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Zakład Anatomii Prawidłowej
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Jurand Domański

Holistyczne badania nad wybranymi muzealnymi kolekcjami medycznymi

ROZPRAWA DOKTORSKA
Cykl publikacji powiązanych tematycznie

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„Spojrzenie”
Poeta i Anatom
Inaczej się przygląda kwiatom.

- Jan Sztaudynger

Pragnę podziękować mojemu Promotorowi pomocniczemu,
serdecznemu Koledze
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1. CYKL PRAC STANOWIĄCYCH ROZPRAWĘ DOKTORSKĄ

1. **Jurand Domański**, Zygmunt Domagała, John E. Simmons, Marta Wanat: *Terra Incognita in anatomical museology - a literature review from the perspective of evidence-based care.* Ann Anat. 2023 Jan;245:152013. doi: 10.1016/j.aanat.2022.152013.
   IF: 2,976; Punktacja Ministerialna: 100 punktów

2. **Jurand Domański**, Adriana Janczura, Marta Wanat, Katarzyna Wiglusz , Magdalena Grajzer, John E Simmons, Zygmunt Domagała, Jacek C Szepietowski: *Preservation fluids of heritage anatomical specimens -a challenge for modern science. Studies of the origin, composition and microbiological contamination of old museum collections.* J Anat. 2023 Apr 6. doi: 10.1111/joa.13854.
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3. **Jurand Domański**, Rafał Białynicki-Birula, Urszula Nawrot, Elżbieta Piątkowska, Zygmunt Domagała, Jacek C Szepietowski: *Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland.* Adv Dermatol Allergol 2023, doi.org/10.5114/ada.2023.127197
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2. OMÓWIENIE ROZPRAWY DOKTORSKIEJ

2.1 WSTĘP

Historyczne muzea uczelniane są depozytariuszami unikalnego dziedzictwa. W przypadku muzeów medycznych wyjątkowość tych instytucji jest szczególna, gdyż zabezpieczają jak i eksponują obiekty, które ze względu na swoją właściwość są niemożliwe do odtworzenia. Przykładowo Muzeum Anatomiczne Uniwersytetu Medycznego we Wrocławiu zabezpiecza kolekcje zabytkowych płodów teratologicznych utworzoną na początku XIX wieku. Znajdują się tam okazy, które najprawdopodobniej prezentują wady rozwojowe powstałe w wyniku chorób zakaźnych okresu ciąży. Wiele z tych schorzeń jest współcześnie eradykowanych lub ich przebieg jest zmieniony w wyniku rozwoju nowoczesnych technik medycznych jak i skutecznych działań prewencyjnych. Efektem tego postępu medycyny jest niezwykła unikalność takiej kolekcji.. Zbiory te, niezwykle 'wrażliwe' na odbiór społeczny, nie mogłyby być eksponowane w innych instytucjach, stąd muzea medyczne to fenomen, legitymizowany etosem medycznym.

Z punktu widzenia pragmatyzmu naukowego, muzea medyczne przechowują atrakcyjny materiał badawczy lub też same w sobie stanowią obiekt badań. Ochrona zbiorów muzealnych, winna być podyktowana choćby z tego powodu, jeśli nie z oczywistych, moralnych powódek. Unikalność tych zbiorów wyklucza jednak stosowanie niesprawdzonych czy niezweryfikowanych naukowo metod ochrony. Bez ich opracowania nie ma możliwości wdrożenia sposobów ochrony dziedzictwa medycznego. Z kolei brak badań uniemożliwia opracowanie metod. Współczesna nauka powinna zatem podjąć (i) badania służące zarówno poznaniu właściwości samych obiektów muzealnych (tzn. badania podmiotowe zbiorów), (ii) badania służące opracowaniu metod ochrony zbiorów i wreszcie (iii) weryfikacji metod badań naukowych na zbiorach muzealnych/muzeach.

Obiekt muzealny jako materiał badawczy wyróżniają jego wielopłaszczyznowe właściwości w tym historyczny kontekst, rozumiany jako przyczyny jego powstania i ekspozycji, techniki wykonania, zmiany dokonywane w czasie (zamierzone i samoistne - niezamierzone) jak i fluktuacje w identyfikacji samego obiektu na przestrzeni czasu. Z tego powodu badania naukowe zbiorów muzealnych są wymagające. Konieczna jest analiza naukowa z różnych perspektyw - z różnych dziedzin nauki i ich wzajemna konfrontacja. Jedynie holistyczna determinacja projektowanych badań umożliwia obiektywne poznanie tego unikalnego materiału badawczego i uzyskanie pełnych i jednocześnie obiektywnych

wyników, będących skutkiem wzajemnego przenikania się czynników kształtujących obiekt badawczy. Ta interdyscyplinarność, zarówno w kontekście badań naukowych jak i podejmowanych działań ochrony zbiorów wydaje się korespondować z medycyną pojmowaną jako sztuka: dyscypliną nauki jak i praktyki leczniczej – *'art and practice'*.

2.2 CELE

Rozprawa skupia się na przedstawieniu koncepcji badania zbiorów historycznych zbiorów muzealnych opartej na interdyscyplinarności.

2.2.1 Szczegółowe cele badań

1. przeprowadzenie badań podmiotowych wybranych zbiorów muzealnych w koncepcji interdyscyplinarnej
2. weryfikacji metod naukowych mogących posłużyć lepszemu poznaniu właściwości badanych obiektów muzealnych
3. opracowania innowacyjnych metod ochrony cennego dziedzictwa na podstawie efektów przeprowadzonych badań

2.2.1.1 Szczegółowe cele badań artykułu pierwszego

Celem badania było odnalezienie opracowań, których wyniki mogą odegrać rolę:
- w uzyskaniu głębszego zrozumienia technik i procedur konserwacji zbiorów anatomicznych;
- w planowaniu interwencji konserwatorskich, których efektem były zmiany w strukturze obiektów muzealnych;
- w optymalizacji planowanych badań na materiale muzealnym celem zapewnienia jego jak najlepszego zachowania
- w planowanych modernizacjach zabytkowej przestrzeni muzealnej przy jednoczesnym zachowaniu jej historycznej integralności.

Dodatkowymi celami pracy były:
- ocena stanu dostępnych badań i ich rodzajów, które mogłyby mieć praktyczne zastosowanie w modernizacji muzeów anatomii człowieka,

- próba kategoryzacji istniejących publikacji naukowych dotyczących muzeów anatomicznych i opieki nad zbiorami muzealnymi
- lepsze zrozumienie miejsce muzeów anatomii człowieka, historycznie związane z uniwersytetami, w ogólnej klasyfikacji i taksonomii muzeów

2.2.1.2 Szczegółowe cele badań artykułu drugiego

- celem badań było określenie składu chemicznego płynów konserwujących zabytkowe zbiory anatomiczne oraz przeprowadzenie oceny mikrobiologicznej płynów konserwujących i próbek.
- ocena, czy proste metody wykrywania składu chemicznego płynów konserwujących są przydatne w pracy konserwatorskiej/analizach badawczych i czy wyniki uzyskane za ich pomocą można porównać z wynikami otrzymanymi poprzez zastosowanie najnowszych, zaawansowanych technik analitycznych.

2.2.1.3 Szczegółowe cele badań artykułu trzeciego

- przeprowadzenie oceny mikrobiologicznej zabytkowych modeli dermatologicznych i otaczającego ich środowiska sali muzealnej.
- zbadanie, czy kolekcja zabytkowych mulaży dermatologicznych jest skolonizowana przez określone mikroorganizmy i czy stanowią one zagrożenie dla zdrowia otoczenia, w tym zwiedzających muzeum.

2.3 MATERIAŁ I METODY

2.3.1 Materiał i metody artykułu pierwszego

W pracy zastosowano metodę przeglądu systematycznego literatury dla dwóch haseł: ‘ anatomy museum’ i ‘museum technoques’ opartą o wyszukiwanie w bazach PubMed, Ebsco, Google Schoolar, z zastosowaniem innowacyjnych kryteriów AQUA (Anatomical Quality Assurance (AQUA) Checklist and the Anatomical Quality Assessment (AQUA) Tool). W celu zwiększenia czułości metody użyto dodatkowe kryteria eliminujące prace nieporównywalne, nieistotne, lub nie powiązane z badaniami/ochrona zbiorów muzealnych lub muzeami anatomicznymi.

Uzyskane w ten sposób rekordy poddano analizie porównawczej, na podstawie której określono strukturę klasyfikacji prac w poszczególnych grupach tematycznych oraz zdefiniowano kwalifikatory dla każdej z tych grup.

2.3.2 Materiał i metody artykułu drugiego

Badaniom poddano zabytkowe zbiory anatomiczne przechowywane w Zakładzie Anatomii Prawidłowej Uniwersytetu Medycznego we Wrocławiu. Ocenie poddano zbiory przechowywane w Muzeum Anatomicznym (grupa kontrolna - 6 okazów) i w podziemiach Zakładu (grupa badawcza - 10 okazów). Badania podzielono na trzy wątki: badania historyczne, chemiczne, mikrobiologiczne.

2.3.2.1 Badania historyczne

Dokonano przeglądu literatury naukowej w celu doboru odpowiednich metod laboratoryjnych do oceny składu płynów konserwujących wykorzystując wyniki dwóch przeglądów literatury: (i) przeglądu systematycznego z artykułu pierwszego prezentowanego w przewodzie (ii) przeglądu niesystematycznego opartego na pracach opublikowanych w czasopiśmie "Museologica" i ocenie wykazów literatury z monografii "Fluid preservation: a comprehensive reference". Dodatkowo przeanalizowano współczesne monografie dotyczące historii Uniwersytetu Medycznego we Wrocławiu, archiwalne prace naukowe Kierowników Instytutu Anatomii z XIX-XX wieku.

2.3.2.2 Badania chemiczne

W badaniach chemicznych analizowano płyny konserwujące i porównywano użyteczność zastosowanych metod. Zastosowano (i) metody analityczne farmakopopealne oparte na prostych reakcjach chemicznych oraz (ii) metody oceny zaawansowane: chromatografia gazowa - trójkątna kwadrupolowa spektrometria mas tandem [GS-MS/MS], spektroskopia w podczerwieni z transformacją Fouriera [FTIR], optyczna spektroskopia emisyjna z plazmą indukcyjnie sprzężoną [ICP-OES], ocena pH.

2.3.2.3 Badania mikrobiologiczne

Do oceny pobierano próbki płynów konsekrujących oraz wymazy z powłok preparatów anatomicznych, które posiewano na pożywkę płynną - bulion z glukozą i podłoże stale –

agar Sabouranda z chloramfenikolem. W kolejnej serii posiewów zastosowano dodatkowo inne stałe – agar z krwia i podłoże Mc Conkeya. Wyrosłe kolonie bakteryjne izolowano i analizowano w MALDI-TOF (matrycowa desorpcja laserowa/jonizacja spektrometria mas typu time-of-flight),. Z części okazów pobrano zeskrobiny, które barwiono metodą Giemzy i analizowano pod mikroskopem. W przypadku wzrostu grzybów, kolonie analizowano pod mikroskopem (preparat bezpośredni – grzyby pleśniowe), natomiast grzyby drożdżopodobne poddano analizie w MALDI-TOF.

2.3.3 Materiał i metody artykułu trzeciego

Ocenie mikrobiologicznej poddano zabytkowe mulaże dermatologiczne i środowisko Muzeum Mulaży Uniwersytetu Medycznego we Wrocławiu. Za pomocą jałowej wymazówki pobierano wymazy z powierzchni woskowej mulaży – z 5 gablot muzealnych, po 5/7 okazów z każdej. Wymazy pobierano również z gablot muzealnych zarówno z wewnętrznej jak i zewnętrznej strony. Oceny środowiska muzeum dokonano poprzez analizę kontaminacji powietrza (filtracja), wymazy z podłogi i ścian oraz wyciski mikrobiologiczne wykonane z grzbietów gablot muzealnych.

Materiał z wymazów i z błon filtracyjnych wytrząsano i posiewano na podłoża dla bakterii (agar z krwią) oraz dla grzybów (agar Sabouranda z chloramfenikolem). W przypadku wzrostu kolonie izolowano i analizowano – bakterie w MALDI-TOF, grzyby – analizowano ogólną morfologie i preparaty bezpośrednie barwione lakrofenolem.

Ponadto dokonano niesystematycznego przeglądu literatury dotyczącej historii i przeprowadzonych badań nad mulażami dermatologicznymi z wykorzystaniem wyszukiwarki PubMed. Wykorzystano wyniki przeglądu systematycznego zaprezentowanego w przytaczanym tu artykule pierwszym.

2.4 PODSUMOWANIE WYNIKÓW

2.4.1 Wyniki artykułu pierwszego

Wśród 462 prac poddanych analizie 61% z nich dotyczyło historii, kwestii społecznych lub aspektów muzeologii współczesnej; 8% stanowiły artykuły techniczne; a 31% artykuły badawcze, z których tylko 8% koncentrowało się na badaniach podmiotowych obiektów muzealnych. Znalezione artykuły wykazały przewagę prac z zakresu kulturoznawstwa, z

zaskakująco niewielką liczbą odnoszących się do praktyki muzealnej. Co więcej, w literaturze brakowało konsensusu co do klasyfikacji muzeów anatomicznych. Nie istnieją lub niedostępne są badania w tematyce muzeów anatomii człowieka o wartości praktycznej, mogące stanowić wsparcie kuratorów zajmujących się ochroną dziedzictwa. Publikacja w istotny sposób udowadnia potrzebę prowadzenia badań opartych na metodach naukowych w celu stworzenia rekomendacji i strategii postępowań z unikalnymi historycznymi preparatami medyczno-muzealnymi.

2.4.2 Wyniki artykułu drugiego

Dzięki analizie źródeł historycznych i literatury pozyskano informacje na temat technik konserwacji zbiorów anatomicznych i składów niektórych mieszanin konserwujących stosowanych w XIX. Uzyskano informacje pozwalające częściowo poznać historię badanych kolekcji muzealnych.

W wyniku analiz chemicznych określono niektóre składniki mieszanin konserwujących i ich stężenia. Wykryto m.in. obecność metanolu, etanolu, formaldehydu i glicerolu. Stężenia tych substancji różniły się między próbkami, a ich oznaczenie wymagało zastosowania różnych metod odpowiednich dla poszczególnych składników mieszaniny konserwującej. Żadna z zastosowanych metod analitycznych nie była wystarczająco selektywna aby mogła być zastosowana w podobnych badaniach indywidualnie.

W badaniach mikrobiologicznych z wymazów pobranych z próbek anatomicznych wyizolowano zarówno bakterie, jak i grzyby. Flora bakteryjna była mniej liczna niż flora grzybowa. Wśród bakterii wyizolowano środowiskowe bakterie Gram-dodatnie *Bacillus cereus, Bacillus thuringiensis* oraz rzadką bakterię z rodzaju *Cupriavidus*, natomiast wśród grzybów wyizolowano grzyby drożdżopodobne *Candida boidinii i Geotrichum silvicola* oraz grzyby *pleśniowe Penicillium sp.* i *Fusarium sp*. Jednak ocena mikroskopowa wykazała większą różnorodność mikroorganizmów, których nie udało się wyizolować klasycznymi metodami hodowlanymi.

2.4.3 Wyniki artykułu trzeciego

Ocenie mikrobiologicznej poddano 32 historyczne mulaże dermatologiczne i ich otoczenie (muzeum). Wymazy z okazów okazały się pozytywne w 28% przypadków, wyizolowano głównie *Micrococcus luteus*. Flora wyizolowana z powietrza i zewnętrznych powierzchni

gablot muzealnych była znacznie bogatsza. Oznaczono bakterie i grzyby środowiskowe, a także organizmy prawdopodobnie związane z florą szpitalną: *Pseudosomonas spp., Paebacillus sp., Acinetobacter sp.*. Odnaleziono bardzo nieliczne opracowania dotyczące sposobów ochrony i konserwacji mulaży, natomiast dostępne w literaturze badania były poświęcone głównie identyfikacji zbiorów i ich zastosowania w dydaktyce.

2.5 WNIOSKI

1. Dane literaturowe odnoszące się technicznych aspektów pracy z anatomicznymi obiektami muzealnymi są wysoce ograniczone i nie stanowią jednoznacznego wsparcia do badań naukowych z zakresu muzeologii anatomicznej.
2. Etanol, metanol, formaldehyd i gliceryna w różnych stężeniach to najczęstsze składniki płynów konserwujących muzealne mokre preparaty anatomiczne.
3. Brak jest jednej wysoce selektywnej metody analitycznej oceny jakościowej i ilościowej płynów konserwujących historyczne preparaty anatomiczne.
4. Flora mikologicznych mokrych preparatów anatomicznych jest bogatsza od bakteryjnej i wykazuje większość różnorodność.
5. Muzealne eksponaty mulaży dermatologicznych wykazują znikomą kontaminację mirobiologiczną, przy bogatszej florze bakeriologicznej i mikologicznej całego środowiska muzeum.
6. Badania medycznych obiektów muzealnych wymagają podejścia interdyscyplinarnego z uwzględnieniem przynajmniej aspektów historycznych, chemicznych i mirobiologicznych.
7. Wskazany jest dalszy rozwój badań nad medycznymi obiektami muzealnymi celem stworzenia wytycznych dotyczących zasad ochrony i pracy z tym dziedzictwem.

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3. ARTYKUŁ PIERWSZY:

Terra Incognita in anatomical museology - a literature review from the perspective of evidence-based care.

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Minireview

Terra Incognita in anatomical museology – A literature review from the perspective of evidence-based care

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ABSTRACT

Background: The care of historical collections in anatomical museums is a highly specialized subject requiring advanced knowledge. When searching for practical information on this subject, the authors were not able to find appropriate literature based on scientific research. The absence of this literature is probably due to the specialized nature of the subject and the poorly defined classification of this type of museum.

The purpose of this study is to conduct a systematic literature review to identify (i) the current state of knowledge of anatomical museology and (ii) the nature and determinants of ongoing research on anatomical museum objects.

Materials and methods: A systematic search of the main electronic databases (PubMed, Google Scholar, Scopus) was conducted to identify relevant studies. The records retrieved were categorized according to thematic similarity and scientific content. Based on these groupings, statistics were created based on the number of eligible papers in each particular group.

Results: 61 % of the papers retrieved addressed the history, social issues, or related aspects of contemporary museology; 8 % were technical papers; and 31 % research papers, of which only 8 % were focused on the museum object as the topic of the research. The paper retrieved showed a predominance of works in cultural studies, with surprisingly few applicable to museum practice. Furthermore, there was a lack of consensus in the literature regarding the classification of anatomical museums.

Conclusions: Anatomical museology is a poorly defined concept in the scientific literature and it is a rare topic in contemporary work by anatomical practitioners. The literature review revealed that the debate about the fate of anatomical museums encompasses a broad spectrum of diverse, often disparate scientific fields as well as economic factors that influence the present status and future of these institutions. For these reasons, museum object research is problematic in design, may not be considered worthwhile, or is unattractive from an institutional perspective. The literature survey showed that there is a paucity of work in the available modern literature that provides significant support for museum anatomists.

1. Introduction

1.1. Anatomical museums - current challenges

The decline in the importance of anatomical museums in their conventional form as teaching institutions provokes a discussion about the role that these institutions now play. Academic museums, which used to be a core resource for teaching, have declined in importance in favour of illustrated textbooks, atlases, and digitised collections that are accessible online (Gulczyński et al., 2018; Zhao et al., 2020). In the present climate, in which institutional decisions are based primarily on the economics of operations rather than on

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0940-9602/

historical importance and tradition, the question of the value of maintaining these institutions is highly relevant (Marreez et al., 2010). Europe has many anatomical museums with long histories that are expensive and cumbersome to maintain. In most cases, the historical nature of their exhibition galleries requires the preservation of their original style and the specimens stored therein require specialist care. Due to the fact that anatomical museology is a unique, niche topic and thus difficult to access, the preservation of these institutions is complicated.

Museums play a special role in science as storehouses of material that is often unique and not available today. Museum specimens are useful not only didactically but also in research; for the evaluation of rare, eradicated diseases; analysis of congenital anomalies; retrospective studies; and much more. However, when working with such objects, it is impossible not to take into account to their historical properties, which are often related to the history of the institution and changing conservation trends over time. For example, when conducting research on animal material, it must be understood that different fixing and preserving chemicals and techniques influence the preservation quality of the tissues in different ways that affect the final morphological structure of the specimen. There has been very little such research on human anatomical collections (Simmons, 2014, p.25) and it remains unclear whether the results of research on animal specimens in museums can be applied to human anatomical specimens and, if so, what the relationship is between the two. Thus, the consideration of historical factors when conducting a study is necessary, albeit difficult, due to the complicated histories of many museums in Europe, the multi-layered process of specialization during the development of these organizations, and the insufficient number of studies of museum objects utilizing appropriate laboratory techniques.

1.2. The anatomical museum as a university museum

The history of museums is obscure and complicated (Simmons, 2016), but it is known that the earliest anatomical museums evolved from teaching institutions in which dissections were performed and anatomical specimens were prepared. Over time, some specimens and models passed into private ownership and were exhibited in cabinets of curiosities, which slowly evolved into modern museums beginning in the late 17th century (Lindauer, 2010; Simmons, 2016). In a treatise on museology published in 1727 (Caspar Friedrich Neickel's *Museographia*), it was recommended to exhibit at one end of the gallery "all kinds of anatomical objects, predominantly human objects, as in mummies, embalmed and anatomical children whose skeletons are displayed along with those of fully grown adults and other anatomized parts of beasts and men artificially prepared in balsam and varnish" (Kaufman, 2021, p. 170).

Over time, some exhibitions in anatomical theaters evolved into a specific type of institution—the academic anatomical museum, which responded to the broad and diverse needs of the society, not only the academic community. There is a question regarding the establishment of anatomical museums—the available scientific data indicate that dedicated anatomical museums did not began to develop dynamically until the 18th and 19th centuries, although there were exhibitions of anatomical specimens in academic museums earlier than that (Cole, 1949). According to the philosophy of science of the time, the first true anatomical museums associated with universities combined primitive, pathological, and comparative anatomy. With the evolution of didactical methodology, these disciplines became separated; it happened, for example, in Breslau at the end of the 19th and beginning of the 20th century, which resulted, among other things, in a reduction in the number of animal specimens (Hasse, 1911; Otto, 1827, 1814). A similar process of specialization was experienced at this time by other anatomical museums in Europe (Monza et al., 2019). This change stemmed from the development of the chemical sciences which made it possible to better protect cadavers from biological agents of deterioration (Musiał et al., 2016). As a result, exhibition halls of anatomical museums were gradually filled with a variety of human anatomical specimens. Their appearance, preparation techniques, and methods of protection varied, depending on the time of their creation and the place and method of their exhibition and storage. Because of this, each specimen has great historical value. This exceptional and historic quality of human anatomical preparations deposited in anatomical museums makes them not only valuable from the medical and didactic perspectives, but also interesting from the scientific and artistic point of view (Gulczyński et al., 2018).

