abcam

Product datasheet

Human VEGF ELISA Kit ab222510

SimpleStep ELISA

★★★★★ 1 Abreviews 21 References 19 图像

概述

产品名称 人VEGF ELISA试剂盒

检测方法 Colorimetric

精确度

批次内

样品	n	Mean	SD	CV%
Supernatant	5			5.4%

批次间

样品	n	Mean	SD	CV%
Supernatant	3			5.5%

样品类型 Cell culture supernatant, Saliva, Milk, Urine, Serum, Cell culture extracts, Hep Plasma, EDTA Plasma, Cit plasma, Cerebral Spinal Fluid

检测类型 Sandwich (quantitative)

灵敏度 2.7 pg/ml

范围 12.5 pg/ml - 800 pg/ml

回收率

特定样本回收率

样品类型	平均%	范围
Saliva	109	105% - 118%
Milk	95	91% - 98%
Urine	95	91% - 102%
Serum	109	98% - 116%
Cell culture extracts	89	80% - 94%
Cell culture media	86	81% - 89%

样品类型	平均%	范围
Hep Plasma	89	84% - 92%
EDTA Plasma	98	95% - 99%
Cit plasma	101	95% - 107%
Cerebral Spinal Fluid	92	89% - 95%

检测时间 1h 30m

实验步骤 One step assay

种属反应性 **与反应:** Human
不与反应: Cow

产品概述 Human VEGF ELISA Kit (ab222510) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of VEGF protein in edta plasma, hep plasma, saliva, serum, urine, cell culture extracts, milk, cell culture supernatant, and cit plasma. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human VEGF with 2.7 pg/ml sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

- Single-wash protocol reduces assay time to 90 minutes or less
- High sensitivity, specificity and reproducibility from superior antibodies
- Fully validated in biological samples
- 96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (**ab203359**) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

INTERFERENCE

Serial dilutions of recombinant human VEGFR1, VEGFR2, and VEGFR3 were prepared starting at 4 ng/mL and tested for interference. No interference was observed.

SPECIES REACTIVITY

This kit recognizes human VEGF protein.

CALIBRATION

This immunoassay is calibrated against a highly purified human VEGF. The NIBSC/WHO unclassified purified human VEGF preparation 02/286 was evaluated in this kit.

The dose response curve of the unclassified standard 02/286 parallels the SimpleStep standard curve. To convert sample values obtained with the SimpleStep human VEGF kit to approximate NIBSC 02/286 units, use the equation below.

NIBSC 02/286 approximate value (IU/mL) = 0.0011 x SimpleStep human VEGF value (pg/mL).

说明

VEGF is a secreted growth factor of PDGF family active in angiogenesis, vasculogenesis and endothelial cell growth both in fetus and adult. Alternative splicing produces many isoforms including major isoforms VEGF121, VEGF165 and VEGF189 in human. VEGF expression is induced by hypoxia. It is regulated by growth factors, cytokines, gonadotropins, nitric oxide, hypoglycemia and oncogenic mutations. VEGF induces endothelial cell proliferation, promotes cell migration, inhibits apoptosis and induces permeabilization of blood vessels. VEGF dimers bind to the FLT1/VEGFR1 and KDR/VEGFR2 receptors, induce their homodimerization and autophosphorylation. VEGF165 and VEGF145 interact with NRP1/Neuropilin.

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances.
It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.

平台

Pre-coated microplate (12 x 8 well strips)

性能

存放说明

Store at +4°C. Please refer to protocols.

