Volume 63 155 Number 4, 2015

http://dx.doi.org/10.11118/actaun201563041417

SOIL SCIENTIFIC RESEARCH METHODS USED IN ARCHAEOLOGY – PROMISING SOIL BIOCHEMISTRY: A MINI-REVIEW

Valerie Vranová¹, Theodore Danso Marfo¹, Klement Rejšek¹

¹ Department of Geology and Soil Science, Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic

Abstract

VRANOVÁ VALERIE, DANSO MARFO THEODORE, REJŠEK KLEMENT. 2015. Soil Scientific Research Methods Used in Archaeology – Promising Soil Biochemistry: a Mini-review. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis*, 63(4): 1417–1426.

This work seeks to review soil scientific methods that have been used and are still being used in archaeology. This review paper aims at emphasising the importance of soil science practice to archaeology thus adding a scientific analytical nature to the cultural nature of archaeology. Common methods (physical, chemical and biochemical) used to analyse archaeological soils and artefacts is touched on and their strengths and shortcomings duly noted to become the base for future research. Furthermore, the authors made emphasis on distinctive excavating/sampling methods, biochemical analyses focused on distinctive features of plough-land and soil organic matter mineralization, Counter Immunoelectrophoresis (CEIP) method by the presence of proteins testing, carbon analyses such as carbon-14 dating techniques, soil phosphorus studies and geochemical analyses of hematite Fe_2O_3 and cinnabaryte HgS contents. It is obvious that, the future of archaeology is in the soil because the soil harbours information of the past hence the synergy between soil and archaeological research has to be strengthened and archaeology made a prime agenda by soil scientists by expanding the analyses scope of total phosphorus extraction and giving attention to soil magnetism.

Keywords: soil properties, soil biochemistry, phosphorus starch, lipid, protein

INTRODUCTION

Soil biochemistry basically has its starting points from carbohydrate compositions, characterisation of their biotic origins and their isolations (Paul, 2014). The theory itself comes from the biochemical nature of changes in microbial biomass carbon, contrasting plant carbon, and animal carbon. A forward march of this part of soil biochemistry will be, thus, focused on the interrelationships between soil carbohydrate composition and soil physics leading to understanding both origins and stability of soil saccharides, and soil organic matter as a whole. For that reason, both mobility of organic polymers, their aliphatic, cyclic and aromatic nature, and the isotopic composition of soil organic matter related to human settlements, vegetation changes and natural soil processes have been scrutinized in the mirror of archaeological records. Therefore, the objectives of the paper include both a selection of soil scientific research methods used in archaeology and a discussion of their interpretative potential.

A lot of people have described archaeology in several ways for its 150 years of study (Daniel, 1950; McGlade and van der Leeuw, 1997). The differences in the descriptions can largely be pinned on the changes in the focus of the subject matter. Archaeology is becoming more and more associated with science and taking into account human actions. Basically, descriptions or definitions given to archaeology depict the perception the authors have towards the subject matter. In a nutshell, archaeology is more of an investigative research, which seeks to uncover what was done by humans that once lived in an area (Sumner, 1999). The Society of American Archaeology describes archaeology as a field of anthropology which analyses the physical remains of ancient and recent past lives to understand human culture (Reyman, 1992). On the other hand, the soil is heterogeneous

and generally complex to study. For this reason, soil is studied in a lot of disciplines (examples are; forestry, agriculture, geology, geography, chemistry etc). This has led to the development of so many methods to studying various concepts within the soil. Again, the soil is seen by these disciplines in different lenses. It is seen mostly as a sediment or growth medium for organisms (geologists see the soil as sediments, pedologists are more concerned with soil formation whereas agriculturalist and foresters are concerned with its productivity). Archaeology will bring another dimension because all the above would be interpreted from a human cultural angle. Although archaeologists, most of the time dig through soil layers for artefacts to get an idea of what happened on the site so many years ago, the soil itself can also be an important storehouse of information hence soil analysis (physical, chemical or biochemical) can help archaeologists to date sites and get to know the major activities the people there did (e.g. soil analysis on fertility can help archaeologist understand the farming systems practiced in ancient times).

The Scandinavia has closely linked archaeology to geology, dating and agriculture. An archaeologist by the name, J. J. A. Worsaae (1821–1886) from Denmark successfully resolved the mysterious sediment formation on the sea shell middens and since then archaeology in Scandinavia has always had strong association with geology (Briggs, 2005). Due to the increased interest in the economics of ancient times and landscape, there have been heightened interests in soil within archaeology (Sherratt, 1997). These connections led to the development of environmental archaeology and geoarchaeology (Butzer, 1971; Brown, 1997). The book "Soil science in Archaeology" written by Susan Limbrey was a great contribution to wed soil and archaeology together (Mackney, 1976). This giant step made soil micromorphology a vital part of soil study in archaeology and it is still developing (Courty et al., 1990). It is clear that in the early stages of archaeology practice in Europe, pedology seemed strong. In America, Karl Butzer gave an ecological angle to archaeology and this later became known as geoarchaeology (Butzer, 1980). Although the geology angle of archaeology in America seemed stronger in earlier times, the pedology angle has never been out of the scene. Soil erosion has since the beginning of soil study been seen as the greatest soil degradation factor hence soil erosion became an integral part in geoarchaeology (Boardman and Poesen, 2006; White, 2013).

From the above, it is obvious that efforts have been made in the past to link archaeology and soil analysis. This mini-review will throw more light on the need to properly merge soil scientific methods especially soil biochemistry with archaeological practices. The synergy between these two fields of study will be beneficial to both and most importantly lead to a more realistically precise archaeological predictions.

Soil Physical Analysis in Archaeology

Perfect description of soils helps archaeologists to understand what was done on a particular site in the past (Holliday, 2004). The most used physical features of soil in archaeology are soil structure, aggregate stability, colour and texture (Moore *et al.*, 1993). These are primarily used to date archaeological sites.

