

DNAmix 2021 — Inter-Laboratory Variation in Interpretation of DNA Mixtures Study

Interpretation, Comparison, and Statistical Analysis (ICSA) Subtest Instructions

Contents

1	Overview	1
2	ICSA Settings Selection.....	2
3	Comparison Packets: DNA Mixture Profiles and Reference Profiles.....	3
	3.1 Amp/CE Settings.....	4
	3.2 Preparation of DNA Mixture Profiles.....	4
	3.3 Data Provided.....	5
4	Reporting Statistical Analysis Results.....	6
5	ICSA Subtest Questions	7

1 Overview

The *Interpretation, Configuration, and Statistical Analysis (ICSA) Subtest* is the fourth and final phase of the DNAmix 2021 study. In this subtest you will be assigned 8 *Comparison Packets* (each containing one DNA mixture profile and one or more reference profiles). All DNA profiles (mixture and reference) will be provided as electropherograms (HID file format). Each electropherogram you are assigned will be prepared using the specified combination of *Amp/CE Settings* you selected in *Mixture Configuration Selection*.

For each *Comparison Packet*, participants will be asked to provide categorical comparison conclusions and statistical analysis responses for two scenarios:

- **General case** — The categorical conclusion and supporting statistics that would be provided if there is no reason to assume that a close relative of the person of interest may be a suspect. Participants will also be asked to optionally provide their supporting statistics computed using only the 20 CODIS core loci for the general case. For participants who report likelihood ratios, this is expressed as $LR=(H_p = \text{the person of interest is a contributor to the mixture} / H_d = \text{an *unrelated individual* is a contributor to the mixture})$.
- **Sibling case** — (Optional) The categorical conclusion and supporting statistics that would be provided if a sibling (but not an identical twin) of the person of interest is also a suspect, but for whom a DNA profile is not available. For participants who report likelihood ratios, this is expressed as $LR=(H_p = \text{the person of interest is a contributor to the mixture} / H_d = \text{a *sibling of the POI* is a contributor to the mixture and the POI is NOT a contributor to the mixture})$. Participants will only report sibling case categorical conclusions and statistics if their SOPs allow such in casework.

For each *Comparison Packet*, participants will also be asked to provide assessments of suitability and number of contributors (as in the *NoC Subtest*).

The *ICSA Subtest* will be accessible via a link on the Participant Homepage of the [DNAmix 2021 website](#). When the *ICSA Subtest* initially becomes available, all registered participants will be notified via e-mail. Prior to the launch of the *ICSA Subtest*, an “*ICSA Beta Test*” will be temporarily available, which will consist of one comparison packet for review. Although the *ICSA Beta Test* is not required, participants are highly encouraged to complete it as practice and are welcomed to provide feedback for improving the *ICSA Subtest* (e.g., clarity of questions, functionality of software, etc.). Once the *ICSA Subtest* becomes available, the *ICSA Beta Test* will be removed from the website.

Conduct your interpretations, comparisons, and statistical evaluations of each DNA mixture profile and respond to each of these questions based upon the policies and validated procedures in your Standard Operating Procedures (SOPs), using the same considerations and diligence that you would employ for

operational casework samples. Your responses should go through technical review/quality assurance as specified by your laboratory's SOPs.

Please review the "[DNAmix2021 — Glossary](#)" prior to beginning the *ICSA Subtest* for details about the acronyms and terminology as specifically used in this study.

Names of commercial manufacturers are included for the systems that are frequently used in laboratories; inclusion does not imply endorsement by the study team.

2 ICSA Settings Selection

In the *ICSA Subtest*, you will report your statistical responses for the population databases you use operationally, for each of the following populations:

- Black/African American
- White/Caucasian
- Combined Asian (or East Asian or Chinese)
- Combined Hispanic (or SW Hispanic or Mexican-American)
- Overall Combined

For example, if in your casework you use a Black/African American population database and a White/Caucasian population dataset (but do not use any of the others), you will report statistical responses using only those two tables.

Before you start the *ICSA Subtest*, you must indicate whether you use each type of population database and which specific databases you use in *ICSA Settings Selection*, which is a brief online questionnaire accessed from the [DNAmix 2021 website](#).

