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Standard for Training in Statistical Calculations Used for Forensic Short Tandem Repeat (STR) DNA Data

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Foreword

This standard defines the minimum requirements ~~for~~to be met in a forensic DNA analyst training program ~~in~~for the application of statistics to autosomal and Y-STR DNA profiling results. The aim is to provide a framework for quality training ~~that will result~~resulting in consistency within a laboratory and in the forensic DNA community.

~~This document was revised, prepared, and finalized as a standard by the DNA Consensus Body of the AAFS Standards Board. The draft of this standard was developed by the Human Forensic Biology Subcommittee of the Organization of Scientific Area Committees (OSAC) for Forensic Science.~~

The American Academy of Forensic Sciences established the Academy Standards Board (ASB) in 2015 with a vision of safeguarding Justice, Integrity and Fairness through Consensus Based American National Standards. To that end, the ASB develops consensus based forensic standards within a framework accredited by the American National Standards Institute (ANSI), and provides training to support those standards. ASB values integrity, scientific rigor, openness, due process, collaboration, excellence, diversity and inclusion. ASB is dedicated to developing and making freely accessible the highest quality documentary forensic science consensus Standards, Guidelines, Best Practices, and Technical Reports in a wide range of forensic science disciplines as a service to forensic practitioners and the legal system.

This document was revised, prepared, and finalized as a standard by the DNA Consensus Body of the AAFS Standards Board. The draft of this standard was developed by the Human Forensic Biology Subcommittee of the Organization of Scientific Area Committees (OSAC) for Forensic Science.

Questions, comments, and suggestions for the improvement of this document can be sent to AAFS-ASB Secretariat, asb@aafs.org or 410 N 21st Street, Colorado Springs, CO 80904.

All hyperlinks and web addresses shown in this document are current as of the publication date of this standard.

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Keywords: *random match probability, likelihood ratio, DNA interpretation, statistics, training, DNA standard*

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Standard for Training in Statistical Calculations Used for Forensic Short Tandem Repeat (STR) DNA Data

1 Scope

This standard ~~outlines~~provides the ~~minimum~~ requirements for a forensic DNA laboratory's training program ~~in~~for the use of statistical calculations and values reported for forensic autosomal and Y short tandem repeat (STR) DNA data.

2 Normative References

The following reference is indispensable for the application of the standard. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ANSI/ASB Standard 022, *Standard for Forensic DNA Analysis Training Programs*[1].

3 Terms and Definitions

For purposes of this document, the following definitions apply.

~~**3.1**~~
~~**avuncular index**~~
~~The likelihood ratio that evaluates the hypothesis that the tested individual is the biological uncle (or aunt or niece or nephew) of the profile donor versus the hypothesis that the tested individual is unrelated to the profile donor.~~

~~**3.2**~~**3.1**
Combined Probability of Exclusion
CPE
The probability that a randomly selected individual would be excluded as a contributor to the DNA mixture. If the single-locus exclusion probabilities are independent, and if P_j is the probability of exclusion at locus j, then the combined probability of exclusion is $1- \prod_j (1 - P_j)$.

~~**3.3**~~**3.2**
Combined Probability of Inclusion
CPI
The probability that a randomly selected individual would not be excluded (i.e., is included) as a contributor to the DNA mixture. If the single-locus exclusion probabilities are independent, and if P_j is the probability of exclusion at locus j, then the combined probability of inclusion is $\Pi_j (1 - P_j)$.

~~**3.4**~~**3.3**
counting method
A method for estimating genotype, sequence, or haplotype frequency by direct counting of the number of times a genotype, sequence, or haplotype is observed in a database and dividing by the number of samples in that database. This method is commonly used for estimating frequencies in populations for mitochondrial DNA and Y-STR DNA haplotype results.

[1] Available from: www.aafs.org/academy-standards-board.

