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ACHIEVEMENTS IN WINTER PEAS BREEDING PROGRAM

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Abstract

The development of the winter pea crop represents a major challenge to expand plant protein production in temperate areas. Breeding winter cultivars requires the combination of freezing tolerance as well as high seed productivity and quality. In this paper we present data obtained from the F3 and F4 lines of winter peas from the four hybrid combinations (Specter/F95-927; F98-492/Windham; F95-927/CHECO; Specter/CHECO) obtained in theNARDI-Fundulea. A number of 121 lines (81 F3 lines and 40 F4 lines descendants from F3*) selected from winter/winter and winter/spring crosses pea genotypes, have been tested in two years 2015 and 2016 in the field of NARDI Fundulea. At these lines were determined winter hardiness, earliness, yield and plant height comparing with the winter peas control (Specter, Checo and Windham). The results of this preliminary study suggested that it is possible in the breeding of winter peas to realize, in the same time, a genetic progress for high yield, good level of winter hardiness, plant height and earliness.*

***Key words**: winter pea, breeding, winter hardiness, yield.*

INTODUCTION

Field pea (*Pisumsativum L.*) is one of the most important grain legumes in the world. Its grain is a major source of plant-based dietary protein for animals.

Kosev (2010) said that field pea can provide protein-rich feed and improve the sustainability of organic systems. The share of agricultural land that is under organic agriculture approaches 4% in EU and 7% or more in Scandinavian countries, Italy, Austria and Greece, and it may reach 25% in EU by 2030.

Field pea continues to be an important crop worldwide both for food and feed and as a rotational crop with other cultures. The pea breeding programs are based on yield and yield components. Pea grain yield is a quantitative trait which is affected by many genetic and environmental factors.

Cultivars adapted to winter sowing have been developed and deployed in Europe and north-west USA giving the potential for better yields because of a longer growing season, higher biomass production, and earlier maturity to avoid late season drought and heat stress (Hanocq et al., 2009). The introgression of the *Hr* allele, which delays flower initiation until after the main winter freezing period shave passed, permitted to obtain some cultivars with notably improved winter hardiness (Lejeune-Hénaut et al., 2008).

Delayed floral initiation helps some forage pea genotypes to escape the main winter freezing periods, as susceptibility to frost increases during the transition to the reproductive state (Lejeune-Hénaut et al. 1999). Numerous studies describe the physiological and phenological effects of the main loci governing the transition to flowering in pea, such as *Lf* and *Hr*, known to delay floral initiation of autumn-sown peas until a longer day length is reached in the following spring (Lejeune-Hénaut and Delbreil 2009). The oldest winter pea cultivars carry the dominant allele, *Hr*, but some of them have *hr* allele (Bourion et al. 2002). A study of one population of recombinant inbred lines (RILs) allowed detection of six quantitative trait loci (QTL) for frost tolerance, which is in agreement with oligogenic determinism of frost tolerance in pea. In this population, the most explanatory QTL was found to colocalize with the *Hr* locus (Lejeune-Hénaut et al. 2008). Further studies in the same genetic background gave an insight in the genetic determinism of physiological traits potentially involved in cold acclimation, showing for example the colocalization of QTLs for raffinose concentration or RuBisCO activity with QTLs for frost tolerance on linkage groups 5 and 6 (Dumont et al. 2009).

Winter peas have a good level of frost resistance, and are also characterized by a large foliage development in spring which favours lodging and fungal diseases in humid conditions (Lejeune-Heaut et al., 1999).

In temperate climate, winter crops are the majority andfrost damage is an important factor reducing crop yields, especially in regions where winter is regularly severe (Biarnès et al., 2016). The winter forage pea varieties are suitable for arid regions. The utilization of winter forage pea in the conditions with a high water deficit, results in higher and more stable aboveground biomass and protein yields, enhancement of the ratio between symbiotic and fertilizer nitrogen in organic farming and the possibilities for more economic use of the agricultural land (Castel et al., 2017).

