

(*Trifolium pratense* L.)

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Germination and seed production of red clover (*Trifolium pratense* L.) from over-sown grassland and arable land

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SUMMARY

Red clover is the main forage crop in Slovakia. It is used for oversowing into grassland for five decades. This study was focused on laboratory germination, seedling establishment and seeds production of red clover from oversown grassland and arable land. Experiment was established in mountain region of Slovakia in Liptovská Teplička (960 m a.s.l.). Cultivar Mazurka was used in sowing rate of 400 seeds per square meter and spring sowing date. Laboratory germination test showed the best performance of seeds which comes from arable land. Significantly lower germination was recorded for seeds from oversown grassland. The native clover had the

lowest germination. In real conditions the seedling establishment was investigated. There were significant differences between the establishment of red clover on arable land vs. semi-natural grassland. Establishment of both oversown red clover into grassland was lower in comparison to arable land. Red clover seed production was the highest on arable land. Significantly lower values were recorded for every single treatment from grassland in comparison to arable land, but their particular differences were on the same level of significance. In summary, the red clover seed production from oversown was poor compared with its counterpart from arable land, but it perform better in reuse as oversowing into grassland.

Key words: germination, seed production, *Trifolium pratense*, grassland, arable land.

INTRODUCTION

Oversowing is a technology for improvement of grassland production and its quality. An idea to direct drill into existing grassland with plant species of good quality and outstanding performance is simple theoretically but it often ends with the failure (Kraj ovi , 1968). As Frame (1992, 2005) states sown seeds are introduced into a hostile environment where competition from the existing sward can be intense. It is a common experience that good initial germination and seedling emergence are followed by a rapid decrease in survival as various adverse factors come into play (Frame, 1992, 2005). So more than fifty years of research over this topic there have been tested different technic to seed bed preparation, sowing date (spring vs late summer/early autumn), sowing rate (reduced vs full ones), seed treatment (seed coated with fertilizers, and herbicides, pesticides, molluscicides, etc.), fertilization (timing, rate, ratio among elements including microelements), and,

(Kraj ovi , 1968).
Frame (1992, 2005)

(Frame, 1992,

2005).

Frame (1992)

Turnbull et al. (2004)

Aronne (2017),
 10^7 (Tilman, 2004)

Zuppinger-Dingley et al. (2014)

Dingley et al., 2014). (Zuppinger-

Züst et al. (2012)

Arabidopsis thaliana (L.) Heynh.
)

pratense L.) (Trifolium

(Kraj ovi , 1968)

of course, their combinations and different grasses, legumes and other herbs, as well.

According to Frame (1992) seed germination, establishment, and subsequent development and survival, all of them have to be encouraged. And as is given in work of Turnbull et al. (2004) large-seeded plants are competitive superior and their establishment in grassland is better compared with small-seeded plant species. Seed germination and seedling establishment in some direct drilling technology of a plant species are critical phases or bottlenecks *sensu* Aronne (2017) to set a population in a native grassland and seedling must both survive and increase mass by a factor of 10^3 to 10^7 or more to become a reproductive adult (Tilman, 2004).

However, one possible important question which has not been tested until now is a provenance of seed used in oversowing. For example, in an ecological experiment with species assemblages Zuppinger-Dingley et al. (2014) found selection for increased niche differentiation. It is the same plant species mixtures performed better when provenance comes from mixtures than these species were grown in monocultures. Selection in diverse mixtures may therefore increase species coexistence and ecosystem function in natural communities and may also allow increased mixture yield in agriculture or forestry (Zuppinger-Dingley et al., 2014). But, a plant species losses their valuable features as found Züst et al. (2012) in their evolutionary experiment: *Arabidopsis thaliana* (L.) Heynh. losses their defences (chemical and trichome density) in just five generation without attack of aphid herbivores.

Red clover (*Trifolium pratense* L.) is main forage plant species in mountain areas of Slovakia (Kraj ovi , 1968) with tradition of growing on arable land dated

18 (Dan ík et al., 1976).
 : (i)
 (ii)
 (*Trifolium pratense* L.)
 (*Polygono-
 Trisetion* Br.-Bl. ex Marshall 1947)

- back into second half of 18th century (Dan ík et al., 1976). It is used in
 - oversowing into grassland for five decades. Our research hypotheses were:
 - (i) the amount of red clover's seed produced after oversowing into grassland
 - is worse than from arable land, but (ii) it germinate and establish better than
 - counterpart from arable land when is oversowing is repeated.

The objective of this research was the laboratory germination, seedling establishment and seeds production of tetraploid red clover (*Trifolium pratense* L.) in oversown grassland (*Polygono-
 Trisetion* Br.-Bl. ex Marshall 1947) and on arable land.

MATERIAL AND METHODS

()
 (960 m . .) (4n)
 1 1.
 925 mm (525 mm)
 4.5 °C
 (9.5 °C)
 (,),
 (ACITA) (Halás,
 2001)
 3,30 mg.kg⁻¹ 97,02 mg.kg⁻¹)

We have been carrying out field experiment in a mountain area (Low Tatras Mts.) of Slovakia, in the cadastre of Liptovská Teplí ka village (960 m a.s.l.) with red clover cultivar Mazurka (4n) in a way outlined as are given in Table 1, and Scheme 1. In this Mountainous Plant Production Area is long-term sum of rainfall 925 mm annually (525 mm over growing period) and long-term average temperature is 4.5°C (9.5°C in growing period). The maternal substrate is formed by carbonates (limestones, dolomites), on which the neutral to weakly acidic rendzina soil was developed. According to Agricultural Central Inspection and Testing Authority of Slovakia (ACITA) (Halás, 2001) plant available concentrations of both phosphorus and potassium were low (3.30 mg.kg⁻¹ of P and 97.02 mg.kg⁻¹ of K, respectively).

1.

e

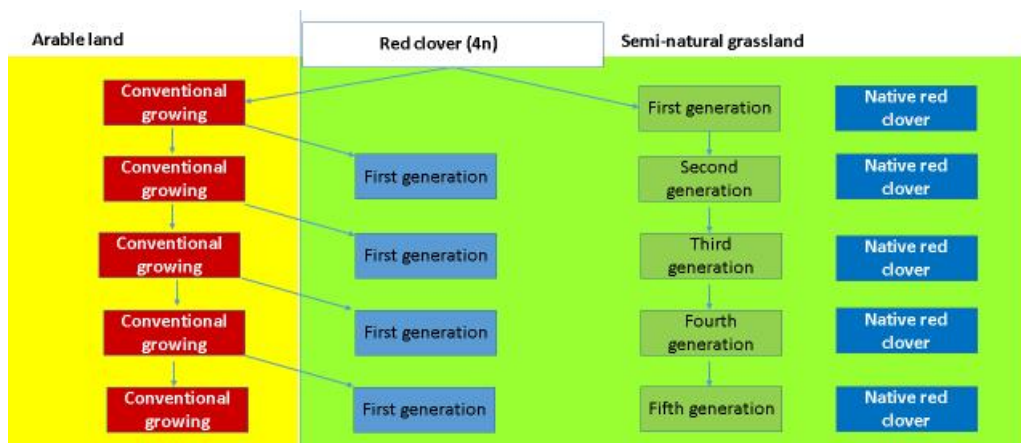
Table 1. Phases of cultivated red clover on arable land and oversown into permanent grassland

/Year	/Activity with red clover
2013	/Germination and establishment
2014	() / Seed production of red clover from both arable land and oversowing (first generation)
2015	/ Germination and establishment
2016	() / Seed production (second generation)
2017	/Germination and establishment
2018	() / Seed production (third generation)
2019	/Germination and establishment
2020	() / Seed production (fourth generation)
2021	/Germination and establishment
2022	() / Seed production (fifth generation)

Note: bold marked year means planned action

1.

Scheme 1. A schematically idea how do we manipulate with red clover over the course of the experiment



2013 . -
Polygono-Trisetion Br.-Bl.
 Marshall 1947 ..

In 2013 a part of semi-natural grassland, *Polygono-Trisetion* Br.-Bl. ex Marshall 1947 alliance, was ploughed and sown of red clover without a cover crop. And another part of the grassland was oversown. Sowing rates for oversowing of one-half to full rates are

400
 30 kg.ha⁻¹ (30 kg.ha⁻¹, 60 kg.ha⁻¹)
 3 × 5 m.
 28-
 Statit Custom QC.

recommended generally. We have been used the reduced sowing rate of 400 seeds per square meter and spring date of sowing.

Fertilizers (nitrogen 30 kg.ha⁻¹, phosphorus 30 kg.ha⁻¹ and potassium 60 kg.ha⁻¹) are applied on arable land after sowing immediately and on oversown grassland after first cut – in the germination and establishment phase. In the seed production phase we have been applying phosphorus and potassium fertilizers in the spring time, only. All plots had dimensions 3 × 5 m. All treatments (with two repetition) are mown three times over growing period (in the first phase of germination and establishment) and only one cut in second phase focused on seed production. Botanical composition is carried out before every cut. Amount of seed production are collected from two sub-plots per plot (for even ripening we use a desiccant). Laboratory germination is determined under standard International Seed Testing Association procedure and field establishment of red clover is evaluated on 28th day after sowing. All records compared with control – semi-natural grassland with presentation of native red clover (but without field establishment, that is, native red clover was not oversown).

All data were processed by Statit Custom QC.

RESULTS AND DISCUSSION

2
 (2013, 2015
 2017).
 (2013 .)

Meteorological conditions in critical months for germination, establishment or red clover’s seed ripening and long-term averages are given in Table 2. Over-sowing was carried out in the mid May and readings of seedlings was done in the mid-June (2013, 2015 and 2017). Only in the first year of the experiment (2013) were meteorologically favourable conditions for germination. Reduced rainfall and lower than average

2015 2017 .

temperature negatively influenced the germination and establishment in 2015 and 2017. On the other hand lower than average sum of rainfall in August and September caused that red clover seed ripening had required longer time. Shortly, shortage of rainfall throughout the first phases dampened germination and establishment, and wet conditions in the late summers/early autumns reduced seed production of red clover.

2.

Table 2. Sum of rainfall and average temperature in critical month for germination and establishment phase and seed production phase

Meteorology	Year	Phase	May	June	August	September
Rainfall (mm)	2013	G/E	143.0	154.7	58.1	86.5
	2014	S	164.1	80.8	74.1	46.0
	2015	G/E	83.7	22.1	21.5	132.6
	2016	S	47.0	69.8	91.4	61.2
	2017	G/E	102.6	69.8	133.4	130.6
	Long-term average		78.1	85.7	74.1	57.3
Temperature (°C)	2013	G/E	11.9	15.4	17.8	9.5
	2014	S	10.7	14.9	14.2	12.4
	2015	G/E	11.2	15.1	19.3	12.2
	2016	S	9.8	14.6	13.9	12.2
	2017	G/E	10.5	14.9	16.3	9.9
	Long-term average		12.0	15.2	16.4	11.3

Note: G/E – germination and establishment, S – seed production

Results of red clover in laboratory conditions suggest the seeds obtained from arable land germinate significantly better than ones raised in oversown grassland. Native red clover seeds had the lowest laboratory germination.

These differences were statistically significant (ANOVA repeated measures: Model $F_{3, 12} = 17.8$, $P < 0.0001$). Germination of both oversown treatments

(ANOVA : $F_{3, 12} = 17.8$, $P < 0.0001$).

($t = -1.7, P = 0.18$).

was not different (t -test $-1.7, P = 0.18$). Our partial second hypothesis (laboratory germination) is not confirmed. And all tested seeds have germinated after scarification.

3.

Table 3. Laboratory germination of red clover's seed

Treatment	Standard germination (% ± S.E.M.)	Germination after scarification (%)
Arable land	55.4 ± 4.1 ^c	100
First generation only (2)	31.1 ± 6.1 ^b	100
All (2) generation in oversowing	40.7 ± 1.8 ^b	100
Native red clover	15.3 ± 2.5 ^a	100

(2)

S.E.M -

Treatment 'First generation only' means every bi-year oversowing into semi-natural grassland, and All (2) generation treatment means repeatedly oversowing of red clover is repeatedly oversown red clover.

S.E.M – standard error of mean.

(. . -)

4.

ANOVA

($F_{2,33} = 6.94, P = 0.003$).

115.5, $P = 0.001$,

95.5, $P = 0.007$,

).

$P = 0.55$).

Red clover seedling establishment in the tested treatments are given in Table 4. There are significant differences between establishment of red clover on arable land vs semi-natural grassland ANOVA repeated measures revealed (Model $F_{2,33} = 6.94, P = 0.003$).

Establishment of both oversown red clover into grassland was lower than on arable land (t -test 115.5, $P = 0.001$, between first generation and arable land, and t -test 95.5, $P = 0.007$, between repeatedly oversown red clover and red clover on arable land).

Non-significant difference was detected when we compared establishment between red clover establishments oversown into semi-natural grassland (t -test $-20.0, P = 0.55$). Here, we partially answered second hypothesis

(establishment of red clover): on average, after three readings, red clover does not differ where its seed is produced between arable land (first generation treatment) or from oversown grassland (three generation in oversowing, yet).

Frame (2005) has reported 47 per cent seeds of red clover can establish on arable land. Our results with 46.1 per cent (Table 3) are the same. Britaák (2008) found in oversowing of the same species, but different cultivars (Vesna – 4n, Nodula – 4n, Rezista – 4n, and Tábor – 2n), approximately the same rate (27.1, 27.3, 23.3 and 21.0, respectively).

(Vesna-4n, Nodula-4n, Rezista-4n Tabor-2n),

(establishment of red clover): on average, after three readings, red clover does not differ where its seed is produced between arable land (first generation treatment) or from oversown grassland (three generation in oversowing, yet).

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4.

Table 4. Average establishment of red clover in the fields (three cycles)

/Treatment	/Seedling per square meter (pcs ± S.E.M)	/Percentage of sowing rate (%)
Arable land	184.8 ± 27.8 ^b	46.1
First generation only (3)	69.3 ± 22.2 ^a	17.3
/ All (3) generations in oversowing	89.3 ± 19.5 ^a	22.3

: 2014 2016.
2018 . -

(5).
8
11 (ANOVA
 $F_{3, 28} = 8.9, P < 0.0001$).

(t- 207.4, 205.4 200.4 ,

- Seed production of red clover was carried out in two years: 2014 and 2016. Next will be in this 2018 year. The best seed production of red clover was detected unsurprisingly on arable land (Table 5). This treatment outperformed the others about 8 to almost 11 times (ANOVA repeated measures $F_{3, 28} = 8.9, P < 0.0001$). All pairwise comparisons between seed production of red clover from arable land and treatments from grassland were statistically significant (t-test 207.4, 205.4, and 200.4 for comparison with first generation, native red clover and all (2) generation in oversowing, respectively). Seeds of red

(2) -

).

2014 . (37,2 kg.ha⁻¹) -

Ilavská (2016) 2016 . (113,4 kg.ha⁻¹). -

" " 119 kg.ha⁻¹. -

(-

54,4 kg.ha⁻¹). Holúbek et al. -

(2007) 100 -

400 kg.ha⁻¹ -

).

2016 . -

402,8 kg.ha⁻¹ .

Holubek et al. (2007) -

400 700 kg.ha⁻¹, Frame -

(2005). Cagaš (2018) -

kg.ha⁻¹ 186,8 -

324,8 kg.ha⁻¹ 2016 . -

(-

a -

)

- clover from any treatments of semi-natural grassland did not differ statistically.

Average seed production in 2014 (37.2 kg.ha⁻¹) was marginally lower than in 2016 (113.4 kg.ha⁻¹). Ilavská (2016) found that in 2014 seed production of red clover cultivar Mazurka was 119 kg.ha⁻¹. In our experiment it was lesser than half of this production (arable land 54.4 kg.ha⁻¹). Holúbek et al. (2007) reported typical yield from 100 to 400 kg.ha⁻¹ of red clover seed in Slovakia. We have to note our growing conditions are not suitable for seed production, but the opposite is true for oversowing (or forage production). Even if in this harsh condition can be some opportunity to produce seeds. In 2016, red clover on arable produced 402.8 kg.ha⁻¹ of seeds. It was in the range presented by Holubek et al. (2007) and lower limit of range 400 to 700 kg.ha⁻¹ reported by Frame (2005). Cagaš (2018) informed us about average seed production of red clover on a country level of the Czech Republic. On average, 186.8 kg.ha⁻¹ was produced in 2014 and 324.8 kg.ha⁻¹ in 2016.

So, our expectation in the first hypothesis (seed production of red clover oversown into grassland is worse than their counterpart on arable land) is fulfilled.

5.

