

Antifeedant Activity of *Calotropis procera* Leaf Extracts Against *Cnaphalocrocis medinalis*

M. Kannan^{*1}, S. Arumugam² and G. Balasubramanian³

Received: 10 Oct 2020 | Revised accepted: 27 Dec 2020 | Published online: 05 Jan 2021
© CARAS (Centre for Advanced Research in Agricultural Sciences) 2021

ABSTRACT

Calotropis procera plants have possessed so many bioactive compounds against agricultural insects. A large number of leaves were collected shade dried and powdered then prepared petroleum ether, methanol and aqueous solution at various concentrations. The antifeedant activity was assayed using a leaf cut choice and leaf dip assay method. *C. medinalis* is one of the agricultural pests. This pest is highly infected in the paddy field and causes severe damages to paddy leaves. To apply the solvent extraction of various concentrations of *C. procera* against *C. medinalis*. From the results, after applying the solvent extraction to the reduced feeding area of the leaves. The leaf extract solution to prohibit the feeding activity on *Cnaphalocrocis medinalis* larvae methanol, petroleum ether and aqueous extracts in 1000ppm concentration showed antifeedant activity as 85.42, 73.73 and 68.70% respectively.

Key words: *Calotropis procera*, *Cnaphalocrocis medinalis*, Solvent extracts, Paddy leaves, Antifeedant activity

Antifeedant is defined as a chemical that inhibits feeding without killing the insect directly while the insect remains near the treated foliage and dies through starvation [1]. The bioactive compounds of plant origin are considered as an ecologically safe alternative and the plant extracts with complex mixtures of compounds have been widely investigated for their insecticidal, repellent, ovicidal, antifeedant and antiopposition properties [2]. The antifeedant activity of botanicals against insects has been studied in many countries. Qualification of the antifeedant effect of botanicals is of great importance in the field of insect pest management [3]. From an ecological point of view, antifeedants are very important since they never kill the target insects directly and allow them to be available to their natural balance. Higher antifeedant index normally indicates a decreased role in feeding [4]. In general, the antifeeding effect of plant extracts depend mainly on insect species, however, the plant structure-activity relationship associated with its components on insect feeding is complex and no clear trends emerge [5].

Natural antifeedants are mainly planted substances of various chemical groups, particularly effective insect antifeedants are triterpenes [6]. Sesquiterpene lactones and alkaloids, cucurbitacins, quinines and phenols [7]. The phytochemicals produced in response to insect pest attack, affect feeding and Ovi-position of insects on the plants [8]. Many authors reported that the antifeedant effect may also due to the chemical constitutes of plants such as alkaloids, flavonoids, terpenes, tannins and sterols [9]. Isolation of the bioactive ingredients responsible for such antifeedant activity

and morphological deformities could possibly facilitate development of new formulations for effective activity at lower concentrations, thereby making them economically low-cost products [10]. Antifeedant property of plant extracts brings about retardation of growth and ultimately results in the death of the insect. However, compounds which do not display antifeedant property are reported to have growth regulatory activity [11]. The antifeedant and insect growth-regulating effects of *M. azedarach* extracts are known for many insects [12]. Rice is one of the prominent crops of the world and widely cultivated in India. China, Thailand, Japan and Indonesia are the other countries which widely cultivate paddy. In India. West Bengal is the leading rice producer in India which is followed by Uttar Pradesh, Telangana, Andhra, Punjab, Orissa, Bihar, Chhattisgarh, Tamil Nadu, Karnataka, Maharashtra, Assam and Haryana [13]. Tamil Nadu has achieved the position of being the premier paddy producing a state in India. In agriculture, insects affect directly the growing part of the crop and cause severe damage resulting in revenue loss. The rice leaf folder (RLF) *Cnaphalocrocis medinalis* (guenee), is a predominant foliage feeder and one of the most destructive pests, affecting in all the rice ecosystems in Asia [14]. The larvae fold the leaves longitudinally by stitching the leaf margins and feed by scraping the green mesophyll tissue from within the folded leaves.

