

## Microbiological indices of soil quality fertilized with dairy sewage sludge

S. Jezierska-Tys<sup>1</sup> and M. Frac<sup>2\*</sup>

<sup>1</sup>Department of Agricultural Microbiology, University of Life Sciences, Leszczyńskiego 7, 20-069 Lublin, Poland

<sup>2</sup>Institute of Agrophysics, Polish Academy of Sciences, Doświadczalna 4, 20-290 Lublin 27, Poland

Received May 9, 2008; accepted June 17, 2008

**Abstract.** The study was performed on a pot experiment in which a grey-brown podzolic soil was amended with two different doses of dairy sewage sludge (30 and 75 t ha<sup>-1</sup>) – 1 and 2.5% of DSS. During periods of incubation in different terms of analyses (7, 14, 30, 60, 90 and 120 days after the incorporation of dairy sewage sludge) the following analyses were done: so-called total number of bacteria, so-called total number of filamentous fungi, number of cellulolytic bacteria, respiration activity and dehydrogenase activity. Dairy sewage sludge applied to the soil caused stimulation of the total number of bacteria, fungi and cellulolytic bacteria and respiration activity and inhibition of dehydrogenase activity in a grey-brown podzolic soil.

**Keywords:** dairy sewage sludge, microorganisms, respiration activity, soil

### INTRODUCTION

One of the methods of utilization of sewage sludge is its application in agriculture (Fernandez *et al.*, 2007; Siuta, 2002). The most favourable – in terms of fertilization of crop plants – are sludges of food industry sewage, including dairy sewage sludge (Siuta, 2001), as sludge formed in the process of dairy sewage treatment is rich in nutrients for plants and is characterised by high levels of organic matter (Ciećko *et al.*, 2001; Magrel, 2003). Organic and mineral compounds introduced in soil with sewage sludge have a significant effect on microbial populations and their biochemical activity (Sullivan *et al.*, 2005). The objective of this study was to acquire knowledge on the effect of dairy sewage sludge on the total number of bacteria and fungi, cellulolytic bacteria, and on the respiratory and dehydrogenase activity in a grey-brown podzolic soil.

### MATERIAL AND METHODS

For the purposes of the study, a pot experiment was established, with dairy sewage sludge on grey-brown podzolic soil. The soil had the following composition: sand fraction 65%, silt fraction 19%, fine silt and clay fraction 16%. The soil had acid reaction (pH 4.8) and contained 0.4 g kg<sup>-1</sup> N, 4.5 g kg<sup>-1</sup> C, 5.3 g kg<sup>-1</sup> P and 8.7 g kg<sup>-1</sup> K. The dairy sewage sludge was introduced into soil at the following doses: 30 and 75 t ha<sup>-1</sup> (1 and 2.5% of DSS), mixed and watered to 60% of the total water capacity. Each pot contained 4 kg of soil. The experiment was carried out in three replications. The soil samples were incubated at 20°C through 4 months. The soil was collected from pots and analysed after 7 (I), 14 (II), 30 (III), 60 (IV), 90 (V) and 120 (VI) days of incubation. The following parameters were determined in the soil: so-called total number of bacteria on a medium with soil extract and K<sub>2</sub>HPO<sub>4</sub>, so-called total number of filamentous fungi on medium of Martin (1950), number of cellulolytic bacteria on liquid medium with cellulose (Rodina, 1968), respiration activity with the method of Rühling and Tyler (1973), dehydrogenase activity with the method according to Thalmann (1968).

The results obtained were processed statistically with the method analysis of variance. Significance of differences was determined using the Tukey test, at  $p = 0.05$ .

### RESULTS

Figure 1a presents the seasonal so-called total number of bacteria in the grey-brown podzolic soil amended with 30 and 75 t of dairy sewage sludge. Introduction of the sludge to

\*Corresponding author's e-mail: m.frac@poczta.fm

the soil at the dose of 30 t caused insignificant seasonal changes in the total number of bacteria compared to the control treatment, while at the dose of 75 t the sewage sludge caused a significant increase in the population of the studied microbial group, as illustrated in Fig. 1a.

The numbers of fungi in soil with sewage sludge doses applied in the experiment are presented in Fig. 1b. Analysing the seasonal numbers of those micro-organisms presented in Fig. 1b one may note distinct stimulation of the studied microbial group by the sludge dose of 75 t compared to the results obtained in the control treatment and in the treatment with the lower dose of the sludge. This is also supported by the mean values for the particular experimental treatments, given in Fig. 1b.

