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STEM RUST ANALYSES IN SOME LOCAL EGYPTIAN WHEAT CULTIVARS USING SPECIFIC MOLECULAR MARKERS FOR STEM RUST RESISTANCE AND GENE POSTULATION

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ABSTRACT

Well tested molecular markers for their linkage with important genes for stem rust resistance are recommended to use for developing varieties with durable rust resistance. This study aimed to use ten of the available markers for stem rust resistance genes i.e *Sr39, Sr28* , *Sr26, Sr24, Sr33, Sr43, Sr25, Sr38* , *Sr31* and *Sr55* with ten commercial wheat cultivars i.e Misr1, Misr2, Sids11, Sids12, Gemmiza9, Gemmiza10, Gemmiza11, Saha93, Sakha94 and Giza168 to be analyzed for polymorphism among the selected cultivars based on specific markers for stem rust resistance. In addition, the present study aimed to postulate stem rust resistance genes in the ten Egyptian spring wheat cultivars at seedling stage using 14 pathotypes identification of stem rust (*Puccinia graminis f.sp. tritici*) in 2018/19. Samples were collected during 2017/2018 growing season. All the tested primers produced amplifications products except for the *Sr39* primer sequences. A total of 86 bands were produced by the ten selected specific primers, 6 of which were polymorphic. Maximum level of polymorphism was 60%, which was observed with the primers *Sr25*. The dendrogram grouped the

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genotypes into three clusters, cluster one included Sids12, Sakha 94 and Misr1, while the second cluster contained Sids 13, Gemmiza9, Gemmiza 10, Gemmiza 11 Sakha 93 and Giza 168. Misr 2 was unaccompanied with any other cultivars in the third cluster. Genetic similarity coefficients ranged from 0.10 to 0.88. Comparison between the different Sr's monogenic lines and the local cultivars revealed the probability of genes for resistance in these cultivars. Misr-2 in seedling stage was more resistance to the tested rust isolates than all the other tested cultivars. It probably has stem *Srs ; 39, 28, 26, 25, 38, 55* and other genes for stem rust resistance. *Sr 28* (80 %) proved to be the most common gene that was postulated in most wheat varieties followed by *Sr 25* (70 %). Specific markers coupled with postulation study provided convenient tool to test stem rust resistance in cultivars which may enhance the design of breeding program against stem rust in wheat in future.

Key words: Sr genes, stem rust specific markers, postulation study, Egyptian wheat

INTRODUCTION

Wheat is the main cereal crop in Egypt followed by maize and rice. Egypt has different climatic conditions from the North Delta to Upper Egypt. Stem rust (caused by *Puccinia graminis* Pers. f. sp. *tritici* Eriks. & E. Henn.) is suitable for the climatic conditions of Central and Upper Egypt (Draz *et al.* 2018). The infection is usually expected to begin at the beginning of April and continues until May. The disease emerges from the end of March or beginning of April on the surfaces of the leaf, stem and the spike. To combat rust diseases, one of the basic methods which can be followed is the cultivation of varieties that have a long-lasting or durable resistance under field conditions.

Approximately 50 stem rust resistance (Sr) genes have been recognized and mapped to specific chromosome locations (McIntosh *et al*. 2008). Stem rust genes providing resistance against stem rust are originating from wheat and its wild types as example, *Sr39* was transferred to the hexaploid wheat genotype Marquis from *Aegilops speltoides* (Kerber and Dyke, 1990). The *Sr28* stem rust gene had been earlier reported on chromosome arm 2BL (McIntosh 1978). Sr26 is one of the little identified major resistance genes effective against the *Sr31*-virulent race Ug99 (TTKSK) and its *Sr24*-virulent derivative (TTKST) (Knott 1961, Liu *et al* 2009). *Sr24* compromises resistance to most races of stem rust, including the virulent race Ug99 (TTKSK) now established in East Africa and Ethiopia. Incidence of virulence to this major resistance gene has been reported in South Africa (Mago *et al* 2005 and India (Bhardwaj 1990) . Also, *Sr24* is not effective against the more recent variant of UG99, designated TTKST. *Sr33* was first discovered in annual grass species *Aegilops tauschii* and transferred to chromosome arm 1DS of wheat. This gene offers an average level a resistance to several *stem rust* races including Ug99 associated races (Periyannan et al, 2013).

In wheat breeding programs to pyramid different resistance genes in to wheat elite cultivars, markers assisted selection (MAS) can facilitate transfer major genes conferring resistance to stem rust disease. There is still inadequate data on the presence or absence of Sr resistance genes in local Egyptian wheats based on specific markers. The objectives of this study aimed (1) use ten of the available specific markers for stem rust resistance gene i.e *Sr39*, *Sr28*, *Sr26*, *Sr24*, *Sr33*, *Sr43*, *Sr25*, *Sr38*, *Sr 31* and Sr 55 with ten commercial wheat cultivars i.e. Misr1, Misr2, Sids11. Sids12. Gemmiza9. Gemmiza10, Gemmiza11, Saha93, Sakha94 and Giza168 to analyzed polymorphism among the selected cultivars. (2) postulate the presence of stem rust genes in the selected Egyptian wheat cultivars using monogenic lines of stem rust resistance genes tested in the seedling stage using 14 pathotypes of stem rust (*Puccinia graminis f.sp. tritici*) from collected samples of 2018/2019

MATERIALS AND METHODS

Ten commercial Egyptian wheat cultivars were used to study their resistance against stem rust disease based on specific markers for stem rust resistance (Table 1). Cultivars were provided by Agriculture Research Center (ARC), Egypt. These cultivars were selected because they are highly productive, tolerant to some biotic and abiotic stresses and are recommended by the ministry of Agriculture, Egypt (Abdel-Lateif and Hewedy 2018).

