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IDENTIFICATION OF WHEAT GERMPLASM RESISTANT TO LEAF, STRIPE AND STEM RUST USING MOLECULAR MARKERS

Abstract. Wheat leaf rust, stripe and stem rust are major wheat diseases in Kazakhstan that reduce yield and quality and cause considerable economic damage. This study utilized winter wheat germplasm from different national and international nurseries to evaluate their value for genetic and breeding programs directed towards improvement of wheat rust resistance in Kazakhstan. Based on the data from field experiments, the most valuable sources, combined resistance to both leaf and stripe rust were 16 lines and cultivars (28.6%), including mainly entries from CIMMYT and IWWIP. Nineteen entries (30.6%) had high level of resistance to leaf rust in the field tests. Thirty-three entries (53%) were effective to control stripe rust. In our study 22% wheat accessions studied had polymorphic band linked to leaf rust resistance gene *Lr10*. Based on rust reactions and data of molecular analysis, 3% entries were found to have *Lr19/Sr25*, 11% entries – *Lr26/Sr31/Yr9/Pm8*, 43% entries – *Lr34/Yr18*, 12% entries – *Lr37/Yr17/Sr38*, 17% – *Lr68* gene and 6% entries – *Yr10* gene. Only one line from IWWIP nursery showed presence of *Yr15* gene. Out of 38 studied entries, the fragment of DNA associated with *Sr22* gene in 13 wheat entries observed. Gene *Sr22* was identified in five Kazakhstani and in 8 Belarusian wheat entries. The results obtained used for developing wheat cultivars resistant to rust.

Key words: wheat, rust species, molecular markers, resistance genes.

Introduction. The region of Central Asia is one of the most important wheat areas in the world. Currently Kazakhstan produces 18-20 million tons of wheat grain [1,2]. Wheat production in Kazakhstan is seriously constrained due to rust diseases, including stem rust caused by *Puccinia graminis* f. sp. *tritici*, stripe rust caused by *Puccinia striiformis* f.sp. *tritici* and leaf rust caused by *Puccinia recondita* f. sp. *tritici* [3,4]. In the period between 2001 and 2007 in north Kazakhstan, epidemic development of *Puccinia triticina* *Eriks* occurred four times (2002, 2003, 2005 and 2007). Leaf rust had severe development in south and south-east of Kazakhstan. Infection on commercial cultivars was up to 20%, collection accessions in demonstrative plots were damaged up to 20-70% [3,5,6]. Currently, more than 110 leaf rust, 86 stem rust, and 83 stripe rust resistance genes have been reported in wheat or wild relatives, most conferring race-specific resistance [7]. Some of the important stem rust resistance genes are *Sr2*, *Sr23*, *Sr24*, *Sr25*, *Sr31*, *Sr33*, *Sr35*, *Sr36*, *Sr38*, *Sr45*, *Sr50*, *SrTmp*, and *Sr1RSAmigo*. Important stripe rust resistance genes are *Yr2*, *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr17* and *Yr27*; and leaf rust resistance genes are *Lr9*, *Lr14a*, *Lr16*, *Lr17a*, *Lr21*, *Lr22*, *Lr24*, *Lr26*, *Lr32*, *Lr39* and *Lr41* [8,9]. In order to more reliably select and deploy disease resistance it is very important to use molecular-genetic markers tightly linked to this trait.

The current study undertaken to asses of 62 wheat cultivars and lines developed in Kazakhstan and in International Centers ICARDA-CIMMYT and Kazakhstan to stripe, leaf and stem rust and to screen wheat material using molecular markers.

Materials and methods. The 62 wheat entries were evaluated for stripe, leaf and stem rust at the adult plant stage under natural conditions. Field test, according to complete randomized block design with three replicates were conducted during the 2017-2019 cropping seasons at the experimental station in v. Almalybak, Almaty oblast, Kazakhstan. The wheat variety, susceptible check Morocco, was planted all-

around the experimental field in order to create a stripe and leaf rust epidemic. Disease severity and adult plant response to stripe and leaf rust recorded following McIntosh et al., 1995 [10]. Evaluation of the development of stripe and leaf rust was carried out in phase milky-wax ripeness according to procedure adopted in CIMMYT. Stripe and leaf rust infection type (IT) and disease severity (DS; percentage leaf area infected) based on the modified Cobb's scale; the IT data for stem rust using the methods described by Stakman and Levine (1922) were analysed [11]. Disease severity was recorded following Peterson et al. (1948) [12].