Figure 1c illustrates the seasonal numbers of cellulolytic bacteria in the particular experimental treatments. Noteworthy is the high number of the studied bacteria on analysis dates II and IV, especially in the treatment with the higher dose of the sludge. During the whole period of the study the numbers of cellulolytic bacteria were stimulated by both sludge doses applied in the experiment. Also notable is the decrease in the numbers of cellulolytic bacteria on analysis dates V and VI. The mean values for the particular experimental treatments, presented in Fig. 1c, confirm the significant stimulation of the population of the microbial group under study by the sludge doses applied.

Mean values of respiratory activity in the experimental treatments for the individual dates of analyses are illustrated in Fig. 2a. Dairy sewage sludge, at doses of both 30 and 75 t, caused an increase in microbiological activity of the soil as measured by the amount of emitted CO<sub>2</sub>, on analysis dates I, II and III, above the level recorded in the control treatment. On analysis dates IV and V, in the treatments with the sludge, lower respiratory activity was recorded compared to the treatment without the sludge. In the final stage of the experiment, in the soil with dairy sewage sludge dose of 30 t, an increase was observed in respiratory activity to a level above that in the control treatment.

Dairy sewage sludge, applied at the doses of 30 and 75 t, caused a notable decrease in the dehydrogenase activity of the grey-brown podzolic soil on the initial three dates of analysis (with exception of higher dose of sludge). On later dates of analysis the activity was on the level of values obtained in the control treatment, but also lower. Seasonal trends in the effect of the sludge affected the mean values of dehydrogenase activity in the individual experimental treatments, as illustrated in Fig. 2b.

