CASE REPORT

The Tarawa Project Part I A Multidisciplinary Approach to Resolve Commingled Human Remains from the Battle of Tarawa

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ABSTRACT: As part of the U.S. Navy's campaign against the Japanese during World War II, the Battle of Tarawa in November 1943 resulted in over 1,100 U.S. and over 6,000 Japanese and Korean conscript casualties on and around Betio Island, Tarawa Atoll, Republic of Kiribati. Casualties were buried in isolated and mass graves, which were negatively affected by the wartime renovations of the island, resulting in only ~47% of the remains being recovered in the late 1940s. The nature of the wartime burials, multiple postwar disinterments and reinterments, identification efforts in the late 1940s, and postwar anthropogenic changes to the island by the local population have contributed to the commingling of casualties. The Defense POW/MIA Accounting Agency established the Tarawa Project in 2016 to assist in the sorting, association, and identification of the often commingled remains recovered directly from the Republic of Kiribati and 94 caskets of Battle of Tarawa unknowns disinterred from the National Memorial Cemetery of the Pacific, Honolulu, Hawai'i. The project has been able to confirm a minimum number of individuals (MNI) of 243 based on unique mitochondrial DNA (mtDNA) sequences, while inventory of ~9,000 elements indicates a MNI of 131 from disinterments and 96 from recent field recoveries. The Tarawa Project has used a combination of historical research, DNA testing, chest radiograph and odontological comparisons, and forensic anthropological and material evidence analyses to help identify 41 of the 84 total service members accounted for from the Battle of Tarawa since the 1940s.

KEYWORDS: forensic anthropology, commingling, Tarawa

Introduction

The Defense POW/MIA Accounting Agency (DPAA) is responsible for the search for, recovery of, and identification of unaccounted-for U.S. service members from past conflicts; the DPAA Scientific Analysis Directorate (DPAA-Laboratory) established the Tarawa Project in the autumn of 2016 to assist with the identification of U.S. casualties from the Battle of Tarawa. This project is tasked with resolving the commingling of casualties (including the segregation of the U.S. from Japanese/Korean remains), associating remains recovered directly from the Tarawa Atoll and/or disinterred from the National Memorial Cemetery of the Pacific (NMCP), and proposing potential identifications of service members to the science director/medical examiner. The nature of the battle, the complex post-depositional history of

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Received 5 June 2018; Revised 18 August 2018; Accepted 21 August 2018 the casualties, and the ongoing recovery efforts highlight how remains can become commingled and the need to have a multidisciplinary approach to resolving this commingling.

Even though the list of unaccounted-for U.S. casualties from the Battle of Tarawa is generally known, the wartime burial procedures combined with the postwar recovery and identification efforts have resulted in a complex commingled assemblage. The post-depositional changes to the island, the extended interval since the incident, the poor preservation of the remains, and the large size of the assemblage all affect the ability to resolve the commingling and identify the unaccounted-for U.S. casualties (Adams & Byrd 2014). Further, the prevalence of an open context of commingling in this assemblage, where the primary burial location is unknown, limits the utility of the burial records associated with a particular casualty (Puerto et al. 2014). Similar to current-day disaster victim identification (DVI) scenarios, a multidisciplinary approach is necessary to systematically analyze the remains and resolve the commingling, especially given the likelihood of non-U.S. remains commingled with U.S. remains in the assemblage (Puerto et al. 2014).

Historical Background

In November 1943, U.S. forces began an initiative to invade the Japanese-occupied Gilbert Islands (now the Republic of Kiribati), termed Operation Galvanic. The largest assault took place on Betio Island in the Tarawa Atoll from 20 to 23 November 1943, resulting in a large number of casualties (Alexander 1993). The U.S. Second Marine Division reported 997 marines and 30 sailors (medical corpsmen) dead, 88 marines missing and presumed dead, and 2,292 injured (Alexander 1993). The reported numbers for the Japanese forces included 4,690 dead and 146 Japanese and Korean prisoners taken (Alexander 1995; Wukovits 2006). Due to post-battle accounting efforts, the initial reported numbers have been increased to ~1,150 U.S. and ~6,000 Japanese and Korean casualties.

