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TRACING THE INTRODUCTION OF DOMESTIC SMALL BOVIDS IN AUSTRAL AFRICA USING PALAEOPROTEOMICS

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INTRODUCTION

- In Austral Africa, no remains of domestic caprines (sheep *Ovis aries* and goat *Capra hircus*) has never been found in archaeological assemblages. This suggests that those domestic animals have been introduced from the Near East to East Africa and then to the southern part of the continent (MacHugh, Larson & Orlando 2017, Vigne 2011).
- Classical morphological determination of caprines from bone remains is often tricky due to the similarities between close related species as well as between domestic and wild species, fragmentation of the remains and bone diagenesis.
- Paleoproteomics has revealed as a powerful approach for the distinction of domestic caprines (Buckley *et al.* 2010). We used this technique on remains from two Austral African sites considered as the oldest pastoral sites of the region: Leopard Cave in Namibia (Pleurdeau *et al.* 2012) and Toteng in Botswana (Robbins *et al.* 2008). Our objective was to ascertain the presence or absence of domestic animals at these sites.
- Three different sample preparation protocols were tested in order to identify the best conditions for remains from arid environment. Bottom-up proteomics analysis was performed on an ESI-Q-TOF device with a data dependent autoMS/MS method. Modern samples from referential collections (MNHN) were also analyzed to constitute a database of unreported diagnostic peptides for species identification.

MATERIALS

ARCHAEOLOGICAL

3 dental remains morphologically identified as caprines

26 unidentified remains (due to taphonomy)

9 presented here

Pleurdeau et al. 2012, Robbins et al. 2008

MODERN

Bones and teeth from domestic and wild African species

Goat *Capra hircus*
Brandberg breed

Sheep *Ovis aries*
Damara breed

Impala *Aepyceros melampus*
Collections Anatomie Comparée MNHN

Springbok *Antidorcas marsupialis*
Collections Anatomie Comparée MNHN

METHODS

ESTIMATION OF THE ORGANIC PHASE PRESERVATION

Fourier-Transform Infrared Spectroscopy (FT-IR)

Lebon *et al.* 2016, Cersoy *et al.* 2016

SAMPLE PREPARATION

Deminerilization: Tris-EDTA 0.5 M, pH 7.4, HCl 1M, 20 min; HCl 0.6M, 4h

Solubilization: NH₄CO₃ 50 mM, 3 h, 65°C, 350 rpm

Digestion: Trypsin « Proteomics Grade » (Promega) 1 µg/µL, 18h, 37°C, 350 rpm

MASS SPECTROMETRY ANALYSIS

- UHPLC : Ultimate 3000-RSLC (ThermoSci), Acclaim RSLC Advantage II column (2,2 µm, 2,1x100 mm, ThermoSci);
- MS : ESI-Q-TOF positive mode, m/z 200-3000
- Analysis : AutoMS/MS data

Bucker Maxis II UHPLC-MS/MS (ESI-Q-TOF)

RESULTS

ESTIMATION OF THE ORGANIC PHASE PRESERVATION

Alpha 2 chain collagen type I

Alpha 2 HS glycoprotein

MS/MS spectrum of [M+H]⁺ ion of LCPCDPLANDSR peptide with a phosphorylation (S) (m/z 853.90, 3,2 min) of one Leopard Cave's sample, deminerilized with Tris-EDTA.

MS/MS spectrum of [M+H]⁺ ion of TPIVQGPSVPGGPVR peptide (m/z 730.92, 4,6 min) of one Leopard Cave's sample, deminerilized with Tris-EDTA.

Site	Species	Peptide	m/z
TOT 14	<i>O. aries</i>	644 GAAGIPEGK 652	m/z 799,44 (2 ⁺)
TOT 09	Suids	145 LCPCDPLANDSR 159	m/z 853,90 (2 ⁺)
TOT 11 & 15	Equids	333 TPIVQGPSVPGGPVR 348	m/z 730,41 (2 ⁺)

Biochronology & geographic repartition Kingdon 1997

CONCLUSION

- Identification of the best protocol for arid environments remains : Tris-EDTA
- Identification of specific markers of unpublished species (*Antidorcas marsupialis*) on 2 different proteins
- Attribution of 4 morphologically undetermined remains
- Identification of sheep at Toteng → confirmation of domestication at this site
- Re-attribution of Leopard Cave remains → wild species: no domestication at this site

ARCHAEOLOGICAL IMPLICATIONS