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# EFFECT OF SYNTHETIC ORGANOMINERAL COMPLEXES ON THE PROCESS OF RESPIRATION OF A HAPLIC LUVISOL

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**Summary.** The study was concerned with the effect of synthetic organomineral complexes SOMC (humic acid with Na-montmorillonite at various pH levels (3–7) and in the presence of $Al_2O_3$), added to samples of a Haplic Luvisol, on soil microbial $CO_2$ evolution. A model incubation experiment was performed, with the division for basal respiration (BR) at 28°C for a period of 7 days and long-term respiration (LTR) at 25°C for a period of 35 days. The measurement of soil microbial activity was the amount of evoluted $CO_2$ and the reduction of $O_2$ concentrations per unit of time (rate of respiration). The pH in $H_2O$ and in 1 M KCl was determined after 7 and 35 days of incubation. A considerable decrease was observed in soil pH (in $H_2O$), both during the BR (5.95–6.60) and the LTR (5.13–5.85). The addition of synthetic organomineral complexes with $Al_2O_3$ of low initial pH (3 and 4) had an inhibiting effect on soil microbial respiration both during the BR and the LTR. A lower soil microbial respiration was observed during the LTR as compared to the BR (on average by a factor of 3 and 2.6, respectively), for $CO_2$ evolution and reduction in $O_2$ consumption. Values of the $CO_2 : O_2$ ratio equal to 1 : 1 or slightly above one indicate the dominance of aerobic processes. Also, a positive correlation was observed between $CO_2$ evolution and $O_2$ consumption.

**Key words:** respiration, humic acids, sodium-montmorillonite, $CO_2$ evolution, $O_2$ consumption, $CO_2/O_2$

## INTRODUCTION

The role of organic-mineral complexes occurring in soils has been the object of numerous studies [Signer and Huang 1993, Stevenson 1994, Jones and Kochian 1996, Księżopolska 2001, 2004, 2005, Qing Huang *et al.* 2008]. According to Stevenson [1994], several mechanisms explain the formation of complexes between organic and mineral components of soils. Chemical reactions in soil mostly occur on the surface of soil organomineral components, for example ion exchange and adsorption, ion diffusion, and the acid-base balancing. The reactions are directly or indirectly affected by surface properties of soil organomineral components. Organic matter can be closely associated with the mineral matter in soil. The processes are closely related with decrease in the soil pH, which increases mobility of many metals in the soil, including aluminium [Sposito 1989, Geoffery and Gadd 2004, Weber and Karczewska 2004, Ronella *et al.* 2004, Gruba 2009]. Aluminium occurs in the largest amounts in the Earth crust (approx. 8%), and therefore its release to the soil solution as a result of low pH has been the subject of numerous studies [Kotowski *et al.* 1994, Kotowski and Pawłowski 1995, Filipek and Dechnik 1995, Hu and Boyer 1996, Badora 2002, Ronella *et al.* 2004].

Changes in the physicochemical properties of soil as a result of acidification influence both the quantity and the quality of soil microorganisms [Kurek 2002, Khan and Scullion 2000]. The soil microbial activity is dependent not only on the soil pH, organic and mineral compounds, and relative content of particular forms of metals in the soil, but also on soil temperature. Microorganisms develop the best at temperatures of 20–30°C [Kobus 1995]. Respiration is one of the fundamental living processes of soil organisms, and especially microorganisms, and depends on a number of factors such as: organic substrate, moisture, soil temperature and density, pH, time of incubation, presence of heavy metals, pesticides, and many others [Gliński and Stępniewski 1985, Khan and Scullion 2000, Swantson *et al.* 2002, Kartina *et al.* 2003, Weber and Karczewska 2004, Geoffery and Gadd 2004, Renell *et al.* 2004].

The soil microbial activity can be estimated by the amount of evolved $CO_2$ or of $O_2$ consumption in a sample incubated for specific periods at specific temperatures [Klimanek *et al.* 1979, Edwards 1982, Klimanek 1994, Mercik *et al.* 1999, Cerhanova *et al.* 2006, Freschet *et al.* 2008].

