

## **CROP PROTECTION PROGRAMME**

**Novel technologies for the control of the African armyworm *Spodoptera exempta* on small holder cereals in East Africa developed, evaluated and promoted**

**R7954 (ZA0460)**

### **FINAL TECHNICAL REPORT**

**1 January 2002 – 1 July 2004**

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### **Executive Summary**

*A very brief summary of the purpose of the project, the research activities, the outputs of the project, and the contribution of the project towards DFID's development goals. (Up to 500 words).*

African armyworm is a serious migratory pest of pasture and grain crops in Africa and outbreaks occur every year in primary outbreak areas of Tanzania and these can, under favourable conditions then move across Tanzania and migrate to many parts of East and Southern Africa. Existing controls based upon chemical insecticides are too expensive to meet more than 30% of the needs of poor farmers largely due to cost and availability. This project, in collaboration with the Tanzanian Ministry of Agriculture, sought to develop and evaluate alternative novel controls based upon the use of a natural pathogen of armyworm, the *Spodoptera exempta* nucleopolyhedrovirus (SpexNPV) and a local botanical neem.

The project has demonstrated that SpexNPV can be used to control armyworm outbreaks as effectively as chemical pesticides whether applied by ground or aerial application systems currently used in Tanzania. This is the first use of aerial application for any NPV biopesticide in Africa. Infusions of neem leaf and seed, widely available to small farmers in outbreak areas although not as effective as chemicals or NPV also has control potential. The project has also shown that by spraying outbreaks of armyworm with SpexNPV and harvesting the killed insects substantial quantities of NPV can be collected at low cost. The project, through collaboration with EMBRAPA (soybean research Institute) in Brazil, has evaluated the cheap low technology NPV formulations developed by EMBRAPA and now used for pest control on 2 million hectares. The conclusions are that this technology could be adopted in Tanzania to produce SpexNPV at less than 6 US\$ per hectare in place of current chemical cost of 16.50US\$. This work has attracted the support of and part funding by the migratory pest programme of USAID.

Ecological studies have confirmed that SpexNPV is widely present in armyworm but mainly as a covert non symptomatic infection, the first record of such an NPV infection in Africa. This work has led to important new insights into host/virus relations in migratory insects and may be of major importance in understanding armyworm population dynamics. A large number of SpexNPV isolates have been collected and characterised and study of their genetics and biology is continuing with UK research council funding.

The Tanzanian Ministry of Agriculture has directed its armyworm control agency to develop a strategy to adopt SpexNPV in place of chemical control. It is also implementing and evaluating the use of neem in a community armyworm forecasting and control project co funded by the USAID and the Government of Tanzania.

## Background

Tanzania is one of the poorest countries in Africa with a per capita GNI of \$270 and 35% of the population below the national poverty line. Agriculture accounts for about 45% of the GDP, 65% of the total exports, and occupies 80% of the employed population. The poverty reduction strategy paper recognises agriculture as critical to the Tanzania Development Vision. The country has a dual agricultural economy with a smallholder sub-sector and a commercial/large scale sub-sector. Smallholder farms of rain fed agriculture dominate the agricultural landscape, and are estimated to number around 4.6 million. They account for most of the food grown in the country. Food crops dominate agricultural production, with maize by far the single most important cereal crop. However, recent increases in area planted to maize have not been matched by increased production (figure 3), indicating reduced yield/ha. Cereal yields in East Africa are highly variable due to the vagaries of climate and pest attack, amongst other reasons. This variability is a key constraint to rural development, causing periodic crisis and hardship for rural communities.

An important pest of cereals in East Africa is the African Armyworm *Spodoptera exempta* (*Lepidoptera: Noctuidae*) is a migratory pest of cereals, pasture and rangeland and is widely distributed in Africa, south of the Sahara. East, central and southern parts of the continent are the most seriously affected. The adult moth migrates with prevailing winds, so outbreaks are linked to the seasonal progression of the Inter-Tropical Convergence Zone and the associated rainfall. In East Africa the first outbreaks of the season are usually in central or northern Tanzania or southeast Kenya. From there the outbreaks tend to spread north and west to other parts of Tanzania and Kenya, and to Burundi, Rwanda, Uganda, Sudan, Ethiopia, Eritrea, Somalia and Yemen. A southern spread of outbreaks from southern Tanzania to Malawi, Mozambique, Zambia, Zimbabwe, Botswana, Swaziland and South Africa also occurs, but there is much less known about this progression.

**Table 1. Armyworm outbreaks in Tanzania (1989-2002)**

Season Year	Area Infested (Hectares)	Number of Districts affected (out of total of 113)
1989/1990	28,768	27
1990/1991	15,214	20
1991/1992	517,233	54
1992/1993	34,844	39
1993/1994	45,504	31
1994/1995	4,798	22
1995/1996	3,187	13
1996/1997	577	5
1997/1998	35,174	17
1998/1999	311,560	44
1999/2000	50	1
2001/2002	157,942	26

Poor farmers are especially vulnerable to armyworm outbreaks. The majority of cereals in East Africa are produced in the long rains (January-May) when armyworm outbreaks are most frequent. The losses due to armyworm are virtually zero in some years and immense in others. Yield losses within an outbreak area may be very high although 30% loss in recently planted maize has been estimated as an average loss (Scott 1991).

Reducing the impact of armyworm attack in Tanzania currently hinges around the timely supply and use of effective agro-chemicals, supported by a centralized forecasting unit (Pest Control Services, PCS). Improved forecasting and control of armyworm is aimed at minimizing the

damage caused by the pest, and so improving rural livelihoods. The most widespread method of controlling armyworm outbreaks is to spray the larvae with chemical pesticides. Insecticides are effective (Rose *et al.*, 2000) and due to the sporadic nature of outbreaks, selection pressure for resistance to pesticides is probably low.

In addition to the usual approach to control by spraying pests that are damaging a crop, studies on the progression of armyworm outbreaks led to the development of an approach known as 'strategic control' (Cheke and Tucker, 1995). As the origin of many outbreaks can be traced back to earlier outbreaks upwind controlling earlier outbreaks could reduce the occurrence of later outbreaks. Strategic control therefore entails finding and controlling 'critical' outbreaks that would otherwise lead to subsequent outbreaks. In practice the logistic and financial constraints have meant that this approach has never been fully implemented.

Due to the catastrophic nature of outbreaks in some years, control of armyworm has often been seen as the responsibility of central governments. In Tanzania the government has frequently distributed pesticides and spraying equipment to the affected areas. For example, during the 1991/92 cropping season, 80,000 liters of insecticides together with 4995 knapsack sprayers and 1000 ULVA sprayers were distributed. Unfortunately this was still far short of what would have been required to control the outbreaks.

In Tanzanian governments Pest control services (PCS) of the ministry of agriculture and food security (MoAFS) is responsible nationally for armyworm control. In 1996 it requested NRI to join in a collaboration to explore novel ways of controlling armyworm. The MoAFS felt that increasing concern about the environmental effects of wide scale pesticide use and the high cost of imported pesticide used for control made long term reliance on chemical controls unwise. It was decided to seek to develop better controls based upon biological control.

Among the most promising was an NPV virus was identified as an important pathogen of armyworm by Brown & Swaine (1965) and Swaine (1966). This NPV is a naturally occurring specific virus of *S. exempta* that causes a highly infectious disease of the armyworm. The NPV is rarely apparent in primary outbreaks of the pest only appearing later in the season after which it spreads rapidly. In the later stages 98% of outbreaks show infection and it frequently causes the collapse of later outbreaks (Rose *et al.* 1997). Some observations later suggested that although NPV is found naturally to occur in armyworm outbreak areas it can be highly localised effecting only small parts of the outbreak area (McKinley 1975).

Under a previous project R6746 the SpexNPV was evaluated in the laboratory glasshouse in UK and finally in 1999 in the field in Tanzania. These showed that this agent could kill armyworm when applied using locally available equipment. The early trials indicated that an application rate of  $5 \times 10^{11}$  OB per hectare could kill armyworm. As this was equivalent to the virus load of only 50 infected larvae it seemed to require only a minimal dose that would be relatively cheap to produce.

These promising results encouraged CPP to set up a follow on project to pursue this work further. The main addition to the work was to include components to look at the use of local botanical neem as a possible tool in armyworm management.

Although armyworm is a pest all over east Africa the project in this phase decided to continue to concentrate its technical research in Tanzania which had both a good infrastructure for armyworm work and where the government agencies were very supportive of novel control and contributed greatly to the progress of the research.

## Project Purpose

The purpose of this project was to develop and evaluate two novel approaches to controlling African armyworm in order to reduce financial losses to smallholder and subsistence farmers and improve food security in East Africa. These approaches were the use of armyworm nucleopolyhedrovirus (NPV), a naturally occurring biological control agent, and neem, a locally available botanical agent. Both could be used to replace environmentally damaging and expensive synthetic chemical pesticides. The project would also develop a strategy to enable these sustainable and appropriate technologies to be available to poor farmers.

## Research Activities

*This section should include detailed descriptions of all the research activities (research studies, surveys etc.) conducted to achieve the outputs of the project. Information on any facilities, expertise and special resources used to implement the project should also be included. Indicate any modification to the proposed research activities, and whether planned inputs were achieved.*

### 1.1 Initial project workshop

A three-day problem specification and planning workshop was held at Arusha, Tanzania, from 9<sup>th</sup> to 11<sup>th</sup> October 2001. 22 participants attended the workshop, representing farmers, District Agricultural Officers, chemical suppliers, extension officers and various Government departments such as pesticide registration and district management. The main aim of the workshop was to explore the current state of armyworm control with regard to forecasting and the control methods used. This information was then used to develop appropriate plans for the testing and introduction of novel control methods. The workshop also identified additional issues to be included in the socio-economic survey. A detailed report describing the workshop process and the giving details of the discussions which took place and the ideas which emerged is attached in Appendix 1.

### 2.1 Bioassay programme

To initially evaluate NPV and neem bioassays of candidate formulations were conducted at NRI using protocols previously described in the FTR of the previous project R6746. Bioassays of wild type SpexNPV as used in field trials was carried out at NRI and reported under this output. Bioassays of different SpexNPV clones were and on different hosts were carried out at CEH Oxford using similar protocols and are reported under the ecology output 5.

An evaluation of the literature on neem and possible botanicals was carried out in 2001. This involved a survey of literature, visits to the GTZ IPM project in Tanzania then actively promoting neem and data sharing with the USAID armyworm project in Kenya who had also tested neem. While no formal report on neem was completed discussion of the results of these activities by the project team led to a decision to go ahead with neem in field trials in the 2002 season.

### Activity3.1 Evaluation of Neem

This activity was a desk study based upon published data and some experimental work. It used data from bioassays carried out in the laboratory on neem as part of the bioassay activity 2.1 along with field data from the USAID funded work of Broza and Brownbridge on the use of neem for armyworm control reports of which were generously provided to the project. Local data from Tanzania on suitability availability and supply of neem was collected by the project socio-economist Dr Oruko as part of baseline studies under activities 6.1, 6.2 and 6.3. Additionally findings from the CPP funded project on botanicals in West Africa (R 7373) were also useful. It had been hoped to draw on the outputs of the GTZ funded project on Neem based in Arusha 1996-2000 but these were not forthcoming.

#### *4.1 Field trials of NPV formulations and neem products*

Field trials of NPV formulations and neem products were carried out in Tanzania in small plot trials in 2001, 2002 and 2004. Field trials had been planned for 2003 but no outbreak materialised due to the drought so the planned trials could not be implemented. The NPV used was produced and formulated in UK using the Tanzania wild type SpexNPV. Initially trials of neem used neem oil as a standard form of known potency to check efficacy of neem against armyworm. Later trials used locally available neem seeds and leaf extracts. Details of these trials are given in the individual field trial reports attached as appendices 2, 3, and 4.

#### *4.2 In large-scale field trials of NPV*

Several large scale field trials of NPV up to 0.5 ha were carried out in 2001, 2002 and 2004 and full details of these trials are reported in appendices 2 and 3. An aerial application trial was also carried out in 2004 and this is reported in fully in appendix 4.

#### *5.1 An investigation of the transmission of SpexNPV 5.2 Prevalence of SpexNPV in Tanzania assessed*

This work was carried out in through fieldwork Tanzania with molecular and genetic studies completed at the Centre for Ecology and Hydrology Oxford. The work started with CPP funding but CEH were able to expand it with additional funding from NERC to a full PhD study jointly supervised by Dr Cory of Oxford and Dr Wilson of Stirling university. This PhD, by Ms E Redman is due for completion in late 2005 and molecular work analysing samples to determine the prevalence of SpexNPV in armyworms from different areas of Tanzania is still underway.