The large number of fatalities, small size of Betio Island, and rapid rate of decomposition necessitated quick burial of the casualties (Alexander 1993). Often trench burials were established using bulldozers with casualties placed shoulder to shoulder wrapped in ponchos and a single marker erected to signify a whole unit (e.g., a single wooden cross inscribed simply with "D-2-18" to refer to D Company, Second Battalion, 18th Marine Regiment; Alexander 1993) or inscribed as "unknown" (Wukovits 2006). These burial practices resulted in incomplete documentation of grave locations and the individuals interred in them as well as the accidental commingling of remains (e.g., appendages overlapping with adjacent remains). At least five compulsory cemeteries were created on Betio Island by the assault troops prior to 24 November 1943, with a total of 43 isolated gravesites or cemeteries reported by 1946 (Steere & Boardman 1957; Wukovits 2006). Further, U.S. Navy Construction members ("Seabees") physically modified Betio Island (1943-1946) to meet wartime needs and "beautified" the cemeteries, resulting in burials becoming disassociated from their corresponding markers (Steere & Boardman 1957).

In March 1946 the American Graves Registration Service (AGRS) arrived at Betio Island to recover the war dead and began consolidating U.S. casualties from Betio and its neighboring islands into a single Lone Palm Cemetery, but they often found remains skewed from or no remains associated with burial markers (Steere & Boardman 1957). By May 1946 the AGRS had recovered and interred 532 remains (~50% of U.S. casualties) into the Lone Palm Cemetery. Just over half of these remains were interred with name associations through in-field analysis of grave markers, dental charts, and/or identification media. The rest were designated with an X-number (e.g., X-001) and buried as an unknown. Due to the lack of trained recovery personnel and the speed of the 1946 operations, many remains became commingled and/or were not completely recovered. In 1947 the AGRS disinterred, chemically treated with a paraformaldehyde powder, and relocated the remains from the Lone Palm Cemetery to the Schofield Barrack's Central Identification Laboratory (CIL), Oahu, Hawai'i, for processing and analysis.

Due to the return of the local residents and population growth, the landscape was further altered after the war (e.g.,

houses built on top of cemeteries), including evidence of the cemeteries (e.g., removal of grave markers). Remains have been intermittently recovered from Betio Island since the end of World War II due to accidental discovery through natural disasters and infrastructure developments, as well as formal excavation by the DPAA, its predecessor organizations, and nongovernmental organizations. The nature of these recoveries often resulted in groups of commingled remains being unilaterally turned over to the U.S. government or remains being reburied near their primary discovery location and later excavated from the secondary burial. Thus, remains accessioned into the DPAA-Laboratory have been primarily commingled assemblages of articulated portions of remains and miscellaneous unarticulated elements with minimal associated provenience information, which represent U.S., Japanese, and Koreans casualties from the Battle of Tarawa and Kiribati nationals. In rare instances, intact in situ burials from wartime trench features have been recovered.

Historical Analysis

The Schofield-CIL processed all casualties from the Lone Palm Cemetery between 1947 and 1949. Laboratory personnel confirmed AGRS field identifications and identified many of the unknowns through anthropological assessment and comparison to antemortem dental records. Historical records of the Schofield-CIL anthropological analyses indicate that 48 of the 92 unknowns processed through the CIL showed some level of commingling, usually duplicated or nonmatching elements. Further, multiple unknowns (X-numbers) believed to be commingled were often processed simultaneously, with elements being removed from or exchanged between unknowns in an effort to segregate the remains. For example, the CIL assumed that continuous X-numbers had a shared provenance when in most cases they came from different cemeteries. Further, a 1948 AGRS Review Board approved the burial of all of the unknowns as a group. Sets of remains were sorted by element in preparation for the group burial, but approval was later rescinded and the 92 unknowns were interred at the NMCP between 1949 and 1950. Subsequent to these "individual" interments, one casket containing the remains of at least 10 individuals recovered from Betio Island in the late 1960s after storms, and another casket containing at least two individuals recovered in the 1970s during infrastructure construction (analyzed by the CIL-HI without success), were interred at the NMCP.

