

# Biochar addition to the soil limits initial development of red clover (*Trifolium pratense* L.)

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## Abstract

Addition of biochar to the soil is recognized as one of the possible measures for moderation of climate change caused by the rising carbon emission to the atmosphere. Aim of this experiment is to explore the effect of addition of biochar to the soil and its effect on initial development of red clover. Experiment was set “in vitro” in a climate chamber. Length of leaves, mass of leaves, quantity of root nodules, and leaf absorption of red segment of spectrum were measured. Results show that differences between the biochar treated group and the control group in leaf mass and length were not significant. Number of root nodules in the biochar treated group was 53.3 % higher than in the control group. Absorption of red segment of spectrum in the control group was higher, suggesting higher rate of photosynthesis what is in accordance with more vigorous appearance of plants. All results show that fresh addition of biochar to the soil probably limits availability of some nutrients to plants.

Key words: biochar, carbon fixation, red clover, carbon sequestration, terra preta

## Dodatak biougljena supstratu usporava početni razvoj crvene djeteline (*Trifolium pratense* L.)

### Sažetak

Dodatak biougljena tlu jedna je od mogućih mjera posrednog ublažavanja klimatskih promjena uzrokovanih rastućom emisijom CO<sub>2</sub> u atmosferu. Cilj ovog istraživanja bio je utvrditi utjecaj dodatka biougljena tlu na početni razvoj crvene djeteline. Pokus je proveden „in vitro“ u klima komori. Mjereni parametri su dužina listova, masa listova broj korjenovih kvržica i lisna apsorpcija crvenog dijela spektra. Rezultati pokazuju da razlike između biougljenom tretirane grupe i kontrolne grupe u duljini listova i lisnoj masi nisu signifikantno različite, dok je broj korjenovih kvržica u biougljenom tretiranoj grupi bio 53,3% veći u odnosu na kontrolnu grupu. Apsorpcija crvenog dijela spektra u kontrolnoj grupi bila je veća što sugerira na veću razinu fotosintetsku aktivnost. Dobiveni rezultati pokazuju kako svježe dodavanje biougljena tlu vjerojatno ograničava biljkama dostupnost pojedinih nutrijenata iz tla.

Gljučne riječi: biougljen, fiksacija ugljika, crvena djetelina, sekvencijacija, terra preta

## Introduction

Carbon emission in atmosphere by human activity recently reached level of 15.4 Pg CO<sub>2</sub> year<sup>-1</sup> (Woolf et al., 2010). Naturally, CO<sub>2</sub> from atmosphere can be fixed through chemical and mostly biological processes, where photosynthesis converts between 150 and 175 Pg of carbon per year (Welp et al., 2011) what is not enough to stop rising trend of emission due to its cycling nature. Solution for carbon storing is to put it into soil in inert form, biochar (carbonised organic mater). Idea is not new, and practice of adding biochar to the soil i.e. *terra preta* is known to improve its fertility and has been practiced for thousands of years in the Amazonian basin. Carbon stored in soil in that way is stable for a long time, improving its fertility by rising ion exchange capacity and soil available water capacity. It also has positive effect on soil microorganisms and regulates soil acidity (Woolf et al. 2010). Biochar has positive effect on nutrient and agrochemical retention in soil reducing their leaching in to groundwater (Laird et al. 2010). It lowers needs of mineral fertilizers and sequestrates heavy metals in soil (Beesley and Marmiroli, 2011). Biochar production and implementation in agriculture can sequesterate up to 1,3 Pg of CO<sub>2</sub>, year<sup>-1</sup> without competing food production or agriculture land conversion (Woolf et al. 2010). The aim of this research is to explore the effect of addition of biochar to the soil and its effect on initial development of red clover.

## Material and methods

Impact of biochar addition to the soil was investigated in experiment set at Grassland research center, Faculty of Agriculture, University of Zagreb. Red clover (*Trifolium pratense*) was used as test model and sown in 6 experimental pots, where 3 presented control group with plain soil, and 3 with addition of biochar in volume ratio biochar:soil 1:3. Soil used in experiment is natural dystric brown soil from pasture on Medvednica mountain (15° 57' 44,74" - 15° 58' 53,65" λ E, and 45° 54' 43,40" - 45° 55' 53,07" φ N ) with pH 6.45. Experiment was conducted *in vitro* in climate chamber Microclima 1000 (Snijders Scientific, Netherlands) under controlled conditions and under two regimes for 80 days (Table 1, Table 2).

