In this issue:

Review
*Glomus coccygeum: a review*

Original article
*Detection and genotyping of hpv-dna through different types of diagnostic platforms in liquid-based cervical-cytology samples*

Case reports
Pathologica storica
*Pathologica ai tempi della Spagnola*

Proceedings
*IV Meeting Nazionale Gruppo Italiano Paleopatologia*
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CONTENTS

Review

Glomus coccygeum: a review
M. Bisceglia, S. Bisceglia, C. Ciampi, G. Panniello, C. Galliani

Original article

Detection and genotyping of hpv-dna through different types of diagnostic platforms in liquid-based cervical-cytology samples
B. Cassani, G. Soldano, D. Finocchiaro, S. Conti, A. Bufamante, G. Lemorini, G. Bufamante

Case reports

Unclassified sex cord/gonadal stromal testis tumor with a "pure" spindle cell component: a case report
C. Spairani, S. Squillaci, A. Pitino, F. Montefiore, W. Fusco

Calcifying aponeurotic fibroma: a core biopsy-based diagnosis
F. Motta, S. Scavo, G.M. Vecchio, G. Fuccio-Sanzà, F. Nicolosi, G. Magro

Lymphnode metastasis of thyroid cancer misinterpreted as lateral aberrant thyroid 40 years before identification of primary tumor. Case report and review of the literature

Pathologica storica

Pathologica ai tempi della Spagnola
C. Patriarca, C.A. Clerici

Proceedings

IV Meeting Nazionale Gruppo Italiano Paleopatologia
Glomus coccygeum: a review

M. BISCEGLIA1, S. BISCEGLIA2, C. CIAMP1, G. PANNIELLO3, C. GALLIANI4

1 Anatomic Pathology, School of Biomedical Sciences, Etromapmacs Pole, Lesina (FG), Italy;
2 Polyclinic of Modena, Modena, Italy; 3 Unit of Clinical Dermatology, Polyclinic “Ospedali Riuniti” di Foggia, Italy; 4 Department of Pathology, Children’s Minnesota, Minneapolis & St. Paul, MN, USA

Key words
Glomus coccygeum • Sucquet-Hoyer canals • Arterio-venous anastomosis • Glomus tumor • Coccygodinia

Summary
With limited information about the coccygeal glomus found in classic textbooks, we deemed it necessary to review the subject. The illustrations presented in this article derive from four coccygeal glomera incidentally encountered during examination of pilonidal disease specimens. Familiarization with its microanatomical features may help to avoid inappropriate interpretation of this enigmatic structure.

Introduction
A glomus body is a spheroidal to ovoid dermal microanatomical structure that constitutes an arteriovenular glomeriformis anastomosis. Each glomus consists of one or more arterial segments that branch into a number of glomic arterioles surrounded by rows of round, uniform, epithelioid contractile cells, that drain into a collecting venous plexus. These sphincteric anastomoses, the essential parts of the glomus, are eponymously known as the Sucquet-Hoyer canals after J.P. Sucquet (1840-1870), a French anatomist, and H. Hoyer (1864-1947), a Polish anatomist, who first described them. Endowed with a high-sympathetic tone, the glomus body is involved in skin thermoregulation acting as a sphincter to control the flow of blood by locally mediated axon reflex responses, to bypass or enable the capillary bed, and prevent loss or dissipate heat.

Knowledge of the function of cutaneous arteriovenous anastomoses in man dates back to the early 1930s when the work of the British physiologists and cardiologists Thomas Lewis (1881-1945) and George W. Pickering (1904-1980) as well as the work of the British cardiologist Ronald T. Grant (1892-1989) and the American cardiologist Edward F. Bland (1901-1992) were published. These arteriovenous anastomoses are scarce in the newborn, but develop rapidly during infancy and early childhood. In the elderly, the glomera regress, sclerose, and diminish in number, thus explaining the increase susceptibility to thermoregulatory disturbances affecting the extremes of life.

Glomera are frequently encountered in the hands and feet, chiefly in the deep dermis of the digital pads and nail beds, but they also occur in the ears, nasal and alimentary mucosa (where the function is related to absorption), thyroid, erectile tissue, and many other sites including the coccygeal region.

In 1860, Hubert von Luschka (1820-1875), a German surgeon and anatomist, first identified the glomus coccygeum at the ventral tip of the coccyx in his dissection studies of the pelvis, naming it “glandula coccygea.” Luschka compared glomus coccygeum with the carotid body (“glomus caroticum”), a chemoreceptor, thinking that both were glands. In 1865, the Swiss-born Julius Arnold (1835-1915) described the “glandulae coccygeae” even on the ventral surface of sacrum, recognizing their vascular origin along the median sacral artery, and naming them “glomeruli arteriosi coccigei.” He considered both glomus coccygeum and “glomus” caroticum as true glomeruli. In 1902, these microanatomical structures were included in the paraganglion system by the Austrian histologist Alfred Kohn (1867-1959). In 1907, the non-paraganglionic (non-chromaffin) nature of the glomus coccygeum was demonstrated histochemically.
by another Austrian Mediziner, the anatomic pathologist Oskar Stoerk (1870-1926). However, credit is due to William H. Hollinshead (1906-1986), an American anat
omist, who in 1942 categorically established anatomical and physiological distinctions between the “glomerus” coccygeum and “glomerus” caroticum (carotid body) 9.

Several terms have been used in the past, referring to the same glomus coccygeum, such as coccygeal body, glomus coccygeicum, pericoccygeal glomus, Luschka’s body, gland of Luschka, coccygeal gland, and “glandula coccygea”.

Glomus coccygeum is homologous to the “caudal glomeruli” (also called “glomerula caudalia”), which are non-nutrient arteriovenous anastomoses present in variable number (up to 15) – according to species – in tailed mammals, from rodents to monkeys 9-16. However, while there is evidence that caudal arteriovenous anastomoses in animals play an important role in thermoregulation 15-17, glomus coccygeum in humans (and in other tailless primates as well) represents a phylogenetic vestigial rest, that is an atavistic organ loosing the normal thermoregulatory function as the dermal glomera of other sites. In fact, the subcutaneous panniculus, where the glomus coccygeum resides, serves as an insulator, not as a dissipator or absorber of heat.

However, the precise function of this structure in humans remains speculative 18. A presumptive hematopoietic function via an immunomodulatory activity of the sympathetic nervous system has been recently proposed 19. There is limited data, if any, regarding glomus coccygeum in classic textbooks of anatomy and physiology, and therefore in this article we review the world literature on the subject.

Anatomic and clinical findings

There is no recorded evidence of this type of arteriovenous shunt in fetuses between 10-18 weeks-gestation, when a presumptive anlage for the coccygeal body may be an arterial plexus derived from a tortuous median sacral artery admixed with nerve fibers and sympathetic ganglion cells 20. However, well-formed coccygeal glomera have been observed for decades in previable fetuses (145-170 mm crown rump length) 21. Postnatally, the glomus coccygeum lies deeply buried in the adipose tissue immediately below or just ventral to the tip of the coccyx, near the center of the natal cleft, in the vicinity of the anococcygeal ligament, between the branches of the median sacral artery and vein, innervated by the pelvic sympathetic plexus 18-20,22-26 (Fig. 1).

Glomus coccygeum, which is endowed of up to 4 arterial segments may be either single 22, binodal, or multiple 24,27, or present as a network of glomus bodies in the coccygeal region 12,24, with a few of them occasionally located even in coccygeal vertebrae 24,25.

It is one of the largest glomera in humans 18,25, measuring between 1 and 5 mm (mean diameter: 3.5 mm in a large study) 22,25.

The prevalence of glomus coccygeum is uncertain. Its presence was nearly constantly documented in three anatomic autopsy studies as it was found by different investigators in 5 out of 5 28, in 29 of 32 26, and in 17 of 20 24 coccygectomy specimens either from adults 28, elderly 26, or individuals of pediatric and adult ages 24. Instead, in anatomic studies of specimens submitted for surgical pathology, glomus coccygeum was identified in 13 of 40 (32.5%) coccygectomy specimens resected in individuals with coccygodinia 23 and in 18 of 37 (48.6%) coccygeal bones removed during rectal resection for advanced rectal and uterine carcinomas and for various other reasons 27.

As a normal structure, the coccygeal body is inapparent. In five cases of coccygodinia which had been ascribed to pericoccygeal glomus tumors 29-32, which were most likely normal coccygeal glomera, the symptomatology subsided after coccygectomy, but in 3 cases there were radiographic abnormalities of the coccyx or intraoperatively proven fractures. And there are large series of patients treated with coccygectomy for both traumatic and idiopathic coccygodinia in which no remarkable features involving glomus coccygeum were encountered in most of the cases, except for histological degenerate changes in the sacrococcygeal or intercoccygeal discs in some 25,32,34.

In clinical practice glomus coccygeum is usually an incidental finding in excised specimens from sacrococcygeal areas for diverse causes, such as pilonidal dis-
Histological features

Glomus coccygeum may exhibit a well-circumscribed or multinodular appearance, mainly comprised of small arterioles surrounded by a mantle of epithelioid myoid cells, rich of unmyelinated nerve fibres and embedded in loose to dense fibrous connective tissue. Due to the variable proportion of the constitutive elements of the glomus body, some authors distinguish the following variants: the glomus cell nodule-dominant type, an intermediate mixed morphology with a mixture of nodules and vessels, and the vascular-dominant type with scattered individual glomic cells (Figs. 2-6).

Immunohistochemistry and electron microscopy

Immunohistochemically, the glomic cells of the glomus body in general are immunoreactive for vimentin, collagen type IV (external basal lamina), muscle specific actin, and alpha-smooth muscle actin (Fig. 2B), and negative for epithelial markers, EMA, endothelial markers (CD31 and FVIII-RA), and neuroendocrine markers, such as chromogranin and synaptophysin, with low proliferative activity. Conflicting findings have been reported for neuron specific enolase (mostly positive), desmin, and S-100 protein (mostly negative). CD34 was expressed in 3 cases, focally in 2 and diffusely in one. Ultrastructurally, pericellular external lamina, subplasmalemmal vesicles, bundles of actin microfilaments, groups of membrane bound organelles including mitochondria and endoplasmic reticulum can be seen in glomus cells, but no membrane bound endocrine granules have ever been observed.

Histological differential diagnosis

In a compilation of tumoral and pseudotumoral entities of the soft tissues that surfaced over the preceding quarter of a century published in 2006, two of the authors (MB; CAG) made reference to the potential pitfall for the “glomus coccygeum of Luschka”. In fact, because of its considerable size and multiple anastomotic channels, coccygeal glomus mimics glomus tumor, that is a tumor mostly occurring in distal extremities which was firstly described in 1924 by the French-born, Canadian, histopathologist Pierre Masson (1880-1959). As a matter of facts, P. Masson applied it the name “glomus” (“neuromyoarterial glomus tumor”, also called “arterial angioneuromyoma”) to depict its similarity to the glomus coccygeum of Luschka, hence the term glomus tumor has occasionally been used synonymously (but erroneously) to refer to a normal glomus coccygeum. Parenthetically, glomus tumor was also alternatively named “glomangioma” in 1935 by the American pa-
Histopathologist Orville T. Bailey (1909-1998), who believed it represented a subgroup of (hem)angiomas. Importantly, glomus coccygeum may be easily misinterpreted as a coccygeal glomus tumor by the unwary histopathologist, and in fact a series of glomus coccygeal tumors, presumed to be normal coccygeal glomera, have been reported by several authors. Bell and collaborators first exposed the potential for confusion, and Albrecht and Zbieranowski emphasized the risk of this misinterpretation. No additional coccygeal glomus tumors have been published since, and subsequent publications all warned against misdiagnosis and overdiagnosis of this normal microanatomical structure. Therefore, it seems that the glomus body in this specific location is not particularly susceptible to neoplastic transformation. However, although any coccygeal glomus tumor should be viewed with scepticism, 4 cases of solitary “true” coccygeal benign glomus tumors have been reported so far in the literature: 2 of the 3 cases by Nutz and Stelzner, and the cases by Llombart et al. and Kim et al. These tumors, which were clinically a “palpable lesion”, and of 1 to 2 cm in size, exhibited the same morphologic features of glomus tumors occurring in the usual extracoccygeal sites. Benign glomus tumors are expansile lesions, which cause the classic triad of pain, tenderness, and cold sensitivity, sometimes eroding the adjacent bone surface. There is no record of inherited glomus vascular malformations, malignant glomus tumor, or glomus tumors of uncertain biologic behaviour involving the glomus coccygeum. Extracoccygeal glomus vascular malformation or glomuvenous malformation (multiple “familial glomangiomomas”) are either localized (glomangiomatosis) or widespread, with an autosomal dominant pattern of inheritance.
Generally, malignant glomus tumors are deeply seated, measure 2 cm, exhibit atypical mitotic figures, moderate to high nuclear grade, and at least 5 mitotic figures/50 high power fields. Glomus tumors of uncertain biologic behaviour exhibit high-mitotic activity and are superficially located, or of large size only, or deep seated only. No histological variants of glomus tumors, either with oncocytic features or symplastic high grade nuclei, have been reported in the coccygeal region. Other entities which may possibly enter the differential diagnosis with normal glomus coccygeum are the following: intradermal melanocytic nevus, paraganglion and paraganglioma, neuroendocrine tumor, adnexal skin tumor, and metastatic carcinoma.

Intradermal melanocytic nevus is less organoid and immunohistochemically is strongly positive for S-100 protein and negative for alpha-smooth-muscle actin. Paraganglioma (paraganglioma of the filum terminale might be in point) is a tumor arising from the paraganglion system. If carefully examined, it looks morphologically different, is positive for argyrophilic stains and immunoreactive for neuroendocrine markers, and shows typical neuroendocrine granules when ultrastructurally examined. Although the glomus coccygeum is not related to the paraganglia, with which it may be confused, we would like to alert the reader about the incorrect use, in the older literature as well as among clinicians of some discipline even today, of the name “glomus” for other dif-
ferent microanatomical chemoreceptoral structures of the paraganglion system (glomus caroticum, glomus jugulare, glomus tympanicum, ...), which erroneously glomus coccygeum one and a half century ago was ascribed to. Adnexal skin tumors (mainly eccrine acrospiroma), neuroendocrine tumors, and metastatic carcinomas have different morphologies and can be easily excluded with the support of immunohistochemistry, mainly evidencing their consistent cytokeratin immunoreactivity.

