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GENETIC STRUCTURE OF RUSSIAN AND KAZAKHSTANI LEAF RUST CAUSATIVE AGENT *Puccinia triticina* Erikss. POPULATIONS AS ASSESSED BY VIRULENCE PROFILES AND SSR MARKERS

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Abstract

Leaf rust caused by *Puccinia triticina* Erikss. is an economically significant disease of spring wheat in the West-Asian Russia and Northern Kazakhstan. Successful wheat breeding for leaf rust resistance necessitates characterization of *Lr* gene effectiveness, the impact of new wheat varieties on the pathogen virulence, and isolation between populations of *P. triticina*. Until now, nobody used a uniform infectious material in *P. triticina* population study, as it was collected from a different set of varieties in each region. Thence, the virulence assessment data were significantly influenced by the effects of host plants. We were the first to compare the structure of *P. triticina* populations in the West-Asian Russia and Northern Kazakhstan on virulent and molecular genetic properties, using as a source of infectious material a single set of the wheat samples dated 2016 that were tested there within the framework of the Kazakhstan-Siberian Spring Wheat Improvement Network Program (KASIB). Ninety one single-pustule isolates have been tested in the virulence analysis, including 13 isolates from Chelyabinsk, 28 from Omsk, 6 from Kurgan, 16 from Akmolinskaya, 16 from Pavlodarskaya, and 12 from Karagandinskaya provinces. Eleven phenotypes of virulence have been identified with the use of 20 isogenic Thatcher lines with *Lr* genes (Tc*Lr*). The similar phenotypes were found on cultivars Duat, Tertsia, Omskaya 35, Pamyati Azieva, Saratovskaya 29, Chebarkulskaya 3 and line Eritrosperum 85-08. The Russian pathotypes found on variety Astana and lines Lutescens 1003 and Lutescens 6/04-4 differed from Kazakhstan ones in virulence to Tc*Lr11*, and those colonizing variety GVK 2074/4 and line Lutescens 715 differed in avirulence to Tc*Lr18*. More significant differences in the virulence rang between regional populations have been observed on the line Lutescens 34/08-1 and the variety Rodnik. All studied *P. triticina* isolates were avirulent to Tc*Lr19*, Tc*Lr24* and virulent to Tc*Lr3a*, Tc*Lr3bg*, Tc*Lr3ka*, Tc*Lr14a*, Tc*Lr14b*, Tc*Lr16*, Tc*Lr17*, Tc*Lr30*. The variation in virulence frequencies was observed on the wheat lines with the genes *Lr1*, *Lr2a*, *Lr2b*, *Lr2c*, *Lr9*, *Lr15*, *Lr18*, *Lr20*, and *Lr26*. Virulence to *Lr9* was found in the pathogen populations collected from the wheat samples Duet, Tertsia, Chebarkulskaya 3, Lutescens 34/08-19 carrying this gene. The differences in populations on virulence were found using the indices of genetic distances Nei D and Fst, the analog for binary data in AMOVA. The Nei index values indicated a

high similarity between the majority of the Northern-Kazakhstani and Russian populations, except of those from Chelyabinskaya and Karagandinskaya provinces. According to the Fst index, the high similarity was found between the populations from Omsk, Kurgan and Northern Kazakhstan. The Chelyabinsk population appeared to be close to the Kurgan but varied from others. In the test, a total of 46 isolates and 9 genotypes have been identified using 12 SSR markers. In this, 21 polymorphic alleles were found in the studied set. The Nei and Fst indices revealed the moderate differences between the Chelyabinsk and Karaganda populations and a high similarity between other populations studied. The current survey defined the high similarity of leaf rust pathogen populations in West-Asian Russia and Northern Kazakhstan, assuming the existence of a single fungus population in the studied territories. In order to prevent the *P. triticina* epiphytoties on the adjacent territories of Russia and Kazakhstan, the constant updating of wheat varieties and higher genetic diversity are strongly recommended. In addition, the varieties should be grown according to the «mosaic distribution» scheme using optimal areas for genetically homogeneous varieties.

