J. Plant Prot. and Path., Mansoura Univ., Vol. 6 (6): 951 - 968, 2015

EVALUATION OF SOME RICE CULTIVARS AND LINES TO *Fusarium moniliforme (Verticilloides)* THE CAUSAL FUNGUS OF RICE BAKANAE DISEASE AND ESTIMATION OF YIELD LOSS

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ABSTRACT

The experiments of this study were conducted at lab, greenhouse and the experimental farm of Rice Research and Training Center (RRTC), Sakha, Kafr El-Sheikh during 2013, 2014 and 2015 seasons.. Results indicated that all the tested isolates of *F. moniliforme* (21) varied in their virulence. However, the high virulent isolates were obtained from isolates no, 12, 2 and 1 which gave 79.33, 73.33 and 72.0% ; infection respectively. While, isolate no. 6 was avirulent one. Also, results indicated that all rice cvs. differed in their susceptibility to infection with tesed isolates of *F. moniliforme*. Sakha 101 and Giza 177, were the most susceptible cvs. Hybrid 1 and Hybrid 2 exhibited the highest infection percentage. Survival percentage of *Fusarium moniliforme* in the infected grains or soil gradually after 2-10 months, then decreased tremendously after two years of storage either in a screenhouse or at room temperature. The results indicated that the highest number of dead(infected) plants were (17.67 plant/pot) was obtained from seedling infection 100% at 10 DAT compared with (0.5 plant/pot) of healthy 100 %. Moreover, the highest number of infected plant (14 plant/pot) was obtained from seedling infection 65 % followed by (12.00 plant/pot) from seedling infection 75 % at 45 DAT compared (0.70 plant/pot) of healthy 100 %. Data indicated that, the highest grain yield (4.962, 4.899 and 4.856 t/fed) was in treatment of spray at 5 DAF followed by spray at 15 DAF(4.826, 4.685 and 4.697 t/fed) that obtained from Beam, Sumi8 and Vitavax spray treatments, respectively compared with control (4.125 t/fed) at 2013 season.

Keywords: Rice, Bakanae, Foot rot , *Fusarium moniliforme*

INTRODUCTION

Rice (*Oryzae sativa* L.) is one of the most important cereal crops cultivated in Egypt and many of the tropic countries of the world, which feeds more than half of the world's population (Singh *et al*., 2013). The common annual cultivated area in Egypt is 1.1 – 1.2 million feddans, produce 4.0 – 4.8 million tons of rough rice for local consumption as well as exportation. In 2014; growing season, the area of rice crop reached 1.5 million feddans, produced about 6 million tons of paddy rice with an average of 4.00 t/fed, which was considered one of the highest average yield in the world per feddan (RRTC, 2014). However, rice crop is subjected to biotic and abiotic agents. In Egypt, the major rice diseases are fungal diseases such as rice blast, brown spot and other minor diseases. Recently, bakanae disease of rice caused by *Fusarium moniliforme* which is later identified as *F. fujikuroi* (Nirenberg, 1976) the anamorph stage is *Gibberella fujikuroi* Sawada (Nirenberg, 1976). Bakanae disease was observed for the first time in Egypt at 2001 season on Giza 177 and Sakha 101 cultivars at Kafr El-Sheikh governorate, and subsequently has been recorded in other Nile Delta Governorates on Giza 177, Giza 178, Sakha 101, Sakha 102, Sakha 104 and Hybrid 1 rice cultivars (Gabr, 2010). However, the pathogen *F. moniliforme* was isolated earlier from rice grains and root rots of Egyptian rice cultivars as indicated in earlier studies (Sehly, 1974 and Hassan, 2008). Recently; the rice disease profile has been changed in response to altering in rice production situations the diseases like bakanae, sheath rot, false smut and grains discoloration which were minor and occurring sporadically in the past are newly emerging diseases and causing considerable yield losses (Kumar *et al*., 2013). Bakanae is a Japanese word which means bad or naughty seedling refers to the abnormal elongation, “thin noodle seedling”, “foolish seedling”, and “stupid rice crop” (Sun and Snyder, 1981). Bakanae is one of the newly emerged increasing problems of rice, particularly on basmati rice in India during recent years (Pannu *et al*., 2012). It is one of the major fungal diseases of rice including blast, sheath blight, brown spot and sheath rot in India (Sharma and Thind, 2007). It is difficult to develop bakanae resistant rice varieties due to the high genetic variation of the causal pathogens (Serafica and Cruz, 2009).Therefore bakanae has a major concern in the affected rice growing areas of Egypt and also becoming more alarmed threat for sustainable rice production in other parts of the rice growing world. The research on bakanae disease and its sustainable management is need of hour and must be given top priority for disease free quality seed production, realizing higher yield potential of aromatic rice and to get edge in rice trade at international market before it is too late. The study aims to evaluation of rice cultivars and some promising lines to disease elongation rice plants (Bakanae) and the ability of spores fungus to survival in grain and soil. Also, study of a good method to get the healthy grains without spores of *F. moniliforme*.

MATERIALS AND METHODS

1. Isolation of the causal organism:

Diseased specimens were cut into 1 to 2 cm pieces, sterilized for two minutes by immersing in 0.5 % sodium hypochlorite solution (NaOH); rinsed twice in sterilized distilled water and then; placed onto potato dextrose agar medium (PDA). The plates were incubated at 25 °C for 5 to 7 days and the developed fungi were purified using single spore or hyphal tip techniques according to Hansen (1926).

