IN VITRO TRIAGE OF ANTI PRURITIC COMPOUNDS

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LEARNING OBJECTIVE: To evaluate a novel complex of active ingredients for its capacity to inhibit several itch receptor activation and inflammatory mediator release *in vitro.*

TAKEAWAY MESSAGE: The novel multi-ingredient complex effectively targets multiple itch pathways and reduces inflammatory mediator release, offering a promising multi-target approach to break the itch-scratch cycle and neuroinflammation in AD.

Conflict of Interest:

L. Canchy, C. Cheng F. Juchaux and N. Ade are employees of L'Oréal.

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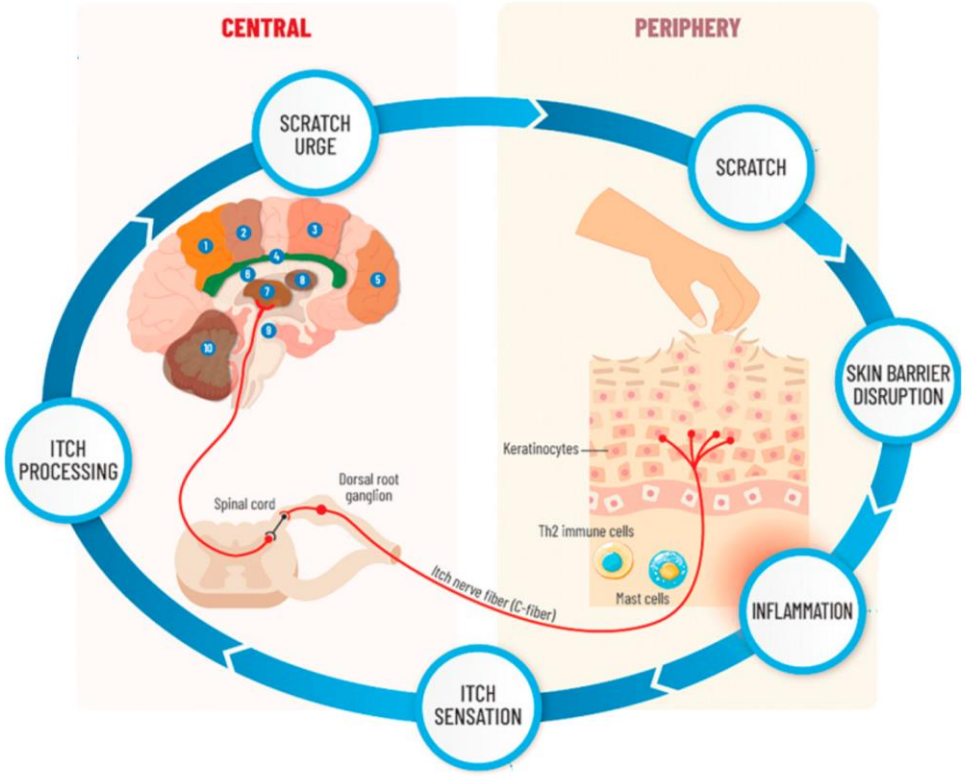
INTRODUCTION

CHRONIC PRURITUS MOST BURDENSOME SYMPTOM OF AD

For 96% of AD patients, one of the **most important** treatment goals is **to be free of itching**.[1]

The sensation of atopic itch is mediated by the interplay between epidermal barrier dysfunction, upregulated immune cascades, and the activation of structures in the peripheral and central nervous systems.

Vicious **ITCH-SCRATCH CYCLE**
exacerbates skin barrier dysfunction and inflammation.