Table 1 Pedigree of the ten studied bread wheat genotypes

Genotypes	Pedigree
Misr-1	OASIS/SKAUZ//4*BCN/3/2*PASTOR
Misr-2	SKAUZ/BAV 92
Sids-12	BUC//7C/ALD/5/MAYA74/ON//1160.147/3/BB/GLL/4/CHAT"S" /6/MAYA/VUL//CMH74A.63014*SX
Sids-13	ALMAZ.19=KAUZ"S"// TSI/ SNB"S"
Gemmiza-9	ALD'S'/HUAC'S'//CMH74.630/5X
Gemmiza-10	Maya 74
Gemmiza-11	BOW"S"/ KVS"S"// 7C/ SERI 82/3/ GIZA 168/ SAKHA 61
Sakha93	Sakha 92/TR 810328 S 8871-1S-2S-1S-0S
Sakha94	OPATA/RAYON//KAUZ. CMBW90Y3280-0TOPM-3Y-010M-010M-010Y-10M-015Y-0Y-0AP-0S.
Giza168	MRL/BUC//Seri. CM93046-8M-0Y-0M-2Y-0B-0GZ

Molecular Markers

DNA extractions

DNA was extracted from young leaf tissue of wheat cultivars using Plant DNAzol (Promega, Madison, WI). The DNA was then suspended in 200 ml of TE buffer, and DNA concentration was quantified by spectrophotometry (TKO100 Fluorometer, Hoefer Scientific Instruments, Holliston, MA).

Allele specific molecular markers analysis

Twelve specific primers for stem rust resistance (Table 2), which were previously identified to be associated with stem resistance genes in wheat were screened for amplification and polymorphism in the 10 Egyptian wheat cultivars. The PCR reaction mixture (25 µl total) consisted of 50 mM KCl and 10 mM Tris–HCl (pH 8.8), 2 mM $MgCl_2$, 125 mM of dNTP, 50 ng of each primer, 1.0 unit of Taq polymerase (Promega, Madison, WI), and 20 ng of genomic DNA. Amplification was carried out in C1000 Touch™ Thermal Cycler (Bio-Rad Laboratories Inc., Hercules, CA), using different programs for each markers Table 2) The amplified PCR products were gel fractionated on 2.5% Agrose gel. Gel images and marker data were processed using Quantity One Software v. 4.0.1 (Bio-Rad Laboratories, Hercules, CA).

Molecular markers data scoring and analysis

Each marker band was scored as present (1) or absent (0), each of which was treated as an independent character. Similarity between the cultivars was analyzed on the basis of the scores. A dendrogram was constructed based on Jaccard's similarity coefficient using the markers data for all wheat cultivars following the unweighted pair group method (UPGMA) (Sokal and Michener 1958)

Postulation of stem rust resistance genes

The ten tested Egyptian wheat varieties i.e. Misr-1, Misr-2, Sids-12, Sids-13, Gemmiza-9, Gemmiza-10, Gemmiza-11, Sakha-93, Sakha94 and Giza 168 and 10 monogenic lines *Sr39, Sr28* , *Sr26, Sr24, Sr33, Sr43, Sr25, Sr38* , *Sr31, Sr55* of stem rust resistance genes were tested in the seedling stage using 14 pathotypes of stem rust (*Puccinia graminis f.sp. tritici*) from collected samples of 2018/2019 growing season, this work in greenhouse and laboratory of wheat Diseases Department, Plant Pathology Research Institute (PPRI), A.R.C., Giza during the period (2018-2019).

Table 2 List of primers name, forward and backward primers sequence, genes and polymorphism results

