Repatriation and Identification of Finnish World War II Soldiers

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Aim To present a summary of the organization, field search, repatriation, forensic anthropological examination, and DNA analysis for the purpose of identification of Finnish soldiers with unresolved fate in World War II.

Methods Field searches were organized, executed, and financed by the Ministry of Education and the Association for Cherishing the Memory of the Dead of the War. Anthropological examination conducted on human remains retrieved in the field searches was used to establish the minimum number of individuals and description of the skeletal diseases, treatment, anomalies, or injuries. DNA tests were performed by extracting DNA from powdered bones and blood samples from relatives. Mitochondrial DNA sequence comparisons, together with circumstantial evidence, were used to establish the connection of the remains to the putative family members.

Results At present, the skeletal remains of about a thousand soldiers have been found and repatriated. In forensic anthropological examination, several injuries related to death were documented. For the total of 181 bone samples, mtDNA HVR-1 and HVR-2 sequences were successfully obtained for 167 (92.3%) and 148 (81.8%) of the samples, respectively. Five samples yielded no reliable sequence data. Our data suggests that mtDNA preserves at least for 60 years in the boreal acidic soil. The quality of the obtained mtDNA sequence data varied depending on the sample bone type, with long compact bones (femur, tibia and humerus) having significantly better (90.0%) success rate than other bones (51.2%).

Conclusion Although more than 60 years have passed since the World War II, our experience is that resolving the fate of soldiers missing in action is still of uttermost importance for people having lost their relatives in the war. Although cultural and individual differences may exist, our experience presented here gives a good perspective on the importance of individual identification performed by forensic professionals.
In the course of the Second World War (WWII) Finland fought three separate wars (1): the Winter War (1939-1940) and Continuation War (1941-1944) against the Soviet Union, and the so-called Lapland war against Germany (1944-1945). Altogether 93,500 Finnish soldiers lost their lives or went missing in the hostilities, the absolute majority of these on the Russian front. In contrast to many other nations involved in the war, Finns did not primarily bury the deceased soldiers in military field cemeteries, but instead transported the bodies to their home towns for burial whenever possible. Despite these efforts, 13,000 soldiers went missing in action (MIA) or were left on the Soviet territory, most of these during the relatively short period of war in summer 1944.

There has been a growing public interest in finding and repatriation of the remains of Finnish soldiers after the previous battlegrounds became accessible after the collapse of Soviet Union in 1991. The repatriation was enabled by an agreement between Finland and Russian Federation signed in 1992. This marked the start of field searches, repatriation, identification, and reburial of Finnish soldiers, an effort that still continues. In Finland, these were initially coordinat-
tion and by the ACMDW from 1998 onwards, performed mostly with the aid of voluntary field workers, mainly from Finland but also from Russia. Based on written or oral information collected from archival documents or from war veterans, the searches were primarily targeted to areas where remains of Finnish soldiers were likely to be encountered. During this field collection phase, trained forensic professionals have rarely been present at the sites. Therefore, basic information from the site of the remains was carefully documented, but no special attention was paid to recording the injuries or number of individuals at each recovery site. Field workers ascertained the nationality of the bodies based on items around the remains (e.g., vestiges of military uniform or other equipment) or from other circumstantial evidence. The exhumed human remains recognized as Finnish soldiers were transported and temporarily stored in the Lutheran Church of Vyborg, Russia. After each summer field period, the remains were then repatriated and stored by ACMDW in Helsinki, Finland, for further examination in the Department of Forensic Medicine, University of Helsinki (DFM-UniHel) or, in cases of no lead for identity, for ceremonial military funeral. The organization and the process are schematically presented in Figure 2.

**Submission for forensic investigation**

The prerequisite for submitting the skeletal remains for further forensic anthropological and DNA analysis was that a metal identification tag (ID-tag) was found at the field site with the remains. Based on the personal ID number on these tags, information of the likely bearer of the found tag was requested from the Military Archives of Finland, where the data of their residency at the time of disappearance or death are also filed (online in Finnish: [http://tietokannat.mil/j/][1]). Occasionally, other associated oral or written information and/or personal belongings, such as wedding rings, were used as criteria for submission. ID-tags were not considered as sufficient proof of identity, as they could rarely be reliably associated with the remains of a single person.

