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# Seedling Evaluation, Gene Postulation, and Characterization of Bread Wheat Genotypes against Wheat Stem Rust (Puccinia graminis f. sp. tritici) Pathotypes in the Highlands of Bale, Oromiya, Ethiopia

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## Abstract

Wheat stem rust, caused by Puccinia graminis f. sp. tritici (Pgt), is one of the most destructive diseases of wheat worldwide. In Ethiopia, the disease is widespread in humid and warm wheat-growing environments, particularly in central and southeastern regions. Understanding resistance genes to Puccinia graminis f. sp. tritici is a fundamental requirement for the effective utilization and deployment of resistant sources in integrated wheat stem rust management. Field and greenhouse experiments were conducted to evaluate slow-rusting resistance and to investigate stem rust resistance spectra in bread wheat genotypes at the seedling stage during the 2021/22 main cropping season. A total of 121 bread wheat genotypes were included in the study. Field experiments were conducted at Sinana and Agarfa, while seedling evaluations were carried out under greenhouse conditions at Debre Zeit Agricultural Research Center. The genotypes were tested against seven dominant stem rust races: TTTTF, TTKTT, TTKTTF, TTRTF, TKTTF, TKKTF, and TTKSK (UG99), which were originally obtained from Ambo Agricultural Research Center. Using 20 near-isogenic lines (NILs), four resistance genes—Sr11, Sr36, Sr30, and Sr24—were postulated in 15 wheat genotypes. Twenty genotypes showed resistance, while 88 genotypes carried unidentified Sr genes. A total of 28 genotypes exhibited intermediate to susceptible reactions at the seedling stage (infection types ranging from 2+ to 4). Among the evaluated genotypes, three varieties—Kingbird, Pavon 76, and Sofumer—exhibited slow-rusting resistance with an area under disease progress curve (AUDPC) value of less than 300. Fifteen genotypes were identified as true slow-rusting types based on combined field and seedling evaluations. These genotypes, carrying Sr11, Sr36, Sr30, and Sr24 genes, along with the identified slow-rusting lines, can be utilized in future breeding programs aimed at developing durable resistance to wheat stem rust. Further studies incorporating additional NILs and molecular markers are recommended to effectively identify both seedling and adult plant resistance (APR) genes in bread wheat genotypes.

## INTRODUCTION

Diseases are among the major biotic factors limiting wheat productivity in Ethiopia. More than 40 diseases have been reported on wheat in the country. Among these, fungal diseases such as rusts (stem, stripe, and leaf rust), Fusarium head blight, Septoria blotch, *Helminthosporium* spp., and tan spot are the most prevalent and economically important [5]. Wheat stem rust, caused by *Puccinia graminis* f. sp. *tritici*, is one of the most devastating diseases and can result in up to 100% yield loss on susceptible varieties, particularly in the Arsi and Bale zones of Oromia Region [Hodson, 2014; 41].

Ethiopia is considered one of the global hotspots for wheat rust pathogens. East Africa, including Ethiopia, has been recognized as a hotspot for stem rust due to the emergence of new virulent races such as TTKSK, commonly known as Ug99. This race was first identified in Uganda in 1998 [48] and was the first known race with virulence to the Sr31 resistance gene, which is located on the 1BL.1RS translocation from rye (*Secale cereale*). Ug99 was detected in Ethiopia in 2003 [3], and most Ethiopian wheat germplasm was found to be susceptible or partially susceptible to this race and its variants [34].

Another virulent race, TKTTF, was first detected in Ethiopia in 2012 at trace levels and reached epidemic proportions in southeastern Ethiopia by 2013, particularly on the widely grown cultivar Digalu, which carries the SrTmp gene (Hodson, 2014). Severe stem rust epidemics in 2013 and 2014 caused up to 100% yield loss in affected areas [5]. Unlike Ug99, TKTTF is genetically unrelated to the Ug99 race group.

Recent studies have reported the presence of multiple Pgt races in Ethiopia, indicating high pathogen diversity and rapid evolution. Hei et al. (2018) identified nine Pgt races (TKTTF, TTTTF, TTKSK, TRTTF, PKTTF, TTTSF, SJPQC, JRCSF, and JRCQC) from samples collected in 2014 and 2015. In 2018, a new race, TTKTT, was detected in Ethiopia [23]. This race overcomes most known resistance genes except Sr36 and is particularly important due to its virulence to SrTmp, Sr24, and Sr31, indicating increasing variability and virulence in Ethiopian Pgt populations.

Stem rust resistance follows the gene-for-gene hypothesis proposed by Flor (1956). Race-specific (vertical) resistance is usually effective only against certain pathogen races and is often associated with hypersensitive reactions. However, such resistance is prone to breakdown due to the emergence of new virulent races [16]. In contrast, race-nonspecific or adult plant resistance (APR) is generally more durable and is expressed quantitatively at later growth stages. APR can be assessed using epidemiological parameters such as disease severity, infection rate, and area under the disease progress curve (AUDPC) [32].

Slow-rusting resistance is an important component of durable resistance, as it reduces disease development and spread over time [18]. Incorporating slow-rusting and APR genes into commercial cultivars has been recognized as an effective strategy for achieving long-term control of rust diseases. However, little is known about the resistance status of many widely grown Ethiopian wheat genotypes.

Host resistance remains the most effective and environmentally friendly method of managing stem rust. Pyramiding multiple resistance genes into a single cultivar is considered the most reliable strategy for durable resistance. Identification of resistance genes through gene postulation is an important approach for characterizing resistance in wheat genotypes. Gene postulation involves testing genotypes with known and unknown resistance genes against multiple pathogen races and comparing infection type patterns [11; 30].

Understanding the genetic basis of resistance in Ethiopian wheat germplasm is essential for developing effective breeding strategies. Identifying resistance genes in elite and commercial varieties provides valuable information for deploying resistant cultivars and monitoring pathogen virulence dynamics. Therefore, this study was designed with the following objectives was to investigate wheat stem rust resistance spectra in bread wheat genotypes and commercial varieties at the seedling stage and to determine slow-rusting resistance in elite bread wheat genotypes under field conditions.

## MATERIALS AND METHODS

**Description of the Study Areas**

The field experiment was conducted at Sinana and Agarfa Research Stations (Figure 1). These locations are recognized as hotspot areas for both yellow rust and stem rust diseases. The seedling test was carried out under greenhouse conditions at Ambo and Debre Zeit Agricultural Research Center. Sinana Agricultural Research Center (SARC) is located at 7°7′3" N and 40°13′38" E at an altitude of 2458 meters above sea level (m.a.s.l) in Bale Zone, approximately 463 km southeast of Addis Ababa. The area receives an average annual rainfall of about 590 mm during the main cropping season and 560 mm during the Belg (spring) season. The monthly average minimum and maximum temperatures are 9.42°C and 21.16°C, respectively. Agarfa Research Station is situated at 7.26° N and 39.84° E at an altitude of 2489 m.a.s.l in the cool sub-humid agro-climatic zone of Bale. The average annual rainfall of the area ranges from 1000 to 1100 mm, and the dominant soil type is pellic vertisol. Ten-year historical weather data, including rainfall, maximum and minimum temperature, and relative humidity, were collected and summarized (Appendix Table 10).

**Plant Materials**

A total of 121 bread wheat genotypes were used in this study. These included 18 commercial varieties, 100 advanced breeding lines, and three standard checks (Appendix Table 2). The plant materials were obtained from the National Wheat Breeding Program at Kulumsa Agricultural Research Center (KARC) and Sinana Agricultural Research Center.

In the greenhouse experiment, the susceptible wheat variety McNair 701 (CItr 15288) was used as a susceptible check in seedling tests. In the field experiment, three widely grown bread wheat varieties namely; Dure, Digalu, and Hidasse were used as checks.

**Stem Rust Races Used in Seedling Tests**

Seven stem rust races were obtained from the National Plant Protection Center at Ambo (Table 1). These races were originally identified from samples collected from different wheat-growing regions of Ethiopia. Race designations were based on the letter-code nomenclature system [50; 53], further modified to delineate races in the TTKSK (Ug99) lineage [27]. All isolates were derived from single pustules, multiplied in isolation, and stored at –80°C. To obtain sufficient and fresh inoculum, each isolate was multiplied on the susceptible wheat variety McNair 701 (CItr 15288). The selected races were chosen based on their prevalence and virulence spectra (Table 1).

**Table 1.** Virulence/avirulence spectra of *P. graminis* f. sp. *tritici* races used in seedling tests at Debre Zeit Agricultural Research Center during 2021/22

| Races | Virulence | Avirulence |
|---|---|---|
| TKTTF | 5, 21, 9e, 7b, 6, 8a, 9g, 36, 9b, 30, 17, 9a, 9d, 10, Tmp, 38, McN | 11, 24, 31 |
| TTTTF | *5, 21, 9e, 7b, 11, 6, 8a, 9g, 36, 9b, 30, 17, 9a, 9d, 10, Tmp, 38, McN* | *24, 31,* |
| TTRTF | *5, 21, 9e, 7b, 11, 6, 8a, 9g,36, 9b, 17, 9a, 9d, 10, Tmp, 38, McN* | *30, 24, 31,* |
| TTKTF | *5, 21, 9e, 7b, 11, 6, 8a, 9g,9b, 30, 17, 9a, 9d, 10, Tmp, 38, McN* | *36, 24, 31* |
| TTKTT | *5, 21, 9e, 7b, 11, 6, 8a, 9g, 9b, 30, 17, 9a, 9d, 10, Tmp, 24, 31, 38, McN* | *36* |
| TKKTF | *5, 21, 9e, 7b, 6, 8a, 9g, 9b, 30, 17, 9a, 9d, 10, Tmp, 38, McN* | *11, 36, 24, 31,* |
| TTKSK (UG99) | 5 21 9e 7b, 11 6 8a, 9g, 9b 30 17, 9a, 9d, 10 ,31 38, MCN | 36, Temp, 24 |

**Seedling Test Procedures**

Seedling evaluations were conducted in the greenhouse at Debre Zeit Agricultural Research Center (DZARC) following the standard procedures described by Jin et al. (2007). Five to seven seeds of each bread wheat genotype, along with commercial varieties and the susceptible check, were planted in 3 cm diameter plastic pots containing a soil mixture of soil, compost, and sand in a ratio of 2:1:1. Differential lines were planted alongside the test genotypes. The experiment was arranged in a Completely Randomized Design (CRD) with two replications. Seven-day-old seedlings were inoculated with selected Pgt races when the first leaf was fully expanded. Spores from each isolate were suspended in mineral oil (Soltrol 170) and sprayed onto seedlings using an atomizer. After inoculation, the seedlings were left for a few minutes to allow the oil to evaporate. Plants were then lightly misted with water to create artificial dew, facilitating spore germination. The inoculated pots were covered with plastic bags (145 mm × 235 mm) and secured at the base with rubber bands to prevent cross-contamination [3]. The seedlings were incubated in a dark dew chamber for 18–24 hours at 18°C and approximately 95% relative humidity. Subsequently, they were exposed to fluorescent light for three hours before being transferred to a growth room/greenhouse for disease development. Infection types (ITs) were assessed 14 days after inoculation using the 0–4 scale described by [67]. Infection types of 0, ;, 1, and 2 were considered resistant (low infection types), while 3 and 4 were considered susceptible (high infection types) (Figure 2).

