Effect of PGML Excipient Mixture in a Transdermal System on the in Vitro Transport of Estradiol Across Skin

Gretchen D. Irion,1 Michael D. Garrison,1 and William Abraham1,2

Received May 5, 1995; accepted July 18, 1995

Purpose. To measure the effect of a combination of excipients from a silicone based pressure sensitive adhesive (PSA) on drug transport across skin.

Methods. Partitioning of propylene glycol monolaurate (PG-ML) from silicone PSA and a solution formulation into the stratum corneum (SC) was measured using radiolabeled PG-ML. Transport of a model drug, estradiol, as well as PG-ML across skin were measured in vitro using heat separated epidermis from human cadaver skin.

Results. The PG-ML partitioning into SC showed a saturation and was independent of the formulation. The local dielectric of the lipid bilayers of the SC showed an increase as a result of PG-ML uptake, as determined by the decrease in fluorescence lifetime of a lipophilic probe incorporated into the SC. However, there was no alteration of lipid packing in SC.

Conclusions. The PG-ML and estradiol transport showed a good correlation over 3 days, suggesting that the two species are co-transported across the epidermis.

KEY WORDS: transdermal drug delivery; pressure sensitive adhesive; transport enhancement; fluorescence spectroscopy; estradiol.

INTRODUCTION

Transdermal drug delivery has become an attractive alternative in controlled drug delivery (1). The stratum corneum (SC) of the epidermis provides the major barrier to percutaneous penetration of drugs (2,3). This diffusional barrier is often overcome with the aid of excipients in the transdermal formulations (4,5). These excipients are thought to act as solubilizers, fluidizers, plasticizers or vehicles. The excipient could increase the solubility of the drug in the adhesive, thereby increasing the concentration in the delivery system in situations where the drug is present in a powdered form in the adhesive. It could partition into the skin and perturb the highly organized lipid bilayers of the SC, increasing the permeability of the skin (4). The excipient could act as a plasticizer in the adhesive, increasing the diffusion of the drug in the adhesive. It could also act as a solvent or as a vehicle, facilitating the transport of the drug across skin. Excipients are used individually or in combination in transdermal systems to provide any one or a combination of the above effects resulting in an enhanced delivery of drugs across skin (4,6).

One of the first steps in the development of a transdermal drug delivery system is to perform a formulation feasibility and optimization study from a solution formulation before exploring a suitable pressure sensitive adhesive (PSA) platform. Quite often, the transdermal flux of the drug observed from a PSA matrix formulation is less than from a solution formulation. The literature is replete with reports from solution formulations on enhancer effects (4-6). However, there are only a few studies reported from solid matrix formulations (7).

In the present investigation, we have measured the uptake of an excipient, propylene glycol monolaurate, into skin and its transport across skin from a silicone based PSA and a liquid formulation. Propylene glycol monolaurate, a fatty acid ester, is used to enhance the transport of drugs from transdermal systems (8,9). The transport of a model drug, estradiol, was measured to determine if there was any correlation between the transport of drug and excipient from a given formulation. The objective of the present study is to understand the effect of a combination of excipients from a silicone based PSA on skin. Building on our earlier work (10), we have used a frequency-domain fluorescence spectroscopic technique to determine the effect of these excipients on the lipid order of the SC.

MATERIALS AND METHODS

A mixture (PGML) of propylene glycol monolaurate (PG-ML, 52.9% by weight), propylene glycol dilaurate (PG-DL, 32.7%), methyl laurate (ML, 9.2%), and propylene glycol (PG, 5.2%) obtained from Gatetofosse (France) was used without further purification. The composition of this mixture was determined by gas chromatography using a HP-5890 gas chromatograph with a Restek RTX-35 capillary column and a FID detector. Radiolabeled PG-ML was synthesized using PG and 1-14C lauric acid (American Radiolabeled Chemicals, Inc., St. Louis, Missouri). One mCi (3.66 mg) of 14C lauric acid was added to a mixture of 2 mg of unlabeled lauric acid and 5.36 mg of PG and the resulting reaction mixture of lauric acid and PG (1:2.5, mole ratio) in hexane was mixed with 0.3M Boron Trifluoride (BF3) ethyl etherate catalyst in diethyl ether and the reaction mixture was maintained at 70°C for 6h under nitrogen. The excess catalyst was removed by drying under a stream of nitrogen and the 14C PG-ML was isolated from the reaction mixture by preparative thin layer chromatography. The reaction mixture was dissolved in chloroform, applied on a 1 mm thick silica gel plate (Alltech Associates), and eluted in hexane:ethyl ether:acetic acid (69:30:1, by volume). Commercial PGML mixture was used as the standard to identify the different bands. The PG-DL eluted the furthest on the plate, followed by lauric acid and PG-ML eluted about one-third the distance from the origin. The 14C PG-ML fraction from this plate was purified on a second plate using a polar solvent system (5% methanol in chloroform). In this solvent system, the 14C PG-ML moved one-half the distance from the origin, while the lauric acid (which was a cross-contamination from the first fraction) was retained close to the origin. The final yield of PG-ML was 0.73 mg or ~10%.

Two different donor formulations were used in the up-
take and transport study. Neat PGML liquid spiked with 4μCi of 14C PG-ML was used as a solution formulation. A silicone PSA formulation was made by adding a known amount of PGML to a silicone solution in Freon (non end capped, 58% solids from Dow Corning) to a final composition of 14% by dry weight. The solution was spiked with 2μCi of 14C PG-ML. The liquid adhesive was stirred for 2h before casting the film at a thickness of 5 mil. A polyester film was used as the backing and as the release liner for the patches. The specific activity of 14C PG-ML in the final formulations were 25.5 μCi/g of PG-ML in the neat PGML liquid and 24.9 μCi/g of PG-ML in the silicone formulation.

