Ancient Starch Research: Goals, Pitfalls, Possibilities

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Starch preservation and reactivity

Bio
Bob Tyler is Professor of Food and Bioproduct Sciences and Associate Dean (Research and Graduate Studies) in the College of Agriculture and Bioresources at the University of Saskatchewan. His teaching and research interests over forty years have included grain chemistry, crop utilization, and food processing and product development, with particular emphasis on pulse crop quality, component fractionation, and starch and protein functionality. He has interacted extensively with the agri-food industry in Saskatchewan in a problem-solving and technical service role on projects related to starch functionality and utilization, particularly in binding and extrusion-expansion applications.

Abstract
Starch exists in grains and seeds primarily as an energy store to be mobilized during germination. The insolubility of starch granules and their relative resistance to degradation are advantageous biologically and are responsible for the durability of granular starch in an archaeological context. Despite this inherent durability, starch is subject to a variety of physical, chemical and enzymatic modifications under appropriate conditions which range from mild to severe. This presentation will provide an overview of some of these modifications, with emphasis on those of most relevance to archeology and diagenesis.
Starch morphology and diversity

Bio
Les Copeland has been teaching and conducting research in the University of Sydney for over forty years on grain chemistry, food starches, plant biochemistry, agricultural science, and the origins of the human diet. He has been the primary mentor of 34 PhD completions and he was awarded the AACC-I 2014 Excellence in Teaching Award. Les is a former Dean of Agriculture, and he was the Foundation President of the Australian Council of Deans of Agriculture. He is a graduate of the University of Sydney, a Fulbright Alumnus and he has held research positions at Yale University, the University of Buffalo, the University of California, Davis, and the Australian National University. Les is Editor-in-Chief of the AACC-I journal Cereal Chemistry and the MDPI journal Agriculture.

Abstract
Starch is a plant polysaccharide of considerable importance for modern humans. It is a macro-constituent of many foods, the major source of dietary energy and an important industrial product. The morphological variability of starch between botanical species is used as a tool in archaeological research for identifying sources of plant-derived foods in ancient diets. This presentation will give an overview of the morphological diversity of starch, its structure and functional properties, and analytical methods, relevant to an archaeological context.
New tools for an Archeologist's toolkit

Bio
Ramaswami Sammynaiken obtained his BSc. Hons. in chemistry from McMaster University, MSc in Physical Chemistry from the University of Guelph and his PhD from the University of New Brunswick. His interest is in the electronic and geometric structure and properties of molecules and materials. Dr. Sammynaiken worked as a temporary faculty member at several universities in Canada before moving to University of Saskatchewan in 2000 to set up and manage the Saskatchewan Structural Sciences Centre. In the years between graduate school and the University of Saskatchewan, Dr. Sammynaiken’s research shifted from structure of transient of molecules to the structure and properties of hard and soft solid materials. This shift and the years at the University of Saskatchewan have converted him to an interdisciplinary scientist who only sees problems to be solved in science and not the walls between disciplines. Dr. Sammynaiken has 96 peer-reviewed publication and has given invited lectures that range from public lecture in palaeontology to photonic materials.

Abstract
Archaeologists are turning to instrumental methods of analysis to glean more information and at the same time instrumentalists are looking to expand into new areas of application. Two disciplines which appear to be diverse are meeting with full force. Great success can be achieved if we take the time to communicate and understand each other’s “language”. I will introduce the new instrumental methods that can be applied to archaeology with the aim that you will want to know more about the methods. Time will also be spent on discussing, sampling, sample handling and sample preparation for instrumental methods – this is not limited to archaeology but all branches of analytical sciences. In conclusion, I will show how archeology, paleontology and current industry challenges are reduced to a common vein, problem solving, for the analytical scientist.
Stable Isotope Variations across Plant Parts: Can we tell Starchy Tissue Apart?

Bio
Robert Patalano is a PhD candidate in the Department of Anthropology and Archaeology, University of Calgary, Canada. His research interests include climate change and human evolution, Stone Age archaeology, and stone tool residue analysis. Bob is investigating prehistoric human-environment interactions using stable isotopes at the FLK-W site of Olduvai Gorge, a site that provides evidence of early Acheulean technology from 1.7 million years ago. His focus is on climate reconstruction and modeling of Pleistocene environments to understand the effective human response to such things as water availability, variations in plant communities, and relative temperatures.

