National Laboratory for HIV Reference Services
National HIV and Retrovirology Laboratories
National Microbiology Laboratory
Public Health Agency of Canada

HTLV Serology Quality Assessment Program

Revised Summary for Panel HTLVSER 2017Oct27

2017Oct27 HTLV Serology Panel		
Panel Sample	**True Status**	**Labs Reporting Incorrect Status**
A	HTLV-I Ab Positive	
B	Negative	
C	HTLV-I Ab Positive	
D	HTLV-II Ab Positive	
E	Negative	

All participants were able to provide either the correct serology status and/or recommendation

Errors observed based on results submitted(s):

All participants reported the correct final status for all samples in the 2017Oct27 HTLV serology panel.

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Revised Final Report for Panel HTVLSER 2017Oct27

Originally Issued 12-Dec-2017 and Revised 22-Jan-2018

Introduction

The NLHRS distributed the 2017Oct27 panel and the 2018Apr19 panel on October 11th 2017. This final report is specific to the 2017Oct27 panel only and it is publicly available; however the identity of participants is not disclosed. This report is revised due to the transcriptional error found in the summary page and page 5 of the report. The corrections being made are highlighted in grey.

Panel Samples, HTLV Test Kits and Data Entry

- *Panel Composition* – Panel 2017Oct27 consisted of five samples; two HTLV negative samples (B, E), two HTLV-I positive samples (A, C) and one HTLV-II positive sample (D). Testing and characterization by the NLHRS are presented in Appendix 1. Panels were prepared and sent to 16 participants including the NLHRS on October 11th, 2017. The deadline for data entry was October 27th, 2017.

- *HTLV Test Kits* – 4 different assays were used by the 16 participants excluding the NLHRS (Figure 1). The majority of participants, 86% (13/16) performed screen testing only. One laboratory performed confirmatory testing in the absence of a screen test.

- *Data entry* - The NLHRS Quality Assessment Program used the web based Survey Monkey system to capture results.

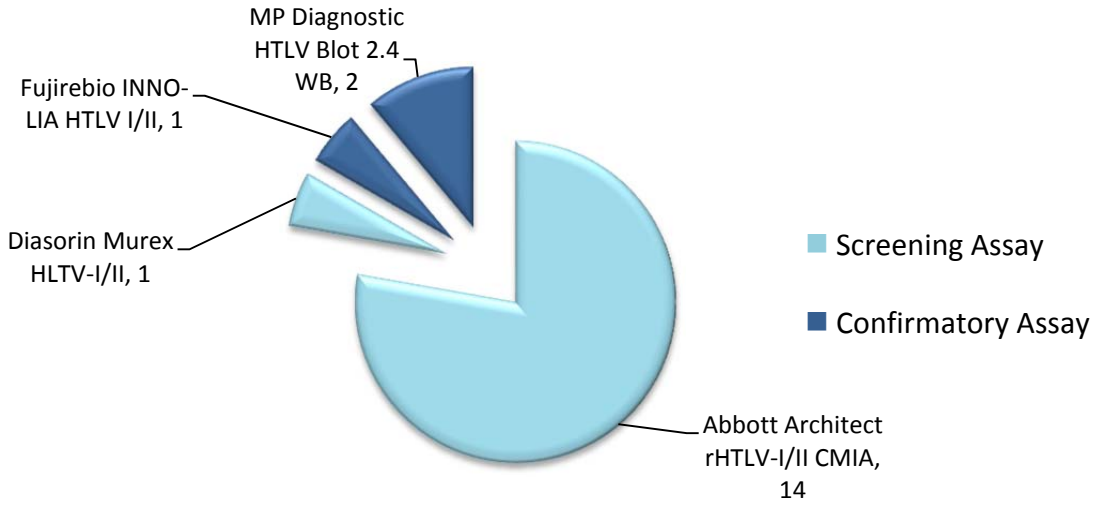

Figure 1: Assays used by the participants in the NLHRS 2017Oct27 HTLV serology panel (excludes the NLHRS)

Homogeneity and stability

- The homogeneity and stability of the 2017Oct27 HTLV serology panel is assessed by comparing the participant's results (including the NLHRS) collected after the test event is over with the results of the panel's characterization performed by the NLHRS prior to the test event
- There were no indication of heterogeneity or instability of the panel samples as the data submitted by the participants is consistent with the characterization of the panel members (Table 1 and Appendix 1)

External QC and QA activities

1. *External quality control (QC) material* - Used in addition to controls provided in kits allows users to detect technical problems and assay sensitivity from lot to lot.
   - 9 participants (56.3%, 9/16) reported using external QC material.

