



# Efficacy of Rectal Tacrolimus for Induction Therapy in Patients With Resistant Ulcerative Proctitis

Ian C. Lawrence,<sup>\*,†</sup> Angela Baird,<sup>\*</sup> Daniel Lightower,<sup>‡</sup> Graham Radford-Smith,<sup>§,||</sup> Jane M. Andrews,<sup>¶,#</sup> and Susan Connor<sup>\*\*,††,§§</sup>

<sup>\*</sup>Harry Perkins Institute of Medical Research, School of Medicine and Pharmacology, University of Western Australia, Murdoch, WA, Australia; <sup>‡</sup>Centre for Inflammatory Bowel Diseases, Saint John of God Hospital, Subiaco, WA, Australia; <sup>§</sup>IBD Research Group, QIMR Berghofer Medical Research Institute, University of Queensland School of Medicine, Brisbane, Queensland, Australia; <sup>¶</sup>Department of Gastroenterology, Royal Brisbane and Women's Hospital, Brisbane, Queensland, Australia; <sup>#</sup>Department of Gastroenterology and Hepatology, Royal Adelaide Hospital, Adelaide, SA, Australia; <sup>\*\*</sup>University of Adelaide, School of Medicine, SA, Australia; <sup>††</sup>Department of Gastroenterology and Hepatology, Liverpool Hospital, Sydney, NSW, Australia; <sup>††</sup>South Western Sydney Clinical School, University of NSW Medicine, Sydney, NSW, Australia; and <sup>§§</sup>Ingham Institute for Applied Medical Research, South Western Sydney Clinical School, UNSW Australia, Sydney, NSW, Australia

**BACKGROUND & AIMS:** Resistant ulcerative proctitis can be extremely difficult to manage. Topically administered tacrolimus, however, may be effective in difficult-to-treat proctitis. This was a randomized, double-blind, placebo-controlled induction trial of rectal tacrolimus in patients with active ulcerative colitis.

**METHODS:** Eleven patients received rectal tacrolimus (0.5 mg/mL), and 10 placebo, for 8 weeks. The primary endpoint was clinical response by using the Mayo Clinic score.

**RESULTS:** A planned interim analysis after 20 patients had completed the study demonstrated highly significant differences between the groups and the study was closed because of ethical considerations with patients already recruited allowed to complete the study. The primary endpoint was met in 8 of 11 patients receiving rectal tacrolimus and 1 of 10 patients receiving placebo (73% vs 10%;  $P = .004$ ). Of the secondary endpoints, 5 patients with rectal tacrolimus achieved clinical remission compared with none receiving placebo (45% vs 0%;  $P = .015$ ). Mucosal healing at Week 8 was achieved in 8 patients receiving rectal tacrolimus compared with 1 (73% vs 10%) receiving placebo ( $P = .004$ ). The Inflammatory Bowel Disease Questionnaire increased  $\geq 16$  points over baseline in 5 of the tacrolimus and 2 (45% vs 20%) of the placebo patients ( $P = .36$ ). Finally, the average partial Mayo score was numerically lower in the tacrolimus-treated group compared with placebo at Week 2 ( $4.3 \pm 0.74$  vs  $5.8 \pm 0.64$ ;  $P = .15$ ) and Week 4 ( $3.7 \pm 0.96$  vs  $5.8 \pm 0.6$ ;  $P = .08$ ) but was significantly lower at Week 8 ( $3.3 \pm 1.2$  vs  $6.7 \pm 0.62$ ;  $P = .01$ ). There were no safety issues identified with rectal tacrolimus use.

**CONCLUSIONS:** Rectal tacrolimus was more effective than placebo for induction of a clinical response, clinical remission, and mucosal healing in resistant ulcerative proctitis ([Clinicaltrials.gov registration: NCT01418131](https://clinicaltrials.gov/ct2/show/NCT01418131)).

**Keywords:** Ulcerative Colitis; Proctitis; Tacrolimus; Blinded Randomized Clinical Trial.

Ulc<sup>er</sup>ative colitis (UC) is thought to result from an interaction between environmental and genetic factors with disease activity characterized by a life-long course of remissions and exacerbations; however, approximately 20%–55% of patients with UC suffer, at some stage, a severe attack requiring hospitalization.<sup>1</sup> The Montreal classification<sup>2</sup> divides the distribution of UC into ulcerative proctitis (E1; limited to the rectum), left-sided colitis (E2; up to the splenic flexure), and extensive colitis (E3; beyond splenic flexure).

