Electronic Supplementary Material (ESI) for Analytical Methods.

Method development for Forensic Oil Identification by Direct Analysis in Real Time-Time of Flight Mass Spectrometry

Krishnaja Tikkisetty[a], Taylor Filewood[a], Jeffrey Yan[a], Honoria Kwok[a], Pamela Brunswick[a], Robert Cody[b], Dayue Shang[a*]

[a]Science and Technology Branch, Pacific Environmental Science Centre, Environment and Climate Change Canada, Pacific and Yukon Laboratory for Environmental Testing, North Vancouver, British Columbia, Canada.

[b] JEOL USA Inc., Peabody, Massachusetts, United States.

*Email: dayue.shang@ec.gc.ca;

Supplementary Data:

Tables:

Table S1: Background information for Round Robin 2017-2020 tests

Round Robin Year	Oil Description
2017	Oil slick spill in the Norwegian Sea. Some of the oil was ignited. Spill sample A was artificially weathered to replicate the weathering of the spilled oil and then heating to over 250 °C. Spill sample B is the ignited spilled oil. Sources A, B and C were three potential suspected sources.
2018	An oil spill was detected in a bay of the sea after a rainy day, which was later traced back to a storm drain water pipe. Three source oil samples were taken from three gas stations that were further upstream from the pipe. The spill sample was taken from the still storm drainpipe water.
2019	Two spill samples were collected from a spill that occurred in a lagoon off the coast of the Mediterranean Sea. Each spill sample was taken from a different location of the lagoon. One source sample was taken from a ship that was in the vicinity at the time of the spill. The other two source oils were collected from a lubricating-oil producing factory in the surroundings.

2020	A thick slick of oil emulsions was spotted in a port in the winter. The following summer, films of oil were found in the same location. One spill oil sample was taken from the winter spill and the other spill sample was collected from the summer oil films. Source oils were taken from nearby land-based, re-fuelling pipelines. One theory is that a leak occurred during re-fueling. Another possibility is that the pipes or connected oil tanks were over filled.

Table S2: Experimental information for Round Robin oil GC/FID and GC/MS

Method	**Description**
GC/FID	An Agilent 6890JN gas chromatograph that was equipped with a flame-ionization detector as well as an Agilent 7683 autosampler. The apparatus was operated using by Agilent OpenLab CDS software rev. C.01.07.SR2. Analysis was performed on a Restek Rtx-5 fused silica column (30 m x 0.32 mm d x 0.25 um film thickness) with helium carrier gas at 1.8mL/min. The inlet temperature was set to 250^{o}C with 25:1 split injection. The oven temperature started at 45^{o}C with a 2 min hold time. Afterwards, the temperature increased by 8^{o}C per minute until the temperature reached 325^{o}C, with a total run time of 61.25 min. Performance quality control wasw confirmed by the analysis of Supelco ASTM D2887 calibration mix (HCID locator). GC/FID chromatographic profiles were visually assessed. Spreadsheet diagnostics were also used in analysis.
GC/MS/MS Analysis	Analysis of targeted compounds was performed on an Agilent 7890B GC. A Agilent 7683 autosampler and 7010 triple quad mass spectrometer completed the apparatus. Pseudo Multiple Reaction ion Monitoring was used for analysis. A Restek Rtx-5MS fused silica column (30m x 0.25 mm id x 0.25 um film thickness) with helium gas (1.0 mL/min) used for analyte separation. The oven temperature started at 42^{o}C with a 2 min hold time. Afterwards, the temperature

	increased by 5.5°C per minute until the temperature reached 330°C at a hold time of 16 minutes, with a total run time of 70.364 min. Confirmation of instrument performance was achieved via analysis of QC samples of Sintef oil at 0.1 mg/mL in DCM analyzed in each analytical sequence.

Table S3: File Names for Round Robin Oils

Sample Name	**Sample Type**
Spill A	RR2017 Spill
Spill B	RR2017 Spill
Source A	RR2017 Source
Source B	RR2017 Source
Source C	RR2017 Source
Spill	RR2018 Spill
Source A	RR2018 Source
Source B	RR2018 Source
Source C	RR2018 Source
Spill 1	RR2019 Spill
Spill 2	RR2019 Spill
Source 1	RR2019 Source
Source 2	RR2019 Source
Source 3	RR2019 Source
Spill 1	RR2020 Spill
Spill 2	RR2020 Spill
Source A	RR2020 Source
Source B	RR2020 Source
Source C	RR2020 Source
Spill 1	RR2021 Spill
Spill 2	RR2021 Spill
Source A	RR2021 Source
Source B	RR2021 Source
Source C	RR2021 Source

Table S4: Model Information

Model Description	Number of spectra in the training set	Number of Features	Mass Range	Number of Principal Components Used	Variance covered (%)	DAPC LOOCV scores (%)	DAPC External Validation Score (%)
RR2017	18	920	81.06986-	8	96.47	100	83.33

			799.66675				
RR2018	18	166	95.08488-840.64246	11	96.64	94.74	100
RR2019	18	1178	71.08664-981.13654	8	97.81	100	100
RR2020	18	1022	81.0695-787.63696	10	95.34	100	100
RR2021	18	159	135.11737 - 635.38574	5	87.80	100	100

Table S5: Results versus Actual

Spill Sample	DART/TOFMS Prediction	Prediction Agreement	Prediction Average Confidence (%)	Classical Analysis Classification
RR2017 Spill A	B	8/8	99.10	B
RR2017 Spill B	B	8/8	98.98	B
RR2018 Spill	C	8/8	100.00	C
RR2019 Spill 1	2	8/8	94.67	2
RR2019 Spill 2	2/1	4/8	100.00	1
RR2020 Spill 1	B	8/8	100.00	B
RR2020 Spill 2	B	8/8	98.88	B
RR2021 Spill 1	B	5/8	100.00	B
RR2021 Spill 2	B	8/8	100.00	B

Figures:

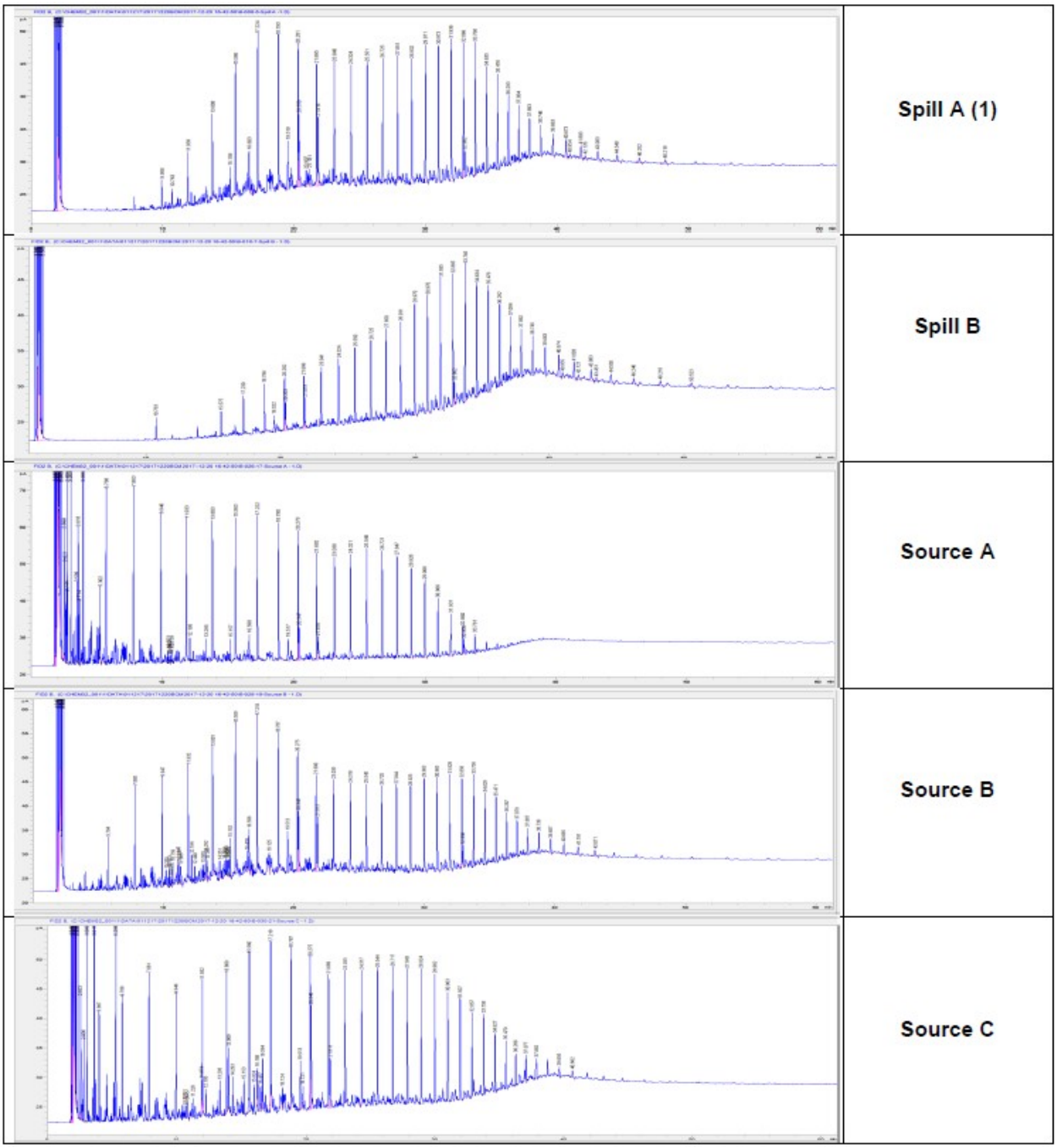

Figure S1: GC/FID chromatography for Round Robin 2017 samples

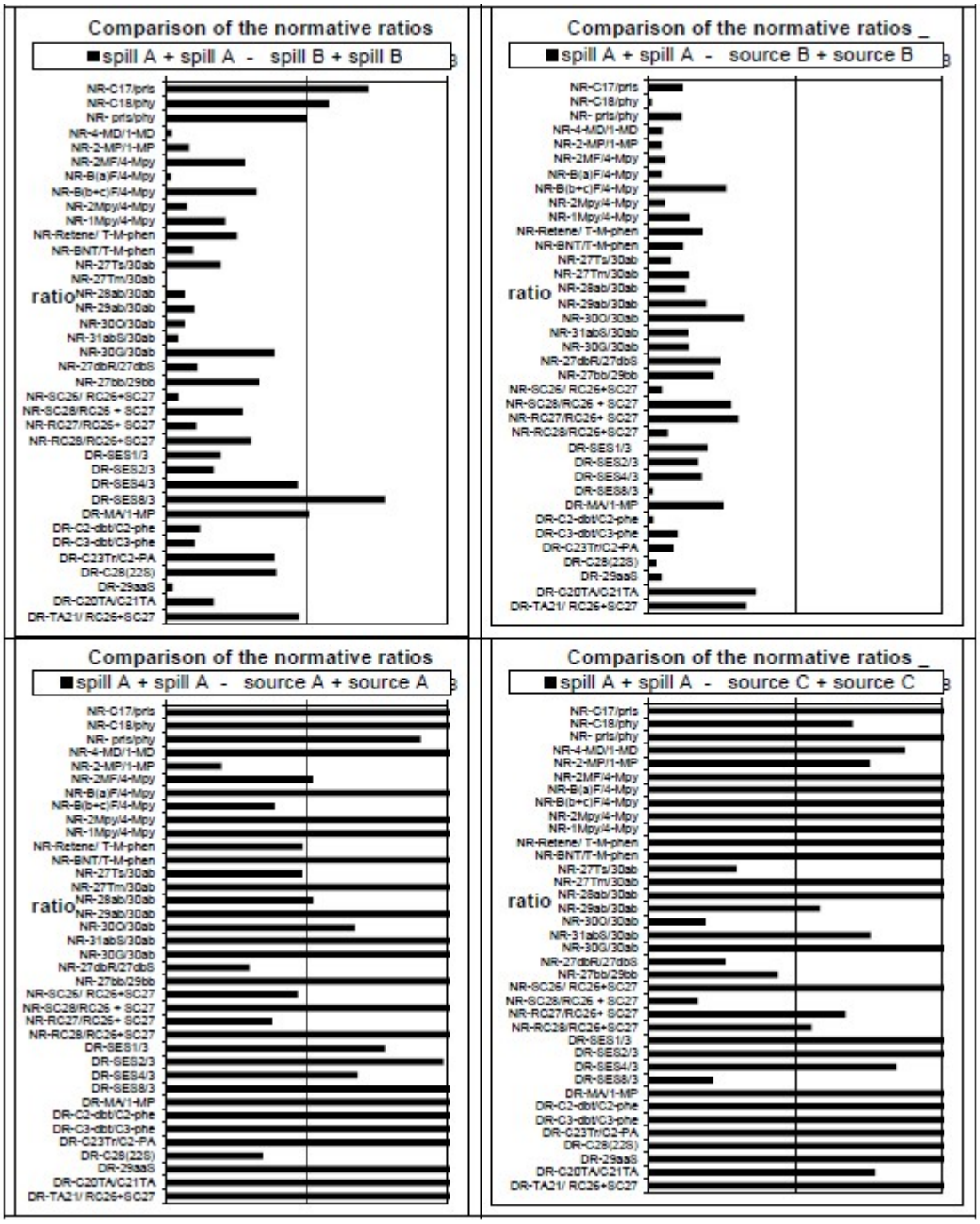

Figure S2: Round Robin 2017 GC/MS Comparison of Diagnostic ratios (normative) of Spill A compared with Spill B and Source oils

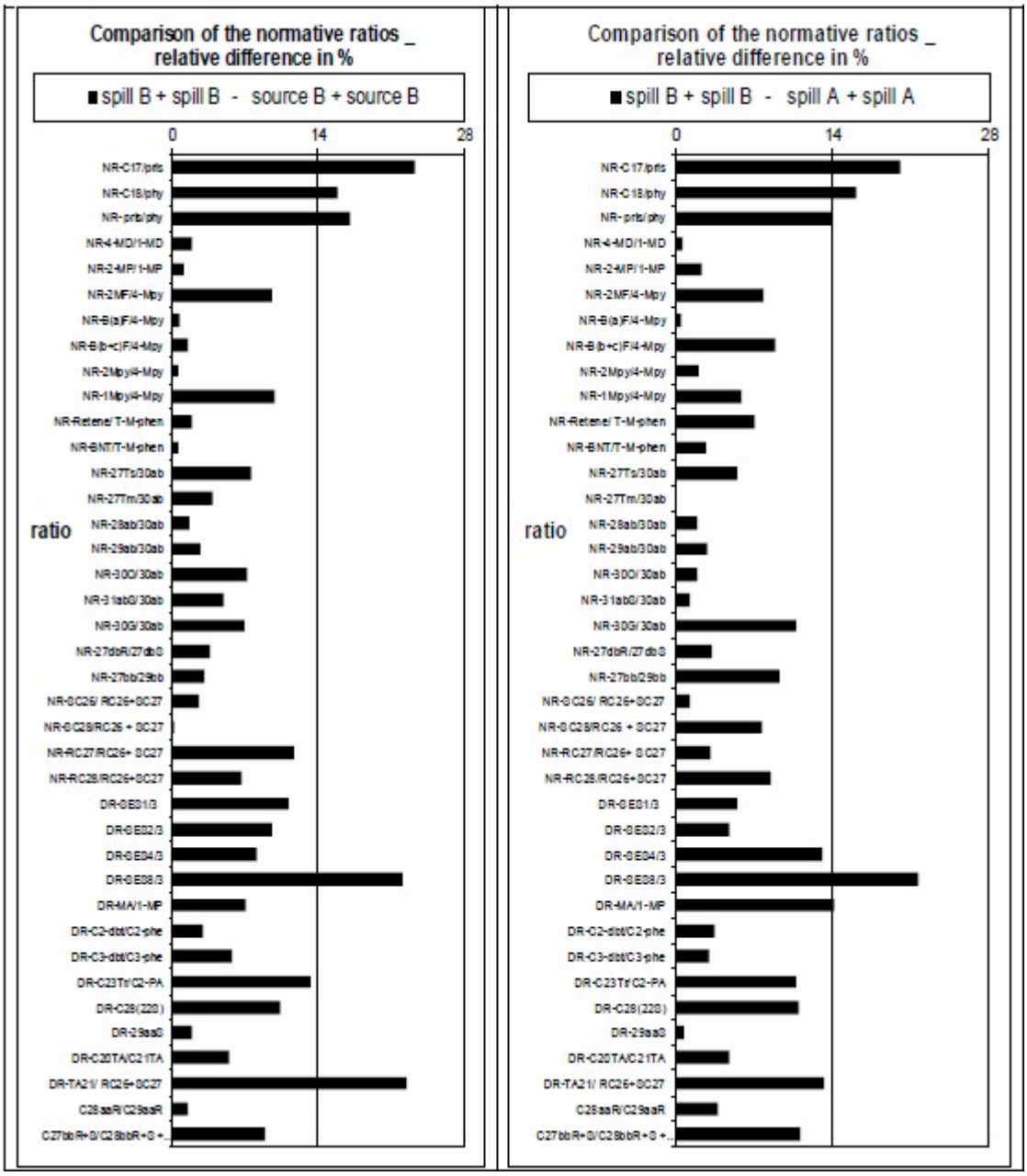

Figure S3: Round Robin 2017 GC/MS Comparison of a diagnostic and normative ratios of Spill B compared with Spill A and Source B