No	Primers name Primers sequence	Annealing temperature	Gene	No. of amplified	No. of polymorphic	Degree of polymorphis
1	: Sr39F : 5'- AGA GAG AGT AGA AGA GCT GC -3' Sr39R : 5'- AGA GAG AGA GCA TCC ACC -3'	58°C	Sr 39	0	0	0.0
2	wmc332-F 5'-CAT TTA CAA AGC GCA TGA AGC C -3' wmc332-R 5'- GAA AAC TTT GGG AAC AAG AGC A -3'	60°C	Sr28	14	3	21.4
3	BE518379-F 5'- AGC CGC GAA ATC TAC TTT GA -3' BE518379-R 5'- TTA AAC GGA CAG AGC ACA CG -3'	60°C	Sr26	9	1	11.1
4	BARC71-F 5'- GCG CTT GTT CCT CAC CTG CTC ATA -3' BARC71-R 5'- GCG TAT ATT CTC TCG TCT TCT TGT TGGT T -3'	56°C	Sr24	3	1	33.3
5	BARC152-F 5'- CTT CCT AAA ATC GGG CAA CCG CTT GTT G -3' BARC152-R 5'- GCG TAA TGA TGG GAG TGG CTA TAG GGC AGT T -3	50°C	Sr33	10	0	20.0
6	cssu45-F 5'- CGA GTT TCA ATA CTT CGC CC -3? cssu45-R 5'- GAT TAC TAT GCA ATA GGG CCC -3	60°C	Sr45	10	0	0.0
7	Gb-F 5'- CAT CCT TGG GGA CCT C -3' Gb-R 5'- CCA GCT CGC ATA CAT CCA -3	50°C	Sr25	15	3	60.0
8	VENTRIUP 5'- AGG GGC TAC TGA CCA AGG CT -3' LN2 5'- TGC AGC TAC AGC AGT ATG TAC ACA AAA -3'	50°C	Sr38	10	0	0.0
9	CD926040-F GTTGGCTTGGCTACTGCTTT CD926040-R AGCATTCAGCTCTGTGAGCA	60°C to 55°C	Sr13	9	1	11.1
10	CFD23-F 5'- TAG CAG TAG CAG CAG CAG GA -3' CFD23-R 5'- GCA AGG AAG AGT GTT CAG CC -3	60°C	Sr55	6	2	16.7
	Total			86		

Rust data were recorded as infection types (Johnston, 1961). All plant materials were grown in plastic pots, with 6 cm. diam. Each pot contained four genotypes one in each corner in clockwise order. Inoculation and incubation procedures were carried out according to methods adopted by Stakman (1962). Rust reaction was recorded on the first leaf, 12 days after sowing. Rust data were scored as infection type (IT) i.e. R = (O, O; 1 and 2) and S = (3 and 4) which were designated as LIT (low infection type) and HIT (high infection type). Genes were postulated following the method of Browder and Eversmeyer (1980) and Statler (1984).

		Host B (unknown)	
		Resistant	Susceptible
Host A (known)	Resistant	LIT = LIT	LIT = HIT
	Susceptible	HIT = LIT	HIT = HIT

RESULTS AND DISCUSSIONS

Marker validation

To estimate the polymorphism and specificity for the available markers, the present study screened ten markers associated with 10 stem rust resistance genes among 10 local wheat cultivars. Using DNA from leaf tissue, primers were analyzed with polymerase chain reaction (PCR) products using agarose gels. The tested primers amplified clear fragments except for the Sr39 primer sequences (Table 2). Genetic diversity among the local wheat cultivars was based on the resulted polymorphism. A total of 86 bands were produced by the ten selected specific primers, 6 of which were polymorphic (Table 2, Figure 1). Maximum level of polymorphism (60%) was observed for the primers Sr25. Genetic similarity coefficients was ranged from 0.88 (Gemmiza 9 and Gemmiza11) to 0.10 (Gemmiza10 and Misr2) (Table 3). Markers validations was carried by Long-Xi Yu 2009 to assess the polymorphism for the presented markers using fifty eight markers linked with twenty one stem rust resistance genes among sixteen wheat lines. Their results showed that 46 (80%) of the markers produced strong bands and, of those, 35 (75%) showed polymorphism. Sr25 and Sr26 are effective against of Ug99 variants races, TTTSK and TTKST (Singh *et al.* 2006; Jin *et al*, 2007).

Sr39 gene is known to be effective to all currently known pathotypes of *Puccinia graminis* f. sp. *tritici* (Pgt) with Ug99 (TTKSK) and its mutants (Mago *et al* 2009). Recent work with Sr28 gene revealed its presence in wheat landrace PI 177906 which conferring resistance to stem rust at seedling stage (Babiker *et al* 2017), *Sr24* compromises resistance to most pathotypes of stem rust (Bhardwaj 1990, Mago 2005). *Sr33* was initially detected in wild wheat Aegilops *tauschii* this gene offers a moderate resistance to many Puccinia *graminis* sp. *tritici* diseased races (Periyannan *et al* 2013). Stem rust resistance gene *Sr45* originates from *(Aegilops tauschii)*, the D-genome progenitor of *T. aestivum*. This gene was detected in RL5289, which was also the source material for the leaf rust resistance of Lr*21* gene (Kerber 1969). *Sr25* and the associated leaf rust resistance gene *Lr19* were translocated onto the long arm of wheat chromosomes 7D (Friebe *et al* 1994) and 7A (Zhang *et al.* 2005). The use of germplasm having *Sr25*/*Lr19* was primarily narrow because of linkage with another *Thinopy*rum *ponticum* derived gene causing disagreeable yellow flour. Knott (1980) established a mutant line, ‘Agatha-28’, having *Sr25*/*Lr19* with reduced yellow color due to a mutation in the *PSY-E1* gene (Zhang and Dubcovsky 2008). A chromosome locus with (25-38 cM) having three rust resistance genes was translocated between the short arms of *Triticum ventricosum* 2NS and the hexaploid wheat chromosome 2AS (McIntosh1995)._*Sr13* is the single known gene that provide resistance against the TTKS complex of *Puccinia graminis* sp. *tritici*: the TTKSK (Ug99) race and its variants, TTTSK and TTKST (Knott 1962, Klindworth *et al* 2007, Simons *et al* 2011,). Reports derived from molecular mapping experiments showed that blends of moderately effective APR gene Sr2 with 3 to 4 extra APR genes such as Sr55, Sr57, Sr58, Sr56, *and* other undesignated quantitative trait loci confer satisfactory to high levels of resistance to stem rust. Numerous markers have been obtained and used in screening polymorphisms and rust resistance genes in different wheat genotype.

