

## Stem formation at alfalfa varieties and correlative dependences with some main parameters

### Стъблообразуване при сортове люцерна и корелативни зависимости с някои основни параметри

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#### Abstract

During the period 2006-2009 in IFC-Pleven were studied 9 alfalfa varieties: Europe, Prista 2, Prista 3, Prista 4, Obnova 10, Pleven 6, Dara, Multifoliate and Dama. In the first year of its development alfalfa formed on average 1.42 number of stems per plant. During second and third year their quantity increased to 2.14 and 3.83 number whereafter during fourth year they decreased to 3.28. In all years (except the first year) was found a trend of increase in stem number from spring to autumn regrowth with average values 2.11, 2.85 and 3.05 respectively. Average for the four-year period the greatest number of stems (over the average for the nine studied varieties – 2.67) formed the Multifoliate variety followed by Europe, Prista 4, Obnova 10 and Dama. The varieties which are characterized by less density of stand (number of plants per unit area) had a greater number of stems per plant ( $r = -0.530$ ). Correlations with mean and high positive value were found between stem formation and amount and distributions of rainfall during vegetation period ( $r = 0.989$ ), year of alfalfa development ( $r = 0.861$ ), nodulation ( $r = 0.763$ ), weight of root mass ( $r = 0.411$ ) as well as correlations with mean negative value between number of stems and percentage of damaged stems by *Apion seniculus* ( $r = -0.456$ ).

**Keyword:** alfalfa, correlations, stems, varieties

#### Detailed Abstract

През периода 2006-2009 г. в ИФК-Плевен са проучени 9 сорта люцерна (Европа, Приста 2, Приста 3, Приста 4, Обнова 10, Плевен 6, Дара, Многолистна и Дама) за установяване стъблообразуването при тази култура и корелативните зависимости с някои основни параметри. Опитът е изведен по блоковия метод, в 4 повторения и големина на опитната парцела от 5 m<sup>2</sup>.

През първата година на развитието си люцерната формира средно 1.42 броя стъбла/растение. През втората и третата година количеството им нараства до 2.14 и 3.83 бр., след което през четвъртата година те намаляват до 3.28 бр.

През всички години (с изключение на първата) се установява тенденция на нарастване в броя на стъблата от пролетния към есенния подраст (със средни стойности съответно 2.11, 2.85 и 3.05), кореспондиращо с нарастващата сума на валежите.

Средно за четиригодишния период най-голям брой стъбла (над средния за деветте проучвани сорта – 2.67) формира сорт Многолистна, следван от Европа, Приста 4, Обнова 10 и Дама. Сортовете, характеризиращи се с по-ниска плътност на тревостоя (брой растения на единица площ) имат по-голям брой стъблата/растение ( $r = -0.530$ ). Установени са корелации със средна и висока положителна стойност между формирането на стъблата (стъблообразуването) и количеството и разпределението на валежите през вегетационния период ( $r = 0.989$ ), годината на отглеждане на люцерната ( $r = 0.861$ ), грудкообразуването ( $r = 0.763$ ) и теглото на кореновата маса ( $r = 0.411$ ), както и корелация със средна отрицателна стойност между броя на стъблата и плътността на посева ( $r = -0.530$ ). Между броя на стъблата и степента на повредени от *Apion seniculus* стъбла съществува отрицателна корелация ( $r = -0.456$ ) като в зависимост от силата на взаимодействие сортовете се отнасят в три обособени групи. Приста 4 и Многолистна се открояват със силно изразена отрицателна корелационна зависимост (над  $-0.666$ ) – I група, следвани от Европа, Приста 2, Дама и Плевен 6 със средна отрицателна корелация от  $-0.333$  до  $-0.666$ ) – II група и Приста 3, Обнова и Дара със слаба отрицателна корелационна зависимост (под  $-0.333$ ) – III група. Разликите по отношение на силата на корелационните коефициенти при проучваните сортове вероятно се дължи на определени различия в биохимичния състав, които влияят върху разглежданата зависимост брой на стъблата и степен на нанесена от хоботника повреда.

