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Integrated Protection of Tomato Crops against *Tuta absoluta* in Open Ground Conditions in the South-East Part of Kazakhstan

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Abstract: There is a need to develop strategies for the protection of tomato crops that would provide high-quality produce safe for the consumer without environmental pollution. The objective of the study is to deploy an integrated system for the protection of open-field tomato crops from *Tuta absoluta* using biological agents (*Bracon hebetor* Say., *Trichogramma*, biopreparations, pheromone traps) and devices in the conditions of southeast Kazakhstan to obtain environmentally safe produce. Experiments on the protection of tomato crops from pests are conducted by applying integrated protection measures with the use of entomophages, biopesticides, pheromone traps, and low-hazard pesticides on the tomato field of the Musa private farm. Per the results of the experiments on protecting open-field tomato plants from *Tuta absoluta*, the effectiveness of the employed instruments reaches up to 90%. The obtained results can be used to protect tomato plants accounting for pest density and the phase of crop development. Thus, based on the results obtained, it can be concluded that the primary elements of the integrated system for the protection of tomato crops against *Tuta absoluta* should include the application of biological preparations, entomophages and, if necessary, low-hazard insecticides, as well as the use of pheromone traps to detect the appearance of the moths and capture them en masse.

Keywords: Plant Protection, *Tuta absoluta*, *Bracon Hebetor* Say, *Trichogramma Achaeae*, Biopreparations, Pheromone Traps

Introduction

Agriculture is among the leading industries in terms of anthropogenic load. The chemicalization of land causes significant damage to the environment (Akhtar *et al.*, 2021; Martirosyan *et al.*, 2022). About 2 million tons of pesticides are used worldwide every year. Their residues are found in 40% of the tested samples of grains, berries, fruits, and vegetables. Every year, 25 million cases of pesticide poisoning are registered around the world, including 20,000 fatalities, not to mention those human diseases that are progressive (Larramendy and Soloneski, 2014; Tudi *et al.*, 2021).

Today, the intensification of agriculture around the world, including in Kazakhstan, has become an extremely urgent economic task at the national and international levels. In this context, the problem of protecting crops and yield from pests is a critical issue. The application of an integrated crop protection system including Unmanned Aerial Vehicles (UAVs), biological agents, and biopesticides is one solution to this problem that is growing increasingly notable as an alternative to chemical agents and a full replacement for them since the biological method as a selective effect on insect pests while being harmless to humans and the environment (Erol *et al.*, 2021).

Tomatoes prevail in the structure of cultivated areas of vegetable crops in all categories of farms in the region, taking up 20 thousand ha, or 21.2% (Moldashev *et al.*, 2017; Iztleuov *et al.*, 2020). Regrettably, the current yield of tomatoes does not fully satisfy the needs of the population of the regions of Kazakhstan and exports to the northern regions. For this reason, tomatoes are imported to Kazakhstan from the neighboring countries, mostly Uzbekistan, Kyrgyzstan, and China (Iztleuov *et al.*, 2020).

One of the reasons behind the low yield of tomatoes is damage to the crop by a wide range of pests and diseases. At present, one of the topical and acute problems faced by agriculture in Kazakhstan, especially in its southern regions, is the South American tomato moth *Tuta absoluta* (Erol, 2021), which has the status of a quarantine pest in the country (Campos *et al*., 2017).

The high level of harmfulness of the tomato moth is due to some characteristic features of this insect. An adult female is capable of laying more than one hundred eggs in her lifetime and given the rapid development of offspring, she may produce thousands of small caterpillars that actively devour tomato crops and plantings (Campos *et al*., 2021).

Feeding on the leaf mesophyll, the caterpillars destroy the photosynthetic system (Cherif *et al*., 2018). Major leaf damage can lead to defoliation. If the fruit is damaged, the growth processes are disturbed, in some cases deformed and, as a consequence, marketable qualities are lost.

By various estimates, the losses in yield and marketable quality of products as a result of damage by this pest can reach from 35 to 100% (Esenali Uulu *et al*., 2017; Prasannakumar *et al*., 2021).

Unfortunately, to date, the fight against *Tuta absoluta* in the regions of Kazakhstan in the open ground involves the use of chemical pesticides, which can lead to detrimental effects on biocenosis (Esenali Uulu *et al*., 2017).

