# INTRODUCING THE PHYTOLITH ANALYSIS: A SUITABLE METHOD IN PALAEOECOLOGY AND LANDSCAPE ECOLOGY

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**Abstract:** The use of silicified plant microremains in palaeoenvironmental reconstructions of landcsape evolution has not been in common use yet. Not only are phytoliths bearers of significant imformation package of soil generation process and landscape evolution, but they can be utilised to reveal important questions related to archaeology and ancient human activities.

This paper intends to give a general overview of the discipline and demonstrates the use of phytolith analysis through a case study. The examination of the buried soil profiles and cultural layers of the Lyukasmound, which is situated near Hajdúnánás on the Great Plains of Hungary, resulted in novel information on the ancient vegetational patterns and environmental conditions of the Holocene. By using the analysis of these plant microfossils, questions regarding the construction and use of kurgans could also be answered.

## Introduction

#### The definition: phytoliths

The nomenclature of silicified plant microremains varried in the past centuries. From their discovery to recent days they were denominated as '*silica cells*', '*plant opal*', '*biogenic opal*', '*opal phytoliths*' and '*silica phytoliths*'.

METCALFE (1960) reports on silicified cells in the epidermal tissues of the Monocotyledons. He points out that both the short- and longcells of grass species (*Gramineae*) can be filled with anorganic silica and the taxonomic specification of these microscopic particles are significant towards the maturity of the vegetative organs. HARASZTY (1979) stresses that the silica accumulation of the grass species (*Gramineae*) are higher compared to that of the Dicotyledons. Although the term '*phytolith*' is not mentioned in his work, the form of silificied cells in the leaves are described in details. PIPERNO (1988) defines phytoliths as particles of hydrated silica formed in the cells of living plants. GOLYEVA (2001a) adds that the morphology of phytoliths resembles that of the host plant cells'.

Not only siliceous, but calcareous bodies were also called phytoliths previously (MULHOLLAND and RAPP 1992). Due to detailed studies of phytolith production, deposition and dissolution the calcium-oxalate crystals are usually not examined as taxa specific particles recently. They are not diagnostic of any plant family and their preservation in geological sediments and soils are low (PIPERNO 1988).

The main advantage of phytoliths lies in their useful qualities. The importance of phytolith analysis is affiliated with the unique attributes of these silicified plant remains,

also called biolilths, deriving from plants, which are in most cases taxa specific. They are highly resistant againts physical and chemical wheathering processes in sediments. Due to these highly important attributes they can be utilised as palaeoecological markers while reconstructing ancient environment.

## The role of silica and the phytolith generation process in plants

Silica is by far the most abundant element on Earth after oxygen. Soluble silica in the groundwater is the result of various wheathering processes of silicate minerals. The process of uptake of soluble silica by plants from the groundwater through their roots is determined by internal, genetical and external, environmental factors. Numerous soil qualities, such as pH – the amount of water in the soil – sesquioxid and organic matter content can affect the uptake in decisive manners, while climatic conditions are also regulator factors. The age and most importantly the taxanomic affinity of the plant itself plays the main role (PIPERNO 1988).

Contradictory results were published on the role of silica in plants. Japanese experts showed that monosilicic acid enters the xylem of rice shoots against a concentration gradient, (OKUDA and TAKAHASHI 1964) whereas BARBER and SHONE (1966) demonstrated with their experiments that the initial uptake of silicic acid by barley (Hordeum vulgare) root is passive, but the movement of the above mentioned material across the root into the transpiration stream is done actively by the plant. VAN DER WORM (1980) examined five different plant species and found that the uptake of silicic acid is done through active transport in sugar cane (Sacharum officiarum), wheat (Triticum aestivum) and rice (Oryza sativa). The experiments of ILER (1979) resulted in undoubted findings in terms of the importance of silica in plantlife. The resistance to pathogenic organisms has grown bigger in some cultivated plants because of more intensive silica accumulation. CHEN and LEWIN (1969) demonstrated that the silica itself is a vital element for the Equisetaceae. The statement of KAUFFMAN et al. (1985) that the essential role of silica in plantlife hasn't been proven unarguable is true, though there are loads of evidences that silica is a vital element amongst plant species and its accumulation are of certain benefits to the living organism in case of the accumulator types.