2. Identification:

The different isolates of the causal fungus were identified according to the morphological characteristics and microscopic examination at Plant Pathology Laboratory at Rice Research & Training Center (RRTC) at Sakha, Kafr El-Sheikh Gov. using the key of imperfect fungi (Barnett and Hunter, 1972; Nelson *et al.,* 1983 and Summerell *et al.,* 2003).

3.Pathogenicity test:

Samples of diseased rice materials showing bakanae disease symptoms were collected from different rice cultivars grown in the different Nile Delta Governorates. Twenty one isolates of *Fusarium moniliforme* were obtained from rice cultivars. Grains of Sakha 101 rice cultivar (the most sensitive cv. to rice bakanae disease) were soaked in spore suspension (4 x 10^5 spores/ml) of the tested isolates for 48 h., then incubated for 48 h., and then 50 grains were seeded in each plastic pot (15 x 15 cm. diameter), the pots were arranged in rows, each row included 3 pots as replicates for each isolate. The pots were kept in the greenhouse at 30-35 °C and fertilized one time with urea 46.5% N at 3g/pot. The untreated check was seeded with 50 grains previously soaked in 100 ml of distilled water for 48 h. and incubated for another 48 h. The germination percentage (%) was recorded before sowing and number of infected plants and plant height (cm) were recorded 30 days after sowing.

4. Survival of *Fusarium moniliforme*:

Grains and soil used for studying the survival of *F. moniliforme* fungus were stored at room temperature and screenhouse as natural conditions for two years from 1st December 2012 to 1st October 2014.

Grains

Infected grains of Sakha 101; the susceptible rice cv. ; collected from artificially inoculated plots by *F. moniliforme* as spore suspension (4 x 10^5 spores/ml) at flowering stage in the previous season (October, 2012) were stored at room temperature and screenhouse made of perforated iron nest as natural conditions and evaluated every two months up to two years. Fifty grains were sown in sterilized soil in plastic pots (15 x 15 cm. in diameter) with three replicates for each treatment. All pots were kept in the greenhouse at 30-35 ºC and fertilized one time with urea 46.5% N at 3g/pot. Percent of germination was recorded after sowing and number of infected plants were estimated 30 days after sowing.

Soil

Artificially inoculated soil using corn meal sand medium were prepared. Glass bottles of 500 ml capacity containing 190 g clean moistened sand and 10 g corn meal, were autoclaved for 30 min. at 1 atm, then inoculated with *F. moniliforme* and incubated at 28 ºC for 15 days, as described by Pratt and Janke, 1980. The inoculum was added as soil infestation 3 % (w:w). The treated soil were stored at room temperature and screenhouse as natural conditions and evaluated every two months up to two years. Healthy grains of Sakha 101 rice cv were seeded in artificially infested soil, or sterilized soil to serve as control (fifty grains per pot). All the pots were kept inside the greenhouse at 30-35 ºC and fertilized as mentioned before and three replicates were used for each treatment. The germination percent was recorded after sowing and number of infected plants were estimated 30 days after sowing.

5. Evaluation of certain rice cultivars and promising lines toward infection with *Fusarium moniliforme*:

The response of thirteen rice cultivars namely: Giza 177, Giza 178, Giza 179, Giza 181, Giza 182, Sakha 101, Sakha 102, Sakha 103, Sakha 104, Sakha 105, Sakha 106, Hybrid1 and Hybrid 2 and seven lines as promising lines GZ 7112-6-20-1, GZ 8710-3-2-1-1, GZ 9057-6-1-3-2, GZ 9461-4-2-3-1, GZ 9626-2-1-3-2-3, GZ 9626-2-1-3-2-4 and GZ 9807-6-3-2-1 to infection with ten isolates of *Fusarium moniliforme* were studied in pots (15 cm in diameter). Hundred grains of each cultivar and line were soaked in spore suspension concentration at 4.5x 10^5 spores/ml of each isolate of the fungus for 48h then incubated for 48h at (30 ºC). The grains of each cultivars and lines were soaked in the tap water for 48h then incubated for 48h at (30 ºC) as cheek. The pots were kept in the greenhouse at 25–30 ºC and fertilized once as mentioned before where three replicates were used for each cultivar. The germination percent was recorded before sowing and number of infected plant and plant height were recorded 30 days after sowing.

6. Effect of infection levels of bakanae rice disease on yield loss:

The experiment carried out under screenhouse conditions during 2014 season under artificial inoculation to study the effect of levels infection on yield loss. Twenty infection levels i.e. 5%, 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95% and 100% of infection seedling by *Fusarium moniliforme* and 100% healthy seedlings were tested. Seeds of Sakha 101cv. was soaked in spore suspension of the fungus at (6x10^7 spore/ml) for 48 hours then sown in sterilized soil for 25 days as infected plant. Seeds of Sakha 101cv. soaked in sterilized water for 48 hours then sown in sterilized soil for 25 days as healthy plants. After 25 days, five seedlings per hill were transplanted in four hills per pot (60 cm in diameter). This experiment was performed in complete randomized design with three replicates for each level. The pots were fertilized two times with urea 46.5% N at 15g/plot as basle dose and 10g/plot 30 days after transplanting. The number of infected plants, number of dead plants due to infection, number of infected plants with panicles, number of infected plants without panicles, height of healthy and infected plants (cm), No. of tillers, 1000 grains weight (g), biomass (g/pot) and grain yield (g/pot) were recorded.