Inflammation confined to the rectum (E1) occurs in approximately 25% of patients with UC and although this

**Abbreviations used in this paper:** CD, Crohn's disease; IBDQ, Inflammatory Bowel Disease Questionnaire; QOL, quality of life; TNF, tumor necrosis factor; UC, ulcerative colitis.

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results in distressing symptoms, including stool frequency, tenesmus (a feeling of incomplete evacuation), fecal urgency, and bleeding, it can often be managed within the community with topical agents.<sup>3</sup> Medication-resistant ulcerative proctitis, however, is extremely challenging to manage. When rectal medications fail, oral agents including mesalamine, azathioprine/6-mercaptopurine, and steroids may be used, but they do not always help. The anti-tumor necrosis factor (TNF)- $\alpha$  medications may also be an effective treatment and can have a response rate of 68% and a remission rate of 40% by 2 weeks.<sup>4</sup> The recent inclusion of the new anti-integrin therapy, vedolizumab, on many formularies has offered another potentially effective option.<sup>5,6</sup> These biologic therapies, however, are still not universally effective, are systemic, and carry a considerable cost burden.

Tacrolimus and cyclosporin are classical calcineurin inhibitors and are widely used as immunosuppressive medications and have demonstrated promising results in the management of UC.<sup>7,8</sup> Calcineurin, or protein phosphatase 2B, is a ubiquitously expressed cytosolic Ser/Thr protein phosphatase that is highly conserved in eukaryotes.<sup>9</sup> It has the ability to dephosphorylate a broad range of proteins and can regulate interleukin-2, interleukin-4, and interferon- $\gamma$  expression,<sup>10</sup> and modulate the activity of transcription factors, such as nuclear factor- $\kappa$ B.<sup>11</sup> Enhanced nuclear factor- $\kappa$ B activity is well described in Crohn's disease (CD) and UC and induces the proinflammatory cytokine interleukin-1 $\beta$ , interleukin-6, and TNF- $\alpha$  expression. It is primarily through the reduction in the levels of these cytokines that clinical remission is thought to be achieved.

The efficacy of oral tacrolimus has been examined in the management of medication-resistant CD and UC. Unfortunately, most of these studies have been open-labelled with only 1 randomized-controlled trial reported in UC.<sup>12</sup> This demonstrated a short-term clinical improvement but without a significant increase in remission, potentially because of low patient numbers. Despite this, there are numerous open-labelled studies in both UC and CD that suggest efficacy in the short-term, and with promising long-term data.<sup>13-18</sup> The evidence suggests, however, that the blood trough levels should be at least 10  $\mu$ g/L to achieve the best efficacy (therapeutic range, 5-20  $\mu$ g/L), but the higher the trough level the more likely a patient will suffer an adverse effect.<sup>19</sup> These, unfortunately, can be numerous and include hypertension, nausea and diarrhea, hematologic abnormalities, and renal impairment.<sup>13</sup> Increase in the rate of skin cancers is also a concern,<sup>20</sup> a side effect that has been supported by animal studies.<sup>21</sup>

Use of topical tacrolimus has been effective in the treatment of perioral and perineal inflammation in pediatric patients with CD with resolution of symptoms in 75%.<sup>22</sup> Work examining topical perianal tacrolimus therapy in adult patients with CD also demonstrated clinical efficacy,<sup>23</sup> and although tacrolimus

is absorbed well transdermally,<sup>24</sup> only low trough levels of tacrolimus were detected in the blood.<sup>23</sup> In these preliminary studies, the use of topical tacrolimus was associated with very few side effects. Long-term topical use, as with oral formulations, may be associated with an increased risk of skin cancer formation. Epidemiologic evidence, however, suggests that the risk is low and localized only to the tacrolimus-treated sun-exposed skin.<sup>25-27</sup>

A pilot study by our group demonstrated that 75% (6 of 8) of patients with resistant distal colitis responded and achieved a clinical remission of their disease following 4-8 weeks of tacrolimus rectal ointment.<sup>28</sup> A dose of 0.3-0.5 mg/mL, 3 mL twice a day, was identified in most patients to induce remission. In these patients, tacrolimus trough levels were taken regularly and were either undetectable (<1.5  $\mu$ g/L), subtherapeutic (<5  $\mu$ g/L) or, at their highest, in the low therapeutic range (therapeutic range, 5-20  $\mu$ g/L). The ointment was well tolerated without any systemic adverse effects. Use of the same rectal tacrolimus preparation in more than another 20 patients by the chief investigator has also been associated with a clinical response in up to 75% of patients following 4 weeks of therapy.

