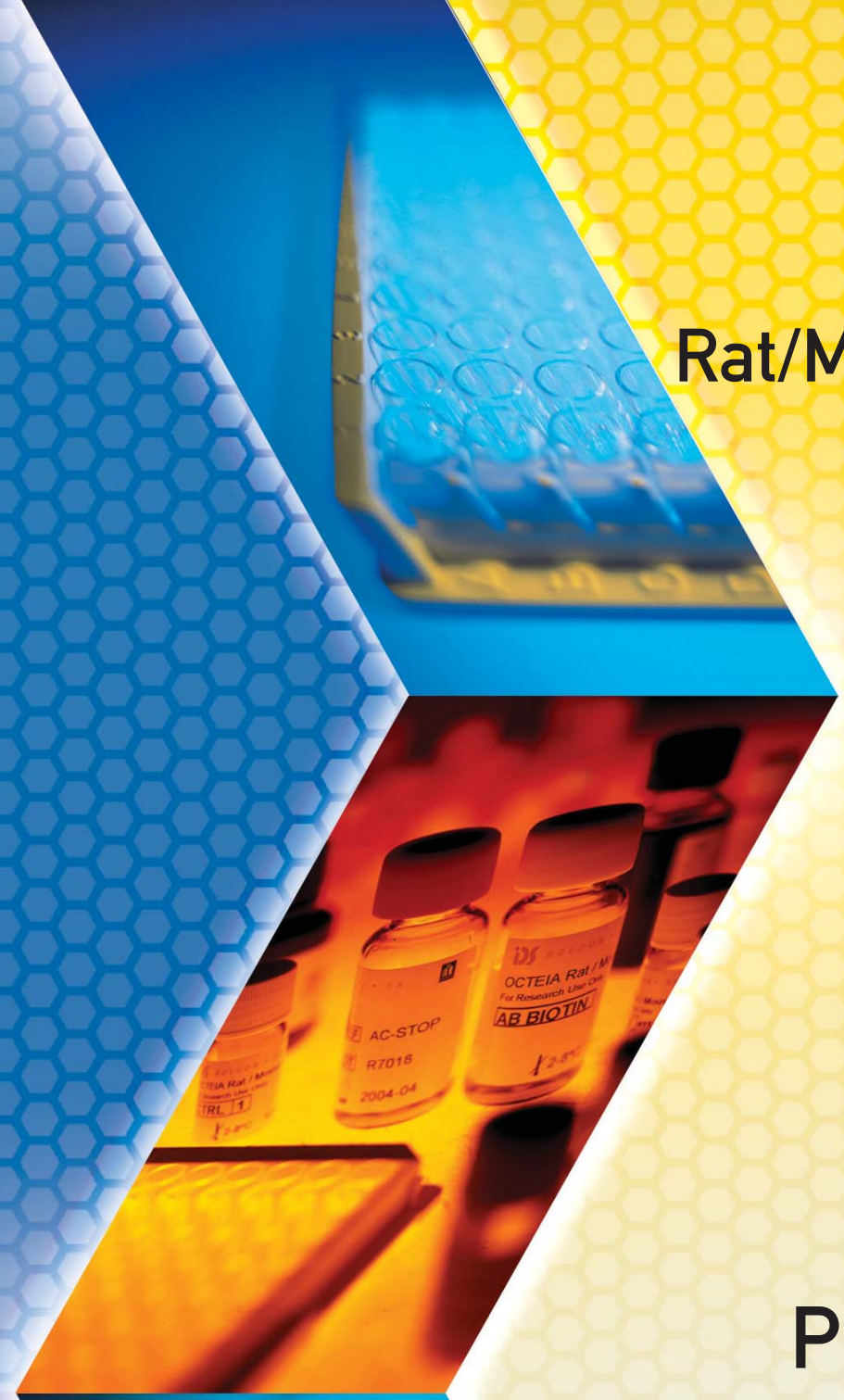


OCTEIA™ Rat/Mouse Insulin-like Growth Factor-I (IGF-I) ELISA*

Product Support

IDS

OCTEIA Rat/Mouse IGF-I

Catalogue Number AC-18F1

Features

Simple sample dilution eliminates the need for solvent extraction.

