Supplementary Material:

Supplemental Table 1 – High-resolution metabolomics analysis of the tryptophan metabolic pathway. Baseline measurements of all detected metabolites with confirmed chemical identities in the tryptophan metabolic pathway were compared in persons with active TB disease versus controls without active TB disease and analyzed for change over time with effective active TB treatment. These metabolites were also compared in persons with latent TB infection (LTBI) versus uninfected controls at baseline and analyzed for change over time after treatment for LTBI. P-values for each comparison are displayed. Comparisons where the metabolite of interest was detected in <90% of samples are listed as not available (NA). Raw metabolite intensity measurements were $\log_2$ transformed and compared using limma for cross-sectional analyses and repeat measures limma for analyses of change over time.

	Tryptophan	Kynurenine	Kynurenine/ Tryptophan Ratio	5-Hydroxy- indoleacetate	Indole-3- acetaldehyde	Indole-3- ethanol	Indole	Indole-3- acetate	Indole-3- acetamide	Anthranilate	3-Hydroxy- anthranilate	2-Amino- phenol	Picolinate
Study 1 - Active TB Disease													
All cases of active TB (n=174) vs controls with (n=20) and without (n=37) LTBI	**4.71E-13**	**1.44E-07**	**<2.2E-16**	0.92	0.31	**0.05**	NA	0.1	0.4	0.34	0.46	**0.005**	0.58
South Africa HIV positive MDR-TB cases (n=64) vs controls with (n=20) and without (n=37) LTBI	**5.76E-12**	**2.48E-08**	**<2.2E-16**	0.64	0.64	0.12	NA	**0.02**	0.84	0.35	0.57	**8.15E-06**	0.76
South Africa HIV negative MDR-TB cases (n=21) vs controls with (n=20) and without (n=37) LTBI	**1.69E-06**	0.43	**2.49E-06**	0.07	0.16	0.18	NA	0.96	0.71	0.25	0.86	NA	0.48
Georgia HIV negative DS-TB cases (n=89) vs controls with (n=20) and without (n=37) LTBI	**1.36E-08**	**3.36E-06**	**1.11E-14**	0.73	0.24	**0.05**	NA	0.61	0.09	0.26	0.31	**0.05**	0.21
South Africa change during 2 years of MDR-TB treatment (n=17)	**0.009**	**1.14E-04**	**1.39E-06**	0.2	0.31	0.09	NA	0.08	0.37	0.78	0.76	0.2	0.24
Georgia change during first 4 months of DS-TB treatment	**3.62E-11**	**3.54E-07**	**<2.2E-16**	**0.04**	0.87	0.27	NA	0.46	0.62	**0.03**	**0.01**	0.81	0.18
Study 2 - Latent TB Infection													
All persons with LTBI (n=60) vs uninfected controls (n=40)	**0.007**	0.79	**0.006**	0.3	0.61	NA	**0.01**	0.68	NA	**0.05**	NA	0.15	0.66
Change during and after treatment for LTBI (n=28)	**0.008**	0.47	**5.26E-04**	0.4	**0.02**	NA	0.18	0.76	NA	0.96	NA	0.3	0.54

Supplemental Table 2 – Transcriptomics Analysis. Tables A-N contain a summary of the analysis of *IDO-1* and *IDO-2* transcripts for each data set acquired from the Gene Expression Omnibus (GEO) database. Each study is identified by its GEO accession number and contains the $\log_2$ fold change ($\log_2$ FC) in gene expression, test statistic (t) and p-value in persons with active tuberculosis (ATB) and latent tuberculosis infection (LTBI) versus healthy controls (HC) and persons with diseases other than TB and no evidence of infection with *M. tuberculosis* (OD). All comparisons were stratified based on whether participants were HIV-positive (P) or HIV-negative (N).

Table A: GSE37250

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs OD	IDO1	0.22	0.97	0.33
		IDO2	0.10	1.53	0.13
	ATB vs LTBI	IDO1	0.26	1.77	0.08
		IDO2	-0.06	-1.35	0.18
	LTBI vs OD	IDO1	-0.04	-0.2	0.85
		IDO2	0.16	2.37	0.02
P	ATB vs OD	IDO1	0.95	4.30	2.01E-05
		IDO2	0.11	1.71	0.09
	ATB vs LTBI	IDO1	0.03	0.18	0.86
		IDO2	-0.02	-0.56	0.58
	LTBI vs OD	IDO1	0.93	4.11	4.52E-05
		IDO2	0.14	2.05	0.04

Table B: GSE39939

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs LTBI	IDO1	-1.02	-3.17	0.002
		IDO2	-0.006	-0.1	0.92
	ATB vs OD	IDO1	0.61	2.48	0.01
		IDO2	0.01	0.23	0.82
	LTBI vs OD	IDO1	1.64	4.70	5.81E-06
		IDO2	0.02	0.26	0.8
P	ATB vs OD	IDO1	0.26	0.88	0.38
		IDO2	0.03	0.52	0.61

Table C: GSE39940

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs LTBI	IDO1	-0.2	-1.05	0.3
		IDO2	0.08	2.03	0.04
	ATB vs OD	IDO1	0.48	2.93	0.004
		IDO2	0.07	2.28	0.02
	LTBI vs OD	IDO1	0.68	3.83	0.0002
		IDO2	-0.002	-0.08	0.93
P	ATB vs OD	IDO1	0.53	2.52	0.01
		IDO2	0.1	2.33	0.02

