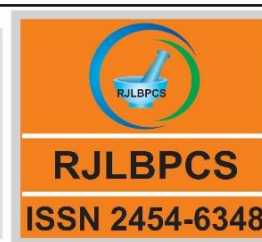


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Research Journal of Life Sciences, Bioinformatics,
Pharmaceutical and Chemical SciencesJournal Home page <http://www.rjlbpcs.com/>**Original Research Article****DOI - 10.26479/2018.0402.22****OBSERVATION ON THE DIFFERENT DEVELOPMENTAL STAGES OF
FRESH WATER SNAIL *Lymnaea Stagnalis* AFTER THE TREATMENT
WITH DOCETAXEL****S. Manekar^{1*}, P. Mahobiya² and Keerty Shrivastava³**

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ABSTRACT: *Lymnaea* is a genus of small to large sized air breathing fresh water snails, aquatic pulmonate gastropod molluscus in the family- Lymnaeide, the pond snail. These snails are harmful pests of various valuable crops. We investigated the effect of docetaxel on the egg mass of *Lymnaea*. Effects of these chemical were examined by recording mortality. To save our valuable crops from these pestiferous snails it is very necessary to control their population.

KEYWORDS: *Lymnaea stagnalis*, docetaxel, mortality, pestiferous, development.

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1. INTRODUCTION

India is an agriculture based country. Our most of the economy depends on it. Different types of habitats and agriculture fields are found in our country and different kinds of toxicants are used to prevent the vegetation from the harmful species of animals improving the yield and increasing the productivity by controlling their growth. Molluscus are one of the most, harming aquatic vegetation. This investigation is based on *Lymnaea* which comes under the family lymnaeide, the pond snail. Lymnaeids are distributed worldwide as observed [1]. *Lymnaea* is found all over the world. *Lymnaea* acts as a pest for the aquatic vegetation and harm them. These snails are harmful pests of various valuable crops and directly decline the productivity area which affects the economy of our country. To save our valuable crops from the disaster of these pestiferous snails and to control their

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population it is essential to study the development of these pestiferous snails. The facet study on embryology of snail is either missing or lacking. Many scientists attempted to work on some aspect of snail development [2, 3, 4]. The work of docetaxel toxicity on *Lymnaea stagnalis* is yet scanty, so this attempt was performed to evaluate of docetaxel toxicity on different developmental stages of *Lymnaea stagnalis*. The study revealed that embryo development was the most sensitive stage after treatment with docetaxel.

2. MATERIALS AND METHODS

Common pond snails of *Lymnaea stagnalis* belonging to family Lymnaeidae were selected for the present investigation. Sexually mature specimens of *Lymnaea stagnalis* were collected from botanical garden pond of Bhopal Lake by fishing nets or picking by hands and were kept in glass containers or troughs (2-5 liter water capacity). The collected snails were acclimatized for 7 days under laboratory conditions [5]. The water was replaced with fresh water 3 times in a week. They were fed regularly with aquatic vegetation e.g. Hydrilla to avoid the stress of starvation. The young ones hatched from the freshly laid egg masses of *Lymnaea stagnalis* were used for the experimental purpose. The egg masses laid by these snails were introduced to different concentration of docetaxel drugs separately in glass Petri dishes (50 ml capacity) in triplicate, used in the present investigation. Fresh egg masses with subsequent developmental stages of *Lymnaea stagnalis* of F₀ generation were introduced via media to different concentrations of antitubulin drugs and the data was collected in triplicate and calculated the values of LC₁₀₀, LC₅₀, LC₀ and sub-lethal concentration were detected out for each group separately and data was summarized in table no.1 by analysis method [6]. Each egg masses contain about 30 egg capsules. Docetaxel is used in the treatment of various cancers. Docetaxel is a taxoid anti neo plastic agent. The trade name of docetaxel is taxotere or docecad. It is obtained from the young shoot of the pacific yew tree. Docetaxel promotes the assembly of micro tubules from tubulin dimers and stabilizes microtubules by preventing depolymerization. This stability results in the inhibition of the normal dyanamic reorganization of the microtubule network that is essential for vital interface and mitotic cellular functions. In addition docetaxel induces abnormal arrays or bundles of microtubules throughout the cell cycle and multiple asters of microtubules during mitosis. In all types of eukaryotic cells, microtubules containing tubulin protein are found and play a major role in cell division.

