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Virulence Spectrum of *Puccinia graminis* f. sp*. tritici* in Northwest Ethiopia

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ኢትዮጵያ ውስጥ ስንዴን ከሚያጠቁ በሽታዎች መካከል የግንድ ዋግ አንዱ እና ዋነኛው ነው፡፡ ይህንን በሽታ የሚያመጣው ተዋስያን አካል የአየሩን ምቹ ሁኔታ በመጠቀም እራሱን እየቀያየረ በሽታ የሚቋቋሙ የስንዴ ዝርያዎችን በማጥቃት ከፍተኛ የሆነ የምርት ቅነሳ ያደርጋል፡፡ የዚህ ጥናት ዓላማ የነበረው በሰሜን ምዕራብ ኢትዮጵያ ውስጥ ያለውን የስንዴ የግንድ ዋግ ዝርያ መለየት ነው፡፡ ሰማንያ ስድስት ናሙናዎች በ2010 እና በ2011 የምርት ዘመን ተሰብስበው 20 ድፍሬንሻሎችን በመጠቀም አራት (TTTTF, TKTTF, TKPTF, and TTRTF) የስንዴ ግንድ ዋግ ዝርያዎችን ለመለየት ተችሏል፡፡ ከእነዚህ አራት ዝርያዎች ውስጥ በ2010ና በ2011 በተከታታይ በአማካይ በመቶኛ TTTTF 60.4 እና 60 የሚሆነውን የሚሸፍን ሲሆን በሁለተኛነት ደረጃ TKTTF 38.7 እና 37.3 ሸፍኖ ታይቷል፡፡ ሌሎች የግንድ ዋግ ዝርያዎች በሁለቱም የምርት ዘመን በአማካይ በመቶኛ 1 የሚሆነውን የሸፈኑ ሲሆን እነዚህ ሁለት ዝርያዎች የተገኙትም፤ TKPTF በ2010 ከደቡብ ጎንደር ፋርጣ ሲሆን TTRTF በ2011 ከሰሜን ጎንደር አምባ ጊዮርጊስ ላይ ብቻ ነው፡፡ የግንድ ዋግ የሚቋቋሙ ዘረመልን የያዙ ድፍሬንሻሎች እንደ Sr5, Sr21, Sr9e, Sr7b, Sr6, Sr8a, Sr9g, Sr36, Sr17, Sr9a, Sr9d, Sr10, SrTmp, Sr38 እና SrMcN ሁሉም በተለዩ የዋግ ዝርያዎች የተጠቁ ሲሆን Sr24 እና Sr31 ዋግ የሚቋቋሙ ዘረመልን የያዙ ድፍሬንሻሎች ግን ሙሉ በሙሉ የተለዩትን የዋግ ዝርያዎችን መቋቋም ችለዋል፡፡ በአሁኑ ጥናት Sr31 እና በአሁኑና ከዚህ በፊት ኢትዮጵያ ውስጥ በተደረጉ ጥናቶች Sr24 ዘረመልን የሚያጠቃ የግንድ ዋግ ዝርያ ያለመኖሩ ስለተረጋገጠ እነዚህን ዘረመሎች ከሌሎች የግንድ ዋግ መቆጣጠሪያ መንገዶች ጋር በመጠቀም አሁን ያለውን የግንድ ዋግ ዝርያ መቋቋም የሚችል የስንዴ ዝርያ ማዳቀል ይቻላል፡፡