Soil microorganisms effuse the organic acids and „siderophores" which complex and mobilise some kind of metals from insoluble sources, for example: Fe from $Fe_2O_3$ and Al from $Al_2O_3$, Mn from $Mn_2O_3$ [Kurek and Jaroszuk 1993, Hu X and Boyer 1996, Kurek 2002].

The objective of the study was to determine the effect of synthetic organomineral complexes, i.e. humic acid with Na-montmorillonite of differentiated pH (3–7) and in the presence of $Al_2O_3$ on soil microbial activity estimated by $CO_2$ evolution and $O_2$ consumption in a Haplic Luvisol.

## MATERIALS AND METHODS

### Synthetic organomineral complexes

In order to prepare the synthetic organomineral complexes (SOMC), the following materials were used: Na-montmorillonite-Wyoming-S-Wy-2 (Na-M) (from the Department of Geology, University of Missouri, Columbia, USA), humic acid extracted from a chernozem soil using the Schnitzer and Stevenson method [Schnitzer and Schluppli 1989, Stevenson 1994] and aluminium oxide ($Al_2O_3$) purchased from the Polish Chemical Reagents-POCH.

The isolated humic acids were complexed with Na-montmorillonite. For this, the mineral (4 g) was suspended in 0.1 mol $L^{-1}$ NaOH at a ratio of 1 : 10 (w/v) and amended with humic acids (HA) (10%) and 10% of Al (in the form of $Al_2O_3$). The suspensions were adjusted to pH (3, 4, 5, 6, (±0.2) and 7 (±0.5). The pH was adjusted with 0.1 M KOH and 0.1 M $HNO_3$ and matured for 24 h with occasional shaking [Księżopolska 2001, 2004, 2005]. The resulting organomineral complexes were filtrated out, dried in a vacuum oven at 20°C over concentrated $H_2SO_4$ and stored at room temperature until analysed.

**Soil samples**

For the investigations soil samples were taken from lessive soil (Haplic Luvisol) of a sandy texture (46% sand, 31% silt, 23% clay), pH in $H_2O$ 5.19, in 1 M KCl 4.46, with low content of organic carbon 5.7 g $kg^{-1}$, available K 113.0 mg $kg^{-1}$ and available P 49.9 mg $kg^{-1}$.

**Model experiment**

Synthetic organomineral complexes (SOMC) (0.25 g) were added to soil samples (2.5 g) from the arable (0–20 cm) horizon Ap. This portion was placed in 20 glass bottles, divided into 2 series of 10 bottles each, watered to 20% of moisture content and covered with rubber stoppers.

The model study involved the use of SOMC according to the following system: soil + SOMC (pH 3); soil + SOMC + $Al_2O_3$ (pH 3); soil + SOMC (pH 4); soil + SOMC + $Al_2O_3$ (pH 4); soil + SOMC (pH 5); soil + SOMC + $Al_2O_3$ (pH 5); soil + SOMC (pH 6); soil + SOMC + $Al_2O_3$ (pH 6); soil + SOMC (pH 7); soil + SOMC + $Al_2O_3$ (pH 7).

Then the samples were incubated in two steps, and gases evolved from the soils were collected with the use of a syringe [Apfelthaler 1994, Klimanek 1994, Mercik *et al.* 1999, Cerhanova *et al.* 2006, Freschet *et al.* 2008].

The 1$^{st}$ step at 28°C for 7 successive days. The value of respiration after 7 days is named by Apfelthaler [1994] basal respiration (BR). Gases were collected each day at fixed hour.

The 2$^{rd}$ step at 25°C for 5 weeks with respiration measurements at 1 week intervals, named according to Klimanek [1994] long term respiration (LTR). Gases were collected at the end of each week.

The above mentioned 2 types of respiration, BR and LTR, were analysed and temperatures of 25°C and 28°C were taken as optimal for bacteria development [Kobus 1995].

