Our Dogs DNA
What can geneticists do with it?

(first published in the Newsletter of the Society for the Preservation of Desert Bred Salukis)
and in **Sloughi World**

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During the past recent years, as a result of the development of techniques that enable scientists to study genetic material, many of us have been approached to provide samples of our dogs DNA. The legitimate question is then to ask: what do scientists study exactly with our dogs DNA? What do their studies tell us, what do they not tell us? How do they differ from one another, as different laboratories study different aspects of the DNA?

What is DNA?

All genetic information is built of DNA (Deoxyribonucleic Acid), a giant double helical molecule that consists of four distinct units, called bases, arranged in two parallel strands. These bases are A, T, C, G (Adenine, Thymine, Cytosine, Guanine). A interacts with T, C interacts with G. These interactions form bridges that hold the double stranded DNA together in a full mirror complement of each other, thus forming steps on a spiraling ladder. In essence, the DNA is a biological digital storage system that has four signals (the four bases) to code for proteins.

Altering the information encoded in the orderly arrangement of bases on the DNA requires altering the sequence of bases. Such an alteration is called a mutation. Such mutations can do a number of things. In many cases, mutations change a single base. Thus, instead of an A it will substitute that for a G, for example. Such base alteration rates typically range in the order of several hundreds or thousands of years, depending on the sequence in question.

The genome is all the inherited information or DNA carried from one generation to the next in a given being. Several genomes have thus far been sequenced: Human, Mouse, Rat, Dog, Chimpanzee and Chicken for example. A comparison revealed that dogs and humans share approximately 90% of their genes.

Geneticists study various aspects of the DNA, and in doing so, their studies obtain results that provide us with different insights. Here are some of the major areas of research.

Studies of the mitochondrial DNA:

The genetic information (DNA) of all mammals is stored predominantly in the nucleus of each cell. In addition, a small fraction of DNA is also found in mitochondria, the power plants of each cell. Mitochondrial DNA (mtDNA) is only inherited from the female to her offspring. Both male and female dogs inherit their mitochondrial DNA from their mothers. No male mtDNA contributes to the next generation. Therefore each male is a dead end with respect to inheritance of male mtDNA information. Analyzing the sequence of mtDNA provides thus exclusive information about the mother's side of a given dog- the mother, grandmother, great grandmother, etc… Since mtDNA, like any other DNA, encodes information in the arrangement of the four bases, one can compare the arrangements of these four bases to study similarities of animals that result from common inheritance. Thus, determinig the arrangement of the four bases (sequencing) of a stretch or segment of the mtDNA will provide genetic information about the inherited relatedness of dogs. Such an analysis would be comparable to ordering all of the letters on this page into a single line without any interruptions and comparing this with files

similarly obtained of pages from other books. Logically, only faithful copies of the page would show a high resemblance in the sequence of letters over such a long stretch.

Method of sampling: hairs and cheek swabs of males and females preferably not related and from known geographical origins.

The studies of the mtDNA of dogs tell us something about the origins of the female lines of a dog breed and its relationship with female lines of other breeds of dogs, various species of wolves or other wild Canids, for example. In other words it tells us something about the Natural History of dog breeds, how and where dog breeds evolved as a result of domestication thousands of years ago.
When looking at the pedigree of a dog, the mtDNA is provided by the female line inherited from the dam, her dam, etc all the way up to the very first female of that particular female line which is typically not represented in the pedigree, and may in some cases be as far in the past as hundreds or thousands of years.
Figure 1 shows the pedigree of a male dog, in which the female line that provides the mtDNA is highlighted in red. In our case Nibal's mtDNA is provided by Chalila and her uninterrupted previous female line. To access the mtDNA of bitches in the middle of the pedigree one would sample dogs of previous generations such as Fahel, for the mtDNA of Chamisa's line to Sa'ida, or Aswad for the mtDNA of Fehda's line to Biskra, and Damir for the mtDNA of Vahima's line and so on.

