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Autopsy doesn't always tell all: The importance of exhuming skeletal remains of cemetery unidentified decedents

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ABSTRACT

The process of personal identification differs according to the state of preservation of the corpse, becoming more complicated when dealing with remains altered by taphonomic variables. Since 2015, the staff of the Laboratory of Forensic Anthropology and Odontology (LABANOF) of the University of Milan has been engaged in recovering the skeletons of 36 unknown people from the cemeteries of the city of Milan, to redraw their biological profiles and give them back the possibility of being identified. Of the 36 starting skeletons, 7 have been identified and therefore are not the object of this study, 25 individuals were previously subjected to an autopsy examination and 4 skeletons were studied for the first time in the present work. The post-mortem data of all the individuals who had previously undergone autopsy and had not yet been identified (n = 25) have been retrieved from the archives of the Institute of Forensic Medicine in Milan and allowed for a comparison with those obtained from the new anthropological study of each exhumed skeleton. The autopsy information alone lacked important details. In all cases, the anthropological examination proved to be capable of detailing the biological profile of the unknowns even after a long period of inhumation (even more than 20 years). The limitations of instrumental investigations in achieving the goal single-handedly and the importance of recovering the unknown skeletons from cemeteries for identification purposes are discussed.

1. Introduction

Every year a large number of unidentified bodies arrive at various forensic institutes located in Italy and the rest of the world and, despite all efforts, many of them remain unidentified [1-18]. Table 1. shows the data on the number of bodies that remained unidentified despite the identification attempts.

The situation worsens when these bodies are buried without an identification process having been conducted or without waiting for the latter to bear fruit. In Italy, usually only in cases where a crime is suspected, the prosecutor may request an autopsy to ascertain the cause of death "after making the necessary investigations for identification" (Art. 116 of the Regulation for the Implementation of the Code of Criminal

Procedure) [19]. Steps forward on national territory were taken with the foundation, in 2007, of the Office of the Special Commissioner for Missing Persons, which is responsible in Italy for updating data on unidentified corpses and missing persons [20]. In the same year, the National Register of Unidentified Corpses was established. In 2010, a new information system was then introduced, called Ri.Sc. ("Ricerca Scomparsi") with the aim of comparing data on missing persons with unidentified corpses [21]. On March 6th, 2015, the first memorandum of understanding was signed between the extraordinary commissioner and some prosecuting authorities in Lombardy, including that of Milan [22]. The Laboratory of Forensic Anthropology and Odontology (LABANOF) was then involved in the identification of unknowns who died in hospital and unidentified corpses and remains found in the area of competence,

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Table 1

Report on the phenomenon of missing persons and bodies without identity in Italy and the rest of the world. The study by Andereev et al. considered only males aged between 25–54.

Author	Country	Period	Bodies remained unidentified
Commissario Straordinario del Governo per le persone scomparse [18]	Italy	1974–2020	942
Paulozzi et al. [3]	USA	1979-2004	10.748
Hanzlick et Smith [1]	Georgia- USA	1988-2002	59
		2003-2004	6
NamUs [17]	USA	2007-2022	14.164
Cattaneo et al. [4] Mazzarelli et al. [14]	Milan-Italy	1995–2019	100
Cavard et al. [5]	Paris-France	2003-2009	18
Andreev et al* [2]	Izhevsk- Russia	2004–2005	634
Evert [6]	Pretoria- South Africa	2005–2008	848
Chattopadhyay et al. [8]	Calcutta- India	2010-2011	505
Reid et al. [13]	Cape Town- South Africa	2010-2017	2.476
Blau et Rowbotham [16]	Victoria- Australia	1960–2020	132

including those not connected with a crime [22]. The ultimate aim was, concerning forensic pathologists and anthropologists, to adequately collect the post-mortem data necessary for the drafting of the Ri.Sc form for the unidentified bodies and the minutes that the public prosecutor must draw up in the case of the death of one or more persons without identity (Art. 78 of the Italian Civil State law) [23]. In 2023, the protocol was renewed and saw more extensive regional involvement.

