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RESEARCH ARTICLE

Studies on acute and chronic toxicity of cadmium to freshwater snail *Lymnaea acuminata* (Lamarck) with special reference to behavioral and hematological changes

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Abstract Molluscs have long been regarded as promising bioindicator and biomonitoring subjects for heavy metals as molluscs are highly tolerant to heavy metals and exhibit high accumulation in their body. In spite of several previous studies about the impact of cadmium on molluscs, little information exists in literatures concerning the toxic effects of cadmium on *Lymnaea acuminata*, especially pertaining to behavioral and hematological changes as these are considered effective bioindicators and biomonitoring variables for detecting heavy metals in polluted water bodies. In the present study, the median lethal concentrations of cadmium chloride to snail, *Lymnaea acuminata*, were estimated to be 9.66, 7.69, 6.26, and 5.54 mg/L at 24, 48, 72, and 96 h, respectively. For behavioral studies, variable test concentrations of cadmium from 0.00 to 10 mg/L were used. The clumping tendency, crawling activity, and touch reflex in the exposed snails were gradually decreased with higher concentrations at 72 and 96 h. For measuring the hemocyte numbers in the circulating hemolymph of snail during chronic cadmium exposure, two sublethal doses of cadmium (10 and 20% 96-h LC_{50}—0.55 and 1.11 mg/L, respectively) were used. A significant variation ($p < 0.05$) from the control at all exposure times (7, 14, 21, and 28 days) was recorded at 1.11 mg/L concentration. The total count of circulating hemocytes was significantly reduced ($p < 0.05$) compared to the controls at both concentrations of cadmium exposure at all time periods except 14 and 21 days exposure at 0.55 mg/L where values were non-significantly increased. In comparison between two sublethal doses, blood cells were significantly ($p < 0.05$) lowered at 1.11 mg/L cadmium treatment. Considering the behavioral and hematological data, it seems possible to forecast the physiological state of snails in cadmium-contaminated water bodies and these findings can be used in determining the safe disposal level of cadmium in aquatic ecosystem.

Keywords Lethal concentration · Cadmium · *Lymnaea acuminata* · Behavioral changes · Hemocyte

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Introduction

The rapid industrialization is one of the major causes of heavy metal pollution in aquatic ecosystem. Among heavy metals, cadmium possesses a significant ecological problem for its ability to be accumulated in living organisms in a cumulative manner (Jensen and Bro-Rasiriussen 1992; Alazemi et al. 1996). Cadmium can enter the environment from various anthropogenic sources, such as by-products of zinc, lead, and copper mining, and smelting, coal combustion in the thermal power plants, iron and steel production, pigments, fertilizers, and pesticides (Hodgson 2010; Gad 2005). Though cadmium occurs naturally in the environment in insignificant amounts, its release in the environment is steadily increasing due to increase in anthropogenic activities causing severe pollution of soil and aquatic ecosystems. Aquatic organisms mainly absorb cadmium from polluted water through respiratory and digestive systems and body surface without significant

excretion and are prone to be affected with this heavy metal (Rainbow and White 1989; van Hatton et al. 1989).

Aquatic organisms are very much susceptible to any change or stress in their environment as they are in direct contact with water (Al-Attar 2005). In general, molluscs play an important role in the balance of nature, and also act as good bioindicator, in determining the degree of pollution of water among the aquatic organisms especially to monitor heavy metal toxicity (Nurnberg 1984; Kambale and Potdar 2010). Among the freshwater molluscs, *Lymnaea* sp. has been used as a suitable test organism in ecotoxicological studies (Russo and Lagadic 2004; Das and Khangarot 2010) and acts as a promising heavy metal toxicity bioindicator in pollution research.

In aquatic organisms like molluscs, the blood remains in the direct contact with their surroundings as molluscs have open circulatory system. Thus, blood physiology of aquatic organisms may be considered as a vital index to monitor health status of the organisms as well as an indicator of water pollution (Kori-Siakpere et al. 2006). Toxic metal ions are known to cause deleterious impact on the various blood parameters in the aquatic organisms including molluscs as they form complexes with the structural proteins, enzymes, and nucleic acids and interrupt their normal physiological functions (Chakraborty et al. 2008; Vinodhini and Narayanan 2008; Chakraborty and Ray 2009). Gastropods possess a well-developed innate defense system where hemocytes play an important role. Its role in the metal transportation and metabolism in molluscs is also well established (Suresh and Mohandas 1989; Suresh 1990). Since gastropods have an open circulatory system, any change in relation to stress immediately gets reflected in the blood. Variations in the hematocyte number present in the hemolymph in gastropods due to the changing external environment thus act as a sensitive indicator to monitor stress and also the physiological state of the organism (Mohandas et al. 1989; Guria et al. 2003; Ray et al. 2013).

