Development of anthracnose- (Colletotrichum gloeosporioides (Penz.) Penz & Sass.) resistant narrow-leaved forage lupine breeding lines

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Breeding for disease resistance is one of the most difficult tasks in breeding work. Rapid mutation of pathogen populations, their ability to continuously form a large amount of races differing in virulence and aggressiveness are the main obstacles in the development of disease-resistant lupine varieties. In 2001 three, in 2001–2003 seven narrow-leaved forage lupine breeding lines developed by the individual selection methods and characterised by a high resistance (7–9 points) to anthracnose (Colletotrichum gloeosporioides (Penz.) Penz & Sass.), rapid growth rate and high seed yield (2.2–2.7 tha⁻¹) were tested in competitive variety testing trials. The investigated lines are a valuable material from the viewpoint of genetics, breeding and agronomic characteristics which will be used in subsequent breeding work, and the most promising lines will be transferred to an official variety testing.

Key words: narrow-leaved forage lupine, individual selection, variety, resistance to fungal diseases

INTRODUCTION

One of the key challenges that Lithuania has currently to deal with is the rational use of land, since any agricultural activity either increases or declines natural soil fertility. Consequently, it is very important to select a farming method that would prevent any direct or indirect damage to the environment.

Recent research has shown that the methods increasing soil biological fertility are most promising. Of all known microorganisms, most efficient nitrogen fixators are legume bacteria in symbiosis with legume plants [1, 2]. Depending on these bacteria and legume crop properties and growing conditions, the symbiotically fixed nitrogen can total from 45 to 460 kg/ha per year [3], therefore in many countries researchers suggest a rational use of mineral nitrogen and a maximal use of biological nitrogen. With a growing demand for ecologically pure agricultural produce, increasingly more attention is being paid to ecological agriculture, which restricts the use of fertilisers, pesticides and herbicides. Therefore in the situation of increased environmental pollution legume crops are of a special value in ecological agriculture.

One of the oldest legume crops, grown worldwide, suitable not only for forage production as a protein source but also for soil culturing, increasing its natural fertility is lupine [4]. Only two lupine species, yellow lupine (Lupinus luteus L.) and narrow-leaved lupine (Lupinus angustifolius L.), are currently grown as cultivated crops in Lithuania.

Lupine has been grown in Lithuania since olden times, however, the area under this crop is not large. The main reason why the area sown with lupine is declining is the spread of new fungal diseases. Lupine anthracnose (Colletotrichum gloeosporioides (Penz.) Penz & Sass.) [5, 6] is one of the most harmful lupine fungal diseases, which occurs on all lupine species. Anthracnose affects seed, stems, pedicels, inflorescences and pods at different stages of lupine ontogenesis (except for seedling and complete maturity stages). Well visible lesions with clusters of pink spores form on different parts of a plant. At affected spots the stems and inflorescences break, pods shrivel and fall off. The least resistant to this disease is yellow lupine. Owing to this fact, yellow lupine is not cultivated in Lithuania at the present time.

Narrow-leaved forage lupine and narrow-leaved lupine for green manure are noted for a high resistance to anthracnose. This lupine species is characterised by a rapid growth rate during all stages of development, higher resistance to fungal diseases, and a shorter growing season. The key directions for lupine breeding are productivity increase, seed quality improvement, disease and pest resistance enhancement.
The objective of our work was to develop high-yielding, fungal disease-resistant, early maturing varieties of narrow-leaved forage lupine suitable for cultivation in all climatic zones of Lithuania.

**MATERIALS AND METHODS**

Tests of the new narrow-leaved forage lupine breeding lines for anthracnose resistance were carried out during the final stage of the breeding process – at competitive variety testing trials during 2001–2003. The trials were set up at the LIA Vokë Branch in a six-course crop rotation, with preceding crop spring cereals. The soil of the experimental site was sod podzolic slightly podzolized on calcareous gravel, with medium acidity, low in humus (2.0–2.1), nitrogen 0.096–0.117%, phosphorus 113.2–147.3 mg kg⁻¹, potassium 126.4–153.3 mg kg⁻¹.

