## HOW DIFFERENT MULCH MATERIALS REGULATE SOIL MOISTURE AND MICROBIOLOGICAL ACTIVITY?

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## Abstract

As a result of climate change, the frequency of droughts across Europe is showing an increasing trend. The solution to this problem can be various soil cover techniques, which help to preserve soil moisture and soil biological activity, thereby increasing crop yield. In our research, we examined how different mulch materials affect the regulation of soil moisture and the microbiological activity of the soil. The experiments were set up on two sites: a sandy soil with a low soil organic matter content (Királyhalom, Serbia) and an coarse loamy soil with a higher water capacity (Thessaloniki, Greece). The treatments were set up in 4 repetitions with pepper plants (Capsicum annuum L.). Daily intensive irrigation with wool mulch, agrotextile, straw mulch and control treatments was used on the Serbian site. The irrigation was carried out every 6 days at the Greek site, where the treatments were as follows: wool mulch with plants, wool mulch without plants, plants without mulch and the control without plants. The yield was measured, and the moisture content,  $\beta$ -glucosidase activity and active (permanganate oxidizable) carbon content of the soils at the time of sampling were examined every 2 weeks. In the Serbian area, wool mulch showed significantly lower  $\beta$ -glucosidase activity compared to the other treatments. This was presumably due to the good water absorption capacity of the wool mulch and the fact that the soil could not aerate in addition to the intensive irrigation, thus the enzyme activity decreased. All mulch treatments increased the active carbon content compared to the untreated control, with the straw mulch to the greatest extent. In the extensively irrigated Greek soil,  $\beta$ -glucosidase was significantly higher in pepper plots covered with wool mulch compared to the control area without plants, but also higher compared to pepper plots not covered with wool mulch. The higher yield results measured on the mulched plots indicate that, in addition to water retention, biological activity also plays a major role in the development of yields. The effect of each mulching method on soil biological activity depends on the degradability of the mulch material, the frequency of irrigation, and the soil's waterholding capacity.

**Keywords:** *living mulch, agro textile, wool mulch,*  $\beta$ *-glucosidase, soil active carbon* 

# HOGYAN SZABÁLYOZZÁK A KÜLÖNBÖZŐ MULCS ANYAGOK A TALAJNEDVESSÉGET ÉS A MIKROBIOLÓGIAI AKTIVITÁST?

# Összefoglalás

A klímaváltozás következtében az aszály gyakorisága Európa szerte növekvő tendenciát mutat. Erre a problémára megoldást jelenthetnek a különböző talajtakaró technikák, amelyek segítenek a talajnedvesség és talajbiológiai aktivitás megőrzésében, ezáltal a terméshozam növelésében. Kutatásunk során arra kerestük a választ, hogy a különböző mulcs anyagok hogyan hatnak a talajnedvesség szabályozásán keresztük a talaj mikrobiológiai aktivitására. A kísérleteket két termőhelyen, egy gyenge humusz tartalmú homoktalajon (Királyhalom, Szerbia)

és egy nagyobb vízkapacitású vályog talajon (Thesszaloniki, Görögország) állítottuk be 4 ismétlésben paprika növénnyel (Capsicum annuum L.). A szerbiai termőhelven napi gyakoriságú intenzív öntözést alkalmaztunk gyapjúmulcs, agroszövet, szalmamulcs és kontroll kezelésekkel. A görögországi termőhelyen 6 naponta történt az öntözés, ahol a kezelések a következők voltak: gyapjúmulcs növénnyel, gyapjúmulcs növény nélkül, növény mulcs nélkül és a növény nélküli kontroll. Mértük a terméshozamot, valamint 2 hetente vizsgáltuk a talajok *mintavételkori nedvességtartalmát*, β-glükozidáz aktivitását és aktív (permanganát oxidálható) széntartalmát. A szerbiai területen a gyapjúmulcs a többi kezeléshez képest szignifikánsan alacsonyabb ß-glükozidáz aktivitást mutatott. Ennek feltehetően az lehetett az oka, hogy a gyapjúmulcs jó vízfelvevő képessége és az intenzív öntözés mellett a talaj nem tudott szellőzni, ezálatal az enzim aktivitás csökkent. Mindegyik mulcs kezelés növelte az aktív széntartalmat a takaratlan kontrollhoz képest, a legnagyobb mértékben a szalmamulcs esetében. Az extenzíven öntözött görögországi talajon a  $\beta$ -glükozidáz a gyapjúmulccsal takart paprika parcellák esetében szignifikánsan magasabb volt a növény nélküli kontroll területhez képest, de magasabb volt a gyapjúmulccsal nem takart paprika parcellákhoz képest is. A mulcsozott parcellákon mért magasabb terméseredmények rámutatnak arra, hogy a vízmegtartás mellett a biológiai aktivitásnak is nagy szerepe van a terméshozamok alakulására. Az egyes mulcsozási módok talajbiológiai aktivitásra gyakorolt hatása a mulcsanyag lebomló képességétől, az öntözés gyakoriságától és a talajok víztartó képességétől is függ.

