

## DEVELOPMENT OF *LYCORIELLA INGENUA* AND *BRADYSIA IMPATIENS* ON DIFFERENT PHASES OF *AGARICUS* COMPOSTS

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### ABSTRACT

Fungus gnats are small, dark colored, mosquito like fragile insects. They can be encountered all around the world, excluding places with extreme climate. Gnats prefer habitats that are damp with high relative humidity and rich in organic debris. Most of the species are not considered to be harmful in agriculture, but some may cause serious damage, especially in forcing of vegetable or ornamental plants. In mushroom production, however, they are considered as the most destructive pests; the damage caused by them alone can result in huge loss for mushroom farmers. In the Hungarian mushroom industry, fungus gnats damage mainly white-button mushroom, oyster mushroom and pioppino productions. *Lycoriella ingenua* (DUFOR, 1839) and *Bradysia impatiens* (JOHANNSEN, 1912) are the two most commonly found fungus gnat species from the family *Sciaridae* on Hungarian mushroom farms.

We have conducted an experiment with the two aforementioned species, to find out, whether they can fully develop into adults, feeding purely on *Agaricus* compost diet. We used unspawned white button mushroom compost (phase 2) and spawned compost which has been colonized by the mycelia of *Agaricus bisporus* (phase 3). We did not conduct any experiment with the first compost phase, as it undergoes pasteurization, which eliminates any pests in it. We recorded emerging gnats only from phase 2 compost. Neither *L. ingenua*, nor *B. impatiens* could complete its development on phase 3 compost diet, furthermore not even pupae were observed in these breeding pots. From pots containing phase 2 compost, a total of 1607 adults emerged. The number of *B. impatiens* adults was 653, and 954 for *L. ingenua*. Sex ratios for the two species were different. For *B. impatiens*, the number of females was 1.36 times greater than that of males, in *L. ingenua* there were 1.7 times more males than females in total. Swarming took approximately 6 days for *B. impatiens* and 10 days for *L. ingenua*. From the experiment we can conclude that phase 3 compost, which is well interwoven with *Agaricus bisporus* mycelia is not suitable for *B. impatiens* and *L. ingenua* to complete their whole life cycle.

**Keywords:** fungus gnat, *Agaricus bisporus*, compost, mushroom cultivation

### INTRODUCTION

Insects from the family *Sciaridae* can be encountered all around the world. In nature, the sciarid flies can be found within tree trunks colonized by fungi, under bark or in manure piles or under fallen leaves as well (BINNS, 1981). Most of the species feed on soil dwelling fungi and are not deemed to be harmful in agriculture (MEAD AND FASULO, 2001). Nevertheless, there are some which can damage agriculturally important plants such as ornamentals and vegetables (HUNGERFORD, 1916; MEAD AND FASULO, 2001). In mushroom cultivation, the fungus gnats cause their damage directly and indirectly. The direct damage is attributed to the larvae, which feed on the compost, thus taking away nutrient from the mushroom mycelia. Additionally the excrement of larvae inhibits the growth of mycelia. In some cases, the larvae can damage the forming pinheads as well, and may burrow themselves in the stalk of mushrooms (LEWANDOWSKI ET AL., 2004). The indirect damage is caused by the adult insects, which act as vectors for fungal pathogens of edible mushrooms (SHAMSHAD, 2010). Every segment of the mushroom industry provides

an ideal habitat for fungus gnats. During compost production, the different phases are transported by the help of conveyer belts, wheel loaders and trucks, from which some compost unintentionally falls off, thus creating potential food source for gnats where they can develop undisturbed. Mushroom production requires manual labor, that inevitably leads to some compost ending up on the floor and remaining uncollected. These conditions are always available, since mushroom production is continuous throughout the calendar year.

## MATERIAL AND METHOD

### Fungus gnat species

During our experiment, we worked with the two most commonly found sciarids in Hungarian mushroom production, *Lycoriella ingenua* and *Bradysia impatiens*. *L. ingenua* is reported to be the most serious pest in mushroom production, especially in white button mushroom cultivation (MENZEL AND MOHRING, 1997), while *B. impatiens* tends to be more common in plant forcing (CLOYD, 2008) but may be present at mushroom farms as well. The morphology of the two species does not differ significantly at the first sight. Both species are small, 2-5 mm long, delicately structured, dark brownish or black colored insects, with large compound eyes and long antennae. The life cycle and environmental needs are identical for the two species, and the damage symptoms do not differ either. Identification of the two species can be done visually, they differ in the armature of the fore tibia, the male genitalia and the length of the antennal flagellomeres.