A number of plants were identified in several developing countries for their pesticidal activities. The plant is toxic and is one of the few plants not eaten by grazing animals. Due to its toxicity, the latex extracted from the stem has traditionally been used to make poison arrows. The latex is highly toxic to human eyes and produces sudden painless dimness of vision with photophobia. Latex of *Calotropis procera* was studied for its inflammatory effects using pedal oedema and air pouch models of inflammation in rats [15]. *Calotropis procera* belongs to the family Asclepiadaceae and

*M. Kannan
drarumugam85@gmail.com

^{1,3}P.G. and Research Dept. of Zoology, Arignar Anna Govt. Arts College, Cheyyar - 604 407, Thiruvannamalai District, Tamil Nadu, India

is a soft wooded, evergreen perennial shrub and growing widely throughout the tropical and sub-tropical regions of Asia and Africa. This plant is popularly used in their latex in traditional medicine worldwide [16]. The use of the plants, plant extracts and pure compounds isolated from natural sources has always provided a foundation for modern pharmaceutical compounds [17]. *Calotropis* species is traditionally used for the treatment of bronchitis, pain, asthma, leprosy, ulcers, piles, spleen, tumors, liver, abdomen and dyspepsia; it is also used frequently for cold, fever, diarrhea, rheumatism, indigestion, eczema and jaundice.

MATERIALS AND METHODS

Plant collection and solvent extract

Calotropis procera plant leaf were collected from Thiruvannamalai District. The plant material was identified by using the book of Medicinal Plants by S.G. Joshi. Large quantity of *C. procera* were collected and washed thoroughly in clean water, and kept in shade for air-drying. They were then dried in the laboratory and were individually ground to a fine powder. The powder material was weighed and mixed in various solvent as aqueous, methanol and petroleum ether for preparation of extracts (the ratio of 1:10 w/v). The mixture was stirred for 30 minutes and left to stand for a period of 72 hours each and then filtered. The filtered content was then subjected to evaporation in a rotary evaporator until solvents were completely evaporated to get the solidified solvent extracts. After this they were preserved in tightly corked labeled bottles and stored in a refrigerator until further use. 10 mg of plant extracts were dissolved in 1 ml acetone and then dissolved in 10 ml distilled water to get 1000 ppm as stock solution. Concentrations of 200, 400, 600, 800 and 1000 ppm were prepared from stock solution.

Antifeedant activity

Antifeedant activity was assayed using a leaf cut choice - test [18], [19] and 'leaf dip assay' method [20]. The fresh ADT-43 paddy leaves measuring 8cm length were dipped in the test tube at various concentration (200, 400, 600, 800 and 1000 ppm) and after 30 minutes shade drying the leaves were arranged in Petri dish (15cm dia) plant extract of *C. procera* against the larvae of *C. medinalis* lined with a moist filter paper disc. The control leaves were treated with distilled water alone. The IV and V instar larvae were allowed to feed on the treated leaf discs only once and the experiment was continued for 48h. The test was carried out in glass Petri dish (15cm via) closed with another Petri dish. At each tested concentration three replicates of two individually kept larvae were tested. The larvae were allowed to feed for 48h and unfed areas of the left-over leaf discs were measured by graph method and percent feeding and antifeedant activity calculated based on the formula of [21].

$$\text{Percent feeding} = \frac{\text{Area given for feeding} - \text{Corrected area left}}{\text{Area given for feeding}} \times 100$$

$$\text{Antifeedant activity (\%)} = \frac{\text{Left disc consumed by the larvae in control} - \text{Left disc consumed by the larvae in treated}}{\text{Left disc consumed by the larvae in control} + \text{Left disc consumed by the larvae in treated}} \times 100$$

Statistical analysis

All data were subjected to analysis of variance (ANOVA) implemented in SPSS package and the means were separated using Duncan's multiple range test [22]. All the observed effects were considered for statistical significance at $P < 0.05$.

