

# U-PLEX<sup>®</sup> NHP Eotaxin-3 Assay



[www.mesoscale.com](http://www.mesoscale.com)<sup>®</sup>

## Ordering Information

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## Scientific Support

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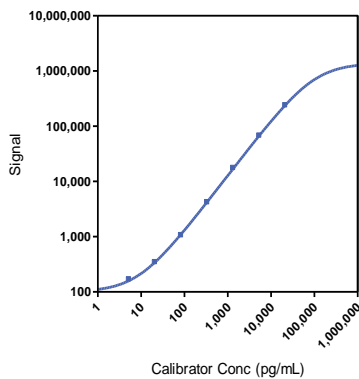
## Company Address

MESO SCALE DISCOVERY<sup>®</sup>  
A division of  
Meso Scale Diagnostics, LLC.  
1601 Research Boulevard  
Rockville, MD 20850-3173 USA

	Available as part of U-PLEX Biomarker Group 1 (NHP) multiplex combination: K15068L-1/-2/-4
<b>Product Options</b>	Individual assay: K156UEK-1/-2/-4; Antibody Set: B26UE-2/B26UE-3 For more ordering options, please visit <a href="http://www.mesoscale.com">www.mesoscale.com</a>
<b>Instrument Compatibility</b>	SECTOR <sup>®</sup> Imager 2400, SECTOR Imager 6000, MESO <sup>®</sup> SECTOR S 600, MESO QuickPlex <sup>®</sup> SQ 120
<b>Sample Type</b>	NHP (Cynomolgus monkey and Rhesus monkey) serum, EDTA plasma, and cell culture supernatants
<b>Assay Protocol</b>	Refer to the U-PLEX Biomarker Group 1 (NHP) product insert available at <a href="http://www.mesoscale.com">www.mesoscale.com</a>

The U-PLEX platform was designed to provide ultimate flexibility for detection of biomarkers in a wide variety of sample types. This datasheet provides the representative performance of the NHP Eotaxin-3 Assay tested on U-PLEX plates run as a multiplex. The data were generated during the development of the assay and do not represent the product specifications. Under your experimental conditions and with your specific multiplex, the assay may perform differently than the representative data shown. U-PLEX assays are available in multiplex format with other compatible assays. The same assay can also be used to detect a single analyte on MSD GOLD<sup>™</sup> Small Spot Streptavidin plates.

## Representative Calibration Curve and Sensitivity



Assay	Median LLOD (pg/mL)	LLOD Range (pg/mL)
Eotaxin-3	7.3	5.9-9.1

The calibration curves used to calculate analyte concentrations were established by fitting the signals from the Calibrators using a 4-parameter logistic (or sigmoidal dose-response) model with a  $1/Y^2$  weighting. Analyte concentrations were determined from the electrochemiluminescence signals by back-fitting to the calibration curve. The limit of detection (LLOD) is a calculated concentration corresponding to the signal 2.5 standard deviations above the background (zero Calibrator).

## Precision

	Control	Average Conc. (pg/mL)	Average Intra-run Conc. %CV	Inter-run Conc. %CV
Eotaxin-3	High	9,355	9.4	18.5
	Mid	914	7.2	18.1
	Low	95	7.5	14.9

**For Research Use Only.**  
**Not for use in diagnostic procedures.**

Controls were made by spiking Calibrator into assay diluent at 3 levels within the quantitative range of the assay. Average intra-run concentration CV is the average %CV of the control replicates within an individual run. Inter-run concentration %CV is the variability of controls across multiple runs.

# MSD® U-PLEX Assays

## Spike Recovery

	Spike Level	Serum		Plasma		Cell Culture Media	
		Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range	Average % Recovery	% Recovery Range
Cynomolgus Monkey	High	110	105-116	112	89-158	125	118-133
	Mid	105	100-111	108	61-160	128	101-142
	Low	103	97-111	106	54-170	128	95-145
Rhesus Monkey	High	137	116-150	139	131-146	125	118-133
	Mid	132	114-144	139	127-145	128	101-142
	Low	133	110-142	133	122-140	128	95-145

Normal serum, EDTA plasma, and cell culture media were spiked with Calibrators at 3 levels. Undiluted samples were tested to determine the expected concentration of the analyte. Samples may require additional dilution with assay diluent to reduce matrix effects.

% Recovery = (measured concentration / expected concentration) x 100

## Tested Samples

	Sample Type	Serum	Plasma	Spiked Serum	Spiked Plasma
Cynomolgus Monkey	Median (pg/mL)	ND	NA	811	760
	Range (pg/mL)	ND-214	NA	773-914	726-816
	% Detected	10	0	100	100
Rhesus Monkey	Median (pg/mL)	ND	ND	586	493
	Range (pg/mL)	ND-19	ND-13	572-833	427-786
	% Detected	10	10	100	100

ND = non-detectable (< LLOD), NA = not applicable due to 0% detected

Normal serum and EDTA plasma samples were tested without dilution prior to the assay. Spiked serum and spiked plasma represent samples that were spiked with Calibrator and/or cell culture supernatants derived from cells (PBMCs and THP-1) stimulated with different compounds in vitro.

## Dilution Linearity

	Serum			Plasma			Cell Culture Media		
	Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range	Fold Dilution	Average % Recovery	% Recovery Range
Cynomolgus Monkey	2	91	86-95	2	99	89-109	2	92	86-99
	4	90	83-96	4	95	86-112	4	87	80-91
	8	88	81-93	8	95	84-116	8	81	74-87
Rhesus Monkey	2	111	109-114	2	88	81-94	2	92	86-99
	4	105	99-111	4	85	82-88	4	87	80-91
	8	110	105-118	8	79	77-83	8	81	74-87

Normal serum, EDTA plasma, and cell culture media were spiked with recombinant Calibrator and tested at different dilutions. Undiluted samples were tested to determine the expected concentration of the analyte. Samples may require additional dilution with assay diluent to reduce matrix effects.

% Recovery = (measured concentration / expected concentration) x 100

# MSD U-PLEX Assays

## Specificity

To assess specificity, Eotaxin-3 Antibody Set was tested individually against a larger panel of recombinant nhp analytes for nonspecific binding (CTACK, ENA-78, Eotaxin, Eotaxin-3, Fractalkine, G-CSF, GM-CSF, IFN- $\alpha$ 2a, IFN- $\gamma$ , IL-1 $\alpha$ , IL-1 $\beta$ , IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12/IL-23p40, IL-12p70, IL-13, IL-15, IL-16, IL-17A, IL-18, IP-10, I-TAC, MCP-1, MCP-4, MDC, MIP-1 $\alpha$ , MIP-1 $\beta$ , MIP-3 $\alpha$ , MIP-3 $\beta$ , SDF-1 $\alpha$ , TARC, TNF- $\alpha$ , TNF- $\beta$ , TPO, and VEGF-A). Nonspecific binding was less than 0.5%.

$\% \text{ Nonspecificity} = (\text{nonspecific signal} / \text{specific signal}) \times 100$

## Diluent Compatibility

The data included in this document has been collected using Diluents 3 and 43. MSD offers a range of assay and antibody diluents for separate purchase. Depending on your assay needs, other diluents may be tested.

## Assay Components

**Calibrator:** NHP Eotaxin-3 is included in Calibrator 2 blend. The full-length recombinant protein expressed in *E. coli* is used.

**Antibodies:** The U-PLEX NHP Eotaxin-3 Assay uses mouse monoclonal antibody for capture and mouse monoclonal antibody for detection.

**Assay generation:** B

Note: This datasheet contains representative assay performance data. In custom multiplex formats, the assay may perform differently than the representative data shown.

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