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OVERVIEW

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The role of forensic anthropological techniques in identifying America's war dead from past conflicts

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Abstract

The Scientific Analysis Directorate of the U.S. Department of Defense's (DoD) Defense POW/MIA Accounting Agency (DPAA) is a unique entity within the U.S. Government. This agency currently houses the world's largest, accredited skeletal identification laboratory in the world, in terms of the size of the scientific staff, global mission, and number of annual identifications. Traditional forensic anthropology is used for the formation of a biological profile (biological sex, stature, population affinity/ancestry, and age) as well as trauma and pathologies that may be compared with historical records and personnel files. Since World War II, various scientists associated with DoD have conducted base-line research in support of the identification of U.S. war dead, including, but not limited to, histology, the use of chest radiography and clavicle comparison, and statistical models to deal with commingling issues. The primary goal of the identification process of the Scientific Analysis Directorate is to use all available historical, field, and forensic methods to establish the most robust and defendable identification as scientifically and legally possible.

This article is categorized under:

- Forensic Anthropology > Age Assessment
- Forensic Anthropology > Sex Assessment
- Forensic Anthropology > Ancestry Determination

1 | INTRODUCTION

The recovery and identification of U.S. war dead has a long history stretching back to the U.S. Civil War (1861–1865). However, it was only in the mid-20th century, during the U.S. involvement in World War II (1935–1945), that the U.S. Department of Defense (*née* U.S. Department of War) made the endeavor more systematic and scientific with the employment of physical anthropologists in support of identifications. This marked an important deviation in general protocol but would be focused on temporary identification facilities associated within specific conflicts. However, in 1976, permanent laboratory facilities were opened on the island of O'ahu in the State of Hawai'i and established as the first permanent facility for the identification of U.S. war dead.

Currently, the Scientific Analysis Directorate (SA) of the Department of Defense's (DoD) Defense POW/MIA Accounting Agency (DPAA) is the largest employer of full-time forensic anthropologists and forensic archeologists in the world. The SA is the DoD's primary skeletal identification facility involved in the search, recovery, and identification of U.S. military personnel missing from past U.S. conflicts since World War II, including the Korean War, the Cold War, the Vietnam War, and Operation Enduring Freedom (or "the Gulf Wars"). Occasionally, the SA will recover and identify service members from previous conflicts, such as the War of 1812, the U.S. Civil War, and World War I, but the agency's U.S. Congressional mandate is restricted to wartime losses where hostilities have ceased from World War II and later. However, the SA can provide domestic and international humanitarian assistance as approved by the Secretary of Defense or request through the local combatant Commands (e.g., U.S. Indo-Pacific Command or USINDOPACOM). The legal mandate for the DoD to conduct this mission can be found in National Defense Authorization Act (NDAA) 1996 and its successor NDAA 2010. NDAA 1996 stated only three methods of legal identification of unknown persons were acceptable, but the rewrite of the law as part of NDAA 2010 reduced this to a single method. NDAA 2010, Paragraph B, Section 1513 (Definitions), Title 10, United States Code states: "…(B) the remains of the person are recovered and, if not identifiable through visual means as those of the missing person, are identified as those of the missing person by a practitioner of an appropriate forensic science…" Thus, the only current legal form of identification of U.S. missing personnel as of 2010 is through forensic analysis.

2 | SCIENTIFIC ANALYSIS DIRECTORATE

SA currently employs over 150 forensic anthropologists and support staff in three laboratories. The main facilities associated with the SA are located on Joint Base Pearl Harbor-Hickam (JBPHH), near Honolulu, Hawai'i (HI) on the island of O'ahu and generally focused on remains recovered from the Pacific Theater, including Southeast Asia and India. Another fully outfitted laboratory is located on Offutt Air Force Base in Omaha, Nebraska (NE) with personnel generally focused on remains recovered from the European-Mediterranean (including Africa) regions (although this facility was the primary facility for the analysis of the USS *Oklahoma* remains for the December 7, 1941 attack on Pearl Harbor, Hawai'i)). Three forensic odontologists are in the HI laboratory (currently three military dentists) and one civilian forensic odontologist is at the NE facility. Another laboratory facility is located at Wright-Patterson Air Force Base in Dayton, Ohio (OH). This facility currently houses two analysts conducting physical analysis of life support (survival gear, flight gear, uniforms, etc.) in support of the identification; additional Life Support Specialists are assigned to the HI laboratory for analysis and field operations. The laboratories are organized into several sections that are overseen by a senior laboratory manager/supervisor: Anthropology (HI/NE), Odontology (HI/NE), Life Support (HI/NE), Field Sciences and Material Evidence (HI/OH), Special Projects (HI), and Case Management (HI).

Investigation and recovery (excavation) is the primary source of remains. These include historically focused archival work as well as following investigative leads in the field to interview witnesses and detail landscapes related to battlefield losses or downed aircraft incidents (both terrestrial and aquatic). Each recovery or excavation is unique, but standard archeological procedures are used to record three-dimensional provenience of significant evidence and possible human remains. Details related to the investigation and recovery processes can be found in Emanovsky and Belcher (2012). Other sources of remains are unilateral turnovers to US Government officials as well as disinterment of unknown burials at various US military cemeteries throughout the world as well as local cemeteries in host nation locations (see Box 1). According to the DPAA website (www.dpaa.mil), over 81,000 are missing from WWII, the Korean War, the Vietnam War, the Cold War, and the Gulf War, and other conflicts. 75% of those losses are from the Indo-Pacific region and over 41,000 (primarily from WWII) are thought to be lost at sea (ship losses, aircraft losses at sea, etc.). These may not be recoverable with the current level of technology because exact locations of loss are not known and often the depth of the loss exceeds 1000 feet below the sea surface (Table 1).

The skeletal identification laboratories in HI and NE are accredited under the Forensic Accreditation program of ANSI National Accreditation Board (ANAB). The SA has been accredited under various accrediting bodies (all subsumed under the current ANAB) since 2003 and follows general guidelines of accreditation for federal forensic facilities. Accreditation follows the ISO/IEC 17020:2012 guidelines related to consistency, accuracy, and competency. Accreditation inspections are conducted on an annual basis with major inspections occurring every 5 years. A Lead Quality Assurance Manager (QM) is based in HI along with a Deputy QM Manager and a Deputy QM in NE. Quality assurance is based on the Laboratory Standard Operating Procedures (SOP), or Laboratory Manual, involving aspects of analysis, safety, security, and work products. More detailed discussions of the day-to-day operations within the SA can be found in Holland, Byrd, and Sava (2008).

