

SOME ASPECTS REGARDING THE CULTIVATION OF *HYPERICUM PERFORATUM* SPECIES UNDER CONDITIONS OF CENTRAL MOLDAVIA

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ABSTRACT

Hypericum perforatum L. (St. John's Wort) is a medicinal crop with multiple therapeutical traits from which, it stands out the antiviral effect, with the possibility to obtain some anti-AIDS drugs. During 1996-1999, at Secuieni Agricultural Research Station, the establishment of sowing time and optimum fertilization level in St. John's Wort was investigated, with a view to get crops with increased productivity. Among the four experimental sowing times (first sowing time: 10-20 August, second sowing time: 10-20 September, third sowing time: 20 October - 5 November, and fourth sowing time: 1-10 April) the sowing in the second decade of August, in which the plants emerged in autumn, was the most efficient, giving the greatest dry inflorescence yield - 86.8 q/ha in three years of cropping: first year - 10.7 q/ha, second year - 39.7 q/ha and third year - 36.4 q/ha. The greatest dry inflorescence yield - 84.1 q/ha (first year - 6.1 q/ha, second year - 39.9 q/ha and third year - 38.1 q/ha) was obtained in the case of fertilization with 30 t manure/ha applied under ploughing and of annually chemical fertilization with 45 kg N + 45 kg P₂O₅/ha.

Key words: fertilization variants, *Hypericum perforatum* L. (St. John's Wort), sowing time.

INTRODUCTION

Hypericum perforatum L. (St. John's Wort) is a medicinal crop used from antiquity, as tea, in treatment of some hepatic-bilious diseases and as St. John's Wort unguent (flowers soak in oil at 40-50°C) as scaring cure of wounds (Bojor et al., 1997; Crăciun et al., 1994; Popescu, 1984; Simionescu, 1973; Treben, 1994). Recent chemical studies have revealed the antidepressing and antiviral effect of St. John's Wort hydroalcoholic extracts (Bombardelli and Morazzoni, 1995; Brandle, 1999; Grigorescu and Silva, 1997; Kight, 1997).

In order to utilize the entire yielding potential of different medicinal crops, adequate technologies for each pedoclimatic area, are necessary. The sowing time and fertilization are the factors which largely determine the obtention of crops with increased yielding potential.

The foreign special literature recommends, for the success of St. John's Wort crops, their

establishment with seedlings produced in cold beds or peat pots (Bezzi, 1999; Brandle, 1999; Kight, 1997; Pola, 1999). Poland and Romania are the only countries in which the St. John's Wort is directly cropped into field, at the beginning of winter, but it is not specified if these recommendations are based on rigorous experiments (Coiciu and Racz, 1962; Crăciun, 1976; Păun et al., 1986; Păun, 1995).

As regards fertilization, it is mentioned that the chemical fertilizers with nitrogen, phosphorus and potassium as well as the manure, positively influence the inflorescence yield, the gains ranging between 30 and 50% (Bezzi, 1999; Brandle, 1999; Laza and Racs, 1975; Păun et al., 1986; Păun 1995; Pola, 1999). The fertilizer rates differ very much from one author to another, thus, the recommended manure rates are: 10, 15, 25, 30 till 40 t/ha; nitrogen rates: 30, 35, 60, 80, 100 kg active ingredient/ha; phosphorus rates: 20, 25, 50, 90 kg active ingredient/ha and potassium rates: 10, 30, 35 and 60 kg active ingredient/ha.

Having in view the therapeutical value of St. John's Wort and the contradictory data from scientific literature, studies regarding the cultivation of this species have been initiated at Secuieni A.R.S., during 1996-1999. The paper presents the results regarding the influence of sowing time and fertilizers.

MATERIALS AND METHODS

The experiments were placed on a cambic chernozem well supplied with nutrients (clay - 2.55%; C : N rapport - 11.1; P₂O₅ - 195 ppm; total nitrogen - 0.15%), in randomized blocks. In an experiment, the influence of sowing time was gradually studied, as follows: first sowing time - second decade of August; second sowing time - second decade of September; third sowing time - at the beginning of winter and fourth sowing time - in early spring.

