

# TOTAL AND HOT-WATER EXTRACTABLE CARBON RELATIONSHIP IN CHERNOZEM SOIL UNDER DIFFERENT CROPPING SYSTEMS AND LAND USE

## ODNOS UKUPNE ORGANSKE MATERIJE I UGLJENIKA RASTVORLJIVOG U TOPLOJ VODI U ČERNOZEMU NA RAZLIČITIM SISTEMIMA RATARENJA I KORIŠĆENJA ZEMLJIŠTA

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### ABSTRACT

A study was conducted to determine the hot water extractable organic carbon (HWOC) in 9 arable and 3 non arable soil samples on Haplic Chernozem. The hot water extractable carbon represents assimilative component of the total organic matter (OM) that could contain readily available nutrients for plant growth. The obtained fraction of organic carbon (C) makes up only a small percentage of the soil OM and directly reflects the changes in the rhizosphere. This labile fraction of the organic matter was separated by hot water extraction at 80°C. In our study the HWOC content in different samples ranged from 125 mg g<sup>-1</sup> to 226 mg g<sup>-1</sup>. On the plots that are under native vegetation, higher values were determined (316 mg g<sup>-1</sup> to 388 mg g<sup>-1</sup>). Whereas samples from arable soils were lower in HWOC. It was found that this extraction method can be successfully used to explain the dynamics of the soil OM. Soil samples with lower content of the total OM had lower HWOC content, indicating that the preservation of the OM depends on the renewal of its labile fractions.

**Keywords:** organic matter, hot water extractable carbon, Chernozem, cropping systems

### Detailed Abstract in Native Language

U radu su prikazani rezultati ispitivanja sadržaja organskog ugljenika ekstrahovanog toplom vodom (HWOC) sa oraničnih parcela i zemljišta pod prirodnom vegetacijom. Za potrebe ovog istraživanja uzeti su uzorci na 9 različitih sistema ratarenja sa različitim kombinacijama đubrenja, odnosno sa livade, šume i ruderalnog staništa. Ugljenik rastvorljiv u toploj vodi se smatra labilnom frakcijom (LF) organske materije koji se dobija ekstrakcijom u vodenom kupatilu na 80°C. Analiza ukupne organske

materije (OM) je pokazala da postoji opadajući trend njenog sadržaja u zemljištu. Uzorci poreklom sa ne poljoprivrednog zemljišta su imali najveći sadržaj ukupne OM. Poređenjem oraničnih površina, parcele đubrene stajnjakom imaju veći sadržaj ukupne OM u poređenju sa parcelama koje se đubre samo sa mineralnim đubrivima. Sadržaj HWOC na oraničnim površinama se kretao od  $125 \mu\text{g g}^{-1}$  do  $226 \mu\text{g g}^{-1}$ . Na parcelama pod prirodnom vegetacijom dobijene su veće vrednosti HWOC ( $316 \mu\text{g g}^{-1}$  do  $388 \mu\text{g g}^{-1}$ ). Utvrđeno je da primenjena metoda ekstrakcije može da posluži u opisivanju dinamike OM zemljišta. Uzorci zemljišta sa manjim sadržajem ukupne OM imali su i manji sadržaj HWOC što ukazuje da očuvanje OM zavisi od obnavljanja njene labilne frakcije.

**Ključne reči:** organska materija, ugljenik rastvorljiv u toploj vodi, černoze, sistemi ratarenja

## INTRODUCTION

Throughout the history the explanation of the soil organic matter (OM) concept has had a long evolution and it is explained in detail in the work of Manlay et al. (2007) and includes three periods of (a) humic period, (b) mineralist period and (c) the ecological period. As a result, today the OM in soil is simultaneously defined as the qualitative ecological component and the chemical substance. One of the common viewpoints of "ecological" and "chemical" approaches is that the OM can be explained as a mixture of abundant chemical fractions that differ in the decomposition intensity. The complexity arises from the pathways of the OM synthesis, climatic conditions, soil characteristics, land use patterns and vegetation cover. In order to obtain a fraction of the OM relevant to the identification of changes in its content in the soil caused by agricultural practices or to preserve its level there are many physical, chemical and biochemical methods (Cheng and Kimble, 2001). At the same time, the use of new techniques in organic chemistry (chromatography, analytical pyrolysis, nuclear magnetic resonance, isotopes, etc..) confirmed the soil OM as "one of the most complex natural substances" or ecosystem components that differs in polymericity and aromaticity (Skjemstad et al., 1997). Several national studies have shown a decline in the OM content with tillage, insufficient fertilization, and removal and burning of crop residue (Nesic et al., 2008; Sekulić et al., 2010; Ličina et al., 2011; Belić et al., 2013). Patterns of reduced OM have been observed regardless of climate, soil type or grown crops. Generally, the rate of loss slows as SOC levels reach a new equilibrium that depends on tillage practices and the level of C inputs returned to the soil as crop residue or animal manures. On the other hand at natural soil the OM changes are caused solely by the pedogenetic processes. The active soil organic matter (the fresh, labile, nutritious part of the humus) is greatly affected by the applied agrotechnical measures. Therefore labile OM is the most dynamic reservoir of the organic carbon in the soil (Janzen et al., 1998) and an indicator of the soil production capacities (Bremer et al., 1995). This fraction makes up only a few percent of the total soil OM and is directly dependent on the changes in the rhizosphere. It can be described with the free granular OM which is not absorbed – closed inside the structure aggregates and the intra-aggregate granular OM which is inside the structure aggregates (Cambardella i Elliott, 1993). Due to a great significance of the soil OM, which is the presumption of the soil fertility and productivity, it is necessary to determine the nutritious part of the OM, i.e. the labile fraction immediately accessible to the plants. Since the soil organic matter content

