

Chair : **Sree Kanthaswamy (SK)**, School of Veterinary Medicine, University of California Davis, Davis, USA.

Approximately 150 attended the animal forensic genetics workshop.

Of the nine standing committee members Drs Joy Halverson and Steve Fain were not able to attend the conference.

The forensic workshop was called to order at 8 am and was conducted in two sessions chaired by Dr Sree Kanthaswamy (SK). The first session consisted of a brief introduction to animal forensic science and the ISAG Animal Forensic Standing Committee by SK followed by four lectures on:

- "Analysis of mitochondrial DNA in dogs: the pattern of geographical differences among populations across the world" by Dr Peter Savolainen, School of Biotechnology, Royal Institute of Technology, KTH, Albanova, SE-106 91 Stockholm, Sweden;
- "Developmental validation of a sensitive and discriminating panel of novel canine STR markers for forensic testing" by Beth Wictum et al., Veterinary Genetics Lab, UC Davis, CA, USA;
- 3. "The effect of inbreeding on match probability calculations: towards a consensus approach for non-human systems" by Dr Rob Ogden, Wildlife DNA Services and Tepnel Research Products and Services, Edinburgh, UK, and,
- 4. "Animal Forensic Science in Argentina" by Dr Guillermo Giovambattista, IGEVET, Facultad de Ciencias Veterinarias, Universidad Nacional de La Plata. Calle, Argentina.

See Abstracts in Appendix 1.

The second session of the workshop included a formal meeting with the following agenda:

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- 1. Forensic and non- forensic testing (definition);
- 2. Present Guidelines from Budowle et al. 2005;
- 3. Proficiency tests v. Comparison tests;
- 4. Accreditation/Certification;
- 5. Databases;
- 6. Standing committee members;



1. Forensic and non- forensic testing

At the Brazil meeting in 2006, it was clear that many ISAG affiliated labs were not sure whether they were performing forensic services or not. To help answer this question, the standing committee had to define what animal forensic genetics entailed to differentiate between forensic and non-forensic application in genetic testing and reporting. Based on the feedback and approval by the workshop participants, animal forensic genetics was defined as "The application of relevant genetic techniques and theory to legal matters, or enforcement issues, concerning animal biological material".

It was also moved and seconded that there be a minimum set of guidelines under which laboratories performing forensic applications operate.

2. Present Guidelines from Budowle et al. (2005) were presented.

For minimum guidelines for forensic applications and to improve lab performance refer to Budowle et al. (2005) which outlines standard criteria in human forensics that can be easily adopted in animal forensics. This motion was also carried. This augmentation of the minimum standard may result in a two layer system which would enable ISAG-affiliated "strictly-routine" parentage laboratories to upgrade existing protocols to those of forensic standards.

3. Proficiency tests v. Comparison tests

Definition proficiency test (PT): an essential element of a successful QA/QC system. "It is used periodically to demonstrate the quality performance of the DNA typing laboratory" - Budowle et al. (2005), and PTs can be required for accreditation (see below). In the next two years the standing committee will establish strategies to facilitate the performance of PTs among member labs including the use of outside agencies.

4. Accreditation/Certification

It was also moved that forensic service providing laboratories acquire internationally recognized accreditation. Because ISAG will not become involved in the accreditation of service genotyping laboratories or institutions (furthermore, ISAG does not regard participation in its Comparison Tests as indicating that a lab is internationally accredited), within the next two years the standing committee will have to determine which agency and what type/level of accreditation would be optimal for animal forensic labs affiliated with ISAG.



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5. Databases, next phase publishable guidelines (Sample collection, handling, protocols, databases, allelic ladders, nomenclature, coordination of reports).

SK discussed an example of a recently assembled domestic dog STR database that is available in the US National Institute of Standards and Technology website http://www.cstl.nist.gov/biotech/strbase/. This database and associated publication by Kanthaswamy et al. (submitted) are a result of collaboration among several ISAG members including Joy Halverson, QuestGen Forensics, Eric Johnston, MMI Genomics, Mikko Koskinnen, Finnzymes, Andreas Hellmann; Budeskriminallampt, Germany, Steve Fain, US Fish and Wildlife National Forensic Lab, SK, California Nat. Primate Research Center/Dept. of Anthropology, UC Davis; it is a good example for new public datasets that can be publicly accessed and shared freely among member and non-member labs. The standing committee is willing to aid in the development of genetic databases to facilitate analysis and interpretation of data and publication of forensic research and casework, as well as guidelines for sample collection, handling, protocols, databases, allelic ladders, nomenclature, and coordination of reports. For submitting research casework articles for peer-reviewed publications, several journals were recommended including the Journal of Animal Genetics, J Forensic Science, International J of Legal Medicine, Forensic Science International and Legal Medicine. The standing committee members may assist member labs with publications as it is their duty to answer inquiries relevant to animal forensic genetics from ISAG members, other scientists and members of the public.

