

## Development Characteristics and Life Cycle of a Sciarid Fly (*Lycoriella* sp.) in Indoor Rearing

### 실내 사육에 의한 버섯파리(*Lycoriella* sp.)의 발육 단계별 특성 및 생활사\*

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**ABSTRACT** This study was carried out to investigate the developmental characteristics and life cycle of sciarid fly (*Lycoriella* sp.), an important mushroom pest. The sciarid fly was reared in a growth chamber at  $21 \pm 1^\circ\text{C}$  and  $55 \pm 5\%$  of relative humidity with a 16h light, 8h dark cycle and *Flammulina velutipes* was used as diet. The egg was oval with 0.17 mm in diameter and 0.27 mm in length. Average egg period was 4 days. Body length of each instar from the 1st to 3rd was 0.7, 1.5, and 4.5 mm, respectively. Average weight of each instar was 1.9, 15.4 and 93.6  $\mu\text{g}$ , respectively. In the 4th instar larvae, however, developmental characteristics of female and male became different by body length, weight and segment size. Body length and weight of the 4th instar female larvae were 5 mm and 162  $\mu\text{g}$ , and those of the male larvae were 3.5 mm and 90  $\mu\text{g}$ , respectively. The larval periods of female and male were respectively 13.5 and 13 days. Pupal length of female and male was 3.5 and 2.7 mm, and average weight was 136  $\mu\text{g}$  and 65  $\mu\text{g}$ , respectively. Adults were emerged from female and male pupae on 5.5 and on 5 days after pupation, respectively. Average longevity of male and female were 7 and 5 days, respectively. The weight of female was two-fold heavier than the male. After copulation, female laid approximately 130 to 150 eggs on/in the sporophore of mushroom. In conclusion, total lifespan of male and female were 29 and 28 days, respectively.

**KEY WORDS** Developmental characteristics, Sciarid fly, *Lycoriella* sp., Life cycle, Indoor rearing

**초 록** 버섯 재배지에 있어서 주요 해충인 Sciarid fly(*Lycoriella* sp.)의 생활사와 발육단계별 특성을 조사하였다. 버섯 파리는 팽이버섯(*Flammulina velutipes*) 균사를 먹이로 공급하고  $21 \pm 1^\circ\text{C}$ ,  $55 \pm 5\%$  습도, 16L:8D 조건의 항온기에서 누대 사육하였다. 버섯 파리의 알은 직경 0.17 mm, 길이 0.27 mm의 타원형으로 알 기간은 평균 4일이었다. 1령으로부터 3령까지 유충의 체장은 각각 0.7, 1.5와 4.5 mm이었고, 평균 체중은 각각 1.9, 15.4와 93.6  $\mu\text{g}$ 이었다. 4령기 유충의 암컷과 수컷은 체장, 마디 크기 및 체중에 의해 형태적 특징으로 뚜렷하게 구별할 수 있었다. 4령유충 암컷의 평균 체장과 체중이 각각 5 mm와 162  $\mu\text{g}$ 이었고 수컷은 체장이 3.5 mm, 체중이 90  $\mu\text{g}$ 으로 조사되었고, 유충 기간은 암컷이 13.5일, 수컷이 13일이었다. 번데기 기간의 암컷과 수컷의 체장은 각각 3.5와 2.7 mm로 나타났으며 평균 체중은 각각 136  $\mu\text{g}$ 과 65  $\mu\text{g}$ 으로 암수간 차이가 현저하였다. 성충은 용화 이후 암컷이 5.5일, 수컷은 5일 경과한 후 우화하였고, 성충 기간은 암컷이 평균 5일 수컷은 7일이었다. 암컷의 체중은 수컷보다 2배 이상 높은 것으로 조사되었고, 교미 후 암컷은 130에서 150개의 알을 버섯 균사 위와 속에 산란하였다. 이상의 결과로, 버섯 파리 암컷과 수컷의 경과 일수는 각각 평균 29일 과 28일로 조사되었다.