RESULTS AND DISCUSSION

C. medinalis treated with three solvent extracts of *C. procera* leaves were tested the properties of antifeedant activity and presented in (Table 1, Fig 1). Highest antifeedant activity of 85.42% was observed in methanol extract at 1000ppm concentration and lowest effect of 34.23% was observed in aqueous extract at 200ppm concentration in methanol extract showed good antifeedant activity with increasing concentrations as 45.24% (200ppm), 50.98 (400ppm), 59.61% (600ppm) 77.30% (800ppm) and 85.42% (1000ppm). Next to methanol extract, petroleum ether possesses antifeedant properties as 38.16% (200ppm) 42.12% (400ppm), 51.10% (600ppm), 66.16% (800ppm) and 73.73% (1000ppm). Aqueous extract were observed lowest antifeedant activity with comparing of other two extracts, as 34.23% in 200ppm, 43.66% in 400ppm, 53.56% in 600ppm, 65.74% in 800ppm and 68.70% in 1000ppm. Maximum antifeedant activities of 85.42, 73.73 and 68.70% were found in methanol > petroleum ether > aqueous respectively at 1000ppm concentration. The percent leaf area feeding was decreased with increasing concentrations. Percent feeding was ranging from 82.66% (control) to 6.50(1000ppm) in methanol extract 31.16% in 200ppm, 26.83% in 400ppm, 20.91% in 600ppm, 10.58% in 800ppm and 6.50% in 1000ppm were observed feeding activity in methanol extract. Next to methanol extract, petroleum ether and aqueous extracts almost same feeding activity in all concentrations at 200, 400,600,800 and 1000ppm as 37.00, 33.66, 26.75, 16.83, 12.5 and 40.50, 32.41, 25.00, 17.08 and 15.33% in respectively (Fig 2). All the plant extracts were found to protect leaf area when compared to control.

Table 1 Percent antifeedant action of leaf extracts of *Calotropis procera* on fourth instar larvae *C. medinalis*

Leaves extracts	Concentration (ppm)	Leaf area consumed (cm ²)	Percent feeding	Antifeedant index
Petroleum Ether	Control	9.92±0.16a	82.66	-
	200	4.44±0.02b	37.00	38.16
	400	4.04±0.15c	33.66	42.12
	600	3.21±0.01d	26.75	51.10
	800	2.20±0.03e	16.83	66.16
Methanol	1000	1.50±0.01f	12.5	73.73
	Control	9.92±0.16a	82.66	-
	200	3.74±0.02b	31.16	45.24
	400	3.22±0.01c	26.83	50.98
	600	2.51±0.02d	20.91	59.61
Aqueous	800	1.27±0.02e	10.58	77.30
	1000	0.78±0.02f	6.50	85.42
	Control	9.92±0.16a	82.66	-
	200	4.86±0.01b	40.50	34.23
	400	3.89±0.02c	32.41	43.66
	600	3.00±0.02d	25.00	53.56
	800	2.05±0.18e	17.08	65.74
	1000	1.84±0.01f	15.33	68.70

Different alphabets notified significant level at 0.05% DMRT

Plant substances acting as antifeedants are found in all the compound groups of secondary plant metabolism. Several plant secondary metabolites are known antifeedants and they possess various chemicals such as triterpenes, sesquiterpenes, lactones and alkaloids, cucurbitacines, quinines and phenols. Some plant families include numerous species containing bioactive substances, amongst which are volatile oils especially terpenes are reported to contain antifeedant properties against various lepidopteran agricultural pests [23]. Antifeedants offer first line of crop protection against notorious insects. Any substances that reduces food consumption by an insect can be considered as an antifeedant or feeding deterrent [24]. The most antifeedents, the modes-of-action are directed at the taste cells. A typical gustatory sensillum in an insect contains receptors selective for deterrents and others for stimulants (such as sugars and amino acids). Although most antifeedent likely act by stimulating a deterrent receptor, that in turn sends a signal (do not feed) to