**Postulation of Stem Rust Resistance Genes**

Gene postulation is one of the most rapid and cost-effective methods for identifying seedling resistance genes in wheat. In this study, a classical gene-for-gene approach was used to postulate resistance genes in the tested bread wheat genotypes. Infection type patterns of the test genotypes were compared with those of 20 known stem rust differential lines. The presence of one or more resistance genes was postulated by matching the infection type pattern of each test genotype with that of known differential lines [43]. Seven dominant stem rust races were inoculated on 121 bread wheat genotypes and 20 differential lines. The reaction patterns of the test genotypes were compared with those of the differentials to identify possible Sr genes present in the bread wheat genotypes (Table 2).

**Table 2.** Description of infection types used in classifying stem rust reactions on wheat seedlings [53].

| Host response | IT | Disease symptoms |
|---|---|---|
| Immune | 0 | No uredinia or other macroscopic sign of infection |
| Nearly immune | ; | No uredinia, but hypersensitive necrotic or chlorotic flecks present |
| Very resistant | 1 | Small uredinia surrounded by necrosis |
| Moderately resistant | 2 | Small to medium uredinia often surrounded by chlorosis or necrosis; green island may be surrounded by chlorotic or necrotic border |
| Heterogeneous | X | Random distribution of variable-sized uredinia on single leaf |
| Moderately susceptible | 3 | Medium-sized uredinia that may be associated with chlorosis |
| Susceptible | 4 | Large uredinia without chlorosis |

**Field Testing Procedures and Experimental Design**

The field experiment was conducted using a total of 121 bread wheat genotypes planted at Sinana and Agarfa Research Stations during the 2021/22main cropping season. The experiment was laid out in a simple lattice design with two replications at each location. Spreader rows consisting of a mixture of highly susceptible varieties (Kubsa, Hidasse, and Digalu) were planted perpendicular and adjacent to the test plots to enhance disease development. Each experimental plot measured 0.8 $m^2$ (1 m × 0.8 m) and consisted of four rows spaced 20 cm apart. The spacing between plots and replications was 0.4 m and 1.5 m, respectively. The total experimental area at each location was approximately 485.1 $m^2$ (21 m × 23.1 m). A seed rate of 150 kg $ha^{-1}$ was used. Fertilizers were applied at rates of 100 kg $ha^{-1}$ NPS and 100 kg $ha^{-1}$ urea. Urea was applied in two splits: half at planting and half at booting stage. All other agronomic practices were conducted according to local recommendations.

**Data Collection**

Data were collected on both plot and plant bases. Four randomly selected plants per plot were used for recording plant-based data, and mean values were used for analysis.

**Disease Assessment**

Stem rust severity was assessed six times at seven-day intervals, starting from the first appearance of disease symptoms. Disease severity was recorded using the modified Cobb scale [45].

Host response was categorized as: R = Resistant, MR = Moderately resistant, M = Intermediate, MS = Moderately susceptible, S = Susceptible

| Symbol | Field response | Constant values |
|---|---|---|
| R | Resistant | 0.2 |
| MR | Moderately resistant | 0.4 |
| M | Intermediate | 0.6 |
| MS | Moderately susceptible | 0.8 |
| S | Susceptible | 1.0 |

**Disease Parameters**

**Average Coefficient of Infection (ACI)**

ACI was calculated by multiplying disease severity with a constant value assigned to the host response (Stubbs et al., 1986).

**Area under Disease Progress Curve (AUDPC) and Relative AUDPC (rAUDPC)**

AUDPC was calculated using the formula of [76]:

AUDPC = $\Sigma [(Xi + Xi+1)/2] (ti+1 - ti)$, Where: Xi = disease severity at ith observation, ti = time at ith observation. rAUDPC was calculated by dividing AUDPC by the maximum potential AUDPC.

**Final Rust Severity (FRS) and Disease Progress Rate (r-value)**

FRS was determined from the last disease severity score combined with the host response coefficient. Disease progress rates were estimated using logistic and Gompertz models (Berger, 1981). The best-fitting model was selected based on the coefficient of determination ($R^2$).

**Agronomic Data**

The following agronomic parameters were recorded: Days to 50% heading, Days to 90% maturity, Plant height (cm), Spike length (cm), Number of productive tillers, Number of kernels per spike, Thousand kernel weight (g), Grain yield (g $plot^{-1}$), converted to t $ha^{-1}$

**Data Analysis**

All quantitative data, including ACI, FRS, AUDPC, disease progress rate, rAUDPC, yield, and yield-related traits, were subjected to two-way Analysis of Variance (ANOVA) using R software version 4.1.3. Mean comparisons were performed using Duncan's Multiple Range Below is your

## RESULTS

**Seedling Stage Evaluation at greenhouse**

Seedling evaluations of 121 bread wheat genotypes against seven (7) *Puccinia graminis* f. sp. *tritici* (*Pgt*) races revealed significant variation in infection types (ITs). The susceptible control McNair 701 exhibited compatible reactions (IT 3–4) to all races, confirming successful inoculation. Resistant genotypes, including BW182037, BW182463, and EBW192861, displayed low ITs (0–2), indicating strong seedling resistance. The genotypes were categorized as resistant, susceptible, or heterogeneous based on their IT responses. The majority of genotypes exhibited resistant reactions with infection types of 1 and 2 (Table 4). However, many varieties were susceptible to the TTKTT race, particularly the widely cultivated popular wheat genotypes. Some breeding genotypes showed susceptible reactions with IT scores of 3 and 4 (Table 4). Resistant reactions to races TTTTF, TTKTT, TTKTF, TKTTF, TTRTF, TTKSK (Ug99), and TKKTF were observed in 56, 68, 50, 90, 49, 84, and 121 genotypes, respectively (Table 4). This indicates that incompatible reactions were common among many advanced bread wheat genotypes and some commercial varieties. Race TKKTF showed the highest frequency of avirulent interactions, followed by TTKSK (UG99). In contrast, races TTRTF, TTKTF, TTTTF, and TTKTT exhibited relatively higher frequencies of virulent interactions.

**Table 4: Infection responses of selected bread wheat genotypes to seven Pgt races at seedling stage**

| IT rxns | TTTTF | TTKTT | TTKTF | TKTTF | TTRTF | TTKSK | TKKTF |
|---|---|---|---|---|---|---|---|
| "0"or";" | 8 | 23 | 2 | 1 | 4 | 9 | 0 |
| "1"or"1$^{+}$" or "1$^{-}$" | 28 | 27 | 25 | 51 | 18 | 42 | 75 |
| "2"or"2+"or"2-" | 20 | 18 | 23 | 38 | 27 | 33 | 46 |
| Incompatible interaction | 56 | 68 | 50 | 90 | 49 | 84 | 121 |
| "3" | 65 | 53 | 72 | 33 | 75 | 39 | 3 |
| "4" | 2 | 0 | 1 | 2 | 0 | 2 | 0 |
| Compatible interaction | 67 | 53 | 73 | 35 | 75 | 41 | 3 |
| Heterogeneous(H) | 0 | 0 | 0 | 3 | 0 | 0 | 0 |

** Infection types observed on seedlings at 14 days post-inoculation using a 0 to 4 scale according to Stakman et al. (1962). Infection types of ;, 1, 2, or X are considered low (resistant), whereas ITs of 3 or 4 are considered high (susceptible). Heterogeneous genotypes contained both resistant and susceptible plants.*

**Postulation of Stem Rust Resistance Genes**

Based on ITs and comparison with differential lines, resistant genotypes were postulated to carry genes Sr2, Sr24, and Sr31. Some genotypes combined seedling resistance with adult plant resistance (APR), indicating potential durable resistance. The gene postulation was classified in four groups.

**Group I: Genotypes with Single Seedling Resistance Genes**

Fifteen bread wheat genotypes were postulated to possess specific Sr resistance genes based on comparison of their IT responses with those of 20 near-isogenic lines (NILs). When the IT response of a test genotype matched that of a NIL, the corresponding Sr gene was considered present. Resistance genes Sr11, Sr36, Sr30, and Sr24 were postulated in 7 (5.6%), 2 (1.6%), 1 (0.8%), and 5 (4.1%) genotypes, respectively (Table 5).

