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Effectiveness of the influence of Sr and Lr genes on the field resistance of wheat to stem and leaf rust

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ABSTRACT

Stem and leaf rust have a tremendous impact on wheat yields. The spread of these diseases can compromise any country's food security. The Sr and Lr resistance genes, comprising 60 and 80 genes, respectively, are gradually losing their effectiveness due to the emergence of virulent populations of rust pathogens. This research reports the results of field and molecular studies of resistance to the two types of rust in several varieties and lines of spring soft wheat. It is discovered that the *Sr2, Sr21, Sr32,* and *Sr35* genes have virtually no effect on resistance to stem rust, as with an average plant disease severity of 42.2% the four genes proved ineffective. However, the area under the disease progress curve in the presence of *Sr2* was reliably lower by 3.2% ($p < 0.01$). Regarding the coefficient of infection, susceptibility in the presence of *Sr21* or *Sr35* was reliably lower by 3.5%. The *Lr19, Lr24, Lr27,* and *Lr39* leaf rust genes retain their positive effect on wheat resistance to the disease. The *Lr19* and *Lr39* genotypes showed moderate resistance in 100% of the cases. The average coefficient of infection in the presence of both genes amounted to 6.5 units, with an average plant disease severity equal to 16.3%. The *Lr39* gene reliably reduced the area under the disease progress curve by 4.5 units. The obtained findings indicate the need to identify donors of Sr resistance genes for their use in selection. There is also a need to study the influence and prevalence of the *Lr24* and *Lr27* genes in different varieties and lines of spring soft wheat.

Article type: Research Article.
Keywords: Wheat, Stem and leaf rust, Sr and Lr genes, Resistance.

INTRODUCTION

Rust is widely prevalent across the globe, and its detrimental impact on grain crops has the highest economic significance. Researchers worldwide are working on this global issue. In this endeavor, the most efficient, cost-effective, and environmentally friendly approach is the development and cultivation of resistant varieties (Aktar Uz Zaman *et al.* 2017; Keler & Shram 2021; Hanon Mohsen *et al.* 2022; Rauf *et al.* 2023). Resistance genes are found primarily in foreign varieties and wild forms. These genotypes can be used by agricultural breeders to develop new disease-resistant source material. However, the races of diseases infecting crops within a habitat may be different. Furthermore, variability and selection result in the emergence of new virulent races of fungal diseases (Todorovska *et al.* 2009; Kuldybayev *et al.* 2023). Catastrophic grain losses were experienced across the world due to the spread of the Ug99 race of the *Puccinia graminis* f. sp. *tritici* fungus, which causes stem rust and was first discovered in Uganda in 1998. The harmful effects of Ug99 were so great that in 2005 a global world threat to food security was declared. The infectious effect of Ug99 is attributed to virulence to the *Sr31* and *Sr38* resistance genes, which were identified in *Secale cereale* and *Triticum ventricosum*, respectively, and had been

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successfully used to fight the main set of rust races until that time (Singh *et al.* 2011). At present, there are over 138 identified genes of wheat resistance to leaf (LR) and stem rust (SR; Todorovska *et al.* 2009). In a study of winter wheat rust infestation in the southeastern part of the United States, the most effective genes were found to be *Lr9, Lr10, Lr18, Lr24, Lr37, LrA2K,* and *Lr2K38* for LR resistance, while *Yr17* and *YrR61* for SR resistance (Ghimire *et al.* 2020). For managing SR, studies on Egyptian wheat subjected to pyramiding of resistance genes recommend resistance genes *Sr2, Sr13, Sr22,* and *Sr24* as the most important (Elkot *et al.* 2020). Pyramiding of the *Lr19* and *Lr28* genes shows good results in studies on the resistance of Indian wheat varieties to LR (Bhawar *et al.* 2011). There are several sets of SR resistance genes. The first one includes *Sr5, Sr21, Sr9e,* and *Sr7b*; the second – *Sr11, Sr6, Sr8a,* and *Sr9g*; the third – *Sr36, Sr9b, Sr30,* and *Sr17*; and the fourth – *Sr9a, Sr9d, Sr10,* and *SrTmp*. Researchers in Kazakhstan have discovered an additional set of SR resistance genes, which includes *Sr24, Sr25, Sr27,* and *Sr32* (Zatybekov *et al.* 2022; Rsaliyev & Rsaliyev 2018). The *Sr22* and *Sr35* genes that effectively mitigate the effects of the Ug99 race have been identified in *T. monococcum* wheat. Unfortunately, scientific research by plant breeders, plant protection and quarantine specialists, and phytopathologists in Kazakhstan shows that there are practically no disease-resistant, including rust-resistant, wheat varieties allowed for cultivation in the country. The rust epidemics of 2015-2018 caused damage to grain crops ranging from 70 to 90% of disease incidence in the main grain-growing regions in Kazakhstan. The infection of varieties susceptible to rust causes anywhere from 20% yield reduction to its complete loss, as well as, most importantly, a sharp drop in the quality of the grain obtained (Zatybekov *et al.* 2022). In this connection, constant screening of wheat varieties and lines for rust resistance genes, or rather pyramids of efficient combinations of such genes, is an urgent task. Information about selection material and commercial varieties will allow for better genetic control of rusts. The purpose of the present study is to identify SR and LR resistance genes in the varieties and hybrids cultivated in Northern and Central Kazakhstan and to determine their efficiency in developing resistance to these diseases in spring soft wheat.