ICSA Settings Selection includes the following questions:

1. Please indicate which Black/African American population database you use in your casework, and will use in this study (also described as "Combined African American, Bahamian, and Jamaican" or "Black African/Caribbean"):
 - 1.a FBI STR Population Data: African American Allele Frequencies (includes African American, Bahamian, and Jamaican) (Expanded 2015 STR Allele Frequency Table)*
 - 1.b NIST 1036 Revised AfAm (2017)[†]
 - 1.c Other (Please specify: _____)
 - 1.d We do not use a Black/African American population database
- 1.1 [if you use a Black/African American population database] Please specify the value of theta that you use for the Black/African American population database that you use in casework, and will use in this study (Note: negative values and values greater than 0.2 will be regarded as typographical errors): _____
2. Please indicate which White/Caucasian population database you use in your casework, and will use in this study:
 - 2.a FBI STR Population Data: Caucasian Allele Frequencies (Expanded 2015 STR Allele Frequency Table)
 - 2.b NIST 1036 Revised Cauc (2017)
 - 2.c Other (Please specify: _____)
 - 2.d We do not use a White/Caucasian population database

* Current (2015) FBI population databases can be found at [https://www.fsigenetics.com/article/S1872-4973\(16\)30142-9/fulltext](https://www.fsigenetics.com/article/S1872-4973(16)30142-9/fulltext) (in the supplemental table <https://www.fsigenetics.com/cms/10.1016/j.fsigen.2016.07.022/attachment/bdb74544-07b8-4a23-ad28-46b3362de740/mmc1.xlsx>).

[†] Current (2017) NIST population databases can be found at <https://strbase.nist.gov/NISTpop.htm>.

- 2.1 *[if you use a White/Caucasian population database]* Please specify the value of theta that you use for the White/Caucasian population database that you use in casework, and will use in this study (Note: negative values and values greater than 0.2 will be regarded as typographical errors): _____
3. Please indicate which Combined Asian population database you use in your casework, and will use in this study. If you do not use a Combined Asian population database in casework, please indicate the East Asian or Chinese population database you use in your casework, and will use in this study (if applicable):
- 3.a *NIST 1036 Revised Asian (2017)*
 - 3.b *Other Combined Asian population database (Please specify: _____)*
 - 3.c *Other East Asian or Chinese population database (Please specify: _____)*
 - 3.d *We do not use a Combined Asian, East Asian, or Chinese population database*
- 3.1 *[if you use a Combined Asian/ East Asian/Chinese population database]* Please specify the value of theta that you use for the Combined Asian, East Asian, or Chinese population database that you use in casework, and will use in this study (Note: negative values and values greater than 0.2 will be regarded as typographical errors): _____
4. Please indicate which Combined Hispanic population database you use in your casework, and will use in this study. If you do not use a Combined Hispanic population database in casework, please indicate the Southwestern Hispanic or Mexican-American population database you use in your casework, and will use in this study (if applicable):
- 4.a *FBI STR Population Data: Southwest Hispanic Allele Frequencies (Expanded 2015 STR Allele Frequency Table)*
 - 4.b *NIST 1036 Revised Hispanic (2017)*
 - 4.c *Other Combined Hispanic population database (Please specify: _____)*
 - 4.d *Other SW Hispanic or Mexican-American population database (Please specify: _____)*
 - 4.e *We do not use a Combined Hispanic, SW Hispanic, or Mexican-American population database*
- 4.1 *[if you use a Combined Hispanic/SW Hispanic/Mexican-American population database]* Please specify the value of theta that you use for the Combined Hispanic, SW Hispanic, or Mexican-American population database that you use in casework, and will use in this study (Note: negative values and values greater than 0.2 will be regarded as typographical errors): _____
5. Please indicate which Overall Combined population database you use in your casework, and will use in this study:
- 5.a *NIST 1036 Revised All (2017)*
 - 5.b *Other (Please specify: _____)*
 - 5.c *We do not use an Overall Combined population database*
- 5.1 *[if you use an Overall Combined population database]* Please specify the value of theta that you use for the Overall Combined population database that you use in casework, and will use in this study (Note: negative values and values greater than 0.2 will be regarded as typographical errors): _____

3 Comparison Packets: DNA Mixture Profiles and Reference Profiles

In the *ICSA Subtest* you will be assigned a total of 8 *Comparison Packets*. Each *Comparison Packet* includes one DNA mixture profile, one or more reference profiles, positive and negative controls, and allelic ladders. No case information will be provided. All DNA mixture profiles and reference profiles are electropherograms, provided to participants as HID files.

