Investigation of some wild annual vetch (*Vicia* L.)

Nijolė Maršalkienė*

Aleksandras Stulginskis University, Studentų St., 11, Akademija, Kaunas Distr., Lithuania Investigation of annual vetch (*Vicia villosa* Roth., *V. angustifolia* L., *V. hirsuta* Gray) cenopopulations has been carried out since 1998. Gene fund is being collected, morphobiochemical characteristics and adaptive potential are being studied. Plant mass, stem, leaf and inflorescence ratio in the mass and crude protein content have been measured. Plant growth phonological stages are recorded. Diseases and damage done have been described.

Experiments have shown that by preserving the characteristics specific to the cenopopulations the tested species (*Vicia villosa* Roth, *V. angustifolia* L., *V. hirsuta* Gray) readily adapt to different year diverse ecological conditions. *V. villosa* was distinguished for the largest biodiversity. Averaged data suggest that according to the crude protein content in the plant aboveground mass, the stem, leaf and inflorescence ratio in the plant mass of *V. villosa* and some samples of *V. angustifolia* were more valuable than in that of cultivated *V. sativa*. For detailed tests and application in plant breeding cenopopulations were selected according to earliness of flowering, productivity, crude protein content, and disease resistance.

Key words: Vicia villosa, V. angustifolia, V. hirsuta, biometric parameters, crude protein, diseases

INTRODUCTION

World plant biodiversity, its conservation and prospects for practical use are becoming an increasingly pressing problem in the 21st century. The relevance is primarily determined by the rapid extinction of plant species and their populations (Crane, 2001; Raven, 2000). It is forecasted that by the end of the 21st century about 2/3 of all organisms will have become extinct. Over the last millennium nearly 10% of the total number of plant species have become extinct (Raven, 2000). Such intensive extinction of species can cause an ecological and biological disaster and irreversible disappearance of genetic resources (gene fund).

On the other hand, the constantly growing number of population necessitates increasing of food and forage resources. Besides forest felling, resulting in cultivated land area increasing, the use of fertiliser and chemical plant protection agents as well as genetically modified plant varieties is steadily increasing (Shevchenko et al., 2001). These human activities also exert a great effect on extinction of plant species and decline their biodiversity (Sokolov, Marchenko, 2002).

^{*}Corresponding author. E-mail: nijole.marsalkiene@asu.lt

As an alternative to the chemisation of agriculture and the spread of genetically modified varieties, organic agriculture has been rapidly developing worldwide since 1990. The purpose of organic agriculture is to produce ecologically clean produce (Lotter, 2003; Buzuk et al., 2002), minimise environmental pollution, conservation phytobiodiversity and to use it for human needs. National resources collection, storage, reproduction and use for rational needs in plant breeding are one of the tasks of genetic resources. Knowledge of multiplication, ecological peculiarities and response to changing environmental and anthropogenic factors makes conservation of plant species possible. A promising way to conserve phytobiodiversity is introduction, creation of collections, versatile tests of plant characteristics, selection of promising forms for breeding. There are about 50 000 well-eaten spontaneous plant species (Agaev, 1999). Only a small part of these species have been domesticated. Extinction of species and populations promotes search for and selection of valuable spontaneous species. The species under investigation are assessed not only as forage (Fraser et al., 2004; Sheaffer, Sequin, 2003) but also according to their ability to adapt to changing climatic conditions, genetic stability of major quality parameters, disease and pest resistance (Skvorcov, Kuklina, 2002). The chief object of domestication is not the species as a whole, but its individual ecotypes. The priority work is investigation of species biodiversity and selection of promising ecotypes.

The genus vetch (*Vicia*) belongs to the Fabaceae family. About 200 species are known, of which about 15 grow in Lithuania. The annual vetch *V. sativa*, *V. angustifolia*, *V. villosa*, *V. hirsuta* are typical plants of agrophytocenoses. *V. hirsuta* and *V. angustifolia* are attributed to the weed group. *V. sativa* and *V. villosa* are domesticated. *V. villosa* is seminatural, grows in phytocenoses as a weed.

Investigations of vetch species plant composition and parameters as well as biochemical makeup are being done (Lahuta et al., 2004; Trusov et al., 2004; Rogozhkina, Monakhova, 1999; Ficev et al., 2003), medicinal properties are studied (Babaskina, 1998), cariological tests and agrotechnical experiments are conducted (Stupakov et al., 1999; Khramoy, 1999), plant breeding is performed (Tyurin, Gulakova, 1989), vetch root system and interaction of *Rhizobium* bacteria are investigated (Pandey et al., 2004).

