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Review of CPHP Project R6773 -005

Control of storage pests with formulations of entomopathogenic fungi such as *Beauveria bassiana*

February 1999

This project review was commissioned by the Manager of the Crop Post-Harvest Research Programme (a component of DFID's Renewable Natural Resources Research Strategy). It was undertaken by:

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and

Jonathan Kydd of the Department of Agricultural Economics and Business Management, Wye College, University of London

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List of Abbreviations and Acronyms

CABI	CAB International
CABI ARC	CAB International Africa Regional Centre
CPHP	Crop Post Harvest Programme
DFID	Department for International Development (UK)
ICIPE	International Centre for Insect Physiology and Ecology
IIBC	International Institute of Biological Control (now CABI Bioscience)
IITA	International Institute of Tropical Agriculture
IPM	Integrated Pest Management
KARI	Kenya Agricultural Research Institute
LUBILOSA	Lutte Biologique contre les Locustes et les Sauteriaux
ODA	Overseas Development Administration (UK)
PAC	Programme Advisory Committee
PM	Project Memorandum Form
NRI	Natural Resources Institute
NRIL	Natural Resources International Limited
RNRRS	Renewable Natural Resources Research Strategy

1. Executive summary

1.1 This research project review was commissioned by the Manager of the Crop Post-Harvest Research Programme (a component of DFID's Renewable Natural Resources Research Strategy). It was undertaken by Tanya Stathers of the Food Security Dept., NRI, University of Greenwich; and Jonathan Kydd of the Department of Agricultural Economics and Business Management, Wye College, University of London.

1.2 This is the first formal review of Project R6773, which was approved for funding at a cost of £388,522 for a 52 month period, December 1996 to March 2001. The Project Memorandum had originally anticipated reviews in March 1998 and March 1999. March 1999 is an important "review point" for this project as the Programme Manager, advised by the Programme Advisory Committee (PAC), has to determine whether the project should be extended until its anticipated end date and, if so, what activities should be funded.

1.3 The purpose of the research project is to develop a control strategy against storage pests (notably *Prostephanus truncatus* and *Sitophilus zeamais*) based on entomopathogenic fungi. There is a body of professional opinion which holds that mycopesticides have the potential to provide the basis of storage pest control solutions which are environmentally benign, safe for humans and cheap. However, given the limited amount of existing research and development into the use of mycopesticides in storage systems, there is a respectable case for research which seeks to investigate and develop these technologies.

1.4 Section 3 of this review summarises the views of the PAC during the discussion leading up to approval. In essence the project was seen as worthwhile on balance: a substantial risk of failure needed to be set against the potentially very high impact that a successful research outcome could have on poverty reduction, via the stabilisation of rural livelihoods.

1.5 Section 4 reviews progress to date against outputs set out in the projects logframe. A mixed picture emerges. Out of 12 outputs, "good progress" has been made with one-third, "some progress" with one-third and "little or no progress" with one-third.

1.6 Section 5 discusses in more detail the potential contribution of this research to CPHP Semi Arid Production System Output 2, "effective and sustainable grain management systems developed" in the light of information which the project has already developed.

1.7 Section 6 considers the short to medium term outlook for the project and concludes that an amended activity plan will be required if funding is approved until 2001.

1.8 Section 6 also concludes that *high priority* should be given to:

- Uptake pathways and potential commercial partners
- Improving the persistence of entomopathogenic fungi within stores via reformulation and further studies of secondary cycling
- The interaction between technologies using entomopathgoenic fungi and existing storage pest management methods
- Technologies, cultural practices and farmer perceptions of mycopesticides;

Lower priority could be given to:

- Conidial longevity modelling (this, as noted below, is not accepted by the project leader)

1.9 Section 7 provides findings and recommendations covering:

For findings: the quality of the management and scientific procedures; nature and usefulness if the information and techniques which have been developed and potential development impact.

Key recommendations:

- **Recognising Project R6773 as an expensive "high risk/high payoff" investment, funding should be supported as envisaged in the PM for a further 24 month period March 1999 - March 2001, subject to some redesign to increase the likelihood of a development payoff.**

The redesign should include:

- Formalising a revised Activity Plan along the lines of the discussion above of Table 4 and the discussion below.

- Information dissemination activity should be partly refocused on potential manufacturers and distributors of the technology. This should be aimed at engaging their interest and it would be desirable to obtain private investment commitments to the near-market stages of the research.

- Thinking through the socioeconomic work, including the conduct of farmer trials and the estimation of the regional significance of the results of the trials.

- Assurance that satisfactory arrangements have been made to cover Susan Smith's role (a key staff member leaving the project).

- Tighter management control to ensure funds for activities not covered during the proposed timeframe are retained and used when time permits. This will help to ensure that agreed outputs are achieved within the budget. Any changes to the original PM which may affect activities and funds, need to be agreed by CPHP programme manager as and when they arise.

- The redesign may imply some input into the project by social scientists with expertise in (i) farmer trials and (ii) business development expertise to develop and manage links with potential private sector investors.

2. Overview of Project R6773 Control of Storage Pests with Formulations of Entomopathogenic Fungi such as *Beauveria bassiana*

The project is located in the Crop Post Harvest Research programme of DFID's RNRRS Research Strategy. It is relevant to "semi-arid" and "high potential" production systems. The physical location of the work is Kenya, for logistical reasons, but the project memorandum (PM) states that modeling would seek to establish the regional relevance of technologies developed. The project purpose (i.e. goal) is "environmentally sustainable and safe post-harvest and pathogen control methods and procedures developed and promoted".