Keywords: *Puccinia triticina* Erikss., leaf rust, common wheat, populations, virulence, *Lr*-genes, SSR markers

Spring wheat is the main cereal crop in the Western Siberia, the Urals and Northern Kazakhstan. Leave rust (causal agent *Puccinia triticina* Erikss.) is the most prevailing and harmful disease limiting wheat yields in these regions [1-3]. With its epiphytotic development, wheat losses can reach 15-30 % [4].

Most of productive varieties cultivated in Western Siberia, the Urals and Northern Kazakhstan are susceptible to leaf rust with varying degree [2, 3]. Shuttle breeding within the framework of the CIMMYT (International Maize and Wheat Improvement Center) Kazakhstan-Siberian Spring Wheat Improvement Network Program (KASIB), leverages global genetic resources to improve the carried out breeding programs efficiency, including on leaf rust resistance [1, 5].

Strategies for genetic wheat protection are based on the creation and introduction of leaf rust resistant varieties, which requires data about the resistance genes efficiency to its causal agent (*Lr*-genes), virulence changes in the pathogen populations under the influence of new varieties, isolation between populations and the potential genes flow between them. In structure of pathogen population, frequency of virulence genes is the most important criterion, but does not always characterize it adequately [7, 8], because the concentration of virulence genes in the fungus population is closely linked to the selective pressure of cultivated wheat varieties. Despite this, in Russia and abroad most population studies of the causatvel agent of leaf rust were performed on contagious material collected from a different set of varieties in each region [9, 10].

Application of the selectively neutral molecular markers to compare DNA polymorphisms of a pathogen makes it possible to refine and to add the results of virulence analysis. Such markers are indispensable in the pathways researches of organism migration, formation of the uniform epidemiologic regions and emergence of areas [7]. Molecular markers (RAPD — random amplification of polymorphic DNA, AFLP — amplified fragment length polymorphism, SSR — simple sequence repeat) have been involved in *Puccinia triticina* study since the mid-1990s [11], with the most extensive use of microsatellites. Molecular markers were used to describe the structure of *P. triticina* populations in the North and South American continents, in Western Europe, in the Middle East, Central Asia and the North Caucasus [4, 8, 12].

We were the first to use infectious material which was collected from a single set of wheat varieties at several geographical locations, which reduces the selective influence of host plants on the results of virulence evaluation. In this case, a high population similarity to the causative agent of leaf rust in the Urals, Western Siberia and Northern Kazakhstan is identified, which confirmes the suggestion of the uniform fungus population in these territories.

The aim of the work is to analyze the genetic structure of *Puccinia triticina* populations in the West-Asian Russia and Northern Kazakhstan for viru-

lence and microsatellite loci.

Techniques. Samples of *Puccinia triticina* Erikss. Were collected in 2016 on 15 wheat varieties, which were studied within the frame of the KASIB program in the Chelyabinsk, Omsk, Kurgan regions (Russia) and Akmolinskaya. Karagandinskaya, Pavlodarskaya regions (Kazakhstan). In 2016, in all these regions, the weather conditions was favorable for leaf rust development, prevalence of the infection among the varieties varied from 0 to 100 %.

The isolates were cultured by laboratory method as per the description [13]. Virulence of 91 single-pustule isolates (2-3 from each studied wheat sample) was analyzed with the use of 20 almost isogenic Thatcher lines with *Lr* genes (Tc*Lr*). For this, 3 grains of each Tc*Lr* line were seeded in the soil. Seedlings (10-14-day old, leaf 1 phase) were inoculated with spore suspension (10^6/ml) and placed in a Versatille Environmental Test Chamber MLR-352H (SANYO Electric Co., Ltd., Japan) at 22 °C and 75 % humidity. On day 10 the results were estimated according to E.B. Mains and H.S. Jackson scale range [14]: 0 — no signs, 0; — necrosis without pustules, 1 — very small pustules surrounded by necrosis, 2 — medium size pustules, surrounded by necrosis or chlorosis, 3 — medium size pustules without necrosis, 4 — large pustules without necrosis, X — different types of pustules on one leaf, chlorosis and necrosis are presented (plants with the X type reaction considered as susceptible).

