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Physiological Infective Characteristics of JRCQC and TTRTF Durum Specific *Pgt* Races on Wheat Cultivars

Ashagre Asnakew Zewde

Debre Zeit Agricultural Research Center, Ethiopian Institute of Agricultural Research, Debre Zeit, Ethiopia

Email address:
ashagre.asnakew@gmail.com

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Abstract: Wheat is an important industrial grain crop which is nominated as strategic for food. Nowadays, wheat is also cultivated in lowlands. Ethiopia is endowed as wheat producing potential mainly at three regions in the country, SNNPRS, Oromia and Amhara regions are potential wheat producing regions. Productivity of wheat in Ethiopia 2.96 t/ha is lowervas compared to global average, 3.65 tha^{-1} it is due to poor Agronomic practices, poor farm managements, limited potential yielding variety, poor soil fertility, rain distribution, limited diseases and insects managements. So the objective is to search new sources of resistance gene for the selected stem rust races. The experiment was conducted at Ambo Agricultural Research Center (AmARC) in 2019 using 78 wheat variants (19 released commercial varieties, 23 landraces, 35 EBI genotypes and McNair as susceptible check). Among those tested 77 tested materials: Oda, Leliso, Asassa, Cocorit 71, LD-357, DW/LRce 1, DW/LRce 2, DW/LRce 3, DW/LRce 6, DW/LRce8, DW/LRce 17, DW/LRce 18, DW/LRce 15, DW/LRce 16, DW/LRce 17, DW/LRce 20), 203792, 231541, 226807, 204519, 226233, 208267, 210795 and 203790) had shown low infection type (IT:; fleck to 2) to TTRTF virulence race. The results indicated tested wheat cultivars have different levels of resistance to the tested races. The results enables by doing further research it provides; the development resistance varieties to stem rust.

Keywords: Cultivars, Races, Resistance, JRCQC and TTRTF

1. Introduction

Wheat is one of the prehistoric crops which provide major energy requirement of the human diet across the world. Wheat is one of the globally popular cereal crops covers about 15% of the total cereal crop produced in the world [10]. It is an important industrial grain crop which ranks second among the most important cereal crops in the world after rice [6] In Ethiopia, wheat is strategic crop food security. Nowadays, wheat is also cultivated in lowlands [4]. The three regions in the country, SNNPRS, Oromia and Amhara regions are potential wheat producing regions. Productivity of wheat at in Ethiopia means of wheat production of 2.96 t/ha is lower as compared to the global wheat yield, 3.65 tha^{-1}. The low productivity and instability wheat productivity in Ethiopia is caused by biotic and abiotic stresses. Among biotic factors, the prominent diseases of wheat that currently contribute to significant yield losses include the fungal diseases rusts especially rust fungi is the major one.

Stem rust known as "shifty enemies" rapidly reproduced by mutation and recombination the pathogens to break the resistance genes [13]. *Pgt* is adapted to broader range of conditions, making it the most widespread of the three rust diseases [2]. Ethiopian highland areas are categorized as hot spot for the development of epidemics [7, 1]. The epidemics of virulent race; Digelu (TKTTF) race has caused local epidemics was occurred at Bale and Arsi [7].

Till now about 14 new races (TTKSK, TTKTF, TTKTK, JRCQC, TKTTF, TTKSC, TRTTF, SRKSC RRKSF, TTTH, HKPPF, HKNTF, TKKTF and RRTTF) have been recorded in Ethiopia [4]. Understanding the genetic effects of resistance, and knowing the available resistance genes in the germplasm help wheat breeders to formulate an efficient breeding program for the achievement of durable resistance breeding [9]. The protection of new varieties against Pgt races can be increased by gene pyramiding.

The repeated wheat production, favorable climates, presence of alternate host Berberry holstii and sexual recombination in Ethiopia favors for the rapid evolution and virulence diversity of the pathogen [8].