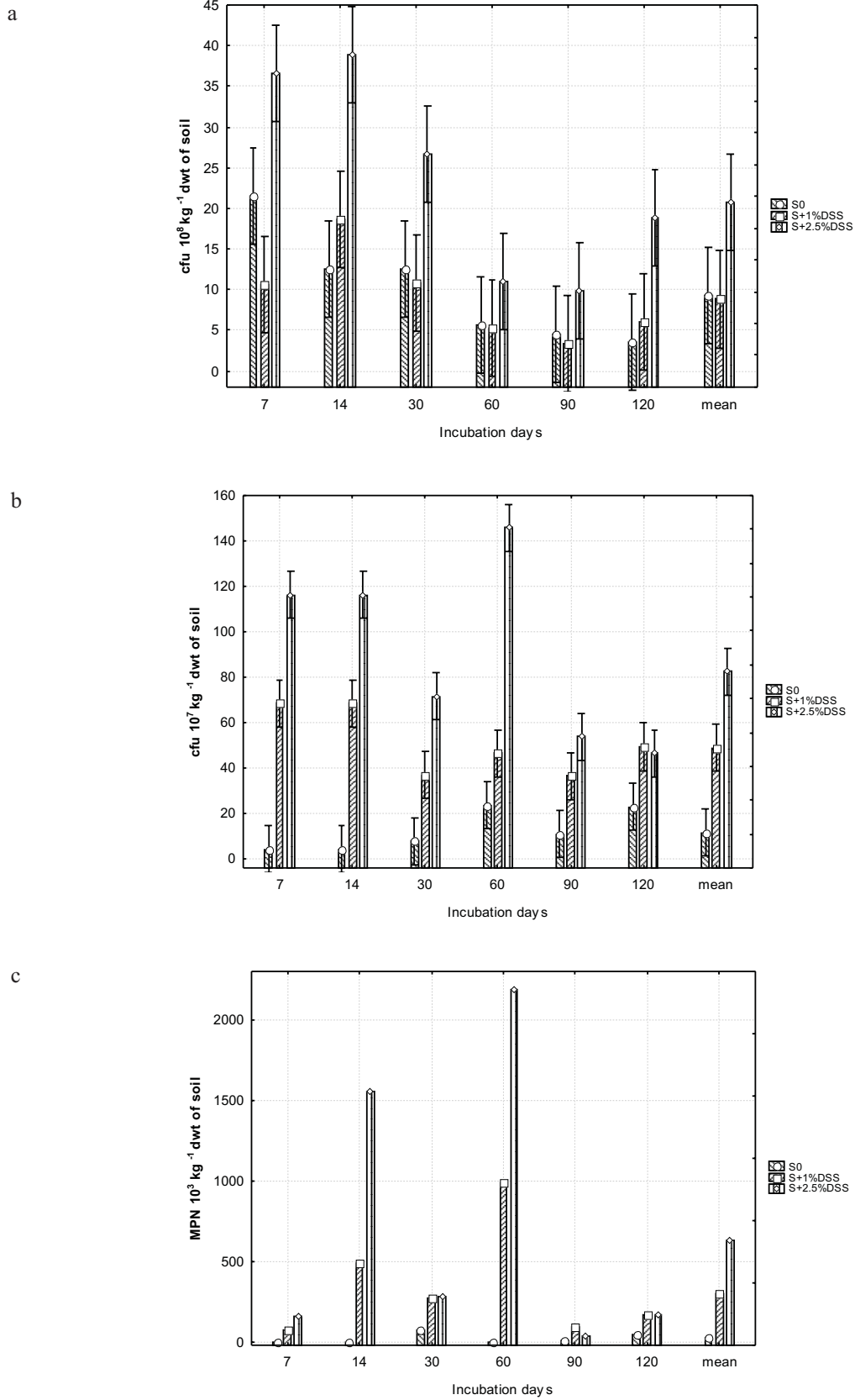
## DISCUSSION

Sewage sludge, and especially sludge originating from dairy sewage treatment plants, due to its high content of organic matter and biogens may be conducive to changes in microbial populations and their biochemical activity (Jeziarska-Tys and Frąc, 2005). In the soil environment

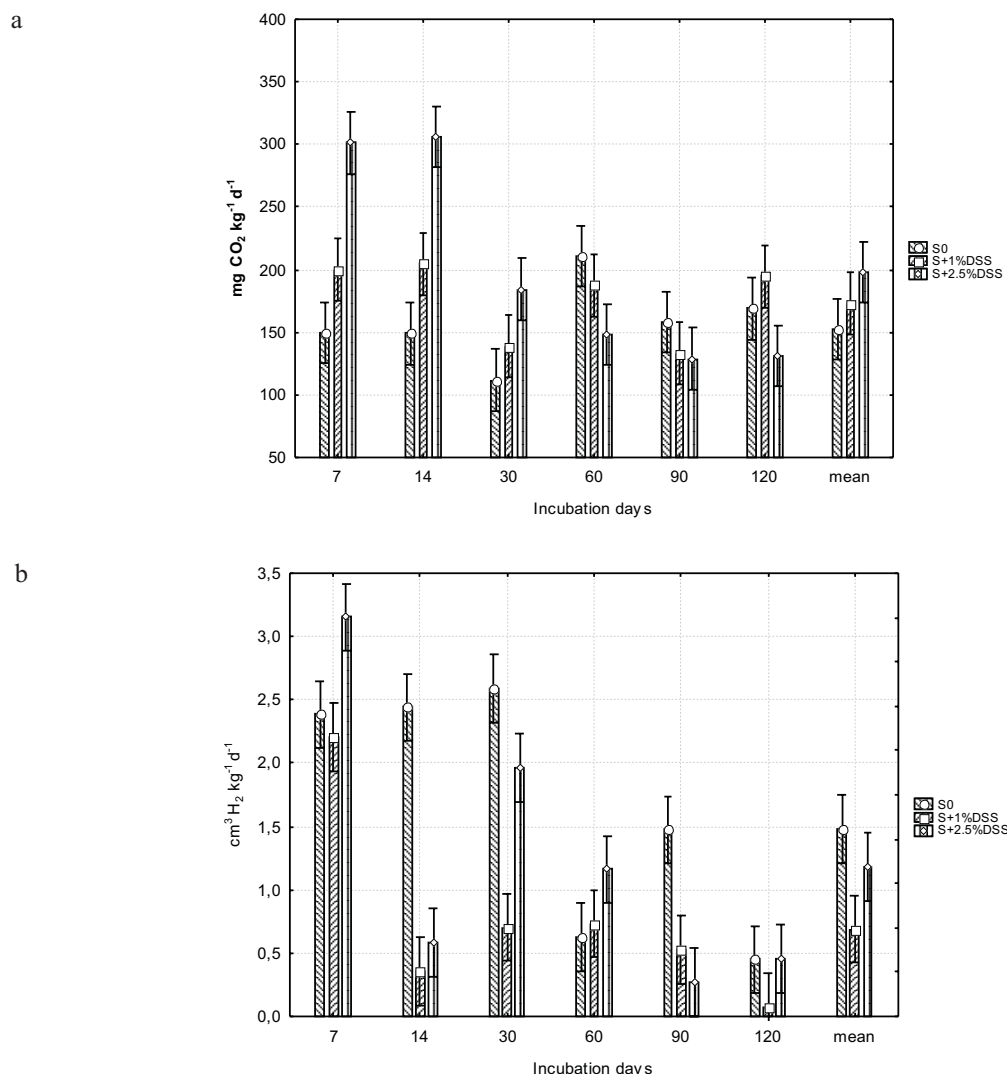
there occur various microbial groups which, with the help of enzymes, catalyse transformations of organic and mineral compounds introduced into the soil. According to some authors (Alkorta *et al.*, 2003; Janvier *et al.*, 2007; Nannipieri *et al.*, 2003), soil is a highly complex biological system which is subject to dynamic changes under the effect of biotic and abiotic factors. The use of microbiological and biochemical properties of soil for the estimation of changes taking place in soil environment as a result of *eg* application of sewage sludge is fully justified. The microbiological indices providing information on changes in the soil environment as a result of application of anthropogenic factors, such as *eg* sewage sludge, include the size of populations of particular microbial groups, including the total numbers of bacteria, fungi, or micro-organisms with specific nutritional requirements *ie* cellulolytic bacteria. A study conducted by these authors showed that dairy sewage sludge applied at the doses of 30 and 75 t had a stimulating effect on the microbial groups under study *ie* on the total number of bacteria, fungi, and bacteria mineralising cellulose. The effect of dairy sewage sludge on the microbial groups under study depended on the applied dose of that organic waste. As fungi and cellulolytic bacteria require greater amounts of organic carbon for their growth (Paul and Clark, 2000), the treatment with the higher dose of the sludge, 75 t ha<sup>-1</sup>, was characterised by their notably greater numbers. A similar effect of sewage sludge on bacterial and fungal populations was observed by Lima *et al.* (1996). Also the present authors, in an earlier study, found a favourable effect of dairy sewage sludge on the total numbers of bacteria, fungi, and cellulolytic bacteria, in laboratory and field experiments alike (Jeziarska-Tys and Frąc, 2005, 2007; Jeziarska-Tys *et al.*, 2005).

