Journal of Agricultural Sciences
Vol. 53, No 2, 2008

UDC: 582.542.11 : 632.51 : 631.84
Original scientific paper

THE EFFECT OF CROP DENSITY AND APPLIED NITROGEN ON THE INTERACTION BETWEEN *Lolium italicum* AND *Galium aparine*

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Abstract: The effect of different densities of Italian ryegrass (*Lolium italicum* L.: G_1- 5 kg/ha seeds, 60 cm row spacing; G_2- 5 kg/ha seeds, 20 cm row spacing; G_3- 20 kg/ha seeds, 60 cm row spacing, and G_4- 20 kg/ha seeds, 20 cm row spacing) and different rates of applied nitrogen fertilizer (N_1- 0 kg N/ha; N_2 - 50 kg N/ha; N_3- 100 kg N/ha and N_4- 150 kg N/ha) on SPAD values under interaction between the crop and cleavers (*Galium aparine* L) was investigated. A trifactorial trial (4 sowing rates x 4 fertilization rates x 4 sampling times) with three replications was set up in a random block design with plot size 10 m^2 on a anthropogenious soil in the Šabac area in 2005/2006. SPAD was measured at 30 random plants of *L. italicum* and *G. aparine* on each plot four times during vegetation (V_1- tillering; V_2 – stem extension; V_3- flagleaf, and V_4- earing), and nitrogen content in plants was then computed. Relative nitrogen content in plant material was used as a parameter of competitive interaction between the cultivated and weed species. Total nitrogen in plant material was also analyzed. The data acquired showed that SPAD values were normally higher in *L. italicum* than in *G. aparine* under almost all crop densities and nitrogen rates applied, and maximum values were recorded in the third growing stage (flagleaf) at G_3 density and treatment rate of 100 kg N/ha. The highest biomass yield (6 033.0 kg/ha) and seed yield (1 129.0 kg/ha) of Italian ryegrass were achieved in the same variants, i.e. at crop density of 5 kg/ha seeds, 60 cm row spacing and 100 kg N/ha fertilizer, and the crop developed in competition with the weed species *G. aparine* at an average population density of 15-25 plants/m^2.

Key words: competition, density, *Galium aparine*, *Lolium italicum*, SPAD-502, nitrogen.

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Introduction

One of the central issues in fighting against weeds is development of acceptable weed control strategies that would reduce the use of herbicides. Elements of such a strategy can also be found in the competitive capacities of cultivated crops against weed plants (Liebman et al., 2001). In that context, optimal fertilization of crops may reduce the competitive weed vs crop interaction (DiTomaso, 1995), giving the crop a slight advantage in terms of utilization of soil nutrients. Nitrogen may significantly influence the yield of a crop (Camara et al., 2003), but it does not mean that by simply increasing nitrogen content in a soil it would be possible to interfere with the competitive interaction between the crop and the weed. This is all the more true as many weed species are remarkable nitrogen consumers (Hans and Johnson, 2002), so that in a situation of high infestation there would remain less nitrogen in soil for the crop, thus leading to its decreased yield. Weeds not only reduce the amount of nitrogen available to cultivated plants, their growth is often improved under conditions of elevated nitrogen contents in soil (Supasilapa et al., 1992). Hitherto research has proved that in terms of growth and development many weed species depend more heavily on nitrogen content in soil than crop plants (Blackshaw et al., 2003). Nitrogen fertilization can therefore often increase the competitive capacities of weeds more than the crop would benefit from it (Ampong-Nyarko and DeDatta, 1993). Considerations of the relative competitiveness of species are therefore associated with the competitors' strength, i.e. their biological and ecological properties. For example, wheat is known to be a weaker competitor than weeds under conditions of high nitrogen contents in soil (Taylor, 1999).

Galium aparine L., a frequent and damaging weed species in Serbia, especially in small crops in which it causes the highest losses (Vrbničanin and Šinžar, 2003). The weed is widespread throughout Europe from northern Norway (70 N°) to southern parts of the Mediterranean (40 N°). It is an early spring species germinating and emerging in autumn (IX-X) or in spring at temperatures of 6-12 ºC, and it flowers and bears fruit from June to October. A special negative effect of *G. aparine* is its ability to stick to the above-ground parts of cultivated plants and thus suffocate them and make harvest difficult by clogging the machinery, and it contaminates the harvested material (its seeds/fruits remain in the harvested crop) (Froud-Williams, 1985). *G. aparine* is a major consumer of soil nutrients, especially of nitrogen and phosphorus (Taylor, 1999). It is a good indicator of high availability of nitrogen in soils, which is further indicated by the index of trophic regime of habitats, which is 5 on a 1-5 scale, a value shared by species that are indicators of highly fertile soils (Kojić et al., 1997).