the feeding centre in the insects central nervous system, some antifeedents are thought to block or otherwise interfere with the perception of feeding stimulant [25]. The antifeedant and oviposition deferent activities were more prominent than the knock down effects [26]. In case of leaf hoppers and plant hoppers, disruption of growth resulted in reduction in size and weight of insects after feeding on plants treated with crude or commercial neem formulations. Consequently, the proportion of nymphs becoming adults was also affected [27]. However, in lepidopterous insects larval pupal intermediaries were observed. Aqueous extracts of *Calotropis procera* and *Datura stromonium* display about 90% feeding against *H. armigera* [28]. This study displays 69% feeding against *Cnaphalocrocis medinalis* larvae with aqueous extract. Leaf extracts of *Calotropis procera* against *Musca domestica* to indicate the antifeedant properties which may be due to the different compounds present in the extract possessing different bioactivities [29].

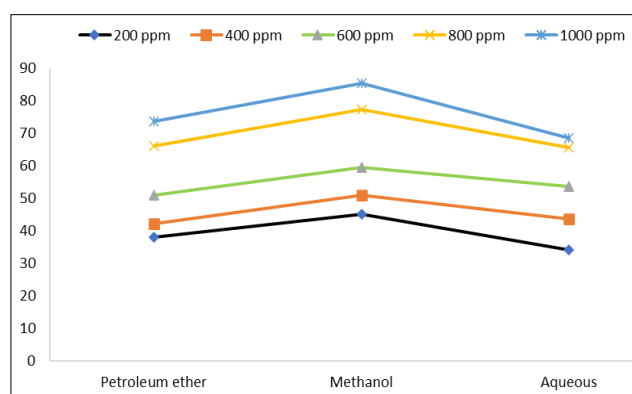


Fig 1 Percent antifeedant effect of *Calotropis procera*

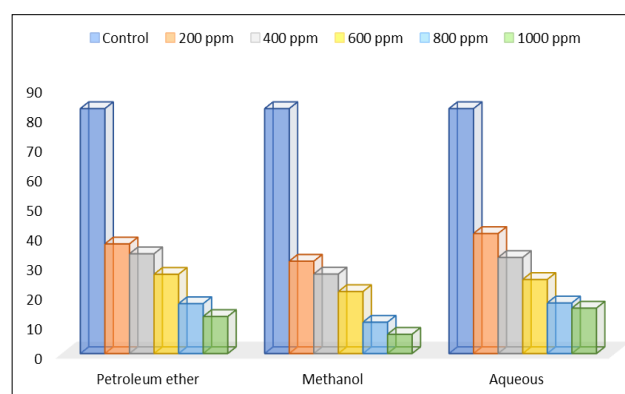


Fig 2 Percent feeding activity of *Calotropis procera*

In this study also evaluated the various solvent extract of *C. procera* leaves against *C. medinalis* pest. The percentage of feeding was decreased with increasing concentrations, in the same time antifeedant activity increased with increasing concentrations [30]. Methanol extracts showed high antifeedant deferent at 1000ppm (85.42%) concentration, on the other two solvent extracts has contain moderate activity followed by petroleum ether (73.73%) and aqueous (68.70%) extract [31]. Antifeedant and larvicidal activity of acetone, chloroform, ethyl acetate, hexane and methanol peel, leaf and flower extracts of *Citrus sinensis*, *Ocimum canum*, *Ocimum sanctum* and *Rhinacanthus nasutus* against lepidopterans suggest their potential as an ideal ecofriendly approach for the control for the agricultural pests [32]. Whereas the observation of this study reveals *C. procera* plants have been performed good antifeedant properties against *C. medinalis* larvae.

Combination of an antifeedant with a physiological toxin is another choice to develop a sustainable pest management strategy based on plant products [33]. The toxicity effect may be attributed to the secondary metabolites

[34], which have been isolated from various plant parts. These tend to affect insects in several ways such as disrupting major metabolic pathways and causing rapid death, acting as deterrents, photostimulants or antifeedants or modifying oviposition [35]. The methanol extracts of *C. procera* leaves applied very effective control of *C. medinalis* on the paddy field.

