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A new diffusion cell – an automated method for measuring the pharmaceutical availability of topical dosage forms

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Summary

A cyclic system, incorporating a novel diffusion cell, that can measure the pharmaceutical availability of an active principal in a topical dosage form is presented. It is proposed as an aid to pre-formulation and as a quality control tool.

Introduction

There is a need for a simple, universal system that can measure the pharmaceutical availability of topical medicaments and which can be readily applied to all types of topical dosage forms (ointments, creams, transdermal patches,...). A review of the various designs of diffusion cell used for the study of percutaneous absorption in vitro has been published recently (Scott, 1986). Briefly, these consist of a donor compartment and a receptor compartment, separated by a membrane. The receptor fluid may simply be stirred in the receptor compartment ("static") or may be pumped through this chamber in an open ("flow-through") or closed ("cyclic") system. A list of guidelines for the use of such systems has recently been published (Skelly et al., 1987). The purpose of this study was to develop an analogous system to quantitate the pharmaceutical availability of a topical dosage form (Thoma, 1987; Shah et al., 1988). The release profile of a drug from its vehicle is obviously of fundamental importance.

The principal uses envisaged for such a system are two-fold. Firstly, as a screening test for experimental formulations and, thus, as an aid to preformulation by comparing the performance of experimental dosage forms. Secondly, as a quality control measure, to compare the release profile of different lots of a finished product. Intra- and inter-lot comparisons of the topical dosage form would provide, respectively, a measure of the homogeneity and the stability of the manufacturing process. The stability of this parameter should also be considered when expiry dates are determined. The system should be easy to use, reproducible and not necessarily require the incorporation of a radiolabelled ligand.

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A preliminary account of this work has been presented (Martin et al., 1987).

Materials and Methods

Apparatus

(1) A cyclic system including a pump, a new flow-through diffusion cell, a detection device and a reservoir is proposed (see Fig. 1).

Pump. An HPLC pump (Gilson 302) ensures that the receptor fluid is cycled from the reservoir, at a controlled rate, to the detector and the flow cell.

Diffusion cell ("CIRD" cell). The flow-through diffusion cell consists of a sintered glass disk onto which a membrane is clamped with an aluminium ring (see Fig. 2). The internal dimensions of the ring defines the contact area (10 cm^2). The receptor fluid is pumped up through the sintered glass disk, thus coming into contact with the underside of the membrane. Good contact between the receptor fluid and the membrane is thus assured even in the presence of cycling air bubbles. The receptor fluid flows off the disk, is collected in a funnel and pumped to the reservoir. The sintered glass disk also provides physical support for the relatively large surface area of membrane used, thus minimising the risk of puncture. The temperature of the cell is controlled by a water jacket connected to a circulating thermostatic bath

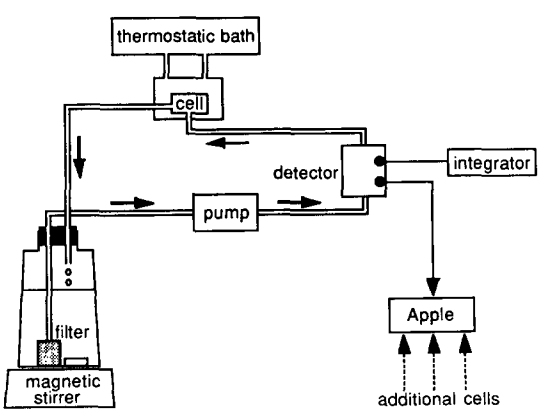

Fig. 1. Schematic diagram of the apparatus.

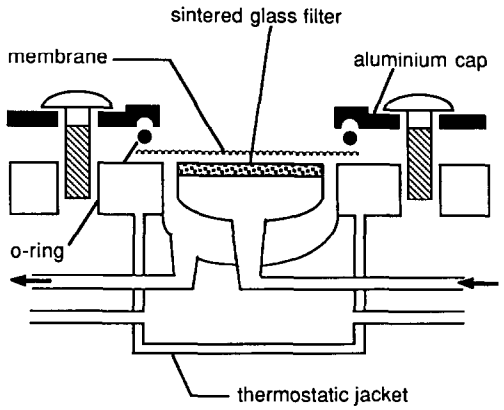

Fig. 2. Cross-section of the diffusion cell.