Virulence phenotypes were classified according to the North American system [15]. For this purpose, 20 Tc*Lr* lines were divided into five sets: 1 — Tc*Lr1*, Tc*Lr2a*, Tc*Lr2c*, Tc*Lr3a*; 2 — Tc*Lr9*, Tc*Lr16*, Tc*Lr24*, Tc*Lr26*; 3 — Tc*Lr3ka*, Tc*Lr11*, Tc*Lr17*, Tc*Lr30*, 4 — Tc*Lr2b*, Tc*Lr3bg*, Tc*Lr14a*, Tc*Lr14b*; 5 — Tc*Lr15*, Tc*Lr18,* Tc*Lr19*, Tc*Lr20*. The alphabetic phenotype code was determined using the software package VAT (Virulence Analysis Tool; https://en-lifesci.tau.ac.il/profile/kosman/vat). Indices of interpopulation differences by M. Nai (Nei D) and Φ_{PT} (analog of F_{st} for binary data in AMOVA, analysis of molecular variance) were calculated using the GenAlEx (Genetic analysis in Excel 6.5; http://biology.anu.edu.au/GenAlEx).

A total of 46 single pustule *P. triticina* isolates, including Chelyabinskaya 7, Kurganskaya 5, Omskaya 14, Akmolinskaya 8, Pavlodarskaya 8 and Karagandinskaya 4 (1-2 isolates from each studied wheat sample) were used for SSR analysis. Procedure of pathogen collection was similar to described by J.A. Kolmer et al. [4]. DNA was isolated according to A.F. Justesen et al. technique [16]. The polymorphism of 12 microsatellite loci (PtSSR50, PtSSR55, PtSSR61, PtSSR91, PtSSR92, PtSSR151, PtSSR152, PtSSR158, PtSSR161, PtSSR164, PtSSR173, and RB35) was determined. The used SSR markers are designed to assess genetic diversity of *P. triticina* [4, 8, 10, 12]. PCR protocols and primer sequences are presented in the original papers [17, 18] (ABI Prism 3500 genetic analyzer, Applied Biosystems, USA; Hitachi, Japan). The SSR allele sizes were determined with GeneMapper 4.1 software. To assess the effect of cultivated varieties on the pathogen virulence, *Lr* genes were identified in wheat samples. Markers of genes *Lr1* (WR003), *Lr9* (SCS5), *Lr10* (FI.2245/*Lr10-6*/r2), *Lr26* (SCM 9) and *Lr34* (L34DIN9/Lr34Plus) were used [19]. PCR protocols and primers for marking varieties and lines were as per original works cited by A. Serfling et al. [19], the PCR was carried out with a C1000TM Thermal Cycler (Bio-Rad, USA).

Statistical results processing of SSR analysis, including P calculation, was carried out with GenAlEx 6.5 software (genetic analysis in Excel, 6.5; http://biology.anu.edu.au/GenAlEx). Intrapopulation genetic diversity of microsatellite loci in fungi was described by the following indices: the mean number of alleles per locus (Na), number of effective alleles (Ne), expected (H_e) and

1. Wheat varieties infected by *Puccinia triticina* Erikss., with identified phenotypes of virulence and SSR genotypes of the pathogen in different geographic populations (2016)