The goal of our research is to deploy an integrated system for the protection of open-field tomato crops against *Tuta absoluta* using biological instruments (*Bracon hebetor* Say., *Trichogramma*, biological agents, pheromone traps) and UAVs in the conditions of southeast Kazakhstan to obtain environmentally safe vegetable produce.

Materials and Methods

Location of the Study

Research on the dynamics of the proliferation of *Tuta absoluta* and the development of measures to control it in open field conditions was conducted in 2021 on the Musa farm in the Enbekshikazakh district, Almaty region, Republic of Kazakhstan. The fields of the Sapar and Yultuz farms located in the same region were observed as control plots.

The tomato variety cultivated on the experimental plot is the Apache F1 hybrid. This is a hybrid with very early flowering and a plant height of up to 35 cm. It has uniform oblong fruits, resistant to cracking. The plant structure completely covers the fruit. This is the variety most commonly cultivated in the open field in the Almaty region.

Objects of the Study

The object of research is the *Tuta absoluta* moth. The means of pest control deployed in the study include:

Trichogramma achaeae-a parasitoid wasp that exclusively controls *Tuta absoluta* eggs (Bioline Agrosciences, UK).

Bracon hebetor Say-a parasite of scale fly caterpillars, Zhiembaev Kazakh Research Institute of Plant Protection and Quarantine LLP.

Bitoxybacillin (BTB)-biological preparation against lepidopterans and other pests. Spores and cells of culture-producer Bacillus thuringiensis var. thuringiensis, Sibbiofarm, Russian Federation.

Actarophyt-a biological preparation, a complex of natural avermectins, which are produced by the beneficial soil fungi Streptomyces avermitilis, Enzyme, Ukraine.

Coragen-broad-spectrum insecticide against insect pests, active agent chlorantraniliprole, hazard class 3. DuPont, France.

Another control agent used is the pheromone of *Tuta absoluta*-a rubber dispenser containing artificially synthesized pheromone identical to the natural pheromone of *Tuta absoluta*. Complete with a yellow glue trap, Russell IPM, Turkey.

Sampling Methods

The research used sampling methods to estimate tomato moth egg density presented in Alfredo H.R. Gonring *et al*. (2020). The eggs were counted on plants in the reproductive stage. The entire canopy of each plant was divided into three sections: Apical, middle, and basal. The pairs of leaves between these sections (apical/middle and middle/basal) were considered to belong to the transitional regions between these canopy sections. The leaves were numbered in ascending order from the apex of the plant. Thus, the first leaf on the plant is the most apical, etc. Egg counts of *T. absoluta* were estimated by direct counting the number of eggs. Monitoring of the phytosanitary condition of tomato fields was carried out weekly long before the beginning of the growing season and until the end of the flight of the pest imago, from the third ten-day period of March to the first ten-day period of November.

Information about the phytosanitary condition of tomato fields was collected using UAV DJI Phantom4 Multispectral (DGI, China) equipped with a multispectral camera shooting in additional parts of the spectrum: Red Edge (RE): 730 ± 16 nm, Near-Infrared (NIR): 840 ± 26 nm.

The primary concept of the research was to perform investigations utilizing a ground description of the territory and route overflight of the points to compare them to enable the identification of the images.

Field studies were conducted during the growing season (May-August) of 2021. In the course of preparatory work, the coordinates of the corners of the polygon to be shot were determined and the characteristic points on the ground were placed and coordinated, which served as reference points for georeferencing the orthophoto plane during further processing of images. To monitor the variability of the phytosanitary situation in the field during the growing season, aerial observations were carried out in the periods of the initial phase of growth and development, the middle of the growing season, and crop maturity.