Table 5. Seed production of red clover in the fields (two cycles completed)

/Treatment	/Seed production (kg.ha ⁻¹)
/Arable land	228.6 ± 67.3 ^b
/First generation only	21.2 ± 7.5 ^a
(2)	28.2 ± 10.0 ^a
All (2) generation in oversowing	
/Native red clover	23.2 ± 8.2 ^a

CONCLUSIONS

- We are in the half of experimentation with obtaining of seeds of red clover from arable land and
- oversowing of this species into grassland.
- Ability of red clover to germinate and establish better in grassland after oversowing does not differ significantly when provenance of seed is from arable land. But, there is some trend favouring reuse seed from oversowing in all tested indicators.

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" 89, 5800

Forage yield in competitive variety testing of tetraploid perennial ryegrass

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SUMMARY

2007-2009 () -
() 4
, 2
: 1.
-
NBG, 2. SBG
3. Roy 4. Pandora.
(kg ha⁻¹)
,
,
, %.
,
,
-
7390,7 kg ha⁻¹ NBG 6589,9 kg ha⁻¹

During the period 2007-2009 in the Institute of Forage Crops (IFC) - Pleven on black soil without irrigation competitive variety trials (CVT) for forage with 4 variants tetraploid perennial ryegrass, two Bulgarian breeding populations: 1. NBG - T, 2. SBG - T and two Belgian varieties - standards 3. Roy, and 4. Pandora was carried out. The biological and agricultural values were studied in terms of forage yield and its elements to describe and apply with the breeding numbers for official state variety testing. The forage fresh and dry matter yield (kg ha⁻¹) by cuts, years, total and average for the period, and its seasonal and total distribution were determined as participation and percent. It was found that Bulgarian tetraploid perennial ryegrass breeding populations are durable, highly forage productive and with high adaptive potential of the conditions of the country. Average annual forage dry matter yields are similar and higher values for the Bulgarian breeding tetraploid populations – 7390.7 kg ha⁻¹ for NBG and 6589.9 kg ha⁻¹ for SBG. The excess of the

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 (Paul, 1988; Cai et al., 2014; Katova, 2016;
 Katova et al., 2016). Van
 Bockstaele (1998) Lamote et al. (1999),
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has been found that tetraploids have many positive qualities: better leafness, higher plants, higher forage and seed productivity, higher water and carbohydrates content in the cells, and higher digestibility by diploids (Paul, 1988; Cai et al., 2014; Katova, 2016; Katova et al., 2016). According to Van Bockstaele (1998) and Lamote et al. (1999), the most important differences between diploid and tetraploid varieties are: a darker green leaf color; faster germination and sward establishment; a greater number of stem leaves; a larger 1000 seeds weight; higher WSC content and lower cell wall thickness; better palatability; higher level of intake of fresh biomass by ruminant; better disease resistance; higher nutritional value; better absorption of nitrogen; lower dry matter content.

The aim is to investigate the biological and economic properties in terms of forage production in order to declare the first tetraploid selection numbers for official state variety testing.

MATERIAL AND METHODS

2007 03 e
 - -
 , 4 - 2
 :
 1. NBG – T, 2. SBG – T
 - - 3. Roy, 4.
 Pandora. 200 kg
 ha⁻¹ P₂O₅ N – 50 kg ha⁻¹. N
 60 kg ha⁻¹ . .
 4
 5 m² 12 cm
 50 kg ha⁻¹.
 75%
 20.04.
 - 14.05.2007 .

In 2007, on April 3, a field experiment was set up – Competitive Variety Trail for forage from perennial ryegrass in IFC - Pleven on leached black soil without irrigation, with 4 variants – 2 Bulgarian tetraploid breeding populations: 1. NBG – T, 2. SBG – T two Belgian varieties – standards – 3. Roy, and 4. Pandora. 200 kg of ha⁻¹ P₂O₅ and N-50 kg of ha⁻¹ have been introduced before sowing. Each year is imported twice in equal quantities N 60 kg ha⁻¹ a. s. in the spring and autumn. The scheme is a Block Method in 4 replications, a plot size of 5 m² at 12 cm spacing and a sowing rate of 50 kg ha⁻¹. The germination and tillering were reported in 75% of emerged plants on 20.04. and on 14.05.2007.

One cut for the year was made in the year of trail establishment on 31.10.2007. In

31.10.2007 .
 kg ha⁻¹
 (3628,6 kg ha⁻¹),
 29,31%,
 1.
 , I , 31.10.2007 .
 Roy (4142,0
 NBG
 27, 34

productivity are our focus on competitive variety testing.

There was a harvest - one cut in the year of establishing on the trail on 31.10.2007. Table 1 shows the yields of fresh and dry mass of CVT for forage and percentage of dry matter. The highest yield is the standard variety Roy (4142.0 kg ha⁻¹) and the breeding population NBG (3628.6 kg ha⁻¹), where the percentage of dry matter is higher 27, 34 and 29.31%, respectively.

1. , (kg ha⁻¹)
 , I , 31.10.2007 .

Table 1. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, I cut - 31.10.2007

Variant	Fresh mass	Dry matter, %	Dry mass	Rank
1. NBG	12380	29,31	3628,6	2
2. SBG	13770	25,16	3464,5	3
3. Roy	15150	27,34	4142,0	1
4. Pandora	9830	24,12	2371,0	4

2008 .
 2
 2
 %
 I (29.04.08).
 - SBG,
 NBG, . .

In 2008, three cuts in CVT for forage for Bulgarian numbers and 2 for Belgian varieties were harvested. Table 2 shows the yields of fresh and dry biomass and % dry matter for I cut (29.04.08). The highest dry mass yield is SBG, followed by NBG, ie. Bulgarian numbers.

2. , kg ha⁻¹
 , I , 29.04.2008 .

Table 2. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, I cut - 29.04.2008

Variant	Fresh mass	Dry matter, %	Dry mass	Rank
1. NBG	31427,2	21,23	7856,8	2
2. SBG	37874,6	21,74	9468,6	1
3. Roy	28448,9	28,57	7112,2	3
4. Pandora	25692,1	25,14	6423,0	4

3
 II
 (12.06.08).
 ,

In Table 3 are the data on the forage yield from the 2nd cut (12.06.08). There was a change in the ranks, the first position being occupied by NBG (2818.6

NBG (2818,6 kg ha⁻¹),
 Pandora (2054,0), SBG
 (1772,1 kg ha⁻¹).
 ,
 -
 29.10.2008 .
 (4). NBG -
 SBG, 1253,5
 880 kg ha⁻¹ .

kg ha⁻¹) and the second of the Pandora
 variety (2054.0), while SBG was in the
 4th position (1772.1 kg ha⁻¹). Due to the
 three-month drought and high summer
 temperatures, the plants fell in summer
 dormancy and the third regrowth was
 formed only with Bulgarian numbers and
 was harvested on 29.10.2008 (Table 4).
 NBG has a higher forage yield of SBG,
 respectively 1253.5 and 880 kg ha⁻¹ dry
 matter.

3. , kg ha⁻¹
 , II , 12.06. 2008 .

Table 3. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, II cut - 12.06. 2008

Variant	Fresh mass	Dry matter, %	Dry mass	Rank
1. NBG	11274,4	27,05	2818,6	1
2. SBG	7088,2	29,05	1772,1	4
3. Roy	8152,3	30,06	2038,1	3
4. Pandora	8216,0	28,06	2054,0	2

4. , kg ha⁻¹
 , III , 29.10. 2008 .

Table 4. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, III cut - 29.10. 2008

Variant	Fresh mass	Dry matter, %	Dry mass	Rank
1. NBG	5014,2	32,06	1253,5	1
2. SBG	3523,4	33,24	880,9	2
3. Roy	-	-	-	-
4. Pandora	-	-	-	-

5
 .
 SBG –
 12121,5 kg ha⁻¹, NBG –
 11928,2 kg ha⁻¹.
 Roy Pandora – 8813,6 kg
 ha⁻¹,
 37,03% 35,35%.

Table 5 shows the annual forage
 yield of dry matter and the ranges for
 these key characteristics. The first
 position is SBG - 12121.5 kg ha⁻¹,
 followed by NBG - 11928.2 kg ha⁻¹.
 Compared to the average standard of
 Roy and Pandora - 8813.6 kg ha⁻¹, the
 excess is 37.03% and 35.35%
 respectively.

5.
, 2008 .

(kg ha⁻¹)

Table 5. Annual dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, 2008

Variant	I I cut	II II cut	III III cut	Total	Rank	% to average St
1. NBG	7856,8	2818,6	1253,5	11928,9	2	135,35
2. SBG	9468,6	1772,1	880,9	12121,5	1	137,53
3. Roy	7112,2	2038,1	0,00	9150,3	3	103,82
4. Pandora	6423,0	2054,0	0,00	8477,0	4	96,18
Average from St (3 and 4)	6767,6	2046,1	0,00	8813,6		100

6
%
,
-
65,86 - 23,96 - 10,51%.
75%
SBG -
7%
III
6.
%

Table 6 shows the distribution of the yield in % of the annual yield by cuts, with relatively the most uniform for NBG, respectively 65.86 - 23.96 - 10.51%.

The other three numbers account for over 75% of the annual yield in the I cut and the remaining share in the second cut for the Belgian varieties, and for the Bulgarian SBG - a significant share - 7% occupies the third cut.

, 2008 .

Table 6. Dry matter yield distribution, % by cuts of tetraploid perennial ryegrass, 2008

Variant	I I cut	II II cut	III III cut	Sum 2008
1. NBG	65,86	23,96	10,51	100
2. SBG	78,11	14,62	7,27	100
3. Roy	77,73	22,27	0	100
4. Pandora	75,77	24,23	0	100

2
7.
%
(28.04.09).
NBG (4312,7 kg ha⁻¹),
SBG (2896,4 kg ha⁻¹), . .

For the third year are harvested three cuts for the Bulgarian numbers and 2 for the Belgian varieties. Table 7 presents the data on the yield of fresh and dry biomass and % dry matter for the first cut (28.04.09). The highest dry matter yield is NBG (4312.7 kg ha⁻¹), followed by SBG (2896.4 kg ha⁻¹), ie. Bulgarian numbers.

7. , I , 28.04.2009 .
Table 7. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, I cut -28.04.2009

Variant	Fresh mass	Dry matter	Dry mass	Rank
1. NBG	16840,0	25,61	4312,7	1
2. SBG	11750,1	24,65	2896,4	2
3. Roy	3030,1	32,23	976,6	3
4. Pandora	2119,7	31,74	672,8	4

8. II
 (16.06.09).
 NBG (1779,6 kg ha⁻¹),
 SBG (931,0 kg ha⁻¹).
 23.10.2009 . (9). NBG
 522,2 356,2 kg ha⁻¹
 SBG,

In Table 8 are the data on the forage yield from the 2nd cut (16.06.09). Ranking stability is observed, with the first position occupied by NBG (1779.6 kg ha⁻¹) and second by SBG (931.0 kg ha⁻¹). Due to drought and high summer temperatures, the plants fell into summer dormancy and the third cut was formed only for the Bulgarian numbers and was harvested on 23.10.2009 (Table 9). NBG has a higher forage yield of SBG, respectively 522.2 and 356.2 kg ha⁻¹ dry matter.

8. II , 16.06. 2009 .
Table 8. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, II cut - 16.06. 2009

Variant	Fresh mass	Dry matter	Dry mass	Rank
1. NBG	4230,1	42,07	1779,6	1
2. SBG	1980,0	47,02	931,0	2
3. Roy	2219,9	39,34	873,3	3
4. Pandora	1390,0	48,98	680,8	4

9. III , 23.10. 2009 .
Table 9. Fresh and dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, III cut, 23.10. 2009

Variant	Fresh mass	Dry matter	Dry mass	Rank
1. NBG	1640,1	31,84	522,2	1
2. SBG	1080,0	32,98	356,2	2
3. Roy	-	-	-	-
4. Pandora	-	-	-	-

10

6614,5 kg ha⁻¹,
 kg ha⁻¹.
 Roy Pandora – 1601,8 kg ha⁻¹,
 412% 261%.

NBG –
 SBG – 4183,6

Table 10 shows the annual yield of forage dry matter and the ranges for this key characteristic. The first position is NBG - 6614.5 kg ha⁻¹, followed by SBG - 4183.6 kg ha⁻¹. Compared to the average standard of Roy and Pandora - 1,601.8 kg ha⁻¹, the excess is 412% and 261% respectively. The explanation of the great differences in the productivity of the Bulgarian and Belgian tested variants is the durability, which is characteristic only for the local adapted breeding numbers. For similar results under Japan's conditions, which are less favorable for perennial ryegrass cultivation, tetraploid varieties show better productive and adaptive potential (Cai et al., 2014).

(Cai et al., 2014).

10.
 , 2009 .

kg ha⁻¹

Table 10. Annual dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, 2009

Variant				Total	Rank	% to average St
	I cut	II cut	III cut			
1. NBG	4312,7	1779,6	522,2	6614,5	1	412,94
2. SBG	2896,4	931,0	356,2	4183,6	2	261,18
3. Roy	976,6	873,3	0,00	1849,9	3	115,49
4. Pandora	672,8	680,8	0,00	1353,6	4	84,50
Average from St (3 and 4)	824,7	777,1	0,00	1601,8		100

11

%

65,2 - 26,9 - 7,9%.
 49,7%

I () .

11.

, 2009 .

%

Table 11. Dry matter yield distribution, % by cuts of tetraploid perennial ryegrass, 2009

Variant				Sum 2009
	I cut	II cut	III cut	
1. NBG	65,2	26,9	7,9	100
2. SBG	69,2	22,3	8,5	100
3. Roy	52,8	47,2	0	100
4. Pandora	49,7	50,3	0	100

2009 .) (2007 –
 12
 4-
 -
 NBG (22172,0 kg ha⁻¹),
 SBG (19769,7 kg ha⁻¹), . .
 -
 -
 28-62 % NBG 14-45 %
 SBG.

For the three-year period (2007-2009) according to the VCU methodology in Table 12, the yield data for dry biomass is shown in total and average for the period and as a percentage against the average of the 4 numbers and the average of the two Belgian varieties.

With the highest total dry matter yield is NBG (22172.0 kg ha⁻¹), followed by variant 2 (19769.7 kg ha⁻¹), ie. Bulgarian numbers. The excess over the above standards is 128-162% for NBG and 114-145% for SBG respectively.

12. , kg ha⁻¹
 (2007-2009 .)

Table 12. Dry matter yield (kg ha⁻¹) of tetraploid perennial ryegrass, (2007-2009)

Variant	/ Dry matter forage yield , (kg ha ⁻¹)				Annual average	Ran k	% to	
	2007	2008	2009	Total			St (3+4)	% to average St (1+2+3+4)
1. NBG	3628,6	11928,9	6614,5	22172,0	7390,7	1	162	128
2. SBG	3464,5	12121,6	4183,6	19769,7	6589,9	2	145	114
3. Roy	4142,0	9150,3	1849,9	15142,2	5047,4	3	111	87
4. Pandora	2371,0	8477,0	1353,6	12201,6	4067,2	4	89	70
% to St (3+4)					4557,3		100%	
% to St (1+2+3+4)					5773,8		100%”	

13
 , %
 100%.
 16,2%
 27,4%
 NBG,
 Roy.
 53,8 % NBG 69,5%
 Pandora.
 - NBG 30
 % SBG 21,2%,
 -
 Pandora 12, 2% Roy. 11,1%

Table 13 shows the distribution of dry mass yield per year, per share, % of the total yield for the survey period, assumed to be 100%. In the year of sward establishment the share of the yield is from 16.2% for perennial ryegrass NBG, up to 27.4% for Roy variety. In the second year, maximum productivity is achieved for all variants, with a share of total productivity ranging from 53.8% for NBG to 69.5% for Pandora perennial ryegrass variety. The yield as a share of total productivity in the third year is the highest for NBG 30% and SBG 21.2%, while the Belgian lowest for the whole period 11.1% for Pandora and 12.2% for Roy.

13.

%

, 2007-2009 .

Table 13. Dry matter yield distribution, % by years of tetraploid perennial ryegrass, 2007-2009

/ Variant	2007	2008	2009	/ Sum 2007-2009
1. NBG	16,2	53,8	30,0	100
2. SBG	17,5	61,3	21,2	100
3. Roy	27,4	60,4	12,2	100
4. Pandora	19,4	69,5	11,1	100

2008 -

2009 -

1/3 1/2

()

10). NBG SBG 1 2 (

45 62 %.

ha⁻¹ 4067,2 kg ha⁻¹,

– 7390,7 kg ha⁻¹

NBG 6589,9 kg ha⁻¹ SBG.

Roy

Pandora, 5047,4 kg

2008 is more favorable for perennial ryegrass grassland grown for forager, while 2009 is limiting - 1/3 to 1/2 of the forage yield of the previous year. Of interest are the varieties which are ecologically stable, i.e., with high production potential under favorable conditions and with good productivity in adverse (limiting) environmental conditions. More stable in the unfavorable 2009 are the Bulgarian breeding populations NBG and SBG ranks 1 and 2 (Table 10).

