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ScienceDirect
Quaternary Research 67 (2007) 328–336
QUATERNARY RESEARCH
www.elsevier.com/locate/yqres

An assessment of changes in properties of steppe kurgan paleosoils in relation to prevailing climates over recent millennia

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Received 28 October 2005
Available online 15 February 2007

Abstract

Comparative analysis of morphological and chemical properties of the soil chronosequence on Kastanozems soils in the steppe zone of the Russian Plain, which included paleosoils buried beneath kurgans erected ca. 2000 BC, AD 50, AD 200, and AD 1250 was performed to reconstruct the paleoenvironmental conditions in this archeologically important region. Paleoenvironmental dynamics were traced using the state of microbial communities of paleo and modern soils (including the dynamics of total and glucose-reactive biomass, and the abundance of microorganisms grown on selected media). We demonstrate that the share of the glucose-reactive microorganisms in the microbial community, the ecological–trophic structure, and oligotrophicity index might serve as indicators of the state of microbial communities and be used for paleoenvironmental reconstructions. The morphological–chemical and microbial properties confirm an arid period ca. 2000 BC, slightly wetter conditions ca. AD 50, and more humid conditions ca. AD 1250.

Keywords: Steppe kurgans; Paleosoils; Climate change; Chemical properties; Microbial communities

Introduction

About 6000 yr ago, burial sites emerged as hills ("kurgans" or burial mounds) in the Eurasian steppes. Funeral ceremonies connected with the erection of these burial mounds have existed for more than 50 centuries up until the 15th century. With a lapse of time, however, kurgans have become an integral part of the steppe landscape. Kurgans are important objects of study in the humanities field, especially archeology and ethnography, because they provide information about spiritual and material culture of steppe inhabitants during prehistory and the Middle Ages.

In Russia, increasing attention has been drawn to paleosoils buried beneath the kurgans. Field and laboratory investigations on these burial mounds have given a rise to a new scientific approach, referred to as archeological soil science (Demkin, 1997). Soils are one of the few natural formations integrating prevailing climatic, lithological, geochemical, biological, and hydrological conditions at the time of their formation and subsequent evolution. Being *in situ* and buried (conserved) beneath kurgans, paleosoil profiles maintain their "paleoecological memory" of the history of the environment.

For their decoding, the discipline of soil science provides many opportunities for obtaining information on morphology and stratigraphy, level of humification, concentrations of carbonates and salts, and microbial activity of paleosoils. Quantitative and qualitative estimates of these parameters enable researchers to make inferences about particular prevailing environmental conditions at the time of erecting the monument and prior to it. Comparative analysis of paleosoil profiles that have developed under similar conditions (e.g., those that are in close vicinity, occupy the same relief position, are composed of the same parent material) but buried during different time windows (i.e. forming what is known as a soil chronosequence) provides important data on the evolution of soils and landscapes within a specific region.

When studying archeological monuments, chemical soil properties are often used to indicate the location, peculiarities, and economy of settlement (Eidt, 1985), but they also may be

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0033-5894/$ - see front matter © 2007 University of Washington. All rights reserved.
doi:10.1016/j.yqres.2007.01.001

used as stratigraphic markers (Haynes and Vance, 1968; Bettis and Thompson, 1982; Goldberg, 1986). Investigations of paleosoils beneath archeological monuments of different ages make it possible to reconstruct climate dynamics within historical time periods that are of particular interest (Ranov and Davis, 1979; Reider, 1980; Paulissen and Vermeersh, 1987; Weiss et al., 1993; Dalfes et al., 1997; Demkin, 1997; Fedoroff et al., 1997; Gerasimenko, 1997; Aleksandrovskiy et al., 2001).

Amongst these important indicators of soil-forming process, the characterization of soil microbial communities is a promising yet underdeveloped field. Microorganisms are an essential part of soil composition and are maintained for an indefinite period of time due to their ability to convert into a resting (dormant) state under stressful conditions (Roszak and Colwell, 1987). They may be used in studies of the dynamics of the natural environment on geological time scales (Zvyagintsev et al., 1985; Khlebnikova et al., 1990; Brockman et al., 1992; Friedman, 1993). However, there is little information on the state of microbial communities in paleosoils buried beneath archeological monuments (Demkina et al., 2000, 2003; Khomutova et al., 2004). We suggest that in surface and buried soils in dry steppe and semi-arid regions, an essential and significant component of the microbial community is maintained in this dormant state. It is therefore important to be able to estimate soil microbial biomass, which would comprise the cells at different stages of their life cycle.

The aim of this study is to analyze morphological and chemical properties and characterize microbial communities in paleosoils buried beneath kurgans of different ages in the steppes of Eastern Europe (Russian Plain), and to use this information to reconstruct paleoclimatic conditions over the past 4000 yr. In order to fulfill this aim, the following objectives were undertaken: (i) provide a comparative analysis of profile stratigraphies; (ii) determine the organic carbon content and concentrations of water-soluble salts, carbonates, and gypsum; and (iii) characterize microbial biomass and population densities of different trophic groups of microbial communities in paleosoils of different ages and in modern surface soils.

Site location

The study region is located in the dry steppe zone of the Lower Volga towards the south end of the Privolzhskaya Upland (Fig. 1). The climate is moderately continental, with mean annual precipitation of 350 mm and mean annual temperature of 7 °C. The key site (comprising a set of kurgans) is located on the high first above-floodplain terrace of the right bank of the Ilovlya River (left tributary of River Don) (Fig. 1). The terrace is well expressed in the topography and has a distinct 10-m bench above the level of the floodplain. Elevation of the surface is 50–60 m. The terrace is cut by ravines and gullies that provide good conditions for its drainage and deep (>10 m) level of ground waters within the Holocene. Therefore, it may be assumed that ground waters did not influence the soil-forming process. The ground waters have insignificant mineralization (less than 1 g/l) of sodium–hydrocarbonate composition.

