DNAmix 2021 Mixture Analysis Study

Frequently Asked Questions

Last updated 11 April 2022

See the end for the most recent additions

Note: All questions received over the course of the study will be answered here as well as in direct email responses, unless the questions are specific to an individual participant. This allows us to provide the same information to all participants. We may modify the wording of some questions we receive to remove lab-specific references, or to combine multiple similar questions.

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1. *Q: Who may participate?*

   A: Participation will be open to all forensic laboratories that conduct DNA mixture interpretation as part of their SOPs; non-U.S. laboratories are welcome to participate if they report interpretations in English. Participation in the study requires the participants to agree to use the same diligence in performing these analyses as used operationally in casework, and to use their laboratory's SOPs in performing these analyses.

2. *Q: Are you looking for only a single response per lab, or could multiple analysts within my lab participate?* (added 30 June)

   A: The responses must go through technical review/quality assurance as specified by the laboratory's SOPs. As long as that is satisfied, multiple responses from a given laboratory are acceptable. We refer to multiple participants from a lab as "subunits" (as discussed in the instructions).

3. *Q: For the NoC and ICSA subtests, will data be provided from only a single amplification kit or the amplification kit which is actually used by the participant laboratories?* (added 30 June)

   A: We will prepare HID files for several popular combinations of amplification kit, CE instrument, and amplification cycle (collectively "Amp/CE Settings"). Specifics will be determined by the results of the Configuration Questionnaire completed as part of registration.

4. *Q: Is a cell phone really necessary for registration? I do not have a work cell, and am hesitant to use my personal cell for this.* (added 30 June)

   A: Unfortunately, we do require a cell phone number for registration. Security approval for the website requires multi-factor authentication by texting a code to a cell phone — this level of security is required to protect both the participants' information, and the DNA profiles included in the study. The cell phone number will only be used for that purpose, will never leave the server, and is not accessible by the study staff. If this is a burden, let us know: we are looking into other alternatives, but at least for now this is our only option.

5. *Q: I had a question regarding the subunits. All of the qualified analysts in my section would like to participate in the study, so I was considering splitting up into groups of two, with one completing the analysis/interpretation and the other completing the technical review. Would it be reasonable to split up each phase (the NoC subtest and the ICSA subtest) between the two analysts? Could I allow half of the NoC subtest to be completed by analyst 1, have it tech reviewed by analyst 2, and then allow analyst 2 to complete the second half of the subtest and have analyst 1 tech review? I want to allow as much participation in the study as possible while still having everything technically reviewed, if possible.* (added 30 June)

   A: The simple answer: How you assign staff to perform the subtests is your laboratory's decision. More broadly, with context: From a human subjects research standpoint, the participants in this study are laboratories (or

subunits within laboratories) — not individuals. The study team does not need (or want) visibility into whether a participating subunit is an individual or team: we only ask that the labs do what they can to make this as close to casework as possible, including tech review, and that the responses from subunits within a lab be independent.

6. *Q: Will all 4 studies be opened to a subunit even if that individual misses a particular study? For example, if an individual signs up as a subunit but misses study 1, will subsequent studies then become unavailable to that individual?* (added 30 June)

   A: We encourage participants to complete all 4 phases of the study, and they will be made available in order. Since the 1st 2 phases (the P&P Questionnaire and Casework Scenario (CS) Questionnaire) are just questionnaires, we assume that participants will complete those prior to the last 2 phases (NoC Subtest and ICSA Subtest): our analyses of the NoC and ICSA responses will rely on the P&P and CS responses.

7. *Q: What is the study schedule? What is the timeframe for participating labs to return results?* (updated 11 April)

   A: Our current planned schedule is as follows:
   - *Registration will be open for new participants through 06 Mar 2022.*
   - *P&P results will be accepted through 10 Mar 2022.*
   - *Casework Scenarios results will be accepted through 13 Mar 2022.*
   - *NoC results will be accepted through 19 Apr 2022.*
   - *ICSA results will be accepted through 05 June 2022.*

8. *Q: During Registration, there is a spot on the Informed Consent Form to add the "Laboratory Name" and "Laboratory Representative's Name," to give consent to take part in the study. Should the name be one of the individuals in the subunit, or just one representative per laboratory?* (added 7 July)

   A: On the Consent Form, the Laboratory Representative is an authorized representative of the lab: it is the lab's discretion who that is, and could be the point of contact, a manager, lab director, or legal counsel. If multiple subunits from a lab are participating, it is the lab's discretion whether there is a single lab representative on the Consent Form or a different one for each subunit. The name of the lab representative from the Consent form does not have to be the point of contact from the Registration form (but it can be). (The point of contact does need to be different for each subunit.)

