

Evaluation of the *in vitro* Human Skin Percutaneous Absorption of Testosterone in PCCA VersaBase[®] Anhydrous HRT

SUMMARY: The human skin percutaneous absorption of PCCA Micronized Testosterone in PCCA VersaBase Anhydrous HRT and PCCA VersaBase[®] Cream was evaluated *in vitro*, and it was shown that VersaBase Anhydrous HRT performs comparably to our industry-leading VersaBase Cream in facilitating testosterone absorption. Since VersaBase Anhydrous HRT has a water activity below 0.6 ($A_w < 0.6$), it may provide an excellent option for compounding pharmacists who rely on anhydrous topical formulations for extended default beyond-use dates (BUDs) without compromising on permeation performance.

Introduction:

Testosterone is the main hormone associated with sexual desire in both men and women. In the U.S., there are about 43% of women between 18 to 59 years of age experiencing sexual dysfunction [1]. Hypoactive sexual desire disorder (HSDD) is a significant medical condition recognized by FDA, but currently there is no FDA-approved androgen therapy for it [1]. Transdermal testosterone has been shown beneficial and safe to use in women with HSDD in several clinical trials [2-4], and is commonly prescribed, but only available as compounded medication. VersaBase Cream is one of the suggested vehicles to deliver testosterone in women, and has been receiving positive feedback from patients.

PCCA VersaBase Anhydrous HRT is a new proprietary base for topical delivery of female hormones into and through the skin with extended stability [5]. Compared with VersaBase Cream that contains water, VersaBase Anhydrous HRT has a water activity below 0.6 ($A_w < 0.6$), classifying it as an anhydrous base. This allows extended default beyond-use dates (BUDs) for preparations that do not have stability studies.

The purpose of this study is to compare the percutaneous absorption of micronized testosterone incorporated in PCCA VersaBase Anhydrous HRT and in VersaBase Cream (Table 1.) in an *in vitro* dermatomed skin model.

Testosterone 1mg/Gm Topical Gel (VersaBase Anhydrous HRT)	
Testosterone USP Micronized CIII (Yam)	0.1%
Propylene Glycol USP	5%
Base, PCCA VersaBase Anhydrous HRT	94.9%

Testosterone 1mg/Gm Topical Cream (VersaBase Cream)	
Testosterone USP Micronized CIII (Yam)	0.1%
Propylene Glycol USP	5%
Base, PCCA VersaBase Cream	94.9%

Table 1. Compounded formulas (PCCA formulas 13468 and 11641^a) were used in the percutaneous absorption study.

^a Modification was made to F11641 in order to compare with F13468.

Methodology:

Skin Preparation

The percutaneous absorption of testosterone was measured using human cadaver abdomen skin tissue from three Caucasian female donors. Dermatomed skin samples were purchased from BioIVT (Westbury, NY) and were cryopreserved and stored at -20°C in tightly sealed plastic bags. Prior to use, the skin samples were defrosted and then soaked in diffusion medium for at least 30 min at room temperature. The samples were visually checked for any significant damages, such as cuts, or holes. Skin tissues from 3 donors and 3 replicates were used for each compounded formula.

Franz Cell Diffusion

The Franz diffusion system (surface area of 1.77 cm²) was used in this study. The diffusion cells were mounted in the diffusion apparatus and the physiological diffusion medium was added to the receptor compartment. A skin integrity test was performed using a Precision LCR meter. Intact skin has transcutaneous electrical resistance at least 2 times greater than the diffusion medium. The finite dose, approximately 5 mg/cm² of the compounded formula, was applied on each skin sample using a positive displacement pipette and a pellet pestle to spread the product across the skin surface. The receptor solution (HBSS #14175-079, 25 mM HEPES, #15630-080 and 50 µg/mL Gentamicin, #15750-060, Gibco) was stirred magnetically at 600 rpm with the water jacket temperature maintained at 32±0.5°C. During the exposure period, samples of the receptor solutions (1 mL) were removed at predetermined time points: 2, 4, 6, 8, 12 and 24 hours after applying the compounded formula.

Testosterone Quantification

The quantification of testosterone in receptor solution was performed by ELISA (Cayman, Ann Arbor, MI) following manufacturer's instruction.