(Haake D8). An inverted crystallisation dish (50 ml) is placed over the membrane as a draft shield.

Reservoir. The cell effluent is pumped to the reservoir, a 50 ml conical flask equipped with a magnetic stirrer. This ensures that the diffusion products are mixed rapidly, as more than 80% of the total circulating volume of about 50 ml is in the reservoir.

Membrane / receptor fluid. The choice of membrane and receptor fluid is dictated by the physical properties of the drug to be studied. The membrane should be chemically inert towards the formulations under investigation and the drug must be sufficiently soluble in the receptor fluid. The experiments described here concern the lipophilic anti-psoriatic drug, anthralin. We used a silicone rubber membrane (Silastic 500-3, 250 μm thickness, Dow Corning) and *n*-octanol (Merck art.820931) as the receptor fluid. However, cellulose acetate membranes with an aqueous receptor fluid have, for example, been used successfully with more hydrophilic drugs (unpublished results).

Detector. This is connected in between the pump and the diffusion cell for two reasons. The diffusion products are mixed into the "total" circulating volume before passing through the detector and thus the signal observed reflects better the release of the drug. The passage of the receptor fluid through the detector is accompanied by a considerable drop in pressure; if the diffusion cell is placed in between the pump and the detector

the increased pressure in the circulating fluid can cause the membrane to distort. An HPLC-UV detector (Waters 441), set at 365 nm, was used for the studies reported here. However, any on-line non-invasive HPLC detector could probably be used (i.e. fluorescence, refractive index,...). The appropriate choice for each case is determined by the physical properties of the drug being studied.

Data acquisition. The detector is connected to an Integrator (Waters 740) or via an analogue-to-digital convertor (U-Micro) to a micro-computer (Apple II^+). The "Labplot" programme (Biosoft, Elsevier) was used to treat the digital input to the computer. This latter system can collect the signals from up to 4 cells simultaneously. Data acquisition is thus entirely autonomous and the potential errors involved with manual sampling are avoided.

Data treatment. The data is converted into a table of optical density against time. These values are entered into a statistical software package (RSE, BBN Software Products), average values determined and the placebo subtracted. The cell is "calibrated" for each product by measuring the optical density produced when standard solutions are circulated. This permits the results to be expressed as a function of the amount of drug that has been liberated against time. The experiments are performed in triplicate to check reproducibility. A graph of the amount of drug liberated versus the square-root of time is then prepared (see Fig. 3). The apparent release constant (B) is calculated, by linear regression, assuming the relation:

$$Q = B\sqrt{t} + C$$

where Q is the amount of product released at time t and C is the intercept with the y-axis. This is the most general version of the equation derived from Fick's laws (Higuchi, T., 1960, 1961; Higuchi, W.I., 1962, 1967; Higuchi, W.I. and Higuchi, T., 1960; Koizumi and Higuchi, W.I., 1968) to describe the release of a drug from topical formulations. The intermediate points were used for the regression analysis. The initial points, considered to be in the lag-time period, were not included. Equally, the final points, which show that the rate

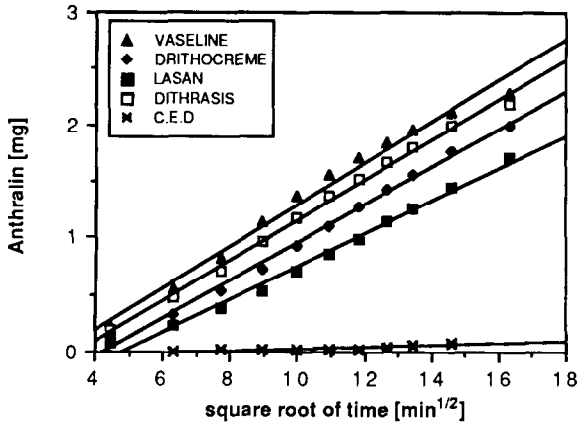

Fig. 3. Release of anthralin (1% w/w) from some proprietary and experimental formulations.

of diffusion reduces at the end of the experiment, were ignored.