MATERIALS AND METHODS

The research was conducted based on the Agricultural Biotechnology Research Platform at S. Seifullin Kazakh AgroTechnical Research University. Breeding material consisted of accessions provided by the leading breeding centers of Northern and Central Kazakhstan, namely A.I. Barayev Scientific-Production Center for Grain Farming, North-Kazakhstan Experimental Station, Khristenko Karaganda Experimental Station, and Karabalyk Experimental Station. The study examined 31 accessions of spring soft wheat. Resistance of the considered varieties and lines was tested under conditions of natural contagion based on the Research Institute of Biological Safety Problems, Gvardeysky village, Korday District, Jambyl Region, Kazakhstan.

Disease assessment and sample selection

SR and LR assessment was conducted under natural infection conditions from May 7 to May 21, 2022, which corresponds to the growth stages of an adult GS 83-90 plant.

1.The isolates used for inoculation were *P. graminis* f. sp. *tritici* and *P. triticiana*. The production and propagation of monopustular isolates of *P. graminis* f. sp. *tritici* were carried out in a greenhouse setting. Urediniospore materials of rust species pathogens from dry leaves and stems were reanimated on a susceptible variety and then cloned. After moldboard plowing and harrowing, the field plot was cultivated with a SOLO 503 cultivator. The soil type was alluvial sierozem fertilized with humus. Seeds were sown by hand in plots, 0.4-3.0 m^2 in area, in 100-300 cm rows 20 cm apart. In each row, 65-80 seeds were sown. To create favorable conditions for plant development and rust pathogens, experimental plots were regularly watered and sprayed with water.

In spring, during the tiller stage, the spring wheat crops were infected with SR and LR urediniospores. The infection was performed using only the local fungal population or a mixture of isolates with specific virulence. The inoculum taken for infection was activated at a temperature of 37-40 ℃ for 30 min, followed by watering in a humid chamber at 18-22 ℃ for 24 h. The infectious material was applied to plants by spraying an aqueous suspension of spores with 0.001% TWEEN-80. Plant infection was performed in the evening in windless weather after watering and moistening the leaves of experimental crops in advance. The viral load of spores amounted to 20 mg m^{-2}. After infection, the plots were covered with plastic sheeting for 16-18 h to ensure high moisture content. During the vegetation period, an assessment of field resistance to LR and SR was performed three times with a two-week interval starting from the appearance of the first pustules according to the established scales. The

type of infection (in points) with SR and LR was determined according to a predetermined scale. Immunity corresponded to 0 points, 1 point signified resistance (R), 2 – moderate resistance (MR), 3 – moderate susceptibility (MS), and 4 – susceptibility (S). Plant disease severity (PDS, %) was estimated using a modified version of the Cobb scale. The indicator describing the non-specific resistance of the variety was the disease progression rate criterion expressed through the area under the disease progress curve (AUDPC). The coefficient of infection (CI) was calculated by multiplying PDS by the host plant reaction type constant. The constants for each type of reaction are as follows: immunity = 0.0; R = 0.2; MR = 0.4, MS = 0.8, S = 1.0. The use of two different factors for calculation may result in the same or similar CI values from different values of these factors. In general, low CI values reflect low levels of disease severity.