**Measurements**

After each day in the case of the BR measurements, and after every week in the case of the LTR, the amount of $CO_2$ evolution and $O_2$ consumption were determined using a Shimadzu GC-14A gas chromatography apparatus. The Shimadzu GC-14A was equipped with thermal conductivity detector (TCD) – temperature 60°C, column filled with Porapak (Q) – temperature 40°C, and gas flow velocity was 60 ml min [Włodarczyk 2000].

After 7 and 35 days the pH was determined in $H_2O$ and in 1 M KCl (pH Meter 761 Calimatic, Kinck, Berlin, Germany).

The results were statistically elaborated, including analysis of variation, using the Statgraff software (95% LSD), and analysis of regression using the Excel software. The calculated relations were then described with linear, power, logarithm and exponential equations. The description of the relationship analysis was made using the best fitted function.

## RESULTS AND DISCUSSION

**pH of soil samples with the synthetic organomineral complexes**

Soil reaction is one of important factors determining the proper course of many soil physical, chemical and biological processes. Table 1 shows that after 7 days of incubation at 28°C (BR) there was a change in the soil reaction to acid,

Table 1. The pH value (in $H_2O$ and in 1 M KCl) of soil samples after 7 and 35 days of incubation of a Haplic Luvisol

| Variant of experiment | pH after 7 days (BR) | | pH after 35 days (LTR) | |
|---|---|---|---|---|
| | $H_2O$ | 1 M KCl | $H_2O$ | 1 M KCl |
| Soil + SOMC (pH 3) | 5.95 | 5.50 | 5.13 | 4.14 |
| Soil + SOMC + $Al_2O_3$ (pH 3) | 6.33 | 5.55 | 5.69 | 4.75 |
| Soil + SOMC (pH 4) | 6.14 | 5.86 | 5.30 | 4.25 |
| Soil + SOMC + $Al_2O_3$ (pH 4) | 6.50 | 5.85 | 5.51 | 4.55 |
| Soil + SOMC (pH 5) | 5.98 | 5.53 | 5.35 | 4.28 |
| Soil + SOMC + $Al_2O_3$ (pH 5) | 6.48 | 5.51 | 5.77 | 5.03 |
| Soil + SOMC (pH 6) | 6.55 | 5.54 | 5.85 | 5.38 |
| Soil + SOMC + $Al_2O_3$ (pH 6) | 6.60 | 5.55 | 5.77 | 5.05 |
| Soil + SOMC (pH 7) | 6.57 | 5.56 | 5.85 | 5.40 |
| Soil + SOMC + $Al_2O_3$ (pH 7) | 6.30 | 5.50 | 5.84 | 5.42 |

SOMC – Synthetic organomineral complexes (Humic acids + Na-montmorillonite +/- $Al_2O_3$)

as a result of the activity of microorganisms in the soil (pH in $H_2O$ from 5.95 to 6.60, and pH in 1 M KCl from 5.50 to 5.86). On the other hand, after 35 days of incubation at 25°C [LTR], a change of soil reaction was observed, towards more acid (pH in $H_2O$ from 5.13 to 5.85, and in 1 M KCl from 4.14 to 5.42). These observations are in agreement with the opinions of numerous authors, e.g. Signer and Huang [1993], Kotowski *et al.* [1994], Filipek and Dechnik [1995], Badora [2002], Geoffery and Gadd [2004] that in soils with pH in KCl > 5.5 aluminium is bound in the crystalline lattices of primary and secondary minerals, in salts, in insoluble hydroxides, while stable complexes of aluminium with organic acids are formed at soil pH values below 6.5. Mobile aluminium appears only in soils with pH below 5.5. In such soils acid hydrolysis of aluminosilicates takes place, and silica and aluminium hydroxides appear. With persisting low pH, aluminium hydroxide dissociates successive hydroxyl groups and appears in the soil solution as so-called mobile aluminium [Jones and Kochian 1996, Khan and Scullion 2000, Geoffery and Gadd 2004].

