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Microbial-based Production System: A Novel Approach for Plant Growth and Pest and Disease Management in Greenhouse-grown Peppers (*Capsicum annuum* L.)

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ABSTRACT

A fundamental shift to a total system approach for crop management in greenhouses is urgently needed to resolve escalating economic and environmental consequences of long-lasting undesired effects of synthetic pesticides used in combating agricultural pests. The aim of this study was to examine a potential new approach i.e. Microbial-based Production System (MPS) for greenhouse-grown peppers. For this purpose, a two-year experiment in greenhouse was carried out in southwestern Turkey (Antalya) in 2011 and 2012, and only microbial-based products were used to suppress and control invertebrate pests (insects, mites, nematodes, gastropods, etc.) and diseases. In addition, biostimulants, inoculants, and bioyield enhancers were used for plant growth, being supported with three macro elements (NPK: Nitrogen– Phosphorus–Potassium) that are considered to be essential elements for plant growth and development. A conventional plot, largely based on the use of synthetic chemical inputs, such as fertilizers and pesticides, was included as the control. The efficacy of the MPS was evaluated by monitoring the population development of the key arthropod pests, such as the cotton whitefly *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae), the western flower thrips *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) etc. and their natural enemies compared with that in a Conventional Production System (CPS). The results showed significantly lower numbers of the key pests, but higher numbers of natural enemies were seen in the MPS of greenhouse-grown peppers compared with the CPS throughout the study. Total yield was relatively higher in the CPS than the MPS in both experimental years.

Keywords: Arthropod pests, Biostimulants, **C**onventional production system, Microbial products.

INTRODUCTION

Turkey, due to its unique geographical location as well as the ecological advantages, is the homeland and product center of many of the horticultural crops (Agaoglu *et al*., 1997). During 2010, Turkey, with a total annual vegetable production of 25.8 million tons, ranked fourth in the world after China, USA and India (Anonymous, 2012). Approximately 5.8 million tons of the total annual fruit and vegetable production in the country is produced in greenhouses covering an area of 56,000 ha. The vast majority of greenhouse production in Turkey consists of tomato (3 million ton yr^{-1}), cucumber (1 million ton yr^{-1}), watermelon (720,000 ton yr^{-1}) and pepper (450,000 ton yr^{-1}) (TUIK, 2011).

Antalya (southwestern Turkey) has a huge potential in the vegetable production of

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Turkey and ranks top, especially in greenhouse production and in production for fresh consumption. Approximately 47% of the total greenhouse area of the country is situated within the boundaries of Antalya, and protected vegetable cultivation is an important part of this area. Within greenhouse-grown vegetables in the province, pepper (*Capsicum annuum* L.), both in terms of variety and planting area, has been increasing, and currently ranks third in terms of production quantity after tomato and cucumber. However, the acceleration achieved in pepper production has not fully realized the expected benefits, especially in terms of the exportation. Although pepper export of Turkey increased 14% compared to the previous year, it is about 2% of the total exports of fresh fruits and vegetables. According to the latest figures, only about 69 thousand tons of the total pepper production in 2011 could be exported (AKIB/MEU, 2012).

Effective pest and disease management is an integral part of any crop production system (Dhawan *et al*., 2009). In greenhouse pepper cultivation with the Conventional Production System (CPS) in Antalya, control of pests and diseases is based on intensive and excessive use of synthetic chemicals. For this reason, greenhouse-grown peppers are at the top of the ranking, of the crops detected with residues exceeding the Maximum Residue Level (MRL). According to European Food Safety Authority (EFSA), the analysis of the results of the 2009 EU-coordinated program has shown that 1.2% of the 10,553 samples exceeded the MRL, while 37.4% of samples had measurable residues above the analytical reporting level but below or at the MRL. The highest percentage of samples exceeding the MRL was identified for table grapes (2.8%), followed by peppers (1.8%) (EFSA, 2011). In 2011, Turkey was the third among the countries having the most residual risks declared by the Rapid Alert System for Food and Feed (RASFF) of the European Union (RASFF, 2011). Among the residual risks to the fresh vegetables exported by Turkey, pepper has been one of the most noticeable products, and the residual risks about fresh peppers constitute approximately 60% of all the notifications (RASFF, 2011). All of these indicate that the main obstacle to the export of pepper of Turkey to EU seems to be the residue problem.

In contemporary greenhouse production, alternative production systems to eradicate or minimize the long-lasting undesired effects of synthetic chemical pesticides are necessary. The present study aimed at developing and testing a new approach (Microbial-based Production System: MPS) for greenhouse-grown peppers. In this approach, only microbial-based products/by-products are used for plant protection and plant nutrition (except for three macro nutrients: NPK: Nitrogen–Phosphorus–Potassium) throughout the growing season, comparing with the CPS that is used by the majority of pepper growers in southwestern Turkey.

MATERIALS AND METHODS

Test Materials

Within the MPS evaluated in this study, only microbial-based products/materials were used to control pests and diseases. Besides, some microbial-based products such as biostimulants, inoculants, and bioyield promoters were used for plant growth. All the microbial products/materials used in the study were provided by Bioglobal Inc. (Antalya, Turkey), and the product details are summarized in Table 1.