Abstract

Stem rust caused by Puccinia graminis f. sp. tritici is one of the most important diseases of wheat in Ethiopia. The pathogen can produce new races that can overcome resistant varieties and cause epidemic under favorable environmental conditions resulting in serious yield losses. The study was carried out to determine the virulence spectrum of Puccinia graminis f. sp. tritici in northwest Ethiopia. Eighty-six stem rust samples were collected during 2017 and 2018 cropping seasons and analyzed on 20 standard stem rust differential lines which resulted in the identification of four races (TTTTF, TKTTF, TKPTF, and TTRTF). Of these races, 60.4 and 60% of the isolates were TTTTF followed by 38.7 and 37.3% of TKTTF (Digalu race) in 2017 and 2018, respectively. These two races accounted for almost 99% of the stem rust population. The least virulent races were TKPTF and TTRTF that accounted 1% in both seasons and were detected only at single location (Farta in South Gondar in 2017 and Amba Giorgis in North Gondar in 2018). The differential hosts carrying resistance genes Sr5, Sr21, Sr9e, Sr7b, Sr6, Sr8a, Sr9g, Sr36, Sr17, Sr9a, Sr9d, Sr10, SrTmp, Sr38 and SrMcN were susceptible to the four races identified in northwest Ethiopia. Two differential hosts carrying the resistant genes Sr24 and Sr31 were effective against all the four races identified in this study. Absence of virulence against lines carrying Sr31 in the present and Sr24 in the present and previous studies suggests that these genes may serve as a source of resistance in combination with other wheat stem rust management systems to the current rust races in northwest Ethiopia.

Introduction

Wheat is one of the most important cereals cultivated in Ethiopia. It is largely grown in the highlands of Ethiopia, constitutes 15.6% of the annual cereal production, and plays an important role in supplying the population with various nutritional foods (CSA 2017). It is grown at altitudes ranging from 1500 to 3000 m, between 6-16^o N latitude and 35-42^o E longitude. The most suitable agro-ecological zones, however, fall between 1900 and 2700 m. l. and the major wheat producing areas in Ethiopia are located in Arsi, Bale, Gondar, Gojam, Illuababora, Shewa, Sidama, western Hararghe and Tigray zones (Bekele *et al.* 2000).

Wheat is widely grown in the northwest part of Ethiopia (Gondar and Gojam). It covers over 290,000 hectares of land and yielded over 760,000 tons in 2016/17, which is 58% of the total Amhara region wheat production (CSA 2017). Though the edaphic conditions of Gondar and Gojam are conducive to produce wheat, the average productivity (2.4 t/ha) is in general low when compared to the national average yield of 2.6 tha^{-1} (CSA 2017). The low productivity is attributed to several factors including biotic (diseases, insect pests and weeds) and abiotic (moisture, soil fertility, etc) and adoption of new agricultural technologies (Zegeye *et al*. 2001). Among these factors, diseases play significant role in yield reduction. Among the various diseases affecting wheat, stem rust caused by the fungus *Puccinia graminis* f. sp. *tritici* has been the most devastating under favorable conditions. This disease is also known as black rust of wheat due to the abundant production of shiny black teliospores that form at the end of the growing season or with unfavorable conditions (Singh *et al.* 2002). Under favorable environmental conditions, stem rust can cause yield losses of up to 100% in susceptible wheat varieties (Roelfs 1985b). The yield loss due to this disease is usually greatest when the disease becomes severe before the grain is completely formed, but the resistance level of the varieties grown, the weather conditions, and the onset of the disease (Luig 1985; Roelfs 1985a) generally influence yield losses. In Ethiopia, yield losses due to stem rust have been reported to be in the range of 61-100% depending on the susceptibility of the variety and environmental conditions (Bechere *et al*. 2000).

The high virulence diversity and evolution rate of *Puccinia graminis* f. sp. *tritici* (Admassu *et al.* 2009) and the variability of virulence in *Pgt* populations, the ability of urediniospores disseminated by wind over long distances and an exponential reproduction capacity makes wheat stem rust a threat to wheat production (Leonard 2001). According to Singh et al. (2006) and Periyannan *et al.* (2013), Ethiopia is considered as a hot spot for the development and spread of new stem rust races. These new races have reduced the number of major rust resistance genes that are available for use (Kolmer 2005). According to Leppik (1970), the highlands of Ethiopia are considered as hot spots for the development of stem rust diversity. Previous studies showed that most previously identified races were virulent on most of varieties grown in Ethiopia (Admassu *et al*. 2009; Naod 2004). Beyene (2018) also reported that wheat growing areas of Ethiopia particularly Amhara, Oromia and southern Ethiopia are hot spot areas for appearance of virulent pathotypes and/or races of stem rust. Therefore, the study was carried out to determine the virulence spectrum of *Puccinia graminis* f. sp. *tritici* in northwest Ethiopia.