BOX 1 Where do the remains come from?

Skeletal remains come from a variety of sources into the SA, but ultimately, they are recovered or found in vast areas of conflict in geographic Theaters of Operation from World War II, the Korean War, the Cold War, the Vietnam Conflict, and certain regions of the Global War on Terrorism. Specific materials are usually turned over to the SA from the following sources or activities:

- Unilateral turnovers. These are sets of remains that are considered from third-party sources, often private citizens.
- Recovery operations. These are remains that are recovered due to excavations as part of bilateral (or trilateral in some cases) agreements and excavations outside of the U.S., including Southeast Asia, Pacific Islands, Europe, etc. and are often referred to as joint operations or joint recoveries. Additionally, recovery operations may represent U.S.-only personnel, particularly within U.S. territorial boundaries.
- Partnerships. With the stand-up of the DPAA, a new Directorate, Partnerships and Innovations, exists to multiply opportunities for external partners (primarily academic institutions and nongovernmental organizations, such as Project Recover [Project Recover - Keeping Americas Promise] or History Flight [Home - History Flight, Inc.]) to conduct historical and scientific research, but also to coordinate field operations within the Indo-Pacific and Europe-Mediterranean regions.
- Disinterments. These sets of remains are associated with exhumations of remains buried in an unidentified or unknown status in U.S. National cemeteries or host nation cemeteries around the world.

3 | FORENSIC ANTHROPOLOGY'S ROLE IN PAST CONFLICT ACCOUNTING

The biological profile is the crux of the identification process for deceased U.S. military personnel and the most common contribution of the forensic anthropologist. The biological profile provides the skeletal data that can be compared to the Individual Deceased Personnel Files (IDPF) or the Official Military Personnel File (OMPF). The biological profile typically encompasses the biological sex, the skeletal age, the height or stature, and the population affinity. Other individuating information such as healed fractures or skeletal pathologies also may be included in the personnel files. Additionally, skeletal trauma is analyzed to determine the consistency of trauma to the death incident. The contents of the personnel files vary according to time period or conflict but may have records related to induction into the Armed Services (medical physical and dental examinations), dental charts of varying quality, dental radiographs, chest radiographs (usually to assess for tuberculosis), circumstances of loss, etc.

The SA uses a variety of methods to attempt the most holistic identification as possible. The biological profile can allow the segregation of possible individuals (a "short list" of candidates) to examine against other nonbiological evidence (such as identification tags or cards, bracelets, wedding bands, etc.). Often the positive identification is based on the odontological (dental) radiographs or certain types of DNA results. This holistic method is characterized by a "Venn" diagram in Figure 1 which represents the totality of the SA identification process, including historical analysis, forensic archaeology, material evidence analysis, forensic odontology, DNA analysis, and forensic anthropology. It should be noted that due to the specialty analyses, DNA as performed by the US Armed Forces Medical Examiner's Office at Dover Air Force Base is not discussed in this current article. This is not to say that this analysis is unimportant to the identification of US war dead.

Many of the methods used to determine the biological profile are "tried-and-true" methods within the discipline and follow procedures one may see in any introductory or advanced forensic anthropology textbook (e.g., Byers, 2016; Christensen, Passalacqua, & Bartelink, 2019; Langley & Terigsini-Tarrant, 2017). The analysis of skeletal materials spans U.S. conflicts from the mid-20th century to the present; thus, specific protocols and analytical techniques are dependent on the various historical populations. For pre-1960 populations (World War II-era 1940–1946 and Korean War-era 1950–1953), standards developed by McKern and Stewart (1957) are used to determine stature and age. Post-1960 populations use a variety of standards to determine stature and age, more coincident with the modern forensic data bases and

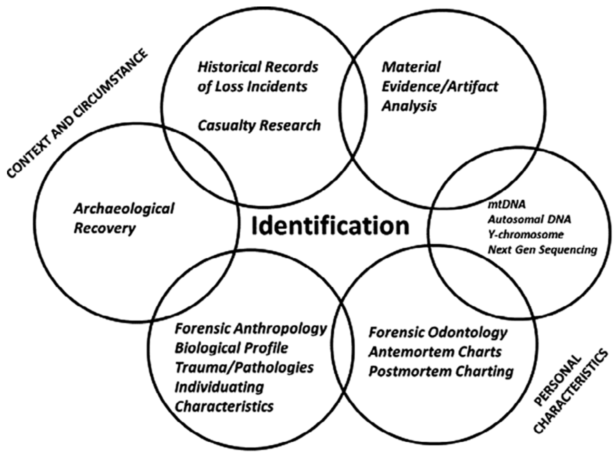

FIGURE 1 Overlapping sequences of research and lines of evidence that focus on the identification of a missing U.S. service member

techniques. The basic biological profile provided by staff forensic anthropologists includes age, biological sex, population affinity or ancestry, stature, assessment of trauma, and discussion of other individuating features of the human skeleton.

3.1 | Age

Age estimation refers to the age-at-death of an unknown individual. The SA uses a variety of methods focused on dental development, degeneration of the *os coxae* and sternal rib ends, epiphyseal growth and fusion, and maxillary sutures. In most methods, an age estimate is derived from macroscopic observations, primarily from examining bone growth or degeneration, dental eruption, and tooth crown and root formation (however, see Algee-Hewitt, 2017 for a full discussion on age estimation in forensic anthropology and inherent assumptions).

3.1.1 | Dental remains

Dental remains can assist in age estimation using procedures typically confined to dental eruption and calcification. The dental age and age-interval are determined by the closest match between the reference standard and the observed development. In terms of dental development, tooth formation begins with the cusps and terminates in the closure of the root apices; most often these are determined from radiographs and focused on the third molar using the dental calcification tables and figures of Mincer et al., (1993). Another system is the eruption pattern and timing of the gingival emergence of teeth. To determine gingival eruption, teeth are examined, either macroscopically or microscopically for either occlusal or interproximal wear. The proposed eruption pattern then is compared to a known standard eruption table (such as American Dental Association, W300 Chart, W368 Plaque, *Development of Human Dentition*).