The choice of the products of microbial origin for inclusion in this system was based on our previous field tests and studies, which indicated their effectiveness against target pests and diseases but were safe for non-target organisms including predaceous fauna and other beneficial arthropods (Erler *et al*., 2013; 2014). With the exception of three macro nutrients (NPK), no synthetic

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Table 1. Products/materials used in the Microbial-based Production System (MPS) throughout the study.[a]

Product/Material (Formulation type)	Active ingrad [ai l^{-1} (g)]	Company	Why use	Recommended dose (ml or g da^{-1} or 100 l-1 water)[b]	Application date	Application form
Oxy (Liquid)	Active Oxygene (14%) Ethane Peroxy acid (4)	Bioglobal Inc	Disinfectant (Used only before planting)	10 l da^{-1}	1 August 2010	Drip irrigation
EndoRootsSoluble® (WP)	Mycorrhizal fungi (23.5% *Glomus* spp. and *Gigasporamargarita* g^{-1})	Bioglobal Inc	Root stimulant	350 g 50 l^{-1} water	25 September and 5 October 2010[c]	Drip irrigation
MET 52® (EC)	*Metarhiziumanisopliae*strain F52 (5.5×10^{9}conidia ml^{-1})	Novozymes Biologicals	Bio-insecticide	100 ml 100 l^{-1} water	First, 18 October 2010 and then monthly intervals	Aerial spraying
Vitormone® (EC)	*Azotobacter* spp.	Bioglobal Inc	Bio-fertilizer	150 ml 100 l^{-1} water	First, 10 November 2010 and then monthly intervals	Aerial spraying
Milastin® (WP and EC)	*Bacillus subtilis*	KanBiosys Pvt Ltd	Bio-fungicide (Against fungal diseases)	15 g or ml 10 l^{-1} water	First, 15 November 2010 and then monthly intervals	Aerial spraying
Netisin Plus (WP)	Nematophagousfungi (*Arthrobotrys*spp., *Paecilomyceslilacinus*and *Verticillium*spp.	Bioglobal Inc	Bio-nematicide (Control of plantparasiticnematodes)	500 g da^{-1}	First, 30 September 2010 and then monthly intervals	Drip irrigation
VitormoneDrip (EC)	*Azotobacterchroococum*and *Azotobactervinelandii*	Bioglobal Inc	Liquid bio-fertilizerfor root growth	250 ml da^{-1}	First, 30 September 2010 and then monthly intervals	Drip irrigation
Combat Plus (WP)	Organic nitrogen (at least 30%)	Bioglobal Inc	Bio-organic fertilizer (For control of damping-off disease)	250 g da^{-1}	First, 30 September 2010 and then monthly intervals	Drip irrigation
Thuricide (WP)	*Bacillus thuringiensis* var. *kurstaki* (20%)	Sandoz Crop Protection	Bio-insecticide (For control of caterpillars)	100 g 100 l^{-1} water	25 September 2010	Aerial spraying
Biosaps (SC)	Cow manure and bio-gas waste plus PGPR (Plant Growth Promoting Rhizobacteria)	Bioglobal Inc	Pure bio-organic fertilizer	150 ml 100 l^{-1} water	First, 5 September 2010 and then monthly intervals	Drip irrigation
Indazal® (EC)	Azadrachtin (%)	Bioglobal Inc	Insecticide/Acaricide	180 ml 100 l^{-1} water	First, 1 October 2010 and then monthly intervals	Aerial spraying
BroadBand® (EC)	*Beauveria bassiana* (Min 4×10^{9} conidia ml^{-1})	Becker Underwood	Bio-insecticide /-Acaricide	150 ml 100 l^{-1} water	13 and 23 October 2010; 20 and 30 April 2011	Aerial spraying
SubtilexFoliar® (WP)	*Bacillus subtilis* MB 600	Bioglobal Inc	Bio-fungicide	50 g 100 l^{-1} water	10 November, 10 December 2010, 10 January, 10 February, and 10 March 2011	Aerial spraying

[a] All the products/materials used in the first year of the study were also applied in the second year of the study (almost in the same application times). [b] All the products/materials used in the study were applied at their recommended label rates. [c] The plants were transplanted into the greenhouse in single row in mid-September for both growing years.

Table1 continued…

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Continued of Table1.

Product/Material (Formulation type)	Active ingrad [ai l^{-1} (g)]	Company	Why use	Recommended dose (ml or g da^{-1} or 100 l-1 water)[b]	Application date	Application form
BiosapsFoliar® (EC)	Cow manure and bio-gas waste plus PGPR (Plant Growth Promoting Rhizobacteria)	Bioglobal Inc	Pure bio-organic fertilizer	100 ml 100 l^{-1} water	First, 1 November 2010 and then monthly intervals	Drip irrigation
Miller® (EC)	-	Bioglobal Inc	*Non-ionicspreader* sticker	25 ml 100 l^{-1} water	Used together with each of the aerial applications	Aerial spraying
KillDew Plus (EC)	Microbial by-product	Bioglobal Inc	Bio-fungicide (Against powdery and downy mildews)	125 ml 100 l^{-1} water	1 December 2010; 1 January, 1 February, 1 March, and 1 April 2011	Aerial spraying
Botanigard (SC)	Beauveria bassiana (2.11×10^{10}conidia ml^{-1})	Futureco Bioscience	Bio-insecticide (Especially, control of thrips)	175 ml 100 l^{-1} water	30 November and 30 December 2010; 15 January, 15 February, 15 March, and 15 April 2011	Aerial spraying
FinishFoliar (WP)	Microbial by-product	Bioglobal Inc	Bio-fungicide (For control of fungal diseases of foliage)	25 g 100 l^{-1} water	20 November, and 20 December 2010; 20 January, 20 February 20 March, and 20 April 2011	Aerial spraying
Finish M (WP)	Microbial by-product	Bioglobal Inc	Bio-insecticide (Especially, for control of thrips)	50 g 100 l^{-1} water	10 December 2010	Aerial spraying
FinishSoil (WP)	Microbial by-product	Bioglobal Inc	Bio-insecticide (Especially, for control of thrips in prepupal and pupal forms)	100 g 1500 m^{-2}	15 November 20110	Drip irrigation
Liquid Sulphur	-	Bioglobal Inc	Fungicide/Acaricide	200 cc/100 l water	22 January 2011	Aerial spraying
Bio-Sulphur (EC)	-	Bioglobal Inc	Fungicide/Acaricide	300 cc/100 l water	20 December 2010	Aerial spraying
BP (WP)	-	Futureco Bioscience	Fungicide	500 g/100 l water	12 February 2011	Aerial spraying
VegexFos (SC)	Soapfosforik 55%	Terralia Inc	Bio-insecticide (Control of aphids)	400 cc/100 l water	1 January and 20 April 2011	Aerial spraying
VegexKuneka (EC)	Plantsaps % (Neemextract, *Tymusserpyllum* and*Tymusv ulgaris*)	Terralia Inc	Bio-insecticide	300 cc/100 l water	25 November 2010; 10 January 2011	Aerial spraying
Chitinase (SC)	-	Bioglobal Inc	Control of spider mites	400 cc/ 100 l water	10 November 2010; 15 May 2011	Aerial spraying

[a] All the products/materials used in the first year of the study were also applied in the second year of the study (almost in the same application times). [b] All the products/materials used in the study were applied at their recommended label rates. [c] The plants were transplanted into the greenhouse in single row in mid-September for both growing years.

chemical inputs, such as fertilizers and pesticides, were used throughout the study. To further identify the effectiveness of MPS, it was compared with the CPS, in which synthetic chemicals were used throughout the study (Table 2). The CPS, largely based on the use of synthetic chemical inputs such as fertilizers and pesticides, is commonly used by the pepper growers in southwestern Turkey.

