

*Case reports***A unique case of naturally occurring mummification of human brain tissue****S. Radanov¹, S. Stoev¹, M. Davidov³, S. Nachev², N. Stanchev¹, and E. Kirova¹**¹Department of Forensic Medicine and Deontology, and ²Department of Pathological Anatomy, Biomedical Research Institute, Medical Academy, Blv. P. Slavejkov 34, 1431 Sofia, Bulgaria³Institute of Cell Biology and Morphology, Bulgarian Academy of Sciences, Acad. Georgi Bonchev St., Block 25, 1113 Sofia, Bulgaria

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Summary. When skulls and bones were exhumed from a mass grave in Bulgaria and subjected to medicolegal examination they were found to originate from 39 humans aged 36–60 years old who had been buried approximately 45–50 years ago. Solid structures which strongly resembled shrunken human brain tissue were found inside 2 intact skulls. Among other bones 5 similar structures were found one of which was an almost entirely preserved human brain, and the others were fragments from different regions of the human brain. Samples of these structures were immersed in 15% aqueous glycerol solution to soften and were examined by light and electron microscopy. Samples of this material and of fresh human brain were subjected to elementary atomic spectral analysis. These complex studies indicated the samples to be naturally mummified human brain tissue and that this process had occurred due to specific conditions within the cranial cavities after burial.

Key words: Human brain tissue – Natural mummification – Histology – Histochemistry – Electron microscopy – Elemental analysis

Zusammenfassung. Bei Ausgrabungen in Bulgarien wurden Schädel und Knochen von 39 Individuen im Alter zwischen 35 und 60 Jahren, die vor ca. 45–50 Jahren begraben wurden, gefunden. Bei der nachfolgenden gerichtsmedizinischen Expertise wurden in zwei intakten Schädeln und zwischen den Knochen anderer Schädel sieben harte Gebilde (zwei ganze und fünf Fragmente), die wie geschrumpfte menschliche Gehirne aussahen, identifiziert. Entsprechende Proben von diesem Material wurden für 3–4 Tage mit 15% wässriger Lösung von Glycerol weicher gemacht und danach lichtmikroskopisch und elektronenmikroskopisch untersucht. An entsprechenden Stückchen von frischem menschlichem Gehirn und den gefundenen Proben wurde eine Atomspektalanalyse zur Bestimmung des chemischen Bestandes an Elementen durchgeführt. Die komplexe Untersuchung des Materials ergab, daß es sich bei diesem Fund um ei-

nen Sonderfall von natürlich mumifizierten menschlichen Gehirnen handelt, die durch sonderbare Bedingungen nur im Schädel der begrabenen Individuen entstanden sind.

Schlüsselwörter: Gehirn – Mensch – Naturalische Mummifikation – Histologie – Histochemie – Elektronenmikroskopie – Atomspektalanalyse

Introduction

Mummification (natural, artificial, complete, partial) of human corpses has been known for thousands of years [1, 2, 7–9, 12, 14]. It is generally held that natural mummification occurs due to defined environmental conditions such as dry air, good ventilation and high temperature [1, 3, 8, 13]. Skin is initially affected by the mummification process while viscera, being readily decomposed in the early post mortem period, are changed to dry structureless plates and membranes [1, 2, 6, 9, 10]. One of the structures undergoing particularly rapid destruction is the human brain. Nevertheless, one case has been reported of brain tissue which was preserved 10 years after burial of the corpse [8].

At the Loshin Site in the vicinity of Dobrinishte Village (Bansko Community, Sofia District) the excavation of a mass grave revealed completely skeletonized skulls and bones 30–40 cm below ground level, in loose and stony soil which was well exposed to sunlight. Medicolegal examination ascertained that these were bones from 39 humans (38 male and 1 female) in the age range 35–60 years old. Firearm wounds in the frontal or temporal region were found in 12 skulls. In 25 cases multifragmental fractures of the skulls were found which had been caused by hard blunt objects or by firearms. Structures were strongly resembled human brain tissue, although greatly reduced in size, were found inside 2 intact skulls. Among the examined bones 5 other similar structures were found one of which was an almost entirely preserved human brain, and the others were fragments from different regions of the human brain.

The work described here was undertaken to verify this observation and included gross examination, light and electron microscopy and comparative elemental (emission) analysis with mummified and fresh tissue samples.

