Preliminary Report on Impact of Immulite 2000 Cortisol antibody change October 2020 including evaluation of manufacturer recommended adjustment factors

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Updated 3rd November 2020 (First report 23rd October, Second Report 27th Oct, Third Report 29th October)

Based on 466 results from 10 labs submitted through an online shared spreadsheet.

The most up-to-date version of this document is here.

# Raw results from Lot 550 and beyond

See next section for evaluation of manufacturer recommended adjustment factors

Serum Overall (canine)

Overall there is negative bias in the results following the antibody change (-23%). The bias varies across concentrations which may make the implementation of simple equipment correction factors difficult as a means to address the change.

	N =	401	
		Minimum	**Maximum**
Before 550		27	1380
550 and after		27	1311

Average Bias nmol/L		-40
Average Bias %		-22.8

Cortisol range nmol/l	n	Average %Bias	% Bias range	
<50	77	-15.1	-46.7	3.0
50-150	152	-26.9	-134.5	3.9
150-250	66	-24.7	-82.1	14.7
250-500	65	-21.9	-56.8	13.8
>500	41	-20.2	-55.8	2.6
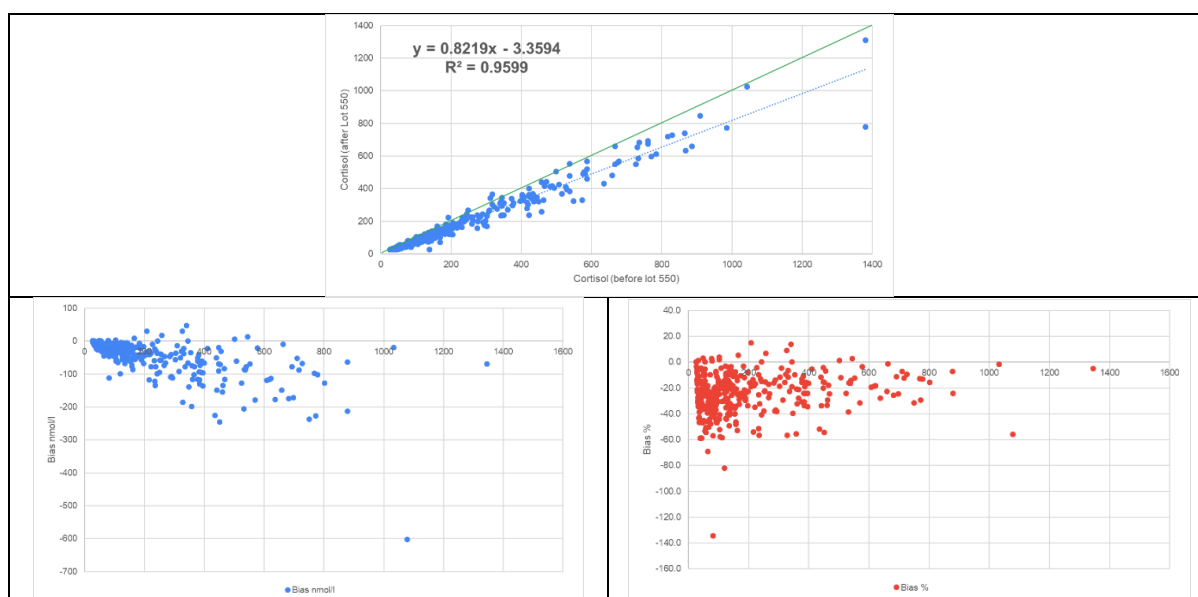
If adjustment of cut-off values is a consideration, the following are the outcome of simple linear regression.

Original Cut-off	Regression derived adjusted cut-off
40	29
50	37
138	110

150	120
250	203
500	410
550	452
600	493

Subjective visual assessment of paired results may also help in deciding new interpretative guidance:

For 40nmol/l:

Previous	New
38	29.8
38	27.6
38	30
38.6	32.8
38.6	28.1
39	30
39.5	27.9
39.5	29.8
40	29.5
41	33
41	29
41	35
41.9	27.6
42	33
42	32

e.g. 30nmol/L

For 50 nmol/L:

Previous	New
48	30.3
48	36
49	50.5
49.1	34.2
49.7	42.8
51	41.9
51	47.7
51	46
52	37.5
52.1	28.4

e.g., 40nmol/L

For 138 nmol/l:

Previous	New
135	106
136	131
136	75
138	27.6
139	104
141	115
141	128
141.6	131.1
142	119
142	113
142	123
142	105

e.g., 110 nmol/l

For 150nmol/l:

Previous	New
145	140
147	103
149	127
149	139
150	113
151	110
152	107
153	98.2
154	109
155	99

e.g., 120nmol/L

For 250 nmol/L:

Previous	New
241	204
244	244
246	223
248.7	266.6
255	224
260	183

e.g., 200 nmol/l

For 500 nmol/l

Previous	New
472	441
483	414
488	417
494	403
499	505
508	425
516	367
527	411
530	395

e.g., 400nmol/l

For 550 nmol/L:

Previous	New

For 600 nmo/l:

Previous	New

538	477		574	328
538	384		577	491
538	552		579	502
549	323		582	494
574	328		588	461
577	491		588	568
			588	519
			635	430

e.g. 450 nmol/L

e.g., 500 nmol/L

Trilostane samples (within 12hrs of dose, canine)

On earlier versions of this evaluation, it appeared that the "within 12 hours of Trilostane" subset was less dramatically affected. However, with more data-points, it appears that this subset has similar bias to the "all canine sera" data. A separate analysis was performed because manufacturer information includes a change in antibody cross reactivity with adrenal steroid pre-cursors. Trilostane has been reported to cause an accumulation of adrenal steroid precursors.

	N =	32	
	Minimum	**Maximum**	
Before 550	27	384	
550 and after	27	348	

Average Bias nmol/L	-19	
Average Bias %	-20.2	

Cortisol range nmol/l	n	Average %Bias	% Bias range	
<50	10	-6.4	-27.5	1.2
50-150	11	-37.7	-134.5	1.0
150-250	9	-15.9	-40.7	14.7
250-500	2	-11.9	-14.0	-9.8
>500	0			
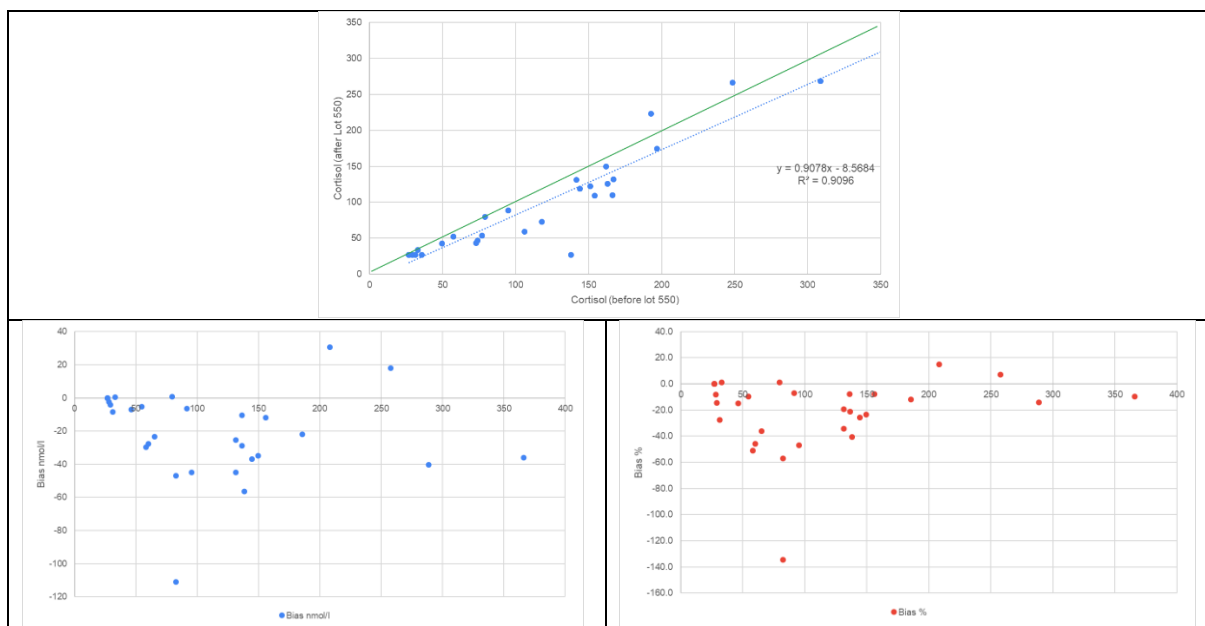
This data subset included trilostane monitoring ACTH stimulations tests (usually with 6 hours of trilostane dose). It did not include "pre-Vetoryl-cortisol" which is usually sampled at 12 or 24hrs after administration.