7. Effect of spraying fungicides at heading stage on rice bakanae disease:

A field experiment were conducted at the experimental farm of Rice Research and Training Center (RRTC), Sakha, Kafr EL-Sheikh during 2013 and 2014 seasons to investigate the effect of different fungicides spray on the production of healthy seeds free from bakanae rice disease. The treatments were arranged in a randomized complete block design with three replicates. Nursery beds of the rice cultivar Sakha 101 was seeded at the rate of grains 60 Kg/feddan and soaked in spore suspension of *F. moniliforme* at (5.5 x 10^6 spore/ml) for 48 hours then sown on 15 may, and the experiment was transplanted about one month later in plots measured(3 x 3.5 m^2), with four plants/hill. Seedlings were transplanted in the flooded paddy field on June15.

The nitrogen fertilizer was added as Urea (46.5 % N) at the rate of 60 units of nitrogen per Feddan. Two thirds of nitrogen dose was incorporated to top 15cm of the dry soil as a basal application before transplanting, other cultural practices were undertaken as recommended. Five fungicides (Vitavax 200 75% WP 2g/L, Beam 75% WP 0.5g/L, Fuji-one 40% EC 2ml/L, Sumi8 5% EC 1ml/L, Copral 50% WP 5g/L) were applied at three dates started from flowering stage with 10-days intervals. The germination percentage, percentage of infected grains and grain yield (Ton/feddan) were recorded.

RESULTS AND DISCUSSION

1. Symptoms detection

Our observation through the whole growing season showed that Infected seedlings often progressively die from the seedling stage through to maturity. If infected plants survive to heading, the panicles they produce are usually sterile. The flag leaf on mature plants is noticeable by its elevated, more horizontal stance. The affected plants may be several inches taller than normal plants, thin, yellowish green and may produce adventitious roots at the lower nodes of the culms (Figure 1). Diseased plants bear few tillers and leaves dry up quickly. The affected tillers usually die before reaching maturity; when infected plants survive, they bear empty panicles (Webster and Gunnel, 1992; Heong *et al*., 2005). Saremi (2005) found that diseased plants senesce and die, mycelium of the fungus may emerge from the nodes and may be visible above the water level. After the water is drained, the fungus sporulates profusely on the stems of diseased plants. The sporulation appears as a cottony mass and contaminates healthy seed during harvest. The bakanae pathogen can over winter as spores on the coat of infested seeds. It can also over winter in the soil and plant residue. However, infested seeds are the most important source of infection.

2&3. Isolation and identification of the causal organism and Pathogenicity tests:-

Pathogenicity of all obtained isolates (21 ones) were tested on Sakha 101 rice cultivar, the most susceptible one. Disease incidence percent was estimated by counting number of the infected plants per hundred plants. Data presented in Table (1) indicate that all the tested isolates varied in their virulence. However, the high virulent isolates were isolates No, (12, 2 and 1) which gave (79.33, 73.33 and 72.00 % infection percentage respectively). While, isolate no. 6 was avirulent one. Data in Table 1 clear that germination percent generally ranged from 70.33 % in case of isolate no. 17, isolated from Gharbia to 87.33 % in isolate no. 20 from Beheria governorate. However, plant heights reflected the level of infection of the different isolates. Sun and Snyder (1978) reported that the fungus is more easily isolated on selective agar media such as those containing quintoozene. They added for good sporulation, culture must be kept under continuous light. Siddiqi *et al*., (1995) found that maximum growth of *F. moniliforme* was observed after 8 days of incubation on maltose, lactose, sucrose and starch compared with 11 days on other carbon sources. Of the 15 carbon sources, macro-conidial and micro-conidial production were highly produced on maltose, lactose and sucrose after 8 days of incubation at 25±2 ^{o}C degrees. Aurangzeb *et al*., (2003) found that potato dextrose agar medium (PDA) was the best for mycelial growth followed by Wakasman's agar, Basal medium, Czapeck's dox agar and Richard's medium. The fungus showed significant growth within a range of 25-30 ºC, the best growth was observed at 30 ºC. The effect of light and dark periods on fungal growth was insignificant, however significant results were obtained when light was given for a continuous period of 24 hours.

Table (1):Pathogenicity tests of isolates of *Fusarium moniliforme* isolated from rice cultivars grown in four governorates during 2014 growing season using Sakha 101 rice cultivar.

Isolate no.	Governorate	Source of isolate	Germination %	Infection %	Plant height (cm)	
					Healthy	Infected
1	Kafrelsheikh	Giza 177	81.33	72.00	15.55	26.66
2		Sakha 101	83.67	73.33	15.78	29.11
3		Sakha 104	81.67	45.67	15.78	26.00
4		Giza 171	81.00	62.33	15.11	25.33
5		Sakha 101	82.00	62.33	15.78	27.78
6		Maize	78.33	0.017	14.44	14.44
7		Sakha 102	80.33	24.00	14.00	25.67
8		Sakha 105	75.67	54.67	16.00	29.55
9		Hybrid 1	78.00	53.33	14.66	26.44
10	Dakahlia	Sakha 104	82.33	67.00	14.44	29.78
11		Giza 178	73.67	25.00	15.55	25.78
12		Sakha 101	72.00	79.33	15.55	32.00
13		Giza 177	74.00	61.00	15.11	25.33
14		Sakha 102	79.67	12.67	15.33	27.78
15	Gharbia	Sakha 101	85.67	62.67	15.33	25.78
16		Giza 177	83.33	51.67	14.00	26.44
17		Sakha 101	70.33	13.67	15.11	26.88
18	Behera	Sakha 101	72.33	51.67	14.66	27.33
19		Giza 178	83.33	17.00	15.33	27.55
20		Giza 177	87.33	53.67	15.33	26.00
21		Giza 177	82.33	61.33	15.78	25.55
Control			92.33	5.67	16.33	29.55
L.S.D. 5 %			3.425	1.215	0.820	2.526

