Physiological Specialization of *Pyricularia grisea* on Rice in Egypt From 2008 to 2010

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A total of 142 isolates of *Pyricularia grisea* were isolated from five Governorates and tested for pathotype diversity on rice seedlings of eight international differential varieties, included twelve Egyptian commercial cultivars and ten Japanese differential varieties (JDVs). Tested isolates were differentiated into thirty pathotypes. For the year 2008, IH race group represented the most frequent phenotype, being 42.22%, followed by IG and II, being 15.56% for both. However, in 2009, ID and IG were the common race groups, being 37.88 and 22.72% in respectively. In 2010, ID race group was still the common race group (64.52%), followed by IB (19.35%) and IC (12.9%). Egyptian commercial rice cultivars tested against *P. grisea* isolates displayed variable reactions. Cultivars Giza 178, Sakha 102, Sakha 103 and Hybrid 1 were resistant to all isolates. While, cvs. Giza 177, Reiho and Giza 181 rice cultivars were infected by 5, 10 and 17 out of the tested isolates, respectively. The susceptible rice cultivars Giza 176, Sakha 104, Giza 159, Sakha 101 and Giza 171 displayed different levels of susceptibility. Concerning the effectiveness of resistance genes, Pi-Z gene was the highest effective in blast control with 93.80%, followed by Pi-i plus Pi-K^s with 87.22%. In opposite, Pi-ta^2 and Pi-a were the least effective genes with 55.77 and 22.78%, respectively. The variability in the pathogen and the resistance identified of some genotypes used in this study are further investigated to develop superior, adapted germplasm for use in rice breeding programs in Egypt.

Keywords: Pathogenicity, physiological races, *Pyricularia grisea,* resistance genes and rice blast.

Rice (*Oryza sativa* L.) is the second most important cereal crop in Afro-Asian continent. It constitutes the essential food for about 54% of the world's population. In Egypt, rice is the second staple food after wheat, and is very important for local consumption and for exportation. The annual area planted with rice is about 598,319 hectares. Paddy production is about 5,909,596 tons and the national yield productively is about 9.88 t/ha (Anonymous, 2012). Rice blast disease caused by *Pyricularia grisea* (the imperfect stage of *Magnaporthe grisea*) is widespread and damaging disease in most rice growing areas of the world (Ou, 1985). Rice diseases, especially rice blast, may affect annual rice production by about 5% in normal or mildly infected seasons. In epidemic seasons, yield losses may reach as high as 30-50% (Sehly *et al.,* 2002a). The most economic and safety way to control this

disease is by cultivating resistant cultivars. Unfortunately, in most cases, the resistance of these cultivars is overcome in few years after cultivar release (Veillet *et al.,* 1996 and Sehly *et al.,* 2008). Many physiological races were identified on the international differential varieties (IDVs) to different groups as international proposed races, *i.e.* IA, IB, IC, ID, IE, IF, IG, IH and II (Atkins *et al.,* 1967; EL-Wahsh and Ammar, 2007 and Sehly *et al.,* 2008). Prabhu *et al.* (2002) examined the pathogenic diversity of *P. grisea* isolates retrieved from 14 upland rice cultivars for five years. Inoculation was performed on 32 genotypes with 85 mono-conidial isolates under controlled greenhouse conditions. Based on the reaction pattern of eight IDVs, eleven pathotypes IB-1, IB-9, IB-41, IB-45, IB-33, IB-37, IC-1, IC-9, IC-25, ID-9 and IG-1 were identified. Dias Neto *et al.* (2010) evaluated diversity of 479 *P. grisea* pathotypes on new commercial irrigated rice fields in Brazil. The reaction of 250 *P. grisea* isolates from three trap nurseries indicated the presence of 45 international *P. grisea* races belonging to seven pathotypes (physiological races) groups IA - IG. Analysis of 229 isolates, from scattered fields, revealed the presence of 33 different international races belonging to eight different pathotypes groups.

