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VARIATION IN THE RESISTANCE SPECTRA OF ETHIOPIAN WHEAT CULTIVARS TO STEM RUST (*PUCCINIA GRAMINIS* F. SP. *TRITICI*) AND ITS EPIDEMIOLOGICAL IMPLICATION

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ABSTRACT: Multipathotype test was conducted to study the resistance spectra of Ethiopian hexaploid and tetraploid wheats to stem rust (*Puccinia graminis* f.sp. *tritici*) infection. Seedlings of 14 durum (*Triticum durum*), 10 emmer (*T. dicoccon*), 15 bread (*T. aestivum*) wheat cultivars and accessions, and 33 stem rust differential lines were inoculated with 18 different stem rust races in the greenhouse experiment. The observed infection types (ITs) were categorized as low infection type (LIT) and high infection type (HIT). The number of LITs (resistance spectrum) exhibited by each variety and accession was described in terms of low infection type frequency (LIT*f*). The results obtained indicated that three bread wheat varieties, KBG-01, Bobitcho, Megal and one durum wheat variety, Ude possess widest resistance spectrum (LIT*f* = 18, resistant to all the 18 races) while three bread wheat varieties, Ketar, Galama, Shinna and one durum variety, Gerado exhibited the narrowest spectrum (LIT*f* = 7 and 8). ACC-213155–2 of emmer wheat type showed resistance to 16 races while ACC-212883–3 was resistant to 11 races. The highest range of variation in the resistance spectrum was observed among durum wheat genotypes while variation recorded among emmer varieties was the least. LIT*f* values of each variety gave clues to the epidemiological relationship between LIT*f* values and pathogen co-evolution. In cases where gene-for-gene interaction works, varieties with highest LIT*f* values exert higher selection pressure on the pathogen population than those varieties with lower LIT*f* values and consequent upon this hypothesis the importance of assessing LIT*f* values of each variety in designing and implementing vertical resistance management strategy is discussed..

Key words/phrases: Low infection type frequency, *Puccinia graminis* f.sp. *tritici*, resistance spectra, stem rust, wheat

INTRODUCTION

Wheat in Ethiopia is represented by hexaploid (2n = 6X = 42, AABBDD) and tetraploid (2n = 4X = 28, AABB) species. Bread wheat (*Triticum aestivum* L.) is a widely grown hexaploid wheat while durum wheat (*T. durum* Desf.) and emmer wheat (*T. dicoccon* Schrank) are the two cultivated tetraploid wheats. Stem or black rust of wheat caused by the fungus *Puccinia graminis* f.sp. *tritici* Eriks & Henn (*Pgt*) is an important disease on wheat worldwide (Roelfs *et al.*, 1992). *Puccinia graminis* f. sp. *tritici* is an obligate biotroph, heteroceous in its life cycle and heterothallic in mating type (Alexopoulos *et al.*, 1996). The stem rust disease is one of the major yield limiting factors and the current threat to Ethiopian wheat production due to the detection of a new physiological race, TTKS (Ug99) against *Sr31* resistant gene in East Africa (Pretorius *et al.*, 2000). Ethiopia is one of the hot spot areas for the development of wheat stem rust complex (Leppik, 1970). Surveys conducted during the 2003/2004 cropping seasons in Ethiopia have indicated that most wheat varieties have lost their resistance to stem rust shortly after release and that the main reason for the susceptibility of wheat varieties against stem rust is the vertical resistance breeding strategy adopted and a narrow genetic base on which the breeding for

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resistance has been founded (Naod Beteselassie, 2004).

Despite the limitations and drawbacks of developing varieties with vertical resistance such as formation of new races that overcome the resistance gene, its ineffectiveness against multi-races, and its short life span, it still remains the most widely adopted strategy of most wheat breeding programs because it is easy to manipulate, and exhibits complete resistance until new virulent race appears. The epidemiological reasons why vertical resistance favours the appearance of new races were best explained by the concept of boom and bust cycle (Knott, 1989). Epidemiological and genetical factors such as types and levels of host resistance, extent of pathogen variability and virulence spectra influence the rate of change in the virulence frequency dynamics of cereal rust fungi (Vanderplank, 1982; Burdon, 1987; Groth, 1984). Such epidemiological analyses were the conceptual frameworks to suggest and develop vertical resistance management options such as regional gene deployment and pyramiding, varietal mixtures and multilines (Knott, 1989).

