BREEDING ASPECTS OF LOW TEMPERATURE TOLERANCE IN WHEAT

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Wheat resistance to low temperatures is complex in nature and dependant on variety as well as on interaction with the environment. In unfavorable conditions for hardening, even varieties resistant to cold may exhibit higher susceptibility to low temperatures. Over a two-year period (2003-04), tests were conducted in cold chamber at–15˚C lasting 24, 36, 48 and 60 hours. The 23 wheat varieties analyzed were divided into four groups according to year of release. In the first group, which was mostly made up of foreign introductions, the average plant survival rate across treatments was 87%. The different origins of the varieties from this group were a significant factor behind the differences in their resistance levels. The second group, comprised of the first domestic varieties, had an average plant survival rate of 89%, while the third, containing the dominant varieties from the 1980s, had a plant survival rate of 88.4% on average. In the fourth group, consisting mostly of varieties that are

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currently commercially grown in the country, the average plant survival rate was 86.6%. Thanks to high temperatures present during the process of hardening, there was greater plant damage and percent winterkill in the second year than in the first. The resistance of the varieties to low temperatures has been maintained at a level enabling successful and stable production under the cold temperature stress conditions of southeastern Europe.

Key word: breeding, low temperatures, variety, wheat

INTRODUCTION

Securing high and stable crop yields is one of the primary tasks of agricultural production. The attainment of stable yields depends not only on the agroecological conditions and genetic potential for yield but on other factors as well. When developing a variety or hybrid, therefore, it is necessary to bring into accord a large number of traits that directly or indirectly influence the end results of breeding.

Most plant species become exposed to the effects of unfavorable environmental factors during the vegetation period. Yields of the major crops may be reduced by as much as 50% environmental stresses such as low and high temperatures, drought, salinity, and a deficit or toxicity of a particular chemical element (LEVITT 1980). In the winter, without snow, wheat plants can withstand temperatures of -18 to -20°C for up to 48 hours with only minimum leaf damage, whereas in the spring they die at -8 to -10°C (SAULESCU & BRAUN 2001). Yields of different small grains species in southeastern Europe in some years depend to a considerable extent on resistance to low temperatures the plant is exposed to in the course of the growing season.

The modern theory of low-temperature resistance defines this characteristic as the ability of a plant to withstand cold under a given set of environmental conditions in the context of its phylogenetic and ontogenetic development (JEVTIC 1986). The frost tolerance of a plant differs from one stage of plant development to another (FOWLER e*t al*., 1996; MAHFOOZI *et al.*, 2001). Wheat varieties respond differently to low temperatures and this response depends in part on the time when exposure to such temperatures occurs in the autumn, winter or spring (VEISZ & SUTKA 1989). Characteristic of this trait is that there is little to no damage to the plant despite the action of low temperatures, so the resulting yields remain satisfactory (OLGUN e*t al*., 2005).

The objective of this paper was to establish if wheat resistance to low temperatures has improved over the years as a result of breeding. At the same time, we wanted to determine if the low temperature resistance of wheat varieties commercially grown in the country at the present time is sufficient for successful production.

MATERIAL AND METHODS

A two-year trial with 23 wheat varieties from different breeding periods was carried out during 2003-04. The varieties were divided into four groups based on the year of their release. The first group included mostly foreign introductions such as Banatka, Bankut 1205, San Pastore, Bezostaja 1 and Libelula. The second comprised domestic varieties grown on larger areas (Sava, Zlatna Dolina, Partizanka, NSR-2 and KG 56). The third consisted of varieties that were dominant in the 1980s (Balkan, Jugoslavija, Zvezda, Skopljanka and Lasta). The fourth and final group included the majority of the most widely grown commercial varieties in our country at the present time (Evropa 90, Pobeda, NSR-5, Renesansa, Zlatka, Ljiljana, Sonata and Vila).