3.1.2 | Skeletal remains

Various regions of the skeleton can be used for age estimation; most importantly, these areas are characterized by the surfaces of nonmovable joints that systematically change or degenerate with age. General procedures include macroscopic observation of developmental stages; the selection of an appropriate standard (dependent on sex and date of death [pre- or post-1960 development]); and recording of the estimated age that is the closest match between the remains and the published standard or exemplar. The most important methods are in the pelvic girdle utilizing the pubic symphyses and auricular surfaces. For the pubic symphyses, various cast sets and graphic exemplars are used (Brooks & Suchey, 1990; McKern & Stewart, 1957).

The auricular surface corresponds to the surface of the sacro-iliac joint of the ilium. Auricular surfaces can be evaluated for age-at-death using a variety of methods (e.g., Buckberry and Chamberlain, 2002; Lovejoy, Meindl, Pryzbeck, & Mensforth, 1985; Osborne, Simmons, & Nawrocki 2004). Osborne et al. (2004) is the preferred method for the

SA. When the Lovejoy et al. (1985) system is used, the six phase age intervals described by Osborne et al., 2004 is used. For the fourth rib, the Işcan et al. (1984, 1985) cast sets of known age phases for White females and males are used in the SA, regardless of population affinity. If any other rib is used, the analyst is required to consult Yoder et al. (2001).

The sutures of the maxillary palate fuse in an age-related sequence. Age estimation using the maxillary sutures follows the Mann, Jantz, Bass, and Willey (1991) method. This method requires macroscopic examination of the sutures of the palate (incisive, posterior median palatine, transverse palatine, including the extension of this suture in the greater palatine foramen), and the anterior median palatine. Based on this examination, a missing individual is assigned an age using Figure 2 in Mann et al. (1991: 783) with the individual's age estimate based on the suture with the oldest age estimate.

3.1.3 | Growth & development

Early growth and development of cranial and postcranial elements are well documented and occur in a predictable chronological sequence. During the final stages of development, epiphyses fuse to the diaphyseal shafts at and are a relatively accurate means of estimating skeletal age in individuals under the age of 25 years. In all bones, the stage of development can be observed macroscopically. Specific techniques used within the SA include McKern and Stewart (1957) and Bass (2005).

Although not in typical military-based populations, Fazekas and Kósa (1978) provides metric data for fetal remains (along with summary data published in Schaefer, Black & Scheuer, 2009). Various measurements (typically lengths and breadths) are described and can be utilized to place the unknown specimen in a fetal age category (e.g., 2.5 lunar months). These tables are used to determine suitable age ranges and are preferably reported in prenatal weeks. Additionally, Cunningham, Scheuer, and Black (2016) provide a comprehensive guide to aging skeletons of juveniles and is consulted for individuals believed to be 17 or younger.

3.2 | Biological sex

Biological sex determination is performed by standard nonmetric and/or metric assessment procedures that examine dimorphic characteristics of the pelvis, cranium, and postcranial skeleton. Estimation of sex is typically based on two tenets: (1) the generalization that males are larger (more robust) and display more prominent muscle attachments than females; and (2) the differences between sexually dimorphic features, usually found on the pelvic girdle and the skull.

When the pelvis and skull are missing or incomplete, postcranial remains, usually the humerus and the femur, may be used. In cases where other postcranial elements are used, the procedure used is documented and referenced. Dimorphic characteristics and features are often relative, requiring the analyst to draw upon professional training and knowledge of human osteology. Various metric techniques (e.g., discriminant function analysis in FORDISC) may be used for various skeletal elements but given that the remains of many missing service members are highly fragmentary and specific landmarks can be ill-defined (such as with the *os coxae*), nonmetric techniques may be more reliable.

The *os coxae* is typically the most common set of skeletal elements for the determination of biological sex. Nonmetric estimation is generally done with an initial assessment of the overall size and morphology of the pelvic girdle (Buikstra and Ubelaker, 1994; Phenice, 1969). Walker (2008: 45) provides empirical probabilities of being male for a given score of each cranial trait based on these scoring systems. Buikstra and Ubelaker (1994) also have scoring criteria related to the skull, including the nuchal crest, mastoid process, supraorbital margin, glabella, and mental eminence.

In general, metric analyses for ancestry and sex estimation at the DPAA focuses on craniometrics and the use of FORDISC and/or the (hu)MANid program (Berg & Kenyhercz, 2017a; Jantz & Ousley, 2005). FORDISC sex estimates are based on the Forensic Data Bank at the University of Tennessee.

Postcranial indicators include: the humerus (Rogers, 1999), clavicle (Rogers, Flournoy, & McCormick, 2000), and femur (Stewart, 1979, p. 210). Nonmetric determinations, based on general size and robusticity, are made visually and most are relative, requiring the analyst to draw upon their professional training and knowledge of human osteology. Estimates, based on the Forensic Data Bank at the University of Tennessee, can be done using FORDISC.

FORDISC can determine sex using a combination of various postcranial elements (Jantz & Ousley, 2005). The osteometrics used in FORDISC are described in Moore-Jansen et al. (1994, pp. 63–71); although the more recent *Data*

Collections Procedure Manual 2.0 by Langley et al. (2016) defines some measurements differently and should be consulted but as of this date, it is not part of the current SA Laboratory Manual. In instances where FORDISC may be ambiguous or the analyst may need additional results to strengthen sex estimates, various other metric methods using the cranium, humerus, femur and proximal tibia are available. Among these are Jantz & Moore-Jansen, 1988 (multiple bones) and Holland, 1991 (proximal tibia).

3.3 | Population affinity

Population affinity or ancestry refers to a group of people who historically shared a geographic origin and, thus, still share some common genetic or physical expression related to that origin. The human genotype, along with environmental and cultural factors, correlates to systematic and discernible patterns of phenotypic variation. Therefore, ancestry assessment (as practiced at the SA) is the classification of a set of remains into one of several broad geographic groups based on shared skeletal morphology. Currently, it must be noted that the definitions and use of the concepts of ancestry are a point of debate within the discipline. Readers should consult Bethard and DiGangi (2020) and Stull et al. (2020) for current discussion regarding terminology and a call for new analytical techniques.