Soil Structure and Aggregate Stability

Soil aggregates are categories of soil particles that bind to each other more tightly than to adjacent particles. Aggregate stability is the ability of soil aggregates to resist collapse when subjected to forces such as those associated with tillage as well as water or wind erosion. Aggregate stability is a sign of organic matter content, biological activity as well as nutrient cycling processes in the soil (Hrabovska et al., 2014). The age of soil has influence of its structure. The older a soil is, the stronger the aggregates and younger have weak aggregates hence archaeologist can use this general age rule to some extent when it comes to dating of sites. Additionally, the stability or instability of soil aggregates gives archaeologists a hint of other factors of interest such as carbon, nitrogen or phosphorus.

Colour

A geochemical analysis of hematite Fe₂O₃ and cinnabaryte HgS contents focused on their accumulation as remnants of usage of pigments for some human rituals and funerals as well as their combined colour effect for the production of red ceramics makes their investigation a crucial soil scientific research method needed in archaeology. Top soils of ancient times are mostly not preserved in-situ but at most archaeological excavation areas or sites. Ancient pits filled with former topsoil is the common practice (Eckmeier and Wiesenberg, 2009; Lauer et al., 2013; Lauer et al., 2014). Soil colour is very important to archaeologists because it is a key determinant of age (Jones and MacGregor, 2002). In North-West Germany, a dark brown soil filling represents Neolithic period, grey-brown represents the Iron Age and light grey represents the Roman era. Soil colour as a dating parameter is very crucial because in areas where no artefacts are found, it becomes the only factor there is to date the site. The Munsell Soil Colour Charts as commonly practiced in soil science is also used to some extent by archaeologists. Its description of soil colour limits the dating and quantitative nature of archaeological practice. For instance, not every possible colour can be found and light exposure generates several perceptions of colour. Spectrometry presents a fast and precise way to determine soil colour (Torrent and Barrón, 1993). Soil colour and its chemical properties can be measured with this method because spectrometry produces values that are possible to analyse statistically. Soil forming processes as well as other soil features produces

the colour of soil material hence soil colour is a sign or indicator of what materials were present in soils from prehistoric to even modern times (Barrett, 2002; Rossel et al., 2006). Addition and removal of some particular compounds characterises human influence in soil formation. Benzene polycarboxylic acid molecular method is mostly employed to spot charcoal carbon composition in soil (Hammes et al., 2007). Eckmeier and Gerlach (2012) tested soil spectra measurements in two regions northwest Germany (Luvisols) and central Germany (Chernozems). Pit fillings from Neolithic to Iron Age were sampled. After the analyses, a key conclusion they came to was that though UV/Vis (Ultravioletvisible spectroscopy) presents a fast way of analysis, it can only be used after the method has been calibrated. Furthermore, the difference in colour of soil for the different archaeological periods was clear but could not be explained (Eckmeier and Gerlach, 2012).

Soil Texture

Distribution of particle size is very important to archaeologists. This is because, it is a stable soil property and it is used to assess the level of soil formation and spot imbalances in soil profiles. Particle size analyses encompasses the determination of soil separates greater than 2 mm thus sand (2-0.05 mm), silt (0.05-0.002 mm) and clay (< 0.002 mm) (Klute, 1986). Soil texture analysis can be done as follows; sieving when separates are big thus greater than 0.05mm and hygrometer or pipette method for smoother separates < 0.05 mm (Bouyoucos, 1962). Soil textural analyses were used to compare the degree of pedogenesis of two different pedons of Busia in Western Kenya. The outcome of this research showed that both pedons had clayey subsoil but the first had a sandy clay top soil and the second, sandy clay loam. The silt clay ratio of both pedons was very low hence both pedons represents a highly weathered materials and the pedon with sandy clay top soil being the more weathered of the two since its silt/clay ratio was smaller (Kenbeney et al., 2014).

Soil Chemical Analysis in Archaeology

Phosphorus: Phosphorus analysis of soils has been the most explored by archaeologists when soil science is concerned (Leonardi et al., 1999). Phosphorus analysis is an indication of past human activity and it is frequently employed in archaeological research. High amounts of phosphate in soil is a clear sign that those that once lived there, probably used the site for disposal of refuse with high organic matter content from human addition of manure, food residue, animal remains, urine etc. (Conway, 1983; Schlezinger and Howes, 2000). Soil phosphorus analysis takes into account two basic components; extraction of soil phosphorus and measuring the amount of phosphate in extract (Provan, 1971). The principle of extraction is to break the molecular bonds phosphorus forms with other substances using reagents. The amount of reagent added has a bearing on its capacity of extraction and the reagent's strength is measured by how easy it dissociates in solution. A lot of archaeological interest in soil phosphorus has been on soil available phosphorus (Menon et al., 1989). This might be so because there are a lot of simple on-field methods to measure soil available phosphorus but essentially, the main reason is that, agriculturists found it very important. Almost all the early analysis on soil phosphorus in archaeology was on available phosphorus. Available phosphorus measurements were developed to measure nutrient availability to plants hence it was mainly for the agricultural sciences (Bray, 1929). There are a number of problems associated with how methods of analysing available phosphorus is interpreted hence might not be the ultimate indicator for archaeological purposes. It should also be noted that different plants take-up different quantities of phosphorus from the same soil hence available phosphorus will only roughly say something about phosphorus status in the soil and not necessarily relate to phosphorus existing in nature (Dietz, 1957).