To investigate the prevalence of SpexNPV and the transmission of the virus insects and virus-killed cadavers were collected from Tanzania in 2002, 2003 and 2004. The cadavers were taken back to CEH Oxford for molecular studies and bioassays. From these samples 60 different genetic variants of the SpexNPV were identified using DNA restriction endonuclease analysis. A number of these were cloned using in vivo cloning and bioassays run to evaluate their pathogenicity and speed of kill against the standard (mixed) wild type NPV.

Live insects collected in Tanzania were used to initiate a laboratory colony which could be used to monitor the presence of asymptomatic NPV infection. A reverse transcriptase polymerase chain reaction (RT-PCR) technique was developed for the detection of vertically transmitted SpexNPV in the different armyworm stages. The PCR targets two genes, the polyhedrin gene, which codes for the NPV polyhedra structural protein, and the *lef 8* gene, which is an RNA polymerase. This technique allows for the detection of very small amounts of both SpexNPV DNA and RNA in all life stages of the armyworm. Details of the experimental techniques used are given in the report appendix 5.

Ecological work additional to the original PMF focussed on the role of armyworm grazing on the later generation of high levels of cyanide by cyanogenic grasses (the dominant grasses in many areas) and the possible link with cattle poisoning. This interesting aspect of armyworm damage came to the team's attention in 2001 and was followed up by Dr Wilson, without any additional funding in the field seasons 2001-2002. This study used cyanide test kits to assess levels of cyanide in grasses, armyworms feeding on them after outbreaks of armyworm in 2001 and 2002. Experiments also looked at cyanide contents of armyworm feeding on cyanogenic grasses and non cyanogenic crops in the laboratory details of these are presented in appendix 19.

#### *6.1 A strategy for locally producing NPV developed*

#### *6.2 Field production of the NPV developed*

#### *6.3 Methods for harvesting and storing NPV developed*

The above activities were carried out through field and laboratory trials as well as visits of PCS staff to Brazil to study low cost production and visits by Brazilian staff to Tanzania. Detailed

methods are reported in appendices 2, 3, 4 8 and 26. Some activities were additional to the original project and were co-funded by USAID.

#### *7.1 A baseline socio-economic study of current armyworm control measures 7.2 A Socio-economic study to assess the feasibility of the new technologies*

A socio-economic survey was designed to study current control measures and to assess the feasibility of new technologies was carried out between 2002 and 2004. The survey was implemented in four districts, Hai and Korogwe in the Northern Zone, and Dodoma and Kilosa in the Central Zone. The districts were selected during the stakeholder workshop held in October 2001. The survey was carried out in two stages (1) Individual farmer interviews (2) Focus group discussions. The survey was led by the CABI ARC socio-economist Dr Jemimah Njuki and Tanzanian armyworm co-ordinator Mr Mushobozi. A full report on the socio-economic activities with the survey tool is attached in appendix 6.

#### *8.1 International dissemination to scientists (papers)*

To disseminate the projects findings to scientists several avenues were used, including (1) oral presentations to scientific meetings. (2) articles in scientific periodicals and (3) peer reviewed papers. Briefing notes on the project and press statements were produced and circulated (see appendices 21 & 22) to Tanzanian and world press agencies and websites. Following this various reporters/writers contacted project staff or visited PCS Arusha and NRI for interviews and stories in several scientific journals (New scientist, Appropriate technology, British Industry News and International Pest Control) were produced on the work of project (see appendices 9-15).

#### *8.2 Regional dissemination to extension services & scientists*

To disseminate to extension services and scientists in the region a number of information leaflets have been produced and distributed. These are attached as appendices (20, 21, 22 24). Mass media in Tanzania and East Africa have also been targeted using press releases to TV and newspapers. National TV news filmed the aerial spray trials in 2004 and this formed the basis of an item on national news. The BBC world service were approached and showed interest in filming the field trials in 2004 but filming schedules did not coincide with this year's armyworm outbreak. It had been intended to hold a final project workshop in early 2004 but due to absence of outbreaks in 2003 the trials planned for v2003 had to be carried out in February-may 2004 leaving no time for a final workshop. This is now planned for early 2005 under a planned extension.

### **Outputs**

*The research results and products achieved by the project. Were all the anticipated outputs achieved and if not what were the reasons? Research results should be presented as tables, graphs or sketches rather than lengthy writing, and provided in as quantitative a form as far as is possible.*

All the major outputs of the project except the original plan to have a final workshop were achieved. The plan to have a final workshop was changed after discussions with the CPP because the failure of any armyworm outbreaks to appear in the 2003 season meant the extensive series of planned field trials for that year could not take place. To complete the scientific trials therefore the 2004 season, originally intended only for dissemination and demonstration outputs, was used to carry out the field trials of neem and NPV as well as the NPV production trials previously planned for 2003.

In 2004 additional aerial spray trials not originally planned were undertaken as a result of additional USAID funds to carryout this aspect of the work. The additional USAID money also funded a visit by Brazilian expert Dr F Moscardi to Tanzania to instruct PCS staff in low cost NPV production.

The objective of evaluating and developing SpexNPV as a novel control against armyworm was fully achieved and this is now in the process of adoption as National policy by Tanzania. The neem work was less advanced but promising field results have encouraged USAID to fund trial evaluation in five districts of Tanzania.

### **Output 1. Initial project workshop report.**

The project workshop set the scene for the main project and enabled the outputs and activities to be discussed in detail with stakeholders. Its discussions were especially useful in defining the issues for the socio-economic work on developing a strategy for implementing novel control. Brainstorming was used to identify key variables affecting all aspects of armyworm management.

It in particular highlighted important areas later developed in the socio-economic work concerning the vulnerability of key groups of farmers and the factors that determined their vulnerability. Key issues highlighted were:

1. The financial constraints to armyworm control by poor farmers.
2. The lack of access to agro-inputs in poorer districts
3. The limited access to sprayers in poor areas and the shortcoming of existing provisions for armyworm control.
4. Availability of products such as neem was thought to be very limited and needed to be quantified.
5. Costs of some pesticides were high and in short supply during outbreaks.
6. The lack of funding at all levels severely limits the ability of national and local government to offer support to farmers. Farmers tend not to budget for uncertain events like armyworm outbreaks. Some farmers lack both access to extension officers and the resources to treat armyworm infestations.
7. A key point identified was that the resources of farmers determined what technologies they could use for control. Larger farmers or cash cropping families have access to ULV and LV sprayers and the cash to buy pesticides. Small subsistence farmers have to share spray equipment if they have access at all and little money to buy pesticides. The desirability of developing controls not needing sprayers (bait based) was considered important if these smallholders were to be helped.

The workshop helped to define the issues the socioeconomic survey was to address. In particular it identified the need to examine the feasibility and resources available to different groups of farmers to carry out control.

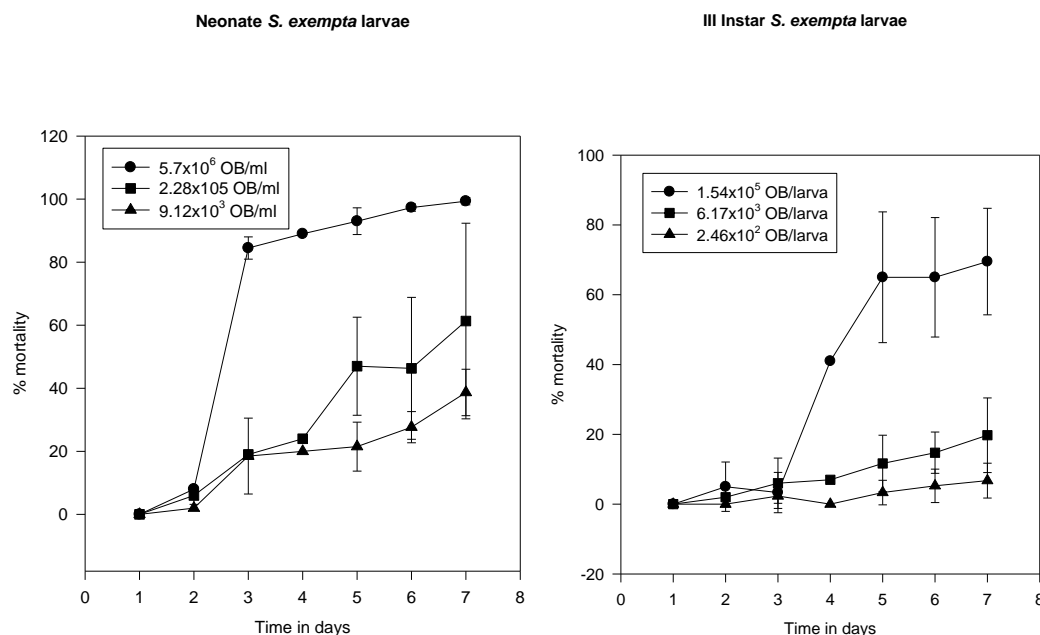
A detailed report describing the workshop process and the giving details of the discussions which took place and the ideas which emerged is attached in Appendix 1.

### **Output 2 Laboratory evaluation of candidate Neem and NPV formulations**

The laboratory work programme continuing from the earlier project confirmed that SpexNPV was highly pathogenic to all instars of SpexNPV from neonates to III instar and that importantly could produce high mortalities within 4 days after application. Such a rapid action was essential if damage to crops was to be prevented. However the results confirmed that if application was delayed until the larvae reached IVth instar then overall kill was reduced to less than 60% even with high rates (5 x10<sup>12</sup> OB ha) of application. Thus while laboratory results showed that SpexNPV could be highly effective in the laboratory they emphasized the crucial importance of confirming its efficacy and reliability under operational field conditions.



**Fig 1 Bioassays of newly hatched and III instar *S.exempta* showing mortality over time.**



In collaboration with CEH Oxford there was some work on producing bait formulations of SpexNPV. However it was found that baiting with SpexNPV had a number of practical problems. Baits with SpexNPV could be produced using homogenized wheat seedlings in an agar gel and were acceptable to armyworm larvae when fresh. However when tested in the field it was found the baits dried out rapidly and became unattractive to larvae. It was considered difficult to produce a bait significantly more attractive to larvae than the new flushes of grass they normally attack. Also the amount of bait needed to treat large areas would have to be considerable (>10kg/ha). The storage at the necessary controlled temperatures (4-8 deg C) distribution and application presented serious problems.

Since no appropriate bait formulations existed that met the requirement and developing such a new formulation was not within the resources or the outputs of this project no further work on this was pursued.

Bioassays of the efficacy of different SpexNPV clones and determining the efficacy of SpexNPV on different hosts were carried out at CEH Oxford and are reported under the ecology output 5.

Laboratory tests on neem oil produced in Tanzania confirmed that it was effective as an aqueous suspension at 1% in controlling armyworm larvae. The laboratory work on neem was at an early stage when we were able to access the outputs of a USAID project on area wide armyworm control led by Broza that had just ended. As part of this project both neem and other alternatives to pesticides had been evaluated in Kenya. This enabled the team to get sufficient evidence of neem's efficacy to move ahead to field trials with neem in 2001. The laboratory programme of neem work was then halted in favor of field based work in Tanzania.

### **Output 3 A review of the potential of neem against armyworm**

The research on the use of botanical insecticides such as Neem against African armyworm is limited though more is known of its activity against other Spodoptera species in Asia (Schmutterer 1995). There has been some work on *S.exempta* using purified azadiractin by Isman (1995 & 1999) showing that

compounds in neem are highly active as anti-feedants and moulting disruptors. The perennial problem with neem though is that locally available sources (either neem leaf or seeds) may vary in activity depending upon strain and growing conditions. A USAID project led by Broza and his team (1999) working in Kenya had showed that 5% neem seed extracts from East Africa gave satisfactory control on rangeland and in maize. There have also been successes reported with use of botanical insecticides including Neem by local farmers in Tanzania. There was a GTZ funded PM project up until 2001 to promote the use of neem in vegetable farming though this has since finished. The project also benefited from the outputs from CPP project R7373 that looked at optimising the use of botanicals in Ghana.

It was known that neem is grown widely in some areas of Tanzania as a shade plant. However much of this is in the coastal lowlands far from the outbreak areas so that supply might be a serious issue. Some is grown in armyworm outbreak areas inland but is considered by farmers and crop protection extension officers to be of inferior quality. This linked in with reports gathered from farmers during the workshop and socio-economic survey about the poor efficacy of the neem from outbreak areas such as Dodoma.

Thus in respect of neem there were some real questions concerning both its efficacy and issues of supply that needed to be answered before its utility as an armyworm control for Tanzania could be determined. However if it did prove efficacious and supply was present in outbreak areas it would offer a simple cheap and available tool for poor smallholders who had little or no access to other control technologies. The work to answer to these questions is covered in outputs 4 and 6 below.

