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**BIOLOGICAL ACTIVITY OF MAIN TYPES
OF POLISH SOILS**

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1. INTRODUCTION

Biological activity is a characteristic feature of soils which sets them apart from mother rocks. This activity is connected mainly with the upper (humic) layer of soil and has close connection with soil aeration which is related to all the processes of production, consumption, and transfer of gases within the soil-plant-atmosphere continuum. This term also denotes the gas exchange between soil and the atmosphere, its availability for microorganisms and plant roots, as well as oxidation-reduction processes and enzyme activity (Gliński and Stepniewski 1985).

The knowledge of biological activity of soils is closely connected with evaluation of environment condition towards its hazard to human health and life. So far no comprehensive (wider, sufficient) biological characteristics of Polish soils can be found in our literature.

The aim of this paper is to present the ability of chosen Polish soils concerning N₂O production, consumption and release (emission) to the atmosphere, soil respiration, dehydrogenase activity and CH₄ oxidation. This ability is based on data from analyses of soil samples collected in the Database of Polish arable mineral soils (Bieganowski *et al.* 2013) published in various papers, mainly by Włodarczyk and Brzezińska *et al.* in the years 2000-2012.

2. INDICES OF BIOLOGICAL ACTIVITY OF SOILS

Numerous studies have shown that soil biological activity is closely related to soil fertility and quality (Burns 1978, Gliński and Stepniewski 1985; Włodarczyk *et al.* 2002a, Koper and Piotrowska 2003, Koper *et al.* 2003, Gajda and Martyniuk 2005, Bielińska *et al.* 2008, Brzezińska *et al.* 2011b, Brzezińska *et al.* 2012, Lemanowicz *et al.* 2013). Biological activity of soils may be expressed by N₂O production, consumption and release to the atmosphere, soil respiration, dehydrogenase enzyme activity, and CH₄ oxidation.

Soils are important sources of a number of greenhouse gases such as water vapour, CO₂, CH₄ and N₂O. In general, most N₂O is formed from denitrification in an oxygen deficient environment, although it can also be produced from chemolithotrophic nitrification in aerobic conditions (Włodarczyk *et al.* 2011). Nitrous oxide is a greenhouse gas that is ca. 300 times more effective at radiative forcing than CO₂ on a mole basis. Agricultural soils are the most significant anthropogenic sources of nitrous oxide. Increasing N-inputs into agricultural soils are suspected to be responsible for increasing N₂O emission into the atmosphere (Szarlip *et al.* 2010).

Nitrous oxide production and consumption is affected by many physical and biochemical factors, such as NO_3^- content, redox potential, organic matter availability, soil texture, soil pH, and soil moisture content. These factors interact in a complicated manner with microorganisms in the soil, creating a large spatial and temporal variability in denitrification released and consumption. Soil can remove atmospheric N_2O under conditions favourable for N_2O reduction (Szarlip *et al.* 2010, Włodarczyk 2000, Włodarczyk *et al.* 2005). Understanding of the processes related to nitrous oxide formation and uptake may be useful in predicting of N-fertiliser fate in soil (Szarlip *et al.* 2010).

The significance of the respiration of soil microorganisms consists in their vital role in the decomposition and transformation of soil organic matter. Several biochemical processes are responsible for carbon dioxide (CO_2) production, such as aerobic heterotrophic respiration, denitrification, fermentation, methane oxidation or methane production. When aerobic microorganisms prevail, the production of CO_2 in soil is closely related to oxygen uptake. Microbial respiration is influenced by several factors, both biotic and abiotic (Brzezińska *et al.* 2011a, Burns 1978, Włodarczyk *et al.* 2003) and the amount of CO_2 released from soil depends on soil texture, pH, and the content of native organic matter. However, temperature, moisture, as well as easily available substrate supply (e.g. sugars, proteins, lipids, root exudates etc.) are equally important factors, and the current soil conditions strongly affect CO_2 production. In fact, soil respiration fluctuates, and changes in time and space, depending on the soil air-water conditions (Brzezińska *et al.* 2011b, Brzezińska *et al.* 2006, Oyonarte *et al.* 2012, Walkiewicz *et al.* 2012).

The aerobic microbial activity in soil reaches its maximum at about 60% of soil water holding capacity (WHC). If water content increases, then diffusion of substrates and O_2 becomes less and more limiting, respectively (Skopp *et al.* 1990). Water saturation has dramatic consequences for gas diffusion processes in soil, as gases diffuse 10,000 faster in air than in water (Gliński and Stepniewski 1985). Consequently, one of the main effects of flooding is a lower pool of available O_2 (Stepniewski *et al.* 2005) and a several-fold change in the soil dehydrogenase activity (DHA), being the important enzymes of microbial respiration metabolism (Brzezińska *et al.* 1998). Soil air-water conditions determine the availability of O_2 and other terminal electron acceptors, and affect the population and activity of soil microorganisms (Gliński and Stepniewski 1985, Włodarczyk *et al.* 2003). Dehydrogenase enzymes are considered to play an essential role in the initial stages of the oxidation of soil organic matter. They transfer hydrogen and electrons from oxidised C-substrates to acceptors. Many different intracellular enzymes or enzyme systems contribute to the total soil dehydrogenase activity

(Burns 1978). Among the various soil indicators, DHA is one of the most adequate, important and sensitive bioindicators relating to soil quality and fertility (Brzezińska *et al.* 1998, Włodarczyk *et al.* 2002a, Wolińska and Stepniewska 2011). It is used as a measure of any soil disruption posed by pesticides, heavy metals and other soil contaminants, and improper management practices.

Recent literature reviews on greenhouse gas emissions and terrestrial C and N cycles have highlighted that soil microbial populations play a central role in regulating the major greenhouse emissions (Owens and Xu 2011). Methane (CH₄) is present in the atmosphere at the average concentration of 1.78 ppm, and its amount doubled during the past 200 years, which is especially problematic given its global warming potential being up to 40 times that of CO₂ (Dlugogencky *et al.* 1994, Mancinelli 1995, Shindell *et al.* 2009). Its main sources are wetlands, rice field soils, and landfills, but a part of its production in anaerobic zones is oxidised in the aerobic parts (Le Mer and Roger 2001). Methane oxidising bacteria (methanotrophs) play a significant role for CH₄ sink in soils. The key enzyme in methanotrophy is methane monooxygenase (MMO) which converts methane to methanol. Determination of the kinetics of methane oxidation in soils is described in detail by Saari *et al.* (2004), Baani and Liesack, 2008; Walkiewicz *et al.* (2012).

3. EXPERIMENTAL CONDITIONS

The soils investigated comprised several taxonomic units, mainly: Eutric Cambisols, Haplic Luvisols, Haplic Phaeozems but also Eutric Histosols, Calcaric Regosols, Mollic Gleysols and Haplic Podzols formed from various textural classes: sands, silts, clays and loams. The results concerning the biological activity of soils were also related to such soil properties as soil texture, Corg, pH, Eh, ODR and reduced iron (Fe II) content. Eutric Cambisols, Haplic Luvisols and Haplic Phaeozems occupy about 90% of mineral arable soils in Poland (Gliński *et al.* 1991). On the surface of land they form a mosaic of various soil taxonomic units. Soils defined in some papers as Calcaric Regosols should be included to Eutric Cambisols.

The samples from the upper (0-30 cm) horizons, representing tested soils, were examined in model (laboratory) experiments with standardized conditions (soil aeration status and temperature), which allowed to express rather the potential of soils to perform the given processes than their current conditions.

The data derive mainly from analyses of soil samples stored in the Database of Polish Arable Mineral Soils of the Institute of Agrophysics, Polish Academy of Sciences in Lublin. The Database (Bieganski *et al.* 2013) includes a lot of in-

formation about the basic properties of soils characteristics. It was elaborated for about 1 000 representative profiles of soils in Poland. Soil profiles are numbered from 1 to 1000. Soil No. in tables and figures shows localization (name of place and geographic coordinates) of profile (Gliński *et al.* 1991).

In general, the experimental procedure of the biological activity of soils was as follows:

Air-dry samples, sieved through a 1 mm sieve, were placed in glass jars (vessels), flooded with water (pF 0), but sometimes soil moisture equal to pF 1.5 and 2.2 was established, with and without C and N substrate addition, were incubated in aerobic, flooded or anaerobic (N₂ atmosphere) conditions, at 20°C. Measurements of N₂O, CO₂, O₂, CH₄ concentrations in the headspace gas were made chromatographically, and also Eh, pH, ODR, Fe²⁺ and dehydrogenase activity in the suspension were measured in open vessels with the use of known methods (Aleksandrova and Naidenova 1967, Casida *et al.* 1964, Gliński and Stępniewski 1985, Malicki and Walczak 1983).

Analyses concerned:

1. Production, consumption and release (emission) of nitrous oxide (N₂O) in soils,
2. Soil respiration,
3. Dehydrogenase activity in soils,
4. Methanotrophic activity and methane oxidation in soils.

4. RESULTS

4.1. Production, consumption and release (emission) of N₂O in soils

Samples of 14 Cambisols derived from sand, silt and loam were incubated under flooded conditions at 20°C with nitrate addition (Włodarczyk 2000). The potential ability of soils to produce N₂O in conditions where NO₃⁻ content was a non-limiting factor (100 mg NO₃⁻-N per kg corresponding to 300 kg NO₃⁻-N per ha in 20 cm top soil layer) and the potential ability of soils to consume N₂O were estimated. The phase of N₂O production lasted about 2-21 days. It was found that initial generation of nitrous oxide was followed, after a maximum, by its subsequent absorption. The results showed that these soils were emitters (cumulative production N₂O ranged from 11.4 to 66.5 mg N₂O-N kg⁻¹ of soil) as well as reducers (daily sink of N₂O ranged from 1.3 to 66.5 mg N₂O-N d⁻¹ kg⁻¹ of soil). The range of reduction of N₂O under the investigation conditions was from 10 to 100% of produced N₂O, depending on the kind of soil and time of incubation. Generation of N₂O in Cambisol under anaerobic conditions was shown to be sig-

nificantly related to dehydrogenase activity ($R^2 = 0.69$, $P < 0.001$). The $\text{CO}_2:\text{N}_2\text{O}$ ratio in the gases evolved during incubation increased curvilinearly with Eh and decreased with N_2O production (Włodarczyk *et al.* 2002a). The absorption rate of N_2O was highly correlated with N_2O efflux from flooded soils (Fig. 1) (Włodarczyk *et al.* 2011).

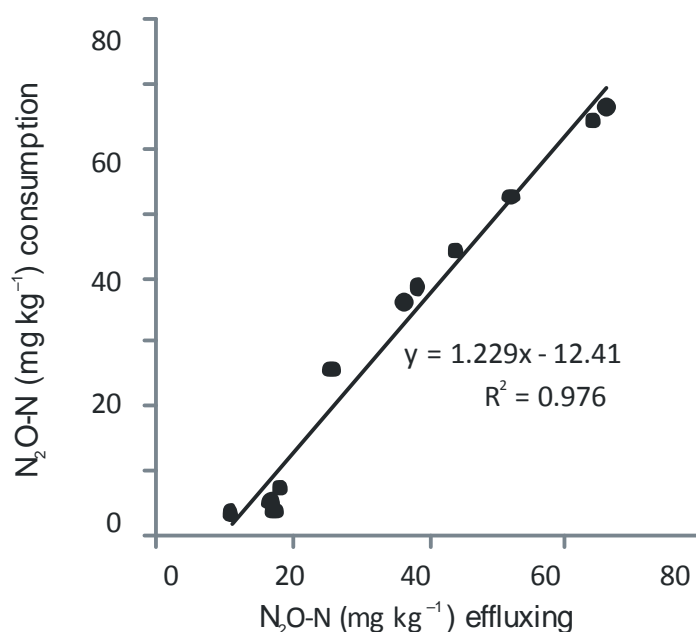


Fig. 1. Maximum N_2O consumption versus maximum cumulative N_2O efflux in flooded Cambisols (Włodarczyk *et al.* 2011)

Samples of Eutric Cambisol soils were also incubated, for 7 days, under flooded conditions in N_2 atmosphere with acetylene (C_2H_2) addition (Włodarczyk *et al.* 2003). In terms of their denitrification activity, the soils were divided into two groups – of lower (I) where diurnal N_2O emission ranged from 1.10 to 6.84 mg $\text{N}_2\text{O-N kg}^{-1} \text{ d}^{-1}$, and higher (II) activity where diurnal N_2O emission ranged from 9.40 to 47.2 mg $\text{N}_2\text{O-N kg}^{-1} \text{ d}^{-1}$. Production of nitrous oxide lasting 3-5 days was followed by its absorption in the case of three soils out of fourteen (where acetylene content dropped below the level blocking N_2O reduction).

Samples of Cambisol derived from sand and Histosol were incubated anaerobically with the addition of KNO_3 (100 mg $\text{NO}_3^- \text{-N kg}^{-1}$ and 2% C_2H_2) for the determination of N_2O emission or with the addition of 1% N_2O for the determina-

tion of N_2O sorption (Włodarczyk *et al.* 2002b). The rates of N_2O , nitrate and redox potential at 20°C were measured over 14 days. The Histosol showed about 4 times higher denitrification activity (as measured by N_2O emission and NO_3^- depletion) than the Cambisol. In turn, the Cambisol was characterised by better capacity for nitrous oxide sorption than the Histosol.

In another experiment, thirteen arable Polish topsoils (0-30 cm) used in the study were Calcaric Regosols (according to FAO/ UNESCO) developed from sand, loamy sand, sandy silt, silty sand and sandy loams. The soils showed a large variation of texture, pH, Corg, and endogenous NO_3^- content. Soil samples were originated from various regions and represented almost the entire territory of Poland. Incubation was under flooded conditions with NO_3^- addition (100 mg NO_3^- -N per kg). Soils developed from sand showed average total nitrous oxide production of about 15 mg N_2O -N kg^{-1} and the value of redox potential corresponding to the maximum N_2O content was +213 mV. Soils developed from silt showed the highest average total denitrification (about 61 mg N_2O -N kg^{-1}) and the corresponding redox potential was +204 mV, while in soils developed from loam, the corresponding values were 36 mg N_2O -N kg^{-1} and +210 mV, respectively. The production of N_2O was accompanied by a drop of redox potential by 84, 105, and 53 mV for sandy, silty and loamy soils, respectively (Włodarczyk *et al.* 2005).

Szarlip *et al.* (2010) examined N_2O production and uptake in Luvisol, Cambisol and Phaeozem soils (Tab. 1) in different (7) variants of aeration, moisture and soil enrichment with organic substrate:

1. Production und uptake of N_2O in control wet soils incubated under aerobic conditions (KT variant),
2. Production und uptake of N_2O in control soils incubated under flooded conditions (KZ variant),
3. Production und uptake of N_2O in wet soils enriched with substrate, incubated under aerobic condition (DT variant),
4. Production und uptake of N_2O in soils enriched with substrate, incubated under flooded conditions (DZ variant),
5. Production und uptake of N_2O in wet soils enriched with substrate, incubated under anaerobic conditions (DB variant),
6. Uptake of added N_2O in wet soils incubated under aerobic conditions (PT variant),
7. Uptake of added N_2O in soils incubated under anaerobic conditions (PB variant).

Table 1. Basic characteristic about tested soils (Szarlip *et al.* 2010)

Soil type	Soil No.	% content of			N _{og} (%)	OM (%)	pH (H ₂ O)	N-NO ₃ ⁻ (mg kg ⁻¹)	N-NH ₄ ⁺ (mg kg ⁻¹)
		sand	silt	clay					
Cambisol (loamy sand)	302	63.5	33.0	3.5	0.110	00.53	7.7	55.80	36.74
Cambisol (loamy sand)	733	68.0	29.6	2.5	0.100	2.09	6.4	4.73	36.69
Luvisol (loamy sand)	27	77.0	21.0	2.0	0.115	1.76	6.5	0.77	36.40
Luvisol (loamy silt)	554	37.7	58.5	3.8	0.105	1.83	5.9	3.90	25.96
Phaeozem (loess)	691	13.9	79.7	6.4	0.115	1.89	7.2	2.97	61.29
Phaeozem (loess)	794	14.7	79.0	6.3	0.155	1.96	7.6	20.29	31.34

OM – organic matter.

The results obtained are gathered in Tables 2-7 and in Figures 2-8 as examples of different soils (Cambisols, Luvisol and Phaeozem). They show variations in data dependent on soil properties, aeration conditions, amendments concerning the amount of N₂O production and rate, N₂O uptake and rate in soils.

The highest values of N₂O production in aerated and flooded conditions found, were as follows:

- 1.96 and 20.22 mg N₂O-N kg⁻¹ – for the amount of produced N₂O under aerobic conditions in control and enriched soils, respectively;
- 0.30 and 9.07 mg N₂O-N kg⁻¹d⁻¹ – for N₂O production rate under aerobic conditions in control and enriched soils, respectively;
- 17.24 and 65.24 mg N₂O-N kg⁻¹ – for the amount of produced N₂O under flooded conditions in control and enriched soils, respectively;
- 3.70 and 23.59 mg N₂O-N kg⁻¹d⁻¹ – for N₂O production rate under flooded conditions in control and enriched soils, respectively.

The highest values of N₂O uptake in aerated and flooded conditions found, were as follow:

- 0.14 and 1.41 mg N₂O-N kg⁻¹ – for the amount of N₂O uptake under aerobic conditions in control and enriched soils, respectively;

- 0.02 and 0.21 mg N₂O-N kg⁻¹ d⁻¹ – for N₂O uptake rate under aerobic conditions in control and enriched soils, respectively;
- 2.59 and 22.83 mg N₂O-N kg⁻¹ – for the amount of N₂O uptake under flooded conditions in control and enriched soils, respectively;
- 0.28 and 6.95 mg N₂O-N kg⁻¹ d⁻¹ – for the amount of N₂O uptake rate under flooded conditions in control and enriched soils, respectively;

The highest values of N₂O production and uptake under anaerobic conditions in enriched soils found, were as follow:

- 14.45 mg N₂O-N kg⁻¹ – for the amount of produced N₂O under anaerobic conditions in enriched soils;
- 9.92 mg N₂O-N kg⁻¹ d⁻¹ – for N₂O production rate under anaerobic conditions in enriched soils;
- 14,45 mg N₂O-N kg⁻¹ – for the amount of N₂O uptake under anaerobic condition in enriched soils;
- 4.96 mg N₂O-N kg⁻¹ d⁻¹ – for N₂O uptake rate under anaerobic conditions in enriched soils, respectively.

The highest values of N₂O uptake rate after N₂O addition to soils found, were as follow:

- 26.35 mg N₂O-N kg⁻¹ d⁻¹ – for N₂O uptake rate of added N₂O under aerobic conditions;
- 78.30 mg N₂O-N kg⁻¹ d⁻¹ – for N₂O uptake rate of added N₂O under anaerobic conditions.