The efficacy of topical tacrolimus in UC is further supported by a separate pilot study that examined topical tacrolimus in patients with resistant distal colitis. In this study clinical and histologic improvement was achieved in 10 of 12 patients with proctitis by 4 weeks. Again no major side effects were reported and the preparation was well tolerated.<sup>29</sup> The mechanism of action of tacrolimus seems to be local, rather than systemic, immune suppression because the administration of rectal tacrolimus inhibits the activation of immune cells within the intestinal mucosa and not systemically.<sup>30</sup> Thus because the studies and observations demonstrate encouraging results in a difficult-to-treat patient population, a randomized double-blinded placebo-controlled trial was warranted.

## Materials and Methods

### Study Design

This was randomized, double-blind, placebo-controlled UC induction study conducted at 4 specialist inflammatory bowel disease centers in Australia (Fremantle Hospital, WA; Royal Brisbane and Women's Hospital, Qld; Royal Adelaide Hospital, SA; and Liverpool Hospital, NSW). The protocol was approved by the Eli and Edythe Broad Foundation, which funded the work, and by each human ethics committee at each center. Because the medication was not approved by the Therapeutic Goods Administration in Australia, approval to use the medication in a clinical trial was obtained from the Therapeutic Goods Administration for each participating site. All patients gave written informed consent.

The study was conducted and reported in accordance with the protocol available at [ClinicalTrials.com](https://clinicaltrials.gov/ct2/show/NCT01418131) (registration: NCT01418131).

### *Tacrolimus Rectal Ointment Preparation*

The preparation of the tacrolimus rectal ointment was undertaken in the hospital pharmacy of each of the participating sites. Gloves and mask were worn when making the preparation. The final concentration of tacrolimus in the rectal ointment was 0.5 mg/mL.<sup>28</sup> To create the tacrolimus preparation, 5 mL of propylene glycol was slowly mixed into the desired amount of tacrolimus powder on a clean glass slab. To this 70 mL of paraffin liquid BP was gradually added by serial dilution and triturate until evenly mixed. This process was repeated with 125 mL of paraffin white soft BP. The resulting cream was packed into tubes and labelled. The cream is stable for more than 30 days when stored at room temperature and more than 90 days if refrigerated. The preparation was formulated using the paraffin liquid BP/paraffin white soft BP base for ease of rectal use. To create the placebo, the same process was followed but without the addition of the tacrolimus powder. A total of 3 mL of the tacrolimus/placebo ointment was applied rectally, via an applicator, twice a day by the patient over the 8-week period. Patients received their rectal preparation at the randomization and the Week 4 visit.

### *Study Population*

Eligible patients were 18 years or older, and had a diagnosis of UC made in accordance with established international criteria based on clinical, endoscopic, histologic, and radiologic features<sup>2</sup> of >3 months duration. All patients had active inflammation defined as a Mayo score of 6–12 (range, 0–12, with higher scores indicating more active disease)<sup>31</sup> that was limited to a maximum of 25 cm from the anal verge. All patients had failed conventional therapies of either oral and/or rectal mesalamine and/or oral and rectal steroids, or were intolerant of these medications. Patients on rectal preparations were allowed to continue their rectal medication up to the day before commencing the rectal tacrolimus trial. Patients taking oral mesalamine had used it continuously for 4 weeks and were on a stable dose for 2 weeks before the screening visit, whereas patients on oral corticosteroids had used them continuously for 4 weeks and were on a stable dose of 30 mg or less for 2 weeks before the screening visit. Patients on azathioprine/6-mercaptopurine or methotrexate had used them for a minimum of 12 weeks with a stable dose for 4 weeks before the screening visit.

Patients were ineligible for this study if they had a known hypersensitivity/allergic reaction to rectal/oral tacrolimus, were pregnant or breast-feeding, or suffered from uncontrolled hypertension or chronic renal failure.

Patients were also ineligible for the study if they had received an anti- TNF- $\alpha$  medication, or any trial or biologic agent, within 8 weeks of the screening visit. Other exclusion criteria included patients with drug abuse, alcohol dependence, dementia or an inability to understand the trial requirements, patients with colitis extending more than 25 cm from the anal verge, and patients who have previously been treated with a rectal tacrolimus preparation. Subjects were discontinued from the study if they revoked their consent, or at the discretion of the principal investigator, based on disease, side effects, or poor compliance.

### *Screening and Baseline Studies*

Assessments performed before randomization included physical, blood tests, and stool analysis for enteric pathogens. Demographic data were collected. Eligible patients were scheduled for a visit within 7 days of randomization, when a sigmoidoscopy was performed and baseline Mayo Clinic scores and scores on the Inflammatory Bowel Disease Questionnaire (IBDQ; range, 0–224, with normal quality of life [QOL] defined as a score of  $\geq 170$  points with higher scores indicating a better QOL and a QOL response defined as a change of  $\geq 16$  points)<sup>32</sup> were determined.

### *Randomization Procedures*

Patients were randomly assigned, in a 1:1 ratio, to receive either 0.5 mg/mL, 3 mL twice a day, of rectal tacrolimus or placebo for 8 weeks. Patients who did not respond by Week 4 were allowed to withdraw and received open-label rectal tacrolimus. Randomization was performed centrally with a computer-generated randomization schedule. Permitted concomitant UC medications included aminosalicylates, glucocorticoids, and immunosuppressive agents that were continued at stable doses throughout the 8-week period.

### *Follow-up*

Patients were seen at Weeks 2, 4, and 8 during the study. The patient filled in a Patient Diary Card with the date and time of administration of the rectal preparation. This diary card was checked for compliance by the investigators at each patient visit. At Week 2 and 4, a partial Mayo score (consisting of the Mayo score minus the sigmoidoscopy subscore; range, 0–9, with higher scores indicating more active disease)<sup>33</sup> was calculated. All adverse events were noted. IBDQ scores and blood samples for serum chemical, hematologic, and tacrolimus trough level testing were obtained at each visit. Sigmoidoscopy was performed at Week 8, or at study withdrawal if this was after at least 4 weeks on the trial medication, and the full Mayo score calculated.

## Outcomes

The primary outcome was the clinical response (Mayo Score) at 8 weeks defined as a reduction in the Mayo Clinic score of  $\geq 3$  points and a decrease of  $>30\%$  from the baseline score, with a decrease of  $\geq 1$  point on the rectal bleeding subscale or an absolute rectal bleeding score of 0 or 1.<sup>31</sup> Secondary outcomes at Week 8 were clinical remission, defined as a Mayo Clinic score of  $\leq 2$  and no subscore  $>1$ , and mucosal healing, defined as an endoscopic subscore of 0 or 1.<sup>31</sup> Other secondary endpoints at 8 weeks were the effect of rectal tacrolimus on mucosal healing, changes in the partial Mayo Score,<sup>33</sup> and changes in the health-related QOL through the use of the IBDQ.<sup>32</sup> Safety and tolerability of rectal tacrolimus were assessed with adverse events classified with the use of the Medical Dictionary for Regulatory Activities 25 version 15.

## Study Oversight

The study was designed and implemented by the chief investigator supported by a grant from the Broad Foundation. The study was monitored by the chief investigator's site. Local investigators and their participating institutions agreed to maintain confidentiality of the data. The chief investigator wrote the manuscript with intellectual input obtained from each of the authors. All authors had access to the study data and reviewed and approved the final manuscript. All the authors made the decision to submit the manuscript for publication.

## Statistical Analysis

A sample size calculation determined that 20 patients per group was considered to be sufficient to see a difference between treatment and placebo group for the primary outcome measure with a significance level of 5% and power of 80%. Effects of time, treatment, and the time by treatment interaction were tested and reported. Estimated differences and 95% confidence intervals for differences are given for each time point. For all analyses the impact of potentially confounding demographic variables, such as sex and age, were considered. Student *t* test, chi-square, and analysis of variance analysis were undertaken with *P* values  $< .05$  considered significant.

An interim analysis was written into the protocol and was to be undertaken by a blinded independent statistician and gastroenterological clinicians as detailed in the study design after 20 patients had completed the 8-week study to determine safety and to determine that the study should continue. The study could be terminated at this point for either safety or ethical considerations.

## Results

### Randomization and Baseline Characteristics

A total of 28 patients were assessed for inclusion into the study. Of these 21 were enrolled and included for analysis. Seven patients did not meet the inclusion criteria of disease limited to the distal 25 cm of the colon. In the trial, 11 patients were randomly assigned to receive tacrolimus, 0.5 mg/mL twice a day, and 10 were allocated to the placebo arm of the study. The baseline characteristics were similar between the placebo group and rectal tacrolimus group (Table 1). The Mayo endoscopy subscore at inclusion in the placebo group was 3 in 4 patients and 2 in 6 patients. The Mayo endoscopy subscore in the tacrolimus group was 3 in 6 patients and 2 in 5 patients. The mean Mayo score at