Italian ryegrass (*Lolium italicum* L. syn *L. multiflorum* L.) is a bushy annual grass originating from southern Europe. Its cultivation began in Italy in the 13th century. In Serbia it is a dense-growing winter crop characterized by fast growth that secures quick tillering, high yielding potential, fitness for reduced cultivation, good adaptability to heavy and moist soils, etc. Annual demands for Italian ryegrass seeds in Serbia are around 200 t, and 50% and provided from domestic sources (Nikitović and Radenović, 1996). Seed production of this species is one of the most cost-efficient branches of industrial grass seed production (Young et al., 2001). Nitrogen is a primary cause of limited growth as well as forage and seed yields of ryegrass (Griffith and Chastain, 1997). In Italian ryegrass seed production, nitrogen applied at a rate of 80-150 kg N/ha during spring fertilization secures seed yields of 1200-2100kg/ha (Cookson, 2000). On the other hand, nitrogen overdose may cause crop lodging, extension of vegetation or dangerous leaching of nitrates into various ground waters (Griffith, 2000, Silberstein et al., 2002). Nitrogen fertilization stimulates tillering of Italian ryegrass, consequently affecting the crop vegetation area, and increasing intra- and inter-species competition (Jewiss, 1972, Simon and Lemaire, 1987).

Recent research has indicated a close association between chlorophyll content and content of nitrogen in the leaf (Nageswara et al., 2001). This is logical with regard to the fact that nitrogen is mostly present in the leaf in its chlorophyll, so that by measuring chlorophyll content we may also indirectly quantify a plant's nitrogen status (Moran et al., 2000). The use of SPAD meters for measuring chlorophyll and iron contents has been justified for several crops: rice (Takeba et al., 1990), maize (Dwyer et al., 1995), wheat, barley, triticale (Iqbal and Wright, 1997, Ponce, 1998, McCloskey t al., 1998, Spaner et al., 2005, Dhima and Eleftherohorinos, 2005), sugarbeet (Sexton and Carroll, 2002), Italian ryegrass (Sunaga et al., 2006), etc.

This research aimed to investigate the effect of sowing densities of Italian ryegrass (*L. italicum*) on the seed, and of different rates of nitrogen fertilization on the SPAD values of both the cultivated crop and the weed species *G. aparine*. Based on SPAD readings, it is possible to calculate the relative content of nitrogen in plants (leaves) and then proceed to consider the competitive relationship between the crop and wild-growing weed species, whose numbers will be approximately the same in all plots. Furthermore, we determined the yields and uptaken nitrogen contents at different sowing densities and different fertilization rates, which allowed us to estimate an optimal sowing density of the crop and optimal nutrition under given agro-ecological conditions.

Material and Methods

Details of the experiment:

A field trial was set up on a anthropogenious soil at a private farm in north-western Serbia (Štitar village, N 44º 47´ 53"; E 19º 35´ 60", 83 m al) in 2005/2006. Before setting up the trial, the soil was analyzed for its main chemical properties: pH (potentiometric measurements with glass electrode), humus content (dichromatic method by Tjurin, Mineev et al., 2001), total nitrogen (semi-micro Kjeldahl method, Bremner, 1996) and content of available nitrogen (Bremner method, 1965). The main agrochemical characteristics of the soil on which the trial was set up are presented in Table 1.

Tab. 1. – The main agrochemical characteristics of soil before trial set-up

Site	Depth (cm)	pH (KCl)	Humus (%)	Total N (%)	Available N (ppm)		
					NH_4-N	NO_3-N	Sum
Štitar	0 - 40	5,40	2,80	0,20	13.9	8,7	22,6

Regarding soil pH, the tested soil belongs to the category of acid soils, and it is medium humic regarding humus content, and it falls in a group of soils well supplied in total nitrogen. The soil is also well provided with available nitrogen, primarily ammonia, which is consistent with the pH of the examined soil (Haynes, 1986).

The cultivated crop, a tetraploid Italian ryegrass (*Lolium italicum* L.) cv. Tetraflorum, was sown in the autumn of 2005 at four sowing rates (two seed rates x two sowing densities). The weed species, cleavers (*Galium aparine* L.), was undercropped in the spring of 2006 as a competitor species over soil nutrients (nitrogen). Cleavers density varied from plot to plot, ranging between 15 and 25 (maximum 35) plants per m^2. Spring nutrition with calcium ammonium nitrate (CAN) was carried out in March 2006. The data on sowing time of Italian ryegrass, sowing rates, nitrogen fertilizer rates, abundance of the weeds species *G. aparine* in plots, times of SPAD readings, times of plant sampling for laboratory analysis (determination of nitrogen content in leaves) and harvest date are shown in Table 2. A tri-factorial trial (4 sowing rates x 4 fertilization rates x 4 sampling times) was set up as a random block design on 10 m^2 plots (2.5 x 4 m). All treatments included three replications.