**Table 5: Postulated Pgt resistance genes in tested bread wheat genotypes**

| SN | Genotypes | TTTTF | TTKTT | TTKTF | TKTTF | TTRTF | TTKSK | TKKTF | Postulated gene |
|---|---|---|---|---|---|---|---|---|---|
| 18 | BW120053 | 2- | 3+ | ;1+ | ;1 | ;1+ | ;1 | ;1+ | Sr24 |
| 67 | BWRVT(P#7) | 2- | 3- | ;1+ | 3- | 2 | ;1+ | ;1+ | Sr24 |
| 98 | BWRVT(P#11) | ; | 3- | 2 | ;1 | 2+ | ;1+ | ;1 | Sr24 |
| 110 | Wane | ;1 | 3+ | ;1+ | 2- | ;1+ | ;1+ | ;1+ | Sr24 |
| 112 | Shorima | ;1 | 3- | 2+ | ;1+ | 2+ | ; | ;1+ | Sr24 |
| **Sr24** | **LcSr24Ag** | **;1+** | **4** | **2+** | **2+** | **;1+** | **2** | **;1** | |
| 11 | EBW192361 | 3- | 3+ | 3+ | 2+ | 3+ | 3+ | 2- | Sr11+ |
| 12 | EBW192370 | 3+ | 3+ | 3+ | ;1 | 3+ | 2 | 2- | Sr11+ |
| 64 | EBW192005 | 3 | 3- | 3 | ;1 | 3- | 3+ | 2- | Sr11 |
| 40 | EBW192328 | 3+ | 3- | 3+ | ;1 | 3- | 3+ | 2+ | Sr11 |
| 65 | EBW192900 | 3 | 3- | 3+ | 2- | 3+ | 3+ | ;1 | Sr11 |
| 84 | BW182285 | 3 | 3 | 3+ | 2- | 3+ | 3+ | 2+ | Sr11 |
| 114 | M/Walabu | 3+ | 3+ | 3+ | ;1+ | 3 | 3- | ;1+ | Sr11 |
| **Sr11** | **ISr11-Ra** | **3** | **4** | **3+** | **;1+** | **;1+** | **3+** | **3** | |
| 46 | BW182493 | 3+ | 2+ | 3 | 3 | 3+ | 2 | 2- | Sr36 |
| 48 | BW182540 | 3+ | 2+ | 3- | 3- | 3 | ;1+ | ;1+ | Sr36 |
| **Sr36** | **W2691SrTt-1** | **3+** | **2-** | **2** | **2-** | **3-** | **3-** | **;1+** | |
| 37 | EBW192325 | 3+ | 3+ | 3- | 3 | 2 | 3+ | 2+ | Sr30 |
| **Sr30** | **BtSr30Wst** | **3+** | **3** | **3+** | **3+** | **3** | **2** | **3-** | |

**Group II: Genotypes without Known Resistance Genes**

One breeding line (EBW192988) exhibited high infection types to all Pgt races, similar to the universally susceptible check McNair 701. Therefore, this genotype was postulated to contain no known seedling resistance genes. However, this line showed partial resistance in field tests, recording 15MS at Sinana and 10MS at Agarfa.

**Group III: Genotypes Resistant to All Tested Races**

A total of 20 genotypes exhibited resistance to all seven Pgt races at the seedling stage (Table 6). These genotypes showed consistently low ITs. However, additional isolates with different virulence patterns and more NILs are required to accurately postulate the specific resistance genes present in these genotypes.

**Table 6: List of genotypes resistant to all Pgt races at seedling stage**

| SN | BW genotypes | TTKTT | TTKSK | TTTTF | TKTTF | TTRTF | TTKTF | TKKTF |
|---|---|---|---|---|---|---|---|---|
| 5 | BW172831 | ;1 | ; | ;1 | ;1 | 2+ | 2+ | ;1+ |
| 9 | BW172713 | ;1+ | ;1+ | ; | ;1 | ; | 2+ | 2- |
| 17 | BW120042 | ; | ; | ;1 | ;1 | ;1 | ;1 | ;1+ |
| 26 | BW173353 | ;1+ | ;1+ | ;1+ | 2- | 2- | ;1+ | ;1+ |
| 27 | BW182052 | ;1+ | ;1 | ;1 | ;1+ | 2+ | ;1+ | ;1+ |
| 29 | ETBW 9077 | ; | ;1+ | ;1 | ;1 | 2- | ;1+ | ;1+ |
| 43 | BWRVT(P#1) | ; | ;1 | 2- | ;1+ | ;1 | ;1 | ;1+ |
| 44 | BW182463 | 0 | ;1 | ;1+ | ;1+ | ; | ;1 | ;1+ |
| 47 | BWRVT(P#2) | ;1 | ;1 | 0 | ;1 | ;1 | 2+ | ;1+ |
| 82 | BW182037 | 2 | ;1+ | ;1 | 2- | ;1+ | 2+ | ;1+ |
| 87 | BW182767 | ;1 | ;1+ | ;1+ | ;1+ | ;1+ | ;1+ | ;1+ |
| 91 | EBW192861 | ;1 | ; | ;1 | ;1 | ;1+ | ; | ;1 |
| 102 | Obora | ;1 | ;1 | ;1 | ;1 | ;1 | ;1 | ;1+ |
| 108 | ATLAS | ; | 2 | ;1 | ;1 | ;1+ | ;1 | ;1+ |
| 109 | Lemu | ; | ;1+ | ;1 | ;1+ | ;1 | ;1+ | ;1+ |
| 111 | Danbal | ; | ;1+ | ;1 | 2- | ;1+ | 2+ | ;1+ |
| 115 | Mandoyu | ;1 | ;1 | ;1+ | ;1+ | ;1+ | ;1+ | ;1+ |
| 117 | Galan | ; | ;1 | ;1 | ;1+ | 2 | ;1+ | ;1+ |
| 120 | Dure | ; | ;1+ | ;1 | 2- | ;1 | ;1 | ;1+ |
| 121 | Sinja | 2- | ;1 | ;1 | ;1+ | ; | ;1 | 2- |

**Group IV: Genotypes with Unidentified Sr Genes**

Eighty-eight genotypes exhibited differential reactions to the seven races but did not match the IT patterns of any known NILs (Appendix Table 3). These genotypes possessed resistance to at least one race, but their gene identities could not be confirmed (Figure 3).

**Analysis of Variance for Disease and Agronomic Traits**

Highly significant differences ($P < 0.01$) were observed among genotypes for days to heading, days to maturity, spike length, number of kernels per spike, thousand kernel weights, and grain yield across locations (Table 8). This indicates the presence of substantial genetic variability among the tested genotypes. Location effects were highly significant for most traits, indicating environmental influence on disease development and yield performance. Genotype × environment interaction was also highly significant ($P < 0.01$) for major traits including FRS, AUDPC, rAUDPC, and grain yield (Table 7). This demonstrates that genotypes responded differently under varying environmental conditions, emphasizing the importance of multi-location testing.

Table 7: Analysis of variance of wheat stem rust disease parameters on the bread wheat genotypes at Sinana and Agarfa, during 2021/22main cropping season.

| Characters | Source of Variation | | | | | |
|---|---|---|---|---|---|---|
| | REP | Block (REP) | Genotypes | Error | Mean | CV |
| **Sinana** | | | | | | |
| FRS | 0.25$^{Ns}$ | 0.122 $^{Ns}$ | <2e-16 *** | 4.6 | 26.47 | 8.11 |
| ACI | 0.026 * | 0.0003 *** | < 2e-16*** | 2.15 | 14.55 | 10.07 |
| AUDPC | 0.013* | 0.0002*** | < 2e-16*** | 3346 | 515.42 | 11.22 |
| rAUDPC | 0.013* | 0.0002 *** | < 2e-16*** | 12.1 | 31.00 | 11.22 |
| **Agarfa** | | | | | | |
| FRS | 0.774 $^{Ns}$ | 0.74$^{Ns}$ | <2e-16*** | 7.15 | 17.55 | 15.29 |
| ACI | 0.539 $^{Ns}$ | 0.03* | <2e-16*** | 0.29 | 28.73 | 8.52 |
| AUDPC | 0.5704 $^{Ns}$ | 0.01* | <2e-16*** | 369 | 209.09 | 9.19 |
| rAUDPC | 0.5704 $^{Ns}$ | 0.01* | <2e-16*** | 4.7 | 23.61 | 9.19 |

FRS (Final rust severity), AUDPC (Area under disease progressive curve), rAUDPC (Relative Area under Disease progress curve), CV (Coefficient of Variation), * significant, ** = moderately resistant, *** highly significant, Ns= non-significant

Table 8: Combining Analysis of yield and yield related traits on bread wheat genotypes at Sinana and Agarfa, 2021/22, during main cropping season.

| Characters [a] | Source of Variation [b] | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | REP (DF=1) | Block (REP) (DF=84) | Genotypes (120) | Location (DF=1) | Genotype Loc (DF=120) | Error (DF= 199) | Mean[c] | CV [d] |
| DH | 0.07 $^{ns}$ | 0.074$^{ns}$ | 0.00034 *** | < 2e-16 *** | 0.16 $^{ns}$ | 1.83 | 71.73 | 1.89 |
| PH | 0.005** | 0.04 * | < 2e-16 *** | 0.26ns | 3.26e-14 *** | 15.22 | 84.57 | 4.61 |
| DM | 3.16e-13*** | 0.08 $^{ns}$ | 3.16e-13*** | 0.004** | 0.0004*** | 1.83 | 120.53 | 1.12 |
| SPS | 0.68 $^{ns}$ | 0.14$^{ns}$ | <2e-16*** | 0.083 $^{ns}$ | 0.9957 $^{ns}$ | 1.44 | 17.34 | 6.92 |
| SPL | 0.007 ** | 0.11$^{ns}$ | 0.28$^{ns}$ | 0.0003*** | 0.46$^{ns}$ | 0.47 | 8.19 | 8.38 |
| NKPS | 0.012* | 0.02* | 5.02e-07 *** | < 2e-16 *** | 1.88e-05 *** | 26.60 | 42.30 | 12.19 |
| NPT | 0.61$^{ns}$ | 0.17$^{ns}$ | 0.214$^{ns}$ | 0.268 $^{ns}$ | 0.536 $^{ns}$ | 0.38 | 23.08 | 2.66 |
| TKW | 0.0006*** | 0.24$^{ns}$ | < 2e-16 *** | 0.124 $^{ns}$ | 0.0003*** | 9.30 | 28.94 | 10.54 |
| GYD | 0.008*** | 0.62$^{ns}$ | < 2e-16 *** | 4.66e-13 *** | < 2e-16 *** | 0.09 | 3.45 | 8.70 |

DH = days to heading, DM = days to maturity, PH = Plant height, NPT = Number of productive tiller, NSPS = Number of spiklets per spike, SPL = Spike length, NSPS = Number of seed per spike, GYD = Grain yield, CV = Coefficients variation, * significant, ** = moderately resistant, *** highly significant, ns= non-significant

**Field Assessment of Slow Rusting Resistance**

**Final Rust Severity (FRS)**

Considerable variation in final rust severity was observed among genotypes at both locations, ranging from trace (TR) to 70S reactions. At Sinana, 53% of advanced genotypes showed FRS equal to or lower than the resistant check (Dure), while 47% showed higher FRS. Eight genotypes and two commercial varieties showed higher severity than the susceptible check Digalu. At Agarfa, 60% of genotypes exhibited FRS equal to or lower than Dure, while 40% showed higher severity. Therefore, many genotypes in this study possessed useful levels of adult plant resistance.

**Coefficient of Infection (CI)**

At Sinana, 78% of advanced genotypes and 66.6% of commercial varieties exhibited high levels of adult plant resistance (CI = 0–20). Similar trends were observed at Agarfa, where most varieties recorded low CI values.