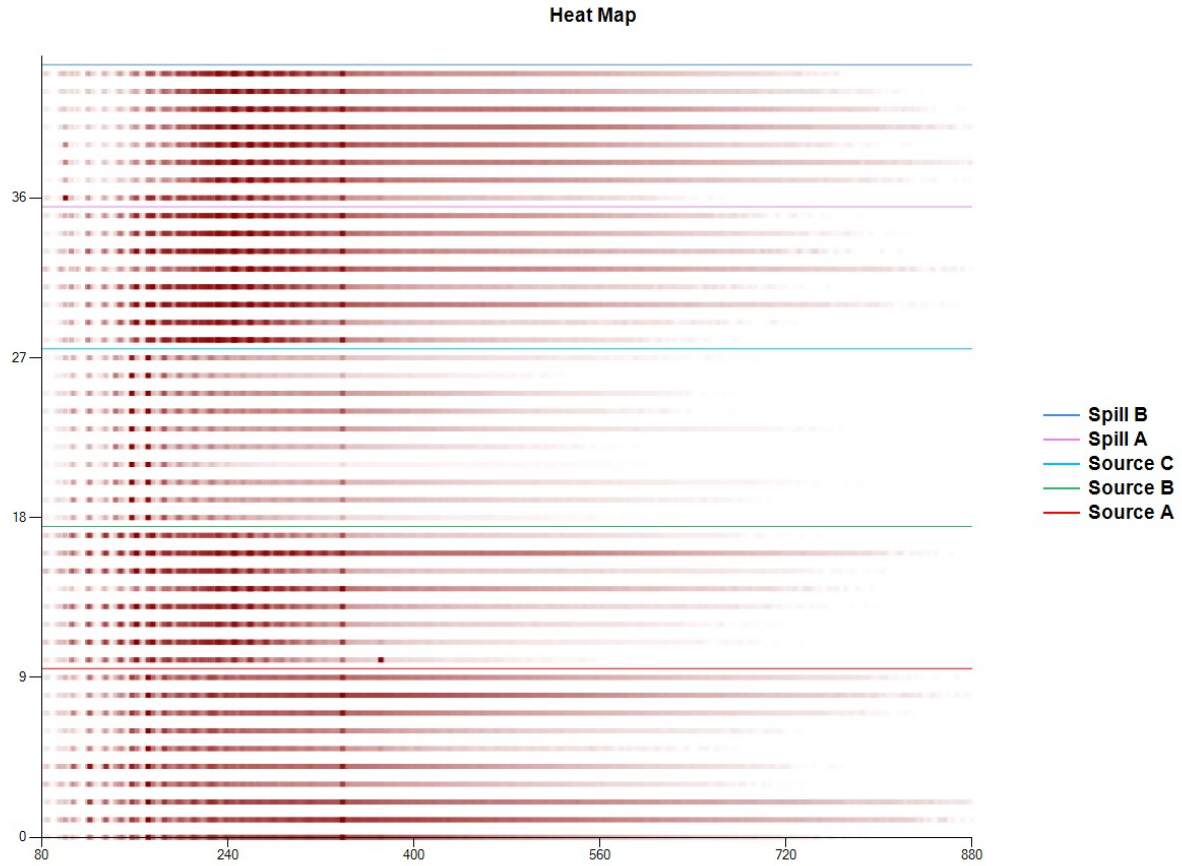

Figure S4: Round Robin 2017 Heatmap

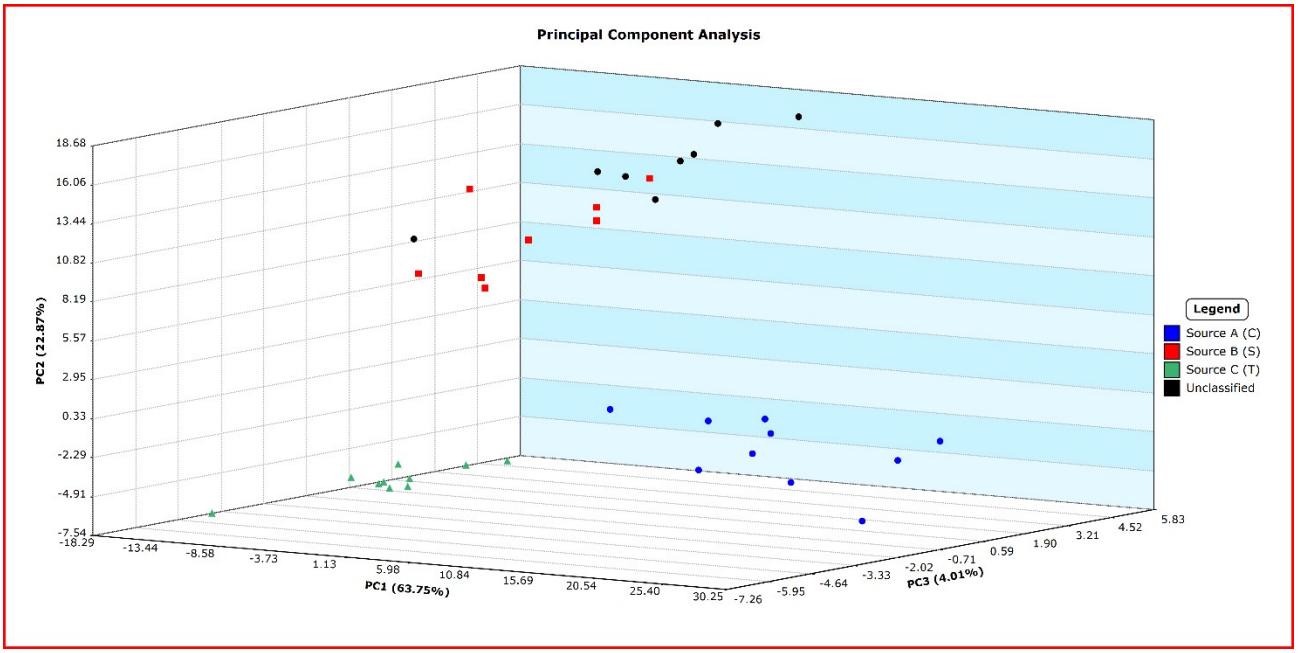

Figure S5: Round Robin 2017 Spill A PCA

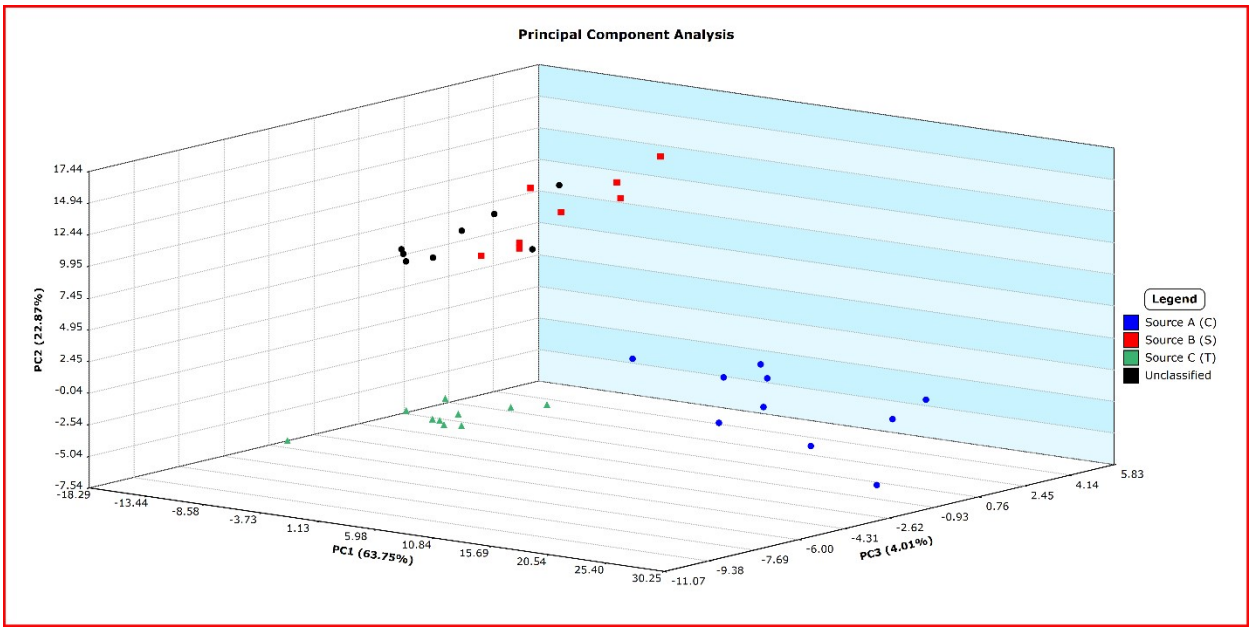

Figure S6: Round Robin 2017 Spill B PCA

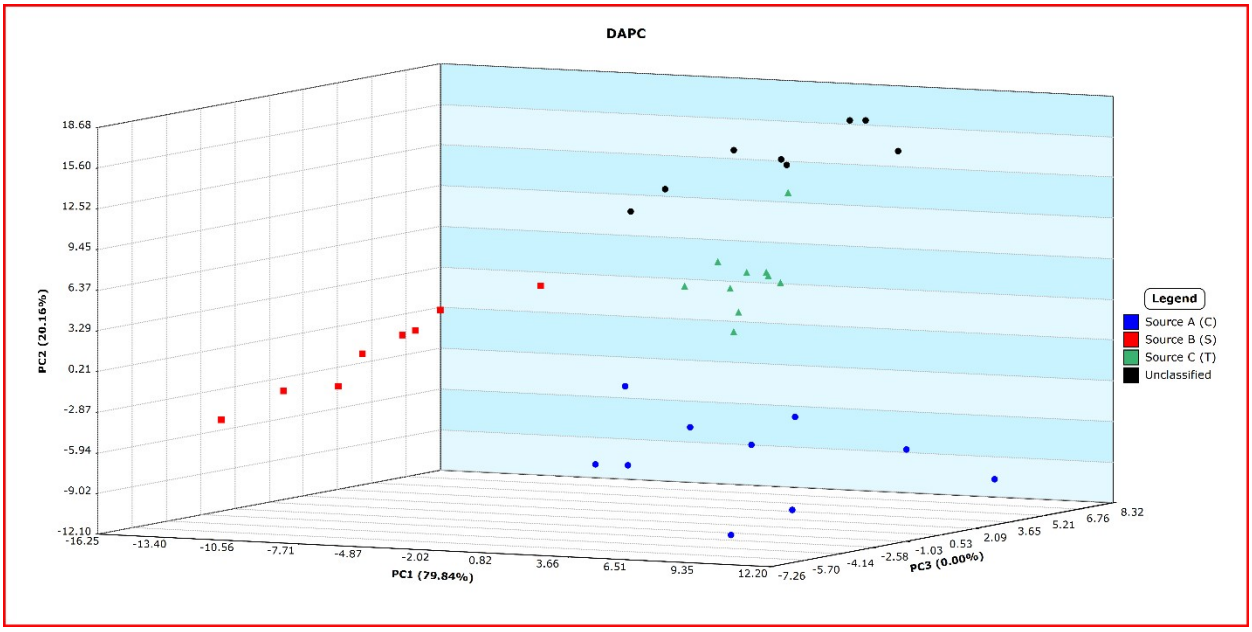

Figure S7: Round Robin 2017 Spill A DAPC

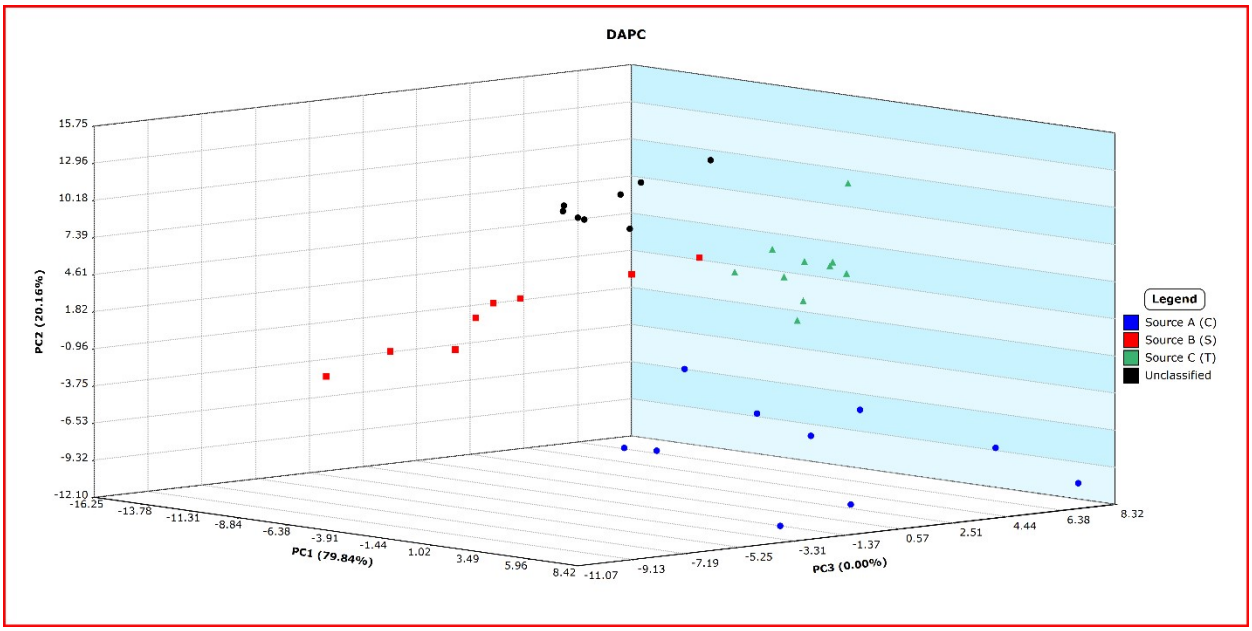

Figure S8: Round Robin 2017 Spill B DAPC

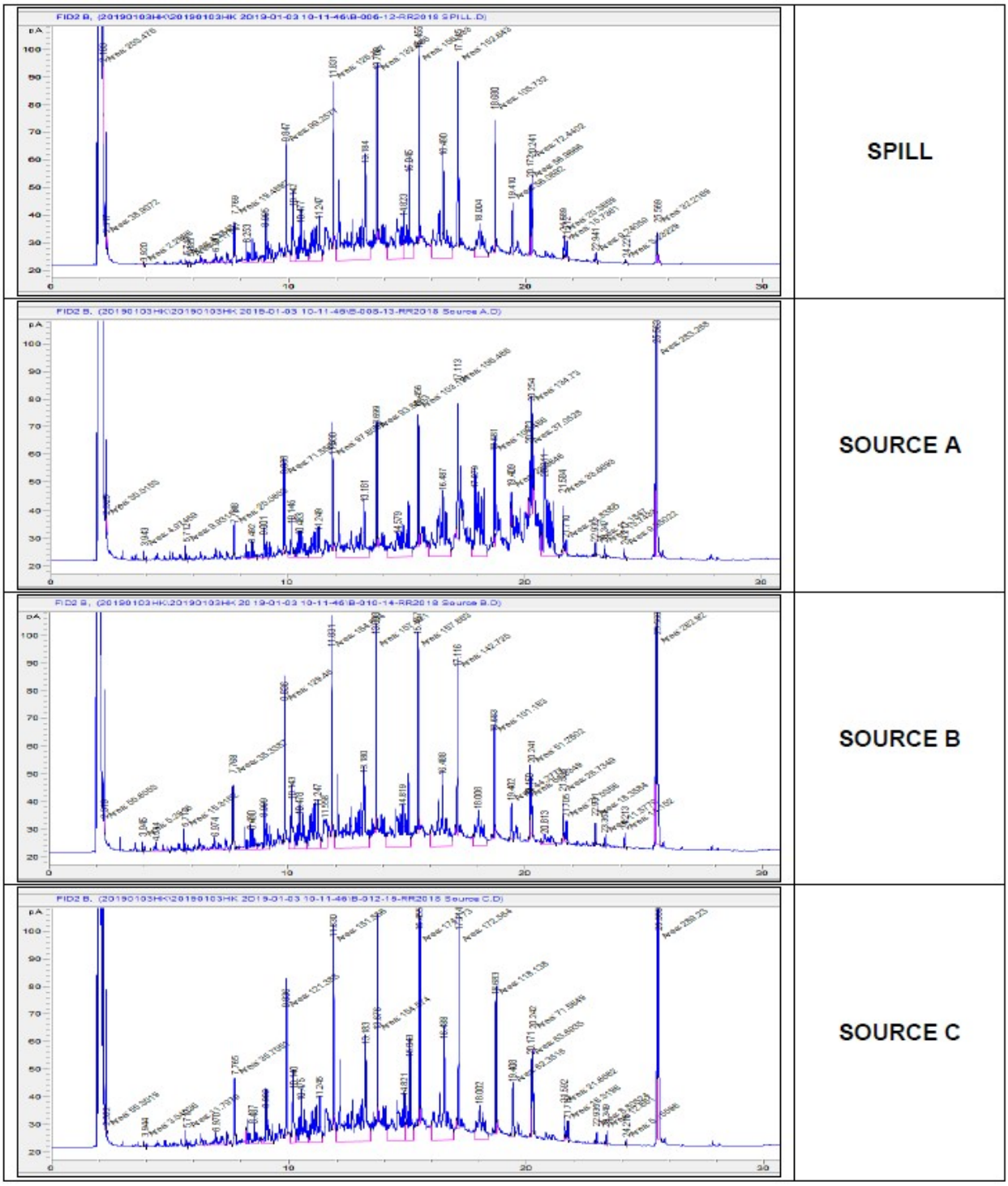

Figure S9: GC/FID chromatography for Round Robin 2018 samples

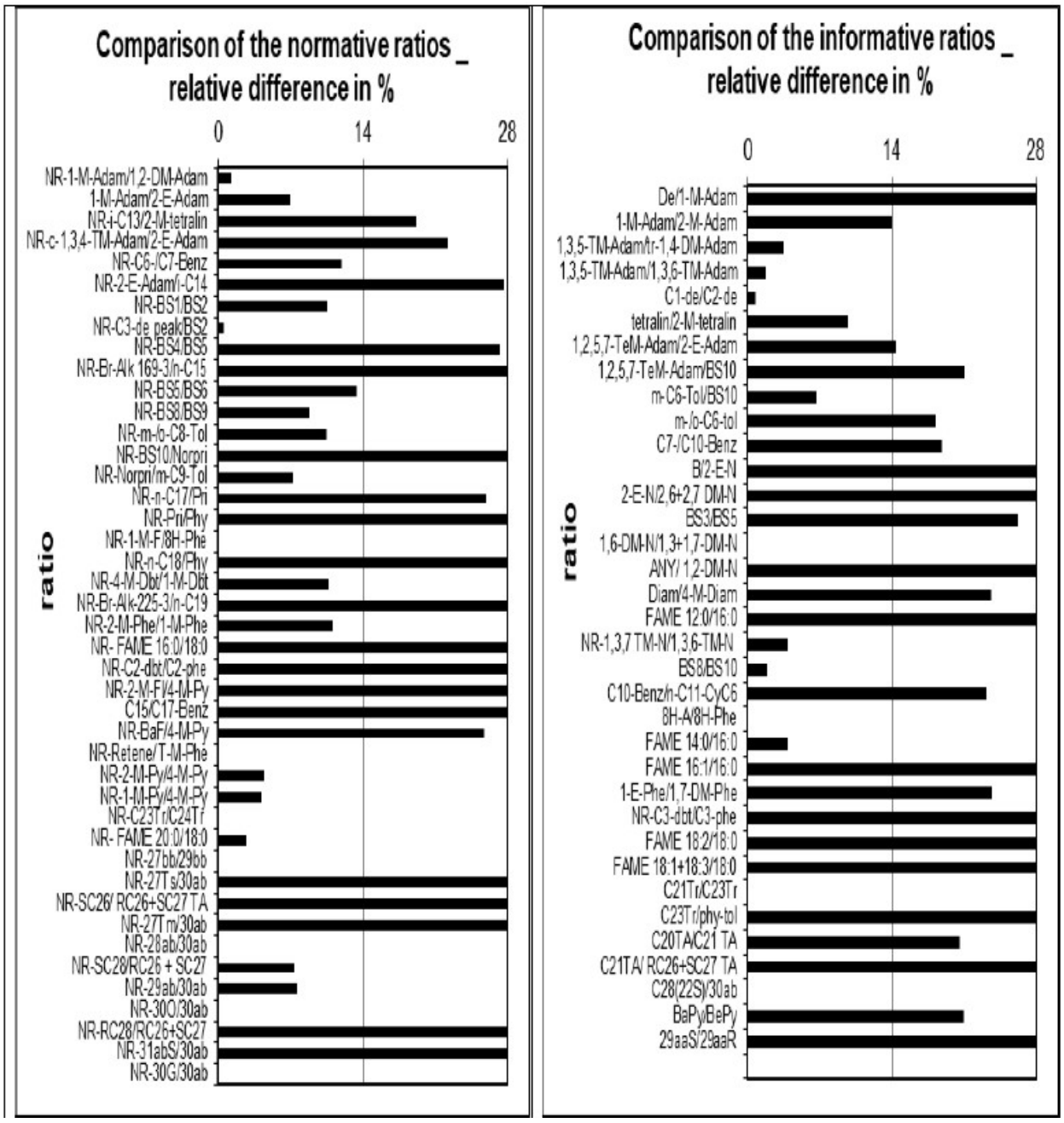

Figure S10: GC/MS comparison of Diagnostic ratios of Spill compared with Source A

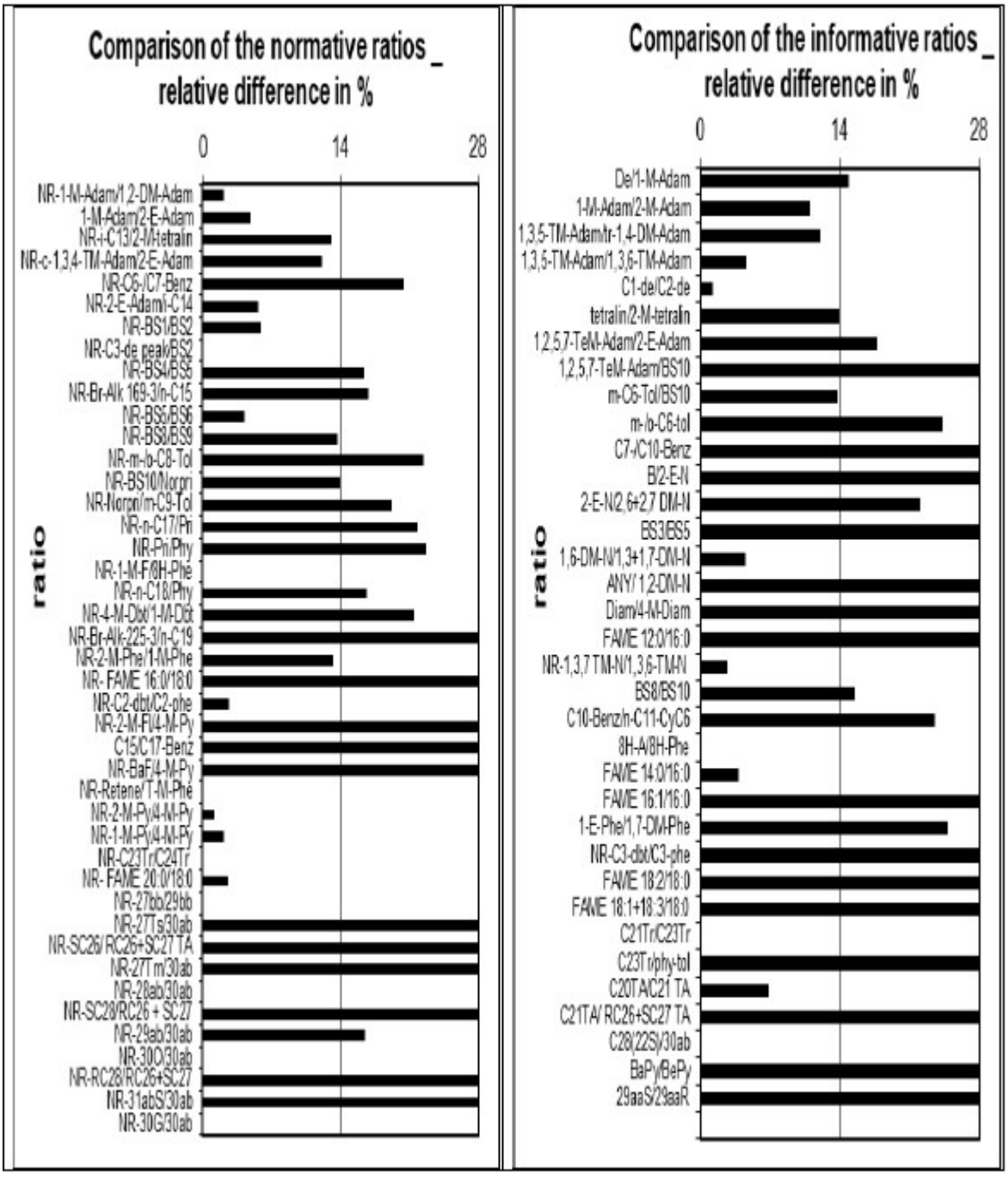

Figure S11: GC/MS comparison of Diagnostic ratios of Spill compared with Source B

