Application of Chaetomium species (Ketomium®) as a new broad spectrum biological fungicide for plant disease control: A review article

K. Soytong*1, S. Kanokmedhakul2, V. Kukongviriyapa3 and M. Isobe4

1Department of Plant Pest Management Technology, Faculty of Agricultural Technology, King Mongkut's Institute of Technology, Ladkrabang, Bangkok 10520, Thailand; *e-mail: kasem_soytong@excite.com
2Department of Chemistry, Faculty of Science, Khon Kaen University, Khon Kaen 40002, Thailand
3Department of Pharmacology, Khon Kaen University, Khon Kaen 40002, Thailand
4Laboratory of Organic Chemistry, Graduate School of Bioagricultural Sciences, Nagoya University, Chikusa, Nagoya 464-8601, Japan


Research and development of biological control agents for use against plant diseases have been undertaken for several years in both government and private sectors, as natural agents are needed to take the place of chemical fungicides. The problem associated with the use of hazardous chemicals for disease control has received increasing attention worldwide, due to the fact that pathogens become resistant to chemical fungicides and the resulting environmental pollution and ecological imbalances. The implementation of practical integrated biological control technology to control plant diseases has been successfully introduced to growers in China, Philippines, Russia, Thailand and Vietnam by using new broad spectrum biological fungicides from Chaetomium (Thailand Patent No. 6266, International Code: AO1N25/12 and registered as Ketomium® mycofungicide). Since 1989, the biological product has been developed and improved from 22-strains of Chaetomium cupreum CC01-CC10 and Chaetomium globosum CG01-CG12 in the form of pellet and powder formulation. The formulation has successfully been applied to infested field-soils with integrated with cultural control measures and organic amendments for the long-term protection of Durian (Durio zibethinus L.) and Black Pepper (Piper nigrum L.) caused by Phytophthora palmivora, Tangerine (Citrus reticulata Blanco) caused by Phytophthora parasitica and Strawberry (Fragaria spp.) caused by Phytophthora cactorum, Wilt of Tomato (Lycopersicon esculentum L.) caused by Fusarium oxysporum f. sp. lycopersici and Basal rot of Corn (Zea mays L.) caused by Sclerotium rolfsii against Phytophthora rot. Bio-control technology of plant diseases has successfully been demonstrated, either completely or partially, when integrated with other control measures for appropriate disease management. Biological products are useful, not only for the protection against plant diseases, but can also be used for curative effects of plant diseases, and also promote plant growth.

Key words: biological control, Chaetomium, mycofungicide.
Introduction

*Chaetomium* species are normally found in soil and organic compost. *Chaetomium* is one of the largest genera of saprobic ascomycetes with more than 300 species worldwide (Arx *et al.*, 1986; Soytong and Quimio, 1989). *Chaetomium* species are potential degraders of cellulosic and other organic material and can be antagonistic against various soil microorganisms. *Chaetomium globosum* and *C. cochlioides* are antagonistic to species of *Fusarium* and *Helminthosporium* (Tveit and Moore, 1954). It has been found that by using specific strains of *C. globosum* it is possible to obtain good control over many plant pathogens. By coating seeds of corn with spores of *Chaetomium globosum* it was possible to prevent seedling blight caused by *Fusarium roseum* f. sp. *cerealis* 'graminearum' (Chang and Kommedahl, 1968; Kommedahl *et al.*, 1981). Such seed coating treatments were also found to reduce disease incidence of apple scab caused by *Venturia inequalis* (Heye and Andrews, 1983; Cullen and Andrews, 1984; Cullen *et al.*, 1984; Boudreau and Andrews, 1987). It has also been reported that some isolates of *C. globosum* produce antibiotics that can suppress damping-off of sugar beet caused by *Pythium ultimum* (Di-Pietro *et al.*, 1991). A further isolate of *C. globosum* was found to be antagonist against *Rhizoctonia solani* (Walter and Gindrat, 1988) and *Alternaria brassicicola* (Vannacci and Harman, 1987) and also reduced the quantity of sporulation of *Botrytis cinerea* on dead lily leaves exposed in the field (Kohl *et al.*, 1995). One strain of *C. cupreum* has also been reported to be antagonistic against *Phomopsis* and *Colletotrichum* spp which are soybean pathogens (Manandhar *et al.*, 1986).

