Imprecision:

Reproducibility of the Follistatin assay was determined in a study using two Serum pools. The study included a total of 12 assays, three replicates of each per assay (n=36). Representative data were calculated based on NCCLS EP5-A guidelines and are presented in the following table.

Sample	Mean conc.	Within run		Between run		Total	
	(ng/mL)	SD	%CV	SD	%CV	SD	%CV
QC 1	1.22	0.04	3.59%	0.06	5.21%	0.07	6.33%
QC 2	2.69	0.07	2.69%	0.08	2.82%	0.10	3.90%

Cross reactivity and specificity:

The antibody pair detects Human and Equine Follistatin in the assay. Other related analytes at the concentration in the table below did not show any cross-reactivity.

Sample No.	Cross-reactant	Concentration (ng/mL)	% Cross-reactivity
1	Inhibin A	10	ND
2	Inhibin B	1.25	ND
3	Activin A	50	ND
4	Activin B	50	ND
5	Activin AB	50	ND
6	Alpha-2-Macroglobulin	40	ND
7	Myostatin	50	ND
8	AMH	50	ND
9	FSTL-3	48	ND

Linearity:

Based on dilutions of the three serum samples containing various Follistatin levels diluted with Calibrator A/sample diluent the percent recovery on individual samples is represented in the following table:

Sample	Dilution Factor	Expected Conc. (ng/mL)	Observed Conc. (ng/mL)	% Recovery
1	Neat	4.28	Neat	NA
	1:2	2.14	2.10	99%
	1:4	1.07	1.03	96%
	1:8	0.53	0.53	100%
2	Neat	3.60	Neat	NA
	1:2	1.80	93	107%
	1:4	0.90	1.01	112%
	1:8	0.45	0.53	118%
3	Neat	5.46	Neat	NA
	1:2	2.73	2.90	106%
	1:4	1.36	1.50	110%
	1:8	0.68	0.67	98%

Recovery:

Known amounts of Follistatin were added to four serum samples containing different levels of endogenous Follistatin. The concentration of Follistatin was determined before and after the addition of exogenous Follistatin and the percent recovery was calculated.

Sample	Endogenous Conc.(ng/mL)	Expected Conc. (ng/mL)	Observed Conc. (ng/mL)	% Recovery
1	1.45	1.73	1.64	95%
		1.64	1.57	96%
		1.56	1.40	90%
2	2.88	2.44	2.28	93%
		2.59	2.34	90%
		2.71	2.56	95%
3	2.88	11.26	12.00	107 %
		8.47	8.56	101%
		6.23	6.64	107%
4	1.41	10.53	11.42	108%
		7.49	7.82	104%
		5.06	5.03	99%

Interference:

When potential interferents (hemoglobin, triglycerides and bilirubin) were added at least at two times their physiological concentration to control sample, Follistatin concentration were within ± 10% of the control as represented in the following table.

Interferents	Analyte Conc. (mg/Ml)	Unspiked Sample Value (ng/mL)	Spiked Sample Value (ng/mL)	% Difference
Hemoglobin	1.35	8.60	9.06	5.35
		3.91	3.76	-3.84
Triglycerides	5.00	1.50	1.52	1.33
		3.91	3.84	-1.79
Bilirubin	0.80	8.92	8.60	-3.59
		1.54	1.50	-2.60

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures.

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Ansh Labs
445 Medical Center Blvd.
Webster, TX 77598-4217, U.S.A.