BIPESCO INTERIM REPORT (IR 5)

1 OUTLINE OF MAJOR ADVANCES

During the last six months, the BIPESCO-team increased its activities in order to complete two dossiers pertaining to the anamorphic stages of the entomogenous fungi *Beauveria brongniartii* and *Metarhizium anisopliae*. The dossiers will ultimately contain information on those agents currently being developed for the control of major subterranean crop pests. These pests include *Melolontha melolontha*, *Phyllopertha horticola, Amphimallon solstitialis, Strophosoma* spp. *and Otiorhynchus* spp. The information conforms with the EU Commission Directive 91/414 Appendix IIB and IIB. Comments will be included to show expert authorities which of the proposed methodologies are applicable to fungal biocontrol agents.

New formulations were evaluated to see if they improved spore viability and efficacy. Formulated inoculum of *Beauveria brongniartii* was incorporated in biodegradable matrices for soil application and attempts were made to improve the stability of postharvest inoculum of both *Metarhizium* and *Beauveria*.

A major goal in the third year of this project was to complete the ongoing field studies in Austria, Denmark, Germany, Italy and Switzerland (sub-task 6.1 to 6.3) and to initiate semi-field trials to examine the efficacy of the new, appropriate formulations. Current trials should be completed this autumn but already these together with the results of earlier studies suggest that some BIPESCO strains could be commercialised. In light of this, the BIPESCO team wish to submit a proposal to the EU, in the next call, to demonstrate the commercial potential of these agents.

The susceptibility and behaviour of a range of non-target invertebrates to *B. brongniartii* and *M. anisopliae* strains were evaluated. Side effects of the application of *B. brongniartii* were studied on earthworms in field trials and in bioassays. Populations of earthworms at two trial sites were assessed and no differences were found in biodiversity of earthworm species (e.g.

2 ACHIEVEMENTS

Three more milestones have been achieved in the last six months: (i) Molecular methods and tools for characterising fungal strains; (ii) biochemical methods and tools for characterising fungal strains; (iii) media which increase production of inoculum. The non-confidential information is already published and/or in press in international, refereed scientific journals.

The BIPESCO-papers also provide information that will help in the registration of fungal BCAs. They reflect the highly synergistic interactions between the partners and close co-operation of the BIPESCO team. A reference list can be downloaded from the BIPESCO Homepage **http:\\bipesco.uibk.ac.at**.

Furthermore, the first field study on *Melolontha melolontha* control has been completed within this reporting period. Melocont®-Pilzgerste, the commercial product based on barley kernels colonised by *Beauveria brongniartii*, was tested against the common cockchafer *Melolontha melolontha* in large field trials over a period of six years. The barley kernel product was applied in pastures in Austria, Italy and Switzerland with a slit seeder at various times of the year. Highest efficacy of the product was achieved by incorporating the inoculum into the soil at a depth of 3 to 10 cm. The results of field trials conducted between 1995 and 2000 with the barley kernel product indicated that the density of *B. brongniartii* increased continuously after each of the five applications performed between autumn 1994 and autumn 1997. Microsatellite marker analysis showed that the applied strain and re-isolated strains were identical. Application of the *B. brongniartii* barley kernel product resulted in sufficient suppression of cockchafer populations after only 2 years of application. Similar results are expected for the control of vine weevils with *Metarhizium anisopliae*.

In April 2001 an International Symposium was co-organised by BIPESCO on " Bioactive Fungal Metabolites - Impact and Exploitation, at University of Wales, Swansea, UK. More than 260 research scientists, registration authorities and extension services from 46 nations attended this meeting. Papers presented at the conference were particularly welcomed as submissions to Mycological Research and will be published in the year 2001/2002.

Selected members of the BIPESCO team will prepare and submit an application for an EU demonstration project. If successful, the project will provide information on the commercial viability of fungal BCAs for the control of subterranean pests. Already several grower organisations and small medium size enterprises have shown an interest in participating in this project.

The BIPESCO team is helping to organise the "Third Meeting of the *Melolontha* Subgroup IOBC wprs Working Group "Integrated Control of Soil Pests" which is to be held 24th-26th September 2001 in Aosta, Italy. This meeting provides an excellent opportunity for all participants to join with others in a discussion of an important issue concerning the relevance of *Beauveria brongniartii* in a practical context. The programme will offer the possibility to discuss issues pertaining to the successful control of the cockchafer (*Melolontha melolontha*).

An International Conference organised by BIPESCO will finalise our activities in January 2002. The tentative title is "Experiences on biocontrol of cockchafer larvae and vine weevils with entomopathogenic fungi ". Europe colleagues will be invited to attend this meeting in Vienna. The first announcement will be made in September

3 LITERATURE PUBLISHED OR IN PRESS

Abendstein, D. & Strasser, H. (2001) Oosporein accumulation during growth of *Beauveria brongniartii* in submerged culture. International Symposium: Bioactive Fungal Metabolites - Impact and Exploitation, 22. bis 27 April 2001, Swansea, p. 39.

Abendstein, D., Schweigkofler, W. & Strasser, H. (2001) Insecticidal, Antifeedant and growth inhibitory properties of oosporein on selected pest organism. IOBC wprs Bulletin, in press.

Chikwenhere, G.P.; Vestergaard, S. (2001) Potential effects of *Beauveria bassiana* (Balsmo) Vuillemin on *Neochetina bruchi* Hustache (Coleoptera: Curculionidae), a biologica control agent of water hyacinth. Biological Control, 21, 105-110.