#### **Output 4 Field trials of NPV and Neem for control of armyworm completed and evaluated**

This output was the major part of the work in 2001, 2002 and 2004. In 2003 there were no outbreaks so that the field work for this planned final field season was postponed to 2004 just before the end of the project. The trials in 2004 included an aerial application trial that was not included in the original PMF but was made possible due to additional support for the novel control work from USAID (Project 50-4001-3 F037). From work in a previous CPP funded project (R5746) we had some limited field data that showed that SpexNPV applied at between  $1 \times 10^{11}$  to  $5 \times 10^{12}$  infective particles or occlusion bodies (OB) per hectare could produce mortalities in excess of 90% in treated armyworm in the field (Grzywacz 1999).

The first major objective was to identify the lowest effective application rate of SpexNPV. A rate of  $1 \times 10^{11}$  OB per hectare would require the NPV from only 25-50 infected larvae while  $5 \times 10^{12}$  OB per hectare would require at least 2500 infected larvae and the cost of such a rate might easily be prohibitive.

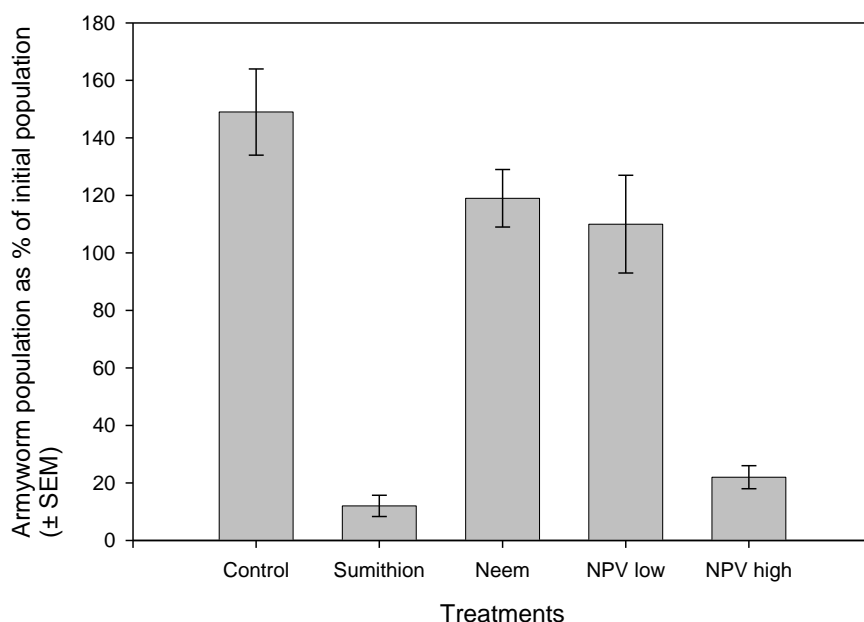
Trials were all initially run on pasture, as it was felt that moving onto crops before the technologies were tested and evaluated was too risky a strategy. In 2003 and 2004 maize trials were also planted but in 2003 no outbreaks occurred and in 2004 the trial was aborted when natural SpexNPV appeared in the control patches. These occurrences emphasize the difficulty of getting good data on armyworm whose outbreak patterns are so temporally and spatially uncertain. The extensive natural occurrence of SpexNPV can also make it difficult to assess the effectiveness of artificial application in some years.

The 2001 field trials consisted of a replicated small-plot trial and a single treatment large plot trial on pasture. The trial included 3 different rates of SpexNPV as well as a control and an insecticide. However heavy rainfall on the trial sites shortly after application produced a massive decline in all treated plots so made the results meaningless. The large plot trial used only one SpexNPV application rate  $1 \times 10^{12}$  OB per hectare compared to a control. The NPV treatment led to a 96% reduction in armyworm numbers by day 7 after application, though the armyworm counts in the control plot fell by 76% so although the difference was significant it wasn't conclusive. A problem

with slower acting agents such as SpexNPV or IGR insecticides is that when armyworm populations are high, as here  $> 600 \text{ m}^2$ , unless the grass is lush in uncontrolled plots larvae eat out all the grass within a few days then migrate from the control plots making comparisons difficult. Full details of results are given in trial report appendix 2.

In the 2002 armyworm season infestations were widespread with 157,000 ha in Tanzania being infested during the season. It was therefore possible for the project to set up three separate field trials. These included a replicated small plot trial to test application rates, and two larger trials of motorised mistblower application. Full details of the trials are given in Appendix 3. The results of the replicated plot trials were very encouraging and showed that SpexNPV applied at a rate of  $1.0 \times 10^{12}$  occlusion bodies, (OB) /ha is capable of controlling armyworm outbreaks as effectively as insecticide (fig 2).

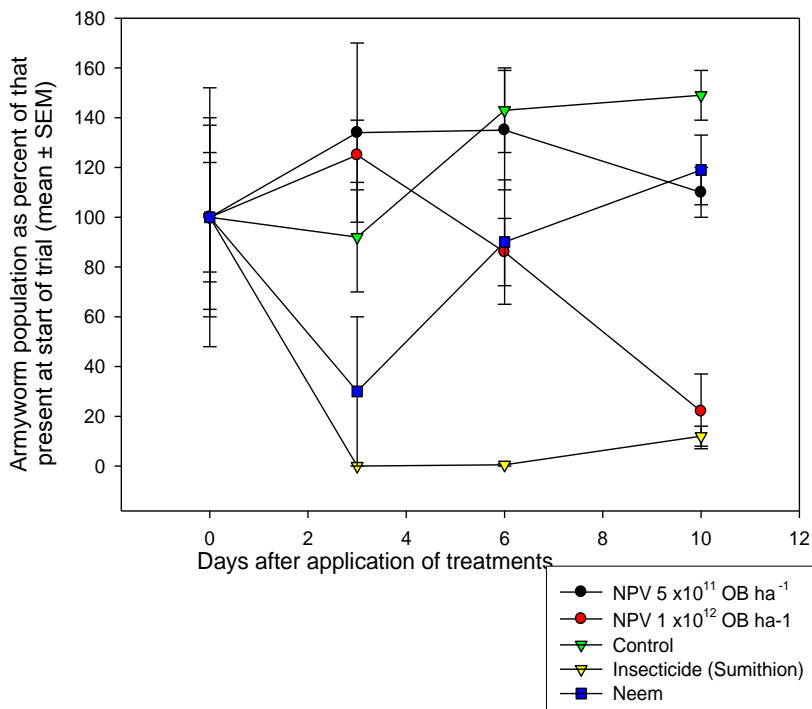
**Figure 2 Counts of armyworm numbers 10 days after application of different treatments to replicated plot trial Luckylucky farm Tanzania 2002.**



A low application rate of  $5.0 \times 10^{11}$  OB/ha was also evaluated but was found to be ineffective. In the previous replicated trials reported on in 1999 (Parnell & Dewhurst NRI 245), application rates of  $1.0 \times 10^{11}$  OB /ha and  $5.0 \times 10^{11}$  OB/ha had shown that good control of armyworm. In the trial reported on here however, a dose of  $5.0 \times 10^{11}$  did not perform well in control of larvae. It was conclude that a rate of  $5.0 \times 10^{11}$  OB/ha should be not be considered for control purposes unless further evidence establishes it as effective. It may be noted here that application rates for a number of other Lepidopteran NPVs used for field control are similar to a level of  $1 \times 10^{12}$  OB per ha (*Mimestra brassicae* NPV  $1 \times 10^{13}$ , *Helthios zea* NPV  $1.5 \times 10^{12}$ , *Helicoverpa armigera*  $1.5 \times 10^{12}$ , *Spodoptera exigua* SpexNPV  $1 \times 10^{12}$ ).

If the time course of the experiment is examined as shown in figure 3 it can be seen that while the insecticide acts rapidly and knocks down populations by day 3 in the SpexNPV at  $1 \times 10^{12}$  this took 10 days to achieve an equivalent reduction.

**Fig 3. Results of replicated plot trial shown with armyworm counts as percentage of counts in plots at start of trial.**



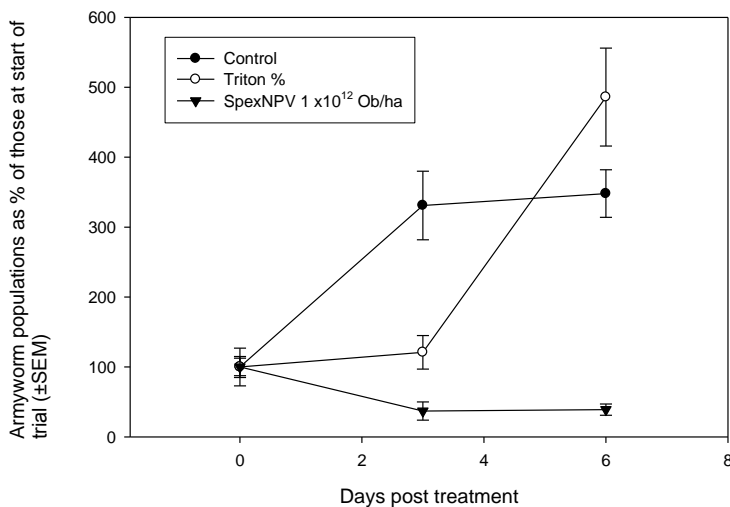
The slower speed of action of NPVs over synthetic insecticides was a known limitation. Previous replicated trials in 1999 had shown that if applied early enough good control could be achieved by 5-7 days. The 6-10 day delay seen here if not improved on could present a problem to using SpexNPV on crops though might be acceptable for strategic control on rangeland.

In this replicated trial neem was also tested. To test the use of neem it was decided to use a high quality product neem oil, to avoid the possible inconsistencies of local seed or leaf extracts, reported by other CPP projects on neem use in Africa (Belmain 2002) to determine that neem per se has sufficient efficacy before moving on to look at low technology local neem infusions. Application of neem in the form of neem oil at 1% to control armyworm showed promise and reduced larval densities within 3 days of application but armyworm numbers bounced back in these plots indicating its control efficacy and persistence were less than that of the synthetic chemical insecticide Sumithion (fenitrothion).

Thus while neem was not as effective as NPV or insecticide it was felt as the socio-economic survey had confirmed neem's widespread availability to poor farmers in some key outbreak districts such as Dodoma that its evaluation showed be continued though focussing on aqueous suspension crushed neem seed and leaf decoctions

The two large scale trials carried out that year one compared SpexNPV to the triton formulation without active ingredient and this showed the blank did not suppress armyworm populations when sprayed onto hatching insects. Here counts fell in SpexNPV treated plot

**Fig 4 Armyworm counts in NPV, blank formulation and control plots at Luckylucky farm 2002**

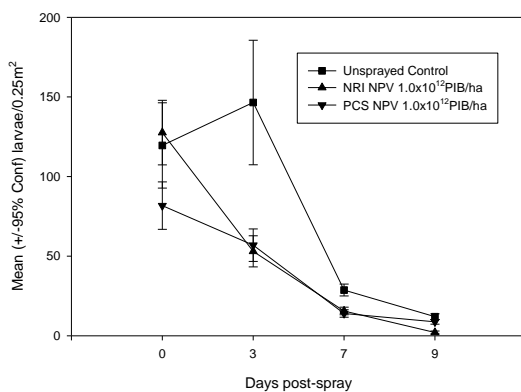


after only three days indicating a more acceptable speed of action could be achieved if application was carried out early enough.

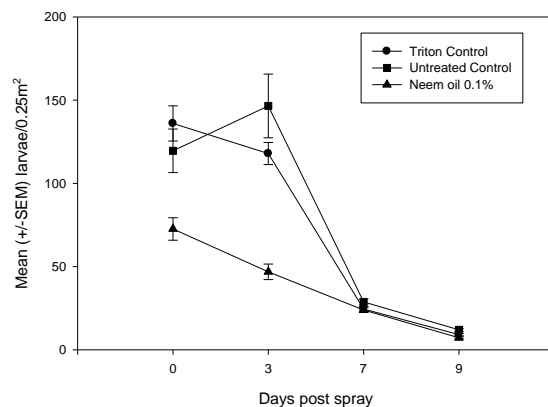
The second large scale trial compared neem SpexNPV produced in a laboratory and SpexNPV harvested the previous year from a field site and stored at ambient temperature for a year. Again a blank formulation of Triton was included.

The results presented in Fig 5 and 6 show that both NPV treatments and the neem produced falls in armyworm counts on treated plots by day 3 and a strong reduction by day 7.