As stated earlier, a lot of the soil phosphorus research in archaeology uses methods easily applied on the field. The ring or spot test is a vivid example and it is used to test for easily removable available soil phosphorus (Eidt, 1973). Nitric acid was originally used for this method but later hydrochloric or sulphuric acids were used because it gave better results. The main drawback of this method is that it only gives qualitative data which is not easily reproducible hence not so reliable. Though the ring or spot test method is simple and can be done on-field, the above mentioned drawback has pushed some archaeologists to reject it completely (Bakkevig, 1980). A semi-quantitative method which employs the use of colorimetry was developed. This is similar to the spot and ring test method but its added advantage is that it is more quantitative (Watanabe and Olsen, 1965). Rypkema et al. (2007) also reported the development of modern soil phosphate analytical method which employs the use of spectrometer, it is simple, can be done on-field in just 6 minutes and fully quantitative. The only drawback is that the set-up is expensive and results are not easily explainable (Rypkema et al., 2007). Total phosphorus analysis became the focus of archaeologist but because of its complexity, research on total phosphorus was minimal (Dietz, 1957). The most used total phosphorus methods are; digestion in HClO₄, fusion with Na₂CO₃ as well as sequential digestion in H₂SO₄, H₂O₂ and HF (Proudfoot, 1976). Total phosphorus measurements gives quantitative data that can be compared to each other unlike available phosphorus hence for now it might be the best indicator of human phosphorus inputs. Lendakova and Grigelova (2012) made a strong case for the use of concentrated nitric acid followed by photometry to determine total phosphorus content in archaeological samples. They came to

this assertion when they conducted phosphate analysis at the Olomouc-Nemilany archaeological site. The prominent positive features reported in their publication titled "Phosphate analysis of sediment from the archaeological site Olomouc-Nemilany" were its time and cost effectiveness but the authors could not ascertain the method's reliability considering the fact that it is currently not commercially employed. The major drawback of total phosphorus measurement is that, it includes all of inorganic phosphorus which is much higher than human phosphorus input (Holliday and Gartner, 2007). This drawback will affect areas with high levels of natural phosphorus.

The first method employed to extract phosphorus, mainly available phosphorus is based on citric acid (2%). The authenticity of this method has been questioned since the huge amounts of phosphorus extracted may possibly include other forms of phosphorus (organic and inorganic phosphorus). Ruttenberg (1992) and Leonardi et al. (1999) did some comparative analysis of some phosphorus extraction methods in the context of archaeology and their conclusions were as follows: i) the difference existing between total phosphorus measured by fusion and that measured by perchloric acid digestion increases as the ratio of coarse sand in the soil also increases; ii) organic and total phosphorus were analysed and compared. Both were good for archaeological analysis but the latter is faster and cheaper; iii) perchloric acid extraction of total phosphorus generated the closest correlation with man influenced soils. 5 β-stanols has proved to be a useful biomarker for soil analysis in archaeology. It gives a clear picture of the past manure application unlike total phosphorus (Evershed, 1997; Ernée and Majer, 2009; Birk et al., 2011; Prokeš et al., 2013).

Calcium

Detailed role of soil calcium as an important component of identifying food preparation areas of the past, its presence in the ash of burning charcoal and its high content in teeth and bones makes its investigation a crucial soil scientific research needed in archaeology. Middens are mainly the sites for bone and ash disposal and these two materials are composed of significant amount of calcium. As time wear on, the calcium from these middens becomes part of the soil matrix hence could be measured. Carr (1982) suggested that higher calcium levels on a site is an indication that the site was once a garbage or refuse dumping ground hence areas kept clean will have lower calcium levels. Systematic sampling of a site should that matter record markedly higher calcium content areas and marked as middens that once had a lot of bone and ash. Sullivan and Kealhofer (2004) in their archaeological research spotted marl and shellfish leftovers using calcium analysis. Cook and Heier (1965) found out that, calcium, carbon and nitrogen were almost always found at the centre of the sites under archaeological investigation. This brings to bear that, calcium levels in archaeological soil suggests the level of past occupation of the site.

Calcium analyses has been another soil scientific research focus for archaeologists due to the importance of both accumulation of primary/ secondary carbonates and calcite micrite. Retallack (2001) as well as Schaetzl and Anderson (2005) made a summary of the distinctive roles of carbonates and calcium silt as the interfaces of pedology with the other natural sciences.

Carbon

Carbon analysis is most of the time applied as C-14 dating. Carbon dating is used to establish the age of ancient organic materials and the features they contain. Human activities play a vital role in the formation of soil organic matter. Soil organic matter found in sites of previous human habitation has higher carbon thus about 2 times more than sites with almost no human habitation historically (Holliday, 2004). Analysing soil organic matter requires a systematic collection of soil samples from a potential site. Non-soil materials such as stones and roots are removed, air-dried, burned in a regulated temperature for a time period after which the carbon percent is ascertained considering the pre and post burning states.

In terms of the role carbon studies play as a soil scientific research method used in archeology, the authors reinforced the individual determinations of four groups of carbon compounds: i) plant carbon, ii) microbial and biomass carbon, iii) stabile carbon forms, and iv) labile carbon forms. With respect to carbon-14 dating techniques, Aitken and Martin (2003), Schoeninger (2010) and Taylor as well as Bar-Yosef (2014) present a comprehensive review of both particular dating methods and soil age determinations.

Nitrogen

Human occupation leaves a great deal of nitrogen (Rapp and Hill, 2006) but nitrogen analysis just like calcium analysis is not as reliable as phosphorus analysis for archaeology hence there are just a few available literature on nitrogen and calcium analysis for archaeology. Holliday (2004) notes that nitrogen is relatively more volatile hence its analysis may be of no use on very old sites. The detailed role of nitrogen can be found in the chapter which focuses on soil biochemistry.

Other Elements

Trace metals content in the soil can also give an indication of what happened years back on the site under investigation by archaeologists. Trace metals can be analysed using the following methods; nitric-perchloric digestion, chelate extraction (effective on calcareous soils), acid extractants and magnetic susceptibility (Parnell *et al.*, 2002). Barium and magnesium detection in soil of sites under archaeological investigation indicates high level of organic refuse or garbage disposal. Lead

and Mercury detection is a sign that the major occupation at the site under investigation was predominately craft production (Alloway and Alloway, 1995).

