ASB Standard 078, First Edition
~~2022~~2023

Standard for Training in Forensic Autosomal Short Tandem Repeat (STR) ~~Data~~ and Y-STR DNA Data Interpretation and Comparison

Standard for Training in Forensic Autosomal Short Tandem Repeat (STR) ~~Data~~ and Y-STR DNA Data Interpretation and Comparison

ASB Approved Xxxxx ~~2022~~2023

ANSI Approved Xxxxxx ~~2022~~2023

410 North 21st Street
Colorado Springs, CO 80904

This document may be downloaded from: www.aafs.org/academy-standards-board

Foreword

This standard defines the minimum requirements to be met in a forensic DNA analyst training program for autosomal STR data and Y-STR data interpretation and comparison. The aim is to provide a framework for quality training that will result in consistency 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 Biological Data Interpretation and Reporting 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.

Questions, comments, and suggestions for the improvement of this document can be sent to AAFS-ASB Secretariat, asb@aafs.org or 401 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.

ASB procedures are publicly available, free of cost, at www.aafs.org/academy-standards-board.

Keywords: *training, interpretation, mixture deconvolution, comparison, DNA standard, autosomal STR, Y-STR*

Table of Contents *(to be finalized prior to publication)*

Standard for Training in Forensic Autosomal Short Tandem Repeat (STR) ~~Data~~ and Y-STR DNA Data Interpretation and Comparison

1 Scope

This standard ~~defines~~outlines the minimum requirements to be met in a forensic DNA analyst training program for autosomal and Y-STR data interpretation and comparison. This standard excludes training for DNA sequencing including the derivation of STR profiles from DNA sequencing 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*~~, First Edition, 2019~~[~~a~~]. [b].

3 Terms and Definitions

For purposes of this document, the following definitions apply.

3.1
analytical threshold
The minimum height requirement at and above which detected peaks on a STR DNA profile electropherogram can be reliably distinguished from instrument background noise; peaks above this threshold are generally not considered noise and are either artifacts or true alleles.

3.2
degradation
The fragmenting, or breakdown, of DNA by chemical~~,~~ or physical means.

3.3
drop-in
~~Allelic peak~~Peak(s) in an electropherogram that are not reproducible across multiple independent amplification events.

3.4
drop-out
Failure of an otherwise amplifiable allele to produce a signal above analytical threshold because the allele was not present or was not present in sufficient quantity in the aliquot that underwent PCR amplification.

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

3.5
inclusion
A conclusion for which an individual cannot be excluded as a potential contributor of DNA obtained from an evidentiary item based on the comparison of known and questioned DNA profiles (or multiple questioned DNA profiles to each other); a statement of inclusion does not confirm that an individual is a source of the DNA.

3.6
inconclusive
A statement provided as the conclusion when testing results are insufficient or lacking in quality and/or quantity, as defined by the laboratory, for comparison purposes; the data are inadequate to draw any meaningful conclusions.

3.7
inhibition
~~The act of interfering~~Active interference with or ~~preventing~~ prevention of the synthesis of DNA during ~~the amplification process of~~ the polymerase chain reaction (PCR).

3.8
match
When used in a DNA testing report, a match refers to genetic profiles that show the same types at all loci tested in common; a match statement does not confirm that an individual is the source of the DNA.

3.9
mixture
DNA typing results originating from two or more individuals.

3.10
mutation
A change in DNA sequence; an alteration or change of an allele at a particular locus resulting in genetic inconsistency between a biological or cellular parent and offspring.

3.11
off-scale STR data
Data produced when the emitted fluorescence from the PCR products being measured saturates the detector in an electropherogram; may result in flat-topped peaks for STR alleles and pull-up peaks in one or more color channels corresponding to the off-scale peak.

3.12
peak height ratio
The relative ratio of two peaks at a given locus ~~in~~. Mathematically, the ratio may be calculated in two ways: 1) the peak height (or area) of the longer length allele divided by the peak height (or area) of the shorter length allele, or 2) The shorter peak height (or area) divided by the taller peak. This is commonly expressed as a ~~diploid~~percentage.

~~3.11~~3.13
preferential amplification
A situation where one allele of a heterozygous ~~single-source sample~~pair at a locus is amplified by PCR with greater efficiency than the other allele.

~~3.12~~3.14
single source
DNA typing results originating from one individual.