Genomic DNA was extracted at two-leaf seedling stage for each individual plant using the CTAB method [13]. The DNA concentration for each sample was adjusted to 30 ng/μl. Samples were genotyped using the markers designed to detect alleles of the *Yr*, *Lr* and *Sr* genes. PCR amplification was undertaken in 25-μl volumes, in thermal cycler (Bio-Rad, France). For each PCR reactions, reaction volume contained 30 ng of template DNA, 0.5 of each primer (1pM/μl) synthesized by Sigma-Aldrich, 2.5 μl dNTP mixture (2.5 mM of each nucleotide) (ZAO Sileks, Russia), 2.5 μl of MgCl₂ (25mM), 0.5 μl TaqDNA polymerase (0.5 U, 5 U/μl (ZAO Sileks, Russia), 2.5 μl 10X PCR buffer (ZAO Sileks, Russia), 16.0 μl of MQ-H2O. Temperature profiles consisted of an initial denaturation at 94°C for 3 min, and then 45 cycles of the following program: 94°C for 1 min, 60°C for 1 min, and 72°C for 2 min, and final extension in 72°C for 10 min. The amplification products were separated on 2%-agarose gels in the TBE buffer (45mM Tris borate, 1 mM EDTA, pH 8); gene-Ruler™, 100 bp DNA Ladder (Ferments, Lithuania). Gels were visualized on Gel Documentation System (Gel Doc XR+, BIO-RAD, Hercules, USA) for documentation of allele types in cultivars.

Results. The resistance evaluation (infection types and severity) of the 62 entries in field test are listed in Table. Accessions studied had variable reactions to leaf and stripe rust from 40MS to high resistant (0-R). The maximum disease recorded for entries studied was 40MS for leaf rust and 30MS for stripe rust, compared to 100S for susceptible check Morocco. Based on field reactions, the 62 entries could be classified into 3 groups. The first group consisted of 16 lines and cultivars that were resistant to both leaf and stripe rust (IT – 0-5R). This group consisted of 28.6% entries with codes 18, 23, 24, 26, 27, 30, 31, 32, 34, 38, 39, 41, 42, 43, 58, 59 and includes mainly entries from CIMMYT and IWWIP, as shown in Table. The second group consisted of nineteen entries (30.6%) that have high level of resistance to leaf rust in the field tests. The third group consisted of thirty-three entries (53%) that were resistant to stripe rust in the field and have great effectiveness to control stripe rust. The remaining 10 entries were moderately susceptible (MS) to both leaf and stripe rust. Sixty two wheat cultivars (lines) were examined by using molecular markers for eight *Lr* and *Yr* genes against the fungal pathogen of wheat (table). Data presented in Table illustrates, that leaf rust resistance gene *Lr10* was identified in 14 entries used (22.6%), including five CIMMYT lines, seven IWWIP lines and one cultivar from Kazakhstan. In these accessions DNA amplification products 310 bp, corresponding to STS marker of *Lr10* – F1.2245Lr10-6/r2. The disease severity in these entries to leaf rust was estimated between 0 – 20MS. Amplification products of marker for *Lr19/Sr25* was found in two Russian accessions Lyubava and 113/00i-4, accounting for 3,2% of 62 wheat entries studied, suggesting that leaf rust resistance was from *Lr19*. The dominant marker *Gb* amplified a 130 bp fragment only in the *Lr19/Sr25*-positive lines and no PCR product was obtained in wheat lines that lack this complex. Marker analyses indicate that *Lr26/Yr9/Sr31/Pm8* gene block is present in approximately 11% of tested cultivars (lines) (Table). Disease severity in these entries to leaf rust was estimated between 5MR – 40MS, and to yellow rust – 5R – 20MS. Amplification products 150 bp, csLV34 corresponds with homozygous resistant allele of *Lr34/Yr18* gene were detected in 27 wheat entries, including 15 CIMMYT lines, 6 IWWIP lines and 6 cultivars from Kazakhstan. These 27 cultivars (lines) accounting for almost 44% of genotypes studied, suggesting that the stripe rust resistance of this breeding material was from *Lr34/Yr18* (Table). Disease severity in these entries to leaf rust was estimated between 5MR – 40MS, and to yellow rust – 5R – 20MS. Eight of 62 tested cultivars showed the 262-bp diagnostic DNA fragment associated to *Lr37/Yr17*. Of the 62 cultivars (lines) identified to carry these resistance genes in our study, 8 accessions (13%), including 2 lines from CIMMYT and 6 IWWIP lines amplified 262 bp fragment indicating the presence of the *Lr37/Yr17/Sr38* resistance gene block. *Lr37/Yr17* carriers have been found from immune to susceptible against field collection of leaf and stripe rust at adult plant stage. Disease severity in these entries to leaf rust was estimated between 0 – 40MS, and to stripe rust – 0 – 30MS. Amplification products of marker for *Lr68* gene were found in 11 wheat accessions

