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Screening Of Bioactive Components Of Azadirachta Indica A. Juss. And Their Efficacy Against The Red Flour Beetle Tribolium Castaneum (Herbst)

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SCREENING OF BIOACTIVE COMPONENTS OF *AZADIRACHTA INDICA* A. JUSS. AND THEIR EFFICACY AGAINST THE RED FLOUR BEETLE *TRIBOLIUM CASTANEUM* (HERBST)

THESIS
SUBMITTED FOR THE DEGREE
OF
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By
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January, 2016

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DEDICATED

to

My Beloved Parents

DECLARATION

I hereby declare that the entire work submitted as the thesis entitled, “Screening of bioactive components of *Azadirachta indica* A. Juss. And their efficacy against the red flour beetle *Tribolium castaneum* (Herbst)” in the Department of Zoology, University of Rajshahi for the degree of Doctor of Philosophy is the result of my own investigation. The thesis contains no material which has been accepted for the award of any other degree or diploma elsewhere, and to the best of my knowledge, the thesis contains no material previously published or written by another person, except where due reference is made in the text.

January, 2017
Rajshahi University

(Rogena Yeasmin)
Candidate

CERTIFICATE

This is to certify that the thesis entitled “Screening of bioactive components of *Azadirachta indica* A. Juss. And their efficacy against the red flour beetle *Tribolium castaneum* (Herbst)” submitted for the degree of Doctor of Philosophy is bonafide original research work of Rogena Yeasmin, carried out at the Department of Zoology, University of Rajshahi under my supervision.

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Rogena Yeasmin

Abbreviation of the special words used in the text

#U	=	Number of insects used
% kill	=	Insects killed percent
+ve	=	Positive
µg	=	microgram
µl	=	micro liter
$CHCl_3$	=	Chloroform
cm^2	=	centimeter square
Cr %	=	corrected mortality percent.
df	=	degree of freedom.
E. Pr	=	Empirical Probit.
et al.,	=	and others (author)
EtOAc	=	Ethyl Acetate
Ex Pr	=	Expected Probit
F Pro	=	Final Probit
Fig.	=	Figure
fr	=	factor (s)
h	=	hour (s)
HPLC	=	High Perform Liquid Chromatography
i.e.	=	that is
KI	=	Number of insects killed
LC_{50}	=	concentration required to kill 50% of test organisms
LD_{50}	=	dose required to kill 50% of test organisms
LDose	=	Log dose
MeOH	=	Methanol
mg	=	milligram (s)
ml	=	milliliter
mm	=	millimeter
mp	=	melting point
nm	=	nanometer
NMR	=	Nuclear Magnetic Resonance
PDA	=	Potato Dextrose Agar
Rf	=	Retention factor
TLC	=	Thin Layer Chromatography
-ve	=	Negative
Weight	=	Weighting coefficient
Wk Pro	=	Working probit
χ^2	=	Chi-squared

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CONTENTS

ABSTRACT

The chloroform and methanol extracts of the leaves, flower, root bark, root wood, seed, stem bark and stem wood of *Azadirachta indica* were tested against *Tribolium castaneum* adults through residual film assay. According to the intensity of activity observed through mortality of the adult beetles the potentiality of the chloroform extracts could be arranged in a descending order of seed (107.0412µg/cm^2) > leaf (113.3073µg/cm^2) > stem wood (177.580µg/cm^2) > root wood (192.5573µg/cm^2) > stem bark (244.4488 µg/cm^2) > flower (259.3435 µg/cm^2) > root bark (480.3277 µg/cm^2) and for the methanol extracts, seed (222.3965 µg/cm^2) > root wood (418.4427µg/cm^2) > leaf(447.2792µg/cm^2) > stem bark (457.6257µg/cm^2) > stem wood (492.0781µg/cm^2) > flower (752.3578 µg/cm^2) > root bark (1011.733 µg/cm^2) for 96h of exposure. Due to prolongation of exposure no alteration of the results was observed other than proportional increase in the number of mortality. However, mortality was observed just within 24h of exposure, which is very special potentiality in dose-mortality experiments.

All the chloroform and methanol extracts of the flower, leaves, root bark, root wood, seed, stem bark and stem wood of *A. indica* have been applied against the larvae of *T. castaneum* for the detection of their biological activity (including lethality, prolongation of larval instars, causing deformity in body, abnormality in any of the biological parameters). According to the intensity of activity against

the 1st instar larvae of chloroform extract the result could be arranged in the following order: seed> root wood > stem bark > stem wood> flower. In case of methanol extracts, the results were as follows: stem wood > root bark > seed > root wood>flower>stem bark>leaf. For 2nd instars, the results were leaf > flower> root wood> stem bark> root bark > seed > stem wood for chloroform extract after 72 hours respectively. For the methanol extracts the results were as follows: leaf > flower> root wood> root bark > stem wood > seed > stem bark after 72 hours respectively. In case of 3rd instars larvae the results were stem bark > root wood> stem wood > seed > flower> root bark> leaf for chloroform extract after 72 hours respectively. For the methanol extracts the results were stem bark > root wood> seed >stem wood > flower> root bark> leaf for after 72 hours respectively. For the 4th instars larvae against the chloroform extracts the results were as follows: root wood > root bark> seed >flower > leaf > stem wood >stem bark after 72 hours respectively. For the methanol extracts, the results were flower > root bark> seed >root wood > leaf > stem wood >stem bark respectively. The larval mortality showed a possibility of raising toxicity by the magnification of the amount of ingestion of the treated food. Besides mortality of the larvae and abnormality in changing instars, as well as differences in size were also observed. The number of death has been increased just proportional to that of the age of the larvae, which indicates the increase in volume of food intake by the larvae as well.

All the test extracts of leaves, flower, seed, root bark, root wood,

stem bark and stem wood of *A. indica* collected in chloroform and methanol showed repellent activity against adult beetles of *T. castaneum*. The F values have been established were 51. 03662, 253.5068, 43.04438, 83.58911, 75.79346, 75.94017, 64.50964, 50.44838, 25.82928, 61.28114, 45.56164, 34.5519, 35.75216 and 21.5157 for the analysis between doses and 6.778143, 3.007724, 5.447409, 3.835164, 1.400522, 1.856993, 5.669432, 4.258362, 5.590989, 0.876118, 4.630108, 3.285364, 1.990562 and 0.989226 for the analysis between time interval for seed, stem wood, stem bark, root wood, root bark, flower and leaves of Chloroform and Methanol extracts respectively.

Among the tested $CHCl_3$ and MeOH extracts all the rest offered repellency at 0.01% level of significance ($P<0.001$) According to the intensity of repellency the result could be arranged in a descending order: In case of chloroform extract stem bark >root wood> seed >flower> stem wood> leaf> root bark and for the methanol extracts seed> stem wood>stem bark> root bark> root wood> flower> leaf extract.

The cytotoxic effect of the above mentioned extracts was also found promising. The seed extract was found to offer the highest mortality of the nauplii, while the LC_{50} values were 520.1635, 24.50645 and 5.942745ppm for the chloroform extracts; 906.5301, 61.17362 and 18.24789ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively. The LC_{50} values for the stem bark extract were 1042.544, 196.883 and 24.53654ppm for the chloroform extracts; 6030.069, 167.7432 and 34.2457ppm for

the methanol extracts. The LC_{50} values for the stem wood extract were 3711.381, 94.12271 and 45.16339ppm for the chloroform extracts; 1641.063, 92.75699 and 48.30029ppm for the methanol extracts. The LC_{50} values for the flower extract were 933.4176, 67.70986 and 26.04309ppm for the chloroform extracts; 18450.49, 113.4081 and 24.50362ppm for the methanol extracts. For the leaf extract the LC_{50} values were 3476.365, 101.4525 and 51.38413ppm for the chloroform extracts; 9577.411, 455.9743 and 160.1078ppm for the methanol extracts. The LC_{50} values for the root bark extracts were 987.7583, 28.04569 and 23.26771ppm for the chloroform extracts; 1030.155, 57.71285 and 26.29665ppm for the methanol extracts. The LC_{50} values for the root wood extracts were 838.2706, 36.47875 and 8.40184ppm for the chloroform extracts; 5187.234, 82.83993 and 23.38707ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively.

According to the intensity of activity the results of the extracts against the brine shrimp nauplii could be arranged in the following order: seed > root wood >root bark> stem bark> flower>stem wood> leaf for the chloroform extract and seed > root wood > flower> root bark> stem bark> stem wood> leaf for the methanol extracts.

The antibacterial activity of *A. indica* extractives collected in $CHCl_3$ and MeOH were tested against 14 bacteria (6 Gram-positive bacteria) *S. aureus, B. cereus, B. megaterium, B. subtilis, S. lutea, S.-ß -haemolyticus* and (8 Gram-negative bacteria) *S. typhi, S. dysenteriae, S. shiga, S. sonnei, S. boydii, E. coli, P. aeruginosa*

and *Proteus sp.* at concentrations of 50 and 200 µg/disc along with a standard antibiotic, ciprofloxacin 30 µg/disc.

The seed (chloroform) extract *S. aureus*, *B. cereus*, *B. megaterium*, *B. subtilis*, S*. lutea* , *S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii, E. coli* and *Proteus sp*. were responsive with inhibition zones 06,13, 11, 10, 10, 12, 14, 12, 10, 13, 13 and 12 mm for 50 and 200 µg/disc application and for the methanol extract *S. aureus*, *B. cereus*, *B. subtilis*, *S. lutea, S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii*, *E. coli* and *Proteus sp*. were responsive with inhibition zones11, 10, 12, 09, 12, 11, 09, 10,12, 05 and 10 mm respectively for the same doses; while the inhibition zones for the standard Ciprofloxacin 30µg/disc were 30, 28, 28, 30, 28, 30, 30, 29, 29, 28, 28, 28,28 and 28 mm for the above mentioned test agents respectively. For the stem bark (chloroform) extract only *S. aureus*, *B. megaterium*, *B. subtilis*, *S. lutea*, *S.-ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *P. aeruginosa* were responsive with inhibition zones 12, 11, 10, 11, 12, 13, 12, 11, 12, 13mm and 7mm for 200 and 50 µg/disc application and for the methanol extract *S. aureus*, *B. megaterium*, *B. subtilis*, *S. lutea*, *S.-ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *P. aeruginosa* were responsive with inhibition zones 10, 09, 10, 10, 11, 12, 13, 10, 12, 11mm and 08, 10, 09mm respectively for the same doses; while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 28, 30, 28, 30, 29, 29, 29, 28, 28, 29, 29 and 29 mm for the above mentioned test agents respectively. The stem wood extract (chloroform) was responsive to *S. aureus*, *B. cereus* , *B. megaterium*, *B. subtilis*, *S.- ß –*

haemolyticus, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *Proteus* sp. with inhibition zones 10, 08, 10, 12, 10, 12, 10, 09, 10 and 10 mm for 200 µg/disc application and for the methanol extract *S. aureus*, *B. megaterium*, *B. subtilis, S.- ß -haemolyticus*, *S. typhi*, *S. dysenteriae*, *E. coli* and *Proteus* sp. with inhibition zones 08, 09, 11, 07, 10, 09, 07 and 10mm respectively for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30µg/disc were30, 30, 30, 30, 32, 30, 30, 32, 30, 30, 30, 31, 30 and 30 mm for the above mentioned test agents respectively.

The flower extract (chloroform) was responsive to *B. cereus*, *B. subtilis*, *S.- ß -haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii* , *S. shiga*, *E. coli* and *P. aeruginosa* with inhibition zones 12, 11, 13, 13, 12, 12, 11, 14 and 12mm for 200 **µ**g/disc application and the methanol extract was responsive to *B. subtilis*, *S.- ß - haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli and P. aeruginosa* with inhibition zones 10, 12, 11, 10, 11, 10, 13 and 10mm respectively for the same doses; while the inhibition zones for the standard Ciprofloxacin 30**µ**g/disc were 30, 30, 30, 30, 32, 30, 32, 30, 30, 30, 31, 32, 32 and 30 mm for the above mentioned test agents respectively. The leaf (chloroform) extracts were responsive to *S. aureus, B. megaterium, S.-ß-haemolyticus*, *S. typhi*, *S. boydii*, *S. lutea, E. coli* and *P. aeruginosa* with inhibition zones 12, 11, 12, 12, 12, 10, 12 and 10 mm for 200 µg/disc application and the methanol extract was responsive to *S. aureus*, *B. megaterium, S. typhi*, *S. lutea*, *S. boydii*, *E. coli* and *P. aeruginosa* and with inhibition zones 09, 10, 11, 10, 12, 10 and 09 mm for the same doses; while the inhibition zones for the standard

Ciprofloxacin 30 µg/disc were 30, 30, 32, 30, 30, 32, 32, 30, 30, 30, 32, 32, 32 and 30 mm for the above mentioned test agents respectively. In case of the root bark extract (chloroform) *B. cereus*, *B. megaterium*, *B. subtilis, S.- ß -haemolyticus*, *S. typhi*, *S. shiga*, *S. boydii*, *E. coli* and *Proteus* sp. were responsive with inhibition zones 13, 12, 13, 13, 13, 12, 09, 12 and 13mm for 200 µg/disc application, and for the methanol extract *B. cereus*, *B. megaterium*, *B. subtilis, S.- ß –haemolyticus, S. typhi, S. shiga* and *E. coli* were responsive with inhibition zones 11,10,12, 10,11,09 and 11mm for the same doses; while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 32, 30, 30, 32, 30, 30, 30, 32, 30, 32, 30 and 30mm for the above mentioned test agents respectively.

The root wood (chloroform) extracts were responsive to *S. aureus, B. megaterium, B. subtilis, S.-ß-haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. sonnei, S. boydii, E. coli* and *Proteus* sp. with inhibition zones 12, 13, 12, 13, 14, 13, 13, 12, 13 and 10 mm for 200 µg/disc application, and the methanol extract was responsive to *S. aureus*, *B. megaterium, S. lutea*, *S.- ß –haemolyticus, S. shiga*, *S. dysenteriae, S. boydii, E. coli* and *P. aeruginosa* with inhibition zones 10,11, 11, 09, 12, 10, 12, 10, 11 and 09 mm for the same doses; while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 30, 32, 30, 30, 28, 29, 29, 30, 28, 30, 30 and 28mm for the above mentioned test agents respectively.

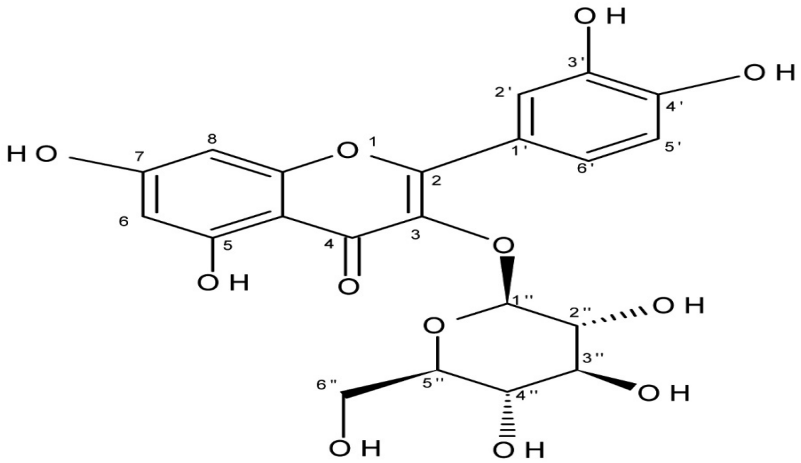
Among all the $CHCl_3$ and MeOH extracts only CHCl3 extracts of the seed and the root wood were subjected to evaluate the minimum inhibition zones. The MIC value of the chloroform extract of the seed was 128µg/ml against *B. cereus,* 64µg/ml against *S.- ß –haemolyticus* and 32µg/ml against *S. dysenteriae*. The MIC values of the chloroform extract of root wood were 128µg/ml against *S. - ß –haemolyticus;* 64µg/ml against *B. megaterium* and 32µg/ml against *S. typhi*.

Antifungal activity of the *A. indica* extractives collected in chloroform and methanol were tested against six pathogenic fungi *F. vasinfectum*, *A. fumigatus*, *A. niger*, *A. flavus, C. albicans* and *P. notatum* at concentrations of 50 and 200µg/disc along with a standard Nystatin (50µg/disc). For the flower extract (chloroform) *A. flavus, A. niger*, *P. notatum* and *C. albicans* were responsive with inhibition zones 13, 12, 12, 11mm and for the methanol extract 10, 10, 14 and 10mm for 200 µg/disc application, while the inhibition zones for the standard nystatin 50 µg/disc were 20, 20, 22, 22, 21 and 20mm for the above mentioned test agents respectively. In case of the leaf extracts (chloroform) *A. niger*, *A. flavus*, *P. notatum* and *C. albicans* were responsive with inhibition zones 14, 13, 12, 12mm and for the MeOH extract 12, 11, 10, 11mm for 200 µg/disc application, while the inhibition zones for the standard nystatin 50µg/disc were 20, 20, 22, 22, 20 and 20mm for the above mentioned test agents respectively. For the root bark extracts (chloroform) *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 14, 13, 10, 11mm

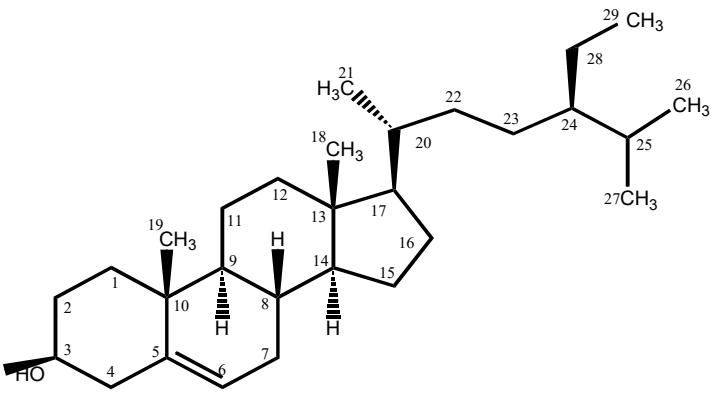
and for the methanol extract 13, 11, 08, 10mm for 200 µg/disc application; while the inhibition zones for the standard nystatin 50 µg/disc were 20, 20, 23, 23, 22 and 20mm for the above mentioned test fungi. In case of the root wood extract ($CHCl_3$) *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones15, 13, 13, 12mm and for the MeOH extract 12, 11, 12, 11mm for 200 µg/disc application; while the inhibition zones for the standard nystatin 50µg/disc were 20, 20, 23, 23, 22 and 20 mm for the above mentioned test agents respectively. In case of the seed extract (chloroform) only *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 16, 14, 12,11mm and for the methanol extract 13, 12, 10, 09 mm for 200 µg/disc; while the inhibition zones for the standard nystatin 50 µg/disc were 23, 24, 24, 24, 23 and 23mm for the above mentioned test fungi respectively.

For the stem bark extract ($CHCl_3$) only *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 13, 12, 13, 11mm and for the MeOH extract 12, 10, 11, 10mm for 200 µg/disc application; while the inhibition zones for the standard nystatin 50 µg/disc were 24, 24, 25, 25, 23 and 23 mm for the above mentioned test agents respectively. For the stem wood extract ($CHCl_3$) only *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 13, 12, 13, 12mm and for the MeOH extract 12, 10, 11, 10mm for 200 µg/disc application; while the inhibition zones for the standard nystatin 50 µg/disc were 23, 24, 25, 25, 23 and 23 mm for the above mentioned test agents respectively.

The leaf, stem bark, root wood and seed of *A. indica* shows alkaloids, carbohydrates, flavanoids, glycosides, phenol, protein, resins, saponnins, tannins and sterols of different solvents. The phytochemical screening was performed with chloroform and methanol extracts of *A. indica*. Through activity guided chromatographic fractionation two bioactive compounds have been isolated from the leaf extracts of the test plant *A. indica.* The $CHCl_3$ extract of the leaf yielded 2 compounds, i) Quercetin3-ß-D-glucoside.ii) ß-sitosterol



Quercetin 3-ß-D-glucoside. A_1



ß-sitosterol

The MIC values of the pure compound A_1 were 32µg/ml against *B. cereus*, 16µg/ml against *S. - ß –haemolyticus* and 64µg/ml against *S. dysenteriae* and of the compound A_2 were 64µg/ml against *B. cereus*, 64µg/ml against *S. - ß –haemolyticus* and 32µg/ml against *S. dysenteriae*.

Chapter-1

General Introduction

1.1 Introduction:

Plants have been a source of herbal remedies throughout the history of mankind. Various medicinal plants have been used for years in daily life to treat diseases all over the world (Nisri *et al.,* 1999). According to recent estimates by the World Health Organization (WHO) more than 3.5 million people in the developing world rely on plants as source of medicine for various ailments. Over 20,000 plants have medical values and many plants are yet to be explored for their potentials. Ethno pharmacologists, Botanists, Microbiologist and natural product chemists are combing the Earth for phytochemical and “leads” which could be developed for treatment of infectious diseases. While 25 to 50% of current pharmaceuticals are derived from plants, they are used as antimicrobials. Plants are rich in a wide variety of secondary metabolites such as tannins, terpenoids, alkaloids and flavonoids, which have been found *in vitro* to have antimicrobial properties (Chattopadhyay *et al.,* 1993*)*.

Plants have formed the basis of traditional medicine systems to maintain human health for thousands of years and have been used as valuable sources of natural products. The medicinal plants are laden with numerous effective pharmacological agents that provide an alternative means of therapy to various infections caused by drug resistant bacteria or dreadful diseases like cancer and other

physiological disorders (Neha *et al*., 2012). Storage of food grains must have commenced much before the cultivation of crops began. The wandering man getting an access to extra food must have collected for consumption when it was not available. He must have kept it at safe place to protect it from insect and other herbivores. From simple storage structures made of mud and other locally available plant materials it has now grown into modern big silos for bulk storage of food grains.

The application of synthetic pesticides to control agricultural pests has been a standard practice. However, with the growing evidence regarding detrimental effects of many of the conventional pesticides safer means of pest management has become very crucial. The loss of food grain during storage due to various insect pests is a very serious problem. More than 2000 species of field and storage pests annually destroy approximately one third of world's food production, valued at more than US $ 100 billion, among which highest losses (43% potential production) occur in developing Asian countries (Ahmed and Grainge, 1986). Annual post-harvest losses resulting from insect damage, microbial deterioration and other factors are estimated to be 10-25% of worldwide production (Matthews,1993). Climate and storage conditions, especially in tropics, are often highly favorable for insect growth and development; control of these insects by chemical insecticides has then serious drawbacks (Sharaby, 1988). The unsystematic use of chemical pesticides have given rise to many obvious serious problems, including genetic resistance by pest species, toxic residues, increasing costs of

application, environmental pollution, hazards from handling, etc. (Champ and Dyte, 1976; Ahmed *et al.*, 1981; Pacheco *et al.,* 1990; Sartori *et al.*, 1990; Rajendran and Narasimhan, 1994; Subramanyam and Hangstrum, 1995; Jembere *et al.,* 1995; Okonkwo and Okoye, 1996).

Stored grain loss in weight and quality of products due to insects is a serious problem world wide. It is estimated that stored grain loss of over 10% occur each year due to insect pests among the stored houses throughout the world and tropics, in particular, *Tribolium castaneum* is a major secondary pest of processed or damaged grains (Danahaye *et al*., 2007).

All insecticides are poisons and the degree of toxicity varies greatly among them. Insecticide's mode of actions involve all the anatomical, physiological and biochemical responses to the chemical poison. Moreover, the fat present in the organism also undergoes reaction with the treated substance. It normally blocks metabolic processes in insects, but this is done in different ways by different compounds. According to their mode of action, the major groups of the most frequently used insecticides are i) nerve poison, ii) muscle poison and iii) physical toxicants (Pedigo, 1996).

Safe and inexpensive insecticides coupled with simple application methods are needed at the rural level (Periera and Wohlgemuth, 1982). In many areas of the world locally available materials are widely used to protect stored products (Golob and Webley, 1980). In the near past the search for naturally occurring antifeedants against pests of field crops and storage has been intensified

(Islam, 1983). A number of investigators isolated, identified and screened chemical compounds from different parts of many botanical families for insect feeding deterrence and growth inhibitor (Jacobson *et al.,* 1975; Bernays and chapman, 1977; Doskotch *et al.,* 1977; Jacobson, 1977; Carpentier *et al.,* 1979; Jurd and Manners, 1980; Menn, 1980).

Renewed interest in botanical pest control agents are motivated by three major objectives: i) to encourage traditional use of simple formulation of locally available plant materials by farmers who can not afford commercial insecticides; ii) to identify sources of new botanical pesticides for commercial extraction; and iii) to elucidate the chemical structure of active principles. Botanical pest control agents extracted on large scale may also be used to replace for supplement the activity of existing synthetic pesticides against refractory pests. Structural elucidation of the active constituents may provide further inside into structure activity relationships. Novel metabolites identified may serve as models for chemical synthesis of new pesticides with more desirable properties. The general pathway of the whole work is given in the Fig. 1 and 2; however the present work could be ahead of purification and pure compound bioassay. To trace the lead components for their further use to enhance the quality of human life it is necessary to go through several steps given in the following manner.

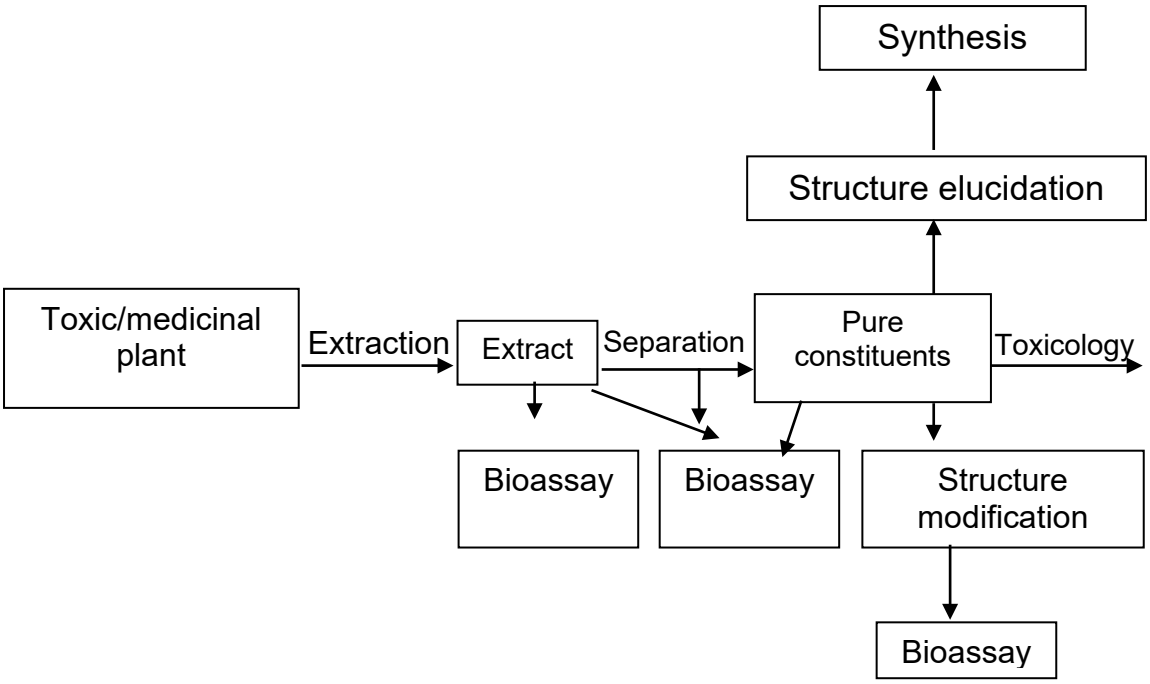


Fig.1: The basic pathway from the plant to the bioactive constituents (After Hostettmann, 1995).

In recent years bioactive principles from natural origin have been subjected to investigation for pest control agents as well as for remedies of diseases without residual or side-effects. Since plants may contain hundreds or even thousands of metabolites there is currently a resurgence of interest in the vegetable kingdom as a possible source of new lead compounds for introduction into therapeutically screening programs (Hostettmann *et al.,* 1995). In fact, plant species is a vast repository of chemical substances that protect plants from attacks by phytophagous insects. Some of these chemicals may repel or kill the insects or deter them from feeding, ovipositor and reproduction. These properties of the plants are of a great value in protecting stored commodities from insect infestation (Munakata, 1977).

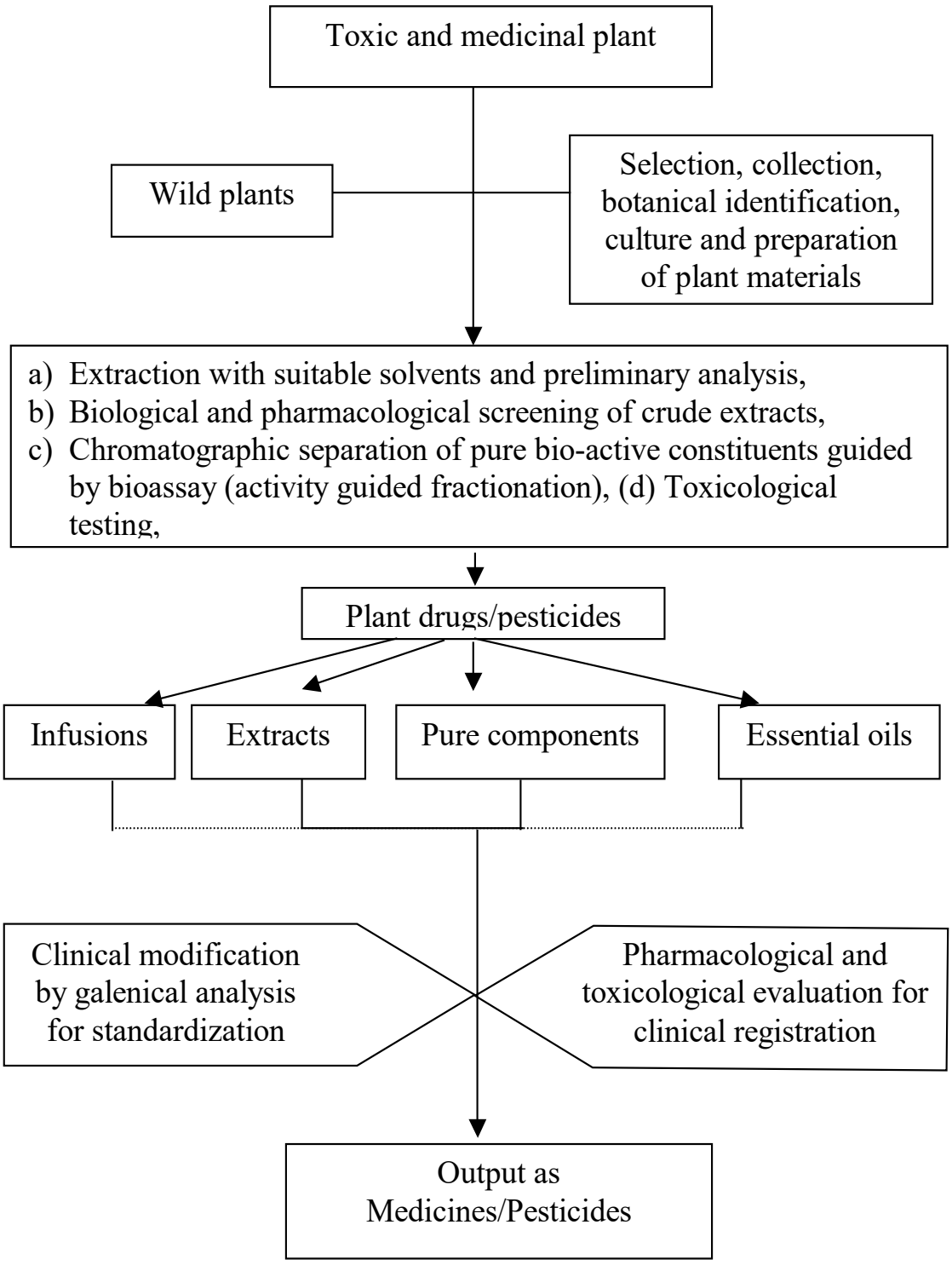


Fig. 2: Phytochemical investigation towards the outputs

(After Hostettmann *et al.,* 1995).

Until now only a small part of the plant kingdom (estimated at 2, 50,000 -5, 00,000 species around the globe) has been investigated

phyto chemically and the fraction subjected to biological and pharmacological screening is even lower. Amongst the most promising of the natural products investigated to date are metabolites. Although only about 10,000 secondary plant metabolites have been chemically identified, the total number of plant chemicals may exceed 4,00,000.They are a vast commucopia of defense chemicals, comprising repellents, feeding deterrents, ovipositor, growth inhibitors, sterility and toxicant etc. (Champagne, *et al.*, 1989).

However, farmers have been using plant extracts in pest control for centuries. This method of pest control provides an ideal source of low cost, safe and effective pesticides. Extracts of plant material rely on the solubility of the active components and it may cause repellent to insects (Sighamony *et al.,* 1984), antifeedant or other type of bioactivities against insects (Jayasinghe and Fujimoto, 1990; Adalla *et al.,* 1993; Facknath and Kawol, 1993; Morallo-Rejesus *et al.,* 1993; Kim *et al.*, 1994; Niber, 1994; Ho *et al.,* 1997; Jannet *et al.,* 2000).

Constituents of many aromatic plants are used for flavoring or medicinal purpose has been found to possess insecticidal properties. Surveys of desert and semi desert plants have revealed a range of sesquiterpenes, benzopyrans, chromenes and prenylated quinines that are cytotoxic (Bell *et al.*, 1990). Some plant families may accumulate a restricted number of anti-insect chemicals, so-called secondary metabolites, whilst other possesses a wide variety of different structural compounds. Secondary metabolites from plants include alkaloids, terpenoids,

phenolics, flavonoids, chromenes and other minor chemicals can affect insects' life in several ways: they may disrupt major metabolic pathways and cause rapid death, act as attractants, deterrents, phagostimulants, antifeedants or an agent to modify oviposition. These compounds may act as fumigants against stored product insects (Huang *et al*., 2000a), contact insecticides (Huang *et al*., 1997, 1999b, 2000b; Huang and Ho, 1998), antifeedant or repellent effects (Chiam *et al*., 1999; Park *et al.*, 2003a, b).

Bangladesh has a great treasure of promising plants. More than 500 plants growing or available in Bangladesh have been reported to possess medicinal properties of some description or other and have been enumerated in the literature of indigenous drugs (Ghani, 1998).

Neem (or Nim) which belongs to the family Meliaceae, originated from South Asia, but grows widely in India, Pakistan and other tropical and sub-tropical parts of the world (Bokhari and Aslam, 1985;Von Maydell, 1986). The tree was introduced in Nigeria from Ghana, and it was first grown from the seeds in Maiduguri, in the then Bornu Province (now Borno State), Nigeria, in 1928 (National Research Council, 1992; Nwoeabia, 1994). In Northern Nigeria, the neem plant is used in traditional circles for the treatment of general body pain after child delivery, pyorrhea, and intestinal worms (Bokhari and Aslam, 1985).

Neem (*Azadirachta indica*) is one of the very few trees known in the Indian subcontinent (Puri, 1999). This tree belonged to

Meliaceae family, and grows rapidly in the tropic and semi-tropic climate. It is also observed that this tree could survive in very dry and arid conditions. (Puri, 1999). The Neem tree is an incredible plant that has been declared the Tree of the 21st century by the United Nations (Puri, 1999). In India, it is variously known as 'Divine Tree', 'Life giving tree', 'Nature's Drugstore', 'Village Pharmacy' and 'Panacea for all diseases'. It is one of the major components in Ayurvedic medicine, which has been practiced in India since many centuries. Extracts from the Neem tree (*A. indicia*) also called 'Dooryard' in Nigeria are most consistently recommended in ancient medical texts for gastrointestinal upsets, diarrhoea and intestinal infections, skin ulcers and malaria (Schmutterer, 1995). All parts of Neem plant such as leaves, bark, flower, fruit, seed and root have advantages in medical treatment and industrial products. Its leaves can be used as drug for diabetes, eczema and reduce fever. Barks of Neem can be used to make toothbrush and the roots has an ability to heal diseases and against insects. (Puri, 1999). The seed of Neem tree has a high concentration of oil. Neem oil is widely used as insecticides, lubricant, drugs for variety of diseases such as diabetes and tuberculosis (Puri, 1999).

India encouraged scientific investigations on neem tree as part of his program to revitalise India tradition and also increase commercial interest on neem (Stix,1992). Neem plant (*Azadirachta indica*) has been of great benefit in human health due to its biochemical, pharmacological, and medicinal properties. Biological

and medicinal properties of *Azadirachta indica* was demonstrated by Sarita Khatkar *et al*., (2014)

According to the history of potentiality *d*ifferent parts of this plant, i.e. flower, leaf, root bark, root wood, seed, stem bark and stem wood have been subjected to biological screening for their possible use in the modern pest control strategy.

1.2. Back ground information on the test plant:

1.2.1. Whereabouts of the titled plant:

Neem (*A. indicia)* is thought to have originated in Assam in northeast India, and Myanmar, where it is common throughout the central dry zone. Later it became naturally distributed throughout much of the Indian subcontinent, particularly in drier areas. Neem name was derived from the Sanskrit *Nimba*, and it was known as the curer of all illness. The neem tree was intimately connected with the everyday life of Indians.

A. indica, Neem (Hindi), *Vembu* (Tamil)) is a tree in the mahogany family Meliaceae. It is one of two species in the genus *Azadirachta*, and is native to India and Pakistan growing in tropical and semi-tropical regions. Its fruits and seeds are the source of neem oil. Other vernacular names include Neem (Nepali, Urdu), Nim (Bengali), Nimm (Punjabi), Arya Veppu (Malayalam), Azad Dirakht (Persian), Nimba (Sanskrit, Oriya), Limdo (Gujarati language) Kadu-Limba (Marathi), Dongoyaro (in some Nigerian languages), Margosa, Neeb (Arabic), Nimtree, Bevu (Kannada),*Kodu nimb* (Konkani), (Kohomba, Sinhala), Tamar (Burmese), (Sdao, Khmer), (Sadaw, Thai), (Hebrew), Paraiso

(Spanish), and Indian Lilac (English). In East Africa it is also known as *Muarubaini* (Swahili), which means *the tree of the 40*, as it is said to treat 40 different diseases, and in Somalia it is known as "Geed Hindi" which means "the Indian tree".

Neem is a fast-growing tree that can reach a height of 15–20 metres (49–66 ft), rarely to 35–40 metres (115–130 ft). It is evergreen, but in severe drought it may shed most or nearly all of its leaves. The branches are wide spread. The fairly dense crown is roundish or oval and may reach the diameter of 15–20 metres (49–66 ft) in old, free-standing specimens.The neem tree has been used for more than 4,500 years in the Indian sub- continent. The Indian physician's charaka (2nd century AD) and susruta (4th century AD), whose books provided the foundation of the Indian system of natural treatment, the Ayurveda, also mention the tree and its medical use. In Ayurveda the neem tree was called the 'Sarva Roga Nivarini' (one that could cure all ailments and ills). At the beginning of this century the neem tree was still highly estimed by Indian emigrants and they took it along to the places where they settled. Thus, the neem tree was introduced in places like Australia, East and sub- Sahelian Africa, South East Asia, and South America. Pioneering work in the possible commercial use of Neem oil and cake had been done by the Indian Institute of Science in Bangalore as early as the 1920s.

Pioneering work in the possible commercial use of Neem oil and cake had been done by the Indian Institute of Science in

Bangalore as early as the 1920s. In the last two decades research on neem has been intensified and many of the trees agricultural and medical properties were rediscovered. Today, Neem plays a major role in the rural industry of India and projects for the commercial use of Neem have been successfully introduced in other countries. The green pinnate leaves of neem have a very bitter taste and garlic- like smell.

1.2.2. Morphological attributes and systemic position of *A. indica*:

Neem is a member of the Meliaceae family. The only congener is *A. excelsa*. Its Sanskrit name, 'arishtha' means 'reliever of sickness' and it is considered as the 'kalpavriksh of kalyuga'. The Persian name of neem is *'Azad- Darakth- E- Hind'* which means 'Free tree of India'. Neem can be regarded as a valuable plant source for the rationalization of its use in traditional medicine and for modern drug development. Neem has a far wider array of uses than any other known herb. The first recorded use of neem is attributed to an ancient Indian culture over 4,500 years ago due to its medicinal properties. Neem provides shade, ornamental look, shelterbelt, fuel wood and construction material, and also helps in degraded land reclamation and soil conservation activities. *Azadirachta indica* is tropical evergreen tree, native to India and Burma; it has been transplanted to Africa, the Middle East, South America and Australia It is especially suited to semi-arid conditions and thrives even in the poorest soil with rainfalls as little as 18 inches (450 mm) per year and temperatures up to 50° C (120° F). It may grow up to 50 feet (15 m) tall and live for 200 years. The

lifespan of the Neem tree is described to be anywhere between 150 to 300 years. Neem is evergreen but can shed most of its leaves under dry conditions. The compound (pinnate) leaves are alternate, 20–40 cm long, with 20–30 dark green, serrated leaflets, each about 3–8 cm long. The terminal leaflet is often absent. Young leaves are reddish to purplish in colour. Neem has a strong root system with single deep tap root and extensive lateral roots. Ripe fruit of neem are about 2 centimeters (cm) long and oval shaped. Inside the fruit there is a light colored seed about 1.5 cm long. The neem tree with many white flowers which smell of honey appear for the first time when the tree is 2 to 3 years old, and the tree bear fruit after 3 to 5 years. Neem trees can grow in areas which have between 400 millimeters (mm) and 1500mm of rain each year. It grows best at an altitude of less than 1,500 meters. Neem trees survive very hot temperatures, up to 44°C and as low as 4°C. Some people reported neem trees surviving light frost.

The neem tree (*A. indica)*, is a tropical evergreen with a wide adaptability, native to India and Burma, it has been transplanted to Africa, the Middle East, South America and Australia. Neem can grow into a big tree to a height of about 20 to 35 m. Its canopy of leaves makes it a useful shade tree. It is planted along roads and avenues in the towns and villages of India. Its blossoms are small, white flowers with a very sweet, jasmine-like scent. Its edible fruit is about 3/4 of an inch (2 cm) long, with white kernels. A neem tree generally begins bearing fruits at three to five years of age, and can produce up to 50 kg of fruit annually when mature. The pinnate leaves have a very bitter taste and a garlic-like smell. The

trunk is relatively short, straight and may reach a diameter of 1.2 m (about 4 feet). It is classified as a bush.

Plate 1: *Azadirachta indica*

Leaves: The opposite pinnate leaves are 20-40 cm (8 to 16 inch) long, with 20 to 31 medium to dark green leaflets about 3-8 cm (1 to 3 inch) long. The petioles are short. Very young leaves are reddish to purplish in colour. The shape of mature leaflets is more or less asymmetric and their margins are dentate with the exception of the base of their basis copal half, which is normally very strongly reduced and cuneate or wedge-shaped. (Ganguli, 2002).

Flowers: The (white and fragrant) flowers are arranged auxiliary, normally in more-or-less drooping panicles which are up to 25 cm (10 in.) long. The inflorescences, which branch up to the third

degree, bear from 150 to 250 flowers. An individual flower is 5-6 mm long and 8-11 mm wide. Protandrous, bisexual flowers and male flowers exist on the same individual. Flowers are used to make a curry called ugadi pachadi.

Fruit: The fruit is a smooth (glabrous) olive-like drupe which varies in shape from elongate oval to nearly roundish. The fruit skin (exocarp) is thin and the bitter-sweet pulp (mesocarp) is yellowish-white and very fibrous. The mesocarp is 0.3-0.5 cm thick. The white, hard inner shell (endocarp) of the fruit encloses one, rarely two or three, elongated seeds (kernels) having a brown seed coat (Ganguli, 2002). Seeds usually fall to the ground and might stay there or be carried away with rain water. Occasionally they are dispersed away from the parent tree by birds which give them a greater chance of growing into a healthy new plant. Neem oil is obtained from the seeds.

The neem tree is noted for its drought resistance. Normally it thrives in areas with sub-arid to sub-humid conditions, with annual rainfall 400–1,200 mm (16–47 in). It can grow in regions with an annual rainfall below 400 mm, but in such cases it depends largely on ground water levels. Neem can grow in many different types of soil, but it thrives best on well drained deep and sandy soils. It is a typical tropical to subtropical tree and exists at annual mean temperatures between 21–32 °C (70–90 °F). It can tolerate high to very high temperatures and does not tolerate temperature below 4 °C (39 °F). Neem is a life-giving tree, especially for the dry coastal, southern districts of India and Pakistan. It is one of the very few shade-giving trees that thrive in the drought-prone areas. The

trees are not at all delicate about the water quality and thrive on the merest trickle of water, whatever the quality. In India it is very common to see neem trees used for shade lining the streets or in most people's back yards. In very dry areas the trees are planted in large tracts of land.

Phylogenetic position of *A. indica*

Kingdom: Plantae (plants)

Subkingdom: Tracheobionta(Vascular plants)

Superdivision: Spermatophyta (Seed plants)

Division: Magnoliophyta (Flowering plants)

Class: Magnoliopsida (Dicotyledons)

Subclass: Rosidae

Order: Sapindales

Family: Meliaceae (Mahogany family)

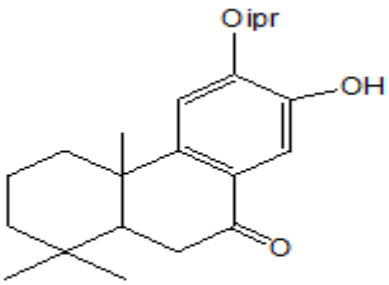
Genus: *Azadirachta*

Species: *Azadirachta indica*

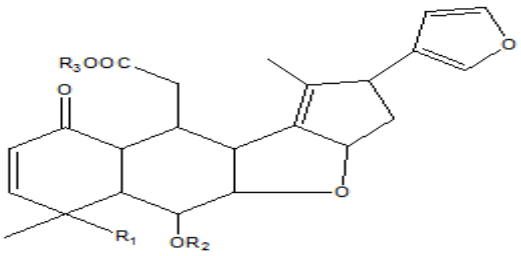
1.2.3. Chemical constituent and properties of neem:

Chemical investigations of neem were undertaken by Indian pharmaceutical chemists in 1919, whereby they isolated acidic principle in neem oil, which they named as 'margosic acid".

However, real chemical research originated in 1942 with isolation of three active constituents, viz, nimbin, nimbidin and nimbinene. In 1963 an Indian scientist extensively examined the chemistry of the active principles of neem. Following the discovery of neem kernel as a locust feeding deterrent, its chemistry has grown considerably. Several compounds have been isolated and characterized. The main feature is that most of them are chemically similar and biogenetically derivable from a tetracyclicterpenes. These are also called liminoids (azadirachtin, meliantrol, salanin etc.) bitter principles and occur in other botanical species as well (Rutaceae and Simaroubaceae). The unraveling of high complex structural features and biogenetic interrelationship represent classic piece of work on natural product chemistry. From the practical side these compounds also exhibit a wide variety of biological activity, for example, pesticides, antifeedants, and cytotoxic properties. Levaesmaily yield quercetin (flavonoid) and nimbosterol (ß- sitosterol) as well as number of liminoids (nimbin and its derivatives). Quercetin (a polyphenolic flavonoid) is known to have antibacterial and antifungal properties. The neem constituent belonging to chemically diverse classes have been divided into two major sections viz. I) isoprenoids, II) non-isoprinoids. The later category comprises glycerides, polysaccharides, sulphurones compounds, flavonoids and their glycosides, amino acids, aliphatic compounds etc**.** Aktar *et al*., (2000).Structure of some constitituents are given below (Fig.3).

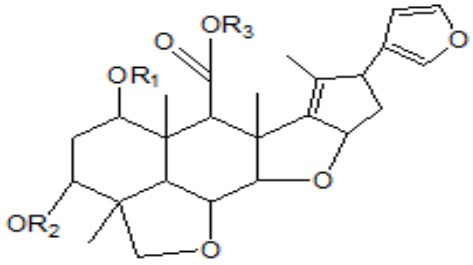


Margosone



Azadirachtin A

Where R1=COOMeR2=Ac R3=Me



Salanin

Where R1=Tg (Tiglica C = 4), R2=Ac, R3 =Me

The structure of some of these bioactive compounds has been presented in Figure 1.Nimbidin; a major crude bitter principle extracted from the oil of seed kernels of *A. indica* demonstrated several biological activities. From this crude principle some tetranortriterpenes, including nimbin, nimbinin, nimbidinin, nimbolide and nimbidic acid have been isolated (Siddiqui, 1942;

Mitra *et al*., 1971).Nimbidin and sodium nimbidate possess significant dose dependent anti-inflammatory activity against carrageenininduced acute paw oedema in rats and formalin-induced arthritis (Bhargava *et al*., 1970; Pillai and Santhakumari 1981).Oral administration of nimbidin demonstrated significant hypoglycemic effect in fasting rabbits (Pillai and Santhakumari 1981). A significant antiulcer effect was observed with nimbidin in preventing acetylsalicylic acid, indomethacin, stress or serotonin-induced gastric lesions as well as histamine or cysteamine-induced duodenal ulcers (Pillai and Santhakumari 1984; Pillai *et al*., 1978). Nimbidin can also suppress basal as well as histamine and carbachol-stimulated gastric acid output and may act as an antihistamine by blocking H_2 receptors, thereby helping as an antiulcer agent (Pillai and Santhakumari 1985). The spermicidal activity of nimbidin and nimbin was reported in rats and human (Sharma and Saksena 1959). Nimbidin also demonstrated antifungal activity by inhibiting the growth of *Tinea* rubrum (Murthy and Sirsi 1958). *In vitro*, it can completely inhibit the growth of *Mycobacterium tuberculosis* and was also found to be bactericidal (Murthy and Sirsi 1958). Diuretic activity was also reported for sodium nimbidinate in dogs (Bhide *et al*., 1958). Nimbolide has been shown to exert antimalarial activity by inhibiting the growth of *Plasmodium falciparum* (Rochanakij *et al*., 1985; Khalid *et al*., 1989). Nimbolide also shows antibacterial activity against *S. aureus* and *S*. coagulase (Rojanapo *et al*., 1958). Gedunin isolated from neem seed oil has been reported to possess both antifungal (Rao *et al*., 1977) and antimalarial (Khalid *et al*., 1989) activities.

Azadirachtin highly oxygenated C-secomeliacins isolated from neem seed and having strong antifeedant activity (Kraus, 1995; Govindachari, 1992; Butterworth and Morgan, 1968) has been demonstrated to have antimalarial property as well. It is inhibitory to the development of malarial parasites (Jones *et al*., 1994). Mahmoodin a deoxygedunin isolated from seed oil has been shown to possess moderate antibacterial action against some strains of human pathogenic bacteria (Devakumar and SukhDev, 1996). Condensed tannins from the bark contain gallic acid, (+) gallocatechin, (–) epicatechin, (+) catechin and epigallocatechin, of which gallic acid (–) epicatechin and catechin are primarily responsible for inhibiting the generation of chemiluminescence's by activated human polymorph nuclear neutrophil (PMN) (Vander Nat *et al*., 1991), indicating that these compounds inhibit oxidative burst of PMN during inflammation. Three tricyclic diterpenoids, margolone, margolonone and isomargolonone isolated from neem stem bark are active against *Klebsiella*, *Staphylococcus* and *Serratia* species (Ara *et al*., 1989). Sulphur-containing compounds such as cyclic trisulphide and tetrasulphide isolated from the steam distillate of fresh, matured neem leaves have antifungal activity against *Trichophyton* mentagrophytes (Pant *et al*., 1986). Several polysaccharides from neem exhibit various biological effects. A polysaccharide extracted from bark inhibits carrageen in-induced inflammation in mouse (Kakai Tokkyo Koho, 1984).Two water-soluble polysaccharides GIa and GIb isolated from the bark of *Melia azadirachta*, demonstrated strong antitumour effect with complete regression of the tumors, when administered in mice at a

daily dose of 50 mg/kg for four days from 24 h after subcutaneous inoculation of Sarcoma-180 cells (Fujiwara, *et al*., 1982). Two more polysaccharides, GIIa and GIIIa isolated from *M. azadirachta* bark also showed significant anti-inflammatory effect on carrageen in-induced oedema in mice (Fujiwara *et al*., 1984). Two polymers isolated from an aqueous extract of neem bark possess anticomplement activity, amongst which the compound NB-II, a peptidoglycan of lower molecular weight was found to be more potent (Vander Nat *et al*., 1987, 1989). Some active ingredients (phytosterol fraction) isolated from the lipid part of neem fruits, exhibit antiulcer activity in stress induced gastric.

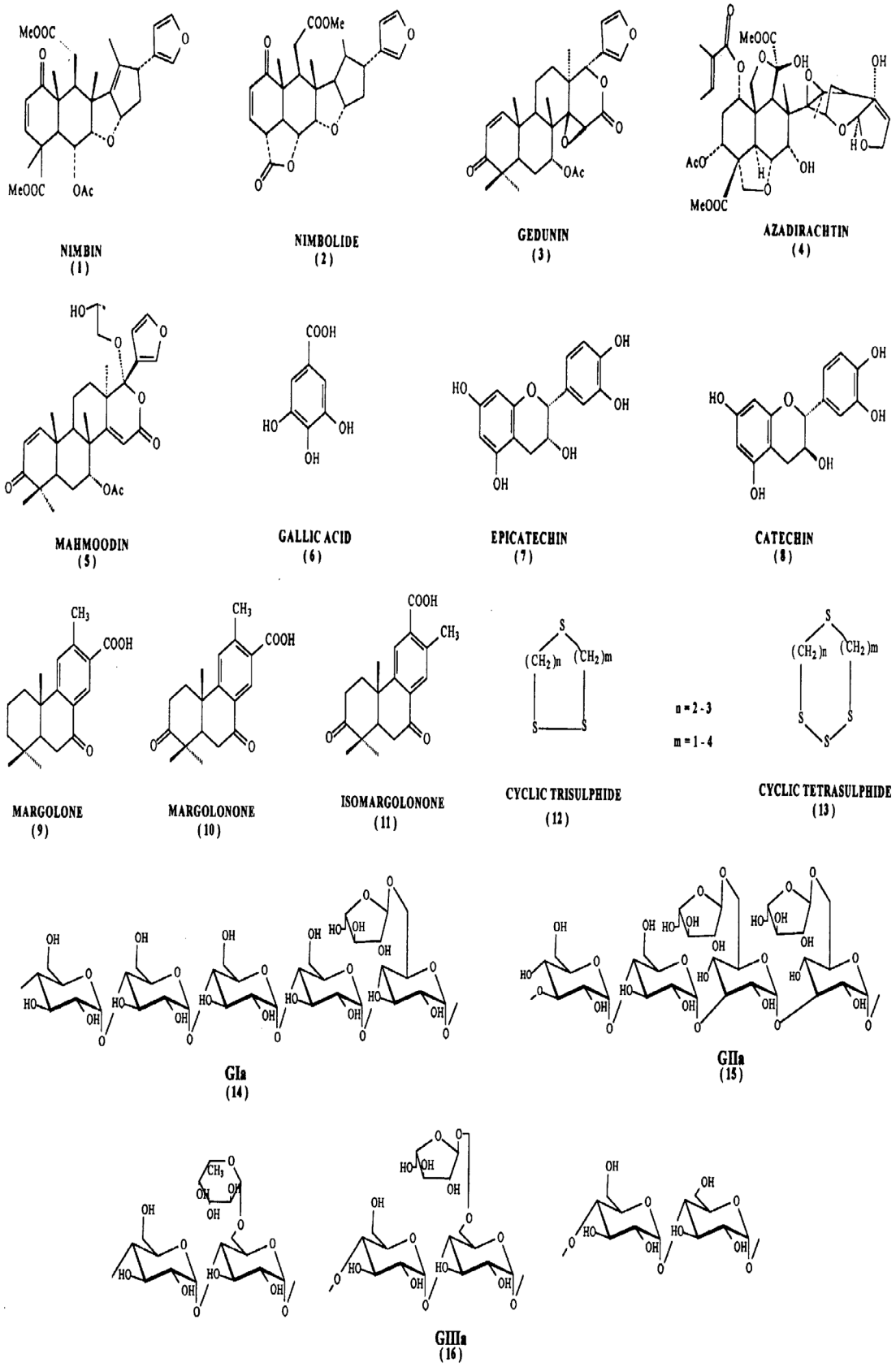


Figure 3. Structure of neem compounds

More than 135 compounds have been isolated from different parts of neem. The compounds have been divided into two major classes: isoprenoids [like diterpenoids and triterpenoids containing protomeliacins, limonoids, azadirone and its derivatives, gedunin and its derivatives, vilasini type of compounds and C-secomeliacins such as nimbin, salanin and azadirachtin] and non-isoprenoids, which are proteins/amino acids and carbohydrates [polysaccharides], sulphurous compounds, polyphenolics such as flavonoids and their glycosides, dihydrochalcone, coumarin and tannins, aliphatic compounds, etc. 'Phyto' is the Greek word for plant. There are many families of phytochemicals and they help the human body in a variety of ways. Phytochemicals may protect human from a host of diseases. Phytochemicals are non-nutritive plant chemicals that have protective or disease preventive properties. Plant produces these chemicals to protect itself but recent research demonstrates that many phytochemicals can protect humans against diseases. There are many phytochemicals in fruits and herbs and each works differently.

The recorded use of plants in the treatment of aliments dates back to antiquity. Plants are recognized for their ability to produce a wealth of secondary metabolites and mankind has used many species for centuries to treat a variety of diseases. Many of these natural products have been shown to present interesting biological and pharmacological activities and are used as chemotherapeutic agents or serve as the starting point in the development of modern medicines (Abubakar *et al.*, 2010). It is estimated that today, plant materials are present in, or have provided the models for 50% of

Western drugs. The term qualitative phytochemical analysis refers to the procedures involved in establishing and proving the identity of the phytochemical constituents present in the crude plant extract. The pharmacological actions of crude drugs are determined by the nature of their constituents.

The late Pakistani scientist Salimuzzaman Siddiqui was the first scientist to bring the plant to the attention of phyto pharmacologists. In 1942 while working at the Scientific and Industrial Research Laboratory at Delhi University, India, he extracted three bitter compounds from neem oil, which he named nimbin, nimbinin, and nimbidin respectively (Ganguli, 2002). The seeds contain a complex secondary metabolite Azadirachtin. Several chemical compounds have been identified and scientists feel that there are many more compounds yet to be identified in neem. Other than sodium, potassium, salts, it contains chlorophyll, calcium, phosphorus, iron, thiamine, riboflasium, nicocin, vitamin C, carotene, and oxalic acid. The chemicals classified are:

- Nimbin: anti-inflammatory, anti-pyretic, anti-histamine, anti-fungal
- Nimbidin: anti-bacterial, anti-ulcer, analgesic, anti-arrhythmic, anti-fungal
- Ninbidol: anti-tubercular, anti-protozoan, anti-pyretic
- Gedunin: vasodilator, anti-malarial, anti-fungal
- Sodium nimbinate: diuretic, spermicide, anti-arthritic
- Quercetin: anti-protozoal
- Salannin: insect repellent

- Azadirachtin: insect repellent, anti-feedant, anti-hormonal

Other chemicals that form its therapeutic value are:

- Limonoids
- Terpenoids and steroids
- Tetranortarpenoids
- Fatty acid derivatives like margosinone and margosinolone
- Coumarins like scopoletin, dihydrosocoumarins
- Hydrocarbons like docosane, pentacosane, hetacosane, octacosane etc.
- Sulphur compounds
- Phenolics
- Flavonoglycosides
- Tannins

The highest concentrations of the active ingredients are found in the seed and oil, however the active ingredients are also found in lesser amounts in the bark and the leaves

1.2.4. Social utilities and folk medicinal use of the titled plant:

Medicinal plants are rich source of novel drugs that forms the ingredients in traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates, bioactive principles and lead compounds in synthetic drugs**.** WHO pointed out that more than 80% of world's population depends on plants to meet their primary health care needs**.** However, overexploitation of the selected medicinal plant species lead to the reduction in number of plants in the wild and inclusion of their name in the red data book 2. Neem (*Azadirachta*

indica) commonly called 'Indian Lilac' or 'Margosa', belongs to the family Meliaceae, subfamily Meloideae and tribe Melieae. *Azadirachta indica* has been used medicinally throughout history by many different cultures. Many compounds have been found in the exudates of the, *Azadirachta indica* plant that have been used medically by humans.

For thousands of years the beneficial properties of Neem (*Azadirachta indica*) have been recognized in the Indian tradition. Each part of the neem tree has some medicinal properties (Biswas *et al*., 2002). Use of neem dates back to pre-historic period as its use is mentioned in Sanskrit language which is one of the oldest languages of the world. Keeping of neem leaves between folds of cloths, leather goods and mixing them with grain destined for storage, generation of smoke to drive away mosquitoes are well known traditional practices. These are still being used with satisfaction in India, Pakistan and SriLanka. Of the different parts of the neem tree it is the neem leaves which have been used extensively in India and some other neighboring countries. Their use, however, differ in different regions**.** For example *bukhari* a common storage structure in Uttar Pradesh (India) is made of plant materials.

Various parts of the neem tree have been used as traditional ayurvedic medicine in India from time immemorial (Varma, 1976). The medicinal utilities have been described, especially for leaf, fruit and bark (Thakur *et al*., 1981). Neem oil and the bark and leaf extracts have been therapeutically used as folk medicine to control leprosy, intestinal helminthiasis, respiratory disorders, constipation

and also as a general health promoter (Kirtikar and Basu, 1935). Its use for the treatment of rheumatism, chronic syphilitic sores and indolent ulcer has also been evident (Kirtikar and Basu, 1975). Neem oil finds use to control various skin infections (Chopra *et al*., 1956). Bark, leaf, root, flower and fruit together cure blood morbidity, biliary afflictions, itching, skin ulcers, burning sensations and pthysis (Mitra, 1963). However, apart from these uses, there are several reports on the biological activities and pharmacological actions of neem based on modern scientific investigations.

Anti-inflammatory, antipyretic and analgesic activities:

The chloroform extract of stem bark is effective against carrageen in-induced paw oedema in rat and mouse ear inflammation (Tidjani *et al*., 1989). Inflammatory stomatitis in children is cured by the bark extract (Lorenz, 1976). Antipyretic activity has been reported in neem oil (Murthy and Sirsi, 1958). A methanol extract of the leaves exerts antipyretic effect in male rabbits (Okpanyi and Ezeukwv,1981). The plant also possesses analgesic activity mediated through opioid receptors in laboratory animals (Vohra and Dandiya,1992). Anti-inflammatory and antipyretic activities in various extracts have been reviewed (Jacobson, 1986).

Immunostimulant activity:

The aqueous extract of neem bark possesses anti complement activity, acting both on the alternative as well as the classical pathway of complement activation in human serum (Vander Nat *et al*., 1987). Recently, an aqueous extract of stem bark has been shown to enhance the immune response of Balb-c mice to sheep

red blood cells *in* vivo (Njiro and Kafi-Tsekpo, 1999). The aqueous extract of leaf also possesses potent immunostimulant activity as evidenced by both humeral and cell-mediated responses (Sen *et al*., 1992; Ray *et al*., 1996).

Hypoglycemic activity:

Aqueous extract of neem leaves significantly decreases blood sugar level and prevents adrenaline as well as glucose-induced hyperglycemia (Murty *et al*., 1978). The aqueous leaf extract when orally fed, also produces hypoglycemia in normal rats and decreased blood glucose levels in experimentally-induced diabetes in rats (El-Hawary and Kholief, 1990). Aqueous leaf extract also reduces hyperglycemia in streptozotocin diabetes and the effect is possibly due to presence of a flavonoid, quercetin (Chakraborty *et al*., 1989). Recently, hypoglycemic effect was observed with leaf extract and seed oil, in normal as well as alloxan-induced diabetic rabbits (Khosla *et al*., 2000).

Antiulcer effect:

Neem leaf aqueous extract producesantiulcer effect in rats exposed to restraint–cold stress or ethanol orally by preventing mucus depletion and mast cell degranulation (Garg *et al*., 1993). An aqueous extract of neem bark has been shown from our laboratory to possess highly potent antacid secretary and antiulcer activity and the bioactive compound has been attributed to a glycoside (Bandyopadhyay *et al*., 1998).

Antimalarial activity:

Neem seed and leaf extracts are effective against malarial parasites (.Khalid *et al*., 1989; Butterworth and Morgan 1968). Components of the alcoholic extracts of leaves and seeds are effective against both chloroquin-resistant and sensitive strains of malarial parasite (Badani *et al*., 1987). Recently, neem seed extract and its purified fractions have been shown to inhibit growth and development of asexual and sexual stages of drug sensitive and resistant strains of the human malarial parasite *P. falciparum* (Dhar *et al*., 1998).

Antioxidant activity:

The antioxidant activity of neem seed extract has been demonstrated *in vivo* during horse grain germination, which is associated with low levels of lipooxygenase activity and lipid peroxides (Rao *et al*., 1998). An antioxidant principle has also been isolated, which is a potent inhibitor of plant lipooxygenases.

Safety evaluation of neem compounds and marketed formulations:

Nimbidin produces sub-acute toxicity in adult rats after daily administration of 25, 50 or 100 mg/kg for six weeks (Kanungo, 1996). A significant hypoglycaemic effect was observed by feeding nimbidin to fasting rabbits (Pillai and Santhakumari, 1981). Nimbidin also has spermicidal activity (Sharma and Saksena, 1959). Nimbolide, a major chemical component of neem seed oil, and nimbic acid were found to be toxic to mice when given intravenously or intraperitoneally (Jacobson, 1995; Glinsukon *et*

al., 1986). Nimbolide and nimbic acid at a lethal dose cause death in most animals by dysfunction of kidney, small intestine and liver as well as by marked and sudden drop of arterial blood pressure.

The structure having been made is plastered with mud containing crushed neem leaves. The plastering is done on inner wall and the top. In Punjab (India and Pakistan) neem leaves extract is sprinkled on wheat straw packed at botton of *palli* 2-3 days prior to storing grain (Jilani and Saxena, 1988). A survey conducted in 8 Indian states and 2 Pakistan showed that farmers used 'handful' to 5-10 kg. of air dried neem leaves per 100 kg of grain (paddy/ rice, wheat, corn and soghum) either alone or in combination with leaves of *Vitex negundo* or *Pongamia pinnata* for protecting stored grain (Ahmed and Grainge, 1986; Ahmed *et al*, 1988; Hegde *et al*., 1988).Users found it effective in preventing entry of beetle and weevils if neem leaves are mixed with freshly harvested grains immediately on storage. The surveyed farmers were satisfied with protection obtained for their 3-6 months storage period. Also the treatment did not adversely affect the grain's cooking quality as most of the residue of leaves gets removed during winnowing. There is also practice in India and more commonly in Pakistan to prepare paste of leaves and then paint with small straw broom inside *padolla* or *dehri* (earthern structures 1-2 metre tall used for storing grain. It is closed with earthern lid. Grain is taken out through a hole at the base. The grain may remain pest free for a year). Farmers in Sri Lanka burn neem leaves to generate smoke to fumigate stored paddy and pulses (Ranasinghe, 1984)

Traditionally Neem was used in Ayurveda for a number of conditions. It is one of the main ingredients in every blood purification formula used in Ayurveda and it appears in most diabetic formulas as well. It is also used for arthritis, rheumatism, the removal of external and internal parasites, including malaria and fevers and as an insect repellent.

Neem leaf extract has been prescribed for oral use for the treatment of malaria by Indian Ayurvedic practitioners from time immemorial. Recently, a clinical trial has been carried out to see the efficacy of neem extract to control hyperlipidemia in a group of malarial patients severely infected with *P. falciparum*. The lipid level, especially cholesterol, was found to be lower during therapy when compared to non-malaria patients. Reports are available regarding the use of neem to treat patients suffering from various forms of cancer. One patient with parotid tumor and another with epidermoid carcinoma have responded successfully when treated with neem seed oi1 Neem leaf aqueous extract effectively suppresses oral squamous cell carcinoma induced by 7,12-dimethylbenz[a] anthracene (DMBA), as revealed by reduced incidence of neoplasm. Neem may exert its chemo preventive effect in the oral mucosa by modulation of glutathione and its metabolizing enzymes. NIM- 76, a refined product from neem oil, was studied in 10 human volunteers, where intra-vaginal application before sexual intercourse could prevent pregnancy with no adverse effect on vagina, cervix and uterus. Different studies have shown that neem has effects against certain diseases like eczema, acne, and some skin problems like dry Skin, wrinkles,

dandruff, itchy Scalp, skin ulcers and warts are other conditions that can be effectively resolved by the use of soaps, lotions, and creams, containing neem leaf extracts and oil.