So far, several management techniques have been developed to use as well to reduce damage caused by stem rust disease; but is not enough and unable to meet the strategy. In search of new source of resistance against *Pgt* races to develop commercial varieties is the only option to insure healthy food and safe environment. Discovery of resistant gene for stem rust and continuous assessment for the presence of *Pgt* races will update management methods for the disease, races diversity and virulence spectrum of *Pgt* in selected regions of Ethiopia. In general, in order to combat the effects of stem rust disease on wheat production, it is advisable to use improved genotypes which have effectively resistant to the disease. To achieve this, it is vital to identify resistant wheat genotypes by evaluating their resistance to a diversified stem rust races both at seedling and adult plant stage. The objective is to search new resistance sources durum cultivars for durum specific stem rust races.

2. Materials and Methods

The experiment was conducted at Ambo Agricultural Research Center (AmARC) laboratory. The evaluation of durum wheat cultivars seedlings to the selected dominant races of Pgt was conducted at Ambo Agricultural Research Center. A total of 78 wheat variants; 19 commercial varieties, 23 landraces, 35 EBI genotypes and McNair as susceptible check were used. were used for the experiment For this study, spores of the dominant stem rust races previously identified in Ethiopia were multiplied on the McNair and collected in separate test tubes for inoculation. The JRCQC and TTRTF Pgt races were used for seedling resistance evaluations which are earlier isolated from durum wheat replicated three times done by CRD.

Sterilized soil composed of three growing Medias; sand, soil and farmyard manure mixed at the ratio of 2:1:1 by volume were used. Seventy eight durum variants (25 released varieties and 52 Cultivars; "McNair" as susceptible standard check which do not have known gene reaction. Of these materials seed were raised in 5cm diameter pots by spreading the seeds on filter paper in Petri dishes, moisten with water to allow the radicles sprout and sprouted seeds were planted in to growing pots.

Spores suspension were prepared in Soltrol 170 then inoculated onto a week cultivars and old McNair seedlings. The suspension was adjusted to 4-5 mg spores per 1ml lightweight mineral oil suspension and inoculated onto seedlings of the differentials using spore inoculators. The inoculated seedlings were placed on a table for 30 minutes until the Soltrol evaporate then the seedling is moistened with fine droplets of distilled water and placed in the incubation chamber in a dark at 18-22°C and 98-100% RH followed by exposure to light for 3-4 hours to facilitate infection. Natural daylight was supplemented with an additional 4 hrs/day that emitted by cool white fluorescent tubes arranged directly above plants [7]. The humidifier switched on for about 1:30 hours, so the seedlings to have enough moisture for the whole dark period to condition facilitate the initial infection. Then after, the seedlings were transferred to glass compartments in the greenhouse a temperature of 18-25°C and relative humidity of 60-70%.

Infection types (ITs) was scored; 14 days after inoculation using 0-4 score scale described by [16]. The IT readings of 3 (medium-size uredia with/without chlorosis) and 4 (large uredia) were regarded as susceptible. Other readings, i.e. 0 (immune/fleck), 1 (small uredia with necrosis) and 2 (small to medium uredia with chlorosis) were resistant (Figure 1).

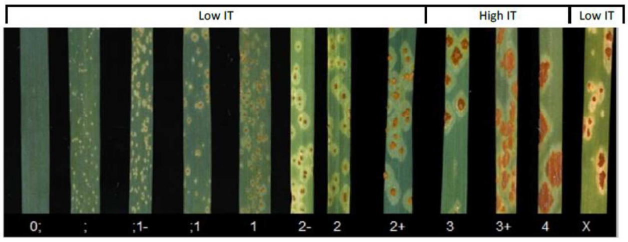

Figure 1. *IT of Pgt; Source: [16] Stackman et al., (1962).*

Seeds were soaked in distilled water for three days to initiate sprouting and five seeds of sprouts including susceptible check were separately planted in 5 cm diameter plastic pots (Figure 2). After sprout to start to grow; selected races 4mg spores in 1ml lightweight mineral oil suspension were inoculated. Inoculated plants were moistened with fine droplets of distilled water by using atomizer after 30 minutes of inoculation. For incubation, seedlings were placed in a dew chamber for 18 hrs dark period at 18-22°C and 98-100% RH Seedlings were allowed to dry for 2 hrs. Following this, the seedlings were transferred from dew chamber to glass compartments in the greenhouse.

