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"Integrated Pest Management in Processing Tomato Fields in Ethiopia"

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Executive Summary

The potato tuber moth (PTM), *Phthorimaea operculella* (Zeller) has recently became an important tomato fruit pest in Ethiopia. It is unclear why tomato, reported earlier as a least preferred host, emerged as a major host. Cultivars of tomatoes were reported to show differential levels of resistance to the attacks by the PTM. Possible reasons for the increased importance of PTM on tomatoes may be (a) the evolution of PTM populations specifically adapted to feed on tomatoes; (b) the introduction of susceptible tomato cultivars; (c) natural enemies of the PTM might have been prevented from searching and attacking PTM larvae in tomato fields.

The evolution of a PTM race specialized to deal with tomato was ruled out (see the 1999/2000 annual report). To determine the observed susceptibility of tomatoes to the PTM and the subsequent information obtained on the presence of variation in the susceptibility of tomatoes to the PTM the following studies have been carried out: field monitoring using sex pheromone baited traps and direct counting of infested leaves and fruits per plant, the latter done for three classes of fruits (immature green, maturing green and ripening); study of the interactions of the PTM with trichomes of the three tomato cultivars; determination of survival, growth and development of PTM larvae on tomato vs. potato foliage and among the fruits of the three tomato cultivars; and determination of the parasitism level on PTM larvae in the foliage of the three tomatoes and a potato cultivar. These studies were carried out using laboratory and field facilities of the Melkassa research center of the Ethiopian Agricultural research organization (EARO).

Highlights of the results obtained include the following: The leaves of the cherry cultivar provided better resource for maximum PTM larvae survival at preblossom stage of the crops than the leaves of the processing, fresh-market cultivars and potato. Conversely, the fruits of the fresh-market cultivar provided the best resources for PTM larvae survival, growth and development. Natural parasitism of PTM larvae in tomato fields was extremely low, whereas in potato fields, an average of 15% larvae parasitism was recorded. The field cage experiment results were consistent with the production field results and a mean of 23.18% parasitism was obtained in potato.

General Background

Adhanom and Tessema (1984) studied the host preference of ovipositing potato tuber moth (PTM), *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae) under field and controlled conditions at Melkassa (8°26'N39°21'E) and Awassa (7°05'N38°29'E), in Ethiopia. They showed that *Datura stramonium* L., *Nicotiana tabacum* L. and *Solanum melongena* were superior hosts than tomato. Varela and Bernays (1988) reported a similar trend. In Ethiopia, in less than a decade after the report of Adhanom and Tessema (1984), the magnitude of attack on tomato fruits by the PTM accounted over 94% of the fruitworm damage in Melkassa (Tsedeke and Gashawbeza 1994). In the upper Awash Agro Industry, in Merti and Nura Era farms, in 1997, fruits from tomatoes planted on 60 ha, which had cash value of about US$ 900 000, were totally discarded due to PTM damage. The possible reasons for the increased importance of the PTM on tomatoes include the following: (a) A PTM population specifically adapted to feed on tomatoes might have evolved and (b) susceptible tomato cultivars might have been introduced into the tomato production system and (c) natural enemies of the PTM might have been prevented from searching and attacking PTM larvae in tomato fields. Traynier (1975) suggested that PTM populations that infest potato crops may be behaviorally different from those infesting tomato crops and he proposed that local races of the PTM that have adapted to feed on tomato might have developed.

