International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

INDIGENOUS PLANT EXTRACTS FOR THE CONTROL OF INDOPLANORBIS EXUSTUS (DESHAYES)

[1]Bency Scaria and [2]Koshy P.M*

Post Graduate and Research Department of Zoology, St. Stephens College, Pathanapuram

drkoshypm@gmail.com

Snails belong to the phylum Molluscs are the second largest group of the animal kingdom after insects. Many species of freshwater snail belonging to the family Planorbidae are intermediate hosts of highly infective fluke (trematode) larvae of the genus Schistosoma which cause schistosomiasis. The present study is to find out potential plant biocides against the common dreaded aquatic pulmonate vector snail of India namely Indoplanorbis exustus (Deshayes). The preliminary screening aqueous (cold and hot water) leaf extract of *Sphagneticola calendulacea* on *Indoplanorbis exustus* at the concentration of 1000 mgL^{-1} at room temperature 28 ± 2°C, showed 100% mean mortality rate on the first day of incubation. Definitive screening of the molluscicidal activity of the cold water (ambient) leaf extract The mean mortality rates of the snail are 100, 77.78, 55.56, 33.33 and 11.11% at the doses of 500, 450, 350, 300 and 250 mgL^{-1} respectively. The hot water (35°C) leaf extract of Sphagneticola *calendulacea* on the snail showed a mean mortality percentage of 77.8, 66.67, 44.44 and 22.22% observed in concentrations 400, 350, 300, 250 and 200 mgL^{-1}. The LC_{50} value of molluscicidal activity of the cold water leaf leaf extract of Sphagneticola *calendulacea* on the snail *Indoplanorbis exustus* was found out to be **329.76 mgL^{-1}** and hot water (35°C) **250.38 mgL^{-1}**

Key words: Schistosomiasis, *Indoplanorbis exustus, Sphagneticola calendulacea*

Introduction

Molluscs are the second largest group of the animal kingdom after insects and they are important invertebrates of the world, divided into 6 classes. Many species of freshwater snail belonging to the family Planorbidae are intermediate hosts of highly infective fluke (trematode) larvae of the genus *Schistosoma* which cause schistosomiasis, also called bilharziasis, in Africa, Asia and the Americas. The digenic trematodes or flukes are parasites of vertebrates and to complete their larval stages need one or more intermediate hosts. Snail habitats include almost all types of freshwater bodies ranging from small temporary ponds

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

and streams to large lakes and rivers. *I. exustus* is best known as the intermediate host responsible for the transmision of *Schistosoma nasale* , *Schistosoma spindale* and *Schistosoma indicum.* Indeed *I. exustus* may be the sole natural intermediate host for these three *Schistosoma* species on the Indian sub-continent (Ravindran *et al*., 2007). Indoplanorbis exustus is responsible for the transmission of Schistosoma nasale, Schistosoma spindale and Schistosoma indicum as well as other trematodes such as Echinostoma spp. and some spirorchids (Liu *et al*., 2010). In view of the wide geographical range of this snail and its importance as a host for several species of *Schistosoma*, there is a need to understand the phylogenetics and dispersal history of *Indoplanorbis*. *Indoplanorbis exustus* is a hermaphroditic invasive snail species with high fecundity. Within one year of introduction the snail is able to colonize habitats with well-established populations of other pulmonate and prosobranch snails.

It is important, therefore, to control the snails by all means. Controlling snails through chemical method involves large scale use of pesticides which is not only expensive but may also prove hazardous to the health of livestock and man. The only commercially available chemical molluscicide, niclosamide (Bayluscid), is very expensive. A 50% wettable powder of niclosamide ethanolamine salt (WPN) is widely used for snail control in China (Jing Xia Jing Xia *et al.*, 2014). The necessity for alternatives to chemical control of snail and the growing interest and imperativeness of biological control methods have been emphasised by Joshi (1990).

A study has been carried out to evaluate the predatory potential of S. urinator adult on two freshwater snails that serves as intermediate hosts of Schistosoma. The adult bug could kill and consume the two intermediate hosts: Bulinus truncatus and Biomphalaria alexandrina (Younes *et al.*, 2017).

