The Prevention of Colitis by E Prostanoid Receptor 4 Agonist through Enhancement of Epithelium Survival and Regeneration

Guang-Liang Jiang, Amelia Nieves, Wha Bin Im, David W. Old, Danny T. Dinh, and Larry Wheeler

Departments of Biological Sciences (G.-L.J., A.N., W.B.I., L.W.) and Medical Chemistry (D.W.O., D.T.D.), Herbert Research Center, Allergan, Inc., Irvine, California

Received July 20, 2006; accepted September 27, 2006

ABSTRACT

Inflammatory bowel disease (IBD) is often triggered and/or exacerbated by nonsteroidal anti-inflammatory drugs (NSAIDs). Among various prostanoids affected by NSAIDs, prostaglandin E₂ (PGE₂), in particular, seems to play critical roles in IBD via the EP4 receptor, one of the four PGE₂ receptor subtypes (EP₁–₄). An EP₄ agonist, [3-[(1R,2S,3R)-3-hydroxy-2-[1E,3S]-3-hydroxy-4-[3-(methoxymethyl)phenyl]-1-buteny]-5-oxocyclopentyl][thio]propyl[thio]-acetic acid, C₂₂H₃₀O₆S₂ (ONO-AE1–329), for example, when topically applied, has been reported to ameliorate typical colitis symptoms by suppressing the production of cytotoxic cytokines in the dextran sodium sulfate (DSS)-induced colitis model. EP₄ agonists are also known, however, for their ability to protect epithelial cells from apoptosis in vitro, which may contribute to the protection of mucosal barrier functions. To investigate this potential application, we have tested another EP₄-selective agonist in the DSS-indomethacin mouse colitis model. 7-[2-(3-Hydroxy-4-phenyl-but-1-eny]-6-oxo-piperidin-1-yl]-heptanoic acid methyl ester, C₂₃H₃₃NO₄ (AGN205203), an analog from the 8-aza-pipiderinone series of EP₄ agonists, is metabolically and chemically more stable than the ONO agonist, because of its lack of oxidizable sulfur atoms in the α-chain and of 11-OH group, a potential source of β-elimination reaction. Treatment of mice subcutaneously with AGN205203 at 3 mg/kg/day minimized colitis symptoms, such as weight loss, diarrhea, and colonic bleeding. Further histological examination of colons revealed healthy surface columnar epithelial cells free of erosion and ulceration compared with those without the drug treatment. At cellular level, the drug treatment decreased colon epithelial apoptosis, prevented goblet cell depletion, and promoted epithelial regeneration. AGN205203 may be unique among known EP₄ agonists for its metabolic and chemical stability, and it is amenable to systemic applications for the prevention and recovery of IBD.

Inflammatory bowel disease (IBD), including ulcerative colitis (UC) and Crohn’s disease (CD), affect approximately 1.4 million United States patients with 15,000 to 30,000 new cases annually at a mean age between 30s and early 40s. IBD patients suffer from body weight loss, diarrhea, fecal blood, and pain. Such symptoms could last for 15 to 25 years, alternatively between exacerbation and remission, thus severely affecting the quality of patient life and retarding the growth of young patients (Loftus, 2004; Isaacs et al., 2005). General consensus in the field is that IBD may arise from compromised colonic mucosal barrier functions that allow colonic antigens access to submucosal monocytes, which, upon activation, initiate innate immune responses and trigger cytotoxic cytokine production. Prevention and recovery of IBD thus largely depend on the integrity and maintenance of colonic mucosal barrier functions, which are compromised by inflammations along the entire bowel wall in CD and at the mucosal surface in UC. Current therapies primarily aim at the symptomatic remission by anti-inflammatory agents such as aminosalicylates and/or immunosuppressive agents, such as steroids, purine analogs, and monoclonal antibodies against cytokotins (Colombel et al., 2004; Isaacs et al., 2005). These thera-
pies have been successful in alleviating symptoms during outbreaks, but they have not been effective in maintaining remission and preventing relapses under various regimens, including combination therapies (Colombel et al., 2004; Loftus et al., 2004; Isaacs et al., 2005). Moreover, severe side effects of immunosuppressants limit their long-term use (Colombel et al., 2004; Loftus et al., 2004; Isaacs et al., 2005). Eventually, 25% UC and 60% CD patients have to undergo colectomy in 10 to 15 years after the onset of IBD (Bernstein and Nalbantoglu, 2006).