**Basal respiration and long-term respiration**

Table 2 presents the cumulative $CO_2$-C and $O_2$ consumption during 7 days (BR) and 35 days (LTR) of incubation. On the final day of incubation, the content of $CO_2$-C varied from 63.1 to 123.7 mg $kg^{-1}$ of soil in the case of the BR, and from 126.1 to 207.2 mg $CO_2$-C $kg^{-1}$ of soil in the case of the LTR. In the case of both the BR and LTR a decrease was observed in the evoluted $CO_2$ from soil samples with the SOMC plus $Al_2O_3$ at initial pH of 3 and 4. Formulation of conclusions using 95% intervals of credibility (LSD) confirmed the significance of the decrease, with the exception of the LTR at pH 4 (Tab. 2). The soil pH seems

Table 2. The cumulative $CO_2$ evolution and $O_2$ consumption during 7 days (BR) and 35 days (LTR) of incubation of a Haplic Luvisol

| Variant of experiment | Basic respiration (BR) | | Long-term respiration (LTR) | |
|---|---|---|---|---|
| | $cm^3$ C-$CO_2$ $kg^{-1}$ soil | $O_2$ % | $cm^3$ C-$CO_2$$kg^{-1}$ soil | $O_2$ % |
| Soil + SOMC (pH 3) | 112.0 | 16.7 | 200.1 | 12.4 |
| Soil + SOMC + $Al_2O_3$ (pH 3) | 79.2 | 18.1 | 126.1 | 16.0 |
| Soil + SOMC (pH 4) | 117.4 | 16.7 | 184.7 | 13.4 |
| Soil + SOMC + $Al_2O_3$ (pH 4) | 102.4 | 17.6 | 175.9 | 13.8 |
| Soil + SOMC (pH 5) | 63.1 | 18.7 | 134.9 | 15.7 |
| Soil + SOMC + $Al_2O_3$ (pH 5) | 123.7 | 16.7 | 188.5 | 12.8 |
| Soil + SOMC (pH 6) | 112.0 | 17.0 | 203.5 | 12.3 |
| Soil + SOMC + $Al_2O_3$ (pH 6) | 121.9 | 16.7 | 179.6 | 14.5 |
| Soil + SOMC (pH 7) | 110.0 | 17.7 | 207.2 | 15.2 |
| Soil + SOMC + $Al_2O_3$ (pH 7) | 117.1 | 16.8 | 184.3 | 14.2 |

SOMC – Synthetic organomineral complexes (Humic acids + Na-montmorillonite +/- $Al_2O_3$)

to preclude the possibility of the presence of free aluminium and its negative effect on the respiratory processes on soil microorganisms. However, there is a clear inhibiting effect of the SOMC of pH 3 with the addition of $Al_2O_3$ as compared to the SOMC without $Al_2O_3$ (Tab. 2). On the other hand, the lower values of the LTR, where the soil pH decreases below 5 (Tab. 1), could have been related to the presence of free Al.

## Statistical analysis

### Basal respiration (BR)

During the process of basal respiration (BR), in soil + SOMC with an addition of Al, statistically significant increase was found in the emission of $CO_2$ within the range from pH 3 to 5 (Fig. 1). No statistically significant differences were observed between the samples with an addition of Al and without it (Fig. 2).