Experimental Site and Design

The study was carried out for 2 consecutive years, fall-winter growing period of 2010-2011 and 2011-2012, in a greenhouse (≈0.2 ha) located in the Campus of Akdeniz University (36° 53' N; 30° 39' E, altitude 39 m), Antalya, Turkey. The greenhouse was well ventilated by a 1-m high side opening with insect protecting net and forced ventilation by an electrically operated fan. The soil type of the greenhouse was sandy-loam, and the soil characteristics were as follows: pH: 7.9, Lime: 15.9%, Electric Conductivity (EC): 2.5 mmhos cm^{-1}, Organic matter: 1.44%.

After soil disinfection with solar energy (solarization), in which the soil was first wetted by drip irrigation and then covered with a transparent polyethylene fil m (30μ thickness) for 6 weeks before planting, the greenhouse was arranged as per layout of the experiment, by dividing it into two separate compartments for MPS and CPS, with clear polycarbonate sheet (Thickness: 12 mm, 100% Virgin Bayer Material, UV layer: One side UV protective layer) in order to prevent potential interference between the two production systems. Biologically- and conventionally-grown seedlings (for MPS and CPS, respectively) of three-weeks old pepper (*Capsicum annuum* L. cv. Urartu) from a local company (Fitar Seedling Inc., Antalya, Turkey) were transplanted into the greenhouse in single row in mid-September for both growing years. The plants spacing within each row was 0.5 m and between two rows was 1 m. The *cv.* Urartu, which is one of the most common kapia type pepper varieties grown in the region, was used in the study. The research was conducted using a randomized factorial experimental design.

Crop Management, Pest and Disease Control

After transplanting, the plants were watered using drip irrigation, applying 8 L of water per plant each week. As the plants grew, all lateral shoots were manually removed and poles were employed to support single stems. All other cultural practices (mulching, pruning etc.) were applied uniformly in both MPS and CPS compartments of the greenhouse during the growing period. In the present study, plant growth stimulating substances of microbial origin were used for plant nutrition only in the MPS compartment of the greenhouse (Table 1). The choice of these microbial-based materials in the study and the amounts used were based on our previous field tests. Since healthy plant production requires complimentary foliar applications of nutrients additional to soil applications, in this study, the plants were supported with foliar applications of some bio-fertilizers, such as Vitormone®, which includes cysts of *Azotobacter chroococcum* Beijerinck suspended in clay based liquid formulation with microbial metabolites that fix atmospheric nitrogen and secrete siderophores, vitamins, and organic acids to avoid any nutrient deficiency (Abdel Latef, 2013).

All the fertilizers including NPK were incorporated into the top 5 cm of soil.

During the application of fertilizers in both MPS and CPS compartments, pepper yield (expected ~70 tons per ha) and available nutrient levels in the soil were taken into consideration. The fertilization was applied at a rate of 200 kg ha^{-1} N, 100 kg ha^{-1} P_2O_5 and 200 kg ha^{-1} K_2O as a modification of Gunay (1992) and Vural *et al*. (2000).

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Table 2. Products/materials used in the Conventional Production System (CPS) throughout the study.[a]

Product/Material (Formulation type)	Active ingrad [ai l^{-1} (g)]	Company	Why use	Recommended dose (ml or g da^{-1} or 100 l^{-1} water)[b]	Application date	Application form
Previcur Energy SL 840	530 g l^{-1} Propamocarb+310 g l^{-1} Fosetyl	Bayer	Against Damping-off Diseases	400 ml da^{-1}	First, 30 September 2010/2011 and then monthly intervals[c]	Drip irrigation
Rimon Supro 10 SC	100 g l^{-1} Novaluron	Flogaz	Against Caterpillars	60 ml da^{-1}	25 September 2010	Aerial spraying
Hypnose 05 SG	5% Emamectin benzoate	Safa Tarım AŞ	Against Caterpillars	30 g da^{-1}	13 October 2010; 20 April 2011	Aerial spraying
Dicarsol 50 WP	500 g kg^{-1} Formetanate	AMC-TR	Against thrips	100 g 100 l^{-1} water	30 November and 30 December 2010; 15 January, 15 February, 15 March, and 15 April 2011	Aerial spraying
Woliam Targo 063 SC	45 g l^{-1} Chlorantraniliprole +18 g l^{-1} Abamectin	Syngenta	Against Cotton leafworm	80 ml 100 l^{-1} water	23 October 2010; 30 April 2011	Aerial spraying
Luna Experience SC 400	200 g l^{-1} Fluopyram+200 g l^{-1} Tebuconazole	Bayer	Against powdery mildew	30 ml 100 l^{-1}	1 December 2010; 1 February, and 1 April 2011	Aerial spraying
Teppeki	50% Flonicamid	Sumitoma	Against aphids	15 g 100 l^{-1} water	1 January and 20 April 2011	Aerial spraying
Emarebeno 02 EC	20 g l^{-1} Emamectin benzoate	Koruma Klor	Against Cotton leafworm	70 ml 100 l^{-1} water	25 November 2010; 10 January 2011	Aerial spraying
Accolade EC	1000 g l^{-1} Dimethyl Disulfide	Cerexagri	Against Root-knot nematodes	45 l da^{-1}	15 August 2010	Drip irrigation
Aremon 10 SC	100 g l^{-1} Pyriproxyfen	SAFA	Against whiteflies	50 g 100 l^{-1} water	First, 1 October 2010 and then monthly intervals	Aerial spraying
Article	100 g l^{-1} Kresoxim methyl+ 200 g l^{-1} Boscalid	Hektaş	Against powdery mildew	50 ml 100 l^{-1} water	1 January, 1 March 2011	Aerial spraying
Agrimek	18% Abamectin	Syngenta	Against mites	30 ml 100 l^{-1} water	First, 13 October 2010/2011 and then monthly intervals	Aerial spraying

