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OCCURRENCE OF WHITE-TIP NEMATODE, *Aphelenchoides besseyi* IN CERTAIN RICE CULTIVATIONS AT SOUTH DAKAHLIA GOVERNORATE, EGYPT AND ITS MANAGEMENT UNDER FIELD CONDITIONS.

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ABSTRACT

Examination of the vegetative stage, as well as panicles at harvest (fresh) and seed samples from farmer's store (12 months) of rice cvs. Sakha 101 & 103 and Rehio grown in South Dakahlia fields for *Aphelenchoides besseyi* symptoms on leaves recorded the whitening of the leaf tip for the vegetative stage, and the shortened twisted, chlortic strips along one edge of the leaf for the flowering and harvest stages of the rice cultivars studied. Results also indicated that a total of 18.27% of the stored grains and 6.66% of the fresh grains were obviously infested with *A. besseyi*. Moreover, rice cv. Sakha 101 appeared to be the most susceptible to *A. besseyi* with infestation levels amounted to 22.22% and 7.5% and number of nematodes per 100 seeds 115 and 30 individuals for stored and fresh rice seeds, respectively. Panicles of rice cv. Sakha 101 with white-tip disease symptoms were obviously shorter by 16.08%, lighter by 38.63%, decreased by 53.71% of 1000 grains weight with sterile grains amounted to 41.53% as compared to those of panicles without any clear disease symptoms. Number of *A. besseyi* was higher in the diseased panicles than those without apparent disease symptoms, which was amounted to 160 and 15 individuals per 100 seeds, respectively. Length of flag leaf averaged 5.3 cm in plants with disease symptoms.

With respect to *A. besseyi* management on rice plant cv. Sakha 103 during the rice growing season 2003 with the tested four chemical pesticides as well as the two plant extracts data revealed that all tested materials obviously reduced nematode population. Moreover, periwinkle *Vinca rosea* accomplished the highest percentage of nematode reduction with value of 55.71% in shoot, followed by Dimathoate (25.88%), whereas thorne apple, gave the least value of nematode reduction (1.62%) when compared with the untreated plots, respectively. Moreover, the bioagent, *V. rosea* achieved the highest reduction percentage of nematode population in grains (83.3%) and surpassed all tested compounds in increment grains weight (61.4%) followed by Dimathoate (54.6%).

Keywords: Rice cultivations, Symptoms, white-tip disease, *Aphelenchoides besseyi*, Infestation level, Yield, pesticides, Plant extracts, periwinkle, *Vinca rosea*, thorn apple, *Datura stramonium*.

INTRODUCTION

White-tip disease of rice *Oryza sativa* L. caused by the seed-borne nematode, *Aphelenchoides besseyi*, Christie, 1942 has been widely distributed in many rice growing countries in Asia, Tropical America, formely USSR and Africa (Franklin & Siddiqui, 1972, Fortuner & Williams, 1975 and Ou, 1985). It was recently recorded by Amin (2002) for the first time in Egypt

