

Liposome size is usually determined by Coulter counting for vesicles larger than 1 μm diameter, or quasi-elastic laser light scattering if smaller than 1 μm diameter. Relative particle size of small, unilamellar vesicles and the degree of contamination with heterogeneous large particles can also be measured turbidimetrically (4). Electron microscopy remains the only accurate, albeit time-consuming method of determining absolute liposome size and mean number of lamellae per lipid vesicle. However, a great deal can be learned with a good phase-contrast microscope equipped with polarizing filters. It is an indispensable tool of liposomology. Both visual and microscopic inspection provide the investigator with a good idea of the approximate size and "lamellarity" of the preparation.

B. Liposome Classes

Size and number of lamellae have guided the standardization of liposome nomenclature. Each liposome category, general methods for producing them, and visual properties will be discussed later, and the reader is referred to appropriate references for details of each method. Also, recent reviews are available that discuss methods of liposome preparation and characterization in greater detail than will be discussed here (5-8; and also Vol. 1-3 of *Liposome Technology* (G. Gregoriadis, ed.)).

Multilamellar vesicles, known as MLVs, are distinguished by being larger than 0.1 μm in diameter and generally having more than five lamellae enclosing the aqueous core. They are prepared simply by drying a thin film of lipid on a surface and hydrating with buffer (1). Thin films are used preferably because the large surface area of exposed lipid facilitates liposome formation and efficient entrapment of aqueous phase. Alternatively, MLV dispersions can be formed by homogenizing or sonicating dried lipid granules suspended in aqueous solution. Care must be taken to ensure that such a preparation is not contaminated with unhydrated lipid. To the naked eye, an MLV dispersion appears milky and opaque at 0.1% lipid (w/v) or greater. The MLVs appear as cross-sectioned, onionlike structures of variable size and shape under high magnification in the phase-contrast light microscope. These preparations can display a Maltese cross birefringent pattern when observed under crossed polarizing filters, if there are sufficient lamellae per vesicle and the interbilayer distance is relatively small and evenly spaced.

The limiting size of a liposome is about 0.03 μm diameter because of the radius of curvature imposed by lipid geometry (Fig. 2). Of necessity, these smallest of vesicles have only one lipid bilayer and, thus, are known as small unilamellar vesicles (SUVs). The