От направения анализ на варианса по отношение на стъблообразуването се установява, че годината на отглеждане на люцерната оказва най-силно, доминиращо влияние и доказан ефект върху този показател – 87.0% от общото вариране (A). Значително по-слабо изразено и недоказано (4.5%) е влиянието на фактор B (сортове) върху варирането на показателя брой стъбла. Взаимодействието между двата фактора (A×B) е незначително и силата на влияние е 4.1%.

**Ключови думи:** люцерна, корелации, стъбла, сортове

## Introduction

The productivity and nutritive value of alfalfa defined it as a leading perennial legume forage crop (Sauvant et al., 2002; Đukić, 2002; Radović et al., 2004; Dinić and Đorđević, 2005, Stanisavljević et al., 2008a). According to Volenec et al. (1987) the alfalfa productivity could be described by three components: plants per unit area, number of stems per plant, productivity per stem.

Stanisavljević et al. (2008b) found a correlation with mean positive value between number of stems and dry matter productivity ( $r = 0.493$ ) and Marinova and Petkova (2010) established mean ( $r = 0.35$ ) to strong correlation ( $r = 0.89$ ) between number of stems per plant and green mass yield as the values of correlation coefficient varied depending on the variety (hybrid) and the age of stand. As a result of study of 43 alfalfa germplasm (varieties, populations, inbred lines and crossbreeds) Petkova and

Marinova (2006) reported that the productivity ( $\text{g}\cdot\text{plant}^{-1}$ ) correlated in a greater degree with number of stems ( $r = 0.64$ ) than with the plant height ( $r = 0.39$ ) and number of plant ( $r = 0.54$ ).

According to Jones (1928) the number of stems is determined by the plant vitality and the expressing of genetic potential of that characteristic is influenced by the environmental conditions. As main factors influencing the number of stems some authors indicated the meteorological conditions, soil conditions (Cowett and Sprague, 1962, 1963; Stanisavljević et al., 2008b) and varietal appurtenance (Stanisavljević et al., 2008b). With determining importance for that parameter were the amount and distributions of precipitations as well as the age of plants i.e. the year of alfalfa growing (Stanisavljević et al., 2008b). According to other researchers the number of stems was in close dependence with the density of alfalfa stand (number of plants per  $\text{m}^2$ ) as between these parameters is found a negative correlation (Rumbaugh, 1963; Bolger and Meyer, 1983; Volenec et al., 1987; Kephart et al., 1992). Lloveras et al. (2006) reported for variation from 2.0 to 3.68 stems per plant in conditions of different density of alfalfa stand and Marinova and Petkova (2010) – from 3 to 6 stems per plant depending on the variety (hybrid) and year of growing. The number of stems was a parameter which had influence on the degree of damaged stems by *Apion seniculus* Kirby and the weevil preference for certain alfalfa varieties (Nikolova and Georgieva, 2010).

The purpose of investigation was to study the stem formation in different alfalfa varieties and its correlative dependences with some main parameter.

## Material and methods

The experimental work was conducted during the period 2006-2009 in Institute of forage crops, Pleven. The subject of the study were 9 alfalfa varieties: Europe (France), Prista 2 (Bulgaria), Prista 3 (Bulgaria), Prista 4 (Bulgaria), Obnova 10 (Bulgaria), Pleven 6 (Bulgaria), Dara (Bulgaria), Multifoliate (Bulgaria) and Dama (Bulgaria). The soil type was slightly leached chernozem. The experiment was realized by the block method with sowing rate  $25 \text{ kg}\cdot\text{ha}^{-1}$  (row spacing 11.5 cm), in 4 replications and plot size of  $5 \text{ m}^2$ . For determination the number of stems per plant at harvest (early flowering stage) of the spring, summer and autumn regrowths were taken soil monoliths with dimensions  $20\times 200\times 30 \text{ cm}$  (width/length/depth) as the number of reported plants was from 50 to 431. The soil monoliths were used also to determining weight of root mass ( $\text{g}\cdot\text{plant}^{-1}$ ) and nodulation (nodule number $\cdot\text{plant}^{-1}$ ).

