DIVERSITY OF WINTER COMMON WHEAT VARIETIES FOR RESISTANCE TO LEAF RUST CREATED IN THE V. M. REMESLO MYRONIVKA INSTITUTE OF WHEAT

Hanna Kovalyshyna, Yuliia Dmytrenko, Oksana Tonkha, Oleksandr Makarchuk, Oleksandr Demydov, Oleksandr Humeniuk, Natalia Kozub, Anatolii Kareliv, Igor Sozinov, Mikhailo Mushtruk

ABSTRACT

The results of the investigations of resistance winter common wheat varieties to leaf rust are given. The high resistance to the pathogen model varieties that contain resistance genes: Lr9, Lr19, Lr37, Lr42 + Lr24, Lr43 (Lr21 + Lr39) + Lr24, Lr9 + Lr26, Lr10 + Lr24 are ascertained. The genes Lr13, Lr34, Lr37 in combination with other resistance genes provides long-term protection to leaf rust wheat. Winter wheat varieties, created at the V. M. Remeslo Myronivka Institute of Wheat, contain resistance genes Lr23, Lr24, Lr26, Lr34. The varieties Vesta, Snizhana, Demetra are protected by the resistance genes Lr26 + Lr34, variety Zolotokolosa – Lr24 + Lr34, Ekonomka – Lr3 + Lr26, Myronivska storicna – Lr3 + Lr23 + Lr10 + Lr26. The allele Lr34 (+) is contained in varieties: Kryzhynka, Vesta, Snizhana, Volodarka, Demetra, Vdiachna, Pamiati Remesla, Sviatkova, Podolianka, Berehynia myronivska, MIP Dniprianka, and Balada myronivska. Sustainability is an important element of an integrated system of plant protection against many diseases, and to ensure increased yields it is necessary to create and distribute sustainable varieties that will be an environmentally promising way to develop the agro-industrial complex of Ukraine.

Keywords: common winter wheat; breeding; variety; resistance; rust; genes

INTRODUCTION

The leaf rust (Puccinia recondita f. sp. tritici Rob. Ex Desm.) is one of the most common and harmful wheat diseases. The disease leads to significant losses of grain yield (Novohatka, 1979). The level of wheat yields loss to 30% during the epiphytotic rust according to the Food and Agriculture Organization of the United Nations (FAO) (El-Khoury, 2009). The population of the pathogen Puccinia recondita differs from the high adaptive ability. High variability virulence of the fungus leads to the accumulation of pathogens that capable of the genes of wheat resistance (Palamarchuk et al., 2019).

The most justified, economically sound, and environmentally safe method of fighting to disease is to creating resistance varieties. The effectiveness of breeding to rust resistance can be improved by using different Lr – genes (Zheplinska et al., 2019).

More than 90 resistance genes to the rust-leaf pathogen are identified and characterized by chromosomal localization and efficiency at the wheat genome and its relatives (Kovalyshyna and Dmytrenko, 2017), which information is collected annually and is published in the Catalogue of gene symbols for wheat (McIntosh et al., 2003a; McIntosh et al., 2007b; McIntosh et al., 2008c; McIntosh et al., 2009d; McIntosh et al., 2010e; McIntosh et al., 2011f; McIntosh et al., 2012g; McIntosh et al., 2014h; McIntosh et al., 2016i; McIntosh et al., 2017j; McIntosh et al., 2018k; McIntosh et al., 2019i). Most genes of resistance to rust causative agents identified in cultivars of wheat descend from their wild relatives. Thus, according to the information provided in the catalogs of genetic symbols, about half of the known resistance genes to leaf rust are allogenic, introgressive into Triticum aestivum species from different types of wheat, aegilops, wheat grass, and others (Leonova et al., 2013). The almost all effective rust resistance genes at the territory of Ukraine, except Lr10 and Lr23, are allogenic, transferred to Triticum aestivum from other species: Aegilops speltoides – Lr28, Lr35, Lr36, Lr47, Lr51, Lr66; Aegilops tauschii – Lr1, Lr21, Lr22a, Lr32, Lr39, Lr42; Triticum timopheevi – Lr18, Lr50; Thinopyrum elongatum – Lr19, Lr29, Lr24; Secale cereale – Lr25, Lr26, Lr45; Aegilops umbellulata – Lr19, Lr76; Triticum spelta – Lr44, Lr63, Lr71; Triticum dicoccoides – Lr53, Lr64; Aegilops triuncialis – Lr58, Lr7r; Triticum timopheevi ssp. viticosum – LrTt1; Aegilops ventricosa – Lr37; Aegilops kotschyi – Lr54; Elymus trachycaulis – Lr55; Aegilops Sharonensis – Lr56;
Aegilops geniculata – Lr57; Aegilops peregrine – Lr59; Triticum turgidum – Lr61; Aegilops neglecta – Lr62; Triticum monococcum – Lr63. Therefore, in breeding must be taken into account the fact that in the wheat genome the effective Lr-genes are introduced not in its "pure" form, but closely linked with other genes that are undesirable to use in breeding (Dinh et al., 2020; Lodgering et al., 1970).