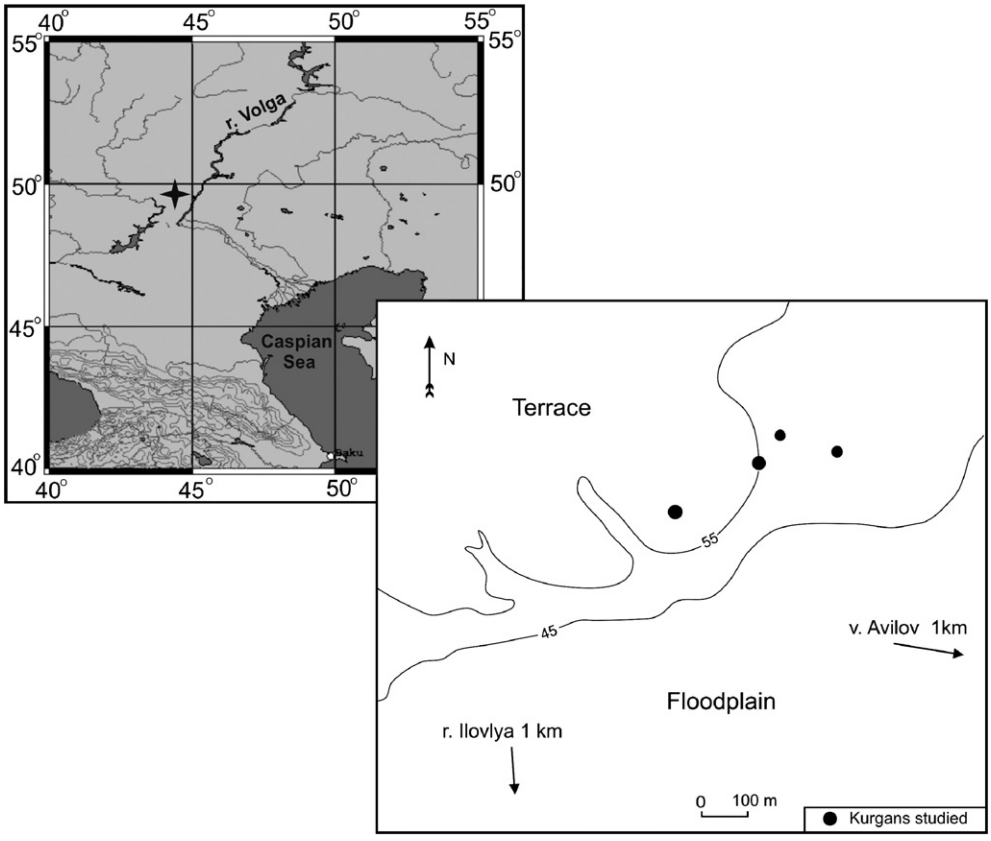

Figure 1. Location of the “Avilov” burial site studied.

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

The "Avilov" burial site studied is 1 km to the west of the village Avilov (Ilovlinsky district, Volgogradskaya oblast). Archeological excavations were performed by the expedition of Volgograd State University, led by Dr. I. V. Sergatskov. Here, kurgans are arranged in a compact group on an area with vegetation cover consisting mainly of *Artemisia incana* and *Festuca sulcata*. All objects were locations in a region of deep, modern, chestnut solonchakous soils (Kastanozems) with low spatial variability in their morphological and chemical properties. Solonetzic spots are not found within this area. Parent material consists of a uniform loess-like carbonated salty loam and lies at depths of more than 4 m. In summary, all paleosoils and modern soils investigated here are located in close vicinity on the same elements of the topography, they occupy the same geochemical position in the landscape, and all have similar lithologies. Therefore, we regard these paleosoils as a soil chronosequence and hypothesize that the main reason for the changes in their morphological, chemical, and microbial properties is determined by climatic factors, particularly the dynamics of humidity in the region.

Materials and methods

Here we present data from modern soils and paleosoils of archeological monuments of different ages. Buried paleosoils are dated according to the succession of steppe societies from Eastern Europe and the remains of the cultural periods over the last 6000 yr (Skripkin, 2005). The chronology is based on archeological toolkits including pottery, weapons, and other artifacts typical for burial rites of the tribes of the respective archeological cultures. The erection of kurgans is dated back to the Bronze Epoch (3000–2000 BC; the Catacomb culture), Early Iron Age (1st century: middle Sarmatian culture; late 2nd to early 3rd century: late Sarmatian culture) and the Middle Ages (early 13th century: Polovetskaya culture).

Field morphological analysis of profiles of each kurgan paleosoil and modern soil included estimates of the depth of genetic horizons, depths of carbonate, salts, and gypsum accumulations (Kovda, 1973). These were all determined from within trenches of each kurgan studied and through all walls of modern soil pits. Average values of all parameters were determined. Soil samples from paleosoils and modern soils were air-dried, averaged, and sieved through a 2-mm screen prior to laboratory analyses. The content of humus, carbonates, gypsum, soluble salts (sum of the contents of Na^+, Ca^{2+}, Mg^{2+}, Cl^-, SO_4^{2-}), pH values, and particle-size distribution were determined by standard methods (Methods of Soil Analysis, 1994).

