EVALUATION AND UTILISATION OF LATVIAN FLAX GENETIC RESOURCES IN BREEDING

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Abstract
Repatriated flax accessions of Latvian origin from the N. I. Vavilov Institute of Plant Industry (Russia), the Flax Research Institute (Russia) and the Institute of Plant Genetics and Crop Plant Research (Germany), and best flax lines breed in the Agricultural Science Centre of Latgale were investigated. Several qualitative and quantitative traits, such as technical plant height, number of seed vessels, logging resistance, vegetation period, yield of straws and seeds, bast fibre, oil content and quality were evaluated. Results of three year field trials were summarized. Some local genotypes and lines were better than the standard fibre flax variety ‘Vega-2’ and the standard oil flax variety ‘Līrīna’ in several agriculturally important traits. For intensification of the breeding process we were looking for the possibility of using biotechnology methods for obtaining additional flax breeding source material. After cultivation in calli culture regenerated plants could perform to a higher the so-called somaclonal variation. We applied this method to the Latvian local line ‘Rīgaer B’. Best explants, medium compositions as well as calli growing and regeneration conditions were detected.

Key words
Flax, breeding, genetic resources, calli culture, antioxidants

Introduction
Before World War II, Latvia was among the biggest flax exporters in the world, and flax was growing on more than 60,000 ha. Flax breeding started in Latvia in 1923. At least six varieties were bred in Latvia by selection from the best local and Russian landraces. After World War II, flax growing was not considered as an important economic resource for the Latvia region, and therefore, the flax area was gradually reduced. In 1970, flax breeding in Latvia was cancelled. Fibre flax breeding was started in Latvia again in 1992 (Rashal and Stramkale, 1998), oil flax growing and breeding was started again less than ten years ago. On the first stage field trials on more than 400 foreign varieties was performed, nevertheless high yielding early-ripening flax varieties suitable for growing in the changeable Latvian weather conditions were not found. Therefore both the Latvian Gene Bank of Cultivated Plants and the Agricultural Science Centre of Latgale started repatriation of the flax accessions of Latvian origin. More than 40 accessions were received from several foreign collections: the N. I. Vavilov Institute of Plant Industry (Russia), the Flax Research Institute (Russia) and the Institute of Plant Genetics and Crop Plant Research (Germany).

Flax is a crop with a rather low variability of agriculturally important traits and the creating of new varieties by using traditional breeding methods is time consuming. Nowadays an actual task is applying the biotechnology approach to intensify the plant breeding process. One of widely used methods for this purpose is inducing somaclonal variation in regenerated plants from calli culture. Somaclonal variation has been described for many plant species (Larkin and Scowcroft, 1981; Leike, 1985; Brar and Jain, 1998; Rutkowska-Krause et al., 2003). Flax plants with resistance to biotic and abiotic stress, increased plant height and increased number of seeds were found among the somaclones (Poliakov, 2000; Rutkowska-Krause et al., 2003). It shows that the in vitro method could be useful for obtaining additional source material for flax breeding to facilitate the flax breeding process.
The aim of this study is the evaluation of repatriated local flax genetic resources, identification among them of useful accessions for breeding, and elaborating optimal conditions for calli inducing and regeneration from the best local flax breeding material.

**Materials and Methods**

Evaluation. The evaluation of 40 repatriated flax accessions and the 10 best fibre flax breeding lines of the Agricultural Science Centre of Latgale was done in field experiments (random block design, two replications) and in laboratory experiments during 2004 – 2006. Accessions were compared with the standard fibre flax variety ‘Vega 2’ or with the standard oil flax variety ‘Lirina’. Agriculturally important traits, such as technical plant height, number of seed vessels, logging resistance, vegetation period, yield of straws and seeds, bast fibre, as well as oil content and quality, including balance of unsaturated fatty acids (Jodine number), were determined.

Calli culture. Latvian landrace ‘Rigaer B’ with excellent adaptation to Latvian climatic conditions, good fibre content and seed yield but with too small technical plant height were used to form calli cultures. Calli cultures were established by a method elaborated earlier (Grauda et al., 2005). Leaves and stem fragments (length 0.5 cm) were used as explants. All explants were placed on the Murashige and Skoog (MS) basal medium with 1.5 mg/l 2,4-D (dichlorophenoxyacetic acid) (added after autoclaving).