Resistance to low temperatures was determined in cold chamber at -15°C after 24, 36, 48 and 60 hours (HRISTOV e*t al*., 1997). The field/lab method of testing tolerance towards low temperatures is a combination of field conditions for plant development and laboratory conditions (low temperatures) for the testing of said tolerance. Sowing is done in the open in early October. The seeds are planted in plastic containers (R=0.22m , h=0.2m) in a circular pattern at a distance of 2 cm from the edge of the pot. In each treatment, 3 x 20 seeds of each cultivar are planted. The soil with which the containers are filled is taken from a field subjected to normal preparation for sowing and fertilizer application. This ensures that the plants are grown in conditions similar to those in the field. At the two-to-three-leaf stage, the plants are thinned so that every other plant remains in the container with sufficient living space. The plants develop under the influence of environmental conditions until they are transposed to the chamber in December for the purpose of resistance testing. In case of snow, the pots are covered with plastic foil. Cold chamber testing is conducted at -15ºC in a series of treatments lasting from 24 to 72 hours. To avoid temperature shock, the temperature is lowered to -15ºC gradually. At the tillering node depth, the temperature is about -11.5ºC on average. After the completion of the treatment, the temperature is gradually increased and the plants are left at 0ºC for 24 h. Afterwards, they remain in the greenhouse at 10-12ºC for another five days, after which the extent of winterkill is assessed visually on a scale of 0 to 10 (0 – no winterkill, 10 – complete winterkill of leaf area). Subsequently, the plants are exposed to 20-22ºC and irrigated to facilitate the regeneration of the surviving plants. Once this is over, the number of surviving plants is determined and a measurement is made of the longest part of the surviving leaf (LPSL), which is directly correlated with the leaf survival rate (LSR) of each plant individually. The calculation of LSR from LPSL is done using a scale (Tab.1) proposed by MISIC (1965).

Changes of this trait occurring in the varieties from the different breeding periods were monitored through percent winterkill and leaf survival rate using linear regression (MALETIC 2005).

Tab. 1. Scale for determining leaf survival rate after exposure to low temperature

LPSL (cm)	10	9.9	9.8	9.6	9.4	9.1	8.6	8.0	7.3	6.5	5.5	4.5	3.4	2.6	1.9	1.3	0.9	0.6	0.3	0.1	0
LSR (%)	100	95	90	85	80	75	70	65	60	55	50	45	40	35	30	25	20	15	10	5	0

RESULTS AND DISCUSSION

Conditions for hardening were satisfactory in 2003/04 (Year I). Starting at the beginning of October, the mean daily temperature decreased gradually up until the moment the plants were placed in cold chamber in the second half of December. The only exception was the third ten-day period of November, when the temperature rose to about 10°C for a few days (Fig.1).

Figure 1. Mean daily temperatures in 2003 and 2004
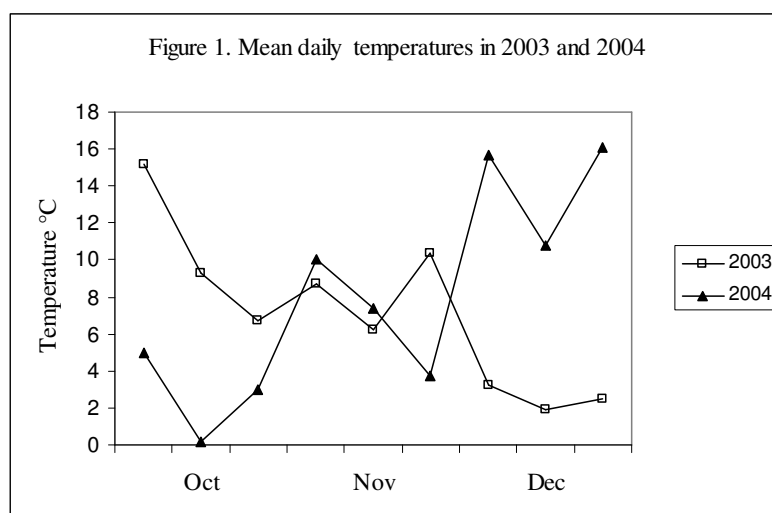
The cold chamber tests of the 2004/05 growing season (Year II) were preceded by extremely low temperatures in October, moderately low ones in November, and extremely high ones in December (Fig.1). These unfavorable temperatures first resulted in slowed-down emergence, poor rooting and insufficient tillering and then in very poor hardening and preparation of the plants for winter.

In the first year of the study, the favorable hardening conditions resulted in a plant survival rate of 90.5%. The unfavorable temperatures of the second year led to increased plant susceptibility to low temperature effects and a decline in the plant survival rate (85%) after the cold chamber test (Tab.2).

