

## **Supplementary Material**

**Supplementary file 1.** Precision and accuracy for quality control standard of mesterolone

**Supplementary file 2.** Robustness testing (n = 6)

**Supplementary file 3.** Recovery studies (n = 6)

**Supplementary file 1.** Precision and accuracy for quality control standard of mesterolone

Conc. (ng)	Analyst 1						Analyst 2		
	Intra-day			Inter-day			Found* (ng)	R.S.D. %	S.E.
	Found*	R.S.D.	S.E.	Found*	R.S.D.	S.E.			
300	304.57 ± 2.05 (ng)	0.67	1.18	302.37± 3.01 (ng)	0.99	1.73	300.57± 1.25 (ng)	0.41	0.72
500	499.43 ± 1.29	0.25	0.74	503.45± 2.07	0.41	1.19	500.62± 0.68	0.13	0.39
700	702.64 ± 1.33	0.18	0.76	700.52± 1.44	0.20	0.83	701.06± 0.51	0.07	0.29

\*Values are mean of n = 6.

**Supplementary file 2. Robustness testing (n = 6)**

Standard level	Mobile phase composition		Nature of TLC		Time from spotting to chromatography		Time from chromatography to staining		Time for staining		Time from staining to scanning	
Amount (ng/spot <sup>-1</sup> )	Amount detected (ng ± SD)	R.S.D. (%)	Amount detected (ng ± S.D.)	R.S.D. (%)	Amount detected (ng ± SD)	R.S.D. (%)	Amount detected (ng ± SD)	R.S.D. (%)	Amount detected (ng ± SD)	R.S.D. (%)	Amount detected (ng ± SD)	R.S.D. (%)
300	301.2 ± 2.25	0.74	300.2 ± 2.61	0.86	303.29 ± 3.47	1.14	298.17 ± 2.31	0.77	302.53 ± 2.07	0.68	302.14 ± 3.5	1.16
500	505.02 ± 3.86	0.76	503.6 ± 1.95	0.38	498.52 ± 2.50	0.50	502.83 ± 2.35	0.46	506.13 ± 2.13	0.42	502.43 ± 2.33	0.46
700	705.23 ± 2.48	0.35	699.46 ± 1.51	0.21	704.04 ± 1.57	0.22	700.65 ± 1.64	0.23	706.45 ± 2.52	0.35	703.25 ± 1.27	0.18

**Supplementary file 3.** Recovery studies (n = 6)

Sample	Standard spiked %	Theoretical ng spot <sup>-1</sup>	Mean recovery %	RSD %
M1	50	351.5	98.3	0.45
	100	468.7	99.2	0.56
	150	585.9	97.5	0.32