

# Alloplasmic recombinant lines (*H. vulgare*)-*T. aestivum* with 1RS.1BL translocation: initial genotypes for production of common wheat varieties

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Alloplasmic lines are formed when the cytoplasm of one species is replaced by the cytoplasm of another as a result of repeated recurrent crosses of wide hybrids with the paternal genotype. Since the cytoplasm replacement results in new intergenomic interactions between a nucleus and cytoplasm leading to variability of plant characteristics, alloplasmic lines with restored fertility can be an additional source of biodiversity of cultivated plants. Earlier, recombinant alloplasmic lines (*H. vulgare*)-*T. aestivum* designated as L-17(1)–L-17(37) were formed from a plant with partially restored fertility of the BC<sub>3</sub> generation of barley-wheat hybrid *H. vulgare* (cv. Nepolegayushchii) × *T. aestivum* (cv. Saratovskaya 29). This male-sterile hybrid was consistently backcrossed with wheat varieties Mironovskaya 808 (twice) and Saratovskaya 29, and Mironovskaya 808 had a positive impact on the restoration of fertility. This paper presents the results of investigation into a group of recombinant alloplasmic lines (L-17F<sub>4</sub>), as well as into doubled haploids (DH) lines – alloplasmic DH-17-lines obtained from anther culture of alloplasmic lines (L-17F<sub>2</sub>). The most productive of these lines were used as initial breeding genotypes. Hybrid form Lutescens 311/00-22 developed from the crossing of the alloplasmic DH(1)-17 line (as maternal genotype) with euplasmic line Com37 (CIMMYT), the source of the 1RS.1BL wheat-rye translocation, proved to be successful for breeding. The presence of the 1RS.1BL translocation in the genome of the Lutescens 311/00-22 form and the L-311(1)–L-311(6) alloplasmic lines isolated from it did not lead to a decrease of fertility or sterility in the plants. This indicates that the chromosome of the 1BS wheat does not carry the gene(s) that determine the restoration of fertility in the studied (*H. vulgare*)-*T. aestivum* alloplasmic lines. Alloplasmic lines L-311(1)–L-311(6) showed their advantage in comparison with the standard varieties for resistance to leaf and stem rust, yield, and grain quality. The breeding tests performed at Omsk Agricultural Scientific Center, Agrocomplex "Kurgansemena", Federal State Unitary Enterprise "Ishimskoe" (Tyumen Region), using alloplasmic lines L-311(5), L-311(4) and L-311(6) resulted in varieties of spring common wheat Sigma, Uralosibirskaya 2 and Ishimskaya 11, respectively.

Key words: alloplasmic lines (*H. vulgare*)-*T. aestivum*; DH-lines; translocation 1RS.1BL; varieties of common wheat.

## Аллоплазматические рекомбинантные линии (*H. vulgare*)-*T. aestivum* с транслокацией 1RS.1BL: исходные генотипы для создания сортов мягкой пшеницы

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Аллоплазматические линии образуются при замещении цитоплазмы одного вида на цитоплазму другого в результате повторяющихся возвратных скрещиваний отдаленных гибридов с отцовским генотипом. Так как при замещении цитоплазмы между ядром и цитоплазмой возникают новые межгеномные взаимодействия, приводящие к изменчивости признаков растений, аллоплазматические линии с восстановленной фертильностью могут служить дополнительным источником биоразнообразия культурных растений. Ранее в наших работах были получены рекомбинантные аллоплазматические линии (*H. vulgare*)-*T. aestivum*, обозначенные как L-17(1)–L-17(37), сформированные от растения с частично восстановленной фертильностью BC<sub>3</sub> поколения ячменно-пшеничного гибрида *H. vulgare* (Неполегающий) × *T. aestivum* (Саратовская 29). Этот мужско-стерильный гибрид был последовательно беккроссирован сортами пшеницы Мироновская 808 (дважды) и Саратовская 29, где сорт Мироновская 808 оказал влияние на восстановление фертильности. В статье представлены результаты изучения группы рекомбинантных аллоплазматических линий L-17F<sub>4</sub>, а также линий гаплоидов с удвоенным числом хромосом – аллоплазматических ДГ-17-линий, полученных в результате культивирования пыльников линий L-17F<sub>2</sub>. Наиболее продуктивные из изученных линий включены в селекционный процесс. Успешной для селек-