PTM has shown differential reaction to tomato cultivars (Tsedeke and Gashawbeza, 1997). This indicates that there might have been present biochemical (allelochemicals) and/or morphological factors (Smith et al. 1994 & Thomas and Waage, 1996) in the resistant tomato cultivars. These factors might be present in the leaves, flowers and/or fruits of the PTM resistant tomatoes, which the PTM use as its niches and hence discouraged the PTM from successfully colonizing them. Therefore, it is thought important to investigate and elucidate the resistance factors, morphological and/or biochemical in the resistant tomato cultivars. Considering the feeding niche of the PTM, morphological factors such as trichomes, may be involved in discouraging oviposition and settlement of colonizing larvae. Allelochemical factors contained in the tomatoes leaf lamella and fruits may be involved in reducing survival, growth and development of colonized larvae. Both factors, in combination or separately, might have contributed to the observed differences in the amount of fruit damage sustained by cv. Ceres (cherry type, resistant to PTM), cv. Serio (Processing type, resistant to PTM) and cv. Marglobe (Fresh-market type, susceptible to PTM). Beside their direct effects on the PTM, tomato allelochemicals and morphological traits may also interfere with the activities of the natural enemies of the PTM in tomato plots. Therefore, investigations on the interaction of tomato hosts and PTM and tomato host, PTM and its natural enemies have been conducted.

Section 1A. Research objectives during this reporting period:

1. Determining the population dynamics of PTM on three cultivated tomatoes and a potato cultivar.
2. Quantifying the impact of leaf trichomes on PTM in three tomato cultivars.
3. Assessing, through feeding bioassay, the biological performance of the PTM on tomato and potato leaves and tomato fruits.
4. Assessing the PTM natural enemies situation in tomatoes viz. potato and determine the natural parasitism of PTM in the two crops.

Section 1B. Research Accomplishments

Field and laboratory studies in Melkassa: Progress report (2000/2001 Tomato crop production season).

1.Population dynamics of the PTM in tomato and potato

1.1. Sex Pheromone trap Monitoring of PTM

1.1.1. Materials and Methods

Sex pheromone baited traps were set in potato and tomato plots after the transplanted tomatoes were established and potato plants were successfully emerged. There was installed one such trap in a plot area of 260 m^2 at 40 cm height. There were three tomato cultivars each representing the three cultivated tomato types: Cherry, Processing and fresh-market. The potato was included as the standard PTM susceptible host. Each tomato and the potato were planted in four plots. The trapped PTM males were collected everyday and recorded. This was continued for several days after the last tomato fruits picking. For the sake of presentation the data were pooled to weeks and the statistical analysis made on the pooled data on weekly bases.

1.1.2. Results

The results in figure 1 show that potato is the preferred host by the PTM. The difference was confirmed to be very high ($p<0.0001$). On the other hand the difference in the number of visiting PTM adults among the tomato cultivars was not significant. Implying that the tomatoes are not different in attracting or deterring the visitors. The general trend was that the PTM male activity increased towards the end of the crop season and corresponded with the peak harvesting of the tomatoes and the maturity of the potato tubers. Though there were three peaks observed during the vegetative growth stage of the crops, all were found to be significantly lower than last peak. This gives a clue again that both in tomatoes and potato there may be some factors, which did not allow free visitation by PTM adult males and also the build up of PTM within the plots.

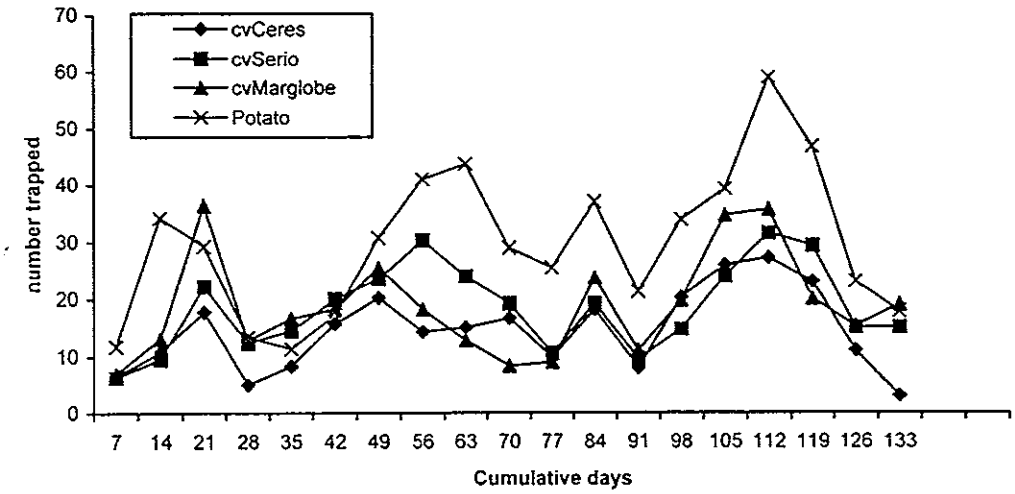

Fig 1. Mean population dynamics of potato tuber moth males in tomato and potato plots.