The observations related to damage of *Apion seniculus* began during the second year of alfalfa development (2007). Correlation coefficients ( $r$ ) were calculated on base of stem number in first alfalfa regrowth and the relevant percentages of damaged stems as the pest harmed in first regrowth.

The mathematical data processing was done by the method of variance analysis with the software product Statgraphics Plus.

## Results and Discussion

The study period (2006-2009) is characterized with higher annual average daily temperature ( $12.7 \text{ }^\circ\text{C}$ ) and less sum of rainfall (541 mm) compared to the same for the previous 20-year period ( $12.2 \text{ }^\circ\text{C}$  and 562 mm respectively) – Figure 1. The

separate experimental years had variable conditions as secured in greatest degree in regard to rainfall was 2009 (613 mm) and in least degree – 2008 (469 mm). The annual average daily temperature was in the limits from 12.2 (2006) to 13.6 °C (2007).

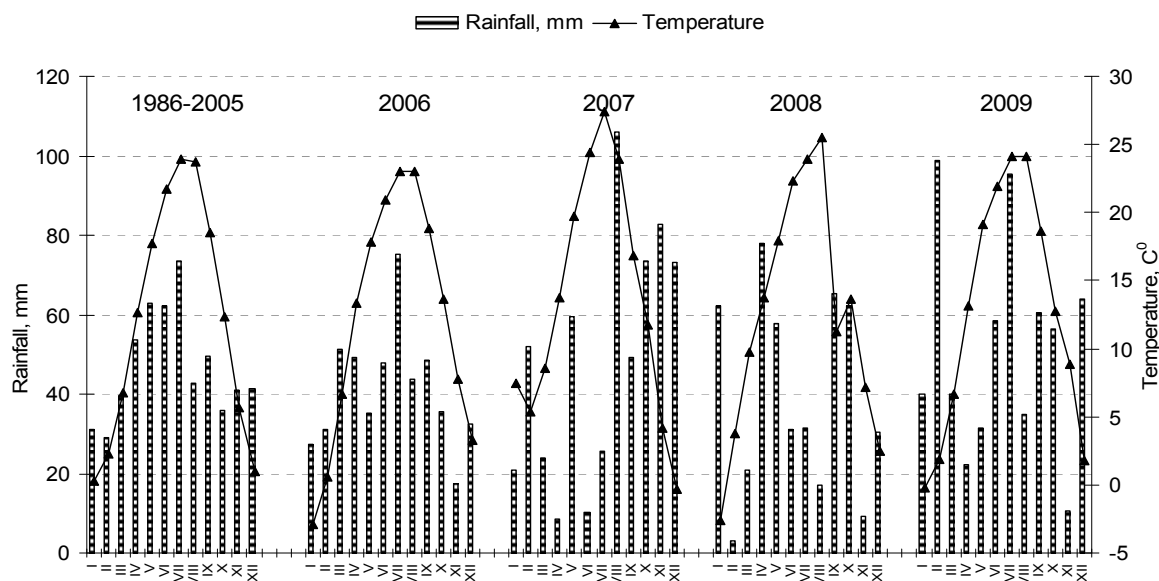


Figure 1. Meteorological characteristic

Фигура1. Метеорологична характеристика

The mean number of stems per plant during the first year of alfalfa development at harvesting of spring regrowth was 1.26 (Figure 2). The differences among varieties were still slightly pronounced and the value of variation coefficient was low (VC=8%). With the progress of vegetation the number of stems increased reaching a maximum in the summer regrowth (1.70) whereafter in the autumn regrowth it decreased (1.31). The variation in summer and autumn regrowth had higher values (19 and 15% respectively) and it was determined as an average (Dimova and Marinkov, 1999). The averaged data by regrowths showed that in the year of establishment of alfalfa stand the greatest number of stems per plant formed Obnova 10 and Prista 3 and during the second experimental year – Multifoliate and Obnova 10. In second year the values of studied parameter increased on average by 50.7% compared to the first year. In all varieties was observed a raising of stem number from spring regrowth (1.42) to autumn regrowth (2.54) as within separate regrowths the variation was from 7 (spring regrowth) to 14% (autumn regrowth).