Each *Comparison Packet* includes one or more reference profiles:

- All comparison packets include 1 reference profile designated as “person of interest” (POI), which for the purposes of this study indicates an individual whose contribution to the mixture is in question (such as an alleged perpetrator).

- All comparison packets that are simulated sexual assault kits (SAKs) include 1 reference profile labeled “victim,” which for the purposes of this study indicates the complainant in a sexual assault from whom simulated sexual assault kit samples are collected.
- Some comparison packets that are simulated SAKs may include 1 reference profile labeled “consensual partner,” which for the purposes of this study indicates an individual known to have had consensual intimate contact with a victim of a sexual assault.
- Some non-SAK comparison packets may include 1 reference profile labeled “expected contributor,” which for the purposes of this study indicates a known individual who is expected or assumed to be a contributor to a DNA mixture profile, such as the owner of an item or a member of a household.

You will have access to only one *Comparison Packet* at a time: to avoid the possibility of administrative errors or misunderstandings, you must submit your responses for a DNA mixture profile before downloading the next DNA mixture profile.

3.1 Amp/CE Settings

In order to represent the SOPs of as many participating laboratories as feasible, electropherograms were prepared using four combinations of “Amp/CE Settings” (which refers to a specific combination of amplification kit, amplification cycles, volume of amplification reaction, CE instrument, and injection time and voltage). The combinations of Amp/CE Settings that have been implemented were the four most commonly-used Amp/CE settings selected by registered participants,* and use the following abbreviations:

- “6C29” — Promega PowerPlex Fusion 6C, 29 amplification cycles, 25µL amplification volume, ABI 3500xl, 1.2kV, 24 seconds (equivalent to ABI 3500: 1.2kV, 15 seconds†)
- “GF28” — Applied Biosystems GlobalFiler, 28 amplification cycles, 25µL amplification volume, ABI 3500xl, 1.2kV, 24 seconds (equivalent to ABI 3500: 1.2kV, 15 seconds)
- “GF29” — Applied Biosystems GlobalFiler, 29 amplification cycles, 25µL amplification volume, ABI 3500xl, 1.2kV, 24 seconds (equivalent to ABI 3500: 1.2kV, 15 seconds)
- “ID28” — Applied Biosystems AmpFLSTR Identifier Plus, 28 amplification cycles, 15µL amplification volume, ABI 3500xl, 1.2kV, 12 seconds (equivalent to ABI 3500: 1.2kV, 7.5 seconds)

You will be assigned mixtures that were prepared using the Amp/CE setting option that you chose during *Mixture Configuration Selection*, prior to the *NoC Subtest*.

3.2 Preparation of DNA Mixture Profiles

The DNA used to create the mixture profiles for this study came from various sources, including buccal, blood, and tissue samples. There were no simulated/contrived profiles; all DNA profiles in this study are from real people. DNA samples were extracted prior to mixing.

Mixtures were quantified using ABI Quantifiler Trio on an ABI 7500 real-time PCR instrument. The mixture quantification results (including the total amount of DNA amplified, amount of male DNA, and degradation index) will be included with the mixtures in the *ICSA Subtest*.

Various volumes of DNA were pipetted into a single tube to make a large mixture stock. That stock was then aliquoted and amplified in each of the four amp kits (see “Amp/CE settings” above for amplification volumes and cycles). The ABI 9700 thermocycler was used for amplification, using the specific Amp/CE settings and other standard manufacturer recommended settings. The ABI 3500xl was used for capillary electrophoresis

* Registered participants were contacted by email and were given a deadline of 23 August 2021 to indicate preferences for Amp/CE settings.

† Based upon discussions with ThermoFisher, we are proceeding with the assumption that injection for X seconds on an ABI 3500@1.2kV can be considered equivalent to 1.6X seconds on an ABI 3500xl@1.2kV.

(CE), using injection time and voltage settings specified in the Amp/CE Settings (see above); settings for run time, run voltage, capillary length, polymer type, etc. use the default settings specified for each amp kit.

GeneMapper (v1.5; incorporated into ABI 3500xl) was used to create HID files. We are not providing PDFs (images) of the electropherograms because creating such PDFs implements decisions regarding the analytical threshold (AT) value and the utilization of stutter filters, and we want all such decisions to be made by the participants.

Every effort was made with respect to quality assurance in creating these mixtures. Note that in some cases there may be artifacts (such as pull-up) present, as may be found in ordinary casework — please review the controls provided.

