

# Distinguish Behavioral Characteristics of *Diasemiopsis Ramburialis* (Duponchel, 1833) and *Nomophila noctuella* (Denis & Schiffermuller, 1775) (Lepidop-Tera: Crambidae) on *Azolla* spp.<sup>†</sup>

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**Abstract:** It is generally accepted that biological invasions have major ecosystem impacts and create new types of interaction between local and exotic species. *Azolla* spp. invasion caused different interactions between local and exotic species in the northern region of Iran. *Diasemiopsis ramburialis* and *Nomophila noctuella* are two spilmelinae moths found feeding on *Azolla* spp. in these regions. They have overlapping feeding periods on *Azolla* spp. and immature stages would be confused easily. *N. noctuella* was recorded as a polyphagous and pest species while *D. ramburialis* had no other reported host plant beside *Azolla* spp., therefore, distinguishing these species from each other was so important for anyone who was working on *Azolla* spp. management in these Areas. Behavioral characteristics of both species were studied for five years in the laboratory and natural habitats. The results indicated that despite the similarities between immature stages, there are some distinctive behavioral characteristics that would be useful for primary identification of each species in the field. *D. ramburialis* larvae constructed well-shaped and strong shelters, and produced more silken webs and remained in the shelter most of the time. In addition pupation occurs near the feeding site in this species. *N. noctuella* larvae have wandering behavior and prefer to move in margins, therefore in this species, pupation occurs far from the feeding site. These results indicated that *D. ramburialis* is more adapted to feed *Azolla* spp. and *Azolla* spp. are main host plants for this species while *N. noctuella* chose them randomly.

**Keywords:** Spilmelinae; *Azolla* spp., Behavioral characteristic

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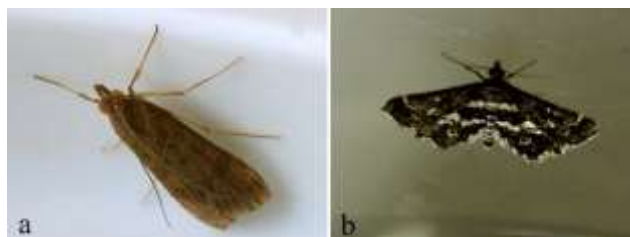


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## 1. Introduction

Spilmelinae (Guenée, 1854) is the largest subfamily of Crambidae; consist of 4026 recorded species from 327 genera [1]. In recent years, it has been indicated that *Diasemiopsis ramburialis* (Duponchel, 1833) and *Nomophila noctuella* (Denis and Schiffermuller, 1775) (Figure 1), two snout moths from spilmelinae, feed on *Azolla* spp. in the northern region of Iran and have overlapping feeding periods (Table 1) [2,3]. *Diasemiopsis* was described by Munroe (1957) with *Hydrocampa ramburialis* (Duponchel, 1833) as type species. Only one other species, *Diasemiopsis leodocusalis* (Walker, 1859), is currently assigned to this genus [4]. *Diasemiopsis ramburialis* is a cosmopolitan species [5] and *Azolla* spp. are known as the first host plants of this moth [2]. *Nomophila* was described by Hübner (1825) with *Pyralis hybridalis* (Hübner, 1796) as type species. 17 identified species were assigned to this genus [4,6] and *N. noctuella* is mainly separated in the old word [7]. This moth is

known as a polyphagous species associated with numerous host plants and a pest for some crops [8,9,5]



**Figure 1.** Adults (a: *Nomophila noctuella*, b: *Diasemiopsis ramburialis*).

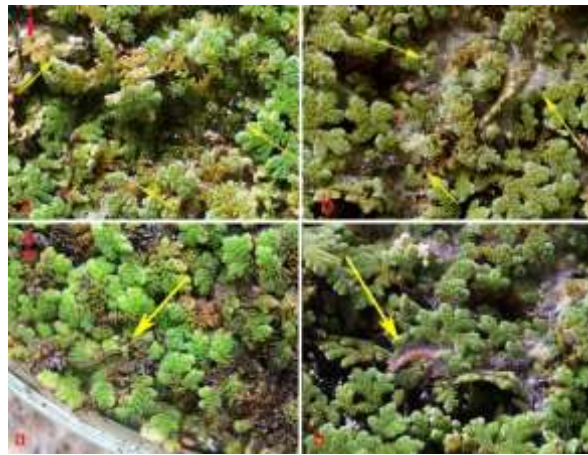
*Azolla* spp. are considered as alien species in Iran. Over the last few years, this obnoxious weed has invaded many wetlands in the northern regions of Iran [10–12,2] and due to eradication of native plants, the insects that feed on these plants either had to use new plants as a food resource or die. Our studies indicated, that *D. ramburialis* and *N. noctuella* feeds on *Azolla* spp. in northern region of Iran and have overlapping feeding periods. The larvae of both species are non-aquatic, somewhat similar morphologically and have shelter making behavior with almost the same patterns. As mentioned before, *N. noctuella* is recorded as a polyphagous species and a pest for some crops, while, for *D. ramburialis*, *Azolla* spp are only recorded host plants. The specific objective of this paper is to compare behavioral characteristics of *D. ramburialis* and *N. noctuella* on *Azolla* spp. in order to recognize each species during their overlapping feeding period.

