

## TOXICITY OF SELECTED MEDICINAL PLANTS EXTRACTS AGAINST RICE WEEVILS

Tariq Javed<sup>1, 4</sup>, M. Farhanullah Khan<sup>1</sup>, Habiballah Rana<sup>1, 6</sup> \*, Hina Qasim<sup>5</sup>, M. Faheem Akbar<sup>2</sup>, Jahangir Khan Achakzai<sup>3</sup>, M. Sohail Khan<sup>1</sup>, S. Naveed Ahmed Hashmi<sup>4</sup> and M. Imran<sup>7</sup>

<sup>1</sup>Laboratory of toxicology, Department of Zoology, University of Karachi, Karachi, Pakistan

<sup>2</sup>Department of Agriculture & Agribusiness Management, University of Karachi, Pakistan

<sup>3</sup>Institute of Biochemistry, University of Balochistan, Quetta, Pakistan.

<sup>4</sup>Department of Zoology, Government College for Men, Karachi, Pakistan.

<sup>5</sup>Department of Gynecology, Baqai Medical University Hospital, Toll plaza Karachi, Pakistan

<sup>6</sup>Department of Zoology, Government Degree College, Gulzar e Hijri, Karachi, Pakistan.

<sup>7</sup>Department of Botany, Government Degree College, Gulzar e Hijri, Karachi, Pakistan.

\*Corresponding author: Tel: + 92 300 7067 397, Email address: drhabibrana@gmail.com

---

### ABSTRACT

Many medicinal plants have been used traditionally to control a wide range of ectoparasites, pest insects and vectors of different diseases and for the treatment of many human diseases. This study aimed to understand the toxicity and larvicidal activity of *Annona squamosa* and *Guaiacum officinale* extracts against *Sitophilus zeamais*. Results of different experiments confirmed the higher efficacy of plant extracts. Lower doses of *Guaiacum officinale* caused 19.00% mortality while 72.00% mortality was recorded under higher doses. Similarly under the toxic effects of *Annona squamosa* 36.00% mortality was recorded under lower doses while 100.00% mortality was recorded under higher doses.

**Key words:** Toxicity, Vectors, Parasites, Medicinal plants, Toxicity, Acute.

---

### INTRODUCTION

Many plants have been used to treat a wide range of diseases. According to World Health Organization (WHO) report, nearly 80% human population of under developing countries rely on traditional medicine therapies for their routine health problems Masimba *et al.* (2016). Plants have a wide range of chemical substances that are responsible for biological activities. In order to understand their specific potentials extensive research is being carried out by many researchers throughout the world. Many phytochemicals also act as an alternate source as mosquito control agents (Wink, 1993). Phytochemicals are not only active for control of pests but also reduce the risk of potentially unfavourable ecological effects. They also prevent the possibility of development of resistance due to prolonged use of synthetic chemicals (Monzon *et al.*, 1994). Bio-insecticides are plant derivatives. They are used to control stored grain pest. They have multi-directional mode of action such as Toxicant, Anti-feedant, Anti-deterrent, Repellent, Attractant and Grain protectant (Omar *et al.*, 2012).

This study evaluates the potentiality of toxicity of *Annona squamosa* and *Guaiacum officinale*. *Annona squamosa* belongs to the family Annonaceae. Different parts of the plants like leaves, bark, fruits and seeds have been used to treat the different diseases and to control the wide variety of pest insects (Ansante *et al.*, 2015). *Guaiacum officinale* belongs to the family Zygophyllaceae. It is widespread in tropical, subtropical and warm temperate, often in drier areas.

### MATERIALS AND METHODS

**Rearing / Culture technique:** Insects (*Sitophilus zeamais*) were reared in the laboratory in dark at 29 - 31°C temperature with relative humidity of 65% – 75% on whole maize grain and rice, respectively as food, in glass jars covered with muslin cloths. About 2 weeks adult weevils were used in the experiment.

**Preparation of Plant Extracts:** The plant extracts were obtained from the fresh leaves of the *Annona squamosa* and *Guaiacum officinale*. The leaves were cleaned and processed separately. Air dried and weighed 100g each after grinding in a miller, materials were dipped in 100 mL of ethyl alcohol and homogenized with the help of homogenizer and left for 48 hours, for the extraction of dissolved contents. After 48 hours the contents were carefully collected through a filter paper and stored in a glass container. Remaining materials were again dipped in

fresh 100 mL ethyl alcohol and left for 24 hours. This process was repeated three times to extract all the contents. The ethanolic extracts were concentrated up to 50 mL under vacuum in a rotary evaporator at 40 °C and stored in volumetric flask for further experiments.

**Toxicity Testing by Contact Method:** The toxicity estimation of plant extracts were carried out by contact method in the 0.5 - 10 $\mu$ L extract to test insects under the control laboratory conditions i.e., 29 - 31°C temperature and relative humidity of 65 - 75%, in complete darkness. The control batches were kept in all cases to observe the effect of other unseen factors.