In the present study, chronic toxicity of cadmium to mature freshwater snail, *Lymnaea acuminata* (class: Gastropoda; family: Lymnaeidae), was performed to determine the alteration in circulating hemocytes in the hemolymph. Behavioral changes were also assessed in this snail as an acute effect of the same toxicant.

Materials and methods

For bioassays, healthy, active, and mature freshwater snails, *Lymnaea acuminata* with mean shell height 1.98 ± 0.12 cm and mean weight 0.89 ± 0.36 g, were used as test organisms. The snails were collected from the unpolluted local ponds, washed up with tap water, and acclimatized in the laboratory condition for 72 h. For acclimatization, they were kept in well-aerated aquarium filled with unchlorinated tap water at room temperature (27 ± 0.45 °C) and provided with some aquatic macro-vegetation (*Hydrilla* sp. and *Pistia* sp.) as their natural food and resting place. The water was changed at every 24 h to avoid detritus load.

Analytical grade of cadmium chloride, $CdCl_2 \cdot H_2O$ (purity 98%, molecular weight 201.32 g/mol; E. Merck, Germany), was used as the test chemical. Water chemical analysis and the bioassays were done following the standard methods outlined in the American Public Health Association (APHA 2012). Static replacement bioassays were used for the 96-h tests (acute toxicity tests and behavioral study) in the laboratory. The underground water stored in the overhead tank was used as a diluent medium. The test medium was replaced every 24 h by freshly prepared test solution to avoid the interference of different abiotic factors with the animals' performance. The cadmium content of water was analyzed on a regular basis by flame atomic absorption spectrophotometry (Varian model spectrAA–10, AAS) according to the modified method of Brewer et al. (1969) as described in FAO (1975). The cadmium content of water was detected very near to the expected values.

During acute (96 h) bioassay, tests with snails were conducted in 15-L glass aquaria each holding 10 L of water for the determination of acute toxicity. Tap water stored in the glass aquaria (temperature 27 ± 0.45 °C, pH 7.4 ± 0.21, free CO_2 8.0 ± 0.21 mg/L, DO 5.54 ± 0.42 mg/L, alkalinity 176 ± 7.01 mg/L as $CaCO_3$, hardness 120 ± 7.0 mg/L as $CaCO_3$) was used as a diluent medium. A set of four aquaria was exposed to one single concentration of cadmium chloride to make four replicates per concentration while determining acute toxicity of the pollutant. Each set of tests was accompanied by four replicates of control. Ten test organisms were used in each replicate. While conducting acute toxicity test, different concentrations of the toxicant were prepared by weighing the required quantity of the chemical compound which was then added directly to the test medium. The test medium was then stirred with a magnetic stirrer for uniform mixing of the test chemical compound. Initially, rough range finding tests were conducted for the test organisms to determine the dose range at which mortality occurs. The selected test concentrations of cadmium chloride finally used for the determination of median lethal concentrations (LC_{50}) to the snails were 0, 1.5, 3.0, 4.0, 5.5, 6.0, 6.5, 7.0, 8.5, 9.0, 9.5, and 10.0 mg/L. Test organisms were not fed 24 h before and during the bioassays. The vitality of the snails was checked frequently (four times/day) using soft tweezers, and they were considered dead when there was no response to physical stimulation. The number of dead organisms was counted every 24 h and removed immediately from the test medium to avoid any organic decomposition and oxygen depletion.