For Belgian varieties, forage productivity is strongly influenced by environmental conditions, as the conditions under which they are selected are characterized by different values of temperatures and rainfall.

CONCLUSIONS

The Bulgarian tetraploid selection populations of perennial ryegrass have been found to be long-lasting, high-productive for forage and with high adaptive potential for the country's conditions. The average annual forage yield data are close and higher for the Bulgarian selection tetraploid numbers – 7390.7 kg ha⁻¹ for NBG and 6589.9 kg ha⁻¹ for SBG. The excess over the standards is 145 to 162%. The Belgian variety Roy is higher in forage yield than Pandora, 5047.4 kg ha⁻¹ and 4067.2 kg ha⁻¹, respectively.

Varieties and populations have a different multi-year strategy and differ in

NBG	-	-	the number of cuts and distribution of yields. The highest forage yield has the NBG as a result of the largest number of cuts and the most even distribution over seasons over the years, followed by SBG, which has the same number of cuts.
SBG,	-	-	Belgian varieties have less longevity under our conditions, lower forage yields, a smaller number of cuts, as forage productivity is strongly influenced by environmental conditions and they are selected under conditions characterized by different values of temperatures and precipitation.

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Breeding assessment of the water-soluble carbohydrates content in perennial grasses accessions

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SUMMARY

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2017 . -
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35 , 50
21 (15 6 , 11
10 ,
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2 , 5
3 , - 6 (3
, (1987)
, ,
,
Kannenber (1978)

- The aim is to determine the water-soluble carbohydrates (WSC) content of collections accessions of perennial ryegrass, cocksfoot, tall, meadow and red fescue and to make the selection. During the period 2015-2017 in the IFC-Pleven three collections were studied on leached black soil, rain fed conditions, by block method in 2 repetitions a total of 35 accessions, each of with 50 individual plants: perennial ryegrass – 21 (15 varieties and 6 ecotypes, 11 tetraploids and 10 diploids, from Bulgaria, Belgium, Romania and China); fescue – 8 (3 – tall, 3 – red and 2 meadow, form them 5 varieties and 3 ecotypes, from Bulgaria and Romania); cocksfoot – 6 (3 varieties and 3 ecotypes from Bulgaria and Romania). The WSC content was determined by Ermakov et al. (1987) and average, minimum, maximum values, standard deviations by regrowths and years, and total mean for the collections. According to Frances and Kannenberg (1978) the breeding assessment was done by genotypes distribution against the collections average values for WSC and coefficient of variation. It was found that WSC content varied according to the plant

Melverde, Meracoli, Melpetra,

BGR 1120.

: Adela, Atoluka Ravnogor.

(Cunningham et al., 1994; Humphr ys et al., 2010; Katova, 2005, 2016).

Festuca

(Jones

species, genotype (variety or ecotype, ploidy level, group of maturity) and seasonal differences (regrowths and years), also growing conditions. The highest WSC content was measured in perennial ryegrass, followed by fescues and the lowest in cocksfoot. The perennial ryegrass genotypes: Meltador, Melverde, Meracoli, Melpetra, Harmoniya and Ravnogor with increased WSC content above year average value for the collection during the two years of the study were selected, as the tetraploid varieties had higher WSC content. The cocksfoot genotypes with higher WSC content above collection average were selected: Dabrava and BGR 1120. The WSC content in the fescue occupies an intermediate position between the perennial ryegrass and the cocksfoot and genotypes of reed and red fescue with a higher content than the average annual value for WSC of the collection were selected: Adela, Atoluka Ravnogor.

Key words: perennial ryegrass, cocksfoot, fescue, varieties, ecotypes, water-soluble carbohydrates, breeding assessment

INTRODUCTION

Perennial ryegrass is one of the most researched and valuable temperate pasture grasses in the world (Cunningham et al., 1994; Humphr ys et al., 2010; Katova, 2005, 2016). Cocksfoot has a high and stable quality biomass yield with a large range of climate tolerance. It is frost-, shade-, heat and drought tolerant with good regrowth ability. Fescues are very diverse grasses which are important components of natural, permanent, and intensively managed grasslands, lawns, and turfs, and are used for conservation purposes. The content of water-soluble carbohydrates (WSC) has a beneficial effect on the intake, digestion and use of feed (Jones and Roberts, 1991, Miller et

and Roberts, 1991; Miller et al., 2001; Halling et al., 2004, Masahito, 2011).

(Jolaosho et al., 2009; Luscher et al., 2014). Naydenova and Vasileva (2016) Kirilov and Vasileva, (2016)

(Wilkins and Lovatt 2007, Wilkins et al., 2010) (Rasmussen et al., 2009).

N (Lee et al., 2001; Evans et al. 2011, Miller et al., 2001).

()

(Humphreys, 1994).

pH (Frame, 1994),

(Hayward and McAdam,1988; Smith et al., 1998). (Haigh, 1990),

(3,7 %
15 %).

al., 2001, Halling et al., 2004, Masahito, 2011). Increasing the WSC content improves protein - carbohydrate balance, both in plants and animals, increases digestibility of proteins and reduces the content of nitrates in the environment (Jolaosho et al., 2009; Luscher et al., 2014). Naydenova and Vasileva (2016) and Kirilov and Vasileva, (2016) found that the cocksfoot had a better quality of the tall fescue feed and was more preferred than the sheep. In the last decade, few improved ryegrass varieties had been bred for increase WSC concentration, in UK (Wilkins and Lovatt 2007; Wilkins et al. 2010), and New Zealand (Rasmussen et al., 2009). Most published data suggest that animals consuming grasses with high WSC concentrations were able to utilize the protein in their diet more efficiently, resulting in increased liveweight gain, milk production and lower loss of N (Lee et al., 2001; Evans et al., 2011; Miller et al., 2001). WSC and crude protein (CP) are the main components of the plant cell content (Humphreys, 1994). The chemical composition of the grass feed is influenced by the following factors: development phase, plant part, annual season, soil pH and fertilization status (Frame, 1994), and of course by genotype. Higher WSC content is important for perennial ryegrass varieties used in dry areas, which are characterized by a decrease in digestibility in the summer and its nutritional value in autumn (Hayward and McAdam, 1988; Smith et al., 1998). According to Haigh (1990), the required WSC content to provide fermentation for silage of non-additive perennial grasses is at least 3.7% of the fresh mass (or about 15% of the dry mass). The WSC content is one of the main quality criteria for fermentation in silage (Wilkinson et al., 1981). Beever (1993) believes that the breeding of fodder grasses must increasingly be linked to the animal's requirements for a suitable type of diet,

(Wilkinson et al., 1981).
 Beever (1993) ,
 -
 ,
 -
 -
 Carlier
 (1994) “ ”
 50-60 %
 12% 100%
 . Carlier and Van Waes (2000)
 , % :
 - 47, - 15,
 15, - 3, - 10,
 - 25.
 -
 170 (min 46, max 315) g kg⁻¹ (McDonald
 et al., 1981), -
 . Humphreys and Turner (2002)
 27%
 -
 -
 -
 -
 -
 (Bugge, 1978; Marais et al.,
 1993; Humphreys, 1989 a, b, c; Jafari, 2012;
 Katova and Ilieva, 2017).
 -
 (Gilliland et al., 2002, Ghesquiere and
 Baert, 2014; Katova, 2005, 2016).
 -
 (Frandsen, 1986).
 -
 ,
 (Fojtik and Horák, 1991).
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 (Fulkerson
 et al., 2004).
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improving the availability of carbohydrates
 and controlling the degradability of the
 protein.
 According to Carlier (1994), the "ideal"
 grass to the animals should contain 50-
 60% of accessible carbohydrates and
 12% of protein with 100% absorption.
 Carlier and Van Waes (2000) report the
 following chemical composition of
 perennial ryegrass, in% of DM: non-
 extractable substances - 47, CP-15,
 WSC-15, fat 3, ash -10, crude fiber -25.
 Among the grasses of moderate climate,
 perennial ryegrass has the highest WSC
 content - an average of 170 (min 46, max
 315) g kg⁻¹ (McDonald et al., 1981), and
 the lowest in the cocksfoot. Humphreys
 and Turner (2002) found a 27% variation
 of WSC in perennial ryegrass varieties.
 The genetic variation of WSC in the
 perennial ryegrass is higher than that of
 the CP or the DM digestibility (Bugge,
 1978; Marais et al., 1993; Humphreys,
 1989 a, b, c; Katova and Ilieva, 2017).
 The tetraploid varieties have a higher
 WSC content (Gilliland et al., 2002,
 Ghesquiere and Baert, 2014; Katova,
 2005, 2016). In many studies, a negative
 correlation between yield and quality
 parameters has been found (Frandsen,
 1986). In some tetraploid varieties of
 perennial and Italian ryegrass the level of
 CP remains unchanged, while the content
 of WSC and, due to palatability, increase
 (Fojtik and Horák, 1991). WSC often have
 higher inheritance than other quality
 indicators. The most productive and
 persistent perennial ryegrass varieties
 tend to have a higher WSC content
 (Fulkerson et al., 2004). The increased
 content of WSC in new varieties of
 perennial ryegrass is considered a
 progress in the plant breeding.
 The aim is to determine the water-
 soluble carbohydrates (WSC) content of
 collections accessions of perennial
 ryegrass, cocksfoot, tall, meadow and red
 fescue and to make the selection of the

- stable genotypes with higher WSC content.

MATERIAL AND METHODS

2015 .
 3
 () 35
 ,
 50/50 cm,
 50
 - 25 25 -
).
 da⁻¹ . . 6 kg N
 (NH₄NO₃)
 .
 5-7 cm.
 - 4
 .
 -1 -
 (*Lolium perenne* L.) 21
 (15 6 ; -
 - 9, - 9, -
 2 - 1; - 10
 11 ; 2 -
Festuca - 8 (3
 (*Festuca arundinacea* Schreb.), 2
 (*Festuca pratensis* L.)
 (*Festuca rubra*),
 3 3 , 4
 3 1 ; 3 - (*Dactylis*
glomerata L.) 6
 (3 3 , 4
 2).
 :
 2016 . 2 (15), 3
 (11, 12, 13,14, 17, 31, 32, 33, 34) 4
 (1,2,3,4,5,6,7,8,9,10,16),
 2017, 3 (33), 4 (1,5,6,7,15,31,32,34)
 5 (2,3,4,8,9,10,11,12,
 13,14,16 17); - 2016 -

In the autumn of 2015 in the experimental field of IFC - Pleven, three collections (KN) with a total of 35 numbers – varieties and ecotypes of perennial grasses, with individual plant layout, by seedlings at a distance of 50/50 cm , each sample is represented by 50 individual plants – 25 for fodder and 25 for seeds) were established. During the vegetation, twice a year, individual fertilization of the plants with 60 kg N ha⁻¹ a.s. in the form of ammonium nitrate (NH₄NO₃), spring and autumn. The experiments were carried out in non-irrigating conditions. The harvesting of the green mass was made individually by manual mowing with a sickle at a height of 5-7 cm. The first cut is in the beginning of heading, according to maturity groups, and the next one is at approximately 4 weeks interval. The characters are measured in the second and third year of establishment of the collections. Collection nursery KN1 involves – perennial ryegrass (*Lolium perenne* L.) 21 accessions (15 varieties and 6 ecotypes; by origin – Bulgarian – 9, Belgian – 9, Romanian – 2 and Chinese – 1; by ploidy level – 10 diploids and 11 tetraploids; KN 2 – *Festuca* – 8 accessions (3 tall fescue (*Festuca arundinacea* Schreb.), 2 meadow fescue (*Festuca pratensis* L.) and 3 red fescue (*Festuca rubra* L.), from them 5 are varieties and 3 ecotypes, 4 Bulgarian, 3 Romanian and 1 Belgian; N 3 – (*Dactylis glomerata* L.) involves 6 accessions of cocksfoot (3 varieties and 3 ecotypes, 4 Bulgarian and 2 Romanian). Various number of cuts for different collection specimens are implemented annually: for perennial ryegrass in 2016 - 2 (15), 3 (11, 12, 13,14, 17, 31, 32, 33, 34) or 4 cuts (1,2,3,4,5,6,7,8,9,10,16), and during 2017 - 3 (33), 4 (1,5,6,7,15,31,32,34) or 5 cuts (2,3,4,8,9,10,11,12,13,14,16 17); for cocksfoot – 2016 – 3 cuts

3 (25,26,27,28,29,30), 2017 – 5
 (30) 6 (25,26,27,28,29)
 – 2016 . - 2 (19,22,23,24)
 3 (18,20,21),
 2017 . - 5 (21,22,23,24) 6
 (18,19,20 35).

al. (1987).

Excel, P=0,05)
 (min max),
 (), (SD)
 (CV, %).
 CV,
 : 10%; >10-20%, >20 %
 (Dimova and Marinkov, 1999). Fransis
 and Kannenberg (1978)

, % CV, %.

(25,26,27,28,29,30), 2017 – 5 (30) or
 6 cuts (25,26,27,28,29) and for fescue
 – 2016 - 2 (19,22,23,24) or 3 cuts
 (18,20,21), and 2017 - 5
 (21,22,23,24) or 6 cuts (18,19,20 and
 35).

Chemical analyses: Water soluble carbohydrates (WSC), % DM – their content is determined by the colorimetric method of Ermakov et al. (1987). The method is based on the color change of the copper glycerate by boiling with aqueous extracts of sugars. For analysis, average samples were taken from each accession of the plant samples, dried at 60 ° C to constant weight. The WSC are extracted with hot water, hydrolysed with hydrochloric acid and copper glycerate is added.

Statistical data processing: Computer data processing (using Excel, at P = 0.05) involves a dispersion analysis. WSC data are characterized by: marginal values (min and max), arithmetic mean (x), standard deviation (SD) and coefficient of variation (CV,%). Variation is considered weak, moderate or strong at CV values, respectively: up to 10%; > 10-20%, and > 20% (Dimova and Marinkov, 1999). According to Fransis and Kannenberg (1978) with average WSC content and mean variation coefficient are selected for genotypes of each species exceeding the collectins average for the two consecutive years. As a major criterion for selection of elite genotypes, the mean arithmetic values of WSC content of forage dry mass,% and CV,% were used.

RESULTS AND DISCUSSION

It was found that WSC content of perennial grasses varied according to the plant species, genotype (variety or ecotype, ploidy level, group of maturity) and seasonal differences (regrowths and years), also growing conditions (fertilization, drought, cold, low and high

air temperatures etc. The highest WSC content was measured in perennial ryegrass, followed by fescues and the lowest in cocksfoot.

The average values for WSC in 2016 for perennial ryegrass are higher than in 2017 - 8.09% and 6.22%, respectively (Table 1 and Table 2). Significant variations in regrowths and genotypes are observed. In 2016, the first and fourth regrowth had the highest average content of WSC - 9.68% and 9.83%, and the third and the second with lower ones. - 7.12% and 6.15%. It is known that in drought the concentration of fructoses and other water-soluble carbohydrates in the perennial ryegrass decreases (Karsten and MacAdam, 2001), thus decreasing the osmotic potential and improving the water status of the plants as well as in the post-drought cutting, the WSC concentration in the biomass the next growth decreases by 50%, b.c. most of the synthesized WSC are used for growth after defoliation (Norris and Thomas, 1982). These facts could explain the low WSC values found in this study. The WSC concentration varies depending on the season and time of day. It increased from early March to 240 g kg⁻¹ DM in early May and declined sharply to 140-70 g kg⁻¹ DM in August (Smith and Elgersma, 2004). The maximum value for WSC is 14.5% and 14% for Sokolare and Harmoniya, respectively (Table 1). In 2017, the trend is similar - the first growth 9.38% and the second 8.15%, followed by fourth, second and third with the lowest WSC content. Lawson and Kelly (2014) and Katova (2005) reported such a trend. The maximum value for WSC is 12% for the new tetraploid Bulgarian varieties Tetrany and Tetramis (Table 2). Figure 1 shows the distribution of accessions for 2016 with WSC content and CV,%. In the first quadrant are the stable accessions with a high content of WSC - 7 and 5 of them are tetraploid, while in the second quadrant are the samples with a high content of WSC but

air temperatures etc. The highest WSC content was measured in perennial ryegrass, followed by fescues and the lowest in cocksfoot.

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with a high coefficient of variation, i. e. unstable - 2. A total of 9 accessions of perennial ryegrass are of interest to the selection. In figure 2, a total of 12 samples exceed the average for WSC for the 2017 for perennial ryegrass collection. The perennial ryegrass genotypes with increased WSC content above year average value for the collection during the two years of the study were selected: Meltador, Melverde, Meracoli, Melpetra, Harmoniya and Ravnogor (Figures 1 and 2). It is confirmed that tetraploid varieties have a higher WSC content in our study (Jung et al, 1996; Smith et al. 2001; Gilliland et al. 2002, Ghesquiere and Baert, 2014; Katova, 2016).