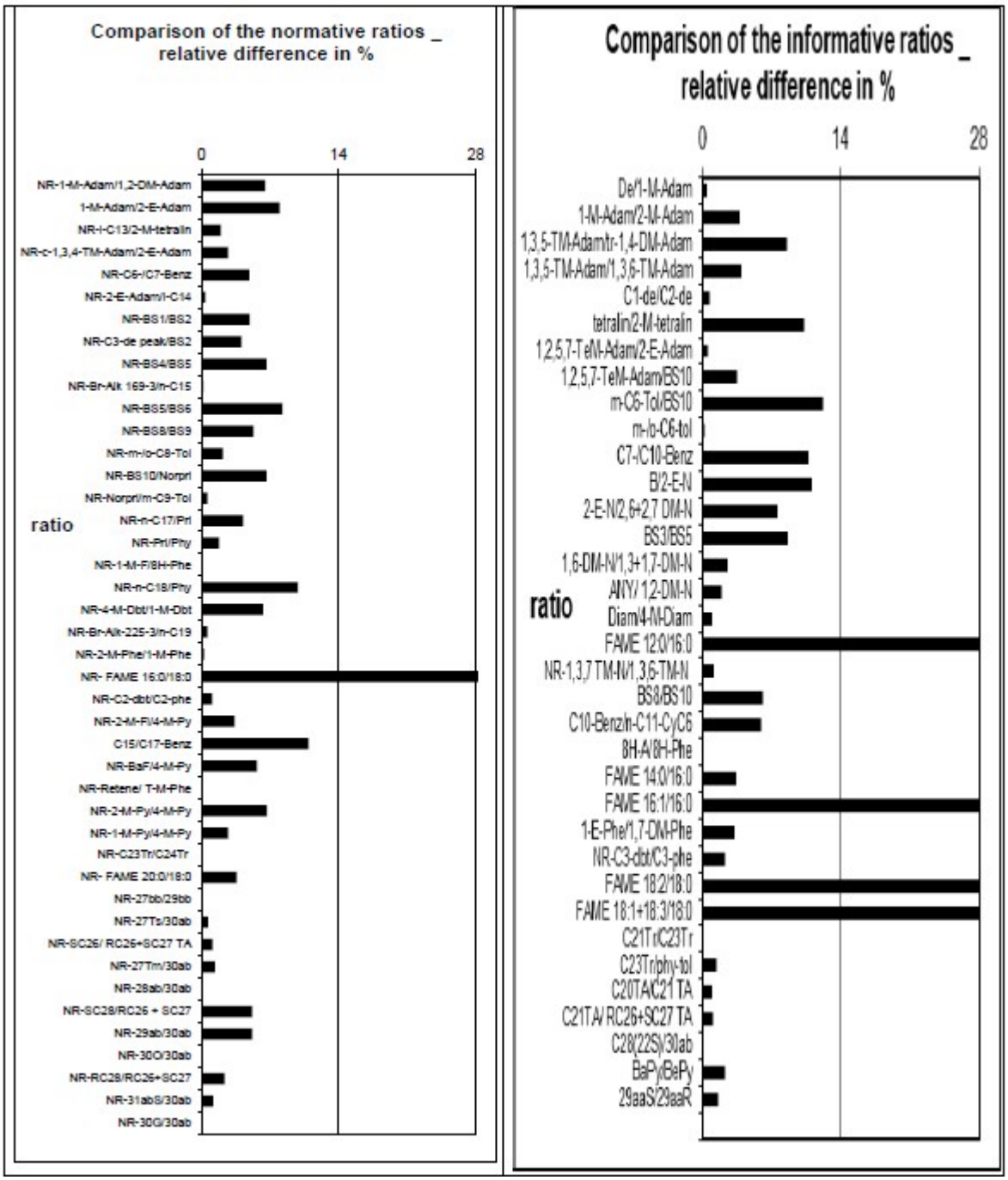

Figure S12: GC/MS comparison of Diagnostic ratios of Spill compared with Source C