It is a rather difficult task creating such varieties, and over time, they lose their resistance by the emergence of new races, pathogenic strains, and climatic changes. Trends in climate warming affect on the deterioration of the phytosanitary condition of crops (Mushtruk et al., 2020).

Scientific hypothesis

The scientific hypothesis is founded on identifying nature inheritance and manifest resistance genes to exciter of leaf rust. It is attaining by investigation of composition population exciter of disease and identifies resistance genes at collectible samples soft wheat. It is making it possible to increase the resistance gene pool and creating new heterogeneous varieties of soft wheat.

MATERIAL AND METHODOLOGY

The assessment varieties (Table 1) of winter wheat were created at the V. M. Remeslo Myronivka Institute of Wheat for resistance to leaf rust made in conditions of the artificial infectious background of the wheat pathogen. Experiments were in the field conditions at a field infectious nursery of the Plant Protection Department of the V. M. Remeslo Myronivka Institute of Wheat. The climate is temperate continental. The average annual air temperature is 7.6 °C. The sum of effective temperatures above 5 °C is 3000 °C. The duration of the frost-free period becomes an average of 165 days. The average annual amount of decline is 310 – 570 mm.

A suspension of a mixture of spores isolated from the local leaf rust population was used for inoculation. Wheat varieties were inoculated a mixture of spores with tacle in the ratio of 1:100 by a technique of Geshele (1971) in a tubing phase – beginning form ears in condition artificial infectious nursery. The spore load was 0.015 kg of urediniospores per one m² of sowing. The assessment of resistance was conducted dynamically every 10 days (Tkachyk, 2014). The variety Myronivskas 10 was used as a susceptible standard. The accounts defeat by the causative agent were evaluated on the scale according to Trybel et al. (2010) and Bober et al. (2020). A DNA was isolated from weight 25 – 40 mg of the plant material, obtained by grinding of 5 grains in ceramic mortars to a homogeneous powder and the further selection and weighing (Babaiants, 2011). DNA isolation was conducted using sets of Diatom™ DNA Prep100 (NEOGENE, Ukraine) according to a standard procedure with certain modifications (Trybel et al., 2010).

The caSNP12 marker was used in investigations of genetic material for identification allelic status of the resistance gene to wheat leaf rust Lr34, (Radchenko and Odintsova, 2008). PCR was performed on the amplifier GeneAmp® PCR System 2720 (Thermo Fisher Scientific, Massachusetts, USA) using sets GenPak® PCR Core according to the manufacturer's method. The primer sequences are as follows: for caSNP12F – 5'-TCCCCAGTTTAACTCTTG-3'; for caSNP12R – 5'-CATTCAGTCACCTCGACGC-3' (NEOGENE, Ukraine).

The conditions of the PCR were in line with the requirement recommended by the developer. As a result of PCR with a mixture of primers flanking the caSNP12 marker (Strahov, 1951). The stable allelic state of the markers (- Lr34 +) answered amplicons with a length of 234 p. n., the sensitivity was the absence of amplicons (Vyerchenko et al., 2019).

Fragments received from PCR were separated in 1.8% agarose gel. Ethyl bromide was used as an intercalating agent for DNA monitoring in an ultraviolet. The system VISION Gel (Scie-plas, Great Britain) was used for gel documentation. The length of the clearest and reproducible bands was determined using DNA marker O’Gene Ruler 50 bp Plus DNA Ladder (Fermentas, Lithuania).

Statistical analysis

To obtain information on the number and interaction of resistance genes, the obtained ratios of classes of resistant and susceptible plants (actual) were compared with one of the theoretically expected cleavages using the chi-square (χ²) correspondence criterion. The assumption that the difference between the actually obtained and theoretically expected splits is random was rejected if χ²exceeded the critical χ²critical. (χ²0.05 = 3.84). An error of results in statistical analysis p = 0.05. Statistical processing was performed in Microsoft Excel 2016 using the analytical application XLSTAT from Addinsoft for Microsoft Excel. Values were estimated using mean and standard deviations.