ции оказалась гибридная форма Лютесценс 311/00-22, полученная от скрещивания аллоплазматической ДГ(1)-17-линии с эуплазматической линией Com37 (СИММТ), источником пшенично-ржаной транслокации 1RS.1BL. При отсутствии транслокации 1RS.1BL в геноме формы Лютесценс 311/00-22 и выделенных из нее аллоплазматических линий Л-311(1)–Л-311(6) не привело к снижению фертильности растений или их стерильности. Это указывает на то, что хромосома пшеницы 1BS не несет ген(ы), определяющие восстановление фертильности у изученных в настоящей работе аллоплазматических линий (*H. vulgare*)-*T. aestivum*. Линии Л-311(1)–Л-311(6) показали их преимущество по сравнению с сортами-стандартами по устойчивости к бурой ржавчине, стеблевой ржавчине, урожайности, качеству зерна. В результате селекционных испытаний в Омском аграрном научном центре, Агрокомплексе «Курган-семена», на предприятии «Ишимское» Тюменской области на основе аллоплазматических линий Л-311(5), Л-311(4) и Л-311(6) созданы сорта яровой мягкой пшеницы Сигма, Уралосибирская 2 и Ишимская 11 соответственно.

Ключевые слова: аллоплазматические линии (*H. vulgare*)-*T. aestivum*; ДГ-линии; транслокация 1RS.1BL; сорта мягкой пшеницы.

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Alloplasmic lines are formed as a result of repeated recurrent crosses of wide hybrids with the paternal genotype and combine the cytoplasm of the maternal species with the nuclear genome of the pollen parent (Tsunewaki, 1996). The new nuclear-cytoplasmic interactions formed due to cytoplasm replacement can cause epigenetic modifications of nuclear genes (Soltani et al., 2016), leading to changes at the level of transcription and metabolism (Crosatti et al., 2013; Soltani et al., 2016). These processes may lead to plant developmental disorders (Suzuki et al., 1995), changes in resistance to stress factors (Dhitaphichit et al., 1989; Keane, Jones, 1990; Buloychik et al., 2002; Talukder et al., 2015) and changes in the manifestation of agronomically important traits (Ekiz et al., 1998; Liu et al., 2002; Atienza et al., 2008; Klimushina et al., 2013). The most common sign of a nuclear-cytoplasmic conflict included in alloplasmic lines is cytoplasmic male sterility (CMS) (Tsunewaki, 1996; Bentolila et al., 2002).

From a practical perspective, alloplasmic lines are obtained from different species of cultivated plants and characterized by CMS that represents a system for obtaining hybrid seeds in heterotic breeding (Cisar, Cooper, 2002). Given that new intergenomic interactions arise during cytoplasm replacement, alloplasmic lines with restored fertility can be an additional source of biodiversity in cultivated plants (Liu et al., 2016). For wheat alloplasmic lines, examples of their practical use are not numerous. The VPM1 alloplasmic line carrying the cytoplasm of *Aegilops ventricosa* Tausch was the source of two

commercial varieties of wheat Reason and Rendezvous (Jones et al., 1998) resulting from hybridization of the amphiploid (*Ae. ventricosa* × *T. persicum*) with common wheat variety Marne. This alloplasmic line and the varieties derived from it are resistant to a number of fungal pathogens due to the introgression of *Ae. ventricosa* genes *Pch-1* and *Sr38/Lr37/Yr17* (Delibes et al., 1988; Friebe et al., 1996). Common wheat variety 'Xiaoshan 2134' carrying the cytoplasm of *Ae. crassa*, is characterized by high grain quality, resistance to salinity and high yield (Liu et al., 2002). The authors ascribe these traits to the effect of nuclear-cytoplasmic heterosis. The lines of common wheat with cytoplasm *Triticum timopheevii* and *Secale cereale* characterized by tolerance to drought (Semenov et al., 2014) and high-quality gluten characteristics are considered promising for breeding (Klimushina et al., 2013; Semenov et al., 2016). With the use of rye cytoplasm, new forms of triticale have been obtained (Gordey et al., 2011).

Earlier, we reported the production of the recombinant alloplasmic lines of common wheat derived from back-cross progenies of barley-wheat hybrids *H. vulgare* × *T. aestivum* (Pershina et al., 1998) and restoration of their fertility (Pershina et al., 1999a). Alloplasmic lines (*H. vulgare*)-*T. aestivum* characterized by different fertility levels proved to be valuable models for studying the variability of both nuclear and organellar (mitochondrial and chloroplast) genomes in the process of nuclear-cytoplasmic co-adaptation (Bildanova et al., 2004; Aksyonova et al., 2005; Pershina et al., 2014). Some

of the recombinant alloplasmic lines with full fertility restoration were included in breeding of common wheat (Belan et al., 2017).

The aim of presented research was to generalize and analyse the results of obtaining and studying of recombinant and introgressive alloplasmic lines (*H. vulgare*)-*T. aestivum*, which have been successfully used in breeding and have become a source of the new varieties of spring common wheat.

