Samea multiplicalis (Guenée, 1854) (Lepidoptera, Pyralidae): a potential agent in the biological control of *Salvinia molesta* DS Mitchell (Salvineaceae)

Samea multiplicalis (Guenée, 1854) (Lepidoptera, Pyralidae): um agente potencial no controle biológico de *Salvinia molesta* DS Mitchell (Salvineaceae)

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Abstract: The interactions between *Samea multiplicalis* (Guenée, 1854) (Lepidoptera, Pyralidae) and *Salvinia molesta* DS Mitchell (Salvineaceae) were followed during a year in an urban lake on the karstic plateau of Lagoa Santa, Minas Gerais, south-east Brazil. Field data and results from experiments under laboratory conditions were used to test the hypothesis that larvae of *Samea multiplicalis* (Lepidoptera, Pyralidae) could be used as controlling agents for *Salvinia molesta* DS Mitchell (Salvineaceae). Results demonstrated that, although capable of consuming up to 12% of the annual production of *S. molesta* DS Mitchell (Salvineaceae), the larvae of *S. multiplicalis* (Lepidoptera, Pyralidae) can only be used as a controlling agent under certain conditions in which *Cyrtobagous salvineae* (Calder and Sands) (Coleoptera, Curculionidae), a reported efficient controlling agent, is not so effective. The results also suggested that not only *S. multiplicalis* (Lepidoptera, Pyralidae) but also association with the variations in water temperature and ortho-phosphate levels is the major determinants for the growth rates of *S. molesta* DS Mitchell (Salvineaceae) in studied lake.

Keywords: insects fauna, aquatic macrophytes, feeding preference, Lake Olhos d'Água.

Resumo: As interações entre *Samea multiplicalis* (Guenée, 1854) (Lepidoptera, Pyralidae) e *Salvinia molesta* DS Mitchell (Salvineaceae) foram seguidas durante um ano em um lago urbano no karst do planalto de Lagoa Santa, Minas Gerais, no sudeste do Brasil. Dados obtidos no campo e resultados de experimentos, em laboratório, foram utilizados para testar a hipótese de que larvas de *Samea multiplicalis* (Lepidoptera, Pyralidae) poderiam ser utilizadas como agentes de controle para *Salvinia molesta* DS Mitchell (Salvineaceae). Os resultados demonstraram que, embora capazes de consumir até 12% da produção anual de *S. molesta* DS Mitchell (Salvineaceae), as larvas de *S. multiplicalis* (Lepidoptera, Pyralidae) só podem ser utilizadas como agente de controle, sob certas condições nas quais *Cyrtobagous salvineae* (Calder and Sands) (Coleoptera, Curculionidae), um agente eficiente no controle de *S. molesta* DS Mitchell (Salvineaceae), não é eficaz. Os resultados também sugeriram que, não somente as densidades de *S. multiplicalis* (Lepidoptera, Pyralidae), mas também as variações na temperatura da água e orto-fosfato são os principais determinantes para as taxas de crescimento de *S. molesta* DS Mitchell (Salvineaceae) no lago estudado.

Palavras-chave: fauna de insetos, macrófitas aquáticas, preference alimentar, Lago Olhos d'Água.

1. Introduction

Salvinia molesta DS Mitchell is a free-floating aquatic pteridophyte that has created plagues in some tropical waters outside Brazil, its original distribution area. Forno and Harley (1979) appointed that *Salvinia molesta* only occur naturally at altitudes of up to 500 meters and above the Tropic of Capricorn, between 24° 05' S and 32° 05' S. After been introduced in Sri Lanka in 1939 the species quickly spread, reaching Africa, India, South East Asia and Australia (Room, 1990). *S. molesta* has been collected in the States of São Paulo, Paraná, Santa Catarina and Rio Grande do Sul (Forno and Harley, 1979).

Forno and Bourne (1984) studied a fauna of arthropods associated with *Salvinia* spp. in six localities in Brazil, reporting the occurrence of *Cyrtobagous* sp. (Coleoptera; Curculionidae), *Samea multiplicalis* (Guenée, 1854) (Lepidoptera; Pyralidae) and *Paulinia acuminata* (De Geer)

(Orthoptera; Pauliniidae) as being the three possible agents in the biological control of *Salvinia molesta*. *Cyrtobagous salviniae* (Calder and Sands), reported as an efficient agent in the biological control of *Salvinia molesta*, was introduced successfully in Australia (Room, 1990). However, according to Miller and Wilson (1989) *C. salviniae* is not efficient in cool waters, possessing little amounts of nutrients. As pointed by Pelli (1998a) nutrients is an important variable that determines the density of insects.