During the third and fourth experimental years the increase in number of stems per plant continued as in third year it was in limits from 2.32 to 5.66 and in fourth year – from 2.40 to 4.30 numbers. The data during these years were unidirectional in regard to maxima (at varieties Europe, Prista 4 and Multifoliate) and minima (at Pleven 6, Dara and Prista 2). The differences among varieties strengthened whereat the values of variation coefficient were higher – from 10 to 17% in third year and from 13 to 22% in fourth year.

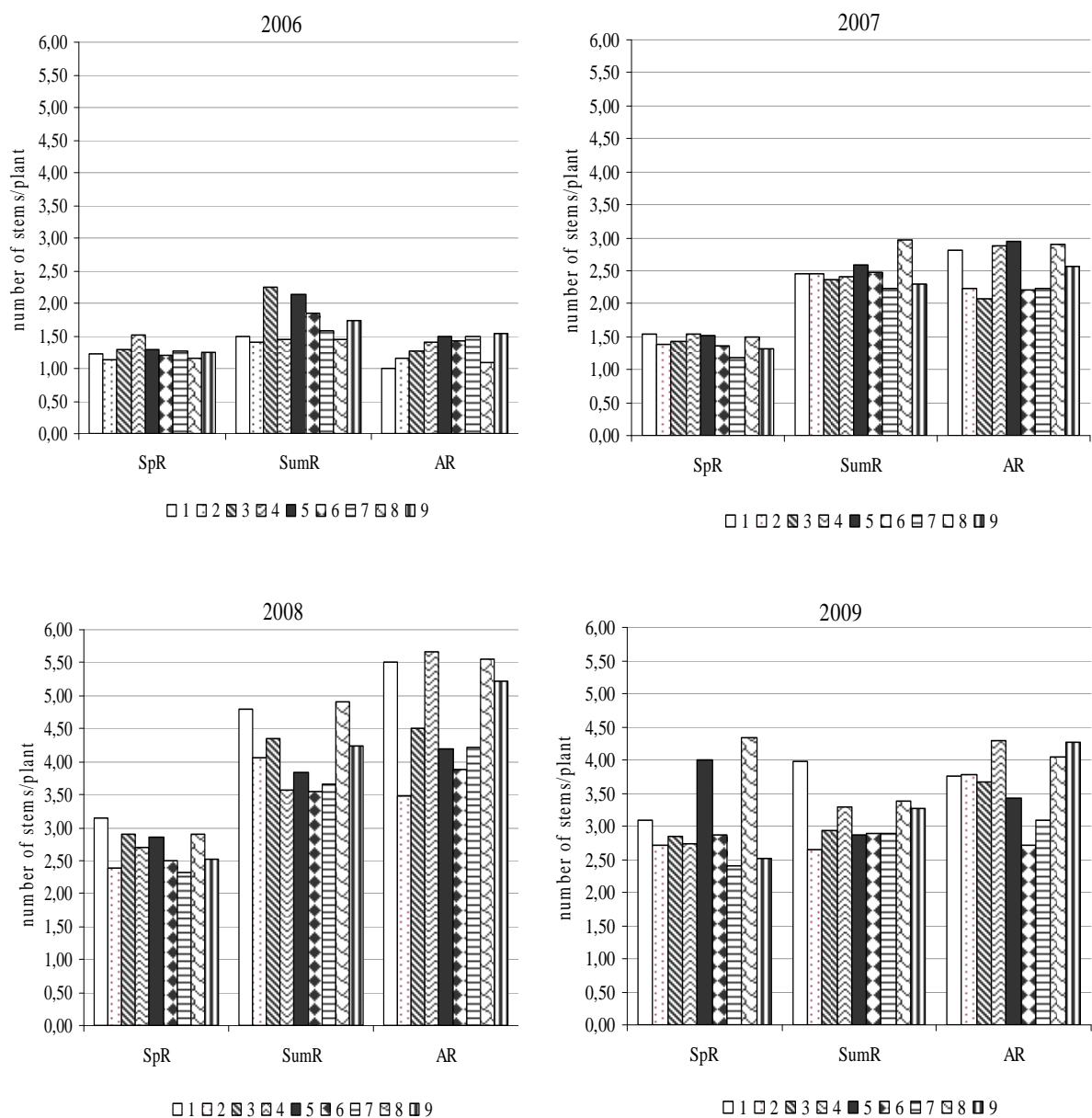


Figure 2. Number of stems per plant at alfalfa varieties

Фигура 2. Брой стъбла на растение при сортове люцерна

Legend: SpR-spring regrowth, SumR-summer regrowth, AR-autumn regrowth; 1-Europe, 2-Prista 2, 3-Prista 3, 4-Prista 4, 5-Obnova 10, 6-Pleven 6, 7-Dara, 8-Multifoliolate, 9-Dama

The averaged data by years showed an unproportional increase in the stems from first (1.42) to second (2.14) and third year of alfalfa development (3.83) whereafter in fourth year they decreased (3.28) (Table 1). Significant differences among varieties within first year were not found but in second year Multifoliolate formed the greatest stem number which was statistically significant ( $p < 0.05$ ) compared to Dara and Prista 3. The next two years distinction among varieties was deepened as Multifoliolate and Europe had with 24.5 and 21.2% more stems (for 2007 and 2008, respectively) in comparison with the rest of varieties.

During all years (except the first year) was outlined a trend of increase in stem number from spring to autumn regrowth (with average values 2.11, 2.85 and 3.05 respectively) corresponding with rising sum of rainfall. The results were in support of established determination (Stanisavljević et al., 2008b) of studied parameter from the growing year (age) of alfalfa and amount and distribution of rainfall during vegetation period.