### Collection of specimens and establishing the fungus gnat cultures

The necessary insect-material was collected from a mushroom farm in Ócsa, on the date of 05.02.2018. Initially, we collected more than 500 insects, which were transported to the Department of Vegetable and Mushroom Growing of Szent István University for further studies. Because it is impossible to collect “pure” populations of single species at growers, we had to establish these pure colonies in laboratory. Mating pairs of fungus gnats were isolated from the rest of the group. After the mating process was completed, we isolated the inseminated females individually in breeding pots containing phase 2 *Agaricus* compost (one female per breeding pot). For species identification, we collected the male gnats after mating, and examined the hypopygium. Based on the outcome of this identification process, we determined which breeding pots contained *Lycoriella ingenua* and *Bradysia impatiens*. In the trial, we used phase 2 and phase 3 *Agaricus* composts, and for each of the species we have used 12 replicates. The breeding pots were created by filling the composts into plastic containers (870 ml) and covering them with plastic-fiber veils. Each of the breeding pots contained only one inseminated female. During the experiment, the pots were stored in an environmental control chamber in the absence of light, at 23 °C with the relative humidity set to 85%. We evaluated the results by recording the number of emerged adults in the breeding pots according to species and sex from the start of the swarming, until the end of it. At the end of each day, the asphyxiated adults were not released back into the breeding pots.

## RESULTS

### Number of emerged adults

We have recorded a total of 1607 adults independently from sex or species, which meant an average of 66.9 adults per breeding pot. The mortality rate was between 33.1% and

55.4% compared to the average number of eggs laid by the isolated females (100-150). We have recorded zero number of *Lycoriella ingenua* or *Bradysia impatiens* on phase 3 Agaricus compost in either experimental trials. From the start of the experiment, we have collected a total of 954 *L. ingenua* and 653 *B. impatiens* adults on phase 2 Agaricus compost (Figure 1). This means that the average number of eggs laid was 79.5 for *L. ingenua* and 54.4 for *B. impatiens*.

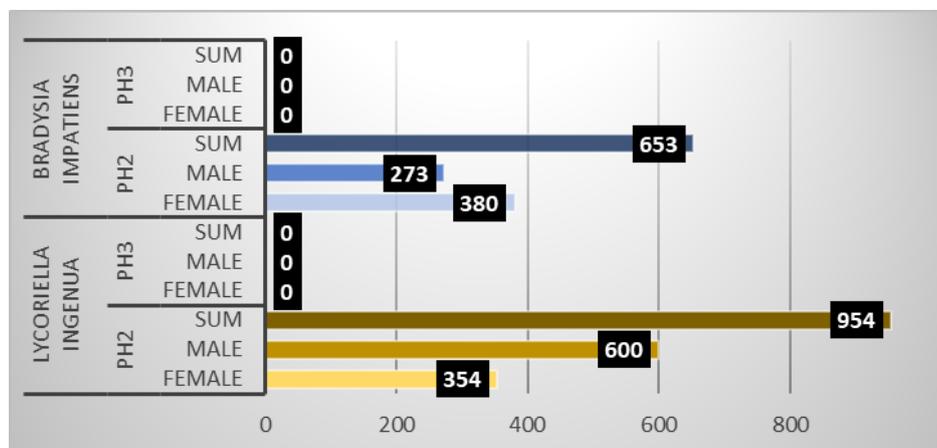


Figure 1. Total number of emerged adults by species and sex

#### Distribution of male and female insects

In the case of *Bradysia impatiens* a higher number of female adults emerged than males (380 and 273). This phenomenon was reversed in *Lycoriella ingenua*, where the emerged males were in greater number than female insects (600 and 354). The population of *L. ingenua* was composed of 63% male and 37% female adults, which gave the sex ratio of 1.69 male : 1 female. The distribution of sex was more even in *B. impatiens*, the sex ratio was 1.36 female : 1 male, which meant that the population was 43% males and 58% females, respectively (Figure 2).