Table 8 shows N₂O production (cumulative amount, mg N₂O-N kg⁻¹) and uptake rates (mg N₂O-N kg⁻¹ d⁻¹) in different aeration conditions, and without or with C and N enriched soils (Szarlip 2010). The highest values of N₂O production and uptake in tested soils, in all variants of aeration and soil enrichment, were 65.24 mg N₂O-N kg⁻¹ for N₂O production and 22.83 mg N₂O-N kg⁻¹ for N₂O uptake.

In control soils, flooded conditions increased N₂O production from 1.96 to 17.24 mg N₂O-N kg⁻¹ and N₂O uptake from 0.14 to 2.59 mg N₂O-N kg⁻¹. In enriched soils the corresponding values were from 20.22 to 65.24 mg N₂O-N kg⁻¹ for production and from 1.41 to 22.83 mg N₂O-N kg⁻¹ for uptake.

The rates of production and uptake were also very differentiated. For control soils the production rates were in the range of 0.30-3.70 mg N₂O-N kg⁻¹ d⁻¹ and for enriched soils from 9.07 to 23.95 mg N₂O-N kg⁻¹ d⁻¹, which was higher in flooded conditions (23.95 mg N₂O-N kg⁻¹ d⁻¹) than in aerobic and anaerobic conditions (9.07 and 9.92 mg N₂O-N kg⁻¹ d⁻¹, respectively).

Table 2. Production and uptake of N₂O in control wet soils incubated under aerobic conditions (KT variant). (Szarlip *et al.* 2010)

Soil type	Soil No.	N ₂ O production			N ₂ O uptake			% of produced
		The highest amount of produced N ₂ O	The highest production rate	The highest amount of N ₂ O uptake	The highest uptake rate	mg N ₂ O-N kg ⁻¹ d ⁻¹	Day	
		mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹	mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹			
Cambisol	302	0.688	0.083	14-21	–	–	–	–
Cambisol	733	0	–	–	–	–	–	–
Luvisol	27	0	–	–	–	–	–	–
Luvisol	554	0.169	0.024	14-21	0.000	–	–	–
Phaeozem	691	0.044	0.011	10-14	0.044	0.006	14-21	100
Phaeozem	794	1.957	0.301	1-3	0.137	0.017	7-10	7

Table 3. Production and uptake of N₂O in control soils incubated under flooded conditions (KZ variant). (Szarlip *et al.* 2010)

Soil type	Soil No.	N ₂ O production			N ₂ O uptake			% of emitted
		The highest amount of produced N ₂ O	The highest production rate	The highest amount of N ₂ O uptake	The highest uptake rate	mg N ₂ O-N kg ⁻¹ d ⁻¹	Day	
		mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹	mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹			
Cambisol	302	5.763	2.275	0-1	0.749	0.113	7-10	13
Cambisol	733	0.501	0.088	3-7	0.070	0.017	7-10	14
Luvisol	27	0	–	–	–	–	–	–
Luvisol	554	0.553	0.553	0-1	0.254	0.085	1-3	46
Phaeozem	691	0.678	0.161	1-3	0.251	0.045	7-10	37
Phaeozem	794	17.24	3.696	1-3	2.586	0.283	14-21	15

Table 4. Production and uptake of N₂O in wet soils enriched with substrate addition incubated under aerobic condition (DT variant). (Szarlip *et al.* 2010)

Soil type	Soil No.	N ₂ O production			N ₂ O uptake		
		The highest amount of produced N ₂ O	The highest production rate	The highest amount of N ₂ O uptake	The highest uptake rate	% of emitted	Day
		mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹	mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹		Day
Cambisol	302	1.487	0.132	0.0	–	–	–
Cambisol	733	7.526	1.290	0.677	0.099	14-21	9
Luvisol	27	5.601	0.968	1.344	0.116	14-21	24
Luvisol	554	7.739	3.608	1.470	0.209	3-7	19
Phaeozem	691	0.004	0.002	0.004	0.001	10-14	100
Phaeozem	794	20.22	9.069	1.415	0.184	7-10	7

Table 5. Production and uptake of N₂O in soils enriched with substrate addition incubated under flooded conditions (DZ variant). (Szarlip *et al.* 2010)

Soil type	Soil No.	N ₂ O production			N ₂ O uptake		
		The highest amount of produced N ₂ O	The highest production rate	The highest amount of N ₂ O uptake	The highest uptake rate	% of produced	Day
		mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹	mg N ₂ O-N kg ⁻¹	mg N ₂ O-N kg ⁻¹ d ⁻¹		Day
Cambisol	302	65.24	23.59	22.83	3.216	3-7	35
Cambisol	733	3.812	3.812	3.812	1.444	1-3	100
Luvisol	27	0.174	0.087	0.174	0.044	3-7	100
Luvisol	554	7.547	7.547	7.547	3.428	1-3	100
Phaeozem	691	4.457	2.228	4.279	0.962	3-7	96
Phaeozem	794	16.71	16.71	13.87	6.949	1-3	83

Table 6. Production and uptake of N₂O in wet soils enriched with substrate addition incubated under anaerobic conditions (DB variant), (Szarlip *et al.* 2010)

Soil type	DB	Soil No.	N ₂ O production			N ₂ O uptake		
			The highest amount of produced N ₂ O	The highest production rate	The highest amount of N ₂ O uptake	The highest uptake rate	% of produced	
			mg N ₂ O-N kg ⁻¹ Day	mg N ₂ O-N kg ⁻¹ d ⁻¹ Day	mg N ₂ O-N kg ⁻¹ Day	mg N ₂ O-N kg ⁻¹ d ⁻¹ Day		
Cambisol		302	5.216	5.216	5.216	2.608	1-3	100
Cambisol		733	2.663	2.663	2.663	1.325	1-3	100
Luvisol		27	4.055	1.910	4.055	1.014	3-7	100
Luvisol		554	6.216	6.216	6.216	3.108	1-3	100
Phaeozem		691	14.45	6.795	14.453	2.579	3-7	100
Phaeozem		794	9.920	9.920	9.920	4.960	1-3	100

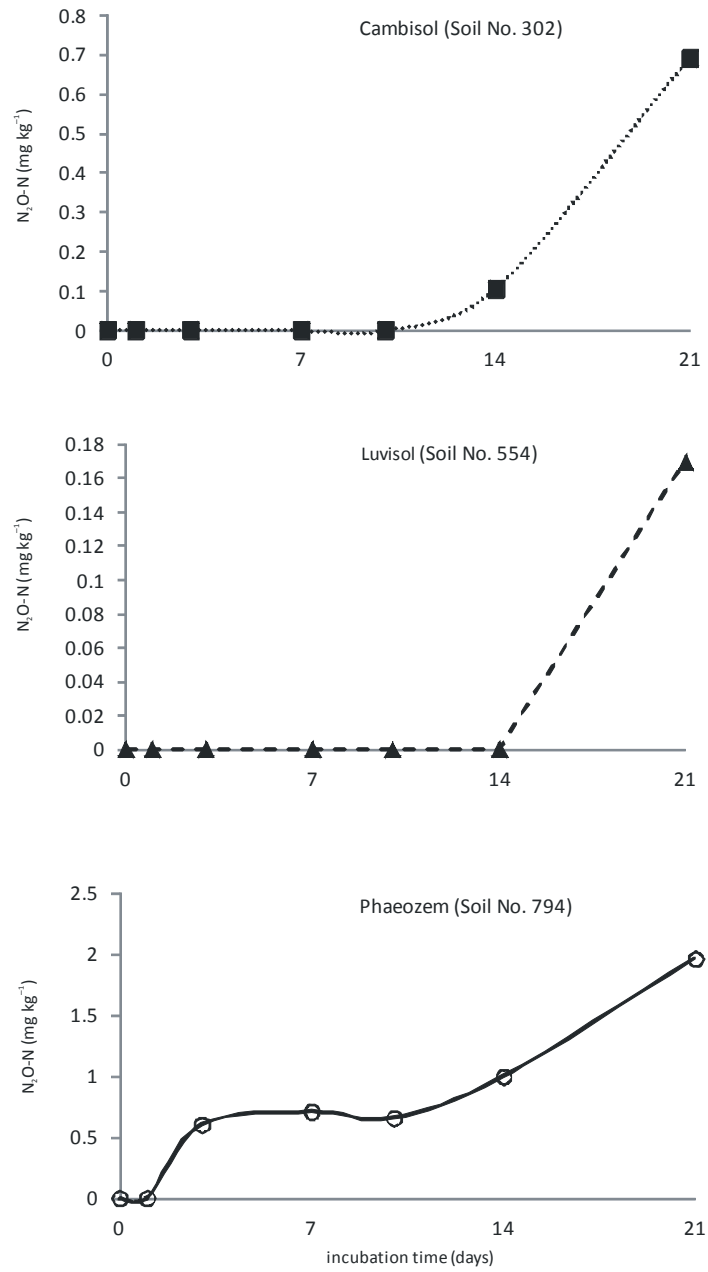


Fig. 2. Changes in the concentration of N_2O in control soils incubated under aerobic conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

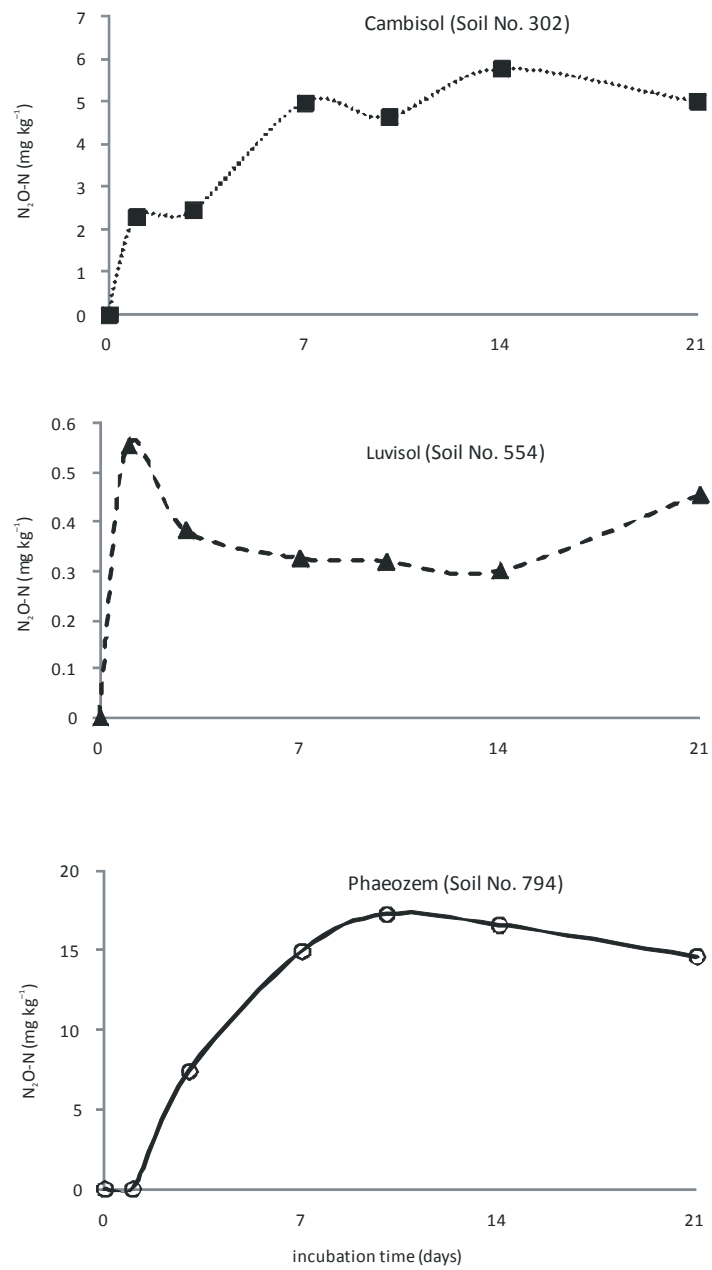


Fig. 3. Changes in the concentration of N₂O in control soils incubated under flooded conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

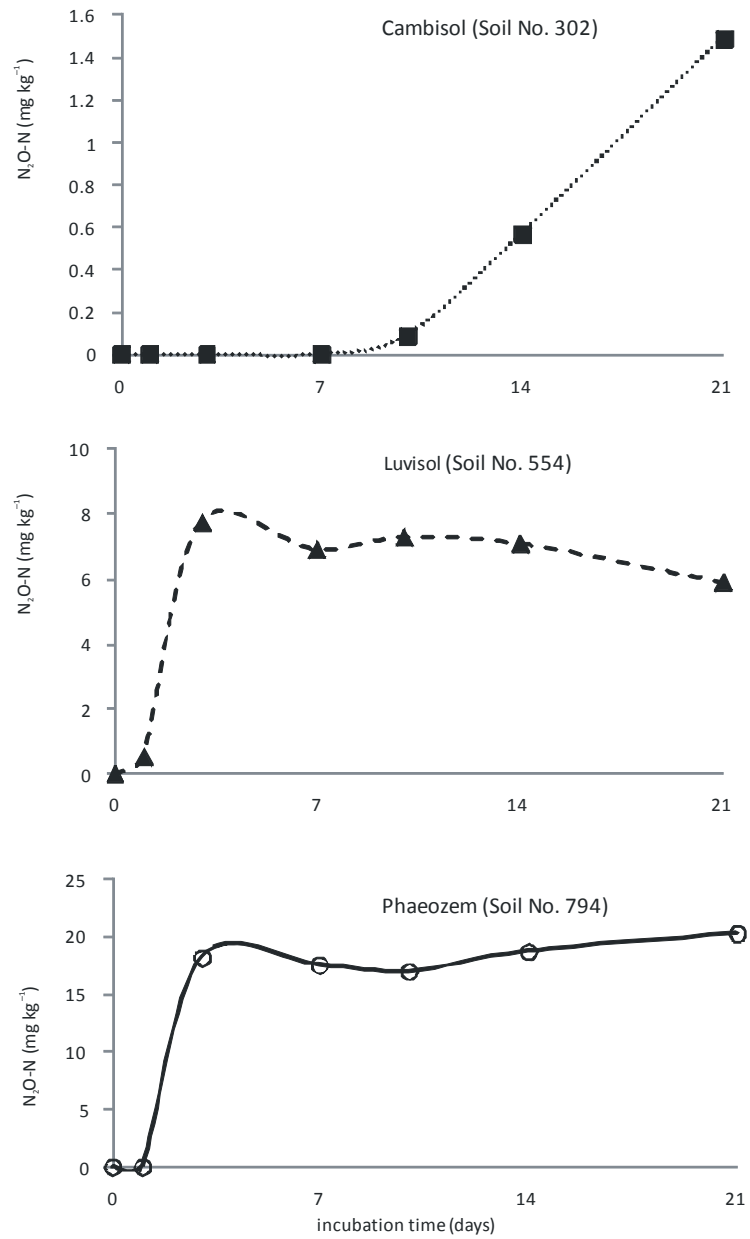


Fig. 4. Changes in the concentration of N_2O in control soils enriched with organic substrates incubated under aerobic conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

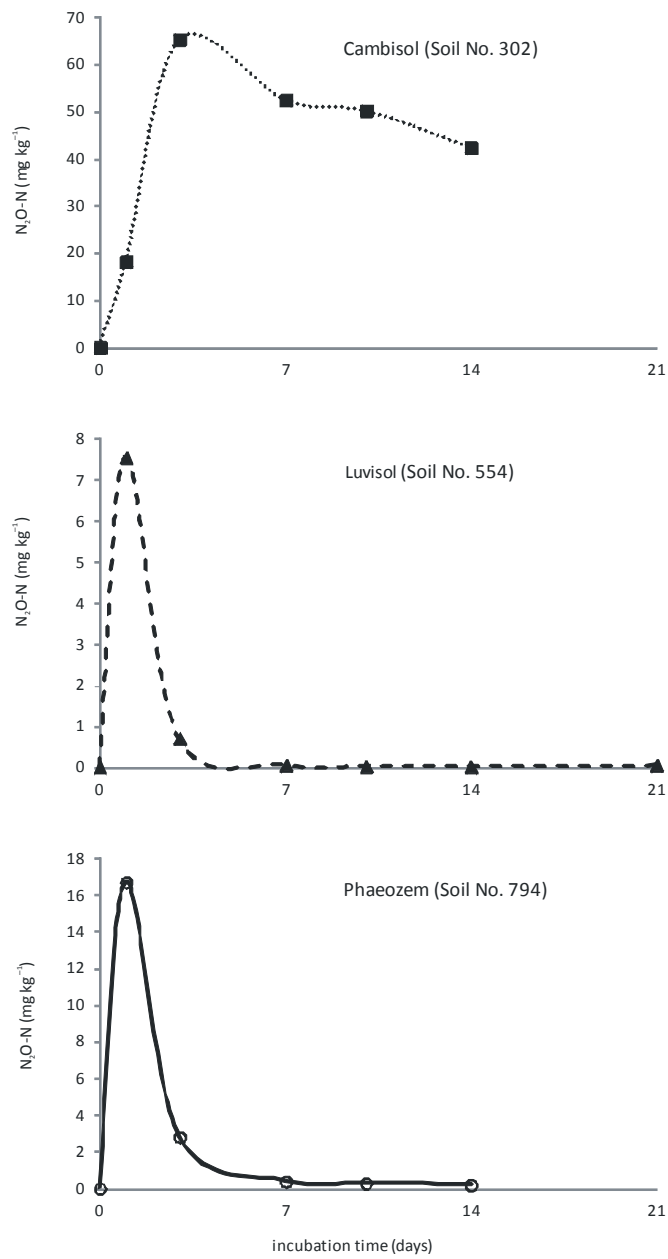


Fig. 5. Changes in the concentration of N₂O in control soils enriched with organic substrates incubated under flooded conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