For microbiological analysis, soil samples taken from A1, B1, and B2 horizons of both modern soils and from each paleosoil profile, were placed into sterile plastic bags and transported to the laboratory. Until measurements were undertaken, the samples were kept in conditions similar to the moment of sampling with respect to moisture and temperature. For estimating the organic carbon related to total microbial biomass (C-MB), the microbial fraction was extracted from the soil and the mass of the fraction, the amount of organic carbon, and the completeness of fractionation were determined. The microbial fraction was extracted from the soil based on procedures described by Faegri et al. (1977) and Khomutova et al. (2004) with some modifications. Samples (ca. 6 g) were mixed with 60 ml of 0.5% sodium pyrophosphate and subjected to ultrasonic treatment (two exposures of 30 s with a pause of 30 s between at a constant temperature of 4 °C) (Ramzay, 1988). The solution of sodium pyrophosphate was then added to the soil suspension to the final volume of 300 ml, mixed, and the soil residue was reduced to a pellet using centrifugation (2000 rpm for 30 min). The supernatant was decanted and the treatment of soil residue was repeated twice, as outlined above. The supernatants from all three treatments were combined and the microbial fraction again reduced to a pellet (5000 rpm, 90 min). The fraction was dried at 105 °C, and its mass was determined. The amount of organic carbon in the isolated fraction was determined by wet combustion. Soil samples were analyzed in triplicate, with the amount of organic carbon determined in 3–6 replicates.

The completeness of extraction of the microbial fraction from soils was estimated by measuring the number of microbial colony-forming units (CFU) in the combined final supernatant and final soil residue. In this case, the number of colony-forming microorganisms was used as a "label" of the whole microbial community. Microbial CFU were determined by agar plating ("soil agar", SA). The plates were inoculated with 0.2 ml of final supernatant and the same volume of water suspension of the final soil residue (in optimized dilutions). The plates were incubated at 22 °C and counted after 7 days. The analyses were carried out in five replicates. All solutions were sterilized prior to use and kept at 4 °C.

Carbon content within the glucose-reactivated microbial biomass was measured using the substrate-induced respiration method (Anderson and Domsch, 1978). Soil samples (2 g) were placed into flasks (15 ml), incubated 1.5–2 h, and ventilated. Then the glucose solution was put at concentration 10 mg/g soil (the resulting moisture of soil was 60–65% field capacity). The flasks were sealed and incubated 3–4 h at 22 °C. The concentration of CO_2 emitted from soil was measured by gas chromatography (Chrom-5, Czech Republic) and the rate of substrate-induced respiration was calculated as μg C/g soil/h. In order to calculate the amount of carbon within the glucose-reactivated microbial biomass (C-SIR), the calculation coefficient was used (Anderson and Domsch, 1978).

Total number and relative abundance of microorganisms of different trophic groups of microbial communities in buried paleosoils and modern soil were determined. This was done by measuring microbial colony forming units (CFU) by plate counting on 3 different types of carrier media: (i) rich medium (RM, in 1 l of tap water: 3 g nutritional agar, 3 g peptone, 1 g triptone, 1 g yeast extract, 1 g glucose, 20 g agar; Ananyeva and Vassilieva, 1985); (ii) on soil agar (SA, in 1 l of tap water: 200 g sterile soil, 20 g agar); and (iii) nitrite agar (NA, in 1 l of tap water: 2 g $NaNO_2$, 1 g Na_2CO_3, 0.5 g K_2HPO_4, 20 g agar;

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

Tepper, 1976). An index of oligotrophy was calculated as the ratio between CFU obtained on soil agar and rich medium (SA/RM ×100%; Nikitin and Nikitina, 1978). All measurements were performed in three replicates and mean values with standard errors for each parameter determined.

Results and discussion

For the dry-steppe zone of Russia, an important factor in the soil-forming process is climate humidity. Variations in annual precipitation greatly influence the formation, expression, and subsequent disappearance of different soil properties. Several soil-archeological studies have demonstrated that kurgan paleosoils of different ages are reliable indicators of centennial-scale climate variability (e.g., Ivanov, 1992; Aleksandrovskiy, 2002; Borisov, 2002; Demkin et al., 2004, 2006). Comparative analysis of different soil parameters (morphological, chemical, microbiological) can thus increase the detail and reliability of climatic reconstructions.

Morphological and chemical properties of paleo and modern soils

The main chemical properties of modern soil and kurgan paleosoils of different ages are presented in Table 1. Weighed average contents of soluble salts (0–200 cm layer), gypsum (0–100 cm layer), and carbonates (0–50 cm layer) are presented in Figure 2. The pit of modern soil in the center of kurgan group was typical for its vegetation cover, lithology, and geomorphology. The soil was characterized as chestnut, weak solonetzic, and deep saline. The content of physical clay (<0.01 mm) particles changed from ca. 31% to ca. 47% across the profile. Texture differentiation was quite distinct in the upper part of the profile, with the content of clay (<0.001 mm) particles in B1