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In another experiment, the influence of fertilizers was gradually studied, in the following variants: V_1 – unfertilized; V_2 – 45 kg a.i. N/ha + 45 kg a.i. P_2O_5 /ha; V_3 – 90 kg a.i. N + 90 kg a.i. P_2O_5 /ha; V_4 – 30 t manure/ha and V_5 – 30 t manure/ha + 45 kg a.i. N + 45 kg a.i. P_2O_5 /ha. The manure was applied under ploughing at the establishment of the crop and the chemical fertilizers were annually applied, as follows: the phosphorus fertilizers in autumn and the nitrogen ones in spring, being incorporated by weedings. It was manually sown, at the soil surface, because the St. John's Wort seeds require light for germination, with a seed rate of 4 kg/ha, at 50 cm distance between rows. Before sowing, the field was packed with a roller. The „De Secuiei” population was sown with seed from the previous year yield, because the seed postmaturation process has a length of one year. The experiment regarding the influence of fertilization was sown at the beginning of winter. The preceding crop was the spring two-row barley. Weedings were performed between and on rows (two-three), depending on the weed infestation degree and climatic conditions of the year. The harvesting was manually performed, by cutting the stems on a length of 20–25 cm from the peak to the base, in the stage of 30–50% open flowers. The row material was naturally dried, under shadow. The hypericine content was determined only in 1999, at the dry inflorescences, according to spectrophotometric method, at the „Stejarul” Biological Research Center of Piatra Neamț. Data processing was made by ANOVA test. During the research

study, the climatic conditions for St. John's Wort were favourable. From the thermic viewpoint, the years were close to the multiannual average (8.5°C), the deviations ranging between -0.3°C (1997–1999) and +0.3°C (1998–1999).

As regards the rainfall, the agricultural year 1996–1997 was the most rainy, the deviations vs. multiannual average (545.1 mm) being of +151.6 mm. In the other two years, the deviations were +10.7 and +37.3 mm, respectively.

RESULTS AND DISCUSSION

The results concerning yield represent the average of two experiments for each year of cropping.

In the first year, only one cutting was obtained and the sowing time significantly influenced the dry inflorescence yield (Table 1). The greatest yield (10.7 kg dry inflorescence/ha) was obtained in the first sowing time, in the second decade of August, the gain vs. control (third sowing time – at the beginning of winter) being of 4.8 q/ha (81%). At this sowing time, the plants emerged from autumn and entered into the winter in two leaflet stage. The plant density at emergence was of 10.5 pl./m². In the second sowing time – the second decade of September and control – sown at the beginning of winter, the yields obtained were of 6.3 and 5.9 q dry inflorescence/ha, respectively.

Table 1. Influence of sowing time on dry inflorescence yield at *Hypericum perforatum*L., first year of cropping one cutting (1996–1997)

Variants	Dry inflorescence yield				Density at emergence pl./m ²	Plant height at harvest- ing cm	Date of emergence
	q/ha	%	diff.	signif.			
First sowing time: 10–10 August	10.7	181	4.8	***	10.5	45.2	1–10 September
Second sowing time: 10–20 September	6.3	106	0.4	-	9.2	38.2	1–10 May
Third sowing time (winter): 20 October – 5 November	5.9	100	control	-	8.9	37.9	1–10 May
Fourth sowing time (early spring): 1–10 April	-	-	-	-	8.6	20.0	30 May – 10 June

Drying output = 4.22

LSD 5% = 0.85 q/ha;
1% = 1.22 q/ha;
0.1% = 1.80 q/ha.

At these two sowing times, the plants emerged in the spring (1–10 May), the density of emergence being about 9 pl./m² at both sowing times (8.9–9.2). In the first year of cropping, the fourth sowing time – in the spring (1–10 April), the plants emerged during 30 May–30 June and did not reach the technical maturity.

In the second year of cropping, two cuttings were obtained and the highest increase of dry inflorescence yield (39.7 q/ha) was registered at the first sowing time – the second decade of August, the gain vs. control – the third sowing time, at the beginning of winter – being of 1.9 q/ha (5%) (Table 2).

The control and the other sowing times (the second sowing time – the second decade of September and the fourth sowing time – in

spring) registered considerably equal yields, between 37.4 and 37.8 q/ha.