1.2. Field monitoring of the PTM on tomato and potato leaves

1.2.1. Materials and methods

The monitoring of PTM larvae and PTM mined leaves was initiated one week after the sex pheromone trapping began and has been carried out every week until the tomatoes started bearing fruits. On each sampling date and on each of the plots wherein the traps were installed, 25 plants were selected at random and examined for PTM larvae and PTM mines.

1.2.2. Results

The number of larvae and the number of mines per plant were significantly higher in the potato plots (Figs 2 & 3). The difference among the three tomato cultivars was not significant. The general trend was that the larvae in active mines and the number of active and non-active mines per plant increased in the last phenological stages of the vegetative phase. This comparison of the host preference of the PTM in the natural setting has clearly shown that given the opportunities, it is still the potato, which is preferred by the PTM as its best feeding niche when compared with tomatoes. The reduced attack sustained by the tomatoes in the presence of potato may imply that there are some tomato plant linked factors, which reduce the biological performance of PTM larvae in tomatoes.

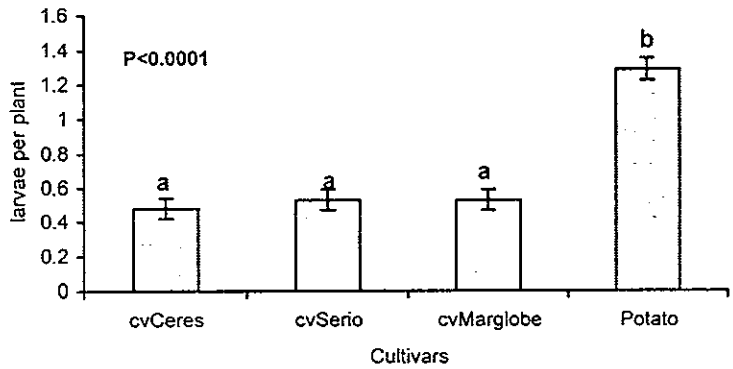

Fig 2. Mean number of PTM larvae per plant recorded on the leaves of the tomato cultivars and the potato

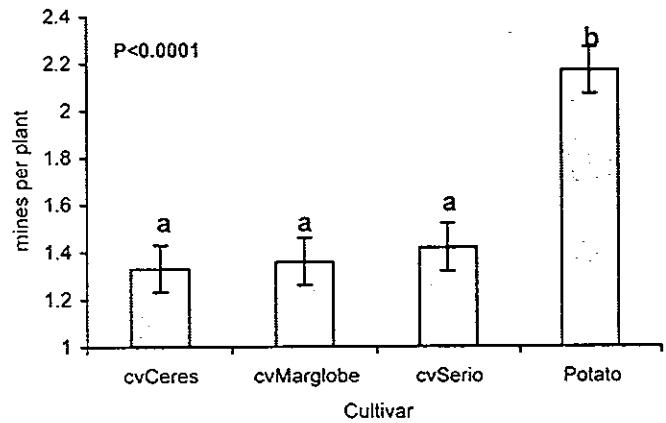

Fig 3. Mean number of PTM mines per plant recorded on the leaves of the tomato cultivars and the potato.

1.3. Field monitoring of the PTM in fruits

1.3.1. Materials and methods

This was carried out by random sampling, on weekly bases, of 25 plants per plot and picking all the borne fruits on it and categorizing them as immature green, maturing green and ripening. All these fruits were then examined for PTM infestation, by checking for the typical hole bored by PTM in tomato fruits by entering under the peduncle, on each and every fruit. The number of PTM larvae present and infested fruits per plant has been recorded on each of the ten sampling dates, which corresponded with the subsequent tomato fruits picking dates.

