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Effects of various agricultural practices on persistence of the inundative applied entomopathogenic nematodes, *Heterorhabditis bacteriophora* and *Steinernema feltiae* in the field

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Summary. Effects of different agricultural practices such as fertilization (organic and NPK), irrigation (dripping and sprinkling), plough tilling and use of the herbicide, Trifluralin EC were tested on two different Turkish isolates of the entomopathogenic nematodes, *Steinernema feltiae* (Filipjev, 1934) (Rhabditida: Steinernematidae) and *Heterorhabditis bacteriophora* (Poinar, 1976) (Rhabditida: Heterorhabditidae) in the field condition. Experiments were conducted between March and November, and replicated in the following 2 years, 2005 and 2006 in Ankara. As all practices together affected negatively only *H. bacteriophora* in the second year, *S. feltiae* was not affected by these practices together in both years. *S. feltiae* persisted longer than *H. bacteriophora* in the years. Based on the lack of detection, the shortest persistence was found in the herbicide treated area and tilled plot area in the two years for *S. feltiae* and *H. bacteriophora*, respectively.

Key words: Agricultural practices, entomopathogenic nematodes, *Heterorhabditis bacteriophora*, persistence, *Steinernema feltiae*.

Efficacy of applied entompathogenic nematodes (EPNs) is closely related to the selected strains or species and also environmental and technical conditions as well. These characteristics directly influence the survival of the nematodes in a period of time without a host and their ability to find and infect a host (Womersley, 1993).

Several factors such as soil type (Kung *et al.*, 1990a), humidity levels in soil (Kung *et al.*, 1991), temperature (Griffin, 1993; Grewal *et al.*, 1994) and soil pH (Kung *et al.*, 1990b) affect the persistence and infectivity of EPNs in the soil. Additionally, understanding of EPN population dynamics over the seasons and their possible synchronisation with host life cycles is also very limited (Smits, 1996). To this end, most of the investigations on the persistence of EPNs have shown a strong relationship with the soil environment. However, there are a few studies on the effect of vegetation and agricultural practises on EPNs (Susurluk, 2005). In the present study, plough tilling, dripping and sprinkle types of irrigation, organic and NPK fertilization and use of a selected herbicide, Trifluralin EC were tested on the persistence of two inundative released Turkish EPNs, *Steinernema feltiae* (Filipjev, 1934) and *Heterorhabditis bacteriophora* (Poinar, 1976) in a two-years period.

MATERIAL AND METHODS

Experiment scheme. Experiments were conducted between March and November in the two sides of the same field in 2005 and 2006 separately. Region of the experimental field in Etimesgut-Ankara has an average altitude of 908 m.

Nematode isolates, propagation and storage. The EPN species used, *S. feltiae* and *H. bacteriophora*, were isolated from the soil in Ankara-Turkey and identified by PCR-RFLP (Susurluk *et al.*, 2001). The nematodes were reared on the last-instar wax moth, *Galleria melonella* L. (Lepidoptera: Galleriidae) at 25°C, according to Wiesner (1993). After harvesting, the nematodes were stored at 5°C in Ringer solution (laboratory standard) containing NaCl 9 g, KCl 0.42 g, $CaCl_2$ x $2H_2O$ 0.37 g, $NaHCO_3$ 0.2 g and aqua dest 1000 ml for 2 weeks before applications.

Preparation of the experimental fields. A field that had not been cultivated for 3 years was selected for the experiments in Etimesgut, Ankara. The treatments were arranged in 7 adjacent plots

(2 x 10 m) in the field. Experimental design was composed of the following agricultural practices: Plough tilling, organic and NPK fertilization, dripping- and sprinkle-type watering, herbicide treatment and a control free of any treatment were placed side by side with 1 m intervals. Additionally, soil samples from each plot were chemically analyzed by Sugar Institute in Etimesgut. The analyses indicated that, each plot had the same soil pattern for 2005 and 2006. Precipitation and soil temperatures were weekly measured and recorded.