Organic matter introduced into the soil with sewage sludge may cause changes not only in microbial populations, but also in their enzymatic activity and rate of respiration. Studies by certain authors (Lai *et al.*, 1999; Saviozzi *et al.*, 2002) indicate that dehydrogenase activity may be used as an index of changes in the biological activity of soil environment under the effect of organic fertilization. The results obtained in our study showed that the applied doses of dairy sewage sludge caused a reduction in dehydrogenase activity of grey-brown podzolic soil. This was probably related with rapid depletion of respiratory substrates by soil microorganisms, as evidenced by the results obtained in the treatment with the higher dose of the sludge. In that experimental treatment dehydrogenase activity was on a significantly higher level compared to the treatment with the lower dose of the sludge. This is also supported by results obtained by the authors mentioned above (Saviozzi *et al.*, 1999).

According to certain authors (Fernandes *et al.*, 2005; Garcia-Gil *et al.*, 2002), measurement of the rate of respiration is also one of the methods for determination of soil microbial activity. The respiratory activity of soil depends, among other things, on the availability of organic matter and



**Fig. 1.** Changes of total number of: a – bacteria, b – fungi, and c – cellulytic bacteria; in grey-brown podzolic soil amended with dairy sewage sludge under aerobic incubation. Vertical bars indicate the standard error of the averages. S0 – control soil, S – soil, DSS – dairy sewage sludge.



**Fig. 2.** Changes of: a – respiration, b – dehydrogenase activities in grey-brown podzolic soil amended with dairy sewage sludge under aerobic incubation. Explanations as on Fig. 1.

on the size of microbial populations. This suggestion is supported by the results of our study. Both doses of the sludge, 30 and 75 t, caused an increase in the populations of the microbial groups under study and in the level of respiratory activity. Notably higher respiratory activity was characteristic of the treatment with the higher dose of dairy sewage sludge.

#### CONCLUSIONS

1. Dairy sewage sludge at doses of 30 and 75 t ha<sup>-1</sup> caused stimulation of the total number of bacteria, fungi and cellulolytic bacteria in a grey-brown podzolic soil.
2. The applied doses of the sludge had an inhibiting effect on dehydrogenase activity in the studied soil.
3. A stimulating effect of dairy sewage sludge on the respiratory activity of the soil was observed.

