

**URINARY PROSTACYCLIN ASSAY LAYOUT SHEET**  
for use with Assay Designs' Catalog No. 900-025 and 901-025 (formerly 90025 and 90125)

• **DILUTION TABLE FOR MAKING STANDARDS 1-5:**

<b>Std.</b>	<b>Diluent Vol. (µL)</b>	<b>Vol. Added (µL)</b>	<b>Urinary Prostacyclin Conc. (pg/mL)</b>
1	980	20, Stock	2,000
2	750	250, Std. 1	500
3	750	250, Std. 2	125
4	750	250, Std. 3	31.25
5	750	250, Std. 4	7.81

• **ASSAY PROTOCOL FLOW CHART:**

<b>Well I.D.:</b>	<b>Blank A1, B1</b>	<b>TA C1, D1</b>	<b>NSB E1, F1</b>	<b>Bo G1, H1</b>	<b>Stds. A2 - B3</b>	<b>Samples C3 - H12</b>
Standard Diluent	---	---	100µL	100µL	---	---
Assay Buffer	---	---	50µL	---	---	---
Std. and/or Sample	---	---	---	---	100µL	100µL
Conjugate	---	---	50µL	50µL	50µL	50µL
Antibody	---	---	---	50µL	50µL	50µL
Incub. 2 hours @ RT, shaking						
Asp. & Wash 3 x 200µL						
Conjugate	---	<b>5µL*</b>	---	---	---	---
Substrate	200µL	200µL	200µL	200µL	200µL	200µL
Incub. 45 min. @ RT						
Stop Solution	50µL	50µL	50µL	50µL	50µL	50µL

\* Conjugate for TA must be diluted 1:10 in Assay Buffer. Add 500 µL of A.B. to a clean test tube. From this volume, remove 50 µL of A.B. and, using the same measuring device, add 50 µL of Urinary Prostacyclin Conjugate.

• **URINARY PROSTACYCLIN PLATE LAYOUT:**

A1 Blank	A2 Std 1	A3 Std 5	A4	A5	A6	A7	A8	A9	A10	A11	A12
B1 Blank	B2 Std 1	B3 Std 5	B4	B5	B6	B7	B8	B9	B10	B11	B12
C1 TA	C2 Std 2	C3	C4	C5	C6	C7	C8	C9	C10	C11	C12
D1 TA	D2 Std 2	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12
E1 NSB	E2 Std 3	E3	E4	E5	E6	E7	E8	E9	E10	E11	E12
F1 NSB	F2 Std 3	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
G1 Bo	G2 Std 4	G3	G4	G5	G6	G7	G8	G9	G10	G11	G12
H1 Bo	H2 Std 4	H3	H4	H5	H6	H7	H8	H9	H10	H11	H12

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

1st Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_ Notes: \_\_\_\_\_

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

2nd Incub.: Start Time \_\_\_\_\_ Temp. \_\_\_\_\_

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