BIOLOGICAL CONTROL

Predatory and Parasitic Activity of *Aphelinus asychis* (Hymenoptera: Aphelinidae) Following Exposure to the Entomopathogenic Fungus *Paecilomyces fumosoroseus* (Deuteromycotina: Hyphomycetes) Under Different Humidity Regimes

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An. Soc. Entomol. Brasil 28(4): 661-673 (1999)

Atividade de Predação e Parasitismo de *Aphelinus asychis* (Hymenoptera: Aphelinidae) Após Tratamento com o Fungo Entomopatogênico *Paecilomyces fumosoroseus* (Deuteromycotina: Hyphomycetos) Sob Diferentes Condições de Umidade Relativa

RESUMO - O efeito do hyphomyceto *Paecilomyces fumosoroseus* (Wise) Brown & Smith sobre as atividades de predação e parasitismo de *Aphelinus asychis* Walker, um parasitóide comum do pulgão-russo-do-trigo, *Diuraphis noxia* (Mordvilko), foi estudado sobre três diferentes umidades relativas. Três lotes de 20 fêmeas de *A. asychis* foram tratadas com 5,2 x 10^4 esporos/cm^2 (2 x DL_{95} para *D. noxia*). Cada fêmea foi então individualmente mantida numa placa de Petri com janela de ventilação provida com folhas de cevada e 20 *D. noxia* do 3º instar. Os lotes de 20 fêmeas tratadas foram mantidos em uma das seguintes umidades relativas: 67-73%, 85-89% e 94-98%. Números iguais de insetos não tratados foram mantidos nas mesmas condições (testemunha). A cada 24 horas, durante uma semana, os parasitóides sobreviventes eram transferidos para nova placa de Petri provida com folhas frescas e 20 pulgões. Após cada período de 24 horas de exposição e para cada parasitóide, os pulgões mortos foram contados e as folhas contendo os pulgões vivos foram colocadas sobre plantas de cevada a fim de permitir o reestabelecimento destes afídeos. Nove dias após a transferência dos pulgões, as múmias originadas foram contadas. O número total de afídeos consumidos e de múmias originadas do ataque dos parasitóides tratados, incubados a 94-98% de umidade relativa, foram significativamente menores do que os números correspondentes resultantes de parasitóides não tratados. Existe um declínio significativo de predação e de parasitismo para as

fêmeas tratadas e uma redução progressiva no número de parasitóides sobreviventes. A morte de todos os parasitóides tratados, incubados por sete dias à umidade de 94-98% após tratamento, e o alto nível de micose dos cadáveres demonstraram suscetibilidade do adulto de *A. asychis* ao fungo *P. fumosoroseus* em condições de alta umidade relativa. Contudo, a compatibilidade do fungo e do parasitóide nas baixas umidades relativas indica bom potencial para seu uso combinado.

PALAVRAS-CHAVE: Insecta, *Diuraphis noxia*, parasitóide, controle biológico, interação entre inimigos naturais.

ABSTRACT - The effect of the hyphomycete *Paecilomyces fumosoroseus* (Wise) Brown & Smith on the predatory and parasitic activity of *Aphelinus asychis* Walker, a common parasitoid of the Russian wheat aphid, *Diuraphis noxia* (Mordvilko), was investigated under three different humidities. Three cohorts of 20 female *A. asychis* were treated with 5.2×10^4 spores/cm^2 (twice the LD_{95} for *D. noxia*). Each female was then individually maintained in a ventilated Petri dish in which were placed three barley leaves and 20 third-instar *D. noxia*. Cohorts of 20 treated females were each held at one of the following relative humidities along with an equal number of untreated control insects: 67-73%, 85-89%, and 94-98%. Every 24 hours over a period of one week, the surviving parasitoids were transferred into a new Petri dish provided with fresh leaves and 20 aphids. After each 24-hour period of exposure and for each parasitoid, dead aphids were counted and the leaves with live aphids were transferred to barley plants to permit the re-establishment of the aphids. Nine days after the transfer of aphids, the mummies were counted. The total number of aphids consumed and mummies originated from the treated parasitoids incubated at 94-98% RH was significantly lower than in the control group. There was a significant decline in predation and parasitism per treated female per day and a progressive reduction in the number of treated parasitoids surviving each day. The death of all treated parasitoids incubated for 7 days at 94-98% RH after treatment and the high level of mycosis in cadavers demonstrate the susceptibility of adult *A. asychis* to *P. fumosoroseus* at high relative humidity. However, the reduced suceptibility of parasitoids at lower humidities means that may still be possible to construct an integrated program of pest management using parasitoids and pathogens.