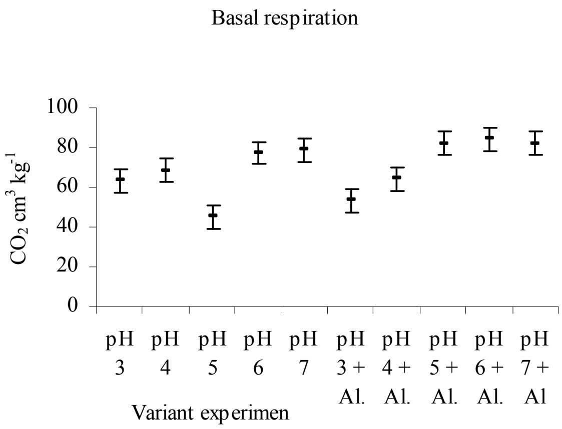


Fig. 1. Evolution of $CO_2$ from samples of Haplic Luvisol during the basal respiration (BR) (all variants pH with and without $Al_2O_3$)

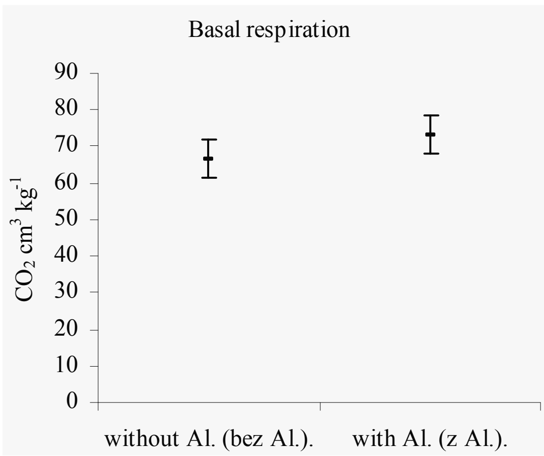


Fig. 2. Evolution of $CO_2$ from samples of Haplic Luvisol during the basal respiration (BR) (variant without $Al_2O_3$, variant with $Al_2O_3$)

## Long term respiration (LTR)

Analysing $CO_2$ emission during for all the variants of the experiment, i.e. the soil + SOMC with Al and without, it was found that the sample with Al at pH 3 had an inhibiting effect on $CO_2$ emission. Significant differences were found between samples of pH 5 and 7 with Al and without it (Fig. 3). Generally, the rate of respiration was significantly higher for the variant without Al (Fig. 4).

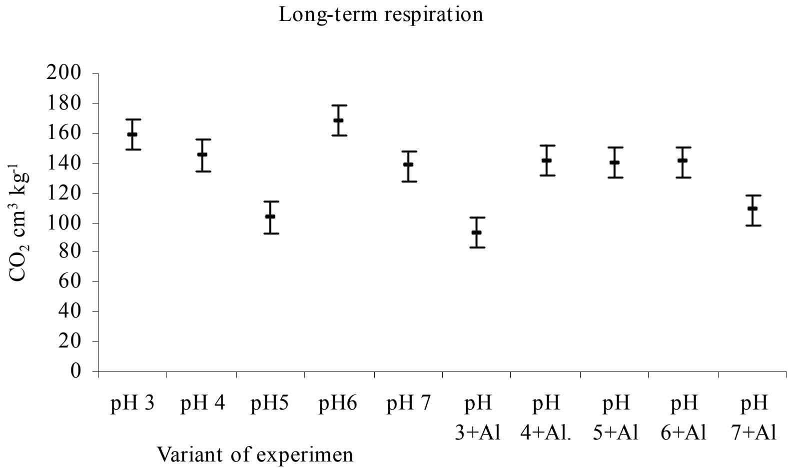


Fig. 3. Evolution of $CO_2$ from samples of Haplic Luvisol during the long-term respiration (LTR) (all variants pH with $Al_2O_3$ and without $Al_2O_3$)

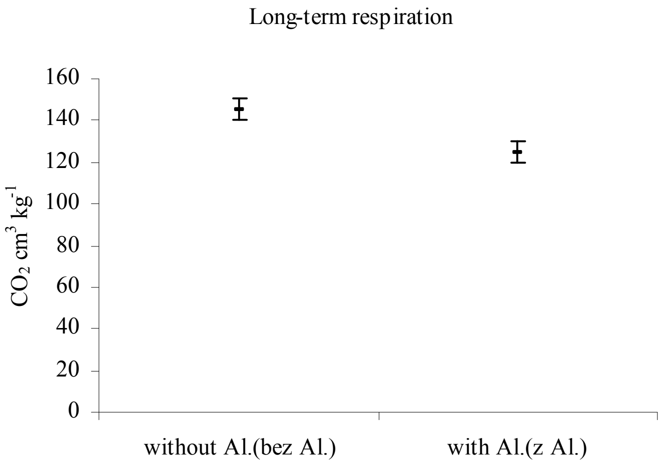


Fig. 4. Evolution of $CO_2$ from samples of Haplic Luvisol during the long-term respiration (LTR) (variant without $Al_2O_3$, variant with $Al_2O_3$)