3. RESULTS AND DISCUSSION

In the present investigation in the control group, mortality started on 25th days. Cleavage is spiral, after 3 to 4 hrs egg masses laid by snails in control group but after treatment cleavage started after 8± 2 with the treatment of docetaxel in *Lymnaea stagnalis*. In treatment groups, Blastula period also enlarged compare to control group. The young snail hatchability percentage was very low with high percentage of mortality after high concentration of docetaxal treatment compare to control groups. Data on the observation of different parameters on egg masses of snails was recorded and

summarized in table no. 2.

Table: 1 Data on toxicity of docetaxel on egg masses of *Lymnaea Stagnalis*.

S.No.	Name of drug	Concentration of the drug	Duration	Mortality (%)	Lethal Conc. Value
1.	Docetaxel	0.07%	72	100%	LC ₁₀₀
2.		0.04%	72	50%	LC ₅₀
3.		0.02%	72	Nil	LC ₀
4.		0.01%	72	Nil	Sub lethal Concentration

Result: 0.01% concentration of docetaxel was considered as sublethal concentration value.

Table: 2 Developmental Data of *Lymnaea stagnalis* after Treatment with Different Concentrations of docetaxel

Groups	Conc. of the docetaxel drug	Total No. of egg capsules	No. of egg complete cleavage	No. of egg complete blastula	No. of egg complete gastrula	No. of trochophore formed	No. of veliger complete formed	Total No. of young snails hatched	No. of young snails survived upto adulthood	Percentage survivality of young snails	
Control	No trace of any docetaxel drug	50	50	50	50	50	50	50	49±1	98-100%	
Experimental groups treated with docetaxel (Sublethal concentration)	0.07% (LC ₁₀₀)	50	13±1	12±1	10±1	9±1	7±1	5±1	2±1	None	0.00%
	0.04% (LC ₅₀)	50	34±1	31±1	30±1	29±1	28±1	27±1	25±1	24±1	48-50%
	0.02% (LC ₀)	50	49±1	49±1	49±1	49±1	49±1	49±1	49±1	49±1	96-98%
	0.01% (Sublethal concentration)	50	50	50	50	50	50	49±1	49±1	49±1	98-100%

In the present investigation egg laying was observed throughout the year in *Lymnaea stagnalis* were laid in the large number from July to October as also reported [3, 7, 8, 9, 10]. *Lymnaea stagnalis* were oviparous. Eggs were laid two or three hours after copulation. Generally the eggs were laid in the early hours in the morning from about 6 to 9 A.M., though sometimes eggs were laid at other hours of the day as also observed in the present investigation. During oviposition, as the eggs were laid in succession and were sticky in the fresh condition. Development evidently begins as soon as the eggs were laid. Apparently all the eggs were fertilized and were capable of development under favorable conditions and no sterile eggs were observed in control group in the present investigation. The gelatinous secretion was large in amount in treated groups in comparison to the control groups and the amount was much more secreted in docetaxel treated groups, more probably indicated hypersecretion due to toxicity. Fresh egg masses laid by the snails of F₀ generation were introduced to different concentrations of docetaxel drugs and data was recorded and summarized in Table No. Cleavage is spiral in the present investigation. Cleavage begins about 2^{1/2} to 3 hours after the eggs masses were laid in the control group of *Lymnaea* but it started after 5±2 hours and 4±2 hours and after 5±2 hours and 6±2 hours after treatment with docetaxel in *Lymnaea stagnalis*. The dose and duration of treatment dependent increase in the duration of cleavage has been observed in the experimental snails of *Lymnaea stagnalis*. It was observed that blastula period was increased in docetaxel treatment. In the present study the gastrulation period increased by 4±2 hours in docetaxel. This stage was found to be more susceptible as mortality occurred in the later gastrula stages. After organogenesis and morphogenesis specific two larvae have been formed. These two larval forms are trochophore and veliger larvae during development of these experimental snails *Lymnaea stagnalis*. In the present investigation in *Lymnaea spp.* the trochophore larval period was prolonged by 5±2 hours in docetaxel. The veliger larval period was increased by 7±2 hours in docetaxel treatment. High percentage of mortality was observed in this developmental stage but lesser than trochophore larval stage. Though after full development in treated groups some young larvae were weakened so that they were unable to break the egg capsule and died due to starvation and that was the potent cause of the higher percentage of mortality during hatching of young snails. In the present investigation it could be concluded that the increase in developmental period was dose and duration of treatment dependent. High percentage of mortality and low percentage of fecundity was achieved by the treatment of different types of docetaxel suggests that these toxicants are able to control the population density of these pestiferous snails while inhibiting their development at any stage. In the present investigation egg laying was observed throughout the year in *Lymnaea stagnalis* and it was observed that egg masses were laid in large number from July to October as also reported [3, 7]. In the present investigation it was observed that the egg masses swelled and turned somewhat viscous in treated groups. Shrinkage of egg masses was observed sooner and more clearly after alkaloid treatment. Their colour changed from dark yellow to whitish owing to the dissolution of yolky

material within the egg capsules. The contours of the egg capsules were more irregular in egg masses treated with docetaxel as also reported [7, 8, 9, 10, 11, 12, 13, 14, 15]. In the present investigation it was observed that in the control group the development evidently begins as soon as the eggs laid. Apparently all the egg capsules were fertilized and capable of development under favorable conditions. No sterile eggs were observed in Prosobranch gastropods, *Pila globosa* and *Lymnaea stagnalis* respectively [16, 17, 18, 19]. Immediately after gastrulation, the blastopore elongates in the anterior direction and lastly the anterior end of the slit closes giving rise to the stomodaeum as mouth near the points of its closure. The mesoderm originates from two mesodermal teloblasts, which are derived from the daughter cells of 4d as reported [20] as also observed in the present investigation in *Lymnaea stagnalis* and *Lymnaea acuminata* as also observed in some pulmonates [7, 8, 9, 19, 21, 22]. It was observed in control and experimental groups of *Lymnaea* spp. that these larval stages were developed within the egg capsules as also observed in some Gastropods [23] and in *Pila globosa* [24]. In the present investigation high percentage of mortality in larval stage of both spp. of *Lymnaea* was observed at the time of torsion as also observed in *Lymnaea stagnalis* after nuvan and baygon administration and in *Lymnaea stagnalis* after antitubulin drug treatment respectively [22, 25, 26,] while in the present investigation larval development arrest has been observed in large number of egg capsules in *Lymnaea stagnalis* due to the intoxication of docetaxel. In the present investigation in *Lymnaea stagnalis* it was observed that the snails developed from the treated egg masses had thin and transparent shell due to decalcification as also observed in *Lymnaea stagnalis* after nuvan and methyl parathion exposure [27], after thiourea and DDT application in *Gyraulus convexiusculus* [28], after treatment with some pesticides and *Lymnaea* spp. after treatment with some pesticides [21], in *Lymnaea* spp. and *Gyraulus* spp. after treatment with some pesticides [8, 9] and in *Lymnaea* spp. after treatment with antitubulin drug [3].

4. CONCLUSION

It could be concluded from the present investigation that the docetaxel used for the treatment, were able to arrest the development at any stage, in which larval stages were most susceptible stage. The anomalies of the organs were resulted into development of small sized, malformed young snails formed.

5. ACKNOWLEDGEMENT

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6. CONFLICT OF INTEREST

Authors have no conflicts of interests.

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