KEY WORDS: Insecta, *Diuraphis noxia*, parasitoid, biological control, natural enemy interaction.

The Russian wheat aphid, *Diuraphis noxia* (Mordvilko) (Hemiptera: Aphididae) has been responsible for large losses in the production of wheat and other cereal grains in North America, South Africa, and other areas into which it has been introduced (Robinson 1993). For a variety of environmental and other factors, a sustainable approach for its control, using integrated methods including biological control, has been pursued. Since 1989, researchers at the USDA European Biological Control Laboratory (EBCL) have conducted a program of foreign exploration in a search for natural enemies of *D. noxia*, including pathogens, parasitic Hymenoptera and predatory insects (Hopper *et*

al. 1995). One of the most commonly observed parasites of this aphid is *Aphelinus asychis* Walker (Hymenoptera: Aphelinidae), a solitary internal parasitoid of *D. noxia* and at least 38 other aphid species (Kalina & Starý 1976). *A. asychis* may regulate aphid populations by adult host-feeding and parasitism (Esmaili & Wilde 1972, Michel 1973).

The importance of the Hyphomycetes as natural and biological control agents of *D. noxia* has only recently been studied. Feng *et al.* (1990) studied the pathogenic activity of *Verticillium lecanii* (Zimm.) and *Beauveria bassiana* (Bals.) Vuill. and, more recently, Mesquita *et al.* (1996) demonstrated the potential of *Paecilomyces fumosoroseus* (Wise) Brown and Smith as a microbial control agent of *D. noxia*. In order to utilise these fungi in an integrated pest management program combined with insect natural enemies for the control of *D. noxia*, their compatibility must be thoroughly studied under a variety of environmental conditions.

Fungi are highly dependent on the microclimate surrounding their hosts. The requirement for high humidity and suitable temperatures that enable germination, growth, and penetration of insects host by entomopathogenic fungi is well documented (Latgé & Papierok 1988, Ferron *et al.* 1991). The range of temperatures that enable growth of *P. fumosoroseus* has been studied by Vidal *et al.* (1997) and the effect of humidity on growth and infectivity of this species has been reported by Landa *et al.* (1994). Although high humidity is necessary for optimal entomopathogenic activity, penetration and subsequent death of *Bemisia argentifolii* Bellows & Perring (Hemiptera: Aleyrodidae) have been reported under field conditions with approximatively 40% relative humidity (Carruthers & Wraight, unpublished data cited in Lacey *et al.* 1996). The objective of our research was to quantify the effect of *P. fumosoroseus* on oviposition and predation by *A. asychis* at three different humidities.

Material and Methods

Fungus and insects. The isolate of *P. fumosoroseus* (ARSEF 4501) used in our studies originated from infected *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) collected on cotton in Multan (Punjab) Pakistan (Lacey *et al.* 1993). Procedures used for culture and viability determination were previously described by Mesquita *et al.* (1996). In brief, after passage through *D. noxia* the fungus was cultured on nutrient agar (SMDAY medium, in g/l : 10 neopeptone, 20 dextrose, 20 maltose, 20 yeast extract, 10 agar), in 9 cm diameter Petri dishes. A suspension of aerial conidia was prepared by superficially scraping conidia from the surface of 12-14 d-old cultures and adding them to 10 ml of sterile distilled water without surfactant, in a 60 ml glass with beads, for posterior homogenization. The glass was then shaken for five minutes in a mechanical agitator after which a 10^{-2} dilution of stock was used to determine conodial concentration. The conidial suspension was then diluted to a concentration of 1.4×10^8 conidia/ml. Conidial viability was determined by inoculating Petri dishes containing SMDAY medium and calculating the number of germinated conidia/100 conidia in four separate areas was counted after 18-20 h at 24 °C.