Mortality rate at different concentrations and at different time of exposure (24, 48, 72, 96 h) was analyzed using the

computer software R version 2.14.0 (US EPA 1999) and probit analysis by Finney (1971) for determining median lethal concentrations (LC_{50}) with 95% confidence limits of cadmium to the test organisms at different time of exposure. Correlation analysis of mortality rate with different concentrations and different time of exposure (24, 48, 72, and 96 h) was analyzed following the methods described by R Development Core Team (2011) and Gomez and Gomez (1984). The behavioral changes of the test organisms in respect of their clumping tendency (lower snail strongly attached to the substratum whereas the top snail bound to the shell surface of the lower one), crawling activity (extremely slow-paced movement either on the surface or on the wall of the aquarium), and touch reflex (a slight stimulus eliciting a local contraction of the musculature under the shell) exposed to the toxicant during acute toxicity were recorded systematically by naked-eye observation following the method of Rand (1985). Scoring of behavioral changes was performed independently by two observers and the scores were collated and average was calculated and quantified in terms of −/+/++/+++ (− = none; + = mild; ++ = moderate; +++ = strong).

Chronic toxicity tests were also conducted in 15-L glass aquaria having similar water qualities as used in acute (96 h) studies, each holding 10 L of water in the laboratory condition for 28 days using two different sublethal concentrations (10 and 20% of the 96-h LC_{50} value) of cadmium and a control. The aquaria were arranged in three blocks each with three aquaria as per randomized block design (Gomez and Gomez 1984), thereby giving three replicates for each of the two sublethal concentrations and control. Each aquarium was stocked with ten healthy, mature, active, and properly acclimatized snails for hematological study under sublethal exposure. In aquaria, they were provided with their natural food like *Hydrilla* sp. and *Pistia* sp. The blood samples of the treated and non-treated *L. acuminata* were analyzed for hemocyte count. The hemolymph samples of the snails were analyzed at every 7 days interval with a total exposure period of 28 days following the method described by Suresh (1990). For this study, the snails were taken out from the aquaria and were washed with cold water in order to remove the feces and excess mucus. Before collection of hemolymph, the water adhering to the snail was soaked and the foot cleaned with tissue paper. The hemolymph was collected from the sinus using a needle fitted with heparinized capillary tube. Hemolymph was also collected by touching the foot with the tip of a micropipette. As a result, the snail was forced to retract deeply into its shell and extruded hemolymph. In this way, about 0.2 mL of oozing hemolymph was collected aseptically from each snail and was immediately stored at 4 °C to avoid hemocyte clumping. An aliquot of fresh extracted hemolymph of each specimen of *L. acuminata* was placed on Neubauer hemocytometer for determination of total hemocyte count per milliliter of hemolymph (Caprette 2007). Post-collection, the first two drops of hemolymph were expelled while the succeeding drops were discharged in one of the V-shaped wells of hemocytometer with a capillary tube. The cells were allowed to settle for 1 to 2 min and then determination of cell numbers was carried out after 2 min.

The mean values of hematological parameter for the control and experimental snails were subjected to one-way analysis of variance (ANOVA) at 0.01 and 0.05 levels using the computer software R version 2.14.0 followed by Duncan's Multiple Range Test (DMRT) for determining significant differences among the means (Gomez and Gomez 1984).

Results

The median lethal concentrations (LC_{50}) at different time of exposure (24, 48, 72, and 96 h) with 95% confidence limits of cadmium to *Lymnaea acuminata* are given in Table 1. No mortality was observed in the control group during the experiment. Significant co-relationship ($p < 0.01$) was observed between mortality rate of the snails and exposure concentrations at all the exposure times (24, 48, 72, 96 h) as tabulated in Table 2. On the other hand, the co-relationship between mortality rate and exposure times was significant ($p < 0.01$) at all concentrations except 3.0 and 5.5 mg/L (Table 3).

The behavioral responses observed in the test organisms exposed to cadmium chloride are summarized in Table 4. The clumping tendency and crawling activity of the snail were low to moderate at control and lower doses of cadmium but were increased with the increasing concentrations of the toxicant at 24 h exposure. With the passage of time, the clumping tendency and the crawling activity gradually decreased with higher concentrations and increased at lower concentrations at 72 and 96 h exposure. No such clumping and crawling effects were recorded at 8.5–9.5 mg/L of 96 h exposure. Touch reflex of the snail was observed at all the concentrations of 24 and 48 h exposures but it was found to decrease at the higher concentrations with the progress of time. Touch reflex was completely absent at 9.0 and 9.5 mg/L of 72 h and at 8.5–9.5 mg/L of 96-h treatments (Table 4).