Sample	Russia						Kazakhstan					
	Chelyabinskaya Province		Omskaya Province		Kurganskaya Province		Akmolinskaya Province		Pavlodarskaya Province		Karagandinskaya Province	
	I, %	P/SSR	I, %	P/SSR	I, %	P/SSR	I, %	P/SSR	I, %	P/SSR	I, %	P/SSR
Astana2	50S	–	40S	TGTTR/D	70S	TGTTR/D	90S	TGPTR/D	50S	TGPTR/G	45S	–
ГВК 2074/4	100S	–	80S	THTTM/D	70S	–	50S	–	20S	THTTR/B	10S	THTTR/A
Duat	50S	PQTKH/F	50S	PQTKH/F	–	–	–	–	–	–	–	–
Lutescens 1003	20MS	–	100S	TGTTR/F	40S	–	90S	TGPTR	50S	TGPTR/G	30S	–
Lutescens 1003	10MR	–	5MR	–	20S	PQTKH/B	5S	TQTTR/B	20S	–	0	–
Lutescens 1003	50S	THTTR/B	10MR	THTTR/B	10S	–	5S	THPTR/B	20S	THPTR/B	0	–
Lutescens 1003	50S	–	100S	TGTTM/H	60S	–	70MS	TGTTR/B	50S	–	20S	TGTTR/C
Omskaya 35	50S	–	100S	TGTTR/F	70S	TGTTR/G	70S	TGTTR/C	50S	–	–	–
Memory of Aziev	50S	–	100S	TGTTR/C	80S	TGTTR/C	90S	TGTTR/C	40S	–	40S	–
Rodnik	20S	THTTR/A	100S	TGTTR/A	30S	–	50S	TGTTR/F	30S	MGTKH/B	10S	–
Saratovskaya 29	50S	–	100S	TGTTR/F	50S	–	100S	–	30S	TGTTR/F	70S	–
Stepnaya 53	80S	–	80S	TGTTM/B	60S	–	50S	–	50S	TGTTM/B	5MS	TGPTR/C
Terzia	20MS	–	40S	–	60S	TQPTR/B	90S	–	40S	TQPTR/D	–	–
Chebarkulskaya 3	50S	CQPKG/E	5S	CQPKG/E	60S	–	10S	CQPKG/E	20S	–	0	–
Eritrosperum 85-08	50S	THTTR/B	5MR	THTTR/B	0	–	10S	–	10S	–	0	–

Примечание. I — infestation, P/SSR — phenotype/SSR genotype S — response type 3-4 points, MR — response type 1-2 points, MS — response type X. Dashes in the table mean that samples were not analyzed..

observed (H_o) heterozygosity, fixation index (F) and Shannon's index (I). Genetic differences between the populations were determined by Nei D indices on M. Nei and F_{st}, which were calculated by AMOVA (GenAlEx) (for 999 permutations). Dendrogram of the genetic similarity in virulence and microsatellite loci among regional populations was constructed in the NTSYSpc 2.21 software for the Φ_{PT} and F_{st} indices. The data of microsatellite analysis and virulence assessment were compared using the Mantel test based on the distances between the relevant matrices for the virulence and SSR markers (for F_{st} and Nei D indices).

Results. Ecogeographic stude]y of wheat samples in the Western Siberia, the Urals and Kazakhstan are held annually according to the KASIB program to realize population researches of the causative agent of leaf rust on a vast territory (over 1000 km) and a single set of varieties, This excludes the selective pressure of the host plant which may interfere with the results of virulence analysis.

Using 20 Tc*Lr*-lines, 11 virulence phenotypes were detected (Table 1). Phenotypes identical in all locations were detected on varieties Duat (PQTKH, av.: Tc*Lr2a*, Tc*Lr2b*, Tc*Lr15*, Tc*Lr19*, Tc*Lr24*, Tc*Lr26*), Terzia (TQPTR, av.: Tc*Lr18*, Tc*Lr19*, Tc*Lr24*, Tc*Lr26*), Omskaya 35 (TGTTR, av.: Tc*Lr9*, Tc*Lr19*, Tc*Lr24*, Tc*Lr26*), Memory of Aziev (TGTTR), Saratovskaya 29 (TGTTR), Chebarkulskaya 3 (CQPKG, av.: Tc*Lr1*, Tc*Lr2a*, Tc*Lr2b*, Tc*Lr2c*, Tc*Lr11*, Tc*Lr15*, Tc*Lr19*, Tc*Lr20*, Tc*Lr24*, Tc*Lr26*) and on Eritrosperum 85-08 (THTTR, av.: Tc*Lr9*, Tc*Lr19*, Tc*Lr24*) line. Russian and North Kazakhstan isolates obtained from Astana 2, Lutescens 1003 (TGPTR, TGTTR) and Lutescens 6/04-4 (THPTR, THTTR) samples differed in virulence to Tc*Lr11,* and from ГВК 2074/4 (THTTM, THPTM) lines and Luteszens 715 (TGPTM, TGTTM) — to Tc*Lr18.* Karaganda isolates (TGPTR) from Stepnaya 53 variety differed from the Omsk and Pavlodar isolates (TGTTM) from the same varieties in virulence to Tc*Lr18* and avirulence to Tc*Lr11.*