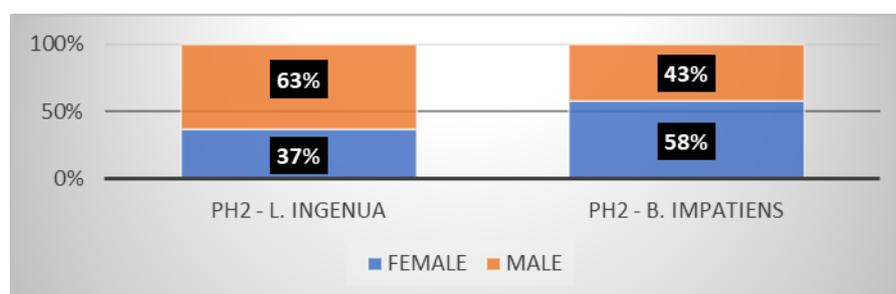


Figure 2. Sex ratio of *L. ingenua* and *B. impatiens*

#### Emergence dynamics of *Bradysia impatiens*

First insects were recorded on the 12<sup>th</sup> of March; 47 female and 73 male insects were collected. The following day, we asphyxiated 89 females and 95 males more. On the 14<sup>th</sup> of March, the number of captured females exceeded that of the males (109 females to 66 males). After this date, even the number of female adults started to decline. On the 17<sup>th</sup> of March, we only collected 11 female insects, and on the 18<sup>th</sup>, no gnat was collected from any of the breeding pots. The duration of swarming took approximately 6 days (Figure 3).

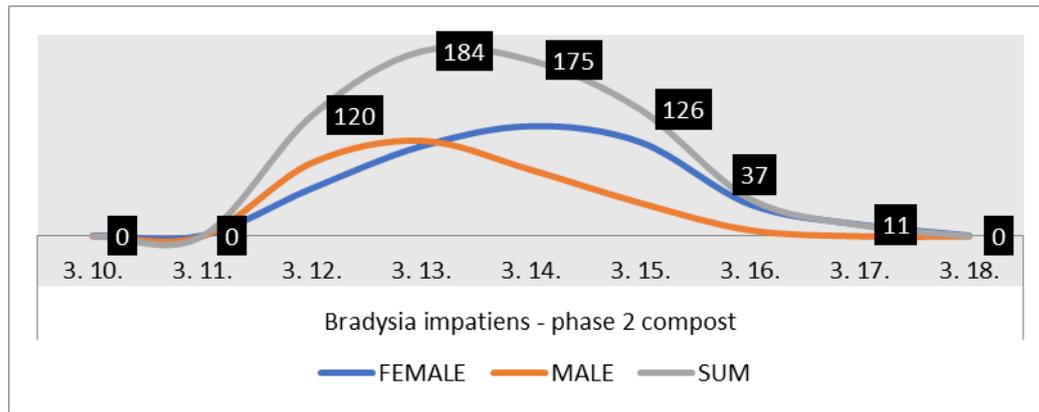


Figure 3. Emergence dynamics of *B. impatiens*

### Emergence dynamics of *Lycoriella ingenua*

On the 12<sup>th</sup> of March we have captured a single female and 24 male gnats from the breeding pots. A sharp and steady rise in the number of male adults could be seen until the 15<sup>th</sup> of March, from which onward the abundance of male gnats decreased strongly. The last male gnats were collected on the 18<sup>th</sup> of March. In the case of female gnats, a slow but steady increase was observed, the peak number of captured gnats was on the 16<sup>th</sup> of March (75), after that, the number of females slowly decreased. The last female specimens were asphyxiated on the 21<sup>st</sup> of March. The swarming of *L. ingenua* took approximately 10 days (Figure 4).