Current scientific interest in anatomical collections stems from the study of the progress of medical science, and from the need to assess the anatomical variability and the dynamics of changes in preserved human structures. Historical collections are often the only source of knowledge regarding medical, sociological, and cultural aspects of anatomical specimens (Corradini, 2015; Marreez et al., 2010).

1.3. Restoration of anatomical specimens

The historical legacy can be a source of difficulties in terms of taking proper care of specimens stored in a museum (Bocaege et al., 2013). Due to their value, experimental activities which may irretrievably damage specimens cannot be used with most historic museum specimens. Conservation procedures carried out on such specimens require a broad understanding of historical practices and scientific history and the intellectual trends and scientific philosophy of past times, knowledge of the context in which a given collection was created, the specific techniques used at a given time, and the contemporary state of anatomical knowledge (Edwards and Edwards, 1959; Häner, 2015).

The uniqueness of particular specimens and the problem of determining the scope of work carried out at the time of their creation pose major difficulties in conservation and restoration. For example, the Wroclaw Museum's collection of teratological foetuses contains specimens showing traces of surgical cuts and sutures, but for many years it was impossible to determine the purpose of these operations. An explanation was finally discovered in 19th century references which revealed that some specimens were sectioned in order to describe an anomaly, then their viscera were filled with packets of straw and the skin carefully sutured. The straw colour of their preservative fluids today probably results from the use of these fillers (Otto, 1841).

The challenge of revising the 19th and 20th century historical collection of the anatomical museum in Wroclaw resulted in the need to find scientific publications that addressed technical aspects of the care and management of historical collections in anatomical museums. This project focuses on the search for literature on the preparation, care, and conservation of human anatomical specimens in museums, a specific group of publications that provides clues and guidelines of a proven and predictable nature, making them useful for museum work.

The aim of this project is to review existing scientific publications in the field of anatomical museology in order to find studies whose results may play a role:

- in gaining a deeper understanding of the techniques and procedures for the conservation of anatomical collections;
- in the planning of conservation interventions and other deliberate alterations that resulted in changes to the structure of museum objects;
- in optimizing planned research on museum material to ensure its safety;

- in planned modernizations of the historic museum space whilst maintaining its historical integrity.

An additional aim of this project is to assess the status and types of research in this area that may have practical application for the modernization of museums of human anatomy, and to attempt to categorize these reports. Furthermore, the authors desire to better understand the place of human anatomical museums—which are historically linked to universities—in the overall classification and taxonomy of museums.

2. Materials and methods

2.1. Overview

In order to find useful publications, assess the amount and status of the research, and evaluate the accessibility of the publications, an independent search was conducted for scientific publications available online through a systematic review. The advantage of this research method is that it allows reliable proportions of different groups of publications to be created in a further step.

2.2. Search strategy

A multi-phase literature search was conducted using available electronic databases, including PubMed, Google Scholar, and Scopus, in order to identify the scientific publications appropriate for analysis. Two separate review sessions were conducted using the keywords "museum anatomy" and "museum techniques" (Fig. 1). Publications in any language and time frame were accepted and the resulting list of records was sorted according to best matches. All forms of publications were included, except those matching the exclusion criteria. The AQUA criteria were used throughout the search process to increase the reliability of the literature review (Henry et al., 2017; Tomaszewski et al., 2017).

Difficulties in assessing the qualifications of studies were resolved by all authors unanimously. The results from the three database searches were sorted and summarised to avoid repetition, which resulted in 474 qualified publications for the “anatomy museum” search (Fig. 1). The additional keyword, "museum techniques", was used to avoid omitting relevant publications, and repetitions were eliminated as noted above. There were 308 qualified publications for the second keyword (Fig. 1).

2.3. Selection criteria

Exclusion criteria eliminated publications relating to exhibitions and/or research in the fields of botany, archaeology (e.g., Egyptian mummies), paleontology (fossils), and art. In addition, papers on museums of medicine (history of medicine, forensic medicine, surgery, etc.) and veterinary medicine were omitted if they did not address anatomical collections or techniques. Records concerning genetic or microbiological banks were also rejected. Non-scientific research literature (i.e., not presenting scientific studies) such as catalogues, albums, and museum guides was omitted.

Conference papers, unavailable works, and studies containing incomplete or insignificant data were excluded as well. Books (scientific monographs) were not emphasized because of the difficulty of assessing their eligibility and their low probability of usefulness with regard to the objectives of this work (see Discussion concerning humanities monographs).

Research on non-human animal museum specimens was accepted, as well as papers on zoological museums and collections if they concerned mammals. Works on natural history museums, nature museums, and anthropological collections were accepted if they referred to human anatomy, vertebrate anatomy (mammals), or

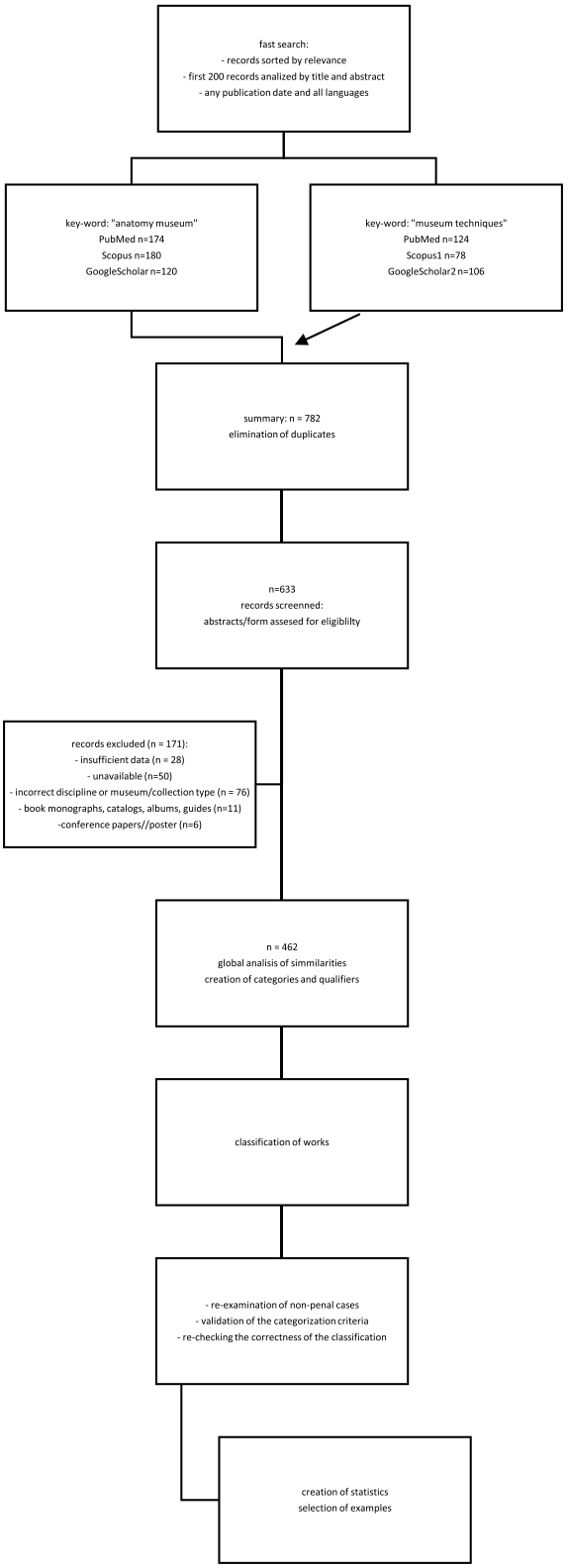

Fig. 1. Systematic search process for the keywords anatomy museum” and “museum techniques” presented as a flowchart of the selection of articles and their compilation. In order to achieve a high level of sensitivity of search results and identify all possible studies, the search strategy was adapted in some cases by modifying the keyword or restricting the disciplines. [1]additional criteria: limit to: medi, bioc, vete, dent. [2]word-key: “museum techniques anatomy”.

comparative anatomy. In addition, papers on anatomical models, wax models or moulages, or plastination were included in the analysis. Research papers on general museums were analyzed individually, as well as works whose object of study was to evaluate the effectiveness or safety of a technique or research method. These were qualified if they had a place in human anatomy collections. Chapters from edited monographs were accepted. All papers not meeting the above exclusion criteria were accepted.

2.4. Data analysis (records screened)

The authors reviewed all articles independently, analyzing the title, abstract, structure, materials and methods, and results, including tables and figures. Works that were in any way unusual or problematic were read in full. Works that were assessed differently by independent reviewers were read in their entirety.

2.5. Data compilation

Following the comparative analysis of the articles (based on discussion and consensus), the researchers determined the structure for the classification of papers in specific thematic groups, and the essential qualifiers for each group were defined. Commonalities among the papers were emphasized in the analysis.

The form of the articles, scientific discipline, concepts and types of research, choice of research methods, objectives, and subject matter were compared to identify global tendencies. On this basis, a first categorization of the papers was made. The articles in each category were then re-analyzed and compared in order to verify conformation with the formulated rules and to validate them, making the method more sensitive.

A selection of articles was made from each group for discussion in the review.

2.6. Statistics

Groups of publications relevant to the aim of the project were compared quantitatively and are presented in the form of a diagram.

3. Results

Three main types of publications were categorized: (i) historical works (i.e., descriptive works on the history of museology); (ii) articles addressing contemporary issues (contemporary museology); and (iii) articles related to museum materials. Within the three types, categories and subcategories were created if additional relationships were found. These qualifiers helped to clarify the categorization of the articles. On this basis, the publications were divided into several primary categories:

- Articles concerning historical topics (Table 1)
- Works on social and ethical issues (Table 2)
- Issues in contemporary museology (Table 2)
- Object-based research (Table 3)
- Studies of research methods (Table 3)
- External research papers (Table 3)
- Non-specific research (Table 3)
- Technical and methodological papers (Table 3)

The rules of classification and examples of reviewed papers are presented in tabular form. Each paper was included in one category only. When the nature of a given article corresponded to more than one category, the authors selected the category which was most significant in the work.

For clarity of analysis, the percentage share of particular categories among the selected publications was determined (Fig. 2).

4. Discussion

An evaluation of world literature, based on scientific criteria, indicated the presence of a significant disproportion of studies concerned with cultural and social issues compared to practical articles (Fig. 2). The lack of the latter is a significant problem for researchers and practitioners who deal with preservation of anatomical specimens in museums. However, many available articles describe anatomopathological collections, which was surprising, but confirms the assumption that many anatomical museums had different functions in the past. Nowadays, such institutions could be defined as hybrid museums because medical education in the eighteenth and nineteenth centuries was not limited to the anatomy of one species (Alberti and Hallam, 2013, pp. 6–7).

In the past, there was less specialization of museums based on scientific disciplines. In addition, the teaching of anatomy involved not only learning gross anatomical structures but also basic microscopic techniques and pathology. Many universities provided comprehensive teaching so that a properly educated doctor was familiar with more than just human anatomy (Bonnel et al., 2019; McLachlan and Patten, 2006).

Even the relatively new Anatomical Institute in Breslau (Wrocław), established by scientists from diverse disciplines, manifests a multi-layered narrative of anatomy in the nature of its museum collections. Of the sixteen thousand specimens (collected by several generations of eighteenth- and nineteenth-century anatomists), at least half are preparations of animal material that have survived until today. In this sense, anatomical museums can be a starting point for research into the history of the institutions to which they belong. Using preserved collections, it is possible to deduce the scientific interests, directions of scientific development, didactic methods, and cultural and social conditions of the past, which proves the thesis stated above that museums contain tangible evidence of the history of their development.

Given the broad context of the education of nineteenth- and twentieth-century anatomists, purely anatomical museums are a modern phenomenon. Describing anatomical collections in the 16th century, one historian of science has pointed out that they “expressed the knowledge and purpose of founders whose major preoccupations were certainly not anatomical, nor did the anatomists themselves in these pioneer days display any enthusiasm for museums”(Cole, 1949). Because many anatomical museums served a wider context than just medical teaching, it is reasonable to distinguish a separate category of anatomical museums alongside medical museums (Simmons, 2014)

On the other hand, it was primarily the teaching and practice of medicine that drove the development anatomical museums. “Human anatomy museums (and especially those in teaching hospitals) gathered their specimens from the unfortunate patients of the adjacent wards, whose body parts radically shifted in meaning during transfer and preservation. By working on these preparations, the surgeon-curator rendered them property *[museal objects]*, …in the process shifting from subject to object.” (Alberti, 2005, p. 563) In the nineteenth century, specific conditions favorable to these collecting tendencies arose, linked to the formation of clinical complexes and their predominant therapeutic-scientific role. In the university cities, the "districts of institutes and clinics" physically brought the clinical wards closer to the theoretical units, enabling both scientific and therapeutic activities to be carried out simultaneously by anatomists-practitioners such as dentists, physicians, and surgeons (Wójtowicz, 2020, chap. 1). Due to these historical and cultural differences, the authors suggest recognizing an additional

Table 1
Articles concerning historical topics.

Category	Qualifier	Description	Sample publications
History of institution n = 53	Institutional emphasis (e.g., descriptions of breakthrough moments for the institution or collection).	• Historical papers—usually describe several intertwining issues. The reasons for the establishment of a museum and its particular collections are presented and the historical-cultural context is determined which makes it possible to establish the development of the institution over time and examine its individual fate. • Specifies objects in the collections and describe their current condition and preparation techniques. • The present situation is presented against the background of history, which provides a basis for reflection on the (sociological) evolution of the museum's role over time. Such articles are usually of a hybrid nature (e.g., include an analysis of sources, elements of review, case studies, comparative research). Sometimes it is difficult to classify these papers unambiguously. • Due to their general, descriptive nature, the methodological aspect of the studies is poorly expressed in these papers.	(Boer et al., 2018; Bonnel et al., 2019; Heylings, 1990)
Biographies n = 39	Biography of a museum's creator(s) in reference to the history of the institution.	• The main theme focuses on the life and output of a specific individual. Sometimes collecting is presented as a continuum of the work of subsequent generations of curators. • The influence of a specific person's activities on the development of the museum. • Description of techniques developed by the museum's creator(s), their original solutions, and collector's and institutional activities. The effect of their output on the local or pan-European environment is analyzed. • Based on these data, the history of the institution is formulated.	(Boer et al., n.d.; Patzak and Winter, 2013;Summerly and Macintyre, 2021)
Articles concerning historical sociology [socio-historical] n = 12	Sociological and psychological research concerning the past; articles focusing on general social phenomena	• Research on the relationship between the museum and the public (society) in the past. • Development of social acceptance of anatomical exhibitions. • Social responses to anatomical exhibitions in the past.	(Fakiner, 2016; Guerrini, 2003; Knoeff, 2015)
History of science, cultural and anatomical museology n = 52	Descriptions of issues in science and the academic community and their evolution over time	• The role of the museum in preserving the continuity of heritage, including intangible cultural and scientific values. • Chronological approach to the establishment of institutions and development of anatomical collecting from a global perspective. • Research limited to elite or specialist social groups • Collecting trends. • History of didactic anatomical methods in medicine using anatomical museums. • Methods of acquiring museum specimens and research material.	(Kamath et al., 2014a, 2014b; Mitchell et al., 2011; Paluchowski et al., 2016)
History of museum techniques n = 16	Chronological development of a given technique.	• The author of a given technique is presented; its effects and evolution over time are described (e.g., articles concerning wax moulage artists) • Technique instructions, typical of technical notes, are omitted. • The importance of particular techniques in the history of didactics and/or science/museums.	(Cooke, 2010; Lotti et al., 2006; Ortug and Yuzbasioglu, 2019)

category of museological classification called anatomical-university museums.

In the context of museum research, the number of articles devoted to the theory of anatomical museums is small. As stated by Felip Cid, "Medical museology depends on research on the collections, since this will produce heuristic interactions that will revert to museographical thinking, which, despite emphasizing the containers more than the contents, it is evident raises museological signals to be resolved" *[Museología médica depende de las investigaciones sobre los fondos, puesto que originarán unas interacciones heurísticas que revertirán en unos replanteamientos museográficos, los cuales, pese a imponer más los continentes que los contenidos, es evidente que suscitan signos museológicos a resolver]* (Cid, 2007, pp. 738–739).

4.1. Historical topics

There is a lack of articles that systematize the key concepts of anatomical museology and why it is distinct (Monza et al., 2019). The usefulness of publications from the non-scientific studies group, from a practical perspective, is that they can help (by comparison) to understand the general context of the times in which the development of anatomical collections took place (Dittmar and Mitchell, 2016). Descriptions of historical methods can be helpful in identifying museum specimens and the techniques used in their preparation (Cooke, 2010), however, in these papers the technical aspects are not described precisely enough to be useful for reproducing the old preparation methods. According to Samuel

Table 2
Issues in contemporary museology.

Category	Subcategory	Qualifier	Description	Recent publications
Social and ethical issues n = 70	Social research n = 43	Articles that systematically investigate society and social changes.	• Communication theories. • Research on the relationship between the museum and the public. • Sociological, psychological, and pedagogical articles concerning modern museums. • Projects combining various didactic-related scientific disciplines. • Current didactical issues in anatomy	(Monza et al., 2019; Sane et al., 2015; Yudhawasthi et al., 2019)
	Discussions combining different fields of social sciences n = 27	Reflective, philosophical, and ethical topics.	• Ethical and legal issues concerning anatomical exhibitions. • Ongoing challenges facing museums and the future of museums	(O'Sullivan and Jones, 2015; Sane et al., 2015;Wakefield, 2007)
Modern technologies n = 36	Modern solutions	Articles concerning the establishment and function of modern anatomical museums.	• Articles devoted to the techniques of digitization, online museums, and modern methods of presenting collections and educational technologies.	(Gangully et al., 2003; Mogali et al., 2019)

Alberti, "To understand the intellectual, political, and cultural significance of the collections they house, then, we need to know about museums' practice. We need to look beyond the displays and texts to the instruments and techniques, and the materiality of objects (…)"(Alberti, 2015).

Among the publications in the socio-historical and history of science, cultural, and anatomical museology categories, most papers deal with social or scientific phenomena related to a single city or institution. Similar analyses in a global context are far less common (Kamath et al., 2014a, 2014b; Mariño Gutierrez et al., 2019). Based on the papers in these categories, and knowing the time frame of the creation of a particular collection, it is possible to make deductions about the reasons and background of its creation.

4.2. Social and ethical issues

Works covering contemporary issues in museology and social aspects related to anatomical museums are classified in this category. Some of these (e.g., social research) are useful for the creation or management of contemporary anatomical exhibitions, while others focus on didactics and the use of museums for education.

A variety of topics are addressed in the papers in the discussion that combine different fields in the social sciences group. For example, several articles discuss the role and future of anatomical museums (Ferrari et al., 2001; Wakefield, 2007). Another topic addressed in some of these papers is the issue of the Body Worlds and similar exhibitions and their ethics (Jones, 2002; Preuß, 2008).

The topic of ethics appears in articles addressing legal and social ramifications related to the complex issue of understanding anatomical collections as human remains and cultural heritage (Monza and Licata, 2015; Ousley et al., 2005; Sane et al., 2015; Svanberg, n.d.). Collecting issues are particularly relevant today in relation to historical collections, which were created at a time of different ethical standards in science than are accepted today. According to many of the authors, no other type of museum is as ethically sensitive and at the same time as controversial as museums of human anatomy.

Articles in the modern technologies category focus on virtual exhibitions and the use of technology in the digitization of collections. Similar papers on the handling of historical collections would be useful.

4.3. Issues in contemporary museology

This group of publications includes technical and methodological descriptions that could be used during collection renovation or verified by other research to better understand and protect museum collections.

4.4. Research work unrelated to museology

A large number of studies conducted on museum material can be used as reference points as a control test for this research. This is most readily seen in anthropological, paleoanthropological, and molecular research. For example, an exemplary paper by Göbbel and his team describes the occurrence of congenital diseases using historical material collected by several generations of anatomists of the Meckel family (Göbbel et al., 2007). Hampl and colleagues used skulls from anatomy departments and museums that were measured and evaluated to assess the topography of the mastoid foramen (Hampl et al., 2018). These papers demonstrate that anatomical museums house and protect specimens of substantial scientific importance, and proves the paradox of modern times that with technological progress, (i.e., the development of new research methods) museum collections become more scientifically useful. At

Table 3
Articles based on research on museum material.

