GREEN MANURING WITH CRIMSON CLOVER AS AN ALTERNATIVE TO MINERAL FERTILIZATION IN MAIZE PRODUCTION: ONE SEASON RESULTS FROM NORTHEAST CROATIA

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ABSTRACT

Role of mineral fertilizers is to complement the soils' indigenous supply of minerals for crop plants nutrition. Among the mineral nutrients, nitrogen is generally considered as the most yield-contributing. Obtaining nitrogen from legumes is potentially more sustainable than from industrial sources. Crimson clover (*Trifolium incarnatum* L.) has long been appreciated as cover crop grown for green manuring due to its capability for binding the atmospheric nitrogen into plant-available form. Green manuring effects of crimson clover for succeeding maize crop were tested in 2019 year on two locations of Northeastern Croatia differing in soil texture: 1) on the site A where the soil was compact and dominated by clay and 2) on the site B where the soil was loose and dominated by loam. The research was conducted in a year with favourable rainfall during maize vegetation. Results have shown that green manuring with crimson clover for maize as the main crop delivered somewhat lower maize grain and aboveground mass yields than the conventional full-dose mineral fertilization on deep fertile soils around Osijek (Northeastern Croatia). When compared to conventional agronomy, green manuring was associated with additional agrotechnical operations required for crimson clover cover crop establishment and its herbage incorporation into soil prior to seeding main maize crop. Research should be continued in more different environments (year × location combinations) in order to produce reliable evaluation of crimson clover's green manuring services.

***Keywords**: green manuring, crimson clover, maize, yield*

INTRODUCTION

Mineral fertilizers complement the soils' indigenous supply of minerals for crop plants. Among the mineral nutrients, nitrogen is generally considered as the most yield-contributing element. Before the availability of relatively inexpensive nitrogen fertilizers, legume cover crops were commonly used as green manure to supply nitrogen needs of the succeeding crops [1]. Nitrogen-based fertilizers have increased agricultural productivity, but have shown detrimental effects to the environment and human health [2]. Obtaining nitrogen from legumes is potentially more sustainable than from industrial sources [3]. According to the cited authors, main objections to the use of industrially synthesized nitrogen fertilizers are their links to eutrophication of water bodies, groundwater contamination with nitrates, stratospheric ozone destruction and global warming. In addition, considerable energy consumption is associated with mineral N fertilizers. Lal [4] in his review presented the average energy requirement of 90 MJ/kg of N for the production of ammonium nitrate and 101 MJ/kg of N for urea. Fertilizers production is inevitably followed by energy consumption used for packaging, storage and distribution of fertilizers. From the farmers' point of view, it is crucially important to consider that mineral nitrogen fertilizers application diminishes soils' fertility in a long run, mainly due to acceleration of humus degradation and depletion of indigenous nitrogen reserves [5]. According to the cited group of authors, long-term sustainability may require a gradual transition from intensive synthetic nitrogen inputs to legume-based crop rotations. Crimson clover (*Trifolium incarnatum* L.) has long been appreciated as cover crop grown for green manuring due to its capability for binding the atmospheric nitrogen into plant-available form. However, there is lack of research and practical experience dealing with crimson clover application as a green manure in Croatia and in broader Pannonian region as well. We decided to check the crimson clover's green manure effects in maize crop production due to two reasons: 1) maize is one of the most nitrogen consuming crops and 2) maize crops occupy the greatest share of Croatian arable area [6]. Crimson clover's green manure effects were compared with conventional mineral nitrogen fertilization and unfertilized control.

EXPERIMENTAL

Green manuring effects of crimson clover for succeeding maize crop were tested in 2019 year on two locations differing in soil texture: Site A, on the location near Tenja (lat. 45.487, long. 18.733), southern from Osijek, where the soil was compact and dominated by clay, and Site B, on the location western from Osijek (lat. 45.546, long. 18.648), where the soil was loose and dominated by loam. Preceding crops, soil fertility indicators and conventional fertilization recommendations according to the Soil Testing Laboratory of the Faculty of Agrobiotechnical Sciences Osijek differed between locations (Table 1).