~~3.13~~3.15
stochastic threshold
The peak height value in a DNA electrophoretic profile above which it is reasonable to assume that, at a given locus, allelic drop-out of a sister allele in a heterozygous pair has not occurred in a single source DNA sample; due to the possibility of shared alleles in mixed samples, the presence of allele peaks above the stochastic threshold is no guarantee that allele drop-out did not occur in mixed DNA sample profiles.

~~3.14~~3.16
stutter
An artifact of polymerase chain reaction (PCR) amplification typically observed one or more repeat units smaller or larger than a short tandem repeat (STR) allele in a DNA electrophoretic profile, may result from strand slippage during PCR amplification. A stutter peak is generally of lower relative fluorescence units (RFU) than the allele peak.

~~3.15~~3.17
tri-allelic pattern
The detection of three alleles in one individual at a particular short tandem repeat (STR) locus.

3.18
variant allele
A non-standard form of an allele due to a point mutation, an insertion or a deletion relative to other commonly seen alleles.

4 Requirements

4.1 General

~~Based upon the laboratory procedures, some of the requirements in this section may be omitted from the training program.~~

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 forensic autosomal and Y-STR data interpretation and comparison;

b) the laboratory's applicable validation studies;

c) literature used to support validation;

d) literature used to support the laboratory's interpretation and comparison protocol;

e) applicable literature as assigned by the trainer.

f) literature on the effects of cognitive bias in decision-making processes associated with forensic DNA analysis.

4.2.2 The knowledge-based training component of the laboratory's training program shall provide the trainee with ~~a~~ basic ~~understanding of~~instruction on the steps for forensic autosomal and Y-STR data interpretation~~.~~ and comparison. The training shall provide instruction on the interpretation ~~parameters~~and comparison criteria used by the laboratory, how the ~~parameters~~criteria were determined by the laboratory and any limitations of the ~~laboratories~~laboratory's validation studies (such as mixtures with number of contributors above what the laboratory considered during validation). The training shall also address documentation requirements of decisions made during the interpretation ~~process.~~and comparison process, to include a basic understanding of the risks of bias and potential for human error. It is critical the training includes manual interpretation and comparison even when software tools may be used. The training shall include, at minimum, the following topics, in 4.2.2.1 through ~~.~~4.2.2.3.

4.2.2.1 Quality control indicators:

a) positive controls;

b) negative controls;

c) internal lane standards;

d) primer peak;

e) allelic ladder.

4.2.2.2 Data suitable for interpretation and/or comparison:

a) factors in data interpretation:

1) peak height thresholds:

i) analytical threshold,

ii) stochastic threshold;

2) artifacts:

i) drop-in/drop-out,

ii) stutter (forward and backward),

iii) spike,

iv) pull-up,

v) other [e.g., (-A), dye blobs];

3) peak height ratios;

4) PCR inhibition;

5) DNA degradation;

6) preferential ~~and differential~~ amplification;

7) allele sharing (for mixtures);

8) data too limited and/or too complex

~~7)~~9) other considerations (e.g., mutations, tri-allelic patterns, ~~microvariants~~variant alleles, off-scale STR data).

b) requirements for single source data interpretation and comparison;

c) requirements for mixture data interpretation ~~including mixture sample types and number of contributors~~and comparison;

~~a) limitations of mixture interpretation (e.g., if the laboratory is validated to interpret four person mixtures, the trainee needs to be trained on four-person mixture interpretation and understand why the laboratory does not interpret mixtures with more than four contributors. See 4.2.1-b.);~~

d) considerations of mixture interpretation and comparison;;

1) mixture assumptions:

i) determination of number of contributors,

ii) appropriate conditioning/assumption of expected (known) individuals,

iii) estimating the ratio of contributors in mixtures,

iv) biological relatives in mixtures;

~~1) mixture considerations:~~

~~i) stutter including how stutter can mask or elevate a minor component in a mixture,~~

~~ii) allelic drop-in/drop-out,~~

~~iii) allele sharing in mixtures on apparent contributor ratios,~~

~~iv) inhibition and degradation,~~

~~v) tri-allelic patterns, mutations, etc.;~~

~~vi) data too limited, or too complex.~~

2) mixture deconvolution:

i) major component(s),

ii) minor component(s),

iii) foreign component(s~~);~~) (i.e., alleles not belonging to an assumed contributor)

3) probabilistic genotyping (if utilized by the laboratory):

i) scientific principle of probabilistic model,

ii) hypothesis development,

iii) statistical methodology,

iv) limitations of software;

e) limitations of mixture interpretation and comparison (e.g., if the laboratory is validated to interpret four person mixtures, the trainee needs to be trained on four-person mixture interpretation and comparison and understand why the laboratory does not interpret mixtures with more than four contributors.)