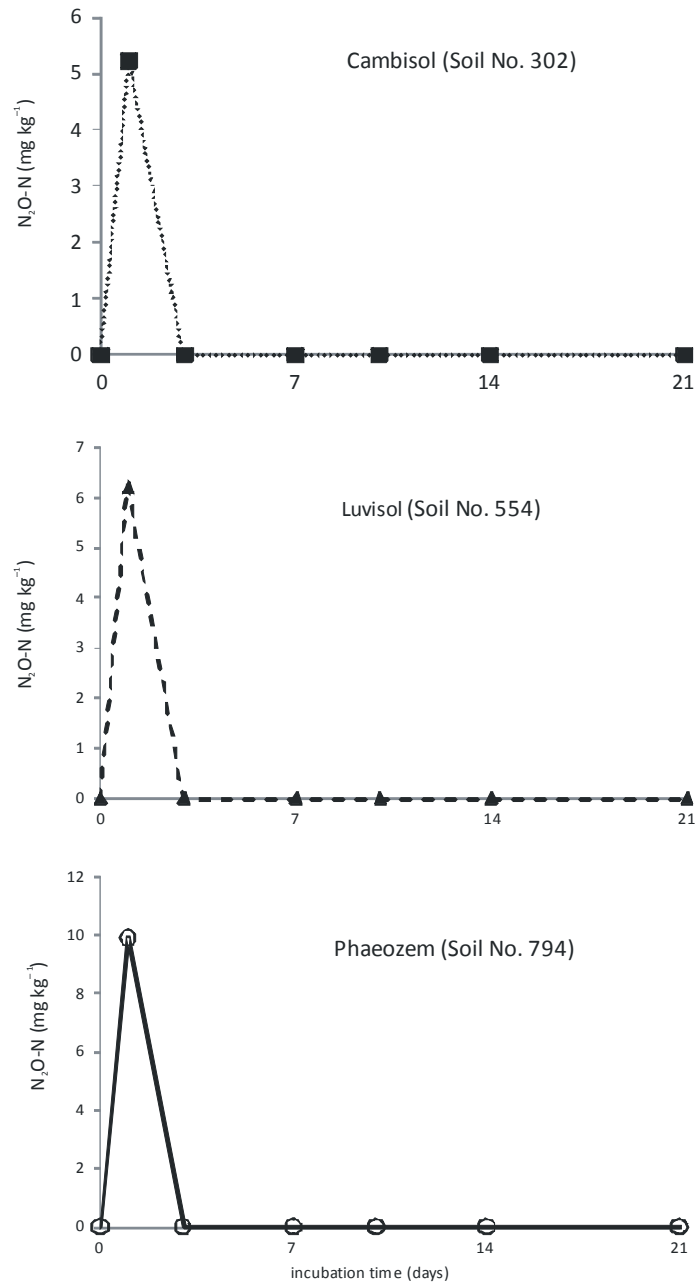


Fig. 6. Changes in the concentration of N_2O in control soils enriched with organic substrates incubated under anaerobic conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

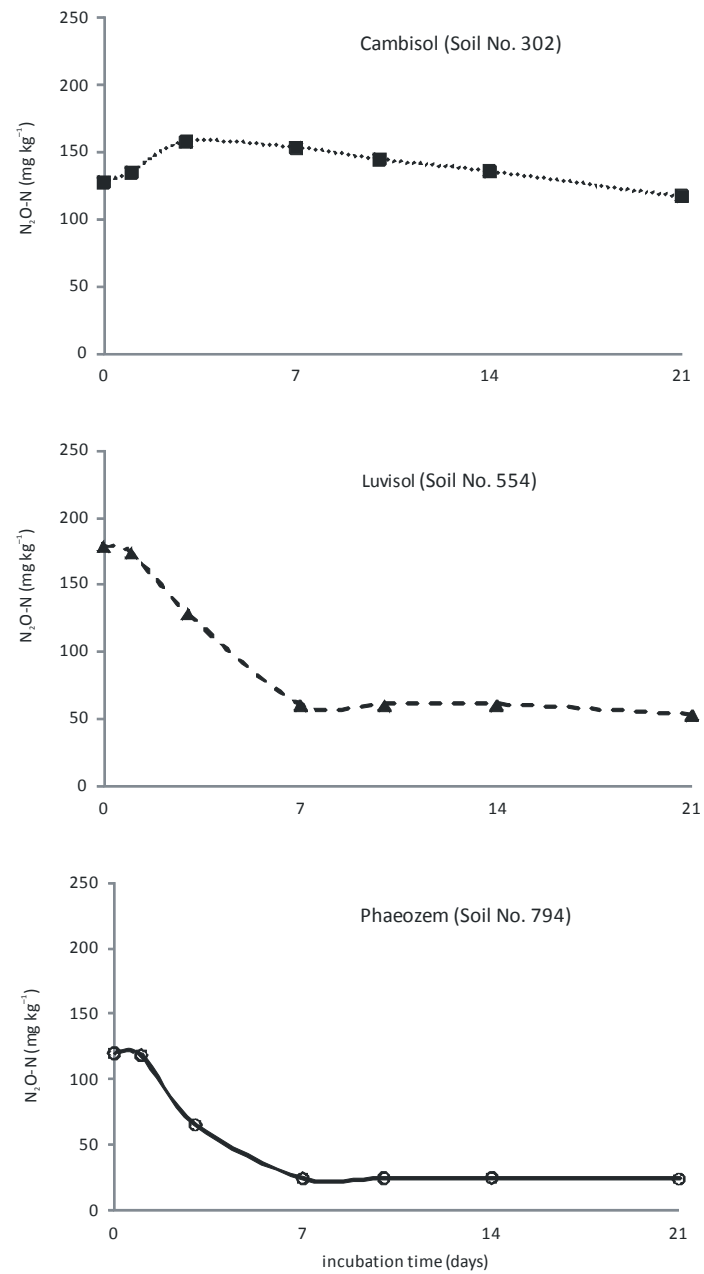


Fig. 7. Uptake of added nitrous oxide to soils incubated under aerobic conditions. (Szarlip *et al.* 2010)

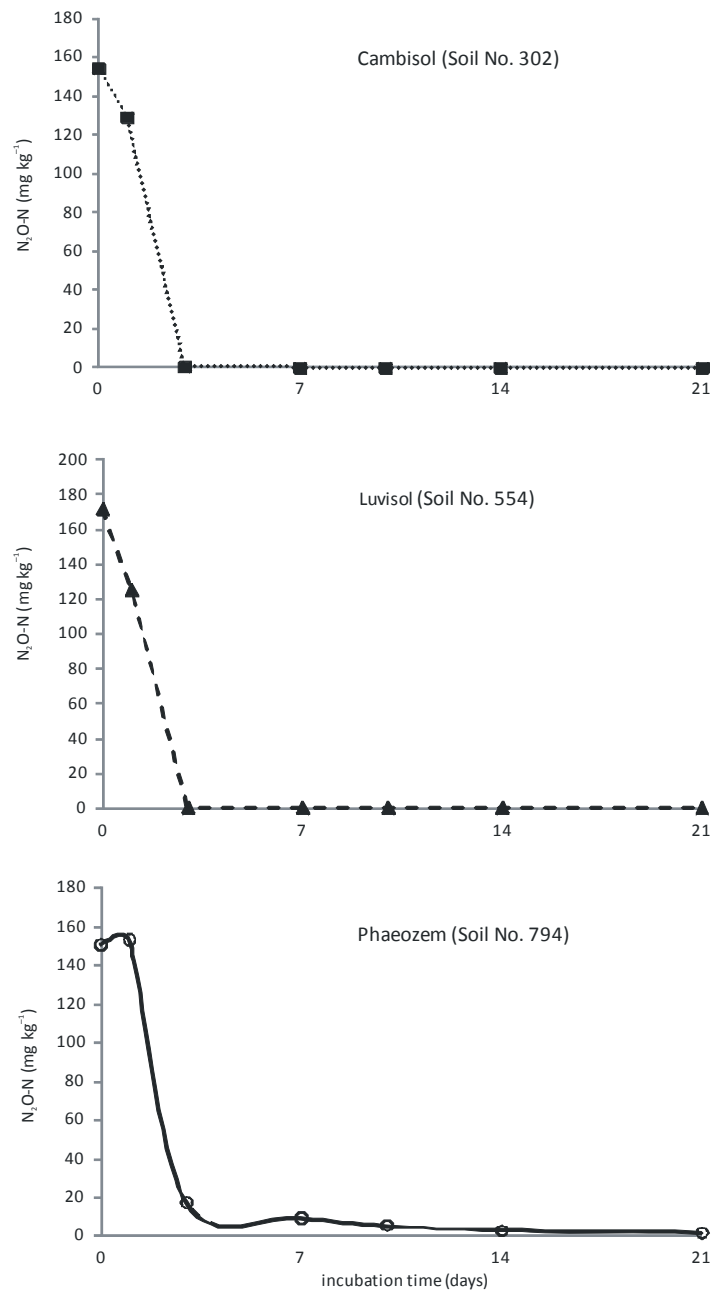


Fig. 8. Uptake of added nitrous oxide to soils incubated under anaerobic conditions. Note different scales on the graphs (Szarlip *et al.* 2010)

Table 7. The highest uptake rates of added N₂O (106-171 mg kg⁻¹) to soils incubated under aerobic and anaerobic conditions (PT and PB variants) (Szarlip *et al.* 2010)

PT		Aerobic conditions			Anaerobic conditions		
Soil type	Soil No.	mg N ₂ O-N kg ⁻¹ d ⁻¹	Day	% of maximum	mg N ₂ O-N kg ⁻¹ d ⁻¹	Day	% of maximum
Cambisol	302	2.93	7-10	26	64.50	1-3	
Cambisol	733	3.44	14-21	25	78.30	1-3	100
Luvisol	27	3.42	14-21	16	24.09	0-1	100
Luvisol	554	23.19	1-3	71	62.56	1-3	100
Phaeozem	691	5.65	14-21	19	22.71	3-7	100
Phaeozem	794	26.35	1-3	80	67.78	1-3	90

Table 8. The highest values of N₂O production and uptake amount (mg N₂O-N kg⁻¹) and rate (mg N₂O-N kg⁻¹d⁻¹) under different aeration conditions and without or with C and N addition (Szarlip *et al.* 2010)

N ₂ O-N	Control soils		Enriched soils		
	aerobic	flooded	aerobic	flooded	anaerobic
	Amount				
Production	1.96	17.24	20.22	65.24	14.45
Uptake	0.14	2.59	1.41	22.83	?
	Rate				
Production	0.30	3.70	9.07	23.95	9.92
Uptake	0.02	0.28	0.21	6.95	4.96
			26.31*		78.30*

*after N₂O addition.

Uptake rates were much lower than production rates (0.02-0.28 mg N₂O-N kg⁻¹ d⁻¹ in control soils and 0.21-6.95 mg N₂O-N kg⁻¹ d⁻¹ in enriched soils) and were also higher in flooded conditions (6.95 mg N₂O-N kg⁻¹ d⁻¹) than in aerobic and anaerobic conditions (0.21 and 4.96 mg N₂O-N kg⁻¹ d⁻¹, respectively). N₂O addition to soils caused very high N₂O uptake rate in anaerobic conditions (78.30 mg N₂O-N kg⁻¹d⁻¹) and less in aerobic conditions (26.35 mg N₂O-N kg⁻¹d⁻¹).

Włodarczyk *et al.* (2004b) carried out an experiment on nitrate stability in Cambisols and Phaeozems formed from loess. Soils amended with KNO₃ in doses of 50, 100, 300 and 500 mg N₂O-N kg⁻¹ dry soil were incubated under flooded condi-

tions. The soils differed as to the denitrification capacity for high 60-90 N₂O-N kg⁻¹ (first group) and low 10-20 N₂O-N kg⁻¹ (second group). The experiment allowed to determine N₂O-N emission maxima: cumulative (124 N₂O-N kg⁻¹), diurnal (26.9 mg N₂O-N kg⁻¹ d⁻¹), NO₃⁻-N denitrified (41.6%), and diurnal absorption (2.72 mg N₂O-N kg⁻¹ d⁻¹) and N₂O-N absorbed (32.5%). The maximum denitrification of tested soils is shown in Figure 9. The maximum denitrification activity of the first soil group showed a very high non-linear correlation with NO₃⁻-N at the moment of maximum N₂O concentration (total NO₃⁻-N minus N₂O-N emitted). The denitrification rate increased with NO₃⁻-substrate within its entire concentration range under study. The denitrification rate of the soils of the second group increased with the concentration up to 100 mg N₂O-N kg⁻¹ and remained at the same level at higher concentrations. Figure 9 shows maximum emission of N₂O for non-amended soils vs. native NO₃⁻-N content.

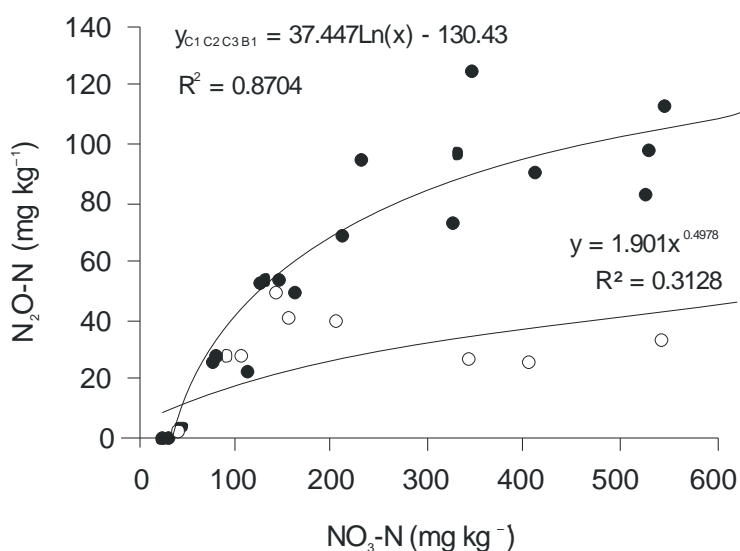


Fig. 9. The maximum denitrification during anaerobic incubation of Cambisols and Phaeozems amended with NO₃⁻ for the soils with high (●) and low (○) denitrification capacity (Włodarczyk *et al.* 2004b)

4.1.1. Redox potential (Eh) effect

Soil is heterogeneous and commonly has both aerobic and anaerobic sites (Stepniewska 2011). The soil oxidation-reduction status (Eh) has been shown to be an important factor affecting soil biological activity and transformations of natural compounds in soils. The oxygen status in soil, which is inversely propor-

tional to soil moisture, appears to be one of the key factors influencing N_2O production and consumption. The reduction of NO_3^- in soil suspensions occurs sequentially at the corresponding soil redox potential (Eh) values (Włodarczyk *et al.* 2003, 2005, Szarlip *et al.* 2010).

Włodarczyk (2000) indicated that N_2O emission and absorption in soil derived from different parent material can occur when Eh falls below 300 mV under flooded conditions. Nitrous oxide content in the headspace existed in equilibrium with nitrous oxide content in soil within a narrow redox potential interval +190-(+240) mV with maximum at about +200 mV (Fig. 10). The redox potential about +200 mV is the limit value between the production of nitrous oxide and its consumption (Włodarczyk 2000, Włodarczyk *et al.* 2005).

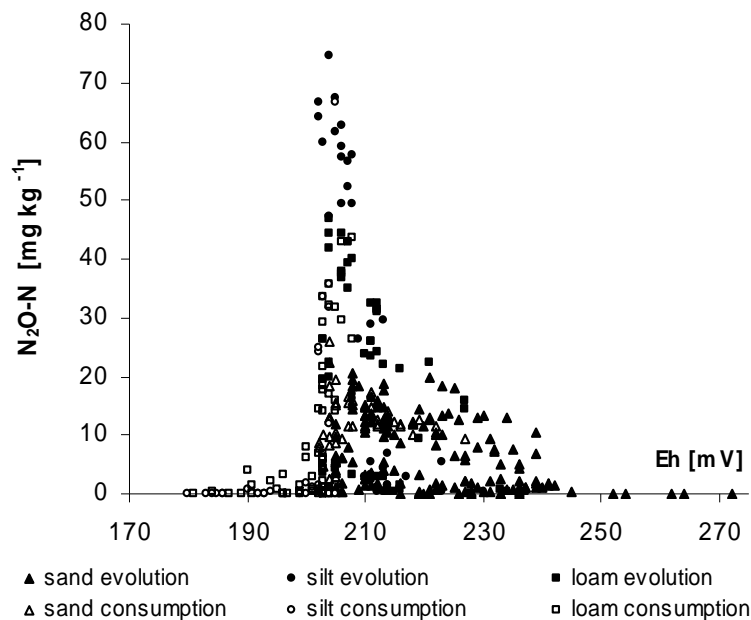


Fig. 10. Relationship between the N_2O content and the redox potential of the evolution and consumption phases in soils derived from different parent materials (Włodarczyk *et al.* 2005)

In loess soils (Cambisols and Phaeozems) incubated under anaerobic conditions nitrous oxide was formed at redox potentials below +200 mV and started to disappear at negative Eh values (Włodarczyk *et al.* 2004b).

Pre-incubated Cambisols investigated under anaerobic conditions were characterised by a very wide range of redox potential measured for the maximal cu-

ulative N₂O emission - from +417 to +233 mV. The beginning of N₂O emission from the light textured soils was observed above +400 mV, while from the heavier textured soil below +400 mV. N₂O emission was correlated with soil redox potential (Włodarczyk *et al.* 2003).

Production and reduction of N₂O in Cambisol derived from different parent material were nonlinearly correlated with redox potential ($R^2 = 0.906$ and $R^2 = 0.966$, respectively). Redox potential showed a negative correlation with pH value ($R^2 = 0.685$). Eh value decreased with decreasing of NO₃⁻-N in the range from about 10 to 100 mg NO₃⁻-N kg⁻¹ of soil. The highest daily reduction of nitrate was observed in a narrow range of Eh (+200-+210 mV). The boundary nitrate concentration, resulting in a distinct drop of redox potential, was about 100 mg NO₃⁻-N kg⁻¹ (Włodarczyk 2000).

The final cumulative N₂O production in anaerobic Cambisol derived from sand, silt and loam decreased linearly with Eh (Włodarczyk *et al.* 2002a). Redox potential was not influenced by the form of N which was added to Cambisol derived from sand, incubated anaerobically (Włodarczyk 2002b).

Włodarczyk *et al.* (2005) found very high correlation coefficients of the head-space nitrous oxide content versus the redox potential, both for evolution and consumption phases. In almost all the cases significant curvilinear correlations were found. They were positive for the consumption phase and negative for the production phase. The highest influence, both on production and consumption of N₂O, was found in the case of silt fraction.

4.1.2. pH effect

One of the factors significantly affecting nitrate reduction process including the process of N₂O emissions and absorption is undoubtedly pH. The optimum pH for N₂O emission via denitrification varies with species and age of the microbial population and nitrate concentration, but most denitrifiers have optimum pH for growth between 6 and 8 (Szarlip *et al.* 2010).

Włodarczyk (2000) found that under flooded conditions the maximum emission of N₂O from Calcaric Regosols (Cambisols) was observed at pH range between 4.5 and 6, but maximum absorption of nitrous oxide occurred at pH from about 5.5 to about 7.

The final cumulative N₂O production increased curvilinearly with pH value in Cambisol incubated under anaerobic condition (Włodarczyk *et al.* 2002a).

