The hardships and obstacles Sherry Jung faced during the course of her research project all seemed to melt away the day she finally obtained significant and reproducible results. Her contribution to the advancement of dermatology is only one of Sherry’s many accomplishments. During her time at UCI, she has been an HSEP Medical Volunteer, a Peer Educator for the Counseling Center, and has served as a Biological Sciences Representative on the ASUCI Legislative Council. She is also a founding member and the current editor-in-chief of Med Times. Sherry’s advice to future undergraduate researchers is to start early, learn from your mistakes, be patient, and stay motivated.

The role of the skin is to provide a barrier to the external environment, rendering absorption of therapeutic drugs, including corticosteroids, problematic. Preliminary testing has shown that fatty acids (linoleic acid) incorporated into structurally configured polymers can act as penetration enhancers. This modification alters the barrier properties of the stratum corneum and the migration of topical drugs such as cortisol through human skin. Previous unpublished studies using an amine compound, polyoxyalkyleneamine D 400 (polyamine D 400), have suggested that topical corticosteroid solutions supplemented with novel polymers improve the penetration of therapeutic drugs. Sets of unique polymers, synthesized in this laboratory, were selected for initial assessment of penetration enhancement using the in vitro Franz diffusion model. The penetration and retention of cortisol into the skin layers were determined by measuring radiolabeled drug levels at the experimental endpoint by liquid scintillation counting. Linoleic acid + polyamine D 400 polymer achieved statistically higher cortisol penetration through the skin when compared to the commercial standard or the vehicle control. In the future, these unique polymers may be used as penetration enhancers to improve transdermal delivery of other topical drugs of therapeutic interest.

The rewards and experiences derived from mentoring undergraduate students through the School of Biological Sciences are mutual. The skills acquired through participation in research programs at UCI carry over into our students’ professional lives, and their publications help to further their career development. The current project by Sherry Jung has served to illuminate our understanding of the penetration of a common dermatological drug when used in combination with a custom-synthesized polymer. The results obtained point to direct applications in the field of dermatology as the polymeric compounds may be further modified for targeted-drug delivery. The avenues of research in this area are numerous, and we anticipate ongoing investigations using novel polymeric penetration enhancers.
Introduction

The skin provides a physical barrier to the harmful effects of the external environment. In so doing, it also interferes with the absorption of topical therapeutic drugs, including corticosteroids. Numerous vehicles and penetration enhancers have been synthesized to increase transdermal delivery of drugs. In a previous study, polyethylene glycol (PEG) was shown to have significant effects on drug penetration when skin structures were hygroscopically manipulated (Sarpotdar et al., 1986). Preliminary research has also shown that fatty acids linked to structurally configured polymers act as penetration enhancers, modifying the barrier properties of the stratum corneum and allowing migration of drugs in human skin (Martin, 1993). In another study, amines and unsaturated fatty acids also exhibited skin penetration enhancement (Gwak and Chun, 2002). Previous studies using an amine compound, polyoxyalkyleneamine D 400 (polyamine D 400), suggest that increased permeability of topical corticosteroid solutions supplemented with novel polymers may improve penetration of therapeutic drugs (Chess, unpublished data).

The purpose of this study was to evaluate the permeability of a topical corticosteroid solution that is supplemented with novel polymers comprised of PEG-, amine-, and fatty acid-linked compounds in order to either enhance penetration in the skin and/or facilitate transdermal delivery for systemic drugs. The unique polymers were comprised of a diisocyanate with a fatty acid or methoxy-PEG of given molecular weight linked to another fatty acid or polyamines of various molecular weights. The present study was an initial investigation of these novel polymeric additives.

Methods and Materials

Synthesis of Polymers

Amphipathic fatty acid-amine compounds (those exhibiting both hydrophilic and lipophilic characteristics), lipophilic fatty acid-fatty acid compounds, and hydrophilic PEG-amine compounds were synthesized by first reacting diisocyanate (R1) with either methoxy-PEG 550 (R2, Figure 1), or a fatty acid (R2, Figure 2), then linking the resulting prepolymer with a fatty acid or polyamines of various molecular weights (R3). The R3 subunit varied in molecular weight.

Fifteen polymeric materials and compounds were synthesized for preliminary assessment of penetration enhancement using the Franz diffusion model; upon analysis, only eight exhibited promising results. These eight polymers included: two PEG-amine compounds, methoxy-PEG 550-polyamine D 400 (formulation No. 16B) and D 2000 (No. 17B); and one fatty acid-fatty acid compound, linoleic acid-linoleic acid (No. 14). In addition, five fatty acid-amine compounds were synthesized: linoleic acid-polyamine D 230 (No. 21) and two formulations each of D 400 (No. 20, No. 22) and D 2000 (No. 11, No. 18B), which differed in synthesis. Linoleic acid compounds No. 20 and No. 22 had molecular weights of 502 and 428, respectively. No. 11 was synthesized overnight, while No. 18B was completed with a 3 hr process (Table 1).

Test Materials

Using 60% ethanol as the topical vehicle, 5% of each polymeric compound was prepared as stock and combined with the representative drug, 1% hydrocortisone. Cortisol in 60% ethanol served as a control, while a known commercial 1% hydrocortisone lotion was used as a standard. To radiolabel the formulations, 25 µl 3H-cortisol was dried under nitrogen gas and then mixed with 5 ml of each compound, control, and the lotion standard.

In Vitro Assay

A total of 3-6 Franz chambers (6.0-11.0 ml volume) were used for controls and standards, while 6-9 chambers were used for testing each compound. Each set of chambers was mounted in a console equipped with magnetic stirring motors and a circulating water bath manifold. Cell reservoirs were filled with isotonic phosphate buffered saline.
supplemented with 4% bovine serum albumin (BSA), and maintained at 37 °C for each assay (Figure 3).