Category	Subcategory	Qualifier	Description	Recent publications
Object-based research n = 37	Descriptive research n = 18	Analytical papers using observation, comparison, and source analyses (without a technical or laboratory aspect).	• Specimen-based research such as examination of collections for cognitive purposes. • Exploration of a museum object to establish its properties or identity. • Research based on museum objects. • Papers that analyze collections individually (e.g., identification studies of a museum objects) • Descriptive research, often based on the analysis of sources • Technical papers using research methods specifically designed or adapted to the characteristics of the museum collection and its characteristics.	(Conde-Salazar Gómez and Heras Mendaza, 2015; Scarani et al., 2001; Skrzat et al., 2018) (Boer et al., 2017; Panzer et al., 2012; Santi et al., 2019)
	Technical and laboratory studies n = 19	Interdisciplinary research using laboratory and technical tools, standard analytical methods.		
Study of research methods n = 16		Presentation and analysis of the application of a modern, specialized research method	• Scientific papers testing the effectiveness of a particular research method on museum-historical material • Technical descriptions and discussion of the application of research and diagnostic methods • Rarely analyze a specific museum specimen but most often a group of similar objects. • The conclusions obtained in the context of museum objects are rather general and derivative to the aim of the presented study.	(Burrell et al., 2015; Lauridsen et al., 2011; Tsangaras and Greenwood, 2012)
Research work unrelated to museology n = 65		Collections are the object but not the subject of research.	• Research motivated by external, non-museological ideas • Research that does not result from the needs of a museum object. • Cognitive conclusions based on the field of study rather than the material analyzed. • Articles analyzing a group of collections as a study or control group.	(Göbbel et al., 2007; Oostra et al., 2005; Sedivy et al., 2011)
Non-specific research n = 28		Conservation and research issues irrespective of museum type	• Research covering different types of museums, results can be applied to anatomical museums. • Studies with practical value and/or containing technical elements • The type of museums in which the research was conducted does not play a significant role in the research methodology. • The methodology of the work, if adapted, could be useful in an anatomical museum	(Gibson et al., 1997; Samide and Smith, 2015; Shen, 2021)
Technical notes n = 38	Pure technical notes n = 22	Technical work without analysis of the museum aspect.	• Works on museal, anatomical. or conservation techniques. • Articles containing technical instructions for reproducing a particular effect or type of preparation. • No information concerning the durability of a described technique over time.	(Brundelet, 1963; Hamilton, 1977; Magro et al., 2015)
	Technical-analytical notes n = 16	Technical papers containing an analysis of a given method described in the museum context.	• Detailed description of a given method and its implementation. • Articles that analyze a method described in a wider context or research context. • The choice of methods is based on observation or investigation • The durability or usefulness of the method is based on additionally performed verifications or research.	(Dwivedi et al., 2014; Prabhu et al., 2015; Sandhyamani et al., 2005)

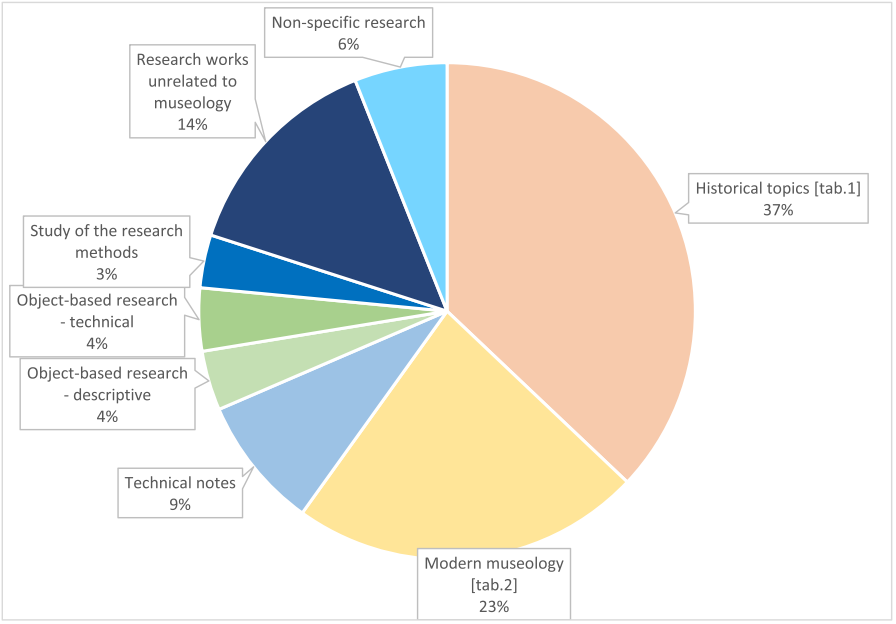

Fig. 2. Percentage of each type of research work.

the same time, museums, as the depositories of this scientific potential, are failing due to economic reasons (Kemp, 2015).

4.5. Object-based research

In the opinion of the authors, proper research and museological articles are those that explore the museum from within (Table 3). Identifying collections and understanding the specimens they contain is the reason for scientific research concerning a specific method and its effects. Although this group of articles has been divided into (i) descriptive (non-laboratory) and (ii) technical (laboratory) categories, theoretical (anatomical, collector) and practical knowledge (e.g., anatomical techniques) are necessary to understand the issues and broad context of the studies in both cases. Research is usually interdisciplinary, consisting of the following stages: (1) collection analysis; (2) evaluation of written source materials; (3) comparative or laboratory research; and (4) comparison of results with other available research. The reasons for creating these articles vary but include (i) studies of specific museum specimens (Santi et al., 2019), which is to say, to learn about the properties of an object (Drifi and le Floch-Prigent, 2009) or its identification (McCulloch et al., 2002); (ii) research concerning a museum section (Boer et al., 2018); or (iii) source research (McCulloch et al., 2002; Skrzat et al., 2018).

Whereas in past centuries the effectiveness of the techniques used were verified by observation (Alberti and Hallam, 2013, p. 3) or by trial and error, the modern stock of analytical possibilities seems inexhaustible. Therefore, the small number of available scientific publications devoted to this topic in indexed scientific journals is surprising (Simmons, 2014, p. 66)i

4.6. Study of research methods

Developments in genetics and molecular biology have created new uses for museum collections (Baker, 1994; Burrell et al., 2015). Genome analyses are used in biodiversity studies (Ewart et al., 2019), animal evolution, systematics, climate change (Bi et al., 2013), and epidemiology (Tsangaras and Greenwood, 2012).

In the context of anatomical museums, laboratory methods (e.g., PCR, Sanger's method) as well as medical diagnostic methods (e.g., magnetic resonance, computer tomography) are used in object-based research to identify animal species that constitute specimens in the collection or to diagnose diseases or anomalies presented by anatomical specimens (Boer et al., 2017; Oostra et al., 1998). Microbiological analyses are used to identify and study the genomes of microorganisms extracted from museum materials(Kemp, 2015; Tsangaras and Greenwood, 2012), but we did not find research that analyzed the microbiomes of anatomical specimens for the design of conservation efforts, even though bacteria and fungi may be a significant factor in damaging specimens (Simmons, 2014, pp. 56, 109-119)

The difficulty of determining which papers belong in this group was due to their thematic relationship with the others (technical notes, object-based research, and general museum research publications), which is one of the limitations of the method used. The relevance of these works lies in the fact that the techniques presented are adapted to a particular type of research material, which is to say, museum-anatomical material. In some of the papers, the authors recognize the importance of protecting the specimens that are studied and of creating research methods that are not destructive (Hofreiter, 2012; McDonough et al., 2018; Wisely et al., 2004). These actions—dictated by a concern for heritage rather than research pragmatism—are worth singling out as ethical models.

4.7. Technical and methodological papers

The few technical works that were found mostly dated from the 20th century. The majority of these are hard to access, and the techniques described in them are difficult to use. For this review, more than half of the technical papers were rejected from the analysis due to lack of availability. A relatively small number of contemporary technical notes concerning museum and anatomical techniques have been found. Perhaps these topics are no longer attractive or have been superseded by a form of technical papers subordinate to research objectives, the development of which is easier to obtain funding for.

Technical notes may be appropriate for creating new collections, but most of them are not useful for conservation work. Another reason for the lack of new papers may be the fact that many historic

museums are now facing the more significant problem of survival (Marreez et al., 2010). Hence, the topic of enriching the collection and studying its historical development becomes a secondary issue. Traditional specimens (and specimen preparations) seem to be less attractive than alternatives such as digitized images or 3D projection techniques, especially in less traditional environments.

This situation can be related to the history of the anatomy department in Breslau. In the 19th century Prof. Hans Carl Leopold Barkow built up a large collection of vascular injected mummies including both animal and human specimens, which he used for extensive research on angiology and syndesmology (Barkow, 1869). These preparations were added to the anatomical museum and were popular research objects in the following centuries, which was a decisive factor in the choice of Prof. Carl Hasse as the successor to Prof. Barkow. It is not known which of the reasons for the creation of the collection—scientific or didactic—was primary (perhaps both?). From today's perspective, the research methodology would be designed differently (e.g., using angio-CT/x-ray with contrast), resulting in research data without the need for time-consuming preparation techniques. In the past, many specimens in museum collections were created as a by-product of scientific research. Due to the evolution of research methods, many anatomical techniques have been abandoned, resulting in fewer specimens being created that have exhibition potential.

One of the aims of the present analysis is to provide scientific support for institutions planning to revitalize anatomical museums. This was the basis of the search for (i) detailed technical notes on old museum techniques together with their contemporary analysis and (ii) modern conservation and exhibition methods supported by research on their usefulness (e.g., conservation, reactivity of substances, visual effects). During the analysis of the available literature, few such articles in the latter category were found.

In his article, an example of pure technical work, Moore outlines very meticulously the techniques for replacing preservative fluid, installing the preparation, and sealing museum the containers (Moore, 1980). This is an example of very useful information for everyday conservation practice. In one chapter of the *Handbook of Autopsy Practice* (Ludwig and Edwards, 2002), the authors reviewed selected techniques, mainly based on earlier publications, including three leading works in the field of anatomical museum techniques (D. H. Tompsett, 1956; Edwards and Edwards, 1959; and Pulvertaft, 1950). Unfortunately, these publications are now largely outdated in the light of modern technological developments. Sandhyamani and colleagues, evoking the ethos of ancient anatomical practices, presented a rare technique for recoloring slides and refined it (Sandhyamani et al., 2005). The meticulous analysis of its effectiveness at the cellular level and the retrospective data on the sustainability of the method are significant contributions. Unfortunately, few similar works with such instrumental accuracy have been found. Thus, although there are articles referencing some of the past practices in collections (e.g., monographs devoted to so-called anatomical museum techniques), only a few of the papers contain the much-needed analytic retrospective. For example, Kaiserling's method (developed in the 19th century) is consistently repeated in review articles, but in the long history of museology, specimen preparations were preserved using many different substances, sometimes particular to specific institutions, and even Kaiserling's method has several variations (Toledano, 2016). Knowledge of these methods and verification of their effectiveness are essential in conservation work if it is to be historically informed.

4.8. Non-Specific Research

The authors were surprised by the potential application of these papers to anatomical museums and as a source of inspiration in designing their own research. For example, the paper "A diffusion tube sampler for the determination of acetic acid and formic acid vapours in museum cabinets" investigates the concentration of a toxic substance in museum spaces using a meter (Gibson et al., 1997). Similar studies measuring the concentration of formic acid (an oxidation product of formaldehyde) in anatomical museums could help identify occupational exposure of museum workers and would be useful for assessing the quality of the seals of containers of preservative liquids displayed in the museum. Two other studies report on the use subjective tests to investigate the effect of corrosive materials in the museum environment (Samide and Smith, 2015; Shen, 2021).

There are probably other such publications, but they were not identified in the search, and no works with similar themes were found in reference to human anatomy museums. This is further proof that the niche of anatomy museums is still poorly understood.

5. Summary

The significant disproportion of descriptive (historical-social) and research publications probably results from the difficulties concerning the creation of the latter. While museum and historical topics are extremely popular, practical research requires the establishment of interdisciplinary teams, a wide range of research tools, and, consequently, financial resources. Furthermore, during the course of the research there is no certainty as to the intended results. These types of articles are intended for a narrow audience, which may further limit the publication of these sorts of papers.

The available literature may provide a general idea of the history of a given collection, but it is rarely sufficient to provide guidelines for its care and maintenance.

The authors are aware of the existence of valuable publications in the subject area which have not been indexed in searches. Many of these are published in specialized journals with limited distribution which are of limited availability. Similarly, it is probable that the general contribution of papers concerning cultural and social aspects of anatomical museums is much larger than was shown in this study because much of this knowledge is contained in existing books and journals that were not indexed in the search, which is a limitation of the analysis.

It needs to be emphasized that the available articles about the history of past anatomical methods and practices have limited utility for anatomical practitioners whose goal is to provide scientific care and maintenance for anatomical museums. Unfortunately, the authors who undertake publications on this subject usually do not come from institutes of anatomy and thus their publications are either merely descriptive or are very theoretical and devoid of significant practical advice. Without detailed historical knowledge about a given technique, step-by-step instructions, and contemporary equivalents for archaic reagents, it is usually not possible to repair a damaged specimen or preparation, reconstruct a missing part, or repair damage while simultaneously respecting the specimen's historical integrity.

The results of this review indicate that in the case of anatomical museums, particularly university anatomical museums, the literature generally considers these institutions in terms of the social, philosophical, and historical aspects of museology, while ignoring technical issues. This is surprising because these museums were created by scientists (anatomists-practitioners), doctors, anthropologists, ethnologists, and the artists and craftsmen who worked on their behalf. Anatomy is an extremely practical science that requires sublime technical skills. Research requires material that the researcher has to prepare and evaluate—this inspires the creative process, but it is the actual techniques—the craftsmanship that evolves into art—that create the anatomical preparation. Thus, the making of an anatomical museum object requires intention, intellectual input, and technical contributions that get one's hands

dirty. Many papers were found that analyze anatomical preparations with exceptional aesthetics, but in these works the effect is described rather than its results. The impression made by the specimen, the emotional message, the sociological context, etc., are all emphasized, but there is no concomitant mention of what the specimen actually represents, its anatomical value is, or how was it created. What craftsmanship did the anatomist show? What actually determined the preparation's creation and for what purpose? Would it be ethical to create an anatomical specimen for aesthetic purposes only?

It should also be noted that ".every object in an anatomical museum is a product of time-specific practices," while "Every action—including alterations to an object—has an effect on the history of the collection" (Häner, 2015). For this reason, both the scientist and the conservator working on a museum object must consider the historical integrity of the object. The interdisciplinarity of this issue emphasizes the need for thorough preparation and input from different, often unrelated disciplines.

There is a dearth of publications with retrospective and prospective analyses that provide information concerning the application of anatomical techniques and their usefulness. This is an unfilled research niche, perhaps caused by low interest in the topic, or the gap may be the result of the decrease in importance of anatomical museums in the last century due to the availability of information via the Internet, the popularization of images of the corpse (e.g., the Human Body Exhibition and Body Worlds) (Alberti and Hallam, 2013, p. 13), or attractive didactic aids that have made the process of anatomical education independent of museums (Marreez et al., 2010; Sugiura et al., 2019; Wakefield, 2007).

For economic reasons, some museums have been closed or their exhibition areas or collections have been reduced. In addition, the need for space is putting increasing pressure on museums to favor laboratories which are considered to be of greater importance (Marreez et al., 2010; Wakefield, 2007). As a result of the availability of data, universities no longer feel the need to present tangible forms of the scientific output of their researchers, hence the role of academic museums in image creation has declined and is often considered to be an extravagance; this function seems to have been taken over by online exhibitions.

The anatomical museum in Wrocław faces yet another problem. Due to warfare, theft, and the transfer of specimens to other cities after war, the original [German] collection was reduced by three-fourths and the museum exhibition area was reduced by half. To this day the whereabouts of the lost specimens and preparations have not been established.

The problem of the museum's historical identity and adaptation to changing times (Turk, 1994) and the change in the traditionally used communication systems (Yudhawasthi et al., 2019) affects even institutions with an established position in European culture. The subject of concern is the future of these institutions and the actions to be taken to secure their funding (Turk, 1994). From a pragmatic point of view, modern science, with its increasing interest in historical collections, needs museum collections. For this reason, the topic of collections care will remain relevant, even as anatomical museums are facing destruction in the present age.

6. Conclusions

The classification of anatomical museums is a complex issue, requiring knowledge and comparative analysis of their origins around the world. Different categories have been used to categorize contemporary museums, whose form was established when they were founded, compared to museums with a long history, whose structure and functions have evolved over time. The care of heritage is the responsibility of the institutions that have custody of them, because they are part of the cultural patrimony of all mankind, not merely their owner. As we have shown, anatomical museums have had an impact on the development of science and are still useful for research, therefore it is important to maintain them for posterity. The scientific community has a special responsibility for these institutions, hence observations and research on them should be encouraged and disseminated, because knowledge is a common good.

The authors hope that this text will inspire all those who care for museum collections to be proactive and bold in presenting their observations and conclusions, as each one contributes to the general awareness of the importance of these museums.

6.1. Limitation of method

Considering museology as a humanistic science, the method used here may seem too mechanical, but the authors have applied it deliberately. It needs to be emphasized that there are sources that can be helpful in working on museum objects which were not included in this review (e.g., catalogs, albums, conference reports, archival handbooks on anatomical techniques). However, it is not technically possible to compare these sources with the available scientific research publications reviewed here. The aim of this review is, in fact, to find empirical studies in the medical and natural sciences as well as technical papers on the specific phenomenon that human anatomical museums represent. Systematic review is one of the most sensitive methods used in medical science to identify and analyze such publications. Moreover, the authors have adapted this review methodology to objectively assess the quantitative proportions of the given categories of works and thus infer scientific and publishing trends, which is not possible with any other method.

With regard to the method used, it is important to explain the selection of the key word "museum techniques." In the review, information was sought on broad activities involving museum objects, so for this reason it was not possible to limit the search only to anatomical preparations, although these constitute the bulk of the anatomy museum exhibitions. The term "anatomical preparation" is not the same as "museum specimen." For example, a preparation as a museum specimen is a coherent whole, established by its maker, consisting of properly preserved tissues, the container in which they are stored, and the way that they are displayed, described, and sealed, and intended for long-term preservation. Although there is no consensus in the literature on the nomenclature for the activities involved in preparing museum specimens (for example, the "anatomical techniques" of Thompsett; the "medical museum techniques" of Edwards and Edwards; and the "museum techniques" of Pulivneart), the authors have decided that the term "anatomical techniques" is insufficient to describe the work of creating museum specimens. This is further supported by the fact that museum specimens do not necessarily consist of tissues (e.g., corrosion preparations, plaster models, moulages). The term anatomical techniques may refer to dissection or preparation techniques such as procedures for eliminating certain types of tissues in order to separate them and visualize others, which constitute the subject of the preparation, or to preparations intended to be used for a specific purpose and discarded (not preserved). Thus, preparation creates a specimen, but not necessarily a museum object.

Ethical statement

I testify on behalf of all co-authors that our article submitted to Annal of Anatomy:

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CRediT authorship contribution statement

JD: Conceptualization, Methodology, Investigation, Writing – original draft. ZD: Methodology, Writing – original draft. JES: Writing – review & editing. MW: investigation.

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4. ARTYKUŁ DRUGI

Preservation fluids of heritage anatomical specimens -a challenge for modern science. Studies of the origin, composition and microbiological contamination of old museum collections.

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ORIGINAL ARTICLE

Preservation fluids of heritage anatomical specimens — a challenge for modern science. Studies of the origin, composition and microbiological contamination of old museum collections

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Abstract

Anatomical museums preserve specimens of great historical value and undiscovered scientific potential. However, frequently these collections lack documentation of the techniques of preparation and the composition of preservative substances (conservation principles). This poses a huge problem for the care and preservation of these materials, more so because understanding this issue requires knowledge of fundamentals from different scientific disciplines. The aim of the research was to obtain information about the composition of substances used to preserve historic specimens, as well as to conduct a microbiological assessment of the specimens to detect possible factors causing their deterioration. Furthermore, we wanted to fill an existing gap in the literature, as there is a lack of reports on analytical methods that could be successfully applied by anatomists involved in the daily care of museum collections in human anatomy departments. The starting point was the analysis of the sources and history of the collections, on which basis the choice of research methods was made. Methods based on simple chemical reactions and specialised methods (such as gas chromatography-tandem mass spectrometry, Fourier transform infrared spectroscopy, inductively coupled plasma optical emission spectroscopy) were used in the analyses of the composition of fluids. Microbiological analyses were based on culture and isolation methods, analysis of microscopy slides and matrix-assisted laser desorption/ionisation time-of-flight mass spectrometry analysis. As a result of these analyses, some components of the preservative mixtures and their concentrations were determined. The presence of methanol, ethanol, formaldehyde and glycerol was detected, among other chemicals. The concentrations of these substances were different between the samples and their determination required the use of a variety of methods suitable for the individual components of the preservative mixture. In microbiological tests, both bacteria and fungi were isolated from swabs taken from anatomical specimens. The bacterial flora was less numerous than the fungal flora. Among the bacteria, environmental Gram-positive *Bacillus cereus*, *Bacillus thuringiensis* and a rare bacterium of the *Cupriavidus* genus were

isolated, whereas among the fungal organisms, the yeast-like fungi *Candida boidinii* and *Geotrichum silvicola* as well as mould fungi *Penicillium* sp. and *Fusarium* sp. were detected. However, the microscopic evaluation showed a greater diversity of microorganisms, which may be related to the fact that many environmental bacteria cannot be cultured using classical methods, but can be observed under the microscope. The results of the research made it possible to draw conclusions about the mutual influence of physical, chemical, and microbiological factors on the condition of historical anatomical specimens. In the course of the research, information was obtained on the processes which could have taken place during the storage of these collections. Maintaining the integrity of a container housing a preserved anatomical specimen has a major impact on maintaining the concentration of preservative fluid and keeping the specimen environment sterile. Many conservation procedures for historical specimens carried out nowadays pose a risk of destroying valuable specimens, as well as a health risk for the person carrying out the work. The exploration of the topic of conservation of anatomical specimens, especially those that lack documentation of their origin, is a key issue in current research on historical collections of anatomical specimens.

KEYWORDS
anatomy museum, chemical composition, microbiological contamination, preservative fluids

1 | INTRODUCTION

Measurements of fixed and/or preserved tissues are commonly used in morphological sciences (Waltenberger et al., 2021). For developmental and foetal anatomy, formaldehyde-preserved research material is the main source for morphological analysis (Beger et al., 2021; Grzonkowska et al., 2021; Paruszewska-Achtel et al., 2018).

It has been shown in animal examples that formaldehyde constricts specimens, changing the original morphological dimensions (Patten & Philpott, 1921). It has also been proven that different chemical mixtures affect the dimensions of organs fixed or preserved with them in different ways (Bakıcı et al., 2017). The effects of preservatives on human tissue are difficult to predict.

Foetal developmental studies are often conducted on material collected and preserved in the distant past, because due to legal changes and advances in medical science, obtaining such specimens is virtually impossible today (Dudek et al., 2018; Krzyżak et al., 2014; Masłoń et al., 2013). In addition, medical developments have eliminated many of the diseases of pregnancy and the puerperium that were commonly observed in the past (Hokama & Binns, 2012). These factors have resulted in reduced availability of fresh foetal specimens depicting concrete infectious diseases, malformations, or congenital syndromes. Thus, the progress of medical science has indirectly strengthened the importance of historical museum collections—preserved historical foetal specimens have become a unique educational and research source (Paluchowski et al., 2016).

A significant limitation to the usefulness of historical anatomical specimens is the sparse amount of data that has survived to the present day. Many collections survived the periods of war in the 20th century in a limited form, with the technical documentation usually severely damaged and dispersed. The lack of technical information makes the contemporary conservation of the anatomical specimens difficult. The methods of preservation and the chemical composition of the historic preservative fluids remain unknown (Domański et al., 2023).

To preserve museum collections in the best possible condition, conservation activities should be based on methods with proven safety and effectiveness. Research on human foetuses has been conducted in the Department of Anatomy of the Medical University of Wrocław since the end of the Second World War (Goździewski et al., 1985; Nizankowski, 1967). The rich collection of teratological and developmental specimens stored in the Museum within the Department of Anatomy indicates that the tradition of this research in Wrocław is much longer and is connected with the pre-WWII period (Domański et al., 2023; Otto, 1841).

In the course of inventory work in the basement of the 19th-century building of the Department of Anatomy of the Medical University of Wrocław, at the turn of 2019/2020, interest was aroused by a neglected anatomical collection that consisted mainly of teratological specimens. The extent of their conservation is unknown. This discovery initiated efforts to rescue and conserve the collection, which was most likely hidden in the basement of the building during World War II to avoid damage during bombing.

An interdisciplinary team was formed to undertake conservation work on the stored anatomical specimens in order to save the collection and conduct a study to provide new insights into the effects of preservatives on the condition of the tissues. A variety of chemical and microbiological analyses were carried out for this purpose, as well as a literature review and analysis of historical sources.

The aim of our research was to determine the chemical composition of the preservative fluids and conduct a microbiological evaluation of the preservative fluids and the specimens.

