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Original Article

Larvicidal Activity Of Croton Sparciflorus Morong (Euphorbiaceae) Leaf Extracts Against Three Vector Mosquitoes

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ABSTRACT

The present investigation was undertaken to assess the larvicidal potential of the leaf extract from the medicinal plant *Croton sparciflorus* Morong (*C. sparciflorus*) against the medically important mosquito vectors, *Aedes aegypti, Anopheles stephensi* and *Culex quinquefasciatus*. Twenty five III instar larvae of three mosquitoes are exposed to different concentration (50-250) and were assayed in the laboratory by using the protocol of WHO 2005; The 24 h LC50 values of the C. The *Sparciflorus* leaf extract was determined following Probit analysis. Among the plant extract tested against *Aedes aegypti, Anopheles stephensi* and *Culex quinquefasciatus* the pronounced lethal activity was recorded against C. sparciflorus extract in the experimental larvae of *A. Stephen*s with an LC_{50} and LC_{90} value of 28.88 and 65.35 ppm. It is accomplished that the highest larvicidal activity against A. stephensi was obtained with ethyl acetate extract of *C. sparciflorus*. The present investigation revealed that the possible utilization of *C. sparciflorus* to control the mosquito menace to a greater extent. Thus, paving the way for further exploration of identification and isolation of active principles present on the selected plant.

Keywords:

Croton sparciflorus, *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefaciatus*, Larvicidal activity.l .

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INTRODUCTION

Mosquitoes represent a significant threat to human health because of their ability to vector pathogens that cause diseases that afflict millions of people worldwide (WHO 2010). Several species belonging to genera *Aedes, Anopheles* and *Culex* are vectors for the pathogen of various diseases like dengue fever, dengue hemorrhagic fever, malaria, Japanese encephalitis and filariasis (Borah et al., 2010; Rahuman 2009; Samuel 2010). *Aedes aegypti* is known to carry dengue and yellow fever; malaria is carried by Anopheles *stephensi*; and filarial disease by *Culex*. The dengue fever incidence has increased fourfold since 1970 and nearly half the world's population is now at risk. In 1990, almost 30% of the world population, 1.5 billion people, lived in regions where the estimated risk of dengue transmission was greater than 50% (Hales et al., 2002). An outbreak of Chikungunya virus disease emerged in the southwest Indian Ocean islands in 2005, spread out to India, and resulted in an ongoing outbreak that has involved >1. 5 million patients, including travelers who have visited these areas (Taubitz et al., 2007). Anopheles stephensi are major malaria vectors in India. With an annual incidence of 300-500 million, malaria is still one of the most important communicable diseases. Currently, about 40% of the world's population live in areas where malaria is endemic (Wernsdorfer and Wernsdorfer, 2003). *Culex quinquefasciatus*, a vector of lymphatic filariasis, is widely distributed in tropical zones with around 120 million people infected worldwide and 44 million people having common chronic manifestation (Bernhard et al., 2003).The present proliferation of these diseases is not only due to higher number of breeding places in urban agglomeration, but also due to increasing resistance of mosquitoes to current commercial insecticides such asorganochlorides, organophosphates,

carbamates and also to biological insecticides (Goettel *et al.*, 1992; Das and Amalraj, 1997; Yadav *et a*l., 1997).

Mosquito control has been becoming increasingly difficult because of the indiscriminate uses of synthetic chemical insecticides which have an adverse impact on the environment and disturb ecological balance. Majority of the chemical pesticides are harmful to man and animal, some of which are not easily degradable and spreading toxic effects. The increased use of these insecticides may enter into food chain, and thereby, the liver, kidney, etc., may be irreversibly damaged. They even result in mutation of genes and these changes become prominent only after a few generations (Ghosh *et al.*, 1991). These problems have highlighted the need for the development of new strategies for selective mosquito control. Phytochemical are advantageous due to their eco-safety, target-specificity, non development of resistance, reduced number of application, higher acceptability, and suitability for rural areas. Botanicals can be used as alternative to synthetic insecticides or along with other insecticides under integrated vector control programs. The plant product of phytochemical, which is used as insecticides for killing larvae or adult mosquito bites. Phytochemicals obtained from the whole plant or specific part of the plant by the extraction with different types of solvent such as aqueous, methanol, chloroform, benzene, acetone, etc., depending on the polarity of the phytochemical. Some phytochemicals act as toxicant (insecticide) both against adult as well as larval stages of mosquitoes, while others interfere with growth and growth inhibitor or with reproduction or produce an olfactory stimulus, thus acting as repellent or attractant (Markouk *et al.*, 2001).

