

**Table 1**Analytical method validation<sup>a</sup> and stability data for vinpocetine and apovincaminic acid in dam plasma, amniotic fluid, and fetal homogenate.

	Plasma	Amniotic fluid	Fetal homogenate
<b>Vinpocetine</b>			
Concentration range (ng/mL or ng/g) <sup>b</sup>	0.5 to 100	0.5 to 100	5–1000
Linearity (r) <sup>c</sup>	≥ 0.999	0.999	0.998
LOQ (ng/mL or ng/g) <sup>d,e</sup>	0.5	0.5	5
LOD (ng/mL or ng/g) <sup>f</sup>	0.05	0.05	0.7
Accuracy (%RE)			
Intra-day	– 3.3 to 4.3	– 7.5 to 12.7	– 7.3 to 5.1
Inter-day	– 2.3 to 2.3	NA	– 2.0 to 2.7
Precision (%RSD)			
Intra-day	0.7 to 9.6	0 to 4.7	0.5 to 8.9
Inter-day	1.7 to 6.1	NA	1.4 to 8.3
Absolute recovery (%) <sup>g</sup>	89.6 to 124.4	81.2 to 102.0	103.8 to 150.3
Dilution verification, n = 6 (% RE, %RSD) <sup>h</sup>	– 7.9, 5.9	4.0, 2.3	6.9, 7.6
Extracted sample stability, n = 4 (%RE) <sup>i</sup>			
Ambient temperature (7d) <sup>j</sup>	– 7.5 to – 12.9	– 0.2 to 12.4	– 8.8 to – 0.4
Freeze-thaw (3 cycles)	– 16.5 to – 6.0	0.4 to 8.6	– 8.8 to – 2.9
Matrix stability (%RE) <sup>k</sup>			
Freeze-thaw (3 cycles)	– 14.8 to – 14.7	– 8.7 to – 3.0	1.3 to 4.7
Storage (61d)	– 12.9 to – 8.4	– 13.9 to – 9.1	4.2 to 6.0
<b>Apovincaminic acid</b>			
Concentration range (ng/mL or ng/g) <sup>b</sup>	0.5 to 100	0.5 to 100	
Linearity (r) <sup>c</sup>	≥ 0.995	0.997	0.998
LOQ (ng/mL or ng/g) <sup>d,e</sup>	0.5	0.5	5
LOD (ng/mL or ng/g) <sup>f</sup>	0.0567	0.140	0.777
Accuracy (%RE)			
Intra-day	– 16.3 to 16.3	– 7.6 to 7.8	– 2.8 to 6.9
Inter-day	– 7.8 to 6.7	NA	– 1.6 to 2.0
Precision (%RSD)			
Intra-day	0 to 23.7	0–10.7	0.8 to 15.2
Inter-day	3.2 to 14.4	NA	2.8 to 9.8
Absolute recovery (%) <sup>g</sup>	160.2 to 249.7	131 to 172	119.3 to 159.3
Dilution verification, n = 6 (% RE, %RSD) <sup>h</sup>	3.9, 8.9	– 4.9, 8.5	– 2.9, 3.7
Extracted sample stability, n = 4 (%RE) <sup>i</sup>			
Ambient temperature (7d) <sup>j</sup>	– 2.4 to – 5.4	– 7.3 to 14.7	– 14.1 to – 2.4
Freeze-thaw (3 cycles)	– 13.7 to 3.6	– 10.5 to 5.7	– 9.8 to 2.7
Matrix stability (% RE) <sup>k</sup>			
Freeze-thaw (3 cycles)	– 7.6 to 10.6	5.2 to 10.2	– 9.1 to 2.2
Storage (61d)	– 22.0 to – 12.4	– 11.4 to – 1.9	– 7.5 to – 2.2

<sup>a</sup> Full validations were performed in plasma and fetal homogenate. A partial validation was performed for amniotic fluid using plasma method.<sup>b</sup> Plasma and amniotic fluid concentrations are expressed as ng/mL and fetal homogenate as ng/g.<sup>c</sup> For both vinpocetine and AVA, plasma and amniotic fluid curves were fitted with linear 1/x weighted regression and fetal homogenate curves were fitted with linear 1/x<sup>2</sup> weighted regression.<sup>d</sup> LOQ, limit of quantitation; LOD, limit of detection, RE, relative error; RSD, relative standard deviation; NA, not applicable.<sup>e</sup> Experimental LOQ, is the lowest concentration used in standard curve. Target values are given.<sup>f</sup> Estimated as the 3 times the standard deviation of the LOQ (n = 6 replicates).<sup>g</sup> Values given are the range for the concentration range validated.<sup>h</sup> Highest concentration verified were: plasma and amniotic fluid, 1000 ng/mL; fetal homogenate, 10,000 ng/g.<sup>i</sup> Values given are the range for 3 QC concentrations at 1, 30 and 75 ng/mL in plasma and amniotic fluid and 7, 320 and 750 ng/g in fetal homogenate.<sup>j</sup> Values given are mean percent recovered after refrigerator, autosampler or ambient temperature storage at least 7 days.<sup>k</sup> Values given are for 2 target QC concentrations at 1 and 75 ng/mL in plasma and amniotic fluid and 7 and 750 ng/g in fetal homogenate.