Single-factor ANOVA was carried out with mean values of hemocyte numbers in the circulating hemolymph of *L. acuminata* exposed to two sublethal doses (10 and 20% 96-h LC_{50} value) of cadmium (0.55 and 1.11 mg/L). The results showed a significant variation of hemocyte numbers ($p < 0.05$) at all time period (7, 14, 21, and 28 days) only at 1.11 mg/L cadmium treatment (Table 5). In this experimental setup, the number of hemocytes in control snails (0.00 mg/L) at 7,14, 21, and 28 days remained constant which lowered significantly ($p < 0.05$) at 1.11 mg/L cadmium treatment as the cadmium exposure period increased from 7 to 28 days. However, at 0.55 mg/L cadmium treatment, no lowering of hematocyte numbers in comparison to control was observed at

Table 1 Median lethal concentrations (LC_{50}) with 95% confidence limits of cadmium to *Lymnaea acuminata* at different period (h) of exposure (control group theoretical spontaneous response rate = 0.0000)

Period (h)	Median lethal concentrations (LC_{50}) (mg/L) with 95% confidence limits	Slope ± SE	Intercept ± SE
24	9.66 (8.36–14.15)	4.47 ± 1.28	0.60 ± 1.12
48	7.69 (6.71–9.48)	3.84 ± 0.99	1.60 ± 0.84
72	6.26 (5.29–7.35)	4.75 ± 1.06	1.22 ± 0.86
96	5.54 (4.71–6.28)	5.43 ± 1.08	0.96 ± 0.87

14 and 21 days. Thus, significant variation ($p < 0.05$) was only observed during the experiment period at higher dose of treatment. During the study, no mortality of the snails in the control was observed and the mortality rate of the exposed snails was also insignificant in respect of control group.

Discussion

Pollution of the aquatic ecosystem due to cadmium has been a serious concern for quite some time (Gad 2005). In aquatic ecosystem, cadmium is readily absorbed by the organisms directly from the water in its free ionic form (AMAP 1998). In this connection, snails are the most susceptible aquatic group to cadmium toxicity (Kaviraj and Das 1990). This may be due to direct contact of their vital organs to the toxicant or to the absence of operculum in cases of Lymnaeidae which is in agreement with the result of the present investigation. The 96-h median lethal concentration of cadmium to *Lymnaea acuminata* was found to be 5.54 mg/L in the present study which nearly corresponds with the 48-h LC_{50} value of cadmium to the snail, *Filopaludina martensi* (5.01 mg/L) (Piyatiratitivorakul and Boonchamoi 2008), and 72-h LC_{50} value to *Melanoides tuberculata* (5.24 mg/L) (Shuhaimi-Othman et al. 2012). Similar findings were also recorded to the mud crab, *Eurypanopeus depressus* (72-h LC_{50} 4.9 mg/L) (Collier et al. 1973), tropical grass shrimp, *Palaemon northropi* (96-h LC_{50} 6 mg/L) (Chung 1980), and Atlantic oyster drill, *Urosalpinx cinerea* (96-h LC_{50} 6.6 mg/L) (WHO 1992). This near correspondence in median lethal concentration values between *Lymnaea acuminata* and already mentioned invertebrates (crabs, shrimps, and oysters) may be attributed to the fact that all these organisms have open circulatory systems; open circulatory systems evolved primarily in phylum molluscs and arthropods, thus similar impact due to cadmium exposure may be expected in such animals. However, it was observed in our experimental setup the fresh water snails were insensitive to cadmium in acute exposure, with a very high LC_{50} value of 5.54 in 120 mg/L hardness water which is several times greater than other sensitive freshwater organisms in 100 mg/L hardness water (US EPA 2016). This high LC_{50} value can be attributed to the increased hardness of water and the difference in hardness of water can influence the median lethal concentration as evidenced by Datta et al. (2003). In addition to the physicochemical properties of water, another contributing factor—the age of organisms (Weir and Walter 1976; Wang et al. 2010)—can also cause variation in LC_{50} values among species (Kaviraj and Das 1990) or even between closely related species (Okocha and Adedeji 2011). Susceptibility of *L. acuminata* to the lethal effect of cadmium was duration and concentration dependent as recorded in the present study. This result corresponds with the findings of Das and Khangarot (2010) on *L. luteola* exposed to cadmium. Many workers have also reported similar findings in other aquatic organisms exposed to cadmium (Nelson et al. 1976; Singh et al. 2010; Tiwari et al. 2011; Dhara et al. 2014).