Multipathotype testing that involves the use of large number of races and differential lines is useful to study and understand different aspects of host-pathogen interaction (Flor, 1971; McVey and Roelfs, 1975; Sawhenay, 1995). McVey (1991) and Marais *et al.* (1994) have also used multipathotype testing method to postulate stem rust resistance *(Sr)* genes and showed the resistance spectra of some wheat genotypes.

In Ethiopia, various efforts have been made to study different aspects of wheat-*Pgt* interactions (Temam Hussien, 1984; Masresha Aklilu, 1996; Ephrem Bechere *et al.*, 2000). However, the variation in the resistance spectra of wheat varieties against stem rust and its epidemiological implication such as selection pressure on the pathogen was overlooked. Several promising wheat varieties were released and their production life-span has increasingly become short. As a result of this, newly released varieties soon become out of production due to stem rust attack even before being widely adopted. Combination of variation assessment of *Pgt* virulence and or wheat varieties resistance spectra provides useful information to devise a better breeding and disease management strategy to use released varieties in wheat production. However, to this date, no recorded information is available about resistance spectra of Ethiopian wheat cultivars to stem rust.

The objectives of this study, therefore, were to assess the extent of variation in the resistance spectra of Ethiopian hexaploid and tetraploid wheat varieties and accessions against stem rust, and the effect of epidemiological implication of the variation as affecting pathogen evolution. The importance of these in the management strategies of stem rust disease on wheat in Ethiopia is discussed.

MATERIALS AND METHODS

Testing materials

Three categories of wheat including 15 bread, 14 durum and 10 emmer wheat varieties and accessions were used. A susceptible bread wheat variety, *Morocco,* was used as a control (Table 1). Eighteen stem rust races that were characterized and maintained according to the method of Naod Beteselassie (2004) at the Kulumsa Agricultural Research Center (KARC), Ethiopia were used (Table 2). The study was conducted under greenhouse condition at KARC in 2004.

Multipathotype test

Six seeds of each variety and accession were planted in 9-cm diameter plastic pots to raise seedlings. The plastic pots were filled with a mixture of soil, compost and sand in ratio of 1:1:1 (v/v). The spore suspensions were prepared by mixing urediospores of each race in FC-40 mineral oil in a separate beaker. The spore concentrations were adjusted to 4×10^6 spores/ml. Seven-day-old seedlings of each variety and accessions were then inoculated by spraying the spore suspensions with an atomizer. Inoculated seedlings were incubated in a growth chamber at 20±2°C, 98% RH for 24 dark hours. The seedlings were then transferred to the greenhouse benches until disease scoring was done.

Disease assessments

Disease assessments were conducted twice at 12 and 14 days after inoculation according to the method of Stakman *et al.* (1962) that is infections types (ITs) of each seedling were scored by using a 0 to 4 seedling scoring scale (0 = Immune, = Nearly immune, 1 = Very resistant, 2 = Moderately resistant, X = heterogeneous, 3 = Moderately susceptible, 4 = Susceptible).

Table 1. Ethiopian wheat varieties and accessions evaluated for their resistance to 18 stem rust (*P. graminis* f.sp. *tritici*) races.

No.	Wheat varieties/lines	PEDIGREE
	Bread wheat	
1.	Enkoy	(HEBERARD SEL/WIS2451)x(FR-FN/Y)A
2.	Mitike	(FSYR20.6/87 BOW28) x (RBC (ET1297))
3.	Kubsa	ATTILA
4.	Galama	4777(2)//FKN/GB/3/PVN"S"
5.	Tuse	COOK/VEE"S"//DOVE"S"/SERI
6.	Megal	F3.71/TRM//BUC"S"/3/LIRA"S"
7.	Ketar	COOK/VEE"S"//DOVE"S"/SERI/3/BIY" S"/COC
8.	Shinna	GOV9AZ//MUS"S"/3/R37/GHL121// KAL/bb/4/ANI"S"
9.	Hawi	CHIL/PRL
10.	Simba	PRINIA
11.	Dodota	BJY/COC//PRL/BOW
12.	Sirbo	MILAN
13.	KBG-01	(300/SM+501M)/HAR 1709
14.	Bobitcho	BURRION
15.	HAR 2504	PASTOR
	Durum wheat	
1.	Arendeto	NA
2.	Gerado	JORRO
3.	Cocorit -71	NA
4.	LD-357	LD357/CI8155 ND58-40
5.	Bichenna	NA
6.	Quamy	AOS//PGO/Cii/3/jo69/CRA//GS/SB A81/3/FGO/CRA/5/FGO/DON/6/HU L,CD75533-A
7.	Asasa	NA
8.	Ginchi	NA
9.	Ude	NA
10.	Dz-966-SY	NA
11.	Dz-1935	NA
12.	Dz-95457	NA
13.	Dz-293-2DZR	NA
14	Dz-1052	NA
	Emmer wheat	
1.	Sinana-01	Acc. 216074-1
2.	ACC-212883-2	Accessions
3.	BC-050	Accessions
4.	BC-042	Accessions
5.	ACC-214540	Accessions
6.	Local Emmer	Accessions
7.	Local Emmer	Accessions
8.	ACC-215522	Accessions
9.	ACC-22406	Accessions
10.	ACC-213155-2	Accessions
	Morroco variety	Susceptible control