Winter peas may soon become an alternative increasingly more reliable and profitable crop canola.Genetic variation hasbeen reported for vegetative-stage frost tolerance (Swensen and Murray, 1983; Bourionet al. 2003), and there has been some genetic analysis of differences for frost tolerance, and six chromosomal regions affecting winter frost tolerance were identified. Genetic analysis of frost tolerance is a prerequisite for the development of lines that are tolerant to frost at reproductive stages (Shafiq, 2012).

The aim of this work was to appreciate the yield performance and other traits, mainly the winter hardiness of several winter pea lines in the climatic conditions from NARDI-Fundulea.

MATERIALS AND METHODS

In this study the plant material used initially consisted in 19 lines from each the four hybrid combinations (Specter/F95-927; F98-492/Windham; F95-927/CHECO; Specter/CHECO) and 5 parents, tested in two years (2015 an 2016) at NARDI Fundulea.

In 2015, the first 81 F3 peas lines were tested in four preliminary micro-cultures, each of them with 25 variants, one rep besides checks (parents of these lines: Specter, Windham, Checo, F95-927 and F98-492), on a plot of 6m^2 harvested area.

In 2016, 40 lines of peas F4 generation (descendants from F3 generation presented above) were tested in preliminary micro-cultures, each of them with 25 variants, one rep besides (parents of these lines), on a plot of 6m^2 harvested area. The lines from F4 generationare more advanced and more stable than lines from F3 generation.

The two years of testing were climatically different, so that, 2015 winter was slight enough, with a short period with negative temperatures of - 20°C (the end of January), with a snow layer of 20 cm which has protected the crop. There are no damages registered due to frost. The early spring was normal, fact that led to restart the vegetation under optimum conditions.

In contrast to the 2015, the 2016 winter was more sever, with a short period with negative temperatures of - 14,5°C (the begining of January), without snow, which realised a good differentiation of genotypes regarding the winter hardiness. The early spring, allowed the restart the vegetation in the begining of February, with an average air temperature higher with 9,5 °C than normal during 50 years. Under these conditions, the peas genotypes were highly differentiated versus other years, regarding flowering precocity depending their reaction to daylight. The varieties peas originating from USA, Specter and Windham cultivars, with a requirement of a longer photoperiod – 12 hours/day - did not passed to the generative stage, in spite of the high temperature registered in the bigining of February.

The level of resistance to winter hardiness was estimated in the field, early in the spring, in a scale 1 to 9, where score 1 is very resistance and 9 very susceptible. Plant height was measure in cm, total length of plant from the ground till the top to the end of flowering time. The earliness was appreciated like number of days from 1st January till the end of flowering time and yield as kg/ha.

The statistical analyses of data have been evaluated by ANOVA, correlations and linear regressions between study traits.

RESULTS AND DISCUSSIONS

Yield performances of 40 peas lines of F4 generation and 81 lines F3, tested in advance trials and respectively in preliminary trials in 2015 and 2016, (Figure 1 A and B) showed a large variation.

Among of F3 lines (bulk) 27% of them and 31% from the F4 lines, have achieved superior yields than the winter checks, Windham, Specter and Checo. There are several lines which realized 4.5 till 5 t /ha. That mains it is possible to be realize an important genetically progress for yield from such germplasm.

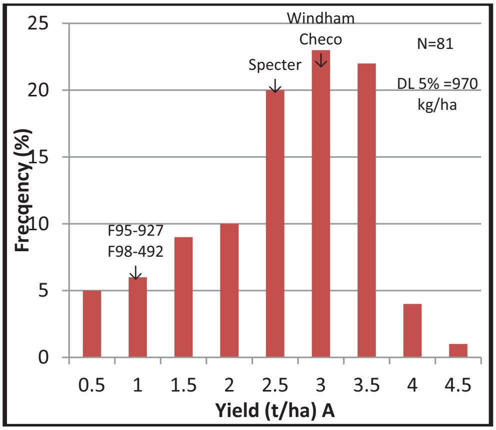

Figure 1.A. Distribution of 81 F3(A) winter pea lines after their yield performnces (t/ha) in 2015