[a] All the products/materials used in the first year of the study were also applied in the second year of the study (almost in the same application times). [b] All the products/materials used in the study were applied at their recommended label rates. [c] The plants were transplanted into the greenhouse in single row in mid-September for both growing years.

During the winter months, a heating system (a diesel burner heating system in which air distribution is provided by means of perforated polyethylene ducts at ground level) was installed in the greenhouse to maintain optimum growing temperatures at around 15°C during the cool days and cold nights, because temperatures lower than the optimum would alter plant metabolic systems to slow growth and hinder fruit set. In addition, InfraRed (IR)+AntiFog (AF)-added heat seal polyethylene film (with high transparency) was used with the aim of contributing to the greenhouse heating. During nights and cold days, IR additive decreases heat loss by 2-5°C by keeping the heat inside the greenhouse. As for AF additive, it provides the moisture condensed on the film surface forming uniform water layer and this prevents the water droplets falling on the plants.

At the beginning of each growing season, the key pests and diseases affecting greenhouse pepper cultivation in the Antalya district were listed, and each was matched at least with one bio-pesticide (Table 1). Periodic surveys were carried out throughout the growing season for the detection of the presence or absence of a key pest and disease.

Data Collection and Statistical Analysis

The comparative evaluation of MPS and CPS was made by sampling the populations of key pests, namely, the cotton whitefly *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae), the thrips [the western flower thrips *Frankliniella occidentalis* (Pergande) and the onion thrips *Thrips tabaci* Lindeman (Thysanoptera: Thripidae)], the leafminers *Liriomyza* spp. (Diptera: Agromyzidae), the aphids [the cotton aphid, *Aphis gossypii* Glover and the green peach aphid, *Myzus persicae* (Sulzer) (Homoptera: Aphididae)] and the mites [the carmine spider mite *Tetranychus cinnabarinus* (Boisduval) (Acarina: Tetranychidae) and the broad mite *Polyphagotarsonemus latus* (Banks) (Acari: Tarsonemidae)].

The adult populations of cotton whitefly, leafminers and aphids were sampled by yellow sticky traps (dimensions: 20 W×25 L cm) at bi-weekly periods throughout the growing season (from the beginning of October to the end of February). The immature forms of cotton whitefly were sampled from leaf samples taken at weekly intervals. Leaf sampling method was also used to detect the presence or absence of other pests or diseases, such as powdery and downy mildews (Lapchin and Shtienberg, 1999). Thrips populations were sampled using blue sticky traps (dimensions: 10 W×25 L cm) with pheromones capsules (ThripLine® ams; Syngenta-Bioline, Antalya, Turkey) and flower sampling (Greer and Diver, 1999; Lapchin and Shtienberg, 1999; Natwick *et al*., 2007). Traps were renewed biweekly, and the capsules monthly. Flowers of each sampling unit (10 plants per treatment) were taken, i.e. a pepper plant was randomly selected and three fully opened flowers were collected. Every single pepper plant that was used for sampling was marked in order not to serve for the following week's sampling. Each sampled flower was placed in a plastic bag on which the number of sampling units (30 flowers from each treatment) and strata had been marked.

Spider mite populations in both MPS and CPS compartments of the greenhouse were monitored by sampling randomly the newly expanded and mature foliage. In random sampling, leaf samples were taken weekly throughout the study, and all life stages (eggs, larvae, nymphs and adults) of the mite were counted from leaf samples. Since the broad mite was found in association with apical leaf-curling symptoms on peppers (Coss-Romero and Peña, 1998), infested plants were inspected weekly, and changes in broad mite population and plant damage over time were measured from leaf samples by counting the broad mites per leaf.

At each leaf sampling, 60 young/fully-expanded leaves were randomly collected

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from 20 plants (by selecting three leaves from the top, middle, and bottom strata) per compartment (MPS or CPS) and examined under a stereo-microscope in the laboratory (Lapchin and Shtienberg, 1999). In trap sampling, sticky traps were uniformly hung in both compartments at the rate of 1 trap 20 m^{-2} (50 traps in each compartment) (Greer and Diver, 1999; Natwick *et al*., 2007; Atakan and Bayram, 2011). The traps were placed each growing season shortly after transplanting and were hung by a rope 10 cm above the plants according to plant heights when the plants were about 30-60 cm tall. At each sampling, the traps were replaced with fresh traps and were transported in special wooden cases to avoid adherence to each other or other surfaces. In the laboratory, adults of the pests mentioned above as well as their natural enemies (predators and parasitoids) were counted with the help of a head-band magnifier. Both sides of each trap were counted carefully. The sampling program in both 2010-2011 and 2011-2012 fall-winter growing periods was initiated at the beginning of October and continued to the end of February.

For disease management, applications were generally intended for the management of Powdery mildew caused by an obligate fungal pathogen, *Leveillula taurica* (Lev.) Arn. and root-crown rot disease caused an oomycete plant pathogen, *Phytophthora capsici* Leonian since they are the most common and destructive diseases of peppers in the study area. Disease ratings pertaining to both MPS and CPS were calculated using the following formula (Chaube and Pundhir, 2009):

Disease incidence (%) = $(N_{dp} \times 100)/N_t$

Where, N_{dp}= No. of diseased plants (whole plant or plant part) and N_t= Total no. of plants or plant parts examined.