1.3.2. Results

Fruit infestation began in green immature fruits in all the cultivars, but significantly different among the three and was highest in cv Marglobe. The same trend continued through to the ripening stage and the fruits of the cv Marglobe were the most attacked. Comparison of the cultivars based on infested

fruits without considering fruit stages even was consistent with the outcomes for the different fruit classes (Fig 4).

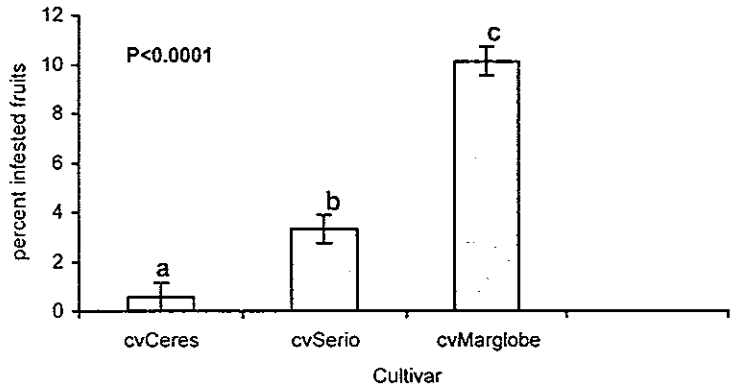

Fig 4. Mean percent infested fruits per plant recorded in the three tomato cultivars.

2. Influence of leaf tomato trichomes on the potato tuber moth

2.1. Determination of total and type VI glandular head trichomes on tomatoes leaves

2.1.1. Materials and methods

The density of total leaf hairs and a specific glandular head trichome called type VI densities on the leaves of the three tomato cultivars were determined at two specific developmental stages: preblossom at nine true leaf stage and blossom at least at 50 % flowering. Each time five fully expanded leaves were cut at random locations on five separate plants in the PTM activities monitoring field by taking all precautions to protect loss of the glandular head trichomes at sampling time. From each leaf two leaf disks were cut out after removing dust on the leaves using gently flowing tap water. Then transferred to a microscope slide and examined at 100X magnification under a compound light microscope.

2.1.2. Results

The total leaf trichome density per field of vision at 100X magnification was significantly higher on the cv. Marglobe and cv Ceres had much lower density of total trichome cover Fig 5. But, total reversal and significantly higher number of the type VI glandular head trichomes was observed on the leaves of cv Ceres in both stages of development (Fig 6). Whereas the difference in the total trichome cover was found to be insignificant at blossom stage. In general, however the distribution of the glandular trichomes was found to be highly variable and the proportion of type VI to total was low in the cv. Marglobe and cv. Serio. To determine the possible influence of trichomes on the PTM, larvae settling and females' oviposition responses were evaluated.

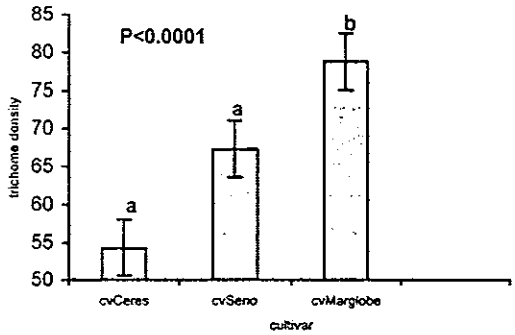

Fig 5. Mean number of total leaf trichome on the leaves of three tomato cultivars per field of vision per cultivar at 100X magnification.

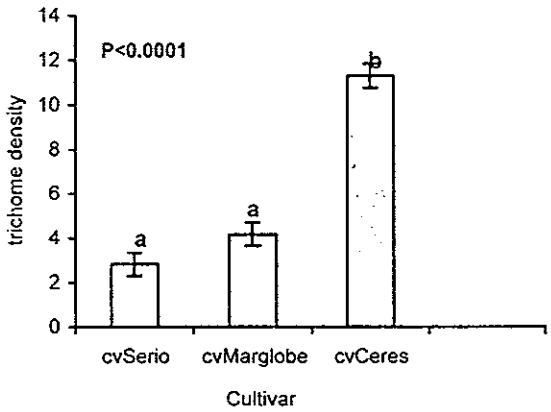

Fig. 6. Mean number of type VI glandular head trichomes per field of vision per cultivar at 100X magnification.