组件	1 x 96 tests
10X Human VEGF Capture Antibody	1 x 600µl
10X Human VEGF Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
50X Cell Extraction Enhancer Solution (ab193971)	1 x 1ml
5X Cell Extraction Buffer PTR (ab193970)	1 x 10ml
Antibody Diluent 4BI	1 x 6ml
Human VEGF Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent NS (ab193972)	1 x 50ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit

组件	1 x 96 tests
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

功能 Growth factor active in angiogenesis, vasculogenesis and endothelial cell growth. Induces endothelial cell proliferation, promotes cell migration, inhibits apoptosis and induces permeabilization of blood vessels. Binds to the FLT1/VEGFR1 and KDR/VEGFR2 receptors, heparan sulfate and heparin. NRP1/Neuropilin-1 binds isoforms VEGF-165 and VEGF-145. Isoform VEGF165B binds to KDR but does not activate downstream signaling pathways, does not activate angiogenesis and inhibits tumor growth.

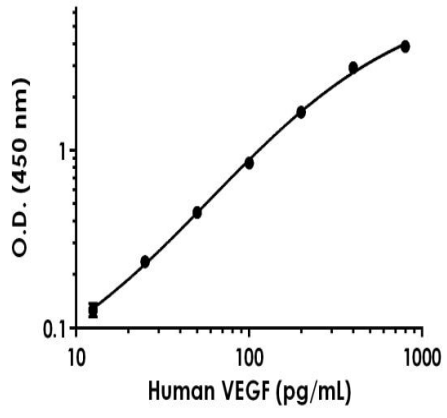
组织特异性 Isoform VEGF189, isoform VEGF165 and isoform VEGF121 are widely expressed. Isoform VEGF206 and isoform VEGF145 are not widely expressed.

疾病相关 Defects in VEGFA are a cause of susceptibility to microvascular complications of diabetes type 1 (MVCD1) [MIM:603933]. These are pathological conditions that develop in numerous tissues and organs as a consequence of diabetes mellitus. They include diabetic retinopathy, diabetic nephropathy leading to end-stage renal disease, and diabetic neuropathy. Diabetic retinopathy remains the major cause of new-onset blindness among diabetic adults. It is characterized by vascular permeability and increased tissue ischemia and angiogenesis.

序列相似性 Belongs to the PDGF/VEGF growth factor family.

细胞定位 Secreted. VEGF121 is acidic and freely secreted. VEGF165 is more basic, has heparin-binding properties and, although a signicant proportion remains cell-associated, most is freely secreted. VEGF189 is very basic, it is cell-associated after secretion and is bound avidly by heparin and the extracellular matrix, although it may be released as a soluble form by heparin, heparinase or plasmin.

图片



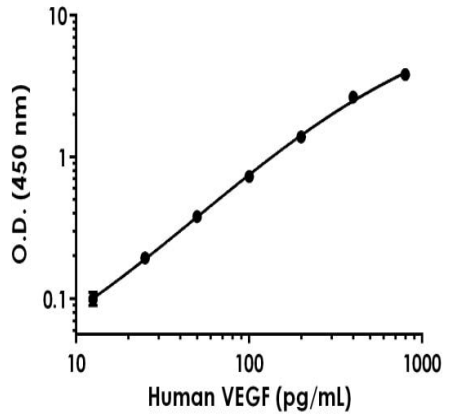
Example of human VEGF standard curve in Sample Diluent NS + 1X Enhancer.

The VEGF standard curve was prepared as described in Section 10. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.

Standard Curve Measurements			
Concentration (pg/mL)	O.D 450 nm		Mean O.D
	1	2	
0	0.104	0.100	0.102
12.5	0.236	0.221	0.229
25	0.345	0.332	0.338
50	0.558	0.540	0.549
100	0.962	0.940	0.951
200	1.740	1.750	1.745
400	3.068	2.987	3.027
800	3.959	3.954	3.957

Example of human VEGF standard curve in Sample Diluent NS + 1X Enhancer.

The VEGF standard curve was prepared as described. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.