Furthermore, 60 plants were marked randomly from each production system to determine yield per plant, total yield, and yield from each harvest. We used a total of 4,000 plants in MPS and CPS to observe the aforementioned parameters. We harvested the peppers when they reached commercial harvest maturity stage at approximately 25-30 days intervals during the entire growing season.

The data pertaining to yield and incidence of some important diseases attacking peppers in both MPS and CPS during both growing periods were analyzed using the Statistical Analysis System software program, version 9.0 (SAS Inst., Cary, NC, USA) and treatments means were statistically compared using LSD's multiple range test ($P \leq 0.05$).

RESULTS

The population development of the key pests mentioned above and their natural enemies in both MPS and CPS during the two sampling growing periods is presented in Figures 1-3. Only sticky trap (yellow or blue) captures for the adults of *B. tabaci*, *Liriomyza* spp., *A. gossypii* and *M. persicae*, *F. occidentalis* and *T. tabaci* as well as their natural enemies (predators and parasitoids), and leaf sampling counts for the immature stages of insect pests and all life stages of the mite pests are presented here.

In both growing season, adult flights of *B. tabaci* and the thrips (*F. occidentalis* and *T. tabaci*) began at the beginning of October, reached a maximum at the beginning or in the middle of December, and increased continuously from mid-January to the end of February (Figure 1). The flight activity of *Liriomyza* spp. and the aphids (*A. gossypii* and *M. persicae*) began towards the end of October or at the beginning of November in both growing seasons, and reached a

maximum after the end of January. When comparing sticky trap captures in both growing seasons, the weekly mean numbers of adults of key pests caught in MPS were generally less than those caught in CPS. However, numbers varied considerably from year to year, and more substantial treatment effects were observed in the first year of the study (Figure 1).

As to the leaf sampling counts, MPS had lower numbers of sampled key pests

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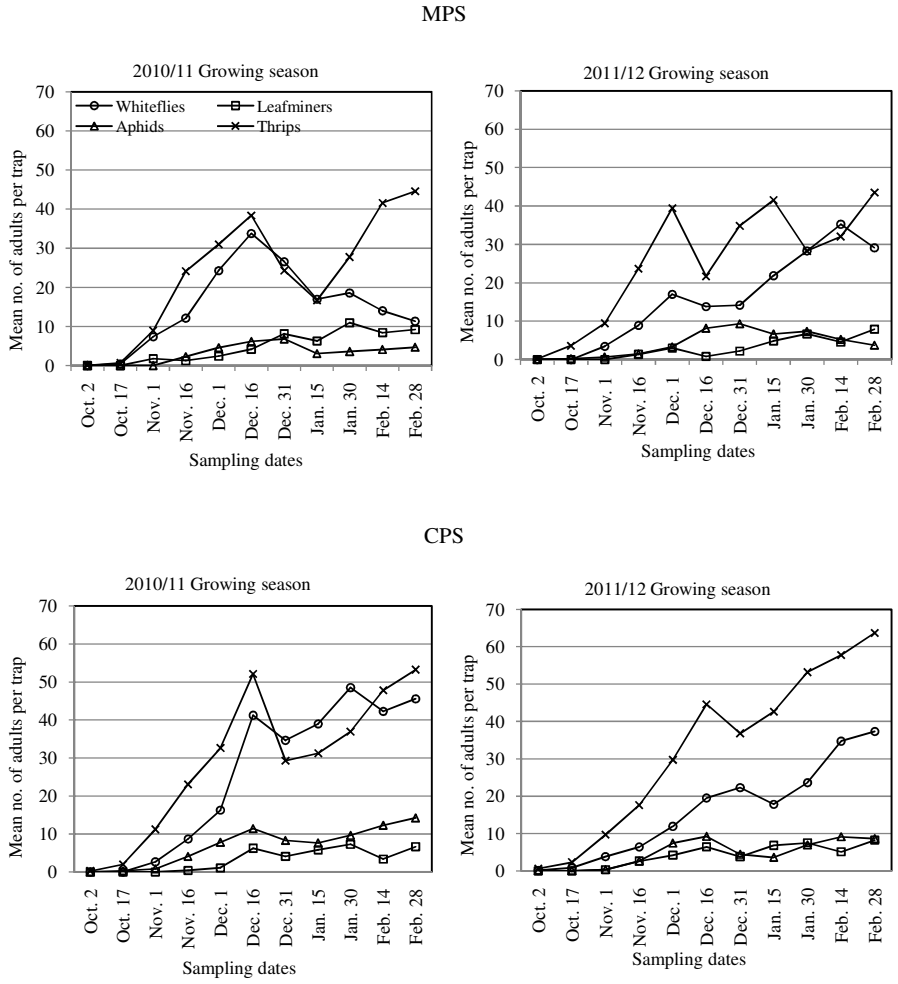

Figure 1. Population dynamics of key pests, the cotton whitefly (*Bemisia tabaci*), the thrips (*Frankliniella occidentalis* and *Thrips tabaci*), the leafminers (*Liriomyza* spp.), and the aphids (*Aphis gossypii* and *Myzus persicae*) caught in sticky traps in the Microbial-based Production System (MPS) and the Conventional Production System (CPS) throughout the 2010/2011 and 2011/2012 growing seasons.

compared with CPS (Figure 2). Whereas weekly mean numbers of sampled pests varied considerably from year to year, higher numbers of pests (except for spider mites) were observed in both production systems in the first year of the study.