**Family association**

After obtaining initial information on the possible identity, the identification was further attempted using mtDNA data. In order to obtain reference DNA profiles for the comparisons, the ACMDW sought for living maternal relatives from the local church and national parish registers. Blood samples from the voluntary relatives were taken in the nearest health care centers or at the DFM-UniHel. Blood samples were sent to the Laboratory of Forensic Biology in the DFM-UniHel via mail, with no costs to the donor. Most reference samples were from first-degree relatives (sisters or brothers) of the missing soldiers, but more distant relatives were also willing to participate by request. In total, 111 reference samples have been analyzed to date.

**Investigation of the skeletal remains**

The recovered remains for which evidence for the identity existed, were transported to the DFM-UniHel, where they were investigated in an autopsy room. After manual cleaning of the

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[1]: http://tietokannat.mil/j/
remains, forensic anthropologist and pathologist attempted to reconstruct the skeletons and to define the minimal number of individuals (MNI) per coffin. In addition, the remains were examined to determine the sex and to document any signs of disease, surgical treatment, skeletal anomalies, or injuries. The age and stature estimation was performed, but they were considered of secondary value for the identification. After anthropological investigation, bones suitable for DNA analysis were sampled. In many instances, a forensic geneticist participated in this stage of investigation both for mutual educational and immediate discussion purposes.

The preferred samples for DNA analyses were 10-20 cm sections of long, intact bones such as femur, tibia, or humerus. At times, DNA-profiles of several bones from one coffin were sampled in order to correctly determine the minimal number of individuals in each coffin. For this purpose, smaller bones were sampled in addition to the preferred compact long bones. Occasionally small bones or bone fragments were the only available samples, from which the macroscopically best preserved ones were chosen for DNA analysis. To date, altogether 181 bone samples from 84 sites were collected for DNA analyses (Table 1).

### DNA analysis

The identification of the remains was based on mtDNA sequence comparisons combined with other available information. Prior to DNA extraction, bone sample surfaces were first cleansed mechanically using sterile toothbrushes. Possible contaminants were removed by rinsing the bones once with sterile water, once with 1/10 dilution of hypochlorite solution containing 8% of active chlorine and finally with 70% ethanol. Bone samples were then desiccated in sterile chamber at room temperature for >72 hours.

After this initial treatment, DNA extraction proceeded with grinding the compact bone tissue into powder using a dental hand drill (Faro, Milan, Italy) or a microdismembrator (B. Braun Biotech, Melsungen, Germany). DNA was extracted from 0.2 to 1.0 g of the powder, using 6 mL of extraction buffer (10 mM TRIS pH 8, 100 mM NaCl, 50 mM EDTA pH 8, 0.5% SDS) and 150 µL Proteinase K (0.5 mg/mL). The solution was incubated at +56°C overnight, after which the extraction proceeded with standard phenol-chloroform method (2). DNA was concentrated with Centrex UF-2 (Schleicher & Schuell, Dassel, Germany) columns and further purified using QIAquick-purification kit (QIagen, Hilden, Germany). DNA from reference blood samples was extracted with the Chelex extraction method.

To minimize the risk of contamination, extraction of the blood samples and bone samples were carried out in separate rooms. In the bone extraction room, all equipment was irradiated with UV-light and non-disposable equipments and surfaces were treated with ethanol and hypochlorite. Only a small number (<6) of bone samples were extracted at one time and reagent blanks were used to detect possible contaminations.

HVR-1 (16024-16385; 326 bp) and HVR-2 (72-340; 269 bp) segments of the mtDNA control region were amplified separately and sequenced using primers H16391-L15997 and
H408-L048. In cases where the protocol failed to give reliable results, an alternative protocol with smaller amplicons was used. In these cases, both segments were amplified in two parts using primer pairs L15997-H16236, L16159-H16391, L048-H285, and L172-H408.

