Resource Summary Report

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SP3 (Single-Pot, Solid-Phase, Sample-Preperation) Protein Extraction for Dental Calculus

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Protocol Information

URL: https://dx.doi.org/10.17504/protocols.io.bfgrjjv6

Authors: Shevan Wilkin, Richard Hagan, Sandra Hebestreit, Madeleine Bleasdale, Ayushi Nayak, Li Tang, Traci N Billings, Nicole Boivin, Kristine Richter

Summary: Dental calculus from archaeoloigcal samples is now commonly extracted for palaeoproteomic analysis. This protocol combines an EDTA based pretreatment and a SP3 (single-pot, solid-phase-enhanced) protein extraction to prepare samples for LC-MS/MS analysis. This protocol is for moderate to well preserved archaeological calculus samples with a starting weight of 5-15 mg of calculus. We recommend batch sizes of 2-10 calculus samples, 1 extraction control (we use a powdered bone sample from an achaeological sheep bone as this should not be modern material), and 1 extraction blank. For the first time doing the protocol in a lab, we recommend using flanking blanks to identify any contaminants as from handling or from solutions.This protocol is based upon the original SP3 protocol published in Hughes et al. (2014) and the developments for palaeoproteomics of bone (Cleland et al., 2018). When using please cite the following three publications the protocol is based upon as well as the DOI for this protocol.Hughes, C. S., Foehr, S., Garfield, D. A., Furlong, E. E., Steinmetz, L. M., & Krijgsveld, J. (2014). Ultrasensitive proteome analysis using paramagnetic bead technology. Molecular Systems Biology, 10, 757. https://doi.org/10.15252/msb.20145625Hughes, C. S., Moggridge, S., Müller, T., Sorensen, P. H., Morin, G. B., & Krijgsveld, J. (2019). Single-pot, solid-phase-enhanced sample preparation for proteomics experiments. Nature Protocols, 14(1), 68–85. https://doi.org/10.1038/s41596-018-0082-xCleland, T. P. (2018). Human Bone Paleoproteomics Utilizing the Single-Pot, Solid-Phase-Enhanced Sample Preparation Method to Maximize Detected Proteins and Reduce Humics. Journal of Proteome Research, 17(11), 3976–3983. https://doi.org/10.1021/acs.jproteome.8b00637

Associated Publications:

Bleasdale, M., Richter, K.K., Janzen, A. et al. Ancient proteins provide evidence of dairy consumption in eastern Africa. *Nat Commun* **12**, 632 (2021). https://doi.org/10.1038/s41467-020-20682-3

Affiliations: Max Planck Institute for the Science of Human History, Department of Archaeology University of York, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History, Max Planck Institute for the Science of Human History

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