Tab. 2. – Details of the experiment

Sowing date	October 2005
Crop density (G) (kg seeds x cm row spacing)	G_1- 5 kg/ha seeds, 60 cm row spacing G_2- 5 kg/ha seeds, 20 cm row spacing G_3- 20 kg/ha seeds, 60 cm row spacing G_4- 20 kg/ha seeds, 20 cm row spacing
Galium aparine density (plants/m^{-2})	15-25 (max 35)
Sampling time (V)	V_1- tillering V_2- stem extension V_3- flagleaf V_4- earing
Spring nitrogen application (kg N/ha, March 2006)	N_1- 0 kg N/ha N_2- 50 kg N/ha N_3- 100 kg N/ha (CAN, 27% N) N_4- 150 kg N/ha

Harvest date End of June 2006

Meteorological data for the trial area were provided from an automatic meteorological station in Sremska Mitrovica (Table 3)

Tab. 3.- Precipitation and average temperatures during the experiment period 2005-2006

2005/06	Sept	Oct	Nov	Dec	Jan	Feb	March	April	May	June	July	Aug	
Temperature	18.0	13.0	5.3	2.3	-1.0	1.3	5.7	13.3	16.7	19.7	23.3	20.0	11.5
Precipitation	38	8	20	62	30	40	62	63	33	104	38	154	652
rainy factor	2.1	0.6					10.9	4.7	2.0	5.3	1.6	7.7	

Monthly precipitation and monthly air temperature means are presented only for the trial months.

Parameters measured:

A nondestructive optical method based on light absorption and/or reflection by intact plants/leaves was used (Richardson et al., 2002). A hand-held chlorophyll meter (SPAD-502, Minolta Corp., Ramsey, NJ) was used for measuring light absorption at different wave lengths: 660 nm and 940 nm (Nageswara et al., 2001). Red light is absorbed exclusively by chlorophyll, while near infrared light is the "recommended wavelength" used for adjusting the differences in leaf structure. Based on SPAD readings that represent the light absorbed by chlorophyll (i.e. chlorophyll concentration in leaves) and using a standard curve, it was possible to determine nitrogen content in plants since nitrogen accounts for most of the chlorophyll molecules. Thus, the SPAD chlorophyll meter can indirectly determine nitrogen status in a plant.

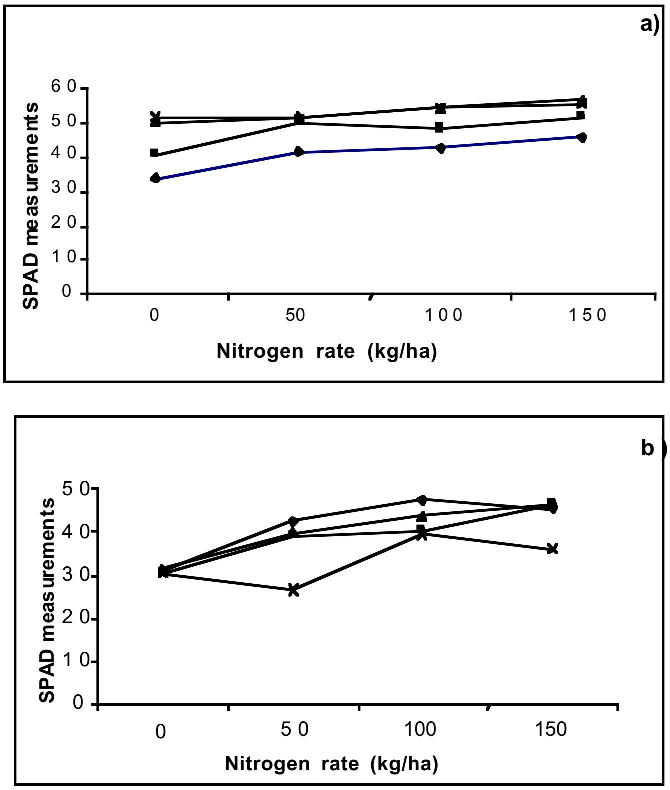
SPAD readings were carried out on leaves of the second, third and fourth nodes from the tip of the main shoot axis on 30 randomly chosen plants. After SPAD readings, 10 leaves were removed from each plant, the leaves were dried in a dryer at 80 °C/48 h, and their dry weight was measured and nitrogen content determined. The Kjeldahl method was used for determining total nitrogen content in plants, and plant samples were digested in H_2SO_4 in the presence of the catalytic mixture K_2SO_4 : $CuSO_4$: Se = 100 : 10 : 2. Distillation was carried out with 33% NaOH and titration with 0.005 M H_2SO_4 (Musinger and McKinney, 1982). A standard curve was constructed based on SPAD readings and nitrogen values from laboratory analysis. For making the standard curve, random SPAD readings were made for each replication in each variant. Based on standard curve-sand "n" SPAD readings, it is possible to evaluate nitrogen status in a plant, which is especially convenient for fast screening in the field in order to check nitrogen availability to plants.

Data analysis:

Experimental data were statistically processed by ANOVA methodology and the differences between treatments investigated were tested with t-test at 0.05, 0.01 and 0.001 levels of significance.

Results and Discussion

Figures 1a and b show the average SPAD values for crop density G_1 (5 kg/ha seeds and 60 cm row spacing) at each of the four stages in which measurements were made (from tillering to earing) for *L. italicum* and *G. aparine* depending on nitrogen rates applied.