Soluble silica enters the rhizoderm in the form of monosilicic acid at soil pH 2–9 (DEBRECZENI and SÁRDI 1999). Three differet locations within the plant are suitable to deposit silica. The membrane silification is the result of cell wall deposition (*incrustation*), while the most resistant, diagnostic phytolith forms are produced through the infilling of the cell lumen. The deposition of silica in the intercellular spaces of the cortex is also known (PIPERNO, 1988). Due to the incrustation of the absorbed silica the physical and chemical qualities of the cell wall changes (HARASZTY 1979). During the plant's metabolism these incrustations strenghten the cell wall, so they grow more resistant against pressure impacts occuring during the transpiration.

So phytoliths are anorganic intra- and intercellular products of the plant. They are the result of a process by which certain living higher plants deposit silica in an intracellular or extracellular location after absorbing silica in a soluble state from groundwater. (PIPERNO 1988).

# Short historical review of phytolith analysis

LOEUWENHOEK was the first to observe calcium phytoliths in 1675 (MULHOLLAND and RAPP 1992). The discovery of opal phytoliths is connected to the work of a German scientist, called STRUVE, who reported on silicified cells from living plants in 1835 (PIPERNO 1988).

The first classification approaches were done by EHRENBERG in Berlin at the German Academy of Sciences, where he observed silicified grains deriving from soils and airborne dust (TWISS 2001). His artificial classification system consisted of four *'paragenera'*-s. With his observation that phytoliths are taxaspecific on family level, he established the theoretical principles for future researches in the field of palaeoenvironmental studies.

PIPERNO (1988) divides the history of phytolithic researches into four discrete periods. After 'the discovery and exploratory stage' (1835– ca. 1900), during the so called 'botanical phase' (ca. 1900–1936) many German experts were engaged in studies of production, morphology and taxonomy as well. The main event of this period was that the first ever archaeological applications of phytolith analysis had been carried out. During the 'period of ecological researches' (1955– ca. 1975) the center of the researches moved to the North American continent. Experts began to utilise the method as a scientific tool of palaeoecology. The first ancient flora reconstruction projects were undertaken. The morphological description of grass-producted phytolith forms were extended in decisive manners. In the 'phase of modern archaeological pedology, palaeoecology, archaeobotany, etc.) implemented the methods of phytolith analysis to gain informative data on e.g. ancient agricultural habits, soil and flora development issues of the Quaternary period.

Refering to MULHOLLAND and RAPP (1992) two types of researches are currently under way in phytolith studies. One is engaged in systematics, while the other deals with the interpretation of phytolith assemblages recovered from sediments. Palaeoecological studies have applied opal phytoliths to a wide variety of samples. Palaeoenvironmental reconstruction has been made from glacial age sediments (TWISS et al.1969) as well as more recent sediments (MULHOLLAND and RAPP 1992, KAMANINA 1997a, 1997b)

# The utility of phytolith analysis in various disciplines

The spread of a more compound view of archaeology, which implements the methodology of various interdisciplines, such as geology, geomorphology, palaeontology (palaeobotany and archaeozoology) is already traceable during the XVIII. Century, though the real breakthrough was still to come, as a result of the XIX. Century. Besides the approaches of the above mentioned disciplines the role of complex pedological examinations steadily increases (BARCZI et al. 2004; BARCZI et al. 2006). In accordance with this recognition experts began to apply phytolith analysis in palaeoenvironmental reconstructions besides the well-tried examination types, such as palynology, malacology, charcoal analysis and the analysis of other organic remains. The most significant studies in the field of the so called 'archaeological pedology' were carried out on the European Territory of Russia (ETR) (ALEXANDROVSKIY and GOLYEVA 1997, ALEXANDROVSKIY et al., 2000, GOLYEVA et al. 1995, GOLYEVA and KHOKHLOVA 2003). Researches of the above mentioned discipline in an ecological and pedological context have recently appeared in Hungary as well (BARCZI 2003, BARCZI et al. 2004a, 2004b, 2004c, Joó et al. 2003, SÜMEGI 2001; 2003).