#### REFERENCES

- Alkorta I., Aizpurua A., Riga P., Albizu I., Amezaga I., and Garbisu C., 2003. Soil enzyme activities as biological indicators of soil health. *Rev. Environ. Health*, 18, 65-73.
- Ciećko Z., Wyszowski M., and Rolka E., 2001. Chemical characteristics of sewage sludge from dairy treatment plants (in Polish). *Zesz. Probl. Post. Nauk Roln.*, 477, 313-318.
- Fernandes S.A.P., Bettiol W., and Cerri C.C., 2005. Effect of sewage sludge on microbial biomass, basal respiration, metabolic quotient and soil enzymatic activity. *Appl. Soil Ecol.*, 30, 65-77.
- Fernandez J.M., Plaza C., Hernandez D., and Polo A., 2007. Carbon mineralization in an arid soil amended with thermally-dried and composted sewage sludges. *Geoderma*, 137, 310-317.
- Garcia-Gil J.C., Plaza C., and Polo A., 2002. Sewage sludge effects on biological and biochemical parameters in a degraded soil. *Waste Manag. Environ.*, 56, 341-350.

- Janvier C., Villeneuve F., Alabouvette C., Edel-Hermann V., Mateille T., and Steinberg C., 2007.** Soil health through soil disease suppression: Which strategy from descriptors to indicators? *Soil Biol. Biochem.*, 39, 1-23.
- Jezierska-Tys S. and Fraç M., 2005.** The effect of fertilization with sewage sludge from a dairy plant and with straw on the population numbers of selected microorganisms and respiration activity of brown soil. *Polish J. Soil Sci.*, 38, 145-151.
- Jezierska-Tys S. and Fraç M., 2007.** The influence of dairy sewage sludge and FYM on the numbers and biochemical activity of microorganisms that participate in the transformations of soil nitrogen. *Polish J. Environ. Stud.*, 16, 686-693.
- Jezierska-Tys S., Fraç M., and Fidecki M., 2005.** Influence of fertilization with sewage sludge from dairy on respiration activity and number of bacteria and fungi in brown soil (in Polish). *Zesz. Probl. Post. Nauk Roln.*, 506, 205-212.
- Lai K.M., Ye D.Y., and Wong J.W.C., 1999.** Enzyme activities in a sandy soil amended with sewage sludge and coal fly ash. *Water Air Soil Poll.*, 113, 261-272.
- Lima J.A., Nahas E., and Gomes A.C., 1996.** Microbial populations and activities in sewage sludge and phosphate fertilizer-amended soil. *Appl. Soil Ecol.*, 4, 75-82.
- Magrel L., 2003.** The possibility of biogas production during digestion of liquid manure and sewage from municipal and dairy wastewater treatment plants (in Polish). 2nd Int. and 13th Nat. Conf. Renewable energy sources. The new insights into sewage sludge. Part II. February 3-5, Częstochowa, Poland, 467-478.
- Martin J., 1950.** Use of acid rose bengal and streptomycin in the plate method for estimating soil fungi. *Soil Sci.*, 19, 215-233.
- Nannipieri P., Ascher J., Ceccherini M.T., Landi L., Pietramellara G., and Renella G., 2003.** Microbial diversity and soil functions. *European J. Soil Sci.*, 54, 655-670.
- Paul E.A. and Clark F.E., 2000.** *Soil Microbiology and Biochemistry* (in Polish). UMCS Press, Lublin, Poland.
- Rodina A., 1968.** *Microbiological Methods of Water Analysis* (in Polish). PWRiL Press, Warsaw, Poland.
- Rühling A. and Tyler G., 1973.** Heavy metal pollution and decomposition of spruce needle litter. *Oikos*, 24, 402-415.
- Saviozzi A., Biasci A., Riffaldi R., and Levi-Minzi R., 1999.** Long-term effects of farmyard manure and sewage sludge on some soil biochemical characteristics. *Biol. Fert. Soils*, 30, 100-106.
- Saviozzi A., Bufalino P., Levi-Minzi R., and Riffaldi R., 2002.** Biochemical activities in a degraded soil restored by two amendments: a laboratory study. *Biol. Fert. Soils*, 35, 96-101.
- Siuta J., 2001.** *Management of wastes in the environment* (in Polish). *Zesz. Probl. Post. Nauk Roln.*, 477, 275-285.
- Siuta J., 2002.** *Utilization of Wastes* (in Polish). Institute of Environment Protection Press, Warsaw, Poland.
- Sullivan T.S., Stromberger M.E., Paschke M.W., and Ippolito J.A., 2005.** Long-term impacts of infrequent biosolids applications on chemical and microbial properties of semi-arid rangeland soil. *Biol. Fert. Soils*, 42, 258-266.
- Thalman A., 1968.** Zur methodik der Bestimmung der Dehydrogenase aktivität in Boden mittels Triphenyltetrazoliumchlorid (TTC). *Landwirtsch. Forsch.*, 21, 249-258.