External QC and QA activities

2. *Quality Assurance (QA) programs*- Allows participants to evaluate their overall use of the assay and reporting of the results.
   - 13 participants (81.3%, 13/16) reported participation in other quality assurance programs. Figure 2 illustrates the different institutions which participants are enrolled in.

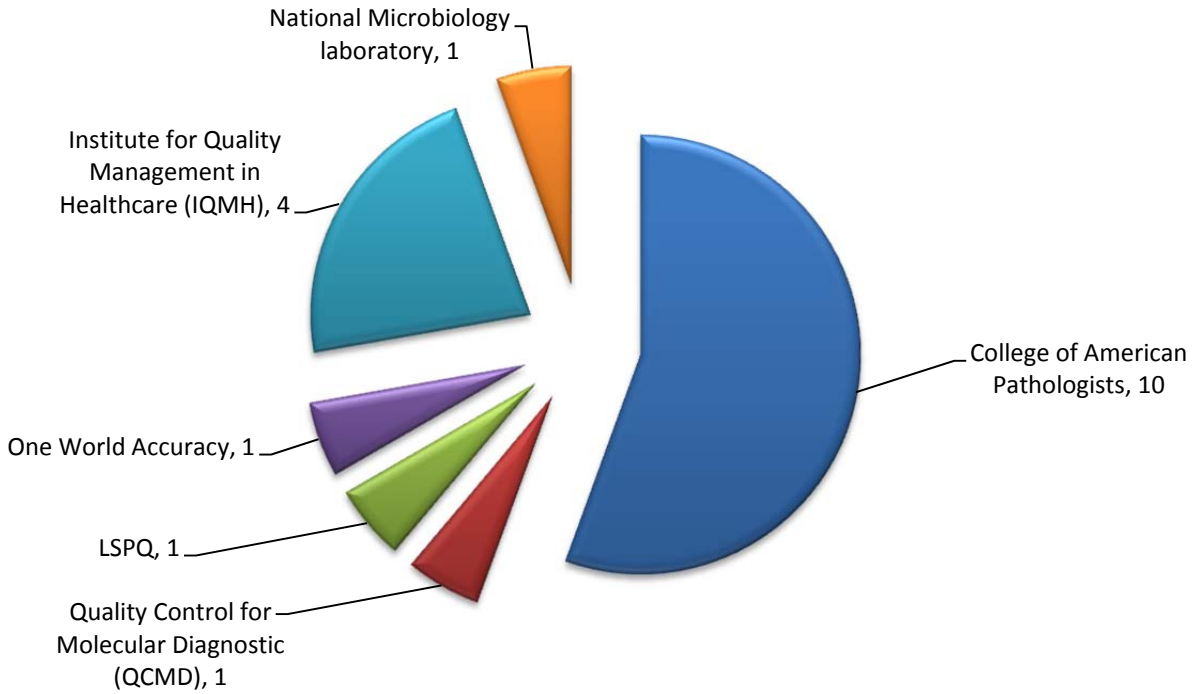

Figure 2: Distribution of external quality assurance programs which participants are enrolled in other than the NLHRS QAP

Participant's feedback

- The participants were asked to give their feedback and suggestions on how the NLHRS as a proficiency testing provider for HTLV serology could improve.
- 16 of 16 participants have provided feedback and found the NLHRS HTLV serology quality assurance program to be satisfactory (Figure 3).
- When asked which area the NLHRS could improve upon, an overwhelming response of allowing the users to print their results before results submission was identified (Figure 4)
- 4 participants were satisfied with the current format while 2 participants had no comments on which area the NLHRS could improve on.