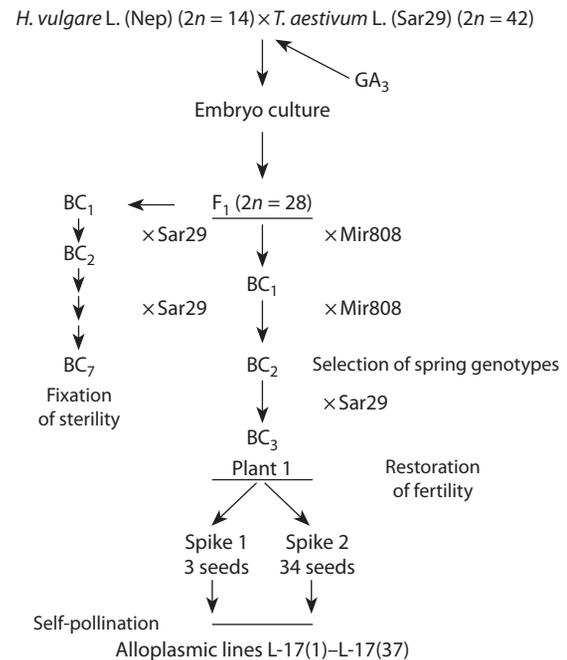
## Materials and methods

**Production of recombinant and introgressive alloplasmic lines (*H. vulgare*)-*T. aestivum*.** The Nepolegayushchii variety of cultivated barley was crossed with spring common wheat Saratovskaya 29, and the pollinated flowers were treated with a solution of gibberellic acid. Using embryo cultivation, hybrid plants *H. vulgare* × *T. aestivum* were grown and were characterized by male sterility but female fertility (Pershina et al., 1998). The F<sub>1</sub> hybrids were consistently backcrossed with wheat Mironovskaya 808 (twice) and Saratovskaya 29. Among the plants from the BC<sub>3</sub> generation, one partially fertile plant was isolated (three seeds were set in one spike, and 34 in another) (Pershina et al., 1999a). Each fertile plants grown from these seeds became the source of recombinant alloplasmic lines designated L-17(1)–L17(37) (Fig. 1). To form each subsequent generation of alloplasmic lines, the seeds from the main spike of the most productive plant were selected. Alloplasmic DH-lines DH(1)-17F<sub>2</sub>, DH(1)-17F<sub>2</sub>, and DH(1)-17F<sub>2</sub> were derived by anther culture of alloplasmic line L-17F<sub>2</sub> (Pershina et al., 1999b).

In this paper, we present the results of a study of seven alloplasmic lines: L-17(3)F<sub>4</sub>, L-17(4)F<sub>4</sub>, L-17(9)F<sub>4</sub>, L-17(12)F<sub>4</sub>, L-17(18)F<sub>4</sub>, L-17(21)F<sub>4</sub>, and L-17(24)F<sub>4</sub>; three alloplasmic DH lines: DH(1)-17F<sub>2</sub>, DH(2)-17F<sub>2</sub>, and DH(3)-17F<sub>2</sub>; and six introgression alloplasmic lines: L-311(1)F<sub>10</sub>–L-311(6)F<sub>10</sub>. The introgression alloplasmic lines were isolated from the hybrid form L-311/00-22F<sub>5</sub> obtained as a result of the crossing of alloplasmic line DH(1)-17F<sub>2</sub> with euplasmic line Com37 (Belan et al., 2010). Using the results of chromosome C-banding, E.D. Badaeva identified the wheat-rye translocation 1RS.1BL in the alloplasmic lines of group L-311 (Pershina et al., 2013).

### Growing conditions and study of alloplasmic lines.

Alloplasmic lines L-17F<sub>4</sub> and DH-17F<sub>2</sub> were grown in the field of Institute of Cytology and Genetics near Novosibirsk in 1999 when there was no mass development of leaf rust or stem rust pathogens. Control wheat line Saratovskaya 29 (Sar29) was used to obtain barley-wheat hybrids and alloplasmic lines (*H. vulgare*)-*T. aestivum*. The alloplasmic lines of the L-311F<sub>10</sub> group were grown in the field of the Institute of Cytology and Genetics in 2017. Euplasmic line Om37 carrying translocation 1RS.1BL and isolated from variety Omskaya 37, and alloplasmic line L-17(3)F<sub>12</sub> were used as controls. Plants were grown on plots 50 cm wide in rows of 10, with a distance of 25 cm between rows. The germination rate was determined by the number of seedlings. During harvesting, the height of



**Fig. 1.** Development of a set of recombinant (*H. vulgare*)-*T. aestivum* alloplasmic lines L-17 (Pershina et al., 1998, 1999a).