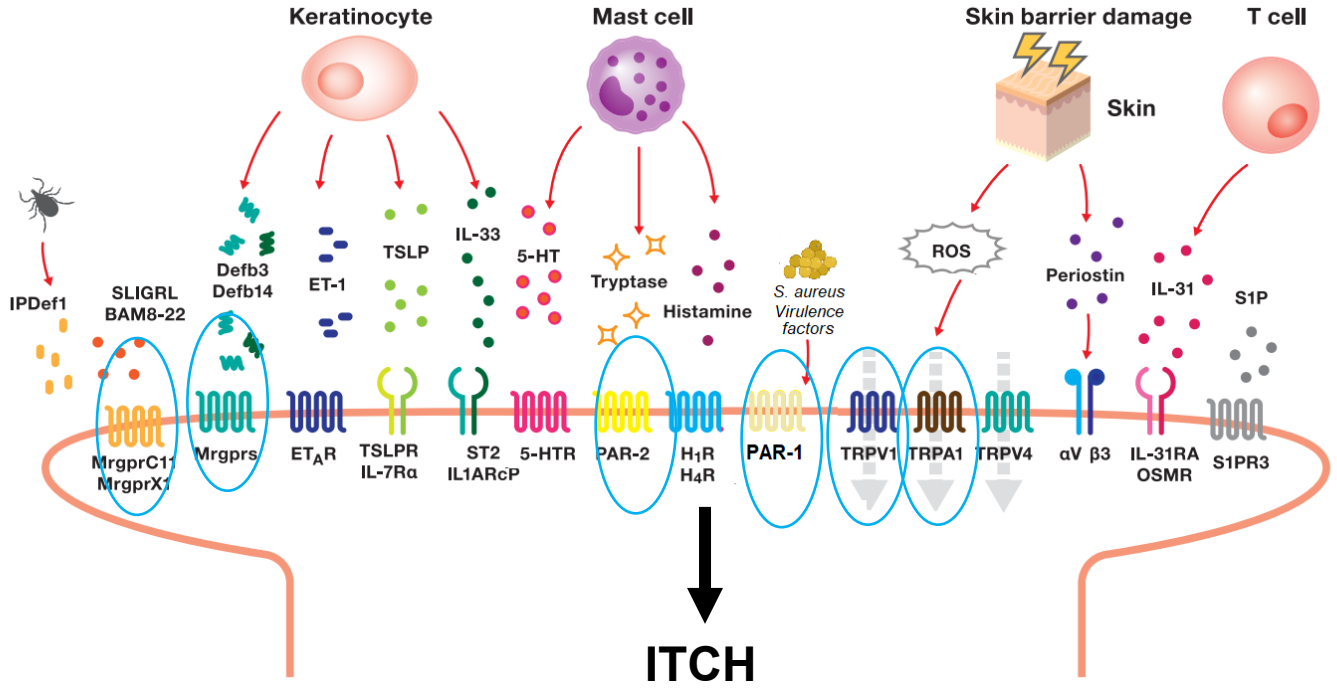


Adapted from M. Steinhoff et al. (2025),Dermatitis. 2025 Aug 14

1. Augustin M et al. J Eur Acad Dermatol Venereol. 2020;34(1):142-152.

INTRODUCTION

At a cellular level, itch signals are primarily transmitted by specialized intra-epidermal nerve fibers (IENF) through various receptors such as Protease-Activated Receptors (PAR-2 and PAR-4) and, transient receptor potential vanilloid 1 (TRPV1) channel, or the Mas-related G-protein-coupled Receptor (MRGPR) family. Furthermore, inflammatory mediators like thymic stromal lymphopoietin (TSLP), which is a key regulator of the Th2 response contribute to sustaining this itch-scratch vicious circle.



Adapted from M. Tominaga, K. Takamori (2022), Allergology International ;71:265-277

MATERIAL & METHODS

The objective of this study was to evaluate a novel complex of active ingredients for its capacity to inhibit itch receptor activation and inflammatory mediator release *in vitro.*

To do so, we assessed the inhibitory effects of the compounds on various itch receptors of interest on recombinant cell lines using fluorimetry to measure calcium mobilization. Then, inflammatory mediator release was measured using ELISA in a reconstructed human epidermis (RHE) mimicking atopic dermatitis with dexamethasone (DEX) as positive control. For each test, pretreatment with individual compounds or the multi-ingredient complex was performed prior to stimulation with specific agonists or a cytokine cocktail.

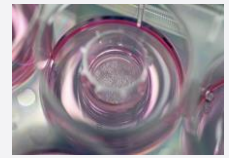


Illustration of reconstructed human epidermis (RHE)

EXPERIMENT 1

INGREDIENT A
Lysate of *Vitreoscilla filiformis,* a bacteria that has been cultivated in La Roche-Posay Thermal Spring water

INGREDIENT B
Anti-protease peptide extract

INGREDIENT C
Obtained from *Ophiopogon japonicum tuberous roots*

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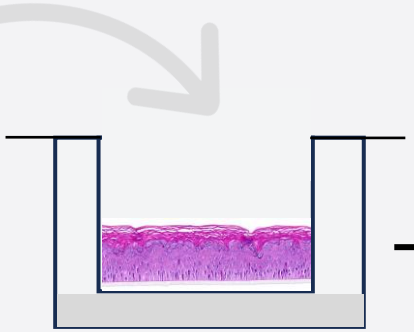
PAR-2, PAR-4, TRPV1, TRPA1, MRGPRX1, MRGPRX2, Mrg D

EXPERIMENT 2

multi-ingredient complex (A+B+C)



SkinEthic™ RHE mimicking AD

ELISA

TLSP
RANTES,
and CCL2

Cytokine mix-stimulated TH2 condition
[IL-4 + IL-13+ TNF-a + Poly(I:C)]

