

## ***Batavia* skeletal research**

### **Bone Chemistry Analyses of (BAT) A15508 + (BAT) A15831: Interim technical report for the Australian National Maritime Museum, Sydney**

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## **Molecular analyses of human skeletal remains associated with the *Batavia* mutiny of 1629**

### **Australian Research Council (ARC) project**

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#### **The Project**

The wrecking of the *Batavia* and the ensuing massacre is well documented in both Australian non-fiction and scientific literature. The location of the massacre is one of Australia's earliest known historical archaeological sites, having occurred well before European colonisation. This project is designed to both contribute new and significant knowledge of events surrounding the mutiny, and to better understand the general life standards of these seafaring 17th-century European peoples. Following a brief introduction to events surrounding the *Batavia* mutiny, I will outline the research objectives and methods, and make some comment on the significance of the expected outcomes. I conclude with a proposed research plan and timeline.

#### **Introduction**

Carrying a complement of approximately 316 people, the *Batavia* embarked from Amsterdam on 29 October 1628, destined for Batavia (modern day Jakarta). Cramped on board were men, women and children of various socio-economic backgrounds and nationalities, including VOC officers and crew, in addition to naval cadets, passengers and soldiers (Drake-Brockman, 1963). Originally sailing alongside a fleet of six other ships, the *Batavia* was subsequently separated, and wrecked on Morning Reef in the Houtman Abrolhos off Australia's west coast on 4 June 1629. The ship's Commander, Francisco Pelsaert, had survivors landed on nearby Beacon Island, and then embarked on a rescue voyage. During Pelsaert's absence, an unsuccessful mutiny attempt resulted in the murder of at least 125 people (van Huystee, 1998).

Human skeletal material has been recovered from excavations of the *Batavia* land sites since the 1960s. Four individual burials were discovered between 1960 and 1964. A further six individuals were recovered from a multiple burial between 1994 and 2001. All are believed to be victims of the slaughter. Some brief descriptions of the multiple burial materials have been made by Pasveer *et al.*, (1998) and Pasveer (2000). More detailed analyses of the total sample were made by Franklin (2001); Franklin & Freedman (2003, 2006). We have permission from the Western Australian Museum to study the *Batavia* skeletal material, which comprises 10 individuals: 7 adults (sex assessed morphologically as 6 ♂ & 1 ♀); 3 juveniles (sex unknown).

## Aims

**1. Sex determination:** there are abundant methods available for determining sex in various skeletal elements. Those methods fall into two broad categories: the first is the visual assessment of morphological features; the second is based on linear measurements and multivariate statistics (Phenice 1969). The accuracy of correct sex determination is known to fall markedly when the material is poorly preserved (Franklin *et al.* 2005). Consideration must also be given to the skeletal age of an individual, as sexing standards for children are especially unreliable (Franklin *et al.* 2006). DNA analysis is the definitive solution to both of these inherent problems, and will allow us to determine the sex of each of the *Batavia* individuals using markers on the X and Y chromosome.

**2. Maternal and paternal lineages:** there have been a number of interpretations of the Beacon Island multiple burial. It was suggested that the burial is that of the family of the *Batavia*'s Predicant (minister), including his wife and six of their seven children (Pasveer 2000). The most plausible theory, on the basis of the reconciliation of morphological evidence and historical documentation, is that this is the burial of sick individuals killed early in the mutiny (Franklin & Freedman 2003, 2006). Using genetic markers from DNA of the multiple burial individuals, we can identify if any familial relationships exist, and thus confirm or refute the Predicant theory.

**3. Commingled remains:** modern construction in the immediate vicinity of the multiple grave resulted in disarticulation of the skulls and associated postcranial skeletons of two individuals. The damaged remains were subsequently recovered in two stages during the 1994 and 1999 field seasons. It is now uncertain whether these skulls have been assigned to their correct postcranial skeletons; a tentative association was made on the basis of morphological assessment (size and development of muscle attachments and metrical dimensions—Franklin & Freedman 2006). DNA identifies individuals uniquely and is thus appropriate to conclusively re-associate the disarticulated remains.

**4. Isotope analysis of diet:** stable isotope analysis of human remains is a valuable research tool in archaeological sites, such as burials, where traditional dietary evidence may be missing or out of context. Carbon ( $\delta^{13}\text{C}/\delta^{12}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}/\delta^{14}\text{N}$ ) isotope analysis of bone collagen taken from the *Batavia* individuals will afford an understanding of the roles that marine and terrestrial resources, and wild and cultivated plants, played in their diets (Katzenberg & Weber 1999). This analysis will allow some insight into potential dietary differences in these individuals based on gender, social status and age (Privat & O'Connell 2002).