Conclusions

**TAKE-HOME-MESSAGES:**

- It is surprising how little attention is devoted to glomus coccygeum in standard textbooks of human anatomy and histology.
- It is a normal, likely phylogenetic vestigial anatomical structure with similar cytoarchitectural constituents as the dental Sucquet-Hoyer canals.
- Its functional significance is uncertain, even enigmatic, given its ectopic location in the hypodermis.
- Its constituents seem to be disinflected to pathological alterations such as hyperplastic and neoplastic proliferation.
- In summary, familiarization with this seldom observed microanatomical structure of the coccygeal region is necessary to avoid overinterpretation as a pathological one.

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Detection and genotyping of HPV-DNA through different types of diagnostic platforms in liquid-based cervical-cytology samples

B. CASSANI, G. SOLDANO, D. FINOCCHIARO, S. CONTI, A. BULFAMANTE, G. LEMORINI, G. BULFAMANTE
U.O.C. di Anatomia Patologica, Citogenetica e Patologia Molecolare, P.O. San Paolo, ASST dei Santi Paolo e Carlo, Milano, Italia; Dipartimento di Scienze della Salute, Università degli Studi di Milano, Italia; Servizio di medicina di laboratorio per il programma di screening lombardo del tumore della cervice uterina (D.D.G. n. 12386 del 28 novembre 2016)

Key words
Cervical cancer • HPV test • Screening

Summary

Background. At present cervical cancer represents the second most common cancer in women worldwide and it reaches a global mortality rate of 52%. Only the early detection and the adequate treatment of pre-neoplastic lesions and early-stage cervical cancer decrease the mortality rate for this type of cancer. Cervical carcinoma screening, as a method of second prevention, is currently feasible through molecular research of high-risk HPV genotypes and in lots of organized screening programs the Pap-test is performed only in women with positive HPV-test. Currently, there are various diagnostic platforms detecting and molecular genotyping HPV, which are based on different procedures, determining uneven viral genotypes panels and using diverse type of vials to collect and store the samples. Previous studies have pointed out that DNA-HPV test can be negative in pre-neoplastic lesions, even of high grade, or in presence of cervical cancer. Therefore, it’s important to assess the risk of false negative diagnoses using DNA-HPV molecular test, because in this circumstance women do not undergo immediately Pap-test, but they are submitted to second round screening with DNA-HPV test after 5 years: this protocol could increase the incidence of “interval cancers”. The present study aims at comparing the results of HPV detection and genotyping on liquid based cervical cytology, using some of the most relevant diagnostic platforms in commerce.

Methods. The study is based on a group of patients which went to their private gynecologist in a contest of opportunistic screening. The vial used in the examined population has been EASY-PREP® preservative solution (YD Diagnostics CORP-Republic of Korea); liquid cervical cytology sampling has been done using a single device (plastic brush), allowing to collect simultaneously cytological material from exocervix and endocervix (Rovers® Cervex-Brush®). The diagnostic platforms employed have been the following: A) Digene HC2 HPV DNA Test, on RCS System (QIAGEN); B) BD Onclarity™ HPV test, on automate platform BD Viper™ LT (Becton Dickinson); C) Xpert® HPV, on GeneXpert® Infinity Systems platform (Cepheid). Every platform researched high-risk HPV genotypes panels (hr-HPV). Part of the clinical records has also been analyzed through PCR and genes L1 and E6/E7 complete sequencing, in order to further typing the viral population.

Results. We have examined 1284 samples of women aged 16 to 73 years: 1125 have been tested using HC2 procedure, 272 samples with Onclarity method, 159 with Xpert® method and 55 samples have been analyzed using PCR and sequencing of gene L1 and gene E6/E7. HPV-DNA was detected with Onclarity method in 15,07%, with Xpert® method in 13,83% and using HC2 procedure in 12,27% of samples. The comparison between the three molecular methods revealed diagnostic discrepancies in 3,14% of our records between Onclarity test and Xpert® method and in 2,20% (6/272) between HC2 test and Onclarity test. Globally, in 431 tests, compared using different diagnostic platforms, discrepant diagnoses, referring to hr-HPV presence or to detected genotype, have been observed 11 times (2,55%). Genotype 16 appeared the most expressed in the positive samples (20,99%), whereas genotype 18 resulted the less expressed in the examined population (4,94%).

Discussion. The present study highlights the following: 1) Positive results’ percentage for high-risk HPV-DNA genotypes, deriving from the three diagnostic platforms used and with the same vial to collect and store samples, does not significantly vary on the basis of the type of equipment and it is congruent with the Italian percentage already detected during organized screening programs. 2) Even the molecular diagnostic approach could give false negative results, preventing the detection in the screened population of cervical HPV-related lesions and theoretically endangering women to develop “interval cancer”. 3) In the population examined, genotype 16 has been the most expressed, whereas genotype 18 was among the less frequently detected. Other genotypes often noticed have been: 56-59-66 (Onclarity P3 group), 31, 51 and 35-39-68

Correspondence
Gaetano Bulfamante, Head Unit of Human Pathology, Cytogenetics and Molecular Pathology, St. Paolo Hospital, ASST Saints Paul and Charles, Milan, Italy - Tel. +39 02 503 23167 - 23180 - Fax +39 02 503 23168 - E-mail: gaetano.bulfamante@unimi.it
(Onclarity P2 group). This remark emphasizes the importance of HPV infection and genotypes distribution’s continuous monitoring, considering that HPV-vaccines planned in Italy in the “National vaccination prevention program 2017-2019” are not specific for the majority of these genotypes. 4) The necessity to improve the screening program to identify cervical carcinomas and pre-neoplastic cervical lesions is remarked by the detection during HPV-test of possible coinfection (present at least in 8.76% of our records). In fact, the risk of development of cervical cancer might be associated with type-specific interactions between genotypes in multiple infections and, in addition, other genotypes, not targeted by quadrivalent HPV-vaccine, can increase the risk of cervical carcinoma. 5) As there’s a different combination of HPV-genotypes in diagnostic categories used by the HPV screening platforms, it’s important that anyone who is in charge of this diagnostic analysis promotes among clinicians the adequate rendition of the laboratory’s data in the patient records, reporting both the diagnostic result and the method through which it has been obtained.

Introduction

At present cervical cancer represents the second most common cancer in women worldwide 1. More than 85% of cervical cancers develops in low-income or resource limited countries 2, whereas in 2010, the invasive cervical cancer rate in the US was 7.5 per 100.000 women 3. This type of cancer reaches a global mortality rate of 52%. Around 90% of these deaths affects low or medium income countries 4 and it is expected that by 2030 98% of cervical cancer deaths will occur in these same countries 5. The early detection and the adequate treatment of pre-neoplastic lesions and early-stage cervical cancer decrease significantly the mortality rate for this type of cancer 6. Indeed, still nowadays the healing chance is low when cervical cancer is diagnosed at a later stage of disease. In 2010 the 5-year survival rate in the US was 91% when the diagnosis of invasive cervical cancer was made at an early stage of disease, however the same survival rate decreased to 16% in late-stage cancer 7. The conventional Pap-test (Papanicolaou smear) has historically been the mainstay of pre-neoplastic lesions detection and cervical cancer screening; recently it has been introduced a new method, called “liquid-based cervical cytology” (LBCC) 8. The LBCC has one main convenience: it’s possible to perform on a single sample both the Thin-Layer Cervical Cytology and the molecular HPV research. Many previous studies 9-16 have supported the usefulness of molecular research of specific DNA-HPV genotypes as a primary screening method, saving the LBCC just for women with positive HPV test. In this particular occurrence, it’s convenient that the two tests (HPV-test and Pap-test) are realized from the same liquid-based cytological sample (co-testing), in order to reduce the number of false negatives of HPV-test or of Pap-test 17-22. Lately, the Government of Lombardy has approved a regional screening plan which establishes the early detection and the adequate treatment of cervical cancer deaths will occur in these same countries 5. The diagnostic platforms employed in the present study have been collected during the second half of 2016 and the first half of 2017. The vial used in the examined population has been EASYPREP™ preservative solution (YD Diagnostics CORP., Republic of Korea); liquid-based cervical cytology sampling has been done using a single device (plastic brush), allowing to collect simultaneously cytological material from excocervix and endocervix (Rovers® Cervix-Brush®). Within three months, in the patients who underwent Pap-test only it has been performed high-risk HPV-test, using the sample’ stock. Likewise, within three months, part of the samples was analyzed through a second type of DNA-test, using a different diagnostic platform. The diagnostic platforms employed in the present study have been the following:

- BD Onclarity™ HPV test, on automate platform BD Viper™ LT (Becton Dickinson);
- Xpert® HPV, on GeneXpert™ Infinity Systems platform (Cepheid);
- Digene HC2 HPV DNA Test, on RCS System platform (QIAGEN).

Table I compares high-risk genotypes panels detected by the various diagnostic platforms with the standard required from Lombardy for its screening program. The molecular and cytological diagnoses have been conducted independently by operators in a blind trial; when the same sample was tested with different platforms in order to identify HPV, operators were working ignoring...

Methods

The present study is not based on a population recruited through an organized screening program: all patients, during spontaneous access to their private gynecologist, underwent “liquid based cervical cytology” on which it has been performed the co-test Pap/HPv or just the Pap test.

Our clinical records have been collected during the second half of 2016 and the first half of 2017. The...
first analysis result. Pap-test diagnoses are not evaluated in the present study, because it is focused on HPV-test diagnostic concordance rate using different platforms: in fact, organized screening programs, which establish molecular test as the primary test to perform, do not prescribe Pap-test execution if molecular test is negative. Part of the clinical records has also been analyzed through PCR and direct sequencing of L1 and E6/E7 viral genes region, in order to further typing the viral population. Our Human Pathology Unit cooperates with external quality control for HPV screening (VEQ HPV Screening), organized by Lombardy Government. Results’ statistical significance has been evaluated according to Chi-Squared test (one tailed). A p-value of ≤ 0.05 was considered as statistically significant.

Results

We have examined 1284 samples of women aged 16 to 73 years. Table II displays the age distribution. Among these, 1125 have been tested using HC2 procedure, 272 samples have been analyzed with Onclarity method, 159 with Xpert® method (Fig. 1) and 55 samples have been analyzed using PCR and sequencing of gene L1 and genes E6/E7. HPV-DNA was detected with Onclarity method in 15.07% of samples, with Xpert® method in 13.83% of samples and using HC2 procedure in 12.27% of samples. These rates are not significantly different according to a statistical data analysis (HC2 versus Onclarity: p = 1.54; Onclarity versus Xpert: p = 0.12; HC2 versus Xpert: p = 0.31). The comparison between the three molecular methods revealed diagnostic discrepancies in 3.14% of our records (5/159) between Onclarity and Xpert (respectively 3 positive samples using Onclarity and negative using Xpert; and 2 positive samples using Xpert and negative using Onclarity) and it revealed discrepancies in 2.20% of our records (6/272) between HC2 test and Onclarity test (respectively 6 negative samples using Onclarity and positive using Xpert; and 2 positive samples using Xpert and negative using Onclarity) and it revealed discrepancies in 2.20% of our records (6/272) between HC2 test and Onclarity test (respectively 6 negative samples using Onclarity and positive using Xpert; and 2 positive samples using Xpert and negative using Onclarity). The PCR analysis and L1, E6/7 viral genes sequencing revealed 1 positive sample for HPV-16 and 2 positive samples for HPV-18: this samples were negative according to HC2 method; in these three samples, all belonging to women older than 34 years, the Pap-test has always been positive for HPV-related lesions. Globally, referring to 431 tests evaluated with different methods, discrepant diagnoses of hr-HPV have been recorded 11 times (2.25%): HC2 High-Risk HPV DNA Test has diagnosed as “negative for hr-HPV infection” 6 of 272 samples (2.20%) co-tested with other platforms (Onclarity), Onclarity method has diagnosed as “negative for hr-HPV infection” 2 of 159 samples (1.26%) co-tested with Xpert and Xpert has diagnosed as “negative for hr-HPV infection” 3 of 159 samples (1.88%) co-tested with Onclarity. Among 81 HPV-test

<table>
<thead>
<tr>
<th>HPV genotypes considered in Lombardy screening program</th>
<th>BD Onclarity™ HPV test</th>
<th>Xpert® HPV</th>
<th>Digene HC2 HPV DNA Test</th>
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<tbody>
<tr>
<td>16</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>18</td>
<td>X</td>
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<td>X</td>
<td>X</td>
</tr>
<tr>
<td>68 (optional)</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
</tbody>
</table>

Tab. II. Age distribution in the examined population.

<table>
<thead>
<tr>
<th>Age</th>
<th>Number of patients</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 25</td>
<td>122</td>
<td>9.5%</td>
</tr>
<tr>
<td>25-33</td>
<td>327</td>
<td>25.5%</td>
</tr>
<tr>
<td>34-64</td>
<td>794</td>
<td>61.8%</td>
</tr>
<tr>
<td>&gt; 64</td>
<td>41</td>
<td>3.2%</td>
</tr>
<tr>
<td>Total</td>
<td>1284</td>
<td>100%</td>
</tr>
</tbody>
</table>
positive samples, it has been possible to identify the infecting genotype (Tab. III). The most detected genotype has been number 16 (20.99%), followed by P3 Onclarity - affecting genotype (Tab. III). The most detected genotype positive samples, it has been possible to identify the intermediate or low risk ones (Tab. IV).