## 2. Material and methods

Laboratory colonies were established by collecting larvae from *Azolla* spp. located on waterways and experimental rice fields at the Rice Research Institute of Iran (RRII) (N37°12'22.2", E49°38'40.7", 17 alt.) from September to November in 2013 to 2018. we used disposable plastic containers (18×13 cm, diameter by height) as mating chamber and plastic boxes (24 × 17 × 10 cm, length by width by height) as rearing chambers, each covered with transparent cellophane with small holes in the cellophane for ventilation. In the mating chamber there were three small containers (6 × 5 cm diameter by height) each filled with 50 cc of distilled water in 18cm diameter chambers. Two containers were filled with 10gr *Azolla* spp. and in the third container a sugar cube dissolved and filled with tissue papers in order to provide adults with food supplements in case that they need food. For rearing chambers boxes were filled with 500cc distilled water and 200gr *Azolla* spp.. All chambers were kept in 25–27 °C and 16: 8h (L: D) photoperiod. In mating chambers a pair, a male and female, were released in each chamber upon emergence. For rearing experiments first or second instar larvae were released in rearing chambers (20 individuals in each box). Water and food materials were renewed once in three days.

## 3. Results

*Diasemiopsis ramburialis* and *N. noctuella* larvae feed on *Azolla* spp. leaves and cause two types of damages. Direct damages were caused by larvae feeding activity. Since larvae of both species are non-aquatic, newly hatched larvae built shelter by binding *Azolla* spp. leaves together. In the laboratory, *D. ramburialis* larvae produced more webs compared with *N. noctuella* (Figure 2).



**Figure 2.** amount of produced webs (1) and larvae pathway(2) (a): *Nomophila noctuella*, (b): *Diasemiopsis ramburialis*).

Behavioral observations of both species indicated that *D. ramburialis* larvae move in corridors that made by silken webs among the *Azolla* spp. leaves. Therefore the larvae pathway on *Azolla* spp. in this species were distinctive (Figure 2b1,b2). They rarely left these corridors and pupation always occurred in these shelters (Figure 3b1,b2). In *N. noctuella* the larval pathway was under the *Azolla* spp. leaves and larvae just bind over leaves to make a union layer in order to move under the leaves. As the result, they produced fewer amounts of the webs and the larvae pathway were not as clear as *D. ramburialis* (Figure 2). In addition, *N. noctuella* larvae wandered on *Azolla* spp. regularly and leave frass near the pathways (figure 2a1) and due to wandering behavior of larvae in *N. noctuella* pupation occurred in places that were somewhat far from feeding sites (Figure 3a1,a2).



**Figure 3.** Pupation site (1) and pupa (2) (a): *Nomophila noctuella*, (b): *Diasemiopsis ramburialis*).

More behavioral studies indicated *N. noctuella* larvae prefer to move in the feeding site margins (Figure 4). Indirect damages on *Azolla* spp. were caused by larvae feeding damage and frass that facilitated the infection of *Azolla* spp. with bacteria and fungi.



**Figure 4.** *Nomophila noctuella* larvae pathway in margins.

#### 4. Discussion

It is generally accepted that biological invasions have major ecosystem impacts [13& 14]. After first introduction in 1986, *Azolla* spp. covered some of the aquatic ecosystems of the northern region of Iran [11] and due to unbalanced and disproportionate environmental conditions, many of migratory or native species consumed *Azolla* spp. Studies showed that *D. ramburialis* and *N. noctuella*, feed on *Azolla* spp. in the northern region of Iran. These species have overlapping activity periods and due to shelter making behavior they almost have the same feeding pattern. However, some of specific behavioral characteristics of both species could be useful for recognizing them. *Diasemiopsis ramburialis* larvae made well shaped and strong shelter on *Azolla* spp.. Considering non-aquatic larvae of this species, making a strong shelter on the aquatic host plant provide more protection for the larvae. Therefore making strong shelters would be an adaptive behavior in response to feed on an aquatic host plants. In addition, non-wandering behavior of the larvae which led to pupation in concealed sites would be another beneficial adaptive behavior that protects the larvae and pupa from predators in aquatic ecosystems. Insects adapt with their host plant and mediate their feeding behavior and strategy in response to host plant chemical components and morphological characteristics [15,16]. Therefore these results indicate that *D. ramburialis* is adapted to feed on the *Azolla* spp. perfectly.

Unlike the *D. ramburialis*, *N. noctuella* is a polyphagous species [5]. This moth can feed on many host plants and its feeding behavior on *Azolla* spp. indicates that it is not so adapted to feed on the aquatic plants. Despite the shelter making behavior of larvae, shelters on *Azolla* spp. were not so strong. Therefore the larvae preferred to move near the margins. In addition wandering behavior of the larvae exposed larvae and pupa to aquatic predators easily. However due to polyphagous behavior of the larvae and high abundance of *Azolla* spp. this species chose to feed on the new host plant. Beside *D. ramburialis* and *N. noctuella*, there are other fern-feeding group of moths from Heliothelinae (Amsel, 1961) in crambidae [17]. Therefore, feeding on *Azolla* spp. in *N. noctuella* could be based on phylogenetically connections as well.

#### 5. Conclusion

*Azolla* spp. invasion caused different interactions between local and exotic species in the northern region of Iran. *Diasemiopsis ramburialis* and *N. noctuella* are two spilomelinae snout moths found feeding on *Azolla* spp. in the northern region of Iran. There was no record of these species or their biology on *Azolla* spp. worldwide. We studied each species life history on *Azolla* spp. before. However further studies indicated that they have overlapping feeding periods on *Azolla* spp. and would be confused easily. Since *N. noctuella* was recorded as a polyphagous and pest species it was important to be distinguished from *D. ramburialis*. Behavioral studies indicated that compared with *N. noctuella*, *D. ramburialis* is more adapted to feed on *Azolla* spp. Therefore, *Azolla* spp. could be main host plants for this species. Much more studies need to confirm this hypothesis but it is sure that *D. ramburialis* and *N. noctuella* would be important biotic resistance factors for *Azolla* spp. in the northern region of Iran.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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