~~3.5~~3.4
haplotype
A set of linked DNA variations, or polymorphisms, that tend to be inherited together (e.g., commonly used for human Y-chromosome or mitochondrial analysis). A haplotype can refer to a combination of alleles or to a set of single nucleotide polymorphisms (SNPs) found ~~on the same~~along a single chromosome that tend to be inherited together.

~~3.6~~3.5
Hardy-Weinberg equilibrium
A state in which allele and single locus genotype frequencies do not change (on average) from one generation to the next in a population. When alleles in a population are independent, allele and genotype frequencies are related through the Hardy-Weinberg principle: for a locus with 2 alleles P and Q at frequencies of p and q, homozygotes for P are found at frequency p^2, homozygotes for Q are found at a frequency of q^2, and heterozygotes are found at a frequency of 2pq. Use of the theta correction ~~removes~~allows the ~~need to assume~~assumption of Hardy-Weinberg equilibrium in the population for which a frequency database is constructed. See **theta correction**.

~~3.7~~3.6
Identical by Descent
IBD
Identical alleles that are copies of the same ancestral allele without mutation; this is a subset of identical by state (IBS).

~~3.8~~3.7
Identical by State
IBS
Identical alleles that may or may not be copies of the same ancestral allele.

~~3.9~~3.8
inbreeding
Mating of two persons who are more closely related than if they were chosen at random. It increases the frequency of homozygous genotypes above the expected for a randomly mating population in Hardy-Weinberg equilibrium.

~~3.10~~3.9
kinship analysis
Comparison of genetic profiles of two or more individuals to evaluate alternative degrees of biological relatedness.

~~3.11~~3.10
Likelihood Ratio
LR
A likelihood ratio is defined by a ratio of two conditional probabilities: the ~~probability~~probabilities of the evidence given each of two mutually exclusive and competing propositions. ~~In forensic science applications, the likelihood ratio is used as an expression for the meaning of scientific evidence and as measure for its value.~~
ASTM E1732-12[mod][2]

[2] Available from ASTM, at www.astm.org

~~3.12~~3.11
linkage equilibrium
~~Linkage equilibrium describes the situation in which the haplotype frequencies~~**/disequilibrium**
When two or more genetic loci appear to segregate randomly in a given population ~~have the same value that,~~ they ~~would have if the alleles~~are at ~~each locus were combined~~equilibrium. When the loci do not segregate randomly, they are at ~~random. If both Hardy-Weinberg and linkage equilibrium hold, then random match probabilities may be multiplied over loci~~disequilibrium.

~~3.13~~3.12
mutation rate
The relative frequency at which mutations have been observed at a specific genetic locus; generally estimated as the number of mutations observed in parent-offspring pairs divided by the total number of pairs examined.

~~3.14~~3.13
Paternity or Maternity Index
PI/MI
~~The likelihood~~Likelihood ratio ~~that evaluates~~in which the ~~hypothesis~~numerator is the probability of the DNA data given the proposition that the tested individual is the biological ~~mother or biological father~~parent of the profile donor ~~versus the hypothesis~~, and the denominator is the probability of the DNA data given the proposition that the tested individual is unrelated to the profile donor.

3.14
proposition
A statement that is either true or false. In the context of evidence evaluation, propositions should be formulated in pairs: the paired propositions should be mutually exclusive (i.e., both cannot be correct at the same time) and exhaustive in the context of the case (i.e., one should not consider all propositions as default, but only those that are thought to be of interest to the court).

3.15
Random Match Probability
RMP
The probability of randomly selecting an unrelated individual from the population who could be a potential contributor to an evidentiary profile.

3.16
reverse parentage
Likelihood ratio in which three individuals have been profiled—the child and two questioned biological parents. More specifically, the probability of observing the data if the child is the biological child of the alleged parents, divided by the probability of observing the data if ~~two~~

~~randomly selected people are the parents of~~the tested individuals are unrelated to the child.

