Results of the study of the modern gene pool of narrow-leaved lupine

P A Ageeva, N A Potchutina, M V Matyukhina and O M Gromova

Federal Williams Research Center of Forage Production & Agroecology, All-Russian Lupin Scientific Research Institute, Berezovaya Street, 2, p/o Mitchurinsky, Bryansk, Russia

Corresponding author’s e-mail: lupin_mail@mail.ru

Abstract. Narrow-leaved lupine (Lupinus angustifolius L.) is a valuable leguminous fodder crop adapted to a wide range of soil and climatic conditions. Modern fodder varieties of narrow-leaved lupine have a low content of alkaloids in the grain of 0.02-0.10%, a high protein content of 32.0-38.0% and a number of other nutrients valuable for fodder production. In the collection nursery 2018-2020 Under the conditions of a changed climate, forage varieties and specimens of two morphological groups were studied: branched with different degrees of blocking of lateral branching and spike-like. The results of the structural analysis of the gene pool for the main elements of productivity and its morphobiological features are presented. According to the productivity of plants in the indeterminate group, the varieties USN 53-236, Narrow-leaved 53-02 and Hybrid 1215 (7.7-6.1 g / plant) were distinguished; by weight 1000 pcs. seeds - varieties Belorozovy 144 and Crystal (136.4-120 g); by the number of seeds in a bean - Belorozovy 144 and Narrow-leaved 53-02 and the Belarusian cultivar Alliance (4.1-4.2 pcs.). In the group of spike-like forms, the cultivar Epigonal 1215 (3.2 g / plant) was distinguished by grain productivity, by the weight of 1000 seeds - the Belarusian cultivar Talent (102.2 g). As a result of the assessment of the collection material in terms of the height and weight of plants, a variety meter was identified that exceeded the control, variety Vityaz, by 15.0-27.7 cm: Belorozovy 144, SN 78-07 and Hybrid 1215. The growing season of the studied numbers varies in a wide range of 80-101 days. The group of spike-shaped numbers is characterized by ultra-early maturity (71-75) days.

1. Introduction

Narrow-leaved lupine (L. angustifolius L.) is an annual, highly leafy herb. Stem erect, partially full, slightly pubescent; the number of leaves along the main stem varies from 18 to 40. The color of the flowers: blue, purple, pink, white-pink, pink-white, white. Seeds are medium, round, rounded-oval. The peel of the seeds is smooth, the color is different: white, creamy without a pattern and with a varied pattern, gray-mottled, rusty, red-brown, black [1]. Number of chromosomes: 2n - 40. Wild forms of narrow-leaved lupine are bitter and contain more than 2% alkaloids in seeds. Selection feed and food varieties and varieties have a low alkaloids content of 0.02-0.10%, protein 28-40%, fat 4-8%.

The narrow-leaved lupine grows wild in countries around the Mediterranean. Narrow-leaved lupine, represented by modern varieties, is the earliest of the large-seeded lupine species introduced into culture. The length of the growing season varies from 80 to 110 days, depending on the cultivar, weather conditions and the level of soil fertility. In terms of the rate of initial growth, it compares favorably with...
other species. It lacks the phase of the basal rosette, therefore, immediately after the cotyledons emerge on the soil surface, active growth of the stem begins. The most favorable conditions for obtaining a high yield of narrow-leaved lupine seeds are provided at an average daily temperature of 16 ... 17 °C and 200 ... 250 mm of precipitation for the period from germination to maturation. The northern border of the area of its cultivation for seeds corresponds to 580, and of determinate spike-like varieties - 600 north latitude [2].

The specificity of the culture is that it has a high nitrogen-fixing ability. Thanks to symbiotic nitrogen fixation, lupine provides itself and enriches the soil with biological nitrogen. In symbiosis with nodule bacteria (Rhizobium lupine), lupine is capable of fixing up to 300 kg of atmospheric nitrogen per hectare of sowing, therefore it is distinguished by its relative undemanding to soil fertility. Narrow-leaved lupine gives good yields of grain and green mass on soils with an average level of fertility and a soil reaction of pH 5.0-6.0, (tolerates from 4.5 to 7.0). It is tolerant to low temperatures; the optimum temperature for seed germination is +9 ... +120°C (minimum +2 ... +40°C), the seedlings can withstand frosts down to -70°C. Narrow-leaved lupine is a moisture-loving culture. The optimum soil moisture for the formation of a high yield corresponds to 60-70% of its full field moisture capacity [3]. The narrow-leaved lupine differs from other species by the greatest tolerance to the harmful fungal disease - anthracnose.

Narrow-leaved lupine is a historically ancient culture, but the creation of a gene pool with altered mutant genes began only in the twentieth century. The work on mobilization and comprehensive study of the modern gene pool of narrow-leaved lupine in changing environmental conditions is relevant.