Table 2. Percent overwintering success of wheat plants during two years (2003-04)

Variety	Year of release	% plant survival rate			% leaf survival rate		
		Year 1	Year 2	Mean	Year 1	Year 2	Mean
Banatka	1930	100.0	95.0	97.5	58.5	46.8	52,6
Bankut 1205	1931	100.0	92.5	96.3	60.6	43.3	51,9
San Pastore	1940	62.5	47.5	55.0	49.9	21.3	35,6
Bezostaja 1	1959	100.0	100.0	100.0	71.4	51.5	61,4
Libelula	1965	92.5	80.0	86.3	58.0	40.5	49,3
Group 1 mean		91,0	83.0	87.0	59.7	40.7	50.2
Sava	1970	95.0	88.1	91.6	53.6	38.0	45,8
Zlatna Dolina	1971	87.5	82.0	84.8	53.1	41.6	47,3
Partizanka	1973	100.0	100.0	100.0	70.8	55.1	62,9
NSR 2	1975	75.0	65.0	70.0	51.3	26.8	39,0
KG 56	1975	100.0	97.5	98.8	58.9	51.0	54,9
Group 2 mean		91,5	86.5	89.0	57.5	42.5	50.0
Balkan	1979	95.0	87.5	91.3	51.3	45.0	48,1
Jugoslavija	1980	75.0	67.5	71.3	58.6	27.8	43,2
Zvezda	1982	100.0	97.5	98.8	68.9	50.8	59,8
Skopljanka	1982	90.0	83.5	86.8	54.4	38.4	46,4
Lasta	1987	97.5	90.0	93.8	60.0	48.0	54,0
Group 3 mean		91,5	85.2	88.4	58.6	42.0	50.3
Evropa 90	1990	87.5	84.0	85.8	53.1	28.0	40,6
Pobeda	1990	95.0	93.4	94.2	58.5	39.5	49,0
NSR 5	1991	70.0	65.5	67.8	46.9	26.8	36,8
Renesansa	1994	80.0	76.0	78.0	49.1	34.3	41,7
Zlatka	1997	92.5	90.0	91.3	64.9	37.8	51,3
Ljiljana	2000	97.5	96.0	96.8	67.3	43.4	55,3
Sonata	2000	87.5	84.0	85.8	53.6	33.9	43,8
Vila	2001	95.0	91.5	93.3	50.8	37.1	43,9
Group 4 mean		88,1	85.1	86.6	51.7	38.9	45.3
Overall mean		90,5	85.0	87.8	56.9	41.0	49.0
$LSD_{0.05}$		0.81	2.93	2.15	0.76	0.82	0.77
$LSD_{0.01}$		1.09	3.92	2.85	1.01	1.10	1.02

Resistance to low temperatures is a variety-specific wheat trait. It is not constant and plants acquire it as they prepare for the winter and go through the process of hardening (GALIBA *at al*., 2000). The longer the transition from high, autumn temperatures to low, winter ones, the better the temperatures become in terms of hardening.

In the first group of varieties, the lowest plant survival rate over the two years on average was found in (55%) and the highest in Bezostaja 1 (100%). In the second, third and fourth groups, the poorest and best performers were NSR-2 (70%) and Partizanka (100%), Jugoslavija (71.3%) and Zvezda (98.8%), and NSR-5 (67.8%) and Ljiljana (96.8%), respectively (Tab.2).

Thanks to intensive breeding, outstanding progress has been made in the first domestic varieties (second group) with regard to all the production/technological characteristics (MLADENOV e*t al*., 2006), resistance to low temperatures included (89%) (Tab.2). However, after the considerable initial increase in the resistance, a decrease in plant survival rate was observed in the varieties of the newer generations. This was in large part due to the poor overwintering success of the varieties from the fourth group in the first study year, whereas in the second the resistance levels of the varieties from the third and fourth groups were the same (Fig 2).