NA = pedigree information not available

Data summary and analysis

Infection types from 0–2 were summarized as resistance and 3–4 as susceptible. For ease of analysis, low infection types (LITs) were recorded as 1 and high infection types (HITs) as 0 (McVey, 1991). The number of LITs exhibited by each genotype for all races was counted and low infection type frequency (LITf) value was assigned for each genotype. Data were interpreted based on the facts and assumptions of gene-for-gene concept (Flor, 1946; 1971).

Table 2. Wheat stem rust (caused by *Puccinia graminis* f.sp. *tritici*) races and their virulence spectra.

Pgt-code*	None effective *Sr* genes
TKM/J**	5, 6, 7b, 8a, 9d, 9e, 9g, 10, 12, 13, 15, 17, 19, 21, 22, 24, 25, 26+9g, 27, 29, 32, 36, 37
SKM/J	5, 6, 8^a, 9d, 9e, 9g, 12, 13, 15, 17, 18, 19, 21, 22, 25, 26+9g, 27, 29, 36, 37
TTM/J	5, 6, 7b, 8a, 8b, 9d, 9e, 9g, 10, 11, 12, 13, 14, 15, 17, 19, 21, 22, 24, 25, 26+9g, 32, 36, 37
STM/J	5, 6, 8^a, 9e, 9g, 10, 11, 12, 13, 15, 17, 18, 19, 21, 22, 24, 26+9g, 25, 27, 29, 32, 36, 37
TTL/K	5, 6, 7b, 8a, 8b, 9d, 9e, 9g, 11, 12, 13, 15, 18, 19, 21, 22, 26+9g, 27, 28, 29, 32, 36,37
TKR/J	5, 6, 7b, 8a, 9b, 9d, 9e, 9g, 10, 12, 13, 14,17, 18, 19, 21, 22, 24, 25, 26+9g, 27, 29, 32, 36, 37
TKM/J	5, 6, 7b, 8a, 9d, 9e, 9g, 12, 13, 15, 17, 21, 22, 24, 25, 26+9g, 27, 29, 36, 37
TTM/H	5, 6, 7b, 8a, 8b, 9d, 9e, 9g, 11, 17, 21, 22, 25, 28, 36, 37
SKM/J	5, 6, 8^a, 9d, 9e, 9g, 10, 12, 13, 14, 15, 17, 18, 19, 21, 22, 24, 25, 27, 26+9g, 29, 32, 36, 37
JKM/G	6, 8a, 9d, 9e, 9g, 12, 13, 15, 17, 21, 25, 32, 36, 37
STB/J	5, 6, 8^a, 9d, 9e, 9g, 10, 11, 12, 13, 14, 15, 19, 21, 22, 24, 25, 26+9g, 29, 32, 37
NFB/B	5, 8a, 8b, 9e, 9g, 19
KDB/D	7b, 8^a, 9e, Tt-3+10, 15, 19, 21, 22, 25, 37, 26+9g
FSF/K	6, 7b, 8a, 8b, 9d, 9e, Tt-3+10, 10, 11, 12, 13, 15, 17, 18, 19, 25, 28, 29,30, 31, 37
QHB/C	5, 6, 9g, 13, 19, 21, 24, 25, 28, 29, 8b
STH/K	5, 6, 8^a, 9b, 9d, 9e, 9g, 10, 11, 13, 14, 15, 17, 18, 21, 28, 31, 32, 37
SJB/J	5, 6, 8^a, 9d, 9e, 10, 12, 13, 15, 21, 22, 24, 25, 26+9g, 32, 37
SRL/J	5, 6, 9d, 9e, 10, 11, 12, 13, 14, 18, 21, 26+9g, 27, 29, 32, 36, 37

* Pgt-code was used to name the races (Roelfs and Martens, 1988)