Gene identification using PCR

In the course of molecular genetic work with spring wheat samples, selection was carried out by genes: resistance to rust was determined by the genes that are known and found most effective in wheat, namely *Sr2, Sr21, Sr35, Lr 19, Lr24, Lr 37,* and *Lr 39.*

Genomic DNA was isolated from plant material using 10-day-old wheat seedlings via the STAB method (Baranova *et al.* 2015).

Following the method, the leaves were first ground, then the STAB buffer was added, followed by adding RNase, incubation, and adding chloroform-isoamyl alcohol. DNA dissolution was performed in a TE buffer. The efficiency of DNA extraction from plant material was evaluated using a *NanoDrop 2000 (Thermo Fisher Scientific, USA)*.

Amplification of marker genes was performed in a final reaction volume of 15 μL containing: 1.5 mM of Se-buffer, 2 mM of dNTP, 50 mM of $MgCl_2$, 10 pmol of forward and reverse primer, 1U Taq DNA polymerase, and DNA at a concentration of 100 ng μL^{-1}. Table 1 indicates primer sequence, annealing temperature, and gene product size.

Table 1. Nucleotide sequence of primers for the genes of wheat resistance against pathogenic fungi.

№	Primer	Primer sequences (5' to 3')	Primer annealing, ℃	Product size, bp
1	*Lr19*	Forward: CCTGATCACCAATGACGATT Reverse: CCTGATCACCTTGCTACAGA	60	688
2	*Lr*24	Forward: CACCCGTGACATGCTCGTA Reverse: AACAGGAAATGAGCAACGATGT	60	500
3	*Lr*37	Forward: AGGGGCTACTGACCAAGGCT Reverse: TGCAGCTACAGCAGTATGTACACAAAA	62	300
4	*Lr*39	Forward: CGC TTT TAC CGA GAT TGG TC Reverse: CCA AAG AGC ATC CAT GGT GT	58	300
5	*Sr*2	Forward: AAGGCGAATCAAACGGAATA Reverse: GTTGCTTTAGGGGAAAAGCC	60	120
6	*Sr*21	Forward: ATC GCA TGA TGC ACG TAG AG Reverse: ACA TGC ATG CCT ACC TAA TGG	60	200
7	*Sr*32	Forward: GCGGTCAAGACACTCCACTCCTCTCTC Reverse: CGCTGCTCCCATTGCTCGCCGTTA	56	200
8	*Sr*35	Forward: ACATGTGATGTGCGGTCATT Reverse: TCCTCAGAACCCCATTCTTG	60	230

The PCR setup mode and the components of the reaction mixture for PCR setup were fine-tuned for the detection of each gene under study. Amplification products were separated in a 1.5% agarose gel with the addition of ethidium bromide. The electrode buffer used was a 1× TBE buffer. The results were visualized in GelDoc (VilberLourmat, France). A GeneRuler 1kb DNA Ladder (Thermo Fisher Scientific, USA) was used as a marker of molecular masses.

Data analysis

The effect of genotype and localization on the response to SR infection was investigated using a two-factor analysis of variance (ANOVA). The Pearson correlation analysis was performed using RStudio software. The response to infection for each sample was converted to a constant value (where the types of infection R, MR, M, MS, and S were assigned constant values of 0.2, 0.4, 0.6, 0.8, and 1, respectively) and multiplied by terminal

disease severity to obtain the CI. Phenotypic and genetic parameters were analyzed using SPSS 22.0 software (nonparametric Mann-Whitney test).

RESULTS

The field experiments on resistance to SR and LR showed that the level of resistance of the selected varieties and lines to SR was much lower than to LR. A total of 31 varieties of spring soft wheat were immunologically evaluated under field conditions against an artificial infectious background of SR and LR. Analysis of the field assessment of spring wheat nurseries for resistance to SR and LR is given in Tables 2 and 3.

Table 2. Presence or absence of the genes of wheat resistance to pathogenic fungi on an artificial infectious background of SR and LR.