**Statistical Analysis:** The mortality data was corrected through probit analysis. SPSS and Biostat 2009 software were used to analyze the data. Solution of selected samples was prepared in different concentrations by the help of Charles equation,  $C_1V_1=C_2V_2$  (Langmuir *et al.*, 197).

## RESULTS AND DISCUSSION

It was observed that toxicity of *Guaiacum officinale* was determined 19.00% mortality was recorded under lower doses and 72.00% mortality was recorded under higher doses (Table 1). Similarly toxicity of *Annona squamosa* leaves extract were recorded 36.00% mortality under lower doses and 100.00% mortality was recorded under higher doses (Table 3) in the present study.

Table 1. Probit analysis of mortality data of *Guaiacum officinale* leaves extract against *Sitophilus zeamais*.

Dose ( $\mu$ L)	Total insects	Insects killed (%)	Probit Percent mortality	Difference	Chi-square	Probit (Y)
2.00	100	19	0.1818	0.8142	0.03645	4.1222
3.00	100	35	0.3555	-0.556	0.00869	4.6151
4.00	100	49	0.5045	-1.4521	0.04180	4.9750
5.00	100	62	0.6207	-0.0729	8.57811	5.3050
6.00	100	72	0.7085	1.1400	0.01834	5.5824

Table 2. Percentile probit mortality analysis under toxic effects of *Guaiacum officinale* extract against *Sitophilus zeamais*.

Percentile	Probit (Y)	Log10[Dose (Stimulus)]	Standard Error	Dose (Stimulus)	Standard Error
1	2.6732	-0.1633	0.0969	0.6866	0.1544
5	3.3548	0.0598	0.0693	1.1477	0.1840
10	3.7183	0.1788	0.0550	1.5094	0.1917
16	4.0056	0.2728	0.0441	1.8743	0.1905
20	4.1585	0.3229	0.0385	2.1034	0.1866
25	4.3258	0.3777	0.0327	2.3860	0.1799
30	4.4760	0.4268	0.0280	2.6719	0.1723
40	4.7471	0.5156	0.0215	3.2776	0.1626
50	5.0000	0.5983	0.0199	3.9660	0.1817
60	5.2529	0.6811	0.0234	4.7989	0.2591
70	5.5240	0.7699	0.0310	5.8867	0.4203
75	5.6742	0.8190	0.0360	6.5922	0.5472
80	5.8415	0.8738	0.0420	7.4780	0.7243
84	5.9944	0.9239	0.0477	8.3918	0.9238
90	6.2817	1.0179	0.0588	10.4208	1.4152
95	6.6452	1.1369	0.0732	13.7050	2.3217
99	7.3268	1.3600	0.1008	22.9076	5.3664

Table 3. Probit analysis of mortality data of *Annona squamosa* leaves extract against *Sitophilus zeamais*.

Dose (µl)	Total insects	Insects killed (%)	Probit Percent mortality	Difference	Chi-square	Probit (Y)
2.00	100	36	0.3296	3.0325	0.2785	4.6419
3.00	100	67	0.7164	-4.694	0.3074	5.4394
4.00	100	89	0.9021	-1.211	0.0162	6.2266
5.00	100	99	0.9679	2.2008	0.0500	7.3267
6.00	100	99	0.9895	0.0491	2.4408	7.3267

Table 4. Percentile probit mortality analysis under toxic effects of *Annona squamosa* leaves extract against *Sitophilus zeamais*.

Percentile	Probit (Y)	Log10[Dose (Stimulus)]	Standard Error	Dose (Stimulus)	Standard Error
1	2.6732	-0.0263	0.0476	0.9413	0.1034
5	3.3548	0.0920	0.0374	1.2359	0.1066
10	3.7183	0.1551	0.0321	1.4292	0.1057
16	4.0056	0.2049	0.0280	1.6031	0.1035
20	4.1585	0.2315	0.0259	1.7041	0.1018
25	4.3258	0.2605	0.0237	1.8219	0.0995
30	4.4760	0.2866	0.0218	1.9346	0.0971
40	4.7471	0.3336	0.0186	2.1559	0.0923
50	5.0000	0.3775	0.0161	2.3853	0.0883
60	5.2529	0.4214	0.0143	2.6390	0.0866
70	5.5240	0.4685	0.0134	2.9409	0.0907
75	5.6742	0.4945	0.0135	3.1228	0.0970
80	5.8415	0.5236	0.0141	3.3387	0.1082
84	5.9944	0.5501	0.0150	3.5491	0.1226
90	6.2817	0.6000	0.0175	3.9809	0.1608
95	6.6452	0.6631	0.0217	4.6033	0.2301
99	7.3268	0.7814	0.0309	6.0445	0.4304