Field responses of wheat entries to leaf and yellow rust and molecular identification of *Lr* and *Yr* resistance genes among wheat entries

Accessions	Origin*	Field responses**		<i>Lr</i> 10	<i>Lr</i> 19/ <i>Sr</i> 25	<i>Lr</i> 26/ <i>Yr</i> 9/ <i>Sr</i> 31	<i>Lr</i> 34/ <i>Yr</i> 18/ <i>Pm</i> 38	<i>Lr</i> 37/ <i>Yr</i> 17/ <i>Sr</i> 38	<i>Lr</i> 68	<i>Yr</i> 10	<i>Yr</i> 15
		Leaf rust	Stripe rust								
1	2	3	4	5	6	7	8	9	10	11	12
338-K1-1//...MLT	CIMMYT	5MR	5MS	0	0	0	1	0	0	0	-
KINACI97/4/.../MLT	CIMMYT	10MR	5MR	1	0	0	1	0	0	0	-
AGRI/...PYN/BAU	CIMMYT	5R	30MS	0	0	0	1	0	0	0	-
TAM200/3/.../MLT	CIMMYT	5R	30MS	0	0	0	1	0	0	0	-
WRM/4/FN/...ATR71/	CIMMYT	20MR	5MR	0	0	0	1	0	0	1+	-
T-2003...1D13.1/MLT	CIMMYT	10MS	20MS	0	0	0	1	0	0	0	-
TAM10...BONITO-36	CIMMYT	5MR	10MR	0	0	0	1	0	0	0	-
TX87V161...ATTILA	CIMMYT	30MS	20MS	0	0	1	0	0	0	0	-
PASTOR/.../KARL	CIMMYT	10MR	5R	1	0	0	0	0	1	0	-
EMB16/.../3/LIRA	CIMMYT	10MR	5R	0	0	0	0	0	0	0	-
EMB16/CBRD//.../MCD/3 /LIRA	CIMMYT	10MR	5MR	0	0	1	0	0	0	0	-
AUS4930.6/.../4/ ZARGANA4	CIMMYT	20MR	5MR	0	0	0	1	0	0	0	-
AUS4930.7/2*PASTOR.../ KAUZ	CIMMYT	10MR	5R	1	0	0	1	0	0	0	-
AUS4930.7/.../5/TAM200/ KAUZ	CIMMYT	10MR	5R	1	0	0	1	0	0	0	-
338-K...ZARGANA-3	CIMMYT	20MS	5R	0	0	0	0	1	0	0	-
338-K...ZARGANA-3	CIMMYT	30MS	5R	0	0	0	1	0	0	0	-
338-K1-1//ANB...BCN	CIMMYT	10MR	0	1	0	0	1	0	0	0	-
DALNI...GAL VEZ87	CIMMYT	5R	5R	0	0	0	1	0	0	0	-
TREGO/.../KAUZ	CIMMYT	20MS	10MS	0	0	0	1	0	0	0	-
DIAMONDB...KAUZ	CIMMYT	10MR	5R	0	0	0	1	1	0	0	-
U11AGEC-1 (Krasnodar 99)	Russia - IWWIP	20MR	15MR	1	0	0	0	0	1	0	-
U11AGEC-2 (VEE...200/KAUZ)	IWWIP	5MS	5R	0	0	0	0	0	1	0	-
U11AGEC-3 (GAN...WRB860365)	IWWIP	0	0	0	0	0	1	0	1	0	-
U11AGEC-4 (GANSU170 WRB860365)	IWWIP	0	0	0	0	0	0	0	1	0	-
U11AGEC-5 (AMSE...F4105W2.1)	IWWIP	20MR	50S	1	0	0	0	0	1	0	-
U11AGEC-6 (ZOLOTA...MILAN)	IWWIP	0	5R	1	0	0	0	0	0	0	-
U11AGEC-7 (4WON-IR-.../MOS)	IWWIP	0	5R	0	0	0	0	0	1	1+	-
U11AGEC-8 (YMH/.../KAUZ)	IWWIP	20MR	5R	1	0	0	0	1	0	0	-
U11AGEC-9 (CADET.../CANON)	IWWIP	30MS	5R	0	0	0	0	0	1	0	1
U11AGEC-10 (AGRI/.../KAUZ)	IWWIP	0	5R	1	0	0	0	0	0	0	-
U11AGEC-11 (HK1/6/.../3NAI60)	IWWIP	0	5R	1	0	0	1	0	1	0	-
U11AGEC-12 (CATBIR...HE 1)	IWWIP	0	5R	0	0	0	1	0	0	0	-