Neem oil:

Neem oil is a vegetable oil pressed from the fruits and seeds of neem plant (*Azadirachta indica*). Neem oil is generally light to dark brown, bitter and has a rather strong odour that is said to combine the odours of peanut and garlic. It comprises mainly triglycerides and large amounts of triterpenoid compounds, which are responsible for the bitter taste.Neem oil also contains steroids (campesterol, beta-sitosterol, stigma sterol) and a plethora of triterpenoids of which azadirachtin is the most well-known and studied. The azadirachtin content of neem oil varies from 300ppm to over 2500ppm depending on the extraction technology and quality of the neem seeds crushed (Puri,1999).

Neem gum:

Neem Gum is a clear, bright and brown-coloured gum obtained from the trunk of neem. This is as a result of certain metabolic mechanism of plants and trees. The gum is a multipurpose by product either water soluble or absorbs water to form a viscous solution.

Neem honey:

In India, neem trees are a major source of honey bee forage. Honey obtained from the Neem tree has more medicinal properties. Neem honey is composed primarily of water, fructose and glucose (22.88%), sucrose (7.46%), ash (0.06%), free acid (20.8 mg/kg). The honey is light amber in colour and its viscosity is low. The taste is good although slightly bitter. Neem honey improves eye sight and is harmless for diabetic patients. It is also used to treat eye disorder by applying as netranjan (eye-liner). It is very beneficial in care of burning sensation of the body. Since Neem is believed to be a great blood purifier and good for the eyes, Neem honey is highly valued.

Neem as a vegetable:

The tender shoots and flowers of the neem tree are eaten as a vegetable in India. Neem flowers are very popular for their use in Ugadi Pachhadi (soup-like pickle), which is made on Ugadi day in the South Indian States of Andhra Pradesh, Tamilnadu and Karnataka. A soup like dish called Veppampoo Rasam (Tamil) translated as ‘neem flower rasam’ made of the flower of neem is prepared in Tamil Nadu. Neem is also used in parts of mainland Southeast Asia, particularly in Cambodia, Laos (where it is called kadao), Thailand (where it is known as sadao or sdao), Myanmar (where it is known as tamar) and Vietnam (where it is known as sau dau and is used to cook the salad: goi sau dau). Even lightly cooked, the flavour is quite bitter and thus the food is not enjoyed by all inhabitants of these nations, though it is believed to be good

for one's health. Neem Gum is a rich source of protein. In Myanmar, young neem leaves and flower buds are boiled with tamarind fruit to soften its bitterness and eaten as a vegetable. Pickled neem leaves are also eaten with tomato and fish paste sauce in Myanmar.

Plate 2: Leaves and fruits with *A. indica*

1.3. Aim of this work:

Quite a good number of plants have been identified and utilized for insecticidal and medicinal purpose till to date. But it is true that a large number of plants have still been untouched or less investigated from which significant results can be obtained to control the pests of crops and human disease problems.

A. indica is one of such plants that has been studied a lot phytochemically and only a few studies have been done with its medicinal properties, but in details a very few works have been done till to date on its use in the control of crop pests. Keeping this in mind the present investigations were undertaken.

Objectives of the present work

i. Screening of *A. indica* plant extractives for insecticidal potential against *T. castaneum* through dose-mortality bioassay establishing the LD_{50} values;

ii. Screening of *A. indica* plant extractives for phytochemical effect using various process.

iii. Further screening of the same through larvicidal effect, as well as special efficacy against their growth and development offering larval mortality, changes of duration at each instar, arresting their growth and metamorphosis thereby giving deformity during adult emergence;

iv. More screening of the test materials using adult beetles by repellency test to see whether or not the extracts contain any potential to repel the stored grain pests;

v. Screening the test materials through cytotoxicity test against *A. salina* nauplii, which is a standard technique to detect the bioactive potentials of the test plant applied in the aquatic medium;

vi. An overall reevaluation of the chloroform and methanol extracts of the leaves, flowers, root wood, root bark, stem bark, stem wood and seeds were done through anti microbial assay, to justify their efficacy through inhibition of their growth;

vii. To isolate, purify and characterize the bioactive compound(s) from the promising extracts and to evaluate the efficacy of the purified compounds against the selected test agents using any suitable biological assay;

viii. To comment on the future perspectives of the test plant for the control of crop pests based on the achieved results.

Chapter-2

Review of literature

The research was carried out to observe the effect of plant extracts of different parts of *Azadirachta indica* against *Tribolium castaneum* and other insects with micro organisms. Therefore, literatures some way linking to the subject of interest from home and abroad are reviewed and outlined below gradually. Literature on the concern topic was searched for making a progress in the present work. Many plant oils/extracts have used against many vectors. Also many botanical extracts are found to be active as pesticide/ insecticide. Toxicological effects of neem (*Azadirachta indica*) Kanair (*Nerium oleander*) and Spinosad (Tracer 240 SC) on the red flour beetle (*Tribolium castaneum*) (Herbst) was studied by Asifa Hameed *et al*., 2012. Ethanolic extracts of Kanair was found least effective against *Tribolium* sp. In comparison with neem extracts and spinosad. Maximum mean mortality (38.13%) at 48h exposure time with maximum dose (2.5%) and minimum (15.63%) mortality at 24h with 0.5% dose. Neem showed maximum mortality (45.63%) was found at exposure time maximum dose of 2.5% and minimum Control (16.88%) was at 24h with 0.5% concentration. Maximum mortality (53.75%) was found at 48h with 2.5% concentration and minimum (16.87%) at 24h interval with 0.5% concentration.

Pruthi (1937) first proved scientifically the insecticidal effect of neem. “Azadirachtin a microcrystalline compound isolated from neem kernel extract is a promising larvicide’s against *Culex*

pipiens. Naturally occurring bio pesticides could be an alternative to chemical pesticides" (Abdelouaheb *et al.,* 2009). It has been reported that it possesses many substances which interfere with insect molting, food uptake, reproduction and provides a nontoxic insect controlling agent for use in agriculture. These cause growth inhibition, abnormal development, elongation of larval period and no pupation (Ascher et al., 1984; Isman, 1993; Ladd, 1984; Mari, 1989; Naqvi *et al*., 1991, 1994).

Aqueous neem kernel extracts were used for warding off insect attack on crops. Neem leaf juice is used for expelling worm and curing jaundice and skin diseases. Oil from nuts and leaves is a stimulant insecticide and antiseptic. It inhibits feeding in a variety of insects and also inhibits ecdysis at much lower concentrations (Mari & Watanabe, 1989). This prevents the insect larvae from developing into mature insects which could further multiply and produce new generations. It blocks receptor of ecdysteriods which are needed for larval development (Govindachari, 1992). Azadirachtin also increased residence time in the feeding and nonfeeding immature stages, larva treated with 1.6µg of azadirachtin for example, had significant longer larval periods than did untreated larvae; length of prepupal and pupal stages was extended (Ladd, 1984). Lin and Liu (2006) studied properties and efficacy of pesticides from neem tree and found them effective antifeedants for pest control. Azadirachtin were growth inhibitors. They interfere with neuroendocrine regulation of juvenile and molting hormone titers (Rembold, 1988). Toxicity and abnormalities caused by neem fractions, RBU-9, RB-b and

Margosan-OTM were determined against fourth instars larvae of *Aedes aegypti,* partially emerged adults were found with crumpled and entangled legs in puparium (Naqvi *et al.,* 1994).Recent studies encouraged the investigation of insecticidal properties of plant-derived extracts and concluded that they are environmentally safe, degradable, and target specific (Senthil *et al.,* 2006). Neem is a natural insecticide and is nonhazardous to man and other mammals (Oudegans, 1991). Neem components show multiple effects against different insects such as mosquitoes, flies, triatomine bugs, cock-roaches, fleas, lice and ticks (Mulla and Su, 1999; Ruskin, 1992). Neem leaf and seed extracts also showed efficacy against stored grain pests (Sharif *et al.,* 2007). Many biologically active compounds have been isolated from neem, *Azadirachta indica* A. Juss, including triterpenoids, azadirachtin (Butterworth and Morgan, 1971). Azadirachtin is a mixture of seven isomeric compounds as Azadirachtin-A to Azadirachtin-G of which Azadirachtin-E is the most effective insect growth regulator (Verkerk and Wright, 1993). Azadirachtin possess insecticidal, ovicidal, antifeedant and growth inhibiting effects against many insect pests (Akou-Edi, 1984; Schmutterer, 1990; Vietmeyer, 1992; Nawrot and Harmatha, 1994), including the storage pests (Jilani and Su, 1983; Ivbijaro, 1983a, b; Makanjuola, 1989).

The production of eggs, hatching and adult emergence in *Callosobruchus maculatus* and adult emergence in *Sitophilus oryzae* were significantly reduced when raised on cowpeas and maize respectively treated with extracts of neem leaf and seed (Makanjuola, 1989). Ivbijaro (1983a) also reported a significant

reduction in egg laying of *C. maculatus* on cowpeas mixed with neem seeds. Neem oil treatment also reduced oviposition, inhibited adult emergence and development of *C. maculatus, C. chinensis* and *C. analis* (Yadav, 1985; Babu *et al.*, 1989). That Azadirachtin inhibits the release of prothoracicotropic hormones and allatotropins (Banken and Stark, 1997), thereby affecting metamorphosis in insects (Schmutterer and Rembold, 1995) is well documented. The extract from bark, leaves, fruits and root have been used to control leprosy, intestinal helminthiasis and respiratory disorders in children **(**Nat *et al*., 1987) The bark extract is also used as tonic, astringent and useful in relieving fever, thirst, nausea, vomiting and skin diseases Sengupta *et al*.,1960. The immunomodulatory activity of the neem bark extract has also been reported **(**Schmutterer H, 1995). The medicinal and industrial uses of various parts of neem tree and the compounds isolated have been reviewed (Fujiwara *et al*., 1996).

Dental caries is caused by acidogenic and acid uric Gram-positive bacteria, primarily the *streptococci mutans*, *lactobacilli* and *actinomycetes* (Alviano and Alviano, 2007). The periodontal diseases have been linked to anaerobic Gram-negative bacteria such as *Porphyromonas gingivalis*, *Actinobacillu*s species, *Prevotella* species and *Fusobacterium* species (Loesche, 2007). *Candida albicans* is the most important causative organism of oral candidiasis which is a common oral fungal infection (Sheila *et al*., 2006). Since various parts of *A. indica* are known to possess antimicrobial properties, we tried to explore the antibacterial and antifungal activities of neem twig against selected oral cariogenic

and periodontal pathogens. Variable results have been observed in different studies of antimicrobial activities of neem against bacterial and fungal organisms. Aqueous extract of *A. indica* did not show any significant activity against the isolates obtained from the oral cavity namely *Staphylococcus auricularis*, *Micrococcus* species, *Acinetobacter lwoffii* and *C. albicans*. (Parthasarathy and Thombare, 2013). Aqueous extract was found to be less effective in antimicrobial activity in comparison to ethanol extract in a study conducted on bark and leave extracts of neem (Rathod *et al*., 2012). In a study which evaluated the antimicrobial activity of leaf extracts of *A. indica*, ethanolic and dichloromethane extracts were found to be more effective among the different extracts used (Rajasekaran *et al*., 2008). In their study, methanol extract was found to be most effective followed by petroleum ether and ethyl acetate. Extract of dichloromethane exhibited effective zone of inhibition only against prevotella species and aqueous extract failed to demonstrate effective zone of inhibition against any tested organisms. Similar findings have been observed in other study, where methanol extract of *A. indica* leaves was reported to have highest antibacterial activity compared to chloroform extract which exhibited moderate to good antibacterial activity (Koona and Budida, 2011). Thus inhibitory activities of plant extracts may be both organism and solvent dependent as it has been observed in other study (Rajasekaran *et al*., 2008). Different parts of *A. indica* have been reported to exhibit varying degrees of antimicrobial activities against bacterial and fungal species.

Neem extract has been reported to have antidiabetic, antibacterial and antiviral activity (Kirtikar and Basu, 1987). Almost every part of the tree has been in use since ancient times to treat a number of human ailments and also as a household pesticide. The extract from bark, leaves, fruits and root have been used to control leprosy, intestinal helminthiasis and respiratory disorders in children (Chattopadhyay *et al.,* 1993). Flavonoids, flavono-glycosides, dihydrochalocones, tannins and others are also important constituents of bark, leaves, fruits and flowers of neem. The biological activities and medicinal properties of neem have recently been reported (Venugopal and Venugopal, 1994). Imaran khan *et al*., 2010 studied that phytochemical analysis of *A. indica* leaves by using different solvent such as Petroleum ether, chloroform, methanol show the presence of triterpenes, glycosides and fatty acids. Other phytochemicals studied in this analysis were absent in all extract of leaves. Antibacterial activity of *A. indica* was analyzed by previous workers showed that the chloroform extract of leaves possess significant activity, than petroleum ether and methanol extracts. Early studies proved ethanol as the most efficient solvent for extracting broad spectrum of antibacterial compounds from plants. Himal paudel chhetri *et al*., (2008) reported that the ethanolic extract of *A. indica* whole plant shows presence of flavonoids and tannins only. Similarly the extract of *A. indica* is active against *E. coli* followed by *Staphylococcus aureus*. Earlier observation done by (Srinivasan *et al*., 2001) also showed the antifungal and antibacterial activity of *A. indica*.

Neem (*Azadirachta indica*) is a widely prevalent tree, mainly cultivated in India subcontinent (Karl, 1997). Various parts of the tree have been used as traditional Ayurvedic medicine in India (Brahmachari, 2004). Neem oil was often administered orally, for deforming and constipation and is applied topically to relieve rheumatism, ulcer, itching and cure chronic skin diseases (Aggarwal and Dhawan, 1995). There is evidence that neem oil has acaricidal, antibacterial, antifungal, antimalarial, antiparasitic and anti-inflammatory as well as immunomodulatory properties in different animal species (Mulla and Su, 1999; Biswas *et al*., 2002; Brahmachari, 2004; Gossé *et al*., 2005; Du *et al*., 2007, 2008, 2009; Xu *et al*., 2010; Zhang *et al*., 2010). Due to its efficacy, biodegradability and minimum side effects, azadirachtin, a tetranortriterpenoid obtained from neem seeds, has emerged as a natural biopesticide (Locke, 1995; Martinez, 2002).

According to Mulla and Su (1999) and Biswas *et al*., (2002) neem oil extracted from the seeds of *Azadirachta indica* has versatile medicinal properties, including antifertility, antifungal, antibacterial, immunostimulant, antipyretic and acaricidal activities. Chloroform and petroleum ether extracts of neem oil have also been found to exhibit potent acaricidal activity against *Sarcopte scabiei* var. *cuniculi* larvae (Du *et al*., 2008, 2009). Neem extract was also found by Da-Costa *et al*., (2010) to have inhibited the fungal growth (*i.e.* mycelia dry weight, diameter of colony and growth rate) of *Aspergillus flavus* on solid media at concentrations from 0.5 to 5.0% v/v, although it significantly increased sporulation in the same conditions. Bhutta *et al*., (2001) tested 32 different seed

diffusates against *Aspergillus alternata* and *Fusarium solani* and found that the diffusates from *Corriander sativum* and *Memoranda charata* exhibited inhibitory effects at 0.5% and 1% concentrations. Other observations were recorded against *Alternaria solani* by using *Allium cepa* extract (Khallial, 2001). Locke (1995), Martinez (2002) and Da-Costa *et al.*, (2010) reported that due to the antifungal efficacy of neem seed extract, its biodegradability and minimum side effects, azadirachtin, a tetranortriterpenoid obtained from the seed has emerged as a natural biopesticide. In addition, the percentage inhibition against the tested fungi were found to increase at different rates by increasing the concentration of neem leaf and seed extract with the result that neem seed organic extracts had higher inhibition percentage than that of neem leaf organic extracts.

An important member of Meliaceae family *Azadirachta indica* (Neem) is well known for its unique characters of fast growth and resistance to the drought conditions (Dalziel, 1955). These unique characters make all parts of the tree a rich source of traditional drugs (Biswas *et al.,* 2002; Ngure *et al*., 2009). Recently, neem has been of ecological importance and is effective as pesticide against about 200 insect species. Moreover, it has antiseptic, antifungal, antibacterial, antipyretic, anti-malaria, anti-diabetic and anti-fertility properties among several other uses (Nok *et al*., 1993, Natarajan *et al*., 2003; Fredros *et al*., 2007; Mbaya *et al*., 2010). Neem elaborates a vast array of biologically active compounds that are chemically diverse and structurally variable with more than 140 compounds isolated from different parts of the tree (Subapriya

and Nagini, 2005). Quercetin and ß-sitosterol were the first polyphenolic flavonoids purified from neem fresh leaves and were known to have antibacterial and antifungal properties (Mahmoud *et al*., 2011).

Abubakar *et al*., (2010) showed that leaves of *Tamarindus indica* extract possess better antifungal properties when compared to fruit and stem extracts. Leaf is one of the major accumulators of bioactive compounds and has therefore preferred for therapeutic purposes (Maji *et al.,* 2010). Mahmoud et *al.*, (2011) reported that leaf extracts of neem had a characteristic effect on human pathogenic fungi. Shivpuri *et al.*, (1997) noticed that the extracts in ethanol of *A. indica* had fungi toxic properties against five pathogenic fungi when tested under laboratory conditions at concentrations ranging between 500 and 1000 µg mL1. Verma *et al.*, (1998) found that a purified fraction (ethyl acetate: chloroform, 3:1) of extracts in methanol from neem seed coat showed strong antifungal activity against *A. niger* and *Curvularia lunata* with MIC of 250 ppm. They found also that the extracts in petroleum ether from the neem leaves were highly active at a lower MIC (100 ppm) against the same pathogens. Kishore *et al.*, (2001) reported that ethanolic leaf extracts of *A. indica* inhibited the conidial germination of *Phaeoisariopsis personata* by 90% to control late leaf spot of groundnut. More than 135 compounds have been isolated from different parts of neem. The compounds have been divided into two major classes: isoprenoids [like diterpenoids and triterpenoids containing protomeliacins, limonoids, azadirone and its derivatives, gedunin and its derivatives, vilasinin type of

compounds and C-secomeliacins such as nimbin, salanin and azadirachtin] and non-isoprenoids, which are proteins/amino acids and carbohydrates [polysaccharides], sulphurous compounds, polyphenolics such as flavonoids and their glycosides, dihydrochalcone, coumarin and tannins, aliphatic compounds, etc. Most of the compounds have the fungi static ability (Asif, 2012). Overall, methanol and chloroform extracts showed considerable antifungal activity over *n*-hexane in this study. This study suggested that some compounds are more active in methanolic extracts than they were present in the chloroform extracts in the present study. Similarly, Rizwana *et al.*, (2012) reported that alcoholic extracts possessed more antibacterial activity than chloroform.

Biu *et al*., (2009) observed presence of anti-nutrients like saponins, tannins, glycosides, alkaloids, terpenes and flavenoids in the aqueous extracts of the leaves of *Azadirachta indica* (neem). According to Benneth and Wallsgrove (1994) and Grayer and Harbourne (1994), a large number of constitutive plant compounds have been reported to have anti-fungal activity and well known examples include glycosides, phenols, saponins and glucosinolates. Osbourn (1996) stated that many saponins exhibit potent anti-fungal activity and are often present in relatively high levels in healthy plants and as a result have been implicated as determinants of a plant's resistance to fungal attack. Price *et al*., (1987); Fenwick *et al*., (1992) and Hostettman and Marston, 1995 stated that a number of other properties associated with saponin compounds included piscidal, insecticidal and molloscicidal

activity; allelopathic action; and anti-nutritional effects. Umar *et al*., (2002); Makein *et al*., (2007) and Wikipedia, 2007 pointed out that Azadirachtin extracts from the seeds, leaves and bark of the neem tree has been reported to have strong biological activities against insect pest, but with very low toxicity to mammals and the environment generally.

Chattopadhyay *et al*., (2004) reported antiulcer activity of neem leaf extract. The extract of neem dose-dependently inhibits gastric lesions induced by restraint–cold stress, ethanol and indomethacin. In stress ulcer model, neem extract is more effective than ranitidine but less effective than omeprazole. Mechanism of antiulcer effect of neem (*Azadirachta indica*) leaf extract is due to its action on H+-K+-ATPase. Badam *et al*., (1999) evaluated *in vitro* antiviral activity of neem (*Azadirachta indica. A. Juss*) leaf extract against group *B coxsackieviruses*. Antiviral activity of methanolic extract fraction of leaves of neem (*Azadirachta indica A. Juss*) (NCL-11) was studied for its antiviral activity and mechanism of action against Coxsackie B group of viruses. Mohanty *et al*., (2008**)** carried out antifungal activity of neem (*A. indica)* against *Lagenidium giganteum* and *Metarhiziumanisopliae* in PYG and Emerson's YpSs agar media. The minimum inhibitory concentration of neem oil for *L. giganteum* showed higher than that for *M. anisopliae*. The minimum fungicidal concentration of neem (*A. indica)* oil in PYG medium was lower than in YpSs for both fungi.

Pendse *et al*., (1977) reported anti-inflammatory, immuno suppressive and some related pharmacological actions of the

water extract of neem in albino rats and immuno suppressive effect in albino rabbits. It significantly inhibited acute inflammatory response evoked by carrageen in a doss of 50 mg/100 g given orally and intraperitoneally. In chronic inflammation produced by crctcn-oil in granuloma pouch technique, 20 mg/1 00 g of the water extract significantly inhibited granulation tissue response; the reduction in oxidative response and increase in the weight of adrenal glands were not significant. A significant inhibition of primary and secondary phases was observed Inadjuvantinduced arthritis. It significantly inhibited antibody formation by typhoid “H” antigen. Mild analgesic effects of its own as well as potentiation of morphine analgesia were possessed by the extract but it was devoid of antipyretic effect. Neem oil produced an increase in the cutaneous capillary permeability. The capillary permeability increasing action of neem oil was discernible 1 h after its application and persisted over 4 h. Histamine action was manifested within 0.5 h and lasted upto 2h. of its injection. Capillary permeability action was not observed at 24h of the application of test substances. Normal saline had no effect on capillary permeability. Investigation showed that neem oil produced increase in vascular permeability. It is likely that direct injury to mast cell granules by neem oil may be responsible for increase in vascular permeability by producing chemical injury at the site of local injection.

Bopana *et al*., (1997) reported antidiabetic and antihyperlipaemic effects of neem seed kernel powder on alloxan diabetic rabbits. In alloxan diabetic rabbits there was a significant ($P<0.001$) increase

in fasting blood glucose and urine sugar and there was a significant decrease (P<0.001) in body weight and total hemoglobin content. There was a significant increase in body weight and hemoglobin level, and a significant decrease in fasting blood glucose (FBG) and urine sugar in diabetic rabbits treated with NP, glibenclamide, insulin and in combination of NP and glibenclamide.Though the entire antidiabetic drugs used significantly decreased the FBG levels, combination therapy of NP (250 mg/kg) and glibenclamide (0.25 mg/kg) p to all the other groups. There was a significant (P<0.001) reduced greater reduction in FBG as compared amelioration of body weight and total hemoglobin content in the diabetic phosphates increased considerably in alloxan diabetic rabbits compared to the normal control. Treatment with various antidiabetic agents in the above experiments significantly reduced the enzyme activity. Treatment of NP with glibenclamide produced a significant (P<0.001) decrease of HMG CoA reductase, alkaline phosphates and serum acid phosphates activity when compared to other experimental antidiabetic agents. Liver glucose 6-phosphatase (G6P) and serum lactate dehydrogenase (LDH) activity significantly (P<0.001) reduced in alloxan diabetic rabbits. On the contrary, Hexokinase activity significantly increased by other experimental antidiabetic agents. The most significant (P<0.001) changes were observed in the combination of NP (250 mg/kg) and glibenclamide (0.25 mg/kg). From our experiments we have found out that, though both NP and glibenclamide produced significant fall in lipid parameter and enzyme activities, the changes were more prominent when

combination of NP and glibenclamide were used. Khosla *et al*., (2000) reported antinociceptive activity of *A. indica* (neem**)** in rats Tail flick reaction time was significantly increased in rats both with leaf extract and seed oil. Naloxone pretreatment partially reversed the antinociceptive action of both leaf extract and seed oil. GAA induced writhing was reduced with both neems extract and seed oil. Neem extract was more potent than seed oil.

Lloyed *et al*., (2005) reported the anticandidal activity of *A. indica* .Hexane and alcoholic extract of neem seed was found to have promising anticandidal activity. Olabinri *et al*., (1992) carried out experimental classification of the antioxidant capacity of the leaf, stem and root barks of *A. indica.* The ferric reducing antioxidant power (FRAP) and total phenolic concentration of the leaf, stem and root barks of *M. indica* and *A. indica* growing in Ogbomoso, Nigeria were evaluated in vitro. Only the leaf of *A. indica* belonged to good FRAP. Both the stem and root bark of *A. indica* and all the parts of *M. indica* investigated belonged to high FRAP. Experimental results revealed that the antioxidant capacity ranged from 6.80 - 9.20, 12.40 - 13.00 and 10.20 -13.203 mM of reduced Fe3+ for the leaf, stem and root bark, respectively in *A. indica*. In *M. indica*, the antioxidant capacity ranged from 12.20 - 15.20, 11.00 - 11.80 and 11.20 - 12.20 mM of reduced Fe3+ for the leaf, stem and root bark, respectively. The total phenolic concentration and antioxidant capacity of *M. indica*, stem bark showed a high significant positive correlation (r = 0.9439; p = 0.05). The total phenolic concentration of the root bark of *A. indica* showed a high positive significant correlation with antioxidant capacity (r= 0.9850;

p= 0.05). All the plant parts examined might be exploited in clinical medicine as protective factors because of their good and high antioxidant capacities.

The phytochemical test results indicated high scores for saponins, moderate scores for tannins and glycosides while alkaloids, terpenes and flavonoids had low scores. According to Anyanwu and Dawet (2005) these constituents found in plants are known to have anti protozoal and anti bacterial activities. Flavonoids especially, are of a potential benefit to human health (Jouad *et al.*, 2001).

Azadirachtin extracts from the seeds, leaves and bark of the Neem tree has been reported to have strong biological activities against insect pests, but with very low toxicity to mammals and the environment, generally (Umar *et al*., 2002; Makeri *et al*., 2007; Wikipedia, 2007). Registered Neem insecticide formulations Neemros® and Neemroc EC® have also been found to be effective against insect pests of vegetables but safe to their natural enemies (Akol *et al.*, 2001).

Neem leaves are eaten as vegetable, and twigs are used as toothbrushes. Neem is a nature's pharmacy (Vietmeyer, 1992). Today, researchers are saying that neem could be called "a wonder tree" and eventually expect it to benefit everyone on the planet. The medicinal properties of neem have been known since time immemorial. The earliest ayurvedic literature refers to the benefits of all parts of this majestic tree - fruit, leaf, bark, flower and root (Schmutterer, 2002; Subapriya and Nagini, 2005) Neem

elaborates a vast array of biologically active compounds that are chemically diverse and structurally complex (Vietmeyer, 1992; Siddiqui *et al*., 1992; Garg *et al*., 1998; Ramesh and Bal Subramanian, 1999; Koul, *et al*., 2003; Kaur *et al*., 2004; Koul, 2004; Senthil *et al*., 2006). More than 140 compounds have been isolated from different parts of neem. It's strong garlic odour (alliaceous) and its medicinal properties have been attributed to the presence of sulphur containing compounds (Nadkarni and Nadkarni, 1954; Dey and Mair, 1973; Balandrin, *et al*., 1988; Mubarak and Kulatilleke, 1990; Koul; 2004) and a number of primary amines and secondary amines were detected (Atawodi and Spiegel alder, 1994). Neem is used in treatment of various skin diseases (Dhawan and Ratnaik, 1993) and it has antibiotic properties (Sharma, 1993). Neem has been demonstrated to exhibit anti-inflammatory, antipyretic, antiarthritic (OKpanyi and Ezeukwa, 1981; Kaur *et al.,* 2004), antihyperglycaemic (Murthy *et al.*, 1978), diuretic(Binde *et al.,* 1958) immuno modulatory (Upadhyay *et al.,* 1993; Arivazhagan *et al*., 2000), antiulcer (Dorababu *et al*., 2004), antimicrobial (Patel and Trivedi, 1962); Khan and Wassilew, 1987; Zeitlin *et al*., 1997; Kusumran *et al.,* 1998; Badam *et al*., 1999; Sai Ram *et a*l., 2000; Udeinya, 1993; Parida *et al*., 2002; Siddiqui *et al.,* 1992), antimutagenic and anticarcinogenic potential effect (Kusamran *et al.,* 1998). Neem is one of the most promising botanical insecticides at present (Dimetry, 1993; Moustafa 1993. Its products are known to have strong pesticidal properties (Schmutterer *et al*., 1981; Schmutterer and Ascher, 1984, 1987; Hadis *et al.,* 2003, Koul, 2004),

ascaricidal (Capinera and Froeba 2007) and larvicidal (Okumu *et al.,* 2007). Regarding neem oil, it has reported anti-fertility; stimulate immune response (Upadhyay *et al.,* 1992), spermicidal and abortifacient effects (Riar *et al.,* 1990; 1991) and contraceptive potential (Sinha *et al.,* 1984a, b; Garg *et al.,* 1994, 1998; Sharma *et al.,* 1996), and many other variable biological activities (Subapriya and Nagini, 2005).

The immunosuppressive viral diseases, of which IBD threaten the poultry industry by causing heavy mortality and economic loss of production (Balamurugan and Kataria, 2006). Neem is traditionally being used as curative against certain fungal and bacterial diseases. However, evaluation of its antiviral properties is limited to few viruses' Viz. small pox, Fowl pox, polio and HSV as assessed by virus inhibition assay (Rao *et al.,* 1969; Rai and Sethi, 1972; Reddy and Sethi, 1974). Neem leaves have been reported to suppress HIV, Dengue virus type-2, group B Coxsackie (Upadhyay *et al.,* 1993; Badam *et al.,* 1999; Parida *et al.,* 2002). A fraction from neem oil (NIM-76) has also been reported to suppress Polio viruses (Sai Ram *et al.,* 2000).

2.1. *Tribolium castaneum* Herbst. (The red flour beetle):

2.1.1. Description and identification:

Tribolium castaneum (Herbst.) commonly known as the red flour beetle, belongs to the family Tenebrionidae, order Coleoptera. The flour beetles are known by a number of common names. Grain millers refer to species of *Tribolium* and other closely related

beetles of similar appearance as (flour beetles), (flour weevils), (red weevils), or (bran bugs). The accepted common name of *T. castaneum* is the rust red flour beetle (Good, 1936). *T. castaneum* adult is 3 to 4 mm in length, parallel sided and reddish brown in colour. The antennae are composed of eleven segments with the last three comprising the club. The compound eyes are partly divided horizontally by a back ward projection of the head.

2.1.1.1. Distribution and host range:

T. castaneum is cosmopolitan, occurring all over the world wherever stored cereal products are to be found. As it lives inside buildings and may easily be carried from place to place in small quantities of foodstuffs, these beetles are likely to be recorded from practically any part of the world. *T. castaneum* is essentially an insect of warm climates (Good, 1936).

2.1.1.2. Biology of *Tribolium castaneum* (Herbst.):

2.1.1.3. Life history:

2.1.1.4. Mating, pre- oviposition period and oviposition rate:

Mating usually begins within a day or two after adult emergence and probably continues at frequent intervals throughout the life span of the insect. Good (1936) stated that the pre-oviposition period of *Tribolium* species might range from four to an indefinite number of days according to temperature. He also stated that adults emerging during winter when kept under room temperature did not lay eggs until the approach of warm weather. Khalifa and Badawy (1955 a) reported that the shortest pre-oviposition period of *T. castaneum* was 4.9 days and the longest was 12.92 days at

the beginning of winter. Howe (1962) found that the shortest pre-oviposition period was 10 days, but it can extend to 15 days at 25^0C and 70% R.H. It is more difficult to have good estimates of the oviposition rate of most stored products insects than it is to measure their developmental period. The total number of eggs laid by a female of *T. castaneum* was 360.4 ± 27.9 eggs (Good, 1936; Howe, 1962). It was also stated that the fecundity varied with food. Robert (1985) stated that each female of *T. castaneum* may deposit 400 to 500 eggs (average 450 eggs) depending on the food quality. In *T. castaneum* all unmated females laid fewer eggs than did the mated ones. The average number of eggs laid by mated and unmated females was 139.1 and 45.1 respectively (Khalifa and Badawy, 1955a).

2.1.1.5. Incubation period and development:

2.1.1.6. Incubation period:

Howe (1956) reported that eggs did not hatch at any humidity at 15°C or 17.5°C nor at 10% R. H at 40°C. Good (1936) reported total mortality of eggs under uncontrolled humidity at 35°C, and high mortality at 32°C. He also added that 30°C appeared to be close to the optimum incubation temperature. Good (1936) studied egg incubation period of the red flour beetle on different types of food including middling, bran, whole wheat flour, corn meal, oat meal and white flour. He found that eggs kept at room temperature in April (temp. ranged from 18.5°C to 28.5°C and R.H ranged from 22 to 43%), required an average of 8.8 days to hatch. Eggs kept at room conditions in November (temp. ranged from 18 °C to 29 °C and R.H ranged from 27 to 47%), required 8 to 11 days with an average of 10 days. Khalifa and Badawy (1955b) stated that hatching did not occur below a temperature of 16°C.

2.1.1.7. Larval development:

The larvae are fairly active and live more or less concealed in the food in which they bury themselves if disturbed. They are a little less tolerant than eggs to extreme conditions (Howe, 1962). Both Howe (1962) and Shazali (1982) concluded that 35°C and 70 % or higher %R.H. were optimum conditions for larval development. Good (1936) reported that there was no fixed number of Laval instars, the number ranging from 5 to 11 or more, and that the usual number was 7 or 8. This variation is due to both external conditions, such as food, temperature and to inherent individual

characteristics. The larval period of *T. castaneum* ranged from 22 to 100 days depending on temperature and food. Good (1936) also stated that the optimum temperature for development approached 30°C. Khalifa and Badawy (1955b) reported a short larval period of 21.1 days in August (Mean temp. 30°C) and a much longer period of 40.5 days at the beginning of March (Mean temp. 15° − 17°C).

2.1.1.8. Pupal development:

The pupae when compared with the eggs and the larvae seem to be less affected by external conditions. Good (1936), Khalifa and Badawy (1955 b) and Howe (1956) stated that the food of the larva had no obvious effect on the duration of the pupal stage, which was similar for larvae reared on wheat or groundnuts. Khalifa and Badawy (1955b), who reared *T. castaneum* under uncontrolled conditions stated that the shortest pupal period was 5.8 days in July (Mean temp. 30°C) and the longest was 18.5 days in October (Mean temp. 20°C). Howe (1956) and Shazali (1982) reported that temperature had a significant effect on the pupal period and that the shortest pupal period of *T. castaneum* (4.5 days) was obtained at 35°C at which pupal mortality was low.

2.1.2. Total development:

In most insects the developmental period from egg to adult varies considerably according to the prevailing environmental conditions. This variability is mostly due to the varying rates of growth of the larval stage, which is greatly affected by environmental conditions. Khalifa and Badawy (1955 b) and Howe (1956) stated that the optimum conditions for rapid development of *T. castaneum* from

oviposition to the emergence of the adult lies between 35°C and 37.5°C at 70 % R.H. They added that the development of one complete generation exceeds 30 days. Shazali (1982) reported that the conditions for the shortest development of *T. castaneum* are 35°C and 70 to 80 % R.H., and the longest development occurred at 25°C. It is clear that optimum conditions for *T. castaneum* result in a short life cycle, which contributes to a very high rate of increase. Such rate of increase could not be sustained for long and would be reduced by the effects of cannibalism, parasitism, predation, disease and competition for space and food. Successful dispersal is achieved by flying and is not only dependent upon the movement of infested food. In late afternoons many individuals fly from the surface of infested sacks. The beetles may also be observed flying from any storage facilities (Krishnamurthy *et al*., 1987).

2.1.3. Nature of damage and economic importance:

T. castaneum is frequently referred to as a secondary pest since it is unable to feed on or attack sound grains (Howe, 1956). It can survive on dry commodities and is particularly troublesome on milled cereals and animal feeds, but does not multiply rapidly on dry cereal grains, if these are undamaged and are free of grain fragments or other dockage (Anon, 1986)). *T. castaneum* prefers the embryo and may feed on whole kernels, if the moisture content is 12 % or higher. In addition to grains, *T. castaneum* attacks dried fruits (Robert, 1985). Shazali (1982) reported that *T. castaneum* is able to damage whole sorghum grains at high temperature and relative humidity, by feeding on microscopic lesions. Furthermore

about 20 % of the kernels are usually damaged during harvesting and threshing processes in the Sudan. Thus, the so -called secondary pests might not need the help of primary pests. Where these pests are present in large numbers the flour becomes grayish and discolored and will mould more quickly than clean flour. Some times the disagreeable, pungent odor given off by the insect scent glands (Quinones) is incorporated into the flour, giving it disgusting taste and odor (Good, 1936; Anon, 1986).

2.1.3.1. Control of the flour beetle:

Basically, control is aimed at the crevices, where the insect hides within bagged products. Several physical and biological methods are used (Shazali, 1987).

Hygiene methods:

Hygiene is the first step towards minimizing losses caused by the pests during storage. Cleanliness of the premises is one of the most important means of minimizing losses caused by pests during storage. The removal of residues and dirt would also make pesticide treatments effective (Krishnamurthy *et al*., 1987)

Physical methods:

The physical methods which are used to minimize the number of

insects are keeping the temperature beyond the optimum range for the insects, reducing the moisture content of the grains to a lesser extent than that needed by the insects for development, irradiation, sticky traps, sieving, sunning, cold storage (Shazali, 1987) and

airtight storage such as in the underground pits (matmoras), drums, plastered bins and plastic bags (Mc Farlane, 1970).

Biological methods:

These include many natural enemies, such as predators and parasites mainly hymenopterans and hemipterous species attacking some insects of stored products, but the intensive use of pesticides applied to the stores caused drastic reduction in their populations. For this reason attempts have been made to use these natural enemies (Shazali, 1987). The only successful biocontrol method, which may economically be applied, is the use of the bacterium, *Bacillus thuringiensis* (Mc. Ganghey, 1976).

Chemical control:

The types of chemical available for use against insect pests of stored products are some fumigants (curative treatment) and also some liquid and powder formulations of contact insecticides (prophylactic treatment).

Chapter-3

General Materials and Methods

3.1. Selection of test organisms:

Field trials and laboratory experiments were carried out to assess the efficacy of the neem extracts with respect to concentration, dose, and treatment area. The neem-based extract was provided as a semi-solid formulation. To confirm the identity of the principal chemical compounds within the extract biochemical analyses were carried out. Crucial to any investigation of plants with biological activities is the availability of suitable bioassays for monitoring the required effects. The test systems should ideally be simple, rapid, reproducible and inexpensive. If active principles are only present at low concentration in the crude extract then bioassay should be high enough sensitive for their detection. Another factor of special relevance to plant extracts is the solubility of the sample and finding a suitable system can pose problems.

The bioassays to carry on tests for insecticidal activities, larvicidal activities and also for repellent potentials of the extractives of *A. indica. Tribolium castaneum* was selected, because it is an easy cultivable and noble laboratory insect. The life histories made these insects as popular choice as test insects for biological studies. For cytotoxicity test *Artemia salina* was selected, since it is being used in such cases as a model test agent. A number of bacteria and fungi were selected to carry out further efficiency tests of the extractives.

3.1.1. Collection and culture of *T. castaneum:*

Adult beetles of *T. castaneum* used in the present investigation were collected from the stock cultures of the Insect research Laboratory, Department of Zoology,University of Rajshahi, Bangladesh and reared as mass-cultures and subcultures to be used in the experimentations. The brine shrimp cysts were collected from any of the aquarium shops of Rajshahi sahab bazar.

Mass cultures were maintained in plastic containers (1200ml) and sub-cultures in beakers (1000 ml) with the food medium. The beakers were kept in an incubator at 30^0C ± 0.5^0C without light and humidity control. Each container and beaker contained 250g and 150g of food respectively. About 200 adults in each container and 100 adults in each beaker were introduced. The cultures were checked in regular interval of 3 days and eggs and larvae were separated to increase properly. A crumpled filter paper was placed inside each container and beaker for easy movement of the beetles. The containers and beakers were covered with muslin cloth tightly fixed with the help of rubber bands to avoid possible escape of the beetles (Plate -).

Plate 3: Cultures of *T. castaneum* in an incubator.

Plate 4: *T. castaneum* under natural conditions.

3.1.2. Preparation of food medium:

Fresh whole-wheat flour was used as a standard food medium for the insect species. The flour was sterilized at 120°C for 6 h in an oven. A standard mixture of flour and brewer's yeast at 19:1 ratio (Park and Frank, 1948; Park, 1962) was used as food medium throughout the experimental period. Both flour and yeast were previously passed through a 250 micrometer sieve and sterilized. The prepared food was not used until at least 15 days after sterilization in order to have its moisture content being equilibrated with the environment (Khan, 1981; Mondal, 1984c).

3.1.3. Collection of eggs:

In regular interval the eggs were collected by sieving the food medium by two sieves of 500 and 250 mesh separating the adults and eggs respectively following the methods of Khan and Selman (1981).These eggs were then transferred to petri dishes (90 mm diam.) containing a filter paper at the bottom and incubated at 30^0C (Mondal and Perween,1997).

3.1.4. Collection of newly hatched larvae:

After 4/5 days the larvae hatched out under the provided condition and the newly hatched larvae were then collected with a fine brush and shifted to the fresh food medium. The larvae are yellowish white in color and long cylindrical in shape. It appears 1 mm in length after hatching.

3.1.5. Collection of matured larvae:

The larval instars were determined by counting the number of exuviate (larval skin) deposited in the food medium according to Good (1936), and Mondal (1984a). Two days old larvae were considered as 1st instar larva while 2nd, 3rd, 4th and 5th instar larvae were considered on 3rd, 6th, 9th and 12th day from hatching respectively. Depending on these days according to larval instar 16 days old larvae have been considered as matured larvae. Larval cultures were maintained in an incubator at the same temperature without light and humidity control. The food medium was changed after every three days by a fresh one to avoid contamination by the larvae (Park, 1934; Mondal, 1984c).

3.1.6. Collection of adults:

A huge number of beetles were reared to get a regular supply of the newly formed adults. When sufficient adults produced in the sub-cultures, they were collected from the food medium. For this purpose some pieces of filter paper were kept inside the beaker on the food. Adults crawled upon the paper and then the paper was taken out with a set forceps. Beetles were then collected in a small beaker (100 ml) with the help of a fine brush.

3.2. Collection and culture of brine shrimp nauplii for cytotoxicity test:

Brine shrimp lethality assay was performed according to the simplified method of Meyer *et al*., (1982). There are many species within the genus of *Anostraca*, but the *A. salina* is very nice to grow, since the rate of successful hatches is very high. To conduct

cytotoxicity test the brine shrimp nauplii were used because of its easy hatching and easy to use in the experiment. The eggs (cysts) were collected from aquarium shops. For their easy hatching and use the requirements were as follows:

- Salt water**:** 1.5 - 3 tablespoons of marine salt was added to 1 liter of pond water;
- Temperature**:** 26-28ºC (80-82ºF);
- Light**:** The beaker was placed near a window with sunlight;
- Aeration**:** Picking up some water carefully with a spoon and let it drop back to the beaker at least twice a day [but a small aquarium pump with a little air-stone is better];
- Special attention**:** Brine shrimp eggs are sometimes very buoyant. Swirling of the water was done to knock down the eggs.

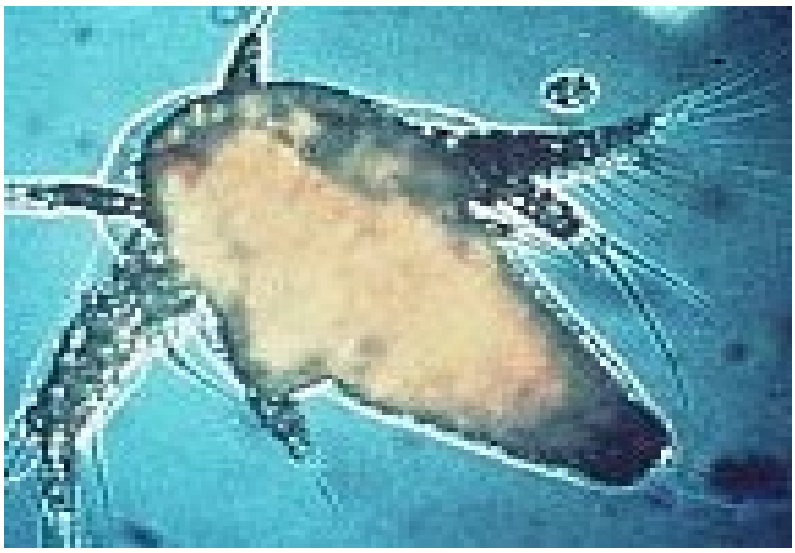


Plate 5 : *Artemia salina* (Brine shrimp) nauplii

The cysts absorbed water and hatched after 24-48 hours, depending on their environment. Freshly hatched *A. salina* called

nauplii and have a size of just 0.25mm (0.01inch). They molt like any other crawfish. When they grow to adult they molt about 17 times. Freshly hatched nauplii were used in this experiment.

3.2.1. Selection of microorganisms as test agents:

3.2.1.1. Test agents for antibacterial activity:

Antimicrobial activity of any plant or parts of a plant can be detected by observing the growth response of various microorganisms to the extracts of a plant or parts of a plant, which is placed in contact with them. Fourteenth pathogenic bacterial isolates were selected for the test, 6 of which were gram positive and the remaining 8 were gram negative (Table).

Table1: List of the pathogenic bacteria used in this investigation

Serial No.	Name of test organism	Strain #
Gram positive		
1.	*Staphylococcus aureus*	ATCC-259233
2.	*Bacillus cereus*	-
3.	*Bacillus megaterium*	QL-38
4.	*Bacillus subtilis*	QL-40
5.	*Sarcina lutea*	-
6.	*Streptococcus- β-haemolyticus*	CRL
Gram negative		
7.	*Salmonella typhi*	-
8.	*Shigella dysenteriae*	AL-35587
9.	*Shigella shiga*	-
10.	*Shigella sonnei*	AJ-8992
11.	*Shigella boydii*	AL-17313
12.	*Escherichia coli*	FPFC-1407
13.	*P. aeruginosa*	-
14.	*Proteus sp.*	-

3.2.1.2. Collection and culture of test bacteria:

Microbiological cultures can be grown in petri dishes of different sizes that have a thin layer of agar-based growth medium. Once the growth medium in the petri dish is inoculated with the desired bacteria, the plates are incubated at the best temperature for the growing of the selected bacteria (for example, usually at 37 degrees Celsius for cultures from humans or animals, or lower for environmental cultures).These organisms of pure culture were primarily collected from the Department of Microbiology, University of Dhaka; Institute of Nutrition and Food Science (INFS), University of Dhaka and the Plant Pathology Laboratory of the Department of Botany, University of Rajshahi, and were further cultured at the Molecular Biology Laboratory, Institute of Biological Sciences, University of Rajshahi.

Culture media:

A number of culture media are available to use in the demonstration of antibacterial activity of the test substances. These are:

i) Nutrient agar medium

ii) Nutrient broth medium

iii) Mueller-Hinton medium

iv) Tryptic Soy broth (TSB) medium

v) Trypticase Soy agar medium

vi) *Staphylococcus* defined medium

vii) Adams and Roe medium

viii) NTH agar or broth medium

While the nutrient agar medium was adopted to conduct experiments in this investigation.

Table 2: List of the composition of nutrient agar medium.

Ingredient	Amount
Bactopeptone	0.5 gm
Sodium chloride	0.5 gm
Bactoyeast extract	1.0 gm
Bactoagar	2.0 gm
Distilled water	100 ml
pH	7.2 ± 0. 1 at 25°C

Preparation of the nutrient agar (DIFCO) medium:

The growth of bacteria in the research, teaching or clinical laboratory is of great importance. This is because research labs may need the bacteria to perform a specific task, the teaching lab needs the bacteria for learning and /or the clinical lab functions to identify disease-causing bacteria for appropriate treatment. Bacteria, however, are a bit finicky when it comes to growing on artificial (man-made) media. Not all bacteria grow optimally on the same kind of medium, nor do all bacteria grow optimally at the same temperature (but that's another experiment). Although the growth of bacteria on different kinds of media will be studied in a later experiment, the preparation of media so bacteria can be "planted" to grow is the focus of this experiment. Additionally, this experiment will supply the student with the basic laboratory skills for media preparation.

The instant nutrient agar (DIFCO) medium was weighed and then reconstituted with distilled water in a conical flask according to specification measurement (2.3% W/V). It was then heated in a water bath to dissolve the agar until a transparent solution was obtained.

Preparation of fresh culture of the pathogenic organisms:

The nutrient agar medium was prepared and dispersed in a number of clean test tubes to prepare slants (5 ml in each test tube). The test tubes were plugged with cotton and sterilized in an autoclave at 121°C and 15 lbs/sq. inch pressure for 15 minutes. After sterilization, the test tubes were kept in an inclined position for solidification. These were then incubated at 37.5°C to ensure sterilization. The test organisms were transferred to the agar slants from the supplied pure cultures with the help of an inoculating loop in an aseptic condition. The loop was burnt after each transfer of microorganisms to avoid contamination very carefully. The inoculated slants were then incubated at 37.5°C for 24 hours to assure the growth of test organisms. These fresh cultures were used for the sensitivity tests.

Selection of test method:

The primary assay can be done in three ways, such as-

(a) Diffusion method;

(b) Dilution method; and

(c) Bioautographic method.

However, the diffusion method was used in this investigation.

Principles of the diffusion method:

Diffusion assay (Barry, 1976) is based on the ability of antibiotics to diffuse from a confined source through the nutrient agar gel and create a concentration gradient. If the agar is seeded or streaked with a sensitive organism, a zone of inhibition will result where the concentration exceeds the minimum inhibitory concentration (MIC) for the particular organism. In this method, measured amount of the test samples are dissolved in definite volumes of solvent to give solutions the known concentrations (µg/ml). The sterile (BBL, Cocksville, USA) filter paper (5 mm diam.) discs were impregnated with known amounts of the test substances and dried. These test material discs were placed on plates containing nutrient agar medium seeded with the test organisms. These plates were kept at a low temperature (40C) for 24 hours to allow maximum diffusion. A number of events took place on the discs simultaneously that includes-

i) The dried discs absorb water from the agar medium and the material under test is dissolved.

ii) The test material diffuses from the discs to the surrounding medium according to the physical law that controls the diffusion of molecules through agar gel.

iii) There is a gradual change of test material concentration in the agar surrounding each disc.

To determine the most optimal concentration of extracts to be used in this study, sterile 7.5 mm filter paper discs were treated with 50 and 200 µl of the chloroform and methanol extracts (while the only

solvents were used as control). The bacteria were inoculated on full–strength nutrient agar (Qualigens Fine Chemicals, Prod # 58673) by suspending loops in sterile de-ionized water. The bacterial suspension was then smeared on agar plates with a sterile glass-rod to ensure the entire surface of the agar had an even coating of the bacterial suspension. The test plates were divided into several areas and one filter paper disk was placed on each of the areas. The plates are then kept in an incubator (37ºC) for 12-18 h to allow the growth of the organisms. If any of the test materials has antimicrobial activity, it will inhibit the growth of microorganisms just giving a clear distinct zone called 'zone of inhibition'. Biological activity of the *A. indica* components on bacterial growth was quantified in this way by measuring the diameter of zones of inhibition (in term of mm) deducing the size of the treated filter paper discs. The size of the inhibitory zone depends principally on the following factors-

i) Intrinsic antimicrobial sensitivity of the test sample.

ii) Growth rate of the test microorganisms.

iii) Diffusion rate of the freshly seeded test organisms.

iv) Concentration of the freshly seeded test organisms.

v) Amount of test sample on disc.

vi) Thickness of the test medium in the Petri dishes.

vii) Composition of the culture medium.

viii) Size of inoculum

ix) Time of incubation

x) Temperature of incubation.

Test materials used:

I) Chloroform and methanol extracts of different parts of *A. indica*.

II) Amoxicillin, (30µg/disc) as standard disc.

Secondary assay:

The simple assay quantities the relative potency, such as Minimum Inhibitory Concentration (MIC), of the lowest concentration of an antimicrobial agent required to inhibit the growth of the microorganisms *in vitro.*

Serial #	Apparatus and reagents to conduct antibacterial assay
1.	Crude extracts of chloroform and methanol
2.	Standard disc (Amoxicillin -30µg/disc).
3.	Chloroform and methanol
4.	Alcohol (95%)
5.	Filter paper discs (Sterilized)
6.	Petri dishes (120 mm diam.)
7.	Inoculating loop
8.	Sterile cotton
9.	Test tubes
10.	Sterile forceps
11.	Micropipette (10 µl-100 µl)
12.	Nose mask and hand gloves
13.	Spirit burner and match box.

14. Rectified spirit
15. Nutrient agar media(DIFCO)
16. Laminar air flow unit (Bio-craft and Scientific Industries, India)
17. Incubator (Osk-9639A, Japan)
18. Refrigerator (Artston, Italy)
19. Autoclave (ALP Co. Ltd. KT-30L, Japan)

Sterilization procedure:

The antibacterial screening was carried out in a laminar air flow unit and all types of precautions were highly maintained to avoid any type of contamination during the test. UV light was switched on for half an hour before working in the laminar hood to avoid any accidental contamination. Petri dishes and other glass-wares were sterilized in the autoclave at 121°C temperature and a pressure of 15 Ibs/sq. inch for 15 minutes. Micropipette tips, culture media, cotton, forceps, blank discs etc. were also sterilized.

3.2.1.3. Test agents for antifungal activity:

Plant derived compounds may offer potential leads for novel agents against systemic fungal diseases (Hufford and Clark, 1988) in man and plants. Chloroform and methanol extracts of *A. indica* samples (leaves, root bark, root wood, stem bark, stem wood, flower and seeds) were used in this investigation for the detection of antifungal potentials.

Table 3: List of the pathogenic fungi used in this investigation.

Serial No.	Name of test organisms
1.	*Fusarium vasinfectum*
2.	*Aspergillus fumigatus*
3.	*Aspergillus niger*
4.	*Aspergillus flavus*
5.	*Candida albicans*
6.	*Penicillium notatum*

3.2.1.4. Collection and culture of test fungi:

General purpose media that are commonly used for fungal culture are Sabouraud dextrose, malt extract and less commonly brain heart infusion medium. To prevent contamination of the medium by bacteria, chloramphenicol is used, but prevents the growth of *Actinomyces*, which others grow well on Sabouraud dextrose agar. For reducing the frequency of environmental fungal growth, cycloheximide is added, but this reduces the yield of many opportunistic fungi including *Aspergillus* spp.,*Cryptococcus neoformans* and Mucorales isolates. Therefore if cycloheximide is used, one agar plate not containing it should also be used in parallel. The fungal strains used in the sensitivity tests are given above. The pure cultures of the strains were collected from the Department of Pharmacy, Univesity of Rajshahi and cultures were maintained in the Molecular Biology Laboratory, Institute of Biological Sciences, University of Rajshahi, Bangladesh

Culture media:

Potato dextrose agar (PDA) media were used to perform the antifungal activity tests and for the maintenance of the subcultures of the test organisms. The composition of the medium is given below:

Table 4: Composition of the PDA medium.

Ingredient	Amount
Potato	20.0 gm
Dextrose	2.0 gm
Agar	1.5 gm
Distilled water	100.0 ml

Preparation of the media:

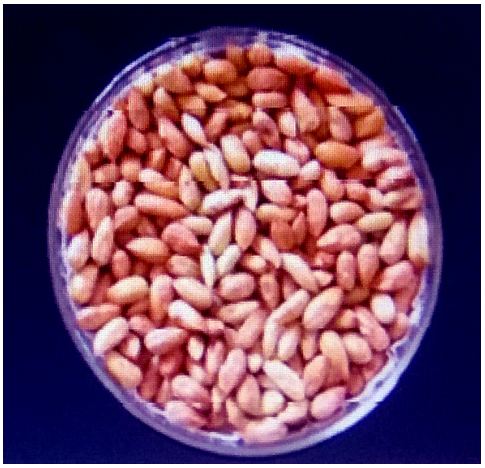
The constituents of the media were accurately weighed and dispersed in a conical flask with distilled water. It was heated in water bath to dissolve the ingredients until a transparent solution was obtained. The pH of the medium was adjusted to 5.6. The volume was adjusted by adding distilled water and sterilized in an autoclave at 121°C and 15 lbs/sq. inch pressure for 15 minutes.

3.3.1. Collection of plant materials:

In order to arrive at useful compounds in the shortest possible time, careful selection of plant material is obviously very important. Random collection is one method but it is more judicious to base the selection on certain criteria. By way of illustration, plants used in traditional medicine are more likely to provide pharmacologically active compounds (Huxtable,1992).Similarly, folk used or popularly

known very common toxic plants could be taken with desirable output, and one of the rotenone producing plant, *A. indica* has been selected for a thorough investigation.

Seeds (with endocarp)



Seeds (without endocarp)

Fruits

Stem wood

Plate 6: Different parts of *A. indica*

Stem bark

Root wood and Root bark

Leaves (Dry)

Leaves (Green)

Plate 7: Different parts of *A. indica*

Plate 8: Dust of different parts of *A. indica*

Incase of very small plants, such as herbs, shrubs, grass, etc. normally the whole plant is subjected for extraction, because the distribution of constituents generally not vary too much. The presence of constituents in the heart-wood may disappear in the leaves; similarly constituents in the roots may not be the same that present in the fruits. Being a large timber plant, the distribution of compounds in different parts of this plant is obviously different and thus different parts of *A. indica* viz. leaves, root bark, root wood, flowers, seeds, stem bark and stem wood have been collected from the Rajshahi University Campus and Meherchandi area near Rajshahi University.

3.3.2. Chemical extraction of the collected materials:

The fresh plant materials were processed through the following way-

Leaves: Leaves were spread out to dry without heaping the material together. It was done under the shade avoiding direct sunshine.

Flower: Flowers were collected and were spread out to dry without heaping the material together. It was done under the shade avoiding direct sunshine.

Root bark: Roots were collected by digging up without damaging them and shook and brushed away excess soil without washing them with water. The root bark was collected by striping out from the stem, and cutting them into small pieces as thin as possible and were dried thoroughly in a well-ventilated place.

Root wood: After removal of the root-bark, the root-wood was collected and cutting into small pieces as thin as possible, and were spread out to dry thoroughly under a shade.

Seeds: Peeling out the fruit shells the seeds were cut into small pieces and spread out to dry under a shade.

Stem bark: Stem bark was processed by striping them out from the stem and cutting into small pieces as thin as possible and thoroughly dried in a well-ventilated room.

Stem wood: After peeling out the bark, the stem-wood was processed by cutting them into small pieces as thin as possible and dried under a shade.

All the plant materials were individually powdered in a grinder machine. The powdered materials were weighed and placed in separate conical flasks to add sufficient amount of chloroform and methanol (500g × 1500ml × 3 times followed by filtration through Whatman filter paper at 24 h interval in the same collection flask) to yield the first extracts of the leaves, root bark, root wood, flowers, seeds, stem bark, and stem-wood separately (Plate -).The

output extracts were poured in to glass vials and preserved in a refrigerator at 4ºC with proper labeling (Plate 8 and 9). For each of the samples two solvents have been used separately and successively.

Plate 9: Filtration of chloroform and methanol extracts.

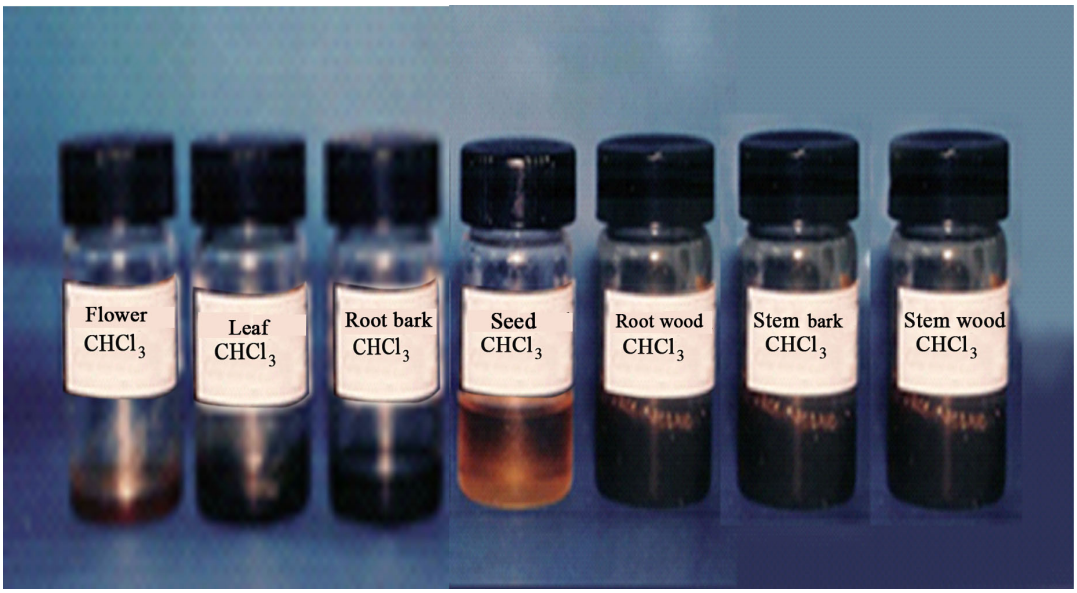


Plate 10: Chloroform extracts in vial

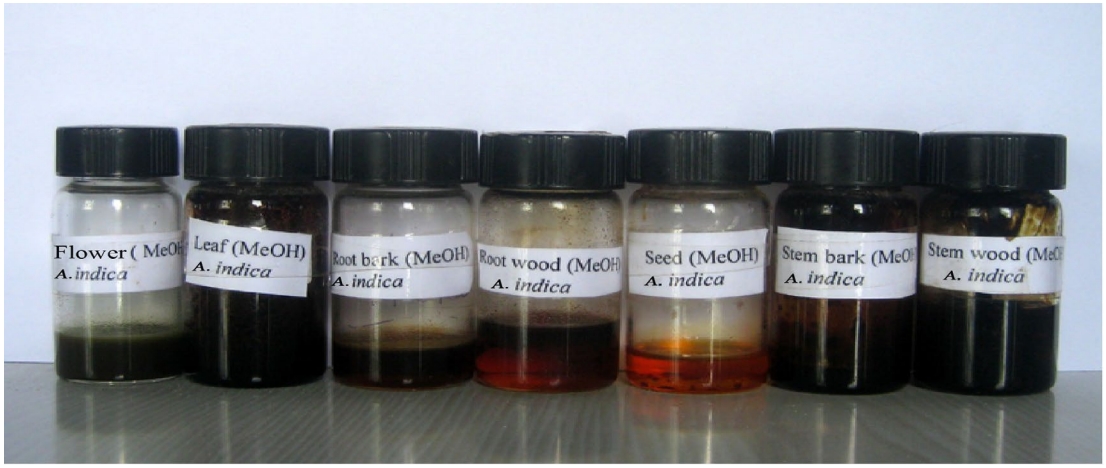


Plate11: Methanol extracts in vial

The pathway for the extraction, in detail, used in this investigation is given in Fig.

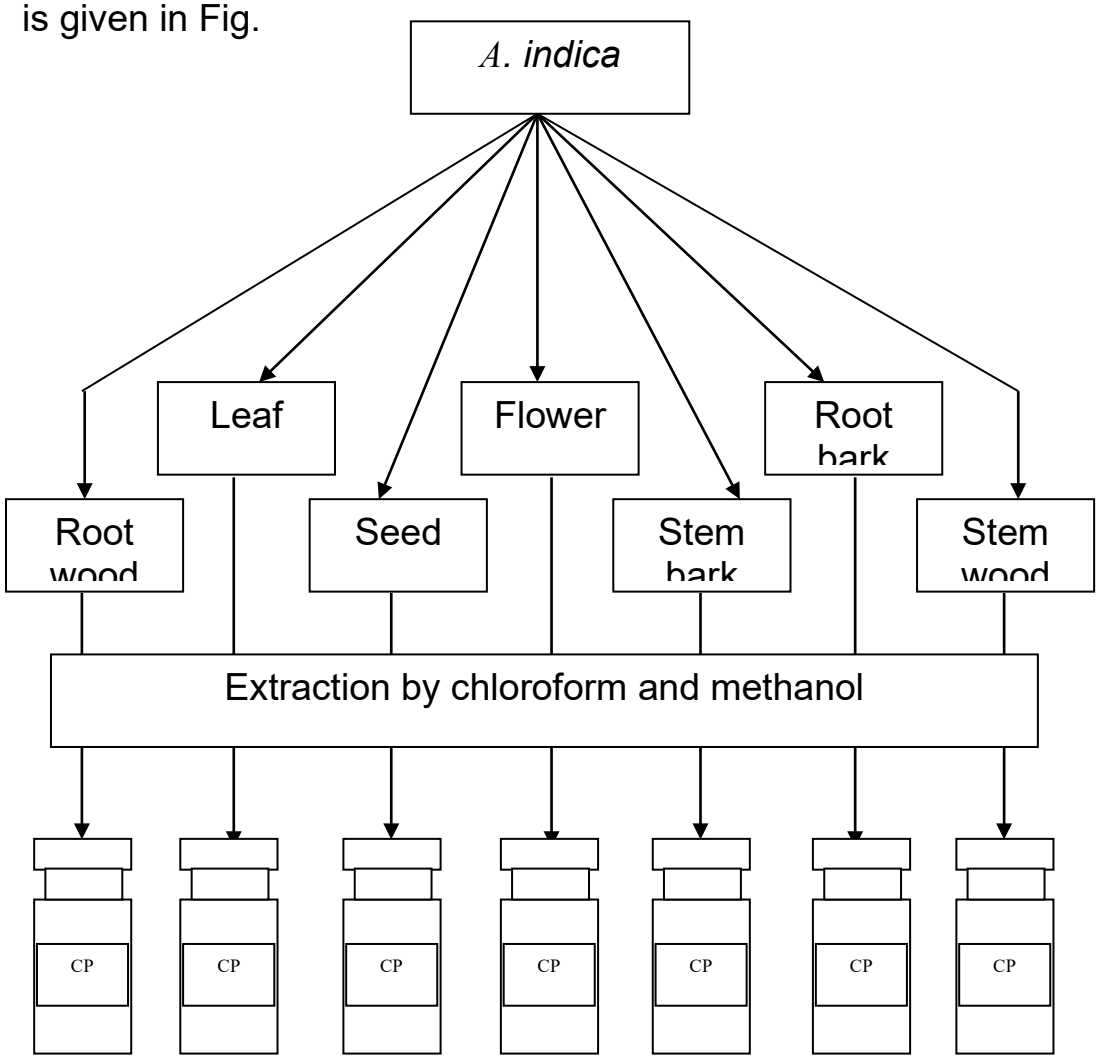


Fig.4 : Collection of extracts from different parts of *A. indica*

3.4.1. Crude extract bioassay:

For the selection of bioassays to employ in research on plant constituents, the first step is to choose suitable target organisms. The complexity of the bioassay has to be designed as a function of the facilities and resources available. A list of bioassays taken in this investigation is shown in below:

Types of tests	**Test agents**
Insecticidal	*T. castaneum* (Hbst.) adults
Larvicidal	*T. castaneum* (Hbst.) larvae
Repellent activity test	*T. castaneum* (Hbst.) adults
Cytotoxicity test	*A. salina*
Antimicrobial activity test:	
1. Antibacterial	Fourteenth pathogenic bacteria
2. Antifungal	Six pathogenic fungi
Phytotoxic activity test	

3.4.1.1. Preparation of doses for insecticidal assay:

This is also one basic application method for doses of toxic substances to any insect population. The test material has been dissolved in an organic solvent with a certain concentration to apply to a Petri dish of known surface area. After application being volatile the solvent evaporates out immediately simply with the atmospheric temperature. Thus, the ingredient goes to make film

on the surface of the Petri dish. Released insects within this captivity might have contact with the substance distributed evenly on the floor. However, being covered with the upper lid of the Petri dish there could have a captive environment with the extract distributed even in the air inside and may cause mortality by suffocation. Mortality suffocation may cause promptly if there is any volatile bioactive principles in the test material.

All extracts were diluted with the solvents in which they were extracted and the actual amount of extracted matter in a dose was recorded (Plate-). The application of dose was carried out by residual film method (Busvine, 1971). A general concentration for each of the extracts was selected as 10 mg/2ml as the stock dose for surface film application to make other successive doses by serial dilution to give 4160, 3640, 3120, 2600, 2080 ,1560, 1040 , 520 ,260 ,130 and 65 µg/cm^2 concentrations.