Skin uptake and transport studies were performed using Franz cells across a diffusional area of 0.785 cm². The estradiol transport studies were done using multiple pieces of skin from different donors, while the rest of the experiments were done using three pieces of skin from the same donor. The patches with the backing material were applied to pieces of heat separated human epidermis obtained from human cadaver skin, and the receiver fluid was analyzed for 14C PG-ML by liquid scintillation counting, at intervals of 24h for 3 days. The amount of 14C PG-ML left in the patch and in epidermis were also analyzed at each time point. After the flux studies, the epidermis was removed from the diffusion cell, and the adhesive was peeled away. The patches and the backing were extracted in methanol:methylene chloride (1:1, by volume) overnight and analyzed for 14C PG-ML. The epidermis was digested in 1 ml of Solvable (0.5M sodium hydroxide, NEN Research Products) overnight and then counted. In one of the time points, i.e., the third day sample, the SC was separated from viable epidermis by trypsin digestion. The epidermis was placed dermal side down on a filter paper soaked in 1% trypsin for 4 to 6h at room temperature and then at 4°C overnight. The partially digested tissue was agitated in a fresh 0.5% trypsin solution for 1h resulting in a translucent sheet of SC, which was rinsed in distilled water, digested in Solvable and counted for PG-ML. The trypsin digests along with the filter paper were counted for PG-ML in the viable layers. Day 3 samples were analyzed for 14C PG-ML in the SC as representative samples. In the samples where neat PGML was the donor, the epidermis was rinsed three times in 3 ml portions of unlabeled PGML to remove any PG-ML from the epidermis surface before digesting in Solvable. In a separate experiment, SC was separated from pieces of epidermis by trypsin digestion as described above. The SC pieces of known area were then equilibrated with PGML liquid mixture in a scintillation vial. The PGML mixtures was spiked with 14C PG-ML to a specific activity of 15 μCi/g of PG-ML. After 72h equilibration, the pieces of SC were rinsed three times in unlabeled PGML to remove any 14C PG-ML from the surface, and the tissue was treated with trypsin identical to the method described above. After rinsing in distilled water, the SC pieces were digested in Solvable and analyzed for 14C PG-ML. The trypsin fractions along with the filter paper and the water rinses were pooled and counted for 14C PG-ML as a check for the amount of PG-ML that could have leached out during the different experimental steps. In the estradiol transport experiments, the drug was present at 5% by weight in the silicone PSA formulation and was present as a saturated solution in the PGML liquid formulation. The estradiol in the receiver fluid was analyzed by reverse-phase HPLC using a C-18 column.

For fluorescence experiments, the epidermis was treated similar to the transport studies, but the donor formulation did not contain any radiolabeled PG-ML or estradiol. The epidermis was equilibrated for 18h in a side-by-side diffusion cell, with a donor solution of a fluorescent probe, 1,6-diphenyl-1,3,5-hexatriene (DPH) in water. The aqueous solution of DPH was prepared by adding 5μl of 2mM DPH solution in ethanol to 1.5 ml of water corresponding to a final concentration of 6.7μM. After pretreatment with DPH, the epidermis was treated with the PGML liquid and the silicone PSA formulations separately, as described above in the uptake studies. At the end of the treatment period (24h to 72h), the epidermis was removed from the diffusion cell, mounted on an aluminum block and placed in the sample holder for spectral acquisition. The samples in which the epidermis was treated with the silicone-based formulations, the spectral acquisition was performed with the adhesive on the epidermis. All fluorescence measurements were performed using an SLM-Aminco MHR 4850 spectrophotometer. Fluorescence lifetimes and anisotropy were measured using the phase modulated 325 nm line of a HeCd laser as described elsewhere (10 - 12).

RESULTS

Skin Uptake Data. The uptake of PG-ML into epidermis showed tissue saturation after day 2 from the silicone but not from the PGML donor, as seen in Fig 1. The distribution of PG-ML between the SC and the viable layers of epidermis is shown in Table I. The amount of PG-ML per cm² of SC was comparable from both the silicone and the PGML formulations, suggesting tissue saturation. The amount of PG-ML per unit area of the viable layers of the epidermis was three-fold higher from the PGML formulation compared to the silicone. The amount of 14C PG-ML lost from the SC during trypsin treatment and water rinses was 11.5 ± 2.1% of the total 14C PG-ML in the SC, which is within the standard deviation of the SC-uptake data shown in Table I. The data in Table I are not corrected for this PG-ML loss.

Transport Data. The transport data for PG-ML across epidermis is shown in Fig 2. The amount of PG-ML transported across epidermis gradually increased with time up to 3 days from both silicone and PGML liquid formulations. The amount of PG-ML transported across epidermis from the PGML mixture was six-fold higher than that from the silicone PSA formulation. Estradiol transport across epidermis from these two formulations is shown in Fig 3. The estradiol transport from the PGML liquid formulation was about 15 times that from the silicone PSA formulation. The drug transport across the epidermis from these two formulations showed a good correlation with the PG-ML transport over a period of 3 days, as seen in Fig 4.

Fluorescence Data. Table II shows changes in lifetime and limiting anisotropy for DPH in epidermis treated with different formulations. The lifetime decreased after 3 days of exposure to PGML from both formulations. The decrease in lifetime was observed even after 24h exposure to PGML from the neat liquid and was twice that from the PSA-treated epidermis. The limiting anisotropy showed no decrease after