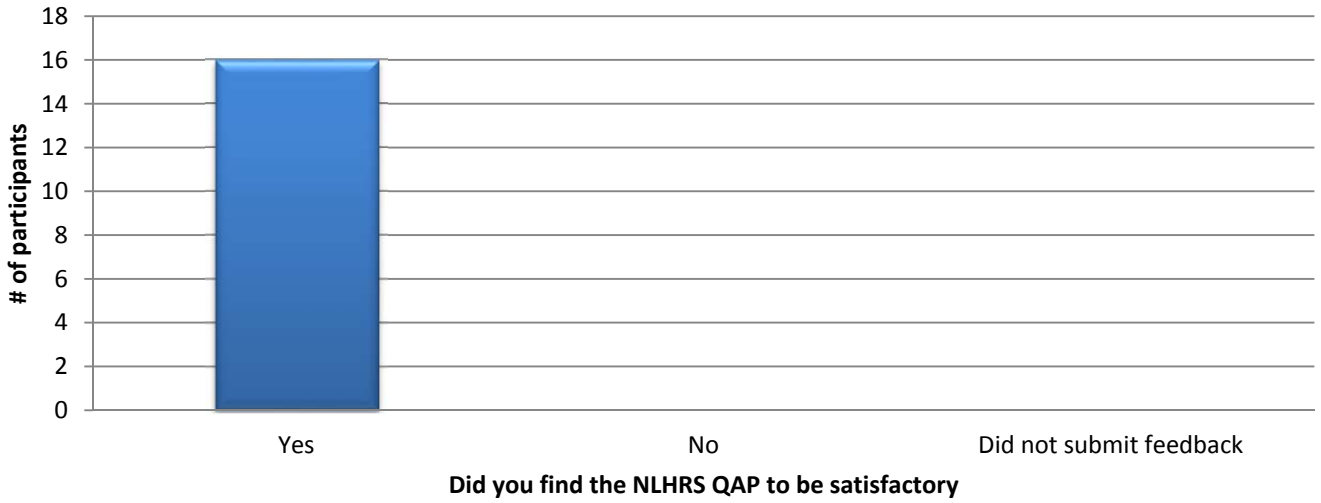

Figure 3: Participant's feedback for the NLHRS as a PT provider

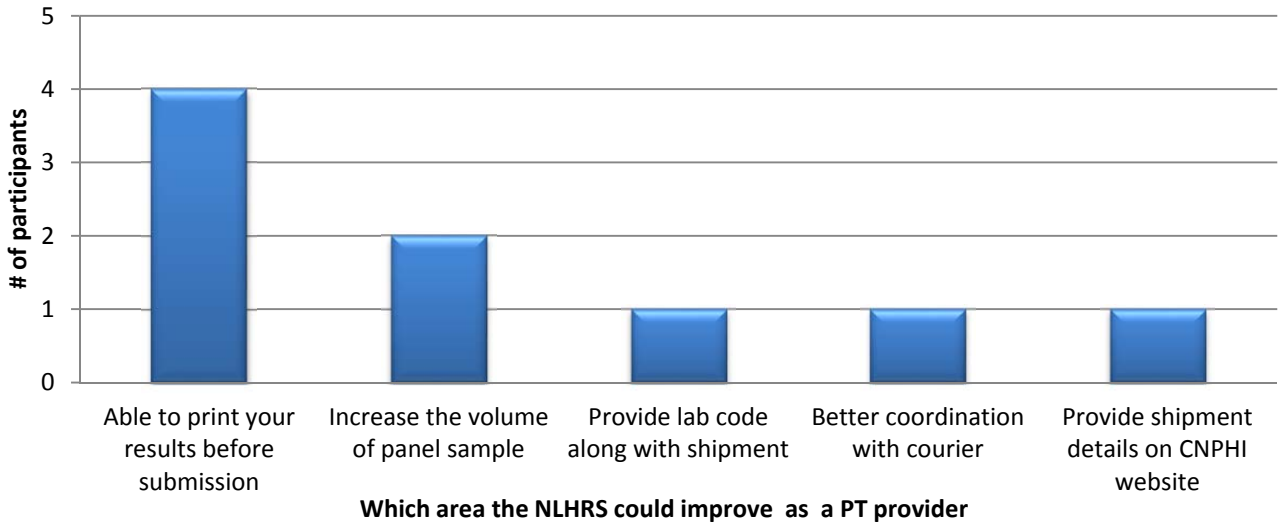

Figure 4: Participant's suggestions for improvement for the NLHRS HTLV serology quality assurance program