Table 1. Preceding crops, soil fertility indicators and conventional mineral fertilizer recommendations for maize production on experimental locations

Parameter	Site A	Site B
Preceding crop	unfertilized forage grass mixture	oilseed rape
Dominant soil texture component	clay	loam
Soil pH(H_2O)	6.37	6.30
Soil pH(KCl)	5.77	5.51
Humus (%)	2.24	1.83
AL extracted P_2O_5 (mg/100 g of dry soil of ploughing layer)	63.63	17.09
AL extracted K_2O (mg/100 g of dry soil of ploughing layer)	35.73	19.82
Recommended total N fertilization based on soil's humus content (kg/ha)	210	215
Recommended P_2O_5 fertilization (kg/ha)	0	95
Recommended K_2O fertilization (kg/ha)	0	215

Four fertilization variants were tested in the field experiment: green manuring with crimson clover (CC), full-dose mineral fertilization (FM), half-dose mineral fertilization (HM) and zero-fertilization (ZF). In CC and ZF variants, no mineral fertilizer was applied. On both locations, prior to the crimson clover cover crop establishment, soil was plowed (in early September 2018, uniformly in all variants as well) to the depth of 30 cm and thereafter seedbed (for CC variant only) was prepared with rotary harrow. Crimson clover convar. Inkara was broad-seeded with 20 kg/ha of pure live seeds in mid-September 2018 and thereafter lightly harrowed, on both locations. It emerged before mid of October 2018 and was well developed before the onset of winter in December 2018. Crimson clover chopping and incorporation into the soil was performed in May 2019 (beginning of May on site A and at the end of May on site B). Prior to chopping and incorporation of the crimson clover, cover-crop samples from 1 m^2 area in each replication were collected for weighing the fresh herbage. From these samples, subsamples (of about 200 g fresh-weight) were taken for dry matter (DM) and nitrogen (N) content determination. The rest was returned to the experimental plots in order to be incorporated into the soil. Subsamples were air-dried for a week and then oven-dried on 70 °C during eight hours to obtain dry weights. N content (in dry matter (DM)) was measured by Kjeldahl method [7].

In FM, HM and ZF variants soil was plowed at the same time and depth as in the CC variant (all variants uniformly, in early September 2018) and was left until the spring tillage with rotary tiller. Mineral fertilizers were applied manually (by hand-spreading) before the spring rotary tillage, according to the conventional recommendations either in full or halved dose, respectively (Table 2).

Table 2. Mineral fertilizers application in full mineral (FM) and half mineral (HM) fertilization variants

Parameter	Site A		Site B	
	FM	HM	FM	HM
Urea 46 % N, incorporated into soil with spring rotary tillage, before maize sowing (kg/ha)	310	155	310	155
NPK 0:20:30, incorporated into soil with spring rotary tillage, before maize sowing (kg/ha)	0	0	500	250
KAN 27 % N, incorporated into soil with the first inter-row cultivation of maize (kg/ha)	250	125	250	125
Total given N (kg/ha)	210	105	210	105
Total given P_2O_5 (kg/ha)	0	0	100	50
Total given K_2O (kg/ha)	0	0	150	75

