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The influence of mineral nutrition and humic acids on the intensity of photosynthesis, as well as the yield and quality of seeds, roots, and aboveground phytomass of milk thistle *Silybum marianum* (L.) Gaertn. in marginal growing conditions

Original Paper

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Abstract Milk thistle *Silybum marianum* (L.) Gaertn. is one of the most researched medicinal plants, but at the same time it belongs to crops with wide industry potential whose growth requirements for marginal agroecological conditions or the response to agronomic interventions has not yet been sufficiently studied. The aim of the present research was to verify the effect of mineral nutrition (NPK 200 kg/ha) and humic acids applied to the soil (HUMAC Agro 250 kg/ha) on the main biometric parameters and the intensity of photosynthesis of milk thistle grown under less favourable climatic conditions of the semi-cold and humid foothills in Central Europe. The highest seed yield was achieved with the NPK variant in terms of dry matter 0.534 t/ha, in the HUMAC Agro variant 0.254 t/ha and with the untreated control 0.087 t/ha. The soil before the trial establishment was characterised by low nutrient content. The use of mineral fertiliser on NPK treatment eliminated the influence of low PK nutrients content of the soil, influencing the crop growth and yield/quality, while no doses of NPK were used on the HUMAC Agro variant and the untreated control. For both of the two sequences created (temperature and light), the highest photosynthesis rate was measured on untreated control (20.115 and 12.386 mmol/m^2/s^1), markedly lower on HUMAC Agro (16.386 and 9.653 mmol/m^2/s^1) – and the lowest on the NPK (10.933 and 7.813 mmol/m^2/s^1, respectively), in inverse proportion to the crop yield. Therefore, the size of the leaf area of the crop was decisive for the increased crop yield. The polynomial trend line of the photosynthesis rate according to the temperature sequence shows the photosynthesis rate and the torrential decrease by temperature increasing on untreated control compared with both treated variants. Both of these threats, mineral nutrition and use of humic preparations to soil, although they reduce the photosynthesis rate, have a wide range of complex effects that provide an opportunity to optimise the growth and yield of milk thistle. Ideally, the mineral nutrition and humic preparations should be used in combination with the crop of milk thistle for medicinal and energy purposes.

Keywords *milk thistle – phytomass – yield quality – photosynthesis rate – humic acids – marginal conditions*

INTRODUCTION

Milk thistle *Silybum marianum* (L.) Gaertn. is one of the medicinal plants most subject to medical research (Karkanis et al., 2011; Martinelli, 2020), but it also belongs to crops with wide industry potential whose growth requirements for marginal agro-ecological conditions have not yet been sufficiently studied. For the development of the best production technologies, it is also necessary to understand the reaction of milk thistle to different phytotechnical interventions, which involve different inputs, levels, and interactions. Therefore, polyfactorial as well as fractional factor field and container-designed experiments are very agronomically useful (Andrzejewska et al., 2015; Bielski, 2021).

At present, there is a need for experiments designed to analyse the response of milk thistle to key factors forming the crop yield and its quality (NPK nutrition, sowing date, stand organisation, seed calibration, seed priming, growth regulators and weed control), which are extended to other

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Table 1. Average content of nutrients in the soil (0-30 cm) and their categorisation, determined according to the Mehlich III methodology – stated before the beginning of the experiment.

parameter	Nt, mg/kg	P, mg/kg	K, mg/kg	Ca, mg/kg	Mg, mg/kg	pH / KCl	C-ox, %	Humus, %	C/N
value / category	1074 midle	29.7 low	80.2 low	3474.2 high	258.3 high	7.03 neutral	1.101	1.897 low	10.2

stimulatory factors that have been little studied in medicinal crops and also include the use of humic preparations (Jahan et al., 2016; Naeeni et al., 2018; Školníkova et al., 2019).

In Slovakia, milk thistle has been the most cultivated medicinal plant for almost two decades (Haban et al., 2009, 2016). The traditional use of seeds (sutures) of this crop is associated mainly with the medicinal use of the components of the silimarin complex, especially on liver regeneration (Cwalina-Ambroziak et al., 2012 and others), but the phytomass harvest potential and its chemical composition suggest possibilities for a much wider use (Andrzejewska et al., 2015). Therefore, efforts to study and verify its suitability for other uses are expanding worldwide, such as seed yields (Ahmad et al., 2014; Vaknin et al., 2008) and aboveground phytomass (Martinelli, 2020; Tsiaousi et al., 2019). This only increases the perspective of growing milk thistle, thus clarifying the requirements for breeding purposes (Alemardan et al., 2013; Andrzejewska et al. 2015; Hammami et al., 2020) and at the same time increasing the need for professional management of agronomic techniques (Bielski, 2021; Martinelli, 2020; Mosavikia et al., 2020).