Table continuation											
1	2	3	4	5	6	7	8	9	10	11	12
U11AGEC-13 (ZANDER...BCN)	IWWIP	30MS	5R	0	0	0	1	0	0	0	–
U11AGEC-15(MILAN..WEAVER)	IWWIP	0	5R	0	0	0	1	1	1	0	–
U11AGEC-16 (DORADE-...MLT)	IWWIP	40MS	5R	0	0	1	0	0	0	0	–
U11AGEC-17 (DORADE.../MLT)	IWWIP	20MS	5R	0	0	1	0	0	0	0	–
U11AGEC-21 (Drujba)	Russia	5MS	5R	0	0	0	1	0	1	0	–
U11AGEC-24 (PYN.../ Bluegil)	IWWIP	0	5R	0	0	0	0	1	0	0	–
U11AGEC-25 (4WON-IR...MOS)	IWWIP	0	5R	0	0	0	0	1	0	0	–
U11AGEC-26-(GRECUM BAU)	IWWIP	10MS	5R	0	0	1	0	0	0	0	–
U11AGEC-27 (PBII013...STAR)	IWWIP	0	5R	0	0	0	0	0	0	0	–
U11AGEC-28 (PYN...Bluegil)	IWWIP	0	5R	0	0	0	0	1	0	0	–
U11AGEC-29 (PYN/... Bluegil)	IWWIP	0	5R	0	0	0	0	1	0	0	–
U11AGEC-30 (PSK...MNCH)	IWWIP	10MS	5R	1	0	0	0	0	0	0	–
Egemen	KZ	10MS	20MS	1	0	0	0	0	0	0	–
Almaly	KZ	30MS	20MS	0	0	0	1	0	0	0	–
Bezostaya 1	KZ	40MS	30MS	0	0	0	1	0	0	0	–
Karasai	KZ	30MS	20MR	0	0	0	0	0	0	0	–
Maira	KZ	20MR	30MS	0	0	0	1	0	0	0	–
Mereke	KZ	40MS	30MS	0	0	0	1	0	0	0	–
Naz	KZ	40MS	30MR	0	0	0	0	0	0	0	–
Nureke	KZ	20MS	10MR	0	0	0	1	0	0	0	–
Ramin	KZ	30MS	10MR	0	0	0	1	0	0	0	–
Sapaly	KZ	30MS	20MR	0	0	1	0	0	0	0	–
Yubileynaya 60	KZ	40MS	20MS	1	0	0	0	0	0	0	–
Akdan	KZ	20MR	30MS	0	0	0	0	0	0	1+	–
Lyubava	RU	0	30MS	0	1	0	0	0	0	0	–
KSI 16/12	BEL	20MS	0	0	0	0	0	0	0	1+	–
KSI 27/12	BEL	20MS	0	0	0	0	0	0	0	0	–
113/00i-4	RU	5MR	10MR	0	1	1	0	0	0	0	–
Steklovidnaya 24 (local susceptible check)	KZ	30S	80S	0	0	0	0	0	0	0	–
Morocco (susceptible check)	Morocco	100S	100S	0	0	0	0	0	0	0	–