In all variants, maize was sown the same day when the CC cover crop was incorporated into soil (early May at Site A and end of May at Site B). Maize was seeded at stand density of 8 plants per m^2, distributed into rows with inter-row distance of 0.7 m. Choice of maize hybrid was adjusted to the sowing term (KWS Capitolis, later FAO 400 at Site A and KWS Camparis, earlier FAO 400 at site B). Maize plots in all experimental variants were cultivated by inter-row cultivation. Weeds were additionally controlled with combined herbicides rimsulphuron (13 g/ha) and dicamba (243,6 g/ha), sprayed in five visible leaves vegetative stage of maize. Field experiment was arranged in a complete random block design (CRBD) with four replications [8]. Basic plots were 5.6 m wide and consisted of eight maize rows, with distance between rows of 70 cm, and 10 m long (56 m^2 per plot). Unexpectedly, at the Site B the fourth replication was unintentionally destroyed, therefore allowing measurements only from the first three replications. By the end of October 2019, plants were harvested by cutting them at height of 25 cm from soil. Plants from the four inner rows were taken for measurements, while the two outer rows from left and right side were excluded in order to eliminate the border-effects. Plants from the first and last 1.5 m of each row were also excluded for the same reason. The aboveground maize mass was separated into ears and the rest (stalks and leaves), and thereafter fresh-weighed. Subsamples (about 500 g each) were taken for dry matter (DM) content measurements. Maize subsamples were air-dried for two weeks and thereafter oven-dried at 70 °C for one whole day in order to obtain dry weights. Dry matter yields were calculated into DM yields per hectare. Ears were later separated into grains and cobs in order to calculate the grain DM yield. Total above-ground herbage DM yield was calculated as a sum of ear and rest plant DM yield and referred as whole-crop maize yield in the further text. The applied Complete Randomized Block Design (CRBD) of field experiment has enabled the separation of total experimental variance into parts caused by fertilization variants and caused by the blocked arrange of replicates by performing the analysis of variance [8]. The residual variance was used for calculation of standard error of difference and least significant difference (LSD, at the confidence level 95 %; [8]). Statistics were calculated and results graphically presented by using MS-Excel software.

Average monthly temperatures and precipitation during the field trial were measured on the nearest meteorological station at Klisa airport near Osijek (Figure 1) and compared to the closest station with records in a referent 30-year period (1962 - 1990, Slavonski Brod).

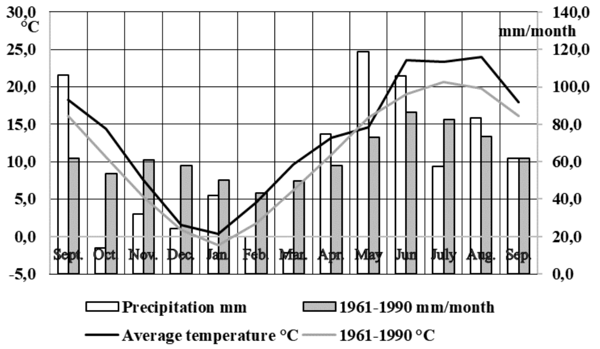

Figure 1. Meteorological data from the station at Klisa airport near Osijek collected during the field trials, from September 2018 until September 2019, and 30-year average from meteo-station in Slavonski Brod

RESULTS AND DISCUSSION

Crimson clover developed abundant herbage mass prior to its chopping and rotary tilling incorporation into the soil, with considerable nitrogen content (Table 3). Favourable precipitation in September 2018 and air temperatures during that autumn and following spring (Figure 1) have probably contributed to CC's quick emergence, fast development and large herbage mass when tilled into soil.

Crimson clover (CC) herbage DM found on Site A was similar to previous research of Rannels and Wagger [9] who found 5.2 t/ha of crimson clover herbage in their two-year study in North Carolina (USA). Nitrogen concentrations in our trial were lower than in Rannels and Wagger [9] research, who found 2.59 and 2.16 % of N in DM during April (11 April 1990 and 17 April 1991, respectively). They established their subsequent maize by no-till seeding, when crimson clover was in the phase 50 % anthesis (50 % bloom). Presented results corresponded to their variant with autumnal seeding of CC. They have also found that CC can be self-reseeded when left to grow and mature between maize rows, thus saving the costs of CC seeding in subsequent years. The trial was set on sloped sandy loam soil. In Georgia (USA), Touchton et al. [10] have found CC herbage DM about 4.7 t/ha in 1979 and 1980, with nitrogen content 1.81 and 1.85 % in DM, for each year, respectively, in the phase of CC seed maturity, just prior to subsequent no-till sorghum planting in early June. The soil was sandy-loam. Bakhtyari et al. [11] have found 6.09 t/ha of CC dry matter in Iran, in their first cut, with no aftermath. Much greater herbage DM found on Site B was due to prolonged vegetation, until the off bloom (Table 3), and probably due to more favourable soil texture (Table 1), structure and overall soil fertility.