For identification purposes, these ancestry categories used in the SA are broadly consistent with "racial" assessments from the military antemortem records. A final assessment of ancestry classifies the remains into one of three broad, geographical ancestral groups (i.e., African, Asian, or European). A modifier to this final assessment (e.g., probable) is left to the discretion of the analyst but must be documented in the accompanying bench notes. Analysts also may choose to include a parenthetical qualifier to the overall assessment of ancestry. This may include, but is not limited to, distinctions of ethnicity, such as European (White); European (Hispanic); African (Black); Asian (SE Asian); Asian (Hispanic); and Asian (Pacific Islander). If an analyst cannot make a final determination, the remains are classified as "Indeterminate." A finding of indeterminate can reflect insufficient data, ambiguous results, or both.

Ancestry assessment is primarily conducted by analyzing the variability of morphoscopic and osteometric characteristics of the cranium and mandible. Secondarily, postcranial skeletal morphometrics may be used to assess ancestry. The SA uses five nonmetric or morphoscopic cranial traits as defined by Hefner (2009): bone morphology; bony feature morphology; cranial suture shape; presence/absence data (e.g., postbregmatic depression); and feature prominence/protrusion. Optimized Summed Scored Attributes (OSSA) utilizes six cranial nonmetric traits (Hefner & Ousley, 2014) and is appropriate as a test to separate American Whites and Blacks. OSSA is a nonparametric method that compresses morphological variation into two classes using the anterior nasal spine, the inferior nasal aperture morphology, the interorbital breadth, the nasal aperture width, the nasal bone structure, and the postbregmatic depression. The shape and size of teeth appear to be strongly with genetic variation in human populations and the expression of specific dental traits and morphology may assist in assessing population affinity (e.g., Edgar, 2009; Hanihara, 1967; Irish, 1997; Rhine, 1990; Scott and Turner, 1997; Turner 1990; Turner et al., 1991).

Using the standard osteometric data collection procedures (Buikstra and Ubelaker, 1994; Moore-Jansen et al., 1994), analysts may use craniometrics to conduct ancestry assessment using FORDISC (Jantz & Ousley, 2005). (hu)MANid (Berg & Kenyhercz, 2017a) can also be used for ancestry. Other methods include Holliday and Falsetti (1999), a discriminant function analysis that compares African American and European-American males and females on seven postcranial methods; Stewart (1962) which provides a method of discriminating between White, Black, and Native American femora by examination of the anterior curvature and torsion; Gilbert and Gill (1990) which provide a sectioning point between Whites and Blacks versus Native Americans based on the subtrochanteric measurements of the proximal feature; and, finally, Wescott (2005), who analyzed the shape of the proximal femur to determine ancestry of Native American, Polynesia, American Black, American White, and Hispanic groups using the platymeric index. Wescott (2005: 288–289) does caution that each group displays considerable within-group variation making discrimination between populations difficult (see also Tallman & Winburn, 2011).

3.4 | Stature

Stature estimation procedures at the SA can be divided into two types: (1) those designed to provide point estimates (a stature) or estimation of the most probable stature of the unknown individual; and (2) those that perform formal comparisons between bone measurements from unidentified remains and statures of candidates for the identification. Stature estimation requires the analyst to consider the following: the "type" of stature targeted by the test; the appropriateness of the reference data set used to generate statistical models; the age of the person whose stature is being compared; the most desirable estimation model to use in the comparison; and the proper statistical treatment of the reference data during its comparison with an unidentified specimen.

Specific test methods include using FORDISC, whose models are applicable to American Black and White females and males. These models are based on long bone lengths and include those of Trotter and Gleser (1952, 1958) as well as those from the Forensic Anthropology Databank (FDB). The FDB models are used when working with recent deaths, as in criminal cases, or post-1960 deaths. The Trotter and Gleser models are generally more appropriate for cases involving the identification of military personnel lost in past conflicts, particularly pre-1960. Additional methods included those based on the sex and ancestry assessment as well as the adjustments based on estimated age of the population (e.g., Choi, Chae, Chung, & Kang, 1997; Genovés, 1967).

3.5 | Trauma

Trauma is injury or disruption of living tissue by an outside force. The identification of trauma is important and allow observations to be correlated to the conflict incident (battlefield, aircraft crash incident, etc.) as most of the U.S. war dead have died in combat. Trauma is classified based on the timing (antemortem, usually distinguished by healed bone; perimortem, usually at or around the time of death) as well as type of trauma, including sharp force, blunt force, deceleration impact events (i.e., aircraft crashes), and projectiles (such as bullets and shrapnel).

3.6 | Individuating characteristics

Individuating characteristics are unique to each individual and can include pathological conditions, anomalies, and indicators of stress that manifest in skeletal tissues. Such traits found within skeletal and dental remains have a potential to directly contribute to circumstantial or even positive identification. Recognizing these traits is dependent on the overall completeness and preservation of the evidence. However, the SA uses two criteria for individuation: (1) relative rarity (the more uncommon the trait, the more potential it may have in contributing to identification); and (2) the relative likelihood of a trait being recognized in vivo and subsequently documented. For any type of individuating characteristics, it is best to not stray beyond your level of expertise, particularly with pathologies. Analysts are expected to consult pertinent literature such as Aufderheide and Rodríguez-Martín (1998), Di Maio (1999), Galloway (1999), Mann and Hunt (2012), and Ortner (2003). The most common mistakes in conducting a differential diagnosis are over-reaching, too narrowly restricted, and/or unsupportable results. The biological profile is an important place to start with the identification of missing service members. However, due to large numbers of service members having similar basic profile (male, White or Black, early 20s in age, around 5′10 to 6′ tall), dental radiographs and charting are essential in most identifications.

4 | FORENSIC ODONTOLOGY'S ROLE IN PAST CONFLICT ACCOUNTING

Odontology's primary role in the past conflict effort deals with the rendering of an opinion as to the identification of unknown remains. These cases can be extremely challenging, dealing with varying quality and quantity of dental records, interpretation of dental records/treatment entries, the potential for erroneous entries/transcription errors, the use of multiple numbering systems (military and civilian), and the lack of antemortem (before death) dental radiographs. The comparison of an individual's dental radiographs with those of the remains is the most reliable method of identifying a set of unknown remains (Luntz, 1977). Except for the Vietnam War and subsequent conflicts, dental radiographs are nearly nonexistent in the personnel files for those lost in previous wars.

Antemortem dental evidence for U.S. service members from the Korea War and WWII include generic charting (extraction patterns with or without generic restorations), dental records with or without treatment entries, and detailed chartings of restorations and extraction patterns (Shiroma, 2016a). Periodically present in their personnel files were civilian dental records. An antemortem profile is developed from the available dental records.