Note: *Origin include countries: KZ - Kazakhstan, RU – Russia, Bel – Belorussia, IWWIP – International Winter Wheat Improvement Program; **Field responses, max from three years, rust severity (%): MR – moderately resistant, MS – moderately susceptible, S – susceptible; “1”, “0”, “1+0” and “–” refer to having same polymorphic bands as, different polymorphic bands with the linked markers, heterozygote allele of gene, and no polymorphic bands, respectively.

(17.5%) (table). The disease severity in these entries to leaf rust was estimated between 0 – 30MS. Four genotypes, one CIMMYT, one IWWIP line, one Kazakh cultivar and one Belorussian line amplified four PCR products (200 bp and 180 bp) for resistant and susceptible alleles of *Yr10*, indicating that these four

entries appeared to be heterozygotes (Table). Disease severity in these entries to stripe rust was estimated between 0 – 5MR. Out of 62 genotypes tested for *Yr15* the expected PCR product was not amplified in any of the entries, excluding IWWIP line U11AGEC-9. The expected size of the fragment amplification for locus Xbarc8 coupled to resistant allele of *Yr15* gene is 250 bp (table). The disease severity in these entries was as 5R. To identify sources of *Sr* resistance genes PCR analysis was performed. The gene of *Sr22* is localized on the short arm of chromosome 7A. PCR analysis with primers to the *Xcfa2123* SSR locus located at a distance 6 of cM from the *Sr22* gene was done for identification of the *Sr22* carriers [14].

PCR analysis showed that of 38 studied entries, the fragment of DNA associated with gene *Sr22* with size 245 bp in 13 samples wheat was observed. Gene *Sr22* was identified in 5 Kazakhstani and in 8 Belarusian wheat entries.

Discussion. Utilization of foreign germplasms is the best way to solve the problem of development new rust resistant cultivars. Based on data from field test can be concluded that the most valuable sources, combined resistance to both leaf and stripe rust were 16 lines and cultivars (28.6%), including mainly entries from CIMMYT and IWWIP. Nineteen entries (30.6%) had high level of resistance to leaf rust in the field tests. Thirty-three entries (53%) were resistant to stripe rust in the field and have great effectiveness to control stripe rust. Application of molecular genetic markers allowed to identify efficient rust resistance genes in wheat cultivars and hybrids [15,6,16,17,18,4,19,20].

Under molecular screening in this study eight genes were detected: three seedling genes (*Lr10*, *Lr19*, *Lr26*) and three adult plant resistance genes (*Lr34*, *Lr37*, *Lr68*). Wheat accessions for the presence of two *Yr* genes – *Yr10* and *Yr15* was also screened. In our study using STS marker, F1.2245Lr10-6/r2, we detected the diagnostic fragment in 15 entries (22.6%). In moderately resistant lines (cultivars) carrying *Lr10* neither *Lr34* nor *Lr68* were detected, suggesting a combination of *Lr* genes different than *Lr10* + *Lr34* + *Lr68* associated with high or moderate level of resistance to leaf rust. Molecular data confirmed the detection of the *Lr19* diagnostic fragment only in two entries. *Lr19* in combination with other gene, *Lr26*, was more effective in the field. In our study marker analyses indicate that *Lr26/Yr9/Sr31* gene block was present in seven (11%) of tested cultivars (lines). Because of its agronomic benefits (yield increase, rusts and powdery mildew resistance), the chromosome 1R of rye (*Secale cereale* L.) has been widely used in wheat breeding.