The project memorandum summarises the project as existing:

> To investigate the role of entomopathogenic fungi such as *Beauveria bassiana* to control storage pests, especially *Prostephanus truncatus* and *Sitophilus zeamais*, but also more minor species.
>
> Extensive surveys will be carried out to obtain specific isolates which will be characterised and bioassayed against the pests and a biocontrol agent, the predator *Teretrius nigrescens*.
>
> Subsequently, methods of formulating these isolates for maximum efficiency will be investigated.
>
> Mass production will produce sufficient inoculum for testing under field storage conditions, along with studies of the environmental tolerance of fungal conidia within these formulations and the development of a model of conidial viability under storage conditions.

Elaborating the project purpose, the PM says that the aim is the development of a control strategy against storage pests (to field trial level) based on entomopathogenic fungi. The mycopesticides developed should be environmentally benign, cheap and safe for humans. The demand for the research is due to the fact that storage pests are a major source of crop loss and that since the late 1970s the larger grain borer *Prostephanus truncatus* has become a serious addition to the pest complex[1]. A 1996 East and Central Africa Storage Pest Management Workshop noted:

- That there is a need for sustainable alternatives to chemical pesticides;
- That *P. truncatus* cannot be considered in isolation from other storage pests;
- New IPM components, in addition to *Teretrius nigrescens* need to be adopted.

Contextual material

The use of synthetic chemical insecticides and fumigants to control insect pests in stored grain has given rise to environmental and health concerns and is hampered by reduced efficacy as insects develop resistance. These problems may be overcome by the adoption of integrated pest management strategies, important components of which are biological control methods. Biological control employs beneficial

[1] Subsequent to the PM, studies in Kenya (including the present one) have found that the LGB appears to be a much less serious pest than it is in Tanzania and Ghana.

organisms as pest control operators. Rather than applying a chemical insecticide, parasitic, predatory or pathogenic organisms are reared and released to control the pests. In many cases the biological control agents will reproduce and spread after the initial release, thus a single inoculation may be sufficient to establish lasting control.

Biological control makes up much less than 0.5% of the world market for pest control solutions (Yudelman *et al*, 1998). However, this figure is misleading as it is often only when natural and often unacknowledged biological control mechanisms breakdown, that pest populations increase to a level requiring pest control solutions. The self sustaining nature of many biological pest control solutions and patenting issues associated with living organisms makes them unattractive to commercial producers under free market forces. At present, biological control is facing the same problem as most other new technologies in pest management - difficulty in overcoming the market appeal of chemical control (Yudelman *et al*, 1998). However, the application of microbial agents such as entomopathogenic fungi within the closed system of a store, is likely to be similar in technique and rationale to the application of chemical protectants. This may make it more attractive to commercial exploitation.

Although laboratory data is accumulating from studies of pathogens, parasitoids, and predators of stored product pests, very few full-scale field experiments to evaluate the efficacy of biological control agents of stored products pests have been performed (Brower *et al*, 1995). It is critical that the performance of biological control agents in actual stores is evaluated because of the potentially unpredictable influences of scale, environmental conditions, pesticide residues, interspecies interactions and management practices. The results from such trials are needed to provide confidence in the level of control expected from applying a prescribed biological control program.

Timeframe

In discussing the timeframe for the development of a biological pesticide technology, the PM states that successful development would take a number of years. Initial work has demonstrated the theoretical feasibility of the concept because indigenous, but non-specific, isolates have been found which are virulent to the major pests. The present project would reach the stage of full scale field trials by 1999.

Personnel and Collaborators

The project leader is Dr David Moore, Research Group leader of the Insect Pathology Section of the International Institute of Biological Control (IIBC), Ascot, Berks (now known as CABI Bioscience (Ascot)). Also central to the project is another CABI Bioscience scientist, Susan Smith (who resigned as from January 1999).

Collaborating institutions are IIBC, Kenya (now known as CABI Africa Regional Centre (CABI ARC)) and Reading University (Dr Tran Hong). Dr George Oduor, a Kenyan Agricultural Research Institute (KARI) insect pathologist has been seconded to CABI ARC for a portion of the work, and now leads the project in Kenya, replacing Susan Smith.

The project logframe

The logframe (Table 1) lists twelve outputs. Section 4 of this report reviews progress against these outputs.

Table 1: LOGICAL FRAMEWORK (Project level)

Narrative Summary	Measurable Indicators	Means of Verification	Important Assumptions
Goal: (Purpose in given, Primary log frame)			(Goal to Primary Goal)
Improved on-farm cereal and legume storage	Losses in quality and quantity of stored grain caused by storage pests decreased	Loss assessment surveys by government and development agencies	Farmers accept storage pest control strategies as practical and economically feasible
Purpose: (Output= Purpose in given, 1°log frame)			(Purpose to Goal)
Environmentally sustainable and safe post-harvest and pathogen control methods and procedures developed and promoted	Enhancement of yield and quality of grain by decreased storage pest damage	Reports of NARS, extension services and development agencies	Adoption of control strategies not constrained by social or economic factors. Strategies promoted by NARS and extension services.
Outputs: (Activity in given, 1° log frame)			(Output to Purpose)
1. Establishment of a collection of fungal isolates of known virulence against *Prostephanus truncatus* and *Sitophilus zeamais*	Surveys completed Collection established	Availability of isolates, quarterly and annual reports, Project reports; published papers	Fungal isolates occur
2. Molecular characterisation of selected isolates	Laboratory data; characterisation facilities established	ditto cf reports etc	Characterisation facilities established
3. Host range studies of most virulent isolates	Laboratory data	ditto	Laboratory facilities available
4. Biological studies of most virulent isolates	Laboratory data	ditto	ditto
5. Development of formulations	Formulations developed and tested	ditto; formulations available	ditto
6. Population dynamics studies of populations of *P. truncatus* and *S. zeamais*	Laboratory data	ditto cf reports etc	ditto
Project Review (March 1998)			
7. Mass production facilities and techniques developed	Mass production facility established	ditto; Physical presence of facility	Mass production facility established
8. Major field trials conducted	Field data	ditto cf reports etc	Experimental results satisfactory
9. Workshop	Workshop conducted	Workshop proceedings	Experimental results satisfactory
Project Review (March 1999)			
10. Conidial longevity model developed	Longevity model available	Reports, published papers	Experimental results satisfactory
11. Experimental control strategy developed	Strategy available for testing	Published research and review papers	
12. Information disseminated			In general it is possible that the research may show that the techniques will prove to be ineffective or not cost effective; this is what the project aims to determine. However under most scenarios the circumstances should permit successful accomplishment of the research

