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ORIGINAL PAPER

THE FERTILIZER VALUE OF POST-HARVEST RESIDUES OF *TRIGONELLA FOENUM-GRAECUM* L.*

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Abstract

Legume plants are environmentally friendly because of the low-cost cultivation costsas well as the good quality of post-harvest residues. The aim of this study was to determine the fertilizer value of post-harvest residues of fenugreek plants grown under various environmental conditions. The value of the aerial parts of fenugreek plants left in the soil after harvest was analyzed in a pot experiment with different seed inoculation treatments (*Rhizobium meliloti* and not inoculated), different doses of K fertilizer (0.0; 1.25; 2.50; 3.75 g K per pot) and different soil moisture content (50-60% and 30-35%) during the growing season. K fertilization and water stress differentiated the dry weight of the aerial parts of fenugreek plants. Water deficit during the flowering and pod formation phases significantly decreased the dry weight of pericarps, leaves and stems, which was lower by about 17, 3 and 14%, respectively. An increase in a K dose led to a decrease in the concentrations of N and Ca in pericarps, N, P, Ca and Mg in leaves, and Na, Ca and Mg in stems. Seed inoculation increased N (11.5%) and Ca (7.2%) accumulation in straw. Water stress modified the uptake of minerals by decreasing the accumulation of Mg, Ca and K. Biological N_2 fixation was influenced by a dose of K fertilizer and water availability. Biological N_2 fixation by *R. meliloti* root nodule bacteria ranged from 27.96 to 32.08 µg per plant. Nitrogen was most effectively fixed in the treatment supplied with 2.50 g K per pot. Water deficit during flowering and pod formation decreased biological N_2 fixation by 4.5%.

Keywords*:* fenugreek, biologically N_2 fixed, macronutrients.

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INTRODUCTION

The goal of sustainable development is to promote food, environmental and energy security. Plants of the botanical family *Leguminosae* play an important role in this model of development in the European Union, including Poland. One of the aims of the Polish policy in this aspect is to create a supportive environment for improving the food and feed protein security (Resolution of the Council of Ministers 2009). Legume plants are capable of fixing atmospheric nitrogen, which lowers greenhouse gas emissions during the production of N fertilizers (Święcicki et al. 2007). The average greenhouse gas emissions associated with the production of N fertilizers in the EU is estimated at 58 860.6 g eq. CO_2 kg^{-1} of pure compound (Żak et al. 2014). Protein-rich plants are grown for seeds, but plant residues can be incorporated into soil after the main crop has been harvested (Hertog et al. 2011). Post-harvest residues can be simply defined as the aerial biomass of annual crop residues (Monreal et al. 2005). The cited authors calculated the aerial biomass by measuring the weight of residues left in the field after crop harvest. In fenugreek, the post-harvest residue of the aerial parts is straw, including pericarps, leaves and stems.

Post-harvest residues supply the soil with organic compounds and nutrients, and their dry weight is estimated at 1.5-8.0 ha^{-1}. Kristensen (2002) showed that the plant and the protein-rich fixed from 41 to 54 kg N per ton seed. Leguminous plans also contribute to a reduction in human labor, in particular in the cultivation of winter crops (Wani et al. 1995, Święcicki et al. 2007).

Legumes are also a source of nutritionally balanced hay and silage for livestock (Sadeghzadeh-Ahari et al. 2009). The content of mineral elements in the biomass of agricultural crops is modified mainly by agronomic and environmental factors (soil, climate and agricultural treatments) (Chojnacki, Boguszewski 1971). Lignified stems of fenugreek plants constitute biomass for energy generation purposes. The profitability of fenugreek production should be analyzed not only from the environmental point of view, but also based on agricultural and economic considerations. According to Bieńkowski et al. (2015), the production value of fenugreek in Poland ranges from 1760 to 1966.6 USD ha^{-1}. Knowledge of nutrient concentrations in the post-harvest residues of leguminous plants is essential in sustainable agriculture. Therefore, the aim of this study was to determine the fertilizer value of the post-harvest residues of fenugreek plants grown under various environmental conditions.

