In Vitro Characterization of the Percutaneous Absorption of Tramadol into Inner Ear Domestic Feline Skin Using the Franz Skin Finite Dose Model

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Poster #

Purpose

The aim of this study is to characterize the percutaneous absorption of two tramadol formulations (Tramadol 100 mg/g in Pluronic Lecithin Organogel (PLO) and Tramadol 100 mg/g in Phospholipid Base), when applied to the inner ear of domestic feline skin, *in vitro*, using the Franz skin finite dose model[1]. This model has proven to be a valuable tool in predicting *in vivo* percutaneous absorption of topically applied drugs and, therefore, was selected to characterize the absorption of tramadol into and through the *ex vivo* inner ear skin of domestic feline donors[2]. Tramadol, a synthetic opioid that exerts analgesic effects by binding to μ-opioid receptors as well as inhibiting neuronal reuptake of norepinephrine and serotonin, can be used for the treatment of domestic feline pain. Tramadol can be incorporated into transdermal bases such as PLO or Phospholipid Base for transdermal delivery[1, 3].

Methods

The percutaneous absorption of two tramadol formulations were evaluated using normal feline ventral inner ear skin obtained from two different donors, a domestic short hair feline female (donor 1) and male (donor 2). The skin from the donors were cut into multiple sections to fit nominal 0.8 cm^2 Franz diffusion cells, a chamber that cultures the skin cells and allows for the condition of the skin to be maintained at a temperature and humidity that matches *in vivo* conditions. Within the chambers, the cells were mounted on a diffusion apparatus so that the epidermal surface is exposed to the laboratory environment while the dermal layer is immersed in receptor solution[3], which consists of 0.1x Phosphate-Buffered Saline (PBS) with 0.1% Volpo, pH 7.4 ± 0.1. Using the finite dose technique, a variable finite dose (e.g. 25 μg/cm^2) of each formulation was applied to the outer surface of each skin section. At 2, 4, 8, 12, 24, 32, and 48 hr following dose application, the receptor solution was removed, replaced with fresh receptor solution, and a predetermined volume of aliquot was saved for subsequent High Performance Liquid Chromatography (HPLC) analysis. Following collection of the last receptor solution sample at 48 hr, skin surfaces were washed and the intact skin was extracted in 80 : 20 ethanol : water overnight. Tramadol content within the surface wash and the skin were determined via HPLC analysis of the extractant sample[1].

Results

To characterize the percutaneous absorption of tramadol, a total of 4 parameters were determined for each chamber, as follows: total absorption, rate of absorption, surface wash, and skin content. Mean values and standard deviation were calculated for each parameter across the 2 donors. Total absorption (μg) was the sum of tramadol content (μg) within the 7 samples collected over 48 hr. Rate of absorption, presented as flux (μg/cm^2/hr) of tramadol into receptor solution, was determined by dividing the amount of tramadol absorbed during a time interval and the length of that interval. Surface wash (μg) and skin content (μg) refers to the amount of tramadol found on the skin surface and within the skin after 48 hr, respectively. Results of this study show that both PLO and Phospholipid Base were capable of facilitating the absorption of tramadol across *ex vivo* feline inner ear skin (Figures 1 and 2). However, the mean total absorption of tramadol, when in Phospholipid Base (100.40% ± 3.24), was higher than when in PLO (90.28% ± 14.70). When examining rate of absorption, the decline in rate was steadier for tramadol in Phospholipid Base, forming a more predictable and reliable absorption profile in comparison to the rapid decline and fluctuations in rate for tramadol in PLO. Tramadol content within surface wash was also lower, when in Phospholipid Base (3.65% ± 0.14), than when in PLO (10.45% ± 14.42). This difference indicates that more tramadol was absorbed when in Phospholipid Base. While very minute amounts of the applied dose were found within the skin, tramadol content in the skin was 1.09% ± 0.07 for Phospholipid Base formulation and undetectable for PLO formulation. The higher tramadol skin content may be due to the ability of Phospholipid Base to partition into various layers of the skin to a greater extent than PLO[1].

Conclusions

The higher mean total absorption, steadier decline in flux, lower surface wash, and higher skin content seen with the tramadol in Phospholipid Base formulation show that Phospholipid Base may be a more appropriate transdermal base for the transdermal delivery of tramadol in comparison to PLO. Knowledge of this study's results can be used to predict the *in vivo* rate and extent of percutaneous absorption of tramadol in domestic felines. The prediction can guide practitioners and pharmacists, when prescribing and compounding with tramadol for feline use, to select a transdermal base with the best and most predictable percutaneous absorption potential[1].

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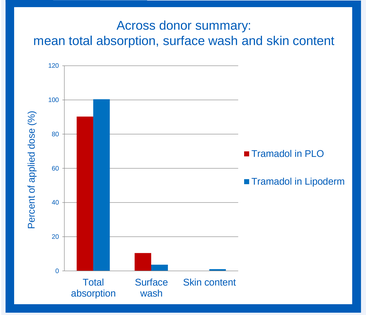

Figure 1. Tramadol absorption profile.

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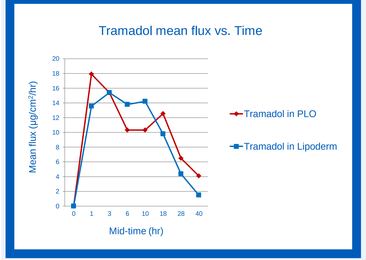

Figure 2. Mean flux (μg/cm^2/hr) results across donors for the two formulations.

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2. Takasuga, S., Yamamoto, R., Mafune, S., Sutoh, C., Kominami, K., Yoshida, Y., Ito, M. and Kinoshita, M. (2011). *In-vitro* and *in-vivo* transdermal iontophoretic delivery of tramadol, a centrally acting analgesic. *Journal of Pharmacy and Pharmacology*, 63(11): 1437.

3. Franz, T.J., Lehman, P.A. and Raney, S.G. (2009). Use of excised human skin to assess the bioequivalence of topical products. *Skin Pharmacology and Physiology*, 22: 276–286.

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