Fertilization. The most commonly composed and organic fertilizers were selected in the present study. Two plots were used for the fertilizations. While one of the plots was treated with fertilizer containing nitrogen (N), phosphorus (P) and potassium (K), the other one was treated with cow manure as an organic fertilizer. Composed fertilizer consists of 20% total nitrogen (5.5% nitric nitrogen, 8% ammoniacal nitrogen and 6.5% ureic nitrogen), 20% phosphorus pentoxide (P_2O_5), 20% potassium oxide (K_2O), and also 0.05% boron (B), 0.001% copper (Cu), 0.20% iron (Fe), 0.10% manganese (Mn), 0.005% molybdenum (Mo) and 0.01% zinc (Zn). Organic fertilizer-cow manure- consists of 2% total N, 1.7% P_2O_5, 4% K_2O, and 0.001% B, 0.001% Cu, 0.006% Mn, 0.0006% Mo, 0.003% Zn and 0.0001% Cobalt (Co). Composed NPK and organic cow fertilizers were applied at doses of 15 and 3000 kg per acre, respectively (Anonymous, 2006). After fertilization, each plot was tilled very slightly in order to mix manures and soil.

Irrigation. Two plots that are not placed side-by-side were irrigated with two different techniques; dripping and sprinkling. Dripping was done with tiny pipes (T-Type) and 2 l water m^{-2} was applied to both plots. However, the water in sprinkling method was performed from 3 m height onto the soil. Irrigation was monthly repeated for both plots.

Tillage. One plot related to plough-tilling study was gently tilled for a depth of 20 cm by using of a tractor, which has the plough system. The tilled plot was then treated with disc harrow.

Herbicide application. The herbicide Trifluralin EC 480 g l^{-1}, which is commonly used in cereal fields at pre-emergence, was selected. The herbicide was applied at a dose of 200 ml per acre (=0.2 ml in 100 ml water m^{-2}) in 100 l water. Two rods of each 1.5 m of herbicide sprayer are equipped with three spraying nozzles (0.5 mm diameter) in a distance of 50 cm. The applications were performed using 3-5 bar pressure at a velocity of 1 m s^{-1}.

Nematode application. The applications of *S. feltiae* and *H. bacteriophora* were performed following day after all agricultural practices were done, except tillage. In order to determine effectiveness of tillage, the nematodes were applied one day before tilling. In control plot, only EPNs were applied, but no agricultural practices were performed. Since the general recommendation for commercial application dose is 0.5 million DJs m^{-2} (infective or dauer = enduring juvenile), (=50 DJs cm^{-2}) (e-nema GmbH), each plot was treated with a dose of $0.5x10^6$ DJs/1.2 l water. The application was done by using the sprayer described above, however, a separate sprayer at 4-5 bars. Prior to the applications, tap water and EPN formulations were gently mixed in a plastic barrel and then the EPN solution was transferred into the spraying tanks. To prevent sedimentation of the nematodes in the tank, the tanks were well shaken before the application.

Check of viability rate and pathogenicity of the EPNs before and after spraying. During the application, the number of EPNs per cm^{-2} was assessed by placing Petri dishes in the field and the number of living nematodes was subsequently counted in the laboratory. In order to obtain the number of applied DJs per area immediately after application, 10 plastic Petri dishes (9 cm diameter) were placed in 1 m intervals onto the soil surface in the experimental plots. After application, the Petri dishes were collected and the nematode suspensions were rinsed into plastic tubes with Ringer solution. Then, the number of DJs per tube was counted in the laboratory to calculate the amount of DJs applied per cm^2 soil. This method was conducted during each plot application. To compare the fitness of EPNs before and after spraying, nematode performance was tested in a bioassay with *G. mellonella*. Nine cm diameter Petri dishes were filled with moist silver sand (10% w/v) with a 0.1-0.5 mm particle size, which had been sterilized at 80°C for 12 h. Five last instar *G. mellonella* larvae were transferred into the dish and 100 DJs per insect were added. The experiments were done in 10 replicates.

Soil sampling and insect baiting. Soil samples were collected at 1 m intervals from each plot. Ten soil samples were collected per plot; with 3 replications for each plot. The procedure of the soil sample collection was also performed prior to the EPN application, in order to detect possible endemic EPN species in the experimental field. Collected soil samples before EPNs application in the parcels including control were baited with *G. mellonella* larvae three times one week interval. Soil samples were collected every month after

EPN application. Each sample had approximately 120 cm^3 (1.78 cm diameter and 12 cm height) soil from an area of 10 cm^2. The samples were placed in a plastic bag and transported to the laboratory. The samples were kept at 4°C until analyze. The nematodes were extracted by using the greater wax moth, *G. mellonella*, bait technique according to Bedding & Akhurst (1975). Soil was homogenised by hand and each soil sample was placed in a 125 ml plastic box (5 x 5 x 5 cm) with 3 last instar larvae of *G. mellonella*. After the larvae had been added, the boxes were turned round and stored at 25±2°C. Three days later, dead larvae were dissected in Ringer solution and nematodes were counted using a stereomicroscope.

