Approved and investigational JAK/TYK2 inhibitors attenuate inflammatory pathways and promote mucosal homeostasis in IBD patient-derived organoids

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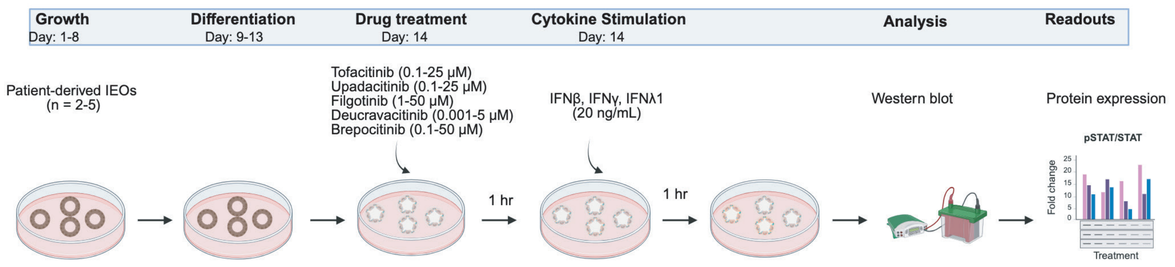
Background & Research Questions

Janus kinase (JAK)-Signal Transducer and Activation of Transcription (STAT) pathway is a key regulator of inflammatory signaling in ulcerative colitis (UC). JAK inhibitors prevent signaling downstream of multiple pro-inflammatory cytokines[1]. While most studies have focused on how JAK inhibitors suppress immune cell mediated responses, their direct effect on the epithelium remains less known.

- How does the selectivity of five selected JAK inhibitors, tofacitinib, upadacitinib, filgotinib, brepocitinib and deucravacitinib differ in inhibiting the interferon-mediated activation of STAT1, STAT3, and TYK2 in intestinal epithelial organoids (IEOs)?
- What are the key transcriptional programs and biological pathways induced in IEOs following exposure to interferon (IFN)γ, IFNλ1, TNF and IFNγ + TNF?
- Do pretreatment with either upadacitinib (JAK1-specific) or deucravacitinib (TYK2-specific) modulates these IFNγ, IFNλ1, TNF and IFNγ + TNF-induced transcriptional signatures?

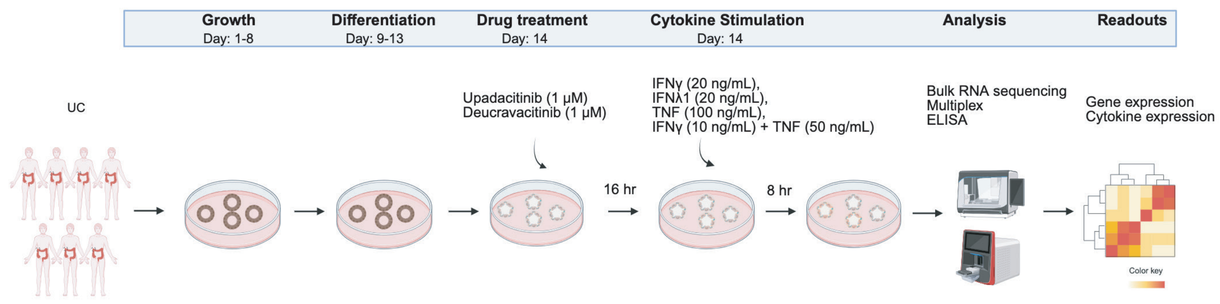
Results

1 JAK/TYK2 inhibitors dose-dependently inhibit phosphorylation of STAT1, STAT3, and TYK2

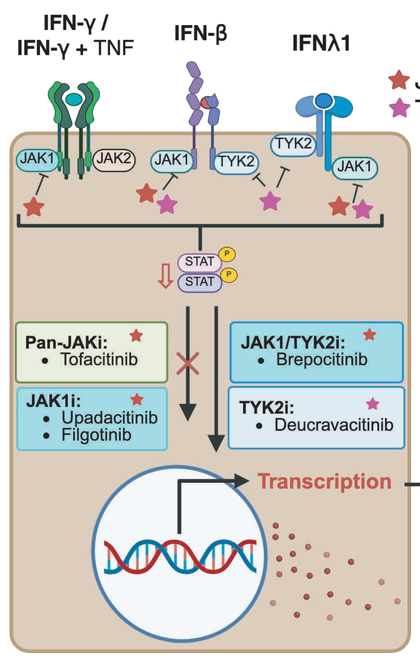


- IFN subtypes trigger unique STAT1 and STAT3 phosphorylation patterns.
- Only IFNβ and IFNλ1 induce TYK2 phosphorylation.
- Potency ranking: Upadacitinib > tofacitinib > brepocitinib > filgotinib > deucravacitinib.