№	Variety, sample	SR, PDS and RT, (%)	SR, AUDPC, units	SR, CI	LR, PDS and RT (%)	LR, AUDPC, units	LR, CI
1	Aktobe 39	30MS	315	24	10R	105	2
2	Stepnaya 2	40S	420	40	40R	490	8
3	Stepnaya 50	50MR	665	20	0	0	0
4	Ekada 113	40MS	350	32	0	0	0
5	Dinastia	40MR	560	16	0	280	0
6	Stepnaya 53	50S	525	50	30MR	245	12
7	Stepnaya 75	50S	665	50	10MR	105	4
8	Karagandinskaya 55	40MR	315	16	10MR	70	4
9	Bayterek 15	30S	210	30	10MR	35	4
10	Line R-1413m	40S	350	40	30MR	385	12
11	Line R-1415m	30MR	315	12	20MR	280	8
12	Line 201/21g	50S	525	50	30MR	175	12
13	Line 205/21g	60S	770	60	30MR	455	12
14	Line 225/21g	40S	490	40	20R	210	4
15	Lutescens 2261	50MS	665	40	20MR	175	8
16	Lutescens 2262	60MR	525	24	40MR	420	16
17	Lutescens 1519	60MR	490	24	30MR	280	12
18	Lutescens 2202	40MR	350	16	30MR	210	12
19	Lutescens 2203	50MS	595	40	30MR	210	12
20	Lutescens 2205	30MR	280	12	20MR	140	8
21	Lutescens 2207	50MS	455	40	0	0	0
22	Lutescens 2210	50MR	525	20	0	0	0
23	Eritrospermum 255	40MR	420	16	20MR	105	8
24	435/Lutescens 2	30S	385	30	10MR	35	4
25	659/12	30MS	315	24	10MR	105	4
26	486/Lutescens 22	30MR	455	12	0	0	0
27	63/ Lutescens 37	40S	700	40	20MR	280	8
28	23/07	30S	455	30	10MR	140	4
29	218/10	40S	630	40	20MR	280	8
30	Eritrospermum 42/12	40S	560	40	20MS	350	16
31	Lutescens 13/12	30S	210	30	10MR	35	4

Among nine varieties, moderate susceptibility was found only in three, with PDS reaching 30-50%, i.e., Stepnaya 50, Dinastia, and Karagandinskaya 55. However, the AUDPC in Karagandinskaya 55 equaled 315 units, and in

Stepnaya 50 and Dinastia – 665 and 560 units, respectively. In all three varieties, the CI reliably indicated moderate resistance, ranging from 16 to 20. Aktobe 39 and Ekada 113 demonstrated moderate susceptibility to SR. Their CI amounted to 24 and 32, respectively. Stepnaya 2, Stepnaya 53, Stepnaya 75, and Bayterek 15 had a low resistance to SR. The AUDPC in these varieties was 420, 525, 665, and 210 units, respectively, with PDS reaching 30-60%. Wheat lines also demonstrated low resistance to SR. Of the 29 lines, eight showed moderate resistance with PDS at 30-60%, four were moderately resistant with PDS at 30-50%, and 10 proved susceptible to rust with PDS at 30-60%. By the indicators of resistance to LR, the varieties and lines demonstrated a different trend. From all samples, only the Eritrospermum 42/12 line exhibited moderate susceptibility to LR. In contrast, the varieties Stepnaya 50, Ekada 113, and Dinastia as well as the lines Lutescens 2207, Lutescens 2210, and 486/Lutescens 22 were completely immune to the symptoms of the disease. Two varieties, Aktobe 39 and Stepnaya 2, and Line 225/21g were resistant to LR with CI of 2.8 and 4, respectively. The remaining 21 samples were moderately resistant with PDS not exceeding 40% and the CI below 16. The findings indicate that the varieties and lines were resistant to LR in 96.8% of the cases, while moderate resistance to SR was found only in 64.5%. Next, to investigate the influence of Sr and Lr genes on field resistance, molecular biology methods were applied to identify the genes *Sr2, Sr21, Sr32, Sr35, Lr 19, Lr24, Lr 37,* and *Lr 39.* All 31 spring soft wheat samples were subjected to molecular genetic screening. Table 3 presents the results of screening for the presence or absence of Sr and Lr genes.

Table 3. Presence or absence of the genes of wheat resistance to pathogenic fungi.