Data analysis. Mean ± SE of the recovered EPNs from each plot and each part of 50 cm sample from plots baiting with the *Galleria* larvae was calculated. Statistical differences of viability rate and pathogenicity of EPNs were analyzed by variance (ANOVA) followed by Least Significant Difference (LSD). The results showed significant differences at $P<0.05$ levels (Statistica, 1991). For the pathogenicty tests, total insect mortality was corrected using Abbott's formula (Abbott, 1925). Interactions between annual means of detected DJs for the plots treated by each practice and each nematode, for each year were compared by student-t test at $P<0.05$. The statistical analyse was performed by using the Xlstat Pro. 7.0.

RESULTS

Analysis of the soil and meteorological data in the experimental fields. It was determined that each plot had the same level soil character before nematode application. With respect to the results of the soil analyse, no major differences were found between the plots for both years (Table 1).

During the investigation, soil temperature and precipitation were measured in every week after application. As measured soil temperatures were from 12 to 29°C, % humidity of the soil varied 10 and 55% in 2005. Temperature and humidity in 2006 were between 10-27°C and 15-46%, respectively.

Assessment of the nematodes before and after application. To detect indigenous EPN populations, total 840 soil samples collected from parcels in March of the years of 2005 and 2006 were analysed three times by dissection of bait insect. However, no indigenous EPN was detected in the plots of the experimental field. Pathogenicity and viability of both nematode species were not significantly different ($p<0.05$) before and after application. Percent viability and pathogenicity before and after application in 2005 were 97% and 94% for *H. bacteriophora* and 98% and 96% for *S. feltiae*, respectively. Pathogenicity and viabilities before and after application in 2006 were 95% for *H. bacteriophora* and 98% for *S. feltiae*. There were no significant differences between first and second year on the viability and pathogenicity of the nematodes before and after application (Table 2). As a result, the application had a negligible impact on nematode viability and pathogenicity. Present results of viability and pathogenicity of the both nematodes before and after nematode application also indicated that the application system used in this study had no negative effect on the nematodes.

Persistence of the nematodes in the plots. The results indicated that *S. feltiae* persisted longer than *H. bacteriophora*. Tillage and herbicide application had a negative effect on both nematodes. In the herbicide-applied plot, DJs of *S. feltiae* and *H. bacteriophora* reached max 22 and 18 DJs cm^{-2},

Table 1. Results of the chemical analysis of soil samples collected from each experimental plot. Numbers indicated in the table show minimum and maximum values.

Location	Depth (cm)	Dried soil (g)	Hydrometer value g/l		Correction factor for 20°C		Sand (%)	Silt (%)	Clay (%)	Soil texture
			40 sec.	2 h.	40 sec.	2 h.				
Exper. area	15-20	50	30-32	19-20	32.45-34.16	21.25-22.52	31.68-32.98	23.28-24.05	45.04-46.11	Clay

Location	Water Saturation %	Water Saturated Soil pH	Total Salinity %	$CaCO_3$ %	Organic Content %	Nutrients (kg/ da)	
						(P_2O_5)	(K_2O)
Exper. area	75-77	7.12-7.25	0.14-0.16	26.79-26.83	3.87-4.35	15.98-17.85	156.85-159.12

Table 2. Viability rate and pathogenicity of the released nematodes recorded before and after application in 2005 and 2006.

Nematode species	Before appl. (%)	After appl. (%)	Viability rate (Before appl./ After appl.)	Before appl. (%)	After appl. (%)	Patho. rate (Before appl./ After appl.)
S. feltiae (2005)	91.5±3.1 ^{a}	89.9±2.7 ^{a}	0.98	86.8±3.7 ^{a}	83.6±3.8 ^{a}	0.96
S. feltiae (2006)	89.7±4.2 ^{a}	88.2±3.0 ^{a}	0.98	87.4±4.0 ^{a}	85.8±2.8 ^{a}	0.98
H. bacteriophora (2005)	85.9±4.8 ^{b}	83±6.4 ^{b}	0.97	84±3.9 ^{b}	79±3.5 ^{b}	0.94
H. bacteriophora (2006)	83.9±2.8 ^{b}	80±7.2 ^{b}	0.95	85±3.3 ^{b}	81.1±2.7 ^{b}	0.95