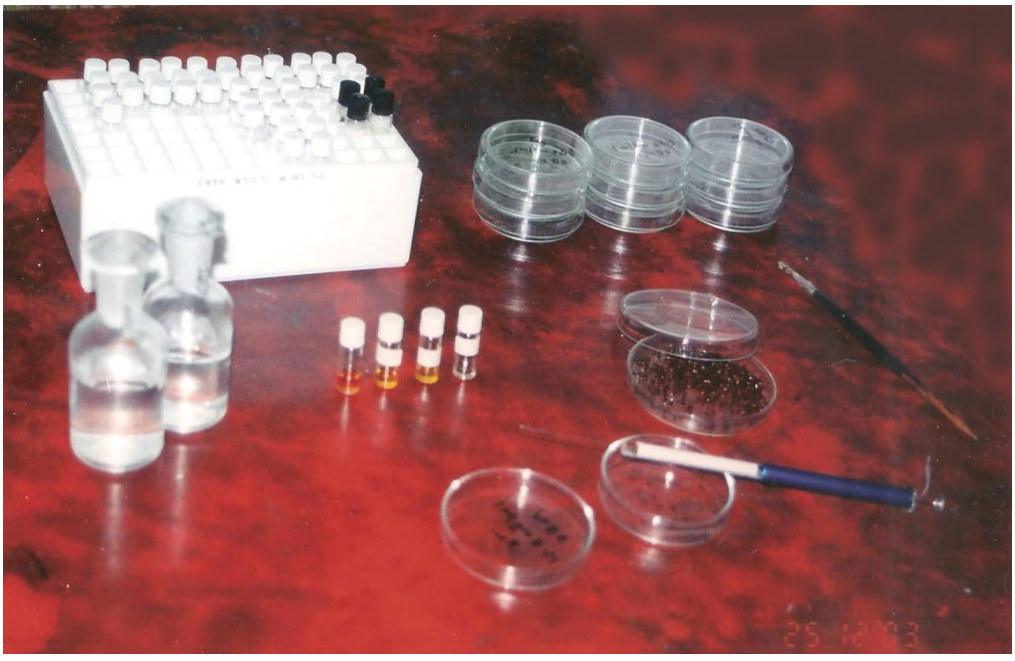


Plate12: Preparation of doses for surface film test

3.4.1.2. Application of doses on insects:

To conduct surface film activity test 70 mm Petri dishes were taken for all the doses and for their replications. One ml of each of the doses were poured into the lower part of the Petri dish and allowed them to dry out. Being volatile the solvent was evaporated out within a few minutes. Ten insects were released in each of the treated Petri dish. A control experiment by applying the only solvent into the Petri dish was also set at the same time under the same conditions (Plate 13).

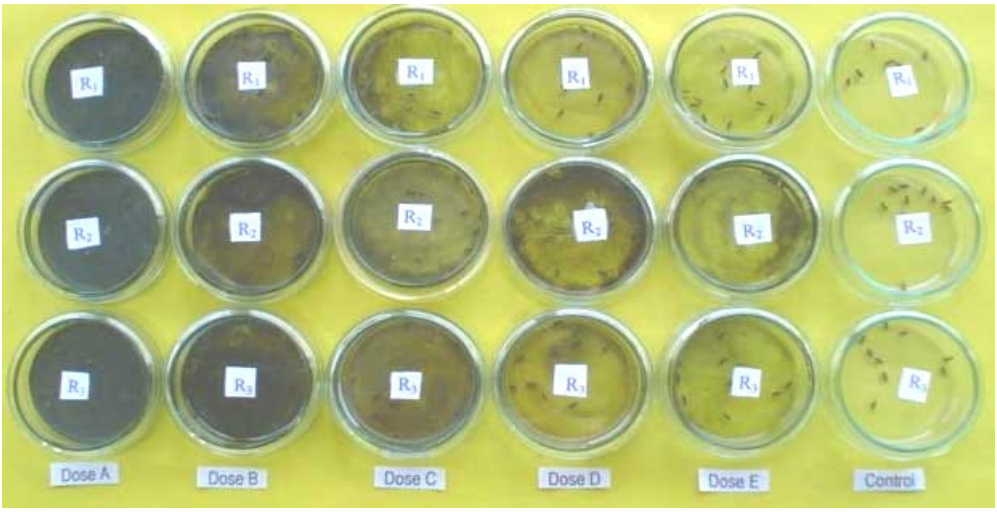


Plate 13: Bioassay using the plant extracts against *T. castaneum* adults by surface film method.

3.4.1.3. Reading and analysis of data for insecticidal activity:

The experimental petri dishes were placed in a secured place at room temperature. The whole experiment was observed from time to time and the mortality was counted after every 24h, 48h, 72h and 96h and the data was recorded. A simple microscope was used to check each and every beetle by tracing natural movement of its organs. In some cases hot needle was taken closer to the

bodies (without movement) to confirm death. Attention was also paid to recovery of the insects if occurred.

The mortality recorded was corrected by the Abbott's (1925) formula in the following manner:

$$P_r = \frac{P_o - P_c}{100 - P_c} \times 100$$

Where,

P_r = Corrected mortality (%)

P_o = Observed mortality (%)

P_c = Control mortality (%), sometimes called natural mortality (%).

Then mortality percentages were subjected to statistical analysis according to Finney (1947) and Busvine (1971) by using software developed in the Department of Agricultural Environmental Science, University of Newcastle upon Tyne, U.K. The dose-mortality relationship was expressed as a median lethal dose (LD_{50}).

3.4.1.4. Preparation and application of doses for larvicidal assay:

Effect of toxicity of A*. indica* extracts against larvae of *T. castaneum* was assessed by observing their chronic action on any stage of the beetles' life span. The selected food medium (1 g of whole wheat flour in a vial for each dose) was treated with different doses of the extracts of *A*. *indica* to release selected number of larvae in each of the units. Changes in all the developmental

stages were observed from time to time including mortality. Any sort of abnormality in their growth was observed.

3.4.1.5. Reading and analysis of data for larvicidal activity:

The vials containing the larvae along with their treated food were kept on the culture rack without light, humidity and temperature control. The recorded mortality was analyzed according to Finney (1947) and Busvine (1971) as it was done in the previous experiments with adult beetles.

3.5. Preparation of doses for the repellency test:

A general concentration for each of the extracts was selected as stock dose for surface film application to make other successive doses from it by serial dilution to give 1888 to as less as 15 µg/cm^2 (1888, 944, 472, 236, 118, 59, 30 and 15 µg/cm^2 concentration for leaf, root bark, root wood, flower, seed, stem bark and stem wood extracts of different parts of *A. indica*.

3.5.1. Application of doses for repellency of insects:

The repellency test used was adopted from the method (No. 3) of McDonald *et al*., (1970) with some modifications by Talukder and Howse (1993, 1994). Half filter paper discs (Whatman No. 40, 9 cm diam.) were prepared and selected doses of all the $CHCl_3$ extract separately applied onto each of the half-disc and allowed to dry out as exposed in the air for 10-15 minutes. Each treated half-disc was then attached lengthwise, edge-to-edge, to a control half-disc with adhesive tape and placed in a Petri dish (9 cm diam.), the inner surface of which was smeared with flu on to prevent insects escaping. The orientation of the same was changed in the replica

to avoid the effects of any external directional stimulus affecting the distribution of the test insects. Ten adult insects were released in the middle of each filter-paper circle (Plate- 12). Each concentration was tested five times. Insects that settled on each half of the filter paper discs were counted after 1 h and then at hourly intervals for 5 h. No significant difference was detected between the repellency of only solvent impregnated and untreated filter papers in tests designed to check for any possible influence of $CHCl_3$. The average of the counts was converted to percentage repellency (*PR*) using the formula of Talukder and Howse (1993, 1995):

$$PR = 2(C - 50),$$

Where, C is the percentage of insects on the untreated half of the disc. Positive values expressed repellency and negative values for attractant activity.

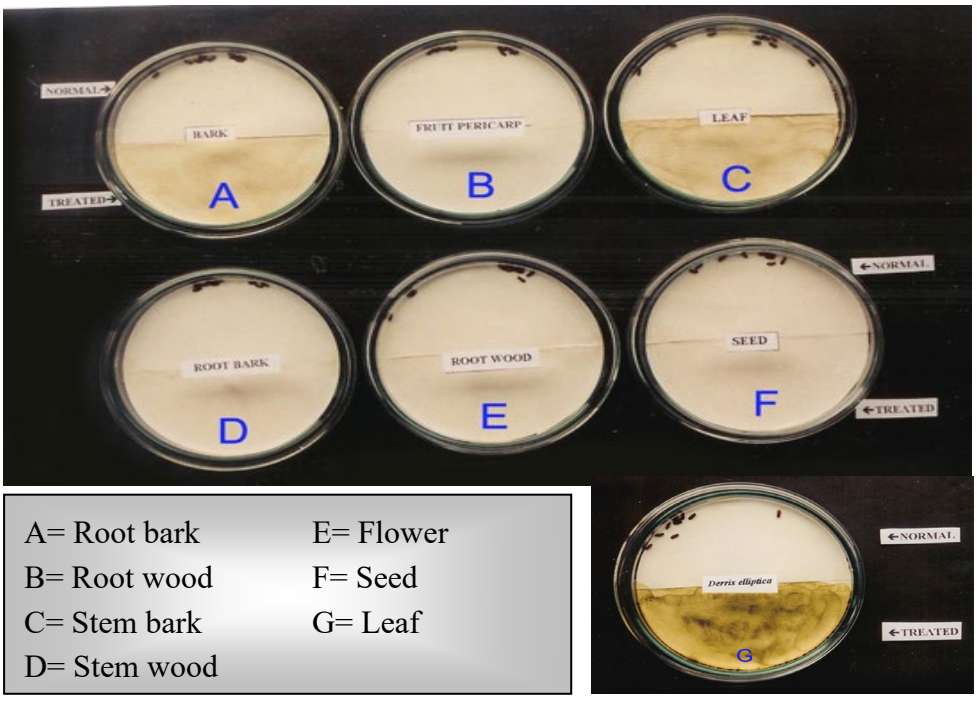


Plate14: Experiment for repellency test

Reading and analysis of data for repellency:

Repellency was observed for one-hour interval and up to five successive hours of exposure, just by counting the number if insects in the treated and non-treated part of the filter paper spread on the floor of the 90 mm Petri dish. The values in the recorded data were then calculated for percent repellency, which was again developed by arcsin transformation for the calculation of ANOVA.

Culture of *A. salina* for cytotoxicity test:

Artemia is a genus of aquatic crustaceans known as brine shrimp. *Artemia*, the only genus in the family Artemiidae, has changed little externally since the Triassic period. The historical record of the existence of *Artemia* dates back to 982 from Urmia Lake, Iran, although the first unambiguous record is the report and drawings made by Schlösser in 1756 of animals from Lymington, England (Alireza Asem, 2008). *Artemia* populations are found worldwide in inland saltwater lakes, but not in oceans. *Artemia* are able to avoid cohabiting with most types of predators, such as fish, by their ability to live in waters of very high salinity (up to 25%) (Martin Daintith,1996).

The ability of the *Artemia* to produce dormant eggs, known as cysts, has led to extensive use of *Artemia* in aquaculture. The cysts may be stored for long periods and hatched on demand to provide a convenient form of live feed for larval fish and crustaceans (Martin Daintith, 1996). Nauplii of the brine shrimp *Artemia* constitute the most widely used food item, and over 2000

tones of dry *Artemia* cysts are marketed worldwide annually. In addition, the resilience of *Artemia* makes them ideal animals for running biological toxicity assays and it is now one of the standard organisms for testing the toxicity of chemicals. A breed of *Artemia* is sold as a novelty gift under the marketing name *Sea-Monkeys*

Test materials used in this experiment:

(i) *A. salina* Leach (brine shrimp eggs or cysts)

(ii) Sea salt (non-ionized NaCl)

(iii) Small tank with perforated dividing dam to hatch the shrimp

(iv) Lamp to attract the nauplii

(v) Pipette (1 ml and 5 ml)

(vi) Micropipette (10-200 µl adjustable)

(vii) Test tubes (5 ml)

(viii) Magnifying glass

(ix) Different sizes beaker

Since the lethality test involves the culture of brine shrimp nauplii, i.e., the nauplii should be grown in the seawater, while the seawater contains 3.8% of sodium chloride. Accordingly 3.8% sodium chloride solution was made by dissolving sodium chloride (38 gm) in distilled water (1000 ml) and was filtered off. The P^H of the brine water thus prepared was maintained between 8 and 9 using $NaHCO_3$.

Brine water was taken in a small tank and shrimp eggs (1.5 gm/L) were added to one side of the perforated divided tank with constant oxygen supply. Constant temperature (37ºC) and sufficient light were maintained to give the sufficient aeration. After 48 hours, matured shrimp as nauplii (larvae) was collected and used for the experiment.

Preparation and application of doses on *A. salina:*

Chloroform and methanol extracts of the *A. indica* samples were applied against the brine shrimp nauplii. For the leaves, root bark, root wood, stem bark, stem wood and flowers samples 4mg were initially dissolved in 200 µl of pure dimethylsulfoxide (DMSO) to make them hydrophilic before adding 19.98 ml of water to get a concentration of 100 ppm for each of the samples separately which were used as stock solutions for all the extracts and from these concentrations other successive doses were prepared separately for each of the extracts through serial dilution method. Then a series of following concentrations made from the stock solutions were 200,100, 50, 25, 12.5, 6.25 and 3.125 ppm for all the extracts separately. In case of the seed extract 2 mg was initially dissolved in 100 µl of pure dimethyl sulfoxide (DMSO) to make it hydrophilic before adding 19.98 ml of water to get a concentration of 50 ppm which was used as the stock solution for the seed extract. Then a series of following concentrations was made from the stock solution as 100, 50, 25, 12.5, 6.25, 3.125, 1.563 and 0.7765 ppm for the seed extract. Brine shrimp eggs were hatched in simulated seawater to get nauplii. Test samples were prepared by the addition of calculated amount of DMSO

(dimethylsulfoxide) for obtaining desired concentration of the test sample. The nauplii were counted by visual inspection and were taken in vials containing 5 ml of seawater. Then samples of different concentrations were added to the pre-marked vials with the help of a micropipette. The vials were left for 24 hours and then the nauplii were counted again to find out the cytotoxicity of the test agents and compared to the results with positive control.

Reading and analysis of data for cytotoxicity:

The test tubes containing the nauplii along with the treated brine water were kept on a rack near the window in the laboratory. The recorded mortality was analyzed according to Finney (1947) and Busvine (1971) as it was done in the previous phase of experiments with both adults and larvae.

Preparation and application of doses for antimicrobial assays:

Preparation and application of doses on bacteria:

Preparation of the test plates:

The test plates were prepared according to the following procedure:

The nutrient agar medium prepared previously was poured in 15 ml quantity in each of the clean test tubes and plugged with cotton pads.

(i) The test tubes and the petri dishes were sterilized in an autoclave at 121°C and 15 lbs/sq. inch pressure for 15 minutes

and were transferred into a laminar air flow unit and then allowed to cool down to 45°C to 50°C.

(ii) The test organisms were transferred from the fresh subculture to the test tubes containing 15 ml autoclaved medium with the help of an incubating loop in an aseptic condition. Then the test tubes were shaken by rotation to get a uniformed suspension of the organism.

(iii) The bacterial suspensions were immediately transferred to the sterile Petri dishes in an aseptic area. The Petri dishes were rotated several times, first clock wise and then anticlockwise to assure homogenous distribution of the test organisms. The media were poured into Petri dishes in such a way that it could give a uniform depth of approximately 4 mm.

(Iv) Finally, when the medium was cooled down to room temperature in a laminar air flow unit, it was stored in a refrigerator (at 4°C).

Preparation of the discs treated with the test samples:

For the preparation of the discs containing chloroform and methanol extracts the following procedures were utilized. Three types of discs were prepared for antimicrobial screening. These are as follows:

(a) Sample discs- Sterilized filter paper discs having 5 mm in diameter (BBL, Cocksville U.S.A.) were prepared with the help of a punch machine and were taken in a blank Petri dish. Sample solution of desired concentration (10 μg/disc) was applied on the discs with the help of a micropipette in an aseptic condition. These

discs were left for a few minutes for complete removal of the solvent.

(b) Standard discs- These are used to compare the antibacterial activity of the test materials. In the present study, discs containing (30 μg/disc) of the antibiotic Amoxicillin were used as standard discs for the comparison with the extract treated ones.

(c) Control/blank discs- These were used as negative controls to ensure that the residual solvents on the filter paper were not active themselves. These were prepared in the previous manner applying only solvent to the discs and were used to examine the effect of the solvents used.

Preparation of test samples:

In both cases, the doses were prepared 50 and 200 µg/discs separately of chloroform and methanol extracts and the standard Amoxicillin was used 30 µg/disc.

Placement of the discs and incubation:

(i) By means of a pair of sterile forceps, the dried crude extract discs and standard disc were placed gently on the solidified agar plates seeded with the test organisms to ensure contact with the medium.

(ii) The plates were then kept in a refrigerator at 4°C for 24 hours in order to provide sufficient time to diffuse the antibiotics into the medium.

(iii) Finally, the plates were incubated at 37.5°C for 24 hours in an incubator.

Precaution:

The discs were placed in such a way that they were not closer than 15 mm to the edge of the plate and were placed apart enough to prevent overlapping of the zones of inhibition.

Measurement of the zones of inhibition:

After incubation, the antibacterial activities of the test samples were determined by measuring the diameter of inhibitory zones in term of mm with a transparent scale.

Preparation and application of doses on fungi:

Preparation of the test plates:

The test plates were prepared according to the following procedure:

(i) About 10 ml of distilled water was poured in several clean test tubes and plugged with cotton pads.

(ii) The test tubes, Petri dishes, glass rods, cotton pads and the medium were sterilized by autoclave and then transferred to the laminar air flow cabinet.

(iii) About 6 ml of the medium was poured carefully into the medium sized Petri dishes separately and were rotated several times, first clockwise and then anticlockwise to assure homogenous thickness of the medium and allowed to cool down and solidity at about 30°C.

(iv) The test tubes containing distilled water were inoculated with fresh culture of the test fungi and were shaken gently to form a uniformed suspension of the

organism because of their high prevalence of sporulation process.

(v) Separate piece of cotton were immerged in the test tubes with the help of individual glass rods and gently rubbed the medium. The pieces of cotton were then discarded.

(vi) Finally, the plates were stored in a refrigerator (4°C) overnight.

Preparation of the discs containing test samples:

(a)**Sample discs-** Sterilized filter paper discs (5 mm diam.) were taken by the forceps in the plates. Crude extracts of chloroform (50 µg and 200 µg) were applied on separate discs with the help of micropipettes in an aseptic condition. These discs were left for a few minutes for complete removal of the solvent.

(b)**Standard discs-** These were used to compare the antibacterial activity of the test material. In the present study, ready-made Nystatin 50 µg/disc was used as standard disc for comparison with the efficacy of the test extracts.

(c)**Control/blank discs-** These were used as negative controls to ensure that the residual solvent on the filter paper were not active themselves. These were prepared in the previous manner applying only solvent to the discs and were used to examine the effect of the solvents used.

Determination of Minimum Inhibitory Concentrations (MIC) for the antibacterial agents:

Minimum Inhibitory Concentration (MIC) may be defined as the lowest concentration of an antimicrobial drug to inhibit the growth of the test organism. There are two methods of experiments to determine the MIC values are as follows:

i) Serial tube dilution technique or turbid metric assay (Reiner, 1982).

ii) Paper disc plate technique or agar diffusion assay (Bauer *et. al.*, 1966).

Here 'Serial tube dilution technique' was followed using nutrient broth medium to determine the MIC values of chloroform extracts against the following 3 gram positive and 2 gram negative pathogenic bacteria.

Gram positive bacteria:

(a) *Streptococcus-β-haemolyticus*

(b) *Bacillus cereus*

(c) *Bacillus megaterium*

Gram negative bacteria:

(c) *Shigella dysenteriae*

(d) *Salmonella typhi*

Preparation of inoculum:

Fresh cultures of the test organisms were grown at 37.5°C for two days on the nutrient agar medium. Bacterial suspensions were

then prepared in sterile nutrient broth medium in such a manner that the suspension contains 107 CFU/ml. These suspensions were used as inoculate.

Preparation of the sample solution:

The stock solution was prepared by dissolving 1.024 mg of crude extracts in 2ml of DMSO. Thus the solution with a concentration of 1.024 mg/ml was obtained.

Procedure of serial tube dilution technique:

(i) Twelve (12) autoclaved test tubes were taken, nine of which marked as 1, 2, 3, 4, 5, 6, 7, 8 & 9 and the rest were assigned as Cm = (medium), Cs = (medium + compound) and Ci = (medium + inoculum).

(ii) One ml of sterile nutrient broth medium was added to each of the 12 test tubes.

(iii) One ml of the sample solution was added to the first test tube and mixed well.

(iv) One ml content from the 1^{st} test tube was transferred by the sterile pipette to the 2^{nd} test tube and mixed uniformly. Then 1 ml of this mixture was transferred to the 3^{rd} test tube. This process of serial dilution was continued up to the 9^{th} test tube.

(v) One drop (10 µl) of properly diluted inoculum was added to each of the 9 test tubes and mixed well.

(vi) For the control test tube, 1 ml of the sample solution was added and mixed well, while 1 ml of this mixed

content was discarded. This was to check the clarity of the medium in presence of diluted solution of the compound.

(vii) 10 µl of the inoculum was added to the control test tube Ci to observe the growth of the organism in the medium used.

(viii) The control test tube Cm, containing medium only was used to confirm the sterility of the medium.

(ix) All the test tubes were incubated at 37.5°C for 18-24 hours. The MIC is the lowest drug concentration at which there is no growth of the organism.

Phytochemical screening:

The phytochemical analysis of all the extracts of *A. indica* were subjected to qualitative phytochemical screening to identify the presence of alkaloids, flavonoids, carbohydrates, gum, reducing sugars, saponins, steroids, tannins and terpenoids using the established methods as described by Harborne, 1998 and Sazada *et al.*, 2009. For alkaloids, saponins, tannins, glycosides, anthraquinones, terpenes, and flavonoids was carried out using the methods described by (Harborne 1973, 1993; Sofowara, 1993; Trease and Evans, 1989). The different parts of *A. indica* were extracted with the required solvent and necessary reagent added to the right quantity of the extract. All observations were recorded. Briefly, Alkaloids, flavonoids and tannins were respectively tested with Wagner reagent, concentrated HCl and 0.1% ferric chloride. Molish reagents and concentrated sulfuric acid for gum, sulfuric

acid for steroids, α-napthol and sulfuric acid for reducing sugar and chloroform and concentrated HCl for terpenoids were used as reagents. Saponin was identified based on the ability to produce suds.

Test for tannins:

About 0.01g of the crude extract was boiled in 20 ml of water in a boiling tube. Few drops of 0.1 % of $FeCl_3$ were added. Formation of brownish green or a blue black colouration indicated the presence of tannins.

Test for saponins:

About 0.01 g of the crude extract was boiled in 20 ml of distilled water in a water bath. Then it was mixed with 5 ml of distilled water and it was shaken well. Stable persistent froth indicated the presence of saponins.

Test for phlobatanins:

About 0.01 g of the crude extract was boiled with 1 % aqueous hydrochloric acid. A deposition of a red precipitate indicated the presence of phlobatanins.

Test for flavanoids:

About 0.01 g of the crude extract was dissolved in 2 ml of ethanol solvent. Con. HCl and Mg turnings were added. Formation of yellow colour indicated the presence of flavanoids.

Test for steroids:

About 0.01 g of the crude extract was dissolved in 2 ml of ethanol solvent. 2 ml of acetic anhydride and 2 ml of con H_2SO4 were

added. A colour change from violet to blue or green indicated the presence of steroid

Test for cardiac glycosides:

0.01g of the crude extract was dissolved in 2 ml of ethanol and then 2 ml of glacial acetic acid containing one drop of ferric chloride solution was added. This was underplayed with 1 ml of con. H_2SO4. Appearance of brown ring indicated the presence of the cardiac glycosides.

Test for alkaloids:

About 0.01 g of crude extract was dissolved in ethanol and it was divided into two parts. Few drops of Mayer's reagent were added to one part. A creamy white precipitate indicated the presence of alkaloids. Few drops of Wagner's reagent were added to other part. A red-brown colour precipitate indicated the presence of alkaloids.

Chapter-4

Isolation, Purification and Evaluation of the Purified Compounds

A large number of chemicals have been developed for the control of plant diseases. But due to overgrowing awareness of the hazardous side effects of these chemicals, more and more emphasis is being given to the use of biocontrol agents. Now major challenge is felt in the field of plant pathology to introduce some ecofriendly and safe alternative control strategies for agriculture, which led researchers to turn their attention to plants and microorganisms as sources of biocontrol agents. For the separation of pure substances the availability and choice of chromatographic techniques are essential for the successful program involving in the investigation of biologically active plant constituents. The aim is to have maximum yield with minimum effort (to reduce the time and cost of the separation procedure). Preparative separation techniques can be tedious and time consuming, especially when complex mixtures, such as crude plant extracts have to be dissolved.

Over the passed decade or so, several new techniques have been introduced, leading to acceleration and simplification of different separation problems (Hostettmann *et al.,* 1998; Marston and Hostettmann, 1991; Hostettmann *et al.,* 1991). However, there is no universal technique capable of solving every isolation problem. All methods have advantages and limitations so much, so that the best results are often obtained by a combination of two or more of

these. Among the most important preparative separation techniques employed in the isolation and purification of plant constituents thin layer chromatography and open column chromatography have been used simultaneously in this investigation since we do not have other equipment available in our laboratory. Thin layer chromatography was used to select the slurry or the solvent system for the successful run of the open column chromatography.

Isolation techniques in general:

Preparative TLC plates for the separation of compounds:

To select the solvent system for the run of an open column separation was made on the preparative thin layer chromatographic plates. Aluminum backed pre-coated preparative thin layer chromatographic (TLC) plates (20 × 20cm) with silica gel GF_{254} with 0.5mm thickness and active in the usual manner (Merck, Germany) were used in this regard. The sample was applied on the active plates with the help of a gradient micropipette as a narrow band at 1 cm above the lower edge of the plate to make sure that the sample was not washed away when the plates were placed inside the TLC chamber with the solvent system. The plates were then developed in the usual manner. A concentration of 10 mg/ml of the sample in the solvent of extraction offered 100 μg/spot when 10 μl for each of the samples spotted. The chromatograms then developed within a conventional camber (Camag) using the following solvent systems: Ethyl Acetate: n-

Hexane = 3: 1 and Chloroform: Ethyl Acetate = 3: 2 for the $CHCl_3$ extracts.

Revelation of compounds (spots) on TLC by reagent spray:

The Godin reagent (Godin, 1954) is the mixture of the equal volume of 1% ethanolic solution of vanillin and 3% aqueous solution of parchloric acid. After spraying the reagent on the dried TLC 10% ethanolic solution of H_2SO_4 is also applied in the same way before drying the plate at 100°C to reveal the compound spots in different colors. The properly developed plates were dried and viewed visually after Godin reagent spray. The developed chromatogram was examined carefully to find different bands on the basis of the difference in colors and concentration of substances in each of the bands, and thus several compounds were detected. The R_f values of the separated compounds were calculated on a developed chromatogram using the pre-established solvent system. The R_f values were calculated by the following formula.

$$R_f = \frac{\text{Distance traveled by the compound}}{\text{Distance traveled by the solvent}}$$

Plate 15: Compounds on point C the upper Brownish is the compound 1, and the lower Yellowish is the compound 2

Open column chromatography for the isolation of the compounds:

Of the methods in the solid phase category, open column chromatography is very popular and used extensively. It can include non-exchange resins, polymeric columns, gel-filtration and chromatography over silica gel or chemically modified silica gel. Open column chromatography has a high load capacity but the separation time is long and the resolution is respectively low.

The stationary phase for the open column chromatography was silica gel Si60 (60-170 mesh and 230-400 mesh) (Merck) and glass column of different size (32×2.5 cm, 25×2 cm, 25×1.5 cm, etc.) were used. Cotton pads washed with acetone, chloroform and Methanol were used at the base of the gel column. A similar cotton cloud was used at the top of the column (after application of the sample and the solvent) to protect destruction of the sample layer (Plate). Selected solvent systems were used as eluents and the elution rate was 1 ml/min.

For fractionation of the selected extracts with a view to isolate biologically active compounds they were subjected to biological assay. Repetition of the same assay all along the successive fractionations was required until the purification of the target compound(s), and thus a suitable bioassay technique was selected in this regard. Considering the bioactive potentials the leaf extract of *A. indica* was the target extract for activity guided fractionations; while 200 µg of these samples were used on the

fungus inoculated agar plates to detect their biological activity to trace the presence of the bioactive compound(s). A number of plant pathogenic fungi, *F. vasinfectum, A. fumigatus, A. flavus, A. niger and C. albicans* were used in this regard. Potato dextrose agar (PDA) medium was used to perform these antifungal activity tests through disc diffusion method. Clear zones were observed against a dark background that had been produced by the fungus itself.

Cylindrical columns made of glass; drown at one end to from a narrow tube. The lower constricted end of the column was fitted with a stop cock for controlling the flow of the effluent. The column was made by pouring down the slurry of the silica gel (70-230 mesh and 230-400 mesh) in the suitable solvent and allowing the silica gel to settle down. The pouring of slurry that was selected earlier by thin layer chromatography was continued until the column of desire height was obtained. The solvent layer should always be kept above the absorbent bed to avoid cracking of the column. At the end of preparation of the column a little amount of the slurry kept on the upper surface of the gel matrix for the convenience in application of the extract in dissolved state.

Plate16: Open column used in the experiment.

Gel filtration for purification of the isolated compounds:

Open columns are used to apply sephadex LH-20 (Pharmacia) for the chromatography of exclusion. The separation of the methanolic extracts done with MeOH (100%) as the eluent and for the $CHCl_3$ soluble samples $CHCl_3$ and MeOH (1:1) system were used. The eluent allowed about 0.5 ml/min.

Compounds targeted for isolation:

In the present study isolation of the pure compounds from the leaf of *A. indica* was done mainly by open column chromatography (OCC), while thin layer chromatography (TLC) was used as a

supporting tool. The selection of the test extracts for the isolation was done depending on their biological activities.

Isolation of compounds from the leaf of *A. indica:*

For the first fractionation LH_{20} (Pharmacia) was used as the stationary phase and $CHCl_3$ and MeOH (1:1) was the eluent on a glass column of 2.5 × 32 cm for 1g of the leaf extract. Elution time was adjusted to yield 1 ml/min. It gave 80 tubes, which were then spotted on TLC to run and reveal the compounds by reagent spray. Six fractions were made for tubes 1-10 (Fr. I), 11-24 (Fr. II), 25-35 (Fr. III), 36-50 (Fr. IV), 51-68 (Fr. V) and 69-80 (Fr. VI). Biological assay with fungi indicated Fr. III for the presence of bioactive components there in and it was then subjected to fractionation. Selecting a solvent system by TLC, a slurry of cyclohexane and acetone (3:1) was applied on a glass column of 2×25 cm which was packed with silica gel (70-230 mesh, 43 gm) (Sigma). The elution was kept similar to that of the previous one. This fractionation yielded 82 tubes and TLC was made for all of them to get six sub-fractions: tubes 1-12 (Sfr. I), 13-25 (Sfr. II), 26-40 (Sfr. III), 41-54 (Sfr. IV), 55-65 (Sfr. V) and 66-82 (Sfr. VI). Biological assay with the test fungi indicated Sfr. IV for the presence of bioactive components and that was then subjected to fractionation. Again selecting a solvent system by TLC a slurry of Ethyl acetate and n-Hexane (3:1) was applied on a glass column of 1.5 × 25 cm was packed with 25 gm silica gel (230-400 mesh, Sigma). The elution was kept similar to that of the previous one. The fraction yielded 70 tubes for 6 fractions for tubes 1-10 (Ssfr. I), 11-20 (Ssfr. II), 21-29 (Ssfr. III), 30-40 (Ssfr. IV), 41-55 (Ssfr. V)

and 56-70(Ssfr. VI). Biological assay of these fractions against the test fungi indicated Ssfr. II for the bioactive compound, which was a pure compound of 30 mg of off-white powder and was named Compound A_1(Fig.5).

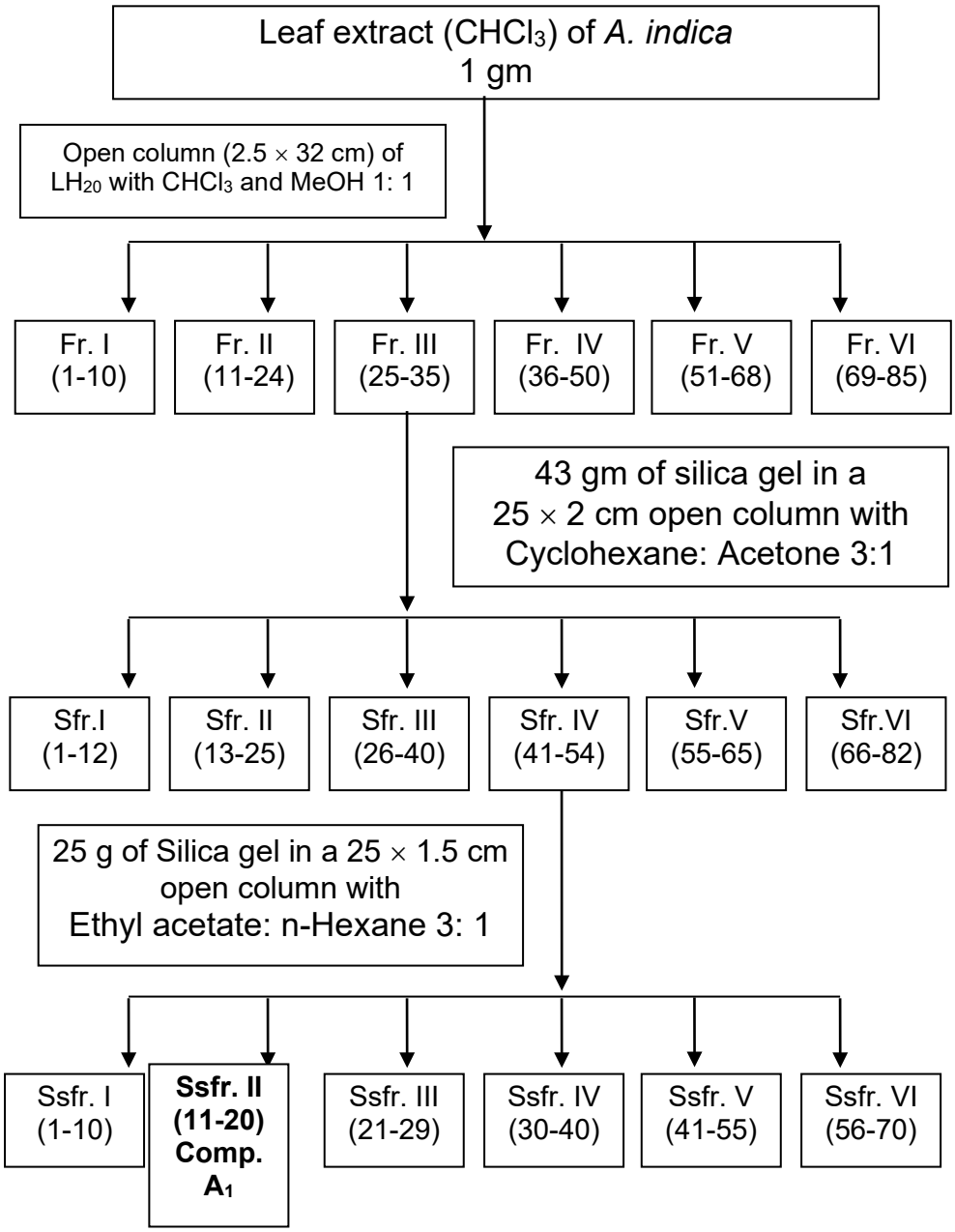


Fig.5: Isolation pathway of the compound A_1 from the leaf of *A. indica*

🗗 Isolation of compounds from the leaf of *A. indica:*

Sephadex LH_{20} (Pharmacia) was used as the stationary phase and $CHCl_3$ and MeOH (1:1) was the eluent on a glass column of 2.5 × 32 cm for 500 mg of the leaf extract. Elution time was adjusted to yield 1 ml/min. which yielded 75 tubes. The collections were then spotted on TLC to run with the solvent system n-Hexane: Ethyl acetate 5:1 and revealed by the Godin (Godin, 1954) reagent spray. Five fractions were made for tubes 1-13 (Fr. I), 14-30 (Fr. II), 31-45 (Fr. III), 46-60 (Fr. IV) and 61-75 (Fr. V). Biological assay with fungi indicated that the Fr. III contains bioactive components there in and it was then subjected to fractionation with a solvent system chloroform: Ethyl acetate, 5:1 glass column of 2 × 25 cm packed with silica gel (70-230 mesh, 40 gm, Sigma). The elution was kept similar to that of the previous amount. This fractionation yielded 75 tubes and TLC was made for them to get six sub fractions: 1-14 (Sfr. I), 15-30 (Sfr. II), 31-45 (Sfr. III), 46-55 (Sfr. IV), 56-70 (Sfr. V) and 71-75 (Sfr. VI). Again biological assay with the test fungi indicated that the Sfr. VI. Contains the presence of bioactive components and this was then subjected to fractionation selecting a solvent system by Chloroform: Ethyl acetate (3:2). A glass column of 1.5 × 25 cm was packed with 28 gm silica gel (230-400 mesh, Sigma). The elution was kept all along same as used in the previous one. This fractionation yielded 77 tubes for 6 fractions for tubes 1-12 (Ssfr. I), 13-25 (Ssfr. II), 26-38 (Ssfr. III), 39-55 (Ssfr. IV), 56-67 (Ssfr. V) and 68-77 (Ssfr. VI). Ssfr IV appeared to have a single compound and it was traced bioactive by antifungal activity test, while this purified compound was 26 mg in amount and was named Compound A_2 (Fig. 6).

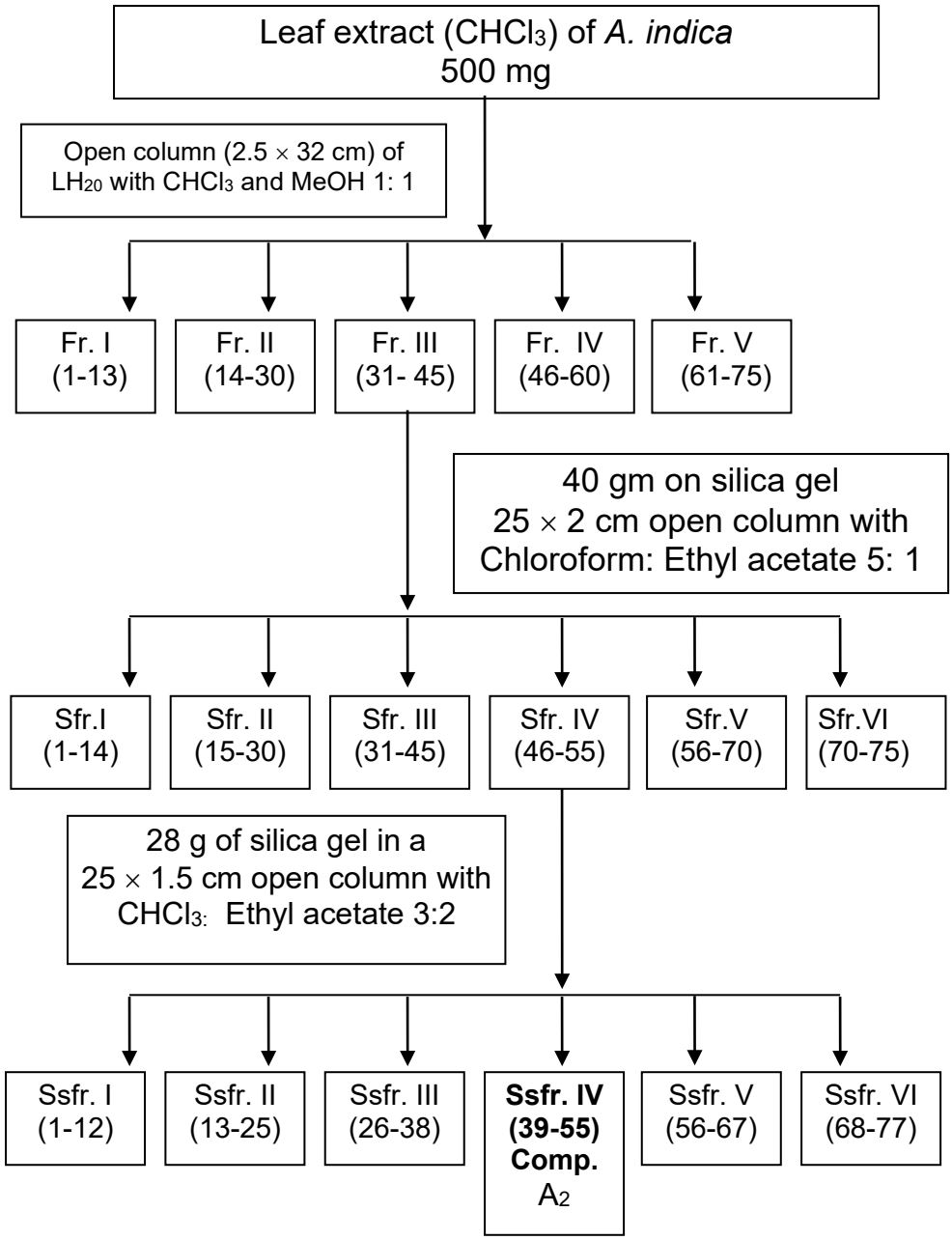


Fig.6: Isolation pathway of the compound A_2 from the leaf of *A. indica*

🗗 Physical remarks of the purified compounds:

The isolated compounds and there physical statures have been presented in (Table 48). Compound A_1 and A_2 were isolated from the leaf extracts of *A. indica*. However, all the two compounds were subjected to NMR analysis, as well as their biological activity tests have been carried out.

Table 5: Compounds purified from *A. indica* leaf extracts.

Solvent of extraction	Compound	Retention factor (R*f*)	Physical identity of the compounds	Coloration after Godin reagent spray
$CHCl_3$	A_1	0.32	White powder	Brownish
	A_2	0.53	Needle like	Gray

Characterization of the leaf compounds through analyses of NMR spectra:

Compound 1

A. Purity analysis of compound 1

The purities of compound 1 were detected by TLC. The results (Plate -) indicated compound 1 showed only one brownish spot along the chromatography plate. That is to say compound 1 should be a relative pure substance.

Structure Analysis of the Compound 1:

The hydrogen and carbon spectral analysis of the compound 1 was obtained by nuclear magnetic resonance spectroscopy. The hydrogen Spectrum analysis of the compound-1 (Table) showed that the high field below δ 4 was the saturated hydrogen spectrum. The methyl should be connected with oxygen, due to shield, at

lower field δ 3.85 (3H, s). The triple peaks at δ 0.88 (3H, t) can be thought as a terminal methyl that's connected with CH_2, which was considered a benchmark of hydrogen spectrum integral curve (Fig.7)

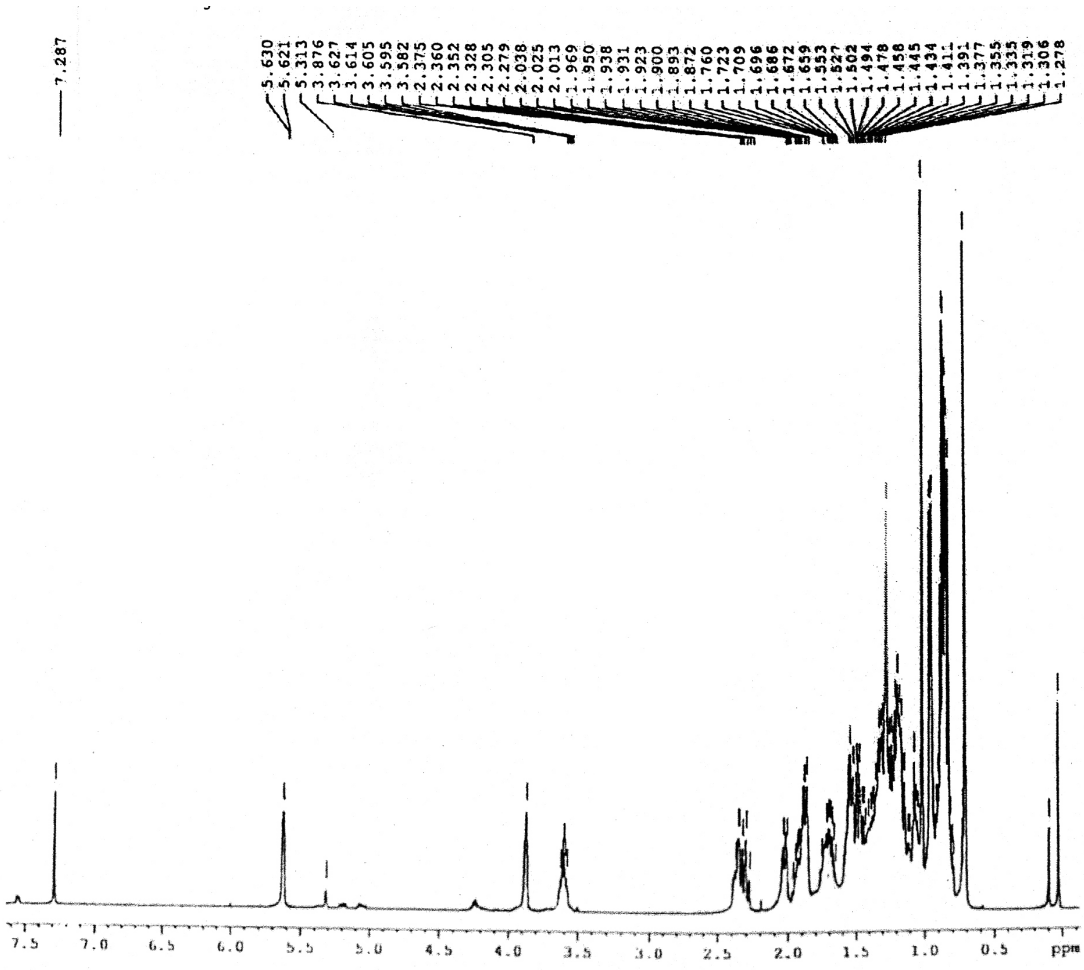


Fig. 7: ^1H NMR spectrum of compound A_1

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Creation Date/Timem: 5/10/05 15:11:10
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Scan 9-5:39. Entries=106.100% Int.=7526

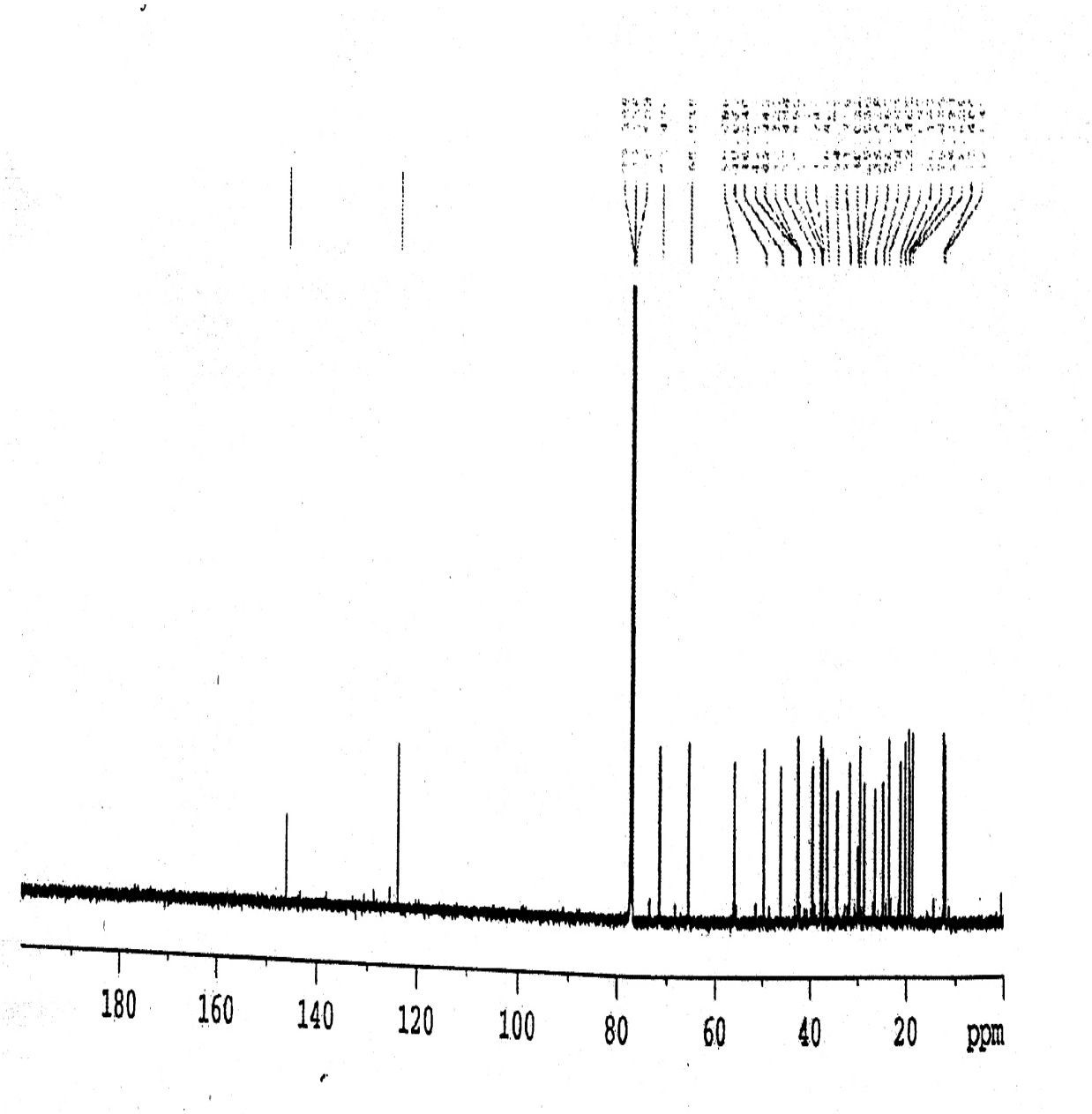


Fig. 8: ^{13}C- NMR spectrum of compound A_1

Table 6: 1H-NMR data of compound A_1

Position of Proton	δ value (J in Hz)
3	3.6 (1H, septet J= 6.5Hz)
8	5.63 (1H, d, J= 5.0Hz)
7	3.87 (1H,s)
18	0.60 (3H,s)
19	1.25 (3H,s)
21	0.99 (3H,s)
26	0.85 (3H,s)
27	0.84 (3H,s)
29	0.90 (3H,s)

Table 7. ^{13}C-NMR spectral data of compound A_1

Carbon Number	Chemical shift (δ Value in ppm)	Carbon Number	Chemical shift (δ Value in ppm)
C-1	37.3	C-16	28.3
C-2	31.7	C-17	56.1
C-3	72.7	C-18	12.0
C-4	42.3	C-19	19.4
C-5	140.8	C-20	36.1
C-6	121.7	C-21	19.3
C-7	31.9	C-22	34.0
C-8	31.9	C-23	29.2
C-9	50.1	C-24	50.1
C-10	36.3	C-25	26.1
C-11	21.1	C-26	19.0
C-12	39.8	C-27	19.4
C-13	42.3	C-28	23.1
C-14	56.8	C-29	11.9

In summary, from the above data have seen that the structure of the compound 1 is very similar with Quercetin. So the structure of the compound 1 was proposed as a derivative of Quercetin 3-ß-D-glucoside.

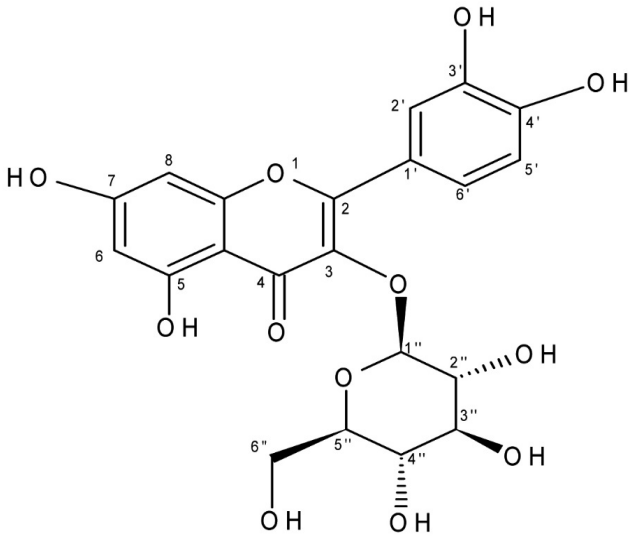


Fig.9: Quercetin 3-ß-D-glucoside.A_1

Compound 2:

A. Purity analysis of compound 2

The purities of compound 2 were detected by TLC. The results (Plate 17) indicated compound 2 showed only one gray spot along the chromatography plate. That is to say compound 2 should be a relative pure substance.

B. Structure Analysis of the Compound 2

Characterization of β-sitosterol (compound A_2)

The crystalline compound (m.p.132-134^{0}C) isolated from chloroform extract was found to be homogenous on TLC plates using different solvent systems and gave positive test for alcohol

and steroid with Salkowski and Liebermann-Burchard reactions and negative test for phenol with ferric chloride reagent and for alkaloid with Dragendorff's reagent. From the positive tests for steroid and alcohol given by the compound A_2, the compound was assumed to be sterol. The melting point 132-134^{0}C is in a good agreement with the melting point given for β-sitosterol in the literature (Directory of organic compound 1965). The final identity of the compound as β-sitosterol was confirmed by analysis of its ^{1}H-NMR spectral data (Figure). In its ^{1}H-NMR spectrum, the compound A_2 exhibited two tertiary methyl proton peaks at δ0.68 (3H, s, H-18) and δ0.85 (3H, s, H-19), three secondary methyl proton peaks at δ0.91 (3H, d, J = 6.42 Hz, H-26), δ 0.87 (3H, d, J = 6.42 Hz, H-27) and δ1.01, (3H, d, J = 6.5 Hz, H-21) and one primary, methyl proton peak at δ0.90 (3H, t, J = 6.5 Hz, H-29). The spectrum also exhibited a broad double at δ5.36 (1H, d, J = 4.6 Hz) attributed to be a double bonded proton typical for H-6 and multiple at δ3. 53 (1H, m) integrated for one proton which could be H-3 of a steroidal skeleton. Other signals appeared between δ0.9 ~ 2.4 were due to the methylene and methane protons. These assignments are in good agreement for the structure of β-sitosterol cited in the literature.

From the above ^{1}H-NMR and ^{13}C-NMR data, the compound A_2 is identified and confirmed as β-sitosterol. Though it is a known compound but is the first report of isolation from the leafs of *A. indica.*

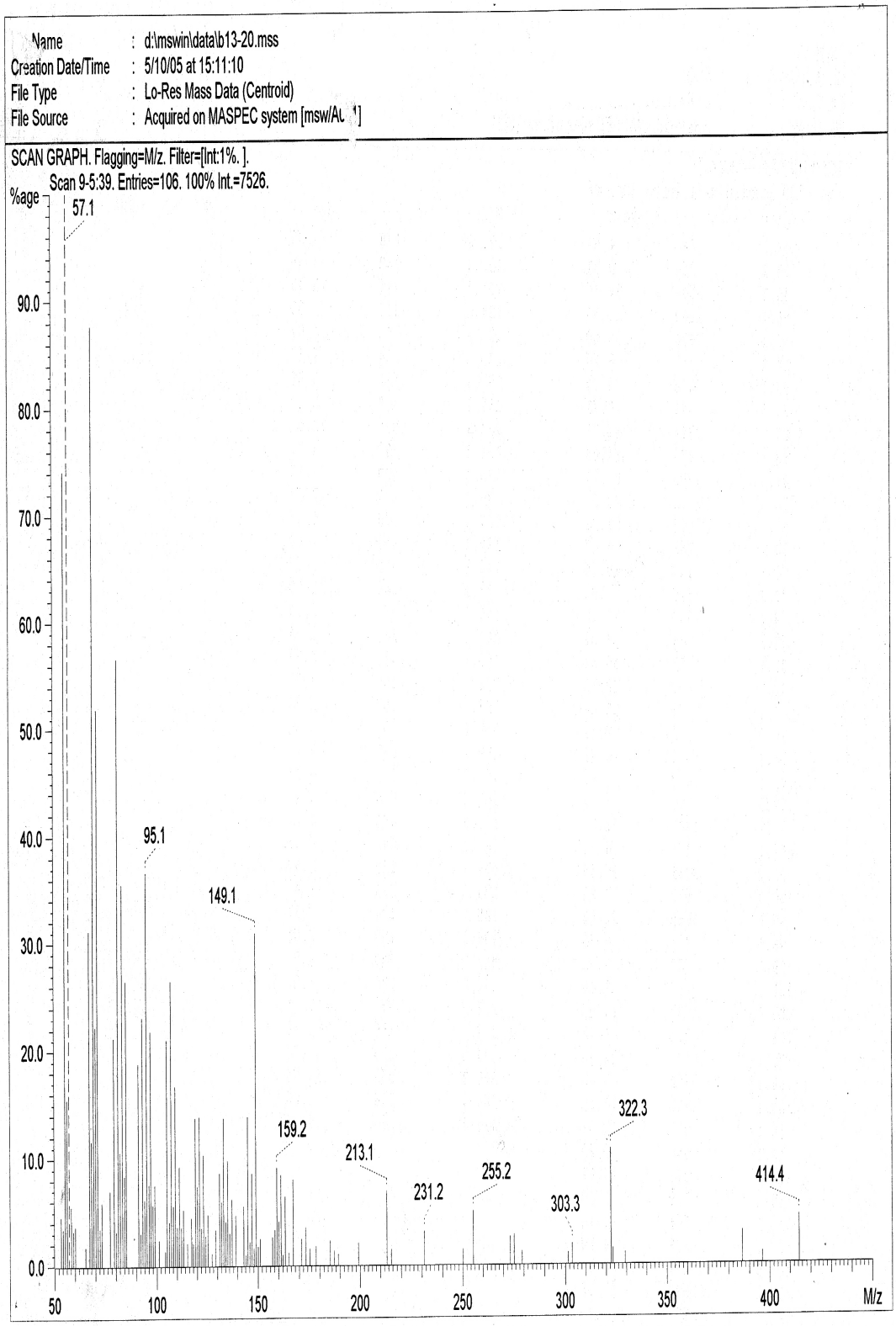


Figure- 10: ^{1}H NMR spectrum of compound (A_2)

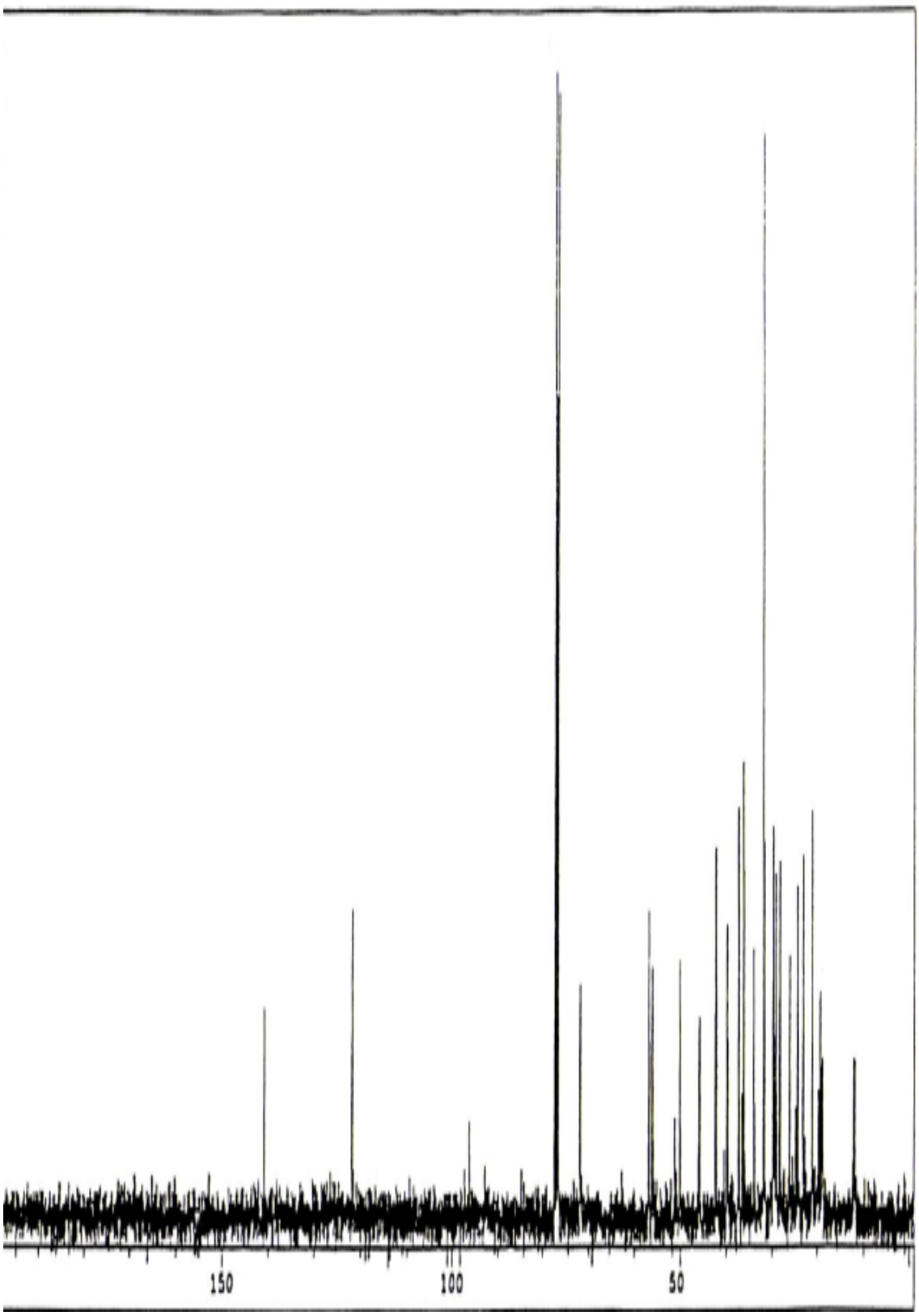


Figure-11: ^{13}C- NMR spectrum of compound (A_2)

Table 8. ^{1}H **-NMR spectral data of compound** A_2

Position of Proton	Chemical shift (δ Value in ppm, J in Hz)
H-3	3.53 (1H, m)
H-6	5.33-5.35 (1H, tdd, J = 4.26 & 1.0 Hz)
H-18	0.66 (3H, s, CH3)
H-19	1.23 (3H, s, CH3)
H-21	0.90 (3H, s, CH3)
H-26	0.84 (3H, s, CH3)
H-27	0.82 (3H, s, CH3)
H-29	0.80 (3H, s, CH3)

Table 9. ^{13}**C-NMR spectral data of compound** A_2

Carbon No.	**Chemical shift (δ value in ppm)**	**Carbon No.**	**Chemical shift (δ value in ppm)**
C-1	37.3	C-16	28.3
C-2	31.7	C-17	56.1
C-3	72.7	C-18	12.0
C-4	42.3	C-19	19.4
C-5	140.8	C-20	36.1
C-6	121.7	C-21	19.3
C-7	31.9	C-22	34.0
C-8	31.9	C-23	29.2
C-9	50.1	C-24	50.1
C-10	36.3	C-25	26.1
C-11	21.1	C-26	19.0
C-12	39.8	C-27	19.4
C-13	42.3	C-28	23.1
C-14	56.8	C-29	11.9
C-15	24.3	-	-

Chemical properties:

It gave color on TLC with vanillin sulfuric acid spray reagent on heating the plates at 110^0C until coloration took place.It gave red ring in Salkowski reaction and in Liebermann Burchard reaction which indicate the presence of steroid.

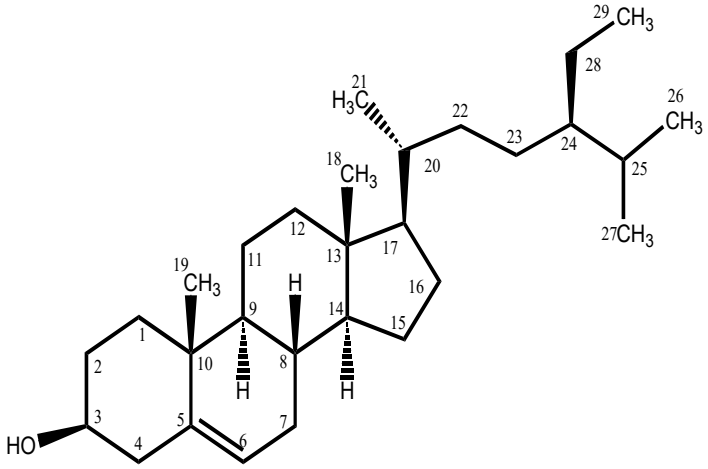


Fig: 12: ß-sitosterol

Bioactivity of the purified leaf compounds of *A. indica:*

Biological activities of the purified compounds were assessed through antimicrobial activity tests. Doing bioassay with the purified compounds has been a major target in this investigation, however the amount of the purified compounds being insufficient after their use in the NMR it was impossible to carry out their biological assays mentioned in the objectives. The two purified compounds A_1 and A_2 showed good antimicrobial activities against the selected pathogenic bacteria and fungi, while the intensity of activity is mentioned in the previous chapter.

Chapter-5

Results

Bioactivity of the crude extracts:

Insecticidal activity against *Tribolium castaneum* adults:

All the chloroform and methanol extracts of the stem bark ,stem wood, flower, leaves, root bark, root wood, and seeds of *A. indica* were tested against *T. castaneum* adults through residual film assay at doses of 7077.141, 3538.571, 2830.856, 2653.928, 2123.142, 1769.286, 1415.428, 884.643, 707.714 and 353.857 µg/cm^2 on the surface of the Petri dishes, where the test insects were released to observe mortality or any sort of behavioral changes due to the action of the extracts compared to their controls. The results have been presented in Appendix Tables (----) for the mortality recorded. To trace acute toxicity (if exists) an observation of mortality was made after 24h after application of the doses, however usual observations were made after 48h, 72h and 96h of exposures.

The data was subjected to probit analysis and the LD_{50} values were shown in Tables (----). The seed extract was found to offer the highest mortality of the beetles, while the LD_{50} values were 52900.91, 3035.954, 323.7229 and 107.0412µg/cm^2 for the chloroform extracts; 153634.400, 9099.510, 1174.775 and 222.3965 µg/cm^2 for the methanol extracts after 24h, 48h, 72h and 96h of exposures respectively. In case of stem bark extracts the

LD_{50} values were 250054.200, 10277.430, 1744.959 and 244.4488 µg/cm^2 for the chloroform extracts; 32645.240, 217184.200, 8083.415 and 457.6257 µg/cm^2 for the methanol extracts after 24h, 48h, 72h and 96h of exposures respectively.

Observation after 24h assured acute toxicity positively, however, the LD_{50} value was simply larger. Depending on toxicity the stem wood extract gives LD_{50} values 65273.730, 11569.140, 3162.549 and 177.580 µg/cm^2 for the chloroform extracts; 25037.140, 3493.696, 826.733 and 492.0781 µg/cm^2 for the methanol extracts after 24h, 48h, 72h and 96h of exposures respectively. For the flower extracts the LD_{50} values were 540902.700, 6911.297, 1523.749 and 259.3435 µg/cm^2 for the chloroform extracts; 449643.300, 254095.200, 4813.655 and 752.3578 µg/cm^2 for the methanol extracts after 24h, 48h, 72h and 96h of exposures respectively. In case of leaf extracts, the LD_{50} values were 36852.300, 2059.099, 856.4559 and 113.3073 µg/cm^2 for the chloroform extracts; 29009.690, 14538.500, 1528.169 and 447.2792 µg/cm^2 for the methanol after 24h, 48h, 72h and 96h of exposures respectively; which was just followed by the root bark extract to give the LD_{50} values 70982.010, 65832.731, 4261.189 and 480.3277 µg/cm^2 for the chloroform extracts; 240501.900, 15825.490, 2975.506 and 1011.733 µg/cm^2 for the methanol extracts after 24h, 48h, 72h and 96h of exposures respectively. The root wood extracts give LD_{50} values 23620.850, 1215.395, 408.6851 and 192.5573 µg/cm^2 for the chloroform extracts and 14412.800, 6449.728, 1146.651 and 418.4427 µg/cm^2 for the

methanol extracts after 24h, 48h, 72h and 96h of exposures respectively.

According to the intensity of activity observed through mortality of the adult beetles the potentiality of the stem bark,stem wood, flower, leaves, root bark, root wood, and seed extracts could be arranged in a descending order of chloroform extracts are as follows: seed> leaf> stem wood> root wood> stem bark> flower> root bark and for the methanol extracts the results could be arranged according to their potentiality seed > root wood >leaf > stem bark > stem wood > flower>root bark extracts.

The overall assessment of toxicity of *A. indica* extracts are very much promising and their efficacy on stored grain pests might have future to be used as a control agent or tool. It may open its possibility as a control agent for the insect pests as well.