MATERIALS AND METHODS

Fenugreek plants were grown in a pot experiment in the greenhouse. The experimental factors were the doses of K fertilizer, seed inoculation and water stress during the growing season. Fenugreek plants were grown in light soil composed of heavy loamy sand (*Particle size distribution* … 2009) with pH of approximately 5.5 in 1 M KCl. The chemical composition of soil was as follows: P – 107, K – 124, and Mg – 19 mg kg^{-1} of soil. Potassium (K_2SO_4) fertilizer was applied at the following doses: 0 g pot^{-1} (K0, control), 1.25 g pot^{-1} (K1.25), 2.50 g pot^{-1} (K2.50) and 3.25 g pot^{-1} (K3.25). The following seed inoculation treatments were applied: (1) no inoculation (control) and (2) inoculation with *Rhizobium meliloti*. Water was supplied at: (1) 50-60% of available water capacity (control) where plants were watered every day, and (2) 30-35% of available water capacity (water deficit) between the flowering stage and the pod formation stage. Fenugreek plants were fertilized with 0.5 g N ($CO(NH_2)$, 0.5 g P ($Ca(H_2PO_4)$ H_2O) and 0.3 g Mg ($MgSO_4$ H_2O) (Żuk-Gołaszewska et al. 2015). For each combination there were 6 replications (pots) of the experimental factors. Each pot contained 9.5 kg of soil seeded with 10 plants. During the plant growing season no pests were observed.

Residual biomass (pericarps, leaves and stems) was mineralized in concentrated sulfuric acid (VI) with hydrogen dioxide as an oxidant (BÜCHI Speed Digester K – 439). Total N content was determined calorimetrically with hypochlorite (Baethgen, Alley 1989). Phosphorus (P) content was determined by the vanadium-molybdenum method. Colorimetric measurements were performed on a UV-1201V spectrophotometer, Shimadzu Corporation Kyoto, Japan. The concentrations of K, calcium (Ca) and sodium (Na) were determined by atomic emission spectrometry (AES) (Jenway LTD PFP 7, UK). Magnesium (Mg) content was determined by atomic absorption spectrometry (AAS) on a Shimadzu AA-6800 apparatus (Ostrowska et al. 1991).

The amount of biologically fixed N was calculated from the formula:

$$SNF = N_{aerial\ biomass} \cdot N_{fix}\ (1+N_{root+stubble} + N_{immobile}) \quad (1)$$

where:

SNF – the amount of N fixed symbiotically by fenugreek (mg per plant);

$N_{aerial\ biomass}$ – N accumulated in aerial biomass;

N_{fix} – 0.74 (similarly to peas) – percentage of symbiotically fixed N in the total N accumulated in the dry matter of aerial biomass;

$N_{root+stubble}$ – 0.25 (similarly to lupine) – percentage of N fixed symbiotically in roots and stubble in the total N accumulated in the aerial biomass of legumes;

$N_{immobile}$ – 0.25 (light soil) – percentage of symbiotically fixed N immobilized in soil organic matter in total symbiotically fixed N in the aerial biomass in the final growth stage (Hùgh-Jensen, Schjoerring 2001, Pietrzak 2011).

The results were processed statistically by analysis of variance, and the effects of the experimental factors on the analyzed parameters were inferred from the Snedecor's *F* distribution. Mean values were compared based on $HSD_{0.01}$ determined by the Tukey's test. All calculations were performed with the use of Statistica 12 software.

RESULTS

The aerial biomass of fenugreek plants is presented in Table 1. The total aerial biomass of fenugreek plants decreased significantly (by more than 12%) only in response to water stress (30-35% of available water capacity). The dose of K fertilizer and water availability during the growing season significantly differentiated the dry weight of aerial plant parts (pericarps, leaves and stems). Leaf and stem biomass was the highest in plants fertilized with 2.50 g K, and the pericarp mass was the highest in plants fertilized with 3.75 g K per pot. The dry weight of all aerial plant parts was significantly lower under water deficit (30-35% of available water capacity). In water-stressed plants, the dry weight of pericarps, leaves and stems was reduced by 17.07%, 3.3% and 13.85%, respectively.

In plants grown from inoculated seeds, only the dry weight of leaves was significantly greater than in plants grown from non-inoculated seeds, and it was determined at 408.9 mg DM per plant.

