

**Table S2.** Analytical validation data of the developed methods for the determination of endogenous steroids in VAMs.

Analytes	Linearity (R <sup>2</sup> )	LOQ	Intra-Day Precision (CV%)	Inter-Day Precision (CV%)	Accuracy	Matrix Effect (%)	Recovery (%)	Hematocrit Effects	Stability	Ref
T	0.9995	0.78 ng/mL	3.6% (2 ng/mL) 5.2% (10 ng/mL) 10.4% (25 ng/mL )	8.1% (2 ng/mL) 4.5% (10 ng/mL) 11.6% (25 ng/mL)	1.6% (2 ng/mL) 0.9% (10 ng/mL) 3.8% (25 ng/mL)	116%	89%	Not evaluated.	> 80% at RT after two months	[27]
T; ET; DHT; DHEA	> 0.99	T and ET: 1.0 ng/mL; DHEA and DHT: 1.5 ng/mL	T: 7% (LOQ) 6.4% (375 ng/ml) 5.9% (750 ng/ml)  ET: 6.8% (LOQ) 6.2% (375 ng/ml) 5.8 % (750 ng/ml)  DHT: 7% (LOQ) 6.9% (375 ng/ml) 6.7% (750 ng/ml)  DHEA: 7%(LOQ)	T: 6.6% (LOQ) 5.9% (375 ng/ml) 5.3% (750 ng/ml)  ET: 6.3% (LOQ) 5.8% (375 ng/ml) 5.5 % (750 ng/ml)  DHT: 6.9% (LOQ) 6.6% (375 ng/ml) 6.3% (750 ng/ml)  DHEA:6.9% (LOQ), 6.4 % (375 ng/ml), 6.1% (375 ng/ml)	89-97%	T: 6.1-8.7 % ET: 4.2-7% DHT: 3.2-5.1% DHEA:6.8-8.8 %	LOQ T: 77.3 ± 3.3 ET:79.1± 4.6 DHT: 81.6 ± 2.6 DHEA: 90.1 ± 3.7  375 ng/ml T: 78.4 ± 3.8 ET:79.1 ± 3.1 DHT: 80.4 ± 2.7 DHEA: 90.2 ± 3.4  750 ng/ml T: 80.3 ± 2.4 ET: 82.0 ± 2.8 DHT: 82.7 ± 1.9 DHEA: 92.0 ± 2.7	HCT effects NA (urine specimen)	Stability Results after 1 year, at RT:  LOQ T: 84% ET: 87 DHT: 84 DHEA: 83  375 ng/ml T: 85 ET: 88 DHT:86 DHEA: 84  750 ng/ml T: 85 ET: 88 DHT: 88 DHEA: 86	[28]
Cortisol (in Human); Cortisone (in Human); Corticosterone (in Rat) 11-Dehydrocorticosterone	0.993 - 0.999	-	Cortisol: 2% Cortisone: 32% Corticosterone: 18% 11-Dehydrocorticosterone: 29%	-	-	-	cortisol-d4: >96%  corticosterone-d8: >80%	HCT effect present: HCT-adapted volume factors are mandatory.	-	[29]
T; A4; 17-OHP	> 0.999	T: 1 nmol/L A4: 1nmol/L 17-OHP: 4nmol/L	T: 3.2 % (3.3 nmol/L) 3.0 % (12.5 nmol/L) 2.2 % (24.1 nmol/L)  A4: 3.8 % (3.0 nmol/L) 2.7 % (10.7 nmol/L) 2.4 % (19.7 nmol/L)  17-OHP: 6.5 % (4.8 nmol/L) 3.1 % (20.8 nmol/L) 3.5 % (38.1 nmol/L)	T: 5.3 % (3.3 nmol/L) 4.5% (12.5 nmol/L) 5.7% (24.1 nmol/L)  A4: 7.6% (3.0 nmol/L) 7.6 % (10.7 nmol/L) 5.8% (19.7 nmol/L)  17-OHP: 7.5% (4.8 nmol/L) 4.4% (20.8 nmol/L) 7.2% (38.1 nmol/L)	T: -0.6 % (3.3 nmol/L) 1.3 % (12.5 nmol/L) 1.8 % (24.1 nmol/L)  A4: -5.4 % (3.0 nmol/L) -8.8 % (10.7 nmol/L) -5.7 % (19.7 nmol/L)  17-OHP: -7.1 % (4.8 nmol/L) 6.7 % (20.8 nmol/L) 12.5 % (38.1 nmol/L)	"The imprecision (%CV) and bias were determined to be <10%"	101 - 104% (T)  95 - 100% (A4)  96 - 98% (17-OHP)	-12.8% - 4.3%	14 days at RT, 20 days at -20 °C, Storage at 60°C not acceptable  (Acceptance criteria: ± 15.0%)	[30]

Abbreviations: Testosterone (T), Androstenedione (A4), 17-hydroxyprogesterone (17-OHP), Epitestosterone (ET), Dihydrotestosterone (DHT), Dehydroepiandrosterone (DHEA)