APPLICATION OF PHYSIOLOGICAL AND BIOCHEMICAL METHODS FOR COMPARATIVE ANALYSIS OF THE FUNCTIONAL ACTIVITY AND COMPOSITION OF HYDROLYTIC MICROBIAL COMPLEXES OF MODERN AND BURIED CHESTNUT SOILS AND BURIED PERMAFROST SOILS

Koltsova E.. Tyapkina A.. Manucharova N.
Lomonosov Moscow State University. Moscow. Russia

BACKGROUND

The properties of paleosols as indicators of biosphere development gain increasing attention of researchers for evaluation of the possible consequences of global changes of environment and climate. Recently the subject of paleomicrobiome long-term sustainability mechanisms and transformation of microorganisms required for survival in subsurface sediments became a subject of great scientific interest. The question about functional activity and structure of microbiological community in subsurface sites remains open. Proven that microbiological community stays “preserved” and keeps original properties from the moment of burial, paleosoils can be considered as natural collections of microorganisms and may have a great biotechnology potential.

AIM OF THE STUDY

The comparative analysis of the functional activity and composition of hydrolytic microbial complexes of modern and buried chestnut soils and buried permafrost soils

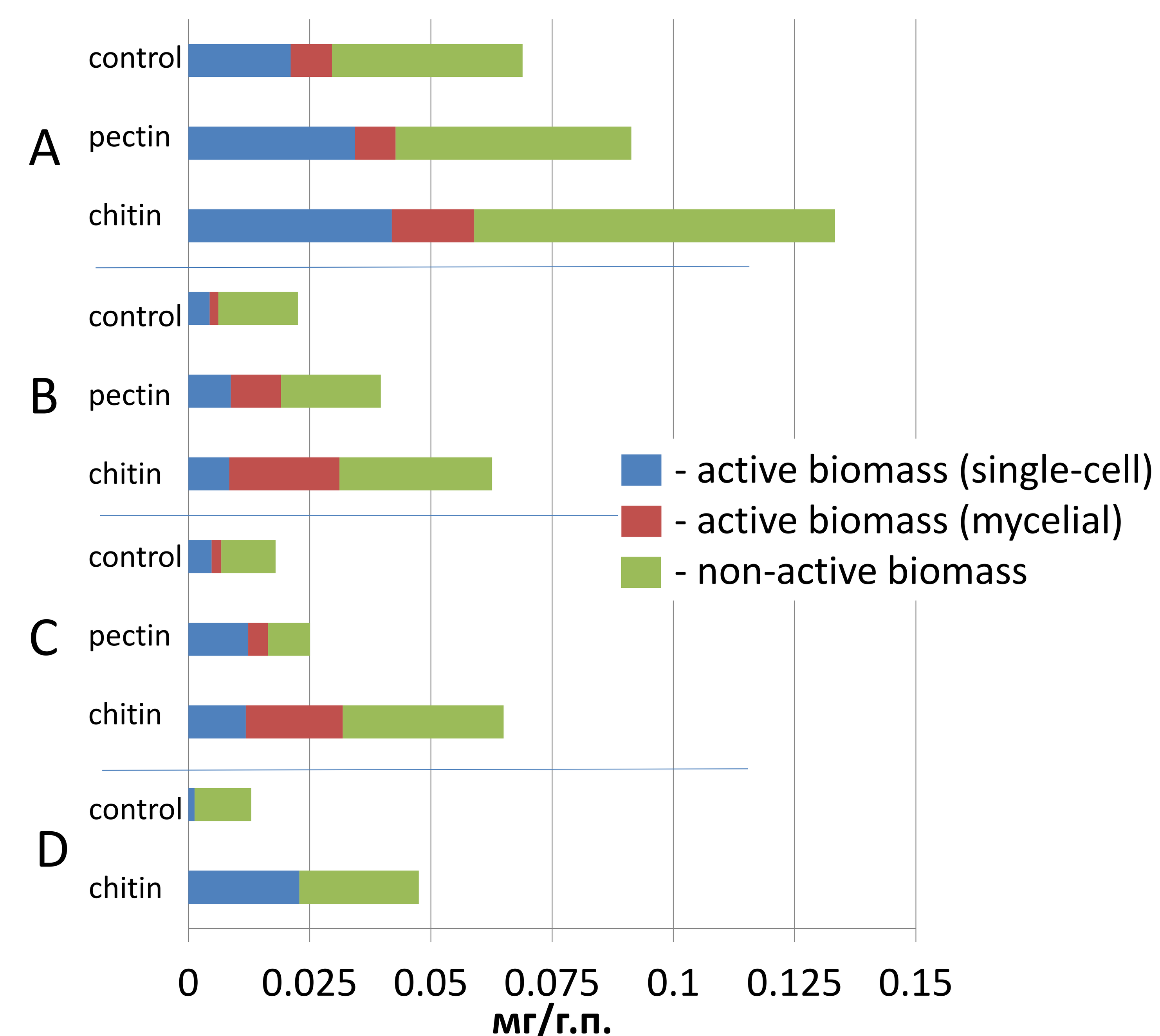
MATERIALS & METHODS

Subjects of the study were the buried subkurgan paleosoils (deposition depth 0.5 and 2.5 m, burial age 3500 and 4500 years respectively), modern chestnut soils and buried permafrost marine terrace sediments (deposition depth 9 m). The structure of the hydrolytic microbial complex was determined by the microcosm method with initiation of microbial succession by humidification and introduction of purified polysaccharides: chitin (ICN Biomedicals. Germany) and pectin (Sigma. Germany) at concentrations of 0.2%. Soil humidified with water (1 mL/5 g soil) without a substrate was used as a control. Bacterial cell numbers, mycelium length, and biomass of actinomycetes and fungi were determined by the fluorescence microscopy (Axioskop 2 plus, Zeiss, Germany) on days 0, 3, and 10. Diversity and abundance of metabolically active cells representing individual phylogenetic groups were determined using fluorescence in situ hybridization (FISH).

RESULTS

		Modern soil			Paleosoil. buried ~3500 years ago.			Paleosoil. buried ~4500 years ago.			Buried parmafrost sediments		
	substrate	Biomass. mg/g of soil	%	p	Biomass. mg/g of soil	%	p	Biomass. mg/g of soil	%	p	Biomass. mg/g of soil	%	p
Procaryotes (single-celled)	control	0.060±0.007	100	-	0.021±0.003	100	-	0.012±0.002	100	-	0.013±0.004	100	-