## The $CO_2 : O_2$ ratio

A good test of the activity of microorganisms is the measurement of the amount of evoluted $CO_2$ and of $O_2$ consumption per unit of time (rate of respiration).

Table 3 presents the daily evolution of $CO_2$ and $O_2$ consumption as expressed in $cm^3$ of gas per kg of soil $d^{-1}$, the $CO_2 : O_2$ ratio and, comparatively, the rate of respiration for the LR for the initial 7 days of incubation. Distinctly lower

Table 3. The daily evolution of $CO_2$ and $O_2$ consumption during the basal respiration (BR) and long-term respiration (LTR) of the Haplic Luvisol

| Variant of experiment | $CO_2$ evolution $cm^3\ kg^{-1}\ d^{-1}$ | | | $O_2$ consumption $cm^3\ kg^{-1}\ d^{-1}$ | | | Values of the $CO_2 : O_2$ ratio | |
|---|---|---|---|---|---|---|---|---|
| | $BR_{7d*}$ | $LTR_{35d}$ | $LTR_{7d}$ | $BR_{7d}$ | $LTR_{35d}$ | $LTR_{7d}$ | BR (28°C) | LTR (25°C) |
| Soil + SOMC (pH 3) | 71.2 | 25.2 | 61.4 | 61.4 | 24.8 | 52.1 | 1.2 | 1.0 |
| Soil + SOMC + $Al_2O_3$ (pH 3) | 49.1 | 15.6 | 31.3 | 41.4 | 14.3 | 27.7 | 1.2 | 1.1 |
| Soil + SOMC (pH 4) | 72.5 | 22.8 | 53.7 | 62.5 | 21.9 | 45.6 | 1.2 | 1.0 |
| Soil + SOMC + $Al_2O_3$ (pH 4) | 64.8 | 22.1 | 53.9 | 49.1 | 20.6 | 40.3 | 1.3 | 1.1 |
| Soil + SOMC (pH 5) | 38.3 | 16.7 | 36.4 | 33.6 | 15.4 | 30.1 | 1.1 | 1.1 |
| Soil + SOMC + $Al_2O_3$ (pH 5) | 76.0 | 23.2 | 51.5 | 62.2 | 23.7 | 44.8 | 1.2 | 1.0 |
| Soil + SOMC (pH 6) | 68.8 | 25.2 | 66.6 | 56.8 | 25.1 | 55.4 | 1.2 | 1.0 |
| Soil + SOMC + $Al_2O_3$ (pH 6) | 75.5 | 22.2 | 56.3 | 62.5 | 18.9 | 46.2 | 1.2 | 1.2 |
| Soil + SOMC (pH 7) | 65.4 | 21.0 | 51.0 | 47.6 | 16.6 | 35.0 | 1.4 | 1.3 |
| Soil + SOMC + $Al_2O_3$ (pH 7) | 72.5 | 22.8 | 59.4 | 60.9 | 19.4 | 46.9 | 1.2 | 1.2 |
| Average | 65.4 | 21.7 | 52.2 | 53.8 | 20.1 | 42.4 | 1.2 | 1.1 |