A total of 30 mg of each spiked formulation was applied and gently rubbed onto 1.7 cm² samples of human cadaver skin, which were then placed onto the diffusion apparatus (Figure 4).

Drug penetration was determined by sampling the reservoir over time and calculating cumulative percent recovery. At 1, 8 and 24 hr, 1.0 ml reservoir fluid samples were obtained for scintillation counts, and the sample volume was replaced in each chamber. After 24 hr, each skin sample was gently washed with a mild detergent and subsequently wiped with gauze. After the skin was removed from the diffusion chamber, the stratum corneum was separated by sequential cellophane tape stripping. The epidermis and dermis were separated by briefly heating the tissue in a microwave oven and scraping the epidermal layer from the dermis using a #10 scalpel.

**Data Analysis**

The penetration and retention of the radioactive drug into the reservoir and skin compartments were quantified by use of liquid scintillation counting. Calculations of the total recovery in each of the reservoir and skin compartments were based on percent of total radioactive dose applied. The mean percent concentration in the compartments for each compound was averaged for all experiments. Data were analyzed using Student’s t-test; p ≤ 0.05 indicates significance.

**Results**

Based on the current study, the fatty acid-amine group (five novel formulations) demonstrated increased transdermal penetration of cortisol (Figure 5). One formulation (No. 20) exhibited significant cumulative penetration into the reservoir as compared to the standard (p < 0.001). In addition, when comparing the penetration of No. 20 to the fatty acid-fatty acid group (No. 14) and the PEG-amine group (No. 16B or No. 17B), a significant increase of drug retention in the epidermis was observed (p ≤ 0.002). Furthermore, all formulations, as well as the standard, exhibited increased transdermal absorption over the control.

<table>
<thead>
<tr>
<th>Chemical Group</th>
<th>Formulation No.</th>
<th>R2 subunit</th>
<th>R3 subunit</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PEG – AMINE (Hydrophilic)</strong></td>
<td>16B</td>
<td>Methoxy-PEG 550</td>
<td>Polyamine D 400</td>
</tr>
<tr>
<td>17B</td>
<td>Methoxy-PEG 550</td>
<td>Polyamine D 2000</td>
<td></td>
</tr>
<tr>
<td><strong>FATTY ACID - FATTY ACID (Lipophilic)</strong></td>
<td>14</td>
<td>Linoleic acid</td>
<td>Linoleic acid</td>
</tr>
<tr>
<td><strong>FATTY ACID – AMINE (Amphipathic)</strong></td>
<td>21</td>
<td>Linoleic acid</td>
<td>Polyamine D 230</td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>Linoleic acid (MW 502)</td>
<td>Polyamine D 400</td>
</tr>
<tr>
<td></td>
<td>22</td>
<td>Linoleic acid (MW 458)</td>
<td>Polyamine D 400</td>
</tr>
<tr>
<td></td>
<td>11</td>
<td>Linoleic acid (Long-term synthesis)</td>
<td>Polyamine D 2000</td>
</tr>
<tr>
<td></td>
<td>18B</td>
<td>Linoleic acid (Short-term synthesis)</td>
<td>Polyamine D 2000</td>
</tr>
</tbody>
</table>

Table 1

Chemical groups and R subunit designations of polymers tested
Discussion

Although the intrinsic nature of the integument is to provide a barrier to the external environment, it also precludes the absorption and retention of topically applied therapeutic drugs. The skin structure is composed of three resilient layers: stratum corneum, epidermis, and dermis. The amalgamation of these transitional layers and their physical properties and physiological forces designates the skin as an ideal component in regulating absorption and retention. Absorption and retention are dependent on many factors, including pH, molecular weight, solubility, electrolytes, polarity, and especially the lipophilic/hydrophilic nature of the molecules involved. Based on our results, the transdermal absorption of cortisol was increased with the use of amphipathic fatty acid-amine formulations as compared to hydrophobic fatty acid-fatty acid or hydrophilic PEG-amine vehicles.

According to the mosaic theory, proteins in the skin may take up water and the resulting swollen membranes may become permeable to water-soluble substances (Rothman, 1954). In this study, alcohol in 40% water was used as a topical vehicle, which may have provided enough hydration to cause expansion of the membrane, thereby enabling penetration of topically applied cortisol.

Because the skin is composed of alternating lipophilic and hydrophilic layers, amphipathic compounds that possess both properties may absorb into the skin more effectively. Ideally, amphipathic compounds may form spherical micelles (Figure 6), which minimize the lipophilic-hydrophilic interface and overcome physiological forces in the skin. These micelles may invert their structural formation in the presence of these forces during transdermal penetration.

The effectiveness of topically applied therapeutic drugs is dependent upon the retention in skin layers and the transdermal absorption into specific target tissues. This study serves as an initial evaluation of novel polymeric compounds that may be useful as potential vehicles for cutaneous or systemic drug delivery. Additional testing of these novel compounds, utilizing other therapeutic drugs, is indicated by these results. These unique polymers may also be used as penetration enhancers in combination with other drugs of therapeutic interest for topical or transdermal delivery, and can be custom-synthesized to target specific skin compartments.

Acknowledgements

I would like to thank Dr. Jerry L. McCullough, Professor of Dermatology, for his gracious support. I also wish to thank Mr. Samuel Chess, Mr. Michael Plowman, and Ms. Anna Jane Eliseo for their expertise, encouragement and support with this project.
Works Cited

Chess, S. Unpublished data, UCI College of Medicine, Department of Dermatology, 1994.