Such inadequacies in current therapies could arise from their failure to mend compromised colonic mucosal barrier functions involving epithelial cell layers, mucus, and goblet cells in IBD patients. This hypothesis is supported by several articles reporting that the impaired integrity of colonic mucosal epithelial barrier is central in the course of IBD (Stein et al., 1998; Lim and Hanauer, 2004). Therefore, strengthening of epithelial resistance to noxious stimuli and enhancement of intestinal repair would provide novel and effective approaches for the treatment of IBD (Dighe, 2001; Yu et al., 2004).

NSAIDs and COX-2 inhibitors often trigger and worsen IBD in humans (Thiefin and Beaugerie, 2005; Meyer et al., 2006). Among various prostanoids affected by COX inhibitors, PGE₂ has received much attention because of its important roles in gastrointestinal physiology. Functionally, PGE₂ interacts with the four receptor subtypes (EP1–4). EP1 and EP3 primarily contribute to inflammatory responses, whereas EP4 promotes both cell survival and growth by activating antiapoptotic and proliferative cellular signaling pathways (Fujino et al., 2003; Hase et al., 2003; Hoshino et al., 2003; Goulet et al., 2004; Joseph et al., 2005). Moreover, EP4 is constitutively expressed in the colonic epithelium and further induced during IBD, along with PGE₂ (Wiercinska-Drapalo et al., 1999; Northey et al., 2000; Takafuji et al., 2000; Nitta et al., 2002). Recently, EP4 antagonists have been reported to impair epithelial proliferation in the colon (Kabashima et al., 2002). Few studies, however, have delineated effects of EP4-selective agonists on the survival of epithelia and Goblet cells during IBD. Earlier studies primarily concerned immunosuppressive effects of EP4 agonists (Nitta et al., 2002). For example, an EP4 agonist, ONO-AE1–329, inhibited the production of cytotoxic cytokines when topically applied in the dextran sodium sulfate (DSS)-induced colitis model.

In this study, we examined the effect of an EP4-selective agonist, AGN205203, on colonic epithelial and Goblet cells in the mouse DSS-Indo colitis model. The drug is metabolically and chemically more stable than the ONO agonist, because it has no oxidizable sulfur atoms in the α-chain and no 11-OH group, a potential source of β-elimination reaction that abolishes its interaction with EP4 (Fig. 1) (Old and Dinh, 2004).

![Fig. 1. Chemical structure of the piperidinone analog AGN205203.](image)

**Materials and Methods**

**Ligand Binding and Functional Tests.** Competition binding experiments were performed in a medium containing Hanks’ balanced salt solution, 20 mM HEPES, pH 7.3, 2 × 10⁵ cells or membranes (−80 µg of protein) from HEK-293 cells stably expressing the human EP4 receptor, 10 nM [3H]PGE₂, and various concentrations of test compounds in a total volume of 300 µL as described in detail previously (Alberts et al., 2003). cAMP assay was carried out using AlphaScreen cAMP assay kits (PerkinElmer Life and Analytical Sciences, Boston, MA) following the manufacturer’s instructions. Intracellular Ca²⁺ was monitored using a fluorometric imaging plate reader Tetra system and assay kits from Molecular Devices (Sunnyvale, CA) following manufacturer’s instructions. All assays were carried out in HEK-293 cells heterologously and stably expressing each of the eight human recombinant prostaglandin receptors. For Ca²⁺ signals, hEP2, hEP4, and hDP were coexpressed with a chimeric G protein, Gαq, which converts Gs signal to Ga ααα ααα, and hEP3 with a chimeric G protein, Gαq (Hata and Breyer, 2004). Each receptor-selective agonist induced Ca²⁺ signals with subnanomolar or nanomolar EC₅₀ values. Subtype-selective compounds used here are PGE₂ for EP1, EP2, EP3, and EP4; BW245C for DP; 17-phenyl prostaglandin F₂α for F prostaglandin receptor, carbacyclin for prostanoyl receptor, and U-46619 for thromboxane receptor.

**Animals and Treatments.** Female C57BL/6 mice at 8 weeks were purchased from Charles River Laboratories, Inc. (Wilmington, MA). Animal housing and handling procedures were approved and performed according to the guidelines of Allergan’s animal care and use committee. All of the chemicals and reagents were obtained from Sigma-Aldrich (St. Louis, MO), unless noted otherwise.