Figure 2. Overwintering of plants
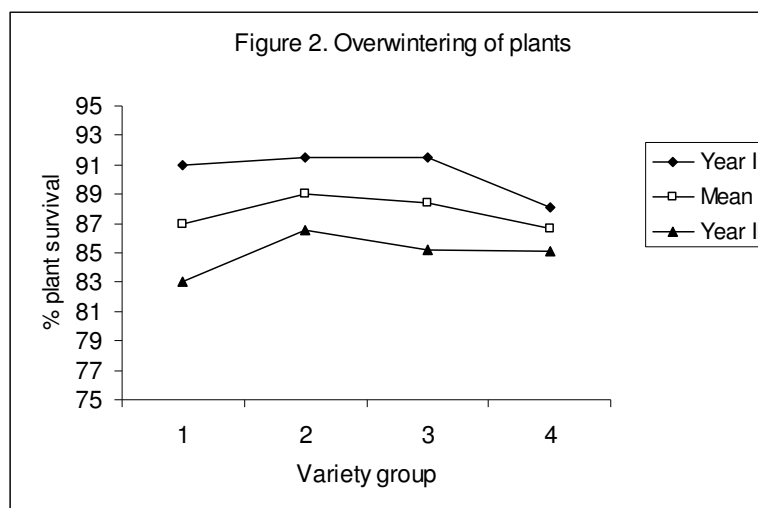
Since resistance to low temperatures is determined first and foremost by genes as the units of heredity (FOWLER *at al.,* 1999; HRISTOV e*t al.*, 1998; MLADENOV e*t al*., 1998; SUTKA 2001) and also by plant growth and development and environmental effects during the growing season (LEVITT 1980), there was less variation in plant survival rate between the study years in the varieties from the

fourth group (Tab. 2). This indicates that, despite having lower tolerance of low temperatures, these varieties have greater stability of the trait concerned. Unfavorable environmental conditions can greatly disrupt the normal functioning of a plant organism, if said organism is unable to resist such effects. Using their genetically determined ability for adaptation to agroecological conditions (FOWLER & LIMIN 1997), varieties from the fourth group are able to adapt to and overcome environmental stress conditions without any major detriment to yield and quality as the most important agronomic traits.

One of the goals of most breeding programs worldwide is to maintain resistance to low temperatures in the commercial varieties at its existing level (BRAUN & SAULESKU 2002). In the varieties from our study, resistance to low temperatures has remained at practically the same level, as the increase of said ability as expressed by the regression coefficient was found to be 0.3%/variety (Fig. 3). This should come as no surprise given the fact that the pedigrees of almost all newer wheat varieties include the varieties Bezostaja 1 and Partizanka, which have an excellent overwintering ability. Although these two genotypes belong to earlier breeding periods, their genetically determined resistance has made it possible for this trait to be passed on to the progeny as well. To what extent a given trait is expressed depends on a large number of factors (VEISZ & SUTKA 1990). It is important, however, that the degree of this expression should be such as to enable successful growing in the agroecological area concerned.