Table10: LD_{50}, regression equation, $\chi 2$ values and 95% confidence limits of flower and leaf extracts of *A. indica* against *T. castaneum* after 24, 48, 72 and 96h of treatment.

Test extract		Time exposed	LD_{50} value µg/cm^2	95% Conf. Limits		Regression equation	χ^2 Value (df)
				Lower limit	Upper limit		
Flower	Chloroform	24h	540902.70	3.37315	8.674E+10	Y = 2.603 +0.418X	0.1691(3)
		48 h	6911.297	530.8342	89983.02	Y=3.185+ 0.473X	2.133E-02(3)
		72 h	1523.749	519.1728	4472.133	Y=3.547+ 0.457X	0.148(3)
		96h	259.3435	28.99703	2319.515	Y =3.864+ 0.471X	2.661E-02(3)
	Methanol	24 h	449643.30	32.5642	3.452E+06	Y=3.593+0.249X	6.375E-02(3)
		48h	254095.20	38.5011	1.677E+09	Y =2.181+ 0.522X	0.254(3)
		72 h	4813.655	432.4115	53586.13	Y =3.475+0.414X	6.361E-02(3)
		96h	752.3578	127.7164	4432.026	Y =4.134+0.301X	0.184(3)
Leaf	Chloroform	24h	36852.30	385.3198	35245.79	Y =2.458+0.557X	5.935E-02(3)
		48 h	2059.099	1108.395	3825.246	Y =1.890+ 0.939X	0.6018476(3)
		72 h	856.4559	459.4218	1596.608	Y =2.614+ 0.813X	0.4344726(3)
		96h	113.3073	6.037319	2126.529	Y =3.926+ 0.523X	0.3142581(3)
	Methanol	24h	29009.69	2.899E-02	2.903E+14	Y =2.609+ 0.370X	9.324E-02(3)
		48 h	14538.50	399.2724	5293.83	Y =3.022+ 0.475X	0.229532(3)
		72 h	1528.169	698.0223 t	3345.597	Y =2.997+ 0.629X	0.28724(3)
		96h	447.2792	127.1399	1573.53	Y=3.426+ 0.594X	0.1975682(3)

Table11: LD_{50}, regression equation, $\chi 2$ values and 95% confidence limits of root bark and root wood extracts of *A. indica* against *T. castaneum* after 24, 48, 72 and 96h of treatment.

Test extract		Time exposed	LD_{50} value µg/cm²	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Root bark	Chloroform	24h	70982.01	298.7637	1.686E+07	Y =2.012+ 0.616X	0.2835674(3)
		48 h	65832.731	6.50053	9.320E+07	Y =3.156+ 0.383X	0.3384838(3)
		72 h	4261.189	618.3115	29366.64	Y =3.244+ 0.484X	0.6619892(3)
		96h	480.3277	98.38282	2345.072	Y =3.795+ 0.449X	0.8699672(3)
	Methanol	24h	240501.900	40.22166 t	1.4380E+09	Y =2.228+ 0.515X	8.164E-02(3)
		48 h	15825.49	980.0414	255546.50	Y =2.147+ 0.679X	0.2034314(3)
		72 h	2975.506	1124.038	7876.648	Y =2.339+ 0.766X	0.5990839(3)
		96h	1011.733	445.3582	2298.383	Y =3.268+ 0.576X	1.968E-02(3)
Root wood	Chloroform	24h	23620.85	172.8482	32279.46	Y =3.199+ 0.412X	6.884E-02(3)
		48 h	1215.395	547.9739	2695.722	Y =3.194+ 0.586X	0.4004541(3)
		72 h	408.6851	224.8994	742.000	Y =1.504+ 1.339X	2.070941(3)
		96h	192.5573	74.1513	500.036	Y =1.901+ 1.357X	1.020651(3)
	Methanol	24h	14412.80	3.770E-02	5.509E+13	Y=3.135+ 0.303X	0.1079621(3)
		48 h	6449.728	969.3614	4291.3E+13	Y =2.603+ 0.630X	0.3760615(3)
		72 h	1146.651	589.842	2229.087	Y =2.854+ 0.701X	1.936571(3)
		96h	418.4427	132.3817	1322.646	Y =3.224+ 0.678X	0.668766(3)

Table12: LD_{50}, regression equation, $\chi 2$ values and 95% confidence limits of seed and stem bark extracts of *A. indica* against *T. castaneum* after 24, 48, 72 and 96h of treatment.

Test extract		Time exposed	LD_{50} value µg/cm²	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Seed	Chloroform	24h	52900.91	99.69377	2.807E+07	Y =2.982+ 0.427X	0.1880722(3)
		48 h	3035.954	607.318	15176.57	Y =3.375+ 0.467X	3.478E-02(3)
		72 h	323.7229	80.55176	1300.982	Y=3.352+ 0.656 X	0.4663468(3)
		96h	107.0412	10.58678	1082.276	Y =3.589+ 0.695X	0.3290119(3)
	Methanol	24h	153634.400	46.15917	5.1134E+08	Y =2.658+ 0.452X	0.1452589(3)
		48 h	9099.51	675.918	122501.800	Y=2.859+0.541X	1.526E-02(3)
		72 h	1174.775	306.4728	4503.159	Y =3.942+ 0.344X	4.725E-02(3)
		96h	222.3965	16.25487	3042.791	Y =3.998+ 0.427X	0.3080139(3)
Stem bark	Chloroform	24h	250054.200	111.5826	5.6036E+08	Y =2.409+ 0.480X	0.1759168(3)
		48 h	10277.430	881.0349	119888.200	Y =3.281+ 0.428X	3.414E-02(3)
		72 h	1744.959	915.3052	3326.627	Y =2.342+ 0.820X	0.5358367(3)
		96h	244.4488	11.30526	5285.62	Y =3.688+0.549X	0.6432109(3)
	Methanol	24h	32645.240	0.1466581	7.2666E+13	Y =2.586+ 0.371X	9.118E-02(3)
		48 h	217184.200	19.47927	2.4214E+09	Y =3.101+ 0.356X	6.792E-02(3)
		72 h	8083.415	1045.788	62480.82	Y =3.263+ 0.444X	0.4730903(3)
		96h	457.6257	5.592158	37449.120	Y =4.255+ 0.280X	2.562E-02(3)

Table13: LD_{50}, regression equation, $\chi 2$ values and 95% confidence limits of stem wood extracts of *A. indica* against *T. castaneum* after 24, 48, 72 and 96h of treatment.

Test extract		Time exposed	LD_{50} value µg/cm²	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Stem wood	Chloroform	24h	65273.730	1.224E-03	3.479E+16	Y =3.075+ 0.283X	9.210E-02(3)
		48 h	11569.140	1927.985	69422.270	Y =2.394+ 0.641X	0.829187(3)
		72 h	3162.549	1491.555	6705.562	Y =2.728+ 0.649X	0.1924589(3)
		96h	177.580	0.905008	34844.66	Y =4.205+ 0.353X	0.6384363(3)
	Methanol	24h	25037.140	1119.11	5601.41	Y =2.631+ 0.539X	0.1649764(3)
		48 h	3493.696	1511.623	8074.704	Y =2.841+ 0.609X	0.1863411(3)
		72 h	826.733	388.0266	1761.442	Y =1.518+1.194X	1.239829(3)
		96h	492.0781	210.316	1151.319	Y =0.547+ 1.654X	0.1123829(3)

Larvicidal activity against *T. castaneum:*

All the chloroform and methanol extracts of the flower, leaves, root bark, root wood, seed, stem bark and stem wood of *A. indica* have been applied against the larvae of *T. castaneum* for the detection of their biological activity (including lethality, prolongation of larval instars, causing deformity in body, abnormality in any of the biological parameters). In the pilot experiment all extracts showed promising results and for this reason several doses of 12-,6-,3-,1.50 and 0.75 mg/g for flower, leaves, root bark, root wood, seed, stem bark and stem wood extracts were established with three replications for the final experiment on the dose-mortality assay, where the test insects were released into treated food medium to observe mortality or any sort of abnormality due to efficacy of the extracts compared to the controls. The observations were made by

24h, 48h and 72h of interval. Mortality of the larvae and abnormality in changing instars, as well as differences in size were observed. In the developmental stages, in some cases, heterogeneity in the development of larvae was traced carefully. Some of the pupa became black or dark brown in color, also an abnormal golden pupa was found. The larvae that survived for long in the larval stage were in shrunk form and were not easily moving. Number of dead individuals decreased drastically in 72h. The results have been presented in Appendix Tables (---------) for the mortality recorded. According to the intensity of activity against the 1st instar larvae of chloroform extract the result could be arranged in the following order: seed> stem bark > root wood > stem wood> flower> root bark> leaf. In case of methanol extracts, the results were as follows: stem wood> root bark> seed> root wood> flower> stem bark> leaf after 72h respectively. For 2nd instars, the results were leaf > flower> root wood> stem bark> root bark > seed > stem wood for chloroform extract and for the methanol extracts the results were as follows: leaf > flower> root wood> root bark > stem wood > seed > stem bark after 72 hours respectively. In case of 3rd instars larvae the results were stem bark > root wood> stem wood > seed > flower> root bark> leaf for chloroform extract and for the methanol extracts the results were stem bark > root wood> seed >stem wood > flower> root bark> leaf after 72 hours respectively. For the 4th instars larvae against the chloroform extracts the results were as follows: root wood > root bark> seed >flower > leaf > stem wood >stem bark and for the methanol extracts the results were flower > root bark> root wood > seed > leaf > stem wood >stem

bark after 72 hours respectively. The LD_{50} values of the different parts of *A. indica* extract against the larvae of different solvents at the 1st instar were 18.76248, 12.66460, 7.438530, 33.052883, 22.274063,12.768684, 30.355246, 22.274066, 11.172065, 46.507186, 24.175765, 19.809616, 19.093132, 13.631894, 8.348996,18.185246, 15.502789, 8.883758, 7.996566, 5.584668, 4.546655, 21.271215, 20.779286, 9.409836, 12.31076, 9.237628, 3.456354, 20.126657, 15.169220, 9.425903, 22.27406, 6.308404, 4.505973, 514.51894, 15.38510, 14.3854, 12.856148, 9.666136, 5.660547, 14.018987, 10.705654 and 7.37698 mg/g respectively after 24h, 48h and 72h of exposures respectively against the chloroform and methanol extracts; for 2nd instar larvae the LD_{50} values were13.35709, 10.98855, 6.275992, 15.60706, 14.00541, 7.962568, 15.92349, 11.64476, 6.501557, 17.00807, 12.57389, 6.453151, 297.56647, 273.12738, 89.27206, 682.47485, 491.35428, 302.24278, 383.16239, 225.27249, 55.42979, 682.47486, 273.12739, 97.59105, 466.30250, 139.47975, 90.13719, 463.53029, 460.63568, 454.04937, 682.47487, 302.24279, 75.59956, 1390.6339, 851.94329, 466.30258, 682.47486, 302.24279, 281.53658, 2687.0685, 491.35426 and 302.24278 mg/g respectively after 24h, 48h and 72h of exposures respectively against the chloroform and methanol extracts. For 3rd instar larvae the LD_{50} values were 466.30259, 136.03048, 39.61924, 467.3026, 383.16238, 157.16654, 292.24750, 203.90819, 139.47976, 466.30200, 251.58975, 211.48936, 383.16238, 109.50198, 76.55148, 470.24668, 228.37415, 203.90817, 14.29405, 9.299592,

5.512042, 21.72716, 9.99665, 6.242771, 22.48768, 17.39089, 6.043458, 31.68779, 22.07263, 8.984072, 15.92349, 8.690649, 5.929032, 18.76248, 12.97696, 6.881171, 15.92349, 11.93817, 5.384638, 28.84459, 22.48768 and 9.688208 mg/g respectively after 24h, 48h and 72h of exposures respectively against the chloroform and methanol extracts. In case of the 4th instar larvae the LD_{50} values were 16.95141, 10.34685, 6.239028, 17.00807, 9.199899, 5.222975, 14.29405, 11.43628, 6.448529, 46.11358, 18.76248, 11.93817, 14.29405, 9.903644, 5.051757, 27.74103, 19.53158, 6.882175, 14.73041, 9.670546, 5.413227, 31.54786, 16.51215, 9.569064, 14.42565, 11.76264, 5.67895, 25.53965, 13.470, 9.66295, 846.20699, 444.64856, 32.924669, 7266.5765, 697.7399, 68.312635, 7450.9956, 192.0266, 39.52767, 43235.54, 105.2427 and 63.79169 mg/g respectively after 24h, 48h and 72h of exposures respectively against the chloroform and methanol extracts. The results have been shown in Tables (-------). The larval mortality showed a possibility of raising toxicity by the magnification of the amount of ingestion of the treated food. The number of death has been increased just proportional to that of the age of the larvae, which indicates the increase in volume of food intake by the larvae as well.

Table14: Larvicidal effect of Flower, leaf, root bark and root wood extract of *A. indica* against *T. castaneum* larva (1st instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Flower	chloroform	24	18.76248	6.555629	53.69897	Y= 2.725+1.002 X	0.73854(3)
		48	12.66460	4.819793	33.27781	Y=3.167+0.872X	1.95926(3)
		72	7.438530	3.518182	15.70000	Y=3.415 +0.848X	1.05836(3)
	methanol	24	33.052883	7.830971	139.5093	Y=2.523+0.984X	1.57912(3)
		48	22.274063	5.81500	85.32300	Y=3.028+0.840X	1.16333(3)
		72	12.768684	4.45800	36.56800	Y=3.314+0.801X	1.78937(3)
leaf	chloroform	24	30.355246	8.967541	102.7525	Y=2.068 +1.182X	0.152573(3)
		48	22.274066	5.814798	85.32262	Y=3.028+0.840X	1.163330(3)
		72	11.172065	4.385187	28.46285	Y=3.278+0.8412X	1.297401(3)
	methanol	24	46.507186	7.134844	303.1486	Y=2.682+0.869X	0.364152(3)
		48	24.175765	6.122689	95.45926	Y=2.962+0.855X	0.449951(3)
		72	19.809616	4.792457	81.88301	Y=3.305+0.738X	0.693476(3)
Rroot bark	chloroform	24	19.093132	8.733655	41.74056	Y=1.625 +1.480X	3.253348(3)
		48	13.631894	6.482191	28.66753	Y=2.416+ 1.211X	3.967207(3)
		72	8.348996	4.142447	16.82719	Y= 3.148 +0.965X	1.529515(3)
	methanol	24	18.185246	7.342437	45.03997	Y =2.360+ 1.168X	1.006329(3)
		48	15.502789	5.389241	44.59557	Y=3.057+ 0.889X	0.524635(3)
		72	8.883758	4.117786	19.16593	Y=3.232+0.908X	0.891757(3)
R root wood	chloroform	24	7.996566	3.984326	8.723152	Y=2.397+1.470X	2.29703 (3)
		48	5.584668	3.313658	7.343968	Y=2.295+1.628X	0.82403(3)
		72	4.546655	5.013438	5.013438	Y=2.594+1.547X	0.68745(3)
	methanol	24	21.271215	6.821656	66.32765	Y=2.706 +0.986X	1.504621(3)
		48	20.779286	5.764886	74.898	Y=3.035+0.848X	2.856905(3)
		72	9.409836	4.200247	21.08091	Y =3.243+ 0.890X	1.282427(3)

Table15: Larvicidal effect of seed, stem bark and stem wood extract of *A. indica* against *T. castaneum* larva (1st instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
seed	chloroform	24	12.31076	7.287057	20.79762	Y = 1.504+ 1.673X	0.50407(3)
		48	9.237628	5.537157	15.41111	Y = 2.230+ 1.410X	2.12349(3)
		72	3.456354	1.912658	3.434968	Y=2.534 +1.751X	0.34166 (3)
	methanol	24	20.126657	7.647386	52.97000	Y = 2.297+1.174X	2.122684(3)
		48	15.169220	5.534841	41.57396	Y =2.986+ 0.924 X	2.00734 (3)
		72	9.425903	4.545616	19.54580	Y=3.052+ 0.987X	2.514602(3)
Stem bark	chloroform	24	22.27406	5.814798	85.32262	Y =3.028+0.840X	1.16333(3)
		48	6.308404	3.404555	11.68903	Y=3.291+0.949X	1.98589(3)
		72	4.505973	2.441486	8.316161	Y =3.632+0.828X	1.15289(3)
	methanol	24	514.51894	5.650212	37.30826	Y=2.911+0.967X	1.13181(3)
		48	15.38510	4.101972	57.704260	Y=3.475+0.697X	0.25507(3)
		72	14.3854	2.875615	12.85992	Y = 3.634+ 0.767X	8.7E-02(3)
stem wood	chloroform	24	12.856148	0.603954	25.80911	Y=2.356+1.2539X	0.597335(3)
		48	9.666136	4.651048	20.08884	Y =3.0159+0.100X	1.401576(3)
		72	5.660547	3.05997	10.47127	Y=3.418 +0.903X	3.060968(3)
	methanol	24	14.018987	4.202658	12.137465	Y=2.636 +1.597X	0.64269 (3)
		48	10.705654	2.973654	6.709896	Y=3.728 +1. 658X	0.164369(3)
		72	7.37698	5.56070	13.96809	Y=3.699+0.293X	4.09607(3)

Table16: Larvicidal effect of Flower, leaf, root bark and root wood extract of *A. indica* against *T. castaneum* larva (2nd instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	χ^2 Value (df)
				Lower limit	Upper limit		
Flower	chloroform	24	13.35709	6.91369	25.80559	Y=2.080 + 1.375X	1.912054(3)
		48	10.98855	5.726405	21.08623	Y= 2.520 +1.216X	3.177353(3)
		72	6.275992	4.460706	11.86809	Y=2.594 +1.293X	5.076075(3)
	methanol	24	15.60706	8.070065	30.18321	Y =1.608+1.548X	0.806482(3)
		48	14.00541	32.61558	86.01404	Y=2.712+1.066X	1.155865(3)
		72	7.962568	4.562784	13.89557	Y=2.741+ 1.189X	1.161095(3)
leaf	chloroform	24	15.92349	7.07521	35.83746	Y= 2.325+1.216X	0.5411558(3)
		48	11.64476	5.891689	23.01556	Y=2.512+1.205X	0.6711731(3)
		72	6.501557	3.898881	10.84164	Y=2.881 +1.170X	1.026411(3)
	methanol	24	17.00807	7.326695	39.48226	Y=2.265+1.227X	1.152259(3)
		48	12.57389	5.534317	28.56771	Y=2.830+1.039X	0.9562092(3)
		72	6.453151	3.973338	10.48065	Y= 2.778+1.230X	1.547712(3)
root bark	chloroform	24	297.56647	4.071503	21747.63	Y=3.654+0.545X	5.3442E-02(3)
		48	273.12738	2.754858	27078.91	Y=3.829+0.481X	4.5517E-02(3)
		72	89.27206	5.00674	1591.755	Y=3.947+0.541 X	0.408191(3)
	methanol	24	682.47485	2.314314	201256.9	Y=3.493+0.532X	1.3568E-02(3)
		48	491.35428	6125063	394165	Y=4.010+0.368X	1.2356E-02(3)
		72	302.24278	3.008951	30359.66	Y=3.774+0.495X	0.268800(3)
root wood	chloroform	24	383.16239	4.624563	31746.43	Y=3.465+0.594X	1.3624E-02(3)
		48	225.27249	5.222331	9717.434	Y=3.655+0.572X	1.6992E-02(3)
		72	55.42979	6.426384	478.1001	Y=3.953+0.601X	4.4575E-02(3)
	methanol	24	682.47486	2.314314	201256.9	Y=3.493+0.532X	1.3568E-02(3)
		48	273.12739	2.754858	27078.91	Y=3.829+0.481X	4.5517E-02(3)
		72	97.59105	4.343658	2192.625	Y=3.989+0.508X	7.1392E-02(3)

Table17: Larvicidal effect of seed, stem bark and stem wood extract of *A. indica* against *T. castaneum* larva (2nd instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	χ^2 Value (df)
				Lower limit	Upper limit		
seed	chloroform	24	466.30250	4.949317	43932.83	Y=3.192+0.678X	0.200049(3)
		48	139.47975	8.041382	2419.306	Y=3.543+0.680X	0.145455(3)
		72	90.13719	7.008613	1159.247	Y=3.760+0.635X	0.255499(3)
	methanol	24	463.53029	4.949317	43932.83	Y=3.191+ 0.678X	0.200049(3)
		48	460.63568	361576	4678510	Y=3.795+ 0.388X	0.269941(3)
		72	454.04937	0.3036665	678905.1	Y=4.143+0.323X	2.58E-02(3)
Stem bark	chloroform	24	682.47487	2.314314	201256.9	Y=3.493+0.532X	1.36E-02(3)
		48	302.24279	3.008951	30359.66	Y=3.774 +0.495X	0.2688(3)
		72	75.59956	3.743789	1526.607	Y=4.105+0.477X	9.06E-02(3)
	methanol	24	1390.6339	5.138E-02	3.7622E+07	Y=4.086+0.291X	0.114463(3)
		48	851.94329	0.7165104	1012975	Y=3.808 +0.407X	0.445677(3)
		72	466.30258	4.949317	43932.83	Y=3.191 +0.678X	0.200049(3)
stem wood	chloroform	24	682.47486	2.314314	201256.9	Y=3.493+0.532X	1.35E-02(3)
		48	302.24279	3.008951	30359.66	Y=3.77+0.494X	0.2688(3)
		72	281.53658	1.561521	50759.78	Y=3.990+0.413X	2.25E-02(3)
	methanol	24	2687.0685	0.3856401	1.8722E+07	Y=3.415+0.463X	0.106453(3)
		48	491.35426	6125063	394165	Y=4.009+0.368X	1.23E-02(3)
		72	302.24278	3.008951	30359.66	Y=3.77 +0.494X	0.2688(3)

Table18: Larvicidal effect of Flower, leaf, root bark and root wood extract of *A. indica* against *T. castaneum* larva (3rd instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Flower	chloroform	24	466.30259	4.949317	43932.830	Y=3.191+0.678X	0.2000492(3)
		48	136.03048	7.876344	2349.347	Y=3.579+0.667X	0.3559666(3)
		72	39.61924	8.229901	190.729	Y=3.831+0.732X	0.3208647(3)
	methanol	24	467.3026	4.949317	43932.83	Y=2.191+ 0.678X	0.245049 (3)
		48	383.16238	4.624563	31746.430	Y=3.465+0.594X	1.3624E-02(3)
		72	157.16654	4.730486	5221.689	Y=3.801+0.546X	0.1731033(3)
leaf	chloroform	24	292.24750	6.531942	13075.510	Y=3.252+0.709X	0.7538161(3)
		48	203.90819	3.651742	11385.940	Y=3.834+0.506X	7.0361E-02(3)
		72	139.47976	8.041382	2419.306	Y =3.544+0.679X	0.1454554(3)
	methanol	24	466.30200	4.949317	43932.830	Y=3.191+0.678X	0.2000492(3)
		48	251.58975	4.821364	13128.530	Y=3.620+0.575X	0.6678355(3)
		72	211.48936	6.239701	184.629	Y=2.664+1.005X	0.7569203(3)
root bark	chloroform	24	383.16238	4.624563	31746.430	Y=3.465+0.594X	1.3624E-02(3)
		48	109.50198	7.786949	1539.840	Y=3.670+ 0.652X	0.20367621(3)
		72	76.55148	5.194382	1128.167	Y=3.978+0 .543X	7.19599E-1(3)
	methanol	24	470.24668	2.269123	97452.560	Y=3.695+0.489X	0.2175069(3)
		48	228.37415	1.389216	37542.560	Y=4.070 +0.394X	0.11586(3)
		72	203.90817	4.949317	43932.830	Y=3.191+0.678X	0.2000492(3)
root wood	chloroform	24	14.29405	6.896696	29.62576	Y=2.242+1.280X	0.8471909(3)
		48	9.299592	4.82285	17.93181	Y = 2.843+ 1.096X	0.7402325
		72	5.512042	2.62264	9.48182	Y=2.997+1.149X	2.142655
	methanol	24	21.72716	7.46617	63.22772	Y=2.468+1.084X	0.5662975(3)
		48	9.99665	5.429061	18.40706	Y=2.542+1.230X	1.256035(3)
		72	6.242771	3.740099	10.42009	Y=2.947+1.144X	0.5566368(3)

Table19: Larvicidal effect of seed, stem bark and stem wood extract of *A. indica* against *T. castaneum* larva (3rd instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
seed	chloroform	24	22.48768	6.482878	78.00485	Y=2.830+0.923X	0.300242(3)
		48	17.39089	3.877473	78.00008	Y=3.540+0.652X	0.155325(3)
		72	6.043458	3.140479	11.62988	Y=3.442 +0.875X	3.793177(3)
	methanol	24	31.68779	36.278734	159.9233	Y=2.933+0.827X	0.501237(3)
		48	22.07263	4.80424 to	101.4106	Y=3.297+0.726X	1.855071(3)
		72	8.984072	3.855749	20.9333	Y=3.385+0.827X	8.984072(3)
Stem bark	chloroform	24	15.92349	7.07521	35.83746	Y=2.324+1.215X	0.541156(3)
		48	8.690649	4.280932	17.64275	Y=3.109+0.975X	0.738543(3)
		72	5.929032	3.238254	10.85567	Y=3.330+ 0.942X	2.20622 (3)
	methanol	24	18.76248	6.555629	53.69897	Y=2.724+1.001X	0.738543(3)
		48	12.97696	5.167974	32.5856	Y =3.030+0.932X	0.554608(3)
		72	6.881171	3.751891	12.62044	Y=3.147+1.008X	2.442274(3)
stem wood	chloroform	24	15.92349	7.07521	35.83746	Y=2.324+1.215X	0.541156(3)
		48	11.93817	4.826464	29.52884	Y=3.125+0.903X	1.41518(3)
		72	5.384638	3.163387	9.165595	Y=3.224+1.026X	2.364758(3)
	methanol	24	28.84459	3.748128	221.9804	Y=3.522 +0.601X	1.216453(3)
		48	22.48768	6.482878	78.00485	Y=2.830+0.923X	0.300243(3)
		72	9.688208	3.572338	26.2745	Y=3.552+0.729X	1.81846(3)

Table20: Larvicidal effect of Flower, leaf, root bark and root wood extract of *A. indica* against *T. castaneum* larva (4th instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
Flower	chloroform	24	16.95141	7.718403	37.22924	Y=2.008 +1.342X	0.3967962(3)
		48	10.34685	5.681353	18.84363	Y=2.416+1.283X	0.8912372(3)
		72	6.239028	4.096548	9.502015	Y=2.475+1.406X	1.143011(3)
	methanol	24	17.00807	7.326695	39.48226	Y=2.264+1.226X	1.152259(3)
		48	9.199899	5.266581	16.07079	Y=2.469 +1.288X	1.0194(3)
		72	5.222975	3.46128	7.881324	Y=2.729+1.322X	1.422516(3)
leaf	chloroform	24	14.29405	6.896696	29.62576	Y=2.241+1.280X	0.8471909(3)
		48	11.43628	5.401934	24.21142	Y=2.787 +1.075X	1.309761(3)
		72	6.448529	3.889667	10.69077	Y=2.869+1.178X	1.698786(3)
	methanol	24	46.11358	7.581674	280.474	Y=2.551+0.919X	0.8320189(3)
		48	18.76248	6.555629	53.69897	Y=2.724+1.001X	0.7385426(3)
		72	11.93817	4.826464	29.52884	Y=3.125+0.903X	1.41518(3)
root bark	chloroform	24	14.29405	6.896696	29.62576	Y=2.241+1.280X	0.8471909(3)
		48	9.903644	5.42245	18.08817	Y=2.526+1.240X	2.146212(3)
		72	5.051757	3.867013	9.470812	Y=2.703+1.289X	1.853952(3)
	methanol	24	27.74103	8.339966	92.27432	Y=2.283+1.112X	1.625594(3)
		48	19.53158	6.878268	55.46159	Y=2.616+1.041X	1.763982(3)
		72	6.882175	4.493163	10.54143	Y=2.344+1.446X	3.231087(3)
root wood	chloroform	24	14.73041	7.106956	30.53135	Y=2.164+1.308X	1.474346(3)
		48	9.670546	5.555684	16.83313	Y=2.350+1.335X	1.142679(3)
		72	5.413227	3.39452	8.632451	Y=2.953 +1.181X	0.7115708(3)
	methanol	24	31.54786	7.169451	138.8205	Y=2.696+0.922X	1.34199(3)
		48	16.51215	6.224204	43.80498	Y=2.761+1.010X	2.076032(3)
		72	9.569064	4.401974	20.80134	Y=3.152+0.933X	2.244703(3)

Table 21: Larvicidal effect of seed, stem bark and stem wood extract of *A. indica* against *T. castaneum* larva (4th instar).

Test extract		Time exposed (h)	LD_{50} value mg/gm	95% Conf. Limits		Regression equation	$\chi 2$ Value (df)
				Lower limit	Upper limit		
seed	chloroform	24	14.42565	8.743	23.800	Y =3.751+1.078X	0.150 (3)
		48	11.76264	6.659	20.775	Y =3.898+1.029X	0.594 (3)
		72	5.67895	2.591	12.443	Y = 4.218+1.037X	0.702 (3)
	methanol	24	25.53965	14.159	46.063	Y =3.646+0.962X	0.34400(3)
		48	13.470	7.182	25.264	Y =4.004+0.882X	0.234 (3)
		72	9.66295	7.665	22.856	Y =3.796+1.036X	0.694 (3)
Stem bark	chloroform	24	846.20699	107.583	6655.92	Y =3.027+0.674X	0.168 55(3)
		48	444.64856	93.614	2111.985	Y =3.175+0.690X	0.189 66(3)
		72	32.924669	18.637	58.163	Y = 3.840+0.765X	0.18966 (3)
	methanol	24	7266.5765	248.445	212533.80	Y =1.755+0.840X	0.647 (3)
		48	697.7399	208.878	2330.734	Y =2.645+0.828X	0.954 (3)
		72	68.312635	55.608	125.575	Y=3.569+0.745X	0.9510(3)
Stem wood	chloroform	24	7450.9956	2903859	1.912E+12	Y =3.238+0.300X	0.123 (3)
		48	192.0266	88.802	415.237	Y =3.351+0.722X	0.155 (3)
		72	39.52767	24.212	64.528	Y =3.429+0.984X	0.574 (3)
	methanol	24	43235.54	31.943	5.852E+07	Y = 3.073+0.416X	0.101 (3)
		48	105.2427	60.326	183.602	Y=3.348+0.817X	0.105 (3)
		72	63.79169	39.563	102.857	Y = 3.337+0.921X	1.228 (3)

Repellency against *T. castaneum* adults:

All the test extracts of leaves, flower, seed, root bark, root wood, stem bark and stem wood of *A. indica* collected in chloroform and methanol showed repellent activity against adult beetles of *T. castaneum* at dose levels 251.415, 125.707, 62.853, 31.427 and 15.713 µg/cm^2 on filter paper. The data was recorded with 1 hour interval for up to 5 hours of exposure and the percent repulsion data was then subjected to ANOVA after transforming into arcsine percentage values Appendix Tables (- to -); and the result has

been presented in Tables (---).The F values have been established were 51. 03662, 253.5068, 43.04438, 83.58911, 75.79346, 75.94017, 64.50964, 50.44838, 25.82928, 61.28114, 45.56164, 34.5519, 35.75216 and 21.5157 for the analysis between doses and 6.778143, 3.007724, 5.447409, 3.835164, 1.400522, 1.856993, 5.669432, 4.258362, 5.590989, 0.876118, 4.630108, 3.285364, 1.990562 and 0.989226 for the analysis between time interval for seed, stem wood, stem bark, root wood, root bark, flower and leaves of Chloroform and Methanol extracts respectively.

Among the tested $CHCl_3$ extracts all the rest offered repellency at 0.01% level of significance ($P<0.001$) According to the intensity of repellency the result could be arranged in a descending order: In case of chloroform extract stem bark >root wood> seed >flower> stem wood> leaf> root bark and for the methanol extracts seed> stem wood>stem bark> root bark> root wood> flower> leaf extract and in all the cases significant differences.

Table22: ANOVA results of repellency by *A. indica* extracts against *Tribolium castaneum* adult

Test material	Extract	Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*
Seed	Chloroform	Dose effect	13061.85	4	3265.462	51. 03662***	6.55E-09
		Time effect	1734.736	4	433.684	6.778143	0.00217
		Error	1023.723	16	63. 98272		
		Total	15820. 31	24			
Seed	methanol	Dose effect	21582.13	4	5395.533	253.5068***	3.01E-14
		Time effect	256.0606	4	64.01515	3.007724	0.04996
		Error	340.5373	16	21.28358		
		Total	22178.73	24			
Stem wood	Chloroform	Dose effect	9054.179	4	2263.545	43.04438**	2.27E-08
		Time effect	1145.836	4	286.4591	5.447409	0.005796
		Error	841.3807	16	52.58629		
		Total	11041.4	24			
Stem wood	methanol	Dose effect	18940.43	4	4735.108	83.58911***	1.63E-10
		Time effect	869.0087	4	217.2522	3.835164	0.022683
		Error	906.3588	16	56.64743		
		Total	20715.8	24			

(*** =Highly significant, ** = Significant).

Table23: ANOVA results of repellency by *A. indica* extracts against *Tribolium castaneum* adult

Test material	Extract	Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*
Stem bark	Chloroform	Dose effect	18683.48	4	4670.871	75.79346***	3.43E-10
		Time effect	345.2359	4	86.30897	1.400522	0.278477
		Error	986.0207	16	61.6263		
		Total	20014.74	24			
Stem bark	methanol	Dose effect	14036.64	4	3509.161	75.94017***	3.38E-10
		Time effect	343.2433	4	85.81081	1.856993	0.167359
		Error	739.3528	16	46.20955		
		Total	15119.24	24			
Root wood	Chloroform	Dose effect	17908.33	4	4477.083	64.50964***	1.15E-09
		Time effect	1573.874	4	393.4686	5.669432	0.00488
		Error	1110.428	16	69.40176		
		Total	20592.63	24			
Root wood	Methanol	Dose effect	14838.64	4	3709.66	50.44838***	7.14E-09
		Time effect	1252.534	4	313.1335	4.258362	0.015525
		Error	1176.541	16	73.53379		
		Total	17267.72	24			

(*** =Highly significant, ** = Significant).

Table24: ANOVA results of repellency by *A. indica* extracts against *Tribolium castaneum* adult

Test material	Extract	Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*
Root bark	Chloroform	Dose effect	12866.88	4	3216.721	25.82928**	8.29E-07
		Time effect	2785.157	4	696.2893	5.590989	0.005184
		Error	1992.605	16	124.5378		
		Total	17644.64	24			
Root bark	Methanol	Dose effect	17587.89	4	4396.973	61.28114***	1.69E-09
		Time effect	251.4488	4	62.8622	0.876118	0.49982
		Error	1148.013	16	71.75083		
		Total	18987.35	24			
Flower	Chloroform	Dose effect	12092.57	4	3023.141	45.56164**	1.5E-08
		Time effect	1228.882	4	307.2205	4.630108	0.011271
		Error	1061.644	16	66.35277		
		Total	14383.09	24			
Flower	Methanol	Dose effect	8799.327	4	2199.832	34.5519**	1.1E-07
		Time effect	836.6831	4	209.1708	3.285364	0.038049
		Error	1018.679	16	63.66746		
		Total	10654.69	24			

(*** =Highly significant, ** = Significant).

Table25: ANOVA results of repellency by *A. indica* extracts against *Tribolium castaneum* adult

Test material	Extract	Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*
Leaf	Chloroform	Dose effect	10685.99	4	2671.497	35.75216**	8.61E-08
		Time effect	594.9606	4	148.7401	1.990562	0.144549
		Error	1195.563	16	74.72267		
		Total	12476.51	24			
Leaf	Methanol	Dose effect	5586.538	4	1396.634	21.5157**	2.83E-06
		Time effect	256.8519	4	64.21298	0.989226	0.441442
		Error	1038.598	16	64.91235		
		Total	6881.987	24			

(*** =Highly significant, ** = Significant).

Cytotoxicity against *A. salina* nauplii:

The dose-mortality assay of *A. indica* extracts against the brine shrimp (*A. salina*) nauplii has been done through test tube treatment method for all the test extracts of leaves, flower, seed, root bark, root wood, stem bark and stem wood. Most of the test extracts showed remarkable dose-mortality effects against the 1 day nauplii of *A. salina* and the result has been presented in Appendix Tables (---). The seed extract was found to offer the highest mortality of the nauplii, while the LC_{50} values were 520.1635, 24.50645 and 5.942745 ppm for the chloroform extracts; 906.5301, 61.17362 and 18.24789ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively. The LC_{50} values for the stem bark extract were 1042.544, 196.883 and 24.53654 ppm for the chloroform extracts; 6030.069, 167.7432

and 34.2457 ppm for the methanol extracts. The LC_{50} values for the stem wood extract were 3711.381, 94.12271 and 45.16339 ppm for the chloroform extracts; 1641.063, 92.75699 and 48.30029 ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively .The LC_{50} values for the flower extract were 933.4176, 67.70986 and 26.04309 ppm for the chloroform extracts; 18450.49, 113.4081 and 24.50362 ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively. For the leaf extract the LC_{50} values were 3476.365, 101.4525 and 51.38413 ppm for the chloroform extracts; 9577.411, 455.9743 and 160.1078 ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively. The LC_{50} values for the root bark extracts were 987.7583, 28.04569 and 23.26771 ppm for the chloroform extracts; 1030.155, 57.71285 and 26.29665 ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively. The LC_{50} values for the root wood extracts were 838.2706, 36.47875 and 8.40184 ppm for the chloroform extracts; 5187.234, 82.83993 and 23.38707 ppm for the methanol extracts for 30 min, 24h and 48h of exposures respectively (Table---).

To consider acute toxicity (if exists) of the extracts a reading of data is made after 30 min. of exposure, and in this case the result was positive, while the LC_{50} values were comparatively larger. According to the intensity of activity the results of the extracts against the brine shrimp nauplii could be arranged in the following order: seed > root wood >root bark> stem bark> flower>stem wood> leaf for the chloroform extract and seed > root wood > flower> root bark> stem bark> stem wood> leaf for the methanol extracts.

Table26: Cytotoxicity through dose-mortality of *A. indica* flower, leaf and root bark extracts (Chloroform and Methanol) against *Artemia salina* nauplii after 30 min, 24h and 48h of treatment.

Test extract		Time exposed	LC_{50} value (ppm)	95% Conf. Limits		Regression equation	χ^2 Value (df)
				Lower limit	Upper limit		
Flower	Chloroform	30 min	933.4176	279.0945	3121.768	Y = 2.477+0.850X	1.48225(3)
		24h	67.70986	47.34112	96.84238	Y=3.550 + 0.792 X	0.5898743(3)
		48h	26.04309	19.34113	35.06738	Y= 3.445+1.099X	2.688568(3)
	Methanol	30 min	18450.49	140.349	24255.31	Y=3.196 + 0.423X	7.055E-03(3)
		24h	113.4081	75.2666	170.878	Y =3.241+0.856X	0.4319(3)
		48h	24.50362	16.54686	36.28648	Y =3.820+ 0.849X	0.136219(3)
Leaf	Chloroform	30 min	3476.365	284.25	42515.80	Y =2.905+ 0.592X	0.2450733(3)
		24h	101.4525	61.43791	167.5287	Y=3.680 +0.658X	0.2389603(3)
		48h	51.38413	36.80554	71.73725	Y =3.600+ 0.818X	0.4642525(3)
	Methanol	30 min	9577.411	202.3107	453396.30	Y =3.137+ 0.468X	0.2887631(3)
		24h	455.9743	182.6174	1138.515	Y =2.945+ 0.773X	0.6033821(3)
		48h	160.1078	94.85948	270.237	Y =3.237+0.799X	2.159527(3)
Root bark	Chloroform	30 min	987.7583	255.7718	3814.602	Y =2.772+ 0.744X	0.1191883(3)
		24h	28.04569	17.10174	45.99303	Y =4.085+ 0.632X	0.6404152(3)
		48h	23.26771	15.22007	35.57056	Y =3.899+ 0.806X	0.8789311(3)
	Methanol	30 min	1030.155	257.5667	4120.177	Y =2.813+ 0.726X	5.345154E-02(3)
		24h	57.71285	38.8379	85.76086	Y =3.785+ 0.690X	0.1849041(3)
		48h	26.29665	17.77245	38.90932	Y =3.831+ 0.823X	0.1627083(3)

Table27: Cytotoxicity through dose-mortality of *A. indica* root wood, seed ,stem bark and stem wood extracts (Chloroform and Methanol) against *Artemia salina* nauplii after 30 min, 24h and 48h of treatment.

Test extract		Time exposed	LC_{50} value (ppm)	95% Conf. Limits		Regression equation	χ^2 Value (df)
				Lower limit	Upper limit		
Root wood	Chloroform	30 min	838.2706	129.3991	5430.47	Y =3.179+ 0.623X	0.1919718(3)
		24h	36.47875	23.963	55.53144	Y =3.928+ 0.687X	0.1976204(3)
		48h	8.40184	5.092476	13.86181	Y =4.256+ 0.805X	0.6388855(3)
	Methanol	30 min	5187.234	139.4302	192981.3	Y =2.902+ 0.565X	0.5579266(3)
		24h	82.83993	45.49681	150.8338	Y =3.640+ 0.709X	0.5536576(3)
		48h	23.38707	17.1553	31.88254	Y =3.790+ 0.884X	0.123806(3)
seed	Chloroform	30 min	520.1635	128.2581	2109.577	Y =3.017+ 0.730X	2.811E-03(3)
		24h	24.50645	17.66922	33.98941	Y =3.841+ 0.834X	5.491E-02(3)
		48h	5.942745	3.268797	10.80404	Y =4.376+ 0.806X	8.294E-02(3)
	Methanol	30 min	906.5301	134.699	6100.985	Y =3.090+ 0.646X	7.057E-02(3)
		24h	61.17362	32.74389	114.2874	Y =3.963+ 0.580X	7.998E-02(3)
		48h	18.24789	14.0748	23.65829	Y =3.587+ 1.120X	0.8352814(3)
Stem bark	Chloroform	30 min	1042.544	254.2184	4275.452	Y=2.839+ 0.716X	0.1087818(3)
		24h	196.883	108.0891	358.6199	Y =2.839+ 0.716X	0.1355801(3)
		48h	24.53654	17.16986	35.06388	Y =3.703+ 0.933X	0.3615456(3)
	Methanol	30 min	6030.069	262.4669	138538.1	Y =2.938+ 0.546X	8.504391E-02(3)
		24h	167.7432	89.26254	315.2249	Y =3.492+ 0.678X	0.4133606(3)
		48h	34.24572	24.98084	46.94675	Y =3.572+ 0.931X	0.5406418(3)
Stem wood	Chloroform	30 min	3711.381	297.0884	46364.49	Y =2.828+ 0.608X	0.6351118(3)
		24h	94.12271	61.21007	144.7326	Y =3.532+ 0.744X	0.3724823(3)
		48h	45.16339	32.41925	62.91725	Y =3.629+ 0.829X	1.065525(3)
	Methanol	30 min	1641.063	263.9264	10203.94	Y =2.965+ 0.633X	8.963585E-02(3)
		24h	92.75699	59.8282	143.8093	Y =3.576+ 0.724X	7.130241E-02(3)
		48h	48.30029	34.94275	66.76403	Y =3.580+ 0.843X	0.2553291(3)

Antimicrobial activities of the test extracts:

Antibacterial activity:

The anitbacterial activity of *A. indica extractives* collected in chloroform and methanol of leaves, flower, seed, root bark, root wood, stem bark and stem wood were tested against 14 bacteria (6 Gram-positive bacteria) *S. aureus, B. cereus, B. megaterium, B. subtilis, S. lutea, S.-ß -haemolyticus* and (8 Gram-negative bacteria) *S. typhi, S. dysenteriae, S. shiga, S. sonnei, S. boydii, E. coli, P. aeruginosa* and *Proteus* sp. at concentrations of 50 and 200 µg/disc along with a standard antibiotic, Ciprofloxacin 30µg/disc. The results obtained are shown in Tables (-----).

Antibacterial activity of the seed extracts:

Antibacterial activity of the seed (chloroform and methanol) extracts:

For the seed (chloroform) extract *S. aureus*, *B. cereus*, *B. megaterium*, *B. subtilis*, *S. lutea*, *S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii*, *E. coli* and *Proteus sp*. were responsive with inhibition zones 06, 13, 11, 10, 10, 12, 14, 12, 10, 13, 13 and 12 mm for 50 and 200 µg/disc application and for the methanol extract *S. aureus*, *B. cereus*, *B. subtilis*, *S. lutea, S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii*, *E. coli* and *Proteus sp*. were responsive with inhibition zones 11, 10, 12, 09, 12, 11, 09, 10, 12, 05 and 10mm respectively for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30µg/disc were 30, 28, 28, 30, 28, 30, 30, 29, 29, 28, 28, 28, 28 and 28 mm for the above mentioned test agents respectively.

Table 28: Antibacterial activity of the seed (chloroform and methanol) extracts of *A. indica and* the standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Ciprofloxacin 30 µg/disc
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	
Gram positive bacteria					
S. aureus	06	13	-	11	30
B. cereus	-	11	-	10	28
B. megaterium	-	10	-	-	28
B. subtilis	-	10	-	12	30
S. lutea	-	12	-	09	28
S.-ß -haemolyticus	-	-	-	-	30
Gram negative bacteria					
S. typhi	-	14	-	12	30
S. dysenteriae	-	12	-	11	29
S. shiga	-	10	-	09	29
S. sonnei	-	-	-	-	28
S. boydii	-	13	-	10	28
E. coli	-	13	-	12	28
P. aeruginosa	-	-	-	-	28
Proteus sp.	-	12	05	10	28

Antibacterial activity of the stem bark (chloroform and methanol) extracts

For the stem bark (chloroform) extract only *S. aureus*, *B. megaterium*, *B. subtilis*, *S. lutea*, *S.-ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *P. aeruginosa*, were responsive with inhibition zones 12, 11, 10, 11, 12, 13, 12, 11, 12, 13 and 7mm for 200 and 50 µg/disc application and for the methanol extract *S.*

aureus, *B. megaterium*, *B. subtilis*, *S. lutea*, *S.-ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *P. aeruginosa* were responsive with inhibition zones 10, 09, 10, 10, 11, 12, 13, 10, 12, 11mm and 08, 10, 09mm respectively for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 28, 30, 28, 30, 29, 29, 29, 28, 28, 29, 29 and 29 mm for the above mentioned test agents respectively.

Table 29: Antibacterial activity of the stem bark (chloroform and methanol) extracts of *A. indica* and the standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Ciprofloxacin 30 µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /disc	
Gram positive bacteria					
S. aureus	-	12	-	10	30
B. cereus	-	-	-	-	30
B. megaterium	-	11	-	09	28
B. subtilis	-	10	-	10	30
S. lutea	-	11	-	10	28
S.-ß –haemolyticus	07	12	08	11	30
Gram negative bacteria					
S. typhi	-	13	-	12	29
S. dysenteriae	-	12	10	13	29
S. shiga	-	-	-	-	29
S. sonnei	-	-	-	-	28
S. boydii	-	11	-	10	28
E. coli	-	12	09	12	29
P. aeruginosa	-	13	-	11	29
Proteus sp.	-	-	-	-	29

Antibacterial activity of the stem wood (chloroform and methanol) extracts:

The stem wood extract (chloroform) was responsive to *S. aureus*, *B. cereus*, *B. megaterium*, *B. subtilis*, *S.- ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. boydii*, *E. coli* and *Proteus sp.* with inhibition zones 10, 08, 10, 12, 10, 12, 10, 09, 10 and 10 mm for 200 µg/disc application and for the methanol extract *S. aureus*, *B. megaterium*, *B. subtilis*, *S.- ß -haemolyticus*, *S. typhi*, *S. dysenteriae*, *E. coli* and *Proteus sp*. with inhibition zones 08, 09, 11, 07, 10, 09, 07 and 10mm respectively for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30µg/disc were30, 30, 30, 30, 32, 30, 30, 32, 30, 30, 30, 31, 30 and 30 mm for the above mentioned test agents respectively.

Table 30: Antibacterial activity of the stem wood (chloroform and methanol) extracts of *A. indica* and the standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Ciprofloxacin 30µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /disc	
Gram positive bacteria					
S. aureus		10	-	08	30
B. cereus		08	-	-	30
B. megaterium		10	-	09	30
B. subtilis		12	-	11	30
S. lutea		-	-	-	32
S.- ß -haemolyticus		10	-	07	30
Gram negative bacteria					
S. typhi		12	-	10	30
S. dysenteriae		10	-	09	32
S. shiga		-	-	-	30
S. sonnei		-	-	-	30
S. boydii		09	-	-	30
E. coli		10	-	07	31
P. aeruginosa		-	-	-	30
Proteus sp.		10	-	10	30

Antibacterial activity of the flower (chloroform and methanol) extracts:

The flower extract (chloroform) was responsive to *B. cereus*, *B. subtilis*, *S.- ß –haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii* ,*E. coli* and *P. aeruginosa* with inhibition zones 12, 11, 13, 13, 12, 12, 11, 14 and 12mm for 200 µg/disc application and the methanol extract was responsive to *B. subtilis*, *S.-ß -haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. shiga*, *S. boydii*, *E. coli* and *P. aeruginosa* with inhibition zones 10, 12, 11, 10, 11, 10, 13 and 10mm respectively for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30µg/disc were 30, 30, 30, 30, 32, 30, 32, 30, 30, 30, 31, 32, 32 and 30 mm for the above mentioned test agents respectively.

Table 31: Antibacterial activity of the flower (chloroform and methanol) extracts of *A. indica* and the standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Ciprofloxacin 30µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /disc	
Gram positive bacteria					
S. aureus		-	-	-	30
B. cereus		12	-	-	30
B. megaterium		-	-	-	30
B. subtilis		11	-	10	30
S. lutea		-	-	-	32
S.- ß -haemolyticus		13	-	12	30
Gram negative bacteria					
S. typhi		13	-	11	32
S. dysenteriae		12	-	10	30
S. shiga		12	-	11	30
S. sonnei		-	-	-	30
S. boydii		11	-	10	31
E. coli		14	-	13	32
P. aeruginosa		12	-	10	32
Proteus sp.		-	-	-	30

Antibacterial activity of the leaf (chloroform and methanol) extracts:

The leaf (chloroform) extracts were responsive to *S. aureus, B. megaterium, S.-ß-haemolyticus*, *S. typhi*, *S. boydii, S. lutea, E. coli* and *P. aeruginosa* with inhibition zones 12, 11, 12, 12, 10, 12, 12 and 10 mm for 200 µg/disc application and the methanol extract was responsive to *S. aureus*, *B. megaterium, S. typhi*, *S. lutea*, *S.*

boydii, *E. coli* and *P. aeruginosa* with inhibition zones 09, 10, 11, 10, 12, 10 and 09 mm for the same doses (Table 29); while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 32, 30, 30, 32, 32, 30, 30, 30, 32, 32, 32 and 30 mm for the above mentioned test agents respectively.

Table32: Antibacterial activity of leaf (chloroform and methanol) of *A. indica* and standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Ciprofloxacin 30µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /disc	
Gram positive bacteria					
S. aureus	-	12	-	09	30
B. cereus	-	-	-	-	30
B. megaterium	-	11	-	10	32
B. subtilis	-	-	-	-	30
S. lutea	-	12	-	11	30
S.- ß -haemolyticus	-	12	-	-	32
Gram negative bacteria					
S. typhi	-	12	-	10	32
S. dysenteriae	-	-	-	-	30
S. shiga	-	-	-	-	30
S. sonnei	-	-	-	-	30
S. boydii	-	10	-	12	32
E. coli	-	12	-	10	32
P. aeruginosa	-	10	-	09	32
Proteus sp.	-	-	-	-	30

Antibacterial activity of the root bark (chloroform and methanol) extracts:

In case of the root bark extract (chloroform) *B. cereus*, *B. megaterium*, *B. subtilis, S.- ß -haemolyticus*, *S. typhi*, *S. shiga*, *S. boydii*, *E. coli* and *Proteus sp*. were responsive with inhibition zones 13, 12, 13, 13, 13, 12, 09, 12 and 13mm for 200 µg/disc application, and for the methanol extract *B. cereus*, *B. megaterium*, *B. subtilis, S.- ß –haemolyticus, S. typhi, S. shiga* and *E. coli* were responsive with inhibition zones 11,10,12, 10,11,09 and 11mm for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 32, 30, 30, 32, 30, 30, 30, 32, 30, 32, 30 and 30mm for the above mentioned test agents respectively.

Table33: Antibacterial activity of the root bark (chloroform and methanol) extracts of *A. indica* and the standard Ciprofloxacin.

Test organisms	Diameter of zone of inhibition (in mm)				
	Chloroform extract		methanol extract		Ciprofloxacin 30µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /disc	
Gram positive bacteria.					
S. aureus	-	-	-	-	30
B. cereus	-	13	-	11	30
B. megaterium	-	12	-	10	32
B. subtilis	-	13	-	12	30
S. lutea	-	-	-	-	30
S.- ß -haemolyticus	-	13	-	10	32
Gram negative bacteria					
S. typhi	-	13	-	11	30
S. dysenteriae	-	-	-	-	30
S. shiga	-	12	-	09	30
S. sonnei	-	-	-	-	32
S. boydii	-	09	-	-	30
E. coli	-	12	-	11	32
P. aeruginosa	-	-	-	-	30
Proteus sp.	-	13	-	-	30

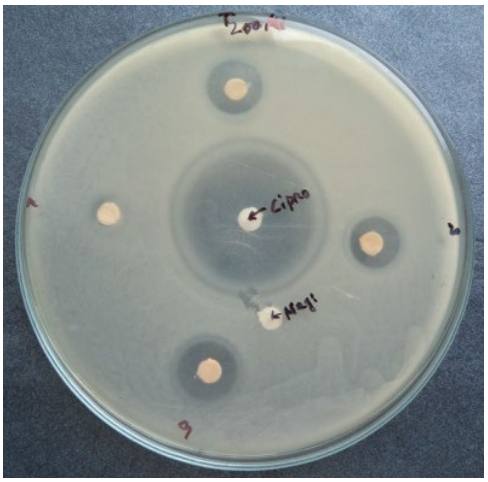
Antibacterial activity of the root wood (chloroform and methanol) extracts:

The root wood (chloroform) extracts were responsive to *S. aureus, B. megaterium, B. subtilis, S.-ß-haemolyticus*, *S. typhi*, *S. dysenteriae*, *S. sonnei, S. boydii, E. coli* and *Proteus* sp. with inhibition zones 12, 13, 12, 13, 14, 13, 13, 12, 13 and 10 mm for 200 µg/disc application, and the methanol extract was responsive to *S. aureus*, *B. megaterium, S. lutea*, *S.- ß –haemolyticus, S.*

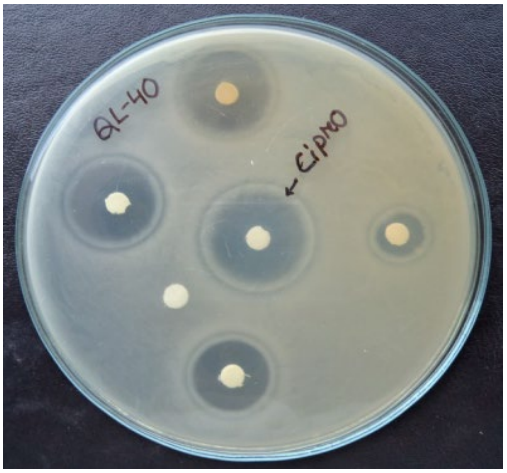
shiga, *S. dysenteriae, S. boydii, E. coli* , *P. aeruginosa* and with inhibition zones 10,11, 11, 09, 12, 10, 12, 10, 11 and 09 mm for the same doses (Table); while the inhibition zones for the standard Ciprofloxacin 30 µg/disc were 30, 30, 30, 32, 30, 30, 28, 29, 29, 30, 28, 30, 30 and 28mm for the above mentioned test agents respectively.

Table34: Antibacterial activity of root wood (chloroform and methanol) Of *A. indica* and standard Ciprofloxacin.

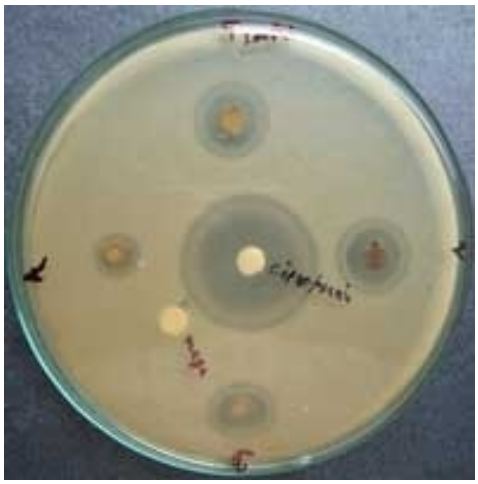
Test organisms	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Ciprofloxacin 30µg/disc
	50µg /disc	200µg /disc	50µg /disc	200µg /dic	
Gram positive bacteria					
S. aureus	-	12	-	10	30
B. cereus	-	-	-	-	30
B. megaterium	-	13	-	11	30
B. subtilis	-	12	-	11	32
S. lutea	-	-	-	-	30
S.- ß -haemolyticus	-	13	-	09	30
Gram negative bacteria					
S. typhi	-	14	-	12	28
S. dysenteriae	-	13	-	10	29
S. shiga	-	-	-	-	29
S. sonnei	-	13	-	12	30
S. boydii	-	12	-	10	28
E. coli	-	13	-	11	30
P. aeruginosa	-	-	-	09	30
Proteus sp.	-	10	-	-	28



Salmonella typhi



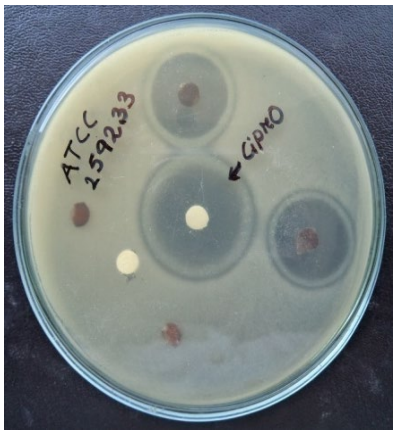
Bacilus subtilis

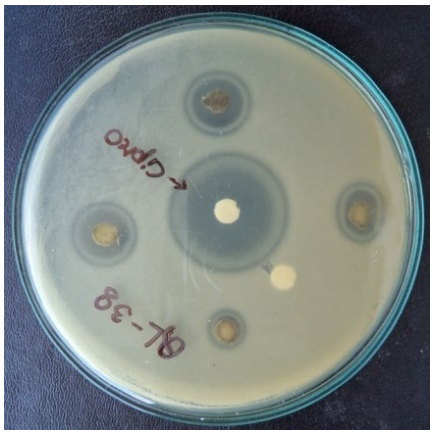


Shigella sonnei

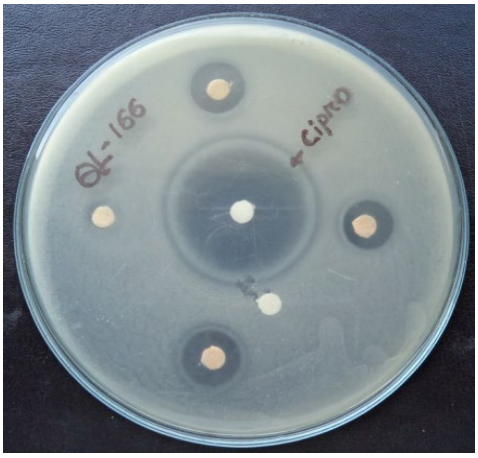
Shigella dysenteriae

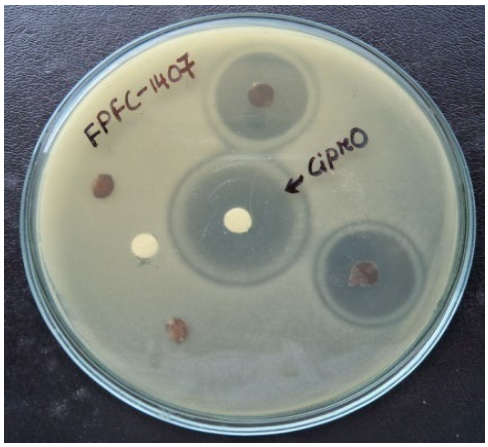
Figure17: Zone of inhibition of each Petri dish measured by standard antibiotic Ciprofloxacin.





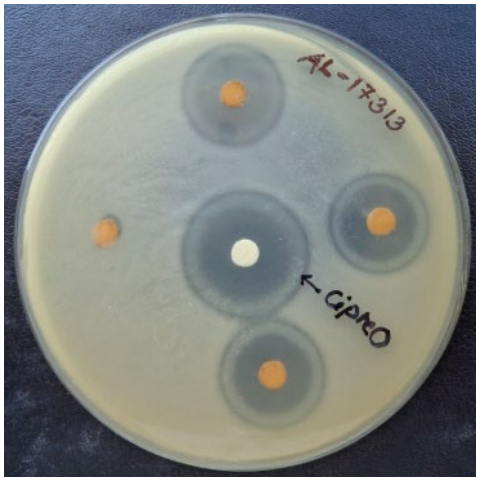
Staphylococcous aureus ***Bacillus megaterium***



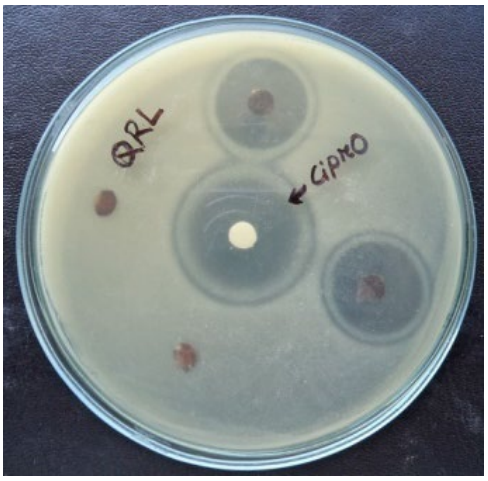


Sarcina lutea ***Escherichia coli***

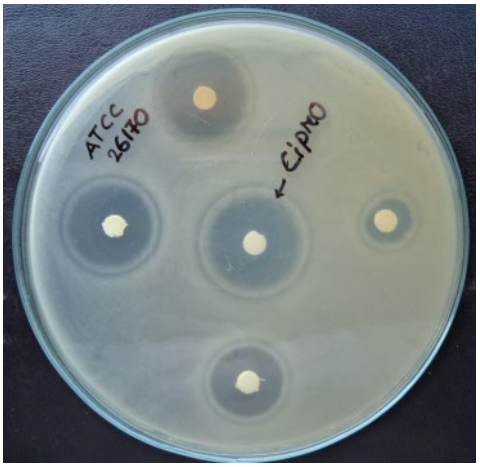
Figure18: Zone of inhibition of each Petri dish measured by standard antibiotic Ciprofloxacin.



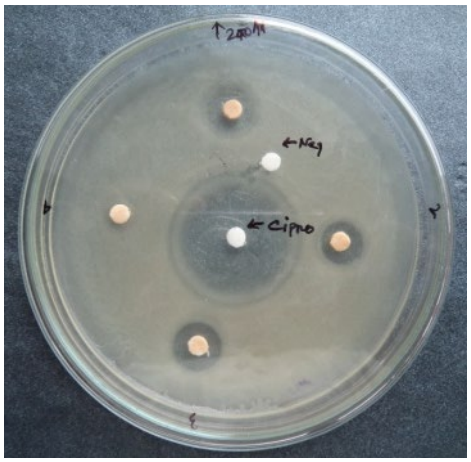
Shigella boydii



Pseudomonas aeruginosa



Shigella shiga



Proteus sp.

Figure19: Zone of inhibition of each Petri dish measured by standard antibiotic Ciprofloxacin.

Minimum inhibitory concentrations (MICs) against test bacteria

Among all the $CHCl_3$ and MeOH extracts of the flower, leaves, root bark, root wood, seed, stem bark and stem wood of *A. indica* only CHCl3 extracts of the seed and the root wood were subjected to evaluate the minimum inhibition zones just depending on the intensity of activity. This was done due to lack of adequate laboratory supports. The results of the MIC values have been presented in Tables (-----). The MIC value of the chloroform extract of the seed was 128µg/ml against *B. cereus,* 64µg/ml against *S.- ß –haemolyticus* and 32µg/ml against *S. dysenteriae*.

Table35: Minimum inhibitory concentrations (MICs) of the chloroform extract of seed against three pathogenic bacteria.

Test tube No.	Nutrient broth medium added (ml)	Seed extract (µg/ml)	Inoculum added (µl)	*S. -ß- haemolyticus*	*S. dysenteriae*	*B. cereus*
1	1	512	10	-	-	-
2	1	256	10	-	-	-
3	1	128	10	-	-	-
4	1	64	10	-	-	+
5	1	32	10	+	-	+
6	1	16	10	+	+	+
7	1	8	10	+	+	+
8	1	4	10	+	+	+
9	1	2	10	+	+	+
10	1	1	10	+	+	+
Cm	1	0	0	-	-	-
Cs	1	512	0	-	-	-
Ci	1	0	10	+	+	+
Results of MIC values in (µg/ml)				64	32	128

"+" = Growth "-" = No growth

The MIC values of the chloroform extract of seed were 128µg/ml against *B. cereus*, 64µg/ml against *S. - ß –haemolyticus* and 32µg/ml against *S. dysenteriae.*

Table36: Minimum inhibitory concentrations (MICs) of the chloroform extract from the root wood against three pathogenic bacteria.

Test tube No.	Nutrient broth medium added (ml)	root wood extract (µg/ml)	Inoculum added (µl)	*S. - ß - haemolyticus*	*S. typhi*	*B. megaterium*
1	1	512	10	-	-	-
2	1	256	10	-	-	-
3	1	128	10	-	-	-
4	1	64	10	+	-	-
5	1	32	10	+	-	+
6	1	16	10	+	+	+
7	1	8	10	+	+	+
8	1	4	10	+	+	+
9	1	2	10	+	+	+
10	1	1	10	+	+	+
Cm	1	0	0	-	-	-
Cs	1	512	0	-	-	-
Ci	1	0	10	+	+	+
Results of MIC values in (µg/ml)				128	32	64

"+" = Growth "-" = No growth.

The MIC values of the chloroform extract of root wood were 128µg/ml against *S. - ß –haemolyticus;* 64µg/ml against *B. megaterium* and 32µg/ml against *S. typhi*.

Antifungal activity:

Antifungal activity of the *A. indica* extractives collected in chloroform and methanol of flower, leaves, root bark, root wood, seed, stem bark and stem wood were tested against six pathogenic fungi *F. vasinfectum*, *A. fumigatus*, *A. niger*, *A. flavus, C. albicans* and *P. notatum* at concentrations of 50 and 200µg/disc along with a standard Nystatin (50µg/disc). The results obtained are shown in Tables (--).

Antifungal activity of flower (chloroform and methanol) extracts:

For the flower extract (chloroform) *A. flavus, A. niger*, *P. notatum* and *C. albicans* were responsive with inhibition zones 13, 12, 12, 11mm and for the methanol extract 10, 10, 14 and 10mm for 200 µg/disc application (Table) while the inhibition zones for the standard Nystatin 50 µg/disc were 20, 20, 22, 22, 21 and 20mm for the above mentioned test agents respectively.

Table37: Antifungal activity of flower (chloroform and methanol) extracts of *A. indica* and standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Nystatin 50µg/disc
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	
F. vasinfectum	-		-	-	20
A. fumigatus	-	-	-	-	20
A. niger	-	12	-	10	22
A. flavus	-	13	-	10	22
P. notatum	-	12	-	14	21
Candida albicans	-	11	-	10	20

Antifungal activity of the leaf (chloroform and methanol) extracts

In case of the leaf extracts (chloroform) *A. niger*, *A. flavus*, *P. notatum* and *C. albicans* were responsive with inhibition zones 14, 13, 12, 12mm and for the MeOH extract 12, 11, 10, 11mm for 200 µg/disc application (Table), while the inhibition zones for the standard Nystatin 50µg/disc were 20, 20, 22, 22, 20 and 20mm for the above mentioned test agents respectively.

Table38: Antifungal activity of leaf (chloroform and methanol) extracts of *A. indica* and standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Nystatin 50µg/disc
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	
F. vasinfectum	-	-	-	-	20
A. fumigatus	-	-	-	-	20
A. niger	-	14	-	12	22
A. flavus	-	13	-	11	22
C. albicans	-	12	-	10	20
P. notatum	-	12	-	11	20

Antifungal activity of root bark (chloroform and methanol) extracts:

For the root bark extracts (chloroform) *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 14, 13, 10, 11mm and for the methanol extract 13, 11, 08, 10mm for 200 µg/disc application (Table); while the inhibition zones for the standard Nystatin 50 µg/disc were 20, 20, 23, 23, 22 and 20mm for the above mentioned test fungi.

