Journal of Archaeological Science 41 (2014) 631–641

Contents lists available at ScienceDirect

Journal of Archaeological Science

journal homepage: http://www.elsevier.com/locate/jas

Species identification of archaeological marine mammals using collagen fingerprinting ☆

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A R T I C L E I N F O

Article history:
Received 12 June 2013
Received in revised form 27 August 2013
Accepted 28 August 2013

Keywords:
Marine mammals
ZooMS
Archaeological collagen
Species identification
Proteomics
Cetaceans
Pinnipeds

A B S T R A C T

Throughout human history, coastal and marine resources have been a vital part of human subsistence. As a result archaeological faunal assemblages from coastal sites often contain large quantities of skeletal remains indicative of human interaction with marine mammals. However, these are often hard to identify due to a unique combination of factors regarding the procurement, utilisation, morphological and physical characteristics of marine mammal bones. These factors often result in a large number of archaeological cetacean and pinniped specimens fragmented beyond visual recognition, being labelled ‘whale’ or ‘marine mammal’. In this paper we report the development of a **Zoo**archaeology by **M**ass **S**pectrometry (ZooMS) method of collagen fingerprinting, for efficient and low cost discrimination of a wide range of marine mammal species including cetaceans and pinnipeds. We apply the technique to more than fifty archaeological specimens from seven different North Atlantic sites ranging from the Mesolithic until the Early Modern period.

1. Introduction

Coastal activities have been important to hominids from the earliest times, with studies on Neanderthals at different sites across Gibraltar providing evidence that marine mammal exploitation predates modern humans (Erlandson, 2001; Sabin, 2005; Stringer et al., 2008). These species continue to be significant to present day populations, especially to coastal communities and most importantly as a source of food. Marine mammals comprise the cetaceans (whales, dolphins and porpoises), pinnipeds (earless (true) seals, eared seals (sea lions and fur seals) and walruses) and sirenians (sea-cows). These groups are not related and differ markedly in ecology and behaviour. However, due to geographical constraints, only cetaceans and pinnipeds are discussed further in this paper. Pinnipeds are distinguished from cetaceans by their ability to move on land as well as water and have limbs configured to allow this transition. Cetaceans never leave the water and have non-weight bearing forelimbs modified to act as flippers and hindquarters represented only by vestigial pelvises. As a result of this evolutionary commitment, post-cranial cetacean bones are mostly composed of cancellous bone with a thin cortical layer, fewer distinct morphological features and a lower density of mineral in their bones to aid buoyancy (Gray et al., 2007). This lower mineral density reduces the likelihood of archaeological bone preservation in relation to those of terrestrial mammals.

There is a large body of evidence for marine mammal exploitation in communities living on the coasts of the North-Eastern Atlantic, North Sea and Baltic since the prehistoric period (Storå and Lõugas, 2005). The manner of this exploitation can provide important insights into the cultural and technological achievement of a society, partly because of the relative difficulty in exploiting the various cetacean species (Erlandson, 2001; Mulville, 2002). The regular occurrence of cetacean bones at archaeological sites (e.g., Clark, 1947; Hallén, 1994; Herman and Dobney, 2004; MacGregor, 1985) has fuelled a long-running debate about their procurement (Erlandson, 2001; Savelle, 1997). In particular, there is considerable

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http://dx.doi.org/10.1016/j.jas.2013.08.021

interest in whether cetaceans were obtained as an occasional "windfall" due to natural stranding events, or were actively hunted (e.g. Clark, 1947; Erlandson, 2001; Gardiner, 1997; Mulville, 2002) and, if deliberately procured, were they captured with harpoons from the shore, from small boats, or was a more active strategy of hunting whales at sea used?

Whilst marine mammal remains are present on sites from prehistory onwards, the earliest written reference for the exploitation of marine mammals comes from the late 7th century AD text by Adomnán on St Columba, who lived in the 6th century AD. Later, an early Christian text written by Bede in 731 AD mentions the hunting of both seals and cetaceans whilst later still there are accounts of herding, stranding and slaughter of small whales in the Western and Northern Isles of Scotland and in Iceland (Fenton, 1997; Kristjánsson, 1986). Overall, however, there is a paucity of detailed information about marine mammal exploitation in the early written records (Szabo, 2008). It is therefore important to examine archaeological bone assemblages if we are to understand the prehistoric and early historic human interaction with these mammals.

Cetaceans provide food, hide, blubber, sinew, fuel, containers, tool-making material and structural elements (Clark, 1947, 1952; Erlandson, 2001; Mulville, 2002; Savelle, 1997). Toothed whales also provide additional material for carving or ornamentation. Seals supply food, hide, sinew, storage containers from stomach and pericardium, teeth for decoration and blubber (Clark, 1952; Grigson, 1981). Although it is possible that complete seal carcasses may be processed at a settlement, it is unlikely that the bones of larger cetaceans would be transported from the site where an animal was accidentally beached, or a carcass was landed, unless there was some specific reason (Erlandson, 2001; Mulville, 2002; Savelle, 1997). Indeed many cetacean bones recovered from archaeological excavations tend to show evidence of modification, burning or *in situ* structural use. Whale bones were used as building material, for tools such as chopping blocks, and as craft items such as gaming pieces (Childe, 1931; Erlandson, 2001; Mulville, 2002; Savelle, 1997; Smith and Kinahan, 1984; Whitridge, 2002; Harrison et al., 2008; Kristjánsson, 1986; Mehler, 2007). This subjects them to further fragmentation and makes their identification through visible inspection more difficult (Eldjárn, 2000; Erlandson, 2001; Mulville, 2002).