The nutrient content (N, P, K, Na, Ca, Mg) of fenugreek pericarps, leaves and stems is presented in Table 2. Leaves were most abundant in nutrients, as they contained around 20 g N, 8 g P, 7 g Na and 7 g Mg kg^{-1} DM. Leaves were also characterized by the highest content of K, which increased proportionally to the doses of K fertilizer. Potassium levels were

Table 1

Aerial biomass of fenugreek plants

Experimental factor	Pods	Leaves	Stems	Aerial biomass
	(mg DM per plant)			
K dose (g per pot)				
0.00	187.4	327.7	668.1	1273.6
1.25	242.2	388.6	743.1	1296.9
2.50	253.9	435.9	748.3	1359.3
3.75	266.2	399.8	689.7	1289.4
$HSD_{0.01}$	4.5	5.7	5.8	NS
Rhizobium meliloti				
Non-inoculated seeds	238.8	367.1	711.8	1278.4
Inoculated seeds	236.0	408.9	712.8	1331.2
$HSD_{0.01}$	NS	6.6	NS	NS
Available water capacity (%)				
50-60	259.5	394.5	765.3	1389.4
30-35	215.2	381.5	659.3	1220.3
$HSD_{0.01}$	5.3	6.6	6.7	31.5

NS – non-significant differences

Table 2

Macronutrient concentrations in the aerial biomass of fenugreek plants

Experimental factor	Pericarps						Leaves						Stems					
	(g kg^{-1} DM)																	
	N	P	K	Na	Ca	Mg	N	P	K	Na	Ca	Mg	N	P	K	Na	Ca	Mg
K dose (g per pot)																		
0.00	12.31	5.91	22.85	5.51	21.21	4.78	21.15	8.57	19.78	7.01	50.36	7.79	6.48	2.56	9.35	5.34	13.94	3.89
1.25	11.36	6.00	28.63	5.07	18.97	5.00	20.28	8.32	37.52	7.08	44.00	7.51	6.56	2.57	17.90	4.60	12.09	3.33
2.50	10.50	5.96	33.75	5.34	17.05	5.32	21.08	8.04	52.20	6.86	39.95	7.33	6.34	2.58	24.91	4.40	9.62	3.27
3.75	9.45	5.80	38.92	4.99	15.34	5.58	19.56	7.83	65.21	6.42	32.60	7.20	6.87	2.46	33.25	4.34	8.25	3.12
$HSD_{0.01}$	0.14	NS	0.49	NS	0.20	0.09	0.22	0.07	0.55	NS	0.37	0.07	0.10	0.03	0.34	0.14	0.13	0.05
Rhizobium meliloti																		
Non-inoculated seeds	10.56	6.08	31.60	5.17	17.75	5.46	20.04	8.56	45.37	6.92	41.70	7.69	6.09	2.54	21.79	4.71	10.69	3.42
Inoculated seeds	11.25	5.75	30.48	5.29	18.54	4.88	20.99	7.82	41.99	6.77	41.76	7.22	7.03	2.55	20.91	4.63	11.26	3.38
$HSD_{0.01}$	0.16	0.09	0.56	NS	0.23	0.10	0.26	0.08	0.64	NS	NS	0.09	0.12	NS	0.39	NS	0.16	NS
Available water capacity (%)																		
50-60	10.90	6.12	31.73	5.39	18.39	5.73	18.73	8.41	45.51	6.79	44.12	8.04	5.71	2.46	20.56	4.54	10.81	3.31
30-35	10.92	5.71	30.35	5.06	17.90	4.60	22.31	7.97	41.85	6.90	39.34	6.87	7.41	2.63	22.14	4.80	11.14	3.50
$HSD_{0.01}$	NS	0.09	0.56	NS	0.23	0.10	0.26	0.08	0.64	NS	0.43	0.09	0.12	0.04	0.39	NS	0.16	0.05

NS – non-significant differences

the lowest in control plants (without K fertilization) at 19.78 g kg^{-1} DM. In the treatment fertilized with 3.75 K per pot, the K content of leaves increased more than three-fold. A similar trend was observed in pericarps and stems. Higher doses of K fertilizer decreased the N and Ca content of pericarps, the concentrations of N, P, Ca and Mg in leaves, and the Na, Ca and Mg content of stems.

Seed inoculation with *Rhizobium meliloti* significantly increased the content of N in aerial biomass (by 6.5%, 4.7% and 10.4%, respectively) and the concentration of Ca in pericarps and stems (by 4.4% and 5.3%, respectively). Fenugreek plants grown from inoculated seeds were characterized by a lower content of P, K and Mg in pericarps and leaves, and a lower concentration of K in stems.