The colony of *D. noxia* started from collections made in Montpellier (southern France) and has been maintained at the EBCL since 1991. The cohort of aphid nymphs (± 24 h maximal age difference) for our tests was obtained from the colony and was reared on young barley plants (*Hordeum vulgaris* L. var. 'Clarine') at 24°C according to the method described by Gruber & Dureseau (1978), until they matured to third instars. Identification of this stage was based on developmental time for this species, relative size of the nymphs, and the number of antennal segments (Aalbersberg *et al.* 1987).

The strain of *A. asychis* used in our study originated from mummies collected from potted plants infested with *D. noxia* placed in a

wheat field in the vicinity of Montpellier during 1993. The mated parasitoid females utilized in all experiments were 4-5 days-old and were produced by mass rearing on *D. noxia* at EBCL. After emergence, parasitoids were transferred to a small maintenance colony of *D. noxia*, in order to pre-condition the females with regards to parasitism and predation. The *A. asychis* colonies were held in an incubator at 22-24°C, 55-65% RH, and photoperiod of 16:8 (L:D) until they were used.

Bioassay procedure. Once per week, during a three-week period, a cohort of 20 female of *A. asychis* was anesthetized with CO_2, placed on filter paper in a Petri dish, and treated with 2 ml of $1.4x10^8$ spores/ml suspension using a Potter spray tower (Potter 1952). This concentration corresponds to a coverage of 5.2 x 10^4 spores/cm^2 and is equivalent to 2 x LD_{95} of *P. fumosoroseus* for third instar *D. noxia* (Mesquita *et al*. 1996). Twenty additional dishes containing only filter paper were also treated each week with the same quantity of fungus. The lid of each dish had a 2 cm diameter ventilation window covered by gauze (0.1 mm mesh). Twenty third-instar *D. noxia* on three 5 cm pieces of barley leaves with their bases wrapped in moist cotton were placed in each of the dishes after treatment. A rectangle of parafilm (5 cm x 3.5 cm) was placed under the wet cotton that was used to provide water to the leaves to avoid moistening the filter paper. Individual *A. asychis* females that had been treated with fungus were introduced into each dish to ensure 24 h of contact of parasitoids with substrate that had been treated with *P. fumosoroseus*. The lids of the Petri dishes were then sealed with parafilm and the 20 dishes were placed in each of three plastic boxes (35 cm x 25 cm x 13 cm) containing different relative humidities : 67-73%, 85-89%, and 94-98%. The temperature during the test varied from 22 to 24ºC, and the photoperiod was of 16:8 (L:D).

After the initial 24 h, the *A. asychis* females were transferred to new, untreated Petri dishes and provided with 20 aphids on freshly cut barley leaves. The petri dishes were then returned to the humidity boxes.

Three cohorts of 20 *A. asychis* females treated with 2 ml of distilled water served as controls for each of the three RH regimes. Every 24 hours, the female parasitoids were transferred into non-treated Petri dishes with three barley leaves and 20 aphids as described above. This provision of fresh leaves and aphids was repeated daily for seven days for treated and untreated parasitoids.

After each 24-hour period of exposure, all aphids were removed from each Petri dish and dead aphids were counted. Treated dead female *A. asychis* were placed on water agar (3 g agar/l water) in 5 cm Petri dishes to confirm infection with *P. fumosoroseus*. Leaves holding live aphids which had been exposed to parasitoids were placed on separate barley plants (8-12 cm) per female per day to permit the re-establishment of the aphids. A plastic, cylindrical cage 25 cm long and 3.8 cm in diameter, supplied with five ventilation openings (1.5 cm diameter) covered by gauze, was used to isolate the plants. Nine days after the transfer of aphids, the mummies were counted.