In conditions of present study the correlation analysis of data showed a stronger dependence of stem formation with the amount and distribution of rainfall during vegetation period ( $r = 0.989$ ) at variation of correlation coefficient from 0.827 (Prista 4) to 0.999 (Europe and Prista 2). The age of alfalfa stand determined in less degree the number of stems (albeit with high value of  $r = 0.861$ ) as at some varieties (Obnova 10, Prista 2) the correlation was stronger pronounced ( $r = 0.917$ ) and at others (Prista 3) – weaker pronounced ( $r = 0.798$ ).

Average for the period 2006-2009 the greatest number of stems (over the average for the group) formed Multifoliate variety followed by Europe, Prista 4, Obnova 10 and Dama. Excepting Dama these varieties are characterized with lower density of the stand (number of plants per unit area) (Georgieva and Nikolova, 2012) and significantly higher number of stems to other varieties. With significantly the lowest values at this parameter are distinguished Dara, Pleven 6 and Prista 2.

Table 1. Mean number of stems per plant at alfalfa varieties during the period 2006-2009

Varieties	2006	2007	2008	2009	2006-2009
Europe	1.24 a*	2.27 ab	4.48 c	3.61 ab	2.90 cd
Prista 2	1.23 a	2.03 ab	3.32 a	3.04 ab	2.41 a
Prista 3	1.60 a	1.96 a	3.91 ab	3.15 ab	2.66 ab
Prista 4	1.45 a	2.27 ab	3.97 bc	3.44 ab	2.78 bcd
Obnova 10	1.63 a	2.35 ab	3.63 ab	3.43 ab	2.76 bc
Pleven 6	1.49 a	2.02 ab	3.31 a	2.82 a	2.41 a
Dara	1.44 a	1.88 a	3.40 ab	2.79 a	2.38 a
Multifoliate	1.23 a	2.46 b	4.46 c	3.92 b	3.02 d
Dama	1.50 a	2.06 ab	3.99 bc	3.35 ab	2.73 bc
average	1.42	2.14	3.83	3.28	2.67

\*values in each column followed by same letters are not significantly different ( $p < 0.05$ )

The number of stems was a main parameter characterizing the degree of infestation by *Apion seniculus* Kirby (*Coleoptera: Curculionidae*) and had influence on the preference of insect at damage infliction. The reason for this was the difference in stem number at different varieties. With the greatest number of stems are distinguished Europe and Multifoliate for which varieties the damage was the weakest and they showed tolerance to weevil damage (Nikolova and Georgieva, 2010).