Note: Data present mean ± SE within the row (before and after application in viability or pathogenicity) followed by the same letters are not significantly different (P>0.05) according to the Least Significant Differences (LSD) test. Pathogenicity (%) was calculated considering mortality in the controls using Abbott's formula (Abbott, 1925). Each nematode was compared each other in two years.
Viability: ^{a} ANOVA: F=2.4124; df=3, 36; P=0.2383, ^{b} ANOVA F=1.9825; df=3, 36; P=0.2481.
Pathogenicity: ^{a} ANOVA: F=0.4509; df=3, 36; P=0.7270, ^{b} ANOVA: F=5.5489; df=3, 36; P=0.1698.

respectively. In the ploughed plot, 19 DJs of *S. feltiae* cm^{-2} and 12 DJs of *H. bacteriophora* cm^{-2} were recorded in the first year. However, in the second year, more DJs of *H. bacteriophora* were found in plots with either ploughed or herbicide treated. In opposite, no significant differences were detected for *S. feltiae* between the years (p=0.807) (Fig. 1, 2).

Persistence of the nematodes in the plots. The results indicated that *S. feltiae* persisted longer than *H. bacteriophora*. Tillage and herbicide application had a negative effect on both nematodes. In the herbicide-applied plot, DJs of *S. feltiae* and *H. bacteriophora* reached max 22 and 18 DJs cm^{-2}, respectively. In the ploughed plot, 19 DJs of *S. feltiae* cm^{-2} and 12 DJs of *H. bacteriophora* cm^{-2} were recorded in the first year. However, in the second year, more DJs of *H. bacteriophora* were found in plots with either ploughed or herbicide treated. In opposite, no significant differences were detected for *S. feltiae* between the years (p=0.807) (Fig. 1, 2).

The collection and evaluation of soil samples was continued until no more nematodes were detected. Regardless to the time of disappear, *S. feltiae* and *H. bacteriophora* became disappear firstly in the ploughed and the herbicide treated plots simultaneously in the first year. However, in the second year, *S. feltiae* became disappear firstly at the herbicide-treated plot, whereas *H. bacteriophora* became disappear firstly at the ploughed plot. *S. feltiae* and *H. bacteriophora* were able to survive more in irrigated plots than the others. Additionally, more DJs were detected from sprinkling irrigated plot than dripping irrigated for both nematodes (Fig. 1, 2).

Interactions between the nematodes and the practices. The differences between the tilled plots with the plough control parcels were only found significant in both years for DJs of *H. bacteriophora* (p=0.028 for 2005 and p=0.024 for 2006). However, differences between the herbicide treated and control plots were statistically significant for two years for *S. feltiae* (p=0.047 for 2005 and p=0.031 for 2006). Differences of detected DJs of *H. bacteriophora* between all practices applied in second year and control were also significant. The other practices were not significantly different for both nematodes in experimental years. Details of the interactions were statistically summarised in the Table 3.

DISCUSSION

Persistence of released EPNs in the agro-environment is one of the major factors in sustainable effects of EPNs. Many studies mainly in the laboratories have tested the persistence of EPNs under various conditions in soil. However, outdoor studies on persistence or establishment are

very limited. After application, nematodes rapidly disappear in the applied area (Molyneux, 1985; Kung *et al.*, 1991). The data generally indicated a viability of weeks rather than months and a gradual

