

THE FIRST RECORD OF ENTOMOPATHOGENIC FUNGUS *Beauveria bassiana* (DEUTEROMYCETES) ON THE HIBERNATING PUPAE OF *Cameraria ohridella* (LEPIDOPTERA: GRACILLARIIDAE)



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Introduction

Invasive pest

The horse chestnut, *Aesculus hippocastanum* L. (Fig. 1), is an important ornamental tree in Europe. It is attacked by the horse chestnut leaf-miner, *Cameraria ohridella* Deschka et Dimic (Fig. 2), an important invasive lepidopteran pest.



FIGURE 1: A horse chestnut tree in a city park.



FIGURE 2: Adult of the horse chestnut leaf-miner, *C. ohridella*.

Since its first record in Macedonia in 1985 it rapidly spread and colonized major parts of Europe including Denmark, south of Sweden, Belorussia and Ukraine. The pest encounters favourable conditions for its development also in the Czech Republic because of a limited spectrum of natural enemies.

Pest control

Present methods of *C. ohridella* control are based on application of non-selective insecticides and composting or burning of leaf litter. However, these methods also kill beneficial organisms including natural enemies of *C. ohridella*.

Aim

- This work is a part of the project aimed at developing of a new, environment-friendly biopesticide against *C. ohridella*.
- Here we report the results of the first task focused on obtaining native strains of entomopathogenic fungi from *C. ohridella*.

Materials and Methods

The occurrence of entomopathogenic fungi spontaneously infecting *C. ohridella* was investigated in samples collected from heavily infested horse chestnut leaves. The samples of the leaves with diapausing *C. ohridella* pupae were collected in autumn 2007 in České Budějovice, South Bohemia, the Czech Republic (49°N).

The leaves were dissected and the pupae (Fig. 3) were individually kept at room temperature and high humidity conditions suitable for development of mycosis. The strains of the fungus were isolated from individual infected pupae using growth medium (Sabourau's agar) and are deposited in CCEFO (Culture Collection of Entomopathogenic Fungi Olešná) in the Czech Republic.



FIGURE 3: Diapausing pupa of *C. ohridella* in a chamber.

A pilot experiment to test the virulence of one of the isolated strains (CCEFO.071.BB) was conducted under the following conditions:

1. Water suspension of conidia obtained from a 20 days-old culture were prepared at concentration 5×10^7 per milliliter.

2. Suspension was applied using the dip-test.

3. Treated pupae were kept at $22 \pm 1^\circ\text{C}$, 95-100% R.H. and 18L:6D photoperiod.

Results

Development of mycosis was observed on numerous diapausing pupae. Most of the pupae were infected by saprophytic fungi, only few were covered by mycelia with conidiophores and conidia of *Beauveria bassiana* (Bals.) Vuill. (Fig. 4–6).



FIGURE 4: Hibernating pupae of *C. ohridella* infected by *B. bassiana*.

Totally, three strains of this fungus species were isolated (CCEFO.069.BB-CCEFO.071.BB). Moreover, one strain of *Paecilomyces fumosoroseus* (syn. *Isaria fumosorosea*, CCEFO.018.PFR) was also isolated. When diapausing pupae of *C. ohridella* were treated with the CCEFO.071.BB strain, 36% mortality ($n=100$) was recorded after 72 h of incubation.