Figure 3. Linear regresion for plant overwintering
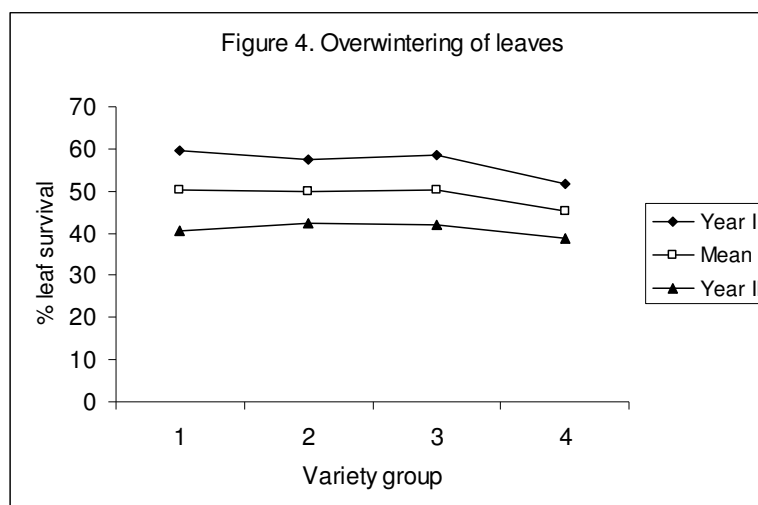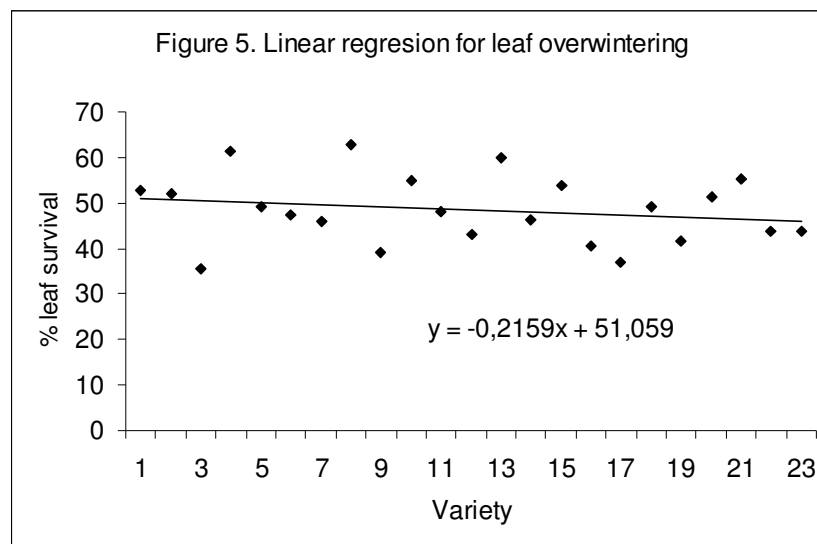
$$y = 0,0313x + 87,211$$
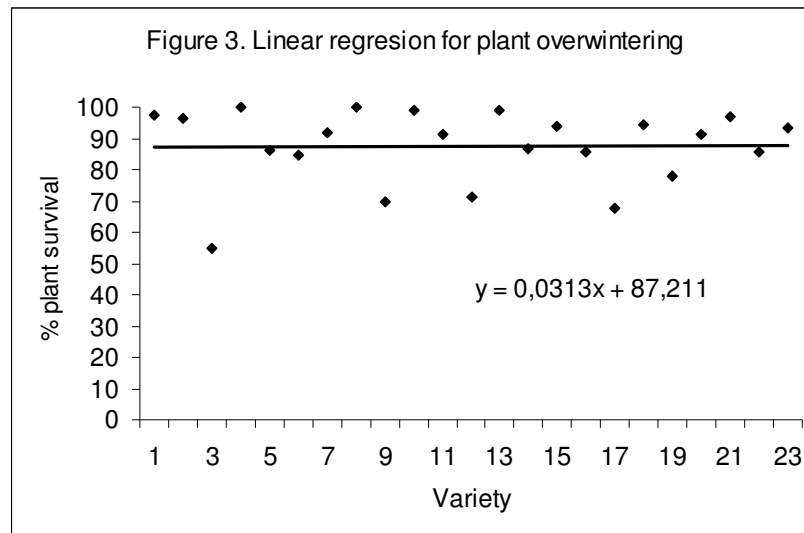
Leaf survival rate serves as an auxiliary indicator of resistance to low temperatures. Leaf and plant survival rates are positively correlated, as more resistant genotypes suffer less leaf damage, primarily as a result of the more rapid

expulsion of water from the cell into the intercellular space (GUSTA *at al.*, 1975). In some genotypes, however, more intensive growth and tillering in the earlier stages of development result in a more luxuriant above-ground plant mass, which is more susceptible to winterkill. Use of this parameter as an independent indicator of the resistance could lead to an incorrect conclusion. The absolute values of the leaf survival rate do not pertain directly to the winterkill of the tillering node itself, i.e. to the survival of the plant as a whole (HRISTOV *et al.*, 1998a).

In the present study, there was a much greater difference between the two study years with respect to leaf survival rate (57.5 and 39.4%, respectively) than with regard to plant survival rate. The warm December of the second year was favorable to leaf area development, but such conditions had negative influence on the preparation of plants for the winter. The highly luxuriant plant mass containing large amounts of free water in the cells suffered greater damage, which did not have a major impact on the plants' ability to survive the winter. The destructive effects of low temperatures resulted in some different genotype responses within the four groups of varieties analyzed in the paper. In the first and third groups, the ranking of the varieties according to their resistance levels remained unchanged. In the second group, the varieties Sava and Zlatna Dolina switched places. The most changes were observed in the fourth group with its more recently developed varieties (Tab.2), which is indicative of the large influence of environmental factors.

The average leaf survival rate was balanced in the first three groups but significantly reduced in the fourth (Fig. 4). The regression coefficient shows a decrease of 2 %/variety (Fig.5). Definitely contributing to this was the use, especially in the newer varieties, of divergent germplasm originating from areas in which resistance to low temperatures is not a limiting factor of crop production.