Superior precision
Intra assay <8% and inter-assay <13%.

Excellent Sensitivity
<82ng/mL.

Colorimetric detection eliminates the use of radioactivity. Provides long shelf life.

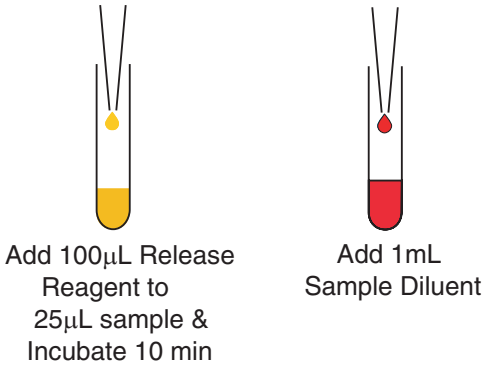
Specificity for both Rat and Mouse IGF-I.

Rapid results in under 4 hours with minimal hands-on time.

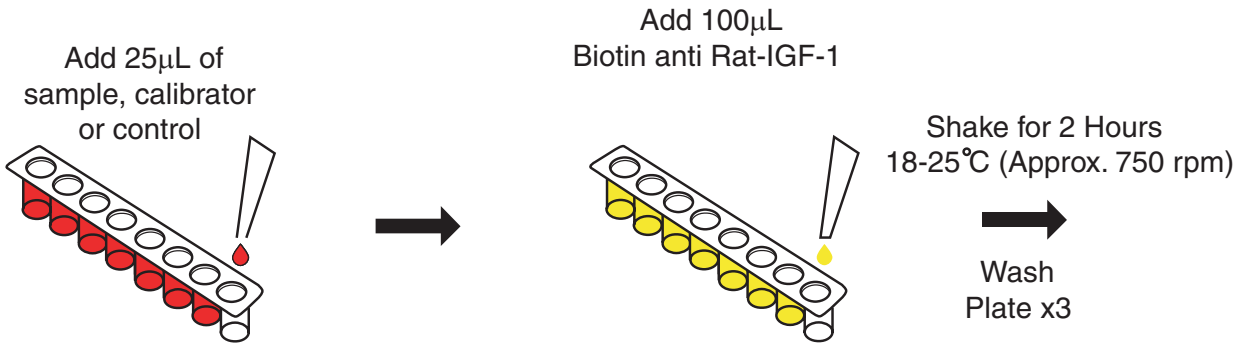
The Kit

The OCTEIA Rat/Mouse IGF-I kit is a two-site immunoenzymometric assay [IEMA] for the quantitative determination of Insulin-like Growth Factor I in rat and mouse serum or plasma. The method incorporates a sample pre-treatment to avoid interference from binding proteins.

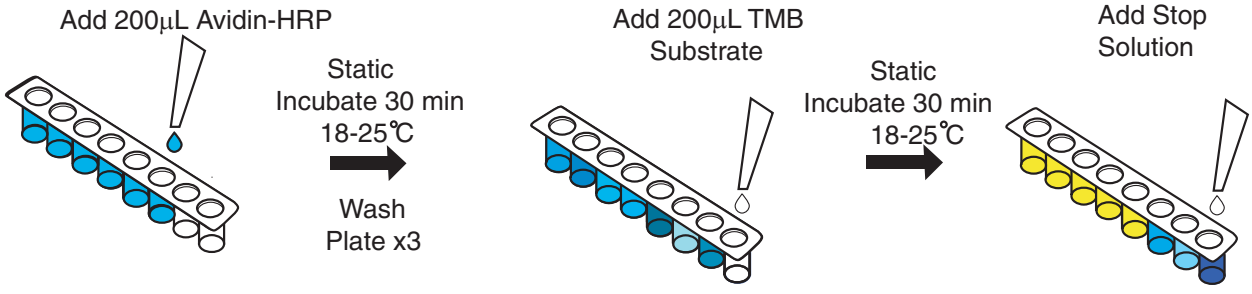
Assay Method



100µL of Release Reagent is added to 25µL of sample or kit control in appropriately labelled tubes. The tubes are vortex mixed and incubated for 10 minutes. 1mL of Sample Diluent is then added to each tube to dilute the sample ready for assay.