Materials and Methods

The study was conducted at Ambo Agricultural Research Center using samples collected from northwest Ethiopia (Gojam and Gondar).

Sample collection, production of mono-pustule isolates and multiplication

Collection of stem rust samples was done in major wheat growing zones of northwest Ethiopia from farmers' wheat fields and trial plots during the 2017/18 and 2018/19 cropping seasons. Three samples of stem rust infected leaf sheath and /or stem of wheat plants were collected and cut into small pieces of 5-10 cm in length using scissors, and placed in paper bags after the leaf sheath has been separated from the stem to keep the leaf sheath dry. Three samples were collected from each field. The samples were labeled and transported to Ambo Agricultural Research Center for analyses. Eighty-six stem rust samples were used for final analyses from 110 collected samples. South Gondar, North Gondar, East Gojam, and West Gojam were represented by 30, 15, 20 and 21 samples, respectively.

Development of mono-pustule isolates and multiplication of these isolates was done on the universally rust susceptible variety "McNair", which does not carry known stem rust resistance genes. Five seedlings of "McNair" were raised in 8 cm diameter pots containing sterilized soil, sand, and manure in a ratio of 2:1:1 mixture, respectively. Inoculations were done following the methods and procedures developed by Stakman *et al.* (1962). Bulked urediniospores from each field was suspended in lightweight mineral oil, Soltrol 170 and sprayed onto seven-day-old seedlings of McNair using atomized inoculator (Vacuubrand inc., Wertheim, Germany). After 20 minutes, distilled water was sprayed to inoculated plants using bottle sprayer. Seedlings were allowed to dry for about 1-2 hours before placing them in an incubation chamber. They were kept in an incubation chamber for 18 hours in the dark at 18-22°C followed by exposure to light for 3-4 hours to provide a condition favorable for infection. Then, the seedlings were transferred from the dew chamber to growth chamber where conditions were adjusted at 12 hours photoperiod, a temperature of 18-25°C and relative humidity of 60-70% (Stubbs et al. 1986). After ten days of inoculation, leaves containing a single fleck that produce single pustule were selected from the base of the leaves. The remaining seedlings in pots were removed using scissors. Only two leaves with single pustule were separately covered with cellophane bags and tied up at the base with a rubber band to avoid cross contamination (Fetch and Dunsmore 2004). Single isolate was developed from each field and eighty-six isolates were used for the final race analyses. After two weeks of inoculation, spores from each mono-pustule were collected using power operated vacuum aspirator and were stored separately in gelatin capsules. Multiplication of spores was done by inoculating seven-day-old 'McNair' seedlings with a suspension of urediniospores of each mono-pustule isolates. Each mono-pustule isolates were multiplied in separate pots following the procedures mentioned earlier. The urediniospores descending from one pustule made up of a single isolate.

Inoculating differential lines and race designations

The modified North American stem rust differentials set containing 20 lines was used for the race analysis; and seeds of the differential hosts were obtained from Ambo Agricultural Research Center. Five seeds of each of the twenty wheat stem rust differential lines and one susceptible variety 'McNair' were grown in 8 cm diameter pots containing sterilized soil, sand and manure in a ratio of 2:1:1 mixture, respectively, in the greenhouse. The susceptible variety 'McNair' (without *Sr* gene) was used to ascertain the viability of spores inoculated to the differential hosts. Each rust isolate derived from single pustule was suspended in Soltrol 170. The suspension was adjusted to 4 x 10^5 spores per ml of oil and was sprayed onto seedlings of the differential lines (Table 1) following the procedure of Stakman *et al.* (1962).