Screening of *Chaetomium* species and strains isolated from soils for use as potential biological control agents commenced in Thailand in 1989. Reports indicate that strains of *C. cupreum* and *C. globosum* are able to suppress plant pathogens such as *Curvularia lunata*, *Pyricularia oryzae* and *Rhizoctonia oryzae* *in vitro* (Soytong, 1989, 1992a). Viable spores of *Chaetomium* spp. were able to reduce tomato wilt caused by *Fusarium oxysporum* f.sp. *lycopersici* in greenhouse and field trials (Soytong, 1990, 1992b) and also prevent basal stem rot of corn caused by *Sclerotium rolfsii* (Soytong, 1991). Twenty-two strains of *C. cupreum* and *C. globosum* have been found to inhibit isolates of various plant pathogens (Soytong and Soytong, 1997). These effective strains of *Chaetomium* have been formulated into bio-pellets and bio-powders for the biological control of plant diseases and have been patented and registered under the name Ketomium®. Their efficacy against tomato wilt (*F. oxysporum* f. sp. *lycopersici*) and basal rot of corn (*S. rolfsii*) has been successfully evaluated in laboratory and greenhouse tests (Soytong, 1992).
This paper reviews the development of Ketomium®, its evaluation with field trials, its patenting, biosafety testing and mechanisms of controlling disease and increasing crop production.

**Field Evaluations with Ketomium®**

In order to patent the *Chaetomium* strains (named Ketomium®) as a mycofungicide we carried out the following field evaluations on various crops in order to test the activity of the mixed strains prepared as biopellets and biopowders.

**Tomato**

Heavy reduction in tomato crop yields result from infection with *Fusarium* in Thailand (Soytong, 1992). *Fusarium*-suppression tests and pathogen suppression tests were carried out for tomato. In the *Fusarium*-suppression tests soil that was already infected with *Fusarium* was used for planting. The Ketomium® application was however applied to the soil two months before the seedlings were planted in the plot. In the pathogen suppression tests plants were grown in the *Fusarium* infected plots without the addition of Ketomium® and when disease symptoms appeared Ketomium® was applied. No Ketomium® applications were made in the control while a further trial involved the application of the chemical fungicide Pentachloronitrobenzene (PCNB). These trails are described in detail by Soytong and Soytong (1997).

Ketomium® in the form of biopellets and biopowders were applied to *Fusarium*-infested soils where tomatoes were growing and it was found that Ketomium® suppressed pathogen growth and thus disease symptoms. In *Fusarium*-suppressive soils, it was shown that the tomato plants treated with Ketomium® biopowder and the chemical fungicide PCNB completely prevented damage by *F. oxysporum f. sp. lycopersici*. The disease incidence in plants treated with biopellets (22%) was significantly lower than in the non-treated plants (43%). Ketomium® in biopellet and biopowder form were as effective in reducing infection rates in the tomato plants as treatment with PCNB (Soytong and Soytong, 1997).

**Maize**

Basal rot of maize caused by *Sclerotium rolfsii* is a serious disease in lowland fields in Thailand. Ketomium® was also effective in the control of pathogens and disease suppression when applied to maize. There was a distinct difference in disease incidence between the treated plants and the non-treated controls. In the pathogen suppression test, disease incidence in the biopellet,
biopowder and PCNB treated plants were 14.5, 15 and 16%, respectively. These were significant improvements, compared with the 23.5% disease incidence in the control. In the disease suppression tests, the same pattern was observed: disease incidence percentages of 16, 15.75 and 15.25 for the biopellet, biopowder and PCNB treated plants, respectively, compared with 25.55% in the control. It was shown that the mycofungicide, in both forms, was equally as effective as the fungicide PCNB in our experiments (Soytong and Soytong, 1997).