~~d)~~f) Comparison of evidentiary data to reference data (as applicable):

1) match,

2) consistent,

3) inclusion/cannot be excluded,

4) exclusion,

5) inconclusive.

~~**1.1.1.1** Data unsuitable for interpretation and comparison:~~

~~a) Data too limited.~~

~~b) Data too complex.~~

4.3 Practical Training

4.3.1 ~~The practical component of the~~The laboratory's training program shall provide the trainee with sufficient practical instruction for the trainee to obtain the skills for forensic autosomal and Y-STR data interpretation and comparison protocol(s) used by the laboratory ~~to include, at minimum all components in 2~~.

~~1.1.2 The protocol(s)~~At a minimum, the practical portion of the training program shall ~~be observed by the trainee~~include the observation of a trained analyst performing the processes at least once~~.~~

~~NOTE This can be done by direct observations and supplemental case file review~~ or until ~~the protocols are~~ clearly understood~~.~~

~~4.3.2 Practical~~ with exercises ~~shall be~~representative of the range, type, and complexity of routine casework or database samples processed by the laboratory.

~~4.3.2~~4.3.3 At a minimum, the practical portion of the training program shall include hands-on exercises representative of the range, type, and complexity of routine casework or database samples processed by the laboratory that include using the laboratory's own data.

~~4.3.3~~4.3.4 The number and quality of samples interpreted by the trainee shall include manual data review and automated data analysis methods, including all validated software programs in use by the laboratory, as applicable~~, and~~. The training shall be appropriate to demonstrate the trainee's ability to follow the laboratory's forensic autosomal and Y-STR data interpretation and comparison protocol(s~~) and to produce reliable and accurate results.~~).

4.4 Competency Component

4.4.1 General

The ~~competency component of the~~ laboratory's training program shall ~~demonstrate~~include knowledge-based and practical competency testing in the application of forensic autosomal and Y-STR data interpretation and comparison protocol(s) as used by the laboratory. The format of the test(s) and the criteria for passing the competency test shall meet ~~section~~Section 4.3 of ANSI/ASB Std 022, *Standard for Forensic DNA Analysis Training Programs*.

4.4.2 Knowledge-based Competency

The trainee shall successfully complete a knowledge-based test covering the critical information obtained during the training of forensic autosomal and Y-STR data interpretation and comparison protocol(s). ~~The format of the test(s) shall be at the discretion of the DNA technical leader or comparable authority.~~ The test(s) shall cover, at a minimum~~, the topics outlined under 2.~~:

a) the theoretical and scientific basis of forensic autosomal and Y-STR data interpretation and comparison;

b) the function of the reagent controls used in forensic autosomal and Y-STR data interpretation and comparison;

c) the quality controls steps pertaining to forensic autosomal and Y-STR data interpretation and comparison;

d) the laboratory's procedures pertaining to forensic autosomal and Y-STR data interpretation and comparison.

4.4.3 Practical Competency

The trainee shall successfully complete a practical competency test covering each of the forensic autosomal and Y-STR data interpretation and comparison protocol(s) for which he or she will be independently authorized~~. All types~~ to perform. Samples representative of ~~samples~~the range, type, and complexity for which the trainee will be authorized to interpret shall be included in the practical competency test. The trainee, at a minimum, shall be able to satisfactorily perform the following:

a) properly and accurately execute forensic autosomal and Y-STR data interpretation and comparison protocol(s);

b) apply the laboratory's interpretation and comparison procedures to a variety of evidentiary casework -or database-type samples, as applicable to the trainee;

c) operate relevant equipment, instrumentation, and software used in the laboratory for forensic autosomal and Y-STR data interpretation and comparison protocol(s);

d) correctly document work performed in accordance with laboratory procedures.

5 Conformance

In order to demonstrate conformance with this standard, the laboratory shall meet Section 5 of the ANSI/ASB ~~Std 022.~~Standard 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 literature 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.

