Journal of Cultural Heritage xxx (2016) xxx–xxx

Available online at
ScienceDirect
www.sciencedirect.com

Elsevier Masson France
EM|consulte
www.em-consulte.com/en

Original article

Investigation of the recent microbial degradation of the skin of the Chinchorro mummies of Ancient Chile

Alice DeAraujo[a,*], Archana Vasanthakumar[a,1], Marcela Sepulveda[b,2], Vivien Standen[c,3], Bernardo Arriaza[d,4], Ralph Mitchell[a,5]

[a] *Laboratory of Applied Microbiology, School of Engineering and Applied Sciences, Harvard University, Cambridge, MA, United States*
[b] *Universidad de Tarapacá, Laboratorio de Análisis e Investigaciones Arqueométricas, Instituto de Alta Investigación, Arica, Chile*
[c] *Universidad de Tarapacá, Departamento de Antropología, Arica, Chile*
[d] *Universidad de Tarapacá, Instituto de Alta Investigación, Arica, Chile*

ARTICLE INFO

Article history:
Received 11 May 2015
Accepted 9 November 2015
Available online xxx

Keywords:
Mummies
Microorganisms
Skin
Biodegradation
Keratinolytic
Collagenolytic

ABSTRACT

Biodeterioration of cultural heritage artifacts due to microbial activity presents a significant challenge to conservators and museums around the World. A collection of Chinchorro mummies recovered from the Atacama Desert (the oldest artificial mummies ever found, dating back to 5050 B.C.E.) has been stored in the Universidad de Tarapacá, northern Chile. Over the past ten years, accelerated deterioration of some mummies has been documented. Blackening and exudation of some areas of their remaining skin is causing disfigurement of the mummies and poses a threat to the collection, also for mummies in situ exposed to the natural environment. This study was designed to provide a broad analysis of the skin microbiota of Chinchorro mummies and, investigate the relationship between the presence of microbes and the recent discoloration and biodegradation of the Chinchorro mummies' skin. Microorganisms isolated from degraded Chinchorro mummy skin samples were similar, based on ribosomal RNA analysis, to bacteria found in the human skin microbiome (predominantly, *Bacillus*, *Staphylococcus*, and *Methylococcus* spp.) and commonly occurring fungi (predominantly, *Penicillium* and *Aspergillus* spp.). Some of these microorganisms were able to utilize collagen and/or keratin as the sole carbon source in vitro. We determined the activity of the collagenase/gelatinase enzymes produced by these microorganisms when grown on pig skin, which was used as a surrogate for human skin. The concentration of hydroxyproline, a measure of collagenous protein degradation by the microorganisms, increased with increasing relative humidity. We demonstrated that keratinolytic and collagenolytic opportunistic microorganisms were likely responsible for the recent degradation phenomenon.

1. Research aims

A collection of Chinchorro mummies (the oldest artificial mummies ever found, dating back to 5050 B.C.E.) is being stored in the museum at Universidad de Tarapacá, in Arica, the northernmost region of Chile. In the last decade, a blackening and oozing of mummy skin have been documented. This degradation has coincided with increases in humidity, leading to the hypothesis that climate and/or environmental change could have brought about alterations in the skin environment, thereby making the skin more susceptible to degradation. The objective of this new study was three-fold: to investigate the role of microorganisms in the discoloration of the Chinchorro mummy skin, to study the biochemical processes underlying biodeterioration, and to investigate the changes in environmental conditions that precipitate biodeterioration of the Chinchorro mummies' skin. The data obtained in this study will facilitate the development of methods to

* Corresponding author. 58, Oxford Street, Room 301, Cambridge MA 02138, United States. Tel.: +1 617 495 4180; fax: +1 617 496 1471.
E-mail addresses: alice.usa@hotmail.com (A. DeAraujo), archanav@seas.harvard.edu (A. Vasanthakumar), marcelaasre@gmail.com, msepulveda@uta.cl (M. Sepulveda), vivien.standen@gmail.com (V. Standen), barriaza@uta.cl (B. Arriaza), mitchell@seas.harvard.edu (R. Mitchell).
[1] Address: 58, Oxford Street, Room 301, Cambridge MA 02138. Tel.: +1 617 495 3307; fax: +1 617 496 1471.
[2] Address: Antofagasta 1520, Arica, Chili. Tel.: +56 58 2230304.
[3] Address: 18 de Septiembre 2222, Casilla 6D, Arica, Chili. Tel.: +56 58 2205551.
[4] Address: Antofagasta 1520, Arica, Chili.
[5] Address: 29, Oxford Street, Room 126, Cambridge MA 02138. Tel.: +1 617 496 3906.