**검색어** 발육 단계별 특성, 버섯 파리, *Lycoriella* sp., 생활사, 실내사육

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## INTRODUCTION

Mushrooms have been consuming for health foods and herbs for a long time, and, nowadays, its consumption is rapidly increasing due to the large income and the improvement of dietary life. Therefore, the growing area of mushrooms is also expanding, and it became an important source of income. Amongst the mushrooms, champignon (*Agaricus bisporus*), p'yogo (*Lentinus edodes*) and agaric (*Philomyces confusa*) are mainly cultivated in Korea. However, there are a lot of pest problems causing serious damages on the mushroom as other crops. It is known that the pests in the mushroom culture are mainly caused by mites (*Rhizoglyphis* spp., *Histiostoma* spp.), termites (*Reticulitermes speratus*), longi-corn beetles (*Moecchotypha diphysis*) and mushroom flies (*Lycoriella* sp. and *Mycophilla* sp.) (Kim 1990, Kim and Hwang 1996). For champignon and agaric cultivation, mushroom flies considered as the main and common insect pests, and Han *et al.* (1977) reported that among the mushroom flies, Sciarid type, *Lycoriella* sp. and Cecid type, *Mycophilla* sp. are the major pests causing severe damages in Korea. The damages on the mushroom are usually caused by larvae, and they feed on the mycelium of the mushrooms, or, burrow or tunnel into the stems and caps of maturing mushrooms. Furthermore, these pests can transport other pathogens such as pyemotid (pigmy) mite, nematodes and mold spores (Kinrus 1978, Wetzel 1981, Kim and Hwang 1996).

For the control of these pests, insecticides such as chlorpyrifos or diazinon have been widely used (Cantelo 1979, Argauer and Cantelo 1980, Kim 1990, Jhune *et al.* 1990). However, those chemicals caused several side effects such as environmental risks including toxicity to human, domestic animals or beneficial insects. Instead of chemical insecticides causing the aforementioned risks, researchers in other countries tried to find other solutions such as juvenile hormones like as methoprene, nematodes like as *Steinernema feltiae*, and microorganisms such as *Bacillus thuringiensis*, although it is yet far from the practical use (Cantelo 1979, Cantwell and Cantelo 1984, Nickle and

Cantelo 1991, Gouge and Hague 1994). On the contrary, the relevant studies on the mushroom flies are totally lacking in Korea, especially on taxonomical, ecological and physiological studies.

This study was carried out to make some ecological foundation as a part of effective control of mushroom flies causing aforementioned severe damages in Korea.

## MATERIAL AND METHODS

Adult mushroom flies, *Lycoriella* sp. were collected with a standard aspirator at a mushroom houses in Chilgok, Kyungbook on 1995. The mushroom flies collected were maintained and reared in plastic petri dishes (9×1.5 cm) at 21±1°C in a growth chamber with 55±5% of relative humidity. Spawns of a mushroom, *Flammulina velutipes*, were grown on Potato Dextrose Agar medium in the same condition at 21±1°C in a growth chamber with 55±5% of relative humidity for 13~16 days, and were supplied as the diet for the larvae.

To investigate the developmental characteristics and life cycle, the eggs, which were laid on/in the spawn bed and the plastic petri dish, were observed and measured the size with objective micrometer (0.01 mm) under a stereo microscope (×40) every 12 hours. Also, the weight of egg was measured with a chemical balance during the same observation period. Hatching was decided when the larvae perfectly escaped from the eggshell. All measurements were repeated three times, and calculated the average and standard error. Sexual difference of the larvae became distinct from the 4th instar. The characteristics for distinction of sexual difference such as body weight, length and the segment size were also measured. Furthermore, the time elapsed, body weight and length for pupae and adults were also estimated.