Yessenbekova et al., 2014, identified *Lr34/Yr18* genes in 14 germplasms from CIMMYT and in 6 wheat entries from Kazakhstan. In our study, occurrence of adult plant resistance genes *Lr34* in 27 (44%) and *Lr68* in 11 entries (18%) was confirmed [21]. In present study combinations of *Lr34* with seedling resistance genes *Lr10* in five entries observed and provided moderate level of resistance. The same resistance was observed in entries with combination of APR *Lr68* with *Lr10*. The most level of resistance to leaf rust had the IWWIP line U11AGEC-11 with immune reaction to leaf rust. Of the 62 cultivars/lines studied, eight accessions (12.7%) had the *Lr37/Yr17/Sr38* resistance genes block. In our previous study it was found that *Lr34* provided some protection, *Lr37* occurred in cultivars/lines L-1090, Krasnovodapadskaya 210 and Madsen; *Lr19* and *Lr68* were present in cultivars Pallada and Yegemen [22,6,18].

Based on recent evaluation in China and Kazakhstan, [23,5,24] stripe rust resistance genes *Yr5*, *Yr10*, *Yr15* are still effective and could be useful in breeding programs. Out of 62 genotypes tested for *Yr15* the expected PCR product for locus Xbarc8 coupled to resistant allele of *Yr15* gene not amplified in any of the entries, excluding the IWWIP line U11AGEC-9.

So, in our study marker *Lr10* found in entries with moderate resistance and moderate susceptible reaction. Using molecular markers, the *Lr10* gene identified in 15 entries; the *Lr19/Sr25* gene, in two entries; the *Lr26/Yr9/Sr31* gene block, in seven entries; the *Lr34/Yr18/Pm38* gene block, in 27 lines; the *Lr68* gene, in seven entries; the *Yr10* gene, in four entries and *Yr15* gene, in one entry. Four CIMMYT lines and thirteen IWWIP lines were resistant to leaf/stripe rust, respectively. To identify sources of effective stem rust resistance *Sr22* gene PCR analysis with primers to the *Xcfa2123* SSR locus PCR analysis performed. The fragment of DNA associated with gene *Sr22* in 13 samples wheat observed. Gene *Sr22* was identified in five Kazakhstani and in eight Belarusian wheat entries.

Conclusion. Thus, due to the threat of disease epiphytotics, it is necessary to create new donors of stripe and leaf rust resistance and wheat breeding material based on them. We selected a number of wheat lines resistant to *P. recondita* f. sp. *tritici*, *P. striiformis* f.sp. *tritici* and *P. graminis* sp. *tritici*. The selected

material revealed the resistance to the Kazakh population of rusts. These genotypes were intensively involved in crosses in national breeding programs for wheat improvement. To accelerate the breeding process, we will continue selection of cultivars (lines) that are resistant to disease using molecular markers linked with this trait

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МОЛЕКУЛАЛЫҚ МАРКЕРЛЕРДІ ПАЙДАЛАНА ОТЫРЫП, ҚОҢЫР, САРЫ ЖӘНЕ САБАҚТЫ ТАТҚА ТӨЗІМДІ БИДАЙДЫҢ ФЕРМОПЛАЗМАЛАРЫН СӘЙКЕСТЕНДІРУ