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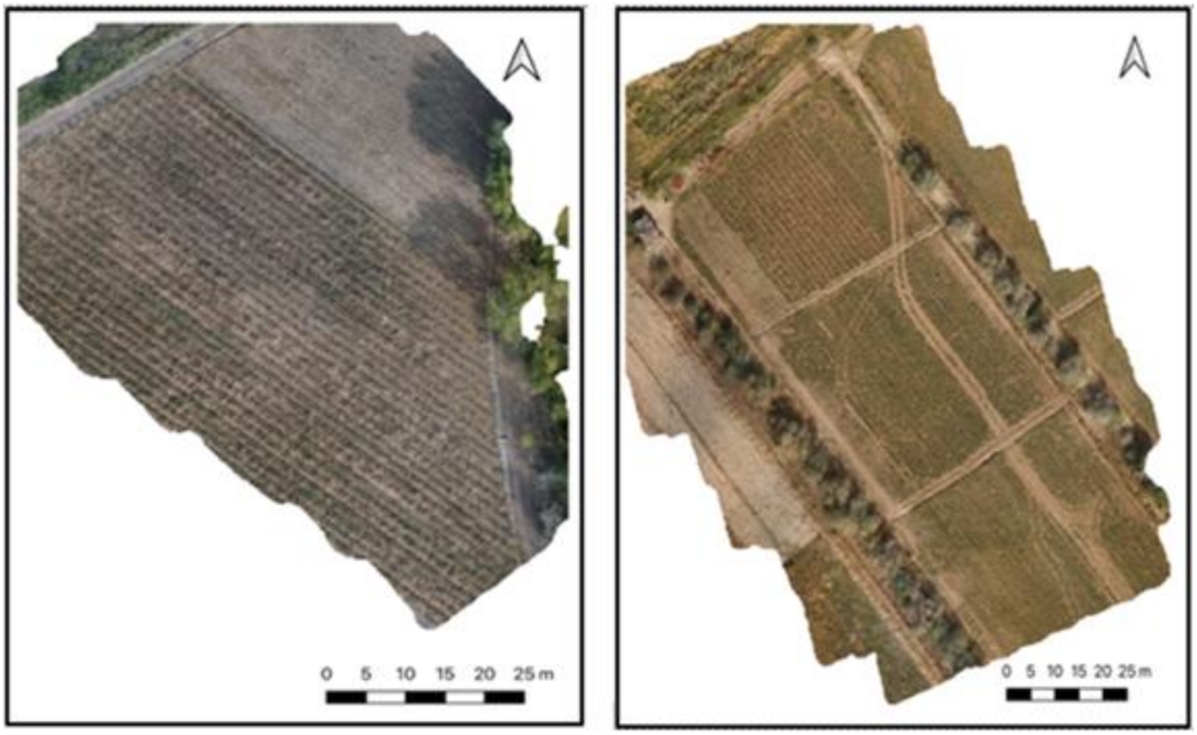

(a) (b)

Fig. 1 : An example of orthophoto maps of the experimental and control sites in the visible range of the electromagnetic spectrum: (a) experimental plot, (b) control plot

Fig. 2: Glue trap with *T. absoluta* pheromone

Desk work consisted in performing a cycle of operations aimed at studying the decoding features and formatting the cartographic materials in the infrared and visible emission of the electromagnetic spectrum.

Pre-processing included creating an orthophoto map of the captured terrain. The number of scenes depended on the size of the test polygons. The images were georeferenced based on UAV data, with coordinates of the central points of the images (Fig. 1).

The obtained aerial photos from the multispectral camera installed on the UAV were subjected to automatic classification in GIS software products Agisoft Meta shape Pro (photogrammetry) and QGIS (post-processing and coloring of maps).

The primary host of *T. absoluta* is the tomato plant (*Lycopersicon esculentum* L.). Many literary sources suggest that *T. absoluta* can also feed on eggplants (*Solarium melongena* L.), potatoes (leaves and stems only), peppers (*Capsicum sp*.), and other plants of the Solanaceae family. Even though eggplant and pepper crops were located some 20 m away from the experimental plot, no damage was found on these cultures. In the first ten-day period of October, traces of the phytophage were observed everywhere on Solanum villosum (*Solanum zelenetzkii*).

To take timely and prompt measures to protect the crop, it is important to have information about the state of the quarantine pest population in its dynamics. For this purpose, glue traps with pheromones for *T. absoluta* were hung in the fields planted with tomatoes (Fig. 2).

The traps allow us to precisely and quickly establish the time of appearance of the pest, as well as to further observe the dynamics of activity of *T. absoluta* (Joshi *et al*., 2018; Poudel and Kafle, 2021). One trap was used for three weeks. In the experimental plot, 50 plants were inspected for phytophage infestation and the number of pest-infested plants was counted to determine the degree of infestation.

The degree of tomato moth damage to plants is calculated using the formula:

$$D = Ax100 / H \quad (1)$$

where:
D = Degree of damage, %
A = The number of plants with pests detected
H = The total number of plants inspected

Testing of chemical and biological preparations in the field was carried out according to the methodological guidelines (Metodicheskie Ukazaniia, 2015). The agents used to control the pest on the test plot included bioinsecticides-Bitoxybacillin (3 kg/ha) and Actarophyt (2l/ha), entomophages-*Trichogramma achaeae* and *Bracon hebetor* Say and low-hazard insecticide Coragen (0.15 l/ha). Treatments were performed at different times throughout the growing season, considering the distribution of the pest.