**Area Under Disease Progressive Curve (AUDPC) and Relative AUDPC**

AUDPC proved to be a reliable indicator of slow rusting resistance. At Sinana, AUDPC values ranged from 29.1 to 1583.8, while at Agarfa they ranged from 6 to 885. Genotypes with AUDPC < 300 were classified as having high partial resistance. Thirty-one genotypes at Sinana and 98 genotypes at Agarfa fell into this category. Relative AUDPC (rAUDPC) values further supported these classifications, with many genotypes showing values below 30%, indicating good slow rusting resistance.

**Disease Progress Rate**

Disease progress rate were used to compare disease progresses among the bread wheat advanced genotypes and commercial varieties. Accordingly, Logistic and Gompertz models of the temporal analysis model were used for this experiment. Higher coefficient of determination (R2) and lower standard error (SE) were identified on the Gompertz model than logistic model. Data had better fitted into the gompertz model than logistic model. Therefore, the disease progress rate was estimated and compared to using a Gompertz model. Infection rate (Inf-rate) is a function of per unit days. In this experiment, highest disease progress rate were observed on the BW172093 (r=0.13) of bread wheat genotypes followed by EBW192375 (r=0.11) variety and genotypes Galan (r=0.0.08) which indicated that wheat stem rust could increase 0.13, 0.11 and 0.008 per 7 days, respectively at Sinana experimental site during 2021/22 cropping season. However, the lowest Disease progress rate (0.001) has been observed on the BW172713 genotypes which indicated that wheat stem rust severity were increased 0.001 units per days which presented in the Appendix Table 7.

At Agarfa site the same way of analysis was followed that of the Sinana test site. At this location, the highest disease progress rate was calculated for the variety Hidasse (0.18), followed by bread wheat line EBW192380 (0.14) which indicated that wheat stem rust could increase 0.18 and 0.14 per days, respectively at Agarfa experimental site during 2021/22cropping season. However, From Gompertz Model the lowest disease progress rate (0.0007) was observed on the BW172713 genotypes which indicated that wheat stem rust severity were increased 0.0007 per days which presented in the Appendix Table 8.

**Identification of True Slow-Rusting Genotypes**

At sinana tested site the tested wheat genotypes were categorized into two distinct groups for wheat stem rust slow- rusting resistance, based on the AUDPC values. Wheat genotypes exhibited AUDPC values less than 300 ranged from 29.10 to 281.4 showed high level of partial resistance or slow-rusting resistance to wheat stem rust, consisted of thirty one (31) bread wheat genotypes (Appendix Table 5). However, in this tested location high AUDPC values more than 300 ranged from 315.00 to 1583.8 which indicated low slow-rusting or low partial resistance, presented in the Appendix Table 5. At Agarfa tested site, on the basis of area under disease progressive curve, 98 bread genotypes and commercial varieties showed high partial resistance or slow-rusting of wheat stem rust which showed less than 300 AUDPC value ranged from 6 to 294 (Appendix Table 5). In contrary, 23 bread wheat genotypes showed low slow rusting resistance or High AUDPC values which ranged from 303 to 885 which indicated the bread genotypes were affected by wheat stem rust; it is presented in Appendix Table 5. In this tested location wheat yellow rust severity were confounded and cause that the severity of wheat stem rust were reduced.

High partial resistance and moderate partial resistance of bread genotypes have been identified based on slow-rusting parameters. Among the tested genotypes 15 namely; 45, 48, 64, 65, 67, 69, 70, 78, 84, 86, 98, 99, 103, 105, and 107 were identified to be presented high partial resistance to wheat stem rust (Table 9). But, genotype 107 showed high AUDPC value which indicated it might group in moderate partial resistance. Those bread wheat genotypes were screened based the performance of high seedling infection types, low AUDPC, less 30% of rAUDPC, CI, and final rust severities. This indicated that bread wheat genotypes which showed high infection types and low disease parameters showed high partial resistance. In the same ways, Moderate partial resistance have been identified on 13 bread wheat genotypes namely; 2, 7, 28, 38, 40, 54, 64, 68, 88, 95, 97, 101, and 110 bread wheat genotypes. The in both Sinana and agarfa tested site, 31 bread wheat genotypes and commercial varieties were identified based of Area under disease progressive curve, and the intermediate to susceptible reactions at seedling stage for TTKTT which were identified while the experiment conducted in 2021 of main cropping season. Because, the TTKTT race dominantly identified during survey of 2021 of main cropping season (source: Ambo wheat stem rust race analysis report).

Generally, in this present study, 14 advanced bread wheat genotypes showed true slow rusting based on infections types (between 2+ and 3+) and field assessment which showed less than 300 AUDPC values (Table 9). In this study, high level and frequency of resistance were detected in advanced genotypes than in commercial varieties. Hence, these advanced breeding genotypes could be exploited for improvement of wheat stem rust resistance in common wheat.

Table 9: The identified high partial resistance of wheat stem rust on 15 the bread wheat genotypes during 2021/22main cropping season.

| | | Sinana | | | | | | Agarfa | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Geno | TTKTT | FRS | CI | ACI | AUDPC | rAUDPC | r-value | FRS | CI | ACI | AUDPC | rAUDPC | r-value |
| 45 | 2+ | 5MS | 4 | 3 | 91.4 | 5.5 | 0.02 | 5ms | 4 | 1 | 38 | 4 | 0.01 |
| 48 | 2+ | 10MSS | 8 | 5 | 165.2 | 9.9 | 0.02 | 5m | 3 | 2 | 51 | 6 | 0.01 |
| 64 | 3- | 30S | 12 | 10 | 414.6 | 24.9 | 0.03 | 25ms | 20 | 7 | 222 | 25 | 0.03 |
| 65 | 3- | 5MS | 1 | 1 | 40.3 | 2.9 | 0.01 | 5m | 3 | 2 | 56 | 6 | 0.01 |
| 67 | 3- | 10ms | 3 | 3 | 82.3 | 4.9 | 0.02 | 10m | 6 | 3 | 89 | 10 | 0.02 |
| 69 | 3 | 5S | 3 | 3 | 95.7 | 5.8 | 0.02 | 5ms | 4 | 2 | 44 | 5 | 0.01 |
| 70 | 3+ | 15MSS | 8 | 7 | 268.6 | 16.2 | 0.09 | 15ms | 12 | 3 | 270 | 15 | 0.02 |
| 78 | 3 | 15MS | 7 | 7 | 241.5 | 14.5 | 0.01 | 15ms | 12 | 6 | 228 | 26 | 0.02 |
| 84 | 3 | 5MS | 4 | 1 | 43.4 | 2.6 | 0.01 | 5s | 5 | 2 | 48 | 5 | 0.01 |
| 86 | 33+ | 20S | 20 | 7 | 248.5 | 14.9 | 0.03 | 20mss | 18 | 7 | 218 | 25 | 0.03 |
| 98 | 3- | 10S | 10 | 3 | 124.6 | 7.5 | 0.02 | 10ms | 8 | 2 | 67 | 8 | 0.01 |
| 99 | 3+ | 15S | 15 | 8 | 262 | 15.8 | 0.03 | 25m | 15 | 5 | 177 | 20 | 0.03 |
| 103 | 3+ | 20ms | 16 | 8 | 272.3 | 16.4 | 0.03 | 10m | 6 | 2 | 55 | 6 | 0.04 |
| 105 | 2+ | 15ms | 12 | 6 | 206.5 | 12.4 | 0.03 | 15ms | 12 | 6 | 194 | 22 | 0.03 |
| 107 | 3+ | 25s | 25 | 11 | 434 | 26.1 | 0.03 | 20mss | 18 | 8 | 249 | 28 | 0.03 |

AUDPC (Area under disease progressive curve), rAUDPC (Relative Area under Disease progressive curve), R-value (infection rate)

Table 10: The identified Moderate partial resistance of wheat stem rust on 13 of bread wheat genotypes during 2021/22 main cropping season.

| Geno | TTKTT | FRS | CI | ACI | AUDPC | rAUDPC | r-value |
|---|---|---|---|---|---|---|---|
| 2 | 3- | 30s | 30 | 17 | 605.5 | 36.4 | 0.02 |
| 7 | 3 | 30s | 30 | 15 | 495.3 | 29.8 | 0.03 |
| 28 | 3 | 30S | 30 | 18 | 627.9 | 37.8 | 0.04 |
| 38 | 2+ | 30S | 30 | 15 | 514.5 | 30.9 | 0.04 |
| 40 | 3- | 30S | 30 | 16 | 548.5 | 33 | 0.05 |
| 54 | 2+ | 30S | 30 | 16 | 568.4 | 34.2 | 0.04 |
| 64 | 3- | 30S | 12 | 10 | 414.6 | 24.9 | 0.03 |
| 68 | 3- | 30MSS | 17 | 16 | 617.8 | 37.2 | 0.03 |
| 88 | 3+ | 30S | 30 | 20 | 645.8 | 38.8 | 0.03 |
| 95 | 3 | 30S | 30 | 20 | 715.8 | 43.1 | 0.07 |
| 97 | 3 | 30MS | 24 | 18 | 561.8 | 33.8 | 0.04 |
| 101 | 3 | 30s | 30 | 16 | 537.1 | 32.3 | 0.04 |
| 110 | 3+ | 30mss | 27 | 16 | 557.9 | 33.6 | 0.04 |

Geno= genotypes, AUDPC=Area under disease progressive curve), rAUDPC=relative Area under Disease progressive curve)

Table 11: Bread wheat genotypes showed less than 30% rAUDPC across locations in the 2021/22 main cropping season

