INVESTIGATION OF SOIL FERTILITY AFFECTED BY BIODIESEL BY-PRODUCT IN MICROCOSM EXPERIMENT

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Abstract: On the grounds of expedience, versatile investigation of industrial by-products has to be considered to protect the environment. This consideration has been revealed the field of biodiesel production. Glycerol, which is a by-product from biodiesel production is available in such a large amount, that it can not be disposed in the traditional utilization ways. This glycerol is contaminated and the cleaning process is rather expensive. The pollutants originated from oil seed proteins, containing carbohydrates, as useful plant nutrients, and also potassium hydroxide is used as catalyser and methanol used for transesterification, which ingredient can be utilisable. There is a noticeable tendency, where manufacturers try to recover methanol during the process in order to reach a higher technological efficiency. Potassium content of potassium hydroxide is a plant nutrient. In addition alkalinity can be important in compensation of soil acidity.
According to our previous investigations on hygroscopic ability of crude glycerol influences the germination of plant seeds. As an easily degradable organic matter stimulates microbial activity in the soil and alters the C/N ratio, which can cause a temporary nitrogen deficiency. The nutrient supplying capacity of soil influenced by glycerol was examined in microcosm experiment. Approximate observation is capable to monitor the complex ecological effect for a small scaled soil-plant-animal system. The soils were treated by glycerol and potassium nitrate. *Lolium perenne* as indicator plant and *eisenia fetida* as indicator animal were applied for this investigation.

Keywords: glycerol, biodiesel, by-product, microcosm experiment, *eisenia fetida*

Introduction

The European biodiesel producers typically use rapeseed oil transesterified by methyl alcohol (Kovács, 2000). As a result of transesterification contaminated glycerol are produced too. Glycerol's contamination level is higher if the initial material is used vegetable oil (Kovács et al., 2012). Decontamination of this by-product is expensive. Without extraction of these surrogates cosmetic and chemical industries cannot enable it (Kovács, 2011). Contamination of glycerol originates from vegetable; it contains typically proteins and carbohydrates. If we add it to the soil, this contamination can improve soil fertility. Soil microorganisms can utilize glycerol as nutrient (Papanikolaou et al., 2008; Temudo et al., 2008). These organisms are able to transform glycerol to valuable materials (Barbirato et al., 1998; Johnson and Taconi, 2007; Yazdani and Gonzalez, 2007).
Carbohydrates and similar forms of organic matters have a significant impact on soil nutrient supplying capacity (Gulyás and Füleky, 1994; Kátai et al., 2005). The effect mainly influenced by the mobility of nitrogen. Glycerol can change the soil C/N ratio. Added organic matter can significantly change C/N ratio of the soil. Increasing the nitrogen content of the soil results a temporary immobilization (Tisdale and Nelson, 1966). Glycerol is an easily accessible and utilizable carbon source to microorganisms (Lee et al., 2001; Tickell, 2003). Glycerol can increase microbial activity and this also contributes to an increased amount of available nutrients in the soil. Mineral nitrogen forms which can be taken up by immobilized plant by microbes only can be bonded temporary because these will be mobilized again. (Tolner et al., 2010). Temporary absorption of nitrogen can inhibit nitrate leaching from the ground soil (Tolner et al.,

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2012). The correct usage of the industrial by-product may contribute to achieve environmentally friendly industrial and agricultural production. (Tóth et al, 2011). An industrial by-product that can affect soil fertility cannot be evaluated only according to their influence on the plants. Complex ecological systems that model the effect of microcosm can be tested through experiments (Parmelee et al., 1993; Uvarov, 1993). In the microcosm systems effects on animal organs are also examinated such as effects on plants. Indicator animals can be collembolas (Bakonyi et al., 2006; Seres et al., 2004), earthworms (Uvarov, 1995), or manure worms (*eisenia fetida*) (Clark and Coats, 2006).