In addition, we wanted to assess whether simple methods for detecting the chemical composition of preservatives were useful and whether they could be compared with recent advanced analytical techniques (e.g., gas chromatographery-triple quadrupole tandem mass spectrometry [GS–MS/MS], Fourier transform infrared spectroscopy [FTIR], inductively coupled plasma optical emission spectroscopy [ICP-OES], matrix-assisted laser desorption/ionisation time-of-flight mass spectrometry [MALDI-TOF MS]).

2 | MATERIALS AND METHODS

The process of material selection, the choice of research methods, and the selection of techniques used were multistaged and followed the objectives of the project (Figure 1).

The results from the three research cycles (source studies, chemical analysis, and microbiological analysis) were compared, as aspects of this researches were found to be strongly associated. This resulted in conclusions that are valuable for conservation as well as scientifically relevant.

2.1 | Material

The material came from the collection of historical museum anatomical specimens deposited in the Anatomical Museum of the Department of Anatomy of Wroclaw Medical University.

The study group consisted of 10 jars, selected from the recently found collection of foetal specimens.

The basic criterion for qualification was visual defects of the anatomical specimens maintained in an unspecified preservative fluid. Major defects were considered to be any indication of deterioration detected during the visual examination of the specimens such as discolouration, sedimentation, oxidation observed directly on the specimen, precipitates in the bottom of the containers, or significant loss of clarity of the fluid. All specimens were stored in their original jars.

The control group consisted of six teratological specimens stored in the exhibition area of the Anatomical Museum.

The closures were effectively sealed to the jars with a hard black binder. The specimens within the group were characterised by morphological similarity, state of preservation, visual aspects of preservative fluids, and identical containers.

Descriptive (Table S1) and photographic (Figure 2) documentation was created. The parameters were described: (i) container (shape of the jar, condition of the closure), (ii) organoleptic characteristics of the preservation fluid (clarity, colour, impurities, presence of paraffin), and (iii) anatomical preparation (colour, tissue fixation, nature of the lesions examined, material with which the body cavities were stuffed, if visible).

Samples were taken from the surfaces of anatomical specimens and their preservative fluids, using aseptic techniques for microbiological, mycological, and chemical analyses. To obtain samples suitable for analysis, the fluid was aspirated using a catheter into a 100 mL syringe from different depths of the jar and then injected into clean, sterilised glass tubes.

2.2 | Source studies

A review of the scientific literature was carried out in order to select appropriate laboratory methods for the evaluation of the composition of preservative fluids (Figure 3a).

The literature was searched for information on methods of preserving anatomical specimens and possible methods of diagnosing fluid composition. The results of two literature reviews were used: (i) a systematic review (Domański et al., 2023) based on a search of scientific papers in PubMed, Medline, and Ebsco databases and (ii) a non-systematic review based on papers published in the journal 'Museologica' and evaluation of literature lists from the monograph 'Fluid preservation: a comprehensive reference' (Simmons, 2014). Additionally, monographs on the history of the Medical University of Wrocław (Kozuschek, 2002, 2007) were analysed.

The results of these reviews determined the choice of methods for the next stages of the research.

2.3 | Chemical composition analyses

Specialised chemical composition analyses were carried out on the fluids taken from the specimens in the study group and control group (Figure 3b).

FIGURE 1 A diagram of the research process (simplified).

FIGURE 2 Selected specimens from the study and control groups. (a) Museum specimen M30. The apparent excellent condition of the specimen is illusory. A white deposit has collected on the surface of the skin, visible around the mouth and arms. (b, c) Specimen A40 in its jar and removed from the container. The fluid is visibly turbid, sediment is in the bottom of the container, and flocs are floating near the surface. The face of the specimen is covered with sediment. (d) Specimen A20. Heterogeneous dark lesion on the knee. (e, f) Specimen A65 in its jar and removed from the container. Note the low level of preservative fluid and brown deposits on the surface of the specimen.

The samples were analysed to determine the identity of the substances composing the fixative and preservative fluids, based on information acquired during the analysis of the source components (ethanol, phenol, glycerine, formaldehyde, and acetic acid were typed).

The analyses were conducted in two steps: first, the fluids were characterised by simple chemical reactions with a low-cost test, in order to determine the presence or absence of the substances typically composing the solution.

The reactions were selected according to the recommendations of the Polish Pharmacopoeia, 11th edition, or those supported in the literature (Morisson & Boyd, 1985; Robins et al., 2011; Simmons, 2014).

A standard blind test was conducted before each chemical reaction was carried out.

Selected qualitative samples are described in Table 1.

The pH of the preservative fluids was also determined at this stage.

The pH measurement was obtained with a meter using a combined glass electrode (pH & Ion-Meter, GLP22+, Crison Instruments S.A., Spain). The electrode was calibrated based on standard buffer

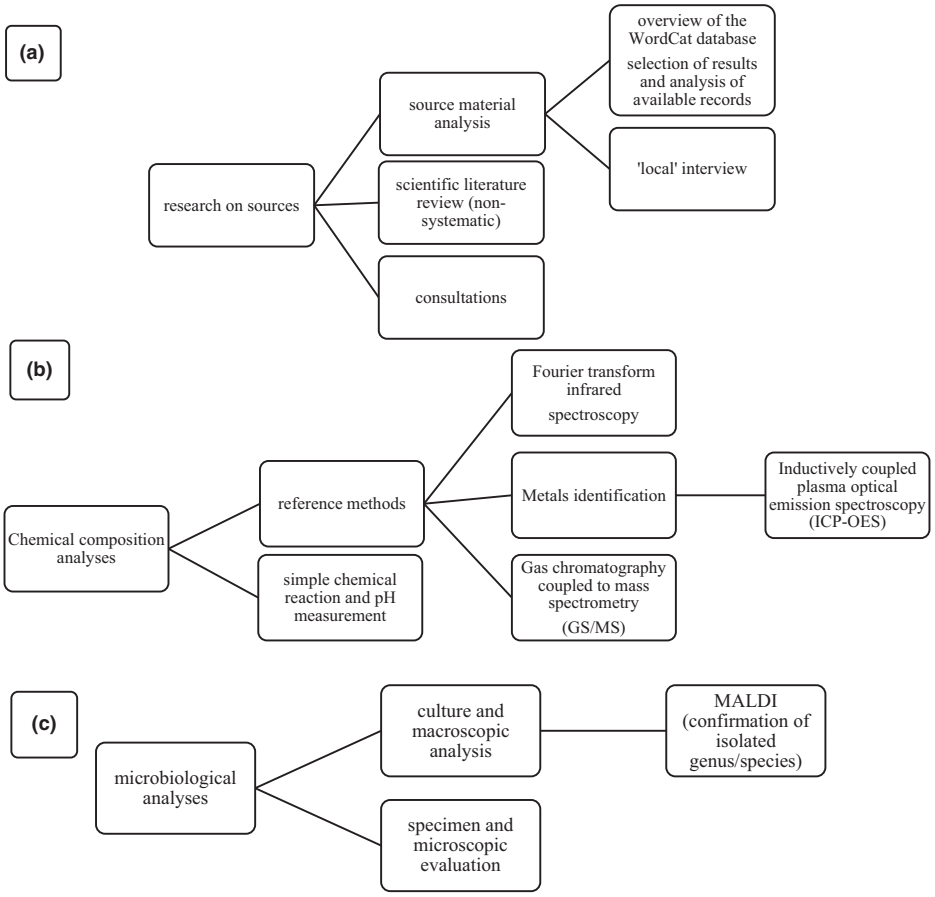

FIGURE 3 The process of particular stages of the research. (a) Search for and analysis of source materials. (b) Analysis of chemical composition and observations of simple chemical reactions and pH measurement based on additional tests. (c) Microbiological analyses.

solutions (pH 4.00, 7.00, 10.00). All measurements were repeated in triplicate and then averaged.

To compare the accuracy of the achieved results, a qualitative and quantitative analysis was performed using reference methods. These tests constituted the second step in the chemical evaluation of the preservative fluids from both study groups. GS–MS/MS, FTIR, and ICP-OES were used to complement the chemical and biological research.

2.3.1 | Determination of formaldehyde, ethanol, and methanol by gas chromatography/triple mass spectrometry

Formaldehyde standard stock solution (37.4 wt % in water), *O*-(2,3,4,5,6-pentafluorobenzyl) hydroxylamine hydrochloride (PFBHA), formaldehyde *O*-pentafluorophenylmethyl-oxime (10 mg/1 mL in water) and cyclohexanone 1 mg/1 mL (in water), methanol (99.9%), ethanol (99.9) were purchased from Merck KGaA. Deionised water was used as blank water. PFBHA solution was prepared by diluting 10 mg of PFHBA in blank water to a concertation of 0.1%.

A 1 mL aliquot of each sample was added to 20 mL headspace vials for analysis without dilution and pre-filtration. 50 µL PFBHA solution and 1 µL of 1% cyclohexanone in water were added, and each vial was immediately sealed with a Teflon-coated silicone septum. The sample was mixed, incubated at room temperature for 4 h, and heated at 60°C for 30 min in the headspace bath to reach gas-liquid equilibration. The headspace syringe was set at 100°C and the sample volume was 1 mL.

Determination was performed on an Agilent 7890B/7000D GC–MS/MS system, equipped with a split/splitless inlet, a PAL RSI 85 headspace tool and MSD Chem Station software. The column was a 60 (m)×0.320 (mm)×0.5 (µm) DB WAX UI (Agilent Technologies). Injection was in a split mode with a helium, 1.8 mL/min constant flow set at 35°C as a carrier gas. The injection temperature was set at 200°C and the split rate was 25:1. The temperature programme of the column was held at an initial temperature of 35°C for 5.66 min, then increased to 100°C at 8.8°C/min, held at 100°C for 1.7 min, then increased to 220°C at 13.3°C/min, held at 220°C for 3.39 min, and increased to 240°C at 22.1°C/min, held at 240°C for 3.43 min. The MSD transfer line was set at 240°C, the source temperature at 230°C (70 eV), quad temperature at 150°C, scan range 20–300 *m/z*. For quantification, the

TABLE 1 Selected simple chemical reactions for the analysis of preservative fluids. Methods were selected based on information obtained through source studies (Simmons, 2014; Thede, 1996).

Verified substance	Test	Reagents	Positive result	Negative result	Comment
Ethanol (the test gives a positive result for *primary and secondary alcohols) *at room temp. the result for 1st-degree alcohols will be negative	Lucas test	$ZnCl_2$ anhydrous in HCl (concentrated)	Turbidity of the solution (milky colour)	No noticeable changes at room temperature	It is recommended to use a high concentration of ethanol (75%) for purposes of increasing liquid volume, especially in exhibits where mould appears (Simmons, 2014)
Glycerine	Reaction with $Cu(OH)_2$ (unheated)	Freshly precipitated $Cu(OH)_2$	Sapphire solution without precipitate	No visible changes	Glycerine is a popular addition to conservative mixtures. It is believed to form a 'protective layer' on the tissues of the preparation, preventing them from drying out when the liquid is drained
Formaldehyde	Schiff test	Aqueous solution of fuchsine saturated with sulfur oxide (SO_2)	Purple colouration (trityl cation)	No change in colour or non-violet colouring of the solution	The presence of formaldehyde, as a constituent of museum fluids, has been confirmed in studies of the solutions of other Museums, including the Mütter Museum of the College of Physicians of Philadelphia (Simmons, 2014) The formalin concentration for specimens which yielded a result during the HCl titration averaged above 10%. Nominally, the formalin concentration of storage solutions is about 4% (Thede 1996)
Phenols or carboxylic acids	Reaction with $FeCl_3$	Diluted solution of $FeCl_3$	Green, blue, violet or purple colouration—phenol present	No change in colouring of the solution Red colouration—compounds able to enolise	
Acetaldehyde, ethanol	Iodoform test	Solution of iodine in potassium iodide	Yellowish iodoform needles are formed	No iodoform needles	May be formed as a result of oxidation

analyses were registered in Sim Mode and following masses were recorded: formaldehyde *O*-pentafluorophenylmethyl-oxime 181, cyclohexanone: 55, ethanol: 45, methanol: 31.

2.3.2 | Fourier transform infrared spectroscopy

The spectra measurements were recorded on a Thermo Scientific Nicolet iS50 FT-IR spectrometer (ATR method) with DTGS detector and KBr beam splitter. The ATR-FT IR spectra were detected in the range of 500–4000 cm^{-1}, at room temperature with a spectral resolution 4 cm^{-1} and number of scans was 32.

2.3.3 | Inductively coupled plasma optical emission spectroscopy

The third step of the chemical composition analyses consisted of performing ICP-OES to detect the presence of selected metals in the preservative fluids.

Three samples of fluids were tested for the presence of metals (one sample from the control group, and two samples from the research group [A19, A65, and N2]).

Mineralisation was performed on an Ertec Magnum II apparatus in HNO_3 solution (65% puriss p.a.), then transferred to 25 mL flasks. The resulting solution was clear and transparent. This

process was repeated four times. The elemental content (As, Zn, Al, Cu, Pb, Hg, Fe) of the samples was then measured using an iCAP 7400 DUO ICP emission spectrometer from Thermo Fisher Scientific.

2.4 | Microbiological studies

Samples were simultaneously cultured on bacterial and fungal media according to the following procedures: (i) specimen containers were transferred to the anatomical laboratory, and samples were removed while maintaining the greatest possible sterile environmental conditions (UV lamp, surface disinfection) (Figure 3c). The samples were taken using disposable bacteriological loops from all lesioned areas and by rubbing the sterile loop along the entire length of the preserved specimen. The material collected was then inoculated on culture media. Single-use sterile pipettes were used to collect fluid samples for culturing. (ii) Samples from the control group specimens were taken directly at the museum—the containers were disinfected externally, the seal removed, and the closures removed. Using sterile equipment, a catheter was inserted to aspirate fluid samples at various depths. The catheter was brushed against the skin of the specimen and the resulting 'mist' was aspirated. The fluid was inoculated on culture media.

Each sampling for microbiological tests was repeated.

When growth was observed on the microbiological media, identification was carried out according to standards for bacteria and fungi.

2.4.1 | Bacteria

In the first stage of the study, cultures were cultured on: (i) nutrient broth with glucose (BTL Poland) for aerobic bacteria culture, (ii) Schaedler broth with vit K3 (bioMerieux) for anaerobic bacteria, and universal thioglycolate medium (BTL Poland).

Media were incubated up to 10 days at 37°C in an incubator (Wamed KBC-65G) and checked every 24 h. When turbidity was observed the material was cultured on solid media Columbia agar +5% sheep blood (bioMerieux) and incubated at 37°C under aerobic conditions up to 48 h, and under anaerobic conditions for 48 h.

Cultured colonies were identified using MALDI-TOF MS (Bruker, MALDI Biotyper).

Identification results obtained by the MALDI-TOF MS method were analysed based on a score value. Those with low strength of evidence were rejected.

Bacteriological cultures were repeated—for this step, liquid media with thioglycolate (for aerobic and anaerobic bacteria) were selected. From positive cultures, material was cultured on selective constant media: (i) MacConkey Agar II (Becton Dickinson) and (ii) Columbia Agar with 5% Sheep Blood (Becton Dickinson).

In addition, gram-stained slides were made from scrapings of biofilm coatings from some specimens. The following reagents were used to make microbiological slides: Crystal violet (bioMerieux), Lugol's fluid (bioMerieux), Ethyl alcohol (bioMerieux), and Alkaline fuchsin (bioMerieux).

After staining, slides were evaluated under a Carl Zeiss light microscope at 1000× magnification and then imaged using a Cell*F (Olympus) system.

2.4.2 | Fungi

Test materials were cultured on solid media—Sabouraud medium with 4% chloramphenicol (BTL Poland). When fungal growth was suspected on liquid thioglycolate medium, additional isolations were made on solid Sabouraud medium. Cultures were incubated for up to 2 weeks at 28°C (Memmert Incubator IN110) and checked every 24 h. When fungal growth was observed, macroscopic evaluation of colonies was performed, taking into account their morphology (colour, reverse, obverse, colony structure). In addition, cultured colonies were screened on chromogenic media (Biomerieux chromID® Candida) and direct preparations were made in saline for microscopic evaluation. Taxonomy of fungi was established to genus. For mould fungi determination was based on the appearance of the fungus on the medium and the preparation of the culture; for yeast-like fungi, cultures were sieved onto chromogenic media and the genus was determined based on the appearance of the colonies obtained. MALDI-TOF MS method (Bruker, MALDI Biotyper) was used for species identification of yeast-like fungi colonies grown on Sabouraud medium. The results of the spectroscopic analyses were compared with those of the mycologist as described previously.

3 | RESULTS

3.1 | Source studies

So far, very little research has been conducted on preservative fluids in the context of human anatomy museums. It is likely that in the past, both common preservation methods—those published in the then-available literature, as well as new recipe variations specific to a particular scientific centre were used in practice (Simmons, 2014, pp. xi–xx, 66).

The history of the anatomical collection in Breslau dates back to 1745 when the Surgeons School was established. The collection was created by Professor Adolph Wilhelm Otto (1786–1845), and his successors, Prof. Hans Carl Barkow (1798–1873) and Prof. Carl Hasse (1841–1921). In one of the monographs on the angiological collection, Prof. Barkow mentioned that some wet preparations were stored on the museum grounds in zinc or lead containers (Barkow, 1869). The catalogues of the museum collections created by Professors themselves provide relevant data, listing 'foetal specimens immersed in the spirit of wine', but it is not known if the specimens were teratological.

The collected accounts of current and former employees of the Department of Anatomy show that the teratology specimens that remained after World War II were moved from the attic to a new location in the basement. Some were donated to other universities. Moreover, the preservative fluid in some of the containers was or supplemented using substances such as ethanol, glycerine, aqueous phenol solution, or methanol. However, there is no detailed information about the composition and volume of the topped-up preservatives.

This research has not yielded data that allows us to unequivocally determine how the historical anatomical specimens in the collection of the anatomical museum were conserved. In one article, which refers to local traditions, three formulas traditionally used to preserve anatomical preparations after 1945 are mentioned: (i) ethanol, formalin, and water; (ii) ethanol, glycerine, and water; (iii) and ethanol, chloroform, and acetic acid (Janiszewska et al., 2018). The method of sealing the containers is also discussed (Pluta et al., 2019).

3.2 | Chemical composition analyses

In the course of chemical composition analyses, formaldehyde, alcohol, and glycerin were detected. The oscillatory spectra, recorded with the ATR (attenuated total reflection) technique, are shown in Figures S1–S5. The results of the simple chemical reactions and the pH measurements, gas chromatography coupled to mass spectroscopy (GS/MS), and FTIR are presented in the Figure 4; Table S2. A comparison of the concentration of the assayed substances in individual specimens is shown in the graph (Figure 4).

3.2.1 | Results of reference methods

3.2.1.1 | Gas chromatography coupled to mass spectrometry

Chromatographic analysis revealed the presence of formaldehyde and the first-order alcohols methanol and ethanol. Ethyl alcohol was detected in all tested fluids. Methanol was found in samples A63 and A77, and trace amounts were also detected in the fluid of samples A19, A38 and A40 (Figure 4; Table S2).

3.2.1.2 | Fourier transform infrared spectroscopy

The analysis proves that the samples studied contain formaldehyde (all tested samples), and secondary alcohols. Some samples show the presence of glycerol (samples A93, C2, M30), which contains two primary and one secondary alcoholic groups. A detailed description of the interpretation of the results is provided in Figure S1.

3.2.1.3 | Inductively coupled plasma optical emission spectroscopy

During the determination of the average metal content of the selected fluid samples, the presence of zinc, iron, and copper was detected in samples A19 and N2, and aluminum content was additionally determined in sample A65 (Table S3). Mineralisation was carried out reproducibly.

3.2.2 | Results of simple chemical reaction and pH measurement

The results of the reaction can be seen in Figure S6.

$FeCl_3$: The colouration of samples by reactions with $FeCl_3$ may indicate the presence of phenol derivatives, for example, nitrophenols (yellow colour) (Pasto & Johnson, 1979) or other compounds capable of enolisation (red colour) (Broumand, 1952). However, performing

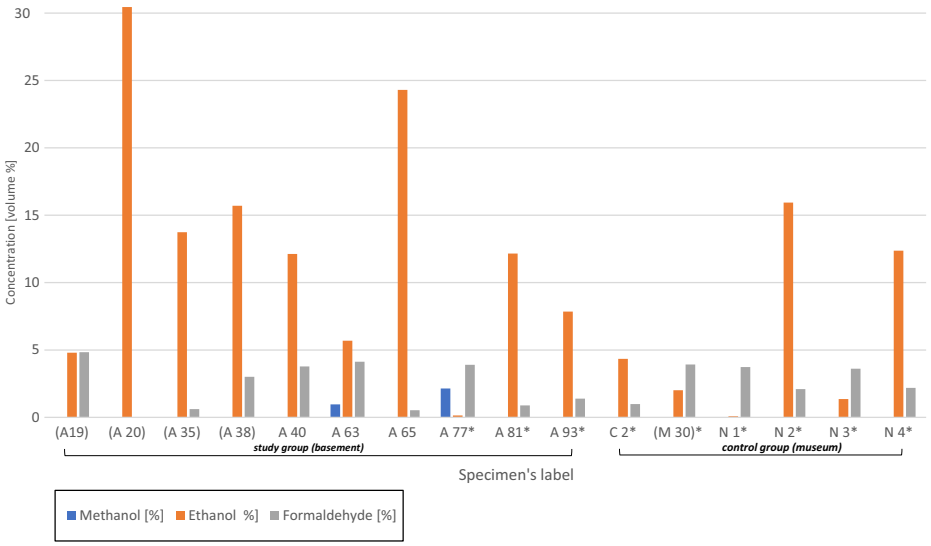

FIGURE 4 Concentration levels of methanol, ethanol and formaldehyde in assayed samples. Asterisk indicates containers with a reliable seal (specimens no. A77 through N4). Parentheses indicate specimens with visual contamination (either microbiological or physicochemical). Compare with results presented in Figure 6.

an analysis using FTIR spectra to confirm the presence of phenol derivatives in preservative liquids did not show signs of their presence. The failure to obtain a purple solution allows us to exclude the presence of phenol itself in the tested samples (Apostica et al., 2018; Moeller & Shellman, 1953).

Cu(OH)$_2$: Trials with Cu(OH)$_2$ provided results that were difficult to interpret. In some samples, there was a colour change to sapphire (suggesting the presence of glycerol) but without a concomitant dissolution of the precipitate (Hassan et al., 2001; Seibert & Long, 1925). An unambiguously positive result was obtained only for M30.

Lucas test: After performing the Lucas test, no significant changes in the colour or structure of the liquids were observed, indicating the absence of alcohols or the presence of only first-order alcohols in the tested solutions.