Table D: GSE41055

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs LTBI	IDO1	0.07	1.42	0.16
		IDO2	-0.1	-2.01	0.05
	LTBI vs HC	IDO1	-0.03	-0.53	0.6
		IDO2	0.06	1.11	0.28
	ATB vs HC	IDO1	0.05	0.9	0.38
		IDO2	-0.05	-0.91	0.37

Table E: GSE42825

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	0.88	2.12	0.04
		IDO2	0.3	0.78	0.44
	ATB vs OD	IDO1	-0.14	-0.31	0.76
		IDO2	0.11	0.26	0.8

Table F: GSE42826

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	1.75	3.84	0.0002
		IDO2	0.43	2.07	0.04
	ATB vs OD	IDO1	1.2	2.55	0.01
		IDO2	0.1	0.48	0.64

Table G: GSE42830

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	1.33	2.42	0.02
		IDO2	0.45	1.27	0.21
	ATB vs OD	IDO1	1	1.85	0.07
		IDO2	0.07	0.2	0.85

Table H: GSE69581

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
P	ATB vs LTBI	IDO1	1.04	2.95	0.005
		IDO2	0.25	0.96	0.34

Table I: GSE73408

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs LTBI	IDO1	0.41	1.58	0.12
	ATB vs OD	IDO1	0.75	2.98	0.003
	LTBI vs OD	IDO1	0.34	1.36	0.18

Table J: GSE83456

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	1.42	8.88	3.66E-16
		IDO2	0.03	0.26	0.8
	ATB vs OD	IDO1	-0.13	-0.76	0.45
		IDO2	0.09	0.75	0.45

Table K: GSE19435

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	0.83	1.28	0.21
		IDO2	0.3	1.65	0.12

Table L: GSE19439

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	0.73	1.38	0.18
		IDO2	-0.07	-0.32	0.76
	ATB vs LTBI	IDO1	1.01	2.06	0.05
		IDO2	0.12	0.63	0.53
	LTBI vs HC	IDO1	-0.28	-0.56	0.56
		IDO2	-0.19	-0.96	0.34

Table M: GSE19442

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs LTBI	IDO1	1.73	4.05	0.0002
		IDO2	0.26	2.86	0.01