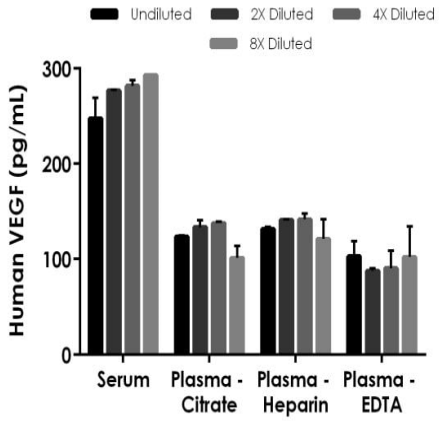
Example of human VEGF standard curve in 1X Cell Extraction Buffer PTR.

The VEGF standard curve was prepared as described in Section 10. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.

Standard Curve Measurements			
Concentration (pg/mL)	O.D 450 nm		Mean O.D
	1	2	
0	0.101	0.099	0.100
12.5	0.207	0.192	0.199
25	0.304	0.284	0.294
50	0.495	0.467	0.481
100	0.853	0.805	0.829
200	1.547	1.425	1.486
400	2.879	2.634	2.757
800	3.962	3.895	3.928

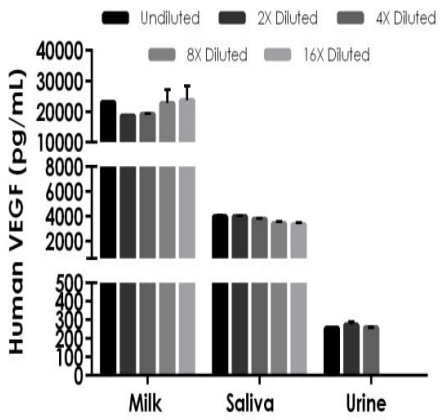
Example of human VEGF standard curve in 1X Cell Extraction Buffer PTR.

The VEGF standard curve was prepared as described. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.



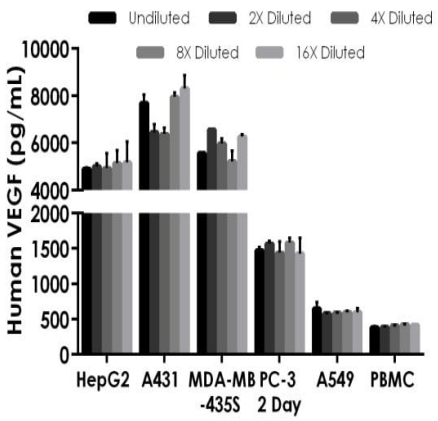
Interpolated concentrations of native VEGF in human serum and plasma samples.

The concentrations of VEGF were measured in duplicates, interpolated from the VEGF standard curves and corrected for sample dilution. Undiluted samples are as follows: serum 50%, plasma (citrate) 50%, plasma (heparin) 50% and plasma (EDTA) 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean VEGF concentration was determined to be 268.9 pg/mL in neat serum, 123.9 pg/mL in neat plasma (citrate), 133.8 pg/mL in neat plasma (heparin), and 95.76 pg/mL in neat plasma (EDTA).



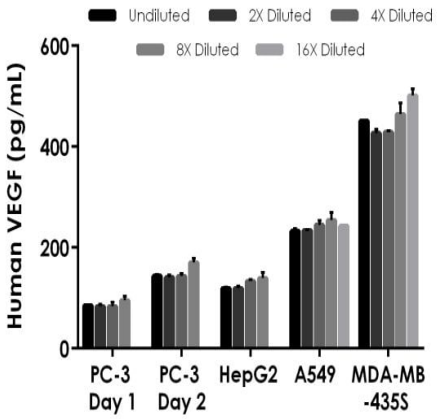
Interpolated concentrations of native VEGF in human breast milk (de-fatted), saliva, and urine samples.

The concentrations of VEGF were measured in duplicates, interpolated from the VEGF standard curves and corrected for sample dilution. Undiluted samples are as follows: breast milk 2%, saliva 12.5%, and urine 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean VEGF concentration was determined to be 21487 pg/mL in neat human breast milk (de-fatted), 3712 pg/mL in neat human saliva, and 262.0 pg/mL in neat human urine.