Statistical analyses. The analysis of variance (ANOVA) was used, after data transformation, to test the effect of RH on the total number of aphids killed due to host-feeding and on the total number of mummies formed per *A. asychis* during one week. The effect of RH and number of days of incubation after treatment with *P. fumosoroseus* on the number of aphids killed by predation by *A. asychis* and on the number of mummies formed was also analyzed. The interaction between RHs and days post-treatment for treated and untreated parasitoids was considered in this analysis. The mean number of data were compared using Tukey's or Student's t test. The cumulative mortality of *A. asychis* seven days after treatement and the percentage of mycosed insects were submitted to a Kruskall-Wallis non-parametric test and the results were compared using the Wilcoxon test. Statistical analyses were performed using SAS software (SAS Institute, 1989).

Results

Predatory activity. The total number of *D. noxia* killed by *A. asychis* females treated with fungus was significantly reduced at 94-98% RH (F = 11.09; DF = 2; P = 0.0001), but did not differ significantly among the controls for the three RH groups. At the highest RH, the total number of aphids killed by treated *A. asychis* was significantly lower than that killed by untreated insects (t = 4.81; DF = 25.4; P = 0.0001) (Fig. 1).

For treated parasitoids, the interaction between RHs and days post-treatment was significant for the number of aphids killed by surviving parasitoids per day (F = 6.30; DF = 11; P = 0,0001) (Table 1). This number varied significantly among the RHs and was lowest from the third day onward for the highest RH (F = 5.48; DF = 2; P = 0.0071). At this humidity, the surviving parasitoids did not host-feed on day 6 and were all dead on day 7. At 85-89% RH, a significant difference in predation was only observed seven days after application of the fungus, when compared to the second day. At the lowest RH, there was no significant decline in predation over the 7-day post-treatment period. In addition to the observed decline in predation, the number of surviving *A. asychis* also progressively de-

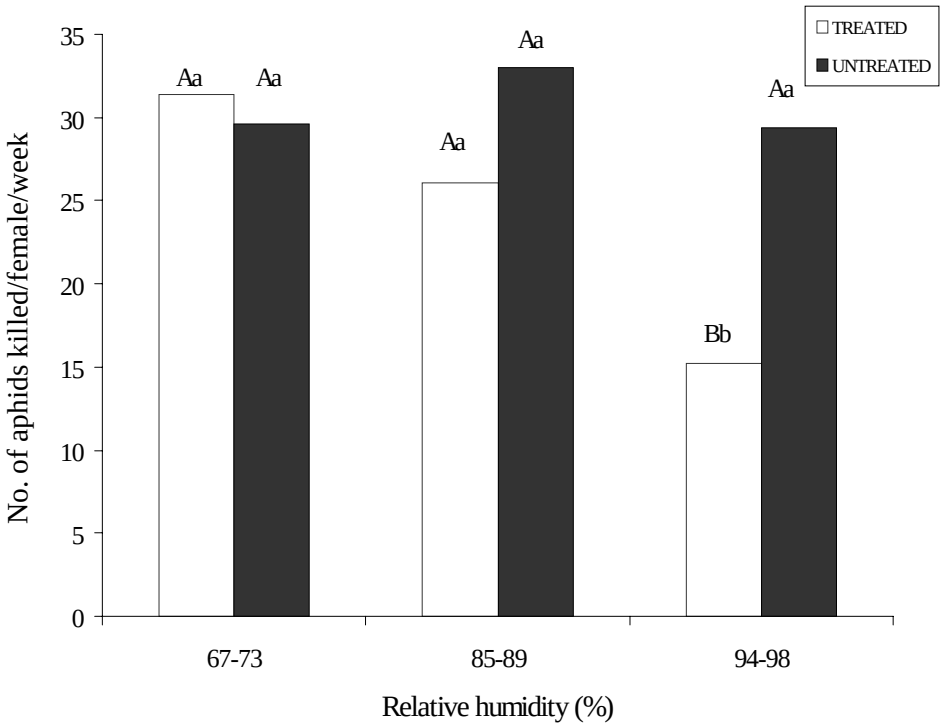

Figure 1: Number of aphids killed due to host-feeding, per female of *A. asychis* treated and untreated with *P. fumosoroseus*, under three humidity regimes. Means with the same uppercase letters within the same humidity (bars of different color), and the same lowercase letters at different humidities (bars of the same color) are not significantly different (P<0.05) using Student's t test and Tukey's test respectively.