Figure 2. *Selected durum wheat cultivars for seedling resistance type.*

3. Result and Discussion

Lemma stated that durum wheat varieties Foka and Yerer, LD 357, Bichena, Arsi robe and Kilinto were resistant to TTTTF for stem rust isolates [11]. In the seedling test, most durum wheat cultivars are resistant to stem rust [18]. Ethiopian durum wheat landraces signifies as sources traits for disease resistance [5, 14]. Similar results were also reported for wild Emmer wheat, Durum wheat and Bread wheat [12, 17] infection types of *P. tritici* on wheat cultivars.

Inefficiency of resistant gene in Ethiopia indicates that it is due to evolvement of virulence races on species specific crop durum wheat which are TTRTF and JQCRC and broad virulence spectrum of the strains. TTRTF has virulence for most wheat cultivars. The standard check for seedling resistance, McNair was susceptible to all races at the seedling stage in the greenhouse showing 3- infection types. Likewise, high percentage of 60.26% and 79.49% of durum cultivars are resistance to TTRTF and JRCQC stem rust races, respectively, with low infection types fluctuating from (flecks) to (2+). Race JRCQC is less virulent for tested durum cultivars than TTRTF, because it has showed high percentage of infection. Promising resistant with high resistance percentage 79.49 cultivalrs was recorded for JRCQC races but lower resistant varieties were recorded for TTRTF (Figure 3). The high frequency of susceptibility was 39.74% for TTRTF followed by JRCQC which made 20.52% of the evaluated varieties susceptible (Figure 3). Races JRCQC possess virulence on stem rust resistance genes *Sr13* and *Sr9e* on durum tested seedlings.

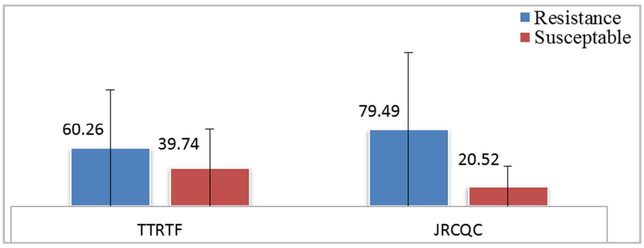

Figure 3. *TTRTF and JRCQC races infection range on wheat cultivars.*

3.1. Identified Durum Cultivars with Reaction of; Fleck and 2

Out of 77 durum wheat varieties evaluated at the seedling stage, none of cultivars has showed complete resistance (zero IT) while 28 cultivars: Kilinto, Foka, Oda, Leliso, Asassa, Cocorit 71, LD-357, Tesfaye, DW/LRce 1, DW/LRce 3, DW/LRce 6, DW/Lrce 10, DW/Lrce 12, DW/Lrce 13, DW/Lrce 14, DW/Lrce 15, DW/Lrce 16, DW/Lrce 20, EBI-222644, EBI-231541, EBI-226807, EBI-214507, EBI-226233, EBI-208267, DW/Lrce 8, EBI-204519, EBI-214422 and EBI-203790 had shown resistance infection types (IT = ";" or fleck to 2) for both races as compared to susceptable check McNair (Table 1). According durum wheat varieties Foka, LD 357 and Kilinto were resistant to TTTTF for stem rust isolates [11]. The use of these cultivars in for resistance breeding can be helpful for the advancement of new resistant wheat commercial variety. In the seedling test, most durum wheat cultivars are resistant to stem rust.

