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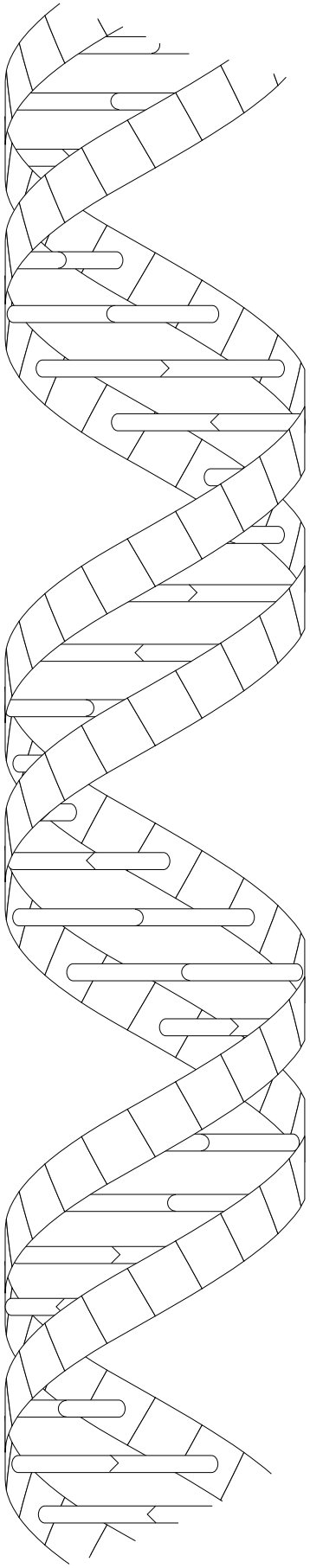


Secrets of the Ancient World Revealed Through DNA

A lecture presented to the ESS by Dr. Scott Woodward, Professor of Microbiology, Brigham Young University, 20 April 2001

Summarized by Judy Greenfield

DNA contains the history of who we are and where we've been. Each human genome contains 3,000,000,000 bits of information; if the information contained in a single cell's DNA was typed out in 12-point type, it would stretch from Denver to Tierra del Fuego! It is only in the last dozen years at most that scientists have been able to decode DNA, thanks to a process invented in 1985 called polymerase chain reaction, which Dr. Woodward said works like a "molecular Xerox machine," enabling sections of DNA to be split into its base pairs, amplified and replicated in a test tube.

One of the goals of sequencing human DNA is to reconstruct genealogies and relationships between individuals. The more closely individuals are related, the more genes they share. When individuals share a genetic sequence, they probably have a common ancestor, and family trees can be built based on this genetic evidence.

An individual's mother and father each contribute half of that person's genes. Two types of DNA have been investigated in cells from human bodies; mitochondrial (mtDNA) and nuclear DNA. Mitochondrial DNA resides outside the nucleus of the cell and is inherited solely from the mother; nuclear DNA derives from both the mother and father but is many orders of magnitude less abundant than mtDNA and is much more difficult to obtain for study.

While DNA is easily obtained from the tissues (e.g. blood, hair, skin) of living people, it is much more difficult to recover from ancient remains because DNA is one of the first things to undergo decomposition by enzymes within the cells. Since arid conditions arrest decomposition, it has been possible to obtain DNA samples from human remains preserved by desiccation, including mummification (both natural and artificial). Bodies have been found naturally preserved *via* desiccation in the deserts of China, South America, and, of course, Egypt. Human tissue also has been preserved in acidic bogs and by freezing. DNA is composed of very long sequences of four organic molecules: Guanine, Cytoscene, Adenoscene, and Thynene. The sequences determine all the characteristics of each individual's DNA, and thus his or her genetic makeup. Decay breaks the strands of DNA into fragments, so extracting and sequencing DNA from ancient individuals remains difficult. A further complication arises when comparing the DNA from two long-dead individuals, since sometimes different regions of each individual's genomes are sequenced and therefore do not correlate.