during a survey of plant parasitic nematodes in the paddies of Dakahlia and Sharkia governorates in the Nile Delta. *A. besseyi* caused variable yield losses in different countries ranging from 14.5 to 46.7% in Japan (Nishizawa & Yamamoto, 1951), 40 – 50% in U.S.A. (Atkins & Todd, 1959) 29 to 46% in Taiwan (Hung, 1959), 41 to 71% in formely USSR (Tikhonova, 1966) and 20 to 60% in India (Rao *et al.*, 1985). *A. besseyi* can survive for one year inside the rice seeds and 53 days in water under 10°C (Qiu *et al.*, 1991). The white-tip nematode infected the rice plant ecto parasitically in the beginning and penetrated the rice flowers and hibernated beneath the seed glumes as fourth stage juveniles and adults (Nandakumer *et al.*, 1975). Once the nematodes revive, leave the seeds to attack new rice seedlings. The occurrence and regulation of *A. besseyi* recovered from soaking either twenty grams of straw or one hundred rice grains of four cultivars i.e. Sakha 101, Giza 172, 171 and Reiho for 48 or 48 and 96 hrs at 25 ± 2°C revealed that Giza 171 and Reiho achieved the highest levels of nematode infestations in rice grains as well as straw, whereas Sakhia 101 showed the least values (Khalil and El-Sherif, 2003). Management of rice white tip nematode, *A. besseyi* was studied by some workers (Kumar & Sivakumar, 1998; Tacconi *et al.*, 1999; Tiwari, 2000, El-Sherif & Khalil, 2003 and Amin & Al-Shalaby, 2004). Amin and Al-Shalaby (2004) studied the efficacy of disinfestation by soaking rice seeds cv. Giza 77 in different concentration of cadusafos (Rugby 20%) or oxamyl (Vydate 24%) for 24 hrs and found that cadusafos was more effective than oxamyl in increasing the percentage of nematode mortality. Moreover, cadusafos at 200 and 400 ppm and oxamyl at 400 ppm showed maximum nematode mortality without damage on seed sprouting. Therefore, white-tip disease of rice caused by *A. besseyi* in Egypt with its symptoms in certain selected rice fields at Simbellawian county, South of Dakahlia governorate were chosen: (1) to study the status of this nematode through [illegible] of rice plants within the growing season of 2002 and 2003 for [illegible] cultivars i.e. Sakha 101, 103 and Rehio, (2) to determine the [illegible] its infestation in the farmer's seed stock of rice yield of 2002 season in comparison with those of rice yield of 2003 season at harvesting time as well as its effects on yield components and (3) to determine the effect of certain chemical pesticides i.e. Malathion, Dimathoate, Cartan, Carbo El-Nasr as well as two plant leaf extracts i.e. thorne apple and periwinkle on controlling *A. besseyi* on rice cv. Sakha 103 under field conditions during the growing season of 2003.

MATERIALS AND METHODS

Incidence and Distribution of the Disease:

During rice growing season of 2002, certain locations in fields of Shubrakeballa and Sobaeen villages at Simbellawian district, South Dakahlia governorate of naturally infested rice seedlings cvs. Sakha 103, 101 and Rehio exhibiting symptoms of white tip disease caused by *A. besseyi* were selected and marked by bamboo stakes for collecting their leaves at vegetative stage, flowering and panicles at harvest in order to be used in this study. Seeds of each rice cultivar i.e. Sakha 103 and 101; and Rehio that

were harvested on October 1st, 2002, were separately stored in paper bags at room temperature 25 ± 2°C for 12 months until September 30th, 2003 as farmer's seed stocks. On the 1st of October 2003, samples of each rice cultivar under study that was marked during rice growing season at the same locations were collected for estimating the actual infestation level in the field (harvesting time) in comparison with those of the farmer's seed stock of rice growing season of 2002. Then, twenty rice seeds of each cultivar of the rice growing seasons of 2002 and 2003 were split open and separately soaked in distilled water (Cobb, 1918) over night at 25 ± 2°C. The extracted nematodes were examined, identified microscopically, counted and recorded.

Based on the presence of nematodes, seed samples per cultivar of each rice yield/season, were indexed at different levels of infestation. The infestation level in the seed stock (stored) was compared with the actual infestation level in the field (at harvesting time).

Effect on Yield Components:

Ten apparently healthy panicles and others showing white tip symptoms were collected from infested fields planted with rice cv. Sakha 101 at the same locality of Simbellawian district. Length and weight of these panicles were determined and recorded, and the panicles were separately threshed. Filled and sterile grains in each panicle were determined, counted and recorded. Then, the 1000 grains weight as well as the nematode population in 100-grains/rice cultivar tested were estimated and recorded.

