Journal homepage:https://www.environcj.in/

Environment Conservation Journal

ISSN 0972-3099 (Print) 2278-5124 (Online)

Molluskicidal activity of synthetic organic compounds (on the foot of *Achatina fulica* (Bowdich)

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ARTICLE INFO

Received : 29 December 2023
Revised : 09 February 2024
Accepted : 24 March 2024

Available online: 18 April 2024

Key Words:
Achatina fulica foot
Histopathology
Molluskicide
Synthetic organic compound

ABSTRACT

Terrestrial snails are economically destructive agricultural pests that cause great damage to vegetables and crops. The present study was performed to evaluate the toxicity of synthetic organic derivatives and subsequent histopathological alterations in the feet of *Achatina fulica* (Bowdich) after exposure to (E)-1-(4-bromophenyl)-3-(4-methylthiazole-5-yl) prop-2-en-1. Accordingly, the LC50 of the synthetic organic derivative was determined to be 1.4206±0.0981. These chemicals can easily penetrate the body of mollusks either by feeding or crawling on the surface, and the foot is the first point of contact. Histopathological and ultrastructural alterations were observed in the foot of *Achatina fulica* (Bowdich), as these organs play important roles in locomotion and mucus secretion. These alterations include degeneration of the normal structure of the cell by rupturing of the outer protecting membrane, and a large gap is created between the cells. The longitudinal muscle band is destroyed by increasing the space. These results are important from an economic point of view since these synthetic chemical compounds were used as molluskicdes for the control of *Achatina fulica* (Bowdich), which is an agricultural pest.

Introduction

Achatina fulica (Bowdich), which is also called the giant African snail due to its large size, is native to East Africa. It is an invasive, exotic species that was introduced in many countries as a polyphagous pest (Raut & Baker, 2002). It feeds nearly 500 different types of plants (Capinera,2011). It is the most popular pest species from ecological, economic, and medical points of view (Mead,1979; Raut & Ghose,1984; Srivastava,1984; Raut & Barker, 2002; Fontinilla,2007). *Achatina fulica* is a nocturnal species, and its feeding, locomotory and reproductive activities are greatest at night (Ademolu *et al*, 2010). It is a hermaphrodite organism, but to fertilize its eggs, snails need to mate with each other. The eggs hatch within 10-12 days (Lange, 1950). *Achatina fulica* (Bowdich) accepts food both qualitatively and quantitatively and eats a wide range of vegetables and crops (Raut & Barker,2002). The World Conservation Union (IUCN-International Union of Conservation of Natura and Natural Resources) listed *Achatina fulica* as one of 100 invasive species in the world (Lowe *et al*., 2000). It is an omnivorous, carpophagus feeder that can even scavenge and sometimes eat its own species. The requirement for food is nonspecific, which allows them to be successfully adapted to new areas. The fecal matter of *Achatina fulica* produces an undesirable smell, and its stool contaminates plants, which prevents the feeding of plants by other organisms (El-okda,1984; Kassab *et al.,* 1964). Therefore, large losses of vegetables, cash crops, ornamental plants, seed oil, pulses, and forest trees occur (El- Deeb *et al.,* 1999; El-okda,1979,1981, Lokma, 2007; Shahaway *et al.,* 2008). Several methods and strategies have been used for controlling snails. The easiest and simplest method is to collect and destroy egg masses and snails, but this method is time-consuming, very expensive and

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requires considerable manpower for many days (Mahrous *et al*., 2002). Biological control is one of the most ideal methods for controlling pests, but research on this topic has not been conducted. Therefore, ultimately, we have to use chemical compounds. The purpose of the present study was to assess the histopathological alterations in the feet of *Achatina fulica* (Bowdich) exposed to the synthetic organic compound (E)-1-(4-bromophenyl)-3-(4-methylthiazole-5-yl) prop-2-en-1 on adult snails as a part of a control measure, and the foot is an organ of locomotion.

Material and Methods

Experimental snail, laboratory culture and acclimatization

Adult *Achatina fulica* were collected from infested agricultural fields of paddy, cabbage, papaya, and groundnut early in the morning and late in the evening from the Niphad, Dindori, Devlali camp, etc., of the Nashik District during the monsoon season, as this period is favorable for their growth and reproduction. The snails were brought to the laboratory and acclimatized in a glass aquarium to simulate natural conditions. The aquarium was covered with a mosquito net on top containing approximately 4 cm of moist soil, providing the aquarium with the natural habitat of *Achatina fulica* (Bowdich). Water was sprinkled at regular intervals to maintain humidity, and snails were fed with green leaves of cabbage, coriander and papaya leaves.