Table39: Antifungal activity of root bark (chloroform and methanol) extracts of *A. indica* and the standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Nystatin
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	50µg/disc
F. vasinfectum	-	-	-	-	20
A. fumigatus	-	-	-	-	20
A. niger	-	14	-	13	23
A. flavus	-	13	-	11	23
C. albicans	-	10	-	08	22
P. notatum	-	11	-	10	20

Antifungal activity of root wood (chloroform and methanol) extracts

In case of the root wood extract ($CHCl_3$) *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones15, 13, 13, 12mm and for the MeOH extract 12, 11, 12, 11mm for 200 µg/disc application (Table); while the inhibition zones for the standard Nystatin 50µg/disc were 20, 20, 23, 23, 22 and 20 mm for the above mentioned test agents respectively.

Table 40: Antifungal activity of root wood (chloroform and methanol) extracts of *A. indica* and standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Nystatin
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	50µg/disc
F. vasinfectum	-	-	-	-	20
A. fumigatus	-	-	-	-	20
A. niger	-	15	-	12	23
A. flavus	-	13	-	11	23
C. albicans	-	13	-	12	22
P. notatum	-	12	-	11	20

Antifungal activity of Seed (chloroform and methanol) extracts:

In case of the seed extract (chloroform) only *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 16, 14, 12,11mm and for the methanol extract 13, 12, 10, 09 mm for 200 µg/disc (Table); while the inhibition zones for the standard Nystatin 50 µg/disc were 23, 24, 24, 24, 23 and 23mm for the above mentioned test fungi respectively.

Table 41: Antifungal activity of seed (chloroform and methanol) extracts of *A. indica* and the standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	chloroform extract		methanol extract		Nystatin 50µg/disc
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	
F. vasinfectum	-	-	-	-	23
A. fumigatus	-	-	-	-	24
A. niger	-	16	-	13	24
A. flavus	-	14	-	12	24
C. albicans	-	12	-	10	23
P. notatum	-	11	-	09	23

Antifungal activity of stem bark (chloroform and methanol) extracts

For the stem bark extract ($CHCl_3$) only *A. niger*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 13, 12, 13, 11mm and for the MeOH extract 12, 10, 11, 10mm for 200 µg/disc application (Table); while the inhibition zones for the standard Nystatin 50 µg/disc were 24, 24, 25, 25, 23 and 23 mm for the above mentioned test agents respectively.

Table 42: Antifungal activity of stem bark (chloroform and methanol) extracts of *A. indica* and the standard Nystatin.

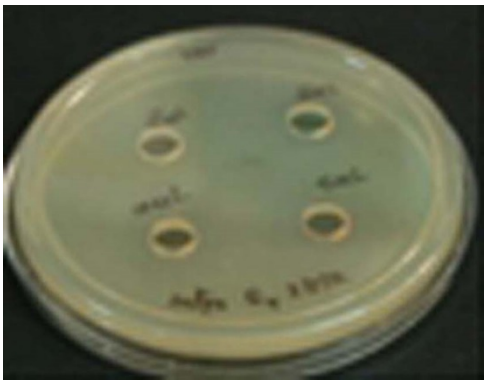
Test Fungi	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Nystatin
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	50µg/disc
F. vasinfectum	-	-	-	-	24
A. fumigatus	-	-	-	-	24
A. niger	-	13	-	12	25
A. flavus	-	12	-	10	25
C. albicans	-	13	-	11	23
P. notatum	-	11	-	10	23

Antifungal activity of stem wood (chloroform and methanol) extracts

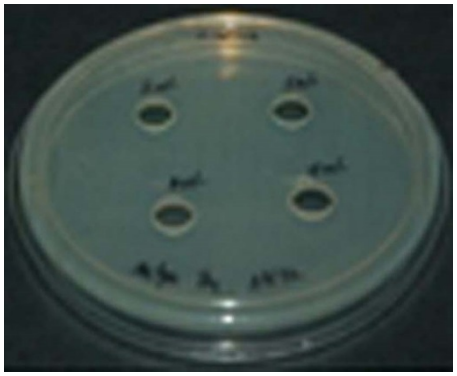
For the stem wood extract ($CHCl_3$) only *F. vasinfectum*, *A. flavus, C. albicans* and *P. notatum* were responsive with inhibition zones 13, 12, 13, 12mm and for the MeOH extract 12, 10, 11, 10mm for 200 µg/disc application (Table); while the inhibition zones for the standard Nystatin 50 µg/disc were 23, 24, 25, 25, 23 and 23 mm for the above mentioned test agents respectively.

Table 43: Antifungal activity of stem wood (chloroform and methanol) extracts of *A. indica* and the standard Nystatin.

Test Fungi	Diameter of zone of inhibition (in mm)				
	Chloroform extract		Methanol extract		Nystatin
	50µg/disc	200µg/disc	50µg/disc	200µg/disc	50µg/disc
F. vasinfectum	-	-	-	-	23
A. fumigatus	-	-	-	-	24
A. niger	-	13	-	12	25
A. flavus	-	12	-	10	25
C. albicans	-	13	-	11	23
P. notatum	-	12	-	10	23

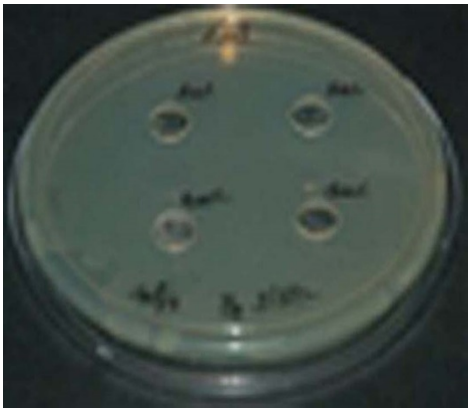


Candida albicans



Aspergillus niger

Penicilium notatum



Aspergillus flavus

Plate20: Zone of inhibition of each Petri dish measured by standard nystatin.

Bioassay of the purified compounds

All the purified compounds of *A. indica* isolated from the leaf were active against Gram positive and Gram negative bacteria and against the selected fungi; and the result is presented in (Table --). The result of the Minimum Inhibitory Concentration (MIC) tests have been presented in Tables (--).

Antibacterial activity of the purified compounds

Among the test bacteria *B. cereus*, *B. megaterium, B. subtilis, S.-ß –haemolyticus* , *S. typhi*, *S. dysenteriae S. shiga, S. sonnei*, *Proteus sp*. and *P. aeruginosa* were responsive to the A_1 and A_2 compounds with the zones of inhibition given in the (Table -) below in comparison to the inhibition by the standard Ciprofloxacin:

Table 44: Antibacterial activity of pure compounds A_1 and A_2 of *A. indica* and the standard ciprofloxacin

Test organisms	Diameter of zone of inhibition (in mm)		
	A_1	A_2	Ciprofloxacin
	200 µg/disc	200 µg/disc	30 µg/disc
Gram positive bacteria.			
S. aureus	-	-	30
B. cereus	14	12	30
B. megaterium	12	11	31
B. subtilis	13	12	30
S. lutea	-	-	30
S. - ß –haemolyticus	14	15	28
Gram negative bacteria			
S. typhi	13	12	30
S. dysenteriae	14	12	30
S. shiga	12	10	31
S. boydii	-	-	29
E. coli	-	-	30
S. sonnei	12	10	28
P. aeruginosa	14	12	30
Proteus sp.	13	11	28

Minimum inhibitory concentrations (MICs) of the purified compound A_1 against test bacteria:

Table 45: Minimum inhibitory concentrations (MICs) of the purified compound A_1 against test pathogenic bacteria.

Test tube No.	Nutrient broth medium added (ml)	Compound A_1 (µg/ml)	Inoculum added (µl)	*S. -ß- haemolyticus*	*S. dysenteriae*	*B. cereus*
1	1	512	10	-	-	-
2	1	256	10	-	-	-
3	1	128	10	-	-	-
4	1	64	10	-	-	+
5	1	32	10	+	-	+
6	1	16	10	+	+	+
7	1	8	10	+	+	+
8	1	4	10	+	+	+
9	1	2	10	+	+	+
10	1	1	10	+	+	+
Cm	1	0	0	-	-	-
Cs	1	512	0	-	-	-
Ci	1	0	10	+	+	+
Results of MIC values in (µg/ml)				16	64	32

"+" = Growth "-" = No growth

The MIC values of the pure compound A_1 were 32µg/ml against *B. cereus*, 16µg/ml against *S. - ß –haemolyticus* and 64µg/ml against *S. dysenteriae.*

Minimum inhibitory concentrations (MICs) of the purified compound A_2 against test bacteria:

Table 46: Minimum inhibitory concentrations (MICs) of the purified compound A_2 against test pathogenic bacteria.

Test tube No.	Nutrient broth medium added (ml)	CompoundA_2 (µg/ml)	Inoculum added (µl)	*S. -ß- haemolyticus*	*S. dysenteriae*	*B. cereus*
1	1	512	10	-	-	-
2	1	256	10	-	-	-
3	1	128	10	-	-	-
4	1	64	10	-	-	+
5	1	32	10	+	-	+
6	1	16	10	+	+	+
7	1	8	10	+	+	+
8	1	4	10	+	+	+
9	1	2	10	+	+	+
10	1	1	10	+	+	+
Cm	1	0	0	-	-	-
Cs	1	512	0	-	-	-
Ci	1	0	10	+	+	+
Results of MIC values in (µg/ml)				64	32	64

"+" = Growth "-" = No growth

The MIC values of the pure compound A_2 were 64µg/ml against *B. cereus*, 64µg/ml against *S. - ß –haemolyticus* and 32µg/ml against *S. dysenteriae*.

Antifungal activity of the purified compounds:

Among the test fungi A. niger, *A. flavus, C. albicans* and *P. notatum* were responsive to the A_1 and A_2 compounds with the zones of inhibition given in the (Table 39) below in comparison to the inhibition by the standard nystatin.

Table 47: *In vitro* antifungal activity of compounds A_1 and A_2 of *A. indica* and the standard nystatin.

Test Fungus	Diameter of zone of inhibition (in mm)		
	A_1	A_2	**Nystatin 50µg/disc**
	200 µg/disc	**200 µg/disc**	
F. vasinfectum	-	-	20
A. fumigatus	-	-	21
A. niger	14	12	18
A. flavus	14	12	20
C. albicans	14	11	18
P. notatum	13	11	18

Effects of phytochemical screening of plant extract of *A. indica:*

The present study was carried out on the plant extracts revealed the presence of medicinally important bioactive compounds. The plant *A. indica* leaf, stem bark, root wood and seed shows alkaloids, carbohydrates, flavanoids, glycosides, phenol, protein, resins, saponnins, tannins and sterols of different solvents. The phytochemical screening was performed with chloroform and methanol extracts of different parts of *A. indica*. The leaves of *A.*

indica were showed alkaloids, carbohydrates, flavanoids, glycosides, phenol, resins, saponnins, tannins and fat were present in methanol extracts. Proteins and steroids were absent for methanol extracts. Carbohydrates, resins, fat and steroids were absent in chloroform extracts (Table48**).**

Table 48: Phytochemical screening of leaf extracts of *A. indica*

Class of compounds indicated	Chloroform	Methanol
Alkaloids	+	+
Carbohydrates	-	+
Flavanoids	+	+
Glycosides	+	+
Phenols	+	+
Proteins	+	-
Resins	-	+
Saponins	+	+
Tannins	+	+
Steroids	-	-
Fat	-	+

'+' = Presence; '-'= Absence

Preliminary phytochemical analysis of the different solvent extracts of the stem bark of *A. indica* alkaloids, flavonoids, glycosides, phenol, Terpenoids, saponnins and tannins were present for methanol extracts but Carbohydrates, proteins and steroids are absent. In case of chloroform extract, only alkaloids, terpenoids, saponnins and tannins were present but others are absent (Table 49).

Table 49: Phytochemical screening of stem bark extract of *A. indica*.

Class of compounds indicated	Chloroform	Methanol
Alkaloids	+	+
Carbohydrates	-	-
Flavonoids	-	+
Glycosides	-	+
Phenols	-	+
Proteins	-	-
Terpenoids,	+	+
Saponins	+	+
Tannins	+	+
Steroids	-	-

'+' = Presence; '-'= Absence

In case of root wood of *A. indica* alkaloids, flavanoids, phenol, tannins, Saponins and steroids are present for chloroform extract but Carbohydrates, glycosides, proteins and resins are absent. For methanol extract only Carbohydrates, proteins, resins and saponins are absent (Table50).

Table 50: Phytochemical screening of *A. indicia* root wood extracts.

Class of compounds indicated	Chloroform	Methanol
Alkaloids	+	+
Carbohydrates	-	-
Flavonoids	+	+
Glycosides	-	-
Phenols	+	+
Proteins	-	-
Resins	-	-
Saponins	+	-
Tannins	+	+
Steroids	+	+

'+' = Presence; '-'= Absence

For the of seeds alkaloids, carbohydrates, phenols, proteins, saponins and tannins were present in the chloroform extracts and flavanoids, glycosides, steroids and resins were absent. In case of methanol extract alkaloids, carbohydrates, flavanoids, phenols, proteins, resins and tannins were present but glycosides, steroids and saponins were absent (Table 51).

Table 51: Phytochemical screening of seed extracts *of A. indica*.

Class of compounds indicated	Chloroform	Methanol
Alkaloids	+	+
Carbohydrates	+	+
Flavanoids	-	+
Glycosides	-	-
Phenols	+	+
Proteins	+	+
Resins	-	+
Saponins	+	-
Tannins	+	+
Steroids	-	-

'+' = Presence; '-'= Absence

Table 52: Summary of biological activity of the (chloroform and methanol) extracts of *A. indica* at a glance.

Test types	Activity traced	Flower		Leaves	
		Chloroform	Methanol	Chloroform	Methanol
Insecticidal activity	*T. castaneum*	√	√	√	√
Larvicidal activity	*T. castaneum*	√	√	√	√
Repellency	*T. castaneum*	√	√	√	√
Cytotoxicity	*A. salina*	√	√	√	√
Antimicrobial	Antibacterial activity	√	√	√	√
	Antifungal activity	√	√	√	√
Phytochemical		×	×	√	√

'√'= activity found, '×'= activity not found, '---'=weak activity found

Table 53: Summary of biological activity of the (chloroform and methanol) extracts of *A. indicate* at a glance.

Test types	Activity traced	Root bark		Root wood	
		Chlorofom	Methanol	Chloroform	Methanol
Insecticidal activity	*T. castaneum*	√	√	√	√
Larvicidal activity	*T. castaneum*	√	√	√	√
Repellency	*T. castaneum*	√	√	√	√
Cytotoxicity	*A. salina*	√	√	√	√
Antimicrobial	Antibacterial activity	√	√	√	√
	Antifungal activity	√	√	√	√
Phytochemical		×	×	----	√

'√'= activity found, '×'= activity not found, '---'=weak activity found

Table 54: Summary of biological activity of the (chloroform and methanol) extracts of *A. indica* at a glance.

Test types	Activity traced	Stem bark		Stem wood		seed	
		Chloro form	Metha nol	Chloro form	Metha nol	Chloro form	Metha nol
Insecticidal activity	*T. castaneum*	√	√	√	√	√	√
Larvicidal activity	*T. castaneum*	√	√	√	√	√	√
Repellency	*T. castaneum*	√	√	√	√	√	√
Cytotoxicity	*A. salina*	√	√	√	√	√	√
Antimicrobial	Antibacterial activity	√	√	√	√	√	√
	Antifungal activity	√	√	√	√	√	√
Phytochemical		----	√	×	×	---	√

'√'= activity found, '×'= activity not found, '---'=weak activity found

Chapter-6
Discussion

The use of phytochemicals as well as plant products such as powder, oil and extracts for the control of stored-product insect pests has agricultural importance and recently has received much more attention because these insecticidal compounds are safer than the synthetic pesticides and can be easily obtained from plants with less sophisticated methods. Being situated in the Oriental Region (Sub Tropical) Bangladesh has a huge diversity of species with a plenty of promising plants of which *Azadirachta indica* was taken into consideration, because this plant has been appeared into human notice as a source of phytochemical screening of leaf and stem in various extracts i.e. petroleum ether, benzene, chloroform, acetone, methanol, rectified spirit and water shows that there is presence of alkaloids, carbohydrate, proteins, tannins, Saponin anthraquinone glycosides, cardiac glycosides, flavanoides and phenolic compounds, quinone, steroids. As a native plant of Bangladesh much attention was not paid to work out its potentiality or for its further possibilities of contribution to the overall development of our country. Thus, the present experiments have been carried out to investigate the extractives of different parts of *A. indica* viz. flower, leaves, root bark, root wood, seed, stem bark and stem wood for their insecticidal, insect larvicidal and insect repellent activity against *Tribolium castaneum*, cytotoxicity

against *A. salina*, phytochemical potential and antimicrobial activity against a number of pathogenic bacteria and fungi.

The chloroform and methanol extracts of the flower, leaf, root bark, root wood, seed, stem bark and stem wood of *A. indica* showed insecticidal potentials and their LD_{50} values have been established as well. The seed extracts (of both the solvents) offered highest mortality of *T. castaneum* beetles, however the comparative larger doses of the root bark extracts indicate weaker action of the two test extracts offered mortality within 24 hours of application just to prove their acute toxicity. Both the seed was found to possess bioactive potential(s) with comparatively higher insecticidal activity after the root bark extract against *T. castaneum adults* through surface film assay.

The present results support the previous works Redfern et al., (1981) reported 83% mortality in second and fourth-instar larvae of *Spodoptera frugiperda* in response to treatment with 10 µg of Azadirachtin (insecticide). In the present case, 4.5µg/larva (Coopex) and 12.5µg/larva (N-9) caused 90% and 62% mortality, respectively, but the higher toxicity of Coopex in this case may have been due to the mode of action. N-9 controls the population by physiological mechanisms, like the IGR effect, while pyrethrum kills by toxic effect. Higher control by Coopex (90%) was due to its toxic action. Ladd (1984) reported that the LD_{50} of Azadirachtin against *Popillio japonica* was 0.1µg/larva. The LD_{50} reported by Ladd (1984) is very low in comparison to the present study, which may be due to the different compounds used and tolerance levels

of the insects. Ladd (1984) also reported the direct proportionality of the compound to insect mortality.

These results also support the results of Meisner et al., (1981) who reported the residual effect of some neem products on the larvae of *Spodoptera littoralis.* In field trials the neem suspensions on sugar beet leaves had the highest residual activity. On cotton, almost no protection of the leaves was obtained with any of the products tested. On lucern, all the products showed good residual activity after 24 h when applied at 0.6% concentration, where as at 0.2% only the seed suspension was active.

The present results support the previous works (Hanifah *et al.,* 2011) studied acaricidal activity of *Chymbopogon citratus* and *A. indica* against House Dust Mites (*Dermatophagoides farinae*) and *Dermatophagoides pteronyssinus.* At 50%concentration of lemongrass resulted in 91% mortality in both the species. At 50% concentration of Neem resulted in 40.3% mortality of topical activity in *D. pteronyssinus* and 15.7% against *D. farinae*. Contact mortality was 8.0% and 8.9% against both the moths. The Effect of ethanol plant extracts on three storage grain pests of economic importance was studied by (Manzoor *et al*., 2011). They found that the screening of plant extracts from wild species of plants for insecticidal properties could lead to the discovery of new agents for pest control.

Asifa Hameed *et al.,* (2012) studied toxicological effects of neem (*A. indica*) Kanair (*Nerium oleander*) and Spinosad (Tracer 240 SC) on the red flour beetle (*T. castaneum*) (Herbst).They noticed that the ethanol extracts of Kanair was found least effective against *Tribolium* sp. In comparison with neem extracts and spinosad. Maximum mean mortality (38.13%) at 168 h exposure time with maximum dose (2.5%) and minimum (15.63%) mortality at 24 h with 0.5% dose. Neem showed maximum mortality (45.63%) was found at exposure time maximum dose of 2.5% and minimum control (16.88%) was at 24 h with 0.5% concentration.

The present results also support the results of Najafabadi *et al*., (2015) which they demonstrated the repellency and toxicity of three plants leaves extraction against *Oryzaephilus surinamensis* L. and *Tribolium castaneum* Herbst. The results revealed that all of the tested materials had repellent and lethal effects against the tested pests as compared to untreated check. The plant extracts were mixed with grain 10mg/g of grains. Comparison of test plant extracts on *O. surinamensis* showed that the Mint extract was the most effective causing 48.30±4.01 mortality percent. Datura and Neem extracts with 35.26±3.21 and 25.60±2.33 mortality percent were the next levels. But, the plant extracts effect on *T. castaneum* revealed that Datura, Neem and Mint extracts were the most effective with 21.42±2.31, 16.66±1.54 and 15.95±1.89 mortality percent, respectively.

No such previous works have been traced that reveals repellent activity of the *A. indica* extracts through experiments directly on *T. castanium* adults, while it was done in the present investigation to establish high degree of repellent potentials of the flower, leaf, root bark, root wood, seed, stem bark and stem wood extracts of the experimental plant. Repellency by the $CHCl_3$ extracts of *A. indica* against *T. castaneum* adults was very much promising, while all the extracts found to repel at 0.01% level of significance ($P<0.001$) except the stem bark extract which was found active at 0.1% level of significance ($P<0.01$). The repellency record triggers a hope for the use of *A. indica* extracts as repellents since most of the extracts repelled the beetles significantly. Singh *et al.,* (2001) carried out antifeedant activity tests of some famous (of bioactive potentials) plants including a related species that help understanding repellent potentiality of the *Azadirachta* extractives. These included the extracts from the leaves and roots of *Achyranthus aspera*, *Acorus colomus*, leaf and oil of *A. indica*, leaves of *Chrysanthemum cinerariefolium*, *Derris elliptica* and *Datura alba* under laboratory conditions. Among the treatments the least antifeedant effect was observed with the leaf extract of *Annona squamosa* showing 60.43 per cent damage to the treated pods.

These findings also supports the results of Diabaté *et al*., (2014) studied the toxicity, antifeedant and repellent effect of *Azadirachta indica* (A. Juss) and *Jatropha carcus* L. aqueous extracts against *Plutella xylostella*. The repellent effect of these products was evaluated by bioassay using choice tests. The results indicate that

the aqueous extracts based on neem and jatropha seeds and leaves act as potential repellent activities to *P. xylostella* larvae. In fact, the aqueous neem seeds extracts 80 g/L (T1) (36.67%) and 50 g/L (T5) (36.67%), and of jatropha seeds 80 g/L (T2) (24.17%) and 50 g/L (T6) (36.67%), and aqueous extracts of jatropha leaves 67 g/L (T8) (27.5%) and T' (30.00%) were on repellent class II. Their repellency activities were superior to those of insecticides Decis (18.33%) and Cypercal (6.67%), aqueous neem leaves extracts 67 g/L (T3) (19.17), T (8.33), T'' 1 (5.83) and T'' 2 (14.17) which were on repellent class I.

These results are in agreement with the results of Hanif *et al*., (2016) who demonstrated the insecticidal and repellent activities of essential oils of three medicinal plants towards insect pests of stored wheat and the study was carried out to assess the efficacy of essential oils of some indigenous plants *Melia azadarach* (Bakain), *Azadirachta indica* (Neem) and *Datura stramonium* (Datura) for their potential repellent and mortality efficiencies against three most important insect pests of stored grain (*Tribolium castaneum*, *Rhyzopertha dominica* and *Trogoderma granarium*). Experiment was performed at Grain Research, Training and Storage Management Cell of the Department of Agri. Entomology, University of Agriculture Faisalabad. Three concentrations viz., 5%, 10% and 15% of essential oils with three replications were applied. They noticed that *Azadirachta indica*, showed the most effective repellant against *Tribolium castaneum* and *Rhyzopertha dominica* with maximum of 77.66% and 81.48% repellency respectively. While *Datura stramonium* depicted highest repellency

(76.43%) against *Trogoderma granarium*. For mortality assay, data was collected after 24h, 48h and 72hr of treatment with plant oils. The highest mortality of *Tribolium castaneum* and *Trogoderma granarium* was observed against *Datura stramonium* which was 28.82 and 24.30% respectively. While in case of *R. dominica* observed maximum mortality was 25.45% against *A. indica*.

To find dose-mortality against multicellular organism's larvicidal activity tests were also carried out in the present investigation. The $CHCl_3$ extracts of different parts of *A. indica* were applied against the larvae of *T. castaneum*. The mortality has been raised proportional to the magnification of the amount of ingestion of the treated food by the larvae, which was thus proportional to their ages. No previous record was found on the biological assay of the *A. Indica* extracts against *T. castaneum* larvae, however different larvicidal assays have been carried out by the previous workers with the *Azadirachta* spp. (Schmutterer H, 1990, Abdelouaheb *et al.*, 2009, Chavan, 1984; Virendra *et al.,* 2009; Aliero *et al.,* 2003; Senthil *et al.,* 2006), and these results support the findings of the present investigation. Larvicidal activity of *Azadirachta indica* against various species of mosquitoes has been observed by various researchers (Chavan, 1984; Virendra *et al.,* 2009; Aliero, 2003; Abdelouaheb *et al.,* 2009; Senthil *et al.,* 2006). The extracts produced some abnormalities in larvae. Larval and pupal intermediates were observed. Partially emerged adults showed crumpled legs and entangled in pupation. All these abnormalities were also reported by Naqvi (1987). Correspondingly, the extracts of neem leaves in different solvents (petroleum ether, ether and

EtoH) were evaluated for mosquito (C*ulex pipiens fatigans)* larvicidal activity according to W.H.O. method. The 1% petroleum ether extract showed 100% mosquito larvicidal activity, it also had good residual activity (for 144hr) at 0.2% (Chavan, 1984). Neem-based pesticides are now extensively used in agriculture practices all over the world. It contains azadirachtin, which is a predominant insecticidal active ingredient, having antifeedants, ovipositional deterrence, repellency, growth disruption, sterility and larvicidal action against insects (Schmutterer H, 1990). There are various reports of control of mosquito breeding under field conditions. An emulsion of neem oil in water was found to be effective in controlling breeding of *Cx. quinquefasciatus*, *An. stephensi* and *Ae. aegypti* in pools, tanks and coolers up to 2 to 3 weeks (Batra et al., 1998), whereas an application of neem cake powder resulted in drastic reduction in the late instar larvae and pupae of culicine mosquitoes in paddy field (Rao *et al*., 1992). Dhar *et al*., (1996) demonstrated the inhibitory effect of neem oil volatiles on geotropic cycle in *An. stephensi* and *An. culicifacies*. A neem oil formulation containing 32% neem seed oil (an equivalent of 0.03% azadirachtin), an emulsifier (5%) and 63% isopropanol (solvent) was investigated for its larvicidal activities against *An. gambiae* (Okumu *et al*., 2007). Generally, insecticides enter insects by cuticular penetration and/or ingestion and then pass throughout the interior of the insect to the site of action (Matsumura, 1975). Although their reports provided some evidence regarding the action site of the bioactive compounds, even though the mechanism causing mortality of mosquito larvae is remained

unknown and needs to be studied further. However, these studies demonstrated and emphasized the potential of those plants against *A. aegypti* larvae and its benefit to developing new types of larvicide's used for mosquito control.

These findings also supports the results of Aditi et al., (2011) studied the larvicidal properties of the extracts of *A. indica*. Laboratory reared larvae were exposed to 1ppm concentration of *A. indica.* Result showed that the *A. indica* elicited 70-99% mortality to larvae. The extract of *A. indica* was found to be significantly effective in controlling *Culex larvae*.

Cytotoxicity test was also carried out which revealed good results of activity of the extractives of the experimental plant. The test materials were found cytotoxic; while the test was carried out through dose-mortality assay it was possible to establish the LC_{50} values as part of this experimentation. According to the intensity of activity the results of the extracts against the brine shrimp nauplii could be arranged in the following order: seed > root wood >root bark> stem bark> flower>stem wood> leaf for the chloroform extracts and seed > root wood > flower> root bark> stem bark> stem wood> leaf for the methanol extracts and the toxicity offered by the extracts were very much promising. In support of these findings screening results for cytotoxicity by many previous researchers done on an allied species of *Azadirachta* were available. These findings support the ethanolic extracts of *Derris scandens* (Roxb.) Benth, along with other test extracts showed cytotoxicity (IC_{50}<30 µg/ml) against lung and prostate cancer cell lines (Acharya and Thomas, 2007). Another similar work was also

available done on cytotoxicity. These tests showed LC_{50} of petroleum ether, chloroform and methanol extracts on *A. salina* Leach as 1.14, 1.1, and 54.9mg/l respectively. Chemical analysis revealed the presence of fatty acids, steroids, triterpenoids, alkaloids, phenols, and phenyl propionates, tannin, and mucilage in the extracts (Uyub *et al.,* 2010).

These results are in agreement with the results of Diabate *et al*., (2014) demonstrated the LC_{50} values of insecticide Decis and Cypercal were 0.11 and 1.26 g/L respectively. LC_{50} of aqueous extracts of jatropha seeds, neem seeds, neem leaves and of jatropha leaves were 9.32, 17.45, 116.45 and 169.95 g/L respectively. For the mixture of aqueous extracts of jatropha seeds and of neem leaves, LC_{50} were 12.08. The toxicity of different products were ranked more effective to least effective on *P. xylostella* larvae by comparing the LC_{50}/CU report. Thus, all aqueous extract of neem and jatropha were more toxic by ingestion to the insecticides Decis and Cypercal. Aqueous extract of Jatropha seeds was most toxic. It was followed respectively by the mixture of aqueous jatropha seeds extract and neem leaves, aqueous neem seed extract, aqueous neem leaves extract and by aqueous jatropha leaves extracts.

These findings also support the results of (Cohen *et al*., 1996) which they demonstrated that Nimbolide shows potent cytotoxic effect on N1E-115 neuroblastoma (mouse), 143B.TK- osteosarcoma (human) and Sf9 (insect) cultured cell lines with LC_{50} value of 4–10 mM. Other limonoids like epoxyazadiradione and salanin show cytotoxic effect at LC_{50} value of 27 and 112 mM

respectively. Nimbidin, deacetylnimbin and azadirachtin are practically nontoxic. Acetyl cholinesterase (AchE), Na+ K+, and Ca++– ATPase are significantly inhibited, while Mg2+– ATPase level increases significantly in rat brain when treated orally with 80, 160 and 320 mg/kg of vepacide, an active ingredient from neem seed oil, daily for 90 days126. Several studies were performed with Margosan 'O', an extract of neem seeds. However, no apparent toxic manifestations were noticeable in rats or mice110. LC_{50} of Margosan 'O' is more than 2 ml/kg in albino rabbits when tested for acute dermal toxicity (Kanungo, 1996). However, Margosan 'O' showed minimal irritation in both eyes (Kanungo, 1996) when applied to one washed and one unwashed eye of albino rabbits over seven days. NIM-76, a volatile fraction of neem oil, possesses antifertility activity when applied before coitus in rats, rabbits and rhesus monkeys (Riar, *et al.*, 1991).

These findings were toxic to mosquito larvae with LC_{50} value of 11 ppm and also reported to possess insect growth regulators (Gianotti *et al*., 2008). Azadirachtin acts as anti-ecdysteroid and kills larvae by growth inhibition effect (Zebit CPW,1984) Neem-based bio pesticides and neem extracts have a wide range of effects against insect pests including repellence, feeding, toxicity, sterility and growth regulator activity and are relatively safe towards non- target biota with only minimal risk of direct adverse effects on aquatic biota from contamination of water bodies (Kreutzweiser ,1997) Allelochemicals such as azadirachtin, nimbin, nimbidin, nimbolides, nimolic acid, salannin, melianttriol, azadirachtol present in neem affect the biochemical and

physiological processes of insect system and nullify the insect detoxification mechanism thereby not allowing the pest to develop resistance. As an emulsifiable concentrate, the neem oil formulation had greatly reduced sized particles and evenly mixed within the water column with a few suspended particles on the water surface. The spread of these fine particles probably increased the efficacy of formulation. it has the advantage of being eco-friendly, effective and ability to prevent the development of pest resistance.

Some earlier workers reported various plant materials including essential oils in checking the multiplication of pests in stores (Krishnamurti and Rao, 1944; Su *et al.,* 1972). The pongam oil from the seeds has already been marketed as karanjin bio pesticide by SOM Phytopharma (India) Limited [http://www.somphyto.com], along with anonym, and azadirachtin. Toxicological effects of neem (*Azadirachta indica*) was marked by **(**Hameed *et al***.,** 2012).The Pepper Research Station, Panniyur, India carried out an experiment on a mixture of pongam oil and neem-seed oil to apply as a biopesticide against *C. maculatus* and the SAARC Documentation Center, India recorded it as an excellent result. The antioxidant activity of herbal extract is due to various biochemicals, which act as inhibitors of the process of oxidation, and thus have diverse physiological role in the body. The free radicals are in those molecules/atoms that possess an unpaired electron in their outermost orbit, which is capable of inducing chain reaction and thereby damaging different types of cells, resulting into accelerated aging, various diseases, stress,

and also hampering the defense mechanisms of the body. Various Indian medicinal herbs of common use are known to have remarkable antioxidant properties; while 25 Indian medicinal plants including *A. indica* were tested to show antioxidant potentials. Different parts of the selected plants were tested individually for their antioxidant activity using established DPPH Radical Scavenging Method, and the *A.* spp. plants gave activity ranging from 45-75% (Kamal *et al.,* 2004).

The flower, leaves, root bark, root wood, seed, stem bark and stem wood of *A. indica* $CHCl_3$ and methanol extracts were subjected to evaluate the antimicrobial activity (on some selected pathogenic bacteria and fungi) and the minimum inhibitory action just depending on the intensity of activity against the selected Gram positive and Gram negative bacteria. The activities were found very much promising. The seed and root wood extracts offered the MIC values 128 µg/ml against *B. cerus* and 128 µg/ml against *S. -ß- haemolyticus* 64µg/ml against *S. -ß- haemolyticus*, *B. megaterium* and 32 µg/ml against *S. dysenteriae* and *S. typhi.*

These results are in agreement with the results of Nishimura *et al*., (1997) reported antibacterial activity of crude bark extract of neem (*A. indica*) against *streptococcus sobrinus*. Antibacterial activity of acetonic and aqueous extracts of neem bark examined on agar plates by using strain of *streptococcus sobrinus*. Asthana *et al*., (2006) reported antimicrobial entity from the cyanobacterium *Fischerella sp.* isolated from bark of Neem (*A. indica*) tree. The active principle in a methanolic extract of the laboratory-grown cyanobacterium, *Fischerella* sp. isolated from neem (*A. indica*) tree

bark found to be active against *Mycobacterium tuberculosis*, *Entero bacteraerogenes*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Escherichia coli* as well as three multi-drug resistant *E. coli* strains *in vitro* assays. Antimicrobial activity was evaluated by using the slightly modified Kirby Bauer Disk Diffusion Susceptibility Method. These findings also supports the results of (Khan, and Wassilew, 1987) which they demonstrated that the extracts of neem leaf, neem oil and seed kernels are effective against certain human fungi, including *Trichophyton*, *Epidermophyton*, *Microsporum*, *Trichosporon*, *Geotricum* and Candida. High antimycotic activity with extracts of different parts of neem has already been reported (Jacobson, 1986). Oil from the leaves, seeds and bark possesses a wide spectrum of antibacterial action against Gram-negative and Gram-positive microorganisms, including *M*. *tuberculosis* and streptomycin resistant strains (Chopra *et al*., 1952). *In vitro*, it inhibits *Vibrio cholerae*, *Klebsiella pneumoniae*, *M. tuberculosis* and *M*. pyogenes (Satyavati *et al*., 1976).

These results are in agreement with the results of (Uwimbabazi *et al*., 2015) which they explained that the assessment of antibacterial activity of neem plant (*A. indica*) on *Staphylococcus aureus* and *Escherichia coli*. The results obtained after experiment showed that *S. aureus* strains responded differently to both ethanol and aqueous leaf extracts. On this strain they used both dried and fresh leaves and the comparison was done based on the inhibition zones obtained after incubation. The results shows the

inhibition zones in cm, formed by aqueous and ethanol extracts from fresh *A. indica* leaves.

The plant *A. indica* leaf, stem bark, root wood and seed shows alkaloids, carbohydrates, flavanoids, glycosides, phenol, protein, resins, saponnins, tannins and sterols of different solvents. The phytochemical test results indicated high scores for saponins, moderate scores for tannins and glycosides while alkaloids, terpenes and flavanoids had low scores. According to Anyanwu and Dawet (2005) these constituents found in plants are known to have anti protozoal and anti bacterial activities. Flavonoids especially, are of a potential benefit to human health (Jouad *et al.*, 2001).

These results are in agreement with the results of Emran *et al*., 2015 which they demonstrated that the results of Phytochemical, Antimicrobial, Cytotoxic, Analgesic and Anti-Inflammatory Properties of *A. indica*: A Therapeutic Study. In this study the ethanol extract, n-hexane extract and chloroform extract of *A. indica* were first evaluated for phytochemical study. The phytochemical screening of the three extracts of *A. indica* exhibited the presence of important secondary metabolites such as flavanoids, terpenoids, steroids and tannins.

These findings also support the results of Biu *et al*., 2009 which they explained the results of Phytochemical screening of *Azadirachta indica* (Neem) (Meliaceae) in Maiduguri, Nigeria. In this study, Saponins had high scores in the extract, tannins and glycosides indicated moderate scores, while alkaloids, terpenes,

flavanoids, reducing sugars, pentoses and whole carbohydrates showed low scores. Anthraquinones, ketones and monosaccharides were not detected from the extract. It was concluded that the extract contains pharmacologically active constituents.

Azadirachtin is a mixture of seven isomeric compounds as Azadirachtin-A to Azadirachtin-G of which Azadirachtin-E is the most effective insect growth regulator (Verkerk and Wright, 1993). Azadirachtin possess insecticidal, ovicidal, antifeedant and growth inhibiting effects against many insect pests (Akou-Edi, 1984; Schmutterer, 1990; Vietmeyer, 1992; Nawrot and Harmatha, 1994), including the storage pests (Jilani and Su, 1983; Ivbijaro, 1983a, b; Makanjuola, 1989).

The present findings also fit well with those of (chourasiya et al 2012) who demonstrated the isolation of quercetin- from the leaves of *Azadirachta indica* and antidiabetic study of the crude extracts.

The Azadirachta extractives also screened for nematicidal activity by the previous workers. The aqueous extract of the Azadirachta leaf was evaluated in the laboratory against the tea red spider mite (Oligonychus coffeae) by directly spraying the test material onto the leaf discs. Bioefficacy, ovicidal and adulticidal action and ovipositional deterrence of the test plant were then confirmed by (Roobakkumar *et al*., 2010).

Azadirachtin extracts from the seeds, leaves and bark of the Neem tree has been reported to have strong biological activities against insect pests, but with very low toxicity to mammals and the

environment, generally (Umar *et al*., 2002; Makeri *et al*., 2007; Wikipedia, 2007). Registered Neem insecticide formulations Neemros® and Neemroc EC® have also been found to be effective against insect pests of vegetables but safe to their natural enemies (Akol *et al.*, 2001).

Therefore, the wide use of the neem plant is attributable to the presence of these bioactive compounds, which may explain its many traditional uses against various ailments. Further research is recommended on this as a confirmation.

Some earlier workers reported various plant materials including essential oils in checking the multiplication of pests in stores (Krishnamurti and Rao, 1944; Su *et al.,* 1972; Sangapa, 1977). The pongam oil from the seeds has already been marketed as karanjin biopesticide by SOM Phytopharma (India) Limited [http://www.somphyto.com], along with annonin, and azadirachtin. Toxicological effects of neem (*Azadirachta indica*) was marked by **(**Hameed *et al***.,** 2012).The Pepper Research Station, Panniyur, India carried out an experiment on a mixture of pongam oil and neem-seed oil to apply as a biopesticide against *C. maculatus* and the SAARC Documentation Center, India recorded it as an excellent result.

So, insecticidal potentials of *A. indica* or of its related species needs no further proof, while certain other experimental results carried out by different researchers worldwide mentioned here,

were tested against different other multicellular organisms, viz. nematodes, mites and brine shrimp nauplii.

So far the results of the present investigation concern the biological activity of *A. indica* is of course promising, which was further authenticated by the outcomes of the previous researches around the world discussed in this chapter. Bangladesh being the homeland of this famous plant might have a bright future of earning foreign currency by exporting different products or preparations of this plant. The seeds offered the highest toxicity to the majority of the test agents, and 100% highest activity to the multicellular test organisms; followed by the root extracts. So, seeds could be an export item, as well as the root products (for what there is an international market), and easy formulation of the products is necessary while the folk use of the *Azadirachta* products given hints in this regard.

Through isolation of the bioactive substances of the most active parts it was attempted to find the active ingredients for biodegradable insecticides, but the amounts yielded were so little, and it was then hard to carry out all the bioactivity tests with the purified compounds. To achieve the goal with much success in proper utilization of this promising plant and for the marketing of its products it is necessary to launch a huge investigation for further substantial empirical assessments.

Conclusion

Neem, the versatile medicinal plant is the unique source of various types of compounds having diverse chemical structure. Very little work has been done on the biological activity and plausible medicinal applications of these compounds and hence extensive investigation is needed to exploit their therapeutic utility to combat diseases. A drug-development programme should be undertaken to develop modern drugs with the compounds isolated from neem. Although crude extracts from various parts of neem have medicinal applications from time immemorial, modern drugs can be developed after extensive investigation of its bioactivity, mechanism of action, pharmaco therapeutics, toxicity and after proper standardization and clinical trials. As the global scenario is now changing towards the use of nontoxic plant products having traditional medicinal use, development of modern drugs from neem should be emphasized for the control of various diseases. In fact, time has come to make good use of centuries-old knowledge on neem through modern approaches of drug development. For the last few years, there has been an increasing trend and awareness in neem research. Quite a significant amount of research has already been carried out during the past few decades in exploring the chemistry of different parts of neem. Several therapeutically and industrially useful preparations and compounds have also been marketed, which generates enough encouragement among the scientists in exploring more information about this medicinal plant. An extensive research and development work should be undertaken on neem and its products for their better economic and therapeutic utilization.

Appendix Table 1: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	5	16.67	17	4.05	4.044	4.037	13.17	4.046
2123.142	3.326	30	5	16.67	17	4.05	3.992	4.062	12.15	3.993
1415.428	3.150	30	4	13.34	13	3.87	3.918	3.878	12.15	3.920
707.714	2.850	30	3	10.00	10	3.72	3.791	3.72	10.08	3.794
353.857	2.548	30	3	10.00	10	3.72	3.791	3.72	10.08	3.794

Results:

Y = 2.602808 + 0.4181305 X

Chi-squared is 0.1691845 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is5.733119

LD_{50} is 540902.7µg/cm^2

95% confidence limits are 3.37315 to 8.673639E+10 µg/cm^2

Appendix Table 2: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	13	43.33	43	4.82	4.815	4.838001	18.81	4.816
2123.142	3.326	30	12	40.00	40	4.75	4.758	4.74	18.48	4.757
1415.428	3.150	30	11	36.67	37	4.67	4.677	4.659	18.03	4.674
707.714	2.850	30	10	33.33	33	4.56	4.538	4.544	17.43	4.532
353.857	2.548	30	8	26.66	27	4.39	4.400	4.39	16.74	4.389

Results:

Y = 3.185217 + 0.4726539 X

Chi-squared is 2.133322E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.83956

LD_{50} is 6911.297µg/cm^2

95% confidence limits are 530.8342 to 89983.02 µg/cm^2

Appendix Table 3: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	17	56.67	57	5.18	5.130	5.165	19.02	5.123
2123.142	3.326	30	16	53.33	53	5.08	5.072	5.075	19.11	5.066
1415.428	3.150	30	14	46.67	47	4.92	4.989	4.915	19.02	4.985
707.714	2.850	30	13	43.33	43	4.82	4.849	4.838	18.81	4.848
353.857	2.548	30	12	40.00	40	4.75	4.708	4.74	18.48	4.710

Results:

Y = 3.546648 + 0.4566106 X

Chi-squared is 0.1479325 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.182913

LD_{50} is 1523.749µg/cm^2

95% confidence limits are519.1728 to 4472.133 µg/cm^2

Appendix Table 4: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	21	70	70	5.52	5.499	5.51	18.03	5.488
2123.142	3.326	30	20	66.66	67	5.44	5.437	5.429	18.03	5.429
1415.428	3.150	30	19	63.33	63	5.33	5.352	5.318	18.48	5.346
707.714	2.850	30	17	56.67	57	5.18	5.204	5.202	18.81	5.205
353.857	2.548	30	16	53.33	53	5.08	5.056	5.075	19.11	5.063

Results:

Y = 3.863903 + 0.4706527 X

Chi-squared is 2.660871E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.413875

LD_{50} is 259.3435µg/cm^2

95% confidence limits are 28.99703 to 2319.515 µg/cm^2

Appendix Table 5: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	5	16.67	17	4.05	3.969	4.062	12.15	3.981
2123.142	3.326	30	4	13.34	13	3.87	3.907	3.878	12.15	3.916
1415.428	3.150	30	3	10.00	10	3.72	3.818	3.72	11.1	3.824
707.714	2.850	30	3	10.00	10	3.72	3.668	3.73	9.060	3.667
353.857	2.548	30	2	6.67	7	3.52	3.517	3.519	8.07	3.510

Results:

Y = 2.180794 + 0.5215927 X

Chi-squared is 0.2537117 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.404997

LD_{50} is 254095.2µg/cm^2

95% confidence limits are 38.5011 to 1.676952E+09µg/cm^2

Appendix Table 6: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	9	30	30	4.48	4.449	4.48	16.74	4.452
2123.142	3.326	30	8	26.67	27	4.39	4.419	4.39	16.74	4.421
1415.428	3.150	30	8	26.66	27	4.39	4.377	4.394	15.96	4.377
707.714	2.850	30	7	23.34	23	4.26	4.304	4.266	15.96	4.302
353.857	2.548	30	7	23.33	23	4.26	4.231	4.252	15.09	4.227

Results:

Y = 3.593121 + 0.2488788 X

Chi-squared is 6.374812E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.652869

LD_{50} is 449643.3µg/cm^2

95% confidence limits are 32.5642+3.452E+6 µg/cm^2

Appendix Table 7: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	14	46.67	47	4.92	4.887	4.942	18.81	4.904
2123.142	3.326	30	13	43.33	43	4.82	4.840	4.838	18.81	4.852
1415.428	3.150	30	12	40.00	40	4.75	4.776	4.74	18.48	4.780
707.714	2.850	30	11	36.67	37	4.67	4.664	4.659	18.03	4.655
353.857	2.548	30	10	33.33	33	4.56	4.552	4.544	17.43	4.530

Results:

Y = 3.474663 + 0.4142152 X

Chi-squared is 6.361366E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.682475

LD_{50} is 4813.655µg/cm^2

95% confidence limits are 432.4115 to 53586.13 µg/cm^2

Appendix Table 8: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	18	60	60	5.25	5.185	5.24	19.02	5.173
2123.142	3.326	30	16	53.33	53	5.08	5.146	5.065	19.02	5.135
1415.428	3.150	30	16	53.33	53	5.08	5.092	5.075	19.11	5.082
707.714	2.850	30	15	50	50	5.00	4.999	4.99	19.02	4.991
353.857	2.548	30	14	46.	47	4.92	4.907	4.915	19.02	4.901

Results:

Y = 4.133716 + 0.3011669 X

Chi-squared is .1843769 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.876425

LD_{50} is 752.3578µg/cm^2

95% confidence limits are 127.7164 to 4432.026 µg/cm^2

Appendix Table 9. Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	8	26.67	27	4.39	4.379	4.394	15.96	4.379
2123.142	3.326	30	7	23.33	23	4.26	4.310	4.266	15.96	4.309
1415.428	3.150	30	7	23.33	23	4.26	4.213	4.252	15.09	4.212
707.714	2.850	30	5	16.66	17	4.05	4.046	4.037	13.17	4.044
353.857	2.548	30	4	13.33	13	3.87	3.880	3.873	11.1	3.876

Results:

Y = 2.457657 + 0.5567423 X

Chi-squared is 5.935478E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.566465

LD_{50} is 36852.30 µg/cm^2

95% confidence limits are 385.3198 to 3524579µg/cm^2

Appendix Table 10. Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	17	56.67	57	5.18	5.132	5.165	19.02	5.130
2123.142	3.326	30	16	53.33	53	5.08	5.017	5.075	19.11	5.012
1415.428	3.150	30	12	40.00	40	4.75	4.854	4.76	18.81	4.847
707.714	2.850	30	9	30.00	30	4.48	4.576	4.46	17.43	4.564
353.857	2.548	30	8	26.66	27	4.39	4.299	4.388	15.09	4.282

Results:

Y = 1.889938 + 0.9385532 X

Chi-squared is 0.6018476 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.313677

LD_{50} is 2059.099 µg/cm^2

95% confidence limits are 1108.395 to 3825.246µg/cm^2

Appendix Table 11. Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	21	70	70	5.52	5.435	5.51	18.03	5.422
2123.142	3.326	30	19	63.33	63	5.33	5.334	5.318	18.48	5.321
1415.428	3.150	30	16	53.33	53	5.08	5.190	5.065	19.02	5.177
707.714	2.850	30	14	46.66	47	4.92	4.943	4.915	19.02	4.933
353.857	2.548	30	12	40.00	40	4.75	4.697	4.74	18.03	4.688

Results:

Y = 2.614321 + 0.813474 X

Chi-squared is 0.4344726 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.932705

LD_{50} is 856.4559 µg/cm^2

95% confidence limits are 459.4218 to 1596.608µg/cm^2

Appendix Table 12. Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	24	80	80	5.85	5.754	5.83	15.96	5.731
2123.142	3.326	30	22	73.33	73	5.61	5.683	5.61	16.74	5.665
1415.428	3.150	30	21	70	70	5.52	5.582	5.5	17.43	5.573
707.714	2.850	30	20	66.66	67	5.44	5.410	5.429	18.03	5.416
353.857	2.548	30	18	60	60	5.25	5.238	5.28	18.81	5.258

Results:

Y = 3.925557 + 0.5230322 X

Chi-squared is 0.3142581 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.054258

LD_{50} is 113.3073 µg/cm^2

95% confidence limits are 6.037319 to 2126.529µg/cm^2

Appendix Table 13. Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	4	13.33	13	3.87	3.883	3.873	11.1	3.886
2123.142	3.326	30	4	13.33	13	3.87	3.837	3.873	11.1	3.840
1415.428	3.150	30	3	10.00	10	3.72	3.772	3.72	10.08	3.774
707.714	2.850	30	3	10.00	10	3.72	3.771	3.72	10.08	3.775
353.857	2.548	30	2	6.666	7	3.52	3.548	3.519	8.07	3.552

Results:

Y = 2.609282 + 0.3699346 X

Chi-squared is 9.324336E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.462543

LD_{50} is 29009.69 µg/cm^2

95% confidence limits are 2.899154E-02 to 2.902783E+14µg/cm^2

Appendix Table 14. Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	12	40.00	40	4.75	4.673	4.74	18.03	4.662
2123.142	3.326	30	10	33.33	33	4.56	4.613	4.551	18.03	4.603
1415.428	3.150	30	9	30.00	30	4.48	4.528	4.46	17.43	4.519
707.714	2.850	30	8	26.66	27	4.39	4.384	4.394	15.96	4.376
353.857	2.548	30	7	23.34	23	4.26	4.240	4.252	15.09	4.233

Results:

Y = 3.021963 + 0.4752018 X

Chi-squared is 0.2290532 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.16252

LD_{50} is 14538.5 µg/cm^2

95% confidence limits are 399.2724 to 529383µg/cm^2

Appendix Table 15. Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	18	60.00	60	5.25	5.174	5.24	19.02	5.168
2123.142	3.326	30	16	53.33	53	5.08	5.096	5.075	19.11	5.090
1415.428	3.150	30	14	46.66	47	4.92	4.987	4.915	19.02	4.979
707.714	2.850	30	12	40.00	40	4.75	4.799	4.74	18.48	4.790
353.857	2.548	30	11	36.67	37	4.67	4.612	4.659	18.03	4.600

Results:

Y = 2.997115 + 0.629013 X

Chi-squared is 0.28724 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.184172

LD_{50} is 1528.169 µg/cm^2

95% confidence limits are 698.0223 to 3345.597µg/cm^2

Appendix Table 16. Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	21	70.00	70	5.52	5.487	5.51	18.03	5.475
2123.142	3.326	30	20	66.66	67	5.44	5.413	5.429	18.03	5.402
1415.428	3.150	30	18	60.00	60	5.25	5.308	5.24	18.48	5.297
707.714	2.850	30	16	53.33	53	5.08	5.130	5.065	19.02	5.118
353.857	2.548	30	15	50.00	50	5.00	4.950	4.99	19.02	4.940

Results:

Y = 3.426053 + 0 .5938126 X

Chi-squared is 0.1975682 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.650579

LD_{50} is 447.2792 µg/cm^2

95% confidence limits are 127.1399 to 1573.53µg/cm^2

Appendix Table 17. Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	6	20	20	4.16	4.138	4.17	14.13	4.138
2123.142	3.326	30	5	16.67	17	4.05	4.058	4.037	13.17	4.061
1415.428	3.150	30	4	13.33	13	3.87	3.948	3.878	12.15	3.953
707.714	2.850	30	4	13.33	13	3.87	3.758	3.894	10.08	3.767
353.857	2.548	30	2	6.67	7	3.52	3.568	3.519	8.07	3.582

Results

Y = 2.011662 + 0.6160064 X

Chi-squared is 0.2835674 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.851149

LD_{50} is 70982.01µg/cm^2

95% confidence limits are 298.7637 to1.686435E+07 µg/cm^2

Appendix Table 18. Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	10	33.33	33	4.56	4.470	4.57	16.74	4.476
2123.142	3.326	30	8	26.67	27	4.39	4.424	4.39	16.74	4.429
1415.428	3.150	30	7	23.34	23	4.26	4.358	4.266	15.96	4.362
707.714	2.850	30	7	23.33	23	4.26	4.245	4.252	15.09	4.246
353.857	2.548	30	6	20.00	20	4.16	4.132	4.170	14.13	4.131

Results

Y = 3.155897 + 0.3827177 X

Chi-squared is 0.3384838 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.818442

LD_{50} is 65832.731µg/cm^2

95% confidence limits are 46.50053 to 9.320226E+07 µg/cm^2

Appendix Table 19. Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	15	50.00	50	5	4.899	5.02	18.81	4.914
2123.142	3.326	30	13	43.33	43	4.82	4.842	4.838	18.81	4.854
1415.428	3.150	30	11	36.67	37	4.67	4.761	4.662	18.48	4.768
707.714	2.850	30	10	33.33	33	4.56	4.623	4.551	18.03	4.623
353.857	2.548	30	10	33.33	33	4.56	4.485	4.57	16.74	4.477

Results

Y = 3.243517 + 0.4839424 X

Chi-squared is 0.6619892 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.629531

LD_{50} is 4261.189µg/cm^2

95% confidence limits are 618.3115 to 29366.64 µg/cm^2

Appendix Table 20. Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	21	70.00	70	5.52	5.342	5.5	18.48	5.346
2123.142	3.326	30	17	56.67	57	5.18	5.287	5.202	18.81	5.290
1415.428	3.150	30	16	53.33	53	5.08	5.210	5.098	18.81	5.210
707.714	2.850	30	16	53.33	53	5.08	5.077	5.075	19.11	5.076
353.857	2.548	30	15	50.00	50	5.00	4.944	4.99	19.02	4.940

Results

Y = 3.79532 + 0.4492497 X

Chi-squared is 0.8699672 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.681538

LD_{50} is 480.3277µg/cm^2

95% confidence limits are 98.38282 to 2345.072 µg/cm^2

Appendix Table 21. Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	5	16.67	17	4.05	4.009	4.037	13.17	4.006
2123.142	3.326	30	4	13.33	13	3.87	3.943	3.878	12.15	3.942
1415.428	3.150	30	4	13.33	13	3.87	3.851	3.873	11.10	3.851
707.714	2.850	30	3	10.00	10	3.72	3.693	3.73	9.060	3.696
353.857	2.548	30	2	6.66	7.0	3.52	3.535	3.519	8.07	3.541

Results:

Y = 2.22816 + 0.5151049 X

Chi-squared is 8.163643E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 5.381119

LD_{50} is 240501.9µg/cm^2

95% confidence limits are 40.22166 to 1.43806E+09µg/cm^2

Appendix Table 22. Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	9	30.00	30	4.48	4.506	4.46	17.43	4.492
2123.142	3.326	30	8	26.67	27	4.39	4.418	4.39	16.74	4.407
1415.428	3.150	30	8	26.66	27	4.39	4.296	4.388	15.09	4.288
707.714	2.850	30	5	16.66	17	4.05	4.084	4.037	13.17	4.083
353.857	2.548	30	4	13.33	13	3.87	3.874	3.873	11.1	3.878

Results:

Y = 2.146664 + 0.6794698 X

Chi-squared is 0.2034314 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 4.199357

LD_{50} is 15825.49µg/cm^2

95% confidence limits are980.0414 to 255546.5µg/cm^2

Appendix Table 23. Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	15	50.00	50	5	4.980	4.99	19.02	4.983
2123.142	3.326	30	14	46.66	47	4.92	4.886	4.942	18.81	4.888
1415.428	3.150	30	12	40.00	40	4.75	4.754	4.74	18.48	4.753
707.714	2.850	30	8	26.67	27	4.39	4.528	4.376	17.43	4.522
353.857	2.548	30	8	26.66	27	4.39	4.301	4.394	15.96	4.292

Results:

Y = 2.338804 + 0.766129 X

Chi-squared is 0.5990839 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.473561

LD_{50} is 2975.506µg/cm^2

95% confidence limits are 1124.038 to 7876.648 µg/cm^2

Appendix Table 24. Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	18	60.00	60	5.25	5.248	5.28	18.81	5.258
2123.142	3.326	30	17	56.66	57	5.18	5.180	5.165	19.02	5.186
1415.428	3.150	30	16	53.33	53	5.08	5.083	5.075	19.11	5.084
707.714	2.850	30	14	46.66	47	4.92	4.916	4.915	19.02	4.910
353.857	2.548	30	12	40.00	40	4.75	4.751	4.74	18.48	4.737

Results:

Y = 3.268065 + 0.5763386 X

Chi-squared is 1.968241E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.005066

LD_{50} is 1011.733µg/cm^2

95% confidence limits are 445.3582 to 2298.383 µg/cm^2

Appendix Table 33: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	11	36.66	37	4.67	4.635	4.659	18.03	4.620
2123.142	3.326	30	10	33.33	33	4.56	4.582	4.544	17.43	4.569
1415.428	3.150	30	9	30.00	30	4.48	4.508	4.46	17.43	4.496
707.714	2.850	30	8	26.66	27	4.39	4.380	4.394	15.96	4.372
353.857	2.548	30	7	23.33	23	4.26	4.254	4.252	15.09	4.248

Results:

Y = 3.19942 + 0.4117216 X

Chi-squared is 6.883776E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.373296

LD_{50} is 23620.85µg/cm^2

95% confidence limits are 172.8482 to 3227946µg/cm^2

Appendix Table 34: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	18	60.00	60	5.25	5.202	5.28	18.81	5.214
2123.142	3.326	30	17	56.66	57	5.18	5.132	5.165	19.02	5.142
1415.428	3.150	30	14	46.66	47	4.92	5.032	4.925	19.11	5.038
707.714	2.850	30	13	43.33	43	4.82	4.862	4.838	18.81	4.862
353.857	2.548	30	12	40.00	40	4.75	4.691	4.74	18.03	4.686

Results:

Y = 3.193704 + 0.5855628 X

Chi-squared is 0.4004541 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.084718

LD_{50} is 1215.395µg/cm^2

95% confidence limits are 547.9739 to 2695.722µg/cm^2

Appendix Table 35: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	27	90	90	6.28	6.178	6.27	12.15	6.125
2123.142	3.326	30	26	86.67	87	6.13	66.004	6.087	13.17	5.958
1415.428	3.150	30	21	70	70	5.52	5.757	5.51	15.96	5.722
707.714	2.850	30	17	56.66	57	5.18	5.335	5.162	18.48	5.319
353.857	2.548	30	16	53.33	53	5.08	4.914	5.065	19.02	4.916

Results:

Y = 1.504299 + 1.338637 X

Chi-squared is 2.070941 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.611389

LD_{50} is 408.6851µg/cm^2

95% confidence limits are 224.8994 to 742 µg/cm^2

Appendix Table 36. Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	29	96.66	97	6.88	6.576	6.759	8.07	6.584
2123.142	3.326	30	28	93.33	93	6.48	6.406	6.491	9.060	6.414
1415.428	3.150	30	25	83.33	83	5.95	6.168	5.948	12.15	6.175
707.714	2.850	30	23	76.66	77	5.74	5.760	5.734	15.96	5.766
353.857	2.548	30	20	66.66	67	5.44	5.353	5.422	18.48	5.358

Results:

Y = 1.900698 + 1.35663 X

Chi-squared is 1.020651 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 0.254365

LD_{50} is 192.5573µg/cm^2

95% confidence limits are 74.1513 to 500.036µg/cm^2

Appendix Table 37: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	6	20.00	20	4.16	4.176	4.17	14.13	4.180
2123.142	3.326	30	6	20.00	20	4.16	4.138	4.17	14.13	4.142
1415.428	3.150	30	5	16.67	17	4.05	4.084	4.037	13.17	4.089
707.714	2.850	30	5	16.67	17	4.05	3.992	4.062	12.15	3.998
353.857	2.548	30	4	13.33	13	3.87	3.900	3.878	12.15	3.906

Results:

Y = 3.134514 + 0.3029002 X

Chi-squared is 0.1079621 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.158749

LD_{50} is 14412.80 µg/cm^2

95% confidence limits are 3.770656E-02 to 5.509097E+13µg/cm^2

Appendix Table 38: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	13	43.33	43	4.82	4.780	4.818	18.48	4.775
2123.142	3.326	30	12	40.00	40	4.75	4.703	4.74	18.48	4.696
1415.428	3.150	30	9	30.00	30	4.48	4.593	4.46	17.43	4.585
707.714	2.850	30	8	26.67	27	4.39	4.406	4.39	16.74	4.396
353.857	2.548	30	7	23.34	23	4.26	4.218	4.252	15.09	4.206

Results:

Y = 2.602638 + 0.6293048 X

Chi-squared is 0.3760615 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.809541

LD_{50} is 6449.728 µg/cm^2

95% confidence limits are 969.3614 to 42913.73E+13µg/cm^2

Appendix Table 39: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	20	66.67	67	5.44	5.267	5.462	18.81	5.275
2123.142	3.326	30	17	56.67	57	5.18	5.181	5.165	19.02	5.187
1415.428	3.150	30	13	43.33	43	4.82	5.060	4.825	19.11	5.064
707.714	2.850	30	13	43.33	43	4.82	4.854	4.838	18.81	4.853
353.857	2.548	30	12	40.00	40	4.75	4.647	4.74	18.03	4.642

Results:

Y = 2.853815 + 0.7014981 X

Chi-squared is 1.936571 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.059431

LD_{50} is 1146.651 µg/cm^2

95% confidence limits are 589.842 to 2229.087µg/cm^2

Appendix Table 40: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	23	76.67	77	5.74	5.596	5.696	17.43	5.564
2123.142	3.326	30	20	66.67	67	5.44	5.508	5.416	17.43	5.477
1415.428	3.150	30	18	60.00	60	5.25	5.383	5.24	18.48	5.358
707.714	2.850	30	17	56.67	57	5.18	5.168	5.165	19.02	5.155
353.857	2.548	30	15	50.00	50	5.00	4.954	4.99	19.02	4.950

Results:

Y = 3.223551 + 0.6776109 X

Chi-squared is 0.668766 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.621636

LD_{50} is 418.4427µg/cm^2

95% confidence limits are 132.3817 to 1322.646µg/cm^2

Appendix Table 25. Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	9	30	30	4.48	4.451	4.48	16.74	4.456
2123.142	3.326	30	8	26.66	27	4.39	4.398	4.394	15.96	4.403
1415.428	3.150	30	7	23.33	23	4.26	4.324	4.266	15.96	4.328
707.714	2.850	30	7	23.33	23	4.26	4.196	4.284	14.13	4.199
353.857	2.548	30	5	16.66	17	4.05	4.070	4.037	13.17	4.071

Results:

Y = 2.981806 + 0.4272701 X

Chi-squared is 0.1880722 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.723463

LD_{50} is 52900.91µg/cm^2

95% confidence limits are 99.69377 to 2.807107E+07µg/cm^2

Appendix Table26. Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	15	50	50	5.00	4.986	4.99	19.02	4.986
2123.142	3.326	30	14	46.66	47	4.92	4.928	4.915	19.02	4.927
1415.428	3.150	30	13	43.33	43	4.82	4.848	4.838	18.81	4.845
707.714	2.850	30	12	40.00	40	4.75	4.712	4.740	18.48	4.705
353.857	2.548	30	10	33.33	33	4.56	4.574	4.544	17.43	4.564

Results:

Y = 3.374793 + 0.4667058 X

Chi-squared is 3.478241E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.482295

LD_{50} is 3035.954µg/cm^2

95% confidence limits are 607.318 to15176.57µg/cm^2

Appendix Table 27. Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	23	76.66	77	5.74	5.635	5.73	16.74	5.618
2123.142	3.326	30	21	70.00	70	5.52	5.549	5.50	17.43	5.536
1415.428	3.150	30	19	63.33	63	5.33	5.428	5.321	18.03	5.420
707.714	2.850	30	17	56.67	57	5.18	5.222	5.202	18.81	5.223
353.857	2.548	30	16	53.33	53	5.08	5.015	5.075	19.11	5.025

Results:

Y = 3.352186 + 0.6564545 X

Chi-squared is 0.4663468 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 2.510173

LD_{50} is 323.7229µg/cm^2

95% confidence limits are 80.55176 to 1300.982µg/cm^2

Appendix Table 28. Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	26	86.67	87	6.13	6.010	6.087	13.17	5.989
2123.142	3.326	30	24	80.00	80	5.85	5.921	5.87	14.13	5.902
1415.428	3.150	30	23	76.67	77	5.74	5.796	5.734	15.96	5.780
707.714	2.850	30	21	70.00	70	5.52	5.583	5.5	17.43	5.570
353.857	2.548	30	20	66.67	67	5.44	5.369	5.422	18.48	5.361

Results:

Y = 3.588591 + 0.6954293 X

Chi-squared is 0.3290119with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 2.029551

LD_{50} is 107.0412µg/cm^2

95% confidence limits are 10.58678 to 1082.276µg/cm^2

Appendix Table 29. Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	7	23.33	23	4.26	4.215	4.252	15.09	4.216
2123.142	3.326	30	6	20.00	20	4.16	4.159	4.17	14.13	4.160
1415.428	3.150	30	5	16.67	17	4.05	4.080	4.037	13.17	4.080
707.714	2.850	30	4	13.33	13	3.87	3.945	3.878	12.15	3.945
353.857	2.548	30	4	13.33	13	3.87	3.810	3.873	11.1	3.808

Results:

Y = 2.657667 + 0.4516222 X

Chi-squared is 0.1452589 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.186488

LD_{50} is 153634.4µg/cm^2

95% confidence limits are 46.15917 to 5.113498E+08 µg/cm^2

Appendix Table 30. Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	12	40.00	40	4.75	4.736	4.74	18.48	4.726
2123.142	3.326	30	11	36.67	37	4.67	4.668	4.659	18.03	4.658
1415.428	3.150	30	10	33.33	33	4.56	4.572	4.544	17.43	4.563
707.714	2.850	30	8	26.67	27	4.39	4.408	4.39	16.74	4.400
353.857	2.548	30	7	23.33	23	4.26	4.244	4.252	15.09	4.237

Results:

Y = 2.858917 + 0 .5408117 X

Chi-squared is 1.526594E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.959018

LD_{50} is 9099.51µg/cm^2

95% confidence limits are 675.918 to122501.8 µg/cm^2

Appendix Table 31. Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	17	56.67	57	5.18	5.144	5.165	19.02	5.132
2123.142	3.326	30	16	53.33	53	5.08	5.097	5.075	19.11	5.088
1415.428	3.150	30	15	50.00	50	5.00	5.032	5.00	19.11	5.028
707.714	2.850	30	14	46.67	47	4.92	4.920	4.915	19.02	4.924
353.857	2.548	30	13	43.33	43	4.82	4.807	4.838	18.81	4.820

Results:

Y = 3.942465 + 0.344479 X

Chi-squared is 4.725695E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.069955

LD_{50} is 1174.775µg/cm^2

95% confidence limits are 306.4728 to 4503.159 µg/cm^2

Appendix Table 32. Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.856	3.452	30	21	70	70	5.52	5.482	5.51	18.03	5.471
2123.142	3.326	30	20	66.67	67	5.44	5.425	5.429	18.03	5.418
1415.428	3.150	30	18	60.00	60	5.25	5.346	5.24	18.48	5.343
707.714	2.850	30	18	60.00	60	5.25	5.211	5.28	18.81	5.214
353.857	2.548	30	16	53.33	53	5.08	5.075	5.075	19.11	5.086

