

DispoEquilibrium DIALYZER™ Samples from 25µl to 75µl



Advantages:

- Easy to use & disposable
- Small sample volumes: 25µl to 75µl each chamber
- Rapid dialysis due to ultra-thin membrane
- High-quality regenerated cellulose membranes with MWCOs of 5,000 and 10,000 Daltons
- Leak-proof

Applications:

- Protein and Protein-drug binding assays
- Receptor binding assays
- Ligand binding assays
- Protein-protein and Protein-DNA interactions

Harvard Apparatus/Amika's DispoEquilibrium DIALYZER is a single-use product for interaction studies and is currently the only such device on the market. The DispoEquilibrium DIALYZER is leak-proof and provides high sample recovery (almost 100 percent). This system is designed for one-time use with samples such as radiolabeled compounds, avoiding the hassle associated with cleaning the DIALYZER after use.

Each chamber has a capacity of 25 to 75µl. The DispoEquilibrium DIALYZER utilizes high-quality regenerated cellulose membranes with MWCO's of 5,000 or 10,000 Daltons. Sample recovery is very easy through centrifugation or via removal with micropipettes.

DispoEquilibrium DIALYZERS

Membrane MWCO (Daltons)	Qty. of 25	Qty. of 50	Qty. of 100
5,000	SDIS 050JE	SDIS 050KE	SDIS 050ME
\$	85.00	141.00	255.00
10,000	SDIS 100JE	SDIS 100KE	SDIS 100ME
\$	85.00	141.00	255.00

Other MWCO are available upon request

Extra Loading Pippette Tips, pkg. of 100	NP 74-2222
\$	35.00

96-Well Equilibrium DIALYZER™ Samples from 50µl to 200µl



Key

● Membrane

Advantages:

- 96-well format
- Individual membrane for each well
- Small sample volumes: 50µl to 200µl
- Ultra-thin regenerated cellulose membranes
- Membranes are free of sulfur and heavy metal contamination
- High well-to-well reproducibility
- Excellent sample recovery (>95%)

Applications:

- Protein and Protein-drug binding assays
- Receptor binding assays
- Ligand binding assays
- Protein-protein and Protein-DNA interactions

The single use, 96-Well Equilibrium DIALYZER is a novel product for the simultaneous assay of 96 samples. Each well in this system has a separate membrane and thus eliminates the possibility of sample cross-contamination. Reproducibility is very high across the different wells of the Equilibrium DIALYZER and sample recovery is excellent. Wells 96-Well are sealed with 8-cap strips. Thus a row of wells or all 96 wells can be used depending on the specifications of the experiment. The 96-Well Equilibrium DIALYZER utilizes high-quality regenerated cellulose membranes available with MWCO's of 5,000 or 10,000 Daltons.

Catalog No.	\$	Description
SDIS 9605EN	383.00	96-Well Equilibrium DIALYZER Plate Membrane MWCO 5,000 Daltons, pkg. of 1
SDIS 9610EN	383.00	96-Well Equilibrium DIALYZER Plate Membrane MWCO 10,000 Daltons, pkg. of 1
SPLR 0000.1	710.00	Single Plate Rotator, pkg. of 1
SPLR 0008H	3,172.00	8-Plate Rotator Oven, pkg. of 1
SPLR 0000	825.00	Dual Plate Rotator, pkg. of 1

Introduction to Equilibrium Dialysis

Equilibrium dialysis is a specific application of the general phenomenon of dialysis that is important for the study of the binding of small molecules and ions by proteins. It is one of several methods currently available but its attractive feature continues to be its physical simplicity.

The objective of an equilibrium dialysis experiment is usually to measure the amount of a ligand bound to a macro-molecule. This is typically done through an indirect method because in any mixture of the ligand and macro-molecule, it is difficult to distinguish between bound and free ligand. If, however, the free ligand can be dialyzed through a membrane, until its concentration across the membrane is at equilibrium, free ligand concentration $C_{L(f)}$ and the following data can be measured:

Temperature (absolute)	T
Concentration of binding component, e.g. protein	$C_{P(o)}$
Starting concentration of ligand	$C_{L(o)}$
Final concentration of free ligand	$C_{L(f)}$

From which the following parameters can be derived directly:

Concentration of bound ligand	$C_{L(b)}$
Free fraction (of ligand)	f
Bound fraction (of ligand)	b
Degree of binding or saturation fraction	r

Data obtained from several experiments at a range of temperatures and with varying initial concentration of ligand can provide other binding parameters:

Association constant	K
Number of binding sites	n
Binding capacity	N

Further, the thermodynamics of the binding reaction can be derived:

Change of free energy	ΔG
Enthalpy change	ΔH
Entropy change	ΔS

Since equilibrium exists, the value $C_{L(f)}$ is the same on both sides of the membrane. (Note: where charged species are involved the Gibbs-Donnan effect can upset the equilibrium unless moderately concentrated salts are in solution; say 0.6% NaCl).

Hence:
$$C_{L(o)} = C_{L(f)} + C_{L(b)} + C_{L(b)}^*$$

*It is essential to correct this equation to take account of any ligand which might be bound to the membrane.

$$C_{L(b)} = C_{L(o)} - 2 \times C_{L(f)}$$

The free fraction f is given by:

$$f = \frac{C_{L(f)}}{C_{L(o)} - C_{L(f)}}$$

The bound fraction b is: $b = 1 - f$

The degree of binding or saturation fraction r is:

$$r = \frac{C_{L(b)}}{C_{P(o)}}$$

If the protein concentration is known, the Scatchard plot can be used to determine binding constants and the number of binding sites. If the protein concentration is unknown, the absolute number of binding sites is replaced by binding capacity N .

In the former case, values of r would be plotted on the abscissa against $r/C_{L(f)}$ on the ordinate. If only one class of binding sites is present, the Scatchard plot results in a straight line with slope equal to $-K$ see Fig. 1.

The intercept on the abscissa give the value n . If two classes of binding sites are involved, the plot takes the form of an hyperbola. In this case, the asymptotes have slopes equal to $-K$ for each class of site, and their intercepts on the abscissa give the two values for n . The intercept between the curve and the abscissa is equal to the sum of the two values for n , see Fig. 2.

The free energy change is obtained simply by substituting the appropriate values in the following equation:

$$\Delta G = -RT \ln K$$

Where R is the gas constant.

ΔH can be obtained from a graph based upon an integrated form of the van't Hoff equation.

$$\ln K = \frac{-\Delta H}{RT} + C$$

In this case a plot $\ln K$ versus $1/T$ has a slope of $-\Delta H/R$. Once a value for ΔH has been found it can be substituted into: $\Delta G = sH - T\Delta S$ to obtain a result for the entropy change ΔS .

Fig. 1.

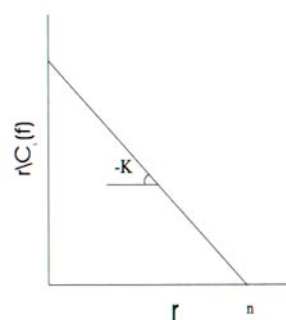
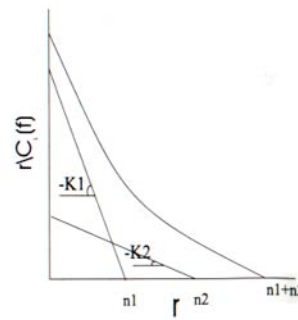


Fig. 2.



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 Weder, H.G., Schildknecht, J., Lutz, R.A. and Kesselring, P., *Eur. L. Biochem.* 42, 475-481 (1974)
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