

COMPARATIVE STUDIES OF ANNUAL LEGUMES

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Abstract: The aim of present study was to get comparative data on forage productivity and quality of forage of Czech cultivars of legumes in the conditions of Northern Bulgaria and respectively to select species and genotypes with the potential for successful introduction in the structure of forage production in Bulgaria. Five species of forage crops were observed and the respective cultivars: Egyptian clover (*Trifolium alexandrinum* L.), cv. Faraon; crimson clover (*Trifolium incarnatum* L.), cv. Kardinal; annual bird's-foot-trefoil (*Lotus ornatopoides* L.) cv. Junak; black medick (*Medicago lupulina* L.) cv. Ekola and white melilot (*Melilotus albus* L.), cv. Adela. The studied legumes differed significantly in their fodder productivity. They are ranked in the following order of DM yield: white melilot - black medick - annual bird's-foot-trefoil - Egyptian clover - crimson clover. The productivity and participation of Egyptian clover and crimson clover in grasslands varied significantly in years. The biomass of crimson clover had the highest content of crude protein (15.24%) and the lowest of crude fiber (21.69%) and no digestible components. According to the comprehensive evaluation of data on productivity and forage quality of studied annual legumes, black medick could be defined as the species with the highest potential for cultivation in the conditions of the Central Northern Bulgaria. It is characterised by high productivity of green mass and dry matter, it has regrowing ability, it is distinguished by a high content of crude protein (14.92%) and crude fat (4.66%), optimal content of neutral and acid detergent fibers (34.67 and 24.99%) and with high levels of hemicellulose content (9.68%). Energy value of forage of that species, assessed by means of feed unit of milk (FUM) and growth (FUG) was assessed as very high (FUM – 0.69/kg DM and FUG – 0.63/kg DM).

Key words: annual legumes, yield, fodder quality

Introduction

The importance of annual legume as forage crops is significantly less than perennial ones. However, in many cases because of specific agro-ecological conditions or requirement of the sustainable agriculture, forage production for ruminants is successfully based on annual species (*Simić and Vučković, 2014*). In Bulgaria, only peas and vetch are used as a annual legumes (*Kirilov, 2016*). The introduction of new practices for increasing the seasonal productivity and forage quality of temporary swards by annual legume requires studies on plant material of different origin. Most of the studies, which have already been conducted, include species, which have a greater significance as fodders for regions with Mediterranean climate (*Goranova et al., 2007; Vasileva et al., 2011; Vasileva and Vasilev, 2012, Naydenova et al., 2014*). It was found that only some of these legumes show good adaptability to the conditions of Northern Bulgaria (*Vasilev, 2006, 2009; Mitev et al., 2013*).

The aim of present study was to get comparative data on productivity and quality of Czech cultivars of annual legumes in the conditions of Northern Bulgaria and to select species and genotypes with the potential for successful introduction in the production structure of forage crops in Bulgaria.

Material and methods

The experiment was carried out at the Experimental Station on Soybean – Pavlikeni, during the period 2013-2014. The region is characterized by temperate continental climate and altitude of 114 m. Soil type is leached chernozem, with an average reserve of phosphorus and nitrogen and a good reserve of potassium. According to rainfall density for the period of April/October, the year of legumes establishment was relatively drier, with a rainfall amount closer to the normal for the region (423 mm, compared to 367 mm, average for 50 year period) - Fig.1. There was more humidity and more favourable distribution of rainfalls in 2014, with a rainfall amount in the vegetation period of 579 mm.