In the conditions of the mild climate of Slovakia, milk thistle is grown on fertile soils with a good water regime (Haban et al. 2009, 2016), as well as in conditions of marginal soil, such as less fertile heavy and light soil with a disturbed water regime, up to periodic water stress. The agronomic hypothesis predominates that if the plant forms a strong root system, it can withstand different conditions, including less favourable ones (Andrzejewska et al., 2011; Karkanis et al., 2011).

The aim of the presented research was to verify the influence of mineral nutrition and soil humic preparations on the main circular root and the intensity of photosynthesis of milk thistle grown in less favourable submontane climate conditions in Central Europe, while we also investigated the effects on yield and quality of seedlings and aboveground phytomass, including the impact on the main biometric parameters of the stand.

MATERIALS AND METHODS

Plant material, soil conditions, and agronomy

A field experiment with the vegetation of *Silybum marianum* (L.) Gaertn. was established under the operating conditions of the processing company Agrokarpaty in the village of Plavnica, with plantations in the foothills of northeastern Slovakia with a mildly cold and humid climate. An experiment covering an overall area of 4 ha was concentrated on a plot with medium heavy clay soil, at an altitude of 540 m (above sea level) within close proximity to the Poprad river. In Table 1 we present the chemical properties of the site, determined by the analysis of soil samples from layers of topsoil of 0–30 cm taken in May 2020 before the start of the experiment.

The MIREL variety (a variety characterised by its origins in the Czech Republic and by both high *silychristin* and *silybinin* content, chemotype A) was sown on May 20, 2020, at a depth of 3 cm and a sowing rate of 350,000 seedlings per ha, and commercial seeds were used. Basic tillage to a depth of 24 cm was done in the fall, during the spring preparation prior to sowing; NPK nutrition, and humic preparation HUMAC Agro were incorporated into the soil at a depth of 15 cm. Differentiated nutrition (Table 2) for a dose of 200 kg/ha NPK consisted of 30 kg/ha N, 30 kg/ha P_2O_5, and 30 kg/ha K_2O. After emergence, the stand was treated against weeds with Pendigan (*pendimethalin*, 400 g/l) at a dose of 3.0 l/ha.

Photosynthesis measurement

The intensity of plant photosynthesis was measured in-*situ* on 21 July 2021 with the LCPro-SD instrument in the phenophase of full thistle flowering (BBCH 69). On each variant, a fully vegetating leaf was measured (flag leaf -3) for a duration of 112 minutes within a sequence of 24 three-minute steps with a gradual change in the intensity of the photosynthetically active FAR light (at a stable temperature) and subsequent temperature (at a constant FAR intensity). Overview of accompanying parameters of the tested range of temperature and light sequence is part of the evaluation (Table 11, Figures 1 and 2).

Laboratory analyses

Collection of plant and soil samples for biometric and laboratory analysis was carried out on the day of harvest, 27 August 2020; the stand was not desiccated before harvest. Samples were analysed in the laboratory NPPC-VUA Michalovce, where biometric measurements were also performed on plant samples. The qualitative content of the components of the silimarin complex *silychristin* (SC), *silydianin* (SD), *silybinin A* (SB A), *silybinin B* (SB B), *isosilybinin A* (ISB A), and *isosilybinin B* (ISB B) as *flavonolignans* (SC + SD + SB A + SB B + ISB A + ISB B) with relative retention times (RRT) in comparison with *silybin B* are listed in order of the components 0.61, 0.65, 0.96, 1.00, 1.04 and 1.07 of seeds

Table 2. Dosage rate of mineral nutrients NPK (ox. f.) and humic preparation HUMAC Agro per plots monitored in kg/ha.

treatment / soil amendment	NPK	HUMAC Agro	untreated CONTROL
NPK (nutrient content ratio 15:15:15%, P and K in oxide form)	200	0	0
HUMAC Agro (content 62% of humic acids)	0	250	0