**Table 1.** Demographic and Baseline Characteristics With Concomitant Medications

|  | Placebo<br>n = 10 | Tacrolimus<br>n = 11 | P<br>value |
|--|-------------------|----------------------|------------|
| Age, y                                   | 39.0 $\pm$ 4.8    | 48.4 $\pm$ 4.9       | NS         |
| Male sex, n (%)                          | 4 (40)            | 8 (73)               | NS         |
| Age at diagnosis, n (%)                  |                   |                      |            |
| A1, $\leq 16$                            | 0 (0)             | 0 (0)                |            |
| A2, 17–40                                | 8 (80)            | 5 (44)               | NS         |
| A3, $>40$                                | 2 (20)            | 6 (56)               |            |
| Disease duration, y $\pm$ SEM            | 7.2 $\pm$ 1.3     | 9.2 $\pm$ 1.9        | NS         |
| Family history IBD, n (%)                | 2 (20)            | 2 (18)               | NS         |
| Smoking, n (%)                           |                   |                      |            |
| Never                                    | 2 (20)            | 2 (18)               |            |
| Ex-smoker                                | 8 (80)            | 9 (82)               | NS         |
| Current                                  | 0 (0)             | 0 (0)                |            |
| EIM, n (%)                               |                   |                      |            |
| None                                     | 8 (80)            | 8 (73)               | NS         |
| Joints (arthritis/arthritis)             | 2 (20)            | 3 (27)               | NS         |
| Eyes (iritis/episcleritis)               | 0 (0)             | 1 (9)                | NS         |
| Skin (EN, PG)                            | 0 (0)             | 0 (0)                |            |
| PSC                                      | 0 (0)             | 0 (0)                |            |
| Mayo score ( $\pm$ SEM)                  | 9.6 $\pm$ 0.5     | 8.6 $\pm$ 0.4        | NS         |
| Mayo Endoscopy Subscore, n (%)           |                   |                      |            |
| 3  | 4 (40)            | 6 (55)               | NS         |
| 2  | 6 (60)            | 5 (45)               | NS         |
| Partial Mayo score ( $\pm$ SEM)          | 7.1 $\pm$ 0.4     | 6.3 $\pm$ 0.4        | NS         |
| IBDQ score ( $\pm$ SEM)                  | 160 $\pm$ 9       | 141 $\pm$ 6          | NS         |
| Concomitant medications<br>for UC, n (%) |                   |                      |            |
| Mesalamine                               |                   |                      |            |
| Oral/topical                             | 8 (80)            | 8 (72)               | NS         |
| Glucocorticoids                          |                   |                      |            |
| Oral                                     | 3 (30)            | 2 (18)               | NS         |
| Topical                                  | 2 (20)            | 4 (36)               | NS         |
| Immunosuppressants                       |                   |                      |            |
| Azathioprine/6-mercaptopurine            | 5 (50)            | 5 (44)               | NS         |
| Methotrexate                             | 0 (0)             | 1 (9)                | NS         |
| Anti-TNF- $\alpha$                       | 0 (0)             | 0 (0)                | NS         |

EIM, extraintestinal manifestations; EN, erythema nodosum; IBD, inflammatory bowel disease; NS, not significant; PG, pyoderma gangrenosum; PSC, primary sclerosing cholangitis; SEM, standard error of the mean.

Week 0 in the tacrolimus group was  $8.6 \pm 0.4$  compared with  $9.6 \pm 0.5$  ( $P = .11$ ). There were also no significant differences observed between the groups neither in the other demographic or baseline characteristics nor in medication history between the groups.

In the placebo group, 60% of patients had failed oral steroids, 30% topical steroids, 90% oral mesalamine, and 50% an immunomodulator medication. In the tacrolimus group 64% of patients had failed oral steroids, 36% topical steroids, 73% oral mesalamine, and 64% an immunomodulator medication. All had failed topical mesalamine treatment. No patient in either group had been treated with an anti-integrin and only 1 patient in the tacrolimus group had failed an anti-TNF medication.

### Outcomes

An interim analysis was undertaken, as per the protocol, after 20 patients had completed the 8-week study. Because of the highly significant differences identified between the groups, across multiple endpoints, it was decided that ethically the study should be closed with any patients already commenced on the study allowed to complete the study. No safety issues were identified.

A total of 21 patients completed the study. Two of the placebo patients and 1 rectal tacrolimus patient withdrew after the Week 4 visit (2 at Week 6 and 1 at Week 4), as allowed by the protocol, 2 because of perceived lack of efficacy and 1 because of pregnancy. These patients underwent a sigmoidoscopy examination and were included for analysis of the primary and secondary outcomes as per the protocol.