decreases, there is a need to define in which way this reflects on the changes in the labile fraction content and to which extent it can affect the soil productivity.

## MATERIAL AND METHODS

The long-term stationary experiments crop rotation „Plodoredi“ (Milošev, 2000; Šeremešić, 2012), and a long-term experiment „IOSDV“ - Der Internationale Organische Stickstoff- Dauerdüngungsversuch (Starčević et al., 1997) that were involved in the study are located at the Institute of Field and Vegetable Crops of Novi Sad. The trial with crop rotations was set up in 1946/1947, and it still exists with lesser changes. The 3-year crop rotation consisted of maize, soybean and winter wheat, the 2-year crop rotation consists of wheat and maize, and maize and winter wheat are grown in the monoculture. The IOSDV experiment was set up in 1984/1985 and it involves a 4-year crop rotation which consists of sugar beet, winter wheat, maize and spring barley. The methodology of the experiment performance involves the application of the organic and mineral fertilizers or the plowing of the crop residue according to the following layout:

1. A2B1- Animal manures 40 t ha<sup>-1</sup> with no N fertilizer, without plowing the crop residue-(BØ).
2. A2B4-Animal manures 40 t ha<sup>-1</sup>+ 100 kg N ha<sup>-1</sup>, without plowing the crop residue- (B2).
3. A1B5 -mineral N fertilizers 200 kg ha<sup>-1</sup> without plowing the crop residue--(A4).
4. A3B5-Plowing the crop residue+ 200 kg ha<sup>-1</sup> N (C4).
5. Unfertilized wheat two-field which was not fertilized from its origin and with plowing the crop residue since 1987(N2).
6. Unfertilized wheat three-field which was not fertilized from its origin and with plowing the crop residue since 1987(N3).
7. Wheat monoculture mineral N 100 kg, 50 kg ha<sup>-1</sup> in the autumn + 50 kg ha<sup>-1</sup> u prihranjivanju, with plowing the crops residue(MO).
8. Fertilized wheat two-field mineral N 100 kg (50 kg ha<sup>-1</sup> in the autumn + 50 kg ha<sup>-1</sup> supplemental fertilizing) with plowing the crops residue(D2)
9. Fertilized wheat three-field mineral N 100 kg (50 kg ha<sup>-1</sup> in the autumn + 50 kg ha<sup>-1</sup> supplemental fertilizing) with plowing the crops residue(D3)

Non agricultural soil samples were taken from the following locations:

10. Natural (virgin) soil –virgin soil, ruderal habitat where weed of a ruderal type of vegetation mainly grows(RS)
  11. Meadow – virgin soil (N) 45° 20,549', (E) 19° 51,492', a 81 m elevation(L)
  12. Oak forest– virgin soil (N) 45° 20,591', (E) 19° 51,374', a 83 m elevation(S)
- All of the treatments examined are from the soil type Chernozem on loess..

