

**CRP (human) ELISA ASSAY LAYOUT SHEET**  
for use with Enzo Life Sciences' Catalog No. ENZ-KIT102-0001

• **TABLE FOR MAKING STANDARDS 1-5:**

Std.	AB42 Vol. (µL)	Standard Vol. Added (µL)	CRP Conc. (ng /mL)
1	520	80, stock	810
2	300	150, Std. 1	270
3	300	150, Std. 2	90
4	300	150, Std. 3	30
5	300	150, Std. 4	10

• **ASSAY PROTOCOL FLOW CHART: Vol. Added (µL)**

Well ID:	Blank A1, B1	NSB C1-D1	Bo E1-F1	Standards G1-B3	Samples C3-H12
Assay Buffer 42	---	150µl	100µl	---	---
Standards or Samples	---	---	---	100µl	100µl
CRP Conjugate	---	50µl	50µl	50µl	50µl
CRP Antibody	---	---	50µl	50µl	50µl
Incub. 2 hour @ RT, shaking & sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 4 x Full well vol. (~400µl)	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
SA-HRP	---	200µl	200µl	200µl	200µl
Incub. 30 min. @ RT, shaking & sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 4 x Full well vol. (~400µl)	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
TMB Substrate	200µl	200µl	200µl	200µl	200µl
Incub. 30 min. @ RT, shaking, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Stop Solution 2 (1N HCl)	50µl	50µl	50µl	50µl	50µl
Read OD at 450 nm	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒

**CRP PLATE LAYOUT:**

A1 Blank	A2 Std 2	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12
B1 Blank	B2 Std 2	B3	B4	B5	B6	B7	B8	B9	B10	B11	B12
C1 NSB	C2 Std 3	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12
D1 NSB	D2 Std 3	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12
E1 Bo	E2 Std 4	E3	E4	E5	E6	E7	E8	E9	E10	E11	E12
F1 Bo	F2 Std 4	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
G1 Std 1	G2 Std 5	G3	G4	G5	G6	G7	G8	G9	G10	G11	G12
H1 Std 1	H2 Std 5	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. \_\_\_\_\_ 3<sup>rd</sup> Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 End Time \_\_\_\_\_ Temp. \_\_\_\_\_ End Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 2<sup>nd</sup> Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_  
 End Time \_\_\_\_\_ Temp. \_\_\_\_\_