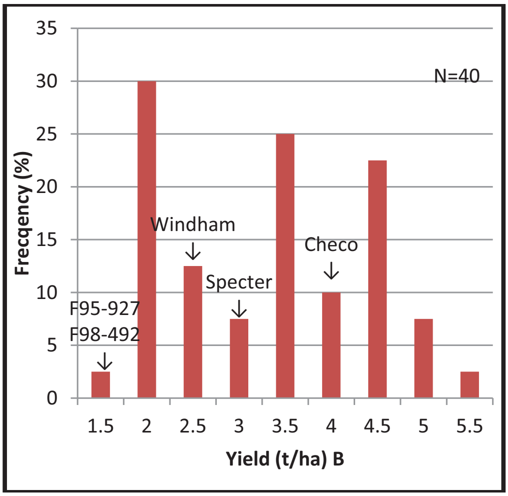

Figure 1.B. Distribution of 40 F4 winter pea lines after their yieldperformances (t/ha) in 2016

Also, it is very important to select the perspective lines with good yield potential but in the same time to recombine an acceptable earliness for Romanian climate conditions. The distribution of the 40 of F4 lines for precocity (Figure 2), divides the breeding material in two groups: late forms which take this trait from American peas cultivars Windham and Specter, and early forms similar with spring peas parents F98-492 and F95-927 and respectively winter peas cultivar Checo.

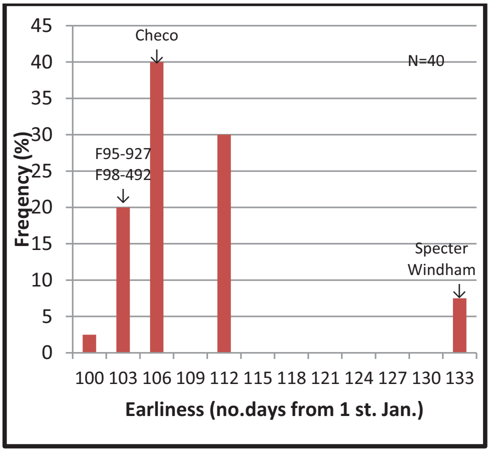

Figure 2. Distribution of 40 F4 winter pea lines after their earliness

Based on molecular assay, using molecular marker AA175, the parents used in this study have split into two groups: Specter and Windham carry *Hr* allele and Checo, F95-927, F98-492 carry *hr* allele (data not shown).

According to this distribution, the most of the lines, 82.5% approximately, were in the early group.

The distribution of the 40 lines from F4 generation for plant height (Figure 3) indicated the presence of pea lines with differentplant height, from 80 cm till 210 cm. Most of them are shorter than tallest parent Specter, only a few have been the same plant high like Specter. What it is interesting that many of the lines had intermediate plant height, between short plant parents, Windham and Checo and height plant parent Specter. That mains the possibility to realize the new varieties with different plant height, that being important for end use of this varieties, for seed or for silage.

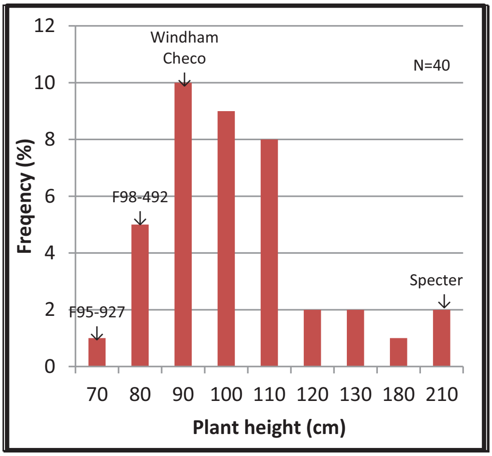

Figure 3. Distribution of 40 F4 winter pea lines after their plant height (cm)

The data for winter hardiness presented in figure 4, suggest that it is easier enough to recover the winter hardiness level like winter parents, in the many lines, even from the winter/spring crosses. From the distribution of the 40 F4 lines, it is possible to see that 60% of them are the same level of winter hardiness like winter parents, Checo, Windham and Specter.