Needless to say, in the absence of an autopsy, a significant amount of relevant information necessary for identification is missed [24-26,27]. However, to the best of our knowledge, no study quantified for the loss of information even when an autopsy was performed. In the same year of the signature of the first agreement protocol for unidentified bodies (2015), LABANOF initiated the project for the recovery of unclaimed skeletons from the city's main cemeteries. This was possible in accordance with the Article 43 of Presidential Decree No. 28 (10 September 1990), which grants Universities to recover and collect skeletons that remained unclaimed for ten years after burial, exclusively for study purposes [28]. If not recovered, unclaimed remains are in fact transferred to a common ossuary, losing any possibility of identification [28,29]. In this study, 29 skeletons of unknown persons exhumed from the main cemeteries in Milan underwent anthropological analyses. This allowed to adequately fill in the Ri.Sc forms [30] for inclusion of all of them in the national database Ricerca Scomparsi. For 25 of them, it was possible to compare anthropological results with autopsy and anthropological data obtained and stored at the Institute of Forensic Medicine in Milan and at LABANOF at the time of death. The results concerning the biological profile and the observation of trauma and prostheses, lead to a discussion on the insufficiency of the autopsy examination alone towards identification requirements. Moreover, considering limitations of imaging methods, the possibility of early exhumation and anthropological examination of the remains of cemetery unknowns is also discussed.

2. Materials and methods

Since 2015, 36 unidentified cemeterial skeletons have been recovered from the Maggiore, Lambrate and Bruzzano cemeteries of the city of Milan. The unknown individuals were buried between 1992 and 2005 and exhumed between 2013 and 2019. The exhumation process normally involves removing most of the soil using excavators, and the remaining material with trowels. Once the skeleton has been exposed and taken out of the ground, the soil beneath the body is collected and then sieved in the laboratory. The recovered remains are placed in metal boxes fitted with a tag indicating sex, date of death, field and burial site and transferred to the university. In some instances, the exhumations were carried out by cemetery workers without any anthropological support [31]. Personnel of LABANOF actively participated in the exhumation operations as of 2016.

Seven skeletons were excluded from the study as they were positively identified and then returned to their families. In Italy it is not possible to study skeletons identified and claimed by their relatives. Their identification took place by means of genetic examinations (n = 3), odontological analysis (n = 2, of which one by X-ray image comparison and the other through the superimposition of an ante-mortem photo of the missing person with the three-dimensional image of a post-mortem dental cast realised on the unknown body) and by visual recognition, associated with the dental superimposition [32] of ante-mortem photos of the missing person and post-mortem pictures of the unknown body (n = 2).Of the remaining 29, an autopsy had been performed for 25 individuals at the time of death, so the related autopsy report and the additional post-mortem data were searched for in the archives of the Institute of Forensic Medicine in Milan and of LABANOF and were retrospectively assessed. This allowed a comparison with the reports of the anthropological examination of each skeleton. The skeletons of the remaining 4 individuals were instead examined for the first time in the course of this work. In three cases, the absence of an autopsy was attributable to a death which had occurred in hospital from known causes and therefore not further investigated; in one case an autopsy was performed in a different institution, resulting in the absence of a report for comparison. The study was conducted blindly, cleaning and analysing the skeletons from an anthropological point of view, and then comparing the information obtained with the autopsy data from the recovered documentation. Once the comparison was made, it was finally possible to fill in the postmortem (PM) Ri.Sc form with all the information concerning the unknown individuals.