Designation: Nep – barley variety Nepolegayushchii; varieties of common wheat: Sar29 – Saratovskaya 29, Mir808 – Mironovskaya 808. GA<sub>3</sub> – gibberellic acid.

the plants, the number of productive spikes, the length of the main spike, the number of spikelets per main spike, the number of seeds per main spike, and the frequency of plants with full fertility (more than 35 seeds in the main spike) were estimated.

To confirm the presence of the translocation 1RS.1BL, the SCAR marker iag95 linked to the genes *Lr26* and *Sr31* localized in the short arm of the rye chromosome 1R (Mago et al., 2002), and genomic *in situ* hybridization (Mukai, Gill, 1991) were used in the lines included in this study. At least 20 plants were used per treatment. The differences between the average values of the lines were compared using *t*-tests. Data were analysed using Statistica v.7.0.61.0.

Alloplasmic hybrid form L-311/00-22 and alloplasmic lines L-311(1)–L-311(6) were grown in the field of Laboratory of Spring Common Wheat of Omsk Agrarian Scientific Center, following the methods described in (Belan et al., 2017). Since 2007, the L-311 alloplasmic lines have been tested according the full breeding scheme for BN-1, BN-2, and BN-3 (breeding nurseries of the first, second, and third year of study) and in a competitive variety trial nursery. Assessments of resistance to powdery mildew (*Blumeria graminis* f. sp. *tritici*), leaf rust (*Puccinia recondita* f. sp. *tritici*), and stem rust (*Puccinia graminis* f. sp. *tritici*) were made according to previously described methods (Belan et al., 2017). In addition, yield and grain quality parameters (1000-grain weight, seed protein content) were determined. Using analogous methods, alloplas-

**Table 1.** Characteristics of the alloplasmic lines of L-17F<sub>4</sub> and DH-17F<sub>2</sub> (Novosibirsk, field, 1999)

Genotypes	Height of plants, cm	Number of productive spikes	Length of the main spike, cm	Number of spikelets per main spike	Number of seeds		1 000-grain weight	% of FF plants
					per main spike	per plant		
Sar29	96.3 ± 2.0	4.3 ± 0.3	9.1 ± 0.1	13.8 ± 0.2	40.1 ± 1.1	152.3 ± 13.4	45.8 ± 0.7	80
L-17(9)F <sub>4</sub>	87.5 ± 2.1 (**)	3.8 ± 0.2	8.2 ± 0.2 (***)	12.9 ± 0.2 (**)	38.1 ± 1.9	122.8 ± 10.9	42.9 ± 0.9 (*)	60 (*)
L-17(18)F <sub>4</sub>	93.2 ± 1.9	2.9 ± 0.3 (**)	7.7 ± 0.1 (***)	12.1 ± 0.2 (***)	33.3 ± 1.7 (**)	88.6 ± 12.6 (**)	43.0 ± 0.8 (*)	65
L-17(4)F <sub>4</sub>	96.4 ± 1.3	5.4 ± 0.6	9.3 ± 0.2	13.6 ± 0.4	42.8 ± 2.1	194.6 ± 20.5	44.5 ± 0.8	85
L-17(21)F <sub>4</sub>	94.6 ± 1.6	5.2 ± 0.4	9.1 ± 0.1	14.2 ± 0.3	44.8 ± 1.5 *	192.1 ± 14.6 *	43.5 ± 0.6 (*)	95
L-17(12)F <sub>4</sub>	100 ± 1.7	5.6 ± 0.3 **	9.2 ± 0.1	14.5 ± 0.3	41.8 ± 1.0	197.9 ± 15.7 *	44.4 ± 0.7	95
#L-17(3)F <sub>4</sub>	85.6 ± 2.5 (**)	6.1 ± 0.3 **	9.4 ± 0.2	14.2 ± 0.3	42.6 ± 1.4	199.7 ± 15.4 *	44.9 ± 0.9	75
#L-17(24)F <sub>4</sub>	99.8 ± 1.7	6.6 ± 0.3 ***	9.3 ± 0.1	15.5 ± 0.2 ***	42.4 ± 0.9	227.9 ± 12.7 ***	45.7 ± 0.6	90
#DH(1)-17F <sub>2</sub>	96.1 ± 1.1	6.7 ± 0.3 ***	9.3 ± 0.2	15.2 ± 0.1 ***	45.4 ± 1.2 **	238.4 ± 12.3 ***	45.9 ± 0.5	85
#DH(2)-17F <sub>2</sub>	80.2 ± 1.3 (***)	5.5 ± 0.3 ***	9.3 ± 0.2	14.8 ± 0.2 **	45.2 ± 1.6 *	188.8 ± 10.9 *	46.6 ± 0.4	100
#DH(3)-17F <sub>2</sub>	90.9 ± 1.3 (*)	6.7 ± 0.4 ***	10.0 ± 0.1 ***	15.4 ± 0.2 ***	44.6 ± 1.4 *	239.7 ± 15.9 ***	44.9 ± 0.5	90