**Tangerine**

Integrated biological control of *Phytophthora* rot of Tangerine using mycofungicide under commercial growing conditions was investigated in Thailand for two years and the experiments were repeated four times. Tangerine (*Citrus reticulata*) is an important economic crop in Thailand. The major problem encountered in the citrus orchard is root and basal rots caused by *Phytophthora parasitica*. The pathogen has invaded citrus planting areas over the previous 30 years and the growers traditionally apply fungicides such as metalaxyl and allelate. In some cases the infected citrus trees have slowly died over several years. Ketomium® has been applied to orchard soils infested with *P. parasitica* and integrated with liming to adjusting soil pH and the addition of organic compost every four months to prevent root and stem rot of Citrus. Ketomium® treatments significantly reduced disease incidence by 47.25% when compared to the fungicide (metalaxyl). Incidentally, the inoculum of *P. parasitica* in soil at a depth of 15 and 30 cm was significantly lower with Ketomium® treatment than the non-treated soils. The pathogen inoculum at the upper soil profile had a higher population than the lower soil profile. It was also observed that Ketomium® treatment produced significantly higher growth parameters in the Citrus plants than with chemical fungicide treatments. Chaetomium treatments also gave significantly better fruit yields (52.35 kg/plant) than the fungicide treatments (27.79 kg/plant) as shown in (Fig. 1 and Tables 1, 2).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Disease Index</th>
<th>Disease reduction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 3 6 9</td>
<td>1 3 6 9</td>
</tr>
<tr>
<td>Chaetomium</td>
<td>6.71 a b 3.05 b 2.30 b 1.85 b</td>
<td>16.71 35.41 47.25</td>
</tr>
<tr>
<td>Control</td>
<td>6.90 a 3.40 a 3.50 a 3.90 a</td>
<td>- - -</td>
</tr>
</tbody>
</table>

1Disease index: 1 = no disease and 10 = severe disease.  
2Average of four repeated experiments. Means with the same common letters in each column are not significantly different.
Fig. 1. Comparative study between before and after application of Ketomium® for plant disease control. (Top = citrus tree before experiment; Bottom = citrus tree after experiment).

Table 2. Percentage colonization by *Phytophthora parasitica* after applying mycofungicide in the citrus orchard

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Soil depth</th>
<th>Colonization (%)&lt;sup&gt;1,2&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>15 cm</td>
<td>30 cm</td>
</tr>
<tr>
<td></td>
<td>Mar</td>
<td>May</td>
</tr>
<tr>
<td>Chaetomium</td>
<td>60a</td>
<td>40a</td>
</tr>
<tr>
<td>Control</td>
<td>58a</td>
<td>55b</td>
</tr>
</tbody>
</table>

<sup>1</sup>Colonization (%) = number of colonized leaf discs with *P. parasitica* / total number of tested leaf discs × 100.

<sup>2</sup>Average of four repeated experiments. Means with the same common letters in each column are not significantly different.

**Black Pepper**

The application of Ketomium® in pellet form was conducted in soils infested with *P. palmivora* in Chantaburi Province, Thailand to prevent root and stem rots of Black Pepper (*Piper nigrum*). The experiment was repeated four
Fig. 2. Comparative study between before and after application of Ketomium® for plant disease control. (left = black pepper before experiment, right = black pepper after experiment).