**Fig 5 Armyworm counts in 0.5 ha plot trials at M'ringa 2002, SpexNPV treatments and control**



**Fig 6 Armyworm counts in 0.5 ha plot trials at M'ringa Neem and control at M'ringa 2002**



However control populations also fell by day seven so the results cannot be considered as unequivocal. The movement of larvae out of control plots as a result of high numbers, here approximately 600 per m<sup>2</sup>, consuming all the food and then migrating is a feature in some trials.

The data on infections of larvae showed that the laboratory SpexNPV treated plots NPV infections exceeded 50% but there was also a high background mean level of infection in 18-23% in all the other treatments.

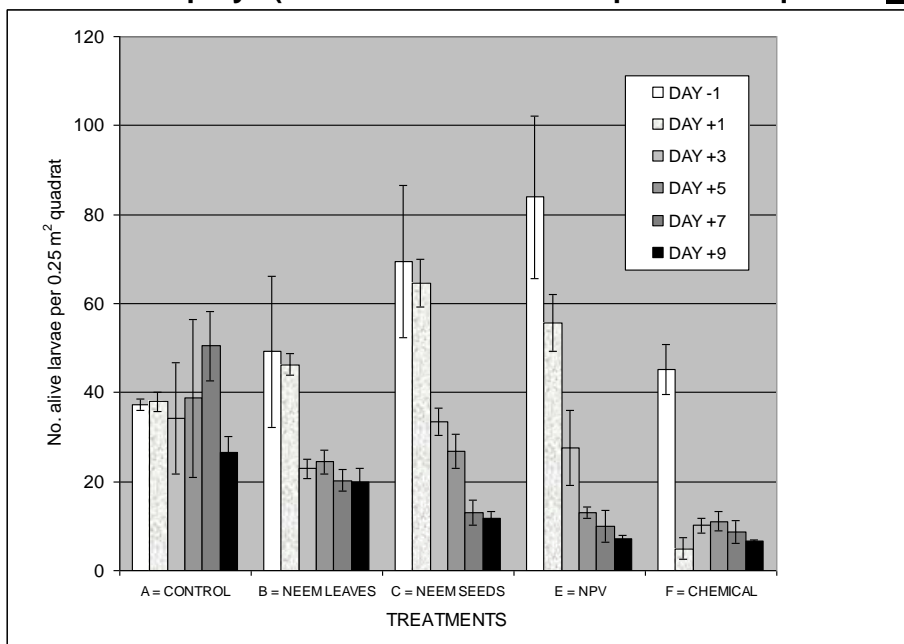
Thus the 2002 trials while showing some promising results did not produce conclusive results showing that SpexNPV could act fast enough or was consistent enough to act as a replacement for insecticides.

The next trials were in 2004 as 2003 was a drought year with no serious outbreaks probably due to the inadequate rains. In this year there were extensive outbreaks in Tanzania and two large scale trials were carried out a ground application trial and an aerial application trial.

The ground spray trial was carried out on 0.5 ha plots on grassland near Arusha. Application of NPV was again at  $1 \times 10^{12}$  OB per hectare using motorised knapsack sprayers the other treatments were neem leaf extract (25 Kg per hectare) and neem seed extract (2.5 Kg per hectare). All details of the trial are in appendix 4.

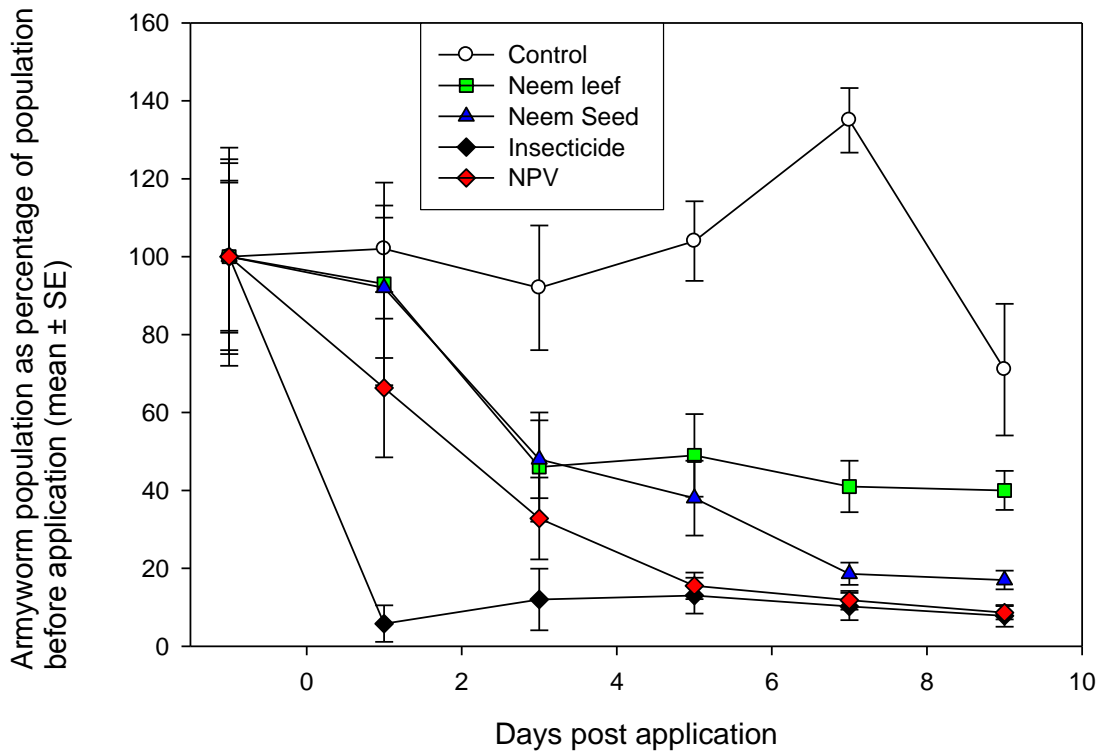
Figure 6 shows the results in terms of armyworm counts per  $0.25 \text{ m}^2$ . Insecticide again acted rapidly producing significantly lower counts than control by day 1 ( ) the NPV by day 5 (Chi squared 53.8,  $df=4$ ,  $P= <0.001$ ). By day 7 both the two neem treatments were significantly different to the control (chi squared 72.35,  $df=4$ ,  $P= <0.001$ ). On day 9 counts in the neem seed plot were significant lower than in the neem leaf treatment (Chi squared 93.17,  $df=4$ ,  $P= <0.001$ )

**Figure 6: Densities of armyworm larvae in each plot following treatment with various insecticidal sprays (mean number of larvae per  $0.25 \text{ m}^2$  quadrat  $\pm$  s.e.).**



as the populations varied between plots these may be more clearly visualised if viewed as in Figure 7 where they are shown as percentages of the starting population.

**Figure 7: Results of ground application of neem and SpexNPV on armyworm populations presented as a percentage of initial populations in each treatment.**



From these results it may be observed that both neem treatments show some promise though they act more slowly than the very fast acting insecticide or the slower SpexNPV. The lower population reduction seen with the neem leaf is a problem for its promotion as a control agent as this is the most available form of neem during armyworm outbreak periods. Outbreaks generally occur in Tanzania before neem sets seed. The difference may be due to the lower level of active ingredient or perhaps a higher proportion of activity as the more volatile fractions that disappear more quickly after application at the generally high (27-35 deg C) ambient daytime temperatures during the outbreak season.

However for farmers lacking access to other controls that have neem and these may account for 50-80% in some outbreak areas neem leaf may be useful in reducing crop damage.

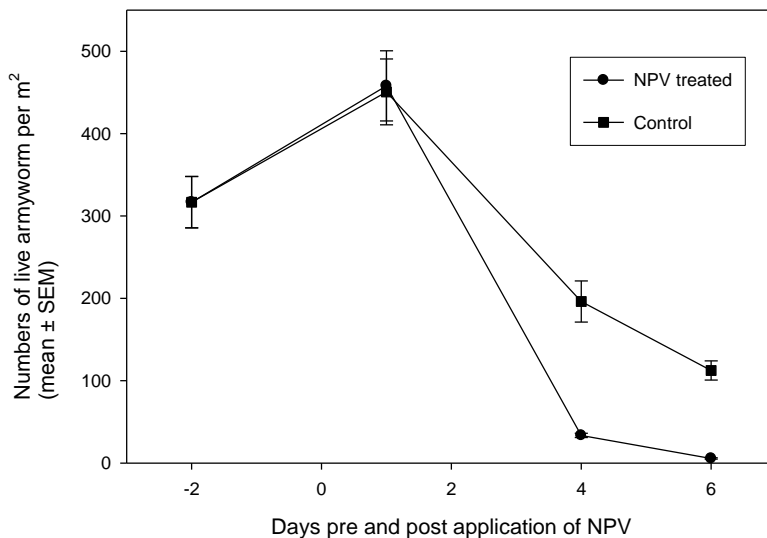
#### *The aerial spray trials*

The aerial trial carried out in 2004 was the first ever aerial application of SpexNPV. It was applied using commercial aircraft using standard hydraulic sprayers at 10 liters per hectare (for full details see appendix 4). The outbreak sprayed on pasture near M'ringa was scouted on the Wednesday seven days after a pheromone trap on a nearby research station recorded high moth numbers. The farmer agreed to let a trial be run and a spray plane applied the NPV on Friday two days after the outbreak was located while the larvae were hatching.

The results of quadrat counts of numbers of larvae at both the sprayed and unsprayed sites are shown in Figure 8. It can be seen clearly that the armyworm population in the NPV plot increased

after initial scouting as larvae continued to hatch out. By the fourth day after application the population of live larvae in the NPV plot fell dramatically and then continued to decline until by day 6 the outbreak declined to nearly zero in the sprayed area. In the control plot the population also declined as larvae matured but remained by day 6 at greater than 100 per m<sup>2</sup> a sufficient level to produce heavy damage to the pasture. Counts in the control were significantly higher than for the NPV plot on day 4 (Chi sq. = 48.65, df = 1, P=<0.0001) and day 6 (Chi sq. = 60 df=1. p = <0.0001).

**Figure 8 Counts of live armyworm in untreated control and NPV sprayed plots at M'ringa Estate NPV trial 17-27<sup>th</sup> March 2003.**



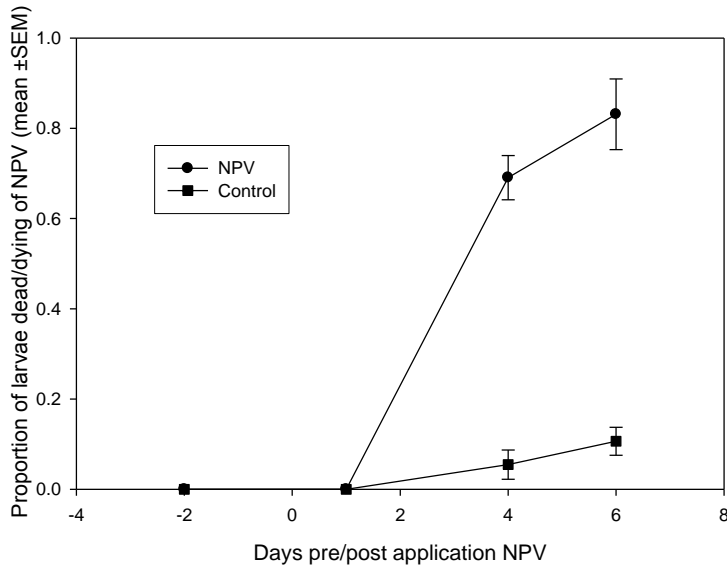
The farm manager noticed that as early as the third day after spraying larvae in the NPV sprayed plot were observed to be “sickly”. It is likely that this early appearance corresponded to the widespread onset of NPV infection and suggests that larval feeding would have ceased in the majority of larvae by day 3 after application. This is important because it has always been put forward as a drawback of some biological pesticides that they act too slowly to be useful in preventing crop damage by pests.

In these trials the SpexNPV showed itself to be relatively fast acting, comparable to that taken by IGR insecticides, and so does seem to be a more promising alternative to conventional pesticides for this pest than in other species. Fungal biopesticides used for migratory pest control can take 7-10 days to kill hosts and NPVs of some other tropical caterpillar pests only become lethal 5-7 days after infection.

In the NPV sprayed plot the decline in live insects was accompanied by the appearance of many NPV infected and killed larvae hanging from grass stems and on the ground (see plate 1). The graph in Figure 9 shows that by day 4 over 70% of larvae counted in quadrats were dead of NPV and this rose to more than 80% by day 6.



**Fig 9 Proportion of larvae in plots killed or infected by NPV in larvae counted in untreated control and NPV treated plots at M'ringa 2004**



In the control areas NPV killed larvae were relatively few at 5% by day 4 and only 11% by day 6. The occurrence of NPV deaths in the control plots appeared noticeably more clumped than that in the NPV plots with hotspots of NPV infection in a few quadrats contrasting with no deaths in most quadrats counted.