**Discussion**

The risk of invasive cervical cancer considerably decreases in women who periodically undergo Pap-test and/or DNA-HPV test. The conventional Pap-smear has been proven to represent a critical tool to diagnose cervical pre-neoplastic lesions and cervical early stage cancer. Its effectiveness has been further enhanced after the introduction in current clinical practice of LBCC. The Pap-test remains, however, an exam whose diagnostic results are profoundly influenced by human subjectivity (high inter and intra operator variability referring to diagnostic criteria) and by the level of expertise/tiredness of screeners. Currently, molecular identification of high-risk HPV genotypes (hr-HPV test) is used as primary test in several cervical cancer screening programs, because its ability to identify high grade intra-epithelial cervical lesions is considered statistically superior than the cytological one. However, the application of molecular HPV-test as primary cervical carcinoma screening tool unfolds several uncertainties. To date, several large available cervical cancer series have documented that HPV-test is negative in 10 to 19% of women with biopsy-confirmed cancer. The test power to detect cervical adenocarcinoma varies from approximately 32 to 100%, depending on the geographic region and tumor subtype. Human papillomavirus DNA is detected in 80 to 100% of the 3 most common histological subtypes of cervical adenocarcinoma (endocervical, endometrioid and intestinal subtypes), whereas it’s rarely detected in non-mucinous subtypes, such as clear cell, serous and mesonephric adenocarcinoma. In addition, the gastric type, which includes minimal-deviation adenocarcinoma, was shown to be unrelated to HPV infection. Cervical adenocarcinomas constitute about 5 to 27% of all cervical carcinomas: their number varies between different countries and it is globally increasing. Eventually it has been observed that Pap-test cervical carcinoma screening can occasionally detect endometrial carcinomas or endometrial atypical glandular cell (AGC); these pathological entities are negative using HPV-test and therefore, primary HPV-test screening, instead of Pap-test, may result in losing the possible benefits of early diagnosis of endometrial cancer. According to several strategies of cervical cancer screening, the algorithm connecting cervical cytology and hr-HPV test is still debated and it’s influenced by both economic available resources and patient characteristics (such as age; organized screening program versus spontaneous patient request of exam or occasional medical indication during gynecological visit – the so called “opportunistic screening” (OS)).

Starting from a single liquid based cervical cytology sample, several diagnostic algorithms are possible:

(a) the two tests can be always and simultaneously performed (co-testing);  
(b) it’s possible to initially search the virus presence (definition of “presence of infection”) and then it can
be performed LBCC on infected women, in order to verify ongoing cervical disease) 16;
c) there’s the possibility to perform firstly the Pap-test and secondly the molecular HPV-test in women with positive Pap-test result (this algorithm is recommended in women younger than 34 years, due to high incidence of sub-clinical infections in young women) 16;
d) it can be performed firstly the Pap-test and then the HPV-test only in women presenting cytological atypia of undetermined significance (ASC-US; ASC-H; AGC referring to The Bethesda System) 61, so to determine if these cytological atypia are HPV-related. The Pap/HPV co-test is considered the best strategy for cervical carcinoma screening in women aged from 30 to 65 years, because, compared with HPV-test only, co-testing is more sensitive for the detection of lesions ≥ CIN3 40 62-64. Moreover, co-testing reduces the number of cervical carcinoma false negatives (particularly referring to glandular-type) which, instead, can occur using HPV-test as primary screening tool; co-testing allows to occasionally detect also endometrial adenocarcinoma or metastasis. Using HPV-test for primary cervical carcinoma screening exposes the patients to false negative results and non-detected patients HPV-infected do not undergo immediately Pap-test, but they are submitted to second round screening, always using HPV-test, after 5 years. This protocol might expose them to the risk of developing the so called “interval cancers” 65.

Our study, conducted on a population of women aged between 16 and 73 years, deriving from spontaneous screening, has demonstrated that there aren’t statistically significant differences between HPV-test results from 3 different diagnostic platforms currently used for cervical carcinoma screening (HC2 versus Onclarity: p = 1,54; Onclarity versus Xpert: p = 0,12; HC2 versus Xpert: p = 0,31). Positive case percentage for high-risk HPV-DNA genotypes, acquired through the three diagnostic platforms used and with a single vial to collect and store the samples, varied from 12,27 to 15,07% (Δ = 2,8%, not statistically significant) and this percentage is comparable with that deriving from previous studies and with the use of different sample vials. Italian average rate of positive HPV-test result during organized screening programs in women aged from 25 to 64 years is between 6,7% and 7,9% 66 67, but variability range among different screening programs is between 4,3 and 13,9% 67. Besides, it has to be considered that the population of our study consisted of 35% women with less than 34 years and that in this age group HPV-test positivity rate can even reach 34,4% 68.

HPV-test is nowadays regarded as an “objective test”, free from diagnostic errors, whereas it’s stressed the intrinsic “subjectivity” of cytological diagnosis, strongly operator-dependent. This pervasive opinion has contributed to the decision of using HPV-test as the primary screening tool in women aged 34 to 64, using the Pap-test as a “clinical triage” only in HPV-test positive women. As far as our samples are concerned, when on the same sample different diagnostic platforms performing HPV molecular research are used, it’s possible to notice discrepancies among the results: non-concordance diagnostic rate varied from 2,20 to 3,14%. Especially, in 11 women (2,55% of cases), one of the two diagnostic platforms compared has detected hr-HPV (representing a screening positive case), while the other has not detected hr-HPV (representing a screening negative case). Moreover, the re-analysis with PCR has revealed in other three cases, negative using HC2 test, the presence of hr-HPV genotypes: yet, this discrepancy could be due to the sensitivity of HPV-test methods committed to screening programs, which is deliberately inferior than PCR/gene sequencing, nevertheless it’s notable that the Pap-test of each of the three women was positive for HPV-related lesions and that all the women were older than 35 years. These elements point out that even the molecular diagnostic approach could give false negative results, being sometimes unable to identify in the screened population women affected by cervical HPV-related lesions. A screening program based on hr-HPV-test performed on 100,000 women could in theory produce about 2.500 false negative diagnoses. This observation should lead to a serious consideration, that is to compare the costs of HPV screening test, which uses the Pap-test only as a clinical triage in positive patients, with the costs deriving from assisting and curing women with “interval cancers”. These costs should also be examined in view of those of other screening strategies, such as: a) co-testing “DNA HPV-test/LBC Pap-test” screening; b) LBC-Pap test screening to identify patients with ongoing disease, and consecutive RNA-HPV test for prognostic evaluation and treatment protocol definition in women with cervical lesion. Recent studies have highlighted that Human Papillomavirus E6/E7 mRNA test has a significantly higher specificity and overall accuracy for HSIL or worse lesion than HPV-DNA test and that, therefore, it may be useful in clinical risk management 69, particularly in women younger than 35 years 70. It has also been observed that MiR-21-5p upregulation, MiR-34a downregulation and human telomerase RNA component (hTERC) amplification are associated with aggressive progression of CC 71, suggesting that these genetic markers could be usefully employed in screening programs for the triage of LBC Pap-test positive patients.

In our selected population, the most detected genotype was genotype 16 (20,99%), while genotype 18 was among the less observed (4,94%). It has been noticed a prevalence higher than 10% for genotypes 56-59-66 (Onclarity “P3” group) (17,28%), for genotype 31 (14,81%), for genotype 51 (12,34%) and for genotypes 35-39-68 (Onclarity “P2” group) (11,11%). This remark stresses the importance of HPV genotypes’ distribution continuous monitoring in the population, given that HPV vaccines planned in Italy in the “National vaccination prevention program 2017-2019” are the “bivalent one” (against genotypes 16 and 18) and the “quadrivalent one” (against genotypes 16, 18, 6 and 11) 72.
necessity to improve the screening program to identify cervical carcinomas and pre-neoplastic cervical lesions, even if there’s an ongoing vaccination program, is remarked by the detection during HPV-test of co-infection from more genotypes. The risk of cervical cancer development might be associated with type-specific interactions between genotypes in co-infections and genotypes not targeted by quadravalent vaccine confer 2.94-fold higher risk of cervical carcinoma. Among the samples analyzed with screening diagnostic platforms, our study observed 8.76% of prevalence of co-infection from more hr-HPV genotypes, but this percentage is surely underestimated, because only in a small proportion of HPV positive samples it has been possible to verify the single infectious genotypes. Moreover, the re-examination of 55 cases with PCR and sequencing of gene L1 and genes E6/E7 pointed out that co-infections can be determined by high-risk HPV genotypes but also by intermediate or low risk ones. The prevalence of multiple genotypes (double, triple and quadruple genotypes) in women older than 18, married and clinically symptomatic (abnormal vaginal bleeding/discharge, pain during coitus, lower abdominal pain and clinician suspicion of cervical malignancy) resulted noticeably higher (23.41% of 346 cases) than the prevalence observed in our population. Even if in Italy, in the target population of organized screening programs, the incidence of co-infections is probably inferior (giving the fact that Italian screening programs do not include very young women), the possibility that co-infections might increase the risk of development of cervical carcinoma, also in women HPV-vaccinated, is real and represents another valid reason to perform organized screening programs in the population at risk. An ancillary point deriving from our study is related to the different “gathering method” in “P groups” of various genotypes detected by diagnostic platforms available to date in commerce. Just taking into consideration as an example BD Onclarity™ HPV test e Xpert® HPV (Cepheid), it’s evident how it can be dangerous to report into clinical records of patients with positive HPV-test, only the “P group”, without specifying the method used: in this particular example two women, one diagnosed using Onclarity method and the other using Xpert, who are both “P1” positive, are de facto infected by different genotypes: the first by genotype 33 or 58, the second by genotype 16. It’s therefore important that anyone who is in charge of this diagnostic analysis promotes among clinicians the adequate rendition of the laboratory’s data in the patient records, reporting both the diagnostic result and the method through which it has been obtained.

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DETECTION AND GENOTYPING OF HPV-DNA THROUGH DIFFERENT TYPES OF DIAGNOSTIC PLATFORMS IN LIQUID BASED CERVICAL-CYTOLOGY SAMPLES

Unclassified sex cord/gonadal stromal testis tumor with a “pure” spindle cell component: a case report

C. SPAIRANI1, S. SQUILLACI2, A. PITINO1, F. MONTEFIORE3, W. FUSCO1
1 Division of Anatomic Pathology, Hospital “San Giacomo”, Novi Ligure (AL), Italy; 2 Division of Anatomic Pathology, Hospital of Vallecamonica, Esine (BS), Italy; 3 Division of Urology Hospital “San Giacomo”, Novi Ligure (AL)

Key words
Sex cord/gonadal stromal tumors • Spindle cells • Immunohistochemistry

Summary
Unclassified sex cord/gonadal stromal tumors (SCSTs) composed predominantly of spindle cells are rare. Very few cases have been documented to date. Here, we report a case of “pure” spindle cell tumor of the left testis in a 83-year old man whose morphological and immunohistochemical findings were consistent with a diagnosis of unclassified SCST and review the literature. Owing to the spindle cell pattern, the differential diagnosis with other benign and malignant spindle cell lesions is discussed.

Introduction
The occurrence of benign and malignant spindle cell tumors of the testis is rare. Apart from the well-known possibility that soft tissue and immune accessory-type spindle cell neoplasms can uncommonly arise from the male gonad, Leydig/Sertoli or granulosa cell tumors may only rarely contain spindle-shaped differentiation of the neoplastic elements 1,2. Conversely, a spindle cell predominant component has been reported in few cases of tumors belonging to the wide group of “unclassified sex cord-stromal tumours” according to WHO 2016 classification 3. These rare neoplasms are usually predominantly composed of spindle cells with intermixed occasional epithelioid elements and/or Leydig cells 4,5. To date, only three (3 out of 6) of such cases were entirely formed by spindle cells 4,6. Herein, we report the clinical and pathologic features of a new case of “pure” spindle cell tumor arising in the testis, consistent with the designation of “unclassified sex cord-stromal tumor”; we discuss problems in the differential diagnosis, and review the pertinent literature too.

Case report
In February 2015, a 83-year-old man was admitted to the Division of Urology of Hospital “San Giacomo”, Novi Ligure (AL), because of a slow-growing, painless, firm nodule in his left testis of several months’ lasting. Apart from diabetes mellitus and hypertension, he also had a past history of prostatic nodular hyperplasia, diagnosed by microscopic examination of the specimen obtained from transurethral resection. Urinalysis, liver enzymes, serum alpha-fetoprotein, and serum beta-human chorionic gonadotropin were all within normal limits. Scrotal ultrasound revealed a well-circumscribed, heterogeneous, prevalently solid with some cystic lacunes, nodular mass in the left testis, measuring 7 x 7 x 5.5 cm. A magnetic resonance imaging confirmed the presence of a mass with near-complete replacement of the left testis (Fig. 1). The patient underwent radical orchiectomy.

Authors’ note
This work was presented in abstract form (poster presentation) at the 7th Three-year Congress of Anatomic Pathology, Genova, Italy, November 23-26, 2016.

Correspondence
Salvatore Squillaci, Division of Anatomic Pathology, Hospital of Vallecamonica, via Manzoni 142, 25040 Esine, Italy - Tel. +39 0364 369256 - Fax +39 03643 69257 - E-mail: salvatore.squillaci@outlook.it
The neoplastic cells had indistinct pale to eosinophilic cytoplasm, and exhibited round to elongate, vesicular in appearance, nuclei with frequent tapered ends, inconspicuous to small nucleoli and occasional grooves (Fig. 3A). At times, small, tight clusters of tumor cells with more dark nuclei and scant cytoplasm stood out in some examined sections. Mitotic figures were occasionally observed, ranging from 0 to 1 per 10 high-power fields. At the periphery of the neoplasm seminiferous tubules were not trapped by the tumor cells and the surrounding testicular parenchima appeared to be normal (Fig. 2B). There was no evidence of necrosis, hemorrhage and vascular invasion. At immunohistochemical examination, tumor cells stained brightly and diffusely for vimentin and S-100 protein; staining for smooth-muscle actin and inhibin was intense but more irregular, confirming the diagnosis of unclassified SCST with a "pure" spindle cell component (Figs. 4 A-D). Furthermore, the tumor had a low MIB-1 index (< 5%). Stains for epithelial markers (EMA, Cam 5.2 and AE1/AE3 cytokeratins), desmin, CD34, CD99, Melan-A and PLAP were uniformly negative. Follow-up after 45 months was negative for recurrence or metastasis.

