

Elongation of Fatty Acids from Skin Care and Cleansing Formulations and their Involvement in Lipid Synthesis for Different Fitzpatrick Skin Types

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Background

We have clearly shown that fatty acids when properly formulated into care and cleansing products are elongated and converted to ceramides in the stratum corneum and contribute to efficacy and skin health [Murphy et al, Sci Rep. 2022]. Differences in the levels and composition of lipids have been reported in Fitzpatrick Skin Types IV-VI, therefore it is of interest to us to establish the benefit of our technology across these skin types.

Objective

To investigate whether free fatty acid (FFA) ingredients present in properly formulated skin care and cleansing products deliver and are incorporated into skin of different Fitzpatrick Skin Types (FST) to help provide a healthier skin lipid barrier.

Methods

Two IRB-approved clinical studies were conducted on healthy female participants, using marketed formulations for a petroleum jelly containing lotion, isethionate syndet bar and mild amino-acid bodywash. To track palmitic acid conversion the non-deuterated palmitic acid in the formulations was replaced by d31-palmitic acid (d31PA).

In-vivo Study 1

- 12 healthy females
- 35-50 years
- FST: I-III
- Forearms (Non-dry skin)
- Syndet bar and lotion with d31PA

In-vivo Study 2

- 11 healthy females
- 35-50 years
- FST: I-III
- Forearms (Non-dry skin)
- Legs (Moderately dry skin)
- Body wash d31PA

After 4 weeks of application, tape strips were collected for analysis of elongated FFA and deuterated sphingosine (as a measure of ceramide production) by liquid chromatography tandem mass spectrometry (LC/MS/MS).

Ex vivo human skin explants from abdomen or breast from donors with a range of FST were dosed topically with d31PA over 4 days. Explants were cultured under normal cell culture conditions (37°C, 5% CO2) and harvested on day 5. Lipids were extracted and analysed for elongated fatty acids species and for sphingosine by LC/MS/MS.

Results (FFA elongation, *in vivo*)

LC/MS/MS analysis showed that after 4 weeks of product application, FFA are elongated up to C26:0.

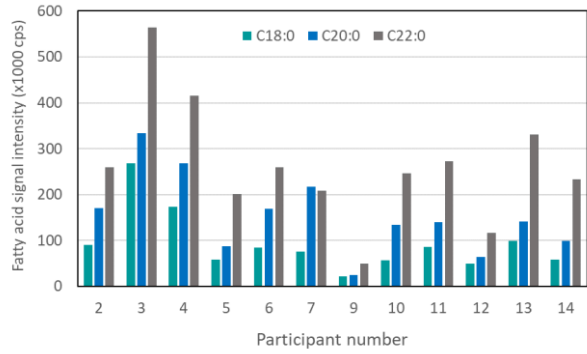


Figure 1 (*in-vivo*): Detectable levels of elongated C18:0, C20:0 and C22:0 (longer chain fatty acids) were observed from all participants using lotion with d31PA (C16:0) at 28 days.

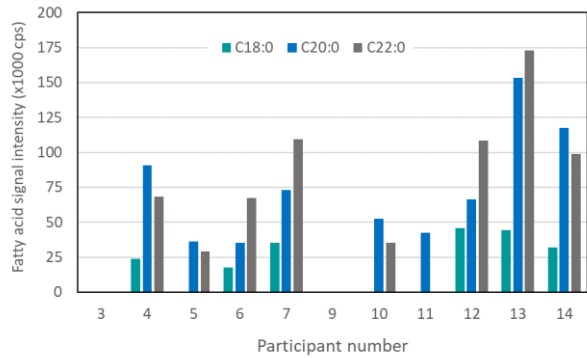


Figure 2 (*in-vivo*): Detectable levels of elongated C18:0, C20:0 and C22:0 (longer chain fatty acids) were observed from 9 of 11 participants using bar with d31PA (C16:0) at 28 days.

Elongation of d31PA was also demonstrated after 28 days application via a body wash (study 2) – data not shown

Results (ceramide production, *in vivo*)

LC/MS/MS analysis showed that after 4 weeks of product application, FFA are used in the formation of ceramides and free sphingosine in human skin.

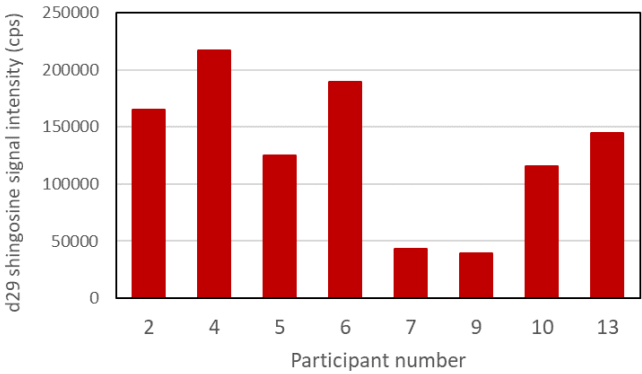


Figure 3 (*in-vivo*): Formation of ceramide (measured as d29 sphingosine) was observed from all participants using lotion with d31PA (C16:0) at 28 days.