Table 3. Population dynamics of *Phytophthora* inoculum and antagonistic fungi in rhizosphere soil of Black Pepper.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Propagules (cfu/soil g⁻¹)</th>
<th>Colonization (%)&lt;sup&gt;12&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Months</td>
<td>1</td>
</tr>
<tr>
<td><em>Chaetomium</em></td>
<td>2.7 a&lt;sup&gt;2&lt;/sup&gt;</td>
<td>2 a</td>
</tr>
<tr>
<td>Metalaxyl</td>
<td>3.1 a</td>
<td>2.8 b</td>
</tr>
<tr>
<td>Control</td>
<td>3.1 a</td>
<td>3.6 c</td>
</tr>
</tbody>
</table>

<sup>1</sup>Colonization (%) = number of colonized leaf discs with *P. palmivora* / total number of tested leaf discs × 100.

<sup>2</sup>Average of four repeated experiments. Means with the same common letters in each column are not significantly different.

Ketomium® was applied at the rate of 20 g/plant and incorporated with liming and organic compost at intervals of four months for one year. The application of Ketomium® resulted in significant disease suppression with disease incidence at 22.66%. Ketomium® treatment did not significantly reduce disease times.
incidence when compared to metalaxyl treatment (disease incidence 21.88%. Ketomium® and metalaxyl treated plots however, had significantly lower disease incidence than the non-treated plots which had the highest disease incidence of 71%. Inoculum of *P. palmivora* in rhizosphere soil of Black Pepper was also significantly lower following Ketomium® and metalaxyl treatments than in non-treated soils. Thus, Ketomium® treatment gave significantly higher plants (average of 208 cm), followed by metalaxyl treatment (177 cm) and non-treatment (71 cm) (Fig. 2 and Tables 3, 4).

Table 4. Disease incidence and plant stands of Black Pepper after applying mycofungicide comparison with Metalaxyl to control *Phytophthora* rot in the field.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Plant stands (cm)</th>
<th>Disease incidence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Month 1</td>
<td>4</td>
</tr>
<tr>
<td>Chaetomium</td>
<td>33.08a</td>
<td>75.3b</td>
</tr>
<tr>
<td>Metalaxyl</td>
<td>33.61a</td>
<td>75.1b</td>
</tr>
<tr>
<td>Control</td>
<td>33.91a</td>
<td>60.4a</td>
</tr>
</tbody>
</table>

1 Disease incidence (%) = number of infected plants/total number of tested plant (healthy and infected) × 100.
2 Average of four repeated experiments. Means with the same common letters in each column are not significantly different.

**Strawberry**

Strawberry Crown Rot invaded the northern part of Thailand in the last few years. Biological control of Strawberry Crown Rot caused by *Phytophthora cactorum* using Ketomium® was evaluated in the infested soils planted with Strawberry at Fang District, Chiangmai Province and Maesai District, Chiangrai Province in Thailand during 1996 to 1997. Ketomium® mycofungicide in pellet form was applied at the rate of 0.04 g/plant added to soil media with 5 g of organic compost in plastic bags with seedlings of Strawberry before transplanting to the field. Chemical fungicide treatments served as a comparison. The percentage of dead plants and strawberry yield were not significantly different with the application of Ketomium® or chemical fungicides. The application of Ketomium® into the field-soil before planting however, resulted in significantly lower percentages of dead plants than Ketomium® treatments in plastic bags.

The population dynamics of antagonistic, plant pathogenic and saprobic fungi in rhizosphere soils of Strawberry were also evaluated for competitive growth in the treated and non-treated soils. It was found that the population of antagonistic fungi, *Chaetomium globosum*, and *C. cupreum* was significantly higher than that of the pathogen, *Phytophthora cactorum*, but this relationship was not shown in all experiments. This indicates that some limiting factors are
involved in biological control of plant diseases in the field, such as the ability
of antagonists to grow, soil pH, organic matter content, soil aeration, moisture
and chemical residues.

