FINAL TECHNICAL REPORT

SUSTAINABLE MANAGEMENT OF THE WHITEFLY, *BEMISIA TABACI*, AND TOMATO LEAF CURL VIRUS (ToLCV) ON TOMATO IN INDIA

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Executive Summary

The project's purpose was to develop and promote sustainable and cost-effective management practices for two principal researchable constraints, *Bemisia tabaci* and ToLCV, thereby improving both the quantity and quality of Indian tomato production.

In the first year of the project, a socio-economic survey was carried out to assess farmers' perceptions of the problem and 100% of farmers reported ToLCV to be their most serious problem.

Research activities involved field experiments into the effect of beneficial insect augmentation, mycopesticides, and ToLCV resistant varieties on the rate of spread of ToLCV into the tomato crop. Of these options, the ToLCV-resistant tomato lines showed the most promise in terms of delivering developmental impact and this was the output for which there was the greatest demand from tomato farmers. This was especially evident from the feedback obtained from farmers who attended the end of project farmer-field day/workshop.

Epidemiological data were also collected and incorporated into a mathematical model that was used to assess and identify potential novel control techniques. Those that increased and decreased, respectively, the vector emigration and immigration rate had the greatest potential for reducing the spread of the disease.

The outputs of the project were the production of more than ten inbred F_5 ToLCVresistant tomato lines with acceptable horticultural characteristics; *B. tabaci* parasitoids and predators (beneficial insects) in Karnataka identified and their potential for reducing *B. tabaci* populations and ToLCV incidence assessed; Indian fungal isolates assessed and compared with existing mycopesticides in field trials against *B. tabaci* and ToLCV; a simulation model built to determine the potential impact and conditions under which the proposed IPM practices are likely to be most successful; a report on farmers' perceptions and management practices related to *B. tabaci* and ToLCV and on socio-economic factors affecting the adoption of new varieties and IPM management strategies; recommendations for pest management practices, developed through farmer participation, for improved tomato production through control of *B. tabaci* and ToLCV; IPM recommendations published as a leaflet in Kannada accessible to extension workers and farmers and peer-reviewed research articles published in international journals.

The project outputs have contributed directly to the project purpose and therefore DFID's developmental goals by developing and disseminating measures that ameliorate the effects of ToLCV and *B. tabaci* on tomato growers in South India.

Background

In the recent past, over-reliance on conventional insecticides in vegetables and cotton production has resulted in the evolution of several, new, highly resistant whitefly, *Bemisia tabaci* (Gennadius), biotypes. These 'aggressive' strains outcompete the original biotypes, are highly mobile and, worldwide, cause hundreds of millions of pounds worth of damage annually. The development of alternative management practices, therefore, is vitally important both to prevent the evolution of additional aggressive biotypes and to cope with those already in existence.

In India, tomato is both a high value crop which is exported to the Middle East and an important subsistence vegetable grown mainly by women. *B. tabaci* is currently the most serious pest, causing direct damage as well as transmitting tomato leaf curl virus (ToLCV). ToLCV incidence is positively correlated both with the numbers of *B. tabaci* present and with yield loss (Saikia & Muniyappa, 1989). When populations of *B. tabaci* are high, 90 - 100% of plants can become infected resulting in a yield loss of 40 - 100% (Saikia & Muniyappa, 1989). With associated risks both to their health and the environment, farmers currently use large quantities of costly broad-spectrum insecticides in increasingly unsuccessful attempts to control *B. tabaci* and ToLCV.

In response to this potentially devastating problem, the University of Agricultural Sciences at Bangalore (UASB) initiated a research programme to introduce ToLCV-resistance genes into edible tomatoes through conventional plant breeding techniques (Muniyappa *et al.*, 1994). The UASB is committed to an Integrated Pest Management (IPM) approach to pest problems and has uptake channels for research through its Directorate of Extension and its laboratory-to-land programme, which conducts field trials with farmer participation. The UASB is keen to collaborate with IPM researchers in areas outside its field of expertise and the proposed project will address these needs.

In addition to whitefly and/or virus-resistant cultivars, sustainable, environmentally acceptable and potentially cheap methods of *B. tabaci* and ToLCV control include the use of beneficial insects and biological (myco) pesticides. These alternatives are mutually compatible and their interactions can provide multiplicative, rather than additive levels of control. For instance, due to their greater virulence to their adapted host, mycopesticides are intrinsically less harmful to natural enemies than conventional insecticides. Parasitism and predation can often proceed successfully with newly infected insects, while those already parasitised at the time of mycopesticide application normally complete their development. Indeed, predators and parasitoids in some cases have been implicated in the spread of pathogenic fungi within the crop (Lacey *et al.*, 1994).

The ODA paper on assistance to Renewable Natural Resources (RNR) in India identifies Karnataka State as an area where the rural poor are geographically concentrated and an impact on poverty seems feasible. In the same paper, whiteflies are mentioned in the section on indicative project profiles to meet RNR strategy objectives.

B. tabaci and ToLCV

In several studies, a very strong correlation has been recorded between the incidence of ToLCV and the size of the *B. tabaci* population (Anzola & Lastra, 1985; Ioannou & Iordanou, 1985; Cohen *et al.*, 1988; Saikia & Muniyappa, 1989; Verma *et al.*, 1989a; Singh, 1990). When populations of *B. tabaci* are high, 90 - 100% of susceptible plants can become infected with a consequent yield loss of between 40 - 100% (Shaheen, 1983; Jeyarajan *et al.*, 1988; Saikia & Muniyappa, 1989). Disease symptoms are particularly severe if the plants are infected at an early stage of development (Saikia & Muniyappa, 1986).

B. tabaci has a wide host range and Sastry (1984) identified 5 weed and 3 ornamentals as hosts and sources of inoculum for ToLCV. In the Kalyani area, India, *B. tabaci* and virus were found on 17 plant species including tomato, okra and bean, *Phaseolus vulgaris* (Verma *et al.*, 1989b). In the off-season in Rajastan, *B. tabaci* and TCLV survived on cucurbits and weeds (Bhardwaj & Kushwaha, 1984). Saikia & Muniyappa (1989) found that *B. tabaci* lives on 173 plant species around Bangalore and can transmit ToLCV to 23 of them. The incidence of ToLCV is at its highest in areas where tomato is grown continuously, year after year, and lowest where tomato has only recently been introduced (Ramappa *et al.*, 1994).

In response to this potentially devastating problem, the University of Agricultural Sciences at Bangalore (UASB) initiated a research programme with the long term aim of introducing ToLCV-resistance genes into edible tomatoes through conventional plant breeding techniques (Muniyappa *et al.*, 1991; Muniyappa *et al.*, 1994). The UASB is committed to an Integrated Pest Management (IPM) approach to pest problems and has uptake channels for research through its Directorate of Extension and its laboratory-to-land programme which conducts field trials with farmer participation. It also has links with the National Centre on Integrated Pest Management (NCIPM) which has a project on the management of ToLCV in India (Puri, 1994).

Predators and parasitoids

In the Indian subcontinent as a whole, at least 20 different species are known to attack *B. tabaci*. In India, however, there has been relatively little research on the predators and parasitoids of *B. tabaci* and the parasitoid complex remains poorly understood (Muniyappa, pers. comm.; Polaszek, pers. comm.). Of the 20 known beneficial species, 12 belong to a single genus, *Encarsia*, and are extremely difficult to differentiate (Polaszek *et al.*, 1992). In the past, *Encarsia* wasps have been highly successful in biocontrol programmes against whiteflies (Onillon, 1990).

In Europe, *Encarsia formosa* is produced commercially for release in glasshouses where it can successfully reduce populations of whitefly to acceptable levels. In combination with other natural enemies such as the predatory mirid bug *Macrolophus caliginosus* and the coccinellid *Delphastus pusillus*, even better levels of control can be achieved (5 - 10 *B. tabaci* per plant, Koppert Biological Systems, pers. comm.).

Field conditions are very different from the glasshouse environment and the short duration of vegetable crops grown in rapid succession favours the whitefly and limits the initial numbers of natural enemies on the crop, unless their numbers are artificially augmented or local reservoirs are present on native or weedy hosts. Crop phenology and diversity of surrounding vegetation are therefore extremely important in the build up of whitefly, virus and beneficial insect populations (Hoelmer, 1994). Even without natural enemy augmentation, parasitism of *B. tabaci* on unsprayed tomato and cauliflower in Egypt reached 78 - 84% (Abdel-Fattah *et al.*, 1984; Abdel-Gawaad *et al.*, 1990; Shalaby *et al.*, 1990).

Where pesticides are used extensively, the complex of natural enemies attacking whiteflies is adversely affected and parasitism rates are can fall to zero (Sharaf, 1984; Mohan, 1988).

Host-plant resistance

In one study, 1306 tomato cultivars, breeding lines and accessions were screened for ToLCV resistance under laboratory and field conditions. Among these genotypes, 9 showed

resistance, 37 exhibited mild and 41 moderate infection. The 43 Indian commercial tomato cultivars and hybrids were all highly susceptible (Muniyappa *et al.*, 1994). Resistance in lines of *Lycopersicon hirsutum* differed from other species in that it was shown to be due to glandular trichomes which entrapped *B. tabaci* before it could transmit the virus (Channarayappa *et al.*, 1992; Muniyappa *et al.*, 1994). Chelliah & Srinivasan (1983) also report that a sticky, glandular exudate on the aerial parts of *L. hirsutum* caused the death of *B. tabaci* individuals.

Some accessions of tomato are virtually free of the disease under field conditions, although they are potential hosts as inoculation by *B. tabaci* or grafting produced 96 - 100% infection (Shoba & Arumugam, 1990).

Mycopesticides

A diverse variety of entomopathogenic fungi attack *B. tabaci* and, under certain conditions, produce epizootics that result in significant control. Such epizootics have been reported from India and Pakistan and in one case the causal agent was identified as *Paecilomyces farinosus* (Nene, 1972), although its potential as a bioinsecticide has not been tested. The three most commonly recorded fungi attacking *Bemisia tabaci* under natural conditions are *Beauveria bassiana* (*Bb*), *Paecilomyces fumosoroseus* (*Pfr*) and *Verticillium lecanii* (*Vl*). All three fungi grow readily in culture on simple substrates to produce abundant conidia and all three have been developed as commercial biopesticides: *Bb* as "Naturalis L" by Troy Biosciences and as "Mycotrol WP" by Mycotech Corporation in USA for *Bemisia, Pfr* as "PFR 97" by W.R. Grace in USA for *Bemisia* in glasshouses and *Vl* as "Mycotal" by Koppert in Netherlands for *Trialeurodes* in glasshouses

These products are either emulsions (Naturalis L) or water-dispersible granules (PFR 97 and Mycotal) for application in water at high volume. The conidia of *Pfr* and *Bb* are hydrophobic and also suitable for application in ULV oil-based diluents. This technology has been tested against grasshoppers for *Bb* in the USA and West Africa and for the related fungus *Metarhizium flavoviride* against grasshoppers and locusts in West Africa, South Africa and Australia. *Vl* conidia are hydrophilic and emulsified formulations for ULV application have not yet been developed, although experimental technology exists.

Formulations, especially those in oil or emulsions, can enhance infectivity by lowering the LD50 and also by allowing infection to occur at low humidities. Studies by Smith (1993) at IIBC and in Trinidad have indicated that both emulsions and ULV oil formulations can be used successfully under conditions of low humidity. Emulsions may hold the most promise, because they can by used with conventional, low cost knapsack sprayers already familiar to most smallholder farmers.

Pfr is of particular interest because it is most frequently recorded causing natural epizootics in *B. tabaci* populations and therefore seems to have the greatest potential to spread following application. The secondary effects of pathogen application, where insects are infected from the inoculum produced on the cadavers of those infected by the initial spraying, has not been much studied but indications are from IIBC's locust work that it may be of importance, especially where humid conditions ensure sporulation on the dead insects. The possibility therefore exists that a spray application will cause both an initial kill and a more long-term effect.

Individual isolates of entomopathogenic fungi vary in their host range. A strain of *Pfr* isolated in Florida showed a wide host range, with insects in several host orders susceptible in laboratory trials. "Naturalis L" is also recommended for a wide range of target insects on cotton. Registration in USA requires data on host range and both fungi have been been

registered successfully (Wright, 1995). Naturalis-L is currently also being registered in Australia and India (Troy Biosciences, pers. comm.). Records of infection of beneficial Hymenoptera by *Vl*, *Bb* or *Pfr* are rare; both *Pfr* and *Bb* have been recorded from some beneficial Coleoptera or Diptera and *Bb* is a common pathogen of Coleoptera. Epizootics in beneficial insects are very rarely recorded in the field, probably because beneficials do not usually reach such high densities as pest species. Studies in the USA and Holland on the parasitiods *E. formosa* and *A. aleyrodis* suggest that they can act in a complementary manner and can even serve as a fungal vector (Lacy *et al.*, 1994).

Epidemiology and modelling

There is currently an absence of research effort in India in this potentially extremely useful and cost-effective area. Integrated pest management by definition requires consideration of the species complex, the different stages in the life-history of each species, the mortalities acting on these stages and how this integrates over time. Such a challenge lends itself to mathematical modelling (Waage & Barlow, 1993). Analytical modelling of pest-parasitoid-insecticide interactions in many agro-ecosystems has helped to identify a spectrum of possible outcomes ranging from resurgence, through self-cancelling effects to additivity and even synergism (Waage, 1989). Simulation modelling of particular pest systems has provided precise guidelines on the optimisation of insecticide use, options for resistance management and the avoidance of pest resurgence (Holt *et al.*, 1992; De Souza *et al.*, 1995). Simulations of potential pest management options also allow the most promising to be selected before more time consuming field trials are attempted (Holt *et al.*, 1990).

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Project Purpose

Indicative output 1.15 Improved methods for the management of insect pests of fruit and vegetables particularly whitefly and spider mites developed and promoted.

To improve both the quantity and quality of Indian tomato production through the sustainable and cost-effective management of two principal researchable constraints, *Bemisia tabaci* and ToLCV.

Research Activities

- Plant breeding and virus research is already ongoing at the UASB and 1306 tomato 1. cultivars, breeding lines and accessions have been screened for ToLCV resistance under laboratory and field conditions with the long term aim of introducing ToLCV/B. tabaci resistance genes into edible tomatoes. Research in this area will continue during the lifetime of the project and the most promising accessions or hybrids will provide the material for field trials investigating interactions between the virus, B. tabaci, the host plant, the beneficial insects and the entomopathogenic fungi. If resistant hybrids become available from the private sector, these will also be included in the trials. Field trials will have plots containing wide marginal guard rows to reduce effects due to insect mobility. The field trials will involve factorial experiments looking at the main effects and interactions between tomato variety, augmentation of beneficial insects and mycopesticide use. At a minimum, each factor will have two levels and each treatment will be replicated four times. The effects that we are looking for will be large and therefore this degree of replication should be sufficient to obtain significant differences. Later trials will involve more than 2 levels to get response curves. Data on whitefly numbers, ToLCV incidence, beneficial insect numbers and percentage mortality due to fungi will be collected at least three times during the growing season. When measurements are made over time, the relevant repeated measurement analysis will be used. Yield quantity and quality in the different treatments will be assessed at the end of the trials (years 1 - 3).
- 2a. Collection and identification of beneficial insects both within tomato fields and the surrounding agro-ecosystem (years 1 and 2). Expertise and training will be provided by NRI and IIE, with the necessary equipment for taxonomic research available at IIE.
- 2b. Literature survey to establish the most promising indigenous predator and parasitoid species in terms of ease of mass rearing and their regulatory effect on B. tabaci populations (years 1 and 2). Data bases, e.g. on Chalcidoidea, available at IIE.
- 2c. An innovative way of augmenting beneficial insect numbers during the crucial early season period would be to release them onto netting-protected seedlings 1 2 weeks before transplanting (protected seedlings usually have sufficient *B. tabaci* present to sustain a beneficial insect population). Selected beneficial insect species will be reared on a small scale for field trials to evaluate this or similar techniques (years 2 and 3). Expertise provided by NRI and UASB. Insect rearing facilities at UASB will require modification for the maintenance of *B. tabaci* and beneficial insect cultures and a new small glasshouse will be required. Trial sites are available at UASB.
- 3a. Existing mycopesticide products, Naturalis-L, PFR-97 and Mycotrol WP will be sought and their efficacy tested at IIBC in comparison with at least two Indian fungal isolates from the USDA-ARS collection (Year 1). *B. tabaci* will be reared at NRI and given to IIBC for the bioassay work. IIBC has already studied several fungi species for their use as control agents against *B. tabaci* (Smith, 1993).
- 3b. Dr Prior will visit Bangalore to provide advice on the mycopesticide component of the field trials and on application technology (years 1-3). Application equipment is available from IIBC and UASB.

- 4a. Data on ToLCV and *B. tabaci* rates of spread within plots will be collected during the field trials of 1, 2c and 3b and will be used, in addition to the quantity and quality of the yield, to assess the efficacy of the treatments. NRI and the IIE have considerable experience in the vector ecology and natural enemies of whiteflies arising from previous projects in Kenya, Côte d'Ivoire, Malawi, Tanzania and Uganda (Polaszek *et al.*, 1992; Thresh *et al.*, 1994; Fishpool *et al.*, 1995).
- 4b. Data will be collected on the *B. tabaci* population and ToLCV incidence on the different types of vegetation (including weeds) surrounding the trial sites. The UASB has the expertise and equipment to detect ToLCV infected plants and insects using the triple antibody sandwich (TAS-ELISA) technique. NRI has a comparative advantage in quantitative virus-disease epidemiology, statistical and simulation modelling, with spatial lattice and differential equation models currently being developed to examine crop disease dynamics with a view to developing IPM strategies and recommendations (e.g. Holt & Chancellor, in press). A simulation model will be built using parameter estimates obtained from statistical modelling.
- 5a. Survey and identification of different types of tomato farmers and other stakeholders (*eg.* nursery growers, agrochemical suppliers). Analysis of the role of women in vegetable production. Identification of farmers willing to participate in further project research (year 1).
- 5b. Survey of tomato farmers' perceptions of and responses to *B. tabaci*, ToLCV, beneficial organisms (linked with outputs from 2a), resistant varieties and IPM in Karnataka (year 2).
- 5c. Social and economic feasibility studies of proposed measures developed in 1-4, including cost comparisons. Feedback of results into activities 1-4 (years 2&3).
- 6a. Development of most succesful ToLCV/*B. tabaci* control measures in on-farm trials. Carried out by farmers and UASB extension workers and project researchers.
- 6b. Farmers' demonstrations of control measures and workshop for the discussion and dissemination of research findings. To include farmers, researchers and extension workers from Karnataka as well as other Indian states (year 3).
- 7a. Handbook(s) or poster(s) designed for extension staff and farmers produced in local language (incorporating information from activities 1 6 above) (year 3).
- 7b. Data analysis and papers written (years 1 3).

The above research activities were carried out as planned and the inputs were achieved. The following additional activities took place.

Additional funding 1996/97: Provision of training in PCR techniques to Dr Muniyappa, UASB at NRI and project staff at UASB in collection and identification of natural enemies of Bemisia tabaci

Dr Muniyappa received training in PCR at NRI which provided him with the background and information to set up a molecular genetics laboratory at the UASB. This is now in operation and PCR reactions are currently being carried out. Please see additional funding 1998/99 below.

Project staff were trained in the collection and identification of natural enemies by Dr Polaszek during a 5 day visit to the UASB. This enabled the beneficial insect component of the project to be carried out successfully. Additional funding 1998/99: Travel and subsistence costs for Dr Colvin and Dr Muniyappa to give presentations at the 2nd International Workshop on Bemisia and Geminiviral Diseases, San Juan, Puerto Rico, 7-12 June 1998

Drs Colvin & Muniyappa travelled to the 2nd International Workshop on Bemisia and Geminiviral Diseases, San Juan, Puerto Rico, 7-12 June 1998 where the work of the project was presented.

Additional funding 1998/99: Training for project staff at UASB in PCR methodologies and techniques for assessing seasonal ToLCV inoculum pressure; also funding for Dr Colvin to visit to UASB to organise and run a workshop/farmer field day for local tomato farmers, extension workers and researchers

Dr Richard Thwaites travelled to Bangalore and spent 7 days training UASB staff (Mr H.M.Venkatesh and Dr H.A. Prameela) in PCR methodologies and techniques for assessing seasonal ToLCV inoculum pressure. The visit was highly successful, as ToLCV was detected both in extractions from plant material and from individual adult *B. tabaci*. This will enable a considerable quantity of new work to be attempted and will potentially greatly increase our understanding of the ToLCV pathosystem with consequent benefits in disease management.

A workshop/farmer-field day was organised and held on the 26th and 27th of February 1999. A small booklet listing the timetable, abstracts and participants has been compiled.

Outputs

Output 1: At least one ToLCV/B. tabaci-resistant tomato variety assessed for use in IPM

Tomato leaf curl virus (ToLCV) is a disease of tomato which can cause severe yield loss. Infected plants have curling, wilted leaves and are often stunted in growth. If the plants are infected early then there may be no yield at all. Leaf curl disease is caused by a gemini virus and spread through whiteflies (*Bemisia tabaci*). The whiteflies have a wide host range and can transmit ToLCV to many different plant species.