Room et al. (1984) observed that *Cyrtobagous* sp. (called by Room, 1990 as *C. salvineae*) had achieved control of *S. molesta* in Australia, but *Samea multiplicalis* isn't efficient, probably due a right temperatures experienced. The work of these authors was made with estimates of the population in the field, but didn't quantified the damage caused by *Samea multiplicalis*.

The present study had as objective to estimate the damage caused to *Salvinia molesta* by *Samea multiplicalis*. Complementary, it assessed the feeding preference in relation to the age of the leaves, and the growth of *Salvinia molesta* in the presence or absence of *Samea multiplicalis*.

1.1. Study area

Lake Olhos d'Água is a natural urban lake located in Lagoa Santa, Minas Gerais, (19° 44'-19° 33' S and 44° 05'-43° 50' W). The lake belongs to the karstic lakes system of the Lagoa Santa plateau, having a maximum width of 110 m, maximum length of 520 m and maximum depth of 3.5 m during the rainy season.

2. Material and Methods

2.1. Field studies

Samplings were performed in a permanent littoral station, from February 1991 until March 1992. The samplings of *Samea multiplicalis* and the estimations of the growth rates (gross and net) of *Salvinia molesta* were conducted at approximately 10 days' intervals (Pelli and Barbosa, 2007). The densities of *Samea multiplicalis* were estimated by using a special collector possessing a net of 0.25 mm size mesh and an area of 214 cm^2. In each sampling 10 random samples were collected. The estimations of the growth rates of *S. molesta* were made by the count number of leaves of least five modular units. The leaves were marked with sewing needle and line. The new leaves were counted and the rates calculated.

The samples were packed in polythene bags and fixed immediately with 10% formalin solution. In the laboratory the samples were washed (tap water) after which sub-samples were selected under stereomicroscope for examination. The sample of *S. molesta* from each sample was weight before drying at 70° C to constant weight. The dry weight was taken as a parameter for the calculation of density, expressed in number of individuals per gram of dry weight (ind/d.w.) of *Salvinia molesta*.

2.2. Laboratory work

Laboratory experiments were conducted on Petri dishes in order to characterize a possible feeding preference in relation to distinct parts of *Salvinia molesta*. "Immature" leaves (leaves located near the apical bud - less than 4 ramets) were offered to the larvae of *Samea multiplicalis* and "mature" leaves (leaves located far from the apical bud - more than 4 ramets). The number of leaves and their positions on the dishes are shown in Figure 1.

The observations were made daily in the morning, recording the number of leaves of *Salvinia molesta* eaten (at least 90%) by the larvae of *Samea multiplicalis*, after which all the leaves were replaced. Each Petri dish contained a single larva of *Samea multiplicalis*.

The first experiment was conducted using six Petri dishes and 24 observations; the second one with six Petri dishes and 28 observations while for the third experiment were used only two Petri dishes and nine observations.

The larvae of *Samea multiplicalis* were separated into three classes, named "small", "medium" and "big" as suggested by Whiteman and Room (1985) and Semple and Forno (1987). This characterization is based on the width of the head capsule, assuming with Smock (1980) that this is the best allometric dimension to characterize the larvae of Lepidoptera.

Experiments in aquaria were also conducted in order to determine the effect of the larvae of *S. multiplicalis* on the growth and standing-crop of *S. molesta*. Plants previously washed with lake water were put in six aquaria of dimensions 20 x 12 x 12 cm. Three aquaria received one "big"

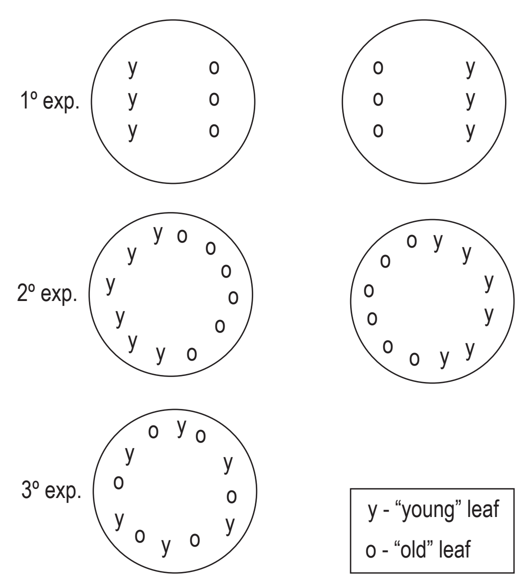

Figure 1. Experimental design used to determine the feeding preference of *Samea multiplicalis*.