The primary endpoint was met in 8 of the 11 patients receiving rectal tacrolimus and 1 of the 10 patients receiving placebo by having a clinical response (73% vs 10%;  $P = .004$ ) at Week 8 (Figure 1, Table 2). The mean Mayo score at Week 8 in the tacrolimus group was  $4.5 \pm 1.3$  compared with  $8.8 \pm 0.8$  ( $P = .01$ ; Figure 2).

Of the secondary endpoints, 5 of the patients receiving rectal tacrolimus achieved clinical remission,

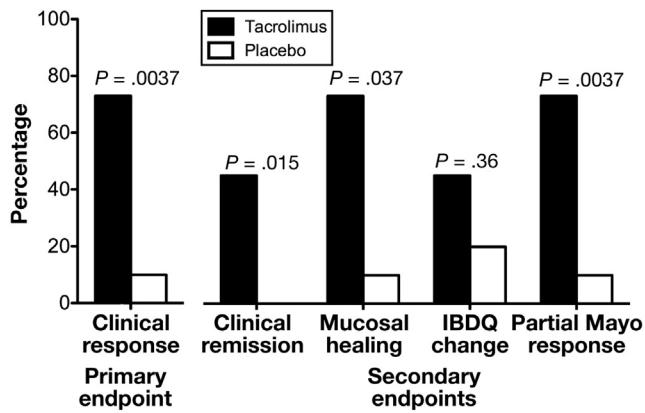


Figure 1. Primary and secondary endpoints at Week 8.

Table 2. Outcome Measures at Week 8

|                       | Placebo (%)<br>(n = 10) | Tacrolimus (%)<br>(n = 11) | P<br>value |
|-----------------------|-------------------------|----------------------------|------------|
| Clinical response     | 1 (10)                  | 8 (73)                     | .0037      |
| Clinical remission    | 0 (0)                   | 5 (45)                     | .015       |
| Mucosal healing       | 1 (10)                  | 8 (73)                     | .0037      |
| Change in IBDQ        | 2 (20)                  | 5 (45)                     | .36        |
| Partial Mayo response | 1 (10)                  | 8 (73)                     | .0037      |

whereas none of the patients receiving placebo achieved remission (45% vs 0%;  $P = .015$ ). Mucosal healing at Week 8 was achieved in 8 of patients receiving rectal tacrolimus compared with 1 patient receiving placebo (73% vs 10%;  $P = .004$ ). The IBDQ was  $\geq 170$  points (normal range) at baseline in 1 patient randomized to receive rectal tacrolimus and 3 (9% vs 30%) of patients randomized to receive placebo. At Week 8, the IBDQ had increased  $\geq 16$  points over baseline in 5 of the tacrolimus and 2 of the placebo patients (45% vs 20%;  $P = .36$ ). Finally, the partial Mayo score was reduced by  $\geq 3$  points in 8 of the patients receiving rectal tacrolimus compared with 1 patient taking placebo (73% vs 10%;  $P = .0037$ ). The average partial Mayo score at each visit is shown in Figure 3. At Week 0 the scores were similar ( $6.3 \pm 0.4$  vs  $6.9 \pm 0.3$ ;  $P = .21$ ) in both groups but was noted to be numerically lower in the tacrolimus-treated group compared with placebo at Week 2 ( $4.3 \pm 0.74$  vs  $5.8 \pm 0.64$ ;  $P = .15$ ) and Week 4 ( $3.7 \pm 0.96$  vs  $5.8 \pm 0.6$ ;  $P = .08$ ) but was significantly lower at Week 8 ( $3.3 \pm 1.2$  vs  $6.7 \pm 0.62$ ;  $P = .01$ ).