4.1.3. Effect of NO_3^- and C organic content in soil as a substrate for denitrification

Total denitrification fluxes (N_2O plus N_2) are directly proportional to soil NO_3^- concentrations when the other important component, readily metabolised organic substrate, is also present and non rate-limiting. When a lack of metabolisable organic matter limits potential denitrification, N_2 plus N_2O fluxes do not increase with increasing NO_3^- concentration (Szarlip *et al.* 2010)

A laboratory study with six loess soils (three Cambisols and three Phaeozems) incubated under anaerobic conditions examined the effect of a wide range of NO_3^- doses on soil redox potential and N_2O emission or absorption. Due to the fact that loess soils are usually well-drained and are expected to be absorbers during prevailing part of the season, the study aimed at determination of the conditions decisive for the process of transition from emission to absorption. On the basis of the response to soil nitrate level, two groups of soils were distinguished – with high and low denitrification capacity. The soil denitrification activity showed Michaelis-Menten kinetics with respect to soil nitrate content with K_M in the range of 50-100 mg NO_3^- -N kg^{-1} . The percentage of nitrates converted to N_2O increased linearly with nitrate concentration in the range from 25 to 100 mg NO_3^- -N kg^{-1} up to 43% and decreased linearly at higher concentrations reaching practically zero at concentrations of about 600 mg NO_3^- -N kg^{-1} . No denitrification was observed below 25 mg NO_3^- -N kg^{-1} . Nitrous oxide absorption in soil occurred only at nitrate concentrations up to 100 mg NO_3^- -N kg^{-1} and in this concentration range it was proportional to the denitrification rate (Włodarczyk *et al.* 2004b).

Under flooded conditions, the tested Cambisol showed different activity in the reduction of nitrate, in which the NO_3^- reduction took place simultaneously with the reduction of N_2O , and the soil in which the reduction of N_2O began after depletion of nitrate. The range of reduced nitrate fluctuated from 22 to 100% depending on the kind of soil and time of incubation (Włodarczyk 2000).

The total N_2O amount in Calcaric Regosols (Cambisols) under anaerobic incubation reached from 3 to 91% of the initial nitrate – N content depending of the soil type (Włodarczyk *et al.* 2003). The percentage of nitrate reduced equals 35, 97 and 100% for Cambisols incubated under flooded conditions derived from sand, loam and silt, respectively (Włodarczyk *et al.* 2011).

The percentage of NO_3^- -N denitrified to N_2O -N in Cambisol incubated under anaerobic condition was positively correlated with organic matter content in soil ($R^2 = 0.52$, $P < 0.01$).

Denitrification rate and sink of nitrous oxide in Cambisol incubated under flooded conditions showed high correlation with mineralization of organic matter ($R^2 = 0.906$ and $R^2 = 0.913$, respectively) (Włodarczyk 2000).

Diurnal N_2O production in 14 Cambisols incubated under flooded conditions was positively correlated with C_{org} content and diurnal CO_2 emission ($R^2 = 0.95$, $P < 0.001$) (Włodarczyk *et al.* 2003).

4.1.4. Role of acetylene

Acetylene (C_2H_2) is well known for its inhibitory properties and interaction with such microbial processes as nitrogen fixation, nitrous oxide reduction, ammonium, methane and ethylene oxidation, methane and ethylene production, anaerobic methane oxidation, and respiration. C_2H_2 has several advantages as an inhibitor of biochemical processes in soil. It can be used during short-term incubations since it rapidly moves through air-filled pores in soil, is highly soluble in water, and its addition to soil will neither alter the water content nor cause any displacement of water-soluble compounds in soil. Although C_2H_2 is not a natural product (its main sources are coming from anthropogenic activity connected with motorization and industry), the ability of bacteria to grow on C_2H_2 has been observed in different soils and sediments (Brzezińska *et al.* 2011a).

The effect of C_2H_2 on CO_2 production and O_2 uptake by microbial biomass (C_{mic}) under different air-water conditions in Histosol and was examined by Brzezińska *et al.* (2011a). Soil samples (silty Cambisol and Histosol) were enriched with C_2H_2 and incubated at 20°C under wet (60% WHC) or flooded conditions. Soils differed in their capacity for C_2H_2 consumption. Histosol utilised more C_2H_2 than Cambisol, maximum 54.03 vs. 19.25 mmol kg^{-1} , respectively. C_2H_2 uptake was influenced by the air-water conditions, and it was faster and larger in flooded than in wet conditions (16.2 and 7.81 mmol kg^{-1} , respectively). On average, 80% and 53% of initial C_2H_2 disappeared from the headspace of flooded and wet soil, respectively. Regression analysis showed that net CO_2 production, net O_2 consumption, and C_{mic} (calculated as differences between enriched with C_2H_2 and control variants) were linearly positively correlated with the quantity of consumed acetylene.

4.1.5. Soil texture effect

Soil texture is a good predictor of denitrification rates at the landscape scale part because it captures the interaction between water content and soil porosity with respect to gas and solute diffusion path length (Szarlip *et al.* 2010).

Our studies carried out on different soils, where the denitrification process occurred under conditions of flooding, as well as in soil without access of oxygen, indicate a strong correlation between both efflux and N_2O consumption and soil texture (Włodarczyk 2000, Włodarczyk *et al.* 2003, 2005, 2011).

The N₂O absorption rate for the three Calcaric Regosols (Cambisols) of different texture, incubated anaerobically, was 0.16 mg N₂O-N kg⁻¹ day⁻¹, 20.6 mg N₂O-N kg⁻¹ day⁻¹ and 3.3 mg N₂O-N kg⁻¹ day⁻¹ in sandy, loamy and silty soils, respectively (Włodarczyk *et al.* 2003). The rate of N₂O disappearance averaged 0.56, 4.08 and 11.7 mg N₂O-N kg⁻¹ day⁻¹ in sandy, loamy and silty soils, respectively (Włodarczyk *et al.* 2005).

The total amount of N₂O release from Calcaric Regosols (Cambisols) under flooded conditions was highest in the silty soils and the lowest in the sandy soils (Fig. 11). Average daily N₂O production in flooded soils was negatively correlated with the >0.05 mm fraction and positively with the finer fractions *i.e.* with the 0.05-0.002 mm and <0.002 mm fraction (Włodarczyk *et al.* 2005).

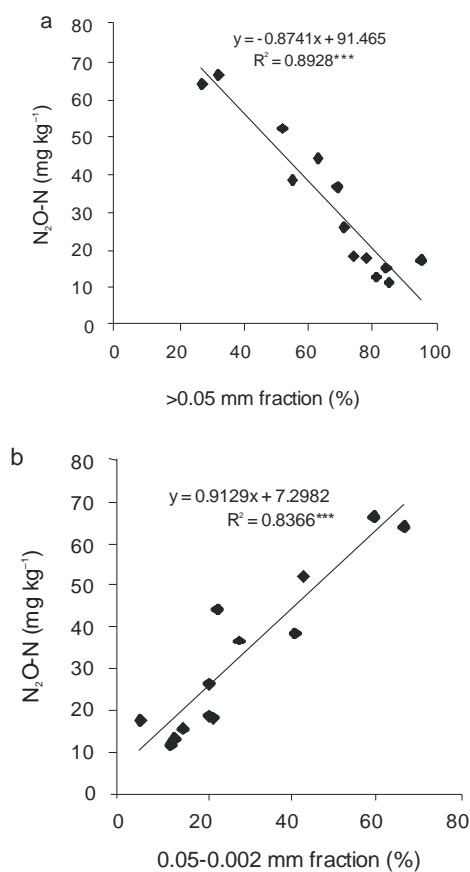


Fig. 11. Total N₂O-N content in the headspace versus the content of particle size fractions (Włodarczyk *et al.* 2005, 2011)

The rate of N₂O consumption for individual Calcaric Regosol (Cambisol) soils was positively correlated with the 0.05-0.002 mm fraction and negatively with the >0.05 mm fraction. N₂O reduction to N₂ began earlier in finely (*e.g.* loam) than in coarsely textured (*e.g.* sand) soils (Włodarczyk *et al.* 2005).

Total N₂O consumption in Cambisols under flooded conditions ranged between 3.3 and 66.5 mg N kg⁻¹, and constituted 32.9, 99.2, and 100% of the produced N₂O for the sandy, loamy and silty soil samples, respectively. The tested soils were characterized by various ratios of N₂O emitted to consumed. Most of the sandy samples were characterized by a weak capacity for N₂O production and consumption, while loamy and silty soils were characterised by a good or very good capacity for N₂O production and consumption (Włodarczyk *et al.* 2011).

4.1.6. Temperature effect

The influence of temperature in the range of 4-20°C on the rate of denitrification of Cambisol and Phaeozem (Włodarczyk *et al.* 2001) is shown in Figure 12. The average maximum cumulative efflux of N₂O was 1.7 and 1.2 times higher in the range of 4 to 10°C and from 10 to 20°C, respectively. The Q₁₀ for the rate of denitrification was 8.4 and 4.2 in the temperature ranges of 4-14°C and 10-20°C, respectively.

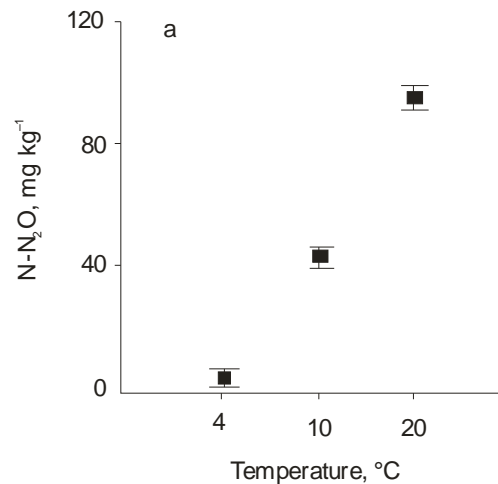


Fig. 12. N₂O-N content in the headspace as a function of temperature in Phaeozem (a) and Cambisol (b) (average values of 7 days of incubation) (Włodarczyk *et al.* 2001)

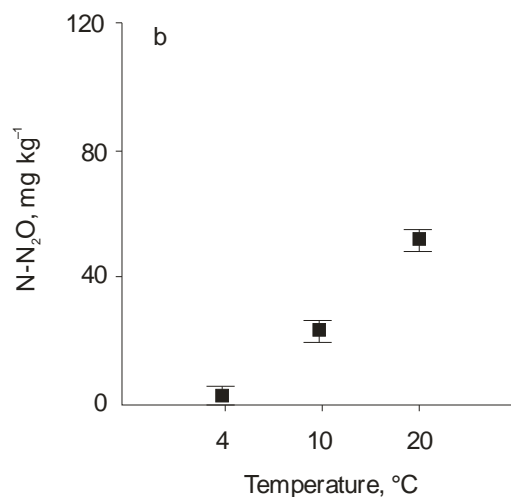


Fig. 12. Cont. N₂O-N content in the headspace as a function of temperature in Phaeozem (a) and Cambisol (b) (average values of 7 days of incubation) (Włodarczyk *et al.* 2001)

4.2. Soil respiration

The respiration activity (CO₂ production and O₂ consumption) of 28 soils (Tab. 9), representing typical soils of the territory of Poland (16 Cambisols, 9 Luvisols and 3 Phaeozems), incubated at 20°C for 14, 24 or 56 days under aerobic (wet) or flooded conditions, amended or not with C and N substrate, was studied in four variants (Gliński *et al.* 2010):

1. Flooded soils,
2. Wet soils,
3. Flooded soils amended with C and N substrate,
4. Wet soils amended with C and N substrate.

The results of measurements of changes in CO₂ production and O₂ consumption in the soil headspace during incubation are shown in Tables 10-17 and Figures 13-20.

In flooded conditions, the highest production of CO₂ at the end of incubation was in the range of 213-523 mg CO₂-C kg⁻¹ for Cambisols, 345-474 mg CO₂-C kg⁻¹ for Luvisols and 475-599 mg CO₂-C kg⁻¹ for Phaeozems. The lowest O₂ consumption was in the range of 12.3-15.7% v/v for Cambisols, 10.2-11.3% v/v for Luvisols and 12.2-15.9% v/v for Phaeozems (Tabs 10 and 14, Figs 13 and 17).

Table 9. Basic characteristic of tested soils (Gliński *et al.* 2010)

Soil type	Soil No.	% of soil fraction (mm)			Organic matter (%)	pH (H ₂ O)
		1-0.1	0.1-0.02	<0.02		
Cambisol	23	58	28	14	0.73	6.3
Cambisol	34	50	25	25	1.19	6.9
Cambisol	116	79	11	10	0.64	7.4
Cambisol	197	87	12	1	0.79	6.0
Cambisol	250	83	8	9	0.81	6.0
Cambisol	302	67	22	11	0.58	7.7
Cambisol	317	49	47	4	0.42	6.8
Cambisol	834	70	17	13	0.32	7.5
Cambisol	528	83	12	5	0.78	6.5
Cambisol	541	64	24	12	0.65	6.7
Cambisol	714	5	68	27	0.98	7.6
Cambisol	733	72	20	8	1.01	6.4
Cambisol	760	87	9	4	0.88	6.0
Cambisol	785	0	58	42	0.90	6.8
Cambisol	755	0	71	29	0.70	6.6
Cambisol	914	76	16	8	0.87	6.3
Luvisol	27	88	6	6	0.86	6.5
Luvisol	74	49	42	9	0.47	6.2
Luvisol	122	42	43	15	0.83	6.9
Luvisol	173	75	19	6	0.90	6.0
Luvisol	327	58	25	17	0.70	6.4
Luvisol	425	40	39	21	0.71	6.7
Luvisol	433	65	21	14	0.56	6.3
Luvisol	554	38	57	15	1.12	5.9
Luvisol	633	44	40	16	0.59	6.8
Phaeozem	601	1	50	49	0.83	7.1
Phaeozem	691	2	41	57	1.86	7.2
Phaeozem	794	0	68	32	1.06	7.6

In wet soils, the highest production of CO₂ at the end of incubation was in the range of 142-476 mg CO₂-C kg⁻¹ for Cambisols, 208-450 mg CO₂-C kg⁻¹ for Luvisols and 420-486 mg CO₂-C kg⁻¹ for Phaeozems. The lowest O₂ consumption was in the range of 12.2-17.9% v/v for Cambisols, 13.3-14.3% v/v for Luvisols and 13.3% v/v for Phaeozems (Tabs 11 and 15, Figs 14 and 18).

In flooded soils amended with C and N substrate the highest production of CO₂ at the end of incubation was in the range of 410-751 mg CO₂-C kg⁻¹ for Cambisols, 444-733 mg CO₂-C kg⁻¹ for Luvisols and 619-787 mg CO₂-C kg⁻¹ for Phaeozems. The lowest O₂ consumption was in the range of 9.6-14.4% v/v for Cambisols, 13.4-13.9% v/v for Luvisols and 10% v/v for Phaeozems (Tabs 12 and 16, Figs 15 and 19).

In wet soils amended with C and N substrate the highest production of CO₂ at the end of incubation was in the range of 455-538 mg CO₂-C kg⁻¹ for Cambisols, 552-635 mg CO₂-C kg⁻¹ for Luvisols and 397-623 mg CO₂-C kg⁻¹ for Phaeozems. The lowest O₂ consumption was in the range of 9.6-14.4% v/v for Cambisols, 13.4-13.9% v/v for Luvisols and 10% v/v for Phaeozems (Tabs 13 and 17, Figs 16 and 20).

The results of the CO₂ production and O₂ consumption in the soils, measured after 14 days, are presented in Figure 21. In wet soils the cumulative CO₂ release by Cambisols, on average 188.24±55.9 mg CO₂-C kg⁻¹, was close to the amount of CO₂ produced by Luvisols (196.78 ± 32.5 mg CO₂-C kg⁻¹), while both amounts were lower by about 40% than CO₂ released by Phaeozems (307.14 ± 33.8 mg CO₂-C kg⁻¹, p < 0.001).

Similar differences between soil types were observed when the soils were incubated under flooded conditions. The CO₂ production in flooded soils was slightly higher than in wet soils, especially in Phaeozems which accumulated on average 356.32 ± 15.8 mg CO₂-C kg⁻¹. During this incubation period, CO₂ production was accompanied by oxygen uptake from the headspace, in the range from approximately 3% v/v in Luvisols and Cambisols to 4.53 ± 0.5 and 5.26 ± 0.1% v/v in Phaeozems (when incubated under wet and flooded conditions), respectively. Longer incubation resulted in the accumulation of up to about 600 mg CO₂-C kg⁻¹, and consumption of O₂ from the headspace down to 10% v/v (Gliński *et al.*, 2010). Figure 22 shows CO₂ versus O₂ in the headspace during 60-day incubation of tested topsoils (all results included, n = 716). Analysis of regression showed that even if various soils were included, the correlation between CO₂ and O₂ remained significant (r = 0.913, p < 0.001). It can be observed, however, that individual Luvisol and Cambisol showed respiration activity apparently lower than most of the tested soils, i.e. lower final CO₂ of about 230 mg C kg⁻¹ and lower O₂ uptake resulting in higher residual O₂ in the headspace at the end of the incubation (about 14% v/v).