Activities:	Budget summary £		(Activity to Output)
1a Intensive survey through southern Kenya and opportunistic surveying in northern Tanzania	Staff costs 141,276.90 Overheads 94,645.60 Equipment 28,200.00 Travel 14,420.00	Fungi available, reports, papers	Civil stability permits surveys
1b Collection of isolates from culture collections (UK)	Miscellaneous 110,000.00	ditto	
	Total 388,542.50		
2a Establishment of characterisation facilities in Kenya		Project report	Laboratory facilities and personnel available
2b PCR characterisation of isolates		Project report; published papers	
3 Bioassays of virulent isolates against main target insects, *Teretrius nigrescens* and additional storage pests		ditto	
4a Determine temperature tolerances of selected isolates		ditto	
4b Determine temperature growth responses of selected isolates		ditto	
4c Determine effects of humidity fluctuations on longevity of selected isolates		ditto	
5 Test formulations of fungi for virulence, effective dose transfer and tolerance to storage		ditto	
6 Laboratory studies to evaluate effects if fungus on model storage systems with *P. truncatus* and *S. zeamais*		ditto	
7a Mass production facilities established in Kenya		Project report	
7b Mass production techniques developed		Project report; published papers	
8 Field trials using farmer's storage facilities			
9 Workshop organised and conducted		Workshop proceedings	
10a Determination of effects of humidity and temperature on survival of *B. bassiana* conidia		Project report	
10b Relate 8 to meteorological data from GIS to develop practical field model		Model available; project reports; published papers	
11 Preparation of control strategy		Published papers;	Again under most reasonable assumptions the research work should be feasible
12 Preparation of papers, conference presentation, workshop		conference proceedings; workshop proceedings	

3. Introduction to the Review

The context of this project review
This research project under DFID's Crop Post-Harvest Research Programme was submitted for funding with a total value of £388, 552, to cover work in the 52 month period from December 1996 to March 2001, inclusive.

The project was approved in November 1996 for the value of £281,039, for support until the end of March 1999. Thus March 1999 was built in as a major "review point", on the assumption that funding of the remaining 24 month period to March 2001 would be conditional on the project being able to demonstrate: (a) good progress in achieving projects outputs and an acceptable likelihood of achievement of project purpose, and (b) that continued funding would secure further substantial progress in outputs and towards the purpose.

Programme Advisory Committee (PAC) Comments on the Project Memorandum
Funding for the research project was supported by the PAC. One member was of the opinion that the lethal effects of some mycopesticides against storage pests suggests that this is a fruitful field to explore. This member also felt that the literature review pointed to probable success in two key areas: (a) screening for other entomopathogenic fungi and (b) optimising the presentations of such agents in formulations which give maximum stability. Another member thought that the project was carefully thought out and based on a proven approach. This member saw the project outputs, if successful, as demonstrating the potential for major developmental gains in post-harvest systems. However, this member noted that further dissemination and uptake phases would be required to check downstream assumptions and whether socio-economic issues present any problems.

Despite being broadly favourable, some PAC members thought that the project was high-risk, speculative and expensive. One member explained that this perception of high risk resulted not from the technical aspects but:

> "because it is moving straight from the speculative initial project, which has shown promising results but does not guarantee a practical methodology, into a large and long-term programme of research across the strategic, applied and adaptive spectrum. The logframe says as much by saying that it is unlikely that they will not be able to complete the research programme (i.e. spend all the money) but that there is a risk that the end result will be an ineffective technique."

Another member commented that it was unlikely that robust and widely acceptable methods would be available by the end of the project.

PAC members were emphatic that steps needed to be taken as early as feasible in the research process to probe whether a viable technique was likely to be developed. This

would require investigation of practical application technology(ies) for formulations based on entomopathogenic fungi and socio-economic assessments of these technologies. One member commented that a workable approach to such investigation would be to undertake some economic modeling at an early stage to confirm suitability for farmer uptake. This member indicated that this could be done by, developing realistic assumptions as to how formulations would be used and marketed to farmers and other crop storers.

The current reviewers note that key socio-economic questions include:

- how formulations might be applied;
- their efficacy and persistency;
- human health implications (relative to alternatives) for both operators and consumers;
- whether private firms or public agencies would produce and distribute the formulation (and what might be the incentives for the former and possible subsidy costs for the latter);
- acceptability[2] and cost/benefit to users

Conduct of the review

The review was carried out over December 1998 to January 1999 by a scientist qualified and experienced in the research area (Tanya Stathers of NRI, University of Greenwich) and an agricultural development economist (Jonathan Kydd of Wye College, London University).

The reviewers examined project documents, including progress reports and publications and met with the two members of the research team (Dr Moore and Ms Smith) at the CABI Bioscience laboratory in Silwood Park, Ascot.

By coincidence one of the reviewers (Tanya Stathers) had taken the opportunity to visit the new CABI ARC laboratories while in Nairobi in April 1998, Dr George Oduor had kindly showed her around and discussed the project.

