



## Protein Standards for Mouse Inflammation ELISA Strip

Catalog Number EA-1052

(For Research Use Only)

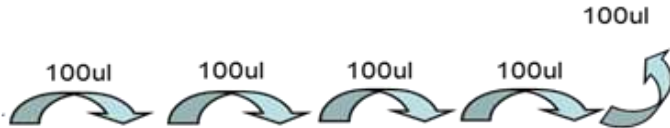
<u>Tube</u>	<u>Protein</u>	<u>Stock Conc.</u>	<u>Volume</u>
#1	Mouse TNF $\alpha$	400ng/ml	10ul
#2	Mouse IL-1 $\beta$	400ng/ml	10ul
#3	Mouse IFN $\gamma$	400ng/ml	10ul
#4	Mouse IL-6	400ng/ml	10ul
#5	Mouse IGF-1	400ng/ml	10ul
#6	Mouse MCP-1	400ng/ml	10ul
#7	Mouse Leptin	400ng/ml	10ul
#8	Mouse TGF $\beta$	400ng/ml	10ul

### Preparation of protein standard dilutions

1. Add 200ul of Diluent buffer the wells of the first strip, and add 100ul of Diluent buffer to the wells of the rest strips according to the following table.
2. Add appropriate amount of protein standards to the first strip according to the table 1.
3. Use multi-channel pipette to mix the dilutions and transfer 100ul to the next dilution wells.
4. Repeat the transfer until 5<sup>th</sup> strip. Trash 100ul from 5<sup>th</sup> strip after mixing. Do not transfer to the 6<sup>th</sup> Strip

**Note: Substrate incubation time may vary due to different antibodies reactivity. Stronger signals (Strong blue color) could be stopped early after 5 minutes. Weaker signals should be incubated for 10-30 minutes. Because MIP-1a has a weaker signal than other cytokines' signals, it may require longer incubation time. Always stop protein standards along with samples from the same row at the same time.**

**Table 1: Dilution of protein standards**



<u>Protein</u>	<u>Added to 1st Strip</u>	<u>1st Strip</u>	<u>2nd Strip</u>	<u>3rd Strip</u>	<u>4th Strip</u>	<u>5th Strip</u>	<u>6th Strip</u>
Mouse TNF $\alpha$	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-1 $\beta$	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IFN $\gamma$	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-6	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IGF-1	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse MCP-1	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse Leptin	4ul	200ul	100ul	100ul	100ul	100ul	100ul
Mouse TGF $\beta$	4ul	200ul	100ul	100ul	100ul	100ul	100ul
			<b>1:2</b>	<b>1:4</b>	<b>1:8</b>	<b>1:16</b>	<b>Blank</b>

**Table 2: Concentrations of Protein Standard Dilutions**

<b><u>Protein</u></b>		<b><u>1:2</u></b>	<b><u>1:4</u></b>	<b><u>1:8</u></b>	<b><u>1:16</u></b>	<b><u>Blank</u></b>
Mouse TNF $\alpha$	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IL-1 $\beta$	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IFN $\gamma$	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IL-6	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IGF-1	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse MCP-1	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse Leptin	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse TGF $\beta$	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank