



# The Search for Egypt's Lost Queen Hatshepsut

Hatshepsut, Egypt's most powerful female ruler, reigned more than 3,000 years ago. After she died mysteriously, her name and monuments were systematically erased. Earlier this year, Dr. Zahi Hawass, Egypt's Secretary General of the Supreme Council of Antiquities, and his team of archaeologists identified the mummy of Hatshepsut from a single tooth. To confirm this remarkable discovery, researchers are performing nuclear and mitochondrial DNA analyses. This project led to a collaboration between the Discovery Channel, the Supreme Council of Antiquities, and Applied Biosystems that established the first laboratory in Egypt dedicated to testing ancient DNA samples.

Three thousand years before Europeans set foot in the Western Hemisphere, the pharaoh Hatshepsut ruled the Eighteenth Dynasty of Egypt. As one of the few female pharaohs in one of the most powerful and highly advanced civilizations of the ancient world, Hatshepsut ruled for more than twenty years, longer than any other female ruler of an indigenous dynasty. A visionary and highly successful leader, Hatshepsut commissioned hundreds of construction projects, rebuilt the country's trade networks, and presided over a flourishing economy.

When Dr. Zahi Hawass announced that he had positively identified a mummy as that of Queen Hatshepsut, he described the discovery as "the most important find in Egypt's Valley of the Kings since the discovery of Tutankhamun in 1922."

### The Challenges of Deciphering Ancient DNA

Ancient DNA research, which retrieves DNA sequences from museum specimens, archaeological finds, fossil remains, and other unusual sources, became feasible only with the advent of enzymatic amplification of specific DNA sequences [1]. For archaeological specimens, the retrieval of ancient DNA sequences is far from routine, as the researcher has to contend with the fact that very little and often damaged DNA survives in ancient tissues [2].

Because Egypt does not permit antiquity samples to leave the country, DNA analysis was impossible, as no genetics facility existed. Now, with the establishment of an Applied Biosystems

equipped laboratory, this has changed. According to Dr. Hawass, "Before, I completely refused to take DNA samples and send them out of the museum to be sequenced. But now, we have, for the first time, a DNA laboratory here in the museum." Dr. Hawass adds, "I really believe that all of the science and archeology should happen at the actual site or in the museum."

Dr. Hawass and his team first tested two unidentified female mummies who were thought to be Hatshepsut, and then compared their DNA with that from the mummies known to be Hatshepsut's father and maternal grandmother. The Egyptian team is conducting a combination of nuclear and mitochondrial DNA testing on the mummies. Nuclear DNA provides the most informative DNA profile because it contains information from both parents; however, it is more difficult to obtain a sufficient sample of nuclear DNA from degraded tissues, as cells contain only a single nucleus. Mitochondrial DNA is passed down only through the maternal side of a family, but because there can be thousands of mitochondria in each cell, mitochondrial DNA tests can supplement the analysis in cases where nuclear DNA testing fails to yield results.

### Applied Biosystems Joins the Effort

Applied Biosystems provided the project with DNA analysis instruments, reagents, software, and training. This included a GeneAmp® PCR System 9700 for DNA amplification, a 3130 Genetic Analyzer for DNA analysis, Analysis Software, and an

AmpF/STR® MiniFiler™ PCR Amplification Kit, the world's first commercially available reagent kit for generating genetic profiles from aged, compromised, or damaged DNA.