For the purpose of determining the soil OM content in the soil samples, the titrimetric Tyrin method was used and the soil organic matter content was calculated by multiplying the C content in the sample with the correction factor  $f=1,724$ . This factor is based on the assumption that the soil OM consists of 58% of the organic carbon (USDA, 1996). To separate the labile fraction of the hot water extractable organic matter (HWOC) in the soil the modified Ghani (2003) procedure was used. In our study a 0-20 cm layer of soil was analyzed as the significant changes were expected in that layer. The samples were obtained in a disturbed state and were kept in the laboratory air-dried, up until the moment the analysis was performed. Grinding the samples in a mill and sieving through a sieve with 2 mm diameter preceded the

analysis. 10 g of soil sample were put in a 50 ml cuvette (< 2 mm) and 40 ml of distilled water was added to the air-dried soil. The cuvettes were put into a horizontal shaker on 30 rpm for a period of 30 min. After that the samples in the cuvettes were transferred into a steam hot-water bath on a 80 °C temperature, for a period of 16h. The next phase involved the centrifugation on a MSEC centrifuge (Measuring & Scientific Equipment LTD., London) with 5000 rpm for a period of 20 minutes. After the centrifugation, the supernatant was filtrated through a 0,45 µm filter. The filtrate was put into oven until absolutely dry. After drying in the oven the filtrate (dry residue) was analyzed for the organic C content according to Tyrin method.

The data on the organic matter content and the HWOC were statistically assessed using the analysis of the variance method on a significance level of  $\alpha=0,05$ , and the LSD test was used for individual comparisons of the treatments' means. The linear regression was conducted without transforming the data. The data were statistically processed by using the statistical program STATISTICA 8.0 series 608c.

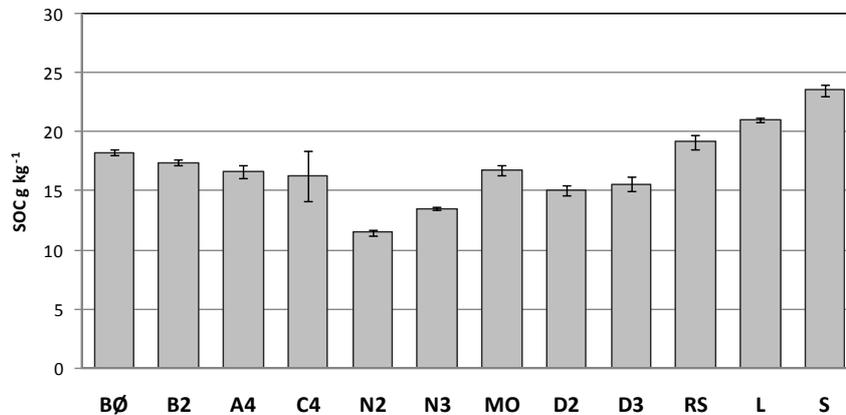
## RESULTS AND DISCUSSION

### *Total soil organic matter content*

By analyzing the cropping system on a long-term experiments and comparing the values determined in the earlier studies (Molnar and Stevanović, 1986; Starčević et al., 1997) significant changes were determined. The assumption is that the soil undergoes the stabilization period of the OM level after the experimental setup, as a consequence of synchronizing with the agro-ecological conditions, the intensity of the applied management practice and other soil characteristics (Blum, 2008).

Subsequent to experimental set up, OM increase was observed on the BØ and B2 plots, where animal manures were used compared with earlier studies (Starčević et al., 1997). The decline of the OM can be explained in 3 ways: (1) natural loss of the OM which began when the arable land was established, (2) the loss due to plowing and/or deepening the arable layer, (3) the loss which can be attributed to the intensifying of the mineralization due to temperature increase (Sleutel, 2006). The loss of the OM from the soil due to tillage is regularly attributed to the natural fertility loss, after deterioration of the water, air and temperature soil regime (Diaz-Zorita et al., 1999), or poor manipulation with crop residue (Sekulić et al., 2010). The soil samples obtained from the natural soils (ruderal, meadow and forest) had OM level lower than samples taken 50 years ago on the same location (Živković et al., 1972). When compare the OM content on the plots fertilized by animal manures (BØ i B2) and native soil (RS, L i S), differences were determined, which speaks in favor of the fact that the application of the organic fertilization on the cropped soil can affect the preservation of the OM, in soils with lower OM content. These results coincide with the research on a long-term experiments in Bad Lauchstadt-u (Blair et al., 2006) where similar results were observed. The assumption is that the steady OM pool in the soil temporarily can lead to a balanced state called „equilibrium“. In our agro-ecological conditions, on the unfertilized treatments N2 and N3, (which began in 1946/47) the stabilization cycle of the OM level lasted for approximately 40 years. Our results correspond with ne with Jankinson (1991) study who concluded that the OM stabilization period varies in the 30-50-year interval for Rothamsted experiments. The stable OM content at the unfertilized plots could be also explained with presence of  $Ca^{2+}$  (Six et al., 2002). Therefore it can be said that the establishment of the OM equilibrium in our condition is possible only if the soil OM level is low or lasts temporarily for a period of several years if there is a high level of OM in soil. A relatively high OM was determined on a wheat MO compared to the fertilized 2-year