2.1. Anthropological examination

The anthropological study aimed at reconstructing the biological profile including sex, age, stature, population affinity, presence of pathologies and/or skeletal abnormalities as well as the analysis of dental characteristics. Each unknown skeleton was first removed from the metal crate and then laid in an anatomical position. An assessment of the preservation state and the inventory of the recovered elements followed. In almost all the subjects, the remains were completely skeletonized. The skeletal remains were dry-cleaned using toothbrushes and steel spatulae to remove residues. Persistent soft tissues were removed through maceration in water and cleaning by mechanical action, with the aid of tweezers and a synthetic bristle brush. The bones were left to dry at room temperature. A complete biological profile was created for all individuals. Results obtained in the sex determination, age estimation and trauma analysis are presented. The choice of study methods was adapted to the state of preservation and the type of bone elements present for each skeleton. Morphological [33-35] and metric methods [36-40] were applied to determine sex. The morphological estimation of age was based on the aspect of the pubic symphysis [41], the auricular surface of the ileum [42,43] the sternal end of the 4th rib [44] and the alteration of the acetabular joint and the auricular surface [45]. In some cases, the degree of ossification of the sternal end of the clavicle was assessed [46-48]. Dental methods evaluating the degree of change in pulp chamber width on canines [49] and other monoradicular teeth [50] were also applied. When no other method was applicable, cranial [51,52] and palatine [53] sutures were observed. Trauma analysis consisted of the identification of antemortem (AM) trauma and the distinction between perimortem signs of trauma and postmortem damage [54-56]. The possible presence of prostheses or other signs suggestive of a surgical intervention were also noted.

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The authors wish to present three of the most interesting cases for a better explanation of the results of the study.

3. Results

The anthropological examination allowed the determination of male (88 %, n = 22) or female sex (12 %, n = 3), even for the sole case in which it was not possible at autopsy. In three cases, the autopsy records reported the co-presence of male genitalia and breast prostheses, indicating a transgender individual which could not be inferred from the sole analysis of the skeletal remains.

Age could not be determined at the autopsy in 24 % (n = 6) of the cases. In the cases where the two types of data corresponded, the age ranges estimated by anthropology allowed a better classification, especially for individuals aged between 17 and 30 years. The anthropological study allowed four individuals, which were misclassified as adults at the time of the autopsy, to be recognised as adolescents.

Evidence of trauma was observed in only 8 % (n = 2) of the unknown bodies during the autopsy, whereas it was found in 84 % (n = 21) exhumed skeletons. In two cases where skeletal trauma were mentioned at autopsy, the description was incomplete compared to what observed on the skeleton.

In four individuals, the autopsy revealed the presence of prostheses (three breast implants and one dental implant) which were found in three cases on anthropological examination (fragments of breast implants in 2 cases and dental prosthesis in the other).

Fig. 1 shows the number of bodies recovered throughout 7 years. The exhumation was in some cases carried out by the cemetery workers without any anthropological support, which often led to only partial recovery of the remains. Since 2016, in 19 out of 29 cases, the exhumation was carried out in the presence of anthropological personnel with an almost complete recovery of skeletal remnants. However, the maximum number of exhumations planned by the cemetery took place in 2013 (n = 8), prior to the lab's intervention. This meant that in about a third of the cases, the skeletal recovery was only partial.

4. Three particular cases

The first case presented is that of a male individual found dead in the toilets of Milan's Central Station. Sex, which was in any case reported in the cemetery documents and evident at autopsy, was determined assessing cranial and pubic traits [33–35]. On anthropological examination, it was not possible ascertain the gender evident at autopsy (transgender). Age-at-death estimation was between 20–30 years, because of incomplete fusion of the sternal end of the clavicle (Fig. 2) [48]. The estimated age was therefore lower than that visually



Fig. 2. Medial end of the clavicle of individual 01/1996 not completely ossified.

attributed by the sector during the autopsy examination (30–35 years). The presence of two ante-mortem traumas at the IV and V rib on the left side was noted, whereas the autopsy only reported the presence of scars (18 scars and 4 scar areas). The breast implants described at the autopsy, although fragmented, were found associated to the skeletal remains.