Table N: GSE19444

HIV Status	Comparison Groups	Gene	Log$_2$ FC	t	P.Value
N	ATB vs HC	IDO1	0.45	1.01	0.32
		IDO2	0.16	1.23	0.22
	ATB vs LTBI	IDO1	0.32	0.84	0.40
		IDO2	0.09	0.79	0.43
	LTBI vs HC	IDO1	0.13	0.29	0.77
		IDO2	0.07	0.56	0.58
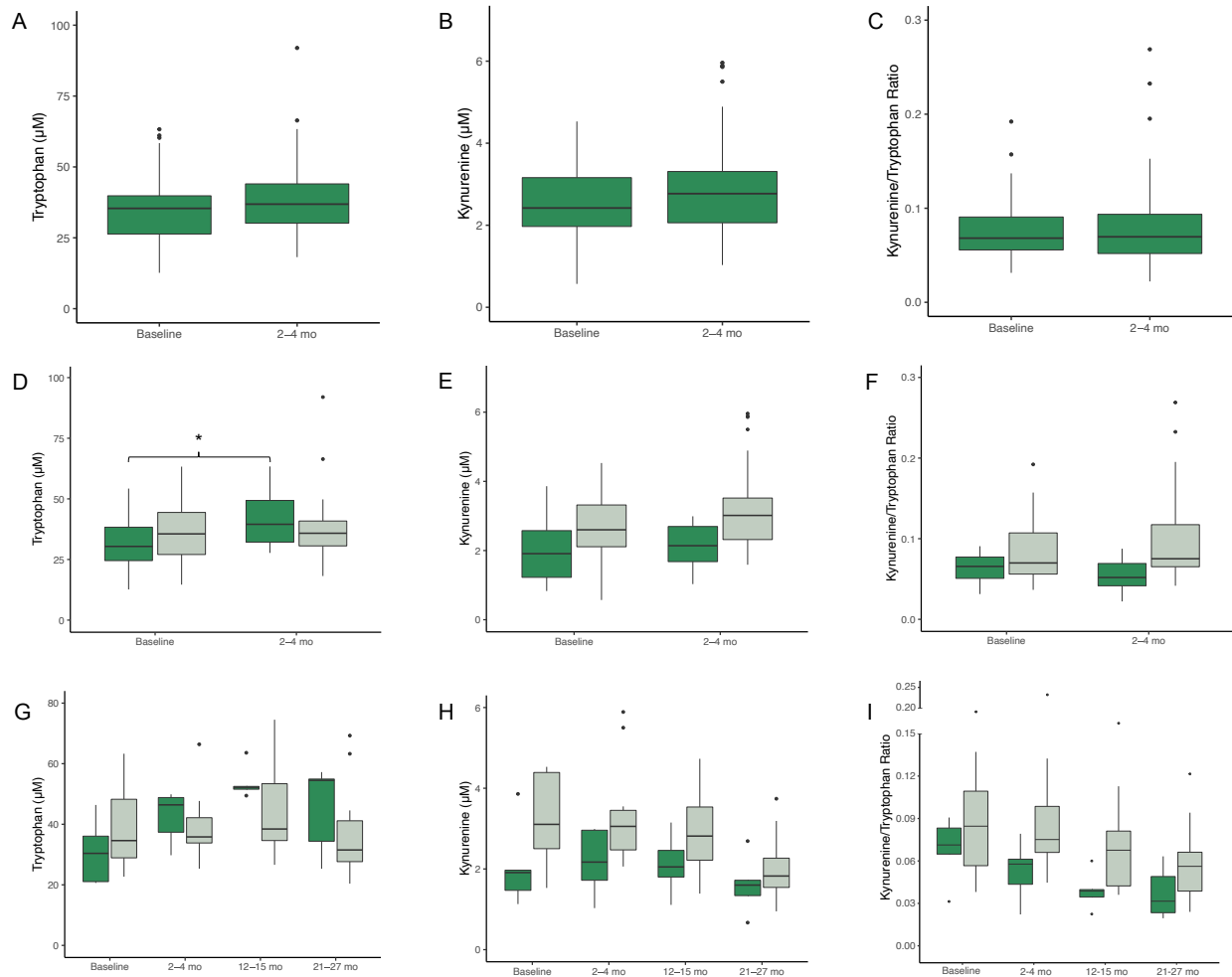
Supplemental Figure 1 – Tryptophan catabolism during multidrug resistant TB treatment in persons with and without HIV co-infection. (A) Plasma tryptophan concentration, (B) kynurenine concentration and (C) the kynurenine/tryptophan (K/T) ratio were unchanged in South Africans with multidrug-resistant (MDR)-pulmonary TB after 2-4 months of treatment versus baseline. (D-F) HIV-positive (n=26; gray) and HIV-negative (n=11; green) persons with active TB disease demonstrated similar trends in plasma tryptophan concentrations, kynurenine concentrations and the K/T ratio after 2-4 months of MDR-TB treatment. (G-I) HIV positive (n=12) and HIV negative (n=5) MDR-TB patients with plasma samples over the 2-year treatment period also demonstrated comparable trends in plasma tryptophan, kynurenine and the K/T ratio.