Note: FF – full fertility. The difference in comparison with the parent Saratovskaya 29 line is significantly less at (\*)  $p < 0.05$ ; (\*\*)  $p < 0.01$  and (\*\*\*)  $p < 0.001$ ; and significantly more at \*  $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ . # indicates the lines included in breeding.

mic line L-311(4) has been studied in the Agrocomplex “Kurgansemena” competitive variety trial nursery since 2013, and alloplasmic line L-311(6) – since 2014 in the Enterprise “Ishimskoe” (Tyumen Region) competitive variety trial nursery.

### Results and discussion

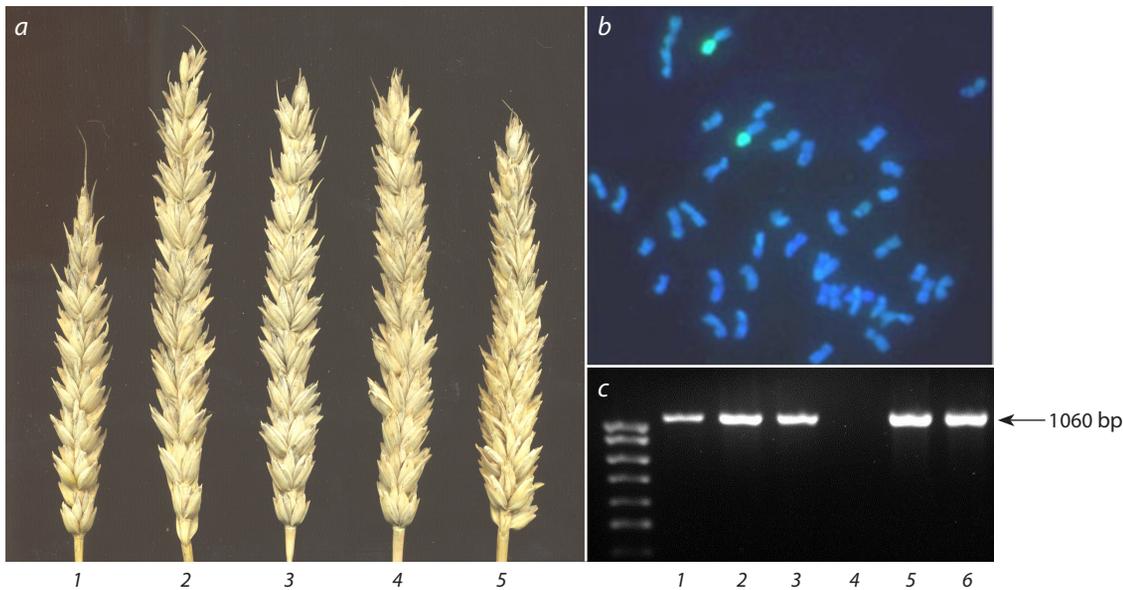
In this study recombinant alloplasmic lines (*H. vulgare*)-*T. aestivum* were investigated, in which barley variety Nepolegayushchii (Nep) was a source of the cytoplasm, and the recombinant nuclear genome was formed with the use of common wheat varieties Saratovskaya 29 (Sar29) and Mironovskaya 808 (Mir808) (see Fig. 1) (Persina et al., 1999a). Use of the Mir808 variety in recurrent crosses of male sterile barley-wheat hybrid *H. vulgare* (Nep) × *T. aestivum* (Sar29) had a positive effect on fertility restoration: one of the plants of the BC<sub>3</sub> generation set seeds after self-pollination (see Fig. 1).

According to our data, the Mironovskaya yarovaya wheat variety recovered from Mir808 (Dorofeev et al., 1987), along with the Ulyanovka and Pyrotrix 28 varieties (Persina et al., 1999a, 2014), which also restored full fertility of common wheat in the barley (*H. vulgare*) cytoplasm. When the backcrossing of barley-wheat hybrids *H. vulgare* × *T. aestivum* (Sar29) included only the Sar29 variety in the backcrossed progenies, fixation of complete sterility occurred (see Fig. 1) (Persina et al., 1999a, 2014). In addition, depending on the genetic diversity of wheat varieties included in the backcrossing of barley-wheat hybrids, numerous alloplasmic lines (*H. vulgare*)-*T. aestivum* with different levels of fertility were obtained, some of which, when self-pollinated,

segregated into plants with low fertility including those being completely sterile (Persina et al., 1999a, 2014).