More significant differences in the virulence spectrum between geographical populations were observed on line Lutescens 34/08-19 (PQTKH and TQTTR) and Rodnik variety (THTTR, MGTKH, TGTTR). These samples are less affected by pathogen (Lutescens 34/08-19 — from 0 to 20 %, Rodnik — from 10 to 50 %) then others studied (see Table 1). Despite some differences between the Russian and Kazakh populations, which were collected from several wheat samples, in general, we did not observe significant changes in the detected virulence spectrum of the pathogen. The phenotype detected in Chebarkulskaya variety 3 (CQPKG) has 10 alleles of virulence, that is less than in other wheat samples where the number of identified phenotypes is from 12 (MGTKH) to 17 (TNTTR).

Using PCR technique, genes *Lr26* (GVK 2074/4, Erythrosperum 85-08), *Lr9* and *Lr10* (Duet), *Lr9* and *Lr34* (Lutescens 34/08-19), *Lr26* and *Lr1* (Lutescens 6/04-4), *Lr10* and *Lr34* (Omskaya 35), *Lr*10 (Memory of Aziev, Saratovskaya 29, Stepnaya 53), *Lr1* and *Lr10* (Rodnik), *Lr9* (Tertsia, Chebarkulskaya 3) were identified in infectious samples. Genes *Lr1*, *Lr9*, *Lr10*, *Lr26* and *Lr34* are ineffective in Western Siberia, the Urals and Northern Kazakhstan conditions [4, 6, 12]. Meanwhile, their specific combinations can enhance field resistance of wheat [19], which was observed in this experiment for Lutescens line 34/08-19 with *Lr9* and *Lr34* genes (see Table 1).

Lr19 and *Lr24* genes showed high efficiency against Russian and Kazakh pathogen populations (Table 2), while *Lr3a*, *Lr3bg*, *Lr3ka*, *Lr14a*, *Lr14b*, *Lr16*, *Lr17* and *Lr30* genes were widespread and absolutely ineffective.

In testing on Tc*Lr1*, Tc*Lr2a*, Tc*Lr2b*, Tc*Lr2c*, Tc*Lr9*, Tc*Lr15*, Tc*Lr18*,

Tc*Lr20*, Tc*Lr26* lines, the virulence frequency varied among isolates. Virulence to *Lr9* line was found in subpopulations collected on wheat samples with this gene (Duet, Tercia, Chebarkulskaya 3, Lutescens 34/08-19), to *Lr26* — in subpopulations from HVA lines 2074/4, Lutescens 6/04-4 and Erythrospermum 85-08 carrying *Lr26* gene. In general, the differences between regional populations in frequency of virulence to Tc*Lr9* and Tc*Lr26* depend on the presence of infectious material from the wheat plants carrying these genes. All isolates which were virulent to *Lr9* lines were avirulent to *Lr26* lines. This should suggests that the combination of *Lr9* and *Lr26* genes can provide effective protection of wheat plants against leaf rust, as observed for the genes combination *Lr19* + *Lr26*, *Lr19* + *Lr37* and *Lr19* + *Lr25* [20]. Avirulence to Tc*Lr20* was detected only in samples collected from Chebarkulskaya 3 variety. Certain differences between Russian and Kazakhstani populations were observed for virulence to Tc*Lr11*, i.e. the virulence was more often in case of *Lr11* in Russian populations.

2. Frequency (%) of *Puccinia triticina* Erikss. isolates virulent to Tc*Lr* wheat lines in the studied geographical populations (2016)

Thatcher line with *Lr*-gene	Russia			Kazakhstan		
	1	2	3	4	5	6
Tc*Lr19*. Tc*Lr24*	0	0	0	0	0	0
Tc*Lr1*	76.9	92.9	100	93.8	100	100
Tc*Lr2a*	46.2	85.7	80.0	93.8	87.5	100
Tc*Lr2b*	76.9	92.9	80.0	93.8	87.5	100
Tc*Lr2c*	76.9	92.9	100	93.8	87.5	100
Tc*Lr9*	23.8	14.3	40.0	18.8	12.5	0
Tc*Lr11*	76.9	92.9	80.0	50.0	50.0	62.5
Tc*Lr15*	46.2	85.7	80.0	93.8	87.5	100
Tc*Lr18*	100	78.6	100	100	87.5	100
Tc*Lr20*	76.9	92.9	100	93.8	100	100
Tc*Lr26*	46.2	21.4	0	12.5	25	25
Tc*Lr3a*. Tc*Lr3bg*. Tc*Lr3ka*. Tc*Lr14a*.Tc*Lr14b*. Tc*Lr16*.						
Tc*Lr17*. Tc*Lr30*	100	100	100	100	100	100
Number of isolates	13	28	6	16	16	12