Kulcsszavak: élő mulcs, agroszövet, gyapjúmulcs, β-glükozidáz, talaj aktív szén

### Introduction

Population growth, global warming, and climate change affect the water supply of the agriculture (KADER et al., 2017a). One of the biggest problems for agricultural sectors is drought. The drought causes significant economic, social and environmental damage, the severity of which has increased in recent times in the Carpatian Basin as well. Drought makes agricultural production impossible without irrigation, especially on sandy soils with low water capacity. Wind erosion is an other significant problem, especially in poorly structured sandy soils (KERTÉSZ et al., 2012a). The effect of wind erosion can be indirect or direct. Its direct effect means worse damage, for example, the crop area decreases, and the productivity of the soil decreases (JAKAB et al., 2010; MEZŐSI et al., 2017).

Different soil covers can be a solution to this problem. The soil covers used in agriculture can be classified into three groups: natural, artificial (e.g. agro textile), and special (e.g.sand) (KADER et al., 2017a). Natural mulches can be living and non-living (e.g. straw) mulch. Waste fibres such as wool can be the basis of innovative soil water storage technologies (MARCZAK et al., 2022). Soil covers help conserve soil moisture (KERTÉSZ et al., 2012; FEKETE et al., 2021), increase yield (GHOSH et al., 2016; KOVÁCS et al., 2020), and prevent weeding (PARMAR et al., 2013; SHARMA et al., 2022) and soil erosion (SZARKA et al., 2015; MADARÁSZ et al., 2021). Plants and living organisms need soil moisture for nutrition uptake. Water is not only a necessary essential transport medium for substrates but also plays an important role in hydrolysis. Therefore, microbial activity is regulated by soil water content (YAN et al., 2015; KOCSIS et al., 2015; KOTROCZÓ et al., 2020). CHAKRABORTY et al., (2008) described in their article that the combination of irrigation and soil cover ensures better water absorption in the case of winter wheat, and the number of irrigations is reduced due to the surface cover effect. Soil covers also help maintain soil health (NGOSONG et al., 2019).

Most mulches can be said to increase the soil microbe population (MANNA et al., 2018), and their activity (LI et al., 2018; BUTCARU et al., 2020). The microbiological properties of the soil are closely related to most of the soil's organic matter content (DEMETER et al., 2013;

BÉNI et al., 2017; KOTROCZÓ and FEKETE, 2020) and water content (BOROWIK et al., 2016; KOCSIS et al., 2022). Organic materials protect the moisture and structure of soils, reducing climatic stress effects, and soil life ensure the cycle of nutrients and the availability of reserve nutrients and bound water for plants (BIRÓ, 2002). The water content in the soil affects the physiological state of microorganisms. Therefore, properly moist soil supports more diverse microbial communities (BOROWIK et al., 2016). Fungi and bacteria release extracellular enzymes such as phosphatase and  $\beta$ -glucosidase. These enzymes have an important function in the mineralization of soil organic matter (KOTROCZÓ et al., 2014). So, mulching has an important function in increasing the water-holding capacity and biological activity of soils, and in increasing their ability to supply nutrients. However, very little scientific data can be found on the soil biological effects of different mulch materials (KADER et al., 2017a; LAL et al., 2020). Most of the research focuses primarily on soil moisture, soil temperature and the effects on the crop yield (KADER et al., 2017b; ADEKALDU et al., 2021; MARCZAK et al., 2022).

Therefore, the aim of our research was to investigate how different mulch materials (wool mulch, agro textile, straw) affect the activity of microbes in the soil and the crop yield under irrigated field conditions.