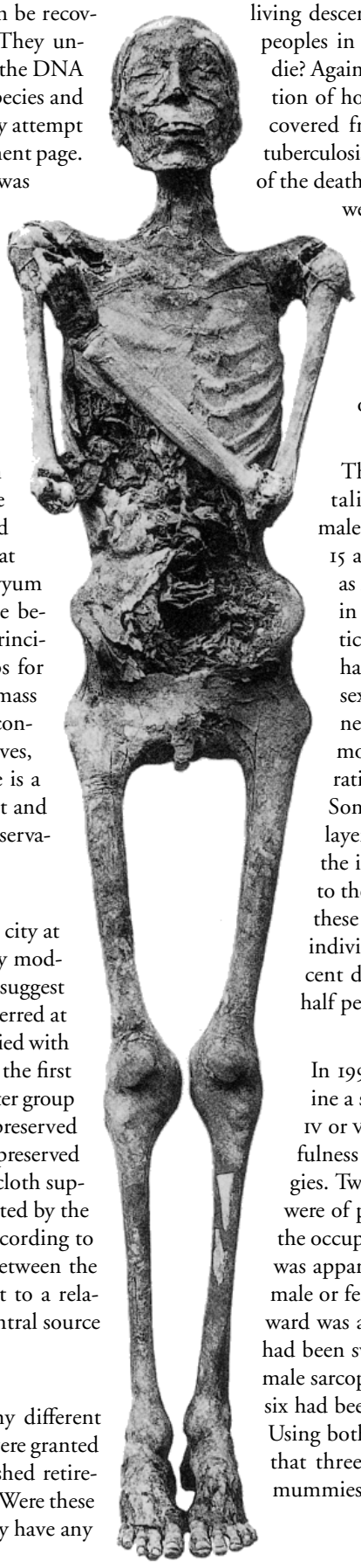
Which tissues of ancient human remains provide the best source of DNA? After experimenting with lung, bone, brain and various tissues, Dr. Woodward was surprised to find that teeth are the best source for DNA. Because tissue samples can be readily contaminated by ubiquitous modern DNA (in the form of shed cells), the outer layer of the tooth first is vigorously cleaned. The interior of the tooth then is drilled to obtain a "clean" sample and the pulverized tooth used for DNA testing. DNA derived from teeth is qualitatively in better condition and more robust than DNA obtained from other tissues.

Woodward and his team found that DNA can even be recovered from products manufactured from animals. They undertook a project to extract, magnify, and sequence the DNA from the Dead Sea scrolls in order to identify the species and individual animals from which it was made. An early attempt revealed that ibex skin was used for one such parchment page. The team later discovered that a 27-page scroll was manufactured from the skins of 23 individual goats. A student at Hebrew University is creating a reference collection of DNA from ibex, goat, sheep, and gazelle bones recovered from archeological sites in Israel. When complete, it should be possible to match the individual goats used to make the parchment with the sites from which they came, thereby identifying their places of origin.

Woodward provided another fascinating way in which DNA had been used to further knowledge about an ancient people. Beginning in 1991, he and his team researched the DNA of individuals buried at the 300-acre cemetery, Fag el Gamous, in the Fayyum (west of Saleh, near the Meidum pyramid). In use between 200 BCE and 400 CE, the cemetery consists principally of two types of graves: limestone shaft tombs for the wealthy and powerful, and the cluster burials or mass graves for commoners. Eighty percent of the graves contained multiple burials. In the limestone shaft graves, corpses were stacked one on top of another. There is a 400 to 500 year difference between the bottommost and uppermost burials, with the number of graves conservatively estimated to be 135,000.

Who were these people? There is no known ancient city at the site—unless it has been completely obscured by modern buildings and structures—but burial practices suggest that two major populations appear to have been interred at Fag el Gamous cemetery. The oldest bodies were buried with their heads facing west but later ones, from around the first century CE, face east. Woodward suggests that the latter group were early Christians. The older burials are poorly preserved and consist merely of skeletons; the latter are better preserved and include the linen burial cloths. Was the burial cloth supplied by a "manufacturer" or was it most likely created by the individual families as a sort of cottage industry? According to Wordward, comparisons of genetic relationships between the deceased and particular types of burial cloth point to a relatively widespread cottage industry, rather than a central source as the origin of the cloths.

Egypt was a cosmopolitan country, home to many different groups of people. It is known that Roman soldiers were granted land in the Fayyum after their service and established retirement villas in the Fayyum. Many questions remain. Were these Greeks and Romans? Were they Christians? Do they have any living descendants? Can they be genetically tied with living peoples in Egypt or elsewhere? And finally, how did they die? Again, DNA testing has helped shed light on the question of how the people in the mass graves died. DNA recovered from disease organisms, including cholera and tuberculosis, indicate that epidemics may be involved in some of the deaths. Indeed, two adults and a child buried together were determined through DNA testing to be "consistent with a two-generation family" of mother, father and child. They evidently died suddenly, probably from plague or an epidemic. Osteoporosis and abnormal growths also show up in the archeological record; these and other genetic diseases possibly played a role in the deaths of some of these people, too.



The older burials show a high degree of infant mortality, both from disease and infanticide. The male:female ratio of both infants and those older than 15 are skewed for reasons not yet determined. Twice as many males as females over age 15 are represented in the cemetery, perhaps as a result of female infanticide, which was known from written records to have been practiced. Woodward's team is currently sexing all the infant burials using DNA because it is nearly impossible tell males from females based on morphology of the infant skeletons. Thus far, the ratio seems to be 1.5 times as many males as females. Some of the deceased died a violent death. In one layer, representing a 30 to 50-year time period, half of the individuals died violently (e.g., by an axe wound to the head or a broken neck). The average lifespan of these unknown people was 35 years, with only a few individuals in their 50s and 60s represented. Forty percent died before they reached their fifth birthday and half perished before they were fifteen years old.