In Egypt, many investigators studied the physiological races of rice blast. Sehly *et al.* (2000) inoculated 45 *P. grisea* isolates on IDVs. They found that IH-1 (36.6%) was the most common race, followed by ID, IA and IG race groups with 17.8, 13.3 and 13.3%, respectively, while, avirulent race group II was 9%. EL-Shafiey (2002) isolated 8 and 15 *P. grisea* isolates from different rice cultivars and weeds, respectively. The blast rice isolates were related to six races on IDVs, while weed isolates were related to four races. Also, Gabr (2004) collected 40 isolates of *P. grisea* from different rice cultivars and weeds. These isolates were identified to six race groups as (IB, IC, ID, IF, IG and IH). Marchetti (1994) mentioned that in the last 30 years, the Pi-K^h and Pi-ta^2 genes have been introduced into US rice cultivars; already present with Pi-Z, Pi-i, Pi-a and Pi-K^s genes. The most effective of these against US pathotypes of *P. grisea* are Pi-ta^2, Pi-K^h and Pi-Z. Sehly and Bastawisi (1995) evaluated 20 single isolates in each season in the period from 1988 to 1993. They stated that Pi-a in Aichi Asahi, Pi-K^s in Shin 2 and Pi-ta in Yashiro-mochi showed lower levels of resistance. Pi-i plus Pi-K^s in Ishikarishiroke and Pi-ta^2 in Pi No.4 showed moderate level of resistance. However, Pi-Z in Fukunishiki and its allele Pi-Z^t in Toride1 were completely resistant in 1988 only. While, Pi-K in Kanto 51, Pi-K^m in Tsuyake and Pi-b in BL-1 showed high level of resistance genes in the same period. Malavolta *et al.* (2009) tested 71 monosporic of *P. grisea* collected from Brazil during 2004-2006. They found that *pi-ta*^{2} gene in Pi No 4 was resistant to 71 tested isolates. While, 63.4, 59.1 and 53.5% of these isolates were virulent on cvs. Aichi Asahi, Yashiro-mochi and Ishikarishiroke, respectively. The introductions of new blast virulent races, possess a great challenge for breeders in their efforts to achieve durable resistance to the *P. grisea*. Identification of virulence phenotypes in rice blast populations is crucial for development of resistant cultivars.

This investigation was aimed (i) to study the pathogenic diversity and geographical distribution of *P. grisea* populations from region to region and from season to another under Egyptian conditions, (ii) to study the current status of *P. grisea* isolates on commercial rice varieties and (iii) effectiveness of blast resistant genes (Pi- genes) in JDVs against *P. grisea* isolates.

Materials and Methods

Blast sample collection from rice:

One hundred forty two isolates of *P. grisea* were isolated from 19 rice entries, *i.e.* Sakha 101, Sakha 104, Giza 159, Giza 171, Nishihikari, Fukunishiki, BL-1, M 202, M201, Pi.No4, Usen, Reiho, B9C-MD-3-3, B46-1B-PN-3, IR70554-48-1-2, IRTP21662 (IRB- LA-A), Hybrid 2, IR-82225-11-3-1 and IR-82737-B-182. Blast isolates were collected from five rice Governorates, *i.e.* Beheira, Dakahliya, Gharbiya, Kafr El-Sheikh and Sharkiya during 2008, 2009 and 2010 growing seasons (Table, 1).

Preparation of tested inducers:
Isolation and preservation of rice blast fungus (P. grisea):

This study was carried out at the greenhouses of Rice Pathol. Lab, Plant Pathol. Res. Inst., ARC and the Fac. Agric., Kafr El-Sheikh Univ. Blast infected rice leaves and panicles were cut into 1-2 cm pieces, sterilized by immersing in 1.0% sodium hypochlorite solution (NaOH) for 2 min, rinsed twice with sterilized distilled water and placed onto sterilized filter paper in Petri dishes, then incubated at $25\pm2^{o}C$ for 24 h under continuous fluorescent light. Single conidium, of each isolate, was generated by streaking conidia from sporulating lesions on water agar medium for 24 hr. The germinating single conidia were picked up and transferred onto water agar medium for additional 24 hours. The tip of a single hyphae was cut and grown on banana dextrose agar medium (BDA) on 3x3 cm disc of sterilized filter paper. When the filter papers were completely occupied by the fungal growth, the paper discs were individually transferred into dry sterilized Petri dishes. About one week later, the dried filter papers having the fungus isolates were cut into small pieces. Pieces obtained from each isolate were altogether introduced into a plastic vial and kept at $-20^{o}C$ for long term storage according to the technique of Mekwatanakarn *et al.* (1999).

Inoculum preparation:

Five plates, inoculated by each isolate, were incubated at $28\pm2^{o}C$ for 10 days. All Petri dishes were left under fluorescent light for 4 days to enhance the sporulation, which estimated by adding 10 ml distilled water to each Petri dish, and then the spores were harvested by spatula. The filtrate was received in a test tube and stirred using the electrical shaker. Spores were counted using the haemocytometer slide and adjusted to $5x10^{4}$ spores ml^{-1} for pathogenicity tests.