The abundance of various groups of natural enemies sampled each growing season in both MPS and CPS is presented in Figure 3. The parasitoid complex including primarily parasitic wasps, *Aphidius colemani* Viereck and *A. ervi* (Haliday) (Hymenoptera: Braconidae) was the most abundant group of natural enemies in both growing seasons. However, higher numbers of parasitoids were caught on the sticky traps in MPS compared with those in CPS in both years. *A. colemani* was the only dominant parasitoid encountered in both production systems in both growing seasons. The three most abundant predator groups in either production system were the heteropterans, the chrysopids (Neuroptera) and the coccinellids (Coleoptera). Other groups of predators (for example, the

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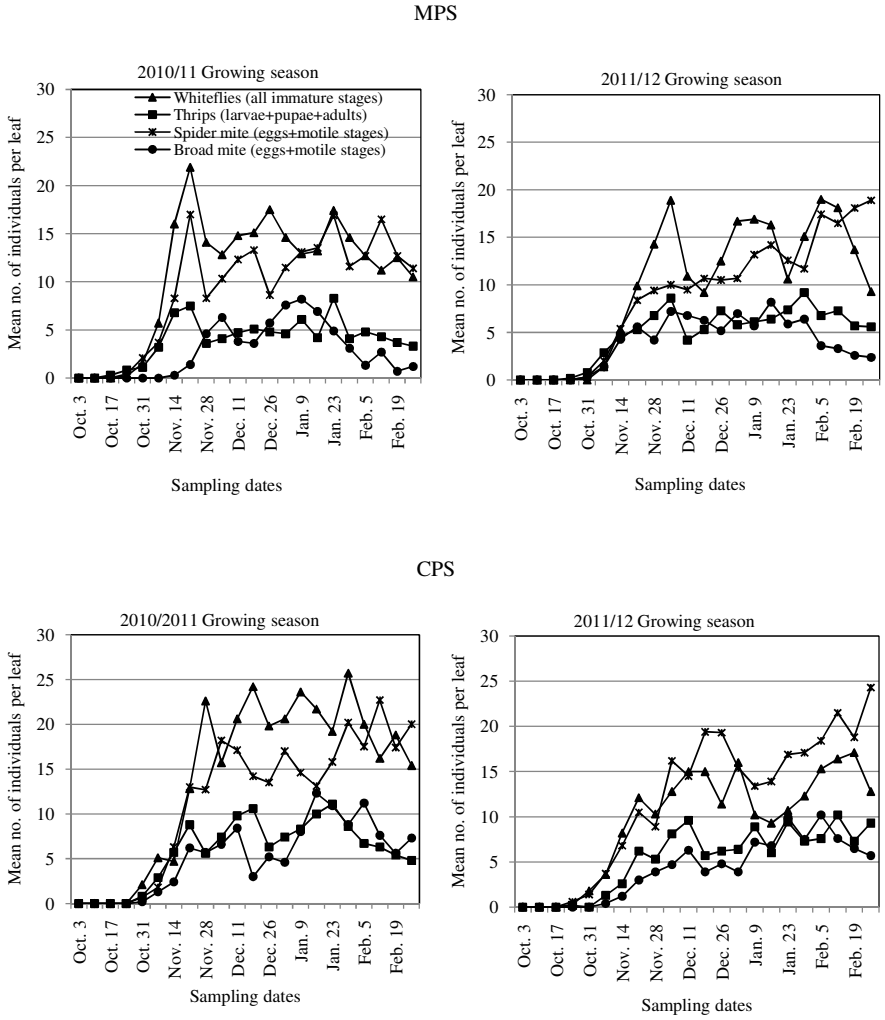

Figure 2. Population development of key pests, the cotton whitefly (*Bemisia tabaci*), the thrips (*Frankliniella occidentalis* and *Thrips tabaci*), the carmine spider mite (*Tetranychus cinnabarinus*) and the broad mite (*Polyphagotarsonemus latus*) from leaf samples taken weekly throughout the study in the Microbial-based Production System (MPS) and the Conventional Production System (CPS) throughout the 2010/2011 and 2011/2012 growing seasons.

dipterans) were rarely observed in sticky trap captures in both production systems during the sampling period. In leaf sampling, in addition to these three predator groups of insects, predatory mites were the most common predators found during the study. The dipterans were rarely observed in both production systems during the sampling period each year. When both production systems were compared in terms of total numbers of natural enemies sampled during the study, MPS had higher numbers of sampled natural enemies compared with CPS (Figure 3).

It was apparent from the results that the products/materials used in both MPS and CPS did not fully prevent the damage caused by powdery mildew and root-crown rot disease. Nevertheless, lower disease ratings were obtained from the MPS in both

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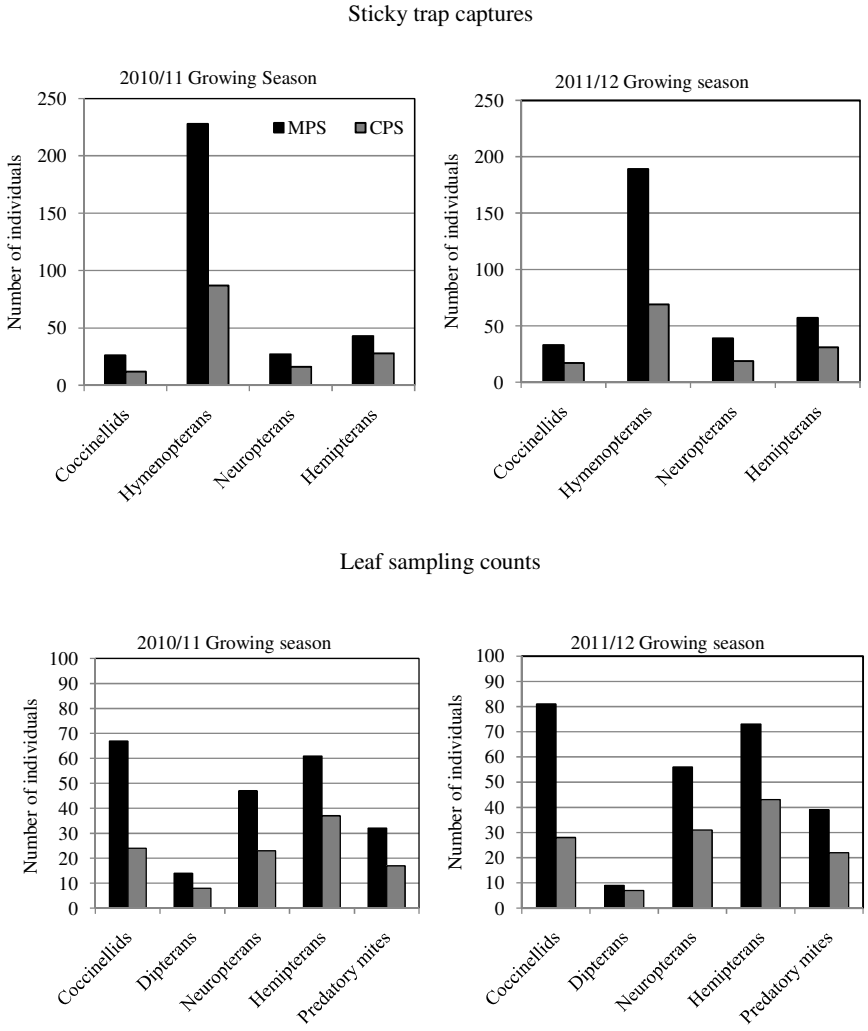

