2002

Presented at:
2018 Keystone Symposia Conference
March 24–28, 2018
Royal Dublin Society, Dublin, Ireland

Probing the Role of the MC1 Receptor in Resolution With a Selective MC1 Receptor Agonist—PL-8177

Carl Spana,[1] Marie Mahklina,[1] Wei Yang,[1] John Dodd[1] | [1]Palatin Technologies, Inc, Cranbury, NJ, USA

Melanocortin-1 (MC1) Receptor Agonists for Irritable Bowel Disease (IBD)

- MC1 receptor agonism is an endogenous mechanism that downregulates inflammatory/immune responses[1]
- MC1 receptors are upregulated in IBD and expressed on the cell surface of intestinal epithelia[2]
- Alpha-melanocortin stimulating hormone (α-MSH) is an endogenous agonist of 4 of the 5 melanocortin receptors (MC1, and melanocortin-3, -4, and -5), and was shown in numerous animal models to prevent and reverse intestinal inflammation[1,2]
- The MC1 receptor has the highest affinity for α-MSH[3]
- However, it is difficult to determine which specific melanocortin receptor is responsible for the effects of α-MSH on intestinal inflammation
- Experimental studies were conducted to determine whether PL-8177, a highly selective MC1 receptor agonist (**Table 1**), demonstrates actions and efficacy similar to those of α-MSH in preventing and reversing intestinal inflammation

Table 1. In Vitro Selectivity of PL-8177 Compared With Endogenous MC1 Receptor Agonists

	Functional-CEREP (EC50; nM)				
	MC1r	MC2r	MC3r	MC4r	MC5r
α-MSH	4.47	>10,000	9.8	10.8	560
ACTH	980	4.8	390	350	4100
PL-8177	0.57	>10,000	>10,000	510	>10,000

ACTH, adrenocorticotropic hormone; α-MSH, alpha-melanocortin stimulating hormone; MC1, melanocortin-1.

PL-8177: Preclinical Proof of Principle Overview

- A series of studies with PL-8177 were conducted to assess/establish:
  - — In vitro activity and safety profile
  - — Preclinical proof of principle/proof of mechanism data
  - — Formulation assessments to allow oral dosing