Table 10. Emission of CO₂ (mg kg⁻¹) during incubation of tested soils at pF 0 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)													
		1	3	5	7	10	14	18	21	24	28	35	42	49	56
Cambisol	23	6.778	77.10		145.5	175.5	214.9	237.7	270.8		321.2	364.5	405.6		
Cambisol	34	39.90	138.9		234.4	277.5	333.9	366.5	410.1		449.9	488.0	523.0		
Cambisol	116	29.88	88.27		144.0	179.4	216.9	241.6	269.2		280.2	309.4	347.5		
Cambisol	197	20.60	73.27	109.7	124.1	150.1	171.6	190.9	206.6	226.1	233.7	264.4	295.3	306.0	305.3
Cambisol	250	5.016	45.76		79.59	92.06	109.7	119.8	134.1		159.9	189.8	213.2		
Cambisol	302	24.33	74.30	91.65	110.5	144.1	171.0	176.6	182.1	202.1	213.0			262.6	
Cambisol	317	29.79	71.95		105.2	120.8	144.0	159.7	190.8		195.1	218.0	230.6		
Cambisol	834	31.49	63.97	82.21	101.5	120.5	151.9	163.8	182.7	188.7	208.6			238.3	
Cambisol	528	31.53	101.6		159.6	187.6	228.8	248.5	266.4		287.7	312.2	323.9		
Cambisol	541	31.11	98.05		157.0	187.2	223.6	241.0	27.03		286.9	321.4	341.6		
Cambisol	714	52.44	114.2	143.6	180.1	204.5	248.9	268.7	285.1	306.7	346.5			409.3	
Cambisol	733	5.913	55.51		116.5	135.2	158.3	169.2	192.2		225.9	263.6	285.0		
Cambisol	760	6.485	52.67		96.65	112.4	135.8	150.9	167.7		202.4	236.0	254.6		
Cambisol	785	38.56	107.0	163.8	198.5	232.9	280.8	314.2	336.2	360.7	379.7			446.2	
Cambisol	755	39.74	121.2	166.5	197.7	230.2	282.2	302.7	319.4	344.1	362.7			438.4	

Cambisol	914	12.43	80.26	167.9	198.0	240.1	250.1	293.0	323.1	379.4	400.2
Luvisol	27	11.26	69.69	114.0	163.5	198.2	238.6	256.2	267.3	317.4	393.8
Luvisol	74	8.749	60.11	90.1	105.6	145.2	161.1	182.8	19.50	200.7	213.9
Luvisol	122	10.07	79.98	127.1	162.9	243.3	273.0	302.5	323.8	340.3	420.7
Luvisol	173	27.04	98.66	148.5	171.1	209.4	282.7	310.1	333.4	348.0	388.8
Luvisol	327	32.87	80.37	110.8	132.1	156.6	188.0	215.8	226.2	248.4	253.3
Luvisol	425	31.38	108.4	149.2	184.8	219.6	278.8	311.1	329.5	357.3	390.4
Luvisol	433	28.07	73.34	100.2	115.6	137.4	160.0	187.9	187.4	210.7	219.9
Luvisol	554	38.23	85.15	118.9	134.6	169.1	202.1	242.2	269.7	274.4	298.3
Luvisol	633	23.85	66.49	95.17	109.4	130.6	151.5	176.6	193.9	206.7	212.5
Phaeozem	601	43.86	143.4	203.7	253.7	313.4	392.9	418.0	437.3	467.3	503.3
Phaeozem	691	53.84	150.9	211.5	262.0	302.4	364.7	394.1	418.0	455.6	485.0
Phaeozem	794	62.87	129.1	168.9	206.4	260.7	311.3	327.5	335.5	366.1	400.2
											279.6
											275.8
											599.1
											550.1
											475.1

Table 11. Emission of CO₂ (mg kg⁻¹) during incubation of tested soils at pF 1.5 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)															
		1	3	7	10	14	17	21	28	31	35	38	42	45	49	56	
Cambisol	23	6.003	18.62	140.5	163.5	206.5	232.6	277.7	326.7	388.7						455.2	476.2
Cambisol	34	44.34	117.2	214.7	254.0	297.5	328.0	361.9	391.4	396.7	416.4	428.3	440.7	445.8	460.9	474.7	
Cambisol	116	68.71	120.2	184.3	214.2	242.6	259.0	280.6	298.5	305.1	315.0	319.1	330.3	332.6	345.6	355.3	
Cambisol	197	11.93	66.37	130.2	142.9	171.0	194.7	194.6	226.4	256.8						284.1	290.0
Cambisol	250	7.257	9.018	63.46	84.32	96.99	110.4	118.9	137.6	148.7	158.8	167.2	175.5	183.7	191.8	202.0	
Cambisol	302	26.66	57.71	104.8	124.1	145.0	152.1	167.6	176.6	197.8			206.7		218.3	224.7	
Cambisol	317	34.20	60.32	92.6	104.4	117.0	124.2	133.3	133.5	131.9	131.6	131.3	133.2	137.0	139.3	142.4	
Cambisol	834	22.65	59.85	106.8	125.6	144.4	152.7	172.6	180.5	204.5			203.3		227.6	221.7	
Cambisol	528	41.14	96.30	158.4	183.1	208.0	223.6	231.2		251.9	253.3	274.3	273.0	284.8	288.7	299.9	
Cambisol	541	14.94	65.68	145.4	174.4	204.4	216.0	237.9	263.2	287.0			305.8		323.1	335.4	
Cambisol	714	59.47	126.3	191.0	208.9	236.0	256.7	279.0	321.9	356.5					376.2	365.0	
Cambisol	733	6.601	19.66	101.1	117.0	136.8	154.4	171.7	205.6	223.0	238.1	247.7	256.0	270.5	277.3	290.3	
Cambisol	760	7.068	14.91	82.5	100.8	122.1	138.0	152.3	177.9	192.4	207.9	218.5	226.8	235.2	244.4	269.4	
Cambisol	785	6.236	60.65	172.1	223.5	253.2	271.0	298.2	337.5	370.0			393.9		435.3	433.1	
Cambisol	755	7.054	72.17	185.1	208.0	244.6	269.2	297.7	341.5	379.4					405.2	398.7	

Cambisol	914	3.808	30.60	126.8	164.2	185.8	213.7	232.2	267.5	302.5	344.5	367.2	381.4
Luvisol	27	8.138	54.32	140.9	175.6	200.8	226.0	244.1	295.6	312.5	329.7	358.6	398.0
Luvisol	74	8.329	47.21	99.69	116.2	132.7	146.4	155.7	174.0	178.9	191.9	197.1	208.4
Luvisol	122	4.116	6.986	118.8	175.8	219.4	231.9	250.8	286.5	327.8	346.9	365.1	375.3
Luvisol	173	25.98	87.06	177.1	202.2	237.3	260.3	284.2	320.9	342.3	381.4	411.3	450.2
Luvisol	327	25.79	84.19	149.5	162.1	193.6	209.7	226.2	257.0	288.5		320.2	339.2
Luvisol	425	9.539	33.87	133.2	172.3	212.9	229.9	274.3	308.8	345.1	357.7	387.4	399.9
Luvisol	433	23.43	83.41	131.1	143.6	166.8	180.7	199.2	224.5	249.6		275.4	271.3
Luvisol	554	46.89	122.1	188.8	202.7	235.2	255.4	286.2	314.8	346.7		363.4	369.6
Luvisol	633	14.09	64.94	133.0	140.8	172.2	184.3	208.6	227.7	250.6		275.1	269.0
Phaeozem	601	11.68	71.12	215.4	266.2	314.2	333.6	375.0	393.5	434.6	458.5	478.7	486.4
Phaeozem	691	13.02	81.30	219.0	273.6	303.4	339.3	363.9	362.8	407.3	409.0	415.4	435.2
Phaeozem	794	59.91	141.6	225.0	265.5	303.8	298.5	324.8	340.4	362.8	388.9	404.0	420.6

Table 12. Emission of CO₂ (mg kg⁻¹) during incubation of tested soils amended with C and N substrates at pF 0 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)						
		1	3	5	7	10	14	21
Cambisol	23	100.0	390.6	485.7	533.3	566.9	621.6	699.1
Cambisol	34	119.3	335.1	442.0	533.2	587.2	629.4	728.2
Cambisol	116	89.78	263.7	324.1	353.4	399.2	437.6	546.3
Cambisol	197	104.9	328.7	381.8	422.4	455.8	486.2	510.9
Cambisol	250	62.19	291.6	414.7	447.1	466.0	494.8	551.7
Cambisol	302	107.6	226.6	260.4	296.1	323.5	360.8	410.7
Cambisol	317	67.31	249.3	305.5	333.0	379.9	397.3	438.1
Cambisol	834	86.38	232.8	274.5	316.5	354.7	396.1	452.9
Cambisol	528	105.4	285.4	356.3	397.7	445.7	461.8	526.1
Cambisol	541	108.4	311.1	378.6	439.8	487.5	521.9	596.2
Cambisol	714	96.00	256.6	309.5	349.7	387.3	435.9	495.6
Cambisol	733	88.23	329.6	442.1	524.6	576.0	624.9	675.1
Cambisol	760	90.46	336.5	455.3	506.7	570.2	569.3	609.5
Cambisol	785	152.9	375.3	451.9	518.7	573.9	591.8	655.0
Cambisol	755	116.4	343.8	422.0	470.0	505.6	572.1	636.3
Cambisol	914	134.1	418.8	527.0	600.6	656.9	666.3	751.5
Luvisol	27	62.25	354.8	467.2	525.6	575.4	589.7	664.1
Luvisol	74	49.01	266.1	345.2	384.9	396.8	418.8	491.4
Luvisol	122	76.75	344.7	481.7	542.1	619.2	644.8	733.1
Luvisol	173	97.09	363.4	452.2	502.9	548.6	564.2	665.3
Luvisol	327	102.8	278.5	336.9	360.5	381.3	416.0	444.8
Luvisol	425	89.98	333.6	446.8	514.3	556.5	622.5	699.9
Luvisol	433	70.15	255.6	330.8	371.3	405.3	441.8	483.9
Luvisol	554	82.43	262.0	315.6	354.7	391.2	429.3	485.8
Luvisol	633	96.85	280.9	354.0	382.5	427.9	449.3	486.6
Phaeozem	601	132.2	412.5	500.5	571.8	647.9	702.1	786.9
Phaeozem	691	152.0	363.5	452.7	551.7	622.0	659.6	755.9
Phaeozem	794	167.5	308.9	360.4	417.5	483.1	522.5	618.9

Table 13. Emission of CO₂ (mg kg⁻¹) during incubation of tested soils amended with C and N substrates at pF 1.5 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)						
		1	3	5	7	10	14	21
Cambisol	116	102.0	335.3	390.8	428.8	454.0	471.0	537.9
Cambisol	302	80.62	294.4	332.7	370.0	380.9	405.4	455.0
Cambisol	317	49.40	282.2	332.4	362.2	377.8	407.6	465.6
Cambisol	713	122.9	343.3	406.1	445.4	453.1	486.4	545.9
Cambisol	733	21.59	168.1	337.4	404.4	458.3	492.1	554.3
Luvisol	27	14.26	163.2	344.2	429.4	479.8	520.7	596.4
Luvisol	122	24.94	236.5	401.9	476.2	530.9	564.6	635.4
Luvisol	554	84.24	333.3	390.9	419.7	447.6	494.7	552.2
Phaeozem	691	13.10	224.5	301.1	331.7	337.8	363.7	396.8
Phaeozem	794	154.3	377.7	447.2	472.2	489.0	565.4	623.0

In general, respiration was the greatest in Phaeozems (on average 262 mg CO₂-C kg⁻¹). The Cambisols and Luvisols presented respiration activity 30% lower (on average about 165 mg CO₂-C kg⁻¹) (Fig. 23). The soil unit did not influence significantly the uptake of O₂ which was about 17% v/v.

Cambisol and Phaeozem developed from loess, amended with CH₄, showed different levels of respiration activity during 11-day incubation (Brzezińska *et al.* 2004). Cambisol evolved 320 mg CO₂-C kg⁻¹ and consumed 8% vol. O₂, whereas Phaeozem evolved 403 mg CO₂-C kg⁻¹ and utilized 11% O₂. It means that O₂ consumption and CO₂ production were, on average, 85 to 30% higher, respectively, in CH₄ amended soil as compared to the control.

Daily O₂ consumption by Gleysol and Podzol, found by Włodarczyk *et al.* (2004a), is shown in Figure 24, and the rates of CO₂ production and O₂ consumption are shown on Table 18.

The greatest CO₂ production and O₂ consumption were observed at pH 5.5-7.5. Soil respiration was high at Eh values of 500-600 mV. Moreover, decrease of Eh below 300 mV induced higher CO₂ evolution and O₂ consumption.

The influence of temperature in the range of 4-20°C on the rate of respiration of Cambisol and Phaeozem (Włodarczyk *et al.* 2001) is shown in Figure 25. It increased in both soils with temperature increase, from about 6 CO₂-C kg⁻¹ at 4°C to about 60 CO₂-C kg⁻¹ at 20°C.

Table 14. Changes of O₂ concentration (% v/v) during incubation of tested soils at pF 0 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation time (days)													
		1	3	5	7	10	14	18	21	24	28	35	42	49	56
Cambisol	23	20.93	20.12	19.18	18.84	18.04	17.71	16.91	16.42	15.60	14.93				
Cambisol	34	20.66	19.29	17.88	16.97	15.91	15.21	13.89	12.78	11.82	11.16				
Cambisol	116	20.28	19.38	18.25	17.56	16.65	16.22	15.51	14.97	14.26	13.72				
Cambisol	197	20.76	19.94	19.48	18.95	18.80	18.49	18.07	17.82	17.63	17.36	16.43	15.73	15.13	15.01
Cambisol	250	20.98	20.56	19.99	19.88	19.58	19.62	19.12	18.81	18.20	17.80				
Cambisol	302	20.58	20.08	19.70	19.35	18.67	18.17	17.75	17.65	17.21	17.06			15.70	
Cambisol	317	20.85	20.38	19.80	19.54	19.13	18.94	18.82	18.02	17.60	17.33				
Cambisol	834	20.61	20.21	19.94	19.67	19.14	18.76	18.23	17.83	17.58	17.33			16.46	
Cambisol	528	20.74	19.82	18.89	18.34	17.68	17.28	16.65	16.19	15.46	15.00				
Cambisol	541	20.70	19.95	19.18	18.67	17.87	17.67	17.06	16.59	15.80	15.09				
Cambisol	714	20.23	19.29	18.48	17.95	17.41	16.50	16.13	15.65	15.29	14.75			12.33	
Cambisol	733	20.87	20.36	19.69	19.38	18.86	18.75	18.27	17.81	17.44	16.78				
Cambisol	760	20.87	20.41	19.83	19.57	19.27	19.06	18.57	18.17	17.55	17.24				
Cambisol	785	20.67	19.81	19.05	18.49	17.93	17.22	16.34	16.29	15.85	15.46			13.92	
Cambisol	755	20.59	19.70	19.08	18.64	17.74	17.33	16.66	16.44	15.53	15.93			14.10	

Cambisol	914	20.85	20.02	18.94	18.50	17.69	17.48	16.79	16.16	15.28	14.67				
Luvisol	27	20.88	20.16	19.44	18.64	18.09	16.48	17.26	16.75	16.01	15.59	14.96	14.34	14.07	
Luvisol	74	20.92	20.20	19.86	19.42	18.83	18.50	18.16	17.94	17.72	17.23	16.63	16.44	15.88	
Luvisol	122	20.98	20.16	19.54	18.80	17.83	17.39	16.94	16.62	16.23			14.77		
Luvisol	173	20.73	19.90	19.15	18.79	17.85	17.66	16.98	16.65	16.20	15.92	14.97	13.79	12.81	12.21
Luvisol	327	20.60	20.01	19.60	19.51	18.44	18.31	17.78	17.56	17.27	17.08	16.26	15.78	15.09	14.58
Luvisol	425	20.81	19.99	19.30	18.86	18.19	17.39	16.45	16.48	16.01	15.46			14.01	
Luvisol	433	20.58	19.87	19.46	19.19	18.87	18.45	17.98	17.76	17.50	17.24	16.64	15.98	15.47	14.84
Luvisol	554	20.64	19.62	18.72	18.44	17.82	17.22	16.41	15.99	15.32	14.89	14.14	13.48	13.07	12.86
Luvisol	633	20.73	20.15	19.80	19.63	18.76	18.81	18.40	18.16	17.90	17.81	17.20	16.66	16.34	15.91
Phaeozem	601	20.70	19.61	18.64	18.86	16.93	15.70	15.04	14.61	14.04	13.37			11.30	
Phaeozem	691	20.50	19.38	18.40	17.82	16.66	15.71	15.05	14.42	13.65	13.81			10.28	
Phaeozem	794	20.18	19.10	18.30	17.57	16.38	15.50	14.90	14.37	13.75	13.38			11.06	

Table 15. Changes of O₂ concentration (% v/v) during incubation of tested soils at pF 1.5 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)															
		1	3	7	10	14	17	21	23	28	31	35	38	42	45	49	56
Cambisol	23	20.52	20.36	18.88	18.60	18.02	17.06	17.02	16.96	16.18		15.26		14.68	13.92		13.82
Camhisol	34	20.30	19.33	18.33	17.31	16.61	15.91	14.97	14.51	13.74	13.64	13.40	1.32	12.90	12.77	12.59	12.24
Camhisol	116	19.77	18.95	17.96	17.39	16.78	16.39	15.97	15.86	15.42	15.47	15.29	15.14	14.90	14.86	14.60	14.36
Cambisol	197	20.43	19.82	18.87	18.65	18.28	18.18	17.90	17.49	17.39		16.74		15.72	15.84		15.70
Cambisol	250	20.65	20.65	20.03	19.78	19.51	19.34	19.20	19.09	18.96	18.83	18.68	18.55	18.42	18.32	18.23	17.91
Canibisot	302	20.53	20.10	19.44	19.09	18.52	18.30	17.81	17.88	17.67		17.54		17.22	17.18		17.01
Cambisol	317	20.44	20.12	19.76	19.61	19.37	19.30	19.21	19.07	19.02	18.97	19.01	18.96	18.82	18.70	18.81	18.54
Cambisol	834	20.62	20.02	19.53	19.24	18.45	18.65	18.20	18.07	17.61		17.32		17.29	17.20		16.97
Cambisol	528	20.30	19.51	18.75	18.41	17.96	17.74	17.21	17.31	17.03	16.97	16.72	16.41	16.03	15.87	15.28	15.34
Cambisol	541	20.77	20.03	19.12	18.74	18.31	17.95	17.59	17.59	17.22		16.77		16.20	15.45		14.87
Cambisol	714	19.73	18.74	17.78	17.38	17.22	16.73	16.37	15.91	15.55		14.49		13.99	13.70		13.71
Cambisol	733	20.68	20.49	19.57	19.35	19.08	18.86	18.56	18.37	18.11	17.90	17.58	17.53	17.32	17.09	17.09	16.59
Cambisol	760	20.71	20.57	19.74	19.55	19.30	19.07	18.78	18.62	18.30	18.25	18.00	17.88	17.60	17.51	17.29	16.91
Cambisol	785	20.79	20.09	18.71	18.02	17.64	17.35	16.55	16.77	16.44		15.79		15.24	14.62		14.12
Cambisol	755	20.53	19.68	18.29	17.98	17.44	17.05	16.63	16.22	15.87		15.36		15.00	14.55		14.20

Cambisol	914	20.80	20.45	19.26	18.71	18.18	18.06	17.67	17.50	17.23	16.64	15.98	15.64	15.35			
Luvisol	27	20.70	20.08	19.04	18.59	18.19	17.85	17.26	17.11	16.62	16.53	16.26	15.99	15.66	14.95	14.31	
Luvisol	74	20.71	20.13	19.54	19.29	19.02	18.85	18.46	18.35	18.18	18.12	17.89	17.74	17.42	17.39	17.26	17.09
Luvisol	122	20.85	20.75	19.39	18.55	18.08	17.76	17.55	17.36	16.91	16.46	16.06	15.85	15.62			
Luvisol	173	20.53	19.69	18.69	18.30	17.85	17.57	17.11	16.79	16.37	16.15	15.79	15.50	14.96	14.70	14.17	13.35
Luvisol	327	20.33	19.56	18.81	18.12	18.05	17.95	17.62	17.22	16.94	16.27	15.45	15.01	14.70			
Luvisol	425	20.83	20.46	19.10	18.75	18.18	17.88	17.34	17.05	16.64	16.20	15.80	15.49	15.21			
Luvisol	433	20.29	19.51	18.94	18.76	18.39	18.24	17.85	17.93	17.27	16.59	16.31	15.82	15.78			
Luvisol	554	19.93	18.88	17.80	17.54	16.97	16.68	16.01	15.30	15.01	14.36	14.23	13.85	13.78			
Luvisol	633	20.39	19.77	18.99	18.82	18.40	18.14	17.95	17.68	17.47	17.12	16.85	16.61	16.30			
Phaeozem	601	20.55	19.96	18.20	17.51	16.80	16.36	15.87	15.75	15.37	14.75	14.15	13.84	13.29			
Phaeozem	691	20.72	19.75	17.95	17.33	16.59	16.22	15.69	15.56	15.37	14.75	14.14	13.73	13.37			
Phaeozem	794	19.98	18.81	17.53	16.61	15.72	15.51	15.09	15.65	15.13	14.42	14.05	13.76	13.34			