[redacted] Dental comparison of X-[redacted] to the casualty list.					
Tooth #	X-[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
1	X	X	V	V	X
2	O-S, O-S, L-S	O-S	O-S	O-S, O-S, L-S	O-S
3	MO-S (or G), O-S (or G), F-S (or G)	X (documented)	X (documented)	MO-G, O-S, L-S	X
4	V	V	V	V	V
5	V	V	V	V	V
6	V	V	V	V	V
7	V	V	V	V	V

FIGURE 2 Example of postmortem (the "X-file number") to Antemortem (the servicemembers-redacted) dental chart comparisons resulting in exclusion of three candidates with one candidate remaining; green cells are concordant, yellow are explainable discrepancies and red are unexplainable discrepancies (F, facial; G, gold; L, lingual; M, mesial; O, occlusal; S,silver; V, virgin; X, extraction)

The dental remains are examined, and a postmortem (after death) charting is performed, radiographed, and photographed. A profile then is developed from the findings of the postmortem examination (Figure 2). The postmortem profile is compared to the antemortem profiles of individuals lost in an incident. The use of a dental computer software with the ability to compare the dental profile of a set of unknown remains quickly and efficiently to the antemortem dental database of individuals lost in previous conflicts is essential in incidents with many casualties. Examples of this type of program/software include WinID and the Centralized Accounting and Repository Information System (http://www.abfo.org). The result of the computerized comparison is a list of ranked individuals. The odontologist should carefully review/analyze the results, as the program provides a list of possible candidates and not a recommendation for identification to a set of remains (Shiroma, 2016a).

A review of the Forensic Odontology Reports written at the SA revealed 69% of the antemortem/postmortem comparisons included explainable discrepancies. Explanations for the various types of discrepancies include undocumented treatment, third molars charted incorrectly as missing, differing opinions by antemortem and postmortem examiners regarding the specific teeth present and missing (e.g., molar and premolar patterns), errors in treatment documentation, and differing opinions of antemortem and postmortem examiners regarding specific surfaces restored (Shiroma, 2016b). Thus, the odontologist must always consider variations while interpreting restorative care/extraction patterns in the remains and in an individual's dental record and have knowledge that charting or treatment documentation errors in an individual's dental record are always possible.

Based on the interpretation of the observed characteristics/findings and antemortem/postmortem comparisons, one of the following opinions may be rendered by the forensic odontologist: positive identification, probable identification, possible identification, exclusion, and insufficient evidence. The odontologist should base their opinion on the strength of the antemortem/postmortem comparison, considering not only the number of concordances, but also on distinct restorative care and extraction patterns (Shiroma, 2019).

5 | NEW METHODOLOGIES FOR PAST CONFLICT ACCOUNTING

The SA is not a static entity but is constantly working to validate traditional methods or create new methods and technologies that will support current methods as well as focusing on different biological components that can be preserved in human skeletal materials. This requires a scientific staff that is trained in current research methods and leadership that is interested and supportive of research and funding related to new techniques and the use of new equipment. The following vignettes represent four major areas of research endeavors that have been supported by the Agency and its predecessors: chest radiograph comparisons, histomorphology, isotopic analysis, and osteometric sorting for commingled remains.

5.1 | Chest radiographic comparison

During the early efforts of CILHI and JPAC to identify disinterred remains, it was found that the Korean War Unknowns had been treated with formaldehyde during the initial processing of the remains. Unfortunately, this inhibited DNA extraction and analysis (although it should be noted that mtDNA sequencing is now possible with the development and the use of Next Generation Sequencing). To mitigate these difficulties with identification, anthropologists at the CILHI, the JPAC, and now the DPAA developed a Chest Radiograph Comparison program to effect individual positive identifications without the use of DNA.

Upon induction, many service members were screened for tuberculosis using chest radiography. These antemortem radiographs are available for approximately 72% of those missing from the Korean War (Stephan et al., 2014). Once an Unknown burial is disinterred from a cemetery, anthropologists can compare the antemortem chest radiographs of those on the short list of candidates to postmortem radiographs of the remains. Specifically, the clavicles and upper vertebrae (the third cervical vertebra through the third thoracic vertebra) are radiographed in a position representative of standard chest radiography. The morphology of these skeletal elements is examined for concordances in shape and similarities in cortical and trabecular densities to assess if an individual is a positive match or can be excluded (Figure 3). A blind test of this method has demonstrated an accuracy of 88% for trained examiners, which included the use of highly eroded remains (Stephan, Winburn, Christensen, & Tyrrell, 2011).

In cases where the historical information for an unknown individual is sparse or inaccurate, short lists of candidates may be obtained through computer-based analysis of radiographs (Stephan et al., 2014). This *Clavicle Matching Program* uses elliptical Fourier descriptors to compare the outlines of the clavicles from the remains captured through 3D scans to the clavicle outlines traced from antemortem chest radiographs. This comparison produces a ranked list of service members that are most like the remains. The radiographs of the most likely candidates can then be visually compared to the postmortem radiographs to assess the possibility of a match. Using this method, correctly matching individuals have been found in the top 10% of the comparison sample 70% of the time (D'Alonzo, Guyomarc'h, Byrd, & Stephan, 2017). Overall, the use of chest radiographs has increased the capacity and timeliness of identifications.

5.2 | Histomorphology

The DPAA recoveries typically take place many years after the initial incident, allowing time and environmental factors to affect bone preservation. After death, bone can undergo significant changes due to its (burial) environment or due to