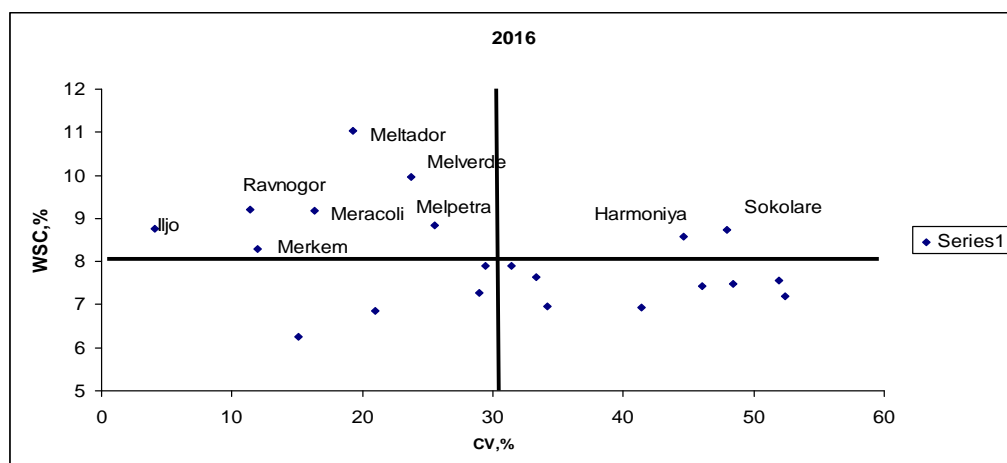
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1. , %

2016 .

Table 1. Water soluble carbohydrates content, % of dry matter of perennial ryegrass accessions in 2016

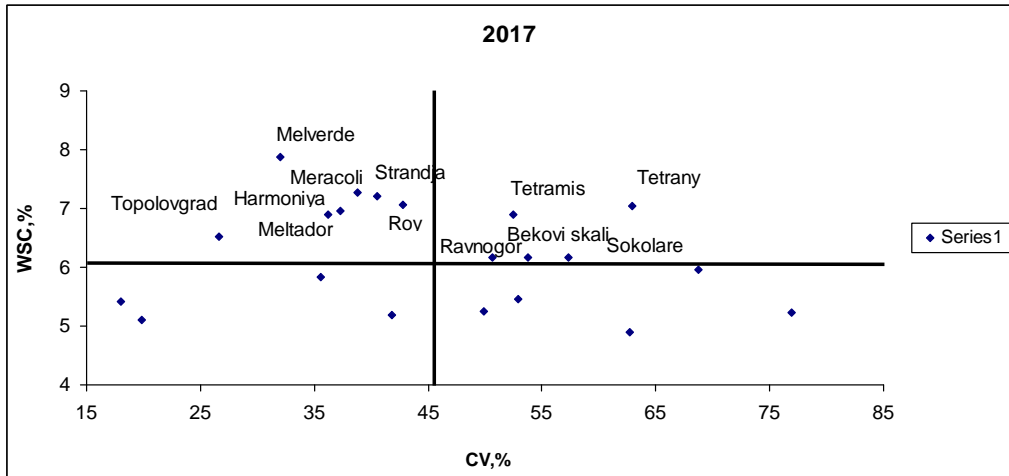
N	Accession	, % WSC content					CV, %
		I cut	II cut	III cut	IV cut	average	
1	Sokolare	14.5	5.6	5.7	9.1	8.73	47.90
2	Harmoniya	14	5.6	6.2	8.5	8.58	44.62
3	Tetramis	8.8	4.3	5.6	9.1	6.95	34.14
4	Strandja	9.7	4.4	4.5	9.1	6.93	41.42
5	Tetrany	10	3.4	5.5	11	7.48	48.42
6	Mara	11	4.5	4.5	9.7	7.43	46.05
7	Ravnogor	9.3	5.2	6.5	10.6	7.90	31.43
8	Topolovgrad	6.5	6.4	5.8	10.4	7.28	28.95
9	Merlinda	8.3	3.5	5	12	7.20	52.45
10	Roy	8.5	5.8	5.5	7.6	6.85	21.00
11	Meltador	12.5	8.6	12		11.03	19.23
12	Meracoli	9.6	7.5	10.4		9.17	16.34
13	Melpetra	9	6.5	11		8.83	25.52
14	Floris	5.8	7.5	10.4		7.90	29.44
15	Iljo	8.5	9			8.75	4.04
16	Bekovi skali	8	5	6.6	11	7.65	33.30
17	CHINA	12	6.2	4.5		7.57	51.97
31	Magura	7	5.2	6.6		6.27	15.08
32	Ravnogor	8.5	8.7	10.4		9.20	11.35
33	Melverde	12.7	8.6	8.6		9.97	23.75
34	Merkem	9	7.6			8.30	11.93
average		9.68	6.15	7.12	9.83	8.09	30.40
min		5.80	3.40	4.50	7.60	6.27	4.04
max		14.50	9.00	12.00	12.00	11.03	52.45
SD		2.34	1.74	2.49	1.29	1.15	14.65
CV, %		24.16	28.22	34.95	13.16	14.25	48.18
LSD 0.01		1.31	0.98	1.40	0.73	0.65	8.23



1.
2016 .
Fig. 1. Distribution of perennial ryegrass accessions according to WSC content in 2016

2. , %
2017 .
Table 2. Water soluble carbohydrates content, % of dry matter of perennial ryegrass accessions in 2017

N	accession	, % WSC content					Average	CV,%
		I cut	II cut	III cut	IV cut	V cut		
1	Sokolare	10.2	4.7	3.6			6.17	57.34
2	Harmoniya	10	7.5	4.4	4.4	10.1	7.28	38.84
3	Tetramis	12	7	2.9	4.1	8.5	6.90	52.46
4	Strandja	11.4	5.4	4.6	5.5	9.1	7.20	40.58
5	Tetrany	12	5.6	3.5			7.03	62.95
6	Mara	10.7	3.7	3.5			5.97	68.72
7	Ravnogor	9.6	5.4	3.5			6.17	50.62
8	Topolovgrad	8.8	7.7	5.2	4.6	6.3	6.52	26.64
9	Merlinda	9	5.7	4	4	6.5	5.84	35.52
10	Roy	11.5	4.5	4.5	6.1	8.7	7.06	42.75
11	Meltador	8.8	4	4.6	7.5	9.6	6.90	36.20
12	Meracoli	10.5	5.8	3.5	7	8	6.96	37.28
13	Melpetra	8.1	4.5	2.2	5	6.1	5.18	41.79
14	Floris	7.5	4.5	1	6.5	6.7	5.24	49.90
15	Iljo	9.5	4.7	1.5			5.23	76.94
16	Bekovi skali	8.8	4.8	1.7	5.5	10	6.16	53.78
17	CHINA	4.5	6.5	4.7	6		5.43	18.02
31	Magura	9	4.1	1.6	4.9		4.90	62.73
32	Ravnogor	8.8	3.6	4			5.47	52.93
33	Melverde	10.2	5.2	8.2			7.87	31.99
34	Merkem	6	5.3	4			5.10	19.90
average		9.38	5.25	3.65	5.47	8.15	6.22	45.61
min		4.50	3.60	1.00	4.00	6.10	4.90	18.02
max		12.00	7.70	8.20	7.50	10.10	7.87	76.94
SD		1.86	1.16	1.58	1.11	1.52	0.87	15.46
CV, %		19.83	22.09	43.32	20.23	18.67	14.00	33.89
LSD 0.01		1.05	0.65	0.89	0.62	0.85	0.49	8.69



2.
2017
Fig. 2. Distribution of perennial ryegrass accessions according to WSC content in 2017

2016 .
 2017
 . - 4,26% 4,04%,
 3 4).
 2016 . -
 (2,72%)
 (4,03%)
 (6,03%).
 8,40
 2017 .
 -
 6,68%),
 , ,
 (1,77%).
 10,5%
 .
 :
 (3 4).
 BGR 1120.
 (6,80%)
 BGR 1120

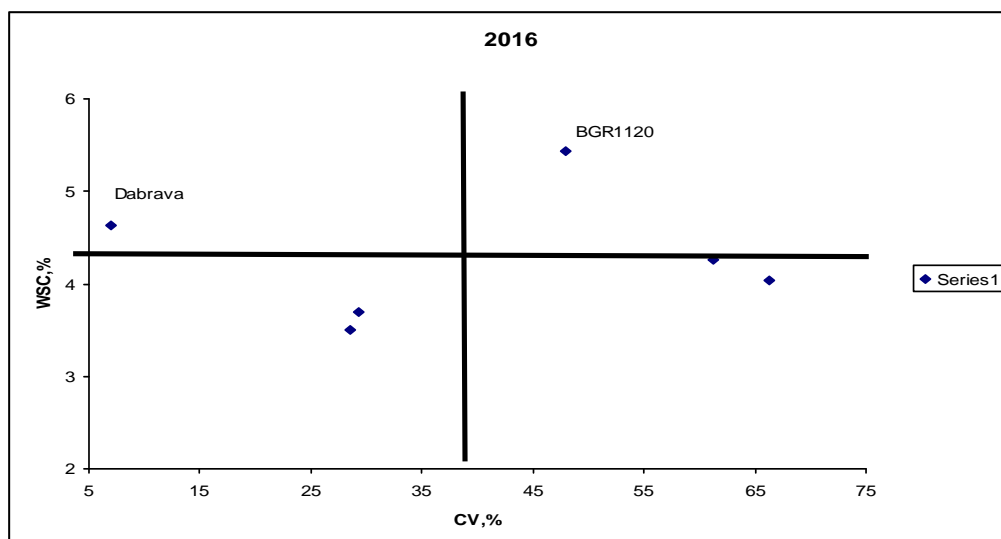
The average values for WSC content in 2016 for the cocksfoot are close to those of 2017 - 4.26% and 4.04%, respectively (Tables 3 and 4). There are differences in the WSC content of the regrowths, the lowest in the first growth (2.72%) in 2016 and increases approximately twice in the second (4.03%) and the third in the third regrowth 6.03%). The maximum value is 8.40% in third regrowth for BGR 1120. In 2017, first and sixth regowths have the highest WSC content (6.80% and 6.68%), decreasing significantly from second, fifth, third and lowest for fourth regrowth (1.77%). The maximum value is 10.5% for Bekovi Scali ecotype. Genotypes of cocksfoot with a higher content than the average annual value for the collection were selected during both study years: Dabrava and BGR 1120 (Figures 3 and 4).

3. , %

2016 .

Table 3. Water soluble carbohydrates content, % of dry matter of cocksfoot accessions in 2016

N	accession	, % WSC content				CV,%
		I cut	II cut	III cut	average	
25	Intensiv	1.5	4.6	6.7	4.27	61.31
26	Magda	1.8	3.3	7	4.03	66.36
27	Dabrava	4.5	4.4	5	4.63	6.94
28	Topolovgrad	2.5	4	4.6	3.70	29.23
29	BGR1120	3.5	4.4	8.4	5.43	48.01
30	Bekovi skali	2.5	3.5	4.5	3.50	28.57
average		2.72	4.03	6.03	4.26	40.07
min		1.50	3.30	4.50	3.50	6.94
max		4.50	4.60	8.40	5.43	66.36
SD		1.11	0.53	1.58	0.70	22.59
CV, %		41.02	13.18	26.16	16.46	56.39
LSD 0.01		1.17	0.56	1.66	0.74	23.76



. 3.

2016 .

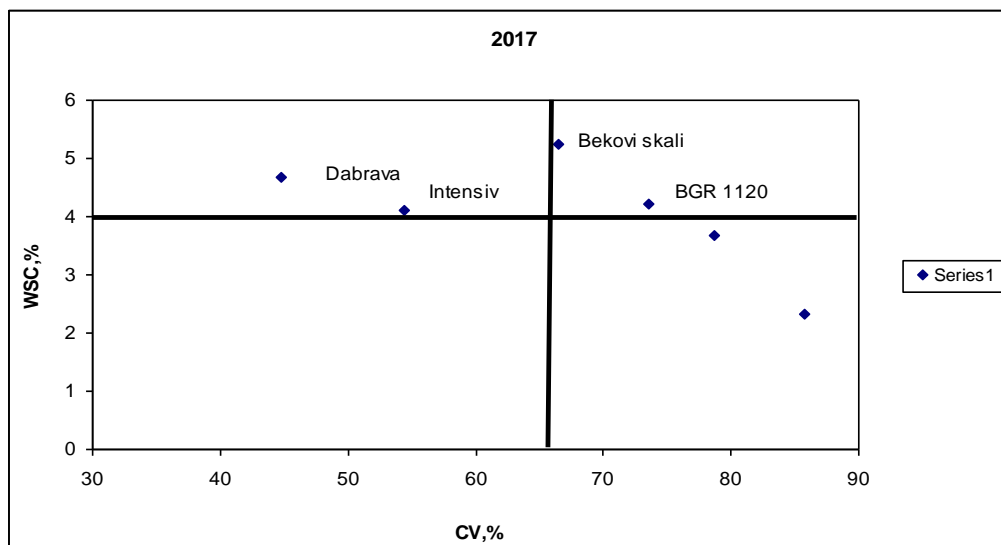
Fig. 3. Distribution of cocksfoot accessions according to WSC content in 2016

4. , %

2017 .

Table 4. Water soluble carbohydrates content, % of dry matter of cocksfoot accessions in 2017

N	accession	, % WSC content							CV,%
		I cut	II cut	III cut	IV cut	V cut	VI	average	
25	Intensiv	4.5	4.5	3.5	1.5	2.6	8	4.10	54.43
26	Magda	8	3.6	1.5	0.7	2	6.3	3.68	78.68
27	Dabrava	7.5	5.5	2.1	3.1	3.5	6.3	4.67	44.74
28	Topolovgrad	2.6	2	0.5	1.2	1.5	6.1	2.32	85.70
29	BGR1120	7.7	2	2.4	1.5	1.9		4.22	73.52
30	Bekovi skali	10.5	3.5	2.5	2.6			5.24	66.54
average		6.80	3.52	2.08	1.77	2.30	6.68	4.04	67.27
min		2.60	2.00	0.50	0.70	1.50	6.10	2.32	44.74
max		10.50	5.50	3.50	3.10	3.50	8.00	5.24	85.70
SD		2.81	1.38	1.01	0.90	0.78	0.89	1.00	15.38
CV, %		41.26	39.21	48.61	51.09	33.82	13.31	24.68	22.86
LSD 0,01		2.95	1.45	1.06	0.95	0.82	0.93	1.05	16.17



. 4.

2017 .

Fig. 4. Distribution of cocksfoot accessions according to WSC content in 2017

2016 . 7,08%,

2017

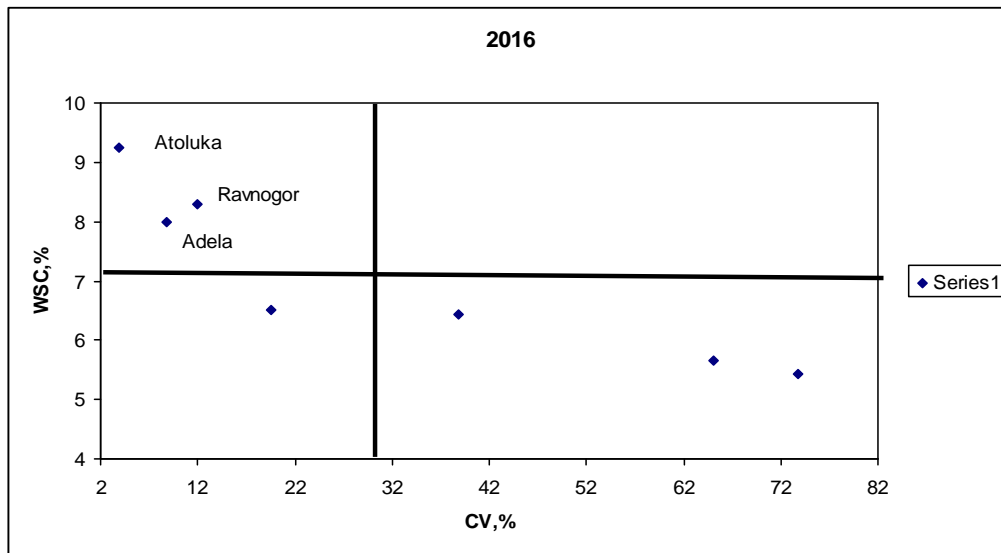
The WSC content in the fescue occupies an intermediate position between the perennial ryegrass and the cocksfoot and genotypes of reed and red fescue. The average annual value for

5,54% (5 6).
 2016 . -
 5,50%
 7,48% 8,33%.
 9,5%
 2017 . -
 (9,0%),
 (4,0%).
 10,7% Adela 10,5%
 Albena.
 Adela, Atoluka Ravnogor (5
 6).