and the 3-year rotation can be explained by a smaller number of operations throughout the year and the length of the land covered with crops on MO. A significant variation of the OM content was determined on the C4 plot which can be explained by the differences in the amount of the crop residue plowed between the repetitions on the plots from which the samples were obtained. The OM content on N2 is only 63% of the OM compared to the higher recorded values at BØ plot. However, if the average soil organic carbon content is  $25,92 \text{ g kg}^{-1}$  found in 1950s on same experimental station (Bogdanović, 1955), a relative yearly loss of the OM on the N2 plot would be  $0,28 \text{ g kg}^{-1} \text{ year}^{-1}$  or  $0,13 \text{ g kg}^{-1} \text{ year}^{-1}$  at the BØ plot. The intensity of OM loss was more pronounced at the beginning, and later it was smaller.



Graph. 1. Total organic carbon content in the investigated treatments ( $\text{g C kg}^{-1}$ )

#### *Labile HWOC content in the soil samples*

Based on the results of the one-way analysis of the variance, it has been determined that the treatments affected the HWOC dynamics, which led to statistical differences among them (Graph 2). The obtained results are deriving from the differences in the cropping technology, i.e. the changes of the initial OM content in the non-agricultural soil. Labile fraction represented with HWOC ranged from  $125 \mu\text{g g}^{-1}$  to  $226 \mu\text{g g}^{-1}$ . On the plots under native vegetation higher values were recorded (from  $316 \mu\text{g g}^{-1}$  to  $388 \mu\text{g g}^{-1}$ ) compared to arable plots. In the Chen et al. (2009) research the average HWOC values in the arable soil of Cambisol ranged from  $375 \mu\text{g g}^{-1}$  to  $273 \mu\text{g g}^{-1}$ , in the plow and the deeper soil layer. The obtained values can be related to the microbiological activity which was more intensive under native soil due to increased humidity of the soil during sampling period. Apart from that, it is considered that the root activity and the fresh organic matter production (root exudates) and the way of manipulation with plant residue also affect the labile fraction of OM. The changes in the light fraction of OM can be a reliable indicator of the intensity of its assimilation by microorganisms. Dalal and Mayer (1986) have determined that the changes of the light fraction are 11 times the size of the stable fraction of the OM. The relationship between HWOC/SOC is an indicator of the labile fraction share of the OM in the total OM. According to our study a large ratio of the labile fraction was determined on the soil samples under native vegetation. Based on these results it can be seen that the unfertilized plots have a HWOC/SOC ratio identical with the fertilized plots, and the highest values of the relationship examined were recorded on the native soil. The HWOC values obtained in our study are 1,1-1,75% of the total soil OM content. According to Ghani et al. (2003) the soils used as grasslands have HWOC reservoir values which vary 3-6%, while Šimon (2008) stated the HWOC values comparable to

our results. According to the results from Soon et al. (2007) on a Luvisol soil type the HWOC is 3,5 do 4,8% of the total OM .

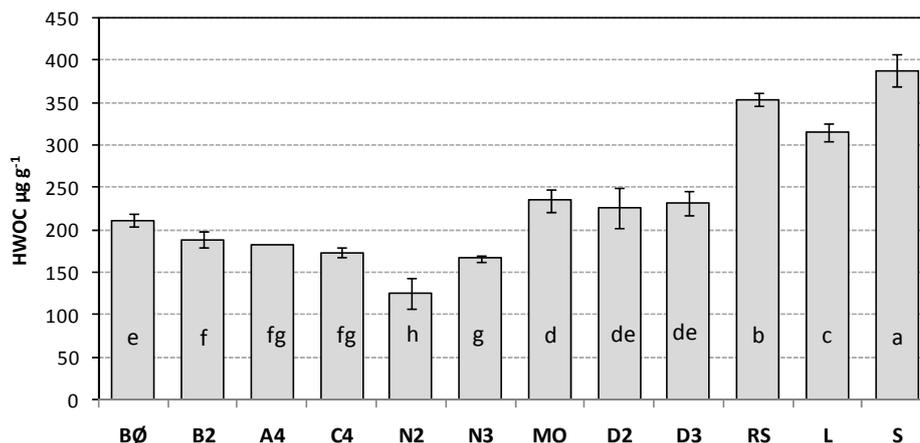
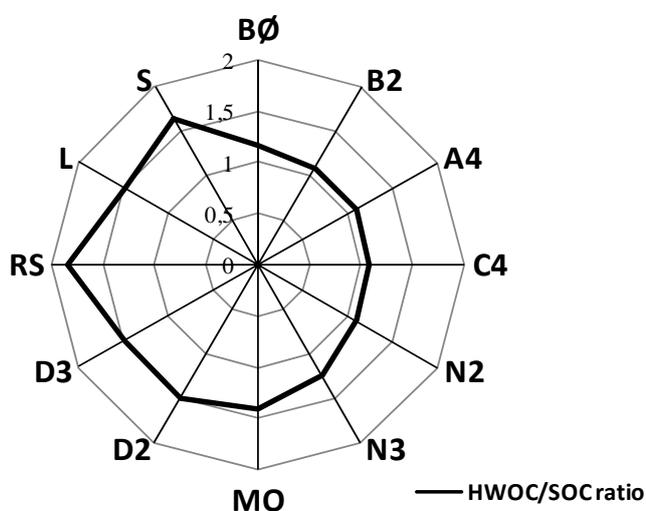