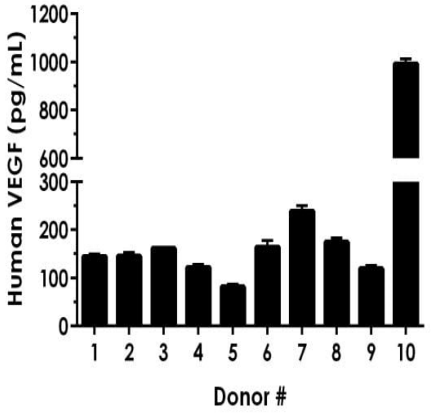
Interpolated concentrations of native VEGF in cell culture supernatant samples.

The concentrations of VEGF were measured in duplicates, interpolated from the VEGF standard curves and corrected for sample dilution. Undiluted samples are as follows: HepG2 5%, A431 5%, MDA-MB-435S 12.5%, PC-3 20%, A549 50%, and PBMC 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean VEGF concentration was determined to be 5027 pg/mL in neat HepG2 supernatant, 7356 pg/mL in neat A431 supernatant (4 Day), 5918 pg/mL in neat MDA-MB-435S supernatant, 1495 pg/mL in neat PC-3 (2 Day), 603.4 pg/mL in neat A549 supernatant, and 405.3 pg/mL in neat PBMC supernatant (PHA-M, 5 Day).



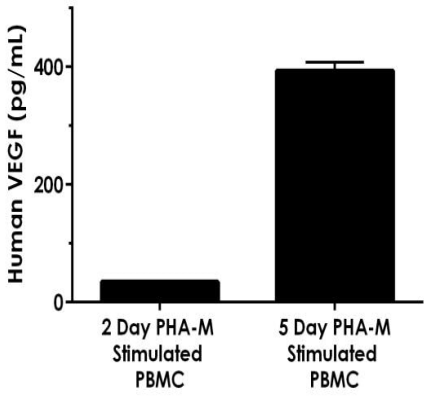
Interpolated concentrations of native VEGF.

Interpolated concentrations of native VEGF in PC-3 cell extract (1 Day), PC-3 cell extract (2 Day), HepG2 cell extract, A549 cell extract, and MDA-MB-435S cell extract samples based on a 300 µg/mL extract load. The concentrations of VEGF were measured in duplicate and interpolated from the VEGF standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean VEGF concentration was determined to be 86.65 pg/mL in PC-3 cell extract (1 Day), 149.6 pg/mL in PC-3 cell extract (2 Day), 127.7 pg/mL in HepG2 cell extract, 241.7 pg/mL in A549 cell extract, and 454.5 pg/mL in MDA-MB-435S cell extract.



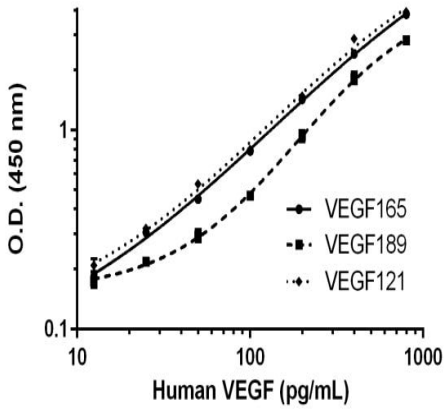
Serum from ten individual healthy human female donors was measured in duplicate.

Interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean VEGF concentration was determined to be 235.0 pg/mL with a range of 81.10 – 993.3 pg/mL.