~~technical leader in defining the breadth and scope of the materials to be reviewed by the trainee.~~

1] ~~This list is not meant to be all-inclusive.~~ ANSI/ASB Standard 022, *Standard for Forensic DNA Analysis Training Programs,* First Edition, 2019.[c]

2] ANSI/ASB Standard 040, *Standard for Forensic DNA Interpretation and Comparison Protocols,* First Edition, 2019.[b]

~~2~~3] Buckleton, J., et al. "Towards Understanding the effect of uncertainty in the number of contributors to DNA stains." *Forensic Science International: Genetics,* vol.1, 2007, pp. 20~~-~~–28.

~~3~~4] Buckleton, J., M. Krawczak, ~~M.,~~ and B. Weir~~, B~~. "The interpretation of lineage markers in forensic DNA testing." *Forensic Science International: Genetics*, vol. 5, 2011, pp. 78~~-~~–83.

~~4~~5] Budowle, B., et al. "Mixture Interpretation: Defining the relevant features for guidelines for the assessment of mixed DNA profiles in forensic casework." *Journal of Forensic Sciences*, vol. 54(4), 2009, pp. 810~~-~~–821.

~~5~~6] Budowle, B., J. Ge, ~~J.,~~X.G. Aranda, ~~X.G.,~~J.V. Planz, A.J.~~V.,~~ Eisenberg, ~~A.J.,~~and R. Chakraborty~~, R~~. "Texas Population Substructure and Its Impact on Estimating the Rarity of Y-STR Haplotypes from DNA Evidence." *Journal of Forensic Sciences*, vol. 54, 2009, pp. 1016~~-~~–1021.

~~6~~7] Butler, ~~John~~J. *Advanced Topics in Forensic DNA Typing: Interpretation,* Elsevier Academic Press, 2015.

~~7~~8] Dror, et al. "Subjectivity and bias in forensic DNA mixture interpretation." *Science and Justice*, vol. 51(4), 2011, pp. 204~~-~~–208.

~~8~~9] Haned, H., et al. "Estimating Number of Contributors to Forensic DNA Mixtures." *Journal of Forensic Sciences*, vol. 56 (1), 2011, pp. 23~~-~~–28.

~~9~~10] Ge, J., B. Budowle, ~~B.,~~J.V. Planz, A.J.~~V.,~~ Eisenberg, ~~A.~~J~~.~~. Ballantyne, ~~J.,~~and R. Chakraborty~~, R~~. "US forensic Y-chromosome short tandem repeats database." *Legal Medicine*, vol. 12, 2010a, pp. 289~~-~~–295.

~~10~~11] Gill, P., et al. "Commentary on Budowle, B., Onorato, A.J., Callaghan, T.F., Della Manna, A., Gross, A.M., Guerrieri, R.A., Luttman, J.C., McClure, D.L. Mixture Interpretation: Defining the

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

relevant features for guidelines for the assessment of mixed DNA profiles in forensic casework." *Journal of Forensic Sciences*, vol. 55(1), 2010, pp. 265~~-~~–268.

~~11]~~12] Gill, P., et al. "An Investigation of the rigor of interpretation rules for STR derived from less than 100 pg of DNA." *Forensic Science International,* vol. 112, 2000, pp. 17~~-~~–40.

~~12]~~13] Gusmão, L., J.M. Butler, ~~J.M.,~~Á. Carracedo, ~~Á.,~~P. Gill, ~~P.,~~M. Kayser, ~~M.,~~W.R. Mayr, ~~W.R.,~~N. Morling, ~~N.,~~M. Prinz, ~~M.,~~L. Roewer, ~~L.,~~C. Tyler-Smith, ~~C.,~~ et al. "DNA Commission of the International Society of Forensic Genetics (ISFG): An update of the recommendations on the use of Y-STRs in forensic analysis." *Forensic Science International*, vol. 157, 2006, pp. 187–197.

~~13]~~14] National Research Council. *The Evaluation of Forensic DNA Evidence*, National Research Council, 1996, Chapters 1~~-~~–6.

~~14]~~15] Perez, et al. "Estimating the number of contributors to two, three and four person mixtures containing high template and low template amounts." *Croatian Medical Journal*, vol. 52(3), 2011, pp. 314~~-~~–326.

~~15]~~16] SWGDAM. ~~SWIGDAM~~SWGDAM Interpretation Guidelines for Autosomal and Y-STR Typing for Forensic DNA Testing Laboratories[d].

[d] Available from: http://media.wix.com/ugd/4344b0_da25419ba2dd4363bc4e5e8fe7025882.pdf

DRAFT

Academy Standards Board
410 North 21st Street
Colorado Springs, CO 80904

www.aafs.org/academy-standards-board