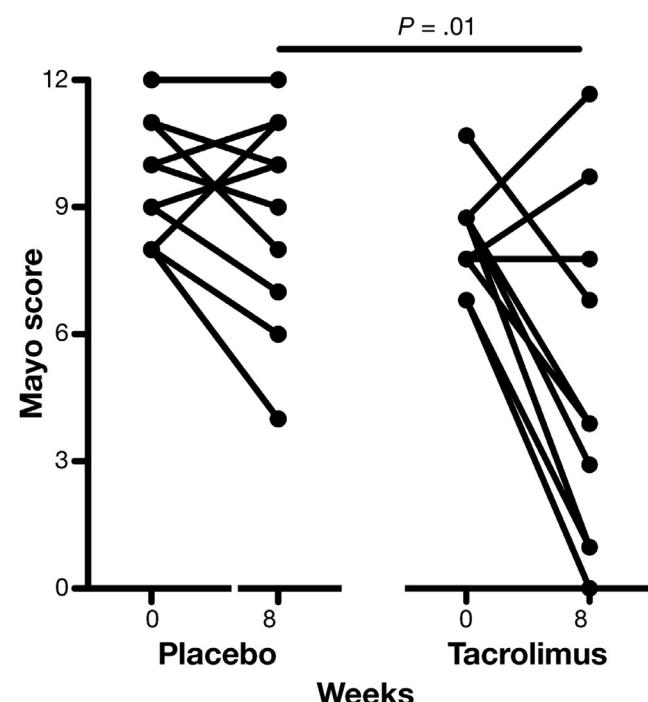
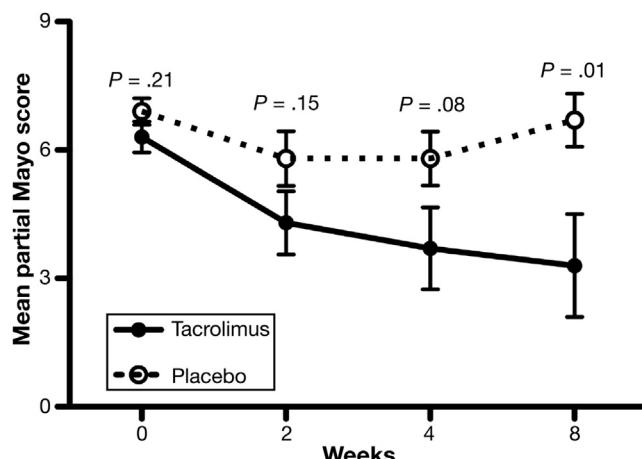


Figure 2. Mayo score at Weeks 0 and 8.



**Figure 3.** Mean partial Mayo score.

#### *Tacrolimus Trough Levels and Safety*

No important differences were observed among the study groups. There was no correlation between the tacrolimus trough levels and clinical outcomes and there was also no correlation between the trough levels and side effects. There were no serious infections and no hospitalizations. No cases of anaphylaxis or serum sickness were observed. One patient in the placebo arm withdrew from the study at Week 6 because of pregnancy without improvement in the colitis symptoms. In consultation with the patient's obstetrician and fetal specialist she was treated with open-label rectal tacrolimus with resolution of her proctitis symptoms. The baby was born at term without any complications. In the placebo arm, there was 1 episode of a throat infection requiring antibiotics and 1 patient complained of burning feet and aching wrists.

Patients were asked to delay administering their rectal preparation until they had had their trough level blood tests taken. The average tacrolimus trough level was  $6.5 \pm 2.2 \mu\text{g/L}$  at Week 2,  $4.2 \pm 1.6 \mu\text{g/L}$  at Week 4, and  $5.2 \pm 2.2 \mu\text{g/L}$  at Week 8 (therapeutic range, 5–20  $\mu\text{g/L}$ ). Tacrolimus trough levels, however, varied from undetectable to  $23.2 \mu\text{g/L}$ . Four of the 11 tacrolimus-treated patients had trough levels  $>10 \mu\text{g/L}$  and it was noted that the blood draw in these patients was later in the day ranging from 11:45 AM to 3:20 PM. If patients had already taken the rectal preparation in the morning before the trough level these would not be true trough levels. With the exclusion of these anomalous levels the average tacrolimus trough level at each time point was subtherapeutic ( $<5 \mu\text{g/L}$ ).

In the tacrolimus treatment arm, 1 patient suffered an upper respiratory tract infection that resolved spontaneously without treatment (tacrolimus trough level undetectable Weeks 2, 4, and 8). One patient noted a mild tremor that did not affect normal activity (tacrolimus trough level undetectable Weeks 2, 4, and 8). Another patient suffered self-limiting dizziness within the first 2 weeks of the study. This patient also had a mild elevation

in the urea  $10.6 \text{ mmol/L}$  (normal range,  $2.9$ – $8.2 \text{ mmol/L}$ ) at inclusion to the study that did not rise with tacrolimus treatment (tacrolimus trough level  $5.1 \mu\text{g/L}$  at Week 2 and then undetectable Weeks 4 and 8). At no point was there a rise in the creatinine and the glomerular filtration rate did not drop over the treatment period. A final patient complained of a headache that resolved with paracetamol (tacrolimus trough level average  $11 \mu\text{g/L}$  with the collection times 1:13 PM and 12:45 PM despite confirmation by the patient diary of administration of the morning rectal tacrolimus dose suggesting that this was not a true trough level). There were 2 further patients with tacrolimus trough levels  $>10 \mu\text{g/L}$  at some stage in the study (maximum level of  $23.2 \mu\text{g/L}$  with a collection time of 2:14 PM and  $12.6 \mu\text{g/L}$  with a collection time 12:55 PM) who did not suffer any adverse effects.