Race identification and pathogenicity test:

Eight international differential rice varieties, *i.e.* Raminad str.3, Zenith, NP-125, Usen, Dular, Kanto 51, CI 8970 S and Caloro (Atkins *et al.,* 1967; Ling and Ou, 1969) and 12 Egyptian commercial rice cultivars (Giza 159, Giza171, Giza 176, Giza 177, Giza 178, Giza 181, Reiho, Sakha 101, Sakha 102, Sakha 103, Sakha 104 and Hybrid 1) were tested to determine the pathotypes and pathogenicity test. In addition, ten Japanese differential varieties, *i.e.* Shin 2 (Pi-K^{s} gene), Toride 1(Pi-Z^{t}), Tusyake (Pi-K^{m}), Kanto 51(Pi-K), Fukunishiki (Pi-Z), Ishikarishiki (Pi-i-Pi-K$^{s)}$, BL-1 (Pi-b), Yashiro-Mochi (Pi-ta), Pi No. 4 (Pi-ta^{2}) and Aichi Asahi (Pi-a) were used to determine the effective resistant genes. All the tested entries were seeded in

plastic trays (30x20x15 cm.). Each tray comprised 10 rows representing eight genotypes and two susceptible rice checks (Sakha 101 and Sakha 104). The trays were kept in the greenhouse at 27 ± 3°C, and fertilized as recommended. Seedlings at 3-4-leaf stage were inoculated by spraying with 100 ml of spore suspension of each isolate by electrical spray gun. Gelatine was added to the spore suspension at a concentration of 2.5 g/l^{-1} (Bastiaans, 1993) to enhance the adhesion of spores on leaf surfaces. The inoculated seedlings were held in a moist chamber with more than 90% R.H. and 27 ± 1°C for 24 h, and then kept under the greenhouse conditions.

Disease assessment:

Seven days after inoculation under greenhouse conditions, blast reaction was scored as the typical blast lesions according to the standard evaluation system (Anonymous, 1996) using 0-9 scale.

R e s u l t s

A total of 142 rice blast isolates were collected during the three growing seasons, 2008, 2009 and 2010 (Table 1). The highest number of isolates was collected from Kafr El-Sheikh (69), followed by Beheira (24), Sharkiya (20), Gharbiya (18), and Dakahliya (11 isolates). Moreover, results presented in Table (2) reveal that the highest number of collected isolates was recorded during season 2009 (66) followed by season 2008 (45) and season 2010 (31 isolates).

Identification of P. grisea physiological races:

The identification of *P. grisea* physiological races was carried out on the bases of the compatibility of the isolates with eight international differential varieties. Results in Table (1) reveal that 30 pathotypes were identified. Only one isolate was found to be related to group IA (IA-41); 12 isolates related to group IB, *i.e.* one isolate for each of IB-29 and IB-47 and two isolates related to each of IB-33 and IB-63 and three isolates related to each of IB-41and IB-61. Fifteen isolates of group IC, *i.e.* one isolate related to each of IC-12, IC-27, IC-31 and IC-32, two isolates related to each of IC-9 and IC-29 as well as seven isolates related to IC-13. Also, 46 isolates of group ID, *i.e.* one isolate related to each of ID-1, ID-7, ID-10, ID-11 and D-16, three isolates related to each of ID-5 and ID-9 and five isolates related to ID-15 and 30 isolates related to ID-13. One isolate of group IE (IE-7) as well as four isolates of group IF, *i.e.* one isolate related to IF-1 and three isolates related to IF-3. Moreover, 23 isolates of group IG, *i.e.* three isolates related to IG-2 and 20 isolates related to IG-1. Also, 33 isolates of group IH (IH-1) and seven isolates of group II (avirulent group).

Geographical distribution of the physiological races:

The distribution of physiological blast races at the five rice Governorates during 2008-2010 period is presented in Table (1). Results show that all race groups were represented at Kafr El-Sheikh Governorate, regardless of its numbers, was considered a hot spot for rice blast infection with 48.59% of the total identified races. However, Beheira came in the second rank with 16.90% of the total identified races, and all race groups were represented except IA, IE, IF and II race groups, followed by Sharkiya and Gharbiya with 14.08 and 12.68%, respectively. Moreover,

Table 1. Geographical distribution and pathotypes of *P. grisea* isolates collected from five rice growing Governorates during 2008 to 2010 seasons, based reaction of eight international differential varieties under greenhouse conditions