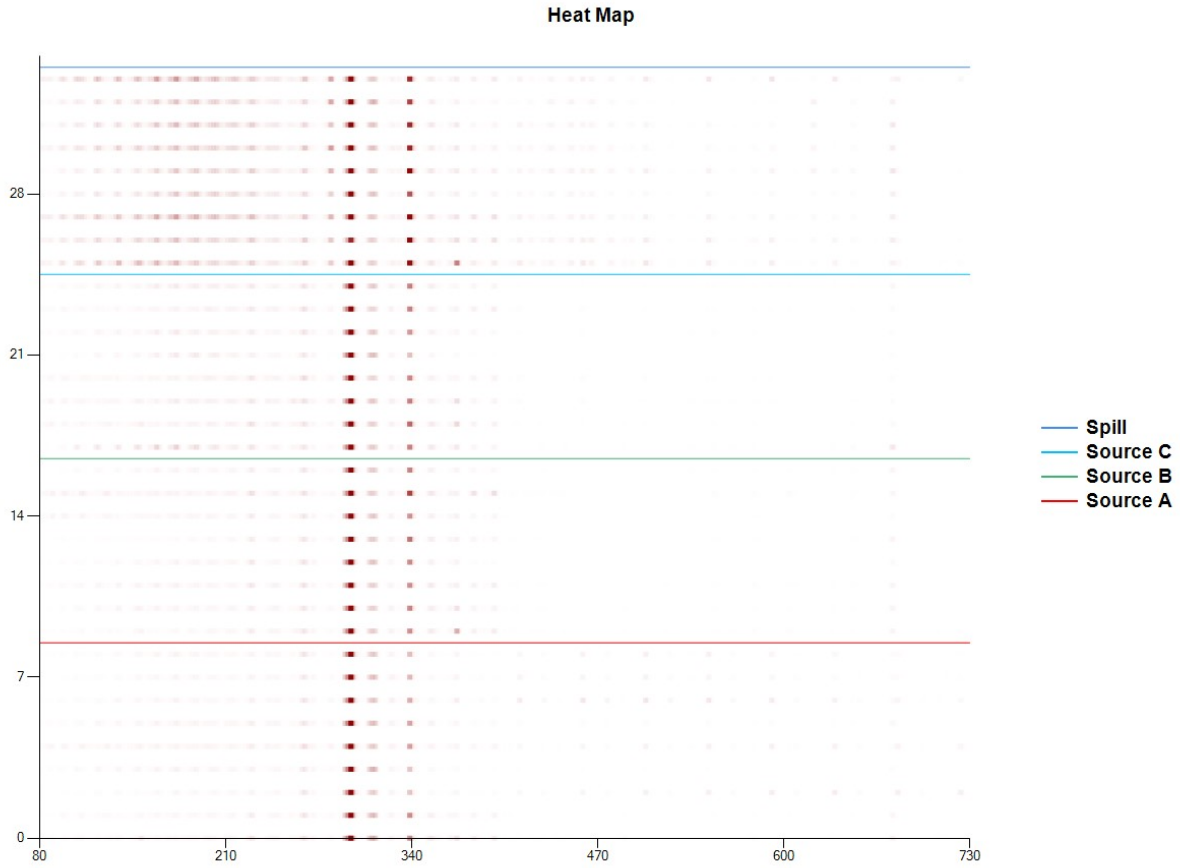

Figure S13: Round Robin 2018 Heatmap

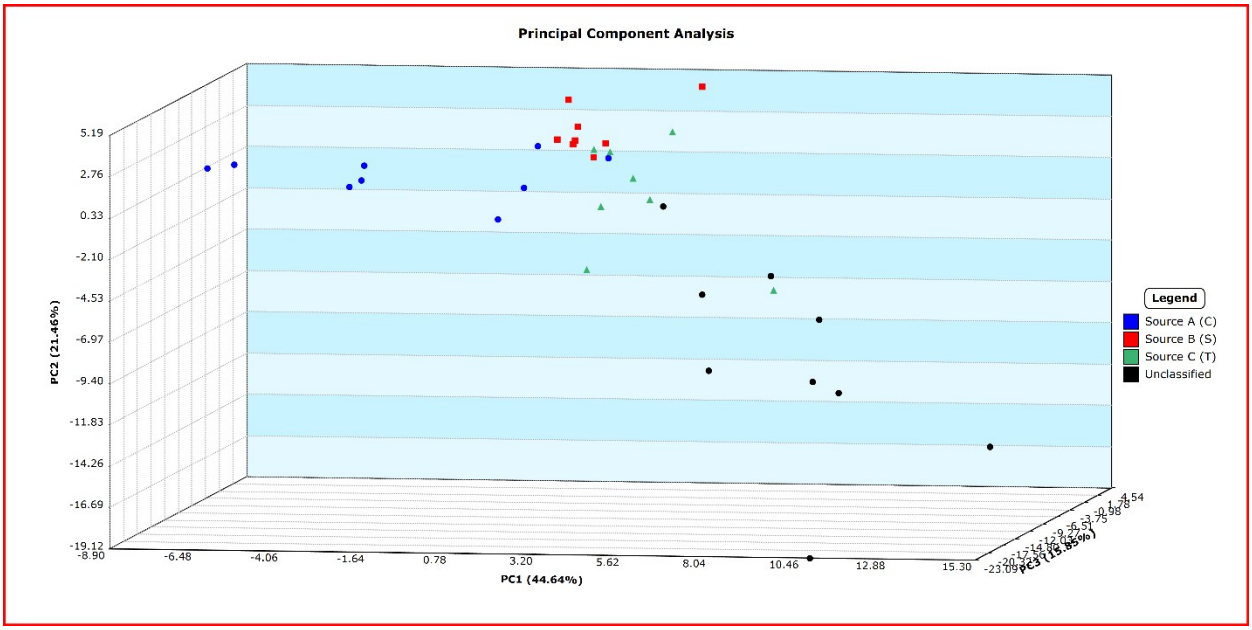

Figure S14: Round Robin 2018 Spill PCA

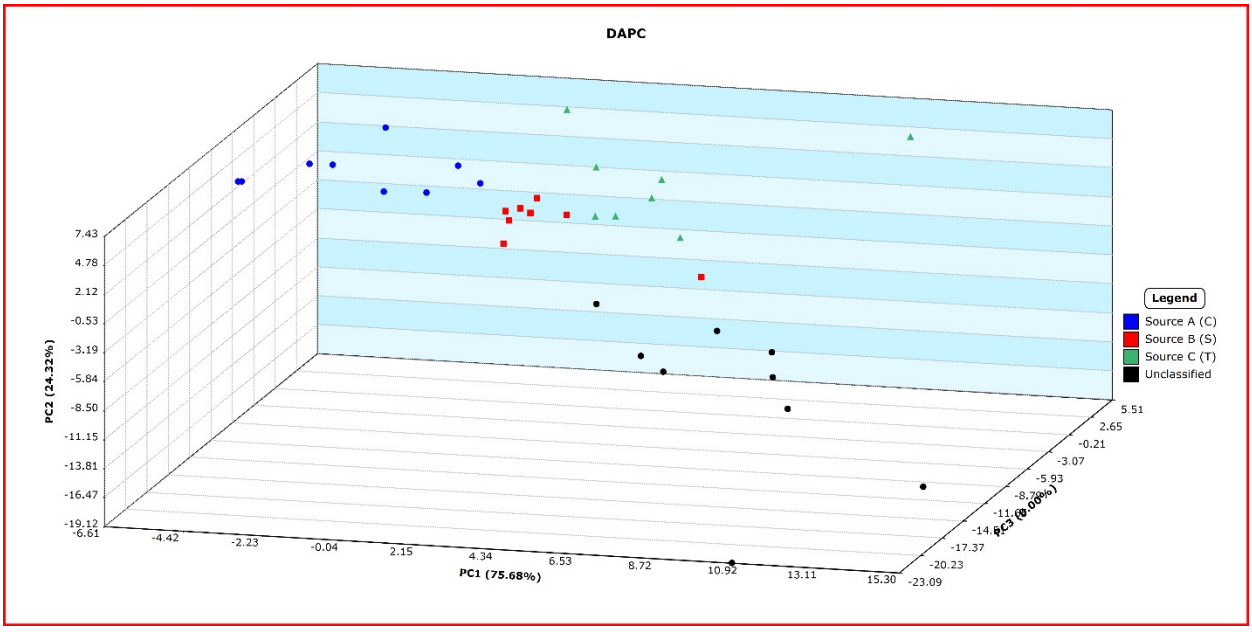

Figure S15: Round Robin 2018 Spill DAPC

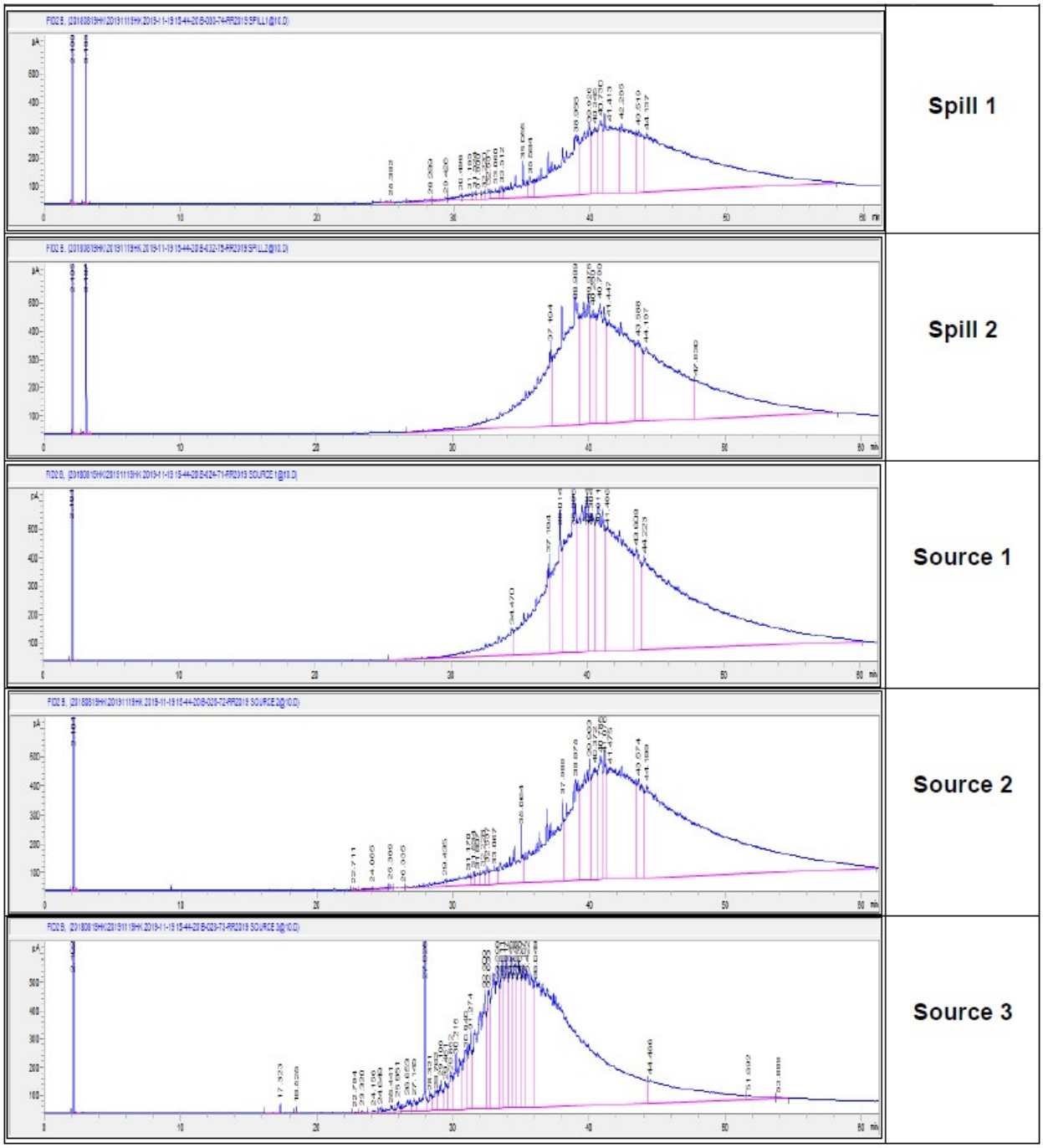

Figure S16: GC/FID chromatography for Round Robin 2019 samples

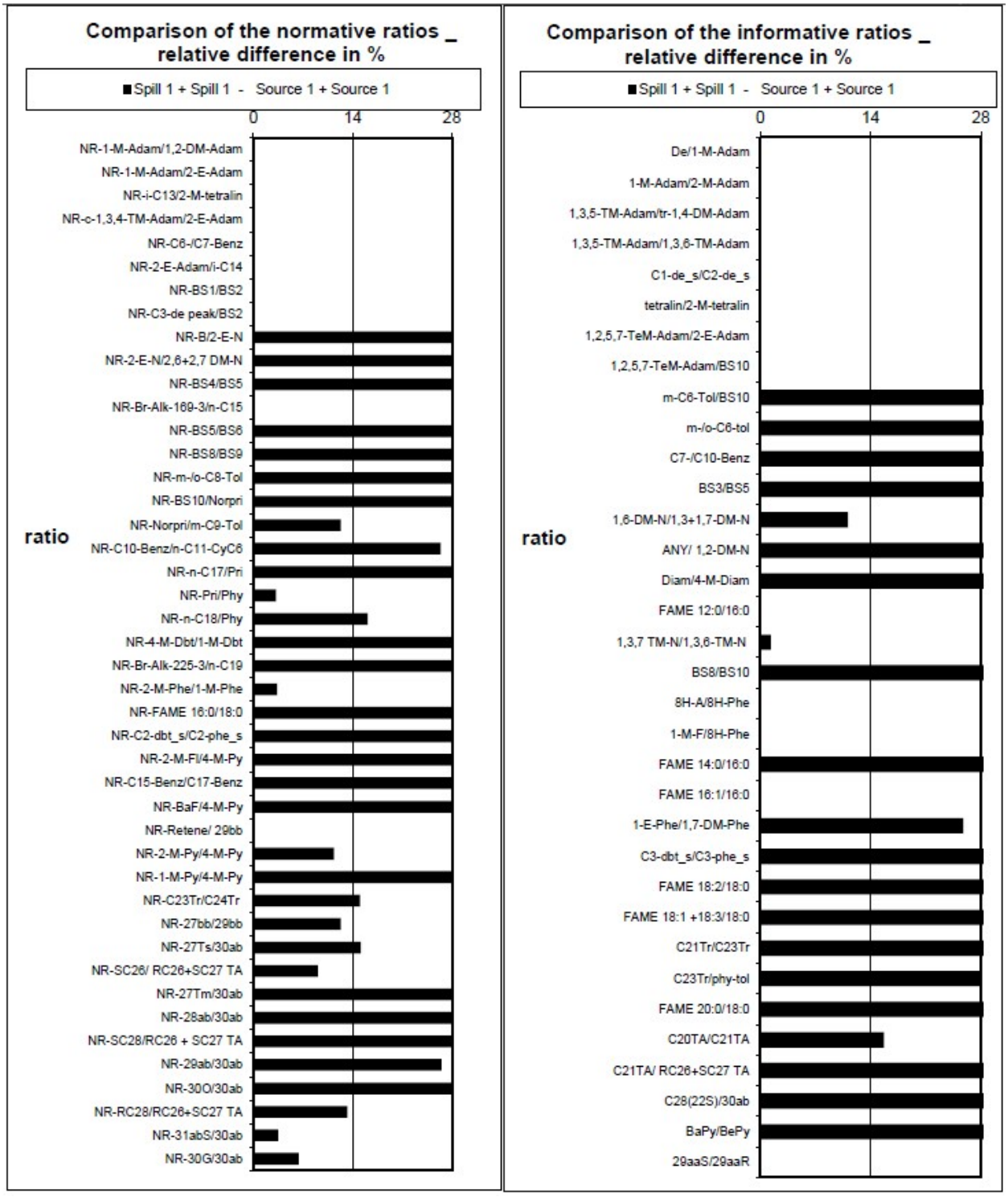

Figure S17: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source 1

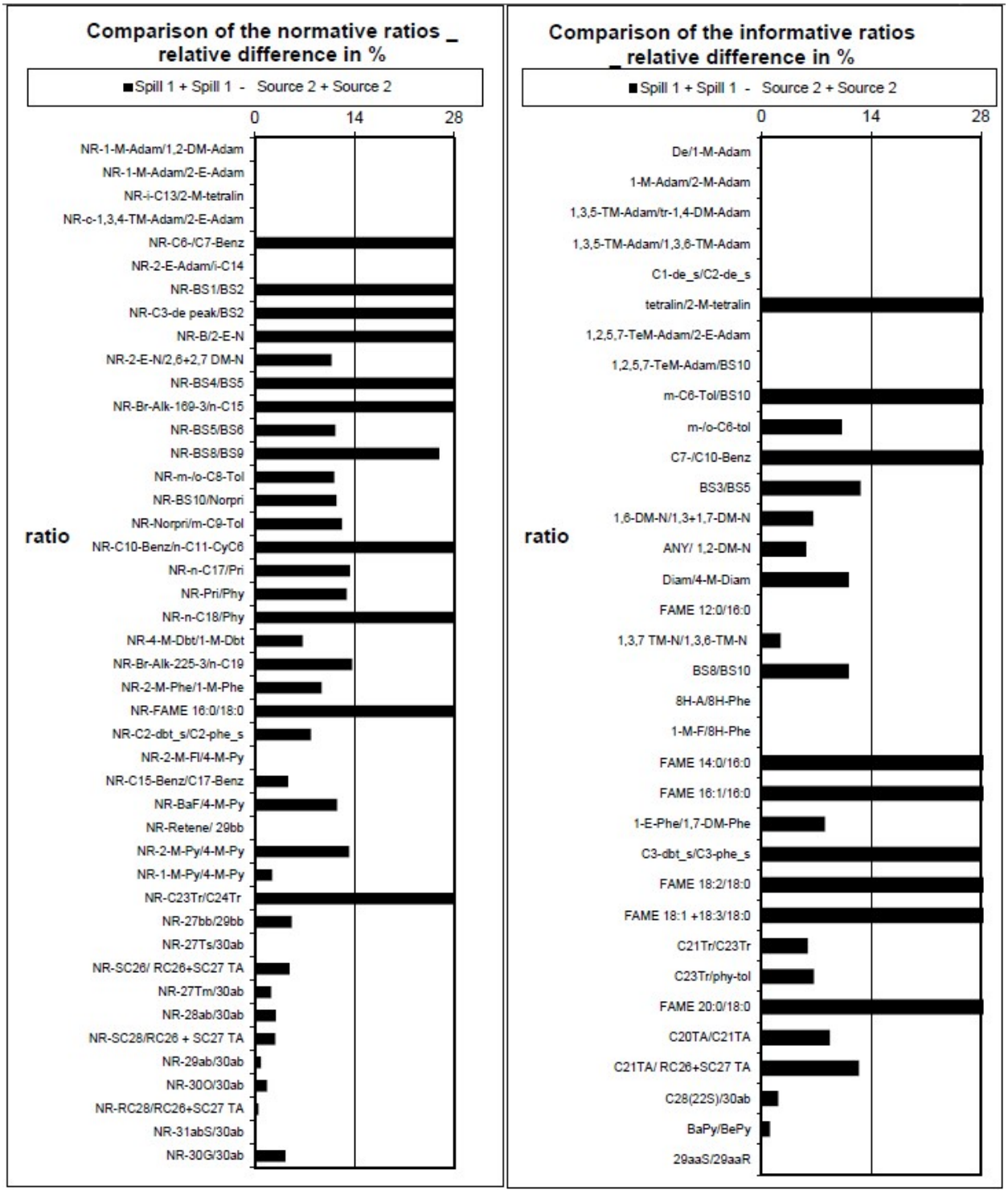

Figure S18: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source 2

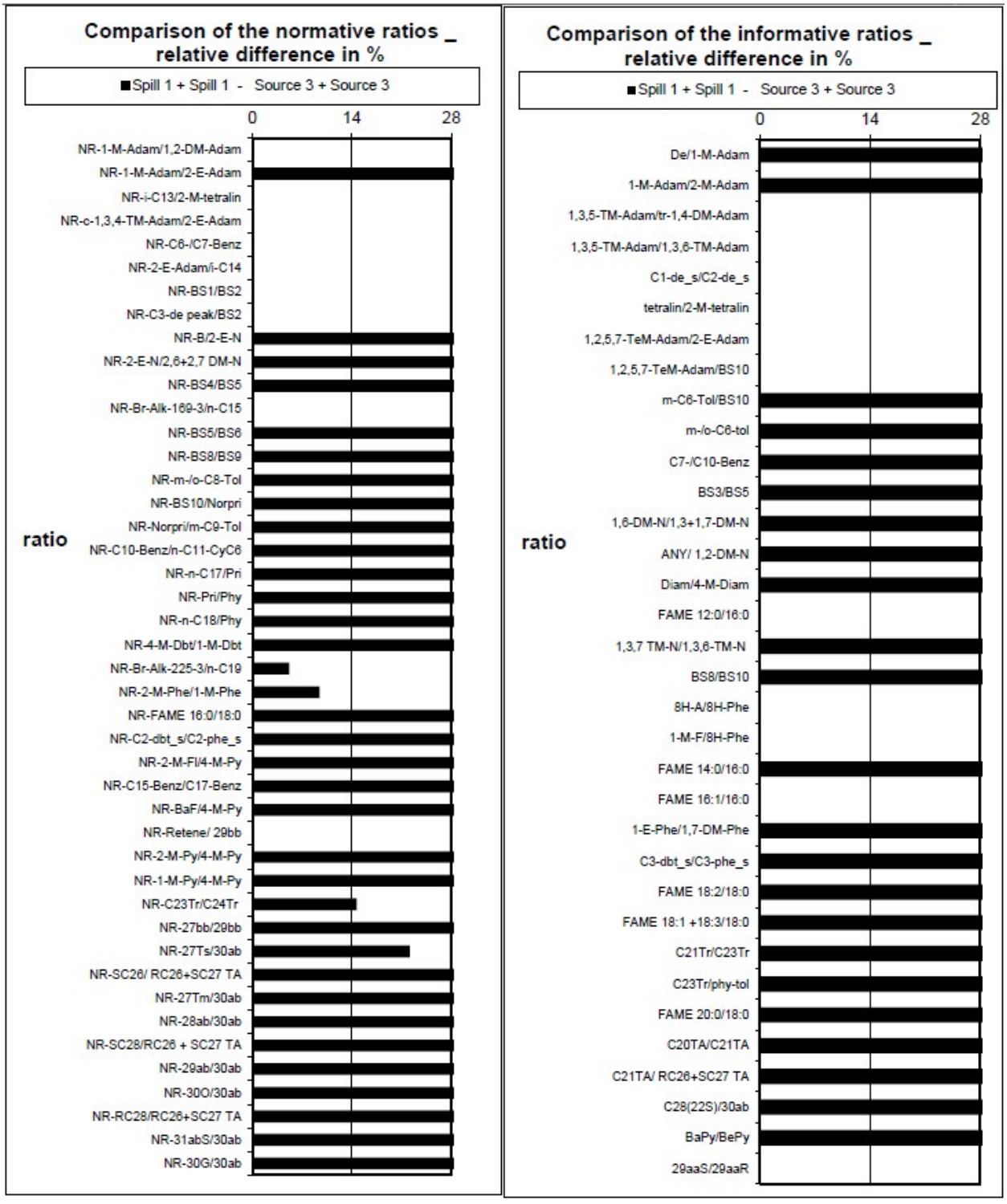

Figure S19: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source 3

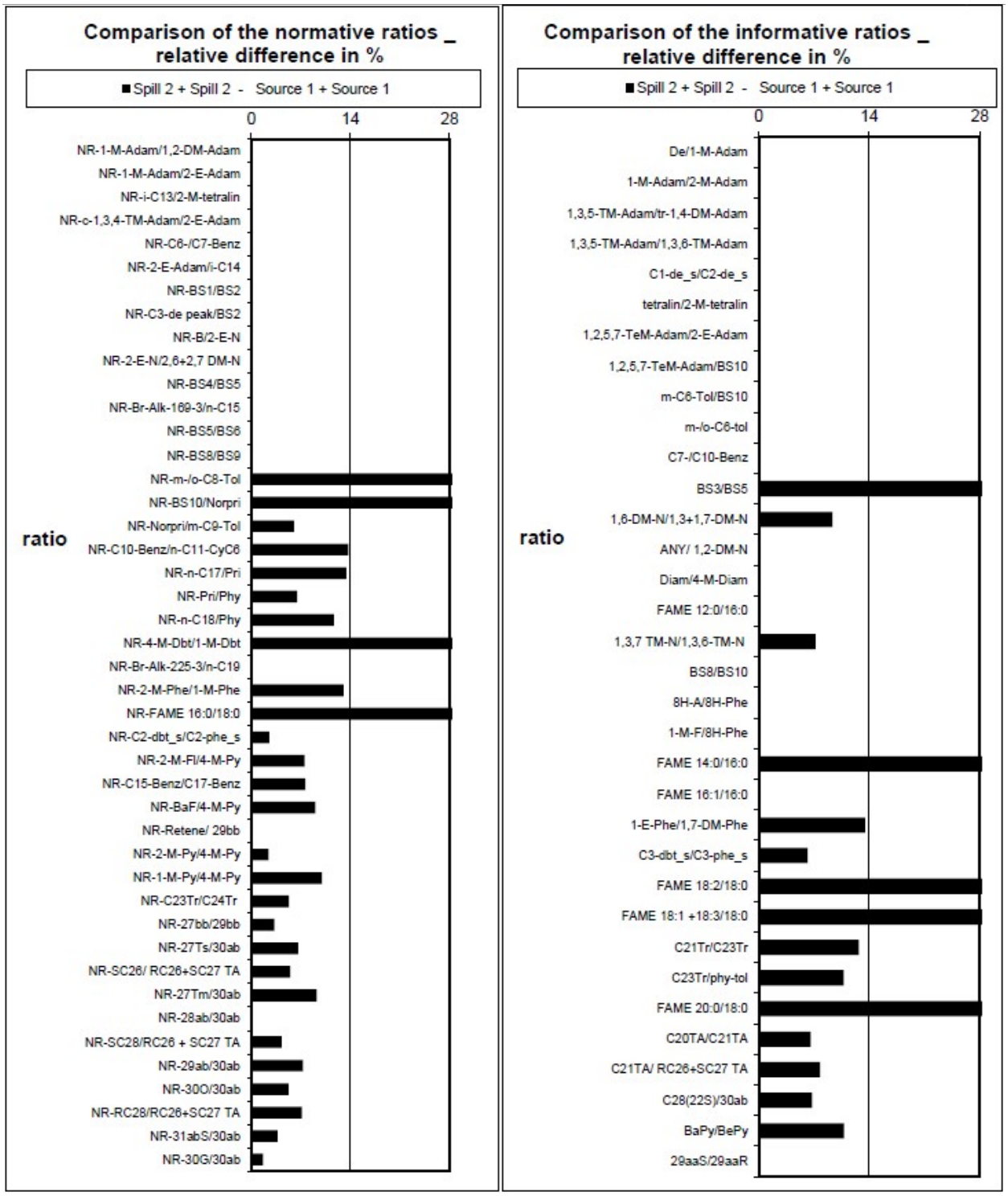

Figure S20: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source 1

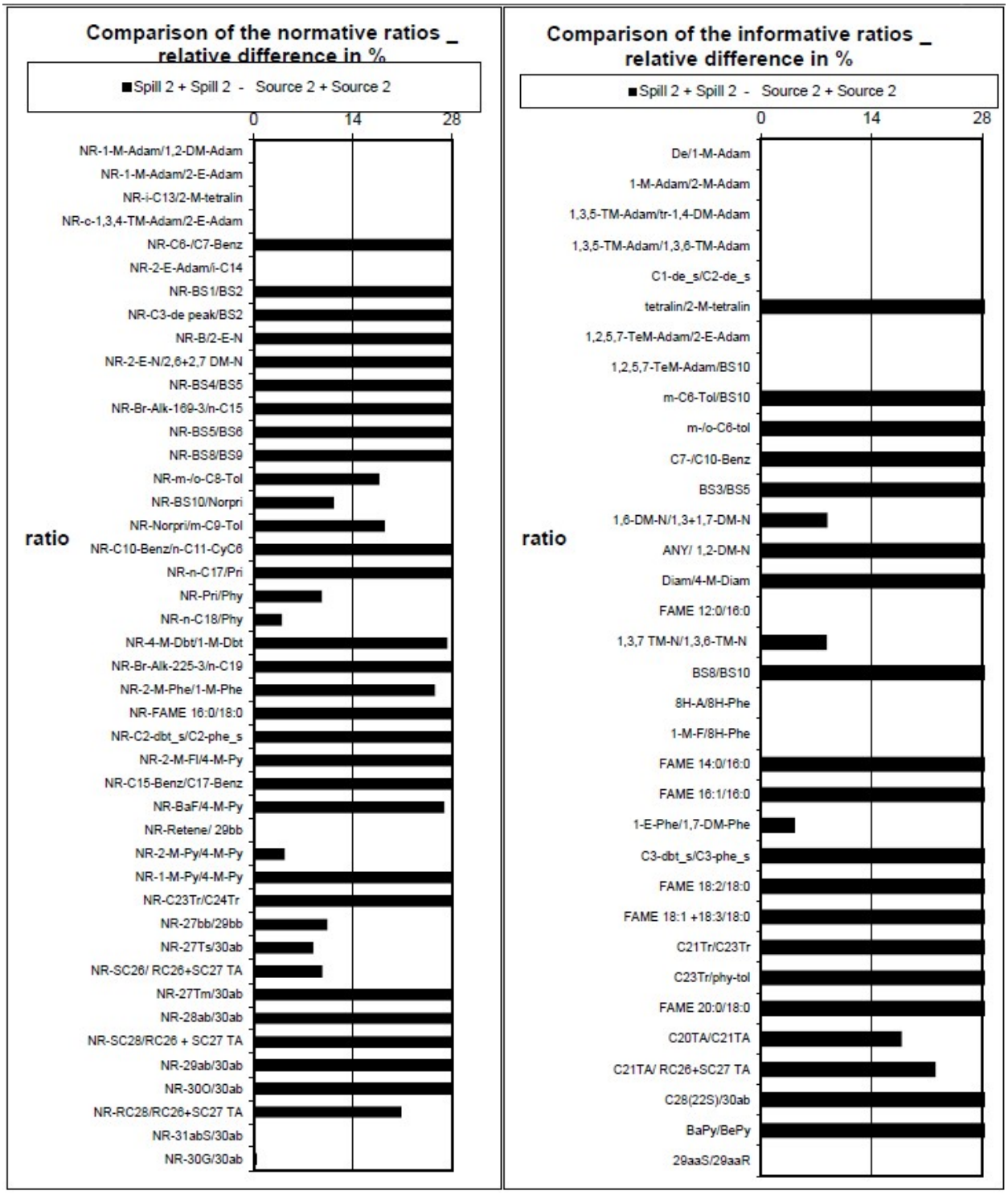

Figure S21: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source 2

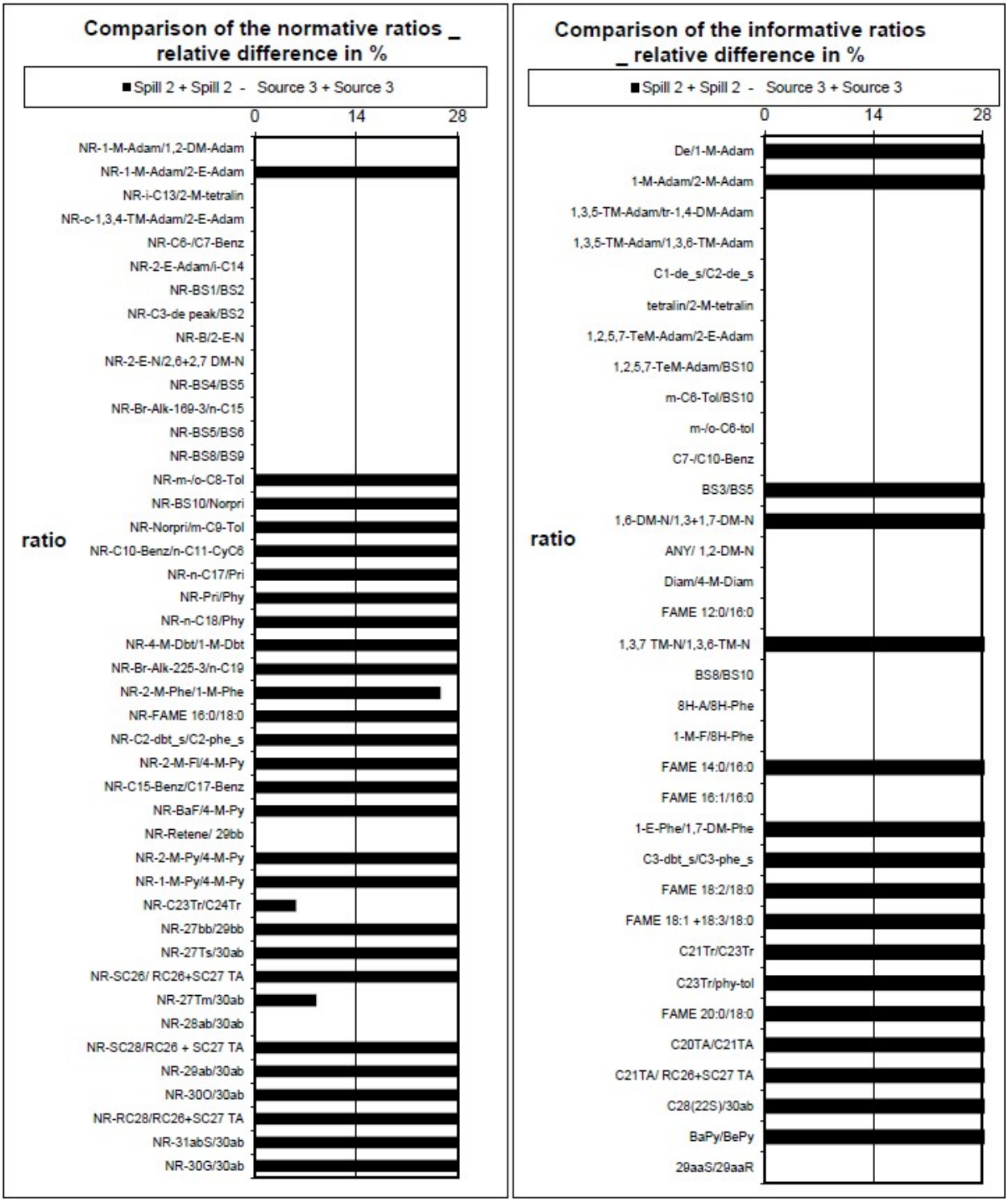

Figure S22: Round Robin 2019 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source 3