Usad male line (Y chromosome)
Damir
Vahima (female)
Aswad
Dubai (male)
Fehda
Biskra (female)
Fahel
Bouh (male)
G'Zal
Mrima (female)
Chamisa
Azhar (male)
Umaia
Sa'ida (female)
Nibal
Raisuli (male)
Emin
Kasira (female)
Ghali
Damir (male)
A'ssisa
Fehda (female)
Lycka
Azhar (male)
Usdan
Sa'ida (female)
Hannah
Badawi (male)
Tiri
Chalila female line (mitochondrial DNA)

Figure 1, example of a pedigree

Studies of the Y chromosome:

The Y chromosome is the chromosome that determines the maleness of a mammal. All males inherit a Y chromosome from their father and an X chromosome from their mother. All females inherit 2 X chromosomes, one from their mother, one from their father, they have no Y chromosomes (like their brothers) and therefore develop into females.

Method of sampling: cheek swabs are collected from male animals preferably not related and from known geographical origins.

Like the mitochondrial DNA studies above, the Y chromosome studies look at the Natural History of Canids. They look at the origins of the male lines of dogs and species of wolves and wild Canids, and compare them to see how they relate to each other, and ultimately how they relate to the findings regarding the female lines of wild and domesticated Canid populations.

In our pedigree example in figure 1, the Y chromosome is inherited through the male line highlighted in blue. In other words Usad and his previous uninterrupted male line provided the Y chromosome to the male dog Nibal.

To have access to other male lines in the middle of the pedigree one would need to sample brothers of the bitches in that pedigree, such as brothers of Lycka, Hannah, Tiri: these would give us the Y chromosomes of the dogs Ghali, Usdan and Badawi respectively.- or brothers of Chamisa, Umaia, Fehda for the sires G'Zal, Azhar and Dubai respectively and so on

This brings me to the question: why is the X chromosome in the bitches not studied? At each generation, a bitch inherits an X chromosome from her sire and an X chromosome from her dam. After a few generations, there is so much reshuffling, that there is currently no way of knowing which X chromosome is inherited only through the bitches, hence it cannot be used to trace patterns of inheritance over multiple generations.

Studies of the mitochondrial DNA and Y chromosome of dogs aim to study as good a cross section of various bloodlines or populations inherent to a breed or species as possible. Therefore representative samples for such studies are those of animals that are not too closely related. Such studies are also based on the geographical origins of the dogs or wild Canid species being studied. Well designed examples of such studies are provided by Dr. Peter Savolainen and collaborators, and others in the field.

DNA and Genetic diseases

Method of sampling: blood samples and cheek swabs of animals affected or not affected by the disease under study.

There are two main avenues in the study of genetic diseases in dogs. One avenue studies the genetic inheritance of dog diseases in specific breeds, the other studies genetic diseases in dogs and their relationships to similar diseases in people.

To track down a genetic disease in a specific breed, blood samples or cheek swabs are first taken from animals affected by the disease being studied and then compared to samples of animals of the same breed not affected by this disease and from lines with no animals affected by that disease. Careful comparison enables geneticists to find where the defect is to be found in the gene (s) responsible for that disease. Once the defect(s) is found a probe is developed that will find the defect gene(s) in dogs of the general population.

For example let us assume that the healthy and the affected sequences of bases are as follows (figure 2)

Healthy "clear" sequence of bases	Affected sequence of bases
A	A
C	C
G	***A***
A	A
T	T

Figure 2, ***A being the defect mutation***

A probe would enable a geneticist to find the animals clear of the disease, affected by the diseases, and the carriers of recessively inherited diseases, who themselves do not show any signs of the disease, but can pass it on (see figure 3)

Healthy "clear" sequence	Healthy "carrier" sequence	Affected sequence
A A	A A	A A
C C	C C	C C
G G	G ***A***	***A*** ***A***
A A	A A	A A
T T	T T	T T

Figure 3, ***A being the defect mutation***

This in turn helps geneticists develop specifics tests which discriminate affected dogs from carriers and from animals clear of that disease in a specific breed. Such genotyping tests are extremely useful for breeders who can then screen their breeding stock for that specific disease and make informed decisions when they select animals for a specific breeding such that none of the pups will be affected by that specific disease. For example crossing healthy "clear" dogs with healthy "carrier" dogs, can only lead to "clear" or "carrier" puppies, not to affected puppies. A good example is the genotyping tests for Progressive Retinal Atrophy available for various breeds of dogs. Such genotyping and careful selection can ultimately eradicate a genetic disease in a given population.