### Validation of Hatshepsut Mummy Using DNA Analysis

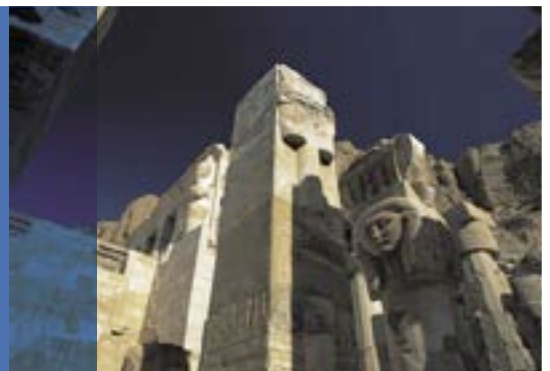
To validate their findings, the researchers extract mitochondrial and nuclear DNA from the unidentified mummies and are comparing it to samples from Hatshepsut's known relatives. A bone marrow biopsy needle was used to retrieve multiple samples from various angles at a single puncture site to minimize damage to the remains. Applied Biosystems reagents and instruments were used to amplify partial sequences of both mitochondrial DNA, which is passed from mother to daughter, and more fragile nuclear DNA, which may reveal Hatshepsut's relationship to the pharaoh Thutmose I and Queen Ahmose Nefertari. The challenges inherent in working with ancient DNA make this type of characterization difficult. Regardless of the result, however, all parties believe that this attempt to analyze ancient DNA is a worthy endeavor. In addition to the Hatshepsut project, scientists will use the new DNA testing facility to examine other Egyptian archeological finds.

#### References:

1. Pääbo S, Poinar H, Serre D, Jaenicke-Despres V, Hebler J, Rohland N, Kuch M, Krause J, Vigilant L, Hofreiter M (2004) Genetic analyses from ancient DNA. *Annu Rev Genet.* **38**:645–679.
2. Ekins JE, Ekins JB, Layton L, Hutchison LA, Myres NM, Woodward SR (2006) Inference of ancestry: constructing hierarchical reference populations and assigning unknown individuals. *Human Genomics*, **2(4)**:212–235.

"It is important in archaeology to use the most up-to-date scientific methods. Now, with the new DNA laboratory, we can go inside the mummies and find out about their family relationships, and possibly identify them with certainty. "

—Zahi Hawass, Ph.D.



**Dr. Zahi Hawass**

World-renowned archaeologist Dr. Zahi Hawass received his Ph.D. in Egyptology from the University of Pennsylvania in 1987. He currently serves as Secretary General of the Supreme Council of Antiquities and Director of Excavations at Giza, Saqqara, and the Bahariya Oasis. Dr. Hawass's dynamic personality and extensive knowledge have sparked global interest in ancient Egypt. He has brought the world of the pharaohs into the homes and hearts of people all over the world through his many television appearances and books for general audiences. Over the course of his long career, Dr. Hawass has received numerous awards and honors. Time Magazine named him as one of the Top 100 Most Influential People of 2005.



“By providing DNA testing technology to Egypt, **Applied Biosystems is helping to advance science and bring our dead pharaohs back to life.** A basement that was once a maze of artifacts is now a cutting-edge scientific lab, the first of its kind dedicated to revealing the mysteries of our mummies.” –Zahi Hawass, Ph.D.



Panel A. Laboratory for Studying Ancient DNA.



Panel B. Collecting Samples.



Panel C. Preparing Samples for DNA Testing.

Figure 1. State-of-the-Art Ancient DNA. (A) New ancient DNA laboratory in the basement of the Egyptian Museum, Cairo. Nicola Oldroyd, an Applied Biosystems senior forensic specialist, training members of the Egyptian scientific team on Applied Biosystems' sequence analysis software. (B) Samples are taken from one of the mummies for DNA testing in hopes of matching the KV 60A mummy (believed to be Hatshepsut) to the queen's known ancestors. (C) Samples from the KV 60A mummy, along with samples from Hatshepsut's relatives, Thutmose I and Ahmose Nefertari, are prepared for DNA testing.



Panel A. KV60 Mummy Determined to be Queen Hatshepsut.



Panel B. Queen Hatshepsut Mummy in the Queen's Pose.

Figure 2. Mummy of Queen Hatshepsut. (A) Dr. Zahi Hawass with the KV60 mummy, which he later identified as Queen Hatshepsut. (B) Mummy of Queen Hatshepsut in the “queen's pose,” with one arm placed diagonally across her chest.