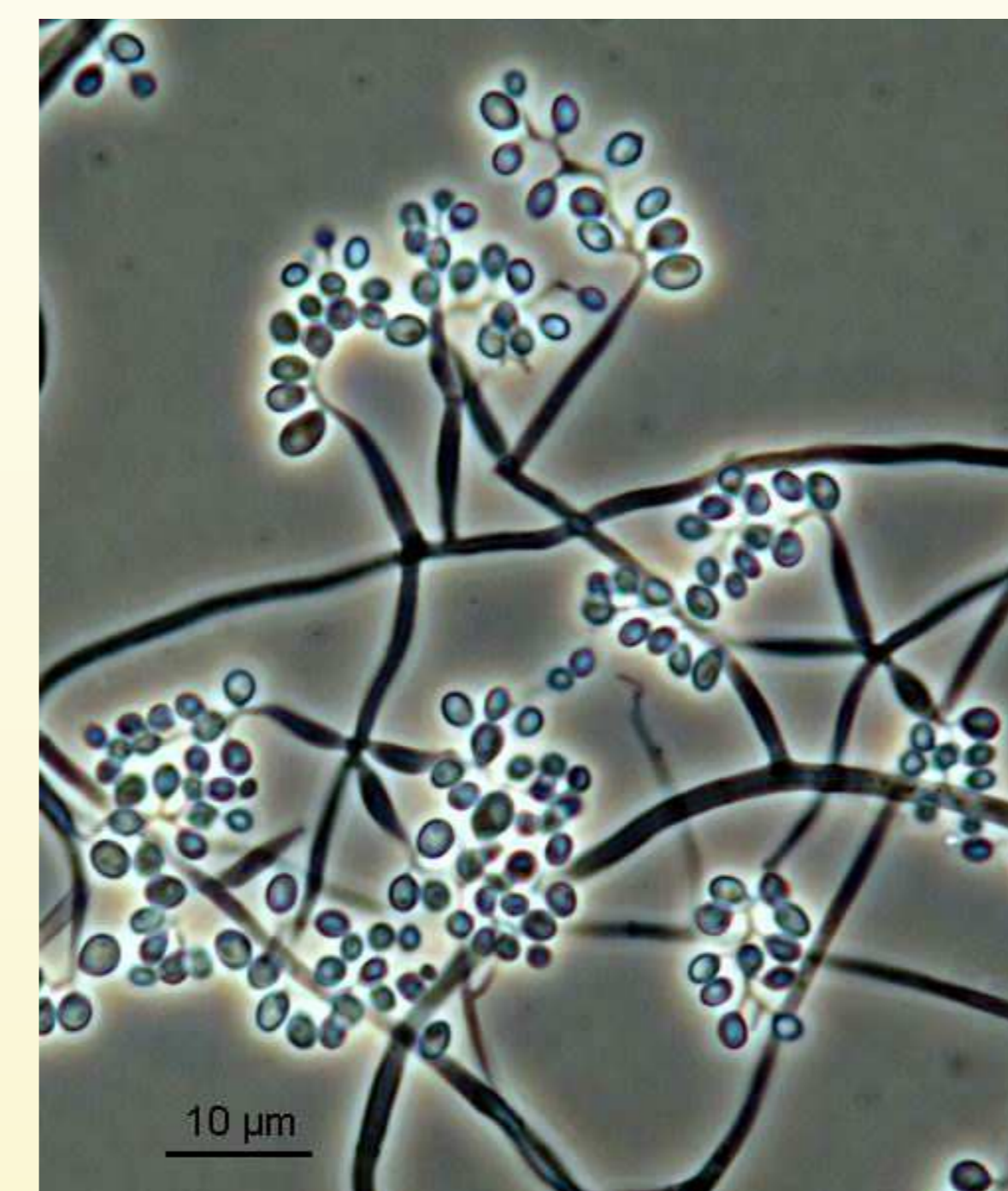


FIGURE 5: Conidiogenous cells with conidia of the fungus *B. bassiana* (photo A. Kubátová).