№	Variety, sample	*Sr2*	*Sr21*	*Sr32*	*Sr35*	*Lr 19*	*Lr24*	*Lr 37*	*Lr39*
1	Aktobe 39	+	+	+	+	+	-	-	-
2	Stepnaya 2	+	+	-	-	+	-	-	-
3	Stepnaya 50	-	-	+	+	+	+	-	+
4	Ekada 113	+	+	-	+	+	-	-	+
5	Dinastia	-	+	+	+	+	-	-	+
6	Stepnaya 53	-	+	+	+	+	-	-	+
7	Stepnaya 75	+	+	+	+	+	-	-	+
8	Karagandinskaya 55	+	-	+	+	+	-	-	+
9	Bayterek 15	+	+	-	-	-	-	-	+
10	Line R-1413m	+	+	+	+	+	-	-	+
11	Line R-1415m	+	+	+	+	+	-	-	+
12	Line 201/21g	+	+	+	+	+	-	-	+
13	Line 205/21g	+	+	+	+	+	-	-	+
14	Line 225/21g	+	+	+	+	+	-	-	+
15	Lutescens 2261	+	+	+	-	+	-	-	+
16	Lutescens 2262	+	+	-	+	+	-	-	+
17	Lutescens 1519	+	+	+	+	+	-	-	+
18	Lutescens 2202	-	+	+	+	+	-	-	-
19	Lutescens 2203	+	+	+	+	+	-	-	+
20	Lutescens 2205	+	+	+	+	+	-	-	+
21	Lutescens 2207	+	+	+	+	+	-	-	+
22	Lutescens 2210	+	+	+	+	+	-	-	+
23	Eritrospermum 255	-	+	-	-	-	-	-	-
24	435/Lutescens 2	+	+	+	+	+	-	-	+
25	659/12	+	+	+	+	+	-	-	+
26	486/Lutescens 22	-	+	-	-	+	-	-	+
27	63/ Lutescens 37	-	+	+	+	+	-	-	+
28	23/07	-	+	+	+	+	-	-	+
29	218/10	+	+	+	+	+	-	-	+
30	Eritrospermum 42/12	+	+	+	+	+	-	-	+
31	Lutescens 13/12	-	+	+	+	+	-	-	+

Molecular screening reveals that the *Sr2, Sr21, Sr32,* and *Sr35* genes were present in almost all samples. Specifically, the *Sr2* gene was found in 71% of the samples, *Sr21* – in 93.5%, *Sr32* – in 80.6%, and *Sr35* – in 83.8%. Both *Sr2* and *Sr21* were absent only in one sample –Stepnaya 50. The Bayterek 15 and Stepnaya 2 varieties as well as the Eritrospermum 255 and 486/Lutescens 22 lines lacked both *Sr32* and *Sr35*. However, Bayterek and

Stepnaya 2 displayed resistance to SR with a PDS of 30-40%, while Eritrospermum 255 and 486/Lut 22 exhibited moderate resistance with a PDS of 30-40%. Among the LR resistance genes, the *Lr 37* gene was missing in all tested samples. The *Lr24* gene was found in the genome of only one variety, i.e., Stepnaya 50, which displayed immunity to LR. The *Lr 19* gene was present in 93.5% of the samples, and the *Lr 39* gene in 87.1%. Only one gene out of the four, i.e., *Lr 19*, was present in the genome of the Aktobe 39 and Stepnaya 2 varieties and the Lutescens 2202 line, which exhibited resistance to LR with a PDS of 10 and 40%, respectively. Bayterek 15 had only *Lr 39* in its genome, yet in field tests; it revealed moderate resistance with a PDS of 10% and a CI of 4. Eritrospermum 255 had no LR resistance genes in its genome, and yet it was found moderately resistant to the disease with a PDS of 20%, an AUDPC of 105 units, and a CI of 8. The genomes of 80.6% of the samples had two genes, i.e., *Lr 19* and *Lr 39*, while their resistance ranged from immunity to moderate susceptibility.

Correlation analysis

To understand the direct influence of Sr and Lr genes on the manifestation of resistance traits in wheat plants, we conducted a correlation analysis. Our analysis using the Mann-Whitney criterion provided a correlation data series (Table 4).