Iodoform test: Ambiguous results were obtained, difficult to both read and interpret. Results varied considerably between specific samples. In most of them, the reaction resulted in a precipitate whose appearance did not resemble iodoform needles. Thus, no clear confirmation or negation of the assumption that an aldehyde or a secondary alcohol was present in the museum liquids was obtained (Morisson & Boyd, 1985). Aldehyde could come, for example, from the oxidation of ethanol, which was most likely a component of the preservative, or it could be a contaminant from samples that were fixed in formaldehyde before transfer to another preservative.

Schiff test: Performing the test resulted in the appearance of a colour (Figure S6f) associated with the formation of a trityl cation and indicative of the presence of an aldehyde in all the tested fluids (Robins et al., 2011).

pH of the solution: most of the liquids were acidic (3.97 ± 0.01–6.20 ± 0.01), but the pH of a few samples approached neutral (6.86 ± 0.01 and 6.98 ± 0.02). However, due to the presence of organic solvents, caution should be exercised in interpreting the results of the pH measurement (Neumann & Crimmen, 2022, pp. 34–35).

3.3 | Microbiological studies

3.3.1 | Culture

Positive cultures were obtained from samples taken from anatomical specimens (Table S4). The microorganisms cultured belonged to yeast-like fungi—*Candida boidinii*, *Geotrichum silvicola* species (Figure 5), moulds—*Penicillium* and *Fusarium* genera, Gram-positive bacteria— *Bacillus cereus*, *Bacillus thuringiensis* and Gram-negative bacteria *Cupriavidus metallidurans* (Figure S7). In the second stage (repetition of culturing), all cultures were negative.

3.3.2 | Results from microscopic evaluation of gram-stained microbiological preparations

Selected anatomical specimens (no. 35, 40, 63, 65) were analysed microscopically (Figure S8). Gram-positive cocci, dispersed or in clusters, Gram-positive coryneform, Gram-positive bacilli, Gram-positive filamentous bacteria resembling actinomycetes, and Gram-negative granules arranged like diplococci, as well as blastospores of yeast-like fungi, were mostly observed. In the majority of samples various impurities, crystals, and epithelial fragments were observed.

4 | DISCUSSION

The historical nature of the research material determined the multifactorial character of the investigations carried out in this study. In this type of research, the first thing to remember is to respect the collection and keep it in the best possible condition, so that it can serve future generations. Ad hoc conservation work, without in-depth historical knowledge of the chemicals and technology used and the circumstances in which the collection was created, can result in damage to unique, historical anatomical specimens.

4.1 | Chemical composition analyses

The composition of the preservative fluid is an essential element in stabilising and fixing anatomical specimens (Waschke et al., 2019). There are many methods of preserving cadavers and anatomical specimens in modern anatomy. However, the gold standard is still formaldehyde (Balta et al., 2015). It has been proven that preservatives affect the appearance as well as the dimensions of the objects immersed in them (Steinke et al., 2012; Yeap et al., 2007). Therefore, it is very important when working with historical material to avoid changing preservation solutions. This fact was behind the decision to search for the historical composition of the preservative fluid used in the collection assessed in this study.

The heterogeneous results and difficulties in interpretation are due to both the diversity of technical analyses and complexity of the analysed material. The influence that the particular components of the solution had on each other over time is unknown. Certainly, the presence of turbidity, non-uniform solution colour, and the presence of precipitates additionally hindered the evaluation of the results. Furthermore, undesirable effects of the lack of conservation supervision in the study group cannot be excluded.

4.1.1 | Reaction with Cu(OH)$_2$ and iodoform test

The advantage of tests based on chemical reactions lies in their simplicity. The disadvantage is the way the results are interpreted, which is based on subjective visual evaluation.

In the Cu(OH)$_2$ cold test, the intriguing fact is that although aqua complexes causing the sapphire colour were detected, there was an additional precipitate in the liquids (Figure S6c). This phenomenon indicates a side reaction with another substance present in the fluid, as the product of the sample itself should only be a copper (II) coordination compound. Another explanation may be the failure to bind

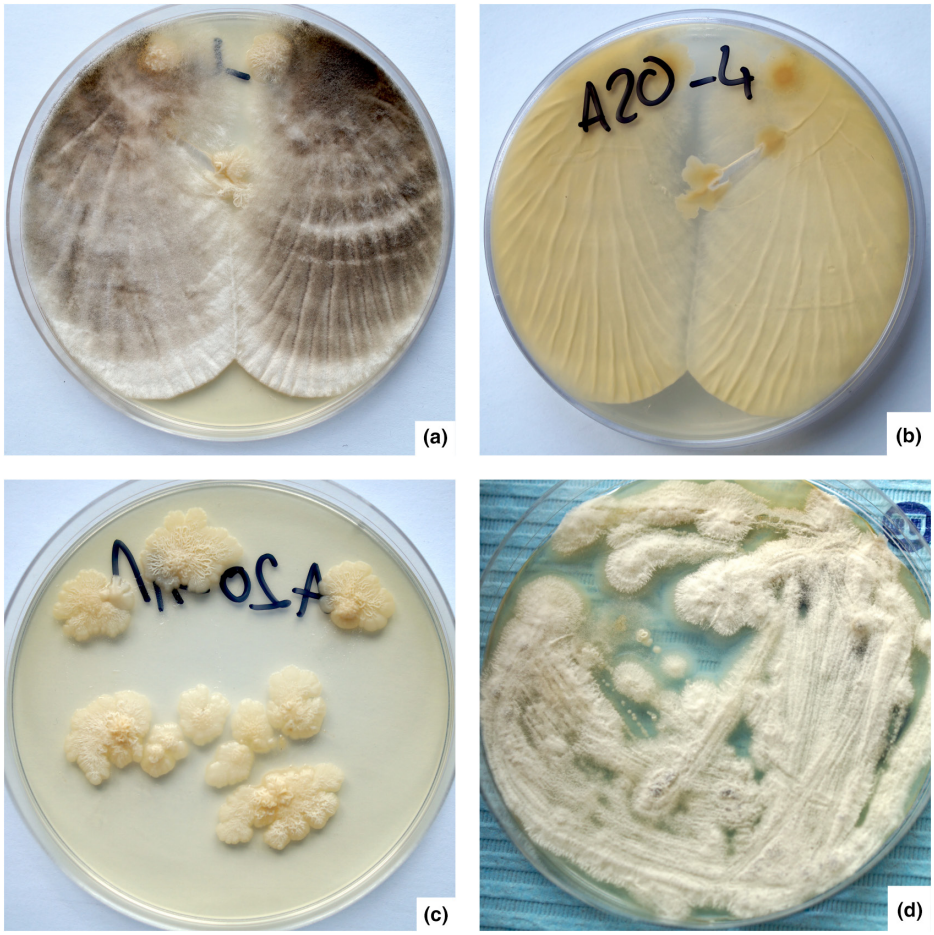

FIGURE 5 Fungi isolated from samples. (a, b). *Penicillium* sp. isolated from specimen A20 on Sabouraud's agar—obverse (a), reverse (b). (c, d) *Candida boidinii* on Sabouraud's agar—big cream colonies with rough growth (c) and *Geotrichum silvicola* on Sabouraud's agar showing off-white/cream colonies with matted appearance (d).

part of the reactant in the complex due to the difficulty of maintaining the stoichiometry of the reaction, resulting from the lack of knowledge of the composition of the preservative fluids. In multi-component samples, the interpretation is complicated by the lack of knowledge of the matrix, which consists of chemical compounds located next to the substance being determined.

The iodoform test provided equally unusual observations in which the flocs that formed did not resemble typical iodoform needles (Figure S6). Nagata and Nishiwaki described a situation in which yellow precipitates were obtained in an iodoform test due to the presence of ethyl acetate in the sample. The possibility of hydrolysis occurring leading to the formation of ethanol makes it difficult to interpret the result unambiguously. Ethanol that forms from ethyl ester, if it is used as a solvent, will also give a positive iodoform test result (Nagata & Nishiwaki, 2021). Because ester compounds and alcohols give a positive iodoform test result, it is impossible to accurately type the compound that is in the fluids based on the iodoform test result.

In the case of both tests, the results could be falsified by sediments present. Their influence on the reaction result could be minimised by prior filtration of the sample. However, this involves the risk that the concentration of the components of the mixture will decrease through absorption by the filter (simultaneous loss of sediment present in the liquid) or, in the case of the analysis of volatile substances, through evaporation. For this reason, the reaction with $Cu(OH)_2$ and the iodoform test is not suitable for testing turbid solutions and should not be recommended for the evaluation of museum conservation fluids.

4.1.2 | Reaction with $FeCl_3$ (phenols, acids)

This test was aimed at verifying the presence of phenol, which would account for the presence of a pungent, suffocating odour when some containers were opened.

Surprisingly the results obtained probably indicate the low sensitivity of this method. Analysis of the literature revealed that iron (III) chloride reacts not only with phenol but also with carboxylic acids. Hence, it is possible to obtain a false positive result. The presence of carboxylic acids in the samples may be indicated by the various non-specific colours obtained (Kapoor, 1988).

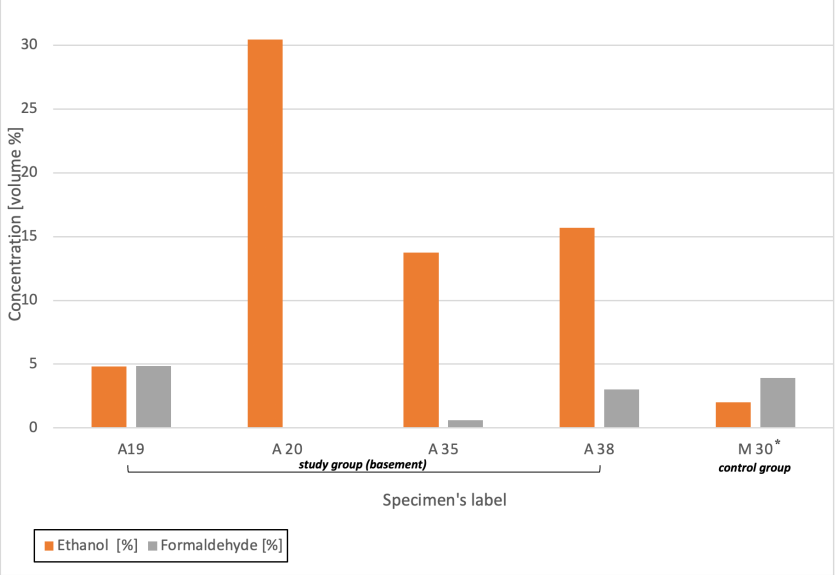

FIGURE 6 Specimens from which microorganisms were isolated and concentrations of preservative fluids. Asterisk indicate jars with a reliable seal.

Acetic acid was typified as a component of the mixtures, based on the assumption that: (i) acetic acid was a component of the recipe; (ii) it is a product of the oxidation of ethanol to aldehyde and acid that occurred due to the leakage of the jars; and (iii) it is a product of the hydrolysis of a weak acid salt. In some methods of conservation of museum preparations, acetates have been used (e.g., the Kaiserling method, the Judah method) (Simmons, 2014, pp. 289–295), the hydrolysis of which can provide weakly dissociating acetic acid to the system. However, the presence of these substances was not demonstrated with further analyses.

4.1.3 | pH measurement

The tested solutions gave curiously low pH results. The acidic pH (Figure 4; Table S2) is most probably caused by the presence of formaldehyde, which in an aqueous solution has a pH of 2.8–4.0 (Simmons, 2014).

The increased acidity of the solution may also be related to the reactivity of metal salts (e.g., Hg, Na, K) which, in order to increase the antiseptic potential, were used in old formulations (Parsons, 1831; Simmons, 2007, 2014; Swan, 1833). The presence of such substances has been demonstrated by research methods conducted on similar museum collections (Simmons et al., 2007; Thede, 1996).

The pH value of fluids is fully dependent on the proportions of the individual components present in the mixture. The impossibility of establishing a complete recipe prevents an unambiguous interpretation of the pH. Ethanol preservatives have slightly acidic to neutral pH (Simmons, 2014) which could, like the addition of methanol, influence the pH of the entire mixture. The exact pH of the alcohol depends on the concentration of the solution and measurement method (Deleebeeck et al., 2021). The pH value is probably somewhat disturbed due to the fact that the tested liquids consist mainly of organic solvents, not water (Neumann & Crimmen, 2022).

4.2 | Formaldehyde

Prior to this study, the presence of formaldehyde in museum fluids was typified and therefore an attempt was made to confirm its presence using a reagent of proven sensitivity that was previously used in museum practice, including to quantify the concentration of airborne formaldehyde in museum rooms (Gibson et al., 2008). A review of the specialist literature showed that the Schiff test is a recognised method for confirming the presence of aldehydes, used by organic chemists (Robins et al., 2011). This test has been used to evaluate museum fluids by some authors (Simmons, 2014, p. 181). The results obtained from the Schiff test clearly indicate the presence of an aldehyde in the preservative fluids.

The identity of formaldehyde, contained in the fluids tested, was confirmed by unambiguous infrared spectra, and its exact amounts were determined by gas chromatography.

The available literature indicates that infrared spectroscopy is one of the most useful methods for identifying chemical compounds (Sablier et al., 2020). The spectra show for example, stretching and deformation vibrations characteristic of specific chemical groups in a given range (Kazarian, 1758). Due to the absorption of infrared radiation, oscillations of molecules occur, such that the valence vibrations of groups of atoms are recorded in their characteristic areas (Thompson, 2018). It is a recognised pharmacopeial method for the identification of numerous chemicals.

Gas chromatography is also a recognised analytical method for the qualitative and quantitative determination of chemical compounds (Al-Rubaye et al., 2017). The separation of individual substances is possible due to their different physicochemical properties which results in discrepancies in migration rate through the chromatograph column, which was chosen for the fluids under study.

From the results obtained during the analysis (Figure 4; Table S2), it is evident that gas chromatography is the best method to accurately determine the presence and concentration of formaldehyde. The Schiff test and FTIR analysis are also suitable to determine the presence of formaldehyde.

The results obtained provide the basis for a broader discussion on the influence of formaldehyde on anatomical preparations.

Formaldehyde, as a fixative, causes tissue hardening in the amino acid crosslinking reaction. In addition, dehydration and darkening of tissues is observed (Simmons, 2014, p. 26).

The long-term effects of formaldehyde on tissues include the breakdown of adipose tissue and the precipitation of water-insoluble free fatty acids, which form flocculent deposits that float on the surface of the preservative fluid. At low temperatures (−5 to +5st) formaldehyde in aqueous solutions undergoes spontaneous polymerisation, the product of which partly converted to paraformaldehyde (Simmons, 2014, pp. 30, 44). This polymer precipitates out of solution causing it to turn cloudy, and settle on the bottom of the container as a white sediment, and can frequently coat specimens as well. Such changes were observed in both the test and control groups in n this study.

The exposure of the preparations to such low temperatures may have been due to the tragic effects of the war when the attic above the museum was bombed. Most of the roof of the building was destroyed, as well as the window frames.

Concentrated formic aldehyde, intended for the preservation of anatomical preparations, is buffered by the manufacturer. This has been the practice since at least the second half of the 20th century (Hayashi et al., 2016). During dilution, the protective action of the buffer is lost and the formic aldehyde becomes reactive. On contact with air, it forms formic acid, which lowers the pH of the solution and consequently decalcifies the tissues. Acetic acid and alcohol, used in the past as a fixatives, may have similar effects.

Considering the fact that formaldehyde has been available in Germany most probably since 1891 and the knowledge about its properties for fixing human tissues has been common since 1893 at the earliest (Musiał et al., 2016, pp. 27–28; Simmons, 2014), anatomical specimens created before this period could not have been fixed or preserved with it. Therefore, insofar as the preparations analysed were created before 1893 (Otto, 1816, p. 7, 1823), the preservative fluid must have subsequently been modified by the addition of formaldehyde, which was previously unavailable. However, even after this period it was possible to use only alcohol (preservation without fixation) (Simmons, 2014, p. 49), or only formaldehyde—especially for 'medium-size foetuses' (Schultz, 1924).

The rationale for the use of spirit for the preservation of the collection under study is supported by its use by German anatomists for scientific research or demonstrations, in which containers could not be permanently sealed and therefore required frequent refilling (Kozuschek, 2007, p. 110). For this reason, cheaper, more widely available, and less complicated to prepare preservative formulas (spirit solutions) were probably used (Brenner, 2014), so that the concentration could be monitored and the evaporating substance replenished as necessary. On the other hand, some of the museum preparations preserved after the war still intrigue us with the natural colour of the tissues and the perceptible, pleasant smell of the preserving liquids, so it is likely that for purposes of permanent exhibition, more sophisticated methods of fixing and preserving the preparations were used. For example, a preparation guide from 1831 lists fluid recipes based on, among other things, turpentine (Parsons, 1831). This has been used as a clearing agent for mercury-injected specimens from the 18th century (Brenner, 2014; Moore, 2006).

4.3 | Alcohols

The Lucas test is a method for the detection of alcohols and the determination of their order. It allows differentiation of the order of the alcohols, exploiting the differences in reactivity with halogens of primary, secondary and tertiary alcohols (Kjonaas & Riedford, 1991; Morisson & Boyd, 1985). The results obtained from the Lucas test indicate the absence of alcohols other than primary alcohols in the museum fluids studied. This is confirmed by gas chromatography coupled with mass spectrometry, which clearly indicates the content of ethanol, that is, primary alcohol, in the preservative fluids, while also indicating the presence of methanol in some of the containers.

The presence of glycerol, a secondary alcohol, was confirmed by FTIR spectra analysis and (probably) by $Cu(OH)_2$ assay. Glycerine is a common additive in preservative fluids, although its properties are debatable for this use (Simmons, 2014). In contrast to FTIR, the Lucas test did not show the presence of glycerol or other secondary or tertiary alcohols in the preservative fluids, which indicates its significantly lower sensitivity than spectra analysis.

In view of the overlapping results obtained for the primary alcohols, carried out independently of each other, the Lucas test can be considered reliable in this context, but perhaps not sensitive enough for the detection of alcohol in a mixture with formaldehyde.

Gas chromatography confirmed the presence of alcohol (ethanol) previously identified in the preservative fluids, but also showed small concentrations of methanol in some of the containers. The methanol content of the samples was confirmed by spectroscopy.

In the control group, the alcohol content of the preservative liquids was low—no more than a maximum of about 13%. suggesting the dominant contribution of formaldehyde (from 1 to almost 4%) as the preservative of these specimens, or perhaps the contribution of other, additional substances not detected in this study.

The yellow precipitates obtained in the iodoform sample may result from the reaction of esters or alcohols in the presence of

methanol as solvent, which would also indicate its presence (Nagata & Nishiwaki, 2021). It is possible that it was added as a stabiliser of formaldehyde. In the case of formaldehyde polymerisation in the preservative fluid, a small addition of methanol or NaOH allows this process to be reversed (Simmons, 2014, pp. 30, 44). The methanol may have been a contaminant of the industrial alcohol or have been used deliberately to denature ethyl alcohol, which was later applied to refill fluid level in the jars. It is also worth noting that the presence of alcohols (e.g., methanol or ethanol) in preservative liquids can cause shrinkage of tissues present (Simmons, 2014).

Unambiguously, the presence of alcohols can only be confirmed by reference methods such as applied here: infrared spectroscopy and gas chromatography. Cersoy and her team report that they could identify ethanol, methanol, and isopropanol alcohol using micro-Raman spectroscopy (Cersoy et al., 2020). Although they suggest or exclude the presence of given group of compounds, analytical reactions give results which are too imprecise to be used as a basis for conclusions.

4.3.1 | Microbiology and specimen environments

Microorganisms present in the environment have different adaptive capacities. Their growth depends, among other things, on the availability of oxygen, pH, nutrients, or humidity. Disinfectants or preservatives can reduce the viability of bacteria and fungi or eliminate them completely from the environment. In the course of this study, it has been shown that some microorganisms tolerate the presence of various compounds contained in preservative fluids and we can freely cultivate these microorganisms using classical microbiological methods. The study of chemical composition allowed us to determine which substances were present in the samples (Figure 6).

In the course of the study, several microorganisms were detected in the cultures from the anatomical preparations, which survived the rough living conditions and the presence of preservatives. It is worth looking at these microorganisms in terms of their ability to adapt to such difficult environmental conditions and the chemical composition of the preservative fluids, for which there is no documentation. Both simple and advanced techniques of chemical composition analysis made it possible to determine the composition of these fluids.

The pH value is one of the factors determining the presence of microorganisms in a given environment. It affects their metabolism and determines the chemical activity of protons involved in, among other things, redox reactions, co-formation of bacterial or fungal surface structures, activity of extracellular enzymes, or interaction with other organic structures (Jin & Kirk, 2018).

The pH also affects the antiseptic potential of formaldehyde. It has been shown that at a steady formaldehyde concentration, with decreasing pH, the susceptibility of some amino acids to formaldehyde fixation decreases, through inactivation of amino and hydroxyl groups (Eltoum et al., 2001). This may be one explanation for the resistance of microorganisms to formaldehyde. The progressive decrease in pH of the preservative fluids of the specimens could therefore be one of the causes of microbial colonisation (Ratzke & Gore, 2018; Tamayo-Arango & Garzón-Alzate, 2018).

The fungus *C. boidinii* has the ability to metabolise short-chain alcohols (including methanol and ethanol) to aldehydes. In addition, it can oxidise formaldehyde to formic acid through formaldehyde dehydrogenase (Sahm, 1977).

The ethanol oxidation capabilities of this yeast have been relatively poorly described. It is known that it assimilates ethanol faster than methanol. Using methanol as the only carbon source in the medium, growth of *C. boidinii* is observed (Sahm & Wagner, 1972).

The growth rate depends strictly on the inoculum of the microorganism. The larger the inoculum in the medium the better the growth (Pilát & Prokop, 1975). However, the ability of this fungus to assimilate and oxidise methanol is limited. The higher the methanol concentration, the more these abilities decrease (Pilát & Prokop, 1975).

Candida boidini tolerates a methanol concentration in the medium of up to 5%, at which level enzymatic activity and growth ceases (Sahm & Wagner, 1972).

The presence of formaldehyde or methyl formate in the medium affects the growth of *C. boidinii* in the presence of methanol and the activity of methanol-degrading enzymes. As a result of their presence, the biomass of the microorganism multiplied in the medium increases, but the methanol utilisation rate decreases. In the presence of formaldehyde, the activities of formaldehyde dehydrogenase and formate dehydrogenase increase significantly, while the activity of methanol oxidase decreases significantly. In the presence of methyl formate, the exact opposite happens—methanol oxidase activity increases and dehydrogenase activity decreases (Aggelis, 2000).