## Expected outcomes and significance

Osteological evidence alone, especially when poorly preserved, provides a particularly limited insight into an individual's life history; the combination that data with molecular evidence will allow a much greater understanding of the demographics, health and lifestyle of this small, but

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historically significant, sample. This project will provide both conclusive sex determinations of, and establish any familial relationships between, the recovered individuals. This evidence is especially important in placing the multiple burial in its correct historical context, thus settling the debate between competing theories. Isotopic analyses will allow us a unique opportunity to assess dietary variation within the *Batavia* individuals. As we expect clear dietary differences between sailors, VOC officers and passengers, this method, in conjunction with the historical literature, may be able to help positively identify the recovered individuals. By conclusively reassigning the commingled remains, we can also begin to look at individual identifying markers on those skeletons, which may also suggest a specific identity. The combination of these research aims will undoubtedly contribute new and significant knowledge about general life standards in this 17th- century European population.

#### **Methods and techniques**

As space is limited, the following is a brief synopsis of the methods; the following sources can be consulted for more detailed treatment: Kaestle & Horsburgh (2002); Schwarcz & Schoeninger (1991). The biological samples removed for analysis is complementary to both DNA and isotopic analysis and will follow these steps.

**Preparation and extraction:** material sampled will be photographed, measured and precise casts made. All extractions and analyses will be performed in a dedicated DNA laboratory (Royal Perth Hospital) to minimise the risk of cross-contamination. Before extraction the surface of the samples will be treated to remove contamination; this will be achieved by treating it with bleach and irradiation with ultraviolet (UV) light. Extraction will involve drilling the sample (both teeth and bone) to expose the surfaces not treated by the decontamination protocols; these fragments are then reduced to a powder.

**DNA analysis:** DNA extraction will be performed following the Phenol-chloroform protocol (see Sambrook et al. 1989). Sex determination – PCR amplification of the Amelogenin gene on the sex chromosomes; simple agarose gel electrophoresis/polyacrylamide gel electrophoresis detection based on the size difference of this gene on the X and Y chromosomes. Family associations—PCR amplification of the HV1 and HV2 regions of the mitochondrial genome with subsequent DNA sequencing of the PCR product. Commingled remains—PCR amplification of known polymorphic STR (short tandem repeats) loci run on an polyacrylamide gel; size differences between the resultant products are then used to exclude or include. DNA analyses will be verified by a dedicated ancient DNA laboratory (GeneTime facility, The University of York, UK).

**Isotope analysis:** following lyophilization (soaking in diethyl ether followed by treatment with HCl and NaOH) the extracted dentine collagen will be combusted in a gas analyser and CO<sub>2</sub> and N<sub>2</sub> gases are analysed on a mass spectrometer. For independent verification of the stable isotope

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analyses, 2mg of collagen obtained from the dentition of each individual will be sent to an independent laboratory (Stable Isotope and Biogeochemistry Group, Curtin University). The multiple burial comprises individuals of different nationalities (English and Dutch) and various socio-economic backgrounds e.g. passengers and enlisted crew. For this reason we would expect clear dietary differences in isotopic ratios; VOC officers and passengers would have had a diet higher in meat compared to an enlisted sailor who would sustain mainly on marine resources. As isotopic composition in bone reflects dietary intake in the last few years of life, we thus may be able to distinguish these individuals on the basis of the ratio of carbon ( $\delta^{13}\text{C}/\delta^{12}\text{C}$  – terrestrial resources) to nitrogen ( $\delta^{15}\text{N}/\delta^{14}\text{N}$  – marine resources) isotopes. Strontium (Sr) isotope composition measured in bone or teeth can be used to infer the geographic region that a person inhabited; different regions tend to have distinct Sr isotope compositions because of local geology and ground water. Sr composition in teeth reflects the average ingested as a child; Sr composition in bone reflects the average over the last 10 years of life. In collaboration with Dr Jurian Hoogewerff (Centre for Forensic Provenancing, University of East Anglia) we will be comparing Sr composition of the *Batavia* victims to contemporary European reference samples already collected.

### Research plan and timeline

**January 2007:** casting and drilling; with the provision of a successful application, individual bone elements to be sampled will be measured, photographed and precise casts made; bone pulp will then be extracted.

**February – March 2007:** extraction of samples; this includes DNA extraction and preparation of bone material for isotope analysis. Control samples will be sent to their respective overseas laboratories (see above).

**April – June 2007:** primary data analysis; in consultation with all associate investigators, this three month period is allocated for DNA (profiling of samples) and isotopic analyses (mass spectrometry).

**July – September 2007:** publications; this phase will involve preparation of manuscripts for publication. It is envisioned that at least 2 papers will be produced, one of which will be aimed for a high impact factor medium such as *Forensic Science International*. We will also prepare conference papers, at least one of which will be given at the *Australasian Society of Human Biology* annual meeting.