Table 1. Daily dynamics of parameters of first regime lasted 53 days

Segment number	Duration of in day (hours)	Luminous intensity (lux)	Relative humidity (%)	Temperature (°C)
1	12	0	90	20
2	2	50,000	85	25
3	8	100,000	85	25
4	2	50,000	85	25

Table 2. Daily dynamics of parameters of second regime lasted 27 days

Segment number	Duration of in day (hours)	Luminous intensity (lux)	Relative humidity (%)	Temperature (°C)
1	8	0	85	20
2	2	50,000	85	25
3	12	100,000	85	25
4	2	50,000	85	25

Experiment ended after 80 days in the beginning of flower emergence, when leaf length and dry leaf mass were measured. Leaves were dried in ventilated oven on 60°C on 48 hours.

Clover leaves were isolated and scanned on ScanJet G3010 (Hewlett-Packard Co, Palo Alto, CA) to obtain raster images which were analysed with Spatial Analyst, ArcGIS (ESRI, Redlands, CA). Zonal statistics was used to determine differences in values of red pixels which present absorption of segment of red spectrum which is involved in photosynthesis.

After isolation, roots were washed from soil and biochar particles and scanned on scanner. Acquired images were processed in ArcMap (ESRI, Redlands, CA) and counted. For statistical analysis and comparison, a simple descriptive statistics – student T-test – was used.

## Results and discussion

In biochar treated group germinate and grew 9 and in control 8 plants. By visual observation was found that leaf colour in biochar treated group was lighter green than control group. Further, plants in biochar treated group had fragile leaf petioles what indicated possible disruption in nutrient uptake or availability. After addition of biochar soil acidity changed from 6.45 to 6.95. Although, plants in biochar treated group with 13.74 cm had average leaf length lower for 1.91 cm than control group with average length 15,65 cm, that difference was not statistically significant ( $p=0.21$ ). Differences in leaf mass between biochar treated and control group statistically also, were not significant,  $p=0.45$  (1.02 g and 0.94 g, respectively). In contrary to recent research results which show that longterm addition and use of biochar in agriculture improve yield and biomass production (Vaccari i sur., 2011), short term addition did not affect photoactive biomass production in initial development of red clover.

On the other side, number of root nodules in biochar treated group was 53.3 % higher than in control group ( $p=0.002$ ), but these nodules were at least twice smaller than those in control group what means that they might have been inactive or low active.

Zonal statistics was performed on scanned images of leaves on 9840 pixels in 8-bit format. Difference between control and biochar group was confirmed and shows greater red light absorption in control group what suggest higher chlorophyll content and photosynthesis (Figure 1).

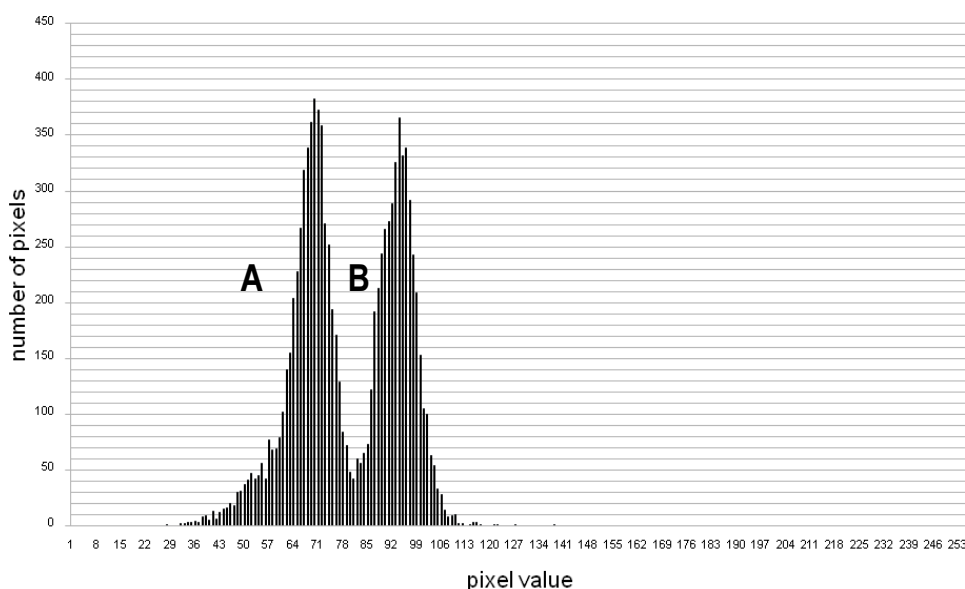


Figure 1. Histogram of pixel values present two distinctive groups: A-control group and B-biochar treated group.

## Conclusions

This experiment showed that fresh addition of biochar to soil in ratio biochar:soil, 1:3 limits initial development of red clover. This effect could be explained by big active surface of biochar which probably binded and blocked soil nutrients making them unavailable to the plants. Comparing results of this experiment with fact that *terra preta* is fertile soil suggest that biochar has to be saturated with nutrients through time and then it becomes nutrient repository with high ion exchange potential.

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