Information hidden in the chemical traits of archaeological soils are most of the time ignored during archaeological investigations but indepth analysis of soils can answer a lot of crucial archaeological and environmental questions posed. Activities of human settlements can easily be detected by the increased levels of plant available phosphorus, calcium, magnesium, zinc and copper in archaeological soils (Hejcman *et al.*, 2013).

The chemical traits of human activities are well kept in fills of sunken subsoil archaeological features juxtaposed with surrounding control samples (Hejcman et al., 2013). Soil depth, rock particle constituent, soil pH, soil texture, soil temperature and soil moisture are crucial soil properties that help in bone decomposition and in finding secret or clandestine graves (Surabian, 2012). Soil pH presents the best clue in detecting grave soils since it affects adipocere formation. Adipocere slows down decomposition. Adipocere formation which is supported by mildly alkaline soil becomes abundant as time wears on (Gordon and Buikstra, 1981). The odour produced by the adipocere is easily detected by cadaver dogs (Rebmann, 2000). The adipocere which happens as a result of anaerobic bacterial hydrolysis of fat in tissue replacing putrefaction of cadaver with a cast of firm fatty tissue in the soil distinguishes grave site soils from cenotaphs and other human settlement sites.

Soil Biochemical Analysis in Archaeology

Analyses of microscopic archaeological remains of biological nature is on the increase nowadays. The main ones of archaeological interest are starch, proteins and lipids.

Starch

It is the main food store of plants (Badenhuizen, 1965). Archaeological analysis takes into account two main starch forms classified based on their function and location. These are transitional starch found mainly in leaves and storage starch found mainly in seeds, roots, fruits etc. (Barton, 2007). Starch is made up glucose molecules and complete granules are not soluble in cold water. The character of starch which is of most interest to archaeologists is how it behaves under wet and heat conditions as well as being chemically stained. Under wet conditions starch imbibes water and swells up but returns to its initial shape when it dries up. This happens only when swelling was not too severe and when the temperature is kept at an optimum level. Otherwise the swelling is irreversible and this situation is termed gelatinisation. Microscopic plant remains have been found on Acheulean artefacts from prehistoric time to modern times hence it is obvious that these remains can survive for a really long time. The most widely used technique for starch residue identification is light microscopy (Olsen, 1988). This method in some cases provides identifications chemical analyses are unable to give but the latter has an advantage when it comes to morphological microscopic research.

Protein

Proteins can be found in plant tissues and all body fluids and tissues e.g. urine, blood, faeces etc. Test of protein remains on artefacts and in soils is done by the Counter Immunoelectrophoresis (CEIP) method. This method is based on the reaction between an antigen and an antibody (Shanks et al., 1999). In this case the antigen is the protein remains on the artefact or in the soil. It is also essential to test soil controls to prevent false positive results generated by bacteria and faecal matter of animals. This method is widely used by archaeologists world-wide. It was used by the Laboratory of Archaeological Science of Califonia State University in the United States of America to test 25 artefacts and 2 soil samples but the results generated 23 negative readings. This can be pinned on several reasons such as insufficient protein residue on or in sample or no identifiable protein residue but in the broader sense, it highlights the imprecise nature of the method hence the need to expand the scope of the method (Yohe and Gibbons, 2013).

Lipid

Lipid is the general term for compounds that cannot dissolve in water. For Archaeologists, the lipids of interest are fatty acids, sterols, waxes, triacylglycerol and terpenes. Lipids have high stability in higher temperatures and its decomposition in temperatures for cooking is small compared to starch and proteins (Fahy et al., 2005). For over 40 years, various techniques have been used to get information about lipid residues in artefacts and soils for archaeological purposes. The most common is the Gas chromatography (with flame ionization detector, mass spectrometry or gas chromatography combustion/isotope ratio methods) (Evershed, 1993). An advancement of lipid identification analysis is the development of biomarkers to give precise results over the last twenty years (Evershed, 2008). This analysis is mainly based on fatty acid composition. Fatty acids give a general archaeological character of residue but it is not able to help trace the exact source that is they can only be classified as of a plant or animal source.

Saccharides

Cheshire (1977) stated that the application of saccharide studies for the broader usage reinforcing that a stability of polysaccharides in soil is not related to their chemical composition but to their unavailability. Such unavailability is caused by either inaccessibility within undecomposed biological residues and/or insolubility resulting from adsorption onto clay. Methodologically, Ball

et al. (1996) gave a description of the differences between the carbohydrates obtained by hot-water extraction and those extracted by acid hydrolysis from the viewpoint of its practical usage. The usage of such studies has to include the division between a specific composition of soil saccharides of plant (Chao et al., 2007) and a specific composition of soil saccharides of microorganisms (Rakhuba et al., 2009). An understanding of these differences would allow us to detect the specific features of the archaeological records.

Soil magnetism as an investigation tool for archaeologists failed to keep pace with current analytical research but it still remains very relevant to archaeology. This method comes in handy if both erosion and sedimentation processes are to be analysed (Singer and Fine, 1989; Blundell *et al.*, 2009).