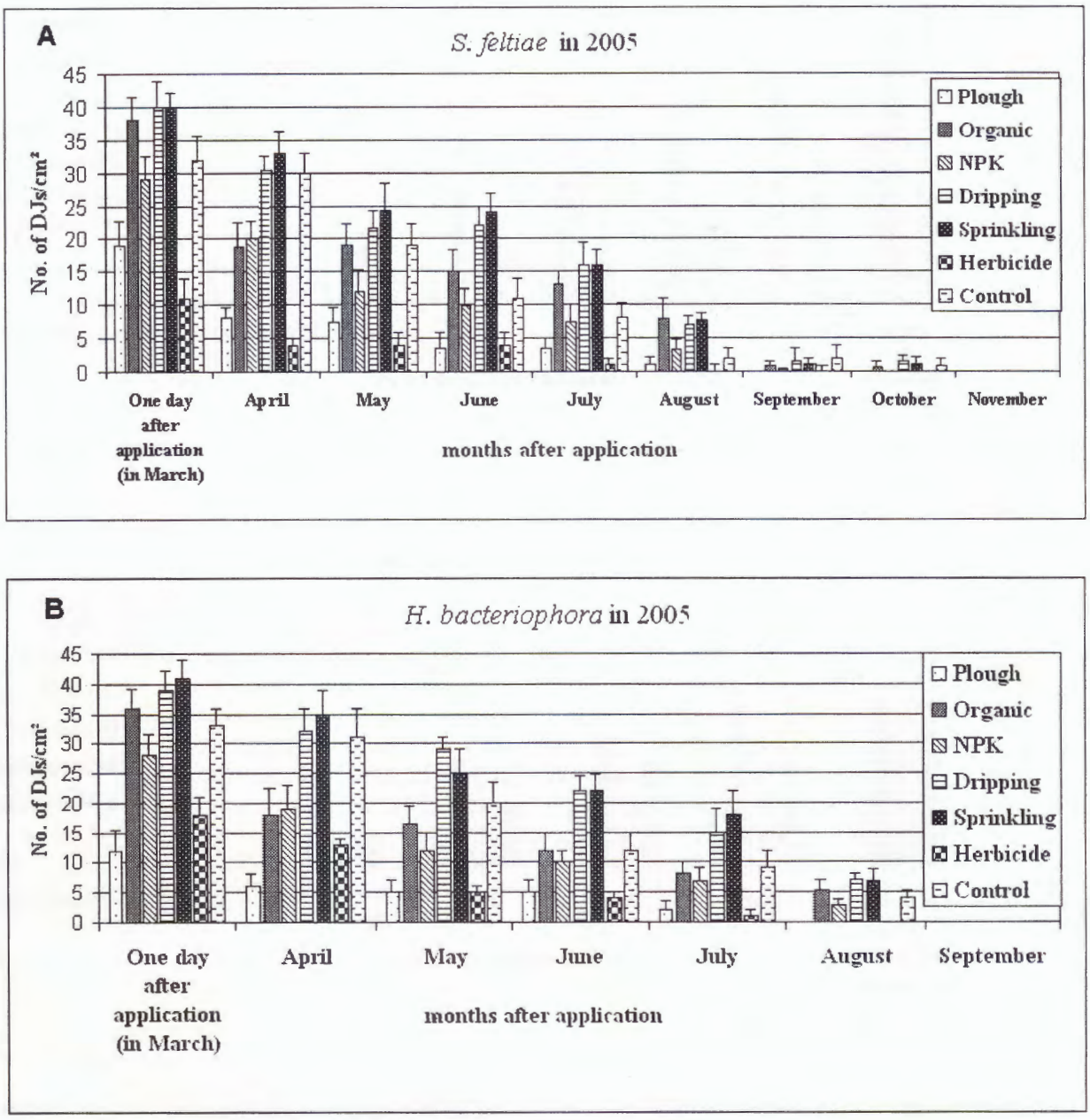
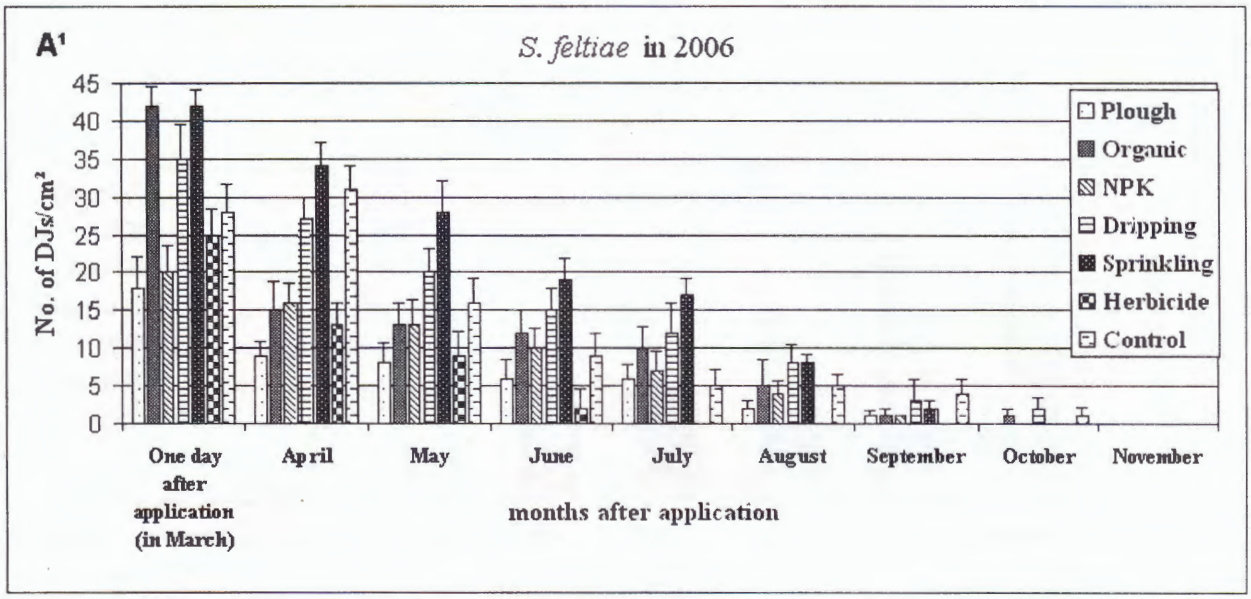