[2] There has been recent work in W. Africa about the acceptability of some mycopesticide application technologies (H. DeGroote pers. com.)

4. Review of progress to date

The table below summarises and comments on progress to date of Project R6773, using the outputs listed in the logical framework. This is followed by discussion of additional areas of the project memorandum, including measurable indicators, means of verification and important assumptions.

Table 2. Summary of project progress to date *(outputs from logical framework and dates from Section D of project memorandum)*

Key: ••• = good progress; •• = some progress; • = little/no progress

	Activity	Status	Progress to date
1	Establishment of a collection of fungal isolates of known virulence against *Prostephanus truncatus* and *Sitophilus zeamais* *(Ongoing)*	•••	Surveys were undertaken in 12 districts of Kenya, during which *B. bassiana* was obtained from 29 stored product insects from 12 farms. Unfortunately the *B. bassiana* isolates used in the early trials were found to be virulent against *P. truncatus* but not very effective against *S. zeamais* (which forms ~80% of the pest population in the stores sampled during the Kenyan survey). Two *B. bassiana* isolates virulent against both *S. zeamais* and *P. truncatus* have since been selected for use in further studies. The surveys were more time consuming than anticipated and as a result other activities were delayed. Selection of *B. bassiana* isolates has been complicated by findings that the virulence differed depending on the method of testing.
2	Molecular characterisation of selected isolates *(Mainly completed by 1998, but ongoing till end of project)*	•	Early during the project Ms Smith was trained in molecular characterisation techniques at CABI Bioscience (Egham). The lack of a constant power supply at the KARI research station at Muguga, made it impossible to carry out any molecular characterisation of the *B. bassiana* isolates collected during the survey. Without molecular characterisation studies, it will be difficult to know how similar the 12 isolates collected are. In order to obtain accurate information about the spread of a fungal isolate, molecular characterisation of any *B. bassiana* infections found would need to be carried out. Samples could either be sent back to the CABI Bioscience (Egham) laboratory or to a suitable laboratory in Kenya. A further training period would have to be undertaken by a Kenyan scientist in order to develop local capacity in these specialised techniques.
3	Host range studies of most virulent isolates *(By 1999)*	•	During the Kiboko field trial a number of storage pest species including: *S. zeamais*, *P. truncatus*, *Tribolium* spp., *Carpophilus* sp., *Sitotroga cerealella* were found to be infected by *B. bassiana*. High levels of *B. bassiana* infection (77-84%) were also observed in dead parasitoids present in the *B. bassiana* treatments in the Kiboko field trial. No specific studies have yet been carried out on the predator *Teretrius nigrescens* or specific parasitoids, these studies are now planned to start in April 99, and supplementary funding for this work has been sought from the Rockefeller Foundation.
4	Biological studies of most virulent isolates *(By 1999)*	••	Studies of the effect of different temperatures on the growth of different isolates have begun. Investigations of temperature tolerance and effect of humidity fluctuation on longevity will be carried out in conjunction with the conidial modeling work (see Activity 10).

5	Development of formulations *(Ongoing but largely completed by 1999)*	••	To date formulations tested include admixing dry conidia with maize and addition of *B. bassiana* spore and pheromone mixtures to hydrogenated fat pellets. Due to the health and safety issues surrounding the use of airborne spores of a respirable particle size, studies are continuing to try and identify alternative ways of formulating the conidial spores. Although, the fat pellets were shown to successfully transfer conidia to *P. truncatus* in the laboratory, they were unsuccessful with *S. zeamais*. Further work will include formulating the conidia in vaseline or wax which could then be used as part of a barrier on sacks, or as a thick layer covering the mouth of clay pots.
6	Population dynamics studies of populations of *P. truncatus* and *S. zeamais* *(By 1999)*	•	Population dynamics studies of *P. truncatus* and *S. zeamais* have not been carried out, although during the recent Entomology and Stored Product Pest Management workshop in Benin useful contacts were made with IITA and other researchers working on storage pests.
7	Mass production facilities and techniques developed *(Facilities established 1997, techniques ongoing but largely established by 1998)*	•••	Laboratories have been set up at the new CABI ARC in Nairobi, functioning from Jan 1998, and three technicians have been trained to a high standard in the laboratory skills needed to isolate, culture and mass produce *B. bassiana* and other entomopathogenic fungi. A small mass production unit is now operational and *B. bassiana* isolates on rice media have been successfully produced to amounts sufficient for the field trial requirements.
8	Major field trials conducted *(Major field experimentation to begin before the end of 1997 and will continue until 1999)*	••	An on-station field trial seeking to simulate farmers storage conditions was set up in October 1997, using dry *B. bassiana* spores mixed with maize, and maize and ash, found no reduction in damage levels in the *B. bassiana* treatments, despite very high levels of *B. bassiana* infection (50-80%) occurring amongst the dead insects during the first nine weeks. After nine weeks the *B. bassiana* infection level dropped dramatically, suggesting that fungal infection was no longer occurring due to reduced spore viability and a lack of significant secondary cycling. Both these factors are important if long term grain protection is to be achieved. Insects were not examined for the presence of internal sporulation, due to heavy work commitments. Since this trial, similar work at IITA in Benin suggests that secondary cycling may occur but after a delayed period (~6 months).
9	Workshop *(1998 – 1999)*	•••	A workshop on 'Entomology and Stored Product Pest Management' was successfully organised by CABI Bioscience and IITA, from 30 Nov. to 3 Dec. 1998 in Benin. 22 participants attended from 11 different countries (Benin, Ghana, Kenya, Nigeria, Tanzania, Togo, Uganda, Zambia, Canada, Denmark, UK). The proceedings and an interactive CD Rom will be produced by CABI Bioscience.
10	Conidial longevity model developed *(Beginning in 1998, completed by 2001)*	••	Modelling work has begun in collaboration with the University of Reading, preliminary results suggest that *B. bassiana* reacts in a similar way to *M. flavoviride* in response to temperature and humidity fluctuations. It is hoped that this study will provide accurate information on the longevity of the fungal spores in different storage conditions and when linked to climatic data be able to predict the success of this microbial control technique in different geographical locations.