To observe the morphological differences between male and female of the 4th instar larvae, a scanning electron microscope (Phillips SEM 515) was used. For the scanning, the larvae were freeze-dried and fuming-fixed by 1% OsO<sub>4</sub> solution at room temperature for 2 hours. And then, samples were washed three

times with 0.1 M PBS (Phosphate Buffered Saline, pH 7.4) at 4°C for 20 minutes. Dehydration was then carried out in 50%, 70%, 80% and 90% ethyl alcohol for 15 minutes each and in 100% ethyl alcohol for 20 minutes. These processes have done three times, and the samples were finally laminated with gold after the critical point dry with CO<sub>2</sub> gas.

## RESULTS AND DISCUSSION

### Egg

In Fig. 1-A, the eggs of *Lycoriella* sp. were laid on/in spawn and plastic petri dish. Shape of egg was oval and the initial color of egg was brilliant-milky and became transparent as time passes. During this period, developing embryo (▶) could be observed. Egg period was approximately 4 days. According to Wetzel (1981), the incubation period was observed as 6 days at 18°C. Thus, it can be presumed that the egg period varies in relation to the incubation temperature. Average weight and size of egg were estimated as 0.9 µg and 0.17×0.27 mm (Table 2).

### Larva

Soon after the hatching, young larvae moved from the hatching site to mycelium, and started to feed immediately. The larvae preferred moderately moist areas

for feeding, and tend to be away from dry areas. This phenomenon was also reported by Wetzel (1981). From the 4th instar larvae, male and female could be mainly distinguished by body sizes characteristics of head and body, such as body size. Body size of female is generally larger than male. Both male and female have 11 segments with brilliant milky-white, and have a pair of spiracle from the 3rd to the 9th segment. Their head is brilliant-black (Fig. 1-B).

Average weight and length of the 1st instar larvae were 1.8 µg and 0.65 mm. The 1st instar larval period was approximately 3 days, and average weight and length were 15.4 µg and 1.65 mm.

Throughout the measurements of the whole larval period, the peak growth was observed during the last stage of the 2nd instar. During this period, the larvae increased 2~4 folds in body weight and length (Table 1.).

It was easy to confirm the differences between male and female due to the size of the 11th segment, especially among the 4th instar larvae, although morphological difference could not be defined. Fig. 2 is a photographs of SEM, and shows the notable morphological differences between male and female of the 4th instar larvae. Comparing the length of the 11th segment between male and female, the length of female was longer than that of male representing as 160 µm and 90 µm, respectively (Fig. 2). Average weight and length of the 4th instar larvae male were 90 µg and 3.5 mm, and those of female were 162 µg and 5.0 mm (Table 1), respectively. Larval period of the 4th instar was slightly different between male and female with approximately 4 days for male and 4.5 days for female. Therefore, the total period of larval stage for male and female were respectively 13 days and 13.5 days. In other study, Wetzel (1981) reported the larval period as 16 days at 18°C. According to Wetzel (1981), larvae and adults of mushroom flies tend to prefer a darkend area to an illuminated area which may be a reason why they normally are not found on the surface of the casing. With this phenomenon of mushroom flies, many scientists studied on the light traps in

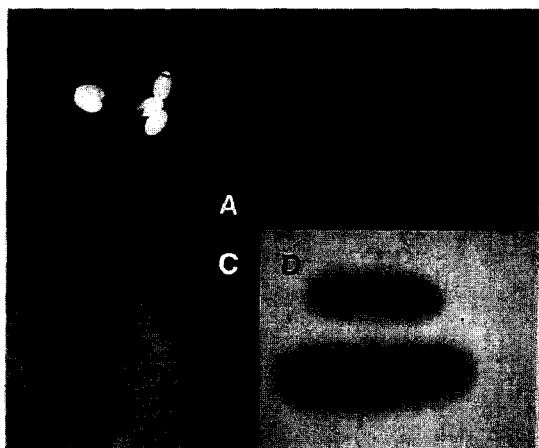


Fig. 1. Sciarid fly, *Lycoriella* sp. A: eggs, B: 4th larvae, C: pre-pupae, D: pupae.