Many studies on plant extract against mosquito larvae have been conducted in the region of the World. Plants may be a source of alternative agents for control of mosquitoes because they are rich in bioactive chemicals, are active against a limited number of species including specific target insects, and are biodegradable. They are potentially suitable for use in integrated pest management programs (Alkofahi *et al.*, 1989, Dharmshaktu *et al.*, 1987, Green *et al.*, 1991). Croton is extensive flowering plants genus in the spurge family, Euphorbiaceae established by Carl Linnaeus in 1737. The common names for this genus are rushfoil and *croton,* but the latter also refers to *Codiaeum variegatum* (Gledhill 2008). It has recently been shown in Kenya that *Croton* nuts, such as those *C. megalocarpus*, are a more economical resource of biofuel than Jatropha (Milich and Lenard 2009). It is used as a potent Hypotensive agent (Radcliffe-Smith 1986) and for the treatment of a variety of ailments like fever, inflammation, hypertension (Dubey *et al.*,1969), and it causes sharp fall in blood pressure (Mandal *et al.*, 2004). Different extracts of this plant show larvicidal activity (Bhakuni and Jain 1981).

Materials And Methods

Plant collection

The leaves of *C. sparciflorus* are collected from Velankanni costal area, Nagai district, Tamil Nadu, South India (Fig. 1). The plant is authenticated by a plant taxonomist from the Department of Botany, Annamalai University. Voucher herbarium specimens have been deposited in the laboratory of Zoology, Annamalai University and Chidambaram.

Fig-1Collection of *C. sparciflorus* from Velankanni costal area

Preparation of the Extract

The dried leaf (100g) was powdered mechanically using commercial electrical stainless steel blender and extracted sequentially with ethyl acetate, hexane, dichloromethane and diethyl ether (500 ml, Ranchem), in a Soxhlet apparatus separately until exhaustion. The extract was concentrated under reduced pressure 22–26 mmHg at 45°C by 'Rotavapour' and the residue obtained was stored at 4°C.

Test Mosquitoes

All tests were carried out against laboratory reared vector mosquitoes viz., *Aedes aegypti (Ae. Egyptian), Anopheles stephensi (An. stephensi)* and *Culex quinquefasciatus (Cx. quinquefasciatus)* free of exposure to insecticides and pathogens, Cyclic generations of vector mosquitoes were maintained at 25-30 OC and 80-90 % relative humidity in the insectariums. Larvae were fed on larval food (powdered dog biscuit and yeast in the ratio of 3:1) and adult mosquitoes were periodically blood-fed to restrained albino mice for egg production.

Larvicidal Activity

Standard WHO protocol with slight modifications was adopted for the study (WHO 1996). From the stock solution, the concentration of 50, 100, 150, 200 and 250 ppm was prepared. Early third instar larvae were introduced in 250 ml plastic cups containing 200 ml of water with each concentration. A control was prepared by the addition of acetone to water. Mortality was recorded after 24 hours. For each experiment, five replicates were maintained at a time. The observed percentage mortality was corrected by Abbott's Formula (Abbott 1925).

Statistical Analysis

Statistical analysis of the study records was carried out with the help of SPSS to find the LC50, LC90, Standard Deviation (±SD), Mortality, Degree of freedom and Regression equations.