Table 1. Number of *D. noxia* killed due to host-feeding by *A. asychis* untreated and treated with *P. fumosoroseus*, per surviving female per day at 3 different relative humidities (RH).

RH (%)	Number of days after treatment with *P. fumosoroseus*[1]						
	1	2	3	4	5	6	7
	Mean number of aphids killed/female/day ± S.E.						
treated *A. asychis*							
67-73	5.2 ± 0.5 Aa	5.3 ± 0.5 Aa	4.3 ± 0.4 Aa	4.4 ± 0.5 Aa	5.3 ± 0.7 Aa	4.5 ± 0.5 Aa	6.4 ± 0.5 Aa
85-89	4.5 ± 0.4 Aab*	5.4 ± 0.3 Aa	4.8 ± 0.5 Aab	4.8 ± 0.4 Aab	4.8 ± 0.4 Aab	3.8 ± 0.5 Bab	3.1 ± 0.6 Bb
94-98	5.8 ± 0.4 Aa	4.8 ± 0.4 Aab	2.9 ± 0.6 Bb*	3.0 ± 0.9 Bb*	2.0 ± 1.1 B b	0.0 Cc*	-
untreated *A. asychis*							
67-73	5.1 ± 0.4 Aa	5.4 ± 0.4 Aa	4.5 ± 0.3 Aab	4.4 ± 0.5 Aab	4.7 ± 0.6 Aab	3.1 ± 0.5 Ab	5.2 ± 0.4 Aa
85-89	6.0 ± 0.4 Aa	5.7 ± 0.3 Aa	5.6 ± 0.4 Aab	5.1 ± 0.3 Aab	4.8 ± 0.3 Aab	4.5 ± 0.4 Aab	4.1 ± 0.3 Ab
94-98	5.9 ± 0.5 Aa	5.3 ± 0.4 Aab	5.1 ± 0.5 Aab	4.8 ± 0.3 Aab	3.7 ± 0.4 Aab	3.7 ± 0.7 Ab	4.2 ± 0.6 Aab

[1]Means followed by different uppercase letters within each column for treated or untreated insects and by different lowercase letters within each row are significantly different ($P<0.05$) according to the Tukey's test for mean separation. For the same RH and the same day, means followed by the symbol *, for treated insects, are significantly different ($P<0.05$) from the corresponding mean of untreated insects, as determined by Student's t test. Means are based on surviving females only.

creased each day, resulting in large differences in the total number of aphids consumed by treated (304) and untreated (588) groups of parasitoids held at the highest humidity.

With the exception of the first day post-treatment at 85-89% RH, the capacity of *A. asychis* to kill its host did not vary significantly between untreated and treated insects at the two lower RHs on any particular day. In contrast, the effect of the treatment was significantly different from controls in the case of the highest humidity ($t = 2.82$; $DF = 28.3$; $P = 0.0085$ for the third day).

Parasitic activity. The total number of mummies resulting from the attack of *A. asychis* was significantly lower at 94-98% RH than at the two other RHs for both treated ($F = 33.72$; $DF = 2$; $P = 0.0001$) and untreated ($F = 3.34$; $DF = 2$; $P = 0.0440$) parasitoids (Fig. 2). At the highest RH, the total number of mummies formed per treated *A. asychis* was significantly reduced compared to untreated insects ($t = 4.71$; $DF = 27.0$; $P = 0.0001$).