Around Bangalore, in Karnataka, India, ToLCV occurs regularly and is a major disease of tomato. The incidence is highest in the hot, summer months.

Screening of tomato varieties at Bangalore initially began with 10 genotypes sent from AVRDC, Taiwan. Figure 1 shows the range of disease progress curves and yields obtained from a selection of these lines.

Over the three year lifetime of the project, screening in both the glasshouse and field has been carried out during five field seasons and several very promising lines are being developed. It will require several more generations of selection before these lines are ready for release to farmers.

Culture of Tomato Leaf Curl Virus

Stock culture of tomato leaf curl virus (ToLCV) was maintained on tomato cultivar Arkavikas / hybrid Rashmi and was maintained in the glass-house by frequently inoculating with whiteflies *Bemisia tabaci*.

Whitefly culture

A virus free *Bemisia tabaci* culture was maintained on cotton, *Gossypium hirsutum*, cv. Laxmi which is immune to tomato leaf curl virus. Healthy cotton plants were frequently introduced into the insect cages ($45 \times 45 \times 30$ cm) while removing old plants. The cages were maintained at a temperature range of 28-35 C in a glass house.

Cages for whitefly acquisition and inoculation

Plastic and polyvinyl chloride (PVC) cages of different sizes ($7.5 \times 2.5 \text{ cm}$; $8 \times 7 \text{ cm}$; $22 \times 6 \text{ cm}$) were used for vector acquisition and inoculation of ToLCV. Cages were prepared by removing the bottom portion and then covering with muslin cloth. A small hole was made in the wall of the cage in order to release adult whiteflies and was then plugged with cotton wool. PVC round bottles prepared in the same way were used for virus acquisition by the adult *B. tabaci*. The tapering end of the bottle was plugged with a cotton-wool ball after inserting a ToLCV infected tomato twig.

Inoculation of tomato seedlings

Different tomato genotypes were sown in the glass-house in pots filled with autoclaved soil and farm yard manure. Eight days after germination the seedlings were transplanted individually in polythene bags.

Adult *B. tabaci_were* collected from the stock culture and released into PVC tubes containing ToLCV infected tomato twigs. Whiteflies were allowed to feed for 24 hours on infected tomato (acquisition access period). Fifteen-day old seedlings were individually covered with plastic / PVC tubes and 10-15 viruliferous adults were released on to each seedling. They were allowed to feed for 48 hours as the inoculation access period. Inoculated seedlings were kept in the glass house for symptom expression.

Screening of AVRDC tomato genotypes for Tomato leaf curl virus by Bemisia tabaci inoculation in glass house

Tomato genotypes ATY-1 (*L.e.* H24), ATY-2 (*L.e.* H36), ATY-3 (*L.e.* TyKing), ATY-4 (*L.e.* TyKing F_2 sel.), ATY-5 (*L.e.* Fiona F_1), ATY-7 (*L.e.* Ty 52), ATY-10 (*L.e. L.pe.* VL 215), ATY-11 (*L.c.* LA 1969), ATY-12 (*L.e.* Ty 50 (susc.)), ATY-13 (*L.e.* TK 70 (susc.)) and local cultivar Arkavikas (local susc. ck.) were sown in the glass house on 17-11-96 and transplanted individually in polythene bags on 4-12-96. The seedlings were individually inoculated using plastic / PVC tubes. 10-15 viruliferous adults were released per seedling. *B. tabaci* adults were given 24 h acquisition access and 48 h inoculation access periods. Seedlings were inoculated three times. After removing plastic tubes, the seedlings were kept in the glass house with subsequent releases of viruliferous whiteflies into the glasshouse. First symptom appearance, number of days taken for full symptom expression, number of infected plants and severity were recorded (Table 1).

Four weeks after inoculation, one ATY-1 plant showed infection and only 3 plants showed infection 8 weeks after inoculation. ATY-1 took 32 to 87 days to show full symptom expression. ATY-10 plants did not show any ToLCV symptoms. ATY-11 seedlings died after few weeks. ATY-13 was very susceptible.

Screening AVRDC tomato genotypes for ToLCV resistance under field conditions

Genotypes ATY-1, 2, 3, 4, 5, 7, 10, 11, 12, 13 and a local check Arkavikas were sown in the

| ATY No. | Tomat | o line | No. plants inoculated | No. of plan (visual) | nts infected | No. of days to First / final symptom Expression | Hybridis. test ³ | Symptoms severity |
|---------|---------------|--------------------------------------|-----------------------|-------------------------|--------------|---|--------------------------------|-------------------|
| | | | | 4 wks | 14 wks | | | |
| ATY-1 | <i>L. e.</i> | H-24 | 37 | 2 | 23 | 28/87 | 0/6 | Mild |
| ATY-2 | <i>L. e.</i> | H-36 | 39 | 29 | 39 | 28/30 | NT | Mild |
| ATY-3 | <i>L. e.</i> | Tyking | 39 | 39 | 39 | 14/21 | NT | Moderate |
| ATY-4 | <i>L. e.</i> | Tyking sel. | 30 | 27 | 30 | 14/30 | NT | Moderate |
| ATY-5 | <i>L. e.</i> | Fiona F1 | 40 | 32 | 40 | 21/30 | NT | Moderate |
| ATY-7 | <i>L. e.</i> | Ty 52 | 38 | 38 | 38 | 15/21 | NT | Mod./Sev. |
| ATY-10 | <i>L. pe.</i> | VL 215 | 36 | 0 | 0 | - | 0/10 | None |
| ATY-11 | L. c. | LA 1969 | 18 | 0 | Dead | - | NT | None |
| ATY-12 | <i>L. e.</i> | Ty 50 (susc.) | 28 | 28 | 28 | 10/15 | NT | V. severe |
| ATY-13 | <i>L. e.</i> | Tk 70 (susc.) | 45 | 45 | 45 | 10/15 | 4/4 | V. severe |
| | <i>L. e.</i> | Arkavikas (local susceptible. check) | 48 | 48 | 48 | 15/21 | 2/2 | V. severe |

Table 1. Response of AVRDC tomato genotypes to inoculation by *B. tabaci* with ToLCV in the glasshouse at Bangalore

Date of sowing : 17/11/96 Date of transplanting: 4/12/196

Date of inoculation: 2-5 January 1997 (10-15 viruliferous *B. tabaci*/seedling; 48 h inoculation access period) repeated 3 times.

Date of final observation: 19-4-97

Sample collected for hybridisation test: 12/5/97, Hybridisation conducted at AVRDC, using the 16i probe.

nursery under field conditions on 7-4-97. Seedlings were transplanted into the field on 6-5-97. A local susceptible check variety was planted all round the plot. Every week incidence was recorded.

For 26 ATY-1 plants, only 3 were infected and symptoms were mild. Growth was moderate and they produced small to medium sized fruits. Severe cracking of fruits was observed. ATY-2 was also less susceptible and showed mild symptoms. Fruit cracking was observed. ATY-12, ATY-13 and Arkavikas were highly susceptible. ATY-11 did not establish (Tables 2, 3 and 4).

Reaction of F_5 AVRDC tomato genotypes to ToLCV under glasshouse and field conditions

In the F_5 generation of screening in the glasshouse, several lines have proved extremely promising in terms of resistance to ToLCV and other fruit qualities (Table 5). This generation was also tested under field conditions and showed similar very promising results (Table 6) to those produced under glasshouse conditions.

Twenty six F_5 tomato genotypes and other eleven genotypes including resistant and susceptible checks were tested against tomato leaf curl virus both under laboratory and field conditions. All F_5 genotypes showed resistance to ToLCV. Seven genotypes which performed well with respect to fruit yield and other horticultural characters were selected for further field trials (TLB 111, 77332 kg/ha; TLB 119, 96665 kg; TLB 122, 80888 kg; TLB 129, 101332 kg; TLB 130, 74888 kg; TLB 134, 94221 kg; TLB 148, 48800 kg; susceptible check TLB 146, 17333 kg; resistant check TLB 147, 44444 kg) (Table 6).

The farmers have evaluated F5 lines by considering resistance to ToLCV, foliage of plants, fruit bearing, size of the fruit, shape of the fruit, transportability, marketability, firmness of fruit and thickness of the skin and taste of the fruits (Table 7).

The most promising seven ToLCV-resistant genotypes have been selected for multilocation on-station trials in the Bangalore area. This work is planned to be the first experiment of the second three year phase of this project. This is the first time systemic ToLCV resistance has been successfully introduced into tomato lines with acceptable horticultural characteristics in India and the demand for our varieties from all stakeholders is clearly evident. As such, we feel certain that there will be a ready uptake of these lines resulting in a large developmental impact.

Inoculation of pepper and chilli genotypes with tomato leaf curl virus by Bemisia tabaci in the glass house

Pepper and chilli lines were inoculated with ToLCV using viruliferous *B. tabaci* to see whether they were susceptible to ToLCV. Pepper and chilli are often grown adjacent to tomato fields and this work was conducted primarily to see whether these crops acted as additional virus sources for one another.

| ATY No. | Tomato line | | No. of No. of plants infected plants (Visual observation) | | | | Hybrid- isation test | No. of days to first/final symptom expression | Symptoms |
|---------|---------------|-------------------------------------|---|-------|-------|--------|----------------------------|--|-----------|
| | | | | 4 wks | 8 wks | 13 wks | | | |
| ATY-1 | <i>L. e.</i> | H-24 | 26 | 0 | 3 | 3 | 0/22 | 35/56 | Mild |
| ATY-2 | <i>L. e.</i> | H-36 | 19 | 1 | 4 | 8 | 4/18 | 28/77 | Mild |
| ATY-3 | <i>L. e.</i> | Tyking | 17 | 3 | 17 | 17 | 1/17 | 35/42 | Mod. |
| ATY-4 | <i>L. e.</i> | Tyking sel. | 5 | 5 | 5 | 5 | 0/4 | 21/42 | Mod./sev. |
| ATY-5 | <i>L. e.</i> | Fiona F1 | 8 | 2 | 8 | 8 | 2/7 | 28/77 | Mod./sev. |
| ATY-7 | <i>L. e.</i> | Ту 52 | 2 | 2 | 2 | 2 | 0/2 | 21/35 | Sev. |
| ATY-10 | <i>L. pe.</i> | VL 215 | 32 | 0 | 0 | 0 | 0/16 | 0 | None |
| ATY-11 | L. e. | LA 1969 | 30 | Dead | - | - | NT | - | |
| ATY-12 | <i>L. e.</i> | Ty 50 (susc.) | 4 | 4 | 4 | 4 | 2/2 (+/-) | 14/28 | Severe |
| ATY-13 | <i>L. e.</i> | Tk 70 (susc.) | 13 | 13 | 13 | 13 | 10/10 (+/-) | 7/28 | V. severe |
| | <i>L. e.</i> | Arkavikas (local susceptible check) | 154 | 154 | 154 | 154 | 5/5 (+/-) | 7/35 | V. severe |

Table 2. Response of AVRDC tomato genotypes to ToLCV under field conditions at Bangalore

Sowing (field nursery) : 7/4/97

Transplanting : 6/5/97

Plot surrounded by local susceptible check.

Final symptom observation : 5/8/97 (13 wk after transplanting).

Samples collected for hybridisation : 18/8/97

Hybridisation done at AVRDC, using the 16i probe.

| | | No. of plants Inoculated | | No. of plants infected (greenhouse) | | No. Res. plants transplanted to field | No. of plants infected (field) | | Symptoms | No. of days to first/ full symptom expression |
|---------|-------------------------------|-----------------------------|-------|-------------------------------------|-----------|--|--------------------------------|-----------|----------|--|
| ATY No. | Tomato line | | V | isual | Hybridis. | - | Visual | Hybridis. | | × |
| | | | 3 wks | 8 wks | | - | 12 wks 16 wks | | | |
| ATY-1 | L. e. H-24 | R1 16 | 0 | 1 | 0/16 | 10 | 0/8 | 4/8 | Mild | 54/54 |
| | | R2 16 | 0 | 0 | 0/16 | 12 | 2/10 | 1/10 | | - |
| ATY-10 | L. pe VL 215 | R1 16 | 0 | 0 | 0/16 | 11 | 0/9 | 0/9 | Mild | - |
| | • | R2 16 | 0 | 1 | 0/16 | 9 | 0/5 | 0/5 | | 51/51 |
| ATY-11 | L. c. LA 1969 | R1 8 | 0 | 0 | 0/8 | 4 | | | - | - |
| | | R2 9 | 0 | 0 | 0/9 | 4 | | | - | - |
| ATY-13 | L. e. TK 70 | R1 16 | 15 | 16 | 1/1 | 0 | | | V. sev. | 12/26 |
| | | R2 16 | 16 | 16 | 2/2 | 0 | | | | 12/26 |
| ATY-14 | L. e. FL 744-6-9-(BL 1163) | R1 16 | 0 | 2 | 1/14 | 10 | 3/9 | 1/9 | Mild | 56/56 |
| | | R2 16 | 0 | 3 | /13 | 9 | 0/9 | 0/9 | | 56/56 |
| ATY-15 | L. e. FL 736 (BL 1165) | R1 16 | 8 | 16 | 2/2 | 0 | | | Sev. | 12/55 |
| | | R2 16 | 10 | 16 | 1/1 | 0 | | | | 9/55 |
| ATY-16 | L. e. FL 699 sp. (BL 1166) | R1 16 | 0 | 0 | 1/16 | 11 | 0/10 | 0/10 | Mild | - |
| | - | R2 9 | 0 | 1 | /9 | 5 | 0/5 | 1/5 | | 33/33 |
| ATY-17 | L. e. FL 699 sp + (BL 1167) | R1 16 | 0 | 1 | 1/16 | 12 | 2/12 | 1/12 | Mild | 33/33 |
| | - | R2 16 | 0 | 3 | /16 | 10 | 0/10 | 0/10 | | 47/55 |
| ATY-18 | L. e. FL 776 (BL 1169) | R1 16 | 2 | 15 | 8/6 | 8 | 4/8 | 1/8 | Mod. | 26/62 |
| | | R2 16 | 4 | 14 | /6 | 8 | 1/2 | 2/2 | | 19/62 |
| ATY-19 | L. e. FL 619 (BL 1170) | R1 10 | 6 | 8 | 1/4 | 5 | 2/4 | 3/4 | Mod. | 12/33 |
| | | R2 0 | - | - | - | - | - | - | | - |
| ATY-20 | L. e. FL 805 (BL 1171) | R1 16 | 2 | 10 | 6/9 | 6 | 1/5 | 0/5 | Mod. | 12/62 |
| | | R2 16 | 6 | 13 | /7 | 3 | 0/1 | 0/1 | | 12/62 |
| ATY-21 | L. e. FL 505 (BL 1172) | R1 16 | 0 | 0 | 0/16 | 11 | 0/4 | 0/4 | - | 0/0 |
| | · · | R2 16 | 0 | 0 | 0/16 | 11 | 0/5 | 0/5 | - | 0/0 |
| | L. e. Arkavikas (local s. ck) | R1 16 | 13 | 16 | | 0 | | | V. sev | 19/33 |
| | | R2 16 | 12 | 16 | | 0 | | | | 19/33 |

Table 3. Response of AVRDC tomato genotypes to ToLCV at Bangalore under glasshouse inoculation and field conditions

Sowing (field nursery): 5/7/97; Transplanting to field: 22/9/97; Inoculation: 2 inoculations: 21/7/97, 15 viruliferous B. tabaci/seedling

2 replications (R1, R2) (randomized block) local susceptible check planted between each genotype. Susceptible Arkavikas planted in the rows for every 2 rows of test plants) Samples from resistant plants taken for hybridization : 18/9/97. Hybridization done at AVRDC, using the 16I probe. Membranes were then reprobed with the Sri Lanka probe. The results are combined here

Samples taken for hybridization : 31/11/97 (= 16 wks after inoculation with whiteflies). Hybridization done at AVRDC, using the Sri Lanka (16i) probe.

| Genotyp | Infected | Average | Average no. | Total fruit | Average | Remarks |
|---------|----------|-------------|-------------|-------------|---------------|-------------------|
| e | / | length / | of fruits | weight / | fruit | |
| | No. of | plant | /plant | plant | weight | |
| | plants | (cm) | | | | |
| ATY-1 | 3/26 | 39.8 | 31.5 | 1157.40 g | 36.70 g | Fruit cracking, |
| | | | | | | small to medium |
| | | | | | | sized fruits |
| ATY-2 | 8/19 | 39.0 | 23.0 | 942.60 g | 40.98 g | Fruit cracking, |
| | | | | | | small to medium |
| | | | | | | sized fruits |
| ATY-3 | 17/17 | 51.9 | 19.8 | 1232.70 g | 62.20 g | Slightly wrinkled |
| | | | | | | fruits |
| ATY-4 | 5/5 | 58.5 | 22.0 | 1377.00 g | 62.50 g | Medium sized |
| | | | | | | fruits |
| ATY-5 | 8/8 | 95.0 | 27.8 | 1317.00 g | 47.30 g | Medium size; |
| | | | | | | indeterminate |
| | | | | | | growth |
| ATY-7 | 2/2 | 40.0 | 40.0 | 1413.00 g | 35.30 g | Small to medium |
| | | | | | | size |
| ATY-10 | 0/32 | 91.4 | - | - | - | - |
| ATY-11 | died | - | - | - | - | - |
| ATY-12 | 4/4 | 38.5 | 10.5 | 445.00 g | 42.30 g | Medium size |
| ATY-13 | 13/13 | 23.7 | 10.2 | 307.10 g | 30.10 g | Fruit size was |
| | | | | | | reduced due to |
| | | 40 - | | 207.00 | 2 0 50 | severe infection |
| Arkavi- | 154/154 | 40.7 | 14.4 | 297.80 g | 20.60 g | Fruit size was |
| kas | | | | | | reduced due to |
| | | | | | | severe infection |

Table 4. Growth and yield characteristics of AVRDC tomato genotypes screened forToLCV resistance under field conditions

Date of sowing: 7-4-97

Date of transplanting: 6-5-97

Date of final harvesting: 20-8-97

Seeds were collected from the individual plants of ATY-1 (21 plants), ATY-2 (18), ATY-3 (17) and ATY-10 (16).

| Sl No. | TLB No. | Entry Number | No. | 3rd | 6th | 9th | 12th | First/final sympton | sympton severity |
|--------|---------|------------------------------|--------------|------|------|------|------|---------------------|------------------|
| | | | inoculated | week | week | week | week | expression | |
| 1 | 110 | CLN 2114 DC1 F1-2-16-8-2 | 12 | 0 | 0 | 0 | 0 | | Resistant |
| 2 | 111 | CLN 2114 DC1 F1-2-29-7-2 | 15 | 0 | 0 | 0 | 0 | | Resistant |
| 3 | 112 | CLN 2114 DC1 F1-2-29-16-22 | 5 | 0 | 0 | 0 | 0 | | Resistant |
| 4 | 113 | CLN 2114 DC1 F1-2-29-20-23 | 12 | 2 | 2 | 2 | 2 | 13 / 15 | 2 Mod, 10 R |
| 5 | 114 | CLN 2114-DC1 F1-2-42-41 | 12 | 0 | 0 | 0 | 0 | | Resistant |
| 6 | 115 | CLN 2114 DC1 F1-2-48-17-4 | 12 | 2 | 4 | 4 | 4 | 13 / 44 | 3 Se, 1 Mod, 8 R |
| 7 | 116 | CLN 2114 DC1 F1-94-25-22-9 | 4 | 4 | 4 | 4 | 4 | 13 / 15 | 4 Se, 9 R |
| 8 | 117 | CLN 2114 DC1 F1-94-29-5-20 | Not germinat | ed | | | | | |
| 9 | 118 | CLN 2114 DC1 F1-94-29-16-24 | 4 | 2 | 2 | 2 | 2 | 13 / 16 | 2 Mod, 2 R |
| 10 | 119 | CLN 2116 DC1 F1-180-50-15-8 | 13 | 0 | 0 | 0 | 0 | | Resistant |
| 11 | 120 | CLN 2116 DC1 F1-270-19-10-6 | 13 | 4 | 6 | 6 | 7 | 13 / 70 | 6 Se, 1 Mi, 6 R |
| 12 | 121 | CLN 2116 DC1 F1-270-19-10-15 | 12 | 1 | 1 | 1 | 2 | 19 / 75 | 1Se, 1 Mi, 10 R |
| 13 | 122 | CLN 2116 DC1 F1-270-19-10-16 | 13 | 3 | 4 | 4 | 5 | 15 / 77 | 4 Se, 1 Mod, 8 R |
| 14 | 123 | CLN 2116 DC1 F1-270-19-10-17 | 15 | 2 | 4 | 4 | 5 | 10 / 70 | 4 Se, 1 Mi, 10 R |
| 15 | 124 | CLN 2121 DC1 F1-26-26-19-2 | 8 | 0 | 0 | 0 | 3 | 70 / 75 | 3 Mi, 5 R |
| 16 | 125 | CLN 2123 DC1 F1-111-2-13-4 | 8 | 2 | 3 | 3 | 3 | 12 / 31 | 2 Se, 1 Mi,5 R |
| 17 | 126 | CLN 2123 DC1 F1- 111-2-13-8 | 7 | 2 | 2 | 2 | 2 | 15/21 | 2Se,5R |
| 18 | 127 | CLN 2123 DC1 F1-111-2-13-16 | 13 | 0 | 0 | 0 | 1 | 70 | 1Mi,12R |
| 19 | 128 | CLN 2123 DC1 F1-120-23-2417 | 14 | 0 | 0 | 0 | 0 | | Resistant |
| 20 | 129 | CLN 2123 DC1 F1-120-23-2424 | 15 | 4 | 4 | 4 | 6 | 12/77 | 4sv,1mod,1Mi,9R |
| 21 | 130 | CLN 2131 DC1 F1-96-46-17-6 | 13 | 1 | 2 | 2 | 2 | 12/31 | 2Se,11R |
| 22 | 131 | CLN 2131 DC1 F1-96-46-17-13 | 15 | 4 | 4 | 4 | 5 | 12/70 | 3Sev,2Mi,10R |
| 23 | 132 | CLN 2131 DC1 F1-96-46-17-15 | 11 | 0 | 0 | 0 | 1 | 77 | 1Mod,10R |
| 24 | 133 | CLN 2131 DC1 F1-96-46-17-18 | 7 | 0 | 0 | 0 | 0 | | R |
| 25 | 134 | CLN 2131 DC1 F1-96-46-17-32 | 15 | 1 | 5 | 5 | 5 | 12/31 | 5sev, 10R |
| 26 | 135 | CLN 2131 DC1 F1-96-46-20-16 | 12 | 2 | 3 | 3 | 3 | 12/31 | 3sev 9R |
| 27 | 136 | CLN 2131 DC1 F1-96-46-20-25 | 11 | 1 | 1 | 1 | 1 | 12/12 | 1Se, 10R |