2016 is 7.08%, while for 2017 it is 5.54% lower (Tables 5 and 6). By regrowths, the values for 2016 are varied, with the first growth having the lowest level of 5.50% and gradually increasing to a second 7.48% and a third 8.33%. The maximum value is 9.5% for the Atoluka ecotype. In 2017, the highest average was for the sixth regrowth (9.0%), followed by fifth, second, third, first and lowest in the fourth regrowth (4.0%). The maximum values are 10.7% for Adela and 10.5% for Albena. Genotypes of tall and red fescue with a higher content than the average annual value for WSC of the collection were selected: Adela, Atoluka : Ravnogor (Figures 5 and 6). In conclusion, despite great fluctuations in WSC due to environmental and plant developmental factors, there is genetic variation within and between forage grasses populations for this character which could be exploited by selection.

5. , % 2016 .
Table 5. Water soluble carbohydrates content, % of dry matter of fescue accessions in 2016

N	accession	, % WSC content				CV,%
		I cut	II cut	III cut	average	
18	F.ar. Albena	1	6.5	8.8	5.43	73.77
19	F.ar. Adela	7.5		8.5	8.00	8.84
20	F. ar. IRGR - Sadovo	1.5	8.5	7	5.67	65.04
21	F.pr. Transilvan	4	6.3	9	6.43	38.90
22	F.r. Capriora	7.4	5.6		6.50	19.58
23	F.r. Ravnogor	7.6	9		8.30	11.93
24	F.r. Atoluka	9.5	9		9.25	3.82
average		5.50	7.48	8.33	7.08	31.70
min		1.00	5.60	7.00	5.43	3.82
max		9.50	9.00	9.00	9.25	73.77
SD		3.33	1.52	0.91	1.44	28.20
CV, %		60.54	20.31	10.89	20.38	88.96
LSD 0.01		3.24	1.48	0.88	1.41	27.45



5. 2016 .
Fig. 5. Distribution of fescue accessions according to WSC content in 2016

6. , % 2017 .
Table 6. Water soluble carbohydrates content, % of dry matter of fescue accessions for 2017

N	accession	, % WSC content						average	CV, %
		I cut	II cut	III cut	IV cut	V cut	VI cut		
18	F.ar. Albena	7	10.5	5.4	2.5	6	8.5	6.65	41.22
19	F.ar. Adela	4.5	10.7	6.5	6	7	10.5	7.53	33.44
20	F. ar. IRGR - Sadovo	6	5.3	3.5	2.6	4.6	8	5.00	38.22
21	F. pr. Transilvan	3.3	4.5	5.9	4	10.1		5.56	48.75
22	F. r. Capriora	3.2	5.5	5.9	4.5			4.78	25.21
23	F. r. Ravnogor	1.8	3.6	3.5	2.6	4.8		3.26	34.68
24	F.r. Atoluka	4.6	6.3	6	5.4	7.7		6.00	19.18
7	F.pr. Merifest	5.5	6.5	2.5	4.4	9		5.58	43.37
average		4.49	6.61	4.90	4.00	7.03	9.00	5.54	35.51
min		1.80	3.60	2.50	2.50	4.60	8.00	3.26	19.18
max		7.00	10.70	6.50	6.00	10.10	10.50	7.53	48.75
SD		1.69	2.63	1.50	1.34	2.07	1.32	1.28	9.67
CV. %		37.60	39.78	30.56	33.46	29.44	14.70	23.11	27.24
LSD 0.01		1.54	2.40	1.36	1.22	1.88	1.20	1.17	8.81

: Meltador,
 Melverde, Meracoli, Melpetra,
 .
 ➤
 2016 .
 2017
 . - 4,26% 4,04%,
 ,
 2016 .
 -
 (2,72%)
 (4,03%)
 (6,03%). 2017 .
 (6,80%
 6,68%),
 ,
 (1,77%).
 -
 : BGR 1120.
 ➤
 2016 . 7,08%, 2017 . -
 5,54%.
 2016 . ,
 - 5,50%
 7,48%
 8,33%. 2017 . -
 (9,0%),
 , , ,
 (4,0%).
 -
 : Adela, Atoluka
 Ravnogor.

- above year average value for the
 - collection during the two years of the
 study were selected: Meltador, Melverde,
 Meracoli, Melpetra, Harmoniya and
 Ravnogor. Tetraploid varieties have a
 higher WSC content.

➤ The average values for
 WSC content in 2016 for the cocksfoot
 are close to those of 2017 - 4.26% and
 4.04%, respectively. There are differences
 in the WSC content of the regrowths, the
 lowest in the first growth (2.72%) in 2016
 and increases approximately twice in the
 second (4.03%) and the third in the third
 regrowth 6.03%). The maximum value is
 8,40% in third regrowth for BGR 1120. In
 2017, first and sixth regrowths have the
 highest WSC content (6,80% and 6,68%),
 decreasing significantly from second, fifth,
 third and lowest for fourth regrowth
 (1.77%). Genotypes of cocksfoot with a
 higher content than the average annual
 value for the collection were selected
 during both study years: Dabrava and
 BGR 1120

➤ The WSC content in the
 fescue occupies an intermediate position
 between the perennial ryegrass and the
 cocksfoot and genotypes of reed and red
 fescue. The average annual value for
 2016 is 7.08%, while for 2017 it is 5.54%
 lower. By regrowths, the values for 2016
 are varied, with the first growth having the
 lowest level of 5.50% and gradually
 increasing to a second 7.48% and a third
 8.33%. In 2017, the highest average was
 for the sixth regrowth (9.0%), followed by
 fifth, second, third, first and lowest in the
 fourth regrowth (4.0%). Genotypes of tall
 and red fescue with a higher content than
 the average annual value for WSC of the
 collection were selected: Adela, Atoluka
 Ravnogor.

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Comparative estimation of alfalfa cultivars regarding some main biological parameters

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SUMMARY

With aim, a comparative characteristic of eight alfalfa cultivars (Europe, Prista 2, Prista 3, Prista 4, Obnova 10, Pleven 6, Dara, Multifoliate.), an estimation of main biological parameters (stand formation, overwintering and factors determining it, accumulation of plant residues and corresponding amounts of N, P and Ca in the soil) was conducted. For carrying out experimental activity, a randomized block method was used, at a row spacing of 11.5 cm and a sowing rate of 2.5 kg da⁻¹. The analysis of variance showed a statistically significant influence of the year and cultivar on the stand formation, expressed as stems m⁻² (plants m⁻² × stems plant⁻¹). The parameter was dynamic and during the particular years, different cultivars demonstrated a higher density. For the 4-year period of alfalfa development was established a general trend of decrease in stand density, with exception of a slight increase in the 3rd year determinative by the variation in the components "plants m⁻²" and "stems plant⁻¹" and the compensatory mechanism

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m⁻²,
 4th
 52.6%
 ().
 25.0 ()
 69.4%
 10
 4 (88.1 - 88.5%).
 (r = 0.750),
 (r = 0.696),
 (r = 0.352),
 4-
 ()
 (0-30 cm)
 10.7 t ha⁻¹,
 225.1 kg N ha⁻¹, 15.4 kg P ha⁻¹ 77.1
 kg Ca ha⁻¹.
 4, 3 6.
 :
 ,
 ,
 ,
 (Radovi et al.,
 2009).
 (Dini et al., 2005,
 Markovi et al., 2007),

between them.

At the end of the 4th experimental year, the number of stems per unit area was reduced by 52.6% on average compared to the 1st year, with a variation of 25.0 (Obnova) to 69.4% (Dara). The evaluation of cultivars regarding sensitivity to unfavorable conditions during the winter period determined a higher percentage of overwintering plants in Multifoliata, Obnova 10 and Prista 4 (88.1 - 88.5%). Overwintering percentage correlated positively with the morphological parameters of root crown diameter (r = 0.750) and root weight plant-1 (r = 0.696) and soluble sugars content in the root mass (r = 0.352), established in the autumn regrowth of stands. At the end of the 4-year period of alfalfa cultivation and after harvesting of the last (autumn) regrowth, accumulated plant residues in the soil layer 0-30 cm were on average 10.7 t ha⁻¹, providing 225.1 kg N ha⁻¹, 15.4 kg P ha⁻¹ and 77.1 kg Ca ha⁻¹, respectively. Cultivars demonstrating higher values in this regard were Prista 4, Dara, Prista 3 and Pleven 6.

Key words: alfalfa, cultivars, stand, overwintering, plant residues, nitrogen

INTRODUCTION

Perennial legumes have an important role in ensuring livestock farming with cheap fodders with high nutritional value and digestibility. Among them, alfalfa is the most widely known and spread worldwide. Due to its rich and variable genetic base, it has a good adaptability to different environmental conditions and a wide range of cultivation (Radovi et al., 2009).

Alfalfa biomass is characterized with a high content of crude protein (Dini et al., 2005, Markovi et al., 2007), vital vitamins and microelements, and is well balanced with respect to amino acids that are

essential for normal growth and development of the animals (Markovi et al., 2007).

(Markovi et al., 2007).

(Bagavathiannan and Van Acker, 2009).

(Karimi et al., 2009),

(Nicolis et al., 2008; Manpong et al., 2009; Oskueian et al., 2011).

Radovi et al. (2009)

(Annicchiarico et al., 2006).

(Radovi et al., 1996).

(Lamb et al., 2005), (Volenec et al., 1987),

(Stanisavljevi et al., 2008).

- essential for normal growth and development of the animals (Markovi et al., 2007).

- Alfalfa is also one of the most famous medicinal plants (Bagavathiannan and Van Acker, 2009). In recent years, an increased interest and using in the agri-food (in the form of sprouts, dehydrated leaves, tea, food supplements) and pharmaceutical industry (Karimi et al., 2009) have been reported, which is determined by the high content of phenols and flavonoids (such as pyrogallol, gallic acid, naringin and quercetin) having antioxidant and anti-inflammatory properties (Nicolis et al., 2008; Manpong et al., 2009; Oskueian et al., 2011).

- According to Radovi et al. (2009), the practical importance of alfalfa is not limited to its valuable forage qualities for livestock production. It also performs other important economic and biological functions: it enriches the soil with nitrogen, it is a good predecessor for many crops, suitable for green manure, improves the physical properties and reduces the salinity of the soil etc.

- Alfalfa is one of the few crops that provide considerable amounts of biomass with minimal inputs. Thereby, beside an important role in traditional livestock farming, it has a meaning for sustainable agriculture and organic production (Annicchiarico et al., 2006). The high and stable yield of the culture is determined by a number of morphological and biological characteristics (Radovi et al., 1996). According to other authors, decisive in this respect are the stand density (Lamb et al., 2005), stem formation (Volenec et al., 1987), longevity, cultivar composition and others (Stanisavljevi et al., 2008).

- The purpose of the study is to compare alfalfa cultivars with respect to

()
 %
 2006-2009
 ()
 8 (*Medicago sativa* L.):
 2, 3, 4, 10, 6,
 11.5 cm
 2.5 kg da⁻¹, 3-
 20/200/30
 cm (/ /)
 (number of stems m⁻²)
 (m⁻² x ⁻¹) %
 (t ha⁻¹)
 4
 (g kg DM⁻¹) (OMA, 1990).
 (Dimova and
 Marinkov, 1999),
 GENES 2009.7.0 (Cruz, 2009).

some main biological characteristics (stand formation and % of overwintering plants in different years of cultivation, quantity and chemical composition of autumn regrowth in the last experimental year).

MATERIAL AND METHODS

The field experiment was carried out during the period 2006-2009 at the Institute of forage crops (Pleven) on a soil type of slightly leached chernozem and non-irrigating conditions of cultivation. Objects of the survey were 8 alfalfa (*Medicago sativa* L.) cultivars: Europe, Prista 2, Prista 3, Prista 4, Obnova 10, Pleven 6, Dara and Multifoliolate. Sowing was conducted by the randomized block method, at a row spacing of 11.5 cm and a sowing rate of 2.5 kg da⁻¹, in a 3-fold replication of the variants. The formation of alfalfa stands in different cultivars was established at harvesting of spring, summer and autumn regrowths (at early flowering). Soil monoliths with dimensions 20/200/30 cm (width / length / depth) were taken, and stems formed per unit area (number of stems m⁻²) (plants m⁻² x shoots plant⁻¹) and % of overwintering plants were calculated. The amount of plant residues (t ha⁻¹) was determined after harvesting the fall regrowth during the 4th experimental year and the nitrogen, phosphorus and calcium content were calculated (OMA, 1990).

The experimental data were processed by using Path coefficient analysis (Singh and Chaudhary, 1979), correlation and variance analysis (Dimova and Marinkov, 1999), software products MS Excel (2003) and GENES 2009.7.0 (Cruz, 2009).

RESULTS AND DISCUSSION

The four-year experimental period was characterized by an average daily temperature and a sum of rainfalls for the period of active vegetation (March-September) in alfalfa of 18.2 ° C and 320

18.2 °C, 320 mm, 2006-2009, 17.8 °C, 384 mm, 17.6 (2006), 19.2 °C (2007), 283 (2007), 351 mm (2006), 1843, (VC) 23%, 6, 3, 2, (VC = 14%), m² 1188, (32.6%), 4, 874 m⁻² (29.3%), (VC=10%), 4,

mm, respectively. These values determined the vegetation period during 2006-2009 as relatively warmer and dry compared to the same for the previous 20-year period, whose values were 17.8 °C and 384 mm. The variation in particular years in the meteorological conditions was considerable, in limits from 17.6 (2006) to 19.2 °C (2007) regarding temperature, and from 283 (2007) to 351 mm (2006) for rainfalls.

The stand formation in different alfalfa cultivars by years and regrowths is presented in Table 1. The average number of stems per unit area (m²) in the year of stand establishment was 1843, with a value of the variation coefficient (VC) of 23%, laying down the fluctuation of the studied parameter as strong. Three cultivars were distinguished with a significantly higher stand density – Dara, Pleven 6 and Prista. 3. In comparison with autumn growth, during the summer and spring regrowth of the 1st experimental year was established a greater number of stems formed per unit area but this trend was not a one-way in the subsequent years. In the 2nd year of alfalfa development, the variation in stand density was considerably less pronounced (VC = 14%). The average number of stems harvested per m² was 1188, and cultivar Europe noted a significant excess (32.6%) above this value compared to all other cultivars. A similar trend was observed in the 4th experimental year, when at an average density of 874 stems m⁻², a statistically significant difference (with 29.3%) compared to the others was demonstrated only by Multifoliate. The 3rd year of alfalfa development was characterized by the least variation in stand density (VC = 10%) as Dara formed the largest number of stems, but with nonsignificant differences to Prista 4, Europe and Multifoliate.

1. (number of stems m⁻²)

Table 1. Stem formation (number of stems m⁻²) in alfalfa cultivars

Cultivar	SpR	SumR	AR	\bar{x}	SpR	SumR	AR	\bar{x}
1 st experimental year				2 nd experimental year				
1	1865	1890	1197	1651 b	1371	2064	1291	1575 d
2	1876	1576	1567	1673 b	1205	1465	1088	1253 a
3	1938	2705	1485	2043 c	1004	1168	1031	1068 ab
4	1871	1446	1752	1690 b	1259	836	1383	1160 ab
5	950	1658	1021	1210 a	981	913	1262	1052 b
6	2092	2797	1946	2278 d	977	1331	1318	1209 ab
7	2353	2997	2262	2537 e	1167	1183	890	1080 a
8	1585	2399	998	1661 b	1075	1118	1124	1106 c
\bar{x}	1816	2184	1529	1843 b	1130	1260	1174	1188
r ₁				0.871				0.784
r ₂				0.141				0.541
3 rd experimental year				4 th experimental year				
1	1145	1493	1441	1360 bc	729	1086	928	914 b
2	1008	1557	1240	1268 ab	567	595	1164	775 a
3	1213	1613	1119	1315 ab	635	572	1314	840 ab
4	1291	1482	1315	1363 bc	510	739	1309	853 ab
5	1094	1065	1133	1097 a	1058	510	1153	907 b
6	859	1398	1588	1282 ab	938	800	650	796 ab
7	947	1612	2120	1559 c	605	1042	686	777 a
8	1047	1714	1270	1344 bc	1228	702	1461	1130 c
\bar{x}	1076	1492	1403	1323	784	756	1083	874
r ₁				0.472				0.346
r ₂				0.605				0.840

* significance at p < 0.05

1-Europe, 2-Prista 2, 3-Prista 3, 4-Prista 4, 5-Obnova 10, 6-Pleven 6, 7-Dara, 8-Multifoliate

SpR-spring regrowth, SumR-summer regrowth, AR-autumn regrowth

r₁ - correlation between stems per m² and plants per m²; r₂ - correlation between stems per m² and stems per plant¹

4-
(m⁻² x m⁻²),
11.4%).
3rd year (11.4%).
" " " " " "
Volenec et al. (1987) Lloveras et al. (2008).