Eilenberg, J.; Jensen, A.B.; Thomsen, L.; Vestergaard, S**.** (2001) Strategier for insektpatogene svampes spredning på friland [Strategies for the dispersal of insect-pathogenic fungi outdoor](In Danish, English summary). Slutkonference @Biologisk og Mikrobiologisk Bekæmpelse af Skadevoldere@, København, 4. Maj, DJF Rapport Nr 49, Markbrug, 39-43.

Eilenberg, J.; Nielsen, C.; Vestergaard, S**.**; Harding, S.; Frølander, A.; Augustyniuk, A. (2001) Biological control of weevils (*Strophosoma* spp.) in Danish greenery plantations. 8th European Meeting in the IOBC/WPRS Working Group: Insect Pathogens and Insect Parasitic Nematodes, Athens, 28/5-1/6. Proceeding in 76.

Eilenberg, J.; Jensen, A.B.; Thomsen, L.; Vestergaard, S**.** (2001) Strategier for insektpatogene svampes spredning på friland [Strategies for the dispersal of insect-pathogenic fungi outdoor](In Danish, English summary). Slutkonference @Biologisk og Mikrobiologisk Bekæmpelse af Skadevoldere@, København, 4. Maj, DJF Rapport Nr 49, Markbrug, 39-43.

Enkerli, J., F. Widmer, C. Gessler & S. Keller (2001) Strain-specific microsatellite markers in the entomopathogenic fungus *Beauveria brongniartii*. Mycol. Res. 105 (in press).

Enkerli, J. & S. Keller (2001) Ist die biologische Kontrolle des Maikäfer-Engerlings wirksam? Medieninformation.

Hozzank, A., S. Keller, O. Daniel & C. Schweizer. Impact of *Beauveria brongniartii* and *Metarhizium anisopliae* (Deuteromycotina: Hyphomycetes) on *Lumbricus terrestris* (Oligichaeta: Lumbricidae). Bull. IOBC/WPRS (in prep.).

Jung, K., Horaczek, A., Zimmermann G.and Viernstein H. (2001) Granular formulations of *Beauveria brongniartii*. SIP, accepted.

Keller, S., P. Kessler & C. Schweizer. Distribution of insect pathogenic soil fungi in Switzerland (manuscript).

Keller, S. & C. Schweizer (2001) Ist das Drahtwurm-Problem ein Pilzproblem? Agrarforschung 8(7) (in press).

Lomer, C.J., Eilenberg, J.E., Langewald, J., Nielsen, C., Vestergaard, S. & Strasser, H. (2001) Current status of registration of *Metarhizium anisopliae* and *Beauveria spp*. under EU regulations. International Symposium: Bioactive Fungal Metabolites - Impact and Exploitation, 22. bis 27 April 2001, Swansea, p. 74.

Pernfuss, B., Schweigkofler, W. & Strasser, H. (2001) Distinction of the entomopathogenic fungal species *Beauveria brongniartii* and *Beauveria bassiana* by comparing their carbon utilisation patterns. IOBC wprs Bulletin, in press.

Pernfuss, B. & Strasser, H. (2001) The monitoring of carbon utilisation as a tool to differentiate entomopathogenic fungi and their virulence. International Symposium: Bioactive Fungal Metabolites - Impact and Exploitation, 22. bis 27 April 2001, Swansea, p. 86.

Schweigkofler W. (2001) Neue Erkenntnisse zum "Engerlingspilz". Untersuchungen der Laimburg zur Stabilität verschiedener Sporentypen. Südtiroler Landwirt 4, 42-43

Schweigkofler W. (2001) Stabilität des “Engerlingpilzes” *Beauveria brongniartii* im Boden. (P). Obstbautagung Meran, Italy 9.1.2001

Strasser, H. (2001) Was tun gegen den Maikäfer? Maßnahmen zur Maikäferbekämpfung im Ackerbau und Grünland. Der Pflanzenarzt, 3: 10-13.

Strasser, H., Vey, A. & Butt, T.M. (2001) Spatial-temporal distribution of toxins of insect-pathogenic fungi. International Symposium: Bioactive Fungal Metabolites - Impact and Exploitation, 22. bis 27, April 2001, Swansea, p. 34.

Strasser, H. (2001) Future needs in risk assessment of fungal BCAs. IOBC wprs Bulletin, in press.

Strasser, H. & Erschbamer, M. *(*2001) Effect of temperature on *Metarhizium anisopliae* conidia germination and vegetative growth. IOBC wprs Bulletin, in press.

Strasser, H. & Enkerli, J. (2001) Biological control of *Melolontha melolontha* with Melocont®-Pilzgerste based on *Beauveria brongniartii*: Long term study in pastures from 1994-2000. SIP, accepted.

Vestergaard, S.; Eilenberg, J. (2001) Ingen sideeffekter ved brugen af *Metarhizium anisopliae* i væksthuse [No side effects of *Metarhizium anisopliae* when used in glasshouses] (In Danish, English summary). Slutkonference @Biologisk og Mikrobiologisk Bekæmpelse af Skadevoldere@, København, 4. Maj, DJF Rapport Nr 49, Markbrug, 71-76.

Vestergaard, S.; Eilenberg, J.; Nielsen, C.; Harding, S. (2001) First field trials to control *Melolontha melolontha* with *Beauveria brongniartii* in Christmas trees in Denmark.

Vestergaard, S.; Eilenberg, J. (2001) Ingen sideeffekter ved brugen af *Metarhizium anisopliae* i væksthuse [No side effects of *Metarhizium anisopliae* when used in glasshouses] (In Danish, English summary). Slutkonference @Biologisk og Mikrobiologisk Bekæmpelse af Skadevoldere@, København, 4. Maj, DJF Rapport Nr 49, Markbrug, 71-76.