Results And Discussion

Results of the larvicidal activity of the ethyl acetate, hexane, dichloromethane and diethyl ether leaf extract of *C. sparciflorus* against *Ae. aegypti, An. stephensi* and *Cx. quinquefasciatus* reported in the present study showed the mosquitocidal properties in the plant signifying their use in mosquito population control (Table 1, 2, 3 and 4). The leaf extracts of *C. sparciflorus* showed larval mortality. *An. stephensi* was more vulnerable followed by *Ae. aegypti* and *Cx. quinquefasciatus*. The ethyl acetate extract of *C. sparciflorus* exhibited the maximum larvicidal activity with LC_{50} and LC_{90} value of 28.88 and 65.35 p.m. against the larvae of *An. stephensi* (Table 1). The screening of local medicinal plants for mosquito larvicidal activity may eventually lead to their use in natural product-based mosquito abatement practices. The results of the present study are equivalent with previous reports. They reported that the Dhanasekaran *et al.*, have that the LC_{50} of ethanol crude extracts of selected indigenous medicinal plants were 82.86ppm, 89.45ppm, 109.37ppm 109.87ppm and 172.31 respectively, against the malarial vector, An. stephensi, dengue vector Ae. aegypti, Japanese Encephalitis vector, *Culex tritaeniorhynchus*. Kaliyamoorthy krishnappa *et al.*, (a) has ethanol extracts of Gliricidia sepium showed larvicidal showed LC_{50} and LC_{90} value of 121.79ppm and 231.98 respectively, against the malarial vector, An. Stephens. Elangovan *et al.*, (a) have leaf extracts of *Corchorus capsularis* the against common malarial vector, *Anopheles stephensi* (LC_{50} and LC_{90}values of 197.34, 205.48, 176.19 and 358.59, 363.42, 334.56 ppm), against dengue vector Ae. aegypti (LC_{50} and LC_{90} values of 222.45, 190.52, 182.06 and 383.06, 354.84, 306.81 ppm), respectively. Krishnappa et al., (b) have hexane, benzene, chloroform and methanol extracts of Adansonia digitata against III instar larvae of medically important human malaria vector mosquito Anopheles stephensi (LC_{50} and LC_{90} values of 111.32, 97.13, 88.55, 78.18 and 178.63, 176.19, 168.14, 155.42 mg/l) respectively. Elumalai (a) et al., have various extracts of *Gymnema Sylvestre* showed LC_{50} values of 34.756, 31.351 and 28.577 respectively, against the Japanese Encephalitis vector, *Culex tritaeniorhynchus*. Elumalai et al.,(b) have that larvicidal, ovicidal and pupicidal activity of Eranthemum roseum against malarial vector mosquito, *Anopheles stephensi* (LC_{50} and LC_{90} values of 121.65, 139.86 ppm and 237.38, 255.51 ppm), respectively. Elumalai *et al.*, (c) have that mosquitocidal activity of Abrus precatorius against chickungunya vector, *Ae. aegypti* (LC_{50} and LC_{90} values of 264.57ppm and 500.76ppm), against the Japanese Encephalitis vector, *Culex tritaeniorhynchus* (LC_{50} and LC_{90} values of 257.73ppm and 496.94ppm),

respectively. Baluselvakumar *et al.*, have plant extracts of Oxystelma esculentum against Anopheles stephensi (LC_{50} and LC_{90} values of 75.46, 68.55, 98.47, 88.24, 63.84 and 140.66, 130.65, 184.10, 169.36 and 122.48 ppm,) respectively. Mullai and Jabanesan have leaf extracts of *C. colocynthis* and Cucurbita maxima showed LC_{50} values of 47.58, 66.92 and 118.74 ppm and 75.91, 117.73 and 171.64 ppm, respectively, against *Cx. quinquefasciatus* larvae. Krishnappa *et al.*, (c) have leaf extracts of Cissus quadrangularis and Combretum ovalifolium showed LC_{50} values of 56.42, 46.37, 47.55, 37.48ppm and 95.47, 85.37, 86.46, 74.53ppm respectively, against *Anopheles stephensi.* Elangovan *et al.*, (b) have that the larvicidal and ovicidal activity of Exacum pedunculatum against the common malarial vector, *Anopheles stephensi* (LC_{50} and LC_{90} values of 127.45, 121.39, 151.96 and 121.24). Karunamoorthi and ilango have that the LC_{50} and LC_{90} of a methanol leaf extract of *Croton macrostachyus* (*C. macrostachyus*) were 89.25 and 224.98 ppm, respectively against late third instar larvae of the malaria vector, *Anopheles arabiensis (A. arabiensis).*

Table 1: Larvicidal activity of Ethyl acetate extracts *C. sparciflorus against A. aegypti, A. stephensi, and Cx. quinquefasciatus.*

Concentration tested	%martality± SD	LC_{50} (ppm)	95% confidence limit		LC_{90} (ppm)	95% confidence limit		*df*	Regression
			LCL	UCL		LCL	UCL		
A. aegypti									
50 ppm	36±2.387								
100 ppm	56±1.516	34.02	28.54	38.57	74.57	68.78	82.26	3	Y=-3.213x+9.897
150 ppm	76±2.280								
200 ppm	92±2.345								
250 ppm	100±0.00								
A. Stephens									
50 ppm	20±1.303								
100 ppm	36±1.414	28.88	1.23	41.35	65.35	51.79	103.66	3	Y=-7.422x+15.93
150 ppm	56±2.915								
200 ppm	72±2.387								
250 ppm	84 ±1.923								
Cx. quinquefasciatus									
50 ppm	36±3.130								
100 ppm	52±2.607	36.22	21.22	46.05	79.89	67.68	103.97	3	Y=-3.340x+10.02
150 ppm	72±3.633								
200 ppm	88±2.792								
250 ppm	100±0.00								