The analysis of the data obtained during the 4-year period of alfalfa development showed a general downward trend in density, expressed as stems m⁻² (plants m⁻² x stems plant⁻¹), with the exception of a slight increase in the 3rd year (average with 11.4%). The reason for this was the fluctuation in the components "plants m⁻²" and "stems plant⁻¹" and the compensatory mechanism between them over time, detailed by Volenec et al. (1987) and Lloveras et al. (2008).

According to the authors, because of

"aging" of the alfalfa stand, the participation of "plant per unit area" and "stems per plant" as components in the stand formation was changing dynamically. Volenec et al. (1987) observed a decrease in crop density during the first 2 to 3 years after sowing, due to high intraspecific competition. Then a slight increase in population density due to a compensatory increase in the number of stems plant⁻¹ was followed. The effect was short-term, after which the decrease in density continued, as the root crown and the formation of stems plant⁻¹ were no longer able to compensate for the losses resulting from the reduction in the number of plants per unit area.

The correlation analysis of the data in this experiment confirmed the above-mentioned dependencies: in the first two years of alfalfa development, the plants per unit area had a determinant role in the stand formation (r_1 was 0.871 and 0.784, respectively) and in the 3rd and 4th year - the stems plant⁻¹ (r_2 was 0.605 and 0.840, respectively). As a terminal result, at the end of the 4th year of alfalfa development, the number of stems per unit area was reduced by 52.6% on average compared to the 1st year, with a variation of 25.0 (Obnova) to 69.4% (Dara).

The analysis of variance showed a statistically significant influence of the year and cultivar on the stand formation (stems m⁻²) in alfalfa (Table 2). With 68.4% of the total variation, the impact of the year (factor A) can be determined as the dominant one. It was conditioned by the essential differences in weather conditions over the four years and by the unequal response of the cultivars to the change in environmental conditions. The action of the cultivar (factor B), as well as the interaction year x cultivar (A x B) was less expressed, respectively by 7.6 and 21.3%.

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2.

Table 2. Analysis of variance for stand formation in alfalfa cultivars

Source of variation	Degrees of freedom	Sum of squares	Mean square	Influence of factors
/ Total	95	1715493		100.0
Factor A- Year ^a	3	1173127	391039*	68.4
Factor B – cultivar	7	130273	186104*	7.6
A x B	21	365256	173931*	21.3
Residual	64	46837	7318	2.7

* significance at $p < 0.05$

Winter resistance is defined as the ability of plants to survive the entire complex of unfavourable conditions during the winter period. As a perennial culture, the successful wintering of alfalfa has an important role in maintaining high stand density, productivity and longevity.

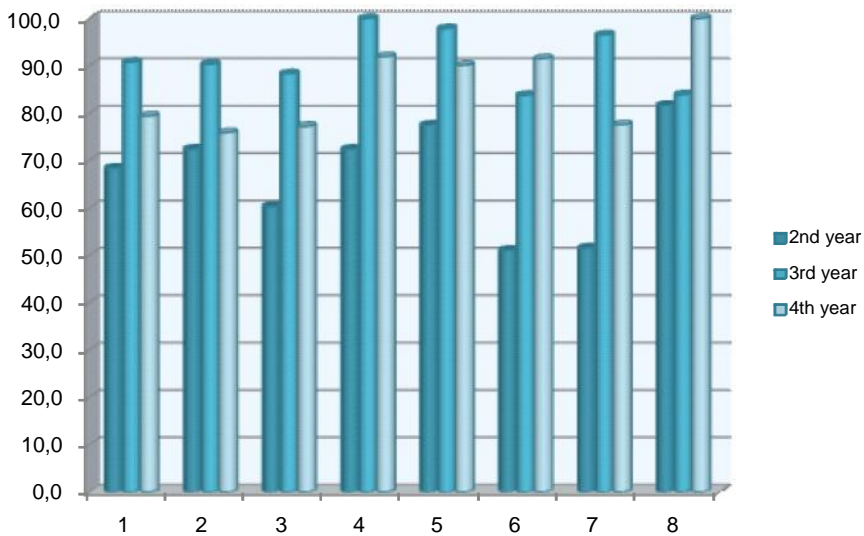
The evaluation of the eight studied cultivars showed well-pronounced trends over the experimental years (Figure 1). In the spring of the 2nd experimental year, the percentage of overwintering plants varied over a wide range (51.1-81.7%), with higher values were demonstrated by Multifoliate, Obnova 10 and Prista 4. Similar trends were also observed during the subsequent years, but with considerably less variation of the parameter.

As a whole, alfalfa exhibited a higher susceptibility to unfavorable winter conditions in the 2nd year (on average 70.0% overwintering plants) than in the 3rd and 4th years (respectively 91.4 and 85.4%). On average, for the investigated period, with higher percentages of overwintering plants were Multifoliate, Obnova 10 and Prista 4 (88.1-88.5%), followed by Europe and Prista 2 (79.5%). Relatively low was the percentage of survived plants in Dara, Pleven 6 and Prista 3 (75.2-75.4%).

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1-Europe, 2-Prista 2, 3-Prista 3, 4-Prista 4, 5-Obnova 10, 6-Pleven 6, 7-Dara, 8-Multifoliata

Fig. 1. Overwintering (%) of plants in alfalfa cultivars by years

(Jung and Larson, 1972; Avice et al., 1997; Dhont et al., 2004; Hakl et al., 2007).

(TNC)

(SS),

15% (Hendershot and Volenec, 1993),

(Jung and Larson, 1972).

Winter- and cold-resistance in alfalfa are associated with a number of physiological and chemical indicators that allow plants to overcome low-temperature stress (Jung and Larson, 1972; Avice et al., 1997; Dhont et al., 2004; Hakl et al., 2007). The physiological status of plants, especially the content of non-structural carbohydrates (TNC) and soluble sugars (SS) accumulated in the root crown and roots can limit the tolerance, especially when they are reduced to critical levels to about 15% of the dry weight.

It has been found that soluble proteins (Hendershot and Volenec, 1993), as well as reduced water absorption, depending on the root system size and root morphology, also have relevance to the considered parameter (Jung and Larson, 1972).

In the present study, correlations with a high positive value were established between the survival percentages in

(r = 0.750,
 (r = 0.696,
 3).
 (r = 0.352),
 Jung and Larson (1972)
 15%

different cultivars and the morphological parameters of root crown diameter (r = 0.750, average for the period) and root weight plant⁻¹ (r = 0.696, average for the period) recorded in autumn regrowths of stands (Table 3). A low value had the correlation with respect to the soluble sugars content in the root mass (r = 0.352), which can be explained by the fact that their average content in the studied varieties over the whole period did not exceed the critical value of 15% stated by Jung and Larson (1972) for sugar content.

3.

%

Table 2. Correlative dependences between %overwintering plants and main parameters of root system in alfalfa

/ Parameters	% / % overwintering plants			
	2 nd year	3 rd year	4 th year	average
Diameter of root crown	0.467	0.462	0.517	0.750
Root mass weight	0.231	0.399	0.446	0.696
Content of soluble sugars	0.246	0.361	0.155	0.352

(Lesznyák et al., 2008).
 ù
 120 (8-12 t/ha)
 40-60 t
 (Ivanov, 1988).
 4-
 ()
 0-30 cm 10.7 t ha⁻¹,

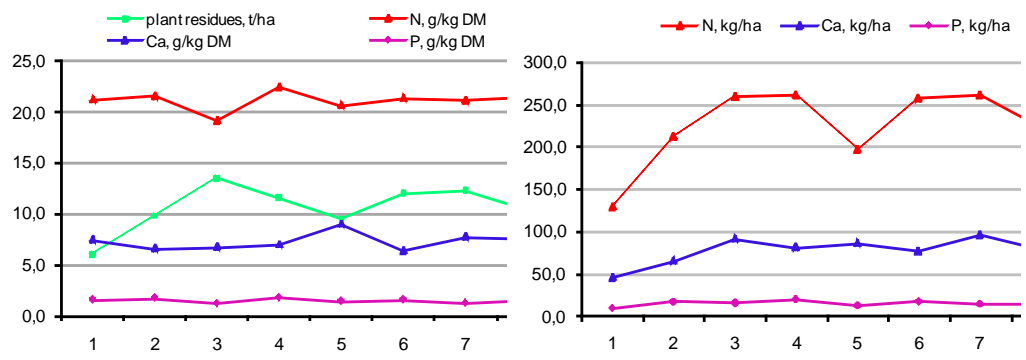
Leguminous crops, including the alfalfa, play an important role in improving nutrition regime of soil, which is of particular importance for the organic production system (Lesznyák et al., 2008). Its well-developed root during the second to the third year of development forms from 80 to 120 centners per hectare (8-12 t/ha) root mass and plant residues in the arable soil layer which is equivalent to 40-60 t fertilizer regarding the content of nitrogen, phosphorus, potassium and other elements (Ivanov, 1988).
 Our data showed that at the end of the 4-year period of alfalfa cultivation and after harvesting of the last (autumn) regrowth, accumulated plant residues in the soil layer 0-30 cm were on average 10.7 t ha⁻¹, whose nutrient content of N, P and Ca

N, P Ca 21.15, 7.28
 1.47 mg kgDM⁻¹ (2).

Kostov Hristozov (1990),
 2% N, 0.7% P₂O₅
 1.3% CaO.

1 t ,
 kg , 7 kg , 13 kg 20

were respectively 21.15, 7.28 and 1.47 mg kgDM⁻¹ (Figure 2). These values were similar to those reported by Kostov and Hristozov (1990), according to which 2% N, 0.7% P₂O₅ and 1.3% CaO were contained in the dried roots of alfalfa. According to the authors, at an average amount of root residues of 1 t per hectare, it enriched the soil with 20 kg of nitrogen, 7 kg of phosphorus and 13 kg of calcium in the active substance, which was more than an average rate of fertilization in the country.



a) b)
 1-Europe, 2-Prista 2,3-Prista 3, 4-Prista 4, 5-Obnova 10, 6-Pleven 6, 7-Dara, 8-Multifoliolate

. 2.) (t ha⁻¹) N, P, Ca (g kgDM⁻¹)
 ;)
 (kg ha⁻¹) N, P Ca

Fig. 2. a) Amount (t ha⁻¹) and content of N, P, Ca (g kgDM⁻¹) of plant residues after 4-year cultivation of alfalfa cultivars; b)Accumulation of N, P and Ca with plant residues per unit area (kg ha⁻¹)

(VC = 21%),
 3 (13.5 t ha⁻¹),
 6
 4, 2 6
 (N: 21.38-22.51 mg kgDM⁻¹; P: 1.51-1.73 mg kgDM⁻¹; Ca: 6.33-8.99 mg kgDM⁻¹),

The variation in the amount of plant residues among the studied varieties was essential (VC = 21%), with higher values being established for Prista 3 (13.5 t ha⁻¹), followed by Pleven 6 and Dara. With a more favorable biochemical composition was the root biomass of Prista 4, Prista 2 and Pleven 6 (N: 21.38-22.51 mg kgDM⁻¹, P: 1.51-1.73 mg kgDM⁻¹, Ca: 6.33-8.99 mg kgDM⁻¹) but as a whole, the variation in the qualitative composition was

10.7 t ha⁻¹,
 225.1 kg N ha⁻¹, 15.4 kg P ha⁻¹
 77.1 kg Ca ha⁻¹.

16.6%
 N, 3
 24.3%
 4
 6 –
 21.0% Ca.

Lesznyák et al. (2008) Bilalis (2015),
 Bilalis (2015)
 Onward, Andros Amorgos.
 , Torricelli (2006)

considerably less pronounced compared with the quantitative one. As sum effect, at an established amount of plant residue of 10.7 t ha⁻¹ (on average), alfalfa accumulated 225.1 kg N ha⁻¹, 15.4 kg P ha⁻¹ and 77.1 kg Ca ha⁻¹, respectively. Cultivars demonstrating higher values in this respect were as follows: Prista 4 and Dara - an excess of 16% compared to the average for the group in terms of the accumulated N, Prista 3 and Dara – an excess with 24.3% for P, and Prista 4 and Pleven 6 – an excess with 21.0% for Ca.

The results confirmed the findings of Lesznyák et al. (2008) and Bilalis (2015), according to which the soil properties were positively influenced after pea cultivation, but this effect correlated directly with different cultivars. Bilalis (2015) found a significant difference among the cultivars in terms of total nitrogen content in the soil and recommended the cultivation of the Onward, Andros and Amorgos varieties.

In addition, Torricelli (2006) noted that the mineral substances in the underground organs and plant residues of alfalfa were available for subsequent crops after degradation of the phytomass.

The inclusion of alfalfa in field crop rotation not only improved soil fertility but also increased the productivity of subsequent crops, which was particularly important for organic farming conditions.

CONCLUSIONS

The analysis of variance showed a statistically significant influence of the year and cultivar on the stand formation, expressed as stems m⁻² (plants m⁻² × stems plant⁻¹), in the eight alfalfa cultivars studied. The parameter was dynamic and during the particular years, different cultivars demonstrated a higher density: 1st year – Dara, Pleven 6, Prista 3; 2nd year – Europe; 3rd year – Dara, Prista 4,

A
 (m⁻² ×
 m⁻²)
 : 1st
 3, 6

2nd - 3rd - 4th ,
 4, - 4-
 3rd , m⁻²
 „ -1, „
 52.6% 1st ,
 25.0 () 69.4% ().
 10 4 (88.1-88.5%).
 (r = 0.750),
⁻¹ (r = 0.696),
 (r = 0.352),
 4-
 () ,
 0-30 cm 10.7 t ha⁻¹,
 225.1 kg N ha⁻¹,
 15.4 kg P ha⁻¹ 77.1 kg Ca ha⁻¹.
 , -
 , 3 6.

Europe, Multifoliate; 4th – Multifoliate.

For the 4-year period of alfalfa development was established a general trend of decrease in stand density, with exception of a slight increase in the 3rd year determinative by the variation in the components "plants m⁻²" and "stems plant⁻¹" and the compensatory mechanism between them. At the end of the 4th experimental year, the number of stems per unit area was reduced by 52.6% on average compared to the 1st year, with a variation of 25.0 (Obnova) to 69.4% (Dara).

The evaluation of cultivars regarding sensitivity to unfavorable conditions during the winter period determined a higher percentage of overwintering plants in Multifoliate, Obnova 10 and Prista 4 (88.1-88.5%). Overwintering percentage correlated positively with the morphological parameters of root crown diameter (r = 0.750) and root weight plant⁻¹ (r = 0.696) and soluble sugars content in the root mass (r = 0.352), recorded in the autumn regrowths of stands.

At the end of the 4-year period of alfalfa cultivation and after harvesting of the last (autumn) regrowth, accumulated plant residues in the soil layer 0-30 cm were on average 10.7 t ha⁻¹, providing 225.1 kg N ha⁻¹, 15.4 kg P ha⁻¹ and 77.1 kg Ca ha⁻¹, respectively. Cultivars demonstrating higher values in this regard were Prista 4, Dara, Prista 3 and Pleven 6.

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„Lebosol®- Total Care” (Medicago sativa)

*,
„”, 7007 ,

Influence of the Lebosol®-Total Care leaf fertilizer on the resistance of young lucerne (Medicago sativa) to leaf pathogens

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SUMMARY

2014 .
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4
“Lebosol®-Total Care”
(
Total Care”
,
,
10-15 cm
, 250 ml/da.

The survey was conducted in 2014 with the specific soil and meteorological conditions of the experimental field of the Institute of Agriculture and Seed Science "Obraztsov Chiflik" - Rousse. The Bulgarian lucerne varieties: Prista 3, Prista 4, Prista 5, Multilist 1 and Roli, created in the Institute, and the French variety – Europe.

The experience is based on the blocking method in 4 iterations and includes variants with the introduction of the “Lebosol® Total Care” product and controls (untreated variants). "Lebosol®-Total Care" is a combination of amino acids, macro and trace elements, with anti-stress and nourishing action. It stimulates photosynthesis and increases the immune protection of plants. Foliar fertilization is carried out in each subplot, twice at plant height of 10 -15 cm and in phaseisation phase at a dose of 250 ml / da. The phytopathological evaluation has been carried out according to the methods, generally accepted for our

country, phytopathological methods.

The aim is to determine the effect of the product on the stability of young lucerne crops on leaf pathogens.

It was found that "Lebosol®-Total Care" leaf fertilization had a positive effect on the resistance of the tested varieties to *Pseudopeziza jonesii* Nannf, *Pseudopeziza medicaginis* and *Uromyces striatus* in all subspecies during the first year of cultivation.

In the first vegetation of crops treated with the product, the attack rate from leaf pathogens decreased from 51.00% to 36,3% for *Pseudopeziza jonesii* Nannf, 49,3% to 35,5% for *Pseudopeziza medicaginis* and from 65,4% to 41% for rust (*Uromyces striatus*).

The "Lebosol® Total Care" product has the strongest positive effect on the resistance of leaf pathogens to Prista 3 crops.