Fenugreek plants exposed to water deficit (30-35% of available water capacity) during flowering and pod formation contained significantly less P, K and Ca in pericarps and leaves than plants with an adequate water supply. Water stress also contributed to an increase in the N content of leaves and the concentrations of all analyzed nutrients in stems.

Nutrient uptake per plant is presented in Figures 1, 2 and 3. The accumulation of the analyzed nutrients, excluding Ca, in aerial biomass was the lowest in control plants (K0) (Figure 1). The highest accumulation of macronutrients, excluding K, was noted after the application of 2.50 g K per pot (N – 16.10, P – 6.75, Na – 7.60, Mg – 7.04 μg per plant). The accumulation of K increased with the doses of K fertilizer and exceeded 58 μg per plant in the treatment fertilized with 3.75 g K per pot, where Ca accumulation decreased by around 25%. Plants grown from inoculated seeds accumulated 15.70 mg of N, 6.23 mg of P, 39.27 mg of K, 7.29 mg of Na, 28.98 mg of Ca

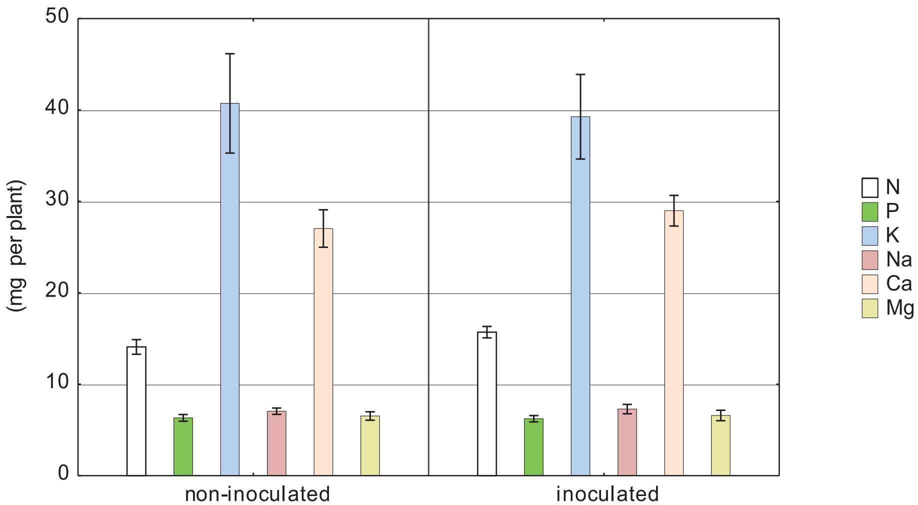

Fig. 1. The effect of inoculation on macronutrient accumulation in the aerial vegetative parts of fenugreek (mean ± 0.95 confidence interval)

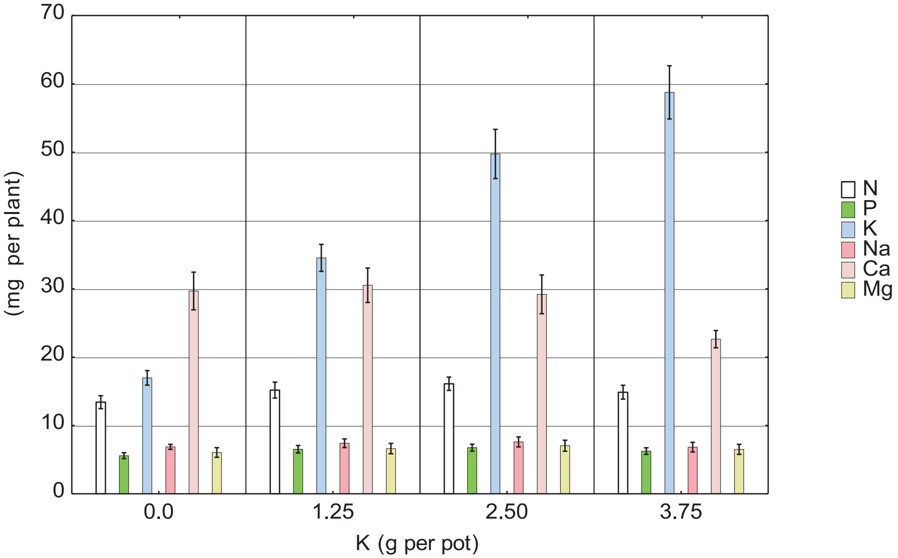

Fig. 2. The effect of K dose on macronutrient accumulation in the aerial vegetative parts of fenugreek (mean ± 0.95 confidence interval)