After four weeks of cultivation for inducing shoot formation calli were placed on the MS medium with 2 mg/l BAP (bensilamiopurine) (Nichterlein, 2003, Rutkowska-Krause et al., 2003). The shoot formation medium after autoclaving was supplemented by one of the following antioxidants: ambiol 1 mg/l, AV-153 1 mg/l, ascorbic acid 1 mg/l, Triovit 1.5 mg/l or the flax seed extract (5 g of seeds were milled and added to 30 ml 35 ºC water, soaked for 30 min and filtered). MS medium supplemented only by 2 mg/l BAP was used as the control. More than 20 calli were placed on each variant of the shoot formation medium. Cultures were grown in the light conditions (16/8 h light/dark, 24 ºC). After four weeks of cultivation on the shoot formation the medium the weight of calli were measured and after eight weeks of cultivation calli regeneration capacity (percent of calli formed regenerants) was recorded.

**Results and Discussion**

Evaluation. Some repatriated accessions and breeding lines were better in several agriculturally important traits (technical plant height, number of seed vessels, number of seeds in a seed vessel, 1000 seeds weight, yield of straws and seeds) in comparison with the standard variety. Most of them showed high logging resistance. The vegetation period (2005. year) was from 76 to 83 days (standard variety ‘Vega 2’ – 83 days).

As it was determined earlier (Grauda et al., 2004) the most important trait for fibre flax breeding is the plant technical height. Four of the investigated repatriated accessions (‘Vietējais 1’, ‘Ošupes 30’, ‘Ošupes 31’, ‘Riger B’) had a rather high value of this trait similar to the fibre flax standard variety ‘Vega 2’ (Fig. 1) (only the best 8 accessions and 6 breeding lines are shown). Those accessions could be suggested as a good initial breeding material for fibre flax breeding. The best fibre flax breeding lines (K15-8/2-13-95 and L11-11/10-97) selected from crosses between repatriated accessions and height yielding foreign varieties, have a statistically significant higher technical height than the standard variety. The bast fibre were ranged depending on the genotype and year of growing from 25.4% (‘Riga Vilnorin’) to 38.9% (T36-26/4-8-94), each of the ten evaluated breeding lines had in average a higher content of bast fibre (30.9 – 36.0 %) than the variety ‘Vega 2’ (29.0 %). The yield of straws was higher than the standard variety (395 g/m²) for all breeding lines and for two repatriated accessions - ‘Ošupes 30’ (400 g/m²) and ‘Riga Originario’ (408 g/m²).
The number of seed vessels for the evaluated material were from 5 to 7 seed vessels, but for two accessions (‘Blue di Riga’ - 11 and ‘Riga 27/12’ - 9) it was significantly higher than the mentioned value. Accessions with better or similar seed yields (Fig. 2) in the three years in the comparison of oil flax standard variety ‘Lirina’ were chosen for the oil content and quality determination. Oil seeds content (Fig. 3) differed depending on the annual conditions. In 2005, they were more favorable for oil flax growing (a sunny summer) and local genotypes (‘Vietējais 1’, ‘Rigaer B’), similarly as the standard variety ‘Lirina’, had higher oil content in comparison with 2004. The Iodine number varied from 143 (‘Rigaer B’) to 186 (‘Blue di Riga’), which is close to the standard variety ‘Lirina’ (193). ‘Rigaer B’ with a good and stable seed yield and oil content is suitable for use as an oil flax variety for oil producing with a low content of unsaturated fatty acids. ‘Blue di Riga’ can be used for the oil production with a high content of unsaturated fatty acids.
Figure 3. Oil content in seeds of flax accessions of Latvian origin in 2004 and 2005

Calli culture. Calli were obtained from both used explant types. Calli formation started two weeks after the culture initiation. Stem segments and leaves developed homogeneous calli structures.

For obtaining a higher number of plants-regenerants the influence of different antioxidants on calli growing and regenerating capacity was tested. After four weeks of growing on a shot formation medium with antioxidants calli induced from leaves were, in general, larger than stem calli (Fig. 4). On calli formed from leaves, only the influence of the synthetic antioxidants ambiol and AV-153 were positive. Ambiol and ascorbic acid have a positive influence on stem calli weight (calli weight was 47 mg heavier on the medium with ambiol and 82 mg heavier on the medium with ascorbic acid). Calli growing was dependent on the interaction of the genotype, explant type, and the used antioxidant.