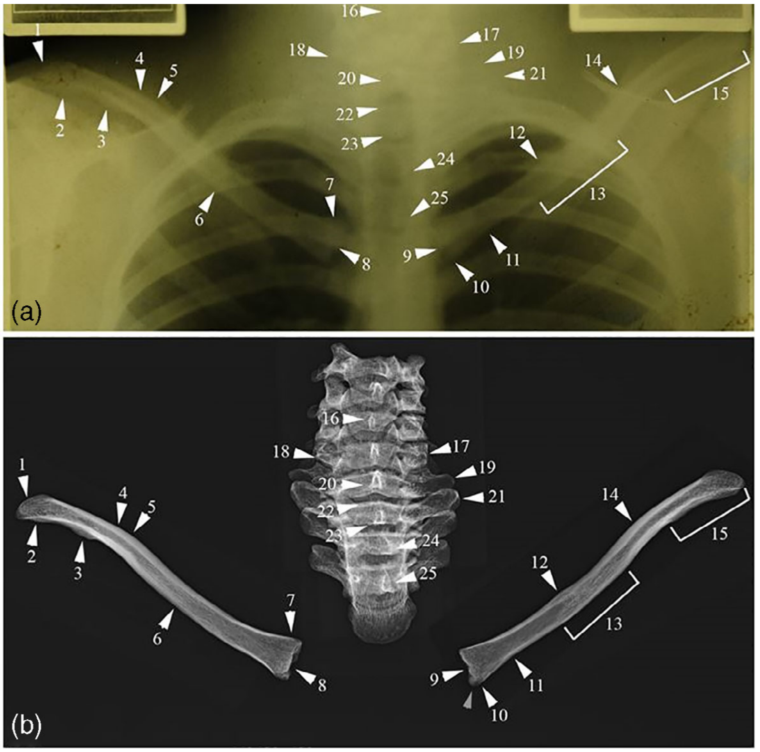

FIGURE 3 Example of a chest radiograph comparison. (a) Antemortem radiograph, (b) postmortem radiograph. The arrows and numbers correspond to points of concordance detailed in the chest radiograph report (CXR)

the loss incident (high impact forces due to a plane crash, heat alteration, etc.). These alterations lead to loss of surface detail, and fragmentation, with a loss of characteristics that inform an anthropologist on the type of bone or even the bone's original species. Additionally, nonhuman bone material may be present on the sites the DPAA investigates, such as animal bone stemming from butchery or food preparation refuse (Benedix, 2004). Commingling of fragmentary human and animal bone, and the question of how to tell human from nonhuman animal bone is a common one (Benedix, 2004). DNA is a powerful method capable of delivering specific and accurate answers to this question but can be time consuming and expensive. This volume of undetermined fragmentary remains at the DPAA necessitates a relatively quick and inexpensive method to distinguish nonosseous material and animal bone from potential human osseous material. Observing bone microstructure (histomorphological study) of bone has been proven as a reliable triage method to differentiate some nonosseous material and nonhuman animal bone from potential human bone (Hillier & Bell, 2007).

Bone is a complex tissue consisting of a mineral and an organic phase, which is organized as mineralized collagen fibrils on the microscopical scale. These mineralized fibrils can be arranged in different histological bone types depending on age, speed of growth, and health (Currey, 2002). The classifications of histological bone type that are important for the question of species differentiation are lamellar bone (a plywood-like, layered organization) and woven (a fibrous organization) bone (Francillon-Vieillot et al., 1990).

Large mammals, such as horses or cattle, grow extremely fast. Lamellar bone is formed slowly, while woven bone can be laid down quickly. A compromise is found in fibrolamellar bone (a combination of woven and lamellar bone), which can grow fast in a radial direction (Currey, 2002). Bone is a living tissue and thus requires blood supply that is provided by a network of blood vessels permeating the bone through canals. The organization of vascular canals in a plexiform manner as well the presence of fibrolamellar bone is an indication of fast growth speed and therefore animal bone (Cuijpers, 2006). Bone microstructure changes with aging, as the initially formed bone structure (primary bone) is replaced with secondary bone structure (Francillon-Vieillot et al., 1990). Secondary bone consists of lamellar bone arranged in concentric rings around vascular canals (osteons). In secondary bone, where remodeling has removed some or all primary bone features, a pattern of osteon banding is indicative of nonhuman animal bone (Mulhern & Ubelaker, 2001). When neither plexiform bone nor osteon banding are present, a histomorphology is considered "inconclusive" and the DNA bone sample is forwarded for DNA analysis as per the DPAA protocols (Tersigni-Tarrant and Byrd, 2013).

To observe bone histomorphology, thin sections of bone need to be made. At the DPAA histology bone samples are taken concurrently with DNA sampling using a Dremel multitool. The histology samples are documented, then embedded in a two-component epoxy resin (Buehler Epo thin) and semi-thin sections (80–100 um) (Tersigni-Tarrant and Byrd, 2013) are cut using a Buehler Isomet 5000. The sections are then mounted on glass slides and analyzed using normal and (circular) polarized light. The use of histomorphology permits about 40% of the bone samples to be triaged as nonhuman animal or nonosseous (Tersigni-Tarrant and Byrd, 2013) allowing such material to be removed as non-evidentiary, significantly alleviating the DNA case load for species determination.

Bone histology within forensic anthropological context can be used for other purposes as well, (e.g., histological age determination, identification of taphonomic processes; see Streeter, 2011 for a discussion of the analysis of age using histological analysis). Research is currently ongoing to explore the practical implementation of these additional uses of histomorphology in the identification process at the DPAA. Another analytical strategy is looking at the isotopic composition of certain hard tissues in a way to reduce case load and create short lists of possible candidates for a missing person.

5.3 | Isotopic analysis

Not every tool in a forensic anthropologist's toolbox can be applied in each case and not every test result provides conclusive evidence to support an identification. Therefore, creation, innovation, and new approaches are often sought out to deal with complex cases. Isotope testing can be an extremely helpful avenue in these instances. Used for decades in bioarcheology (Schoeninger & Moore, 1992) and other forensic science fields (Meier-Augenstein, 2018), applications of isotope testing in forensic anthropology have increased rapidly in the last 30 years (Chesson et al., 2020a). Here, we present a short synopsis of the technique and its utility in human identification; for a more comprehensive review, particularly in relation to post-conflict applications, please see Chesson and Berg (2021).

5.3.1 | Isotopes

Isotopes are different forms of a chemical element that vary in neutron number, and, thus, mass. As an example, most atoms of carbon (C) contain six neutrons and six protons within the nucleus, with six electrons bound to the nucleus. The shorthand for this isotope form of carbon is ^{12}C, representing the sum of neutrons and protons. An additional neutron is present in the rare isotope form ^{13}C while two additional neutrons are present in ^{14}C. Both ^{12}C and ^{13}C are stable isotopes meaning the atoms do not undergo radioactive decay. In contrast, ^{14}C is a radioactive isotope and undergoes decay. All isotope forms of carbon can participate in the same biological and chemical processes, but reactions are affected by the mass differences between isotopes.