MATERIALS AND METHODS

Collection, transportation and maintenance of *I .exustus*

Collection of the snail, *Indoplanorbis exustus* (Fig.1). was made from the Thannikulam pond near Kollaka 9.040587, 76.543286 using long handled sieve, dip net, scoop etc (W.H.O.,1965) from March to May 2018. The snails were brought to the laboratory within 3 hours of their catch, in screw- capped glass bottles filled with the pond water, along with the water plants and stones on which the snails were found in the pond. In the laboratory, the snails were released into dechlorinated tap water in round glass troughs of 30 cm diameter filled to a height of 15 cm, at the rate of 50-60 snails per trough at room temperature (27±2^{o}C). Water plants like Certophyllum sp., and pistia sp., were introduced

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

into water troughs as the snails were reported to feed, crawl and deposit their eggs on these plants (Anantaraman, 1955). The snails were fed ad libitum with dried lettuce and beans following De Bont, Vercruysse, Van Aken, Southgate and Rollinson (1991).

Fig. 1. *Indoplanorbis exustus*

Collection of the plant materials

Sphagneticola calendulacea (Fig.2) belongs to the family Asteraceae which is a very useful herbal medicinal plant. Its common name is Chinese Wedelia and is a flowering plant species in the genus *Sphagneticola*. The leaves can be used in treatment of dermatological disorders, cough, headache, hair loss, strengthening the nervous system, lack of blood, digestive system disorders (Meena *et al.,* 2011). The leaves are used in dyeing grey hair and in promoting the growth of hair (Martin *et al.,* 2003).

Fig. 2. *Sphagneticola calendulacea*

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Preparation of the plant extracts

All the plant materials used in the analyses were fresh because it had been reported that alkaloids could be decomposed by milling of dried plant materials (Farnsworth, 1966). The extracts were prepared following the method of Duncan and Sturrock (1987).

Preliminary screening

Preliminary screening of the aqueous (cold and hot water) leaf, extracts of *L. camara* and *S. calendulacea* for their biocidal activity against *I. exustus* was carried out following the procedure of Duncan and Sturrock (1987).

The extracts of the plant materials that have shown very high levels of molluscicidal activity in the preliminary screening, resulting a mean mortality rate of 100% of the snails within 24 hours of incubation, were alone considered for definitive screening, adopting the procedure of Duncan and Sturrock (1987).

RESULTS

Table 1 Represents the preliminary screening ambient leaf extract of Sphagneticola calendulacea on *Indoplanorbis exustus* at the concentration of 1000 mgL^{-1} at room temperature 28 ± 2^{o}C. The mean mortality rate in the leaf extract is 100% in the first day of incubation. In the control no mortality was noticed. The pH of the control was 7 and in the experimental medium pH 8. The broad band evaluation of hot water leaf extract of *Sphagneticola calendulacea* on the snail *Indoplanorbis exustus* at the concentration of 1000 mgL^{-1} at room temperature 28 ±2^{o}C were represented in table 2. 100% mean mortality was obtained at first day in the incubation medium which extracted at 35, 40, 50^{o}C. At fourth day 88.88% mortality was noticed at 60^{o}C but no mortality noticed in the fifth day of incubation. It was also observed that the lowest mortality was in 35^{o}C. At 60^{o}C no mortality was noticed in the first day of incubation but 33.33, 33.33, 75 % in 2nd, 3rd and 4th days of incubation.

The result of the definitive screening of the molluscicidal activity of the cold water (ambient) leaf extract of *Sphagneticola calendulacea* on *Indoplanorbis exustus* at various concentrations at room temperature are given in Table 3. The mean mortality rates of the snail are 100, 77.78, 55.56, 33.33 and 11.11% at the doses of

500, 450, 350, 300 and 250 mgL^{-1} respectively. At concentration 500 mgL^{-1} 100% mortality was noticed in the first day of incubation. Hot water ambient leaf extract

Definitive screening of the molluscicidal activity of the hot water (35^{o}C) leaf extract of Sphagneticola *calendulacea* on the snail *Indoplanorbis exustus* at various concentrations at

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

room temperature 28 ±2°C recorded in table 5. A 100% mean mortality was noticed in incubation medium at concentration 400 mgL^{-1}. Mean mortality percentage of 77.8, 66.67, 44.44 and 22.22% observed in concentrations
400, 350, 300, 250 and 200 mgL^{-1}. In this case also 100 percentages mean mortality in the fifth day at 400 mgL^{-1}.

