Baricitinib Restores Epidermal Barrier Gene Expression in IL-23–Driven Psoriasiform Inflammation Across Ex Vivo, In Vivo, and Human Keratinocyte Models

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Background

Psoriasis is a chronic immune-mediated inflammatory skin disease driven by dysregulated interactions between keratinocytes and immune cells, with a central role for the IL-23/Th17 axis.[1,2] IL-23 promotes Th17 cytokine responses, including IL-17A, IL-17F, and IL-22, which contribute to keratinocyte activation, inflammatory amplification, and epidermal barrier disruption.[1,2]

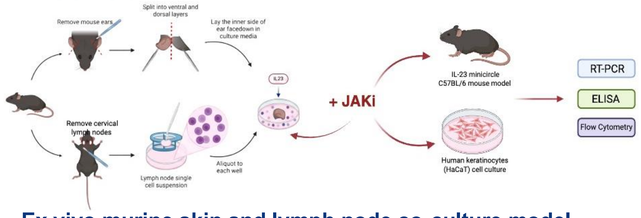
In addition to inflammatory activation, psoriatic inflammation is associated with impaired epidermal barrier gene expression, including suppression of structural barrier genes such as filaggrin and loricrin.[3] Although JAK inhibitors suppress cytokine signaling through JAK/STAT pathways, their effects on epidermal barrier gene regulation remain incompletely understood.[4]

Here, we investigated whether baricitinib, a JAK1/2 inhibitor, suppresses psoriasiform inflammation driven by IL-23 and restores epidermal barrier gene expression across ex vivo, in vivo, and human keratinocyte models.

Objective

To determine whether baricitinib suppresses inflammatory responses driven by IL-23 and restores epidermal barrier gene expression using an ex vivo murine skin and lymph node co-culture model, an in vivo IL-23 minicircle mouse model, and HaCaT human keratinocytes.

Methods



Ex vivo murine skin and lymph node co-culture model

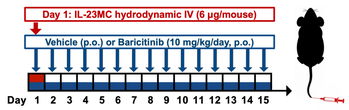
C57BL/6 mouse ear skin was co-cultured with cervical lymph node cells in the presence of recombinant IL-23. After 4–72 hours, ear skin samples were collected for RNA extraction and RT-qPCR analysis of inflammatory and epidermal barrier-associated genes. Culture supernatants were collected for IL-17A ELISA.

JAK inhibitor treatment in the ex vivo co-culture model

Co-cultures stimulated with IL-23 were treated with JAK inhibitors, including baricitinib, upadacitinib, and tofacitinib, at 250 nM or 1,000 nM. Betamethasone (10 μM) was used as a positive anti-inflammatory control. Ear skin samples were collected for RT-qPCR analysis of Th17 cytokine and epidermal barrier-associated genes.

In vivo IL-23 minicircle model and baricitinib treatment

C57BL/6 mice received a hydrodynamic intravenous injection of IL-23 minicircle DNA (6 μg/mouse) on day 1 to induce systemic IL-23 expression and psoriasis-like inflammation.[5,6] Baricitinib (10 mg/kg/day) or vehicle (10% DMSO, 40% PEG300, 5% Tween-80, and 45% saline) was administered once daily by oral gavage from day 1 through day 15. Ear thickness was measured serially. At endpoint, ear tissues were collected for RT-qPCR analysis of inflammatory and epidermal barrier-associated gene expression.

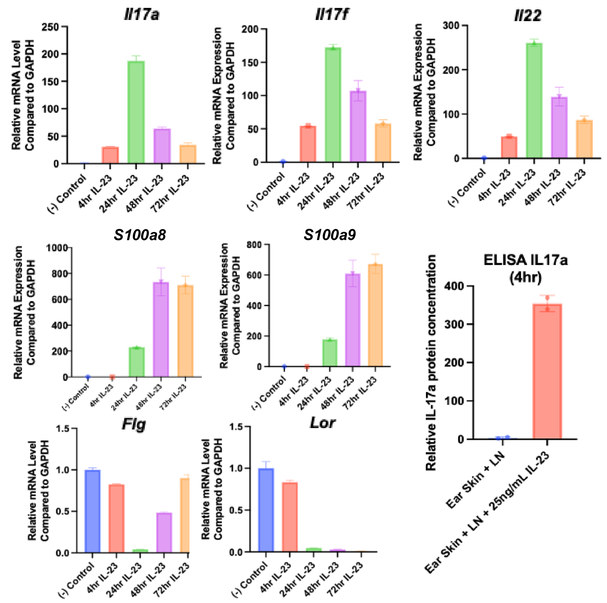


HaCaT human keratinocyte model

HaCaT cells were treated with varying concentrations of baricitinib. Gene expression was analyzed by RT-qPCR, including *FLG* as a structural barrier gene and *MKI67* and *BAX* as markers of proliferation and apoptosis.