◆ *V1-Tillering* ■ *V2-Stem extension* ▲ *V3-Flagleaf* X *V4-Earing*

Fig. 1a,b. – Average SPAD readings for L. italicum and G. aparine depending on nitrogen rate at G_1 crop density

The results show that under G_1 density and growing nitrogen rates, SPAD values of *L. italicum* had an increasing trend at all readings, except at stem extension (V_2) and under 100 kg N/ha rate, when SPAD was on a slight decline. The highest SPAD value was measured under 150 kg N/ha rate in the flagleaf stage (V_3, 56.82±5.19). Also, the highest statistical difference regarding SPAD readings under rising nitrogen rates, compared to control, were found for the rate 150 kg N/ha throughout the vegetation season (Tab. 4). At 50 and 100 kg N application rates, significant statistical differences for SPAD readings against control were most evident in the tillering and stem extension stages (V_1 and V_2). From the aspect of SPAD readings therefore, the rate of 150 kg N/ha was adequate for the crop species *L. italicum*.

Tab. 4. – Statistical significance of the differences in SPAD readings between fertilized and non-fertilized crop variants (density 1)

Control: Fertilization rate	Crop stage	SPAD of *L. italicum*	SPAD of *G. aparine*
0 : 50 kg N/ha	tillering	6.75 **	8.95**
	stem extension	8.88**	6.91**
	flagleaf	0.98 ns	5.76**
	earing	0.37 ns	2.88*
0 :100 kg N/ha	tillering	7.70**	18.12***
	stem extension	6.62**	8.90**
	flagleaf	4.36*	6.50**
	earing	1.95 ns	8.51**
0 : 150 kg N/ha	tillering	10.87**	11.07**
	stem extension	10.92**	12.82***
	flagleaf	6.98**	8.38**
	earing	2.56*	4.41*

* - statistical significance at =0.05 risk; ** - statistical significance at = 0.01 ;
*** - statistical significance at = 0.005 ; ns - no statistical significance

At the same crop density (G_1), SPAD values of *G. aparine* at each time of measurement and applied rate of nitrogen were lower than those of *L. italicum* (Fig. 1b). As in *L. italicum*, the highest SPAD was found in the fourth reading for 150 kg N/ha rate (46.49±5.62). With increasing fertilizer rates SPAD values also increased in the second and third measurements, while in the first and fourth readings SPAD increased only as far as 100 kg N/ha rate, while at 150 kg N/ha it had a decreasing trend. Evidently, SPAD values of *G. aparine* in control were lower in all four readings than those of *L. italicum*. In terms of absolute values, these readings were approximately the same, unlike the SPAD values of the crop, which increased from tillering to earing, at which point the highest SPAD value was measured for *L. italicum* (51.60) and lowest for *G. aprine* (30.04). Therefore, in untreated control, the crop showed a higher ability than the weed species to uptake nitrogen, and formed greater leaf area and showed itself a better competitor than the weed, which is probably associated with earlier tillering of Italian ryegrass and its better nitrogen uptake (P o n c e, 1998). Statistical analysis confirmed significant differences in SPAD values between the fertilized variants and control in all stages of growth, but high nitrogen contents in soil evidently did not increase SPAD values in the weed species *G. aparine*.

SPAD readings for the second crop density G_2 (5 kg/ha seeds and 20 cm row spacing) at all stages of growth of both species and different nitrogen rates are shown in Figures 2a and b.

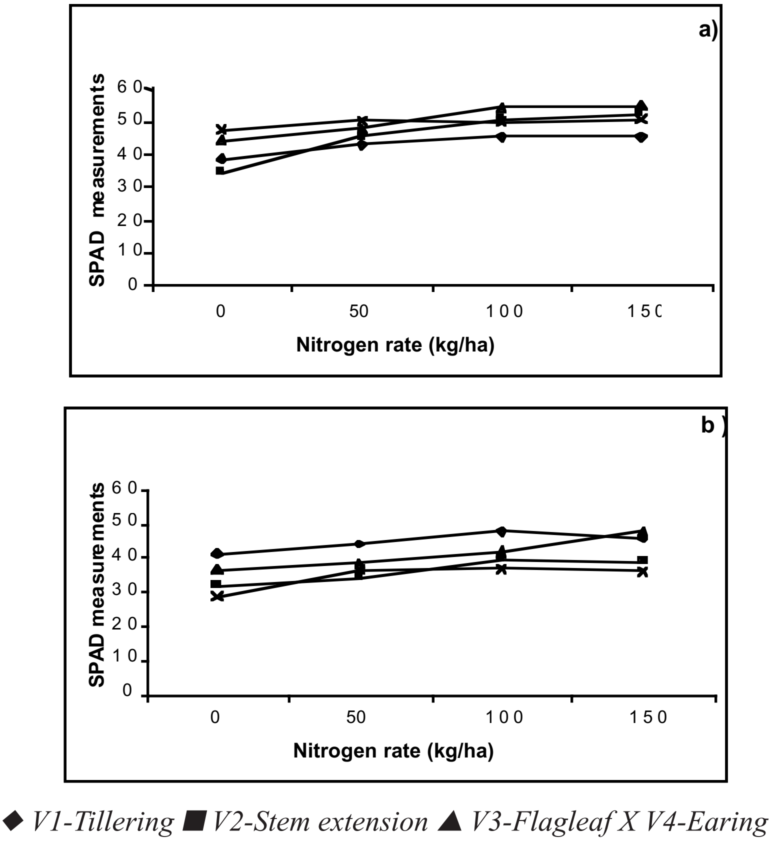


Fig. 2a,b.- Average SPAD readings for L. italicum and G. aparine depending on nitrogen rate at G_2 crop density