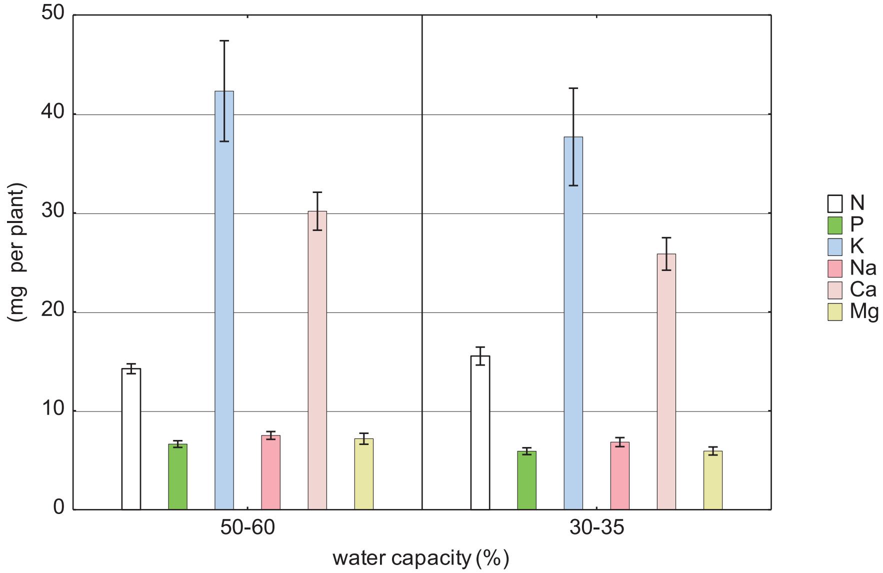

Fig. 3. The effect of water capacity on macronutrient accumulation in the aerial vegetative parts of fenugreek (mean ± 0.95 confidence interval)

and 6.58 mg of Mg per plant. Seed inoculation increased the accumulation of N and Ca (by 11.5% and 7.2%, respectively) as well as Na in straw. Plants grown from non-inoculated seeds were characterized by a somewhat higher content of K (Figure 2).

Water deficit (30-35% of available water capacity) also modified nutrient uptake, and had the most inhibitory effect on the accumulation of Mg, Ca and K, whose content decreased by 17.3%, 16.7% and 12.3%, respectively. Nitrogen was the only nutrient whose accumulation increased in water-stressed plants (by around 9%) – Figure 3.

Biological N fixation by root nodule bacteria ranged from 27.96 to 32.08 mg per plant (Table 3). The amount of biologically fixed N was the lowest in

Table 3

Biological nitrogen fixation

Experimental factor	Symbiotically fixed N	Symbiotically fixed N in soil with fenugreek residues and straw
	(mg per plant)	
K dose (g per pot)		
0	27.96	19.25
1.25	30.19	21.30
2.50	32.08	22.61
3.75	31.43	21.47
$HSD_{0.01}$	0.40	0.20
Rhizobium meliloti		
Non-inoculated seeds	30.82	20.70
Inoculated seeds	30.01	21.62
$HSD_{0.01}$	NS	0.24
Available water capacity (%)		
50-60	31.11	20.91
30-35	29.72	21.40
$HSD_{0.01}$	0.47	NS

NS – non-significant differences

control plants (K0 - 27.96 mg per plant) and the highest (32.08 mg per plant) in plants fertilized with 2.50 g K per pot. A similar trend was noted in the concentration of biologically fixed N in soil containing the post-harvest residues of fenugreek plants. Fenugreek roots and straw enriched soil with N by 19.25-22.61 mg per plant. Seed inoculation with *Rhizobium meliloti* had no significant effect on biological N fixation, whereas water stress during flowering and pod formation significantly decreased the activity of symbiotic bacteria and decreased the amount of biologically fixed N_2 by 4.5%.

DISCUSSION

In the present study, the dry weight of fenugreek biomass was determined by a dose of K fertilizer and available water capacity during flowering and pod formation. FORAWI and ELSHEIKH (1995) demonstrated that under salt stress, the dry weight of shoots and roots in inoculated fenugreek plants cv. Berber increased by 31% and 29%, respectively, in comparison with non-inoculated plants. In the study of DUBEY et al. (2012), the highest straw yield (25.01% higher than in the control treatment) was reported in plants treated with vermicompost (5 t), *Rhizobium* and 40 kg N ha^{-1}. According to the cited authors, the above treatment can increase the seed and biomass yield of fenugreek plants.