5.3.2 | Records of life history

Stable isotopes are found in all human tissues, including bone, teeth, hair, and nails. The isotopic records within these tissues can be used to reconstruct an individual's life history. Carbon and nitrogen isotopes provide information on diet. Plants using different photosynthetic pathways discriminate against $^{13}CO_2$ to varying degrees, with the so-called C4 plants having a greater abundance of ^{13}C isotopes in their tissues as compared to C3 plants (Tipple & Pagani, 2007). The ripple effect for humans comes via food preference: individuals that consume more C4 plants—such as corn and sugar cane—have higher carbon isotope values in their body tissues (denoted $\delta^{13}C$) than individuals who consume mainly C3 plants, which include rice, wheat, and most fruits and vegetables (Valenzuela, Chesson, Bowen, Cerling, & Ehleringer, 2012). The prevalence of C4 and C3 plants in a person's diet also is influenced by the feed of animals that they consume as meat, eggs, or dairy products. Nitrogen isotopes record information on the consumption of animal protein, with animals at the end of longer food chains having higher nitrogen isotope values (denoted $\delta^{15}N$) (O'Brien, 2015). This is due to isotopic fractionation processes that take place with each "step" in a food chain. A terrestrial food chain may contain only three trophic levels (e.g., plant, animal, human), while a marine food chain can contain more (e.g., phytoplankton, zooplankton, small fish, game fish, and human). When discussing human dietary variations, carbon and nitrogen isotopic records are generally measured and interpreted together to form a more complete understanding of an individual's diet.

In contrast to the dietary discrimination possible through analysis of carbon and nitrogen isotopes, isotopes of oxygen and strontium provide geolocation information. The primary source of oxygen isotopes (denoted $\delta^{18}O$) in an individual's tissue is from consumed water; the isotopic composition of water varies across landscapes due to rainfall patterns and geological features. In general, lower $\delta^{18}O$ values of water are found inland at higher elevations and higher latitudes while higher $\delta^{18}O$ values of water are found at lower elevations, typically along coastal margins, or mid- to low-latitude plains/deserts (Bowen et al., 2007). The systematic pattern in water $\delta^{18}O$ values is demonstrated in Figure 4, as an isotope landscape or "isoscape" map. Spatial variation in strontium isotopic composition is related to bedrock age, with regions of older geology having higher strontium isotope values (denoted $^{87}Sr/^{86}Sr$) in the environment (Bataille & Bowen, 2012). Strontium isotopes are recorded by human tissues via diet, as plants incorporate strontium isotopes from the soil into their tissues and are in turn consumed by animals. Like oxygen, strontium isoscape maps can be constructed for local or regional geologic areas.

The "snapshot" of time recorded by isotopes varies from tissue to tissue. Generally, permanent dentition enamel is formed during early childhood and adolescence. Therefore, the isotopic analysis of a permanent tooth can provide information on the diet and geolocation of an individual before adulthood. This record is static; enamel does not remodel and it will not change over time. A dynamic dietary record can be found in bone tissues since bone slowly remodels throughout life. Depending on the bone sampled, an averaged signal representing perhaps 10–25 years of a person's life can be obtained (Hedges, Thomas, & O'Connell, 2007). If available, hair and nail tissues can provide more recent information on diet and geolocation, representing times closer to weeks or months since incorporation (Mancuso and Ehleringer, 2018, 2019; Thompson, Wilson, & Ehleringer, 2014).

5.3.3 | Isotope testing and human identification

The information provided by isotope testing about an unknown individual's diet and geolocation can aid in the process of identification, typically by excluding possibilities (as opposed to directly identifying a person). Because dietary

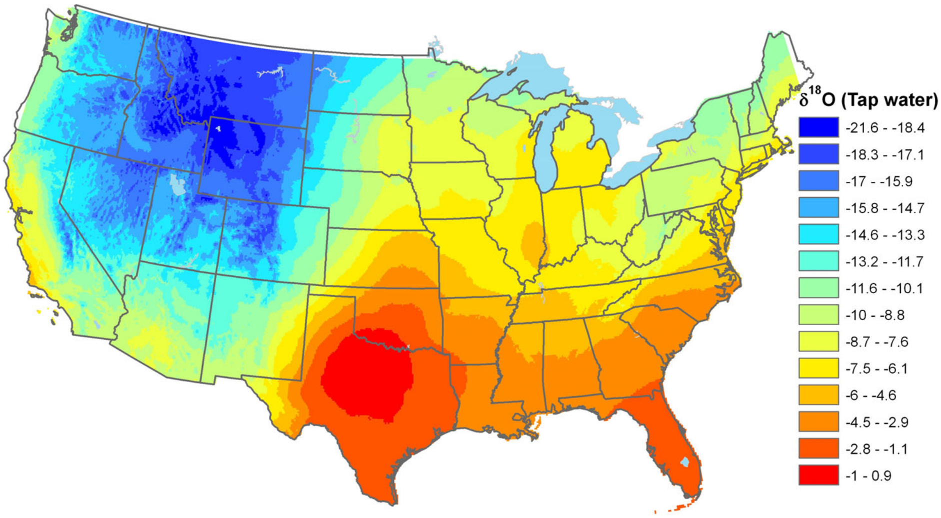

FIGURE 4 Groundwater isoscape for oxygen isotope ratios of tap water in the contiguous USA (from Bowen, Ehleringer, Chesson, Stange, & Cerling, 2007)

preferences and food availability vary between populations, the $\delta^{13}C$ and the $\delta^{15}N$ values of a tissue can be used by the DPAA to determine whether an individual was more likely a resident of southeast Asia—and thus a heavy consumer of rice—or a U.S. American who consumed more corn and sugar cane. An online tool called Isolocate (Berg & Kenyhercz, 2017b) has been developed to assist in population classifications; it's freely available at www.anthropologyapps.com. Isolocate uses custom discriminant functions to compare isotope test results from unidentified remains to reference population databases and produces easy-interpret-statistics and graphics as outputs. It contains populations applicable not only to DPAA casework, but also to casework in the forensics world in general.