Present-Day Analysis

The Tarawa Project team employs a systematic multidisciplinary commingled human remains (CHR) approach following standard DPAA protocols. The six MA- and PhD-level anthropologists making up the team process and analyze the project assemblage both to facilitate the association of skeletal and dental elements to a single individual and as a means to identify missing U.S. service members (Jin et al. 2014; Fig. 1).

Materials

The Tarawa Project assemblage is a compilation of the 94 caskets disinterred from the NMCP and 83 accessions (aka field accessions) of remains recently excavated from Betio Island, Tarawa Atoll, Republic of Kiribati by the DPAA, its predecessor organizations, and nongovernmental organizations (see Fig. 1, Ia and Ib). To date there are nearly 9,000 elements or groups of elements (i.e., complete articulated hands or feet or seriated sets of ribs and vertebrae) in the assemblage. Field accessions comprise ~6,000 of these ~9,000 elements, while remains from disinterred caskets comprise the remainder, with ~75% of the inventorying complete (see Fig. 1, IIa and IIb). The field accessions contain material evidence (ME) and osseous remains ranging from a single element to hundreds of elements. Further, evidentiary material continues to be recovered and accessioned into the Tarawa Project assemblage as a result of the DPAA partnering with nongovernmental organizations to maintain active recovery efforts on Betio Island.

Preservation of remains varies from excellent to poor across the assemblage. Even though most NMCP-derived remains were treated with a paraformaldehyde preservative prior to interment, many of the remains exhibit water erosion and are friable. The remains being recovered from Betio Island are generally in good to excellent condition, but some exhibit evidence of burning and postmortem fragmentation.

Analytical Procedures

All remains are systematically inventoried, each element or group of articulated elements is given a unique designator, and preliminary analyses are conducted and peer-reviewed (e.g., completeness, developmental status, measurements, and articulation or refit with other elements or fragments within the accession; see Fig. 1, IIa and IIb). After the initial inventory and assessment of morphological re-associations, project personnel select specific elements for mtDNA and/or isotope testing. Although destructive, these analyses are an important means to reassociate elements given the lack of context for many of the remains. Isotopes are being used in lieu of mtDNA testing in many instances to help segregate Japanese/Korean remains from U.S. casualties to ensure efficient use of limited resources. Both Sanger and nextgeneration sequencing (NGS) methods are being utilized by the Armed Forces DNA Identification Laboratory (AFDIL), who conducts DNA testing for the DPAA, for yielding mtDNA sequences. All preliminary data are entered into the Commingled Remains Analytics (CoRA) (Pawaskar 2016) web application and database for subsequent analyses.

The segregation portion of analysis is twofold: segregation of U.S. casualties from foreign nationals, and association of elements for individuation (see Fig. 1, IIIa and IIIb). This step utilizes data from DNA and isotope testing, CXR and dental comparisons (Shiroma 2017; Stephan et al. 2011), morphological assessments (e.g., refitting of fragments, joint articulation, visual and/or statistical pair matching [Adams & Byrd 2006; Lynch 2018a; Lynch et al. 2018], and osteometric sorting [Byrd 2008; Byrd & LeGarde 2014; Lynch 2018b]), historical record and archaeological provenience reviews, and/or biological profile criteria (e.g., age, sex, ancestry, stature, trauma, pathology, etc.) to determine if elements are consistent with a single individual. In addition to reassociating skeletal elements, these methods are used to differentiate Japanese and Korean from U.S. casualties during the segregation process (Christensen 2014; Marshall et al. 2018; Shiroma 2017). This series of analytical tests are summarized in a Report of Segregation, which is peerreviewed by another team member (Fig. 1, Va and Vc). Unlike other DPAA projects, segregation of remains is not attempted entirely in the blind since the association of the remains and identification can be intertwined.