4. Evaluation of certain rice cultivars and promising lines towards infection with *Fusarium moniliforme*:

Ten isolates of *F. moniliforme* (isolate No. 1, 2, 5, 6, 8, 10, 11, 14, 16, 20) were used for evaluating susceptibility of 11 cvs., two hybrid namely; (Giza 177, Giza 178, Giza 179, Giza 181, Giza 182, Sakha 101, Sakha 102, Sakha 103, Sakha 104, Sakha 105, Sakha 106, Hybrid1 and Hybrid2) and fife lines namely (GZ 7112-6-20-1, GZ 8710-3-2-1-1, GZ 9057-6-1-3-2, GZ 9461-4-2-3-1, GZ 9626-2-1-3-2-3, GZ 9626-2-1-3-2-4 and GZ 9807-6-3-2-1) Table (2). This experiment was preformed in greenhouse at Sakha Agric. Res. St. during 2015 growing season.

Data in Table (2) indicate that all the tested rice cvs. differed in their susceptibility to infection with isolates of *F. moniliforme*. However, Sakha 101 and Giza 177 were the most susceptible cvs. Hybrid 1 and Hybrid 2 exhibited the highest infection percentage (43.068%, 38.305%, 30.935% and 26.968% respectively). On the other hand; Sakha 104, GZ 9461-4-2-3-1 and GZ 9626-

2-1-3-2-3 were the least susceptible ones (9.410%, 4.502% and 4.235% ;respectively). Also, the results indicated that all the tested isolates differed in their virulent to the tested cultivars and lines. However, isolates No. 16, 1 and 5 were high virulence (32.09%, 24.89% and 23.95% ;respectively). While, isolate No. 6 which was avirulent. The tested lines in the present work varied in their susceptibility to the disease. Similar results were obtained by Cheema *et al*., (1998) and Bagga and Vineet-Kumar (2000), who reported that infection with rice bakanae disease vary according to the tested cultivars. Krishanaveni *et al*., (2003) tested ninety-three rice genotypes for their resistance to the bakanae disease in the glasshouse. Observations were recorded at 7-day intervals from the 15th to the 42nd day after sowing. Twelve genotypes (ADT-40, ADT-44, ADT-41, ASD-16, Amulya, Sabita, Ereimaphou, Prasanna, GR-4, IR-64, Akutphou and MTU 1010) were highly resistant, 19 genotypes were resistant, whereas the remaining genotypes were moderately susceptible, susceptible or highly susceptible to bakanae disease.

Table (2): Evaluation of certain commercial rice cultivars, hybrids and promising lines to the infection with bakanae disease under artificial soil infestation with *Fusarium moniliforme* under greenhouse during 2015 season.

No.	Entry	Mean infection (%)										check	Cultivar . Mean
		Isolate no.											
		1	2	5	6	8	10	11	14	16	20		
1	Giza 177	62.67	74.33	35.33	0.17	25.67	25.00	47.33	14.33	72.00	26.33	0.17	38.305
2	Giza 178	15.00	12.0	7.67	0.17	12.67	11.00	24.00	39.67	9.00	9.00	0.17	14.002
3	Giza 179	18.67	8.67	45.67	0.17	2.33	17.67	30.33	13.33	31.67	24.00	0.17	19.201
4	Giza 181	24.67	15.00	14.33	0.17	21.33	25.00	15.00	7.00	10.67	8.00	0.17	14.102
5	Giza 182	16.00	19.67	7.67	0.17	5.67	13.67	6.67	13.00	72.33	5.00	0.17	15.968
6	Hybrid 1	25.67	28.67	64.67	0.17	13.66	28.67	15.67	25.67	81.33	25.67	0.17	30.935
7	Hybrid 2	1633	14.33	47.67	0.17	8.67	6.00	71.67	51.33	26.33	27.33	0.17	26.968
8	Sakha 101	51.67	63.33	70.67	0.17	32.67	20.33	23.67	32.00	85.00	51.33	0.17	43.068
9	Sakha 102	22.00	25.33	22.67	0.17	12.33	15.00	12.33	47.67	20.00	25.00	0.17	20.235
10	Sakha 103	15.00	5.67	13.67	0.17	15.00	15.00	2.33	45.33	53.33	5.33	0.17	17.068
11	Sakha 104	12.33	1.33	5.00	0.17	5.67	13.00	8.67	20.67	15.00	12.33	0.17	9.401
12	Sakha 105	25.00	13.67	15.00	0.17	12.33	8.33	63.33	5.67	25.00	5.00	0.17	17.335
13	Sakha 106	12.33	13.00	21.33	0.17	10.33	26.00	10.67	15.67	28.33	14.67	0.17	15.235
14	GZ 7112-6-20-1	46.33	20.33	10.67	0.17	5.00	11.00	10.67	35.33	20.67	25.33	0.17	18.535
15	GZ 8710-3-2-1-1	15.33	41.00	43.33	0.17	11.33	24.67	30.33	46.00	14.00	11.33	0.17	23.735
16	GZ 9057-6-1-3-2	11.67	4.67	9.67	0.17	4.33	4.67	5.67	2.67	15.00	5.33	0.17	6.368
17	GZ 9461-4-2-3-1	8.67	1.67	2.67	0.17	1.67	2.67	8.00	15.67	2.67	1.33	0.17	4.502
18	GZ 9626-2-1-3-2-3	1.67	15.67	1.67	0.17	4.33	5.67	5.67	2.67	5.67	2.33	0.17	4.235
19	GZ 9626-2-1-3-2-4	47.33	26.67	26.67	0.17	4.33	7.00	7.67	19.00	30.33	18.67	0.17	18.768
20	GZ 9807-6-3-2-1	51.33	12.33	15.33	0.17	21.67	2.33	15.33	28.33	25.33	11.00	0.17	18.502
Isolate Mean		24.89	20.77	23.95	0.17	11.54	14.04	20.50	23.95	32.09	15.62	18.73	--

