

## Protein Standards for Mouse Oxidative Stress ELISA Strip

Catalog Number EA-1402

(For Research Use Only)

100ul

<u>Tube</u>	<u>Protein</u>	Stock Conc.	<u>Volume</u>
#1	Mouse TNFa	400ng/ml	10ul
#2	Mouse TGFβ	400ng/ml	10ul
#3	Mouse MCP-1	400ng/ml	10ul
#4	Mouse IL-1α	400ng/ml	10ul
#5	Mouse IL-1β	400ng/ml	10ul
#6	Mouse IL-6	400ng/ml	10ul
#7	Mouse IL-10	400ng/ml	10ul
#8	Mouse IL-12	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: For colorimetric kits, 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. Always stop protein standards along with samples from the same row at the same time.

Table 1: Dilution of protein standards

		10	Oul 1	ooul 1	100ul	100ul	5
<u>Protein</u>	Added to 1st Strip	1st Strip	2nd Strip	3rd Strip	4th Strip	5th Strip	6th Strip
Mouse TNFα	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse TGF-β	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse MCP-1	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-1α	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-1β	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-6	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-10	4µl	200ul	100ul	100ul	100ul	100ul	100ul
Mouse IL-12	4µl	200ul	100ul	100ul	100ul	100ul	100ul
			1:2	1:4	1:8	1:16	Blank

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

<u>Protein</u>	Added to 1st Strip	<u>1:2</u>	<u>1:4</u>	<u>1:8</u>	<u>1:16</u>	<u>Blank</u>
Mouse TNFα	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse TGF-β	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 IL-1α	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IL-1β	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 IL-10	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank
Mouse IL-12	8ng/ml	4ng/ml	2ng/ml	1ng/ml	0.5ng/ml	Blank