Management of *A. besseyi* Infesting Rice Plants Under Field Conditions:

To determine the influence of certain pesticides i.e. Malathion 57% EC., Cheminova, 0.0-dimethyl-S-(1,2-dicarbothoxy) ethyl phosphorodithioate; Carbo El-Nasr 10% G., 2, 3-dihydro –2,2-dihydro-2,2-dihydro-2,2-dimethyl benzofural-7-methyl, carbonate; Dimathoate 40% [0,0-dimethyl-S-(methyl-Carbamoyl)] methyl phosphorodithiate, Ctan 10% (Local); Methyl N', N'-dimethyl-N- [(methyl carbamoyl) Oxy]-1-thioo-Xamimidate as well as two aqueous leaf extracts of periwinkle, *Vinca rosea* and thorne apple, *Datura stramonium* (that were prepared according to Khalil (1996) were selected to control *A. besseyi*. An area of 210 m^2 was chosen at Nedia 2 location of Shubrakeballa village, Simbellawian district, South Dakahlia of Northeastern Nile Delta region to carry out this experiment. The selected area was divided into seven plots each with thirty square meters. Each plot served as a treatment with three replicates at 10 m^2 each. Thirty days after sowing rice cv. Sakha 103 seeds, seedlings were transplanted into the previous mentioned area, which was prepared according to the procedure of rice cultivation under the Egyptian conditions. Ten days later, pesticidal applications as well as aqueous leaf extracts tested were added. Treatments were as follows:

(1) Malathion 57% EC., (2) Carbo El-Nasr 10% G., (3) Dimathoate 40% EC., (4) Cartan 10% G (Local), (5) *Vinca rosea*, (6) *Datura stramonium* and without any chemical or plant extracts (N alone).

Malathion or Dimathoate was separately added as spray at the rate of 5 ml/L/replicate, respectively. Each of Carbo El-Nasr or Cartan (Local) was introduced at the rate of 21.28 gm, per replicate, whereas each aqueous leaf

extract of periwinkle or thorn apple at 50% tested as spray at the rate of 12.5 ml/L/replicate, Each treatment was replicated three times.

Two weeks later, NPK fertilizer was added at the recommended rate/replicate. The initial population of *A. besseyi* in 300 hundreds rice seeds cv. Sakha 103 was previoulsy determined. One hundred seeds were soaked in cup filled with tap water at 25 ± 2°C for 48 hr, sieved, collected counted and the average of nematode number was then recorded to be 25 individuals per one hundred of rice grains.

Twenty days after transplanting rice seedlings, 50 grams of rice stems and leaves in each/replicate/treatment were weekly collected in plastic bag between August 25th and September 17th 2003, sent to the Nematology Laboratory and Kept in a refrigerator at 4°C for nematode extraction. Nematode extraction was carried out by cutting the shoot and leaf into small pieces 0.25 cm each, soaked in tap water for 48 hrs at room temperature (25 ± 2°C), sieved (Coff, 1918), collected counted and the average number of *A. besseyi* was determined and recorded. Data were subjected for analysis of variance (Anova) Gomez and Gomez, 1984) and means were compared by Duncan's multiple range test (Duncan, 1955).

RESULTS

The present work revealed that rice plants cvs Sakha 101, 103 and Rehio at the vegetative stage, flowering stage and panicles at harvest, showed widely distribution of *A. besseyi* is in the studied rice area (Simbellawian, South Dakahlia governorate, Egypt). The symptoms on rice leaves e.g. whitening of the leaf tip during the vegetative growth stage and shortened, twisted and crinkled leaf seen during the flowering and harvest stages of rice cv. Sakha 101 are shown in Figures 1 and 2.

Data in Table (1) revealed the white-tip nematode infestation in farmer's seed samples as well as in fresh seed samples at harvest and the average number of *A. besseyi*/100 seeds for both cases. A total of 18.29% of the stored seeds and 6.66% of the fresh seeds were obviously infested with the white-tip nematode. Apparently all rice cultivars, i.e. Sakha 101, 103 and Rehio grown in Simbellawian district, were found to be infested with *A. besseyi* at the rate of infestation reached to 7.14, 22.22 and 15.00% for stored seeds and 3.33, 7.50 and 6.00% for fresh seeds of Sakha 101, 103 and Rehio, respectively.