For the assay, 25µL of calibrator, or pre-treated sample or control is added to the wells of the anti-IGF-I antibody coated microtitre plate. 100µL of the anti-rat IGF-I biotin conjugate is added to all wells and the assay mixture is incubated shaking for 2 hours at room temperature after which the plate is washed to remove sample and unbound anti-rat IGF-I biotin conjugate.



Detection of bound anti-rat IGF-I biotin conjugate is accomplished by addition of 200µL Avidin HRP for 30 minutes after which the plate is washed.

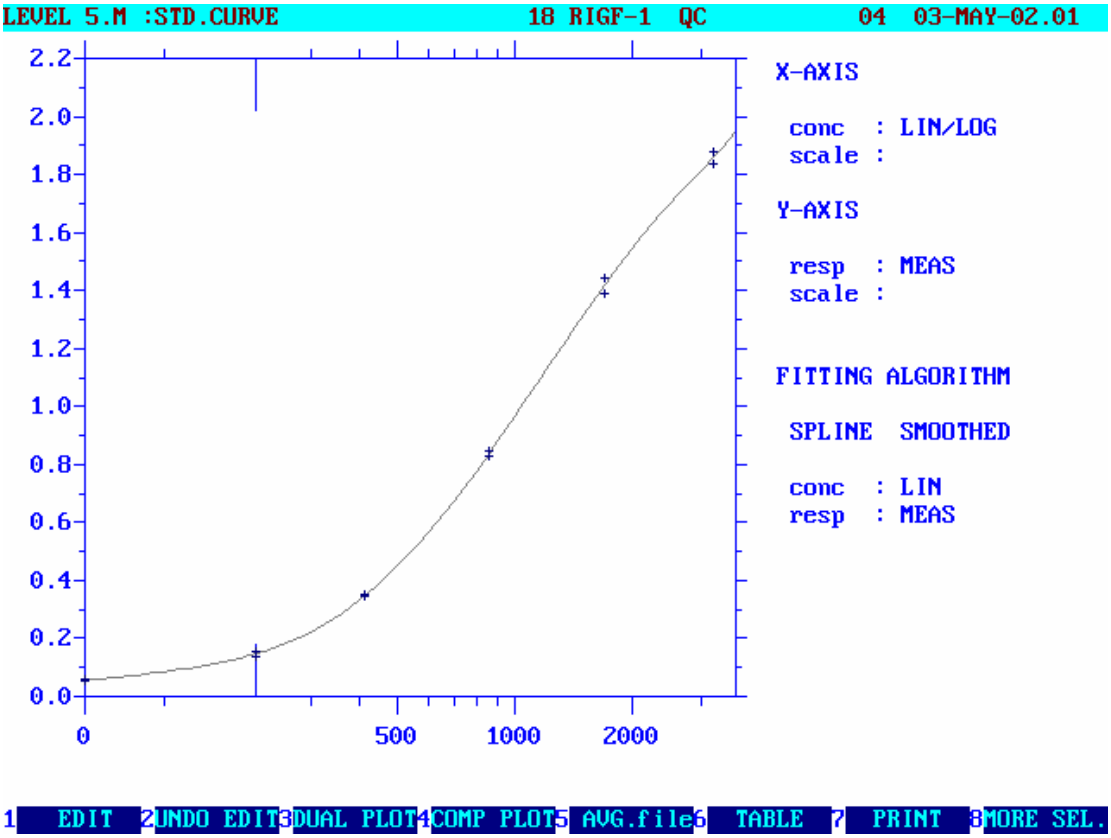
TMB substrate is added, incubated for 20 minutes and the reaction stopped with acid. The absorbance is proportional to the concentration of Rat/Mouse IGF-I. The calibration curve gives serum or plasma Rat/Mouse IGF-I values for unknowns directly.