RESULTS

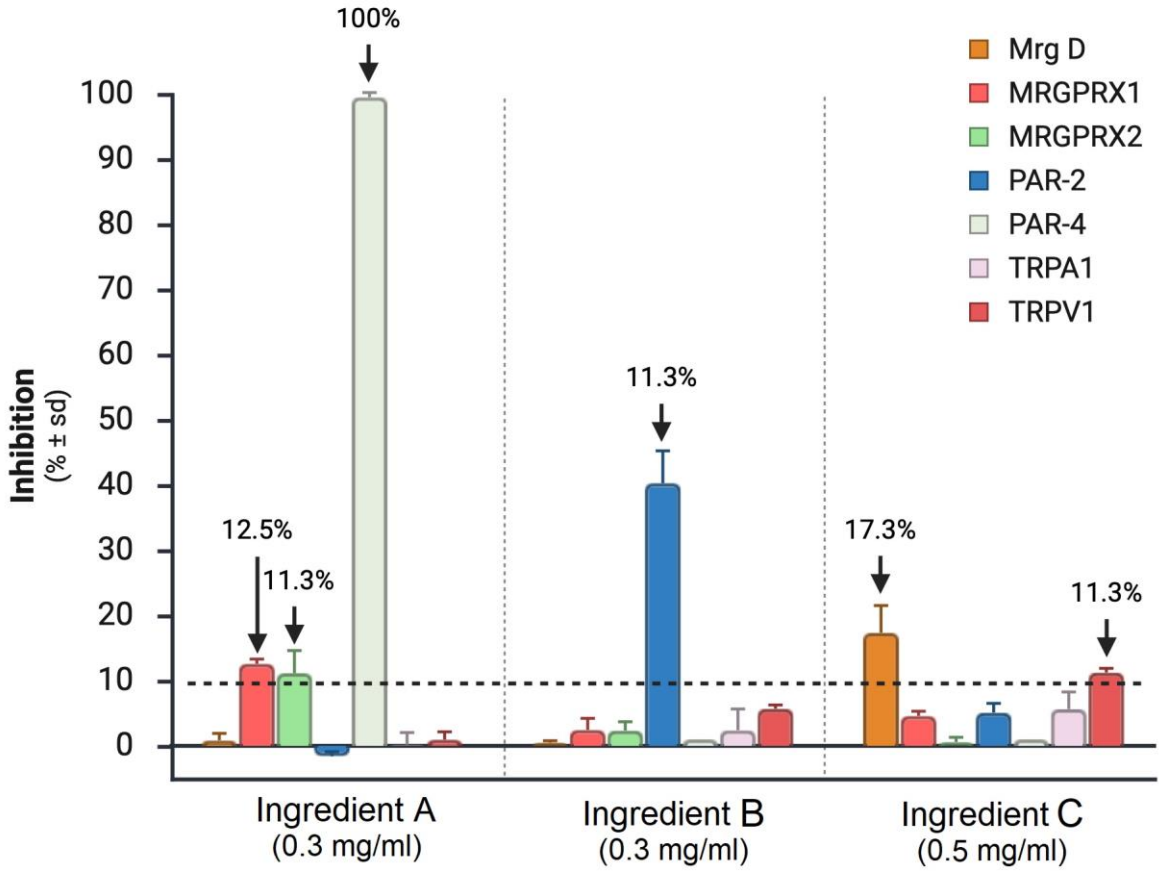
EXPERIMENT 1

Individual compounds demonstrated distinct inhibitory effects (Ca^{2+} influx inhibition) on the different activated itch receptors :

- **Ingredient A** completely inhibited **PAR-4** activation and had a slight inhibitory effect on both **MRGPRX1** and **MRGPRX2**, 12.5% and 11.3% respectively.
- **Ingredient B** showed an inhibitory effect of 11.3% on **PAR-2**.
- **Ingredient C** was able to inhibit **Mrg D** and **TRPV1**, 17.3% and 11.3%.

	PAR-2	PAR-4	TRPV1	TRPA1	MRGPRX1	MRGPRX2	MRG D
A		V			V	V	
B	V						
C			V				V

V: inhibition effect

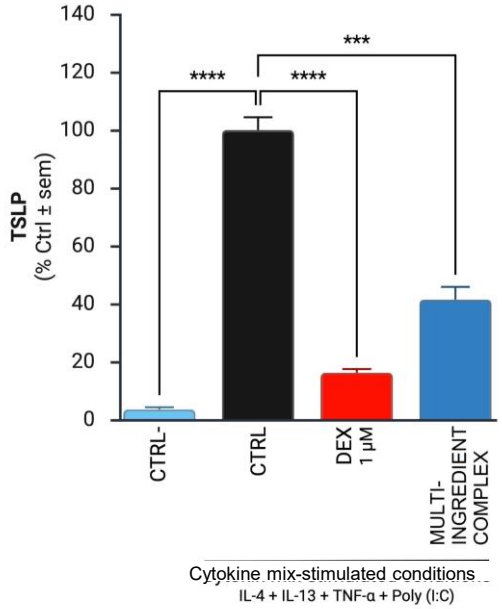


Ingredient A : Bacteria lysate of *Vitreoscilla filiformis* cultivated in LRP Thermal Spring Water
Ingredient B: Anti-protease mix peptide
Ingredient C: Made of oligosaccharides obtained from *Ophiopogon japonicum tuberous roots*

