History of Medicine

Cancer and therapy in the 16th century: the unique case of adenocarcinoma in Luigi Carafa, prince of Stigliano (1511–76)

In 2016, a rare and surprisingly well preserved case of colon adenocarcinoma was discovered in the natural mummy of a 16th century Neapolitan noble, the Prince Luigi Carafa of Stigliano (1511–76; figure 1).[1]

This tumour represents an extraordinary finding, because it is the third malignant soft tissue tumour identified in the mummies of the Neapolitan court preserved in the Basilica of Saint Domenico Maggiore in Naples and the fifth known in the paleopathological literature worldwide.[2]

Autopsy showed well preserved intestinal loops with a tract of enlarged colon with faeces (appendix). Histological analysis showed an exceptional morphology, with an excellent state of preservation of the tissue (appendix), which is contrary to all expectation considering that the intestines are the most putrescible organs of the body. Previous histological evaluation showed a well preserved colon wall with mucosa, submucosa, muscularis propria and visceral peritoneum with villous adenoma, and focal adenocarcinoma having an apparently superficial level of infiltration (stage T1).[2]

Considering the relevance of this case for paleo-oncology, and the new finding of clear invasion of the subserosal adipose tissue (figure 2), further analyses including toxicological investigation and application of several antibodies were done on the soft tissue samples of the tumour, to better determine the histological type of the adenocarcinoma and the treatment to which the patient had been submitted.

Immunohistochemistry showed strong positivity for anti-pan cytokeratin, a typical marker of normal and neoplastic epithelial cells; anti-cytokeratin 20 (figure 2), which identifies normal mature enterocytes, goblet cells, and gastrointestinal epithelial neoplastic cells; CD10, which links to a membrane metallopeptidase (cell and apical surface staining is observed in well differentiated colon carcinoma, pancreas, and prostate); Ki67, a marker of active cell proliferation; and CDX2, a highly specific and sensitive marker of cells of colorectal origin. Immunohistochemistry showed negativity for anti-cytokeratin 7, usually negative in colonic epithelium, which alongside with CDX2-positivity constitutes a pathognomonic panel of primary colorectal cancer; negativity for HER2, a member of the human epitelial growth factor receptor family shows the low aggressivity of the tumour; and against the altered protein p53, which is present in many adenocarcinomas (table).[3]

The immunohistochemical panel (table) allowed to diagnose a well differentiated colon adenocarcinoma infiltrating the soft tissue (stage T3), which developed on a colonic villous adenoma.[4]

Toxicological analysis of colon samples by atomic absorption spectroscopy showed very high toxic concentrations of lead (50 ppm) and copper (53·5 ppm): the normal mean concentration of lead in colon is 0·04 ppm (0·04 median); and is 0·12 ppm (0·11 median) for copper.[5] The same analysis done on a sample of striated muscle tissue showed concentrations of lead (3·1 ppm) and copper (1·4 ppm), which were much lower than those of the colon. The toxic concentrations of lead and copper in the gut, with the resulting abatement of intestinal and putrefaction flora,[6] can explain the exceptional preservation of the tumour cells. To reach this degree of gut concentration, the lead and the copper must have been taken orally or through anal administration with an enema.

The use of lead and copper was prevalent in Renaissance pharmacology, which was influenced by iatrochemical medicine. Already in the 15th century, the so-called virginal milk, a mixture of lead acetate (lead sugar or

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For a full reference list see **Online** for appendix

Figure 1: **Natural mummy of Luigi Carafa**

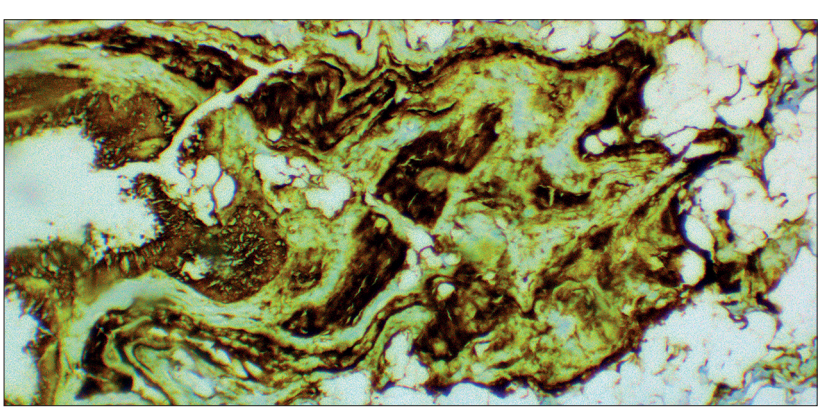

Figure 2: **Evident neoplastic invasion of the subserosal adipose tissue (anticytokeratin 20, magnification ×100)**