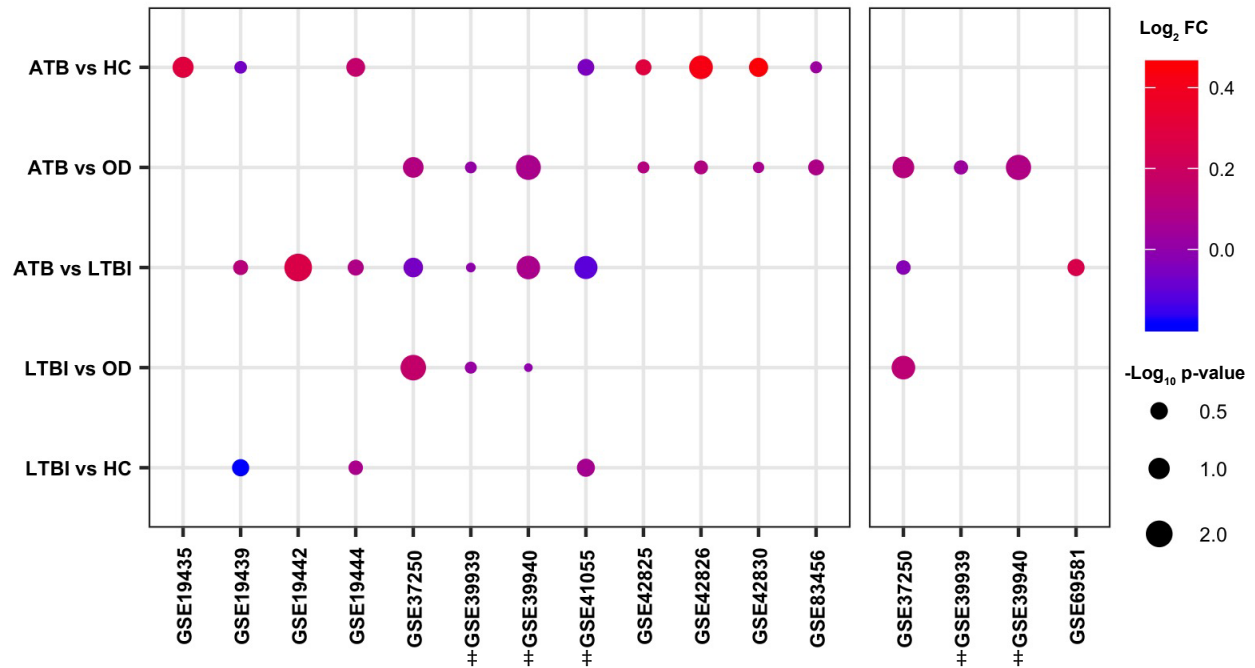
Supplemental Figure 2 – *IDO-2* transcripts in persons with active TB disease and latent TB infection with and without HIV co-infection. The bubble plots represent whole blood measurement of *IDO-2* transcripts in in HIV-negative persons (left box) and HIV-positive persons (right box). Study accession numbers from the publicly available Gene Expression Omnibus (GEO) data repository are shown on the x-axis and each comparison is shown on the y-axis. The color scale for each dot represents the $\log_2$ fold change in detected *IDO-2* transcripts for persons with active TB disease (ATB) and latent TB infection (LTBI) relative to healthy controls (HC) and person with diseases other than TB and no evidence of infection with *M. tuberculosis* (OD). The dot size represents the -log p-value for each comparison. ‡ indicates studies that were performed in children.
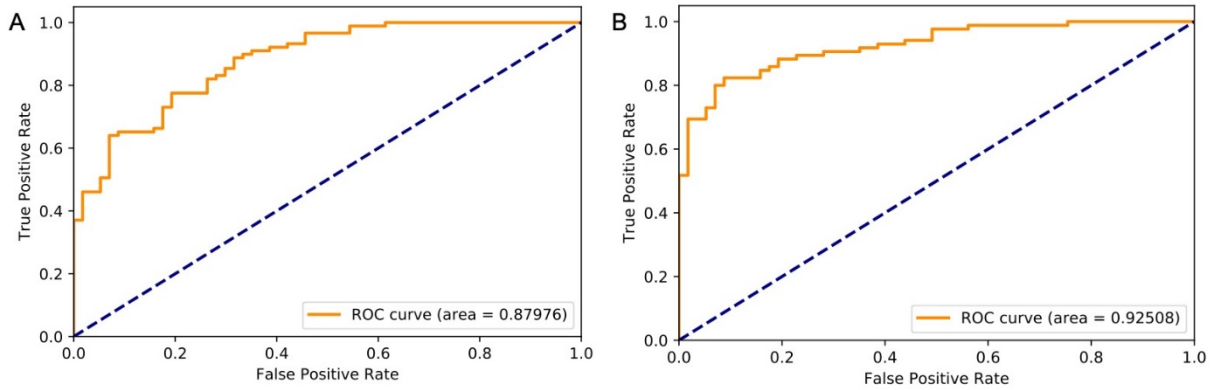
Supplemental Figure 3 – Cross-cohort receiver operator characteristic curves. Logistic