RESULTS

The resistance spectrum exhibited by each bread, durum and emmer wheat varieties and accessions is presented in Fig. 1. The result showed that KBG-01, Bobitcho and Magal bread wheat varieties were resistant to all the 18 races of *P. graminis* f. sp. *tritici*. These varieties have the widest resistance spectra (LIT*f* = 18) among the tested bread wheat varieties. The recorded wide resistance spectra on these bread wheat varieties may be a favorable host factor for the recently obsetved susceptibility of Bobitcho and Magal varieties under field conditions. The other bread wheat varieties such as Shinna, Galama and Ketar that showed resistance to 8 races out of 18 were found to have narrow resistance spectra (LIT*f* = 8). The two widely grown bread wheat cultivars, Galama and Kubsa also exhibited narrow resistance spectra. Galama is resistant to 8 races and Kubsa to 10 out of 18 races under assessment.

Among the tested 15 durum wheat varieties, Ude was the only variety with widest resistance spectra (LIT*f* = 18) while the old semi dwarf variety, Gerado exhibited narrowest resistance spectra (LIT*f* = 7). The three advanced lines of durum wheat: Dz-966-SY, Dz-95457, Dz-293-2DZR showed a resistance spectrum of 14, 15, and 17, respectively. The highest range of variation in the resistance spectrum was also observed among durum wheat genotypes.

In the tested ten emmer wheat genotypes, the widest resistance spectrum (LIT*f* = 16) was observed in ACC-213155-2, Sinana-01 showed resistance to 14 races (LIT*f* = 14) while ACC-212883-2 showed the narrowest resistance spectrum (LIT*f* = 11). The range of variation in the resistance spectrum was found minimum in emmer lines as compared to the range of variation observed in bread and durum wheat varieties and accessions.

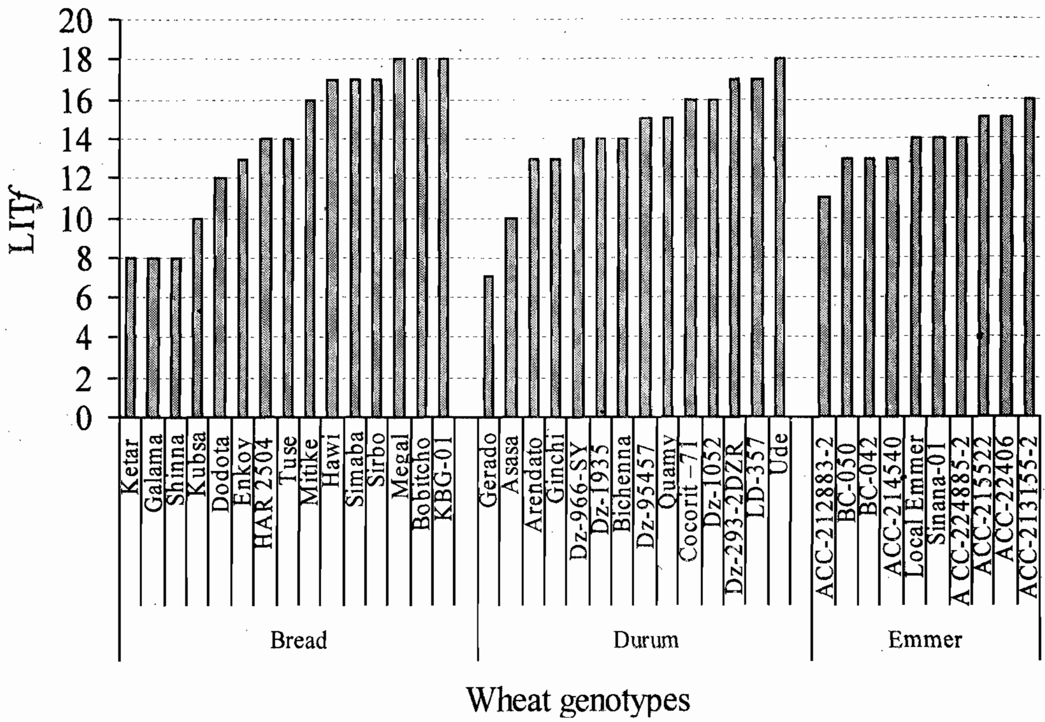

Fig. 1. The resistance spectra exhibited by Ethiopian bread, durum, and emmer wheat varieties and accessions tested against eighteen stem rust (caused by *Puccinia graminis* f. sp. *tritici*) races.