Although, the number of seed samples taken from rice cultivar Sakha 103 which is a new one was high, it was found to be the most susceptible to *A. besseyi* both in terms of infestation level (22.22 and 7.50%) and number of nematodes per 100 seeds (115 and 30 individuals) for stored as well as fresh rice seed, respectively. It was also noticed that fresh seeds always had fewer nematodes than stored ones (Table 1) for all rice cultivars tested, since their average number were 10, 30 and 20 individuals/100 seeds for Sakha 101, 103 and Rehio cvs, respectively.

Data in Table (2) showed the effects of *A. besseyi* on panicle yield in susceptible rice cv. Sakha 101 which indicated that panicles with White-tip disease symptoms were obviously shorter by 16.08% and lighter by 38.63%

as compared to the panicles without disease symptoms. The diseased panicles had few filled grains and 41.53% of grains were sterile. Moreover the weight of 1000 grains from diseased panicles decreased by 53.71% and the nematode population per 100 seeds in diseased panicles (160 individuals) was obviously higher than in panicles without apparent disease symptoms (15 individuals) (15.0) (Table 2). Average length of flag leaf was 5.3 cm in plants with disease symptoms.

Table 1: White-tip nematode infestation in farmer's stored seed and fresh seed samples at harvest in Simbellawian district, south of Dakahlia governorate.

Rice cultivar	Number of seed samples infested with *A. besseyi*		% infestation with *A. besseyi*		Average numbers of *A. besseyi* per 100 seeds	
	Stored	Fresh	Stored	Fresh	Stored	Fresh
Sakha 101	50 (700)	10(300)	7.14	3.33	60	10
Sakha 103	200(900)	60(800)	22.22	7.50	115	30
Rehio	90 (600)	30(500)	15.00	6.00	75	20

*Figures within the parenthesis are total seed samples.

Table 2: Population density of white-tip nematode in rice grains and its influence on yield components in rice cv. Sakha 101 under field conditions at Simbellawian district.

Yield component	Plant without disease symptoms	Plants with disease symptoms	Percent of increase (+) or decrease (-)
Panicle length (cm)	23.00	19.30	-16.08
Panicle weight (g)	4.40	2.70	-38.63
Filled grain (no.)/panicle	119.70	70.10	-41.52
Sterile grain (no.) panicle	14.30	85.50	+497.9
1000 grain weight (g.)	29.60	13.70	-53.71
Nematodes/100 grains	15.00	160.00	+966.66
Flag leaf length (cm)	-	5.3	-

*Means of 10 samples.

Data in Table (3) indicated the impact of four chemical pesticides i.e. Carbo El-Nasir, 10% G, Cartan 10% G, Dimathoate 40% E.C., and Malathion 57% E.C. as well as two aqueous leaf extracts i.e. thorne apple and periwinkle on controlling *A. besseyi* infesting rice plants (shoot & panicles) during the growing season of 2003 under field conditions. It was evident that the periwinkle leaf extract achieved the highest percentage of nematode reduction with value of 55.71%, followed by Dimathoate and Carb El- Nasir with values of nematode reduction percentage reached to 25.88 and 24.47%, respectively. In addition, the applications of thorne apple leaf extract, cartan (granules) and Malathion gave the least values of nematode reduction percentages which were amounted to 1.62, 5.76 and 6.96%, respectively.

Table 3: Impact of four chemical pesticides and two aqueous plant leaf extracts on controlling *Aphelenchoides besseyi* infesting rice plants cv. Sakha 103 during the growing season of 2003 and rice grains yield under field conditions.