The objective of the present study was to investigate and estimate adaptive capacity of *V. angustifolia* L., *V. villosa* Roth., *V. hirsuta* Grey *ex situ*, under the same agrotechnical and meteorological conditions, seeking to conserve the gene fund and select promising forms suitable for practical use.

MATERIALS AND METHODS

The experimental subject was *V. angustifolia, V. hirsuta, V. villosa* species of the genus *Vicia* grown *in situ* (1998–2000) and *ex situ* (2001–2004). *V. sativa* was grown for the sake of comparison. *Ex situ* spring vetch (*V. angustifolia, V. hirsuta, V. sativa*) was sown in the second half of April, winter vetch (*V. villosa*) was sown in the first half of September.

Morphological plant assessment was based on the measurements of stem height, branching point, number of branches, number of pods, seed number per pod, 1 000 seed weight, fresh weight per plant. Crude protein content was determined in air-dried mass (Kjeldahl method), content of carotenoides in green material (colorimetry method), and disease resistance was measured (Šurkus, Gaurilčikaitė, 2002). To estimate the weather data the hydrothermal coefficient (HTC) was calculated (Kudakas, Urbonas, 1983) for the three stages: April–June, July–August, September–October.

Analysis of the material collected *in situ* and *ex situ* was done at the Laboratory of Crop Production Department of Aleksandras Stulginskis University (ASU). Protein and fat analyses were done at the Agrochemical Research Centre of LIA, the content of carotenoids was measured by a Beckman DU-40 spectrometer at the ASU "Tempus" Laboratory. The collections

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were grown and field trials were conducted at the ASU Experimental Station.

Meteorological conditions

In 2002 and 2003 the summer–autumn weather conditions were similar to the long-term mean (Fig. 1), however, the year 2002 was distinguished by hot and droughty weather during July–August and especially rainy autumn (Fig. 2), the year 2004 was also unrepresentative – the beginning of summer was cool and rainy, high temperatures occurred only in the second half of summer. During the experimental years the air temperature of the July–August period was by up to 3.4 °C higher than the long-term mean, the air temperature of the April–June period, except for the year 2004, was by 0.1– 3.4 °C higher than the long-term mean, during 2001–2004 the air temperature of the September–October period varied less (Fig. 2).

In all experimental years the amount of precipitation that fell during April–June was lower than the long-term mean (22.5–122 mm), the amount of precipitation during July–August (except for 2002) and September–October was by 18–46 and 11–106 mm higher. In 2002 especially droughty was the April–June period when the amount of precipitation was by 103 mm lower than the long-term mean, and the July– August period when the amount of precipitation was by 83 mm lower than the long-term mean.

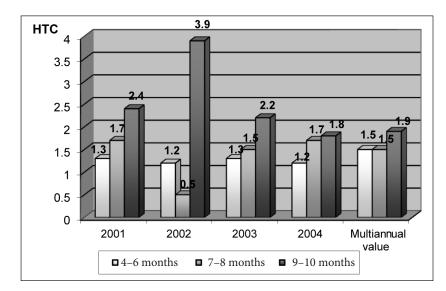


Fig. 1. Hydrothermal coefficient (HTC) of the vegetation period 2001–2004

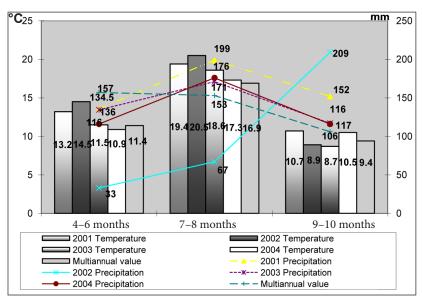


Fig. 2. Meteorological conditions of the vegetation period 2001–2004

RESULTS AND DISCUSSION

Ecogeographical distribution

During expeditions we collected 23 accessions of *V. angustifolia*, 21 accessions of *V. hirsuta*, and 67 accessions of *V. villosa*. The species under investigation most often grew (*V. villosa*winter, *V. angustifolia*, *V. hirsuta*-spring) in cereal agrophytocenoses.

V. angustifolia – narrow-leafed, *V. hirsuta* – hairy vetch often grew together in spring cereal phytocenoses, less often in winter cereal phytocenoses, waste or abandoned land, less frequently in grasslands, sandy loam and light loam soils (pH 5.8–7.0). These species are rather frequent on the whole territory of Lithuania, especially in South and South-east

regions characterised by light soils, and less frequent in northern Lithuania (Fig. 3).