Once remains are classified, the likely U.S. remains can then be submitted for follow-on anthropological analysis and DNA testing, saving time and money in the identification process at the DPAA. Similarly, in the civilian sector, dietary variations can be used to differentiate residents of South and Central America versus U.S. Americans in investigations of unidentified border crossers South and Central Americans rely even more heavily on C4 plants than U.S. Americans while U.S. American tend to eat more meat (Bartelink & Chesson, 2019). For isotopes of oxygen and strontium, the geolocation information that is provided by those elements can be compared to an individual's home of record in their personnel files to substantiate (or refute) a hypothesis regarding identity. In these ways, isotope testing at the DPAA has contributed to the identifications of seven missing U.S. military members to date (e.g., Chesson et al., 2020a; Holland, Berg, & Regan, 2012); at least 10 additional identifications using isotope test results are currently in progress.

Finally, the isotopic records found in tissues may prove useful in the segregation of commingled remains as measurands that complement osteometric data (see below). For this application to be successful, large and reliable datasets on intra-bone and intra-person isotopic variation will be needed and are not (yet) available. However, preliminary investigations suggest the utility of isotope testing in the analysis of commingled remains could be significant (Berg, Bartelink, Yuryang, Shin, & Chesson, 2019; McCormick, Berg, & Chesson, 2020). These innovative applications of isotope testing—to differentiate populations based on dietary variations, to compare with home of record, to separate commingled remains—provide data useful for human identification not easily obtained by other analytical methods.

5.4 | Osteometric sorting for commingled remains

To deal with commingled (mixed) remains of human remains, researchers with the DPAA's predecessors (US Army CILHI, JPAC) had investigated the use of various osteometric measures to reassociate skeletal elements into individuals. This is an essential step to complete the biological profile as well as assess the cause and manner of death. While not the only tool used to segregate mixed assemblages into individuals (age, taphonomy, articulation, and visual pair matching), the size of individuals based on osteometric sorting is an important tool to segregate these individuals (e.g., Byrd, 2008; Byrd & LeGarde, 2014). The DPAA has developed a series of protocols and reference data bases that

TABLE 1 Current U.S. military losses estimated as of April 2, 2021

Congressionally mandated conflicts	Losses
World War II (1941–1945)	72,491
Korean War (1950–1943)	7564
Vietnam War (1963–1976)	1584
Cold War	126
Gulf Wars	5
El Dorado Canyon (Libya)	1

Source: www.dpaa.mil; Accessed April 3, 2021.

are to be used in tandem with the Forensic Databank at the University of Tennessee-Knoxville (see Moore-Jansen et al., 1994) to segregate individuals from these mixed assemblages. Specific osteometrics have been developed and are part of the osteometric suite used at the DPAA.

Since the early 2000s, large, commingled sets of remains appear to be the norm for case work at the DPAA. These come from a variety of sources (see *Sidebar: Where do the remains come from?*), but major sources include disinterments from large mass casualty events (the World War II bombing of Pearl Harbor, HI and the analysis of the remains from battleships like the USS *Oklahoma*, the USS *West Virginia*, and the USS *California* as well as unilateral remains from North Korea) as well as more typical commingled remains from aircraft incidents with large crew complements, such 5 to 10 (or more) individuals on a World War II-era bomber.

6 | CONCLUSION

The DPAA SA Laboratories are, in many ways, cutting edge in terms of processing a large amount of human remains to gain scientifically, legally, and forensically valid identifications of missing U.S. military personnel lost during wartime. The SA has built upon standard biological profile determination and forensic odontology to include avant-garde science and validation of numerous methods of analysis to allow the analysts to have a wide variety of techniques for determining identifications. In the end, the goal is to present the evidence and analysis to the resident DPAA Armed Forces Medical Examiner who will approve and submit a legal identification. It is through this process and using as many lines of evidence as possible that the identification report is created. All forensic reports are peer-reviewed within the laboratory and the entire identification and summary is peer reviewed within the Armed Forces Medical Examiner system. The entire identification report then is presented to the casualty's primary next-of-kin for review before a decision is made on the acceptance of the identification and the disposition of the remains (i.e., how and where the identified person will be buried, usually with full military honors).

The development of new scientific techniques for identification has always been an important part of the military identification process. Since World War II, the U.S. Government has employed physical anthropologists to develop identification processes (see McKern & Stewart, 1957; Trotter & Gleser 1952 for classic examples) and the U.S. Army Quartermaster Corps oversaw fundamental research that the forensic anthropological discipline continues to be used in bioanthropology and forensic anthropology university training. This core principle continues today with developments in chest radiograph matching, osteometric sorting of commingled remains, histological analysis, the uses of different forms of DNA, and, most recently, the development of isotopic signatures and maps to identify missing U.S. service personnel.

In 2009, the National Academy of Sciences published a report to the U.S. Congress entitled *Strengthening Forensic Science in the United States: A Path Forward*. One of the major efforts of this was the concept of validation of forensic studies, particularly related to understanding the known rate of error of specific methodologies. This is extremely important when dealing with various rules of evidence, including the Federal Rules of Evidence and the Daubart and Frye Standards. Thus, much of the research since 2010 at the SA has focused on validation of methodologies that are part of the Laboratory SOPs; any new research must go through a validation process to become part of the SOP, including publication as a peer-reviewed journal article.

Another division of the DPAA, Partnerships and Innovations, has an important role in the expansion of the mission to include a variety of partners, including universities to assist in field work and develop or validate analytical procedures and conduct historical research. Continued laboratory management procedures have developed sophisticated data bases and case management systems for maintaining the large amount of data produced by the DPAA forensic anthropologists (STAR—Laboratory Information Management System). Applications such as the Commingled Remains and Analytics (CoRA) allow the recording of specimen level information for reassociating remains back to individuals. CoRA is a collaborative ecosystem product of a partnership with the DPAA and the University of Nebraska Omaha, College of Information Sciences and Technology.

Some forensic anthropologists may think of the SA Laboratory Standard Operating Procedures (SOPs) or Laboratory Manual as formulaic and stagnant in terms of analysis and methodology, but it should be clear from the discussion above that this is far from the truth. Research and development of new methodologies is part of the dynamic aspect of the SOPs with constant revision, validation, and addition of new methodologies as research continues in the field of forensic anthropology as used for the identification of missing U.S. service members.

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AUTHOR CONTRIBUTIONS

William Belcher: Conceptualization (lead). **Calvin Shiroma:** Writing – original draft (supporting). **Lesley Chesson:** Writing – original draft (supporting). **Gregorgy Berg:** Writing – original draft (supporting). **Miranda Jans:** Writing – original draft (supporting).

DATA AVAILABILITY STATEMENT

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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