Table 1
Chemical properties of buried and modern soils

Horizon, depth, cm	Humus, %	pH_{H_2O}	Salts, %	$CaCO_3$, %	$CaSO_4$, %	Particle-size distribution, %	
						<0.001 mm	<0.01 mm
Chestnut-like paleosoil, pit D-510, the Bronze Ages (ca. 2000 BC: the Catacomb culture)							
A1 74–86	0.6	8.3	1.53	4.9	1.8	22	36
B 86–103	0.4	8.2	1.58	7.8	1.8	23	40
Bca 103–118	Nd[a]	8.5	1.24	13.8	0.9	26	44
BCca 118–145	Nd	8.4	0.93	7.6	0.7	26	41
Cs, g 145–220	Nd	8.3	1.96	13.6	11.4	22	40
Chestnut paleosoil, pit D-503, the Early Iron Ages (ca. AD 50: middle Sarmatian culture)							
A1 48–61	1.4	7.1	0.30	0.1	0	19	33
B1 61–80	1.2	7.6	1.25	0.8	1.7	26	39
B2ca 80–94	0.9	7.8	1.35	14.2	3.6	27	43
BCca 94–147	Nd	8.3	0.58	16.8	0.5	29	45
C 147–175	Nd	8.1	0.42	8.8	0.7	24	34
Cs, g 175–230	Nd	8.1	1.16	8.8	11.0	18	36
Chestnut paleosoils, pit D-509, the Early Iron Ages (ca. AD 200: late Sarmatian culture)							
A1 26–36	2.3	7.7	0.30	0.1	0.8	21	37
B1 36–55	1.2	8.4	2.02	2.4	6.6	28	44
B2ca 55–66	0.9	8.7	1.65	14.2	2.6	28	45
BCca 66–109	Nd	8.8	1.52	16.5	2.1	32	45
C 109–150	Nd	8.9	1.18	11.3	1.8	26	40
Cs, g 150–200	Nd	8.7	1.86	7.6	7.1	26	42
Chestnut paleosoils, pit D-504, the Middle Ages (ca. AD 1250: Polovetskaya culture)							
A1 90–102	1.2	7.6	0.06	0.1	0.1	14	35
B1 102–122	1.1	7.8	0.08	0.2	0.2	27	44
B2ca 122–137	0.8	8.4	0.06	12.4	0.3	31	45
BCca 137–176	Nd	8.6	0.05	15.4	0.1	30	45
C 176–280	Nd	8.4	0.05	8.0	0.2	23	34
C 280–350	Nd	8.3	0.20	3.0	0.4	16	32
Control modern soil, pit D-505 (current time: AD 2000)							
A 0–10	2.2	7.3	0.03	0.1	0	12	31
B1 10–26	1.7	7.6	0.03	0.2	0	22	40
B2ca 26–36	0.8	8.6	0.05	6.7	0	27	45
BCca 36–82	Nd	9.0	0.06	18.4	0.3	27	49
C 82–130	Nd	8.3	0.25	12.3	0.3	28	45
Cs, g 130–200	Nd	8.1	1.39	9.5	1.9	14	32

[a] Nd—not determined.

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

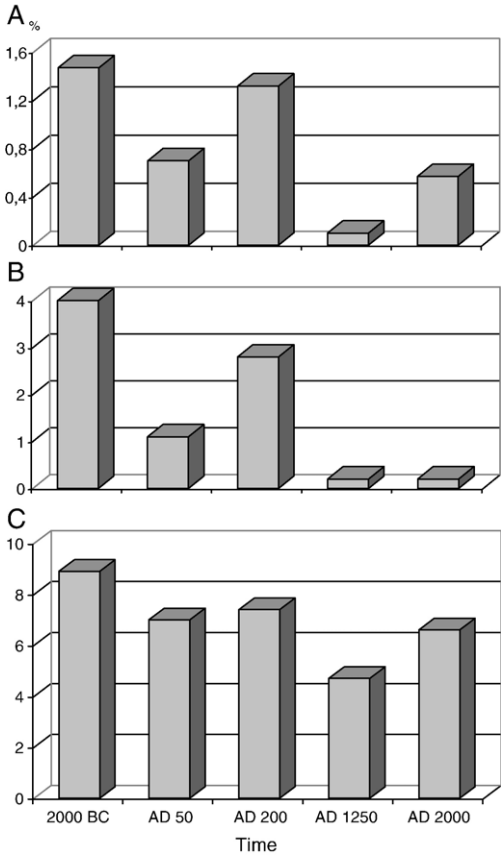

Figure 2. Weighed average contents of (A) soluble salts in 0–200 cm layer, (B) gypsum in 0–100 cm layer, and (C) carbon in 0–50 cm layer of the paleosoils of different ages and modern soil.

horizon almost twice that in the A1 horizon. The thickness of humus layer (A1+B1) averaged 26 cm, while the humus content decreased from 2.2% (A1 horizon) to 0.8% (B2ca horizon). The middle part of the profile had the highest soil reaction (pH 8.6–9.0) and the recorded effervescence depth was 27 cm. Calcareous neoformations were represented by calcite nodules ("white eyes") 6.7 mm in diameter (abundance of 7 nodules per square decimeter) and impregnated with fine calcite crystals. The content of $CaCO_3$ exceeded 18% in the upper 0.5 m layer in the zone of maximum accumulation, with an average content of 6.6%. Accumulations of soluble salts (1.4%) and gypsum (1.9%) were observed at 130 cm depth. Average salt content was about 0.6% (layer 0–200 cm), while that of gypsum was about 0.2% (between 0–100 cm).

The oldest paleosoil was studied beneath the kurgan of the Catacomb culture (from the second half of 3rd to the beginning of the 2nd millennium BC), which could be dated with the findings in the graves from late in the 3rd millennium BC to the beginning of the 2nd millennium BC (Skripkin, 2005). In this paper we assign this paleosoil an age of ca. 2000 BC, pit D-510. It was characterized as chestnut-like calcareous, nonsaline, solonetzic soil, and had no analogs in the current soil cover within the terrace. The paleosoil was of medium loam texture with similar distribution of particles in A1 and B horizons but without signs of salinization. The thickness of the humus layer (A1+B1) averaged 29 cm. Due to diagenesis, the initial humus content (before burial) decreased by 1.5–2 times and did not exceed 0.6% (A1 horizon). The pH values were similar along the whole profile (8.2–8.5), while the effervescence line coincided with the upper boundary of paleosoil; here the content of $CaCO_3$ was almost 5% in the A1 horizon. Calcareous neoformations were mostly of fine calcite impregnation, and there were few nodules of mean size 5.8 mm (with 4 nodules/dm^2). The average $CaCO_3$ content was 8.9% (layer 0–50 cm). The paleosoil was highly saline; accumulation of soluble salts was determined deeper than 55 cm, and that of gypsum deeper than 70 cm; their average contents were 1.5% (layer 0–200 cm) and 4.0% (layer 0–100 cm), respectively.