Colitis was induced in mice with 3 or 5% DSS in drinking water with or without indomethacin. DSS with the molecular weight of 8000 is known to induce colitis at the proximal colon (Kitajima et al., 2000), the most affected region of IBD. The stock solution of indomethacin was prepared in dimethyl sulfoxide with trace Tween 80, and it was added to DSS solution to a final concentration of 4 mg/kg/day (Takeuchi et al., 1986; Kabashima et al., 2002). Normal control group was given drinking water containing only vehicle for 7 days.

AGN205203, the methyl ester of AGN205204, was chosen for the subcutaneous application here, because the ester is readily hydrolyzed to the parent acid in the plasma by endogenous esterases, but it would provide more options for future applications such as subcutaneous depots and transdermal patches because of its more optimal pharmacokinetics and favorable metabolism compared to the parent acid (Old and Dinh, 2004).

**Histology and Immunological Staining.** Colons were divided into proximal, middle, and distal portions and embedded with paraffin. Sections of 5 µm in thickness were prepared and stained with H&E from each portion and examined under the microscope. To observe goblet cells, Alcian Blue staining was used. In brief, sections...
were incubated in 1% Alcian Blue in 3% acetic acid, pH 2.5, for 30 min and 0.1% nuclear Fast Red for 10 s. Alcian Blue-positive goblet cells were counted over the size of a grid (1-mm grid with total 100 squares) at original magnification, 600X in 10 view fields randomly selected from each slide.

To assess cell apoptosis in colon, sections were processed with TUNEL staining according to manufacturer's instruction (Chemicon International, Temecula, CA). In brief, after deparaffinizing, slides were digested with 20 μg/ml proteinase K at 37°C for 10 min. Working strength terminal deoxynucleotidyl transferase was applied at 37°C for 1 h after equilibration. Then, antidigoxigenin conjugated with fluorescence was added. Slides were also counterstained with 37°C for 1 h after equilibration. Then, antidigoxigenin conjugated with fluorescence was added. Slides were also counterstained with 3% H2O2 in methanol for 10 min. Microwave, cooled down at room temperature, and blocked for endogenous peroxidase activity with 3% H2O2 in methanol for 10 min. The person who stained and evaluated the slides was blinded to treatments of the mice. From each section, 10 view fields were randomly chosen at original magnification, 400X through a fluorescence lens and a UV lens, respectively. Cells positive to TUNEL and DAPI were counted. The percentage of TUNEL-positive cells was calculated. One sample consists of the average of 10 view fields from a section.

Cell proliferation in colon was detected using proliferating cell nuclear antigen (PCNA) using a commercially available kit (Biomed, Foster City, CA). In brief, paraffin-embedded sections were deparaffinized in xylene and rehydrated in ethanol. Antigen was retrieved with citrate buffer, pH 6.0, boiled for 5 min twice in microwave, cooled down at room temperature, and blocked for endogenous peroxidase activity with 3% H2O2 in methanol for 10 min. Biotinylated mouse anti-rat PCNA antibody was applied and incubated 1 h at room temperature, followed by streptavidin-peroxidase-labeled horse anti-mouse secondary antibody for 30 min. 3,3'-Diaminobenzidine chromogen was applied finally. DAPI counterstaining was also used. Ten views from each slide were randomly collected at original magnification, 400X. PCNA-positive cells were counted.

**Statistical Analysis.** Data are presented as the means with S.E.M. Data were analyzed using an unpaired two-tailed t test or one-way analysis of variance. A probability (P value) of less than 0.05 was considered significant.

**Results**

AGN205204, the parent acid of AGN205203, is an EP4-selective agonist from the 8-aza piperidone PGE series. In HEK-293 cells expressing recombinant hEP4, the drug bound hEP4 with a K_i value of 81 nM from competition experiments with [3H]PGE_2 and increased cAMP production with an EC_{50} value of 0.08 nM. In contrast, the drug at 10 μM showed no detectable fluorometric imaging plate reader signals in HEK-293 cells heterologously expressing hEP1, human F prostanooid receptor, human prostacyclin receptor, and human thromboxane receptor and also in hEP2 (G_{q5}), hEP3 (G_{q4}), and hDP (G_{q4}), where a chimeric G protein in parentheses was coexpressed, either G_{q4} or G_{q4}, to convert G_{i} or G_{s} coupling to Ca\(^{2+}\) signal, respectively (Hata and Breyer, 2004).

Mice were treated with 3% DSS with or without indomethacin (4 mg/kg/day) in drinking water. The 3% DSS treatment alone showed no considerable effects on body weight, loss of stool consistency, and fecal blood (Fig. 2). Only the combined treatment with DSS and indomethacin led to considerable body weight loss, increased diarrhea score, and colonic bleeding, compared with mice treated with 3% DSS alone or normal control since day 2 (Fig. 2).