In the third year of cropping, two cuttings/year were obtained (Table 3). The dry inflorescence yields obtained in the fourth studied sowing times were practically equal and varied between 36.2 and 36.7 q/ha.

The hypericine content was the same at all the sowing times, 0.1%, which confirms the data from the special literature which mention that the hypericine accumulation in St. John's Wort is, firstly, genetically controlled and followed by the environmental conditions and applied technology (Brandle, 1999).

In the fourth year of cropping, the plants regenerated only in percentage of 10%, which did not justify the experiments maintenance, both for the sowing time and the influence of

Table 2 . Influence of sowing time on dry inflorescence yield at *Hypericum perforatum*L., second year of cropping, two cuttings (1997–1998)

Variants	Dry inflorescence yield, q/ha						Plant height cm
	Yield		I + II	%	diff.	signif.	
I	II						
First sowing time: 10–10 August	27.8	11.9	39.7	105	1.9	*	95.9
Second sowing time: 10–20 September	27.4	10.3	37.7	99	-0.1		97.7
Third sowing time (winter): 20 October – 5 November	26.4	11.4	37.8	100	control		94.8
Fourth sowing time (early spring): 1–10 April	27.2	10.2	37.4	98	-0.4		95.1

Drying output = 4.22 first cutting
3.75 second cutting

LSD 5% = 1.39 q/ha;
1% = 2.00 q/ha;
0.1% = 2.95 q/ha.

Table 3. Influence of sowing time on dry inflorescence yield at *Hypericum perforatum*L., third year of cropping, two cuttings (1998–1999)

Variants	Dry inflorescence yield, q/ha						Hypericine content %	Plant height cm
	Yield		I + II	%	diff.	signif.		
I	II							
First sowing time: 10–10 August	26.2	10.2	36.4	100	0.2		0.1	106.2
Second sowing time: 10–20 September	26.4	10.3	36.7	101	0.5		0.1	105.9
Third sowing time (winter): 20 October – 5 November	26.0	10.2	36.2	100	control		0.1	106.0
Fourth sowing time (early spring): 1–10 April	26.1	10.1	36.2	100	0.0		0.1	106.3

Drying output = 3.77 first cutting
3.89 second cutting

LSD 5% = 0.79 q/ha;
1% = 1.14 q/ha;
0.1% = 1.68 q/ha.

fertilizers.

From the total of yields during the three years of cropping (Table 4), the greatest yield (86.8 q dry inflorescences/ha) was achieved at the first sowing time – the second decade of August, the gain vs. control – the third sowing time at the beginning of winter – being of 6.9 q/ha (9%).

The St. John's Wort dry inflorescence yield was significantly influenced by the applied fertilizers, too.

In the first year of cropping, one cutting only was obtained (Table 5). In comparison with the unfertilized control, the fertilizers applied in rates of 45 kg N + 45 kg P₂O₅/ha and 90 kg N + 90 kg P₂O₅/ha, determined the achievement of some statistically ensured yield gains by 0.6 q/ha (14%) and 0.8 q/ha (19%), respectively. A dry inflorescence gain of 1.4 q/ha (32%) vs. the unfertilized control, was obtained by the application of 30 t manure/ha.

The greatest dry inflorescence yield (6.1 q/ha) was obtained in the variant fertilized with 30 t manure + 45 kg N + 45 kg P₂O₅/ha, the gain vs. control being of 1.8 q/ha (42%).

In the second year of cropping, two cuttings were obtained and the greatest dry inflorescence yield (39.9%) was achieved in the variant fertilized with 30 t manure + 45 kg N + 45 kg P₂O₅/ha, the gain vs. unfertilized control being of 14.6 q/ha (58%) (Table 6). The influence of manure applied in the first year was benefic for the second year too, the yield obtained in the variant fertilized with 30 t manure/ha being 37.9 q/ha and the gain vs. unfertilized control, 12.6 q/ha (50%). In the variants fertilized with chemicals, yield gains were also achieved vs. unfertilized control, by 3.6 q/ha (14%) in the variant fertilized with 45 kg N + 45 kg P₂O₅/ha and by 11.4% in the variant fertilized with 90 kg N + 90 kg P₂O₅/ha.