**Discussion**

Sex cord/gonadal stromal tumors (SCSTs) are a heterogeneous group of neoplasms that account for approximately 5% of all primary testis tumors including various histologic subtypes as Leydig cell tumors, that are the most common, together with Sertoli cell tumors, adult and juvenile granulosa cell neoplasms and mixed tumors. There remains, however, a small group of neoplasms, predominantly spindle cell in their morphology, that are more difficult to classify and as a result, have been described under a variety of names, mostly under the uncommitted term “unclassified sex cord/stromal tumor”. From the published data to date, including those of our patient, among the seven cases of SCST with a predominant spindle cell pattern, only four were completely formed by spindly elements (Tab. I). It is noteworthy that the histological and immunohistochemical features of spindle cell tumors similar to the present case, closely resemble those previously reported for “testicular myoepithelioma” and “testicular fibroma of gonadal stromal origin with or without minor sex elements”. Taking the former into consideration, the term “myoepithelioma” was suggested for ultrastructural findings of a dual, epithelial and myoid (peritubular myoid cell) differentiation. Following the original description in 1991 by Weidner, Du et al. and Kao et al. further delineated a rare testicular neoplasm, distinctly different from other SCSTs, under the term “myoid gonadal stromal tumor”, with less than 10 examples reported to date. Discussion of the categorization of these tu-
mors is beyond the scope of this case report; however, it is also important to mention that the 2016 WHO classification of testicular non-germ cell tumors stated that myoid gonadal stromal tumors and unclassified spindle cell-predominant SCSTs were separate entities. Reticular stains might be helpful in distinguishing them as it envelops groups of sex cord cells (which may be morphologically incospicuous) in the latter. In our case, microscopically, the testicular lesion seemed to fit the morphologic criteria for unclassified SCST with a “pure” spindle cell component, including reticular framework, immunopositivity for S-100 protein, smooth-muscle actin, and inhibin. Co-expression of S-100 protein (7/7) and smooth-muscle actin (7/7) is typical of unclassified SCSTs with predominance of spindle cells. Although inhibin expression was not often previously reported in this type of neoplasm, moderate to diffuse staining for this marker was recently noted in 3/3 studied cases, including the present one. In addition, these tumors were consistently negative for cytokeratins (AE1/AE3) (0/7), including the present case. It is deserving to be noted that nuclear grooves in tumor cells were often observed (5/7).

As some morphological features of unclassified SCSTs with a spindle cell component predominance, mainly nuclear grooves, and their immunophenotype were almost identical to the phenotype of granulosa cell tumors, Renshaw et al. regarded the former as a poorly differentiated variant of the latter. Focal expression of Melan-A in two tumors was consistent with incomplete Leydig cell differentiation and, the co-existence, in one of them, of some morphological and immunohistochemical features indicative of granulosa cell differentiation was also found. The histogenesis of these tumors is still unclear and a larger number of studies is needed before a conclusion about this topic can be drawn.

Unclassified SCSTs with a predominant or pure spindle cell component need to be distinguished from a wide variety of benign and malignant monomorphic spindle cell tumors, analogous to their soft-tissue counterparts, rarely arising from the testis. Immunohistochemically, a moderate reactivity for inhibin was crucial in recognizing our case as unclassified SCST with a “pure” spindle cell component and, accordingly, in excluding benign tumors (leiomyomas, angioleiomyomas, benign periphr-
eral nerve sheath tumors) and sarcomas with a bland-looking spindle cell pattern (low-grade fibrosarcomas, low-grade myofibroblastic sarcomas, inflammatory myofibroblastic tumors, follicular and interdigitating dendritic cell neoplasms) 29. The remaining malignant high-grade spindle cell tumors, such as malignant fibrous histiocytoma and rhabdomyosarcoma, can be easily distinguished on the basis of overt cytological atypia, high mitotic index, atypical mitoses, necrosis and vascular invasion 1.

All the reported cases of testicular unclassified SCST with predominant or pure spindle cell component pursued an innocent course, even though one of them exhibited worrying histologic features (3 mitoses per 10 HPF) 4. Follow-up information was available in all patients with unclassified SCST with predominance of spindle cell component for a mean period of 19.7 months (range = 1 month to 60 months, median = 12 months) 4-6. Pathologic findings suggestive of a high probability of malignant behaviour include large tumor size, infiltrative margins, evidence of lymphatic or vascular invasion, tumor necrosis and high mitotic rate 17. Nevertheless, it is difficult to predict with certainty the clinical behaviour of unclassified SCST with predominant or pure spindle cell component based on histology, also because metastases may occur many years later 1. Thus, as for other SCSTs, careful and long-term follow-up is mandatory for all cases.

Summing up, we report a case of unclassified SCST arising in left testis with a “pure” spindle cell neoplastic component. The clinical history, morphological findings, and immunohistochemical profile support the diagnosis, but the experience with this extremely rare tumor is, however, very limited and thus the proper prognostic features as well as optimal management and treatment have not yet been stated. Additional studies on more large series, collecting long-term follow-up results, are needed to determine the behaviour of these tumors, as this category of SCSTs might have a more favorable prognosis.

References


Calcifying aponeurotic fibroma (CAF) is a very rare tumor of the extremities, which can be difficult to diagnose due to its wide cytoarchitectural pattern. We herein report the clinicopathologic features of a case of CAF localized on the dorsal face of the foot in a 5-year-old male child, diagnosed by needle core biopsy. Differential diagnostic problems are discussed. The present case emphasizes that the diagnosis of CAF can be confidently rendered on core needle biopsy if the main morphological components of this tumor are concurrently present; however, before making the diagnosis of CAF, the clinical and radiological context should be considered.

Calcifying aponeurotic fibroma: a core biopsy-based diagnosis

F. MOTTI, S. SCAVO, G.M. VECCHIO, G. FUCCHIO-SANZÀ, F. NICOLOSI, G. MAGRO

1 Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, Anatomic Pathology, University of Catania, Italy; 2 Sezizione di Dermatologia - ASL 3 Catania, Acireale (CT), Italia; 3 U.O. Ortopedia e Traumatologia, ARNAS Garibaldi, Presidio Garibaldi Nesima, Catania, Italia

Key words
Calcifying aponeurotic fibroma • Needle core-biopsy • Histological diagnosis • Immunohistochemistry

Summary
Calcifying aponeurotic fibroma (CAF) is a very rare tumor of the extremities, which can be difficult to diagnose due to its wide cytoarchitectural pattern. We herein report the clinicopathologic features of a case of CAF localized on the dorsal face of the foot in a 5-year-old male child, diagnosed by needle core biopsy. Differential diagnostic problems are discussed. The present case emphasizes that the diagnosis of CAF can be confidently rendered on core needle biopsy if the main morphological components of this tumor are concurrently present; however, before making the diagnosis of CAF, the clinical and radiological context should be considered.

Introduction

In 1953 Keasbey described a tumor characteristically occurring in the palms and soles of young children, for which the term “juvenile aponeurotic fibroma” was proposed. Subsequent studies revealed that this tumor can occur, not only at any age, but also in various sites including wrist, forearm, elbow, upper arm, neck, abdominal wall, lumbar paravertebral area, leg, ankle, and thigh. On clinical examination CAF presents as a single, firm, painless, slowly growing mass occurring more frequently in males. Multiple lesions have also been reported in the literature. Given its tendency to local invasion, typically into the surrounding fascia or muscle, a high local recurrence rate (up 50%) can be documented after surgical excision. Malignant transformation into fibrosarcoma has also been rarely reported. Gross examination reveals a whitish mass with irregular borders, usually attached to fascia or tendons. The cut section shows a fibrous surface with gritty foci of calcifications. Histologically, two distinct components can be identified: i) fibromatosis-like proliferation of bland-looking spindle shaped cells set in a fibrous stroma; ii) nodules, some of them with chondroid metaplasia, variably calcified, and usually rimmed by plump round to epithelioid cells, as well as multinucleated osteoclast-like giant cells.

Some authors suggested the existence of different phases of tumor growth. The early phase is characterized by high cellularity, with neoplastic cells mainly arranged in fascicles; chondroid or immature cartilaginous foci are present but calcifications are lacking or only focally detectable. Mitoses may be present. Later, the deposition of granular calcifications in the nodular tumor component predominates. During this phase, the calcified areas are surrounded by plump round/epithelioid fibroblasts and osteoclast-like multinucleated giant cells. In the last phase, tumor is predominantly hypocellular with a diffusely fibrotic and calcified stroma. These different histological phases explain the heterogeneous appearance on radiological imaging, which is variable according to the patient’s age, presence of calcifications and osseous involvement. On radiography, CAF appears as a soft tissue mass, with or without fine stippled calcification. Signs of bone involvement, such as scalloping of the cortex and adjacent bone erosion, may be seen. Computed Tomography (CT) scan is helpful to detect the calcified areas. Although magnetic resonance (MRI) is the most accurate tool in the evaluation of soft tissue tumors, MR imaging
features of CAF are not available in the literature, with rare exceptions. Kwak et al. reported a case of radiologically “non-calcified” CAF in a young child, and described the MRI findings as an intensity signal lower than that of muscle on T1WI and T2WI. Hasegawa et al. demonstrated subcutaneous distribution, tendency to local infiltration, close location to fascia or tendon, and heterogeneous enhancement after gadolinium contrast injection. They stated that the low signal intensity on T2WI was attributed to the fibrous component and little cellularity of the mass and this finding is more commonly seen during the initial phase.

We herein report the clinicopathologic features of a case of CAF localized on the dorsal face of the foot in a 5-year-old male child, emphasizing the possibility of needle core biopsy-based diagnosis. Although in one case the diagnosis of CAF was suspected by fine needle aspiration biopsy (FNAC), to the best of our knowledge, this is the first case of CAF diagnosed pre-operatively by needle core biopsy.

**Case report**

A 5-year-old male child presented with a firm, palpable and painless mass on his left forefoot. CT scans revealed a swelling mass, about 17 x 17 mm in size, with elastic consistency and separated from adjacent skeletal segments. The deep component of the lesion was sited between the shaft of the II and III metatarsal bones. Multiple inert calcifications were seen. RMI imaging showed an expansile, oval-shaped subfascial mass, measuring approximately 22 mm in its greatest dimension, hypointense on T1-weighted images, hyperintense on the T2-weighted images, with regular margins delimited by a hypointense rim. The heterogeneous contrast enhancement is due to the simultaneous presence of both hypointense and hyperintense areas likely due to calcifications. No signal abnormalities of bone structures was appreciable (Fig. 1). Clinically and radiographically the mass was interpreted to be suspicious for sarcoma. Fine needle core biopsy was performed.

**Materials and methods**

Bioptic and the relative surgical specimens were fixed in 10% buffered formalin, routinely processed and embedded in paraffin. Two sections were stained with haematoxylin and eosin while additional sections were cut for immunohistochemical procedures. Immunohistochemical studies were performed with the labeled streptavidin-biotin peroxidase detection system using the DAKO automated immunostainer (Glostrup, Denmark). The following antibodies were tested: vimentin (dilution 1:100), α-smooth muscle actin (dilution 1:200), desmin (dilution 1:100), myogenin (dilution 1:100), S-100 protein (dilution 1:500), CD34 (dilution 1:50), pancytokeratins (dilution 1:50), β-catenin (dilution 1:100), EMA (dilution 1:100); INI-1 (dilution 1:100) all from DakoCytomation, Glostrup, Denmark.

**Pathologic findings**

Core biopsy specimen revealed a proliferation of spindle to stellate cells intermingling with multinucleated osteoclastic-like giant cells and set in a fibrous stroma (Fig. 2a-d). The cells showed only a mild nuclear atypia, while mitoses and necrosis were absent. Notably microcalcifications were scattered throughout the fibrous stroma (Fig. 2d). Immunohistochemical analyses showed a diffuse staining only for vimentin. No immunostaining was obtained with α-smooth muscle actin, desmin, myogenin, S-100 protein, CD34, pancytokeratins and EMA. The proliferation of spindle- to stellate-shaped cells with a fibroblastic profile, intermingling with osteoclastic-like cells, set in a fibrous stroma was consistent with the diagnosis of CAF. This suspicion was also supported by the age and the site of the tumor mass. However it was recommended to clinicians of evaluating histologically the entire tumor mass after complete surgical excision. Grossly the surgically-resected tumor presented as a well-circumscribed, 3 cm nodule, firm in consistency and whit-
ish in color. The cut surface showed a fibrous mass with gritty foci of calcifications. Histologically, at low magnification, tumor had a multinodular appearance with hypercellular areas alternating with more hypocellular fibrous areas (Fig. 3a-c). Notably some nodular areas were extensively calcified (Fig. 3c). The neoplastic proliferation was focally present at the resection margins (Fig. 3d). Higher magnification showed highly cellular areas composed of bland-looking spindle cells arranged in long, variably intersecting, fascicles closely reminiscent of desmoid-type fibromatosis (Fig. 4a). These areas were variably blending with hypocellular fibrous areas in which the cells adopted a stellate shape (Fig. 4b-d). Notably a significant number of multinucleated osteoclastic-like giant cells were variably intermingling with the neoplastic cells (Fig. 4b). Mitotic count was low (up 2 mitoses x 10 high power field). Osteoid and/or cartilaginous matrix was absent. Nuclear pleomorphism, atypical mitoses and necrosis were absent. Immunohistochemically neoplastic cells were diffusely stained with vimentin and focally with alpha-smooth muscle actin and WT1 (cytoplasmic staining alone) (Fig. 5). Based on morphological and immunohistochemical features, the diagnosis of CAF was rendered.

Discussion

Recently young-type fibromatoses have been interpreted as an unique group of benign or intermediate, non-metastasizing fibroblastic- myofibroblastic proliferations. Their clinical behavior ranges from a self-limiting mass to locally recurrent tumor. Fibroblasts and myofibroblasts are present in variable proportions and arranged in a wide variety of growth patterns in each single entity and they are usually interspersed in a predominantly fibrous stroma. Their morphological heterogeneity likely reflects their phenotypic plasticity. The following clinicopathologic entities are currently included in the category of young-type fibromatoses: infantile myofibroma (myofibromatosis), fibromatosis colli, infantile digital fibromatosis, fibrous hamartoma of infancy, calcifying aponeurotic fibroma, lipofibromatosis, juvenile nasopharyngeal fibroma, hyaline fibromatosis (juvenile and infantile types). The immunohistochemical expression of vimentin, alpha-smooth muscle actin and WT1 is helpful in revealing the fibroblastic/myofibroblastic profile shared by all these entity.

CAF is a very rare tumor which can be difficult to diagnose due to its wide morphological spectrum. Accordingly differential diagnostic problems may arise with a variety of benign and malignant soft tissue tumors. Unfortunately, radiologic features of CAF, including those of the present case, are not specific, and thus the correct diagnosis is still based on the histological examination. Although the diagnosis of CAF is usually straightforward in surgically-resected specimen if all the diagnostic clues are present, it may be challenging when evaluating needle core biopsies. This is mainly due to
Fig. 3. Surgical specimen. (A) Low magnification showing a tumor with a multinodular appearance, attached to tendon fascia (arrows). (B) Tumor showing hypercellular areas alternating with more hypocellular fibrous areas. (C) Notably some tumor nodules were extensively calcified. (D) The neoplastic proliferation was focally present at the resection margins (tendon fascia).