It has been shown that this fungus uses the metabolic products of some bacteria with which it has a symbiotic relationship, if it is not possible to incorporate the compounds available in the environment into its own biochemical pathways (Sahm & Wagner, 1972). The metabolism of *C. boidinii* may have had an effect on changes in the quantity and quality of preservative fluids.

Formaldehyde or formate is much more toxic to microorganisms than methanol, hence it is assumed that fungi will not grow in their presence. Even low concentrations of formaldehyde, on the order of 0.01%, may be too toxic, but Pilat et al. showed that in formaldehyde with the addition of a small amount of methanol (1%) it was possible to induce the growth of *C. boidinii*. This happened in a range of formaldehyde concentrations from 0.005% to 0.08%. At higher formaldehyde concentrations, inhibition of yeast growth occurred (Pilát & Prokop, 1975). The effect of ambient concentrations of ethanol or formaldehyde on the survival of this yeast is not known.

Candida boidinii in the presence of methanol as the only source of carbon and energy, induces in its cells the biogenesis of peroxisomes, which contain a large number of enzymes responsible for the transformation of various substances, including xenobiotics (van Dijk et al., 2000).

Another yeast-like fungus, *G. silvicola*, isolated from an anatomical sample, has the ability to metabolise, among other substances, glycerol, ethanol, and its ester with acetic acid (ethyl acetate) as carbon sources (Pimenta et al., 2005). *Geotrichum* spp. are yeast-like

fungi widely used in biotechnology, for example in cheese making, in the biodegradation of oil stains and decomposition of cellulose (Hyde et al., 2019). Zhu et al., (2017) showed significant degradation of higher alcohols, such as hexanol and isoamyl alcohol, at low pH by glutamate dehydrogenase produced by the species *G. candidum*.

Cupriavidus metallidurans, which was isolated from one of the samples, is an environmental bacterium that is able to withstand extreme environmental conditions and to metabolise toxic substances such as gold chloride. It has a wide range of adaptations, including the ability to survive in high concentrations of heavy metals, including Fe, Zn, Cu, Hg, which it reduces by incorporating them into metabolic pathways. It does not have the ability to absorb glucose from the environment, but can produce energy by metabolism of acetates, etc. It also produces the enzymes alcohol and aldehyde dehydrogenases and can degrade short fatty acids and aromatic compounds (Janssen et al., 2010; Lal et al., 2013).

Cupriavidus metallidurans, until recently mainly isolated from contaminated soils and sediments, was isolated for the first time in 2011 from clinical material from humans. The isolation was from blood of a patient with sepsis (Langevin et al., 2011; Vojtková & Janulková, 2012).

The detection of *Cupravidius* bacteria may indirectly demonstrate the presence of heavy metals in the preservative solution. There may be several reasons for their presence: (i) intentional components of preservative mixtures, (ii) ions released through contact of the liquid with metal elements of the specimen (such as tags (Barkow, 1869) or 'zinc containers', as suggested by one of the historical sources we found), (iii) ions released from the tissues when exposed to formaldehyde (Simmons, 2014, p. 33), or (iv) contact of the specimen with metal instruments during preparation (Slevin, 1927). Some of the specimens examined were not only fixed and preserved but also dissected (vide Figure 2f), so the latter situation is likely.

ICP analysis provided similar results for samples from the study group specimens A19 and A65. This is surprising, because A19 was a significantly heavier foetus than A65, additionally enriched with a well-preserved placenta. Therefore, it was expected that the preservative fluid of the larger specimen would contain higher concentrations of metal ions, particularly iron, derived from the degradation of erythrocyte haemoglobin contained in the richly vascularised placental structure. Perhaps these ions were utilised by microorganisms, and in this way, the amount of free metals in the fluid remained low.

A case has been described of preparations that have darkened. These studies showed the presence of elemental mercury, perhaps precipitated by the reduction of mercury salts by bacteria living in the preparation jar, or by spontaneous reactions of the components of the preservative liquid. It is difficult to determine the cause of the colonisation effect on the specimens because no bacteria were identified (Simmons et al., 2007, pp. 32–36). However, it is known that bacteria of the genus *Cupravidius* precipitate metals from the solution, which has been imaged microscopically and macroscopically (Reith et al., 2009).

Cupriavidus in this study was isolated from one sample of the three taken from preparation A19, thus the positive smear could have been obtained from only a few specific places on the specimen (the lesions were of yellow colour, around which dense mycelium-like lesions were observed). Due to the disinfection of the preparation, additional verification tests for the presence of bacteria—as carried out in other cases—were not possible.

The Gram-positive bacilli *B. cereus* and *B. thuringiensis* cultured from several specimens are likely to be contaminants introduced from the air into the preservative fluids. These are environmental microorganisms that survive very harsh conditions by transforming into spore-like structures. A spore has a thickened cell wall, discards water, does not metabolise, has no cellular processes, and can therefore remain in the environment for many years. Only appropriate conditions (e.g., a change in pH, nutrients present in the environment) allow it to transform into a vegetative cell and function normally (Cho & Chung, 2020).

Mould of the genera *Penicillium* and *Fusarium* were also isolated. Such microorganisms are present in the air in the form of spores and float freely, so they have the opportunity to contaminate all kinds of samples for microbiological tests. It is therefore difficult to assess the nature of their presence in samples. It may be that contamination with these spores simply occurred during the collection of material to test, resulting in a positive culture.

On the other hand, Moore mentions contamination of specimen jars by fungi of the genus *Penicillium* particularly when preservation fluids contained low concentrations of ethanol/formalin or were accidentally diluted with water (Moore, 1999, pp. 108–109). Similar circumstances in the past probably altered the fluids analysed in this study. It is important to control the fluid concentrations and integrity of the container seal, and perhaps add an antifungal substance such as thymol or menthol.

Microscopic observations revealed a much higher diversity of microorganisms in the samples compared to the results of microbiological cultures. Some of the microorganisms detected in this way, presumably environmental, may be species unknown to traditional bacteriological and mycological culture. The only way to be sure would be to identify the microorganisms using molecular systematic methods, but the utility of these methods is restricted due to their expense.

4.4 | Mutual chemical and microbiological influences: Conservation aspects

On the basis of the chemical composition analyses, it was shown that each of the tested fluids had a distinct, individual composition. This may suggest that the composition was dependent on (i) the original purpose for preserving a given specimen (e.g., as a museum specimen, for demonstration, for research), (ii) the date at which it was created, (iii) the state of the container seal, or (iv) modifications of the fluid (whether intended or spontaneous).

The low concentrations of volatile substances in the preservative fluids may have been due to evaporation, especially if the jars were poorly sealed. The container seals were probably improved by a layer of paraffin oil that coated the meniscus of the solutions (Pluta et al., 2019) (vide Table S1; Figures S1b, S5), but its effect on reducing evaporation or oxidation of the substances in the preservatives has not been described. On the other hand, even the sealed containers from the control group (A93, C2, N3, N4) did not have very high concentrations of volatile substances, so perhaps it was intentional not to use high concentrations, or these specimens are preserved by another 'dominant' substance, which was not detected.

Based on this study it may be concluded that the deterioration of specimen quality is the result of simultaneous and interdependent biochemical and physico-chemical reactions resulting from the overlapping influences of (i) the location of the object, (ii) the specimen storage environment, and (iii) the microorganisms to which the specimen was exposed.

The primary causes of the changes in the anatomical specimens were probably leaky containers and the place where the collection was stored (i.e., a dark, cool, damp cellar with limited ventilation). Evaporation of the components of the preservative mixtures and their oxidation by air reaching the surface of the fluid may have affected not only the volume and concentration of the fluids but also their antiseptic properties. The changes in environmental conditions, due to unsealing the containers, may have stimulated dormant microorganisms to germinate. In the present study, it was shown that in certain concentrations, substances traditionally used in preservation can become nutrients for microorganisms (e.g., ethanol for *C. boidini*, heavy metal ions for *C. metallidurans*) (Figure 7).

This research shows that conservation practice has a substantial impact on the state of specimen preservation. It is risky to make any changes to the structure of historic fluid-preserved preparations, so a complete replacement of the preservative fluid is inadvisable: instead, preservative fluids should be replaced slowly over time (Simmons, 2014, p. 45). The equilibrium between the fluid and the specimen is a factor sensitive to changes. The balance established by the reactions between the specimen tissue and preservation fluid can be easily upset by radical fluid changes, which may induce new physico-chemical reactions that will manifest themselves in a variety of ways that are difficult to predict.

An isolated environment for the specimen limits the influx of microorganisms from outside, helps maintain the antiseptic potential of the preservative solution, and reduces the reactivity of the preservatives. Analysing the results obtained in a more general way (Figures 4 and 6), it appears that the isolation factor plays the most important role in the long-term preservation of anatomical specimens. There was no correlation between the concentration of preservatives and contamination by microorganisms or the degree of specimen pollution.

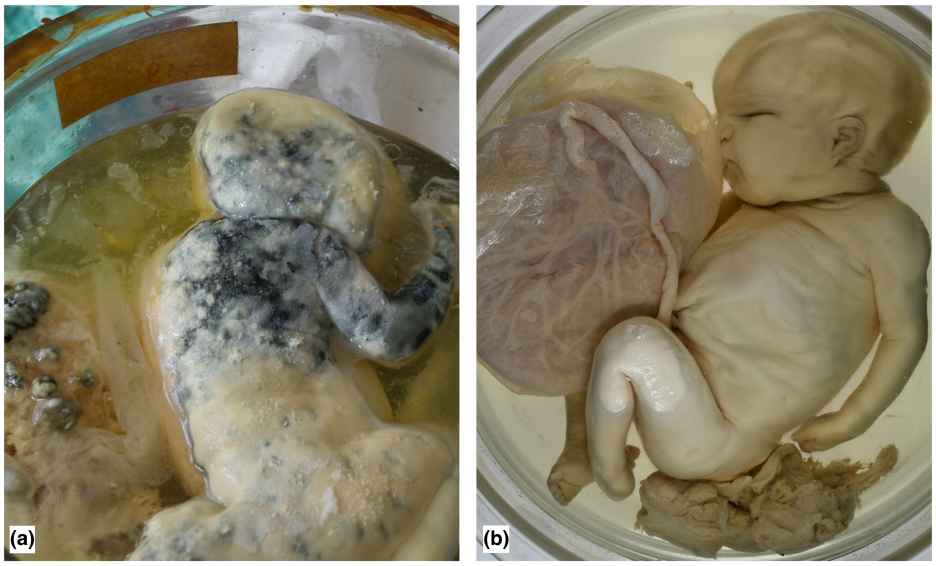

FIGURE 7 Specimen A19 described in the label as '*Fetus et placenta*'. (a) The centimetre-thick, malleable layer (*Candida* spp.) covering the specimen was removed, showing dark colonies (*Geotrichum* spp.) growing underneath. (b) The preparation was gently cleaned mechanically to remove the fungi, then transferred to a new conservation solution at low concentration to slowly 'adapt' the tissues to the new environment. However, intensive fungal growth was soon observed in the liquid. Based on the results of the chemical and microbiological analyses provided, it was decided to inject the preparation and submerge in formalin and, after time, gradually transfer it to increasingly higher concentrations of the alcohol solution. The glass lid was periodically removed and the specimen was monitored. After a year, we noticed the appearance of white deposits (probably *Candida* spp.) on the un-soaked parts of the preparation (e.g. part of placenta, knee). It is likely that the paraffin layer has created a barrier, making the methanol concentration in the empty (air) space of the jar too low or the specimen has been contaminated from the air during checking. Guidelines developed for the care of natural history collections may be useful in the conservation of such specimens (Moore, 1999; Neumann et al., 2022).

Specimen containers with good closures (in both the study and control groups) were in better condition than specimens that may have had their container seals compromised or that showed signs of sloppy interventions (e.g., traces of several layers of multiple seals, poor seal adhesion over the entire surface of the jar-lid junction). Furthermore, the latter containers showed higher concentrations of alcohol, probably the result of repeated refilling in the past.

In the group of specimens with good container seals, there were a few containers that displayed aesthetic defects, for example, white deposits on the bottom, crystalline precipitates on the walls of the jar, oxidised seals. This seems to confirm the thesis presented here that physicochemical factors also influence the visual aspects of a specimen.

It should be emphasised that working with contaminated preparations poses health risks. Microorganisms have a variety of pathogenic characteristics under different environmental conditions. Their virulence level for humans is unknown, but other research has described the isolation of pathogens that are hazardous to humans from fixed human anatomical specimens (Sperry & Sweeney, 1987; Weed & Baggenstoss, 1951). Knowledge of the possible risk factors is therefore extremely important. A variety of chemical substances, often toxic, were used for fixation and preservation, and these can evaporate through inadequate container closures into the museum space resulting in long-term health consequences. Persons undertaking the conservation of historic specimens are particularly vulnerable, as they are most exposed to these chemicals.

Little has been written about the role of microorganisms in the context of anatomical museums. Our study indicates a significant relationship between the microorganisms identified here, the composition of the preservative fluid, and the quality of preservation of anatomical preparations.

5 | CONCLUSIONS

The chemical studies demonstrated the presence of methanol, ethanol, formaldehyde, and glycerol in preservative fluids. The concentrations of these substances differed between the samples, and their determination required the use of a variety of methods to analyse the individual components of the preservative mixtures. The differences between the results obtained from the advanced modern analytical techniques and those obtained from the simple chemical reactions indicate that the latter are not sufficiently selective and are therefore inappropriate for assessing the composition of the preservative fluid independently.

This study shows that historical preservative fluids can be a habitat for many microorganisms. In microbiological tests, both bacteria and fungi were isolated from swabs taken from anatomical specimens. The bacterial flora was less numerous than the fungal flora. This research highlights the fact that microorganisms can be an additional factor in determining the compositions of preservative fluids and, at certain fluid concentrations, microorganisms show resistance to their effects.

The analysis of preservative fluids represents a significant challenge for contemporary anatomical museology. The unique and innovative research carried out by our team provides guidelines for contemporary anatomists and museum professionals. People involved in the protection of museum collections need reliable, fast and easy to implement methods for verifying the composition of the fluid protecting the specimens. However, due to compositional variability simple screening for chemical composition is an ineffective diagnostic tool for the museum anatomist. Instrumental analysis using sophisticated equipment is suitable, but expensive, and requires long lead times, while methods for measuring physical parameters are only suitable for single-component solutions. The chemical tests described in above can be useful for rapid assessment of the composition of simple mixtures, however, the development of guidelines for the analysis of contaminated historic preservative fluids that have been subjected to various modifications requires further research and systematic verification.

The development of such methods is much needed in the day-to-day work of the museum conservator, who often lacks specialist technical facilities.

AUTHOR CONTRIBUTIONS

Jurand Domański: concept/design, acquisition & analysis/interpretation of data (historical research, chemical reactions, microbiological analysis), co-author of the draft and final version of the work. **Adriana Janczura**: acquisition & analysis/interpretation of data (microbiological analysis), co-author of the draft and final version of the work. **Marta Wanat**: acquisition & analysis/interpretation of data (chemical reactions), co-author of the draft and final version of the work (chemical section). **Katarzyna Wiglusz**: acquisition & analysis/interpretation of data (Fourier transform infrared spectroscopy), co-author of the draft version of the work (chemical section). **Magdalena Grajzer**: acquisition & analysis/interpretation of data (gas chromatography coupled to mass spectrometry—GC–MS/MS), co-author of the draft version of the work (chemical section). **John E. Simmons**—critical revision. **Zygmunt Domagała**—co-author of draft version of the work (historical part). **Jacek C. Szepietowski**—critical revision, supervision.

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CONFLICT OF INTEREST STATEMENT

All authors declare that they have no conflicts of interest.

DATA AVAILABILITY STATEMENT

The authors confirm that the data supporting the findings of this study are available within the article and its supplementary materials.

ETHICS STATEMENT

Approval from the local bioethics committee was obtained for the purpose of the study.

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SUPPORTING INFORMATION

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5. ARTYKUŁ TRZECI

Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland.

Original paper

Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland

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Abstract

Introduction: Many historic dermatology departments keep and preserve valuable collections of dermatological moulages.
Aim: The aim of the present research was to find out whether the specimens collected in the Museum of the Department of Dermatology, Venereology, and Allergology of Wroclaw Medical University are colonized by microorganisms, and whether these organisms can pose a risk of damage to this heritage or a health risk to visitors.
Material and methods: In the study 32 historic moulages and their environment (museum) were subjected to microbiological evaluation.
Results: Swabs from moulages turned to be positive in 28% of cases. Micrococcus luteus was mainly isolated. The flora isolated from the air and the external surfaces of the museum display cases was much richer. Environmental bacteria and fungi were determined, as well as organisms probably associated with the hospital flora: *Pseudosomonas* spp., *Paebacillus* sp., Acinetobacter sp.
Conclusions: The close proximity of clinical wards probably influences the composition of the museum environment. The surprisingly low contamination of the moulages may be due to the antiseptic properties of the bee wax from which they were made. Conservation work on the moulages as well as people visiting the museum do not pose significant health risks. However, the small number of studies devoted to this topic limits the conclusions. Further research on medical collections is needed to provide 'evidence-based care' for this heritage.

Key words: moulages, microbiology, museum, dermatology.

Introduction

Wroclaw Medical University in Poland is the depositary of several collections of cultural heritage that survived the turbulent period of World War II and, in accordance with tradition, are exhibited in the museums of individual faculties and/or departments. These collections include wax models that were used as teaching tools for educating future physicians on a daily basis. After establishing a complex of clinics in the 19th century, the university became one of the most modern in Europe. Wax models had several advantages, the most important of which was their durability and legibility, allowing for quick identification. Probably for this reason, it was decided to make a costly investment related to their creation. From 1853, the university employed the outstanding artist Gustavo Zeiller, who made wax models for the Institute of Anatomy and Pathology in Wrocław (formerly: Breslau, Germany) [1]. It is possible that some of the wax models stored in the Anatomical Museum are his masterpieces.

A unique collection of wax models, the so-called dermatological moulages, was created for the purposes of the local dermatology clinic, founded by Heinrich Koebner in 1877. From 1882, this clinic was headed by Albert Neisser, who initiated the creation of the collection of moulages in 1890. At his request, the specimens were created in the building of the clinic by the employed artists: Paul Berliner from 1890 to 1897 and Alfons Kröner from 1897 to 1937. The most intensive production of moulages was 100 pieces per year, making the Breslau

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collection of 2696 objects one of the largest in Europe at the time. A collection of such a large size necessitated the preparation of a special space for exhibition, so in 1906 the former lecture hall was used as a storage place for the moulages, resulting in the creation of the Moulage Museum, which exists to this day [2].

There are 323 original moulages that have survived to the present day, of which 315 are made of wax and 8 of plaster. They are presented in 6 original wooden display cases with glass. The layout of the museum has remained unchanged since its creation (Figure 1).

The protection and maintenance of this unique cultural heritage is a challenge for contemporary times. However, there are not many reports that systematize how to take care of dermatological moulages, although they are part of the collections of many medical universities in Europe [3].

Taking the above into consideration, the current study was undertaken to perform microbiological examinations of dermatological wax models as well as of the neighbouring environment of the museum hall. We aimed to investigate whether the collection of historic dermatological moulages is colonized by specific microorganisms and whether these microorganisms pose a health risk to the environment, including museum visitors. To the best of our knowledge, such studies including dermatologic wax moulages have not been performed so far.

Material and methods

The specimens for microbiological examinations (bacterial and mycological) were swabs obtained from (i) the surfaces of wax moulages, (ii) internal and external surfaces of the cabinets in which they were stored, and (iii) the walls and floor of the museum hall. The air samples taken from the museum's exhibition room were examined. For technical reasons, showcase 6 and its collections were omitted from the study.

Collection and microbiological proceeding of surface samples

Five museum display cabinets (nos. 1–5) were included in the study. The samples were obtained from at least 5 moulages in each case. A total of 32 specimens, with a variety of textures and morphologies, were selected. In display case no. 1, specimens hanging at each height of the display case were selected. In showcases nos. 2–5, moulages were selected that were hung at different heights of the showcase, on either side of perforated boards.

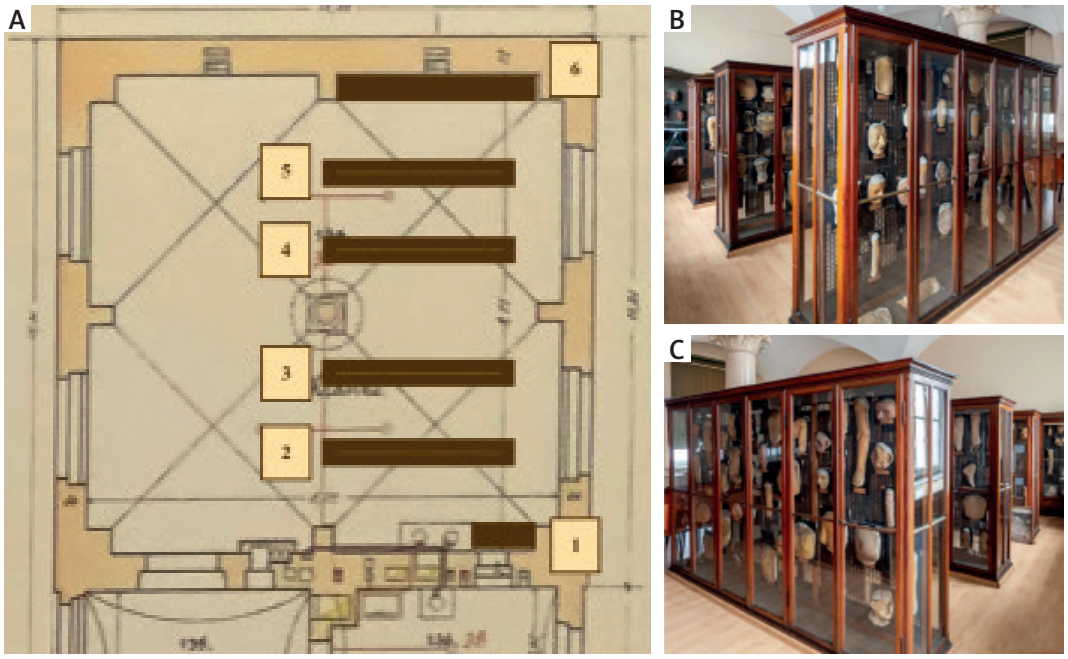

Figure 1. A – The Moulage Museum is a 9.7 × 9.7 m square with a barrel-vaulted ceiling supported by a 4.88 m high column in the centre of the room. The collections hang on perforated metal panels in 6 wooden display cases with glass. Showcases nos. 1 and 6 are set up against the walls to the right and left: showcase no. 1, with plaster moulages, measuring (width × height × depth) – 1.10 × 2.10 × 0.32 m; showcase 6, facing the lecture hall – 3.70 × 2.00 × 0.32 m (omitted from the study). The centre of the museum is occupied by four display cases (no. 2–5) measuring 3.00 × 1.92 × 0.6 m. **B** – View of showcases 1 to 4 (**C**) view of showcases 3 to 6

A sterile swab (Equimed, Poland) moistened with sterile 0.9% sodium chloride solution was used to wipe in 3 different places the waxy surface of a single moulage. Swabs were also taken from the surfaces in the museum space: the upper outer surface of the display cases nos. 2–5 (3 swabs from 3 different display cases), the floor (3 swabs), and the walls (3 swabs taken from a height of 2 m). In addition, material was collected from the top surface of the cabinets nos. 1–5 using Rodac contact plates with Saboraud Dextrose Lab agar (BioMaxima). The imprint plates were opened and the agar side was applied to the top outer surface of each of the 5 cabinets for 10 seconds. Material from one of the impression plates was transferred to Columbia agar with 5% sheep blood (Becton Dickinson) to assess bacterial growth. All samples were transported to a laboratory within an hour and immediately seeded on microbiological media. From each swab, the material was successively inoculated onto the entire surface of the plate with Columbia agar supplemented with 5% sheep blood (Becton Dickinson) for bacteria and Sabouraud Dextrose Agar supplemented with chloramphenicol (Biomaxima, Lublin, Poland) for fungi. The plates were incubated either at 37°C for up to 3 days (bacteria) or at 28°C for up to 14 days (fungi). Every 24–48 h, the cultures were reviewed and the cultured colonies were identified in accordance with standard microbiological procedures (evaluation of colony morphology and microscopic examination – Gram staining for bacteria and lactophenol slides for fungi). For bacteria species identification was obtained using mass spectrometry (MALDI TOFF MS) and a Bruker MALDI Biotyper IVD instrument (Bruker).