11	Experimental control strategy developed	••	The initial field trial highlighted problems, principally related to the persistence of the inoculum and the experimental design. Some of these problems could not have been anticipated before the Kiboko field trial and it is hoped that laboratory studies and field trials in 1999 will help identify more successful control strategies. Once a successful control strategy has been developed, safety studies will have to be carried out on the selected *B. bassiana* isolate. Until these safety studies have been completed all field trials will have to be researcher managed making it difficult to identify socio economic issues associated with this technology. Preliminary studies into farmers' perceptions of mycopesticides have been carried out as part of the LUBILOSA project in West Africa, and issues arising from the LUBILOSA work should be carefully considered by this project. If a successful control strategy is developed, links will need to be established with potential commercial *B. bassiana* production units. At the local level one possibility would be for ICIPE and CABI ARC to set up a pathogen production unit depending on the demand.
12	Information disseminated *(On going)*	•••	Many dissemination outputs were produced during the first two years of the project including 7 scientific papers for submission to refereed journals, 3 published conference proceedings and reviews, and 5 conference presentations (see Appendix for details). Links have been made with other researchers working on stored product pests in Africa.

Key: ••• = good progress; •• = some progress; • = little/no progress

In the discussion below, boxed italic text is taken directly from the project memorandum and is followed by a discussion of relevant points.

15d *Successful development of a biological pesticide would take a number of years. Initial work has demonstrated the theoretical feasibility of the concept as indigenous, but non-specific, isolates have been found which are virulent to the major pests. The present phase would reach the stage of full scale field trials with various formulations of fungi by 1999*

Reviewers' comment

Initial surveys and virulence testing took longer than expected, which resulted in the delay of the formulation work, although admixing of dry spores and use of fat pellets were tested, neither were found to be acceptable and new *B. bassiana* formulations will be tested during 1999. If successful these new formulations will then be used in a full scale field trial.

19a Describe the project activities

Milestones

By March 1998 the most suitable isolate for development will have been selected and practical demonstration of control in laboratory model systems and small scale field experiments must have been demonstrated. Project will be reviewed in March 1998.

By March 1999 the feasibility of small scale mass production of fungal inoculum must be demonstrated. Field trials must have given significant control of storage pests (70% control or better) which may be expected to be improved with further field work and refined formulations. Project will be reviewed in March 1999.

Reviewers' comment

Two *B. bassiana* isolates have now been selected. In both the laboratory and the field, infection leading to mortality has been demonstrated, but no damage reduction has yet been achieved. No review occurred in March 1998.

Small scale mass production of fungal inoculum has been achieved in the new mass production facilities at CABI ARC. Field trials have not yet given significant control of storage pests, certainly not 70% control or better. The current review will be completed by February 1999.

19b What factors could prevent the attainment of:

i) Planned activities

a) Loss of or changes in personnel at IIBC Kenya.
b) Access to laboratory facilities in both Kenya and UK denied
c) Political or civil instability making field trials untenable

Reviewers' comments

a) Significant changes in project personnel have occurred with the unexpected departure of Susan Smith in December 1998. Although her input (previously 50% time) into the project was always to be reduced to 25% time from April 1999 to March 2000, this sudden departure may affect progress during the coming year. Dr Oduor will take over Ms Smith's responsibilities in addition to his own existing project duties in Kenya. As Dr Oduor also has inputs into a number of other RNRRS funded projects (Biorational methods for control of insect pests, Pest management of horticultural crops etc), this additional work load could potentially lead to problems in completing this years proposed activities. David Moore plans to make three visits to Kenya this year to help with the laboratory studies and field trials.

b) There have been no problems accessing laboratory facilities in Kenya or England, although the moving of CABI ARC from KARI Muguga site to Nairobi in January 1998 and the starting up of a new

laboratory will undoubtedly have caused some delays. Power problems in Kenya made it impossible to carry out the planned molecular characterisation work.

c) Field sampling was temporarily interrupted during the Kenyan elections.

Measurable indicators, means of verification and important assumptions for the project are shown in the logical frame (Table 1). As the project lifecycle runs from 1 December 1996 to 31 March 2001, many of the activities will not have been completed, the data from the surveys, laboratory and fields trials is available in quarterly and annual reports and published papers. Data for molecular characterisation, host range and biological studies of the isolates and population studies of the target insect pests is only very preliminary at this stage. Further studies are planned this year that should enable data to be collected on all these areas with the exception of molecular characterisation. The conidial longevity modeling studies have only recently started and the availability of an experimental control strategy is dependent on successful results of this year's studies. As for the important assumptions:

Fungal isolates occur	Pathogenic *B. bassiana* isolates have been discovered on storage insect pests for the first time in sub-Saharan Africa.
Characterisation facilities established	Lack of a constant power supply at the Muguga facilities prevented this from occurring initially. No attempt has since been made to establish facilities at the new CABI ARC Nairobi site
Laboratory facilities available	CABI ARC has established excellent laboratory facilities and three technicians have been trained during the course of the project
Mass production facility established	as above
Experimental results satisfactory	Field trials are yet to show any reduction in insect damage in the *B. bassiana* treatments. The conidial longevity modeling work only began in April 1998 progress to date has been encouraging. Experimental control strategy has not yet been developed the work is still at quite an early stage.