Table 5. Response F_5 AVRDC tomato genotypes to ToLCV under glasshouse conditions

| Sl No. | TLB No. | Entry Number | No. inoculated | 3rd week | 6th week | 9th week | 12th week | First/final sympton expression | sympton severity |
|--------|---------|----------------------------|-------------------|-------------|-------------|-------------|--------------|-----------------------------------|------------------|
| 28 | 137 | CLN 2131 DC1 F1-9646-20-27 | Not germinat | ed | | | | | |
| 29 | 138 | FANTASTIC | 14 | 12 | 14 | 14 | 14 | 12/31 | 14 Sev |
| 30 | 139 | BONNY BEST | 14 | 7 | 14 | 14 | 14 | 12/29 | 14 Sev |
| 31 | 140 | UC-82-L | 13 | 9 | 13 | 13 | 13 | 12/29 | 13 Sev |
| 32 | 141 | FLORIDA MH-1 | 13 | 12 | 13 | 13 | 13 | 12/27 | 13 Sev |
| 33 | 142 | RODADE | 12 | 6 | 12 | 12 | 12 | 15/27 | 12 Sev |
| 34 | 143 | 13R -1(BL-783) | 8 | 5 | 8 | 8 | 8 | 15/30 | 8 Sev |
| 35 | 144 | TSW-10 (BL 1022) | 11 | 6 | 11 | 11 | 11 | 13/27 | Sev |
| 36 | 145 | CLN2052F2 | 11 | 2 | 4 | 4 | 4 | 20/42 | 4 Mod 7R |
| 37 | 146 | CL5915-93 D4(CH 45) | 11 | 11 | 11 | 11 | 11 | 10/15 | Se |
| 38 | 147 | BL837 (H24) | 50 | 0 | 0 | 0 | 0 | 0 | Resistant |
| 39 | 148 | RASHMI | 30 | 30 | 30 | 30 | 30 | 20/42 | Se |

Table 5 continued

Serial No. 1-15, 37,38 & Rashmi : Date of Sowing : 8-9-98. Date of transplanting :15-9-98; Date of inoculation ; 17-9-98

Date of observation ; 8/10 (3rd wk), 29/10 (6th wk), 19-11-98 (9th wk), 10-12-98 (12th wk)

Serial No. 16-36: Date of sowing :15-9-98; Date of transplanting: 23-9-98; Date of inoculation: 29-9-98

Date of observation: 19-10-98 (3rd wk); 9-11-98 (6th wk) 30-11-98 (9th wk) : 21-12-98(12th wk)

Each seedling was inoculated by Bemisia tabaci.

Symptom severity: Se = Severe infection; Mo = Moderate infection; Mi = Mild infection; R = Resistant (no visible symptoms)

| Genotype | Number of | | of plan | | | Average fruit | Yield /ha |
|--------------------|-----------|----------------|---------------|--------------|----------------|----------------------------|-----------|
| | plants | <u>we</u> 3 | eks afte 6 | er plar 9 | nting12 | yield / plant ^a | (kg/ha) |
| TLB 110 | 8 | $\frac{3}{0}$ | 0 | <u> </u> | $\frac{12}{0}$ | 1.650 | 36666 |
| TLB 110 TLB 111 | 8 33 | 0 | 0 | - | 0 | 3.48 | 77332 |
| | 55 7 | | | 0 | | | |
| TLB 112 | | 0 | 0 | 0 | 0 | 3.220 | 71554 |
| TLB 113 | 20 | 0 | 0 | 0 | 0 | 4.520 | 10044 |
| TLB 114 | 21 | 0 | 0 | 0 | 0 | 3.21 | 71332 |
| TLB 115 | 8 | 0 | 0 | 0 | 0 | 3.47 | 77110 |
| TLB 116 | 21 | 1 | 5 | 5 | 5 | 2.29 | 50888 |
| TLB 119 | 28 | 0 | 0 | 0 | 0 | 4.35 | 96665 |
| TLB 120 | 20 | 1 | 5 | 6 | 6 | 4.17 | 92665 |
| TLB 121 | 29 | 1 | 4 | 4 | 4 | 4.04 | 89776 |
| TLB 122 | 22 | 1 | 1 | 2 | 2 | 3.64 | 80888 |
| TLB 123 | 22 | 1 | 1 | 1 | 1 | 3.27 | 72665 |
| TLB 125 | 15 | 2 | 3 | 3 | 3 | 3.59 | 79776 |
| TLB 127 | 11 | 0 | 0 | 0 | 0 | 2.30 | 51110 |
| TLB 128 | 22 | 0 | 0 | 0 | - | 4.71 | 104665 |
| TLB 129 | 29 | 0 | 1 | 1 | - | 4.56 | 101332 |
| TLB 130 | 33 | 1 | 2 | 4 | - | 3.37 | 74888 |
| TLB 131 | 22 | 2 | 4 | 6 | - | 4.09 | 90887 |
| TLB 132 | 12 | 0 | 0 | 0 | - | 3.05 | 67777 |
| TLB 133 | 22 | 0 | 0 | 0 | - | 3.78 | 83999 |
| TLB 134 | 33 | 2 | 3 | 3 | - | 4.24 | 94221 |
| TLB 135 | 10 | 1 | 1 | 1 | - | 2.18 | 48443 |
| TLB 136 | 21 | 0 | 0 | 1 | - | 3.40 | 75554 |
| H-24 | 36 | 0 | 0 | 0 | 0 | 2.00 | 44444 |
| CL 5915 | 20 | 5 | 20 | 20 | 20 | 0.78 | 17333 |
| Rashmi | 113 | 51 | 110 | 113 | 113 | 1.60 | 35555 |
| Arkavikas | 41 | 3 | 18 | 38 | 41 | 1.16 | 25777 |
| Ramya | 36 | 3 | 9 | 30 | 36 | 1.80 | 39999 |

TABLE 6. FIELD EVALUATION OF F_5 tomato genotypes against ToLCV

^a Average of 10 plants Date of sowing : 9.11.98 Date of transplanting : 8.12.98

| Sl. No. | TLB No. | AVRDC No. | Fruit characters |
|------------|---------|---|--|
| 1 | 111 | CLN 2114 DC ₁ F ₁ -2-29-7-2 | Round shaped, slightly less than medium sized fruits, good yielding. |
| 2 | 119 | CLN 2116 DC ₁ F ₁ -180-50-15-8 | Round shaped, medium sized fruits (70-90 gms), deep red, high yielding. |
| 3 | 122 | CLN 2116 DC ₁ F ₁ -270-19-10-16 | Round shape, more than medium size fruits, deep red colour, high yielding, more foliage. |
| 4 | 129 | CLN 2123 DC ₁ F ₁ -120-23-24-24 | Oblong shape, light red, nipple at the basal end of the fruit, high yielding; good in other characters such as fruits / cluster, more foliage etc, except the shape of the fruits. |
| 5 | 130 | CLN 2131 DC ₁ F ₁ -96-46-17-6 | Round shaped, medium sized fruits, high yielding. |
| 6 | 134 | CLN 2131 DC ₁ F ₁ -96-46-17-32 | Oval shaped, medium sized fruits and deep red colour, high yielding and good taste. |
| 7 | 148 | CLN 2116 DC ₁ F ₁ -180-1-1-7 | Oval shape, deep red above medium sized fruits, good yielding. |

Pepper lines AHP-1, AHP-2, AHP-3, AHP-4, AHP-5, AHP-6, AHP-7 (AVRDC lines), California wonder and chilli cv. G-4 were sown in pots containing sterilised soil and kept in the glass house for germination. Seedlings were transplanted in polythene bags at the rate of one per bag.

Adults of B. tabaci were collected from the stock culture and released onto ToLCV infected tomato twigs in PVC tubes for the acquisition access period.

Plastic / PVC tubes were covered on pepper and chilli seedlings. With the help of aspirator 15-20 viruliferous adult whiteflies were released onto each seedling. Allowed to feed for 48 hours as the inoculation access period. Three sequential inoculations were made in this way. Subsequently tubes were removed and viruliferous adult whiteflies were released on the seedlings.

Only one plant in AHP-3 bcame infected and showed vein clearing, vein thickening and curling. It took 66 days to show the symptoms. All the pepper and chilli lines were clean and did not show any visible symptoms. It will be interesting to check whether all these lines are virus free or symptomless carriers. Leaf squashes of these plants were mailed to AVRDC for nucleic acid hybridisation test (Table 8). These results suggest that the ToLCV does not readily infect pepper or chilli.

| Sl no. | Genotype | Visual | Hybridis* | No. of days to | Symptoms | |
|---------|-----------------|---------------|------------|---------------------------------|---------------------|--|
| | | observation | test | full symptom | | |
| | | No. of plants | | expression | | |
| | | W. symptoms / | | | | |
| | | Total exposed | | | | |
| 1. | AHP-1 | 0/8 | 8 | - | No symptoms | |
| 2. | AHP-2 | 0/13 | 13 | - | No symptoms | |
| 3. | AHP-3 | 1/15 | 15 | 66 | Vein clearing, vein | |
| | | | | | thickening, curling | |
| 4. | AHP-4 | 0/16 | 16 | - | No symptoms | |
| 5. | AHP-5 | 0/18 | 18 | - | No symptoms | |
| 6. | AHP-6 | 0/12 | 12 | - | No symptoms | |
| 7. | AHP-7 | 0/14 | 14 | - | No symptoms | |
| 8. | California | 0/30 | 30 | - | No symptoms | |
| | wonder | | | | | |
| 9. | Chilli G4 | 0/30 | 30 | - | No symptoms | |
| Date of | sowing: 11-6-9 | 97 | Dates of g | Dates of germination 22/25-9-97 | | |
| Date of | inoculation: 3- | 7-97, 5-7-97, | Date of t | transplanting: 1-7 | -97 | |

| Table 8. | Inoculation of | Chilli and Pepper | genotypes with | vith tomato leaf | curl virus by B. |
|--------------------|-----------------|--------------------------|----------------|------------------|------------------|
| <i>tabaci</i> in t | the glass house | | | | |

7-7-97.

AAP 24 h; IAP 48 h.

Date of final observation: 17-9-97.

Output 2: Systematics of B. tabaci parasitoids and predators (beneficial insects) in Karnataka determined and their potential for reducing B. tabaci populations and ToLCV incidence assessed

Parasitoids and predators were collected from different places in different cropping systems in Karnataka. Plant shoots infested with parasitoid nymphs were collected and placed into labelled paper bags and were brought back to the laboratory. Each parasitoid nymph was separated along with the leaf and was transferred into a gelatin capsule for adult emergence. The adults were kept for 24 h after emergence in the capsules before they were collected and preserved in 80 % ethanol in a sealed tube. Parasitic wasps and predators were also collected directly using an aspirator and sweep nets in the field. The adults obtained were first card mounted and then permanent slides were prepared by following the canada balasm method as described by Noyes (1982).

The specimens were identified using a key provided for the recognition of previously described *Encarsia* specis which are known to attack *B. tabaci* (Polaszek et al., 1992). Some parasitoids and predators were identified by the International Institute of Entomology and the British Museum (Natural History), London. The parasitoid species *Encarsia transvena* Timberlake, *E. adrianae* Lopez Avela and *Eretmcerus mundus* Mercet were identified. Amongst the parasitoid species, *E. tranvena* was the most commonly occuring in Karnataka (Table 9). Among many predators found in tomato fields, *Axinoscymnus puttarudriahi* Kapoor and Munshi was very common. This predator was also found feeding on eggs and nymphs of *B. tabaci* in the whitefly culture cages (Table 10).

Stock cultures of the parasitoids were established on *B. tabaci* on cotton plants in rearing cages. The cotton plants were heavily infested with *B. tabaci*. These plants were kept in the whitefly culture room in cages until day 12 after oviposition then were set up on a tray. Leaves with whitefly nymphs (third instar) were exposed to the adult parasitoids in the parasitoid culture room.

Beneficial insects in the management of Bemisia tabaci and ToLCV

Nursery: Two nursery beds measuring 12 x 4 ft were prepared and the beds were sown with tomato hybrid Rashmi. The beds were covered with 50 mesh nylon nets. About 3000 *B. tabaci* were relaeased into each nursery bed 15 days after sowing. Thirteen days after relaease of *B. tabaci*, about 500 newly emerged *E. transvena* and *E. adrianae* were released into one of the nursery beds.

Main field: Plots measuring 27 x 36 ft were prepared and three sides of each were covered with 50 mesh nylon nets (3 1/2 ft height) to restrict the movement of beneficial insects from one plot to other. One side was kept open to allow the migration of *B. tabaci* into the tomato plot. Twenty nine days old tomato seedlings were transplanted at a spacing of 3 x 2 ft in four randomized replications.

| Date of collection | Place of collection | Host plant | Parasitoid species |
|--------------------|----------------------------|---------------|--|
| 11/11/1996 | Hebbal, Bangalore | Cotton | Encarsia transvena |
| 12/11/1996 | Hebbal, Bangalore | Cotton | E. adrianae |
| 13/11/1996 | Hosakote, Bangalore | Tomato | E. transvena |
| 21/11/1996 | Darwar | Cotton | E. transvena |
| 22/11/1996 | Hebbali, Darwar | Cotton | E. transvena |
| 08/02/1997 | Hebbal, Bangalore | Cotton | E. transvena |
| 21/03/1997 | Channasandra, Bangalore | Tomato | E. transvena, Eretmoceros mundus |
| 29/03/1997 | Hebbal, Bangalore | Cotton | E. mundus |
| 26/04/1997 | Hebbal, Bangalore | Euphorbia sp. | E. transvena |
| 02/09/1997 | Doddaballapur | Rose | E. transvena, E. mundus |

Table 9. Bemsia tabaci parasitoid species collected from different host plants in Karnataka, south India

| Predator | Family | Desription |
|---|----------------|---|
| <i>Cryptotacnius montrouzier</i> i Mudsant | Coccinellidae | This is an Australian species. Widely distributed throughout the world. Used as biocontrol agent |
| Dolichopodid fly | Dolichopodidae | Adults are general predator of insects |
| Nephus regularis (Sicard) | Coccinellidae | This is a wide spread Indian species and predator of scale insects especially mealy bugs |
| <i>Leptobatopsis indica</i> (Cameron) | Ichneumonidae | It has been recorded from various Lepidopteran hosts |
| <i>Auxinoscymnus puttarudriahi</i> Kappor & Munshi | | An Indian species known to feed on whitefly eggs and nymphs |
| <i>Coccinella</i> transversali Fabricius | Coccinellidae | This is wide spread oriental and Australian species. It is a general aphid predator |

Table 10. Predators commonly found in tomato crops in Karnataka

Beneficial insect augmentation

Parasitoids: Newly emerged *E. transvena, E. adrianae* and *E. mundus* were collected in tubes from the culture room and were released in the center of beneficial insect augmented plots at 200 per plot. Four such releases were made at weekly interval.

Predators: The eggs of *Chrysoperla carnea* Stephens used in the experiments were obtained from the Biocontrol Research Laboratory, Bangalore. The eggs were dusted onto tomato plants along with the filler material @ 10 eggs per plant, a day prior to the date of hatching. Four releases were made at weekly intervals. Incidence of leaf curl was recorded at weekly interval by counting the number of infected plants based on visual symptoms. Cylindrical yellow sticky traps were used to monitor *B. tabaci* adults. Two traps were placed in each plot, one at the center and other on the periphery of the open end of the plot. Adult *B. tabaci* were conted twice weekly and cellophane strips were changed every week.

The incidence of ToLCV was first noticed in the margins of the open end of the plot two weeks after transplanting and later the spread was irregular. The incidence of ToLCV reached 100 % six weeks after planting in both the augmented and control plots and there was no significant differences between the treatments in the rate of ToLCV spread or in the number of adult *B. tabaci* per plant (Fig.2). Parasitism of *B. tabaci* nymphs within the augmented plots (27.2 %) was higher than in the check plots (20.3 %), although the difference was not significant (Tables 11 & 12). Tomato yield in the augmented plots (21.5 tonnes/ha) was significantly higher than that of the check plots (18.7 tonnes/ha) (Table 13). There was no significant difference between the *B. tabaci* catch of the two traps placed at the periphery and inside the tomato plots. The average number of whiteflies per trap during the crop growth was 200 and the population decreased when the crop reached maturity (Table 14). This clearly indicated that the immigration of whteflies started immediately after transplanting and that these immigrants are responsible for the rapid spread of ToLCV in the plots.

Interaction between parasitoids and entomopathogenic fungi

The interaction of parasitoid nymphs and entomopathogenic fungi was studied by using the leaf dip method. The results indicated that nymphs of both *E. transvena* and *E. adrianae* were infected by all the tested entomopathogenic fungi. *V. lecanii* showed maximum infection (60.0 %) on nymphs of both *E. tranvena* and *E. adrianae* which was followed by *B. bassiana* and *P. farinosus* (Table 15). All the three entomopathogenic fungi tested were found to be pathogenic to adult *E. transvena*. *V. lecanii* recorded maximum per cent mortality (91.3 %) followed by *B. bassiana* (72.5 %) after 72 h of treatment (Table 16). Mortality was noticed after 48 h in all the entomopathogenic fungi. *E. adrianae* was also found to be killed by all the three fungi. *V. lecanii* recorded maximim per cent mortality (60.0 %) followed by *B. bassiana* (53.3 %) 72 h after inoculation (Table 17).

A field trial was carried out to examine the effect of augmenting the beneficial insect population by releasing parasitoids at the time of transplanting. Adult and nymph whitefly populations were monitored in the ToLCV-susceptible tomato crop simultaneously and it became evident that disease progression was extremely rapid (Fig. 2) due mainly to plants becoming infected by immigrant adult whitefly. The number of *B. tabaci* nymphs recorded on tomato was very low and therefore very little secondary spread took place. As a result of the low number of offspring produced by *B. tabaci* on tomato, the effect of the beneficial insects was minimal.