* $_{7d,\ 35d}$ – days of incubation
SOMC – Synthetic organomineral complexes (Humic acids + Na-montmorillonite +/- $Al_2O_3$)

rate of respiration was observed in the samples incubated over LTR as compared to the BR, which is most likely related to the lower temperature of incubation and the longer period of incubation, during which exhaustion of readily available organic matter could have taken place, which probably entailed a decrease in the microbial activity. The average rate of $CO_2$ emission was 65.4 and 21.7 $cm^3$ $CO_2$-C $kg^{-1}$ $d^{-1}$, respectively, for the BR and the LTR, while the rate of oxygen consumption was 53.8 and 20.5 $cm^3$ $O_2$ $kg^{-1}$ $d^{-1}$, respectively for the BR and LTR. Also, a lower rate of respiration was observed after 7 days of incubation for the LTR (52.2 $cm^3$ $CO_2$-C $kg^{-1}$ $d^{-1}$ and 42.4 $cm^3$ $O_2$ $kg^{-1}$ $d^{-1}$) as compared to the BR, where the decrease in activity could have resulted primarily from the difference in the temperature of incubation.

The presence of humic acids in the SOMC probably had a stimulating effect on the development of fungi, especially at low pH values of the soil samples (Tab. 1). According to Kim *et al.* [1997], fungi constitute a dominant group among microorganisms decomposing humus substances in terms of the number of species. The organisms decompose humic substances most effectively at pH values within the range from 3 to 5. A considerable role in the decomposition of humic substances is ascribed to ligninolytic enzymes produced by numerous fungi of the families *Ascomycetes* and *Basidiomycetes*.

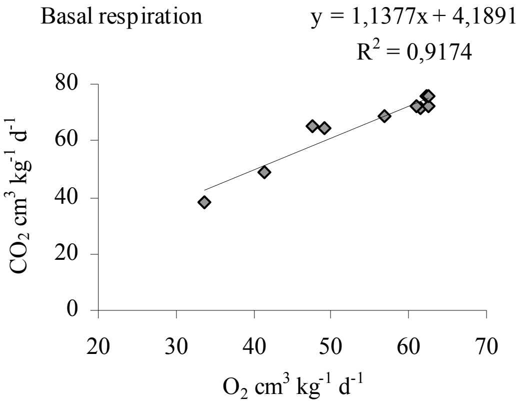


Fig. 5. The relationship between the rate of $CO_2$ evolution and the rate of $O_2$ consumption from samples of Haplic Luvisol during incubation at 28°C (BR), for all variants in final phase of experiment

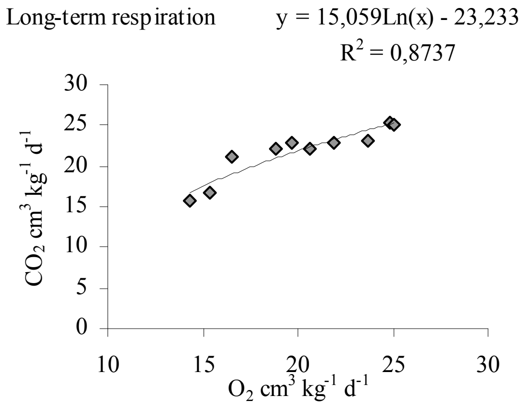


Fig. 6. The relationship between the rate of $CO_2$ evolution and the rate of $O_2$ consumption from samples of Haplic Luvisol during incubation at 25°C (LTR), for all variants in final phase of experiment

The intensity of oxygen consumption and evolution of $CO_2$ in the process of respiration is reflected by the value of the $CO_2 : O_2$ ratio, calculated for the final phase of incubation. Under the conditions of the experiment, the value of the ratio equals 1 or is slightly higher. A somewhat higher value of the $CO_2 : O_2$ ratio was observed in the case of the BR, where the pH value of the soil was higher, which may suggest more intensive bacterial than fungal growth. A value of the $CO_2 : O_2$ ratio higher than 1 suggests that the bacteria, in the process of aerobic respiration, used electron acceptors other than oxygen. Values of the $CO_2 : O_2$ ratio closer to 1 during the LTR indicate the domination of oxygen processes in the environment studied, which would indicate a more intensive growth of fungi, classified among prominent aerobes. According to Bauhus and Bartel [1995], in acid soils a shift is observed in the fungus-to-bacteria biomass ratio in favour of the fungus component. The value of the $CO_2 : O_2$ ratio obtained by these authors is supported by laboratory studies by Kobus [1995] which indicate that the ratio of evoluted $CO_2$ to consumed $O_2$ equals 1 : 1.