9. *Q: If a laboratory opts to have teams of multiple analysts complete the study, for ease & consistency, can one team fill out the study questionnaires for the whole laboratory/unit?* (added 7 July)

   A: We prefer to treat the subunits as independent participants as much as possible, and request that each subunit complete the questionnaires separately.

10. *Q: If the laboratory opts to have teams and one team was unable to hand in their results, would that void the rest of the participants participation from the study?* (added 7 July)

    A: No: we will treat the responses from the subunits independently. Note also that we use partial results from participants who do not complete the entire study (e.g. if a lab responds to the questionnaires and NoC subtest but does not finish the ICSA subtest)

11. *Q: When will the closing period for registration be for this study?* (added 7 July)

    A: We will keep registration open at least through mid-September (to accommodate potential participants who hear of the study at ISHI).

12. *Q: Could you give me an idea of the time it should take to participate in the project?* (added 29 July)

    A: We estimate the time needed for participants for each sample in the Number of Contributors (NoC) Subtest to vary significantly based on the complexity of the mixture, from 5 minutes to over an hour per sample. We estimate the time needed for participants for each sample in the Interpretation, Comparison, and Statistical Analysis (ICSA)

Subtest to vary significantly, up to 3 or 4 hours of human analyst time per sample, not including software processing time.

13. *Q: (Regarding the Policies and Procedures question "Do your SOPs limit interpretation and/or comparison based on a maximum total number of contributors?") My laboratory has different analysis paths depending upon sample/case conditions, each with different limitations on maximum total number of contributors. How should I record my response?* (added 05 August, modified 21 September)

    *A: We recognize that laboratories may have different interpretation/comparison/statistical analysis paths depending upon the case or sample conditions. Given this, please record the single highest possible **total** NoC that your laboratory has the ability to handle for DNA mixture samples.*

14. *Q: In the Casework Scenarios Questionnaire (Phase 2), you ask in question 2 which types of information we USUALLY have available during interpretation of DNA data. When you refer to "Sexual Assault Medical Exam Report," what specifically does this include?* (added 21 September)

    A: The Sexual Assault Medical Exam Report will include details regarding the source of specific samples (e.g., vaginal swab, rectal swab, etc.), the presence of bruises or injuries (e.g., descriptions, photographs, etc.), the medical condition of the victim, and/or whether the victim had consensual sex prior to the assault.

15. *Q: In the Casework Scenarios Questionnaire (Phase 2), you ask in question 2 which types of information we USUALLY have available during interpretation of DNA data. While my laboratory does not have formal statements from witnesses, complainants, or investigators, we do have some information provided to use regarding the case by means of a case history on our submission forms. Can you please clarify how you would like me to indicate as such in question 2.* (added 21 September)

    A: Your description of the case history provided on submission forms would be included in the "Law Enforcement Case File." Please check this box if you USUALLY have access to that case history information.

16. *Q: I was disappointed that the Qiagen Investigator 24plex kit was not included as one of the subset whilst two GlobalFiler settings were chosen instead. Why was this kit not included in the study?* (added 21 September)

    A: We wish we could replicate all the registered participants STR laboratory protocols, but are unfortunately unable to provide all possible Amp/CE settings due to funding and time constraints. In selecting the specific Amp/CE settings options that we would provide, we used the four most commonly-used combinations of settings (as reported by registered participants by 23-Aug). Overall, there was a clear majority of participants who used GlobalFiler / 29 cycles or Fusion 6C / 29cycles, but the remainder of participants had a wide variety of settings. Relatively few participants selected Qiagen Investigator 24plex and those who did had a wide variety of cycle settings, so there was no single set of 24plex Amp/CE settings that would have captured a statistically-useable number of potential participants.

17. *Q: Since we're a laboratory using 24plex with STRmix, since our model maker is not adjusted for any of the kits, will it still make sense for us to move forward (i.e. will model maker for one of the sets be included, SOPS, etc.)?* (added 21 September)

    A: For each Amp/CE setting combination, we will be providing the following data/information:
    - *1 DNA mixture profile (HID file)*
    - *0-3 DNA reference profiles (HID files)*
    - *The following textual information:*
        - *Amp/CE Settings used to create the electropherograms:*
            - *Amplification kit*
            - *Amplification cycles*
            - *Volume of amplification reaction*
            - *CE instrument*

        - *Injection time and voltage*
    - *Quantitation data (as measured by Quantifilier Trio during quantitation of the mixture):*
        - *Total amount of DNA amplified*
        - *Total amount of male DNA amplified*
        - *Degradation index*
- *The following additional HID files, which some participants may wish to use for quality assurance:*
    - *Amplification positive control (HID file)*
    - *Amplification negative control (HID file)*
    - *2 allelic ladders (2 HID files)*

We will not be providing the model maker or SOPs for each Amp/CE setting combination. It is up to the participating laboratory to use the resources available to them.