In 1993–94, Professor Woodward was asked to examine a set of six Egyptian mummies from the Dynasties IV or V. The exercise was to demonstrate again the usefulness of DNA for sexing and reconstructing genealogies. Two of the mummies were of grandparent age, two were of parent age and two were children. The names of the occupants were written on five of the sarcophagi and it was apparent from the face masks and names which were male or female. Or was it? Using DNA and x-rays, Woodward was able to show that the male and female "parents" had been switched, so that the male now resided in the female sarcophagus and *vice versa*. X-rays also showed that all six had been executed, as evidenced by their broken necks. Using both mtDNA and nuclear DNA, it was determined that three related generations were represented by these mummies.

The mummy of king Seti II, Dynasty XIX

Positive identification and the construction of genealogies using DNA was Woodward's goal for the well-known but still mysterious 27 royal mummies dating from Dynasties XVIII through XXI. In ancient times, the royal mummies were treated badly, with tomb robbers often searching for valuables immediately after the deceased was interred. In Dynasty XXI, the High Priest decided to have the plundered and desecrated royal remains re-wrapped and interred in two guarded *caches*. When they were re-wrapped he identified the remains, probably correctly, re-writing their names on the wrappings. These mummies came to light again in the late 1800s and early 1900s.

The mummies of Ramesses III, Ramesses II, Seti I, Amenhotep I and Seknet-ra may be among the royal mummies housed at the Cairo Museum. During the removal of the mummies to better quarters in the museum, Dr. Woodward was given a rare opportunity to harvest detached tissue fragments from these mummies for DNA testing, though no invasive sampling, including endoscopy, was permitted by the Egyptian officials. At this time, Woodward also observed that Thutmoses IV had red hair (natural, not hennaed) and a pierced ear, and that his thoracic cavity contained packages of resin wrapped in linen. Woodward also discovered that Amenhotep I's clavicles had detached and been stuck behind the mask, perhaps an afterthought of some tired workers re-wrapping the mummy!

Professor Woodward was able to glean tissues from 11 of the mummies and sequence the DNA of seven of these individuals. He worked from a hypothetical "tree" to see if DNA evidence would support relationships between some of royals from late Dynasty XVII and early Dynasty XVIII. Ahmose I was supposed to have married his full sister, Seknet-re, which would mean they should share mtDNA (having the same mother) and some of the HLA alleles or nuclear DNA, if they had the same father. This was supported by the DNA evidence. It was assumed that Amenhotep I's mtDNA would be different from Ahmose's, as his mother was probably not part of the lineage. This too was borne out by Woodward's DNA findings. It is possible that Ahmose Nefertari may have been Amenhotep's mother.

Thutmosis I introduces new mtDNA. Was his mother, Seneseneb, a non-royal? The new mtDNA indicates that a pharoah did not necessarily inherit the throne through his mother's line.[1] Thutmosis shares a particular allele with Amenhotep I; conventional wisdom says they were not father and son but DNA evidence implies that they were. The other three mummies sampled do not fit closely with any of the remaining four Woodward sampled. And while this is the most extensive genealogical record (based on DNA) to date of New Kingdom royal mummies, it does not yet answer many questions—such as Tutankhamen's lineage.

Woodward's team, among others, has come tantalizingly close to sampling the tissues of Tutankhamen, "the national treasure" of Egypt. Perhaps permission will be granted once Tutankhamen's possible relations—including Smenkhare, Akhenaten, and Amenhotep III—are sampled and sequenced. The parentage of two fetuses found in Tut's tomb remain a mystery. They were miscarried at five months and eight months. Woodward has now sequenced the mtDNA and nuclear DNA from the older fetus, but genetic information from these two mummies will tell us more when it has been compared with other possible family members.

Now Woodward's team has embarked on yet another ambitious project—to build a reference database of DNA from 13,000 living individuals representing 500 modern populations, worldwide. They hope to compare ancient genome fragments to modern sequences to deduce past population movements and relationships between ancient and modern peoples. For more information, Woodward directed the audience to the project's website: `<http://molecular-geneology.byu.edu/>` and provided an e-mail address: `<molecular-geneology@email.byu.edu>`

NOTE

[1]It should be noted that the theory of a royal "heiress" determining the succession to the Egyptian throne has been rejected on other grounds as well. For discussion, see Gay Robins. 1983. "A Critical Examination of the Theory That the Right to the Throne of Ancient Egypt Passed Through the Female Line in the 18th Dynasty." *Göttinger Miszellen: Beiträge zur ägyptologischen Diskussion* 62:67–77; Gay Robins. 1993. *Women in Ancient Egypt*. London: British Museum Press, 26–27. [EDITOR]