In plots where beneficial insects were released, the yield was just significantly higher although this result is hard to explain in terms of disease progression within the crop. These results suggest that given the effort involved in the mass production of predators and parasitoids, the returns to be obtained in terms of increased tomato yield are neligible.

| | Average number of <i>B. tabaci</i> on each plant | | | | | | |
|-----------------------|--|----------------------|----------------|---------------------------------|------------------------|------------------|--|
| Treatment | Replication | | | | - | | |
| | | Developmental stages | Pupal cases | No. of nymphs parasitised | Total no. of nymphs | % parasitization | |
| 1. Beneficial insects | R1 | 5.40 | 8.50 | 4.05 | 17.95 | 22.56 | |
| | R2 | 5.25 | 9.65 | 6.15 | 21.05 | 29.22 | |
| | R3 | 4.55 | 10.35 | 6.10 | 21.00 | 29.04 | |
| | R4 | 5.15 | 9.85 | 5.85 | 20.85 | 28.05 | |
| Treatment mean | | 5.08 | 9.58 | 5.54 | 20.21 | 27.21 | |
| 2. Check | R1 | 4.50 | 9.25 | 3.00 | 16.75 | 17.91 | |
| | R2 | 4.85 | 10.85 | 4.95 | 20.65 | 23.97 | |
| | R3 | 4.65 | 12.65 | 3.95 | 21.25 | 18.58 | |
| | R4 | 4.90 | 12.45 | 4.50 | 21.65 | 20.78 | |
| Treatment mean | | 4.72 | 11.30 | 4.10 | 20.07 | 20.31 | |

| | abe a generation |
|---|---|
| Table 11. Effect of beneficial insects on <i>B. tabaci</i> developm | nental stages ^{","} in the field |

^a *B. tabaci* counted on 20 plants in each plot ^b Counts were made at the end of crop by destructive sampling.

| Treatment ^a | Mean no. of nymphs parasitised per tomato $plant^{c} \pm SE$ | Р |
|---|--|----------|
| Beneficial insect augmentation ^b | 5.5 ± 0.5 | P = 0.07 |
| Control | 4.1 ± 0.42 | |
| ^a No of replications -4 | | |

Table 12. Effect of beneficial insect augmentation on *B. tabaci* parasitism

^aNo. of replications = 4. ^b Encarsia transvena, E. adrianae.

^cDestructive sampling of 20 plants/replicate at the end of the experiment. Date of sowing: 1/3/97; date of transplanting: 31/3/97; hybrid Rashmi.

| Treatment ^a | Mean yield \pm SE (tonnes ha ⁻¹) | Р |
|---|--|----------|
| Beneficial insect augmentation ^b | 21.5 ± 0.68 | P = 0.02 |
| Control | 18.7 ± 0.59 | |

Table 13. Effect of beneficial insect augmentation on tomato yield

^aNo. of replications = 4.

^b Encarsia transvena, E. adrianae, Chrysoperla carnea.

Date of sowing: 1/3/97; date of transplanting: 31/3/97; hybrid Rashmi.

| Treatment ^b | Weeks after transplanting | | | | | |
|------------------------|---------------------------|--------|--------|--------|-------|-------|
| | Ι | III | V | VII | IX | XI |
| T1 P1 H1 | 12.50 | 12.25 | 19.50 | 13.50 | 6.25 | 4.00 |
| T1 P2 H1 | 9.00 | 11.75 | 19.75 | 15.50 | 7.75 | 3.50 |
| T2 P1 H1 | 17.50 | 14.25 | 20.25 | 18.75 | 7.00 | 2.50 |
| T2 P2 H1 | 17.00 | 14.00 | 20.00 | 14.25 | 6.25 | 3.25 |
| T1 P1 H2 | 14.50 | 18.00 | 36.00 | 26.00 | 16.50 | 7.00 |
| T1 P2 H2 | 18.75 | 18.25 | 44.00 | 25.50 | 10.75 | 5.75 |
| T2 P1 H2 | 22.25 | 18.75 | 31.00 | 32.25 | 15.75 | 5.25 |
| T2 P2 H2 | 22.00 | 21.25 | 32.25 | 24.00 | 9.00 | 4.50 |
| T1 P1 H3 | 63.00 | 93.00 | 156.00 | 131.75 | 57.25 | 36.25 |
| T1 P2 H3 | 51.75 | 117.00 | 188.25 | 155.00 | 57.75 | 34.50 |
| T2 P1 H3 | 82.25 | 100.50 | 136.25 | 136.50 | 45.75 | 29.25 |
| T2 P2 H3 | 79.50 | 113.25 | 192.50 | 176.75 | 45.75 | 28.75 |
| | | | | | | |
| Sem +/- | 5.98 | 4.14 | 13.34 | 11.56 | 3.69 | 2.05 |
| | | | | | | |
| CD (5%) | 16.58 | 11.46 | 36.95 | 32.03 | 10.24 | 5.70 |

cylindrical yellow sticky traps) in the field experiment

Treatment 1 = Beneficial insects (*Encarsia transvena*, *E. adrianae* and *Chrysoperla carnea*); Treatment 2 = Control

^b Two poles in each plot and each pole has three heights (top = H1, H2 = middle, H3 = bottom)

^b Four replications

| Table 15. | The interaction between parasitoid nymphs and entomopathogenic |
|-----------|--|
| fungi | |

| Parasitoid | Entomopathogenic fungi ^a | % infection ^b | |
|--------------|-------------------------------------|--------------------------|--|
| E. transvena | B. bassiana | 53.3 | |
| E. transvena | V. lecanii | 60.0 | |
| E. transvena | P. farinosus | 50.1 | |
| E.adrianae | B. bassiana | 50.0 | |
| E.adrianae | V. lecanii | 60.0 | |
| E.adrianae | P. farinosus | 50.1 | |

^a The concentration used 1 x 10 7 spores/ml ^b Average of 4 repications

Table 16. The interaction between adult Encarsia transvena and entomopathogenic fungi

| Period after treatment (h) | Entomopathogenic fungi ^a | % Mortality ^b |
|----------------------------|-------------------------------------|--------------------------|
| 24 | B. bassiana | 0.0 |
| 48 | B. bassiana | 23.8 |
| 72 | B. bassiana | 72.5 |
| 24 | V. lecanii | 0.0 |
| 48 | V. lecanii | 68.7 |
| 72 | V. lecanii | 91.2 |
| 24 | P. farinosus | 0.0 |
| 48 | P. farinosus | 21.3 |
| 72 | P. farinosus | 45.8 |
| 24 | Check | 0.0 |
| 48 | Check | 3.0 |
| 72 | Check | 7.0 |

^a The concentration used 1 x 10⁷ spores/ml ^b 4 replications; 20 adults per treatment

| Table 17. | The interaction between adult Encarsia adrianae and | Ĺ |
|-----------|---|---|
| entomopa | thogenic fungi | |

| Period after treatment (h |) Entomopathogenic fungi a | % Mortality b |
|---------------------------|----------------------------|---------------|
| | | |
| 24 | B. bassiana | 0.0 |
| 48 | B. bassiana | 18.8 |
| 72 | B. bassiana | 53.8 |
| 24 | V. lecanii | 0.0 |
| 48 | V. lecanii | 30.0 |
| 72 | V. lecanii | 60.0 |
| 24 | P. farinosus | 0.0 |
| 48 | P. farinosus | 27.3 |
| 72 | P. farinosus | 50.1 |
| 24 | Check | 0.0 |
| 48 | Check | 3.0 |
| 72 | Check | 7.0 |

^a The concentration used 1 x 10⁷ spores/ml
^b 4 replications; 20 adults per treatment

This project contributed to the funding of Mr Venkatesh's PhD and he is currently in the process of writing his thesis. It is anticipated that he will be able to produce at least three publications from it and that he will have received his PhD by the end of June 1999.

Output 3: Indian fungal isolates assessed and compared with existing mycopesticides against B. tabaci

Collection, isolation and identification of entomopathogenic fungi

Cotton leaves infested with *B. tabaci* were collected from the whitefly stock culture maintained on cotton. The different hosts plants in and around the University of Agricultural Sciences, GKVK, Bangalore were examined periodically for dead and infected insects. These were collected carefully with a brush and placed into sterile plastic tubes with screw caps, labelled and brought to the laboratory. The collected specimens were surface sterilised with 0.5 per cent sodium hypochlorite solution for 2-3 minutes, rinsed in three changes of sterile distilled water to remove the traces of sodium hypochlorite solution in order to prevent the toxicity of sodium hypochlorite to the fungi. After surface sterilization these were kept in Sabouraud's Dextrose Agar + Yeast extract (SDAY) medium or in Potato dextrose agar (PDA) in petriplates. In the case of sporulating cadavers, the mycelium or the spores were picked up using a sterile inoculation needle and placed directly on the medium in the petriplates. The plates were prepared by pouring 15 to 20 ml of the molten medium per petriplate. The inoculated plates were incubated at room temperature for five to seven days and observed for the growth of fungi. The fungus growth was picked up using a sterile inoculation needle and transferred to SDAY or PDA slants in test tubes. The isolated micro-organisms were further purified by the hyphal tip method and single spore isolation (Tuite, 1969) and stored in the refrigerator at 4°C.

Identification of cultures

Efforts were made to identify the fungi based on morphological characters. The identity was further confirmed by Dr. Moore, Scientist, International Institute of Biological Control, U.K. The results showed that among the isolates, *Aspergillus* spp. was the most commonly encountered fungi followed by *Trichothecium* sp. Even though the cultures isolates were not primary pathogens, these mycoflora associated with whiteflies exert a pressure on the survival of population of whiteflies (Table 18). The cultures of entomopathogenic fungi were also procured from other centres (Table 19).

Pathogenicity of isolates

The fungal isolates locally isolated from whiteflies and other insects proved to be not the primary pathogens of *B. tabaci*. The cultures of *B. bassiana*, *V. lecanii, Paecilomyces farinosus, P. javanicus* and *P. lilacinus* were pathogenic to *B. tabaci*. These fungi re-isolated from the whiteflies were used for further studies.

| Crop | Place of collection | Insect/ stage | Name of isolate | Number of isolates |
|---|-------------------------|------------------------------|--------------------------|--------------------|
| Cotton | Glasshouse, | B. tabaci | Aspergillus sp. | 3 |
| Cotton | Hebbal | (adult) | 1 0 1 | - |
| T | | × / | <i>Trichoderma</i> sp. | 1 |
| Tomato | Glasshouse, | B. tabaci | Phoma ap. | 2 |
| Duinial. | Hebbal | (adult) | Aspergillus sp. | 1 |
| Brinjal | GKVK farm | B. tabaci | Aspergillus sp. | 1 |
| Tobacco | Glasshouse | Aphids | Actinomycetes | 1 |
| Cowpea | Glasshouse | Aphids | Aspergillus sp. | 1 |
| Cotton | Glasshouse | B. tabaci | <i>Mucor</i> sp. (nymph) | 1 |
| Mango | Farm, GKVK | Spiralling | Trichothecium sp. | 1 |
| | | whiteflies | Aspergillus sp. | |
| <i>Phyllanthes</i> sp. | Tomato field | <i>B. tabaci</i> (nymph) | Unidentified | 3 |
| Weeds | Unweeded | B. tabaci | Unidentified | 1 |
| (Euphorbia geniculata, Ageratum sp.) | Sorghum plot, Hebbal | (adult) | | |
| Lantana sp. | Hosakote | <i>B. tabaci</i> (adults) | Aspergillus sp. | 1 |
| | Hosakote | Aphids | Unidentified | 1 |
| Rose | Nagarjuna Agri. | B. tabaci | Unidentified | 1 |
| | Tech Polyhouse | (adult) | | |
| Tomato | Kolar | B. tabaci | Aspergillus sp. | 1 |
| | Talagawara | B. tabaci | Unidentified | 2 |
| | U U | | Aspergillus sp. | 1 |
| | | | Tricchothecium | 1 |
| | Lakkondanahally | Aphid | Mucor sp. | 1 |
| | <u>,</u> | grasshopper | Unidentified | 1 |

 Table 18. Mycoflora associated with Bemisia tabaci and other insects.

| Fungus species ^a | Source | |
|--|--------------------------------|--|
| | | |
| Beauveria bassiana (Balls) Vvill | MTCC ^a , Chandigarh | |
| Paecilomyces farinosus (Holm and Gray) | MTCC, Chandigarh | |
| Brown and Smith | | |
| P. javanicus (Fredrichs and Bally) Brown | MTCC, Chandigarh | |
| and Smith | | |
| Verticillium lecanii (Zimm) Viegas | MTCC, Chandigarh | |
| P. lilacinus (Thom) Samson | IIHR ^b , Banglore | |

^a Microbial Type Culture Collection, Chandigarh

^b Indian Institute of Horticulture Research, Hessaraghatta

Evaluation of different media for the growth and multiplication of entomopathogenic fungi

A study on the evaluation of different media on the growth and multiplication of entompathogens was conducted at room temperature $(28 \pm 1^{\circ}C)$. The growth and sporulation of *B. bassiana* and *V. lecanii* was evaluated on eight different solid media viz., Sabouraud's Maltose Agar + Yeast extract (SMAY), Sabouraud's Dextrose Agar + Yeast extract (SDAY), Potato Dextrose Agar (PDA), Modified Leonolian medium (MLM), Leonolian medium (LM), Yeast Extract Agar (YEA), Malt Extract Agar (MEA) and Potato Carrot Agar (PCA). The results are presented in Table 20 & 21. Statistically high significant differences were observed between media with respect to mycelial growth expressed in terms of colony diameter and sporulation 14 days after inoculation of *B. bassiana*. The maximum colony diameter was recorded on PDA (69.55 mm) followed by YEA (56.60 mm) which was statistically equivalent to that of MEA (53.95 mm), SMAY (21.90 mm) and SDAY (50.45 mm). The least growth was recorded on PCA (36.7 mm).

Sporulation in different media was compared by estimating the spore count of a 5 mm disc cored from the centre of a 14 day old colony. The sporulation was maximum in SDAY extract (4.548×10^8 spores) followed by YEA (2.364×10^8 spores), PCA (1.467×10^8 spores) and MEA (1.22×10^8 spores). Moderate sporulation was recorded on SMAY (8.68×10^7 spores) which was not significantly different to the sporulation produced in PDA (5.07×10^7 spores) and LM (3.41×10^7 spores). Least sporulation was recorded in MLM (1.92×10^7 spores). There was a highly significant difference between media with respect to mycelial growth and sporulation of the 14 day old culture of *V. lecanii*. In the case of *V. lecanii*, maximum mycelial growth was recorded in YEA (35.75 mm) which was on a par with RM (25.25 mm). This was followed by MEA (32.9 mm) and PDA (32.9 mm) and was on a par with LM (32.10 mm). The mycelial growth observed in

SMAY and SDAY were 27.3 mm and 27.2 mm respectively. Least growth was recorded in MLM (26.55).

| Treatment | Media | Mean colony | Spore count / |
|-----------|---|---------------|-------------------------|
| No. | | diameter (mm) | 5 mm disc |
| 1 | Sabouraud's Maltose Agar + Yeast Extract | 51.90 | 8.68 x 10 ⁷ |
| 2 | Sabouraud's Dextrose Agar + Yeast Extract | 50.45 | $45.48 \ge 10^7$ |
| 3 | Potato Dextrose Agar | 69.55 | 5.07×10^7 |
| 4 | Modified Leonolian Medium | 37.20 | $1.92 \ge 10^7$ |
| 5 | Leonolian Medium | 48.40 | 3.34×10^7 |
| 6 | Yeast Extract Agar | 56.60 | 23.64 x 10 ⁷ |
| 7 | Malt Extract Agar | 53.95 | 12.22×10^7 |
| 8 | Potato Carrot Agar | 36.70 | $14.67 \ge 10^7$ |
| | | | |
| | F test | ** | ** |
| | S.Em <u>+</u> | 2.28 | 1.48×10^{7} |
| | C.D. at 5% | 6.56 | 7.08×10^7 |

| Table 20. Effect of different solid | media on the mycelial growth and sporulation of |
|-------------------------------------|---|
| B. bassiana | |

** Significant at P = 0.01

Table 21. Effect of solid media on the mycelial growth and sporulation of Verticillium lecanii

| Treatment | Media | Mean colony | Spore count / |
|-----------|---|---------------|-------------------------|
| No. | | diameter (mm) | 5 mm disc |
| 1 | Sabouraud's Maltose Agar + Yeast Extract | 27.30 | 17.96 x 10 ⁷ |
| 2 | Sabouraud's Dextrose Agar + Yeast Extract | 27.20 | 19.96 x 10 ⁷ |
| 3 | Potato Dextrose Agar | 22.90 | 3.09×10^7 |
| 4 | Modified Leonolian Medium | 26.55 | $0.78 \ge 10^7$ |
| 5 | Leonolian Medium | 32.10 | $1.62 \ge 10^7$ |
| 6 | Yeast Extract Agar | 35.75 | $11.78 \ge 10^7$ |
| 7 | Malt Extract Agar | 32.90 | 6.83×10^7 |
| 8 | Potato Carrot Agar | 28.95 | 3.74×10^7 |
| 9 | Richard's Medium | 35.25 | $1.66 \ge 10^7$ |
| | | staate | starte |
| | F test | ** | ** |
| | S.Em <u>+</u> | 0.86 | $0.26 \ge 10^7$ |
| | C.D. at 5% | 2.37 | 0.72×10^7 |

** Significant at P = 0.01

The above trend was different, however, in the case of sporulation. Sporulation in different media was compared by estimating the spore count from a 5 mm disc cored from the centre of the 14 day old colony. Maximum sporulation was observed in SDAY (1.996 x 10^8 spores) which was closely followed by SMAY (1.796 x 10^8 spores) and YEA (1.178 x 10^8 spores). Moderate sporulation occurred in MEA (6.83 x 10^7 spores), PCA (3.74 x 10^7 spores) and PDA (3.095 x 10^7 spores). Least sporulation was recorded in MLM (7.8 x 10^6 spores).

Evaluation of liquid media for the growth and sporulation of *B. bassiana* **and** *V. lecanii*

The growth and sporulation of the fungus was studied on eight different liquid media viz., Sabouraud's Maltose broth + Yeast extract (SMBY), Sabouraud's Dextrose broth + Yeast extract (SDAY), Potato Dextrose broth (PDB), Modified Leonolian Broth (MLB), Leonolian Broth (LB), Yeast Extract Broth (YEB), Malt Extract Broth (MEB) and Potato Carrot Broth (PCB). The growth in terms of dry mycelial weight and sporulation per 20 ml of the broth was recorded. There were statistically significant differences between difference media with respect to dry mycelial weight and sporulation of *B. bassiana* (Table 22). The mean dry mycelial weight of B. bassiana was maximum in SDBY (410 mg) followed by SMBY (318 mg) and MEB (316 mg). The mean dry mycelial weight recorded in PDB, YEB and LB were 90 mg, 52 mg and 48 mg, respectively. The minimum dry mycelial weight was observed in PCB and MLB (10 mg each). The sporulation in different liquid media was compared by estimating the spores produced in 20 ml of each media after 14 days of inoculation with *B. bassiana*. The sporulation was maximum in SDBY (5.108 x 10^9) followed by MEB (1.938 x 10^9) which is on par with YEB (1.522×10^9) . Moderate sporulation was observed in SMBY (1.038×10^9) and PDB (1.041×10^8) . Less sporulation was produced in PCB (6.03 x 10^7), which was on a par with LB (5.36 x 10^7). Least sporulation was recorded in MLB (2.27 x 10^7). A statistically significant difference was observed between the media with respect to dry mycelial weight and sporulation of V. lecanii. The growth of the fungus in liquid media was expressed in terms of dry mycelial weight produced from 20 ml of broth. The dry mycelial weight produced by different media showed statistically significant differences. Maximum dry mycelial weight was recorded in SDBY (420 mg). This was followed by MEB (334 mg) which was on par with SMBY (320 mg) and RB (298 mg). The dry mycelial weight produced in YEB and PDB were 170 mg and 136 mg which were statistically on par. LB recorded less dry mycelial weight of 68 mg and least was recorded in MLB (12 mg). The sporulation in different liquid media was compared by estimating the sporulation produced in 20 ml of each media 14 days after inoculation with V. lecanii. There was a statistically significant difference in the sporulation of V. lecanii in different broths. Maximum sporulation was recorded in SDBY (2.758×10^9) , followed by RB (1.972 x 10⁹), SMBY (1.948 x 10⁹) and YEB (1.152 x 10⁹). Moderate sporulation was produced in PDB (8.24 x 10^8). Minimum sporulation was recorded in MLB (6.55 x 10^8) and PCB (6.32 x 10^8) (Table 23).