** Letter after slash (/) represents 4th sub-set (supplemental differential lines with *Sr*9a, 9d, 10 *and Sr*28 genes)

DISCUSSION

The success of multipathotype testing in describing the extent of variation in disease resistance depends largely on the number and genotype of pathogen isolates used in the study (Burdon, 1987). The 18 races used in this study were found adequate to show the variability in the resistance spectrum of the 39 wheat varieties and accessions. The results obtained give a clue on the epidemiological and practical significance of knowing the resistance spectrum of the wheat varieties against stem rust.

The level of resistance spectrum expressed in terms of LIT*f* would signify the inherent ability of each Ethiopian wheat variety to stem rust pathogen population. In this study, TKR/J was the most virulent race attacking 25 *Sr* genes, while race NFB/B was the least virulent race attacking 6 *Sr* genes (Table 2). Varieties with high LIT*f* of 18 such as KBG-01, Bobitcho, Megal and Ude would have been expected to fit and maintain their resistance in a stem rust population that contains races with wide virulence spectrum. However, in reality where dynamic system of host-pathogen interaction exists, LIT*f* values could be used to explain epidemiological concepts such as boom and bust cycle, directional and stabilizing selection (Vanderplank, 1982) and magnitudes of fitness (Groth, 1984).

The level of resistance spectrum expressed as LIT*f* was also found to be a quantitative clue of the selection pressure in host-pathogen interaction. Burdon (1987) has stated that variation in the frequency distribution of infection types in a population provide clues concerning the nature and extent of the selection pressure placed on individual plant populations by fungal pathogens. Based on this premise, the epidemiological meaning and logical relationship between the observed LIT*f* of the varieties and the extent of selection pressure they could exert on the pathogen could be postulated as follows: In cases where gene-for-gene interaction works, varieties with highest LIT*f* values (broad resistance spectrum) exerts higher selection pressure on the pathogen population than those varieties with lower LIT*f* values (narrow resistance spectrum). Varieties with broad resistance spectrum cause greater pathogen co-evolution than those varieties with narrow resistance spectrum. Thus, varieties such as KBG-01, Bobitcho, Megal and Ude are the most important to dictate the evolution of new virulent race in the Ethiopian stem rust population than Shinna, Galama, Ketar and Gerado

Broad resistance spectrum is most likely the feature of new varieties with vertical resistance genes. The main reason could be the effect of series of screenings and selections procedures that are biased towards complete resistance. Varieties developed through such vigorous breeding procedures will possess gene(s) that are resistant to most of the prevailing races. Introduction of such new varieties in to the existing host-pathogen system could exert more selection pressure and disturbs the equilibrium as explained in the concepts of boom and bust cycle. Person *et al.* (1976) gave evidences to support the view that changes in the host population cause changes in selection pressure, and changes in the pathogen population rather than of changes due to new mutations. Groth (1984) has discussed the epidemiological reasons why favoured varieties with higher LIT*f* possess higher magnitude of fitness than others. Hence, the variation in LIT*f* values could be taken as a quantitative value that signify the differential levels of selection pressures exerted by different varieties on the pathogen population.

Regional gene deployment, gene pyramiding, multilines, mixtures and varietal diversity are strategies that are designed to lengthen the duration of vertical resistance genes (Vanderplank, 1982; Knott, 1989). Despite all their limitations, these strategies remains the best at present as long as breeding programs could not develop varieties with non-specific resistance genes as required (Roelfs *et al.*, 1992).

The range of variation in the resistance spectrum of emmer lines was found to be less than that of bread and durum wheat varieties. The number of genotypes of the different wheat might not be sufficient to compare them as a separate entity, and suggest their differential role in the Ethiopian stem rust epidemiology. However, there were important differences observed among the three wheat species in Ethiopia in terms of their origin, methods of development (selection or crossing) of a variety, and spatial distribution (extent of cultivation). These differences among the wheat species, could affect the host-pathogen co-evolution, population equilibrium and the rate of stem rust pathogen spread and evolution of new races. Emmer and durum wheat are landraces while all bread wheat varieties were introduced from outside. All emmer genotypes were selections while crossing programs developed from some durum wheat varieties. Emmer wheat grows in pocket sites of the Ethiopian highlands while durum and bread wheat varieties are extensively cultivated in the country. Cultivated Ethiopian emmer wheat accessions were found to be good sources of stem rust resistance (Naod Beteselassie *et al.*, 2007) and hence, they can be alternative sources of resistance to wheat breeding programs.

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