L.S.D 5% **Cultivars 1.4804** **Isolates 1.4401**
Cultivars X Isolates 1.9750

5. Survival of *Fusarium moniliforme*:

Infected rice grains of the susceptible cultivars Giaz 177 and Sakha 101 to *F. . moniliforme* and infested soil were used to estimate the causal fungus survival in two years at screenhouse and room temperature. For grain samples data presented in Table (3) show that, the highest germination (89.33 and 90.67%) were obtained after six month (1^{st} June, 2013) and decreased gradually up to (14.33 and 28.67 %) after two years (1^{st} October, 2014) for screenhouse and room temperature respectively. About infection percent, the highest infection (55.67 and 57.0 %) were obtained (1^{st} December, 2012) and decreased gradually up to (0.33 and 1.40 %) after two years (1^{st} October, 2014) for greenhouse and room temperature respectively.

As regard to soil samples, data present in Table (4) indicat that, the highest germination (94.33 and 91.33%) were obtained after eight month (1^{st} August, 2013), while the lowest germination (55.0 and 43.33 %) were obtained at (1^{st} December, 2012) for screenhouse and room temperature respectively.As for infection percent, the highest infection (88.33 and 92.67 %) were obtained (1^{st} December, 2012) and decreased gradually up to (0.16 and 1.23 %) after two years (1^{st} October, 2014) for greenhouse and room temperature respectively.

Survival percentage of *Fusarium moniliforme* in the infected grains or soil were decreased after two months of storage up to 10, then decreased tremendously after two years of storage either in a screenhouse or at room temperature. Similar results were obtained by Biswas and Das (2003), who reported that the fungus survived up to three months from the date of soil samples collection. The fungus survived in infected rice stubbles kept in soil under natural environmental conditions up to four months. Low temperature favored the survival of the fungus in the infected stubble-pieces. The pathogen survived in the infected rice grains stored at room temperature for considerably ten months. Dodan *et al.*, (1994) in India found that *F. moniliforme* of rice, survived in 15-17% of infected grains of 4 cultivars (Taraori Basmati, Jaya, HKR 120 and PR 109) until the next crop season (mid June).

Sunder and Satyavir (1997) studied the survival of *F. moniliforme* in soil enriched with different nutrients and their combinations from November 1993 to July 1994. Fungal recovery from stubble declined with the incubation period irrespective of amendments, but none of the nutrients at test concentrations (nitrogen 37.5-150, P_2O_5 15-60, K_2O 15-60 mg/kg soil) affected the survival of the pathogen significantly after 8 months of storage of stubble in soil. However, in propagule-infested soil, the higher dose of nitrogen (150 mg/kg) reduced its population (c.f.u./g soil) significantly compared with the unamended controls at the completion of study. Among the various nutrient-combinations, the soil enrichment with NPK, $ZnSO_4$ and $FeSO_4$ significantly reduced the survival of the pathogen in stubble, but none of the combinations affected the fungal recovery from propagule-infested soil significantly after 8 months of incubation.

Table (3) : Effect of storage period on survival of rice bakanae disease (*Fusarium moniliforme*)in infected grins of Sakha 101 rice cultivar.

No.	Period	Screenhouse		Room temperature	
		Germination (%)	Infection (%)	Germination (%)	Infection (%)
1	1st December, 2012	77.00	55.33	76.33	57.00
2	1st February, 2013	86.33	41.67	86.67	52.00
3	1st April, 2013	85.67	31.33	90.67	44.33
4	1st June, 2013	89.33	22.33	90.67	33.33
5	1st August, 2013	83.67	15.67	88.33	28.67
6	1st October, 2013	77.00	10.33	77.33	24.33
7	1st December, 2013	76.67	6.33	76.33	17.67
8	1st February, 2014	62.67	3.67	74.33	15.33
9	1st April,2014	55.00	3.33	61.33	11.33
10	1st June,2014	40.33	0.67	53.33	7.33
11	1st August,2014	22.67	0.33	47.33	4.33
12	1st October, 2014	14.33	0.33	28.67	1.40
L.S.D. 5 %		1.625	1.935	3.228	2.325

Table (4) : Effect of storage period on survival of rice bakanae disease (*Fusarium moniliforme* in the infested soil using Sakha 101 rice cultivar.

No.	Period	Screenhouse		Room temperature	
		Germination (%)	Infection (%)	Germination (%)	Infection (%)
1	1st December, 2012	55.00	88.33	43.33	92.67
2	1st February, 2013	64.67	33.67	51.67	73.33
3	1st April, 2013	74.33	12.33	73.00	49.33
4	1st June, 2013	84.33	5.67	87.33	17.67
5	1st August, 2013	94.33	2.33	91.33	7.33
6	1st October, 2013	91.67	1.67	88.67	1.40
7	1st December, 2013	92.67	0.29	88.67	1.47
8	1st February, 2014	93.67	0.37	89.67	1.23
9	1st April,2014	90.33	0.20	91.67	1.13
10	1st June,2014	89.67	0.29	90.67	1.17
11	1st August,2014	90.00	0.21	90.67	1.10
12	1st October, 2014	91.67	0.16	88.33	1.23
L.S.D. 5 %		2.112	2.125	1.256	3.235