(2) A second cyclic system, incorporating the Franz diffusion cell (contact area 2 cm^2), was also studied (Franz, 1975). The same pump, detector and data acquisition device were used. An extra reservoir, however, was not included. This is because the combined volume of the Franz cell and the connecting tubes was about 10 ml. Thus, the ratio of circulating volume : contact area was the same as the apparatus already described. The cell water jacket was connected to the same thermostatic circulating bath.

Drug formulations

A number of formulations of anthralin were studied: Dithrasis (Laboratoires Galderma), Drithocreme (American Dermal Corp.), Lasan HP1 (Stiefel), a suspension in "Codex" Vaseline, and an experimental Cellulose Ester Disc (Shroot et al., 1984). All contained the same concentration of the drug (1% w/w). A slow current of nitrogen was bubbled through the receptor fluid in the reservoir to purge the system of oxygen. Under these conditions it was shown, by UV spectrophotometry, that anthralin was stable. The solubility of anthralin in n-octanol was measured (2 mg/ml).

Procedure

n-Octanol (50 ml) was placed in the reservoir and a slow stream of nitrogen was started. The

membrane was attached to the diffusion cell and then the pump, the detector and the thermostatic bath were switched on. The system was then allowed to equilibrate for about 2 h. A fresh sample of about 1 ± 0.05 g of the formulation was placed on the membrane for each experiment. The output from the detector was recorded for 10 h after the application of the product.

Results and Discussion

The evaluation of this technique was accomplished in 3 stages. Firstly, the effect of a number of physical parameters of the experimental system on the rate of release of a drug was investigated. In particular, the flow-rate of the receptor fluid and the temperature of circulating thermostatic bath were studied. Using the selected conditions, the new diffusion cell was compared to a Franz cell. Finally, the system was used to compare a number of proprietary and experimental formulations of the anti-psoriatic drug, anthralin.

Influence of temperature and flow-rate

Firstly, the influence of temperature and flow-rate on the liberation of anthralin formulated as a suspension in "Codex" Vaseline (1% w/w) was studied.

Temperature. The release of anthralin from this formulation was measured at 4 temperatures

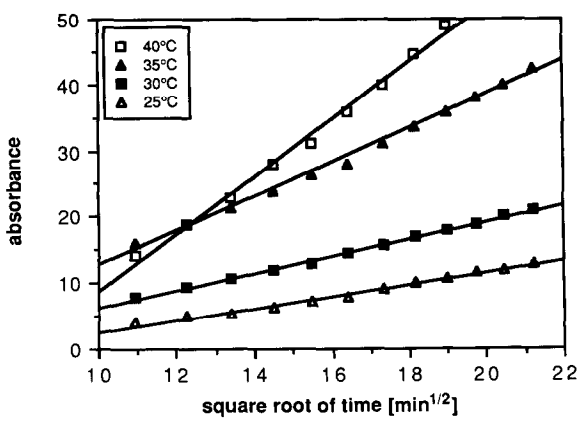

Fig. 4. Effect of the temperature on the release of anthralin (1% w/w) from Vaseline (flow rate = 2 ml/min).

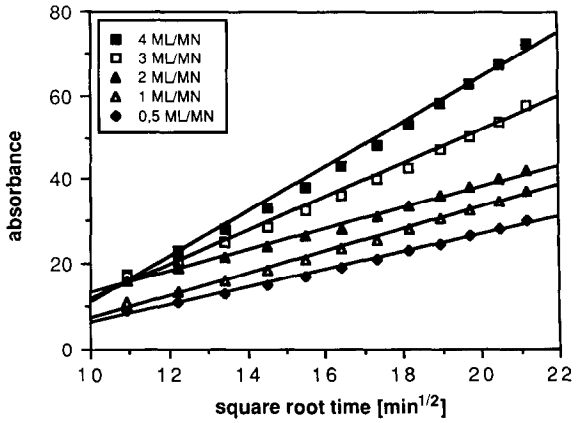

Fig. 5. Effect of the flow-rate on the release of anthralin (1% w/w) from Vaseline (temperature = 35 °C).