Results

Y = 3.998091 + 0.426866 X

Chi-squared is 0.3080139 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.347128

LD_{50} is 222.3965µg/cm^2

95% confidence limits are 16.25487 to 3042.791 µg/cm^2

Appendix Table 45: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	7	23.33	23	4.26	4.258	4.252	15.09	4.256
3538.571	3.549	30	6	20.00	20	4.16	4.112	4.17	14.13	4.112
2653.928	3.424	30	5	16.66	17	4.05	4.052	4.037	13.17	4.052
1769.286	3.248	30	4	13.33	13	3.87	3.966	3.878	12.15	3.967
884.643	2.947	30	4	13.33	13	3.87	3.820	3.873	11.1	3.823

Results:

Y = 2.408829 + 0.4800213 X

Chi-squared is 0.1759168with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.398034

LD_{50} is 250054.2µg/cm^2

95% confidence limits are 111.5826 to5.603668E+08µg/cm^2

Appendix Table 46: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	14	46.66	47	4.92	4.926	4.915	19.02	4.930
3538.571	3.549	30	13	43.33	43	4.82	4.803	4.838	18.81	4.801
2653.928	3.424	30	12	40.00	40	4.75	4.752	4.74	18.48	4.748
1769.286	3.248	30	11	36.66	37	4.67	4.680	4.659	18.03	4.673
884.643	2.947	30	10	33.33	33	4.56	4.557	4.544	17.43	4.544

Results:

Y = 3.281171 + 0.4284342 X

Chi-squared is 3.414214E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.011885

LD_{50} is 10277.43µg/cm^2

95% confidence limits are 881.0349 to 119888.2µg/cm^2

Appendix Table 47: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	21	70.00	70	5.52	5.504	5.5	17.43	5.498
3538.571	3.549	30	19	63.33	63	5.33	5.254	5.358	18.81	5.252
2653.928	3.424	30	16	53.33	53	5.08	5.150	5.065	19.02	5.149
1769.286	3.248	30	14	46.66	47	4.92	5.004	4.925	19.11	5.004
884.643	2.947	30	13	43.33	43	4.82	4.756	4.818	18.48	4.758

Results:

Y = 2.341571 + 0.820051 X

Chi-squared is 0.5358367 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.241785

LD_{50} is 1744.959µg/cm^2

95% confidence limits are 915.3052 to 3326.627µg/cm^2

Appendix Table 48: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	25	83.33	83	5.95	5.856	5.902	15.09	5.803
3538.571	3.549	30	22	73.33	73	5.61	5.663	5.61	16.74	5.638
2653.928	3.424	30	20	66.66	67	5.44	5.583	5.416	17.43	5.569
1769.286	3.248	30	21	70.00	70	5.52	5.470	5.51	18.03	5.472
884.643	2.947	30	19	63.33	63	5.33	5.278	5.358	18.81	5.306

Results:

Y = 3.687769 + 0 .5494671 X

Chi-squared is 0.6432109 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.388188

LD_{50} is 244.4488µg/cm^2

95% confidence limits are 11.30526 to 5285.62µg/cm^2

Appendix Table 41: Dose-mortality effect of stem bark extract (methanol of *Azadirachta indica* against *T. custaneum* after 24 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	5	16.67	17	4.05	4.015	4.037	13.17	4.013
3538.571	3.549	30	4	13.33	13	3.87	3.901	3.878	12.15	3.901
2653.928	3.424	30	4	13.33	13	3.87	3.854	3.873	11.1	3.855
1769.286	3.248	30	3	10.00	10	3.72	3.786	3.72	10.08	3.790
884.643	2.947	30	3	10.00	10	3.72	3.673	3.73	9.060	3.678

Results:

Y = 2.58632 + 0.3705477 X

Chi-squared is 9.118784E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.51382

LD_{50} is 32645.24µg/cm^2

95% confidence limits are 0.1466581 to 7.266633E+13µg/cm^2

Appendix Table 42: Dose-mortality effect of stem bark extract (methanol of *Azadirachta indica* against *T. custaneum* after 48 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	9	30.00	30	4.48	4.470	4.48	16.74	4.471
3538.571	3.549	30	8	26.67	27	4.39	4.362	4.394	15.96	4.364
2653.928	3.424	30	7	23.33	23	4.26	4.317	4.266	15.96	4.320
1769.286	3.248	30	7	23.33	23	4.26	4.254	4.252	15.09	4.257
884.643	2.947	30	6	20.00	20	4.16	4.146	4.17	14.13	4.150

Results:

Y = 3.101123 + 0.3558062 X

Chi-squared is 6.792498E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.336828

LD_{50} is 217184.2µg/cm^2

95% confidence limits are 19.47927 to 2.421494E+09µg/cm^2

Appendix Table 43: Dose-mortality effect of stem bark extract (methanol of *Azadirachta indica* against *T. custaneum* after 72 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	16	53.33	53	5.08	4.977	5.065	19.02	4.974
3538.571	3.549	30	12	40.00	40	4.75	4.847	4.76	18.81	4.840
2653.928	3.424	30	12	40.00	40	4.75	4.793	4.74	18.48	4.784
1769.286	3.248	30	11	36.67	37	4.67	4.716	4.662	18.48	4.706
884.643	2.947	30	11	36.67	37	4.67	4.586	4.656	17.43	4.572

Results:

Y = 3.263176 + 0.444474 X

Chi-squared is 0.4730903 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.907595

LD_{50} is 8083.415µg/cm^2

95% confidence limits are 1045.788 to 62480.82µg/cm^2

Appendix Table 44: Dose-mortality effect of stem bark extract (methanol of *Azadirachta indica* against *T. custaneum* after 96 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	19	63.33	63	5.33	5.324	5.318	18.48	5.333
3538.571	3.549	30	18	60.00	60	5.25	5.243	5.28	18.81	5.248
2653.928	3.424	30	17	56.66	57	5.18	5.209	5.202	18.81	5.214
1769.286	3.248	30	17	56.66	57	5.18	5.162	5.165	19.02	5.164
884.643	2.947	30	16	53.33	53	5.08	5.080	5.075	19.11	5.080

Results:

Y = 4.254948 + 0.2800412 X

Chi-squared is 2.562422E-02with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.660511

LD_{50} is 457.6257µg/cm^2

95% confidence limits are 5.592158 to 37449.12µg/cm^2

Appendix Table 53: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 24 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	6	20	20	4.16	4.165	4.17	14.13	4.162
3538.571	3.549	30	5	16.67	17	4.05	4.078	4.037	13.17	4.078
2653.928	3.424	30	5	16.67	17	4.05	4.042	4.037	13.17	4.042
1769.286	3.248	30	5	16.66	17	4.05	3.990	4.062	12.15	3.992
884.643	2.947	30	4	13.33	13	3.87	3.904	3.878	12.15	3.907

Results:

Y = 3.074816 + 0.282503 X

Chi-squared is 9.210211E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.814739

LD_{50} is 65273.73µg/cm^2

95% confidence limits are 1.224582E-03 to 3.479271E+16 µg/cm^2

Appendix Table 54: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 48 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	12	40	40	4.75	4.862	4.76	18.81	4.863
3538.571	3.549	30	13	43.33	43	4.82	4.670	4.821	18.03	4.670
2653.928	3.424	30	11	36.66	37	4.67	4.591	4.656	17.43	4.590
1769.286	3.248	30	8	26.66	27	4.39	4.479	4.39	16.74	4.477
884.643	2.947	30	7	23.33	23	4.26	4.288	4.252	15.09	4.284

Results:

Y = 2.3939 + 0.6413751 X

Chi-squared is 0.829187 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.063301

LD_{50} is 11569.14µg/cm^2

95% confidence limits are 1927.985 to 69422.27µg/cm^2

Appendix Table 55: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 72 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	17	56.67	57	5.18	5.213	5.202	18.81	5.227
3538.571	3.549	30	16	53.33	53	5.08	5.026	5.075	19.11	5.032
2653.928	3.424	30	15	50.00	50	5.00	4.949	4.99	19.02	4.950
1769.286	3.248	30	12	40.00	40	4.75	4.840	4.76	18.81	4.836
884.643	2.947	30	11	36.66	37	4.67	4.653	4.659	18.03	4.640

Results:

Y = 2.728425 + 0.6490146 X

Chi-squared is 0.1924589 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.500037

LD_{50} is 3162.549µg/cm^2

95% confidence limits are 1491.555 to 6705.562 µg/cm^2

Appendix Table 56: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *T. custaneum* after 96 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	22	73.33	73	5.61	5.593	5.584	17.43	5.565
3538.571	3.549	30	19	63.33	63	5.33	5.474	5.321	18.03	5.459
2653.928	3.424	30	21	70.00	70	5.52	5.424	5.51	18.03	5.415
1769.286	3.248	30	20	66.67	67	5.44	5.354	5.422	18.48	5.353
884.643	2.947	30	17	56.67	57	5.18	5.234	5.202	18.81	5.246

Results:

Y = 4.205425 + 0.3532397 X

Chi-squared is 0.6384363 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.249394

LD_{50} is 177.58µg/cm^2

95% confidence limits are 0.905008 to 34844.66µg/cm^2

Appendix Table 49: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 24 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	12	40	40	4.75	4.711	4.74	18.48	4.704
3538.571	3.549	30	10	33.33	33	4.56	4.548	4.544	17.43	4.542
2653.928	3.424	30	8	26.67	27	4.39	4.481	4.39	16.74	4.475
1769.286	3.248	30	8	26.67	27	4.39	4.386	4.394	15.96	4.380
884.643	2.947	30	7	23.33	23	4.26	4.224	4.252	15.09	4.218

Results:

Y = 2.630763 + 0.5386361 X

Chi-squared is 0.1649764 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.398585

LD_{50} is 25037.14µg/cm^2

95% confidence limits are 1119.11 to 560141µg/cm^2

Appendix Table 50: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 48 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	17	56.66	57	5.18	5.196	5.165	19.02	5.187
3538.571	3.549	30	16	53.33	53	5.08	5.10	5.075	19.11	5.003
2653.928	3.424	30	14	46.66	47	4.92	4.933	4.915	19.02	4.927
1769.286	3.248	30	12	40	40	4.75	4.824	4.76	18.81	4.820
884.643	2.947	30	11	36.67	37	4.67	4.638	4.659	18.03	4.636

Results:

Y = 2.841155 + 0.6092778 X

Chi-squared is 0.1863411 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.543285

LD_{50} is 3493.696 µg/cm^2

95% confidence limits are 1511.623 to 8074.704µg/cm^2

Appendix Table 51: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 72 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	27	90	90	6.28	6.154	6.27	12.15	6.113
3538.571	3.549	30	23	76.67	77	5.74	5.776	5.734	15.96	5.754
2653.928	3.424	30	21	70	70	5.52	5.620	5.52	16.74	5.604
1769.286	3.248	30	18	60	60	5.25	5.399	5.24	18.48	5.394
884.643	2.947	30	17	56.66	57	5.18	5.022	5.175	19.11	5.035

Results:

Y = 1.517816 + 1.193606 X

Chi-squared is 1.239829 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.917365

LD_{50} is 826.733 µg/cm^2

95% confidence limits are 388.0266 to 1761.442µg/cm^2

Appendix Table 52: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *T. custaneum* after 96 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.141	3.850	30	29	96.67	97	6.88	6.922	6.844	4.62	6.915
3538.571	3.549	30	28	93.33	93	6.48	6.426	6.491	9.060	6.417
2653.928	3.424	30	27	90.00	90	6.28	6.220	6.23	11.10	6.211
1769.286	3.248	30	24	80.00	80	5.85	5.930	5.87	14.13	5.920
884.643	2.947	30	20	66.67	67	5.44	5.433	5.429	18.03	5.421

Results:

Y = 0.5471034 + 1.654101 X

Chi-squared is 0.1123829 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.692034

LD_{50} is 492.0781µg/cm^2

95% confidence limits are 210.316 to 1151.319µg/cm^2

Appendix Table 267: Repellency of *T. castaneum* by seed (chloroform) of *A. indicai* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	7	8	8	8.333	8.00	7.666	7.333	7.666	66.66	60.00	53.32	46.66	53.32	54.70	50.77	46.89	43.05	46.89
		R2	7	6	9	8	8															
		R3	9	9	7	6	7															
125.70	10	R1	6	8	6	6	8	7.666	7.000	6.333	6.333	6.666	53.32	40.00	26.66	26.66	33.32	49.89	39.23	31.05	31.05	35.24
		R2	9	7	7	6	7															
		R3	8	6	6	7	5															
62.86	10	R1	6	7	5	5	6	6.333	6.000	5.666	5.333	5.666	26.66	20.00	13.32	6.66	13.32	31.05	26.56	21.39	14.89	21.39
		R2	7	6	6	5	6															
		R3	6	5	6	6	5															
31.63	10	R1	5	6	4	4	5	5.666	5.333	5.333	4.666	5.333	13.32	6.66	6.66	6.68	6.66	21.39	14.89	14.89	14.89	14.89
		R2	6	5	5	5	5															
		R3	6	5	7	5	6															
15.72	10	R1	4	6	3	3	5	5.333	4.000	4.000	3.333	4.666	6.66	20.00	20.00	33.34	-6.68	14.89	26.56	26.56	35.24	14.89
		R2	6	4	4	4	5															
		R3	6	2	5	3	4															

Appendix Table 268: Repellency of *T. castaneum* by seed (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	7	9	7	7	7	6.666	7.000	7.333	7.000	6.333	33.32	40.00	46.66	40.00	26.66	35.24	39.23	43.05	39.23	31.05
		R2	8	6	9	6	6															
		R3	5	6	6	8	6															
125.70	10	R1	5	6	7	5	5	6.000	5.666	6.333	5.333	5.666	20.00	13.32	26.66	6.66	13.32	26.56	21.39	31.05	14.89	21.39
		R2	7	5	7	6	5															
		R3	6	6	5	5	7															
62.86	10	R1	5	5	6	4	4	5.333	5.666	5.666	5.666	5.333	6.66	13.32	13.32	13.32	6.66	14.89	21.39	21.39	21.39	14.89
		R2	6	5	6	6	5															
		R3	5	7	5	7	7															
31.63	10	R1	6	4	5	3	3	4.666	4.666	4.333	4.333	3.666	-6.68	-6.68	-13.34	-13.34	-26.68	-14.89	-14.89	-21.39	-21.39	-31.05
		R2	5	4	5	4	5															
		R3	3	6	3	6	4															
15.72	10	R1	4	3	4	3	2	3.000	2.333	3.333	3.333	2.333	-40.00	-53.34	-33.34	-33.34	-53.34	-39.23	-46.89	-35.24	-35.24	-46.89
		R2	3	2	4	3	3															
		R3	2	2	2	4	2															

Appendix Table 269: Repellency of *T. castaneum* by stem wood (chloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm^2)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	9	10	9	10	8	9.666	9.333	9.000	9.333	8.666	93.32	86.66	80.00	86.66	73.32	75.00	68.35	63.44	68.35	58.89
		R2	10	9	8	9	10															
		R3	10	9	10	9	8															
125.70	10	R1	9	7	8	9	6	8.000	7.666	8.000	7.666	7.000	60.00	53.32	60.00	53.32	40.00	50.77	46.89	50.77	46.89	39.23
		R2	6	8	7	8	8															
		R3	9	8	9	6	7															
62.86	10	R1	7	6	8	7	7	7.666	6.666	6.333	6.666	6.333	53.32	33.32	26.66	33.32	26.66	46.89	35.24	31.05	35.24	31.05
		R2	8	7	6	7	7															
		R3	8	7	5	6	5															
31.63	10	R1	6	5	6	6	5	6.333	5.666	6.333	5.666	5.333	26.66	13.32	26.66	13.32	6.66	31.05	21.39	31.05	21.39	14.89
		R2	7	6	7	7	6															
		R3	6	6	6	4	5															
15.72	10	R1	5	6	5	5	5	5.666	5.333	5.666	5.666	4.333	13.32	6.66	13.32	13.32	- 13.32	21.39	14.89	21.39	21.39	- 21.39
		R2	7	6	7	6	5															
		R3	5	4	5	6	3															

Appendix Table 270: Repellency of *T. castaneum* by stem wood (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	7	8	8	8.333	8.000	7.666	7.333	8.000	66.66	60.00	53.32	46.66	60.00	54.70	50.77	46.89	43.05	50.77
		R2	9	7	8	7	9															
		R3	7	8	8	7	7															
125.70	10	R1	9	7	6	7	6	7.333	7.333	6.333	6.666	6.333	46.66	46.66	26.66	33.32	26.66	43.05	43.05	31.05	35.24	31.05
		R2	8	9	7	6	7															
		R3	6	7	6	7	6															
62.86	10	R1	7	7	5	6	5	6.333	6.000	5.666	6.000	5.666	26.66	20.00	13.32	20.00	13.32	31.05	26.56	21.39	26.56	21.39
		R2	7	6	6	5	6															
		R3	5	5	6	7	6															
31.63	10	R1	6	6	4	5	4	5.333	5.333	5.333	5.000	4.000	6.66	6.66	6.66	0.00	-20.00	14.89	14.89	14.89	00.00	-26.56
		R2	6	5	6	4	5															
		R3	4	5	6	6	3															
15.72	10	R1	5	5	3	4	5	4.333	4.000	4.000	3.666	3.000	-13.34	-20.00	-20.00	-26.56	-40.00	-21.39	-26.56	-26.56	-31.05	-39.23
		R2	5	3	5	3	3															
		R3	4	4	4	4	1															

Appendix Table 271: Repellency of *T. castaneum* by root wood (chloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm^2)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	8	9	7	8.666	8.666	8.333	8.000	8.333	73.32	73.32	66.66	60.00	66.66	58.89	58.89	54.70	50.77	54.70
		R2	9	7	9	8	8															
		R3	7	10	8	7	10															
125.70	10	R1	9	8	7	8	8	8.000	7.666	7.333	7.000	6.666	60.00	53.32	46.66	40.00	33.32	50.77	46.89	43.05	39.23	35.24
		R2	9	8	8	7	5															
		R3	6	7	7	6	7															
62.86	10	R1	8	7	6	6	7	6.666	6.333	6.000	5.666	5.666	33.32	26.66	20.00	13.32	13.32	35.24	31.05	26.56	21.39	21.39
		R2	7	5	7	6	5															
		R3	5	7	5	5	5															
31.63	10	R1	6	5	5	5	6	6.000	5.666	5.333	4.333	5.333	20.00	13.32	6.66	-13.34	6.66	26.56	21.39	14.89	-21.39	14.89
		R2	8	5	6	4	5															
		R3	4	7	5	4	5															
15.72	10	R1	5	6	4	4	5	4.666	5.000	3.666	3.000	4.333	-6.68	0.00	-26.68	-40.00	-13.34	-14.89	00.00	-31.05	-39.23	-21.39
		R2	6	4	4	3	6															
		R3	3	5	3	2	2															

Appendix Table 272: Repellency of *T. castaneum* by root wood (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	9	8	10	9	8	9.666	9.333	9.000	8.666	9.000	93.32	86.66	80.00	73.32	80.00	75.00	68.53	63.44	58.89	63.44
		R2	10	10	9	8	10															
		R3	10	10	8	9	9															
125.70	10	R1	10	9	9	8	9	8.666	8.000	8.000	7.333	8.000	73.32	60.00	60.00	46.66	60.00	58.89	50.77	50.77	43.05	50.77
		R2	8	9	8	7	8															
		R3	8	6	7	7	7															
62.86	10	R1	8	7	8	7	8	7.333	6.666	7.000	6.333	6.666	46.66	33.32	40.00	26.66	33.32	43.05	35.24	39.23	31.05	35.24
		R2	6	8	7	6	5															
		R3	8	5	6	6	6															
31.63	10	R1	6	6	7	6	6	6.000	5.666	6.333	5.333	5.666	20.00	13.32	26.66	6.66	13.32	26.56	21.39	31.05	14.89	21.39
		R2	6	6	6	5	6															
		R3	7	5	6	5	5															
15.72	10	R1	6	5	6	5	4	5.333	4.666	5.666	4.333	4.000	6.66	-6.68	13.32	-13.34	-20.00	14.89	-14.89	21.39	-21.39	-26.56
		R2	4	6	5	4	5															
		R3	6	3	6	4	3															

Appendix Table 273: Repellency of *T. castaneum* by stem bark (choloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	8	9	7	8	9	6.666	7.333	6.666	7.000	7.333	33.32	46.66	33.32	40.00	46.66	35.24	43.05	35.24	39.23	43.05
		R2	7	7	6	6	7															
		R3	5	6	7	7	6															
125.70	10	R1	7	7	6	7	7	6.333	6.000	6.333	6.000	6.333	26.66	20.00	26.66	20.00	26.66	31.05	26.56	31.05	26.56	31.05
		R2	6	7	7	6	6															
		R3	6	4	6	5	6															
62.86	10	R1	5	6	5	6	5	5.666	5.333	5.666	5.333	5.666	13.32	6.66	13.32	6.66	13.32	21.39	14.89	21.39	14.89	21.39
		R2	7	5	6	5	6															
		R3	5	5	6	5	6															
31.63	10	R1	5	5	4	5	4	4.666	4.333	5.333	4.333	4.000	-6.68	-13.34	6.66	-13.34	-20.00	-14.89	-21.39	14.89	-21.39	-26.56
		R2	5	4	5	4	5															
		R3	4	4	7	4	3															
15.72	10	R1	4	3	5	4	4	3.333	3.666	4.000	3.333	3.000	-33.34	-26.68	-20.00	-33.34	-40.00	-35.24	-31.05	-26.56	-35.24	-39.23
		R2	4	4	4	3	3															
		R3	2	4	3	3	2															

Appendix Table 274: Repellency of *T. castaneum* by stem bark (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	9	8	9	7	9	7.666	7.333	7.000	7.666	7.333	53.32	46.66	40.00	53.32	46.66	46.89	43.05	39.23	46.89	43.05
		R2	7	8	7	8	8															
		R3	7	6	5	7	5															
125.70	10	R1	8	7	7	5	6	6.333	6.666	6.000	6.333	6.666	26.66	33.32	20.00	26.66	33.32	31.05	35.24	26.56	31.05	35.24
		R2	6	7	6	7	7															
		R3	5	6	5	7	7															
62.86	10	R1	6	5	6	4	5	6.333	5.666	5.333	5.666	5.666	26.66	13.32	6.66	13.32	13.32	31.05	21.39	14.89	21.39	21.39
		R2	7	6	5	6	6															
		R3	6	6	5	7	6															
31.63	10	R1	5	6	7	3	4	5.333	5.333	5.333	4.666	5.333	6.66	6.66	6.66	-6.68	6.66	14.89	14.89	14.89	-14.89	14.89
		R2	6	5	5	5	6															
		R3	5	5	4	6	6															
15.72	10	R1	4	2	4	2	4	4.333	4.333	4.000	3.333	4.000	-13.34	-13.34	-20.00	-33.34	-20.00	-21.39	-21.39	-26.56	-35.24	-26.56
		R2	5	4	3	4	5															
		R3	4	7	5	4	3															

Appendix Table 275: Repellency of *T. castaneum* by root bark (chloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	8	10	9	8.333	8.000	7.666	8.000	7.666	66.66	60.00	53.32	60.00	53.32	54.70	50.77	46.89	50.77	46.89
		R2	7	9	7	7	7															
		R3	8	6	8	7	7															
125.70	10	R1	9	7	6	8	7	7.666	7.000	7.000	7.000	6.333	53.32	40.00	40.00	40.00	26.66	46.89	39.23	39.23	39.23	31.05
		R2	7	8	7	6	6															
		R3	7	6	8	7	6															
62.86	10	R1	6	5	5	7	5	6.333	5.333	6.000	6.333	5.333	26.66	6.66	20.00	26.66	6.66	31.05	14.89	26.56	31.05	14.89
		R2	7	6	6	5	6															
		R3	6	5	7	7	5															
31.63	10	R1	6	4	4	6	5	5.666	4.333	5.333	5.666	4.666	13.32	-13.34	6.66	13.32	-6.68	21.39	-21.39	14.89	21.39	-14.89
		R2	5	5	5	4	4															
		R3	6	4	7	7	5															
15.72	10	R1	7	3	3	5	3	5.333	3.333	4.333	5.333	3.333	6.66	-33.34	-13.34	6.66	-33.34	14.89	-35.24	-21.39	14.89	-35.24
		R2	5	4	4	4	3															
		R3	4	3	6	7	4															

Appendix Table 276: Repellency of *T. castaneum* by root bark (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	9	9	10	8.666	9.000	8.666	8.666	9.000	73.32	80.00	73.32	73.32	80.00	58.89	63.44	58.89	58.89	63.44
		R2	8	8	8	9	7															
		R3	8	10	9	8	10															
125.70	10	R1	9	8	9	7	9	7.666	8.000	8.333	8.000	8.000	53.32	60.00	66.66	60.00	60.00	46.89	50.77	54.70	50.77	50.77
		R2	8	7	8	9	7															
		R3	6	9	8	8	8															
62.86	10	R1	8	7	7	6	8	6.666	6.666	7.000	6.666	6.666	33.32	33.32	40.00	33.32	33.32	35.24	35.24	39.23	35.24	35.24
		R2	6	6	6	8	5															
		R3	6	7	8	6	7															
31.63	10	R1	6	5	6	5	6	5.666	6.000	5.666	5.333	5.333	13.32	20.00	13.32	6.66	6.66	21.39	26.56	21.39	14.89	14.89
		R2	6	6	5	7	5															
		R3	5	7	6	4	5															
15.72	10	R1	6	4	5	4	3	5.333	4.666	4.333	4.333	3.666	6.66	-6.68	-13.34	-13.34	-26.68	14.89	-14.89	-21.39	-21.39	-31.05
		R2	6	5	4	6	4															
		R3	4	5	4	3	4															

Appendix Table 277: Repellency of *T. castaneum* by flower (chloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	9	8	10	10	9	9.666	9.000	9.333	9.666	9.000	93.32	80.00	86.66	93.32	80.00	75.00	63.00	68.53	75.00	63.44
		R2	10	10	8	9	8															
		R3	10	9	10	10	10															
125.70	10	R1	9	8	9	8	7	8.333	7.666	8.000	8.333	7.000	66.66	53.32	60.00	66.66	40.00	54.70	46.89	50.77	54.70	39.23
		R2	7	9	8	9	6															
		R3	9	6	7	8	8															
62.86	10	R1	8	7	8	7	5	7.000	6.333	6.333	7.333	5.666	40.00	26.66	26.66	46.66	13.32	39.23	31.05	31.05	43.05	21.39
		R2	6	7	6	8	5															
		R3	7	5	5	7	7															
31.63	10	R1	7	6	7	6	4	6.000	6.000	5.333	6.333	5.333	20.00	20.00	6.66	26.66	6.66	26.56	26.56	14.89	31.05	14.89
		R2	6	8	4	7	6															
		R3	5	4	5	6	6															
15.72	10	R1	7	5	6	6	2	5.333	5.666	5.333	5.000	4.000	6.66	13.32	6.66	0.00	-20.00	14.89	21.39	14.89	0.00	-26.56
		R2	5	7	5	6	6															
		R3	4	5	5	3	4															

Appendix Table 278: Repellency of *T. castaneum* by flower (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	9	9	8	9	8.333	7.666	8.333	7.333	8.000	66.66	53.32	66.66	46.66	60.00	54.70	46.89	54.70	43.05	50.77
		R2	9	7	6	7	8															
		R3	6	7	10	7	7															
125.70	10	R1	7	7	8	6	7	6.666	7.333	7.000	6.666	7.000	33.32	46.66	40.00	33.32	40.00	35.24	43.05	39.23	35.24	39.23
		R2	7	8	7	7	8															
		R3	6	7	6	7	6															
62.86	10	R1	6	6	7	5	6	6.000	6.333	6.666	5.666	6.000	20.00	26.66	33.32	13.32	20.00	26.56	31.05	35.24	21.39	26.56
		R2	7	7	6	6	7															
		R3	5	6	7	6	5															
31.63	10	R1	5	5	6	4	5	5.666	5.333	6.333	5.333	5.666	13.32	6.66	26.66	6.66	13.32	21.39	14.89	31.05	14.89	21.39
		R2	6	6	6	6	6															
		R3	6	5	7	6	6															
15.72	10	R1	5	4	4	3	5	5.333	4.333	5.333	4.333	4.666	6.66	-13.34	6.66	-13.34	-6.68	14.89	-21.39	14.89	-21.39	-14.89
		R2	6	5	6	5	3															
		R3	5	4	6	5	5															

Appendix Table 279: Repellency of *T. castaneum* by leaf (chloroform) of *A. indica* with percent repulsion and arcsin transformation data.

Dose ($\mu g/cm^2$)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	9	10	9	10	7	8.666	9.000	8.333	8.666	8.666	73.32	80.00	66.66	73.32	73.32	58.89	63.44	54.70	58.89	58.89
		R2	9	7	8	8	10															
		R3	8	10	8	8	9															
125.70	10	R1	7	8	7	8	7	7.666	7.333	7.666	7.666	7.666	53.32	46.66	53.32	53.32	53.32	46.89	43.05	46.89	46.89	46.89
		R2	8	7	8	7	9															
		R3	8	7	8	8	7															
62.86	10	R1	6	7	6	7	6	6.333	6.666	6.333	6.666	6.666	26.66	33.32	26.66	33.32	33.32	31.05	35.24	31.05	35.24	35.24
		R2	7	6	7	6	8															
		R3	6	7	6	7	6															
31.63	10	R1	5	6	5	6	5	5.666	6.333	5.333	5.666	5.666	13.32	26.66	6.66	13.32	13.32	21.39	31.05	14.89	21.39	21.39
		R2	6	6	6	5	7															
		R3	6	7	5	6	5															
15.72	10	R1	5	4	4	5	5	5.000	5.666	4.333	5.333	4.333	0.00	13.32	-13.34	6.66	-13.34	0.00	21.39	-21.39	14.89	-21.39
		R2	4	5	5	5	5															
		R3	6	8	4	6	3															

Appendix Table 280: Repellency of *T. castaneum* by leaf (methanol) of *A. indica* with percent repulsion and arcsin transformation data.

Dose (µg/cm²)	Insects used	Replication	Hourly observation					Average of hourly observation (Nc)					Percent repulsion (PR) PR = (Nc – 5) × 20%					Arcsin transformation data				
			1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h	1h	2h	3h	4h	5h
251.50	10	R1	10	8	8	9	9	8.666	8.333	8.333	8.000	8.333	73.32	66.66	66.66	60.00	66.66	58.89	54.70	54.70	50.77	54.70
		R2	9	10	8	8	9															
		R3	7	7	9	7	7															
125.70	10	R1	9	8	9	8	8	7.666	7.333	7.666	7.333	7.666	53.32	46.66	53.32	46.66	53.32	46.89	43.05	46.89	43.05	46.89
		R2	8	8	8	7	7															
		R3	6	6	6	7	8															
62.86	10	R1	7	6	6	7	6	6.333	6.333	6.666	6.333	7.000	26.66	26.66	33.32	26.66	40.00	31.05	31.05	35.24	31.05	39.23
		R2	7	7	7	6	7															
		R3	5	6	7	6	8															
31.63	10	R1	6	5	5	6	5	6.000	5.333	6.333	5.666	6.000	20.00	6.66	26.66	13.32	20.00	26.56	14.89	31.05	21.39	26.56
		R2	7	6	8	5	6															
		R3	5	5	6	6	7															
15.72	10	R1	6	5	4	5	5	6.000	5.333	5.666	5.333	4.666	20.00	6.66	13.32	6.66	-6.68	26.56	14.89	21.39	14.89	-14.89
		R2	5	6	7	6	4															
		R3	7	5	6	5	5															

Appendix Table 1: Dose-mortality effect of flower extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	5	16.67	17	4.05	4.044	4.037	13.17	4.046
2123.14	3.326	30	5	16.67	17	4.05	3.992	4.062	12.15	3.993
1415.43	3.150	30	4	13.34	13	3.87	3.918	3.878	12.15	3.920
707.72	2.850	30	3	10.00	10	3.72	3.791	3.72	10.08	3.794
353.86	2.548	30	3	10.00	10	3.72	3.791	3.72	10.08	3.794

Results:

Y = 2.602808 + 0.4181305 X

Chi-squared is 0.1691845 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is5.733119

LD_{50} is 540902.7µg/cm^2

95% confidence limits are 3.37315 to 8.673639E+10 µg/cm^2

Appendix Table 2: Dose-mortality effect of flower extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	13	43.33	43	4.82	4.815	4.838	18.81	4.816
2123.14	3.326	30	12	40.00	40	4.75	4.758	4.740	18.48	4.757
1415.43	3.150	30	11	36.67	37	4.67	4.677	4.659	18.03	4.674
707.72	2.850	30	10	33.33	33	4.56	4.538	4.544	17.43	4.532
353.86	2.548	30	8	26.66	27	4.39	4.400	4.390	16.74	4.389

Results:

Y = 3.185217 + 0.4726539 X

Chi-squared is 2.133322E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.83956

LD_{50} is 6911.297µg/cm^2

95% confidence limits are 530.8342 to 89983.02 µg/cm^2

Appendix Table 3: Dose-mortality effect of flower extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	17	56.67	57	5.18	5.130	5.165	19.02	5.123
2123.14	3.326	30	16	53.33	53	5.08	5.072	5.075	19.11	5.066
1415.43 0	3.150	30	14	46.67	47	4.92	4.989	4.915	19.02	4.985
707.72	2.850	30	13	43.33	43	4.82	4.849	4.838	18.81	4.848
353.86	2.548	30	12	40.00	40	4.75	4.708	4.74	18.48	4.710

Results:

Y = 3.546648 + 0.4566106 X

Chi-squared is 0.1479325 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.182913

LD_{50} is 1523.749µg/cm^2

95% confidence limits are519.1728 to 4472.133 µg/cm^2

Appendix Table 4: Dose-mortality effect of flower extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	21	70	70	5.52	5.499	5.51	18.03	5.488
2123.14	3.326	30	20	66.66	67	5.44	5.437	5.429	18.03	5.429
1415.43	3.150	30	19	63.33	63	5.33	5.352	5.318	18.48	5.346
707.72	2.850	30	17	56.67	57	5.18	5.204	5.202	18.81	5.205
353.86	2.548	30	16	53.33	53	5.08	5.056	5.075	19.11	5.063

Results:

Y = 3.863903 + 0.4706527 X

Chi-squared is 2.660871E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.413875

LD_{50} is 259.3435µg/cm^2

95% confidence limits are 28.99703 to 2319.515 µg/cm^2

Appendix Table 5: Dose-mortality effect of flower extract (methanol) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	5	16.67	17	4.05	3.969	4.062	12.15	3.981
2123.14	3.326	30	4	13.34	13	3.87	3.907	3.878	12.15	3.916
1415.43	3.150	30	3	10.00	10	3.72	3.818	3.72	11.1	3.824
707.72	2.850	30	3	10.00	10	3.72	3.668	3.73	9.060	3.667
353.86	2.548	30	2	6.67	7	3.52	3.517	3.519	8.07	3.510

Results:

Y = 2.180794 + 0.5215927 X

Chi-squared is 0.2537117 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.404997

LD_{50} is 254095.2µg/cm²

95% confidence limits are 38.5011 to 1.676952E+09µg/cm²

Appendix Table 6: Dose-mortality effect of flower extract (methanol) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	9	30	30	4.48	4.449	4.48	16.74	4.452
2123.14	3.326	30	8	26.67	27	4.39	4.419	4.39	16.74	4.421
1415.43	3.150	30	8	26.66	27	4.39	4.377	4.394	15.96	4.377
707.72	2.850	30	7	23.34	23	4.26	4.304	4.266	15.96	4.302
353.86	2.548	30	7	23.33	23	4.26	4.231	4.252	15.09	4.227

Results:

Y = 3.593121 + 0.2488788 X

Chi-squared is 6.374812E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.652869

LD_{50} is 449643.3µg/cm²

95% confidence limits are 4.455762E µg/cm²

Appendix Table 7: Dose-mortality effect of flower extract (methanol) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	14	46.67	47	4.92	4.887	4.942	18.81	4.904
2123.14	3.326	30	13	43.33	43	4.82	4.840	4.838	18.81	4.852
1415.43	3.150	30	12	40.00	40	4.75	4.776	4.74	18.48	4.780
707.72	2.850	30	11	36.67	37	4.67	4.664	4.659	18.03	4.655
353.86	2.548	30	10	33.33	33	4.56	4.552	4.544	17.43	4.530

Results:

Y = 3.474663 + 0.4142152 X

Chi-squared is 6.361366E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.682475

LD_{50} is 4813.655µg/cm²

95% confidence limits are432.4115 to 53586.13 µg/cm²

Appendix Table 8: Dose-mortality effect of flower extract (methanol) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	18	60	60	5.25	5.185	5.24	19.02	5.173
2123.14	3.326	30	16	53.33	53	5.08	5.146	5.065	19.02	5.135
1415.43	3.150	30	16	53.33	53	5.08	5.092	5.075	19.11	5.082
707.72	2.850	30	15	50	50	5.00	4.999	4.99	19.02	4.991
353.86	2.548	30	14	46.	47	4.92	4.907	4.915	19.02	4.901

Results:

Y = 4.133716 + 0.3011669 X

Chi-squared is .1843769 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.876425

LD_{50} is 752.3578µg/cm²

95% confidence limits are 127.7164 to 4432.026 µg/cm²

Appendix Table 9. Dose-mortality effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	8	26.67	27	4.39	4.379	4.394	15.96	4.379
2123.14	3.326	30	7	23.33	23	4.26	4.310	4.266	15.96	4.309
1415.43	3.150	30	7	23.33	23	4.26	4.213	4.252	15.09	4.212
707.72	2.850	30	5	16.66	17	4.05	4.046	4.037	13.17	4.044
353.86	2.548	30	4	13.33	13	3.87	3.880	3.873	11.1	3.876

Results:

Y = 2.457657 + 0.5567423 X

Chi-squared is 5.935478E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.566465

LD_{50} is 36852.30 µg/cm²

95% confidence limits are 385.3198 to 3524579µg/cm²

Appendix Table 10. Dose-mortality effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	17	56.67	57	5.18	5.132	5.165	19.02	5.130
2123.14	3.326	30	16	53.33	53	5.08	5.017	5.075	19.11	5.012
1415.43	3.150	30	12	40.00	40	4.75	4.854	4.76	18.81	4.847
707.72	2.850	30	9	30.00	30	4.48	4.576	4.46	17.43	4.564
353.86	2.548	30	8	26.66	27	4.39	4.299	4.388	15.09	4.282

Results:

Y = 1.889938 + 0.9385532 X

Chi-squared is 0.6018476 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.313677

LD_{50} is 2059.099 µg/cm²

95% confidence limits are 1108.395 to 3825.246µg/cm²

Appendix Table 11. Dose-mortality effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	21	70	70	5.52	5.435	5.51	18.03	5.422
2123.14	3.326	30	19	63.33	63	5.33	5.334	5.318	18.48	5.321
1415.43	3.150	30	16	53.33	53	5.08	5.190	5.065	19.02	5.177
707.72	2.850	30	14	46.66	47	4.92	4.943	4.915	19.02	4.933
353.86	2.548	30	12	40.00	40	4.75	4.697	4.74	18.03	4.688

Results:

Y = 2.614321 + 0.813474 X

Chi-squared is 0.4344726 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.932705

LD_{50} is856.4559 µg/cm^2

95% confidence limits are 459.4218 to 1596.608µg/cm^2

Appendix Table 12. Dose-mortality effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	24	80	80	5.85	5.754	5.83	15.96	5.731
2123.14	3.326	30	22	73.33	73	5.61	5.683	5.61	16.74	5.665
1415.43	3.150	30	21	70	70	5.52	5.582	5.5	17.43	5.573
707.72	2.850	30	20	66.66	67	5.44	5.410	5.429	18.03	5.416
353.86	2.548	30	18	60	60	5.25	5.238	5.28	18.81	5.258

Results:

Y = 3.925557 + 0.5230322 X

Chi-squared is 0.3142581 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.054258

LD_{50} is 113.3073 µg/cm^2

95% confidence limits are 6.037319 to 2126.529µg/cm^2

Appendix Table 13. Dose-mortality effect of leaf extract (methanol) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	4	13.33	13	3.87	3.883	3.873	11.1	3.886
2123.14	3.326	30	4	13.33	13	3.87	3.837	3.873	11.1	3.840
1415.43	3.150	30	3	10.00	10	3.72	3.772	3.72	10.08	3.774
707.72	2.850	30	3	10.00	10	3.72	3.771	3.72	10.08	3.775
353.86	2.548	30	2	6.666	7	3.52	3.548	3.519	8.07	3.552

Results:

Y = 2.609282 + 0.3699346 X

Chi-squared is 9.324336E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.462543

LD_{50} is 29009.69 µg/cm^2

95% confidence limits are 2.899154E-02 to 2.902783E+14µg/cm^2

Appendix Table 14. Dose-mortality effect of leaf extract (methanol) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	12	40.00	40	4.75	4.673	4.74	18.03	4.662
2123.14	3.326	30	10	33.33	33	4.56	4.613	4.551	18.03	4.603
1415.43	3.150	30	9	30.00	30	4.48	4.528	4.46	17.43	4.519
707.72	2.850	30	8	26.66	27	4.39	4.384	4.394	15.96	4.376
353.86	2.548	30	7	23.34	23	4.26	4.240	4.252	15.09	4.233

Results:

Y = 3.021963 + 0.4752018 X

Chi-squared is 0.2290532 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.16252

LD_{50} is 14538.5 µg/cm^2

95% confidence limits are 399.2724 to 529383µg/cm^2

Appendix Table 15. Dose-mortality effect of leaf extract (methanol) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	18	60.00	60	5.25	5.174	5.24	19.02	5.168
2123.14	3.326	30	16	53.33	53	5.08	5.096	5.075	19.11	5.090
1415.43	3.150	30	14	46.66	47	4.92	4.987	4.915	19.02	4.979
707.72	2.850	30	12	40.00	40	4.75	4.799	4.74	18.48	4.790
353.86	2.548	30	11	36.67	37	4.67	4.612	4.659	18.03	4.600

Results:

Y = 2.997115 + 0.629013 X

Chi-squared is 0.28724 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.184172

LD_{50} is 1528.169 µg/cm²

95% confidence limits are 698.0223 to 3345.597µg/cm²

Appendix Table 16. Dose-mortality effect of leaf extract (methanol) of *A. indica* against *T. castaneum* male after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	21	70.00	70	5.52	5.487	5.51	18.03	5.475
2123.14	3.326	30	20	66.66	67	5.44	5.413	5.429	18.03	5.402
1415.43	3.150	30	18	60.00	60	5.25	5.308	5.24	18.48	5.297
707.72	2.850	30	16	53.33	53	5.08	5.130	5.065	19.02	5.118
353.86	2.548	30	15	50.00	50	5.00	4.950	4.99	19.02	4.940

Results:

Y = 3.426053 + 0 .5938126 X

Chi-squared is 0.1975682 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.650579

LD_{50} is 447.2792 µg/cm²

95% confidence limits are 127.1399 to 1573.53µg/cm²

Appendix Table 17. Dose-mortality effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	6	20	20	4.16	4.138	4.17	14.13	4.138
2123.14	3.326	30	5	16.67	17	4.05	4.058	4.037	13.17	4.061
1415.43	3.150	30	4	13.33	13	3.87	3.948	3.878	12.15	3.953
707.72	2.850	30	4	13.33	13	3.87	3.758	3.894	10.08	3.767
353.86	2.548	30	2	6.67	7	3.52	3.568	3.519	8.07	3.582

Results

Y = 2.011662 + 0.6160064 X

Chi-squared is 0.2835674 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.851149

LD_{50} is 70982.01µg/cm^2

95% confidence limits are 298.7637 to1.686435E+07 µg/cm^2

Appendix Table 18. Dose-mortality effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	10	33.33	33	4.56	4.470	4.57	16.74	4.476
2123.14	3.326	30	8	26.67	27	4.39	4.424	4.39	16.74	4.429
1415.43	3.150	30	7	23.34	23	4.26	4.358	4.266	15.96	4.362
707.72	2.850	30	7	23.33	23	4.26	4.245	4.252	15.09	4.246
353.86	2.548	30	6	20.00	20	4.16	4.132	4.170	14.13	4.131

Results

Y = 3.155897 + 0.3827177 X

Chi-squared is 0.3384838 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.818442

LD_{50} is65832.731µg/cm^2

95% confidence limits are 46.50053 to 9.320226E+07 µg/cm^2

Appendix Table 19. Dose-mortality effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	15	50.00	50	5	4.899	5.02	18.81	4.914
2123.14	3.326	30	13	43.33	43	4.82	4.842	4.838	18.81	4.854
1415.43	3.150	30	11	36.67	37	4.67	4.761	4.662	18.48	4.768
707.72	2.850	30	10	33.33	33	4.56	4.623	4.551	18.03	4.623
353.86	2.548	30	10	33.33	33	4.56	4.485	4.57	16.74	4.477

Results

Y = 3.243517 + 0.4839424 X

Chi-squared is 0.6619892 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.629531

LD_{50} is 4261.189µg/cm^2

95% confidence limits are 618.3115 to 29366.64 µg/cm^2

Appendix Table 20. Dose-mortality effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm^2	**Log dose**	**#used**	**#Kill**	**%Kill**	**Corr%**	**Emp probit**	**Expt probit**	**Work probit**	**Weight**	**Final probit**
2830.86	3.452	30	21	70.00	70	5.52	5.342	5.5	18.48	5.346
2123.14	3.326	30	17	56.67	57	5.18	5.287	5.202	18.81	5.290
1415.43	3.150	30	16	53.33	53	5.08	5.210	5.098	18.81	5.210
707.72	2.850	30	16	53.33	53	5.08	5.077	5.075	19.11	5.076
353.86	2.548	30	15	50.00	50	5.00	4.944	4.99	19.02	4.940

Results

Y = 3.79532 + 0.4492497 X

Chi-squared is 0.8699672 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.681538

LD_{50} is 480.3277µg/cm^2

95% confidence limits are 98.38282 to 2345.072 µg/cm^2

Appendix Table 21. Dose-mortality effect of root bark extract (methanol) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	5	16.67	17	4.05	4.009	4.037	13.17	4.006
2123.14	3.326	30	4	13.33	13	3.87	3.943	3.878	12.15	3.942
1415.43	3.150	30	4	13.33	13	3.87	3.851	3.873	11.10	3.851
707.72	2.850	30	3	10.00	10	3.72	3.693	3.73	9.060	3.696
353.86	2.548	30	2	6.66	7.0	3.52	3.535	3.519	8.07	3.541

Results:

Y = 2.22816 + 0.5151049 X

Chi-squared is 8.163643E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 5.381119

LD_{50} is 240501.9µg/cm²

95% confidence limits are 40.22166 to 1.43806E+09µg/cm²

Appendix Table 22. Dose-mortality effect of root bark extract (methanol) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	9	30.00	30	4.48	4.506	4.46	17.43	4.492
2123.14	3.326	30	8	26.67	27	4.39	4.418	4.39	16.74	4.407
1415.43	3.150	30	8	26.66	27	4.39	4.296	4.388	15.09	4.288
707.72	2.850	30	5	16.66	17	4.05	4.084	4.037	13.17	4.083
353.86	2.548	30	4	13.33	13	3.87	3.874	3.873	11.1	3.878

Results:

Y = 2.146664 + 0.6794698 X

Chi-squared is 0.2034314 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 4.199357

LD_{50} is 15825.49µg/cm²

95% confidence limits are980.0414 to 255546.5µg/cm²

Appendix Table 23. Dose-mortality effect of root bark extract (methanol) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	15	50.00	50	5	4.980	4.99	19.02	4.983
2123.14	3.326	30	14	46.66	47	4.92	4.886	4.942	18.81	4.888
1415.43	3.150	30	12	40.00	40	4.75	4.754	4.74	18.48	4.753
707.72	2.850	30	8	26.67	27	4.39	4.528	4.376	17.43	4.522
353.86	2.548	30	8	26.66	27	4.39	4.301	4.394	15.96	4.292

Results:

Y = 2.338804 + 0.766129 X

Chi-squared is 0.5990839 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.473561

LD_{50} is 2975.506µg/cm^2

95% confidence limits are 1124.038 to 7876.648 µg/cm^2

Appendix Table 24. Dose-mortality effect of root bark extract (methanol) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	18	60.00	60	5.25	5.248	5.28	18.81	5.258
2123.14	3.326	30	17	56.66	57	5.18	5.180	5.165	19.02	5.186
1415.43	3.150	30	16	53.33	53	5.08	5.083	5.075	19.11	5.084
707.72	2.850	30	14	46.66	47	4.92	4.916	4.915	19.02	4.910
353.86	2.548	30	12	40.00	40	4.75	4.751	4.74	18.48	4.737

Results:

Y = 3.268065 + 0.5763386 X

Chi-squared is 1.968241E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.005066

LD_{50} is 1011.733µg/cm^2

95% confidence limits are 445.3582 to 2298.383 µg/cm^2

Appendix Table 25: Dose-mortality effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	11	36.66	37	4.67	4.635	4.659	18.03	4.620
2123.14	3.326	30	10	33.33	33	4.56	4.582	4.544	17.43	4.569
1415.43	3.150	30	9	30.00	30	4.48	4.508	4.46	17.43	4.496
707.72	2.850	30	8	26.66	27	4.39	4.380	4.394	15.96	4.372
353.86	2.548	30	7	23.33	23	4.26	4.254	4.252	15.09	4.248

Results:

Y = 3.19942 + 0.4117216 X

Chi-squared is 6.883776E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.373296

LD_{50} is 23620.85µg/cm^2

95% confidence limits are 172.8482 to 3227946µg/cm^2

Appendix Table 26: Dose-mortality effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	18	60.00	60	5.25	5.202	5.28	18.81	5.214
2123.14	3.326	30	17	56.66	57	5.18	5.132	5.165	19.02	5.142
1415.43	3.150	30	14	46.66	47	4.92	5.032	4.925	19.11	5.038
707.72	2.850	30	13	43.33	43	4.82	4.862	4.838	18.81	4.862
353.86	2.548	30	12	40.00	40	4.75	4.691	4.74	18.03	4.686

Results:

Y = 3.193704 + 0.5855628 X

Chi-squared is 0.4004541 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.084718

LD_{50} is 1215.395µg/cm^2

95% confidence limits are 547.9739 to 2695.722µg/cm^2

Appendix Table 27: Dose-mortality effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	27	90	90	6.28	6.178	6.27	12.15	6.125
2123.14	3.326	30	26	86.67	87	6.13	66.004	6.087	13.17	5.958
1415.43	3.150	30	21	70	70	5.52	5.757	5.51	15.96	5.722
707.72	2.850	30	17	56.66	57	5.18	5.335	5.162	18.48	5.319
353.86	2.548	30	16	53.33	53	5.08	4.914	5.065	19.02	4.916

Results:

Y = 1.504299 + 1.338637 X

Chi-squared is 2.070941 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.611389

LD_{50} is 408.6851µg/cm²

95% confidence limits are 224.8994 to 742 µg/cm²

Appendix Table 28. Dose-mortality effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	29	96.66	97	6.88	6.576	6.759	8.07	6.584
2123.14	3.326	30	28	93.33	93	6.48	6.406	6.491	9.060	6.414
1415.43	3.150	30	25	83.33	83	5.95	6.168	5.948	12.15	6.175
707.72	2.850	30	23	76.66	77	5.74	5.760	5.734	15.96	5.766
353.86	2.548	30	20	66.66	67	5.44	5.353	5.422	18.48	5.358

Results:

Y = 1.900698 + 1.35663 X

Chi-squared is 1.020651 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 0.254365 µg/cm²

LD_{50} is 192.5573µg/cm²

95% confidence limits are 74.1513 to 500.036µg/cm²

Appendix Table29: Dose-mortality effect of root wood extract (methanol) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	6	20.00	20	4.16	4.176	4.17	14.13	4.180
2123.14	3.326	30	6	20.00	20	4.16	4.138	4.17	14.13	4.142
1415.43	3.150	30	5	16.67	17	4.05	4.084	4.037	13.17	4.089
707.72	2.850	30	5	16.67	17	4.05	3.992	4.062	12.15	3.998
353.86	2.548	30	4	13.33	13	3.87	3.900	3.878	12.15	3.906

Results:

Y = 3.134514 + 0.3029002 X

Chi-squared is 0.1079621 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.158749

LD_{50} is 14412.80 µg/cm^2

95% confidence limits are 3.770656E-02 to 5.509097E+13 µg/cm^2

Appendix Table 30: Dose-mortality effect of root wood extract (methanol) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm^2	**Log dose**	**#used**	**#Kill**	**%Kill**	**Corr%**	**Emp probit**	**Expt probit**	**Work probit**	**Weight**	**Final probit**
2830.86	3.452	30	13	43.33	43	4.82	4.780	4.818	18.48	4.775
2123.14	3.326	30	12	40.00	40	4.75	4.703	4.74	18.48	4.696
1415.43	3.150	30	9	30.00	30	4.48	4.593	4.46	17.43	4.585
707.72	2.850	30	8	26.67	27	4.39	4.406	4.39	16.74	4.396
353.86	2.548	30	7	23.34	23	4.26	4.218	4.252	15.09	4.206

Results:

Y = 2.602638 + 0.6293048 X

Chi-squared is 0.3760615 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.809541

LD_{50} is 6449.728 µg/cm^2

95% confidence limits are 969.3614 to 42913.73E+13µg/cm^2

Appendix Table 31: Dose-mortality effect of root wood extract (methanol) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	20	66.67	67	5.44	5.267	5.462	18.81	5.275
2123.14	3.326	30	17	56.67	57	5.18	5.181	5.165	19.02	5.187
1415.43	3.150	30	13	43.33	43	4.82	5.060	4.825	19.11	5.064
707.72	2.850	30	13	43.33	43	4.82	4.854	4.838	18.81	4.853
353.86	2.548	30	12	40.00	40	4.75	4.647	4.74	18.03	4.642

Results:

Y = 2.853815 + 0.7014981 X

Chi-squared is 1.936571 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.059431

LD_{50} is 1146.651 µg/cm^2

95% confidence limits are 589.842 to 2229.087µg/cm^2

Appendix Table 32: Dose-mortality effect of root wood extract (methanol) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	23	76.67	77	5.74	5.596	5.696	17.43	5.564
2123.14	3.326	30	20	66.67	67	5.44	5.508	5.416	17.43	5.477
1415.43	3.150	30	18	60.00	60	5.25	5.383	5.24	18.48	5.358
707.72	2.850	30	17	56.67	57	5.18	5.168	5.165	19.02	5.155
353.86	2.548	30	15	50.00	50	5.00	4.954	4.99	19.02	4.950

Results:

Y = 3.223551 + 0.6776109 X

Chi-squared is 0.668766 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.621636

LD_{50} is 418.4427µg/cm^2

95% confidence limits are 132.3817 to 1322.646µg/cm^2

Appendix Table 33: Dose-mortality effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	7	23.33	23	4.26	4.258	4.252	15.09	4.256
3538.57	3.549	30	6	20.00	20	4.16	4.112	4.17	14.13	4.112
2653.93	3.424	30	5	16.66	17	4.05	4.052	4.037	13.17	4.052
1769.29	3.248	30	4	13.33	13	3.87	3.966	3.878	12.15	3.967
884.64	2.947	30	4	13.33	13	3.87	3.820	3.873	11.1	3.823

Results:

Y = 2.408829 + 0.4800213 X

Chi-squared is .1759168with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.398034

LD_{50} is 250054.2µg/cm²

95% confidence limits are 111.5826 to5.603668E+08µg/cm²

Appendix Table 34: Dose-mortality effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	14	46.66	47	4.92	4.926	4.915	19.02	4.930
3538.57	3.549	30	13	43.33	43	4.82	4.803	4.838	18.81	4.801
2653.93	3.424	30	12	40.00	40	4.75	4.752	4.74	18.48	4.748
1769.29	3.248	30	11	36.66	37	4.67	4.680	4.659	18.03	4.673
884.64	2.947	30	10	33.33	33	4.56	4.557	4.544	17.43	4.544

Results:

Y = 3.281171 + 0.4284342 X

Chi-squared is 3.414214E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.011885

LD_{50} is 10277.43µg/cm²

95% confidence limits are 881.0349 to 119888.2µg/cm²

Appendix Table 35: Dose-mortality effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	21	70.00	70	5.52	5.504	5.5	17.43	5.498
3538.57	3.549	30	19	63.33	63	5.33	5.254	5.358	18.81	5.252
2653.93	3.424	30	16	53.33	53	5.08	5.150	5.065	19.02	5.149
1769.29	3.248	30	14	46.66	47	4.92	5.004	4.925	19.11	5.004
884.64	2.947	30	13	43.33	43	4.82	4.756	4.818	18.48	4.758

Results:

Y = 2.341571 + 0.820051 X

Chi-squared is 0.5358367 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.241785

LD_{50} is 1744.959µg/cm^2

95% confidence limits are 915.3052 to 3326.627µg/cm^2

Appendix Table 36: Dose-mortality effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	25	83.33	83	5.95	5.856	5.902	15.09	5.803
3538.57	3.549	30	22	73.33	73	5.61	5.663	5.61	16.74	5.638
2653.93	3.424	30	20	66.66	67	5.44	5.583	5.416	17.43	5.569
1769.29	3.248	30	21	70.00	70	5.52	5.470	5.51	18.03	5.472
884.64	2.947	30	19	63.33	63	5.33	5.278	5.358	18.81	5.306

Results:

Y = 3.687769 + 0 .5494671 X

Chi-squared is 0.6432109 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.388188

LD_{50} is 244.4488µg/cm^2

95% confidence limits are 11.30526 to 5285.62µg/cm^2

Appendix Table 37: Dose-mortality effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* after 24 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	5	16.67	17	4.05	4.015	4.037	13.17	4.013
3538.57	3.549	30	4	13.33	13	3.87	3.901	3.878	12.15	3.901
2653.93	3.424	30	4	13.33	13	3.87	3.854	3.873	11.1	3.855
1769.29	3.248	30	3	10.00	10	3.72	3.786	3.72	10.08	3.790
884.64	2.947	30	3	10.00	10	3.72	3.673	3.73	9.060	3.678

Results:

Y = 2.58632 + 0.3705477 X

Chi-squared is 9.118784E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.51382

LD_{50} is 32645.24µg/cm^2

95% confidence limits are 0.1466581 to 7.266633E+13µg/cm^2

Appendix Table 38: Dose-mortality effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* after 48 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	9	30.00	30	4.48	4.470	4.48	16.74	4.471
3538.57	3.549	30	8	26.67	27	4.39	4.362	4.394	15.96	4.364
2653.93	3.424	30	7	23.33	23	4.26	4.317	4.266	15.96	4.320
1769.29	3.248	30	7	23.33	23	4.26	4.254	4.252	15.09	4.257
884.64	2.947	30	6	20.00	20	4.16	4.146	4.17	14.13	4.150

Results:

Y = 3.101123 + 0.3558062 X

Chi-squared is 6.792498E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.336828

LD_{50} is 217184.2µg/cm^2

95% confidence limits are 19.47927 to 2.421494E+09µg/cm^2

Appendix Table 39: Dose-mortality effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* after 72 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	16	53.33	53	5.08	4.977	5.065	19.02	4.974
3538.57	3.549	30	12	40.00	40	4.75	4.847	4.76	18.81	4.840
2653.93	3.424	30	12	40.00	40	4.75	4.793	4.74	18.48	4.784
1769.29	3.248	30	11	36.67	37	4.67	4.716	4.662	18.48	4.706
884.64	2.947	30	11	36.67	37	4.67	4.586	4.656	17.43	4.572

Results:

Y = 3.263176 + 0.444474 X

Chi-squared is 0.4730903 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.907595

LD_{50} is 8083.415µg/cm²

95% confidence limits are 1045.788 to 62480.82µg/cm²

Appendix Table 40: Dose-mortality effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* after 96 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	19	63.33	63	5.33	5.324	5.318	18.48	5.333
3538.57	3.549	30	18	60.00	60	5.25	5.243	5.28	18.81	5.248
2653.93	3.424	30	17	56.66	57	5.18	5.209	5.202	18.81	5.214
1769.29	3.248	30	17	56.66	57	5.18	5.162	5.165	19.02	5.164
884.64	2.947	30	16	53.33	53	5.08	5.080	5.075	19.11	5.080

Results:

Y = 4.254948 + 0.2800412 X

Chi-squared is 2.562422E-02with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.660511

LD_{50} is 457.6257µg/cm²

95% confidence limits are 5.592158 to 37449.12µg/cm²

Appendix Table 41: Dose-mortality effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* after 24 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	6	20	20	4.16	4.165	4.17	14.13	4.162
3538.57	3.549	30	5	16.67	17	4.05	4.078	4.037	13.17	4.078
2653.93	3.424	30	5	16.67	17	4.05	4.042	4.037	13.17	4.042
1769.29	3.248	30	5	16.66	17	4.05	3.990	4.062	12.15	3.992
884.64	2.947	30	4	13.33	13	3.87	3.904	3.878	12.15	3.907

Results:

Y = 3.074816 + 0.282503 X

Chi-squared is 9.210211E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 6.814739

LD_{50} is 65273.73µg/cm^2

95% confidence limits are 1.224582E-03 to 3.479271E+16 µg/cm^2

Appendix Table 42: Dose-mortality effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* after 48 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	12	40	40	4.75	4.862	4.76	18.81	4.863
3538.57	3.549	30	13	43.33	43	4.82	4.670	4.821	18.03	4.670
2653.93	3.424	30	11	36.66	37	4.67	4.591	4.656	17.43	4.590
1769.29	3.248	30	8	26.66	27	4.39	4.479	4.39	16.74	4.477
884.64	2.947	30	7	23.33	23	4.26	4.288	4.252	15.09	4.284

Results:

Y = 2.3939 + 0.6413751 X

Chi-squared is 0.829187 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.063301

LD_{50} is 11569.14µg/cm^2

95% confidence limits are 1927.985 to 69422.27µg/cm^2

Appendix Table 43: Dose-mortality effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* after 72 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	17	56.67	57	5.18	5.213	5.202	18.81	5.227
3538.57	3.549	30	16	53.33	53	5.08	5.026	5.075	19.11	5.032
2653.93	3.424	30	15	50.00	50	5.00	4.949	4.99	19.02	4.950
1769.29	3.248	30	12	40.00	40	4.75	4.840	4.76	18.81	4.836
884.64	2.947	30	11	36.66	37	4.67	4.653	4.659	18.03	4.640

Results:

Y = 2.728425 + 0.6490146 X

Chi-squared is 0.1924589 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.500037

LD_{50} is 3162.549µg/cm^2

95% confidence limits are 1491.555 to 6705.562 µg/cm^2

Appendix Table 44: Dose-mortality effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* after 96 h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	22	73.33	73	5.61	5.593	5.584	17.43	5.565
3538.57	3.549	30	19	63.33	63	5.33	5.474	5.321	18.03	5.459
2653.93	3.424	30	21	70.00	70	5.52	5.424	5.51	18.03	5.415
1769.29	3.248	30	20	66.67	67	5.44	5.354	5.422	18.48	5.353
884.64	2.947	30	17	56.67	57	5.18	5.234	5.202	18.81	5.246

Results:

Y = 4.205425 + 0.3532397 X

Chi-squared is 0.6384363 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.249394

LD_{50} is 177.58µg/cm^2

95% confidence limits are 0.905008 to 34844.66µg/cm^2

Appendix Table 45: Dose-mortality effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* after 24 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	12	40	40	4.75	4.711	4.74	18.48	4.704
3538.57	3.549	30	10	33.33	33	4.56	4.548	4.544	17.43	4.542
2653.93	3.424	30	8	26.67	27	4.39	4.481	4.39	16.74	4.475
1769.29	3.248	30	8	26.67	27	4.39	4.386	4.394	15.96	4.380
884.64	2.947	30	7	23.33	23	4.26	4.224	4.252	15.09	4.218

Results:

Y = 2.630763 + 0.5386361 X

Chi-squared is 0.1649764 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.398585

LD_{50} is 25037.14µg/cm^2

95% confidence limits are 1119.11 to 560141µg/cm^2

Appendix Table 46: Dose-mortality effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* after 48 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	17	56.66	57	5.18	5.196	5.165	19.02	5.187
3538.57	3.549	30	16	53.33	53	5.08	5.10	5.075	19.11	5.003
2653.93	3.424	30	14	46.66	47	4.92	4.933	4.915	19.02	4.927
1769.29	3.248	30	12	40	40	4.75	4.824	4.76	18.81	4.820
884.64	2.947	30	11	36.67	37	4.67	4.638	4.659	18.03	4.636

Results:

Y = 2.841155 + 0.6092778 X

Chi-squared is 0.1863411 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.543285

LD_{50} is 3493.696 µg/cm^2

95% confidence limits are 1511.623 to 8074.704µg/cm^2

Appendix Table 47: Dose-mortality effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* after 72 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	27	90	90	6.28	6.154	6.27	12.15	6.113
3538.57	3.549	30	23	76.67	77	5.74	5.776	5.734	15.96	5.754
2653.93	3.424	30	21	70	70	5.52	5.620	5.52	16.74	5.604
1769.29	3.248	30	18	60	60	5.25	5.399	5.24	18.48	5.394
884.64	2.947	30	17	56.66	57	5.18	5.022	5.175	19.11	5.035

Results:

Y = 1.517816 + 1.193606 X

Chi-squared is 1.239829 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.917365

LD_{50} is 826.733 µg/cm²

95% confidence limits are 388.0266 to 1761.442µg/cm²

Appendix Table 48: Dose-mortality effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* after 96 h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
7077.14	3.850	30	29	96.67	97	6.88	6.922	6.844	4.62	6.915
3538.57	3.549	30	28	93.33	93	6.48	6.426	6.491	9.060	6.417
2653.93	3.424	30	27	90.00	90	6.28	6.220	6.23	11.10	6.211
1769.29	3.248	30	24	80.00	80	5.85	5.930	5.87	14.13	5.920
884.64	2.947	30	20	66.67	67	5.44	5.433	5.429	18.03	5.421

Results:

Y = 0.5471034 + 1.654101 X

Chi-squared is 0.1123829 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.692034

LD_{50} is 492.0781µg/cm²

95% confidence limits are 210.316 to 1151.319µg/cm²

Appendix Table 49. Dose-mortality effect of seed extract (chloroform) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	9	30	30	4.48	4.451	4.48	16.74	4.456
2123.14	3.326	30	8	26.66	27	4.39	4.398	4.394	15.96	4.403
1415.43	3.150	30	7	23.33	23	4.26	4.324	4.266	15.96	4.328
707.72	2.850	30	7	23.33	23	4.26	4.196	4.284	14.13	4.199
353.86	2.548	30	5	16.66	17	4.05	4.070	4.037	13.17	4.071

Results:

Y = 2.981806 + 0.4272701 X

Chi-squared is 0.1880722 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.723463

LD_{50} is 52900.91µg/cm^2

95% confidence limits are 99.69377 to 2.807107E+07µg/cm^2

Appendix Table 50. Dose-mortality effect of seed extract (chloroform) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm^2	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	15	50	50	5.00	4.986	4.99	19.02	4.986
2123.14	3.326	30	14	46.66	47	4.92	4.928	4.915	19.02	4.927
1415.43	3.150	30	13	43.33	43	4.82	4.848	4.838	18.81	4.845
707.72	2.850	30	12	40.00	40	4.75	4.712	4.740	18.48	4.705
353.86	2.548	30	10	33.33	33	4.56	4.574	4.544	17.43	4.564

Results:

Y = 3.374793 + 0.4667058 X

Chi-squared is 3.478241E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 3.482295

LD_{50} is 3035.954µg/cm^2

95% confidence limits are 607.318 to15176.57µg/cm^2

Appendix Table 51. Dose-mortality effect of seed extract (chloroform) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	23	76.66	77	5.74	5.635	5.73	16.74	5.618
2123.14	3.326	30	21	70.00	70	5.52	5.549	5.50	17.43	5.536
1415.43	3.150	30	19	63.33	63	5.33	5.428	5.321	18.03	5.420
707.72	2.850	30	17	56.67	57	5.18	5.222	5.202	18.81	5.223
353.86	2.548	30	16	53.33	53	5.08	5.015	5.075	19.11	5.025

Results:

Y = 3.352186 + 0.6564545 X

Chi-squared is 0.4663468 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 2.510173

LD_{50} is 323.7229µg/cm²

95% confidence limits are 80.55176 to 1300.982µg/cm²

Appendix Table 52. Dose-mortality effect of seed extract (chloroform) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	26	86.67	87	6.13	6.010	6.087	13.17	5.989
2123.14	3.326	30	24	80.00	80	5.85	5.921	5.87	14.13	5.902
1415.43	3.150	30	23	76.67	77	5.74	5.796	5.734	15.96	5.780
707.72	2.850	30	21	70.00	70	5.52	5.583	5.5	17.43	5.570
353.86	2.548	30	20	66.67	67	5.44	5.369	5.422	18.48	5.361

Results:

Y = 3.588591 + 0.6954293 X

Chi-squared is 0.3290119with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} 2.029551

LD_{50} is 107.0412µg/cm²

95% confidence limits are 10.58678 to 1082.276µg/cm²

Appendix Table 53. Dose-mortality effect of seed extract (methanol) of *A. indica* against *T. castaneum* after 24h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	7	23.33	23	4.26	4.215	4.252	15.09	4.216
2123.14	3.326	30	6	20.00	20	4.16	4.159	4.17	14.13	4.160
1415.43	3.150	30	5	16.67	17	4.05	4.080	4.037	13.17	4.080
707.72	2.850	30	4	13.33	13	3.87	3.945	3.878	12.15	3.945
353.86	2.548	30	4	13.33	13	3.87	3.810	3.873	11.1	3.808

Results:

Y = 2.657667 + 0.4516222 X

Chi-squared is 0.1452589 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.186488

LD_{50} is 153634.4µg/cm^2

95% confidence limits are 46.15917 to 5.113498E+08 µg/cm^2

Appendix Table 54. Dose-mortality effect of seed extract (methanol) of *A. indica* against *T. castaneum* after 48h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	12	40.00	40	4.75	4.736	4.74	18.48	4.726
2123.14	3.326	30	11	36.67	37	4.67	4.668	4.659	18.03	4.658
1415.43	3.150	30	10	33.33	33	4.56	4.572	4.544	17.43	4.563
707.72	2.850	30	8	26.67	27	4.39	4.408	4.39	16.74	4.400
353.86	2.548	30	7	23.33	23	4.26	4.244	4.252	15.09	4.237

Results:

Y = 2.858917 + 0 .5408117 X

Chi-squared is 1.526594E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.959018

LD_{50} is 9099.51µg/cm^2

95% confidence limits are 675.918 to122501.8 µg/cm^2

Appendix Table 55. Dose-mortality effect of seed extract (methanol) of *A. indica* against *T. castaneum* after 72h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	17	56.67	57	5.18	5.144	5.165	19.02	5.132
2123.14	3.326	30	16	53.33	53	5.08	5.097	5.075	19.11	5.088
1415.43	3.150	30	15	50.00	50	5.00	5.032	5.00	19.11	5.028
707.72	2.850	30	14	46.67	47	4.92	4.920	4.915	19.02	4.924
353.86	2.548	30	13	43.33	43	4.82	4.807	4.838	18.81	4.820

Results:

Y = 3.942465 + 0.344479 X

Chi-squared is 4.725695E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.069955

LD_{50} is 1174.775µg/cm²

95% confidence limits are 306.4728 to 4503.159 µg/cm²

Appendix Table 56. Dose-mortality effect of seed extract (methanol) of *A. indica* against *T. castaneum* after 96h of exposure.