6. Effect of infection levels of bakanae rice disease on yield losses:-

This experiment was carried out under screenhouse conditions during 2014 season under artificial inoculation to study the effect of infection levels on yield losses. Data in Table (5) show that, the highest number of dead infected plants (17.67 plant/pot) was obtained from seedling infection 100% at 10 DAT compared with (0.5 plant/pot) of healthy 100 %. While the highest number of infected plants (14 plant/pot)was obtained from seedling infection 65 % followed by (12.00 plant/pot) from seedling infection 75 % at 45 DAT compared with (0.70 plant/pot) of healthy 100 %. Concerning number of the infected plants with and without panicles, the highest number of infected plant with panicles (9.33 plant/pot) was obtained from seedling infection 65 % followed by seedling infection 60 % (7.67 plant/pot), while the lowest (0.70 plant/pot) was obtained from healthy seedling 100%. About the infected plants without panicles, the highest infected plant without panicles (6.00 plant/pot) was obtained from 75 % level compared with the lowest number of infected plants without panicles (0.13 plant/pot) from healthy seedling 100%. Data in Table (6) indicate that the highest plant height (89.00cm) was obtained from seedling infection 70 %, while the seedling infection 80% gave the lowest height (84.00cm). The highest number of tillers (96.33 and 96.00 tiller/pot) was obtained from seedling infection 10 % and those healthy 100 % respectively, while the lowest number of tillers (13.0 tiller/pot) was obtained from seedling infection 100 %. As regard to 1000 grain weight, the highest (27.83g) was obtained from healthy seedling 100 %, while the lowest (12.40g) was obtained from seedlings infected 100 %. About yield losses percent, the highest yield losses (82.65 %) was obtained from seedling infection 100 %, while the lowest (3.18 %) was obtained from seedling infection 5 %. This is in agreement with the findings of (Rathaiah *et al.*, 1991) and (Hossain 2009). Rathaiah reported that an infected hill showed 67% reduction in yield over the adjacent healthy hill of highly susceptible rice variety (Kmj 1-27-2) in India. Likewise Hossain *et al.*, 2007 calculated as 56.80% yield loss on the basis of hill infection, although, the loss of yield in T. Aman was low. Data recorded in the present study also revealed a direct relationship between disease incidence and reduction in yield. This results are in agreement with the findings of (Sunder *et al.* 1997). He stated that each increase in the disease incidence resulted in a simultaneous reduction in yield. Bakanae disease of rice is also a similar type of disease where infected plants die either in seed bed or in field after few weeks of transplanting. If the infected plants survive until maturity, they bear only empty panicle (Ou, 1985). Present study indicate that yield loss caused by bakanae disease is highly correlated with the number of plants infected in a particular field or season. The disease normally cause yield reduction and economical damage, so researchers try to find different ways to manage it, including resistant cultivars (Zhang *et al*., 2005). Yasin *et al*., (2003) found that Bakanae disease infestation levels of 5, 10, 25, 50, 75 and 100% of seedlings were compared with non-infested control in a field trial conducted at the Rice Research Institute, Kala Shah Kaku, Lahore, Pakistan. According to the results, treatments with 5, 10, 25, 50, 75 and 100% infestation had significantly lower paddy yields to be 4.15, 3.95, 3.75, 2.97, 2.45 and 1.87 t/ha, respectively, against 4.45 t/ha paddy yield of the control. Losses of 57.97% were recorded in 100% infested treatment producing 68.40% seedling infection. The study indicated the potential of the disease to cause heavy economic losses. Also, Hossain *et al.,* (2007) reported that bakanae is one of the major diseases of rice in Bangladesh and caused loss of yield up to 25% in the susceptible varieties. The disease is mainly seed borne and some seed dressing chemicals are usually used to control it.

Table (5): Effect of infection levels of bakanae rice disease (*Fusarium moniliforme* on yield losses under screenhouse conditions, season (2014).

No.	Treatment	No. of dead infected plant		No. of infected plant			No. of infected with panicle	No. of infected without panicle
		10 DAT*	20 DAT	20 DAT	30 DAT	45 DAT		
1	Healthy 100 %	0.5	0.133	0.13	0.70	0.70	0.70	0.13
2	Seedling infection 5 %	1.33	0.200	0.70	2.00	2.33	1.10	0.43
3	Seedling infection 10 %	1.00	0.400	1.03	3.00	3.00	3.00	0.43
4	Seedling infection 15 %	1.33	0.700	1.33	3.33	3.67	3.00	1.33
5	Seedling infection 20 %	1.67	0.700	2.33	4.00	4.67	3.00	1.67
6	Seedling infection 25 %	2.33	0.400	1.33	3.67	5.33	3.33	2.00
7	Seedling infection 30 %	2.67	0.700	2.67	6.00	6.00	4.33	1.67
8	Seedling infection 35 %	3.00	1.333	2.33	6.00	5.00	3.33	1.67
9	Seedling infection 40 %	3.33	2.000	3.67	5.00	7.33	4.67	2.67
10	Seedling infection 45 %	4.67	2.000	4.00	5.67	6.67	4.00	2.67
11	Seedling infection 50 %	5.33	2.000	3.67	6.33	6.33	5.00	1.33
12	Seedling infection 55 %	6.00	2.333	5.00	5.00	8.00	5.00	3.00
13	Seedling infection 60 %	6.33	1.333	4.67	6.00	11.67	7.67	4.00
14	Seedling infection 65 %	7.67	1.333	4.67	7.00	14.00	9.33	4.67
15	Seedling infection 70 %	9.00	1.333	6.00	6.00	9.00	6.00	3.00
16	Seedling infection 75 %	10.67	0.700	5.00	5.00	12.00	6.00	6.00
17	Seedling infection 80 %	11.67	2.000	5.33	5.00	8.00	5.00	3.67
18	Seedling infection 85 %	13.00	1.667	6.67	6.33	8.33	4.67	3.67
19	Seedling infection 90 %	14.33	2.000	3.00	4.00	7.33	5.00	3.33
20	Seedling infection 95 %	16.00	2.000	4.00	4.00	6.33	4.00	2.33
21	Seedling infection 100 %	17.67	1.333	3.00	4.00	4.00	2.00	2.67
22	Single plant infection 100 %	17.33	0.200	2.33	4.00	5.00	2.67	2.33
23	Single plant healthy 100 %	2.00	0.133	0.70	1.33	1.67	1.33	0.40
L.S.D. 5 %		1.547	1.124	1.728	2.059	1.751	1.625	1.575