GOLYEVA and KHOKHLOVA (2003) studied early nomad burial mounds in the Orenburg region of Southern Russia to detect anthropogenic impacts on the landscape. Pedological and phytolithic examinations were both applied to achieve the goals of their study. Their results definitely showed the human-induced transformation of soils before and during the process of kurgan constructions.

GOLYEVA (2001a) desrcibes the biomorphic analysis as the microscopic investigation of plant tissues, detritus, phytoliths, pollen and other remains of biota for the reconstruction of ancient pedogenic conditions and the evolution of soils. As every stage of soil development forms its own biomorph profile they are indicators of main vegetation changes in the Holocene. Biomorph particles can be divided into two main microscopic groups, anorganic and organic. Siliceous particles include phytoliths, spicules of sponge species (*Porifera*) and the skeleton of diatoms (*Bacillariophyaceae*). Organic particles are pollen grains, detritus and spores. As organic remains do not persist under dryer environmental conditions the analysis of the anorganic, siliceous particles may result is beneficial information. Sponge spicules and diatoms indicate soils and alluvials evoluted under hydrogenic impacts.

Phytoliths enter the soil through leaf fall and accumulte in the upper horizon of the soil, producing a record of local vegetation (PIPERNO 1988). Unlike pollen grains, the anemochor dispersion of phytolith is not so remarkable, therefore they represent the local vegetation. Though fire and strong wind erosion however, can expose phytoliths to wind transport. In comparison to pollen grains phytoliths occur within many plant parts not only in flower parts during the reproductive lifeperiod. The differences in pollen amount produced by wind- and insect-pollinated species can be a source of misinterpretation of the local and regional flora. In contrast the production of phytoliths are usually equable. The wind-induced dispersion of pollens are significant (MULHOLLAND and RAPP 1992).

MADELLA (1997) undertook the phytolith analysis of a 138 m long sequence, which consisted of an alternance of loess and 11 pedocomplexes, and a modern soil layer. With this study the expert underlined the importance of phytolith analysis of dry terrestrial environments. Not only the higher durability of these opal microfossils but the information carried by the assemblages is far more informative then other micro- and macroremains.

The vegetation changes during the Holocene on the Russian Plains were detectable by implementation of phytolith analysis and pedological examinations of buried soil profiles. The transition from Chernozems to Luvisols was accompanied by the extension of forest areas at the expense of steppelands. ALEXANDROVSKIY et al. (1999) reconstructed Holocene soil development through the pedological, micromorphological and palaeobotanical examinations of buried soil profiles of kurgans. According to their results, the steppe period ended in the middle of the Holocene and afterwards Vertic soil qualities occurred in these chernozem soil profiles due to climatic changes and terminally they changed to Luvisols. ALEXANDROVSKIY (1997, 2000) attracted attention to the importance of climatical changes when studying buried soil profiles. He found that Chernozem soils of steppelands had changed to Luvisols due to the decline of temperature and the increase of climatic humidity. Though these profiles preserved some keyfactors, like palaeocrotovina and relic humus horizons proving their Chernozem past.

GOLYEVA (2001a) showed that biomorphic analysis can provide information that is otherwise unobtainable and can elucidate some controversial questions of soil genesis and cultural changes. The expert studied three soil profiles representing different climatic subzones of the Russian Plain. Changes in vegetation and landscape due to anthropogenic impacts were demonstrable. GOLYEVA could give reasonable answer to relevant cultural changes in the region by seeking the relation between the dispersion of the early nomad Catacomba culture to significant environmental changes.