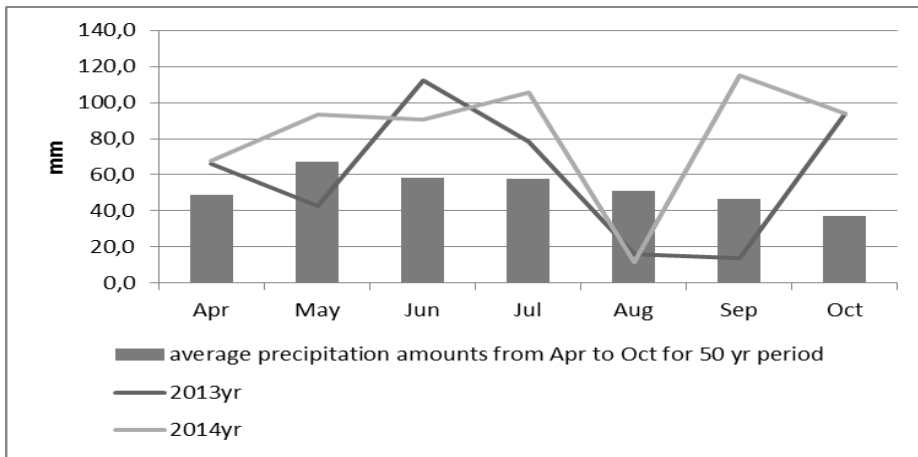


Figure 1. Rainfall amount (mm) for the experimental period

Five species were included in the experiment: Egyptian clover (*Trifolium alexandrinum* L.), cv. Faraon; crimson clover (*Trifolium incarnatum* L.), cv. Kardinal; annual bird's-foot-trefoil (*Lotus ornatopoides* L.) cv. Junak; black medick (*Medicago lupulina* L.) cv. Ekola and white melilot (*Melilotus albus* L.), cv. Adela. The species were studied in pure crop. The randomized block method was used, the number of replications was 4 and the area of the plot was 5m². Mineral fertilization was not applied. Sowing was in rows, with 15 cm space between the rows and sowing rate was 1000 g.s.m⁻². Sowing dates were 12. April 2013 and 14. March 2014. Mowing was conducted at the stage of budding to early flowering of species at a height of 3-4 cm. The following characteristics were observed: plant growth and development, sward botanical composition, yield of green mass and dry matter (t ha⁻¹), regrowing ability, and forage quality. The following indicators for the forage quality have been studied: crude protein (CP,%) according to Kjeldahl; crude fiber (CF,%) according to Weende analysis, crude fat (Cft,%) by extraction in an extractor of Soxhlet type, Ash (%) - ashing in a muffle furnace at 550 °C, humidity content (%) - the sample was dried under a temperature of 105 °C till it reached a constant weight, calcium (Ca,%) - according to (AOAC, 2007), phosphorus (P,%) according to (AOAC, 2007) spectrophotometer (Agilent 8453 UV – visible Spectroscopy System), measuring within the sphere 425 nm, NEF=100 (CP,%+ CFr,%+ CF,%+ Ash,%+ Humidity,%). The fiber composition of cell walls was determined as a percentage of dry matter, which included: neutral detergent fibers (NDF,%), acid detergent

fiber (ADF,%) and acid detergent lignin (ADL,%) according to the *Goering and Van Soest* (1970) analysis, polysaccharides hemicellulose (NDF-ADF) and cellulose (ADF-ADL). The degree of lignification was expressed as a percentage share of ADL/NDF. The potential energy nutritional value of fodder was assessed according to the Bulgarian system as FUM and FUG (*Todorov*, 1995). Equations were used for legumes according to experimental values of CP, CF and NFE and were precalculated according to the coefficients of digestibility. The coefficient for the energy exchange (q) expresses the share of the exchangeable energy (EE) of the net energy (NE), calculated as a part of unit with the equation: $q = EE/NE$.

For the statistical processing of yield data was used two-factor analysis of variance and multiple comparison of yields in years by means of least significant differences (LSD). The statistic program STATGRAPHICS PLUS was used.