LC50= Lethal concentration that kills 50% of the exposed parasite, LC90= Lethal concentration that kills 90% of the exposed parasite. LCL- Lower Confident Limit, UCL- Upper Confident Limit, PPM- Parts Per Million, SD- Standard Deviation, DF- Degree of Freedom.

Table 2: Larvicidal activity of Hexane extracts *C. sparciflorus against A. aegypti, A. stephensi, and Cx. quinquefasciatus.*

Concentration tested	%martality± SD	LC_{50} (ppm)	95% confidence limit		LC_{90} (ppm)	95% confidence limit		*df*	Regression
			LCL	UCL		LCL	UCL		
A. aegypti									
50 ppm	20±1.303								
100 ppm	36±1.414	55.28	49.88	60.42	110.70	101.08	124.29	3	Y= 2.594x+0.651
150 ppm	56±2.915								
200 ppm	72±2.387								
250 ppm	84 ±1.923								
A.stephensi									
50 ppm	24±2.167								
100 ppm	06 ±3.507	63.47	56.23	71.14	143.91	125.56	174.35	3	Y= 1.785x+1.879
150 ppm	48±3.209								
200 ppm	60±4.301								
250 ppm	72±2.949								
Cx. quinquefasciatus									
50 ppm	12±2.121								
100 ppm	08±2.966	69.59	64.08	75.71	129.34	116.91	147.63	3	Y= 2.489x+0.511
150 ppm	44±2.774								
200 ppm	60±3.781								
250 ppm	72±2.683								

LC_{50}= Lethal concentration that kills 50% of the exposed parasite, LC_{90}= Lethal concentration that kills 90% of the exposed parasite. LCL- Lower Confident Limit, UCL- Upper Confident Limit, PPM- Parts Per Million, SD- Standard Deviation, DF- Degree of Freedom.

Table 3: larvicidal activity of dichloromethane extracts *C. sparciflorus against A. aegypti, A. stephensi, and Cx. quinquefasciatus*

Concentration tested	%martality± SD	LC_{50} (ppm)	95% confidance limit		LC_{90} (ppm)	95% confidance limit		*df*	Regression
			LCL	UCL		LCL	UCL		
A.aegypti									
50 ppm	32±1.095								
100 ppm	48±1.673	41.58	18.23	55.47	88.44	71.20	137.20	3	Y=-3.402x+9.978
150 ppm	64±1.816								
200 ppm	80±1.949								
250 ppm	100±0.00								
A.stephensi									
50 ppm	40±2.863								
100 ppm	56±2.302	32.32	26.33	37.18	74.95	68.92	83.02	3	Y=-3.356x+10.16
150 ppm	76±2.683								
200 ppm	92±2.880								
250 ppm	100±0.00								
Cx. quinquefasciatus									
50 ppm	24±2.509								
100 ppm	40±3.209	54.77	48.68	60.49	117.47	105.95	134.50	3	Y= 2.279x+1.197
150 ppm	52±3.033								
200 ppm	68±3.286								
250 ppm	84±2.509								

LC_{50}= Lethal concentration that kills 50% of the exposed parasite, LC_{90}= Lethal concentration that kills 90% of the exposed parasite. LCL- Lower Confident Limit, UCL- Upper Confident Limit, PPM- Parts Per Million, SD- Standard Deviation, DF- Degree of Freedom.

Table 4: Larvicidal activity of Diethyl ether extracts *C. sparciflorus against A. aegypti, A. stephensi, and Cx. quinquefasciatus.*

Concentration tested	%Martality± SD	LC_{50} (ppm)	95% confidence limit		LC_{90} (ppm)	95% confidence limit		*df*	Regression
			LCL	UCL		LCL	UCL		
A. aegypti									
50 ppm	28±1.643								
100 ppm	44±1.483	47.22	40.89	52.73	106.06	96.31	120.09	3	Y=2.426x+1.113
150 ppm	60±1.140								
200 ppm	76±1.341								
250 ppm	88±2.073								
A.stephensi									
50 ppm	32±3.130								
100 ppm	04±3.781	45.04	38.65	50.52	102.62	93.30	115.97	3	Y=2.483x+1.085
150 ppm	60±3.271								
200 ppm	76±3.898								
250 ppm	92±2.280								
Cx. quinquefasciatus									
50 ppm	20±2.302								
100 ppm	36±3.240	60.55	54.70	66.44	123.65	111.35	141.94	3	Y=2.274x+1.084
150 ppm	48±2.607								
200 ppm	64±3.847								
250 ppm	80±3.082								