	pectin	0.082±0.006	137	0.026	0.029±0.005	138	NS	0.021±0.005	175	NS	0.000±0.000	-	-
	chitin	0.113±0.015	188	0.014	0.040±0.008	190	NS	0.044±0.007	366	0.013	0.048±0.005	369	0.002
Procaryotes (mycelial)	control	0.009±0.001	100	-	0.002±0.001	100	-	0.002±0.001	100	-	0.000±0.000	-	-
	pectin	0.008±0.001	88	NS	0.010±0.002	500	0.030	0.004±0.001	200	0.045	0.000±0.000	-	-
	chitin	0.017±0.002	189	0.002	0.023±0.005	1150	0.018	0.021±0.002	1050	0.005	0.000±0.000	-	-
Eucaryotes (mycelial)	control	1.806±0.025	100	-	0.120±0.064	100	-	0.007±0.012	100	-	0.000±0.000	-	-
	pectin	5.883±0.099	326	<0.001	0.063±0.021	52	NS	0.021±0.021	300	NS	0.000±0.000	-	-
	chitin	2.477±0.029	137	<0.001	0.000±0.000	-	-	0.099±0.136	1414	NS	0.000±0.000	-	-

Absolute total biomass values of microorganism groups and their relative values after incubation with substrate (10th day of the experiment). Statistical analysis was performed with pair-sample Student T-test (N=3). P<0.05 represents significantly different values from control samples. NS represents non-significant difference



Active procaryote biomass after humification and introduction of substrate (10th day of the experiment) in samples of: 1 - modern soil; B – soil, buried ~3500 years ago; C – soil, buried ~4500 years ago; D –buried permafrost sediments

RESULTS

The absolute values of biomass in the control soil samples had a gradual decrease of the values with increasing age and depth of deposition of the samples, while the samples of buried soils and buried permafrost sediments responded much better to introduction of substrates (biomass increased 5–7 fold if compared to control) than the samples of modern soils. The fraction of cells identified as metabolically active was comparable in all soil samples (30–40% of the total cell number), except permafrost samples (10%). The rate of increase of the share of active microorganisms in both samples of buried soil and sediments was much higher than in the modern soils and the intensity of response correlated with the age and deposition depth of the sample.

CONCLUSION

Absolute values of total and active biomass gradually decreased with the increase of deposition depth and age of the soil, but the intensity of response to the introduction of the substrate increased with the deposition depth and age of the soil. Supposedly it is due to the fact that the organic substance income to subsurface microbiomes is low and there occurs the selection of species, which are able to hydrolise complex substrates and are capable of fast growth and multiplication.