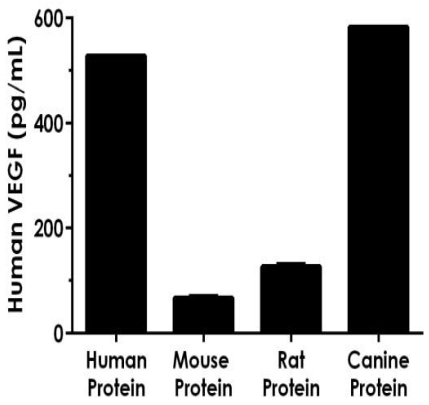
Comparison of VEGF in PHA-M stimulated and unstimulated human PBMC cell culture supernatants stimulated for different durations.

Human PBMC (seeded at $10x10^6$/mL) were cultured for 2 or 5 days in the presence or absence of 1.5% PHA-M. The concentrations of VEGF were measured in three different dilutions of the supernatant samples in duplicates and interpolated from the VEGF standard curve. The interpolated values are plotted (mean +/- SD, n=3). The mean VEGF concentration was determined to be 34.76 pg/mL in 2 Day PHA-M stimulated PBMC cell culture supernatant, 393.7 pg/mL in 5 Day PHA-M stimulated PBMC cell culture supernatant, and undetectable in unstimulated PBMC cell culture supernatant at both 2 and 5 days (not shown).



Serial dilutions of recombinant human VEGF189 and VEGF121.

Serial dilutions of recombinant human VEGF189 and VEGF121 were prepared and assayed in parallel with recombinant VEGF165.



Other species reactivity was determined by measuring a 500 pg/mL protein load of various species recombinant VEGF proteins.

Protein concentrations were interpolated from the human standard curve, and graphing the interpolated concentrations (mean +/- SD, n=2).

Dilution Factor	Interpolated value	50% Human Serum	50% Human Plasma (Citrate)	50% Human Plasma (EDTA)	50% Human Plasma (Heparin)
Undiluted	pg/mL	692.8	698.8	675.6	665.3
	% Expected value	100	100	100	100
2	pg/mL	365.0	366.1	353.1	332.2
	% Expected value	105	105	105	100
4	pg/mL	166.5	179.0	168.8	166.3
	% Expected value	96	102	100	100
8	pg/mL	75.20	72.75	67.96	80.28
	% Expected value	87	83	80	97
16	pg/mL	34.83	34.80	38.87	41.28
	% Expected value	80	80	92	99

Linearity of dilution.

Linearity of dilution is determined based on interpolated values from the standard curve. Linearity of dilution defines a sample concentration interval in which interpolated target concentrations are directly proportional to sample dilution.

Native VEGF was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in Sample Diluent NS + 1X Enhancer.

Recombinant human VEGF was spiked into the following biological samples and diluted in a 2-fold dilution series in Sample Diluent NS + 2X Enhancer and in Sample Diluent NS + 1X Enhancer, see Sample Preparation section for details.

Dilution Factor	Interpolated value	2% Human Breast Milk	12.5% Human Saliva	50% Human Urine	20% PC-3 (1 Day) SN	20% PC-3 (2 Day) SN
Undiluted	pg/mL	461.4	497.3	127.4	123.8	294.4
	% Expected value	100	100	100	100	100
2	pg/mL	187.1	250.2	68.12	69.22	156.5
	% Expected value	81	101	107	112	106
4	pg/mL	96.03	118.3	32.34	35.23	71.87
	% Expected value	83	95	102	114	98
8	pg/mL	56.86	53.86	ND	18.31	39.46
	% Expected value	99	87	ND	118	107
16	pg/mL	29.63	26.14	ND	ND	17.81
	% Expected value	103	84	ND	ND	97

ND – Non-Detectable. O.D. values below the 7th point of the standard curve.

Linearity of dilution.

Native VEGF was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in Sample Diluent NS + 2X Enhancer and in Sample Diluent NS + 1X Enhancer, see Sample Preparation section for details. Cell culture supernatants are represented by SN.