Table 3. Crimson clover herbage dry matter mass and total nitrogen content prior to chopping and incorporation into the soil and maize hybrid seeded

Parameter	Site A	Site B
Term of cover crop – cash crop alteration	beginning of May 2019	end of May 2019
Crimson clover developmental phase	bloom	off bloom
Average cover crop above-ground herbage DM (t/ha) and standard error of mean in brackets	4.4 (0.16)	10.1 (0.50)
Total N concentration in cover crop herbage (on a dry matter basis, %)	2.34	2.07
Average amount of N tilled into the soil with cover crop herbage (kg/ha)	103	207

Weather conditions during the maize vegetation (from May to September 2019) were also favourable, mainly due to a plenty of rainfall (Figure 1). Considering the average whole-crop maize DM yields on Site A, the trial could detect significant difference between the FM and ZF variants only (Figure 2). Average yield of CC variant was found intermediary between the FM and ZF variants, while the HM variant was closer to the FM.

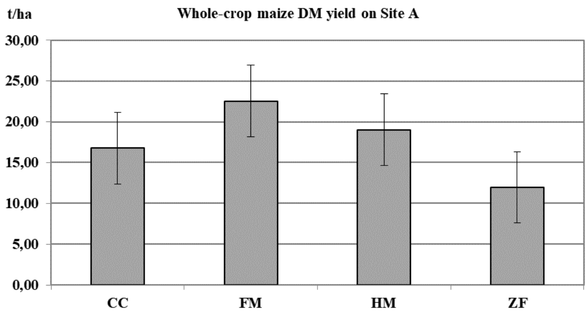

Figure 2. Maize average aboveground DM yields on Site A are presented by columns. Vertical bars present the LSD (95 % confidence)

On Site B trial could not detect any significant differences between fertilization variants (Figure 3), probably due to lesser number of replications (tree instead of four), which has caused greater residual variance and therefore greater LSDs and poorer sharpness of arithmetic means. However, FM and ZF were repeatedly found the extreme variants.

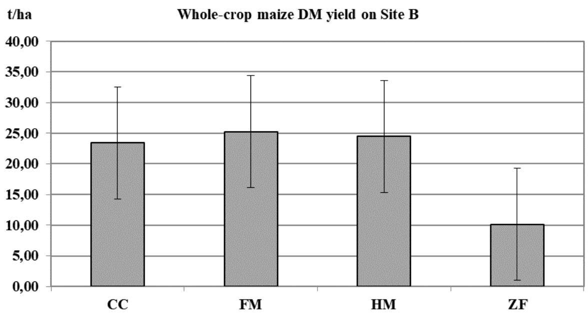

Figure 3. Maize average aboveground DM yields on Site B are presented by columns. Vertical bars present the LSD (95 % confidence)

Average maize aboveground DM yield found in CC variant on Site A was in the range found by Wagger [12] in North Carolina (USA). 34 years ago, he found maize herbage DM yields between 10.04 and 18.90 t/ha, 16 weeks after maize planting, depending on the term of shift between cover crop and cash crop, and depending on year of trial (1984 or 1985). Greater maize herbage DM yield on Site B could be attributed to more favourable soil texture (Table 1) and overall fertility, similarly to the observed greater CC herbage mass on the same location. However, nowadays are expected greater maize herbage and grain yields than 34 years ago due to genetic improvement of maize cultivars during the same period [13].

Considering the average maize grain DM yields on Site A, the trial could detect significant difference only between the FM and ZF variant (Figure 4) again. Average yield of CC variant was found intermediary between the FM and ZF variants, while the HM variant was closer to the FM.

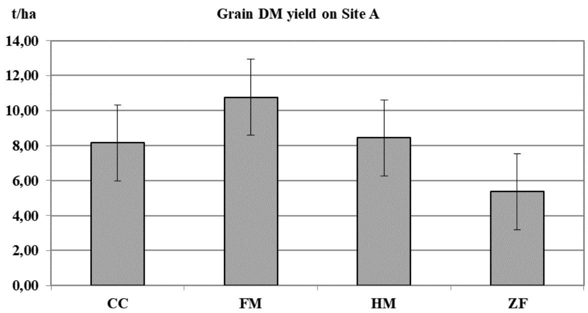

Figure 4. Average maize grain DM yields on Site A are presented by columns. Vertical bars present the LSD (95 % confidence)

On Site B we could not detect significant differences between arithmetic means, probably because of the loss of one replication (Figure 5). Similarly, to whole-crop yields, FM and ZF variants took the extreme values of grain DM yield.