Table 16. Changes of O₂ concentration (% v/v) during incubation of tested soils amended with C and N substrates at pF 0 (Gliński *et al.* 2010)

Soils	No.	Incubation (days)						
		1	3	5	7	10	14	21
Cambisol	23	20.24	17.28	15.92	15.01	14.16	13.15	12.65
Cambisol	34	19.83	17.37	15.86	14.46	13.16	12.41	9.60
Cambisol	116	19.52	16.84	15.55	14.68	13.77	12.92	10.87
Cambisol	197	19.96	17.07	16.21	15.35	14.86	14.19	13.27
Cambisol	250	20.42	18.45	16.56	16.16	15.36	15.07	14.08
Cambisol	302	19.65	18.07	17.18	16.93	16.32	15.58	14.37
Cambisol	317	20.09	17.95	16.85	16.19	15.74	15.43	14.41
Cambisol	350	19.86	17.79	16.75	16.16	15.40	15.05	13.67
Cambisol	528	19.85	17.52	16.27	15.60	14.81	14.48	12.88
Cambisol	541	19.96	17.53	16.32	15.51	14.50	13.92	12.27
Cambisol	713	19.60	16.99	15.79	14.91	13.95	13.09	11.58
Cambisol	733	20.42	18.00	16.52	15.58	14.63	14.03	12.67
Cambisol	760	20.34	17.92	15.74	14.83	13.86	13.77	12.71
Cambisol	785	19.72	17.84	16.14	15.14	14.22	13.70	12.23
Cambisol	795	19.88	17.39	16.11	15.21	14.11	13.10	11.42
Cambisol	914	19.93	16.84	14.97	14.05	12.74	12.52	10.30
Luvisol	27	20.43	17.51	15.87	14.74	13.67	13.09	11.53
Luvisol	74	20.31	17.87	16.63	15.90	15.39	14.74	13.63
Luvisol	122	20.31	17.96	15.99	14.98	13.76	12.67	11.21
Luvisol	173	20.00	17.74	15.78	14.28	14.04	13.06	11.66
Luvisol	327	19.99	17.57	16.92	16.04	15.53	14.97	13.89
Luvisol	425	20.16	17.69	15.89	14.92	13.87	13.14	10.95
Luvisol	433	19.98	17.51	16.29	15.50	14.69	14.14	13.12
Luvisol	554	19.65	16.57	15.72	14.95	14.07	13.21	12.02
Luvisol	633	19.97	17.66	16.48	15.74	15.01	14.47	13.43
Phaeozem	689	19.96	17.27	15.66	14.55	13.22	12.34	10.00
Phaeozem	691	19.53	16.98	15.49	14.33	12.84	12.19	9.82
Phaeozem	794	18.90	16.78	15.66	14.64	13.43	12.76	9.98

Table 17. Changes of O₂ concentration (% v/v) during incubation of tested soils amended with C and N substrates at pF 1.5 (Gliński *et al.* 2010)

Soil type	Soil No.	Incubation (days)						
		1	3	5	7	10	14	21
Cambisol	116	19.19	15.86	15.04	14.58	13.97	13.06	11.95
Cambisol	302	19.30	16.64	16.03	15.66	15.13	14.55	13.94
Cambisol	317	19.96	17.23	16.61	16.21	15.71	14.84	14.42
Cambisol	713	18.92	15.74	14.92	14.34	–	12.94	12.35
Cambisol	733	20.19	18.63	16.82	15.43	–	14.15	13.67
Luvisol	27	20.26	18.56	16.35	15.28	14.28	14.18	12.65
Luvisol	122	20.23	17.68	15.63	14.66	13.46	13.28	12.31
Luvisol	554	19.36	15.92	15.17	14.65	13.95	13.12	12.12
Phaeozem	691	20.31	17.33	16.20	15.71	–	14.60	14.50
Phaeozem	794	18.49	15.25	14.33	13.62	–	11.26	10.45

Table 18. The rate of O₂ consumption and CO₂ production depending on the CH₄ addition to Gleysol and Podzol (Włodarczyk *et al.* 2004a)

Soil	CH ₄ amendment (%)	CH ₄	O ₂	CO ₂	CH ₄ : O ₂ : CO ₂
		(ml kg ⁻¹ d ⁻¹)			
Gleysol	0	0	18.8	11.4	–
	1.25	23.0	39.1	20.4	1 : 0.88 : 0.39
	2.5	45.2	50.4	29.5	1 : 0.70 : 0.40
	5	84.7	78.0	46.2	1 : 0.70 : 0.40
	10	163.2	139.2	78.1	1 : 0.74 : 0.41
	15	124.8	171.5	87.7	1 : 1.22 : 0.61
Podzol	0	0	12.2	10.6	–
	1.25	1.6	13.9	11.5	1 : 1.06 : 0.58
	2.5	2.9	15.7	12.0	1 : 1.21 : 0.50
	5	10.3	26.2	18.8	1 : 1.36 : 0.79
	10	19.1	38.7	25.4	1 : 1.39 : 0.78
	15	39.0	62.4	38.6	1 : 1.29 : 0.72

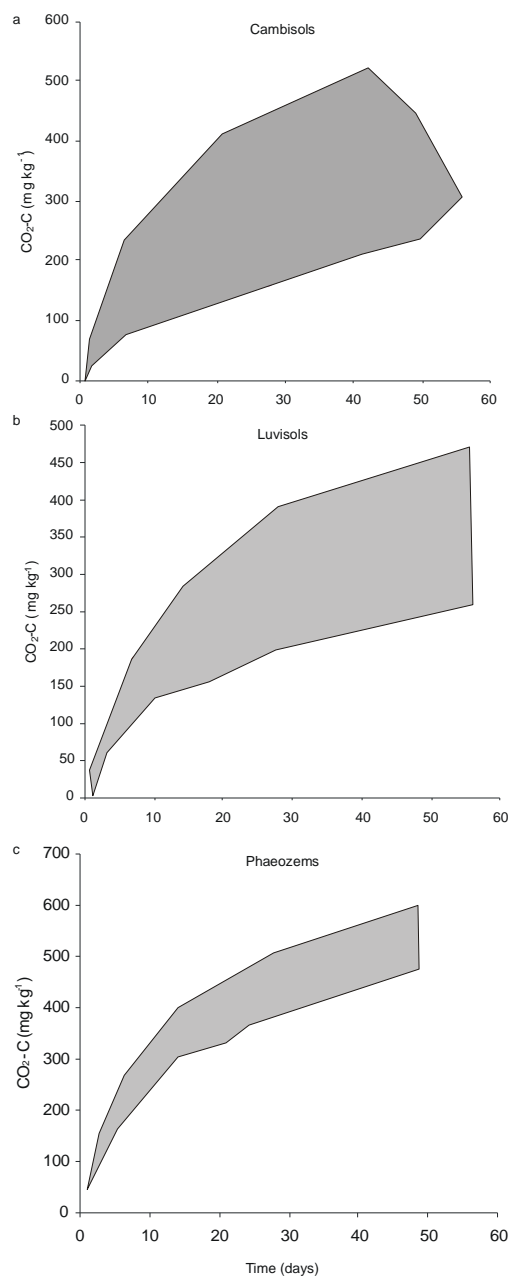


Fig. 13. Areas of cumulative curves of the emission of CO₂ from the Cambisols, Luvisols and Phaeozems incubated at pF 0. Note different scales on the graphs (Gliński *et al.* 2010)

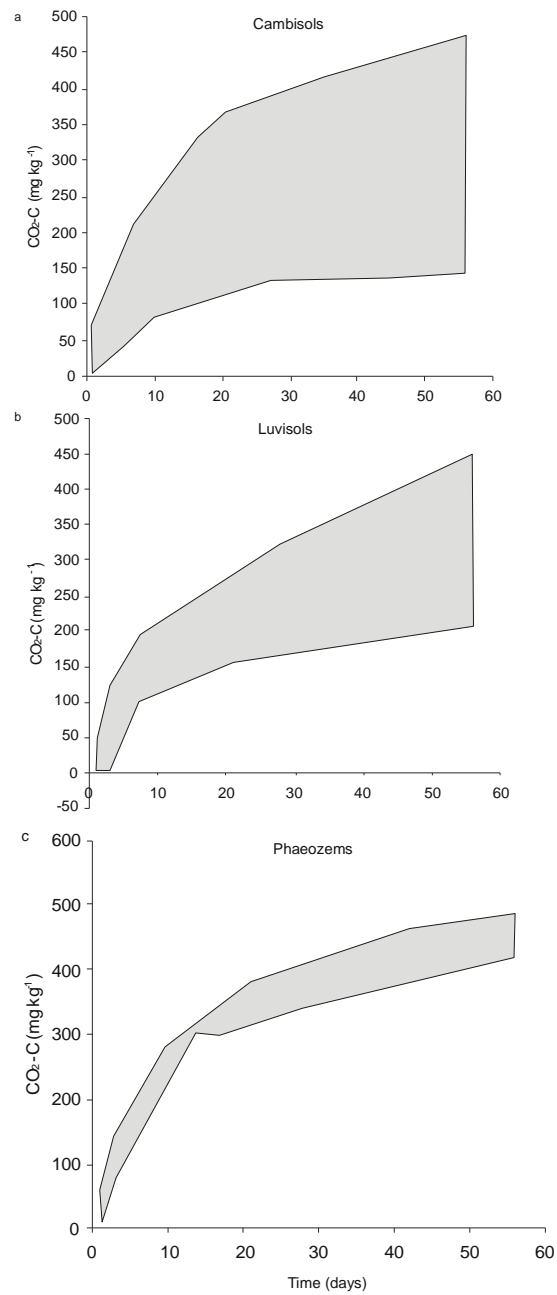


Fig. 14. Areas of cumulative curves of the emission of CO₂ from the Cambisols, Luvisols and Phaeozems incubated at pF 1.5. Note different scales on the graphs (Gliński *et al.* 2010)

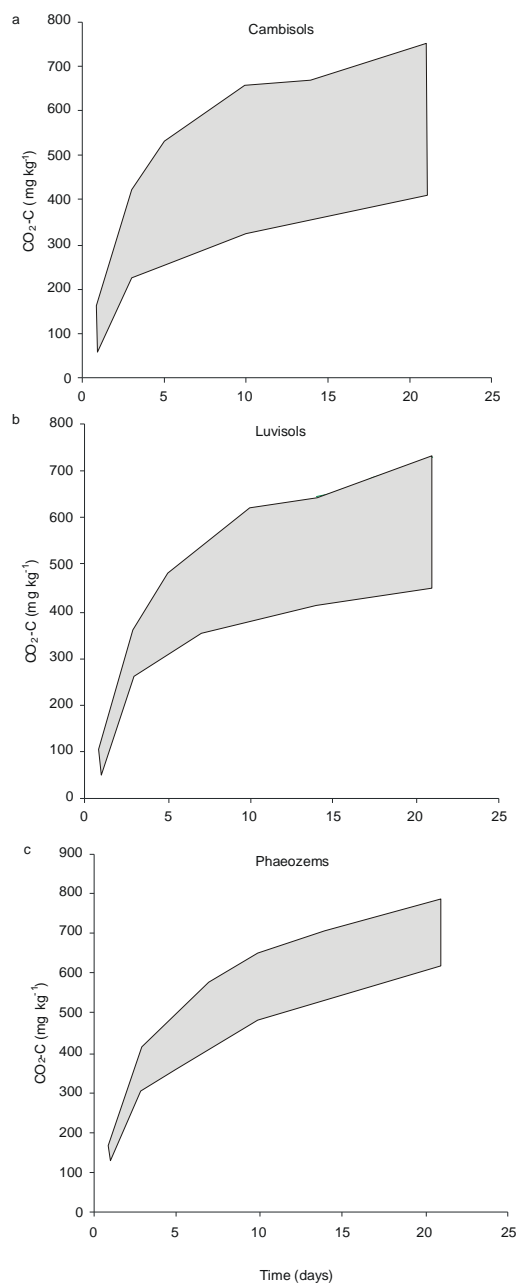


Fig. 15. Areas of cumulative curves of the emission of CO₂ from the Cambisols, Luvisols and Phaeozems incubated with addition of C and N substrate at pF 0. Note different scales on the graphs (Gliński *et al.* 2010)

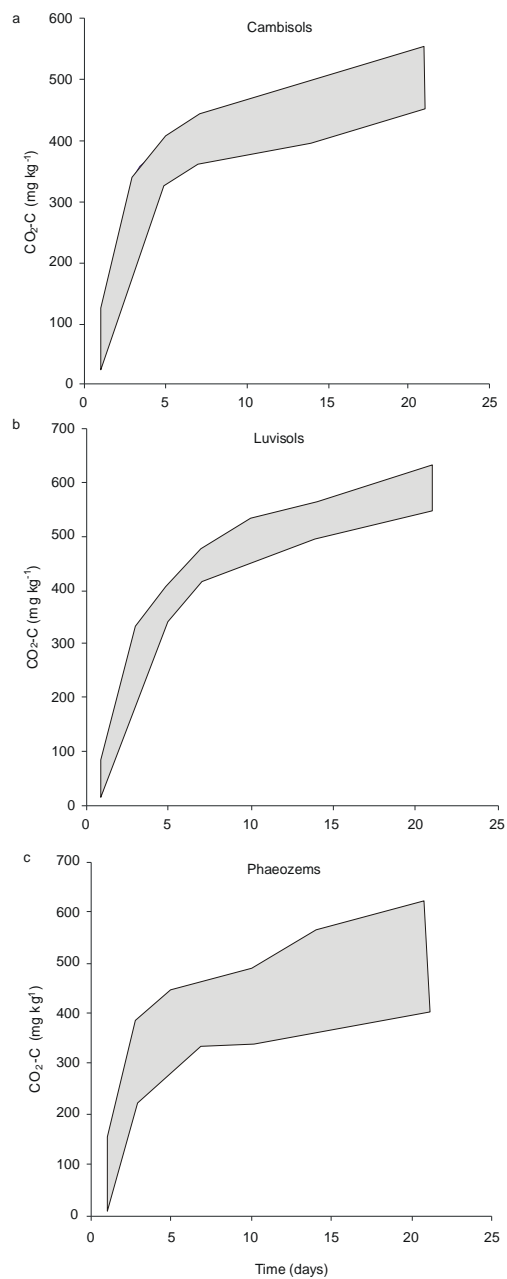


Fig. 16. Areas of cumulative curves of the emission of CO₂ from the Cambisols, Luvisols and Phaeozems incubated with addition of C and N substrate at pF 1.5. Note different scales on the graphs (Gliński *et al.* 2010)

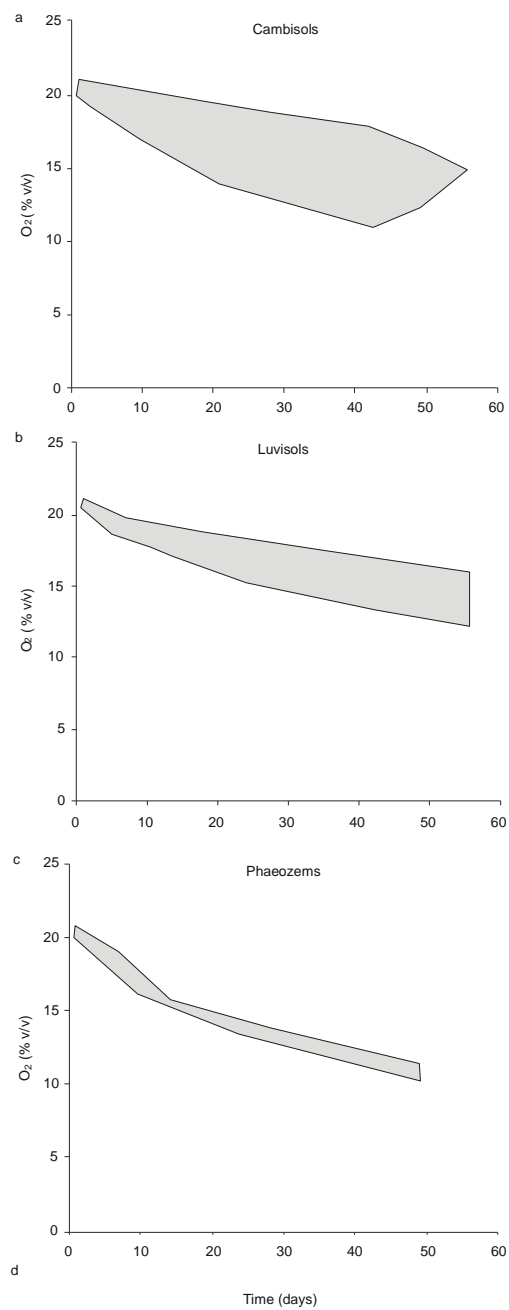


Fig. 17. Areas of changes of O_2 concentration in the headspace of the Cambisols, Luvisols and Phaeozems incubated at pF 0 (Gliński *et al.* 2010)

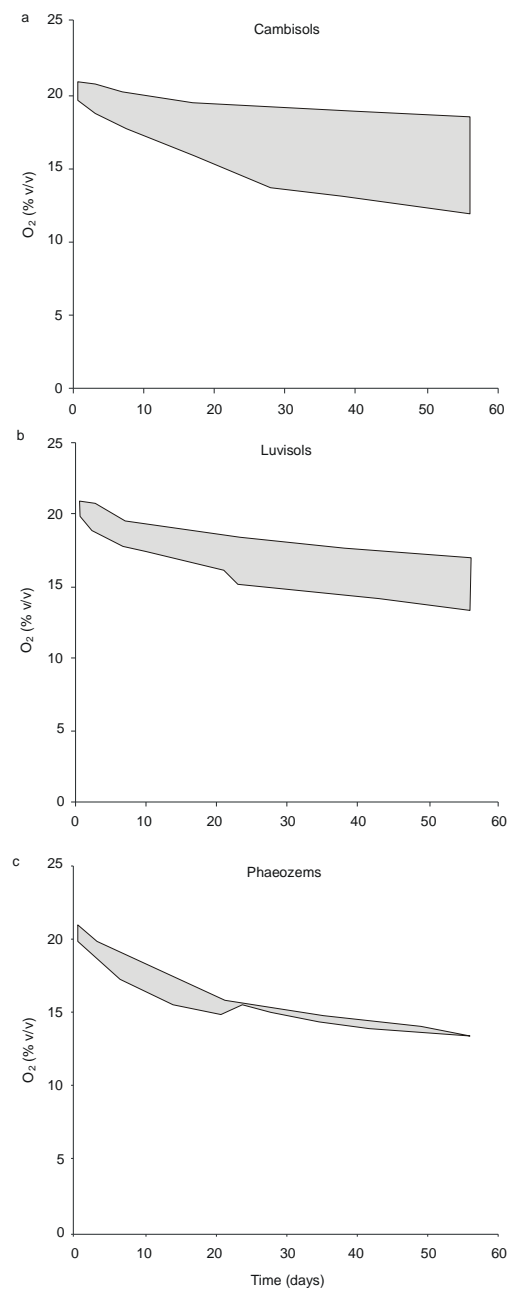


Fig. 18. Areas of changes of O₂ concentration in the headspace of the Cambisols, Luvisols and Phaeozems incubated at pF 1.5 (Gliński *et al.* 2010)