2. Material, methods, conditions
The research material is the varieties and varieties of narrow-leaved lupine of our own selection, as well as those obtained from the VIR and other research institutions of Russia and Belarus, which were studied in the collection nursery in 2018-2020. The counts and observations were carried out according to the methods generally accepted in breeding work. The key points in assessing the gene pool and breeding material is to determine the resistance to cracking of beans and alkaloid. The first trait is determined by the lentus (le) and tardus (ta) genes and is determined by the presence of marker traits; pink color of beans during the filling of seeds and bright yellow inside the valve during the ripening phase. Alkaloidity is constantly monitored by qualitative methods: in the laboratory in winter, Buchard's reagent is used for seeds, in the field, special alkaloid-sensitive paper treated with Dragendorff's reagent is used for vegetative plants. The content of alkaloids in collectible sortonomers promising for the creation of a new starting material is determined by a quantitative analytical method.

When setting up the experiment, we used guidelines for the study of grain legumes [4; five]. Field studies were carried out in the selection crop rotation of the All-Russian Research Institute of Lupine, located in the north-eastern part of the Bryansk region of the Central region. The soils of the experimental plots during the years of study are gray forest, the depth of the arable layer is 22-24 cm, the content of humus – 2,29-2,4%, P2O5 15,1-21,0 mg / 100 g, K2O 14,2- 15,0 mg / 100 g soil, reaction of soil solution pH 5.4.

The meteorological conditions were unfavorable for the realization of the productivity potential of the narrow-leaved lupine, since they did not meet the biological requirements of the crop, which negatively affected the green-mowing and grain productivity.

3. Research results and discussion
The collection under study is diverse in many economic and biological characteristics. By morphotype, the collection includes branched varieties with varying degrees of blocking of lateral branching, paniculate, spike-shaped and fasciated.

There is a wide variety in plant height, color of vegetative, generative organs and seeds. According to the content of alkaloids in the collection nursery, there are low-alkaloid sortonomers with varying degrees of alkaloid and green manure. A feature of the study of the collection of the Institute is the high infectious load of our fields due to the prolonged excessive saturation of the culture. Varieties coming
from VIR or other research institutions suffer greatly and often die from fungal, viral and other diseases. Old varieties are very susceptible to death.

The studied set of genetic resources presented in Table 1 includes fodder varieties and samples of our own selection, as well as Belarusian varieties Mirtan, Guslyar, Alliance, Vanyusha, Talant and the Polish variety Gnome. In the group of branched forms with different degrees of blocking of lateral branching by the number and weight of seeds per plant, the variety narrow-leaved 53-02, USN 53-236 and Hybrid 1215 were distinguished. The number of seeds varies from 64.7 to 69.7 pcs., The weight of seeds - from 6.1 to 7.6 g. The excess to the control, zoned variety Vityaz, was 23.7-33.2% for the first indicator, 5.1-32.7% for the second. Among the spike-shaped forms, the cultivar Epigonal 1215 stood out.

Table 1. Characteristics of collection varieties and varieties of fodder narrow-leaved lupine by productivity elements (2018-2020).

| Narrow-leaved lupine local catalog no. | Variety, variety sample | Seed productivity of the plant | Amount | Weight (g) | Seed bean (pcs.) |
|---------------------------------------|-------------------------|-------------------------------|--------|------------|-----------------|
| Branched forms with varying degrees of blocking of lateral branching | | | | | |
| 1 | Knight, control | 52.3 | 5.8 | 110.7 | 3.8 |
| 2 | Bryansk feed | 46.9 | 5.7 | 123.8 | 3.6 |
| 3 | Belozerny 110 | 43.9 | 4.3 | 100.9 | 3.8 |
| 4 | Change | 48.5 | 5.0 | 102.5 | 3.9 |
| 5 | Crystal | 26.0 | 3.2 | 120.1 | 3.0 |
| 11 | Belorozovyi 144 | 47.6 | 5.8 | 136.4 | 4.1 |
| 12 | Narrow-leaved 53-02 | 64.7 | 7.6 | 116.4 | 4.2 |
| 13 | SN 78-07 | 36.6 | 3.8 | 103.1 | 4.0 |
| 20 | USN 53-236 | 66.1 | 7.7 | 114.7 | 4.1 |
| 53 | Hybrid 1215 | 69.7 | 6.1 | 89.3 | 3.1 |
| 58 | Myrtan | 40.0 | 3.7 | 95.4 | 3.6 |
| 75 | Dwarf | 42.4 | 4.6 | 106.7 | 3.6 |
| 76 | Nemchinovsky 97 | 50.9 | 5.7 | 113.6 | 3.9 |
| 106 | Guslyar | 43.3 | 4.4 | 110.5 | 3.5 |
| 107 | Alliance | 45.0 | 4.6 | 102.2 | 4.1 |
| 108 | Vanyusha | 48.0 | 4.3 | 89.7 | 2.7 |
| Spike-shaped forms | | | | | |
| 93 | Hope, control | 29.8 | 2.7 | 92.6 | 3.5 |
| 94 | Talent | 25.9 | 2.8 | 102.2 | 3.2 |
| 95 | Epigonal 1215 | 35.6 | 3.2 | 77.6 | 3.5 |
| 98 | Fasciant | 32.3 | 3.0 | 79.0 | 3.1 |