Dose µg/cm²	Log dose	#used	#Kill	%Kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
2830.86	3.452	30	21	70	70	5.52	5.482	5.51	18.03	5.471
2123.14	3.326	30	20	66.67	67	5.44	5.425	5.429	18.03	5.418
1415.43	3.150	30	18	60.00	60	5.25	5.346	5.24	18.48	5.343
707.72	2.850	30	18	60.00	60	5.25	5.211	5.28	18.81	5.214
353.86	2.548	30	16	53.33	53	5.08	5.075	5.075	19.11	5.086

Results

Y = 3.998091 + 0.426866 X

Chi-squared is 0.3080139 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.347128

LD_{50} is 222.3965µg/cm²

95% confidence limits are 16.25487 to 3042.791 µg/cm²

Appendix Table 225: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30 min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	23	25.556	26	4.36	4.452	4.36	50.22	4.432
100	1.999	90	19	21.111	21	4.19	4.177	4.208	42.39	4.176
50	1.699	90	15	16.667	17	4.05	3.902	4.062	36.45	3.920
25	1.398	90	8	8.889	9	3.66	3.627	3.663	27.18	3.664
12.5	1.097	90	4	4.445	4	3.25	3.352	3.254	18.72	3.409

Results:

Y = 2.47687+0.8495169X

Chi-squared is 1.48225 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.970076

LC_{50} is 933.4176

95% confidence limits are 279.0945 to 3121.768

Appendix Table 226: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	57	63.333	63	5.33	5.382	5.318	55.44	5.372
100	1.999	90	51	56.667	57	5.18	5.140	5.165	57.06	5.134
50	1.699	90	43	47.778	48	4.95	4.898	4.968	56.43	4.896
25	1.398	90	32	35.556	36	4.64	4.656	4.632	54.09	4.657
12.5	1.097	90	24	26.667	27	4.39	4.414	4.39	50.22	4.419

Results:

Y = 3.550312 + 0.7918972 X

Chi-squared is 0.5898743 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.830652

LC_{50} is 67.70986

95% confidence limits are 47.34112 to 96.84238

Appendix Table 227: Dose-mortality effect of flower extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	73	81.111	81	5.88	5.948	5.908	42.39	5.973
100	1.999	90	66	73.334	73	5.61	5.629	5.61	50.22	5.642
50	1.699	90	63	70	70	5.52	5.31	5.50	55.44	5.311
25	1.398	90	41	45.556	46	4.90	4.991	4.89	57.06	4.980
12.5	1.097	90	32	35.556	36	4.64	4.672	4.632	54.09	4.650

Results:

Y = 3.444655 + 1.098646X

Chi-squared is 2.688568 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.415693

LC_{50} is 26.04309

95% confidence limits are 19.34113 to 35.06738

Appendix Table 228: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30 min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	18	20	20	4.160	4.168	4.170	42.39	4.169
100	1.999	90	15	16.667	17	4.050	4.040	4.037	39.51	4.042
50	1.699	90	13	14.444	14	3.920	3.912	3.924	36.45	3.915
25	1.398	90	10	11.111	11	3.770	3.784	3.778	30.24	3.787
12.5	1.097	90	8	8.889	9	3.660	3.656	3.663	27.18	3.660

Results:

Y = 3.196 + 0.423X

Chi-squared is 7.055E-03 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 4.266008

LC_{50} is 18450.49

95% confidence limits are 140.349 tO 24255.31

Appendix Table 229: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	53	58.889	59	5.23	5.204	5.254	56.43	5.211
100	1.999	90	41	45.556	46	4.90	4.948	4.890	57.06	4.953
50	1.699	90	34	37.778	38	4.69	4.692	4.686	54.09	4.696
25	1.398	90	27	30.000	30	4.48	4.436	4.480	50.22	4.438
12.5	1.097	90	18	20.000	20	4.16	4.180	4.170	42.39	4.180

Results:

Y = 3.241139 + 0.8560416 X

Chi-squared is 0.4319 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.054644

LC_{50} is 113.4081

95% confidence limits are 75.2666 tO 170.878

Appendix Table 230: Dose-mortality effect of flower extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	71	78.889	79	5.81	5.800	5.766	45.27	5.774
100	1.999	90	63	70.000	70	5.52	5.536	5.500	52.29	5.519
50	1.699	90	55	61.111	61	5.28	5.272	5.306	56.43	5.263
25	1.398	90	45	50.000	50	5.00	5.008	5.000	57.33	5.007
12.5	1.097	90	36	40.000	40	4.75	4.744	4.740	55.44	4.752

Results:

Y = 3.820252 + 0.8492097 X

Chi-squared is 0.136219 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.38923

LC_{50} is 24.50362

95% confidence limits are 16.54686 tO 36.28648

Appendix Table 231: Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30 min.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	21	23.333	23	4.26	4.27	4.252	45.27	4.266
100	1.999	90	17	18.889	19	4.12	4.09	4.119	39.51	4.088
50	1.699	90	13	14.444	14	3.92	3.91	3.924	36.45	3.910
25	1.398	90	8	8.889	9	3.66	3.73	3.662	30.24	3.732
12.5	1.097	90	7	7.778	8	3.59	3.55	3.596	24.21	3.554

Results:

Y = 2.90469 + 0.5917074 X

Chi-squared is 0.2450733 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.541125

LC_{50} is 3476.365

95% confidence limits are 284.25 to 42515.8

Appendix Table 232: Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	52	57.778	58	5.2	5.200	5.19	57.06	5.194
100	1.999	90	45	50	50	5	4.100	4.99	57.06	4.996
50	1.699	90	39	43.333	43	4.82	4.8	4.838	56.43	4.798
25	1.398	90	30	33.333	33	4.56	4.600	4.551	54.09	4.600
12.5	1.097	90	25	27.778	28	4.42	4.400	4.42	50.22	4.402

Results:

Y = 3.679797 + 0.6580409 X

Chi-squared is 0.2389603 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.006263

LC_{50} is 101.4525

95% confidence limits are 61.43791 to167.5287

Appendix Table 233: Dose-mortality effect of leaf extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	62	68.889	69	5.5	5.484	5.483	54.09	5.483
100	1.999	90	54	60	60	5.25	5.236	5.28	56.43	5.237
50	1.699	90	42	46.667	47	4.92	4.988	4.915	57.06	4.991
25	1.398	90	37	41.111	41	4.77	4.740	4.766	55.44	4.744
12.5	1.097	90	28	31.111	31	4.5	4.492	4.51	50.22	4.498

Results:

Y = 3.600151 + 0.8182286 X

Chi-squared is 0.4642525 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.710829

LC_{50} is 51.38413

95% confidence limits are 36.80554 to 71.73725

Appendix Table 234: Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30 min.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	21	23.333	23	4.26	4.218	4.252	45.27	4.214
100	1.999	90	15	16.667	17	4.05	4.075	4.037	39.51	4.073
50	1.699	90	12	13.333	13	3.87	3.932	3.878	36.45	3.932
25	1.398	90	11	12.222	12	3.82	3.789	3.836	30.24	3.791
12.5	1.097	90	8	8.889	9	3.66	3.646	3.663	27.18	3.650

Results:

Y = 3.137479 + 0.4678234 X

Chi-squared is 0.2887631 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.981249

LC_{50} is 9577.411

95% confidence limits are 202.3107 to 453396.3

Appendix Table 235: Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	37	41.111	41	4.77	4.718	4.766	55.44	4.723
100	1.999	90	27	30.000	30	4.48	4.491	4.48	50.22	4.491
50	1.699	90	19	21.111	21	4.19	4.264	4.184	45.27	4.258
25	1.398	90	14	15.556	16	4.01	4.037	3.996	39.51	4.025
12.5	1.097	90	12	13.333	13	3.87	3.81	3.873	33.3	3.793

Results:

Y = 2.944775 + 0.7729489 X

Chi-squared is 0.6033821 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.65894

LC_{50} is 455.9743

95% confidence limits are 182.6174 to 1138.515

Appendix Table 236: Dose-mortality effect of leaf extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	50	55.556	56	5.15	5.068	5.15	57.33	5.077
100	1.999	90	39	43.333	43	4.82	4.831	4.838	56.43	4.837
50	1.699	90	27	30.000	30	4.48	4.594	4.46	52.29	4.596
25	1.398	90	22	24.444	24	4.29	4.357	4.298	47.88	4.355
12.5	1.097	90	20	22.222	22	4.23	4.120	4.246	42.39	4.114

Results:

Y = 3.23722 + 0.7996599 X

Chi-squared is 2.159527 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.204413

LC_{50} is 160.1078

95% confidence limits are 94.85948 to 270.237

Appendix Table 237: Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	28	31.111	31	4.5	4.484	4.51	50.22	4.484
100	1.999	90	20	22.222	22	4.23	4.261	4.218	45.27	4.260
50	1.699	90	15	16.667	17	4.05	4.038	4.037	39.51	4.036
25	1.398	90	11	12.222	12	3.82	3.815	3.822	33.3	3.812
12.5	1.097	90	7	7.778	8	3.59	3.592	3.596	24.21	3.588

Results:

Y = 2.771625 + 0.7441186 X

Chi-squared is 0.1191883 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.994651

LC_{50} is 987.7583

95% confidence limits are 255.7718 to 3814.602

Appendix Table 238: Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	62	68.889	69	5. 5	5.562	5.472	52.29	5.539
100	1.999	90	60	66.667	67	5.44	5.368	5.422	55.44	5.349
50	1.699	90	51	56.667	57	5.18	5.174	5. 165	57.06	5.159
25	1.398	90	45	50.00	50	5.00	4.980	4.99	57.06	4.968
12.5	1.097	90	36	40.00	40	4.75	4.786	4.74	55.44	4.778

Results:

Y = 4.084707 + 0.6321671 X

Chi-squared is 0.6404152 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.447866

LC_{50} is 28.04569

95% confidence limits are 17.10174 to 45.99303

Appendix Table 239: Dose-mortality effect of root bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	72	80	80	5.85	5.774	5.83	47.88	5.753
100	1.999	90	61	67.778	68	5.47	5.524	5.444	52.29	5.510
50	1.699	90	53	58.889	59	5.23	5.274	5.254	56.43	5.268
25	1.398	90	44	48.889	49	4.97	5.024	4.975	57.33	5.025
12.5	1.097	90	40	44.444	44	4.85	4.774	4.844	55.44	4.783

Results:

Y = 3.8988 + 0.8057047 X

Chi-squared is 0.8789311 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.366754

LC_{50} is 23.26771

95% confidence limits are 15.22007 to 35.57056

Appendix Table 240: Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	27	30	30	4.48	4.490	4.48	50.22	4.483
100	1.999	90	21	23.333	23	4.26	4.268	4.252	45.27	4.265
50	1.699	90	16	17.778	18	4.08	4.046	4.078	39.51	4.046
25	1.398	90	11	12.222	12	3.82	3.824	3.822	33.3	3.827
12.5	1.097	90	7	7.778	8	3.59	3.602	3.596	27.18	3.609

Results:

Y = 2.812564 + 0.7260228 X

Chi-squared is 5.345154E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.012903

LC_{50} is 1030.155

95% confidence limits are 257.5667 to 4120.177

Appendix Table 241: Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	59	65.556	66	5.41	5.384	5.396	55.44	5.372
100	1.999	90	51	56.667	57	5.18	5.177	5.165	57.06	5.165
50	1.699	90	42	46.667	47	4.92	4.970	4.915	57.06	4.957
25	1.398	90	36	40	40	4.75	4.763	4.74	55.44	4.749
12.5	1.097	90	31	34.445	34	4.59	4.556	4.572	52.29	4.542

Results:

Y = 3.785082 + 0.6897956 X

Chi-squared is 0.1849041 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.761273

LC_{50} is 57.71285

95% confidence limits are 38.8379 to 85.76086

Appendix Table 242: Dose-mortality effect of root bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	69	76.667	77	5.74	5.730	5.734	47.88	5.725
100	1.999	90	62	68.889	69	5.5	5.481	5.483	54.09	5.477
50	1.699	90	52	57.778	58	5.2	5.232	5.228	56.43	5.230
25	1.398	90	43	47.778	48	4.95	4.983	4.94	57.06	4.982
12.5	1.097	90	37	41.111	41	4.77	4.734	4.766	55.44	4.734

Results:

Y = 3.831306 + 0.823082 X

Chi-squared is 0.1627083 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.419901

LC_{50} is 26.29665

95% confidence limits are 17.77245 to 38.90932

Appendix Table 243: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30 min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	26	28.899	29	4.45	4.428	4.45	50.22	4.425
50	1.699	90	20	22.222	22	4.23	4.241	4.218	45.27	4.237
25	1.398	90	15	16.667	17	4.05	4.054	4.037	39.51	4.050
12.5	1.097	90	11	12.222	12	3.82	3.867	3.822	33.33	3.863
6.25	0.796	90	9	10	10	3.72	3.68	3.73	27.18	3.675

Results:

Y = 3.179498 + 0.622738 X

Chi-squared is 0.1919718 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.923384

LC_{50} is 838.2706

95% confidence limits are 129.3991 to 5430.47

Appendix Table 244: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	56	62.222	62	5.31	5.288	5.332	56.43	5.301
50	1.699	90	48	53.333	53	5.08	5.085	5.075	57.33	5.094
25	1.398	90	40	4.445	44	4.85	4.882	4.864	56.43	4.887
12.5	1.097	90	33	36.667	37	4.67	4.679	4.659	54.09	4.681
6.25	0.796	90	28	31.111	31	4.5	4.476	4.51	50.22	4.474

Results:

Y = 3.92757 + 0.6865575 X

Chi-squared is 0.1976204 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.56204

LC_{50} is 36.47875

95% confidence limits are 23.963 to 55.53144

Appendix Table 245: Dose-mortality effect of root wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	73	81.111	81	5.88	5.898	5.834	45.27	5.866
50	1.699	90	67	74.444	74	5.64	5.646	5.640	50.22	5.623
25	1.398	90	61	67.778	68	5.47	5.394	5.448	55.44	5.381
12.5	1.097	90	48	53.333	53	5.08	5.142	5.065	57.06	5.139
6.25	0.796	90	41	45.556	46	4.9	4.890	4.916	56.43	4.897

Results:

Y = 4.256125 + 0.8047338 X

Chi-squared is 0.6388855 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 0.9243744

LC_{50} is 8.40184

95% confidence limits are 5.092476 to 13.86181

Appendix Table 246: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	16	17.778	18	4.08	4.018	4.078	39.51	4.031
50	1.699	90	11	12.222	12	3.82	3.855	3.822	33.3	3.861
25	1.398	90	8	8.889	9	3.66	3.692	3.663	27.18	3.691
12.5	1.097	90	5	5.556	6	3.45	3.529	3.442	24.21	3.521
6.25	0.796	90	5	5.556	6	3.45	3.366	3.466	18.72	3.351

Results:

Y = 2.901641 + 0.5648441 X

Chi-squared is 0.5579266 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.714936

LC_{50} is 5187.234

95% confidence limits are 139.4302 to 192981.3

Appendix Table 247: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	49	54.445	54	5.1	5.048	5.1	57.33	5.058
50	1.699	90	39	43.333	43	4.82	4.84	4.838	56.43	4.845
25	1.398	90	30	33.333	33	4.56	4.632	4.551	54.09	4.631
12.5	1.097	90	25	27.778	28	4.42	4.424	4.42	50.22	4.418
6.25	0.796	90	21	23.333	23	4.26	4.216	4.252	45.27	4.205

Results:

Y = 3.639803 + 0.7090863 X

Chi-squared is 0.5536576 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.91824

LC_{50} is 82.83993

95% confidence limits are 45.49681 to150.8338

Appendix Table 248: Dose-mortality effect of root wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	65	72.222	72	5.58	5.574	5.556	52.29	5.558
50	1.699	90	57	63.333	63	5.33	5.306	5.318	55.44	5.292
25	1.398	90	45	50.00	50	5	5.038	5.00	57.33	5.026
12.5	1.097	90	36	40.00	40	4.75	4.770	4.74	55.44	4.760
6.25	0.796	90	29	32.222	32	4.53	4.502	4.516	52.29	4.493

Results:

Y = 3.789963 + 0.8838996 X

Chi-squared is 0.123806 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.368976

LC_{50} is 23.38707

95% confidence limits are 17.1553 to 31.88254

Appendix Table 249: Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	27	30.000	30	4.48	4.484	4.48	50.22	4.477
50	1.699	90	21	23.333	23	4.26	4.262	4.252	45.27	4.257
25	1.398	90	15	16.667	17	4.05	4.04	4.037	39.51	4.037
12.5	1.097	90	11	12.222	12	3.82	3.818	3.822	33.3	3.818
6.25	0.796	90	7	7.778	8	3.59	3.596	3.596	24.21	3.598

Results:

Y = 3.016606 + 0.7302255 X

Chi-squared is 2.811432E-03 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.71614

LC_{50} is 520.1635

95% confidence limits are 128.2581 to 2109.577

Appendix Table 250: Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	63	70	70	5.52	5.506	5.5	52.29	5.509
50	1.699	90	54	60	60	5.25	5.258	5.28	56.43	5.259
25	1.398	90	45	50	50	5.00	5.01	5.00	57.33	5.007
12.5	1.097	90	36	40	40	4.75	4.762	4.74	55.44	4.756
6.25	0.796	90	29	32.222	32	4.53	4.514	4.516	52.29	4.506

Results:

Y = 3.841274 + 0.8340478 X

Chi-squared is 5.490875E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.389281

LC_{50} is 24.50645

95% confidence limits are 17.66922 to 33.98941

Appendix Table 251: Dose-mortality effect of seed extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	76	84.444	84	5.99	6.006	5.964	39.51	5.988
50	1.699	90	70	77.778	78	5.77	5.756	5.766	47.88	5.745
25	1.398	90	63	70.000	70	5.52	5.506	5.5	52.29	5.503
12.5	1.097	90	54	60.000	60	5.25	5.256	5.28	56.43	5.260
6.25	0.796	90	45	50.000	50	5.00	5.006	5.00	57.33	5.018

Results:

Y = 4.376288 + 0.8058436 X

Chi-squared is 8.293724E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 0.7739871

LC_{50} is 5.942745

95% confidence limits are 3.268797 to 10.80404

Appendix Table 252: Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30 min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	23	25.556	26	4.36	4.376	4.362	47.88	4.382
50	1.699	90	19	21.111	21	4.19	4.18	4.208	42.39	4.187
25	1.398	90	14	15.556	16	4.01	3.984	4.016	36.45	3.993
12.5	1.097	90	10	11.111	11	3.77	3.788	3.778	30.24	3.798
6.25	0.796	90	7	7.778	8	3.59	3.592	3.596	24.21	3.604

Results:

Y = 3.089669 + 0.6459533 X

Chi-squared is 7.057381E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.957382

LC_{50} is 906.5301

95% confidence limits are 134.699 to 6100.985

Appendix Table 253: Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	50	55.556	56	5.15	5.134	5.14	57.06	5.124
50	1.699	90	43	47.778	48	4.95	4.958	4.94	57.06	4.949
25	1.398	90	37	41.111	41	4.77	4.782	4.766	55.44	4.775
12.5	1.097	90	31	34.444	34	4.59	4.606	4.578	54.09	4.600
6.25	0.796	90	26	28.889	29	4.45	4.43	4.45	50.22	4.425

Results:

Y = 3.963283 + 0.5802853 X

Chi-squared is 7.998276E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.786564

LC_{50} is 61.17362

95% confidence limits are 32.74389 to 114.2874

Appendix Table 254: Dose-mortality effect of seed extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
100	1.999	90	75	83.333	83	5.95	5.874	5.902	45.27	5.827
50	1.699	90	59	65.556	66	5.41	5.521	5.388	52.29	5.490
25	1.398	90	51	56.667	57	5.18	5.168	5.165	57.06	5.153
12.5	1.097	90	39	43.333	43	4.82	4.815	4.838	56.43	4.816
6.25	0.796	90	27	30.000	30	4.48	4.462	4.48	50.22	4.479

Results:

Y = 3.587074 + 1.120292 X

Chi-squared is 0.8352814 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.261213

LC_{50} is 18.24789

95% confidence limits are 14.0748 to 23.65829

Appendix Table 255: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30 min.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	28	31.111	31	4.5	4.482	4.51	50.22	4.487
100	1.999	90	21	23.333	23	4.26	4.27	4.252	45.27	4.271
50	1.699	90	15	16.667	17	4.05	4.058	4.037	39.51	4.056
25	1.398	90	11	12.222	12	3.82	3.846	3.822	33.3	3.840
12.5	1.097	90	8	8.889	9	3.66	3.634	3.663	27.18	3.625

Results:

Y = 2.839485 + 0.7158541 X

Chi-squared is 0.1087818 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.018095

LC_{50} is 1042.544

95% confidence limits are 254.2184 to 4275.452

Appendix Table 256: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	45	50	50	5	5.010	5	57.33	5.005
100	1.999	90	37	41.111	41	4.77	4.775	4.766	55.44	4.771
50	1.699	90	31	34.444	34	4.59	4.54	4.572	52.29	4.536
25	1.398	90	21	23.333	23	4.26	4.305	4.266	47.88	4.302
12.5	1.097	90	16	17.778	18	4.08	4.070	4.078	39.51	4.067

Results:

Y = 2.839485 + 0.7158541 X

Chi-squared is 0.1355801 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.294208

LC_{50} is 196.883

95% confidence limits are 108.0891 to 358.6199

Appendix Table 257: Dose-mortality effect of stem bark extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	75	83.333	83	5.95	5.886	5.902	45.27	5.850
100	1.999	90	64	71.111	71	5.55	5.592	5.528	52.29	5.569
50	1.699	90	54	60	60	5.25	5.298	5.28	56.43	5.289
25	1.398	90	44	48.889	49	4.97	5.004	4.975	57.33	5.008
12.5	1.097	90	37	41.111	41	4.77	4.71	4.766	55.44	4.727

Results:

Y = 3.702853 + 0.9333245 X

Chi-squared is 0.3615456 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.389813

LC_{50} is 24.53654

95% confidence limits are 17.16986 to 35.06388

Appendix Table 258: Dose-mortality effect of stem bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30min.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	19	21.111	21	4.19	4.188	4.208	42.39	4.193
100	1.999	90	14	15.556	16	4.01	4.0250	3.996	39.51	4.029
50	1.699	90	12	13.333	13	3.87	3.862	3.873	33.3	3.865
25	1.398	90	9	10.000	10	3.72	3.699	3.73	27.18	3.700
12.5	1.097	90	6	6.667	7	3.52	3.536	3.519	24.21	3.536

Results:

Y = 2.937893 + 0.5454845 X

Chi-squared is 8.504391E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.780322

LC_{50} is 6030.069

95% confidence limits are 262.4669 to 138538.1

Appendix Table 259: Dose-mortality effect of stem bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	48	53.333	53	5.08	5.048	5.075	57.33	5.052
100	1.999	90	40	44.444	44	4.85	4.847	4.864	56.43	4.848
50	1.699	90	31	34.444	34	4.59	4.646	4.578	54.09	4.644
25	1.398	90	25	27.778	28	4.42	4.445	4.42	50.22	4.440
12.5	1.097	90	22	24.444	24	4.29	4.244	4.286	45.27	4.236

Results:

Y = 3.492004 + 0.6778593 X

Chi-squared is 0.4133606with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 2.224645

LC_{50} is 167.7432

95% confidence limits are 89.26254 to 315.2249

Appendix Table 260: Dose-mortality effect of stem bark extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	69	76.667	77	5.74	5.722	5.734	47.88	5.713
100	1.999	90	60	66.667	67	5.44	5.440	5.429	54.09	5.433
50	1.699	90	49	54.444	54	5.1	5.158	5.09	57.06	5.153
25	1.398	90	42	46.667	47	4.92	4.876	4.942	56.43	4.873
12.5	1.097	90	31	34.444	34	4.59	4.594	4.572	52.29	4.593

Results:

Y = 3.571618 + 0.9307811 X

Chi-squared is 0.5406418with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.534606

LC_{50} is 34.24572

95% confidence limits are 24.98084 to46.94675

Appendix Table 261: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 30min. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	22	24.445	24	4.29	4.228	4.286	45.27	4.228
100	1.999	90	14	15.556	16	4.01	4.045	3.996	39.51	4.045
50	1.699	90	10	11.111	11	3.77	3.862	3.771	33.3	3.862
25	1.398	90	9	10.000	10	3.72	3.679	3.73	27.18	3.679
12.5	1.097	90	6	6.667	7	3.52	3.496	3.54	21.42	3.496

Results:

Y = 2.828171 + 0.6084348 X

Chi-squared is 0.6351118 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.569536

LC_{50} is 3711.381

95% confidence limits are 297.0884 to 46364.49

Appendix Table 262: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 24h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	53	58.889	59	5.23	5.230	5.254	56.43	5.243
100	1.999	90	47	52.222	52	5.05	5.013	5.05	57.33	5.020
50	1.699	90	36	40	40	4.75	4.796	4.74	55.44	4.796
25	1.398	90	30	33.333	33	4.56	4.579	4.544	52.29	4.572
12.5	1.097	90	24	26.667	27	4.39	4.362	4.394	47.88	4.348

Results:

Y = 3.532413 + 0.7435739 X

Chi-squared is 0.3724823 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.973695

LC_{50} is 94.12271

95% confidence limits are 61.21007 to 144.7326

Appendix Table 263: Dose-mortality effect of stem wood extract (chloroform) of *Azadirachta indica* against *Artemia salina* after 48h. of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	66	73.333	73	5.61	5.556	5.584	52.29	5.536
100	1.999	90	56	62.222	62	5.31	5.303	5.292	55.44	5.289
50	1.699	90	44	48.889	49	4.97	5.05	4.975	57.33	5.037
25	1.398	90	35	38.889	39	4.72	4.797	4.714	55.44	4.787
12.5	1.097	90	32	35.556	36	4.64	4.544	4.628	52.29	4.538

Results:

Y = 3.628589 + 0.8287544 X

Chi-squared is 1.065525 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.654787

LC_{50} is 45.16339

95% confidence limits are 32.41925 to 62.91725

Appendix Table 264: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 30min of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	25	27.778	28	4.42	4.428	4.42	50.22	4.421
100	1.999	90	20	22.222	22	4.23	4.235	4.218	45.27	4.231
50	1.699	90	16	17.778	18	4.08	4.042	4.078	39.51	4.040
25	1.398	90	11	12.222	12	3.82	3.849	3.822	33.3	3.850
12.5	1.097	90	8	8.889	9	3.66	3.656	3.663	27.18	3.659

Results:

Y = 2.965103 + 0.6329139 X

Chi-squared is 8.963585E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 3.215125

LC_{50} is 1641.063

95% confidence limits are 263.9264 to 10203.94

Appendix Table 265: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 24h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	53	58.889	59	5.23	5.226	5.254	56.43	5.242
100	1.999	90	46	51.111	51	5.03	5.014	5.025	57.33	5.024
50	1.699	90	37	41.111	41	4.77	4.802	4.786	56.43	4.806
25	1.398	90	31	34.444	34	4.59	4.59	4.572	52.29	4.588
12.5	1.097	90	24	26.667	27	4.39	4.378	4.394	47.88	4.370

Results:

Y = 3.575806 + 0.7239164 X

Chi-squared is 7.130241E-02 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.967347

LC_{50} is 92.75699

95% confidence limits are 59.8282 to 143.8093

Appendix Table 266: Dose-mortality effect of stem wood extract (methanol) of *Azadirachta indica* against *Artemia salina* after 48h.of exposure.

Dose ppm	Log dose	#used	#Kill	%Kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
200	2.301	90	65	72.222	72	5.58	5.522	5.556	52.29	5.520
100	1.999	90	53	58.889	59	5.23	5.27	5.254	56.43	5.266
50	1.699	90	44	48.889	49	4.97	5.018	4.975	57.33	5.013
25	1.398	90	36	40.000	40	4.75	4.766	4.74	55.44	4.759
12.5	1.097	90	30	33.333	33	4.56	4.514	4.544	52.29	4.506

Results:

Y = 3.580152 + 0.8431652 X

Chi-squared is 0.2553291 with 3 degrees of freedom

No significant heterogeneity

Log LC_{50} is 1.68395

LC_{50} is 48.30029

95% confidence limits are 34.94275 to 66.76403

Appendix Table 57: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.796	4.922	18.48	4.806
6	1.778	30	8	26.667	27	4.39	4.504	4.376	17.43	4.504
3	1.477	30	6	20.000	20	4.16	4.212	4.15	15.09	4.203
1.50	1.176	30	4	13.333	13	3.87	3.92	3.878	12.15	3.902
0.75	0.875	30	3	10.000	10	3.72	3.628	3.73	9.060	3.600

Results:

Y = 2.724367 + 1.001031 X

Chi-squared is 0.7385426 with 3 degrees of freedom

No significant heterogeneity
Log LD_{50} is 2.27329
LD_{50} is 18.76248 mg/gm
95% confidence limits are 6.555629 to 53.69897 mg/gm

Appendix Table 58: Larvicidal effect of flower extract (chloroform**) of *A. indica* against *T. castaneum* larva (1st instar) after 48 h of exposure.**

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	4.974	5.165	19.02	4.980
6	1.778	30	10	33.333	33	4.56	4.718	4.558	18.48	4.717
3	1.477	30	7	23.333	23	4.26	4.462	4.27	16.74	4.455
1.50	1.176	30	7	23.333	23	4.26	4.206	4.252	15.09	4.192
0.75	0.875	30	5	16.667	17	4.05	3.950	4.062	12.15	3.930

Results:

Y = 3.166985 + 0.8717883 X

Chi-squared is 1.959259 with 3 degrees of freedom

No significant heterogeneity
Log LD_{50} is 2.102592
LD_{50} is 12.6646mg/gm
95% confidence limits are 4.819793 to 33.27781 mg/gm

Appendix Table 59: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	19	63.333	63	5.33	5.186	5.315	19.02	5.176
6	1.778	30	13	43.333	43	4.82	4.929	4.815	19.02	4.921
3	1.477	30	10	33.333	33	4.56	4.672	4.551	18.03	4.666
1.50	1.176	30	8	26.667	27	4.39	4.415	4.39	16.74	4.411
0.75	0.875	30	7	23.333	23	4.26	4.158	4.284	14.13	4.156

Results:

Y = 3.41474 + 0.8471829 X

Chi-squared is 1.058361 with 3 degrees of freedom

No significant heterogeneity
Log LD_{50} is 1.871214
LD_{50} is 7.433853mg/gm
95% confidence limits are 3.518182 to 15.7 mg/gm

Appendix Table 60: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.540	4.74	17.43	4.567
6	1.778	30	5	16.667	17	4.05	4.261	4.048	15.09	4.271
3	1.477	30	4	13.333	13	3.87	3.982	3.878	12.15	3.975
1.50	1.176	30	3	10.000	10	3.72	3.703	3.72	10.08	3.679
0.75	0.875	30	2	6.667	7	3.52	3.424	3.54	7.140	3.383

Results:

Y = 2.522279 + 0.9835311 X

Chi-squared is 1.57912 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.51921
LD_{50} is 33.052883 mg/gm
95% confidence limits are 7.830971 to 139.5093 mg/gm

Appendix Table 61: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.766	4.922	18.48	4.774
6	1.778	30	8	26.667	27	4.39	4.522	4.376	17.43	5.522
3	1.477	30	6	20.000	20	4.16	4.278	4.15	15.09	4.269
1.50	1.176	30	5	16.667	17	4.05	4.034	4.037	13.17	4.016
0.75	0.875	30	4	13.333	13	3.87	3.790	3.894	10.08	3.763

Results:

Y = 3.028001 + 0.8399352 X

Chi-squared is 1.16333 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.347799
LD_{50} is 22.274063 mg/gm
95% confidence limits are 5.815 to 85.323 mg/gm

Appendix Table 62: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	4.978	5.165	19.02	4.978
6	1.778	30	10	33.333	33	4.56	4.744	4.558	18.48	4.737
3	1.477	30	8	26.667	27	4.39	4.51	4.376	17.43	4.496
1.50	1.176	30	7	23.333	23	4.26	4.276	4.252	15.09	4.255
0.75	0.875	30	6	20.000	20	4.16	4.042	4.16	13.17	4.014

Results:

Y = 3.313887 + 0 .8005681 X

Chi-squared is 1.789371 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.106146
LD_{50} is 12.768684 mg/gm
95% confidence limits are 4.458 to 36.568 mg/gm

Appendix Table 63:Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.52	4.544	17.43	4.524
6	1.778	30	6	20.000	20	4.16	4.168	4.170	14.13	4.168
3	1.477	30	3	10.000	10	3.72	3.816	3.72	11.10	3.813
1.50	1.176	30	2	6.667	7	3.52	3.464	3.54	7.140	3.457
0.75	0.875	30	1	3.333	3	3.12	3.112	3.116	4.62	3.102

Results:

Y = 2.068102 + 1.181153 X

Chi-squared is 0.1525726 with 3 degrees of freedom

No significant heterogeneity
Log LD_{50} is 2.482233
LD_{50} is 30.355246 mg/gm
95% confidence limits are 8.967541 to 102.7525 mg/gm

Appendix Table 64: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	#Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.766	4.922	18.48	4.774
6	1.778	30	8	26.667	27	4.39	4.522	4.376	17.43	4.522
3	1.477	30	6	20.000	20	4.16	4.278	4.15	15.09	4.269
1.50	1.176	30	5	16.667	17	4.05	4.034	4.037	13.17	4.016
0.75	0.875	30	4	13.333	13	3.87	3.790	3.894	10.08	3.763

Results:

Y = 3.028001 + 0.8399352 X

Chi-squared is 1.16333 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.347799
LD_{50} is 22.274065mg/gm
95% confidence limits are 5.814798 to 85.32262 mg/gm

Appendix Table 65: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	5.022	5.175	19.11	5.026
6	1.778	30	11	36.667	37	4.67	4.777	4.662	18.48	4.773
3	1.477	30	8	26.667	27	4.39	4.532	4.376	17.43	4.520
1.50	1.176	30	7	23.333	23	4.26	4.287	4.252	15.09	4.267
0.75	0.875	30	6	20	20	4.16	4.042	4.16	13.17	4.013

Results:

Y= 3.277468 +0.8410256X

Chi-squared is 1.297401 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.048133
LD_{50} is 11.172065mg/gm
95% confidence limits are 4.385187 to 28.46285 mg/gm

Appendix Table 66: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.47	4.57	16.74	4.488
6	1.778	30	6	20.000	20	4.16	4.218	4.15	15.09	4.227
3	1.477	30	4	13.333	13	3.87	3.966	3.878	12.15	3.965
1.50	1.176	30	3	10.000	10	3.72	3.714	3.72	10.08	3.704
0.75	0.875	30	2	6.667	7	3.52	3.462	3.54	7.140	3.442

Results:

Y=2.681601+0.8691215X

Chi-squared is 0.3641524 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.66752
LD_{50} is 46.507186mg/gm
95% confidence limits are 7.134844 to 303.1486 mg/gm

Appendix Table 67: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.736	4.818	18.48	4.740
6	1.778	30	8	26.667	27	4.39	4.482	4.39	16.74	4.483
3	1.477	30	6	20.000	20	4.16	4.228	4.15	15.09	4.225
1.50	1.176	30	5	16.667	17	4.05	3.974	4.062	12.15	3.968
0.75	0.875	30	3	10.000	10	3.72	3.72	3.72	10.08	3.710

Results:

Y = 2.962304 + 0.8549606 X

Chi-squared is 0.4499512 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.38338

LD_{50} is 24.175765mg/gm

95% confidence limits are 6.122689 to 95.45926 mg/gm

Appendix Table 68: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum larva* (1stinstar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.818	4.942	18.81	4.839
6	1.778	30	10	33.333	33	4.56	4.604	4.551	18.03	4.617
3	1.477	30	7	23.333	23	4.26	4.390	4.266	15.96	4.395
1.50	1.176	30	6	20.000	20	4.16	4.176	4.17	14.13	4.173
0.75	0.875	30	5	16.667	17	4.05	3.962	4.062	12.15	3.950

Results:

Y = 3.30455 + 0.7381548 X

Chi-squared is 0.6934762 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.296876

LD_{50} is 19.809616mg/gm

95% confidence limits are 4.792457 to 81.88301 mg/gm

Appendix Table 69: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.703	4.922	18.48	4.702
6	1.778	30	7	13.333	13	3.87	4.256	3.912	15.09	4.256
3	1.477	30	3	10.000	10	3.72	3.810	3.72	11.10	3.811
1.50	1.176	30	2	6.667	7	3.52	3.364	3.572	6.24	3.365
0.75	0.875	30	1	3.333	3	3.12	2.917	3.172	3.30	2.920

Results:

Y=1.624853 +1.479758 X

Chi-squared is 3.253348 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.280877
LD_{50} is 19.093132mg/gm
95% confidence limits are 8.733655 to 41.74056 mg/gm

Appendix Table 70: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	4.878	5.202	18.81	4.933
6	1.778	30	9	23.333	23	4.26	4.547	4.264	17.43	4.568
3	1.477	30	5	16.667	17	4.05	4.216	4.048	15.09	4.204
1.50	1.176	30	4	13.333	13	3.87	3.885	3.873	11.10	3.839
0.75	0.875	30	3	10.000	10	3.72	3.554	3.75	8.07	3.475

Results:

Y = 2.415029 + 1.211011 X

Chi-squared is 3.967207 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.134556
LD_{50} is 13.631894mg/gm
95% confidence limits are 6.482191 to 28.66753 mg/gm

Appendix Table 71: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	19	63.333	63	5.33	5.152	5.315	19.02	5.152
6	1.778	30	11	36.667	37	4.67	4.868	4.682	18.81	4.832
3	1.477	30	9	30.000	30	4.48	4.584	4.460	17.43	4.571
1.50	1.176	30	8	26.667	27	4.39	4.300	4.388	15.09	4.281
0.75	0.875	30	5	16.667	17	4.05	4.016	4.037	13.17	3.991

Results:

Y= 3.147225 + 0.9641664X

Chi-squared is 1.529515 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.921634

LD_{50} is 8.348996mg/gm

95% confidence limits are 4.142447 to 16.82719 mg/gm

Appendix Table 72: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.762	4.922	18.48	4.789
6	1.778	30	7	23.333	23	4.26	4.428	4.27	16.74	4.437
3	1.477	30	5	16.667	17	4.05	4.094	4.037	13.17	4.086
1.50	1.176	30	3	10.000	10	3.72	3.760	3.72	10.08	3.734
0.75	0.875	30	2	6.667	7	3.52	3.426	3.54	7.140	3.382

Results:

Y = 2.359889 + 1.168336 X

Chi-squared is 1.006329 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.259719

LD_{50} is 18.185246 mg/gm

95% confidence limits are 7.342437 to 45.03997 mg/gm

Appendix Table 73: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.902	4.990	19.02	4.901
6	1.778	30	10	33.333	33	4.56	4.636	4.551	18.03	4.634
3	1.477	30	7	23.333	23	4.26	4.37	4.266	15.96	4.367
1.50	1.176	30	6	20.000	20	4.16	4.104	4.17	14.13	4.100
0.75	0.875	30	4	13.333	13	3.87	3.838	3.873	11.10	3.833

Results:

Y = 3.056801 + 0.8871393 X

Chi-squared is 0.5246335 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.19041
LD_{50} is 15.502789 mg/gm
95% confidence limits are 5.389241 to 44.59557 mg/gm

Appendix Table 74: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.114	5.24	19.02	5.119
6	1.778	30	12	40.000	40	4.75	4.847	4.76	18.81	4.845
3	1.477	30	9	30.000	30	4.48	4.58	4.46	17.43	4.572
1.50	1.176	30	7	23.333	23	4.26	4.313	4.266	15.96	4.299
0.75	0.875	30	6	20.000	20	4.16	4.046	4.16	13.17	4.026

Results:

Y = 3.231305 + 0.9076765 X

Chi-squared is 0.891757 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.948597
LD_{50} is 8.883758 mg/gm
95% confidence limits are 4.117786 to 19.16593 mg/gm

Appendix Table 75: Larvicidal effect of root wood extract ($CHCl_3$) of *A. indica* against *T. castaneum* larvae (1st instar) after 24h of exposure.

:

Dose	Log dose	# used	# Kill	%kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	22	71.000	71	5.53	5.448	5.51	18.03	5.454
6	1.778	30	16	54.334	54	5.09	5.015	5.075	19.11	5.011
3	1.477	30	8	27.000	27	4.40	4.582	4.376	17.43	4.569
1.5	1.176	30	6	17.000	17	4.05	4.149	4.056	14.13	4.126
0.75	0.875	30	4	13.333	13	3.87	3.716	3.894	10.08	3.684

Results:

Y = 2.397 +1.470 X
Chi-squared is 2.29703 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.770491
LD_{50} is 7.996566 mg/g
95% confidence limits are 3.984326 to 8.723152 mg/g

Appendix Table 76:Larvicidal effect of root wood extract ($CHCl_3$) of *Derris indica* against *T. castaneum* larvae (1st instar) after 48h of exposure.

Dose	Log dose	# used	# Kill	%kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	23	77.000	77	5.74	5.682	5.730	16.74	5.680
6	1.778	30	18	60.000	60	5.25	5.199	5.240	19.02	5.190
3	1.477	30	11	33.333	33	4.56	4.716	4.558	18.48	4.700
1.5	1.176	30	6	20.000	20	4.16	4.233	4.150	15.09	4.210
0.75	.875	30	4	13.333	13	3.87	3.750	3.894	10.08	3.720

Results:

Y = 2.295 +1.628 X
Chi-squared is 0.82403 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.661
LD_{50} is 5.584668 mg/g
95% confidence limits are 3.313658 to 7.343968 mg/g

Appendix Table 77: Larvicidal effect of root wood extract ($CHCl_3$) of *Derris indica* against *T. castaneum* larvae (1st instar) after 72h of exposure.

Dose	Log dose	# used	# Kill	%kill	Corr%	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	26	84.356	84	5.95	5.842	5.902	15.09	5.805
6	1.778	30	20	63.333	63	5.33	5.368	5.318	18.48	5.340
3	1.477	30	13	40.000	40	4.75	4.894	4.76	18.81	4.874
1.5	1.176	30	9	26.667	27	4.39	4.420	4.39	16.74	4.410
0.75	0.875	30	5	17.000	17	4.05	3.946	4.062	12.15	3.944

Results:

Y = 2.594 + 1.547 X
Chi-squared is 0.687456 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.558179
LD_{50} is 4.546655 m/g
95% confidence limits are 2.608365 to 5.013438 mg/g

Appendix Table 78: Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.722	4.922	18.48	4.755
6	1.778	30	7	23.333	23	4.26	4.443	4.27	16.74	4.458
3	1.477	30	5	16.667	17	4.05	4.164	4.056	14.13	4.162
1.50	1.176	30	4	13.333	13	3.87	3.885	3.873	11.10	3.865
0.75	0.875	30	3	10	10	3.72	3.606	3.73	9.060	3.568

Results:

Y = 2.705716 + 0.9856053 X

Chi-squared is 1.504621 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.327792
LD_{50} is 21.271215 mg/gm
95% confidence limits are 6.821656 to 66.32765 mg/gm

Appendix Table 79: Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.778	5.00	18.48	4.798
6	1.778	30	8	26.667	27	4.39	4.536	4.376	17.43	4.543
3	1.477	30	6	20.000	20	4.16	4.294	4.150	15.09	4.287
1.50	1.176	30	4	13.333	13	3.87	4.052	3.873	13.17	4.032
0.75	0.875	30	3	16.667	17	4.05	3.810	4.077	11.10	3.777

Results:

Y = 3.034778 + 0.8479446 X

Chi-squared is 2.856905 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.317631
LD_{50} is 20.779286 mg/gm
95% confidence limits are 5.764886 to 74.898 mg/gm

Appendix Table 80:**Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure**.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.082	5.25	19.11	5.094
6	1.778	30	11	36.667	37	4.67	4.823	4.682	18.81	4.826
3	1.477	30	9	30.000	30	4.48	4.564	4.46	17.43	4.558
1.50	1.176	30	7	23.333	23	4.26	4.305	4.266	15.96	4.290
0.75	0.875	30	6	20.000	20	4.16	4.046	4.16	13.17	4.022

Results:

Y = 3.24287 + 0.8903251 X

Chi-squared is 1.282427 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.973582
LD_{50} is 9.409836 mg/gm
95% confidence limits are 4.200247 to 21.08091 mg/gm

Appendix Table 81: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.954	5.065	19.02	4.981
6	1.778	30	8	26.667	27	4.39	4.475	4.39	16.74	4.478
3	1.477	30	4	13.333	13	3.87	3.996	3.878	12.15	3.974
1.50	1.176	30	2	6.667	7	3.52	3.517	3.519	8.07	3.471
0.75	0.875	30	1	3.333	3	3.12	3.038	3.135	3.93	2.967

Results:

Y = 1.503229 + 1.67287 X
Chi-squared is 0.5040646 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.090283
LD_{50} is 12.31076 mg/gm
95% confidence limits are 7.287057 to 20.79762 mg/gm

Appendix Table 82: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	19	63.333	63	5.33	5.132	5.315	19.02	5.160
6	1.778	30	11	36.667	37	4.67	4.730	4.662	18.48	4.736
3	1.477	30	5	16.667	17	4.05	4.328	4.074	15.96	4.311
1.50	1.176	30	4	13.333	13	3.87	3.926	3.878	12.15	3.887
0.75	0.875	30	3	10.000	10	3.72	3.524	3.75	8.07	3.463

Results:

Y = 2.229132 + 1.409709 X
Chi-squared is 2.123493 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.96556
LD_{50} is 9.237628mg/gm
95% confidence limits are 5.537157 to 15.41111 mg/gm

Appendix Table 83: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larvae (1st instar) after 72h of exposure.

Dose	Log dose	# used	# Kill	%kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	28	90.000	90	6.28	6.196	6.270	12.15	6.173
6	1.778	30	22	73.333	73	5.61	5.667	5.610	16.74	5.646
3	1.477	30	16	53.333	53	5.08	5.138	5.065	19.02	5.120
1.5	1.176	30	10	33.333	33	4.56	4.609	4.551	18.03	4.592
0.75	.875	30	6	20.000	20	4.16	4.080	4.160	13.17	4.066

Results:

Y = 2.534 + 1.751 X
Chi-squared is 0.341659 with 3 degrees of freedom
No significant heterogeneity
LogLD_{50} is 2.356985
LD_{50} is 3.456354 mg/g
95% confidence limits are 1.912658 to 3.434968 mg/g

Appendix Table 84: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.686	4.929	18.03	4.736
6	1.778	30	8	20.000	20	4.16	4.362	4.17	15.96	4.383
3	1.477	30	4	13.333	13	3.87	4.038	3.873	13.17	4.028
1.50	1.176	30	3	10.000	10	3.72	3.714	3.72	10.08	3.676
0.75	0.875	30	2	6.667	7	3.52	3.390	3.572	6.24	3.323

Results:

Y = 2.296055 + 1.173704 X

Chi-squared is 2.122684 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.303772
LD_{50} is 20.126657 mg/gm
95% confidence limits are 7.647386 to 52.97 mg/gm

Appendix Table 85:Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.882	5.098	18.81	4.906
6	1.778	30	8	26.667	27	4.39	4.617	4.389	18.03	4.628
3	1.477	30	7	23.333	23	4.26	4.352	4.266	15.96	4.350
1.50	1.176	30	6	20	20	4.16	4.087	4.16	13.17	4.072
0.75	0.875	30	4	13.333	13	3.87	3.822	3.873	11.10	3.794

Results:

Y = 2.98578 + 0.9235459 X
Chi-squared is 2.007341 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.180963
LD_{50} is 15.169220 mg/gm
95% confidence limits are 5.534841 to 41.57396 mg/gm

Appendix Table 86: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	19	63.333	63	5.33	5.090	5.325	19.11	5.103
6	1.778	30	10	33.333	33	4.56	4.804	4.578	18.81	4.806
3	1.477	30	8	26.667	27	4.39	4.518	4.376	17.43	4.509
1.50	1.176	30	7	23.333	23	4.26	4.232	4.252	15.09	4.212
0.75	0.875	30	5	16.667	17	4.05	3.946	4.062	12.15	3.915

Results:

Y = 3.051451 + 0.9869455 X

Chi-squared is 2.514602 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.974323
LD_{50} is 9.425903 mg/gm
95% confidence limits are 4.545616 to 19.5458 mg/gm

Appendix Table 87: Larvicidal effect of stem bark extract (Chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.766	4.922	14.48	4.774
6	1.778	30	8	26.667	27	4.39	4.522	4.376	17.43	4.522
3	1.477	30	6	20.000	20	4.16	4.278	4.15	15.09	4.269
1.50	1.176	30	5	16.667	17	4.05	4.034	4.037	13.17	4.016
0.75	0.875	30	4	13.333	13	3.87	3.790	3.894	10.08	3.763

Results:

Y = 3.028001 + 0.8399352 X

Chi-squared is 1.16333 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.347799
LD_{50} is 22.27406 mg/gm
95% confidence limits are 5.814798 to 85.32262 mg/gm

Appendix Table 88: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.252	5.462	18.81	5.265
6	1.778	30	13	43.333	43	4.82	4.973	4.815	19.02	4.979
3	1.477	30	10	33.333	33	4.56	4.694	4.551	18.03	4.694
1.50	1.176	30	8	26.667	27	4.39	4.415	4.39	16.74	4.408
0.75	0.875	30	7	23.333	23	4.26	4.136	4.284	14.13	4.122

Results:

Y = 3.291224 + 0.9493623 X

Chi-squared is 1.985899 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.79992
LD_{50} is 6.308404mg/gm
95% confidence limits are 3.404555 to 11.68903 mg/gm

Appendix Table 89: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.366	5.50	18.48	5.352
6	1.778	30	15	50.000	50	5.00	5.114	4.990	19.02	5.103
3	1.477	30	12	40.000	40	4.75	4.862	4.760	18.81	4.854
1.50	1.176	30	10	33.333	33	4.56	4.610	4.551	18.03	4.605
0.75	0.875	30	9	30.000	30	4.48	4.358	4.49	15.96	4.356

Results:

Y = 3.631464 + 0.8275156 X

Chi-squared is 1.152888 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.6537892
LD_{50} is 4.505973 mg/gm
95% confidence limits are 2.441486 to 8.316161 mg/gm

Appendix Table 90: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.918	5.065	19.02	4.920
6	1.778	30	9	30.000	30	4.48	4.633	4.470	18.03	4.629
3	1.477	30	7	23.333	23	4.26	4.348	4.266	16.96	4.338
1.50	1.176	30	5	16.667	17	4.05	4.062	4.037	13.17	4.047
0.75	0.875	30	4	13.333	13	3.87	3.778	3.894	10.08	3.756

Results:

Y = 2.910171 + 0.9666475 X

Chi-squared is 1.131802 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.161935
LD_{50} is 514.51894.mg/gm
95% confidence limits are 5.650212 to 37.30826 mg/gm

Appendix Table 91: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.932	4.99	19.02	4.925
6	1.778	30	11	36.667	37	4.67	4.723	4.662	18.48	4.715
3	1.477	30	9	30.000	30	4.48	4.514	4.46	17.43	4.505
1.50	1.176	30	7	23.333	23	4.26	4.305	4.266	15.96	4.295
0.75	0.875	30	6	20.000	20	4.16	4.096	4.16	13.17	4.085

Results:

Y = 3.475124 + 0.6972134 X

Chi-squared is 0.2550674 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.187101

LD_{50} is 15.38510 mg/gm

95% confidence limits are 4.101972 to 57.70426 mg/gm

Appendix Table 92: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	5.220	5.202	18.81	5.226
6	1.778	30	15	50	50	5.00	4.992	4.990	19.02	4.996
3	1.477	30	13	43.333	43	4.82	4.764	4.818	18.48	4.765
1.50	1.176	30	10	33.333	33	4.56	4.536	4.544	17.43	4.534
0.75	0.875	30	7	23.333	23	4.26	4.308	4.266	15.96	4.303

Results:

Y = 3.632576 + 0.7665 X

Chi-squared is 8.776665E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.7839846.081132
LD_{50} is 14.3854 mg/gm
95% confidence limits are 2.875615 to 12.85992 mg/gm

Appendix Table 93 : Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.968	4.065	19.02	4.962
6	1.778	30	9	30.000	30	4.48	4.595	4.46	17.43	4.585
3	1.477	30	6	20.000	20	4.16	4.222	4.15	15.09	4.208
1.50	1.176	30	4	13.333	13	3.87	3.849	3.873	11.10	3.830
0.75	0.875	30	2	6.667	7	3.52	3.476	3.540	7.140	3.453

Results:

Y = 2.355597 + 1.2538 X

Chi-squared is 0.5973358 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.109111
LD_{50} is 12.856148 mg/gm.

95% confidence limits are 0.603954 to 25.80911 mg/gm

Appendix Table 94: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.086	5.25	19.11	5.094
6	1.778	30	11	36.667	37	4.67	4.795	4.662	18.48	4.793
3	1.477	30	8	26.667	27	4.39	4.504	4.376	17.43	4.492
1.50	1.176	30	6	20.000	20	4.16	4.213	4.15	15.09	4.191
0.75	0.875	30	5	16.667	17	4.05	3.922	4.062	12.15	3.890

Results:

Y = 3.015847 + 0.9994459 X

Chi-squared is 1.401576 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.985253
LD_{50} is 9.666136 mg/gm
95% confidence limits are 4.651048 to 20.08884 mg/gm

Appendix Table 95:Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.274	5.54	18.81	5.295
6	1.778	30	13	43.333	43	4.82	5.014	4.825	19.11	5.023
3	1.477	30	10	33.333	33	4.56	4.754	4.558	18.48	4.751
1.50	1.176	30	9	30.000	30	4.48	4.494	4.48	16.74	4.479
0.75	0.875	30	8	26.667	27	4.39	4.234	4.388	15.09	4.208

Results:

Y = 3.417769 + 0.9026579 X

Chi-squared is 3.060968 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.752858
LD_{50} is 5.660547 mg/gm
95% confidence limits are 3.05997 to 10.47127 mg/gm

Appendix Table 96: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larvae (1st instar) after 24h of exposure.

Dose	Log dose	# used	# Kill	%kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	25	85.667	86	6.13	6.006	6.087	13.17	5.957
6	1.778	30	20	65.667	66	5.44	5.516	5.416	17.43	5.477
3	1.477	30	13	46.667	47	4.92	5.026	4.925	19.11	4.996
1.5	1.176	30	8	30.000	30	4.48	4.536	4.46	17.43	4.515
0.75	0.875	30	5	20.000	20	4.16	4.046	4.16	13.17	4.034

Results:

Y = 2.636 + 1.597 X
Chi-squared is 0.642698 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.479779
LD_{50} is 14.018987 mg/g
95% confidence limits are 3.202658 to 5.137465 mg/g

Appendix Table 97: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larvae **(1st instar) after 48h of exposure.**

Dose	Log dose	# used	# Kill	%kill	Corr %	Emp probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	27	86.987	87	6.13	6.07	6.087	13.17	6.026
6	1.778	30	22	70.000	70	5.52	5.58	5.500	17.43	5.549
3	1.477	30	16	53.333	53	5.08	5.09	5.075	19.11	5.071
1.5	1.176	30	11	33.333	33	4.56	4.60	4.551	18.03	4.594
0.75	0.875	30	6	20.000	20	4.16	4.11	4.170	14.13	4.116

Results:

Y = 3.728 + 1. 658 X
Chi-squared is 0.164369 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.432117
LD_{50} is 10.705654 mg/g
95% confidence limits are 2.973654 to 6.709896 mg/g

Appendix Table 98: Larvicidal effect of stem wood extract (methanol**) of *A. indica* against *T. castaneum* larva (1st instar) after 72h of exposure.**

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.234	5.54	18.81	5.281
6	1.778	30	12	40.000	40	4.75	4.87	4.76	18.81	4.892
3	1.477	30	6	20.000	20	4.16	4.506	4.18	17.43	4.503
1.50	1.176	30	5	16.667	17	4.05	4.142	4.056	14.13	4.114
0.75	0.875	30	5	16.667	17	4.05	3.778	4.126	10.08	3.725

Results:
Y = 3.69894 + 2.2927572 X
Chi-squared is 4.096075 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 1.861892
LD50 is 7.37698mg/gm
95% confidence limits are 5.560706 to 13.96809 mg/gm

Appendix Table 99: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.874	5.098	18.81	4.936
6	1.778	30	8	26.667	27	4.39	4.494	4.39	16.74	4.522
3	1.477	30	4	13.333	13	3.87	4.116	3.904	14.13	4.109
1.50	1.176	30	3	10.000	10	3.72	3.737	3.72	10.08	3.695
0.75	0.875	30	2	6.667	7	3.52	3.358	3.572	6.24	3.281

Results:

Y = 2.079067 + 1.374097 X

Chi-squared is 1.912054 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.040941

LD_{50} is 13.35709mg/gm

95% confidence limits are 6.91369 to 25.80559 mg/gm

Appendix Table 100: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.010	5.25	19.11	5.046
6	1.778	30	10	33.333	33	4.56	4.665	4.551	18.03	4.681
3	1.477	30	5	16.667	17	4.05	4.320	4.074	15.96	4.315
1.50	1.176	30	4	13.333	13	3.87	3.975	3.878	12.15	3.949
0.75	0.875	30	4	13.333	13	3.87	3.630	3.931	9.060	3.583

Results:

Y = 2.519619 + 1.215313 X

Chi-squared is 3.177353 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 2.125712

LD50 is 10.98855mg/gm

95% confidence limits are 5.726405 to 21.08623 mg/gm

Appendix Table 101: Larvicidal effect of flower extract (chloroform**) of *A. indica* against *T. castaneum* larva (2nd instar after 72h of exposure.**

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.234	5.54	18.81	5.281
6	1.778	30	12	40.000	40	4.75	4.87	4.76	18.81	4.892
3	1.477	30	6	20.000	20	4.16	4.506	4.18	17.43	4.503
1.50	1.176	30	5	16.667	17	4.05	4.142	4.056	14.13	4.114
0.75	0.875	30	5	16.667	17	4.05	3.778	4.126	10.08	3.725

Results:
Y = 2.59393 + 1.292272 X
Chi-squared is 5.076075 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 1.861892
LD50 is 6.275992mg/gm
95% confidence limits are 4.460706 to 11.86809 mg/gm

Appendix Table 102: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.776	4.922	18.48	4.823
6	1.778	30	7	23.333	23	4.26	4.342	4.266	15.96	4.358
3	1.477	30	3	10.000	10	3.72	3.908	3.74	12.15	3.892
1.50	1.176	30	2	6.667	7	3.52	3.474	3.54	7.140	3.427
0.75	0.875	30	1	3.333	3	3.12	3.040	3.135	3.93	2.961

Results:

Y = 1.607492 + 1.546745 X

Chi-squared is 0.8064823 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.193321

LD_{50} is 15.60706 mg/gm

95% confidence limits are 8.070065 to 30.18321 mg/gm

Appendix Table 103: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.928	5.065	19.02	4.928
6	1.778	30	9	30	30	4.48	4.613	4.470	18.03	4.608
3	1.477	30	6	20	20	4.16	4.298	4.150	15.09	4.287
1.50	1.176	30	5	16.667	17	4.05	3.983	4.062	12.15	3.966
0.75	0.875	30	3	10	10	3.72	3.668	3.730	9.060	3.645

Results:

Y = 2.71183 + 1.066102 X

Chi-squared is 1.155865 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.146296
LD_{50} is 14.00541 mg/gm
95% confidence limits are 86.014042 to 32.61558. mg/gm

Appendix Table 104: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (2ndinstar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.196	5.24	19.02	5.212
6	1.778	30	14	46.667	47	4.92	4.844	4.942	18.81	4.854
3	1.477	30	7	23.333	23	4.26	4.492	4.27	16.74	4.496
1.50	1.176	30	6	20.000	20	4.16	4.14	4.17	14.13	4.138
0.75	0.875	30	4	13.333	13	3.87	3.788	3.894	10.08	3.781

Results:

Y = 2.740535 + 1.188533 X

Chi-squared is 1.161095 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.901053
LD_{50} is 7.962568 mg/gm
95% confidence limits are 4.562784 to 13.89557 mg/gm

Appendix Table 105: Larvicidal effect of leaf extract (Chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.814	4.942	18.81	4.851
6	1.778	30	8	26.667	27	4.39	4.467	4.39	16.74	4.485
3	1.477	30	5	16.667	17	4.05	4.12	4.056	14.13	4.119
1.50	1.176	30	3	10.000	10	3.72	3.773	3.72	10.08	3.753
0.75	0.875	30	2	6.667	7	3.52	3.426	3.54	7.140	3.387

Results:

Y = 2.323623 + 1.215409 X

Chi-squared is 0.5411558 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.202038
LD_{50} is 15.92349 mg/gm
95% confidence limits are 7.07521 to 35.83746 mg/gm

Appendix Table 106: Larvicidal effect of leaf extract (Chloroform) of *A. indica* against *T. castaneum* larva (2ndinstar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	5.004	5.075	19.11	5.016
6	1.778	30	11	36.667	37	4.67	4.652	4.659	18.03	4.653
3	1.477	30	6	20.000	20	4.16	4.300	4.150	15.09	4.291
1.50	1.176	30	4	13.333	13	3.87	3.948	3.878	12.15	3.928
0.75	0.875	30	3	10.000	10	3.72	3.596	3.750	8.07	3.565

Results:

Y = 2.511452 + 1.204449 X

Chi-squared is 0.6711731 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.066131

LD_{50} is 11.64476 mg/gm

95% confidence limits are 5.891689 to 23.01556 mg/gm

Appendix Table 107: Larvicidal effect of leaf extract (Chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	19	63.333	63	5.33	5.316	5.318	18.48	5.311
6	1.778	30	16	53.333	53	5.08	4.968	5.065	19.02	4.959
3	1.477	30	9	30.000	30	4.48	4.620	4.470	18.03	4.607
1.50	1.176	30	6	20.000	20	4.16	4.272	4.15	15.09	4.225
0.75	0.875	30	5	16.667	17	4.05	3.924	4.062	12.15	3.903

Results:

Y = 2.880345 + 1.169131 X

Chi-squared is 1.026411 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.813017

LD_{50} is 6.501557 mg/gm

95% confidence limits are 3.898881 to 10.84164 mg/gm

Appendix Table 108: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.778	4.922	14.48	4.814
6	1.778	30	8	26.667	27	4.39	4.431	4.39	16.74	4.445
3	1.477	30	4	13.333	13	3.87	4.084	3.873	13.17	4.076
1.50	1.176	30	3	10.000	10	3.72	3.737	3.72	10.08	3.707
0.75	0.875	30	2	6.667	7	3.52	3.390	3.572	6.24	3.337

Results:

Y = 2.264115 + 1.226494 X

Chi-squared is 1.152259 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 2.230655

LD50 is 17.00807mg/gm

95% confidence limits are 7.326695 to 39.48226 mg/gm

Appendix Table 109: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.974	5.065	19.02	4.979
6	1.778	30	11	36.667	37	4.67	4.67	4.659	18.03	4.668
3	1.477	30	6	20.000	20	4.16	4.366	4.17	15.96	4.357
1.50	1.176	30	5	16.667	17	4.05	4.062	4.037	13.17	4.045
0.75	0.875	30	4	13.333	13	3.87	3.758	3.894	10.08	3.734

Results:
Y = 2.829474 + 1.033845 X
Chi-squared is 0.9562092 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 2.09947
LD50 is 12.57389 mg/gm
95% confidence limits are 5.534317 to 28.56771 mg/gm

Appendix Table 110: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.332	5.422	18.48	5.331
6	1.778	30	15	50.000	50	5.00	4.97	4.99	19.02	4.961
3	1.477	30	8	26.667	27	4.39	4.608	4.389	18.03	4.591
1.50	1.176	30	6	20.000	20	4.16	4.246	4.15	15.09	4.222
0.75	0.875	30	5	16.667	17	4.05	3.884	4.077	11.10	3.852

Results:

Y = 2.777095 + 1.228279 X

Chi-squared is 1.547712 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 1.809772

LD50 is 6.453151 mg/gm

95% confidence limits are 3.973338 to 10.48065 mg/gm

Appendix Table 111: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.364	4.394	15.96	4.362
6	1.778	30	6	20.000	20	4.16	4.201	4.15	15.09	4.198
3	1.477	30	5	16.667	17	4.05	4.038	4.037	13.17	4.034
1.50	1.176	30	4	13.333	13	3.87	3.875	3.873	11.10	3.870
0.75	0.875	30	3	10.000	10	3.72	3.712	3.72	10.08	3.706

Results:
Y = 3.653581 + 0.5443193 X
Chi-squared is 5.344272E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 2.473583
LD50 is 297.56647 mg/gm
95% confidence limits are 4.071503 to 21747.63 mg/gm

Appendix Table112: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4.48	4.45	4.48	16.74	4.454
6	1.778	30	7	23.333	23	4.26	4.307	4.266	15.96	4.309
3	1.477	30	6	20.000	20	4.16	4.164	4.170	14.13	4.165
1.50	1.176	30	5	16.667	17	4.05	4.021	4.037	13.17	4.020
0.75	0.875	30	4	13.333	13	3.87	3.878	3.873	11.10	3.876

Results:
Y = 3.828938 + 0.4806597 X
Chi-squared is 4.551387E-02 with 3 degrees of freedom
No significant heterogeneity

Log LD50 is 2.436365

LD50 is 273.12738 mg/gm

95% confidence limits are 2.754858 to 27078.91 mg/gm

Appendix Table 113: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.648	4.74	18.03	4.649
6	1.778	30	8	26.667	27	4.39	4.485	4.39	16.74	4.486
3	1.477	30	7	23.333	23	4.26	4.322	4.266	15.96	4.324
1.50	1.176	30	6	20.000	20	4.16	4.159	4.170	14.13	4.161
0.75	0.875	30	5	16.667	17	4.05	3.996	4.062	12.15	3.999

Results:

Y = 3.946196 + 0.5402143X

Chi-squared is 0.408191 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 1.950716