3.17
sibship index
Likelihood ratio in which the numerator is ~~conditioned on the hypothesis that a sibling~~the probability of the ~~source of the questioned profile in a specimen~~DNA data given the proposition that the two tested individuals are biological siblings, and the denominator is ~~conditioned on the hypothesis~~the probability of the DNA data given the proposition that ~~an~~the two tested individuals are unrelated ~~individual is the source~~.

3.18
source attribution
A declaration which identifies an individual as the source of the DNA that produced an evidentiary single-source or deduced contributor profile. This statement is based on a statistical estimate that meets or exceeds a laboratory-defined threshold.

3.19
theta correction
A ~~value~~method used to adjust statistical calculations that rely on population databases to correct for substructures within populations.

4 Requirements

4.1 General

ANSI/ASB Standard 022, *Standard for Forensic DNA Analysis Training Programs* shall be used in conjunction with this document because ANSI/ASB Standard 022 provides the foundational training program requirements upon which additional specific requirements, such as this document, will be based.

The laboratory's training program shall include all requirements applicable to the work conducted by the laboratory and by the individual in training.

4.2 Knowledge-Based Training

4.2.1 At a minimum, the knowledge-based portion of the training program shall require review of the following:

a) the laboratory’s protocols for statistical applications;

b) the laboratory’s applicable validation studies;

c) literature used to support validation;

~~c)~~d) literature used to support specific calculations and their use in appropriate circumstances;

~~d)~~e) applicable literature as assigned by the trainer;

~~e)~~f) literature on the effects of cognitive bias in decision-making processes associated with statistical calculations used for forensic STR DNA data.

4.2.2 The knowledge-based training component of the laboratory's training program shall provide the trainee with ~~a~~ basic ~~understanding~~instruction of probability and statistics applied to autosomal and Y-STR data to include, at minimum, the following topics.

a) *Population Genetics*

1) laws of Mendelian genetics (the law of segregation and the law of independent assortment);

2) Hardy-Weinberg equilibrium, assumptions, and related evolutionary forces;

3) linkage equilibrium/disequilibrium;

4) use of theta correction to adjust for inbreeding and population substructure~~;~~.

b) *Statistical Foundations*

1) frequency;

2) probability;

3) odds;

4) the laws of probability (e.g., the addition rule and product rule);

5) Bayes' theorem~~;~~.

6) Sources of uncertainty (e.g.~~,~~ modelling uncertainty and sampling variability~~)~~).

c) *Population Allelic Frequency Databases*

1) population database size relative to the population size;

2) sample collection, to include:

i) number of samples,

ii) how population group was determined,

iii) how the database was created, maintained and reviewed,

iv) sampling uncertainty;

3) differences in allele frequencies observed between population databases;

4) mechanisms to account for alleles not observed in the database.

d) *Suitability of data for statistical application*

1) when to perform statistical analyses and which statistical calculation is validated for the type of data obtained and comparison performed;

2) instruction on which loci to include in the statistical analyses when the following circumstances are observed, at a minimum:

   i) no allelic data,

   ii) partial allelic data,

   iii) tri-alleles, duplications/triplications, null alleles, and mutations,

   iv) STR artifacts.

e) *Statistical analysis for autosomal STR data*

1) proper use of statistical calculations to include derivation and applicable equations;

2) statistical calculation method(s) in use by the laboratory, to address:

   i) population substructure,

   ii) mutation rates,

   iii) known relatedness;

3) equation(s) in use by the laboratory, to include:

   i) combined probability of inclusion,

   ii) combined probability of exclusion,

   iii) random match probability (or modification),

   iv) likelihood ratio including formulating propositions;

4) operation of the software program(s) in use by the laboratory, including the underlying equations and review of the output data;

5) source attribution statements~~, if applicable;~~

6) limitations and assumptions of statistical method(s) ~~in use~~used by the laboratory.