Fig. 4. Surgical specimen. (A) Higher magnification showing highly cellular areas composed of bland-looking spindle cells arranged in a fibromatosis-like pattern. (B) Hypercellular areas blending with hypocellular areas containing multinucleated osteoclastic-like giant cells. (C) This area shows the different morphological phases of CAF: hypercellular areas blending into hypocellular and more extensively fibrotic areas with microcalcifications. (D) Tumor area in which spindle-, stellate-shaped cells and multinucleated giant cells coexist.
the fact that CAF often shows a wide variety of cellular composition and growth patterns. Apart from morphology, the patient’s age, tumor site, and radiological features are very helpful for a correct pre-operative diagnostic approach.

We herein report the first case of CAF occurring in the foot of a 5-year-old male child and diagnosed pre-operatively by needle core biopsy. Differential diagnosis on needle core biopsy mainly included desmoid-type fibromatosis, tenosynovial giant cell tumor (localized type) \(^7\) \(^11\) \(^15\) \(^17\), and epithelioid sarcoma \(^17\) \(^18\). Differentiating CAF from desmoid-type fibromatosis can be difficult as the latter can arise in the soft tissues of the proximal extremities. In addition CAF usually contains hypercellular areas which look like desmoid-type fibromatoses \(^7\) \(^11\) \(^15\). In our case the majority of the cells had a spindle shape but they were not arranged in distinct fascicular pattern as they do in desmoid-type fibromatosis. In addition the presence of osteoclast-like multinucleated giant cells and calcifications, along with the absence of immunostaining for \(\beta\)-catenin, argued against the diagnosis of desmoid-type fibromatosis \(^7\) \(^11\) \(^15\). Tenosynovial giant cell tumor, localized type \(^7\) \(^11\) \(^17\), shares some clinical features with CAF in that both lesions present as asymptomatic, slowly-growing masses attached to the tendon sheath and/or joint capsule. Magnetic resonance imaging is helpful in the differential diagnosis as CAF shows speckled calcifications and ill-defined margins, whereas the former has lobulated and well-defined margins \(^11\) \(^14\) \(^17\). In addition erosions of the adjacent bone can be observed in tenosynovial giant cell tumor \(^17\). Although the presence of multinucleated giant cells in the needle core biopsy of our case was reminiscent of tenosynovial giant cell tumor, it was ruled out as the characteristic mononuclear cells, foamy histiocytes and siderophages were lacking \(^7\) \(^11\). The spindle cell component seen in our case is not a feature of tenosynovial giant cell tumor \(^17\). In the present case the identification on needle core biopsy of spindle cells set in a fibrous stroma containing microcalcifications may arise diagnostic problems with epithelioid sarcoma, spindle cell variant \(^17\) \(^18\). However this malignant tumor was ruled out by immunohistochemistry that showed INI1 expression, as well as no immunostaining with both cytokeratins and EMA \(^7\) \(^18\).

The present case emphasizes that the diagnosis of CAF can be confidentially rendered on core needle biopsy if the main morphological components of this tumor are concurrently present: proliferation of spindle to stellate cells set in a fibrous stroma containing osteoclast-like giant cells and microcalcifications. Immunohistochemistry is helpful in revealing the fibroblastic/myofibroblastic profile of the neoplastic cells \(^7\) \(^16\). Before making the diagnosis of CAF, the clinical and radiological context should be taken in consideration by pathologists. However we suggest that the final diagnosis of CAF should be rendered on the surgically-excised nodule.

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**Fig. 5.** Immunohistochemical findings in surgical specimen. (A) Neoplastic cells were diffusely stained with vimentin and only focally with alpha-smooth muscle actin (B). (C) WT1 immunostaining was restricted to cytoplasm of neoplastic cells. (D) INI1 was diffusely expressed in the nuclei of neoplastic cells, ruling out the diagnosis of epithelioid sarcoma.
References

Case report

Lymphnode metastasis of thyroid cancer misinterpreted as lateral aberrant thyroid 40 years before identification of primary tumor. Case report and review of the literature

G. RIVA1, M. VILLANOVA1, G. FRANCIA2, G. VALOTTO1, L. MEZZETTO1, M. TOAIARI1, A. ECCHER1, L. NOVELLI1

1 Department of Pathology and Diagnostic, University and Hospital Trust of Verona, Italy; 2 Department of Endocrinology, Pederzoli Hospital, Peschiera del Garda (VR), Italy; 3 Department of Pathology and Diagnostic, Careggi University Hospital, Firenze, Italy

Key words
Aberrant thyroid • Occult thyroid carcinoma • Ectopic thyroid

Summary

The differential diagnosis between lateral ectopic thyroid tissue with orthotopic normal gland and metastatic thyroid carcinoma is challenging. Lateral cervical site is a very rare location for ectopic tissue since only a few cases have been reported. The peculiarity of this clinical case is the finding of a thyroid carcinoma forty years after surgical resection of the ectopic thyroid lesion. This asynchronous association, never reported in literature, raises the question of the differential diagnosis between a true ectopic aberrant thyroid and an early lymph node metastasis from an occult thyroid carcinoma, evident in the primitive site many years later. Several elements, which will be matter of discussion, seem to favour the latter hypothesis. This case, although isolated, suggests that any lateral cervical mass, comprising thyroid tissue, should be regarded as a metastasis of thyroid carcinoma until proven otherwise. Carefull investigation of thyroid gland is mandatory.

Case report

Ectopic thyroid was described for the first time by Heller in 1749. The prevalence is about 1 case per 100,000-300,00 people. This rare condition is characterized by the presence of thyroid tissue outside the usual location of the normal thyroid, mostly in the pretracheal position, without anatomical and vascular connection with the gland. Lateral cervical site is a very rare location for ectopic tissue to occur, since only few sporadic cases have been reported in literature. It is so rare and for this its presence as a nosological entity is questionable. On the other hand, according to literature, most cases of regional neck lesions associated to a clinically normal thyroid gland at first evaluation, should be considered to be metastases of occult thyroid carcinoma. Our case provides the opportunity to discuss this topic.

We report the case of an 74-year-old man, who presented, in 2014, with a left lateral-cervical mass that he noticed two months earlier. He was affected by non Hodgkin B lymphoma, diagnosed in 2010 and since then in follow-up. His past medical history revealed that in November 1974 he underwent to an excision of a 3-4 cm left lateral-cervical lesion. The surgical examination of the mass did not reveal any anatomical connection with normal thyroid and the lesion appeared firmly adherent to internal jugular vein, connected by small and short vessels. The carotid artery and the vagus nerve were not involved. Pathological examination revealed the presence of thyroid tissue composed of normal hyperplastic cells in a follicular pattern, scanty colloid and massive infiltration of lymphocytes, often arranged in lymphoid follicles. On the basis of these findings the final diagnosis was “ectopic aberrant lateral-cervical thyroid with basedowian histological appearance”. At that time physical examination of the thyroid gland was normal and patient did not show any symptom of thyroid dysfunction. Neither endocrine assessment nor imaging study were carried out. In the following years no clinical and/or instrumental follow-up was performed. Forty years later, in 2014, a
new mass, in left lateral-cervical region became evident. Neck ultrasound imaging showed several lymph nodes on the left side, the greater of which had a main axis of 2 cm with rough calcifications. Furthermore a 1 cm nodule, in the left thyroid lobe, showed a highly suspicion ultrasound appearance. Cytology by fine needle aspiration demonstrated features consistent with papillary thyroid carcinoma. The patient underwent to a total thyroidectomy and left lateral neck compartmental lymph node dissection. Histology confirmed the presence of a multifocal follicular variant of papillary carcinoma (FVPC) localized in the left thyroid lobe (Fig. 1), with capsular infiltration and involvement of the perithyroidal soft tissues. Three out of 19 lymph nodes were positive for metastasis. No lymphocytic infiltration of thyroid tissue was observed. We retrieved the previous case from our archive. It’s revaluation was consistent with a nodal metastasis from papillary thyroid carcinoma. The patient underwent to a remnant ablation therapy with 100 mCi 131 I after rhTSH, with excellent biochemical and structural response after one year of follow-up.

**Discussion**

Lateral ectopic thyroid is due to an aberrant development of the gland, that happens when the cells of lateral part do not join those of the median one 1. Its occurrence is usually in the submandibular region and, less frequently, in the para-jugular or para-carotid region 2-3. This event is far rarer than lingual thyroid, which represents 90% of all cases of ectopic thyroid. Differential diagnosis with lymph node metastasis of thyroid cancer can be very challenging. The cases of ectopic thyroid detected in the lateral cervical region were, in the past, usually interpreted as malignant metastatic lesions 4-6. This event is far rarer than lingual thyroid, which represents 90% of all cases of ectopic thyroid. Differential diagnosis with lymph node metastasis of thyroid cancer can be very challenging. The cases of ectopic thyroid detected in the lateral cervical region were, in the past, usually interpreted as malignant metastatic lesions 4-6. In the series of Nussbaum et al. 27 out of 197 patients with thyroid carcinoma (14%) presented with regional lymph node involvement, which was regarded as “basedowian picture” 7. The crucial problem is whether typical nuclear changes of PTC are always clearly evident in aberrant localizations. Diagnosis of FVPC relies on the typical nuclear alterations, the assessment of which is, in many cases, subjective and leads to a high inter observer variability, mainly when nuclear features are “border line” or faded 8-10. According to some authors it is not always possible, in cytology, separate FVPC from the other follicular lesions because of overlapping features or sporadic presence of nuclear abnormalities 11. The absence of the nuclear features characteristic of FVPC does not rule out the diagnosis since these can be very mild and scattered. Critical analysis of our case should take in account the important progress in medical knowledge and diagnostic methods made in the last decades. Firstly, in the seventies, imaging techniques as neck ultrasonography and computerized tomography were not yet widely available in clinical practice. At that time scintigraphy examination was the only method to assess functional status and morphology of the thyroid gland, as well as to detect ectopic tissue. In our case scintigraphy was not performed so that evaluation of thyroid relied exclusively on neck palpation, which did not show abnormalities. No symptoms related to thyroid dysfunction were recorded, despite the presence of massive lymphocytic infiltration, which was regarded as “basedowian picture”. This latter finding was more likely compatible with the presence of lymphoid tissue, harboring thyroid tissue of adenomatous appearance. Forty years later FVPC was diagnosed, with nodal metastatic involvement in the same site of the previous lesion. This clinical evolution suggests that the lateral mass excised in 1974 could be a metastasis from an occult FVPC, rather than an ectopic lateral thyroid. Besides we must take into account that, in the mid-1970s, FVPC was not yet broadly recognized as separated primary tumor 12-14. Moreover in 1974 not the whole tissue excised was included and evaluated. In conclusion we recommend that diagnosis of lateral ectopic thyroid should be made with great caution in cases of lateral cervical mass. Every lateral-cervical lesion containing thyroid tissue should be considered suspected until proven otherwise. In these cases, thyroid should mandatory be investigated from a clinical, radiological and pathological point of view and total thyroidectomy should be considered in the diagnostic route, in order to rule out an occult papillary thyroid carcinoma.
LYMPHNODE METASTASIS OF THYROID CANCER MISINTERPRETED AS LATERAL ABERRANT THYROID 40 YEARS BEFORE IDENTIFICATION OF PRIMARY TUMOR. CASE REPORT AND REVIEW OF THE LITERATURE

References


Pathologica ai tempi della Spagnola
(Pathologica in the time of “Spanish flu”)

C. PATRIARCA¹, C.A. CLERICI²
¹ Servizio di Anatomia Patologica, Ospedale St Anna, ASST Lariana, Como, Italia; ² Dipartimento di Oncologia ed Emato-Oncologia, Università degli Studi di Milano e SSD Psicologia Clinica, Fondazione IRCCS Istituto Nazionale dei Tumori, Milano, Italia

Riassunto

La pandemia di influenza “spagnola”, che in poche settimane fece in Italia tra le 350.000 e le 600.000 vittime durante l’autunno di cento anni fa, fu largamente discussa dalla comunità scientifica. In questo articolo riportiamo il dibattito avvenuto tra il 1918 e il 1923 all’interno della rivista Pathologica, a proposito delle basi anatomo-patologiche della pandemia e del confronto di dati e opinioni sull’eziologia della grande infezione.

Summary

The pandemic “Spanish flu”, that in a few weeks of the autumn 1918 caused in Italy a number of deaths between 350.000 and 600.000, was widely discussed by the scientific community, although very little of that debate leaked out, because of the military censorship.

In the present article we comment on the original papers describing the hemorrhagic pneumonia, and on discussions about the ideas of the origin of the pandemic infection (Pfeiffer bacillus, vs streptococcus or other bacteria vs a “viral hypotesis”) that occurred in Pathologica during and soon after that ominous pandemia.
tiva della rivista: “riassunto di recenti pubblicazioni” e “notiziario”;
4. il resoconto del congresso di Milano tenutosi dell’april- le del 1919, quando in realtà anche la terza ondata epi- demica dei primi mesi dell’anno si era oramai spenta.

Casistica anatomo-patologica

Nel numero del 15 giugno 1919 6 B. Polettini descrive i reperti di 50 autopsie eseguite all’Università di Pisa tra settembre e ottobre del ’18. Si trattava in 40 casi di indivi- due giovani, in gran parte militari e vi erano anche 5 donne in gravidanza. L’autore riferisce la monotonia dei reperti macroscopici: petecchie emorragiche alla cute del collo e sul torace. Iperemia dell’encefalo e del mi- dollo spinale. All’apertura del torace, scarso era il versa- mento pleurico e pericardico e se il cuore non mostrava segni particolari, a parte qualche petecchia epicardica, era il reperto polmonare ad essere più significativo: pe- tecchie su tutta la superficie pleurica e diffusa “epatiz- zazione” di entrambi i polmoni, con l’eccezione degli apici, ancora areati. Al taglio, colorito scuro ed aree emorragiche alternate ad aree più grigie ed epatizzate, talvolta purulente, mentre i bronchi si presentavano di aspetto congesto e a tratti purulento. Milza incrementata- ta di volume in pochi casi, surreni sempre emorragici e congestione della midollare renale completavano il qua- dro macroscopico.