V. villosa is a winter vetch. The plant is thought to have originated on the Mediterranean littoral. It belongs to the European flora element (Lietuvos TSR flora, 1971). *V. villosa* was specific to rye agrophytocenoses, it occurred less frequently on waste or abandoned land and seldom in spring cereals, potatoes. Winter vetch grows on different acidity (pH 5.1–6.9) sandy loams, in some sites even on sandy soils. *V. villosa* is most widespread in South-east and East Lithuania, far less frequent in South-west and Central Lithuania, it was not found at all in Northern Lithuania. Only one accession of this plant was found in the north-western part of the country (Fig. 4).

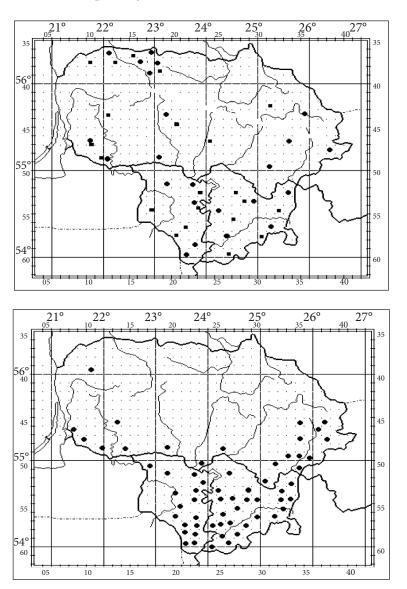


Fig. 3. Finding places of *V. angustifolia* (●) and *V. hirsuta* (■)

Fig. 4. Finding places of *V. villosa* (■)

Investigation of some wild annual vetch (Vicia L.)

Ex situ collection

V. angustifolia. This vetch is thought to be the pioneer of the genus (Sinskayja, 1969). It is characterised by a great phonological and morphological diversity. According to the length of the growing season the distinguished groups differed not only by the beginning of flowering but also by other parameters (Table). Late forms were characterised by taller plants, greater mass, higher 1 000 seed weight and crude protein content, and clearly expressed leaf polymorphism (Fig. 5). Leaves accounted for 52% of the total plant mass of V. angustifolia (Fig. 4). Leaves had 23.7% of protein. Although flowers constituted only a small portion of the total plant mass, the crude protein content present in them had an appreciable effect on the total crude protein content in the aboveground part of a plant. The content of carotenoides in green material is 0.5 mg/g.

V. sativa (sown vetch). This species is found on the whole territory of Lithuania (Gudžinskas, 1999). It was formed through a range of transitional forms from *V. angustifolia* (Sinskayja, 1969). It is the most widespread species of cultivated (domesticated) vetch. Originally it was a food crop known in Europe since the Neolithic times, later it became a forage and green manure crop (Sinskaya, 1969).

In our experiments the V. sativa variety was grown as a control. The seed ripening period of this variety coincided with that of V. villosa early forms (Table). The stem height was equal to that of medium early V. hirsuta, and plant mass was lower than that of V. villosa, however, it was several times higher than that of V. angustifolia and V. hirsute. According to pod productivity this vetch lagged behind V. villosa and V. hirsuta and was distinguished by the seed number per pod. According to this parameter V. sativa was identical to V. angustifolia and surpassed all species tested by 1 000 seed weight. V. sativa and V. angustifolia had a similar number of seed set per pod. Since the number of seed is genetically determined and is only weakly affected by the environmental conditions, it is a relatively stable parameter exhibiting the relatedness of these two species. According to the ratio of stems, leaves and inflorescences in the total plant mass these two species are also very close (Fig. 4). In terms of crude protein percentage in the total plant mass (Fig. 6) V. sativa equals V. hirsuta. Their protein content is lower



Fig. 5. Different forms of *V. angustifolia* in 2002. *a* – late, *b* – average early, *c* – ultra early

