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Audit 9642: DNA IQ Method of Extracting DNA from Casework and Reference Samples Audit

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DNA Analysis FSS (August 2009)

1. Abstract

A follow-up internal audit was performed on the DNA IQ method for extracting DNA from Casework and Reference Samples. The purpose of this audit was to maintain the continuous high quality standards within the DNA Analysis laboratory.

2. Aim

To determine if the DNA IQ extraction process is fit for purpose as well as to ensure that the recommendations from Audit 8227 have been implemented.

3. Background

Through the laboratory quality system (OQI process) a number of adverse quality events were identified on the MultiPROBE® II platforms. Three OQI's (19349, 19477, and 19768) had previously been raised to address contamination events, which were investigated in Audit 8227. This is a follow-up audit to ensure quality measures that were suggested have been implemented successfully. Audit findings and recommendations are outlined in this report.

4. Findings and Observations

Observations from the Off-Deck lysis process are outlined below:

- Table 2 – Table of Reagent Volumes, on Page 5 of the SOP (24897 Automated DNA IQ™ Method of Extracting DNA) is unclear. The table lists reagent volumes all in mL, most of the reagent volumes required are less than 1mL. The addition of a formula to determine reagent volumes would assist in the event of non-standard batch sizes.
- The Auslab OFF-Deck Lysis worksheet has 20% Sarcosyl, this should be 40%. As a result the operator is required to amend the volume on the worksheet.
- The process of transferring the substrate to the spin basket is not defined. Operators can do this in 2 different ways.
- The preparation of the plate map using the BSD program for the validation configuration of batches i.e. Soccerball, is not mentioned in the SOP at all.
- Staff were using a draft copy of the SOP

outlined in the user prompt', and the specific step details are not mentioned at all.

- During the transfer of lysates to the slicprep, some of the tubes are temporarily lifted from their position. No tubes were completely removed from their position.
- The seal on the slicprep plate was not completely sealed on one corner after sealing with the heatsealer.
- It was observed that during the wash stage, a 1mL tip flicked out of the tip chute after being discarded by the fixed tip. One operator has witnessed this occurring 2 times out of 4 runs.
- A 125uL tip could not be removed automatically from the fixed tip on 2 occasions. The operator was required to remove them manually.
- There is currently a check at the end of the extraction process to ensure there is no lysate left in the original nunc tubes. This step may be more useful if performed before the extraction process.
- A new magnet is now being used. The plate now sits on the magnet with no operator intervention.
- It was observed that the de-capper on Platform A has begun to rust.
- The shaker component needs to be removed from the deck every Friday in order to perform weekly maintenance on the Monday. This could be a Workplace Health and Safety concern as the shaker is heavy and is difficult to manoeuvre with the current cabinet doors.

5. Summary

Overall, the recommendations of Audit 8227, Process Audit of the Automated DNA IQ™ System (including Off-Deck Lysis) have been addressed. Some sections are still to be addressed and have been included in this audit's recommendations. Further improvements were identified during this audit and have been outlined below.

6. Recommendations

There have been several places identified where improvements can be made. These are outlined below:

1. The addition of a formula to determine reagents amounts in Section 4 of the SOP.
2. In Table 2 in the SOP, change the reagent volumes to appropriate measurements. Currently all volumes are listed in mL, but for most of the reagents uL would be more appropriate.
3. Update the Auslab Off-Deck Lysis worksheet to state 40% Sarcosyl instead of 20%.

Audit 9642 response

1. The addition of a formula to determine reagents amounts in Section 4 of the SOP.
This point will be addressed in the next revision of the SOP 24897, it has been added as a comment and communicated to the appropriate staff and will be included as an appendix in the future.

2. In Table 2 in the SOP, change the reagent volumes to appropriate measurements. Currently all volumes are listed in mL, but for most of the reagents µL would be more appropriate.
This point will be addressed in the next revision of the SOP 24897; it has been added as a comment and communicated to the appropriate staff.

3. Update the AUSLAB Off-Deck Lysis worksheet to state 40% Sarcosyl instead of 20%.
A change request will be submitted to LISS for amendment of this mistake.

4. The addition of the specific methods for transferring the substrate to the spin basket to the SOP.
This was discussed at length previously amongst the Analytical team and a decision was made to include both methods; as each was valid and an operator was free to choose a method that they would most prefer using.

5. Addition of the method for the preparation of the BSD plate map for the validation configuration of batches i.e. Soccerball to the SOP.
This point will be addressed in the next revision of the SOP 24897; it has been added as a comment and communicated to the appropriate staff.

6. Consider acquiring an automatic de-capping machine. The manual de-capping method is inefficient, difficult in the current cabinet and has the potential to cause contamination events.
Whilst the current manual de-capping of Nunc tubes is inefficient the procedure has been redesigned to minimise the potential for cross contamination. Given the success and increase in efficiency of de-capping with the Pre-PCR automated de-capper; additional de-cappers are a priority but current budget constraints prohibit DNA Analysis purchasing additional units at this time. As noted in the following response, purchase and implementation of improved cabinets has been delayed due to budgeting issues as well

7. Investigate ergonomically designed cabinet doors for the MultiPROBE II platforms. The current cabinet doors are a Workplace Health and Safety concern.
New cabinets have already been applied for and are expected to be delivered during the move to block 3. The Perspex doors slide along a metal channel, to make sliding of the doors easier for the time being – soap will be used on the metal channels. Improved cabinets have not been sourced prior to this date due to budget restrictions.

8. The SOP requires the addition of the tube to batch paperwork sequence checking step.
This point will be addressed in the next revision of the SOP 24897; it has been added as a comment and communicated to the appropriate staff.

9. Implement weekly cleaning of the Flush Wash G13 reservoir.
This will be discussed at an Analytical meeting to determine if cleaning or renewing of the reservoir is necessary.

10. Investigate the bubbles/droplets on the outer surface of the disposable tips.
This has been investigated as part of the DNA IQ re-implementation project. The surface

EXTRACTION BATCH CONTAMINATION

Batch: CWIQEXT20080506_02

Contaminated profile:

- Position No: 7
- Lab No: 365296308
- Case No: QP800235382
- Case details: Person killed by being run over by vehicle.
- Profile details: A mixed DNA profile was obtained with an indication of two contributors. This profile could be separated into major and minor DNA profiles. The major DNA profile matched the reference DNA profile from the deceased and the other profiles from the 'scene' samples. The minor DNA profile was incomplete and matched the DNA profile from Tuuru TARE (complainant in sexual assault matter: QP800088413) where information was obtained.

Contaminating profile:

- Position No: 23 (more likely) or 24
- Lab No: 320124503 or 320124514
- Case No: QP800088413
- Case details: Alleged Sexual Assault
- Profile details: Female DNA profile obtained from 'Lip - wet' and 'Lip - dry' swabs matching the reference sample from Tuuru TARE.

Notes:

- These cases appear to be unrelated.
- The mixed DNA profile was unexpected in the former case.
- There do not appear to be any other instances of obvious/detectable contamination events on this extraction batch.
- The mixed DNA profile was re-amplified and mixture confirmed on GEN9CW20080807_02.
- Recommendation that all samples to be interpreted with caution, by utilising macro results and 'virtual' plate map of extraction batch, and by searching spreadsheet for partial profiles by using auto-filter function. Acceptance to be determined on a case-case basis with checking by competent peer reviewer.