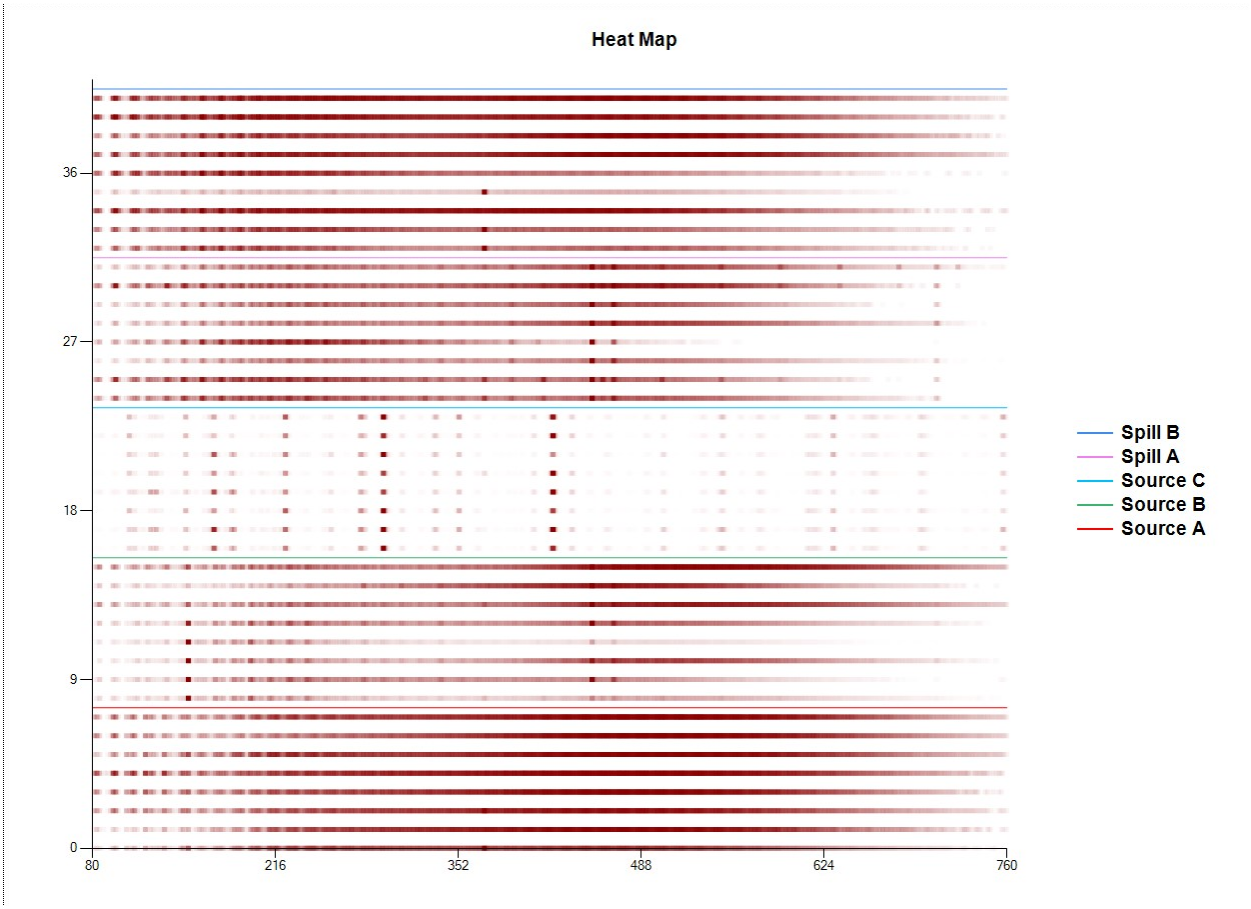

Figure S23: Round Robin 2019 Heatmap

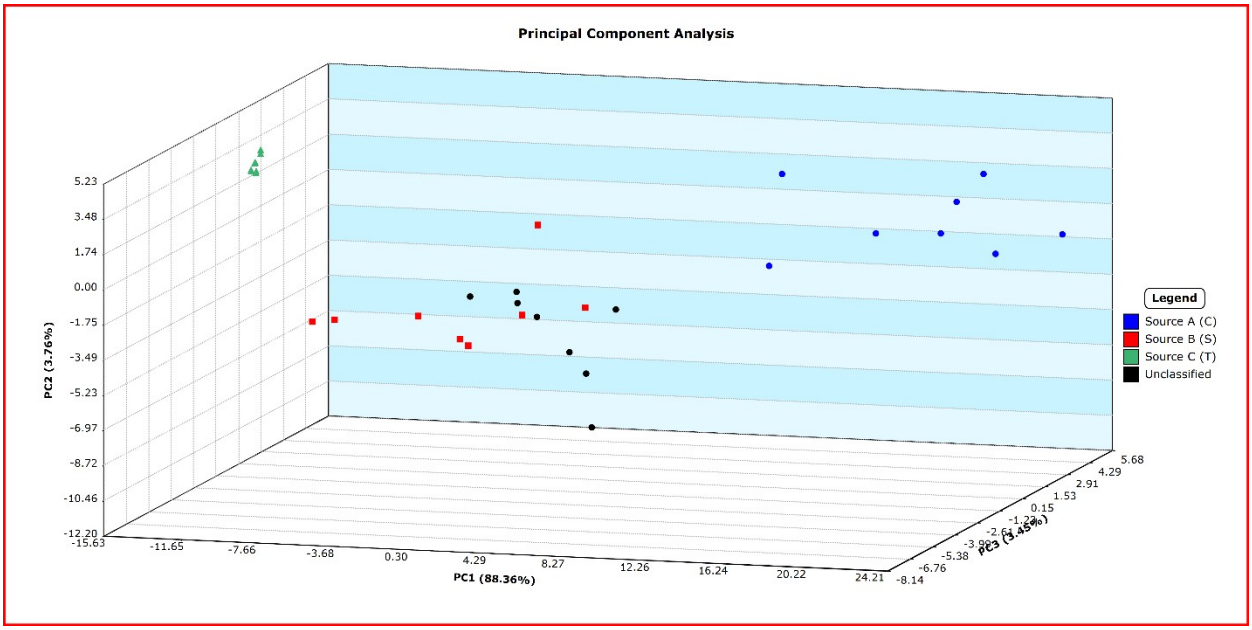

Figure S24: Round Robin 2019 Spill 1 PCA

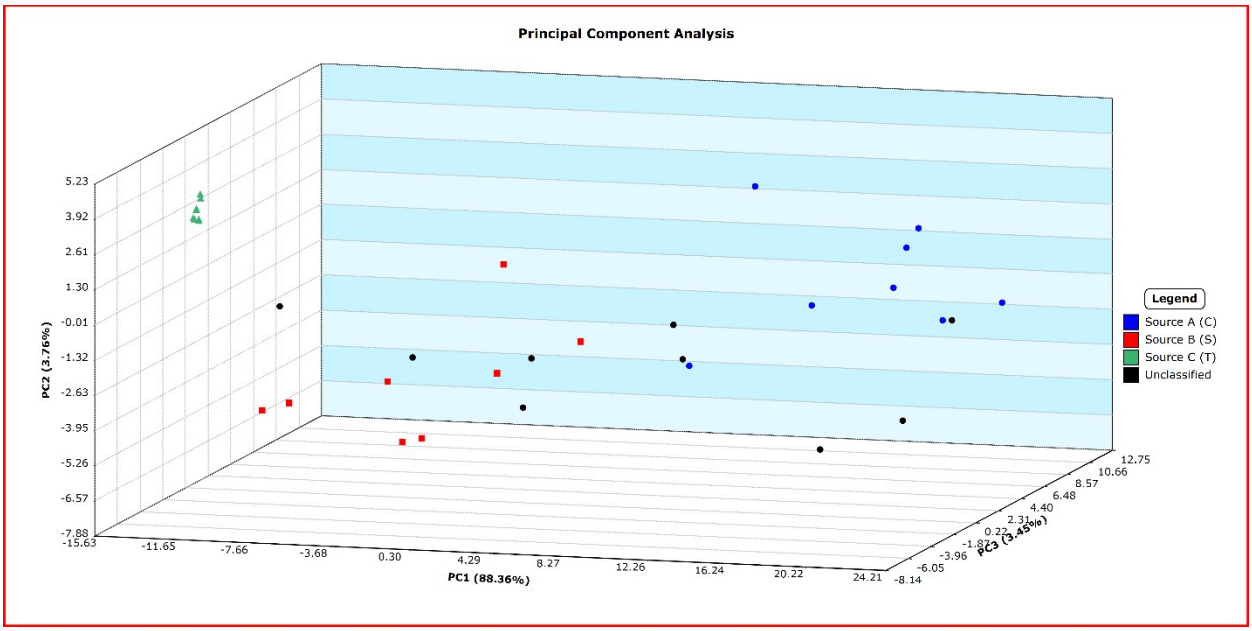

Figure S25: Round Robin 2019 Spill 2 PCA

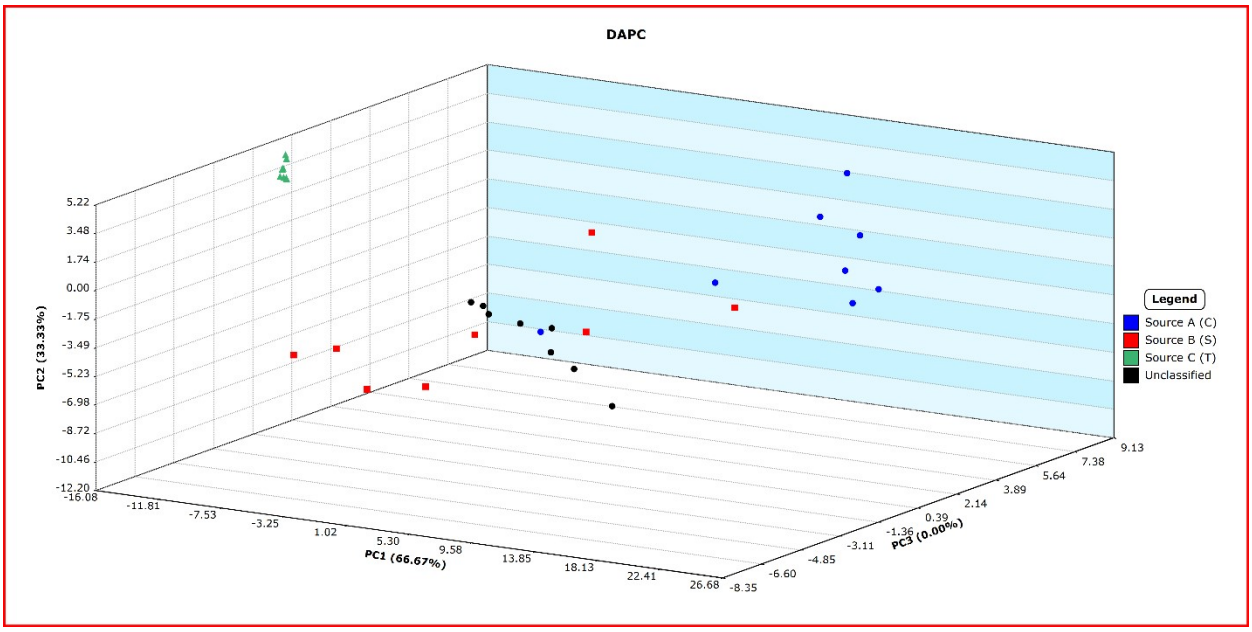

Figure S26: Round Robin 2019 Spill 1 DAPC

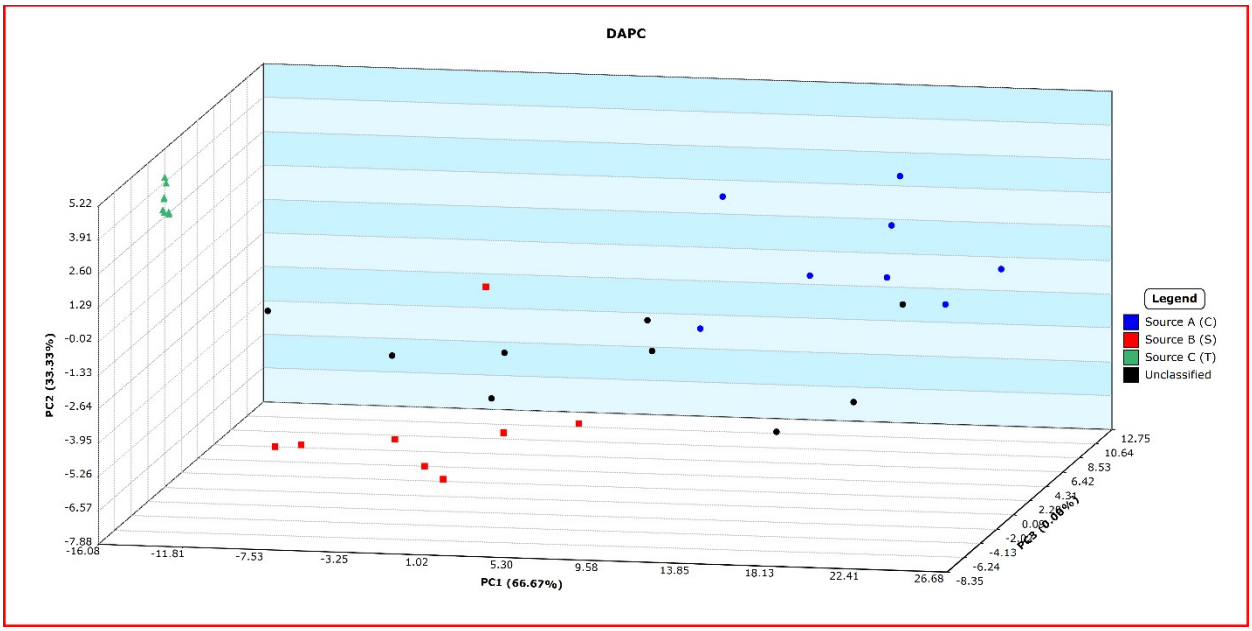

Figure S27: Round Robin 2019 Spill 2 DAPC

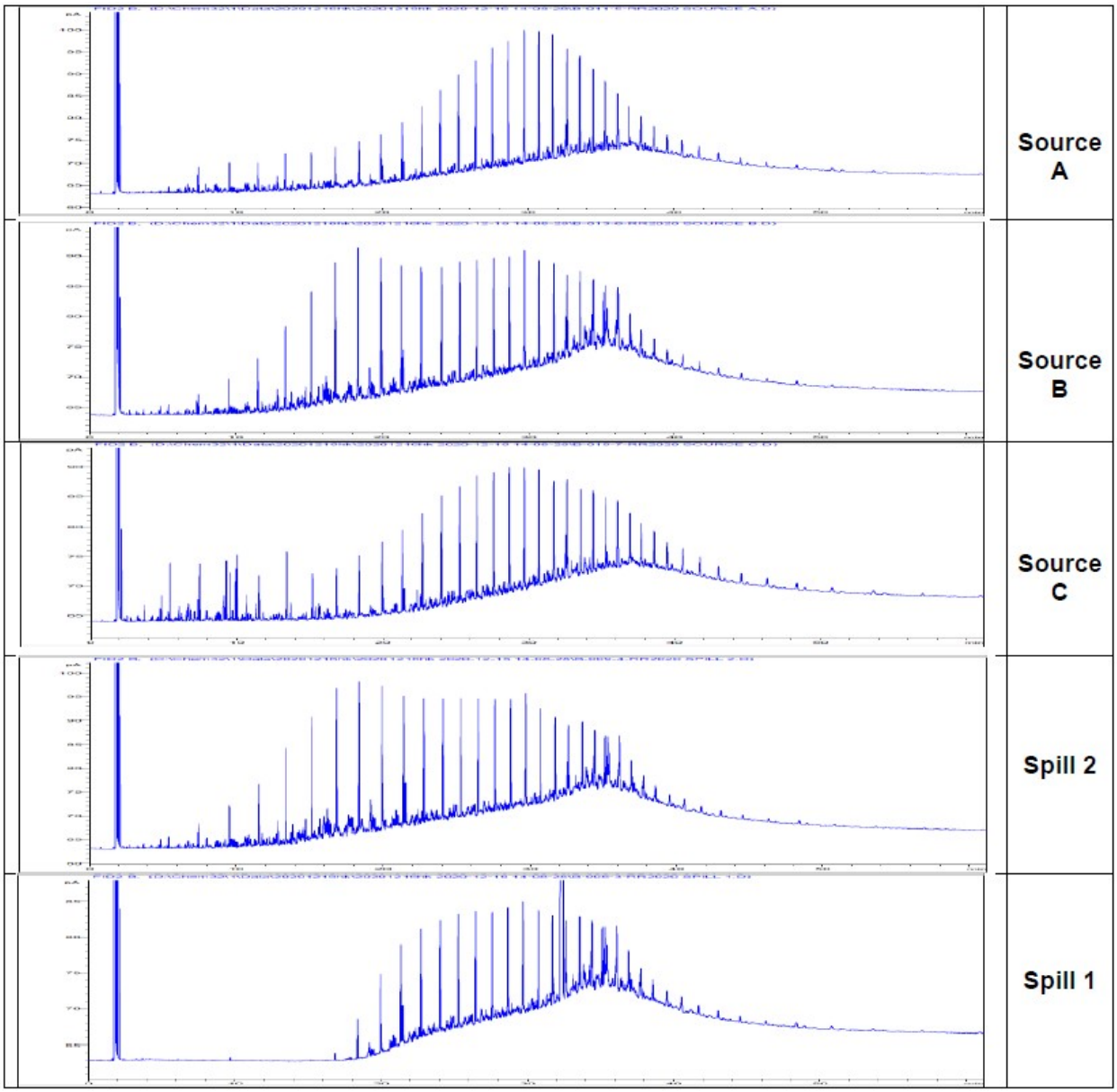

Figure S28: GC/FID chromatography for Round Robin 2020

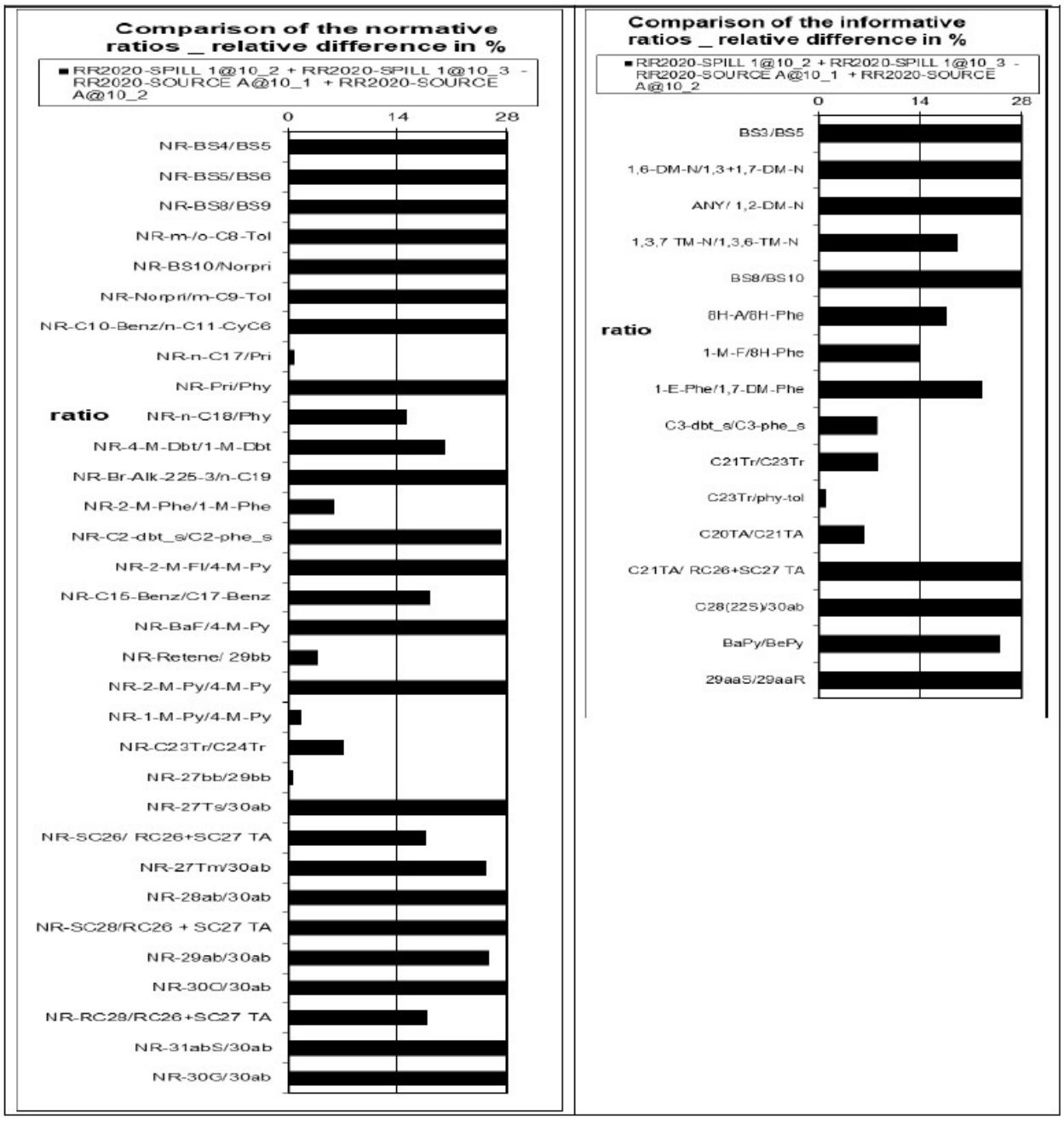

Figure S29: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 1 with Source A