## Material and methods

### Material

The experiment was carried out in two areas with different climates and soil types (in Serbia and Greece). The Serbian site is characterized by carbonate sandy soil with low water capacity and organic matter content (Table 1). The Greek site has coarse-loamy neutral soil with a low organic matter content (Table 1). The average daily mean temperature during the growing season was 26.4 °C at the Greek site and 24.5 °C at the Serbian site.

Site location	Texture	pH (CaCl <sub>2</sub> )	CaCO3 (m/m %)	Soil Organic Matter (m/m %)
Serbian site (Királyhalom)	sandy	7.44	3.57	0.30
Greek site (Thessaloniki)	Greek site coarse- Thessaloniki) loamy 6.9		0.00	1.57

Table 1. Soil physical and chemical properties of the experimental sites.

The experimental plant in both places was pepper (*Capsicum annuum L*.). At the Serbian site, we used pepper of the Amy type, which does not require a support system and has good yields (https://zoldseg-palanta.hu). In the Greek site, we used the Flavorburst F1 variety, which has a high yield, requires a support system and a lot of light (https://www.burpeehomegardens.com). The Serbian site was covered with wool mulch, agro textile, and straw, and there was an uncovered control. There were the following four treatments at the Greek site: a plant with mulch, a plant without mulch, mulch without plant, and uncovered control.

#### Methods

4 plots (replications) were planted per treatment. We planted 10-10 plants on the Serbian plots and 8-8 plants on the Greek plots (approx.  $2 \text{ m}^{-2}$  plots). The pepper were planted at the end of June. To reduce the impact of environmental factors, the experiment was surrounded by a protective strip. Nutrient supplementation was based on the nutrient requirements of the plants. Daily intensive irrigation was used at the Serbian site, while surface irrigation was used every 6 days at the Greek site. The soil sampling was carried out every two weeks (4 times during the growing season) before the actual watering from the upper 0-10 cm soil layer. Also, soil temperature was measured a few times at the Greek site.

In measuring the  $\beta$ -glucosidase activity, we used SINSABAUGH et al., (1999) method with a minor modification, according to KOTROCZÓ et al., (2014) study, increasing the concentration of the buffer and terminal solutions. Active Carbon was measured according to WEIL et al., (2003) with the change in concentration of KMnO<sub>4</sub> used to estimate the amount of carbon oxidized. The method was modified by shaking of 1 g of air-dry soil in 10 mL 0.02 M KMnO<sub>4</sub> solution for 5 minutes and the absorbance measured at 565 nm wavelength using Biochrom Libra S22 spectrophotometer. Soil moisture was determined from the soil sample using a gravimetric method. The yields were characterized by the pepper mass harvested during the entire vegetation period per plant.

All statistical analyses were performed using R version 4.2.1 (R Core Team, 2018) and dplyr version 1.0.10 (WICKHAM and FRANCOIS, 2015) as well as ggplot2 packages. We tested the differences in the effect of treatments towards glucosidase activity among the mulching groups using the Kruskal-Wallis test with subsequent non-parametric Dunn's test comparisons. The assumption of normality of the residuals was checked using the Shapiro-Wilk test. Differences in labile carbon activity among the mulch treatments were tested with an one-way ANOVA followed by posthoc comparisons using Tukey's HSD method. The homogeneity of variances was checked using Levene's test. We report partial eta squared ( $\eta$ 2) values as the effect size indicator for ANOVA, which is essentially the same as the fitted R-squared indicator used in regression analysis). The  $\eta$ 2 measures the proportion of the variability in the outcome variable that can be explained in terms of the predictor. The value of this indicator can range from 0 to 1.00. Zero indicates independence and 1.00 indicates a deterministic relationship.

### **Results and Discussion**

At the Serbian site,  $\beta$ -glucosidase activity differed significantly among the mulch types (H=18.21, p<0.001,  $\eta^2$ =0.34). Follow-up tests demonstrated a significant difference between the straw and wool mulch (p<0.001), between the control and the wool mulch (p<0.01) and between the agro textile and wool mulch (p<0.05) (Fig. 1). The order of the average  $\beta$ -glucosidase activity for each treatment was as follows: straw (1.69 µmol g<sup>-1</sup> h<sup>-1</sup>), agro textile (1.322 µmol g<sup>-1</sup> h<sup>-1</sup>), control (1.45 µmol g<sup>-1</sup> h<sup>-1</sup>) and wool mulch (0.82 µmol g<sup>-1</sup> h<sup>-1</sup>). The highest  $\beta$ -glucosidase activity was therefore measured in the case of organic mulch (SM) that decomposes more easily. While, on average, the lowest activity was experienced in the case of wool mulch.