The dry weight of leaves and stems was highest in treatment K2.50. The effects of K fertilization were also investigated by SHRIVASTAVA (2015). Potassium fertilizer enhanced the activity of nitrate reductase in leaves, stems and roots, which increased the fresh and dry weight of the analyzed plant parts. Water deficit significantly decreased the dry weight of fenugreek plants, which implies that fenugreek is particularly sensitive to water stress during critical stages of growth.

WILCZEWSKI and SKINDER (2005), and WILCZEWSKI (2007) demonstrated that legume plants are more abundant in nutrients than other field crops. In a study by DUBEY et al. (2012), seed inoculation with *Rhizobium,* fertilization at 30-40 kg N ha^{-1} and the application of vermicompost increased the content of N, P and K in fenugreek straw. It should also be noted that the highest dose of K fertilizer significantly decreased the content of N and Ca in fenugreek pericarps and the content of P and Ca in leaves. SHRIVASTAVA (2015) found that K fertilizer influenced the total phenolic content, total chlorophyll content and carotenoid content of fenugreek plants, thus influencing their pharmacological value and antioxidant potential. The concentration of Ca was higher in the stems of water-stressed plants. A comparative study revealed that fenugreek straw was less abundant in nutrients than barley straw. Fenugreek straw contained 58 g of ash kg^{1} DM and 52 g of crude protein kg^{1} DM, and these values were 45% and 15% lower, respectively, than in barley straw (MUSTAFA et al. 1996). In turn, SYMANOWICZ et al. (2017) reported that an application of higher doses of potassium fertilizers significantly reduced the content of nitrogen in seeds, straw and pods of field pea. In another study, spring wheat straw accumulated less macronutrients, in particular N, Ca and Mg, than field pea straw and the straw of spring wheat and field pea mixtures (BURACZYŃSKA, CEGLAREK 2011). Field pea residues and straw were also characterized by the highest macronutrient content. The crude protein content of fenugreek straw was determined at 5.1%, and animals fed fenugreek straw consumed more dry matter ($P < 0.05$) than animals fed the straw of other legume plants (horse bean, soybean).

Each year, around 139-170 million tons of biologically fixed N enter the global N cycle. Nitrogen fixed symbiotically by bacteria (N_2) accounts for 70-80% of that amount (Peoples, Craswell 1992, Cheng 2008). According to Faostat, legumes are ubiquitous around the world, and the amount of N that is symbiotically fixed by these plants is determined mainly by their genotype (Pietrzak 2011, Hussein, Zaki 2013). According to Pietrzak (2011), the amount of molecular N fixed symbiotically by plants is influenced by the harvest date, plant yield and environmental conditions. In the cited field study, yellow lupine fixed 38 kg N t^{-1} DM and peas incorporated 35 kg N t^{-1} DM. Martyniuk (2008) also noted that the amount of symbiotically fixed N_2 and the percentage of plant N are determined not only by the genotype of legume plants and its symbionts, but also by environmental factors and agronomic factors. In this study, biological N fixation was conditioned by the experimental factors. Due to the lower N content of seeds (Żuk-Gołaszewska et al. 2015), higher concentrations of biologically fixed N were found in the post-harvest residues of fenugreek plants exposed to water stress during flowering and pod formation, and in fenugreek plants grown from inoculated seeds. Symbiotically fixed N not only increases legume yields, but also creates a nitrogen-rich environment for successive crops (Martyniuk et al. 2005, Martyniuk 2008).

CONCLUSIONS

The optimal dose of K fertilization was 2.50 g per pot, as it maximized the dry weight of leaves and stems, significantly increased the fertilizer value of fenugreek residues and increased the amount of biologically fixed N to 32.08 μg per plant. Seed inoculation with *Rhizobium meliloti* influenced only the N and Ca content of straw.

Water deficit during flowering and pod formation significantly decreased the dry weight of fenugreek plants, lowered the accumulation of Mg, Ca and K (by 17.3%, 16.7% and 12.3%, respectively), increased the accumulation of N by around 9%, and decreased the amount of biologically fixed N_2 by 4.5%.

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