At crop density G_2, SPAD values of *L. italicum* increased in all measurement stages with the applied nitrogen rates, except in the tillering stage in which SPAD values slightly decreased under the highest fertilization rate (150 kg N/ha), and in the flagleaf stage under the rate of 100 kg N/ha. The highest SPAD value was found in the flagleaf stage (V_3) at 150 kg N/ha rate (54.50±5.23). Also, statistically significant differences were found for this parameter under application of increasing nitrogen rates against control with the highest differences showing at 150 (r= 21.05***) and 100 kg N/ha (r= 16.69***) in the stem extension stage (V_2) (Tab.5). Regarding the weed species *G. aparine,* SPAD values in the first measurement under all applied nitrogen rates were higher than they were for *L. italicum*, but the trend did not hold in subsequent readings (Fig. 2b).

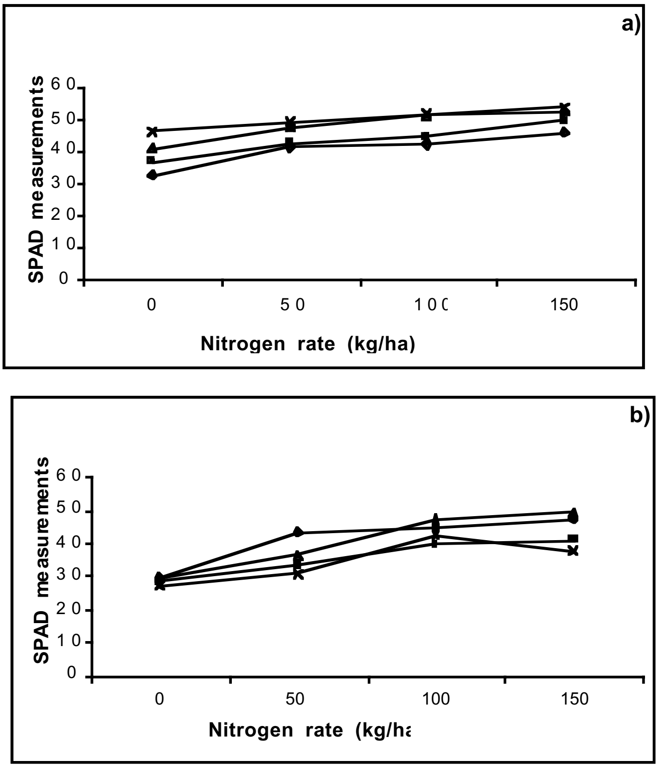
Tab. 5. – Statistical significance of the differences in SPAD readings between fertilized and non-fertilized crop variants (density 2)

Control: Fertilization rate	Crop stage	SPAD of *L. italicum*	SPAD of *G. aparine*
0 : 50 kg N/ha	tillering	5.29***	5.21**
	stem extension	10.93**	2.25*
	flagleaf	3.94*	1.55^{ns}
	earing	2.41*	6.39**
0 : 100 kg N/ha	tillering	6.56**	5.82**
	stem extension	16.69***	8.21**
	flagleaf	10.56**	2.59*
	earing	1.90^{ns}	4.83*
0 : 150 kg N/ha	tillering	6.81**	5.65**
	stem extension	21.06***	6.00**
	flagleaf	8.89**	7.46**
	earing	2.80*	4.39*

* - statistical significance at =0.05 risk ; ** - statistical significance at = 0.01 ;
*** - statistical significance at = 0.005 ; ns - no statistical significance

Figure 2b shows that the highest SPAD value of *G. aparine* was found in the third reading (flagleaf stage) for the application rate of 150 kg N/ha (47.97). Statistically significant differences in SPAD values for this species under growing nitrogen rates and in control were found in nearly all measurements, but the coefficients of correlation were lower than they were for the crop species. Thence, *G. aparine* had a weaker reaction to fertilization at the same crop densities.

Under the third crop density G_3 (20 kg/ha seeds and 60 cm row spacing), SPAD readings were rising for Italian ryegrass with increasing nitrogen rates at all stages of growth (Fig. 3a), and the highest value was recorded under 150 kg N/ha rate in flagleaf stage (53.71±3.66). The significance of differences between SPAD values measured in the fertilized and non-fertilized variants are shown in Table 6. The data for both plant species show statistically significant differences in SPAD values between the control and the variants of growing nitrogen rates.