**Durian**

Durian is a tropical fruit tree originating from Indonesia and Malaysia
and is now distributed in Thailand. The major problem encountered in Durian
orchards is the low quality of yields due to *Phytophthora* rot. *Phytophthora
damilensis* is one of the most economically important stem and root rot of
Durian. Our evaluation of Ketomium® have demonstrated that Ketomium® can
completely prevent stem and root rot of Durian seedlings grown in inoculated
soils in greenhouse tests. The metalaxyl chemical fungicide treated seedlings
had the highest disease prevalence while all the seedlings died in non-treated
control. The integrated biological control tests were carried out from 1994-
1996 in a Durian orchard in an area where the soil is infested with *P.
damilensis* and the disease seriously affects Durian trees. Integrated biological
control comprised good cultural practices, organic amendment, changes in soil
acidity and removal of disease plant parts. This integrated control strategy was
highly successful in controlling the disease in the field after application of
Ketomium® at four months intervals and all treated disease plants recovered
(Pechprome and Soytong, 1997).

**Patenting and Registration of Ketomium®**

Ketomium® a new broad spectrum mycofungicide (Int. cl. 5 AO 1 N
25/12, Thai Patent No. 6266, 20 years, date of issue - 22 February 1994: date
of expiration - 21 February 2014) is a biological product of *Chaetomium
globosum* strains Cg1, Cg2, Cg3, Cg4, Cg5, Cg6, Cg7, Cg8, Cg9, Cg10, Cg11,
Cg12 and *Chaetomium cupreum* strains Cc1, Cc2, Cc3, Cc4, Cc5, Cc6, Cc7,
Cc8, Cc9 and Cc10. Ketomium® was registered by the Department of
Agriculture, Ministry of Agriculture and Co-operatives (No. 458/2539, 2
September 1996) using *C. cupreum* as a mycofungicide for the control of plant
diseases. It has been also registered as a biological fertilizer for degrading
organic matter and for induction of plant immunity and growth stimulant in
Guangxi Province, China and distributed as a biological protection of plant
diseases.

**Development of Ketomium® Formulations and their Shelf-Life**

Formulations of mycofungicides have gradually developed since 1992,
and mycofungicides have now been formulated into both pellet and powder
preparations (Soytong and Soytong, 1997). The 22 strains of *Chaetomium* spp.
are grown on potato dextrose agar for three weeks at room temperature (27-30 C). A spore suspension is then made and added to alginate mixture and dropped through a pasture pipette into a solution of 0.1 M calcium gluconate. The pellets are dried overnight before packaging. The pellets are ground to make biopowder before packaging (Fig. 3).

Compatibility of the Chaetomium strains when mixed together was initially tested and it was found that mixing did not affect activity, but had a synergistic effect. The biopellet and biopowder preparations were formulated and consist of a mixture of 22 effective strains of C. cupreum and C. globosum (Soytong, 1994). Viable spore populations were determined by dilution plate assays before pellet formation, 24 h after, and every two months for a period of one year. During this period pellets were stored at room temperature (27-30 C) in screw-capped jars. To test viability, pellets were dissolved in water with a mixture of $8 \times 10^2$ M KH$_2$PO$_4$ for dilution plating assays. Viable spore populations of the biopowder formulation was assessed in the same way. It was found that following storage the viability of propagules decreased in both formulations. There was a better survival rate in the biopellets (77%) compared to that of the biopowder formulation (57%) following three years of storage.