3.3 Data Provided

In the ICSA Subtest you will be assigned a total of 8 *Comparison Packets*. You will have access to only one *Comparison Packet* at a time: to avoid the possibility of administrative errors or misunderstandings, you must submit your responses for a DNA mixture profile before downloading the next DNA mixture profile.

Each *Comparison Packet* is numbered (ICSA_001 through ICSA_999, shown as “ICSA_XXX” in the table below). Participants are not necessarily assigned the same packets, and the order of assignments varies among participants.

Each *Comparison Packet* is specific to the Amp/CE Settings previously selected by participants (shown as “YYYY” in the table below), using the abbreviations listed in *Section 3.1 (Amp/CE Settings)* [6C29, GF28, GF29, or ID28].

Each *Comparison Packet* includes 1 DNA mixture profile and 1 person of interest (POI) reference profile. *Comparison Packets* that are simulated sexual assault kits (SAKs) include 1 victim (VIC) reference profile, and may include 1 consensual partner (CON) reference profile. Non-SAK comparison packets may include 1 expected contributor (EXP) reference profile. Each mixture or reference profile includes positive and negative controls, and 2 allelic ladders.

Each *Comparison Packet* is contained in a Zip file, downloaded from the DNAmix 2021 website (<https://dnamix.edgeaws.noblis.org/>). The mixture and reference profiles are included in separate subdirectories as shown in the table below.

	Subdirectory	Files	
All packets	ICSA_XXX_YYYY/Mixture/	ICSA_XXX_YYYY_Mixture.HID	DNA mixture profile
		ICSA_XXX_YYYY_Ladder1-Mixture.HID ICSA_XXX_YYYY_Ladder2-Mixture.HID ICSA_XXX_YYYY_NEG-Mixture.HID ICSA_XXX_YYYY_POS-Mixture.HID	Controls for DNA mixture profile
		ICSA_XXX_YYYY_POI.HID	Person of interest (POI) reference profile
		ICSA_XXX_YYYY_Ladder1-POI.HID ICSA_XXX_YYYY_Ladder2-POI.HID ICSA_XXX_YYYY_NEG-POI.HID ICSA_XXX_YYYY_POS-POI.HID	Controls for POI reference profile
Some packets	ICSA_XXX_YYYY/VIC/	ICSA_XXX_YYYY_VIC.HID	Victim (VIC) reference profile
		ICSA_XXX_YYYY_Ladder1-VIC.HID ICSA_XXX_YYYY_Ladder2-VIC.HID ICSA_XXX_YYYY_NEG-VIC.HID ICSA_XXX_YYYY_POS-VIC.HID	Controls for VIC reference profile
		ICSA_XXX_YYYY_CON.HID	Consensual partner (CON) reference profile
		ICSA_XXX_YYYY_Ladder1-CON.HID ICSA_XXX_YYYY_Ladder2-CON.HID ICSA_XXX_YYYY_NEG-CON.HID ICSA_XXX_YYYY_POS-CON.HID	Controls for CON reference profile
	ICSA_XXX_YYYY/EXP/	ICSA_XXX_YYYY_EXP.HID	Expected contributor (EXP) reference profile
		ICSA_XXX_YYYY_Ladder1-EXP.HID ICSA_XXX_YYYY_Ladder2-EXP.HID ICSA_XXX_YYYY_NEG-EXP.HID ICSA_XXX_YYYY_POS-EXP.HID	Controls for EXP reference profile

In some *Comparison Packets*, the positive or negative controls were re-injected, in which case they are in a subdirectory (named POS or NEG) with the associated ladders.

4 Reporting Statistical Analysis Results

To ensure there is no ambiguity in the reported results, all statistical values shall be reported using scientific notation, using the format X.XXE+YY or X.XXE-YY.