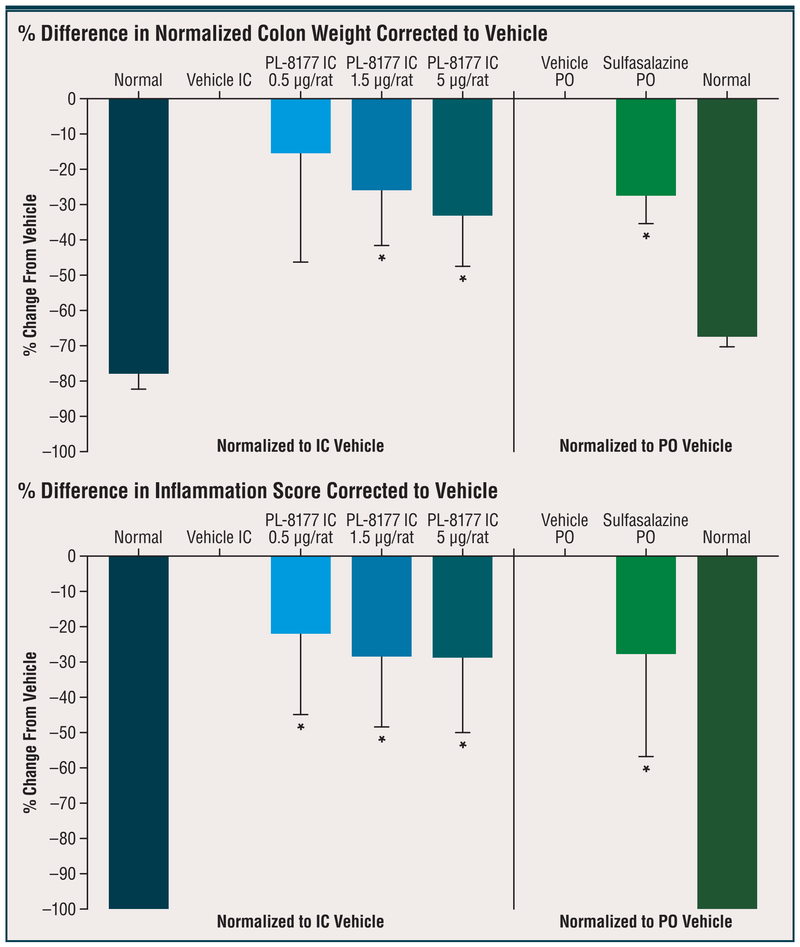
In Vitro Activity and Safety Data

- PL-8177 demonstrated similar inhibition of lipopolysaccharide-induced tumor necrosis factor alpha inhibition compared with the endogenous MC1 receptor agonists α-MSH and adrenocorticotropic hormone (**Figure 1**)
- In a Eurofins lead profile (© Eurofins Scientific 2018), no activity in any of 72 in vitro assays at 10 μM was detected; highlights included no activities in:
  - — Cytochrome P450 enzymes 1A2, 2C19, 2C9, 2D6, and 3A4
  - — Potassium channel hERG
  - — Any of 7 adrenergic receptor subtypes
  - — Any remaining assays included in the panel

Proof of Principle Data

- PL-8177 was evaluated in a cannulated rat model of bowel inflammation
  - — Dinitrobenzene sulfonic acid (DNBS) was administered rectally as a solution in male, 200g Wistar rats to induce inflammation of the bowel lumen
  - — The rats were implanted with a catheter in the proximal part of the ascending colon, which exited out the nape of the neck for dosing access
  - — In groups of 10, the rats were dosed at: 0.5 μg and 5.0 μg PL-8177 and vehicle (sterile water) via intracolonic injection at 24 h, 12 h, and 2 h before and 6 h after DNBS challenge, followed by twice-daily dosing for 5 consecutive days through day 7
  - — Non-cannulated control rats were administered sulfasalazine (positive controls), and vehicle (untreated controls)
- In the DNBS rat model of bowel inflammation, PL-8177 was as active as sulfasalazine (standard of care), and superior to untreated controls, in reducing parameters of bowel inflammation (colon weight and inflammation score) (**Figure 2**)

Figure 2. Effects of PL-8177 and Sulfasalazine on Colon Weight and Inflammation Score in Rats with DNBS-Induced Bowel Inflammation



*$P<0.05$. DNBS, dinitrobenzene sulfonic acid; IC, intracutaneous; PO, oral.

Oral Formulation Assessments

- The pharmacokinetics and pharmacodynamics of an oral capsule formulation of PL-8177 for colon release were evaluated in rats
  - — 24 total Sprague-Dawley rats weighing between 250–350 grams, 7–9 weeks old
  - — Fasted overnight prior to oral dosing with 0.1-mg single capsule of PL-8177; food and water ad libitum
  - — Intestinal and colon contents were collected at specific time points (n=20) or after testing (n=4)
- The oral PL-8177 formulation was released in the colon and progressed through the rat intestinal tract in 9 hours (**Figure 3**)
- In a DNBS model of colitis in rats, oral capsule PL-8177 was evaluated dosed at 10 μg, 20 μg, and 50 μg twice daily (bid), compared with vehicle and sulfasalazine treatment
- The baseline-corrected inflammation score and macroscopic damage score were both significantly lower (improved) with PL-8177 50 μg versus vehicle, and to a similar degree as sulfasalazine (**Figure 4**)

Figure 3. Progression of PL-8177 Via Oral Capsule Administration Through the Rat Intestinal Tract