Legend: **Major** **Intermediate** **Minor**

Table 1: 2017Oct27 HTLV Panel final status reported from participants (includes the NLHRS).					
LAB	**SAMPLE A** **HTLV-I Ab Positive**	**SAMPLE B** **Negative**	**SAMPLE C** **HTLV-I Ab Positive**	**SAMPLE D** **HTLV-II Ab Positive**	**SAMPLE E** **Negative**
HV01	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV02	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV03	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV12	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV15	HTLV-I Ab positive	HTLV-I/II Ab negative	HTLV-I Ab positive	HTLV-II Ab positive	HTLV-I/II Ab negative
HV16	HTLV-I Ab positive	HTLV-I/II Ab negative	HTLV-I Ab positive	HTLV-II Ab positive	HTLV-I/II Ab negative
HV17	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV18	No status provided[1]	HTLV-I/II Ab negative	No status provided[1]	No status provided[1]	HTLV-I/II Ab negative
HV20	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV21	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV22	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV44	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV50	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV55	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV75	HTLV-I Ab positive	HTLV-I/II Ab negative	HTLV-I Ab positive	HTLV-II Ab positive	HTLV-I/II Ab negative
HV76	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative	HTLV-I/II Ab positive[1]	HTLV-I/II Ab positive[1]	HTLV-I/II Ab negative
HV80	HTLV-I Ab positive	HTLV-I/II Ab negative	HTLV-I Ab positive	HTLV-II Ab positive	HTLV-I/II Ab negative

[1] Further action recommended by participant; "Refer for further HTLV testing or request follow-up samples".

Table 2: Level of the different flags and the causes of the flag	
Level of flag	**Causes for flagging**
Major	Incorrect result/status provided
Intermediate	Deviation from kit insert, unresolved status without recommendation
Minor	Minor errors that do not resulted in misinterpretation of the true status of the sample, unresolved status but made a recommendation

Results

- *Return rate* - 100% of the participants returned results by the deadline (16/16).

- *Qualitative Group Analysis* (Table 1)
  - *Sample A (HTLV-I Ab positive)* – All participants provided either a correct serology status and/or recommendation.
  - *Sample B (HTLV-I/II Ab negative)* – All participants provided either a correct serology status and/or recommendation
  - *Sample C (HTLV-I Ab positive)* – All participants correctly identified the sample.
  - *Sample D (HTLV-II Ab positive)* – All participants provided either a correct serology status and/or recommendation
  - *Sample E (HTLV-I/II Ab negative)* – All participants provided either a correct serology status and/or recommendation.

Discussion

All participants returned the correct result for all samples in the 2017Oct27 panel. The participants that submitted results based on the Abbott Architect rHTLV-I/II CMIA were able to detect the HTLV-II in sample D. The two participants performing the confirmatory testing were able to correctly identified sample D as an HTLV-II.

In this panel, the NLHRS have asked the participants to provide feedbacks and suggestions for improvement in the HTLV serology Quality Assurance Program. The NLHRS was pleased to hear from our participants of their opinion on the NLHRS HTLV serology QAP. However, several areas were identified for improvements, such as allowing the participants to print their results before submission. The NLHRS will take the feedback and suggestions into consideration to improve the overall HTLV serology quality assurance program.

Conclusion

The NLHRS would like to highlight the importance of running external quality control material in their assay. External quality control material allows users to detect technical problems and lot to lot variations in the assay sensitivity. The NLHRS would like to suggest to the participants that are currently not using external quality control material in their assay to consider implementing external quality control material into their workflow.