Two kurgans of the Early Iron Age have been attributed to the Sarmatian culture (Skripkin, 2005). The oldest was erected ca. AD 50 (middle Sarmatian culture, pit D-503). The paleosoil during this time can be classified as medium loamy, saline, and deep solonetzic chestnut soil. The upper part of the profile was clearly differentiated by texture and significantly impoverished clay quantities in the A1 horizon. The structure of illuvial solonetzic B1 horizon was fine prismatic. The thickness of the humus layer (A1+B1) averaged ca. 32 cm, while the fixed humus content was 1.4% in A1 horizon (although with due correction for diagenetic changes it could be up to 1.5–2 times higher; Demkin, 1997). The pH values increased along the profile from 7.1 to 8.1–8.3, and the effervescence line registered at a depth of 30 cm. Calcareous neoformations were represented by well-formed nodules of mean size 5.8 mm (10 nodules/dm^2). In the BCca horizon, the content of $CaCO_3$ reached almost 17%, although the average carbonate content was 7.0% (layer 0–59 cm). In the upper part of the profile, soluble salts and gypsum of diagenetical origin (leached from the embankment) were found. Their initial accumulations were from depths of 125 cm. The average salt content was 0.7% (0–200 cm), while that of gypsum was 1.1% (0–100 cm).

The next kurgan attributed to the late Sarmatian culture was dated back to the late 2nd century or early 3rd century (ca. AD 200, pit D-509). The paleosoil was characterized as saline and solonetzic chestnut. It was medium loamy and the profile was differentiated by texture, with well-expressed signs of solonetz process. The thickness of the humus layer (A1+B1) averaged 29 cm, while the content of humus along the profile decreased from 2.3% to 0.9%. The pH values reached 8.7–8.9, and the effervescence line was fixed at the depth of 29 cm. Calcareous neoformations were represented predominantly by impregnation with fine crystalline calcite and fine nodules (size 5 mm, abundance 7 nodules/dm^2). In the accumulation zone (B2ca and BCca horizons), $CaCO_3$ content was ca. 14–16%, although average content of carbonates was lower at ca. 7.4% in the 0–50 cm layer. In the upper part of the profile the veinlets of soluble salts of diagenetical origin were fixed. The initial accumulation of salts (before burial) was found at a depth of 70 cm, while that of gypsum was found at a lower depth of 120 cm. The average soluble salt content was 1.32% (layer 0–200 cm); that of gypsum was 2.8% (layer 0–100 cm).

Paleosoil of the Middle Age was studied in the kurgan of the Polovetsky culture dated back to the early 13th century (ca. AD

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

1250, pit D-504). It was characterized as a residually solonetzic and deep, saline, chestnut soil. In common with the other paleosoils, it had an even medium, loamy texture. The content of physical clay varied between 32% and 45%. The upper part of the profile was differentiated by texture, and here the content of clay was almost twice as high in illuvial B1 horizon compared to the A1 horizon. The solonetzic signs were residual, while the structure of B1 horizon was fine prismatic with abundant dark manganic oxides on ped faces. The thickness of the humus layer (A1+B1) averaged 32 cm, while humus content gradually decreased along the depth from 1.2% to 0.8%. The effervescence line was fixed at a depth of 33 cm. Calcareous neoformations were represented by well-formed, coarse, and abundant nodules (7 mm, 16 nodules/dm^2). The highest content of $CaCO_3$ (over 15%) was fixed in the BCca horizon, while average carbonate content was only 4.7% in the 0–50 cm layer. The 2-m soil-ground depth was leached of practically all soluble salts and gypsum, and the highest contents did not exceed 0.2–0.4%. Below 180 cm, scarce veinlets of gypsum were observed.

Thus, the data obtained suggest that within the last 4000 yr, the soils on the Ilovlya terrace underwent distinct changes. Recently it was established (Demkin et al., 2004) that in the period 3000–3400 BC, saline and solonchakous chestnut paleosoils were developed on the terrace. Due to an abrupt change in soil-forming conditions that occurred 2500–2000 BC because of climate aridization, they were transformed to chestnut-like eroded calcareous paleosoils. These soils were found beneath the kurgan of the Catacomb culture and dated back to ca. 2000 BC. It should be noted that ca. 2000 BC the chestnut-like paleosoils were widely spread on the catchments and high river terraces, not only on the south of the Privolzhskaya upland but also within other natural regions of the Southern Russia steppes (e.g., Yergeny upland, Cis-Caspian lowland, Volga plain; Demkin et al., 2004). During the 2nd millennium BC the soils evolved into zonal chestnut soils, followed by the occurrence of cyclic changes of different soil properties. The regularities of soil evolution on the Ilovlya terrace within the past 4000 yr are in agreement with general regularities of the late Holocene pedogenesis in the steppes of the Lower Volga region. Centennial-scale dynamics of the concentrations of soluble salts, gypsum, and carbonates along the soil profile were regulated predominantly by the level of atmospheric humidity.

