

**PECULIARITIES OF WINTER BARLEY BREEDING
IN RELATION TO LOW TEMPERATURE TOLERANCE**

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Abstract

Mihova, G. and T. Petrova, 2007. Peculiarities of winter barley breeding in relation to low temperature tolerance.

Low temperatures are a main limiting factor for barley production in North Bulgaria. The resistance to this type of abiotic stress is an extremely complex and dynamic character. The necessity to evaluate a number of indices in various combinations additionally complicates both the theoretical investigations on this problem and the breeding process. Increasing frost resistance is a crucial moment in the barley breeding program of Dobroudja Agricultural Institute. As a result from purposeful research work considerable genetic variability was developed. Lines with higher yield stability under stress were selected. A higher correlation was established between low temperature tolerance and winter growth type, poor growth vigor in spring, medium to prostrate growth habit later date to heading and higher plant height. The use of various breeding material and the careful selection in the hybrid population allowed breaking these correlations.

Key words: Barley – Breeding traits – Abiotic stress – Frost resistance

Резюме

Михова Г., Т. Петрова, 2007. Особености в селекцията на зимния ечемик във връзка с толерантността към ниски температури.

Ниските температури са основен лимитиращ фактор за производството на ечемик в Северна България. Устойчивостта към този вид абиотичен стрес е изключително сложен и динамичен признак. Необходимостта от оценка на редица показатели в различни комбинации допълнително усложняват, както теоретичните изследвания по проблема, така и селекционния процес. Повишаването на студоустойчивостта е основен момент в селекционната програма на ечемика в Добруджански земеделски институт. В резултат на целенасочена работа е създадено значително генетично разнообразие. Отбрани са линии, които притежават по-голяма стабилност на добива при стресови условия. Установена е по-силна зависимост на толерантността към ниски температури и зимния тип на развитие, по-бавния темп на развитие през пролетта, междинна до разстлана розетка, удължаване периода до изкласяване и по-голяма височина на растението. Използването на разнообразен изходен материал и внимателното провеждане на отбора в хибридна популация дава възможност тези връзки да бъдат разчупени.

Ключови думи: ечемик, селекционни признаци, абиотичен стрес, студоустойчивост.

INTRODUCTION

The rate and growth of grain production are directly related to the changes in the main climatic factors and the recent more frequent extreme conditions. The unfavorable winters are one of the reasons for the decrease of the barley sowing areas in North Bulgaria. Developing varieties tolerant to low temperatures is a possibility for its stabilization. The main problems in breeding for winter hardiness improvement are the following: the extremely poor world gene pool and the small number of sources suitable for developing genetic diversity; the variety of forms of winter resistance occurrence and the complexity of the genetic systems which determine it; the strong genotype x environment interaction; difficult selection under stress conditions due to changes in the heritability regularities; negative correlations with valuable biological and economic indices. The most common reason for the poor wintering of barley as compared to other cereals is its low cold resistance.

The aim of this investigation was to present the main problems in the developing of initial material and breeding of barley varieties with increased frost resistance.

MATERIAL AND METHODS

The study was carried out during 2003 – 2006 at Dobroudja Agricultural Institute – General Toshevo. Thirty-two breeding barley lines selected among fifteen hybrid populations of different systematic affiliation were subjected to investigation (Table 1). Two groups were formed. The first group included lines with production potential and frost resistance similar to that of the standard varieties; the second group involved lines with increased frost resistance. The two-rowed barley lines of var. *nutans* and var. *erectum* were compared to the standards Obzor, Emon and Kaskadyor 3, and the fodder barley of var. *pallidum* and var. *parallelum* were compared to Veselets and Izgrev. The selection for frost resistance was based on the field evaluation from 2005-2006 and the data obtained from the laboratory testing in freezing chambers by the method of Tsenov and Petrova (1984). The trial was designed within the framework of the preliminary varietal trial, in one replication, the plot area being 10 m². Sowing was accomplished within the period recommendable for North-East Bulgaria. The sowing norm of the two-rowed lines was 400 germinating seeds, and of the poly-rowed lines – 430 germinating seeds. Eight plants were analyzed from each line, and the characters were evaluated by the methods of IPGRI (1994) and UPOV

Table 1. Investigated cross combinations and systematic affiliation of selected lines