Fig. 1. Persistence measured in tilled, fertilized, watered and herbicide applied experimental plots and control as recovered DJs of the released species, *S. feltiae* (A) in 2005, *H. bacteriophora* (B) in 2005. Bars indicate means and vertical lines represent standard errors of the means.

decline in the numbers of living nematodes recovered. However, Susurluk (2005) stated that *H. bacteriophora* applied in bean field in organic farm had persisted for nearly two years. But, *S. feltiae* sprayed in clover cultivated in organic field had been detected for only one year. However, present results are not concordance with that conclusion. Thus, *S. feltiae* was found more resistant than *H. bacteriophora* to the agricultural applications used in this study.



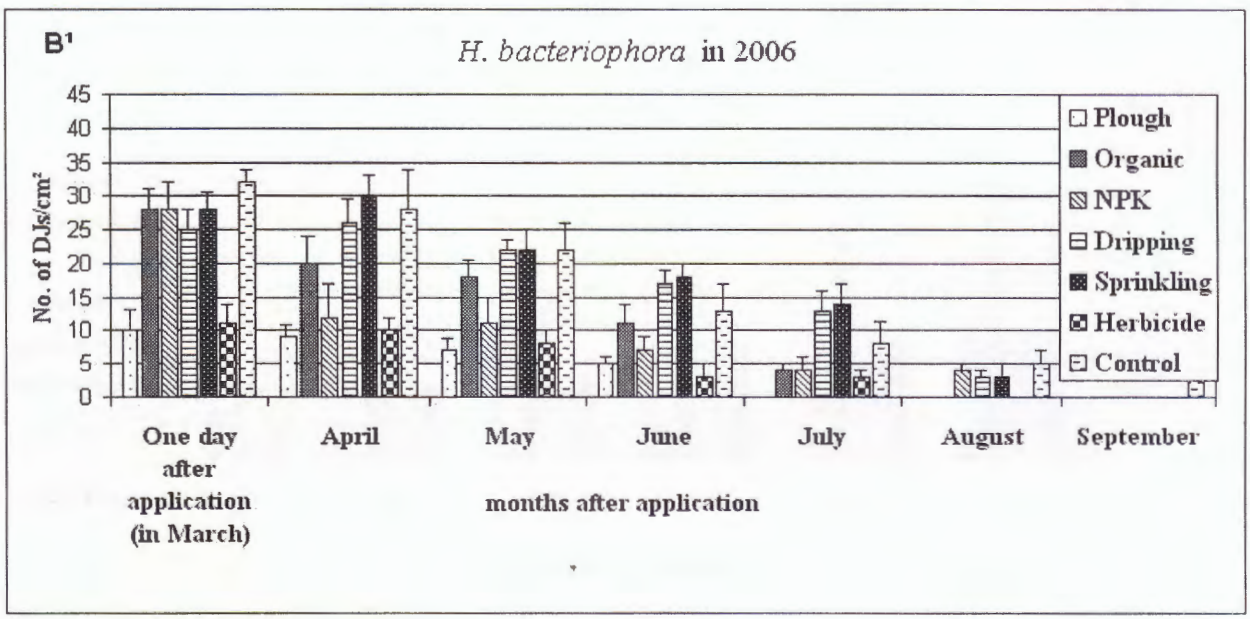


Fig. 2. Persistence measured in tilled, fertilized, watered and herbicide applied experimental plots and control as recovered DJs of the released species, *S. feltiae* (AN) in 2006, *H. bacteriophora* (BN) in 2006. Bars indicate means and vertical lines represent standard errors of the means.