Results and discussion

White melilot exceeded all species included in the study in green and dry matter yield, with the average values for the experimental period of 29.6 and 7.3 t ha⁻¹ (Table 1) respectively. This was also the earliest maturing species, and its productivity varied slightly in years, which determined its good adaptability to the region of testing. It achieved harvest maturity stage in the first half of June, as it regrew quickly and formed a second regrowth up to the middle of July. It participated with 89 and 97% in the biomass composition in the first regrowth, respectively in the first and second experimental year. It is important to note that the economic value of high yield of white melilot could be low because of the bitter taste and specific smell of forage caused by alkaloid coumarin (*Duke*, 1981; *Sanderson et al.*, 1986). In the contemporary selection of grasses is required to be cultivated low-coumarin varieties of that species, which are less productive (*Meyer*, 2005).

High average green mass yields - 21.3 t ha⁻¹, 20.7 t ha⁻¹ and dry matter - 5.8 t ha⁻¹, 5.4 t ha⁻¹ were gathered with black medick and annual bird's-foot-trefoil. In both experimental years, these species had great presence in fodder mass (>60-70%, Table 1), which is a prerequisite for high competitiveness and adaptability. The dry matter productivity of Czech cultivar of black medick was significantly higher than Bulgarian population of that species (3.3-3.6 t ha⁻¹) grown also in the conditions of the Central

Northern Bulgaria (Naydenova *et al.*, 2014) and harvested in the same phenophase.

In the first experimental year, the values of green and dry matter yield, as well as the participation of crimson clover in fodder biomass were significantly higher than in the second year, 21.5 t ha⁻¹, 4.05 t ha⁻¹, 73 % and 11.2 t ha⁻¹, 2.5 t ha⁻¹, 22 %, respectively. Crimson clover, in the spring growth, was equal in productivity with black medick and the annual bird's-foot-trefoil, but it did not regrow after cutting. In conditions of the spring sowing only a part of plants developed generative stems and could be considered that the studied genotype of that species had a winter type of development and its productivity should be studied under conditions of pre-winter sowing. Crimson clover was distinguished by the highest degree of weed infestation – over 52% average for the period.

The productivity and participation of Egyptian clover in biomass also varied significantly by years. In the first year that species had the smallest percentage share in the sward (49%), as well as the lowest productivity of green and dry mass (7.8 t ha⁻¹ and 1.5 t ha⁻¹). In the second experimental year, when also the rainfall amount was higher, the presence of Egyptian clover in sward significantly increased (78%) and respectively the obtained yields of green and dry matter yield were very high (26.4 t ha⁻¹ and 7.8 t ha⁻¹). Variability in values of the main indicators, which determine the economic significance of this crop, is a sign for its slighter adaptability to soil and climate conditions for the region of study.

Table 1. Fresh and dry matter yield and participation of annual legume in fodder biomass

Species	FMY (2013) t ha ⁻¹	FMY (2014) t ha ⁻¹	Mean t ha ⁻¹ yr ⁻¹	DMY (2013) t ha ⁻¹	DMY (2014) t ha ⁻¹	Mean t ha ⁻¹ yr ⁻¹	% in fodder mass (2013)	% in fodder mass (2014)
<i>T. alexandrinum</i>	7.8 ^b	26.4 ^a	17.1	1.5 ^c	7.8 ^a	4.7	49	78
<i>T. incarnatum</i>	22.2 ^b	11.3 ^b	16.8	4.2 ^b	2.5 ^b	3.4	73	22
<i>L. ornithopoides</i>	25.9 ^{ab}	15.4 ^b	20.7	6.2 ^a	4.5 ^b	5.4	77	75
<i>M. lupulina</i>	25.4 ^{ab}	17.2 ^b	21.3	7.4 ^a	4.1 ^b	5.8	70	58
<i>M. albus</i>	31.0 ^a	28.1 ^a	29.6	7.1 ^a	7.4 ^a	7.3	89	97
LSD _{0.05}	7.5	8.3		1.8	2.3			

*values of annual productivity by years, followed by equal letters do not differ significantly at P<0.05
FMY - fresh matter yield; DMY - dry matter yield