Synthesis of chemical compounds

A series of **(**E)-1-(4-fluorophenyl)-3-(4-methyl thiazol-5-yl) prop-2-en-1-ones were synthesized in the research laboratory. Reagents, starting materials, and solvents were purchased from common commercial suppliers. The melting points of the synthesized compounds were determined by the open capillary method on a Veego digital melting point apparatus. The purity of the compounds was checked by TLC using silica gel-G (Merck). (E)-1-(4-Fluorophenyl)-3-(4-methylthiazol-5-yl) prop-2-en-1-one was prepared from the starting material 4-methylthiazole-5-carbaldehyde (1 mol) with 4-fluoroacetophenone (1 mol) and NaOH (1 mol) as a base in the presence of ethanol (5 ml) as the solvent. This reaction mixture was heated for 1 hr. under moderate reflux. The response was tested by TLC. After completion of the reaction, the reaction mixture was poured into ice-cold water, and the solid crude material was filtered and recrystallized in ethanol as the solvent. (E)-1-(4-Fluorophenyl)-3-(4-methylthiazol-5-yl) prop-2-en-1-one has the molecular formula $C_{13}H_{10}FNOS$ (mol. wt. 247.29, m.pt.). 210-212 $^{\circ C}$ and is brown in color (Figure 1).

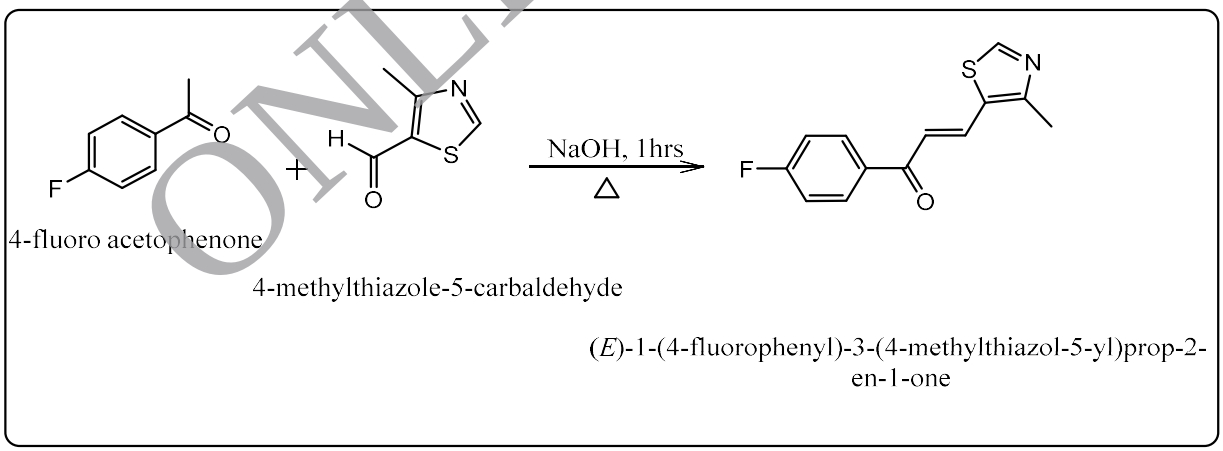

Fig 1: Final product (E)-1-(4-fluorophenyl)-3-(4-methylthiazol-5-yl) prop-2-en-1-one

Experimental Bioassay

For the present study, healthy, active adult snails were selected for toxicity evaluation. The snails were divided into five groups containing 10 snails in each group and were exposed to different concentrations of 10 ppm, 20 ppm, 30 ppm, 40 ppm and 50 ppm for 96 hrs for this experimental work. At

50 ppm, the LC_{50} of the synthetic organic derivative was 1.4206±0.0981. After this, the animals were treated with 50 ppm for 96 hrs, and many behavioral changes were observed, followed by histo pathological alterations in the foot of *Achatina fulica* (Bowdich).

Histomorphology of the normal foot of *Achatina fulica*

The foot is one of the most important muscular organs involved in locomotion. Histologically, it shows the presence of the outermost cuticular layer, which is protective in function. Below this layer, columnar epithelial cells with nuclei were present. The modified unicellular glands are formed from columnar epithelial cells that open through the cuticular layer exterior to the foot surface, which secretes mucus only. Inner to this, a major portion of the foot is occupied by this muscle fiber and the longitudinal and oblique muscle fibers, which form a meshwork with little space, as shown in (Fig-2). The foot in *Achatina fulica* plays an important role in locomotion and mucus secretion, and after treatment with synthetic organic derivatives, the snail becomes lethargic, sluggish, secretes large amounts of mucus and becomes paralyzed (M. Vijaya *et al*., 2013).