Table 22. Effect of liquid broth on the mycelial growth and sporulation of Beauveria

bassiana

| Treatment | Media | Dry mycelial | Sproulation |
|-----------|---|--------------|-----------------|
| No. | | weight (mg) | $(x \ 10^7)$ |
| 1 | Sabouraud's Maltose Agar + Yeast Extract | 318 (17.79) | 103.80 (10.21) |
| 2 | Sabouraud's Dextrose Agar + Yeast Extract | 410 (20.26) | 510.80 (22.39) |
| 3 | Potato Dextrose Agar | 90 (9.44) | 10.41 (3.28) |
| 4 | Modified Leonolian Medium | 10 (3.24) | 2.27 (1.66) |
| 5 | Leonolian Medium | 48 (6.91) | 5.36 (2.42) |
| 6 | Yeast Extract Agar | 52 (7.24) | 152.20 (12.33) |
| 7 | Malt Extract Agar | 316 (17.77) | 193.80 (13.80) |
| 8 | Potato Carrot Agar | 10 (3.24) | 6.03 (2.55) |
| | F test | ** | ** |
| | S.Em ± | 0.40 | $0.68 \ge 10^7$ |
| | C.D. at 5% | 1.16 | $1.89 \ge 10^7$ |

Figures in parentheses are $\sqrt{(x + 0.5)}$ transformed values

** Significant at P = 0.01

Table 23. Effect of liquid broth on the mycelial growth and sporulation of *Verticillium lecanii*

| Treatment | Media | Dry mycelial | Sproulation |
|-----------|---|--------------|-------------------------|
| No. | | weight (mg) | (spores / 20 ml) |
| 1 | Sabouraud's Maltose Agar + Yeast Extract | 320 (17.87) | 19.48 x 10 ⁸ |
| 2 | Sabouraud's Dextrose Agar + Yeast Extract | 420 (20.50) | $27.58 \ge 10^8$ |
| 3 | Potato Dextrose Agar | 136 (11.57) | $8.24 \ge 10^8$ |
| 4 | Modified Leonolian Medium | 12 (3.50) | $6.55 \ge 10^8$ |
| 5 | Leonolian Medium | 68 (7.92) | $5.50 \ge 10^8$ |
| 6 | Yeast Extract Agar | 170 (12.86) | $11.52 \ge 10^8$ |
| 7 | Malt Extract Agar | 334 (18.27) | $14.10 \ge 10^8$ |
| 8 | Potato Carrot Agar | 20 (4.47) | $6.32 \ge 10^8$ |
| 9 | Richard's broth | 298 (17.27) | 19.72 x 10 ⁸ |
| | | | |
| | F test | ** | ** |
| | S.Em <u>+</u> | 0.68 | $0.73 \ge 10^8$ |
| | C.D. at 5% | 1.96 | 2.02×10^8 |

Figures in parentheses are $\sqrt{(x + 0.5)}$ transformed values

** Significant at P = 0.01

Mass production of mycopesticide using the two stage technique

The mass production of *B. bassiana* and *V. lecanii* to apply as mycopesticide in the field was standardised as per Jenkins (1996) with slight modifications. The production of conidia was done in two stages. The first phase was preparation and inoculation of liquid medium and the second stage was the preparation and inoculation of solid rice substrate. The mass production of entomopathogenic fungi viz., *B. bassiana* and *V. lecanii* by the two phase production technique was standardised

The mass production of spores of *B. bassiana* by the two phase production technique was standardised. An average yield of 4.0×10^9 conidia per g of rice inoculum was obtained. The harvested spores were made into the required concentration of spray solution in aqueous Tween 80-0.05 per cent and used for spraying in the field. The mass production of spores of *V. lecanii* by the two phase production technique was standardised. An average yield of 2×10^9 conidia per g of inoculum on rice was obtained. The harvested spores were made into the required concentration of spray solution in aqueous Tween 80-0.05 per cent and used for spraying in the field. The mass production of spores of *V. lecanii* by the two phase production technique was standardised. An average yield of 2×10^9 conidia per g of inoculum on rice was obtained. The harvested spores were made into the required concentration of spray solution in aqueous Tween 80-0.05 per cent and used for spraying in the field.

Studies on mass production of entomopathogenic fungi

Locally available materials such as broken rice, broken maize grains, broken sorghum grains, rice bran and wheat bran were compared as substrates for the mass production of spores of entomopathogens viz., B. bassiana and V. lecanii. One hundred grams of each of the substrates was weighed in polypropelene bags of size 30 x 20 cm. To this, 80 ml of water and 4 ml of peanut oil were added and mixed well. The open end of the cover was plugged with cotton and tied with a rubber band. These were autoclaved at 121°C, 15 psi steam pressure for 30 minutes and after cooling, these were kept outside for one day. On the next day, they were autoclaved again at same temperature, pressure, time combination. After cooling the contents to room temperature, these were placed in a inoculation chamber and the mouth of the bag was carefully opened and inoculated with 10 ml of spore suspension (10^6 spores/ml) of respective fungi and mixed thoroughly. Four replications were kept for each substrate. The mouth of the bag was closed after inoculation and incubated at room temperature $(25 + 1^{\circ}C)$ for 14 days. The contents were mixed once in two days for the uniform distribution of inoculum. After 14 days, the contents were mixed thoroughly and one gram of each was taken and shaken thoroughly, homogenized with known quantity of sterile 0.05 per cent tween 80 solution to prepare spore suspension with uniform distribution of spores. The contents were filtered through muslin cloth and the spore count of the suspension was estimated using a haemocytometer (Lomer and Lomer, 1996).

The maximum sporulation of *B. bassiana* was recorded on broken rice, 1.385 x 10^9 spores per g of inoculum, followed by sorghum grains (3.643 x 10^8) and wheat bran (3.12 x 10^8) which were significantly on par (Table 24). Although least sporulation was recorded in rice bran (1.48 x 10^7), it also produced a good spore load. Maximum spore load was recorded in broken rice (2.645 x 10^9 spores per g of inoculum on which was significantly on par with broken maize (2.333 x 10^9 spores). The differences in the substrates with respect to spore yield was statistically significant. Broken sorghum

yielded about $1.958 \ge 10^9$ spores per g of inoculum which was on a par with wheat bran $1.888 \ge 10^9$ spores. Rice bran yielded the least spore load among the treatments (1.54 $\ge 10^8$ per g of inoculum) which was also a good spore yield (Table 25).

| Treatment No. | Substrate | Spore load / g inoculum |
|---------------|----------------|-------------------------|
| 1 | Broken rice | $1.38 \ge 10^7$ |
| 2 | Broken maize | $11.1 \ge 10^7$ |
| 3 | Broken sorghum | 36.43×10^7 |
| 4 | Rice bran | $1.48 \ge 10^7$ |
| 5 | Wheat bran | 31.22×10^7 |
| | F test | ** |
| | | 3.51×10^7 |
| | S.Em <u>+</u> | |
| | C.D. at 5% | $10.56 \ge 10^7$ |

 Table 24. Effect of locally available substrates on the sporulation of B. bassiana

** Significant at P = 0.01

| Table 25. Effect of l | locally available | substrates on | the sporulation | of Verticillium |
|-----------------------|-------------------|---------------|-----------------|-----------------|
| lecanii | | | | |

| Treatment No. | Substrate | Spore load / g inoculum |
|---------------|----------------|-------------------------|
| 1 | Broken rice | $26.45 \ge 10^8$ |
| 2 | Broken maize | 23.33×10^8 |
| 3 | Broken sorghum | 19.58 x 10 ⁸ |
| 4 | Rice bran | $1.54 \ge 10^8$ |
| 5 | Wheat bran | 18.88 x 10 ⁸ |
| | F test | ** |
| | S.Em + | $2.16 \ge 10^8$ |
| | C.D. at 5% | 6.51×10^8 |

** Significant at P = 0.01

Effect of temperature on the growth and sporulation of *B. bassiana*

A storage study was carried out on the unformulated conidia at temperature 22, 25, 30, 35 and 40°C. At each temperature, five replications were kept. The germination percentage was taken at the start of the experiment and repeated at 4 weeks and 8 weeks after storage. The results of influence of temperatures ranging from 20 to 40°C on the growth and sporulation of *B. bassiana* was statistically significant. Then mycelial growth was maximum at 25°C (50.2 mm) followed by 30°C temperature (42.2 mm). At higher temperatures of 35°C the growth was least (28.6 mm). At 40°C there was no growth at all.

Preparation of inoculum

The spores of *B. bassiana* and *V. lecanii* were harvested from the stock culture grown on SDAY media. The spores were suspended in 0.05 per cent aqueous Tween 80. The spore count was estimated using a haemocytometer and the concentration was adjusted approximately to 1×10^7 spores per ml. Equal volume of this suspension was kept in separate beakers.

Evaluation of entomopathogenic fungi against adult B. tabaci

Isolated cultures were tested for their pathogenicity on healthy *B. tabaci* by inoculating fungi onto B. tabaci. The spore suspension of fungus was sprayed onto leaves of cotton seedlings, air dried and the leaves were inserted into surface sterilized plastic cages (8 x 7 cm). The cage was held in place with a twig. Newly emerged uniform aged adults were released into the cage. High humidity was maintained by covering the whole seedling with a plastic cover with few holes. The entomopathogens, B. bassiana and V. lecanii were evaluated against the adult *B. tabaci*. The percentage of mortality of whitefly adults treated with B. bassiana and V. lecanii were 20 and 5.83 per cent, respectively at 24 h of evaluation. But at 72 h the respective mortality percentage were 100 and 88.3. The control recorded a mortality of 10.83 per cent (Table 26). The symptoms of the infection started as external growth of mycelium all around the body of the infected whitefly. Later, the fungus produced vigorous hyphal growth which covered the entire surface of the host body. There was profuse sporulation and fungal growth on the cadavers. For the V. lecanii infection, the mycelium on the insect was white coloured. In the case of B. bassiana infection, the body turned slightly brown coloured and the fungal colony was slightly yellowish.

Table 26. Percentage mortality of *B. tabaci* adults and developmental stages by entomopathogens in the laboratory.

| | Percent mortali | ty/percent infect | ion of <i>B. tabaci</i> |
|-----------------------|---------------------|-------------------|-------------------------|
| B. tabaci stage | B . BASSIANA | V.LECANII | Control |
| Egg | 0.00 | 0.00 | 0.00 |
| 1 st stage | 69.47 | 71.63 | 0.00 |
| 2 nd stage | 66.07 | 61.47 | 0.00 |
| 3 rd stage | 51.28 | 46.29 | 0.00 |
| Adult | 100 | 88.33 | 10.83 |
| | | | |

Evaluation of entomopathogenic fungi against eggs and early nymphal instars

Bioassay on eggs and early instars of *B. tabaci* was carried out in the laboratory by dipping the leaves infested with the respective developmental stage of the insect in the spore suspension (Rowland et al., 1990). The life stages tried by this method were egg, first and third instar nymphs. The eggs were not affected by the entomopathogens. The percentage of mycosed first instar nymphs recorded at 72 hr was 69.47 and 71.63 per cent for *B. bassiana* and *V. lecanii*, respectively and there was growth of the fungal hyphae on the surface of nymphs. The percentage of mycosed second instar was 66.07 and 61.47 for B. bassiana and V. lecanii respectively and the third instar infection of B. bassiana and V. lecanii were 51.28 and 46.29 per cent respectively. However, within 7 days, the fungal growth spread to other nymphs and completely covered them masking the nymphs because of high humidity (Table 26).

Evaluation of entomopathogens on fourth instar nympth of Bemisia tabaci

The bioassay for determining the pathogenicity of entomopathogenic fungi on fourth instar of B. tabaci was done by the method of Landa et al., (1994) with slight modifications. Leaves infested predominantly with early fourth instar nymphs were taken from cotton seedlings maintained in the stock culture of B. tabaci in the glasshouse. These leaves were washed in sterile distilled water and air dried. After being dried, the infested leaves were examined under a stereomicroscope and early fourth instar whitefly nymphs were carefully removed from the leaf surface with a flattened slides, 20 per slide and arranged in two rows of ten. The slides were then placed in plastic petridishes, until they were ready to be used in the bioassay. The entomopathogens viz., B. bassiana and V. lecanii were evaluated against the eggs, first, second and third instars of nymphs of B. tabaci. The mortality was recorded at 72 h after inoculation.

Three FGDI values were considered to be the most critical values during the assay (0.5, 1.5 and 2.5). Each of these values represented the beginning of a growth phase of the fungal life cycle. The FGDI value of 0.5 represented the first sign of viability of the conidia. The colonization of the host with fungus was defined by an FGDI value of 1.5. This phase of the fungal development is irreversible and the infected host does not recover from infection. An FGDI value of 2.5 represented the initial sporulation of the mycelium on the nymphs. Both the entomopathogens, *B. bassiana* and *V. lecanii* were found to the pathogenic on the early fourth stage nymphs of *B. tabaci*. Mean FGDI values of *B. bassiana* and *V. lecanii* at 24 h of evaluation were 0.15 and 0.29 respectively. At 48 h the FGDI value were 1.025 and 1.40 and at 72 h it was 1.525 and 1.060 respectively for *B. bassiana* and *V. lecanii* (Table 27). The FGDI value of *B. bassiana* and *V. lecanii* at 2.5. The result showed that at 72 h both fungi got established on the nymphs which did not recover from infection. For the nymphs treated with control drops (Tween solution 0.05%) there was no infection and the adult eclosed from the pupa leaving the pupal case behind.

| Period after inoculation (h) | | ^a FGDI value | |
|---------------------------------|--------------------------|-------------------------|---------|
| _ | B. bassiana ^d | V. lecanii ^e | Control |
| 24 | 0.15 | 0.29 | 0.00 |
| 48 | 1.03 ^b | 1.40 | 0.00 |
| 72 | 1.53 ^c | 1.60 | 0.00 |
| 96 | 1.93 | 2.30 | 0.00 |
| 120 | 2.28 | 2.50 | 0.00 |

Table 27. Effect of B. bassiana and V. lecanii on early forth instar nymphs ofB. tabaci

^aFungus growth development index and each value is the mean of 40 nymphs

^bGDI value of 0.5 represents the first sign of viability of fungus

^cGDI value of 1.5 represented the colonisaion of the host with the fungus

^dThe % germination of conidia of *B. bassiana* was 85.17

^eThe % germination of conidia of *V. lecanii* was 95

In vitro evaluation of effect of plant protection chemicals on entomopathogenic fungi

The concentration of the chemicals selected for the *in vitro* evaluation against the entomopathogens were chosen on the basis of the dose recommended for field application. Accordingly, the three doses selected were the recommended dose, half of the recommended and a quarter of the recommended dose.

The systemic fungicides inhibited the mycelial growth of the fungi at all three concentrations tested (Table 28). In case of *V. lecanii* complete inhibition was brought about by Iprodione at all the three concentrations. Among the non significant fungicides maximum inhibition was recorded by Captafol and Captan (Table 29). The maximum inhibition was recorded by Triazophos (44.52) and the least inhibition was recorded by Neemark (22.03) (Table 30).

Table 28. Inhibition of mycelial growth of entomopathogens by Systemic fungicides

Per cent inhibition of mycelial growth over control

| Fungicides | Concentration (ppm) | | | | | | | | |
|---------------|---------------------|------------|-------------|------------|-------------|------------|--|--|--|
| | 1 | 000 | 50 | 00 | 2 | 250 | | | |
| | B. bassiana | V. lecanii | B. bassiana | V. lecanii | B. bassiana | V. lecanii | | | |
| Carbendazim | 100 | 100 | 100 | 60.56 | 100 | 42.77 | | | |
| Iprodione | 74.82 | 100 | 54.55 | 100 | 53.85 | 100 | | | |
| Matalaxyl | 51.75 | 100 | 43.71 | 100 | 28.95 | 66.39 | | | |
| Mean | 75.52 | 100 | 66.09 | 86.85 | 60.93 | 69.72 | | | |
| | | | F test | S. EM K | CD 5% | | | | |
| Fungicides | | | * * | 0.65 | 1.80 | | | | |
| Fungicides x | | | * * | 1.13 | 3.12 | | | | |
| Concentration | 1 | | | | | | | | |

Table 29. Inhibition of mycelial growth of entomopathogens by non-systemic fungicides

| Per cent inhibition of mycelial growth over control | | | | | | | | | | | | |
|---|------------------------|------------------------|------------------------|--|--|--|--|--|--|--|--|--|
| Fungicides | | Concentration(ppm) | | | | | | | | | | |
| | 2000 | 1000 | 500 | | | | | | | | | |
| | B. bassiana V. lecanii | B. bassiana V. lecanii | B. bassiana V. lecanii | | | | | | | | | |

| Mancozeb | 60.84 | 100 | 42.66 | 100 | 38.11 | 100 |
|---|----------------------------------|----------------------------------|----------------------------------|----------------------------------|---------------------------------|---------------------------------|
| Wettable Sulphur | 12.94 | 20 | 11.54 | 15.56 | 1.74 | 11.67 |
| Captafol Chlorothalanil Captan Copper Oxy- Chloride | 72.73 39.86 80.07 27.97 | 41.67 52.22 38.89 33.89 | 69.93 31.47 66.78 12.94 | 25.56 25.00 29.44 24.44 | 69.23 24.48 62.59 6.59 | 16.67 11.11 17.22 1.67 |
| Mean | 49.07 | 47.78 | 39.22 | 36.67 | 33.79 | 26.39 |

Table 30. Inhibition of mycelial growth of entomopathogens by insecticides

| | Per cent inhibition of mycelial growth over control | | | | | | | | | | | | |
|----------------------------|---|------------------------|-------------|------------|-------------|------------|--|--|--|--|--|--|--|
| Insecticide | | Concentration (ppm) | | | | | | | | | | | |
| | X | X ^a X/2 X/4 | | | | | | | | | | | |
| | B. bassiana | V. lecanii | B. bassiana | V. lecanii | B. bassiana | V. lecanii | | | | | | | |
| Triazophos ^b | 50.35 | 35.00 | 47.55 | 34.44 | 35.66 | 20.55 | | | | | | | |
| Monocrotophos ^b | 36.36 | 33.89 | 24.03 | 27.78 | 10.84 | 3.33 | | | | | | | |
| Neemark ^c | 22.73 | 21.67 | 24.48 | 15.00 | 18.88 | 11.11 | | | | | | | |
| Mean | 36.48 | 30.19 | 32.02 | 25.74 | 21.79 | 11.66 | | | | | | | |

^a X is recommendation dose

^bX=1000 ppm

^cX =5000 ppm

Field evaluation of mycopesticides against *B. tabaci* the vector of ToLCV in relation to the spread of ToLCVD (nursery protected with nylon net covering)

A randomised block replicated field trial was conducted to examine the effect of mycopesticides on *B. tabaci* and the incidence of ToLCV and tomato yield. The effect of mycopesticides viz., *B. bassiana* and *V. lecanii* were compared with the insecticide Triazophos and untreated control plots on the direct count of whitefly population (Table 31). The data on whitefly populations in the 2nd, 3rd, 5th, 6th, 7th, 9th, 10th and 12th week

after transplanting (WAP) were significantly different. The whitefly population was lowest in the Triazophos treatment (2.5 at 12 WAP to 13.5 at 7 WAP). This trend continued in all the weeks of observations. In all the weeks, the population of whiteflies in mycopesticide treated plot was less compared to untreated plot. Among the mycopesticides, locally mass produced *B. bassiana* at 10^7 spores per ml was most effective in reducing the population of whiteflies compared to control. The number of whiteflies ranged from 21 at 5 WAP to 3 at 12 WAP in *B. bassiana* – 10^7 spores per ml treated plot and whereas in control it ranged from 24 at 6 WAP to 6.50 at 12 WAP (Table 32).

The ToLCV incidence was minimum in the Triazophos treated plot (5% at 2 WAP to 81% at 5 WAP) which was closely followed by *B. bassiana* – 10^7 spores per ml treated plot (6% at 2 WAP to 83% at 5 WAP). The maximum incidence was recorded in the untreated control plot (15% at 2 WAP to 89% at 5 WAP). The symptoms started 2 WAP and reached a peak of 100 per cent six weeks after planting (Table 33). The results show a statistically significant effect of treatment. Maximum yield (18.49 t/ha) was obtained from the plot treated with the insecticide, triazophos which was closely followed by locally produced mycopesticides of *B. bassiana* (17.91 t/ha). The yield recorded in untreated plot was 8.72 tonnes per ha. The yield obtained from Biorin treated plot was 13.76 tonnes per ha. (Table 34).