Note. 1 — Chelabinskaya Province, 2 — Omskaya Province, 3 — Kurganskaya Province, 4 — Shortadinskaya Province, 5 — Pavlodarskaya Province, 6 — Karagandinskaya Province.

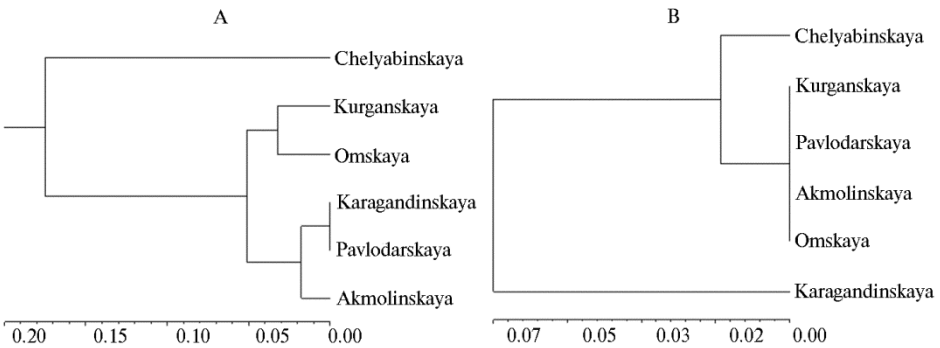

Fig. 1. UPGMA-dendrogram of genetic distances between *Puccinia triticina* Erikss populations for virulence (A) **and SSR markers** (B) (by F_{st}).

By the Nei D index, most of Russian and Kazakhstani populations are highly similar (Nei D = 0.02-0.7). The exceptions are Chelyabinsk and Karaganda populations (Nei D = 0.12). Index Φ_{PT} indicates a high similarity between Omskaya, Kurganskaya and Kazakhstanskaya populations. Chelyabinskaya is similar to Kurganskaya and differes from other studied populations (Fig. 1, A).

Analysis of polymorphism of 12 microsatellite loci detected 9 genotypes (4 in the Chelyabinskaya population, 4 in the Kurganskaya, 7 in the Omskaya, 4 in

the Pavlodarskaya, 5 in the Akmolinskaya, 2 in the Karagandinskaya) (see Table 1). Most of the genotypes belong to two and more populations, and only two are unique (one identified on Lutescens 715 line in Omskaya population, the other on Omskaya 35 variety in Kurganskaya population). A total of 21 polymorphic alleles are found. For most loci, two polymorphic alleles are identified, except for PtSSR55, PtSSR91 and PtSSR164 loci which are monomorphic (Fig. 2 at http://www.agrobiology.ru). We did not find alleles specific for a particular population. Note, we performed this study on ABI Prism 3500 genetic analyzer in which, unlike 4200 DNA Analyzer or 4300 DNA Analyzer (LI-COR, USA) used for similar purposes by other scientific groups [3, 12, 17, 18], sample preparation and analysis are fully automated that, with the same resolution, increases determination accuracy and data comparability.

Genetic diversity of *P. triticina* isolates on microsatellite loci are similar for all studied populations, except Karagandinskaya, which could be due to its low representation in the analysis (4 isolates). The mean number of alleles per locus (Na) varied from 1.2 in the Karagandinskaya population to 1.6-1.7 in the other studied, the number of effective alleles (Ne) was from 1.1 to 1.3-1.4, respectively. The observed heterozygosity (H_o) was higher than expected (H_e) for the Chelyabinskaya, Kurganskaya, Pavlodarskaya and Omskaya populations, which was confirmed by negative values of the fixation index (F). Values of H_o and H_e for Karagandinskaya and Akmolinskaya populations were similar (0.06 and 0.07; 0.16 and 0.19, respectively). Shannon index (I) was identical for all populations (0.3), except Karagandinskaya (0.1).