Comparison between disinterred remains and field accessions is made when the data from preliminary assessments and the segregation process support a possible association between the two (see Fig. 1, IV). For example, when one or more elements return mtDNA sequence data, an effort to directly associate these elements with other elements from that accession or provenience is completed. Additional comparisons are made with relevant field accessions when the original cemetery information for a NMCP disinterment is known and can be linked to an area that has been excavated. Further, historical information from the unknown X-files can be used as a means to track prior efforts to solve commingling issues, as well as identify potential sources of historical laboratory error and commingling. When the X-file indicates commingling or attempted segregation with other cases, all disinterred cases that were simultaneously processed or had elements interchanged at the Schofield-CIL are evaluated for possible associations (see Fig. 1, IV). As this is part of the segregation process, it too is summarized in a Report of Segregation (see Fig. 1, Vb).

The project attempts to associate casualty names to remains once they are successfully segregated into discrete individuals (see Fig. 1, VI). The association of a set of remains to a specific U.S. casualty is accomplished through comparison of DNA sequence data to available family references samples (FRS) for U.S. Battle of Tarawa casualties, postmortem dental charts and radiographs to antemortem dental records and chest radiographs, biological profile criteria to

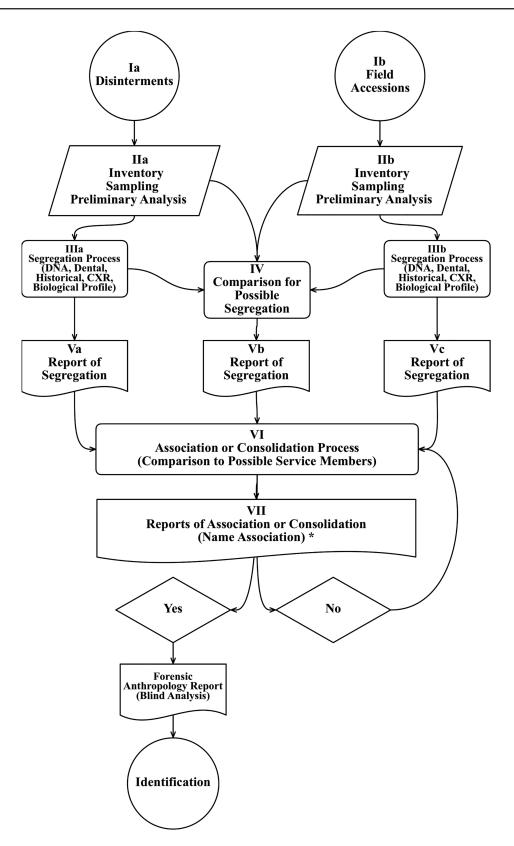


FIG. 1—The Tarawa Project workflow outlining the commingled human remains (CHR) processes. At the completion of the Reports of Association or Consolidation (*), the science director/medical examiner is briefed to determine if a case will move forward toward identification. A nonpartisan analyst will conduct a full forensic anthropological analysis in the blind subsequent to the CHR process.

official military records, and recorded recovery location to disposition records (Fig. 2). For example, when an mtDNA sequence is obtained, these data are used to generate shortlists of service members with matching FRS mitochondrial lines. Conversely, if no sequence data was obtained or there is no FRS match, the initial shortlist is created from a dental or biological profile comparison to all U.S. casualties and/or historical burial location records (e.g., chaplain's book, Marine Corps rosters, and individual casualty cards). Ideally, this initial shortlist is iteratively parsed until one name remains, who represents the most likely candidate (see Fig. 2). When a list cannot be parsed to one name, additional DNA testing may be requested. The additional FRS comparison depends on the type of sequencing method used to obtain the mtDNA sequence; NGS samples will undergo whole genome comparisons, and Sanger-derived samples will undergo nuclear testing (e.g., Y-STR or auSTR testing).