larvae of *S. multiplicalis* (except in the first experiment when two larvae were put in each aquarium); the remaining three aquaria were used as controls. This experiment was repeated three times with different larvae densities of *S. multiplicalis* / *S. molesta* leaves. For the first, second and third experiments one larva of *S. multiplicalis* was used while the densities of leaves of S. molesta were, respectively 27.7, 58.3 and 70.0.

The experiments had duration of four weeks with observations at 7 days intervals. Comparisons of the net growth rates with and without *S. multiplicalis* larvae and the differences in the number of leaves at the beginning and at the end of the experiments in aquaria with and without *S. multiplicalis* larvae were conducted.

From data obtained in the field and in the experiments conducted in aquaria estimates of the rate of damage caused by *S. multiplicalis* were made. Simulations using Qbasic-DOS6 were made in order to estimate the population increase of *S. molesta* with and without *S. multiplicalis*.

The following program was assembled:

```
N1 = x:
FOR i = 1 TO z
N1 = n1 + (n1 * a) – b
PRINT "n1 = ;"; n1; ""
N1 = n1:
NEXT i
```

where:

"x" = number of initial leaves; "z" = interval (in days); "a"= rate of net increase and "b" = daily rate of herbivory.

3. Results and Discussion

According to Room (1990) *S. multiplicalis* only feeds on leaves of *S. molesta*. However, nutritional differences between "immature" and "mature" leaves are likely to affect this feeding preference. So, it is assumed that in the case of a preference for leaves showing signs of senescence (mature leaves), the effect of *S. multiplicalis* on the growth rates and standing-stock of *S. molesta* should be smaller.

The results of the experiments conducted in Petri dishes were analyzed by the test of χ^2 with the correction of Yates (Table 1). This showed that in 92% of the observations of Experiment 1, 89% of Experiment 2 and 88.9% of Experiment 3, the larvae of *S. multiplicalis* preferred to eat "immature" leaves of *S. molesta*, thus rendering an average preference for "immature" leaves in 90% of the observations. These results also suggested an important characteristic of *S. multiplicalis* as a potential controlling agent of *S. molesta*, since in preferring "immature" leaves this species is likely to be more effective in controlling the increase of the aquatic macrophyte than if showed a preference for "mature" leaves, as proposed by Room (1988).

The experiments conducted in aquaria showed that *S. multiplicalis* reduced drastically the net increase rates of *Salvinia molesta*. The results of the *t*-test were that in the first experiment $t = 7.74$ with $p < 0.001$; in the second experiment $t = 5.11$ with $p < 0.001$ and in the third experiment $t = 5.23$ with $p < 0.001$.

The effect of the larvae of *S. multiplicalis* on the standing-stock of *S. molesta* was also recorded by comparing the differences in the number of leaves of *S. molesta* recorded at the beginning and at the end of the aquarium experiments. The results are shown in Table 2.

These showed that, at the densities used, *S. multiplicalis* could cause a significant reduction in the net increase rate and in the standing-stock of *S. molesta*.

Considering that: i) a ramet (or 2 leaves + submerged organ) weights 0.0382428 grams; ii) a "big" larva of *S. multiplicalis* eats an average 1.181 leaves of *S. molesta* per day; iii) the net growth rate of *S. molesta* in the lake, and iv) when the "big" larvae occurred during the study period, it was possible to estimate that during the period 14/II/1991 until 13/XII/1992, *S. multiplicalis* ate approximately 12% of the biomass of *S. molesta* present in the lake, thus being potentially a controlling agent for the growth of this macrophyte.

Such considerations are hypothetically showed in Table 3.

The recorded growth rates for *S. molesta* and *S. multiplicalis* densities are shown in Figure 2. Although the curves show a strong negative correlation between the variables, we believe that not only *S. multiplicalis* but also other factor, notably variations in the water temperature and ortho-phosphate concentrations was major determinants for the growth rates of *S. molesta*.