Many Russian studies were engaged in defining the biomorph and phytolith profile of genetic soil types of the ETR. With the definition of the phytolith profiles (PhP) of different soil types, a classification system of soils was generated. The phytolith profiles have been found to be characteristic of soils fomed in definite biogenocoenoses (KAMANINA 1997a). The studies of BOBROVA and BOBROV (1997) showed that phytoliths are least common in sand soils and they are most abudant in meadow soils. This statement was underlined by KAMANINA's (1997a) investigations as well. KAMANINA added that the big biolith content in meadow soils is attributed apparently to a big productivity of meadow vegetation and to a big content of diatomic algae, while the specific position of these soils in a landscape contributes to this quality too.

The phytolith analysis can be divided into quantitative and qualitative analysis. Morphology and the comparison of phytolith types from ancient and modern horizons are the subject of the qualitative analysis, while the phytolith content in different soil fractions belongs to the topic of the quantitative investigations (KAMANINA and SHOBA 1997).

# A case study: Biomorphic analysis of the excavated Lyukas-mound near Hajdúnánás

# Materials and methods

The research team from the Department of Landscape Ecology lead the excavation of the Lyukas-mound in 2004. The kurgan is situated in the Hajdúhát mesoregion of the Hungarian Great Plain. Due to disruption of the original mound body the research team seized the opportunity to collect the relevant permission for the excavation.

The cross-section wall of the kurgan was prepared to conduct pedological, archaeological and palaeoecological investigations. According to macromorphological observations discrete layers were separated on the cross-section wall and the horizons of the buried soil profile denominated. Besides other examination types, the biomorphic analysis of the layers and the buried soil horizons has been undertaken.

The sampling for biomorphic analysis was done according to PIPERNO (1988) in a vertical column. All 7 samples derive from the upper section of the layers and soil horizons. The laboratory preparation of the soil samples was done according to the standard

method (GOLYEVA 1997, 2001a, GOLYEVA and KHOKHLOVA 2003). This contains several discrete steps whilst the biomorph content of the soil is separated from different soil fractions. The samples are treated with hydrogen peroxide to destroy organic matter (humus). To remove the clay fraction from the samples gravity sedimentation was utilised, then they were filtered through a 0.25 mm mesh sieve. The final step is to centrifuge the samples with a special heavy liquid adjusted to a specific gravity of 2.2–2.4.

After the separation method was completed, a drop (0.5 ml) of each specimen mixed with the same amount of additional glycerin was examined under optical microscope. A magnification ranging from 350x to 700x was applied for the optical observations. The classification system developed by GOLYEVA (2001b) was used to identify and group phytoliths and other biomorph particles. The softly modified, grouped and standardized version of the classification system is presented below (Table 1.). This type of classification groups the indicator forms on the purpose to detect the association type of the ancient environment. It does not focus on species – as identification of ancient phytolith assemblages is usually not possible on species level – but characterises the ancient flora and provides additional information on the environment in terms of its climate and soils.

Denomination	Comment
Main indicator types of the classification system	
Indicator types of coniferous species	Phytoliths of <i>Picea</i> and <i>Pinus</i> spp. Cubic formed particles with peats on their surface.
Trichoms of grasses of forest habitats	Relative big $(30-50 \ \mu m)$ with long base. The apex of the trichom does not extends the basis.
Trichoms of grasses of meadow type habitats	Smaller with round base. The apex of the trichom is long, pointed and extends the base.
Indicator forms of grasses of steppeland habitats (epidermal origin)	Remarkably small (10–15 $\mu$ m) with special rounded end on both side. (Figure 3.)
Indicator forms of grasses of extremely dry, semidessert-like habitats (epidermal origin)	Relative big (20–40 µm) epidermal longcell phytolith with comb-like spine along its body. Typical of grass species inhabiting dry, semi- dessert habitats. A type of xeromorphism.
Reed phytolith	Big size (50–100 $\mu$ m). Nearly cubic formed, with one concave facet. It is a product of the joint of <i>Phragmites australis</i> .

Table 1. Indicator forms of the classification system 1. táblázat Az osztályozási rendszer indikátor csoportjai

Denomination	Comment
Other phytolith forms	
Epidermal longcell phytoliths	All grass species (Gramineae) produce
	longcell phytoliths in their epiderm.
	It varies in a broad range. The amount of these
	forms indicate the biomass production of a
	surface. (Figure 4.)
Phytolith of moss species	Very small (8–10 µm), spherical shape
	(Figure 1.).
Other siliceous indicator types	
Spicules of sponge species	Elongated form with a tubular central channel
	(Figure 2.). Deriving from <i>Porifera</i> species.
Diatoms	Deriving from the <i>Bacillariophyaceae</i>
	classis. Their shape vary on a broad range
	from the oval types to the triangular ones.
Other non-siliceous vegetable indicators	
Grass detritus particles	Organic microscopic remains of grass species
	(Gramineae)
Arbaraal datritus partialas	Organia miarosconia ramaina of trae anacias
Arborear detritus particles	organic incroscopic remains of tree species.

Contd. Table 1. 1. táblázat folytatása



Figure 1. Spherical moss phytolith 1. ábra Gömbölyű moha fitolit

*Figure 2.* Broken spicule of sponge *2. ábra* Egy szivacsfaj törött tüskéje

### Results and their discussion: Palaeoecological, pedogenetical relations

The reconstruction of the ancient flora and environment in the case of the Lyukas-mound is hardly achievable through palynological methods as the preservation of pollen grains in dry terrestrial environments is low. Therefore the biomorphic analysis is a suitable method in this case to obtain data on the palaeovegetation.

In accordance with the results of the biomorph (phytolith) analysis and the pedological examinations the kurgan was erected on a Chernozem type soil formation. This profile can be divided into 3 separate horizons. The lowest one is a structureless, loessy parent material (BARCZI et al. 2006). The biomorphic analysis provides additional information on the evolution of this layer. The presence of diatom and sponge spicules particles is the evidence of hydromorph evolution. On the basis of these facts several theories are conceivable. Either the airborne dust resettled on a watery surface or the postgenetical effect of flooding water was significant to a certain extent. The presence of a few intact diatom forms indicate a partly or periodically submerged evolution of the deposit. The second horizon is characteristic of Chernozem soils. By virtue of the palaeoecological investigations this layer has never formed the surface before. The phytolith distribution and content shows a typical [B]-horizon. The greater abundance of grass detritus remains (5-10%) and the traces of arboreal detritus (1-2%) marks the closeness of a once actual surface. Not more than 5-10% of the counted and identified grains proved to be elongated phytoliths. Most of the observed specimen had a significantly corroded surface, which reflects the first stages of soil development and shows their ancient matter. The most interesting of all was the sample deriving from the uppermost section of the palaeo [A]-horizon. Besides the high, elongated form content (>20%) a huge amount of grass detritus (10-20%) was indentified. Practically all elongated phytolith forms were corroded on their surface, whereas the steppe indicator forms (Figure 3.) appeared to be more intact. If we suppose that steppe phytoliths are younger due to their condition, whilst the others are older and destroyed, a vegetational change from a meadow dominated, more humid landscape to a xeromorphic, arid steppetype can be visualised. Though this is not the only explanation. In case of special microrelief conditions of the surface, mosaics of meadow type and a steppeland vegetation could have inhabited the former surface simultaneously. When featuring the palaeoenvironment the fact of human impact must be also taken into account. In the case of the Lyukas-mound the influence of early nomad cultures could have been significant as the region was inhabited at the time. The quantitative and qualitative observations of the plant detritus deriving from this layer needs further explanation. On one hand the amount of vegetable detritus indicates a surface that can be characterised of high biomass production. While on the other, the participation of arboreal detritus particles among the total detritus content indicates that arboreal species were determining components of the ancient flora that vegetated here in the past. On the whole, the biomorphic analysis resulted in the conclusion that dry, steppe-grassland vegetation with discrete tree species dominated this part of the surrounding landscape at a time when the inhabitants of the region began to erect their kurgan.