◆ *V1-Tillering* ■ *V2-Stem extension* ▲ *V3-Flagleaf X V4-Earing*

Fig. 3a,b. – Average SPAD readings for L. italicum and G. aparine depending on nitrogen rate at G_3 crop density

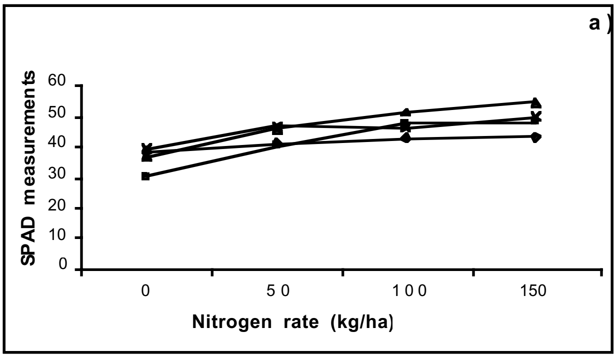
Tab. 6. – Statistical significance of the differences in SPAD readings between fertilized and non-fertilized crop variants (density 3)

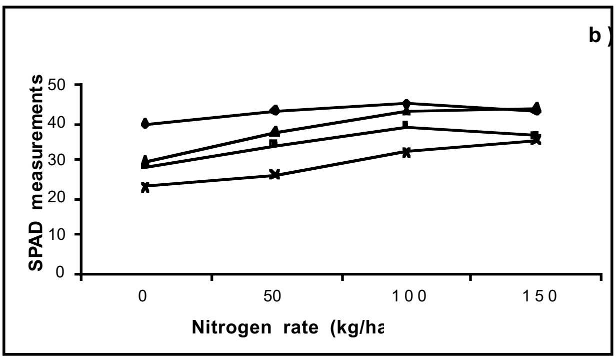
Control: Fertilization rate	Crop stage	SPAD of *L. italicum*	SPAD of *G. aparine*
0 : 50 kg N/ha	tillering	8.46**	12.46***
	stem extension	5.04**	4.12*
	flagleaf	6.06**	5.00**
	earing	2.19*	2.78*
0 : 100 kg N/ha	tillering	9.29**	14.74***
	stem extension	6.35**	9.59**
	flagleaf	10.72**	11.70**
	earing	3.78*	9.84*
0 : 150 kg N/ha	tillering	12.85***	16.58***
	stem extension	10.03**	12.91***
	flagleaf	12.24***	15.54***
	earing	6.62**	6.28**

* - statistical significance at =0.05 risk ; ** - statistical significance at = 0.01 ; *** - statistical significance at = 0.005 ; ns - no statistical significance

SPAD values of *G. aparine* at G_3 density in all measurement stages were growing with the growth of fertilization rate, except in the fourth measurement in which a lower value was read using150 kg N/ha rate (Fig. 3b). The highest SPAD value was read in the flagleaf stage using 150 kg N/ha rate (48.86±6.51). Similar to the former variants, statistically significant differences in SPAD values were recorded between the fertilized and non-fertilized variants, and the differences were higher under 150 kg N/ha rate. Also, the lowest SPAD values of *G. aparine* were found at this crop density compared to all densities in control plots, and they showed little difference regarding measurement stage.

Figures 4 a and b show SPAD values at crop density G_4 (20 kg/ha seeds and 20 cm row spacing) for both plant species. Under this density, SPAD readings under rising fertilization rates had a rising trend for *L. italicum*. The highest SPAD value was found in the flagleaf stage under 150 kg N/ha rate (54.70±4.98). Also, data for the fertilized and control variants show statistically significant difference (Table 7).





◆ *V1-Tillering* ■ *V2-Stem extension* ▲ *V3-Flagleaf X V4-Earing*

Fig. 4a,b. – Average SPAD readings for L. italicum and G. aparine depending on nitrogen rate at G_4 crop density

SPAD values of *G. aparine* in the fourth variant of crop density were higher in the first and second measurements when nitrogen rates were lower (50 and 100 kg N/ha). Such a trend was not recorded in the third and fourth measurements, i.e. SPAD values increased with nitrogen increase in the stages of flagleaf and earing. The highest SPAD value of this species was found under 100 kg N/ha rate in the first measurement stage (44.90). Statistical analysis confirmed significant differences between the fertilized and non-fertilized variants, which was especially evident under treatment with 100 kg N/ha (Table 7). However, coefficients of correlation were far lower than they were for *L. italicum*. We may therefore say that *G. aparine* had a weaker reaction to the fertilizer applied, i.e. the crop better utilized the available nitrogen.

Tab. 7.- Statistical significance of the differences in SPAD readings between fertilized and non-fertilized crop variants (density 4)

Control: Fertilization rate	Crop stage	SPAD of *L. italicum*	SPAD of *G. aparine*
0 : 50 kg N/ha	tillering	2.23*	3.22*
	stem extension	8.22**	0.00ns
	flagleaf	7.48**	5.83**
	earing	5.99**	2.28*
0 : 100 kg N/ha	tillering	4.04*	5.39**
	stem extension	15.92***	9.54**
	flagleaf	12.86***	10.94**
	earing	5.28**	6.60**
0 : 150 kg N/ha	tillering	4.99*	3.37*
	stem extension	18.42***	8.09**
	flagleaf	16.58***	11.68**
	earing	8.29**	8.78**