The soil for lupine was prepared using a conventional technology: deep autumn ploughing, two cultivations in spring. Weeds were controlled by the herbicide Gezagard (2–2.5 kg/ha). The fungicide Kemitar-T (2 l/t seed) was used for seed treatment.

Seven narrow-leaved forage lupine breeding lines were investigated. The yellow forage lupine variety ‘Trakiai’ was used as a control. Since during the last eight years anthracnose had occurred annually, the tests were conducted under natural conditions.

The area of the record plots was 6 m², four replications were done. A randomised plot design was employed.

During the growing season, resistance to fungal diseases was estimated at three plant growth stages: seedling, bud formation – flowering, and shiny pods. The 1-to-9 point scale was used: 1 – very low resistance, diseased plants over 50%, 5 – moderate resistance, diseased plants less than 2.5% [8, 9]. With this end in view, at complete emergence plants were counted in A and C replications, at seedling, bud formation – flowering, and shiny pods stages anthracnose affected plants were counted and removed from the plot. At the complete maturity stage healthy plants were counted and their productivity was estimated. The percentage of fungal-disease-affected plants was identified according to the formula:

\[ P = \frac{n}{N} \times 100, \]

where \( n \) is the number of affected plants and \( N \) is the number of assessed plants [10].

Data on green material and seed yield were processed by statistical methods using the Selekcija software package [11].

To define meteorological conditions, the hydrothermal coefficients HTC were calculated according to the formulation:

\[ HTC = \frac{\Sigma p}{0.1 \Sigma T^0}, \]

where \( \Sigma p \) is the sum of precipitation over a period when the temperature was higher than 10 °C, \( \Sigma T^0 \) if the sum of active temperatures over the same period [12].

In our tests we estimated anthracnose (Colletotrichum gloeosporioides (Penz.) Penz & Sass.) resistance of the new narrow-leaved forage and yellow forage lupine variety ‘Trakiai’ and the effect of anthracnose resistance on seed yield.

**RESULTS AND DISCUSSION**

During the period 2001–2003, two species of lupine were investigated in field trials: 7 early maturing breeding lines of narrow-leaved forage lupine developed by an individual selection method and characterised by a high resistance to anthracnose and a high yield, and a high yielding, medium early yellow forage lupine variety ‘Trakiai’. Currently there are no varieties of narrow-leaved forage lupine included in the official variety list, therefore the yellow forage lupine variety ‘Trakiai’ was chosen as a standard.

The weather conditions during the experimental period varied. As a result, the spread of lupine anthracnose was different, too. During the period 2001–2003, no anthracnose was spotted in the treatments of yellow forage lupine and narrow-leaved forage lupine at the seedling stage due to the early lupine sowing and low air temperatures (8.0–15.3 °C) during the emergence – stem growth stage. Under natural conditions, the first symptoms of disease can manifest themselves during the stem growth period, but most often they are identified at later growth stages. Experimental evidence suggests that the disease severity, incubation period and the number of pathogen’s generations are different under natural conditions and depend on the lupine species, genotype, variety, and earliness. Furthermore, infection development is markedly affected by weather conditions and the plant growth stage at which the disease appears. The greatest harm is done when anthracnose starts to spread at bud formation – flowering stage or shiny pods stage. When lupine has reached the end of milk maturity – beginning of wax maturity, the disease does not cause any serious damage to the seed yield, but the seed gets infected.

