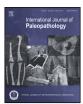
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Histology of ancient soft tissue tumors: A review

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ABSTRACT

Ancient neoplasms diagnosed in the soft tissues of mummies are limited to 18 cases so far, with only 5 malignant tumors. The apparent paucity of neoplasms in ancient populations is sometimes attributed to shorter life spans and fewer oncogenic substances in the environment. However, this paucity may also be a result of the scarcity of autopsies of mummies, together with technical difficulties in detecting neoplastic lesions in mummified tissues. An exception, and example of the benefits of thorough systematic analysis, is the small sample of 10 Renaissance mummies from Naples (15th–16th centuries), in which 3 cases of cancer were found. In order to increase detection of soft tissue tumors, it is imperative that mummies undergo systematic autopsies and histological examinations performed by skilled paleopathologists. This review of the known ancient soft tissue neoplasms demonstrates the state of histology of malignant and benign soft tissue neoplasms in mummies, and the potential for further study. The limitations of paleopathological diagnosis will be discussed and an argument will be made for the use of autopsies and histological analysis on mummified human remains.

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1. Introduction

4.

Although cancer currentlyrepresents the second leading cause of death in advanced countries (Torre et al., 2015), it has been said that the prevalence of cancer may have been lower in the past (David and Zimmerman, 2010). The number of neoplastic lesions documented so far in the paleopathological literature, distributed over a long time span and in different countries, includes about two hundred cases in the skeletal apparatus (Strouhal, 1994; Capasso, 2005; Hunt et al., in this issue). However, there are only eighteen cases of soft tissue tumors in the literature, all discovered by paleopathologists (Fornaciari and Giuffra, 2012) using different methods of rehydration (Ruffer, 1910; Sandison, 1955) and staining for light microscopy (Allison and Gerszten, 1982; Reyman and Dowd, 1980) or electron microscopy (Riddle, 1980).

The main reasons for the rarity of soft tissue tumors in paleopathology, especially malignancies, are apparently the short life span of past populations, the scarcity of mummified remains available in comparison with skeletal remains, and technical difficulties in the detection of neoplastic lesions in mummified tissues. This review of the known ancient soft tissue neoplasms demonstrates the state of histology of malignant and benign soft tissue neoplasms in mummies, and the potential for further study. The limitations of paleopathological diagnosis will be discussed and an argument will be made for the use of autopsies and histological analysis on mummified human remains.

2. Malignant tumors

2.1. Colon

2.1.1. Rectal adenocarcinoma in a Roman Period Egyptian mummy

An abnormal tissue mass found in the rectum of an adult male mummy (West Cemetery #5, Body C) from the Dakhleh Oasis, Egypt, and dating back to the Roman Period, was examined histologically. The rectal wall was well-preserved, with a polypoid mass protruding into the lumen and showing a relatively wellpreserved hyperplastic colonic mucosa in a tubular pattern (Fig. 1a). The nuclei were not preserved, but the central portion of the lesion showed a deep invasion of the epithelium into the underlying muscularis (Fig. 1b, arrow). Although the cellular detail was not well-preserved, the cells were shown to be periodic acid Schiff (PAS)-positive and the reticulin stain showed an epithelial pattern, with clusters of cells surrounded by reticular fibers. Small clusters of similar PAS-positive material were observed deep within the muscularis, revealing a deep invasion through the wall (Zimmerman and Aufderheide, 2010). The histological picture clearly demonstrated degenerated epithelial cells with prevalent non-invasive, adenomatous structure, but some sections showed clear invasion and infiltration into the submucosa of an atypical glandular component. This lesion is consistent with an infiltrating adenocarcinoma occurring in a benign villous adenoma.

2.1.2. Colorectal adenocarcinoma from Naples (15th century)

The anthropogenic mummy of Ferrante I of Aragon (1424-1494), King of Naples, revealed a hollow fibrous structure in the pelvis, identified as the rectum at autopsy. After rehydration, the specimen reached a size of $14 \times 6 \times 1$ cm (Fig. 2a) and on cross-section it appeared dark brown, with many small roundish grey-white areas 0.5-3.0 mm in diameter (Fig. 2b). Histology revealed epithelial tumor cells, disposed in cords, solid nests and glands (Fig. 3a, b), disseminated in a fibrous stroma containing striated muscular fibers (Fig. 4c). The cells were crowded and tall, with abundant cytoplasm and pseudo-stratified pleomorphic hyperchromatic nuclei (Fig. 4a). Bizarre nuclei, with indentations and irregular clumps of chromatin appeared in electron microscopy (Fig. 4b). The mucus was scarce and limited to pseudo-glandular formations, as shown by the specific staining with Alcian-blue and PAS (Fig. 4d). These results pointed out a moderately differentiated mucinous adenocarcinoma infiltrating the muscular-fibrous layers of the small pelvis. A strong intracytoplasmic immunoreactivity of the neoplastic cells for pancytokeratin was shown (Fig. 4e) and a positive staining, showing actively proliferating cells, was observed for Proliferating Cell Nuclear Antigen (PCNA) (Fig. 4f), whereas Prostate-specific Antigen (PSA) and Carcinoembryonic Antigen (CEA) gave negative results. The histological, histochemical and immunohistochemical results clearly indicate a mucinous adenocarcinoma of the digestive tract, most probably of the colorectum (Fornaciari, 1993).

DNA hybridization probe and sequencing analysis of K-RAS exons 1–2 demonstrated the presence of the codon 12 GGT > GAT transition (Marchetti et al., 1996; Ottini et al., 2011). This mutation represents the most frequent mutation of the K-RAS gene in sporadic colorectal cancer and is associated with exposure to chemical carcinogens, probably present in the diet (Saffhill et al., 1985; Kuhnle and Bingham, 2007).

An increased intake of red meat is known to induce a significant (3-fold) increase in fecal N-nitroso compound (NOC) levels, with a range of exposure evident in the feces similar to that from tobacco-specific NOC in cigarette smokers (Fornaciari et al., 1999). A paleonutritional study with Carbon (¹³C) and Nitrogen (¹⁵N) stable isotopes revealed a massive intake of animal proteins caused by a large consumption of meat by King Ferrante (Fornaciari, 2008), and histology revealed severe atherosclerosis of the carotids (Gaeta et al., 2013). In conclusion, the alimentary "environment" of the Neapolitan court of the 15th century, with its abundance of natural endogenous alkylating agents, well explains the *K-RAS* mutation that caused the tumor that killed the Aragonese king over five centuries ago.

2.1.3. Colon adenocarcinoma from Naples (16th century)

Autopsy of the natural mummy of Luigi Carafa, Prince of Stigliano (1511–1576) (Fornaciari, 2006), showed "a hollow organ with material" of about 8×3 cm, identifiable as a portion of a bowel loop with feces. Histology revealed a tract of colon wall with very well-preserved mucosa, submucosa, muscularis propria and vis-

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