- X.XX represents the statistical value, using three significant digits
- E represents an exponential constant of 10
- +/- represent the direction of the resulting exponent (i.e., positive for statistical values ≥ 1 and negative for statistical values < 1)
- YY represents the value of the resulting exponent, using two significant digits (e.g., 06 for an LR of 1,000,000)

Specific statistical values shall be reported as follows:

- For likelihood ratio (LR), report the result of the ratio (i.e., compute H_p/H_d and report the resulting value). For example, an LR of 1 million shall be reported as 1.00E+06. An LR of 1 shall be reported as 1.00E+00.
- For random match probability (RMP) and modified RMP (mRMP), report X (the “denominator”) from the 1 in X result format. In other words, an RMP of 1 in 1 million shall be reported as 1.00E+06 (i.e., using a positive exponent).
- For combined probability of inclusion (CPI) and combined probability of exclusion (CPE), report X (the “denominator”) from the 1 in X result format. In other words, a CPI of 1 in 1 million shall be reported as 1.00E+06 (i.e., using a positive exponent). Do not report the raw probability: a 0.000001% probability of inclusion should be transformed to 1 in 1 million and reported as 1.00E+06.

5 ICSA Subtest Questions

On the [DNAmix 2021 website](#), you will be asked to answer the following questions for **each** of the 8 comparison packets that you are assigned in the *ICSA Subtest*. The *ICSA Subtest* is completed online; this information is provided here as a reference.

As a quality assurance measure, the website will display an image of the electropherogram for the first several loci in the DNA mixture profile for the assigned *Comparison Packet*. Please ensure that you are submitting your responses for the given mixture profile.

After downloading the packet, we recommend clicking HOME in the main menu bar (at the top of the DNAmix webpage) and returning to the ICSA Subtest screen only when you are ready to enter your responses. (Some users have timeout issues if this window is left open.)

For each comparison packet assessed in the ICSA Subtest, you will be asked to review and confirm your responses prior to submission. After submission, your responses are considered final and cannot be changed.

Packet Assignment Details

1. Please re-enter the Participant ID shown at the top of the page (Dxxxx): _____

Note: this information will be used for quality assurance purposes only. The Participant ID (a 5 character alpha-numeric string starting with D) is located at the top right of the Interpretation, Comparison, and Statistical Analysis (ICSA) Page (right about the electropherogram preview image).

2. Please double-check the Comparison Packet number: verify that the number of the HID mixture file and any reference files you are assessing is the same as shown at the top of this page. Please enter that Comparison Packet number here (for example, in the ICSA Beta Test, you would enter 999): _____

The Comparison Packet number is located in the filename, in the electropherogram preview image (located at the top of the ICSA Subtest page of the DNAmix 2021 website), and embedded within the electropherogram data. You do not need to enter the "ICSA_" portion; please only enter the three digit Comparison Packet number.

- 2.1 Please indicate which (if any) reference profiles other than the POI are included in this *Comparison Packet* (check all that apply; leave blank if none):

Comparison packets that include a victim reference profile are simulated SAK packets; these may also include a consensual partner reference profile.

Non-SAK packets may include an expected contributor reference profile.

- 2.1-a Victim (VIC)
- 2.1-b Consensual partner (CON)
- 2.1-c Expected contributor (EXP)

Suitability and Number of Contributors

Questions 3-13 from the *Number of Contributors (NoC) Subtest* will be repeated exactly here, with the addition of Question 9.2 below. Please refer to the *NoC Subtest Instructions* for details about how to respond to each question.

The *ICSA Subtest* includes the following additional question, which is not in the *NOC Subtest*:

- 9.2 *[if indicated a range or minimum NoC]* (Given that you would report a range or minimum number of contributors) What NoC value did you use as a basis for your conclusions/analyses reported for this Comparison Packet regarding the person of interest (POI) as a potential contributor to the mixture sample?

In other words, when conducting your comparisons and statistical analyses for this Comparison Packet what value did you assume for number of contributors to the mixture sample?

Please report all statistical analyses in Q18, 19, and 22 below (if applicable) based upon your response to this question.

9.2-a 1 contributor

9.2-b 2 contributors

9.2-c 3 contributors

9.2-d 4 contributors

9.2-e 5 contributors

9.2-f 6 contributors

9.2-g 7 contributors

9.2-h 8 contributors

9.2-i 1 based analyses with respect to the POI on a range of number of contributors (not a single NoC value)

Note: You will only move on to the Comparison and Statistical Analysis questions below if you indicate that the DNA mixture sample is suitable for comparison and/or statistical analysis.

Comparison Conclusion — General Case

14. What is your conclusion regarding the person of interest (POI) as a potential contributor to the mixture sample?

In other words, did you determine that the POI can be included or excluded as a potential contributor to this DNA mixture profile? Alternatively, were you unable to include or exclude the POI as a potential contributor, thus yielding an inconclusive determination? If your laboratory SOPs do not permit you to report categorical conclusions, please indicate as such.