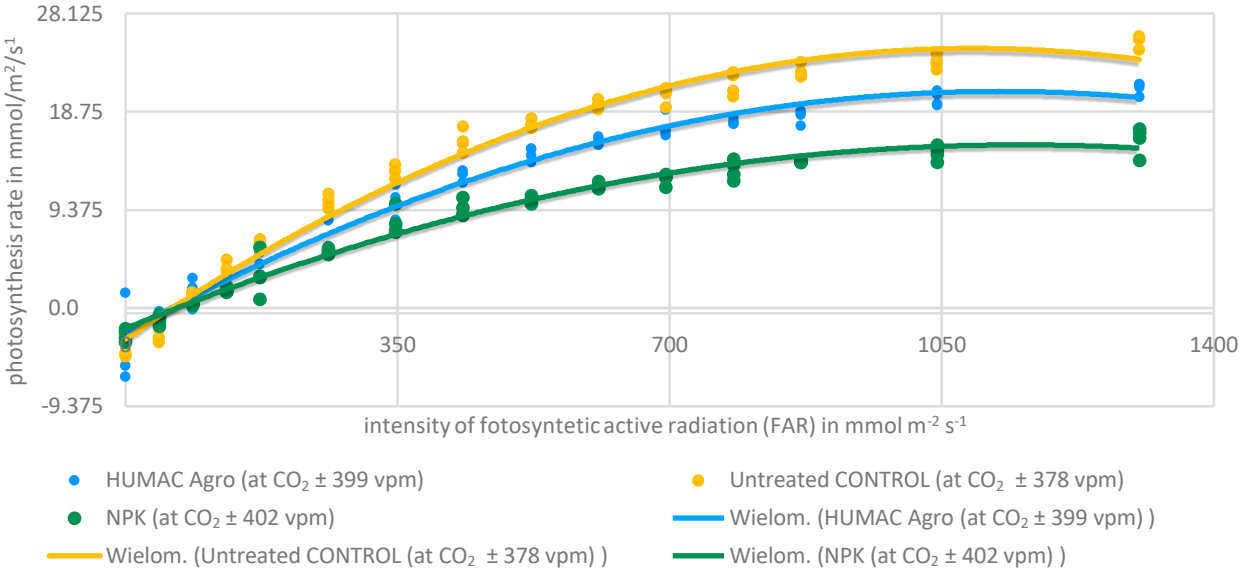


Figure 1. Photosynthesis rate (mmol/m²/s¹) of milk thistle (MIREL, BBCH 69, full flowering) according intensity of FAR and treathments NPK, HUMAC Agro, and the untreated control.

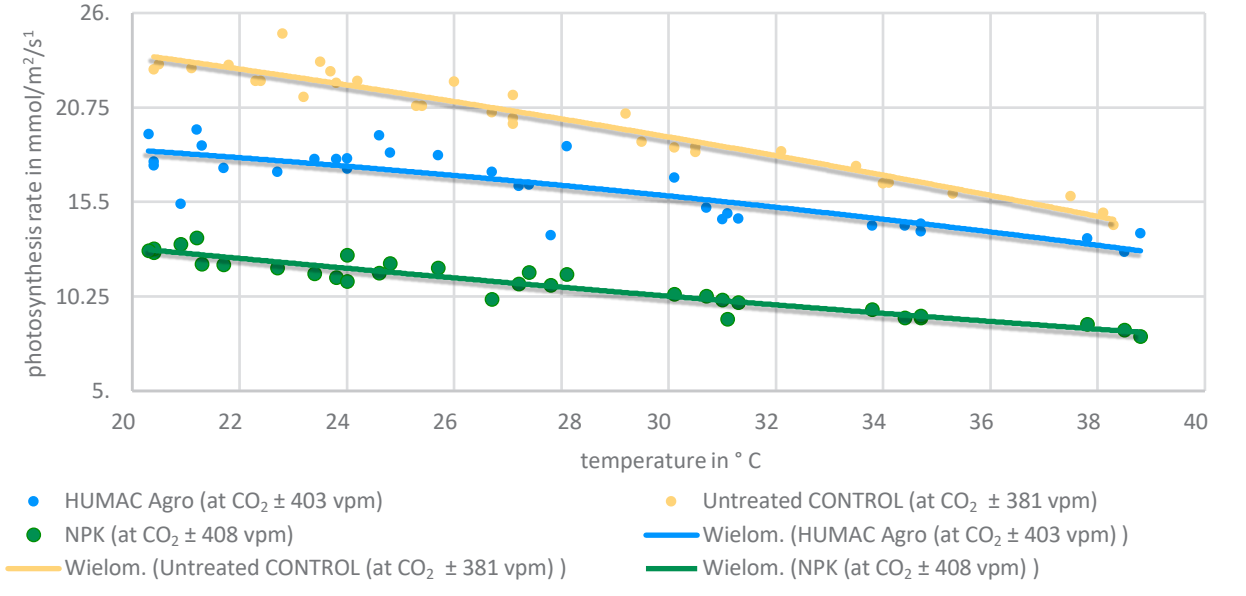


Figure 2. Photosynthesis rate (mmol/m²/s¹) of milk thistle (MIREL, BBCH 69, full flowering) according temperature and treathments NPK, HUMAC Agro and untreated control.