Treated young lucerne crops are characterized by a lower attack index than phytopathogens compared to untreated variants.

Key words: alfalfa, resistance, "Lebosol®-Total Care", index of attack

country, phytopathological methods.

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INTRODUCTION

Lucerne (*Medicago sativa* L.), is the most important grass-fodder plant in Bulgaria and in many temperate climates (arinova et al., 2009; Pachev, 2014). Its importance is due to the high yield of green fodder, protein content and high nutritional value (Huyghe, 2003). It is known for its ability to improve soil structure and is an effective source of biological nitrogen. (Peoples et al., 1995; Vasileva, 2010). It has a life span of up to 5 years, but in some areas of the world the fields may remain productive longer (Summers, 1998).

(Hwang, 2006).

(*Pseudopeziza medicaginis* Lib.),
(*Pseudopeziza jonesii* Nannf.)
(*Uromyces striatus* Schroet.),
(Blajev, 1982; Nikolova, 1990; Ivanova, 2006).
(*Pseudopeziza jonesii* Nannf.),
Ascomycetes,
(Ivanova, 2014).
(*Pseudopeziza medicaginis*),
Ascomycetes,
(*Uromyces striatus* Schroter.,
Basidiomycetes (Ivanova and
Marinova, 2014).

One of the factors to reduce yields and crude protein content (Hwang, 2006) is the attack of leaf pathogens.

The greatest damage that leaf diseases cause is permeation, followed by defoliation.

In our country, the most common leaf pathogens are *Pseudopeziza medicaginis*, *Pseudopeziza jonesii* Nannf., and *Uromyces striatus*, which have been studied to one degree or another (Blajev, 1982; Nikolova 1990; Ivanova, 2006).

Pseudopeziza jonesii Nannf., Class *Ascomycetes*, are observed in all alfalfa substrates until the end of the vegetation. Leaves and stems form oblong, yellow to orange or dark brown V-shaped stains (Ivanova, 2014).

Pseudopeziza medicaginis, class *Ascomycetes*, are characterized by spot-shaped, cut-off black spots that rarely merge.

Another disease that is observed annually on alfalfa is rust *Uromyces striatus* Schroter., Class *Basidiomycetes* (Ivanova and Marinova, 2014). After the plant disease develops, brown uredosporos are formed with a chlorotic wreath around them.

As a result of the patching of the pathogens mentioned, there is an early leafpad, a change in the nutritional value of the feed, a reduction in its quantity and quality.

Changes in plant production, as well as market prices of material production costs, require economic rethinking of agricultural production. In a market economy, competition forces each manufacturer to seek new technological solutions to increase productivity and lower costs to achieve greater returns and production efficiencies (Sabev et al., 2015).

2015).	(Sabev et al.,	Recently many researchers have studied
(Petkova et al., 2007; Sabev et al., 2015)	-	the effects of leaf fertilizers (Petkova et al., 2007, Sabev et al., 2015) in order to
	-	stimulate plant growth and increase their
2015; Ivanova et al., 2016).	(Sabev et al.,	resistance and adaptability to unfavorable
	-	environmental conditions (Sabev et al.,
e	-	2015; Ivanova et al., 2016).
	-	A modern solution for optimizing
(Pachev, 2014).	-	plant nutrition and a very useful event in
	-	agriculture is leaf fertilization. The best
	-	digestible and best-performing are the
	-	complex foliar fertilizers. They deliver the
	-	microelements to the plants in an easily
	-	digestible form and in an amount sufficient
	-	to activate the metabolism processes
	-	(Pachev, 2014).
	-	The use of leaf fertilizers as an
	-	opportunity to further supply the plants
	-	with nutrients directly involved in the
	-	photosynthesis process through the
	-	leaves is important for improving the diet
	-	and quality of produce.
	-	It is essential to introduce bio-nutrients
	-	that have a nutritional and stimulating
	-	effect.
	-	Because they are complex, containing all
	-	necessary nutrients, humic acids and
	-	enzymes, they have a positive influence
	-	on plant growth, increase their resistance
	-	to adverse factors and increase their
	-	productivity (Pavlov and Valchev, 2013).
Valchev, 2013).	(Pavlov and	Foliar fertilization increases yield
(Pachev, 2003; Pachev, 2004; Pachev,	-	and quality of production (Pachev, 2003;
2008; Pachev, 2013).	-	Pachev 2004; Pachev 2008; Pachev,
Lebosol®-Total Care	-	2013).
	-	The aim of the present study is to
	-	determine the effect of Lebosol®-Total
	-	Care on the resistance of young lucerne
	-	to leaf pathogens as there was no data
	-	available in our study on the use of
	-	Lebosol® Total Care in this area.
Total Care	Lebosol®-	

Nannf.) (1990),
(Pseudopeziza jonesii
 10
 20
 0
 1 –
 10%, 3 –
 25 %, 5 – 50%, 7 – 50 75
 % 9
 – 75 %
 (%)
 (Mc Kinney)

Nikolova (1990) and *Pseudopeziza jonesii* Nannf.) on a six-dimensional scale, modified by us.

Approximately 10 samples, including 20 plants that were randomized from the control and leaf fertilization variants, were treated for the degree of attack by leaf pathogens of each variety.

The rate of attack is reported on a 0-9 scale, where with 0 is marked with healthy plants, with 1 – single spot plants or leaf area affected by 10%, 3 – affected area up to 25%, 5 – to 50%, 7 – 50 to 75% of the leaf area and 9 highly infested plants – more than 75% of the leaves. Based on the attack rate, the infestation index (%) (disease development index) was calculated by Mc Kinney's formula for each subgroup and averaged over the year.

Data on precipitation and average monthly air temperatures from the meteorological station of the Institute were used to characterize the meteorological conditions.

RESULTS AND DISCUSSION

The weather conditions during the survey year were comparatively favorable for the growth and development of the young alfalfa (Figure 1).

During the autumn-winter period, extreme temperatures were not observed, and rainfall was slightly above the long-term rate for the area. The rich autumn-winter moisture distribution of the soil ensures even germination and good garnishing of lucerne crops.

The spring was characterized by precipitation and temperatures around the norm. Exceptions were rainfall in May exceeding 100 mm long. In the following months of the young alfalfa vegetation, the temperatures and the amount of

precipitation were the norm, but precipitation was unevenly distributed.

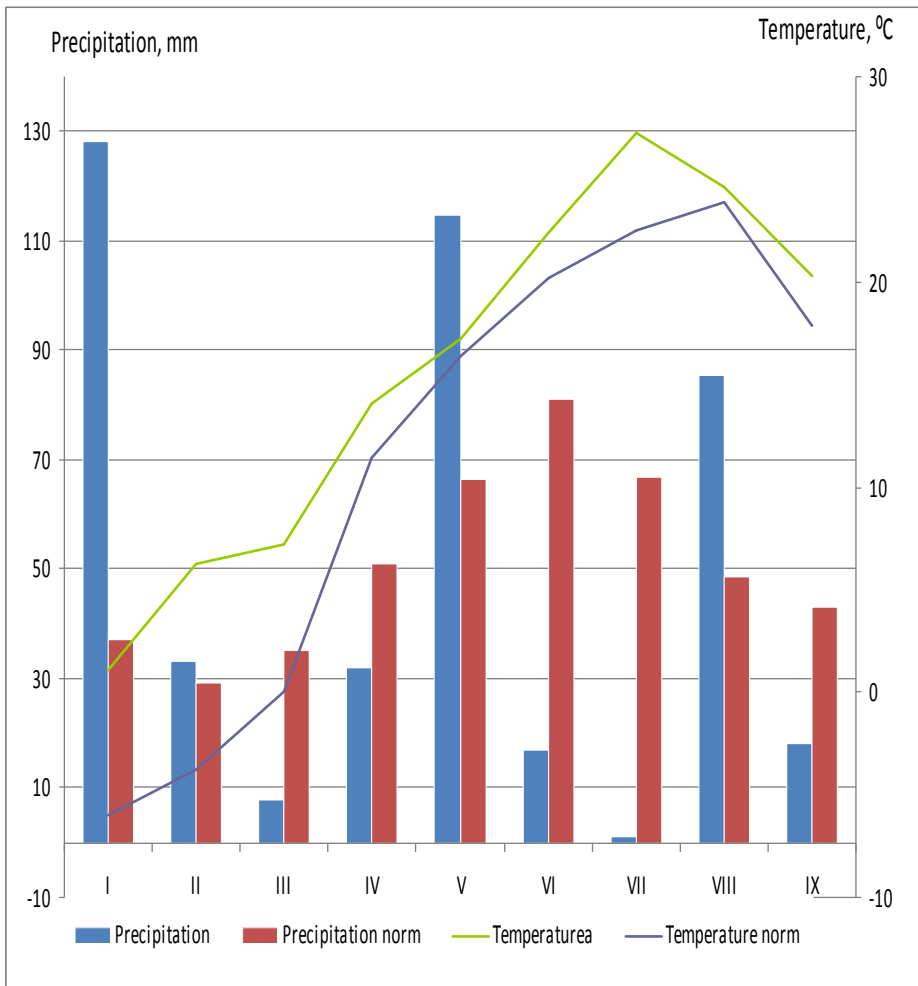


Fig. 1. Precipitation, amount of precipitation and air temperature sums for the 2014

3

(*Pseudopeziza jonesii* Nannf.)
medicaginis)

(*Pseudopeziza*

The young alfalfa managed to show its productive potential, forming 3 sub-populations, but over-humidification at certain times created conditions for the development of fungal diseases.

During the study period, lesions of yellow leaf spots (*Pseudopeziza jonesii* Nannf.) And black leaf spots (*Pseudopeziza medicaginis*) were observed in all subspecies of newly

(*Uromyces striatus*),

planted lucerne plants except rust (*Uromyces striatus*), which developed only in the second and third subtraction.

The leaves and stems of the plants formed oblong, yellow to orange or dark brown to black spots.

The results obtained in the first subsurface for lesions of yellow leaf spots indicate that the control variants strongly attacked the Prista 4 variety, respectively the 55% affected leaf area (Table 1). The reported value for the attack index in Lebosol®-Total Care sowed is 30%, respectively. A strong positive effect from the application of the product is positive for Prista 3 strain (54.3% affected leaf area for the control and 25% for the treated crop (Table 1).

4, 55%
Lebosol®-Total Care, 30%.
3 (54,3%
25%
1).

1.

***Pseudopeziza jonesii* Nannf**

, %

Table 1. Index of attack by *Pseudopeziza jonesii* Nannf. of alfalfa varieties in regrowth and mean for the first production year, %

Varieties	<i>Pseudopeziza jonesii</i>							
	/ ontrol				„Lebosol®- O”			
	1 regrowth	2 regrowth	3 regrowth	Mean	1 regrowth	2 regrowth	3 regrowth	Mean
3 / Prista 3	50%	58%	55%	54.3%	25%	20%	30%	25%
4/ Prista 4	55%	58%	50%	54.3 %	30%	40%	35%	35%
5/ Prista 5	35%	54%	50%	46.3%	35%	50%	35%	40%
1/ Mnogolistna 1	30%	55%	55%	46.6%	35%	35%	35%	35%
/ Roli	50%	50%	50%	50%	30%	55%	45%	43.3%
/ Evropa	50%	55%	60%	55%	35%	30%	55%	40%
/Mean	45%	55%	53.3%	51%	31.6%	38.3%	39.1%	36.3%

(*Pseudopeziza jonesii* Nannf.),

The degree of attack on the second and third pathogenesis of the pathogen (*Pseudopeziza jonesii* Nannf.), Both in the treated variants and the corresponding controls, is the highest. The results show that the strong positive effect of the product in Prista 3 is preserved. Data from the third subspecies shows that Europe’s most powerful attack (60% for control and 55% for foliar application).

(60%

55%

Pseudopeziza jonesii Nannf.,

Lebosol®- tal Care

(2).

On average for the entire vegetation, the values for the *Pseudopeziza jonesii* Nannf. Invasion index for the harvested varieties with foliar application are lower than those of the respective controls. The slightest effect of Lebosol®-Total Care on the resistance to the pathogen, in all substrates and on average for vegetation, was reported in a variety of Europe showing a stronger sensitivity (Table 2).

2.

Pseudopeziza medicaginis

, %

Table 2. Index of attack by *Pseudopeziza medicaginis* of alfalfa varieties in regrowth and mean for the first production year, %

Varieties	<i>Pseudopeziza medicaginis</i>							
	/ ontrol				„Lebosol®- O”			
	1 regrowth	2 regrowth	3 regrowth	Mean	1 regrowth	2 regrowth	3 regrowth	Mean
3 / Prista 3	35%	55%	55%	48.3%	20%	25%	35%	26.6%
4/ Prista 4	30%	55%	55%	46.6%	20%	30%	35%	28.3%
5/ Prista 5	30%	53%	55%	46%	35%	50%	50%	45%
^{1/} Mnogolistna 1	40%	50%	55%	48.3%	35%	40%	45%	40%
/ Roli	35%	55%	55%	48.3%	30%	35%	35%	33.3%
/ Evropa	30%	50%	50%	43.3%	35%	40%	45%	40%
/Mean	33.3%	53%	54.1%	49.3%	29.1%	36.6%	40.8%	35.5%

medicaginis)

(*Pseudopeziza*

(30% - , 50% - , 50% -).

Lebosol®-Total Care,

3,
20 %, 25 % 35 %.

(*Pseudopeziza medicaginis*.),

Lebosol®-Total Care

With respect to the *Pseudopeziza medicaginis* attack, the results obtained show that in the control variants, in all subspecies, lowest values for the black leaf leaf attack index were established for Europe (30% - first sub-row, 50% - second, 50% - third).

In Lebosol®-Total Care variants, the attack on the ground is the lowest in Prista 3, and in the three subpopulations, respectively 20%, 25% and 35% respectively.

The reported *Pseudopeziza medicaginis* index values, both in the treated variants and the corresponding controls, were highest in the third subgroup.

The analysis of the results shows that treatment with the product Lebosol®- Total Care has had a positive effect on

newly created lucerne crops, which is to increase the resistance of all varieties to *Pseudopeziza medicaginis*.

The highest positive effect of Lebosol®-Total Care was found for Prista 3, where the infestation index, on average, of three sub-populations decreased from 49% for the control to 26.6% for the treated variant. The data for all subsets and on average for vegetation characterize Prista 5. as the most sensitive to 45% pathogen.

The results of the phytopathological evaluation of the young lucerne plants in rust (*Uromyces striatus*) (Table 3) show a positive effect of the product on the sustainability of all varieties included in the study

3. ***Uromyces striatus***, %

newly created lucerne crops, which is to increase the resistance of all varieties to *Pseudopeziza medicaginis*.

The highest positive effect of Lebosol®-Total Care was found for Prista 3, where the infestation index, on average, of three sub-populations decreased from 49% for the control to 26.6% for the treated variant. The data for all subsets and on average for vegetation characterize Prista 5. as the most sensitive to 45% pathogen.

The results of the phytopathological evaluation of the young lucerne plants in rust (*Uromyces striatus*) (Table 3) show a positive effect of the product on the sustainability of all varieties included in the study

Table 3. Index of attack by *Uromyces striatus* of alfalfa varieties in regrowth and mean for the first production year, %

Varieties	<i>Uromyces striatus</i>							
	/ ontrol				„Lebosol®- O”			
	1 regrowth	2 regrowth	3 regrowth	Mean	1 regrowth	2 regrowth	3 regrowth	Mean
3 / Prista 3	-	65%	75%	70%	-	22%	28%	25%
4/ Prista 4	-	78%	77%	77.5%	-	37%	33%	35%
5/ Prista 5	-	50%	50%	50%	-	32%	48%	40%
1/ Mnogolistna 1	-	50%	50%	50%	-	40%	50%	45%
/ Roli	-	77%	53%	65%	-	50%	50%	50%
/ Evropa	-	80%	80%	80%	-	48%	54%	51%
/Mean	-	66.6%	64.1%	65.4%	-	38.1%	43.8%	41%

In the first subplot the pathogen did not develop. In the control variants, in the second and third substrates, a strong attack was observed in Prista4 and Europe 77.5 and 80% of the affected leaf area), and slight in the varieties Prista 5 and Multiplex 1 (50%). In foliage crops with Lebosol®-Total Care the lowest attack rate was found in Prista 3, 22% in the second substrate and 28% in the third, followed by Prista 4, respectively 33% and 37% respectively.

In the first subplot the pathogen did not develop. In the control variants, in the second and third substrates, a strong attack was observed in Prista4 and Europe 77.5 and 80% of the affected leaf area), and slight in the varieties Prista 5 and Multiplex 1 (50%). In foliage crops with Lebosol®-Total Care the lowest attack rate was found in Prista 3, 22% in the second substrate and 28% in the third, followed by Prista 4, respectively 33% and 37% respectively.