lable. Earli	lable. Earliness groups of Vicia angustifolia, V. hirsuta,	angustifolia, V. hi		V. villosa, LUA Experimental Station, 2001–2004	tal Station, 200	11-2004			
Farlinace	Length of	% of samples	Ctom	Graan mass of	Branch	Branch number	Number of	Saade eat in	Weight of
species	growing period, days	from the collection	height, cm	one plant, g	From enlobe	From main stem	pods per plant	a pod	1 000 seeds, g
	_			V. angustifolia	tifolia				
Ultra early	75-80	6	35.1 ± 5.4	2.3 ± 0.7	1.9 ± 0.2	2.0 ± 0.2	7.1 ± 0.9	7.6 ± 1.1	7.5 ± 0.9
Early	81-85	22	42.6 ± 7.2	3.2 ± 1.1	2.3 ± 0.4	2.5 ± 0.3	9.4 ± 1.2	8.0 ± 1.1	14.5 ± 1.8
Average early	86–95	60	48.7 ± 9.6	3.6 ± 1.6	1.8 ± 1.0	4.2 ± 0.6	13.2 ± 1.9	7.5 ± 1.6	19.2 ± 2.1
Late	<96	6	54.3 ± 6.4	4.0 ± 0.8	0.9 ± 0.4	4.8 ± 0.3	11.7 ± 1.2	5.8 ± 0.9	22.3 ± 1.4
Average			4.5 ± 12.1	3.3 ± 1.8	1.9 ± 1.3	2.8 ± 1.5	11.6 ± 2.2	7.4 ± 2.1	17.1 ± 3.2
				V. hirsuta	uta				
Early	65-75	20	70.2 ± 5.8	1.1 ± 0.2	I	0.9 ± 0.2	54.1 ± 8.2	2.0 ± 0.00	6.3 ± 1.1
Average early	76–90	57	86.4 ± 7.9	1.9 ± 0.3	I	1.2 ± 0.4	56.5 ± 11.9	2.0 ± 0.01	7.1 ± 1.4
Late	<91	24	91.2 ± 5.3	2.1 ± 0.2	I	1.5 ± 0.3	57.0 ± 7.4	1.9 ± 0.01	7.4 ± 1.2
Average			84.4 ± 14.4	1.7 ± 0.5	I	1.3 ± 0.8	56.8 ± 138	2.0 ± 0.01	7.3 ± 1.6
				V. villosa	sa				
Ultra early	100-105	7	78.4 ± 13.4	21.5 ± 5.6	2.6 ± 0.4	1.2 ± 0.3	$14.6 \pm 4,1$	4.4 ± 0.3	23.3 ± 2.3
Early	106-110	19	101.5 ± 12.1	28.8 ± 4.2	1.8 ± 0.3	1.9 ± 0.3	23.5 ± 8.6	4.7 ± 0.5	28.9 ± 3.1
Average early	111-120	44	121.8 ± 18.2	32.2 ± 7.6	1.0 ± 0.1	3.1 ± 0.9	29.0 ± 11.8	4.5 ± 0.8	30.2 ± 3.4
Late	121-130	16	135.2 ± 16.5	34.4 ± 5.4	1.0 ± 0.1	3.5 ± 0.5	33.8 ± 4.9	4.5 ± 0.5	31.4 ± 2.8
Ultra late	<131	14	154.2 ± 17.2	33.2 ± 6.2	1.0 ± 0.0	3.8 ± 0.4	36.2 ± 6.3	4.3 ± 0.3	29.2 ± 2.0
Average			124.4 ± 28.2	32.6 ± 9.9	1.3 ± 0.7	3.2 ± 1.6	30.5 ± 16.3	4.5 ± 0.9	30.4 ± 4.1
				V. sativa	иа				
'Tverai'	105		84.0 ± 5.9	17.5 ± 2.4	1.0 ± 0.6	3.9 ± 1.2	16.5 ± 3.2	5.2 ± 0.5	54.0 ± 1.2

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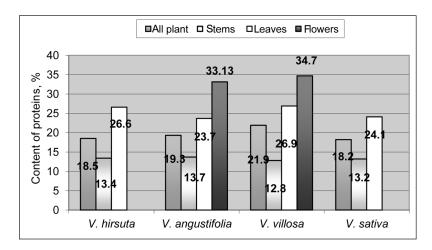


Fig. 6. Protein content of dry vetch mass

than that of *V. angustifolia and V. villosa*. The protein content in leaves which are the most valuable part of the total plant mass is identical to that of *V. angustifolia*.

Vicia hirsuta. During the experimental years the first plants of early forms started flowering at the end of June (Table), and plants of late forms one week later. The parameters of this species cenopopulations were characterised by a relatively small diversity. Populations whose height varied from 70.2 to 91.2 cm were dominant. Leaves accounted for 45.6% of the plant air-dried mass (Fig. 7). Leaves contained 26.6% of crude protein, i. e. by protein content in leaves it was similar to sand vetch and surpassed other species (Fig. 6). The deviation of protein content from the average was relatively small. V. hirsuta was noted for a relatively weak response to diverse ecological conditions of different years. V. hirsuta had the biggest content of carotenoides in green material – 0.59 mg/g. Due to these characteristics and high crude protein content, *V. hirsuta* is a valuable crop for plant breeding and genetic tests (Tyiurin et al., 1989).