№	Cross combinations	Systematic affiliation of selected progeneis	Number of lines
1	Skorohod x BIC 1176	subsp. <i>vulgare</i> var. <i>parallelum</i>	2
2	Hemus x Kozyr	subsp. <i>vulgare</i> var. <i>parallelum</i>	2
3	Knyajich x Secret	subsp. <i>vulgare</i> var. <i>parallelum</i>	2
4	Bastion x Izgrev	subsp. <i>vulgare</i> var. <i>parallelum</i>	2
5	Vavilon x Vesletc	subsp. <i>vulgare</i> var. <i>pallidum</i>	2
6	Makas x Hemus	subsp. <i>vulgare</i> var. <i>pallidum</i>	2
7	Radical x Miraj	subsp. <i>vulgare</i> var. <i>pallidum</i>	2
8	Secret x Toman	subsp. <i>vulgare</i> var. <i>pallidum</i>	2
9	Ruen x H-N-01	subsp. <i>distichum</i> var. <i>nutans</i>	2
10	M-1-94-5 x 104-207	subsp. <i>distichum</i> var. <i>nutans</i>	2
11	Laura x 386-117	subsp. <i>distichum</i> var. <i>nutans</i>	3
12	Ruen x Laura	subsp. <i>distichum</i> var. <i>nutans</i>	2
13	M-1-94-5 x Laura	subsp. <i>distichum</i> var. <i>erectum</i>	3
14	103-493 x H-N-01	subsp. <i>distichum</i> var. <i>erectum</i>	2
15	Ruen x 104-207	subsp. <i>distichum</i> var. <i>erectum</i>	2

(2003). The previous crop was grain peas. The agrotechnical activities, which are not considered in this paper, were performed according to the cultivation practices accepted for this crop.

The statistical processing was realized with the help of the software STATISTICA, version 7.0 for Windows XP.

RESULTS AND DISCUSSION

The combination of meteorological conditions during the years of study allowed evaluating the breeding materials with regard to their production potential and some types of biotic and abiotic stress.

In all three harvest years the autumn conditions favored the normal germination and development of barley. The hardening stages occurred under favorable conditions. Year

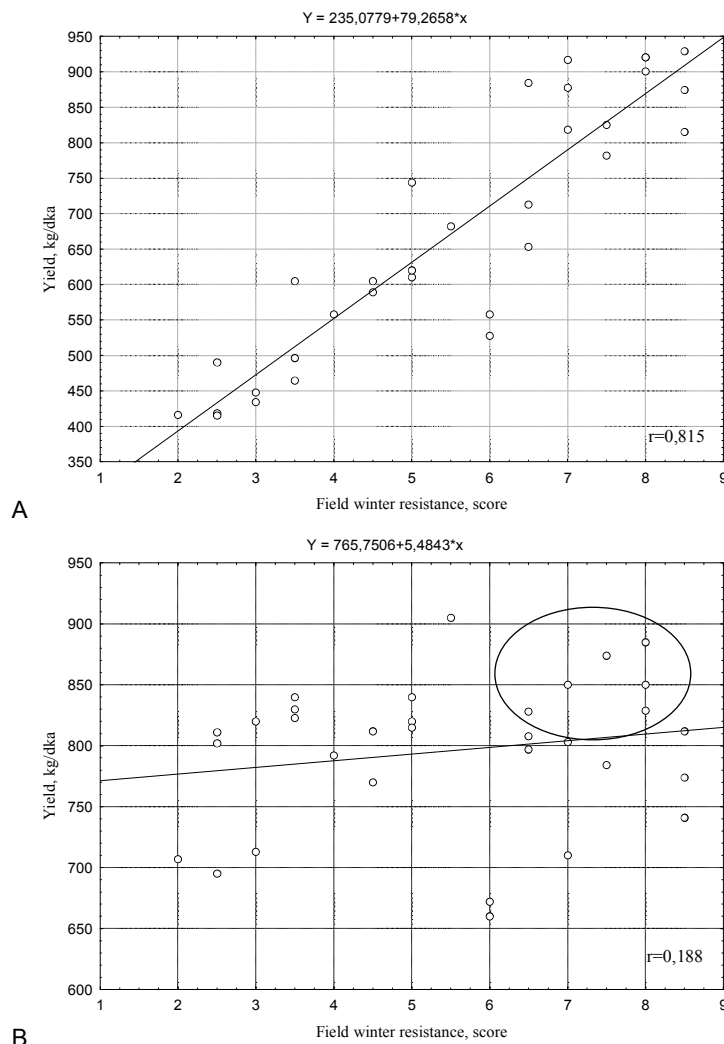


Figure 1. Relationship between grain yield (kg/dka) and field winter resistance (score) under stress (A) and non stress (B).
Field winter resistance, score: 1-very low, 5-moderate, 9-very high