regression was used to determine the accuracy of the plasma kynurenine/tryptophan (K/T) ratio

in predicting active TB disease using the two active TB cohorts as training and validation sets.

The area under the curve (AUC) for the receiver operator characteristic (ROC) curve is

displayed for (A) the Georgia active TB cohort when the model was trained using active TB

patients from South Africa and (B) for the South Africa cohort when the model was trained using

active TB patients from Georgia.

Supplemental Figure 4 – MS/MS spectra provide definitive chemical identification for (A) tryptophan, (B) kynurenine, (C) anthranilate, (D) hydroxyanthranilate, (E) 2-aminophenol, (F) picolinate, (G) indole-3-acetaldehyde, (H) indole, (J) indole acetate, (K) indole acetamide and (L) 5-hydroxyindoleacetate (5-HIAA). For each figure, the top MS and MS/MS spectra are from a standardized reference plasma sample, while the bottom spectra demonstrate enhancement of the relevant MS and MS/MS peaks after spiking plasma samples with purified authentic standards of each compound.

A

Tryptophan – 205.0972 *m/z*
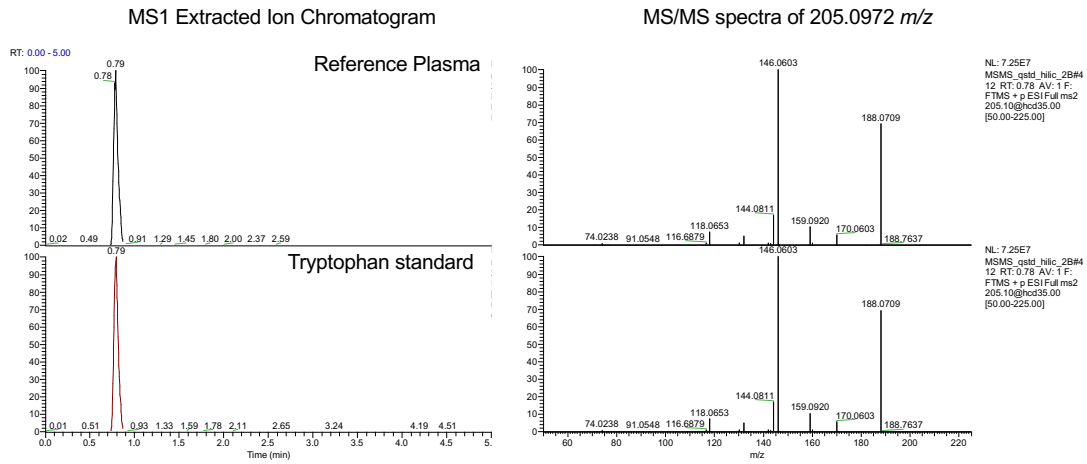
B
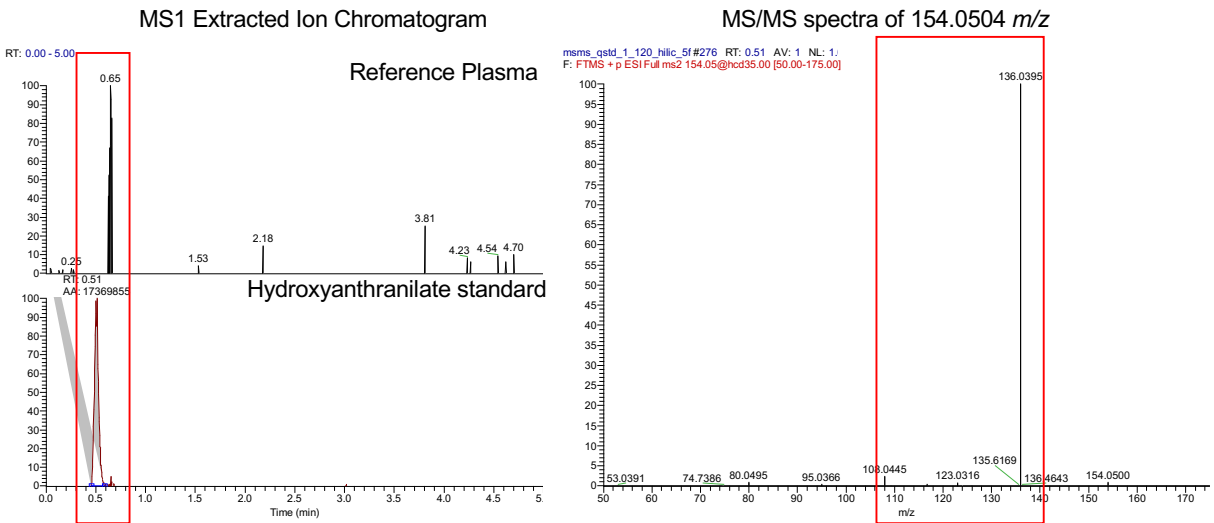
Kynurenine – 209.0921 *m/z*
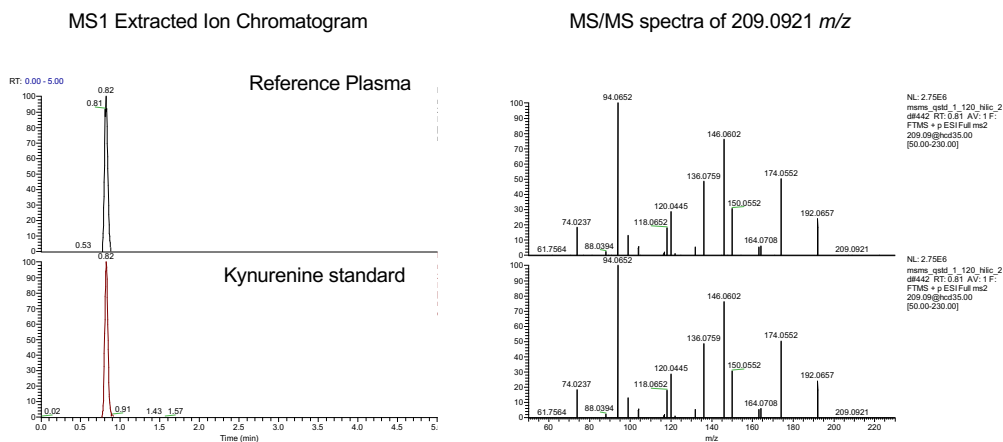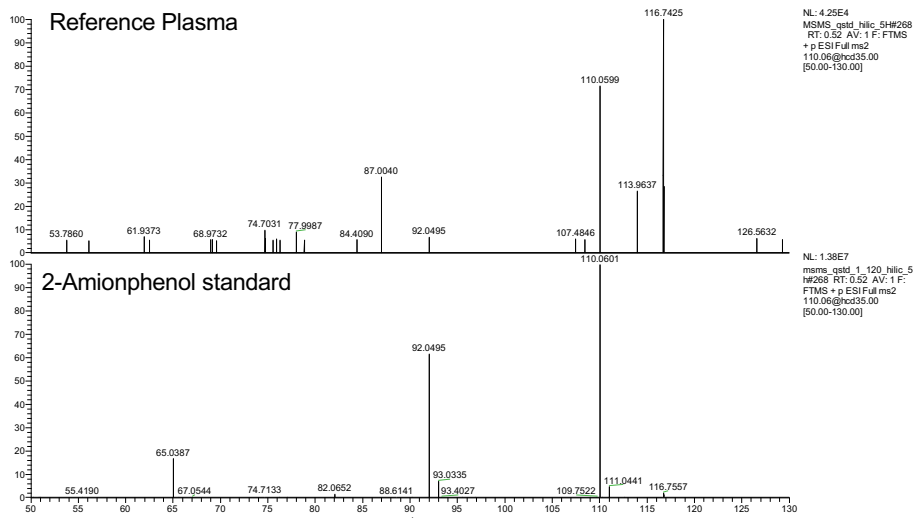
C