Table 3. Weather conditions and evaluation of hydrothermal coefficient, SHMU Plaveč 2020 station (1–2 km from experimental site).

month / parameter	average daily temperature, ° C	sum of precipitations, mm	hydrotermic coeficient, Hk (valuation)
May	10.0	101.7	3.3 (excess)
Jun	16.5	99.5	2.0 (sufficient)
July	17.1	103.6	2.0 (sufficient)
August	18.1	74.6	1.3 (sufficient)
V–VIII, vegetation period of milk thistle	15.4	379.4	2.5 (sufficient)

Hk rating for vegetation period of milk thistle (V–VIII): > 2.00 excess moisture, 1.01–2.00 sufficient moisture, 1.0 precipitation is equal vapor, 0.51–0.99 lack of moisture, 0.31–0.50 drought, < 0.3 catastrophic drought.

and roots, as determined in the laboratory of Moravol s.r.o. by HPLC. The RRTs stated are for guidance only and depend on the current state of the column and mobile phase. The measuring device is a liquid chromatograph operating in gradient elution mode, ie, Knauer assembly, equipped with a column thermostat with Peltier cells, a UV/VIS detector and a manual dispenser. The system also includes Clarity control and evaluation software running on the Windows operating system. Column: Type RP C18 Nucleosil C18; column temperature: 30° C; flow: 0.5 ml/min; detection: 288 nm.

Acid-detergent fiber (ADF), neutral-detergent fiber (NDF), acid-detergent lignin, crude cellulose (CrC), and hemicellulose (HeC) were determined according to the regulation MP SR no. 2,136 / 2,004-100 by extraction systems: Fiber extractor – Fibertest, Model F-6 and Cold Extraction Unit, Model EF-6. Combustion heat and calorific value were determined in the IKA C 5,000 calorimetric system, in accordance with the STN standard ISO 1928.

Weather conditions

Table 3 presents data on the course of weather conditions for the vegetation period of milk thistle. Data were obtained from a meteorological observation station located near the experiment site and are part of the SHMU (Slovak Hydrometeorological Institute) network with guaranteed record quality. In the pre-vegetation period of 2020, the average daily temperature and the total precipitation for January, February, March, and April were as follows: -2.6° C and 16.6 mm; 1.3° C and 60.6 mm; 2.6° C and 16.8 mm; and 6.7° C and 33.9 mm.

RESULTS AND DISCUSSION

Harvest and quality of seeds

Harvest of milk thistle seeds and roots, as well as the values of the main biometric parameters monitored, growth and indicators of seed quality, were significantly differentiated according to the established treatments and variants (Tables 4, 5, 6, 7, and 8). The highest seed yield was achieved with the variant NPK, for which, in terms of dry matter, the yield was 0.534 t/ha; in comparison with the NPK variant (100%), seed yield was 0.254 t/ha with the HUMAC Agro variant (47.6%), resp. 16.3% at 0.087 t/ha, and seed yield for the untreated control.

Differences in the yield of milk thistle seeds according to treatment variants, order of variants (i) NPK, (ii) HUMAC Agro, and (iii) untreated control correlated with the biometric parameters of the stand, with differences and with the order of variants for the monitored stand parameters. Plant height, the number of plants per area, and coverage were highest with the NPK variant and lowest with the untreated control; similarly, the weight of heads at harvest per m^2, the weight of seeds at harvest per m^2 and per plant, as well as above-ground crop phytomass without buds exhibited the same characteristics (Table 4).