For parasitoids treated with fungus, the interaction between RHs and days post-treatment was significant for the parasitic activity ($F = 6.52$; $DF = 11$; $P = 0.0001$). The number of mummies formed per day decreased significantly at the highest humidity from the second day post-treatment ($F = 3.20$; $DF = 5$; $P = 0.0490$) (Table 2). The effect of the fungus on reducing the rate of parasitism (i.e. number of mummies formed/day) was accelerated at the highest humidity. The number of mummies was significantly reduced from the third, sixth, and seventh day on at RH's of 94-98% ($F = 6.84$; $DF = 5$; $P = 0.0001$), 85-89% ($F = 4.30$; $DF = 6$; $P = 0.0007$) and 67-73% ($F = 2.59$; $DF = 6$; $P = 0.0220$), respectively as compared to the first day.

For the two lowest humidity conditions on any given day, the number of mummies formed by *A. asychis* did not differ significantly between treated and untreated insects. On the other hand, the effect of treatment with

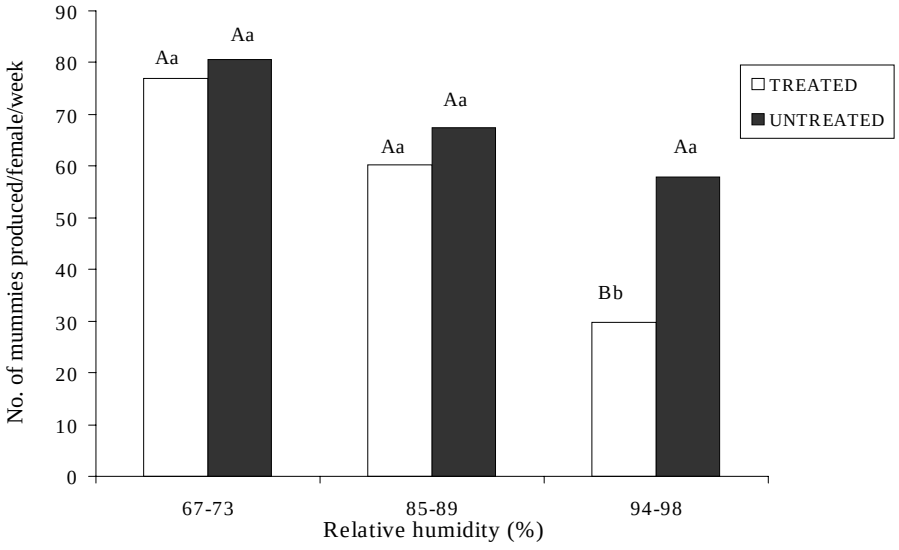

Figure 2: Number of mummies produced during one week, per female of *A. asychis* treated and untreated with *P. fumosoroseus*, incubated under three humidity regimes. Means with the same uppercase letters within the same humidity (bars of different color), and the same lowercase letters at different humidities (bars of the same color) are not significantly different ($P<0.05$) using Student's t test and Tukey's test respectively.

Table 2. Number of mummies resulting from the attack of *A. asychis* untreated and treated with *P. fumosoroseus*, per surviving female per day at 3 different humidities (RH).

RH (%)	Number of days after treatment with *P. fumosoroseus*[1]						
	1	2	3	4	5	6	7
	Mean number of mummies/female/day ± S.E						
treated *A. asychis*							
67-73	13.7 ± 0.7 Aa	12.1 ± 0.8 Aab	12.8 ± 0.8 Aab	11.5 ± 0.8 Aab	13.3 ± 0.7 Aa	12.5 ± 1.2 Aab	9.3 ± 1.2 Ab
85-89	13.2 ± 0.7 Aa	11.1 ± 0.8 Aab	11.4 ± 0.9 Aab	11.2 ± 0.8 Aab	9.3 ± 1.1 Bab	7.9 ± 1.1 ABb	7.3 ± 1.4 Ab
94-98	12.2 ± 0.6 Aa*	9.2 ± 1.0 Bab	5.1 ± 1.3 Bb*	5.8 ± 2.2 Bb*	2.5 ± 0.7 Cb*	2.0 ± 2.0 Bb*	-
untreated *A. asychis*							
67-73	13.3 ± 0.7 Aa	11.5 ± 0.9 Aa	13.6 ± 0.7 Aa	12.4 ± 1.2 Aa	12.7 ± 1.0 Aa	14.3 ± 0.8 Aa	10.4 ± 1.2 Aa
85-89	12.4 ± 0.6 Aa	12.2 ± 0.5 Aa	10.4 ± 0.8 Bab	10.4 ± 0.5 Aab	9.9 ± 0.6 Aab	9.6 ± 0.9 Bab	8.9 ± 0.8 Ab
94-98	9.9 ± 0.5 Ba	10.3 ± 0.6 Aa	10.1 ± 0.9 Ba	10.3 ± 0.8 Aa	9.1 ± 1.0 Aab	8.9 ± 1.4 Bab	5.9 ± 1.3 Ab