2.2. Influence of leaf trichomes on the settling response of neonates of the PTM on tomato and potato foliage

2.2.1. Materials and methods

A no choice experiment was designed using freshly excised leaflets from fully expanded leaves of the three cultivated tomatoes and a standard PTM host potato. Two leaflet sets of five each were prepared at a time from each cultivar of tomato and the potato. One set of leaflets was left unwashed and in the other set, they were washed with Triton 0.1% solution to remove the leaf trichomes and their contents. Each leaflet was then transferred into individual Petri dishes lined with moistened filter paper. Then after five neonates were introduced onto each of the leaflets. Examination under stereomicroscope was made to determine the time it took each of the PTM larvae in each of the leaf disks to settle underneath the leaf sheaths and started sustained feeding. This response was examined and recorded after 15, 30, 60, 120, 180 and 240 minutes. This has been done both on the abaxial and adaxial surfaces of the leaves from the tomatoes and the potato.

2.2.2. Results

The total trichome cover on the leaves of cv Marglobe was shown earlier to be significantly higher than on the others, but this failed to cause any adverse effect on the settling response of the neonates of PTM more than the trichome cover in the other two cultivars. Instead, on the leaf samples taken at preblossom stage, the settling of larvae on cv Marglobe leaves was second to the PTM susceptible host, potato. On the other hand, the type VI glandular head trichome density was found to be higher in cv Ceres in both stages of leaf sampling, however, this too, did not interpret into any significant adverse effect on the settling larvae (Fig 7).

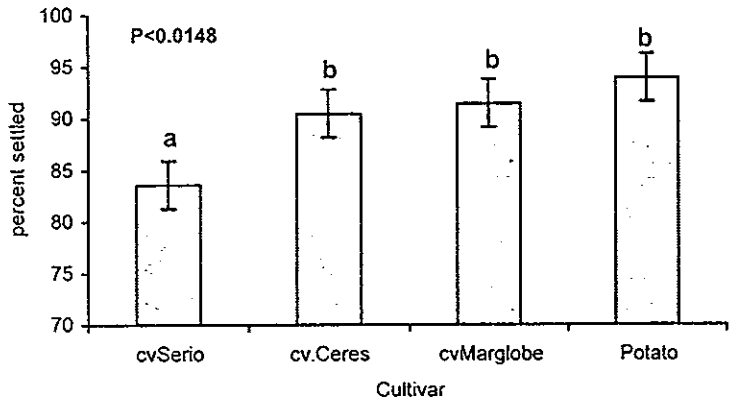

Fig 7. Mean percent settling response of neonate of the PTM on tomato and potato foliage sampled at preblossom stage.

3. Survival growth and development of PTM larvae on the leaves of tomato cultivars and potato

3.1. Materials and methods

The survival, development and growth of neonates of PTM on the foliage of tomatoes and potato have been carried out using excised leaflets. Fully expanded leaves were sampled in the monitoring field from the three tomato cultivars. The leaves were protected from desiccation by covering the severed end with wetted cotton and putting them in cool iceboxes until use. Petri dishes were cleaned with 70% ethanol alcohol and dried and were lined with filter papers and subsequently moistened with heat sterilized water. Following this, the sampled leaves were thoroughly examined for any physical damage or lesions and the healthy ones were cut and transferred into Petri dishes either the abaxial or adaxial surfaces facing up without washing or after washed with Triton 0.1% solution. After this, one neonate PTM larva was introduced into each Petri dish and tightly closed with the aid of rubber bands and transferred into a growth chamber with the inside temperature of 26°C, 12:12 L:D and 70% RH. Each Petri dish has been examined everyday until the developing larva completes its development or die before. Data were recorded on the survival over the test period and between the different larval developmental stages, developmental time for the different instars and the pupae, body weight and

body length of wandering larvae and body weight of pupae and adults. This work was done in preblossom and blossom stages.