Proficiency testing programs are designed not only to test the examination stage but the overall process in patient sample testing. As outlined in Appendix 2, errors in laboratory and medical testing can also occur during the pre-examination stage which includes all elements related to specimen collection.

The quality of HTLV antibody testing overall in Canada remains very high.

We value each laboratory's participation in these QA panels and your suggestions for improvement. The NLHRS is committed to improve all aspect of the HTLV serology proficiency testing program in order to provide quality proficiency testing services to our participants.

If you would like to make an appeal, please submit your concerns to: nlhrs-lnsrv@phac-aspc.gc.ca

Thank you for your participation in the NLHRS Quality Assurance Program

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Appendix 1: Characterization

Summary of NLHRS Characterization of the 2017Oct27 HTLV Panel Samples

The NLHRS 2017Oct27 HTLV Panel Sample Testing Results									
Sample	**Final Status**	**NLHRS Testing**							
		Fujirebio INNO-LIA HTLV I/II Score							
		Interpretation	p19 I/II	p24 I/II	gp46 I/II	gp21 I/II	p19 I	gp46 I	gp46 II
A	HTLV-I Ab positive	HTLV-I Positive	+++	+++	+++	+++	++	+++	-
B	HTLV-I/II Ab negative	Negative	-	-	-	-	-	-	-
C	HTLV-I Ab positive	HTLV-I Positive	+++	+++	+++	+++	++	+++	-
D	HTLV-II Ab positive	HTLV-II Positive	++	+++	+++	++	-	-	+++
E	HTLV-I/II Ab negative	Negative	-	-	-	-	-	-	-

N/T: Not tested

Appendix 2: Troubleshooting

Troubleshooting; common causes of outlying and/or aberrant results in Serology and Molecular Laboratories.

Type of Error	Possible Cause(s)	Pre-Analytical	Analytical	Post-Analytical
Sample mix-up	Can occur during specimen reception or testing. May result in outlying/aberrant results for one or all samples mixed-up.	✓	✓	
Transcription	• Incorrect test ordering by physician	✓		
	• Incorrect shipment address	✓		
	• Selecting the wrong assay for data entry	✓		
	• Interchanging results for two or more specimens			✓
	• Entering incorrect results			✓
	• Entering values in the incorrect field (e.g., OD as S/Co)			✓
	• Entering values in the incorrect unit (e.g., IU/mL instead of log_{10} copies/mL)			✓
	• Using a comma instead of a dot to denote a decimal point			✓
	• Selecting the incorrect assay interpretation or analyte			✓
	• Failure to recommend follow-up testing where necessary			✓
	It is recommended all results that are manually transcribed or entered electronically be checked by a second individual to avoid transcription errors.			
Outlying and/or Aberrant Results (random error)	Sporadic test results identified as outlying and/or aberrant can be classified as random events. Possible causes of random error include:			
	• Incorrect sample storage/shipping conditions	✓	✓	
	• Incorrect test method	✓	✓	
	• Insufficient mixing of sample, especially following freezing		✓	
	• Poor pipetting		✓	
	• Ineffective or inconsistent washing		✓	
	• Transcription errors	✓		✓
	• Cross-contamination or carryover	✓	✓	
	• Presence of inhibitors to PCR		✓	
Outlying and/or Aberrant Results (systematic error)	A series of test results identified as outlying and/or aberrant may be due to a systematic problem. Systematic problems may be due to:			
	• Reagents contaminated, expired or subject to batch variation		✓	
	• Instrument error or malfunction		✓	
	• Insufficient washing		✓	
	• Incorrect wavelength used to read the assay result		✓	
	• Cycling times too long/short or temperature too high/low		✓	
	• Incubation time too long/short or temperature too high/low		✓	
	• Insufficient mixing/centrifuging before testing		✓	
	• Incorrect storage of test kits and/or reagents	✓		
	• Contamination of master-mix, extraction areas or equipment		✓	
	• Ineffective extraction process		✓	
	• Degradation of master-mix components		✓	
	• Suboptimal primer design (in-house assays)		✓	

This table was modified from a report produced by the National Reference Laboratory (NRL), Melbourne, Australia.