Abstract
The isotopic signature of environmental carbon ($\delta^{13}$C) incorporated during plant biosynthesis represent changes in relative temperatures, precipitation/aridity, evapotranspiration of leaf and soil moisture, and the relative abundance of $C_3$ and $C_4$ plants. In addition, a growing dataset demonstrates that stable isotope variations exist between tissues of the same plant, such as roots, nuts, fruits, seeds, and leaves. Can we measure isotope variations in ancient microbotanical residue? Are we able to tell plant storage tissue apart? Is the geochemical signature of plant residue diagnostic, and does this tell us anything about food extraction and processing? Measuring bulk carbon and nitrogen isotopes is one technique we use to analyse variations amongst different plant parts. In addition, using referential material from roots, nuts, and legumes, we characterize the isotope values of modern species, and use preliminary results for this workshop as a platform to discuss the ways forward in exploring human dietary variability.
Assessment of carbohydrate residues in archaeological and sedimentary materials. Learnings and potential technologies from the dark side

Bio
Steve’s research has focused on organic and petroleum geochemistry and more recently on studies of the deep subsurface biosphere where his work helped define microbial processes in oilfields and the base of the deep crustal biosphere. He has also worked on possible transition technologies for zero emission energy recovery, including direct hydrogen or electricity production, from oil and gas fields and for capturing and sequestering carbon dioxide at large scale. Steve was Scientific Director of Carbon Management Canada Inc., supporting Canada’s academic research in the area of decarbonising fossil fuel use. Steve is a Fellow of the Royal Society (FRS), a Fellow of the Royal Society of Canada (FRSC), a Fellow of the Geological Society (FGS) and a Foreign Member of the Norwegian Academy of Sciences and Arts (DNVA).

Abstract
Characterization of sedimentary organic materials has both driven technological developments and also been highly dependent upon them. Thus, with the development of computerized GCMS (1970s), hydrocarbon focused organic geochemistry became a front-line tool for assessment of organic materials in deep time. Further developments in mass spectrometry and data analysis (ESI, APPI, FTMS-1990s+), have permitted the analysis of complex nonhydrocarbon residues, potentially including those related to carbohydrate diagenesis and biomarker potential. In most settings, reactive biopolymers such as proteins and carbohydrates would be expected to be rapidly altered or mineralized during diagenesis, and any useful proxy species would have to be more refractory in nature than their parent components. One class of diagenetic products that may carry key information on processes, may be melanoidins, reaction products of carbohydrates and amino acids. We look at potential markers for carbohydrate signal preservation in archaeological materials and review recent analytical developments, derived from the petroleum industry, that may have utility in archaeological studies.
Towards a diagenetic framework to aid interpretation of ancient starch

Bio
Matthew Collins is Niels Bohr Professor of Palaeoproteomics at the University of Copenhagen and Professor of Archaeology at the University of York. His research focuses on the persistence of proteins in ancient samples, using modelling to explore the racemization of amino acids and thermal history to predict the survival of DNA and other molecules. In particular, he is interested in developing technological solutions of direct practical application in the humanities. Using a combination of approaches (including immunology and protein mass spectrometry) his research detects and interprets protein remnants in archaeological and fossil remains.

Abstract
Using experience from the study of ancient proteins, I will suggest possible approaches to exploring the authenticity of starch from a diagenetic perspective. I will first explore the way in which we have been attempting to use knowledge of diagenetic pathways to assess the authenticity of ancient proteins. I will then consider cases in which these limits are beaten, and ways in which it is possible to test which element is wrong. Is it the model or are the claims not wholly justified? Finally, I will consider the kinetics of starch hydrolysis/gelatinization and explore whether it will be possible to combine this information with experimental studies in order to produce a similar framework for starches.
Modern contamination in ancient starch laboratories: issues and progress

Bio
Alison Crowther is a Postdoctoral Research Fellow in the School of Social Science at The University of Queensland, Australia, and team leader at the Max Planck Institute for the History of Human Science (MPI-SHH) in Jena, Germany. She has previously held research positions at the Universities of Oxford and Sheffield in the UK. Alison’s current research is investigating the origins and development of Indian Ocean trade and interaction with a focus on coastal East Africa, where she has conducted extensive fieldwork since 2010. Her interests include the archaeobotany of early agriculture in the Indo-Pacific, trans-regional maritime trade and crop transfers, and ancient food processing technologies, which she investigates combining micro- and macrobotanical approaches as well as experimental and taphonomic studies.