Istologicamente si osservava lungo tutto il SNC la pre- senza di angetastie e talora di limitate emorragie periva- sali. Nei polmoni dominava l’emorragia alveolare, con rottura dei setti, e “desquamazione” dell’epitelio; ab- bondava il materiale fibrino leucocitario intra-alveolare e peribronchiale. L’autore riferisce anche che l’emor- ragia peribronchiale era tale a volte da creare un vallo di separazione dalla muscolatura mentre in altri campi microscopici l’essudato era tale che “i bronchi rimango- no mascherati”. Insomma Polettini descrive una bronco- polmonite a focolai confluenti a forte impronta emorra- gica peribronchiale, definendo l’alterazione bronchiale segmentaria “quasi irriconoscibile” e anche questo autore conclude in modo un po’ sconsola- to che si tratta di una broncopolmonite da piogioni senza alcuna specificità, se non per l’imponenza della flogo- si e dell’emorragia, e del tutto diversa dalle polmoniti franche lobari, o dalle polmoniti pestose, carbonchiose o da ifomiceti. Nel corso di queste 34 autopsie i patologi avviarono anche analisi colturali, identificando strep- tococchi, tra cui pneumococci, stafilococchi e bacillo di Pfeiffer. Gli autori si dichiararono scettici circa il ruolo eziologico del bacillo di Pfeiffer e concluso: “le no- stre scarse esperienze con filtrati di organi non permet- tono di poterci esprimere sull’importanza che qualche autore ha voluto attribuire all’azione patogena di specia- li virus fibrilabili”.

Eziologia dell’epidemia

È o non è influenza? titola il suo intervento su Patholo- gica D. Pacchioni 8 nel numero del 1 febbraio del 1919. Il titolo era quanto mai opportuno, perché se era a torto accettato da tutti l’aggettivo spagnola (la Spagna, neutra- le nella grande guerra, non censurò mai le notizie sull’e- pidemia in corso, col risultato di apparire agli occhi del mondo la nazione colpita per prima e più massivamente) ovunque si sollevavano obiezioni contro l’uso del termine influenza 2, anche su Pathologica 7, per indicare un’epi- demia dalle conseguenze tanto devastanti. Ricordiamole ancora: tra i 350.000 e i 600.000 morti in Italia, tra i paesi con la maggiore mortalità in Europa assieme al Portogal- lo 1. L’autore si dichiara scettico circa la cosiddetta ipotesi unica, ovvero la tesi secondo cui l’agente responsabile dell’epidemia in atto fosse lo stesso di quella sviluppatisi nel 1890 (la cosiddetta influenza russa, da molti attribuita all’Haemophilus Influenzae, isolato nel contesto di quel- la pandemia da Richard Pfeiffer). Quest’ipotesi si basava sull’osservazione, per altro esatta, che ad ammalarsi fos- sero in gran parte i giovani e dunque proprio gli individui che non potevano essere immunizzati nel corso dell’e- pidemia influenzale di 28 anni prima. L’autore, direttore della clinica pediatrica dell’Università di Genova, sottoli- nea tuttavia come a morire siano soprattutto i 30 e 40 anni, dunque individui entrate in contatto nella prima infanzia con l’agente responsabile di quella pandemia, che colpì a sua dire, e secondo i registri che pubblica nel suo articolo, anche nella fascia infantile. Mancano le faringiti e i dolori muscolari, così tipici dell’influenza, aggiunge Pacchioni. L’autore, che evidentemente era tra chi non credeva nel ruolo causale del bacillo di Pfeiffer a proposito dell’epi- demia del 1890, si esprime a favore del ruolo causale del bacillo di Pfeiffer nell’epidemia attuale, ma in una sua va- riente particolarmente virulenta e in simbiosi, “in associa- zione a delinquere” dice, con streptococchi.
Di diversa opinione è il batteriologo Mario Segale degli ospedali Galliera di Genova, nonché fondatore e direttore scientifico della rivista (per un excursus storico sul suo fondatore vedi www.galliera.it), che avvia le sue indagini già nel giugno del 1918 (all’epoca della prima ondata, meno aggressiva, di febbre spagnola) e pubblica i suoi risultati nel numero del 1 gennaio 1919. Specifica di avere ben presto escluso dagli studi il muco faringeo o il tessuto polmonare, perché troppo abbondante in quelle sedi era la flora batterica concomitante, e di essersi concentrato su colture da sangue delle cavità cardiache, da liquor cefalo rachidiano e da linfonodi ascellari, isolando quello che definisce come un nuovo tipo di streptococco emolitico, il c.d. streptococcus pandemicus capace a suo dire di riprodurre la broncopolmonite emorragica in diversi animali da esperimento, pur ammettendo che i suoi risultati non sono costanti né tanto meno probativi. Lo stesso autore torna sull’argomento con un lavoro originale nel numero del 15 maggio del ‘19 incentrato sullo streptococcus pandemicus e con una review del numero di Pathologica del 15 agosto del 1920. Facendo una rivalutazione della letteratura apparso in quei due anni, l’autore ammette che la massa di prove a sfavore di un ruolo causale del bacillo di Pfeiffer è preponderante; ad esse Segale aggiunge le proprie osservazioni dirette. Egualmente scettico è il prof. Segale a proposito del ruolo causale del bacillo di Pfeiffer, mentre più spesso viene indicato come responsabile lo streptococco emolitico, da solo o in associazione. Infine anche dalle rubriche di Pathologica viene a galla il possibile ruolo causale di un virus invisibile. Questo non impedisce all’anonimo redattore della rubrica, probabilmente lo stesso Mario Segale, che curava in prima persona gran parte dei testi delle rubriche, di presentare nel numero del 15 luglio 1921 un lavoro dell’università di Toronto a sfavore della genesi virale, uscito sul British Journal of Experimental Pathology, con queste parole: “La discussione sulla natura etiologica della grippe è inciampata, come i lettori hanno presente, nella dottrina dei virus filtrabili e tale è l’attrazione anche per gli uomini di scienza per il soprannaturale che i più insignificanti sperimentucoli vennero dichiarati fondamentali e si andò formando un ambiente assolutamente sfiduciato per ogni ricerca obiettiva al di là del presupposto. Storia di ieri. Appare ora un interessante lavoro (…) dove la questione dei virus filtrabili è ripresa a fondo e si dimostra che (…) le lesioni sono transmissibili in serie e sono in rapporto a qualche attività tossica che non è ben stabilita, ma certo indipendente dall’azione di un presunto virus specifico dell’influenza”. Sono affermazioni che possono colpire ma sarebbe da ingenui o da presuntuosi stupirsi di come la strada verso l’invisibile, i virus, banché correttamente postula da molti, sia stata tanto disseminata di obiezioni. Nelle rubriche vi sono inoltre brevi riassunti di lavori anatomo-patologici comparsi principalmente su Archives negli anni ’18 - ’20 e incentrati sui ben noti quadri di broncopolmonite emorragico-suppurativa, talmente massiva da essere caratteristica e definitiva, secondo le affermazioni del patologo Cesaris Damel nel corso del congresso del ’19 a Milano. Per finire nel 1923 Mario Segale torna sul tema del bacillo di Pfeiffer, ribattezzato per l’occasione bacillo di Pfeiffer-Gosio, in sintonia forse con il nuovo spirito autarchico del tempo e in onore del batteriologo italiano Bartolomeo Gosio direttore del Laboratorio centrale della Sanità Pubblica che aveva studiato approfonditamente questo germe e ne aveva sostenuto con forza il ruolo causale. In questo intervento Segale con dovizia di argomenti scagiona definitivamente l’Haemophilus Influenzae e dichiara insoluto il problema della causa microbiologica della grande pandemia del ’18.

La spagnola nelle due rubriche di Pathologica: “riassunto di recenti pubblicazioni” e “notiziario”

Fin dall’autunno del 1918 le rubriche della rivista ospitano decine di segnalazioni della diffusione della malattia. Vengono riportati dati epidemiologici dell’epidemia in corso da fonti Euroasiatiche, nordamericane, ma anche dal Brasile, all’India e al Giappone, che forniscono al lettore italiano informazioni “in tempo reale” sull’entità e sulla gravità dei focolai. Non mancano notizie dell’epidemia nemmeno dall’Alaska e dal Transvaal. Colpisce come già nel ’19, riferendo di articoli esteri (W.G. Mc Callum. Med Record, 10. 5-19) si segnalasse per l’anno prima “un numero fenomenale di polmoniti nei campi di istruzione militare degli USA”. Noi oggi sappiamo come quello sia stato uno dei principali focolai di partenza della pandemia, ma all’epoca ciò non contribui a disinnescare la fama di “spagnola” alla piaga in corso. Negli anni successivi troviamo nello spazio della rubrica di Pathologica decine di segnalazioni di studi eziologici in corso in tutto il mondo, in cui ricorre con fortune alterne il ruolo causale del bacillo di Pfeiffer, mentre più spesso viene indicato come responsabile lo streptococco emolitico, da solo o in associazione. Infine anche dalle rubriche di Pathologica viene a galla il possibile ruolo causale di un virus invisibile. Questo non impedisce all’anonimo redattore della rubrica, probabilmente lo stesso Mario Segale, che curava in prima persona gran parte dei testi delle rubriche, di presentare nel numero del 15 luglio 1921 un lavoro dell’università di Toronto a sfavore della genesi virale, uscito sul British Journal of Experimental Pathology, con queste parole: “La discussione sulla natura etiologica della grippe è inciampata, come i lettori hanno presente, nella dottrina dei virus filtrabili e tale è l’attrazione anche per gli uomini di scienza per il soprannaturale che i più insignificanti sperimentucoli vennero dichiarati fondamentali e si andò formando un ambiente assolutamente sfiduciato per ogni ricerca obiettiva al di là del presupposto. Storia di ieri. Appare ora un interessante lavoro (…) dove la questione dei virus filtrabili è ripresa a fondo e si dimostra che (…) le lesioni sono transmissibili in serie e sono in rapporto a qualche attività tossica che non è ben stabilita, ma certo indipendente dall’azione di un presunto virus specifico dell’influenza”. Sono affermazioni che possono colpire ma sarebbe da ingenui o da presuntuosi stupirsi di come la strada verso l’invisibile, i virus, banché correttamente postula da molti, sia stata tanto disseminata di obiezioni. Nelle rubriche vi sono inoltre brevi riassunti di lavori anatomo-patologici comparsi principalmente su Archivs negli anni ’18 - ’20 e incentrati sui ben noti quadri di broncopolmonite emorragico-suppurativa, talmente massiva da essere caratteristica e definitiva, secondo le affermazioni del patologo Cesaris Damel nel corso del congresso del ’19 a Milano. Per finire nel 1923 Mario Segale torna sul tema del bacillo di Pfeiffer, ribattezzato per l’occasione bacillo di Pfeiffer-Gosio, in sintonia forse con il nuovo spirito autarchico del tempo e in onore del batteriologo italiano Bartolomeo Gosio direttore del Laboratorio centrale della Sanità Pubblica che aveva studiato approfonditamente questo germe e ne aveva sostenuto con forza il ruolo causale. In questo intervento Segale con dovizia di argomenti scagiona definitivamente l’Haemophilus Influenzae e dichiara insoluto il problema della causa microbiologica della grande pandemia del ’18.
Gli atti della riunione italiana per lo studio della pandemia 1918. Milano, 27-28 Aprile 1919  

I professori Mangiagalli, Devoto, Belfanti e Viganò organizzano a Milano questa riunione dal carattere, sottolineano, famigliare e pratico. Già questa precisazione evidenzia implicitamente il clima sanitario che si doveva respirare all’epilogo dell’epidemia. Sin dalle prime battute si intuisce una contrapposizione di punti di vista tra Bartolomeo Gosio, che appare come il difensore dell’ortodossia batteriologica e gli altri, tra cui spicca per incisività degli argomenti il patologo Cesaris Damel. Gli argomenti di Gosio sono molti e il batteriologo si sofferma sui vari terreni di coltura da utilizzare, sulle infezioni sperimentali nel topo e sul ruolo delle tossine del bacillo di Pfeiffer, in grado di provocare infarti polmonari, epatici ed encefalici a suo dire. “Non è esatto che il prof. Pfeiffer non abbia più fiducia nel germe da lui scoperto come causa dell’influenza”, dichiara. “Anche le esperienze fatte vicino a Boston per negare l’azione patogena sull’uomo non hanno grande valore”, aggiunge. Di cosa si trattava: in cambio della libertà, a una sessantina di detenuti venne proposto da medici militari del Chelsea Hospital la nebulizzazione nel naso e nella gola detenuti venne proposto da medici militari del Chelsea Naval Hospital la nebulizzazione nel naso e nella gola di escrementi di malati di grippe ricchissimi di bacilli di Pfeiffer 2 14. Esperimenti simili, in una medicina priva delle attuali regolamentazioni etiche, avvennero anche in altre parti del mondo 2 15, alla ricerca delle cause di una malattia che sembrava a molti sperimentatori limitata alle colonne delle sue rubriche da un vaglio sistematico della letteratura internazionale e da lavori anatomo-patologici e microbiologici originali, oscillava senza pervenire a conclusioni chiare tra ipotesi di un’origine batterica (bacillo di Pfeiffer, magari virulentato, o azione combinata di diversi batteri, in primis nuovi ceppi di streptococchi) e congetture indimostrabili su di una possibile causa virale. Del resto, la natura submicroscopica e intracellularle dei virus collocava le indagini eziologiche della pandemia oltre i limiti della scienza del tempo.

Nondimeno lo studio della guerra di trincea è stato terreno di incontro tra storici e medici 17-19. Nel bilancio finale di una guerra infatti vi sono capitoli dedicati a come le malattie abbiano contribuito al suo svolgimento e come viceversa la guerra abbia condizionato il modo di fare fronte alle emergenze sanitarie. Da questo punto di vista le epidemiche sono un modello straordinario perché il modo di affrontarle dipende strettamente dal mutare delle condizioni socio-politiche, il che rende ciascuna epidemia una storia a sé. Colpisce in questo senso il contrasto dell’epoca tra la vivacità del dibattito, anche all’interno di Pathologica, e quanto poco sia emerso all’esterno. A ben vedere tuttavia, la disinformazione era la risposta coerente con le prospettive di un’epoca di diffuso fabeticismo, nella quale la retorica dominante non sapeva trasformare in vittoria o almeno in eroismi la gestione di una catastrofe come “la spagnola”. Ma oltre alla censura ideologica della stampa interventista e della politica, che temeva anche la demoralizzazione di un esercito impregnato nell’epilogo del conflitto ed era perciò sempre pronta a bollare come disfattista qualsiasi notizia negativa, vi fu l’autocensura imbarazzata della classe medica, che usciva dalla stagione d’oro delle grandi scoperte microbiologiche di fine ottocento e di inizio secolo. Negli anni successivi per concludere, l’ombra della memoria collettiva della grande guerra e di quello che ne è seguito, e la mancanza di interesse di grandi scrittori e giornalisti che sapessero imprimere alle parole influenza spagnola il suono sinistro di una pestilenza, hanno finito per confinare la memoria della pandemia del 1918 nel privato delle storie famigliari.