Dilution Factor	Interpolated value	50% A549 SN	12.5% MDA-MB-435S SN	5% HepG2 SN	5% A431 SN	50% PHA-M PBMC SN (5 Day)
Undiluted	pg/mL	325.8	696.7	245.2	384.6	191.2
	% Expected value	100	100	100	100	100
2	pg/mL	145.3	409.8	125.1	161.1	97.32
	% Expected value	89	118	102	84	102
4	pg/mL	73.49	186.7	61.51	79.70	51.17
	% Expected value	90	107	100	83	107
8	pg/mL	37.53	81.58	32.11	49.70	26.66
	% Expected value	92	94	105	103	112
16	pg/mL	18.61	48.93	16.16	25.98	13.10
	% Expected value	91	112	105	108	110

Linearity of dilution.

Native VEGF was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in Sample Diluent NS + 2X Enhancer and in Sample Diluent NS + 1X Enhancer, see Sample Preparation section for details. Cell culture supernatants are represented by SN.

Dilution Factor	Interpolated value	300 µg/mL PC-3 Cell Extract (1 Day)	300 µg/mL PC-3 Cell Extract (2 Day)	300 µg/mL HepG2 Cell Extract	300 µg/mL A549 Cell Extract	300 µg/mL MDA-MB-435S Cell Extract
Undiluted	pg/mL	85.16	143.9	119.0	232.5	451.4
	% Expected value	100	100	100	100	100
2	pg/mL	41.55	70.46	59.64	117.1	213.3
	% Expected value	98	98	100	101	95
4	pg/mL	20.77	35.78	33.36	61.19	107.2
	% Expected value	98	99	112	105	95
8	pg/mL	11.90	21.31	17.39	31.75	58.03
	% Expected value	112	118	117	109	103
16	pg/mL	ND	ND	ND	15.19	31.34
	% Expected value	ND	ND	ND	105	111

ND – Non-Detectable. O.D. values below the 7th point of the standard curve.

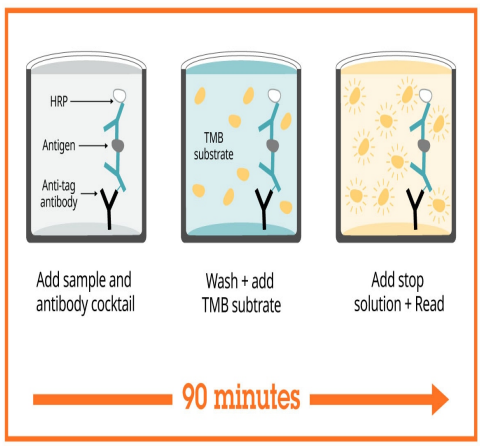
Linearity of dilution.

Native VEGF was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in 1X Cell Extraction Buffer PTR.

Sample Diluent Buffer	n=	Minimal Detectable Dose
1X Cell Extraction Buffer PTR	10	2.9 pg/mL
Sample Diluent NS + 1X Enhancer	14	2.7 pg/mL

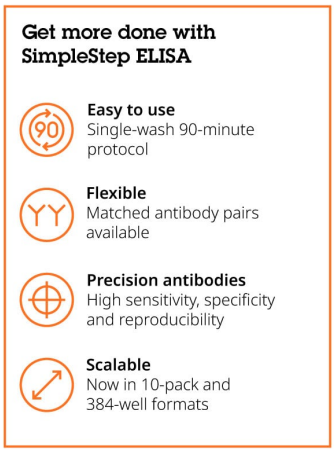
Assay sensitivity.

The MDD was determined by calculating the mean of zero standard replicates and adding 2 standard deviations then extrapolating the corresponding concentration.



Sandwich ELISA - Human VEGF ELISA Kit (ab222510)

SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.



Sandwich ELISA - Human VEGF ELISA Kit (ab222510)

To learn more about the advantages of SimpleStep ELISA® kits see **here**.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

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- Response to your inquiry within 24 hours

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- We investigate all quality concerns to ensure our products perform to the highest standards

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