14.a *Included— the POI is considered a possible contributor (also known as consistent with, support for inclusion, cannot be excluded/eliminated)*

14.b *Inconclusive— the POI can neither be included nor excluded as a potential contributor (also known as uninformative)*

14.c *Excluded— the POI is NOT a possible contributor (also known as eliminated, support for exclusion)*

14.d *We do not use categorical conclusions*

15. Was your conclusion regarding the POI as a potential contributor to the mixture based upon manual comparison, statistical analysis, or both?

In other words, how did you conduct your interpretation, comparison, and/or statistical analysis of THIS mixture sample with respect to the POI? If you did not compute any statistics and reported your conclusion based solely upon a review of the electropherograms (e.g., comparison of alleles, evaluation of peak heights, consideration of stutter or other artifacts, etc.), indicate manual comparison only. If you did not review/compare the electropherograms manually prior to computing a statistic (either manually or using a software tool), indicate statistical analysis only. If you manually reviewed/compared the electropherograms and computed a statistical value, indicate both.

15.a *Manual comparison only* [Go to Additional Comments]

15.b *Statistical analysis only*

15.c *Both manual comparison and statistical analysis*

Statistical Analysis — General Case

16. What type of statistical values are you reporting for this comparison packet?

In other words, please indicate which statistic you computed with respect to the POI in THIS comparison packet. You are permitted to use different statistics on different comparison packets; however, the same type of statistic must be used within a comparison packet.

- 16.a Continuous likelihood ratio (LR)
- 16.b Semi-continuous LR
- 16.c Binary LR
- 16.d Combined probability of inclusion (CPI)
- 16.e Combined probability of exclusion (CPE)
- 16.f Random match probability (RMP)
- 16.g Modified RMP (mRMP)

17. What software was used to calculate the statistical values you are reporting for this comparison packet?

In other words, please indicate which software you used to compute your statistics with respect to the POI in THIS comparison packet. You are permitted to use different software (or to use manual computation/in-house workbooks) on different comparison packets; however, the same software tool must be used to compute all statistics within a comparison packet.

- 17.a I did my calculations manually, or used an in-house software or in-house workbook
- 17.b Armed Xpert
- 17.c CEESIt
- 17.d DNA View Mixture Solution
- 17.e EuroForMix
- 17.f LabRetriever
- 17.g likeLTD
- 17.h LRmix/LRmix Studio
- 17.i PopStats
- 17.j Soft Genetics MaSTR
- 17.k STRmix
- 17.l TrueAllele

18. Statistical responses (General case). Please enter the result of your statistical analysis regarding the POI as a contributor to the mixture, for each of the following population databases.

Report your statistics using the type of statistics you indicated in Question 16, computed using the population database and theta you indicated in ICSA Settings Selection. If you do not use a given type of population database the corresponding question will be disabled.

*Report statistical values in scientific notation, using the format X.XXE+YY or X.XXE-YY.**

For likelihood ratios, use the following generalized propositions, assuming unrelated contributors: H_p = the POI is a contributor to the mixture; H_d = the POI is NOT a contributor to the mixture.

For LR values, report the point estimate of the computed LR (i.e., not an interval/range, a unified statistic, varNOC, stratified statistic, etc.)

- 18.1 *[if Black/African American DB]* Statistical value regarding the POI as a contributor to the mixture (Black/African American population database): _____
- 18.2 *[if White/Caucasian DB]* Statistical value regarding the POI as a contributor to the mixture (White/Caucasian population database): _____
- 18.3 *[if Combined Asian/ East Asian/Chinese DB]* Statistical value regarding the POI as a contributor to the mixture (Combined Asian/East Asian/Chinese population database): _____
- 18.4 *[if Combined Hispanic/SW Hispanic/Mexican-American DB]* Statistical value regarding the POI as a contributor to the mixture (Combined Hispanic/SW Hispanic/Mexican-American population database): _____
- 18.5 *[if Overall Combined DB]* Statistical value regarding the POI as a contributor to the mixture (Overall Combined population database): _____

* Scientific notation format is detailed in Section 4.