Significant differences were found among studied species for protein and fiber content (Table 2). The biomass of crimson clover had the highest

crude protein content (15.24% of dry matter) and the lowest level of crude fiber (21.68%) for a two year period. This species had also the lowest mean content of no digestible components in the forage (Table 3) - lignin (3.08%) and cellulose (17.73%). The decreased content of fractions of the structural fiber components of the cell walls which was observed in crimson clover (NDF – 31.16%, ADF – 20.81% and ADL – 3.08%) could be explained by the fact that forage mass of this species consisted predominantly by leaves.

Black medick was closer to crimson clover in content of crude protein (14.92% of DM) and hemicellulose (9.68%). The content and ratio of NDF (34.67%) and ADL (5.01%) in the forage of Black medick can be defined as optimal (*Oba and Allen, 1999*). This species had the highest content of crude fat - 4.66%. The annual bird's-foot-trefoil was characterized by the lowest content of crude protein (10.07%), neutral detergent fibers (30.55%) and hemicellulose (6.43%), as well as with the highest level of lignification (17.09%). Those values suggest low levels of intake and low digestibility of biomass of that species. In comparison to common bird's-foot-trefoil, grown in the foothill conditions of the Northern Bulgaria (*Churkova, 2012; 2013*), annual bird's-foot-trefoil has a significantly lower content of NDF and ADF and does not differ in relation to content of macroelements.

Significant variation depending on legume species was observed in relation to content of mineral substances, and the content of macroelements of calcium and phosphorus, as well as according to values of their ratio in the forage. The fodder of the annual bird's-foot-trefoil had the highest percentage content of mineral substances (15.63%) and calcium (2.64%). White melilot significantly gave in to the other species in relation to mineral concentration (5.28). The ratio Ca:P was close to the optimal 2:1 (NRC, 2000) only for Egyptian clover (3.79:1) and exceeded the determined one as critical 6:1 for the white melilot (7.41:1) and for bird's-foot trefoil (7.65:1).

Variation among studied species in relation to coefficient of energy exchange (q) could be specified as low - from 0.42 to 0.46 (Table 4). The exchangeable energy (7.34 MJ/kg DM) for crimson clover took the greatest share of the net energy (15.81 MJ/kg DM). The highest energy value of forage, measured as feed unit of milk (FUM) and growth (FUG) was found for crimson clover, black medick and white melilot, and the lowest - for Egyptian clover (Table 4).

Table 2. Chemical composition of annual legume- average for a period of two years (% DM)

Species	CP	CF	Cft	Ash	Ca	P	Ca:P [†]
<i>T. alexandrinum</i>	11.18	33.44	3.02	12.24	2.14	0.565	3.79
<i>T. incarnatum</i>	15.24	21.68	3.61	11.85	2.34	0.420	5.57
<i>L. ornithopoides</i>	10.07	25.26	4.61	15.63	2.64	0.345	7.65
<i>M. lupulina</i>	14.92	24.92	4.66	10.29	1.94	0.321	6.04
<i>M. albus</i>	11.76	32.59	3.16	5.28	1.84	0.248	7.41

CP – crude protein; CF – crude fibers; Cft – crude fats; Ash – mineral composition; Ca – calcium content; P – phosphorus content

[†]-relationship is not expressed as a percentage

Table 3. Content of fiber components of cell walls in annual legumes- average for a period of two years (% DM)

Species	NDF	ADF	ADL	Hemicellulose	Cellulose	Degree of lignification
<i>T. alexandrinum</i>	42.86	30.95	6.13	11.91	24.82	14.30
<i>T. incarnatum</i>	31.16	20.81	3.08	10.35	17.73	9.88
<i>L. ornithopoides</i>	30.55	24.12	5.22	6.43	18.90	17.09
<i>M. lupulina</i>	34.67	24.99	5.01	9.68	19.98	14.45
<i>M. albus</i>	33.60	24.15	3.05	9.45	21.10	9.08