Table 1. χ^2 values and significance level for the experiments conducted in Petri dishes.

Experiment	χ^2	Significance level
1	15.04	p < 0.01
2	15.75	p < 0.01
3	4.00	p < 0.05

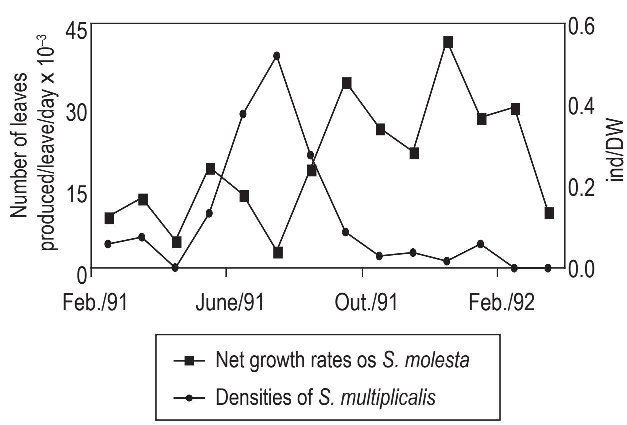

Figure 2. Seasonal variation of the net growth rates of *Salvinia molesta* and densities of *Samea multiplicalis* in lake Olhos d'Água, Minas Gerais.

Table 2. Average number of leaves of *S. molesta* eaten per day by one larva of *S. multiplicalis* in a period of 4 weeks, it is also shown the numbers of larva by leaves, reduction (%) in the number of leaves, the *t*-test values and the level of significance.

Experiment	N° of leaves/larva	N° of eaten leaves/day	%reduction/4 weeks	*t*-test	Significance level
1	27.7	1.115	33	9.51	$p < 0.001$
2	58.3	1.030	31	9.08	$p < 0.001$
3	70.0	1.397	35	9.53	$p < 0.001$

Table 3. Hypothetical number of *S. molesta* leaves and projection to the end of one year.

Date	Number of leaves of *S. molesta*	
	with *S. multiplicalis*	without *S. multiplicalis*
14/02/1991	100,000	100,000
28/01/1992	47,058	53,266

S. multiplicalis has been considered as a second agent in the biological control of *S. molesta* (Forno and Bourne, 1984; Room, 1990; Semple and Forno, 1987; Taylor, 1988; Taylor and Forno, 1987). These authors indicated the potential of *S. multiplicalis* as a controlling agent, but the present study is the first quantification of the damage caused by this species.

In this study 26 larvae of *S. multiplicalis* were maintained under laboratory conditions, from which 50% were parasitised, the others being apparently healthy emerging adults. Knopf and Habeck (1976) found 52% parasitism of larvae of *S. multiplicalis*, whereas Semple and Forno (1987) reported only 22% of parasitised larvae. From the 128 larvae collected in this study, 11% were classified as "big", whereas Semple and Forno (1987) recorded that 24% of the larvae collected in 14 different farms in Australia were classified in this category, from which 0.3% were parasitised.

Comparing the data of Semple and Forno (1987) with the data obtained in this study we conclude that the bigger the index of parasitism the smaller the proportion of big larvae. In environments with a high percentage of parasitism (as reported by Knoph and Habeck (1976)) as well as in lake Olhos d'Água, is possible that the reduction of the parasitism rates promotes an increase in the size of individuals in the population, thus altering the distribution by age, favoring the big larvae.

S. multiplicalis in Olhos d'Água lake consumed more than 12% of the annual production of *S. molesta*; a figure that can be considered relatively low if compared with the population explosion of *S. molesta*.

The density and extent of infestations of *S. molesta* in Olhos d'Água lake are variable but is generally much less than outside their original distribution, like Australia before the biological control. These differences are almost certainly due to attack by natural enemies in original area of distribution.

In lake Olhos d'Água Pelli and Barbosa (1998b) related 114 species of insects associated with *S. molesta*, and only 13 species can damage this aquatic macrophyte. It indict that Olhos d'Água lake can be included in the original area of distribution of *S. molesta* and that *S. multiplicalis*, associated with others species of insects of little importance (Pelli and Barbosa, 1998), can control *S. molesta* populations to densities that do not cause damage to most economic activities.

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