Prolonged exposure to the high concentrations of inorganic NPK fertilizers inhibited the activities of *S. feltiae* and *H. bacteriophora*. In contrast, Bednarek & Gaugler (1997) stated that very short (1 day) exposures to the fertilizers increased the activity and infectivity of EPNs. However, Sturhan (1996) found no such correlation between these factors. The present results indicated that both NPK and organic fertilizers had no affect on either nematode. On the other hand, herbicide Trifluralin EC had a negative affect on *S. feltiae* in both years. This differentiation between *S. feltiae* and *H. bacteriophora* could be explaned with their cuticle structure. The DJs of heterorhabditids remain in the second stage cuticle during detrimental environmental conditions, whereas steinernematids

Table 3. Relationships between all practices together and individual used in the experiment and nematodes in 2005 and 2006. Interactions were analyzed by Student t- test. * = Interaction is statistically significant at P<0.05.

Years	Nematodes	Interactions	df	t-values (observed)	Two-tailed p-value
1.	Both	*S. feltiae X H. bacteriophora*	94	-0.286	0.775
2.	Both	*S. feltiae X H. bacteriophora*	94	-1.721	0.089
1. + 2.	*S. feltiae*	*S. feltiae X S. feltiae*	94	0.245	0.807
1. + 2.	*H. bacteriophora*	*H. bacteriophora X H. bacteriophora*	94	1.592	0.115
1.	*S. feltiae*	Plough X Control	14	1.557	0.142
2.	*S. feltiae*	Plough X Control	14	1.268	0.223
1.	*H. bacteriophora*	Plough X Control*	12	2.558	0.028*
2.	*H. bacteriophora*	Plough X Control*	12	2.576	0.024*
1.	*S. feltiae*	Organic Fert. X Control	14	-0.171	0.867
2.	*S. feltiae*	Organic Fert. X Control	14	0.000	1.000
1.	*H. bacteriophora*	Organic Fert. X Control	12	0.340	0.741
2.	*H. bacteriophora*	Organic Fert. X Control	12	0.776	0.453
1.	*S. feltiae*	NPK Fert. X Control	14	0.878	0.395
2.	*S. feltiae*	NPK Fert. X Control	14	0.684	0.504
1.	*H. bacteriophora*	NPK Fert. X Control	12	0.819	0.432
2.	*H. bacteriophora*	NPK Fert. X Control	12	1.227	0.244
1.	*S. feltiae*	Dripping Irrig. X Control	14	-0.649	0.527
2.	*S. feltiae*	Dripping Irrig. X Control	14	-0.462	0.651
1.	*H. bacteriophora*	Dripping Irrig. X Control	12	-0.854	0.413
2.	*H. bacteriophora*	Dripping Irrig. X Control	12	0.173	0.865
1.	*S. feltiae*	Sprinkling Irrig. X Control	14	-0.785	0.445
2.	*S. feltiae*	Sprinkling Irrig. X Control	14	-0.881	0.392
1.	*H. bacteriophora*	Sprinkling Irrig. X Control	12	-0.936	0.372
2.	*H. bacteriophora*	Sprinkling Irrig. X Control	12	-0.047	0.963
1.	*S. feltiae*	Herbicide X Control*	14	2.174	0.047*
2.	*S. feltiae*	Herbicide X Control*	14	2.436	0.031*
1.	*H. bacteriophora*	Herbicide X Control	12	1.998	0.074
2.	*H. bacteriophora*	Herbicide X Control	12	1.153	0.266
1.	*S. feltiae*	All Practices X Control	55	-0.050	0.960
2.	*S. feltiae*	All Practices X Control	55	-0.337	0.738
1.	*H. bacteriophora*	All Practices X Control	53	-0.223	0.824
2.	*H. bacteriophora*	All Practices X Control*	53	-2.140	0.037*

do not have any cuticle in this stage (Poinar, 1976). Therefore, the cuticle may prevent *H. bacteriophora* against the herbicide. Indeed, DJs are tolerant to short exposures (2-6 h) against most chemical pesticides. However, DJs are highly susceptible to several nematicides likely to be found in the agro-ecosystem (Rovesti & Deseo, 1990; Ishibashi, 1993). Many scientists stated that most insecticides do not interact with EPNs (Zimmermann & Cranshaw, 1990; Ishibashi & Takii, 1993). But, the present study is not in agreement with the results. The result of the herbicide showed that herbicide Trifluralin had also negative affect on *S. feltiae*.