Pathotype	Governorate/season														
	Beheira			Dakahliya			Gharbiya			Kafr El-Sheikh			Sharkiya		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
IA-41	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
IB-29	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-
IB-33	-	-	-	-	-	-	-	-	-	-	-	2	-	-	-
IB-41	-	-	1	-	-	-	-	-	-	-	-	2	-	-	-
IB-47	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-
IB-61	1	-	-	1	-	-	-	1	-	-	-	-	-	-	-
IB-63	-	-	-	1	-	-	-	1	-	-	-	-	-	-	-
IC-9	-	-	-	-	-	-	-	-	-	-	1	1	-	-	-
IC-12	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
IC-13	-	1	-	-	-	-	-	1	-	-	3	2	-	-	-
IC-27	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
IC-29	-	-	-	-	-	-	-	-	-	1	-	1	-	-	-
IC-31	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-
IC-32	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-
ID-1	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-
ID-5	-	-	-	-	-	-	-	1	-	-	1	1	-	-	-
ID-7	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-
ID-9	-	-	-	-	-	-	-	2	-	-	-	1	-	-	-
ID-10	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-
ID-11	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
ID-13	-	6	2	-	-	2	-	4	1	-	4	7	-	-	4
ID-15	-	3	-	-	-	-	-	-	-	-	2	-	-	-	-
1D-16	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-
IE-7	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-
IF-1	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-
IF-3	-	-	-	-	-	-	-	-	-	3	-	-	-	-	-
IG-1	1	1	-	-	-	-	-	3	-	3	4	-	1	5	1
IG-2	1	-	-	-	1	-	-	-	-	1	1	-	-	-	-
IH-1	2	2	-	2	1	-	-	2	-	13	4	-	2	5	-
II-1	-	-	-	3	-	-	-	-	-	3	-	-	1	-	-
Total	24			11			18			69			20		
Frequency (%)	16.90			7.75			12.68			48.59			14.08		

A= 2008 season   B= 2009 season   C= 2010 season

results indicate that race IH-1 was the most frequent included 13 isolates of the 142 tested isolates, followed by ID-13 and IG-1 that included 30 and 20 isolates, respectively. However, pathotypes IA-41, IB-29, IB-47, IC-27, IC-31, IC-32, ID-1, ID-7, ID-10, ID-11, ID-16, IE-7 and IF-1 recorded the lowest frequency, when included one isolate for each race.

The population dynamics of P. grisea:

Results presented in Table (2) show that in the first season (2008), IH group race was the common one as represented by 42.2%, followed by II, IG with 15.6% for each one and IF with 8.9%. While, IB, IC, ID and IE were the minor race groups, and IA group race was absent. In 2009 season, the population dynamics of *P. grisea* was high as compared with the other seasons. The common race groups were ID (37.9%), IG (22.7%) and IH (21.2%) followed by IC which changed from a minor to a major race group with 12.1%. In the last season (2010), ID remained as the most common race group (64.5%), followed by IB (19.3%) and IC group race (12.9%). The race groups, IA, IE, IF, IH and II were absent. During the study period, ID was generally the most common race group, followed by IH and IG groups, While IA, IE and IF were of minor occurrence race groups.

Table 2. Number and percentage of *P. grisea* common race groups collected from 5 rice growing Governorates during 2008, 2009 and 2010 seasons

Race group	Growing season						Total	Common race (%)
	2008		2009		2010			
	Number	%	Number	%	Number	%		
IA	0	0.0	1	1.5	0	0.0	1	0.7
IB	3	6.7	3	4.5	6	19.3	12	8.4
IC	3	6.7	8	12.1	4	12.9	15	10.6
ID	1	2.2	25	37.9	20	64.5	46	32.4
IE	1	2.2	0	0.0	0	0.0	1	0.7
IF	4	8.9	0	0.0	0	0.0	4	2.8
IG	7	15.6	15	22.7	1	3.2	23	16.2
IH	19	42.2	14	21.2	0	0.0	33	23.2
II	7	15.6	0	0.0	0	0.0	7	4.9
Total	45	-	66	-	31	-	142	-

Reaction of commercial rice cultivars to P. grisea isolates

According to the reactions of the tested genotypes against different isolates of *P. grisea* under controlled greenhouse conditions, differences in the levels of resistance among cultivars became apparent. Results presented in Table (3) show that out of the 12 commercial rice genotypes tested only 4 cultivars, *i.e.* Hybrid 1, Giza 178, Sakha 102 and Sakha 103, were resistant to all of the tested isolates. On the other hand, rice genotypes, cvs. Giza 177, Reiho and Giza 181, were infected by 5, 10 and 17 isolates and had 97.0, 92.2 and 88.0% resistance, respectively. The next most susceptible group of cultivars, *i.e.* Sakha 104, Giza 176 and Giza 159, were attacked by 49, 58 and 68 isolates of the 142 tested isolates. Meanwhile, genotypes cvs. Sakha 101 and Giza 171 were the most susceptible; where they were challenged by 95 and 99 isolates with resistance level 33.3 and 29.7%, respectively.