The dependence between these parameters at studied varieties was expressed by correlation analysis (Figure 3). During the second year of alfalfa development (2007) with strongly expressed negative relationship varying from  $-0.691$  to  $-0.897$  between number of stems and percentage of damaged stems by weevil were distinguished Prista 4, Multifoliate and Prista 2. During 2008 the trend was reserved as strong

negative correlation was established and for Europe and Dara. In regard to the rest varieties during the two years was observed mean negative correlation from  $-0.345$  to  $-0.619$ . It should be noted that the dependence between the number of stems and the degree of damaged stems by *Apion seniculus* during the fourth year of alfalfa development (2009) as a whole was weakest expressed and the strength of interaction between two parameters reached to mean negative correlation (Multifoliolate, Prista 4, Prista 2, Dama and Europe). The year is distinguished and with weakest weevil infestation (Nikolova and Georgieva, 2010), with highest variation coefficient of this parameter (46% compared to 36 and 18% for 2007 and 2008) which probably influenced the strength of interaction between the two parameters. With weak negative correlation is distinguished Prista 3 and Plevan 6 ( $r = -0.206$  and  $r = -0.258$  respectively) as at Prista 3 the tendency was observed and during the previous years. Exception from the found negative correlation was found only in 2009 at Obnova 10 and Dara with  $r = +0.251$  and  $r = +0.397$  respectively.

Average for the period 2007-2009 between the number of stems and the degree of damaged stems by *Apion seniculus* was found a negative correlation as depending on the strength of interaction the varieties were related in three groups. Prista 4 and Multifoliolate are characterized with strongly pronounced negative correlation (over  $-0.666$ ) – I group, followed by Europe, Prista 2, Dama and Plevan 6 with mean negative correlation (from  $-0.333$  to  $-0.666$ ) – II group and Prista 3, Obnova10 and Dara with weak negative correlation (below  $-0.333$ ) – III group. Probably the differences in correlative coefficients at studied varieties are due to certain differences in the biochemical composition which influenced the dependence between number of stems and degree of inflicted damage by weevil.

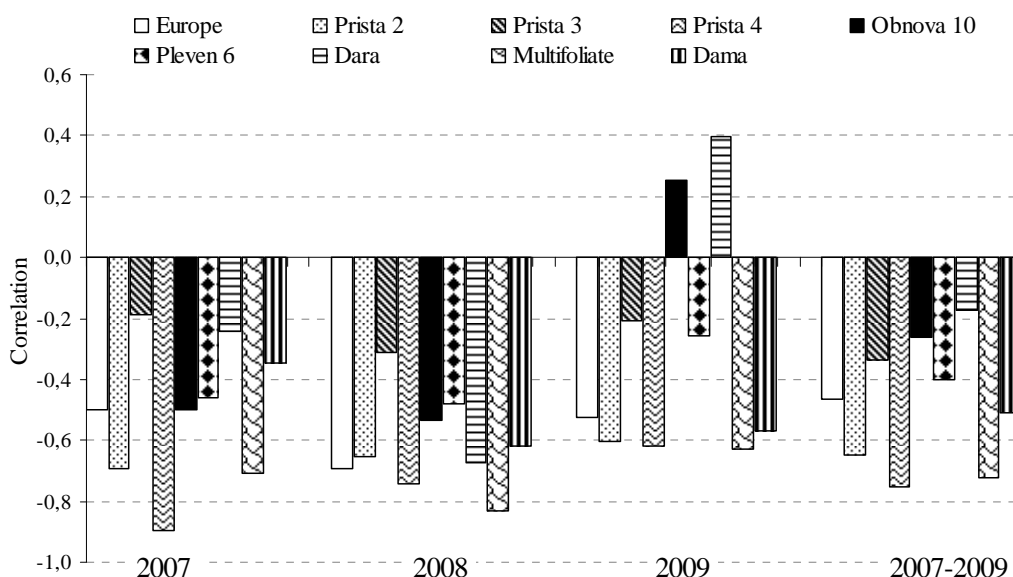


Figure 3. Correlations between number of stems and degree of damaged stems by *Apion seniculus* at alfalfa varieties

