Sunflower OleoDistillate, a new natural PPAR Alpha activator with anti-inflammatory properties

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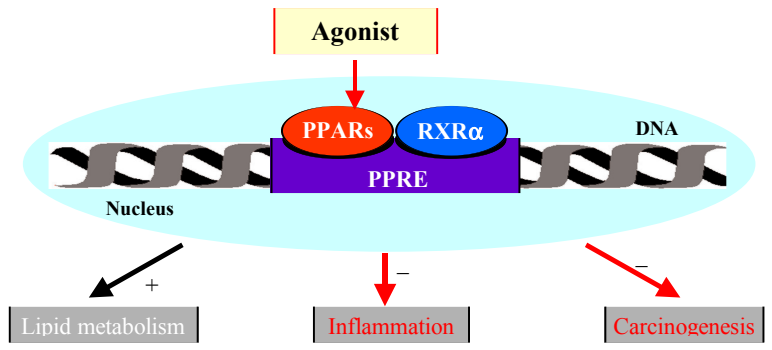
Introduction

The peroxisome proliferator-activated receptors (PPAR) are type II nuclear receptors that are activated by a wide range of fatty acids and synthetic ligands.
Three PPAR isoforms have been identified: PPARα (NR1C1), PPARβ/δ (NR1C2), and PPARγ (NR1C3).
Although PPARs are known to be regulators of lipid metabolism, they have been involved in regulation of inflammation and cell proliferation notably in epidermis (Figure 1). PPARα stimulates differentiation and apoptosis and decreases proliferation both in cultured human keratinocytes and *in vivo* when applied topically to mouse skin (1-3). Topical treatment with PPARα activators accelerates permeability barrier recovery following acute barrier disruption (3). Moreover, treatment with PPARα activators inhibits cutaneous inflammation in animal models of irritant contact dermatitis using phorbol 12-myristate-13-acetate (TPA) and allergic contact dermatitis using oxazalone (4). PPARβ/δ and PPARγ have similar properties in skin.

Figure 1: Mechanism of action of PPARs



Sunflower OleoDistillate has been incorporated in a cosmetic cream formulated specifically for the management of atopic dermatitis (AD). We have shown that this specific cream:

- was able to improve the clinical signs of AD: skin dryness (88%), desquamation (84%), erythema (81%), pruritus (80%) [227 atopic children under paediatricians and dermatologists control]
- was adapted even during atopic flares *
- had a major complementary therapeutic role with a 75% steroid sparing effect *
- was a simple and mandatory way for improving quality of life of atopic children and their parents *

[* 35 pediatricians, 86 children with light to moderate atopic dermatitis, multicentric, randomized, blinded study.]

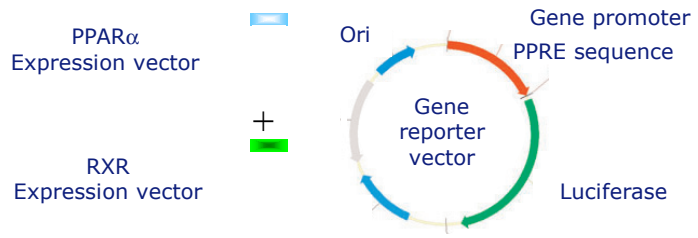
Since PPARs are activated by lipids we have tested the possibility that the Sunflower OleoDistillate could transactivate these nuclear receptors.

Protocol

1. Transactivation assays

CV-1 cells (ATCC CCL-70) were co-transfected like described in figure 2.

Figure 2: co-transfection model



Cells were treated with Sunflower OleoDistillate or Fenofibric acid (as positive control) during 24 hours. PPARα activation was determined by following luciferase activity (luminescence measurement).

2. Phorbol 12-myristate-13-acetate (TPA)-induced ear inflammation

Inflammation was induced by topical application of TPA (in acetone) on mice right ears. Acetone alone was applied on left ears (5).
45 minutes and 4 hours after TPA application, tested compounds were applied on both ears. 18 hours after inflammatory insult induced by TPA, inflammation was assessed as the percentage of increase in ear thickness in the TPA-treated right ears versus the acetone-treated left ear.

3. Real Time PCR analysis

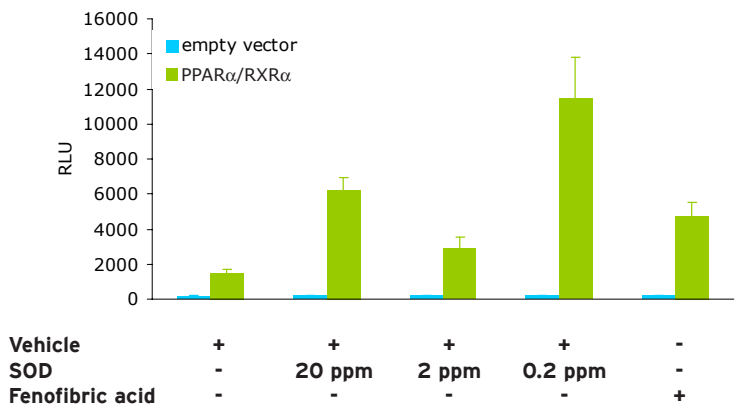
Total RNAs were extracted from ears. TNFα and IL-1β mRNA expression were determined by Real-Time PCR. Results are expressed as percentage of increase between left and right ears.