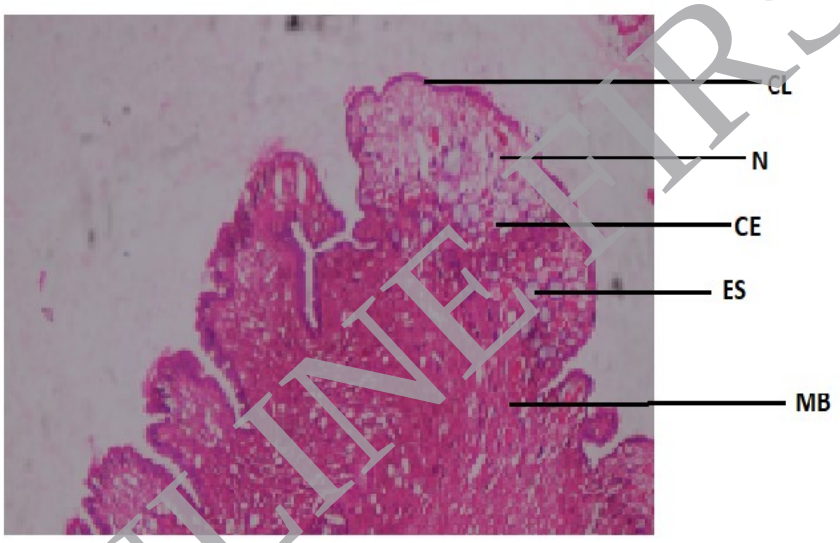

Fig 2: T.S. passing through the Foot of *Achatina fulica* (10X)

CL-cuticular layer, N-nucleus, CE-columnar epithelium, ES-empty space, MB-muscle band

Histomorphology of the treated foot of *Achatina fulica*

Due to treatment with the synthetic organic chemical (E)-1-(4-bromophenyl)-3-(4-ethylthiazole -5-yl) prop-2-en-1, a derivative of the thiazole group, which belongs to a halogen family and acts as a molluskicide for controlling snails, causes histomorphological changes in the feet of *Achatina fulica*. In various places, microhistomorphological alterations were observed in the foot of *Achatina fulica*. The protective membrane was ruptured, and a large gap was created between adjacent cells. The tissue was damaged, and the cells shrank. The normal arrangement of longitudinal and oblique muscles was destroyed at various places that created gaps between the cells (Fig 3). The disturbance of this pattern was observed due to the action of synthetic organic derivatives, which ultimately disturb the normal function of the foot and secrete a large amount of mucus, after which the cells become degenerated, and the behavior of *Achatina fulica* becomes sluggish, motionless, and lethargic. Various pesticides and heavy metals (Cu and Pb) have been used to control gastropod species, but these chemicals are harmful to the environment (Ahmed *et al*., 2016; Otitoloju A.A. *et al*., 2009).

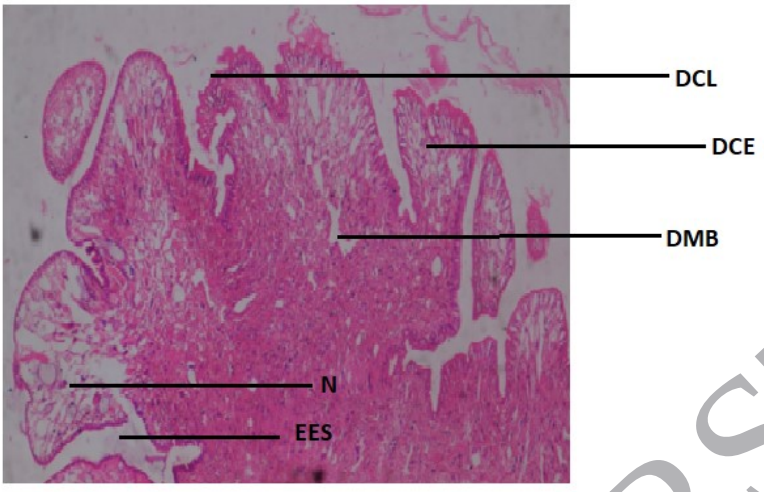

Fig 3: T.S. passing through the treated foot of *Achatina fulica* (10X)

DCL- damaged cuticular layer, N-Nucleus, damaged columnar epithelium, EES- enlarged empty space, DMB-damaged muscle band

Therefore, ultimately, we used this synthetic chemical compound as a molluskicide to control *Achatina fulica* (Bowdich) as a part of the control measures.