Figure 3. The abundance of natural enemies of key pests caught in sticky traps and leaf samples in the Microbial-based Production System (MPS) and the Conventional Production System (CPS) throughout the 2010/11 and 2011/12 growing seasons (black and gray bars indicate yearly total numbers).

growing periods compared with the CPS (Table 3). Other diseases attacking peppers, such as gray mold caused by the fungus, *Botrytis cinerea* (de Bary) Whetzel and bacterial soft rot caused by the bacteria belonging to at least five genera of bacteria including species of *Erwinia, Pseudomonas, Bacillus, Xanthomonas* and *Cytophaga,* were present only in trace amounts in both production systems throughout the study.

Yield per plant for both production systems is presented in Table 4. The effects of both production systems and growing season on yield per plant were found statistically significant ($P \leq 0.05$). The mean yields per plant were 3.69 kg and 3.84 kg in 2010-2011 and 2011-2012 growing season, respectively. The mean yield per plant was higher in CPS (3.90 kg) compared to MPS (3.63 kg). In terms of interactions of production system and growing season, the lowest yield per plant was 3.53 kg in 2010-2011 growing season at MPS and the highest average yield per plant was 3.95 kg in 2011-2012 growing season at CPS.

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A comparison of the total yield between the MPS and CPS found them different (at P≤ 0.05) in both production systems (Table 4). The highest total yield from the CPS was measured in the second year of the experiment (7906.70 kg decare^{-1}). Analyzing the impact of the factor of production system, the total yield was higher in CPS (7,793.35 kg) compared to MPS (7,223.35 kg decare^{-1}) (at P≤ 0.05). The interaction of major factors, i.e. growing season and production system, had a significant impact on total yield, i.e. the least total yield was measured in the first growing season in the MPS (7,060.00 kg decare^{-1}) and the highest total yield was measured in the second year in the CPS (7,906.70 kg decare^{-1}). There were no statistical differences (P> 0.05) between the two growing seasons in CPS.

DISCUSSION

The results from the present study showed that MPS may be a viable alternative to the CPS that is commonly used for growing pepper indoors in southwestern Turkey. According to the results, microbial treatments in both growing periods were more successful in suppressing populations of key pests compared with conventional ones (Figures 1 and 2). Moreover, higher number of predators and parasitoids were observed in MPS compared to CPS throughout the study (Figure 3). This is in conjunction with a number of reports (Roy and Pell, 2000; Lacey *et al*., 2001; Goettel *et al*., 2010) showing that the use of microbial-based products including entomopathogenic fungi is an important practice which could complement other control practices involving some predators and parasitoids in reducing the populations of pest insects and mites in economically important crops. Concerning the disease incidence, the products/materials used in both MPS and CPS did not fully prevent the damage caused by powdery mildew and root-crown rot disease. However, lower disease ratings were obtained from the MPS in both growing periods compared with the CPS (Table 3). In addition, residue analysis results of pepper samples taken periodically from both production systems showed no residues in samples from MPS in both growing periods, but some pesticide residues were present in samples from CPS (the results of residue analyses are not presented here).

There are some principal factors affecting growth, sporulation, infectivity and survival of entomopathogenic fungi, and these are: temperature, Relative Humidity (RH) and solar radiation (Vidal and Fargues, 2007). Principal factor limiting residual activity of entomopathogenic fungi is inactivation of conidia by ultraviolet radiation, therefore,

Table 3. The incidence rates of some important diseases attacking peppers in both Microbial-based Production System (MPS) and Conventional Production System (CPS) during the two growing periods.[a]

Disease and (its agent)	Disease incidence (%)	
	MPS	CPS
	2010/2011 Growing period	
Powdery mildew (*Leveillula taurica*)	33.8a	41.7b
Root-crown rot disease (*Phytophthora capsici*)	1.3a	2.2b
Pepper soft rot (Bacteria belonging to *Erwinia*, *Pseudomonas* etc.)	0.16a	0.3b
	2011/2012 Growing period	
Powdery mildew (*Leveillula taurica*)	38.2a	46.3b
Root-crown rot disease (*Phytophthora capsici*)	1.6a	1.8a
Pepper soft rot (Bacteria belonging to *Erwinia, Pseudomonas* etc.)	0.21a	0.6b

[a] For each of the two production systems tested, means within a line followed by the same lower-case letter are not significantly different (LSD, P≤ 0.05).

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Table 4. Effect of the Microbial-based Production System (MPS) and Conventional Production System (CPS) on total yield and yield per plant in protected pepper growing.