Materials and methods

Gross examination of the material, description of outer appearance and microscopical features:

Histological studies. To soften the material, fragments of varying size were immersed in 15% aqueous glycerol solution for 3–4 days. Blocks approximately 1 cm³ in size were fixed in 10% neutral formalin and embedded in paraffin. Paraffin sections (6–8 μ) were stained with hematoxylin and eosin, Cossa to demonstrate Calcium, Perls to demonstrate iron, Bodian to demonstrate neurofilaments, and metazol-fast-blue to demonstrate myelin [11].

For electron microscope studies, small (5 × 2 mm) blocks of softened material were fixed in 2% OsO₄ in 0.1 M sodium phosphate buffer pH 7.4 for 1 h and embedded in Durcupan after dehydration. Ultrathin sections (μ) were additionally contrasted with lead citrate and uranyl acetate, and observed using a JEM 100B electron microscope at 80 kv.

Sections from 4 mummified brains and 2 fresh human brains were examined by atomic emission spectral analysis to determine the inorganic elemental chemical composition. Two grams of each sample were desiccated under infrared-lamp under mild conditions for 10 days. The samples, cleaned of surface impurities were ground and homogenized with pure carbon powder (1:1 ratio) in agate mortars. Spectra were obtained in triplicate from each sample under the following analytical conditions: PGS-2 diffraction spectrography, revolving diaphragm 3.2, split 15 μm, 1 order of spectra, range 225–345 nm, exposure 120 s. The results were semiquantitative and approximate as precise reference standards were not available.

Results

The preserved structures strongly resembled human brains, although they were hard in consistency and black in color, weighed 57–72 g and were about 22 times smaller than fresh human brains. The volume ratio of pre-

served skull to entire mummified formation was 1450 cm³:71 cm³ (Fig. 1). The brain hemispheres bearing discernible gyri and sulci were easily recognizable as well as the separate parts of the brain (Figs. 2, 3). The cross-section showed a distinguishable, nonuniformly outlined, thin outer band, greyish-black in colour, suggestive of cortex, and below a zone of whitish-grey colour resembling the white substance of the brain. Protruding from the outer surface of the examined formation were some roots of vegetation.

Hematoxylin-eosin stained preparations showed moderately eosinophilic matter devoid of cells. In some areas structures could be observed suggesting old hemorrhages, which gave a positive reaction for iron. Metazol-fast-blue staining demonstrated separate structures resembling fibres in cross-section or transverse section with surfaces very similar to myelin sheaths. Staining for neurofibrils according to Bodian revealed distinct positive fibers (Fig. 4).

Electron microscopical studies

No brain cells were detected in the preparations examined. There were numerous multilamellar structures of