Collection and microbiological proceeding of the air samples

Air was tested using a MicroBio MB1 (220) Bioaerosol Sampler (Cantium Scientific, UK) according to the manufacturer's instructions. Sampling was carried out from 5 different parts of the museum. At each site, air samples were taken at a hight of 1.5 m sequentially onto an open plate with either Saboraud Dextrose Agar supplemented with chloramphenicol (BioMaxima, Lublin, Poland) for fungi (500 l of air per plate) or Columbia agar with 5% sheep blood (Becton Dickinson) for bacteria (300 l of air per plate), with a flow rate of 100 l/min. Incubation of plates and identification of cultured microorganisms was performed according to the standards described above. The cultures were observed every 24–48 h and the number of grown colonies was counted (each time, the fungal growth sites on the plate were marked to avoid counting secondary colonies). The number of fungi and bacteria in the air was calculated using MicroBio counts. The following formula was used for the calculations:

$CFU/m^3 = 1000 \times (n_c/V_s)$,

where n_c is the corrected number of colonies on the plate, and V_s is the volume of the tested air in litres.

Results

Moulages

Positive moulage swab cultures were obtained from 9/32 (28%) of the tested samples. Microbial growth each time was sparse, from 8 samples grown 1–2 bacterial colonies and from a single mould colony (Table 1). All other

Table 1. Results of the analysis of microbial contamination of the dermatological moulages and internal surfaces of the museum cabinets

Cabinet number	Moulages		Internal surfaces of cabinets	
	Number of positive samples/number of samples tested	Identified microorganisms[1]	Number of positive samples/number of samples tested	Identified microorganisms[1]
1	3/6	2× *Micrococcus luteus* 1× unidentified fungi (hialohyphomycetes)	1/1	1× *Rhodotorula* sp.
2	3/7	2× *Micrococcus luteus* 1× unidentified Gram-positive bacilli	4/5	2× *Micrococcus luteus* 1× unidentified Gram-positive cocci (species II) 1× unidentified Gram-positive cocci (species III)
3	1/7	1× *Bacillus pumilus*	2/2	1× *Penicillium* sp. 1× *Micrococcus luteus*
4	1/5	1× *Bacillus pumilus*	0/1	–
5	1/7	1× unidentified Gram-positive bacilli	0/1	–
Total (Nos. 1–5)	9/32 (28%)	–	7/10 (70%)	–

[1]*The value by the organism's name indicates the quantity of positive samples for it*

cultures were negative. The predominant microorganism identified in the cultures was *Micrococcus luteus*.

Interior surfaces of the display cabinets

Microbial growth was obtained from 3/5 cabinets; positive results were obtained from more than half of the swabs taken (70%) (Table 1).

External surfaces of the cabinets

As a result of the examination of the swabs, bacterial growth was obtained from 2/3 and fungal from 3/3 display cases. From each positive sample 2–4 bacterial and 5–10 fungal colonies were obtained. Cultures of samples taken with imprint methods revealed abundant growth of fungi from all 5 showcases tested (at least 50 fungal colonies per 25 cm^3 surface) (Table 2 and Figure 2). In the culture obtained from the transfer of material to the microbiological medium the growth of numerous bacteria was achieved (Table 2).

Floor and walls of the museum

Bacterial and fungal growth was obtained in cultures from the floor swabs (Table 3 and Figure 3). No bacterial and fungal growth was obtained in culture from swabs from the walls.

Museum air

As a result of the analysis, both fungi and bacteria were isolated (Table 4). The mean number of bacteria in a cubic metre of air was 161 CFU/m^3, and the number of fungi was 38 CFU/m^3. The quantitative proportions of microorganisms in the air are shown in Figure 2.

Discussion

The results obtained allow us to state that the space of the cabinets and the moulages protected in them are contaminated to a minor degree by several microorganisms. Probably the tight fit of the historic cabinet doors effectively isolates their interior environment.

It is puzzling why the studied elements were colonized mainly by one and the same species of bacteria, *Micrococcus luteus*. According to Young *et al.* [4], this microorganism is adapted to a limited ecological niche of mammalian skin, while its presence in other ecological niches (water and soil) may be the result of contamination of the epidermis. Contamination of the specimens could have occurred, for example, by touching the moulages with bare hands, but the absence of other identified representatives of the skin flora on these specimens indicates a higher probability of contamination from the ambient air. Obtaining a small number of positive cultures is intriguing. Perhaps the materials from which the castings were made contain substances that are toxic to microorganisms [5] or are colonized by microorganisms that cannot be identified by traditional culture methods.

The unique abilities of microorganisms enable them to survive in environments inaccessible to other living organisms [6]. For example, *Bacillus pumilis*, isolated from one of the moulages, is characterized by high resistance to UV radiation and H_2O_2 disinfecting properties [7]. It tolerates the lack of assimilation of nutrients in the environment of drying irradiation [8]. Meanwhile, *M. luteus* managed to grow from material preserved in amber from 120 million years ago. It is suggested that bacterial growth must have taken place while the resin was still liquid and therefore nutrients were available. Studying

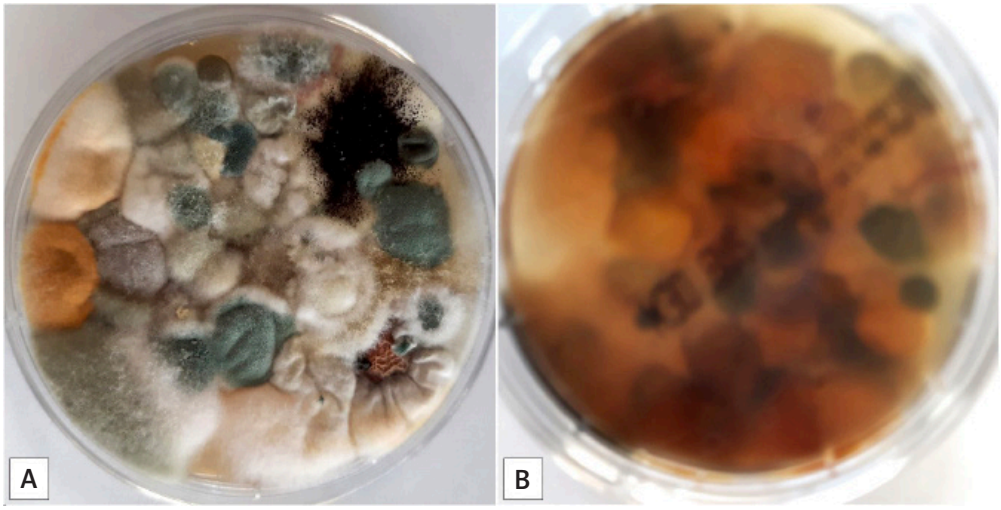

Figure 2. Imprint plate culture from the exterior surface (top) of the museum display cases: **A** – obverse, **B** – reverse

Table 2. Results of the analysis of microbial contamination of the upper surfaces of museal cabinets. The results of cultures and direct impressions were included

Cabinet number	Microorganism cultured from	
	Swab sample	**Imprint plate**
1	Swabs were not taken	*Alternaria* spp.
		Cladosporium
		Fusarium spp.
		Mucor spp.
		Penicillium spp.
		Yeast
2	*Acremonium* spp.	*Mucor* spp.
	Alternaria spp.	Other fungi[1]
	Aspergillus spp.	
	Fusarium spp.	
	Pheohyphomycetes	
	Micrococcus luteus	*Micrococcus luteus* (strain 1)
		Micrococcus luteus (strain 2)
		Paenibacillus lautus
		Pseudomonas plecoglossicida
		Solibacillus silvestris
		Unidentified Gram-positive bacilli (species III)
		Unidentified Gram-positive cocci (species III)
		Unidentified Gram-positive polymorphic bacteria (species I)
		Unidentified Gram-positive polymorphic bacteria (species II)
3	*Cladosporium* spp.	*Mucor* spp.
	Fusarium spp.	Other fungi[1]
	Penicilium spp.	
	Pheohyphomycetes	
	No bacterial growth	
4	*Penicillium* spp.	*Aspergillus niger*
	Aspergillus fumigatus	*Alternaria* spp.
	Chrysonila spp.	*Cladosporium* spp.
	Cladosporium spp.	*Fusarium* spp.
	Fusarium sp.	*Penicilium* spp.
	Hyalohyphomycetes	
	Micrococcus luteus	
	Paenibacillus pabuli	
	Unidentified Gram-positive bacilli (species II)	
	Unidentified Gram-positive cocci (species II)	
5	Swabs were not taken	*Aspergillus niger*
		Alternaria spp.
		Cladosporium spp.
		Fusarium spp.
		Penicilium spp.

[1]*Impossible to isolate and identify due to massive growth of Mucor spp.*

Table 3. Results of the analysis of microbial contamination of museal floor

Sample	Cultured microorganism
A	No fungal growth
	Acinetobacter ursingii
	Unidentified Gram-positive bacilli (species I)
	Unidentified Gram-positive bacilli (species II)
	Unidentified Gram-positive cocci
	Unidentified Gram-positive rod (species I)
	Unidentified Gram-positive rod (species II)
B	*Cladosporium* spp.
	Fusarium spp.
	Bacillus pumilus
	Kocuria rhizophila
	Micrococcus luteus (strain 1)
	Micrococcus luteus (strain 2)
	Staphylococcus hominis (strain 1)
	Staphylococcus hominis (strain 2)
	Unidentified Gram-positive bacilli (species II)
	Unidentified Gram-positive cocci (species V)
C	*Cladosporium* spp.
	Uidentified Gram-positive bacilli
	Micrococcus luteus
	Staphylococcus capitis

the bacteria's genome allowed us to understand some of the enzymes it produces and, on this basis, deduce the metabolic pathways available to it. For example, thanks to succinate dehydrogenase, an isolated strain was able to biotransform succinic acid taken from the environment, one of the components of amber. It also assimilated other resin components such as terpenes [6]. One wonders whether the materials used to make the wax casts contained substances particularly favoured by this species, and what factors prevented it from colonizing the rest of the collection.

The technique of making moulages was a secret. An imprint (negative) was made of the patient's skin using modelling plaster. A mixture of bee wax and various other organic and inorganic substances was poured into the mould thus prepared. The hardened cast was then coloured and "characterized" [9]. Siemiątkowski [10] emphasizes that the evidence of the use of different recipes of wax masses explains the variable durability of historic specimens observed today. In addition, he uses turpentine-based dyes in his conservation work, which may suggest that this natural solvent was used in the past to colour prepared moulds. Moreover, turpentine, along with alcohol, is an important solvent of bee wax [5], which may have been used to prepare the raw material.

It can therefore be suspected that the terpenes (a component of turpentine) contained in the moulages could be a potential food source for bacteria such as *M. luteus*, but the aseptic properties of bee wax and heavy metals contained in the pigments with which the wax was coloured prevented the survival of microorganisms. It is likely that the positive cultures came from specimens that were particularly contaminated with dust.

Studies of the museum environment showed a greater biodiversity of microorganisms than results from inside the display cases. Numerous florae were isolated in air samples and from swabs and impressions from the upper surfaces of the cabinets. Environmental bacteria and fungi, microorganisms forming the human skin flora (*M. luteus, S. hominis, S. capitis*), and pathogenic microorganisms were determined.

The identified bacteria of the genus *Paebacillus* sp. play an ambiguous role in the clinical context. They are likely to contaminate laboratory samples; however, some may also represent the true pathogens and cause nosocomial infections [11]. Similarly identified *Acinetobacter ursinigii* colonizes the hospital environment [12] and may be a casuistic cause of bacteraemia, even in immunocompetent patients [13]. *Pseudomonas* spp. bacteria are sometimes opportunistic human florae, and in conditions of immunosuppression or damage to natural barriers, it can be the aetiology of infections [14].

The air of the building of the local Department of Dermatology in Wroclaw, Poland has already been analysed for fungal spore contamination in a previous study. Our results indicate that the fungal diversity in the museum environment is very similar to that in other areas of the building. We isolated fungi from the genera *Acremonium* sp., *Penicillium* spp., *Fusarium* sp., *Aspergillus* spp., *Mucor* sp., *Rhodotorula* sp., similar to those documented by Łukaszuk *et al.* [15]. However, a comparison of the averaged quantitative data from both studies suggests that the museum air is less polluted than other areas of the same building (Table 5).

Some species of filamentous fungi have a negative impact on human health. Through colonization, they can cause superficial skin infections as well as systemic infections, especially in immunocompromised individuals. In addition, some mycotoxins released into the air or food are harmful to humans. They have various health effects: they are toxic to internal organs, carcinogenic, teratogenic, immunosuppressive, or endocrine disruptors. The occurrence of certain characteristic symptoms caused by exposure to mycotoxins is called mycotoxicosis [16]. *Aspergillus fumigatus* identified here is a source of mycotoxins such as fumagillin, gliotoxin, verruculogen, viriditoxin, and *A. niger*: malformin, oxalic acid, and och-

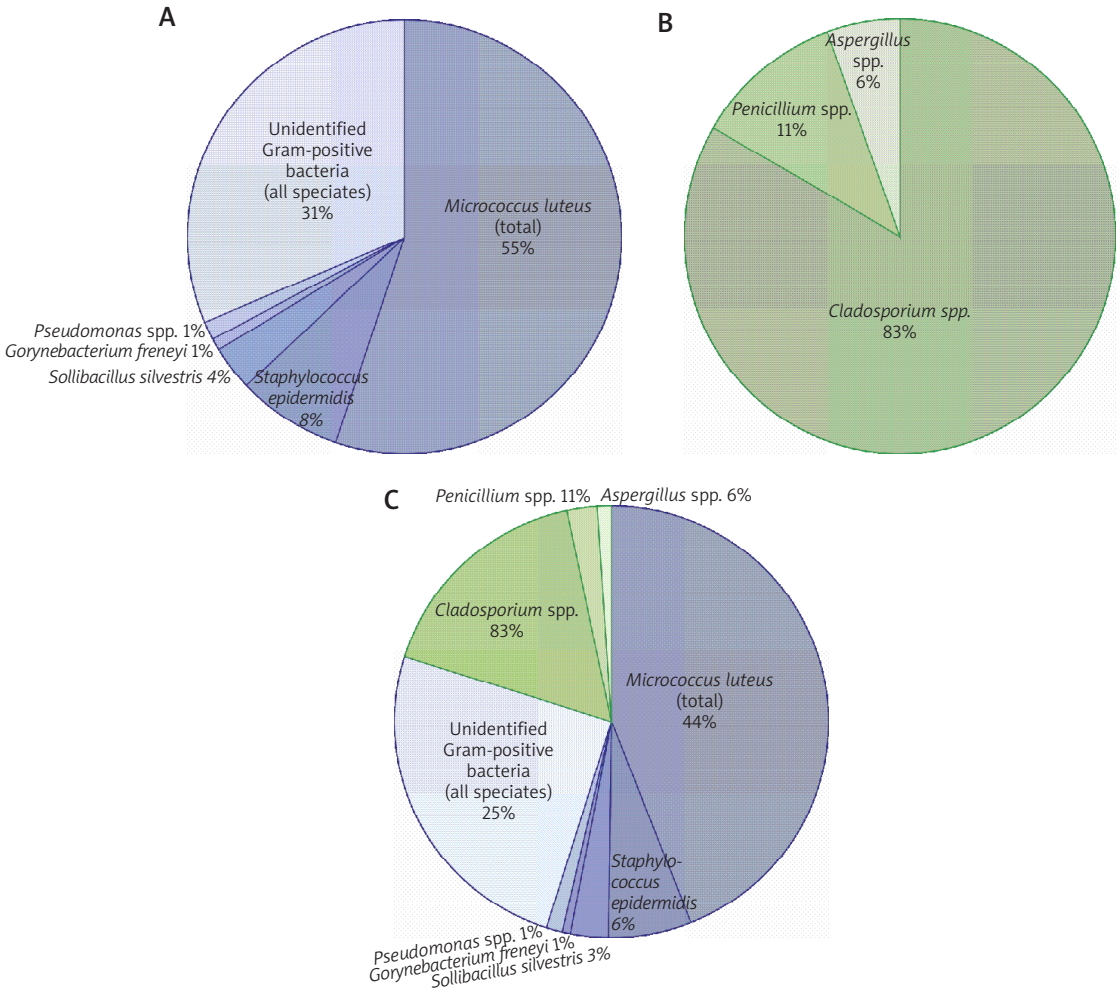

Figure 3. Contribution of particular microorganisms in contamination of museal air in calculation in CFU/m^3. Percentage distribution of bacteria (**A**) and fungi (**B**). **C** – percentage distribution of both fungi and bacteria

ratoxin A. Mycotoxins are released by fungi in response to specific environmental conditions such as temperature and humidity [17]. Therefore, simply identifying a fungus does not equate to exposure to its mycotoxins; for this to happen, the niche inhabited by the fungus must meet certain conditions and (probably) the amount of toxin released into the space must reach the appropriate concentration. The presented comparison (Table 5) shows that the spores of some fungi polluting the air in the museum *(Penicilium* sp. and *Aspergillus* spp.) were probably transferred from other rooms of the building.

Among the airborne fungi identified here, *Cladosporium* sp. was the most prevalent. This fungus is widely distributed on the earth. Its growth medium is paper and wood-based materials, so it commonly inhabits libraries and museums [18, 19]. Its spores contain numerous proteins that irritate the respiratory tract, causing symptoms of inhalant allergies (allergic rhinitis, sinusitis, conjunctivitis, mould asthma, allergic alveolitis) [20]. *Cladosporium* spp. are one of the common causes of allergy in museum employees who are constantly exposed to air contaminated with fungal spores [18, 19]. The occurrence of allergic reactions depends on the concentration of spores in the air and the individual sensitivity of the exposed organism. Different threshold concentrations of airborne spores causing allergic symptoms are proposed in the literature [21]. Studies on the Polish population have shown that a concentration of 2800 *Cladosporium* sp. spores in a cubic metre of air is necessary to induce allergic symptoms in predisposed individuals, and a value of 5000 in

Table 4. Results of microbial air pollution analysis

Microorganisms	Number of colonies						Number of CFU/m^3
	Plate 1 (window 1)	Plate 2 (window 2)	Plate 3 (window 3)	Plate 4 (door)	Plate 5 (table)	Mean	
Cladosporium spp.	18	13	10	20	14	15	30
Aspergillus spp.	2	0	1	1	0	1	2
Other fungi	0	2 *Naganishia albida*[3] (1 colony) (dematiaceous fungi)[4] (1 colony)	0	0	0	0	1
Penicillium spp.	3	2	1	2	2	2	4
Micrococcus luteus (total)[1]	28	28	20	25	17	24	79
Corynebacterium freneyi	0	1	0	0	0	0	1
Pseudomonas spp.	0	1 *Pseudomonas oryzihabitans*	1 *Pseudomonas oryzihabitans*	1 *Pseudomonas plecoglossicida*	0	1	2
Sollibacillus silvestris	4	3	0	0	0	1	5
Staphylococcus epidermidis	7	2	2	0	6	3	11
Unidentified Gram-positive polymorphic bacteria	3	3	6	3	0	3	10
Unidentified Gram-positive bacilli (total)[2]	5 (II)	9 (V)	8 (II)	10 (III)	12 (II)	9	29
Unidentified Gram-positive cocci	2	1	1	5	0	2	6

[1]Several strains of M. luteus were differentiated according to growth morphology.
[2]The number of distinguished species is indicated in brackets.
[3]No exact identification.
[4]Identification obtained from Phoenix Yeast ID-BD metabolic test (Becton Dickinson).

Table 5. Comparison of the microbiological contamination of the museum air with the data obtained by Łukaszuk and Her team

Fungi	[1]Mean number of colonies in museum air [volume of analysed air – 500l]	[1]Mean number of colonies from other rooms in Dermatology Clinic verified by Lukaszuk [volume of analysed air – 100 l]
Cladosporium spp.	15	–
Penicilium spp.	2	36
Aspergillus spp.	1	26
Naganishia albida	1	–
Feohyfomycetes	1	–

[1]Average sum of colonies obtained from all measurement trials divided by their number: (i) museum – 5 measurements from different locations in the museum, (ii) clinic – 5 measurements from different rooms.

all tested subjects [22]. Thus, the number of spores determined in the museum air was not sufficient to cause disease symptoms.

Conclusions

Based on our results, one may hypothesize that the materials from which the moulages are made may prevent the development of microorganisms on their surfaces, and the few isolated microorganisms probably came from dirt (dust). However, when working with these exhibits, we recommend the use of gloves to avoid contamination of the specimens with skin flora, as well as to protect them from the destructive effects of sweat and heat, which can desulpastize the natural wax and damage the object. The main microorganisms isolated from the museum environment were bacteria constituting the natural flora of the skin (of which *M. luteus* was the most numerous) and fungi of the genus *Cladosporium* sp. Compared to other rooms in the building, the air in the museum was slightly polluted with fungal spores. Working with moulages or visiting the museum do not pose a significant health risk, but individual findings of *Aspergillus* spp. fungi in the air (probably transferred from other rooms of the building) require further scrutiny. Air analysis can be used as a monitoring tool. The probable sudden appearance of new species or the observation of changes in the proportions of microorganisms in the air can provide valuable information about the condition of museum objects and health risk factors. Very few studies are devoted to the microbiology of medical school museums. Their location near medical facilities makes their biodiversity unique. Similarly, only a few studies dedicated to the protection of dermatological moulages are available [2, 10]. Articles on the conservation of artwork could be helpful in the care of these collections [23] as well as other rare studies performed, for example, on wax anatomical models [24]. This gap in the literature suggests that heritage medical collections need more attention from the scientific community, particularly with regard to designing solutions for their protection and a better understanding of their properties, especially in an interdisciplinary context [25, 26].