Figure 4. Overwintering of leaves

Figure 5. Linear regresion for leaf overwintering

$$y = -0{,}2159x + 51{,}059$$

Leaf damage has definite negative influence on wheat development later in the season (spring) but does not need to play the decisive role in the fulfillment of genetic potentials for yield and quality. The regeneration ability, which is highly prominent in some genotypes, can considerably mitigate the damage caused by low temperatures (LIMIN & FOWLER 2006). Good regeneration ability and rapid growth under favorable conditions are especially characteristic of spring small grains varieties (RONCEVIC & HRISTOV 2000). With this in mind, it has been observed that winter genotypes whose genetic backgrounds include varieties from a temperate climate most often exhibit excellent regeneration capabilities as well.

Favorable conditions for hardening in the first year of the study contributed to the better overwinteriing success of the plants relative to the second year. With both indicators of resistance to low temperatures that we studied in the paper, however, varieties from the fourth group exhibited greater susceptibility when tested in the cold chamber. As varieties from this group were all released between 1990 and 2001 and their development began in the 1980s, the effects of global warming and changes in microclimatic conditions have become visible. Thanks to the absence of long and severe winters and the genetic adaptation to the environmental conditions, the targeted levels of resistance to low temperatures have now become somewhat lower in wheat breeding. (HRISTOV & MLADENOV 2004). According to DENCIC e*t al*. (2006), the grain yields and technological quality achieved by Novi Sad wheat varieties in commercial production have

shown that their existing cold resistance levels are more than sufficient for the agroecological conditions in which they are grown.

CONCLUSION

Over the two-year study period, the greatest resistance to low temperatures was exhibited by the varieties Bezostaja 1 and Partizanka. Among the newer genotypes, high resistance was found in the variety Ljiljana.

There were significant differences among the four variety groups studied with regard to the trait concerned. In the first group, which was mostly made up of foreign introductions, the average plant survival rate across treatments was 87%. The different origins of the varieties from this group were a significant factor behind the differences in their resistance levels. The second group, comprised of the first domestic varieties, had an average plant survival rate of 89%, while the third, containing the dominant varieties from the 1980s, had a plant survival rate of 88.4% on average. In the fourth group, consisting mostly of varieties that are currently commercially grown in the country, the average plant survival rate was 86.6%.

Thanks to high temperatures present during the process of hardening, there was greater plant damage and percent winterkill in the second year than in the first. The resistance of the varieties to low temperatures has been maintained at a level enabling successful and stable production under the cold temperature stress conditions of southeastern Europe.

In the process of breeding, certain characters of cultivated plants must be maintained at a satisfactory level while simultaneously improving other traits, most importantly the agronomically significant ones such as yield and quality, which is exactly what has been accomplished with the varieties commercially cultivated in Serbia at the present moment.

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ASPEKTI OPLEMENJIVANJA PŠENICE NA NISKE TEMPERATURE

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I z v o d

Otpornost pšenice prema niskim temperaturama je kompleksnog karaktera, pri čemu pored sorte, do izražaja dolazi i interakcija sa spoljnom sredinom. Pri nepovoljnim uslovima za kaljenje biljaka, i otporne sorte mogu ispoljiti veću osetljivost prema niskim temperaturama. U dvogodišnjem periodu (2004-2005), izvršeno je testiranje u hladnim komorama na temperaturi –15˚C u trajanju od 24, 36, 48 i 60 časova. Analizirano je 23 sorte pšenice koje su podeljene u četiri grupe, u zavisnosti od godine priznavanja. U prvoj grupi, u kojoj se uglavnom nalaze introdukovane sorte, procenat preživelih biljaka na svim tretmanima iznosio je u proseku 87%. Različito poreklo sorti koje se nalaze u ovoj grupi, značajno je uticalo na međusobne razlike u otpornosti prema niskim temperaturama. U drugoj grupi, koju čine prve domaće sorte, procenat preživelih biljaka iznosio je 89%, dok su u treću gupu uvrštene sorte koje su bile dominantne 80-ih godina, sa 88,4% preživelih biljaka. Većina sorti koje čine četvrtu grupu, trenutno je zastupljena na proizvodnim površinama u našoj zemlji. Kod ovih sorti, procenat preživelih biljaka iznosio je 86,6%. Visoke temperature u periodu kaljenja biljaka, uslovile su veća oštećenja, odnosno veći procenat izmrzlih biljaka u drugoj godini ispitivanja. Otpornost sorti pšenice prema niskim temperaturama, zadržana je nivou koji obezbeđuje uspešnu i stabilnu proizvodnju u uslovima hladnog temperaturnog stresa jugoistočne Evrope.