

# INSTRUCTIONS



## Liposomics

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# Liposome Drug Encapsulation Assay Kit

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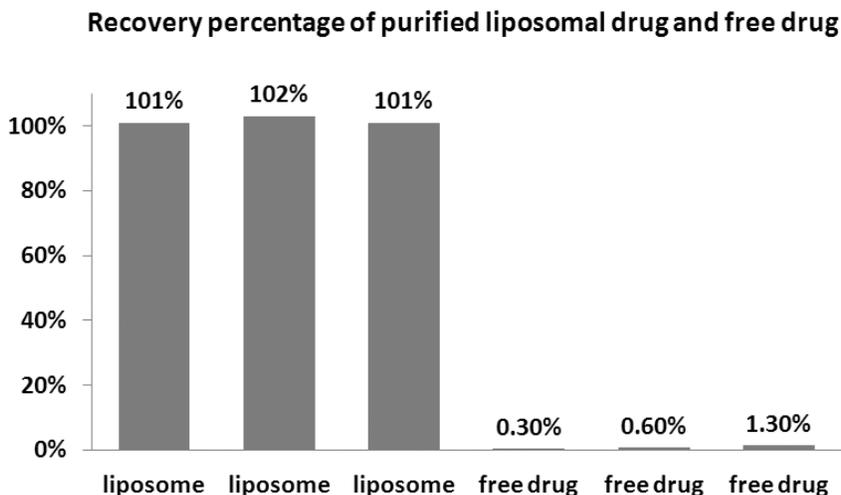
CATALOG NUMBER

LDE10

## INTRODUCTION

Liposome-formulated drug samples may contain free (non-encapsulated) drug molecules that are not encapsulated within the liposomes. Encapsulated drugs may leak out of liposomes during storage or due to exposure of the liposomes to organic solvent, ultrasound vibration or freezing or elevated temperatures. During production of liposomal drugs, drug loading may not be complete leaving certain percentage of the drug non-encapsulated. The **Liposome Drug Encapsulation Assay kit (Catalog number LDE10)** is designed to analyze the percentage of drug encapsulation in liposomes.

The kit is based on spin-column separation of the liposomes from the non-encapsulated drug molecules. It recovers 100 %  $\pm$  2 % liposomes and removes > 98% non-encapsulated drugs. The nearly complete separation between the liposomes and the non-encapsulated drug is based on a combination effect of absorbance and size between liposomes and non-encapsulated drug molecules.



The **Liposome Drug Encapsulation Assay kit (Catalog number LDE10)** includes 30 prepacked spin-columns and 10 ml of elution buffer. It is for measurement of drug encapsulation of 10 liposome samples.

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## ASSAY PROTOCOL

### 1. Sample preparation

Remove any high aggregates by filtration of the samples through a 0.22 µm filter.

### 2. Column preparation

- (1) Use 3 spin columns for each sample (triplicate measurement). Briefly spin the columns to set down the resin. Remove the caps of 1.5-ml Eppendorf tubes and use them as receiver tubes. Remove the bottom tip and the cap of each spin column and insert the column into a receiving tube.
- (2) Spin the columns at 1000 rpm using a benchtop Eppendorf centrifuge for 1 min and discard the elute.
- (3) Repeat Step (2).
- (4) Add 150 µl of the elution buffer, spin the columns at 1000 rpm for 2 min and discard the elute.
- (5) Spin the columns at 1000 rpm for 4 min and change to a clean receiving tube.

### 3. Separation of encapsulated drugs

- (1) Load 50 µl of the liposome sample onto each column.
- (2) Spin the column at 1000 rpm for 2 min.
- (3) Add 150 µl of the elution buffer on the top of the column and spin the column at 1000 rpm for 4 min.
- (4) Mix the elute well by pipetting up-and-down a few times. This is the “Encapsulated” sample.

### 4. Analyze the liposomal drugs

- (1) Prepare the sample for the total drug concentration by mixing 50 µl of the sample and 150 µl of the elution buffer. This is the “Total” sample.
- (2) Measure the light absorbance values of the “Encapsulated” sample ( $A_e$ ), “Total” sample ( $A_t$ ) and the buffer ( $A_b$ )<sup>1</sup>.
- (3) Calculate the encapsulation yield (%)

$$\text{Encapsulation yield (\%)} = (A_e - A_b) \times 100\% / (A_t - A_b)$$

For more information of liposome products and research tools please visit [www.liposomics.com](http://www.liposomics.com).

<sup>1</sup>Note: If the drug concentration is too high, serial dilution is needed to avoid saturated signal. If it is UV absorbance, all the samples are solubilized and measured in ethanol or methanol.