The number of stems was an important parameter related not only to alfalfa productivity (Frakes et al., 1961; Marinova and Petkova, 2010) and stand density

(number of stems per m<sup>2</sup>) (Rumbaugh, 1963; Bolger and Meyer, 1983; Volenec et al., 1987; Kephart et al., 1992) but to main parameters of the root system and degree of infestation by *Apion seniculus*. The results from the present experiment (Table 2) showed correlations with mean and high positive value between the considered parameter and weight of root mass ( $r = 0.411$ ) and nodulation ( $r = 0.763$ ) respectively, correlation with mean negative value between number of stems and stand density ( $r = -0.530$ ) and between number of stems and percentage of damaged stems by weevil ( $r = -0.456$ ). Statistically significant correlations were found only between stem number with nodulation (in 2007) and stand density (in 2007 and 2008).

Table 2. Correlative dependences

Parameters	Number of stems per plant				
	2006	2007	2008	2009	2006-2009
Weight of root mass	0.593	0.310	0.202	0.138	0.411
Nodulation	0.101	0.730*	0.681	0.305	0.763
Stand density (number of plants*m <sup>-2</sup> )	-0.398	-0.476*	-0.458*	-0.425	-0.530
Percentage of damaged stems by <i>A. seniculus</i>	-	-0.347	-0.399	-0.085	-0.456

\* significance at  $p < 0.05$

From the made analysis of variance in regard to the stem formation (Table 3) was established that the year of alfalfa growing had the strongest dominant influence and significant effect on this parameter – 87.0% from the total variation (A). Considerably less pronounced and insignificant was the influence of factor B (varieties) on the variation of stem number. The interaction between two factors (A and B) was inconsiderable and the strength of influence was 4.1%.

Table 3. Influence of the factors of variance on stem formation at alfalfa varieties

Source of variation	Degrees of freedom df	Sum of squares SS	Influence of factor %	Mean squares MS
Total	71	71.7	-	-
Factor A - Year	3	62.4	87.0*	1.0
Factor B - Varieties	8	3.2	4.5	20.8
A x B	24	2.9	4.1	0.4
Pooled error	36	3.2	4.4	0.1

\* significance at  $p < 0.05$

## Conclusions

During the first year of its development alfalfa formed on average 1.42 number of stems per plant. During second and third year their quantity increased to 2.14 and 3.83 number whereafter during fourth year they decreased to 3.28. In all years (except the first year) was found a trend of increase in stem number from spring to



autumn regrowth with average values 2.11, 2.85 and 3.05 respectively.

Average for the four-year period the greatest number of stems (over the average for the nine studied varieties – 2.67) formed the Multifoliate variety followed by Europe, Prista 4, Obnova 10 and Dama. The smallest number of stems had Dara, Pleven 6, Prista 2 and Prista 3. The varieties which are characterized by less density of stand (number of plants per unit area) had a greater number of stems per plant ( $r = -0.530$ ). Correlations with mean and high positive value were found between stem formation and amount and distributions of rainfall during vegetation period ( $r = 0.989$ ), year of alfalfa development ( $r = 0.861$ ), nodulation ( $r = 0.763$ ), weight of root mass ( $r = 0.411$ ) as well as correlations with mean negative value between number of stems and percentage of damaged stems by *Apion seniculus* ( $r = -0.456$ ).