Urine

The ESVE collaboration data is "urine cortisol" rather than "urine corticoid:creatinine ratio".

Presumably as a result of the new antibody having different cross-reactivity profile for urinary corticoid metabolites, the bias impact on urine cortisol (around -70%) seems to be much greater than that on serum.

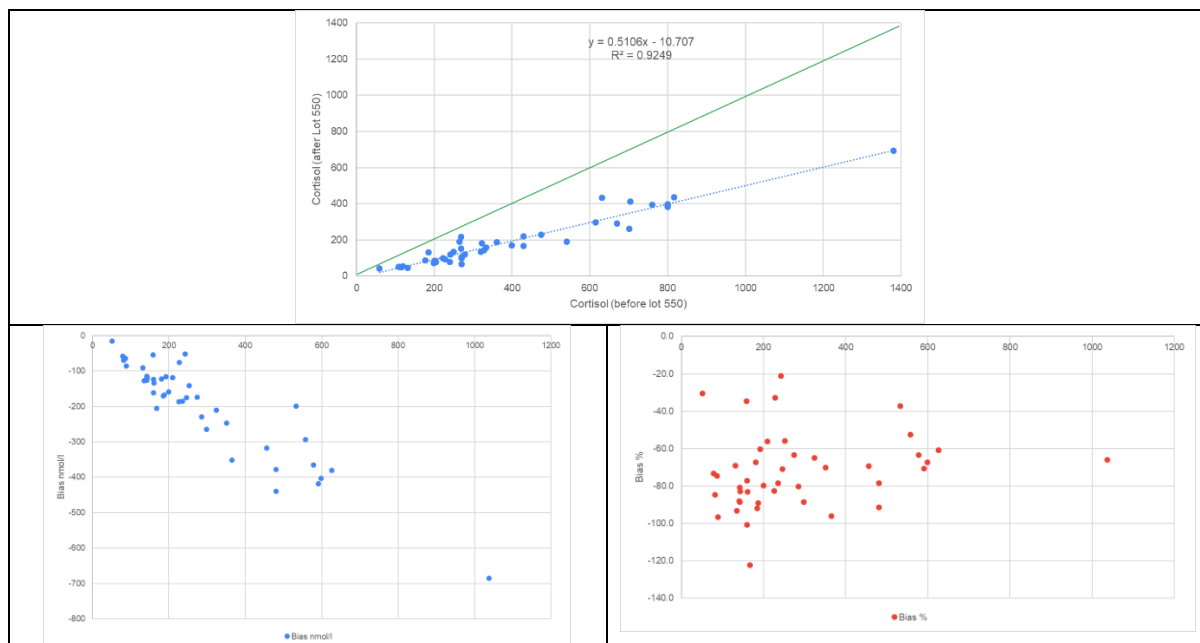|	N=	44

	Minimum	Maximum
Before 550	59.3	1380
550 and after	43.6	695

Average Bias nmol/L	-198
Average Bias %	-72.5

Urine Cortisol range nmol/l	n	Average %Bias	% Bias range	
100-200	7	-75.2	-96.6	-34.6
200-500	25	-75.1	-122.3	-21.1
>500	11	-68.5	-96.0	-37.4

A substantial decrease in cut-off values for UCCR will be required (e.g. to possibly 10 from 30). The coloration does not have creatinine values in the data-file to be able to confirm new suggested cut-offs, but on average it would be around 30% of original.

Other species

Only a very small number of paired results are so far available for horse (n=6) and cats (n=3). So far, the data suggest that the impact on cats may be similar to dogs but that for horses it may be less.

Species	Before Lot 550	After Lot 550	Average	Bias (nmol/l)	Bias %
Feline	74	56	65	-18	-27.7
Feline	98	69	83.5	-29	-34.7
Feline	32	27.6	29.5	-5	-16.9
Equine	126	111	118.5	-15	-12.7
Equine	51	38	44.5	-13	-29.2
Equine	154	173	163.5	19	11.6
Equine	187	171	179	-16	-8.9
Equine	160	152	156	-8	-5.1
Equine	113	108	110.5	-5	-4.5

Results incorporating manufacturer adjustment factors in Lot 550 and beyond

Siemens has proposed the following adjust formula to mitigate the impact of the antibody change. Initially labs would need to be incorporate it LIMS systems but eventually to be included in veterinary specific reagent barcodes and reagent packs.

$$\text{historical value} = (1.1 \times \text{new antibody kit value}) + 4.14 \text{ nmol/L}$$

The ESVE collaboration data has been reanalysed to assess the impact of the manufacturer correction factor.