The summary of the potential name association processes are outlined in a Report of Association or Consolidation, dependent on whether the remains came from a single accession or multiple accessions, respectively, and like all other analytical reports is peer reviewed (see Fig. 1, VII). At this time the medical examiner/science director is briefed on the case and decides on whether the case moves forward toward identification. If they agree with the project's findings, a full forensic anthropology analysis is completed in the blind by an analyst not associated with the Tarawa Project (see Fig. 1). If they do not agree with the findings, additional testing is initiated and the case remains with the project until all concerns are addressed.

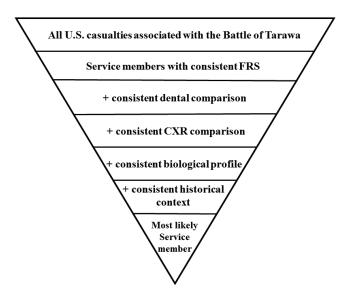


FIG. 2—The process by which a name association is generated during the consolidation/association step. The list of possible individuals is reduced as identification criteria are added until one name is associated with the remains.

Results and Discussion

Extent of Commingling

Thus far, the ~6,000 elements recently recovered from Betio Island represent a minimum number of 96 individuals (MNI). The MNI is based on duplication of hand elements and highlights the larger number of small elements recovered from Betio Island compared to the more diagnostic elements of the skeleton. The majority of the field accessions represent isolated body segments-such as hands, feet, patellae, and miscellaneous elements from throughout the body-and are often associated with material evidence of U.S. origin. This assemblage, when compared to the disinterred accessions, indicates that these elements are the remnants of the AGRS recovery efforts in the 1940s likely left in their original burial locations and that they likely reflect additional portions of some of the remains disinterred from the NMCP. Archaeological field documentation and preliminary mtDNA results indicate that large assemblages of highly commingled, fragmented, and/or burned remains recovered from secondary burial locations are more likely to be Japanese in origin (see Challenges section below for further details).

The ~3,000 elements from the 94 caskets disinterred from the NMCP represent a MNI of 131 individuals based on the duplication of elements and morphological and articulation discrepancies. Present-day analyses of the disinterred NMCP remains confirm the commingling noted in the historical records. Thirty-one of the 94 disinterred caskets exhibit duplication of elements or poor articulation among elements. Preliminary mtDNA results support these conclusions in six cases. Duplication of elements is most common in the hands, feet, and ribs, with 23 unknowns demonstrating this. Six cases exhibit poor articulation between elements, with five involving the major elements of the upper limb. The recent excavation of intact burial trenches from Betio Island revealed the overlap of upper and/or lower limbs between adjacent individuals during their initial interment in 1943. This revelation supports the need for a critical approach to the reanalysis of the disinterred accessions. In these cases, DNA analysis is required to resolve the commingling.

The AFDIL analyses have produced 243 unique mtDNA sequences with Sanger sequencing and 75 complete mitogenome sequences with NGS from 655 samples at the time of publication. These sequences include both U.S. service members and Japanese casualties. Based on FRS and population database searches, 124 of the 243 (~50%) unique Sangergenerated mtDNA sequences are consistent with Asian haplogroups, 92 (41%) of the sequences are European, and 9 (~4%) are undetermined haplogroups (Marshall et al. 2018). The Sanger-derived sequences provide a conservative MNI of 243 for the entire Tarawa Project given the possibility that mtDNA sequences represent multiple individuals and there is overlap between field and disinterred accessions. The chemical treatment of most NMCP remains prevents successful Sanger sequencing of these samples, and the Tarawa Project success rate for NGS is ~47.7% (75 sequences generated from 157). To date there are ~80 NGS samples, 420 Sanger sequence samples, and 40 samples for nuclear testing awaiting analyses, so a more definitive MNI is not possible at this time.

Association of Remains

The systematic inventorying of the field accessions and disinterred Battle of Tarawa unknowns in conjunction with analyses of the historical records and archaeological provenience have allowed the Tarawa Project to associate commingled elements within and between the NMCP and field accessions. Most cases successfully processed through the project since 2016 have been nearly complete skeletons that did not require DNA analysis for reassociation of elements to a single individual. However, 15 of the 41 identified cases have utilized DNA in combination with articulation, osteometric sorting, and provenience to confirm the association of the elements. Seven of these 15 cases required the association of elements from a disinterred accession to one or more field accessions and led to the refitting of additional fragments beyond the DNA-sampled elements in six cases. Two of the 15 cases involved positively associated remains that originated from four different NMCP caskets. To date, six cases have had non-articulating clavicles and/or cervical vertebrae included in an accession based on the positive association of the CXR to the same name as a dental and/or DNA FRS comparison.