Soil biochemistry as a whole seems to be an important tool as well (Craig and Collins, 2000, 2002; D'Anjou et al., 2012; Vranová et al., 2012; Vranová et al., 2013a). This is because it adequately takes into account the following; mineralisation processes, decomposition processes, enzymatic processes and soil organic compounds composition, soil polysaccharides and lipids in particular. First of all, soil organic matter mineralization (Balesdent et al., 1988) including microbial carbon (Cmic) and the metabolic quotient, and soil microbial decomposition (Child, 1995; Haslam, 2004) can be studied effectively. The results of such investigations manifest through the appearance of simple inorganic substances, soluble in the soil solution, differences between naturally occurring and anthropogenic complex organic compounds. Furthermore, such results can be supported by the appearance of simple organic substances, insoluble in the soil solution. Secondly, soil enzymology may be a very promising soil research for archaeology (Dick et al., 1994), especially for phosphomonoesterases (Rejšek et al., 2012), ureases (Formanek et al., 2006) and proteases (Vranová et al., 2013b). This is mainly because of their direct interrelationships and the fact that they remain unchanged after speeding up specific biochemical reactions resulting in formation of new products at the end of the reactions. Turek et al. (2015) presented results dealing with the ratio(s) between galactose/mannose and arabinose/xylose (the GM/ AX ratios) as a method of allowing a differentiation among soil organic matter (SOM). The presence of soil polysaccharides can be evaluated from the viewpoint of a plant stock storage by humans versus an accumulation by natural presence of soil microbial communities. Again, lipids of animal origin such as cholesterol (Hjulström and Isaksson, 2009) in soil samples can serve as another evidence of past human beings settlements. An advantage of soil lipids and associated compounds determinations can easily be substantiated by understanding the sources for waxes (residues of inhibited biological activity), carotenoids (plant pigments) and resins (wood of coniferous tree species).

Biochemical analyses can be combined with other soil scientific investigations. In particular, studies focused on distinctive features of plough-land can be reinforced. Cultivation to the depth of 25 cm, deep ploughing to the depth of 50 cm and layering of organic compounds after a speedy deforestation to colluvial deposits are proposed as promising soil research areas for archaeology.

Excavating or Sampling Recommendations

For the synergy between archaeology and soil science to be first and foremost feasible, the quality of sampling should be high. Adequate sampling can only be done when the questions that require answers are brought up in advance. Archaeological sampling involves different strategies than other purposes of soil sampling such as for soil fertility, soil genesis, etc. Before the excavation is done, the site must be apportioned into sampling or excavation units which should cover the whole site and should not overlap. Most of the time, the choice of units is not clear-cut. Non-Invasive methods such as field-walking followed by aspects of invasive methods such as test-pits may seem the most logical approach of sampling but this approach is not time efficient. Purposive sampling which involves digging targeted areas with already identified possible features or probabilistic way of sampling, if not so much is known in advance about the site. If purposive sampling is employed, the shape and size of the excavation units are likely to be defined by the nature of the visible evidence that points to their location. When it comes to probabilistic sampling, the choice of excavation units is usually either 2 meters wide by digging machine and mostly 30 meters long, or in the case of hand-dug testpits, usually 1 or 2 meters square. Trenches are of a flexible design, cheap, good when it comes to the detection of features but are destructive and have a poor recovery rate for archaeological finds. Testpits are good in the detection of archaeological finds and are generally not destructive but labourintensive hence expensive but highly recommended (Richardson and Gajewski, 2003).

CONCLUSION

It is evidently clear that the soil represents a great storehouse of information for archaeology hence the connection between the two fields thus soil science and archaeology should be tightened. Further research should be done to drastically minimize the main drawbacks of soil total phosphorus analysis. Soil magnetism should be deeply explored as it holds the key to reliable archaeological investigations.

Evaluating the quantity, quality and biodegradability of soil organic matter as influenced by either their plant or microbial origins leads to distinctive features of the archaeological records where inconsistencies in their interpretations can be effectively treated. The prehistoric alterations in the local archaeological soil body profiles documented by archaeological and biochemical evidences and related to microbial and biochemical changes induced by paleoenvironmental amendments and settlement dynamics rotations can be studied from soil scientific points of view. Biochemical analyses of archaeological sediments formed under different ancient activities and prehistoric human influence add considerable evidence for understanding prehistoric patterns in ancient landscapes and their properties.

In terms of future research, the authors will like to devote their effort to focus on the following specific themes i) artificial metal contents in settlements/graves, ii) biochemical methods applied for studies of entombment with graves and without coffins, and iii) biochemical methods applied for studies of soils in settlements and soils in graves.

Acknowledgement

This study was supported by the Grant Agency of the Czech Republic (Grant No. GA15-02453S).

REFERENCES

- AITKEN, M. J. 2003. Radiocarbon dating. In: *Archaeological Method and Theory*. ELLIS, L. (ed.), pp 505–508. New York: Garland Publishing.
- BADENHUIZEN, N. 1965. Occurrence and development of starch in plants. In: *Starch: Chemistry and Technology.* WHISTLER, R. L. and PASCHALL, E. F. (eds), vol. 1, pp 65–100. London, New York: Academic Press.
- BAKKEVIG, S. 1980. Phosphate analysis in archaeology problems and recent progress. *Nor. Archaeol. Rev.*, 13(2): 73–100.
- BALESDENT, J., WAGNER, G. H. and MARIOTTI, A. 1988. Soil organic matter turnover in long-term field experiments as revealed by carbon-13 natural abundance. SSSAJ, 52: 118–124.
- BALL, B. C., CHESHIRE, M. V., ROBERTSON, E. A. G. and HUNTER, E. A. 1996. Carbohydrate composition in relation to structural stability, compactibility and plasticity of two soils in a long-term experiment. *Soil Till. Res.*, 39(3–4): 143–160.
- BARTON, H. 2007. Starch residues on museum artefacts: implications for determining tool use. *J. Archaeol. Sci.*, 34(10): 1752–1762.
- BIRK, J. J., TEIXEIRA, W. G., NEVES, E. G. et al. 2011. Faeces deposition on Amazonian Anthrosols as assessed from 5β-stanols. *J. Archaeol. Sci.*, 38(6): 1209–1220.
- BLUNDELL, A., DEARING, J. A., BOYLE, J. F. et al. 2009. Controlling factors for the spatial variability of soil magnetic susceptibility across England and Wales. *Earth Sci. Rev.*, 95(3): 158–188.
- BOARDMAN, J. and POESEN, J. 2006. Soil Erosion in Europe. In: Soil erosion in Europe: major processes, causes and consequences. J., BOARDMAN and POESEN, J. (eds.), Chapter 36. pp 477–487. John Wiley & Sons, Ltd. [Online]. Available at: http://onlinelibrary. wiley.com/book/10.1002/0470859202. [Accessed: 26 January 2015].
- BOUYOUCOS, G. J. 1962. Hydrometer method improved for making particle size analyses of soils. *Agron. J.*, 54(5): 464–465.