**Across locations of rAUDPC**

| GM | rAUDPC | GM | rAUDPC | GM | rAUDPC | GM | rAUDPC | GM | rAUDPC | GM | rAUDPC |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 80 | 30.03$^{j\text{-}E}$ | 101 | 25.10n-J | 53 | 22.97s-M | 43 | 18.72 y-R | 44 | 15.44 D-S | 69 | 5.39 N-S |
| 60 | 29.72 j-F | 57 | 25.04n-J | 76 | 22.58s-M | 4 | 18.28 z-S | 42 | 13.46E-S | 17 | 5.29 N-S |
| 3 | 29.56 j-G | 64 | 25.01n-J | 49 | 21.52t-N | 121 | 18.12 z-S | 77 | 12.99 F-S | 45 | 4.90 N-S |
| 34 | 28.98 k-G | 32 | 24.56o-K | 108 | 21.34 t-O | 99 | 17.89 z-S | 50 | 12.78G-S | | 63 4.77O-S |
| 68 | 28.34 k-G | 6 | 24.30 p-L | 66 | 21.15t-O | 73 | 17.43z-S | 103 | 11.27 H-S | 83 | 4.77 O-S |
| 20 | 28.25 k-G | 58 | 24.24p-L | 38 | 20.77 u-P | 8 | 17.38 z-S | 47 | 10.85 I-S | 65 | 4.37 P-S |
| 74 | 28.18 k-G | 51 | 24.11p-L | 31 | 20.38v-Q | 105 | 17.16 z-S | 62 | 10.54 I-S | 84 | 4.013Q-S |
| 21 | 27.93 k-H | 27 | 23.88p-L | 75 | 20.11 v-Q | 72 | 16.98 A-S | 35 | 9.25 J-S | 9 | 2.48 RS |
| 85 | 27.85 k-H | 39 | 23.792p-L | 78 | 20.11 v-Q | 30 | 16.48 A-S | 29 | 9.18 J-S | 100 | 2.45 RS |
| 28 | 27.24 l-I | 40 | 23.52 q-L | 52 | 19.85 v-Q | 104 | 16.32B-S | 46 | 9.00 J-S | 79 | 1.85 S |
| 107 | 27.08 m-I | 26 | 23.48q-L | 86 | 19.79w-Q | 71 | 16.30 B-S | 48 | 7.85 K-S | | |
| 37 | 26.11m-I | 1 | 23.08r-M | 7 | 19.57 x-Q | 87 | 15.69C-S | 98 | 7.513 L-S | | |
| 82 | 26.11 m-I | 61 | 23.03$^{s\text{-}M}$ | 5 | 19.28x-Q | 70 | 15.69 C-S | 41 | 6.41 M-S | | |

GM= Genetic materials or genotypes, rAUDPC = Relative Area under Disease Progressive Curve.

**Performance of Bread wheat genotypes for Agronomic traits**

Analysis of variance of individual locations were analysed for mean values, range**,** and standard error of yield and yield traits (Table 11 and Table 12).

**Days to Heading and Days to Maturity of Bread Wheat Genotypes**

At Sinana, genotype BW120063 and BW172440 were early to heading which took them 69 days, while BW184055 and variety Digalu were late in heading took 76 days. Early maturity genotypes observed for BWRVT(P#10) (118 days), followed by genotypes BW184123 and variety Boru took them 119 days to mature, While late mature BW182714 and BW172474 took the 123 days (Table 14). At Agarfa, among the genotypes BW172474 took 65 days followed by BW182714 and BWRVT(P#11) were early to heading took 67 days, whereas variety Digalu were late heading in 77 days. Digalu variety showed late days to heading at the two test locations (Table 11). Genotype/varieties Obora, EBW192880, EBW192867, EBW192856, EBW192031, BW184183, BW184123, Wane and (118 days) were early to matured genotypes, while BW172474 were showed late matured genotypes took 127 days (Table 14

Grain yield of bread wheat genotypes

At both test locations highly significant difference were recorded among the genotypes for plant height, which ranged from 67.8 to 100.25 cm with the mean value of 84.33 cm at Sinana and 67.85 to 102.50 cm with the mean value of 84.77 cm at Agarfa. At Sinana, shortest plant height were observed in variety Mandoyu (65.65 cm), while variety Sanete (100.625cm) were the tallest followed by EBW192005 (95.37 cm), and EBW192928 (94.38 cm). At Agarfa, the shortest genotype were EBW192856 (67.85 cm) followed by BWRVT (P#3) (68 cm), while the tallest were Variety Sanete (102.50 cm) followed by Balcha (101.50 cm) and Shorima (99.5 cm) (Table 11). Presence of highly significant difference in plant height of studied bread wheat advanced genotypes showed the existence of variability indicates selection for plant height could be effective.

At Sinana test site highly significant variability among genotypes for grain yield t ha$^{-1}$ observed, which ranged from 0.31 to 6.36 t ha$^{-1}$ with the mean value of 3.34 t ha$^{-1}$. From this result, BW182463 (6.36t ha$^{-1}$) gave the highest overall grain yield, followed by ETBW 9077 (6.124 t ha$^{-1}$) and ETBW 9548 (5.97 t ha$^{-1}$) presented in table 11. On the other hand genotype Sanete (0.30875) gave the lowest (0.308 t ha$^{-1}$) grain yield followed by genotype Hidasse (0.89250 t ha$^{-1}$) and genotype Digalu (0.97 t ha$^{-1}$), presented in the Table 12. Reason for low yielder on sanete was highest rust (Yellow rust and Stem rust) pressure were occurred on these commercial variety. At Agarfa, the highest grain yield 7.36 t ha$^{-1}$ were recorded for ETBW 9077 followed by BW182463 (7.30 t ha$^{-1}$) BW182491 (7.28 t ha$^{-1}$), ETBW 9077 (7.03 ha$^{-1}$) , ETBW 9548 (7.01), while the lowest yield of 0.65 t ha$^{-1}$ obtained from Sanete variety followed by EBW192319 (0.87 t ha$^{-1}$), kakaba (0.87 t ha$^{-1}$), Hidasse (1.02 t ha$^{-1}$), Digalu (1.03 t ha$^{-1}$) has obtained which presented in the table 12. This result indicated that most of popular commercial variety has produced the lowest grain yield due to the effects of wheat stem rust and wheat yellow rust. Among the test genotypes about 51 genotypes had gave 3.6 to 6.36 t ha$^{-1}$ and about 53 genotypes had gave above maximum national average from 3.6 to 7.3 t ha$^{-1}$ and 3.60 to 6.36 t ha$^{-1}$ and having a good resistant/tolerance against wheat stem rust and wheat yellow rust in compared to the commercial variety at Sinana and Agarfa test locations, respectively (Table 11). This indicated that there will be the profitable genotypes to be released.

Thousand Kernel Weight

At sinana test site, 10.8 to 46.4 TKW were recorded from the test bread wheat. At Agarfa, 15.00 to 47.6 TKW were recorded from the same test bread wheat of sinana (Table 12). Across test locations Sanete variety has significantly reduced the kernel weight due to the effects of wheat stem rust and wheat yellow

rust confound which were worth noting that stem rust caused kernel weight reduction in the genotypes. This indicated a grain yield and TKW has strong relationship or they had highly correlated to each other.

Generally, the highest grain yield over national average were obtained from the tested bread wheat genotypes, 51 and 53 bread genotypes at Sinana and Agarfa tested site in 2021/2022 main cropping season, It was presented in (Table 12). This indicated there would be the best genotypes to exploit in breeding program and helps for variety releasing.

Table 11: Mean variance, standard error, range, of mean of disease parameters, yield and yield related traits at Sinana, in 2021/22

| Character | Mean ± SE | Range | |
|---|---|---|---|
| | | Min. | Max. |
| DH | 72.74±1.34 | 68 | 78.00 |
| DM | 120.7±1.01 | 118.0 | 127.0 |
| SPL | 8.07 ±0.50 | 6.500 | 9.75 |
| SPS | 17.43± 0.89 | 13.00 | 22.00 |
| NPT | 2.627 ± 0.44 | 1.000 | 5.00 |
| NSPS | 39.46 ± 7.41 | 26.00 | 167.25 |
| PH | 84.33 ± 2.85 | 65.00 | 105.00 |
| Tkw | 29.15 ± 1.84 | 10.80 | 46.40 |
| GYD | 3.34 ± 0.13 | 0.31 | 6.36 |
| ACI | 14.5 ± 1.2176 | 1.00 | 46.000 |
| AUDPC | 515.42 ± 47.06 | 28.35 | 1662.50 |
| rAUDPC | 31 .00 ± 2.83 | 1.7 | 100.00 |
| YR | 37.55± 3.75 | 0.80 | 70.00 |

DH= days to heading, DM= Days maturity, SPL=Spikr length, SPS= spike per spike, NPT= number productive tillers, NSPS= number seed per spike, PH= plant height, TKW=Thousand kernel weight, GYD= grain yield, ACI= Average coefficients of infection, AUDPC= Area under disease progressive curve, rAUDPC= relative area under disease progressive curve

Table 12: Mean variance, standard error, range of disease parameters, yield and yield related traits at Agarfa, in 2021/22

| Character | Mean ± SE | Range | |
|---|---|---|---|
| | | Min. | Max. |
| DH | 70.74 ± 0.42 | 64.00 | 77.00 |
| DM | 120.7 ±0.933 | 118.0 | 128.0 |
| SPL | 8.307 ±0.47 | 6.000 | 10.00 |
| SPS | 17.43 ±0.84 | 13.00 | 22.00 |
| NPT | 2.627 ±0.42 | 1.00 | 4.40 |
| NSPS | 45.66 ±3.28 | 26.00 | 61.00 |
| PH | 84.77 ±2.77 | 67.85 | 102.50 |
| TKW | 29.72 ±2.41 | 15.00 | 47.60 |
| GYD | 3.558 ± 0.26 | 0.65 | 7.36 |
| ACI | 6.32 ±0.40 | 0.20 | 27.00 |
| AUDPC | 209.09 ± 14.74 | 5.600 | 885.50 |
| rAUDPC | 0.63 ±1.66 | 0.63 | 100.00 |
| YR | 48.23 ±3.4 | 3.00 | 70.00 |

DH= days to heading, DM= Days maturity, SPL=Spikr length, SPS= spike per spike, NPT= number productive tillers, NSPS= number seed per spike, PH= plant height, TKW=Thousand kernel weight, GYD= grain yield, ACI= Average coefficients of infection, AUDPC= Area under disease progressive curve, rAUDPC= relative area under disease progressive curve