We investigated the effects of AGN205203, an EP4 agonist, on colitis in mice induced by 5% DSS-induced. We observed a rapid body weight loss of 10% within 3 days, along with diarrhea and bloody stool (Fig. 3, A, C, and D). Subcutaneous administration of AGN205203 (3 mg/kg/day) from day -2 to day 7 abolished colitis symptoms; gaining body weight up to 4% instead of weight loss (Fig. 3, A) and lower diarrhea and hemoccult scores compared with vehicle-treated mice (Fig. 3, C and D). Food consumption was less in vehicle-treated mice.
than that in AGN205203-treated or normal control mice (Fig. 3B). Overall, the mice treated with AGN205203 seemed much healthier than the vehicle-treated mice.

Gross observations of the colon and spleen were performed on day 7. The average colon length was 76.6 ± 1.4, 63.4 ± 0.8, and 75.2 ± 1.0 mm for the normal, vehicle-, and AGN205203-treated mice, respectively. The ratio of colon weight to length, which reflects colonic edema and water absorption was 5.9 ± 0.2, 7.5 ± 0.3, and 6.1 ± 0.2 for the normal, vehicle-, and drug-treated mice, respectively (P < 0.0001). The weight of spleen as normalized to the body weight was 0.41 ± 0.03, 1.42 ± 0.06, and 0.96 ± 0.05% for the normal, vehicle- and AGN205203-treated mice, respectively (P < 0.0001), and it was inversely related to the hematocrit level as reported previously (Morteau et al., 2000). Overall, AGN205203 treatment reversed the phenotypes of colitis mice (vehicle-treated), such as significant shortenings of the colon, increased edema and decreased water absorption, and splenomegaly.

Tissues from isolated colons were processed for histological observation. Twenty tissue slides from each group were examined. Figures 4A shows the representative sections. As shown in Fig. 4, top, H&E staining revealed considerable tissue damage in the vehicle-treated group (V), but not in the normal or AGN205203-treated groups. For example, the vehicle-treated colon showed a loss of columnar nuclei at the surface of crypts, the disappearance of the tight conjunctive epithelial cells, and the infiltration of mononuclear cells into eroded ulcerous mucous layers. Inset a to the V panel shows severely ulcerated regions with distorted crypts, destruction of cellular structure, and some mononuclear cell infiltrations. On the bottom panel, Alcian Blue primarily stains mucus polysaccharide (blue) and nuclei (pink). The staining revealed the depletion of goblet cells (P < 0.0001; Fig. 4, A and B) and the loss of top crypt nuclei from the vehicle-treated

![Fig. 3. Clinical observation of colitis mice treated with EP4 agonist AGN205203. Mice were treated with either vehicle only (N), 5% DSS plus indomethacin and vehicle (D+I+V), or 5% DSS plus indomethacin and AGN205203 (D+I+AGN205203) (n = 20/group). A, body weight loss as percentage of initial body weight; ***, P < 0.001, D+I+V group versus N and D+I+AGN205203 groups. B, average daily food intake relative to body weight in mice; ***, P < 0.0001 versus N and D+I+AGN205203. C, diarrhea score for all record points; ***, P < 0.001 between D+I+V and D+I+AGN205203. D, hemoccult score; *, P < 0.01 versus N and D+I+AGN205203.

![Fig. 4. Histopathological observation of colon treated with vehicle (N), 5% DSS plus indomethacin and vehicle (V), or 5% DSS plus indomethacin and AGN205203 (AGN205203). A, H&E staining of colon tissues. Images from each group are shown at original magnification, 200× (top). In the vehicle-treated colon, the loss of columnar nuclei of epithelial cells at the surface of crypts was common; some regions completely lost crypts and formed typical ulcers with mononuclear cells. The inset (a) shows that severe damaged crypts had few enterocytes but mononuclear infiltrates. Bottom, representative images of Alcian Blue staining of goblet cells in colon tissues, at original magnification, 200×. In addition to the depletion of goblet cells, the top one-third or one-half of crypts showed pale nuclei or disappearance of nuclei in the vehicle-treated section. B, quantitation of goblet cells from colon slides as shown in Fig. 5A. ***, P < 0.0001 versus N and AGN205203; n = 20.](https://i.imgur.com/3Q5Q5Q5.png)
mice. No such damage was observed in the AGN205203-treated or control mice.

