RESEARCH/INVESTIGACIÓN

EFFECT OF WHITE-TIP NEMATODE, *APHELENCHOIDES BESSEYI*, ON GRAIN YIELD AND YIELD TRAITS OF SOME JAPONICA RICE CULTIVARS UNDER FIELD CONDITIONS

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ABSTRACT

Tülek, A. 2016. Effect of white-tip nematode, *Aphelenchoides besseyi*, on grain yield and yield traits of some japonica rice cultivars under field conditions. Nematropica 46:8-13.

Trials were conducted on the Thrace Agricultural Research Institute, Turkey in 2013. Three rice cultivars grown in Turkey (Halilbey and the advanced lines, Osmancık/Rus and Kral/Diana from the Turkish national breeding program) and two resistant cultivars from the International Rice Research Institute (Asahi and Rinaldo Bersani) were used to determine the effects of rice white-tip nematode, *Aphelenchoides besseyi*, on yield and yield components of rice. Infested plots that were created with a combination of infected seeds and supplemental inoculation with *A. besseyi* at the seedling stage and uninfested plots were compared. Yield was significantly reduced ($P < 0.01$) in nematode-infested plots in comparison with the control. The cultivars Halilbey, Rinaldo Bersani, Osmancık/Rus, Kral/Diana, and Asahi exhibited 36.2%, 18.1%, 19.3%, 28.2%, and 7.9% yield reduction, respectively. Nematode infection also reduced other agronomic traits including paddy and milled rice 1,000-kernel weight by 4.5% and 6.6%, respectively, and milled rice yield by 7.1%.

Key words: Aphelenchoides besseyi, japonica rice, white-tip nematode, yield.

RESUMEN

Tülek, A. 2016. Efectos del nematodo de las puntas blancas, *Aphelenchoides besseyi*, sobre la producción y características del grano de algunos cultivares de arroz japónica en condiciones de campo. Nematropica 46:8-13.

Los ensayos de campo de se llevaron a cabo en el Instituto de Investigación Agrícola de Thrace, Turquía en 2013. Se determinaron los efectos del nematodo de las puntas blancas, *Aphelenchoides besseyi*, en la producción y componentes de la producción de tres cultivares de arroz cultivados en Turquía (Halilbey y la líneas avanzadas, Osmancık/Rus y Kral/Diana procedentes del programa nacional de mejora Turco) y dos cultivares resistentes procedentes del Instituto Internacional de Investigación del arroz (Asahi y Rinaldo Bersani). Se compararon parcelas no infestadas con parcelas infestadas, que fueron creadas con una combinación de semillas infectadas y una inoculación suplementaria con *A. besseyi* en el estado de plántula. La producción se redujo significativamente ($P < 0.01$) en las parcelas infestadas por el nematodo en comparación con el control. Los cultivares Halilbey, Rinaldo Bersani, Osmancık/Rus, Kral/Diana, y Asahi mostraron 36.2%, 18.1%, 19.3%, 28.2%, y 7.9% de reducción en la cosecha, respectivamente. La infección por nematodos también redujo otras características agronómicas, como el peso de 1000 granos de arroz con cáscara y descascarillado en 4.5% y 6.6%, respectivamente, y la cosecha del arroz descascarillado en 7.1%.

Palabras clave: Aphelenchoides besseyi, arroz japonica, nematodo de las puntas blancas, producción.

INTRODUCTION

The white tip nematode, *Aphelenchoides besseyi* Christie, is a potential pest in rice growing areas of the world (Prasad and Varaprasad, 1992). Yield reductions due to this nematode have been reported to be as high as 71% in USSR (Tikhonova, 1966), 60% in India (Muthukrishnan *et al*., 1974), and 54% in USA (Atkins and Todd, 1959) on susceptible varieties. Seed is the main source of infection and dissemination (Komori et al., 1963).

In several countries, control of *A. besseyi* has been achieved by using nematode resistant rice cultivars (Qu, 1985). It was reported that some rice cultivars in USA including Arkansas Fortuna, Asahi, Bluebonnet, Bluebonnet 50, Improved Blue-bonnet, Century 231, Fortuna, Hill long grain, Nira, Nira 43, Rexoro, Sunbonnet, Texas Patna, Toro, and TP - 49 were resistant and Century 52, Century Patna 231, and Rexark were highly resistant (Cralley and Adair, 1949; Todd and Atkins, 1959) to white tip. In Japan, Tosan 38 is very resistant, Norin 8 and Norin 43 are resistant, and Norin - Mochi 5 and Natsushimo are highly resistant (Goto and Fukatsu, 1956) to this nematode, and in Italy, Rinaldo - Bersani is resistant, and Carnaroli and Pierrot are considered to be highly resistant (Orsenigo, 1954).