2003/2004 proved most successful for barley production. The rainfalls in May and June were decisive for the formation of high yields. The low temperatures registered in the winter of 2004/2005 did not in practice damage the crops since they were accompanied with snow cover. The low radiation temperatures reaching up to -13° ч -14° C were typical for the first decade of April. Simultaneously the air temperature at 2 p.m. was above 10° C. Some discrepancy resulted between the abundant leaf mass formed in autumn and the capability of the roots to provide sufficient water and nutrients. The response of the individual barley biotypes differed radically. The winter type barleys entered active vegetation with difficulty in spite of the longer daylight. On the whole, the mean daily temperatures were within the biological minimum required for the booting stage. As a result, greater differences in the date to heading were observed among the individual biotypes, as compared to previous years. The winter conditions during 2005/2006 were rather severe. At the end of January the absolute minimum temperatures were as low as -20° C, without snow cover. The period of critical low temperatures lasted about a week. Furthermore, during the first decade of February, a second cold period occurred with minimum temperatures up to -16.5° C, again without snow cover. These conditions allowed differentiating the investigated lines with regard to their frost resistance under field conditions. Besides the low negative temperatures, the ice crust formed in consequence also affected the further survival of plants. The direct damages it caused were weaker, however, due to the fact that it affected only the surface soil layer. Spring was favorable for secondary booting and many varieties compensated for the losses. The meteorological conditions following heading, during the period of grain filling and maturation, contributed to the formation of high yields.

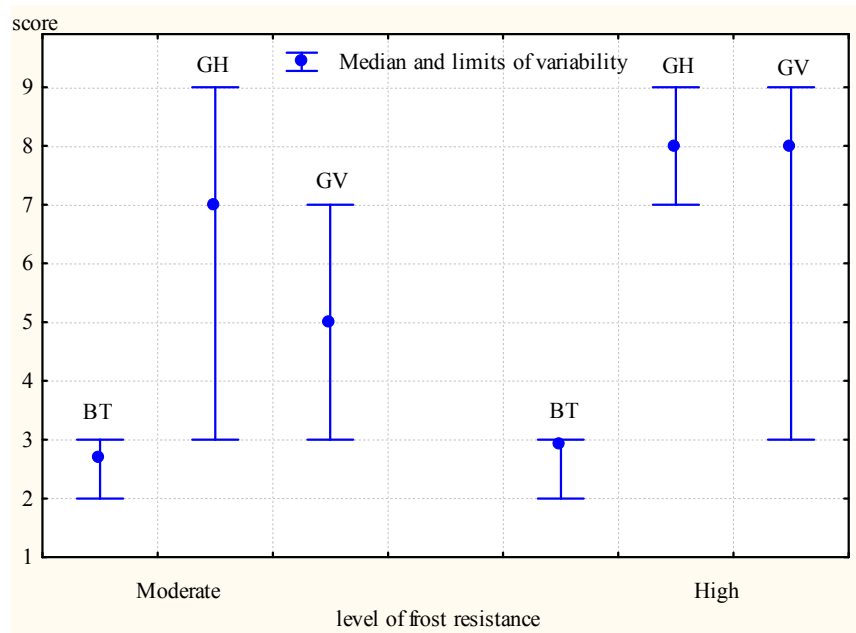


Figure 2. Variation of the biological type of development (BT), growth habit (GH) and growth vigour (GV) in lines of different frost resistance.

BT: 1 spring, 2 facultative, 3 winter

GH: 1 erect, 5 intermediate, 9 prostrate

GV: 1 very high, 5 moderate, 9 very low

Increasing the adaptability of the varieties is one of the possibilities to obtain stable yields from the individual crops. Developing varieties tolerant to various types of stress is

a main task in many breeding programs. The effect of environment on the phenotypic expression of the genetic potential can be summarized in the following three ways (according to Hoffman and Merila, 1999): genotypic differences are expressed only under unfavorable conditions; genotypic differences are expressed only under favorable conditions; norms of reaction are closest under intermediate conditions and diverge towards increasingly favorable and unfavorable conditions. While comparing the regression models of the functional correlation between the yields from the produced lines and the field winter resistance, significant differences were observed (Figure 1).

Under stress, very high correlation was established. The yield formed correlated directly with the field evaluation reading. Without stress, the correlation coefficient was low and insignificant. The analysis of the results showed that a part of the lines with increased winter resistance were responsive to favorable environments. The yield formed was high and comparable to the varieties with wide distribution.