Anthranilate – 138.055 *m/z*

MS1 Extracted Ion Chromatogram

Reference Plasma

Anthranilate standard

MS/MS spectra of 138.055 *m/z*

D
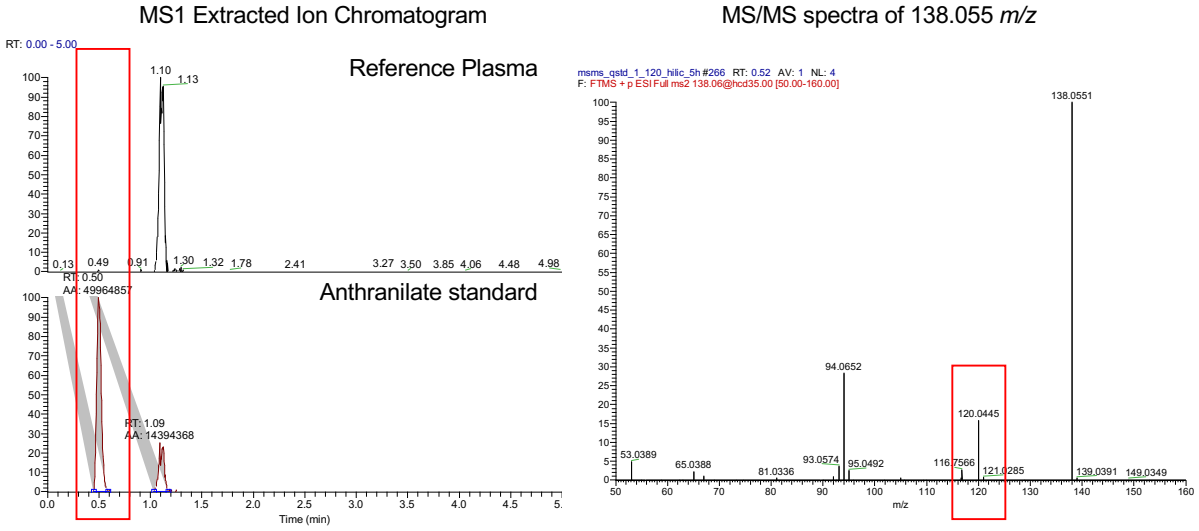
Hydroxyanthranilate – 154.0504 *m/z*

MS1 Extracted Ion Chromatogram

Reference Plasma

Hydroxyanthranilate standard

MS/MS spectra of 154.0504 *m/z*

E

2-Aminophenol – 110.0601 *m/z*

MS/MS spectra of 110.0601 *m/z*

Reference Plasma

NL: 4.25E4
MSMS_qstd_hilic_5H#268
RT: 0.52 AV: 1 F: FTMS
+ p ESI Full ms2
110.06@hcd35.00
[50.00-130.00]

116.7425
110.0599
113.9637
87.0040
53.7860 61.9373 68.9732 74.7031 77.9987 84.4090 92.0495 107.4846 126.5632

2-Amionphenol standard

110.0601

NL: 1.38E7
msms_qstd_1_120_hilic_5
h#268 RT: 0.52 AV: 1 F:
FTMS + p ESI Full ms2
110.06@hcd35.00
[50.00-130.00]

92.0495
65.0387
55.4190 67.0544 74.7133 82.0652 88.6141 93.0335 93.4027 109.7522 111.0441 116.7557

m/z

F

Picolinate – 124.0393 *m/z*

MS1 Extracted Ion Chromatogram

RT: 0.00 - 5.00

Reference Plasma

0.62
0.23
0.27
0.70
1.09 1.61 1.69 1.73 1.94 1.98
1.43
2.55
3.01 3.40 3.54 3.89 4.23 4.36 4.71 4.80

RT: 0.66
AA: 14677472

Picolinate standard

Time (min)

MS/MS spectra of 124.0393 *m/z*

msms_qstd_1_120_hilic_2e #356 RT: 0.67 AV: 1 NL: 6
F: FTMS + p ESI Full ms2 124.04@hcd35.00 [50.00-145.00]

96.0444
124.0394
116.6907
61.9124 74.6717 78.0339 96.9179 106.0288 112.1582 128.1110 133.8499

m/z

G
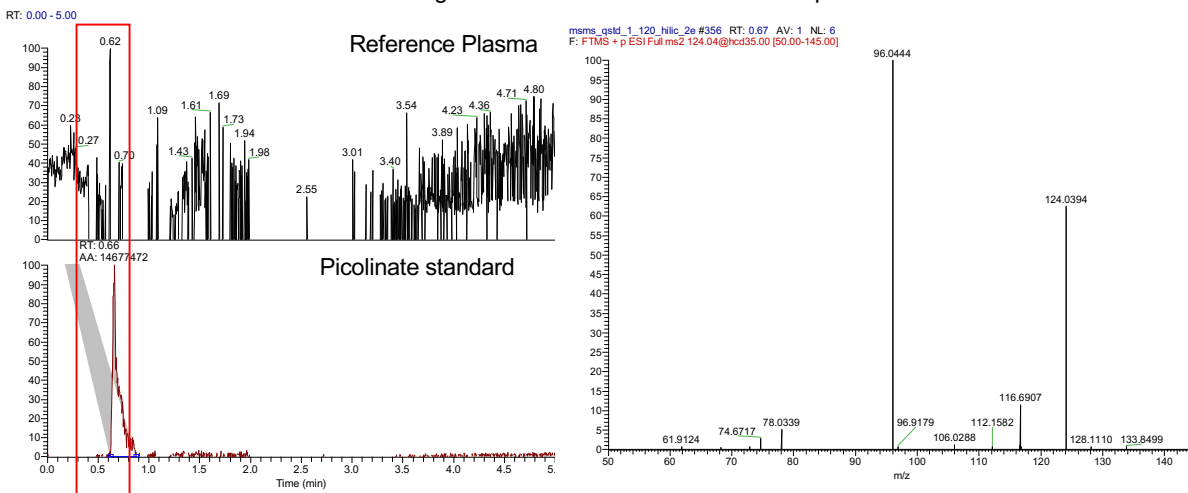
Indole-3-acetaldehyde – 160.0757 *m/z*