Aphelenchoides besseyi is an important nematode pest of rice production in Turkey. The pest was first reported in 1995 in the İpsala and Gönen districts of Edirne and Balıkesir provinces in Turkey (Ozturk and Enneli, 1997). This study aimed to determine yield and quality losses due to white-tip nematode in two advanced lines of rice obtained from the Turkish national breeding program that have been determined to be resistant to *A. besseyi*.

MATERIALS AND METHODS

A trial to estimate yield loss due to *A. besseyi* was established on the experiment station of the Thrace Agricultural Research Institute, Turkey in 2013. The rice cultivar, Halilbey, two advanced breeding lines, Osmancık/Rus and Kral/Diana, and two known *A. besseyi*-resistant control cultivars, Rinaldo Bersani (IRGC – Acc. No. 9468) from Italy and Asahi (IRGC – Acc. No. 33983) from Japan, were studied. Seed of the resistant control varieties, Rinaldo Bersani and Asahi, were obtained from the International Rice Research Institute (Los Banos, Philippines). Seed of the local rice cultivar Halilbey, which was considered a susceptible control for white tip nematode and the advanced rice breeding lines, Osmancık/Rus and Kral/Diana, were obtained from the Turkish rice breeding program. Osmancik/Rus and Kral/Diana were found to be resistant to *A. besseyi* in varietal reaction experiments (Tulek *et al*., 2015 unpublished data). All of the rice cultivars and advanced lines used in these experiments are *Oryza sativa* var. Japonica (Japonica rice).

Infected and nematode-free seeds used in the experiment were grown the year before the trial was initiated. The experiment was established as split plot in a randomized complete block design with three replications. Main plots were infested and uninfested plots and cultivars were evaluated as sub-plots. Seeds were sown to a depth of 5 to 6 cm in plots of 7 m^2 (6 rows, 6 m long × 1.2 m wide with 20 cm between rows) at a rate of 500 seeds per square meter. Intermittent irrigation was performed until seedlings appeared and continous irrigation until harvest, with plants submerged in 15 cm water. All the testing materials were planted in the second half of May.

Infested plots received both naturally infected seeds that were drill-planted in the plots and artificial inoculation with 5,000 *A. besseyi* per m^2. The related methods were as follows: *Aphelenchoides besseyi*-infected seeds (100 of each variety), which had been grown the year before, were used and assayed for nematodes in 5 replications. The values obtained constituted the initial population density in the seedlot. The rate of reproduction was calculated by the formula "Multiplication rate of rice white tip nematode = final population (Pf) / inital population (Pi)". Additionally, 5,000 *A. besseyi* individuals per m^2 were applied in the seedling stage. Inoculum was obtained from two sources. Nematodes were initially isolated from infested seeds and reared on carrot disk in the laboratory (Tülek *et al*., 2009). In addition, nematodes were collected from infected Halilbey seeds produced the previous year. Nematodes were sprayed on to water-filled rice plots 10 d after germination. Nematode-free seeds were obtained by presoaking seed for 3 to 5 hr in cold water then submerging the seed in water at 55-60°C for 15 min (Tülek and Çobanoğlu, 2011a) using an electric Benmari (P-D Industriegesellschaft mbH Typ WB 10 Nr.:634537 DIN 12876 Klasse 1, Dresden, Germany) containing 12 liters of water.

All cultural practices were applied as in conventional rice farming. For the weed control in the plots, penoxulam (Cherokee 25.2 gl, DowAgroSciences) at a rate of 1,000 cm^3 ha^{-1}) and bentazon (Basagran 480 gl, BASF) at a rate of 2,000 cm^3 ha^{-1} were applied prior to rice tillering. The crop was harvested at the end of September when 80% of kernels were straw yellow in color and harvest moisture of rice grain was at 20% to 24% . Following harvest, nematode analysis was done on the grains by assaying three replications of 100 seeds each. Nematode densities and yield data were subjected

to analysis of variance. Means were compared by LSD ($P < 0.05$).

To determine of white-tip ratio in cultivar, the seeds of all varieties used in the experiments were sown by hand in a dry field in a randomized complete block design with three replications, with 1 row as 1 m and a space between the rows of 30 cm, with 100 seeds in each row. The seed beds were then watered. To test the cultivars, the seeds were obtained from infected panicles from the previous year. In addition, artificial inoculation was performed with 5,000 *A. besseyi*/m^{-2}. The plants in each row were reaped during the flowering stage, and the ratios of plants (%) with symptoms of white tip in the flag leaf were determined. On the tillers of the affected plants, the tips of the leaves were bleached for a distance of 3 to 5 cm and shredded.