Results and Discussion:

Testosterone that was applied on the skin passed through the stratum corneum, epidermis and dermis layers of the skin model, and finally reached receptor solution. This process mimics testosterone penetrating through the skin and into systemic circulation *in vivo*. Percutaneous absorption of testosterone in this study refers to the amount of testosterone detected in the receptor solution and is shown in Figure 1. Testosterone was detected as early as 2 hours after skin application, and the amount continued to increase, but not significantly different between the two compounded formulas. By the end of 24 hours, percutaneous absorption of testosterone facilitated by

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VersaBase Anhydrous HRT was 2338 ± 594 ng/cm² of skin surface, which is comparable to 1901 ± 506 ng/cm² by VersaBase Cream.

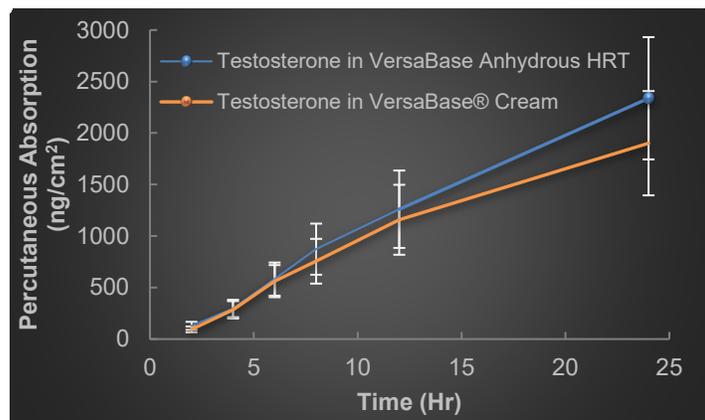


Figure 1. Across donor summary: mean skin percutaneous absorption of testosterone (ng/cm² of skin) in two compounded formulas during 24 hours diffusion. Results were plotted as mean \pm standard error.

In order to understand the kinetics of testosterone absorption in the two compounded formulas, the rate of absorption, or flux rate, was determined in each skin donor and was summarized in Figure 2. The rate of percutaneous absorption shows a rapid penetration upon application and the maximum flux was achieved at approximately 4-6 hours post-application, followed by a slow decline in all donors. There is no statistical significance in flux rate between the two compounded formulas at any time point, suggesting that the two bases have comparable capabilities in delivering testosterone into skin.

In summary, profiles of testosterone in both VersaBase Anhydrous HRT and VersaBase Cream are very similar during 24 hours in the flux rising and declining rate, flux peak time, duration and total amount of testosterone absorption.

Conclusions:

This *in vitro* study performed in the dermatomed human skin model has demonstrated that the proprietary topical base VersaBase Anhydrous HRT facilitates the percutaneous absorption of testosterone across human cadaver skin. The profile of absorption is comparable to VersaBase Cream. One of the desired characteristics for an ideal female hormone delivery base is to produce steady delivery without quick peaking or declining. VersaBase Cream, as our industry-leading base, has already received satisfactory responses from patients. With a similar delivery capability, VersaBase Anhydrous HRT could provide a reliable option to compounding pharmacists to extend default BUDs with the assurance of an excellent permeation performance.

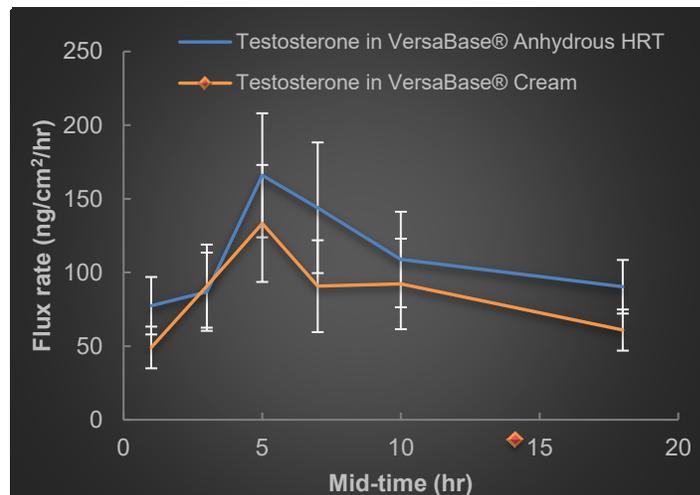


Figure 2. Across donor summary: mean flux rate of testosterone (ng/cm²/hr) in two compounded formulas. Absorption values were reported at midpoint of sample collection. Results were plotted as mean \pm standard error.

References:

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4. Davis S, Papalia MA, Norman RJ, et al. Safety and efficacy of a testosterone metered-dose transdermal spray for treating decreased sexual satisfaction in premenopausal women: a randomized trial. *Ann Intern Med.* 2008;148(8):569-77.
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