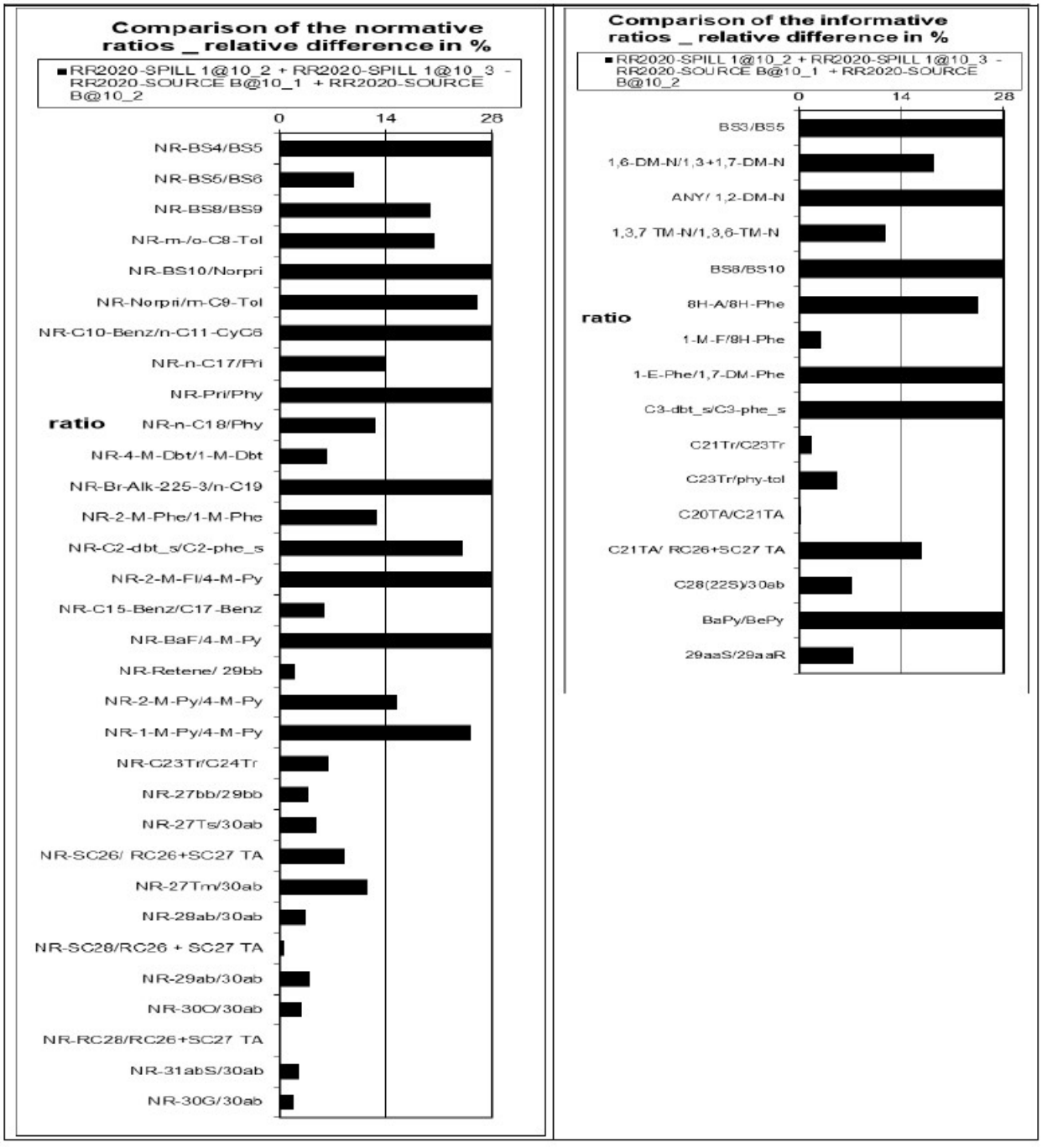

Figure S30: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 1 with Source B

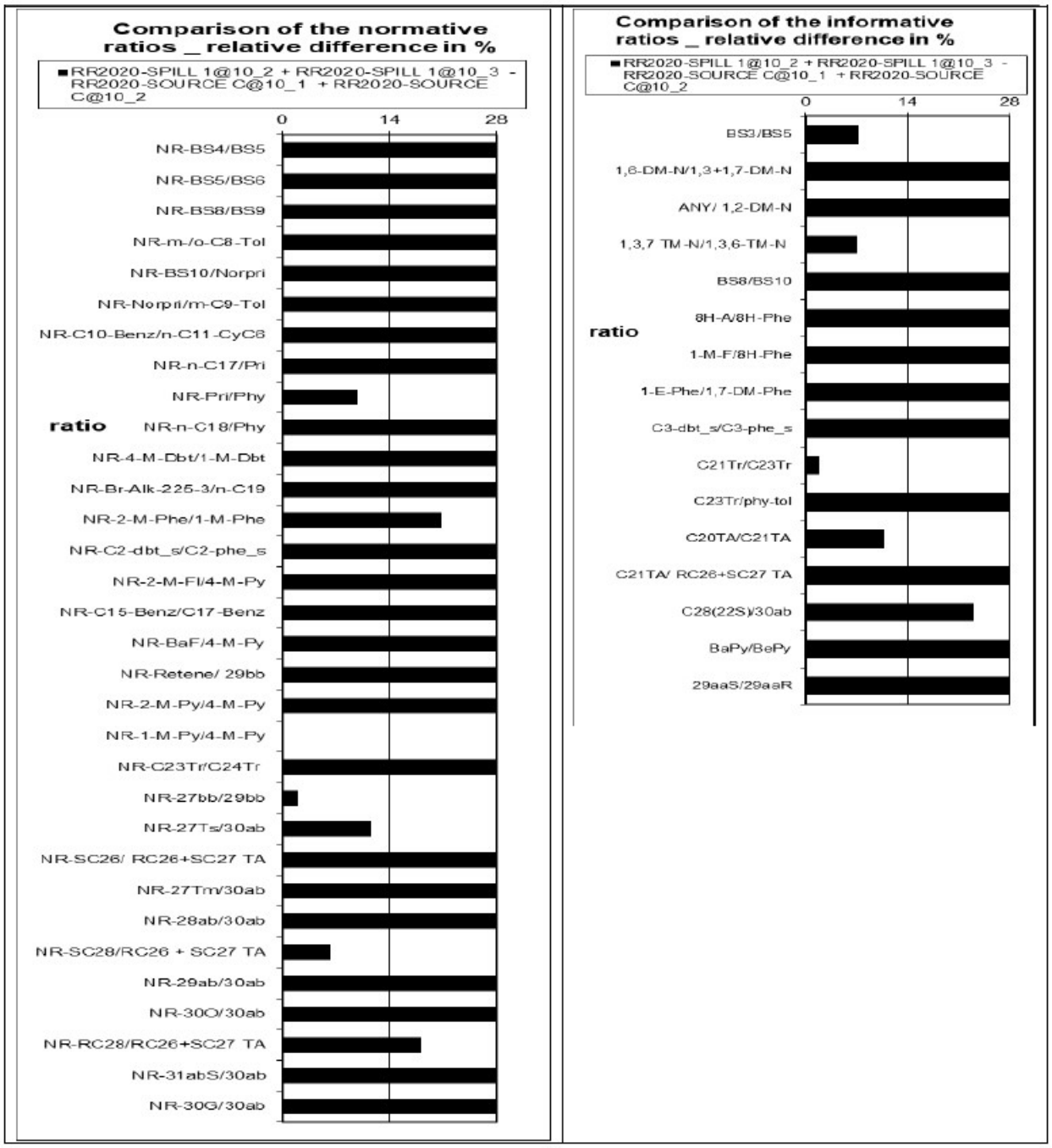

Figure S31: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 1 with Source C

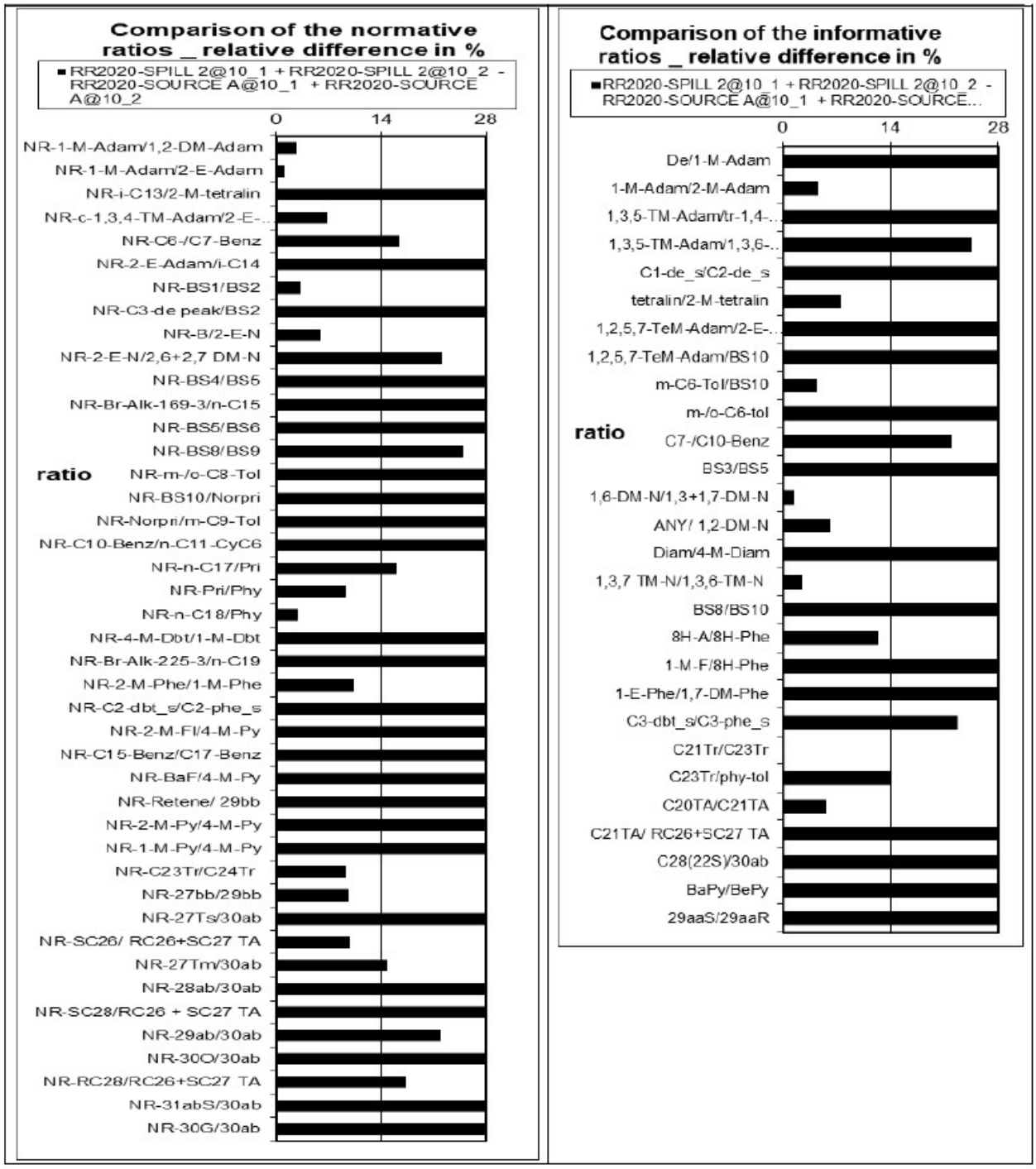

Figure S32: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 2 with Source A

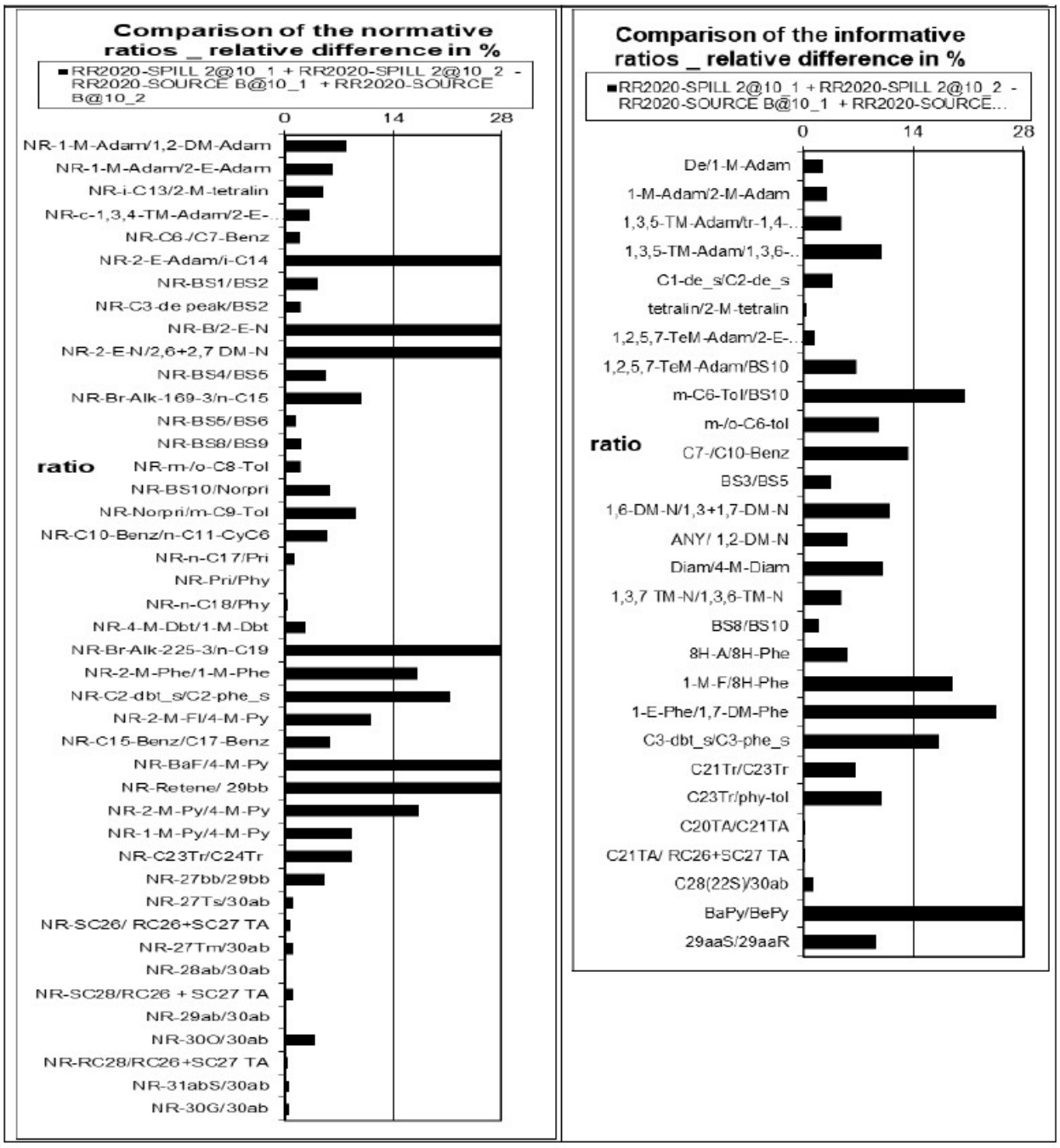

Figure S33: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 2 with Source B

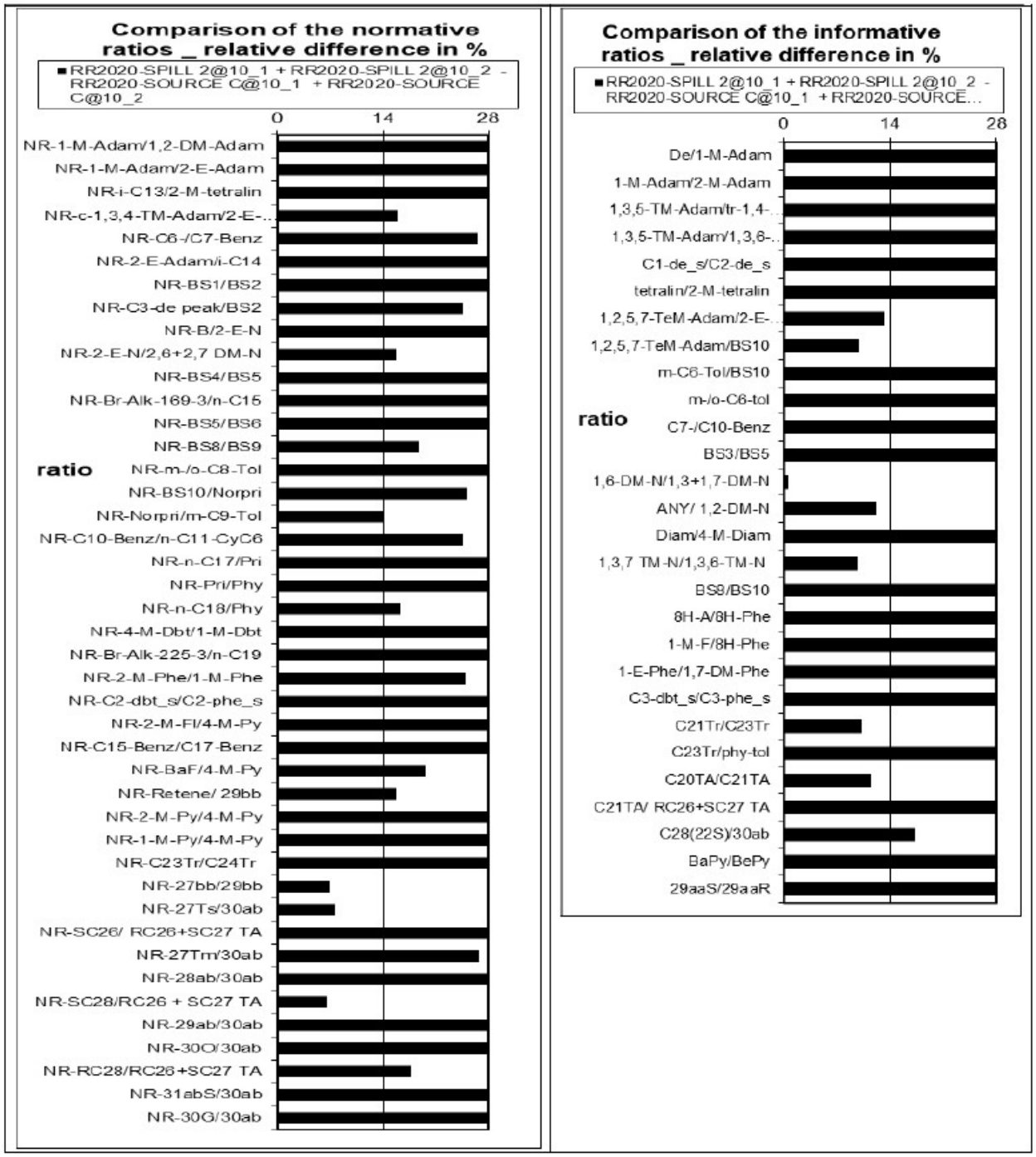

Figure S34: Round Robin 2020 GC/MS Comparison of Diagnostic ratios of Spill 2 with Source C