	Staining	Interpretation	Positivity
Pan cytokeratin	Cytoplasmic	Epithelial cells	++
Cytokeratin 20	Cytoplasmic	Colorectal, bile duct, pancreas, gastric cells	+++
CDX2	Membranous or cytoplasmic	Colorectal cells	+++
Cytokeratin 7	Cytoplasmic	Negative in colonic cells	–
CD10	Membranous or cytoplasmic	Colon, pancreas, and prostate carcinoma	++
Ki67	Nuclear	Cell proliferation	++
HER2	Membranous or cytoplasmic	Neoplastic progression	–
p53	Nuclear or cytoplasmic	Tumour suppressor gene	++

++ indicates strong positivity, +++ very strong positivity, and – negativity.

Table: **Immunohistochemistry of the tumour cells**

saccharum saturni) and water developed by the physician Antonio Guaineri, was used for many pathological conditions.[7]

In the 16th century, the Swiss physician and alchemist Paracelsus strongly promoted the use of metals in the treatment of different diseases. Among these, mercury, antimony, gold, copper, and lead became the subject of interest and experimentation by the iatrochemical physicians.[8]

Although an external use of lead acetate, in particular, was more common, there is clear evidence of the internal use of the compound in the writings of Renaissance and Modern age physicians.[9] Lead acetate was used to stop diarrhoea and gut haemorrhages. In fact, sugar of lead acetate and copper, considered astringent substances, were used orally in the 16th century medicine and later, in particular by followers of Paracelsus, to cure the intestinal bleeding and diarrhoea.[10]

It is possible that Luigi Carafa, who had an advanced stage of the cancer that is likely to have caused melaena, was treated with a similar potion, which explains the extraordinaryly unique preservation of the intestinal tissues and the perfect histological picture of the colon adenocarcinoma. This case represents one of those very rare instances in which direct evidence of ancient medicine can be gained from accurate ancient body examination.

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Spotlight

Will immunotherapy really change radiotherapy?

Opening opinion: Yes

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For more on the **preclinical work combining immunotherapy and standard of care** see *Nat Commun* 2017; **8:** 15618

For more on the **phase 3 trial** see *N Engl J Med* 2018; **379:** 2342–50

For more on **treatment approaches with immunocytokines** see *Clin Cancer Res* 2015; **21:** 1151–60

For more on **patient selection for radioimmunotherapy** see **Series** *Lancet Oncol* 2019; **20:** e452–63

Cancer is a systemic disease and more than 90% of patients with cancer are dying of metastasis while radiotherapy is being used as the loco-regional treatment. This issue has been the formidable challenge of radiotherapy for more than a century, having an effect on locoregional control, but no or limited effect on overall survival. The high-quality translational work and a randomised phase 3 trial in non-small cell lung cancer comparing immunotherapy with standard of care have been game changers. Durvalumab significantly prolonged overall survival compared with placebo (stratified hazard ratio for death 0·68; p=0·0025).

So, is this the definitive solution? Should we just use this unchanged chemoradiotherapy protocol followed by the standard checkpoint inhibitors a few weeks later? My answer to this last question is no, because I believe that immunotherapy will really change radiation oncology. Immunotherapy is a paradigm changer in oncology, although the long-term cure is still unacceptably low. Combining immunotherapy with a proven therapeutic approach is obvious and radiotherapy looks to be ideally suited to this role for several reasons: primarily because of its known safety profile, but also its capacity to mediate robust immunostimulatory effects, which might be able to aid immunotherapy in achieving systemic tumour control.

There are several arguments to investigate fundamental changes of current radiation oncology practice. Nowadays, it is assumed that radiotherapy is, in the long term, most effective when it causes tumour-targeting immune responses. As a consequence, radiotherapy could be administered in doses and schedules that stimulate anticancer immunity rather than kill cancer cells through DNA damage. But we know that the combination of radiotherapy with immunotherapy has a systemic effect, improving the overall survival in non-small-cell lung cancer (PACIFIC trial).

Strong (pre)clinical evidence suggests that key effectors of immunotherapy are CD8-positive lymphocytes. Irradiation of large blood vessels, the heart, and lymphoid