**Biosafety tests**

Acute toxicity tests of Ketomium® were carried out using the suspensions of Chaetomium spp. which consisted of equal concentrations of C. cupreum and C. globosum at the concentration of $9.9 \times 10^7$ spores/ml. A suspension was formulated in alginate sodium and talcum in sterilized water. The test animals were the Swiss-albino mice of both sexes weighing between 30-40 g. Mice were allocated into six groups for each sex. Mice were orally administered by gastric gavage with a Ketomium® suspension as a single bolus dose. Doses administered were divided into three levels for three mice groups (5, 10, and 20 ml/kg body weight, corresponding to $4.95 \times 10^8$, $9.9 \times 10^8$ and $1.98 \times 10^9$ spores/kg body weight) and another three groups fed with corresponding amounts of suspension without Ketomium®, served as control. After mice were fed with Ketomium® or the control formulation, the animals were examined for side effects hourly for 6 h in the first day and daily for another 7 d. Drug response profiles were recorded in a standardized worksheet of Nodine and Siegler (1964) where applicable. Observation was made for screening effects on the central nervous system and signs of intoxication. Drug response profiles were grouped as effects on the awareness, mood, CNS excitation, motor
coordination, muscle tone, reflexes, autonomic signs and death. Special attention was also paid to signs of gastrointestinal disturbance, possibly due to large amount of fungal spores being fed. The number of animals that died in a 7 d period after a single dose was tabulated. In addition to mortality, weight and daily examination for signs of intoxication, lethalogy and behavioral modification were conducted. Mice were housed with four animals per cage in the animal facility of Faculty of Medicine, Khonkaen University. After the observation period, animals were sacrificed and gross examination of internal organs were made and major organs were weighed.

Acute toxicity tests for Ketomium® were evaluated for bio-safety in experimental animals. It should be noted that this acute toxicity study does not ensure safety resulting from chronic exposure to spore formulations and toxicity with regard to some organ systems such as skin, lung and allergy or mutagenesis and carcinogenesis.

The values of mean lethal dose (LD<sub>50</sub>) could not be obtained in this study, since there was no death resulting in experimental animals fed with the highest possible dose. It was concluded that Ketomium® shows no apparent signs of acute toxicity in experimental mice for both sexes at doses of up to 1.98 x 10<sup>9</sup> and 2 x 10<sup>9</sup> spores/ kg body weight.
Fungal Diversity

How Ketomium® works

Ketomium® contains 22 strains of *C. globosum* and *C. cupreum* in the form of a spore power or pellets. *Chaetomium* species are strictly saprobic fungi belonging to the ascomycetes and are lignocellulose degraders (Arx et al., 1986). Specific strains of *Chaetomium* have been shown to stimulate plant growth and produce better crop yields both in the greenhouse and field, following their application as biological products (Chang and Kommedahl, 1968; Soytong and Quimio, 1989; Soytong, 1989; Soytong, 1992a; Soytong, 1992b; Pechprome and Soytong, 1997).

Control mechanisms

The mechanism of plant disease control involving specific strains of *C. globosum* and *C. cupreum* involves the production of antibiotics, e.g. *C. globosum* produces Chaetoglobosin C (Fig. 4) which suppresses the growth of plant pathogens such as *Colletotrichum gloeosporioides*, *C. dematioid*, *Fusarium oxysporum*, *Phytophthora parasitica*, *P. palmivora*, *P. cactorum*, *Pyricularia oryzae*, *Rhizoctonia solani* and *Sclerotium rolfsii* (Soytong and Quimio, 1989; Soytong, 1992b, Soytong and Soytong, 1997; Pechprome and Soytong, 1997).

![Fig. 4. Structure of Chaetoglobosin C produced from *Chaetomium globosum*.](image)

Plant growth stimulant

It has been shown that in greenhouse and field trials, that Tomato, Corn, Rice, Pepper, Citrus, Durian, Bird’s of paradise and Carnation plants treated with Ketomium®, have a greater plant growth and higher yields than non-treated plants. This implies that the mechanism of action for stimulating plant growth is probably confined to our specific strain of *Chaetomium*. It has been
found that some strains of *Chaetomium* produce substantial amounts of ergosterol (Fig. 5). This substance can help to improve the humus layer in soils which in turn leads to higher soil fertility.