The obtained mtDNA sequences were checked and manually edited using software SeqEd v.1.0.3 and SeqScape v.2.0 (both Applied Biosystems, Foster City, CA). The SeqScape software was also used for sequence comparisons of concatenated HVR-1 and HVR-2 sequences from the victim and the putative relatives. For these positive matches, estimation of the power of evidence was based on Finnish mtDNA sequence database of currently approximately 800 unrelated Finns (3 and Palo et al, unpublished results).

Results
Field searches

During field searches in 1993 to 2007 about a thousand Finnish soldiers, who went missing in action or were left on the present-day Soviet territory, have been found and repatriated. All of the identified soldiers have been buried to their home residency graveyards according to the wish of the relatives. Repatriated soldiers whose identities could not have been established have received a ceremonial military funeral in dedicated graves.

Family association

For the DNA comparison, 111 family members were contacted. None of the relatives refused the offer to donate their blood sample for the identification purpose. The majority (57%) of the reference samples was from siblings of the victims, and more distant relatives such as cousins or cousin’s children were also successfully used as reference sample donors. The most distant reference sample was obtained from the soldier’s great-grandmother’s sister’s grandson.

Investigation of skeletal remains

The systematic data of anthropological examination was performed from 2002 to 2007. In this period, the remains from 42 sites were anthropologically investigated. The main aim of the forensic anthropological investigations was to establish the minimal number of individuals (MNI). The MNI varied from 1 to 8 per site and was altogether 101.

Forensic anthropology examination revealed several exit and entrance wounds and other injuries from projectiles were found (see example in Figure 3). In 5 cases, the injury was established to result from a gunshot wound, and in 7 cases caused either by a gunshot wound or shrapnel. In 12 cases, the signs of at least one clear injury due to ammunition were documented, and in some of those pieces of shrapnel were still imbedded in the bone.

Sex determination was possible for 62 individuals, all identified as males.

Signs of diseases included tibial periostitis, and 5 individuals showed signs of a healed long bone fracture. Other skeletal anomalies observed

Figure 3. Reconstructed skeleton of a repatriated Finnish soldier. Fully reconstructed skeletal remains (a) and details of projectile wounds in the right pelvic region (b) and tibia (c) are shown.
were spina bifida, fused vertebrae, and osteoarthritis.

**DNA analysis**

For the total of 181 bone samples analyzed, HVR-1 and HVR-2 sequences were successfully obtained from 167 (92.3%) and 148 (81.8%) samples, respectively. Five samples yielded no reliable sequence data. The quality of the mtDNA and the obtained sequence data varied depending on the sample bone type, with long bones (femur, tibia and humerus) having significantly better (90.7%) success rate than the small bones (51.2%, Table 1).

Among the 167 bone samples possibly or fully sequenced for HVR-1 and/or HVR-2, there were 104 individuals. Of these, concordance with mtDNA lineage to the family reference sample was confirmed for 79 individuals. The most distant reference sample, presenting maternal lineage over nine meioses, yielded a positive mtDNA match.

For 25 individuals mtDNA profiles that did not match the reference profiles were obtained, resulting in exclusion. The identity of these remains remained unresolved.

**Discussion**

The identification of the Finnish WWII soldiers is an ongoing project; 181 samples from approximately a thousand repatriated skeletal remains have been analyzed with DNA methods during 1993-2007. The limiting factor for the number of DNA identifications is the lack of the background information, due to the scarcity of ID-tags or other personal effect associated with the found remains. Due to this, identification of only 20% of the repatriated individuals could have been attempted with molecular methods.

For the total of 181 bone samples analyzed, HVR-1 and HVR-2 sequences were successfully obtained for 167 (92.3%) and 148 (81.8%) samples, respectively. Although five samples yielded no reliable mtDNA sequences, our data suggests that mtDNA preserved rather well in the boreal acidic soil, for 60 years at minimum. As expected, the quality of the mtDNA and the obtained sequence data varied depending on the sample bone type, the success rate being significantly better with long bones (femur, tibia and humerus; 90.0%) than the small bones (51.2%). Naturally, PCR inhibitors can also hinder the analyses of soil-contaminated bones, but these effects can be minimized with careful pre-treatment of the samples as well as with additional purification steps on the extracted DNA. Although the sequencing success rate in the small bones was substantially lower, occasionally these small bones were the only available samples, and at several cases yielded information otherwise unobtainable.

