

Table S7. Analytical validation of the developed method for tryptophan pathway metabolites determination in VAMS

Analytes	Linearity Range (ng/mL); (R ²)		LOQ (ng/mL)	Intra-Day Precision RSD(%)	Inter-Day Precision RSD(%)	Accuracy (% bias)	Matrix Effects (%)	Recovery (%)	Hematocrit effects	Stability	Additional data	Ref
TRP	25-10,000	0.9988	25	4.5% - 7.9%	4.7% - 9.5%	90% - 107%	87% - 96%	86% - 96%	–	86% - 95% (VAMS stored in a clamshell, in a bag with desiccant, protected from light at room temperature for 30 days).	Selectivity No interference peaks at target retention time. Carryover No signal in the blank was higher than background noise.	[10]
NFK	0.3-100	0.9991	0.3	RSD (low, medium, and high concentrations)	RSD (low, medium, and high concentrations)	(low, medium, and high concentrations)	(low, medium, and high concentrations)	(low, medium, and high concentrations)				
KYN	5-1000	0.9989	5									
ATA	0.2-100	0.9993	0.2									
KYNA	0.2-100	0.9992	0.2									
3-HKYN	0.2-100	0.9993	0.2									
XA	0.2-100	0.9994	0.2									
3-HAA	0.2-100	0.9992	0.2									
QUIN	1-500	0.9991	1									
PIC	1-500	0.999	1									
2-AMA	0.1-50	0.9995	0.1							> 85% (extracted samples stored in the autosampler for 24 hours at room temperature).		
5-HTP	0.1-50	0.9994	0.1									
5-HT	1-500	0.9991	1									
5-HIAA	0.1-50	0.9996	0.1									
MEL	0.2-100	0.9994	0.2									
IND	0.1-50	0.9994	0.1									
TA	0.1-50	0.9992	0.1									
IDE	0.1-50	0.9993	0.1							> 85% (extracted samples stored on the benchtop and exposed to all lab conditions for 24 hours).		
IPA	1-500	0.9989	1									
ILA	1-500	0.9991	1									
IAA	1-500	0.9991	1									
SK	0.1-50	0.9994	0.1									
IALD	0.2-100	0.9992	0.2									
IA	0.2-100	0.9993	0.2							87% - 91% for acidic compounds; 90% - 94% for other compounds (VAMS stored in a clamshell, in a bag with desiccant, protected from light at room temperature for > 30 days).		

Abbreviations: L-tryptophan (TRP), N-formylkynurenine (NFK), kynurenine (KYN), kynurenic acid (KYNA), 3-hydroxykynurenine (3-HKYN), anthranilic acid (ATA), kynurenine aminotransferase (KAT), serotonin (5-HT), melatonin (MEL), xanthurenic acid (XA), 3-hydroxyanthranilic acid (3-HAA), quinolinic acid (QUIN), picolinic acid (PIC), 2-aminomuconic acid (2-AMA), 5-hydroxytryptophan (5-HTP), 5-hydroxyindole acetic acid (5-HIAA), melatonin (MEL), indole (IND), tryptamine (TA), 3-indoleethanol (IDE), 3-indolepropionic acid (IPA), indolelactic acid (ILA), 3-indoleacetic acid (IAA), skatole (SK), 3-indolealdehyde (IALD), and indoleacrylic acid (IA), relative standard deviation (RSD).