Results and Discussion

In the present investigation, *Achatina fulica* was exposed to different concentrations (10 ppm, 20 ppm, 30 ppm, 40 ppm and 50 ppm) of the chemical compound (E)-1-(4-bromophenyl)-3 (4-methyl thiazole -5-yl) prop-2-en-1, and at 50 ppm, an LC_{50} (1.4206±0.0981) was found. This finding is also similar to that of Kanawade S *et al*. (2011), who synthesized new thiophenedicarboxamide 2a-c and dicyanothiopheneacetamide 3a-c derivatives for use as molluskicides against *Indoplanorbis exustus* and reported LC_{50} =0.6043 ppm and LC_{50} =0.6511 ppm, respectively; this finding is supported by Toche *et al*. (2009). Both of these findings of synthetic organic derivatives were used to control the species of gastropods. Due to the effects of these chemicals, *Achatina fulica* moves slowly, becomes inactive, is lethargic, and secretes a large amount of mucus, and snails stop feeding; it is assumed that this chemical not only irritates the cells but also affects the digestive gland of *Achatina fulica*, through which the snails stop feeding. The cuticular layer is disrupted by these chemicals, creating space between the cells. Snails move slowly because of damage to the arrangement of muscle fibers in the foot, which leaves a void in the foot area. This result is also consistent with that of Soomro *et al*. (2005), who found that injecting cercaria into *Lymenea acuminata* caused damage to the cuticular layer and a decrease in mucus. The cuticular epithelium layer and the muscle layer organization were also altered by this chemical, resulting in numerous degenerations of the cells in the foot region. This finding is also similar to that of Aukkanimart *et al*. (2013), who reported that the extracts of Camellia seeds and Mangosteen also affect the pattern of muscle fibers present in the foot region, which damages epithelial cells in *Bithyxia siamensis* by creating a gap between epithelial cells and connective tissue. These results are supported by Viard *et al*. (2004). Soomro *et al.* (2005) reported damage to the cuticular layer and shrinkage of mucus-secreting cells in *Lymnaea acuminata* infected with Cercaria, similar to the present results, where this synthetic chemical compound damages the outer cuticular layer and muscle band and is responsible for the secretion of mucus. Venkata M *et al*. (2008) observed the effect of cadmium on the foot of the freshwater mussel *Lamellidens marginalis* (Lam), which disorganizes the outer membrane of the foot, resulting in necrosis of the cell. This result is also similar to the present result,

where after 96 hrs of exposure to the given chemicals, the muscle fibers became distorted by creating a large gap between the cells. This result is also similar to that of Balamurugan *et al*. (2021), where the muscle fibers became damaged in *Lamellidens marginalis* due to the sublethal concentration of oil effluent and epithelial cells being damaged, disorganizing the muscle band from its normal position and increasing the space between the cells. In the present study, the muscle fibers became thinner, and pyknosis of the nucleus and heavy vacuolization followed by muscular atrophy and complete destruction of the muscular structure occurred; these findings coincide with those of Chetty *et al.* (1988). Gaber *et al.* (2022) reported that exposure to methomyl for 96 hrs caused many histomorphological changes in the feet of *Monacha* cartusiana snails. There is distortion of the muscle fiber with the outer layer and necrosis of the mucus-secreting cells, and there is a complete deformity of the outer layer of the foot, which results in the formation of vacuoles in muscle fibers. This result is similar to that of Abdel *et al*. (2020). Further investigations of this synthetic organic compound to increase its toxicity, stability and selectivity should be performed, and its efficacy should be manipulated to characterize its molluskicide activity, which not only causes histopathological changes but also affects the functional properties of *Achatina fulica* (Bowdich).

Conclusion

According to the results of this investigation, (E)-1-(4-bromophenyl)-3-(4-methylthiazole-5-yl) prop-2-en-1 does not kill *Achatina fulica;* instead, it causes it to become lethargic, sluggish, and dehydrated, which causes it to die and cause histomorphic alterations in the foot. Recent developments in the synthesis of bioactive molecules comprising thiazoles and their structural activity have led to a wide range of pharmacological effects. To attempt and stop the massive loss of agricultural products from the Nashik area, derivatives of synthetic organic compounds may be employed as molluskicides to decrease the population of *Achatina fulica*, a notorious pest.

Acknowledgment

The authors are grateful to Principal Dr B.S. Jagdale Loknete Vyankatrao Hiray Arts, Science & Commerce College, Panchavati Nashik-3 for providing all necessary research laboratory facilities during the tenure of this research.

Conflict of interest

The authors declare that they have no conflicts of interest

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