A mean of 28, 12.9 and 11.7 whiteflies per pole were trapped at heights 25-35, 60-70 and 95-105 cm, respectively, on the days immediately after transplanting the seedlings. This showed that a large migration of whiteflies into the tomato crop occurred even on the day of transplanting which may have resulted on plants becoming infected from the first day. Thereby symptoms would expected to be, and were, expressed in some plants 10-14 days after transplanting. Table 35 shows how the catch of whiteflies changes with time after transplanting.

| Sl. | Treatments | Whitefly population ^a weeks after planting | | | | | | | | | | | |
|------------------|--|---|-----------|-------|-------|-------|-------|-------|-------|-------|-------|-------|------|
| No. | | -1 ^b | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
| 1. | <i>B. bassiana</i> -10^7 spores / ml | 22.50 | 17.50 | 13.00 | 12.00 | 21.00 | 14.50 | 7.50 | 6.50 | 9.00 | 5.50 | 5.50 | 3.00 |
| 2. | Biorin – 3ml / litre | 15.50 | 15.00 | 19.50 | 16.50 | 22.50 | 13.50 | 10.00 | 10.50 | 10.50 | 9.50 | 6.00 | 6.50 |
| 3. | <i>V. lecanii</i> - 10 ⁷ spores / ml | 17.00 | 19.50 | 16.50 | 11.50 | 21.50 | 14.50 | 14.50 | 11.50 | 14.50 | 11.00 | 9.50 | 5.50 |
| 4. | V. lecanii – 10^8 spores / ml | 18.00 | 17.50 | 15.00 | 13.50 | 23.00 | 12.50 | 14.50 | 13.00 | 10.50 | 7.00 | 12.00 | 4.50 |
| 5. | Triazophos – 1.5 ml / litre | 22.50 | 11.50 | 12.50 | 12.00 | 12.00 | 8.00 | 13.50 | 8.50 | 7.50 | 6.00 | 7.00 | 2.50 |
| 6. | Tween solution -0.05% | 23.50 | 20.00 | 21.50 | 15.50 | 22.50 | 20.00 | 21.50 | 17.50 | 12.50 | 13.00 | 8.50 | 5.00 |
| 7. | Control (Untreated) | 20.50 | 20.50 | 20.00 | 18.50 | 23.50 | 24.00 | 18.50 | 17.50 | 16.00 | 12.00 | 7.00 | 6.50 |
| | S.Em + | _ | 0.76 | 1.44 | _ | 0.98 | 0.53 | 1.04 | _ | 1.38 | 1.03 | _ | 0.24 |
| | C.D. at 0.05% | - | 2.64 | 4.98 | - | 3.38 | 1.85 | 3.60 | - | 4.76 | 3.56 | - | 0.84 |
| ^a Eac | C.D. at 0.05% ch figure represents whitefly pop | | for 20 pl | ants | - | 3.38 | 1.85 | 3.60 | - | 4.76 | 3.56 | - | 0 |

| Table 31. | Effect of myco | pesticides on the | whitefly pop | ulation wit | hin the tomato crop |
|-----------|----------------|-------------------|--------------|-------------|---------------------------|
| | | | | | · · · · · · · · · · · · · |

^b Count before spraying Date of sowing: 27-4-97 Date of transplanting: 25-7-97

Six sprays given First spray on 3-6-97

Subsequent two sprays were given at weekly intervals;

Fourth spray was given two weeks after third spray followed by two sprays at three weeks intervals.

| Sl. | <i>Treatments</i> Number of whiteflies ^a weeks after planting | | | | | | | | | | | |
|-----|--|------|------|------|------|------|------|------|------|------|------|------|
| No. | | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
| 1. | <i>B. bassiana</i> -10^7 spores / ml | 6.50 | 5.00 | 4.00 | 5.00 | 4.50 | 1.50 | 4.50 | 3.50 | 2.00 | 2.50 | 2.50 |
| 2. | Biorin – 3ml / litre | 4.50 | 1.50 | 1.50 | 4.50 | 5.00 | 2.50 | 4.00 | 2.00 | 1.50 | 1.50 | 4.00 |
| 3. | <i>V. lecanii</i> - 10 ⁷ spores / ml | 5.00 | 2.50 | 5.50 | 3.50 | 4.50 | 2.50 | 4.00 | 1.00 | 1.00 | 2.50 | 4.50 |
| 4. | <i>V. lecanii</i> – 10^8 spores / ml | 6.50 | 1.50 | 3.00 | 1.50 | 2.00 | 1.00 | 2.50 | 1.00 | 1.50 | 1.50 | 3.50 |
| 5. | Triazophos – 1.5 ml / litre | 2.50 | 1.50 | 3.00 | 1.50 | 5.00 | 1.50 | 2.50 | 1.50 | 2.50 | 0.00 | 1.50 |
| 6. | Tween solution -0.05% | 1.50 | 0.50 | 0.00 | 1.50 | 1.50 | 1.00 | 0.00 | 1.00 | 1.00 | 0.00 | 0.50 |
| 7. | Control (Untreated) | 1.50 | 0.50 | 0.50 | 0.00 | 1.00 | 0.50 | 0.00 | 1.00 | 0.50 | 0.00 | 1.50 |
| | S.Em ± | 1.30 | 0.81 | 1.13 | 1.44 | 1.00 | 0.45 | 0.70 | 0.29 | 0.99 | 0.35 | 0.99 |
| | C.D. at 0.05% | 4.57 | 2.80 | 3.92 | 4.97 | 3.46 | 1.56 | 2.62 | 1.00 | 3.42 | 1.19 | 3.44 |

Table 32. Effect of mycopesticides on the mortality of whiteflies on tomato plants in the field

^a Each figure represents whitefly population for 20 plants
 ^b Six sprays given during the cropping period
 1st spray given one week after transplanting

Date of sowing: 27-4-97 Date of transplanting: 25-7-97

Subsequent two sprays were given at weekly intervals;

Fourth spray was given two weeks after third spray followed by two sprays at three weeks intervals.

| Treatment <u>Per cent ToLCVD inc</u> | | | | | |
|--------------------------------------|---------|---------|-------------|---------|-----|
| | | Weeks | after plant | ting | |
| | 1 | 2 | 3 | 4 | 5 |
| B. bassiana | 5.55 | 30.40 | 71.55 | 99.00 | 100 |
| $(10^7 \text{ spores / ml})$ | (13.44) | (33.44) | (57.44) | (85.65) | |
| B. bassiana | 8.34 | 46.30 | 77.90 | 99.00 | 100 |
| $(10^8 \text{ spores / ml})$ | (16.77) | (42.86) | (61.72) | (85.65) | |
| V. lecanii | 7.30 | 21.80 | 68.75 | 99.00 | 100 |
| $(10^7 \text{ spores / ml})$ | (15.67) | (27.54) | (56.05) | (85.65) | |
| <i>V. lecanii</i> (10^8) | 8.15 | 28.70 | 74.70 | 100.00 | 100 |
| spores / ml) | (16.57) | (32.37) | (59.97) | (89.43) | |
| Triazophos | 3.60 | 39.95 | 73.45 | 97.45 | 100 |
| 1.5 ml/litre | (10.57) | (39.19) | (59.14) | (83.87) | |
| Tween solution | 10.10 | 50.90 | 72.75 | 100.00 | 100 |
| 0.05% | (17.45) | (45.52) | (58.56) | (89.43) | |
| Untreated | 14.45 | 56.20 | 82.45 | 100.00 | 100 |
| | (22.30) | (48.56) | (66.14) | (89.43) | |
| | | | | | |
| C.D. at 0.05% | 10.88 | 9.37 | 3.10 | 12.15 | |
| S.E. m+ | 3.14 | 2.71 | 10.72 | 3.51 | |
| | | | | | |

Table 33. Effect of mycopesticide on the incidence of ToLCVD^a,^c (nursery unprotected)

^aSeedlings were unprotected with nylon net covering nursery site was near the source of inoculum

^bDate of sowing 28.2.97 Date of planting 3.4.97

^cTomato cv. Rashmi

Table 34. Effect of mycopesticides on the yield of tomato cv. Rashmi

| Treatment | Yield / plot | Yield |
|---|------------------|--------|
| | (Kg / 300 sq.ft) | (t/ha) |
| <i>B. bassiana</i> (10^7 spores/ml) | 48.36 | 17.91 |
| Biorin 3ml/litre | 37.16 | 13.76 |
| <i>V. lecanii</i> (10 ⁷ spores/ml) | 33.83 | 12.53 |
| <i>V. lecanii</i> (10 ⁸ spores/ml) | 20.53 | 7.60 |
| Triazophos 1.5 ml/litre | 49.93 | 18.49 |
| Tween solution 0.05% | 34.10 | 12.63 |
| Control (Untreated) | 23.50 | 8.72 |
| Ftest | * | |
| S.Em <u>+</u> | 4.60 | |
| C.D. at 0.05% | 15.90 | |

Date of sowing : 27.4.97; Date of transplanting : 27.5.97 * = Significant at 0.05

| Days after Transplanting | <i>B. tabaci</i> counts on yellow cylindrical trap ^b | | |
|-----------------------------|---|--------|-----|
| 1 0 | Bottom | Middle | Тор |
| | | | |
| 1 | 24 | 13 | 8 |
| 2 | 28 | 9 | 3 |
| 4 | 43 | 12 | 4 |
| 7 | 17 | 8 | 22 |
| 14 | 79 | 23 | 14 |
| 21 | 175 | 92 | 53 |
| 28 | 152 | 85 | 25 |
| 34 | 76 | 65 | 41 |
| 40 | 97 | 42 | 10 |
| 46 | 69 | 23 | 16 |
| 52 | 103 | 79 | 58 |
| 58 | 62 | 31 | 24 |
| 64 | 67 | 34 | 12 |
| 70 | 77 | 39 | 29 |
| 76 | 117 | 66 | 18 |
| 82 | 81 | 46 | 33 |
| 88 | 52 | 21 | 16 |
| 91 | 50 | 19 | 11 |

Table 35. Migration of *B. tabaci* into the tomato crop^a

^a Nursery was covered with nylon nets ^b Whiteflies counted on 3rd and 7th day in each week after transplanting.

Output 4: Simulation model built to determine the potential impact and conditions under which the proposed IPM practices are likely to be most successful

Based on the epidemiological discoveries made during the project, a simulation model was built of the ToLCV pathosystem in southern India. This process helped to clarify the importance of several management parameters that are within our control and thus identify the most potentially useful *B. tabaci*/ToLCV management practices and strategies. The manuscript detailing the model was submitted to *Journal of Applied Ecology* and it is still in press (Annex 1).

Output 5: Report on farmers' perceptions and management practices related to B. tabaci and ToLCV and on socio-economic factors affecting the adoption of new varieties and IPM management strategies.

Study of farmers' perceptions and practices

A study was carried out in the districts around Bangalore, to investigate what farmers knew about ToLCV and how they were coping with the disease. The study consisted of group and individual discussions with farmers, plus a structured questionnaire survey of 174 farmers in 15 locations in 5 districts.

Survey sample design

The following sampling procedure was used.

The survey covered 5 districts of Karnataka adjacent to Bangalore:

- Bangalore Urban
- Bangalore Rural
- Tumkur
- Mandya
- Kolar

This area was divided into 3 geographical areas of roughly equal area, each containing 39 taluks. In each geographical area, 5 taluks were randomly selected from the complete list of taluks, making a total of 15. In each of the selected taluks, one Gran Panchayat was randomly selected. In each of the selected Gran Panchayats, one village was randomly selected.

In each of the selected villages a minimum of 10 farmers were randomly selected from a list of tomato farmers in the village. If there were less than 10 farmers within the village, then additional farmers were selected randomly from the adjacent village to make up the total.

In addition, the following farmers in the village were added:

Women tomato farmers Organic tomato farmers (but none were found) Very large tomato farmers (more than 20 acres)

The final sample is shown in table 36.

| District | Taluk | Gram Panchayat | Village | # farmers | # women farmers | # large farmer | Total |
|--------------------|--------------------------|------------------------|-----------------------|--------------|-----------------------|-------------------|-------|
| Bangalore Urban | Bangalore North | Madanayakan- ahally | Kootlupalya | 5 | 0 | 0 | 5 |
| | | | Kammasandra | 5 | 0 | 0 | 5 |
| | Anekal | Vanakahally | Vanakahally | 9 | 0 | 0 | 9 |
| | | , | Singasandra | 1 | 0 | 0 | 1 |
| Bangalore Rural | Magadi | Ajjanahally | Attimgere | 10 | 2 | 0 | 12 |
| | Channapatna | J.Byadarahally | J.Byadarahally | 10 | 0 | 0 | 10 |
| | Doddaballa- pura | Hosahally | Gundumgere | 7 | 0 | 0 | 7 |
| | - | | Kattehindanah ally | 2 | 0 | 0 | 2 |
| | | | Hosahally | 1 | 0 | 2 | 3 |
| Kolar | Kolar | Vadagur | Vadagur | 11 | 0 | 2 | 13 |
| | Bangarapete | N.G.Hulkooru | Kadirayanakup pe | 1 | | | 1 |
| | | | Hulkooru | 9 | | 2 | 11 |
| | Chikkaballap ura | Peresandra | Hale Peresandra | 6 | 0 | 1 | 7 |
| | | | Peresandra | 7 | 2 | 0 | 9 |
| | Bagepally | Devaragudi- pally | Jilakarapally | 6 | 0 | 0 | 6 |
| | | | Devaragudi- pally | 4 | 0 | 2 | 6 |
| | Chintamani | Upparapete | Rayapally | 12 | 1 | 0 | 13 |
| Tumkur | Tumkur | Doddanaravan gala | Kottanahally | 6 | 0 | 0 | 6 |
| | | | Doddanaravan gala | 4 | 0 | 0 | 4 |
| | Kunigal | Taredakuppe | Sulekuppe | 6 | 0 | 0 | 6 |
| | | | Nagenahally | 3 | 0 | 0 | 3 |
| | | | Taredakuppe | 3 | 2 | 0 | 5 |
| | Chikkanayak ana hally | Jayachamaraj apura | Malagondana- hally | 5 | 0 | 0 | 5 |
| | | | Gyarehally palya | 3 | 0 | 0 | 3 |
| | | | J.C.pura | 2 | 0 | 0 | 2 |
| Mandya | Pandavapura | Manikanahally | Shettahally | 7 | 0 | 0 | 7 |
| | | | Manikanahally | 3 | 0 | 0 | 3 |
| | K.R.Pete | Murukanahally | Hemmana- hally | 10 | 0 | 0 | 10 |
| Total | | | | 158 | 7 | 9 | 174 |

Table 36. Selection of farmers in questionnaire survey

Results

Demographic characteristics

The age of the farmers ranged between 22 and 75 with a mean age of 45.0 years. Only 7 of the total 174 farmers were women. In general, the men are considered to be the main decision-makers on tomato production, although their wives often work on the farm as well, and may, for example, supervise other farm labourers. The women who were considered farmers in their own right, tended to be older women who did not have husbands. Their ages ranged from 40 to 70 with a mean age of 54.

The number of years of formal education was low, with almost one quarter of the farmers having no formal education (Table 37). None of the farmers had secondary education. The average number of years of education was 1.9.

| Years of formal education | | | | | | |
|---------------------------|------------|------------|------------|------------|------------|----------------|
| 0 % (n) | 1 % (n) | 2 % (n) | 3 % (n) | 4 % (n) | 5 % (n) | Total % (n) |
| 23.6 (41) | 23.6 (41) | 14.4 (25) | 26.4 (46) | 2.3 (4) | 9.8 (17) | 100 (174) |

Table 37. Years of formal eduation received by farmers

Farming was the main occupation of all those interviewed, except one who was a government employee. 15 farmers had close family members in other salaried jobs (such as teaching, police, doctor).

Farming system

The majority of farmers had farms of between 2 and 10 acres (median size 5.5, average 8.2) (Table 38).

Table 38. Farm size

| Farm size (acres) | | | | | Total | | |
|-------------------|-----------------|------------------|-------------------|------------------|-------------------|-------------------|-----------|
| <2 % (n) | 2- < 5 % (n) | 5- < 10 % (n) | 10- < 15 % (n) | 15- <20 % (n) | 20- < 50 % (n) | 50- <100 % (n) | |
| 1.2 (2) | 30.5 (53) | 43.7 (76) | 11.5 (20) | 4.0 (7) | 8.6 (15) | 0.6 (1) | 100 (174) |

The farming system is a mixed crop/livestock farming system with tomatoes grown in rotation with other vegetables, legumes or cereals. All farmers except one grew cereals (finger millet (ragi) or rice). Most grew other vegetables or legumes and just over half grew mulberry (Table 39).

| Crop | % farmers growing | Number of farmers |
|----------------------|-------------------|-------------------|
| Cereals | 99.4 | 173 |
| Root crops | 0.6 | 1 |
| Vegetables & legumes | 79.3 | 138 |
| Oil nuts | 52.9 | 100 |
| Mulberry | 52.9 | 92 |
| Spices | 51.7 | 90 |
| Flowers | 19.5 | 34 |
| Grapes | 1.7 | 3 |
| Other fruit trees | 17.8 | 31 |
| Timber | 2.3 | 4 |

Table 39. Other crops grown by farmers

Livestock

The majority of farmers owned livestock, particularly cattle or buffalo (Table 40). Only 4% (7) owned no livestock at all. 83.9% owned at least one pair of oxen. However, total number of livestock was relatively small: no one owned 10 or more buffalo or oxen and only 2 people owned 10 or more cattle. For the small ruminants, the largest herd was 60, and 15.5% (27) owned 20 or more. The largest number of poultry was 50, and 12.1% (21) owned 20 or more. No one owned pigs or donkeys.

Table 40. Farmers owning livestock

| Livestock | % of farmers owning | Number of farmers |
|---------------|---------------------|-------------------|
| Oxen | 83.9 | 146 |
| Cattle | 78.7 | 137 |
| Buffalo | 55.7 | 97 |
| Sheep & goats | 38.5 | 67 |
| Poultry | 20.1 | 35 |

Farm machinery

Apart from a pair of oxen for ploughing, few farmers owned large-scale machinery such as tractors (6.9%) or power tillers (1.1%).

Farm Labour

Almost all the farmers worked on their farms themselves (97.1%). In addition most had access to family labour and hired labour (Table 41).

| Labour | 0 | 1 | 2 | 3-5 | 6-10 |
|-------------|-----------|-----------|-----------|-----------|---------|
| Male family | 35.6 (62) | 20.1 (35) | 24.7 (43) | 17.2 (30) | 2.3 (4) |
| Female | 25.9 (45) | 50.6 (88) | 19.5 (34) | 4.0 (7) | 0 |

| Table 41. | Number of labourers | working on the fa | arm |
|-----------|---------------------|-------------------|-----|
|-----------|---------------------|-------------------|-----|

| | | | Hired labour | | |
|---------------------------|---------|----------|--------------|-----------|-----------|
| No. of labourers | 0 | 1-10 | 11-50 | 51-100 | 101-200 |
| % of farmers (<i>n</i>) | 4.0 (7) | 9.8 (17) | 35.6 (62) | 18.4 (32) | 31.6 (55) |

TOMATO PRODUCTION

Tomatoes are grown throughout the year, with 62.6% farmers growing them in all three seasons, 25.3% growing tomatoes in 2 seasons and 21.1% growing in one season only (often depending on the water availability). More farmers grew tomato in the hot summer and rainy seasons (89.7% and 88.5%) than the cool season (70.1%).

In the hot summer season the majority of farmers use irrigation for their tomato crop. In the rainy and cool seasons, most have a mixture of irrigated and rainfed production (Table 42).

| Table 42. | Percentage of farmers | reporting water sources |
|-----------|-----------------------|-------------------------|
|-----------|-----------------------|-------------------------|

| Water source | Hot season | Rainy season | Cool season |
|---------------------|------------|--------------|-------------|
| Irrigated | 83.9 | 0 | 1.1 |
| Irrigated & Rainfed | 5.7 | 84.5 | 66.1 |
| Rainfed | 0 | 4.0 | 2.9 |
| Not growing | 10.3 | 11.5 | 29.9 |

Tomatoes are grown in rotation with other crops. The most common crops grown before or after tomato are ragi (finger millet), groundnut, other vegetables such as brinjal, onion, bendi, beans, grams, sunflower, rice and mulberry.

Farmers usually leave a gap between tomato crops of at least 4 months (Table 43).

Table 43. Percentage of farmers reporting the number of months between plantingtomato on the same plot

| < 4 months | 4-6 months | 7-12 months | 13-24 months | > 24 months |
|------------|------------|-------------|--------------|-------------|
| 3.4 (6) | 60.1 (87) | 38.5 (6) | 7.5 (13) | 0.6 (1) |

31.4% of the farmers listed tomato as their main source of income. 78% of these were growing hybrids.

Main problems in tomato production

Half the farmers were concerned about low prices or fluctuating farmgate price for tomatoes (Table 44). Prices quoted by farmers ranged from as low as Rs0.27 to Rs12.50 per kilo. The mean price was Rs2.61 (std. 1.89). There was no significant difference between the mean prices of hybrid and local tomatoes.

The problems of water shortage and power were often closely linked. Unreliable power supplies meant that the farmers could not pump water when they needed to water the crop.

Other problems reported were the lack of information or extension activities on tomato production. Transport was a problem for 13% farmers and the difficulty of obtaining good quality seed was reported by 6% farmers.

Table 44. Tomato production problems (other than pests and diseases) reported by farmers

| Problem | % farmers reporting $(n = 174)$ |
|---|---------------------------------|
| Price fluctuations, marketing | 50.0 |
| Lack of information & extension | 26.4 |
| Unreliable power supply | 25.3 |
| Water shortage | 14.4 |
| Transport problem | 13.2 |
| Lack of good quality or certified seeds | 5.7 |
| Labour problem | 1.1 |
| Drainage | 0.6 |
| No problems reported | 25.9 |

Varieties grown

Just over half the farmers (51.1%) grew hybrid tomato varieties. Varieties grown are shown in Table 45. There was a very significant difference in yields between hybrid and non-hybrid growers. The average hybrid yield was 9.94 tonnes per acre (Std 9.22). The average non-hybrid yield was 4.34 tonnes per acre (Std 5.16).