5. Contribution of the project to CPHP Semi Arid Production System Output 2: Effective and sustainable grain management systems developed

Insect damage of stored commodities is one of the principal areas of concern for most grain producers. Grain protection methods aim to improve rural livelihoods by increasing household food security and adding value to the harvested crop. Synthetic chemical insecticides are widely applied to stored grain and if used correctly can provide effective insect control. These insecticides, however, are often unavailable, expensive, unreliably formulated, ineffective due to pest resistance and increasingly believed to cause environmental and human health risks. Alternative methods are needed which will provide farmers with choices for cost-effective, safe and sustainable protection against damage and loss.

To date, there has been little research into the use of entomopathogenic fungi in storage systems, mainly due to the belief that the low moisture content of the storage environment is unsuitable for the successful use of fungi. However, laboratory and field studies conducted during this project have shown that a number of stored product pests including *S. zeamais*, *P. truncatus*, *Tribolium* spp., *Carpophilus* sp., *Sitotroga cerealella* can be infected and killed by *B. bassiana*. Isolates of *B. bassiana* have been shown to vary in pathogenicity against different insect species, the Kiboko field trial highlighted how important a knowledge of the composition of the target pest complex can be. These observations suggest that the high specificity of some *B. bassiana* isolates may prevent this technology from being transferable to areas where the pest complex composition differs from that used in the selection of the experimental *B. bassiana* isolate. Two isolates with virulence to both *S. zeamais* and *P. truncatus* have now been collected, and one of these will be used in future trials. No formal studies on the virulence of these fungi against parasitoids and predators has yet been carried out, although very high levels of *B. bassiana* infection (77-84%) were observed in the field trial, and further investigation is crucial (Table 3). Further studies on the compatibility of *B. bassiana* with other control methods, such as the traditionally used ash and chemical insecticides are also needed.

Table 3. Percentage *B. bassiana* infection of insect cadavers collected from the experimental Kiboko stores over a 21 week period.

	Week				
Insect sp.	**3**	**6**	**9**	**14**	**21**
Sitophilus zeamais	67	58	57	<1	1
Prostephanus truncatus	55	74	84	1	2
Tribolium spp.	50	50	12	2	3
Carpophilus sp.	55	30	19	2	3
Sitotroga cerealella	44	82	67	1	4
Parasitoids	77	84	84	2	<1

(Source: S. Smith unpubl. data).

Despite the initially extremely high levels of *B. bassiana* infection of several stored product insect species during the field trial (Table 3), no reduction in damage occurred. One of the disadvantages of biological control agents is that they are often much slower acting than conventional insecticides, feeding damage and oviposition

(egg laying) may already have occurred in the Kiboko field trials before the infected insects died. The sharp decline in *B. bassiana* infection levels between weeks 9 and 14 suggests that the virulence of the original inoculum decreased after 9 weeks in the store and that no significant level of secondary cycling (i.e. sporulation of infected insects leading to ongoing production and release of inoculum) was occurring within the sacks. Grain may need to be stored for more than six months and a biological pesticide needs to achieve a comparable control level and duration to that of a chemical pesticide in order to reduce food losses. Improved formulations may extend the longevity of the spores and a detailed study of secondary cycling may reveal possible techniques for aiding spore dispersal through the store. As fungal isolates vary in their persistence study of the different isolates collected may reveal opportunities for increased longevity, the conidial longevity modelling activities will identify areas in Africa where persistence will be extended.

Many of the farmers interviewed during the survey in Kenya purchased chemical insecticides including Actellic Super dust and malathion (Oduor *et al*, in prep.). Until a successful experimental control strategy using entomopathogenic fungi is developed, any cost comparisons would only be speculative, however production costs of different formulations should be calculated. Dr Moore is also involved in a locust and grasshopper biological control project, the biocontrol agent *Metarhizium flavoviride* used is now being produced both commercially in South Africa and on a small scale locally in Benin, and may act as a useful model.

In the short term, it will be difficult to monitor the 'user acceptability' of any control strategy that is developed, as safety data regarding the specific *B. bassiana* isolate is required before any farmer managed trials could be initiated. However, desk studies on West African farmers' perceptions and views on mycopesticide use should be studied to see whether there are similarities.

6. Short to medium term outlook

Prioritisation of research activities is needed to ensure that outputs are covered and that a successful experimental control strategy is developed before the end of the project. The bar chart of activities in the original PM is shown in Appendix 2, some critical activities have not been completed within the planned timeframe for a number of reasons which were mentioned in Section 4 of this review report. An amended activity plan for the remainder of the project period was constructed by Dr Moore (Table 4).

Potential uptake pathways need to be identified and information produced in a form suitable for dissemination to interested parties.

It is essential that the persistence of the entomopathogenic fungi within the stores is improved if long term control and reduction of damage is to be achieved. This will require reformulation to improve spore longevity and a detailed study of whether secondary cycling is occurring internally within infected insects in the store. If this is so, it will be important to study whether release of this inocolum will be increased by possible cultural practices such as regular rolling of sacks to break up insects and aid release of inoculum etc. The integration of this technology into existing pest management methods needs to be studied, and particular attention should be paid to the effect of the pathogen on natural enemies such as *T. nigrescens*.

If successful field results are obtained in 1999, further work on refining and improving application methods will need to be carried out and tested initially in researcher managed on-farm trials and subsequently by farmers. (Ethical and regulatory considerations prohibit farmer handling of these entomopathogenic fungi until safety data has been collected). Interesting work on farmers perceptions of mycopesticides in relation to grasshopper and locust control in West Africa has started and it is important that issues arising from this work which may also affect the use of pathogenic fungi by farmers in stores are considered as early as possible in the development of a control strategy. As mentioned above, once successful field results are achieved priority should be given to identifying and collaborating with interested public and/or private potential producers.