[1]Means followed by different uppercase letters within each column for treated or untreated insects and by different lowercase letters within each row are significantly different ($P<0.05$) according to theTukey's test for mean separation. For the same RH and the same day, means followed by the symbol *, for treated insects, are significantly different ($P<0.05$) from the corresponding mean of untreated insects, as determined by Student's t test. Means are based on surviving females only.

fungus and incubation at 94-98% RH markedly reduced parasitism relative to untreated insects on all but the second day of the observation period (t = -3.04; DF =29.9; P = 0.0048 for the first day).

Cumulative mortality of *A. asychis* and mycosed insects. For treated insects, the cumulative mortality was 76.5% at the third day for the 94-98%RH. At this humidity, 100% mortality was observed at the end of the test, which was significantly higher compared to the two lower humidities (Table 3) (Kruskall-Wallis test; Chi2 = 14.57; DF = 2; P = 0.0007). For the untreated insects at the seventh day after treatment, cumulative mortality reached 37.5% at the highest humidity, but did not vary significantly from the two other RHs.

At the highest humidity condition, 100% of treated *A. asychis* developed mycosis, significantly higher than those percentages at the higest humidity condition observed in parasitoids held at the two other RHs (Kruskall-Wallis test; Chi2 = 7.35; DF = 2: P = 0.0253).

Table 3. Cumulative mortality (%) of *A. asychis* untreated and treated with *P. fumosoroseus* seven days after treatment and percentage of dead parasitoids with fungal infection.

RH (%)	Cumulative mortality seven days after treatment (%)[1]		Percentage of dead treated *A. asychis* with confirmed infection by the fungus[1]
	treated *A. asychis*	untreated *A. asychis*	
67-73	38.9 Aa	16.7 Aa	57.1 A
85-89	61.1 Aa	22.2 Ab	72.7 A
94-98	100 Ba	37.5 Ab	100 B
	Chi2 = 14.57; DF = 2; P = 0.0007	Chi2 = 2.03; DF = 2; P = 0.36	Chi2 = 7.35; DF = 2; P = 0.0253

[1]Means followed by different letters uppercase in a column and by different lowercase letters in a row are significantly different (P<0.05) as determined by Wilcoxon's test.

Discussion

The majority of the mortality of *D. noxia* that was observed during each 24-h exposure period can be mainly attributed to the predatory behavior of *A. asychis*. Predatory behavior of aphelinids on hosts is considered to be a reproductive strategy for the majority of synovigenic parasitoids (Jervis & Kidd 1986). In the case of *A. asychis*, females emerge with a small number of mature oocytes (Flanders 1950) and the production of eggs depends on the nutrition provided mainly by predation during adult life (Boyle & Barrows 1978, Le Ralec 1995).

The difference between the number of dead aphids, as a function of relative humidity, demonstrates the negative effect of high humidity on the predatory behavior of the parasitoids treated with *P. fumosoroseus*. The fungus was, probably, favoured by high humidity through ennhancement of germination, growth and penetration into parasitoids (Latgé & Papierok 1988).