Yield	Production system	Years [a]		Means [b] (Production system)
		2011	2012	
Yield per plant (kg)	MPS	3.53d	3.73c	3.63b[b]
	CPS	3.84b	3.95a	3.90a
	Means (Years)	3.69b	3.84a	
$LSD_{\%5\ year}$: 0.0709	$LSD_{\%5\ year\times prod.\ sys.}$: 0.1002	$LSD_{\%5\ production\ sys.}$: 0.0709		
Total yield per decare (kg)	MPS	7060.00c	7386.70b	7223.35b
	CPS	7680.00a	7906.70a	7793.35a
	Means (Years)	7370.00b	7646.70a	
$LSD_{\%5\ year}$: 171.88	$LSD_{\%5\ year\times prod.\ sys.}$: 243.07	$LSD_{\%5\ production\ sys.}$: 171.88		

[a] According to LSD test indicating that interactions marked with different lower-case letters differ significantly at P≤ 0.05. [b] Means with different letters according to LSD test statistically different from each other (P≤ 0.05).

the spray applications in both growing periods were made during sunset. High humidity (≥ 80%) after application and optimal daytime air temperature are essential factors in efficacy of the microbial products, especially entomopathogenic fungi (Vidal and Fargues, 2007; Bugeme *et al*., 2008). The region including the study area has a Mediterranean climate, characterized by warm to hot, dry summers and mild to cool, wet winters. Temperature increases gradually during the spring months while humidity decreases. Greenhouse air temperatures increase with time up to 40°C during the late spring period. When temperature inside the greenhouse gets too hot, i.e. 38°C, it exceeds the optimum growing temperature in the range of 18-30°C for fungal products tested and, therefore, the effectiveness of fungal products sharply decreases.

Another important point is that some pests, such as spider mites, broad mite etc., usually feed on the underside of leaves, and all life stages can usually be found there, too. Therefore, spraying technology that better targets the underside of leaves would provide better coverage of the targeted pest and could prolong the viability of conidia and result in an increased interval between applications. Alternatives to conventional spraying, such as ultra low volume application and the use of electrostatic devices, could also be developed for improved targeting of pests.

Based on the results of this study, it was concluded that MPS could be useful for growing peppers indoors by removing residual risks associated with this crop. The use of MPS, in conjunction with good agricultural practices (good hygiene, preservation of biological control agents, good cultural practices including weed sanitation, adequate nutrition and irrigation etc.), may reduce the need of synthetic chemical inputs.

Since pests and diseases spread very rapidly in the warmth of a greenhouse, a careful watch should be kept for the first signs of their presence. If this is properly done, many of them can be controlled effectively by using bio-pesticides. Early identification, correct diagnosis, and the swift implementation of preventative methods are very essential to manage potential problems, and allow us to get on top of most problems before serious damage. For this reason, we carefully checked the greenhouse on a daily basis.

This study showed that CPS generally resulted in higher total yield than MPS. Although MPS showed lower yields, it has a number of advantages such as easy application, production of healthy (non-contaminated) food, maintaining environmentally-friendly production

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system, helping the natural cycle, and generating high income for the growers with more export opportunities. The other systems have equal to slightly lower yields in a range of crops than CPSs, but organic and integrated systems generally have greater economic value, environmental sustainability and energy efficiency (Smolik *et al*., 1995; Drinkwater *et al*., 1998; Reganold *et al*., 2001; Mäder *et al*., 2002; Porter *et al*., 2003; Peck, 2004). Producers have difficulty about both yield and market, and they cannot sell the organic crops with the price they want. There are some difficulties about organic agriculture in Turkey due to low market demand and high input (Bayram *et al*., 2007). In the light of the obtained results in the present study, it could be suggested that MPS may be recommended as a transition model and sustainable agricultural practices for the future. In order to have Turkish horticultural crops better accepted at both national and international levels, there should be the right production system management for farmers. In order to obtain peppers which are safe to consumers and for export promotion, we should establish a Good Agricultural Practices Standard for peppers and other crops.

We believe that this novel non-pesticide approach is healthier and environmentally-friendly production systems compared to the Conventional Production System (CPS) and may also be applicable to the other crops grown both indoor and open field conditions.

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سامانه تولید مبتنی بر میکروب: روشی نوین برای رشد گیاهان و مدیریت امراض و آفات در تولید فلفل گلخانه ای (*Capsicum annuum* L.)

ا. دوگان، ف. ارلر، م. ارکان، ا. اوزگور آتس، ه. سول سابانسی، و ا. پولات

چکیده

امروزه چرخشی اساسی به سوی سامانه ای فراگیر برای مدیریت گیاه در گلخانه به فوریت مورد نیاز است تا بتوان پیآمدهای اقتصادی و زیست محیطی نا خواسته و دراز مدت آفتکش های شیمیایی را که در مبارزه با آفات کشاورزی به کار می روند بر طرف کرد. هدف پژوهش حاضر بررسی یک روش تازه به نام سامانه تولید مبتنی بر میکروب (MPS)برای تولید فلفل گلخانه ای بود. به این منظور، آزمایشی گلخانه ای و دو ساله طی سال های ۲۰۱۱ و۲۰۱۲ در جنوب غربی ترکیه (منطقه آنتالیا) اجرا شد و در آن فقط مواد مبتنی بر تولیدات میکروبی برای کنترل و مبارزه با بی مهرگان (حشرات، کنه، نماتد، شکم پایان، و غیره) و آفت ها به کار رفت. افزون بر این، در این روش محرک های زیستی، مایه تلقیح، و مواد زیستی افزاینده عملکرد(bioyield enhancers) به همراه سه عنصر غذایی پر مصرف(نیتروژن، فسفر، پتاسیم) که برای رشد و تکامل گیاه ضروری هستند مصرف شدند. به عنوان تیمار شاهد، یک کرت به روش رایج (CP) مبتنی بر مصرف نهاده های شیمیایی مانند کود شیمیایی و آفتکش ها اختصاص یافت.. برای تعیین موثر بودن روش MPS، تحولات جمعیت بند پایان اصلی مانند سفید بالک پنبه(Homoptera: Aleyrodidae) (Gennadius) *Bemisia tabaci*، تریپس گل غربی (Thysanoptera: Thripidae) (Pergande) *Frankliniella occidentalis*و غیره، همراه با دشمنان طبیعی آن ها در روش MPS پایش شد و با روش تولید رایج (CP) مقایسه شد. نتایج نشان داد که در طول مدت آزمایش تعداد آفت های اصلی در فلفل گلخانه ای در کرت MPS به طور معنی داری کمتر از روش CP بود در حالی که دشمنان طبیعی آفت ها در روش MPSبیشتر بود. گفتنی است که در هر دو سال آزمایش،عملکرد کل درCP نسبتا بیشتر از MPS بود .

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