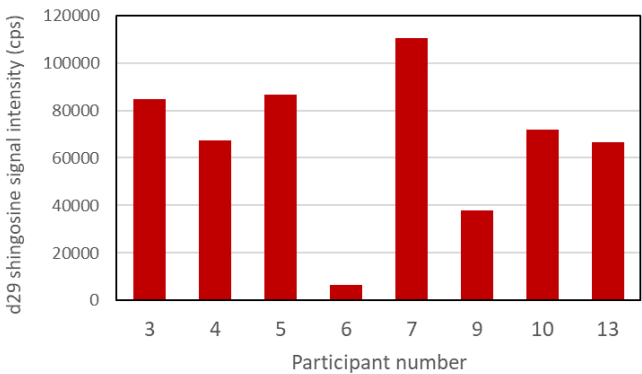
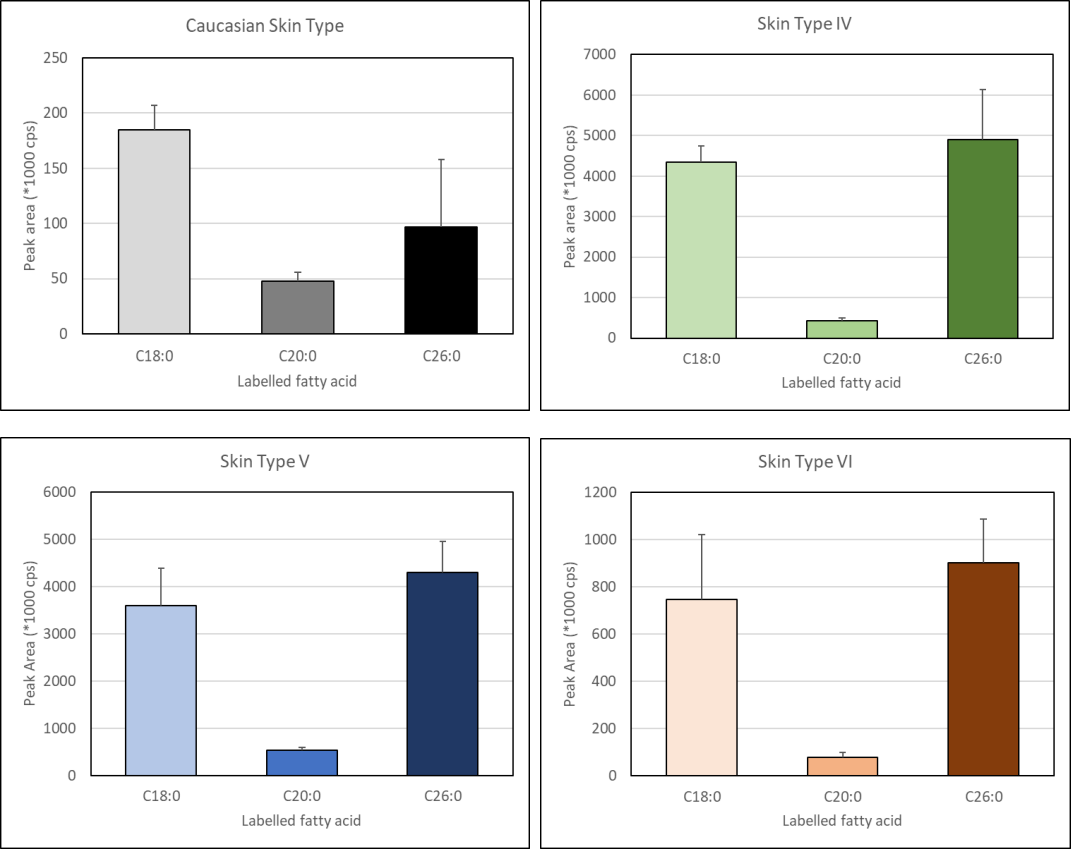


Figure 4 (*in-vivo*): Formation of ceramide (measured as d29 sphingosine) was observed from all participants using syndet bar with d31PA (C16:0) at 28 days.

Results (FFA elongation, *ex vivo*)

The *ex vivo* studies on all FST tested showed deuterated FFA with chain lengths higher than palmitic acid. Hydrolysis of these samples liberated measurable levels of deuterated sphingosine (data not shown) thereby demonstrating the incorporation of exogenous dPA into ceramides as well as into elongated fatty acids.

Figure 5 (*ex-vivo*): Elongation of d31PA (C16:0) into longer chain FFA extracted from *ex vivo* skin samples. Each graph represents the data from a different FST.



Conclusions

In vivo studies showed that shorter chain free fatty acids (palmitic acid), when formulated properly to deliver efficiently in lotions, bars and cleansers, are elongated and incorporated into ceramides.

A robust and reproducible *ex vivo* model has been established which can be used to demonstrate fatty acid elongation and ceramide formation across all FST.

A well-designed cleanser can nourish the barrier with new ceramides and elongated fatty acids. Not surprisingly, well-designed lotions can provide the same benefits to a greater degree. An optimal skin care regimen would include both to provide a healthy well-nourished barrier and is relevant across all FST.