Treatment	Date of sampling				No. of nematodes in shoot (50 g.)**	% of Reduction	No. of nema. per one gm. grain**	% Reduction	Weight of 1000 grains (g.)**	% Increase
	8/25	9/2	9/10	9/17						
Carbo El-Nasr 10% G.	290	170	162	125	186.50^{b}	24.47	6^{a}	50	27.5^{b}	50.18
Cartan 10% G	310	224	192	106	233.00^{a}	5.76	9^{bc}	25	20.3^{c}	32.51
Thorn apple	310	288	225	150	243.20^{a}	1.62	11^{b}	8.3	15.7^{c}	12.73
Dimathoate40% E.C.	246	260	122	105	183.25^{b}	25.88	5^{a}	58.3	30.2^{b}	54.60
Malathion57% E.C.	220	246	246	210	230.50^{a}	6.96	8^{bc}	33.3	25.3^{b}	45.84
Periwinkle	224	90	32	42	109.50^{c}	55.71	2^{a}	83.3	35.5^{b}	61.4
Check	270	340	285	294	297.00^{a}	-	12^{b}	-	13.70^{a}	-

* Each figure represented the mean of three replicates.
** Means with the same letters are not significantly different at P < 0.05
*** Initial population of *A. besseyi* per one gram grains = 80 individuals.
One gram = 40 rice grains (seeds).

Finally, the bioagent, periwinkle leaf extract achieved the highest reduction percentage of nematode population in grains that was amounted to 83.3%. Moreover, it was surpassed all treatments tested in increasing weight of grains (yield) with value of 61.4%.

DISCUSSION

Investigation of rice plants cvs. Sakha 101, 103 and Rehio at the vegetative and flowering stages; and panicles at harvest and seed samples from farmer's store, showed that the white-tip nematode symptoms on leaves, e.g. whitening of the leaf tip during the vegetative stage and the shortened, twisted and crinkled leaf seen during the flowering stage of rice cultivars studied were similar to that recorded in other types of rice (Yoshii & Yamamoto, 1950; Todd & Atkins, 1958 and Ou, 1985). The chlorotic strips along one edge of the leaf of an infested rice plant was regularly considered as an important leaf symptom under Egyptian conditions. The presence of nematode infested grains on the uppermost part of the panicle or throughout the panicle, together with the obviously healthy grains are in agreement with the report made for other cultivars of rice (Steel, 1970).

Examination of both stored as well as fresh seed samples of rice cultivars i.e. Sakha 101, 103 and Rehio tested indicated that a total of 18.27% of the stored seeds and 6.66% of the fresh ones were clearly infested with *A. besseyi*. Moreover, the fresh seeds always had fewer nematodes than the stored ones/100 grains, for all. These results agreed with the findings of Rahman & Miah (1989) and Khalil and El-Sherif (2003), since the levels of infestation ranged from 14.8% to 28.4% for fresh seeds and 45.0% to 81.2% for six months stored seeds/100 grains for rice cvs. Sakha 101, 103 and Rehio, respectively (Khalil and El-Sherif, 2003). The apparent reduced infestation level at harvest could be due to the masking of symptoms on leaves as they die and dry-out. Fukano (1962) in Japan recorded that 30 or

more live nematodes per 100 grains may be the possible economic threshold level in such susceptible variety.

Concerning the management of *A. besseyi* infesting rice cv. Sakha 103 under field conditions during the growing season of 2003, periwinkle, *Vinca rosea* as foliar spray achieved the highest percentage of nematode reduction followed by Dimathoate 40% E.C. and then Carbo-El-Nasr 10% G, with values of 83.3%, 58.3% and 50.0%, respectively, whereas Cartan 10% G gave the least value of nematode reduction when compared with the untreated plots. These results agree with that of El-Sherif and Khalil (2003) who reported that spraying Dimathoate achieved the highest percentage of *A. besseyi* reduction during all rice growth stages. Meanwhile, this bioagent, *V. rosea* as foliar spray application accomplished the highest percentage of rice yield increase, followed by Dimathoate 40% E.C. and Carbo-El-Nasr with values of 61.4%, 54.6% and 50.2%, respectively and reduced *A. besseyi* population on rice plant with values of 55.71%, 25.88%, and 24.47%, under field condition, respectively. However, when *V. rosea* applied as a dried leaf powder in comparison with the same pesticides for controlling *A. besseyi* on rice plants in pots under greenhouse conditions recorded the third rank in suppressing numbers of *A. besseyi* per 100 rice grains (El-Sherif and Khalil, 2003). This may be attributed to its behaviour of parasitism as an ectoparasite pest attacking the rice young leaf where, *V. rosea* as foliar spray applied.