2 Upadacitinib suppresses IFNγ and IFNλ1-induced gene expression, while deucravacitinib has minimal impact on IFNγ stimulation, indicating distinct selectivity in targeting inflammatory pathways.



Mechanisms of action of JAK/TYK2 inhibitors and list of upregulated and downregulated differentially expression genes (DEGs).



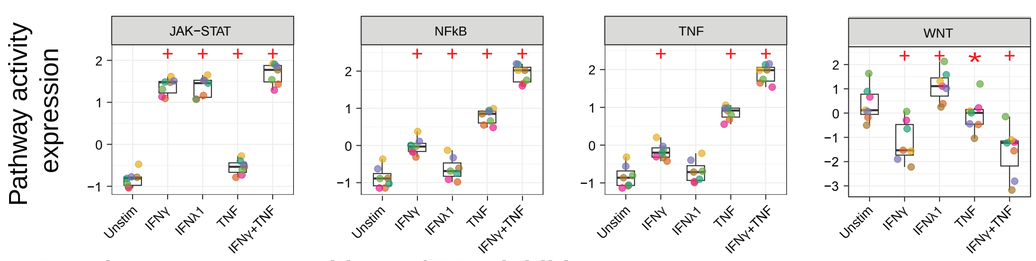
Differentially expressed genes

Contrasts	Number of genes*		
	Unstimulated	Upadacitinib	Deucravacitinib
IFNγ	6357	5389	63
IFNλ1	1069	1369	1131
TNF	4070	9	7
IFNγ + TNF	9067	6988	392

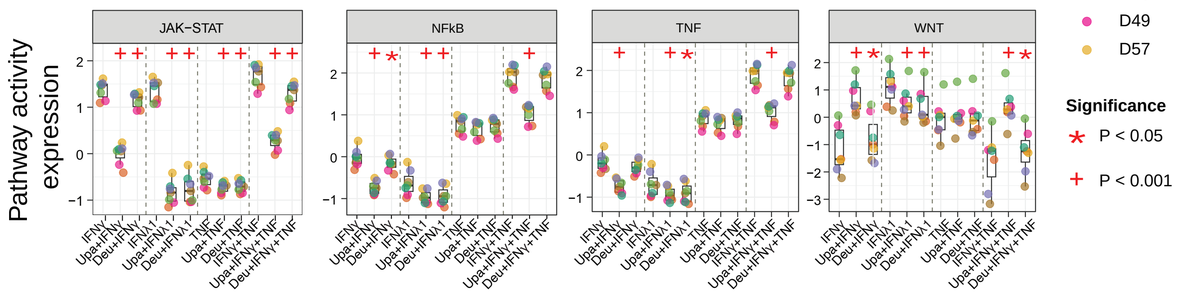
*|log2 foldchange| > 0.3 and p-adjusted < 0.05

3 Upadacitinib inhibits the JAK-STAT, NFkB and TNF pathways while restoring WNT signaling in response to IFNγ, IFNλ1 and IFNγ + TNF stimulation