- BRAY, R. H. 1929. A field test for available phosphorus in soils. 1st edition. Bulletin No. 337. Illinois, Urbana.
- BRIGGS, C. S. 2005. C. C. Rafn, J. J. A. Worsaae, Archaeology, History and Danish national identity in the Schleswig-Holstein question. *Bull. Hist. Archaeol.*, 15(2): 4–25.
- BROWN, A. G. 1997. Alluvial geoarchaeology: floodplain archaeology and environmental change. Cambridge, New York: Cambridge University Press.
- BUTZER, K. W. 1971. Environment and archeology. An ecological approach to prehistory. Chicago, Illinois: Aldine Publ. Co.
- BUTZER, K. W. 1980. Adaptation to global environmental change. *Prof. Geogr.*, 32(3): 269–238.
- CARR, C. 1982. Handbook on soil resistivity surveying: Interpretation of data from earthen archeological sites. Evanston, Ill.: Center For American Archeology Press.
- CHAO, L., XUDONG, Z., BALSER, T. C. 2007. Net microbial amino sugar accumulation process in soil as influenced by different plant material inputs. *Biol. Fertil. Soils*, 44(1): 1–7.
- CHESHIRE, M. V. 1977. Origins and stability of soil polysaccharide. *J. Soil Sci.*, 28(1): 1–10.
- CHILD, A. M. 1995. Towards and understanding of the microbial decomposition of archaeological bone in the burial environment. *J. Archaeol. Sci.*, 22(2): 165–174.
- CONWAY, J. 1983. An investigation of soil phosphorus distribution within occupation deposits from a Romano-British hut group. *J. Archaeol. Sci.*, 10(2): 117–128.
- COOK, S. F. and HEIZER, R. F. 1965. The quantitative approach to the relation between population and settlement size. Archaeological Survey Reports Berkeley, Ca: University of California.
- COURTY, M.-A., GOLDBERG, P. and MACPHAIL, R. 1990. Soils and micromorphology in archaeology. Cambridge: Cambridge University Press.
- CRAIG, O. E. and COLLINS, M. J. 2000. An improved method for the immunological detection of mineral bound protein using hydrofluoric acid

- and direct capture. J. Immunol. Methods, 236(1-2): 89-97.
- CRAIG, O. E. and COLLINS, M. J. 2002. The removal of protein residues from mineral surfaces: Implications for residue analysis of archaeological materials. *J. Archaeol. Sci.*, 29(10): 1077–1082.
- D'ANJOU, R. M., BRADLEY, R. S., BALASCIO, L. N. et al. 2012. Climate impacts on human settlement and agricultural activities in northern Norway revealed through sediment biogeochemistry. *PNAS*, 109(50): 20332–20337.
- DANIEL, G. E. 1950. A hundred years of archaeology. Gerald Duckworth & Co. Ltd.
- DICK, R. P., SANDOR, J. A. and EASH, N. S. 1994. Soil enzyme-activities after 1500 years of tenace agri-culture in the Coica Valley, Perú. *Agric. Ecosyst. Environ.*, 50(2): 123–131.
- DIETZ, E. F. 1957. Phosphorus accumulation in soil of an Indian habitation site. *American Antiquity*, 22(4): 405–409.
- ECKMEIER, E. and WIESENBERG, G. L. 2009. Short chain alkanes in ancient soil are useful molecular markers for prehistoric biomass burning. *J. Archaeol. Sci.*, 36(7): 1590–1596.
- ECKMEIER, E. and GERLACH, R. 2012. Characterization of Archaeological Soils and Sediments Using VIS Spectroscopy. eTopoi. *J. Ancient Studies*, Special Volume 3: 285–290. [Online]. Available at: http://journal.topoi.org/index.php/etopoi/article/view/128/148. [Accessed: 26 January 2015].
- EIDT, R. C. 1973. A rapid chemical field test for archaeological site surveying. *Am. Antiq.*, 38(2): 206–210.
- ERNÉE, M. and MAJER, A. 2009. Uniformita, či rozmanitost pohřebního ritu? Interpretace výsledků fosfátové půdní analýzy na pohřebišti únětické kultury v Praze 9 Miškovicích. *Archeologické rozhledy*, 61: 493–508.
- EVERSHED, R. P. 1993. Biomolecular archaeology and lipids. *World Archaeol.*, 25(1): 74–93.
- EVERSHED, R. P. 1997. 5β-Stigmastanol and related 5β-stanols as biomarkers of manuring: analysis of modern experimental material and assessment of the archaeological potential. *J. Archaeol. Sci.*, 24(6): 485–495.
- EVERSHED, R. P. 2008. Organic residue analysis in archaelogy: The archaeological biomarker revolution. *Archaeometry*, 50(6): 895–924.
- FAHY, E., SUBRAMANIAM, S., BROWN, H. A. et al. 2005. A comprehensive classification system for lipids. *J. Lipid Res.*, 46(5): 839–862.
- FORMÁNEK, P., REJŠEK, K., JANOUŠ, D. et al. 2006. Casein-protease, urease and acid phosphomonoesterase activities in moderately mown and abandoned mountain meadow soil. *Beskydy Bull.*, 2006(19): 53–58.
- GORDON, C. C. and BUIKSTRA J. E. 1981. Soil, pH, bone preservation, and sampling bias at mortuary sites. *Am. Antiq.*, 46(3): 566–571.
- HAMMES, K., SCHMIDT, M. W. I., SMERNIK, R. J. et al. 2007. Comparison of quantification methods