Taking into account the solubility and migration activity, we estimated weighed average contents of these soluble components for the layers of 0–200, 0–100, and 0–50 cm (Fig. 2). The dynamics of the content of soluble salts (Fig. 2A), gypsum (Fig. 2B), and carbonates (Fig. 2C) in these layers were similar in the content and changes were synchronous. The highest contents were found in the chestnut-like soils dated back to ca. 2000 BC. In the chestnut paleosoil dated back to ca. AD 50 these parameters were distinctly lower, and increased in ca. AD 200. The highest leaching of the profile was observed in the paleosoil dated back to the Middle Ages (ca. AD 1250). In comparison, modern soil was characterized by higher salt content (almost 6 times, 0–200 cm layer) and carbonates (almost 1.5 times, 0–50 cm layer).

Table 2
Dynamics of the depth of accumulations of soluble salts, gypsum, and carbonates in the soils studied

Parameters	Time				
	ca. 2000 BC	ca. AD 50	ca. AD 200	ca. AD 1250	AD 2000
Depth of the accumulation of soluble salts, cm	55	125	70	>200	130
Depth of the accumulation of gypsum, cm	70	125	120	180	130
Depth of the effervescence line, cm	From the surface	30	29	33	27

Temporal variability of quantitative indices was in a good agreement with morphological analysis of soil profiles of different ages, particularly with the accumulation depth of salts, gypsum, and carbonates (Table 2). Closest to the surface, they existed in the paleosoil dated back to the Bronze Age, while the deepest existed during the Middle Ages. The regularities of the dynamics of the profile distribution and the content of soluble components indicate that most arid conditions of soil formation took place ca. 2000 BC in the region studied. Dry climatic conditions (but less significant) had fallen to ca. AD 200. In the 1st century of the new era (ca. AD 50) a short-term humid shift of the climate took place. However, more significant increase of atmospheric humidity had fallen to 12th–13th centuries (ca. AD 1250, Middle Age climatic optimum). From the combination of morphological and chemical parameters of the soils studied, we suggest that the climatic conditions most similar to the modern ones existed within first centuries of the new era.

Characterization of microbial communities in paleo and modern soils

We attempted to trace the patterns of environmental conditions derived from the data above in the state in the state of microbial communities from paleosoils and modern soil. The values related to microbial biomass carbon fractionated from A1, B1, and B2 horizons of modern soil constituted 700–1100 μg/g, while the completeness of fractionation was 20–35%. In paleosoil horizons these values varied from 240 to 660 μg/g soil, and the completeness of fractionation was 20–68%. It should be noted that mycelial forms of microbial population (mainly fungi) are not included in the estimates of total microbial biomass (C-MB), which comprises mainly the bacterial cells and spores. However, the mycelial forms in the soils of dry steppe zone constitute an insignificant part of the microbial carbon and do not exceed 7% of C-MB (Borisov et al., in press). From the data obtained here, the values of the carbon related to C-MB were calculated. They constituted 2050–6150 μg/g in modern soil and 620–1750 μg/g in paleosoils (Table 3).

In paleosoils, the level of microbial carbon was 4–7 times lower than in modern soil and varied within the soil chronosequence 1.8–2 times. We did not find a monotonous decrease of C-MB values dependent on the time of burial. In general in A1 and B2 horizons, the dynamics of C-MB within

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

Table 3
Carbon related to the total microbial biomass (C-MB), carbon of the glucose reactive microbial biomass (C-SIR), abundance of microorganisms grown on the soil agar (SA), nitrite agar (NA), and rich media (RM) in the paleosoils of different ages and modern soil

Soil, horizon	C-MB, μg/g soil	C-SIR, μg/g soil	Number of CFU, 10×6 CFU/g soil		
			SA	NA	RM
Chestnut-like buried soil (ca. 2000 BC)					
A1	1655.5±196.6	0.37±0.00	32.0±4.13	1.7±0.13	18.4±1.31
B1	889.9±76.4	0.37±0.00	2.9±0.10	0.5±0.03	3.2±0.10
B2	1188.0±134.5	0.37±0.00	3.6±0.11	0.5±0.03	2.4±0.10
Chestnut buried soil (ca. AD 50)					
A1	952.6±99.8	45.94±3.47	39.1±1.46	2.6±0.05	27.3±1.38
B1	878.3±72.2	9.60±3.75	2.6±0.25	0.5±0.03	3.8±0.15
B2	756.4±62.8	11.51±1.43	5.6±0.12	0.6±0.01	3.1±0.12
Chestnut buried soil (ca. AD 200)					
A1	853.9±80.8	6.50±1.29	29.0±1.01	1.8±0.04	19.9±1.00
B1	772.0±77.6	0.37±0.00	2.2±0.16	0.6±0.03	4.2±0.22
B2	1106.0±105.8	0.37±0.00	4.6±0.09	0.6±0.01	2.5±0.22
Chestnut buried soil (ca. AD 1250)					
A1	1745.7±311.5	13.00±6.37	35.0±1.13	1.9±0.08	19.3±1.06
B1	624.3±65.0	25.92±1.10	4.4±0.09	0.6±0.03	3.8±0.31
B2	1334.0±115.5	93.91±12.11	2.2±0.08	0.4±0.01	2.2±0.11
Chestnut modern soil (AD 2000)					
A1	6149.0±806.5	657.86±18.32	23.5±1.29	1.9±0.15	17.4±1.24
B1	2045.8±179.9	73.98±29.96	5.0±0.19	0.6±0.02	3.7±0.08
B2	5212.4±696.1	16.40±6.56	2.3±0.08	0.4±0.01	2.5±0.09

the soil chronosequence was more expressed than in the B1 horizon, and that is likely to be connected to the development of solonetzic process. The weighed average values of C-MB in paleosoils buried ca. 2000 BC and ca. AD 1250 were higher than those in paleosoils buried at ca. AD 50 and ca. AD 200 and were about 4 times lower than in modern soil samples (Fig. 3A).