Alloplasmic lines L-17(1)–L-17(37) formed from a partially fertile BC<sub>3</sub> generation plant with a  $2n = 42$  did not contain barley chromosomes and were fertile in F<sub>2</sub>–F<sub>3</sub> generations (Bildanova et al., 2004). The germination of the recombinant L-17F<sub>4</sub> alloplasmic lines was similar to the control level (not less than 98 %), and all the plants were fertile (Table 1).

In the majority of alloplasmic lines, the frequency of plants with full fertility (seed set higher than 35 seeds in the main spike) was at the level of the Sar29 parent line, varying from 65 % in L-17(18)F<sub>4</sub> to 95 % in L-17(21)F<sub>4</sub> and L-17(12)F<sub>4</sub> (see Table 1). The full fertility was significantly lower than that of the control (60 %) in alloplasmic line L-17(9)F<sub>4</sub>. The analysis of the productivity characteristics (the number of productive spikes, the length of the main spike, the number of spikelets and seeds per main spike, and the number of seeds per plant) differed between different alloplasmic lines. So, in L-17(9)F<sub>4</sub> and L-17(18)F<sub>4</sub>, the values of some of these parameters were significantly lower than in the Sar29 control line, while in L-17(4)F<sub>4</sub> they reached the control level. Alloplasmic lines L-17(21)F<sub>4</sub>, L-17(12)F<sub>4</sub>, L-17(3)F<sub>4</sub> and L-17(24)F<sub>4</sub> exceeded line Sar29 in the number of seeds per plant. The increase in L-17(21)F<sub>4</sub> was due to an increase in the number of seeds per main spike; an increase of a number of productive spikes in alloplasmic lines L-17(3)F<sub>4</sub> and L-17(12)F<sub>4</sub>; and an increase in the number of productive spikes per plant and the number of spikelets per main spike in alloplasmic line L-17(24)F<sub>4</sub>. For 1000-grain weight, the values of this parameter were the same at the



**Fig. 2.** The proof of the presence of wheat-rye translocation in the alloplasmic lines of L-311(4) $F_{10}$ , L-311(5) $F_{10}$ , L-311(6) $F_{10}$  by the results of GISH and PCR analysis.

*a* – spikes: 1 – alloplasmic line L-17(3) $F_{12}$ ; 2–4 – alloplasmic lines L-311(4) $F_{10}$ , L-311(5) $F_{10}$ , L-311(6) $F_{10}$ ; 5 – euplasmic line Om37. *b* – GISH-analysis of alloplasmic lines L-311(4) $F_{10}$  (1RS chromosome is marked in green). *c* – PCR assay developed for iag95 marker linked with *Lr26* gene: 1 – rye *S. cereale* (control), 2 – Om37, 3 – L-311(4) $F_{10}$ , 4 – L-17(3) $F_{12}$ , 5 – L-311(5) $F_{10}$ , 6 – L-311(6) $F_{10}$ .

control level for the majority of the lines studied and was lower for lines F-17(9) $F_4$ , L-17(18) $F_4$ , and L-17(21) $F_4$  (see Table 1).

These data show that the new nuclear genome that formed as a result of recombination between two genomes of wheat varieties Saratovskaya 29 and Mironovskaya 808 when interacting with the cytoplasm of barley had a positive effect not only on the restoration and maintenance of fertility of alloplasmic lines (*H. vulgare*)-*T. aestivum* but also on the traits that determine the productivity of plants. This is mostly due to the high ecological plasticity and high productivity of Saratovskaya 29 and Mironovskaya 808, which have been grown for a long time in many regions with different soil and climatic conditions and hybridized with each other and with other genotypes of wheat to produce many varieties (Dorofeev et al., 1987).

Another important result of our study is that fertility and the signs of high productivity were also evident in the DH lines derived from alloplasmic line L-17 $F_2$ . Earlier we had been able to obtain 20 green plants grown from anther culture of alloplasmic line L-17 $F_2$  on a modified medium of P-II (Pershina et al., 1999b). Eleven of these plants with  $2n = 42$  were fertile. These plants were used for development of a separate alloplasmic DH line. In the current paper we present the results of a study of the self-pollinated progeny of three alloplasmic DH lines: DH(1)-17 $F_2$ , DH(2)-17 $F_2$ , and DH(3)-17 $F_2$  (see Table 1). In these lines, in comparison with line Sar29, the number of productive spikes, the number of spikelets and seeds

per main spike, and the number of seeds per plant were significantly higher. Due to their high level of fertility and productivity, these alloplasmic DH lines, along with the original alloplasmic lines L-17(3) $F_4$  and L-17(24) $F_4$ , were included in the breeding. In addition, among these alloplasmic lines, we found three more undersized lines such as L-17(3) $F_4$ , DH(2)-17 $F_2$ , and DH(3)-17 $F_2$ .