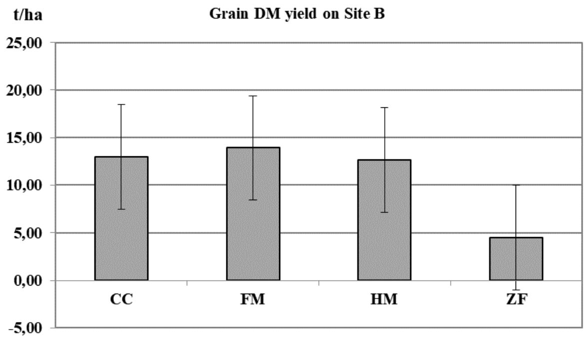

Figure 5. Average maize grain DM yields on Site B are presented by columns. Vertical bars present the LSD (95 % confidence)

Grain DM yield found on Site A was in the range of Wagger [12] research in North Carolina. Maize grain yield after crimson clover cover crop with no mineral N fertilization in Wagger's [12] trial varied between 5.55 and 10.44 t/ha, depending on the year of investigation. Maximum grain yields in his research (9.25 to 11.82 t/ha in favourable 1984 year) were associated with mineral nitrogen fertilization with 200 kgN/ha. However, half-dosed mineral nitrogen mainly did not significantly decrease grain yields in his research. In Ontario (Canada), Coombs et al. [14] found maize grain yields 10.7 and 5.42 t/ha (depending on the year of field trial) after termination of winter CC cover crop, with no mineral nitrogen fertilization. Greater grain yields found on Site B could be attributed to more favourable soil texture (Table 1) and overall soil fertility too, and likely to the genetic improvement of maize grain yields during the last 34 years [13], and lower latitude when compared to the trial in Ontario.

Precious results of CC green manuring are available for another warm-season cereal. Grain yield of no-till planted sorghum into CC mulch varied between 2.8 and 6.8 t/ha without mineral N addition in Georgia (USA), and addition of up to 135 kgN/ha did not give much gain to the sorghum yield [10]. Cover crop mixes of legumes with crucifers provided somewhat poorer green manure services than sole legumes due to their greater C/N ratio [15]. Although vetches (*Vicia* sp.) can bring more atmospheric nitrogen into soil than crimson clover [1, 16], vetches are more suitable for no-till agronomy due to their excessive canopy binding, which can cause difficulties during shredding or plowing-under their herbage (authors' experience).

When compared to conventional agronomy, green manuring in this trial was associated with additional agrotechnical operations required for crimson clover cover crop establishment and its herbage incorporation into soil prior to seeding the main maize crop. Further research is required to estimate total energy used in each of alternative fertilization variants (in fertilizers and in field operations). In order to offset the increase of fossil energy use for CC establishment and incorporation into soil, an animal work for powering the agrotechnical operations could be an option not to be neglected by small agricultural enterprises [17]. Research should be continued in more different environments (year × location combinations) in order to produce reliable evaluation of crimson clover's green manuring services.

CONCLUSION

The research in a year with favourable rainfall during maize vegetation has shown that green manuring with crimson clover for maize as a main crop delivers somewhat lower maize grain and aboveground mass yields than the conventional full-dose mineral fertilization on deep fertile soils around Osijek (Northeastern Croatia). When compared to conventional agronomy, green manuring was associated with additional agrotechnical operations required for crimson clover cover crop establishment and its herbage incorporation into soil prior to seeding main maize crop. Research should be continued in many different environments (year × location combinations) in order to produce reliable evaluation of crimson clover's green manuring services. Further research is required to estimate total energy used in each of alternative fertilization variants (in fertilizers and in field operations).

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