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Dynamics of biomass accumulation in sugar beet, fodder beet and table beet

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SUMMARY

The dynamics of development of Standard Bulgarian varieties of sugar, fodder and table beet of Agricultural Institute – Shumen collection has been studied during the period 2014-2015. The climatic conditions during the test period are with extreme deviations from the climatic norms for the region.

It has been established an intensive accumulation of roots and leaves biomass during September and October. There are significant differences in the dynamics of biomass increase in dependence on the genotype, and it is more significant for the tested fodder beet forms. The sugar and semi-sugar beet hybrids have more sustainable to the media factors biomass accumulation than the direct varieties and parental components.

Key words: sugar beet, fodder beet, root yield, leave mass

INTRODUCTION

One of the ways of solving the problem with the balanced feeding is the inclusion of fodder sugar and semi-sugar beets in the day rations of livestock

Noskaitit, 2000).

(Tanosiunier and

(Tanosiunier and Noskaitit, 2000). The high yields and concentrations of nutritive substances and vitamins, and the good taste qualities condition the use of fodder and semi-sugar beets for fresh forage and their inclusion in the autumn-winter period's rations of the ruminant animals (Badawi et al., 2002).

2002).

(Badawi et al.,

The high sugar content, combined with high productivity makes actual the significance of the sugar beet for the production of organic sweet syrups. The preferred source of raw material for bio-ethanol production in the European countries is the sugar beet (Hinkova and Bubnik, 2001; Oktay and Ozturk, 2004).

(Hinkova and Bubnik, 2001; Oktay, 2004).

The beet is a plant of the moderate climate and the most favorable for its development average day temperature is 15-20 , and the temperature sum, necessary for the entire vegetation is about 2800 . The soil's humidity is the key factor for the beets growing because of the dry continental climate in our country. For its optimum development this crop needs rainfalls of about 550-600 mm for the year. The rainfalls sum in Northern Bulgaria is in the norm, but during the last years there is an uneven distribution of their quantities during the beets development. Unfavorable for the quality indices are the late September rainfalls, causing secondary vegetation and decrease of the sugar and dry matter content in the roots.

15-20 ,

2800 .

550-600 mm.

The summer droughts in July-August are also very unfavorable for the crop development, especially when accompanied by comparatively high daily temperatures and low atmospheric humidity. The results of our researches show (Kikindonov, 2011a) that in extreme climatic conditions of continuous drought the yield of beets is at serious risk.

(Kikindonov, 2011a)

(Uchkunov, 2006), in vitro (Slavova and Kaschieva, 2004; Zayova et al., 2004a; Zayova et al., 2004b). (Kikindonov and Kikindonov, 2012). (Morvan et al., 2000). (Hagihara et al., 2001; Kikindonov, 2014). 1239 (Uchkunov, 2004), (Kikindonov and Kikindonov, 2012), 56 (Kikindonov, 2011b).

The monogerm varieties, hybrids of monogerm male-sterile lines and multigerm pollinators are confirmed to be the best in the beets breeding (Uchkunov et al., 2006), as part of the breeding materials used, are obtained under in vitro conditions (Slavova and Kaschieva, 2004; Zayova et al., 2004a; Zayova et al., 2004b). In Europe usually the triploid hybrids are higher yielding and with better technological qualities than the diploid hybrids, while the diploid hybrids have higher seeds germination (Kikindonov and Kikindonov, 2012). The controlled inclusion of sugar beet varieties of normal and high yield direction in the forage balance increases the productivity of the ruminants without side effects and that is proved. The fodder beet, with its high yields and forage value is a traditional source of fresh forage (Morvan et al., 2000). The semi-sugar beet hybrids combine successfully the high productivity of the fodder beet pollinators with the increased dry matter content and monogermity of the sterile maternal components of sugar beet. This makes them appropriate for mechanized intensive production on bigger areas. (Hagihara et al., 2001; Kikindonov, 2014).

The researches on the dynamics of biomass accumulation in sugar, semi-sugar and fodder beet were discontinued for a certain time. At the same time in the Agricultural Institute - Shumen have been created new beets varieties - Kom, 1239 (Uchkunov, 2004), Diex (Kikindonov and Kikindonov, 2012), the fodder beet varieties Sasha and Preslav, the semi-sugar beet Vessi and Hybrid 56 (Kikindonov, 2011b). The study of the dynamics of accumulation of fresh and dry mass in the roots and leaves of standard varieties would allow to determine the optimum terms of harvesting the varieties used in the practice and to make appropriate schemes of breeding new varieties for forage production.

2014-2015
 3 (Sasha, Preslav, Triga)
 2 (SKG SKg)
 , 5
 -Hybrid 56, Vesi, MS124xSKG,
 Pliska MS1213xSKg, (M-Dx)
 (M 5319R)
 8
 MS6634xM-Dx, MS222xM-Dx, MS201xM-Dx,
 MS015xM-Dx, MS222xM5319, MS201xM5319,
 MS5140xM5319, MS5332xM5319,
 (Diex)
 (Peshtera)
 - Radost 1 Radost 2.
 10.8 m².
 (2x 3x)
 (kg/da)
 19 , 20 , 12
 - 2014 . 10 , 4
 16 - 2015 .
 (Lidanski,
 1988).

- 2014 . (271 mm
 105 mm)
 (Ta 1)
 25.05,

MATERIAL AND METHODS

Three fodder beet varieties (Sasha, Preslav, Triga) and two multigerm fodder beet pollinators (SKG and SKg), 5 diploid and triploid monogerm semi-sugar beet hybrids (Hybrid 56, Vesi, MS124xSKG, Pliska MS1213xSKg,), diploid (M-Dx) and tetraploid (M 5319R) sugar beet pollinators and their eight monogerm hybrids (MS6634xM-Dx, MS222xM-Dx, MS201xM-Dx, MS015xM-Dx, MS222xM5319, MS201xM5319, MS5140xM5319, MS5332xM5319), Standard diploid (Diex) and triploid (Peshtera) monogerm sugar beet varieties, as well as two table beet origins (Radost 1 and Radost 2) have been tested during 2014-2015.

The field tests are realized on a carbonate black soil with a weekly alkaline reaction of the soil solution, under non-irrigation conditions. The randomization of the test field is according the long plot method, in four replications, the area of the harvest plot is 10.8 m². The Group Standard includes two certified fodder beet varieties (diploid and triploid) and two sugar beet varieties (2x and 3x).

The weights of the roots and the leaves' mass have been measured on: 19.08, 20.09 and 12.11. in 2014, and on 10.08, 04.09 and 16.11. in 2015. The results are treated statistically by dispersion analysis according to Lidanski (1988).

RESULTS AND DISCUSSION

The continuous rainfalls during April-May of 2014 slowed the sowing to 25.05 (Table 1), while the normal sowing date for the last years is the end of April. The flooding of the experimental fields, the late rarefaction and hoeing slowed down additionally the development of beet. The vegetation after May run in favourable for beet conditions of wet and cool summer. The active biomass accumulation, sugar and dry matter content continued to the end of November.

1.
 , 2014-2015 .

Table 1. Meteorological conditions in the region of Agricultural Institute - Shumen, 2014-2015

Year	Month	- mm Rainfalls - mm					, C Air temperature	
		/Decades			Sum	Norm	Mean	
		I	II	III				
2014	IV	-	33.8	10.6	44.4	41.0	11.5	
	V	26.0	52.9	147.6	226.5	64.0	15.1	
	VI	37.0	19.9	14.6	71.5	75.0	19.3	
	VII	2.2	4.4	63.9	70.5	60.0	21.9	
	VIII	29.0	37.3	4.0	70.3	42.0	22.5	
	IX	54.3	0.2	-	54.5	28.0	17.5	
	X	4.5	3.0	48.3	55.8	53.0	11.6	
/ Total for the period					593.5	363.0		
2015	IV	32.6	10.5	10.0	53.1	41.0	14.6	
	V	6.3	3.2	6.3	15.8	64.0	20.8	
	VI	2.5	18.6	3.0	24.1	75.0	23.6	
	VII	2.3	6.6	-	8.9	60.0	27.1	
	VIII	0.6	8.8	15.2	24.6	42.0	28.4	
	IX	1.1	40.2	-	41.3	28.0	22.8	
	X	14.9	12.7	26.0	53.6	53.0	15.2	
/ Total for the period					221.4	383.0		

2015 .

27.04

(-)

57 mm

mm,

200

The agro-meteorological conditions in 2015 are unfavorable for beet's development. A continuous drought followed the sowing (27.04.), and this brought to slower germination, irregular initial development and ungraded sowings. The volume of the rainfalls in the region during the vegetation period is extremely insufficient. The growth and the accumulation of biomass in the first three months (May to July) are in conditions of sharp water deficiency – the sum of rainfalls is 57 mm, with normal quantity of 200 mm, and with increased daily air temperatures. In these conditions record low yields are registered for the tested fodder and semi-sugar beet breeding materials.

The differences of the agro-meteorological conditions during the test period makes it possible to assess the influence of the media factors on the

2014

20.09.

kg/da

kg/da.

(2).

1905 kg/da

3839 kg/da

7500 kg/da.

1000

dynamics of growth of the tested varieties, hybrids and pollinators of sugar and fodder beet.

Despite the shorter vegetation in 2014, in the conditions of optimum waterproofing in the period from the end of August to the beginning of November an intensive mass accumulation runs in the roots. In this regard the values of the formed biomass of the fodder beet forms exceed significantly those of the sugar beet in the tests (Table 2). Thus on 19th of August the average root yield of the fodder beet forms is 4562 kg/da, and the mean leaves mass is 3276 kg/da, and on 20.09. are measured on average 5569 kg/da roots with 1905 kg/da mass of leaves. The most intensive biomass accumulation on the first date of measurements is registered for the variety Pliska (5821 kg/da of roots and 3839 kg/da leaves mass). On the second measurement date we note somewhat a leveling of the values of the formed total biomass of over 7500 kg/da. It is characteristic for the sugar beet the more intensive leaves mass accumulation, registered on the first measurement date. With the measurements in September the mass of the roots is already higher than the leaves mass. On the last measurement date we note intensive increase of the mass accumulated in the roots – the weight of the roots has increased on average with about 1000 kg/da. Obviously the late sowing affects negatively the sugar beet root yield. The biomass accumulation in August and September is slower, and the intensive increase of the root weight starts at the end of the vegetation, when a sharp decrease of the leaves mass begins. The differences between the sugar beet forms with different ploidy levels are not so distinct. It is clear that the shortened vegetation does not allow the triploid sugar beet forms to realize at full their productive potential.

2.

, 2014 . ()

Table 2. Dynamics of biomass (roots and leaves) accumulation in sugar and fodder beet origins, 2014

Variant	/ Yield, kg/da					
	I date of measurement		II date of measurement		III date of measurement	
	Roots	Leaves	Roots	Leaves	Roots	Leaves
/ Fodder beet						
1. Sasha - 4 ,	5518	2821	6134	1315	6190	1298
2. Hybrid 56 – 3	4554	2964	6207	1768	6870	1452
3. Preoslav - 2x	4196	4036	5524	2149	6420	1923
4. Vessi - 2x	3821	3250	4409	2087	4860	1179
5. riga - 4	4304	2482	5811	1357	6350	1518
6. Pliska – 3	5821	3839	5945	1958	6480	1530
7. SKG – 4x	3893	3089	4732	1756	4890	1488
8. MS124 x SKG 3x	4696	3196	5463	1881	5990	1351
9. SKg – 2x	4286	3036	5919	2042	6180	1196
10. MS0213xSKg 2x	4018	3618	5189	2637	5450	1732
11. Radost 1 – 2	1089	661	1829	196	2590	268
12. Radost – 2	2246	411	2287	232	2680	179
/ mean	4080	2819	4984	1623	5433	1294
/ Sugar beet						
13. -D - 2	3125	3643	3143	2887	4460	1863
14. MS6634 x M-Dx	3214	3429	3292	2470	4210	1429
15. MS222 x M-Dx	2946	3375	3500	2702	4830	1738
16. MS201 x M-Dx	3536	4089	3524	2024	3760	1446
17. MS015 x M-Dx	3125	3857	3839	2857	4420	1810
18. Diex – 2	2750	3411	3196	2179	4220	1494
19. M 5319R – 4x	3339	4196	3401	2185	3740	1351
20. MS222 x M5319	2857	3232	3030	1923	4180	1863
21. MS201 x M5319	3000	3446	3827	2536	4730	1339
22. MS5140 x M5319	2714	3003	2839	2619	4240	1417
23. MS5332 x M5319	3286	3821	3286	2482	4150	1179
24. Peshtera – 3	2518	3339	3429	2393	4170	1155
/ mean	2894	3382	3275	2403	4151	1509
GD 1 %	910	1017	985	310	900	378
%	4,24	4,68	4,01	3,47	3,65	3,14

- In case of sufficient soil moisture during the vegetation period the late sowing does not affect significantly the dynamics of the biomass formation in table beet.

- In the water deficiency conditions of 2015 the levels and the temp of roots' growth are significantly decreased (able 3). The registered values of the accumulated mass in the roots and the leaves during the measurements in August and September are extremely low

2015 .

(3).

2687 kg/da
899kg/da

- for the fodder beet forms mean 2687 kg/da for roots and 899kg/da for leaves.
- In October, due to the rainfalls, the accumulation of biomass, especially in the roots, is already with a normal intensity.

3.

, 2015 .

Table 3. Dynamics of biomass (roots and leaves) accumulation in sugar and fodder beet origins, 2015

Variant	/ Yield, kg/da					
	1 I date of measurement		2 II date of measurement		3 III date of measurement	
	roots	leaves	Roots	Leaves	Roots	Leaves
/ Fodder beet						
1. Sasha -4	2796	582	2879	628	4589	460
2. Hybrid 56 – 3	2446	497	3050	475	4289	503
3. Preslav - 2x	2561	656	2861	578	4250	700
4. Vessi - 2x	2668	570	2689	418	3325	465
5. Triga - 4	2418	1225	3061	893	4486	1100
6. Pliska – 3	2764	1115	3218	768	4793	873
7. SKG – 4x	2839	1269	3504	968	3861	873
8. MS124 x SKG 3x	2725	890	3643	1060	4675	1073
9. SKg – 2x	2782	1034	3604	678	4850	770
10. MS0213xSKg 2x	3318	1070	4114	830	4221	682
11. Radost 1 – 2	2589	628	3229	465	3750	538
12. Radost – 2	2341	682	3114	713	3582	553
/mean	2687	899	3190	944	4141	872
/Sugar beet						
13. -D - 2	1986	758	3546	1013	2779	663
14. MS6634 x M-Dx	1886	842	2871	682	3975	1038
15. MS222 x M-Dx	1968	778	3846	1023	2369	652
16. MS201 x M-Dx	2789	1039	3379	785	3482	718
17. MS015 x M-Dx	2689	1245	3618	818	4521	745
18. Diex – 2	2814	1373	4243	1163	4704	1395
19. M 5319R – 4x	2925	1210	3473	907	4254	810
20. MS222 x M5319	3243	1110	3936	832	4393	730
21. MS201 x M5319	2996	942	4664	1130	5654	795
22. MS5140 x M5319	2236	1227	3461	1018	4396	955
23. MS5332 x M5319	2925	1285	3354	883	4325	1038
24. Peshtera – 3	2457	1207	3257	918	3896	1429
/mean	2586	1033	3750	962	3964	914
GD 1 %	503	213	619	193	845	194
%	4.92	5.76	4.58	6.29	5.43	6.16

- With water deficiency during the
- summer months the dynamics of biomass
- accumulation in the two table beet origins
- is normal, with an increased intensity

1000 kg -

2015 .

1033 kg/da, 962 kg/da 914 kg/da.

MS 201 x M5319 –

5654 kg/da,

after the first date of measurement. As a result nearly a 1000 kg higher root yield is registered in November.

The sugar beet manifests as more adaptable to the unfavorable climatic conditions. In 2015 the formed leaves mass is with significantly lower values, no matter the date of measurement respectively 1033 kg/da, 962 kg/da and 914 kg/da.. The sugar beet roots' weight increases most intensively in September-October. The triploid sugar beet hybrids succeed to form higher root yield than the diploid hybrids. It is characteristic for the year the evenly decrease of the leave mass.

The decrease of the ratio leave mass: root weight in vegetation water deficiency is significant, as an adaptive mechanism for accumulation of assimilates in roots in unfavorable conditions.

In these unfavorable conditions the high productivity of the hybrid MS 201 x M5319 stands out – the measured root yield in November is 5654 kg/da, significantly higher than the yield of all remaining varieties and hybrids of sugar and fodder beet.

CONCLUSIONS

The biomass accumulation in the tested beet forms is most intensive during September and October. It is registered a strong variation in the dynamics of growth depending on the genotype, and it is more significant for the tested fodder beet forms.

The sugar and semi-sugar beet hybrids have more stable to the media factors biomass accumulation than the direct varieties and the parental components.

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