To date, from a practical point of view, the hybrid combination obtained in Laboratory of Spring Common Wheat Breeding (Omsk Agrarian Scientific Center) by crossing alloplasmic line DH(1)-17 $F_2$  (maternal genotype) with line Com37 (pollinator, provided for the study by A.I. Morgunov from the CIMMYT collection) has been the most effective. In the early breeding nurseries, hybrid alloplasmic form Lutescens 311/00-22 was reproduced as a population, from which, after selection based on agronomical valuable traits, six promising alloplasmic lines were identified: L-311(1), L-311(2), L-311(3), L-311(4), L-311(5), and L-311(6).

Using C-banding, E.D. Badaeva revealed in these alloplasmic lines a wheat-rye translocation identified as 1RS.1BL (Pershina et al., 2013), which was inherited in a number of self-pollinated generations. Figure 2 shows the spikes of alloplasmic lines L-311(4) $F_{10}$ , L-311(5) $F_{10}$ , L-311(6) $F_{10}$  and the presence of translocation of 1RS.1BL in these lines proved by the results of GISH and PCR analysis. Since translocation 1RS.1BL was not detected the alloplasmic line of group L-17, it can be assumed that Com37 used as a pollinator in production of hybrid Lutescens 311/00-22, had this translocation. (Currently,

**Table 2.** Characteristics of alloplasmic lines L-311(1)F<sub>10</sub>–L-311(6)F<sub>10</sub> in comparison with euplasmic line Om37-1RS.1BL and alloplasmic line L-17(3)F<sub>12</sub>

Genotypes	Number of productive spikes	Length of main spike, cm	Number of spikelets per main spike	Number of seeds	
				per main spike	per plant
Om37	5.0±0.5	10.5±0.2	18.6±0.3	41.5±1.2	183.2±18.1
L-311(1)F <sub>10</sub>	5.2±0.3	11.5±0.2**	19.6±0.3*	47.9±1.2**	194.3±18.5
L-311(2)F <sub>10</sub>	6.0±0.4	11.8±0.1***	19.9±0.2***	49.5±0.8***	231.8±18.3
L-311(3)F <sub>10</sub>	6.1±0.5	12.4±0.1***	20.3±0.3***	49.5±1.2***	257.4±16.2**
L-311(4)F <sub>10</sub>	6.8±0.4*	12.4±0.1***	20.9±0.3***	56.8±1.0***	263.5±19.3**
L-311(5)F <sub>10</sub>	6.5±0.5*	12.7±0.3***	20.3±0.4***	50.9±2.1***	252.1±22.2*
L-311(6)F <sub>10</sub>	6.3±0.4*	12.8±0.2***	21.2±0.4***	53.8±1.2***	260.3±22.6*
L-17(3)F <sub>12</sub>	4.7±0.4	8.5±0.2(***)	14.6±0.2(***)	32.6±1.2(***)	153.7±13.4

Note: The difference in comparison with euplasmic line Om37 is significantly greater at \*  $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ ; and significantly less at (\*\*\*)  $p < 0.001$ .

**Table 3.** Results of studying promising alloplasmic lines of group L-311 in a competitive variety trial nursery, Omsk, 2010

Lines	Growing season, days	Productivity, t/ha		1 000-grain weight		Protein content in the grain (N×5.7), %		Lesion of rust disease	
		lines	± of Om33	lines	± of Om33	lines	± of Om33	leaf rust	stem rust
L-311(1)	98	4.75	0.16	47.7	4.6	16.56	1.48	0	10
L-311(2)	97	5.00	0.41	44.9	1.8	18.24	3.16	1	20
L-311(3)	98	5.29	0.7	43.0	-0.1	18.78	3.7	1	5
L-311(4)	98	5.05	0.46	44.4	1.3	17.78	2.7	5	5
L-311(5)	94	5.25	0.66	45.8	2.7	17.64	2.56	5	10
L-311(6)	93	5.00	0.4	45.5	2.4	17.53	2.45	5	10
Om33-St	93	4.59	–	43.1	–	15.08	–	80	50

Note: Om33-St is a variety of spring common wheat Omskaya 33 used as a reference.

the Com37 line and alloplasmic lines DH(1)-17F<sub>2</sub> have been lost.)

The fact that the substitution of chromosome 1BS for 1RS did not result in decrease of fertility or sterility in alloplasmic lines (*H. vulgare*)-*T. aestivum* indicates that the restoration and maintenance of fertility of these lines does not depend on the effect of *Rf*-genes controlling the restoration of wheat fertility in the alien cytoplasm and localized in chromosome 1BS (Tsunewaki, 2015).