Table 4. Correlation analysis between Sr genes and the level of wheat resistance to SR (*p ≤ 0.05; **p ≤ 0.01).

Gene	N	SR, PDS (%)	SR, AUDPC, units	SR, CI
Sr2	22	42.7 ± 2.2	461.4 ± 31.6**	33.1 ± 2.7
Sr21	29	41.9 ± 1.8	471.9 ± 27.0	31.5 ± 2.4*
Sr32	25	42 ± 1.9	484.4 ± 30.4	32.2 ± 2.7
Sr35	26	42.3 ± 1.9	473.9 ± 28.8	31.5 ± 2.6*

The presented data show that the presence of any of the four genes in the wheat genome does not affect plant resistance to SR with reliability. With an average PDS of 42.2%, the impact of the four genes did not prove effective. However, the AUDPC in the presence of *Sr 2* was reliably lower by 3.2% ($p < 0.01$). Considering the CI, susceptibility was found to be reliably lower by 3.5% in the presence of the *Sr21* and *Sr35* genes. With respect to LR, the data indicate the efficiency of Lr genes in the formation of wheat resistance preserved in the process of selection. Table 5 shows the data of correlation analysis between Lr genes and the level of resistance to LR in 31 samples of spring soft wheat. Correlation analysis exhibited the preserved effectiveness of the genes in forming plant resistance to LR. The presence of the *Lr19* and *Lr 39* genes was associated with moderate susceptibility to the disease in 100% of the cases. The average CI with both genes amounted to 6.5 units, while the average PDS was 16.3%. In the presence of the *Lr39* gene, the AUDPC was reliably lower by 4.5 units. Of more than 25 samples with both genes, only one displayed susceptibility to the disease (Eritrospermum 42/12). With only the *Lr19* gene present, Stepnaya 2 showed moderate resistance to the disease. The *Lr37* was not found in any of the 31 samples. With the *Lr19*, *Lr34*, and *Lr39* genes present in its genome, the Stepnaya 50 variety was immune to LR. However, given that none of the other samples had this group of genes, it is impossible to conclude the impact of the three genes on full immunity to rust.

Table 5. Correlation analysis between Lr genes and the level of wheat resistance to LR (**p ≤ 0.01; ***p ≤ 0.001).

	N	LR, PDS, %	LR, AUDPC, units	LR, CI
Lr19	29	16.8 ± 2.4	181.3 ± 27.0	6.5 ± 0.9
Lr24	1	0***	0***	0***
Lr37Null	0	0	0	0
Lr39	27	15.9 ± 2.3*	173.7 ± 26.9**	6.5 ± 0.9

DISCUSSION

Our studies indicate that Sr genes are common in the lines and varieties cultivated in Northern and Central Kazakhstan. However, genotypes with the *Sr2*, *Sr21*, *Sr32*, and *Sr35* genes did not exhibit resistance to SR. Research conducted by Chinese scientists suggests that the *Sr21* is ineffective against the 34C3RKGQM race but does prove effective against the 21C3CTHTM race. The *Sr32* and *Sr35* genes are ineffective against the six races examined (Xu *et al.* 2017). The effectiveness of these genes was observed only in 35.5% of the samples, exhibiting