*** DAT = Days after transplanting**

Table (6) : Effect of infection levels of bakanae rice disease (*Fusarium . moniliforme*) on yield losses under screenhouse conditions, season 2014.

No.	Treatment	Height (cm)		Total branch	1000 g weight (g)	Biomass g/plot	Yield g/plot	Yield losses (%)	Yield ton/fed
		Healthy	infected						
1	Healthy 100%	85.00	117.00	96.00	27.833	274.33	188.33	-	4.745
2	Seedling infection 5 %	87.00	120.67	95.00	27.633	274.33	182.33	3.18	4.594
3	Seedling infection 10 %	86.50	119.67	96.33	27.233	272.33	177.33	5.84	4.468
4	Seedling infection 15 %	86.33	120.00	93.00	27.633	269.33	169.33	10.08	4.266
5	Seedling infection 20 %	86.00	119.33	92.00	27.167	270.00	162.00	13.98	4.082
6	Seedling infection 25 %	85.00	120.67	91.33	26.567	260.00	154.33	18.05	3.888
7	Seedling infection 30 %	86.50	122.33	93.00	26.333	259.33	151.67	19.46	3.821
8	Seedling infection 35 %	87.67	117.67	90.00	26.300	253.33	147.67	21.58	3.720
9	Seedling infection 40 %	88.33	116.00	87.33	25.967	249.33	144.67	23.18	3.645
10	Seedling infection 45 %	86.67	115.00	85.00	25.233	239.67	139.67	25.83	3.519
11	Seedling infection 50 %	86.00	119.00	81.33	25.433	235.00	131.33	30.26	3.309
12	Seedling infection 55 %	85.00	119.33	74.00	24.900	218.33	127.33	32.38	3.259
13	Seedling infection 60 %	87.00	117.00	68.67	23.767	205.00	120.00	36.28	3.023
14	Seedling infection 65 %	86.33	111.00	66.67	23.300	193.67	110.67	41.23	2.788
15	Seedling infection 70 %	89.00	113.67	56.67	22.333	165.67	100.33	46.72	2.528
16	Seedling infection 75 %	86.33	124.67	48.67	22.033	122.33	84.33	55.22	2.124
17	Seedling infection 80 %	84.00	123.33	34.00	21.400	99.67	73.00	61.23	1.839
18	Seedling infection 85 %	85.83	123.67	27.00	17.933	80.67	62.67	66.67	1.579
19	Seedling infection 90 %	84.83	123.67	18.33	16.833	62.67	52.67	72.03	1.326
20	Seedling infection 95 %	85.67	114.67	15.00	15.500	46.33	43.00	77.16	1.083
21	Seedling infection 100 %	87.43	124.00	13.00	12.400	40.00	32.67	82.65	0.823
22	Single plant infection 100 %	86.83	116.00	11.00	11.267	36.00	29.00	84.94	0.730
23	Single plant healthy 100 %	87.83	116.00	104.67	28.233	286.0	192.67	-	4.854
L.S.D. 5%		2.939	7.224	3.829	1.165	9.315	4.651	-	0.117

7. Effect of spraying fungicides at heading stage on rice bakanae disease:-

A field experiment was conducted at the Experimental Farm of Rice Research and Training Center (RRTC), Sakha, Kafr EL-Sheikh during 2013 and 2014 seasons to investigate the effect of different fungicide sprays on production of healthy seeds free from *Fusarium moniliforme* the causal of bakanae rice disease. Data present in Table (7) indicat that, the highest grain yield (4.962, 4.899 and 4.856 t/fed) spray at 5 DAF followed by (4.826, 4.685 and 4.697 t/fed) by spray at 15 DAF were obtained by using Beam, Sumi8 and Vitavax respectively compared to control (4.125 t/fed) at 2013 season. As regard to germination percent, the highest germination (92.33 and 88.0 %) came back to spray at 5 DAF followed by (87.33 and 86.0 %)due to spray at 15 DAF obtained from Sumi8 and Beam respectively in comparison with control (61.33 %) at 2013 season. As for infection percent, the lowest infection (5.67 and 6.0 %) were recorded with spray at 5 DAF by Sumi8 and Beam respectively compared by control (72.67 %) at 2013 season. Krishanaveni *et al*., (2003) reported that the screening and selection against *Fusarium. moniliforme* incidence and severity were correlated significantly and negatively with the yield. Different seed treatment fungicides i.e., Benlate (benomyl), Derosal (carbendazim), Kasuran (kasugamycin), Healthied, Topsin-M (thiophanate-methyl), Dithane M-45 (mancozeb), Bayleton (triadimefon) and Ridomil (metalaxyl) were used in controlling bakanae disease of rice which gave significant reduction in disease incidence and increased seed germination and yield (Dodan *et al.,* 1994; Javaid and Ilyas 1995; Tateishi *et al*., 1998; Biswas and Das 2002; Kabir *et al*., 2006 and Bagga and Sharma 2006).