In our previous work, other genotypes of alloplasmic lines (*H. vulgare*)-*T. aestivum* were obtained in which the introduction of wheat-rye translocation 1RS.1BL into the genome, including a combination with wheat-*Agropyrum* translocation 7DL-7Ai, did not have a negative effect on the fertility (Pershina et al., 2014).

The results of a study into the morphobiological traits of alloplasmic lines L-311(1)F<sub>10</sub>–L-311(6)F<sub>10</sub> in comparison with alloplasmic line L-17(3)F<sub>12</sub> (has no the 1RS.1BL translocation), and euplasmic line Om37 carrying this translocation, are presented in Table 2.

In all the six alloplasmic lines of group L-311F<sub>10</sub> carrying the 1RS.1BL translocation the lengths of the main spike, the number of spikelets and the number of seeds per main spike were significantly higher than in control line L-17(3)F<sub>12</sub> and line Om37. In addition, alloplasmic lines L-311(3)F<sub>10</sub>, L-311(4)F<sub>10</sub>, L-311(5)F<sub>10</sub>, and L-311(6)F<sub>10</sub> exceeded the control line in the number of seeds per plant, and the lines L-311(4)F<sub>10</sub>, L-311(5)F<sub>10</sub>, and L-311(6)F<sub>10</sub> – in the number of productive spikes.

Since rye chromosome arm 1RS carries a complex of genes (*Lr26/Sr31/Yr9/Pm8*) controlling resistance to fungal pathogens (Singh et al., 1990), the varieties with 1RS.1BL have become widespread throughout the world (Rabinovich, 1998; Schlegel, 2018). Mass cultivation of homogeneous varieties of wheat with this translocation has reduced the effectiveness of the gene resistance to fungal pathogens, including appearance of the highly aggressive *Ug99* race virulent to the *Sr31* gene (Pretorius et al., 2000). At the same time, the effect of 1RS.1BL translocation on the manifestation of agronomically important and adaptive

traits depends on the genetic background and plant growing regions (Hoffmann, 2008; Tahmasebi et al., 2015). The gene *Sr31* remains effective for protection against stem rust in different regions of Russia (Gulyaeva, 2017).

From the data obtained a conclusion can be made that the genetic background of the alloplasmic lines of group L-311 was favourable for realization of the positive effect of the 1RS.1BL translocation on both agronomically important and adaptive traits in the breeding trials regions including Omsk, Kurgan and Tyumen regions.

The study of alloplasmic lines L-311(1)–L-311(6) for consecutive self-pollinated generations in breeding nurseries and competitive variety trial nurseries was initially carried out in the fields of the Omsk Agrarian Scientific Center. It should be emphasized that leaf rust expansion had been noted there every year since 2001, and in 2007 the epiphytomy of this pathogen was recorded. Since 2009, the threat of stem rust has increased. In addition, powdery mildew is observed in this region every year.

Studying alloplasmic lines L-311(1)–L-311(6) demonstrated their moderate resistance to powdery mildew. In comparison with variety-standard Omskaya 33 susceptible to leaf and stem rust pathogens, the alloplasmic lines showed complex resistance to local populations of leaf and stem rust (Table 3).

Moreover, these alloplasmic lines exceeded the variety-standard for yield, 1000-grain weight, and seed protein content. Table 3 presents the results of studying alloplasmic lines L-311(1)–L-311(6) in 2010 in a competitive variety trial nursery. According to the tests results obtained from 2010 to 2012, middle-ripened alloplasmic line L-311(5) was identified as the most promising and was transferred in 2013 to the State Committee for Testing of New Varieties as variety Sigma. This variety was included in State Register of Selection Achievements of the Russian Federation in West Siberia in 2016.

The middle-ripened alloplasmic line L-311(4) was studied in the competitive variety trial nursery of Agro-complex “Kurgansemena”. Based on the three year results (2012–2015), variety Uralosibirskaya 2 was transferred to the State Committee for Testing of New Varieties in 2016. This variety was tested in the Urals and West Siberian region. In 2014, middle-early alloplasmic line L-311(6) was studied in the competitive variety trial nursery of Enterprise “Ishimskoe”, Tyumen Region. Based to the four-year data this line was transferred to the State Committee for Testing of New Varieties as variety Ishimskaya 11 in West Siberia in 2017.

In addition, the efficiency of using alloplasmic lines from group L-311 for obtaining hybrids and the alloplasmic DH lines combining resistance genes to fungal pathogens localized in wheat-rye translocation 1RS.1BL with complexes of effective resistance genes from other sources has been shown (Pershina et al., 2013).

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## Conflict of interest

The authors declare they have no conflict of interest.

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