CHAPTER II

MATERIALS AND METHODS

1. Materials.

    1.1 Hexamidine. (Rhone - Poulenc. S.A.)

    1.2 Benzalkonium chloride. (Imperial Chemical Industry).

    1.3 Cetrimide (Imperial Chemical Industry).

    1.4 Cetylpyridinium chloride. (The British Drug House Ltd.)

    1.5 Hydrophilic Ointment U.S.P.XIX

    1.6 Hydrophilic Petrolatum U.S.P. XIX.

    1.7 Polyethylene Glycol Ointment U.S.P. XIX

    1.8 White Ointment U.S.P.XIX

2. Equipments.

    2.1 Cellulose Dialyzer Tubing (Arther H.Thomas Co.)

    2.2 Constant Temperature Water Bath (Hythermco)

    2.3 Electric Stirrer (Fluid Equipment Co.)

    2.4 Single Pan Balance (Sauter)

    2.5 Spectrophotometer Pye Unicam Model SP.1800

3. Methodology.

    3.1 Preparation of Ointment.

        3.1.1 Prepared the following ointment bases,

according to U.S.P.XIX.(19) : Hydrophilic Oihtment U.S.P.XIX (Emulsion Base O/W), Hydrophilic Petrolatum U.S.P.XIX(Absorption Base), Polyethylene Glycol Ointment U.S.P.XIX(Water Soluble Base), and White Ointment U.S.P.XIX (Oleagenous Base).

3.1.2 Ointments containing hexamidine 1.0 mg./g. (0.1 % W/W). They were prepared by incorporating hexamidine in fine powder with ointment bases from 3.1.1 using slab & spatula until receiving a smooth and homogeneous ointment.

3.1.3 Ointment containing hexamidine 1.0 mg./g. in benzalkonium chloride 1:1000 aqueous solution (1,3,5,7,10 and 12 %). Dissolved hexamidine in benzalkonium chloride 1:1000 aqueous solution (1,3,5,7,10 and 12 %), then incorporated this solution with ointment base which selected from 3.1.1 (best ointment base for hexamidine) by using the same procedure.

3.1.4 Ointment containing hexamidine 1.0 mg./g. in cetrimide 1:1000 aqueous solution (1,3,5,7,10 and 12 %). Prepared the same as 3.1.3

3.1.5 Ointment containing hexamidine 1.0 mg/g. in cetylpyridinium chloride 1:1000 aqueous solution (1,3,5,7, 10 and 12 %). Prepared the same as 3.1.3.

3.2 Release from ointments. (1,7,8,16,25,30,31,32)

Used diffusion technique, developed by Bottari,F., et al (7). The capacity of the diffusion cell was 15.9 cm^3.

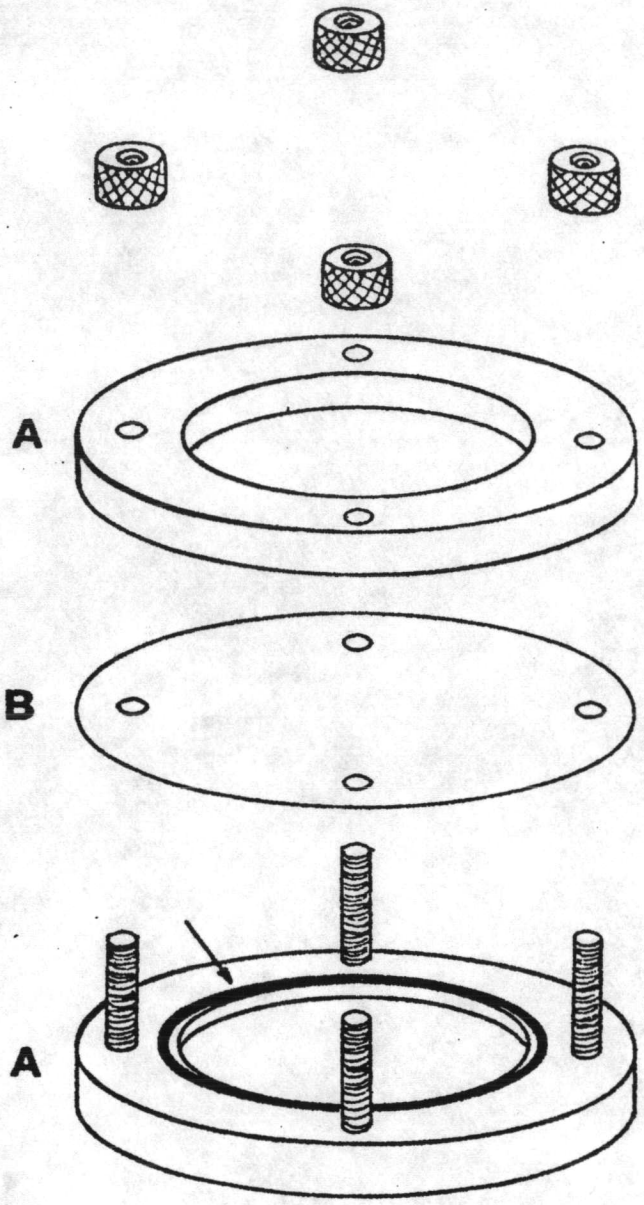

Figure 1 Diffusion cell used for release experiments.

Keys : A plastic cell body and plate.

B cellulose membrane

(Figure 1.) The cell was filled with an ointment, the excess was removed by spatula to produce an even surface, then the cellulose membrane which had already been soaked in water, was carefully placed and pressed on the surface of ointment. A new membrane was used for each release run. The upper part of the cell was then assembled, thus securing the membrane in place. The filled cell was placed in a beaker (internal height 10 cm., internal diameter 7.8 cm.) immersed in a constant temperature bath (37°C). The solution outside the cell was stirred with slow speed. The system was designed to produce resonably fast release rate and to avoid absorption of hexamidine by the cell material. Prewarmed tridistilled water (37°C, 150 ml.) was introduced into the beaker, into which the cell was immediatly placed, and stirring was begun, at 15 minutes time intervals for two hours, pipet 10 ml. sample from the beaker to a 20 ml. test tube and replaced at once with an equal amount of prewarmed tridistilled water. Blank runs, using plain ointment without drug, to fill the diffusion cell and proceeded the same procedure as previously done. Reserved this solution as a blank.

3.3 Analytical Method. (17)

Determined the amount of hexamidine released during the in vitro test by spectrophotometry at 265 nm.