The seeds were characterised by a markedly different moisture content at the time of harvest, which was highest with the HUMAC Agro variant with a value of 32% and lowest with the untreated control with a value of 12.6%. With variant NPK, the moisture content of the seeds was 20.3%. For the bulk density of the seeds and the weight of a thousand seeds, the highest values were observed with the HUMAC Agro variant (665.9 g/l; 21.4 g), lower for the NPK variant (664.3 g/l; 21.1 g), and the lowest in untreated control (653.1 g/l; 20.5 g). The proportion of fully developed mature seeds was highest with the NPK variant (83.8 %), slightly lower with the HUMAC Agro (82.5 %), and the lowest with the untreated control (78.2 %). Immature seeds, red achenes had markedly low values of bulk density (461 g/l) and weight per thousand tons (16.8 g). According to Giuliani et al. (2018) the dark colour of fully ripe fruits is due to the accumulation of condensed tannins in the pericarp of the subepidermal cell layer. The asynchronous flowering and spontaneous release, according to Alemardan et al. (2013), are among the main problems in the cultivation of milk thistle. During the harvest period, the plants have inflorescences at all stages of development results in non-uniform seed ripening. Agronomic measures, as well as breeding efforts, should therefore focus on obtaining plants with unified flowering and reduced seed loss. According to

Table 4. Biometric parameters of stand and above ground phytomass of milk thistle.

treatment / parameter	plant heigh, m	coverage, %	plant density, pieces/m²	plant weight at harvest, g/m²	weight of inflorescence at harvest, g/m²	weight of seeds at harvest, g/m²	weight of seed per plant at harvest, g/plant	aboveground phytomass without inflorescence in 100 % dry matter, t/ha
NPK	1.32–1.54	60–80	31.19	605.42	272.44	79.91	2.56	2.86
HUMAC Agro	1.11–1.36	50–-70	19.76	301.71	144.82	45.65	2.31	1.35
Control	0.69–0.83	20–30	13.38	71.28	39.16	12.73	0.95	0.28

Table 5. Harvest of milk thistle seeds and their basic quality parameters, per treatments monitored.

treatment / parameter	seed moisture at harvest, %	proportion of ripe seeds, %	seed yield at harvest moisture, kg/ha	seed bulk density at 100% dry matter, g/l	thousand seeds weight at 100% dry matter, g	seed yield at 100% dry matter, t/ha
NPK	20.3	83.8	779	664.3	21.1	0.534
HUMAC Agro	32.0	82.5	457	665.9	21.4	0.254
Control	12.6	78.2	127	653.1	20.5	0.087

Table 6. Biometric parameters of the main round root of milk thistle, per treatments monitored.

treatment / parameter	root count, pieces/m²	average root diameter, cm	average root weight, g	root yield, g/m²	root moisture at harvest, %	root yield at harvest moisture, t/ha	root yield at 100% dry matter, t/ha
NPK	31.19	1.19	1.59	48.93	44.3	0.489	0.272
HUMAC Agro	19.76	1.15	1.50	29.65	42.3	0.296	0.171
Control	13.38	0.84	0.75	10.04	27.5	0.100	0.072

Table 7. Content (%) of the components of silimarin complex in seeds of milk thistle.

treatment / parameter	SC	SD	SB A	SB B	ISB A	ISB B	flavonolignans at moisture as received	dry matter as received	flavonolignans at 100% dry matter
NPK	0.814	0.024	0.806	1.235	0.311	0.088	3.278	94.02	3.486
HUMAC Agro	0.870	0.029	0.777	1.193	0.295	0.086	3.167	94.03	3.368
Control	0.750	0.045	0.740	1.131	0.291	0.084	3.042	93.94	3.238

Grest and colleagues (2006), seeds in secondary inflorescence are markedly lower in weight (-56%) compared with the primary inflorescence. Similarly, the weight of the plants and the weight of the main branch are in a positive relationship and are directly or indirectly linked to the yield of seeds. In the definition of the ideotype of plants, ie, stand density of milk thistle, it is therefore appropriate to focus on the high proportion of the main inflorescence and the main branch.

Carrier et al. (2003) found that the average milk thistle plant grown in dry conditions produced 2.0 g, 2.9 g, and 3.4 g of dry seeds per inflorescence of the plant, which corresponded to the harvest in phenophase in the middle of flowering, late flowering, and spontaneous opening of inflorescence, with the highest yield of silymarin detected in the late flowering and spontaneous inflorescence during fruit ripening at the beginning of September.

Table 8. Content (%) of the components of the silimarin complex in the main spinning root of the milk thistle.

treatment / parameter	SC	SD	SB A	SB B	ISB A	ISB B	flavonolignans at moisture as received	dry matter as received	flavonolignans at 100% dry matter
NPK	0.014	0.003	0.015	0.027	0.007	0.005	0.072	91.90	0.078
HUMAC Agro	0.015	0.006	0.014	0.025	0.007	0.005	0.072	91.23	0.079
Control	0.006	0.003	0.005	0.012	0.002	0.001	0.031	91.44	0.034

According to Qavami et al. (2013) The content of silymarin in milk thistle seeds depends on the variety and the geographical and climatic conditions. Also, according to Arampatzis et al. (2019), the impact of climatic conditions on the crop and its quality is demonstrable, whilst the lowest values of silymarin content and harvest of seeds were recorded in a dry year in the season from March to May. The results of Arampatzis et al. (2019) also show that plant density has significant impact on the growth and yield of seeds. The main indicators of growth, such as height, yield of above-ground phytomass, and seeds were highest when plant density was highest.