CONCLUSIONS

All the plants are possessing many bioactive compounds. These bioactive materials are acts as antifeedant deterrent and larvicidal activities. Chemical pesticides are controlling of the pest infection but reduced the soil quality. Hence, now a days the alternative method of plant materials to be used controlling of pest and maintained soil quality. The leaf extracts of *C. procera* prohibit the feeding area on paddy leaves for *C. medinalis* larvae. In furthermore, this plant extracts are controlling of *C. medinalis* as well as maintained toxic free environment.

LITERATURE CITED

- Jeyasankar, Premalatha, Elumalai. 2014. Antifeedant and insecticidal activities of selected plant extracts against *Epilachna* beetle, *Henosepilachna vigintioctopunctata* (Coleoptera: Coccinellidae). *Advances in Entomology* 2(1): 14-19.
- Isman MB, Wan AJ, Passreiter CM. 2001. Insecticidal activity of essential oils to tobacco cutworm, *Spodoptera litura*. *Fitoterapia* 72: 65-68.
- Arivoli S, Samuel T. 2013. Antifeedant activity, developmental indices and morphogenetic variations of plant extracts against *Spodoptera litura*, *Journal of Entomology and Zoological Studies* 1(4): 87-96.
- Pavunraj M, Basker K, Ignacimuthu S. 2012. Efficacy of *Melochiacorchorifolia* L. (Sterculiaceae) on feeding behavior of four Lepidopteran pests. *International Journal of Agricultural Research* 7(2): 58-68.