The significant relation between the rate of $CO_2$ evolution and the rate of $O_2$ consumption due to soil microbial respiration under the conditions of the experiment, both in the BR and the LTR, is confirmed by regression in analysis of the parameters from the last day of incubation (Fig. 5, 6).

## CONCLUSIONS

1. The addition of $Al_2O_3$ with low initial pH (3 and 4) had a significant negative effect on the respiratory activity, both during the basal respiration (BR) at 28ºC and the long-term respiration (LTR) at 25ºC.

2. The soil + SOMC were characterised by much lower respiratory activity in the case of LTR at 25ºC as compared to the BR at 28ºC, on average by a factor of 3 and 2.6, respectively, for the evolution of $CO_2$ and the consumption of $O_2$.

3. Values of the $CO_2 : O_2$ ratio in the soil + SOMC equal to 1 : 1 or slightly higher than 1 indicate the dominance of aerobic processes occurring in the soil.

4. A positive correlation was found between the activity of $CO_2$ evolution and $O_2$ consumption during incubation at 28 and 25°C.

5. Statistical analysis (95% LSD) showed that in the process of BR an addition of $Al_2O_3$ to soil + SOMC caused an increase in $CO_2$ emission and in oxygen consumption, while in the LTR such a relation occurred in the samples without the addition of $Al_2O_3$

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## WPŁYW SYNTETYCZNYCH KOMPLEKSÓW ORGANICZNO-MINERALNYCH NA PROCES RESPIRACJI GLEBY PŁOWEJ HAPLIC LUVISOL

**Streszczenie**. W niniejszej pracy badano wpływ syntetycznych kompleksów organiczno-mineralnych (kwas huminowy z Na-montmorylonitem ± $Al_2O_3$ przy pH 3–7), dodanych do próbek gleby płowej Haplic Luvisol na jej aktywność respiracyjną w warunkach doświadczenia modelowo-inkubacyjnego. Inkubacje prowadzono w temperaturze 28°C przez 7 dni jako oddychanie podstawowe oraz w temperaturze 25°C przez 35 dni jako oddychanie długotrwałe. Badano intensywność wydzielonego $CO_2$ i pobranego $O_2$ metodą chromatografii gazowej, kontrolowano wartość pH po 7 oraz 35 dniach inkubacji oraz wyliczono wartość stosunku $CO_2/O_2$. W badaniach stwierdzono, że dodatek syntetycznych kompleksów organiczno-mineralnych z udziałem $Al_2O_3$ i przy pH 3 i 4 spo-

wodował spadek aktywności respiracji badanej gleby, zarówno podczas oddychania podstawowego, jak i długotrwałego, co potwierdziła analiza statystyczna (95% LSD). Niższą aktywność respiracyjną zaobserwowano podczas oddychania długoterminowego, w porównaniu z oddychaniem podstawowym, tj. średnio 3 do 2,6 razy odpowiednio dla ilości wydzielonego $CO_2$ i skonsumowanego $O_2$. Stwierdzono również istotną zależność pomiędzy ilością wydzielonego $CO_2$ a ilością pobranego $O_2$ zarówno podczas oddychania podstawowego, jak i długotrwałego, co potwierdziła analiza regresji wielokrotnej dla zmiennej zależnej i niezależnej. Dla zależności tych wyznaczono współczynniki korelacji R pojedynczych wyników.

**Słowa kluczowe**: respiracja, kwasy huminowe, Na-montmorylonit, produkcja $CO_2$, konsumpcja $O_2$,wartość stosunku $CO_2/O_2$