Phytopesticides are safe, biodegradable and promising alternate of hazardous insecticides. Larvicidal activity of *Annona squamosa* leaves extract shows 100% mortality at higher doses, this rate was observed after 24hours of exposure. Veni *et al.* (2016) also recorded the 100% mortality (zero hatchability) at higher doses of different plant extracts. Under test plant extracts exhibited very encouraging larvicidal results. Asawalam *et al.* (2012) reported that powder of the *Annona squamosa* leaves extract lead to suffocation and finally death of the stored grain pests. They reported that *Annona squamosa* killed 98.00% adult weevil and even no weevils were alive after few days of exposure. *Annona squamosa* leaves extract also found most promising larvicidal agent against the larvae of *Ae. aegypti*. Dwivedi and Karwasara, (2003) also recorded the same results against the *Culex quinquefasciatus*. Plants could be an alternate source of stored grain pest larvicides because they produce a wide range of bioactive chemicals and mostly are free from hazardous effects. Derivatives of *Annona squamosa* were effective against many agricultural pests (Leatemia and Isman 2004; Trindade *et al.*, 2006). Bioactivity of theses extracts were associated with the presence of triglycerides, alkaloids and acetogenins and synergistic effects of these compounds (Ribeiro *et al.*, 2014c; Ansante *et al.*, 2015). Use of these bioactive compounds could reduce the cost and environmental pollutions. Further extensive research is needed to identify these active compounds. Results of present studies

employ important implications in the control of stored grain pests. Under test plants are easily available in large quantities and can be use locally in the control of the targeted pests.

## REFERENCES

- Ansante, T. F., L. do Prado Ribeiro, K.U. Bicalho, J.B. Fernandes, P.C. Vieira and J.D. Vendramim (2015). Secondary metabolites from Neotropical Annonaceae: Screening, bioguided fractionation, and toxicity to *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae). *Industrial Crops and Products*, 74: 969-976.
- Asawalam, E. F., U.E. Ebere and K.C. Emeasor (2012). Effect of some plant products on the control of rice weevil *Sitophilus oryzae* (L.) Coleoptera: Curculionidae. *Journal of Medicinal Plants Research*, 6(33): 4811-4814.
- Chanda, S. and Y. Baravalia (2011). Brine shrimp cytotoxicity of *Caesalpinia pulcherrima* aerial parts, antimicrobial activity and characterisation of isolated active fractions. *Natural Product Research*, 25(20): 1955-1964.
- Dal Bello, G., S. Padin, C.L. Lastra and M. Fabrizio (2000). Laboratory evaluation of chemical-biological control of the rice weevil (*Sitophilus oryzae* L.) in stored grains. *Journal of Stored Products Research*, 37(1): 77-84.
- Dwivedi, S.C. and K. Karwasara (2003) Larvicidal activity of five plants extracts against *Culex quinquefasciatus*. *Indian J Entomol* , 65(3): 335-8.
- Khan, M. A. and M. Qaiser (2006). Halophytes of Pakistan: characteristics, distribution and potential economic usages. In: *Sabkha ecosystems* (pp. 129-153). Springer Netherlands.
- Leatemia, J. A. and M.B. Isman (2004). Efficacy of crude seed extracts of *Annona squamosa* against diamondback moth, *Plutella xylostella* L. in the greenhouse. *International Journal of Pest Management*, 50(2): 129-133.
- Langmuir, C. H., R.D. Vocke, G.N. Hanson and S.R. Hart (1978). A general mixing equation with applications to Icelandic basalts. *Earth and Planetary Science Letters*, 37(3): 380-392.
- Masimba, P. J., E. Innocent and B. Samwel (2016). Oral acute toxicity study of *annona squamosa* l. Leaves extract and fractions in albino mice. *Journal of Advanced Scientific Research*, 7(1): 38-42.
- Monzon, R. B., J.P. Alvior, L.L. Luczon, A.S. Morales and F.E. Mutuc (1994). Larvicidal potential of five Philippine plants against *Aedes aegypti* (Linnaeus) and *Culex quinquefasciatus* (Say). The Southeast Asian journal of tropical medicine and public health, 25(4): 755-759.
- Omar, K., N.M. Faraj, S.A. Malik and I.M. Al-Farhani (2012). Effect of some medicinal plants extracts and cypermethrin against Khapra Beetle (*Trogoderma granarium* Everts). *Emirates Journal of Food and Agriculture*, 24(2): 120.
- Ribeiro, L. P., J.D. Vendramim, M.S. Andrade, K.U. Bicalho, M. F. G. F. Silva, P.C. Vieira and J.B. Fernandes (2014). Tropical plant extracts as sources of grain-protectant compounds against *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). *Neotropical entomology*, 43(5): 470-482.
- Trindade, R. C. P., J. D. S. Luna, M. R. F. De Lima, P. P. Da Silva and A. E. G. Sant'Ana (2011). Actividad larvica y variacion estacional del extracto de *Annona muricata* en *Plutella xylostella* (Lepidoptera: Plutellidae). *Revista Colombiana de Entomologia*, 37(2): 223-228.
- Veni, T., T. Pushpanathan, and J. Mohanraj (2016). Ovicidal and larvicidal efficacy of *Crataeva magna* (Lour.) DC. (Family: Capparidaceae) against the *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*. *International Journal of Pure and Applied Zoology*, 4 (2): 149-154.
- Wink, M. (1993). Allelochemical properties of the raison d'etre of alkaloids. *The alkaloids: chemistry and pharmacology*, 43: 1-118.

(Accepted for publication October 2017)