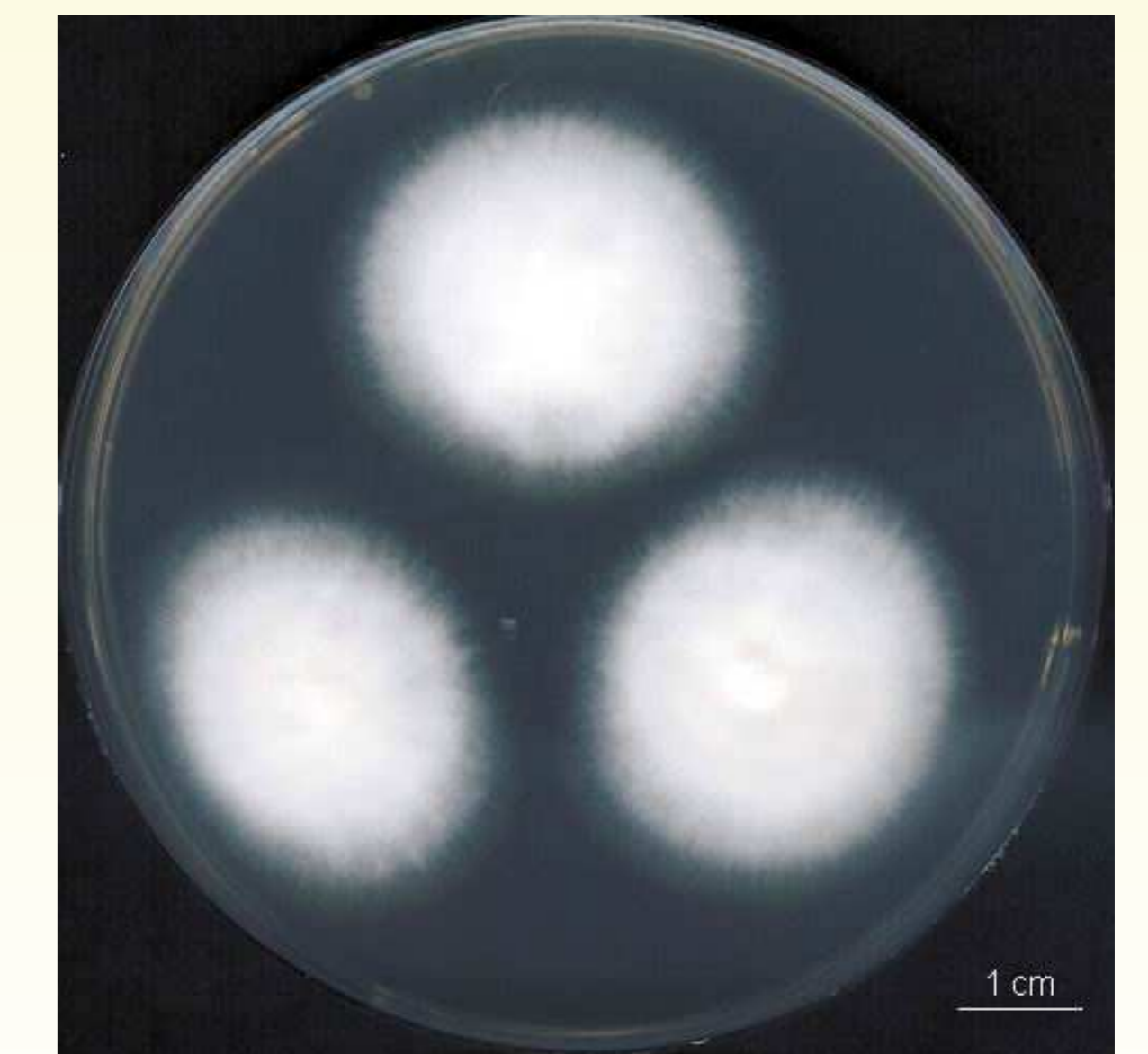


FIGURE 6: Culture of *B. bassiana*, PCA 10 days, 25°C (photo A. Kubátová).

Conclusions

- The discovery of spontaneous infection of *C. ohridella* by entomopathogenic fungus *Beauveria bassiana* is reported for the first time.
- Spontaneous infection of *C. ohridella* by the fungus *Paecilomyces fumosoroseus* already reported (Zemek et al., 2007) was also observed this year.
- Both fungal species were successfully isolated from the pest and cultivated on artificial medium.
- Our preliminary results indicate that *P. fumosoroseus* and *B. bassiana* have a potential to become a new environmentally safe biocontrol agent against *C. ohridella*.

Acknowledgements

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References

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- [2] Zemek R., Prenerová E. & Weyda F. 2007. The first record of entomopathogenic fungus *Paecilomyces fumosoroseus* (Deuteromycota: Hyphomycetes) on the hibernating pupae of *Cameraria ohridella* (Lepidoptera: Gracillariidae) Entomol. Research, 37: A135–136.