The number of mummies resulting from the attack of *A. asychis*, was reduced by an increase in RH and age of the females, for both treated and untreated parasitoids. This reduction was more evident for treated insects maintained at 94-98% RH. However, the decrease in the number of mummies formed relative to RH for untreated insects, showed that parasitic activity of *A. asychis* was more affected by the variation in humidity than predation.

Pathogens can cause a remarkable reduction in fecundity of infected target adult in-

sects (Santiago-Alvarez & Osuna 1988, Fargues *et al.* 1991; Hsiao *et al.* 1992). However, little is known regarding the effects of entomopathogenic fungi on fecundity of beneficial nontarget organisms (Croft 1989, Goettel *et al.* 1990, Brooks 1993). Fransen & Van Lenteren (1994) observed that adults of the parasitoid *Encarsia formosa* Gahan, when emerged from hosts treated with *Aschersonia aleyrodis* Webber, had normal reproduction compared with that of insects emerged from non-treated hosts. However, *A. aleyrodis* is specific for whiteflies and does not directly infect nontarget organisms. The potential for infection of *A. asychis* by *P. fumosoroseus* under ideal conditions for the fungus is greater due to the less specific nature of *P. fumosoroseus* (Lacey *et al.* 1996).

The death of all treated *A. asychis* incubated at 94-98% RH seven days after treatment demonstrated the susceptibility of the adult parasitoids to *P. fumosoroseus* at high humidity (i.e. optimal conditions for fungal development). This susceptibility was confirmed by mycosis in cadavers of *A. asychis* that had been maintained at this humidity.

It is clear that *P. fumosoroseus* can be antagonistic to the predatory and parasitic activities of *A. asychis* under conditions of high humidity. However, this study also indicates that a high degree of compatibility between the fungus and the parasitoid is possible under lower humidities and conditions that are also favorable for the fungus. For example, *P. fumosoroseus* is a highly efficient microbial control agent of *D. noxia* at 70-80% relative humidity, with high mortality (ca 95%) of the aphids within three days of treatment, under laboratory conditions, even when half dosage of the fungus inoculum was used (Mesquita *et al.* 1996). Mesquita *et al.* (1997) studied individual and combined effects of *P. fumosoroseus* and *A. asychis* on confined populations of *D. noxia*, under field conditions, and obtained an additive effect of the fungus and the parasitoid regarding to aphid control, without detrimental effects on the percentage of parasitism or parasitoid emergence when the two agents were used together. Also, Lacey *et al.* (1997) studied the effect of treatment with *P. fumosoroseus* ($2xLC_{95}$ for *D. noxia*) on foraging behavior of the parasitoid with video image analysis 24-96 h after treatment with the fungus and incubation at low (55% RH) or high (>95% RH) relative humidity. The authors were unable to detect any significant difference between control and fungus-treated females that were incubated at the lower humidity in terms of percentage of time walking, turning, resting, and average walking speed. However, fungus-treated insects that were incubated at high relative humidity were significantly less active than their respective controls for percentage of time walking, walking speed, and distance covered.

According to Brooks (1993), there are only a few examples of parasitoids being eliminated as a result of natural epizootics or whose ability to remain as significant control factors of specific insects pests have been compromised as a result of their direct susceptibility to pathogens of their hosts. However, natural and artificial mechanisms may help to minimize antagonism between fungi and parasitoids. These measures include spatial and/or temporal separation of fungal applications to avoid contact with parasitoids and other beneficial organisms when they are most likely to be abundant and or susceptible to fungal diseases. Augmentative and inundative release of aphelinids under drier conditions and applications of fungi under more humid conditions would serve to minimize antagonism and maximize their complementary activity as biological control agents.

Acknowledgments

We thank Lloyd Knutson, Director of the European Biological Control Laboratory for his support and encouragement. We are grateful to Claire Vidal, Marie-Jeanne Perrot-Minnot, Franck Hérard and Nathalie Smits for their constructive comments on the manu-

script. We acknowledge Guy Mercadier for his technical assistance and Cindy Lacey for help with revision of the manuscript.

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Received 26/III/98. Accepted 31/VIII/99.