The first release of trichogramma to the field was performed 5 days after catching an average of 2-3 butterflies per trap per night in the first generation of tomato moths and, respectively, 3-4 days after catching 1.5-2 butterflies in the second and third generations. Trichogramma was released in the evening hours when it was not hot. Trichogramma was dispersed in at least 50 points every 10 m.

Bracon was used against each generation of older *Tuta absoluta* caterpillars three times in a parasite-to-pest ratio of 1:5 (Alpysbayeva *et al*., 2021).

Treatment of tomato fields with biological preparations was carried out using the Qarlyqash UAV (Agrodron, Kazakhstan). Each variant was applied once.
Evaluation of the biological effectiveness of biopreparations and entomophages was calculated according to Abbott's formula 2:

$$E = (A - B) / A \times 100 \quad (2)$$

where E-biological effectiveness; A-the number of eggs/caterpillars before the application of biological agents; B-the number of healthy eggs/caterpillars after the application of biological agents (Moldashev *et al*., 2017; Akhmedova *et al*., 2018; Iztleuov *et al*., 2020).

Monitoring was conducted on days 3, 5, and 7 after treatment.

Statistical Analysis

Statistical processing of the obtained data was carried out by the method of descriptive statistics for assessing biological effectiveness using "Microsoft Office Excel" spreadsheets and "SigmaStat 3.1" application software package.

Results

Preliminary Observations

Preliminary results of the study reveal that the use of UAV imagery data and the NDVI vegetation index clearly shows pixels where the values in the image are close to plus one (where photosynthesis occurs) or approach zero and minus one (no photosynthesis, indicating unhealthy plants), which is the solution to the project problem at this stage.

The NDVI index was calculated for images from the camera, an example is provided in Fig. 3.

Tuta absoluta is harmful from the very beginning of seedling planting. Eggs are laid on the surface of the vegetative organs of plants (Fig. 4a). *Tuta absoluta* caterpillars develop in living plant tissues, eating out mines in leaf parenchyma (Fig. 4b), stem, or fruit, and can damage apical buds of plants. When plants are massively damaged, growth retardation or stoppage is observed.

The Activity of Tuta absoluta Moths

The timing of pest imago flight is determined using pheromone traps. The dynamics of moth trapping are presented in Table 1.

The first *Tuta absoluta* moth was found on a pheromone trap on May 26 in a greenhouse of the Musa farm located in the Kyzylsharyk village 1 kilometer away from the field where research was carried out throughout the vegetation period. The first appearance of the pest on the experimental plot itself was reported on June 10.

The duration of the flight activity of the moths is presented in Fig. 5.