Abstract
Establishing the authenticity of ancient starch requires analysts to demonstrate the archaeological provenance of granules, a key component of which is ruling out modern contamination sources. These may be from on-site or in the laboratory. This paper discusses a systematic study of laboratory consumables and environment at labs in the UK and Canada conducted recently to assess the risk of modern starch contamination. It will describe new protocols developed to reduce contamination and will also briefly look at how other laboratories around the world have begun to adopt similar measures and their reported results.
How do starch granules fossilize?
Soils, microbes, and mineral salts

Bio
Dr. Mercader is an associate professor at the University of Calgary and the principal investigator of the SSHRC-funded project Stone Tools, Diet and Sociality. His interests focus on the climatic and dietary changes that drove human evolution with special emphasis on the African Stone Age from the tropical zone. Dr. Mercader's current research concentrates on a multidisciplinary assessment of ancient environments, diet, subsistence, and technological stability and change in East Africa. Over the years, he has also studied a diversity of archaeological sites in Central, West, and Southern Africa. Dr. Mercader has overseen field and laboratory work in multiple contexts, ranging from undergraduate field schools to highly technical laboratory set-ups.

Abstract
In ancient starch research, the term ‘microfossil’ is a widely publicized misnomer to imply microscopic botanical material, but analytical substantiation that the reported starch granules in the archaeological literature in fact underwent fossilization is missing. Under what conditions could starch granules fossilize? How fast would the process be and would fossilized granules retain diagnostic shape and size? What role does microbial degradation play? Modern observation of starch granules in soils constitutes the baseline to illustrate degradation patterns of starch grains in short time spans; outside human interference or culinary manipulation. Controlled laboratory experimentation provides empirical testing of durability and a systematic record of alterations that may occur at the assemblage level within fixed intervals, over extended periods of time, and under changing variables such as substratum, pH, water, inoculum, and microbial modulation. Synthetic fossilization of soft tissues shows that plant parts permeated by high silica solutes undergo fast silicification by the deposition of opaline silica on polysaccharide surfaces and voids. We have learned that starch granules from Dioscorea permineralize quickly, obtaining variably silicified casts.
Is the location of residues on artefacts a reliable indicator to determine if they are use-related or not?

Bio
Hermine Xhauflair is currently a Fyssen Foundation postdoctoral fellow at the McDonald Institute for Archaeological Research at the University of Cambridge where she founded a use-wear and residue analyses discussion group. She is also a research affiliate of the Muséum National d’Histoire Naturelle of Paris, where she did her PhD. She is involved in the study of several sites in the Philippines such as Tabon Cave, Ille and Pasimbahan. To gather data about past practices and behaviours, she investigates the function of stone tools by the mean of use-wear and residue analyses. She is mostly interested in plant exploitation activities and her approach encompasses a strong experimental component and ethnoarchaeology.

Abstract
Analysing residues on stone tools can reveal precise information about the activities that were conducted with the lithic tool and is a valuable technique to reconstruct past human behaviours. However, it is often difficult to assess the nature of the relationship between residues and the artefact on which they are found and they may be related to contamination from various agents. The location of residues is often used to determine whether or not residues are use-related. I will present the result of systematic mapping of residues and use-wear observed on 99 experimental stone tools used to process plants. Surprisingly, it turned out that residues where not spatially associated to use-wear in the majority of cases.
Potential problems with the starch record in dental calculus

Bio
Amanda G. Henry is an associate professor in the Faculty of Archaeology at Leiden University, where she is studying the role of plant foods in human evolution, and how a behavioral ecology framework can help us understand the foraging choices made by our hominin ancestors. She has been using starch grain analysis as one of many means of understanding dietary behavior in the past.

Abstract
Analyses of starch grains preserved in dental calculus and other archaeological substrates can provide information about diet and behavior, but several factors can bias this record. Several studies have shown that the starch grain record in calculus does not capture the entire suite of foods consumed, and that larger starches are more easily preserved and identified. Furthermore, subsequent post-depositional biasing factors may also alter the starch record, but these are less well-understood. The mechanisms by which starches are preserved in calculus despite exposure to salivary amylase has yet to be explored, and the degree to which calculus may be contaminated or the starches in calculus taphonomically altered is unknown. I hope to discuss some new ideas for testing the antiquity of starch grains from calculus, and the potential for post-deposition contamination in calculus residues.
Phytoliths from starchy tissues. What do we know?