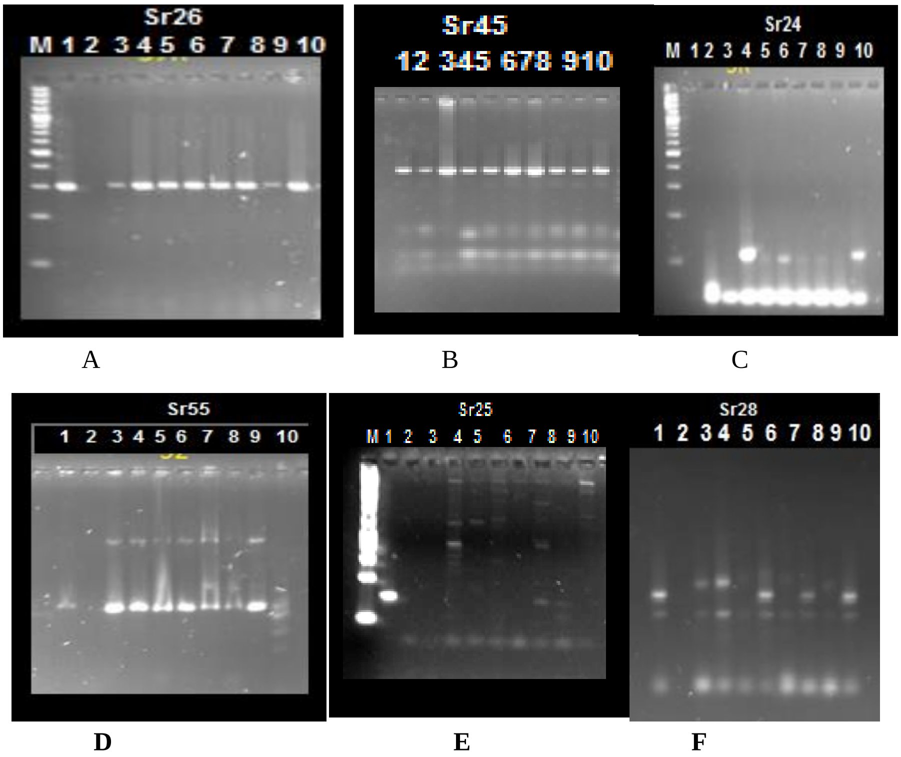

Figure 1. Amplification of specific PCR A for stem rust resistance gene 26, B *Sr* 45; C *Sr* 55; E *Sr*25; F; *Sr* 28 and primers products of ten wheat cultivars. M = 1kbp ladder 1=Misr1, 2= Misr2, 3= Sids12, 4=Sids13, 5 = Gemmiza9, 6 = Gemmiza 10, 7=Gemmiza11, 8= Sakha93, 9=Sakha94, 10=Giza168

Table 3 similarity indices among the studied wheat cultivars

Cultivars	Misr-1	Misr-2	Sids-12	Sids-13	Gemmiza-9	Gemmiza-10	Gemmiza-11	Sahka93	Sahka94	Giza 168
Misr-1	1.00									
Misr-2	0.20	1.00								
Sids-12	0.57	0.17	1.00							
Sids-13	0.36	0.10	0.60	1.00						
Gemmiza-9	0.44	0.13	0.56	0.80	1.00					
Gemmiza-10	0.50	0.10	0.45	0.67	0.80	1.00				
Gemmiza-11	0.50	0.14	0.63	0.70	0.88	0.70	1.00			
Sahka93	0.56	0.11	0.36	0.58	0.55	0.58	0.60	1.00		
Sahka94	0.57	0.17	0.71	0.45	0.56	0.45	0.63	0.50	1.00	
Giza168	0.63	0.13	0.40	0.50	0.60	0.80	0.67	0.70	0.40	1.00

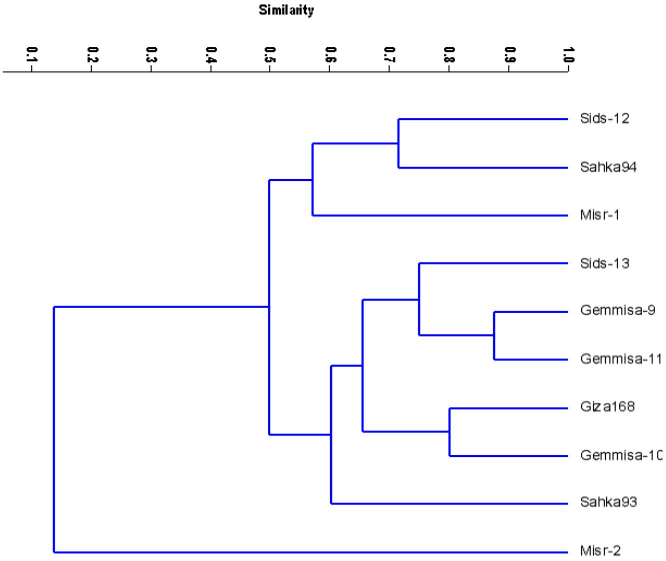

Figure 2 Dendrogram of 10 Egyptian wheat cultivars based on 10 stem rust specific markers.