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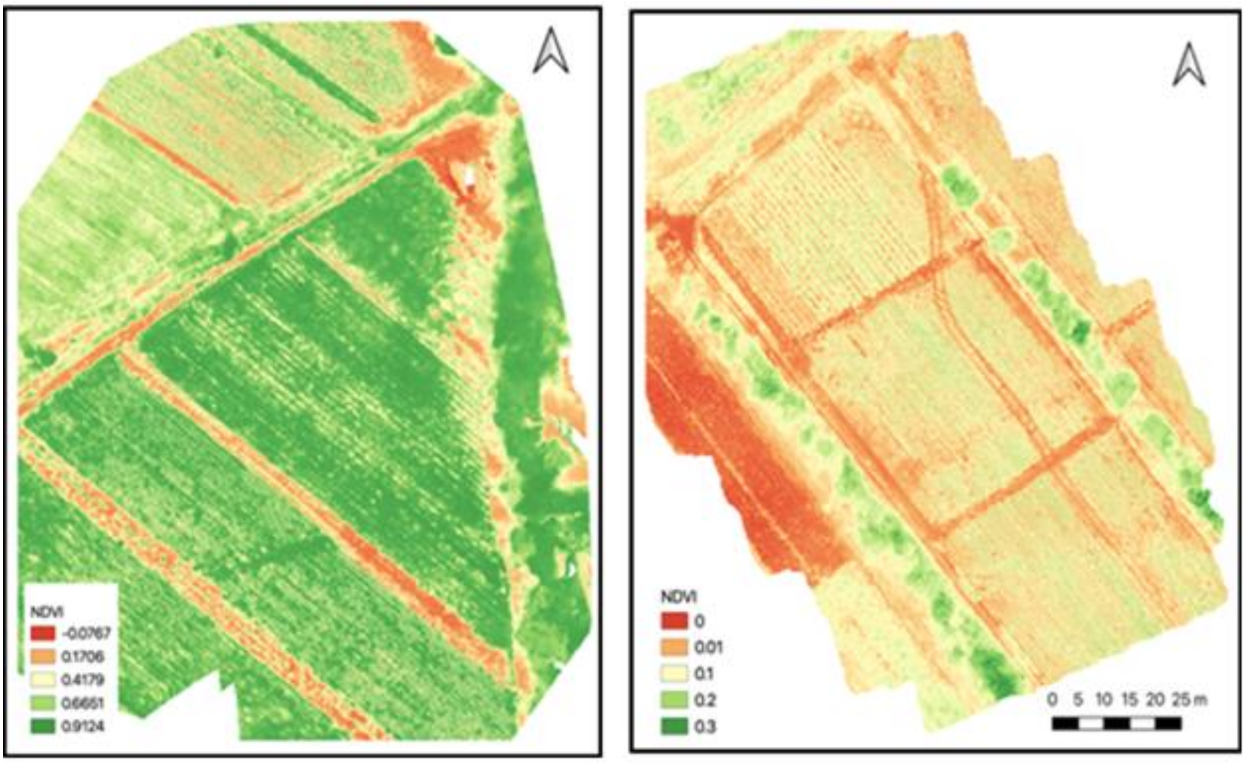
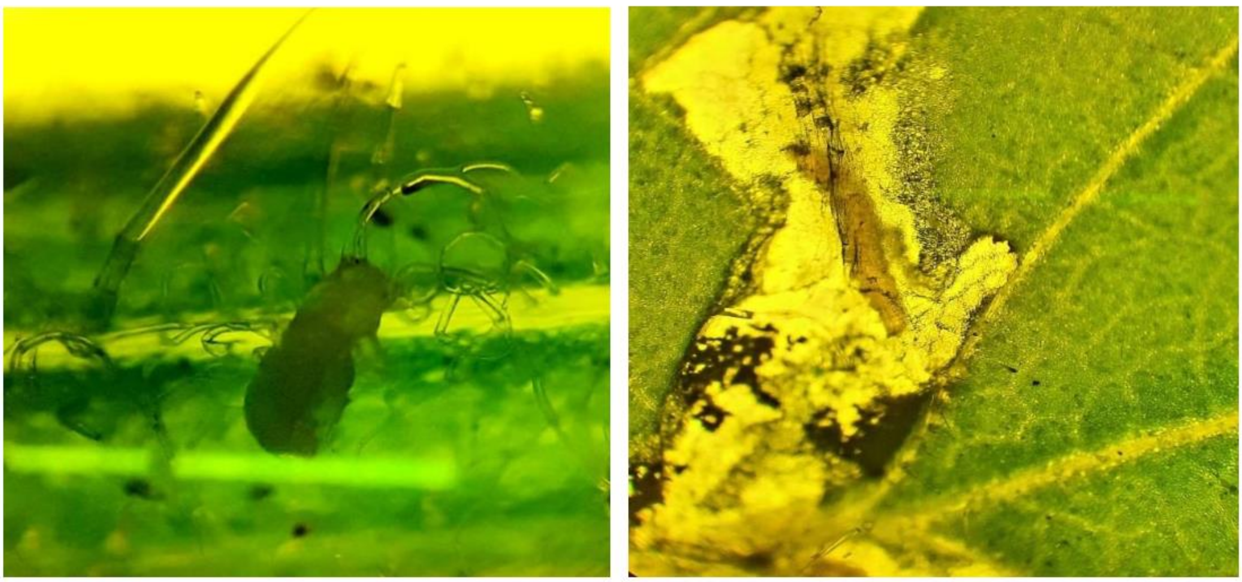


(a) (b)

Fig. 3: The calculated NDVI index: (a) experimental plot, (b) control plot



(a) (b)

Fig. 4: *Tuta absoluta*: A. the caterpillar's exit from the egg; b. caterpillar’s in tomato leaf parenchyma