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Conflicts of interest

The authors declare no conflict of interest.

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26. Domański J, Domagala Z, Simmons JE, Wanat M. Terra incognita in anatomical museology – a literature review from the perspective of evidence-based care. Ann Anat 2023; 245: 152013. DOI: 10.1016/j.aanat.2022.152013.

6. STRESZCZENIE W JĘZYKU POLSKIM

Rozprawa doktorska powstała w oparciu o montematyczny cykl trzech artykułów opublikowanych w międzynarodowych czasopismach naukowych indeksowanych w bazie PubMed i uwzględnionych na liście Journal Citation Reports oraz znajdujących się w wykazie czasopism naukowych Ministerstwa. Artykuły zostały opublikowane w recenzowanych czasopismach o łącznym współczynniku wpływu 7,561 i 320 punktów ministerialnych. Badania sfinansowano grantem FAST (Funduszu Aktywności Studenckich - edycja II) pt. „Muzeum Anatomiczne Uniwersytetu Medycznego we Wrocławiu - historia, nauka, sztuka" o numerze w ewidencji wewnętrznej Uczelni: GMIN.A351.20.006 oraz subwencją Ministra Edukacji i Nauki na zadanie konkursowe pt. „Ocena mikroorganzimów izolowanych ze zbiorów anatomicznych pod kątem zagrożenia zdrowotnego i oporności na substancje dezynfekcyjne", identyfikowanym w systemie SIMPLE: SUBK.A351.23.020.

Pierwszym artykułem spośród cyklu jest praca obejmująca przegląd jak i metaanalizę literatury poświęconej badaniom muzeów i zbiorów muzealnym anatomii człowieka. W pracy przedstawiono stan wiedzy w tym temacie oraz występujące domeny zainteresowań badawczych. Udowodniono zaskakujący brak badań poświęconych konserwacji zbiorów muzealnych jak i brak badań obiektów muzealnych (tzn. badań podmiotowych). W tematyce tej nie odnaleziono satysfakcjonujących badań projektowanych w koncepcji interdyscyplinarnej.

W kolejnej pracy, poświęconej analizie płynów konserwujących zabytkowe preparaty anatomiczne, zaprezentowano wyniki przeprowadzonych badań (badania historyczne, chemiczne i mikrobiologiczne) poświęconych płynom konserwującym zabytkową kolekcje anatomiczną. W ich efekcie ustalono istotne fakty historyczne dotyczące badanej kolekcji, wytypowano i zweryfikowano metody badawcze, oznaczono niektóre substancji chemiczne wchodzących w skład mieszanin konserwujących jak i wyizolowano bakterie i grzyby tworzące florę preparatów muzealnych. Wyniki z tych trzech części pozwoliły stworzyć koncepcję preparatu anatomicznego jako bardzo specyficznego ekosystemu, którego stan determinują wpływające na siebie czynniki zewnętrzne, zachodzące reakcje fizyko-chemiczne i działalność mikroorganizmów. Ustalono nowe fakty użyteczne w praktyce konserwatorskiej, jak i mogące stanowić wsparcie projektowanych w przyszłości badań zbiorów muzealnych. Zidentyfikowano nieznane dotychczas czynniki zagrażające zdrowiu, mogące wystąpić podczas pracy z materiałem muzealnym. W

rezultacie udowodniono, że jednoczesne zastosowanie metod z poszczególnych dziedzin istotnie zwiększa proces poznawczy prowadzonego badania.

Trzeci z artykułów poświęcony jest badaniom Muzeum Mulaży Dermatologicznych Uniwersytetu Medycznego we Wrocławiu. Przeprowadzono badania źródeł historycznych, literatury naukowej poświęconej mulażom woskowym (przegląd niesystematyczny) oraz badania mikrobiologiczne i mikologiczne kolekcji i jej otoczenia. Informacje uzyskane na temat procesów powstawania zbiorów jak i materiałów z jakich je wykonywano pozwoliły wyjaśnić dlaczego są one kolonizowane w niewielkim stopniu. Wyniki z analizy kontaminacji powietrza porównano z innymi dotychczas przeprowadzonymi badaniami prowadzonymi w budynku Katedry Dermatologii, Wenerologii i Alergologii Uniwersytetu Medycznego we Wrocławiu. Ustalono, że muzeum nie jest bardziej zanieczyszczone od reszty pomieszczeń, ale niektóre ze zidentyfikowanych organizmów są charakterystyczne dla oddziałów szpitalnych. Ponadto zaproponowano tę metodę badawczą jako potencjalne narzędzie konserwatorskie do monitorowania zmian zachodzących w przestrzeni muzeum i kolekcji. Dostępne badania zbiorów mulaży przydatnych technicznie są kazuistyczne. Ogranicza to możliwości wnioskowania i wskazuje na potrzebę kontynuację badań tych w przyszłości.

W rozprawie przedstawiono zaistnienie paradoksu, polegającego na tym, że choć dostępne są różne opracowania naukowe podejmujące zagadnienia muzeów medycznych, to (i) badania poświęcone samym zbiorom, tj. badania podmiotowe obiektów muzealnych, (ii) badania weryfikujące użyteczność metod badawczych oraz (iii) badania mające zastosowanie praktyczne np. w konserwatorstwie, są nieliczne. Zaproponowano interdyscyplinarną koncepcję badań materiału muzealnego i muzeów medycznych, jako obiektów badawczych, których analiza poprzez pryzmat pojedynczej dyscypliny nauki, nie pozwala na ocenę obiektywną.

7. STRESZCZENIE W JĘZYKU ANGIELSKIM

The dissertation was based on a montematic series of three articles published in international scientific journals indexed in the PubMed database and included in the Journal Citation Reports list and included in the Ministry's list of scientific journals. The articles were published in peer-reviewed journals with a total impact factor of 7,561 and 320 ministerial points. The research was funded by a FAST (Student Activities Fund - 2nd edition) grant entitled. "Anatomical Museum of the Medical University of Wrocław - history, science, art" with the number in the University's internal records: GMIN.A351.20.006 and a subsidy from the Minister of Education and Science for the competition task entitled. "Evaluation of microorganisms isolated from anatomical collections in terms of health risks and resistance to disinfectants", identified in the SIMPLE system: SUBK.A351.23.020.

The first article among the series is a paper covering a review as well as a meta-analysis of the literature on the study of museums and museum collections of human anatomy. The paper presents the state of knowledge on the subject and the occurring domains of research interest. A surprising lack of research dedicated to the conservation of museum collections as well as a lack of research on museum objects (i.e. subject-based research) was demonstrated. Satisfactory research designed in an interdisciplinary concept was not found in this field.

In the following paper, dedicated to the analysis of the preservation fluids of historic anatomical specimens, the results of the conducted research (historical, chemical and microbiological research) dedicated to the preservation fluids of the historic anatomical collection are presented. As a result, important historical facts about the studied collection were established, research methods were selected and verified, some chemical substances included in preservation mixtures were determined, and bacteria and fungi forming the flora of the museum preparations were isolated. The results of these three parts allowed the concept of the anatomical specimen as a very specific ecosystem, whose condition is determined by influencing external factors, occurring physico-chemical reactions and the activity of microorganisms. New facts have been established that are useful for conservation practice and that can support the projected research of museum collections in the future. Previously unknown health risk factors that can occur when working with museum material were identified. As a result, it was shown that the simultaneous application of methods from

the individual disciplines significantly enhances the cognitive process of the research being carried out.

The third article is devoted to the research of the Moulages museum of the Medical University of Wrocław. Historical sources, scientific literature on wax moulages (non-systematic review) and microbiological and mycological studies of the collection and its surroundings were carried out. The information obtained on the processes of collection formation as well as the materials from which they were made helped to explain why they are poorly colonised. The results from the air contamination analysis were compared with other studies carried out to date in the building of the Department of Dermatology, Venereology and Allergology at Wrocław Medical University. It was found that the museum is not more contaminated than the rest of the premises, but some of the identified organisms are characteristic of hospital wards. Furthermore, this research method was proposed as a potential conservation tool to monitor changes in the museum space and collections. Available studies of technically useful moulage collections are casuistic. This limits the possibilities for inference and points to the need to continue this research in the future.

The dissertation outlines the existence of a paradox in that, although various scientific studies addressing medical museums are available, (i) studies dedicated to the collections themselves, i.e. subject studies of museum objects, (ii) studies verifying the usefulness of research methods, and (iii) studies with practical applications, e.g. in conservation, are scarce. An interdisciplinary concept of research on museum material and medical museums has been proposed, as research objects whose analysis through the prism of a single discipline of science does not allow for objective evaluation.

8. CURRICULUM VITAE

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kierownik projektu	**Projekt: Muzeum Anatomiczne Uniwersytetu Medycznego we Wrocławiu - historia, nauka, sztuka** - w ramach Funduszu Aktywności Studenckich - nabór II organizowany przez Prezydenta Wrocławia w związku z realizacją strategii rozwoju Wrocławia „Przedsiębiorczy Wrocław 2030”, Grant realizowany od 1.10-31.12.2020r. Numer w ewidencji wewnętrznej Uczelni: GMIN.A351.20.006
kierownik projektu	**Subwencja Ministra Edukacji i Nauki na projekt konkursowy 2023 pt. ‘Ocena mikroorganizimów izolowanych ze zbiorów anatomicznych pod kątem zagrożenia zdrowotnego i oporności na substancje dezynfekcyjne.’** kwota subwencji 50.000zł numer w systemie SIMPLE: SUBK.A351.23.020

9. DOROBEK NAUKOWY

(z wyłączeniem prac stanowiących cykl publikacji do Rozprawy Doktorskiej)

9.1 Publikacje w międzynarodowych czasopismach:

1. Domagała, Z., Mrożek, A., Piotrowska, A., Olesińska, N., **Domański, J.**, Kobierzycki, C., Thelen, M., & Śliwa, J. (2022). Expression of selected cytokeratins in human placenta - a preliminary observational study. *Medical Journal of Cell Biology*, *10*, 155–162. https://doi.org/10.2478/acb-2022-0023
   Punktacja Ministerialna: 20 punktów

2. Domagała, Z., **Domański, J.**, Smyczek, N., & Galk, C. (2022). Maceration stage in corrosion cast specimen procedure in anatomy: a minireview. *Folia Morphologica*, *81*, 825–833. https://doi.org/10.5603/fm.a2021.0119
   IF: 1,195; Punktacja Ministerialna: 70 punktów

3. Domagała, Z., Pinkowska, A., Piotrowska, A., **Domański, J.**, Tarkowski, V., Zimmer-Stelmach, A., & Śliwa, J. (2022). Utility of the Movat pentachrome stain technique in the microanatomical analysis of the human placenta. *Italian Journal of Anatomy and Embryology*, *126*, 25–32. https://doi.org/10.36253/ijae-13882
   IF: 0,012; Punktacja Ministerialna: 40 punktów

4. **Domański, J.**, Wanat, M., Ciach, J., Osuch, A., Kurc-Darak, B., Woźniak, S., & Domagała, Z. (2022). Off-line or on-line? - near-peer assisted anatomy education in the time of Covid-19 pandemic – a single center randomized controlled study. *Italian Journal of Anatomy and Embryology*, *126*, 17–24. https://doi.org/10.36253/ijae-13877
   IF: 0,012; Punktacja Ministerialna: 40 punktów

5. Wanat, M., Nowak, B., Świątko, A., Mirkowski, K., **Domański, J.**, Dąbrowski, P., & Domagała, Z. (2022). Trigger points in medical practice - current therapeutic directions. *Medical Journal of Cell Biology*, *10*, 129–137. https://doi.org/10.2478/acb-2022-0020
   Punktacja Ministerialna: 20 punktów

6. **Domański, J.**, Krajewski, P. K., Baran, W., & Szepietowski, J. C. (2021). Chronic upper eyelid oedema in a young boy: a quiz. *Acta Dermato-Venereologica*, *101*, art. https://doi.org/10.2340/actadv.v101.415
   IF 4,437; Punktacja Ministerialna: 100

7. Domagała, Z., **Domański, J.**, Zimmer, A., Tarczyńska, A., Śliwa, J., & Gworys, B. (2020). Methodology of preparation of corrosive specimens from human placenta - a technical note. *Annals of Anatomy-Anatomischer Anzeiger*, *228*, art. https://doi.org/10.1016/j.aanat.2019.151436
   IF: 2,976; Punktacja Ministerialna: 100

9.2 Doniesienia zjazdowe:

1. Mrożek, A., **Domański, J.**, Wanat, M., Ciach, J., Osuch, A., Kurc-Darak, B., Woźniak, S., & Domagała, Z. (2022). Nauczanie anatomii wspomagane rówieśniczo w czasie pandemii COVID - jednoośrodkowe randomizowane badanie naukowe. W (red.), *34. Ogólnopolski Kongres Polskiego Towarzystwa Anatomicznego. Szczecin, 14 - 16 wrzesień 2022 r. Książka streszczeń* (s. 165). Szczecin.

2. **Domański, J.**, Janczura, A., Wanat, M., Wiglusz, K., Grajzer, M., Simmons, J., & Domagała, Z. (2022). AN ANATOMIST- DETECTIVE, the chemical techniques in the service of museologist. W (Red.), *116th Annual Meeting of the Anatomische Gesellschaft - Joint Meeting with the Anatomical Society. Berlin, September 20-23, 2022. Abstract book [online]* (s. 47).

3. Domagała, Z., **Domański, J.**, Wanat, M., & Simmons, J. E. (2022). Terra incognita w muzealnictwie anatomicznym - spojrzenie z perspektywy standardów medycyny opartej na faktach. W (red.), *34. Ogólnopolski Kongres Polskiego Towarzystwa Anatomicznego. Szczecin, 14 - 16 wrzesień 2022 r. Książka streszczeń* (s. 126). Szczecin.

4. Domagała, Z., Janczura, A., Wanat, M., Wiglusz, K., Grajzer, M., Simmons, J., & **Domański, J.** (2022). Microbiological evaluation of selected historical anatomical specimens. W (Red.), *116th Annual Meeting of the Anatomische Gesellschaft - Joint Meeting with the Anatomical Society. Berlin, September 20-23, 2022. Abstract book [online]* (s. 46).

5. Domagała, Z., **Domański, J.**, Zimmer, A., Tarczyńska, A., & Woźniak, S. (2020). Human Placental Corrosion Cast Studies - a technical note. *Journal of Anatomy*, *236*, [129] poz.O097. https://doi.org/10.1111/joa.13163
   19th International Federation of Associations of Anatomists Congress. London, UK, 9th-11th August 2019

6. Domagała, Z., Klekowski, J., **Domański, J.**, Tarkowski, V., & Woźniak, S. (2021). Real-time visualization: early ultrasound exposure in undergraduate medical students. *Journal of Anatomy*, *238*, 195–196 poz.S25.
   https://doi.org/10.1111/joa.13290
   Winter Meeting of the Anatomical Society, Lancaster, 18-20 December 2019. Conference abstract

7. Domagala, Z., Mazurek M., Kozłowski O., Drążyk M., Piotrek O., Thelen M., Woźniak S., **Domański J.** (2023) How to prepare the best cadaver for educational purpose the challenges during vascular access through the common carotid artery during embalming procedure in humans. Anatomical Society Winter Meeting 2022 Nottingham 17th-19th April 2023

8. **Domański J.**, Szepietowski J. C., Woźniak S., Janczura A., Tarkowski W., Simmons J. E., Wanat M., Domagala Z. (2023) Tutankhamun's curse in anatomical museum. Anatomical Society Winter Meeting 2022 Nottingham 17th-19th April 2023

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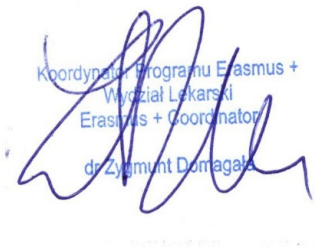
mój udział polegał na współtworzeniu metodologii, pomocy w tworzeniu manuskryptu i akceptacji jego finalnej wersji.



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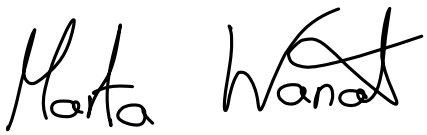

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Oświadczam, że w pracy:

Jurand Domański, Adriana Janczura, Marta Wanat, Katarzyna Wiglusz , Magdalena Grajzer, John E Simmons, Zygmunt Domagała, Jacek C Szepietowski: *Preservation fluids of heritage anatomical specimens -a challenge for modern science. Studies of the origin, composition and microbiological contamination of old museum collections.* J Anat. 2023 Apr 6. doi: 10.1111/joa.13854.

mój udział polegał na nadzorze naukowym i akceptacji finalnej wersji manuskryptu.

Uniwersytet Medyczny we Wrocławiu
KATEDRA I KLINIKA
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Wrocław, 26/05/2023

OŚWIADCZENIE

Oświadczam, że w pracy:

Jurand Domański, Rafał Białynicki-Birula, Urszula Nawrot, Elżbieta Piątkowska, Zygmunt Domagała, Jacek C Szepietowski: *Microbial load of heritage dermatological moulages of the historic university department in Wrocław, Poland.* Adv Dermatol Allergol 2023 May 26. doi.org/10.5114/ada.2023.127197

mój udział polegał na pozyskaniu danych, współtworzeniu metodologii badań, pomocy w tworzeniu manuskryptu i akceptacji jego finalnej wersji.

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Wrocław, 30/05/2023

OŚWIADCZENIE

Oświadczam, że w pracy:

Jurand Domański, Rafał Białynicki-Birula, Urszula Nawrot, Elżbieta Piątkowska, Zygmunt Domagała, Jacek C Szepietowski: *Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland.* Adv Dermatol Allergol 2023 May 26.
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mój udział polegał na pozyskaniu i opracowaniu danych oraz pomocy w tworzeniu manuskryptu.

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Wrocław, 22/05/2023

OŚWIADCZENIE

Oświadczam, że w pracy:

Jurand Domański, Rafał Białynicki-Birula, Urszula Nawrot, Elżbieta Piątkowska, Zygmunt Domagała, Jacek C Szepietowski: *Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland.* Adv Dermatol Allergol 2023 May 26.
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Uniwersytet Medyczny we Wrocławiu
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dr n. med. Zygmunt Domagała
Zakład Anatomii Prawidłowej Uniwersytetu Medycznego we Wrocławiu

Wrocław, 22/05/2023

OŚWIADCZENIE

Oświadczam, że w pracy:

Jurand Domański, Rafał Białynicki-Birula, Urszula Nawrot, Elżbieta Piątkowska, Zygmunt Domagała, Jacek C Szepietowski: *Microbial load of heritage dermatological moulages of the historic university department in Wroclaw, Poland.* Adv Dermatol Allergol 2023 May 26.
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mój udział polegał na pomocy w tworzeniu manuskryptu i akceptacji jego finalnej wersji.

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Uniwersytet Medyczny we Wrocławiu

Wrocław, 26/05/2023

OŚWIADCZENIE

Oświadczam, że w pracy:

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11. ETYKA

KOMISJA BIOETYCZNA
przy
Uniwersytecie Medycznym
we Wrocławiu
ul. Pasteura 1; 50-367 WROCŁAW

OPINIA KOMISJI BIOETYCZNEJ Nr KB – 167/2021

Komisja Bioetyczna przy Uniwersytecie Medycznym we Wrocławiu, powołana zarządzeniem Rektora Uniwersytetu Medycznego we Wrocławiu nr 278/XVI R/2020 z dnia 21 grudnia 2020 r. oraz działająca w trybie przewidzianym rozporządzeniem Ministra Zdrowia i Opieki Społecznej z dnia 11 maja 1999 r. (Dz.U. nr 47, poz. 480) na podstawie ustawy o zawodzie lekarza z dnia 5 grudnia 1996 r. (Dz.U. nr 514 z 2020 r.) w składzie:

dr Joanna Birecka (psychiatria)
dr Beata Freier (onkologia)
dr hab. Tomasz Fuchs (ginekologia, położnictwo)
prof. dr hab. Dariusz Janczak (chirurgia naczyniowa, transplantologia)
dr hab. Krzysztof Kaliszewski (chirurgia endokrynologiczna)
dr prawa Andrzej Malicki (prawo)
dr hab. Marcin Mączyński (farmacja)
Urszula Olechowska (pielęgniarstwo)
prof. dr hab. Leszek Szenborn (pediatria, choroby zakaźne)
prof. dr hab. Andrzej Szuba (choroby wewnętrzne, angiologia
ks. prof. Andrzej Tomko (duchowny)
prof. dr hab. Mieszko Więckiewicz (stomatologia)
dr hab. Andrzej Wojnar, prof. nadzw. (histopatologia, dermatologia) przedstawiciel Dolnośląskiej Izby Lekarskiej)
dr hab. Jacek Zieliński (filozofia)

pod przewodnictwem
prof. dr hab. Jerzego Rudnickiego (chirurgia, proktologia)

Przestrzegając w działalności zasad Good Clinical Practice oraz zasad Deklaracji Helsińskiej, po zapoznaniu się z projektem badawczym pt.

„Ocena anatomiczna mięśni ludzkich położonych w warstwie powierzchownej grupy przedniej mięśni przedramienia u płodów ludzkich"

Zgłoszonym przez **mgr Małgorzatę Suchanecką,** zatrudnioną w Zakładzie Anatomii Prawidłowej Katedry Morfologii i Embriologii Człowieka Uniwersytetu Medycznego we Wrocławiu oraz złożonymi wraz z wnioskiem dokumentami, w tajnym głosowaniu postanowiła **wyrazić zgodę** na przeprowadzenie badań w Zakładzie Anatomii Prawidłowej Uniwersytetu Medycznego we Wrocławiu **pod warunkiem zachowania anonimowości zgromadzonych danych.**

Uwaga: Badanie to zostało objęte ubezpieczeniem odpowiedzialności cywilnej Uniwersytetu Medycznego we Wrocławiu z tytułu prowadzonej działalności.

Pouczenie: W ciągu 14 dni od otrzymania decyzji wnioskodawcy przysługuje prawo odwołania do Komisji Odwoławczej za pośrednictwem Komisji Bioetycznej UM we Wrocławiu.

Opinia powyższa dotyczy projektu badawczego realizowanego z działalności statutowej.

Przewodniczący Komisji Bioetycznej
przy Uniwersytecie Medycznym

prof. dr hab. Jerzy Rudnicki

Wrocław, dnia 22 lutego 2021 r.