The correction factor shifts the reportable range from 27.6 to 1380 to 34.5 to 1446

Serum Overall (canine)

Overall there is negative bias in the results following the antibody change (-23%). The bias varies across concentrations which may make the implementation of simple equipment correction factors difficult as a means to address the change.

N=	401	
	Minimum	**Maximum**
Before 550	<27.6	1380
Adj 550 and after	<34.5	1446
Average Bias nmol/L	-19	
Average Bias %	-8.2	

Cortisol range nmol/l	n	Average %Bias	% Bias range	
<50	77	6.8	-25.1	22.2
50-150	152	-12.1	-121.2	16.9
150-250	66	-12.7	-69.5	25.8
250-500	65	-11.1	-46.2	24.3
>500	41	-10.1	-46.4	12.8
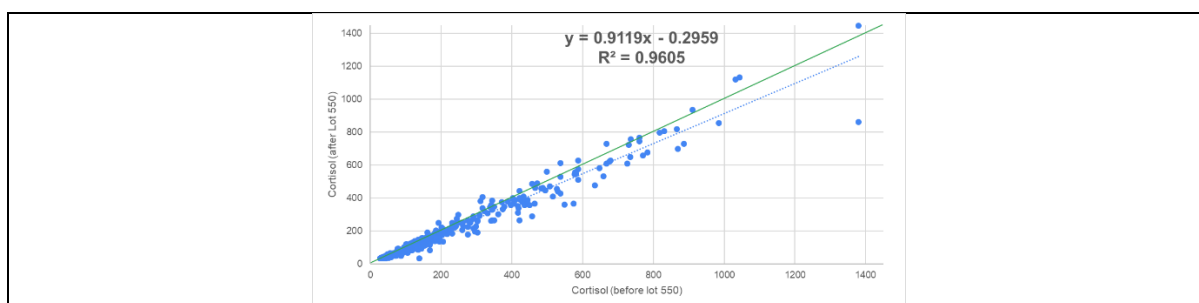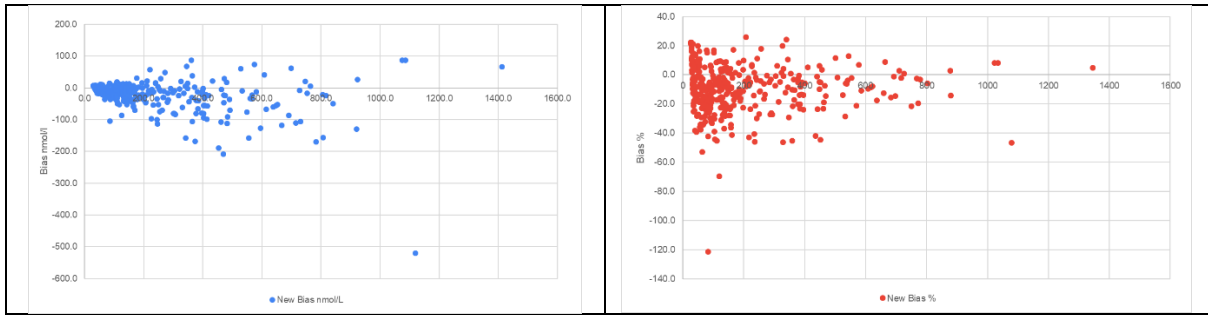
If adjustment of cut-off values is a consideration, the following are the outcome of simple linear regression.

Original Cut-off	Regression derived adjusted cut-off
40	36
50	45
138	126
150	136
250	228
500	456
550	501
600	547

Subjective visual assessment of paired results may also help in deciding new if a change in interpretative guidance is necessary:

For 40nmol/l:

Previous	New
38	36.9
38	34.5
38	37.1
38.6	40.2
38.6	35.1
39	37.1
39.5	34.8
39.5	36.9
40	36.6
41	40.4
41	36.0
41	42.6
41.9	34.5
42	40.4
42	39.3

e.g. ?? No change

For 50 nmol/L:

Previous	New
48	37.5
48	43.7
49	59.7
49.1	41.8
49.7	51.2
51	50.2
51	56.6
51	54.7
52	45.4
52.1	35.4

e.g., ?? no change

For 138 nmol/l:

Previous	New
135	120.7
136	148.2
136	86.6
138	34.5
139	118.5
141	130.6
141	144.9
141.6	148.4
142	135.0
142	128.4
142	139.4
142	119.6

e.g., 125 nmol/l

For 150nmol/l:	
Previous	New
145	158.1
147	117.4
149	143.8
149	157.0
150	128.4
151	125.1
152	121.8
153	112.2
154	124.0
155	113.0

e.g., 135nmol/L

For 250 nmol/L:	
Previous	New
241	228.5
244	272.5
246	249.4
248.7	297.4
255	250.5
260	205.4

e.g., ?? no change

For 500 nmol/l	
Previous	New
472	489.2
483	459.5
488	462.8
494	447.4
499	559.6
508	471.6
516	407.8
527	456.2
530	438.6

e.g., 450nmol/l

For 550 nmol/L:	
Previous	New
538	528.8
538	426.5
538	611.3
549	359.4
574	364.9
577	544.2

e.g. 500 nmol/L

For 600 nmo/l:	
Previous	New
574	364.9
577	544.2
579	556.3
582	547.5
588	511.2
588	628.9
588	575.0
635	477.1

e.g., 550 nmol/L

Trilostane samples (within 12hrs of dose, canine)

A separate analysis was performed because manufacturer information includes a change in antibody cross reactivity with adrenal steroid pre-cursors. Trilostane has been reported to cause an accumulation of adrenal steroid precursors.

	N =	32	
	Minimum	**Maximum**	
Before 550	27.6	384	
Adj 550 and after	34.5	387	

Average Bias nmol/L	-5	
Average Bias %	-4.1	

Cortisol range nmol/l	n	Average %Bias	% Bias range	
<50	10	15.9	-3.1	22.2
50-150	11	-22.9	-120.0	15.1
150-250	9	-3.9	-28.2	25.8
250-500	2	-1.2	-3.1	0.8
>500	0			
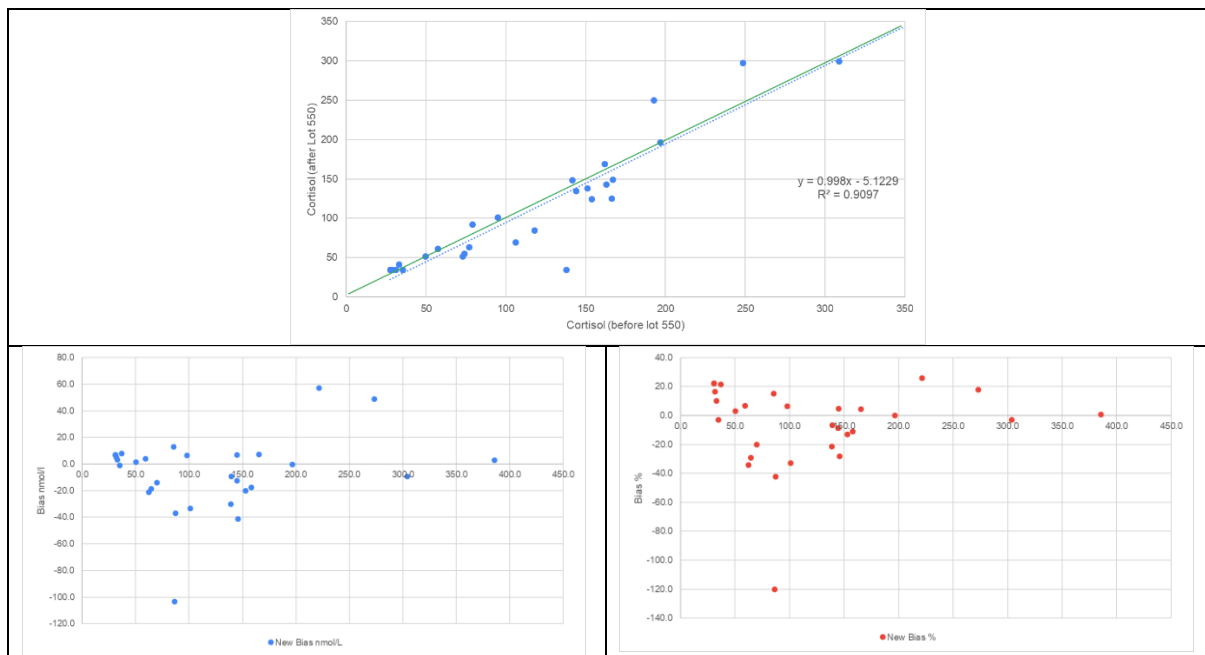
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Urine

The ESVE collaboration data is "urine cortisol" rather than "urine corticoid:creatinine ratio".