Typical Calibration Curve

Example Data

Concentration	Absorbance	s.d.	%cv	B/B$_{max}$ (%)
0 **ng/mL**	0.054	0.006	10.5	2.6
217 **ng/mL**	0.142	0.014	10.0	6.9
411 **ng/mL**	0.357	0.017	4.8	17.4
857 **ng/mL**	0.785	0.040	5.0	38.2
1691 **ng/mL**	1.515	0.060	4.0	73.8
3204 **ng/mL**	2.053	0.117	5.7	100

The calibration curve below is an example. The data is for illustration only and must not be used for the calculation of results.



Sensitivity

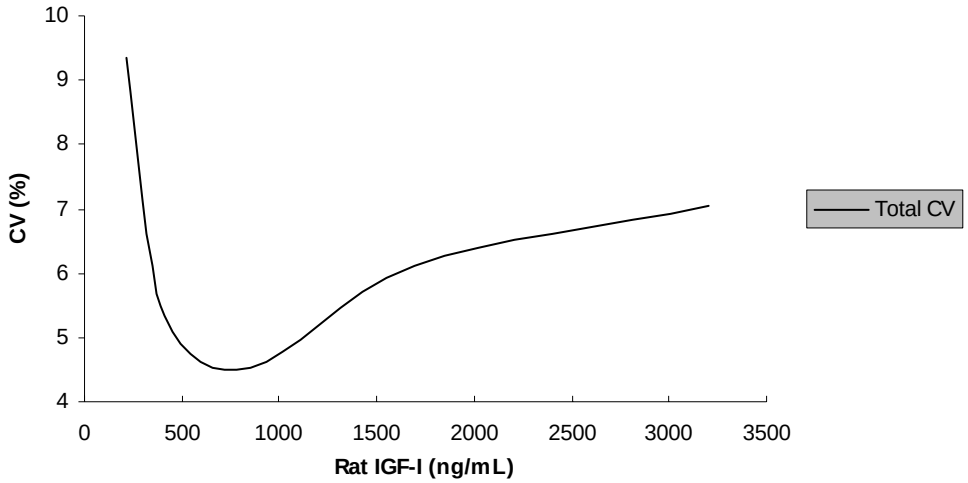
Sensitivity is defined as the concentration corresponding to the mean plus 2 standard deviations of 20 replicates of the zero calibrator.

Sensitivity 82ng/mL

Precision Profile

The profile is constructed from the imprecision of duplicates for calibrators from a total of 10 assays.

The graph below shows the calculated imprecision produced by "Analyse-it™ for Microsoft EXCEL™" software.



Rat IGF-I Concentration (ng/mL)	n	Total s.d.	Total cv
217	10	20.28	9.3%
411	10	21.93	5.3%
857	10	38.85	4.5%
1691	10	103.36	6.1%
3204	10	226.20	7.1%

The "Analyse-it™ for Microsoft EXCEL™" calculated precision range for CV <10% is 217 to 3204 ng/mL.

Within Assay Variation

Within assay variation was determined from duplicates of 10 sample dilutions of each sample, giving 20 assay replicates for each level.

Sample	Mean (ng/mL)	%cv	n
1	1385	5.1%	20
2	1560	5.5%	20
3	877	6.4%	20

Between Assay Variation

Between assay variation was determined from a panel of samples which were assayed using two batches of reagents over several assays.

Sample	Mean (ng/mL)	%CV	N
A	394.0	10.2%	9
B	1128.1	8.0%	9
C	421.4	10.1%	11
D	1091.3	6.2%	11
E	378.2	12.0%	11
F	1024.4	11.1%	11
G	1478.0	5.6%	15
H	1696.8	10.1%	15
I	398.5	12.4%	15
J	1038.0	7.2%	9

Linearity

Linearity was assessed by diluting high samples with low samples prior to assay.