Table 3. Number of pathogenic isolates and resistance percentage of the tested commercial cultivars against 142 blast isolates under greenhouse conditions during 2008, 2009 and 2010 growing seasons

Tested cultivar and hybrid	No. of pathogenic isolates			Resistance (%)			
	2008	2009	2010	2008	2009	2010	Mean
Hybrid 1	0	0	0	100.0	100.0	100.0	100.0
Giza 159	19	32	17	57.8	51.5	45.2	51.5
Giza 171	28	47	24	37.8	28.8	22.6	29.7
Giza 176	21	33	4	53.3	50.0	87.1	63.5
Giza 177	2	3	0	95.5	95.5	100.0	97.0
Giza 178	0	0	0	100.0	100.0	100.0	100.0
Giza 181	0	11	6	100.0	83.3	80.6	88.0
Reiho	2	4	4	95.5	93.9	87.1	92.2
Sakha 101	26	47	22	42.2	28.8	29.0	33.3
Sakha 102	0	0	0	100.0	100.0	100.0	100.0
Sakha 103	0	0	0	100.0	100.0	100.0	100.0
Sakha 104	11	22	16	75.6	66.7	48.4	63.5

Effectiveness of blast resistant genes

According to the reaction of ten Japanese differential varieties (JDVs) against 142 tested isolates, the resistant blast genes (Pi- genes) effectiveness ranged from 22.8 to 93.8% resistance (Table 4). Pi-Z gene from Fukunishiki variety was highly effective in blast control with 93.8% resistance, followed by Pi-i plus Pi-Ks from Ishikarishiroke with 87.2% resistance, Pi-K from Kanto 51 with 83.3% resistance and Pi-b from BL-1 with 83.1% resistance. On the other hand, Pi-ta^2 from Pi No.4 and Pi-a from Aichi-Asahi were the less effective genes against *P. grisea* isolates with 55.8 and 22.8% resistance, respectively.

Table 4. Evaluation of effective resistant genes in JDVs against blast fungus isolates under greenhouse conditions during 2008, 2009 and 2010 seasons

Tested variety	Pi-gene	No. of virulent isolates			Effective resistant gene (%)			
		2008	2009	2010	2008	2009	2010	Mean
Fukunishiki	Pi-Z	2	3	3	95.6	95.4	90.3	93.8
Ishikarishiroke	Pi-i-Pi-K^s	3	6	7	93.3	90.9	77.4	87.2
Kanto 51	Pi-K	4	8	9	91.1	87.9	71.0	83.3
BL-1	Pi-b	5	9	8	88.9	86.4	74.2	83.1
Yashiro-Mochi	Pi-ta	8	11	11	82.2	83.3	64.5	76.7
Shin 2	Pi-K^s	8	23	9	82.2	65.1	71.0	72.8
Toride 1	Pi-Z^t	11	17	12	75.6	74.2	61.3	70.4
Tusyake	Pi-K^m	19	9	14	57.8	86.4	54.8	66.3
Pi No.4	Pi-ta^2	15	23	20	66.7	65.1	35.5	55.8
Aichi Asahi	Pi-a	29	55	26	35.6	16.7	16.1	22.8

Results in Table (5) indicate that Aichi Asahi variety which has Pi-a gene was infected by 110 isolates out of 142 tested isolates. Pi No.4 was in the second rank where it infected by 58 isolates, followed by Tusyake, Toride 1 and Shin 2 rice varieties which have Pi-K^m, Pi-Z^t and Pi-K^s genes which were infected by 42, 40 and 40 isolates, respectively. Pi-Z gene from Fukunishiki variety was infected by eight isolates out of 142, followed by Pi-i plus Pi- Ks from Ishikarishiroke that was infected by 16 isolates. ID group race was highly virulent group on JDVs, followed by IC, IH and IB group races. IE, IA and II were the lowest infected race groups on JDVs by 6, 7 and 7 races, respectively.