RESULTS AND DISCUSSION

The genes of high efficiency against the pathogen are Lr9, Lr19, Lr37, Lr42+Lr24, Lr43 (Lr21+Lr39) +Lr24, Lr9+L26, Lr10+Lr24 discovered by investigation the population of leaf rust on varieties of carriers of effective resistance genes (Leary et al., 2018; Ramanathan et al., 2018). The resistance of the varieties protected by the Lr24 genome is lost. We observed a slight defeat by the pathogen of the carriers of the Lr19 gene, indicating that the population has virulent clones against it. The pestules of leaf rust were marked also on varieties protected by the genome Lr9 in some years. The genes Lr37, Lr42 + Lr24 and Lr43 (Lr21 + Lr39) + Lr24 have high efficiency in recent years. The Lr13, Lr34, Lr37 genes provide the long-term protection of wheat against rust in combination with other genes (Dorofee, 1972; Bharti et al., 2016). The varieties with these genes remain resistant for 20 – 30 years in different countries of the world (Leppik, 1970).

10 highly resistant endemic species of wheat and its relatives with the highest immunity to rust, in particular: Triticum monococcum, T. timopheevii, T. millitinae, T. Zhykovski, T. fungicidum, Hayntriticum, Aegilops umbellulata are distinguished (Varella et al., 2017; Jafary, Szabo and Niks, 2006).
Breeding and Genetics Institute
Academy of Agricultural Sciences of Ukraine, the Plant
M. Remeslo Myronivka Institute of Wheat of the National

20

early

conducted

resistance against leaf rust, based on

Davoyan

Institute of Plant Protection
Ukraine;

Note:

susceptibility

Myronivska 10

Myronivska 264

Myronivska 808

Myronivska 61

Myronivska 27

Myrleben

Myronivska 28

Myronivska 30

Myronivska 31

Myronivska 33

Myrych

Myronivska 65

Kryzhynka

Myronivska 67

Kolumbia

Podolianka

Snižhana

Peresiašlavka

Smuhišanka

Demetra

Vesnianka

Volodarka

Favorytka

Pyvna

Zolotokolosa

Kalynova

Kolos

Myronivschyna

Monotyp

Ekonomka

Pamiatī Remesla

Myronivska storichna

Yuviliar

Myronivskyi

Myrliena

YasMohirka

Slavna

Yavoryna

Lehenda

Myronivska

(standard

susceptibility)

Plant damage, %, the average for 2015 – 2018 pp.
Resistance, number

13.8
9.5
17.5
11.3
8.3
18.3
4.5
18.8
7.5
11.3
10.0
16.3
8.3
15.0
2.8
11.3
10.0
13.8
4.5
15.0
5.3
5.0
5.0
12.5
20.8
13.8
7.5
5.0
5.8
12.5
8.3
6.7
5.8
5.8
6.7
5.8
6.7
5.8
10.0
10.0
26.3

1

5

7

8

6-7

5

7

4-5

7

8

6-7

8

6-7

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7

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6-7

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6-7

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23
Pereiaslavka, Demetra, Favorytka, Pyvna, Kalynova, Kolos Myronivschnya, Monotyp, Ekonomka, Myronivska stornichna, Lehenda Myronivska according to the results of investigations by Kovalyshyna et al. (2020), Lisova (2012) and You-Xiong et al. (2009). The Lr26 gene does not provide high resistance of varieties to leaf rust causative agent, since this gene has lost its effectiveness against the population of the disease as the data in Table 1 show.

The availability of wheat-rye translocation IAL/IRS in the genotype of winter wheat provides resistance to fungal diseases, because it carries a complex of resistance genes – to leaf rust Lr24, stem rust Sr/IRS, powdery mildew Pm17. This translocation was detected in varieties: Kolumbiia, Smuhlanka, Vesnianka, Zolotokolosa, Yasnochirka, Slavna, Yavoryna (Vlasenko, Koliuchyi and Chebakov, 2005; Landjeva et al., 2006). The Lr24 gene provides moderate resistance to leaf rust, varieties have damage of 2.8 – 17.2%.

The varieties Vesta, Snizhana, Demetra are protected by the Lr26 and Lr34 genes, and the Zolotokolosa – by the Lr24 and Lr34. The combination of these resistance genes inhibits the development of leaf rust on winter wheat varieties, damage of the leaf surface is within 10 – 17.5%.