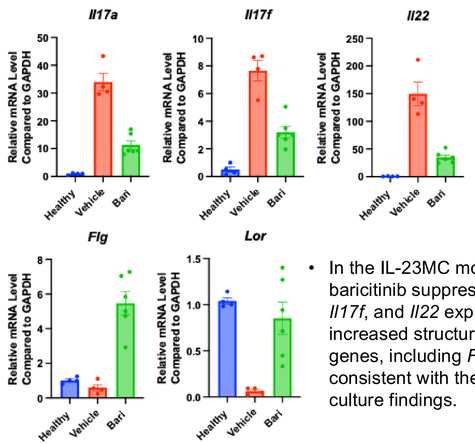
Results

Figure 1. The skin–lymph node co-culture model recapitulates IL-23–driven psoriasiform inflammation



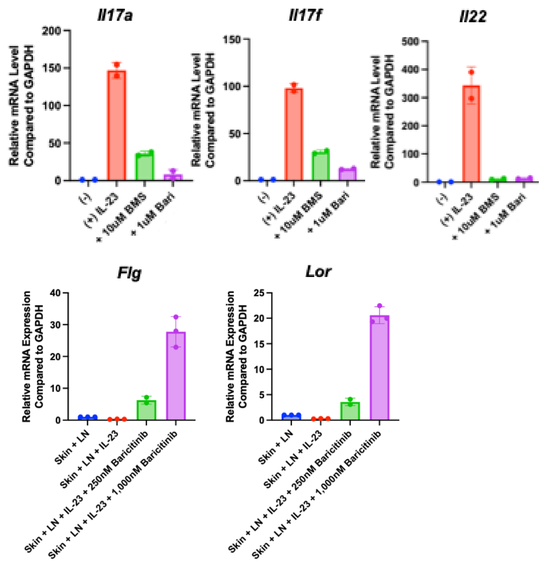
- RT-qPCR analysis showed that recombinant IL-23 induced Th17 cytokine expression in the skin and lymph node co-culture model.
- *Il17a*, *Il17f*, and *Il22* peaked at 24 hours, followed by delayed *S100a8* and *S100a9* induction at 48–72 hours.
- Structural barrier genes, including *Flg* and *Lor*, were markedly suppressed after IL-23 stimulation.
- ELISA further confirmed increased IL-17A protein in culture supernatants as early as 4 hours after IL-23 stimulation.

Figure 3. Baricitinib suppresses inflammatory cytokines and restores barrier genes in the IL-23MC mouse model



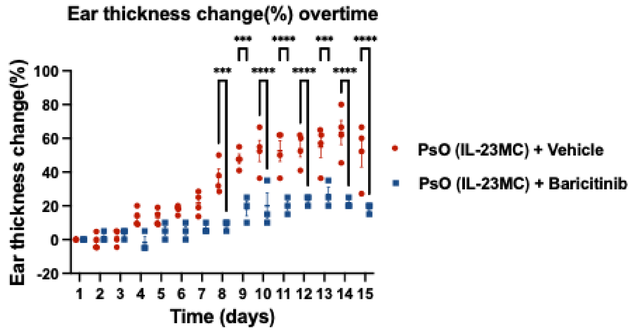
- In the IL-23MC mouse model, baricitinib suppressed *Il17a*, *Il17f*, and *Il22* expression and increased structural barrier genes, including *Flg* and *Lor*, consistent with the ex vivo co-culture findings.

Figure 2. Baricitinib suppresses Th17 cytokine expression and restores structural barrier genes in the ex vivo co-culture model



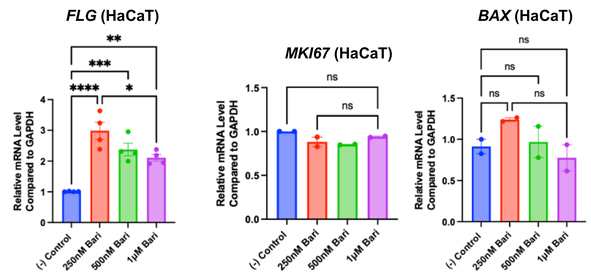
- In the skin and lymph node co-culture model stimulated with IL-23, baricitinib markedly reduced Th17 cytokine gene expression, including *Il17a*, *Il17f*, and *Il22*, with an effect comparable to betamethasone.
- Baricitinib also increased the expression of structural barrier genes, including *Flg* and *Lor*, suggesting that baricitinib suppresses inflammation induced by IL-23 while restoring epidermal barrier gene expression.

Figure 4. Baricitinib reduces ear thickness change in the IL-23MC mouse model



- Baricitinib significantly reduced the percentage change in ear thickness compared with vehicle from Day 8 onward (mixed-effects model with Šídák's multiple comparisons test, adjusted $p < 0.05$).

Figure 5. Baricitinib increases *FLG* expression in HaCaT human keratinocytes



- Baricitinib increased *FLG* expression in HaCaT human keratinocytes without significant changes in *MKI67* or *BAX*.
- These findings suggest that baricitinib increases *FLG* expression independently of major changes in proliferation or apoptosis and may have a direct effect on epidermal barrier gene regulation in keratinocytes.

Discussion

The skin and lymph node co-culture model recapitulated key features of psoriasiform inflammation driven by IL-23, including Th17 cytokine induction, delayed upregulation of downstream inflammatory genes, and marked suppression of structural barrier genes. This supports the utility of the model as an ex vivo platform for evaluating interactions between keratinocytes and immune cells and epidermal barrier disruption.

Baricitinib suppressed Th17 cytokine gene expression induced by IL-23 and restored structural barrier genes in the ex vivo co-culture model. These findings were also observed in the in vivo IL-23MC mouse model, where baricitinib reduced ear thickness change and increased expression of barrier-associated genes, including *Flg* and *Lor*. Together, these results suggest that baricitinib improves psoriasiform inflammation driven by IL-23 through both anti-inflammatory effects and barrier restoration.

In HaCaT human keratinocytes, baricitinib increased *FLG* expression without significant changes in *MKI67* or *BAX*, suggesting that *FLG* induction was not simply secondary to altered proliferation or apoptosis. These findings support a possible intrinsic effect of baricitinib in keratinocytes on epidermal barrier gene regulation.

Overall, baricitinib may contribute to improvement of psoriatic disease not only by suppressing inflammatory cytokine signaling, but also by promoting epidermal barrier gene recovery.

References

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