Table 2 Correlation (r) of mortality rate for *Lymnaea acuminata* with cadmium chloride concentrations at different time of exposure (h)

Exposure time (h)	r ± SE
24	0.93** ± 0.54
48	0.98** ± 0.71
72	0.98** ± 0.90
96	0.99** ± 1.00

**$p < 0.01$

Table 3 Correlation (r) of mortality rate for *Lymnaea acuminata* with exposure times at each concentrations of cadmium chloride (mg/L)

Concentrations of cadmium chloride (mg/L)	r ± SE
1.5	NA
3.0	0.89 ± 0.25
4.0	1.00** ± 0.56
5.5	0.92 ± 0.54
6.0	1.00** ± 0.56
6.5	0.95* ± 0.71
7.0	0.98* ± 0.74
8.5	0.96* ± 0.94
9.0	0.99* ± 0.96
10.0	0.98* ± 1.03

*$p < 0.05$; **$p < 0.01$

Table 4 Impact of cadmium chloride on the behavioral responses of *Lymnaea acuminata* at various concentrations during different hours of exposure. *CA* crawling activity, *CT* clumping tendency, *TR* touch reflex, − none, + mild, ++ moderate, +++ strong

Dose (mg/L)	24 h			48 h			72 h			96 h		
	CA	CT	TR	CA	CT	TR	CA	CT	TR	CA	CT	TR
0.0	++	+	+++	++	+	+++	++	+	+++	++	++	+++
1.5	++	+	+++	++	+	+++	++	++	+++	+++	+++	+++
3.0	++	+	+++	++	+	+++	+++	++	+++	+++	+++	+++
4.0	++	++	+++	++	++	+++	+++	+++	+++	+++	+++	+++
5.5	++	++	+++	++	+++	+++	+++	+++	+++	++	++	++
6.0	+++	++	+++	+++	+++	+++	+++	+++	+++	++	+	++
6.5	+++	+++	+++	+++	+++	+++	+++	+++	++	++	+	++
7.0	+++	+++	+++	+++	+++	+++	++	++	+	+	+	+
8.5	+++	+++	+++	+++	++	++	+	++	+	−	−	−
9.0	+++	+++	++	++	++	++	+	+	−	−	−	−
9.5	+++	+++	++	++	++	++	−	−	−	−	−	−

The alteration in the behavioral pattern is considered the most sensitive indicator of potential toxic effects of cadmium in aquatic organisms (Tiwari et al. 2011). In our present study, *Lymnaea acuminata* showed dose- and duration-dependent behavioral changes highlighting their avoidance reaction to the cadmium toxicity. This observation was in accordance to earlier studies showing similar behavioral activities in the water snail, *Physa integra* and *Taphius glabratus* with their inability to crawl when exposed to such polluted water (WHO 1992; Tantulvesn and Pornprapa 1995). These avoidance reactions may be contributed to the narcotic effects or to the change in sensitivity of chemo-receptors in the exposed organisms (Suterlin 1974). Enzymatic as well as ionic disturbances in the blood or tissue may also be associated with such alteration in animal behavior (Larsson et al. 1981).

Changes in hematological values of aquatic organisms are known to occur in relation to physiological stress, disease, and toxic environmental conditions (Kori-Siakpere et al. 2006). One of the known manifestations of stress in molluscs is the significant fluctuation in the total hemocyte count (Sminia 1981). The number of hemocytes is a factor directly linked with innate defense system of the gastropods which alters defense mechanism, mainly at the cellular level (Suresh and Mohandas 1989). Thus, the alteration of total hemocyte count in gastropods can be considered as one of the most reliable indicators to monitor stress factor and also to assess their physiological state (Mohandas et al. 1989; Auffreta et al. 2006). In this context, mercury is known to cause significant hemocyte mortality in Pacific oyster, *Crassostrea gigas* (Gagnaire et al. 2004). Reduced hemopoiesis was observed in copper-stressed oysters *C. virginica* molluscs (Cheng 1988). Bindya Bhargavan (2008) found time-dependent decrease of hemocyte count in green mussel, *Perna viridis* exposed to copper and mercury. In a separate study by Suresh (1990), the hemocyte count also decreased significantly in *Indoplanorbis exustus* and *Lymnaea acuminata* exposed to copper. In accordance to these previous studies, our present study on cadmium-treated *L. acuminata* showed a decrease in hemocyte count (Table 5) confirming disturbance to the organisms. This particular observation is also in agreement with a previous experimental study by George et al. (1983) on oyster, *Ostrea edulis*, exposed to cadmium. Significant difference in total hemocyte counts as observed in our *L. acuminata* exposed to different doses of cadmium acts as a good indicator of ecological difference. The decrease in hemocyte count in our experimental snails exposed to cadmium may have resulted from hemodilution as a result of mechanism that reduces the concentration of the toxicant/pollutant in the circulatory