- to measure fire-derived (black/elemental) carbon in soils and sediments using reference materials from soil, water, sediment and the atmosphere. *Global Biogeochem. Cy.*, 21(3): GB3016. doi:10.1029/2006GB002914.
- HASLAM, M. 2004. The decomposition of starch grains in soils: implications for archaeological residue analyses. *J. Archaeol. Sci.*, 31(12): 1715–1734.
- HEJCMAN, M., HEJCMANOVÁ, P., HLÁSNÁ-ČEPKOVÁ, P. et al. 2013. Environmental archaeology at the Czech University of Life Sciences Prague – An aplication of new methods for interdisciplinary research. *Interdiscip. Archaeol.*, 4(2): 223–231.
- HJULSTRÖM, B. and ISAKSSON, S. 2009. Identification of activity area signatures in a reconstructed Iron Age house by combining element and lipid analyses of sediments. *J. Archaeol. Sci.*, 36(1): 174–183.
- HOLLIDAY, V. T. 2004. *Soils in archaeological research*. Oxford: Oxford University Press.
- HOLLIDAY, V. T. and GARTNER, W. G. 2007. Methods of soil P analysis in archaeology. *J. Archaeol. Sci.*, 34(2): 301–333.
- HRABOVSKA, B., HAMMEROVA, A., JANDAK, J. et al. 2014. Soil aggregate stability and soil organic matter on chernozems of South Moravia. In: POLÁK, O., CERKAL, R. and ŠKARPA, P. MendelNet 2014 Proceedings of International PhD Students Conference. Brno, Czech Republic: Mendel University in Brno, pp 260–265.
- JONES, A. and MACGREGOR, G. 2002. Colouring the past: the significance of colour in archaeological research. Berg: Bloomsbury Academic.
- KLUTE, A. 1986. Methods of Soil Analysis, Part 1. Physical and Mineralogical Methods. Agronomy Monograph No. 9. 2nd edition. Madison, WI: American Society of Agronomy/Soil Science Society of America.
- LAUER, F., PÄTZOLD, S., GERLACH, R. et al. 2013. Phosphorus status in archaeological arable topsoil relicts. Is it possible to reconstruct conditions for prehistoric agriculture in Germany? *Geoderma*, 207–208: 111–120.
- LAUER, F., PROST, K., GERLACH, R. et al. 2014. Organic fertilization and sufficient nutrient status in prehistoric agriculture? Indications from Multi-Proxy Analyses of archaeological topsoil relicts. *PloS ONE* 9(9): e106244.
- LENĎÁKOVÁ, Z. and GRÍGELOVÁ, A. 2012. Phosphate analysis of sediment from the archaeological site Olomouc-Nemilany. *Geol. Výzk. Mor. Slez.*, 19(1–2). [Online]. Available at: http://www.sci.muni.cz/gap/casop/r2012/026_lendakova12.pdf. [Accessed: 21 May 2015].
- LEONARDI, G., MIGLAVACCA, M. and NARDI, S. 1999. Soil phosphorus analysis as an integrative tool for recognizing buried ancient ploughsoils. *J. Archaeol. Sci.*, 26(4): 343–352.
- MACKNEY, D. 1976. Soil science and archaeology: Susan Limbrey. 1975. pp. xv +384. London: Academic Press. *J. Archaeol. Sci.*, 3(2): 189–190.

- MCGLADE, J. and VAN DER LEEUW, S. E. (eds.). 1997. *Time, process and structured transformation in archaeology.* 1st edition. London and New York: Routledge.
- MENON, R., HAMMOND, L. L. and SISSINGH, H. A. 1989. Determination of plant-available phosphorus by the iron hydroxide-impregnated filter paper (Pi) soil test. SSSAI, 53(1): 110–115.
- MOORE, I. D., GESSLER, P. E., NIELSEN, G. A. et al. 1993. Soil attribute prediction using terrain analysis. SSSAJ, 57(2): 443–452.
- OLSEN, S. L. 1988. Scanning electron microscopy in archaeology. 1st edition. Oxford: British Archaeological Reports.
- PARNELL, J. J., TERRY, R. E. and NELSON, Z. 2002. Soil chemical analysis applied as an interpretive tool for ancient human activities in Piedras Negras, Guatemala. *J. Archaeol. Sci.*, 29(4): 379–404.
- PAUL, E. A. 2014. Soil microbiology, ecology and biochemistry. 4th edition. Academic PRESS.
- PROKEŠ, L., PETŘÍK, J., BERAN, V. et al. 2013. Možnosti statistické a prostorové analýzy hodnot půdních fosfátů na příkladě sekundárně narušených hrobů z Hodonic a Kyjova. Archeologické prospekce a nedestruktivní archeologie v Jihočeském kraji, kraji Vysočina, Jihomoravském kraji a v Dolním Rakousku. Sborník z konference, Jindřichův Hradec 6. 3.–7. 3. 2013, 229–236.
- PROUDFOOT, B. 1976. The analysis and interpretation of soil phosphorus in archaeological contexts. In: DAVIDSON, D. A. and SHAKLEY, M. L. (eds), *Geoarchaeology*. London: Duckworth, 93–113.
- PROVAN, D. M. 1971. Soil phosphate analysis as a tool in archaeology. *Nor. Archaeol. Rev.*, 4(1): 37–50.
- RAKHUBA, D., NOVIK, G. and DEY, E. S. 2009. Application of supercritical carbon dioxide (scCO₂) for the extraction of glycolipids from Lactobacillus plantarum B-01. *J. Supercrit. Fluids*, 49(1): 45–51.
- RAPP, G. R. and HILL, C. L. 2006. Geoarchaeology: The earth-science approach to archaeological interpretation. 2nd edition. New Haven, Ct: Yale University Press.
- REJŠEK, K., VRANOVÁ, V., PAVELKA, M. et al. 2012. Acid phosphomonoesterase (E.C. 3.1.3.2) location in soil. *J. Plant Nutr. Soil Sci.*, 175(2): 196–211.
- REBMANN, A. J., KOENIG, M., DAVID, E. and SORG, M. H. 2000. Cadaver dog Handbook: forensic training and tactics for the recovery of human remains. Boca Raton, FL: CRC Press.
- RETALLACK G. J. 2001. Soils of the Past. An introduction to paleopedology. 2nd edition. Oxford: Blackwell.
- REYMAN, J. E. 1992. Rediscovering our past: Essays on the history of American archaeology. Worldwide archaeology series Vol. 2. 1st edition. Aldershot: Avebury.
- RICHARDSON, M. and GAJEWSKI, B. 2003. Archaeological sampling strategies. [Online]. *J. Stat. Educ.*, 11(1). Available at: http://www.amstat.org/publications/JSE/v11n1/ richardson.html. [Accessed: 21 May 2015].