Table 4 shows planned activities to continue under this project, and others which at present are unfunded. It can be seen that six activities are prioritised (nos: 1, 2, 5, 6, 7 & 9). In our view Activity 9 - 'Conidial longevity work' is less critical to a successful outcome to the project, bearing in mind that successful field control has yet to be achieved with any isolate. It should be noted that Dr Moore disagrees with the reviewers on this point and maintains the view that "longevity predictions will pin-point areas where the conidia will survive longest and where control is most likely to occur, even if this is not Kenya. The dissemination output would also be of value to mycoinsecticide development."

Table 4: Revised activity plan for the remainder of the project (Jan 99 - Mar 2001)

	1 Dec 98 - 31 Mar 99	1 Apr 1999 - 31 Mar 2000				1 Apr 2000 - 31 Mar 2001				
	4	1	2	3	4	1	2	3	4	**Comments**
1. Effects of insect migration (Kenya)	X	X	X							Results will influence design of field experiment
2. Barrier protection (Kenya)	X	X								Result will influence design of field experiment
3. Isolate characteristics (Kenya/UK)		x	x	x	x	x	x	x		Molecular characterisation desirable but not a priority in Kenya
4. Non target effects (Kenya)		x	x	x		x	x	x		Emphasis on *Teretrius nigrescens*
5. Mass production (Kenya)		X	X			X	X			For material to be used in field and laboratory experiments
6. Formulation work (Kenya)		X	X				X			Designed to increase conidial longevity in the field
7. Simulated farmer store trial (Kenya)			X	X	x	x				Experimental store
8. Farmer store trial (Kenya)							X	X		Authentic store
9. Conidial longevity work (UK/Kenya)	X	X		X	X		X	X	X	Field work in Kenya would be desirable.
10. Secondary cycling studies (Kenya)		x	x	x	x	x	x			To test for occurrence and effects of secondary cycling

X = Priority work, x = work organised around time constraints of priority work, hashed areas = unfunded activities

Additionally, consideration needs to be given at this stage to establishing contacts with potential manufacturers and distributers of a control technology. This is because, even though control has not been achieved yet, businesses may be able to provide advice and, possibly, funding and collaboration at the near market stage. Contacts have already been established in Southern Africa through the LUBILOSA work.

7. Recommendations

Findings

- As a scientific process the project has been well undertaken. Not all procedures have been carried out to the planned schedule, and some of the problems encountered have been explicable in terms of the difficulties of operating in developing countries. However, the PM now appears to have been over-ambitious.

- The project has been successful in disseminating its findings, via an impressive range of scientific publications and contacts with other researchers in the field.

- However, communication with potential manufacturers and distributers of this biological control technology has been much less intensive than with the scientific community. Although there are valid reasons for a degree of caution in the dissemination of information to the private sector about scientific work in progress (e.g. patenting issues), eventual uptake pathways will depend on businesses being prepared to invest in the near-market stages of development of the formulations, applications techniques, manufacturing, distribution and marketing. Also until some level of control has been demonstrated in the field it may be premature to start discussions about future supply of the biocontrol agent.

- There are concerns about the departure of Susan Smith and the possibility that over the next 24 months the scientific leadership of this project at CABI ARC, while extremely competent, may be overstretched[3].

- It remains very uncertain as to whether an economically viable pest control technology will result from this research. Field trials involving farmers will not be able to begin until the year 2000 at the earliest. Key questions remain unresolved about the virulence of the isolates, the formulations which might be used, and methods for their application.

- In terms of developmental impact, the project still has a high risk status, although this could fall significantly if favourable results flow from the work planned for 1999/2000. The high risk status has to be balanced against the possibility of very high developmental impact which could result from the development of viable pesticides for use on stored grain based on entomopathogenic fungi.

[3] Since our initial discussion Dr Moore has informed us that CABI ARC are in the process of recruiting an additional insect pathologist/ entomologist, who will cover Dr Oduor's other projects enabling Dr Oduor to commit to the time input expected.

Recommendations

- Recognising Project R6773 as an expensive "high risk/high payoff" investment, funding should be supported as envisaged in the PM for a further 24 month period March 1999 - March 2001, subject to some redesign to increase the likelihood of a development payoff.

- The redesign should include:

  - Formalising a revised Activity Plan along the lines of the discussion above of Table 4 and the discussion below.

  - Information dissemination activity should be partly refocused on potential manufacturers and distributers of the technology. This should be aimed at engaging their interest and it would be desirable to obtain private investment commitments to the near-market stages of the research.

  - Thinking through the socioeconomic work, including the conduct of farmer trials and the estimation of the regional significance of the results of the trials.

  - Assurance that satisfactory arrangements have been made to cover Susan Smith's role.

  - Tighter management control to ensure that funds for activities not covered during the proposed timeframe are retained and used when time permits. This will help to ensure that agreed outputs are achieved within the budget. Any changes to the original PM which may affect activities and funds, need to be agreed by CPHP programme manager as and when they arise.

- The redesign may imply some input into the project by social scientists with expertise in (i) farmer trials and (ii) business development expertise to develop and manage links with potential private sector investors.

8. References

Brower, J.H., Smith, L., Vail, P.V. and Flinn, P.W. (1995). Biological Control. In *Integrated Management of Insects in Stored Products* (eds Bh. Subramanyam and D.W. Hagstrum). Marcel Dekker, Inc. New York, pp. 223-286.

Yudelman, M., Ratta, A. and Nygaard, D. (1998). Pest management and Food Production: Looking to the Future. *Food, Agriculture and the Environment* Discussion Paper #25, IFPRI, Washington DC.