The Baermann funnel method (Hooper, 1986) was used to obtain the nematodes both before sowing and after the harvest. Prior to assay, arbitrarily chosen seeds were separated from hulls (lemma and palea). Since *A. besseyi* individuals were found localized inside hulls and on the brown rice (unmilled rice) at 97% and 3%, respectively (Tülek and Çobanoğlu, 2011b), only hulls were used for nematode extraction. Samples were kept in water in flasks for 48 hr, and bottom-sedimented nematodes were collected into tubes in volumes of 20 ml. The nematodes in two, 1-ml aliquots of the water suspension from each extract were counted in counting dishes, using a stereo-microscope. Two counts were made, and the number of nematodes was averaged. The averages were used to calculate the number of *A. besseyi* in 100 paddy seeds. After harvest, three 100-g paddy rice samples were drawn from each sample and milled rice yield (%) was determined using a rice testing machine, (Zaccaria, PAZ-1-DTA, Brazil).

RESULTS

All five cultivars evaluated in the field trials were naturally infected with *A. besseyi*, although the population density that was detected varied by cultivar (Table 1). At harvest, nematodes multiplied in all cultivars with the exception of Osmancık97/Rus, which had a multiplication rate of 0.5. The nematode was detected in seed of cultivars in all plots that had been planted to infected seed (Table 2). Paddy rice yield was higher in uninfected plots than infected plot where the white tip symptoms were present in the plant in flowering stage for all cultivars except Asahi and Osmancık/Rus. The nematode resulted in a decrease in yield ranging from 7.91% to 36.2%, and 1,000-kernel weights were lowered 1.94% to 9.59% by the nematode.

At the seed infection rate seen in the field trial (114.3 *A. besseyi* per 100 seeds) paddy rice yield was 22% lower ($P < 0.01$) in rice plots planted to infected seed than where nematode-free seed was used (Table 3). Although the 1,000-kernel weights for paddy rice were not different between plots where infected seed were planted and the nematode-free control plots, milled rice 1,000-kernel weights were lower ($P < 0.05$) where infected seed were grown; and milled rice yield was 7.1 % lower ($P < 0.05$) where the nematode was present.

When the relationship between yield and certain other characteristics was examined, a negative correlation ($P < 0.05$) was found between yield and milled rice yield and the number of white-tip nematode in 100 seeds (r = -0.5948 and r = -0.5616, respectively) (Table 4).

DISCUSSION

Cultivar differences in the reaction to *A. besseyi* have been reported by many countries. The presence or absence of white tip and the number of

Table 1. At planting (Pi) and harvest (Pf) *Aphelenchoides besseyi* population density and multiplication rate (Pf/Pi) in five rice cultivars in yield trials.

Rice cultivars	*A. besseyi*/100 grains (Pi)	*A. besseyi*/100 grains (Pf)	Multiplication rate (Pf/Pi)
Rinaldo Bersani	19 c[z]	66.6 c	3.5
Osmancık97/Rus	53 b	28.8 c	0.5
Halilbey	135 a	307.7 a	2.3
Asahi	4 c	8.3 c	2.0
Kral/Diana	72 b	160.0 b	2.2

[z]Data are the means of three replications. Means within columns followed by the same letter are different at $P < 0.05$ by LSD.

Table 2. Decrease in paddy rice and milled rice yield (T ha^{-1}), 1,000-kernel weights (g), and *Aphelenchoides besseyi* density (number/100 seed) in plots planted to infected and uninfected seed.