About 90% of the dog genome is similar to that of Man, and many diseases found in dogs are found in Man as well. This is an area of research which uses the dog as a model to understand inherited diseases in people. In this avenue affected dogs are bred with affected ones and produce affected puppies on which the disease in question can be studied. Good examples of this are the study of Narcolepsia (sudden sleep) in a dog breed with this inherited disease, which helps better understand the same disease in people. Another case is Progressive Retinal Atrophy named Retinitis pigmentosa in people.

Parental DNA/ DNA fingerprinting

Method of sampling: cheek swabs, hair or blood of animals under study

This is the part of the DNA collected and decoded to confirm parentage in breeds of dogs, such as are implemented by various kennel clubs including the American Kennel Club, and in Forensics.

The mitochondrial DNA is the most stable part of the DNA and it thus allows comparisons of relatedness over 1000 of years. For this very reason mtDNA is not useful to discriminate between kin.

Therefore geneticists interested in "dog forensics" will look at the most variable and mutating regions of the DNA, the non-sex linked chromosomes (autosomes). In doing so, they establish a fine grid of genetic information which enables them to establish who is related or not related to whom. In that scenario each individual has a unique DNA fingerprint, a unique combination of highly variable regions of the DNA- hence the expression DNA fingerprinting.

These studies enable us to tell whether a particular dog is in fact an offspring of the claimed sire or dam on his pedigree, or not. In Forensics it helps to match evidence on a crime scene to potential suspects.

DNA fingerprinting and breed characteristics

Method of sampling: cheek swabs of animals preferably closely related

These Studies use DNA fingerprinting to establish relatedness between inbred breeds of dogs as well as differences between breeds. Studying the hyper-variable DNA gives insight into the immediate past (100 generations or so). In comparison, analyzing mitochondrial DNA and Y chromosome gives insight into the past 150.000 thousand plus years.

DNA fingerprinting studies to establish breed characteristics are represented by the studies of Dr. Oestrander and collaborators.

DNA fingerprinting, mitochondrial DNA and Y chromosome studies provide mutually compatible analysis of relatedness with a different level of resolution: mtDNA may not show finer differences between dogs related for less than 5 generations; fingerprinting may randomly lump very distantly related dogs and mtDNA is needed to resolve such ambiguity.

Cloning

Cloning is the exact reproduction of a piece of DNA or of an entire being.

The method for cloning of an entire being is extremely complicated and involves many steps. The sheep Dolly and the Afghan hound Snuppy are the famous clones.

Hundreds of live cells of an individual are collected by a skin biopsy, the skin cells are processed and grown in culture. Hundreds of female donor eggs are extracted. The nucleus of the egg is squeezed out (very delicate technique involved) through a slit in the egg. An entire skin cell is injected into the donor egg through that same slit. An electric charge fuses the egg and the skin cell. The resulting cell is immersed in a chemical bath and starts to divide and grow into an embryo. After several days several such embryos are transferred into a surrogate bitch, hoping that at least one will make it to term. Hundreds of attempts are needed and a fortune will be spent before any success is achieved.

Cloning of an entire being requires cells that are alive. Can such cloning be made from dead cells from cheek swabs? At the moment, NO. Will it be possible to clone from dead cells of cheek swabs in some 20 years? Probably not. Will it be possible in some 100 years? Who knows, but it will still be very costly…Will it be possible to use stored DNA after 100 years? If yes, it could in fact help your breed in a century from now……but who can say?