Conclusioni

Se le guerre si sono spesso rivelate una tragica occasione di progresso scientifico, si può dire che nel caso della spagnola quest’occasione venne mancata. Anche nelle pagine di Pathologica il dibattito, pur arricchito nelle colonne delle sue rubriche da un vaglio sistematico della letteratura internazionale e da lavori anatomo-patologici e microbiologici originali, oscillava senza pervenire a conclusioni chiare tra ipotesi di un’origine batterica (bacillo di Pfeiffer, magari virulentato, o azione combinata di diversi batteri, in primis nuovi ceppi di streptococchi) e congetture indimostrabili su di una possibile causa virale. Del resto, la natura submicroscopica e intracellularle dei virus collocava le indagini eziologiche della pandemia oltre i limiti della scienza del tempo.

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IV MEETING NAZIONALE

GRUPPO ITALIANO DI PALEOPATOLOGIA

L’AQUILA
CONVENTO DI SAN GIULIANO DEI FRATI MINORI
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Dott. Raffaele Gaeta
Prof. Gino Fornaciari

Segreteria Organizzativa
Dott. Luca Ventura
Dott. Gaetano Miranda
Dott.ssa Giulia Tudico
Sessione I
Moderatori: R. Gaeta (Pisa), M. Licata (Varese)

Wounding and death of Ferdinand Francis (1490-1525), marquis of Pescara and winner of the battle of Pavia: archaeo-
llogical and paleopathological evidences

G. FORNACIARI
Department of Civilisations and Forms of Knowledge, University of Pisa, Italy

Ferdinando Francesco d’Avalos, 5th marquis of Pescara (1489-1525), was an Italian condottiero of Spanish origin. As general
of the Spanish army, he participated in the Italian Wars. When Francis I invaded Italy in 1524, d’Avalos was appointed “Lie-
utenant of the Emperor” to repel the invasion. His influence over the veteran Spanish troops and the German mercenaries
kept them loyal during the long siege of Pavia. On February 24, 1525 he defeated and took prisoner Francis I by a brilliant
attack. D’Avalos plan was remarkable for its audacity and for the skill he showed in destroying the superior French heavy
cavalry, which was assailed in flank with a mixed force of harquebusiers and light horses. His health, however, had begun
to give way under the strain of wounds and exposure, during late November, until he died in Milan on December 3, 1525
and was buried in the Basilica of Saint Domenico Maggiore in Naples. It was also possible to establish the tomb
of Ferdinando Francesco d’Avalos in the Basilica of Saint Do-
menico Maggiore in Naples. His body was buried into the common grave of the friars and
found intact when exhumed many years later.

The Blessed Vincenzo dell’Aquila (1430-1504). Canonical Recognition
and paleopathological study

L. YENTURA1, M. IOANNUCCI2, G. TUDICO1, A. RAF-
FAELE3, C. MASCIOCCHI1

1 Division of Pathology, San Salvatore Hospital, L’Aquila, Italy; 2 Diagnostic and Interventional Radiology, Department of Tran-
slational Research and New Technologies in Medicine and Surgery, University of Pisa, Italy; 3 Division of Radiology, San Salvatore Hospital, L’Aquila, Italy.

Vincenzo was born to a humble family in the oldest part of the city, and worked as a shoemaker. In 1448, he entered the Franciscan Regular Observance. He received the gift of prophecy and was kept in great honor by the neapolitan sovereigns. In 1482, Alfonso Duke of Calabria was driving the aragonese army against the Papal States, and asked to meet him. Vincenzo predicted his defeat, actually occurred in the battle of Campomorto (21 August 1482). He also predicted the Prince of Capua (the future King Ferdinando II) a rapid gain of the throne, shortly followed by his death. The Queen of Naples herself used to keep the friar into high consideration. In his last years Vincenzo was affected by “gout” and after death he was greatly honoured by people and authorities. His body was buried into the common grave of the friars and found intact when exhumed many years later.

The fourth Canonical Recognition of the Blessed took place in 2018, following the restoration of his portrait made by Sat-
urnino Gatti in 1509.

External examination and on-site digital radiography allowed to recognize a well-preserved, natural mummy belonging to
an old male subject in good nutritional status. Marked dental wear and parodontal disease, in the absence of caries, were
identified, along with artificial reconstruction of face and feet. Calcium deposits in the left costophrenic angle were displayed
by radiography, and referred to infection or pleural effusion.

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San Davino Armeno (†1050).
Preliminary results of the paleopathological study

A. FORNACIARI1, V. MONGELLI1, L. MELAI2, D. CARAMELLA1, G. FORNACIARI1, V. GIUFFRA1
1Division of Paleopathology, Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa, Italy; 2Service of Radiology, Barbiantini Clinic, Lucca, Italy; 3Division of Diagnostic and Interventional Radiology, Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa, Italy

The preliminary study of the mummy of San Davino Armeno (†1050) was performed by the Division of Paleopathology of the University of Pisa in March 2018. The mummy of San Davino, preserved in Lucca for about a millennium in the Basilica of San Michele in Foro, is one of the oldest natural mummies of saints known in Italy. The study of the mummy included: undressing, macroscopic examination, endoscopy of the oral cavity and CT total body. Macroscopic examination allowed to establish a partially skeletonized natural mummy of a man of about 25 years, 1.70 m tall. The cranial vault, spine, and thorax resulted skeletonized. Skin is preserved on the upper and lower limbs and partially in the pubic region. CT shows the partial preservation of the brain in the posterior skull, and the remains of rectum, prostate and external genitalia. The study of teeth reveals no dental wear, a penetrating caries of the second upper right molar and diffuse periodontal disease, with partial alveolar resorption of the anterior teeth and accumulation of dental calculus on the lower incisors on the lingual side. The muscular insertions of the upper and lower limbs demonstrate a strong muscular activity. On the tibia and fibulas are evident traces of periostitis. Paleopathological study revealed two traumatic lesions of the skull with long-term survival: a superficial sharp force lesion on the left frontal bone (5 cm length, 0.5 cm wide), perhaps caused by a toothed blade, and an irregular elliptical (2 x 1 cm) wound produced by a blunt weapon on the right coronal suture, with evident signs of surgical treatment by a cautery with pentagonal head (2 cm on each side), that created a thin margin (0.5 mm) around the lesion. The surgical treatment is a rare case of 11th sample of cautery applied to a cranial wound, testified by the written source and medical literature especially in Islamic medicine (Albucasis), but very uncommon to observe in the archaeological remains.

References

Histopathological re-evaluation of two “pleural sarcomas” from the Pathology Collection of Turin

L. FERRARI1,2, J. METOVIC2, D. RULLO2, L. VENTURA1
1SC Anatomia Patologica, Cardinal Massaja Hospital, Asti; 2University of Turin, Città della Salute Hospital, Turin; 3U. O. C. di Anatomia ed Istologia Patologica, Ospedale San Salvatore, L’Aquila, Italy

The Pathology Collection of Turin houses around 300 wet specimens dating back to the end of XIX century and the beginning of the XX century. Most of them are in their original jars whose label describes year, necropsy number, and diagnosis. Two cases labelled as “pleural sarcomas” (sarcomi della pleura), an entity considered very rare since that time, underwent modern re-evaluation in order to verify the original diagnosis.

Samples from the specimens were submitted to routine histology, histochemistry (Mason’s trichrome and reticulin) and immunohistochemical stains. The first case dates back to 1896 with an autopsy report of a stomach cancer metastatic to the mediastinum. Grossly, the lung showed disseminated papillary lesions on the pleural surface and within the organ. Histology and histochemistry suggested a non keratinizing squamous carcinoma. Negative cytokeratin 7 and CD68 with internal positive controls were the only reliable immunostains. All these data support the final diagnosis of metastatic squamous cancer from distal esophagus. The second specimen dates back to 1898 and was sampled at the autopsy of a 94-year-old woman with a “soft tumor of the cervix” metastatic to liver and with pleural nodules. Gross examination showed roundish nodules of different diameter on the pleura and within the lung. Microscopy suggested a leymiosarcoma. Cytokeratin AE1/AE3 was negative with positive control in some normal mesothelial cells. The final diagnosis was uterine leiomyosarcoma metastatic to the lung.

Despite the lack of reliable immunohistochemical results, morphological and histochemical details enabled us to address the final diagnosis in both instances. In contrast to other researchers, the long-term preservation in a fluid different from formalin may have hindered immunohistochemistry. According to the doctoral dissertation of Carlo Ascoli in 1894, at the end of the XIX century formalin had only an antiseptic use in Italy.

References

Perimortem reconstruction of an inhabitant of Mutina (Italy): a multianalytical investigation of weapon-related injuries in a Late Antiquity necropolis

M. TRAVERSARI1, A. VAZZANA1, L. M. SCALISE1, C. FIGUS1, S.A. APERCELLA1, L. BUTI1, G. OXILIA1,2, R. SORRENTINO1, S. PELLEGRINI1, C. MATTEUCCI1, L. CALCAGNILE2, R. SAVIGNI1, R.N.M. FEENEY3, G. GRUPPIONI1, S. BENAZZI1,6
1Department of Cultural Heritage, University of Bologna, Ravenna, Italy; 2Department of Oral and Maxillo Facial Sciences, Sapienza University, Rome, Italy; 3Municipal Museum of Archaeology and Ethnology of Modena, Italy; 4CEDAD - CEnter for Dating and Diagnostics Department of Mathematics and Physics “Ennio De Giorgi”,
Human skeletal remains from archaeological contexts occasionally preserve evidence of traumatic injuries from weapons, revealing the degree of interpersonal violence, type of weapons used and the sequence of events; details which may be specific to an historical context. Traumatic lesions are typically analyzed using macroscopic and microscopic methods, which are seldom integrated in the same study. We employed a multi-analytical approach to determine if new or refined information could be gained compared to traditional analyses.

We analyzed interpersonal perimortem sharp-force trauma in the skeletal remains of an adult male recovered during excavations of a 4th-6th century AD necropolis (radioncarbon dated between 380–580 CE) in Mutina (an ancient Roman town in modern Modena, Italy). This period was characterized by frequent devastations from natural disasters and barbaric invasions. Evidence of sharp force trauma was observed in four of 13 individuals recovered, but only one individual (B11) presents a complex violent succession of injuries.

Traumatic lesions were analysed using an integrated multi-analytical approach combining traditional macroscopic examination, light microscopy and 3D digital modelling of injuries. Morphological analyses revealed an absence of bone response and the presence of polished surfaces, which suggest the injuries led to the immediate death of the victim and which typically characterize a metal weapon impact on cortical tissue, respectively. The virtually computed angles between the lesion and Camper’s planes, and the amount of bone removed on the vault, further suggest the movement of the weapon and the energy. From this, we were able to determine the number, size and position of insults, and the direction (angles) of weapon penetration in the bone. The application of a multianalytic methodology provides advanced information on the dynamics of violent deaths useful for interpreting ancient contexts.

References


Head wounds by firearm and sharp weapon at the S. Martino Battle (1859, Italy)

S. MINOZZI, V. GHIROLDI, G. FORNACIARI
Division of Paleopathology, Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa, Italy

On 24 June 1859, the Battles of San Martino and Solferino involved the French Army allied to the Sardinian-Piedmontese Army opposed to the Austrian troops. The victory of the French and the retreat of the Austrians ended the Second Italian War of Independence. Thousands of deaths of both Armies were hurriedly buried in the next days, but in 1869 the skeletal remains were exhumed and placed in a monumental ossuary. The aim of this work was the study of the head injuries caused by firearms and bladed weapons in relation to the armaments and the military tactics used in that period.

Examination of 817 skulls recovered in the San Martino Ossuary revealed traces of war wounds in 90 skulls that were selected and submitted to anthropological and paleopathological studies. The shape, dimensions and features of the wounds were accurately examined to characterize the lesions. The results evidenced different types of injuries, principally caused by firearms with projectiles of different calibers. The investigation of the caliber and the trajectory of the projectiles, as well as the kind of artillery have been based on the diameter and localization of the holes in the skulls. The size of the entry wounds corresponds to the caliber of the bullets used by both armies. Therefore, the army membership of the soldiers could not be distinguished.

The 18% of the skulls were affected by steel weapon wounds mostly localized on the right side of the skull. All the wound characteristics corresponds to those of the weapons used by both armies equipped with sabres, lances, spits or dagger bayonets. The presence of several healed lesions, in some cases surgically treated, testifies a rather intense war period, and the capacity of the soldiers to survive even to severe injuries. The prevalence of gunshot wounds is consistent with the military tactics and with the weapons used in that period: hand-to-hand combat was restricted and bayonet assault was unsuccessful compared to the use of efficient ranged weapons.

From paleopathological issues to the re-excavation of the cemetery area of Saint Agostine in Caravate (Varese)

M. LICATA1, D. CAPUZZO2, A. TOSI1, R.FUSCO1, O. LARENTIS3
1Centre of Research of Osteoarchaeology and Paleopathology, Department of Biotechnology and Life Sciences, University of Insubria; 2Archeosfera S.r.l.s, Sesto San Giovanni (MI), Italy

In 2002, an excavation carried out near the northern perimeter wall of the church of Saint Agostine in Caravate (Varese), accidentally led to the discovery of an ancient cemetery area. The subsequent archaeological investigations brought to light twenty burials dating from the 8th to the 9th century AD. The tombs, oriented according to the canonical order, that is east-west with the west-facing head, contained skeletons in an excellent state of preservation.

From the first anthropological investigations, we registered a significant number of ante mortem injuries to the head level, which could suggest violent episodes in those communities. In addition to this, other degenerative diseases common to adult populations of the time were observed, as well as vitamin deficiencies in young subjects. The anthropological estimates then allowed us to determine an average stature of 160 cm for men and 145-150 cm for women and a life expectancy significantly lower than current standards. The tombs, oriented according to the canonical order, that is east-west with the west-facing head, contained skeletons in an excellent state of preservation.