Table 13: Bread wheat genotypes and commercial genotypes with the highest grain yield (> national average 3.5 t ha-1) at Sinana and Agarfa test site in 2024/25in the main cropping season

| GM | **Sinana** | **Sinana** | | **Agarfa** | | | |
|---|---|---|---|---|---|---|---|
| | GYD | GM | | GM | GYD | GM | GYD |
| 44 | 6.36 $^{a}$ | 31 | 4.21$^{a-o}$ | 21 | 7.36$^{a}$ | **81** | 4.73$^{e-k}$ |
| 29 | 6.12$^{ab}$ | 23 | 4.19 $^{a-o}$ | 44 | 7.30 $^{a}$ | 7 | 4.54$^{f-l}$ |
| 100 | 5.97 $^{a-c}$ | 28 | 4.17 $^{a-o}$ | 45 | 7.29$^{a}$ | 71 | 4.45$^{f-m}$ |
| 26 | 5.62$^{a-d}$ | 83 | 4.15$^{a-o}$ | 29 | 7.03 $^{a}$ | 20 | 4.35$^{g-n}$ |
| 5 | 5.44$^{ae}$ | 87 | 4.11$^{a-o}$ | 100 | 7.01 $^{a}$ | 4 | 4.32$^{g-o}$ |
| 21 | 5.23$^{af}$ | 35 | 4.10$^{a-o}$ | 42 | 6.85 $^{a}$ | 77 | 4.320$^{g-o}$ |
| 43 | 5.23$^{a-f}$ | 48 | 4.08$^{a-o}$ | 27 | 6.84 $^{a}$ | 62 | 4.29$^{g-p}$ |
| 4 | 5.18$^{ag}$ | 7 | 4.08$^{a-o}$ | 58 | 6.00 $^{b}$ | 52 | 4.27$^{g-q}$ |
| 8 | 5.16$^{ah}$ | 34 | 4.06$^{a-o}$ | 5 | 5.88 $^{bc}$ | 40 | 4.26$^{g-q}$ |
| 27 | 5.07$^{ai}$ | 71 | 4.05 $^{a-o}$ | 76 | 5.67$^{b-d}$ | 61 | 4.22$^{h-q}$ |
| 14 | 4.82$^{aj}$ | 39 | 4.03 $^{a-o}$ | 26 | 5.54$^{b-e}$ | 2 | 4.16 $^{i-r}$ |
| 62 | 4.73$^{ak}$ | 58 | 4.013 $^{a-o}$ | 87 | 5.49$^{b-e}$ | 31 | 4.15$^{i-r}$ |
| 42 | 4.73$^{ak}$ | 106 | 4.00$^{a-p}$ | 10 | 5.48$^{b-e}$ | 68 | 4.11$^{i-r}$ |
| 10 | 4.59$^{al}$ | 12 | 3.98$^{a-p}$ | 30 | 5.47$^{b-e}$ | 35 | 4.08$^{i-s}$ |
| 59 | 4.54$^{am}$ | 90 | 3.96$^{a-p}$ | 14 | 5.46$^{b-e}$ | 59 | 4.04$^{i-t}$ |
| 50 | 4.44$^{am}$ | 93 | 3.83$^{a-p}$ | 16 | 5.27$^{b-f}$ | 25 | 3.92 $^{j-u}$ |
| 9 | 4.37$^{an}$ | 109 | 3.83$^{a-p}$ | 13 | 5.16$^{b-g}$ | 18 | 3.88$^{k-u}$ |
| 104 | 4.28$^{ao}$ | 40 | 3.80$^{a-p}$ | 6 | 5.13$^{bh}$ | 22 | 3.88$^{k-u}$ |
| 16 | 4.28$^{ao}$ | 60 | 3.77$^{a-p}$ | 39 | 5.09$^{ch}$ | 95 | 3.84$^{k-v}$ |
| 86 | 4.28$^{ao}$ | 81 | 3.76$^{a-p}$ | 8 | 4.89$^{d-i}$ | 9 | 3.78$^{l-w}$ |
| 41 | 4.27$^{ao}$ | 22 | 3.73$^{a-p}$ | 106 | 4.87$^{d-i}$ | 69 | 3.75$^{l-w}$ |
| 84 | 4.27$^{ao}$ | 3 | 3.69$^{bq}$ | 41 | 4.88$^{d-i}$ | 83 | 3.71$^{l-x}$ |
| 45 | 4.26 $^{ao}$ | 49 | 3.68$^{b-q}$ | 46 | 4.87$^{d-i}$ | 60 | 3.71$^{l-x}$ |
| 15 | 4.23 $^{ao}$ | 30 | 3.60$^{b-r}$ | 50 | 4.86$^{d-i}$ | 15 | 3.67$^{l-y}$ |
| 69 | 4.22 $^{ao}$ | | | 24 | 4.81$^{d-j}$ | 8 | 3.67l-y |
| 76 | 4.15 ao | | | 32 | 4.81$^{d-j}$ | 53 | 3.65$^{l-x}$ |
| 77 | 4.22$^{a-o}$ | | | 12 | 4.79$^{d-j}$ | | |

GM= Genotype material, GYD=grain yield

**Correlation between slow rusting parameters of wheat stem rust**

Pearson's correlation analysis showed highly significant positive correlation ($p<0.001$) between the slow rusting of wheat stem rust parameters (Table 15). In the Table 15, A positive and highly significant correlation coefficients of CI with final rust severity (r=0.96), Area under disease progressive curve (r=0.99) and relative area under disease progressive curve (r=0.99) was observed at sinana in the 2024/25main cropping season. In the same season, at Agarfa positive and highly significant correlation coefficients of CI with final rust severity (r=0.91), Area under disease progressive curve (r=0.89) and relative area under disease progressive curve (r=0.89) was observed.

At both test locations the disease progress rate showed highly significant with coefficient of infection (r=0.64), Area under disease progressive curve (r=0.54), relative area under disease progressive curve (r=0.57) has observed. As compared to the slow rusting parameters of wheat stem rust correlations to themselves, disease progress rate had shown moderate correlation with slow rusting parameters at both test locations (Table 14). This showed that despite severity or the AUDPC were increasing, the rate of infection slowed down over time.

At Sinana Experimental Site, weak (CI-1 only) to strong correlation was observed in between CI-6 with CI-1 (r= -0.02), CI-2 (r=0.61), CI-3 (r= 0.82), CI-4 (r=0.90), and CI-5 (r=0.91), while highly significant (p<0.001) different has been observed in this wheat stem rust parameters in Table 18. CI-1 and CI-6 showed Negative correlation among each other's. CI (1-4) had shown positive correlation. CI (3-5) showed strong correlation with CI-6 despite CI 2 and CI 6 showed moderate correlation. However, non-significant different had shown in between CI-4 with CI-1-3. Likewise, yellow rust was tested to identify its confounding effect with wheat stem rust. Hence, Pearson's correlation coefficients showed non-significant and weak correlation were detected among yellow rust and wheat stem rust parameters. This indicated that, when yellow rust is increased wheat stem rust infection would slow down.

In similarly at Agarfa experimental site, moderately to high significant difference and weak to strong correlation was observed in between CI-6 with CI-1(0.18), CI-3(0.46), CI-4(0.72), CI-5 (r=0.94) (Table 16). However, non-significant difference has been detected in between CI-6 with CI-2 and yellow rust. This is similar with the above discussion with Sinana location.

**Correlation between slow rusting parameters of wheat stems rust and agronomic traits**

Pearson's correlations between slow rusting parameters of wheat stems rust and agronomic parameters like days to heading, plant height, days to maturity, spike length, seed per spike, number of seed per spike and number of productive tillers showed non-significant and weak correlations at sinana in the 2021 main cropping season. However, significant correlations had been detected between the slow rusting parameters average coefficients of infection (ACI), final rust severity (FRS), area under disease progressive curve (AUDPC) and agronomic traits like grain yield and thousand kernel weights. At sinana, the negative correlation has been detected between days to maturity, spike length, number of productive tillers, and number of seed per spike, thousand kernel weights and grain yield with the slow rusting parameters like CI, AUDPC, and rAUDPC (Table 16). Similarly, negative correlation has detected between FRS, and SPL, TKW, GYD and YR (table 16). Positive correlation was observed between FRS with days to heading (DH), plant height (PH), days to maturity (DM), spike length (SPL), seeds per spike (SPS), number of productive tiller (NPT), and NSPS. Also, positive correlations were observed between CI with DH and DM (Table 16).

At Agarfa test site the same correlation coefficients were observed except ACI with DM which showed the significant association and DH with ACI, AUDPC, and rAUDPC showed significant difference to each other's (Table 16). In this test site, negative and weak correlation coeficients had been observed between slow rusting parameters with PH, SPL, NPT, TKW, GYD and YR (Table 19). In the opposite of this result, positive and weak correlation was observed among slow rusting parameters with DH, SPS, and NSPS, reference (Table 16).

Table 14 Correlation coefficients (r) of Disease progress rate with of Wheat stem rust parameters in 2021/2022 main cropping season

| Parameters | AUDPC | rAUDPC | CI | DPR |
|---|---|---|---|---|
| AUDPC | - | | | |
| rAUDPC | 1*** | - | | |
| CI | 0.94*** | 0.89*** | - | |
| DPR | 0.54*** | 0.57*** | 0.64*** | - |

AUDPC= Area under Disease progressive curve, rAUDPC = Relative Area under Progressive Curve, CI= Coefficients of Infection, DPR= Disease Progress rate.

Table 151: Pearson's correlation between wheat stem rust parameters at Sinana (Below diagram) and Agarfa (Above diagram)

| Character | YCI | CI-I | CI-II | CI-III | CI-IV | CI-V | CI-VI | FRS | ACI | AUDPC | SrAUDPC |
|---|---|---|---|---|---|---|---|---|---|---|---|
| YCI | | -0.03ns | 0.04ns | 0.03ns | -0.09ns | -0.15ns | -0.13ns | -0.18* | -0.11ns | -0.11ns | -0.11ns |
| CI-I | -0.07$^{ns}$ | | 0.72*** | 0.43*** | 0.26*** | 0.23** | 0.18** | 0.19** | 0.35*** | 0.36*** | 0.36*** |
| CI-II | 0.62 $^{ns}$ | 0.00*** | | 0.60*** | 0.33*** | 0.17ns | 0.10ns | 0.10ns | 0.33*** | 0.37*** | 0.37*** |
| CI-III | 0.38ns | -0.01*** | 0.79*** | | 0.70*** | 0.48*** | 0.46*** | 0.44*** | 0.66*** | 0.69*** | 0.69*** |
| CI-IV | 0.31* | 0.01ns | 0.64ns | 0.88ns | | 0.75*** | 0.72*** | 0.70*** | 0.86*** | 0.88*** | 0.88*** |
| CI-V | 0.30ns | -0.07*** | 0.54*** | 0.79*** | 0.91*** | | 0.94*** | 0.91*** | 0.95*** | 0.95*** | 0.95*** |
| CI-VI | 0.28ns | -0.02*** | 0.61*** | 0.82*** | 0.90*** | 0.93*** | | 0.97*** | 0.94*** | 0.92*** | 0.92*** |
| FRS | -0.04ns | 0.28*** | 0.60*** | 0.82*** | 0.90*** | 0.94*** | 0.99*** | | 0.91*** | 0.89*** | 0.89*** |
| ACI | 0.40ns | -0.02*** | 0.73*** | 0.92*** | 0.96*** | 0.95*** | 0.96*** | 0.96*** | | 1.00*** | 0.99*** |
| AUDPC | 0.39ns | -0.03*** | 0.74*** | 0.93*** | 0.97*** | 0.95*** | 0.95*** | 0.95*** | 0.99*** | | 1.00*** |
| SrAUDPC | 0.39ns | -0.03*** | 0.74*** | 0.93*** | 0.97*** | 0.95*** | 0.95*** | 0.95*** | 0.99*** | 1.00 | |

FRS (Final rust severity), AUDPC (Area under disease progressive curve), rAUDPC (Relative Area under Disease progressive curve), CV (Coefficient of Variance), YCI (Yellow rust coefficients of infection), CI-1-6 (coefficients of infection first, coefficients of infection second times, coefficients of infection third times, fourth times, five times, six times ), YCI, yellow rust coeficients of infection.