One other major difference in tomato production is that 92.6% hybrid growers used stakes, whereas only 9.8% of other farmers used stakes¹. Staking tomatoes requires a large investment in the cost of the stakes and twine, and in labour requirements to set up and tie the plants. Overall, almost all farmers (96%) had heard of staking, but just over half (54.6%) used them.

| Variety | Type* | | Season | |
|-----------------------|--------|-------------|------------|-------------|
| | | Hot (Besige | Rainy | Cool |
| | | kala) | (Malegala) | (Chaligala) |
| Madanapally, Bile | local | 28.2% (49) | 29.9% (52) | 18.4% (32) |
| madanapally, | | | | |
| Jamoon, Punjab | | | | |
| Chorra | | | | |
| РКМ | local | 4.0% (7) | 2.3% (4) | 2.9% (5) |
| Roma | local | 3.4% (6) | 2.3% (4) | 4.0% (7) |
| Peach Globe | local | 2.9% (5) | 6.9% (12) | 6.9% (12) |
| Sungro & Selections | local | 2.3% (4) | 8.0% (14) | 5.7% (10) |
| Other local | local | 0 | 0.6% (1) | 0 |
| Avinash - II | hybrid | 16.7% (29) | 2.3% (4) | 1.7% (3) |
| Rashmi | hybrid | 13.8% (24) | 13.2% (23) | 11.5% (20) |
| Namdhari, (386, 604) | hybrid | 7.5% (13) | 7.5% (13) | 5.7% (10) |
| Ramya | hybrid | 6.3% (11) | 6.3% (11) | 5.7% (10) |
| Poineer's hybrid | hybrid | 4.6% (8) | 6.9% (12) | 6.9% (12) |
| Rupali | hybrid | 2.9% (5) | 9.2% (16) | 8.6% (15) |
| Mahyco S15, S22, | hybrid | 4.0% (7) | 4.0% (7) | 3.4% (6) |
| S28, S68 | | | | |
| Mahyco Cross B | hybrid | 2.3% (4) | 1.7% (3) | 1.1% (2) |
| Sun seeds (230, 176) | hybrid | 1.7% (3) | 0.6% (1) | 0.6% (1) |
| Other hybrids: Peta - | hybrid | 1.7% (3) | 2.3% (4) | 1.7% (3) |
| 85, Rasika, Kuber | | | | |
| geeta, Naveen, Nath | | | | |
| seeds (summer 15) | | | | |
| No tomatoes grown | | 10.3% (18) | 11.5% (20) | 29.9% (52) |
| Total farmers** | | 100% (174) | 100% (174) | 100% (174) |

Table 45. Percentage of farmers growing different varieties

* Local varieties may come from farmer selections, commercial & research institutes, but are not hybrids and do not require staking

** Farmers may grow more than one variety per season

Pests and diseases

¹ There are a few hybrids which do not require staking.

Pests and diseases reported by farmers are shown in Table 46. All farmers were aware of ToLCV, and listed it as a major problem in the hot summer season, but less so in the cool, dry season. Other diseases reported included leaf spots, wilt, blight, various forms of rots, powdery mildew and damping off. Fruit borers were regarded as the most important insect pest in all seasons and the most difficult to control. Leaf miner was the other insect pest frequently reported. Almost all farmers were unaware of the importance of whiteflies in the spread of ToLCV.

| Hot (Besige kala) | Rainy (Malegala) | Cool (Chaligala) |
|---------------------------------------|--|---------------------------------------|
| ToLCV 100% | Fruit borer 97.4% | Fruit borer 95.1% |
| Fruit borer 88.5% | ToLCV 81.0% | ToLCV 40.2% |
| Leaf miner 26.9% | Leaf spot 29.4% | Wilt 23.0% |
| Wilt 12.8% | Wilt 29.4% | Leaf spot 19.7% |
| Leaf spot 12.2% | Leaf miner 20.8% | Blight 16.4% |
| Blight 12.2% | Blight 15.7% | Leaf miner 13.1% |
| Rots (root, stem, collar, fruit) 4.3% | Rots (root, stem, collar, fruit) 13.8% | Powdery mildew 13.1% |
| Damping off 3.8% | Powdery mildew 12.7% | Rots (root, stem, collar, fruit) 9.0% |
| Other pests 3.8% | Other pests 6.3% | Other pests 6.5% |
| Powdery mildew 1.9% | Damping off 4.5% | Damping off 3.3% |

Table 46. Percentage of farmers reporting main pests and diseases

Pest Management Practices

Application of chemicals was the most commonly-reported pest management practice. Only one farmer said he did not apply chemicals. Farmers view chemicals as very important inputs, and often place the empty pesticide containers on top of the tomato stakes to demonstrate that they have sprayed the field.

There was a significant difference between those growing hybrid varieties and those who do not, in the number of pesticide applications (Table 47). The hybrid growers spray more, averaging between 2 and 2.5 in the nursery and 8 to 8.5 in the main field. Those not growing

hybrids sprayed on average about 0.75 times in the nursery and just over 1.5 times in the main field.

There was no significant differences in the amount of spraying between the different seasons.

| Season | | Number o | of applicati | ons |
|--------|------|----------|--------------|-----|
| | 0 | 1 - 3 | 4 - 5 | > 5 |
| Hot | 23.7 | 66.0 | 9.0 | 1.3 |
| Rainy | 24.0 | 63.6 | 9.7 | 2.6 |
| Cool | 21.3 | 68.9 | 9.0 | 0.8 |

 Table 47a.
 Pesticide applications in the nursery

| Season | Mean | Std. deviation | Minimum | Maximum |
|--------|------|----------------|---------|---------|
| Hot | 1.61 | 1.28 | 0 | 5.5 |
| Rainy | 1.55 | 1.38 | 0 | 8 |
| Cool | 1.54 | 1.21 | 0 | 5.5 |

| | Hot season | | Ra | Rainy season | | | Cool season | | |
|------------------|------------------|------|-----|--------------|------|-----|-------------|------|-----|
| | Mea | Std. | n | Mean | Std. | n | Mean | Std. | n |
| Using | n 2.24 | 1.16 | 88 | 2.29 | 1.38 | 82 | 2.19 | 1.11 | 67 |
| hybrids | | | | | | | | | |
| Not using | 0.79 | 0.90 | 68 | 0.72 | 0.79 | 72 | 0.75 | 0.77 | 55 |
| hybrids | 1.61 | 1.00 | 150 | 1.55 | 1 20 | 154 | 1.54 | 1.01 | 100 |
| Total farmers | 1.61 | 1.28 | 156 | 1.55 | 1.38 | 154 | 1.54 | 1.21 | 122 |

There was a significant difference between the means of hybrid and non-hybrid users (P < 0.000) in all seasons

Table 47b. Pesticide applications in the main field

Percentage of farmers applying number of pesticide applications

| | | Number of applications | | | | | | |
|--------|-----|------------------------|--------|-----------|---------|---------|---------------|--|
| Season | 0 | 1 - 3 | 4 - 5 | 6 - 10 | 11 - 15 | 16 - 20 | > 20 | |
| Hot | 1.3 | 18.6 | 26.3 | 41.7 | 15.4 | 2.6 | 0.6 | |
| Rainy | 0.6 | 19.5 | 31.8 | 37.7 | 5.8 | 3.9 | 0.6 | |
| Cool | 0.8 | 17.2 | 32.8 | 38.5 | 8.2 | 1.6 | 0.8 | |
| | | | | | | | | |
| Season | | Mean | Std. d | leviation | Minimum | Max | kimum | |
| Hot | | 6.43 | 3.99 | | 0 | 22.5 | | |
| Rainy | | 6.06 | 3.94 | | 0 | 22.5 | | |
| Cool | | 6.14 | 3.90 | | 0 | 22.5 | | |
| | | • | | | | | | |
| | | Unt concor | | Dainy | ocon | Coola | 0000 n | |

| | H | ot sease | on | Ra | ainy sea | son | С | ool seas | on |
|---------|------|----------|-----|------|----------|-----|------|----------|-----|
| | Mean | Std. | n | Mean | Std. | n | Mean | Std. | n |
| Using | 8.42 | 4.07 | 88 | 8.07 | 4.38 | 82 | 8.10 | 4.21 | 67 |
| hybrids | | | | | | | | | |
| Not | 3.85 | 1.85 | 68 | 3.76 | 1.22 | 72 | 3.75 | 1.34 | 55 |
| using | | | | | | | | | |
| hybrids | | | | | | | | | |
| Total | 6.43 | 3.99 | 156 | 6.06 | 3.94 | 154 | 6.14 | 3.90 | 122 |
| farmers | | | | | | | | | |

There was a significant difference between the means of hybrid and non-hybrid users (P < 0.000) in all seasons

Chemicals used

Most farmers mixed chemicals together before spraying (92%). The mixture usually consisted of a liquid insecticide and a wettable powder fungicide, but other chemicals could be added such as an additional liquid insecticide, a growth promoter and a foliar fertiliser. The chemicals most commonly used are shown in Table 48.

Herbicide was not used in tomato production. 29.9% of the farmers had heard of weedicide, but only 2.3% had ever tried it.

Over 75% of farmers knew the brand names of the chemicals they were using but it was not clear how many knew much about the active ingredients. For example, a few were using mixtures of similar chemicals (eg. Monocrotophos and Dimethoate).

Most of the insecticides were organo-phosphates. There were some organo-chlorines and carbamates, but relatively few pyrethroids were used. The most popular chemicals were rogar, metacid, ekalux and monocrotophos.

D.M.45 was the most commonly-used fungicide. Bavistin was also popular.

Table 48: Most commonly-used pesticides

| Liquid chemicals | Hot | Rainy | Cool |
|------------------|------------|------------|------------|
| Insecticides | | | |
| Ekalux | 19.2% (30) | 20.8% (32) | 18.0% (22) |
| Endosulfan | 6.4% (10) | 4.5% (7) | 7.4% (9) |
| Metacid | 29.5% (46) | 36.4% (56) | 36.9% (45) |
| Metasystax | 3.2% (5) | 8.4% (13) | 4.9% (6) |
| Monocrotophos | 43.6% (68) | 43.5 (67) | 45.9 (56) |
| (Nuvacron) | | | |
| Dimethoate | 42.9% (67) | 46.1% (71) | 44.3% (54) |
| (Rogar) | | | |
| Sumicidin | 6.4% (10) | 4.5% (7) | 3.3% (4) |
| Growth promotors | | | |
| Spic cytozyme | 7.1% (11) | 4.5% (7) | 4.9% (6) |
| Vipul | 6.4% (10) | 5.8% (9) | 5.7% (7) |
| Multiplex growth | 6.4% (10) | 2.6% (4) | 1.6% (2) |
| promotor | | | |

| Powdered | Hot | Rainy | Cool |
|-----------------|------------|------------|------------|
| chemicals | | | |
| Fungicides | | | |
| Bavistin | 37.8% (59) | 31.8% (49) | 30.3% (37) |
| Captan | 6.4% (10) | 5.8% (9) | 4.1% (5) |
| Copper oxy | 6.4% (10) | 3.9% (6) | 4.1% (5) |
| chloride | | | |
| D.M 45, Indofil | 60.9% (95) | 59.7% (92) | 70.5% (86) |
| M45 | | | |
| Sulphur, sulfex | 7.1% (11) | 7.1% (11) | 5.7% (7) |

Other chemicals used

Insecticides: Cypermethrin, Decis, Fenvalarate, Hostathion (Triazophos), Deltamethrin+Triazophos, Nimbicidin, Demecron, Cymbush, Folidol, Quinolphos, Kelathane, DDT, BHC, Furadan, Carbendizim, *Fungicides:* Blitax, Ridomil, Foltop, Radar, Roko. *Miscellaneous*: Stickers, Streptomycin, Tara oil, Zinc EDTA.

Pesticide application

Only 10 (5.8%) farmers said they owned their own sprayer; the others borrowed or hired equipment. The equipment was either a Gotor or a knapsack sprayer. Gotor sprayers were used by 75.3% of farmers; knapsack sprayers by 33.3% farmers. (9.2% used both). Two farmers had used power sprayers.

Most farmers were involved in applying pesticides themselves (89.7%). 35.6% relied only on family labour, 47.7% hired labour and 17.8% used family and hired labour. Usually several

people were involved in spraying with some mixing the chemicals and holding the hose, while one person sprayed. Men would usually do the spraying whilst both men and women would help in mixing (Table 49). Only 2 people did the pesticide spraying by themselves. Of the 7 women farmers, 5 were involved in pesticide application themselves. On average, between 2 to 3 men plus one woman were involved in pesticide application.

| | Number of labourers | | | | | | |
|-----------------|---------------------|-------|-------|------|------|------|------|
| Labour type | 0 | 1 | 2 | 3 | 4 | 5 | >5 |
| Self | 10.3% | 89.7% | 0 | 0 | 0 | 0 | 0 |
| Family male | 56.3% | 24.7% | 16.1% | 1.7% | 1.1% | 0 | 0 |
| Family female | 77% | 21.3% | 1.7% | 0 | 0 | 0 | 0 |
| Other male | 37.9% | 23.6% | 32.8% | 3.4% | 1.1% | 0.6% | 0.6% |
| Other female | 52.3% | 32.8% | 12.6% | 0.6% | 0.6% | 1.1% | 0 |

 Table 49. Percentage of farmers reporting amount of labour used in pesticide application

Non-chemical pest management

Few non-chemical pest management methods were reported by farmers when asked generally about controlling pests and diseases (Table 50).

Table 50. Other pest management methods reported by farmers

| Other non-chemical sprays (no. of | Other pest control methods (no. of |
|-----------------------------------|---------------------------------------|
| farmers reporting) | farmers reporting) |
| water only (2) | Dusting ash (1) |
| water + urea (2) | Picking boll worm (2) |
| neem oil (2) | Removing infected leaves & fruits (3) |
| water $+$ coconut water (1) | Roguing (2) |
| liquor + DM 45 (fungicide) (1) | Using Pheremone trap (1) |
| | Installing 200W bulb in field to trap |

| insects (1) | |
|-------------|-------------|
| | insects (1) |

Farmers were asked specifically about whether they had heard of, or used, a number of pest management methods (Table 51). About half had heard of using marigolds as a trap crop, and 28% had tried using them. 41% had heard of using neem, and 14% tried it. However, very few had heard of any microbial pesticides such as mycopesticides, viral pesticides or Bt (a bacterial pesticide, *Bacillus thuringinensis*). Use of milk (a traditional remedy) was known of by a few (6%) but not used. Over half had heard of using netting over tomato nursery, but very few had tried it (5%).

| | Control method | | | | | | |
|--------------|----------------|----------------|------|-----|--------------------------------|-----|------|
| % farmers | Netting | Mari- golds | Neem | Bt | Myco- pesticide (Biorin) | NPV | Milk |
| Heard of | 50.6 | 48.3 | 41.5 | 1.1 | 1.7 | 2.3 | 6.3 |
| Used | 4.6 | 28.2 | 13.8 | 0 | 0.6 | 0.6 | 0 |

| Table 51. | Percentage of farmers | s who have heard of | or used pest control | methods |
|-----------|-----------------------|---------------------|----------------------|---------|
| | | | | |

Knowledge of ToLCV

ToLCV was known by a variety of local names, depending on the location and local language of the farmers. All farmers knew of the disease and all claimed to have experienced it on their tomatoes. Most farmers associated the disease with hot weather, and thought that high temperature was the cause of the disease (Table 52). 11.5% of farmers did think that insects were involved in causing or spreading the disease, but only 4 farmers (2.3%) knew that whiteflies were the disease vector. Discussions with groups of farmers revealed that most took little notice of whitefly in the field because they were so small. They were generally regarded as not important; the farmers having no idea that they could spread disease.

| Table 52.Farmers' perceptions of causes and spread of ToLCV | | | | |
|---|-------------------------|--|--|--|
| Caused / spread by: | % of 174 respondents | | | |
| High temperature | 86.2 | | | |
| Waterlogging / very heavy rain | 11.5 | | | |
| Insects (general) | 9.2 | | | |
| Too dry conditions | 5.7 | | | |

| Soil condition | 2.9 |
|----------------|-----|
| Whitefly | 2.3 |
| Virus | 1.7 |
| Do not know | 5.7 |

Note: multiple answers possible

Control of ToLCV

Farmers' stated methods of controlling ToLCV were based on using chemicals (Table 53). However, many farmers were aware that the infected tomato plants did not recover after being sprayed. The farmers were also unspecific about exactly what chemicals should be used and why. Since most farmers are unaware of how ToLCV is spread, only one farmer suggested using netting in the nursery.

Table 53. Farmers' ToLCV controls

| Control measure | % of 174 farmers specifying control |
|-------------------------------|-------------------------------------|
| Use chemical (unspecified) | 39.1 |
| Use insecticide | 32.2 |
| Use insecticide/fungicide mix | 14.9 |

| Water control | 4.6 |
|------------------------|-----|
| Use fungicide | 4.0 |
| Spray urea | 2.3 |
| Rogue diseased plants | 0.6 |
| Use netting in nursery | 0.6 |
| Spray liquor | 0.6 |
| No control | 4.6 |

Sources of information

Other farmers were said to be the main source of information on farming for farmers (Table 54). Over half the farmers obtained information from pesticide dealers particularly on which chemicals to buy. The media was another important source of information. Extension staff were not seen as the main source of information by most farmers.

Table 54. Sources of information on farming and ToLCV

| Source of information | % farmers reporting (n=174) |
|-----------------------|--------------------------------|
| Other farmers | 90.8 |
| Extension staff | 21.8 |
| Pesticide dealers | 54.6 |

| TV, radio, newspapers | 29.9 |
|-----------------------|------|
| Researchers | 3.5 |
| IPM training | 0.6 |
| Other | 4.6 |

On farm trials using netting protected seedling beds in farmers' fields

Farm trials were undertaken in six tomato farmers' fields (Table 55) to check the effect of netting, forty mesh nylon nets were given to the farmers for netting over nursery. At transplanting stage, seedling from the nets were planted seperately with the check. Significant different in yield was noticed between netted and un-netted plots in three farmers fields with an average yield of 5040 kg (netted) over 2784 kg (un-netted), 3946 Kg (netted) over 2053 Kg (un-netted) and 7398.8 Kg (netted) over 4094.6 Kg (un-netted) with net returns of Rs. 3,820/-, Rs. 12,007.5/- and Rs. 7,668/- per acre respectively (Table 56). A farmer used netting over nursery and Avinash-II as resistant variety got net returns of Rs. 24,487.5/- per acre.

| Name of the farmer* | Taluk | Village | Variety grown |
|---------------------|-----------------|--------------------|--------------------|
| Narasappa | Dodda-ballapur | Kattehindana-hally | Local: Madanapally |
| S/oNarasimhaiah | | | |
| Avalakondarayappa | Chikka-ballapur | Peresandra | Hybrid: |
| S/o Pillana | | | Rashmi |
| Srinivas | Chintamani | Rayapalli | Hybrid: Avinash-2 |
| S/o Sriram Reddy | | | |
| Ramakrishna Reddy | Chintamani | Rayapalli | Hybrid: Nandhini |
| S/o Rama Reddy | | | |
| Fayoz Sab | Chintamani | Rayapalli | Hybrid: |
| S/o Aziz Sab | | | Ramya |
| G. Narayanappa | Mulbagal | Seegenahally | Hybrid: Nandhini |
| S/o Gangappa | | - | |
| (Organic farmer) | | | |

Table 55. Farm trial on the management of ToLCV in farmers fields

* Nylon netting over nursery Farmers selection:

1. Low number of sprays (<5), Local varieties

2. Medium number of sprays (5-10), Hybrid varieties

3. High number of sprays (>15), Hybrid varieties

| Name of the Farmer | Area/ variety | Yield (Kg) | | Total yield (Kg) | Payment | Total expenditure | Net returns | Remarks |
|--------------------------|--------------------------|------------|-----------|------------------------|---------|----------------------|-------------|-------------------|
| | | Netted | Un-netted | | Rs. | Rs. | Rs. | |
| 1. Narasappa | 0.5 acre | 1260 | 696 | 1956.0 | 6885 | 4975.0 | 1910.0 | ToLCV, |
| | Madanapally May 1998 | (5040) | (2784) | (3912) | | | (3820.0) | 4% N, >20% UN |
| 2. Avalakonda rayappa | 0.4 acre | 740 | 385 | 1125.0 | 9770 | 4967.0 | 4803.0 | ToLCV, |
| | Rashmi Feb. 1998 | (3946) | (2053) | (2812) | | | (12007.5) | 20% N 40% UN |
| 3. Ramakrishna Reddy | 0.5 acre | 1849.7 | 1023.6 | 2873.3 | 12858.5 | 9023.8 | 3840.0 | ToLCV, |
| | Nandhini April 1998 | (7398) | (4094) | (5746) | | | (7668) | 13% N, >30% UN |
| 4. Srinivas | 0.5 acre | | | 12000 | 25000 | 12756.25 | 12243.75 | ToLCV, |
| | Avinash II April 1998 | | | (24000) | | | (24487.5) | 2% N, 13% UN |
| 5. Narayanappa | 0.5 acre | | | 3750 | 12500 | 9603.75 | 2896.25 | ToLCV, |
| | Nandhini May | | | (7500) | | | (5792.5) | 12% N |
| | 1998 | | | | | | | 36% UN |
| 6. Fayaz sab | 0.5 acre Ramya | | | 1392 | 4270 | 6268.50 | -1998.50 | ToLCV, |
| | June 1998 | | | (2784) | | | (-3997.0) | 14% N 18%UN |

Table 56. Netting trial: farm budget information

Figures in parenthesis indicates value for an acre; N = Netted; UN= Unnetted

Comments provided by tomato farmers about the tomato genotypes being bred by the project during the Farmer-field day/Workshop on the Sustainable management of *B. tabaci* and Tomato Leaf Curl Geminivirus Disease (26th February 1999).

| 1. Mr. Srinivasa Reddy, Rayapalli, Chintamani Taluk: | -Disease resistant varieties are good. -March-June (summer season) is ideal for testing. If they remain resistant during these months then these varieties can be used as resistant source. -Fruit size has to be increased. Fruit should be round with thick skin. -Used nylon net - gave good result, 30% less disease incidence than unnetted |
|--|---|
| 2. Mr. Abdul Latif Sab, Rayapalli, Chintamani Taluk | Size of the fruits are under size, but one or two are correct size. These genetic lines should be grown in summer. In this season ToLCV is not serious in any of the genetic lines. Netting was a good experiment. Netting experiment gave higher yield compared to non netting experiments. |
| 3. Mr. Devendra Reddy, Rayapalli, Chintamani Taluk | Size and skin of tomato fruits are not ideal Thick skin / rind is required Like Avinash-2, skin should be thick and yield should be like Avinash (30.50 t/ha). December-January planted lines / hybrids will have less incidence. |
| Mr. Laxminarayana Reddy, Peresandra, Chikkaballapur Taluk. | -In this season diseases are less -Conduct trials in summer season -TLB-128 & TLB-129 are good lines -If length is reduced and tips are eliminated and changed to a round shape TLB-128 & TLB-129 will be the best variety. -Netting over nursery is very much useful, using this increased yield up to 30% compared to unnetted one. |

5. Mr. Chinnappa,

Haleperesandra & Chikaaballapur Taluk: -From ancient days we have 4 seasons, (1) Chaligala (2) Galigala (3) Bisilugala (4) Malegala.

