

HUMAN INTERLEUKIN-4 ASSAY LAYOUT SHEET
for use with Assay Designs' Catalog No. 900-145

• **TABLE FOR MAKING STANDARD 1-5:**

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 (μL)	Vol. (μL)	Vol Added (μL)	human IL-4 Conc. (pg/mL)
1	240		160, Stock	400
2	240		160, Std 1	160
3	240		160, Std 2	64
4	240		160, Std 3	25.6
5	240		160, Std 4	10.24

• **ASSAY PROTOCOL FLOW CHART:**

Well ID:	Blank A1, B1	S0 C1, D1	Standards E1-F2	Samples G2-H12
Antibody	---	50 μL	50 μL	50 μL
Standard Diluent	---	50 μL	---	---
Std. and/or Sample	---	---	50 μL	50 μL
Incub. 2 hours @ RT, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 3 x 400 μL	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Prepared Streptavidin Solution	---	100 μL	100 μL	100 μL
Incub. 30 min. @ RT, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 3 x 400 μL	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
TMB Substrate	100 μL	100 μL	100 μL	100 μL
Incub. 1 hour @ RT, in the dark	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Stop Solution	100 μL	100 μL	100 μL	100 μL
Read at 450 nm	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒

HUMAN IL-4 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 Std 5	E3	E4	E5	E6	E7	E8	E9	E10	E11	E12
F1 Std 1	F2 Std 5	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. _____

1st Incub.: Start Time _____ Temp. _____

End Time _____ Temp. _____

3rd Incub.: Start Time _____ Temp. _____

End Time _____ Temp. _____

2nd Incub.: Start Time _____ Temp. _____

End Time _____ Temp. _____