Gene Postulation Study

Comparison between the different *Sr׳s* monogenic lines and the local cultivars revealed the probability of genes for resistance in these cultivars (Table 4,5 and 6). Misr-1 was resistance to the tested rust isolates than all Sakha cultivars. It probably has *Sr׳s ; 39, 28, 26, 25, 45* and other genes for stem rust resistance. Misr-2: was more resistant to the tested rust isolates than all Misr-1 cultivars. It probably has stem *Sr׳s ; 39, 28, 26, 25, 38, 55* and other genes for stem rust resistance. Sids-12 showed low infection types to 8 isolates, this cultivar probably carries *Sr,s;*39, 28, 26, 25 and other genes for stem rust resistance this result agree with (Abou-Zeid *et al.* 2014 and Shahin *et al.* 2018). Cv. Sids-13: It was shown in this cultivar low infection types to 9 isolates this cultivar probably carries *Sr,s;* 28, 25, 24, 45 and other genes for stem rust resistance. For Gemmiza 9 the obtained results showed that low infection types to 11 isolates This cultivar probably carries *Sr׳s ;* 26, 25, 33, 45, 38 and other genes for stem rust resistance. Gemmiza 10 showed low infection types to 9 isolates, this cultivar probably carries *Sr׳s;* 28, 26, 33 and other genes for stem rust resistance. Moreover, Gemmiza-11 the obtained results showed that low infection types to 6 isolates This cultivar probably carries *Sr׳s;* 28, 24, 38 and other genes for stem rust resistance. Sakha-93 indicated low infection types to 10 isolates, this cultivar probably carries *Sr;* 28, 25, 55 and other genes for stem rust resistance. Sakha-94 exhibited low infection types to 10 isolates this cultivar probably carries *Sr׳s;* 28, 24, 55 and other genes for stem rust resistance. Giza 168 revealed low infection types to 10 isolates, this cultivar probably carries *Sr;* 25, 45, 38, 55 and other genes for stem rust resistance.

Result showed that, the postulated genes identified within ten commercial wheat cultivars, the number of probable gene(s) for stem rust resistance were postulated by comparing the reaction of these varieties against the reaction of 10 monogenic lines (*Sr's*) inoculated with 14 cultures of *Puccinia graminis f.sp. tritici.* The present results indicated to the possibility of the postulation of 10 stem rust resistance genes in the tested wheat varieties.

In general, Sr 28 (80 %) proved to be the most common gene that was postulated in most wheat varieties followed by Sr 25 (70 %). On the other hand, *Sr 13* was not detected in any of the tested varieties. Similar results were previously reported by Singh and Rajaram, (1991); Singh and Gupta (1991), Singh (1993), Kadkhodaei . (2012), Abou-Zeid (2014), Abou-Zeid *et al.* (2014) and Abou-Zeid *et al.* (2018). Concerning the situation of the tested wheat varieties in relation to the postulated genes, the obtained results indicated the probability of the presence of 6 Sr,s genes in var. Misr-2 followed by Misr-1, Sids-12 and Gemmiza-9 (5), Sids 13 (4), Gemmiza-11 Sakha-93 and Sakha94 (3, each),. These results were in accordance with those previously reported by Singh and McIntosh (1986); McVey (1989); Singh and Rajaram (199l); Singh and Gupta (1991), Singh (1993) Abou-Zeid *et al* (2014), Abou-Zeid . (2014) and Abou-Zeid *et al* (2018).

Table (4): Seedling reactions of 10 resistance genes (*Sr*) against 14 isolates of *Puccinia graminis* under greenhouse condition in 2018/2019.

NO.	**P.g. Isolates / Infection types**														
	Resistance gene (*Sr*)	BTTCS	CBSCT	MKPMT	NKTDT	PHCFG	MBTLN	MTCNT	MTKMT	THLNP	NHKNT	PHTBP	PKSBB	BNTTT	RSSNT
1	**39**	**L**	**L**	**L**		**L**	**L**	**L**		**L**				**L**	**L**
2	**28**	**L**	**L**	**L**	**L**	**L**		**L**	**L**	**L**	**L**		**L**		**L**
3	**26**				**L**	**L**	**L**					**L**			
4	**25**		**L**				**L**				**L**		**L**		
5	**24**		**L**	**L**	**L**			**L**	**L**		**L**	**L**		**L**	**L**
6	**33**				**L**		**L**			**L**	**L**		**L**	**L**	**L**
7	**45**	**L**		**L**	**L**		**L**	**L**	**L**	**L**					**L**

8	**38**			**L**	**L**				**L**		**L**		**L**	**L**	
9	**13**					**L**		**L**						**L**	
10	**55**										**L**		**L**		**L**

L = low infection type **Blank cells = High infection type**

Table (5): Seedling reactions of 10 Egyptian wheat cultivars against 14 isolates of *Puccinia graminis* under greenhouse conditions in 2018/2019.