Appendix 1: List of dissemination outputs from Project R6773 - The control of storage pests of maize with entomopathogenic fungi such as *Beauveria bassiana*

Submitted to refereed journals

S.M. Smith, D. Moore, L. Karanja and E. Chandi. 1999. Formulation of *Beauveria bassiana* in vegetable fat pellets for control of the larger grain borer, *Prostephanus truncatus* (Horn.) *Pesticide Science* (in press).

Oduor, G.I., Smith, S.M., Chandi, E.A., Karanja, L.W., Agano, J. and Moore, D. Occurrence of *Beauveria* spp. on insect pests of stored maize, e.g. *Sitophilus zeamais* Motsch. in Kenya. Submitted to *Journal of Stored Products Research*

Oduor, G.I., Smith, S.M., Chandi, E.A., Karanja, L.W., Agano, J. and Moore, D. Maize and its local storage in Kenya in relation to infestation by storage pests e.g. *Sitophilus zeamais*. To be submitted to *Journal of Stored Products Research*

Smith, S.M., Karanja, L.W., Chandi, E.A., Agano, J.O. and Oduor, G.I. Testing the pathogenicity of the fungi *Beauveria bassiana* amd *Metarhizium anisopliae* to *Prostephanus truncatus* and *Sitophilus zeamais*. In preparation

Smith, S.M., Karanja, L.W., Chandi, E.A. and Oduor, G.I. The effect of mixtures of *Beauveria bassiana* and wood ash on the mortality of *Prostephanus truncatus*. In preparation.

Oduor, G.I., Smith, S.M., Chandi, E.A., Karanja, L.W., Agano, J. and Moore, D. Evaluating the potential of *Beauveria bassiana* to control storage pests of maize in the field I. The effect on insect populations. In preparation.

Smith, S.M., Oduor, G.I., Chandi, E.A., Karanja, L.W., Agano, J. and Moore, D. Evaluating the potential of *Beauveria bassiana* to control storage pests of maize in the field II. Establishment of fungal infection. In preparation

Published conference proceedings and reviews

S.M. Smith, G.I. Oduor and D. Moore (in press). Preliminary investigations into the potential of entomopathogenic fungi for the control of pests of stored maize. Proceedings of IOBC Working Group on Microbial Control in Sustainable Agriculture, August 10-16, 1997, Copenhagen, Denmark.

Susan M. Smith, George. I. Oduor, Ephraim A. Chandi, Lucy W. Karanja, John, O. Agano and Dave Moore (in press). The potential of the insect pathogenic fungus *Beauveria* sp. for control of storage pests of maize in Kenya. Second Biennial Crop Protection Conference (KARI/DFID NARP II), Nairobi, Kenya, 16-17 September 1998.

Moore, D., Smith, S.M., Lord, J.C. Pathogens in Stored Product IPM. In: B. Subramanyan and D. Hagstrum (eds) Alternatives to Pesticides in Stored Product

IPM. Kluwer Academic Publishers, Norwell, Massachusetts, USA. (A chapter in prep.)

Conference presentations

Smith, S.M., Chandi, E.A., Karanja, L. and Moore, D. Control of the larger grain borer, *Prostephanus truncatus* with *Beauveria bassiana*. Presented at *30th Annual Meeting of the Society for Invertebrate Pathology, Banff, Alberta, Canada, 24-29 August, 1997.*

Susan M. Smith, George. I. Oduor, Ephraim A. Chandi, Lucy W. Karanja, John, O. Agano and Dave Moore. Investigating the use of *Beauveria* sp. for control of pests of stored maize in Kenya. Presented at *VIIth International Colloquium on Invertebrate Pathology and Microbial Control in Sapporo, Japan, August 23-28, 1998.*

George Oduor. Current status and promising results of stored grain pest control research at CABI. Presented at *East and Central Africa Maize and Wheat (ECAMAW) Working Group on Controlling Post-Harvest Losses in Maize and Wheat, Addis Ababa, Ethiopia, 28 September - 1 October, 1998.*

Oduor, G., Smith, S., Chandi, E., Karanja, L. Aganoa, J. and Moore, D Survey for fungal pathogens of storage pests infesting maize in Kenya; first records of beauveria spp. Presented at *7th International Working Conference on Stored-product Protection (IWCSPP), Beijing, China October 1998.*

Dave Moore, Susan M. Smith, George I Oduor and Jeff K Waage. Controlling pests of stored maize with fungi - are the problems real or perceived? Presented at *Annual Meeting of the Entomological Society of America, Las vegas, USA 8-12 November, 1998.*

Appendix 2: Bar chart of project activities

	1 Dec 96-31 March 97	1 April 97 - 31 March 1998				1 April 98 - 31 March 1999				1 April 99 - 31 March 2000				1 April 2000 - 31 March 2001			
	1	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Purchase and installation of equipment	■																
Survey for isolates		■	■	■	■												
Collecting of isolates *					■	■	■	■									
Bioassays of isolates *		■	■	■	■	■	■	■	■	■	■						
Characterisation *	■		■			■	■	■	■	■	■						
Biological studies on isolates			■	■	■	■	■	■	■	■	■						
Formulation development		■	■	■	■	■	■	■	■	■	■						
Laboratory and small scale field, population regulation			■	■	■	■	■										
Small scale mass production		■			■												
Project Review March 1998					■												
Mass Production						■	■	■	■	■	■						
Field trials						■	■	■	■	■	■	■					
Conidial Modelling						■	■	■	■	■	■	■	■	■	■	■	■
Project Review March 1999									■								
Information dissemination				■			■	■			■			■	■	■	■

* Activity on these will be greatly reduced after March 1998 with only incidental additions to the isolate collection