The second unknown individual died at the Fatebenefratelli hospital in Milan in 2003. The sex determined on the skeletal remains was consistent with the autopsy report. The examination of the medial end of the clavicle [48] and the fourth rib [44] assessed an age range between 24 and 37 years, which was lower than that found in the autopsy report (26–50 years old). At the same time, the presence of signs of two surgeries, one neurosurgical and the other orthopaedic in the right tibia (Fig. 3), of which only one was appreciated at the autopsy, was observed. Even more remarkable was the evidence on the skeleton of 52 antemortem bone traumas of different ages (four with the corresponding scarring on the skin), that went unnoticed by the forensic pathologist



Fig. 1. Showing the year of exhumation of the 29 skeletons considered for the period 2013—2021. Since 2016, the LABANOF personnel have participated in the recovery.



Fig. 3. Outcomes of surgery with nails and screws to the right tibia of unknown 02/2003.

(eg. Figs. 4-6).

The third and last case is that of a male individual who was hit by a car while walking naked on a highway. Even in this case, no discrepancies were found between the sex observed at autopsy and that anthropologically determined. Because of the poor state of preservation of the skeleton, the age could only be estimated from the cranial sutures (30–60 years) [51]. Five ante-mortem traumas (e.g Fig. 7), not previously noted, were observed on the skeleton. No prostheses or medical devices were found.





Fig. 5. Bone callus on the proximal phalanx of the fifth digit of the left hand of the unknown 02/2003.



Fig. 6. Bone callus at the manubrio-sternal joint in Unknown 02/2003.

5. Discussion

This paper highlighted the nature and extent of the loss of information, which can be drawn from a thorough skeletal analysis even after a long period of inhumation (up to 20 years), and how much can be missed at the autopsy.

At the time of the autopsies, instrumental examinations (X-rays, CT scans) were not routinely performed on all unknown bodies, as is done at our Institute today. Although an important complement, these examinations are not by themselves sufficient to provide the same amount of information as a direct and extensive examination of the skeletal remains. This is true for perimortem trauma but also for antemortem ones, which may often be difficult to observe with the different imaging methods available [24,57–61].

Bearing in mind the variability of fracture repair depending on age,



Fig. 4. Bone callouses on the left ribs of unknown 02/2003.



Fig. 7. Bone callus on the nasal bones of the unknown 03/2004.

location and type of fracture [60], the bone callus is usually clearly visible on X-ray images no earlier than 6–8 weeks after injury [60,62–64]. Furthermore, the two-dimensional projection of radiographs is prone to misinterpretation in the living subject [61], and, in our experience, even more in the deceased. In fact, it has been shown that for the radiographic determination of fracture union, the sensitivity is only 54 % and the specificity 63 % [65] with errors in the estimation of bone fusion approaching 40 % [58]. Orthopaedic surgeons themselves usually rely on the radiographic study alone in less than 50 % of cases, preferring to pair it with the evaluation of a series of clinical and functional criteria [57,66]. These are, however, missed in the cadaver.

Although it is widely accepted that CT scans have a better and earlier ability to detect bone fractures and calluses than radiography, in some cases the time advantages have averaged only a few days (7.3 weeks after injury compared to 8 for radiography) [67]. Furthermore, clinical scanners usually have limited resolution and therefore tend to miss the smallest and thinnest lesions [59]. It should also be considered that as with radiography, the presence of metal prostheses on CT-scan may disturb the detection of any lesions [60,61]. The known limitations of CT have led to an increasing recognition of micro-CT [68–70]. However, the results so far obtained and shared in the literature do not grant the method sufficient autonomy in decision-making [59,71]. In one study, out of fourteen bone fractures, micro-CT detected only five at a first analysis and a further eight only after a second check [69].

Within this domain, increasing confidence has been given to ultrasound. Indeed, it appears that ultrasound signs of bone healing can be identified as early as 1–3 weeks after fracture [61,72]. However, the acoustic impedance of bone can sometimes produce errors in signal interpretation [61] and it can be difficult to evaluate the deep cortical surface [60]. In addition, not all forensic pathologists are familiar with this method. Nevertheless, the possibility of obtaining 3D images of the bone callus makes it more user-friendly [72].