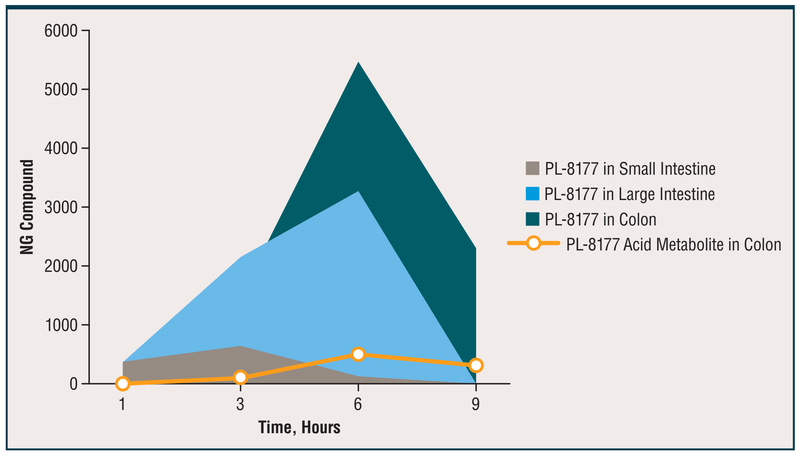
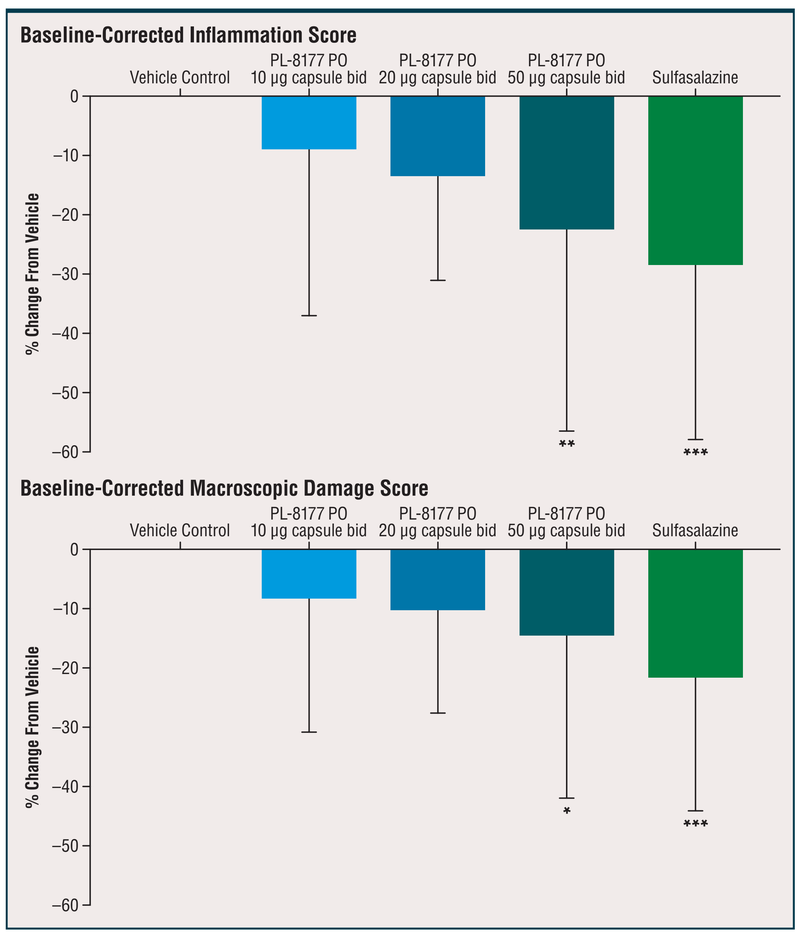
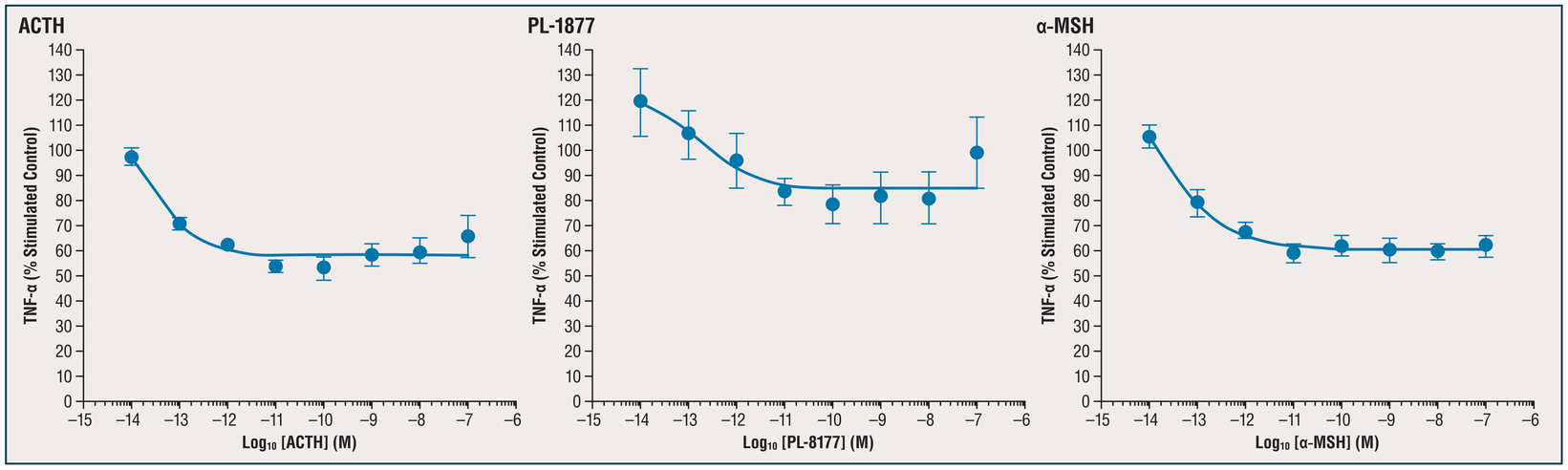