- RUTTENBERG, K. C. 1992. Development of a sequential extraction method for different forms of phosphorus in marine sediments. *Limnol. Oceanogr.*, 37(7): 1460–1482.
- RYPKEMA, H. A., LEE, W. E., GALATY, M. L. et al. 2007. Rapid, in-stride soil phosphate measurement in archaeological survey: a new method tested in Loudoun County, Virginia. *J. Archaeol. Sci.*, 34(11): 1859–1867.
- SHANKS, O. C., KORNFELD, M. and HAWK, D. D. 1999. Protein analysis of Bugas-Holding tools: new trends in immunological studies. *J. Archaeol. Sci.*, 26(9): 1183–1191.
- SHERRATT, A. 1997. *Economy and society in prehistoric Europe: Changing perspectives*. Edinburgh: Edinburgh University Press.
- SCHAETZL, R. and ANDERSON, S. 2005. Soils: Genesis and geomorphology. Cambridge: Cambridge University Press.
- SCHLEZINGER, D. R. and HOWES, B. L. 2000. Organic phosphorus and elemental ratios as indicators of prehistoric human occupation. *J. Archaeol. Sci.*, 27(6): 479–492.
- SCHOENINGER, M. J. 2010. Diet reconstruction and ecology using stable isotope ratios. In: SPENCER, L. C. (ed.), *A companion to biological anthropology*. Oxford: Blackwell, 445–464.
- SINGER, M. J. and FINE, P. 1989. Pedogenic factors affecting magnetic susceptibility of northern California soils. SSSAJ, 53(4): 1119–1127.
- SULLIVAN, K. A. and KEALHOFER, L. 2004. Identifying activity areas in archaeological soils from a colonial Virginia House lot using phytolith analysis and soil chemistry. *J. Archaeological Science*, 31(12): 1659–1673.
- SUMNER, M. E. 1999. *Handbook of soil science*. Boca Raton, Florida: CRC Press.
- SURABIAN D. A. 2012. Soil characteristics that impact clandestine graves. [Online]. Available at: http://www.forensicmag.com/articles/2012/02/soil-characteristics-impact-clandestine-graves. [Accessed: 10 May 2015].
- TAYLOR, R. E. and OFER, B. Y. 2014. *Radiocarbon Dating*. 2nd edition. Walnut Creek, California: Left Coast Press.
- TORRENT, J. and BARRÓN, V. 1993. Laboratory measurement of soil colour: theory and practice. In: BIGHAM, J. M. and CIOLKOSZ, J. E., *Soil Colour*. 1st edition. SSSA Special Publication Number 31. Madison, Wisconsin: SSSA, Inc., 21–33.
- TUREK, J., HADACZ, R., VRANOVÁ, V. et al. 2015. Soil polysaccharides, a biochemistry and dating of the prehistoric occupation in Brandýs nad Labem, Czech Republic. (sent into Geoarchaeology, January 2015).
- VRANOVÁ, V., ZAHRADNÍČKOVÁ, H., JANOUŠ, D. et al. 2012. The significance of D-amino acids in soil, fate and utilization by microbes and plants: review and identification of knowledge gaps. *Plant Soil*, 354(1–2): 21–39.
- VRANOVÁ, V., REJŠEK, K. and FORMÁNEK, P. 2013a. Aliphatic, cyclic and aromatic organic acids, vitamins and carbohydrates in soil: a review.

- The Scientific World J., 2013(2013), 1–15. [Online]. Available at: http://www.hindawi.com/journals/tswj/2013/524239/. [Accessed: 26 January 2015].
- VRANOVÁ, V., REJŠEK, K. and FORMÁNEK, P. 2013b. Proteolytic activity in soil: A review. *Appl. Soil Ecol.*, 70: 23–32.
- WATANABE, F. and OLSEN, S. 1965. Test of an ascorbic acid method for determining phosphorus in water and NaHCO₃ extracts from soil. SSSAJ, 29(6): 677–678.
- WHITE, R. E. 2013. *Principles and practice of soil science: the soil as a natural resource*. 4th edition. John Wiley & Sons.
- YOHE, M. and GIBBONS, S. 2013. Protein Residue Analysis of Twenty-three Artefacts and Two Soil Samples from Site 7K F-11 the Gray Farm Site in Delaware. Vol. II. In: Archaeology/Historic Preservation. Gray Farm Site DRAFT Phase II and III Excavations on the Murderkill River (Sites 7K-F-11 and 7K-F-169) SR 1 Frederica North Grade Separated Intersection Kent County, Delaware Volume I, Volume II and Volume III Agreement 1416, Task 8. [Online]. Available at: http://www.deldot.gov/archaeology/north_frederica/GrayFarmSite/phaseII_III/pdf/volII/SubConsultantRpt5.pdf. [Accessed: 26 January 2015].