Cultivar	Uninfected plot (control)	Infected plot	Decrease in infected plots (%)
	Paddy rice yield (T ha^{-1})		
Rinaldo Bersani	7.7 c[z]	6.3 d	18.1
Osmancık97/Rus	8.1 bc	6.5 d	19.2
Halilbey	9.6 a	6.1 d	36.1
Asahi	8.9 ab	8.2 bc	7.9
Kral/Diana	8.4 bc	6.0 d	28.1
	Paddy rice 1000-kernel weight (g)		
Rinaldo Bersani	36.5 a	35.1 a	3.6
Osmancık97/Rus	31.7 b	29.8 cd	5.8
Halilbey	30.3 bcd	28.9 de	4.7
Asahi	27.3 ef	26.8 f	1.9
Kral/Diana	30.8 bc	27.9 ef	9.5
	Milled rice 1000-kernel weight (g)		
Rinaldo Bersani	26.0 a	24.5 b	6.0
Osmancık97/Rus	23.0 c	21.6 e	5.8
Halilbey	22.5 cd	20.5 f	8.7
Asahi	20.0 fg	19.9 fg	0.6
Kral/Diana	22.1 de	19.6 g	11.1
	Milled rice yield (%)		
Rinaldo Bersani	66.4 a	65.5 ab	1.3
Osmancık97/Rus	66.2 a	66.0 ab	0.2
Halilbey	62.5 bc	60.2 c	3.7
Asahi	55.4 d	42.9 e	22.6
Kral/Diana	61.6 c	55.3 d	10.2
	A. besseyi/100 grains		
Rinaldo Bersani	0	66.6 c	
Osmancık97/Rus	0	28.8 cd	
Halilbey	0	307.7 a	
Asahi	0	8.3 d	
Kral/Diana	0	160.0 b	

[z]Within a category, means not sharing the same letter differ significantly at $P < 0.05$ using LSD.

nematodes at the end of the season have always been considered major indicators of resistance (Cook, 1974). There was no sign of white-tip symptoms on the flag leaf of the resistant control cultivars Asahi and Osmancık/Rus, whereas white-tip ratio was determined in Halilbey, Rinaldo Bersani, Kral/Diana as 39.3%, 2.2%, and 1.4%, respectively. The highest disease incidence based on visible white-tip symptoms was observed at the flowering stage of the crop. Yield losses of up to 57.9% for susceptible cv. Halilbey, have been reported in the field with 77% of plants with white tip symptoms on the infected flag leaves (Tülek and Çobanoğlu, 2010). Tikhinova (1974) reported that white tip of rice periodically is epiphytotic with infection of plants ranging from 30% to 80% and yield losses of 0.5 to 1.8 ton/ha in Povolzhe, Kuban, and Rostov regions.

Resistance to *A. besseyi* is considered to be

Table 3. Paddy rice and milled rice yield (T ha^{-1}) and 1,000 kernel weight (g), nematode density (number of *Aphelenchoides besseyi* per 100 seed), and percent decrease in yield in the treatments.

Components	Uninfected plot (control)	Infected plot	Decrease in infected plots (%)	F-value
Yield (T ha^{-1})	8.5 a^{z}	6.6 b	22.1	309.7**
Paddy rice 1,000-kernel weight (g)	31.2 a	29.8 a	4.5	8.9 ^{NS}
Milled rice 1,000-kernel weight (g)	22.7 a	21.2 b	6.6	42.1*
Milled rice yield (%)	62.4 a	58.0 b	7.1	16.5*
A. besseyi /100 grains	0 b	114.3 a	-	101.7**

(**) $P < 0.01$ (*) $P < 0.05$, NS = Non-Significant
^{z}Within a category, means not sharing the same letter differ significantly at $P < 0.05$ using LSD.

Table 4. Correlation coefficients of some traits with grain yield in 5 rice cultivars.

Trait	Yield	Paddy rice 1000-kernel weight	Milled rice 1000-kernel weight	Milled rice yield
Paddy rice 1000 kernel weight	-0.2526^{NS}	1		
Milled rice 1000 kernel weight	-0.1737^{NS}	0.9810**	1	
Milled rice yield	-0.5948*	0.6614**	0.6460**	1
A. besseyi / 100 grains	-0.5616*	-0.1647^{NS}	-0,2676^{NS}	0.1586^{NS}
A. besseyi /100 grains	0 b	114.3 a	-	101.7**

** = Significant at $P < 0.01$ level, * = Significant at $P < 0.05$ level, NS = Non-Significant using LSD.

genetically controlled and carried by the Japanese cv. Asahi (Nishizawa, 1953), and resistance is one of the best control methods for this nematode. In our yield experiment, Asahi paddy and milled rice 1,000-kernel weight were the least affected of the cultivars. However, milled rice yield was suppressed to the greatest degree in the plots planted to nematode-infected seed compared to the control plots in Asahi. This is thought to be due to Asahi being a late-maturing cultivar that was impacted by a high rate of broken seeds from the early harvest.

The nematode multiplication rate was lowest in the Osmancık97/Rus cultivar. Earlier reports indicate a decrease of less than 15% in seed weight while 30 to 70% decreases were seen with susceptible varieties when the population of nematodes was relatively high (Popova *et al.,* 1980). Asahi variety was tolerant and Rinaldo Bersani and Osmancık97/Rus cultivars were partially tolerant in that study.

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