Table 1. *Identified durum variants with resistance reaction types of; fleck and 2.*

No.	Cultivars	TTRTF	JRCQC
1	Kilinto	;1+	2
2	Foka	;1	2
3	Oda	;1	;1+
4	Leliso	;1	;1+
5	Asassa	;1+	2
6	Cocorit 71	;1+	;1
7	LD-357	;	;1+
8	Tesfaye	2	;1+
9	DW/Lrce 1	;	;1+
10	DW/Lrce 3	2	2
11	DW/Lrce 6	1+	;1
12	DW/Lrce 10	;1	2
13	DW/Lrce 12	;1+	;1
14	DW/Lrce 13	;1c	2
15	DW/Lrce 14	;1	;1
16	DW/Lrce 15	;1+	;1
17	DW/Lrce 16	;	;
18	DW/Lrce 20	;1	2
19	EBI-222644	2	1+
20	EBI-231541	;1	;1
21	EBI-226807	2	;1+
22	EBI-214507	;1+	;1
23	EBI-226233	;1	;1
24	EBI-208267	2	;1+
25	DW/Lrce 8	1	;1
26	EBI-204519	;1	;1
27	EBI-214422	;1	;1
28	EBI-203790	;1	1+
29	McNair	3-	3-

3.2. Identified Durum Variants with Reaction of Moderately Susceptible (2 and 2$^+$)

Infection types with 2 to 2$^+$ are categorized as moderately susceptible for the tested materials. According to wild emmer wheat, durum wheat and bread wheat low infection types to *P. tritici*. Among tested wheat materials: Donmathew, Mangudo, Ude, Bakalcha, Toltu, Alemtena, DW/Lrce 2, DW/Lrce 4, DW/Lrce 7, DW/Lrce 17, EBI-5982, EBI-216078, EBI-231594, EBI-214423, EBI-231565, EBI-214423, EBI-231565, EBI-210795 and EBI-222704 as compared to McNair has shown ITs from (2 to 2$^+$) [17]. These cultivars showed moderate resistance seedling stage for stem rust races.

Table 2. *Identified durum variants with reaction types of (2 to 2$^+$) for TTRTF and JQCRC.*

No.	Cultivars	TTRTF	JRCQC
1	Donmathew	2	2+
2	Mangudo	2+	2-
3	Ude	2+	2
4	Bakalcha	2+	2
5	Toltu	2	2+
6	Alemtena	2+	2+
7	DW/Lrce 2	;1	2-
8	DW/Lrce 4	2	2+
9	DW/Lrce 7	2+	2+
10	DW/Lrce 17	2+	2+
11	EBI-5982	2	2+
12	EBI-216078	2	2+
13	EBI-231594	2	2+
14	EBI-214423	2+	2
15	EBI-231565	2+	2
16	EBI-210795	2	;1
17	EBI-222704	2	2+
18	McNair	3-	3-

3.3. Identified Durum Variants with Reaction of Susceptible (2$^+$ and 3$^-$)

Inefficiency of resistant gene in Ethiopia indicates that it is due to evolvement of virulence races on species specific crop durum wheat which are TTRTF and JQCRC and broad virulence spectrum of the strains. TTRTF *Pgt* race has complex virulence spectrum and spreading rapidly on Sr5, Sr21, Sr9e, Sr7b, Sr11, Sr6, Sr8a, Sr9g, Sr36, Sr9b, Sr17, Sr9a, Sr9d, Sr10, SrTmp, Sr38, and SrMcN; and avirulent on lines with Sr30, Sr24, and Sr31. TTRTF has virulence for most wheat cultivars. Sensitiveness of these cultivars and cultivars indicates that major gene for resistance is not available or they have fewer resistance genes resistance the races. Among tested cultivars; Utuba, Filakit, Obsa, Malefiya, DW/Lrce 5, DW/Lrce 9, DW/Lrce 11, DW/Lrce 19, EBI-208282, EBI-226913, EBI-222687, EBI-214353, EBI-226887, EBI-222600, EBI-222353, EBI-238128, EBI-222629, EBI-222619, EBI-214512, EBI-208311 and EBI-222643 have showed susceptible reaction (Table 3). Released commercial durum wheat varieties are under cultivation; this susceptible variety may use as the epidemics and year to year occurrence of stem rust in Ethiopia.