LD50 is 89.27206 mg/gm

95% confidence limits are 5.00674 to 1591.755 mg/gm

Appendix Table 114: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	6	20	20	4.16	4.186	4.17	14.13	4.185
6	1.778	30	5	16.667	17	4.05	4.025	4.037	13.17	4.025
3	1.477	30	4	13.333	13	3.87	3.864	3.873	11.10	3.865
1.50	1.176	30	3	10	10	3.72	3.703	3.72	10.08	3.704
0.75	0.875	30	2	6.667	7	3.52	3.542	3.519	8.07	3.544

Results:

Y = 3.492854 + 0.5317924 X

Chi-squared is 1.356268E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 2.834087

LD50 is 682.47485 mg/gm

95% confidence limits are 2.314314 to 201256.9 mg/gm

Appendix Table 115: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30	30	4.48	4.486	4.48	16.74	4.488
6	1.778	30	8	26.667	27	4.39	4.377	4.394	15.96	4.377
3	1.477	30	7	23.333	23	4.26	4.268	4.252	15.09	4.267
1.50	1.176	30	6	20	20	4.16	4.159	4.17	14.13	4.156
0.75	0.875	30	5	16.667	17	4.05	4.05	4.037	13.17	4.045

Results:
Y = 4.009365 + 0.3680751 X
Chi-squared is 1.235986E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 2.691395
LD50 is 491.35428 mg/gm
95% confidence limits are 6125063 to 394165 mg/gm

Appendix Table 116: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.426	4.39	16.74	4.417
6	1.778	30	7	23.333	23	4.26	4.271	4.252	15.09	4.268
3	1.477	30	6	20.000	20	4.16	4.116	4.17	14.13	4.119
1.50	1.176	30	5	16.667	17	4.05	3.961	4.062	12.15	3.971
0.75	0.875	30	3	10.000	10	3.72	3.806	3.72	11.10	3.822

Results:

Y = 3.773859 + 0.494341 X

Chi-squared is 0.2688 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 2.480356

LD50 is 302.24278 mg/gm

95% confidence limits are 3.008951 to 30359.66 mg/gm

Appendix Table 117:Larvicidal effect of rood wood extract (Chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.246	4.252	15.09	4.238
6	1.778	30	5	16.667	17	4.05	4.065	4.037	13.17	4.059
3	1.477	30	4	13.333	13	3.87	3.884	3.873	11.10	3.880
1.50	1.176	30	3	10.000	10	3.72	3.703	3.72	10.08	3.701
0.75	0.875	30	2	6.667	7	3.52	3.522	3.519	8.07	3.522

Results:
Y = 3.464888 + 0.5942255 X
Chi-squared is 1.362634E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 2.583383
LD50 is 383.16239mg/gm
95% confidence limits are 4.624563 to 31746.43 mg/gm

Appendix Table 118:Larvicidaleffect of rood wood extract (Chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.404	4.39	16.74	4.399
6	1.778	30	7	23.333	23	4.26	4.231	4.252	15.09	4.227
3	1.477	30	5	16.667	17	4.05	4.058	4.037	13.17	4.055
1.50	1.176	30	4	13.333	13	3.87	3.885	3.873	11.10	3.882
0.75	0.875	30	3	10	10	3.72	3.712	3.72	10.08	3.710

Results:
Y = 3.654891 + 0.5717278 X
Chi-squared is 1.699162E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD50 is 2.352708
LD50 is 225.27249mg/gm
95% confidence limits are 5.222331 to 9717.434 mg/gm

Appendix Table 119: Larvicidal effect of rood wood extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.742	4.74	18.48	4.734
6	1.778	30	10	33.333	33	4.56	4.562	4.544	17.43	4.553
3	1.477	30	8	26.667	27	4.39	4.382	4.394	15.96	4.373
1.50	1.176	30	6	20.000	20	4.16	4.202	4.15	15.09	4.192
0.75	0.875	30	5	16.667	17	4.05	4.022	4.037	13.17	4.011

Results:

Y = 3.9528 + 0.6005473 X
Chi-squared is 4.457855E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.743743
LD_{50} is 55.42979 mg/gm
95% confidence limits are 6.426384 to 478.1001 mg/gm

Appendix Table 120: Larvicidal effect of rood wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	6	20	20	4.16	4.186	4.17	14.13	4.185
6	1.778	30	5	16.667	17	4.05	4.025	4.037	13.17	4.025
3	1.477	30	4	13.333	13	3.87	3.864	3.873	11.10	3.865
1.50	1.176	30	3	10	10	3.72	3.703	3.72	10.08	3.704
0.75	0.875	30	2	6.667	7	3.52	3.542	3.519	8.07	3.544

Results:

Y = 3.492854 + 0.5317924 X
Chi-squared is 1.356268E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.834087
LD_{50} is 682.47486 mg/gm
95% confidence limits are 2.314314 to 201256.9 mg/gm

Appendix Table 121: Larvicidal effect of rood wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4.48	4.45	4.48	16.74	4.454
6	1.778	30	7	23.333	23	4.26	4.307	4.266	15.96	4.310
3	1.477	30	6	20.000	20	4.16	4.164	4.17	14.13	4.165
1.50	1.176	30	5	16.667	17	4.05	4.021	4.037	13.17	4.020
0.75	0.875	30	4	13.333	13	3.87	3.878	3.873	11.10	3.876

Results:

Y = 3.828938 + 0.4806597 X

Chi-squared is 4.551387E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.436365

LD_{50} is 273.12739mg/gm

95% confidence limits are 2.754858 to 27078.91 mg/gm

Appendix Table 122: Larvicidal effect of rood wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	11	36.667	37	4.67	4.662	4.659	18.03	4.650
6	1.778	30	9	30	30	4.48	4.506	4.46	17.43	4.497
3	1.477	30	8	26.667	27	4.39	4.35	4.394	15.96	4.344
1.50	1.176	30	6	20	20	4.16	4.194	4.17	14.13	4.191
0.75	0.875	30	5	16.667	17	4.05	4.038	4.037	13.17	4.038

Results:

Y = 3.988786 + 0.5082985 X

Chi-squared is 7.139802E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD50 is 1.98941

LD50 is 97.59105 mg/gm

95% confidence limits are 4.343658 to 2192.625 mg/gm

Appendix Table 123: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10.000	10	3.72	3.656	3.730	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.214	3.121	5.400	3.257

Results:

Y = 3.190897 + 0.677905 X

Chi-squared is 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 466.30250 mg/gm

95% confidence limits are 4.949317 to 43932.83 mg/gm

Appendix Table124: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.444	4.39	16.74	4.427
6	1.778	30	7	23.333	23	4.26	4.231	4.252	15.09	4.223
3	1.477	30	5	16.667	17	4.05	4.018	4.037	13.17	4.018
1.50	1.176	30	4	13.333	13	3.87	3.805	3.873	11.10	3.814
0.75	0.875	30	2	6.667	7	3.52	3.592	3.519	8.07	3.609

Results:

Y = 3.54343 + 0.6792087 X

Chi-squared is 0.1454554 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.144511

LD_{50} is 139.47975mg/gm

95% confidence limits are 8.041382 to 2419.306 mg/gm

Appendix Table 125: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.600	4.544	17.43	4.585
6	1.778	30	8	26.667	27	4.39	4.398	4.394	15.96	4.394
3	1.477	30	7	23.333	23	4.26	4.196	4.284	14.13	4.203
1.50	1.176	30	5	16.667	17	4.05	3.994	4.062	12.15	4.012
0.75	0.875	30	3	10.000	10	3.72	3.792	3.72	10.08	3.821

Results:

Y = 3.759897 + 0.6343551 X

Chi-squared is 0.2554999 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.954904

LD_{50} is 90.13719 mg/gm

95% confidence limits are 7.008613 to 1159.247mg/gm

Appendix Table 126: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.213	3.121	5.40	3.256

Results:

Y = 3.190897 + 0.677905 X

Chi-squared is 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 463. 53029mg/gm

95% confidence limits are 4.949317 to 43932.83mg/gm

Appendix Table 127: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.296	4.252	15.09	4.298
6	1.778	30	7	23.333	23	4.26	4.179	4.284	14.13	4.181
3	1.477	30	5	16.667	17	4.05	4.062	4.037	13.17	4.065
1.50	1.176	30	4	13.333	13	3.87	3.945	3.878	12.15	3.948
0.75	0.875	30	4	13.333	13	3.87	3.828	3.873	11.10	3.832

Results:

Y = 3.794079 + 0.3872388 X

Chi-squared is 0.2699411 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.114154

LD_{50} is460.63568 mg/gm

95% confidence limits are .361576 to 4678510 mg/gm

Appendix Table 128: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.574	4.544	17.43	4.562
6	1.778	30	9	30.000	30	4.48	4.472	4.48	16.74	4.465
3	1.477	30	8	26.667	27	4.39	4.37	4.394	15.96	4.368
1.50	1.176	30	7	23.333	23	4.26	4.268	5.252	15.09	4.271
0.75	0.875	30	6	20.000	20	4.16	4.166	4.17	14.13	4.174

Results:

Y = 4.142542 + 0.3227044 X

Chi-squared is 2.583397E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.657103

LD_{50} is 454.04937 mg/gm

95% confidence limits are 0.3036665 to 678905.1mg/gm

Appendix Table129: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	6	20.000	20	4.16	4.186	4.17	14.13	4.185
6	1.778	30	5	16.667	17	4.05	4.025	4.037	13.17	4.025
3	1.477	30	4	13.333	13	3.87	3.864	3.873	11.10	3.865
1.50	1.176	30	3	10.000	10	3.72	3.703	3.72	10.08	3.704
0.75	0.875	30	2	6.667	7	3.52	3.542	3.519	8.07	3.544

Results:

Y = 3.492854 + 0.5317924 X
Chi-squared is 1.356268E-02 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.834087
LD_{50} is 682.47487 mg/gm
95% confidence limits are 2.314314 to 201256.9 mg/gm

Appendix Table 130: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.426	4.39	16.74	4.417
6	1.778	30	7	23.333	23	4.26	4.271	4.252	15.09	4.268
3	1.477	30	6	20	20	4.16	4.116	4.17	14.13	4.119
1.50	1.176	30	5	16.667	17	4.05	3.961	4.062	12.15	3.971
0.75	0.875	30	3	10	10	3.72	3.806	3.72	11.10	3.822

Results:

Y = 3.773859 + 0.494341 X

Chi-squared is 0.2688 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 0.2688

LD_{50} is 302.24279 mg/gm

95% confidence limits are 3.008951 to 30359.66 mg/gm

Appendix Table 131: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.738	4.74	18.48	4.725
6	1.778	30	10	33.333	33	4.56	4.59	4.544	17.43	4.581
3	1.477	30	9	30.000	30	4.48	4.442	4.48	16.74	4.437
1.50	1.176	30	7	23.333	23	4.26	4.294	4.252	15.09	4.294
0.75	0.875	30	6	20.000	20	4.16	4.146	4.17	14.13	4.150

Results:

Y = 4.104065 + 0.476937 X

Chi-squared 9.060383E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.878519

LD_{50} is 75.59956 mg/gm

95% confidence limits are 3.743789 to 1526.607mg/gm

Appendix Table 132: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4. 48	4.464	4.48	16.74	4.464
6	1.778	30	8	26.667	27	4.39	4.377	4.394	15.96	4.377
3	1.477	30	7	23.333	23	4.26	4.29	4.252	15.09	4.289
1.50	1.176	30	6	20.000	20	4.16	4.203	4.15	15.09	4.201
0.75	0.875	30	6	20.000	20	4.16	4.116	4.17	14.13	4.114

Results:

Y = 4.085724 + 0.2908732 X

Chi-squared is 0.1144633 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.143212

LD_{50} is 1390.6339mg/gm

95% confidence limits are 5.139258E-02 to 3.762912E+07mg/gm

Appendix Table 133: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar)) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.348	4.266	15.96	4.337
6	1.778	30	7	23.333	23	4.26	4.219	4.252	15.09	4.215
3	1.477	30	6	20	20	4.16	4.09	4.16	13.17	4.093
1.50	1.176	30	5	16.667	17	4.05	3.961	4.062	12.15	3.970
0.75	0.875	30	3	10	10	3.72	3.832	3.72	11.10	3.848

Results:

Y = 3.808434 + 0.4066209 X

Chi-squared is 0.4456774 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.930411

LD_{50} is 851.94329mg/gm

95% confidence limits are 0.7165104 to 1012975 mg/gm

Appendix Table 134: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.213	3.121	5.400	3.257

Results:

Y = 3.190897 + 0.677905 X

Chi-squared is 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 466.30258 mg/gm

95% confidence limits are 4.949317 to 43932.83 mg/gm

Appendix Table 135: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	6	20.000	20	4.16	4.186	4.17	14.13	4.185
6	1.778	30	5	16.667	17	4.05	4.025	4.037	13.17	4.025
3	1.477	30	4	13.333	13	3.87	3.864	3.873	11.10	3.865
1.50	1.176	30	3	10.000	10	3.72	3.703	3.72	10.08	3.704
0.75	0.875	30	2	6.667	7	3.52	3.542	3.519	8.07	3.544

Results:

Y = 3.492854 + 0.5317924 X

Chi-squared 1.356268E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.834087

LD_{50} is 682.47486 mg/gm

95% confidence limits are 2.314314 to 201256.9 mg/gm

Appendix Table 136: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.426	4.39	16.74	4.417
6	1.778	30	7	23.333	23	4.26	4.271	4.252	15.09	4.268
3	1.477	30	6	20.000	20	4.16	4.116	4.17	14.13	4.119
1.50	1.176	30	5	16.667	17	4.05	3.961	4.062	12.15	3.971
0.75	0.875	30	3	10.000	10	3.72	3.806	3.72	11.10	3.822

Results:

Y = 3.773859 + 0.494341 X

Chi-squared 0.2688 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.480356

LD_{50} is 302.24279 mg/gm

95% confidence limits are 3.008951 to 30359.66 mg/gm

Appendix Table 137: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.534	4.544	17.43	4.527
6	1.778	30	8	26.667	27	4.39	4.409	4.39	16.74	4.403
3	1.477	30	7	23.333	23	4.26	4.284	4.252	15.09	4.278
1.50	1.176	30	6	20	20	4.16	4.159	4.17	14.13	4.154
0.75	0.875	30	5	16.667	17	4.05	4.034	4.037	13.17	4.030

Results:

Y = 3.990417 + 0.4121533 X

Chi-squared 2.252841E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.449534

LD_{50} is 281.53658 mg/gm

95% confidence limits are 1.561521 to 50759.78 mg/gm

Appendix Table 138: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.018	4.037	13.17	4.016
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.876
3	1.477	30	3	10.000	10	3.72	3.736	3.72	10.08	3.737
1.50	1.176	30	2	6.667	7	3.52	3.595	3.519	8.07	3.598
0.75	0.875	30	2	6.667	7	3.52	3.454	3.54	7.140	3.459

Results:

Y = 3.414252 + 0.4624146 X

Chi-squared 0.1064537 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.429279

LD_{50} is 2687.06845mg/gm

95% confidence limits are 0.3856401 to 1.872297E+07mg/gm

Appendix Table 139: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4.48	4.486	4.48	16.74	4.488
6	1.778	30	8	26.667	27	4.39	4.377	4.394	15.96	4.377
3	1.477	30	7	23.333	23	4.26	4.268	4.252	15.09	4.267
1.50	1.176	30	6	20.000	20	4.16	4.159	4.17	14.13	4.156
0.75	0.875	30	5	16.667	17	4.05	4.05	4.037	13.17	4.045

Results:

Y = 4.009365 + 0.3680751 X

Chi-squared 1.235986E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.691395

LD_{50} is 491.35426mg/gm

95% confidence limits are 6125063 to 394165mg/gm

Appendix Table 140: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (2nd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.426	4.39	16.74	4.417
6	1.778	30	7	23.333	23	4.26	4.271	4.252	15.09	4.268
3	1.477	30	6	20.000	20	4.16	4.116	4.17	14.13	4.119
1.50	1.176	30	5	16.667	17	4.05	3.961	4.062	12.15	3.971
0.75	0.875	30	3	10.000	10	3.72	3.806	3.72	11.10	3.822

Results:

Y = 3.773859 + 0.494341 X

Chi-squared 0.2688 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.480356

LD_{50} is 302.24278mg/gm

95% confidence limits are 3.008951 to 30359.66 mg/gm

Appendix Table 141: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.214	3.121	5.40	3.257

Results:

Y = 3.190897 + 0.677905 X

Chi-squared 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 466.30259mg/gm

95% confidence limits are 4.949317 to 43932.83mg/gm

Appendix Table 142: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.466	4.39	16.74	4.445
6	1.778	30	7	23.333	23	4.26	4.253	4.252	15.09	4.245
3	1.477	30	6	20.000	20	4.16	4.04	4.16	13.17	4.044
1.50	1.176	30	4	13.333	13	3.87	3.827	3.873	11.10	3.844
0.75	0.875	30	2	6.667	7	3.52	3.614	3.529	9.060	3.643

Results:

Y = 3.578562 + 0.6662048 X

Chi-squared 0.3559666 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.133636

LD_{50} is 136.03048 mg/gm

95% confidence limits are 7.876344 to 2349.347 mg/gm

Appendix Table 143: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.796	4.74	18.48	4.783
6	1.778	30	11	36.667	37	4.67	4.569	4.656	17.43	4.562
3	1.477	30	7	23.333	23	4.26	4.342	4.266	15.96	4.342
1.50	1.176	30	6	20.000	20	4.16	4.115	4.17	14.13	4.122
0.75	0.875	30	4	13.333	13	3.87	3.888	3.873	11.10	3.901

Results:

Y = 3.830366 + 0.7319791 X

Chi-squared 0.3208647 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.597906

LD_{50} is 39.61924mg/gm

95% confidence limits are 8.229901 to 190.7295 mg/gm

Appendix Table 144: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.214	3.121	5.40	3.257

Results:

Y = 2.190897 + 0.677905 X

Chi-squared 0.245049 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 467.30265 mg/gm

95% confidence limits are 4.949317 to 43932.83mg/gm

Appendix Table 145: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.246	4.252	15.09	4.238
6	1.778	30	5	16.667	17	4.05	4.065	4.037	13.17	4.059
3	1.477	30	4	13.333	13	3.87	3.884	3.873	11.10	3.880
1.50	1.176	30	3	10	10	3.72	3.703	3.72	10.08	3.701
0.75	0.875	30	2	6.667	7	3.52	3.522	3.519	8.07	3.522

Results:

Y = 3.464888 + 0.5942255 X

Chi-squared 1.362634E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.583383

LD_{50} is 383.16238 mg/gm

95% confidence limits are 4.624563 to 31746.43 mg/gm

Appendix Table 146: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.498	4.57	16.74	4.511
6	1.778	30	7	23.333	23	4.26	4.339	4.266	15.96	4.347
3	1.477	30	6	20.000	20	4.16	4.180	4.17	14.13	4.182
1.50	1.176	30	5	16.667	17	4.05	4.021	4.037	13.17	4.018
0.75	0.875	30	4	13.333	13	3.87	3.862	3.873	11.10	3.853

Results:

Y = 3.800447 + 0.5461556 X

Chi-squared 0.1731033 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.196359

LD_{50} is 157.16654mg/gm

95% confidence limits are 4.730486 to 5221.689 mg/gm

Appendix Table 147: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.200	4.056	14.13	4.174
6	1.778	30	5	16.667	17	4.05	3.961	4.062	12.15	3.961
3	1.477	30	4	13.333	13	3.87	3.722	3.894	10.08	3.747
1.50	1.176	30	2	6.667	7	3.52	3.483	3.54	7.140	3.534
0.75	0.875	30	1	3.333	3	3.12	3.244	3.121	5.400	3.321

Results:

Y = 3.251909 + 0.7089487 X

Chi-squared 0.7538161with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.465751

LD_{50} is 292.24750 mg/gm

95% confidence limits are 6.531942 to 13075.51mg/gm

Appendix Table 148: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4.48	4.502	4.46	17.43	4.491
6	1.778	30	8	26.667	27	4.39	4.346	4.394	15.96	4.339
3	1.477	30	6	20.000	20	4.16	4.19	4.17	14.13	4.187
1.50	1.176	30	5	16.667	17	4.05	4.034	4.037	13.17	4.035
0.75	0.875	30	4	13.333	13	3.87	3.878	3.873	11.10	3.883

Results:

Y = 3.834423 + 0.5047025 X

Chi-squared 7.036281E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.309435

LD_{50} is 203.90819mg/gm

95% confidence limits are 3.651742 to 11385.94 mg/gm

Appendix Table149: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	8	26.667	27	4.39	4.444	4.39	16.74	4.427
6	1.778	30	7	23.333	23	4.26	4.231	4.252	15.09	4.223
3	1.477	30	5	16.667	17	4.05	4.018	4.037	13.17	4.018
1.50	1.176	30	4	13.333	13	3.87	3.805	3.873	11.10	3.814
0.75	0.875	30	2	6.667	7	3.52	3.592	3.519	8.07	3.609

Results:

Y = 3.54343 + 0.6792087 X

Chi-squared 0.1454554 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.144511

LD_{50} is 139.47976mg/gm

95% confidence limits are 8.041382 to 2419.306 mg/gm

Appendix Table 150: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10.000	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.540	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.214	3.121	5.40	3.257

Results:

Y = 3.190897 + 0.677905 X

Chi-squared 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 466.30200 mg/gm

95% confidence limits are 4.949317 to 43932.83 mg/gm

Appendix Table 151:Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.388	4.266	15.96	4.368
6	1.778	30	7	23.333	23	4.26	4.201	4.252	15.09	4.195
3	1.477	30	6	20	20	4.16	4.014	4.16	13.17	4.022
1.50	1.176	30	4	13.333	13	3.87	3.827	3.873	11.10	3.849
0.75	0.875	30	2	6.667	7	3.52	3.64	3.529	9.060	3.676

Results:

Y = 3.619936 + 0.5748606 X

Chi-squared 0.6678355 1with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.400693

LD_{50} is 251.58975mg/gm

95% confidence limits are 4.821364 to 13128.53 mg/gm

Appendix Table 152: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	4	13.333	13	3.87	3.955	3.878	12.15	3.971
6	1.778	30	3	10.000	10	3.72	3.674	3.73	9.060	3.668
3	1.477	30	2	63.667	7	3.52	3.393	3.572	6.24	3.366
1.50	1.176	30	1	3.333	3	3.12	3.112	3.116	4.62	3.063
0.75	0.875	30	0	0.000	0	0.00	2.831	2.41	2.76	2.761

Results:

Y = 2.663163 + 1.004967 X

Chi-squared 0.7569203 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.325288

LD_{50} is 211.48936 mg/gm

95% confidence limits are 6.239701 to 184.6296 mg/gm

Appendix Table 153: Larvicidal effect of root bark extract (Chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.246	4.252	15.09	4.238
6	1.778	30	5	16.667	17	4.05	4.065	4.037	13.17	4.059
3	1.477	30	4	13.333	13	3.87	3.884	3.873	11.10	3.880
1.50	1.176	30	3	10.000	10	3.72	3.703	3.72	10.08	3.701
0.75	0.875	30	2	6.667	7	3.52	3.522	3.519	8.07	3.522

Results:

Y = 3.464888 + 0.5942255 X

Chi-squared 1.362634E-02 1with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.583383

LD_{50} is 383.16238mg/gm

95% confidence limits are 4.624563 to 31746.43 mg/gm

Appendix Table 154: Larvicidal effect of root bark extract (Chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	9	30.000	30	4.48	4.532	4.46	17.43	4.518
6	1.778	30	8	26.667	27	4.39	4.328	4.394	15.96	4.322
3	1.477	30	6	20.000	20	4.16	4.124	4.17	14.13	4.126
1.50	1.176	30	4	13.333	13	3.87	3.920	3.878	12.15	3.930
0.75	0.875	30	3	10.000	10	3.72	3.716	3.72	10.08	3.733

Results:

Y = 3.669845 + 0.6522216 X

Chi-squared 0.20367621with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.039422

LD_{50} is 109.50198mg/gm

95% confidence limits are 7.786949 to 1539.84mg/gm

Appendix Table 155: Larvicidal effect of root bark extract (Chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	11	36.667	37	4.67	4.694	4.659	18.03	4.684
6	1.778	30	10	33.333	33	4.56	4.53	4.544	17.43	4.520
3	1.477	30	8	26.667	27	4.39	4.366	4.394	15.96	4.357
1.50	1.176	30	6	20.000	20	4.16	4.202	4.15	15.09	4.193
0.75	0.875	30	5	16.667	17	4.05	4.038	4.037	13.17	4.030

Results:

Y = 3.97746 + 0 .542763 X

Chi-squared 7.195998E-1with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.039422

LD_{50} is 76.55148mg/gm

95% confidence limits are 5.194382 to 1128.167mg/gm

Appendix Table 156: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	7	23.333	23	4.26	4.326	4.266	15.96	4.330
6	1.778	30	7	23.333	23	4.26	4.179	4.284	14.13	4.183
3	1.477	30	5	16.667	17	4.05	4.032	4.037	13.17	4.036
1.50	1.176	30	4	13.333	13	3.87	3.885	3.873	11.10	3.889
0.75	0.875	30	3	10.000	10	3.72	3.738	3.72	10.08	3.742

Results:

Y = 3.694959 + 0.4883541 X

Chi-squared 0.2175069 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.672326

LD_{50} is 470.24668 mg/gm

95% confidence limits are 2.269123 to 97452.56 mg/gm

Appendix Table 157: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.596	4.544	17.43	4.583
6	1.778	30	9	30	30	4.48	4.472	4.48	16.74	4.464
3	1.477	30	8	26.667	27	4.39	4.348	4.394	15.96	4.346
1.50	1.176	30	7	23.333	23	4.26	4.224	4.252	15.09	4.227
0.75	0.875	30	5	16.667	17	4.05	4.100	4.056	14.13	4.108

Results:

Y = 4.069945 + 0.394317 X

Chi-squared 0.11586 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.358647

LD_{50} is 228.37415mg/gm

95% confidence limits are 1.389216 to 37542.56 mg/gm

Appendix Table 158: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.098	4.037	13.17	4.073
6	1.778	30	4	13.333	13	3.87	3.877	3.873	11.10	3.869
3	1.477	30	3	10.000	10	3.72	3.656	3.73	9.060	3.665
1.50	1.176	30	2	6.667	7	3.52	3.435	3.54	7.140	3.461
0.75	0.875	30	1	3.333	3	3.12	3.214	3.121	5.40	3.257

Results:

Y = 3.190897 + 0.677905 X

Chi-squared 0.2000492 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.668667

LD_{50} is 203.90817mg/gm

95% confidence limits are 4.949317 to 43932.83mg/gm

Appendix Table 159: Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.862	5.02	18.81	4.903
6	1.778	30	8	26.667	27	4.39	4.499	4.39	16.74	4.517
3	1.477	30	5	16.667	17	4.05	4.136	4.056	14.13	4.132
1.5	1.176	30	3	10.000	10	3.72	3.773	3.72	10.08	3.747
0.75	0.875	30	2	6.667	7	3.52	3.410	3.54	7.140	3.361

Results:

Y = 2.241499 + 1.279955 X

Chi-squared 0.8471909 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.155155

LD_{50} is 14.29405 mg/gm

95% confidence limits are 6.896696 to 29.62576mg/gm

Appendix Table 160:Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.122	5.24	19.02	5.121
6	1.778	30	11	36.667	37	4.67	4.795	4.662	18.48	4.791
3	1.477	30	8	26.667	27	4.39	4.468	4.39	16.74	4.462
1.5	1.176	30	6	20.000	20	4.16	4.141	4.17	14.13	4.132
0.75	0.875	30	4	13.333	13	3.87	3.814	3.873	11.10	3.802

Results:

Y = 2.84282 + 1.09587 X

Chi-squared 0.7402325 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.968464

LD_{50} is 9.299592mg/gm

95% confidence limits are 4.82285 to 17.93181mg/gm

Appendix Table 161:Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	22	73.333	73	5.61	5.398	5.578	18.48	5.388
6	1.778	30	14	46.667	47	4.92	5.055	4.925	19.11	5.042
3	1.477	30	9	30.000	30	4.48	4.712	4.48	18.48	4.697
1.5	1.176	30	8	26.667	27	4.39	4.369	4.394	15.96	4.351
0.75	0.875	30	6	20.000	20	4.16	4.026	4.16	13.17	4.005

Results:

Y = 2.999677 + 1.148745 X

Chi-squared 2.142655 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.741313

LD_{50} is 5.512042 mg/gm

95% confidence limits are 2.62264 to 9.48182mg/gm

Appendix Table 162: Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.702	4.818	18.48	4.721
6	1.778	30	7	23.333	23	4.26	4.388	4.266	15.96	4.394
3	1.477	30	5	16.667	17	4.05	4.074	4.037	13.17	4.068
1.5	1.176	30	3	10.000	10	3.72	3.760	3.72	10.08	3.742
0.75	0.875	30	2	6.667	7	3.52	3.446	3.54	7.140	3.416

Results:

Y = 2.467785 + 1.083531 X

Chi-squared 0.5662975 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.337003

LD_{50} is 21.72716mg/gm

95% confidence limits are 7.46617 to 63.22772mg/gm

Appendix Table 163: Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.082	5.25	19.11	5.098
6	1.778	30	10	33.333	33	4.56	4.725	4.558	18.48	4.727
3	1.477	30	7	23.333	23	4.26	4.368	4.266	15.96	4.357
1.5	1.176	30	5	16.667	17	4.05	4.011	4.037	13.17	3.987
0.75	0.875	30	3	10.000	10	3.72	3.654	3.73	9.060	3.617

Results:

Y = 2.541524 + 1.229328 X

Chi-squared 1.256035 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.999855

LD_{50} is 9.996651mg/gm

95% confidence limits are 5.429061 to 18.40706 mg/gm

Appendix Table 164: Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.334	5.422	18.48	5.324
6	1.778	30	14	46.667	47	4.92	4.990	4.915	19.02	4.980
3	1.477	30	10	33.333	33	4.56	4.646	4.551	18.03	4.636
1.5	1.176	30	7	23.333	23	4.26	4.302	4.266	15.96	4.292
0.75	0.875	30	5	16.667	17	4.05	3.958	4.062	12.15	3.948

Results:

Y = 2.94724 + 1.143358 X

Chi-squared 0.5566368 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.795377

LD_{50} is 6.242771mg/gm

95% confidence limits are 3.740099 to 10.42009 mg/gm

Appendix Table 165: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.736	4.818	18.48	4.748
6	1.778	30	8	26.667	27	4.39	4.464	4.39	16.74	4.471
3	1.477	30	6	20.000	20	4.16	4.192	4.17	14.13	4.193
1.5	1.176	30	4	13.333	13	3.87	3.92	3.878	12.15	3.915
0.75	0.875	30	3	10.000	10	3.72	3.648	3.73	9.060	3.637

Results:

Y = 2.830175 + 0.9225664 X

Chi-squared 0.3002424 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.351945

LD_{50} is 22.48768mg/gm

95% confidence limits are 6.482878 to 78.00485 mg/gm

Appendix Table 166: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.884	4.942	18.81	4.895
6	1.778	30	11	36.667	37	4.67	4.691	4.659	18.03	4.699
3	1.477	30	9	30.000	30	4.48	4.498	4.48	16.74	4.502
1.5	1.176	30	7	23.333	23	4.26	4.305	4.266	15.96	4.306
0.75	0.875	30	6	20.000	20	4.16	4.112	4.17	14.13	4.110

Results:

Y = 3.5395 + 0.6519155 X

Chi-squared 0.1553254 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.240322

LD_{50} is 17.39089 mg/gm

95% confidence limits are 3.877473 to 78.00008 mg/gm

Appendix Table 167: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.246	5.54	18.81	5.261
6	1.778	30	12	40.000	40	4.75	4.993	4.74	19.02	4.997
3	1.477	30	10	33.333	33	4.56	4.74	4.558	18.48	4.734
1.5	1.176	30	9	30.000	30	4.48	4.487	4.48	16.74	4.471
0.75	0.875	30	8	26.667	27	4.39	4.234	4.388	15.09	4.207

Results:

Y = 3.441545 + 0.8749049 X

Chi-squared 3.793177 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.781286

LD_{50} is 6.043458 mg/gm

95% confidence limits are3.140479 to 11.62988 mg/gm

Appendix Table 168: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.642	4.74	18.03	4.651
6	1.778	30	7	23.333	23	4.26	4.397	4.266	15.96	4.402
3	1.477	30	6	20.000	20	4.16	4.152	4.17	14.13	4.154
1.5	1.176	30	4	13.333	13	3.87	3.907	3.878	12.15	3.905
0.75	0.875	30	3	10.000	10	3.72	3.662	3.73	9.060	3.656

Results:

Y = 2.932368 + 0.8267577 X

Chi-squared .5012369 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.500892

LD_{50} is 31.68779mg/gm

95% confidence limits are 36.278734 to 159.9233 mg/gm

Appendix Table 169: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.798	5.00	18.48	4.808
6	1.778	30	8	26.667	27	4.39	4.585	4.376	17.43	4.589
3	1.477	30	7	23.333	23	4.26	4.372	4.266	15.96	4.370
1.5	1.176	30	6	20.000	20	4.16	4.159	4.17	14.13	4.152
0.75	0.875	30	5	16.667	17	4.05	3.946	4.062	12.15	3.933

Results:

Y = 3.297336 + 0.7264376 X
Chi-squared 1.855071 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.343854
LD_{50} is 22.07263 mg/gm
95% confidence limits are 4.80424 to 101.4106 mg/gm

Appendix Table170: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.098	5.25	19.11	5.104
6	1.778	30	11	36.667	37	4.67	4.852	4.682	18.81	4.855
3	1.477	30	10	33.333	33	4.56	4.606	4.551	18.30	4.606
1.5	1.176	30	8	26.667	27	4.39	4.36	4.394	15.96	4.357
0.75	0.875	30	6	20.000	20	4.16	4.114	4.17	14.13	4.108

Results:

Y = 3.384844 + 0.8268123 X

Chi-squared 1.100843 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.953473

LD_{50} is 8.984072 mg/gm

95% confidence limits are 3.855749 to 20.9333 mg/gm

Appendix Table 171: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.814	4.942	18.81	4.851
6	1.778	30	8	26.667	27	4.39	4.467	4.39	16.74	4.485
3	1.477	30	5	16.667	17	4.05	4.12	4.056	14.13	4.119
1.5	1.176	30	3	10.000	10	3.72	3.773	3.72	10.08	3.753
0.75	0.875	30	2	6.667	7	3.52	3.426	3.54	7.140	3.387

Results:

Y = 2.323623 + 1.215409 X
Chi-squared 0.5411558 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.202038
LD_{50} is 15.92349 mg/gm
95% confidence limits are 7.07521 to 35.83746 mg/gm

Appendix Table 172: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.136	5.24	19.02	5.137
6	1.778	30	12	40.000	40	4.75	4. 847	4.76	18.81	4.843
3	1.477	30	9	30.000	30	4.48	4.558	4.46	17.43	4.549
1.5	1.176	30	7	23.333	23	4.26	4.269	4.252	15.09	4.256
0.75	0.875	30	5	16.667	17	4.05	3.98	4.062	12.15	3.961

Results:

Y = 3.108629 + 0.9754102 X

Chi-squared 0.7385426 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.27329

LD_{50} is 8.690649mg/gm

95% confidence limits are 4.280932 to 17.64275 mg/gm

Appendix Table 173: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.3	5.5	18.48	5.288
6	1.778	30	13	43.333	43	4.82	5.014	4.825	19.11	5.005
3	1.477	30	10	33.333	33	4.56	4.728	4.558	18.48	4.721
1.5	1.176	30	9	30.000	30	4.48	4.442	4.48	16.74	4.438
0.75	0.875	30	7	23.333	23	4.26	4.156	4.284	14.13	4.154

Results:

Y = 3.329647 + 0.9421144 X
Chi-squared 2.206224 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.772984
LD_{50} is 5.929032mg/gm
95% confidence limits are 3.238254 to 10.85567 mg/gm

Appendix Table 174: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.796	4.922	18.48	4.806
6	1.778	30	8	26.667	27	4.39	4.504	4.376	17.43	4.504
3	1.477	30	6	20.000	20	4.16	4.212	4.15	15.09	4.203
1.5	1.176	30	4	13.333	13	3.87	3.92	3.878	12.15	3.902
0.75	0.875	30	3	10.000	10	3.72	3.628	3.73	9.060	3.600

Results:

Y = 2.724367 + 1.001031 X

Chi-squared 0.7385426 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.27329

LD_{50} is 18.76248 mg/gm

95% confidence limits are 6.555629 to 53.69897mg/gm

Appendix Table 175: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	16	53.333	53	5.08	4.976	5.065	19.02	4.968
6	1.778	30	10	33.333	33	4.56	4.694	4.551	18.03	4.688
3	1.477	30	8	26.667	27	4.39	4.412	4.39	16.74	4.407
1.5	1.176	30	6	20	20	4.16	4.130	4.17	14.13	4.127
0.75	0.875	30	4	13.333	13	3.87	3.848	3.873	11.10	3.846

Results:

Y = 3.030382 + 0.9320664 X
Chi-squared 0.5546074 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.113173
LD_{50} is 12.97696 mg/gm
95% confidence limits are 5.167974 to 32.5856 mg/gm

Appendix Table 176: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.228	5.462	18.81	5.244
6	1.778	30	12	40.000	40	4.75	4.936	4.74	19.02	4.940
3	1.477	30	9	30.000	30	4.48	4.644	4.470	18.03	4.636
1.5	1.176	30	8	26.667	27	4.39	4.352	4.394	15.96	4.333
0.75	0.875	30	6	20.000	20	4.16	4.060	4.16	13.17	4.030

Results:

Y = 3.147026 + 1.008332 X

Chi-squared 2.442274 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.837662

LD_{50} is 6.881171mg/gm

95% confidence limits are 3.751891 to 12.62044 mg/gm

Appendix Table 177:Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.814	4.942	18.81	4.851
6	1.778	30	8	26.667	27	4.39	4.467	4.39	16.74	4.485
3	1.477	30	5	16.667	17	4.05	4.12	4.056	14.13	4.119
1.5	1.176	30	3	10.000	10	3.72	3.773	3.72	10.08	3.753
0.75	0.875	30	2	6.667	7	3.52	3.426	3.54	7.140	3.387

Results:

Y = 2.323623 + 1.215409 X
Chi-squared 0.5411558 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.202038
LD_{50} is 15.92349mg/gm
95% confidence limits are 7.07521 to 35.83746 mg/gm

Appendix Table 178: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rdinstar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	59.667	57	5.18	5.00	5.165	19.02	5.002
6	1.778	30	10	33.333	33	4.56	4.734	4.558	18.48	4.730
3	1.477	30	8	26.667	27	4.39	4.468	4.39	16.74	4.459
1.5	1.176	30	6	20	20	4.16	4.202	4.15	15.09	4.187
0.75	0.875	30	5	16.667	17	4.05	3.936	4.062	12.15	3.915

Results:

Y = 3.125089 + 0.9027288 X

Chi-squared 1.41518 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.076938

LD_{50} is 11.93817mg/gm

95% confidence limits are 4.826464 to 29.52884mg/gm

Appendix Table 179:Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	22	73.333	73	5.61	5.376	5.578	18.48	5.357
6	1.778	30	13	43.333	43	4.82	5.063	4.825	19.11	5.048
3	1.477	30	11	36.667	37	4.67	4.75	4.662	18.48	4.740
1.5	1.176	30	8	26.667	27	4.39	4.437	4.39	16.74	4.431
0.75	0.875	30	7	23.333	23	4.26	4.124	4.284	14.13	4.122

Results:

Y = 3.224198 + 1.025789 X
Chi-squared 2.364758 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.731157
LD_{50} is 5.384638 mg/gm
95% confidence limits are 3.163387 to 9.165595 mg/gm

Appendix Table 180: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.764	4.922	18.48	4.771
6	1.778	30	9	30.000	30	4.48	4.59	4.46	17.43	4.590
3	1.477	30	7	23.333	23	4.26	4.416	4.27	16.74	4.410
1.5	1.176	30	7	23.333	23	4.26	4.242	4.252	15.09	4.229
0.75	0.875	30	6	20.000	20	4.16	4.068	4.16	13.17	4.048

Results:

Y = 3.522446 + 0.6006159 X

Chi-squared 1.216453 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.460065

LD_{50} is 28.84459mg/gm

95% confidence limits are 3.748128 to 221.9804 mg/gm

Appendix Table 181: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.736	4.818	18.48	4.748
6	1.778	30	8	26.667	27	4.39	4.464	4.39	16.74	4.471
3	1.477	30	6	20.000	20	4.16	4.192	4.17	14.13	4.193
1.5	1.176	30	4	13.333	13	3.87	3.92	3.878	12.15	3.915
0.75	0.875	30	3	10.000	10	3.72	3.648	3.73	9.060	3.637

Results:

Y = 2.830175 + 0.9225664 X
Chi-squared 0.3002424 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.351945
LD_{50} is 22.48768 mg/gm
95% confidence limits are 6.482878 to 78.00485 mg/gm

Appendix Table 182: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (3rd instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.062	5.25	19.11	5.068
6	1.778	30	11	36.667	37	4.67	4.845	4.682	18.81	4.848
3	1.477	30	9	30.000	30	4.48	4.628	4.470	18.03	4.629
1.5	1.176	30	9	30.000	30	4.48	4.411	4.48	16.74	4.409
0.75	0.875	30	7	23.333	23	4.26	4.194	4.284	14.13	4.190

Results:

Y = 3.552113 + 0.7289573 X

Chi-squared 1.81846 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.986244

LD_{50} is 9.688208mg/gm

95% confidence limits are 3.572338 to 26.2745 mg/gm

Appendix Table 183: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.798	4.818	18.48	4.799
6	1.778	30	8	26.667	27	4.39	4.391	4.394	15.96	4.395
3	1.477	30	4	13.333	13	3.87	3.984	3.878	12.15	3.990
1.5	1.176	30	3	10	10	3.72	3.577	3.75	8.07	3.586
0.75	0.875	30	1	3.333	3	3.12	3.17	3.116	4.62	3.182

Results:

Y = 2.007543 + 1.342387 X
Chi-squared 0.3967962 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.229206
LD_{50} is 16.95141mg/gm
95% confidence limits are 7.718403 to 37.22924 mg/gm

Appendix Table 184: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	5.064	5.175	19.11	5.083
6	1.778	30	11	36.667	37	4.67	4.692	4.659	18.03	4.696
3	1.477	30	6	20.000	20	4.16	4.32	4.17	15.96	4.310
1.5	1.176	30	4	13.333	13	3.87	3.948	3.878	12.15	3.924
0.75	0.875	30	3	10.000	10	3.72	3.576	3.75	8.07	3.538

Results:

Y = 2.415731 + 1.282638 X

Chi-squared 0.8912372 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.014808

LD_{50} is 10.34685mg/gm

95% confidence limits are 5.681353 to 18.84363 mg/gm

Appendix Table 185: Larvicidal effect of flower extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.400	5.429	18.03	5.400
6	1.778	30	16	53.333	53	5.08	4.983	5.065	19.02	4.976
3	1.477	30	8	26.667	27	4.39	4.566	4.376	17.43	4.553
1.5	1.176	30	5	16.667	17	4.05	4.149	4.056	14.13	4.129
0.75	0.875	30	4	13.333	13	3.87	3.732	3.894	10.08	3.706

Results:

Y = 2.475242 + 1.406459 X
Chi-squared 1.143011 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.795117
LD_{50} is 6.239028mg/gm
95% confidence limits are 4.096548 to 9.502015 mg/gm

Appendix Table 186: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.778	4.922	18.48	4.814
6	1.778	30	8	26.667	27	4.39	4.431	4.39	16.74	4.445
3	1.477	30	4	13.333	13	3.87	4.084	3.873	13.17	4.076
1.5	1.176	30	3	10	10	3.72	3.737	3.72	10.08	3.707
0.75	0.875	30	2	6.667	7	3.52	3.39	3.572	6.24	3.337

Results:

Y = 2.264115 + 1.226494 X

Chi-squared 1.152259 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.230655

LD_{50} is 17.00807mg/gm

95% confidence limits are 7.326695 to 39.48226mg/gm

Appendix Table 187: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.138	5.24	19.02	5.149
6	1.778	30	12	40.000	40	4.75	4.762	4.74	18.48	4.761
3	1.477	30	6	20.000	20	4.16	4.386	4.17	15.96	4.373
1.5	1.176	30	5	16.667	17	4.05	4.010	4.037	13.17	3.985
0.75	0.875	30	3	10.000	10	3.72	3.634	3.73	9.060	3.597

Results:

Y = 2.469744 + 1.28846 X
Chi-squared 1.0194 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.963783
LD_{50} is 9.199899mg/gm
95% confidence limits are 5.266581 to 16.07079mg/gm

Appendix Table188: Larvicidal effect of flower extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	22	73.333	73	5.61	5.484	5.591	18.03	5.478
6	1.778	30	16	53.333	53	5.08	5.090	5.075	19.11	5.080
3	1.477	30	9	30	30	4.48	4.696	4.470	18.03	4.682
1.5	1.176	30	7	23.333	23	4.26	4.302	4.266	15.96	4.284
0.75	0.875	30	5	16.667	17	4.05	3.908	4.062	12.15	3.886

Results:

Y = 2.728861 + 1.32203 X

Chi-squared 1.422516 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.717918

LD_{50} is 5.222975 mg/gm

95% confidence limits are 3.46128 to 7.881324mg/gm

Appendix Table 189: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50	50	5.00	4.862	5.02	18.81	4.903
6	1.778	30	8	26.667	27	4.39	4.499	4.39	16.74	4.517
3	1.477	30	5	16.667	17	4.05	4.136	4.056	14.13	4.132
1.5	1.176	30	3	10	10	3.72	3.773	3.72	10.08	3.747
0.75	0.875	30	2	6.667	7	3.52	3.410	3.54	7.140	3.362

Results:

Y = 2.241499 + 1.279955 X
Chi-squared 0.8471909 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.155155
LD_{50} is 14.29405mg/gm
95% confidence limits are 6.896696 to 29.62576 mg/gm

Appendix Table 190: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	5.01	5.175	19.11	5.023
6	1.778	30	10	33.333	33	4.56	4.697	4.551	18.03	4.699
3	1.477	30	7	23.333	23	4.26	4.384	4.266	15.96	4.375
1.5	1.176	30	5	16.667	17	4.05	4.071	4.037	13.17	4.051
0.75	0.875	30	4	13.333	13	3.87	3.758	3.894	10.08	3.728

Results:

Y = 2.78658 + 1.075371 X

Chi-squared 1.309761 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.058285

LD_{50} is 11.43628mg/gm

95% confidence limits are 5.401934 to 24.21142 mg/gm

Appendix Table 191: Larvicidal effect of leaf extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.326	5.50	18.48	5.318
6	1.778	30	13	43.333	43	4.82	4.976	4.815	19.02	4.963
3	1.477	30	9	30.000	30	4.48	4.626	4.470	18.03	4.609
1.5	1.176	30	7	23.333	23	4.26	4.276	4.252	15.09	4.254
0.75	0.875	30	5	16.667	17	4.05	3.926	4.062	12.15	3.899

Results:

Y = 2.868699 + 1.177865 X
Chi-squared 1.698786 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.809461
LD_{50} is 6.448529 mg/gm
95% confidence limits are 3.889667 to 10.69077 mg/gm

Appendix Table 192: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.426	4.57	16.74	4.463
6	1.778	30	5	16.667	17	4.05	4.165	4.056	14.13	4.186
3	1.477	30	4	13.333	13	3.87	3.904	3.878	12.15	3.909
1.5	1.176	30	2	6.667	7	3.52	3.643	3.529	9.060	3.632
0.75	0.875	30	2	6.667	7	3.52	3.382	3.572	6.24	3.356

Results:

Y = 2.551239 + 0.9192638 X

Chi-squared .8320189 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.663829

LD_{50} is 46.11358mg/gm

95% confidence limits are 7.581674 to 280.474 mg/gm

Appendix Table 193: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.796	4.922	18.48	4.806
6	1.778	30	8	26.667	27	4.39	4.504	4.376	17.43	4.504
3	1.477	30	6	20	20	4.16	4.212	4.15	15.09	4.203
1.5	1.176	30	4	13.333	13	3.87	3.92	3.878	12.15	3.902
0.75	0.875	30	3	10	10	3.72	3.628	3.73	9.060	3.600

Results:

Y = 2.724367 + 1.001031 X
Chi-squared 0.7385426 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.27329
LD_{50} is 18.76248mg/gm
95% confidence limits are 6.555629 to 53.69897mg/gm

Appendix Table 194: Larvicidal effect of leaf extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	17	56.667	57	5.18	5.00	5.165	19.02	5.002
6	1.778	30	10	33.333	33	4.56	4.734	4.558	18.48	4.730
3	1.477	30	8	26.667	27	4.39	4.468	4.39	16.74	4.459
1.5	1.176	30	6	20.000	20	4.16	4.202	4.15	15.09	4.187
0.75	0.875	30	5	16.667	17	4.05	3.936	4.062	12.15	3.915

Results:

Y = 3.125089 + 0.9027288 X

Chi-squared 1.41518 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.076938

LD_{50} is 11.93817mg/gm

95% confidence limits are 4.826464 to 29.52884 mg/gm

Appendix Table 195: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.862	5.02	18.81	4.903
6	1.778	30	8	26.667	27	4.39	4.499	4.39	16.74	4.517
3	1.477	30	5	16.667	17	4.05	4.136	4.056	14.13	4.132
1.5	1.176	30	3	10.000	10	3.72	3.773	3.72	10.08	3.747
0.75	0.875	30	2	6.667	7	3.52	3.410	3.54	7.140	3.362

Results:

Y = 2.241499 + 1.279955 X
Chi-squared 0.8471909 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.155155
LD_{50} is 14.29405mg/gm
95% confidence limits are 6.896696 to 29.62576 mg/gm

Appendix Table 196: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.076	5.25	19.11	5.103
6	1.778	30	11	36.667	37	4.67	4.720	4.662	18.18	4.730
3	1.477	30	6	20.000	20	4.16	4.364	4.17	15.96	4.357
1.5	1.176	30	4	13.333	13	3.87	4.008	3.873	13.17	3.984
0.75	0.875	30	4	13.333	13	3.87	3.652	3.931	9.060	3.611

Results:

Y = 2.525887 + 1.239663 X

Chi-squared 2.146212 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.995795

LD_{50} is 9.903644mg/gm

95% confidence limits are 5.42245 to 18.08817 mg/gm

Appendix Table197: Larvicidal effect of root bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.380	5.50	18.48	5.383
6	1.778	30	15	50.000	50	5.00	5.002	5.00	19.11	4.995
3	1.477	30	8	26.667	27	4.39	4.624	4.389	18.03	4.607
1.5	1.176	30	6	20.000	20	4.16	4.246	4.15	15.09	4.219
0.75	0.875	30	5	16.667	17	4.05	3.868	4.077	11.10	3.831

Results:

Y = 2.70345 + 1.288834 X
Chi-squared 1.853952 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.781882
LD_{50} is 5.051757mg/gm
95% confidence limits are 3.867013 to 9.470812mg/gm

Appendix Table 198: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	**Log dose**	**# used**	**# Kill**	**% Kill**	**Corr. %**	**Emp. probit**	**Expt probit**	**Work probit**	**Weight**	**Final probit**
12	2.079	30	12	40.000	40	4.75	4.554	4.74	17.43	4.595
6	1.778	30	6	20.000	20	4.16	4.244	4.15	15.09	4.261
3	1.477	30	3	10.000	10	3.72	3.934	3.74	12.15	3.926
1.5	1.176	30	2	6.667	7	3.52	3.624	3.529	9.060	3.591
0.75	0.875	30	2	6.667	7	3.52	3.314	3.572	6.24	3.256

Results:

Y = 2.283466 + 1.111911 X

Chi-squared 1.625594 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.443123

LD_{50} is 27.74103 mg/gm

95% confidence limits are 8.339966 to 92.27432mg/gm

Appendix Table 199: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	14	46.667	47	4.92	4.738	4.922	18.48	4.780
6	1.778	30	8	26.667	27	4.39	4.446	4.39	16.74	4.466
3	1.477	30	4	13.333	13	3.87	4.154	3.904	14.13	4.153
1.5	1.176	30	4	13.333	13	3.87	3.862	3.873	11.10	3.840
0.75	0.875	30	3	10	10	3.72	3.570	3.75	8.07	3.526

Results:

Y = 2.615665 + 1.04086 X
Chi-squared 1.763982 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.290736
LD_{50} is 19.53158mg/gm
95% confidence limits are 6.878268 to 55.46159mg/gm

Appendix Table 200: Larvicidal effect of root bark extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.298	5.54	18.81	5.349
6	1.778	30	13	43.333	43	4.82	4.891	4.838	18.81	4.914
3	1.477	30	6	20.000	20	4.16	4.484	4.18	16.74	4.479
1.5	1.176	30	5	16.667	17	4.05	4.077	4.037	13.17	4.044
0.75	0.875	30	4	13.333	13	3.87	3.670	3.931	9.060	3.608

Results:

Y = 2.343537 + 1.445517 X

Chi-squared 3.231087 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.837726

LD_{50} is 6.882175 mg/gm

95% confidence limits are 4.493163 to 10.54143 mg/gm

Appendix Table 201:Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.826	5.02	18.81	4.884
6	1.778	30	8	26.667	27	4.39	4.463	4.39	16.74	4.490
3	1.477	30	4	13.333	13	3.87	4.10	3.904	14.13	4.096
1.5	1.176	30	3	10.000	10	3.72	3.737	3.72	10.08	3.702
0.75	0.875	30	2	6.667	7	3.52	3.374	3.572	6.24	3.309

Results:

Y = 2.164293 + 1.307853 X
Chi-squared 1.474346 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.168215
LD_{50} is 14.73041mg/gm
95% confidence limits are 7.106956 to 30.53135mg/gm

Appendix Table202:Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar)) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.106	5.24	19.02	5.125
6	1.778	30	11	36.667	37	4.67	4.72	4.662	18.48	4.723
3	1.477	30	6	20.000	20	4.16	4.334	4.17	15.96	4.322
1.5	1.176	30	4	13.333	13	3.87	3.948	3.878	12.15	3.920
0.75	0.875	30	3	10.000	10	3.72	3.562	3.75	8.07	3.520

Results:

Y = 2.349999 + 1.33471X

Chi-squared 1.142679 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.985451

LD_{50} is 9.670546mg/gm

95% confidence limits are 5.555684 to 16.83313mg/gm

Appendix Table 203:Larvicidal effect of root wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	21	70.000	70	5.52	5.414	5.51	18.03	5.408
6	1.778	30	15	50.000	50	5.00	5.059	5.00	19.11	5.053
3	1.477	30	10	33.333	33	4.56	4.704	4.558	18.48	4.697
1.5	1.176	30	8	26.667	27	4.39	4.349	4.394	15.96	4.342
0.75	0.875	30	5	16.667	17	4.05	3.994	4.062	12.15	3.987

Results:

Y = 2.953416 + 1.180638 X
Chi-squared 0.7115708 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 1.733456
LD_{50} is 5.413227mg/gm
95% confidence limits are 3.39452 to 8.632451mg/gm

Appendix Table 204:Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.584	4.74	17.43	4.613
6	1.778	30	7	23.333	23	4.26	4.324	4.266	15.96	4.335
3	1.477	30	4	13.333	13	3.87	4.064	3.873	13.17	4.058
1.5	1.176	30	3	10.000	10	3.72	3.804	3.72	11.10	3.780
0.75	0.875	30	3	10.000	10	3.72	3.544	3.75	8.07	3.503

Results:

Y = 2.695882 + 0.9220272 X

Chi-squared 1.34199 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.49897

LD_{50} is 31.54786mg/gm

95% confidence limits are 7.169451 to 138.8205 mg/gm

Appendix Table 205:Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	15	50.000	50	5.00	4.828	5.02	18.81	4.860
6	1.778	30	9	30.000	30	4.48	4.541	4.46	17.43	4.556
3	1.477	30	5	16.667	17	4.05	4.254	4.048	15.09	4.252
1.5	1.176	30	4	13.333	13	3.87	3.967	3.878	12.15	3.948
0.75	0.875	30	4	13.333	13	3.87	3.680	3.931	9.060	3.644

Results:

Y = 2.76067 + 1.009706 X
Chi-squared 2.076032 with 3 degrees of freedom
No significant heterogeneity
Log LD_{50} is 2.217804
LD_{50} is 16.51215mg/gm
95% confidence limits are 6.224204 to 43.80498mg/gm

Appendix Table 206:Larvicidal effect of root wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.07	5.25	19.11	5.092
6	1.778	30	12	40.000	40	4.75	4.803	4.76	18.81	4.811
3	1.477	30	7	23.333	23	4.26	4.536	4.264	17.43	4.530
1.5	1.176	30	7	23.333	23	4.26	4.269	4.252	15.09	4.249
0.75	0.875	30	6	20.000	20	4.16	4.002	4.16	13.17	3.968

Results:

Y = 3.151846 + 0.9330014 X

Chi-squared 2.244703 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.980869

LD_{50} is 9.569064 mg/gm

95% confidence limits are 4.401974 to 20.80134mg/gm

Appendix Table 207: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	27	90.000	90	6.28	6.247	6.23	11.10	6.230
6	1.778	30	24	80.000	80	5.85	5.918	5.87	14.13	5.906
3	1.477	30	22	73.333	73	5.61	5.589	5.584	17.43	5.582
1.5	1.176	30	18	60.000	60	5.25	5.261	5.28	18.81	5.257
0.75	0.875	30	15	50.000	50	5.00	4.932	4.99	19.02	4.933

Results:

Y =3.751+1.078X

Chi-squared is 0.150 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.159

LD_{50} is 14.42565 mg/g

95% confidence limits are 8.743 to 23.800 mg/g

Appendix Table 208: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	28	93.333	93	6.48	6.336	6.424	10.08	6.267
6	1.778	30	25	83.333	83	5.95	6.009	5.923	13.17	5.957
3	1.477	30	21	70.000	70	5.52	5.682	5.52	16.74	5.647
1.5	1.176	30	19	63.333	63	5.33	5.355	5.318	18.48	5.337
0.75	0.875	30	16	53.333	53	5.08	5.028	5.075	19.11	5.027

Results:

Y =3.898+1.029X

Chi-squared is 0.594 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.070

LD_{50} is 11.76264 mg/g

95% confidence limits are 6.659 to 20.775 mg/g

Appendix Table 209: Larvicidal effect of seed extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	29	96.667	97	6.88	6.599	6.759	8.07	6.604
6	1.778	30	27	90.000	90	6.28	6.287	6.23	11.10	6.292
3	1.477	30	25	83.333	83	5.95	5.975	5.984	14.13	5.980
1.5	1.176	30	21	70.000	70	5.52	5.662	5.52	16.74	5.668
0.75	0.875	30	20	66.667	67	5.44	5.350	5.422	18.48	5.355

Results:

Y = 4.218+1.037X

Chi-squared is 0.702 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 0.754

LD_{50} is 5.67895 mg/g

95% confidence limits are 2.591 to 12.443 mg/g

Appendix Table 210: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	27	90.000	90	6.28	6.190	6.27	12.15	6.149
6	1.778	30	24	80.000	80	5.85	5.890	5.80	15.09	5.860
3	1.477	30	21	70.000	70	5.52	5.591	5.50	17.43	5.570
1.5	1.176	30	18	60.000	60	5.25	5.292	5.28	18.81	5.281
0.75	0.875	30	15	50.000	50	5.00	4.993	4.99	19.02	4.991

Results:

Y =3.646+0.962X

Chi-squared is 0.344 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.407

LD_{50} is 25.53965 mg/gm

95% confidence limits are 14.159 to 46.063 mg/gm

Appendix Table 211: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	26	86.667	87	6.13	6.085	6.087	13.17	6.033
6	1.778	30	24	80.000	80	5.85	5.808	5.80	15.09	5.768
3	1.477	30	20	66.667	67	5.44	5.530	5.416	17.43	5.502
1.5	1.176	30	17	56.667	57	5.18	5.253	5.202	18.81	5.237
0.75	0.875	30	15	50.000	50	5.00	4.976	4.99	19.02	4.971

Results:

Y =4.004+0.882X

Chi-squared is 0.234 with3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.129

LD_{50} is 13.470 mg/g

95% confidence limits are 7.182 to 25.264 mg/g

Appendix Table 212: Larvicidal effect of seed extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	29	94.333	94	6.49	6.346	6.424	11.08	6.367
6	1.778	30	26	84.333	84	5.97	6.109	5.923	14.17	5.959
3	1.477	30	22	71.000	71	5.53	5.692	5.52	17.74	5.648
1.5	1.176	30	18	64.333	64	5.45	5.365	5.318	19.48	5.338
0.75	0.875	30	16	54.333	54	5.36	5.038	5.075	20.12	5.029

Results:

Y =3.796+1.036X

Chi-squared is 0.694 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.080

LD_{50} is 9.66295 mg/g

95% confidence limits are 7.665 to 22.856

Appendix Table 213: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	10	33.333	33	4.56	4.590	4.544	17.43	4.578
6	1.778	30	8	26.667	27	4.39	4.380	4.394	15.96	4.375
3	1.477	30	6	20.000	20	4.16	4.171	4.170	14.13	4.172
1.5	1.176	30	5	16.667	17	4.05	3.962	4.062	12.15	3.970
0.75	0.875	30	3	10.000	10	3.72	3.753	3.72	10.08	3.766

Results:

Y =3.027+0.674X

Chi-squared is 0.168 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.927

LD_{50} is 846.206995 mg/g

95% confidence limits are107.583 to 6655.92 mg/g

Appendix Table 214: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	12	40.000	40	4.75	4.774	4.74	18.48	4.761
6	1.778	30	10	33.333	33	4.56	4.561	4.544	17.43	4.553
3	1.477	30	8	26.667	27	4.39	4.348	4.394	15.96	4.346
1.5	1.176	30	6	20.000	20	4.16	4.135	4.17	14.13	4.138
0.75	0.875	30	4	13.333	13	3.87	3.922	3.878	12.15	3.931

Results:

Y =3.175+0.690X

Chi-squared is 9.481E-02 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.648

LD_{50} is 444.64856 mg/g

95% confidence limits are 93.614 to 2111.985 mg/g

Appendix Table215: Larvicidal effect of stem bark extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	22	73.333	73	5.61	5.609	5.61	16.74	5.599
6	1.778	30	19	63.333	63	5.33	5.377	5.318	18.48	5.369
3	1.477	30	17	56.667	57	5.18	5.146	5.165	19.02	5.139
1.5	1.176	30	14	46.667	47	4.92	4.914	4.915	19.02	4.909
0.75	0.875	30	12	40.000	40	4.75	4.683	4.74	18.03	4.678

Results:

Y = 3.840+0.765X

Chi-squared is 0.18966 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.518

LD_{50} is 32.924669 mg/g

95% confidence limits are18.637 to 58.163 mg/g

Appendix Table 216: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	4	13.333	13	3.87	3.924	3.878	12.15	3.942
6	1.778	30	3	10.000	10	3.72	3.688	3.73	9.060	3.689
3	1.477	30	2	6.667	7	3.52	3.452	3.54	7.140	3.436
1.5	1.176	30	1	3.333	3	3.12	3.216	3.121	5.40	3.183
0.75	0.875	30	1	3.333	3	3.12	2.979	3.172	3.30	2.930

Results:

Y =1.755+0.840X

Chi-squared is 0.647 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 3.861

LD_{50} is 7266.5765 mg/g

95% confidence limits are 248.445 to 212533.80 mg/g

Appendix Table 217:Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (4thinstar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	13	43.333	43	4.82	4.790	4.818	18.48	4.800
6	1.778	30	10	33.333	33	4.56	4.546	4.544	17.43	4.551
3	1.477	30	8	26.667	27	4.39	4.302	4.394	15.96	4.301
1.5	1.176	30	4	13.333	13	3.87	4.058	3.873	13.17	4.052
0.75	0.875	30	3	10.000	10	3.72	3.814	3.72	11.10	3.803

Results:

Y =2.645+0.828X

Chi-squared is 0.954 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.844

LD_{50} is 697.739964 mg/g

95% confidence limits are 208.878 to 2330.734 mg/g

Appendix Table 218: Larvicidal effect of stem bark extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	24	80.000	80	5.85	5.817	5.80	15.09	5.784
6	1.778	30	20	66.667	67	5.44	5.502	5.416	17.43	5.476
3	1.477	30	18	60.000	60	5.25	5.187	5.24	19.02	5.169
1.5	1.176	30	13	43.333	43	4.82	4.873	4.838	18.81	4.862
0.75	0.875	30	10	33.333	33	4.56	4.558	4.544	17.43	4.554

Results:

Y =3.127+1.021X

Chi-squared is 0.176 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.834

LD_{50} is 68.312635 mg/g

95% confidence limits are 44.174 to 105.639 mg/g

Appendix Table 219:Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	5	16.667	17	4.05	4.024	4.037	13.17	4.019
6	1.778	30	4	13.333	13	3.87	3.931	3.878	12.15	3.928
3	1.477	30	4	13.333	13	3.87	3.838	3.873	11.10	3.838
1.5	1.176	30	3	10.000	10	3.72	3.745	3.72	10.08	3.748
0.75	0.875	30	3	10.000	10	3.72	3.652	3.73	9.060	3.658

Results:

Y =3.238+0.300X

Chi-squared is 0.123 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 5.872

LD_{50} is 7450.9956 mg/g

95% confidence limits are 2903859 to 1.912E+12 mg/g

Appendix Table 220: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	18	60.000	60	5.25	5.219	5.28	18.81	5.230
6	1.778	30	15	50.000	50	5.00	5.005	5.00	19.11	5.013
3	1.477	30	12	40.000	40	4.75	4.792	4.74	18.48	4.795
1.5	1.176	30	10	33.333	33	4.56	4.578	4.544	17.43	4.578
0.75	0.875	30	8	26.667	27	4.39	4.365	4.394	15.96	4.360

Results:

Y =3.351+0.722X

Chi-squared is 0.155 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.283

LD_{50} is 192.02665 mg/g

95% confidence limits are 88.802 to 415.237 mg/g

Appendix Table 221: Larvicidal effect of stem wood extract (chloroform) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	25	83.333	83	5.95	5.970	5.984	14.13	5.989
6	1.778	30	22	73.333	73	5.61	5.680	5.61	16.74	5.693
3	1.477	30	20	66.667	67	5.44	5.391	5.422	18.48	5.397
1.5	1.176	30	18	60.000	60	5.25	5.102	5.24	19.02	5.100
0.75	0.875	30	12	40.000	40	4.75	4.813	4.76	18.81	4.804

Results:

Y =3.429+0.984X

Chi-squared is 0.574 with 3 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.597

LD_{50} is 39.52767 mg/g

95% confidence limits are 24.212 to 64.528 mg/g

Appendix Table 222:Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 24h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	6	20.000	20	4.16	4.150	4.17	14.13	4.155
6	1.778	30	5	16.667	17	4.05	4.026	4.037	13.17	4.030
3	1.477	30	4	13.333	13	3.87	3.902	3.878	12.15	3.904
1.5	1.176	30	3	10.000	10	3.72	3.778	3.72	10.08	3.779
0.75	0.875	30	3	10.000	10	3.72	3.654	3.73	9.060	3.654

Results:

Y = 3.073+0.416X

Chi-squared is 0.101 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 4.636

LD_{50} is 43235.51365 mg/g

95% confidence limits are 31.943 to 5.852E+07 mg/g

Appendix Table 223: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 48h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	20	66.667	67	5.44	5.468	5.429	18.03	5.474
6	1.778	30	18	60.000	60	5.25	5.226	5.28	18.81	5.228
3	1.477	30	15	50.000	50	5.00	4.984	4.99	19.02	4.982
1.5	1.176	30	12	40.000	40	4.75	4.742	4.74	18.48	4.736
0.75	0.875	30	9	30.000	30	4.48	4.501	4.46	17.43	4.490

Results:

Y=3.348+0.817X

Chi-squared is 0.105 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 2.022

LD_{50} is 105.24269 mg/g

95% confidence limits are 60.326 to 183.602 mg/g

Appendix Table 224: Larvicidal effect of stem wood extract (methanol) of *A. indica* against *T. castaneum* larva (4th instar) after 72h of exposure.

Dose mg/gm	Log dose	# used	# Kill	% Kill	Corr. %	Emp. probit	Expt probit	Work probit	Weight	Final probit
12	2.079	30	25	83.333	83	5.95	5.767	5.926	15.96	5.734
6	1.778	30	19	63.333	63	5.33	5.483	5.321	18.03	5.457
3	1.477	30	16	53.333	53	5.08	5.199	5.065	19.02	5.180
1.5	1.176	30	14	46.667	47	4.92	4.915	4.915	19.02	4.903
0.75	0.875	30	11	36.667	37	4.67	4.631	4.659	18.03	4.625

Results:

Y = 3.337+0.921X

Chi-squared is 1.228 with 4 degrees of freedom

No significant heterogeneity

Log LD_{50} is 1.805

LD_{50} is 63.79169mg/g

95% confidence limits are 39.563 to 102.857 mg/g

Chapter-7
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