MS1 Extracted Ion Chromatogram

MS/MS spectra of 160.0757 *m/z*

Reference Plasma

I-3-A standard

160.0757 - 117.0575 *m/z*
fragment ion
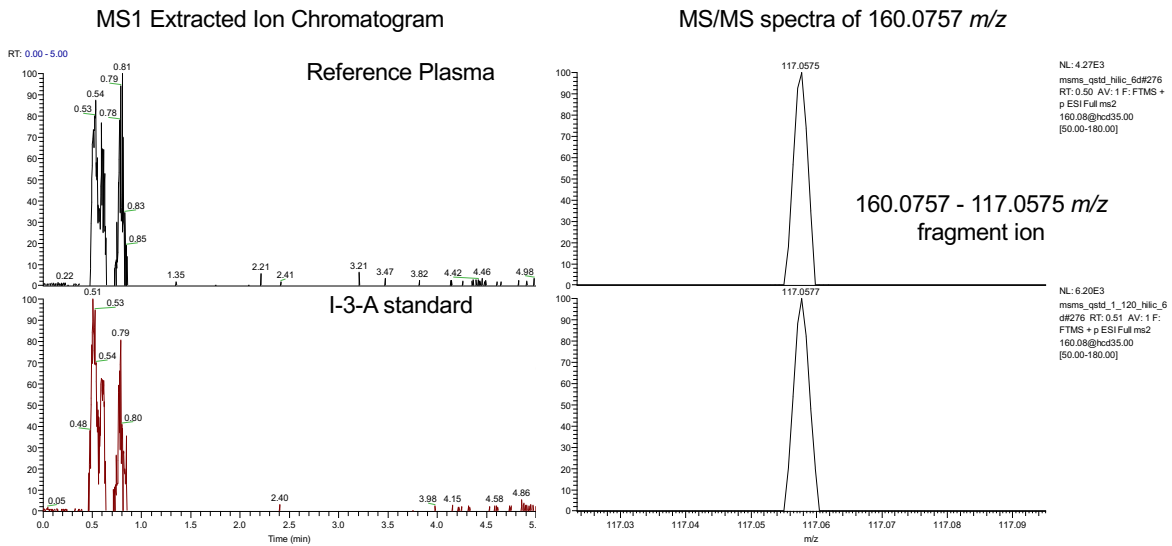
H