Vicia villosa. Winter vetch is an archeophyte that arrived in Lithuania together with winter cereals (Gudžinskas, 1999). All the cenopopulations tested were very varied both in terms of phonological and morphometrical parameters. According to all parameters the plants of very early cenopopulations lagged behind the other groups most considerably (Table). Of all the parameters tested the number of seeds per pod was found to be the least variable. The leaf to stem ratio (Fig. 7) in the total plant mass was similar. The total value of plant mass was increased by inflorescences that contained the largest amount of crude protein (Fig. 6). The content of carotenoides in green material was the smallest one -0.44 mg/g.

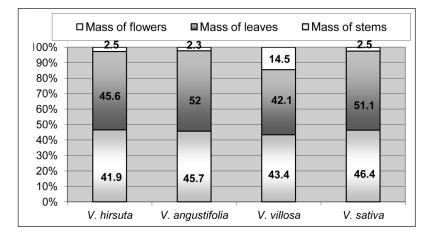


Fig. 7. Proportion of stems, leaves and flowers in vetch

In different years winter vetch cenopopulations varied according to all biometrical parameters. This suggests that this species is polymorphic and can adapt well to changing ecological conditions (Table) without the loss of parameters specific to the cenopopulations.

Our experimental findings indicate that variable weather conditions affected phonological, morphological, productivity, and biochemical parameters of the cenopopulations of all species tested. The relationship between the tested parameters and the place of geographical origin was not found during the experimental period. Warm, sunny weather had a positive effect on herbage protein content. In 2002 the total protein content of species was 17% lower compared with the average of the rest of the years (2001, 2003, 2000).

V. villosa characterised by the longest growing season was most heavily affected by diseases and pests. The end of the growing season was influenced by the spread of diseases. In 2001 and 2003 in the second half of the growing season when the weather was rather hot and wet (Fig. 2), powdery mildew caused by Erysiphe communis Grev. f. vicea Jacz. was identified. About 50% of cenopopulations were found to be affected. In 2004 the growing season for all plants finished at the beginning of August. The wet and cool weather in 2004 promoted the spread of Ascochyta sp. fungus which caused foliar, stem and pod ascochyta. Disease resistance of different cenopopulations was diverse. Nine cenopopulations resistant to powdery mildew and seven populations resistant to ascochyta were discriminated. During 2001-2004 assessments for Bruchus atomarius L. damage on leaves, in flowers and pods were made. Late V. villosa forms were most heavily affected by this pest. Especially heavy pest damage was identified in 2002 when the weather during the July-August period was exceptionally dry and hot (Fig. 1).

All the species tested had populations with valuable characteristics (total plant mass, stem to leaf ratio in the plant mass, length of the growing season, disease resistance) that can be used in breeding, molecular research, and practical needs. A special attention should be drawn to *V. villosa*, as a protein-rich plant producing abundant green material. *V. villosa* provides a good soil cover and is used as a weed control means (Fujii, Araki, 2000; Hanano et al., 1998; Sadeghi, Isensee, 2001), and as a soil amendment (Zhou, Everts, 2004).

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VIKIO (*Vicia* L.) GENTIES VIENMEČIŲ RŪŠIŲ TYRIMAI

Santrauka

Vienmečių vikių (*Vicia villosa* Roth., *V. angustifolia* L., *V. hirsuta* Gray) cenopopuliacijų tyrimai vykdomi nuo 1998 m. Renkamas genofondas, tiriamos morfobiocheminės savybės, adaptacinis potencialas. Nustatyta augalų masė, stiebų, lapų ir žiedynų santykis, žaliųjų proteinų kiekis. Buvo registruojami augalų augimo fenologiniai tarpsniai, apibūdintos plintančios ligos ir jų daroma žala.

Didžiausia bioįvairove išsiskyrė *V. villosa*. Vidutiniais duomenimis, pagal žaliųjų proteinų kiekį augalo antžeminėje masėje visos tirtos rūšys buvo vertingesnės už kultūrinį *V. sativa*. Atskiros cenopopuliacijos lenkė *V. sativa* ir antžeminės masės dydžiu.

Raktažodžiai: *Vicia villosa*, *V. angustifolia*, *V. hirsuta*, biometriniai parametrai, žalieji proteinai, ligos