At the cellular level, we also monitored apoptotic cells using TUNEL staining. Nearly 5% of DAPI-stained cells were TUNEL-positive in colonic mucosal layers from DSS-indo-treated mice. With the drug treatment, however, TUNEL-positive cells decreased to less than 1.3% (Fig. 5B). It should be noted that TUNEL-positive stained cells were mainly located at the surface of mucous layer, with little staining at the bottom of crypts or other layers of the colons (Fig. 5A). We also monitored proliferation of epithelial cells using PCNA as a marker, because gastrointestinal epithelial cells are turned over rapidly. PCNA is an intranuclear polypeptide whose synthesis reaches its maximum during the S phase of cell cycle. Generally, PCNA-positive cells were localized largely at crypt epithelium of the mucous layer and rarely found in submucosal, muscular, or serosa layer (Fig. 6A). With the DSS-indo treatment, PCNA-positive cells are sparsely found only at the bottom of crypts, probably hinting an ongoing, spontaneous repair of epithelial damage, albeit not so robust. In contrast, the AGN205203 treatment abundantly produced PCNA-positive cells, which were localized not only at the bottom of crypts but also at the top of crypts. Overall, PCNA-positive cells in nonvillus as well as villus crypts were significantly greater in number with AGN205203 treatment than those without the drug treatment (Fig. 6B). It seems that AGN205203 may accelerate the regenerative capability of epithelial cells and replenish cells at the crypts much faster. Taken together, the above-mentioned results indicate that EP4 activation may contribute to the maintenance of intact mucosal barrier.

**Discussion**

Low dose of DSS treatment alone induced no obvious colitis symptoms in wild-type mice (Morteau et al., 2000; Shichijo et al., 2005). In contrast, in COX-1 or -2 knockout mice, DSS alone produced severe colitis (Morteau et al., 2000). These observations are also consistent with our current observations that only a combined treatment with DSS and indomethacin at a low dose (4 mg/kg/day) produced significant retardation in body weight gain and bloody diarrhea. In addition, clinically, NSAIDs have been reported to trigger or worsen IBD patients (Thiefin and Beaugerie, 2005; Meyer et al., 2006). These results indicate some protective effects of prostanoids synthesized by COX against colitis development (Morteau et al., 2000). Particularly, PGE$_2$ has been reported to increase in colonic tissues upon DSS treatment (Morteau et al., 2000), and it seems to be the primary prostanoid contributing to such protective effects.

PGE$_2$ interacts with four EP receptor subtypes (EP1–4). Previous studies have shown that, in human colon, both surface and lateral epithelium of crypts constitutively express the EP4 subtype, whereas apex epithelia of crypts constitutively express the EP2 and EP3 subtypes (Takafuji et al., 2000). The EP1 subtype is not detected on colon epithelial cells (Morteau et al., 2000). Interestingly, only EP4$^{-/-}$ among the eight prostanoid receptor knockout mice is the most susceptible to DSS-induced colitis (Kabashima et al., 2002). Moreover, topical application of an EP4 agonist, ONO-AE1–329, reduces cytotoxic cytokine production and amelio-
rates colitis symptoms in DSS colitis model (Nitta et al., 2002). Likewise, in this current study, we have shown that systemic application of a chemically and metabolically stable EP4 agonist, AGN205203, eliminated colitis symptoms, such as body weight loss, reduced food intake, shortening of colon, and severe diarrhea in the mouse IBD model. These results are consistent with the notion that PGE$_2$ via EP4 may ameliorate colitis symptoms.