NDF – neutral detergent fiber; ADF – acid detergent fiber; ADL – acid detergent lignin

Table 4. Energy nutritional value of annual legume- average for a period of two years

Species	NE	EE	q	FUM	FUG
<i>T. alexandrinum</i>	15.93	6.71	0.42	0.61	0.55
<i>T. incarnatum</i>	15.81	7.34	0.46	0.69	0.63
<i>L. ornithopoides</i>	15.20	6.79	0.45	0.63	0.57
<i>M. lupulina</i>	16.47	7.42	0.45	0.69	0.63
<i>M. albus</i>	17.11	7.48	0.44	0.69	0.63

NE – net energy (MJ/kg CB); EE – exchangeable energy (MJ/kg CB); FUM – feed unit of milk (kg DM); FUG – feed unit of growth (kg CB); q – coefficient of energy exchange

Conclusion

According to the comprehensive evaluation of data on productivity and forage quality of studied annual legumes, black medick cultivar 'Ecola' could be defined as the species and genotype with the highest potential for cultivation in the conditions of the Central Northern Bulgaria. It is

characterised by high productivity of green mass and dry matter, it has regrowing ability, it is distinguished by a high content of crude protein and crude fat, optimal content of neutral and acid detergent fibers and high levels of hemicellulose content. Energy value of forage of that species, assessed by means of feed unit of milk (FUM) and growth (FUG) was assessed as very high.

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Komparativno ispitivanje jednogodišnjih mahunarki

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Rezime

Cilj ovog istraživanja je bilo dobijanje komparativnih podataka o produktivnosti i kvalitetu krmnog bilja čeških sorti jednogodišnjih mahunarki u uslovima severne Bugarske i, respektivno, odabir vrste i genotipova sa potencijalom za uspešno uvođenje u strukturu proizvodnje krme u Bugarskoj. Pet vrsta krmnog bilja je bilo uključeno u istraživanje, i to sledeće sorte: egipatska detelina (*Trifolium alexandrinum* L), sorta Faraon; inkarnatska detelina (*Trifolium incarnatum* L), sorta Kardinal; jednogodišnji žuti zvezdan (*Lotus ornithopoides* L.) sorta Junak; hmeljasta vilja (*Medicago lupulina* L.) sorta Ekola i beli kokotac (*Melilotus albus* L.) sorta Adela. Ispitivane mahunarke značajno su se razlikovale u produktivnosti. One su rangirane po sledećem redosledu prinosa SM: beli kokotac – hmeljasta vija - jednogodišnji žuti zvezdan - egipatska detelina - inkarnatska detelina. Produktivnost i učešće egipatske deteline i inkarnatske deteline u travnjaku značajno varira u godinama. Biomasa inkarnatske deteline je imala najveći sadržaj sirovih proteina (15,24%), a najmanji sirovog vlakna (21,69%). Prema sveobuhvatne proceni podataka o produktivnosti i kvalitetu krme ispitivanih godišnjih mahunarki, hmeljasta vija bi se mogla definisati kao vrsta sa najvećim potencijalom za gajenje u travnjacima u uslovima centralne severne Bugarske. Ona se odlikuje visokom produktivnošću zelene mase i suve materije, ima sposobnost ponovnog rasta, ona se odlikuje visokim sadržajem sirovih proteina (14,92%) i sirove masti (4,66%), optimalnim sadržajem neutralnih i kiselih deterđent vlakana (34,67 i 24,99%) i visokim nivoom sadržaja hemiceluloze (9,68%). Energetska

vrednost krme ove vrste, vrednovana korišćenjem hranidbene jedinice mleka (Feed Unit of Milk -FUM) i rasta (Feed unit of growth - FUG) je ocenjena kao veoma visoka (FUM – 0,69/kg SM i FUG – 0,63/kg, SM).

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