![Fig. 5. Structure of ergosterol produced from *Chaetomium globosum*.](image)

**Induction of resistance in plants**

It has been found that the Chaetoglobosin C produced by our strains of *C. globosum* act as alien substances which induce a localised and sub-systemic oxidative burst (OXB) in tested plants e.g. Carrot, Potato, Sweet potato, Tomato and Tobacco. This possibly acts as an induction of plant immunity for disease resistance. OXB in plants can be characterized by rapid generation of active oxygen species (AOS) immediately after exposure to certain stresses. The active oxygen species play an important role in initiating signals for the response of various plant stresses (Doke, 1997). The OXB has been postulated to be an alarm signal in plant tissues against invasion by aggressive microorganisms and it seems to act as an emergency signal for responses against attack by alien substances, and may be related to resistance or protection against plant pathogens (Doke et al., 1991).

**Limitations of Ketomium®, concluding remarks and future studies**

The effectiveness of Ketomium® in plant disease control, especially soil-borne pathogens, integrated with other control measures such as cultural practices, e.g. sanitation, improving water drainage, pruning, removal of disease plant parts, adding organic compost and liming was investigated. These integrated control measures completely prevented damage by various pathogens in the infested fields. The curative effects of the Ketomium® when
applied to the infected plants, combined with the manipulation of the soil environment has been surprisingly successful. Implementation of integrated biological control of plant pathogens should be used to obtain disease control and reduce economic damage and compared with the use of chemical fungicides. The potential exists to use biological techniques to modify the infested soil environment to disfavor the pathogen and increase the resistance of plants.

Our observations indicate that Chaetomium globosum and C. cupreum can adapt to harsh microclimate conditions in infested soils when applied as Ketomium® formulations. Chaetomium spp. have the ability to survive during dry periods and to colonize the organic substrates rapidly in the soils. Most of data on Chaetomium spp. have resulted from laboratory and greenhouse studies. Our studies were conducted in the field for more than four years, with the application of Ketomium® as a broad-spectrum mycofungicide. It has been shown that this biological product can be applied to the field-soils infested with the test pathogens. However, further field trials and observations need to be carried out in order to establish a better integrated approach in using Ketomium®.

This report has shown that Ketomium® is effective and demonstrates the usefulness of an integrated approach in disease control. It discusses some limiting factors involved in biological control of plant diseases in the field such as the ability of the antagonists to grow in the field, and the effects of soil pH, content of organic matter, soil aeration, moisture and chemical residue in the soil. In different soils however, the ecology and climate may possibly result in no or poor biological control. Thus, we need to establish the biological diversity of soil microorganisms, the ecology of microbial antagonists, and the effects of chemical residues in the soil. The above experiments should also be continued in the same infested field-soils to establish whether biological control of plant pathogens can serve as long-term protection.

The expression of antagonistic activity, implying antibiosis, is consistent with the findings of some investigators where antibiotic substances, e.g. chetomin, sterigmatocystin, chaetomin (Udagawa et al., 1979; Sekita et al., 1981) are released by some isolates of Chaetomium spp. such as C. globosum (Arx et al., 1986). Screening for antagonism should not be limited to species, but should consider specific strains within species because these can differ in antagonistic activity (Soytong, 1992a). Several strains of C. globosum have been shown to produce Chaetoglobosin C which plays a role in antibiosis. These strains produce antibiotic substances which induce local and sub-systemic oxidative bursts (OXB) for signaling induction of disease resistance or immunity in plants such tomato, potato, cucumber, carrot and sweet potato.
Acknowledgements

We would like to thank N. Doke, Plant Pathology Laboratory, Department of Biological Resources and Environmental Sciences, Graduate School of Bioagricultural Sciences, Nagoya University, Nagoya, Japan for supporting the oxidative burst test. The research project had been fully supported from the International Foundation for Science (IFS) Sweden from 1989 to 1997 and partly supported by Third World Academy of Science (TWAS), Italy and National Research Council of Thailand. We would like to thank K.D. Hyde for his critical comments on the manuscript.

References


(Received 20 October 2000, accepted 10 April 2001)