Given this, it is up to the discretion of the laboratory whether they choose to move forward to NoC and ICSA using an Amp/CE Settings option that differs from their SOPs. You will have an opportunity to indicate whether or not you choose to move forward in the forthcoming Mixture Configuration Selection. Should you choose to participate in NoC and/or ICSA using Amp/CE Settings that are not equivalent to your SOPs, you will have the opportunity to indicate as such.

Even if you are unable to participate in NoC and ICSA, we encourage laboratories to participate in the first two phases of the study (Policies & Procedures Questionnaire and Casework Scenarios Questionnaire). Please let us know if you have any further questions.

18. Q: I am attempting to submit my responses to the NoC Subtest, but when I click "Submit" after reviewing, I get this error message: "An error occurred while submitting responses." (added 22 November)

A: This error is a result of a browser, network, or timeout issue. To fix this issue, please logout of the DNAmix webpage and close your browser; then reopen a new browser window and log back into the DNAmix webpage. Once you have logged back in, you can re-renter and review your responses and you should be able to successfully submit.

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

19. Q: What changes were made after the ICSA Beta Test? (added 19 January)

A: Two changes were made since the ICSA Beta Test:

- *Responses entered into the website are now automatically saved: if you enter part of your responses for a sample and close the browser, the responses are retained the next time you log into the website.*
- *We added one question (#9.2) to address participants who would report the number of contributors (NoC) as a range (or minimum), but would use a single NoC value when calculating statistics with respect to a person of interest.*

20. Q: There are no Amp/CE choices for the 3130 genetic analyzer; therefore, we do not have comparable peak heights when referring to our analytical and stochastic thresholds. Do you have an alternative solution or should we consider ourselves unable to participate? (added 21 January)

A: We wish we could replicate all the registered participants STR laboratory protocols, but are unfortunately unable to provide all possible Amp/CE settings due to funding and time constraints. In selecting the CE instrument options that we could provide, we evaluated the responses to the Registration Questionnaire (as completed by registered participants by 09-Aug-2021), which indicated a supermajority of participants used the ABI 3500 or 3500xl genetic analyzer. Given this, we opted to move forward using only the ABI 3500 series instruments to prepare the samples in this study, in order to maximize the number of different Amp/CE settings we could accommodate. We will not be providing data for any other genetic analyzers.

Given this, it is up to the discretion of your laboratory whether you choose to move forward to NoC and ICSA using an Amp/CE Settings option that differs from their SOPs in terms of the CE instrument used. Should you choose to participate in NoC and/or ICSA using Amp/CE Settings that are not equivalent to your SOPs, you will have the opportunity to indicate as such.

Even if you are unable to participate in NoC and ICSA, we encourage laboratories to participate in the first two phases of the study (Policies & Procedures Questionnaire and Casework Scenarios Questionnaire).

21. *Q: Should we complete the NoC Subtest (Phase 3) if we are not able to complete the ICSA Subtest (Phase 4) due to time constraints because of our current laboratory initiatives/caseload?* (added 21 January)

    A: Even if you are unable to complete the ICSA Subtest due to time constraints, we encourage laboratories to complete the first three phases of the study (Policies and Procedures Questionnaire, Casework Scenarios Questionnaire, and NoC Subtest). We are also willing to accept any number of ICSA Comparison Packets that you are able to complete in the study period (even if you are unable to complete all 8 assignments).

22. *Q: If multiple individuals from the same lab participate, each individual is asked to register as a "subunit" from a single laboratory. Will each "subunit" receive different mixtures for the NoC and ICSA subtests, or will they be given the same mixtures because they are linked to a single laboratory/point of contact?* (added 27 January)

    A: Different participants will not necessarily receive the same mixtures as each other (in NOC or in ICSA).
    Labs with multiple participants, please note that the participants' responses must be independent: the participants may not collaborate with each other. The point of contact needs to be different for each subunit. (see FAQ#8 above)

23. *Q: Are we expected to report sub-source or sub-sub source likelihood ratios in the ICSA Subtest?* (added 28 January)

    A: In the ICSA Subtest instructions, we indicate that "For LR values, report that point estimate of the computed LR (i.e., not an interval/range, a unified statistic, varNOC, stratified statistic, etc." More specifically, we are expecting laboratories to report LRs computed for the ***sub-source level proposition*** *(not the sub-sub-source level).*

    This follows the best practices recommended by ASB Standard 041 (Formulating Propositions for Likelihood Ratios in Forensic DNA Interpretations, Draft 2021): "Within the capabilities of the analysis approach used, the laboratory should report results for a pair of propositions that addresses the issue of interest. This level is the highest level in the hierarchy for which the forensic scientist can provide information. Hence, the laboratory should report results given sub-source level rather than sub-sub-source level propositions."

    For context, ASB 041 provides the following summary of the hierarchy of propositions: "Proposition pairs are classified by the level of information required to assist the trier of fact: offense (e.g., "Mr. X raped V"), activity (e.g., "Mr. X had intercourse with V"), source (e.g., "The semen came from Mr. X"), sub-source (e.g., "Mr. X is a contributor to this DNA"), and sub-sub-source (e.g., "Mr. X is the minor contributor to this DNA mixture")."

24. *Q: I do not see BP Sentry (the probabilistic genotyping software that my lab and several other U.S. labs use) listed as an option in the ICSA Subtest. Can you please add it to the list of software available?* (added 01 February)

    A: We will add BP Sentry as a response option to Question #17 in the ICSA Subtest, but will not be able to update the website until Weds Feb 2. (We will also add an "Other commercial software not listed above" option for any additional commercial software tools that may be missing.)

    Note that the response options provided in the ICSA Subtest were based upon these sources:
    - *The initial responses were developed in multiple detailed discussions with the DNAmix Working Group, which contains a cross-section of notable experts from across the forensic DNA community*

- *We added additional options indicated by registered participants in the Policies and Procedures Questionnaire as of 13 Jan 2022*
- *We added additional options based on any feedback received from participants who completed the ICSA Beta Test as of 19 Jan 2022*

25. *Q: In the P&P Questionnaire (Phase 1), I am unable to enter my lab's default stochastic threshold value of 1250 RFU. Do you know how to get past the "Enter an integer" error that pops up?* (added 01 February)

A: There was an upper limit of 1000 RFU inadvertently set for this question that has now been removed.
**If anyone has had this issue when entering default values of your lab's analytical threshold (AT) or stochastic threshold (ST) in the P&P Questionnaire, please contact us so we can update your responses accordingly.*

26. *Q: I receive an error when unzipping ICSA packets containing reference profiles for consensual partners (CON). Can you please help me to access these files?* (added 24 February)

A: The name of the subfolder ("CON") refers to a system action or device reference in Windows, which causes unexpected compatibility issues when trying to extract the folder from the ZIP files.

Due to this Windows compatibility issue, all folders previously named "CON" were updated to "CONS." Please note that all study documents and packet contents descriptors will still use the "CON" abbreviation; only the subfolder names were updated.

27. *Q: The consensual partner in my ICSA packet is labeled "EC" in the HID file instead of "CON". Do I have the right file?* (added 28 February)

A: In short, "EC" stands for "Expected Contributor", but you can assume that in this ICSA Comparison Packet the "expected contributor" (EC) is the victim's consensual partner (CON).

For additional background:

- *The initial version of nomenclature for the study grouped all "elimination-type" reference profiles together (this included any victim, consensual partner, or expected contributor).*
- *Based upon discussion with the DNAmix Working Group, we decided to split each of these types of reference profiles out explicitly, as detailed in the instructions (and summarized below). We made this decision to improve clarity.*
    - *Victim (VIC)*
    - *Consensual partner (CON)*
    - *Expected contributor (EXP)*
- *Any HID files that contain "EC" in the header were likely created prior to this change.*
- *Please assume that "EC" corresponds to the type of reference profile indicated in the packet description and the subfolder from which the HID file originated (i.e., VIC, CON, or EXP).*

28. *My laboratory always reports a general LR for cases in which a sibling of the POI is also a suspect (but for whom no DNA is available). While our software does have a specific option to compute an updated LR for the sibling case, we generally do not report it unless specifically requested. Which values would you like us to report for the sibling case questions in ICSA?* (added 03 March)

*A: If your SOPs **ever** allow you to report the sibling case (wherein Hd = a sibling of the POI is a contributor to the mixture), then please provide those LR values here.*

29. *Can you send me a copy of my responses after I submit them?* (added 18 March)

A: Due to human subjects and study restrictions, we are not permitted to release individual study results to anyone during the study, including participants. Confidentiality restrictions require that any results we release are aggregated in such a way that individual participants cannot be identified.

However, you have two options if you would like a copy of your responses:

- *After review but prior to submission, you are more than welcome to screenshot your responses and print/save them for your records.*
- *If you complete all four phases of the study, you will be provided with the opportunity to receive your AnonID. Your AnonID will allow you to review your responses once the study is published.*
    - *Note: results for the NoC and ICSA Subtests will be released after the study close, but we cannot release any responses to the P&P Questionnaire or other responses that could compromise confidentiality.*