Of note was that there were no cases of hyperglycemia, a rise in the serum creatinine, or a drop in the glomerular filtration rate suggesting no nephrotoxicity in any patient with the use of tacrolimus. No episodes of paresthesia, hypertension, insomnia or nausea were reported.

#### **Discussion**

This paper identifies an effective new therapy for the induction of response, and remission, in patients with active distal UC that is resistant to topical and oral therapies. Because of the efficacy of this treatment recruitment the study was terminated early following the interim analysis as the analysis demonstrated a significant effect of treatment with rectal tacrolimus over placebo across multiple assessments. With enrollment of half of the number of patients predicted to be required to show a significant difference, this trial demonstrated that rectal tacrolimus was effective in inducing a clinical response and remission for colitis limited to 25 cm from the anal verge and resistant to the conventional therapies of either oral and/or rectal mesalamine and/or oral and rectal steroids. The primary endpoint of a clinical response at 8 weeks was achieved. The secondary endpoints of clinical remission, mucosal healing, and a partial Mayo score response were also achieved, and numerically more patients had an improvement in the IBDQ in the treatment, over the placebo, arm but this did not reach clinical significance. Longitudinal assessment of the partial Mayo score also demonstrated a treatment benefit.

Toxicity and adverse effects of oral tacrolimus is related to the serum trough levels. Despite the fact that tacrolimus is readily absorbed through the dermis and thus also the rectal mucosa, the trough levels in our patients were generally low and so it is not surprising that the number of adverse events was also low. The trough levels in some patients, however, were high yet this did not correlate to any adverse outcomes and may have been secondary to the administration of the rectal

preparation before the trough blood collection resulting in a peak, rather than trough, tacrolimus blood level.

One patient on the rectal tacrolimus, however, did note the presence of a fine tremor that is a well-recognized side effect of tacrolimus therapy. There were, however, no adverse effects of renal dysfunction, hyperglycemia, paresthesia, hypertension, insomnia, or nausea reported. There were no cases of serious infection and no serious adverse events in either arm. There was 1 pregnancy in a patient in the placebo arm who withdrew from the study at Week 6. Longer-term epidemiologic studies and clinical experience, however, are still required to more fully determine the risk of adverse events from the use of rectal tacrolimus.

The study does have some important limitations. This is an induction study only and it is yet to be proven that the use of rectal tacrolimus is effective, or safe, for long-term use. The dose selected for the study was 0.5 mg/mL administered as 3 mL twice a day. Our initial paper, and further experience, has demonstrated that higher concentrations of 0.8 mg/mL or 1.0 mg/mL tacrolimus in the rectal cream may be effective in patients who do not respond to 0.5 mg/mL. Superiority of these concentrations over the doses in this study, however, have yet to be formally studied, but could offer greater efficacy. Finally, the numbers in this study are small but the level of clinical efficacy and mucosal healing in the treatment arm were highly significant, despite the small numbers, prompting the early closure of the study. Because of the efficacy of the medication further work is planned to be undertaken in resistant pouchitis and perianal CD. Long-term use of the rectal medication is also going to be assessed for safety, efficacy, ability of de-escalation, and withdrawal of the tacrolimus medication and the need for reintroduction of the medication.

In conclusion, rectal tacrolimus is effective for the induction of response, and remission, for patients with active distal UC that has been resistant to topical and oral therapies.

## Supplementary Material

Note: To access the supplementary material accompanying this article, visit the online version of *Clinical Gastroenterology and Hepatology* at [www.cghjournal.org](http://www.cghjournal.org), and at <http://dx.doi.org/10.1016/j.cgh.2017.02.027>.

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**Reprint requests**

Address requests for reprints to: Ian Craig Lawrence, MBBS (Hons), PhD, School of Medicine and Pharmacology, University of Western Australia, Harry Perkins Institute for Medical Research, Fremantle WA, Australia. e-mail: [ian.Lawrence@uwa.edu.au](mailto:ian.Lawrence@uwa.edu.au); fax: +61893822460.

**Conflicts of interest**

The authors disclose no conflicts.