Verification of Quantitative Methods

Purpose	**To show the instrument is fit for use after changing the front and back columns.**
Analyte	**Ethanol, Methanol, Acetone, Isopropanol**
Units of Measure	**g/100 mL**
Analyst Performing Verification Study	**Ashley Ann Johnson, M.S.**
Responsible Supervisor	**Dayong Lee, Ph.D.**
Start Date	**August 16, 2018**
Completion Date	**August 31, 2018**
Primary Matrix	**Blood**
Secondary Matrices	**Serum, Plasma, Alcoholic Beverages, Other Liquid Specimens**
Low Calibrator Concentration	**0.010**
Highest Calibrator Concentration	**0.500 (Ethanol), 0.400 (Methanol, Acetone, Isopropanol)**
Equipment/Instrument	**Headspace 2** **This instrument is fit for use on casework for Alcohol and Other Volatiles Analysis.**
Instrument Serial Number	**Headspace CN15070059** **Gas Chromatograph CN15123149**
Method	**VOLATILES**

Verification Approval

Analyst: Ashley Ann Johnson, M.S. Digitally signed by Ashley Ann Johnson, M.S. Date: 2018.09.20 09:19:44 -05'00' 9/20/18

Date

Responsible Supervisor: Dayong Lee Digitally signed by Dayong Lee DN: cn=Dayong Lee, o=Houston Forensic Science Center, ou=Toxicology, email=dlee@houstonforensicscience.org, c=US Date: 2018.09.20 09:54:38 -05'00' 9/20/18

Date

Verification Study — BIAS AND PRECISION

Analyte: **Ethanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID1**

Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	BQC2	EQC	LMQC
Target Concentration (g/100 mL):		*0.0800*	*0.2029*	*0.0800*	*0.0192*
Run 1	1	0.0790	0.1972	0.0807	0.0199
	2	0.0789	0.2013	0.0808	0.0199
ALC_20180816_AAJ2	3	0.0787	0.2017	0.0812	0.0198
Within Run	**Mean**	**0.0789**	**0.2001**	**0.0809**	**0.0199**
	SD	**0.000153**	**0.002491**	**0.000265**	**0.000058**
	%CV	**0.194%**	**1.245%**	**0.327%**	**0.291%**
	% Bias	**-1.42%**	**-1.40%**	**1.12%**	**3.47%**
Run 2	1	0.0791	0.1968	0.0803	0.0198
	2	0.0793	0.1996	0.0808	0.0199
ALC_20180831_AAJ	3	0.0796	0.1993	0.0808	0.0198
Within Run	**Mean**	**0.0793**	**0.1986**	**0.0806**	**0.0198**
	SD	**0.000252**	**0.001537**	**0.000289**	**0.000058**
	%CV	**0.317%**	**0.774%**	**0.358%**	**0.291%**
	% Bias	**-0.83%**	**-2.14%**	**0.79%**	**3.30%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037); BQC2 (Lot: 1803028); EQC (Lot: 28082014-B)

Acceptance Criteria: **Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.**

Verification Study — BIAS AND PRECISION

Analyte: **Ethanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID2**

Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	BQC2	EQC	LMQC
Target Concentration (g/100 mL):		*0.0800*	*0.2029*	*0.0800*	*0.0192*
Run 1	1	0.0785	0.1975	0.0804	0.0198
	2	0.0783	0.2020	0.0796	0.0197
ALC_20180816_AAJ2	3	0.0778	0.2007	0.0798	0.0197
Within Run	**Mean**	**0.0782**	**0.2001**	**0.0799**	**0.0197**
	SD	**0.000361**	**0.002316**	**0.000416**	**0.000058**
	%CV	**0.461%**	**1.158%**	**0.521%**	**0.293%**
	% Bias	**-2.25%**	**-1.40%**	**-0.08%**	**2.78%**
Run 2	1	0.0795	0.1987	0.0803	0.0198
	2	0.0797	0.2014	0.0810	0.0199
ALC_20180831_AAJ	3	0.0800	0.2015	0.0807	0.0199
Within Run	**Mean**	**0.0797**	**0.2005**	**0.0807**	**0.0199**
	SD	**0.000252**	**0.001589**	**0.000351**	**0.000058**
	%CV	**0.316%**	**0.792%**	**0.435%**	**0.291%**
	% Bias	**-0.33%**	**-1.17%**	**0.83%**	**3.47%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037); BQC2 (Lot: 1803028); EQC (Lot: 28082014-B)

Acceptance Criteria: **Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.**

Verification Study — BIAS AND PRECISION

Analyte: **Methanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID1**

Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0364*	*0.0192*
Run 1	1	0.0354	0.0199
	2	0.0354	0.0198
ALC_20180816_AAJ2	3	0.0354	0.0198
Within Run	**Mean**	**0.0354**	**0.0198**
	SD	**0.000000**	**0.000058**
	%CV	**0.000%**	**0.291%**
	% Bias	**-2.75%**	**3.30%**
Run 2	1	0.0354	0.0196
	2	0.0357	0.0198
ALC_20180831_AAJ	3	0.0362	0.0198
Within Run	**Mean**	**0.0358**	**0.0197**
	SD	**0.000404**	**0.000115**
	%CV	**1.130%**	**0.585%**
	% Bias	**-1.74%**	**2.78%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.