Аннотация. Орталық Азия аймағы дүниежүзіндегі бидай өндірісі бойынша ең маңызды аймақтардың бірі болып табылады. Бидай 15 млн га егіледі, соның ішінде 5 млн га күздік және 10 млн га жаздық бидай. Қазақстан бидайды аса көп өндіруші, сонымен қатар экспорттаушы мемлекеттердің бірі болып табылады және Орталық Азияның азық-түлік қауіпсіздігін қамтамасыз етуде маңызды рөл атқарады. Қазақстандағы бидайдың негізгі аурулары болып қоңыр, сары және сабақты тат аурулары табылады, олар өнімнің сапасы мен өнімділігін төмендетіп, экономикалық шығынға әкеліп соқтырады. Төзімді ұрықтық плазманы халықаралық серіктестік шеңберінде қолдану, Қазақстандағы татқа төзімді жаңа сорттарды шығаруда маңызы болып табылады. Бұл зерттеуде Қазақстанда татқа төзімділікті жақсартуға бағытталған, генетикалық және селекциялық программаларда өнімнің құндылығын бағалау негізінде әртүрлі ұлттық және халықаралық питомниктердің күздік бидай ұрықтық плазмалары қолданылады. Дала жағдайындағы зерттеулердің мәліметтеріне сүйене отырып, қоңыр және сары татқа бірдей төзімділік танытқан, ең құнды көздер болып 16 линиялар мен сорттар (28,6 %) табылады, оларға негізінен CIMMYT және IWWIP үлгілері кіреді. Он тоғыз үлгі (30,6 %) дала жағдайындағы зерттеулер кезінде қоңыр татқа жоғары төзімділік көрсетті. Отыз үш үлгі (53 %) сары татты бақылауға эффективті болды. Молекулалық маркерлердің көмегімен бидайдың сары және қоңыр татына қарсы төзімділік гендері идентификацияланды. Молекулалық маркерлерді қолдана отырып бидайдың 62 сортын сегіз *Lr* және *Yr* төзімділік гендеріне молекулалық скрининг жүргізілді. Осы гендердің ішінде үш төзімділік гені өскін кезіндегі (*Lr10*, *Lr19*, *Lr26*) және 3 төзімділік гені (*Lr34*, *Lr37*, *Lr68*) ересек кезеңдегі төзімділікті қамтамасыз етеді. Сонымен қатар бидай үлгілерін *екі Yr гендеріне*, яғни *Yr10* және *Yr15* гендерін анықтау жұмыстары жүргізілді. *Lr10*, *Lr19/Sr25*, *Lr26/Yr9/Sr31*, *Lr34/Yr18*, *Lr37/Yr17/Sr38*, *Lr68*, *Yr10*, *Yr15* гендері үшін маркерлердің амплификацияланған фрагменттерінің көлемі 310 ж.н, 130 ж.н, 1100 ж.н, 150 ж.н, 262 ж.н, 385 ж.н, 200 ж.н және 250 ж.н құрады. Біздің зерттеуімізде бидай үлгілерінің 22 %-да қоңыр таттың *Lr10* төзімділік генімен байланысқан, STS F1.2245Lr10-6 / r2 маркерінің полиморфты фрагменті анықталынды. Молекулалық зерттеулердің мәліметтері мен татқа реакция негізінде, үлгілердің 3 %-да *Lr19/Sr25* төзімділік гендері, 11 %-да *Lr26/Sr31/Yr9/Pm8*, 43 %-да *Lr34/Yr18*, 12 %-да *Lr37/Yr17/Sr38* төзімділік ген кешендері, 17 %-да қоңыр таттың *Lr68* APR төзімділік гені мен 6 %-да сары татқа *Yr10* төзімділік гендері бар болуы мүмкін екендігі анықталынды. IWWIP тәлімбағының тек бір линиясынан сары таттың *Yr15* төзімділік гені анықталынды. Бидайдың сабақты татына қарсы зерттелген 38 үлгінің 13 линиясында *Sr22* генімен байланысқан, көлемі 245 ж.н. құрайтын ДНК фрагменттері анықталынды. Бұл ген 5 қазақстандық және 8 белорустық бидай үлгілерінде идентификацияланды. Алынған нәтижелер Қазақстанда татқа төзімді бидай сорттарын шығаруда қолданылады. Біздің зерттеулердің нәтижелері, молекулалық генетика әдістерін қолдану нәтижесінде Қазақстандағы селекциялық процесстерді жаңа ғылыми деңгейге шығаруға мүмкіндік береді.