Results

Sunflower OleoDistillate transactivates PPARα *in vitro*

Treatment with the PPARα agonist fenofibric acid induced a 3 times increase of relative luciferase activity (RLU) compared to vehicle treated cells (Figure 3). Interestingly, Sunflower OleoDistillate (SOD) significantly increased luciferase activity. These data suggest that the **Sunflower OleoDistillate is a modulator of PPARα**.
Despite a significant increase of luciferase activity after stimulation with agonists for PPARβ/δ or for PPARγ, Sunflower OleoDistillate didn't seem to activate this two nuclear receptors (Data not shown).

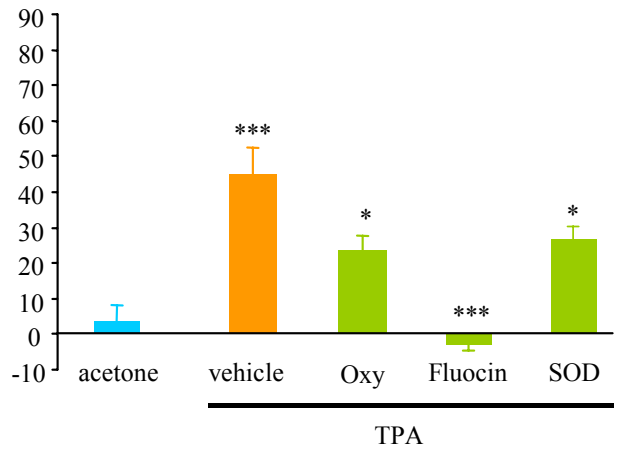
Figure 3: Transactivation of PPARα



Improvement of ear thickness after Sunflower OleoDistillate treatment

The results of ear thickness measurements 18 hours after TPA application are depicted in Figure 4. The reference group treated with the vehicle showed an important increase of ear thickness (45%) after TPA application. Compared to this group, mice treated with Oxysterol or Fluocinolone (positive controls) and Sunflower OleoDistillate (SOD, 2%) displayed a **significant reduction of ear thickness**.

Figure 4: Increase of ear thickness (%)



Normalisation of TNFα and IL-1β mRNA levels in ears of mice treated with Sunflower OleoDistillate

Pro-inflammatory cytokines mRNA levels were measured using Real-Time PCR (Figures 5 and 6). **A significant limitation of the TPA-induced TNFα and IL-1β levels** has been observed in fluocinolone-treated mice as well Sunflower OleoDistillate-treated mice.

Figure 5: Increase of TNFα mRNA (%)

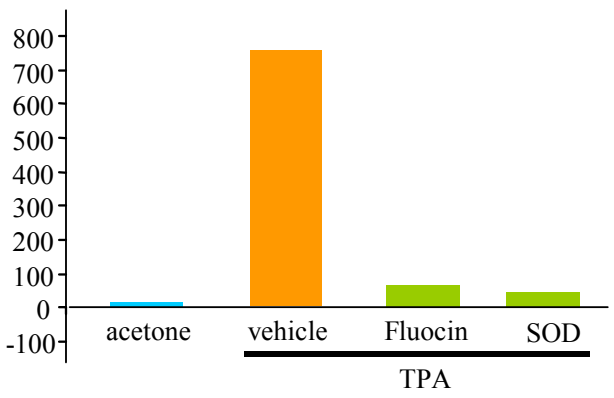
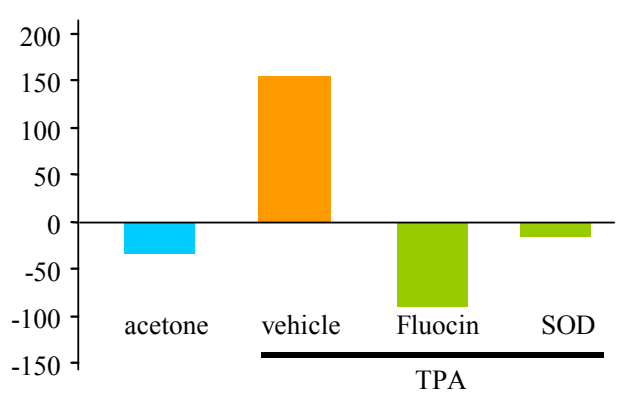


Figure 6: Increase of IL-1β mRNA (%)



Conclusion

By this study, we have shown that Sunflower OleoDistillate is able to activate PPARα *in vitro*. Moreover, this natural compound improves ear inflammation induced by TPA.

Sunflower OleoDistillate thus represents an interesting drug to prevent and treat inflammatory cutaneous disorders.

Since PPARα is involved in lipids synthesis and has anti-inflammatory properties, it is tempting to speculate that the positive clinical outcomes observed after Sunflower OleoDistillate treatment must be due to its ability to activate PPARα.

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