19. *[if Amp/CE Setting Selection is 6C29, GF28, or GF29]* Statistical responses (General case—CODIS core loci*). Please enter the result of your statistical analysis regarding the POI as a contributor to the mixture, for each of the following population databases, computed using only the 20 CODIS core loci. (Note: this question is optional)

Report your statistics using the type of statistics you indicated in Question 16, computed using the population database and theta you indicated in ICSA Settings Selection; use ONLY the 20 CODIS core loci in your computations. If you do not use a given type of population database the corresponding question will be disabled.

Report statistical values in scientific notation, using the format X.XXE+YY or X.XXE-YY.

For likelihood ratios, use the following generalized propositions, assuming unrelated contributors: H_p = the POI is a contributor to the mixture; H_d = the POI is NOT a contributor to the mixture.

For LR values, report the point estimate of the computed LR (i.e., not an interval/range, a unified statistic, varNOC, stratified statistic, etc.)

- 19.1 *[if Black/African American DB]* Statistical value regarding the POI as a contributor to the mixture based upon the 20 CODIS core loci (Black/African American population database): _____
- 19.2 *[if White/Caucasian DB]* Statistical value regarding the POI as a contributor to the mixture based upon the 20 CODIS core loci (White/Caucasian population database): _____
- 19.3 *[if Combined Asian/ East Asian/Chinese DB]* Statistical value regarding the POI as a contributor to the mixture based upon the 20 CODIS core loci (Combined Asian/East Asian/Chinese population database): _____
- 19.4 *[if Combined Hispanic/SW Hispanic/Mexican-American DB]* Statistical value regarding the POI as a contributor to the mixture based upon the 20 CODIS core loci (Combined Hispanic/SW Hispanic/Mexican-American population database): _____
- 19.5 *[if Overall Combined DB]* Statistical value regarding the POI as a contributor to the mixture based upon the 20 CODIS core loci (Overall Combined population database): _____

Comparison Conclusion — Sibling Case

20. In your casework, would you report a conclusion and/or statistics regarding the POI if a sibling (not an identical twin) of the POI is also a suspect, but for whom no DNA profile is available?

In other words, if a POI claims his sibling was the culprit but that sibling cannot be located for a sample, would you report a conclusion and/or statistical value regarding your interpretation, comparison, and statistical analysis of the mixture profile with respect to the POI?

- 20.a No [Go to Q23, Conditioning]
20.b Yes

21. *[if yes]* What conclusion would you report regarding the POI if a sibling (not an identical twin) of the POI is also a suspect, but for whom no DNA profile is available?

In other words, did you determine that the POI can be included or excluded as a potential contributor to this DNA mixture profile (when considering that a sibling is also a suspect)? Alternatively, were you unable to include or exclude the POI as a potential contributor, thus yielding an inconclusive determination? If your laboratory SOPs do not permit you to report categorical conclusions, please indicate as such.

- 21.a *Included— the POI is considered a possible contributor (also known as consistent with, support for inclusion, cannot be excluded/eliminated)*

* A list of the 20 CODIS core loci can be found at: <https://www.fbi.gov/services/laboratory/biometric-analysis/codis/codis-and-ndis-fact-sheet>

- 21.b *Inconclusive— the POI can neither be included nor excluded as a potential contributor (also known as uninformative)*
- 21.c *Excluded— the POI is NOT a possible contributor (also known as eliminated, support for exclusion)*
- 21.d *We do not use categorical conclusions*

Statistical Analysis — Sibling Case

22. Statistical responses (Sibling case). Please enter the result of your statistical analysis regarding the POI as a contributor to the mixture if a sibling (not an identical twin) of the POI is also a suspect (but for whom no DNA profile is available), for each of the following population databases.

Report your statistics using the type of statistics you indicated in Question 16, computed using the population database and theta you indicated in ICSA Settings Selection. If you do not use a given type of population database the corresponding question will be disabled.

*Report statistical values in scientific notation, using the format X.XXE+YY or X.XXE-YY.**

For likelihood ratios, use the following generalized propositions, assuming unrelated contributors: H_p = the POI is a contributor to the mixture; H_d = the sibling of the POI is a contributor to the mixture and the POI is NOT a contributor to the mixture.

For LR values, report the point estimate of the computed LR (i.e., not an interval/range, a unified statistic, varNOC, stratified statistic, etc.)