5. Bruno MFP, Magio AM, Roselli S, Simmonds MSJ. 2002. Antifeedant activity of neoclerodane diterpenoids from *Teucrium arduini*. *Biochemical Syst. Ecol.* 30: 595-599.
6. Beek VTA, Groot AED (1986) Terpenoidantifeedants. Part I. An overview of terpenoidantifeedants of natural origin. *Recueilles Travaux Chimiques des pays-Bass* 105(12): 513-527.
7. Norris DM. 1986. Anti-feeding compounds in chemistry of plant protection (1), sterol Biosynthesis Inhibitors and Anti-Feeding compounds. Academic-Verlag, Berlin. pp 97.
8. Ramya S, Jayakumararaj R. 2009. Antifeedent activity of selected ethno-botanicals used by tribals of vattal hills on *Helicoverpa armigera*. *Jr. of Pharmacy Research* 2(8): 1414-1418.
9. Salama HS, Dimitry NZ, Salem SA. 1971. On the host preference and biology of the cotton leafworm, *Spodoptera littoralis* (Boisd.). *Z. Ang. Ent.* 67: 261-266.
10. Arivoli S, Tennyson S. 2012. Antifeedant activity of plant extracts against *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae) *American- Eurasian Journal of Agriculture & Environmental Sciences* 12(6): 764-768.
11. Kraus W, Baumann S, Bokel M, Keller U, Klenk A, Klingele M, Pohnl H, Schwinger M. 1987. Control of insect feeding and development by constituents of *Melia azadirach* and *Azadirachta indica*. Proceedings of 3rd International Neem Conference, Nairobi, Kenya. pp 375-385.
12. Saxena RC, Epino PB, Cheng WT, Puma BC. 1984. Neem, chinaberry and custard apple: antifeedant and insecticidal effects of seed oils on leaf hopper and plant hopper pests of rice. *In:* (Eds) H. Schmutterer, K.R.S. Ascher. Proceedings of Second International Neem Conference. Rauischholzhausen, Germany. pp 403-312.
13. CIKS. 206. Centre for Indian Knowledge Systems. Package of organic practices from Tamil Nadu.
14. Leo SJ. 2010. Occurrence of rice leaf roller in China and its identification and prevention. *Plant Diseases and Pests* 1: 13-18.
15. Quazi S, Mathur K, Arora. 2013. *Calotropis procera*: An overview of its phytochemistry and pharmacology. *Indian Journal of Drugs* 1(2): 63-69.
16. Goyal M, Mathur R. 2011. Antimicrobial potential and phytochemical analysis of plant extracts of *Calotropis procera*. *International Journal of Drug Discovery and Herbal Research* 13: 138-143.
17. Murti Y, Yogi B, Pathak D. 2012. Pharmacognostic standardization of leaves of *Calotropis procera* (Ait.) R.Br. (*Asclepiadaceae*). *International Jr. Ayurveda Research* 1: 14-17.
18. Isman B, Koul O, Luczynski A, Kaminski J. 1990. Insecticidal and antifeedant bioactivities of neem oils and their relationship to Azadirachtin content. *Journal of Agricultural and Food Chemistry* 38: 1407-1411.
19. Khan ZR, Abenes MLP, Fernandez NJ. 1996. Suitability of graminaceous weed species as host plants for rice leaf folders, *Cnaphalocrocis medinalis* and *Marasmia patnalis*. *Crop Protection* 15: 121-127.
20. Zhuang J, Fu J, Su Q, Li J, Zhan Z. 2009. The regional diversity of resistance of tea green leafhopper, *Empoasca vitis* (Gothé), to insecticides in Fujian Province. *Journal of Tea Science* 29: 154-158.
21. Mondal M. 2018. Traditional methods for economical Paddy cultivation, Agripedia.
22. Duncan DB. 1955. Multiple range and multiple F tests. *Biometrics* 11: 1-42.
23. Nathan S, Dency S, Narmadha G, Suganya, Chung PG. 2006. Efficacy of *Melia azedarach* L. extract on the malarial vector *Anopheles stephensi* Liston (Diptera: Culicidae). *Bioresource Technology* 97: 1316-1323.
24. Isman MB. 2002. Insect antifeedants. *Pesticide Outlook* 13: 152-157.
25. Sharaby A. 1988. Evaluation of some myrtaceae plant leaves as protectants against the infestation by *Sitophilus oryzae* L. and *Sitophilus granaries* L. *Insect Science Appl.* 9: 465-468.
26. Jena M. 2005. Integrated pest management with botanical pesticides in rice with emphasis on neem products. *Oryza* 42: 124-128.
27. Dodia DA, Patel IS, Pathak AR. 1998. Antifeedant properties of some in-digenous plant extracts against larvae of *Helicoverpa armigera*. *Pestology* 19: 21-22.
28. Begum N, Sharma B, Pandey RS. 2011. Evaluation of insecticidal efficacy of *Calotropis procera* and *Annona squamosa* ethanol extracts against *Musca domestica*. *Jr. Biofertil Biopesticide* 1(1): 2-6.
29. Juan A, Sans A, Rabia M. 2000. Antifeedant activity of fruit and seed extracts of *Melia azedarach* and *Azadirachta indica* on larvae of *Sesamia nonagrioides*. *Phytoparasitica* 28: 311-319.
30. Rahman M. 2007. Antifeedant and toxic activity of some plant extracts against larvae of cotton leafworm *Spodoptera littoralis* (Lepidoptera: Noctuidae). *Pakistan Journal of Biological Sciences* 10: 4467-4472.
31. Schmidt GH, Rembold AA, Ahmed I, Breuer M. 1998. Effect of *Melia azedarach* fruit extract on juvenile hormone titer and protein content in the Haemolymph of two species of noctuid Lepidopteran larvae (Insecta: Lepidoptera: Noctuidae). *Phytoparasitica* 26: 283-292.
32. Sehoon. 2006. Effects of *Melia azedarach* L. extract on the teak defoliator *Hyblaea puera* Cramer (Lepidoptera: Hyblaeidae). *Crop Production* 25(3): 287-290.
33. Koul O, Walia S. 2009. Comparing impacts of plant extracts and pure allelochemicals and implications for pest control. *Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources* 4: 049.
34. Dales MJ. 1996. A review of plant materials used for controlling insect's pest of stored products. NRI bulletin NO. 65. Natural Resources Institute Chatham, UK. pp 84.
35. Jilani G. 1992. Botanicals and pheromones in IPM P. *In:* (Eds) Baloch U. K. Integrated Pest Management in food grain. FAO/PARC, Islamabad, Pakistan. pp 50-57.