Previous studies with an ONO EP4 agonist in mouse IBD models were largely focused on its ability to inhibit innate immune responses, namely, the inhibition of cytotoxic cytokine production (Kabashima et al., 2002; Nitta et al., 2002). More importantly, however, EP4 activation has been shown to induce antiapoptotic activities in cultured gastric and intestinal epithelial cells (Hoshino et al., 2003; Joseph et al., 2005), which seem to be critical for preserving and mending mucosal barrier functions. Therefore, we further investigated morphological and biochemical changes in colonic tissues and cells. Histopathological examination showed that tissues from vehicle-treated mice showed a marked loss of surface epithelia, visible erosion and ulcer formation, and infiltration of inflammatory cells. Alternatively, tissues from the AGN205203-treated mice showed a well preserved structure of mucous layer with no loss of surface epithelia, and no visible erosion and ulcer formation. Also of interest is our current observation that EP4 agonist treatment preserves not only epithelial but also goblet cells, which are largely depleted in vehicle-treated mice. It has been reported that goblet cells from both human and experimental animals robustly express EP4, and its expression was further enhanced in colon mucosa during IBD (Northey et al., 2000; Takafuji et al., 2000; Nitta et al., 2002). Also known is that mucus secretion from goblet cells increases upon activation of EP4 but that it is inhibited by NSAIDs (Ohnishi et al., 2001; Shimamoto et al., 2005). Thus, EP4 agonists should bring about robust secretion of mucus through the activation of EP4 abundantly expressed in colonic goblet cells during colitis development and provide a semipermeable, mechanical gel barrier between the lumen and the epithelium. Such actions of EP4 agonist certainly enhance mucosal barrier functions, which are critical in minimizing the penetration of bacteria and toxins to the cells lining the colon (Einerhand et al., 2002; Shaoul et al., 2004). Thus, depletion of goblet cells means the loss of such critical preventive functions in the colon, and it may serve as a histological hallmark for ulcerative colitis in humans and experimental animals.

At cellular level, our current study demonstrated for the first time in vivo that EP4 agonist prevented colitis-induced epithelial apoptosis and promote cell proliferations. Previously, in cultured intestinal and gastric epithelial cells, PGE$_2$ has been reported to inhibit apoptosis and also NSAID-induced apoptosis (Joseph et al., 2005; Redlak et al., 2005). Mechanistically, several cellular kinases and transactivators have been proposed to mediate cell survival and cell growth by EP4 agonists, such as protein kinase A, phosphatidylinositol 3-kinase, extracellular signal-regulated kinases, and growth response factor-1 (Fujino et al., 2003; Fujino and Regan, 2006). Further study would clarify the roles of these kinases in EP4 actions in colitis.

Some concerns exist, however, about a potential contribution of EP4 agonists to abnormal cell growth in the colon. In this study, AGN205203 treatment seems to preserve the normal morphology of crypts with no detectable change in the height of crypts compared with those of the normal group. It seems that the drug reverses only DSS-indo-induced epithelial cell death and colonic crypt reduction. In contrast, it has been known from knockout mice studies that ablation of EP1, EP2, or EP4 reduced the formation of aberrant crypt foci in the colon, suggesting their potential contributions to colon carcinogenesis (Shoji et al., 2004). However, this could be also interpreted that EP4 activation may be necessary but not sufficient for colon carcinogenesis and that EP4-selective agonists may not have carcinogenic potentials without the activation of the other two subtypes of PGE$_2$ receptors.

Another obvious potential side effect of PGE$_2$ analogs is diarrhea, because of the elevation of cAMP in epithelial cells, and the subsequent activation of cAMP-regulated chloride channels. Recently, however, the selective activation of EP4 has been reported to induce primarily G$_i$/phosphatidylinositol 3-kinase/extracellular signal-regulated kinase signaling and much less G$_i$/cAMP signaling (Fujino and Regan, 2006). Therefore, it is reasonable to speculate that EP4-selective agonists could induce diarrhea, but at much milder levels than other nonselective PGE$_2$ analogs on the market. Future study is needed to clarify potential side effect profiles of EP4 agonists, including diarrhea and abnormal cell growth in naive animals.

In summary, IBD is triggered by disruption of epithelial integrity and inadequate repair. AGN205203, an EP4 agonist of the 8-aza piperidinone PGE series, when applied systemically, prevented goblet cell depletion, reduced epithelial cell apoptosis, and enhanced epithelial proliferation, resulting in near normal functions and morphology of the colon in the presence of colitis inducing insults, DSS, and indomethacin. We propose that AGN205203 would be therapeutically useful for prevention and treatment of IBD via monotherapy or and combination therapy with drugs in market, particularly during the maintenance phase of IBD, the success of which depends on the restoration of mucosal barrier functions.

Acknowledgments

We are grateful to Yan Hai (Department of Pathology, Allergan, Inc.) for the excellent work in tissue sectioning and staining. We thank Sonal Vaziran from University of California (Irvine, CA) for apoptosis study. We appreciate the discussion with Adelekan Oyejide (Allergan, Inc.).

References


Address correspondence to: Dr. Guang-Liang Jiang, Department of Biological Sciences, Herbert Research Center, Allergan, Inc., 2525 Dupont Dr., R&D3-2B, Irvine, CA 92612. E-mail: jiang_guang-liang@allergan.com