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1Centre of Research of Osteoarchaeology and Paleopathology, Department of Biotechnology and Life Sciences, University of Insubria; 2Archeosfera S.r.l.s, Sesto San Giovanni (MI), Italy

In 2002, an excavation carried out near the northern perimeter wall of the church of Saint Agostine in Caravate (Varese), accidentally led to the discovery of an ancient cemetery area. The subsequent archaeological investigations brought to light twenty burials dating from the 8th to the 9th century AD. The tombs, oriented according to the canonical order, that is east-west with the west-facing head, contained skeletons in an excellent state of preservation.

From the first anthropological investigations, we registered a significant number of ante mortem injuries to the head level, which could suggest violent episodes in those communities. In addition to this, other degenerative diseases common to adult populations of the time were observed, as well as vitamin deficiencies in young subjects. The anthropological estimates then allowed us to determine an average stature of 160 cm for men and 145-150 cm for women and a life expectancy that never exceeded the mature age.

To clarify the degree of violence within these populations we decided, in May 2018, to resume the archaeological excavation of the cemetery area that in the past had not been investigated. The excavation has brought to light several burials of children with opposed orientation from the others.

References

Pathological evidences from medieval samples. The subjects of Saint Pietro and Paolo church in Brentonico

E. TONINA1, C. PANGRAZZI2, O. LARENTIS2

1B. Bagolini Laboratory, Department of Humanities, University of Trento; 2Centre of Research in Osteoarchaeology and Paleopathology, Department of Biotechnology and Life Sciences, University of Insubria

Archaeological investigations conducted in 2003 by The PAT Archaeological Heritage in the church of Saint Pietro and Paolo in Brentonico (Trentino Alto Adige, northeastern Italy), brought to light a secondary grave, dated to the 13th century, under the Romanesque bell tower. The anthropological investigation estimated a minimum number of 22 individuals. Most of the subjects show severe caretential conditions, highlighted by the constant presence of cribra cranii and periostal reactions diffused on tibiae and femora.

Concerning the arthropathies, the joint of the pelvis appears to be the most compromised, and strong arthritic degeneration is often accompanied by osteochondritic phenomena. Knees and shoulder joints have also degenerated. The arthropathies in the lower limbs are accompanied by the frequent presence of occupational markers and in the upper limbs by a strong development of musculoskeletal markers. In addition to the most commonly visible pathological conditions in osteoarchaeological findings, our sample presents some meaningful pathological conditions, traumas and characters less present in the archaeological record. The skull of an adult male presents an extended antemortem cut on the left parietal bone and a Concha Bullosa, and another skull shows possible peri mortem traumas on the frontal bone. Furthermore, a left shoulder blade presents a fracture with bone remodelling at the acromion level. Other pathological conditions recorded in the sample are: a Brodie abscess on a tibiae that belonged to an adult and an osteochondroma recognizable by a tibiofibular synostosis. We also observed a probable third condyle detected on the occipital condyle’s anterior arch of a female skull. Among the epigenetic traits the crista supramastoidea is observed on several skulls in our samples.

References

Leprosy spread in Italy

M. RUBINI1,2, N. LIBIANCHI1, A. GOZZI1, E. DELLÜ1, P. ZAIO1

1Servizio di Antropologia SABAP-RM-MET e SABAP-LAZ, Italy; 2University of Foggia, Italy

The arrival of leprosy in Italy is a very controversial topic, as is its diffusion in Europe. According to the most accredited theories on a molecular basis, leprosy spread in Eurasia from Eastern Africa about 80,000 years ago (Monot et al 2005) following the migratory routes of H. sapiens “out of Africa”. To date, in ancient time dissemination strategies seem to have followed terrestrial rather than maritime routes (at least until to the late Middle Ages), favored in this by the long incubation period of leprosy and its low pathogenicity. The sugges-

Anthropological and paleopathological analysis of the trepanned skull of Aliano (MT)

G. MIRANDA1, D. MANCINELLI2, A. PREITE1

1Physical Anthropology, L’Aquila, Italy; 2MESVA Department, University of L’Aquila, Italy; 3Pre-Protohistoric Archeology, Chiaromonte (FZ), Italy

The tomb n. 856 was located at the northern end of the necropolis and the deposition plane was placed at over 2 m deep. Consists of a complete individual in an excellent state of conservation. All anatomic districts are present and well represented only the left condyle of the mandible is absent. Anthropological analysis shows that it is a male individual with a strong sexualization at the level of glabella and mastoid and pelvis with a very closed and pronounced ischial incision. The age at death is estimated to be between 43 and 50 years. From classical anthropology, the individual has a height between 138 cm and 163 cm, of robust build and well pronounced ossifications. From the muscular insertions and lengths of humeri and ulnae it is believed that the individual was right-handed. The overall state of health of the individual is substantially good, there are slight osteophyosis at the level of the atlas, the epistropheus and the last lumbar vertebrae, stress injuries on the right clavicle and a strong expression of the pectoralis major muscle. There are very light astragalus and calcaneum eburneations.
On the individual’s skull there is a hole in the center of the left parietal bone of an antero-posterior oblong shape. The dimensions of the hole are for the greater transverse length of 2.54 cm and for the shorter (supero-inferior) length 2.04 cm. An oval that shows clear signs of travelling intra vitam. The edges are net eburne and with clear signs of both external and internal vascularization, which denotes a survival of at least 6 months. Longer times are not excluded. The motivation of the act is not very clear, from a slight longitudinal fracture of about 7.2 cm that seems healed and vascularized, which goes from the left eyebrow to the medial border of the trephination can lead to suspect a random or induced traumatic event. Two are the hypothesis: accidental fall with a fracture of the skull, and a blow with a non-pointed instrument vibrated by a right-handed upward. In a face to face contrast the agressor is of equal height of the traumatized.

The technique used for the surgery is that of scraping ie a slightly sharpened non-pointed and abrasive tool that rubbed with force and continuity on the skull opens a hole, of variable shape depending on the instrument used, from round to oval. The thesis of the traumatic event, however, is not fully supported by post-cranial data, where there are no fractures, lesions or anomalies of any kind to any bone district that can show a fall, an aggression or other. The cause of death for this reason has nothing to do with the traumatic event itself, it is not excluded a secondary cause not found on bones, such as acute infections or other related to them.

**Sessione III**

Moderatori: L. Ferrari (Asti), M. Rubini (Roma, Foggia)

**A possible case of mycosis in a post-classical burial from La Selvicciola (Italy)**

I. MICARELLI, R. PAINE, M. A. TAFURI, G. MANZI

1Department of Classics, Sapienza University of Rome, Italy; 2Department of Environmental Biology, Sapienza University of Rome, Italy

Diagnosing disease from dry bone lesions is often challenging. Nevertheless, when burials exhibit systemic pattern of lesions paleopathologists can link lesion type, frequency and location with a specific illness. While examining a skeleton of an adult male buried from the post-classical necropolis of La Selvicciola (Viterbo, Latium, Italy; 4th-6th centuries AD), a series of skeletal lesions was revealed. This male (t 84/3) was buried without grave goods outside the church walls and W-E oriented. The burial is contemporary with the post-classical phase of the settlement, and it was placed there after the church had been built, earlier than the 7th century AD. The lesions occurred at multiple sites in a random pattern and they are both proliferative and lytic in nature and ranging in size from small to extensive pits. A holistic assessment involving lesion type, frequency and location was used to determine potential diagnosis. Bone has a limited response to agents that stimulate the formation of bony defects. In some cases, osteoblastic response to a pathogen will produce new bone growth. In other cases, osteoclastic activity will lead to bone resorption. Occasionally, these agents will produce both bony growth defects and lytic lesions on the same surface. A comparative assessment of the lesions observed in 84/3 indicates that it does not match any of the lesion patterns specific to tuberculosis, osteomyelitis, metastatic carcinoma, leukemia, and multiple myeloma. So, we compared the lesion pattern of 84/3 to fungal causal agents, as they also cause lytic bone responses. Therefore, determining which fungal agent caused this condition is problematic. Still, key factors useful for circumscribing a potential fungal candidate includes geographical region/ecological information as well as modern clinical case assessments. As a final assessment, it was concluded that a mycosis, specifically Cryptococcus, may have been the cause of these lesions.

**References**


**A medieval case of a possible metastatic neuroblastoma from Vico nel Lazio (FR), XIII-XV century**

P. ZAIO, N. LIBIANCHI, A. GOZZI, E. DELLÜ, M. RUBINI

1Servizio di Antropologia SABAP-RM-MET e SABAP-LAZ, Italy; 2University of Foggia, Italy

A site inspection at the Church of Santa Maria in Vico nel Lazio led to the discovery of a crypt whit numerous burials related to the medieval settlement, which is dated between the 13th and the 15th century. The human remains belongs to both male and female individuals and fully represent the demographic sample for the presence of either adults and infants. We present a skull of a woman which was about 30-35 years old. Despite of the young age she was affected by the complete loss of teeth, with a significant alveolar resorption. Most remarkably the calvarium shows numerous perforations with irregular or even jagged edges, along with the evident cribran on the right parietal bone, and the intense pyogenic activity which involves part of the frontal bone and the nasal bones. The differential diagnosis excludes either diseases that produces lesions similar to the observed ones (such the syphilis and the leukemia) or surgical complications (trepanation). Thus for the strictly characterizing morphology of the “open space” fenestrations, which most probably refer to the metastatic activity of a neuroblastoma.

**References**


**Multiple osteomas in a skeleton from the medieval necropolis of Pava (Tuscany, 10-12th Centuries)**

V. GIUFFRA, G. NACCARATO, M. CASTAGNA, R. LENCIONI, S. CHERICONI, V. MONGELLI, G. RICCOMI, S. CAMPANA

1Division of Paleopathology, Department of Translational Research
A pathological diagnosis of kyphosis in the case of the concealed corpse - the skeleton of Azzio’s Crypt

O. LARENTIS1, I. GORINI1, C. ROSSETTI1, E. TONINA2, M. LICATA1

1Centre of Research of Orearchaeology and Paleopathology, Department of Biotechnology and Life Sciences, University of Insubria; 2B. Bagolini Laboratory, Department of Humanities, University of Trento, Trento, Italy

An archaeological recovery, conducted in 2013, by Lombardy Archaeological Heritage in the crypt of the Monastic Church of Azzio (Varese) brought to light a common ossuary, belonging to the Franciscan order, which then revealed an anomalous burial. A complete skeleton with osteological districts in anatomical connection was lying in supine position above an accumulation of bones.

In a successive archaeological investigation of the crypt in 2015, the skeleton was recovered and transported to our center of research. In addition to the archaeological data, which suggested a case of concealment of the corpse, C14 analysis, dating the skeleton after the funerary use of the crypt by the friars could confirm this hypothesis.

The anthropological investigation revealed that the skeleton belonged to an adult male with an approximate estimated stature of 185 cm.

From the macroscopic analysis, we observed that the skeleton presented an important kyphosis of the high thoracic tract caused by the fusion of the vertebral bodies with the consequent collapse of different elements of the spine. Radiological investigations (CT scan) confirmed the kyphosis diagnosis. This pathological condition probably caused several anatomical alterations especially located in the scapular girdle and at the thorax level.

An in-depth study of pathological conditions in the future will probably carry out new elements useful to the identification analysis of this past case of a concealed corpse.

References


The skeletal remains of Tommaso da Cascina (1379-1460), founder of the San Giuliano Convent

G. TUDICO1, G. MIRANDA2, M. IOANNUCCI3, A. RAFFAELE1, C. MASCIOCCHI1, L. VENTURA1

1Division of Pathology, San Salvatore Hospital, L’Aquila, Italy; 2Physical Anthropologist, L’Aquila; 3Sacred Linsanotheca of the Metropolitan Archibishopy of L’Aquila, Italy; 4Division of Radiology, San Salvatore Hospital, L’Aquila, Italy

The convent of San Giuliano (L’Aquila, central Italy) was built in 1415 by friars coming from Umbria region, supported by the local monk Tommaso da Cascina and the wealthy citizen Nunzio da Fonte, who financed the new foundation. Tommaso was born in Cascina, a no more existing village near L’Aquila. When he was 11 he joined the Clarens (Franciscan hermits), then entered the Franciscan Regular Observance as a lay-brother. In 1418, he was sent to Corsica, but came back three years later to stay in San Giuliano until his death.

His bones are enshrined in a repository kept in the second left chapel of the conventual church. The reliquary appears as a carved, painted wooden casket with a central glass window displaying bones. On the back of the châssé someone wrote “This chest was made on march 30th 1624”.

After opening the box, a mixture of dark, clean, post-cranial bones was evident. Skeletal completeness was 60% and the bones belonged to a male subject, about 160 cm tall, more than 60 years of age at death. A preliminary inspection showed signs of heavy walking activity, Schmorl’s nodes, and mild osteoarthritis of the spine.

A sacral spina bifida occulta, grade II, along with bilateral spondylolysis of the fifth lumbar vertebra and possible spondylolisthesis were evident. Moreover, a sacroiliitis with lytic lesions of both sacrum and left os coxae was noted and confirmed after radiologic investigation. Additional investigations are planned to disclose the real nature of this condition (neoplasm? infection?).

This contribution confirms the importance of reliquaries in preserving human remains of religious historical figures. Modern investigation studies of such relics may be of great help in improving our knowledge about lifestyle and health condition of this particular category of people.

References


Locus parvulorum, children’s pathology of the Saint Mary Nativity church of Segno (Trentino)

O. LAVENTIS¹, E. TONINA²

¹Centre of Research in Osteoarchaeology and Paleopathology, Department of Biotechnology and Life Sciences, University of Insubria, Italy; ²B. Bogolini Laboratory, Department of Humanities, University of Trento, Italy

The funerary area of the Saint Mary Nativity church was excavated in 2004 by the Trentino Archaeological Heritage Department. From the 15th to the end of the 18th century, the space in front of the facade was used only for the deposition of children. 38 primary burials, belonging to children aged between prenatal and puberty, were discovered and underwent anthropological analysis. For better comprehension of the pathological picture of the group, archaeological and anthropological evidences were associated with information acquired from the parish registers of baptisms, marriages, and burials and from the historical archival documents. The samples allowed us to observe the pathological conditions of children from a rural medieval, showing some of the most interesting diseases detectable in the early years of life, such as vitamin C and D deficiency and their co-occurrence, traumatic injuries, endo cranial lesions, carential states or imbalance of the haemopoietic system and dental pathologies. The study of children’s health within a known closed community could also be useful to comprehend the state of health in the adults. This contribution presents a cross section of the state of health and quality of life of the children from Segno and respective communities.

References