Table 26: Pearson's correlation among slow rusting of wheat stem rust parameters and Agronomic traits at sinana (Below diagram) and Agarfa (Above diagram)

| Character | FRS | ACI | AUDPC | rAUDPC | DH | PH | DM | SPL | SPS | NPT | NSPS | TKW | GYD |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| FRS | | 0.91*** | 0.89*** | 0.89*** | 0.2 ns | -0.11 ns | -0.08** | -0.17 ns | 0.01 ns | -0.03 ns | 0.09 ns | -0.24 ns | -0.13 ns |
| CI | 0.96*** | | 1.00*** | 0.99*** | 0.26*** | -0.08 ns | -0.07 ns | -0.14 ns | 0.01 ns | -0.03 ns | 0.08 ns | -0.30*** | -0.24*** |
| AUDPC | 0.96*** | 1.00*** | | 1.00** | 0.24*** | -0.08 ns | -0.07 ns | -0.14 ns | 0.01 ns | -0.03 ns | 0.07 ns | -0.29*** | -0.24*** |
| rAUDPC | 0.95*** | 1.00*** | 1.00*** | | 0.24*** | -0.08 ns | -0.07ns | -0.14 ns | 0.01 ns | -0.02 ns | 0.07 ns | -0.29*** | -0.24** |
| DH | 0.95ns | 0.03ns | 0.03ns | 0.03ns | | -0.05 ns | -0.42*** | -0.04 ns | 0.26*** | -0.03 ns | 0.08 ns | -0.14 ns | -0.06 ns |
| PH | 0.07ns | 0.06ns | 0.06ns | 0.06ns | -0.13ns | | -0.02ns | 0.38*** | 0.03ns | 0.08 ns | -0.04 ns | 0.08 ns | 0.10 ns |
| DM | 0.06ns | -0.04ns | -0.04ns | -0.04ns | -0.13ns | 0.02ns | | -0.13ns | -0.13 ns | 0.12 ns | -0.07 ns | 0.17* | 0.11ns |
| SPL | -0.04ns | -0.01ns | -0.02ns | -0.02ns | 0.03ns | 0.03* | -0.14ns | | -0.01ns | -0.01 ns | 0.00 ns | 0.07 ns | 0.06 ns |
| SPS | 0.00ns | 0.08ns | 0.07ns | 0.07ns | 0.05ns | 0.06ns | 0.00* | 0.2ns | | -0.06 ns | -0.16 ns | -0.05 ns | 0.03 ns |
| NPT | 0.12ns | 0.05ns | 0.05ns | 0.05ns | 0.03ns | 0.05ns | -0.07ns | 0.13ns | 0.22* | | 0.02 ns | 0.01ns | 0.08 ns |
| NSPS | 0.02ns | -0.09ns | -0.09ns | -0.09ns | -0.04 ns | -0.04 ns | 0.04 ns | 0.08 ns | 0.29*** | 0.02 ns | | 0.09 ns | 0.13 ns |
| TKW | -0.08*** | -0.40*** | -0.40*** | -0.40*** | -0.13ns | 0.26*** | 0.06 ns | 0.07 ns | -0.03 ns | -0.03ns | 0.33*** | | 0.74ns |
| GYD | -0.37*** | -0.27*** | -0.27*** | -0.27*** | -0.14ns | 0.21* | 0.03ns | 0.03ns | 0.05ns | 0.05ns | 0.35*** | 0.74*** | |
| YR | -0.25ns | -0.02 ns | -0.03 ns | -0.03 ns | **-0.02ns** | -0.28*** | -0.03 ns | -0.03 ns | 0.04 ns | -0.03 ns | -0.09 ns | -0.51*** | -0.52*** |

DH = Days to heading, DM = Days maturity, PH = Plant height, NPT = Number of productive tiller, NSPS = Number of Spiklet per spike, SPL = Spike length, NSPS = Number of seed per spike, GYD = Grain yield, FRS = Final rust severity, AUDPC = Area under disease progress curve, rAUDPC = Relative area under disease progress curve.

## DISCUSSION

The present study showed that considerable variability in seedling and Slow rusting (adult-plant resistance) among the evaluated bread wheat genotypes against seven pathotypes/races of *Puccinia graminis* f. sp. *tritici*. The wide range of infection types observed at the seedling stage illustrates the presence of diverse resistance sources within the entire tested bread wheat genotypes. Genetic diversity for resistance genes is essential for the development of durable wheat genotypes, particularly under conditions where there is hot spot of pathogen populations evolve swiftly. Recent studies have shown that wheat rust resistance is controlled by complex genetic architectures involving both major race-specific (qualitative) resistance genes and minor race (quantitative loci) contributing to partial resistance [62].

The Postulation of wheat stem rust resistance genes such as Sr24, Sr11, Sr36, and Sr30 among several genotypes further confirms the importance of these genes in wheat improvement programs. Conversely, the susceptibility detected in some genotypes suggests that virulent pathogen races capable of overcoming certain resistance genes remain prevalent in the wheat stem rust infection. The seedling result for the variety Dure contradicted the report of [9], who postulated the presence of Sr5 in this variety. In the present study, Sr5 showed high IT responses to all seven races, indicating that virulent races have overcome this gene. This suggests that Sr5 resistance in Dure has likely been broken due to the evolution of new races. The evolution of new virulence combinations in *P. graminis* populations has been widely documented, emphasizing the need for continuous surveillance and breeding for durable resistance. Global monitoring programs have reported frequent shifts in dominant rust races, which can rapidly render previously resistant genotypes susceptible [62].

The presence of genotypes resistant to all tested races suggests the possibility of novel resistance genes or favorable gene combinations. Such genotypes represent valuable genetic resources for breeding programs aiming to enhance durable resistance. Screening studies of international wheat germplasm collections have similarly reported that only a small proportion of accessions maintain resistance across multiple races, highlighting the importance of identifying and utilizing such broad-spectrum resistance sources [25].

Field evaluations revealed considerable variation in adult-plant resistance among the tested bread wheat genotypes at both locations Agarfa and Sinana during main cropping season in 2021/2022. Several lines exhibited low final rust severity, reduced coefficient of infection, and low AUDPC values, indicating the presence of slow-rusting resistance. Adult plant resistance is typically controlled by multiple minor genes with additive effects and is considered more durable than major race-specific resistance genes. This form of resistance reduces disease development rather than completely preventing infection, thereby providing stable protection across environments and pathogen populations [25].

The results found in this study are consistent with previous research conducted in hot-spot rust environments in Ethiopia, mainly in the Bale and Arsi highlands wheat production, where strong disease pressure where helps to screen effective selection for durable wheat stem rust resistance. These locations are historically known as hot spot to wheat stem rust races particularly UG99 Race. Natural field evaluations in Ethiopian wheat genotypes have demonstrated that only a small proportion of genotypes exhibit resistance at both seedling and adult plant stages, confirming the importance of identifying and incorporating slow-rusting resistance in breeding programs [14].

Global rust surveillance programs led by the International Maize and Wheat Improvement Center (CIMMYT) have emphasized that rust pathogens continue to evolve rapidly, with new virulent races emerging in different regions of the world. Continuous monitoring of pathogen populations is therefore essential to guide breeding programs and ensure the effectiveness of resistance genes deployed in wheat genotypes [65].

Significant variation in grain yield and yield components among genotypes further demonstrated the negative impact of rust infection on wheat productivity. Genotypes with higher wheat stem rust severity generally produced lower grain yield and reduced thousand-kernel weight, indicating the detrimental effects of rust infection on plant physiology. Hidase variety gave the lowest yield due to the high infections of Wheat stem rust. Wheat stem rust infection reduces photosynthetic activity and interferes with assimilate translocation during grain filling, ultimately resulting in yield loss and reduced grain quality. Similar yield reductions associated with rust epidemics have been reported in several wheat production systems worldwide [74].

The results of this study provide valuable insights for wheat breeding programs aiming to develop genotypes with durable resistance to stem rust. The identification of genotypes exhibiting both seedling resistance and adult-plant slow-rusting resistance indicates that these materials could serve as important parental lines for breeding programs and helps to know the variety background for releases.

## CONCLUSION

Wheat stem rust is a re-emerging disease, epidemic disease outbreaks indicate it can pose a threat again to durum and bread wheat production. Particularly, the occurrence of the recent detected Pgt races with a virulence to 19 Sr differential genotypes included Sr24 (TTKTT) has increased the vulnerability of bread wheat to stem rust. The limited presence of effective stem rust resistance genes in improved materials underscore the need for identifying and incorporating new genes in bread wheat breeding programs due to the virulence of TTKTT and others dominant Pgt races including TTTTF, TTRTF, TTKTF, TKTTF, TKKTF and TTKSKS (UG99). To identify new effective stem rust resistance genes in wheat, it is crucial to evaluate the entries against the dominant Pgt races that are virulent to the most widely used resistance genes.

Wheat genotypes that exhibit intermediate (2+) and/or susceptible (>3- ) infection types may possess race non-specific resistance and these genotypes may provide durable resistance when their field assessment results confirm their slow rusting nature. Hence, nominees for source of slow-rusting resistance were those genotypes that exhibited susceptible and mixed (2+ and 3- ) reaction types. Bread wheat genotypes which had slow-rusting resistance, were assessed based the area under disease progress curve or the measurement of the apparent infection rates and coefficients of infection values.