LC_{50}= Lethal concentration that kills 50% of the exposed parasite, LC_{90}= Lethal concentration that kills 90% of the exposed parasite. LCL- Lower Confident Limit, UCL- Upper Confident Limit, PPM- Parts Per Million, SD- Standard Deviation, DF- Degree of Freedom.

Conclusion

It may conclude that natural products as extracts from parts of plants of insecticidal and medicinal values have higher efficiency in reducing the mosquito menace due to their larvicidal toxicity. Further studies on the isolation and purification of compounds followed by in-depth laboratory and field bioassays are needed as the present study shows that there is scope to use C. sparciflorus leaf extracts to control the immature stages of vector mosquitoes.

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References:

1.WHO (2010). Malaria fact sheets No. 94. WHO Report. Geneva: WHO media center.
2.Borah R, Kalita MC, Kar A, Talukdar AK (2010). Larvicidal efficacy of Toddalia asiatica (Linn.) Lam against two mosquito vectors Aedes aegypti and Culex quinquefasciatus. African J Biotechnol; 9 (16): 2527-2530.
3.Rahuman AA, Bagavan A, Kamaraj C, Saravanan E, Zahir AA, Elango G (2009). Efficacy of larvicidal botanical extracts against Culex quinquefasciatus Say (Diptera: Culicidae). Parasitol Res; 104: 1365-1372.
4.Samuel T (2010). Studies on the mosquitocidal activity of Ageratum houstonianum Mill. (Asteraceae) against vector mosquitoes [PhD Thesis]. Tamilnadu, India: University of Madras.
5.Hales S, Wet ND, Maindonald J, Woodward A (2002). Potential effect of population and climate changes on global distribution of dengue fever: an empirical model. Lancet; 360: 830-4.
6.Taubitz W, Cramer JP, Kapaun A, Pfeffer M, Drosten C, Dobler G, et al., (2007). Chikungunya fever in travelers: clinical presentation and course. Clin Infect Dis; 45: 508.
7.Werndorfer G, Wernsdorfer WH (2003). Malaria at the turn of the 2nd to the 3rd millennium. Wien Klin Wochenschr; 115: 2-9.
8.Bernhard L, Bernhard P and Magnussen P (2003). Management of patient with lymphedema caused by fibrosis in northeastern Tanzania: alternative approaches. Physiothe. 89(12): 743-749.
9.Goettel, M.S., Toohey, M.K., Ram, R.C., Pillae, J.S., (1992). Field evaluation of Bti against Aedes vigilax and Culex sitiens in Fiji. Mosquito News 42, 277–278.
10.Das, P.K., Amalraj, D., (1997). Biological control of malaria vectors. Indian Journal of Medical Research 106, 174–197.
11.Yadav, R.S., Sharma, V.P., Upadhyay, A.K., (1997). Field trial of Bacillus sphaericus B-101 (serotype H5a, 5b) against filariasis and Japanese Encephalitis vectors in India. Journal of American Mosquito Control Association 13, 158–163.
12.Ghosh GH (1991). Biopesticide and Integrated Pest Management. A.P. H. Publishing Corporation, New Delhi; 145-146.
13.Markouk M, Bekkouche K, Larhsini M, Bousaid H, Lazrek HB, Jana M (2001). Evaluation of some Moroccan Medicinal plant extracts for larvicidal activity. J Ethnopharmacol; 73: 293-297.
14.Alkofahi A, Rupprecht JK, Anderson JE, Mclaughlin JL, Mikolajczak KL, Scott BA (1989). Search for new pesticides from higher plants. In: Arnason JT, Philogene BJR, Morand P (Eds) Insecticides of Plant Origin. In: ACS Sym. Ser, 387. Am Chem Soc, Washington, DC; 25–43.
15.Dharmshaktu NS, Prabhakaran PK, Menon PK (1987). Laboratory study of the mosquito larvicidal properties of leaf and seed extract of plant Agava Americana. J Trop Med Hyg; 90: 79–82.
16.Green MM, Singer JM, Sutherland DJ, Hibbon CR (1991). Larvicidal activity of Tagetes minuta (Marigold) towards Aedes aegypti. J Am Mosq Control Assoc; 7: 282–286.
17.Gledhill D. (2008). The Names of Plants (4 Ed.). Cambridge University Press. p. 126. ISBN 978-0-521-86645-3.
18.Milich, Lenard (2009). "Environmental Comparisons of Croton Megalocarpus vs. Other Tropical Feedstocks". Africa Biofuel. Retrieved -10-11.
19.Radcliffe-Smith, A (1986). In Flora of Pakistan; Nasir, E.; Ali, S. I., Eds.; Shamim Printing Press, Karachi.
20.Dubey, M. P.; Srimal, R. C.; Dhawan, B. N (1969). Indian J. Pharmacol. 1, 73{75.
21.Mandal, S. C.; Dewanjee, S.; Parimaladeve, B.; Boominathan, R.; Mazumder, R.; Mazumder, A (2004). In Proceedings of the International Society of Ethnobiology-Ninth International Congress; Great, E., Ed. University of Kent, Canterbury, UK.
22.Bhakuni D. S.; Jain, S (1981). Tetrahedron, 37, 3175 {3181.
23.WHO (1996). Report of the WHO informal consultation on the evaluation and testing of insecticides. CTD/WHOPES/IC/96. 1. Giveaway: Control of Tropical Diseases Division.
24.Abbott WS (1925). A method of computing the effectiveness of an insecticide. J Eco Entomol; 18: 265-267.
25.Dhanasekaran S, Krishnappa K, Anandan A and Elumalai K. J (2013). agri. Technology; 9 (1): 29-47.
26.Kaliyamoorthy Krishnappa (2012) (a), Shanmugam Dhanasekaran, Kuppusamy Elumalai. Asian J. trop. med; 598-604.
27.Elangovan A (2012) (a), Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elumalai K. mosquitocidal activities of Corchorus capsularis against a common vector, Anopheles stephensi and a dengue vector Aedes aegypti (Diptera: Culicidae). Int. J. Rec. Sci. Res; 564-568.
28.Krishnappa K (b), Elumalai K, Dhanasekaran S, and Gokulakrishnan J (2012). larvicidal and repellent properties of Adansonia digitata against medically important human malarial vector mosquito Anopheles stephensi. J Vector Borne Dis; 49: 86-90.
29.Elumalai K (a), Krishnappa K, and Dhanasekaran S (2013). Eur. Rev. Med. Pharmacol. Sci; 17:

1404-1410.
30.Elumalai K (2012) (b), Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elangovan A. Larvicidal, ovicidal and pupicidal activity of Eranthemum roseum (VAHL) R.BR. Against malaria vector mosquito, Aedes aegypti (Diptera: Culicidae). Int. J. curr. agricul. sci; 2(7): 28-33.
31.Elumalai K(c), Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elangovan A (2012). Int. J. curr. agricul. SCI; 2 (6): 31-38.
32.Balaselvakumar, Gokulakrishnan J, Elumalai K, Dhanasekaran S, Anandan A, Krishnappa K (2012). Mosquito larvicidal activity of Oxystelma esculentum plant extracts against Anopheles stephensi (Diptera: Culicidae). Int. J. Recent. Sci. Res; 3 (5): 321-324.
33.Mullai K, Jebanesan A (2007). Larvicidal, ovicidal and repellent activities of the leaf extract of two Cucurbitacious plants against filarial vector Culex quinquefasciatus (Say) (Diptera: Culicidae). Trop Biomed; 24 (1): 1-6.
34.Krishnappa K, Mathivanan T, Elumalai A, Jeyasankar A, Dhanasekaran S and Elumalai K (2013). Int J of Interdisci Res And Revs; 01 (04): 11-18.
35.Elangovan A, Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elumalai K (2012). Larvicidal and Ovicidal activities of Exacum pedunculatum (LINN.) (Gentinaceae) against a common malarial vector, Anopheles stephensi LISTON (Diptera: Culicidae). Int. J. Recent. Sci. Res; 3 (6): 559-563.
36.Karunamoorthi K, Ilango K (2010). Larvicidal activity of Cymbopogon citratus (DC) Stapf, and Croton macrostachyus Del. against Anopheles arabiensis Patton, a potent malaria vector. Eur Rev Med Pharmacol Sci; 14(1):57-62.