The breeding materials from the two groups – with moderate and high winter resistance were characterized by some peculiarities, as well (Figure 2).

Biological type. The type of development is determined by complex genetic systems related to vernalization duration and photoperiod susceptibility. These are the two major mechanisms controlling development of cereal plants during ontogenesis and having a key role for their adaptability to environmental complications (Mahfoozi, 2001). In barley, the major photoperiod response genes are located on the short arm of chromosome 2H (*Ppd-H1*) and the long arm of chromosome 1H (*Ppd-H2*). Day length-sensitive *Ppd-H1* promotes flowering in response to increasing day length. The second gene *Ppd-H2* affects flowering under short days but has little effect under long days (Laurie, 1997). Three genes are responsible for vernalization requirement: *Vrn-H1* on chromosome 5H, *Vrn-H2* on chromosome 4H and *Vrn-H3* on chromosome 1H. Most genotypes are monomorphic at *Vrn-H3*, reducing the genetic model to two factors (Yasuda et al., 1993; Laurie et al., 1995). Alleles at the two loci show varying degree of dominance and epistatic interaction. Recent research indicates that vernalization requirements influence the expression of low temperature tolerance genes in cereals (Hayes et al., 1993; Snape et al., 1997). A significant correlation between frost resistance and winter growth habit in wheat has been reported by Brule-Babel and Fowler (1988). According to Shevstov et al. (2003), the combination of various methods for developing genetic variability allows selection of facultative barley varieties with good frost resistance.

In the group of moderate frost resistance, a similar number of lines from the two biotypes were selected, the median being close to the mean arithmetic. Winter lines were predominant in the second group, which implies a more difficult combining of high frost resistance and facultative type of development. However, the variation of this character demonstrates that in case of careful selection of parental forms the combination of the characters is not impossible.

Growth habit (tillering stage). A correlation has been reported between prostrate growth habit and freezing tolerance in wheat. A gene controlling prostrate growth was found to be closely linked with *Fr 1* and *Vrn 1* on chromosome 5A (Roberts, 1990). According to Saulescu and Brun (2001) the correlation is not high enough to use this character as a criterion of winter resistance evaluation. Averaged for the period of investigation, there was a wider variation of the character in the lines with lower frost resistance. In both groups most genotypes were with semi-prostrate and prostrate growth habit. The results were in accordance with a purposeful selection carried out for the character in relation to the tasks of the barley breeding program in DAI (Mihova and Petrova, 2005a).

Early growth vigour. Greater variability of materials was established in the group of lines with increased winter resistance. The character median was drawn towards the upper limit of variation, i.e. the lines with slower growth vigour following winter were predominant. This was an expected result since most lines from the group were of winter growth type. These lines are characterized with longer vernalization period, higher photoperiod

susceptibility, and vegetation in spring is resumed after permanent temperature increase. Ceccarelli et al. (1991) established that the genotypes tolerant to low temperature are with poor early growth vigour. Nevertheless, these authors recommend not to consider this correlation absolute and to search for optimal combinations of characters determining resistance under stress. Mersinkov (2000) reported a relation between the fast growth vigour and the formation of higher protein and lower extract content, which should be taken into account in breeding of malting barley. Lower was the variation of this character in the group with higher productivity and lower frost resistance. The genotypes with moderate values were predominant.