Table 4 . Influence of sowing time on dry inflorescence yield at *Hypericum perforatum* L., yield sum of first, second and third years of cropping (1996–1999)

Variants	Dry inflorescence yield			
	q/ha	%	diff.	signif.
First sowing time: 10–10 August	86.8	109	6.9	***
Second sowing time: 10–20 September	80.7	101	0.8	
Third sowing time (winter): 20 Oct. – 5 Nov.	79.9	100	control	
Fourth sowing time (early spring): 1–10 April	73.6	92	-6.3	°°

LSD 5% = 3.03 q/ha;
1% = 4.36 q/ha;
0.1% = 6.43 q/ha.

Table 5 . Influence of fertilizers on dry inflorescence yield at *Hypericum perforatum* L., first year of cropping one cutting (1996–1997)

Variants	Dry inflorescence yield				Plant height at harvesting cm
	q/ha	%	diff.	signif.	
Unfertilized	4.3	100	control		28.4
45 kg N + 45 kg P ₂ O ₅ /ha	4.9	114	0.6	*	34.2
90 kg N + 90 kg P ₂ O ₅ /ha	5.1	119	0.8	**	38.1
30 t manure/ha	5.7	132	1.4	***	37.9
30 t manure + 45 kg N + 45 kg P ₂ O ₅ /ha	6.1	142	1.8	***	38.4

Drying output = 4.09 LSD 5% = 0.42 q/ha;
1% = 0.59 q/ha;
0.1% = 0.83 q/ha.

Table 6. Influence of fertilizers on dry inflorescence yield at *Hypericum perforatum* L., second year of crop, two cutting (1997–1998)

Variants	Dry inflorescence yield, q/ha						Plant height cm
	Yeild		I + II	%	diff.	signif.	
	I	II					
Unfertilized	17.7	7.6	25.3	100	control	***	81.7
45 kg N + 45 kg P ₂ O ₅ /ha	20.3	8.6	28.9	114	3.6	***	92.3
90 kg N + 90 kg P ₂ O ₅ /ha	26.4	10.3	36.7	145	11.4	***	95.2
30 t manure/ha	27.1	10.8	27.9	150	12.6	***	95.0
30 t manure + 45 kg N + 45 kg P ₂ O ₅ /ha	28.2	11.7	39.9	158	14.6	***	95.3

Drying output = 3.68 first cutting
3.81 second cutting

LSD 5% = 1.19 q/ha;
1% = 1.67 q/ha;
0.1% = 2.36 q/ha.

In the third year of cropping, two cuttings were also obtained (Table 7). The greatest dry inflorescence yield (38.1 q/ha) was obtained in the variant fertilized with 30 t manure + 45 kg N + 45 kg P₂O₅/ha, the gain vs. unfertilized control being of 13.8 q/ha (57%). The variant fertilized with 90 kg N + 90 kg P₂O₅/ha followed with an yield of 36 q/ha, the gain vs. unfertilized control being of 11.7 q/ha (48%).

The hypericine content was not significantly influenced by fertilizers, being almost the same in all variants (0.099% – unfertilized and 0.1% – the other variants).

From the total sum of yield during the three years of cropping (Table 8), the greatest yield (84.1 q dry inflorescence/ha) was achieved in the variant fertilized with 30 t manure + 45 kg N + 45 kg P₂O₅/ha, the gain vs. unfertilized control being of 30.2 q/ha (56%). In the variants fertilized with 30 t manure/ha, at the establishment of the crop, and 90 kg N + 90 kg P₂O₅/ha, annually applied, equal yields were achieved (77.9–77.8 q/ha), the gains vs. unfertilized control, being of 24 and 23.9 q/ha (44%), respectively.

Table 7. Influence of fertilizers on dry inflorescence yield at *Hypericum perforatum* L., third year of cropping, two cuttings (1997–1998)

Variants	Dry inflorescence yield, q/ha						Hypericine content %	Plant height cm
	Yield		I + II	%	diff.	signif.		
	I	II						
Unfertilized	18.0	6.3	24.3	100	control		0.099	86.9
45 kg N + 45 kg P ₂ O ₅ /ha	19.7	8.2	27.9	115	3.6	***	0.1	99.2
90 kg N + 90 kg P ₂ O ₅ /ha	26.1	9.9	36.0	148	11.7	***	0.1	105.3
30 t manure/ha	24.1	10.2	34.3	141	10.0	***	0.1	106.1
30 t manure + 45 kg N + 45 kg P ₂ O ₅ /ha	26.7	11.4	38.1	157	13.8	***	0.1	105.7

Drying output = 3.82 first cutting
3.96 second cutting

LSD 5% = 1.31 q/ha;
1% = 1.85 q/ha;
0.1% = 2.61 q/ha.