In contrast to our results, LI et al., (2016) found that, on strawberry plots,  $\beta$ -glucosidase activity of wool much treatment was higher compared to the control, whereas in our case there was no significant difference. Furthermore, when agrotextile was applied, WANG et al., (2020) found that beta-glucosidase activity was decreased compared to the control, while there was no negative effect in our case.



Figure 1. Data summary of  $\beta$ -glucosidase activity at the Serbian site with a box-andwhisker-plot indicating the outliers and variability outside the upper and lower quartiles, classified by the effect of treatments: C: control, WM: wool mulch, AT: agro textile, SM: straw mulch. The results of the multiple comparisons for a one-way ANOVA model are indicated by the lines above the graph. Statistical significance was determined via *posthoc* test, where '\*'p<0.05 '\*\*'p<0.01 '\*\*\*'p<0.001.

At the Serbian site, the level of active carbon differed significantly among the mulch types  $(F_{3,20}=13.18, p<0.001, \eta^2=0.66)$  (Fig. 2). Follow-up tests demonstrated a significant difference between the agro textile (mean: 361.19 mg kg<sup>-1</sup>) and control (p<0.001) (mean: 249.80 mg kg<sup>-1</sup>). The straw (mean: 387.26 mg kg<sup>-1</sup>) was also significantly different from the control and the wool mulch (mean: 299.57 mg kg<sup>-1</sup>). So we found that all mulches increased the amount of active carbon in the soil compared to the control.

Similar to our results, YU et al., (2007) found that straw mulch can increase soil labile carbon content compared to other treatments. In a study of bacterial and fungal species and soil respiration, BUTCARU et al., (2017) found higher biological activity in areas covered with wood mulch and combined with cover crops than in areas covered with cotton mulch and cover crops.

At the Serbian site, because of the intensive irrigation, there was no major difference in soil moisture among the treatments. The average soil moisture content at the time of soil sampling was 10.54%, 8.72%, 8.33% and 7.89% for agro textile, straw, wool mulch and control plots. HOOVER (2000) also found no difference in soil moisture between different mulch types under intensive irrigation. Soil moisture contents are therefore not always related to soil biological parameters.



Figure 2. Data summary of active carbon at the Greek site with a box-and-whisker-plot indicating the outliers and variability outside the upper and lower quartiles, classified by treatments: C: control, WM: wool mulch, AT: agro textile, SM: straw mulch. The results of the multiple comparisons for a one-way ANOVA model are indicated by the lines above the graph. Statistical significance was determined via *post-hoc* test, where '\*'p<0.05 '\*\*'p<0.01 '\*\*\*'p<0.001

In the case of Greek site, the  $\beta$ -glucosidase activity of control plots (mean: 0.152 µmol g<sup>-1</sup> h<sup>-1</sup>) were significantly lower compared to the plant with wool mulch plots (p<0.001) (mean: 0.956 µmol g<sup>-1</sup> h<sup>-1</sup>) (Fig. 3). In the case of wool mulch without plants, the  $\beta$ -glucosidase activity was also higher (mean: 0.745 µmol g<sup>-1</sup> h<sup>-1</sup>) compared to the control, although the difference was not significant due to the high standard deviation of the data. In contrast to the Serbian site, the  $\beta$ -glucosidase activity in the soil increases as a result of the wool mulch on the Greek site.



Figure 3. Data summary of  $\beta$ -glucosidase activity at the Greek site with a box-andwhisker-plot indicating the outliers and variability outside the upper and lower quartiles, classified by the effect of treatments: C: control, WM: wool mulch without plant, PW: a plant with wool mulch, P: plant without mulch. The results of the multiple comparisons for a one-way ANOVA model are indicated by the lines above the graph. Statistical significance was determined via *post-hoc* test, where '\*'p<0.05 '\*\*'p<0.01 '\*\*\*'p<0.001.

Active carbon levels on the Greek plots also differed significantly among the treatments and follow-up tests ( $F_{3,8}$ =5.57, p<0.05,  $\eta^2$  =0.69) demonstrated a significant difference between wool mulch without plant (WM) (mean:1226.83 mg kg<sup>-1</sup>) and control (p<0.05) (mean: 948.36 mg kg<sup>-1</sup>) (Fig. 4). However, the plant with wool mulch and plant without wool mulch treatments did not differ significantly from each other, although their active carbon concentrations (1032.495 and 1039.605mg kg<sup>-1</sup>) were on average higher compared to the control.

At the Greek site, the average soil moisture content at the time of soil sampling was 21.35%, 15.78%, 15.48% and 10.68% for mulch without plant, plant without mulch, plant with mulch and control. So, soil moisture contents are usually related to soil biological parameters. Soil moisture is therefore a limiting factor for soil biological activity, as confirmed by many authors (YU et al., 2007; LI et al., 2016; BUTCARU et al., 2017).



Figure 4. Data summary of active carbon at the Greek site with a box-and-whisker-plot indicating the outliers and variability outside the upper and lower quartiles, classified by treatments: C: control, WM: wool mulch without plant, PW: a plant with wool mulch, P: plant without mulch. The results of the multiple comparisons for a one-way ANOVA model are indicated by the lines above the graph. Statistical significance was determined via *post-hoc* test, where '\*'p<0.05 '\*\*'p<0.01 '\*\*\*'p<0.001.

As shown in Fig 5, at the Greek site, the highest average soil temperature in all cases was in the control treatment (11 August: mean: 35.84 °C, 17 August: 30 °C, 18 August: 33 °C. 3°C, Aug. 20:33°C), while the lowest soil temperature was in the plant with mulch treatment (PW) (Aug. 11: mean: 29.6°C, Aug. 17: 29.5°C, Aug. 18: 28°C, Aug. 20: 28.3°C). This interaction can be explained by the fact that the presence of wool mulch has some insulating effect (HOOVER, 2000), which helps to regulate soil temperature, and the presence of plants also helps to reduce soil temperature.



Figure 5. Soil temperature (°C) on the left and soil moisture (%) on the right are plotted as a function of treatments at different times at the Greek site.

Table 2. Average pepper yield per plant measured in different treatments on the
research sites.

Serbian site			Greek site		
	Treatment	Average yield (kg/plant)		Treatment	Average yield (kg/plant)
1.	Control (C)	0.504		Plant with wool mulch (PW)	1.512
2.	Straw mulch (SM)	0.850	1.		
3.	Agro textile (AT)	0.808	2.	Plant without wool mulch (P)	1.087
4.	Wool mulch (WM)	0.504			

Environmental factors influence the activity of microorganisms, and consequently the yield, because where conditions were more optimal for the microbes, the yield was higher. Table 2 shows the average yield per plant per treatment. On Serbian sandy soils, straw helped to increase the organic matter content, which also contributed to increased microbiological activity and yields. Similar to the experience of other authors, straw mulch increased yields of winter wheat (HUANG et al., 2005) and maize (SHEN et al., 2012). In contrast, DÖRING et al., (2005) did not observe a significant yield increase in potatoes when straw mulch was applied. In addition to the intensive irrigation, the wool mulch impeded soil aeration by absorbing

moisture, which had a negative effect on microbial activity and consequently on yield. Under extensively irrigated conditions at the Greek site, however, wool mulch increased yields compared to non-covered control plots. CINCINNATI et al., (2012) reported that eggplants covered with wool mulch achieved higher yields. GARTON et al., (2013) also reported significantly higher yields in organic tomato production.

## Conclusion

In this study, the effects of different mulches on soil biological activity and pepper yield under two different field conditions were investigated. We found that under intensively (daily) irrigated conditions on sandy soil, most mulches had no or negative effects on β-glucosidase activity but significantly positive effencts on active carbon. The significantly lower βglycosidase activity in the case of wool mulch suggests that on intensively irrigated soils, wool mulch becomes saturated with water, which inhibits aeration and creates an inadequate habitat for microorganisms, which decreases their activity. We found that, because of organic matter input, active carbon and biological activity increased to the greatest extent in the case of using straw mulch. This is also reflected in the fact that, the yield using straw mulcs slightly exceeded the yield measured in the agrotextile treatment and was significantly higher compared to the control. However, under extensively irrigated situation on soil with higher water holding capacity, the wool mulch can be advantageous over unmulched areas, as its moisture retention capacity helps microorganisms survive and become active, thereby increasing yield. Mulching methods therefore affect crop yields not only through their water retention capacity, but also through their soil biological activity. The effect of each mulching method on soil biological activity and yields depends on the degradability of the mulch material, the frequency of irrigation, and the soil's water-holding capacity.

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