* - statistical significance at =0.05 risk; ** - statistical significance at = 0.01 ; *** - statistical significance at = 0.005 ; ns - no statistical significance

In order to outline a pattern by which one of the two species (*L. italicum* or *G. aparine*) has put available nitrogen to better use, and which is directly correlated with SPAD values, we calculated the average SPAD values (based on 160 measurements in all stages of growth) for different crop densities and rates of nitrogen treatment (Table 8). The results show that the highest average SPAD values for both plant species were found in the variants with top nitrogen rate, meaning that this level of fertilization caused the highest intensity of leaf «greenness», which is positively correlated with chlorophyll content in leaves (Spaner et al., 2005). Those values were higher for the crop species *L. italicum* at all densities and nitrogen rates. In this context, it is worth noting that some authors consider weeds better competitors than crops over nitrogen (Ampong-Nyarko and DeDatta, 1993, Taylor, 1999), which means that different factors, such as agro-ecological conditions, cultivation, fertilization and biological-ecological characteristics of species, can determine which species will be the stronger competitor (Ruiz et al., 2008).

Tab. 8. – Average SPAD values of L.italicum and G. aparine at different densities and nitrogen rates

Crop density	Control	50 kg N/ha	100 kg N/ha	150 kg N/ha
		L . italicum		
G_1	43.94	48.52	49.89	52.41
G_2	41.04	46.53	50.00	50.75
G_3	39.07	44.96	47.36	50.21
G_4	36.07	43.23	46.78	48.71
		G. aparine		
G_1	30.59	37.16	43.06	43.65
G_2	34.60	38.28	41.44	42.23
G_3	28.65	36.03	43.49	43.75
G_4	29.90	33.64	39.62	39.63

A high correlation was detected between SPAD values and nitrogen rates applied to *L. italicum* at all crop densities (G_1- 0.97**; G_2- 0.95**; G_3- 0.98**; G_4- 0.96**). Based on that correlation, we may conclude that the applied nitrogen rates had immediate effect on SPAD values.

The highest SPAD readings for *L. italicum* at all nitrogen rates applied were found under crop density G_1 (5 kg/ha seeds and 60 cm row spacing), which may relate to the lowest intraspecies shading enabling the best utilization of light. Also, the effect of crop density on SPAD values becomes evident by comparing SPAD values from the two extreme densities (G_1 and G_4) under all levels of fertilization of both plant species (Table 8). Besides, the effect of crop density on SPAD readings can also be assessed by comparing the same sowing rates (same amounts of seeds) and different row spacing (5 kg seeds at 60 and 20 cm row spacing; and 20 kg seeds at 20 and 60 cm row spacing), showing that SPAD values tend to be regularly higher at higher row spacing. Therefore, SPAD values were primarily affected by crop density.

Although the average SPAD values of *G. aparine* at all crop densities and nitrogen rates were lower than those of *L. italicum*, a high correlation between SPAD values and fertilization rates was detected in the former species too (G_1- 0.95**, G_2- 0.97**, G_3- 0.95**, G_4- 0.95**). So, in both species a high correlation was found between the applied nitrogen rates and average SPAD values at all crop densities, only the values were higher for *L. italicum*. To some degree, it may be related to crop uniformity and physical dominance of the crop, i.e. the interspecies competition in which the crop was shading *G. aparine*. Therefore, SPAD values of *G. aparine* were directly affected by crop density so that the highest values were recorded at lowest densities, i.e. at the highest row spacing and a same amount of seeds. In contrast to the crop *L. italicum,* where this regularity was quite evident, slight deviations occurred in this case at G_1 and G_2 densities and low nitrogen rates (control and 50 kg N/ha), where SPAD values were higher under higher density.

Table 9 shows the yields of Italian ryegrass (biomass and seed yield) and outtake of nitrogen at different rates of fertilization.

Tab. 9. – Yield of L. italicum and outtake of nitrogen at different crop densities and rates of nitrogen supply

Fertilization rate (kg N/ha)	Biomass kg/ha	Seed yield kg/ha	Nitrogen outtake kg N/ha
		G_1	
control	2 300	591.3	31.74
50	2 500	594.3	53.25
100	5 150	922.3	105.40
150	3 550	741.0	94.43
		G_2	
control	2 700	693.0	41.04
50	2 925	759.0	58.50
100	3 725	865.0	82.32
150	3 700	803.0	97.31
		G_3	
control	4 550	849.0	63.24
50	4 900	920.0	81.83
100	6 033	1 129.0	166.51
150	5 500	968.0	130.30
		G_4	
control	3 258	911.0	40.40
50	4 733	941.0	71.47
100	5 533	953.3	135.0
150	4 900	911.0	139.16