Bio
Makarius is a PhD candidate who started working with the Stone Tools, Diet and Sociality team at the University in Calgary in 2014. He is currently receiving theoretical and practical lab training equipping to the newly emerging analytical approaches, techniques and their application for paleovegetation reconstruction. In his PhD thesis, he is doing microscopic analysis of the soil and plant phytoliths; and the laboratory treatment for starch grain separation as well as phytolith extraction from soil sediments.

Abstract
A systematic extraction and classification of opal silica from cortical tissues in starch-containing plant parts can help build a baseline dataset to interpret ancient starch finds from USOs, nuts, legumes, seeds, and fruits. Because hominins utilized stone tools for a variety of plant processing tasks such as digging and pounding, Early Stone Age implements possibly retain a residue record containing both phytoliths from cortical tissues and starches from the core. With modern reference samples, we test whether there are differences between phytoliths produced in different plant parts across 15 species known to contain abundant starches. The plants studied are taxonomically diagnostic and endemic to Zambezian & Somali-Maasai phytochoria. Our goal is to move from referential baseline to palaeoecological application, and to determine which plants tissues produce phytoliths and the relative abundance. This study will help identify phytoliths from starch-rich plants to infer ancient food processing, and will integrate macrobotanical proxy data into the knowledgebase of plant consumption by our ancestors.
Functional analysis of Acheulean lithic industry of Thiongo Korongo (Olduvai Gorge, Tanzania): the case study for flakes and bifaces

Bio
Patricia Bello Alonso is a predoctoral researcher who since 2015 has been developing her thesis project at Centro Nacional de Investigación de la Evolución Humana (CENIEH), located in Burgos (Spain). Presently, her research is based on functional analysis of lithic remains (flakes and LCT’S) from the Early Acheulean (ca. 1.353 ± 0.035 Ma.) archaeological site of Thiongo Korongo. Using a traceological study of the lithic industry of TK, Patricia plans on reconstructing the economic activities of the Early Acheulean in Africa.

Abstract
In this paper we will present an ongoing research about the function of Acheulean flakes and LTC’s recovered at Thiongo Korongo (TK) (Upper section of Olduvai’s Bed II, ca. 1.353 ± 0.035 Ma.). Our hypothesis is that these assemblages could reflect actual evidence of various activities developed at the site other than knapping. The presence of faunal remains, including different species of large mammals, suggest that butchery and carcass processing activities could potentially have been realized there. The variability of formats and tools present support the idea that other activities (e.g. wood working, digging, tuber / vegetable processing, etc.) could also have been performed at the site. These hypotheses are relevant to our understanding of the extent of activities that took place during the Acheulean and the function, technological variability and the settlement dynamics of the site.
Palaeoenvironmental DNA as a complementary source to ancient starch research in Olduvai Gorge

Bio
Tyler Murchie is a Ph.D. candidate at McMaster University working on sedimentary 'ancient' DNA research at the McMaster Ancient DNA Centre. Tyler's training and research interests lie in archaeological science, having completed his undergraduate and masters degrees at the University of Calgary. He has conducted (and disseminated) research on lithic morphometrics and typology, canid ancient DNA (aDNA) as a culture-historical proxy, protein residues, and plastid ancient DNA with cryospheric wooden manuports from the Northwestern Subarctic. Tyler has published on ancient DNA from archaeological canids recovered from sites on the Canadian Plains, as well as a statistical reevaluation and critique of Northern Plains projectile point typologies using digital scanning.

Abstract
The analytical power of ancient DNA (aDNA) has exploded in recent years with developments in genomic sequencing technologies, targeting strategies, and bioinformatic pipelines. This progress has facilitated a broadening of viable sampling materials as supported by increasingly sophisticated means of authenticating ancient biomolecules. Eurasian palaeoanthropology has seen breakthroughs as a result of aDNA analyses of Neanderthal and Denisovan specimens. While the likelihood of extracting endogenous aDNA from ancient Olduvai hominins is remote, research has found that DNA degradation is highly contingent on conditions of the burial environment, and that specimens exposed to minimal taphonomic processes may harbour short, damaged yet preserved endogenous DNA and proteins. Further, a range of palaeoenvironmental target materials may exist in Olduvai specimens—such as dental calculus, palaeofeces, lithic residues, sediment, and pollen—which can be used to generate new, high-resolution palaeoecological datasets. I will focus here on means by which ancient DNA analysis can complement and strengthen ongoing research developments with ancient starch.
Use-wear analysis in the Early Stone Age: the case of Olduvai Gorge, Tanzania