In the ideal agroecological conditions of the warm lowlands of Slovakia, Haban et al. (2009) studied the impact of intermediate crops, mineral nutrition, and the incorporation of pre-crop post-crop residues into the yield and quality of milk thistle seeds. The seed yields recorded ranged from 1.43 to 1.83 t/ha, and silymarin yields were in the range of 16.5 to 24.6 kg/ha. The content of silymarin complex in the seeds was in the range of 1.5% to 2.0%. Haban et al. (2016), in a similar experiment on an identical site from a later period, state that milk thistle yields ranged from 0.30 to 0.75 t/ha, which is probably closely related to the decline of the genetic value of the farm seed used.

In the agroecological conditions north of Slovakia, humidity and thermal conditions during three years of research, according to Anrdzejewska et al. (2011) caused the yield of seeds to range from 0.55 to 1.68 t/ha and silymarin yield from 13.3 to 35.4 kg/ha, the average silymarin content in the crops was 2.18%. With comparable conditions Cwalina-Ambroziak et al. (2012) state that the milk thistle yield is increased in response to increasing nitrogen utilisation, tested in the range of 0–120 kg N/ha.

Estaji et al. (2016) did not show any demonstrable effect on silybin content based on the tested doses of nitrogen 0–100 kg N/ha. According to Rahimi and Kamali (2012), the delayed sowing time apparently significantly reduces inflorescence for all methods of fertilisation. They concluded that the highest yield of seeds and essential oil was obtained with the earliest sowing date, whilst a declining trend was observed with a sowing delay.

According to Karkanis et al. (2011) milk thistle is considered drought resistant and average rainfall is sufficient even in the semi-arid conditions of Greece. In a Mediterranean environment, in strong drought conditions, milk thistle should be irrigated during seed growth and filling. In the experiments of Afshar et al. (2014, 2015) with declining irrigation, where a mild to severe decline of moisture resulted in a 7% and 27% reduction of seed yield compared with the full irrigation dose. Regardless of the irrigation regime, the application of poultry manure and vermicompost increased the yield of milk thistle. Application of these organic materials to soil had no significant effect on the silymarin and oil content of the seeds, but the silymarin and oil increased owing to the beneficial effect on seed yield.

In view of the findings of Omidbaigi et al. (2003) with the most suitable plant density of 6.7 units per m^2 (at 50 × 30 cm, this is 1.500 cm^2 per plant) it is clear that the organisation of milk thistle growth is strongly influenced by agroecological conditions. This is also confirmed by the conclusions of the same study on the suitability of the sowing date, which states that the best time to sow milk thistle is in the autumn, especially in September. In trials, Omidbaigi studied the effect of sowing dates September 11 and October 17 as autumn sowing and March 10 and April 12 as spring sowing. Within plant density, they monitored growth, seed yield, and active ingredients at 8.0, 6.7, and 5 plants per m^2. According to Haban et al. (2009), in the central European climatic conditions of Slovakia, milk thistle does not hibernate during the winter, and therefore only spring sowing is possible.

Yield and quality of aboveground phytomass and parameters of round root

Similar to seed yield, the yield of aboveground phytomass of milk thistle is significantly different based on the treated variants (Table 4) as well as the diameter and other monitored parameters of the main root (Table 6). The highest dry matter yield of aboveground phytomass without buds was 2.86 with the NPK variant t/ha (100%), significantly lower with HUMAC Agro 1.35 t/ha (47.2%), and lowest with the untreated control 0.28 t/ha (9.8%). The highest weight of the main round root was in the NPK variant 0.272 t/ha (100%), lower with HUMAC Agro 0.171 t/ha (62.9%), and lowest with the untreated control 0.072 t/ha (26.5%). The average thickness of the main round root and its weight were highest for NPK variant (1.19 cm and 1.59 g, respectively 100% and 100%), lower for HUMAC Agro (1.15 cm and 1.50 g, 96.6% and 94.3%, respectively) and lowest for the untreated control (0.84 cm and 0.75 g, and 70.6% and

Table 9. Qualitative indicators of phytomass of milk thistle above ground level according treatments, results in 100% dry matter (heating value at the listed analytical moisture content).