The mass of 1000 seeds, being one of the components of productivity, is of great importance for breeding. In the presented group, the varieties Belorozovyi 144, Bryansk kormovoy and Kristall have an excess to the control on this trait (25.7-9.4 g). Among the spike-shaped forms by the mass of 1000 seeds, the Belarusian cultivar Talent stood out. According to literary sources [6], the mass of 1000 seeds of narrow-leaved lupine in the world collection varies from 37 to 244 g. This trait is polygenic, controlled by genes with various effects (additive, dominant, epistatic). It is known that selection for traits that are controlled with a predominance of additive effects has good performance in most cases.

A long-term study of the collection samples, reflecting the intraspecific diversity by the weight of 1000 seeds, showed that the conditions of the year significantly change the severity of this trait, while maintaining the existing difference between genotypes. The number of seeds in a pod plays a significant role in the life cycle of a plant, determining its potential for reproduction. Analysis of the distribution of
the number of seeds in a pod in varieties and varieties of the world collection showed the species diversity in the range of 2.5-7.4 pieces. As a rule, small-seeded wild forms have better dissemination rates. The combination in one genotype of a bean polyseed and an increased weight of 1000 seeds is one of the tasks of breeding work. In the presented table, the pod distribution according to the control is 3.8 pcs. On this basis, the best indicators are in the varieties Belorozovy 144, Uzkolistny 53-02 and the Belarusian variety Alliance. In normally developed central cluster beans of the Uzkolistny 53-02 variety, the appearance of six-seeded beans was noted. Among the spike-shaped forms, the control variety Nadezhda and the sample variety Epigonal 1215 have the best seed distribution.

Lupine narrow-leaved is distinguished by a large polymorphism of traits. The height of plants is important in the selection of lupine, since this indicator, to a certain extent, characterizes the suitability of the variety for modern technologies. Long-term studies of scientists from different countries (Australia, Russia, Belarus) show that the intraspecific diversity of narrow-leaved lupine in plant height includes forms from 25 to 135 cm.

There is an opinion of a number of researchers [7] that further progress in plant breeding will be most successful through an increase in the total biological crop yield by increasing its height. Genotypes with higher linear growth are capable of forming crops of high optical and biological density. For the narrow-leaved lupine, as a fodder green mowing and grain fodder crop, this approach is interesting. The height of plants of the Vityaz variety under the conditions of changed weather conditions is 50.2 cm. The variety Belorozovy 144 has a significant excess to the control (15.0 cm) (Table 2).

| Table 2. The results of the study of the collection of narrow-leaved lupine for some morpho-biological characters (2018-2020). |
|---------------------------------------------------------------|
| Branched forms with varying degrees of blocking of lateral branching |
| Narrow-leaved lupine local catalog no. | Variety, variety sample | Height (cm) | Weight (g) | Vegetation period (days) | Survival (%) |
| 1 | Knight, control | 50,2 | 14,5 | 81 | 90,3 |
| 2 | Bryansk feed | 49,3 | 14,2 | 84 | 81,7 |
| 3 | Belozerny 110 | 55,1 | 12,3 | 83 | 80,6 |
| 4 | Change | 46,3 | 12,4 | 87 | 74,6 |
| 5 | Crystal | 41,1 | 8,1 | 80 | 71,3 |
| 11 | Belorozovy 144 | 65,2 | 17,1 | 92 | 76,6 |
| 12 | Narrow-leaved 53-02 | 47,7 | 18,1 | 88 | 79,4 |
| 13 | SN 78-07 | 64,6 | 12,6 | 91 | 84,8 |
| 20 | USN 53-236 | 47,4 | 19,0 | 87 | 79,3 |
| 53 | Hybrid 1215 | 77,9 | 20,1 | 101 | 63,9 |
| 58 | Myrtan | 43,3 | 9,0 | 93 | 64,8 |
| 75 | Dwarf | 50,2 | 11,7 | 82 | 70,2 |
| 76 | Nemchinovsky 97 | 45,1 | 15,5 | 84 | 73,4 |
| 106 | Guslyar | 51,4 | 12,0 | 91 | 86,2 |
| 107 | Alliance | 43,6 | 11,0 | 91 | 82,4 |
| 108 | Vanyusha | 57,4 | 12,3 | 95 | 33,3 |
| Spike-shaped forms |
| 93 | Hope, control | 41,5 | 6,3 | 74 | 79,4 |
| 94 | Talent | 37,4 | 5,6 | 71 | 67,3 |
| 95 | Epigonal 1215 | 52,1 | 6,7 | 75 | 84,6 |
| 98 | Fasciant | 25,6 | 5,8 | 72 | 58,4 |
The new variety meter SN 78-07 and Hybrid 1215 also stand out significantly in this indicator (64.6 and 77.9 cm). The height of lupine plants largely depends on the rate of initial growth; decreases as it slows down and increases as it accelerates. Sowings of fast-growing varieties of narrow-leaved lupine are less susceptible to the negative influence of weeds, since they shade the soil surface earlier, creating competition with weeds [8]. In the group of spike-shaped plants in height, the cultivar Epigonal 1215 was distinguished. The biological feature of Fasciant is that it is able to form an increased number of reproductive organs in a small area of a thickened peduncle.