Table 1. Developmental characteristics of larvae of Scliarid fly (*Lycoriella* sp.)

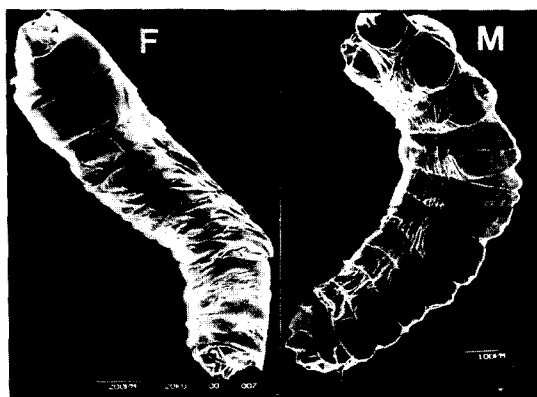
Larval instar	Date												
	1 <sup>st</sup>	2	3	4	5	6	7	8	9	10	11	12	13
Width of	0.05	0.073	0.098	0.15	0.2	0.26	0.307	0.307	0.307	0.35*	0.347	0.35	0.353
Head (mm)	(±0.006)	(±0.003)	(±0.002)	(±0.007)	(±0.025)	(±0.017)	(±0.007)	(±0.007)	(±0.07)	(±0.06)	(±0.003)	(±0.0)	(±0.009)
Length of	0.567	0.7	0.817	1.0	1.3	2.567	3.167	4.167	4.8	0.297**	0.3	0.303	0.303
body(mm)	(±0.067)	(±0.058)	(±0.073)	(±0.058)	(±0.1)	(±0.219)	(±0.088)	(±0.167)	(±0.115)	(±0.176)	(±0.058)	(±0.252)	(±0.033)
Width of	0.1	0.14	0.18	0.203	0.317	0.307	0.45	0.6	0.667	0.7	0.767	0.8	0.807
body(mm)	(±0.012)	(±0.01)	(±0.012)	(±0.003)	(±0.017)	(±0.007)	(±0.029)	(±0.115)	(±0.033)	(±0.0)	(±0.033)	(±0.0)	(±0.007)
Weight (µg)	1.2	1.833	2.6	6.067	8.3	32	70.67	90.33	120	0.557	0.5	0.507	0.54
	(±0.058)	(±0.26)	(±0.115)	(±1.157)	(±0.173)	(±7.7)	(±6.36)	(±8.18)	(±7.87)	(±0.012)	(±0.012)	(±0.007)	(±0.01)
										153	182.3	161	151
										(±1.732)	(±4.631)	(±6.35)	(±1.53)
										86	101	90	82.667
										(±1.732)	(±13.17)	(±5.77)	(±4.33)

\*female, \*\*male

**Table 2. Developmental characteristics of male and female pupa, adult and egg of Sciarid fly, *Lycoriella* sp.**

		pre-pupa	pupa				adult	egg
			1 day	2 day	3 day	4 day		
Length of body(mm)	F*	3.4 (±0.058)	3.03 (±0.067)	3.03 (±0.033)	3.03 (±0.003)	3.03 (±0.003)	3.3 (±0.153)	0.27 (±0.010)
	M**	2.7 (±0.100)	2.57 (±0.030)	2.5 (±0.010)	2.467 (±0.003)	2.5 (±0.006)	2.7 (±0.115)	
Width of body(mm)	F	0.8 (±0.029)	0.747 (±0.009)	0.793 (±0.023)	0.793 (±0.023)	0.81 (±0.001)	0.803 (±0.003)	0.17 (±0.001)
	M	0.643 (±0.023)	0.603 (±0.003)	0.653 (±0.003)	0.643 (±0.007)	0.553 (±0.015)	0.45 (±0.006)	
Weight(ug)	F	136 (±3.464)	135.3 (±1.764)	150 (±5.196)	120 (±3.464)	120 (±1.747)	140.3 (±6.060)	0.907 (±0.058)
	M	65 (±2.887)	65 (±1.560)	67 (±1.550)	65 (±2.150)	55.3 (±2.900)	55 (±5.500)	