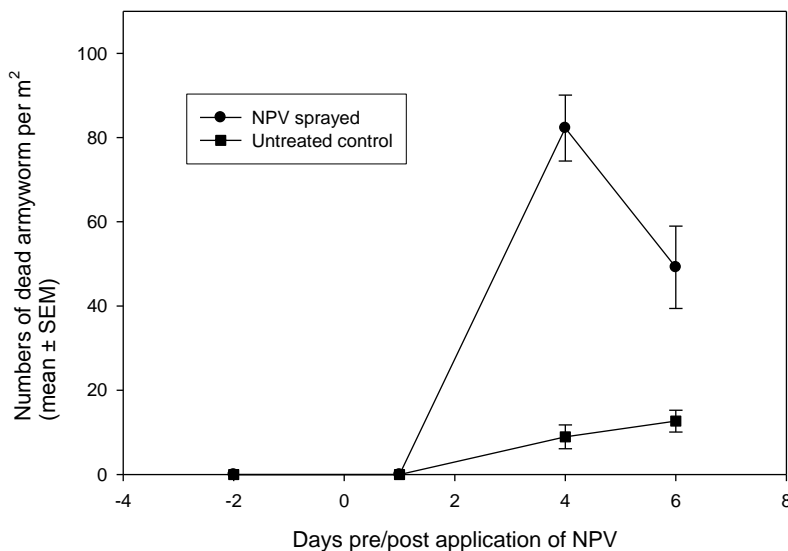
**Plate 1 Dead and dying larvae in NPV sprayed plot hanging from grass**



The numbers of dead larvae seen in the NPV plot were high accounting for about half the numbers of larvae that disappeared. The failure to find an exact correspondence between larval reduction and the number of corpses seen is not surprising as many of these larvae were still very small II-III. These small corpses dry out quickly and were perhaps being blown away, lost in the canopy, or eaten by invertebrate scavengers such as ants which were abundant in these plots. there can

be little doubt that an NPV epidemic was the most likely cause of the armyworm population collapse seen in the sprayed plot.

**Fig 10 Numbers of dead larvae per metre<sup>2</sup> in NPV and control plots in aerial trials of NPV at M'ringa 2004**



The speed of kill by SpexNPV infection seen here is very encouraging. It may partly be due to spraying the larvae as young larvae when infection is very quick to establish and kill young larvae. Glasshouse trials in 1999 showed that larvae infected as III instar suffered 60% deaths by day 4 (Grzywacz et al 2000). However the rapid kill could also be because the African armyworms, unlike many other noctuid larvae, have the habit of feeding actively by day on grass exposed to sunlight. This could result in very high larval body temperature and metabolism which would make the NPV infection cycle much faster. A similar phenomenon has been observed in the NPV infections of temperate species (J S C Cory pers comms).

This observation provides an interesting contrast with the field performance of the fungal biological pesticide "Green Muscle" on brown locusts and some grasshopper. Infected grasshoppers actively "sunbathe" to raise their body temperature and slow infection. Here the sunbathing of infected locusts produces a "fever response" that impedes the development of the disease delaying death and the markedly reducing the effectiveness of the control (Thomas and Blandford 2003).

The effectiveness of SpexNPV in killing armyworm and preventing damage has been clearly demonstrated provided it can be applied early enough during the early larval stages. This however depends upon accurate forecasting and the ability to intervene with application. Discussions with PCS and Mr P Nayland of Airspray Ltd indicate that both forecasters and sprayers are confident this could be achieved if the current forecasting system was scaled up and the SpexNPV was available. Airspray Tanzania Ltd confirmed that many aerial spraying operations already undertaken in Tanzania already requires quick response times similar to those that would be required in NPV armyworm control operations.

The effectiveness of aerially applied NPV is a key finding of the project as aerial application is a major tool in the government's programme of armyworm control and would be the only effective way to implement strategic control in the isolated primary outbreak areas that are far from roads.

In conclusion the field trials have demonstrated that SpexNPV could be used as a substitute for insecticides in armyworm control though this work would need validating on crops as well as the pastures tested here. Neem does show promise but should be evaluated in trial programmes to ensure reliability before promotion nationwide.

### **Output 5 An Ecological study of the role of SpexNPV in armyworm population dynamics and the potential for strategic control using NPV completed.**

This study was completed and has thrown important novel insights into armyworm/virus ecology. The key finding of the ecology study has been to establish that SpexNPV can be present in host insects as a non lethal asymptomatic persistent infection as well as the previously described overt lethal form. This finding has important implications for understanding the ecology of this insect and its virus.

Before this project had been suspected, though never proven, that armyworm NPV could be present in some "latent" form that enabled it to survive during the periods between armyworm outbreaks when host numbers are very low and populations scattered. In particular, it is important to understand how NPV outbreaks could occur in armyworm populations that outbreak in areas where no armyworm had been seen for many years and the existence of an environmental reservoir of NPV was unlikely. Prior to this project it had been accepted wisdom that NPV persists in the absence of hosts in soil, foliage or some other environmental reservoir but this hypothesis seemed poorly to explain the sudden occurrence of NPV in outbreaks of armyworm that occurred in areas where there had been no outbreaks for many years. This project has produced the important ecological finding that in a highly migratory sporadic pest insect an alternative mechanism of pathogen persistence within host populations exists.

The initial ecology component addressed two broad issues; the mechanisms of virus persistence, particularly between generations, and presence and impact of virus diversity in field populations.

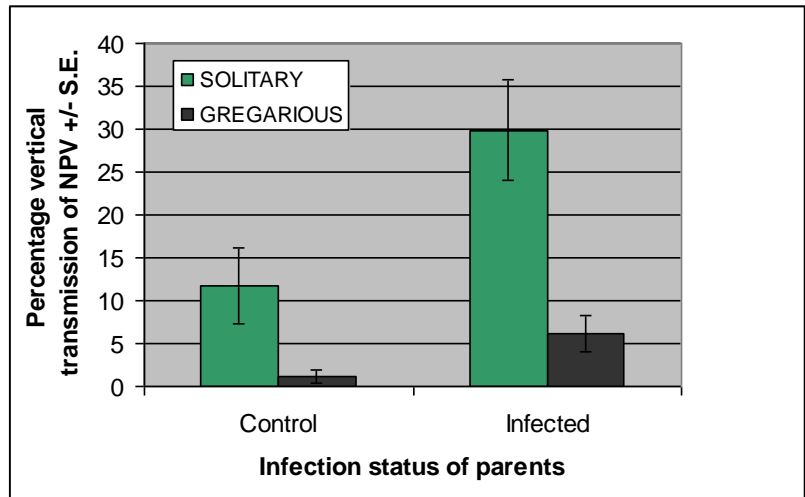
The ecology work has established conclusively that a high proportion of armyworm moths in natural populations carry asymptomatic, persistent, covert infections of NPV, which can be transmitted from adult moths to their offspring via the egg (i.e. transovarial vertical transmission). This was confirmed through successful use of specific RT-PCR technique used to identify low level viral replication in non symptomatic insects that had been reared for several generations after collection from the field without symptoms of disease. The phenomenon of asymptomatic viral infections in insects has been conclusively identified only recently as molecular techniques have been developed and this is the first confirmed finding of an asymptomatic infection in any tropical insect.

This provides a mechanism by which the virus can move between armyworm outbreaks and persist during the dry season and in years when population densities are very low. Virus can be transmitted vertically from adult moths to larvae in an overt (i.e. resulting in disease), in addition to the commoner asymptomatic, covert, form. The work has shown that the incidence of vertically transmitted, overt infection can be increased by infecting larvae with a sub-lethal dose of NPV. Thus, any caterpillars surviving NPV exposure have the potential to pass on a lethal dose of NPV to their offspring.

Armyworm caterpillars reared at low density, the distinctive green solitaria form, are much more susceptible to NPV acquired via horizontal transmission (i.e. by ingesting OBs), than the high

density reared black gregaria form. They are also much more likely to transmit virus as an overt infection to their offspring (see Figure 11 below). This suggests that control in solitaria using SpexNPV, if adopted as part of a strategic control programme, would be even more effective than the field trials carried out to date against gregarious swarming larvae (see output 4).

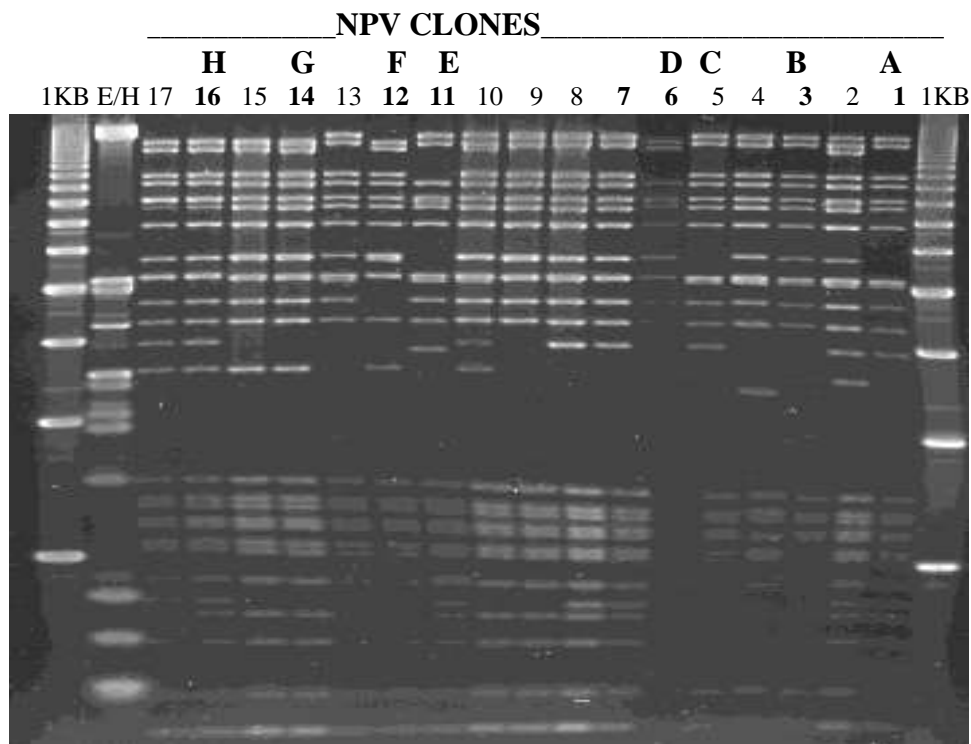
**Figure 11 transmission of NPV in solitary and gregarious phase African armyworm**



A key question looked at was how are these asymptomatic infections in insects triggered into outbreaks of the lethal overt form. Knowing this might enable forecasters to predict when natural NPV outbreaks are likely to be prevalent so reducing the need for control operations. It had been suggested from results in a previous phase that the apparent success low applications of SpexNPV itself might trigger be due to triggering of latent NPV. However despite many attempts to trigger overt NPV from asymptomatic larvae using low doses of SpexNPV and various environmental stressors crowding, heat, high humidity or cooling no trigger for the latent SpexNPV (or any other NPV) was identified in laboratory studies. This question is still of great ecological interest is the subject of continuing research at CEH.

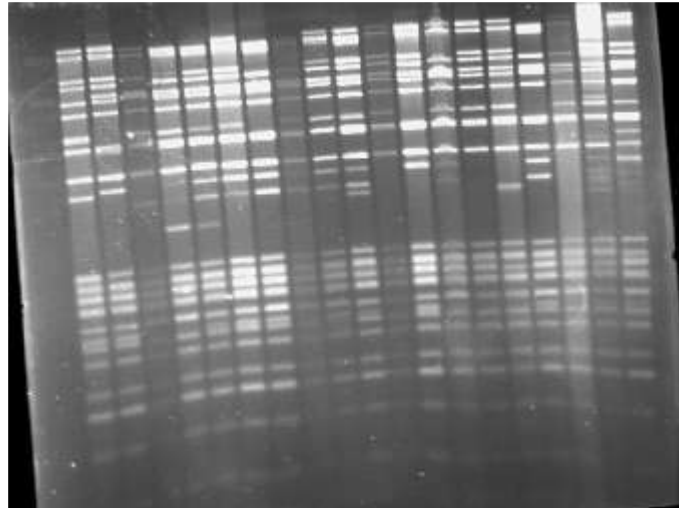
It was evident from the initial analysis of the *SpexNPV* that there was considerable genetic diversity within the virus populations. A study on the virus stock used in trials, originally isolated in 1977, separated out the virus variants using dilution cloning *in vivo*. After 4 rounds of *in vivo* cloning 17 genetically distinct variants were isolated and identified (Fig 12.).