The mean mortality rates of I. exustus are calculated based on the values obtained in the definitive screening. The mean mortality rates of the snail are 100, 77.78, 55.56, 33.33 and 11.11% at the doses of 500, 450, 350, 300 and 250 mgL^{-1} respectively. The conversion values of the concentrations into log. Concentrations and % of kill into probit kill are given in Table 4. The LC_{50} value of molluscicidal activity of the cold water leaf leaf extract of Sphagneticola *calendulacea* on the snail *Indoplanorbis exustus* was found out to be **329.76 mgL^{-1}**.

The percentage mean mortality rate of hot water (35°C) leaf extract of Sphagneticola *calendulacea* on the snail *Indoplanorbis exustus* at various concentrations at room temperature was noted as 22.22, 44.44, 66.67, 77.78 and % at concentrations 400, 350, 300, 250, 200 mg L^{-1} respectively. 100% mean mortality observed in the first day of incubation in 400 mg L^{-1}.pH of the incubation medium was noticed as 8 except in 200 mg L^{-1}. The conversion values of the concentrations into log. Concentrations and the respective mean mortality rates (% kill) and probit kill of the snail are given in Table 6. The LC_{50} value of molluscicidal activity of the hot water leaf extract of *Sphagneticola calendulacea* on the snail *Indoplanorbis exustus* was found out to be **250.38 mgL^{-1}**.

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 1. Preliminary screen g of the molluscicidal activity of the cold water leaf extract of *Sphagneticola calendulacea* on the snail *Indoplanorbis exustus* at the concentration of 1000mgL-1 at room temperature 28 ±2°C

	Control					Experimental				
						Sphagneticola calendulacea				
	Days of Incubation					Days of Incubation				
	1	2	3	4	5	1	2	3	4	5
No. of snail incubated	9	9	9	9	9	9	-	-	-	-
No. of snail dead	0	0	0	0	0	9	-	-	-	-
No. of replicates	3	3	3	3	3	3	-	-	-	-
Mortality rate %					00.00					100.00
SD										00.00
pH	7	7	7	7	7	8	-	-	-	-

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 2. Preliminary screening of the molluscicidal activity of the hot water leaf extract of *Sphagneticola calendulacea* on the snail *Indoplanorbis exustus* at the concentration of 1000mgL^{-1} at room temperature 28 ±2^{o}C.

	Control					Experimental																			
						35 ^{o}C					40 ^{o}C					50 ^{o}C					60 ^{o}C				
	Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
No. of snail incubated	9	9	9	9	9	9	-	-	-	-	9	-	-	-	-	9	-	-	-	-	9	9	6	4	1
No. of snail dead	0	0	0	0	0	6	-	-	-	-	6	-	-	-	-	9	-	-	-	-	0	3	2	3	-
No. of replicates	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
Mortality rate %				00.00					100.00					100.00					100.00						88.88	
pH	7	7	7	7	7	8	-	-	-	-	8	-	-	-	-	8	-	-	-	-	8	8	8	8	-

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 3. Definitive screening of the molluscicidal activity of the cold water leaf extract of *Sphagneticola calendulacea* on the snail *Indoplanorbis exustus* at various concentrations at room temperature 28 ±2°C.

	Experimental																								
	500 mgL^{-1}					450 mgL^{-1}					350 mgL^{-1}					300 mgL^{-1}					250 mgL^{-1}				
	Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
No. of snail incubated	9	-	-	-	-	9	8	6	4	3	9	8	5	3	-	9	8	7	7	6	9	9	9	8	7
No. of snail dead	9	-	-	-	-	1	2	2	1	1	1	2	2	-	-	-	1	-	1	1	0	0	0	1	-
No. of replicates	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
Mortality rate %				100.00					77.78					55.56					33.33					11.11	
pH	8	8	8	8	8	8	8	8	8	8	8	8	8	8	-	8	8	8	8	8	7	7	7	7	7

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 4. Conversion table of the concentrations of the cold water leaf extract of Sphagneticola *calendulacea* in to log concentration and the respective mean mortality rate of (%) *Indoplanorbis exustus* in to probit kill.

Con. Mg L^{-1}	Log. Con.	Mortality %	Probit kill
250	2.397940009	11	3.72
300	2.477121255	33	4.56
350	2.544068044	56	5.15
450	2.653212514	78	5.77
500	2.698970004	100	8.09

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 5 . Definitive screening of the molluscicidal activity of the hot water (35^{o}C) leaf extract of Sphagneticola *calendulacea* on the snail *Indoplanorbis exustus* at various concentrations at room temperature 28 ±2^{o}C.