Identifications

Prior to the inception of the project, identifications of U.S. Battle of Tarawa casualties were sporadic and primarily relied on biological profile and dental record congruency (Figs. 3 and 4). Identifications increased after 2013 as a result of the increase in Betio Island field excavations, FRS DNA availability, and the integration of CXR comparison as a routine laboratory procedure. At the time of publication, the

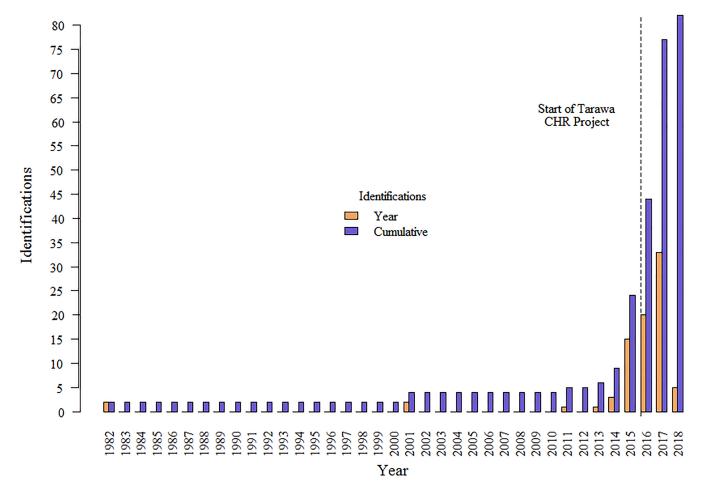


FIG. 3—The number of identified individuals from the Battle of Tarawa since the 1980s. The increase in identifications in recent years is associated with the inception of the Tarawa Project in 2016 and the recovery efforts on Betio Island, Tarawa Atoll, Republic of Kiribati.

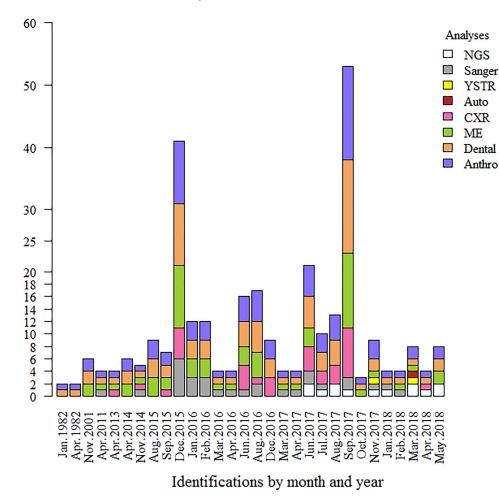


FIG. 4—The types of analyses contributing to individual identifications. The peaks in December 2015 and September 2017 are directly related to the recovery of undisturbed wartime graves. Anthropological and dental analysis contribute to nearly all identifications, but other analytical methods are becoming vital for the successful outcome of a case.

Tarawa Project has assisted with the identification of 41 (~50%) of the 82 unaccounted-for service members from Battle of Tarawa since the 1940s. These numbers are a direct result of the focused efforts of a dedicated commingled human remains team, the approval to disinter the unknowns buried at the NMCP, and the increase in FRS DNA availability. Since 2016, 26 cases utilized a combination of NGS (n=23) or Sanger (n=11) sequencing, as well as nuclear ySTR (n=2) and autosomal (n=1) testing (see Fig. 4).