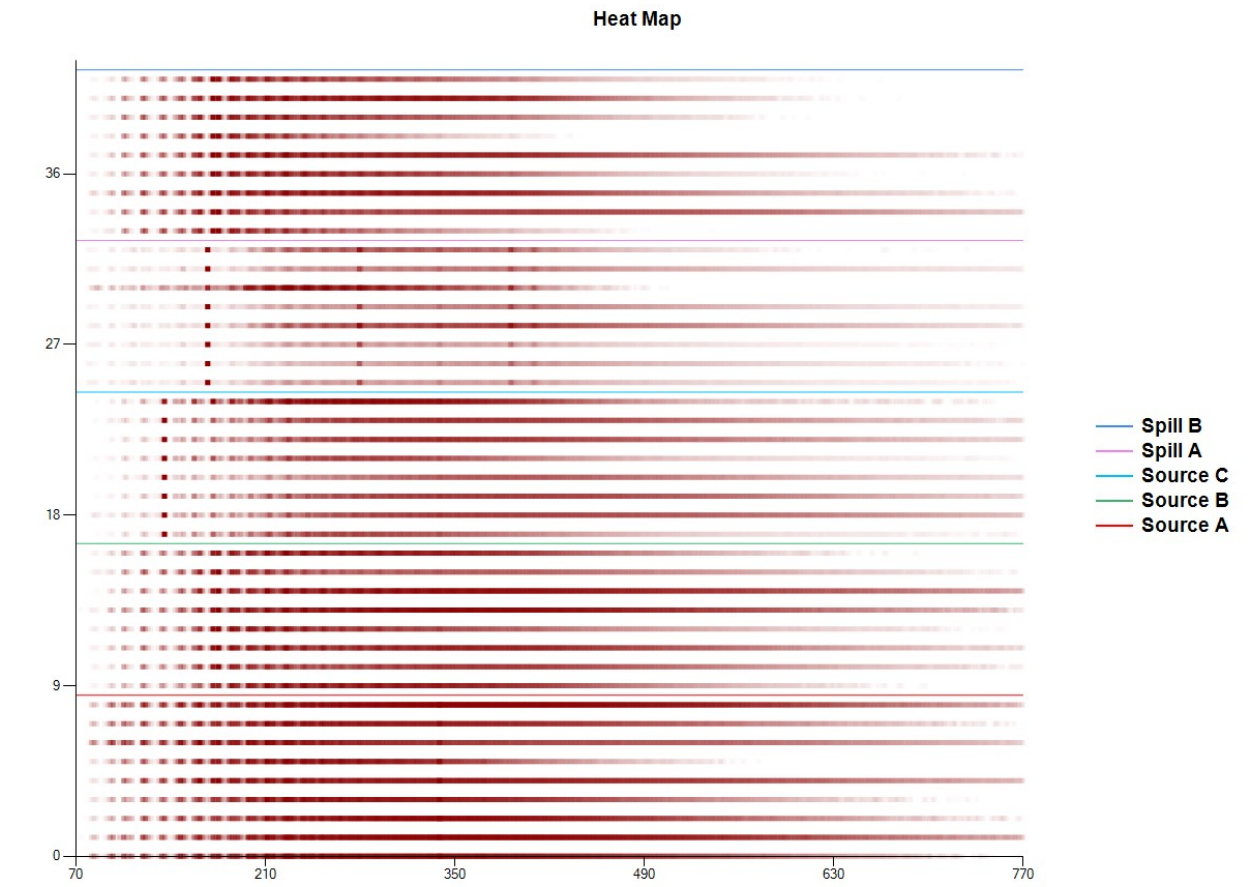

Figure S35: Round Robin 2020 Heatmap

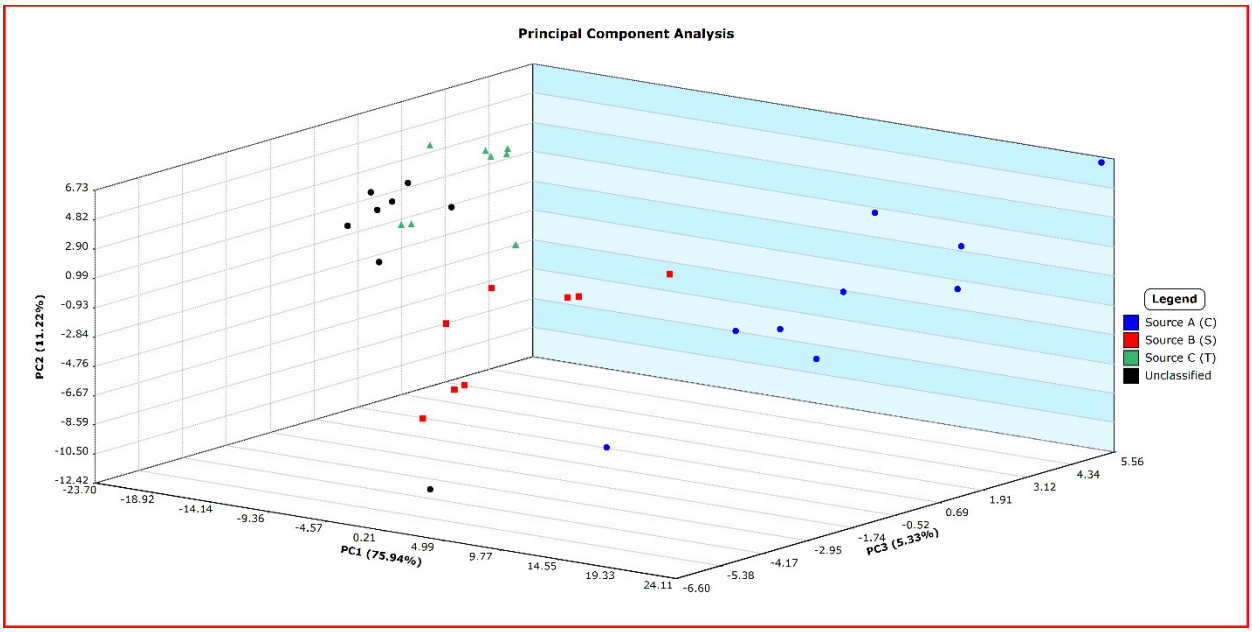

Figure S36: Round Robin 2020 Spill 1 PCA

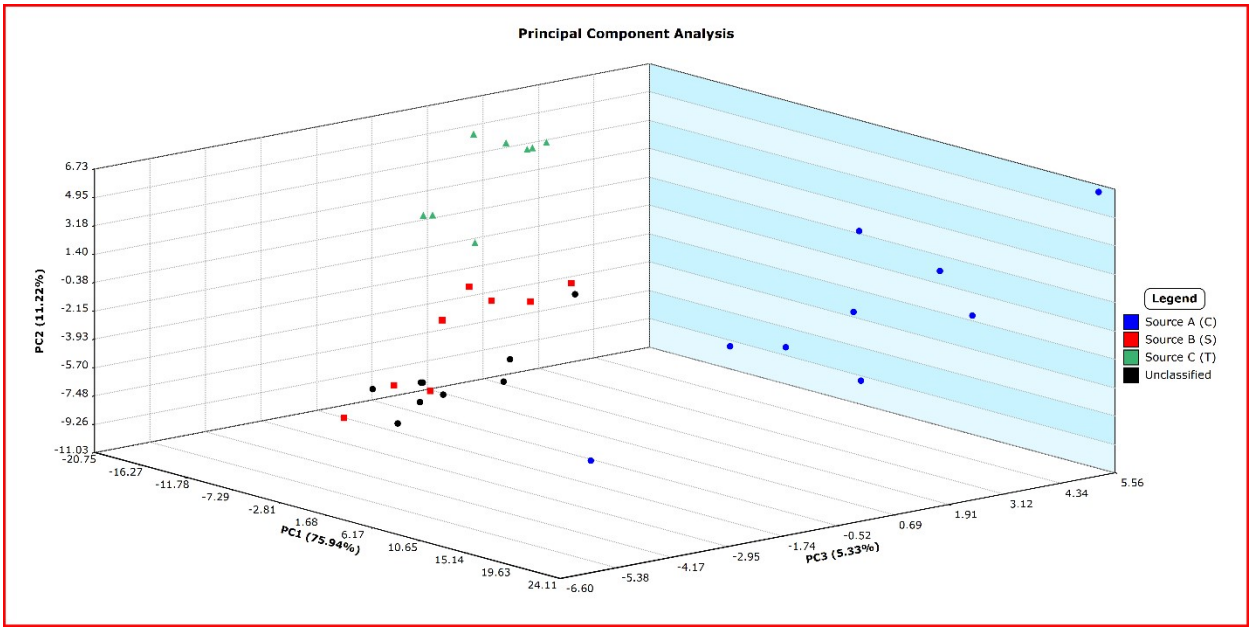

Figure S37: Round Robin 2020 Spill 2 PCA

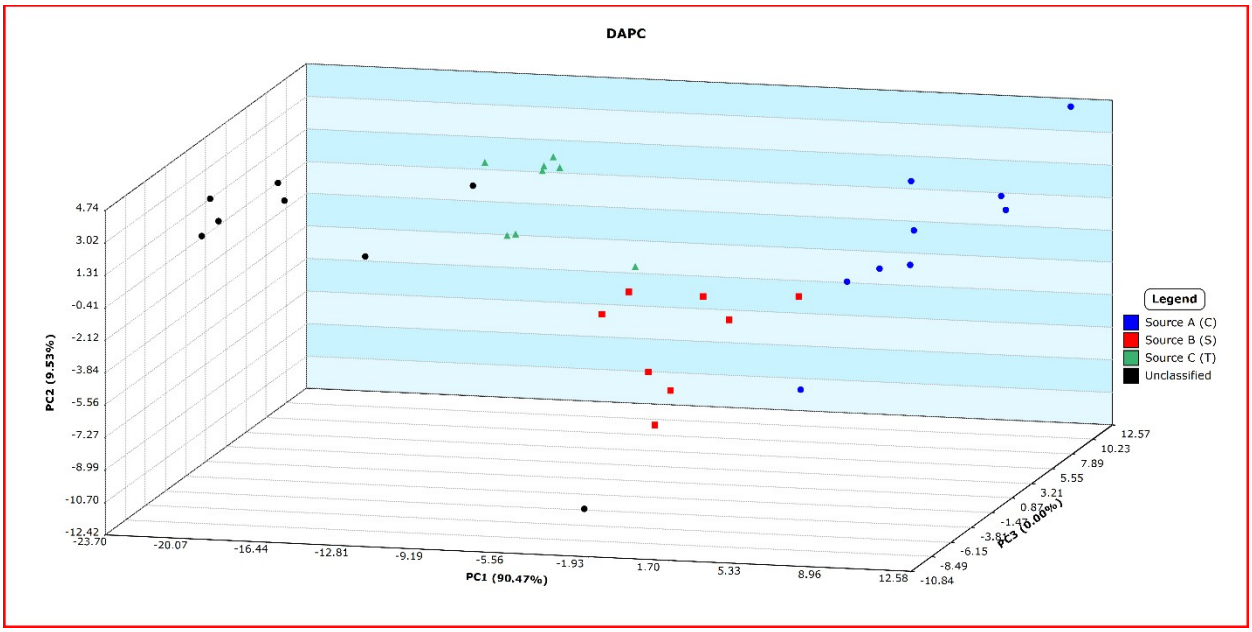

Figure S38: Round Robin 2020 Spill 1 DAPC

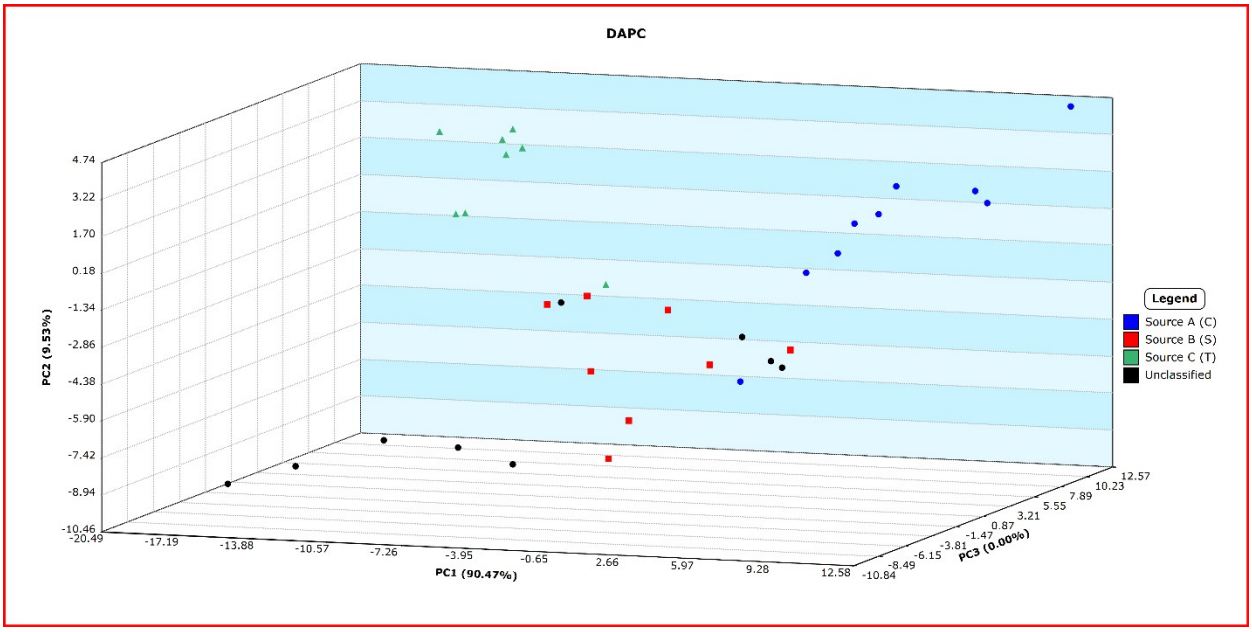

Figure S39: Round Robin 2020 Spill 2 DAPC

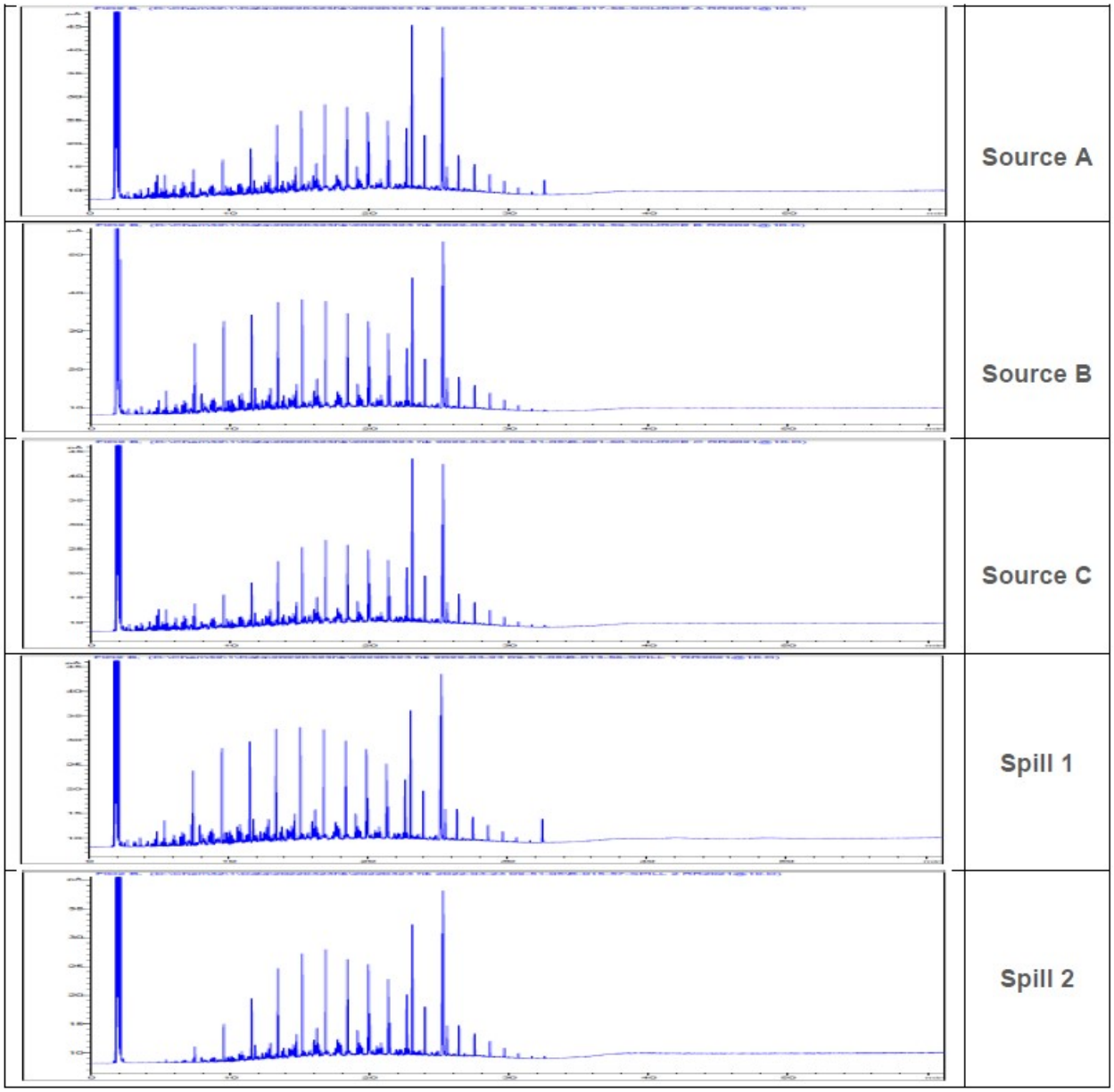

Figure S40: GC/FID chromatography for Round Robin 2021 samples

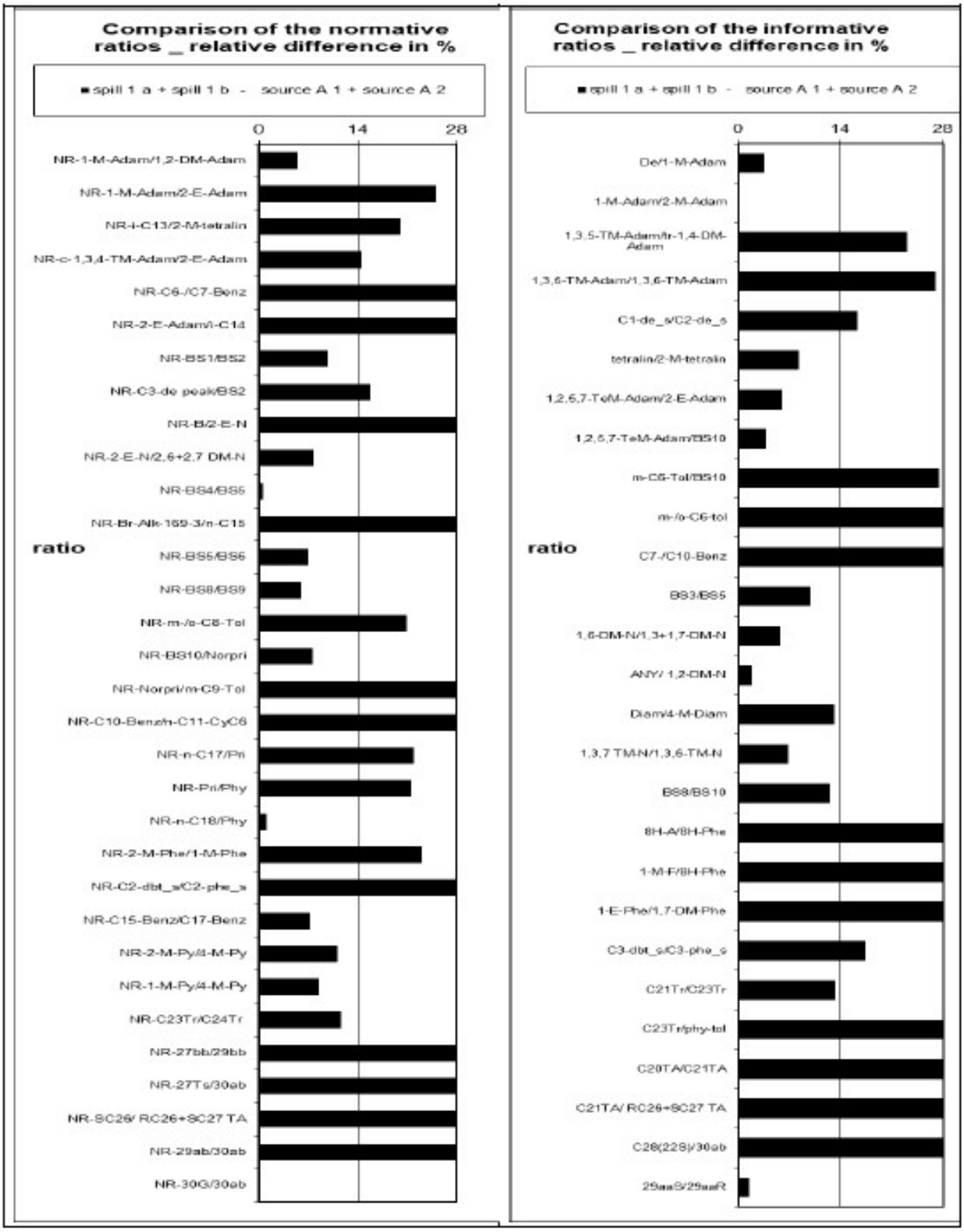

Figure S41: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source A

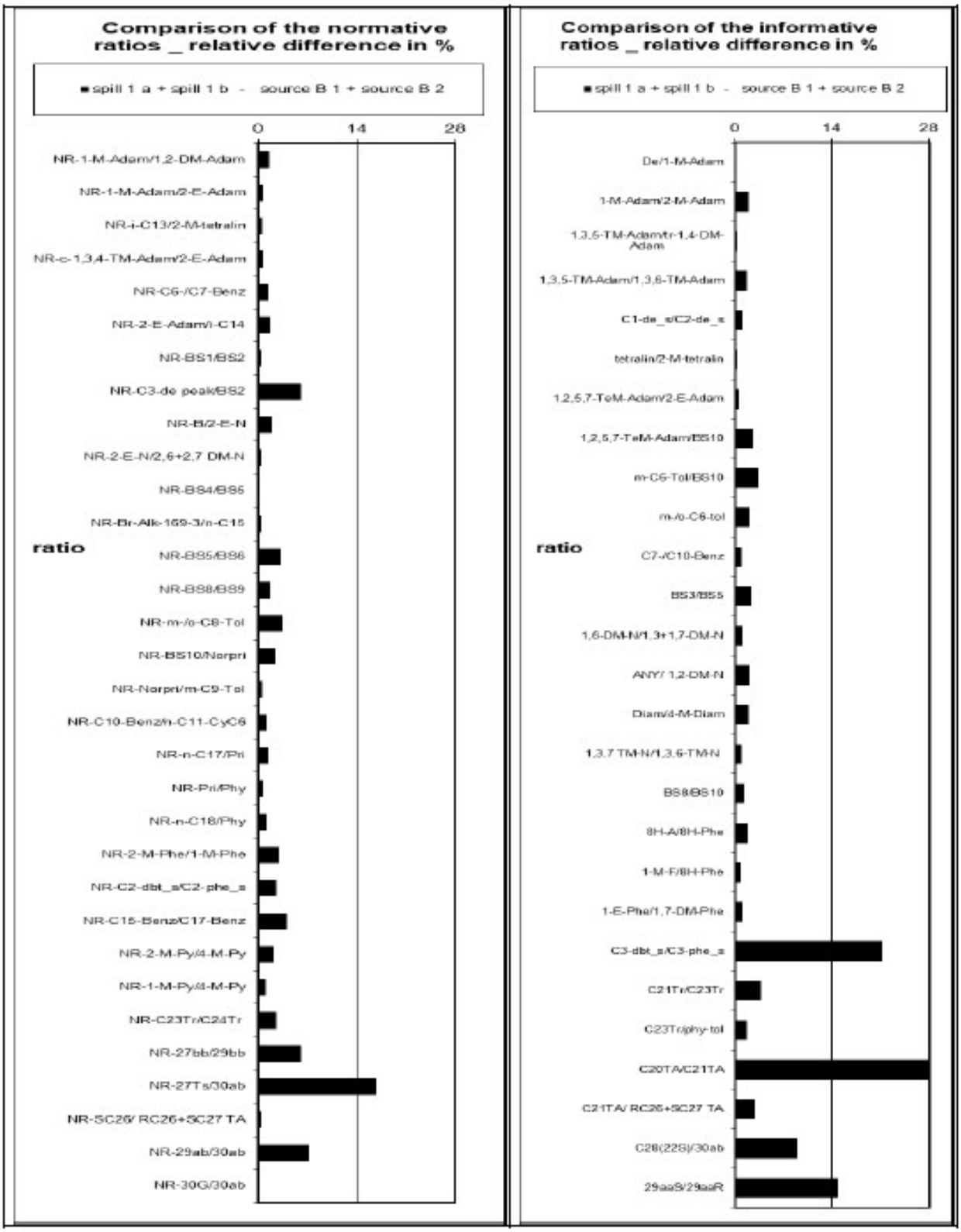

Figure S42: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source B

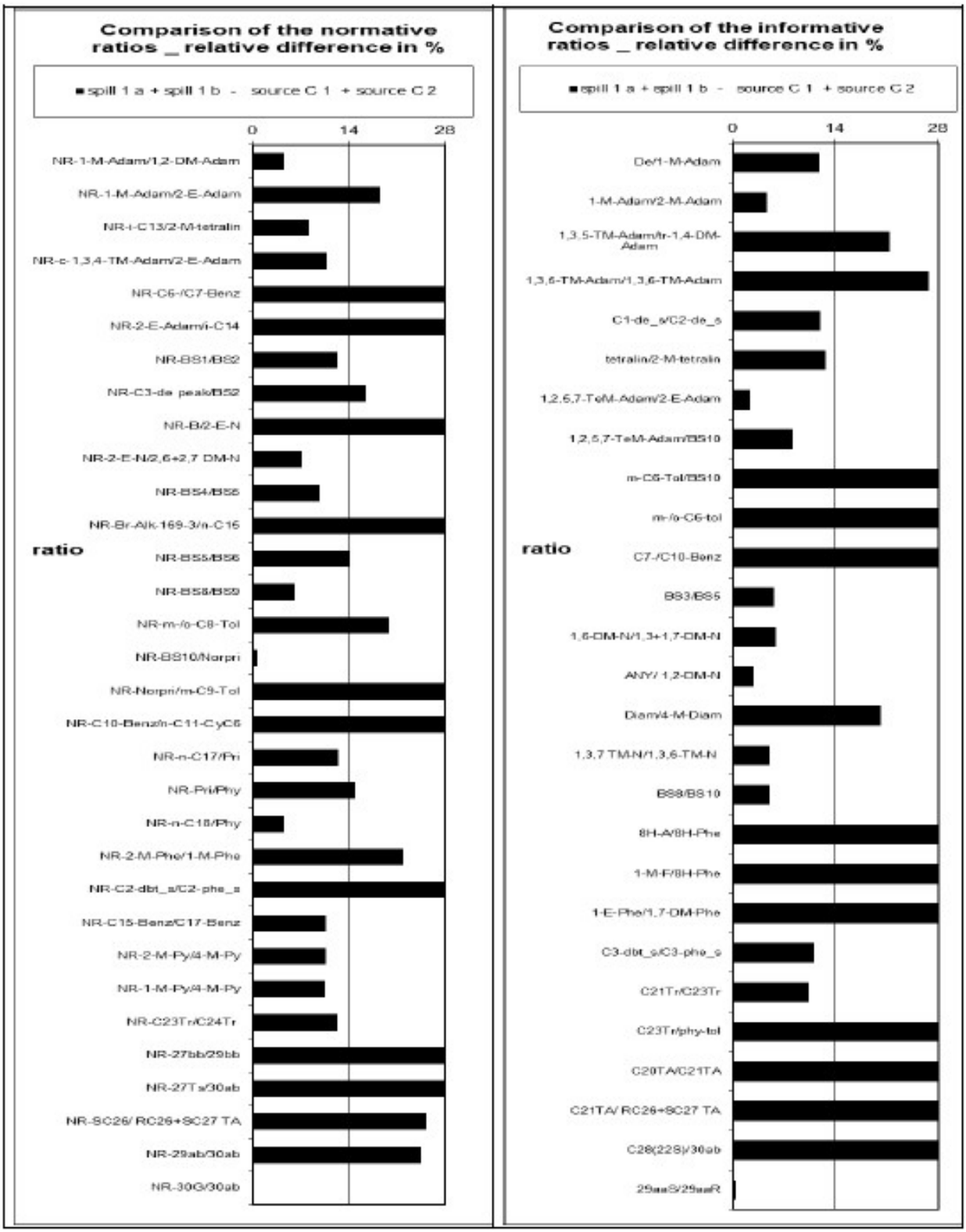

Figure S43: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 1 compared with Source C