Table 3. *Identified durum variants with reaction of susceptible ($2^+$and 3^-).*

No.	Cultivars	Races	
		TTRTF	JRCQC
1	Utuba	2+	3-
2	Filakit	2+	3-
3	Obsa	3-	2-
4	Malefiya	2+	3-
5	DW/Lrce 5	2+	3-
6	DW/Lrce 9	2+	3-
7	DW/Lrce 11	2	3-
8	DW/Lrce 19	2+	3-
9	EBI-208282	2+	3-
10	EBI-226913	2+	3-
11	EBI-222687	2+	3-
12	EBI-214353	2+	2
13	EBI-226887	2+	3-
14	EBI-222600	2+	3-
15	EBI-222353	2+	3-
16	EBI-238128	2+	3-
17	EBI-222629	3-	2+
18	EBI-222619	2+	3-
19	EBI-214512	2+	3-
20	EBI-208311	2+-	3
21	EBI-222643	3-	2+
22	McNair	3-	3-

3.4. Identified Durum Variants with Mixed Reaction Fleck (;) to 3^-

Singh also stated that such genotypes (Table 4) could possess any of the genes either singly or in combination(s) for which the test pathotypes were avirulent [15]. The results of this study also support the fact that Ethiopian durum wheat accessions contain good sources of stem rust resistance (Table 4). Durum wheat has diversified reaction for different wheat stem rust races. The availability of varied gene provide for improvement of commercial varieties with good reaction to stem rust. Selam, DW/LRce 18, 236305 and 222619 of four cultivars used have revealed mixed reactions (; 1^+ to 3^-). Selam, DW/LRce 18, 236305 and 222619 of four cultivars used have revealed mixed reactions (; 1^+ to 3^-). This implies that the materials used has mixed gene may have shown mixed reactions. Mixed reactions for both races were observed on DW/Lrce 18 and 236305. also indicated that an incompatible response to two or more races could be an indication for the presence of an effective major R-gene against multiple races or a hint for multiple (horizontal) disease resistance due to the presence of two or more minor genes [3].

Table 4. *Identified durum variants with mixed reaction fleck (;) to 3^-.*

No.	Cultivars	Races	
		TTRTF	JRCQC
1	Selam	3-	;1+
2	DW/Lrce 18	3-	;1
3	EBI-236305	1+	3-
4	EBI-222353	3-	;1+
5	EBI-203792		
6	EBI-222619	;1	3-
7	EBI-204430	3-	2
8	McNair	3-	3-

4. Conclusion

The study indicated that there were effective cultivars among for races the races tested at seedling stage. The resistant variety will be assumed to have Sr24 and Sr31 resistant gene which can be used for the further evaluation of the virulence range and change rate in the wheat stem rust race populations to use effective resistant gene for wheat improvement. The result indicated that most varieties were susceptible to races TTRTF and JRCQC.

Some of the wheat lines used in the current study displayed highly susceptible infection types. However, some durum wheat such as Kilinto, Foka, Oda, Leliso, Asassa, Cocorit 71, LD-357, Tesfaye, DW/LRce 1, DW/LRce 3, DW/LRce 6, DW/Lrce 10, DW/Lrce 12, DW/Lrce 13, DW/Lrce 14, DW/Lrce 15, DW/Lrce 16, DW/Lrce 20, EBI-222644, EBI-231541, EBI-226807, EBI-214507, EBI-226233, EBI-208267, DW/Lrce 8, EBI-204519, EBI-214422 and EBI-203790 had shown low infection types (IT = ";" or fleck to 2) for both races as compared to McNair have good resistance at the seedling stage. For rusts outbreak, finding such resistance sources is crucial. This finding gives a clue as there are resistance genes against *Pgt* in some of the tested wheat lines. It is thus important to map the resistance genes by using molecular techniques and use these genes to develop resistant cultivars against. Resistance in seedling is complete and monogenic that is effective throughout all growth stages of wheat. Seedling resistance as a response to a combination of isolates was further inferred based on infection type scores.

Conflict of Interest

The author has not declared any conflict of interest on the paper.

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