- 22.1 *[if Black/African American DB]* Statistical value regarding the POI as a contributor to the mixture given a sibling is also a suspect (Black/African American population database): _____
- 22.2 *[if White/Caucasian DB]* Statistical value regarding the POI as a contributor to the mixture given a sibling is also a suspect (White/Caucasian population database): _____
- 22.3 *[if Combined Asian/ East Asian/Chinese DB]* Statistical value regarding the POI as a contributor to the mixture given a sibling is also a suspect (Combined Asian/East Asian/Chinese population database): _____
- 22.4 *[if Combined Hispanic/SW Hispanic/Mexican-American DB]* Statistical value regarding the POI as a contributor to the mixture given a sibling is also a suspect (Combined Hispanic/SW Hispanic/Mexican-American population database): _____
- 22.5 *[if Overall Combined DB]* Statistical value regarding the POI as a contributor to the mixture given a sibling is also a suspect (Overall Combined population database): _____

Conditioning

23. *[If selected 2.1-a, 2.1-b, or 2.1-c]* Did you assume the presence of the victim, consensual partner, and/or expected contributor reference profiles during deconvolution? (check all that apply; leave blank if none apply)

In other words, did you assume that the victim and/or consensual partner (for SAK comparison packets) or the expected contributor (for non-SAK comparison packets) were contributors to the DNA mixture at any point during your interpretation and comparison of the mixture profile? Such assumptions may have been considered/incorporated either manually or using software.

- 23.a *I assumed the presence of the Victim (VIC)*
- 23.b *I assumed the presence of the Consensual partner (CON)*
- 23.c *I assumed the presence of the Expected contributor (EXP)*

* Scientific notation format is detailed in Section 4.

24. *[If selected 2.1-a, 2.1-b, or 2.1-c]* Were your reported statistics conditioned on the victim and/or consensual partner OR on the expected contributor? (i.e., were these accounted for in the propositions, so that the statistics were calculated assuming their presence) (check all that apply; leave blank if none apply)

In other words, did you assume that the victim and/or consensual partner (for SAK comparison packets) or the expected contributor (for non-SAK comparison packets) were contributors to the DNA mixture during your statistical analysis of the mixture profile?

Specifically, did your statistical analyses omit specific alleles/genotypes from your statistical analysis with respect to the POI based upon the genotypes of the victim, consensual partner, or expected contributor? Such omissions may have been incorporated in your statistical analysis conducted via manual computation or a software tool.

- 24.a Statistics were conditioned on the presence of the Victim (VIC)
24.b Statistics were conditioned on the presence of the Consensual partner (CON)
24.c Statistics were conditioned on the presence of the Expected contributor (EXP)

Omitted Loci

25. Did you omit any loci from your statistical computations?

In other words, did you completely ignore any loci in your statistical computations, conducted either manually or using a software tool? These loci would be dropped from your computation entirely (i.e., you would be omitting ALL alleles at a given locus, not just a selection of alleles).

- 25.a Yes
25.b No [Go to Additional Comments]

26. *[If Yes to omitted loci]* Please check all loci that you OMITTED from your statistical computations. (check all that apply; select at least one)

Note: you will only see the set of loci included in the amplification kit that you previously selected in Mixture Configuration Selection.

Names of commercial manufacturers are included for the systems that are the most frequently used by registered participants; inclusion does not imply endorsement by the study team.

Applied Biosystems GlobalFiler (display order)	Promega PowerPlex Fusion 6C (display order)	Applied Biosystems AmpFLSTR Identifiler Plus (display order)
D3S1358	Amel	D8S1179
vWA	D3S1358	D21S11
D16S539	D1S1656	D7S820
CSF1PO	D2S441	CSF1PO
TPOX	D10S1248	D3S1358
Y indel	D13S317	TH01
Amel	Penta E	D13S317
D8S1179	D16S539	D16S539
D21S11	D18S51	D2S1338
D18S51	D2S1138	D19S433
DYS391	CSF1PO	vWA
D2S441	Penta D	TPOX
D19S433	TH01	D18S51
TH01	vWA	Amel
FGA	D21S11	D5S818
D22S1045	D7S820	FGA
D5S818	D5S818	
D13S317	TPOX	
D7S820	D8S1179	
SE33	D12S391	
D10S1248	D19S433	
D1S1656	SE33	
D12S391	D22S1045	
D2S1338	DYS391	
	FGA	
	DYS576	
	DYS570	

Additional Comments

27. Additional comments: Please provide a comment ONLY if there is an issue or a limitation for this comparison packet that you could not adequately address using any of your responses above. (Please limit your responses to 75 words or less.)