http://dx.doi.org/10.1016/j.culher.2015.11.004
1296-2074/

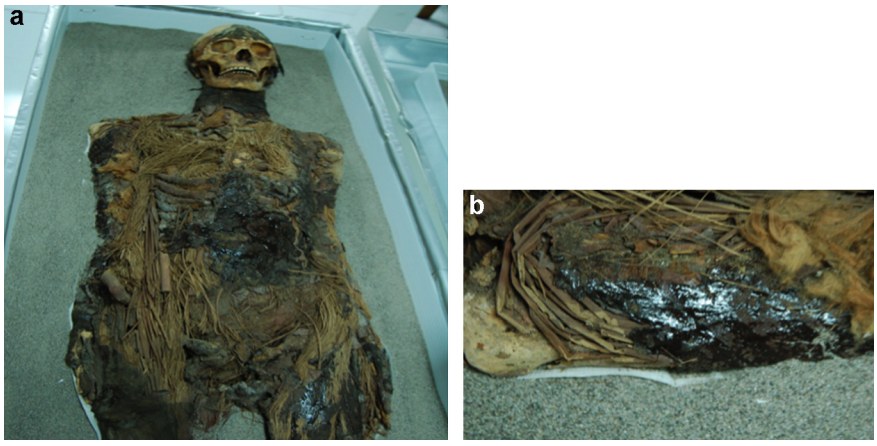

Fig. 1. Recent Chinchorro skin deterioration: (a) Chinchorro mummy and discoloration of some skin areas; (b) close up of the dark area in the right arm. M. Sepulveda [photograph]. Case Morro 1B C1, mummy excavated during 1985.

minimize biodeterioration of this culturally valuable archeological collection.

2. Introduction

Biodeterioration of cultural heritage materials presents a significant challenge to conservation of ancient artifacts. Many cultural heritage materials such as paintings, archival materials, monuments, and ancient human remains have been previously damaged by microbial contamination of these invaluable artistic, cultural and archaeological materials causing corrosion, fouling, rotting, decay, and disfigurement, in processes that liquefy, degrade, or mineralize the various materials [1–4]. The scientific study of ancient human skeletal and artificially mummified remains has gained much attention and appreciation over the past years with the refinement and advancement of laboratory techniques [5,6]. Current methodological approaches and refined laboratory techniques can be used not only to study and learn about the past, but also to preserve and possibly remediate such significant cultural heritage materials.

The Chinchorro mummies, the oldest examples of artificially mummified human remains, reportedly originated in 5050 B.C.E. [7,8], up to one thousand years before Egyptian mummies [9]. The Chinchorro people lived in the geographical region that is currently part of southern Peru and northern Chile. The climate of this area is described as a "mild desert climate". Temperatures range from 18 °C in the winter to 26 °C in the summer. The region is one of the driest in the world, with an average annual precipitation of ~1 mm. In spite of this, the average relative humidity is 70% [10]. However, climatic information from the coastal areas of the Atacama Desert were obtained from the Tarapacá and Antofagasta regions, almost 350 to 900 km to the south of Arica, where the relief features are dramatically different from that in Arica, as indicated by Cereceda and collaborators [10,11]. However, specific long-term studies for this region are not available and are required.

The Chinchorro people mummified all dead bodies, regardless of possible differences in their place in society. The mummies were prepared by removing the organs from the dead bodies and replacing them with animal hair or vegetable fiber. An archeological collection of about 100 natural and artificial Chinchorro mummies is currently stored, with minimal environmental controls, in the Universidad de Tarapacá, northern Chile. These include fetuses, children and all other members of their prehistorical society. The collection was classified according to Arriaza [12] based on the methods used for mummification: natural, red, black, and mud coated. This typology simplified the oldest classification proposed by Uhle [13] and by Allison et al. [7]. The variations in the three principal different mummification processes make their preservation even more challenging.

The mummies have been deteriorating at a slow rate since they were removed from their original sites, especially over the last decade. The problem is worst for mummies in situ in their original funerary place, in the desert. Besides extreme desiccation, which can easily cause breakage of body parts, some areas of the mummies' skin, when it is present, have been rapidly darkening in color over the past few years and therefore further compromising part of the mummified bodies, especially in red and natural mummies. Discoloration of some mummies' skin in the form of a dark exudate started to be documented over the past ten years causing disfigurement of the mummies and posing a threat to the collection (Fig. 1a and b). The dark exudate is found exclusively on the skin of the mummies and is more pronounced in areas of multiple layers or thicker layers of skin and soft tissue [14]. Local observations indicate that the incoming fog from the pacific coast has become denser and the humidity has increased over the past decades. Although no specific studies about climate change in the region have been done, the observed changes occurring in the local environment are a potential explanation for the recent and unexpected discoloration of the mummies' skin. Elevated relative humidity at a surface (70% or higher) is associated with accelerated microbial growth and can lead to problems with mold, corrosion, and decay. Climate-mediated changes in the population of bacteria and fungi on the skin and tissue surfaces may therefore account for some of the observed deterioration.

Since their discovery at the beginning of the twentieth century, extensive research has been focused on the Chinchorro mummies [7,8,12,14–19]. These efforts are mostly the work of anthropologists who have focused their expertise on gathering important information about mummification practices and the cultural context surrounding the Chinchorro, following the pioneering work of Uhle [13,20]. To the best of our knowledge, no studies have surveyed the microbiological constituents of the Chinchorro mummies' skin and the recent accelerated biodeterioration phenomenon. This study is focused on the indigenous microflora and their capacity to cause deterioration of the mummy skin. The skin samples analyzed originated from a female individual from Morro 1b site in 1985.

3. Materials and methods

3.1. Skin samples

Due to the sensitivity and cultural value of the mummy skins, 1-cm^2 pig skin samples were used as a surrogate in the keratinolytic

Please cite this article in press as: A. DeAraujo, et al., Investigation of the recent microbial degradation of the skin of the Chinchorro mummies of Ancient Chile, Journal of Cultural Heritage (2016), http://dx.doi.org/10.1016/j.culher.2015.11.004