3.2. Results

The number of PTM larvae, which survived to complete development and reach adult stage on the foliages of tomato cultivars Serio and Marglobe and Potato, taken at preblossom stage of the crops, was found to be significantly lower than on the foliage of the cultivar Ceres (Fig 8). Whereas, when the leaf samples were taken at blossom stage, the tested PTM larvae survived to complete their development more on the potato foliage and least on Marglobe (Fig 9). Upward trend in the number of PTM larvae surviving on foliage was shown only in the potato cultivar. The highest survival rate on the foliage of cv Ceres did not persist at blossom stage implying that some thing has happened in the foliage of this cultivar, which has negative effect on the developing larvae. One thing clear is that the cultivar is determinate type and does not produce more new leaves after blossoming and depends on leaves from early growth stages. Even then, however, it provided better condition for larvae than the indeterminate cultivar Marglobe. This is one possible indicator that allelochemical buildup occurs in cv Ceres at the latter stage of development.

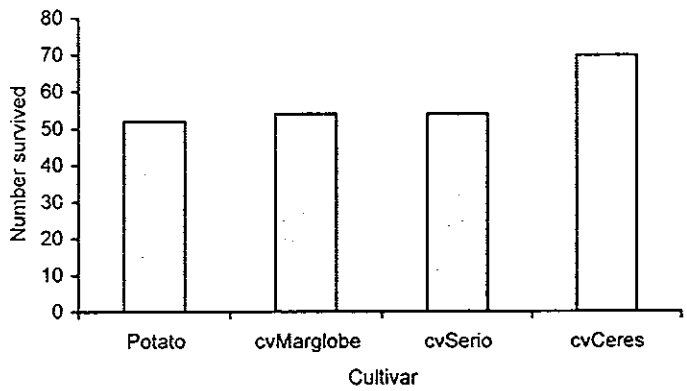

Fig 8. Number of PTM larvae survived to complete their development (n=80) on three cultivars of tomatoes and potato cultivar foliages taken at preblossom stage.

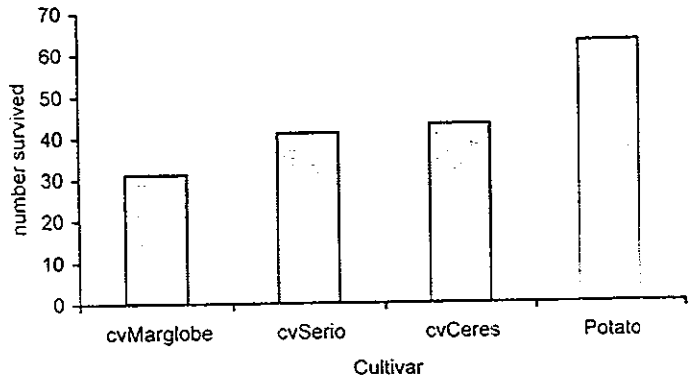

Fig 9. Number of PTM larvae survived to complete their development (n=80) on three cultivars of tomatoes and a cultivar of potato foliages taken at blossom stage.

The total development times for larvae on the three tomato cultivars at preblossom and blossom stage were found to be different. For instance on the cv Ceres, on which foliage more larvae survived at preblossom stage, the development time for the tested larvae was significantly lower than on the other tomato cultivars, however despite the lower survival in potato, the total development time was lowest for larvae reared on its foliage and completed their development. At blossom stage, the development time of larvae on cv Ceres was the highest and was consistent with the lower survival of larvae reared on its foliage at this stage. On cv. Serio, developmental delay was lowered at blossom stage. Whereas on the foliage of Marglobe, this was not significantly different both from cv Ceres and cv Serio at preblossom stage, but at blossom stage, it was significantly lower than on cv Ceres only (Fig 10 & 11). The consistently significantly lower development time for larvae on potato foliage and the increased survival of larvae from preblossom to blossom stage imply that Potato is the best host for the PTM and the leaves of the tomatoes are posing some hurdles on the larvae of PTM.