According to F_{st} index of genetic variations, the most of the studied populations, except for Chelyabinskaya and Karagandinskaya, do not differ for SSR markers ($F_{st} = 0.21$, $P = 0.003$) (see Fig. 1, B). We also obtained similar results by Nei D parameter (0.01-0.03 and 0.07, respectively). The identified differentiation between the Chelyabinskaya and Karagandinskaya populations can be explained by the contrast of wheat varieties from these two regions for *Lr*-genes, geographical remoteness (over 1000 km) of the populations and regional differences in climatic conditions. In the Karaganda region, leaf rust is noted sporadically in years favorable for the development of the pathogen. In the Chelyabinsk region, the disease occurs almost every year and belongs to a potentially dangerous group.

In whole, in comparing Russian and Kazakhstani populations, the indices of genetic distances indicate a high similarity (Nei $D = 0.007$, $F_{st} = 0.003$, $P = 0.31$), which agrees with the results of the earlier analysis of virulence [10, 20]. The low value of genetic divergence index (F_{st}) suggests the existence of a gene flow between the studied populations.

For all the populations except for Chelyabinskaya, the differentiation by virulence and microsatellite loci is quite close in pattern, i.e. by Mantel test for Nei D $r = 0.88$ ($P < 0.001$), for F_{st} $r = 0.43$ ($P < 0.001$). The similarity of Chelyabinskaya population with Akmolinskaya and Pavlodaraskaya populations by microsatellite loci is significantly higher than by virulence.

The tight connections among the West Asian Russian and North Kazakhstan populations indicate the existence of a single fungus population in these regions. Such results are in line with the reports of other researchers and the data obtained earlier [4, 10, 12, 21]. There are several assumptions on the emergence of the infection in the west of Asian Russia and Kazakhstan. The Ural Mountains are a geographical obstacle to spores spread from Europe to Asia, and the local direction of air flows is a physical obstacle [21]. It is shown [22] that the probability of spore migration from the North Caucasus territory to Kazakhstan is extremely

small, since the cyclone operating between the Caspian and Aral Seas and the anticyclone running down from the north along Western Siberia hinder the penetration of air mass. The diseases epiphytoty in Western Siberia and the Urals vs. its depression in the North Caucasus [21, 22], as well as the absence of isolates with virulence to *Lr9* in the North Caucasian populations [23] vs. their high frequency in Asian can be as a confirmation.

According to the opinion of other researchers, the epiphytotic development of leaf rust in Western Siberia and Northern Kazakhstan occurs only due to infection from the southern or southwestern regions of the European Russia [24]. The pathogen transfer from the sowings of the Middle Volga and western regions of Kazakhstan is not excluded [24, 25]. Thus, additionally to the exogenous infection in Western Siberia, there are its own sources independent of European ones [21, 22, 24]. Vegetation of the intermediate host plant *Isopyrum fumarioides* in the region can contribute to a full cycle of the pathogen development. It is reported [26] about the higher plasticity of the *P. triticina fungus* on *I. fumarioides* species**,** compared to the form affecting *Thalictrum* spp., which increases the competitiveness of local isolates. Affected crops of winter and spring wheat in the west of the republic and in neighboring regions of Russia can be the source of infection for Northern Kazakhstan. Favorable conditions for air infection transfer are created due to the prevalence of western winds during the growing season [27]. According to other data [12, 28], the contamination of spring wheat in Northern Kazakhstan occurs during the spore transfer from cultivated winter wheat by air from the southern part of the republic due to the lack of geographic barriers between the north and south of Kazakhstan.

Thus, using the approach which reduces the influence of the host plant on the estimates of virulence of the leaf rust causative agent, we have shown the high similarity of *Puccinia triticina* Erikss. populations in the Urals, Western Siberia and northern Kazakhstan. These data prove the assumption about existence of a single pathogen population in these regions. To prevent epiphytoty of *Puccinia triticina* on the adjacent territories of Russia and Kazakhstan, it is recommended to update constantly the stock of wheat varieties, to improve genetic diversity, to use mosaic sowings of varieties and optimal areas occupied by genetically homogeneous varieties.

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