**Figure 12. Diversity of *S. exempta* NPV variants within the original *S. exempta* NPV isolate NRI #0045.**



Diseased caterpillars were collected during natural armyworm outbreaks also showed considerable genetic variation in insects collected from plots of only 10m<sup>2</sup> (Fig 13). Why this high degree of genetic diversity exists and is maintained is as yet unclear but it suggests that SpexNPV virus host system is ecologically a very interesting one that might give interesting insights into the evolution and genetic diversity of virus dynamics in unstable host populations.

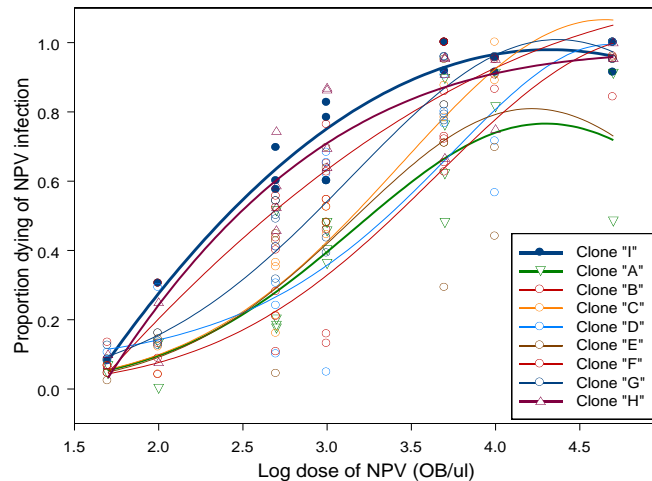
**Figure 13. DNA profiles (EcoRV) of individual *S. exempta* NPV isolates collected Lucky farm , Arusha, Northern Tanzania 2002. Each track represents a single infected caterpillar.**



The work has also shown that there is considerable variation between these clones in their ability to kill armyworms (Fig 14). A bioassay compared 17 *S. exempta* NPV clones against the original mixed virus stock in terms of pathogenicity and speed of kill. These preliminary baseline data, together with REN data, were then used to select the most genotypically and phenotypically diverse clones for further study. Extensive bioassays were carried out on the 8 chosen virus clones (labelled A-H, Fig. 1). Figure 15 shows the dose response curve of the clones against that of the original mixed stock (I) which clearly demonstrates that the wild type NPV has higher pathogenicity than any of the individual NPV clones.

The conclusion is that at present the strategy of using wild type mixed NPV, as used in the field trials, is more effective than the use of cloned NPV lines.

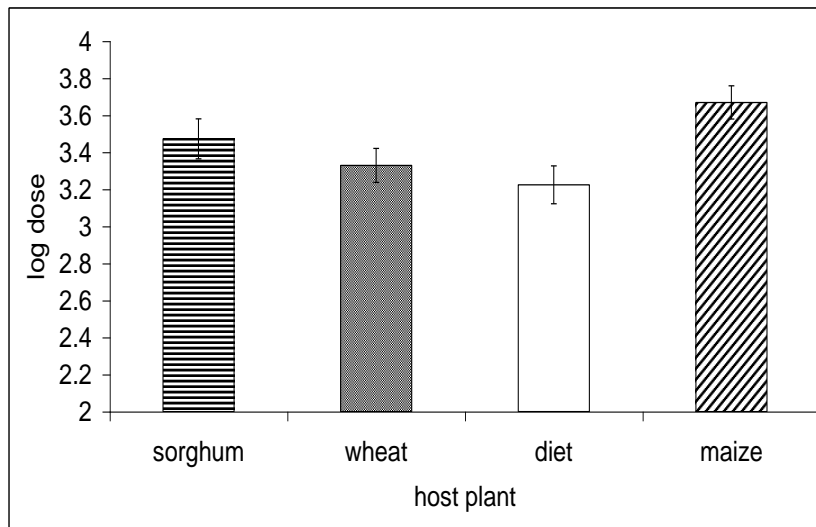
**Figure 14. Dose response of eight *S.exempta* NPV clones compared with the parent wild type virus.**



The project collected over 400 isolates (from single caterpillars) of SpexNPV from Tanzania, and a preliminary screening has indicated that these comprise at least 60 genetical different variants. Investigations of these isolates are continuing at CEH Oxford and University of Lancaster. This collection will form a useful genetic database for future SpexNPV studies and a resource of biodiversity for practical control with SpexNPV. This high level of genetic and phenotypic diversity has implications for the selection of effective control agents for armyworm, and in particular, the consequences of different NPV production methods that might alter the composition of virus material for application in the field.

In addition to studies on genetic and biological diversity of the NPV a short study was carried out to assess whether host plant could influence the infectivity of the virus. Virus was administered to the larvae on maize, sorghum and wheat, in comparison with artificial diet. Larvae fed on maize proved slightly more resistant more resistant to virus than larvae fed on the artificial diet but no major differences in SpexNPV effectiveness on different crops were found. Thus it may be inferred that a single application rate of SpexNPV could be used on all the main target crops.

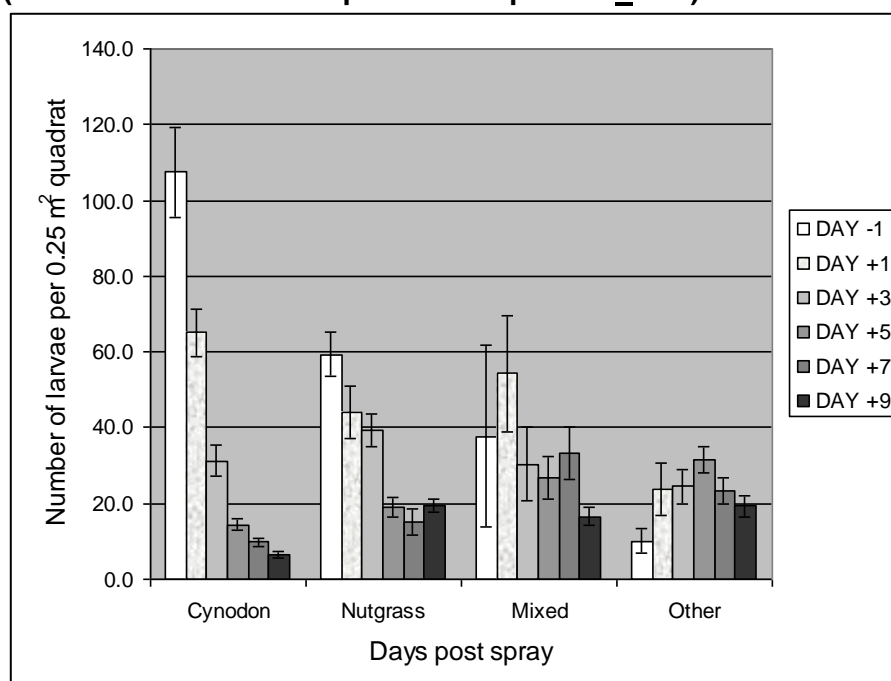
**Figure 15. LD<sub>50</sub> of *Spodoptera exempta* larva inoculated with *S. exempta* NPV on different host plants**



Studies in the field in the last year (2004) on natural armyworm outbreaks showed clearly that armyworm have a strong preference for *Cynodon* grasses over any other plants (see appendix 4) , especially as young caterpillars when they are green and difficult to find. In field trial areas approximately 40% of the quadrats sampled were dominated by star grass (*Cynodon nlemfuensis*); a further 25% were a mixture of *Cynodon* and other species; c. 10% were dominated by nutgrass (*Cyperus* sp.), and the remaining 22% were dominated by a variety of other species, including *Chloris* spp., *Eragrostis* spp. and *Seteria* sp.

Prior to spraying armyworm larval densities were approximately 10 times greater in quadrats dominated by *Cynodon* grasses (mean = 428 larvae per m<sup>2</sup>) than in those dominated by species other than *Cynodon* and nutgrass (40 larvae per m<sup>2</sup>) (c.f. white bars in Figure 16). Thus, even though *Cynodon* dominated just 40% of the quadrats sampled, c. 70% of all larvae counted were in quadrats dominated by *Cynodon*. This suggests that the armyworm larvae were preferentially feeding on *Cynodon*. Given the young age of the larvae at this time (around 5 days old), it is likely that this bias in plant usage is because female moths preferentially lay their eggs on *Cynodon* grasses.

**Figure 16: Density of armyworm larvae in relation to the dominant host plant in the quadrat (mean number of larvae per 0.25m<sup>2</sup> quadrat  $\pm$  s.e.).**



This initial bias in host-plant usage quickly eroded over time, with larval densities appearing to be almost independent of the identity of the dominant host plant species by day +3 post-treatment. This is likely to be a consequence of the larvae having to switch to new host plants when *Cynodon*, and other preferred species, are exhausted.

Fieldwork undertaken in 2002-2003 showed that armyworm caterpillars feeding on *Cynodon* grasses in high densities stimulate them to produce high levels of cyanide (see appendix 19). This finding may explain the previously unexplained reports of cattle deaths in herds feeding on pasture after heavy armyworm attack. Further investigations on the role of armyworm have now been taken up by the Tanzanian Ministry of Agriculture for further follow up to determine the scale of this problem and to validate the trial findings.

The results from the ecology studies have implications for *strategic biocontrol* using NPV:

- 1) Biocontrol will be most effective if we use a mixed isolate of NPV, rather than any of the individual NPV clone (this will also minimise the possible build-up of genetic resistance to sprayed NPV in the insect population)
- 2) Caterpillars surviving NPV biocontrol, but carrying sub-lethal covert infections, are likely to transmit viral infection vertically to their offspring. These larvae have the potential to trigger virus epidemics at any new armyworm outbreaks. Thus, strategic control using NPV can produce a double action: (1) effective biocontrol of larvae at focal outbreak sites and (2) the potential for continued biocontrol at subsequent outbreak sites elsewhere, initiated by migrating insects surviving the initial biocontrol effort.
- 3) SpexNPV could be used at lower rates if it is targeted at the more susceptible, low-density phenotypes (e.g. caterpillars persisting at low densities in off-season breeding grounds near the coasts of Tanzania).



4) As armyworms tend to congregate on *Cynodon* grasses, control using NPV should be targeted at pastures dominated by these grasses, and this may also help to prevent cattle deaths caused by armyworms feeding upon cyanogenic grasses.

**Output 6 A strategy for production and provision of neem and NPV to resource poor farmers in outbreak areas of Tanzania will be completed.**

Understanding farmers' constraints and decision making regarding armyworm outbreaks and control measures is important for developing a strategy for armyworm control. Novel control measures must compare favourably with current options if they are to be adopted. Resource poor farmers are more vulnerable to armyworm outbreaks, so it is important to take resource endowment into consideration when devising improved control. A major output of the project has been the socio-economic work covered in detail in the report in appendix ? This work provided important findings for many of the outputs of the project and stands as an important dataset on farmer attitudes and perceptions of armyworm control and forecasting.

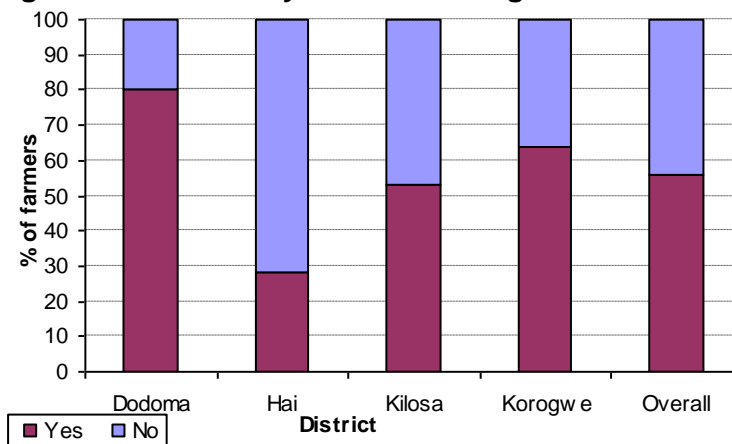
Of farmers interviewed 97% had had armyworm outbreaks on their farms in the previous 7 years while 65% had had an outbreak in the previous year. Despite the high frequency of outbreaks less than 35% of farmers controlled outbreaks. This figure masks significant regional differences with 90% of farmers in wealthier districts using control while in the poorest only 15% report controlling outbreaks. Of those farmers who did not treat outbreaks 94% said that financial constraints were the main factor. Thus armyworm has a disproportionate effect on the poorest farmers and this needs to be considered when developing effective control strategies suitable for this group.

The work in output 4 has indicated that neem from Tanzania in both leaf extract and seed has potential as an armyworm control tool. In developing a strategy for its use however the key issue of supply needs to be considered.