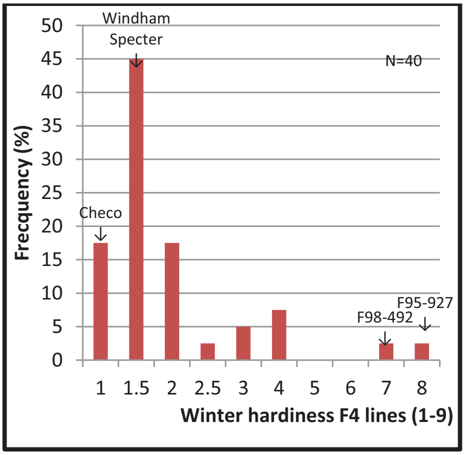

Figure 4.Distribution of 40 F4 winter pea lines after their winter hardiness

Having in view that winter hardiness in winter peas is a very important trait, there was necessary to highlight in what way this trait could be recombined with other important agronomical characteristics, likeearliness to flowering, plant height, grain yield as well as the relationship among other traits as plant height/earliness or yield/earliness. Correlations between different characteristics in 81 F3 lines from 2015 and 40 F4 lines from 2016 were presented in Table 1.

The correlation between winter hardiness and yield in F3 and F4 lines was very distinct significantly negative (r=-0.54 and r=-0.60) (Table1 and figures 5 A and 5 B), what means that in winter peas is absolutely necessary to cultivate genotypes with good level of winter hardiness, to realize high and stable yields.

Also relationship between plant height and earliness should be positively strong enough in some case, what mains that it quite easily to be recombine such characteristics. The correlation, between plant height and winter hardiness, was only in F4 lines negative significantly (r=-0.32), but in the F3 lines, was not significant, that suggests possibilityto select the genotypes which recombine both traits.

Table 1. Correlation coefficients among different traits in F3 and F4 lines of the four hybrid combinations

The generation genotypes	**Correlation between different characters**	**The correlation coefficient**
81 F3 lines tested in micro-cultures in 2015	Winter hardiness/yield	**-0,54*** **
	Winter hardiness/earliness	-0,05ns
	Winter hardiness/plant height	-0,02ns
	Plant height/earliness	**0.45*** **
	Yield/earliness	0.19ns
40 F4 lines tested in micro-cultures in 2016	Winter hardiness/yield	**-0.60*** **
	Winter hardiness/earliness	-0.21ns
	Winter hardiness/plant height	**-0.32***
	Plant height/earliness	0.28ns
	Yield/earliness	**-0.32***

The relationship between yield and earliness, in F3 and F4 peas lines was insignificant and negative significant, (0.19 and -0.32), what mains that later types can realize high yield than earlier types.

Also, the relationship between winter hardiness and earliness, in the all cases, was not significant, that mains it is too easy to select the winter pea form with both traits.