Bio
Sara de Francisco-Rodríguez is a Ph.D. candidate at Universidad de Valladolid, Spain. She has a Five-year degree in History at Universidad de Valladolid (2008-2013) and a Master’s degree in Quaternary, Environmental Change and Human Ecological Footprint at University of the Basque Country (2013-2014). Her current research is focused on lithic use-wear and the functionality of prehistoric collections from Olduvai Gorge, Tanzania. Sara is a recipient of postgraduate grants from the Junta de Castilla y León and the European Social Fund.

Abstract
Use wear analyses are based on the fact that the different functions undertaken with lithic tools leave a variety of alterations on their surface that can be recorded and interpreted in order to determine the type of action performed and type of matter worked. Since 1957, when Sergei Semenov published his seminal book “Prehistoric Technology”, a wide number of functional analyses have been undertaken by archaeologists. For ancient periods, however, use-wear studies are significantly scant due to a number of shortcomings particularly related to conservation and post-sedimentary alterations. In this presentation, we will investigate the use-wear analyses being carried out in a number of archaeological sites currently excavated in Olduvai Gorge, Tanzania (SHK, FLKW, BK, DS). In addition, the advances that we could achieve in our understanding of tool form and function by the combination of use-wear analyses with residue analyses will be explored.
Insights into Ancient Oral Microbiomes Using Starch Preserved in Dental Calculus

Bio
Cory Henderson is a recent graduate from the departments of Biology and Anthropology at the University of North Carolina at Greensboro, and he will begin his career as a Ph.D. student in the department of Biology at the Pennsylvania State University in Fall of 2017. He has conducted field work at Olduvai gorge, and performed research projects in paleolithic archaeology and population genetics. He is broadly interested in the application of genetic and bioinformatic toolsets to address questions about human and non-human primate adaptation.

Abstract
There have been many recent studies that are elucidating the complex nature of modern human oral microbiomes, non-human primate oral microbiomes, and the oral microbiomes of our extinct hominin ancestors, specifically the Neanderthals. This presentation will propose an actualistic project design, where starch granules of modern plants that may have been representative in the diets of extinct hominins are introduced to different microbiota that have been recorded in the oral microbiomes of modern humans, non-human primates, and Neanderthals, to observe how they impact the morphology of the starch granules. The intent of a project of this sort would be to compare the morphological changes to the starch granules in an experimental setting to starch granules preserved in dental calculus, and to use these observations as a proxy for directly analyzing the oral microbiome when DNA might not be preserved.
Microbotanical Controls From Faunal Calculi in the Arusha Region

Bio
Mariam has been working previously at the University of Dar es Salaam and will be beginning her studies at the University of Calgary in the Fall of 2017. She is interested in attaining practical field and laboratory skills in palaeoenvironmental reconstruction, with a focus on the ways in which it influenced change in hominin subsistence patterns, habitat choice, and their plant landscape reconstruction. Specifically, she wants to study phytoliths from modern top soils and plants of different ecoregions in Northern Tanzania and its adjacent areas which will form a more fresh, revised and improved catalogued dataset for ethnobotanical and palaeobotanical studies particularly at Olduvai Gorge and in East Africa. She is also interested in Acheulean, Middle and Later stone tools industries in East Africa.

Abstract
Dental calculus sampled from both living and dead individuals are gaining recognition as an invaluable tool for reconstructing life history. Starch grains, phytoliths, pollen, diatoms, proteins, and DNA often persist within calculus. Therefore, plant particulates from calculus for example, can be a direct indicator of an animal’s plant diet. However, only a few attempts have been made to recover microbotanical remains from dental calculus in Tanzania, and even less so from within the Middle Pleistocene. Drawing on evidence from dental calculus, the main goal of this proposal is to establish microbotanical controls from faunal remains in the Arusha region. This study will employ data from animal teeth collected from the Mid-Pleistocene sites of Makuyuni and Juma’s Korongo, Olduvai Gorge. The anticipated results will establish a control baseline for dental calculus studies in the Arusha region.