6. Standing committee members

The second session confirmed the line-up of standing committee members for another two years since membership in ISAG standing committees. Romy Morrin O'Donnel of the Irish Equine Center proposed that the ISAG parentage labs should also be represented in the standing committee. The motion was discussed, seconded and voted. An additional member, Leanne van de Goor of the Dr. van Haeringen Laboratorium was elected by the membership. It must be noted that Ms van de Goor will be the second person from the Dr. van Haeringen Laboratorium in the present standing committee and there are no rules against having two representatives from the same lab in the ISAG standing committees. Therefore the current membership includes Drs Wim van Haeringen, Ross McEwing, Mikko Koskinen, Steven Fain, Joy Halverson, Paolo Garafano, Andreas Hellmann and SK (Chair) as well as Ms Beth Wictum and Ms van de Goor.



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Tasks for the ten-member Animal Forensic Genetics Standing Committee for the next two years will include:

- 1. In the next two years the standing committee will develop a strategy for implementing PTs among member labs.
- 2. The standing committee will have to determine which agency and what type/level of accreditation would be optimal for animal forensic labs affiliated with ISAG.
- 3. The standing committee will be willing to aid in the development of genetic databases.
- 4. The standing committee members may assist member labs with publications as it is their duty to answer inquiries relevant to animal forensic genetics from ISAG members, other scientists and members of the public.



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Appendix 1

Analysis of mitochondrial DNA in dogs: the pattern of geographical differences among populations across the world

Savolainen, P

Abstract: In a study aimed at unravelling the geographical origin of the domestic dog we have analysed 582 base pairs of mitochondrial DNA in 1,576 dogs from across the world. In this material we find distinct differences in the representation of haplotypes among different geographical regions, and among dog breeds. For example, there are a several haplogroups that are unique to dogs and breeds from East Asia, probably because dogs originated in this region, and many dog breeds belong to a distinct set of a few haplotypes. Interestingly, the European dog population, which represent the largest number of breeds and morphological diversity, is among the least diverse populations for mitochondrial DNA, probably because it was formed via several genetic bottlenecks as the population was derived from East Asia. These patterns are here presented and the possibility for forensic inferences discussed.

Developmental Validation of a Sensitive and Discriminating Panel of Novel Canine STR Markers for Forensic Testing

Wictum EJ, Kun TJ, Truong NV, Lindquist CD, Collins JA and Wong A

Abstract: The molecular analysis of forensic evidence has revolutionized the criminal justice system. Its strength has been used to both incriminate the guilty and free the innocent. While analysis of human biological material has been extensively vetted in courtrooms worldwide, forensic analysis of animal DNA is still gaining acceptance. As the oldest domesticated species, dogs (Canis lupus familiaris) inhabit 39% of households in America and, after humans, are the species of greatest forensic interest. Numerous genetic markers have been employed to individualize canids for parentage verification, breed identification, phylogeny, and diversity assessment; however, forensic analysis requires the application of more stringent marker selection criteria. To address the lack of a standardized canid forensic panel that meets those criteria, tools were developed to mine the 7x dog genome sequence data. Fifteen unlinked highly discriminating tetranucleotide-repeat markers were identified and assembled with a sex-determination marker into a multiplex capable of generating a full profile with less than 0.1 ng of nuclear DNA. This panel has the potential to be not only a valuable tool for the emerging field of veterinary forensic science but also demonstrates utility for parentage verification in highly inbred dog populations and for phylogenetic analyses of unique canid populations.



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The effect of inbreeding on match probability calculations: towards a consensus approach for non-human systems

Ogden, R

Abstract: Non-human DNA profiling is becoming widely used to match trace evidence samples to individual animals involved in legal investigations. The majority of techniques, standards and statistical approaches can be directly transferred from human forensic genetics however certain species differences require special consideration. The calculation of match probabilities for identical human DNA profiles can account for sub-population structure (F_{ST}) and levels of known relatedness (familial matching), but generally ignores inbreeding at the level of the individual (F_{IS}), as this is rarely significant in human populations. In contrast, both domestic and wild animals routinely exhibit very high levels of inbreeding, due to captive or natural breeding systems, and this potentially affects the probability of observing matching DNA profiles, often biasing the results in favor of the prosecution. Corrections for inbreeding and alternatives to match probability equations have been suggested, but there is currently no agreed method for how to account for F_{IS} across a wide range of scenarios. This presentation reviews existing proposals and presents new research on this topic with the aim of initiating discussions and finding consensus.

Animal Forensic Science in Argentina

Giovambattista, G

Abstract: Argentina is one of the major countries that produce, commercialize and export livestock for food, work and sport and pet animals for companionship and enjoyment. For this reason, forensic science has a great impact in this country. In this context, animal forensic cases are common, and their resolutions are mandated by the government, breeder registries, and farmers. Livestock theft is a common criminal activity that causes the loss of several millions of dollars each year and poses a serious human and animal health risk. Additionally, cases of fraudulent trade of livestock and pets, horse doping, contamination or adulteration of human food and animal feed and illegal trafficking of wildlife are also common crimes. To prevent and solve these types of activities, we have designed and implemented The Prevention and Resolution of Livestock Theft Cases in the Buenos Aires Province Plan which includes the creation of a DNA sample library and database representing local animal populations, and a sample management and analysis system. Furthermore, experimental methods have been developed for genotyping pristine and degraded samples, while statistical methods have been used for resolving typical forensic cases in Argentina.