Table 1. Wheat stem rust differential lines used race analysis

Differential line	*Sr* gene	Origin/pedigree
LcSr24Ag	24	Little Club/Agent (CI 13523)
W2691SrTt-1	36	CI12632 *T. timopheevii*
ISr7b-Ra	7b	Hope/Chinese Spring
ISr8a-Ra	8a	Rieti/Wilhelmina//Akagomughi
CnSSrTmp	Tmp	Triumph 64(CI 13679)/ Chinese Spring
Sr31(Benno)/6*LMPG	31	Kavkaz
CnS-T-.mono-deriv	21	Einkorn CI 2433
Trident	38	Spear*4/VPM (PI519303)
ISr9a-Ra	9a	Red Egyptian/Chinese Spring
ISr9d-Ra	9d	Hope/Chinese Spring
Combination VII	17	Esp 518/9
ISr5-Ra	5	Thatcher/Chinese Spring
ISr6-Ra	6	Red Egyptian/Chinese Spring
W2691Sr9b	9b	Kenya 117A
Vernsteine	9e	Little Club//3*Gabo/2*
W2691Sr10	10	Marquis*4/Egypt NA95/2/2*W2691
BtSr30Wst	30	Festival/Uruguay CI0837
CnsSr9g	9g	Selection from Kubanka(CI1516)
ISr11-Ra	11	Kenya C6402/Pusa4/Dundee
McNair 701	McN	CI 15288

Stem rust infection types (ITs) on the differential lines were scored 14 days after inoculation using the 0-4 scale (Stakman *et al.* 1962). Infection types were grouped into two, where, low infection type (resistant) was 0 to 2+; and high infection type (susceptible) was 3- to 4+ (Table 2). Race designation was done by grouping the differential hosts into five subsets (Table 3). Each isolate was assigned using a five-letter race designation based on its reaction on the differential lines (Roelfs and Martens 1988; Jin *et al*. 2008).

Table 2. Description of infection types used in classifying the reactions of stem rust on leaves of wheat seedlings

Class	IT	Description of symptoms
Immune	0	No sign of infection on the naked eye
Very resistant	;	No uredia, but distinct flakes of varying size, usually a chlorotic yellow but occasionally necrotic
Resistant	1	Small uredia surrounded by yellow chlorotic and necrotic area.
Moderately resistant	2	Small to medium sized uredia, typically in a dark green island surrounded by a chlorotic area
Mesothentic/ Heterogeneous	x	A range of infection type from resistant to susceptible scattered randomly on a single leaf caused by a single isolate not mixture
Moderately susceptible	3	Medium sized Uredia. Usually surrounded by a light green chlorotic
Susceptible	4	Large uredia with a limited amount of chlorosis may be diamond shaped
Modified characters		
Lower uredia	=	Uredia much smaller than typical and at the lower limit of the infection type
Small uredinia	-	Uredia smaller than normal
Large uredinia	+	Uredia larger than normal
Largest uredinia	++	Uredia much larger than typical and at the upper limit for the infection type

Table 3. Nomenclature of *Puccinia graminis* f. sp. *tritici* based on 20 differential wheat lines

Pgt – code		Infection types produced on near-isogenic *Sr* lines			
	Set 1	5	21	9e	7b
	Set 2	11	6	8a	9g
	Set 3	36	9b	30	17
	Set 4	9a	9d	10	Tmp
	Set 5	24	31	38	McN
B		Low*	Low	Low	Low
C		Low	Low	Low	High**
D		Low	Low	High	Low
F		Low	Low	High	High
G		Low	High	Low	Low
H		Low	High	Low	High
J		Low	High	High	Low
K		Low	High	High	High
L		High	Low	Low	Low
M		High	Low	Low	High
N		High	Low	High	Low
P		High	Low	High	High
Q		High	High	Low	Low
R		High	High	Low	High
S		High	High	High	Low
T		High	High	High	High

*Source: Roelfs and Martens (1988); Jin et al. (2008) *Low- Infection types 0; 1, and 2 and combinations of these values; **High- Infection types 3 and 4 and a combination of these values.*