Presumably as a result of the new antibody having different cross-reactivity profile for urinary corticoid metabolites, the bias impact on urine cortisol seems to be much greater than that on serum. The correction formula derived for serum does not correct urine results and so new interpretative guidance

	n	44

	Minimum	Maximum
Before 550	59.3	1380
Adj 550 and after	52.1	769

Average Bias nmol/L	-175
Average Bias %	-61.4

Cortisol range nmol/l	n	Average %Bias	% Bias ranges	
100-200	7	-61.7	-82.7	-22.5
200-500	25	-64.2	-112.4	-9.9
>500	11	-59.1	-86.9	-27.2
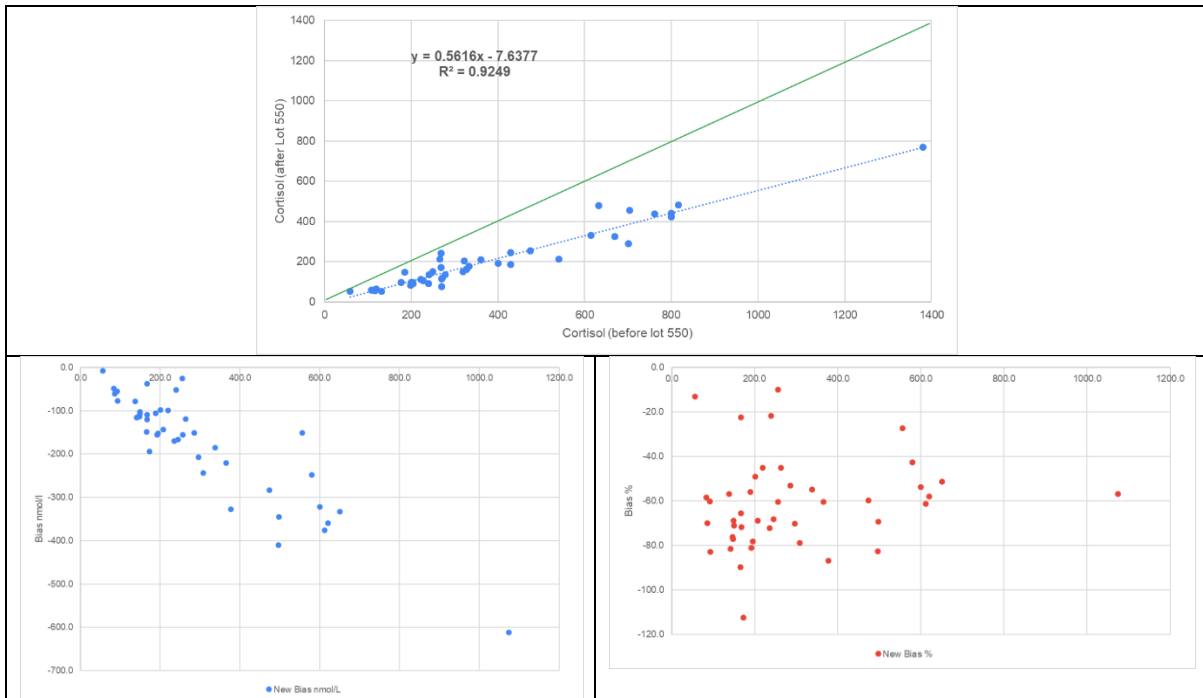
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Other species

Only a very small number of paired results are so far available for horse (n=6) and cats (n=3). So far, the data suggest that the impact on cats may be similar to dogs but that for horses there may be a positive bias. More data are required.

Species	Before Lot 550	Adj After Lot 550	Average	Bias (nmol/l)	Bias %
Feline	74	66	69.9	-8.3	-11.8
Feline	98	80	89.0	-18.0	-20.2
Feline	32	<33.8	32.9	1.8	5.6
Equine	126	126	126.1	0.2	0.2
Equine	51	46	48.5	-5.1	-10.4
Equine	154	194	174.2	40.4	23.2
Equine	187	192	189.6	5.2	2.8
Equine	160	171	165.7	11.3	6.8
Equine	113	123	118.0	9.9	8.4