Fig. 1. Size relation between human skull and mummified brain

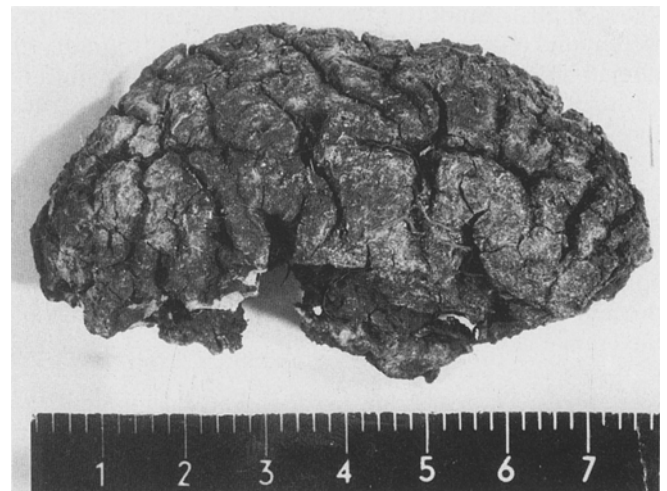


Fig. 2. Outer surface of mummified human brain

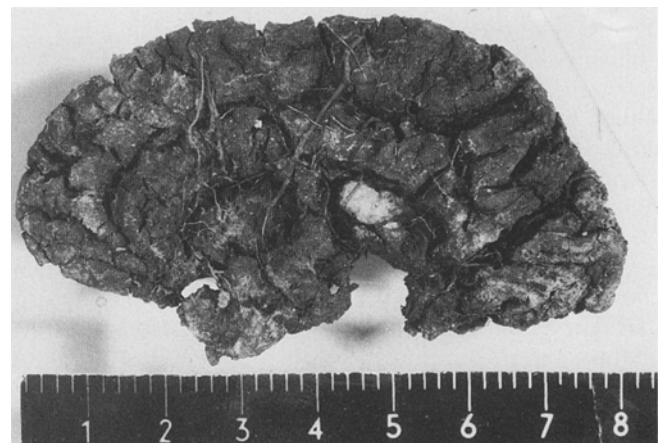


Fig. 3. Inner surface of mummified human brain

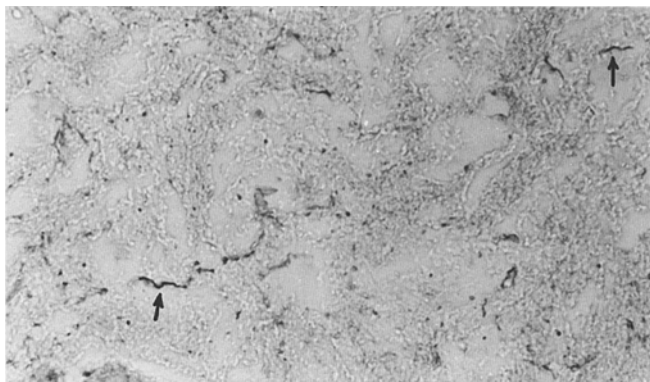


Fig. 4. Neurofibrils (arrows), Bodian, $\times 63$

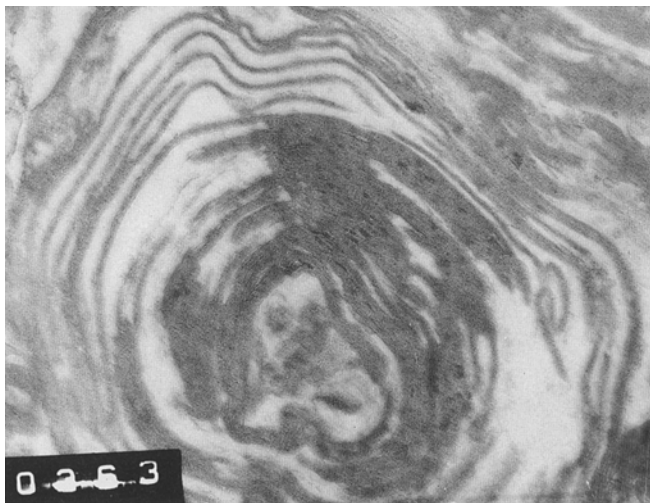


Fig. 5. Myelin – like body with numerous lamellae (electron micrography, $\times 51000$)

varying size, shape, and density of disposition of the lamellae but the central zones showed no axoplasm or axolemma (Fig. 5). Some multilamellar bodies displayed dense masses enclosed in a membrane. Similar dense masses were also seen between lamellar bodies.

Elemental analysis

The semiquantitative data obtained from comparative elemental analysis indicated a similarity between the calcium, copper and lead contents of mummified material and normal human brain. Mummified material contained less phosphorus and sodium, whereas the control brain tissue contained less manganese, silicon, aluminium and titanium.

Discussion

Based on the evidence obtained, it would seem reasonable to assign the case to a unique find: mummified human brain occurring naturally under conditions which caused complete decomposition of soft tissues and organs. This was clearly due to an appropriate microclimatic ambiance developing exclusively within the cranial cav-

ity of the corpses. Here, suitable temperature and ventilation apparently enabled rapid evaporation of intracellular brain fluid [4, 5, 8]. Nevertheless the process must have taken some time since the brain cells had been destroyed. Virtually the only remaining structures found were medullary sheaths of myelinated nerve fibers, although they too showed varying degrees of destruction.

Nerve cell autolysis most probably proceeded under sterile conditions because neither cells resembling microphages nor cell debris were observed in the tissues. A likely contributing factor to brain mummification may have been acute haemorrhage, which is supported by the presence of traumatic bone lesions as well as by the histological findings suggestive of old haemorrhages with the presence of iron-containing material (most probably hemosiderin).

In conclusion, the phenomenon observed is believed to represent a unique case of naturally occurring preservation of human brain tissue, in the presence of complete decomposition of other organs and soft tissues. It should be noted that none of the investigation methods, if used alone, would have allowed a definite medicolegal assessment of brain mummification. However, their combined use made this possible.

The findings of bone changes (complete skeletonization), sprouting of vegetation, deposition of calcium salts and differences in elemental composition relative to fresh human brain were consistent with available preliminary information that there had been a period of about 45–50 years between the death of the individuals and the discovery of the mummified tissue.

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