NO.	Cultivars	***P.g.* Isolates / Infection types**													
		BTTCS	**CBSCT**	**MKPMT**	**NKTDT**	**PHCFG**	**MBTLN**	**MTCNT**	**MTKMT**	**THLNP**	**NHKNT**	**PHTBP**	**PKSBB**	**BNTTT**	**RSSNT**
1	Misr-1	**L**	**L**	**L**	**L**	**L**	**L**		**L**	**L**	**L**				**L**
2	Misr-2			**L**		**L**	**L**	**L**		**L**		**L**	**L**	**L**	**L**
3	Sids-12	**L**	**L**		**L**		**L**	**L**	**L**					**L**	**L**
4	Sids-13	**L**	**L**	**L**	**L**	**L**		**L**	**L**	**L**					**L**
5	Gemmiza - 9	**L**		**L**	**L**		**L**	**L**	**L**	**L**	**L**	**L**		**L**	**L**
6	Gemmiza - 10	**L**			**L**	**L**	**L**		**L**		**L**	**L**		**L**	**L**
7	Gemmiza - 11	**L**					**L**			**L**		**L**		**L**	**L**
8	Sakha93	**L**		**L**		**L**	**L**	**L**		**L**	**L**	**L**	**L**		**L**
9	Sakha94	**L**		**L**		**L**	**L**	**L**	**L**		**L**	**L**	**L**		**L**
10	Giza168	**L**		**L**	**L**	**L**		**L**	**L**		**L**		**L**	**L**	**L**

L = low infection type. Blank cells = High infection type

CONCLUSIONS

Results of this study revealed that Misr 2 was highly diverse from the all studied cultivars based on specific marker analyses. In addition, based on postulation study cultivar Misr 2 was more resistance to the tested rust isolates compared to all cultivars. It probably has stem *Sr's ;* 39, 28, 26, 25, 38, 55 and other genes for stem rust resistance. Evidence obtained in this study will be significant for releasing possibly durable blends of stem rust resistance genes in cultivars.

Table (6): Probable resistance genes for stem rust (*Sr's*) in some Egyptian wheat cultivars.

NO.	**Cultivars**	**Probable *Sr* genes**
1	Misr-1	39, 28, 26, 25, 45, *
2	Misr-2	39, 28, 38, 45, 55, *
3	Sids-12	39, 28, 26, 25, *
4	Sids-13	28, 25, 24, 45, *
5	Gemmiza -9	26, 25, 33, 45, 38, *
6	Gemmiza -10	28, 26, 33, *
7	Gemmiza -11	28, 24, 38, *
8	Sakha-93	28, 25, 55, *
9	Sakha-94	28, 24, 55, *
10	Giza-168	25, 45, 38, 55, *

*=Additional genes may be found.

REFERENCES

Abdel-Lateif, K.S. and O.A. Hewedy. 2018. Genetic diversity among Egyptian wheat cultivars using SCoT and ISSR markers. SABRAO Journal of Breeding and Genetics 50 (1) 36-45

Abou-Zeid, A.M., Abd Elhameed A. S. and M.M.H. Abd El-Wahab 2018. Evaluation of new wheat genotypes with genetic for stem rust resistance diversity and some yield components under Egyptian field conditions. *Egypt. J. Plant Breed. 22(4):849–871.*

Abou-Zeid, M.A. 2014. Identification of yellow rust resistance gene *Yr 18* in Egyptian wheat germplasm by molecular markers. "2nd International Wheat Stripe Rust Symposium" Izmir, Turkey, 28 April-1 May, 2014.

Abou-Zeid, M.A.; Olfat M. Moussa; Mona M. Ragab and S. Sherif 2014. Virulance of *Puccina graminis f. sp. tritici* and postulated resistance genes for stem rust in ten wheat varieties in Egypt. International Journal of Plant & Soil Science, 3(6): 671-684

Babiker, EM, T.C, Gordon, S, Chao, MN, Rouse, R, Wanyera, M, Acevedo, G, Brown-Guedira and Bonman JM.. 2017. Molecular Mapping of Stem Rust Resistance Loci Effective Against the Ug99 Race Group of the Stem Rust Pathogen and Validation of a Single Nucleotide Polymorphism Marker Linked to Stem Rust Resistance Gene *Sr28*. Phytopathology, 107(2):208-215.

Bhardwaj, SC, SK, Nyar, M, Prashar, J, Kumar, MK, Menon, SB. Singh 1990. A pathotype of *Puccinia graminis* f. sp. *Tritici* on *Sr24* in India. In: Cereal Rusts and Powdery Mildews Bulletin, 18:35-38.

Browder, L.E. and Eversmeyer, M.G. 1980. Sorting of *Puccinia recondita*: *Triticum* infection-type data sets toward the gene-for-gene model. Phytopath., 70: 666 - 670.

Draz I., Samar, M. Esmail, M. Abou-Zeid and Y. Hafez (2018). Changeability in Stripe Rust Infection and Grain Yield of Wheat Associated to Climatic Conditions. Env. Biodiv. Soil Security Vol. 2, 143 – 153

Friebe B, J Jiang, D.R. Knott and BS. Gill 1994. Compensation indexes of radiation-induced wheat Agropyron-elongatum translocations conferring resistance to leaf rust and stem rust. . In: Crop Science 34: 400-404.