\*female, \*\*male

**Fig. 2.** Scanning electron microscopy of female (F) and male (M) during the 4th instar larvae.

mushroom fly control. For instance, it was demonstrated that most of mushroom flies were attracted to Blacklight Blue or Blacklight among the tested fluorescent lamps at commercial mushroom farms (Parsells 1974).

### Pupa

At the end of larval stage, the larvae secreted mucus from the tail, adhered on the space of spawn or petri dish, and pupated without making a cocoon. During these days the pupae move a bit by convulsive movements of the abdomen rather than the rhythmic movement of the entire body. Pupal period was also slightly

different with 5 days for male and 5.5 days for female including pre-pupal stage. Wetzel (1981) estimated the pupal period as 6 days at 18°C.

There were no morphological differences between male and female during the pupal stage. However, male and female could be distinguished by the weight and length (Fig. 1-C, D). According to the measurements, the average weight and length of male were 65 µg and 2.7 mm, and those for female were 136 µg and 3.5 mm, respectively.

### Adult

It was known that adults generally consume small quantities of water and other liquids, and they do not feed mushroom tissues. However, they can affect the growth of the mushroom as a carrier of mites, nematodes, mold spores and other pathogens causing severe damage (Kinrus 1978, Wetzel 1981). For the morphological characteristics of adults, wings are folded a part way along the abdomen. They run on/around the surface of spawn bed and petri dish after emergence, and then wings are fully expanded within 30~60 minutes (Fig. 3). Average longevity of male adult was 7 days, while female lived 5 days.

Mating occurred immediately after the emergence, and then, female laid eggs within 48 hours. Average number of eggs oviposited by a female ranges between 130 and 150. Eggs were singly laid or in a group not

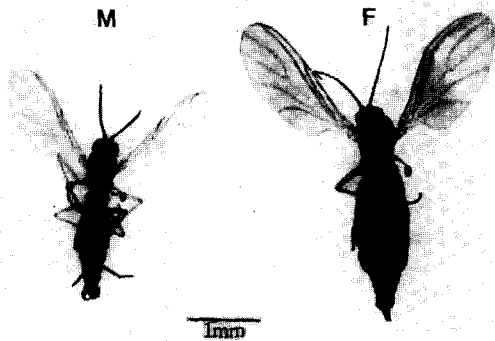


Fig. 3. Adult male and female of Sciarid fly, *Lycoriella* sp.

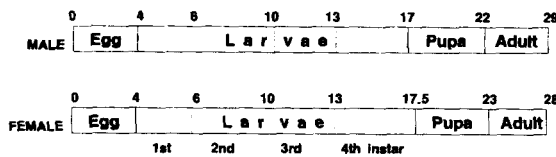


Fig. 4. Life cycle of a Sciarid fly, *Lycoriella* sp. on *Flammulina velutipes* in a growth chamber (21°C and 55% of Rh).

more than 15 eggs.

In conclusion, life history of a Sciarid fly, *Lycoriella* sp. is composed of distinct four stages of egg, larval, pupal and adult. Fig. 3 shows adult male and female. Male lived slightly longer than female. Average length of a generation time for male and female was respectively 29 and 28 days under 21°C. According to Wetzel (1981), average length of a generation time for male and female was respectively 35 to 38 days at 18°C. He noted that the length of generation time varies considerably with temperature and sex.

This study assessed the developmental characteristics and life cycle of mushroom flies (*Lycoriella* sp.). During the study, some problems associated with variation of ecological and physiological characteristics by temperature were uncounted. Therefore, further research should be evaluated on these aspects, especially the effect of temperature on the length of generation time.

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