It should also be considered that, regardless of the method used, in some pathological healing courses the formation of a bone callus does not occur. This is the case for example of both viable oligotrophic nonunions and non-viable atrophic nonunions [60]. In this regard, criteria sometimes used to diagnose the absence of consolidation and the possible occurrence of pseudoarthrosis are the evaluation of the vacuum phenomenon, associated with the presence of gas within the fracture, as well as the presence of liquid in situ [73,74].These factors may be unhelpful in the cadaver as the post-mortem transformation processes are associated with fluid and gas production [75], which could mislead.

In a skeletonized subject it is usually possible to define as antemortem a fracture produced no less than 2–3 weeks before death [76]. Histological investigations performed on bones samples may, however, allow a better appreciation of some features commonly observed in an ante-mortem fracture as early as 4–7 days after injury, such as: the absorption of cortical bone adjacent to the lesion, the first Howship lacunae at the lesion margins and the appearance of the first bone spicules between the lesion ends [77]. This offers the possibility of an earlier and safer diagnosis of ante-mortem trauma of the individual once his skeleton is available.

In all cases presented in our study, the anthropological examination [30,78–81], proved to be capable of detailing the biological profile of the unknown persons. It provided a more accurate estimation of age and an enormous contribution in the detection of antemortem trauma, useful in the identification process. In the three cases examined, the study of the skeleton revealed the presence of a total of 59 ante-mortem traumas and one surgical intervention, none of which had been observed previously. Histological investigations were not necessary because the calluses were all well recognizable. These results lead to consider the insufficiency of the autopsy examination alone for identification purposes.

However, anthropological examination has its limits too. Indeed, even if the skeletal analysis correctly assessed the sex of all individuals, including the one for whom it was unclear at autopsy, it did not allow a confident evaluation of gender in three transgender cases seen at autopsy. Failure to take this into account would mean running the risk of drawing up a misleading biological profile, bringing the search for a missing person phenotypically quite different from the real one [29,82]. For prostheses, having found one less in anthropological examination than in autopsy examination can be attributed to the degradation of some types of prostheses over time. This is the case of breast implants, which unfortunately are also the most common to find [83].

Nonetheless, autopsy examination should still not be considered sufficient on its own to provide all the details necessary to increase the likelihood of identifying an unknown body. Although a harbinger of important information, which may be lost through postmortem transformative processes and the taphonomic effect of the depositional environment [56,78,84–88], it is always advisable to couple it with a careful anthropological study.

A first step to intertwine autopsy and anthropological data is to properly perform bone samples at the time of necropsy (pubic symphysis, fourth rib, mono-radicular tooth, clavicle in younger individuals), to facilitate later age estimation. Moreover, the necroscopic examination should always be preceded by instrumental investigations to gather as much information as possible on the presence of prostheses, potential ante-mortem trauma or other skeletal abnormalities [24,89]. In fact, despite some limitations, which prevent their exclusive use, these methods remain a valuable aid. This practice can also direct the forensic pathologist in the execution of possible further useful sampling for subsequent investigation or even a more thorough examination of the same [24,59].

The second step, if a full anthropological examination was not possible earlier, concerns the moment of exhumation. Exhumation is normally understood as the process of recovering a legitimately interred body to perform a first or second autopsy on the same one [90]. The moment of exhumation is already known by those in the field to be a supplementary opportunity to retrieve information overlooked or difficult to obtain at autopsy [91]. This opportunity becomes a necessity in the case of skeletal remains of unidentified persons. In Italy, an exhumation performed before 10 years from inhumation is considered extraordinary. If this does not happen, 10 years after the burial of a body, the unclaimed remains are routinely exhumed and transferred to a common ossuary, without any further investigation (Presidential Decree 285/90 Art. 83) [28]. In this regard, for many nameless buried skeletal remains, the moment of exhumation represents the last opportunity to attempt the restoration of their identity, so their recovery is justified and urged.