treatment / parameter	ADF, %	ADL, %	CrC, %	HeC, %	NDF, %	heating value J/g (moisture, %)	combustion heat, J/g	phytomass above ground level without inflorescence, g/ha	energy yield, GJ/ha
NPK	59.88	10.25	49.63	4.55	64.43	16,123 (6.83)	16,290	2,863,628	46.649
HUMAC Agro	56.24	7.74	48.49	4.88	61.13	16,055 (7.20)	16,227	1,349,254	21.894
Control	60.36	8.31	52.05	4.81	65.17	16,142 (6.62)	16,306	276,232	4.504

47.2%, respectively). The moisture of the root at harvest was the highest in the NPK variant 44.3% (relatively 100%), lower for the HUMAC Agro variant 42.3% (relatively 95.5%), and the lowest for the untreated control 27.5% (relatively 62.1%).

Qualitative indicators of above-ground phytomass without buds, which are important indicators for the use of phytomass of milk thistle for various energy purposes, are listed in Table 9. ADF content (56.24%–60.39%), ADL (7.74%–10.25%), cellulose (48.49%–52.05%), hemicellulose (4.55%–4.88%), and NDF (61.13%–65.17%) were not significantly differentiated in the phytomass above ground level, according to the variants monitored, similarly to the calorific value (16.06–16.14 kJ/g at comparable humidity, 7.20%–6.62%) and combustion heat (16.23–16.31 kJ/g). According to the yield of of milk thistle phytomass above ground level (without buds) the energy yield was highest in the variant NPK 46.649 GJ/ha. However, with significantly differentiated yields of phytomass above ground level, the energy yield was significantly different depending on the variants monitored. The highest phytomass energy was found in the NPK variant 46.649 GJ/ha (100%), significantly less with HUMAC Agro 21.894 GJ/ha (49.9%), and even less for the untreated control 4.504 GJ/ha (9.7%).

Milk thistle provides suitable above-ground phytomass for various energy uses, however, in the somewhat cold and humid foothill conditions of northern Slovakia, and the yield is not quite satisfactory. It is necessary to take into account the losses during harvest of phytomass, which may be approximately 20%–30% and more. In the conditions of southern Slovakia with warmer lowlands, the yield potential of the phytomass of milk thistle above ground level is higher, if we take into account our own findings and literature data, especially owing to the earlier onset of spring and therefore the possibility of earlier sowing. Southern locations provide the advantage of a higher input of solar radiation, while northern locations provide the advantage of more sufficient moisture. Higher daily temperatures alone during the second half of the milk thistle growing season are not a clear advantage when growing milk thistle.

In the semi-arid conditions of Greece, Tsiaousi et al. (2019) found that milk thistle growth produced 15.49 to 20.89 t/ha of dry matter of phytomass above ground level, in which the yield was reduced by the encroachment of weeds onto the plantation and the gross calorific value of milk thistle phytomass ranged from 15.5 to 16.1 MJ/kg. At an average net energy gain (energy yield reduced by energy deposits in cultivation) in the range of 80.13 GJ/ha to 111.87 GJ/ha, in the conditions of the Mediterranean climate, milk thistle provides suitable phytomass of satisfactory quality for energy use.

According to Martinelli (2020), an analysis of the composition of milk thistle phytomass shows that the contents of substances that are possible to extract, such as ash, lignin, and cellulose, is comparable with other herbal energy crops with these traits show only limited variability. With milk thistle, only xylans form the hemicellulose fraction, and its content, therefore, appears on average lower compared with the phytomass of other herbs.

The course of weather, soil parameters, and intensity of photosynthesis

Milk thistle is sown in the second half of May and harvested in late August. In terms of weather conditions, the vegetation period in the given locality is characterised by sufficient to abundant precipitation with a total of 379.4 mm and an average daily temperature of 15.4° C, (Table 3). Planting did not suffer from a lack of moisture; in May it was even exposed to an excess amount of moisture. The sowing of the stand was postponed by two weeks in comparison with the optimal sowing date for the given area, owing to the delayed onset of spring.