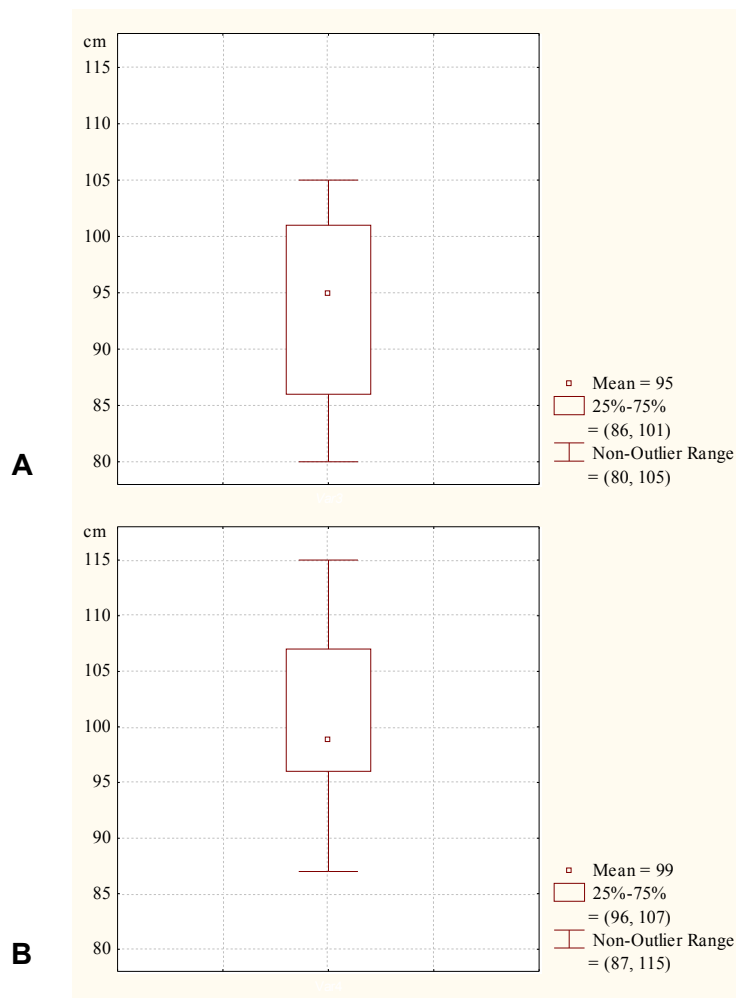


Figure 3. Variation of plant height in lines with moderate (A) and increased (B) frost resistance.

Plant height. One of the problems in breeding for frost resistance is lodging (Shevstov et al., 2004). Degree of lodging is most often related to plant height. Comparing the values of this character, a significantly higher mean value and limits of variation were observed in the group of frost resistant lines (Figure 3). The greater part of them had plant height 96-107 cm. These are high values, especially for 6-row barley which is more inclined to lodging. The results confirmed the positive correlation found by Ceccarelli et al. (1991) be-

tween plant height and resistance to different types of abiotic stress. The considerable variation shows that variability with regard to this character was developed and that selection of forms with desirable values is possible, as well as search for forms with strong and elastic stems.

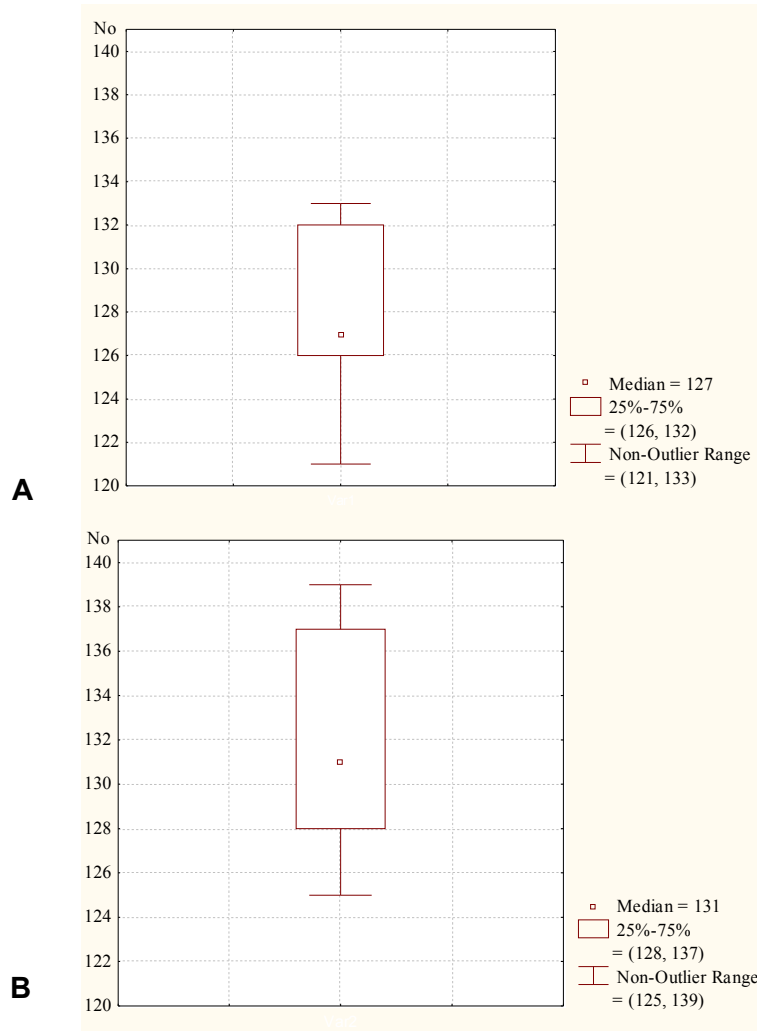


Figure 4. Variation of days to heading* in lines with moderate (A) and increased (B) frost resistance.
*as from 1st January