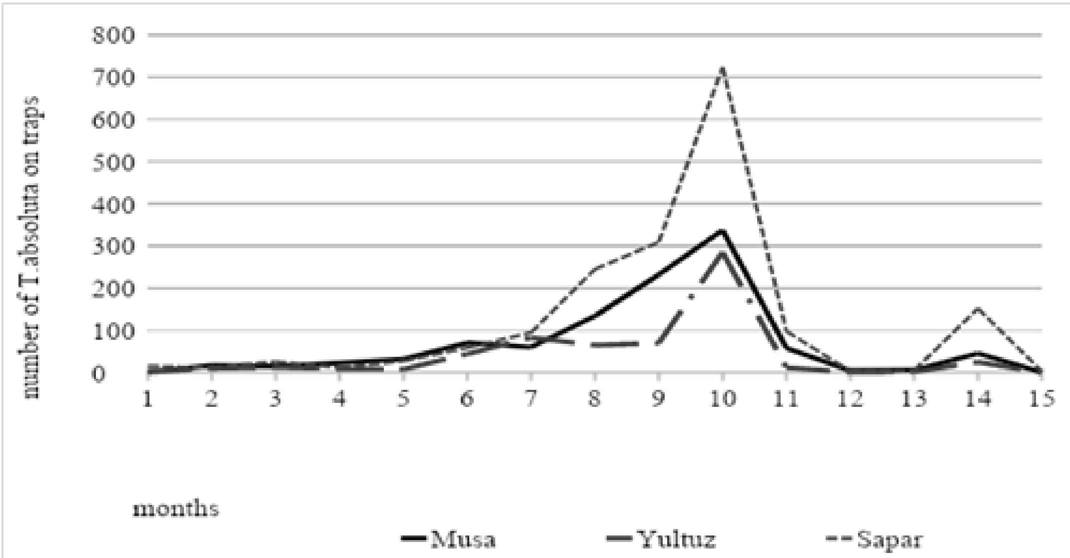


Fig. 5: Dynamics of the flight activity of *Tuta absoluta* moths, Almaty region, Enbekshikazakh district, Republic of Kazakhstan