In this studies location, field assessment for slow-rusting resistance identified on the 28 advanced bread wheat genotypes and commercial genotypes indicates intermediate and susceptible reactions at seedling stage. TTKTT race were used to identify slow ruster. Because, the TTKTT races dominantly identified during survey of 2021 of main cropping season (source: Ambo wheat stem rust race analysis report). True slow-rusters were identified on the 15 bread wheat genotypes 45, 48, 64, 65, 67, 69, 70, 78, 84, 86, 98, 99, 103, 105, and 107 bread genotypes showed which identified based on field evaluation and seedling test (2+ to 3+) exhibiting less than 300 AUDPC values.

Generally, in this present study, 15 advanced bread wheat genotypes showed true slow rusting based on infections types (between 2+ and 3+) and field assessment were found to exhibit a resistant to moderately susceptible response at two field evaluations at sinana and Agarfa test site, while three commercial variety showed good slow rusting among other commercial varieties. Hence, these advanced breeding genotypes could be exploited for improvement of wheat stem rust resistance in common wheat. A higher level and frequency of resistance was detected in advanced genotypes than in commercial varieties. The slow-rusting resistance might not give adequate resistance if exposed to heavy rust epidemics- the commercial cultivars with slow rusting resistance could be deployed with reduced rate of fungicide in hot spot areas.

On the other hand, in this study the Sr resistance genes were postulated to be presented on the bread wheat genotypes. In match with 20 near isogenic genotypes (differential line) four Sr resistance genes were postulated to be presented in the wheat genotypes. Sr11, 36, 30, 24, resistance gene were postulated to be presented in the 7 (5.6%), 2(1.6%), 1(0.8 %), and 4(0.32%) genotypes showed as single seedling resistance. Wheat genotypes that exhibited resistance against all races were identified 20 bread wheat genotypes. However, wheat genotypes carrying multiple stem rust resistance genes did not identified during this study. The un-identified genes have been present in 88 genotypes were possessing at least one low infection types to *Pgt* races which were difficult to identify of Sr resistance. The other genotypes showed unknown Sr resistance gene to be presented in the wheat genotypes. For such cases using more differential genotypes would easily help to postulate the Sr resistance genotypes which to be found in the genotypes.

Therefore, 4 single Sr resistance namely sr 24, sr 30, sr 11 and sr 36; 20 resistance genotypes postulated were identified; 23 wheat genotypes were identified having true slow ruster based on field evaluation and seedling tests. It can be used in future breeding programs for durable slow rusting resistance by tagging of the minor genes responsible for the resistance through molecular marker technology and transferring the genes responsible for the resistance to high yielding genotypes or accumulating different slow rusting genes in a line and increase durability. For further study, to postulate Sr resistance genes using more near isogenic genotypes, using marker assisted, matching genotypic and phenotypic data for wheat genotypes would be important to identify slow rusting resistance or APR, pyramiding R-genes and in combining APR genes with R-genes for Sr resistance gene found in the particular wheat genotypes.

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## Declarations

**Plant material compliance statement**

All plant materials used in this study (bread wheat, Triticum aestivum L.) were obtained from cultivated sources maintained at Sinana agricultural research centers. The collection and use of plant materials complied with relevant institutional, national, and international guidelines and regulations. The experimental work was conducted using officially released varieties and breeding lines; therefore, no specific permission or license was required for their use. All procedures followed standard practices for agricultural research.

**Author contributions**

Zerihun Eshetu: Conceptualization, Methodology, Investigation, Visualization, Validation, Supervision, Writing - Original Draft, Writing - Review & Editing. Zelalem Bekeko: Coordination, Methodology, Edition, supervising and advising. Ayele Badebo; Coordination, Methodology, Edition, supervising and advising.

**Ethics approval and consent to participate:** This study did not involve human participants or animals. All experimental procedures were conducted on plant materials under field and greenhouse conditions in accordance with relevant institutional and national guidelines. Therefore, ethical approval and consent to participate were not required.

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## Figures

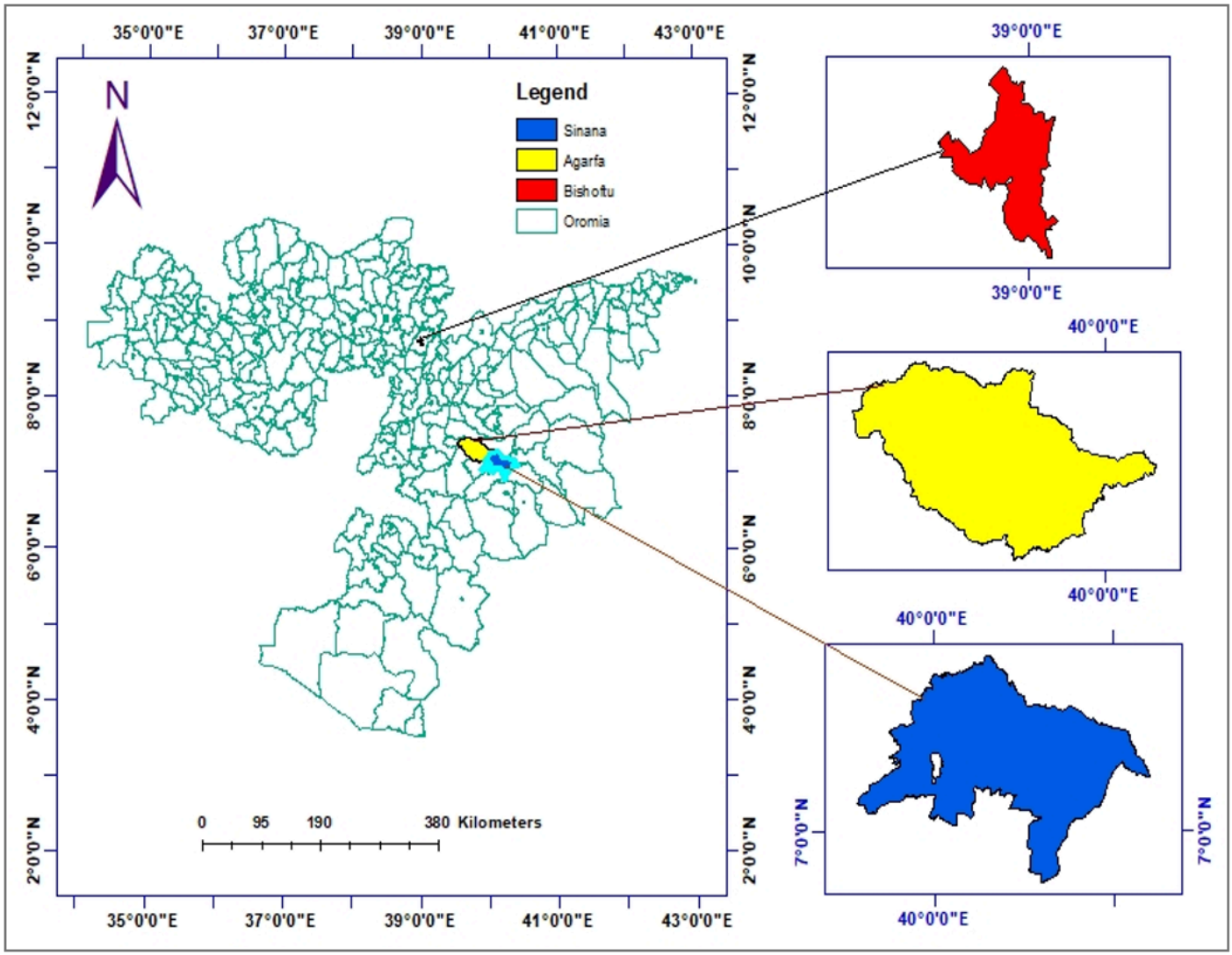



**Figure 1**

Study locations of the experimental sites

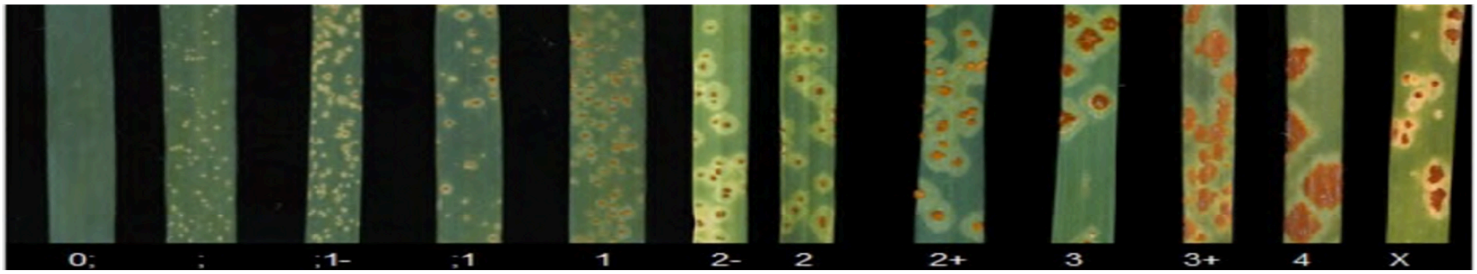



**Figure 2**

Infection types of *P. graminis* f. sp. *tritici* and host response

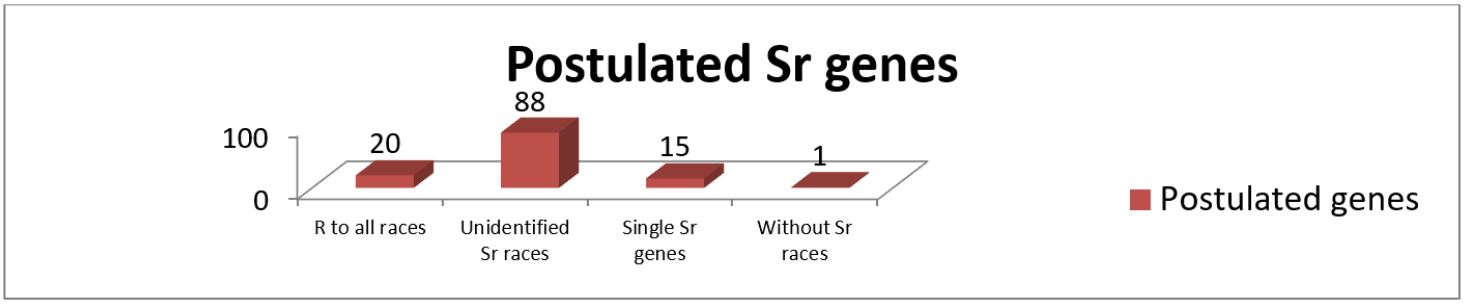



**Figure 3**

**Postulation of bread wheat genotypes for Sr resistance genes**

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