The carbon of glucose reactive microorganisms (C-CIR) in the modern soil decreased down the profile and constituted 658–16 μg/g (Table 3). In paleosoils, the values were considerably lower and varied from nonregistered values (0.37 μg/g) to comparatively high values (94 μg/g). The decrease of this parameter was also not dependent on the time of the paleosoil burial. The specific dynamics of this parameter was observed in paleosoils buried ca. AD 50 and ca. AD 1250: in B1 and B2 horizons in paleosoil buried ca. AD 50, the values of C-SIR were similar, while in the paleosoil buried ca. AD 1259, an increase of C-SIR down the profile was observed. Weighed average value of C-SIR (Fig. 3B) in modern soil was about 210 μg/g, while in paleosoils they were significantly lower but also very different: in paleosoils buried ca. AD 50 and ca. AD 1250 values of C-SIR were higher compared to other paleosoils. C-SIR seems to be an informative parameter and characterizes the level of dormancy and the ability of a part of the microbial community to reactivate and may be considered as a specific physiological characteristic of microbial community. In modern soil the reactive part of the microbial community constituted ca. 5.4% (Fig. 3C). In paleosoils, however, it varied from 0.03–0.21% (paleosoils buried ca. 2000 BC and ca. AD 200, respectively) to 2.2–3.4% (paleosoils buried ca. AD 50 and ca. AD 1250, respectively). We propose that the differences observed may be connected to the specific paleosoil conditions (such as soil moisture content and hence climate humidity) of the respective time windows.

To estimate ecological trophic structure of microbial communities, the abundance of microorganisms (number of colony forming units, CFU) were determined from growth on different types of media: (i) soil agar (SA) when microorganisms utilize dispersed nutrients, (ii) nitrite agar (NA) when microorganisms utilize not readily available organic matter (soil humus), and (iii) rich nutrient medium (RM) when microorganisms utilize readily available organic matter (plant residues) (Table 3). In modern soil, the abundance of microorganisms of all trophic groups studied was highest in the A1 horizon and decreased down the profile. In paleosoils the abundance were highest in the A1 horizon, and these levels were comparable

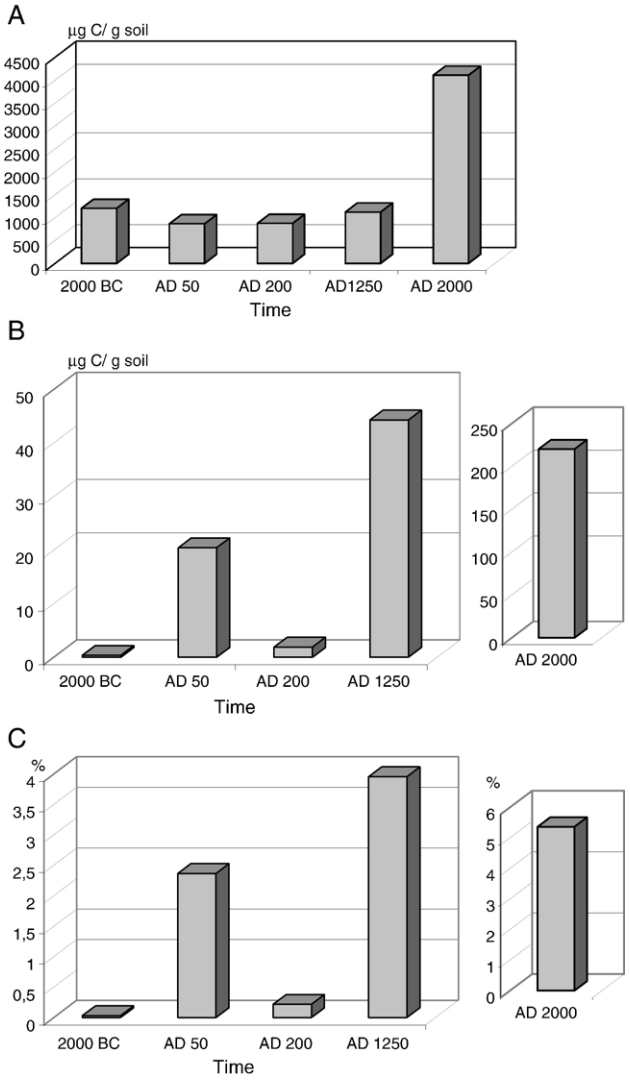

Figure 3. Weighed average estimates (A1+B1+B2 horizons) of carbon of total microbial biomass; C-MB (A); carbon of glucose reactive microorganisms, C-SIR (B, left—paleosoils, right—modern soil); and the share of glucose reactive microorganisms in the community (C, left—paleosoils, right—modern soil).

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

with those in the modern soil (microorganisms utilizing humus, NA) and in a number of cases exceeded it (microorganisms utilizing dispersed elements, SA, and utilizing readily available organics, RM). In the B1 horizon, the abundance were comparable with those of modern soil (NA and RM) or less (SA). In the B2 horizon, the abundance were comparable with those in modern soil (NA and RM) or exceeded modern values (SA).