Table 5 Mean values (± SD) of circulating hemocytes (no./mL) in the hemolymph of *Lymnaea acuminata* exposed to sublethal concentrations of cadmium in water at different days of exposure (7, 14, 21, and 28 days). Common superscript letters between any two mean values indicate their similarity while two different superscript letters indicate significant difference at DMRT, $p < 0.05$ (a and b within columns and m and n within rows)

Dose (mg/L)	Mean values (± SD) of circulating hemocytes (no./mL)			
	7 days	14 days	21 days	28 days
0.00	$91290^{bm} \pm 1531$	$91475^{bm} \pm 1054$	$91600^{bm} \pm 1607$	$91625^{bm} \pm 1358$
0.55	$87114^{abm} \pm 954$	$92082^{bn} \pm 2060$	$93118^{bn} \pm 971$	$87855^{bm} \pm 1212$
1.11	$85567^{an} \pm 1401$	$83516^{amn} \pm 557$	$82620^{amn} \pm 1007$	$81814^{an} \pm 1102$

system (Kori-Siakpere et al. 2006). Bindya Bhargavan (2008) have opined that heavy metals usually get accumulated in the granular hemocytes and overloading results in the decrease in cell viability or cell death in molluscs due to degenerated enzymes and reactive oxygen species. Decrease in total hemocyte count can also be a result of reduced proliferation of hemocytes or movement of cells from circulation into damaged tissues (Pipe and Coles 1995). Besides blood cell death, low inflow of hemocytes from other sites into the hemolymph could also reduce the total hemocyte count (Bindya Bhargavan 2008). Besides, Mohandas et al. (1989) has suggested that hemocytes under stress may also migrate from the circulatory system to the gonads to phagocytose, resulting in lower count of hemocytes in the hemolymph. In contrary to this, total hemocyte count in molluscs has also been reported to increase by heavy metal pollution in some earlier studies (Pickwell and Steinert 1984; Pipe et al. 1995, 1999; Fisher et al. 2000). Pipe and Coles (1995) recorded significantly higher number of circulating hemocytes in mussels under chronic exposure of cadmium. Interestingly, similar trends were also obtained in our current study in the treated snails at lower dose of cadmium for certain period of exposure (Table 5). Such increase in the number of circulating hemocytes may be due to migration of cells from the reservoir compartment to the hemolymph as a result of stress elicited by cadmium ions (Pipe et al. 1999). It may also be due to mitosis of leukocytes or to the continuous hemopoiesis from the amoebocyte-producing organ as reported in *Biomphalaria glabrata* (Jeong et al. 1983). Similarly, Cheng (1988) reported that cadmium stimulated the process of hemopoiesis in molluscs resulting in the elevation of hemocyte level. In freshwater gastropods, the majority of hemocytes are amoebocytes with granules in the cytoplasm (Sminia 1972, 1981; Sminia et al. 1983; Ottaviani 1983; Dikkeboom et al. 1985). It was reported that although metal stress causes reduction in hemocyte counts, mature granulocytes are seldom affected. Molluscs exposed to toxicant would need increased number of granulocytes to remove the overload of toxicants as such granulocytes play important roles in internal defense (Suresh 1990). This is reflected in the increase in the number of hemocytes.

Conclusion

The present findings, thus, highlight the toxicity of cadmium to *Lymnaea acuminata* during their acute and chronic exposure. Acute toxicity studies are among the first steps in determining the water quality required for the sustenance of snails. These studies reveal the toxicant concentrations (viz. LC_{50}) that cause snail mortality even at short-term exposure. The observation on the behavioral responses of the snail in the present study may be an indicative parameter for assessing the toxicity of cadmium in the ecosystem as suggested by Doving (1992). The hematological study clearly shows that heavy metals like cadmium pose a serious threat to the biological functions of snails during their chronic exposure. On the basis of hematological changes, it might be possible to forecast the physiological state of snails in natural water bodies as well as developing new vistas pertaining to the mechanism of action of a toxicant in a molluscan body.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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