Results and Discussion

Virulence structure of stem rust pathogen

Out of the total stem rust isolates collected during the two seasons, 86 viable isolates were tested for their virulence, and four races namely TKTTF, TTTTF, TTRTF and TKPTF were identified. Of these, the races that were detected in both seasons were

TKTTF and TTTTF. Race TKPTF was identified only in 2017 while TTRTF was detected only in 2018 main cropping season. This could be an indication of virulence diversity within the *Pgt* population in northwestern Ethiopia. According to Admassu and Fekadu (2005) similar number but different races were identified from samples collected in 2003 in northwest region (three races in Gojam and one race in Gondar). This showed that the variability of the *Pgt* across locations and seasons. Previous studies also showed being there of TTTTF and TKTTF races of *Puccinia graminis* f. sp. *tritici* in Ethiopia *(*Admassu *et al.* 2009; Hailu et al. 2015).

Table 4. Races of *Puccinia graminis* f. sp. *tritici* and their frequency in northwest Ethiopia in 2017 and 2018 main cropping seasons

Zone	Race	Number of isolates		Frequency %	
		2017	2018	2017	2018
South Gondar	TTTTF	20	-	66.7	-
	TKTTF	9	-	30.0	-
	TKPTF	1	-	3.3	-
North Gondar	TTTTF	1	6	33.3	50
	TKTTF	2	5	66.7	41.7
	TTRTF	-	1	-	8.3
West Gojam	TTTTF	3	10	75.0	58.8
	TKTTF	1	7	25.0	41.2
East Gojam	TTTTF	2	12	66.7	70.6
	TKTTF	1	5	33.3	29.4

Among identified races TTTTF was the most frequent and dominant race with frequencies of 60.4 and 59.8% in 2017 and 2018, respectively (Table 4). The second abundant and virulent race was TKTTF (Digalu race). It had frequencies of 38.7 and 37.4% in 2017 and 2018 cropping seasons, respectively. Races TTTTF and TKTTF accounted for 99% of the stem rust population of northwest Ethiopia in the study period. The remaining two races, TKPTF and TTRTF were the least abundant with frequency of 1%. Race TKPTF and TTRTF were each detected only at single location at Farta from South Gondar in 2017 and Amba Giorgis from North Gondar in 2018, respectively. This study revealed that race TTTTF was the most dominant and virulent of the stem rust population in northwest Ethiopia. It was widely distributed in the major wheat growing parts of the region in both seasons. Lemma et al. (2015) reported that this race was detected in East Shewa zone in central Ethiopia in trace level. TTTTF was detected on samples collected from durum wheat, bread wheat and barley varieties in 2014 and 2015 cropping seasons in Ethiopia (Netsanet et al. 2018). It was also reported in Iran (Afshari *et al.* 2015) and was detected and attacked thousands of hectares of durum wheat in Sicily, Italy in 2016, which is claimed to have caused the largest stem rust outbreak in Europe in recent times (FAO 2017).

The second most dominant race, TKTTF-called Digalu race-, [virulent on 17 stem rust resistance genes (Table 5)] had also wide distribution in northwest part of the country in both seasons. The Digalu race (TKTTF) was first detected on a widely grown bread wheat variety Digalu in 2012 in Ethiopia; and reported to affect large hectares of wheat in Bale and Arsi in subsequent years (Hailu et al. 2015; Worku *et al.* 2016). The newly emerged stem rust race TKTTF caused major damage to wheat production in Ethiopia and most farmers who grew Digalu variety were highly affected (Olivera et al. 2015). This race was making further advances in the country, i.e. central Ethiopia (Gurage) according to the study made by Abrahim et al. (2018). Race TKTTF was also detected in North Africa (Egypt in 2013) and Middle East [Iran in 2010 and Lebanon in 2012 (Olivera *et al.*, 2015)]. The race is highly virulent to the variety 'Digalu' which possesses stem rust resistance gene *SrTmp* that is effective against the Ug99 race group, but ineffective against TKTTF. Data from the present study indicate that three stem rust resistance genes, namely *Sr31*, *Sr24* and *Sr11* were effective against TKTTF (Table 5). Studies indicated that TKTTF does not belong to the Ug99 lineage based on avirulences to *Sr11* and *Sr31* and molecular fingerprints (Olivera et al. 2015).