Moreover, it would be desirable, if possible, to recover bodies without identity earlier to shorten the time needed for their identification. This would ensure less impact on the lives and health of those left behind [92-95]. In Italy, according to Article 82 of the Mortuary Police Regulation [28], it is possible to proceed with an anticipated exhumation if ground conditions allow for more rapid skeletonization of the corpse. The period of inhumation cannot, however, be less than five years unless, as stipulated in Article 83, an anticipated exhumation is requested by the judicial authority or there is an authorisation from the mayor for transport to another burial site or for cremation. In other countries, such as Greece, the timeframe is shorter and generally, 3 to 5 years after the burial of an individual, the remains are exhumed and placed in a metal box, as in Italy (a practice that would seem to have originated in monasteries, due to space constraints) [78]. In Brazil, after five years of inhumation, permission was obtained to recover skeletal remains from cemeteries and to verify the accuracy of the generalities recorded at death by comparing AM data, recorded by the Bom Pastor cemetery administration, and PM data obtained from the anthropological study [79]. In Tunisia, India and Pakistan no time limitations exist to the request for an exhumation [96–98]. However, in India the widespread practice of cremation and scattering of ashes in the Ganges can make the process infeasible [99]. In Pakistan, the obstacle can often be cultural and religious reasons whereby family members have qualms about accepting the process for fear of dishonouring deceased loved ones [98,100-102]. Religious motivations can also be a hindrance to the process for other religions, such as Hebraism. The Halacha, in fact, prohibits disturbing the graves of Jews, even in the case of burials resulting from mass disasters or individual violence, also forbidding autopsies and samplings [103]. In other states too, such as Zambia, the possibility of moving the body from the primary burial place is not culturally accepted, so, if needed, autopsies are often conducted at the place of exhumation [104]. However, when required by judicial needs exhumations are normally conducted [98,102]. Identification should be considered a judicial necessity even when criminal proceedings have not been opened for the buried person.

It is important to emphasise that, in any case, the entire process of recovering skeletal remains must be carried out by competent persons who are aware of what can be found and what can be lost. In Québec, in the period 2000-2005, a study reported the presence of a forensic pathologist at the exhumation site for most cemetery exhumations and illegal exhumations [105]. Even the UNITED Operation (Unknown Names Identified Through Exhumation and DNA), started in Michigan, provided the presence of an FBI technician on site with anthropological training and several anthropologists for exhumation procedures with identification purposes [106]. For this operation anthropologists were usually only employed to assess the consistency of the biological profile of the exhumed with the individual reported buried and to select the most suitable bone remains for DNA sampling; skeletons were then reburied, except for those of infants [106]. In our opinion, definitive recovery of the bone remains is important to allow for more extensive study and avoid further taphonomic damage related to inhumation. In our case, the presence of forensic anthropologists at the time of exhumations, since 2016, proved crucial in the complete retrieval of all skeletal elements as well as in avoiding cases of commingling of remains. Also, the use of excavators for this type of process should be limited because of the danger of commingling and damage the remains. The use of trowels and brushes whenever possible is suggested [106].

All investigations should be conducted with full respect for the person and, where possible, the identified remains should be returned to the family for a respectful burial according to the person's wishes in life.

6. Conclusions

Greater awareness of what information can be lost in the identification process, when and how to prevent this from happening is desirable. The autopsy should only be one step in the identification process, not negligible but not sufficient on its own to provide all the necessary information. Instrumental investigations [24,59,89] before and a thorough anthropological examination [30,78–81] after the autopsy should become a routine part of the identification process. The exhumation of human remains, anticipated, if possible, should be seen as an important opportunity to add and, above all, not to lose information on skeletal unknown remains [91]. The information obtained for each of the individuals involved in this study will be entered into the national missing persons' information system "Ri.Sc" and on the laboratory's website page [107]. For individuals already entered, erroneous or incomplete data will be edited. More studies should shed light on the fate of unknown bodies in the world and the activities that are put in place to ensure that they can be identified. It should not be forgotten that unknown remains correspond to missing persons with even serious repercussions for those who stay and look for them [108].

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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