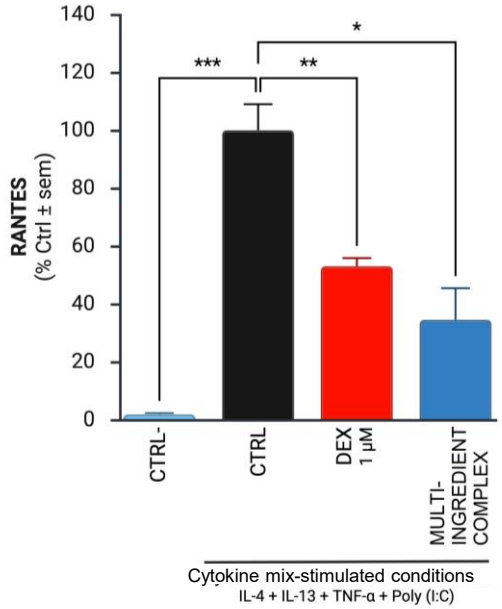
RESULTS

EXPERIMENT 2

In a reconstructed human epidermis model mimicking atopic dermatitis, significant inhibitory effects on the release of TSLP (thymic stromal lymphopoietin), RANTES (regulated on activation, normal T-cell expressed and secreted), and CCL2 (C-C Motif Chemokine Ligand 2) were observed distinctly with individual ingredients.
The multi-ingredient complex further enhanced these anti-inflammatory effects, TSLP ($p<0.001$), RANTES ($p<0.05$) and CCL-2 ($p<0.05$) levels were reduced with no significant difference *vs* dexamethasone known as anti-inflammatory compound (positive control).



*P<0.05 ; **P<0.01; *** P<0,001
Multi-ingredient complex : [A:B:C] (0.4 : 0.06 : 0.1 mg/ml)



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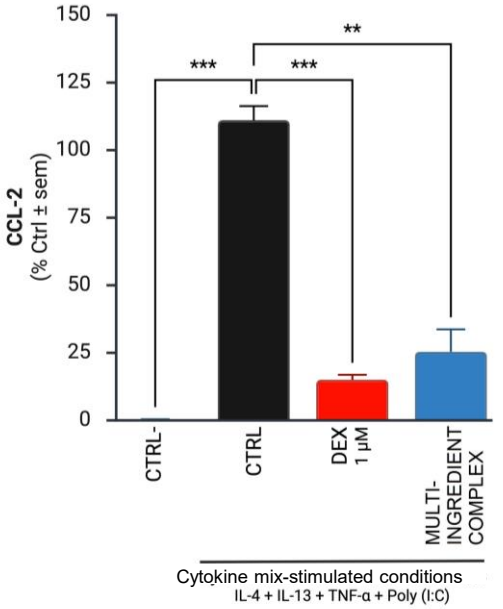

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CONCLUSION

The novel multi-ingredient complex effectively targets multiple itch pathways and reduces inflammatory mediator release, offering a promising multi-target approach to break the itch-scratch cycle and neuroinflammation in AD in the future.

Acknowledgements

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References:

1. Steinhoff M, et al. Breaking the Itch-Scratch Cycle: An Integrative Approach for Atopic Dermatitis. Dermatitis. 2025 Aug 14
2. Augustin M, et al. Characterizing treatment-related patient needs in atopic eczema: insights for personalized goal orientation. J Eur Acad Dermatol Venereol. 2020 Jan;34(1):142-152.
3. Mack MR, Kim BS. The Itch-Scratch Cycle: A Neuroimmune Perspective. Trends Immunol. 2018 Dec;39(12):980-991.
4. Rinaldi G. The Itch-Scratch Cycle: A Review of the Mechanisms. Dermatol Pract Concept. 2019 Apr 30;9(2):90-97.
5. Tominaga M, Takamori K. Peripheral itch sensitization in atopic dermatitis. Allergol Int. 2022 Jul;71(3):265-277.
6. Mollanazar NK, Smith PK, Yosipovitch G. Mediators of Chronic Pruritus in Atopic Dermatitis: Getting the Itch Out? Clin Rev Allergy Immunol. 2016 Dec;51(3):263-292. .