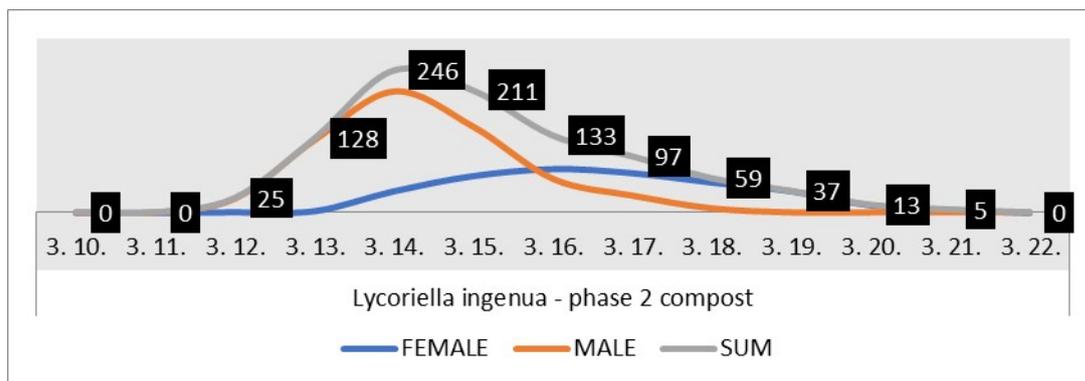


Figure 4. Emergence dynamics of *L. ingenua*

## DISCUSSION

Based on our results, we have concluded that both *Lycoriella ingenua* and *Bradysia impatiens* can develop into adult stage purely on phase 2 *Agaricus* compost. According to our recorded data, we found, that the number of total insects was greater in the case of *L. ingenua*, than *B. impatiens*. Emerging adults of *L. ingenua* were 46% more numerous, than that of *B. impatiens*. However, it did not mean that the fecundity of *L. ingenua* is greater than that of *B. impatiens*. We have not found literature, which would have supported the theory. The difference may have been the result of varying mortality of the two species in this experiment.

The distribution of male and female insects was quite different between the two species. The sex ratio was more skewed towards males in *L. ingenua*, and in the case of *B.*

*impatiens*, the two sexes were more in equilibrium. This is because of the genetic nature of how progenies are formed in the two species. In fungus gnats, the determination of sex and consequently sex ratio in populations, is based on how the X-chromosomes are eliminated during PGE (paternal genome elimination). In certain species of sciarids, the females either produce unisexual progenies (monogenic) or in other species, the progenies deriving from one female can consist of males and females alike (digenic). In monogenic species the sex ratio is closer to 1:1, while digenic species' sex ratio can considerably differ from 1:1. (METZ and SCHMUCK, 1929; HERRICK and SEGER, 1999; ROCHA AND PERONDINI, 2000). In some of the sciarid species, the two phenomena (monogenic and digenic females) can occur at the same time (ROCHA AND PERONDINI, 2000). *B. impatiens* is a monogenic (METZ, 1925; ALBERTS *et al.*, 1981) sciarid species, while *L. ingenua* is digenic (STEFFAN, 1974), which is why the sex ratios were observed different in our studies. Based on our findings we may suggest, that the food source does not influence the sex ratio of progenies. This is further established in our previous study, where we have compared different substrates for the development of *B. impatiens*, and the average sex ratio of the population did not differ from 1:1; it was 46:54 males to females, respectively (KECSKEMÉTI, 2017). However, certain environmental factors, such as temperature may have an effect on the sex ratio of progenies of certain fungus gnat species (CHENG ET AL., 2017).

The fact that zero number of adults have been recorded on phase 3 *Agaricus* compost either in the case of *B. impatiens* or *L. ingenua* is quite intriguing. It was a known fact to us that an *Agaricus* compost fully interwoven with the mycelia of *Agaricus bisporus* is not preferred by fungus gnat females for oviposition, and sub-optimal for larval development. The observed complete inhibition of development, however, was not expected. It is reported that the accumulation of calcium-oxalate crystals found in the *Agaricus bisporus* mycelia negatively affects the development of fungus gnat larvae (BINNS, 1980; WHITE, 1997; CHANG AND MILES, 2004). Previous experiments showed similar results (KECSKEMÉTI, 2017), from which we may suggest, that purely on phase 3 *Agaricus* compost, which is well colonized by *Agaricus bisporus* mycelia, the sciarid fly *B. impatiens* and *L. ingenua* cannot develop into the adult stage.

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