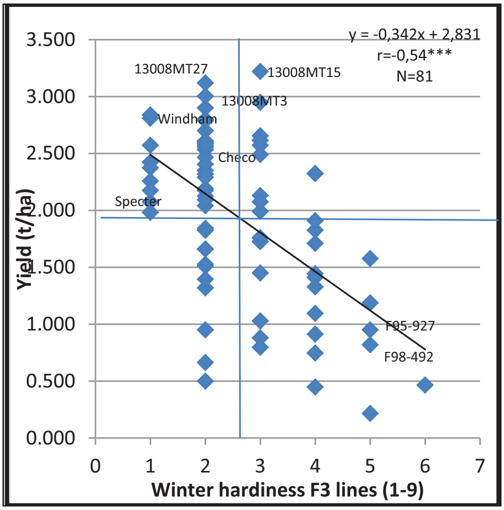

Figure 5.A. Relationship between winter hardiness and yield data of 81 F3 lines

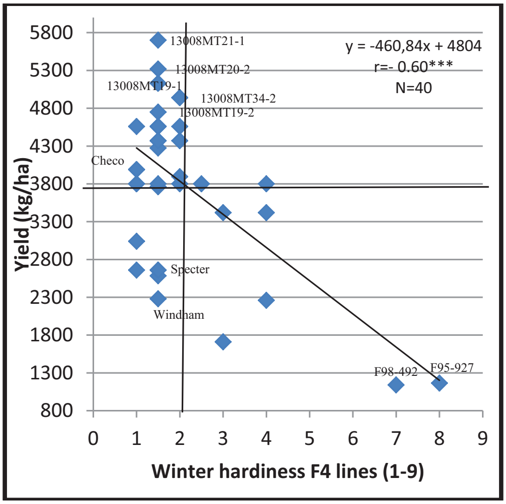

Figure 5.B. Relationship between winter hardiness and yield data of 40 F4 lines

The correlation distributions between yield and winter hardiness of 81 F3 lines and of the 40 F4 lines tested in 2015 and 2016 respectively, presented in Figure 5A and5B, indicated, in the both cases, that there are lines with the same level of winter hardiness like winter parents but with higher yield than this. Such lines are from combination hybrids F95-927/Checo, from F3 were 13008MT2, 13008MT1 and 13008MT5 and from F4, 13008MT21-1, 13008MT20-2, and 13008MT19-1.

CONCLUSION

The data obtained in this study on the F3 and F4 random lines from four hybrid combinations (Specter/F95-927; F98- 492/Windham; F95-927/CHECO; Specter/CHECO) suggested the possibility to achieve simultaneously a genetic progress for high yield, good level of winter hardiness, plant height and earliness, useful in the breeding of winter peas.

REFERENCES

Bourion V, Fouilloux G, Le Signor C, Lejeune-Hénaut I (2002) Genetic studies of selection criteria for productive and stable peas. *Euphytica*127: 261–273.

Castel T., Lecomte C. ,Yves Richard, Lejeune-Hénaut I., Larmure A., (2017) Frost stress evolution and winter pea ideotype in the context of climate warming at a regional scale, OCL, 24 (1) D 106.

Dumont E, Fontaine V, Vuylsteker C, Sellier H, Bodèle S, Voedts N, Devaux R, Frise M, Avia K, Hilbert JL, Bahrman N, Hanocq E, Lejeune-Hénaut I, Delbreil B (2009). Comparative studies of frost tolerance QTL in *Pisum sativum*L. under field and controlled conditions using physiological and phenotypical traits.*Theor. Appl. Genet.*118:1661-1571.

Hanoq E., Jeuffroy, M, Lejeune-Hénaut I, Munier-Jolain N. (2009) Construire des ideotypes pour des systemes varies en pois d´hiver. Innov.Agron. 7, 14-28.

Kosev V., Pachev I., (2010) Genetic Improvement of field pea (Pisumsativum L.) in Bulgaria.Field Veg. Crop Res. 47, 403-408.

Lejeune-Henaut I, Delbreil B (2009) Genetics of winterhardiness in pea.*Grain Legume*52:7-8.

Lejeune-Hénaut I, Bourion V, Etévé G, Cunot E, Delhaye K, Desmyter C (1999) Floral initiation in field-grown forage peas is delayed to a greater extent by short photoperiods,than in other types of European varieties. *Euphytica*109:201-211.

Lejeune-Hénaut I, Hanocq E, Béthencourt L, Fontaine V, Delbreil B, Morin J, Petit A, Devaux R, Boilleau M, Stempniak J-J, Thomas M, Lainé A-L, Foucher F, Baranger A, Burstin, Rameau C, Giauffret C. (2008) The flowering locus Hr colocalizes with a major QTL affecting winterfrost tolerance in Pisumsativum L. *Theor Appl. Genet* 116:1105-1116.

Shafiq S, Mather D, Ahmad M, Paul J (2012) Variation in tolerance to radiant frost at reproductive stages in field pea germplasm. Euphytica 186 (3): 831-845.

Biarnès V, Lecomte C, Lejeune I, Hascoët E. (2016) Pois d'hiver: la resistance au froid bientôt décryptée. PerspectAgric 43: 32–35.