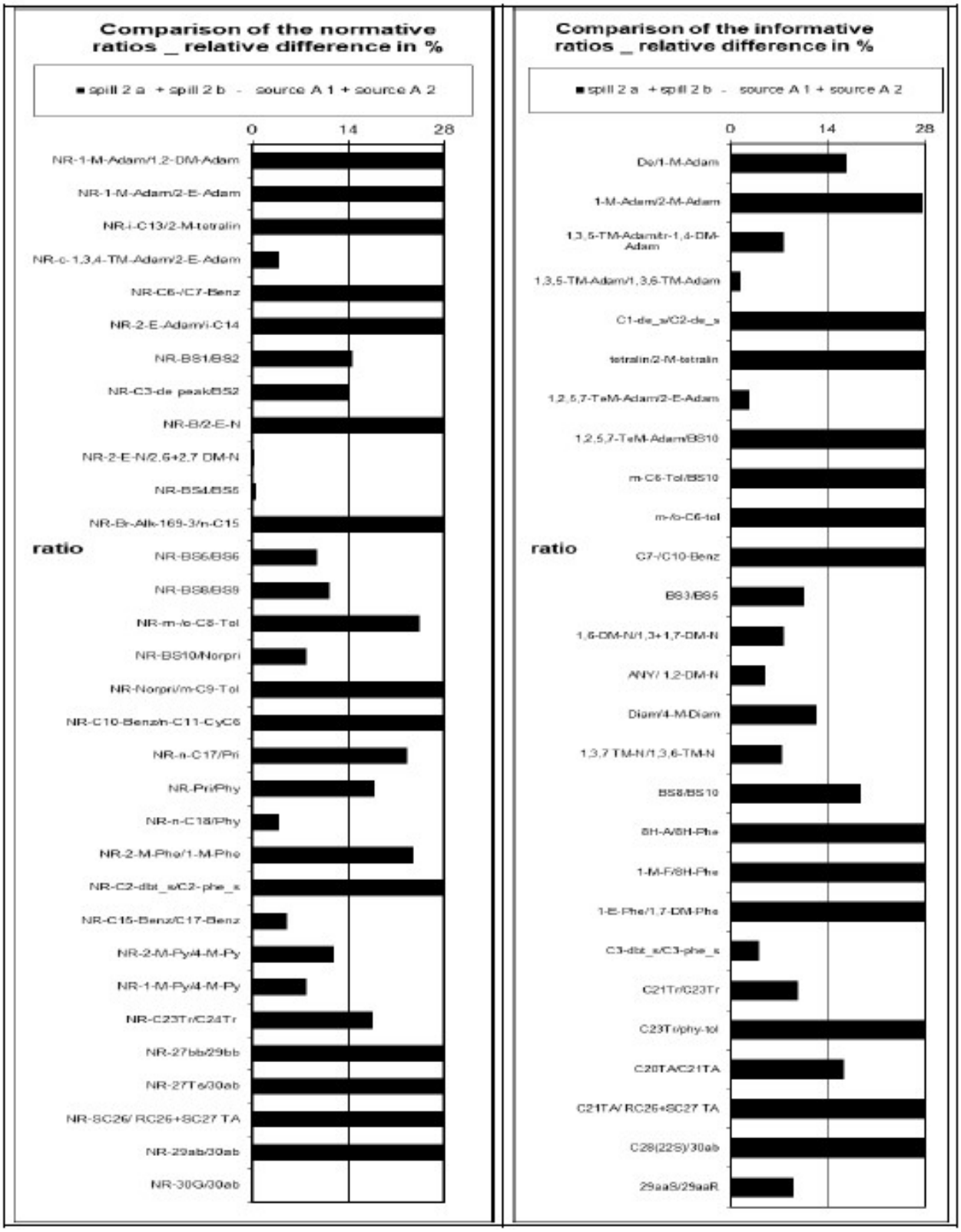

Figure S44: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source A

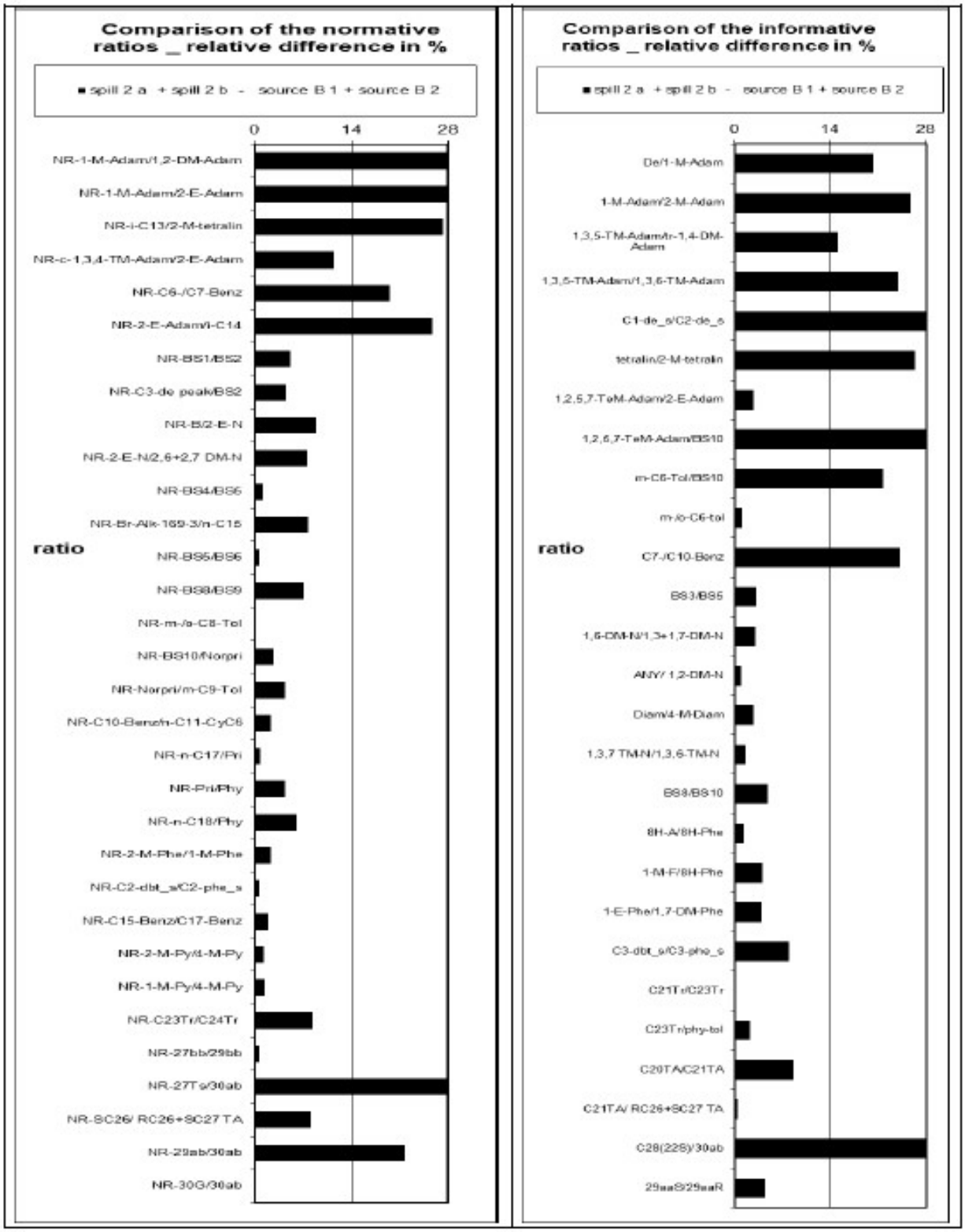

Figure S45: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source B

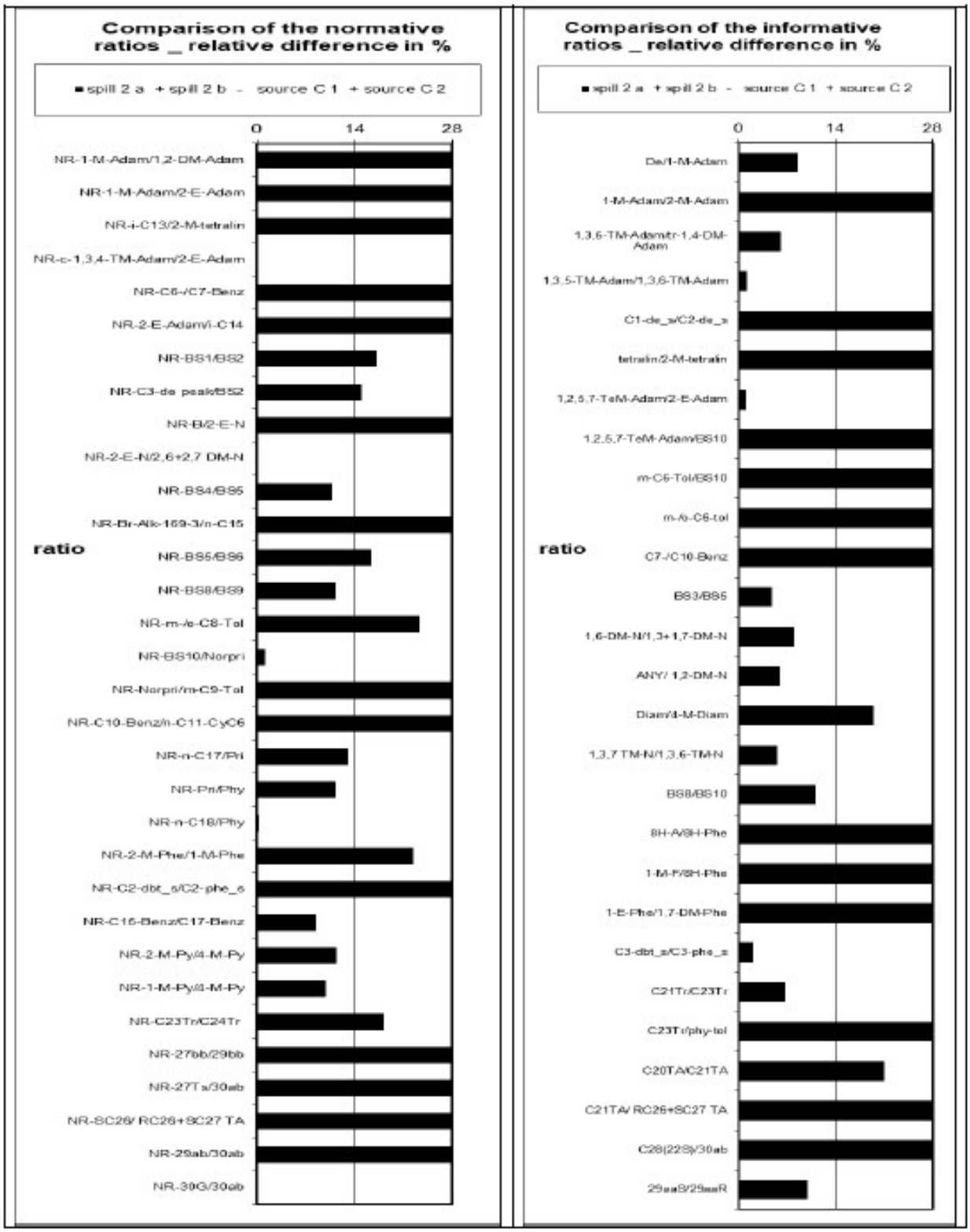

Figure S46: Round Robin 2021 GC/MS Comparison of Diagnostic ratios of Spill 2 compared with Source C

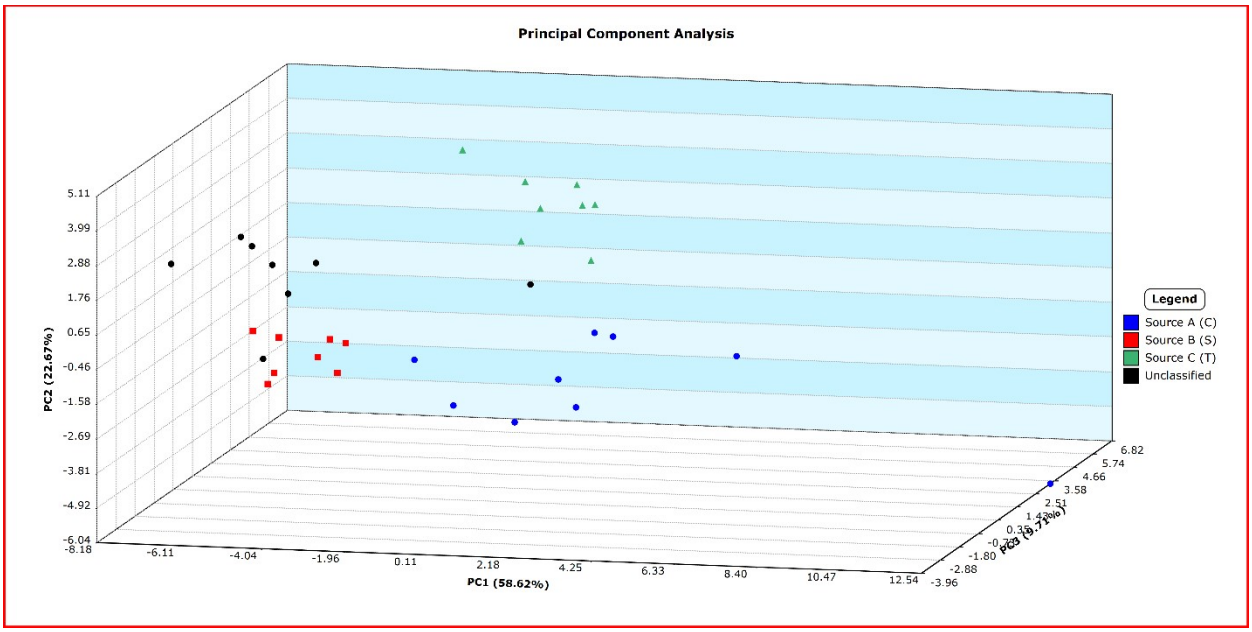

Figure S47: Round Robin 2021 Spill 1 PCA

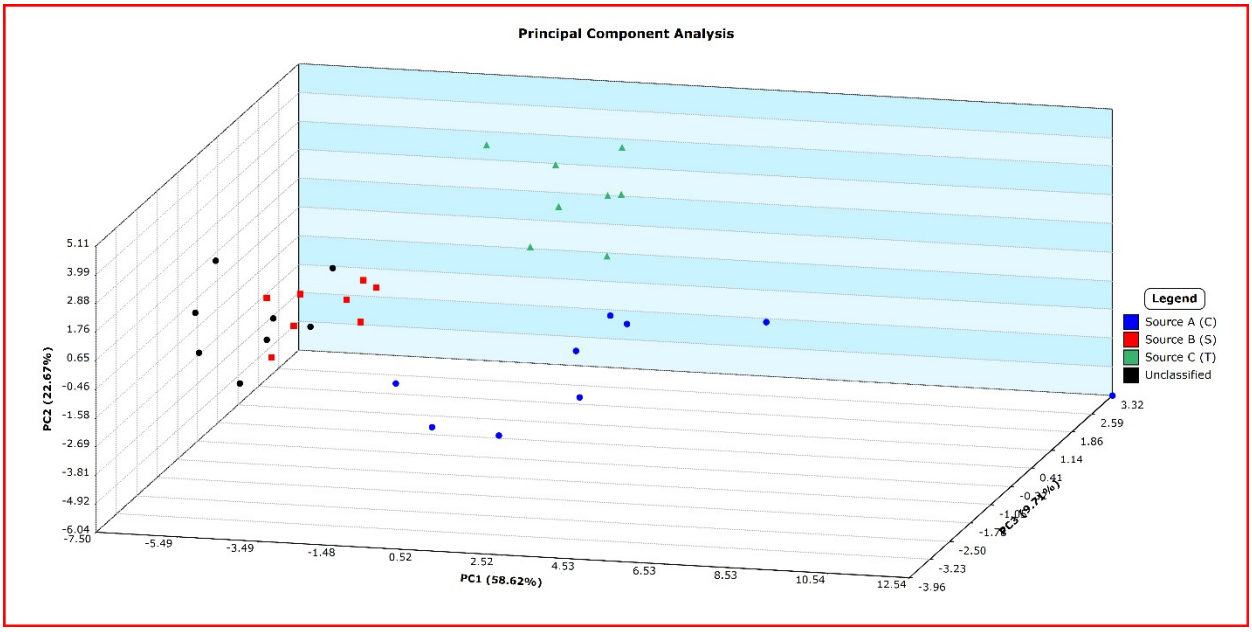

Figure S48: Round Robin 2021 Spill 2 PCA

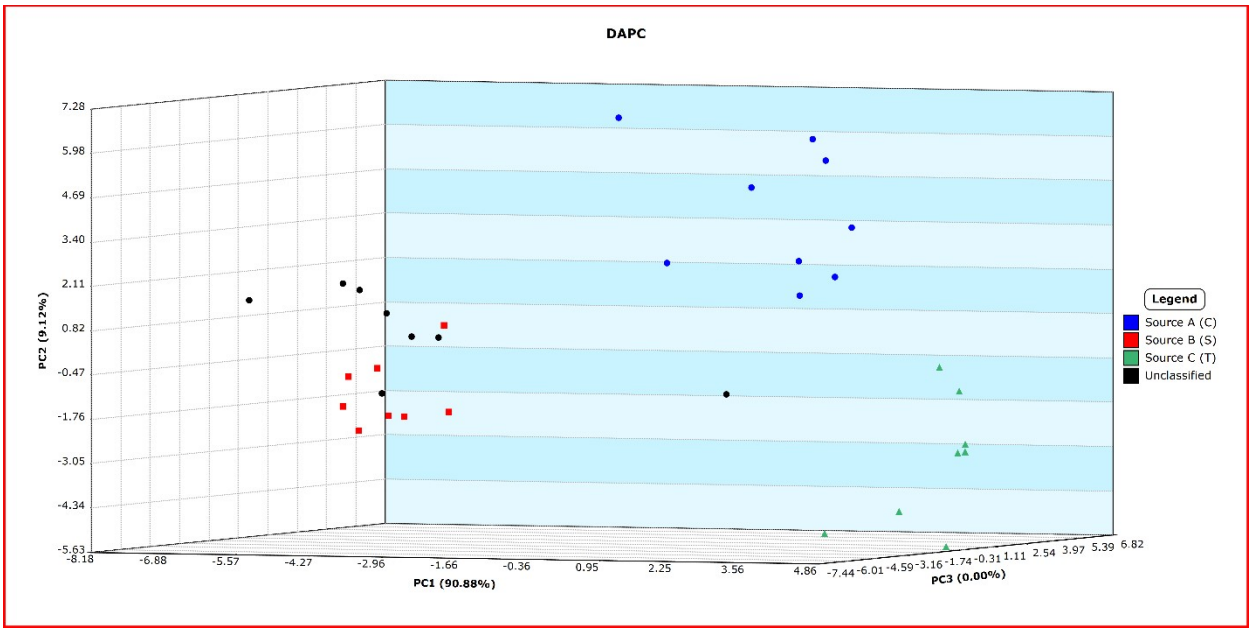

Figure S49: Round Robin 2021 Spill 1 DAPC

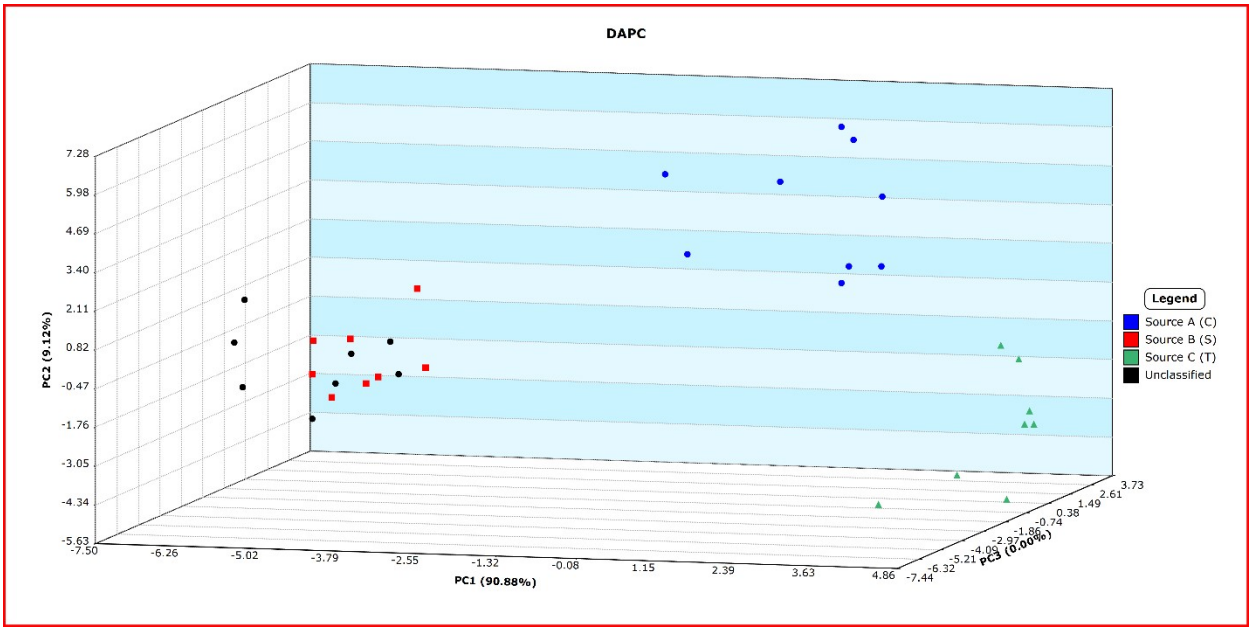

Figure S50: Round Robin 2021 Spill 2 DAPC