Figure 2. Hot water extractable carbon (HWOC) in soil samples

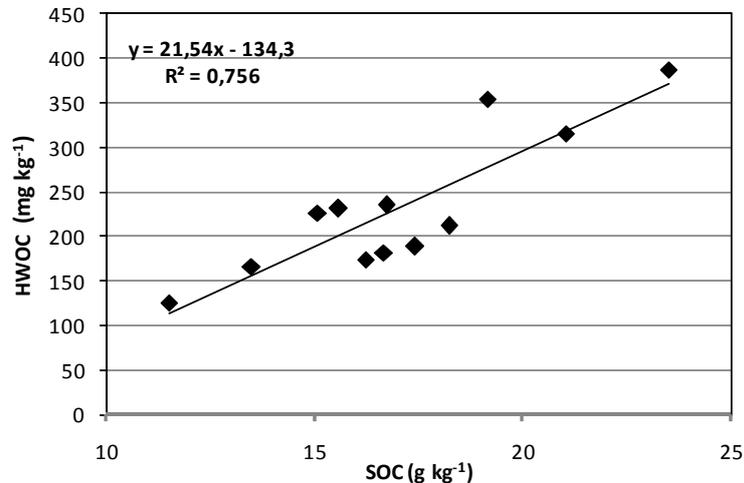
Since the HWOC is considered as an indicator which reflects the microbiological activity to a great extent, the differences which exist between the treatments can derive from the soil biogenesis (Sparling et al., 1998). Based on the above mentioned facts, it is assumed that the number of the microorganisms was not a limiting factor, but the accessibility of the substrate for the microbiological degradation. The total HWOC content might be an indicator of general soil fertility as there is more HWOC in the soil with the higher OM content, due to the processes of intensive transformation of fresh organic matter. As far as the fertilized treatments are concerned, the application of the mineral nitrogen can increase the microbiological activity and therefore the assimilation of the significant part of the labile organic matter. Higher OM and HWOC in the soil are related with aggregation of soil particles (Tobiašová, 2012). Natural forest, meadow and ruderal habitat generally could improve soil aggregation which brings the protection of OM in soil.



Graph. 3. Relationship between hot water extractable carbon and total organic matter

By comparing the SOC and the HWOC contents, a positive correlation ( $r=0,756$ ) was determined. With an increase of the soil OM content unit, the HWOC is also increased by 21,  $\mu\text{g g}^{-1}$ . The obtained result is in compliance with the literature values (Sparling et al., 1998). It is assumed that the higher amount of the total soil OM

affects the more intensive mineralization processes and a higher HWOC content. Therefore the increasing the total OM in soil with manure and crop residue management will lead to higher OM and preservation of soil fertility. Likewise Šeremeši (2012) noted that maintenance of average 3% of OM in the topsoil will require incorporation of 750 C g ha<sup>-1</sup> with crop residues each year.



Graph. 4. Coefficient of correlation between HWOC and SOC

## CONCLUSION

Based on the obtained results, a loss of total soil organic matter was determined, as a consequence of the anthropogenic influence. From the arable soil samples it was determined that the hot water extractable carbon content is 125  $\mu\text{g g}^{-1}$  to 226  $\mu\text{g g}^{-1}$ . The soil samples which originate from the natural habitat, the meadow, the forest and ruderal habitat showed that they have the highest concentration of organic matter labile fraction (316  $\text{mg g}^{-1}$  to 388  $\text{mg g}^{-1}$ ) which indicated its intensive loss on the arable soils. The hot water extractable carbon that comprises only a few percent of the total soil organic matter is the most sensitive part of OM which is directly dependent on the rhizosphere activity. This is why its content can be considered as one of the main indicators of the changes in the organic matter content and is a qualitative indicator of the soil potential for the realization of the soil productivity potential.

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