The Myrliena variety contains the gene Lr23, and the Myronivska stornichna – Lr3, Lr23, Lr10, and Lr26 are ascertaining according to the analysis of the genealogy of varieties. The Lr3, Lr24 genes, protects the Ekonomka variety.

The gene Lr34, which belongs to the group of genes that provide partial resistance in the phase of adult plants, is found in the following varieties: Kryzhynka, Vesta, Snizhana, Volodarka, Demetra, Vdiachna, Pamiati Remesla, and Sviatkova (Kozub et al., 2017; Pirko et al., 2012). Eight varieties exhibit polymorphism for the gene Lr34 – Illichivka, Myronivska 30, Myronivska 32, Myronivska 65, Myronivska 66, Pyvna, Ekonomka, Lehenda Myronivska. Perhaps a small frequency of the dominant allele of the Lr34 gene in the varieties of MIW is related to the difficulty of identifying it in field conditions in the Right-Bank Forest-Steppe zone of Ukraine with the use of classical breeding methods, since for varieties of the Plant Breeding and Genetics Institute – National Center of Seed and Cultivar Investigation of the National Academy of Agricultural Sciences of Ukraine frequency is much higher. The varieties Zolotokolosa and Podolianka also contain the gene Lr34 according to Radchenko and Tishchenko (2010) and Krattinger et al. (2016).

We found that among the 15 newly created varieties, only 3 varieties – Berehynia myronivska, MIP Dniprianka, and Balada myronivska contain the Lr34 allele (+), which is only 20% of the investigated varieties according to the PCR results, using the caSNP12 marker (Table 2).

Table 2 Allelic status of the Lr34 gene in new winter wheat varieties of MIW by results of PCR on the marker caSNP12, 2017.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Entry into the State Register, year</th>
<th>Allelic status of the gene Lr34*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Svitank Myronivskyi</td>
<td>2014</td>
<td>-</td>
</tr>
<tr>
<td>Horlytsia myronivska</td>
<td>2016</td>
<td>-</td>
</tr>
<tr>
<td>Hospodynia myronivska</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>Berehynia myronivska</td>
<td>2016</td>
<td>+</td>
</tr>
<tr>
<td>MIP Kniazhna</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>MIP Vyshyvanka</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>MIP Valensia</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>Myronivska slava</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>Trudvnytsia myronivska</td>
<td>2017</td>
<td>-</td>
</tr>
<tr>
<td>Estafeta myronivska</td>
<td>2018</td>
<td>-</td>
</tr>
<tr>
<td>Vezha myronivska</td>
<td>2018</td>
<td>-</td>
</tr>
<tr>
<td>MIP Dniprianka</td>
<td>2018</td>
<td>+</td>
</tr>
<tr>
<td>Balada myronivska</td>
<td>2018</td>
<td>+</td>
</tr>
<tr>
<td>Hratsia myronivska</td>
<td>2018</td>
<td>-</td>
</tr>
<tr>
<td>MIP Assol</td>
<td>2018</td>
<td>-</td>
</tr>
</tbody>
</table>

Note: "+" – Associated with allele resistance Lr34 (+); "-" – allele present in susceptible varieties Lr34 (-).

CONCLUSION

It has been established that effective against the pathogen of leaf rust the are genes: Lr9, Lr19, Lr37, Lr42+Lr24, Lr43 (Lr21+Lr39) +Lr24, Lr9+Lr26, Lr10+Lr24. The genes Lr13, Lr34, Lr37 in combination with other genes, provide the long-term protection of wheat against leaf rust.

The wheat-rye translocation IBL/IRS was identified in the following varieties: Myronivska 61, Myronivska 27, Mirleneb, Myronivska 28, Myronivska 30, Myronivska 31, Myronivska 33, Myrych, Myronivska 65, Myronivska 66, Kryzhynka, Myronivska 67, Vesta, Snizhana, Pereiaslavka, Demetra, Favorytka, Pyvna, Kalynova, Kolos Myronivschnya, Monotyp, Ekonomka, Myronivska stornichna, Lehenda Myronivska.