Races TKPTF and TTRTF were identified in South Gondar, Farta and North Gondar, Amba Giorgis in 2017 and 2018, respectively, but at low frequencies. According to Lidiya *et al*. (2019), race TTRTF is also reported from Arsi and Bale in trace level. TTRTF was detected from samples collected during the 2015-2016 from the International Stem rust Trap Nursery and commercial wheat fields in Sakha, the most important wheat-growing region in Egypt (Samar and Szabo 2018). Race TKPTF was also reported in Germany wheat fields in summer 2013 (Olivera et al. 2017). The pathogen population for virulence combinations varied slightly from location to location. The differences in race compositions in the study zones might be due to variation in locations and time. According to Roelfs *et al*. (1992), the prevalence of races in a specific season and region depends on the type of wheat cultivars grown and to some extent on the predominant environmental conditions, especially temperature.

There was some variation in the virulence spectrum of the races within northwest Ethiopia. In 2017, the 30 isolates analyzed from South Gondar were assigned to 3 races namely TTTTF, TKTTF and TKPTF. Similarly, the 4, 3 and 3 isolates studied in West Gojam, North Gondar and East Gojam, respectively, belonged to races TKTTF and TTTTF. On the other hand, the 17 isolates each from West Gojam and East Gojam analyzed in 2018 were assigned to 2 races namely TTTTF and TKTTF while the 12 isolates studied in North Gondar belonged to race TTTTF, TKTTF and TTRTF.

In both seasons, the race frequency showed a similar trend as TTTTF was predominant with a frequency of more than 60%, followed by race TKTTF with frequencies of 38.7 and 37.4% in 2017 and 2018 seasons, respectively. The less abundant races, TKPTF and TTRTF, were confined only to single locations, while races TTTTF and TKTTF were present in South Gondar, North Gondar, West Gojam and East Gojam Zones.

Table 5. Virulence /Avirulence spectrum of the *Pgt* races identified in northwest Ethiopia in 2017 and 2018 main cropping seasons

Race	Virulence	Avirulence
TTTTF	*5, 21, 9e, 7b, 11, 6,8a,9g,36,9b,30,17,9a,9d,10, Tmp, 38, McN*	*24,31*
TKTTF	*5, 21, 9e, 7b, 6,8a,9g,36,9b,30,17,9a,9d,10, Tmp, 38, McN*	*11, 24,31*
TKPTF	*5, 21, 9e, 7b, 6,8a,9g,36,30,17,9a,9d,10, Tmp,38, McN*	*9b, 11,24,31*
TTRTF	*5, 21, 9e, 7b, 11,6,8a,9g,36,9b,17,9a,9d,10, Tmp, 38, McN*	*24,30,31*

Virulence to *Sr* resistance genes

The widest virulence spectrum was noted on race TTTTF making 90% of *Sr* genes ineffective. It was virulent against all except *Sr24* and *Sr31* genes. Races TKTTF, TKPTF and TTRTF defeated 85, 80, and 85% of the *Sr* genes in the wheat differential lines, respectively. Of these, TKTTF and TTRTF varied from one another by a single gene change, which were virulent to all differential lines except *Sr11*, *Sr24*, *Sr31* and *Sr30, Sr24*, *Sr31*, respectively. Similarly, TKTTF and TKPTF varied from one another by a single gene change, which were avirulent to *Sr11*, *Sr24*, *Sr31,* and *Sr9b, Sr11, Sr24*, *Sr31,* respectively. The narrowest virulence spectrum was recorded on race TKPTF with 80% of the *Sr* genes ineffective.

