Spleen Weight
GM-CSF Induced pSTAT5 (JAK2/JAK2)
Dex
Lupus Pathology
Dex
Tofacitinib
Ruxolitinib
Dex NDI-031407
Ear IL-17A
*p<0.05

METHODS
We utilized cutting edge proprietary structure-based drug design tools to identify highly selective Tyk2 inhibitors. These inhibitors were characterized for their drug-like properties, potency and selectivity in enzyme and cellular-based assays, and in mouse models of IBD and psoriasis.

RESULTS
Here we describe the identification of NDI-031407, a small molecule catalytic inhibitor of Tyk2, that exhibits potent enzyme and cellular activity, and is highly selective against other Jak family members and against panels of receptors, transporters, ion channels, CYP enzymes and the HER2 receptor. This compound inhibits Tyk2 with an IC50 of 0.2 μM in a biochemical assay, and is 218-, 146-, and 20-fold selective against Jak1, Jak2, and Jak3, respectively. Cell-based potency and selectivity of NDI-031407 was demonstrated in human PBMC assays by blockade of IL-12 induced phospho-STAT1, GM-CSF induced phospho-STAT5, and IL-2 induced phospho-STAT5, with IC50 of 0.10 μM, 4.1 μM and 0.25 μM, respectively. In addition, NDI-031407 inhibited IL-12 induced IFN-γ and IL-2 induced phospho-STAT5, with IC50 of 2.7 μM and 7.0 μM in human and mouse whole blood, respectively. We investigated the in vivo efficacy of NDI-031407 in mouse models of psoriasis and IBD. In an IL-12-induced mouse ear inflammation model, NDI-031407 dose-dependently reduced disease with up to 74% inhibition of ear swelling and 96% inhibition of tissue levels of IL-17A at 100 mg/kg, highlighting the crucial role of Tyk2 inhibition in Th17 mediated pathology with IL-23+T cells. In addition, NDI-031407 was highly efficacious in an imiquimod-induced mouse psoriasis model. NDI-031407 at 100 mg/kg achieved the same effect as dexamethasone in reduction of psoriasis score without body weight reduction. NDI-031407-treated mice had improved skin histology and dose-dependent reduction of spleen weight. To investigate the role of Tyk2 inhibition in Th1-driven pathology, we tested NDI-031407 in a CD4+T cell induced autoimmune arthritis model that resembles the pathology of human Crohn’s disease. NDI-031407 treatment improved disease outcomes by reduction of body weight loss and colonic weight/length ratio, and improved colon histology. 100 mg/kg of NDI-031407 treatment also reduced colon myeloperoxidase (MPO) activity in the colon and spleen weight. We also demonstrated that NDI-031407 is efficacious in suppressing diseases with Th1 and Th17 pathogenic mechanisms. The preclinical safety profile of NDI-031407 is currently under evaluation.

CONCLUSION
Utilizing unique and innovative structure-based drug design technologies, we rapidly designed highly selective and potent Tyk2 inhibitors with superior pharmacological properties, potent therapeutic efficacy in inflammatory disorders. We validated the vital role of Tyk2 in disease pathogenesis of psoriasis and IBD in preclinical mouse models.

**BACKGROUND**
Tyk2, a member of the Jak family kinases, is a key mediator of Th17 and Th1 Pathogenesis.

**METHODS**
We utilized cutting edge proprietary structure-based drug design tools to identify highly selective Tyk2 inhibitors. These inhibitors were characterized for their drug-like properties, potency and selectivity in enzyme and cellular-based assays, and in mouse models of IBD and psoriasis.

**RESULTS**
Here we describe the identification of NDI-031407, a small molecule catalytic inhibitor of Tyk2, that exhibits potent enzyme and cellular activity, and is highly selective against other Jak family members and against panels of receptors, transporters, ion channels, CYP enzymes and the HER2 receptor. This compound inhibits Tyk2 with an IC50 of 0.2 μM in a biochemical assay, and is 218-, 146-, and 20-fold selective against Jak1, Jak2, and Jak3, respectively. Cell-based potency and selectivity of NDI-031407 was demonstrated in human PBMC assays by blockade of IL-12 induced phospho-STAT1, GM-CSF induced phospho-STAT5, and IL-2 induced phospho-STAT5, with IC50 of 0.10 μM, 4.1 μM and 0.25 μM, respectively. In addition, NDI-031407 inhibited IL-12 induced IFN-γ and IL-2 induced phospho-STAT5, with IC50 of 2.7 μM and 7.0 μM in human and mouse whole blood, respectively. We investigated the in vivo efficacy of NDI-031407 in mouse models of psoriasis and IBD. In an IL-12-induced mouse ear inflammation model, NDI-031407 dose-dependently reduced disease with up to 74% inhibition of ear swelling and 96% inhibition of tissue levels of IL-17A at 100 mg/kg, highlighting the crucial role of Tyk2 inhibition in Th17 mediated pathology with IL-23+T cells. In addition, NDI-031407 was highly efficacious in an imiquimod-induced mouse psoriasis model. NDI-031407 at 100 mg/kg achieved the same effect as dexamethasone in reduction of psoriasis score without body weight reduction. NDI-031407-treated mice had improved skin histology and dose-dependent reduction of spleen weight. To investigate the role of Tyk2 inhibition in Th1-driven pathology, we tested NDI-031407 in a CD4+T cell induced autoimmune arthritis model that resembles the pathology of human Crohn’s disease. NDI-031407 treatment improved disease outcomes by reduction of body weight loss and colonic weight/length ratio, and improved colon histology. 100 mg/kg of NDI-031407 treatment also reduced colon myeloperoxidase (MPO) activity in the colon and spleen weight. We also demonstrated that NDI-031407 is efficacious in suppressing diseases with Th1 and Th17 pathogenic mechanisms. The preclinical safety profile of NDI-031407 is currently under evaluation.

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