The chemical properties of the soil monitored, as well as the condition and categorisation before the start of the experiment are provided in Table 1. The condition and categorisation after the end of the experiment are provided in Table 10. The soil on the plot before the establishment of the experiment was characterised by low P (29.7 mg/kg) and K (80.2 mg/kg) as well as low humus content (1.897%). The use of mineral nutrients on the NPK variant partially eliminated (with P sufficient and K insufficient) the effect of the low content of PK nutrients in the soil on the growth and yield of milk thistle, whereas with variants HUMAC Agro and untreated control no dose of industrial nutrients was used, and growth and yield of milk thistle were limited differentially. Compared with the state before sowing, higher content of P and K in the soil after the end of the experiment on each variant treatment (Table 10) can be associated mainly with

Table 10. Average content of nutrients in the soil (layer 0–30 cm) and the categorisation, determined according to the Mehlich III methodology, stated at the end of the experiment.

treatment / parameter	Nt mg/kg	P mg/kg	K mg/kg	Ca mg/kg	Mg mg/kg	pH	C-ox %	humus %	C/N
NPK	1174 very low	101.2 midle	118.2 low	3,220.6 middle	506.1 very high	6.88 neutral	1.850	3.188 middle	15.8
HUMAC Agro	1,510 high	49.2 low	140.3 slight	3,747.5 High	359.7 very high	6.88 neutral	1.087	1.873 low	7.2
Control	1082 low	77.4 slight	92.2 low	3465.1 high	290.6 high	6.99 neutral	1.443	2.487 midle	13.4

Nt evaluated with respect to the content of organic carbon, C / N.

Table 11. Photosynthesis rate (PHR, mmol/m^2/s^1), photosynthetic active radiation (FAR, mmol/m^2/s^1) and accompanying parameters of the sequences tested.

treatment / parameter	the sequence of temperature					the sequence of light (FAR)				
	FAR	CO_2, vpm	range of temperature, ° C	average temperature, ° C	PHR	temperature, ° C	CO_2, vpm	range of FAR	average FAR	PHR
NPK	608	402	20.3–38.8	27.6	10.933	30.3	408	0–1,304	484	7.813
HUMAC Agro	608	399	20.3–38.8	27.6	16.386	30.3	403	0–1,304	509	9.653
Control	608	378	20.4–38.3	27.7	20.115	30.2	381	0–1,304	502	12.386

the date of collection (the effect of higher mineralisation of nutrients in warm August rather than in cold April), while to a much lesser extent we also assume it results from making nutrients available through the root secretions of milk thistle itself (for the main nutrients P and K it shows a different final effect). In the case of large-scale, partly operational experiments, to some extent the surface heterogeneity of the plot plays a role, which is associated with the different humus content according to the treatment variants, where the final state may be distorted more than the nutrient content.

The intensity of milk thistle photosynthesis was significantly different among the variants monitored, similarly to the sequence of temperature changes as well as the sequence of changes in the intensity of photosynthetically active radiation (Table 11, Figure 1 and 2). In both sequences, the highest intensity of photosynthesis was in the untreated control (20.115, 12.386 mmol/m^2/s^1, 100%–100%), significantly lower for HUMAC Agro (16.386 and 9.653 mmol/m^2/s^1, 81.5%–77.9%) and lowest at NPK (10.933 and 7.813 mmol/m^2/s^1, 54.4%–63.1%). The polynomial trend line of the temperature sequence also shows that the intensity of photosynthesis in the untreated control is characterised by a sharper decrease due to increasing temperatures compared with both treated variants.

The performance of photosynthesis is a parameter that is measured and related to a unit of leaf area. Increased milk thistle yield in both treated variants compared to the untreated control is in strong negative correlation with the decrease in photosynthesis performance, ie the size of the leaf is decisive in the formation of the crop area per unit area of land. Although mineral nutrition and the use of humic preparations in the soil reduce the photosynthetic performance of leaves, in a wide range of their actions they provide an opportunity (ideally if they are used in combination) to optimise the growth and yield process of milk thistle.

According to Geneva et al. (2008), increasing the yield and quality of milk thistle seeds using plant growth regulators in combination with soil or foliar mineral fertilisers is associated with the regulation of growth and development, and the aim should be to prolong the reproductive stage and increase the yield of seeds with a high silymarin content. They found that treatment of milk thistle with foliar fertilisers and growth regulators resulted in an increase in mature inflorescences.

CONCLUSIONS

The influence of mineral nutrition and soil humic preparations on the milk thistle was verified, while we investigated its effects on the yield and quality of seeds and phytomass above ground level as well as on the main circular root and on the intensity of photosynthesis.

Although both mineral nutrition and the use of humic preparations in soil reduced the photosynthesis rate of the leaves they have a wide range of complex effects that provide an opportunity to increase and to optimise the growth and

yields of milk thistle. Ideally, the mineral nutrition and humic preparations should be used in combination within the cropping of milk thistle for medicinal and energy purposes.

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