Table 5. Number of blast physiological races infecting JDVs under greenhouse conditions during 2008, 2009 and 2010 seasons

Tested JDVs	Race group									Total
	IA	IB	IC	ID	IE	IF	IG	IH	II	
Fukunishiki	0	2	3	2	0	0	1	0	0	8
Ishikarishiroke	1	4	2	7	0	0	1	1	-	16
Kanto 51	1	5	5	6	0	4	0	0	0	21
BL-1	-	3	3	10	-	-	2	3	1	22
Yashiro-Mochi	-	3	3	13	1	-	4	5	1	30
Shin 2	1	5	8	12	1	1	8	4	0	40
Toride 1	1	7	5	15	1	1	6	4	0	40
Tusyake	1	7	8	9	1	3	5	7	1	42
Pi No.4	1	9	10	18	1	2	5	10	2	58
Aichi Asahi	1	12	14	35	1	2	17	26	2	110
Total	7	57	61	127	6	13	49	60	7	

Discussion

Rice blast, caused by *P. grisea* is one of the most serious biotic stresses in the tropics and subtropics (Veillet *et al.,* 1996 and Sehly *et al*., 2008). A total of 142 isolates were collected during growing seasons of 2008, 2009 and 2010, *i.e.* 45, 66 and 31 isolates, respectively, from five rice Governorates, *i.e.* Beheira, Dakahliya, Gharbiya, Kafr El-Sheikh and Sharkiya. These results are in agreement with the findings of EL-Wahsh (1997) who reported that the comparison of epidemics and physiological races in different locations is often different, since epidemics may vary from year to another and from location to another.

Thirty pathotypes were identified under nine race groups, IA up to II groups according to their reaction on IDVs. Many investigators studied the physiological races (Correa-Victoria and Zeigler, 1993; Chen *et al.,* 2001 and Prabhu *et al.,* 2002). Also, Hunts *et al.* (1986) collected 29 isolates of *P. grisea* from worldwide sources, the isolates were classified into 21 physiological races as IA-1, IA-65, IA-109, IB-1, IB-4 , IB-33, IB-45, IC-17, IC-17A, IC-19, ID-1, ID-8, ID-9, ID-13, ID-15, IE-3, IF-1, IG-1, IG-2, IH-1 and II according to the same genotypes. Sehly *et al.* (1993) identified 60 isolates collected in 1989, 1990 and 1991 growing seasons on the same

genotypes with 20 isolates in each season. They mentioned that five race groups were identified in the three seasons, including IC, ID, IG, IH and II. The identified races in 1989 were IG-1, IH-1, IC-25, IC-29, ID-5and II. In the second season, they were ID-13, ID-15, IC-17, IC-25, IC-29 and IH-1. In 1991 season, ID-13, ID-15, IH-1, IG-1, IC-3, IC-13 and IC-21 were identified. Nutsugah *et al.* (2008) collected 71 *P. grisea* isolates from seven regions where rice is grown in Ghana. The isolates were grouped into 25 pathotypes as IB-1, ID-13, IB-45, ID-1, IB-13, IC-1, IH-1, IB-9, IC-9, ID-9, IC-13, IC-25, IA-9, IF-1, II-1, IB-61, IB -21, IA-2, IB-5, IB-7, IG-1, IC-29, IC-17, IA-88 and IB-29. Dias Neto *et al.* (2010) evaluated the *P. grisea* pathotypes diversity in new commercial rice fields in Brazil. They collected 479 isolates from three trap nurseries and scattered sites. These isolates were identified by the international rice blast differential set. They obtained 85 isolates from the first trap nursery represented race groups IA, IB, IC, ID and IE, 70 isolates from the second trap nursery representing four race groups, *i.e.* IA, IB, IC and ID and ninety five isolates from the third trap nursery represented IA, IB, IC, ID, IE, IF and IG groups. The rest (229 isolates) were collected from scattered fields, represented all group races, except IH race group. They added that the most common race groups were ID, which represented 31.95% from the total races followed by IH and IG race groups. Ou (1985) and Coley (1992) reported that field resistance is effective and important, because complete resistance is subject to be broken down after some years of variety release. This breakdown is due to the variability of the isolates of blast pathogen under field conditions. Sehly *et al*. (2008) identified more than 200 blast isolates collected from 1994 up to 2006 growing seasons in the greenhouse on IDVs as used in the present study under artificial inoculation conditions. They reported that from 1994 up to 1996, IC race group was the common race followed by IB, ID and IG groups, while IA race group was absent. From 1997 up to 2000, all groups were represented; IH and IG groups were the common races. In the third period 2001-2004, IG race group was the common race because it was specific on Sakha 101 rice cultivar and was responsible for its breakdown. In the last period 2005-2006, IG group still was the common race followed by IB and IC race groups. On the other hand, IA, IE, IH and II were absent. The common physiological races suggest the existence of a wide genetic variability of blast race in Egypt. The knowledge of the diversity and prevalence of the physiological races of *P. grisea* fungus in rice production areas is an important step for devising strategies to develop and release new resistant cultivars. These results are in harmony with those of many researchers (El-Kazzaz, 1973; Hunts *et al.,* 1986; Araujo *et al.,* 2005; EL-Wahsh and Ammar, 2007 and Sehly *et al.,* 2008). This may be due to the high level of variability of *P. grisea* especially for the isolates obtained from susceptible rice cultivars. The wide variation of the physiological races of blast isolates in Africa might have been created due to the pressure caused by the diversities of rice cultivars (Telebanco-Yanoria *et al*., 2008).

