



A7 Mystery Solved: The Identification of the Two Missing Romanov Children by Forensic DNA Testing

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After attending this presentation, attendees will understand forensic DNA testing that was conducted on a set of skeletal remains recovered in the summer of 2007 near the former mass grave of members of the Romanov family officially excavated in 1991.

This presentation will impact the forensic community by concluding one of the greatest mysteries of the 20th century.

For over 300 years, the Romanov Dynasty ruled the country of Russia. In 1917 following the Bolshevik revolution, the last Russian Tsar, Nicholas II, abdicated his throne and was eventually exiled to Yekaterinburg with his wife, Tsarina Alexandra, and their five children: Olga, Tatiana, Maria, Anastasia, and the Tsarevich Alexei. Also present with the royal family were four loyal servants: Dr. Botkin, the family physician; Mr. Trupp, valet to the Tsar; Ms. Demidova, maid to the Tsarina; and Mr. Kharitonov, the family cook.

In July of 1918, the Bolsheviks feared an attempt to rescue the Tsar and his family by the White Russian Army. A decision was made by the Bolsheviks to execute the entire family, with the hope that upon hearing of the Tsar's death the will of the people loyal to the Tsar would be broken. In the early morning hours of July 17, 1918 the royal family and their servants were led to the basement of the Ipatiev house where they were being held and were executed.

According to the account written by the lead executioner, Yakov Yurovsky, the Bolsheviks first sought to dispose of the bodies by throwing the remains down an abandoned mine about 20 km outside of

Yekaterinburg, and then attempting to collapse of the mine by exploding grenades down the shaft. This strategy did not work as planned, and the next night the Bolsheviks endeavored to move the remains to another mine shaft about 30 km away. Approximately 2 km from the original mine shaft, their truck broke down in an area known as "pig's meadow." According to reports, the Bolsheviks removed two of the children's bodies from the truck at this location. In an attempt to completely destroy the remains, they dug a shallow grave, doused the bodies in sulfuric acid, and burned them as much as possible. This effort took more time than anticipated, however, and the remaining bodies were doused with sulfuric acid and hastily buried together, "some distance away", in a mass grave.

Approximately five days later, Yekaterinburg was liberated by the White Russian Army, and an attempt by investigators to discover the remains of the Romanov family came to a dead end. In the late 1970s, two Russian citizens obtained a copy of the Yurovsky report and were able to locate the mass grave containing the remains of five members of the royal family and their four servants. Following the fall of the Soviet Union in 1991, the men came forward with their discovery and an official recovery was conducted.

Forensic DNA testing of the remains recovered in 1991 was conducted by Dr. Peter Gill, formerly of the Forensic Science Service and Dr. Pavel Ivanov, a Russian geneticist.^[1] Nuclear STR testing of five loci confirmed the sex of the skeletons and established a familial relationship among the remains of the Tsar, the Tsarina and three of their daughters recovered from the same grave. Mitochondrial DNA testing confirmed a maternal relationship between HRH Prince Philip, the Duke of Edinburgh, and the Tsarina (and her daughters). The Duke of Fife and Princess Xenia were used to match the putative remains of the Tsar. A single point heteroplasmy at position 16169 (C/T) was observed in the mtDNA sequence of the Tsar, whereas his maternal relatives were fixed for 16169 T. In testing conducted at the Armed Forces DNA Identification Laboratory (AFDIL), the identity of the Tsar was further confirmed by the mtDNA heteroplasmy shared with Grand Duke Georgij Romanov, brother of Tsar Nicholas II.^[2]

Despite the overwhelming forensic evidence, doubts about the authenticity of the remains persisted. Skeptics often referenced the two children missing from the mass grave - Alexei and one of his sisters, either Anastasia or Maria. The missing remains also encouraged the beliefs held by some that these two children had somehow miraculously escaped the bullets of the executioners and made their way out of Russia. The most famous case was Anna Anderson, who claimed to be the Grand Duchess Anastasia. Forensic DNA testing eventually disproved her claims.^[3]

After the discovery of the "first" mass grave, several attempts were made in the following years to find the "second" grave, which was believed to be relatively nearby. In the summer of 2007, a group of



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amateur archeologists discovered a set of 44 bone fragments and teeth approximately 70 meters from the “first” grave. In late 2007, the Russian government invited a team of scientists to conduct an independent forensic DNA testing of the remains from the “second” grave.

We will present results from mtDNA, nuclear STR, and Y-STR testing of these remains. DNA testing on the skeletal remains from the “first” grave that conclusively link the two graves will also be presented. The results from the DNA analysis of all three genetic systems confirms that the samples tested from the second grave are one female and one male child of Tsar Nicholas II and Tsarina Alexandra, and resolves the mystery of the missing Romanov children.

The views expressed herein are those of the authors and not necessarily those of the Armed Forces Institute of Pathology, the U.S. Army Surgeon General, or the U.S. Department of Defense. **References:**

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- ² Ivanov PL, Wadhams MJ, Roby RK, Holland MM, Weedn VW, Parsons TJ. (1996) Mitochondrial DNA sequence heteroplasmy in the Grand Duke of Russia Georgij Romanov establishes the authenticity of the remains of Tsar Nicholas II. *Nat Genet.* 12(4): 417-420.
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