## References

- Bolger, T.P., Meyer D.W., (1983) Influence of plant density on alfalfa yield and quality. In: Proc Am Forage and Grassl Conf, Eau Claire, WI 23–26 January 1983. Lexington: American Forage and Grassland Council.
- Cowett, E.R., Sprague, M.A., (1962) *Agronomy Journal*, 54(4), 294–297.
- Cowett, E.R., Sprague, M.A., (1963) *Agronomy Journal*, 55(5), 432–434.
- Dimova, D., Marinkov, E., (1999) *Experimental work and biometrics*. Plovdiv: Agricultural Univ.
- Dinić, B., Đorđević, N., (2005) *Preparing and utilization of silage*. Belgrade: Agricultural Research Institute.
- Dukić, D., (2002) *Biljke za proizvodnju stočne hrane*. Novi Sad: Poljoprivredni fakultet.
- Frakes, R.V., Davis, R.L., Patterson, F.L., (1961) *Crop Science*, 1(3), 207–209.
- Georgieva, N., Nikolova, I., (2012) Density and reduction of the stand at alfalfa varieties (*Medicago sativa* L.). *Banat's Journal of Biotechnology*, 3(2), 18–23. DOI:10.7904/2068 – 4738
- Jones, F.R., (1928) *J.Agr.Sci.* 75, 27-36 by author: Hanson, C.H. (1973) *Alfalfa science and technology*. Madison: American Society of Agronomy.
- Kephart, K.D., Twidwell, E.K., Bortnem, R., Boe, A., (1992) Alfalfa yield component responses to seeding rate several years after establishment. *Agronomy Journal*, 84(5), 827–831. doi:10.2134/agronj1992.00021962008400050013x
- Lloveras, J., Chocarro, C., Freixes, O., Arqué, E., Moreno, A., Santiveri, F., (2008) Yield, Yield Components, and Forage Nutritive Value of Alfalfa as Affected by Seeding Rate under Irrigated Conditions. *Agronomy Journal*, 100(1), 191–197. doi:10.2134/agrojn12006.0333
- Marinova, D., Petkova, D., (2010) Correlation dependences between green matter weight and yield components in alfalfa germplasms and their crosses. *Journal of Mountain Agriculture on the Balkans*, 13(4), 897–904.
- Nikolova, I., Georgieva, N., (2010) Tolerance of Lucerne varieties to *Apion seniculus* Kirby (Coleoptera: Curculionidae). *Agricultural Science and Technology*, 2(1), 44–47.

- Petkova, D., Marinova, D., (2006) Manifestation and stability of the most important traits of lucerne and relationships between them. *Plant Science*, 43(1), 78–80.
- Radović, J., Lugić, Z., Ignatović, S., Delić, D., (2004) Prinos i kvalitet suve materije genotipova lucerke (*Medicago sativa* L.) različitog porekla. *Acta Agriculturae Serbica*, 9(17), 109–115.
- Rumbaugh, M.D., (1963) Effects of population density on some components of yield of alfalfa. *Crop Sci.* 3(5), 423–424.  
doi:10.2135/cropsci1963.0011183X000300050016x
- Sauvant, D., Perez, J.M., Tran, G., (2002) Table de composition et de valeur nutritive des matières destinées aux animaux d'élevage: porcs, volailles, bovins, ovins, caprins, lapins, chevaux, poissons. Paris: INRA.
- Stanisavljević, R., Tomić, Z., Lugić, Z., Milenković, J., Đokić, D., (2008a) Yield and nutritive value of alfalfa cultivars sown at different densities. *Biotechnology in Animal Husbandry*, 24(3-4), 147–156. DOI:10.2298/BAH0804147S
- Stanisavljević, R., Milenković, J., Đokić, D., Štrabanović, R., Vasić, T., (2008b) Yield, yield components and forage quality of alfalfa varieties and their correlation dependence. *Journal of Mountain Agriculture on the Balkans*, 11(5), 896–908.
- Volenec, J.J., Cherney, J.H., Johnson, K.D., (1987) Yield components, plant morphology, and forage quality of alfalfa as influenced by plant population. *Crop Science*, 27(2), 321–326.  
doi:10.2135/cropsci1987.0011183X002700020040x