A Under stimulated condition



B Under pretreatment with JAK/TYK2 inhibitors

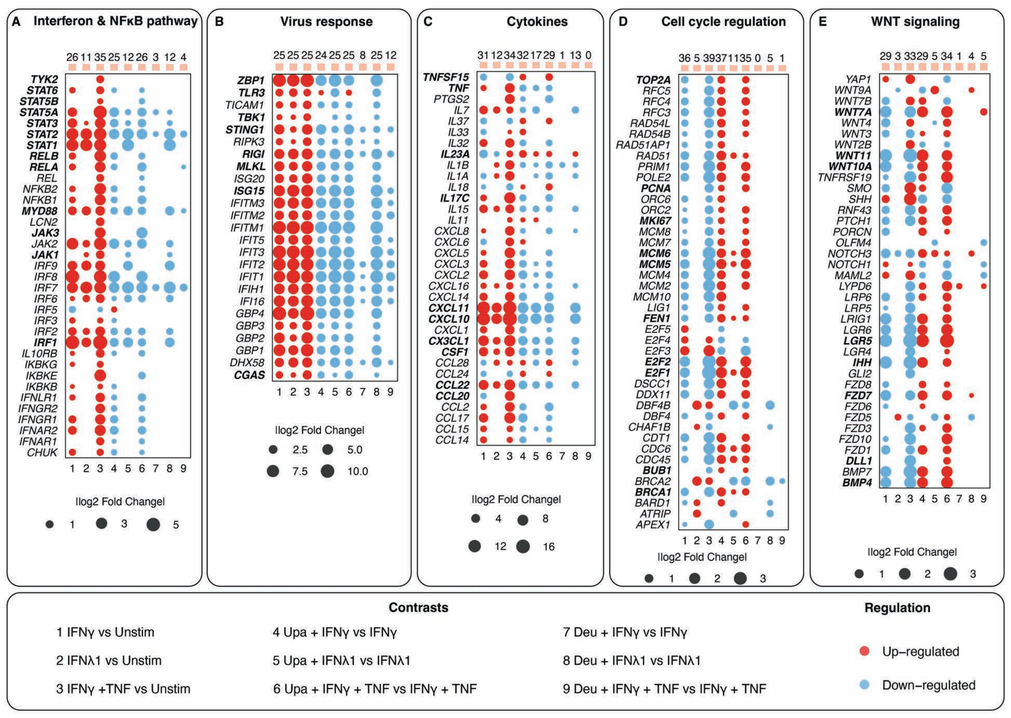


Donors: D16, D48, D52, D65, D40, D49, D57

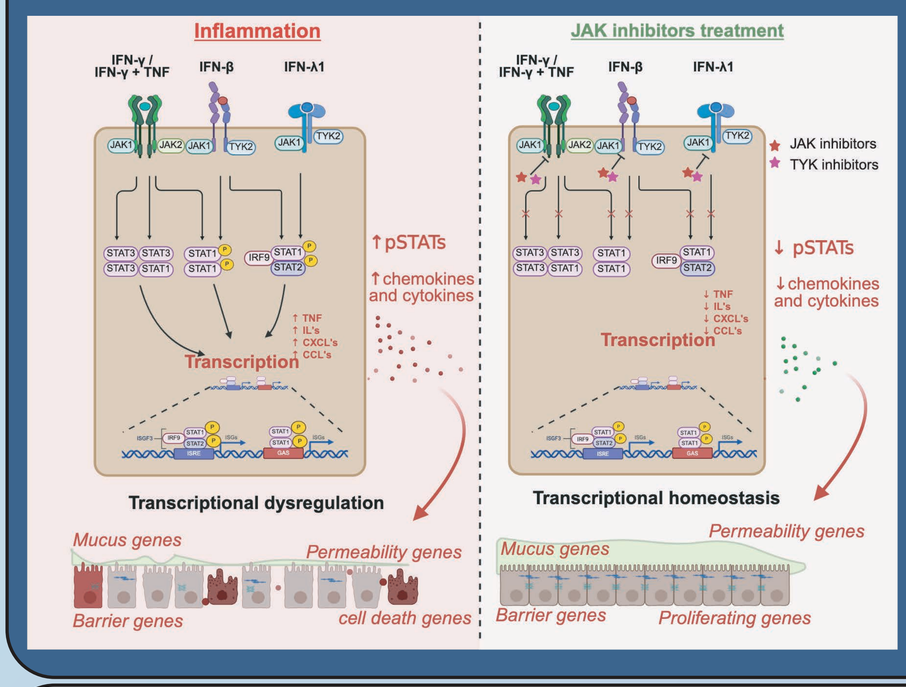
Significance: * P < 0.05; + P < 0.001

Condition

Example of genes associated with inflammatory pathways and WNT and cell cycle regulation processes.



Conclusion



Our comprehensive analysis of JAK/TYK2 inhibitor effects on patient-derived IEOs challenge the current paradigm of JAK inhibitors as solely immune-modulatory agents and reveal their capacity to directly promote genes involved in mucosal healing.

Reference: [1]Hu, X., et al., The JAK/STAT signaling pathway: from bench to clinic. Signal Transduction and Targeted Therapy, 2021. 6(1): p. 402.

Conflict of Interest: The authors declare no conflicts of interest.

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