On the basis of the layer classification of the mound's cross-section wall, the multistage construction of the burial mound is presumable. Alltogether 6 layers were separated besides the 3 horizons of the buried palaeosoil profile.



*Figure 3*. Steppe phytolith form *3. ábra* Sztyeppei indikátorforma

*Figure 4*. Large epidermal longcell phytolith *4. ábra* Nagy méretű epidermális hosszúsejt fitolit

The most informative results stem from the topsection of the second mound. The quantitatvie analysis showed that this has been a surface for a longer time. This statement is underlined by the high amount of phytoliths and plant detritus pieces (Figure 5., PETŐ 2006).

One of the most interesting qualitative observation is in connection with the size of the epidermal longcell phytoliths stemming from this cultural layer. The significantly bigger size of these silicified plant remains are characteristic of pioneer flora. The species that newly inhabited the raw mound surface could produce larger vegetative organs because of the lack of competition (Figure 4.). This event is demonstrable through the biomorphic analysis. Arboreal detritus pieces were also found in the sample. Significant amount of the epidermal longcell phytoliths show xeromorphism, which is an indicator symptom of a dryer environment. Arboreal detritus pieces were also found in the sample. Summarizing the findings the vegetational conditions of the second mound shows resemblance with that of the surrounding territories and with the conditions that were demonstrated from the sample deriving from the palaeo [A]-horizon.

One of the most interesting section was found atop the second cultural layer. The origin of this thin (*ca.* 20 cm) layer is unexplained, unclarified at the recent stage of our researches. What we can tell by virtue of the biomorphic analysis is that this layer is of shortage of biomorphs (phytoliths). The epidermal longcell phytoliths were in all cases smaller than it is expected. There is a perceptible decrease in the percentage of some particles in this sample. Not just a part of grass detritus particles, but the elongated phytoliths have also disappeared in this layer, indicating a decline in biomass production. Light microscopical investigations made it clear that not only quantitative, but qualitative changes occur, which are related with the small size of elongated phytolith forms indicating suppressed vegetation. In case of vegetational degradation, plant species are "forced" to produce smaller phytoliths in their tissues of diminished vegetative organs. Causes of such degradations are likely to be related with human influences on the landscape, like overgrazing. One theory is that the generation of this layer may be the result of an increased soil development process, which was presumably interrupted by anthropgenic impacts.



Figure 5. Vertical biomorph distribution along the cross-section wall of Lyukas-mound kurgan (PETŐ 2006 modified)
5. ábra Vertikális biomorf eloszlás a Lyukas-halom metszetfala mentén (PETŐ 2006 nyomán módodsítva)

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### AZ ŐSKÖRNYEZETTANBAN ÉS TÁJÖKOLÓGIÁBAN ALKALMAZHATÓ MÓDSZERRŐL: A FITOLIT ELEMZÉS

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Kulcsszavak: fitolit elemzés, őskörnyezettan, paleotalajok, kunhalmok, eltemetett talajszelvények, őskörnyezettani rekonstrukció

Összefoglalás: A tájfejlődés őskörnyezettani szempontú rekonstruálásában még nem rutinszerű módszer a kovavázas, növényi mikromaradványok elemzése. A fitolitok nem csak a talaj- és tájfejlődés emlékeit hordozzák, de elemzésüket felhasználhatjuk egyes régészeti kérdések megválaszolására és ősi antropogén hatások kimutatására egyaránt.

Jelen tanulmány egy általános képet kíván festeni az említett témakörről és annak hátteréről, illetve egy esettanulmányon keresztül mutatja be a módszer alkalmazhatóságát. A Hajdúnánás határában emelt Lyukashalom eltemetett paleotalajának és kultúrrétegeinek vizsgálata újfajta adatokat szolgáltatott a holocénbeli őskörnyezet megismeréséhez. Ezen növényi mikrofosszíliák analízise folytán a halom építésére és használatára vonatkozó kérdésekre is választ kaptunk.