Verification Study — BIAS AND PRECISION

Analyte: **Methanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID2**

Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0364*	*0.0192*
Run 1	1	0.0351	0.0196
	2	0.0351	0.0195
ALC_20180816_AAJ2	3	0.0362	0.0195
Within Run	**Mean**	**0.0355**	**0.0195**
	SD	**0.000635**	**0.000058**
	%CV	**1.791%**	**0.296%**
	% Bias	**-2.56%**	**1.74%**
Run 2	1	0.0367	0.0196
	2	0.0367	0.0198
ALC_20180831_AAJ	3	0.0362	0.0199
Within Run	**Mean**	**0.0365**	**0.0198**
	SD	**0.000289**	**0.000153**
	%CV	**0.790%**	**0.773%**
	% Bias	**0.37%**	**2.95%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.

Verification Study — BIAS AND PRECISION

Analyte: **Isopropanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID1**
Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0399*	*0.0192*
Run 1	1	0.0390	0.0199
	2	0.0395	0.0199
ALC_20180816_AAJ2	3	0.0393	0.0198
Within Run	**Mean**	**0.0393**	**0.0199**
	SD	**0.000252**	**0.000058**
	%CV	**0.641%**	**0.291%**
	% Bias	**-1.59%**	**3.47%**
Run 2	1	0.0397	0.0196
	2	0.0396	0.0195
ALC_20180831_AAJ	3	0.0396	0.0198
Within Run	**Mean**	**0.0396**	**0.0196**
	SD	**0.000058**	**0.000153**
	%CV	**0.146%**	**0.778%**
	% Bias	**-0.67%**	**2.26%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.

Verification Study — BIAS AND PRECISION

Analyte: **Isopropanol**
Units: **g/100 mL**
Instrument: **Headspace 2 FID2**
Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0399*	*0.0192*
Run 1	1	0.0391	0.0196
	2	0.0389	0.0196
ALC_20180816_AAJ2	3	0.0386	0.0195
Within Run	**Mean**	**0.0389**	**0.0196**
	SD	**0.000252**	**0.000058**
	%CV	**0.647%**	**0.295%**
	% Bias	**-2.59%**	**1.91%**
Run 2	1	0.0401	0.0199
	2	0.0400	0.0197
ALC_20180831_AAJ	3	0.0401	0.0197
Within Run	**Mean**	**0.0401**	**0.0198**
	SD	**0.000058**	**0.000115**
	%CV	**0.144%**	**0.584%**
	% Bias	**0.42%**	**2.95%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.

Verification Study — BIAS AND PRECISION

Analyte: **Acetone**
Units: **g/100 mL**
Instrument: **Headspace 2 FID1**
Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0416*	*0.0192*
Run 1	1	0.0403	0.0196
	2	0.0400	0.0196
ALC_20180816_AAJ2	3	0.0399	0.0195
Within Run	**Mean**	**0.0401**	**0.0196**
	SD	**0.000208**	**0.000058**
	%CV	**0.520%**	**0.295%**
	% Bias	**-3.69%**	**1.91%**
Run 2	1	0.0416	0.0191
	2	0.0411	0.0189
ALC_20180831_AAJ	3	0.0413	0.0189
Within Run	**Mean**	**0.0413**	**0.0190**
	SD	**0.000252**	**0.000115**
	%CV	**0.609%**	**0.609%**
	% Bias	**-0.64%**	**-1.22%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: **Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.**

Verification Study — BIAS AND PRECISION

Analyte: **Acetone**
Units: **g/100 mL**
Instrument: **Headspace 2 FID2**
Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Matrix: **Blood**

Run Date	Run Order	MQC1	LMQC
Target Concentration (g/100 mL):		*0.0416*	*0.0192*
Run 1	1	0.0400	0.0194
	2	0.0395	0.0192
ALC_20180816_AAJ2	3	0.0391	0.0191
Within Run	**Mean**	**0.0395**	**0.0192**
	SD	**0.000451**	**0.000153**
	%CV	**1.141%**	**0.794%**
	% Bias	**-4.97%**	**0.17%**
Run 2	1	0.0418	0.0192
	2	0.0414	0.0189
ALC_20180831_AAJ	3	0.0416	0.0189
Within Run	**Mean**	**0.0416**	**0.0190**
	SD	**0.000200**	**0.000173**
	%CV	**0.481%**	**0.912%**
	% Bias	**0.00%**	**-1.04%**

Comments: LMQC (Lot: 022818-LMQC); MQC1 (Lot: 1801037)

Acceptance Criteria: **Refer to the Analysis of Alcohol and Other Volatiles by Headspace GC-FID section of the Toxicology Section Analytical Manual Standard Operating Procedures Version 3.1.**

SUMMARY OF VERIFICATION PERFORMANCE

Analyst: **Ashley Ann Johnson, M.S.**
Study Dates: **8/16/18 to 8/31/18**
Instrument: ***Headspace 2***
Matrix: **Blood**

Failed Runs (include dates/reasons):

ALC_20180822_AAJ2 analyzed on 8/22/2018: The FID1A Acetone value for 0.010 g/100 mL Mixed Volatile Calibrator was outside of the acceptance range. The data in this run were not used for verification.

Deviations from SOP:

N/A

Other Notes:

After changing both the front and back columns, retention times were updated in the batch sequence calibration tables. This was accomplished by finding the average retention time per analyte across all calibration levels, updating the calibration table for each analyte, and reprocessing all data with the updated calibration table. The front column retention times were updated using verification run ALC_20180816_AAJ2. The back column retention times were updated using verification run ALC_20180831_AAJ. Following this successful verification, the master VOLATILES.M calibration table will be revised to reflect the updated analyte retention times.

Conclusion:

Headspace 2 is fit for use on casework analysis of ethanol, methanol, isopropanol and acetone after maintenance.