	Experimental																								
	400 mgL^{-1}					350 mgL^{-1}					300 mgL^{-1}					250 mgL^{-1}					200 mgL^{-1}				
	Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation					Days of Incubation				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
No. of snail incubated	9	-	-	-	-	9	8	6	4	3	9	8	5	3	-	9	8	7	7	6	9	9	9	8	7
No. of snail dead	9	-	-	-	-	1	2	2	1	1	1	3	2	-	-	1	1	-	1	1	0	0	1	1	-
No. of replicates	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
Mortality rate %				100.00					77.78					66.67					44.44					22.22	
pH	8	8	8	8	8	8	8	8	8	8	8	8	8	8	-	8	8	8	8	8	7	7	7	7	7

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

Table 6. Conversion table of the concentrations of the hot water (35^{o}C) leaf extract of *Sphagneticola calendulacea* in to log concentration and the respective mean mortality rate of (%) *Indoplanorbis exustus* in to probit kill.

Con. Mg L-1	Log. Con.	Mortality %	Probit kill
200	2.30103	22	4.23
250	2.39794	44	4.85
300	2.477121	66	5.41
350	2.544068	78	5.77
400	2.60206	100	8.09

International Research Journal of Natural and Applied Sciences
ISSN: (2349-4077)
Impact Factor 5.46 Volume 4, Issue 11, November 2017
Website- www.aarf.asia, Email : editor@aarf.asia , editoraarf@gmail.com

DISCUSSION

Preliminary screening of the biocidal activity of the cold water (ambient), hot water, leaf extracts has shown 100% mean mortality rates of the snail I. exustus in 1000 mgL^{-1} concentration. The aqueous extraction used because it was found to be more promising. This could be because water is a universal solvent, used to extract plant products (Das, *et al.*, 2010). Hence in the present study the aqueous extracts used for definitive screening. The present observation is very encouraging when compared with yet another earlier report about the molluscicidal property of different plant extracts.

Phytochemical study showed that the leaves of the plant contain isoflavonoids, bisdesmosidic oleanolic acid saponins and wedelolactones Haldar *et al.*, 2011. Norwedlolactone has also been isolated from alcoholic extract of leaves. Norwedelic acid (III) (5, 6-dihydroxy-2 (2', 4', 6'-trihydroxyphenyl)- benzofuran-3-carboxylic acid) (Masoodi *et al.*, 2011). The molluscicidal activity of the saponins may be due to their characteristic detergent effect on the soft body membranes of the molluscs. However, saponins are structurally diverse and have multiple effects in animal cells .

The $LC_{50} > 100 < 500$ mg/l indicates that the substance is moderately toxic. In the present study the LC_{50} values determined by using regression equation that is statistically significant. The finding of LC50 values found to be **329.76 mgL^{-1} and 250.38 mgL^{-1}**. The results are more promising compared to the LC_{50} values of *Rumex dentatus* (580 ppm), *Lycium schweinfurhii* (740 ppm). The results are very close to the effect of *Momordica charantia* lyophilized fruit powder where the 96 h LC_{50} observed as 318.29 **mgL^{-1}** (Aparna Upadhyay et al., 2013). Prolonged exposure of the snails suffered from higher mortality due to negative effect on their growth (Mohamed *et al.*, 2000). This may be due to the effect of leaf extract on the digestive system and hermaphrodite gland (Ahmed *et al.*, 2014). Aparna Upadhyay *et al.*, 2013 reported that the time-dependent toxic effects of tested plant products may be due to the uptake of active components by snails, which progressively increases in the body with an increase in exposure duration. It is also possible that the active compound(s) could change into more toxic forms in the aquarium water or in the snail's body due to the action of various enzymes. R^2 value explained that more than 80% cases able to explained in the model. With regard to the molluscicidal activity of the cold water leaf and hot water (35^oC) leaf extracts proves the efficacy of *Sphagneticola calendulacea* on *I. exustus* in in the laboratory condition.

References

Anantharaman , M. 1971. The epidemiology of nasal Schistomiasis in animals. ICAR workshop on co-ordinated research project on calf mortality and nasal Schistosomiasis, Bangalore, January 7-10, 1971.