Table (7) : Effect of some fungicides sprays at heading stage on rice bakanae disease (*Fusariu moniliforme*) on Sakha 101 cultivar in the field during 2013 and 2014 seasons.

No.	Treatment	2013			2014		
		Yield ton/fed	% germination	% infection	Yield ton/fed	% germination	% infection
1	Vitavax 200 5 days flowering	4.856	81.00	11.67	4.926	98.67	18.00
2	Vitavax 200 15 days flowering	4.697	76.67	17.67	4.768	87.00	21.67
3	Vitavax 200 25 days flowering	4.562	72.33	25.67	4.430	81.67	29.67
4	Beam 75 % 5 days flowering	4.962	88.00	6.00	4.987	93.33	14.00
5	Beam 75 % 15 days flowering	4.826	86.00	11.67	4.812	87.00	27.67
6	Beam 75 % 10 days flowering	4.356	81.67	27.00	4.530	83.67	39.67
7	Fuji-one 40 % 5 days flowering	4.658	83.33	16.00	4.680	86.67	24.33
8	Fuji-one 40 % 15 days flowering	4.536	80.00	25.00	4.513	80.00	41.33
9	Fuji-one 40 % 25 days flowering	4.365	71.33	39.33	4.482	73.67	50.33
10	Sumi 8 5 % 5 days flowering	4.899	92.33	5.67	4.926	92.00	15.00
11	Sumi 8 5 % 15 days flowering	4.685	87.33	17.67	4.816	87.00	20.00
12	Sumi 8 5 % 25 days flowering	4.361	82.00	24.33	4.515	81.00	35.00
13	Copral 50 % 5 days flowering	4.532	67.67	48.33	4.502	82.67	54.00
14	Copral 50 % 15 days flowering	4.395	69.00	51.67	4.381	71.67	52.67
15	Copral 50 % 25 days flowering	4.320	64.67	55.33	4.395	69.33	59.33
16	Control	4.125	61.33	72.67	4.200	67.33	65.67
L.S.D. 5 %		0.136	5.004	7.087	0.175	5.947	8.002

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تقيـيم بعـض أصـناف الأرز والسـلالات المبشـرة للفطـر فيوزاريـوم مونيليفـورم (فرتيسيلويدز) المسبب لمرض البكانا في الأرز وتقدير الخسارة في المحصول.

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أجريت هذه الدراسة في المعمل والصوبة والمزرعة البحثية لمركز البحوث والتدريب في الأرز سخا، كفر الشيخ خلال مواسم ٢٠١٣ و ٢٠١٤ و ٢٠١٥ . وأشارت النتائج أن جميع العزلات المختبرة تفاوتت في القدرة المرضية. وكانت اقوي العزلات في الشدة المرضية هي العزلات (١٢، ٢ و ١) و التي أعطت اعلي نسبة إصابة (٧٩,٣٣، ٧٣,٣٣ و ٧٢,٠٠ % على التوالي) كانت معزولة من نباتات الأرز. في حين إن العزلة رقم ٦ كانت غير قادرة علي الإصابة وإظهار عرض البكانا كانت معزولة من نبات الذرة. كما أشارت النتائج إلى أن جميع أصناف الأرز والسلالات المبشرة التي تم اختبارها. اختلفت في قابليتها للإصابة بعزلات الفطر فيوزاريوم فرتيسيلويدز. ومع ذلك، كانت الأصناف سخا ١٠١ و الجيزة ١٧٧ وبعض الهجن مثل هجين ١ وهجين ٢ الأكثر حساسية للإصابة بالمرض . انخفضت نسبة البقاء والقدرة علي إحداث العدوى الفطر فيوزاريوم فرتيسيلويدز في الحبوب المصابة أو التربة بعد شهرين من التخزين وحتي١٠ شهور ثم انخفضت بشكل كبير بعد عامين من التخزين علي درجة حرارة الغرفة وفي الصوبة السلكية. وأشارت النتائج التي تم الحصول عليها أن أكبر عدد من النبات المصابة والتي حدث لها موت قبل الظهور فوق سطح التربه (١٧,٦٧ نبات / معاملة) من البادرات المصابة بنسبة ١٠٠٪ بعد ١٠ أيام من الشتل مقارنة ب (٠,٥ نبات / معاملة) من بادرات سليمة ١٠٠٪. في حين تم الحصول على أكبر عدد من النباتات المصابه (١٤ نبات / معاملة) من البادرات المصابة بنسبة ٦٥٪ تليها (١٢,٠٠نبات / معاملة) من البادرات المصابة بنسبة ٧٥٪ بعد ٤٥ يوم من الشتل مقارنة (٠,٧ نبات / معاملة) من نباتات سليمة بنسبة ١٠٠٪. وأشارت النتائج إلى أن أعلى محصول الحبوب (٤,٩٦٢، ٤,٨٩٩ و ٤,٨٥٦ طن / فدان) تم الحصول عليها من المعاملة بالرش بعد خمسة أيام من بداية طرد السنابل تليها (٤,٨٢٦، ٤,٦٨٥ و ٤,٦٩٧ طن / فدان) تم الحصول عليها من المعاملة بالرش بعد خمسة عشرة يوم من بداية الطرد للمبيدات البيم والسومي ٨ والفيتافاكس على التوالي مقارنة بالمعاملة بدون مبيد كانت (٤,١٢٥ طن / فدان) وغيرها من المبيدات الأخرى في موسم ٢٠١٣.