Two indices were calculated to compare the peculiarities of ecological trophic structure of microbial communities: the ratio of the abundance of microorganisms grown on the media used (SA:HA:RM) and an index of oligotrophy (SA:RM × 100). The latter characterizes the ability of microbial community to assimilate nutritional elements from the dispersed state: the higher this index is, the poorer nutritional conditions the soil microorganisms are adapted for, and vice versa. For the case of dry regions, a lower index indicates a higher input of plant residues to the soil associated with more humid environmental conditions. Therefore, both dominating RM microorganisms and an index of low oligotrophy are indicative of an increasing input of plant residues into the soil caused by increasing atmospheric humidity.

Table 4 presents the weighed average ratio values (%) through the profile (A1 + B1 + B2) of the abundance of microorganisms of different ecological trophic groups from the soils studied. The highest proportions of microorganisms utilizing plant residues were found in paleosoils buried ca. AD 50 and ca. AD 200, and also in modern soil samples. For these soils we calculated the lowest index of oligotrophy (Table 4).

These microbiological data allow us to make certain inferences about past prevailing environmental conditions. For example, based on low C-MB and C-SIR values, a low proportion of microorganisms utilizing readily available organics, and a high index of oligotrophy, climatic conditions of the Bronze Epoch (ca. 2000 BC) are inferred to be more arid in comparison to modern times. In contrast, the Early Iron Age can be characterized by more humid climatic conditions (ca. AD 500–ca. AD 200) as determined by the high share of carbon of microorganisms reactivated by glucose in the microbial community, the high share of microorganisms grown on rich media, and low index of oligotrophy. Soil buried in the Middle Ages (ca. AD 1250) is characterized by high total and reactivated by glucose microbial biomass (especially in B1 and B2 horizons) that suggests increased moisture conditions. However, low share of microorganisms grown on rich media and high oligotrophy index point to the reverse.

Table 4
Ecological trophic structure (SA:HA:RM) and oligotrophy index (SA:RM × 100) of microbial communities of paleosoils of different ages and modern soil (weighed average values for the layer A1 + B1 + B2)

Time	SA:HA:RM (%)	SA:RM × 100
ca. 200 BC	59:4:37	157
ca. AD 50	55:4:41	135
ca. AD 200	53:5:42	124
ca. AD 1250	59:4:37	159
AD 2000 (modern)	54:5:41	131

This discrepancy may be explained by undertaking a more detailed analysis of the mound embankment. On the embankment, archeologists had found traces of a large burial fire. Despite the fact that the soil was sampled out of visual fire zone, the influence of high temperature evidently has affected physiological properties of soil microbial communities and their ability to reactivate (A1 horizon) and grow. Therefore, despite high microbial biomass, the activity of the community was depressed and could not reliably reflect natural circumstances of the past. This case confirms our point that microbial communities are useful indicators of the state of natural environment. Strong anthropogenic disturbance (burial fire) impacted specific properties of the microbial community (e.g., its physiological state), which is why in this particular case we believe that more reliable reconstructions of the environmental conditions ought to be based on other microbial parameters, in particular on the amount of total microbial biomass and ability of microorganisms to reactivate in deeper horizons (B1 and B2), and which point to the elevated level of humidity. Thus, humidity conditions occurring during the Middle Ages must be considered as moist and not arid.

To summarize, the prevailing conditions in the steppes of Eastern Europe during the past 4000 yr had been distinctly variable. The state of microbial communities in paleosoils beneath kurgans of different ages allows us to determine that arid conditions existed in the region studied in ca. 2000 BC. By the time of middle Sarmatian culture (ca. AD 50) a short-term increase of atmospheric humidity took place, followed by a subsequent decrease within the late Sarmatian period (ca. AD 200). During Middle Ages (ca. AD 1250), the climate shifted towards wetter conditions.

Conclusions

The morphological and chemical properties contained within soil chronosequences, and the biomass and physiological peculiarities of soil microbial communities, can both be used to interpret changes in climatic conditions in the Russian Plains of Eastern Europe over the last 4000 yr. The paleosoils of archeological monuments of different ages indicate that in the late 3rd millennium BC, an abrupt aridization of the climate took place that reached a maximum ca. 2000 BC, and which led to a paleoecological crisis. This period resulted in the desertification of the landscapes, denudation, dehumification, salinization, and carbonatization of paleosoils. As a result, ca. 4000 yr ago, unusual chestnut-like soils widely spread in the region. Microbial communities of the chestnut-like paleosoils are characterized by low reactivation ability and high degree of oligotrophy, despite their high biomass. The Middle Sarmatian time (1st century AD) is marked by a shift towards an increase of atmospheric humidity (micropluvial), followed over the next two centuries by a dry period. These dynamic features of the natural environment are traced in the properties of microbial communities, specifically the dynamics of glucose reactivated biomass and ecological trophic

https://doi.org/10.1016/j.yqres.2007.01.001 Published online by Cambridge University Press

structure. The Middle Ages (AD 1100–1300) are characterized by a more humid climate, equivalent to the Middle Age climatic optimum. The increase in precipitation during this period led to distinct changes of soil properties, particularly desalinization, displacement of the effervescence line towards deeper layers, increase of humus content, degradation of solonetzic signs, high total microbial biomass, and reactivation ability of microorganisms in B1–B2 horizons. Microbial communities from paleosoils buried under kurgans maintain a number of parameters reflecting the paleoecological conditions at the moment of the kurgan erection, and they seem to be more sensitive—that is, indicative more of short-term climatic fluctuations—than the morphological and chemical properties of paleosoils.

Acknowledgments

The work was supported by Russian Foundation for Basic Research and the Program for Fundamental Research of the Presidium of Russian Academy of Sciences. We would like to thank 2 anonymous reviewers, whose comments greatly improved the manuscript, and Dr. Anson Mackay for the time for improving the language of the manuscript.

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