It is worthy to note that this is the first report in Egypt for using such successful bioagent, *V. rosea* as foliar spray in the management of *A. besseyi* on rice plant under field conditions, however, more research is needed to clarify such method in this respect.

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تواجد نيماتودا القمة البيضاء *Aphelenchoides besseyi* في بعض زراعات الأرز في جنوب محافظة الدقهلية في مصر ومكافحتها تحت ظروف الحقل

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أسفر فحص أعراض الإصابة بنيماتودا القمة البيضاء فى الأرز *Aphelenchoides besseyi* فى مرحلة النمو الخضرى والزهرى والسنابل وقت الحصاد (حديثة) وكذلك عينات حبوب مخزونة (١٢ شهرا) لدى المزارع لكل أصناف الأرز سخا ١٠١، ١٠٣ وريهو بحقول الأرز بجنوب محافظة الدقهلية عن تسجيل إبيضاض قمة أوراق النباتات المصابة فى مرحلة النمو الخضرى وقصرها والتفافها مع وجود شرائط ملونة بطول حافة واحدة لورقة النبات فى مرحلة التزهير والحصاد فى كل أصناف الأرز تحت الدراسة.

كما دلت النتائج أن نسبة ١٨,٢٧% من الحبوب المخزونة و ٦٦,٦% من الحبوب الحديثة وقت الحصاد مصابة بنيماتودا *A. besseyi* بدرجة ملحوظة. وكان الصنف سخا ١٠٣ أكثر الأصناف قابلية للإصابة بمعدل ٢٢ر٢٢% و ٧,٥% محتويا على ١١٥ و ٣٠ فردا لكل ١٠٠ حبة فى الحبوب المخزونة والحديثة على التوالى.

كما كانت سنابل الصنف سخا ١٠١ للنباتات المصابة بمرض القمة البيضاء قصيرة (١٦,٠٨%) وخفيفة الوزن (٣٨,٦٣%) وقليلة فى وزن الألف حبة (٥٣,٧١%) مع وجود حبوب عقيمة بنسبة ٤١,٥٣% إذا ما قورنت بالسنابل التى لم تظهر أعراض المرض. وكان معدل أعداد النيماتودا فى ١٠٠ حبة عالى فى السنابل ذات أعراض مرضية عن تلك بدون أعراض بمعدل ١٦٠ و ١٥ فرد على التوالى. كما كان طول ورقة العلم ٥,٣سم فى النباتات المصابة.

أوضحت نتائج مكافحة نيماتودا القمة البيضاء فى الأرز *A. besseyi* على صنف سخا ١٠٣ خلال موسم النمو ٢٠٠٣ تحت ظروف الحقل باستخدام أربعة مبيدات كيماوية هى الملايثون ٥٧% والدايمثويت ٤٠% (كلاهما رشا) والكاربو النصر ١٠% والكارتان ١٠% (كلاهما نثرا)، وكذا مستخلصين نباتيين (كلاهما رشا) الداتورا والونكا إلى أن كل هذه المواد خفضت تعداد النيماتودا بدرجة ملحوظة على الأوراق، وكذا الحبوب حيث أعطى مستخلص نبات الونكا أعلى نسبة خفض فى تعداد النيماتودا بالأوراق (٥٥,٧١%) يليه مبيد الدايمثويت (٢٥,٨٨%)، بينما مستخلص نبات الداتورا أقل فى قيمة خفض تعداد النيماتودا (١,٦٢%) مقارنة بالنباتات الغير معاملة. كما أعطى مستخلص نبات الونكا أعلى معدل خفض فى تعداد النيماتودا فى الحبوب (٨٣,٣%) وبالتالى زيادة فى محصول حبوب الأرز بمعدل ٦١,٤% يليه فى ذلك مبيد الدايمثويت (٥٤,٦%).

ملحوظة: تم تمويل هذا البحث من وحدة حساب البحوث بجامعة المنصورة.