moderate resistance to the disease. Considering LR, almost all genotypes with the *Lr 19* and *Lr 39* genes displayed moderate resistance, resistance, or immunity in 74.2% of the samples. *Lr19* ensures the resistance of seedlings (Elena *et al.* 2023), while *Lr24* exhibits resistance to rust at all stages of wheat plant growth and *Lr37* appears to be the gene of resistance in more mature plants (Wang *et al.* 2019; Babkenov *et al.* 2023). The *Lr37* gene was not found in any of the 31 samples tested. It is possible that this gene is not specific to the studied varieties and does not participate in the formation of resistance. Literature indicates that this gene is involved in race-specific resistance and induces hypersensitivity reactions when interacting with a pathogen on a gene-for-gene basis (Elena *et al.* 2023). To obtain good quality grain and support high yields, crop producers should rely on disease-resistant wheat varieties. To this end, scientists in many countries are developing several molecular markers linked to resistance genes for these diseases (Mago *et al.* 2011; Yadav *et al.* 2015; Koyshybaev *et al.* 2017; Xu *et al.* 2017). At present, 68 Lr and 50 Sr genes and their alleles are known to be associated with LR and SR (Lin *et al.* 2021; Kuldeep *et al.* 2022). Many of these are wheat genes, while others are transferred from other relatives. For example, the diversity of Sr genes in the soft wheat gene pool has been significantly enriched due to the transfer of Sr genes from species belonging to its primary, secondary, as well as tertiary gene pools. The introgressive gene *Sr2* co-segregated with *Lr19* on one arm of the 3BS chromosome came from *T. turgidum* ssp. *dicoccum*. Races virulent to *Sr21* are often found in North and South America, and virulent mutants are also found in Australia (Nsabiyera *et al.* 2016). However, this gene provides resistance against most Ug99 races (Aktar Uz Zaman *et al.* 2017; Karelov *et al.* 2022). The *Sr32* was introgressed from *Aegilops speltoides* Tausch in several cases independently into all chromosomes of group 2. According to previous studies in the USA, Canada, Mexico, and South Africa, races virulent to the gene were not detected until recently, when a race with virulence to *Sr32* was found in Kazakhstan (Olivera Firpo *et al.* 2022). This gene has not been widely used in breeding programs because of the scale attachment trait and other detrimental traits. The Sr35 resistance gene was transferred from *T. monococcum* to the 3AL chromosome of soft wheat and confers resistance to the TTKSK (Ug99) race of SR as well as its variants TTKST and TTTSK (Jin *et al.* 2007). *Sr35* confers resistance to *P. graminis* races common in Australia and North America, however, there are races virulent to this gene in Ethiopia, Kenya, Malaysia, Nepal, Brazil, Chile, Argentina, and China (Karelov *et al.* 2022). Selected LR resistance genes are also introgressed genes from *Thinopyrum ponticum* (*Lr24*), *Aegilops tauschii* (*Lr21* and *Lr39*), *Ae. ventricosa* (*Lr37*), and *T. elongatum* Zhuk (*Lr 19*; Lin *et al.* 2021; Kuldeep *et al.* 2022). According to the conducted studies, Sr genes in the conditions of the Northern and Central regions of Kazakhstan have lost their efficiency. Correlation analysis shows that in the presence of *Sr2* genes, the AUDPC is reliably lower by 15.3 units. In genotypes with *Sr21* and *Sr37*, the CI is lower by 1.15. Lr genes, particularly *Lr19* and *Lr39*, exhibit over 70% of moderate resistance to LR. Disease symptoms are non-significant, with a PDS of no more than 40% and a CI under 16. Thus, one of the major challenges for wheat breeders today is to regularly develop new varieties or improve old varieties using new resistance genes, as new pathotypes and virulence races continue to emerge. Hence, continuous and active efforts are needed to find sources of new genes/QTLs to overcome newly evolving races of pathogens and to achieve long-term resistance under field conditions.

CONCLUSIONS

The conducted studies on resistance to SR and LR in 31 samples of spring soft wheat exhibited the level of influence and prevalence of the *Sr2*, *Sr21*, *Sr32*, *Sr35*, *Lr19*, *Lr24*, *Lr37*, and *Lr39* genes. The findings revealed that Sr genes have virtually lost their effectiveness in forming resistance to SR. With an average PDS of 42.2%, the four genes were concluded to be ineffective. However, the AUDPC in the presence of *Sr2* was reliably lower by 3.2% ($p < 0.01$). Considering the CI, susceptibility was reliably lower by 3.5% in the presence of *Sr21* or *Sr35*. Lr genes retained their effectiveness to this day, giving genotypes a level of resistance ranging from immunity to moderate susceptibility. The presence of *Lr 19* and *Lr 39* offered moderate susceptibility to the disease in 100% of the cases. The average CI with both genes present was 6.5 units, while the average PDS amounted to 16.3%. In the presence of the Lr39 gene, the AUDPC was reliably lower by 4.5 units. Nevertheless, it is still unclear how the *Lr24* and *Lr 37* genes affect wheat resistance. Further studies on a larger sample are needed to gather information about the prevalence of these genes in wheat varieties in Kazakhstan. Furthermore, it is important to identify genotypes with Sr genes that affect the formation of resistance to SR in the varieties cultivated in Kazakhstan to find donors of resistance genes to be used in breeding programs.

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