f) *Statistical analysis for Y-STR data*

1) detailed instruction on the calculation of haplotype frequencies using ~~the~~ counting ~~method~~methods, to include:

   i) consideration of the differences between the loci that the database samples are typed with and the loci in the amplification kit used by the laboratory,

   ii) instruction on confidence intervals, Y-STR profile probabilities and Y-STR match probabilities,

iii) consideration of the effect of the database source/population and size;

iv) instruction on incorporating Y-STR mutation rates;

~~iii)~~v) instruction on combining statistical values from autosomal and Y-STR data;

2) operation of the software program(s) ~~in use~~used by the laboratory, ~~including the underlying equations and review of the output data.~~to include:

i) underlying equations,

ii) review of the output data,

iii) search types (reduced, masked, transient).

g) *Kinship analysis*

1) statistical calculations for kinship associations including derivation and use, to include, as applicable:

i) the difference between alleles that are identical by state (IBS) or identical by descent (IBD),

ii) how to set-up competing propositions for kinship calculations~~;~~,

iii) how to account for mutations in the kinship calculations;

2) determination of appropriate calculation for the case (identifying the unknown in the relationship scenario), to include:

i) maternity or paternity index,

ii) reverse parentage,

iii) sibship, ~~avuncular~~aunt or uncle, or single grandparent index,

iv) complex family reconstruction.

4.3 Practical Training

4.3.1 The practical component of the laboratory's training program shall provide the trainee with ~~sufficient~~ practical instruction for the trainee to obtain the skills for calculating statistical values ~~for DNA data~~ used by the laboratory~~, to include, at minimum the components in 4.3.2 through 4.3.4:~~.

4.3.2 At a minimum, the practical portion of the training program shall include the observation of a trained analyst performing the processes ~~at least once or until clearly understood~~ with exercises representative of the range, type, and complexity of DNA data from routine casework or database samples processed by the laboratory, at least once or until clearly understood.

4.3.3 Practical exercises shall be representative of the range, type, and complexity of routine DNA data from casework samples processed by the laboratory. Practical exercises shall include the following:

a) the application of statistical analysis to the laboratory's own data;

b) hand calculations for the following, as appropriate: RMP (or modification), single source LR, CPI/CPE, and kinship analysis likelihood ratios;

c) exercises to understand the derivation of the equations involved in the calculation for ~~the following, as appropriate:~~ parentage and kinship analysis likelihood ratios, as appropriate.

4.3.4 The practical exercises performed shall be sufficient to demonstrate the trainee's ability to follow the laboratory's protocols and produce ~~appropriate~~accurate statistical values.

4.4 Competency Component

4.4.1 General

The competency component of the laboratory's training program shall ~~include~~demonstrate knowledge-based and practical competency in the laboratory's protocols for statistical applications. The format of the test(s) and the criteria for passing the competency test(s) shall meet Section 4.3 of the ANSI/ASB Standard 022, *Standard for Forensic Training DNA Analysis Training Programs.*

4.4.2 Knowledge-Based Competency

As applicable to the trainee's job responsibilities, the trainee shall successfully complete (as defined by the laboratory's policy) a knowledge-based test covering the critical information obtained during the training on case record management, forensic DNA report writing, and performing technical and administrative reviews. The test(s) shall cover, at a minimum, the topics outlined in 4.2 and its subsections.

4.4.3 Practical Competency

The trainee shall successfully complete (as defined by the laboratory's policy) a practical competency test covering each of the statistical applications the trainee will be independently authorized to perform. DNA data from samples representative of the range, type, and complexity for which the trainee will be authorized to perform statistical calculations shall be included in the practical competency test~~.~~(s).

5 Conformance

In order to demonstrate conformance with this standard, the laboratory shall meet the requirements outlined in ~~section~~Section 5 of ANSI/ASB Std 022, *Standard for Forensic DNA Analysis Training Programs* and all the requirements set forth in this document.

Annex A
(informative)

Bibliography

The following information provides a list of the resources that may assist the DNA technical leader in defining the breadth and scope of the materials to be reviewed by the trainee. This list is not meant to be all inclusive.

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