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-In Chaligala we harvest very good crop -In Bisilugala, Galigala and Malegala we lose 70% of the crop

-If you develop an adaptable technology for the farmers suitable for the above seasons, it would be appreciated. It would also be appreciated if you could conduct trials, demonstration and seminars in the village rather than in the city.

-ToLCV appears in the crop 15-20 days after transplanting when plants start luxurious growth.

-Putting nylon net to the nursery is good.

-Only 2-3 lines are good.

- -Size of the fruit should be increased.
- -To be tested in summer.
- -Yield loss was up to 50% in cv. Rashmi.
- -Fruits are small in size, should be increased.
- -Only few lines are good.

-Fruits are small in size, should be increased.-Only few lines are good.

- -Fruits are small in size.
- -Crops should be grown in summer and please release the variety which will show resistance to ToLCV.
- -Nylon netting trial is good, it gave higher yield.

-Netting has increasing yield by 20% and also disease incidence is reduced. -3 to 4 lines are good.

- Mr. Dinesh Kumar: Peresandra, Chikkaballapur Taluk.
- Mr. Ramakrishnappa, Kattehindanahally, Doddaballapur Taluk.
- 8. Mr. Narasimhamurthy, Kattehindanahally, Doddaballapur Taluk.
- Mr. Nagaraja, Kattehindanahally, Doddaballapur Taluk.
- 10. Mr. Krishnamurthy, Peresandra, Chikkaballapur Taluk
- Mr. Gopinath Seegenahally, Mulbagal Taluk.
- 12. Mr. G. Narayanappa,

-He has grown Roopali, Rashmi, Namadhari

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Seegenahally, Mulbagal Taluk. varieties & he is now growing local variety. -Netting is useful.

13. Mr. M. Narayanappa, Seegenahally, Mulbagal Taluk. -Not spoken.

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Output 6: Recommendations for pest management practices, developed through farmer participation, for improved tomato production through control of B. tabaci and ToLCV.

INFORMATION AND RECOMMENDATIONS FOR TOMATO LEAF CURL DISEASE MANAGEMENT

What is Tomato Leaf Curl Disease?

- This disease may be called several different local names: malle roga, bindi roga,
- It is a very destructive disease of tomato
- It causes severe yield loss
- Once infected, tomato plants cannot recover

What does Tomato Leaf Curl Disease look like?

[picture of infected plant]

- Plants have curling and small leaves
- Younger leaves may be pale yellow in colour
- Older leaves may look bronze or purple in colour
- Plants infected when young remain stunted and do not grow tall or bear fruit

How do tomato plants become sick with Tomato Leaf Curl disease?

[picture of whiteflies on leaves. Diagram of spread from one plant to another]

- Tomato Leaf Curl Disease is caused by a geminivirus
- Tiny insects called whiteflies feed on plants that are already sick with Tomato Leaf Curl Disease and pick up the virus.
- The whiteflies carry the virus to healthy plants that they feed on
- The healthy plants then become sick with Tomato Leaf Curl Disease.
- The whiteflies spread the disease between plants in a similar way to mosquitoes spreading malaria between people by biting them.
- Tomato Leaf Curl Disease is only spread by whitefly. It is **not** spread by other insects.
- Tomato Leaf Curl Disease is **not** spread through the soil or water or wind or seed.

- Tomato Leaf Curl Disease is more serious in the summer months when the weather is hot
- Tomato Leaf Curl Disease affects some tomato varieties more than others. A few varieties are said to be "resistant to Tomato Leaf Curl Disease" because they do not often become badly infected with the disease.

How can Tomato Leaf Curl Disease be prevented?

Tomato Leaf Curl Disease is difficult to control. But there are things that the farmer can do to prevent the disease.

✤ Chose a tomato variety that resists infection.

Examples are Avinash II,

Check with the Department of Horticulture for the latest information on tomato varieties

✤ Prevent early infection

Plants which are infected early will be stunted with few fruits. Plants which are infected late, will stay produce some fruit. Therefore it is important to protect tomato plants when young.

IN THE NURSERY [PICTURE OF NYLON NET]

□ Use a nylon net to cover the seedbed and stop whitefly from reaching the seedlings

- Use a small (40) mesh nylon net.
- Make a frame to support the net.
- Cover the seedbed completely, making sure there are no gaps where the whitefly could enter
- Make sure there are no holes in the net.
- Do not raise the sides of the net at any time to prevent the whitefly reaching the seedlings

IN THE MAIN FIELD

□ USE OF INSECTICIDES

No chemical can cure a plant once it is sick with Tomato Leaf Curl Disease. However, spraying with insecticide can kill the whitefly and therefore delay the spread of the disease from one plant to another.

- Spray within 3 days after transplanting with an approved insecticide such as Confidor (or Hostathion).
- Alternatives to conventional insecticides are Neem, Neem extract (Neemark) and mycopesticides such as Biorin
- Spray again 8 days after transplanting.

Note:

• Fungicides (for example Dithane) will have no effect on Tomato Leaf Curl Disease.

• Spraying late in the season (after 35 days after transplanting) will have no effect on Tomato Leaf Curl Disease

Ouput 7: Findings of 1 - 5, above, with resulting IPM recommendations published as a handbook(s)/poster(s) and peer-reviewed scientific articles accessible to researchers, extension workers and farmers

The above recommendations have been printed in Kannada (the language of Karnataka) as a leaflet and these were handed out at the Workshop (copy attached). At the time this FTR was written, the English version had not been printed.

See also section 3.

Additional funding 1996/97: Provision of training in PCR techniques to Dr Muniyappa, UASB at NRI and project staff at UASB in collection and identification of natural enemies of Bemisia tabaci

Dr Muniyappa received training in PCR at NRI which provided him with the background and information to set up a molecular genetics laboratory at the UASB. This is now in operation and PCR reactions are currently being carried out. Please see additional funding 1998/99 below.

Project staff were trained in the collection and identification of natural enemies by Dr Polaszek during a 5 day visit to the UASB. This enabled the beneficial insect component of the project to be carried out successfully.

Additional funding 1998/99: Travel and subsistence costs for Dr Colvin and Dr Muniyappa to give presentations at the 2nd International Workshop on Bemisia and Geminiviral Diseases, San Juan, Puerto Rico, 7-12 June 1998

Drs Colvin & Muniyappa travelled to the 2nd International Workshop on Bemisia and Geminiviral Diseases, San Juan, Puerto Rico, 7-12 June 1998 where the work of the project was presented. Please see section 3.

Additional funding 1998/99: Training for project staff at UASB in PCR methodologies and techniques for assessing seasonal ToLCV inoculum pressure; also funding for Dr Colvin to visit to UASB to organise and run a workshop/farmer field day for local tomato farmers, extension workers and researchers

Dr Richard Thwaites travelled to Bangalore and spent 7 days training UASB staff (Mr H.M.Venkatesh and Dr H.A. Prameela) in PCR methodologies and techniques for assessing seasonal ToLCV inoculum pressure. The visit was highly successful, as ToLCV was detected both in extractions from plant material and from individual adult *B. tabaci*. This will enable a considerable quantity of new work to be attempted and will potentially greatly increase our understanding of the ToLCV pathosystem with consequent benefits in disease management.

A workshop was organised and held on the 26th and 27th of February 1999.

Contribution of Outputs

Output 1: (at least one ToLCV/B. tabaci-resistant tomato variety assessed for use in IPM)

In India, *B. tabaci* is an extremely important pest both in its own right and as a vector of plant viruses. More than ten ToLCV-resistant genotypes have been produced through the project's screening and breeding programme that have varying degrees of resistance to the vector, *B. tabaci*. This resistance to ToLCV is an extremely important component of the IPM recommendations for tomato production because it means that the large number of insecticide sprays required to obtain any yield from a ToLCV-susceptible crop can be reduced greatly.

The objectives for this output have been exceeded as at least three of these tomato genotypes were commented on very favourably by local farmers, indicating the great potential of these inbred tomato lines in terms of their developmental impact. This output, therefore, has contributed directly to the project goal and production system purpose.

Output 2: Systematics of B. tabaci parasitoids and predators (beneficial insects) in Karnataka determined and their potential for reducing B. tabaci populations and ToLCV incidence assessed

B. tabaci parasitoids and predators (beneficial insects) in Karnataka were collected, identified and mass produced for field trials. We discovered during the course of the field trials that the numbers of immature *B. tabaci* developing on the tomato crop was relatively low. This is in direct contrast to the situation in other parts of the world were the B-biotype of B. *tabaci* reaches very high numbers on tomato. The beneficial insects which feed and develop on the immature stages of *B. tabaci* had a limited effect on the *B. tabaci* population within the tomato crop but there was almost no effect on the spread of ToLCV. We discovered that this is because almost all the spread of ToLCV into a susceptible tomato crop is caused by viruliferous immigrant adult *B. tabaci*. This has both increased our understanding of the pathosystem and aided the formulation IPM recommendations that are likely to be sustainable.

Augmentation of beneficial insects did produce a small but significant increase in the final tomato yield which was achieved without applying any chemical sprays. This result is important in that it demonstrates the value of preserving the beneficial insect complex within the crop as they also prevented any other major insect pests such as *Helicoverpa armigera* from causing damage. This output, therefore, has also contributed considerably to the project goal and production system purpose.

Output 3: Indian fungal isolates assessed and compared with existing mycopesticides against *B. tabaci*

Several entomopathogenic fungal isolates were collected, mass produced and used in field trials. *B. bassiana* caused significant mortality of nymphs and adult *B. tabaci* within the tomato crop. The yield obtained for the best mycopesticide treatment was slightly less but not significantly different to the conventional insecticide treatment Triazophos. The yield obtained for the *V. lecanii* treatments were significantly lower. These results show that the use of *B. bassiana* as a mycopesticide could potentially be useful in a tomato IPM programme. However, there are several difficulties to be overcome before a *B. bassiana* mycopesticide could be recommended. Some of these are that: it acts like a broad spectrum

insecticide and has a detrimental effect on the beneficial insect population; its widespread use would probably affect the local silk industry; its efficacy would be reduced by fungicides which are often sprayed by farmers; its efficacy is currently not noticably better than a conventional insecticide which may impede its adoption by local farmers.

One of the aims of this project was to try out most of the possible IPM technologies that would be suitable for the ToLCV/*B. tabaci* pathosystem. As such, this work has greatly improved our understanding of the potential for using mycopesticides in this system and therefore has contributed to the project goal and production system purpose. Some additional IPM components have yet to be evaluated and this will form one of the proposed activities in the next phase of the project.

Output 4: Simulation model built to determine the potential impact and conditions under which the proposed IPM practices are likely to be most successful

This output has been extremely useful in bringing together systematically all the information gained from the other project activities and integrating them in order to identify the IPM technologies that have the highest chance of having a sustainable impact. As such, it has aided our understanding of the mechanism driving the annual epidemics of ToLCV and suggested ways in which IPM technologies might be improved.

Output 5: Report on farmers' perceptions and management practices related to B. tabaci and ToLCV and on socio-economic factors affecting the adoption of new varieties and IPM management strategies.

A greatly increased understanding of farmers' current perceptions and management practices related to *B. tabaci* and ToLCV has been obtained. In addition, the socio-economic factors affecting the adoption of new varieties and IPM management strategies have been identified through detailed feedback from farmers. The information that makes up this output, therefore, will greatly facilitate the adoption of improved ToLCV/*B. tabaci* management practices and has laid the foundations for the future additional successes of the project. This output, therefore, has clearly contributed to the project goal and production system purpose.

Output 6: Recommendations for pest management practices, developed through farmer participation, for improved tomato production through control of B. tabaci and ToLCV.

A package of pest management recommendations, developed through farmer participation, for improved tomato production through control of *B. tabaci* and ToLCV has been formulated. This information has started to be disseminated to farmers (see output 7) and is already having an impact. This output, therefore, continues to contribute directly to the project goal and production system purpose.

Ouput 7: Findings of 1 - 5, above, with resulting IPM recommendations published as a handbook(s)/poster(s) and peer-reviewed scientific articles accessible to researchers, extension workers and farmers

An information leaflet containing the IPM recommendations has been published in Kannada, the local language of Karnataka State, South India, and has been given to researchers,

extension workers and farmers. This leaflet continues to be freely available and therefore continues to contribute directly to the project goal and production system purpose and generate developmental impact.

Promotion pathways and follow up action/research to promote the findings of the work to achieve their development benefit

The project's IPM recommendations have been given to the University of Agricultural Sciences' Directorate of Extension and the State Horticultural Department for dissemination to farmers. In the next phase of the project it is intended that the ToLCV resistant genotypes will undergo multilocation and farm trials. If these are successful, these genotypes will be officially released for general cultivation in the state of Karnataka. It is highly probable that these genotypes could also be grown in other parts of India.

The use of these genotypes will accrue significant advantages to resource poor farmers as, unlike hybrid varieties, they are true breeding and seeds can be collected for use in the following season. Further benefits include cost savings from the reductions in pesticide use, improvements to farmers' health, reduction in environmental pollution and pesticide residues in tomato based products.

An ability to grow tomatoes successfully in the ToLCV epidemic season will also increase the farmers' income as tomatoes are an important source of revenue and hence, it will reduce poverty.

Publications and plans for further dissemination

Published:

RAMAPPA, H.K., MUNIYAPPA, V. & COLVIN, J. (1998) The contribution of tomato and alternative host plants to tomato leaf curl virus inoculum pressure in different areas of south India. *Annals of Applied Biology*, **133**: 187-198.

In press:

HOLT, J., COLVIN, J. & MUNIYAPPA, V. (1999) Identifying control strategies for tomato leaf curl virus disease using an epidemiological model. *Journal of Applied Ecology* (in press). (A)

In preparation:

- Anita Cherian K, Muniyappa V., Colvin J. & Moore D. (In prep.) Laboratory evaluation of Indian isolates of *Beauveria bassiana* (Balls) Vvill and *Verticillium lecanii* (Zimm) Viegas against the life stages of *Bemisa tabaci* (Genn.), the vector of geminiviruses.
- Anita Cherian K, Muniyappa V., Colvin J. & Moore D. (In prep.) Evaluation of culture media and locally available substrates for the growth and sporulation of Indian isolates of Beauveria bassiana (Balls) Vvill and Verticillium lecanii (Zimm) Viegas.
- Venkatesh, H.M. (*In prep.*) The use of beneficial insects and other management methods for the control of tomato leaf curl virus and *Bemisia tabaci*. PhD thesis, University of Agricultural Sciences, Bangalore, India. In prep.

Planned:

In the previous three years the project has far exceeded the original expectations and agreed objectives in the amount and quality of experimental research data produced. Dissemination funding will therefore be required in order to enable the following publications to be prepared to a standard that will allow publication in international peer-reviewed journals.

- 1. Anita Cherian K, Muniyappa V., Colvin J. & Moore D. Field trials using mass produced entomopathogenic fungi against B. tabaci and ToLCVD in southern India.
- 2. Anita Cherian K, Muniyappa V., Colvin J. & Moore D. A laboratory study of the effect of fungicides on entomopathogenic fungi.
- 3. Warburton, H., Muniyappa, V., Nagaraju, N., Venkatesh, H.M & Colvin, J. Farmers' perceptions and coping strategies for ToLCV management in southern India.
- 4. Venkatesh, H.M, Colvin J & Muniyappa V. The effect of beneficial insect augmentation on the *B. tabaci* population and ToLCV spread in tomato fields in southern India.
- 5. Venkatesh, H.M, Muniyappa V., Anita Cherian & Colvin J. A laboratory study of the effect of entomopathogenic fungi on beneficial insects in southern India.
- 6. V. Muniyappa, H.M.Venkatesh, M.N. Maruthi, A.S. Padmaja; Hanson, P., S.K. Green & J. Colvin. Screening for resistance in tomato to ToLCV in southern India.
- 7. Etc.

Internal reports:

- 1. Quarterly reports submitted to NRInt. each quarter.
- 2. Annual reports submitted to NRInt. at the end of each financial year.
- 3. Back-to-office visit reports copied to NRInt.

Other dissemination of results, training etc:

Theses:

- CHERIAN, A.K. (1998) Management of tomato leaf curl virus in tomato through the control of its whitefly vector, *Bemisia tabaci* Genn., by entomopathogic fungi. PhD thesis, University of Agricultural Sciences, Bangalore, India.
- MARUTHI, M.N. (1998) Molecular methods of detection and screening tomato genotypes to tomato leaf curl virus and *Bemisia tabaci*. M.Sc. thesis, University of Agricultural Sciences, Bangalore, India.
- GOVINDAPPA, M.R. (1998) Pathogenicity of *Paecilomyces farinosus* against *Bemisia tabaci* (Genn.) the vector of tomato leaf curl virus. M.Sc. thesis, University of Agricultural Sciences, Bangalore, India.

Conference presentations:

- MOORE, D., LUKE, B.M. & THOMPSON, E.C. (1997) Preliminary assessments of entomopathogenic fungi for the control of *Bemisia tabaci* in India. Abstract of poster presented at The Society of Invertebrate Pathologists Meeting, Banff, 24-29 August 1997. (B)
- MUNIYAPPA, V., CHERIAN, A.K., VENKATESH, H.M., NAGARAJU, N., MARUTHI, M.N., CZOSNEK, H., GREEN, S.K. & COLVIN, J. (1998) Tomato leaf curl virus in southern India and its sustainable management presented in 2nd International Workshop on Bemisia and Geminiviral Diseases. 7-12 June 1998, San Juan, Puerto Rico.

- WARBURTON, H., MUNIYAPPA, V., NAGARUJU, N., PALIS, F.L & VILLEREAL, S. Plant virus diseases: farmers perceptions and coping strategies. Poster and abstract in Proceedings of 7th International Congress of Plant Pathology, 9-16 Aug., 1998 Edinburgh, UK.
- COLVIN, J., MUNIYAPPA, V., RAMAPPA, H.K., CHERIAN, A.K., VENKATESH, H.M., NAGARAJU, N., MARUTHI, M.N. & S.K. GREEN. The epidemiology and management of tomato leaf curl virus and Bemisia tabaci in southern India. VIIth International Plant Virus Epidemiology Symposium, Aguadulce, Spain, April, 11-16, 1999.

For projects aimed at developing a device, material or process specify:

a. What further market studies need to be done?

The socio-economic survey carried out in the first phase of the project clearly identified the demand for resistant varieties by tomato farmers throughout Karnataka. It is not envisaged that a further market study needs to be done.

b. How the outputs will be made available to intended users?

The development of the project's ToLCV-resistant tomato genotypes with horticulturally acceptable qualities now requires multilocation testing and trials in farmers' fields before they can be released on a large scale. This requires at least a further two years of trials to be conducted in the peak ToLCV epidemic season of the year as well as the approval of the State Horticultural Department and the University of Agricultural Sciences' Extension Service which will be forthcoming provided the varieties perform well in the trials.

c. What further stages will be needed to develop, test and establish manufacture of a product?

See above.

d. How and by whom, will the further stages be carried out and paid for?

A second phase of the project is necessary because it has been successful in identifying potentially extremely useful IPM measures and technologies that have the potential to impact greatly on the *B. tabaci*/ToLCV problem in India. The second phase will be carried out by the major stakeholders that were involved in the first phase as well as the State Horticultural Department and the University of Agricultural Sciences' Extension Service. A proposal has been prepared and submitted to the DFID Crop Protection Programme to fund this work.