Days to heading. Analyzing a hybrid population produced by crossing winter x spring barley, Griffiths et al. (1997) identified five main genes and eight QTL loci with lower effect, controlling flowering. The main genes were those determining the requirements to vernalization duration, photoperiod susceptibility and one denso dwarfing gene related to retardation of flowering. The eight QTL are relatively unaffected by conditions and termed “*earliness per se*” genes. The investigations on them are of certain interest because their effect is not related to daylight length and the temperature regime. The variation of the character in the groups was similar; in the group of frost resistant varieties, however, the median was significantly higher (Figure 4). The later heading was again related to the fact

that winter growth type lines were predominant in this group. Usually the lines with heading 2-3 days later than the standard varieties give higher yields in the competition trials. This affected to a certain degree the course of selection. The longer duration of the period is not desirable due to the risk of soil drought and high air temperatures during grain filling.

Structural elements of yield. The genetic investigations on barley have revealed approximately 67 QTL peaks related to abiotic stress resistance (Hayes et al., 2003). The biggest cluster of genes determining low temperature tolerance was localized on chromosome 7. The observations showed that besides the higher percent of surviving plants, the general regeneration ability of the genotype was also important for the end productivity. Achieving high values of the structural elements of yield is difficult due to the negative correlations between them. On the other hand, this increases its adaptability under stress during certain stages of the phenological development. No difference was established between the variations of some yield components in the individual groups of lines with different frost resistance (Table 2). Previous investigations showed that under stress correlations between yield components were changed (Mihova and Petrova, 2005b).

Table 2. Variation of yield's structural elements in lines of moderate and high frost resistance

Level of frost resistance	subsp. <i>vulgare</i> var. <i>parallelum</i>			subsp. <i>vulgare</i> var. <i>pallidum</i>			subsp. <i>distichum</i>		
	min	ma	Vc	min	ma	Vc	min	ma	Vc
Spike per plant (number)									
Moderate frost resistance	2.00	3.50	22.13	2.50	3.50	13.75	3.00	4.50	14.62
High frost resistance	2.00	3.50	24.15	2.50	4.00	14.80	3.00	4.00	12.75
Spike length (cm)									
Moderate frost resistance	4.18	4.97	17.3	5.35	7.02	18.5	6.86	7.85	19.3
High frost resistance	4.22	5.21	19.2	5.20	6.94	19.1	6.55	7.98	20.2
Kernels per spike, (number)									
Moderate frost resistance	52.4	59.7	12.1	42.5	49.9	14.1	20.8	26.2	26.8
High frost resistance	53.1	58.1	13.1	40.8	50.2	15.4	21.8	24.1	23.7
1000 kernel weight, g									
Moderate frost resistance	25.7	35.5	9.16	25.8	42.4	12.0	32.3	52.9	11.7
High frost resistance	23.6	32.4	7.52	27.1	43.7	10.5	34.7	51.1	15.9
Harvest index (%)									
Moderate frost resistance	39.4	51.9	27.1	40.2	52.1	19.7	41.1	53.4	21.7
High frost resistance	40.8	52.7	23.4	41.1	54.2	15.2	44.5	55.7	15.6

Harvest index. Harvest index is among the more general criteria for yield evaluation. It includes not only the morphological peculiarities of the plant but also some physiological and biochemical indices. This allows a more efficient assessment of the interaction between the genetic system and the environmental factors.

The differences in the character variation were mostly between the individual systematic groups. Between the groups of different frost resistance they were not significant. It should be taken into account when analyzing the results that the combination of meteorological

logical conditions during the years of investigation was rather various. The summarizing of the obtained data reduces to some extent the effect of the stress conditions.

The mean values of the yield's structural elements and the harvest index of the group of lines with increased frost resistance showed that breeding material has been developed which combines good production potential and tolerance to low temperatures.

CONCLUSIONS

Considerable genetic variability was developed as a result from purposeful work on increasing frost resistance of barley.

Lines of different systematic affiliation were selected which possess higher stability of yield under stress conditions.

A higher correlation was established between low temperature tolerance and winter growth type, poor growth vigor in spring, medium to prostrate growth habit later date to heading and higher plant height.

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