It is characteristic of all tested crop densities that by increasing nitrogen rate the yield is also increased as far as 100 kg N/ha rate. Consequently, the rate of 150 kg/ha cannot be considered adequate for all crop densities (Griffith, 2000). The highest biomass and seed yield were achieved at G_3 density at all levels of fertilization, as well as in control plots. Also, at this crop density the highest yield was achieved under fertilization rate of 100 kg N/ha, when the outtake of soil nitrogen was also highest (166.51 kg/ha, Tab. 9). Therefore, an optimal fertilization of a crop makes it possible to reduce the weed-crop competitive interaction (DiTomaso, 1995), i.e. to secure that the crop has a slight advantage in terms of utilization of soil nutrients. Furthermore, it is evident that the yield achieved was in correlation with SPAD readings at the same sowing rates, i.e. at 20 kg/ha sowing rate the yield was higher when row spacing was 60 cm and SPAD values were also higher (Table 8 and 9), compared to row spacing of 20 cm under the same sowing rate. It means that under the same fertilization rates the yield directly depended on crop density, which indirectly made plants uptake the highest amount of nitrogen at G_3 density. In terms of seed yield, there is the same regularity as we have observed for biomass yield. Under the highest fertilization rate (150 kg N/ha) and all crop densities, biomass yield dropped, and so did seed yield. This fact may be related to an intensive vegetative growth under high nitrogen fertilizer rates, and crop lodging and reduced seed yield (Griffith, 2000, Silberstein et al., 2002).

Conclusion

The data presented here suggest the following conclusions:

At all investigated crop densities, SPAD values were regularly highest under maximum rates of applied nitrogen (150 kg N/ha) in species, *L. italicum* and *G. aparine*.

SPAD values were mostly highest in the third reading, which is the growth stage of flagleaf.

SPAD values were regularly higher for *L. italicum* (V_1=56.82, V_2=54.51, V_3=53.72, V_4=54.70) than *G. aparine* (V_1=46.49, V_2=47.97, V_3=48.86, V_4=43.98) at the same crop densities, nitrogen rates and stages of growth.

A high correlation was detected in both plant species between the applied nitrogen rates and average SPAD values at all crop densities, but the values were higher for *L. italicum*.

At all crop densities, it was characteristic that by increasing nitrogen rate the yield also increased as far as 100 kg N/ha rate; therefore, a 150 kg/ha rate cannot be considered adequate under any of the crop densities tested.

The highest crop yield and nitrogen outtake was achieved at 20 kg sowing density, row spacing of 60 cm and fertilization rate of 100 kg/ha, in which variant the Italian ryegrass crop developed in competition with the weed species *G. aparine* growing at population density of 15-25 plants/m^2.

Acknowledgements

The authors would like to thank students who had worked hard to collect data presented in this paper.

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Received: Maj 09, 2008
Accepted: Juli 10, 2008

UTICAJ GUSTINE USEVA I PRIMENJENOG AZOTA NA INTERAKCIJU *LOLIUM ITALICUM* I *GALIUM APARINE*

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R e z i m e

Ispitivan je uticaj različitih gustina setve italijanskog ljulja (*Lolium italicum* L.: G_1- 5 kg ha^{-1} semena, 60 cm međuredno rastojanje; G_2- 5 kg/ha semena, 20 cm međuredno rastojanje; G_3- 20 kg/ha semena, 60 cm međuredno rastojanje i G_4- 20 kg/ha semena, 20 cm međuredno rastojanje i različitih doza azotnog đubriva (N_1- 0 kg N/ha; N_2- 50 kg N/ha; N_3- 100 kg N/ha i N_4- 150 kg N/ha) na SPAD vrednosti pri interakciji useva i lepljive broć (*Galium aparine* L). Trofaktorijalni ogled (4 norme setve x 4 nivoa đubrenja x 4 vremena uzorkovanja) u tri ponavljanja je zasnovan po slučajnom blok sistemu sa elementarnom parcelom veličine 10 m^2 tokom 2005/2006. godine na području Šapca na antropogeniziranom zemljištu. Četiri puta tokom vegetacione sezone (V_1- faza bokorenja; V_2- faza vlatanja; V_3- faza zastavičara i V_4- faza klasanja) merena je SPAD vrednost na 30 nasumično odabranih biljaka *L. italicum* i *G. aparine* na svakoj parceli, a potom je indirektno računat relativni sadržaj azota u biljkama. Vrednosti relativnog sadržaja azota u biljnom materijalu je korišćen kao parametar kompetitivne interakcije između gajene i korovske vrste. Takođe, rađena je analiza sadržaja ukupnog azota u biljnom materijalu. Dobijeni rezultati su pokazali da je SPAD vrednost gotovo pri svim gustinama setve useva i primenjenim dozama azota bila redovno veća kod *L. italicum* u odnosu na *G. aparine*, s tim što su maksimumi izmereni u trećem vremenu (faza zastavičara) pri gustini G_3 i dozi od 100 kg N/ha. Pri istim varijantama, tj. gustini useva od 5 kg/ha semena i 60 cm međuredno rastojanje i dozi đubriva od 100 kg N/ha je postignut i najveći biološki prinos (6 033.0 kg/ha) i prinos semena (1 129.0 kg/ha) italijanskog ljulja koji se razvijao u kompeticiji sa korovskom vrstom *G. aparine* pri gustini populacije od 15 do 25 biljaka/m^2 u proseku.

Primljeno: 09 maj 2008
Odobreno: 10 jul 2008

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