Indole-3-ethanol – 162.0914 *m/z*

MS1 Extracted Ion Chromatogram

MS/MS spectra of 160.0768 *m/z*

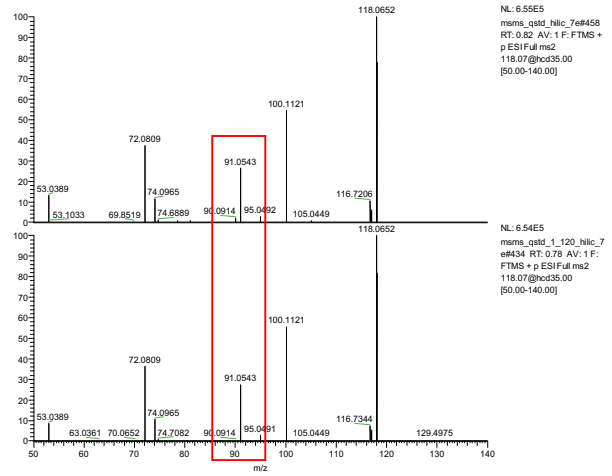Reference Plasma

I-3-E standard

MS/MS not informative
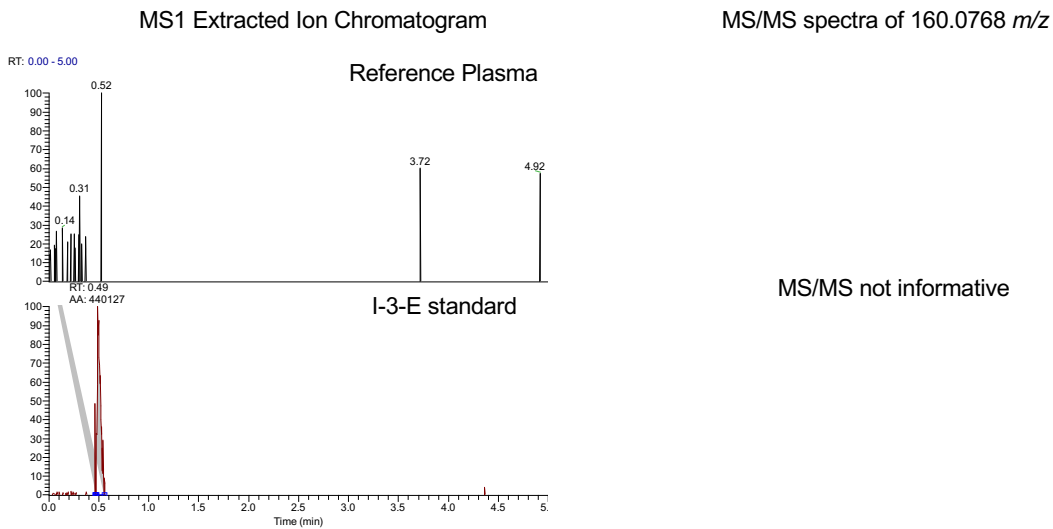
I

Indole – 118.0651 *m/z*

MS1 Extracted Ion Chromatogram

MS/MS spectra of 118.0651 *m/z*
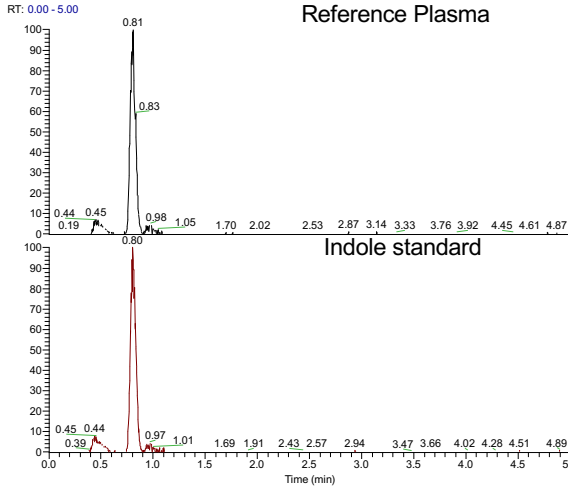
J

Indole acetate – 176.0706 *m/z*

MS1 Extracted Ion Chromatogram
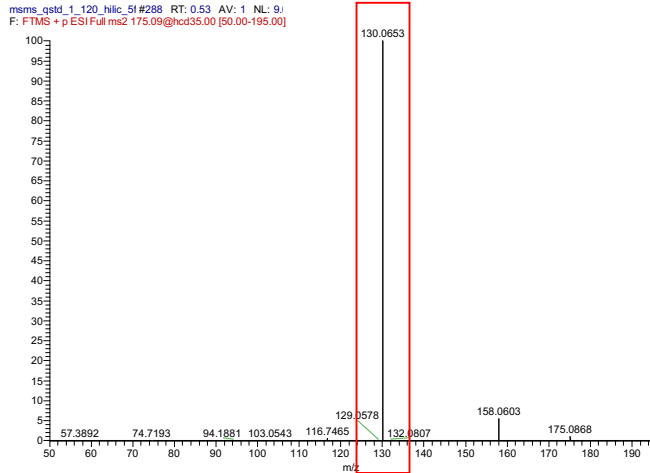
MS/MS spectra of 176.0706 *m/z*
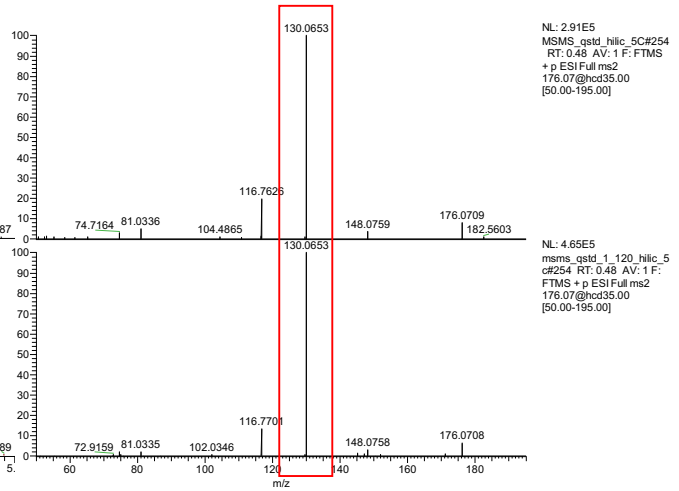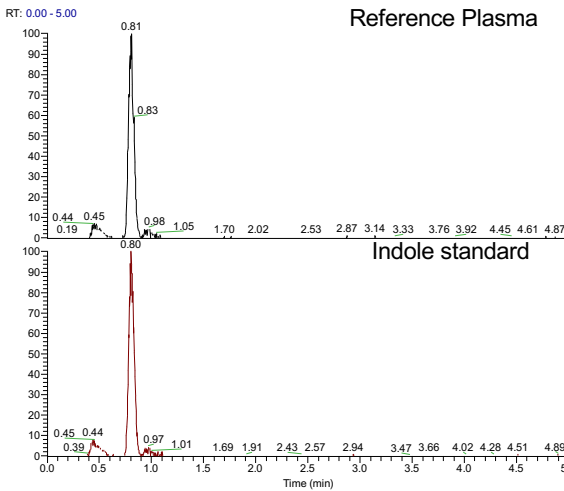
Indole acetamide – 175.0866 *m/z*

K

MS1 Extracted Ion Chromatogram

Reference Plasma

Indole acetamide standard

MS/MS spectra of 175.0866 *m/z*

5-HIAA – 192.0657 *m/z*

L

MS1 Extracted Ion Chromatogram

Reference Plasma

5-HIAA standard
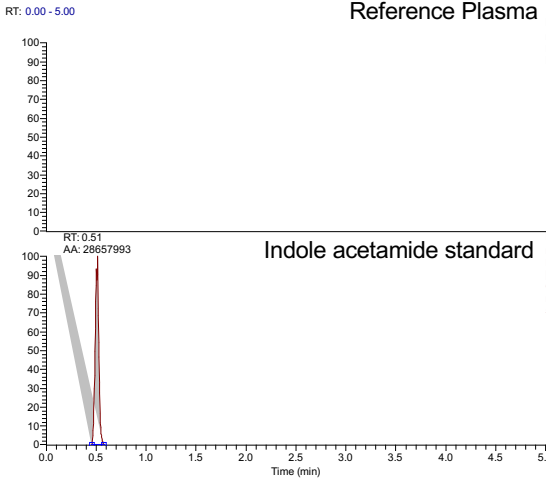
MS/MS spectra of 192.0657 *m/z*