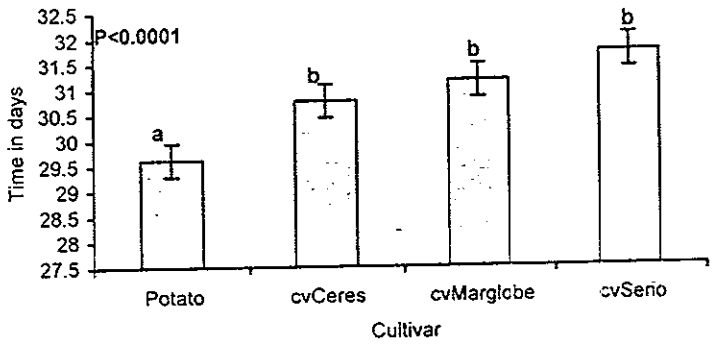

Fig 10. Mean development time for PTM larvae reared on tomato and potato foliages from preblossom stage to reach adult stage.

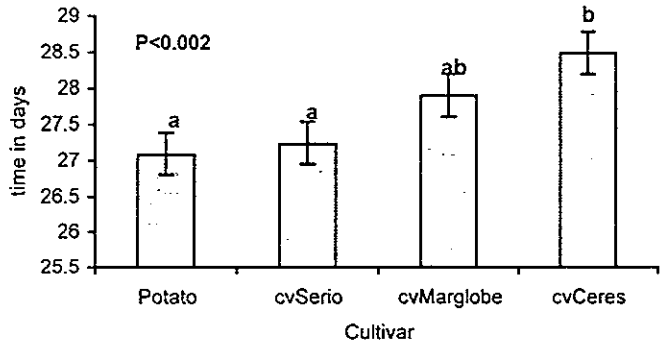

Fig 11. Mean development time for PTM larvae reared on tomato and potato foliages from blossom stage to reach adult stage.

4. Survival growth and development of PTM larvae on the fruits of three tomato cultivars

4.1. Materials and methods

Lightweight cages, which do not cause damage to tomato branches bearing bunch of fruits were made from fine plastic mesh and polystyrene foam. For the cultivar Marglobe, bigger sized cages were made to accommodate one bunch of fruits as its fruits size is much bigger than the other two cultivars fruits. Ten branches per plot and 40 per cultivar were individually caged after counting the number of borne fruits in each caged bunch and introducing neonates of the PTM, twice in number to the caged fruits. The fruits in each cage were collected in separate bag to the lab after 15 days and the number of infested fruits, surviving larvae and the body weight and head capsule size of the recovered larvae were measured. This was done on immature green, maturing green and ripening fruits.

4.2. Results

More larvae survived in the fruits of cv Marglobe ($P<0.0001$) (Fig 12). Moreover results consistent with the natural field infestation of tomato fruits by the PTM were obtained (Fig 13). The body weight gain and developmental stage of PTM larvae recovered from the fruits of cv Marglobe were significantly different from the others. The larvae were heavier in size and reached the fourth instar stage in less than 15 days (data not shown).

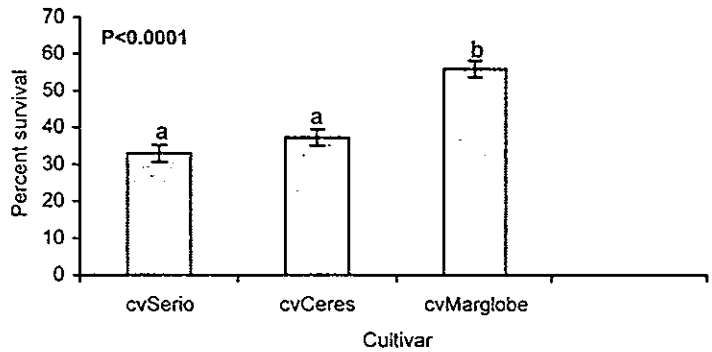

Fig 12. Mean percent survival of PTM larvae in the fruits of the three tomato cultivars.