Jin, Y., R.P. R.W. Singh, R. Ward, M. Wanyera, P. Kinyua, Njau, and Z.A. Pretorius. 2007. Characterization of seedling infection and adult plant infection responses of monogenic Sr gene lines to race TTKS of *Puccinia graminis* f. sp. *tritici*. Types. Plant Dis. 91:1096–1099. doi:10.1094/PDIS-91-9-1096

Johnston, C.O. 1961. The “Y” type infection in the leaf rust of wheat.*Robigo* 15, 1-2.

Kadkhodaei, M.; Ali, D.; M.T.Assad; H.Bahram, and R. Mostowfizadeh-Ghalamfarsa, 2012 Identification of the Leaf Rust Resistance genes *Lr9*, *Lr26*, *Lr28*, *Lr34*, and *Lr35* in a Collection of Iranian Wheat Genotypes Using STS and SCAR Markers. J. Crop Sci. Biotech. 15: 267-274.

Kerber ER, and PL. Dick 1969. Inheritance in hexaploid wheat of leaf rust resistance and other characters derived from *Aegilops squarrosa*. In: Canadian Journal of Genetics and Cytology, 11:639-647.

Kerber ER, and PL Dyke.1990. Transfer to hexaploid wheat of linked genes for adult-plant leaf rust and seedling stem rust resistance from amphiploid of *Aegiolops speltoides* x *Triticum monoccum*. Genome 33:530-537.

Klindworth DL, JD. Miller, Y. Jin and SS. Xu 2007. Chromosomal locations of genes for stem rust resistance in monogenic lines derived from tetraploid wheat accession ST464. In: Crop Science, 47:1441-1450.

Knott D.R. 1980. Mutation of a gene for yellow pigment linked to *Lr19* in wheat. Can J. of Gene. and Cytology, 22:651–654

Knott DR. 1962. The inheritance of rust resistance: IX. The inheritance of resistance to races 15B and 56 of stem rust in the wheat variety Khapstein. Can. J. of Plant Science 42:415-419.

Mago R, HS Bariana, IS Dundas, W Spielmeyer, GJ Lawrence, AJ Pryor and JG Ellis. 2005. Development of PCR markers for the selection of wheat stem rust resistance genes *Sr24* and *Sr26* in diverse wheat germplasm. . In: Theoretical and Applied Genetics, , 111:496-504.

Mago R, Zhang P, Bariana HS, Verlin DC, Bansal UK, Ellis JG, Dundas IS. 2009. Development of wheat lines carrying stem rust resistance gene *Sr39* with reduced *Aegilops speltoides* chromatin and simple PCR markers for marker-assisted selection. In: Theoretical and Applied Genetics. 124:65-70.

McIntosh RA. 1978. Cytogenetic studies in wheat X. Monosomic analysis and linkage studies involving genes for resistance to *Puccinia graminis* f. sp. *tritici* in cultivar Kota. McIntosh RA. In: Heredity, 41:71-82.

McIntosh RA, Wellings CR, Park RF (1995) Wheat rust: an atlas of resistance genes. CSIRO, Australia

McIntosh, RA, Y Yamazaki, J Dubcovsky and WJ Rogers 2008. Catalogue of gene symbols for wheat. Gene symbols. In: McIntosh RA (ed) http://wheat.pw.usda.gov/GG2/Triticum/wgc/2008/GeneSymbol.pdf.

McVey, D.V. 1989. Verification of infection-type data for identification of genes for resistance to leaf rust in some hard red spring wheats. Crop Science 29, 304-307.

Periyannan S, J Moore, M Ayliffe, U Bansal, X Wang, L Huang, Deal K, M Luo, X Kong, H Bariana, R Mago, R McIntosh, Dodds P, Dvorak J, Lagudah E. 2013. The gene *Sr33*, an ortholog of barley *Mla* genes, encodes resistance to wheat stem rust race Ug99. Science, , 341:786-788.

Shahin, A. A.; M. A. Hasan and M. A. Abou-Zeid (2018). Adult plant resistance to stem rust and molecular marker analysis of some Egyptian and exotic bread wheat genotypes. *J. Plant Prot. and Path., Mansoura Univ., Vol.9 (12): 901 – 907.*

Simons K, Abate Z, Chao S, Zhang W, Rouse M, Jin Y, Elias E, Dubcovsky J. 2011. Genetic mapping of stem rust resistance gene *Sr13* in tetraploid wheat (*Triticum turgidum* ssp. *durum* L.). TAG Theoretical and Applied Genetics, , 122:649-658.

Singh R. P., McIntosh R. A., 1986. Cytogenetical studies in wheat XIV. Sr8b for resistance to *Puccinia graminis tritici*. Can. J. Genet. Cytol. 28: 189–197.

Singh, R.P., D.P. Hodson, Y. Jin, J.H. Espino, M.G. Kinyua, R. Wanyera, P. Njau, and R.W. Ward. 2006. Current status, likely migration and strategies to mitigate the threat to wheat production from race Ug99 (TTKS) of stem rust pathogen. CAB Rev. Perspect Agric. Vet. Sci. Nutr. Nat. Resour. 1(054):13.