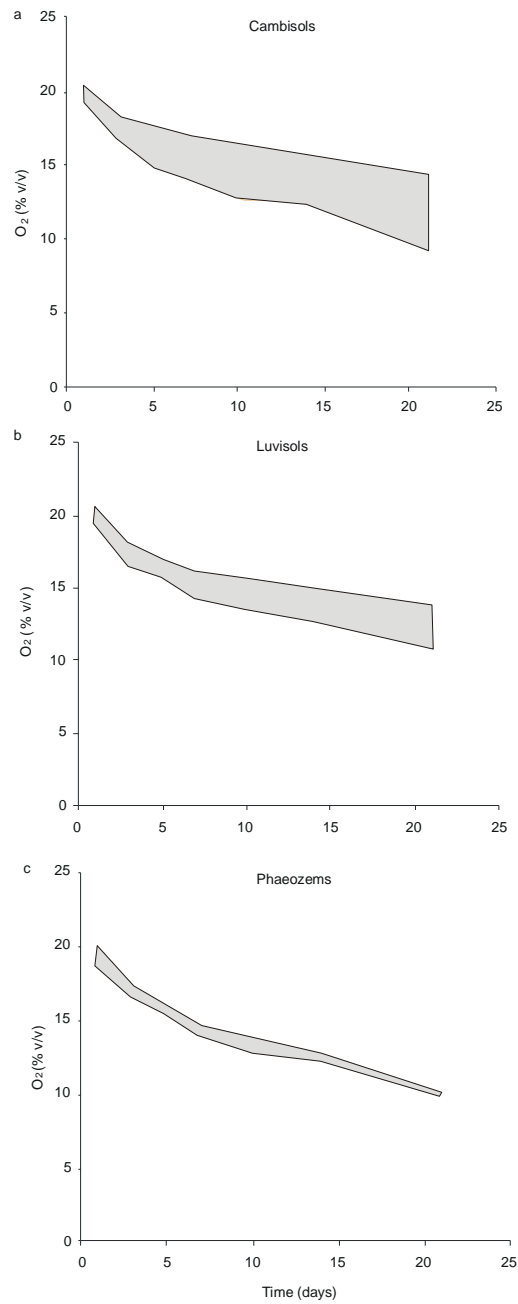


Fig. 19. Areas of changes of O₂ concentration in the headspace of the Cambisols, Luvisols and Phaeozems incubated with addition of C and N substrate at pF 0 (Gliński et al. 2010)

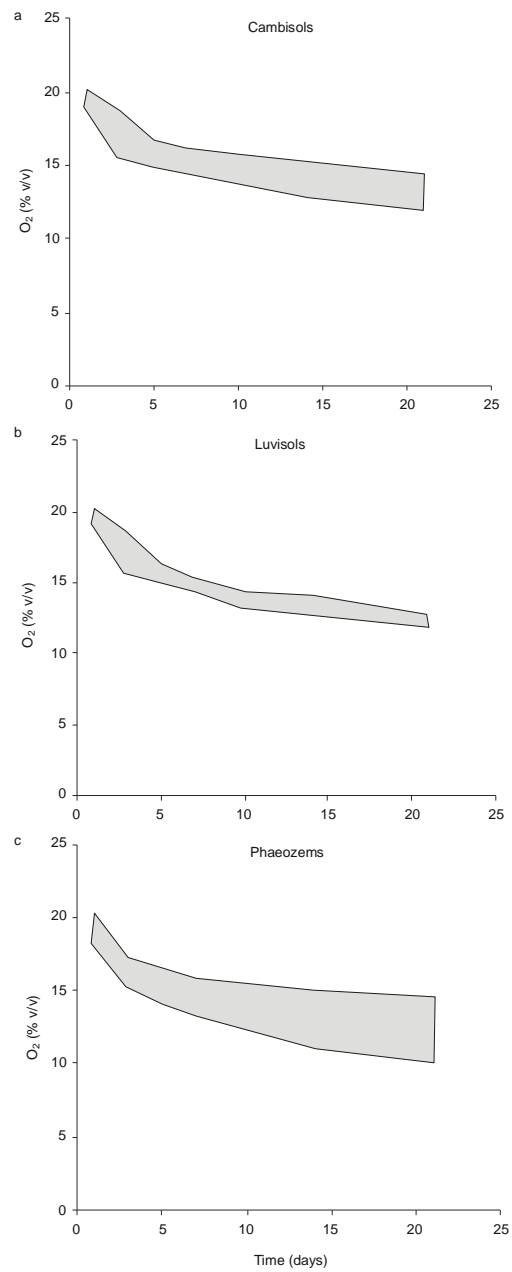


Fig. 20. Areas of changes of O₂ concentration in the headspace of the Cambisols, Luvisols and Phaeozems incubated with addition of C and N substrate at pF 1.5. Note different scales on the graphs (Gliński *et al.* 2010)

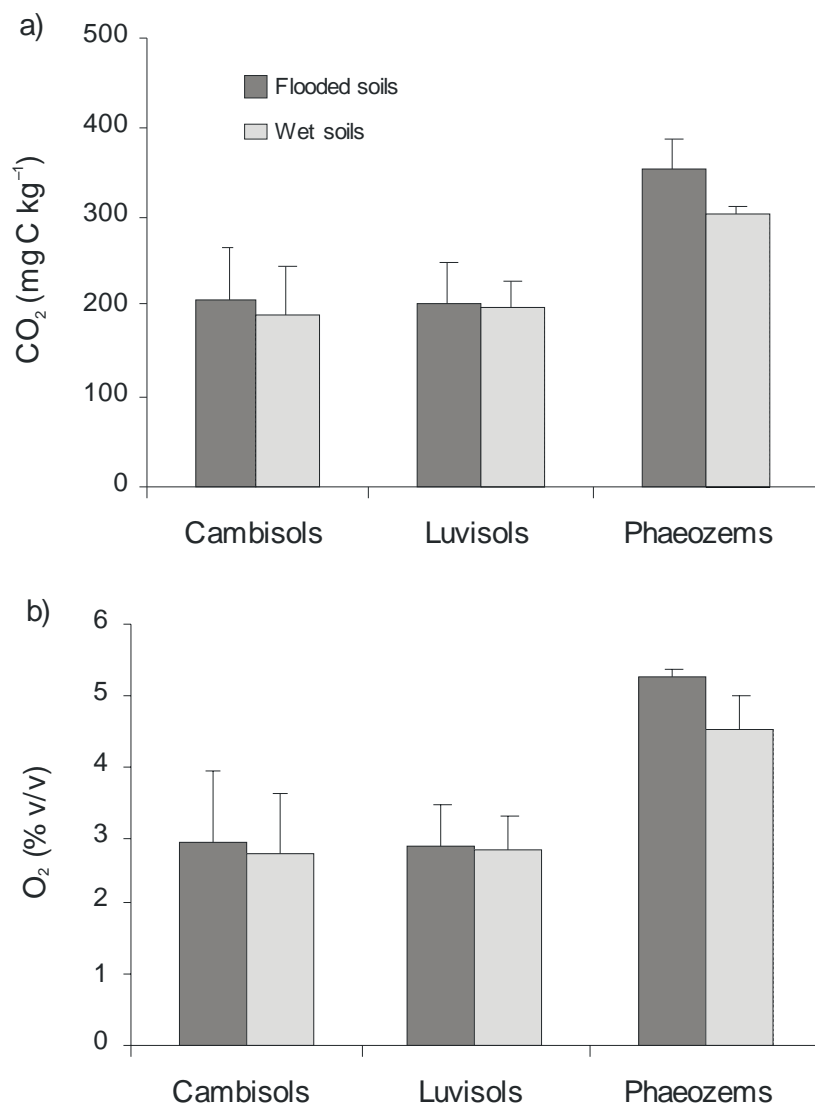


Fig. 21. Respiration activity of 28 (see Table 9) mineral soils of Poland (Cambisols, Luvisols and Phaeozems (average value with standard deviation); a) carbon dioxide produced, and b) O_2 consumed during 14-day incubation of wet or flooded soil samples (pF 1.5 or pF 0, respectively) (Gliński *et al.* 2010)

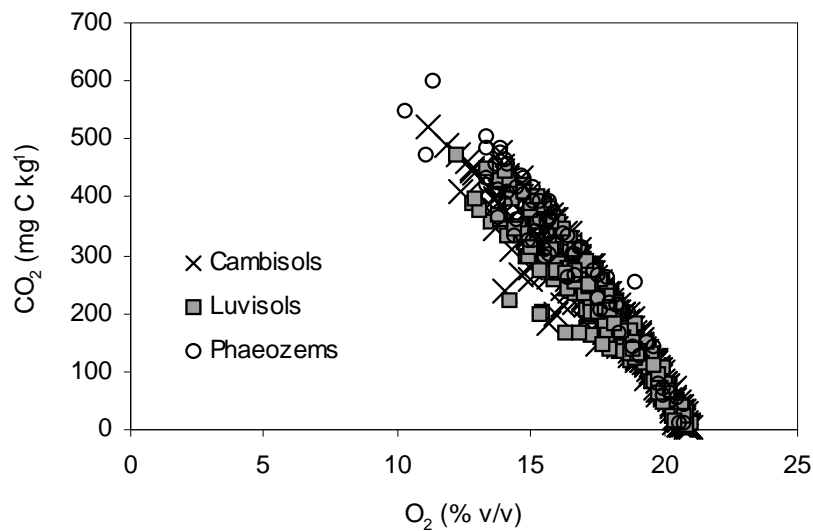


Fig. 22. Cumulative CO₂ vs. O₂ concentration in the headspace for 28 (see Table 9) mineral soils (Cambisols, Luvisols and Phaeozems) incubated at pF 1.5 or under flooding. All measurements over 60-day incubation were included. Points of ambient O₂ in the headspace (app. 21% v/v) and very low CO₂ correspond to the start of the experiment. For linear correlation of all results ($n = 716$), $r = 0.96$, $p < 0.001$) (Gliński *et al.* 2010)

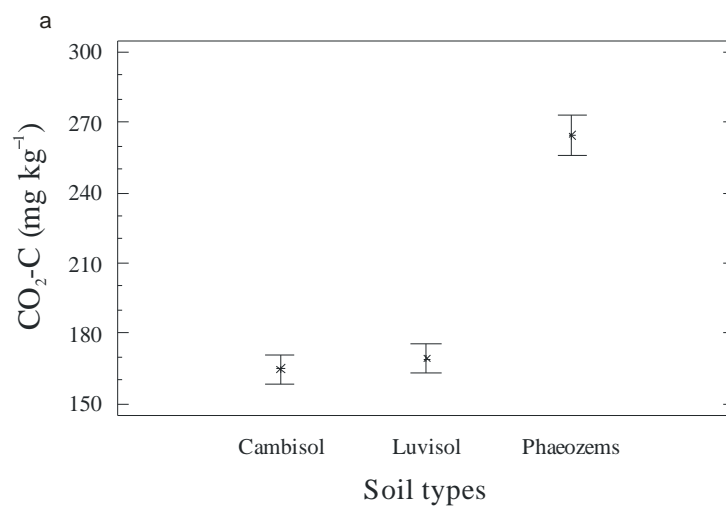


Fig. 23. CO₂ emission from 28 (see Table 9) tested soils: a) average values for Cambisols, Luvisols and Phaeozems; b) average changes with time (Gliński *et al.* 2010)

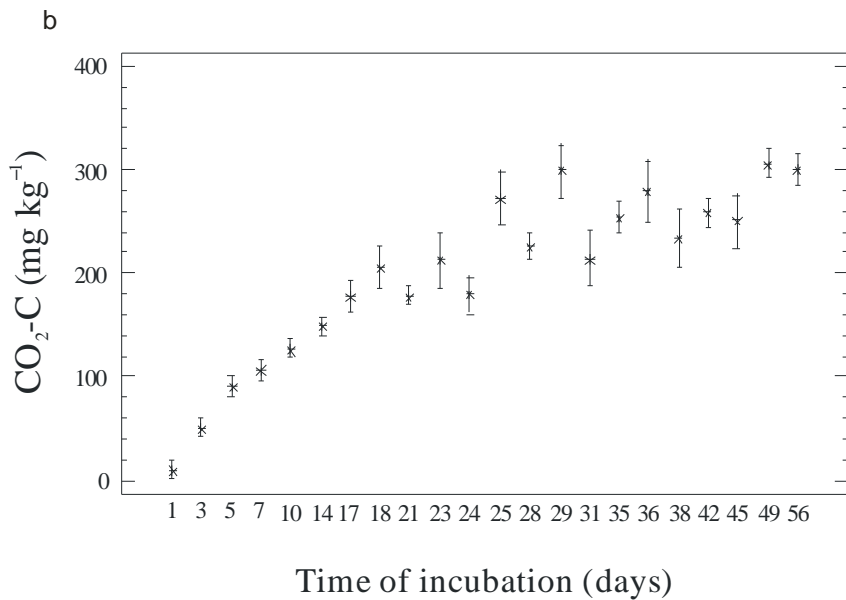


Fig. 23. Cont. CO₂ emission from 28 (see Table 9) tested soils: a) average values for Cambisols, Luvisols and Phaeozems; b) average changes with time (Gliński *et al.* 2010)

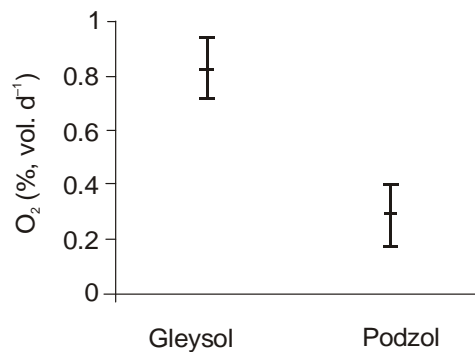


Fig. 24. Daily O₂ consumption by Gleysol and Podzol. The bars represent 95% LSD confidence intervals (Włodarczyk *et al.* 2004a)

Recently, a study was conducted on soils from the Database of Polish Arable Mineral Soils of the Institute of Agrophysics PAS in Lublin, stored for many years and newly downloaded, on ‘Changes in denitrification capacity of selected mineral soils in relation to changes in their content of organic carbon and nitrogen’, sponsored by the Ministry of Science, from which it follows that storage time of soils under drought conditions has a great impact on the activity of aerobic and

anaerobic respiration and denitrification capacity (Włodarczyk, Project Report No. N N310 115338, 2013).

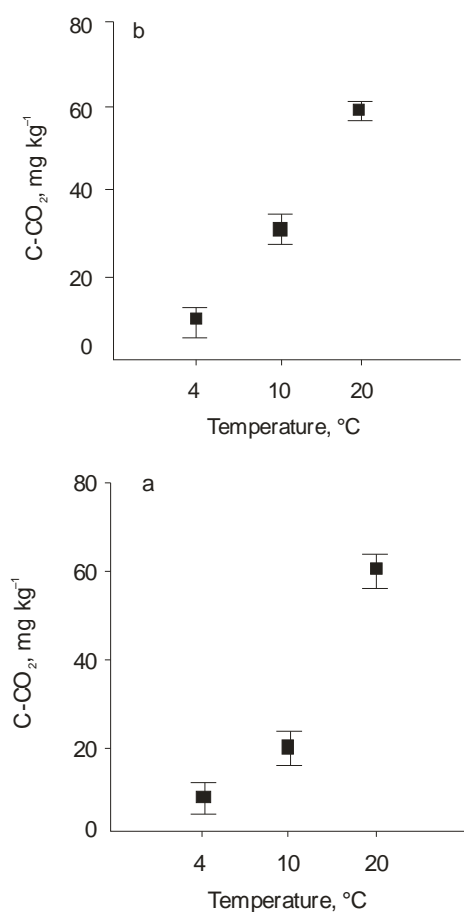


Fig. 25. CO₂-C content in the headspace as a function of temperature in Phaeozems (a) and Cambisol (b) (Włodarczyk *et al.* 2001)

4.3. Dehydrogenase activity

Twenty one Luvisols and Phaeozems developed from loess were tested for their dehydrogenase activity (DHA), redox potential (Eh), oxygen diffusion rate (ODR), and ability of iron reduction to Fe(II). Tested soils were characterized by C_{org} ranges of 0.65-1.04% and 0.83-2.10%, respectively, and showed pH in H₂O in the range from 5.65 to 7.71. The measurements of soil redox properties were preceded by

a 14-day pre-incubation under nine variants being combinations of different temperature and air-water status, namely 10, 20 or 30°C, and soil water tensions of pF 0, 1.7 or 2.2 (corresponding to 0, 5 and 15.9 kPa, respectively) (Brzezińska *et al.* 1998). An increase of temperature by 10°C enhanced the DHA, on average, 2.6 and 4.6 times for pF 2.2 and flooded treatment, respectively. In turn, soil flooding caused on average an 18-fold increase of the dehydrogenase activity with respect to the pF 2.2 treatment. The combined effect of flooding and elevated temperature of 30°C resulted in a strong stimulation of microbial metabolism and, on average, a 129-fold increase of dehydrogenase activity as compared with pF 2.2 at the lowest temperature. The changes in enzyme activity were followed by the reduction of soil system (i.e., a decrease of the Eh and ODR, and accumulation of reduced Fe). The authors reported a significant negative relation for ODR ($r = -0.71$, $p < 0.001$), and a positive one for reduced Fe ($r = 0.90$, $p < 0.001$) (Brzezińska *et al.* 1998).

Fe(III) oxides have previously been shown to be an important electron acceptor of facultative anaerobic bacteria, and intensive reduction of Fe(III) to Fe(II) occurs after oxygen and nitrate depletion in flooded soil (Gliński and Stepniewski 1985). The Eh values in tested soils dropped to a comparable level in both Phaeozems and Luvisols, while maximum Fe(II) which was reached in Phaeozems was by about 20% higher than in Luvisols (Table 19). However, dehydrogenase activity for Phaeozems was >2 times higher than that obtained for Luvisols under identical flooding conditions, at 30°C.