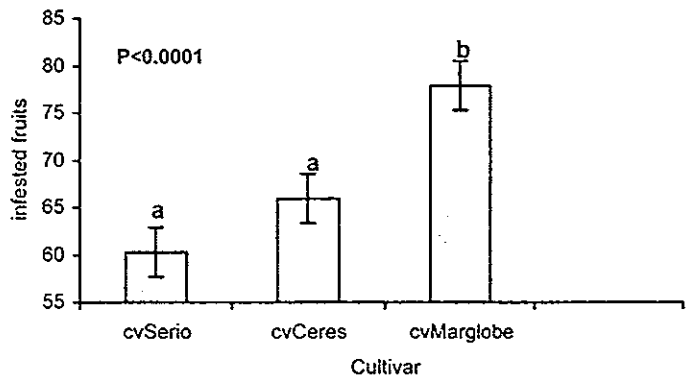

Fig 13. Mean percent PTM infested fruits in the three tomato cultivars.

5. Influence of three tomato cultivars and a potato on the activities of the natural enemies of the PTM larvae

The part of the work has been carried out in Ethiopia. But, more has to be done here in Rehovot to get a complete picture on the interaction. The results will be reported in the final report.

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Section I

I.C. Scientific Impact of Collaboration:

During most of the period covered in this report, Mr. Bayeh Mulatu Aregay, the joint Ethiopian graduate student in this project, completed the second year of fieldwork in Ethiopia. Intensive field monitoring and elaborate laboratory experiments were conducted. In May, Mr. Mulatu arrived in Israel and is already conducting some research as planned. PTM, parasitoid, and predator cultures have been established in Rehovot from field-collected material.

I.D. Description of Project Impact:

The results of the studies presented here have shown that potato is the most important host to the PTM and its natural enemies. PTM populations remain low during the vegetative stages of the potato and tomato plants and they increase late in the growing season at tubers and fruits are set. The tomato cultivar Marglobe allowed more PTM larvae to survive, attain higher body weight and reach the last instar faster. Tomato plants provided enemy free environment to PTM larvae. However, this host may be less suitable for larval development.

I.E. Strengthening of Developing Country Institutions:

Investments in equipment and in facilities, including field facilities, are an ongoing process. They proceed reasonably well, under the circumstances. Much progress has been attained by Mr. Beyeh Mulatu, the Ethiopian PhD student. He is advancing nicely and proving to be good human investment. There are still some institutional constraints, primarily financial ones.

Section II

II.A. Managerial Issues:

The orderly and timely transfer of funds to Ethiopia was a main problem: The initial advance was transferred rapidly, but the next transfer(s) took months to achieve, and the Ethiopian team was left with no financial support for an extended period.

II.B. Budget: Transfer of money from the Ethiopian budget to support the Ethiopian graduate student while he stays in Israel has been made. Israeli travel fund has also been transferred to the same purpose.

II.C. Special Concerns: None

II.D. Collaboration, Travel, Training and Publications:

Collaboration -

Frequent communications take place between the Principal Investigators and Dr. Gebremedhin and Mr. Mulatu. These e-mail and phone communications involve both managerial and scientific issues. While in Ethiopia, Mr. Mulatu often sought the advise of the Israeli PIs that serve, together with Dr. Gebremedhin, as his PhD advisors.

Travel -

Mr. Bayeh Mulatu arrived in Rehovot Israel in May where he is scheduled to conduct research, write his dissertation, and complete all requirements toward his Ph.D. degree at the Hebrew University of Jerusalem.

Publications -

Zhou X., Coll M. and Applebaum SW. 2000. Effect of temperature and photoperiod on juvenile hormone biosynthesis and sexual maturation in the cotton bollworm, *Helicoverpa armigera*: implication for life history traits. Insect Biochemistry and Molecular Biology 30: 863-868.

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II.E. Request for US Embassy (Tel Aviv) and AID Actions: none