Determining the numbers and species of marine mammal remains found at archaeological sites has an important part to play in resolving questions about their procurement and use. However archaeological cetacean bone is relatively fragile, due to its low mineral content, and has often been worked for various purposes. Consequently many archaeological specimens are fragmented beyond morphological recognition, often being labelled 'whale' or 'marine mammal' (e.g. Harrison et al., 2008; McGovern, 2009; Mulville, 2002; Pálsdóttir, 2008). Additionally, not all countries or regions have museum collections with sufficient numbers of cetacean and pinniped skeletons to use for species identification making it even more important to establish relatively cheap alternative methods to visual inspection of comparative morphology.

Three examples illustrate the difficulty of using comparative morphology for cetacean identification from archaeological sites. Firstly, in a collection from seven Western Isles archaeological sites, ranging from the later Bronze Age to the Norse period, only 30 (5.3%) of 568 cetacean bone fragments could be identified to species when compared with the marine mammal collection held at the Natural History Museum, London (Mulville, 2002). Secondly, only seven (5.2%) of 134 cetacean bone fragments recovered from the Iron Age site of Brest Ness in Orkney, could be identified to species using the comparative collection at the National Museums Scotland (Fraser, unpublished). Thirdly there were few specific identifications among an extensive list of cetacean bone recovered from archaeological sites in the Baltic, North Sea and North-West Atlantic (Clark, 1947).

Pinniped identification is often hampered by the remarkable level of intra-specific variation in bone morphology (Amorosi, 1992; Hodgetts, 1999) and this has led to the widespread practice of identifying only a limited number of elements (e.g., cranium, mandible, humerus, ulna and femur) to the species level. Although other elements of the post-cranial skeleton can sometimes be identified, there is often overlap between species, so that many identifications are only made to 'seal' (Hodgetts, 1999). Since Clarks seminal publication many archaeological sites have consistently yielded small but significant proportions of cetacean and pinniped bone but few of these have been ascribed to species, illustrating the difficulties of identification using morphological characteristics. For example, in the Mesolithic shell midden at Cnoc Coig, Oronsay, Argyll and Bute, a number of grey seals were identified but as many again could only be recorded as *probable* grey seal (Grigson and Mellars, 1987); there were similar difficulties in identifying a common seal. At the same site a range of small cetacean bones could not be identified to species, but the authors suggested these probably derived from common porpoise or common dolphin based on size and present day distribution (Grigson and Mellars, 1987). At the site of Northton on the Isle of Harris, neither pinnipeds nor cetaceans could be identified to species. In the Neolithic assemblage, seven bone and tooth fragments were allocated to seal, which was 1% of those recovered. At the Beaker phase 1 of the site, 19 bones (3%) were allocated to seal and two bones (0.3%) were unidentified cetaceans (Finlay, 2006). At the Neolithic and Early Bronze Age period from Tofts Ness in Orkney, 172 bones were allocated only as 'seal', 1.4% of the total collection for that period, and a further 36 bones (0.3%) as cetacean (Nicholson and Davies, 2007).

Specific identification of cetacean remains has particular value in the understanding of species distributions before the time of large-scale commercial whale hunting in the early modern period (Roman and Palumbi, 2003). Zooarchaeological data has also been used in arguments for and against modern whaling (Mulville, 2005), thus the importance of accurate identification and thorough understanding of the nature of whale exploitation has become particularly significant in recent years. Extraction of DNA provides a means to identify such fragments and can also yield considerable information on species, number of individuals at the archaeological site and even in the source population (Foote et al., 2012; Nichols et al., 2007), however it is a relatively time-consuming and expensive process. In this paper we report the development of a **Zoo**archaeology by **M**ass **S**pectrometry (ZooMS) method, initially designed to separate sheep from goat (Buckley, 2008; Buckley et al., 2010), as a tool that can distinguish a wide range of marine mammal species. As described here, it can be used to separate cetaceans and pinnipeds at least to subfamily levels and down to species level in some groups of cetaceans.

1.1. Species identification using collagen peptide mass fingerprinting (ZooMS)

Depending on the conditions, as collagen (Type 1 collagen; COL1) loss in bone is sensitive to temperature, the preservation of collagen molecules in fossils can be sustained for hundreds of thousands or even millions of years; studies have used collagen peptide mass fingerprints from Mediterranean sites >10 Ka (Buckley et al., 2009; Buckley and Kansa, 2011), British Pleistocene fossils ~1.5 Ma (Buckley and Collins, 2011) and Arctic Pliocene