Neem seed appears the more effective formulation for control but in Tanzania seeding occurs after the main armyworm season is past. Thus a strategy to promote neem seed for smallholder armyworm control would require prior investment by farmers in collecting and saving stocks of seed. Interviews with farmers indicate that most do not currently build armyworm control into their cropping plans or budgeting. Thus a strategy to promote neem seed seems unlikely to be taken up by most farmers except in the locations where armyworm attacks are an annual event.

Neem seed might be used as part of government run national armyworm control but here the problem is that the application requires 2.5 Kg per hectare. The problem here is that this would require a major financial and organisation effort to collect, store and distribute the quantities needed. There is currently no support for this option within the Tanzanian system which struggles to distribute existing chemical insecticides whose high activity, low bulk and long term stability make them much easier to implement as a control.

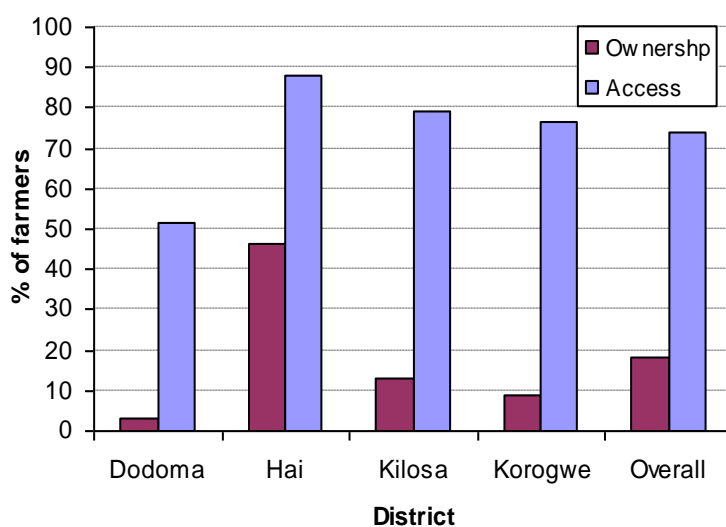
When considering neem leaf as an option 50-80% of farmers have neem trees on their farms. Neem leaf used in the trials here was at 25 kg per hectare so that its use in other than a very local context would be very difficult logistically. In Dodoma 80% had trees but only 35% had used these for pest control and that was mostly for storage pests. Only 10% had used neem for armyworm control.

**Fig 17 Percentage of farmers in key districts having access to neem trees**

Thus it may be concluded that there is scope for the development of neem as a control strategy for poor farmers who have access to neem but knowledge appears to be a serious constraint. The distribution of neem trees suggests considerable opportunities for promoting home preparation of neem for armyworm control in districts such as Dodoma where capacity for purchasing pesticides is very limited.

This approach is currently being promoted by PCS as part of its community forecasting and control projects currently funded by CPP/USAID. This effort needs to be monitored and its success in trial districts reviewed before promotion on a national scale is undertaken.

Access to sprayers is crucial for armyworm control with insecticides/NPV or neem. About 75% of farmers said they had access to a sprayer, either their own or through borrowing/hiring. Hai had the highest percentage of farmers owning or having access to sprayers (88%) because farmers in Hai already use sprayers on their coffee crop. However in the poorer Dodoma only 50% had access but only 3% owned sprayers.

**Fig 18 Access to sprayers by farmers in different districts**

However, access to a sprayer in principle does not mean that it would be available when needed. In the group discussions accessibility of sprayers to farmers was identified as a serious constraint. This lack of access to sprayers by poorer farmers has implications for control methods being

developed. Either farmers have to be given improved access to sprayers, or the pesticides have to be formulated in such a way that they will not require sprayers for application e.g. as bait formulations

The project did some initial work on formulating NPV as baits in the laboratory but found that they dried out quickly and became unattractive to larvae. No long term stable baiting technology exists for NPV. It is possible that using an aqueous gel based formulation combined with barrier technology could give a bait stable at ambient temperatures for 1-2 years. This would however require a significant new research effort and perhaps should not be pursued further until the scope for conventional application is fully exploited.

The work of the socio-economic survey identified that cost was the major constraint to armyworm control for most poor farmers in Tanzania. This poses a serious challenge for any strategy promoting NPV in place of conventional pesticides.

Most NPV pesticides are produced in dedicated facilities where large numbers of susceptible host insects are mass reared, infected, incubated then harvested before finally being processed to extract the multiplied NPV. Such facilities require significant capital investment. The products, especially if produced using a low tech approach in a developing country, are often comparable in cost to insecticides (Warburton and Grzywacz 2001,). Work in the previous project demonstrated that SpexNPV can be produced in dedicated NPV production plants through in vivo production in cultured *S.exempta* (Cherry et al 1997), but the cost is likely to be at least comparable to or higher than existing chemical insecticides, that Tanzania already has difficulty affording. In order to be a really viable option SpexNPV would need to be produced at a cost equal to or preferably lower than the current chemical insecticides

In 1992 under DFID and EU funding a pilot SpexNPV facility was set up in Kenya at the IIBC station in Maguga to be run by DLCO with technical backstopping from NRI (see appendix7). A proposal to establish this on an operating basis for three years at a cost of £549,000 was prepared for donors but not subsequently funded. In the absence of donor support the facility carried out small scale SpexNPV production over the following year but lacking support the initiative seems to have fizzled out by early 1994. Although demonstrating the technical feasibility of producing SpexNPV in Africa it pointed out the organisational and economic problems of setting up dedicated facilities for producing specific biocontrol agents for sporadic migratory pests.

There is a potentially cheaper method for NPV production which is to use artificial infection of natural outbreaks of the pests and harvesting dying infected insects as a source of NPV. This method was originally developed for producing NPV in several species of forest Lepidoptera such as *Euproctis* and *Malcosoma* where a habit of forming nests where broods of hundreds of synchronous foraging caterpillars retreated to tents made it easy to collect infected insects in large numbers. It has not been widely used for other pests because they rarely appear as synchronised outbreaks in the densities that make field collection feasible.

Given the propensity for armyworm to produce outbreaks with very high densities of larvae (>500 per m<sup>2</sup>) this species would seem very promising as a candidate for field production. One such field production system has been developed by the EMBRAPA the soya bean research institute in Brazil for producing *Anticarsia gemmatilis* NPV (AgNPV) at a cost of 1.26US\$ per ha (Moscardi 1999). This is used to produce some 40 tons of infected insects annually that are processed into a biological insecticide used now on some 2 million hectares of soy crop each year

Adopting such a field production system seems the most promising option for Tanzania. Dr Moscardi of EMBRAPA, head of the NPV programme has with support from USAID become involved in the armyworm project to advise on field production. He made a visit to Tanzania in the 2003 armyworm season, which unfortunately experienced few outbreaks. He subsequently wrote

a report (appendix 8) in which he concluded that field production of SpexNPV would be feasible if the EMBRAPA developed protocols for processing were adopted. In the autumn of 2003 Mr Mushobozi of Pest control services Tanzania visited EMBRAPA for a month to be trained in production and processing methods. This visit confirmed Mr Mushobozi in the opinion that these methods seem eminently appropriate for use in Tanzania (appendix 17).

The main challenge to adoption of this system is to assess if field collection of SpexNPV is practicable. The AgNPV is easy to produce partly because when the infected insects die they remain intact. Thus AgNPV can be harvested by collecting dead insects relatively easily and at low cost simply by hand picking the corpses. SpexNPV in contrast is characterised by highly fragile cadavers that split easily releasing the progeny NPV. The difference is due to cathepsin genes present in SpexNPV, but not AgNPV, that are expressed late in the infection cycle and whose products digest the insect skin. Thus the direct harvesting of infected armyworm insects by hand may not be possible without excessive loss of NPV.

Trials to produce NPV from the field started in 2001 season with harvesting work from a natural outbreak of NPV. Full details of this are in the field trial report for 2001 see appendix 2. This showed that the yield from armyworms collected in the field was  $4.62 \times 10^9$  per insect. This was twice that obtained in laboratory reared cultures by artificial infection (Cherry et al 1997). Several collection methods were tried though only cutting grass on which infected larvae were attached, as they commonly are when NPV killed. From an outbreak of 35 larvae per metre it was determined that with simple manual collection it would take one person 2.5 hours to collect enough SpexNPV to treat 2.5 hectares if treated at  $1 \times 10^{12}$  per hectare. Material produced from the field in 2001 was tested in the field in 2002 and found to be as effective as SpexNPV produced by in vivo laboratory culture (see appendix 3 for details). This work was repeated on a larger scale in 2004 which produced similar results (see appendix 4).

The harvesting work in 2001 and 2004 shows that feasible quantities of SpexNPV can be collected from artificially infected armyworm outbreaks. However the system of simple manual collection as used in the Brazilian EMBRAPA system, the model we were hoping to use is not practical for SpexNPV as the cadavers of the current SpexNPV isolate infected larvae are too fragile for direct collection.

Field collection of SpexNPV from artificially infected larvae by collecting grass is a feasible method to produce SpexNPV. The simple manual harvesting systems tested in this trial produced an NPV yield rate of  $4 \times 10^{11}$  OB per man-hour. Given the current SpexNPV application rate is  $1 \times 10^{12}$  OB per ha it would take 2.5 man-hours to harvest enough SpexNPV to treat 1 hectare. Given a six hour working day at a wage of 3 dollars per day this suggests that SpexNPV could be produced by this method not counting processing cost of a maximum of 1.5US\$ per ha.

Clearly the manual collection of armyworm NPV using a simple manual system of using scissors and plastic bags is not likely to be the most cost effective for large scale production. Scaling up to better bulk collection of grass and larvae by say using sickle or scythe would probably increase the rate of collection considerably though to produce the amount required for a national control strategy would probably require mechanisation. The next stage would be to test a mechanical harvesting system incorporating a combined grass cutter and collector.

An alternative approach to this problem of harvesting would be to find and use a cathepsin minus strain of SpexNPV that does not cause the insect body to break up. It is known that a number of different SpexNPV isolates exist and 60 of these have been catalogued as part of this project. All strains so far identified are however cathepsin positive and in all strains so far examined infection is accompanied by cuticular thinning and fragility (J S C Cory pers comms). Thus while an easier to produce cathepsin minus strain of SpexNPV and this may yet be found for the immediate future it would be advisable to work with the current SpexNPV strains.

The current cost of chemical control is an important benchmark against which any new technology needs to be assessed. It was found that currently control costs 8453 TSH (US\$ 7.6) a litre and is used at 2 litres per ha. Any new technology needs to be at least equal in price to (US\$15 a ha) but preferably much cheaper if access to armyworm control technology is to be significantly increased and the poorer farmers to benefit. The distribution of neem trees suggests considerable opportunities for promoting home preparation of neem for armyworm control in districts such as Dodoma where capacity for purchasing pesticides is very limited.

In any strategy to use novel controls then support by the regulatory bodies for initiatives will be crucial. The project team have kept the appropriate regulatory committees from the Tanzanian tropical Pesticides Research institute and key players in biopesticides registration in the picture through meetings and presentations. The secretary of the biopesticides committee visited the 2004 trials and was very impressed with the results. The director of Tropical Pesticides Research Institute has also been briefed and has said would support registration of SpexNPV as biological insecticide.

Thus the enabling environment for adopting SpexNPV in Tanzania is highly favourable in contrast to some other African countries where adoption of baculoviruses and other biopesticides has met many perceptual and regulatory barriers. This may in part be due to the earlier efforts of the research team to raise public awareness and keep the authorities and key policy players well informed right from the start of the project. However it is also because in Tanzania NPV disease was recognised as a naturally occurring phenomenon by armyworm researchers and policy makers before the project started. It is thus it is not perceived as a new and dangerous introduction. This is something that has occurred elsewhere in Africa when attempts to promote a baculovirus, although endemic and widespread, was perceived as a new unknown quantity because it had not previously been reported or recognised.

#### **Output 7 An assessment of the viability of procurement and use of NPV and neem for strategic armyworm control will be completed.**

Strategic armyworm control has been an option under consideration for many years. Strategic control for armyworm focuses on treating primary outbreaks in Tanzania to suppress later progeny outbreaks in croplands in Tanzania and neighbouring countries has a long history although often it is suggested means was the use of chemical insecticides. A comprehensive review of the economic importance of armyworm outbreaks (Scott 1991) and a paper modelling strategic control (Cheke & Tucker 1995) both concluded that the concept was viable and cost effective if employed early in the season.

The socioeconomic survey in this project concluded that the major obstacle to implementing has been cost but also the need for regionally co-coordinated approach to its implementation and funding has been a problem. An additional element in recent years has been move to restrict the use of chemical pesticides in Africa. This has impinged on the willingness of donors to support projects such as a strategic armyworm which would inevitably involve the application of control agents to large areas including national parks.

