

**MOUSE INTERLEUKIN-1 $\beta$  ASSAY LAYOUT SHEET**  
for use with Assay Designs' Catalog No. 900-132

• **TABLE FOR MAKING STANDARD 1-4:**

Reconstitute lyophilized standard with deionized water or Tissue Culture Media. Reconstitution volume is stated on the standard vial label. Let sit 5 minutes, then mix gently.

Std.	Diluent Vol. ( $\mu$ L)	Vol Added ( $\mu$ L)	mouse IL-1 $\beta$ Conc. (pg/mL)
1	300	100, Stock	1,000
2	300	100, Std 1	250
3	300	100, Std 2	62.5
4	300	100, Std 3	15.6

• **ASSAY PROTOCOL FLOW CHART:**

Well ID:	Blank A1, B1	S 0 C1, D1	Standards E1-D2	Samples E2-H12
Antibody	---	50 $\mu$ L	50 $\mu$ L	50 $\mu$ L
Standard Diluent	---	50 $\mu$ L	---	---
Std. and/or Sample	---	---	50 $\mu$ L	50 $\mu$ L
Tap plate gently to mix	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Incub. 2 hours @ RT, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 3 x 400 $\mu$ L	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Conjugate	---	100 $\mu$ L	100 $\mu$ L	100 $\mu$ L
Incub. 30 min. @ RT, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 3 x 400 $\mu$ L	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
TMB Substrate	100 $\mu$ L	100 $\mu$ L	100 $\mu$ L	100 $\mu$ L
Incub. 30 min. @ RT, in the dark	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Stop Solution	100 $\mu$ L	100 $\mu$ L	100 $\mu$ L	100 $\mu$ L

**MOUSE IL-1 $\beta$  PLATE LAYOUT:**

A1 Blank	A2 Std 3	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12
B1 Blank	B2 Std 3	B3	B4	B5	B6	B7	B8	B9	B10	B11	B12
C1 S0	C2 Std 4	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12
D1 S0	D2 Std 4	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12
E1 Std 1	E2	E3	E4	E5	E6	E7	E8	E9	E10	E11	E12
F1 Std 1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
G1 Std 2	G2	G3	G4	G5	G6	G7	G8	G9	G10	G11	G12
H1 Std 2	H2	H3	H4	H5	H6	H7	H8	H9	H10	H11	H12

Kit Lot No. \_\_\_\_\_ Exp. Date \_\_\_\_\_ Date \_\_\_\_\_ Tech. \_\_\_\_\_

1<sup>st</sup> Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 End Time \_\_\_\_\_ Temp. \_\_\_\_\_

3<sup>rd</sup> Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 End Time \_\_\_\_\_ Temp. \_\_\_\_\_

2<sup>nd</sup> Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 End Time \_\_\_\_\_ Temp. \_\_\_\_\_