Materials and methods

The microcosm experiments were set in 1000 cm3 wide-mouthed glass jars and filled with 500 g of soil.
As test plants ryegrass, as the test animals manure worms (*Eisenia fetida*) were used for the experiment. A sandy soil from Fót was applied for treatments. The main properties of this soil: saturation percentage, K_A=28.33, lime content, $CaCO_3$%=8%, pH_{H2O}=8.2, humus content, H%=1.4%, AL-P_2O_5=95 ppm, AL-K_2O=120 ppm.
The solution were: 1000 mg.N.dm^{-1} KNO_3 (7.221 g of KNO_3 were dissolved in 1000 cm^3 solution) and glycerol 5% C content (128.55 g 95% glycerol in 1000 cm^3 solution). Four types of treatments were used. (*Table 1.*).

Table 1. The treatments of the soil in microcosm experiment, Gödöllő, 2012

Treatment	Code	N ppm	N-sol. cm^3	C %	G-sol. cm^3	DV cm^3
Control	C	0	0	0	0	100
Glycerol	G	0	0	0,5	40	60
N (nitrogen)	N	100	40	0	0	60
N + Glycerol	NG	100	40	0,5	40	20

Treatments were replicated in three times and were adjusted to 5-5 worms. 14 days after water saturation of the soil in the attempted expulsion of worms in order to change their mass determined. The microcosms were investigated after 14, 21 and 28 days since the earthworms were put in. It was notable that there were no sings of structural damaging in the soils with glycerol content during the experiments. Significant damaging was experienced in microcosms with glycerol and nitrogen, the control microcosms and also only nitrogen affected microcosms.
We used a program for ANOVA which made by Tolner in Microsoft Office Excel (Aydinalp et al., 2010; Sipos et al., 2009).

Results and discussion

The change of worms weight was evaluated by three factor variance analysis. Factors:

- „A" the three sampling day (day 14, day 21, day 28)

- „B" treatments (Control, Glycerol, Nitrogen, Nitrogen+glycerol)

- „C" the starting date and the observation time points measured masses

The effect of the factor „A" (elapsed time to the sampling) to the other factors calculated average weight did not change significantly (F-rate=0.15).
The effects of the factor „B" (treatments) are evaluated on the other factors calculated average on the worms average mass (*Table 2.*).

Table 2. Worm mass (g) depending on the averages of treatments

Treatment	Control	Glycerol	N (nitrogen)	N + Glycerol	LSD (5%)
"A" mean	0.61	0.69	0.52	0.60	0.08

It is apparent the Nitrogen + Glycerol treatment did not show significant change compared to the control sample. The Glycerol treatment significantly increased the weight of the earthworms, while the nitrogen only treatment considerably decreased that. It shows similar picture to the „AxB" interaction analysis (*Figure 1.*)

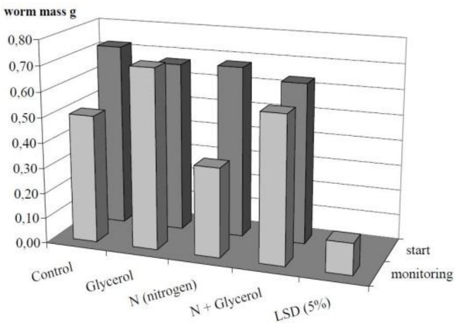

Figure 1. Average mass of the worms at the start time and the average of the monitoring times

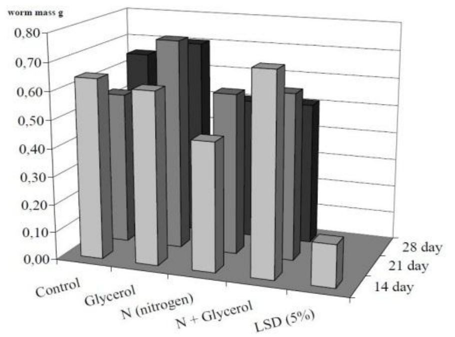

Figure 2. Average mass of the worms depending on the dates of treatments and monitoring

The prime equal average worm's weights are significantly changed due to the treatments. In the case of control and nitrogen only treatment the average of the worms mass are reduced significantly. The average weights of worms are also reduced in the nitrogen only treatment compared with the control sample. Examining the average weight of the worms at different test dates, it is apparent that the nitrogen treatment causes worm mass reductive effect. (*Figure 2.*).

Conclusions

We studied changes in soil fertility with nitrogen and glycerol treatment in complex systems which are includes plants and animals also. Both plant and animal weight changes suggest that the industry by-product as glycerol resulting positive impact on the fertility of soils.

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