In 2001, anthracnose spread in the trial plots of the yellow forage lupine variety ‘Trakiai’ in the middle of the second ten-day period of July at the shiny pods stage. The weather conditions in July were especially conducive to the development of the pathogen: HTC - 1.9, amount of precipitation 94 mm, air temperature 21.0 °C (long-term average 87 mm, 17.4 °C, respectively). The recorded disease incidence in yellow forage lupine reached 63% (Fig. 1), lupine matured at the beginning of the second ten-day period of August and the seed yield was as low as...
During the epiphytoty the narrow-leaved forage lupine breeding lines reached the end of milk ripeness – beginning of wax ripeness, depending on the genotype. The incidence of anthracnose was slightly higher in the later maturing breeding line N 1670, the affected plants amounted to 5.6%, however, this did not have any significant effect on seed yield and quality. Narrow-leaved lupine matured at the end of the third ten-day period of July, the seed yield was from 1.7 to 1.8 t/ha.

The highest anthracnose severity was recorded in 2002. The strong epiphytoty at the end of the third ten-day period of June was determined by the weather conditions conducive to the spread of the pathogen, HTC – 2.02 (the highest temperature of the month 31 °C, total monthly precipitation 69 mm), sufficient amount of infection and the yellow forage lupine variety ‘Trakai’ growth stage – the end of flowering. The disease severity was as high as 95% (Fig. 1). The seed yield was 0.1 t/ha (Fig. 2).

During a strong disease outbreak at the end of June forage lupine reached milk maturity. While investigating the new breeding lines, very high anthracnose resistance was identified for individual genotypes. Anthracnose resistance of the breeding lines 1700, N1701, N1702 was estimated by 9 ponts, affected plants from 0.8 to 1.0%. Seed yield was 1.6–1.85 t/ha. The following lines were found to be less resistant: N1683, N1684, N1703 – affected plants 2.5–2.7%, seed yield 1.25–1.7 t/ha. The lowest resistance was identified for N1670, affected plants accounted for up to 4.6%.

The lowest anthracnose incidence was recorded in 2003. The weather conditions were not favourable for the spread of the anthracnose pathogen, the HTC of the first-second ten day period of June was 0.6. The first symptoms of anthracnose were recorded at the end of the second ten-day period of July in the yellow forage lupine variety ‘Trakai’ at the end of milk maturity. The disease incidence was 12.3% (Fig. 1), seed yield 1.1 t/ha (Fig. 2). Since 1996, the start of lupine anthracnose occurrence in Lithuania, this was the highest yellow forage lupine seed yield obtained in the trials during a seven-year period. Anthracnose did not have any appreciable effect on narrow-leaved forage lupine.

Most heavily anthracnose-affected were found to be the breeding lines N1670 and N1703 – 1.3%. The rest of the breeding lines were affected from 0.5 to 1.0%. Narrow-leaved forage lupine matured within the first ten-day period of August. In 2003 a very high seed yield (3.1 to 3.7 t/ha) was recorded for narrow-leaved forage lupine.

Breeding for disease resistance is one of the most challenging tasks in breeding work. The rapid mutation of pathogen’s populations, its ability to con-
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Continuously form a large amount of races differing in virulence and aggressiveness are the chief obstacles in the development of resistant lupine varieties. When a limited gene fund is used in breeding work, lupine anthracnose can occur in varieties differing in their genetic potential. The presented data show that the development of the infection is influenced by human factors and the pathogen’s characteristics, as well as weather conditions. The world lupine gene fund does not contain any varieties completely resistant to fungal or viral diseases. It is likely that the varieties characterised by partial resistance do not lose this trait for a longer period, and in the years of weak epiphytosis such varieties either do not catch infection or are insignificantly affected, like N1700, N1701, N1702.

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\textbf{References}


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\textbf{ATSARPIO ANTRAKNOZEI (COLLETOTRICHUM GLOESPOROIDES (PENZ.) PENZ & SASS.) SIAURALAPIØ PAŠARINIO LUBINO SELEKCIINIO LINIJO KŪRIMAS}

\textbf{Santrauka}


\textbf{Raktapodþiai:} siauralapiø pašariniai lubinos, individuali atranka, veisli, atsparumas grybinëms ligoms