Anantharaman , M. 1995. Biological control of aquatic snails. *Indian j. vet. Sci. & anim. hus*. 25:65-67.

Aparna, Uupadhyay.Vinay, K. Singh. & Dinesh, K. Singh. 2013. Characterization of molluscicidal component of Moringa oleifera leaf and Momordica charantia fruits and their modes of action in snail Lymnaea acuminate. *Rev. Inst. Med. Trop. Sao Paulo*. 55(4):251-259.

Das, K. Tiwari, R.K.S. and Shrisvastava, D.K. 2010. Techniques for evaluation of medicinal plant products as antimicrobial agent: Current methods and future trends. *Medicinal Plants Research*. 4(2), 104-111.

De Bont, Vercruysse, Van Aken, Southgate and Rollinson. 1991. Studies of relationships between Schistosoma nasale and S.spindale infection and their snail host Indoplanorbis exustus. *J. helminth*.65:1-7.

De Bont,J. Vercruysse, J. Van Aken,D.Warlow, A.Southgate, V.R. and Rollinson,D.1991.use of Enzyme electrophoresis for differentiating Schistosoma nasale and S.spindale infection of Indoplanorbis exustus in Srilanka. *Syst. Parasitol*.20:161-164.

Dunkan,J. and Sturrock, P.F.1987.Laboratory evaluation of potential molluscicides (Page 251-256). In: plant molluscicides (Edr. Mott, K.E.), john Willy and sons Ltd. chichester, New York, Brisbane, torentoand Singapore.

Farnsworth, N.R. 1966. Biological and phytochemical screening of plants. *J. Pharm. Sci*. 55: 225 – 276.

Haldar, P.K. Bhattacharya, S. Dewanjee, S, Mazumdera, U.K.2011. Chemopreventive efficacy of Wedelia calendulaceae against 20- methylcholanthrene-induced carcinogenesis in mice. *Environ Toxicol Pharmacol* . 31: 10-17.

Jing Xia, Yi Yuan, Xingjian, Cu. Penghu, Wei. Guiling, Li. Min, Liu. Jianqiang, Li. Rujuan, Chen.Zhengping, Zhou and Shaofa Nie. 2014. Evaluating the Effect of a Novel Molluscicide in the Endemic Schistosomiasis Japonica Area of China. *Int. J. Environ Res Public Health*. 11(10): 10406–10418.

Joshi, C.O.1990. Screening of molluscicidal activity of plant extracts for the biocontrol of snailintermediatehost. M.Phil thesis, university of Madras, madras.

Martin, K.P. Benna, M.R. Joseph, D. 2003. High frequency axillary bud multiplication and ex- vitro rooting of Wedelia chinensis (Osbeck) Merr. A medicinal plant. *Indian J Exp Biol*. 41(3):262-266.

Masoodi, M.H.Ahmad, B.Wali, A.F. Zargar, B.A. Dar, M.A.2011. Recent developments in phytochemical and pharmacological studies of Wedelia calendulaceae- *A review. Indian J Nat Prod*. 27(1):3-7.

Meena, A.K. Rao, M.M. Meena, R.P. Panda, P. Renu. 2011. Pharmacological and Phytochemical Evidences for the Plants of Wedelia Genus. A Review. *Asian J. Pharm. Res*. 1(1):07-12. 6.

Mohamed. A.M. Bakry, F.A. Heiba, F.N. 2000. Molluscicidal effects of Abamectin on Biomphalaria alexandrina and its infection with Schistosoma mansoni The 1st Inter. Cong. Biolg. Sci. Tanta Univ., Egypt. 1 (2): 207-216.

Ravindran, R. Lakshmanan,B. Ravishankar, C. and Subramanian, H. 2007. Visceral **schistosomiasis among** domestic Ruminants slaughtered in wayanad, south India. *The Southeast Asian journal of tropical medicine and public health*. 38 (6):1008-10.

W.H.O. 1965. Snail control in the prevention of Bilharzizsis. World Health Organization.

Younes, Aly. Hanaa El-Sherief. Fathia Gawish. Marwa Mahmoud.2017. Biological control of snail hosts transmitting schistosomiasis by the water bug, Sphaerodema urinator. arasitol Res DOI 10.1007/s00436-017-5402-5