The work of this project has identified SpexNPV as a candidate natural control that could be applied over large areas without significant environmental problems. The recent OECD report reviewing baculoviruses as pesticides (2002) identified baculoviruses (which includes NPV) as having no harmful environmental effect.

In addition demonstrating that SpexNPV could be produced in the field at a cost very much below existing chemical insecticides it can be seen that adopting SpexNPV as the prime agent in a strategic control strategy would solve some of the economic problems inherent in implementation.

## Output 8 Dissemination of results through papers, publications and mass media

The project has run a successful programme of disseminate its findings and information about its activities particularly since 2002. This has achieved a high public profile for the project in Tanzania and internationally.

Dissemination of project outputs at a national and regional level used a strategy of leaflets, posters, press releases, personal contact with journalists and oral presentations to key policy makers. Talks to various national and regional scientific and agricultural fora were given by W Mushobozi and a PowerPoint talk on the project is attached as appendix 23. A talk on the future of biopesticides including the results of this project will be given by W Mushobozi to a regional conference at Arusha in September 2004.

The most effective mass media channels were the national TV broadcasts (and radio reruns) which attracted wide attention both by the general public and key policy makers in Tanzania. Opinion forming newspapers were also targeted and a copy of the article in the Financial Times for East Africa is attached as appendix 13.

Efforts to disseminate to the international development and scientific community have borne considerable fruit. A number of articles about the work of the project have been produced by project staff or independent journalists who contacted project staff after reading press releases circulated by the project. Examples of are the articles in New Scientist, International Pest control and Biocontrol news and information, all attached as appendices. An oral presentation is to be given to the Society of Invertebrate Pathology, the premier scientific forum for microbial control work at their 2004 meeting in Helsinki. The PowerPoint of this is attached as appendix 26. No peer reviewed papers on the project work have yet been accepted but two papers on the virus ecology entitled (1) "High levels of sub lethal NPV infection in populations of armyworm in Africa" and (2) "Vertical transmission of *Spodoptera exempta* NPV in solitary and gregarious phases of the armyworm *Spodoptera exempta*" are drafted for submission to a high impact scientific journal. The field trial work on NPV and neem will be written up for submission to an international crop protection journal by end of 2004.

A list of outputs of the dissemination of results is given below and these are attached as appendices

### Publications about the project

Anon (2004). Focus on Agriculture: Biological weapons used in battle against armyworm. *International Pest Control*, 46, (2), p96.

Bell B (2004). Natural attack on African Crop pests. *British Industrial News*, January 2004. p7

Daylell T (2004). Eco friendly Pesticides. *New Scientist* p47 24 February 2004

Dixon David (2004). Biological Weapons used against armyworm. *Appropriate Technology* 31, (1) p60-61

Ihucha A (2004) Natural Way to control armyworms sought. Financial times, March 10-16 p10 [Tanzania Kenya]

Grzywacz, D., and Mushobozi, W., (2004) A local solution to African Armyworm" *Biocontrol News and Information* March 25, (1), 7-8N.

Randerson, J., (2003) Corpses of Dead kill living. *New Scientist*, p12. 13 December 2003.

#### Other dissemination

Mushobozi W (2004) Natural tools to control armyworm in Tanzania. News at 2000 ITV , 17 April 2004, 10 minutes, 2000 hours. Tanzania [National TV interview] [in Swaheli]

Mushobozi W., (2004) British and Tanzanian scientists discover a natural way to control armyworm. ITV at 2000 hrs 19<sup>th</sup> February. [National TV interview] [in Swaheli]

Mushobozi W and Grzywacz D (2003) "NPV a new biological control for armyworm in Tanzania" 20 copies. NRI, Chatham, [Poster] [Field & Policy] [English]

Mushobozi W and Grzywacz D (2003) "A new natural control for armyworm in Africa" 50 copies NRI, Chatham [leaflet] [Field] [English] p2

Mushobozi W and Grzywacz D 2003 "A new natural control for an old pest in Africa" 50 copies NRI, Chatham [leaflet] [Policy] [English] p 4

Mushobozi W (2003) New methods for control of armyworm in Tanzania. Ministry of Agriculture and Food Security Armyworm Control & Forecasting Services Tanzania [Oral presentation to Tanzanian National Advisory Plant Protection Committee April 2004]

Redman L (2003) Armyworm outbreaks in Tanzania. Postgraduate Research Seminar. Centre for Ecology and Hydrology, Oxford, 4 September 2003 [Oral presentation]

Wilson and Mushobozi (2002) "NPV disease in African armyworm", 100 copies, PCS and Stirling University. [Poster] [Field & Policy] [English]\*

Wilson K. (2004) African Armyworms and their Natural Enemies IACR Research Seminar series. IACR, Rothamsted, 24th June 2004: [Oral presentation]

Output 9 A strategy to promote and implement novel controls in Tanzania to poor farmers using existing extension services will be developed.

The use of SpexNPV as an armyworm pesticide has been recommended for adoption as national policy by the Tanzanian National Advisory Plant Protection Committee in June 2004. They have directed the Ministry to develop a strategy for the adoption of NPV in place of existing chemical controls (appendix 18). After this is adopted PCS will promote use of SpexNPV through its own channels and the findings of the project will inform the USAID funded armyworm control project. The use of neem is currently being promoted to farmers in several trial districts by PCS, as part of the linked community forecasting and control project funded by USAID.

Output 10 The capacity of agricultural research organizations will be enhanced through training personnel in the production and use of NPV and neem control technologies.

Training of staff in PCS Tanzania in the production and use of SpexNPV and neem was carried out through training visits totalling 3 months to UK (NRI) and 1 month in Brazil (EMBRAPA). The later was co-funded by USAID. Training was also carried out of staff in Tanzania by project staff as part of the field trials programme throughout the project.

### **Contribution of Outputs to developmental impact**

*Include how the outputs will contribute towards DFID's developmental goals. The identified promotion pathways to target institutions and beneficiaries. What follow up action/research is necessary to promote the findings of the work to achieve their development benefit? This should include a list of publications, plans for further dissemination, as appropriate. For projects aimed at developing a device, material or process specify:*

- a. *What further market studies need to be done?*
- b. *How the outputs will be made available to intended users?*
- c. *What further stages will be needed to develop, test and establish manufacture of a product?*
- d. *How and by whom, will the further stages be carried out and paid for?*

The goal of improving access of poor people in Tanzania and other East African countries to affordable armyworm control has been significantly progressed by the project which has demonstrated that there are safer more affordable alternative to chemical pesticides. Through the project having worked directly with the ministry of agriculture in Tanzania outputs have been made available to policy makers and armyworm controlling authorities.

The Tanzanian Ministry of Agriculture has charged PCS to develop a strategy for implementation of SpexNPV as an armyworm control (appendix 18).

Further stages are:

1. The use of SpexNPV on pasture and rangeland has been evaluated but validation trials on key agricultural crops Maize, wheat and barley need to be carried out before it is adopted for use on these.
2. There is need for further work to develop the low cost field production of SpexNPV into a working system but on the basis of work carried out to date this faces no serious technical barriers.
3. The use of neem by farmers for armyworm is being promoted as part of the community forecasting and control project funded jointly by CPP/USAID/GoT. This will evaluate this at an operational level in the forthcoming armyworm season early in 2005.

The project has significantly progressed the DFID CPP programme goal of reducing dependence on synthetic pesticides and by developing cheaper locally producible alternatives has identified a more sustainable alternative.

The strategy now must be to secure adoption by Tanzania and then once this system is fully working it can be demonstrated to other users both national (Kenya, Ethiopia and South Africa) as well as regional and international migratory pest control agencies DLCO and FAO.

Further planned publications are discussed in detail under Output 8.



## Appendices

Appendix 1 Knight J.D. (2001) Planning workshop on the development and implementation of improved forecasting and novel control methods for armyworm in Tanzania. Report of the workshop held at the Southern Africa Management Institute, Arusha, Tanzania, 9<sup>th</sup> – 11<sup>th</sup> October 2001.

Appendix 2 Parnell M., Grzywacz D and Mushobozi W (2001) Field trials of *Spodoptera exempta* nucleopolyhedrovirus biological pesticide against armyworm on rangeland in Tanzania 2000-2001 NRI Report no. 2705 pp17

Appendix 3 Mushobozi W Parnell M and Grzywacz D (2002) Field trials of *Spodoptera exempta* nucleopolyhedrovirus and neem carried out against armyworm on rangeland in Tanzania 2001-2002. NRI report 2720 pp28.

Appendix 4 Grzywacz D., Mushobozi W., and Wilson K., (2004) Report of field trials of novel technologies, *Spodoptera exempta* nucleopolyhedrovirus and neem extracts against African armyworm in Tanzania 2004, NRI Report no 2777. pp 28.

Appendix 5 Cory J S C (2002) Progress report: Novel control methods for armyworm. NERC Centre for Ecology and Hydrology, Oxford. Pp2

Appendix 6 Njuki J., Mushobozi W., and Day R., (2004) Improving armyworm forecasting and control in Tanzania: a socio-economic survey. CABI ARC Nairobi. Pp

Appendix 7 Jones K.J. (1993) FEASIBILITY STUDY OF THE PRODUCTION AND FORMULATION OF SPODOPTERA EXEMPTA NUCLEAR POLYHEDROSIS VIRUS: PROJECT REVIEW. NRI report. 35pp.

Appendix 8 Moscardi F (2003) : Development of Area Wide Strategic Armyworm Control Using *Spodoptera exempta* Nucleopolyhedrovirus (SpemNPV) Consultants Report pp 10.

Appendix 9 Anon (2004). Focus on Agriculture: Biological weapons used in battle against armyworm. *International Pest Control*, 46, (2), p96.

Appendix 10 Bell B (2004). Natural attack on African Crop pests. *British Industrial News*, January 2004. p7

Appendix 11 Daylell T (2004). Eco friendly Pesticides. *New Scientist* p47 24 February 2004

Appendix 12 Dixon David (2004). Biological Weapons used against armyworm. *Appropriate Technology* 31, (1) p60-61

Appendix 13 Ihucha A (2004) Natural Way to control armyworms sought. *Financial times*, March 10-16 p10 [Tanzania Kenya]

Appendix 14 Grzywacz, D., and Mushobozi, W., (2004) A local solution to African Armyworm” *Biocontrol News and Information* March 25, (1), 7-8N.

Appendix 15 Randerson, J., (2003) Corpses of Dead kill living. *New Scientist*, p12. 13 December 2003.

Appendix 16 Redman L (2003) Armyworm outbreaks in Tanzania. Postgraduate Research Seminar. Centre for ecology and Hydrology 4 September May 2003 [Oral presentation]

Appendix 17 Mushobozi W (2004) Study Visit report on NPV production methods to EMBRAPA Brazil. Tanzanian Ministry of Agriculture and Food security report pp 7.

Appendix 18 Kategira F (2004) Letter from Tanzanian Ministry of Agriculture confirming that Tanzania is directing its Pest Control services to adopt SpexNPV as national strategy for armyworm control.

Appendix 19 Wilson K (2002) Interactions between African armyworm (*Spodoptera exempta*) and its food plant (*Cynodon* spp.) A preliminary report. Pp11

Appendix 20 Mushobozi W and Grzywacz D (2003) "NPV a new biological control for armyworm in Tanzania" 20 copies. NRI, Chatham, [Poster] [Field & Policy] [English]

Appendix 21 Mushobozi W and Grzywacz D (2003) "A new natural control for armyworm in Africa" 50 copies NRI, Chatham [leaflet] [Field] [English] p2

Appendix 22 Mushobozi W and Grzywacz D 2003 "A new natural control for an old pest in Africa" 50 copies NRI, Chatham [leaflet] [Policy] [English] p 4

Appendix 23 Mushobozi W (2003) New methods for control of armyworm in Tanzania. Ministry of Agriculture and Food Security Armyworm Control & Forecasting Services Tanzania [Oral presentation to Tanzanian National Advisory Plant Protection Committee April 2004]

Appendix 24 Wilson and Mushobozi (2002) "NPV disease in African armyworm", 100 copies, PCS and Stirling University. [Poster] [Field & Policy] [English]\*

Appendix 25 Wilson K.A. Armyworm Related cattle poisoning [Oral presentation to Tanzanian Ministry of agriculture February 2003]

Appendix 26 Grzywacz D, Parnell M, Mushobozi W and Wilson K. (2004) Development of *Spodoptera exempta* nucleopolyhedrovirus (SpexMNPV) for the control of African armyworm in East Africa. [Oral presentation to Society of Invertebrate Pathology 37<sup>th</sup> Annual meeting, Monday 2 August, Helsinki 1-6<sup>th</sup> August 2004]