Challenges

One of the biggest challenges for the Tarawa Project is segregating U.S., Japanese, and Korean casualties due to the extensive commingling of remains recovered from Betio Island. The project has taken a three-pronged approach to sorting U.S. from Japanese and Korean remains: analysis of skeletal and dental morphology, DNA haplogroup analysis, and isotope analysis. The overall morphology has been most useful with dental remains, especially those with non-Western dentistry (Shiroma 2017). Any elements that can be directly articulated with the dental remains are currently not submitted for DNA sampling. Other elements suspected of being non-U.S. in origin based on recovery context are submitted for haplogroup testing prior to producing a full sequence (Marshall et al. 2018). Elements for which a sequence has already been generated and are unique in the AFDIL population database also undergo haplogroup testing. Based on the mtDNA results and the ongoing recoveries on Betio Island, isotope analysis should allow for the relatively quick segregation of Japanese/Korean remains from U.S. remains and help prioritize mtDNA samples. Isotope analysis is the newest method added to the project's toolkit and, thus, results are not yet reportable.

Another challenge facing this project is the low number of appropriate FRS on file for the unaccounted-for U.S. casualties. AFDIL has an FRS on file for 86% of all of the unaccounted-for from the Battle of Tarawa, but only 69% of these are mitochondrial references. The Tarawa Project is limited in the fact that samples processed through NGS cannot undergo nuclear testing as of yet, so the individuals who only have a paternal FRS on file cannot be compared to these samples. This limits the ability to reassociate elements and/or eliminate individuals from a shortlist.

The Tarawa Project, like other commingled projects, also has had to contend with limited antemortem records and documentation. Chest radiographs are only available for ~25% of all U.S. casualties, and often the only dental records available are from induction records, so may not reflect subsequent dental work. Additionally, archaeological field notes (e.g., provenience information and maps) of recovery operations from the 1940s to present are difficult to use to support the association of remains due to lack of or inconsistencies in the field notes. Further, it is not uncommon to find identification media (e.g., ID tags) with remains that are not the individual; it was common for service members to collect identification tags to account for the fallen, so some individuals may have multiple ID media found with them. For these reasons, the project relies on many lines of evidence from multiple disciplines to support the segregation of remains and the identification of a service member.

Conclusions

The confounding disposition history of the U.S. and Japanese Battle of Tarawa casualties has led to many graves being lost over time. The renewed efforts to account for these service members, through the establishment of the Tarawa Project in 2016, highlight the successes and challenges of undertaking such a complex project. The Tarawa project has assisted in the identification of 41 previously unaccounted-for service members, including 26 unknowns disinterred from the NMCP. Several of these cases relied on the ability to associate elements recently recovered from Betio Island to disinterred remains through a multifaceted approach that included the articulation of elements, refitting of fragments, osteometric sorting, dental and CXR comparisons, and/or DNA testing (for examples of individual identifications, see Scott et al. 2019). Furthermore, the systematic inventory and preliminary analysis of remains have allowed us to determine possible relationships between disinterred remains and field recoveries, focus the DNA sampling strategy employed, establish an isotope initiative for segregating Japanese and U.S. remains, and target areas for excavation on Betio Island. Associations between remains and identifications of service members being made from the Tarawa Project rely on the

totality of evidence and interdisciplinary collaborations from geneticists, historians, archaeologists, odontologists, and anthropologists; no one scientist works in a vacuum.

The formation processes associated with commingled contexts vary significantly (e.g., mass disasters versus humanrights violations); however, there are several commonalties between these different commingled assemblages (Puerto et al. 2014). Forensic practitioners working on CHR projects must contend with the passage of time, preservation of remains and recovery context issues, poor record keeping, and the availability of antemortem data. As the literature on DVI and commingled remains proposes, and as this project highlights, multidisciplinary approaches are vital to successfully identifying individuals from commingled assemblages.

Acknowledgments

We would like to acknowledge all of the service members who have made the ultimate sacrifice; DPAA's strategic partner, History Flight, Inc., for their recovery efforts on Betio Island; and our project team and colleagues for feedback on this paper.

The opinions presented here are those of the authors and do not necessarily reflect those of the DPAA, the Department of Defense, or the United States government.

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95

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