The twelve commercial rice cultivars showed different responses to blast infection. Pathogenicity test revealed that some rice cultivars, *i.e.* Giza 178, Sakha 102, Sakha 103 and Hybrid 1 were resistant to all isolates, Giza 177, Reiho and Giza 181 were infected by 5, 10 and 17 out of 142 tested isolates, respectively. While the rest rice cultivars were infected with the most tested isolates. Kiyosawa (1965)

mentioned that several factors effect on the resistance duration of a newly released variety to rice blast. The first one is the amount of the pathogen around the field, which depends on susceptibility of surrounding varieties and their amount. The second is mutation frequency of avirulence allele to virulence allele. The third is the amount of virulent fungus strains at the time of release of the variety; the last one is multiplication rate of virulent fungus strains between years. Sehly *et al.* (2008) evaluated 14 Egyptian rice cultivars during 1994 up to 2006 growing seasons; these cultivars were distributed at different locations (Multi location test, 18-22 locations yearly) on demonstration farmers، fields at six rice Governorates. They found that Giza 171, Giza 159, Giza 176 and Reiho were susceptible in most of tested locations. While, Giza 181, Giza 177, Giza 178, Sakha 102, Giza 182 and Sakha 103 were resistant at all tested locations during this period. On the other hand, Sakha 101 and Sakha 104 were resistant up to 2003 and became susceptible in 15% of the tested locations starting from 2004 growing season. The level of susceptibility for Sakha 101 was increased from 47.6 to 85.7% in 2005 and 2006 growing seasons. While the susceptibility for Sakha 104 was increased from 28.6 to 66.7% in 2005 and 2006 seasons respectively. These results are in agreement with the cases in different countries such as Thailand (Mekwatanakarn *et al.,* 2000), China (Chen *et al.,* 2001) and Vietnam (Ninh *et al.,* 2006).

These common race groups are represented at all main rice Governorates. IC race group was found at Kafr El-Sheikh, Beheira and Gharbiya Governorates, IB race group at Kafr El-Sheikh, Dakahliya, Beheira and Gharbiya, II race group at Kafr El-Sheikh, Sharkiya and Dakahliya and IF race group was only found at Kafr El-Sheikh. Also, IA and IE race groups were collected from Kafr El-Sheikh Governorate, which is considered a hot spot area because all of the race groups appeared in this location. Rice Research and Training Centre (National Rice Research Program) at Sakha, Kafr El-Sheikh tests annually around five thousand rice genotypes. This may indicate that the existence of new pathotype (s) in some locations raises the inoculum levels in these areas, and enhances the frequency pathotypes. These results are agree with the findings of Sehly *et al*. (2002b) who reported that the distribution of physiological blast races was differed among rice growing Governorates.

Concerning the effective genes on blast control, Pi-Z gene in Fukunishiki followed by Pi-i plus Pi-Ks in Ishikarishiroke were the highest effective ones during the study period. Pi-a gene in Aichi Asahi was the lowest effective one on blast control followed by Pi-K^m gene in Tusyake and Pi- ta^2 gene in Pi No. 4 regardless of the season. Sehly *et al.* (2002b) evaluated 20 isolates of *P. grisea* on JDVs, each with a specific major gene for blast resistance. The isolates represented the six rice Governorates. Pi-Z^t in Toride 1, Pi-Z in Fukunishiki, Pi-b in BL 1, and Pi-K in Kanto 51 were highly effective resistant genes against 20 tested isolates. Pi-ta in Yashiro-Mochi, Pi-K^s in Shin 2, and Pi-a in Aichi Asahi showed 80, 90 and 100% susceptibility to the tested isolates. However, Pi-K^m in Tsuyake and Pi-i in Ishikarishiroke showed moderate levels of resistance, with 40 and 60% susceptibility, respectively. In addition, it was observed that before 1988 growing season, Pi-Z^t in Toride 1 was moderately resistant (46.7% resistance) against the 30 isolates of *P. oryzae*. Pi-Z in Fukunishiki, Pi-K in Kanto 51, Pi-Km in Tsuyake, and