In terms of the weight of plants, an excess to the control, along with tall ones, has a variety meter USN 53-236 and Narrow-leaved 53-02 (2.6-5.6 g). They are distinguished by a monopodial type of branching, due to which this indicator increases.

The length of the growing season and the duration of the passage of individual phenological phases are important in the selection of pairs for crossing and in the process of working with hybrid and breeding material. When cultivated in production, early ripening varieties ensure timely harvesting and obtaining high-grade, high-quality seed material. In the collection nursery, spike-shaped varieties and specimens were distinguished by ultra-rapidity. The duration of their growing season from germination to maturation was 71-75 days. In the group of branched forms, the variation of this trait is quite significant (84-101 days). Varieties Bryanskii kormovaya and Belozerny 110, as well as the Polish variety Gnome, ripened 2-3 days later compared to the early ripening control, variety Vityaz.

The survival rate of plants for harvesting in the Vityaz variety was 90.3%. More than 80% preserved Belarusian varieties Guslyar and Alliance and a number of other varieties and varieties. A low level of plant safety (33.3%) for harvesting was shown by the Belarusian variety Vanyusha, which is distinguished by a productive central brush.

4. Conclusion
The collection varieties and varieties that have emerged as a result of the research on economically valuable traits will be used in intervarietal crosses when creating a new source material using the principle of geographically and genetically distant hybridization. As a result of the rational use of the gene pool, the result of breeding work in a changed climate is the creation of new constant sortonomers, which in terms of grain yield exceeded the standard, the Vityaz variety, by 0.84-1.76 t/ha.

Acknowledgments
Financial support from the Ministry of Science and Higher Education of the Russian Federation within the framework of the PFNI RAS theme (AAAA-A19-119122490147-7) “To improve methods of examination, mobilization and DNA typing of genetic resources of forage and fruit plants in order to identify adaptive and economic potential, create sources and donors of economically valuable traits and properties”.

References
[1] Kuptsov N S and Takunov I P 2006 Lupin – genetics, breeding, heterogenous crops (Bryansk: Klintsy Publ) p 576
[2] Takunov I P 1996 Lupin in Russian farming (Bryansk: Pridesen’e Publ.) p 370
[3] Kosolapov V M, Yagovenko G L, Lukashevich M I, Ageeva P A, Novik N V, Mischenkova N V, Slesareva T N, Isaeva E I, Takunov I P, Pimoxova L I and Yagovenko T V 2020 Lupin – breeding, cultivation and use (Bryansk Publ.) p 304
[4] Vishnyakova M A, Buravtseva T V, Bulyntsev S V, Burlyaeva M O, Semenova E V, Seferova I V, Aleksandrova T G, Egorova G P, Yankov I I, Gerasimova T V and Drugova E V 2010 VIR global collection of grain legume crop genetic resources: replenishment, conservation and studying (Methodological guidelines 2nd ed) ed M A Vishnyakova (St Petersburg: VIR) p 140
[5] Stepanova S, Nazarova N, Kornechuk V (SSSR), Xr. Leman (GDR) and Mikolajchik Ya (PNR) 1983 The wide unified COMECON classifier and the international COMECON classifier list of descriptors for the genus Lupinus (L. Leningrad: VIR Publ) p 33
[6] Gladstones J S and Crosbie G B 1979 Wild lupin introduced into Western Australia to 1973; collection site data, preliminary ratings of field characteristics and disease reactions and measurements of seed protein and oil content. *DAWA, Technical Bulletin* No 43 p 44

[7] Starzhiczkij St 1981 Biological base for agricultural plants’ modeling M: Nauka pp 434-439

[8] Ageeva P A, Matyukhina MV, Pochutina NA 2019 Evaluation of narrow-leafed lupin varieties for some morphological and biological characters. *Multifunctional adaptive feed production* 21(69) pp 20-25 DOI10.33814/MAK-2019-21-69-20-25