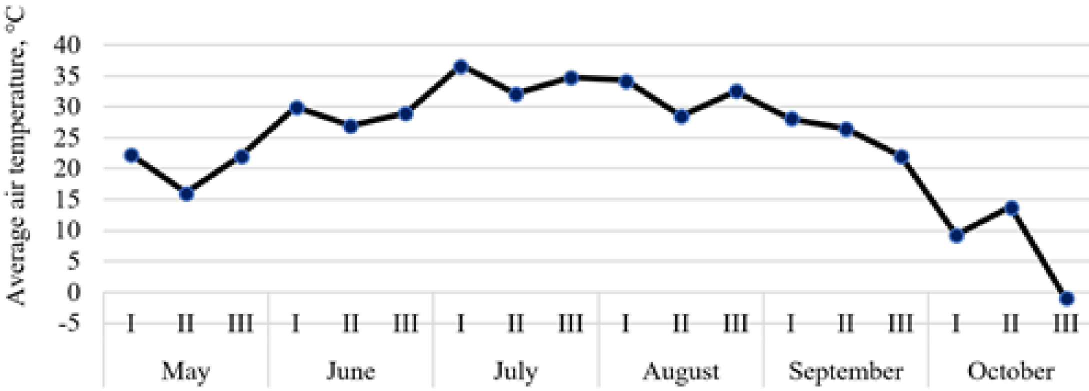


Fig. 6: Average air temperature, C

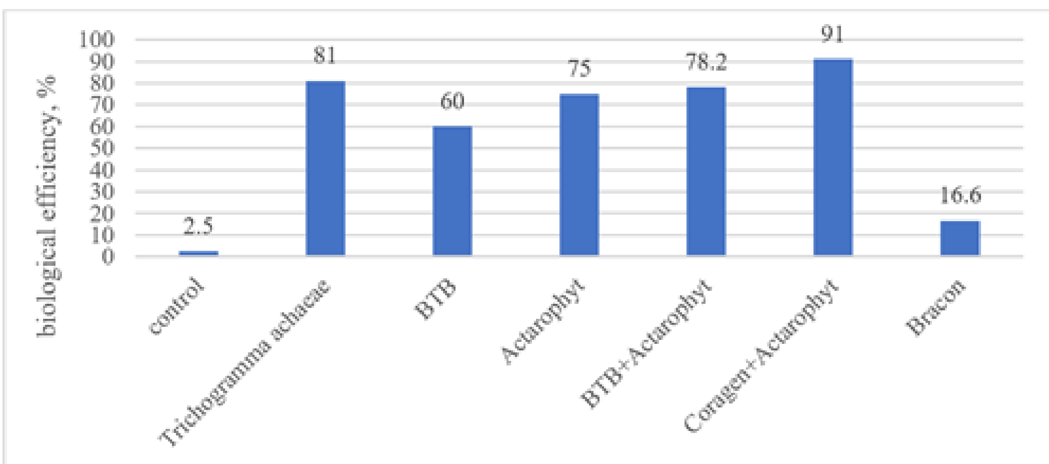


Fig. 7: Biological effectiveness of tomato protection agents in open field conditions, 2021

Table 1: Phenology of *Tuta absoluta* on tomatoes in the conditions of Almaty region, Enbekshikazakh district, Republic of Kazakhstan, 2021

MAY			JUNE			JULY			AUGUST			SEPTEMBER			OCTOBER		
ten-day periods																	
I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III
		T	T	T	T												
				*	*	*	*	*	*								
					¤	¤	¤	¤	¤	¤	¤	¤	¤				
		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
			•	•	•	•	•	•	•	•							
				1	1	1	1	1	1	1	1	1	1				

T-growth, ∗-blooming; ¤-fruiting; + – start of insect flight, flight; • -oviposition; 1-caterpillars *Pest development phenology according to V.V. Verderevskiĭ (1968)

As demonstrated in the above figure, the activity of the moths starts to markedly escalate starting from the 3rd ten-day period of July. While in early summer, the number of moths in the traps is between 2 and 10, in the 3rd ten-day period of July, there is a noticeable increase to 44 and more imago. On the Musa farm, the peak of moth activity is recorded in the first ten-day period of September; during the same period, the number of adults in the control field of the Sapar farm is 723 specimens. This may be because no protective measures against *Tuta absoluta* were taken in this tomato field. On the sites of the Yultuz farm, the activity of the tomato moth is significantly lower compared to the other two farms since the very beginning of the growing season. The flight of *Tuta absoluta* continues until the 3rd ten-day period of October. At this time, there is a sharp decrease in air temperature at nighttime (reaching -1ºC), after which point the activity of the phytophage stops.

A significant increase in the activity of *Tuta absoluta* in all the studied territories is noted since the 2nd ten-day period of August. Sharp drops and rises in the rate of imago flight in late September and early October are associated with temperature fluctuations (Fig. 6). For instance, in early October, the average daytime air temperature can be seen to be below +10°C. At that time, the total number of *Tuta absoluta* moths is no more than 5. Yet 10 days later, with daytime air temperature reaching+15°C, the moths resume their active flight. On the field of the Sapar farm, the number of captured moths is 152, on the Yultuz farm-25 and the Musa farm-45 specimens. With air temperature dropping to -1°C in the 3rd ten-day period of October, the flight of the phipophage ceases. The total number of *Tuta absoluta* captured throughout the growing season is 3,617 specimens.

In the conditions of the Almaty region, the tomato moth produced five generations in 2021.

Biological Effectiveness of Protection Agents

Treatment with biological agents seems to be one of the promising directions of crop protection against pests. In our experiments, together with the release of entomophages, we used biological preparations such as Bitoxybacillin and Actarophyt. At the same time, we also used a low-hazard preparation, Coragen. The results of the tests are shown in Fig. 7.

As demonstrated in Fig. 7, the lowest effectiveness is achieved by BTB with biological effectiveness of 60%. The efficiency of Actarophyt in the open ground proves to be quite high-75%. Notably, a tank mixture of BTB and Actarophyt also demonstrates high biological effectiveness-78.2. 81% effectiveness against *Tuta absoluta* eggs is demonstrated by *Trichogramma achaeae*. The highest effectiveness rate is achieved by treatment with a tank mixture of Coragen and Actarophyt-91%.

As a result of the activities performed using the integrated system for the protection of tomato plants, the Musa farm collected 18 tons of tomatoes from 0.3 ha of land in 2021. Meanwhile, the Sapar and Yultuz farms collected 8 and 9 tons of produce, respectively, from the same area of land. The reason for the loss of yield in the Sapar and Yultuz farms was ill-timed protection against tomato pests, including *Tuta absoluta*, and the use of the same chemical pesticide over the entire vegetation period, which, in turn, may have caused resistance in the pest.