Figure 4. Weight of calli grown on shot formation medium with different antioxidants for 4 weeks depending of explant type

Formation of plants-regenerants started after five weeks of calli cultivation. Regeneration zones on a medium without antioxidants were formed only by stem calli. The use of antioxidants ambiol and AV-153 initiate the regeneration also from calli leaves (Fig. 5). The best results were obtained when antioxidant ascorbic acid or flax seed extract were added to the medium: the regeneration capacity of stem calli was increased up to 5 times. For all genotypes shoot formation was observed after 8 weeks of calli cultivation.
The use of the most appropriate source material is always crucial to achieve breeding goals. The old plant genetic resources usually are characterised by good adaptation to the local conditions but loose to the modern varieties in yield capacity, quality, resistance to new pathogen races etc and therefore could not be used directly in contemporary agriculture. A solution can be found in crossing the best local and modern varieties to combine the favourite characteristics of both materials. Our results of the evaluation of the Latvian flax genetic resources shown, that some of them had very good performance in yield and adaptability. Four genotypes were suggested as good for fibre flax breeding and two genotypes for oil flax breeding in Latvian conditions. Some hybrids obtained from crosses among local genotypes and best foreign varieties were better in several agriculturally important traits in comparison with the standard variety.

Calli culture could be very useful for the broadening of source material for breeding. Formation of flax calli culture is possible from various parts of seedlings (roots, hypocotyls, cotyledons, stems, leaves). Depending on the genotype and explant type the rate of calli formation ranged from 50 to 90% (Poliakov, 2000; Rutkowska-Krause et al., 2003; Yildiz and Özgen, 2004). Our results supported earlier findings that stem fragments and leaves were useful for obtaining flax calli cultures. For the first time it has been demonstrated that the addition of antioxidants or flax seed extracts could significantly increase the shoot formation capacity and can by this broaden the spectrum of initial genotypes for inducing somaclonal variation.

**Conclusion**

Four genotypes were suggested as good for fibre flax breeding and two genotypes for oil flax breeding in Latvian conditions. Some hybrids obtained from crosses among local genotypes and the best foreign varieties were better in several agriculturally important traits in comparison with the standard variety.

Used explants were excellent for calli culture formations (100% of the used explants developed calli). The development of regeneration zones were depended on both from the used explant type and antioxidant flax seed extract but did not depended on the calli weight. By using a the appropriate additions (an antioxidant or flax seed extract) to the shoot formation medium the calli regeneration capacity can be significantly increased.

**References**


ANALYSIS OF SWARD MANAGEMENT FACTORS INFLUENCING Festulolium AND Lolium x boucheanum YIELD FORMATION

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Abstract
Under conditions of the Latvian climate, forage grasses are the main fodder source in cattle breeding. Festulolium hybrids are among the most persistent and productive grasses of the grasses used in many Europe countries, especially in adverse environments. The aim of this research was to investigate dry matter yield formation and the sward persistency of Festulolium and Lolium x boucheanum varieties under the agro-ecological conditions of Latvia. Field trials were established on the sod–podzolic soil and fertilized with N 120 (60+60), N180 (60+60+60), P 78 and K 90 kg ha⁻¹. Forages were harvested three times during the growing season. During the three years of utilization the dry matter yield for festulolium and ryegrasses swards was reliably (P<0.05) dependent on the used variety as well in the nitrogen fertilization rate. The N fertilizer dose increase from 120 to 180 kg ha⁻¹ contributed to significantly to the DM yield increase for all investigated varieties. On average the N fertilizer dose increase to 180 kg ha⁻¹ contributed to DM yield increase by 1.91 t ha⁻¹ or 20 percent. On the basis of the experiments in the years 2003-2006, significant differences in DM yield and winter hardiness were found between first, second and third year of yields. Dry matter yield was found to be strongly dependent on climatic conditions in the particular year of yields and the particular period of regrowth. Analysis of yield distribution between three cuts showed that a year of sward use had a very great effect on the DM yield of the first cut. For the first year it accounted for 51% of the annual yield, in second year the first cut yield reach only 39 % of the annual yield.

Key words
Festulolium, Lolium hybridum, productivity, regrowth, photosynthesis.

Introduction
Many producers of dairy husbandry in Latvia are shifting towards intensive systems of production. The slow growing grass species in grasslands have been replaced by species and cultivars with rapid regrowth and high yield potentials. In this regard perennial ryegrasses are gaining popularity in the Baltic region as they are tolerant to intensive management, but are susceptible to drought and have rather poor winterhardiness (Aavola 2005). In Baltic climate