Figure 26 illustrates the relationship between soil dehydrogenase activity and redox potential measured after the pre-incubation of tested Luvisols and Phaeozems over the entire ranges of water tensions and temperature. The results show that soil physical conditions indirectly influenced soil DHA through changes of soil aeration status, expressed here by soil redox potential. A strong curvilinear relationship between DHA and Eh was observed despite the differences in the soils themselves, and in soil physical conditions. The close relation between dehydrogenase activity and redox potential determined for various soils incubated under the same conditions indicates that the major factor determining dehydrogenase activity in soil is its aeration status. The role of the physical factors in determination of DHA is important as far as they alter the redox potential of the soil. Moreover, these results imply that the electron activity of the soil solution (which is reflected by Eh) is more important for the activity of soil dehydrogenases than the direct oxygen availability determined by ODR. The latter indicator, in turn, is known to be a measure of direct availability of O₂ for plant roots (Gliński and Stepniewski 1985).

Table 19. Maximum values of the dehydrogenases activity (DHA) and reduced iron (Fe(II)), and the lowest redox potential (Eh) observed among tested Luvisols and Phaeozems (averages \pm SD). (Brzezińska *et al.* 1998)

Soil type	DHA (mg TPF g ⁻¹ 20h ⁻¹)	Fe(II) (mg kg ⁻¹)	Eh (mV)
Luvisols	1.08 \pm 0.07	1836 \pm 139	-107 \pm 10
Phaeozems	2.31 \pm 0.09	2217 \pm 166	-144 \pm 10

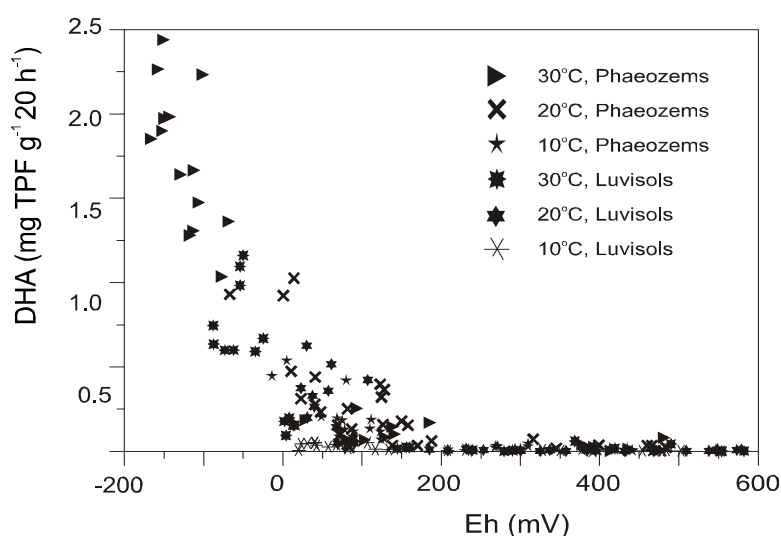


Fig. 26. Relationship between soil dehydrogenases activity (DHA) and redox potential (Eh) in five Luvisols and five Phaeozems measured after 14-day preincubation at 10, 20 and 30°C over the entire range of water tensions (pF 0, 1.7 and 2.2). Correlation coefficient of the whole group of data (multiplicative model, $r = -0.81$, $p < 0.001$, $n = 174$) (Brzezińska *et al.* 1998)

The results of the response of DHA activity to Polish soils reoxidation process are extensively described by Wolińska (2010), Wolińska and Bennicelli (2010), Wolińska and Stepniowska (2011), Stepniowska and Wolińska (2004, 2005). Re-oxidation process in soils is connected with reversion to aerated conditions following their flooding. Changes of such aeration factors as Eg, Eh, and ODR and their fluctuations have an effect on the metabolism of microorganisms and their enzymatic activity. To find the relationships between the microorganism abundance of the soil and the varying aeration parameters during reoxidation processes, based on their indirect effect on the soil oxidation status, Cambidols, Podzols, Histosols, Fluvisols Gleysols, Phaeozems and Leptosols were investigated.

The scheme of one of the experiments concerning three soils (Leptosols, Histosols and Fluvisols) is shown in Figure 27 and the results obtained in Figure 28 and Table 20. DHA was negatively correlated with aeration parameters (pF, ODR, Eg).

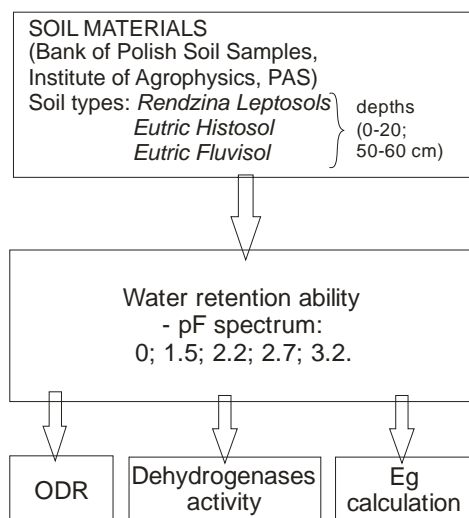


Fig. 27. Scheme of the experiment (Wolińska and Bennicelli, 2010)

Table 20. Statistical significance of differences between DHA and parameters pF, ODR and Eg described by correlation coefficient (R) (95% LSD method, n=15), (Wolińska and Bennicelli 2010)

DHA response	depth (cm)	pF	ODR	Eg
Leptosols	0-20	-0.98***	-0.90**	-0.96**
Histosol	0-20	-0.95***	-0.41*	-0.34*
Fluvisol	0-20	-0.97***	-0.96**	-0.97**

*, **, *** – indicate values significant at the 5, 1 and 0.1% level, respectively.

Heavy metals presence could suppress DHA significantly. Stepniewska and Wolińska (2004, 2005) and Wolińska and Stepniewska (2011) showed that Cd addition to Fluvisol at the concentration of 2 mg kg⁻¹ had a stimulating effect on soil DHA level. However, 10-fold higher Cd amendment (20 mg kg⁻¹) had an inhibiting effect. Trivalent and hexavalent Cr compounds had a noticeable negative effect on soil DHA in Luvisols and Cambisols (Stepniewska and Wolińska 2004).

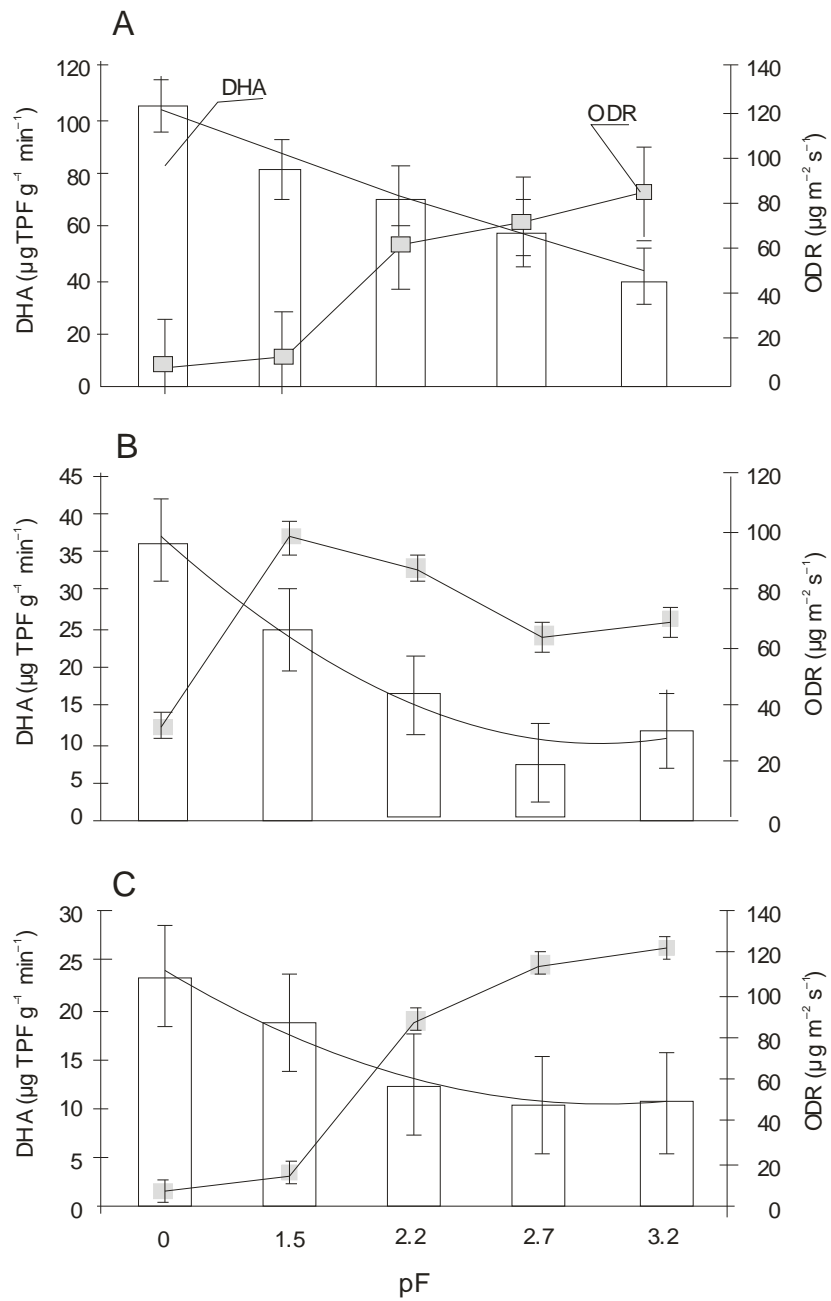


Fig. 28. The response of soil DHA to aeration factors (pF and ODR), of Leptosols (A), Histosols (B) and Fluvisols (C) (Wolińska and Bennicelli 2010)

A study by Brzezińska *et al.* (2004) on the effect of methane addition on DHA of Cambisol and Phaeozem developed from loess showed that DHA activity in methane amended soils was higher, by a maximum of 112 and 60%, respectively, than that in not amended (control) soils.

4.4. Methanotrophic activity and methane oxidation in soils

The influence of methane concentration on the methanotrophic activity of Gleysol and Podzol, characterized by similar granulometric composition and pH but different organic matter content, was examined by Włodarczyk *et al.* (2004a). In their experiment, methane was added, in increasing concentrations up to 15%, to closed vessels containing soil at water capacity of 159 hPa (pF 2.2), incubated at 20°C in O₂ atmosphere through 28 days. Data obtained (see Table 18) show higher methanotrophic activity by Gleysol than Podzol. There was an increase in the range of 0.5 to 10% initial methane for Gleysol and 1.25 to 15% in the case of Podzol. The average daily methane oxidation for Gleysol was 48.2 mg CH₄-C kg⁻¹ d⁻¹ and only 6 mg CH₄-C kg⁻¹ d⁻¹ for Podzol (Fig. 29).

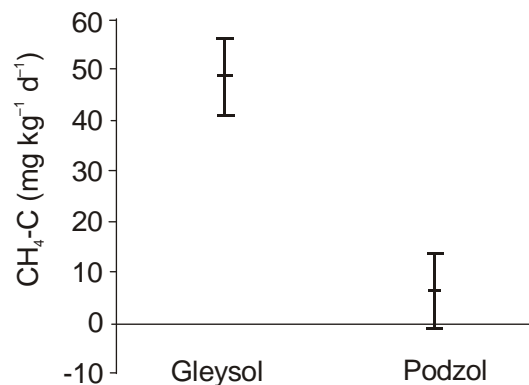


Fig. 29. Daily methane oxidation in Gleysol and Podzol. The bars represent 95% LSD confidence intervals (Włodarczyk *et al.* 2004a)

The kinetic parameters of methane oxidation were determined in three soils: Gleysol, Podzol and Cambisol by Walkiewicz *et al.* (2012). Soils were selected with a similar texture, which determines *in situ* soil air-moisture conditions and, thus, regulates soil methanotropic activity.

Soil samples were incubated at a constant temperature of 25°C, at water capacity of 16 kJ m⁻³ (pF 2.2). The incubation was preceded by 24-day pre-incubation with 10% methane in the headspace.

Kinetic parameters of CH₄ oxidation activity – K_m (the Michaelis constant) and V_{max} (the maximum reaction rate) – were determined (Tab. 21). In conclusion all tested soils showed potential for consumption of added methane. The Gleysol showed the highest methanotrophic activity, while the Cambisol showed the lowest activity and Podzol was in between (Walkiewicz *et al.* 2012).

Table 21. Kinetic parameters of CH₄ oxidation in tested soils (K_m and V_{max} calculated from linear regression of Lineweaver-Burk plots (Walkiewicz *et al.* 2012)

Soil type	K_m (μmol)	V_{max} (μmol g ⁻¹ h ⁻¹)
Gleysol	30.66	0.550
Podzol	19.79	0.443
Cambisol	5.98	0.137

5. CONCLUSIONS

Investigation was conducted on the biological activity of Polish soils, comprising Cambisols, Luvisols Phaeozems and also Podzols, Gleysols and Histosols, representing many textural classes: sands, silts, clays and loams, with various organic matter content and pH values. The potential biological activity of these soils was measured in fixed conditions of their incubation at 20°C, differentiated aeration and moisture status and enriched denitrification substrate on soil samples from the Database of Polish Mineral Soils of the Institute of Agrophysics PAS in Lublin. The results obtained, concerning denitrification, respiration, dehydrogenases activity and methane production in soils, are a reflection of above mentioned soil properties and experiment conditions. N₂O production and uptake in soils modified by soil properties, air-water conditions and organic substrates additions reached the highest values of 65 mg N₂O-N kg⁻¹ for production, 24 mg N₂O-N kg⁻¹ d⁻¹, 23 mg N₂O-N kg⁻¹ for uptake and 7 mg N₂O-N kg⁻¹ d⁻¹ for uptake rate. Soil respiration was the greatest in Phaeozems (on average 262 mg CO₂-C kg⁻¹). The Cambisols and Luvisols presented respiration activity 30% lower (on average about 165 mg CO₂-C kg⁻¹). The soil unit did not influence significantly the uptake of O₂ which was about 17%

v/v. O₂ consumption and CO₂ production in Cambisols and Phaeozems developed from loess and amended with CH₄ were, on average, 85 to 30% higher, respectively, as compared to the control soils. The greatest values of CO₂ production and O₂ consumption in Cambisols and Phaeozems were found at pH 5.5-7.5 and at Eh 500-600 mV. Dehydrogenase activity in Luvisols and Phaeozems reached the highest values of 1.08 and 2.31 mg TPF g⁻¹ 20 h⁻¹, respectively, including aeration, Eh, Fe(II) and temperature effects. Methane addition to Cambisol and Phaeozem developed from loess showed that DHA activity in methane amended soils was higher, by a maximum of 112 and 60%, respectively, than that in not amended (control) soils. Methanotropic activity of Gleysol (average daily methane oxidation of 48.2 mg CH₄-C kg⁻¹ d⁻¹) was much higher than that of Podzol (6 mg CH₄-C kg⁻¹ d⁻¹). This was confirmed by kinetic parameters of CH₄ oxidation activity – *Km* (30.66 μmol) and *Vmax* (0.550 μmol g⁻¹ h⁻¹) for Gleysol, and *Km* (19.79 μmol) and *Vmax* (0.443 μmol g⁻¹ h⁻¹) for Podzol. These parameters for Cambisol were the lowest – *Km* (5.98 μmol) and *Vmax* (0.137 μmol g⁻¹ h⁻¹). Comparison of indicators of soil biological activity of tested soil types on the level of fixed conditions of temperature, air-water and incubation time is shown in Table 22.

Table 22. Soil respiration and dehydrogenases activity (14-day incubation at 20°C, pH 1.5), denitrification (24-hour anaerobic incubation at 20°C) and methanogenic potential (77-day anaerobic flood incubation at 25°C). Each value is an average from three replications (Bieganowski *et al.* 2013)

Soils	Respiration mg CO ₂ -C k ⁻¹	Dehydrogenases μg TPF g ⁻¹ 20 h ⁻¹	Denitrification mg N ₂ O-N kg ⁻¹	Methane production CH ₄ -C k ⁻¹
Cambisols	96.9-297.5	7.22-108.4	0.10-26.6	3.46-279.7
Gleysols	127.5-393.1	14.5-196.3	95-18.8	140.1-520.1
Luvisols	132.7-237.3	1.81-11.2	n.t	n.t.
Phaeozoms	203.4-314.2	16.6-179.0	8.1-18.5	89.7-364.7
Podzols	71.9-195.4	1.21-4.50	1.5-12.9	3.17-193.9

n.t. – not tested.

6. REFERENCES

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7. SUMMARY

The aim of this monograph is to present the ability of Polish soils concerning N₂O production, consumption and release (emission) to the atmosphere, soil respiration, dehydrogenase activity and CH₄ oxidation. Investigated soils comprised several taxonomic units, such as Eutric Cambisols, Haplic Luvisols, Haplic Phaeozems, Eutric Histosols, Mollic Gleysols, Haplic Podzols and Rendzic Lep-tosols, formed from various textural classes: sands, silts and loams. Results obtained were also related to such soil properties as soil texture, Corg, pH, Eh, ODR, reduced iron (Fe II), H₂O₂ and C₂H₂ content. The data derive from analyses of soil samples stored in the Database of Polish Arable Mineral Soils of the Institute of Agrophysics, Polish Academy of Sciences in Lublin. The samples from the upper horizons, representing tested soils, were examined in model experiments with standardized conditions (soil aeration status and temperature), which allowed to express rather the potential of soils to perform the given processes than their current conditions, but data obtained in this manner can be comparable among different soils.

Keywords: biological activity, mineral Polish soils

8. SUMMARY IN POLISH

AKTYWNOŚĆ BIOLOGICZNA GŁÓWNYCH TYPÓW GLEB POLSKI

Badanie potencjalnej biologicznej aktywności gleb Polski dotyczyło powszechnie występujących na terenie Polski gleb brunatnoziemnych (Eutric Cambisols), płwoziemnych (Haplic Luvisols) i czarnoziemnych (Haplic Phaeozems) o zróżnicowanym składzie granulometrycznym, pH i z różną zawartością C_{org} . Były również badane, w niewielkiej liczbie, gleby bielcowe (Haplic Podzols), glejowe (Mollic Gleysols), torfowe (Eutric Histosols) i rędziny (Rendzic Leptosols). Analizy wykonywano w ustalonych laboratoryjnych warunkach, na materiale glebowym zgromadzonym w Bazie Danych Instytutu Agrofizyki PAN w Lublinie. Przedstawiono wiele danych dotyczących potencjalnej aktywności biologicznej gleb Polski, opublikowanych w różnych pracach, dotyczących głównych wskaźników aktywności: produkcji i absorpcji N_2O , oddychania (respiracji), aktywności dehydrogenazowej i metanotroficznej gleb.

Słowa kluczowe: aktywność biologiczna, mineralne gleby Polski

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