Discussion

The covert lifestyle of *Tuta absoluta* contributes to the rapid development of populations resistant to rotating insecticides. Such pest populations cause economic losses and the substitution of one agent with another (Braham and Hajji, 2012) note that insecticides remain an indispensable means of plant protection due to their effectiveness and ease of use. Nevertheless, our practice of control of *Tuta absoluta* demonstrates the feasibility of using integrated plant protection with the biological method of control.

In Kazakhstan, *Tuta absoluta* is the most dominant and harmful pest on tomatoes. The loss of yield to this pest reaches 90-100% in the southern part of the country. Furthermore, research indicates that *T. absoluta* is spreading to new territories in Kazakhstan. In the Almaty region, the phytophage was found to produce five generations in 2021.

Control over the development of *T. absoluta* can only be established with a complex protection system in place. The system of protection has to proceed from the biology of the specific pest and be embedded in the crop cultivation technology. In areas where *T. absoluta* is widespread and chemical agents are used, the high resistance of the species to them is noted. In Argentina, Brazil, Chile, and other countries, the effectiveness of some organophosphorus drugs, pyrethroids, etc. has deteriorated in recent years (Desneux *et al*., 2021). These countries use repeated treatment with insecticides, which accelerates the development of resistance to them in populations of the pest. However, the experience of Spanish vegetable farmers testifies that for now, Confidor, Spintor, and Match remain effective against *T. absoluta*. Preparations based on carbamates (Lannat) and pyrethroids (Decis) can also cause the rapid death of imago and caterpillars. Treatments with pesticides with different mechanisms of action are more effective.

Bacterial preparations have shown fairly reliable results in Spain (Jalapathi *et al*., 2020). Encouraging results were obtained when testing preparations based on entomopathogenic fungi.

Researchers from the Kuwait Institute for Scientific Research (Safat, Kuwait) compared the effectiveness of four biological insecticides, azadirachtin, *Bacillus thuringiensis*, *Steinernema feltiae*, and *Beauveria bassiana*, on their own and in various combinations in protecting tomato crops against *T. absoluta* in laboratory settings and greenhouses. When second instar larvae were released on tomato leaves treated with azadirachtin (3 g/L), *B. thuringiensis* (0.5 g/L), or *B. bassiana* (1.5 g/L), 70-86, 55-65, and 45.5-58.5% pest mortality was observed, respectively. The least effective was the use of *Steinernema feltiae*, which resulted in the death of only 26-42% of pest larvae (EastFruit, 2019).

Our study also demonstrates good results of the application of a biological preparation based on *Bacillus thuringiensis thuringiensis* (Bitoxybacillin) with biological effectiveness reaching 60%, as well as of treatment with Actarophyt (a complex of natural avermectins produced by a beneficial soil fungi *Streptomyces avermitilis*), in which case the biological efficiency was 75%.

In experiments conducted by L.I. Prishchepa, the effectiveness of *Trichogramma achaeae* in the protection of closed-soil tomato plants was 91% (Prishchepa and Voitka, 2013). In our study, the release of *Tr. achaeae* also resulted in a high effectiveness rate of 81%.

Thus, the conducted study reveals that the inclusion of biopesticides, entomophages, and pheromone traps in the integrated strategy of tomato plant protection against *T. absoluta* is an effective and promising endeavor.

Conclusion

Our study employs such methods of control of *Tuta absoluta* as biological preparations (Bitoxybacillin, Actarophyt), entomophages (*Trichogramma achaeae*, *Bracon hebetor Say*), and pheromone traps. The latter allows for prompt detection of the activity of *Tuta absoluta* and, therefore, makes it possible to carry out protective measures in time.

Concerning the results collected in 2021, it is important to note that treatment with biological agents and well-timed and repeated release of *Trichogramma achaeae* (against eggs) and *Bracon hebetor Say* (against older caterpillars) successfully controlled the population of the phitophage. It is also notable that pheromone traps prove to be a valuable tool for integrated control in combination with other measures. Thus, the primary elements of the integrated system for the protection of tomato crops against *Tuta absoluta* include the use of pheromone traps to detect the appearance of the moths and capture them en masse and the application of biological preparations, entomophages and, if necessary, low-hazard insecticides.

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Author’s Contributions

All authors equally contributed to this study.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues are involved.

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