Another result of present study showed that the major impact on the nematode persistence was tillage in the used agricultural practices. In tilled plots, the both nematode populations were found lowest number of recovered nematodes. However, *H. bacteriophora* was found more sensitive against tillage than *S. feltiae*. Favorable soil conditions and the lack of physical disturbance increase the

success of EPNs (Shapiro-Ilan *et al.*, 2002). Under a conventional tillage regime, the soil surface tends to have greater fluctuations in temperature and moisture in comparison to non-tilled or less-tilled soils and EPNs are more frequently detected in reduced tillage regimes (Brust, 1991; Hsiao & All, 1998; Hummel *et al.*, 2002; Millar & Barbercheck, 2002; Shapiro *et al.*, 1996). Brust (1991) reported that lack of tillage regimes and the presence of weeds in the field increased the infection of *G. mellonella* caused by EPNs in the first and subsequent years. Analogously, Susurluk (2005) stated that although enough DJs of *H. bacteriophora* had reached the soil surface after application, only ones positive soil sample was obtained from a potato field tilled strongly. Tilling with a disc harrow and plough has negative effects on the persistence of *H. bacteriophora* in the potato field. These results are in agreement with the present findings especially for *H. bacteriophora.*

Brust (1991) also studied the influence of tillage, weed and irrigation in a cropping system with corn. Irrigation had no influence on the nematode population (*H. bacteriophora*), whereas tillage reduced the population size and persistence of weed increased the population size. The results also confirmed that irrigation prolonged DJs survivals. Dry conditions adversely affect nematode motility and viability. The potential of EPN to survive desiccation, however, is poor (Glaser, 2002). Menti *et al.* (1997) showed that although *H. megidis* survival was superior to that of *S. feltiae*, desiccation tolerance for both species was poor (minutes). Thus, desiccation is considered as a limiting factor, however, it does not have a major impact on nematode survival under normal agricultural conditions. Susurluk (2005) reported that analysis of results on establishment and climatic conditions indicated a correlation between the amount of weekly precipitation (mm) and the number of positive soil samples. His result expressed that optimal precipitation for persistence of *S. feltiae* and *H. bacteriophora* varied from 15 to 30 mm week^{-1}.

The presented data suggest that a single approach on persistence of EPNs in laboratory conditions may not be sufficient for a reliable detection of EPN persistence in the field.

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Alper Susurluk I. Воздействие разных сельскохозяйственных технологий на выживание энтомопатогенных нематод *Heterorhabditis bacteriophora* и *Steinernema feltiae* после их внесения в полевую почву.

Резюме. Исследовано воздействие различных сельскохозяйственных технологий, таких как, внесение удобрений (органических и NPK), орошения (капельного и поливного), пахоты и использования гербицида Trifluralin EC на выживание двух эндемичных турецких штаммов энтомопатогенных нематод *Steinernema feltiae* (Filipjev, 1934) (Rhabditida: Steinernematidae) и *Heterorhabditis bacteriophora* (Poinar, 1976) (Rhabditida: Heterorhabditidae) после их внесения в полевую почву. Эксперименты проводили с мая по ноябрь в 2005 и 2006 годах в районе Анкары. В то время как все сельскохозяйственные технологии оказывали отрицательное воздействие только на второй год на *H. bacteriophora,* никакого отрицательного воздействия на *S. feltiae* не было отмечено в оба года. Нематоды *S. feltiae* показали большую устойчивость в почве в эти годы чем *H. bacteriophora.* Основываясь на данных по повторному обнаружению энтомопатогенных нематод, наиболее низкая выживаемость в течение двух лет наблюдений была отмечена для личинок *S. feltiae* на полях где применяли гербициды, а для личинок H. bacteriophora под вспаханными участками.