(25, 30, 35 and 40 °C). The temperature indicated is that set for the circulating thermostatic bath; the flow-rate (2.0 ml/min) was kept constant. The results are shown in Fig. 4. Over the temperature range studied, the apparent release constant increased regularly. The temperature selected for future experiments was 35 °C. This results in an effective temperature at the membrane/receptor fluid interface of 32 ± 1 °C – the physiological temperature of human skin.

Flow-rate. Using the same formulation, and keeping the thermostatic bath temperature at 35 °C, the liberation of anthralin was measured for flow-rates from 0.5 to 4.0 ml/min. The results are shown in Fig. 5. The flow-rate of 2.0 ml/min was selected as this was the highest rate for which a plot of optical density versus the square-root of time was linear.

Comparison of the diffusion cells

The same formulation of anthralin (1% in "Codex" Vaseline) was used to compare the two diffusion cells. As above, the receptor fluid was *n*-octanol and a Silastic membrane was employed. The flow-rate was set at 2 ml/min and the temperature in the circulating thermostatic bath to 35 °C. Data acquisition and treatment was as above. The observed release constants (18.3 $\mu g/cm^2/min^{1/2}$ for the first system and 22.4 $\mu g/cm^2/min^{1/2}$ in the second) are similar. However, the

flow-diffusion cell we describe here has, in our opinion, two major advantages.

Firstly, the experiment is much easier to perform. The receptor fluid circuit has to be very carefully degassed – a delicate task – when using a Franz cell as any circulating air bubbles tend to collect at the receptor fluid/membrane interface. The design of the 'CIRD' cell ensures that this cannot occur. Secondly, with the Franz cell, the plot of the amount of product liberated against the square-root of time is linear over a relatively short period of time. Thus, the precision and reproducibility of the measurement is reduced.

Comparative liberation of anthralin

In order to test the ability of the system to quantify comparative release characteristics of a series of formulations, a number of proprietary and experimental topical dosage forms of the anti-psoriatic drug, anthralin, were investigated. The commercial formulations chosen were Dithrasis (an ointment specifically designed for 'short contact' therapy), Lasan HP1 and Drithocreme. These were compared to a suspension of the drug in 'Codex' Vaseline (as above) and an experimental delivery device in which anthralin was incorporated into a Cellulose Ester Disc. All 5 formulations contained 1% (w/w) of the drug.

The formulations were compared under the selected conditions defined above. The apparent release constants, determined from Fig. 3, are shown in Table 1. A variation from 0.8 to 18.3 $\mu g/cm^2/min^{1/2}$ was obtained. This demonstrates the first proposed use for this system, namely as an aid to pre-formulation. The apparatus could readily distinguish between the 5 formulations tested. The intra-formulation variation was less than 5% ($n = 3$). This demonstrates the reproducibility of the apparatus and hence, its suitability as an aid to quality control of a given formulation.

TABLE 1

Apparent release constants for the liberation of anthralin from some proprietary and experimental formulations (concentration 1% w/w)

Formulation	Apparent release constant ($\mu g/cm^2/min^{1/2}$)
Vaseline	18.3
Dithrasis	17.7
Drithocreme	16.7
Lasan HP1	14.3
Cellulose Ester Disc	0.8

Conclusion

The diffusion cell presented here has been used successfully to distinguish between a number of topical formulations of the drug anthralin. It has, also, proved to be simple to use, requires minimal surveillance and is very reproducible. Commercial formulations can be analysed directly; thus vehicles of confidential or unknown composition can be compared to experimental systems. The incorporation of a radiolabelled ligand is not necessarily required. The system is very flexible and can be adapted to analyse all types of topical medication.

This system is proposed as a simple, reproducible method to quantify pharmaceutical availability of topical dosage forms and, as such, to aid preformulation and quality control.

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