Table 8. Influence of fertilizers on dry inflorescence yield at *Hypericum perforatum* L., yield sum of first, second and third year of cropping (1996–1999)

Variants	Dry inflorescence yield			
	q/ha	%	diff.	signif.
Unfertilized	53.9	100	control	
45 kg N + 45 kg P ₂ O ₅ /ha	61.7	114	7.8	***
90 kg N + 90 kg P ₂ O ₅ /ha	77.8	144	23.9	***
Manure 30 t/ha	77.9	144	24.0	***
30 t manure + 45 kg N + 45 kg P ₂ O ₅ /ha	84.1	156	30.2	***

LSD 5% = 2.92 q/ha;
1% = 4.11 q/ha;
0.1% = 5.80 q/ha.

CONCLUSIONS

Dry inflorescence yield of St. John's Wort (*Hypericum perforatum* L.) was influenced by the sowing time. The optimum sowing time is the second decade of August, when the seeds found optimum conditions for germination, which favoured the plant emergence in autumn. During the three years of cropping, the greatest yield was obtained – 86.8 q/ha (first year – 10.7 q/ha; second year – 39.7 q/ha and third year – 36.4 q/ha), the gain vs. the control – sown at the beginning of winter, being of 6.9 q/ha (9%).

The manure applied at the establishment of the crop and nitrogen and phosphorous fertilizers, annually applied, had a positive influence on dry inflorescence yield, under the conditions of Secuieni A.R.S.

The greatest inflorescence yield – 84.1 q/ha, the sum of yields obtained during the three years of cropping (first year – 6.1 q/ha; second year – 39.9 q/ha and third year – 38.1 q/ha) was registered in the variant fertilized with 30 t manure + 45 kg N + 45 kg P₂O₅/ha, the gain vs. the unfertilized control being of 30.2 q/ha (56%).

The hypericine content, determined only in 1999, was not significantly influenced by both sowing time and fertilization variants, ranging between 0.099 and 0.1%.

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Table 1. Influence of aluminum ions, in reaction mixture, on the level of saccharasic activity in a reddish-brown soil fertilized with compost with different quantities (glucose+fructose-mg/100 g soil dw/24 hours)

A- Factor	B – Factor – COMPOST (t/ha)								Average (A)	
	b1-0	%	b2-0	%	b3-0	%	b4-0	%		%
a1–without Al ³⁺	b 3287	100	b 4028	100	b 2579	100	b 3472	100	b 3341	100
a2- with Al ³⁺	a 4228	129	a 5019	125	a 3472	135	a 4528	130	a 4312	129
Average (B)	3757 c		4523 a		3025 d		4000 b			
LD P	5%	1%	0,1%							
A	291	673*	2143							
B	101	142	201*							
AB	302	628*	1799							
BA	144*	201	284							

Table 2. Influence of aluminum ions, in reaction mixture, on the level of saccharasic activity in a chernozem mineral fertilized or manured with farmyard compost (glucose+fructose-mg/100 g soil dw/24 hours)

A- Factor	B – Factor – COMPOST (t/ha)										Average (A)	
	b1-0	%	b2-N ₃₂ P ₃₂	%	b3-N ₉₄ P ₉₆	%	b4-N ₁₂₈ P ₁₂₈	%	b5 com-post	%		%
a1–without Al ³⁺	b 1564	100	b 1496	100	b 1459	100	b 1401	100	b 1732	100	b 1530	100
a2- with Al ³⁺	a 1686	108	a 1581	106	a 1684	115	a 1589	113	a 1864	108	a 1681	110
Average (B)	1625 b		1538 d		1571 c		1495 e		1798 a			
LD P	5%	1%	0,1%									
A	7	17	54*									
B	14	20	27*									
AB	19	28	45*									
BA	20*	28	39									