Pi-b in BL 1 showed 86.7, 90, 93.3, and 96.7% resistance against 30 tested isolates, respectively. Highly susceptible genes; Pi-K^s in Shin 2 and Pi-a in Aichi Asahi had 23.3 and 33.3% resistance, respectively. Eizenga *et al.* (2006) screened 91 accessions for presence of the R-genes, out of them five Egyptian entries, *i.e.* Egyptian yasmine, GZ 1368-5-4 and GZ 5594-23-1-2 had Pi-b and Pi-ta, while GZ 5578-2-1-2 has Pi-ta, Pi-K^s, and Pi-Z genes and GZ 5830-48-2-2 had Pi-b and Pi-K^s. Sehly *et al.* (2008) reported that the resistance genes (Pi-a and Pi-K^s) exhibited low levels with 20 and 31%, respectively. Meanwhile, higher levels were obtained in Pi-b, Pi-K and Pi-m genes with 90, 85 and 80%, respectively, during 1994-1996 growing seasons. On the other hand, during 1997 to 2000, higher levels of resistance genes Pi-Z and Pi-K with 95.6 and 93.4% but Pi-a and Pi-ta genes showed lower levels of resistance with 14.3 and 21.1%, respectively. In the last period (2001-2004), Pi-a and Pi-ta^2 genes showed low levels of resistance with 15 and 25%. The rest genes showed high levels and ranged from 60 to 95%. The fluctuation in effective resistance genes from one season to another and between this study and other studies may be due to the prevalence of common physiological races in every season and every location. Bidaux (1976) and Notteghem (1981) described this phenomenon in different countries and observed that virulent strains existed for all the identified genes of vertical resistance and most of the strains possessed virulent genes which were not necessary for their survival. The differential systems consisting of differential varieties and blast isolates are a useful guide to study the resistance and avirulence gene interaction. Identification of genotype and pathotype of a certain variety and blast isolate, respectively, is made possible, by using a standard set of DVs like Japanese differential varieties, which have resistance genes, it would be easier to characterize and differentiate the races of blast pathogens in Egypt. Differential systems would also help in monitoring and characterizing pathogen populations that could facilitate the identification of appropriate breeding strategies and proper gene deployment to rice blast infected areas for effective control measures (Telebanco-Yanoria *et al.,* 2008).

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التخصص الفسيولوجى للفطر بيريكيولاريا جرازيا

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عزله من الفطرالمسبب للفحة الارز بيريكيولاريا جرازيا من
خمس محافظات شهيرة   . وهذه العزلات تم اختبارها على
الأصناف المفرقه العالميه لتحديد سلالاتها الفسيولوجية وكذلك على
لتقيمها و   أصناف يابانيه لتحديد كفاءة جينات المقاومة بها.
اظهرت هذه العزلات تنوعا حيث أتضح انها تتنوع الى   سلاله فسيولوجيه.
العزلات المتحصل عليها خلال موسم   كانت السلالات الفسيولوجيه الشائعة
تتبع المجموعه IH حيث كانت ,   % يليها IG ,II ,   % لكل منهما.
بينما السلالات الفسيولوجيه الأكثر شيوعا خلال موسم   ID, IG
 ,   % على الترتيب وفى موسم
الفسيولوجية الشائعة المتحصل عليها تنتمى الى المجموعة ID حيث بلغت نسبتها
 ,   % يليها السلالات التابعة للمجموعات IC   IB ,   , %
الترتيب. وبخصوص النتائج المتحصل عليها من اختبارات الأصناف التجاريه
المصريه فقد اتضح ان الأصناف جيزه   وهجين
كانت مقاومه لجميع العزلات تحت الدراسه. بينما الاصناف جيزه   وريهو
وجيزه   قد اصيبت بعدد قليل من العزلات   عزله على الترتيب
 .   اسه للأصابه بمرض اللفحة والتى
تمثلت فى جيزة ,   , جيزة ,   و جيزة   قد أظهرت
مستويات مختلفة من الاصابه . اما بخصوص الجينات الفعالة لمقاومة المرض فقد
اظهر الجين Pi-Z أعلى درجة فى المقاومه قدرت بنسبة ,   % يليه Pi-i
Pi-Kˢ ,   %. اما الجينين Pi-ta^2 Pi-a
المرض حيث اظهرت مقاومه بنسبة ,   % على الترتيب.

الجدير بالذكر هنا ان
الموجودة والمتحصل عليها فى بعض التراكيب الوراثية خلال هذه الدراسة تدعو
مزيد من البحث لتطوير تراكيب وراثية متفوقة ومتكيفة يمكن استخدامها فى
برامج تربية الارز فى مصر.