Most of the races were virulent to one or more of the resistance genes (Table 5). For instance, the differential hosts carrying the resistance genes *Sr5*, *Sr21*, *Sr9e*, *Sr7b*, *Sr6*, *Sr8a*, *Sr9g*, *Sr36*, *Sr17Sr9a*, *Sr9d*, *Sr10*, *SrTmp*, *Sr38*, and *SrMcN* were ineffective to all the races identified in the study in both seasons (Table 5). *Sr9b* and *Sr30* were ineffective against 75% of the races while *Sr11* was ineffective against 50% of the races identified (Table 6). On the contrary, stem rust resistance genes *Sr24* and *Sr31* were effective against all races identified in northwest Ethiopia in both seasons.

Even if it is risky to use *Sr31* as a source of resistance as it is ineffective against Ug99 and its variants; they were not detected in this study and it is possible to use resistance gene *Sr31* for the source of resistance for breeding program to the current dominant wheat stem rust races in the northwest Ethiopia. However, *Sr24* has been consistently effective against races prevalent in various parts of Ethiopia over time (Admasu et al. 2010; Abebe et al. 2012; Hailu et al. 2015). This concurs with reports of Roelfs et al. (1992) and CIMMYT (2005) that indicated its effectiveness against many races across the globe. Hence, the national wheat-breeding program may consider utilizing *Sr24* as a source of resistance to stem rust. Virulence against *Sr*5, *Sr*6, *Sr*9a, *Sr*9d, *Sr*9e, and *Sr*17 is common worldwide (Roelfs *et al*. 1992). Study by Admassu and Fekadu (2005) reported the effectiveness of *Sr24* against races studied from 2001 -2004 in Ethiopia. *Sr11* was effective against the second dominant virulent race TKTTF, also known as the 'Digalu race', which is virulent against the widely grown wheat cultivar 'Digalu'.

Table 6. Virulence frequency of *Pgt* races collected in northwest Ethiopia in 2017 and 2018 to single gene wheat differential lines

Stem rust resistance gene (*Sr gene*)	Virulence frequency (%)	Stem rust resistance gene (*Sr gene*)	Virulence frequency (%)
Sr5	100	*Sr30*	75
Sr21	100	*Sr17*	100
Sr9e	100	*Sr9a*	100
Sr7b	100	*Sr9d*	100
Sr11	50	*Sr10*	100
Sr6	100	*SrTmp*	100
Sr8a	100	*Sr24*	0
Sr9g	100	*Sr31*	0
Sr36	100	*Sr38*	100
Sr9b	75	*SrMcN*	100

The present study showed that more than 90, 85, 80 and 85% of the *Sr* genes were ineffective against TTTTF, TKTTF, TKPTF and TTRTF, respectively. The differential host carrying the resistance gene *SrMcN* was ineffective to all isolates studied (Table 5). Admassu et al. (2010) and Lemma et al. (2015) reported similar results.

Conclusion

Stem rust, caused by *Puccinia graminis* f. sp. *tritici* (*Pgt*), is one of the most serious diseases of wheat worldwide. The discovery of new *Pgt* races in Eastern Africa, Ug99 and its variants, brought a new threat to global wheat production. *Pgt* race analysis is very vital for producing resistant varieties and providing management options timely. From this study four races were identified from 86 samples collected from northwest Ethiopia. Of these races, the highly virulent race TTTTF was the most dominant race which accounted for 60.4% of the races identified followed by race TKTTF with a frequency of 38.7%. Races TTRTF and TKPTF with frequency of 1% in the study area were also the least virulent ones. Differential host carrying *Sr24* and *Sr31* genes were effective against all the races identified in the study area followed by *Sr11*, *Sr9b* and *Sr30*. All other *Sr* genes were ineffective to all races. Absence of virulence against lines carrying *Sr31* in the present and *Sr24* in the present and previous studies suggests that these genes may serve as a source of resistance to the current rust races in northwest Ethiopia.

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