Among 167 bone samples successfully sequenced, there were at minimum 104 individuals. Of these, concordance with mtDNA lineage to the family reference sample was confirmed for 79 individuals (76%). Although the majority of the reference samples were from siblings, more distant relatives such as cousins or cousin’s children were also matching the mtDNA sequence from the bones. As an extreme case in our experience was a positive mtDNA match obtained from the soldier’s grandmother’s mother’s sister’s grandson, i.e., over nine meioses. Since there has been great public interest toward the identification of World War II soldiers in Finland, obtaining the reference blood samples has been a relatively easy task after the right persons have been located.

As mtDNA is only a single locus, the power of resolution was limited in certain cases. This indicates the need for additional and more advanced laboratory methods for autosomal STRs or Y-chromosomal markers with greater exclusion power (4,5). However, these markers, residing at the low-copy portions of the genome, are more difficult to score from old material, al-
though promising reports are now available (6,7). Yet, regional differences in the preservation of DNA are likely to exist due to climate and, eg, chemical properties of the soil.

Obviously, the power of exclusion of mitochondrial HVR-sequences are several order of magnitudes lower than values commonly reached with 10-15 biparental STR markers. The routine use of these more informative markers is, however, often precluded by the quality of the bone tissue (template DNA). The distribution of the Finnish mtDNA haplotype frequencies as well as their geographical distribution within Finland is even (4 and Palo et al, unpublished results). The mtDNA data together with associated information have been considered to fulfill the criteria for positive identification by Finnish officials, although orthodox forensic and population genetic interpretation may challenge this operational protocol.

Statistical interpretation of the use of mtDNA is challenging in general. In our case, this is especially demonstrated in the cases from the 1939-1940 Winter War as the infantry companies were usually formed by men of the same village – often resulting in the existence of maternal relatives among the perished. For example, in one case, reference samples for remains of 2 soldiers with putative identity from the same site yielded reference mtDNA profiles differing by one base pair. In this particular case, complete mtDNA control region sequence did not yield any additional data and the identities of these two soldiers remained unconfirmed. Although low in discrimination power, the use of mtDNA allows sampling of rather distant relatives, an advantage of growing importance as the availability of reference samples from closely related family members is gradually diminishing over time.

For 25 samples the identity could not be established. This can be considered rather high proportion, as only the remains with putative identity based on circumstantial evidence were chosen for the mtDNA analysis. This strongly suggests that circumstantial evidence, even identification tags or personal belongings, is not a reliable means of identification.

After Finland became independent in 1917 it has had a national service-based army. Consequently, all healthy men born between 1894 and 1926 were drafted at least in one stage of the World War II. As a consequence, the majority of Finns living today have had their relatives serving on the front. Together with the fact that most of the fighting occurred on areas that had previously been a part of Finland, the events of World War II have had a great effect also on the present day Finns. Both of these factors explain the great public interest in clarifying the fate of the soldiers whose bodies were not recovered. Therefore, collection of the reference blood samples has been fairly straightforward and the relatives have shown great interest toward our attempts in identifying the Finnish soldiers.

In general, the aim of repatriation and reliable identification of the Finnish soldiers with previously unknown fates, and subsequent burials by existing relatives underlines the societal impact of forensic know-how. Although more than 60 years have passed since World War II ended, our experience is also that this is of uttermost importance for people who had lost their relatives in the war. This is shown by the fact that none of the relatives – distant or close – has declined the possibility to aid in the identification process by donating his/her blood sample. Although cultural and individual differences may exist, our data gives a good perspective on the importance of individual identification performed by forensic professionals (8,9).

Acknowledgment
The authors thank Mr. Antti Vuorinen from the Ministry of Education, Mr. Markku Kiikka and Mr. Pekka Pitkänen from ACMDW, and several forensic experts in Finland for their invaluable work in this project. We thank Nils-Erik Lindeman and Osmo Eskola for their help in various phases of the project. We also thank many
of Finnish and Russian volunteers for their continuous activities in the field. Financial support for collecting the population data was provided by Academy of Finland (grants 109265 and 11713 to JUP and 80578/54938: OMLL to AS).

References