**October 2007:** final report; the final phase is reserved for the writing and submission of the final report.

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## **Bone chemistry analysis of (BAT) A15508 and (BAT) A15831: Interim technical report for the Australian National Maritime Museum Sydney**

Three *Batavia* bones (a Rt scapula, Rt ulna and Rt fibula) from the post-cranial skeleton (BAT) A15508 on loan from the Australian National Maritime Museum (ANMM) were drilled by Professor John Watling, University of Western Australia, on 2 August 2007 in order to obtain samples for diagenesis (i.e. the disintegration of mineralised or calcified tissues following decomposition) and isotopic analysis. A sample was also obtained from the skull (BAT) A15831 (believed to be associated with the post-cranial skeleton A15508) for comparative analysis, as per the aims and objectives outlined in Dr Daniel Franklin's ARC research proposal.

The sampling was undertaken in the Conservation Laboratory of the Western Australian Museum under the supervision of Dr Franklin and Myra Stanbury, Curator of Maritime Archaeology. The selection of drill sites took into account the following factors:

- the site should not be visible when the bone is exhibited in its correct anatomical position;
- the site should not obscure natural features of the bone; and
- the site should potentially produce the quantity of sample required with the minimum of drilling.

The sample size required was relatively small and all samples were obtained with a hand-micro-drill fitted with a 3 mm drill bit. Where more than one hole needed to be drilled to acquire the required amount of sample the holes were made adjacent to each other so as to localise the interference. Prior to drilling, the sites were dusted with a soft brush only to remove surface detritus; no chemical and/or other agents were applied to the bone. As per the verbal request from ANMM the drill holes have not been cosmetically filled.

The following images were taken prior to the drilling procedure to record the location of the drill holes. Details are given as to the number of holes drilled in each bone and the sample sizes obtained. Professor Watling will undertake the analyses, and results and interpretation will be forwarded when available.

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### **Bone Chemistry Analyses of (BAT) A15508 + (BAT) A15831**

The colour of the scapula, ulna and fibula from A15508 is basically identical to that of the suspected associated skull A15831.

#### **A15508**

All samples were drilled with a hand micro-drill fitted with a 3 mm drill-bit.

#### **Scapula**

Drilled into the scapula neck – 4 holes.

*Approximately 0.5 grams of bone material removed.*



*Figure 1. Location of Scapula drill site. Photo: Daniel Franklin, Centre for Forensic Science, University of Western Australia.*



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### **Ulna**

Drilled lateral to the semilunar notch in the approximate region of *pronator teres* muscle attachment – 4 holes combined.

*Approximately 0.5 grams of bone material removed.*



*Figure 2. Location of Ulna drill sites. Photo: Daniel Franklin, Centre for Forensic Science, University of Western Australia.*

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### **Fibula**

Drilled inferior to the head (approx 3cm) 1 hole into the region of attachment of *Soleus*.

*Approximately 80 milligrams of bone material removed.*



*Figure 3. Location of Fibula drill site. Photo: Daniel Franklin, Centre for Forensic Science, University of Western Australia.*

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Figure 4. Preparing papers for collection of sample. Photo: Patrick Baker, Western Australian Museum.



Figure 5. Preparing to drill the Ulna. Photo: Patrick Baker, Western Australian Museum.



Figure 6. Professor Watling drilling the Ulna. The sample is collected on the small square of paper, which is then folded into an envelope to contain the material and sealed into a marked polythene bag. Photo: Patrick Baker, Western Australian Museum.

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Figure 7. Equipment for preparing and obtaining bone samples. Photo: Patrick Baker, Western Australian Museum.

### Analyses

With regard to isotopes of the Sydney *Batavia* bones and reuniting the skull, the material will need to be analysed in an external laboratory. This will be done by Dr Jurian Hoogewerff, Centre for Forensic Provenancing, School of Chemistry & Pharmacy, University of East Anglia. New developments in instrumentation have created exciting possibilities for the routine 'non-destructive' isotope and trace-element analysis of small and valuable specimens. Trace elements and natural isotopic profiles may be used to verify the authenticity and/or origin of raw materials, industrial products and materials, illegal drugs, foodstuffs, and human remains. The bio-geochemical Natural Isotope and Trace Element (NITE) signatures consist of elemental and isotopic profiles related to regional climate (H and O isotopes), bio-environment (C and N isotopes) and geology (elements, and S, Sr, Nd, Pb and other isotope systems) (see <http://www.ifr.ac.uk/events/hogewerff.pdf>; [http://www.nitecrime.eu.com/WS\\_Wellington020404.pdf](http://www.nitecrime.eu.com/WS_Wellington020404.pdf)); Hoogewerff & Almirall (eds.), in press).

Dr Hoogewerff used isotopic signatures from teeth and bones of the Alpine Iceman to pinpoint his origins to a few valleys in southern Tyrol in Italy (see Holden, 2003; Müller, et al., 2003; Bahn, 2003: 84–90; South Tyrol Museum of Archaeology website: <http://www.archaeologiemuseum.it>).

The work will be done late in the year with results likely in January 2008.

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