The availability of wheat-rye translocation IBL/IRS was found in varieties: Kolumbiia, Smuhlanka, Vesnianka, Zolotokolosa, Yasnochirka, Slavna, Yavoryna. The Lr34 (+) allele is presented in varieties: Kryzhynka, Vesta, Snizhana, Volodarka, Demetra, Vdiachna, Pamiati Remesla, Sviatkova, Podolianka, Zolotokolosa, Berehynia myronivska, MIP Dniprianka, and Balada myronivska according to the results of the research. As a result of research, it was found that the wheat variety Myronivska Ukrainian selection is quite productive in comparison with other varieties and resistant to pathogens of various diseases, especially to the pathogen (brown rust).

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Contact address:
Hanna Kovalyshyna, National University of Life and Environmental Sciences of Ukraine, Agrobiological Faculty, Department of Genetics, Plant Breeding and Seed Production them Prof. M. O. Zelensky, Heroiv Oborony Street, 13, Kyiv, 03041, Ukraine, Tel.: +38(096)410-45-16, E-mail: hkovalyshyna@gmail.com
ORCID: https://orcid.org/0000-0002-2715-7679
Yulia Dmytrenko, National University of Life and Environmental Sciences of Ukraine, Agrobiological Faculty, Department of Genetics, Plant Breeding and Seed Production them Prof. M. O. Zelensky, Heroiv Oborony Street, 13, Kyiv, 03041, Ukraine, Tel.: +38(098)78-30-971,
Oksana Tonkha, Department of Soil Science and Soil Protection them prof. M. K. Shykuly, National University of Life and Environmental Sciences of Ukraine, 17 Herov Oborony Street, Kyiv 03041, Ukraine, E-mail: oksana16095@gmail.com
ORCID: https://orcid.org/0000-0002-0677-5494

Oleksandr Makarchuk, National University of Life and Environmental Sciences of Ukraine, Agrobiological Faculty, Department of Genetics, Plant Breeding and Seed Production them Prof. M. O. Zelensky, Heroiv Oborony Street, 13, Kyiv, 03041, Ukraine, Tel.: +38(096)335-75-31, E-mail: mcar2010@ukr.net
ORCID: https://orcid.org/0000-0002-3348-676X

Oleksandr Demydov, The V. M. Remeslo Myronivka Institute of Wheat of the National Academy of Agrarian Sciences of Ukraine, 68 Tsentralna Street, Tsentralne, Myronivka district, Kyiv region, 08853, Ukraine, E-mail: a.demidov@meta.ua
ORCID: https://orcid.org/0000-0002-5715-2908

Oleksandr Humeniuk, The V. M. Remeslo Myronivka Institute of Wheat of the National Academy of Agrarian Sciences of Ukraine, 68 Tsentralna Street, Tsentralne, Myronivka district, Kyiv region, 08853, Ukraine, E-mail: alexgynenyk@ukr.net
ORCID: https://orcid.org/0000-0002-1147-088X

Natalia Kozub, Institute of Plant Protection of the National Academy of Agrarian Sciences of Ukraine, 33 Vasylkivska Street, Kyiv 03022, Ukraine, Institute of Food Biotechnology and Genomics of the National Academy of Sciences of Ukraine, 2a Osipovskogo Street, Kyiv 04123, Ukraine, E-mail: natalkozub@gmail.com
ORCID: https://orcid.org/0000-0002-3572-1786

Anatolii Karelov, Institute of Plant Protection of the National Academy of Agrarian Sciences of Ukraine, 33 Vasylkivska Street, Kyiv 03022, Ukraine, Institute of Food Biotechnology and Genomics of the National Academy of Sciences of Ukraine, 2a Osipovskogo Street, Kyiv 04123, Ukraine, E-mail: tolikkarelov@meta.ua
ORCID: https://orcid.org/0000-0001-6548-2504

Igor Sozinov, Institute of Plant Protection of the National Academy of Agrarian Sciences of Ukraine, 33 Vasylkivska Street, Kyiv 03022, Ukraine, Institute of Food Biotechnology and Genomics of the National Academy of Sciences of Ukraine, 2a Osipovskogo Street, Kyiv 04123, Ukraine, E-mail: sia1953@ukr.net
ORCID: https://orcid.org/0000-0002-3621-5746

*Mikhailo Mushtruk, National University of Life and Environmental Sciences of Ukraine, Faculty of Food Technology and Quality Control of Agricultural Products, Department of Processes and Equipment for Processing of Agricultural Production, Heroev Oborony Str., 12 B, Kyiv, 03040, Ukraine, Tel.: +38098941-26-06, E-mail: mixej.1984@ukr.net
ORCID: https://orcid.org/0000-0002-3646-1226

Corresponding author: *