Sample & Dilution:	Observed (ng/mL)	Expected (ng/mL)	Obs / Exp %
Rat Serum A	318.3		
0.750A + 0.250B	629.2	587.8	107%
0.500A + 0.500B	913.3	857.2	107%
0.250A + 0.750B	1261.0	1126.7	112%
Rat Plasma B	1396.1		
Rat Serum C	395.0		
0.750C + 0.250D	720.9	711.7	101%
0.500C + 0.500D	1025.6	1028.4	100%
0.250C + 0.750D	1282.8	1345.0	95%
Rat Serum D	1661.7		
Mouse Serum E	362.2		
0.750E + 0.250F	507.1	499.3	102%
0.500E + 0.500F	663.1	636.4	104%
0.250E + 0.750F	774.0	773.4	100%
Mouse Serum F	910.5		
L (R6426) Rat Plasma G	1016.1		
0.750G + 0.250H	1186.4	1149.3	103%
0.500G + 0.500H	1215.6	1282.4	95%
0.250G + 0.750H	1267.5	1415.6	90%
Rat Plasma H	1548.7		
L (R6246) Rat Plasma I	1024.6		
0.750I + 0.250J	1221.0	1164.7	105%
0.500I + 0.500J	1299.6	1304.9	100%
0.250I + 0.750J	1451.9	1445.0	100%
Rat Serum J	1585.1		
L (R6651) Mouse Plasma K	652.8		
0.750K + 0.250L	803.5	875.0	92%
0.500K + 0.500L	1007.4	1097.2	92%
0.250K + 0.750L	1194.4	1319.3	91%
Mouse Plasma L	1541.5		
		Mean	**100%**

Recovery

Recovery was assessed by adding rat IGF-I to samples prior to assay.

Sample	Sample conc. ng/mL	IGF-I added ng/mL	Measured ng/mL	Recovery ng/mL	Recovery %
1	430	400	853	423	106%
		800	1290	859	107%
2	733	400	1087	355	89%
		800	1519	786	98%
3	667	400	1113	446	112%
		800	1461	661	83%
4	271	400	731	460	115%
		800	1219	948	119%
5	383	400	835	452	113%
		800	1254	871	109%
6	667	400	1018	351	88%
		800	1468	801	100%
7	579	400	1049	470	118%
		800	1360	780	98%
8	243	400	689	446	112%
		800	1150	907	113%
				Mean	**105%**

Interferences

No interference from the following up to and including the following levels:

Rat BP4	8000ng/mL
Human IGF-II	2000ng/mL
Rat IGF-II	2000ng/mL
Bilirubin	300µg/mL
Lipid	20g/L
Haemoglobin	1470mg/dL

Specificity

Analyte	Cross-reactivity
Rat IGF-I	100%
Mouse IGF-I	100%
Rat IGF-II (9000ng/mL)	<0.1%
Human IGF-I (51885ng/mL)	<0.1%

References

1. *Daughaday E, Rotwein, P. Insulin like growth factor I and II. Peptide, messenger ribonucleic acid and gene structure, serum and tissue concentrations. Endocrin Rev 1989: 10: 68-91.*
2. *Baxter, RC, Martin JL, Beniac VA. High molecular weight insulin-like growth factor binding protein complex. J Biol Chem 1989: 264: 11843-11848.*
3. *Rechler M. Insulin-like growth factor binding proteins. Vit Horm 1993: 47: 1-114.*

Recommended Equipment

1. Disposable 12 x 75 mm plastic or glass tubes.
2. Precision pipetting devices to deliver 25 µL.
3. Repeating pipettes to deliver 100 µL and 1 mL, e.g. Eppendorf Multipipette 4780, or similar.
4. Precision multi-channel pipettes to deliver 100 µL and 200 µL.
5. Vortex mixer.
6. Automatic microplate washer (optional).
7. Photometric microplate reader and data analysis equipment.
8. Orbital plate shaker.

Related Products

OCTEIA	**Human IGF-I**	96 wells	AC-27F1

ORDERING INFORMATION

OCTEIA Rat/Mouse IGF-I Cat. No. AC-18F1 96 Wells

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Notes:

Notes:

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