Singh, R.P. 1993. Genetic association of gene *bdv1* for tolerance to barley yellow dwarf virus with genes *Lr34* and *Yr18* for adult plant resistance to rusts in bread wheat. Plant Dis., 77, 1103-1106.

Singh, R.P. and Gupta, A.K. 1991. Genes for leaf rust resistance in Indian and Pakistani wheats tested with Mexican pathotypes of *Puccinia recondita* f. sp. *tritici*. Euphytica 57:27–36

Singh, R.P. and S. Rajaram 1991. Resistance to *Puccinia recondita* f. sp. *tritici* in 50 Mexican bread wheat cultivars. Crop Sci., 31:1472-1479.

Sokal RR, Michener CD. 1958. A statistical method for evaluating systematic relationships. Univ. Kansas Sci. Bull.38:1409–1438.

Stakman EC, Stewart DM, 1962. Loegering WQ. Identification of physiologic races of *Puccinia graminis* var. *tritici*. U.S, Agric. Res. Serv. ARS E.;617:1-53.

Statler, G.D. 1984. Probable genes for leaf rust resistance in several hardred spring wheats. Crop Sci., 14 : 883 - 886.

Zhang W, Dubcovsky J 2008. Association between allelic variation at the Phytoene synthase 1 gene and yellow pigment content in the wheat grain. Theoretical and Applied Genetics, 116:635-645.

Zhang W, AJ Lukaszewski, J. Kolmer, MA. Soria, S. Goyal and J. Dubcovsky 2005. Molecular characterization of durum and common wheat recombinant lines carrying leaf rust resistance (*Lr19*) and yellow pigment (*Y*) genes from *Lophopyrum ponticum* . In: Theoretical and Applied Genetics, 111:573-582.

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تحليل صدأ الساق في بعض أصناف القمح المصري المحلي باستخدام المعلمات الجزيئية المتخصصة لمقاومة صدأ الساق والافتراض الجينى

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يوصى باستخدام المعلمات وذلك لتحديد وجود جينات مهمة لمقاومة صدأ الساق من أجل تطوير أنواع القمح ذات المقاومة لصدأ الساق. وهدفت هذه الدراسة إلى استخدام عشرة من المعلامات المتاحة وذلك لتحديد جينات المقاومة لصدأ الساق هذة الجينات هي *Sr39* ، *Sr28* ، *Sr26* ، *Sr24* ، *Sr33* ، *Sr43* ، *Sr25* ، *Sr38* ، *Sr31* ، *Sr55* مع عشرة أصناف من القمح التجاري ، هي مصر1 , مصر 2, سدس 11, سدس 12, جميزة 9, جميزة 10, جميزة 11, سخا 93, سخا 94 و جيزة 168 وتحليل البلومورفيزم الناتج عن هذة الاصناف بالارتكاز على معلمات جزيئية محددة لمقاومة صدأ الساق. بالإضافة إلى ذلك ، أيضا هدفت الدراسة الحالية إلى افتراض جينات المقاومة لصدأ الساق في هذة الاصناف من القمح محل الدراسة في مرحلة البادرة باستخدام 14 سلالة لمسبب صدأ الساق (*Puccinia graminis f.sp. tritici*) التي تم تعريفها في 19/2018 هذه العينات تم جمعها في الموسم السابق 2018/2017. أنتجت جميع البادئات التي تم اختبارها منتجات تضاعف باستثناء تسلسل برايمر او بادئ الSr39 . تم إنتاج اجمالى 86 باند من خلال عشرة أنواع محددة من البادئات ، 6 منها كانت متعددة الأشكال. كان الحد الأقصى لمستوى تعدد الأشكال 60 ٪ ، والتي لوحظت مع بادئ .Sr25 وقد قسم الديندوغراما التراكيب الوراثية إلى ثلاث مجموعات ، المجموعة الأولى شملت سدس 12 ، سخا 94 و مصر1 ، بينما تضم المجموعة الثانية سدس 13 و جميزة 9 و جميزة 10 و جميزة 11 سخا 93 و جيزا 168 .وكان مصر 2 غير مصحوب بأي أصناف أخرى في المجموعة الثالثة. تراوحت معاملات التشابه الوراثية من 0.88 إلى 0.10. وقد بينت المقارنة بين الاصناف محل الدراسة وال 10 سلالات التى تحمل جين فردي لمقاومة صدأ بتوقع جينات موجودة بالأصناف المحلية نتج عنها هذة المقاومة . الصنف "مصر -2" احتمال ان يوجد به اكبر عدد من الجينات المقاومة, لمقاومتة لاكبر عدد من العزلات من الفطر بالمقارنة بالأصناف الأخرى التي تم اختبارها. من المحتمل أن يكون جينات *Srs ; 39, 28, 26, 25, 38, 55* وجينات أخرى لمقاومة صدأ الساق. ثبت أن Sr 28 (80 ٪) هو الجين الأكثر شيوعا الذي تم افتراضه في معظم أصناف القمح يليه Sr 25 (70 ٪). توفر المعلمات المحددة مقترنة بدراسة افتراضية وجود جين المقاومة لصدأ الساق في الأصناف التي تستخدم في السياسة الصنفية يعزز من تحسين برنامج تربية القمح لمقاومة صدأ الساق في القمح في المستقبل.

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