Figure 4. Effects of Orally Administered PL-8177 Capsule on Parameters of DNBS-Induced Colitis in Rats



*$P<0.05$; **$P<0.01$; ***$P<0.001$. DNBS, dinitrobenzene sulfonic acid.

Figure 1. Lipopolysaccharide-Induced Tumor Necrosis Factor Alpha Inhibition in Human Whole Blood



α-MSH, alpha-melanocortin stimulating hormone; ACTH, adrenocorticotropic hormone; TNF-α, tumor necrosis factor alpha.

Conclusions

- PL-8177 is a highly selective MC1 receptor agonist with in vitro actions similar to those of endogenous MC1 receptor agonists such as α-MSH and ACTH
- In a cannulated rat model of DNBS-induced bowel inflammation, PL-8177 administered via catheter reduced inflammation and colon weight scores to a similar degree as sulfasalazine
- An orally administered capsule formulation of PL-8177, released in the colon, also improved inflammation scores in rats with DNBS-induced bowel inflammation, without systemic exposure
- Based on these data, a per-oral colon release formulation of PL-8177 will be developed for clinical evaluation

Disclosures Carl Spana, Marie Mahklina, Wei Yang, and John Dodd are employees of Palatin Technologies, Inc.

Support These studies were funded by Palatin Technologies, Inc. Editorial assistance was provided by The Curry Rockefeller Group, LLC, which was funded by Palatin Technologies, Inc.

References **1.** Ahmed TJ, et al. Curbing inflammation through endogenous pathways: focus on melanocortin peptides. *Int J Inflamm.* 2013;2013:985815. **2.** Maaser C, et al. Crucial role of the melanocortin receptor MC1R in experimental colitis. *Gut.* 2006;55(10):1415–1422. **3.** Mountjoy KG. The human melanocyte stimulating hormone receptor has evolved to become "super-sensitive" to melanocortin peptides. *Mol Cell Endocrinol.* 1994;102(1–2):R7–R11.