Түйін сөздер: бидай, таттың түрлері, молекулалық маркерлер, төзімділік гендері

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ИДЕНТИФИКАЦИЯ ГЕРМОПЛАЗМА ПШЕНИЦЫ, УСТОЙЧИВОЙ К БУРОЙ, ЖЕЛТОЙ И СТЕБЛЕВОЙ РЖАВЧИНЕ С ИСПОЛЬЗОВАНИЕМ МОЛЕКУЛЯРНЫХ МАРКЕРОВ

Аннотация. Регион Центральной Азии является одним из самых важных пшеничных районов в мире. Пшеница выращивается на 15 млн га, в том числе 5 млн га озимой и 10 млн га яровой пшеницы. Казахстан является одним из крупнейших производителей и экспортеров пшеницы и играет важную роль в обеспечении продовольственной безопасности Центральной Азии. Бурая, желтая и стеблевая ржавчина пшеницы являются основными заболеваниями пшеницы в Казахстане, которые снижают урожайность и качество и наносят значительный экономический ущерб. Использование устойчивой зародышевой плазмы в рамках международного сотрудничества играет важную роль в разработке новых устойчивых к ржавчине сортов для Казахстана. В этом исследовании использовалась зародышевая плазма озимой пшеницы из различных национальных и международных питомников для оценки их ценности для генетических и селекционных программ, направленных на улучшение устойчивости пшеницы к ржавчине в Казахстане. Основываясь на данных полевых экспериментов, наиболее ценными источниками, комбинированной устойчивости как к бурой, так и к желтой ржавчине, были 16 линий и сортов (28,6%), включая в основном образцы из CIMMYT и IWWIP. Десять образцов (30,6%) имели высокий уровень устойчивости к бурой ржавчине в полевых испытаниях. Тридцать три образца (53%) были эффективны для контроля желтой ржавчины. Проведен молекулярный скрининг 62 сортов и линий пшеницы с использованием молекулярных маркеров, разработанных для восьми *Lr* и *Yr* генов устойчивости к ржавчине пшеницы. Три из этих генов (*Lr10*, *Lr19*, *Lr26*) проростков и три гена (*Lr34*, *Lr37*, *Lr68*) контролируют устойчивость к бурой ржавчине на стадии взрослых растений. Кроме того, был проведен скрининг на наличие двух *Yr* генов устойчивости (*Yr10* и *Yr15*). Размеры ПЦР-продуктов маркеров составляет 310 п.н, 130 п.н, 1100 п.н, 150 п.н, 262 п.н, 385 п.н, 200 и 250 п.н для *Lr10*, *Lr19/Sr25*, *Lr26/Yr9/Sr31*, *Lr34/Yr18*, *Lr37/Yr17/Sr38*, *Lr68*, *Yr10* и *Yr15* соответственно. В нашем исследовании 22% образцов пшеницы имели полиморфный фрагмент маркера F1.2245Lr10-6 / r2, сцепленный с геном устойчивости к бурой ржавчине *Lr10*. На основании реакции к ржавчине и данных молекулярных анализов было обнаружено, что 3% образцов, возможно, имеют гены устойчивости *Lr19/Sr25*, 11% образцов – комплекс генов устойчивости *Lr26/Sr31/Yr9/Pm8*, 43% образцов – комплекс генов устойчивости *Lr34/Yr18* APR, 12 % образцов – комплекс генов устойчивости *Lr37/Yr17/Sr38*, 17% образцов – APR ген устойчивости к бурой ржавчине *Lr68* и 6% образцов – ген устойчивости к желтой ржавчине *Yr10*. Только одна линия из питомника IWWIP показала наличие гена устойчивости к полосовой ржавчине *Yr15*. Из 38 исследованных образцов *Sr22* в 13 линиях пшеницы обнаружен фрагмент ДНК с размером 245 п.н., сцепленный с геном *Sr22*. Этот ген был идентифицирован в 5 казахстанских и 8 белорусских пшеничных записях. Полученные результаты используются в Казахстане для создания сортов пшеницы, устойчивых к ржавчине. Результаты нашего исследования создают возможности для перевода селекционного процесса в Казахстане на новый научный уровень благодаря применению методов молекулярной генетики.

Ключевые слова: пшеница, виды ржавчины, молекулярные маркеры, гены устойчивости.

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