

**ASSAY LAYOUT SHEET**  
**for use with Enzo Life Sciences Catalog No. ENZ-KIT190**

• **ASSAY PROTOCOL FLOW CHART:**

	Blank	S0	HC	LC	NC	Samples
Well ID:	A1, B1	C1, D1	E1, F1	G1, H1	A2, B2	C2-H12
Sample Diluent	---	100 µL	----	---	---	---
Standards or Samples	---	---	100 µL	100 µL	100 µL	100 µL
Incub. 30 min. @ 37°C, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 4 x Full well vol. (~350µL)	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
HRP Conjugate	---	100 µL	100 µL	100 µL	100 µL	100 µL
Incub. 15 min. @ 37°C, sealed	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Asp. & Wash 5 x Full well vol. (~350µL)	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
TMB Substrate	100 µL	100 µL	100 µL	100 µL	100 µL	100 µL
Incub. 15 min. @ 37°C in dark	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒
Stop Solution	50 µL	50 µL	50 µL	50 µL	50 µL	50 µL
Read OD at 450 nm	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒	⇒⇒⇒⇒

• **ABBREVIATIONS:**

S0: No Sample Control

HC: High Positive Control

LC: Low Positive Control

NC: Negative Control

**PLATE LAYOUT:**

A1 Blank	A2 NC	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12
B1 Blank	B2 NC	B3	B4	B5	B6	B7	B8	B9	B10	B11	B12
C1 S0	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12
D1 S0	D2	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12
E1 HC	E2	E3	E4	E5	E6	E7	E8	E9	E10	E11	E12
F1 HC	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
G1 LC	G2	G3	G4	G5	G6	G7	G8	G9	G10	G11	G12
H1 LC	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. \_\_\_\_\_

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

End Time \_\_\_\_\_ Temp. \_\_\_\_\_