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Volume 10 • Issue 9 • October 2023

ISSN 2050-4365

Ondansetron for irritable bowel syndrome with diarrhoea: randomised controlled trial

David Gunn, Rabia Topan, Ron Fried, Ivana Holloway, Richard Brindle, Suzanne Hartley, Lorna Barnard, Maura Corsetti, Mark Scott, Adam Farmer, Ayesha Akbar, Maria Eugenicos, Nigel Trudgill, Kapil Kapur, John McLaughlin, David S Sanders, Arvind Ramadas, Peter Whorwell, Lesley Houghton, Phil G Dinning, Qasim Aziz, Alexander C Ford, Amanda Farrin and Robin Spiller

DOI 10.3310/YTFW7874

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Disclosure of interests

Full disclosure of interests: Completed ICMJE forms for all authors, including all related interests, are available in the toolkit on the NIHR Journals Library report publication page at https://doi.org/10.3310/YTFW7874.

Primary conflicts of interest: Robin Spiller has received research grants from Zespri International and Sanofi-Aventis and speakers'f fees from Ardelyx, Menarini and Ferrer. S Mark Scott has received payment or honoraria for lectures, presentations, speakers bureaus, manuscript writing or educational events Medical Measurement Systems (MMS)/Laborie. John McLaughlin is an unpaid trustee and deputy chair, Guts UK charity. Ayesha Akbar has received speakers' fees from the following pharmaceutical companies: Takeda, Dr Falk, Janssen, Tillots and Galapagos. Maura Corsetti is Co-Chief Investigator in a research grant funded by Sanofi, has received consulting fees from Sanofi, Takeda, Mayoly, RB and Arena, and has received payments or honoraria for lectures, presentations, speakers bureaus, manuscript writing or educational events from Sanofi and Mayoly. Lesley Houghton has received consulting fees from Ironwood Pharmaceuticals, USA, and has a leadership or fiduciary role in other boards, societies, committees or advocacy groups, paid or unpaid in Rome V International Working Group – Fundamentals of Neurogastroenterology: Physiological Aspects and Clinical Implications Section Chair, Member of British Society of Gastroenterology Committee – Guidelines for irritable bowel syndrome, and Member of British Society of Gastroenterology Committee – Guidelines for functional dyspepsia. Qasim Aziz has received grants or contracts from any entity from Classado Pharmaceuticals and Allergan, has received payment or honoraria for lectures, presentations, speakers bureaus, manuscript writing or educational events from Bromatec, has participated on a Data Safety Monitoring Board or Advisory Board for the MHRA GRID EAG Committee, has a leadership or fiduciary role in other boards, societies, committees or advocacy groups, paid or unpaid at the European Society of Neurogastroenterology and Motility, and has stock or stock options in My Health Chart, The Physicians' Clinic and London Digestive Centre. Amanda Farrin is a NIHR Senior Investigator, has received research grants from NIHR and was a member of HTA Efficient Study Designs Board (13 October 2014–17 December 2014), NIHR CTU Standing Advisory Committee (1 February 2022–1 May 2026), HTA Funding Committee Policy Group (formerly CSG) (12 March 2014–31 October 2018) and the HTA Clinical Evaluation and Trials Committee (1 November 2014–30 November 2018). Suzanne Hartley has support for the present manuscript, role on the project was partly funded by the project grant (National Institute for Health Research Efficacy and Mechanism Evaluation) and grants or contracts from any entity, role has been partly funded by other NIHR grants. Alexander C Ford has received consulting and/or speakers' fees from Dr Falk, GE Healthcare, Ironwood, Novartis and Takeda Pharmaceuticals, and has received research funding from Dr Falk and Tillotts, UK.

Published October 2023
DOI: 10.3310/YTFW7874

This report should be referenced as follows:

Gunn D, Topan R, Fried R, Holloway I, Brindle R, Hartley S, *et al*. Ondansetron for irritable bowel syndrome with diarrhoea: randomised controlled trial. *Efficacy Mech Eval* 2023;**10**(9). https://doi.org/10.3310/YTFW7874

Efficacy and Mechanism Evaluation

ISSN 2050-4365 (Print)

ISSN 2050-4373 (Online)

Efficacy and Mechanism Evaluation (EME) was launched in 2014 and is indexed by Europe PMC, DOAJ, Ulrichsweb™ (ProQuest LLC, Ann Arbor, MI, USA) and NCBI Bookshelf.

This journal is a member of and subscribes to the principles of the Committee on Publication Ethics (COPE) (www.publicationethics.org/).

Editorial contact: journals.library@nihr.ac.uk

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The EME programme is funded by the Medical Research Council (MRC) and the National Institute for Health and Care Research (NIHR), with contributions from the Chief Scientist Office (CSO) in Scotland and National Institute for Social Care and Health Research (NISCHR) in Wales and the Health and Social Care Research and Development (HSC R&D), Public Health Agency in Northern Ireland.

This report

The research reported in this issue of the journal was funded by the EME programme as project number 15/74/01. The contractual start date was in February 2017. The final report began editorial review in May 2022 and was accepted for publication in February 2023. The authors have been wholly responsible for all data collection, analysis and interpretation, and for writing up their work. The EME editors and production house have tried to ensure the accuracy of the authors' report and would like to thank the reviewers for their constructive comments on the final report document. However, they do not accept liability for damages or losses arising from material published in this report.

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Abstract

Ondansetron for irritable bowel syndrome with diarrhoea: randomised controlled trial

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Background: Irritable bowel syndrome with diarrhoea is characterised by frequent, loose or watery stools with associated urgency, resulting in marked reduction of quality of life. Ondansetron, a 5-hydroxytryptamine-3 receptor antagonist, has been shown to benefit patients with irritable bowel syndrome with diarrhoea.

Objective: To evaluate the effect of ondansetron in irritable bowel syndrome with diarrhoea.

Design: Phase III, parallel-group, randomised, double-blind, multicentre, placebo-controlled trial in 400 patients, with embedded mechanistic studies.

Setting: Hospital, primary care and community.

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Participants: Eighty participants meeting Rome IV criteria for irritable bowel syndrome with diarrhoea.

Intervention: Ondansetron 4 mg (dose titrated up to two tablets three times a day) or matched placebo for 12 weeks.

Main outcome measures: **Clinical** – Primary patient-reported end point was % 'Food and Drug Administration-defined responders' over 12 weeks. Secondary end points were worst abdominal pain intensity, worst urgency, stool consistency, stool frequency, anxiety, depression and dyspepsia at 12 and 16 weeks.

Main outcome measures: **Mechanistic** – Whole gut transit time, faecal water, protease (FP), bile acids and assessment of rectal sensitivity using a barostat.

Results: **Clinical** – The study closed early due to slow recruitment. Between 1 January 2018 and 11 May 2020, 80 patients were recruited and randomised (20% of target), 37 to ondansetron, 43 to placebo. Discontinuations (4 ondansetron; 2 placebo) meant 75 completed the 12-week trial treatment. There were four protocol violations. In the intention-to-treat analysis, 15 (40.5%) on ondansetron were primary end-point responders (95% CI 24.7% to 56.4%), and 12 (27.9%) on placebo (95% CI 14.5% to 41.3%), $p = 0.19$, adjusted OR 1.93 (0.73, 5.11). Pain intensity reduction occurred in 17 (46.0%) on ondansetron (95% CI 29.9% to 62.0%) and 16 (37.2%) on placebo (95% CI 22.8% to 51.7%), $p = 0.32$. Improvement in stool consistency occurred in 25 (67.6%) on ondansetron (95% CI 52.5% to 82.7%) and 22 (51.2%) on placebo (95% CI 36.2% to 66.1%), $p = 0.07$. Use of rescue medication, loperamide, was lower on ondansetron [7 (18.9%) vs. 17 (39.5%)]. Average stool consistency in the final month of treatment reduced significantly more on ondansetron, adjusted mean difference −0.5 [standard error (SE) 0.25, 95% CI (−1.0 to −0.02), $p = 0.042$]. Ondansetron improved dyspepsia score (SFLDQ), adjusted mean difference −3.2 points [SE 1.43, 95% CI (−6.1 to −0.4), $p = 0.028$]. There were no serious adverse events.

Mechanistic – mean (SD). Ondansetron increased whole gut transit time between baseline and week 12 by 3.8 (9.1) hours on ondansetron, significantly more than on placebo −2.2 (10.3), $p = 0.01$. Mean volume to reach urgency threshold using the barostat increased on ondansetron by 84 (61) ml and 38 (48) ml on placebo, $n = 8$, $p = 0.26$. Ondansetron did not significantly alter protease, faecal water or bile acids. Changes in referral pathways substantially reduced referrals, impairing recruitment, which meant the study was underpowered.

Conclusion: Our results are consistent with previous studies and confirmed ondansetron improves stool consistency and urgency but showed minor effect on pain. We plan to undertake a simplified version of this trial overcoming the changed referral pathways by recruiting in primary care, using software linked to primary care records to identify and randomise patients with irritable bowel syndrome with diarrhoea to ondansetron or placebo and remotely follow their progress; thus minimising barriers to recruitment.

Trial registration: This trial is registered as ISRCTN17508514.

Funding: This project was funded by the National Institute for Health and Care Research (NIHR) Efficacy and Mechanism Evaluation programme and will be published in full in *Efficacy and Mechanism Evaluation*; Vol. 10, No. 9. See the NIHR Journals Library website for further project information.

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Report Supplementary Material 1 Dose Titration_SSOP02_V1.0

Report Supplementary Material 2 TRTION Participant Dose Titration Instructions v5.0

Supplementary material can be found on the NIHR Journals Library report page (https://doi.org/10.3310/YTFW7874).

Supplementary material has been provided by the authors to support the report and any files provided at submission will have been seen by peer reviewers, but not extensively reviewed. Any supplementary material provided at a later stage in the process may not have been peer reviewed.

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List of abbreviations

5HT	5-hydroxytryptamine
$5HT_3RA$	5-HT3 receptor antagonist
AE	adverse event
ALT	alanine transaminase
ANOVA	analysis of variance
BAD	bile acid diarrhoea
BP	blood pressure
BSFS	Bristol Stool Form Scale/Score
CI	confidence interval
CNS	central nervous system
CONSORT	Consolidated Standards of Reporting Trials
CTRU	clinical trials research unit
DMEC	Data Monitoring and Ethics Committee
ECG	electrocardiogram
FBA	faecal bile acid
FBC	full blood count
FDA	Food and Drug Administration
FP	faecal protease
FW	faecal water
GP	general practitioner
HADS	Hospital Anxiety and Depression Scale
HAPC	high-amplitude propagated contraction
HRM	high-resolution manometry
IBD	inflammatory bowel disease
IBS	irritable bowel syndrome
IBS-D	irritable bowel syndrome with diarrhoea
IBS-QOL	irritable bowel syndrome–quality of life
IBS-SSS	Irritable Bowel Syndrome Severity Scoring System
IMP	investigational medicinal product
ISRCTN	International Standard Randomised Controlled Trial Register
ITT	intention to treat
MAR	missing at random
NICE	National Institute for Health and Care Excellence
PHQ-12	Patient Health Questionnaire 12
PI	principal investigator
PIC	Participant Identification Centre
PIS	patient information sheet
PP	per protocol
PPI	patient participation and involvement
QoL	quality of life
QTc	corrected QT interval
SAE	serious adverse events
SD	standard deviation
SE	standard error
SeHCAT	selenium homocholic acid taurine
SFLDQ	short-form Leeds dyspepsia questionnaire
SPE	solid-phase extraction
TCA	tricyclic antidepressant
t.d.s.	three times a day
TPH-1	tryptophan hydroxylase-1
VAS	visual analogue scale
WGTT	whole gut transit time

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Plain language summary

Background

Irritable bowel syndrome with diarrhoea is characterised by frequent, loose, or watery bowel movements with marked reduction of quality of life. A previous small study suggested ondansetron benefits patients with irritable bowel syndrome with diarrhoea.

Methods

A clinical trial aiming to recruit 400 patients meeting established criteria for irritable bowel syndrome with diarrhoea from 18 centres throughout the UK. Patients received either ondansetron or placebo for 12 weeks but neither the investigator and nor patient could tell which they were receiving. They recorded their worst abdominal pain, stool frequency and consistency daily. The main end point was the proportion of patients meeting a standard recommended by the U.S. Food and Drug Administration (FDA). Being called a "FDA responder" meant they showed reductions to both pain and days with loose bowel movements. Other less important end points included pain intensity, stool consistency and frequency. We also measured the time for content to pass through the gut (whole gut transit time).

Results

The study closed early due to slow recruitment with 80 patients randomised. There were 40.5% of responders in the ondansetron group and 27.9% in the placebo group; however, due to low numbers these differences could be due to chance. Ondansetron produced a significant improvement in average stool consistency in the final month of treatment. Ondansetron slowed whole gut transit time which increased from baseline to 12 weeks by a mean of 3.8 hours, while it fell 2.2 hours on placebo, a difference unlikely to be due to chance.

Conclusion

These results are consistent with previous studies showing that ondansetron improves stool consistency and slows transit. However, because the numbers recruited were smaller than planned, the apparent improvement in "FDA responder" rate could have been due to chance. A further larger trial is needed to confirm the benefit of ondansetron which should be done in primary care where most patients are to be found.

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Scientific summary

Background

Irritable bowel syndrome (IBS), which affects around 10% of the population, accounts for 1.8 million consultations/year in primary care in England and Wales (0.6 million patients). Symptoms of IBS with diarrhoea (IBS-D) include frequent, loose or watery stools with associated urgency, which can severely limit socialising, travelling and eating out, with resulting marked reduction in quality of life and loss of work productivity. Around one-third of all IBS patients meet Rome criteria for IBS-D. When patients are asked to rank symptoms in order of importance, the erratic bowel habit is rated first, followed by abdominal pain and, for those with diarrhoea, urgency.

Ondansetron, a 5-hydroxytryptamine-3 ($5HT_3$) receptor antagonist, has an excellent safety record for over 20 years as an antiemetic, but is only exceptionally used in the treatment of IBS-D. It has, however, been shown to slow colonic transit and in a small randomised, placebo-controlled, crossover pilot study, benefited patients with IBS-D. While the current trial was ongoing, a separate trial in the USA using a fixed dose bimodal release formulation (3 mg ondansetron +9 mg delayed release formulation) also reported improvement in stool consistency but not pain.

Objectives

Our primary aim was to determine the efficacy of generic ondansetron compared to placebo in controlling the symptoms of IBS-D using the US FDA-recommended combined end point in which a responder is defined as a patient who met the response criteria for both pain and bowel habit for 6 out of 12 weeks of the trial. Secondary end points included its effect on the characteristic abnormalities of stool consistency, frequency and urgency as well as abdominal pain, satisfactory relief of IBS symptoms, mood and use of rescue medication and to determine the effect of 12 weeks ondansetron over the 1 month after discontinuation, as well as safety.

The study also included mechanistic studies to examine the correlation of rectal sensitivity and compliance, faecal bile acids (FBAs) and proteases and postprandial sigmoid motility with the baseline symptoms of our IBS-D patients. We also attempted to determine whether ondansetron significantly altered these biomarkers compared to placebo.

Methods

Treatment of irritable bowel syndrome using titrated ondansetron trial (TRITON) was a multisite, parallel-group, randomised, double-blinded, placebo-controlled trial, with embedded mechanistic studies within selected sites. Our aim was to determine the superiority of ondansetron compared with placebo. We aimed to randomise 400 patients with IBS-D on a 1 : 1 basis to receive either ondansetron or placebo. Both treatments were administered for 12 weeks in oral doses ranging from 4 mg every third day to 24 mg daily. Dose titration was undertaken in the first 2 weeks of the study to avoid constipation, which at a standard dose occurs in one-quarter of patients. This was achieved by frequent consultation with the research nurse, starting with 1 × 4 mg tablet per day and increasing in increments every 2 days to a maximum of 2 tablets thrice daily. If constipation developed, the treatment was stopped to allow the return of bowel movements and then restarted at a lower dose, typically one every alternate day or one every second day. Rescue medication of loperamide was discouraged but allowed exceptionally for uncontrolled diarrhoea and was documented in the daily diary.

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The primary outcome of response for both reduction in pain intensity and improvement in stool consistency was assessed over the 12 weeks post randomisation. Secondary and safety outcomes were measured up to 16 weeks post randomisation. Symptoms that were recorded daily included (1) stool consistency and abdominal pain (measured by both paper diary and daily text message); (2) stool frequency, urgency of defaecation, use of rescue medication (defined as the use of loperamide) over 12 weeks of treatment and the answer to the question in the diary 'Overall, have you had satisfactory relief from your IBS symptoms in the past week?'.

Irritable bowel syndrome symptom severity [measured by the IBS Severity Scoring System (IBS-SSS)], dyspepsia [using the Short Form Leeds Dyspepsia Questionnaire (SFLDQ)], quality of life and mood [using the IBS Quality of Life (IBS-QOL) and Hospital Anxiety and Depression Scale (HADS) questionnaires], and somatic symptoms [using the Patient Health Questionnaire 12 Somatic Symptoms (PHQ-12) questionnaire] were assessed by patient-reported questionnaires at the baseline and 12 weeks post randomisation.

The trial also assessed possible underlying mechanisms of any effect of ondansetron on changes in the primary and secondary end points. Whole gut transit was measured at baseline and 12 weeks using radio-opaque markers and an abdominal X-ray. High-resolution manometry was performed at baseline and after 8–11 weeks of treatment at two centres to assess whether ondansetron decreased the number of high-amplitude propagating contractions (HAPCs) or increased the percentage time occupied by cyclical retrograde propagated contractions. Barostat assessment was performed at baseline and after 8–11 weeks of treatment at two centres in order to assess if ondansetron increases rectal compliance or decreases sensitivity (manifested as increased pressure thresholds for pain and urgency). Stool samples were assessed for faecal water % (FW), faecal protease (FP) and FBAs.

Clinical results

The study closed early due to slow recruitment with just 80 patients randomised; 37 to ondansetron and 43 to placebo. Four patients discontinued ondansetron and one placebo during 12-week randomised treatment. Four were excluded from the per-protocol population due to major protocol violations.

In the intention to treat (ITT) analysis, 15 patients (40.5%) on ondansetron achieved the primary end point response [95% confidence interval (CI) 24.7% to 56.4%], compared to 12 (27.9%) patients on placebo (95% CI 14.5% to 41.3%), $p = 0.19$, adjusted OR 1.93 (0.73, 5.11). Response for pain intensity reduction was achieved by 17 (46.0%) on ondansetron (95% CI 29.9% to 62.0%) and 16 (37.2%) on placebo (95% CI 22.8% to 51.7%), $p = 0.32$, adjusted OR 1.61 (0.63 to 4.12). Response for stool consistency improvement was reported by 25 (67.6%) on ondansetron (95% CI 52.5% to 82.7%) and 22 (51.2%) on placebo (95% CI 36.2% to 66.1%), $p = 0.07$, adjusted OR 2.45 (0.92, 6.52). Overall use of the rescue medication, loperamide, was 39.5% ($n = 17$) on placebo compared with 18.9% ($n = 7$) on ondansetron. However, by week 12, loperamide use fell to 13.5% on ondansetron versus 25.6% on placebo. Average stool consistency in the final month of treatment fell significantly more on ondansetron than placebo, adjusted mean difference −0.5 [standard error (SE) 0.25, 95% CI (−1.0 to −0.02), $p = 0.042$]. Ondansetron improved the dyspepsia score (SFLDQ) significantly more than placebo; the largest reduction being in symptoms of indigestion and nausea. The adjusted mean difference in the total score compared to placebo was −3.2 points [SE 1.43, 95% CI (−6.1, to −0.4), $p = 0.028$]. Ondansetron was well tolerated with most adverse reactions being mild or moderate and not significantly greater than on placebo. The commonest was constipation, reported in 32% on ondansetron and 23% on placebo, of which 75% and 80%, respectively, were rated as mild. Just two patients withdrew citing constipation as the cause.

Mechanistic results

Results are expressed as mean (SD). Comparing baseline and week 12 showed ondansetron increased average whole gut transit 3.78 (9.1) hours on ondansetron significantly more than placebo −2.2 (10.3),

p = 0.01. Mean volume to reach urgency threshold using the barostat increased on ondansetron by 84 (61) ml and 38 (48) ml on placebo, n = 8; the difference was not significant, p = 0.26. Too few underwent manometry to allow meaningful assessment of the effect of ondansetron but anecdotally one patient who had a dramatic clinical improvement showed a loss of HAPCs and an increase in retrograde contractions, but this could have been due to chance. Ondansetron appeared not to significantly alter FP, though overall the increase in whole gut transit time from baseline to week 12 was correlated with a decrease in FP. There were no significant changes in FBAs and no evidence that ondansetron altered these though we did confirm that we had effectively excluded those with bile acid diarrhoea (BAD). The ratio of secondary to primary bile acids, a measure of bacterial metabolism of bile acids, increased substantially on ondansetron from 9.7 (7.08) to 21.4 (32.9) and less so on placebo from 22.84 (58.23) to 28.61 (31.42). However, owing to small numbers and wide variability these differences were not significant.

Limitations

Two previous studies in Nottingham had recruited 120 and 136 IBS-D patients within 2 years so we did not anticipate problems with recruitment. However, changes in referral pathways from primary to secondary care substantially reduced referrals to our coinvestigators who were all in secondary care, thus impairing recruitment. The power calculations required 400 to achieve 90% power to detect a 15% difference in primary end point, so the study is substantially underpowered. Use of loperamide did somewhat complicate interpretation since those on placebo used more rescue medication reducing the size of the effect on transit and stool consistency.

Conclusion

Despite being underpowered for our primary end point, our results are consistent with previous studies and confirmed ondansetron improves stool consistency but showed little effect on pain. Ondansetron significantly slowed whole gut transit time. Ondansetron reduced sensitivity to rectal distension more than placebo without altering compliance, but numbers were too small to achieve statistical significance. This could plausibly contribute to the reduction in urgency and stool frequency but needs repeating with larger numbers to be sure it was not due to chance. We found no evidence that rectal sensitivity was related to either faecal protease or bile acids. The manometry studies were underpowered but anecdotally ondansetron appeared to alter rectosigmoid motor patterns in a way that could reduce inflow of stool to the rectum.

Future work

We plan to do a simplified version of this trial, using an efficient and remote process, to overcome the changed referral pathways by recruiting in primary care. We will search for patients who have had a diagnosis of chronic diarrhoea and the recommended screening including a normal full blood count, a negative tissue transglutaminase (excluding coeliac disease) and a normal faecal calprotectin using software linked to primary care records. This will allow rapid screening of large numbers of patients to identify and approach patients with IBS-D who meet criteria to take part in a randomised trial of ondansetron or placebo, thus minimising barriers to recruitment. We would remove the pain threshold, which would increase the number of eligible patients and facilitate recruitment. Not allowing loperamide as rescue medication would simplify interpretation and dropouts would be treated as treatment failures. Further streamlining by removing all additional tests that were included in the current trial, as well as efficient trial processes, including e-consent, remote blood and stool samples (if required), and online questionnaires would also optimise recruitment.

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Study registration

This trial is registered as ISRCTN17508514.

Funding

This project was funded by the National Institute for Health and Care Research (NIHR) Efficacy and Mechanism Evaluation programme and will be published in full in *Efficacy and Mechanism Evaluation*; Vol. 10, No. 9. See the NIHR Journals Library website for further project information.

Chapter 1 Introduction

Background and rationale

Irritable bowel syndrome (IBS), which affects around 4% of the world population,[1] accounts for around 1% of the 285 million consultations per year in primary care in England and Wales,[2] which is approximately 2.8 million. Around one-third of these patients meet criteria for IBS with diarrhoea (IBS-D). Symptoms include frequent, loose or watery stools with associated urgency, which can severely limit socialising, travelling and eating out. This results in a reduction in quality of life (QoL) and loss of work productivity. When patients with IBS are asked to rank symptoms in order of importance, erratic bowel habit is rated first, followed by abdominal pain and, for those with diarrhoea, urgency.[3] This can often be associated with incontinence, which is socially debilitating, but often under-reported.[4] Current over-the-counter treatments for patients with IBS-D such as loperamide reduce bowel frequency, but do not improve abdominal pain[5,6] and often lead to constipation. The lack of effective treatments results in frequent referrals to secondary care, and such patients have in the past been a significant proportion of gastroenterology outpatients. A previous meta-analysis showed that the 5-hydroxytryptamine-3 receptor antagonists ($5HT_3$RAs) alosetron and cilansetron benefitted such patients, improving stool consistency, and reducing both frequency and urgency of defaecation.[7] However, these drugs had serious side effects, including constipation in 25% of patients and, rarely, ischaemic colitis (1 in 700). Alosetron was initially withdrawn and is now available in the USA, only through a risk evaluation and mitigation strategy and is not available in Europe. Cilansetron never came to market, while ramosetron, another $5HT_3$RA, is only available in Japan, where it is licensed for IBS-D with several good-quality trials confirming its benefit.[8,9]

Ondansetron is a potent, highly selective $5HT_3$RA, which blocks 5HT3 receptors in the gastrointestinal tract. Penetration across the blood–brain barrier is limited, with cerebrospinal fluid (CSF) concentration < 15% of plasma levels so central nervous system (CNS) adverse effects are few. Ondansetron was developed and is currently licenced for use in adults and children for the management of nausea and vomiting induced by cytotoxic chemotherapy and radiotherapy (mediated by local release of serotonin in the gut from enterochromaffin cells), and for the prevention and treatment of postoperative nausea and vomiting. Constipation is an unintended side effect of ondansetron, which was first shown to slow colonic transit 30 years ago.[10,11] Ondansetron is widely used and unlike alosetron has not been associated with ischaemic colitis. We previously performed a randomised double-blind, placebo-controlled, crossover pilot study of two 5-week periods of treatment recruiting 120 IBS-D patients randomised to receive either ondansetron [2 mg up to 8 mg three times a day (t.d.s)] followed by placebo, or placebo followed by ondansetron with washout period of 2–3 weeks.[12] The primary outcome measure for the study was the difference in average stool consistency in the last 2 weeks of treatment, which showed a highly significant improvement with ondansetron versus placebo. We also showed significant benefits for both urgency and stool frequency, with associated slowing of whole gut transit.[12] Despite having limitations, results of this pilot study were very encouraging, supporting our clinical experience of ondansetron's benefits. Recently a novel formulation of ondansetron (Bekinda) comprising 3 mg immediate release and 9 mg slow-release has been shown in a 12-week randomised, placebo-controlled trial to improve stool consistency, though it was underpowered to show significant benefit for pain. Currently there is lack of understanding as to exactly how it works, nor can we predict the individual dose required for optimum effect, which varies widely.[13] One key effect we found, also seen with other $5HT_3$RAs,[14] was a marked reduction in urgency, which may play an important role in improving QoL for patients with IBS-D.[15]

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Potential mechanisms of action of $5HT_3$ receptor antagonists

Transit

5-HT3 receptor antagonists slow transit,[10,11] an effect we found particularly marked in the left colon of IBS-D patients and rectosigmoid region, but the underlying mechanism was unclear.[12]

Tone and motility

An early colonic barostat study showed a reduction in the postprandial rise in colonic tone with ondansetron in both carcinoid syndrome patients[16] and healthy volunteers,[17] which would be predicted to slow colonic transit. Previous studies of the impact of $5HT_3$RAs on human colonic motility[18–20] were however paradoxical in showing that the $5HT_3$RAs tropisetron, alosetron and cilansetron all increased periprandial frequency of colonic contractions, and mean amplitude of contractions in the left colon. We hypothesised that $5HT_3$RAs increase retrograde sigmoid motility, perhaps enhancing 'brake' function,[21,22] which would be a novel mode of action.

Proteases

In our pilot study we showed the decrease in urgency associated with ondansetron use correlated directly with the reduction in faecal protease (FP),[23] but whether this represents a true causal relationship, or just an epiphenomenon, is unclear. FPs have been shown to be increased in IBS-D and, at least in animal models, cause hypersensitivity to rectal distension via activation of protease-activated receptors type 2 (PAR2).[24] We have shown that most FPs are endogenous,[23] representing pancreatic enzymes that have escaped degradation by colonic bacteria. We hypothesise that slowing gut transit with ondansetron reduces FP, by allowing time for bacterial degradation, and that this may contribute to the beneficial effects of ondansetron on urgency. Reducing faecal tryptase might improve the anal soreness that is commonly reported by patients with IBS-D.

Bile acids

Bile acids have also been shown to sensitise the rectum,[25] and elevated faecal bile acids (FBAs) have been shown by several groups in patients with IBS-D.[26] Slowing transit may increase the time for bile acid deconjugation by colonic bacteria, and therefore enhance absorption, but how important this is in reducing rectal sensitivity, compared with the effects on FPs, is unclear.

Genetic factors altering sensitivity to $5HT_3$RAs

We have shown that individuals vary widely in their responsiveness to ondansetron, explaining why trials using fixed doses of $5HT_3$RAs result in severe constipation in around one in four patients. Meta-analysis gives a relative risk of constipation of 4.3 (3.3–5.6).[7] However, when patients were allowed to dose titrate we found that constipation was rare, occurring in only 9% of patients, most of whom responded to dose reduction and only 2% discontinuing ondansetron because of this.[12] However, the required dose of ondansetron ranged from 4 mg on alternate days to 8 mg three times a day (t.d.s.). The reasons for this variation are unclear, but recent evidence suggests that responsiveness to $5HT_3$RAs might be linked to polymorphisms in the genes controlling 5HT synthesis. Serotonin availability in the rectal mucosa is thought to be determined by the activity of the rate-limiting synthetic enzyme tryptophan hydroxylase-1 (TPH-1), which produces serotonin in enterochromaffin cells. A recent small study showed TPH-1 messenger ribonucleic acid (mRNA) levels in rectal mucosa (and thus presumably serotonin synthesis rate) were approximately doubled in responders to another $5HT_3$RA, ramosetron, compared with non-responders, and that this was linked to the TPH-1 genotype.[27] TPH-1 rs211105 minor allele G was found in 44% of non-responders, but only 4% of responders, indicating that possessing the major allele increases responsiveness to the drug. It was also associated with worse diarrhoea, possibly because of the greater 5HT synthesis.

Objectives

The overall aim was to determine the efficacy and safety of ondansetron in patients with symptoms of IBS-D, in particular evaluating the effect on the characteristic abnormalities of stool consistency, frequency and urgency as well as abdominal pain, satisfactory relief of IBS symptoms, mood and use of rescue medication and to determine the effect of 12 weeks ondansetron over 1 month after discontinuation.

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Chapter 2 Methods

Trial design

A multisite, parallel-group, randomised, double-blinded, placebo-controlled trial, with embedded mechanistic studies recruiting patients from primary and secondary care as well as the community.

Participants

Patients were screened using a standardised protocol to establish eligibility using the following criteria.

Inclusion criteria

These were as follows:

- patients had to meet modified Rome IV criteria for IBS-D (see as previously published);
- patients had to be aged ≥ 18 years;
- patients should have completed standardised workup to exclude:
    - microscopic colitis (colonoscopy or flexible sigmoidoscopy with colonic biopsies);
    - bile acid diarrhoea (BDA) [Selenium homocholic acid taurine (SeHCAT) results of > 10%, C4 results of < 19 ng/ml or failed 1-week trial of a bile acid binding agent (colestyramine 4 g t.d.s., colesevelam 625 mg t.d.s. or equivalent)];
    - lactose malabsorption (suggested but not mandated negative lactose breath hydrogen test, negative clinical challenge or failure to respond to lactose-free diet);
    - coeliac disease [tissue transglutaminase (tTG) or duodenal biopsy];
- patients of childbearing potential or with partners of childbearing potential had to agree to use methods of medically acceptable forms of contraception during the trial and for 90 days after completion of trial medication;
- for women of childbearing potential, a negative pregnancy test was performed within 72 hours of confirmation of eligibility;
- weekly average worst pain score had to be ≥ 30 on a 0 to 100-point scale;
- stool diary needed to show stools with a consistency of 6 or 7 on the Bristol Stool Form Scale (BSFS) for 2 or more days per week.

Modified Rome IV diagnostic criteria for irritable bowel syndrome with diarrhoea

Must fulfil the following criteria for the past 3 months:

1. recurrent abdominal pain at least weekly;
2. pain is associated with two or more of the following criteria:
    a. related to defaecation;
    b. associated with a change in frequency of stool;
    c. associated with a change in form (appearance) of stool;
3. symptom onset at least 6 months prior to diagnosis;
4. > 25% of abnormal stools are loose (BSFS 6 or 7) but < 25% are hard (BSFS 1 or 2).

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Exclusion criteria

- Gastrectomy.
- Intestinal resection.
- Other known organic gastrointestinal diseases [e.g. inflammatory bowel disease (Crohn's disease, ulcerative colitis)].
- Unable or unwilling to stop restricted medication, including regular loperamide, antispasmodics (e.g. hyoscine butylbromide, mebeverine, peppermint oil, alverine citrate), eluxadoline, tricyclic antidepressant (TCA) doses > 30 mg/day or other drugs likely in the opinion of the investigator to alter bowel habit. These medicines should be discontinued for a 7-day washout period prior to registration.
- Corrected QT (QTc) interval ≥ 450 milliseconds for men or ≥ 470 milliseconds for women [assessed within the last 3 months by electrocardiogram (ECG)].
- Previous chronic use of ondansetron or contraindications to it.
- Pulse, blood pressure (BP) and laboratory blood values outside the normal ranges according to the site's local definition of normal [assessed within the last 3 months]. Note minor rises in alanine transaminase (ALT) (< 2 × upper limit of normal) will be acceptable, but the patient's general practitioner (GP) will be informed if they remain elevated at the end of the trial.
- Women who were pregnant or breastfeeding.
- Patients currently participating in, or who have been in, a trial of an investigational medicinal product (IMP) in the previous 3 months, where the use of the IMP may cause issues with the assessment of causality in this trial.
- Patients who had started or altered dosing of selective serotonin reuptake inhibitors or TCAs in the last 3 months, or who were planning to change the dose during the trial.
- Patients currently taking and unwilling or unable to stop apomorphine or tramadol (which interact with ondansetron).
- Patients with only stools of consistency 7 on the BSFS for 7 days a week.

Patients taking QT-prolonging or cardiotoxic drugs were reviewed by the local principal investigator (PI) to determine whether they were suitable for the trial.

Intervention

Participants were randomised on a 1 : 1 basis to initially receive either ondansetron 4 mg or matched placebo, one tablet daily. Both treatments were administered in oral doses which ranged from one tablet every third day to a maximum of six tablets daily for 12 weeks. The optimum dose was established by dose titration monitored closely by the research nurse using a standardised advice protocol in the first 2 weeks of the trial to avoid constipation, which at a standard dose occurs in one-quarter of patients. Patients were told to increase or decrease dosage every 2 days according to their stool consistency. If stools became hard or there was no bowel movement on day 2, they were asked to stop the drug for 1 day and recommence at a lower dose going from one tablet daily to one tablet on alternate days. If stools still remained hard or infrequent, they were asked to reduce to one tablet every third day. Continuing loose stools led to the advice to increase the daily dose by one tablet every 2 days, while constipation led to a reduction in dosage. Most patients had established a stable dose by 2 weeks. Patients were reminded to take their medication regularly on each trial visit, as well as during the phone calls within the 2-week titration period. Counts of any remaining IMP were done at visits 4 and 5.

Participant timeline

This is summarised in *Appendix 1*, *Table 34* with details given below.

Visit 1

Potential trial candidates attended their first visit for registration and consent by the PI or delegate. If required, further tests to exclude diagnoses other than IBS-D were arranged. These included a SeHCAT scan, serum C4 level (Edinburgh centre only) or a 1-week trial of a bile acid binding agent to assess for bile acid malabsorption (unless done within the last 5 years), and a colonoscopy (unless done within 2 years, or 5 years if they also had a current normal calprotectin) to assess for microscopic colitis. Baseline serum blood tests, vital signs, demographics (date of birth, gender, ethnicity and smoking history) and an ECG were obtained. Current medications were reviewed, and those unable to discontinue drugs likely to alter bowel habit were unable to enter the trial. Patients on QT-prolonging drugs and cardiotoxic drugs were reviewed by the PI for suitability for the trial, as high-dose ondansetron may increase the risk of QT prolongation and arrhythmias. Eligible and consenting patients were registered and allocated a unique trial ID.

All patients were asked to complete a 2-week daily diary recording stool frequency, stool consistency for each stool (using the BSFS), worst abdominal pain (on a scale of 0–100), worst bowel movement urgency (on a scale 0–100) and if they had used loperamide that day. In addition, patients had the option to be sent two automated text messages each day, something most participants initially agreed to do. The first message asked the patient if they had passed a stool of consistency 6 or 7 on the BSFS. They had to reply with either a yes or no. The second text message asked what their worst abdominal pain score was that day. The patient could respond with a number from a scale of 0–100 (where 0 is no pain and 100 is the worst imaginable pain).

Visit 2

Two weeks later the patient returned to confirm eligibility. The diary was used to confirm they had stool consistency BSFS 6–7 for more than 2 days a week and did not meet the exclusion criteria of having only BSFS 7. They also had to have a weekly average worst pain score ≥ 30. For patient consenting to the whole gut transit study, Transit-Pellet capsules containing markers were dispensed and the abdominal X-ray appointment confirmed for the morning of visit 3. If the patient had consented to one or both mechanistic studies, appointments for baseline assessment were arranged prior to visit 3.

Visit 3

On visit 3, patients underwent a pregnancy test if applicable, whole gut transit assessment by abdominal X-ray (if consented), rigid sigmoidoscopy (if consented), completion of baseline questionnaire booklet [including IBS-SSS, Short-form Leeds dyspepsia questionnaire (SFLDQ), Hospital Anxiety and Depression Scale (HADS), Patient Health Questionnaire 12 (PHQ-12) and IBS-QOL questionnaires], and collection of stool, whole blood and serum samples (if consented). Patients were then randomised and given a 6-week patient diary and the trial medication in accordance with their blinded randomisation allocation. Patients were asked to record the following on a daily basis: stool frequency, consistency of each stool, worst abdominal pain experienced that day, worst bowel movement urgency, number of trial medication capsules taken and whether they had used loperamide that day. Every week the diary required them to respond to the question whether they felt that 'they have had satisfactory relief from their IBS symptoms that week'. If they agreed, the patient continued to receive two text messages each day for the rest of the trial, asking if they had passed a stool of a consistency of six or seven that day, and what their worst abdominal pain was that day.

During the first 2 weeks, patients were contacted every 2 days by the local site team to discuss bowel habit. The dose of ondansetron or placebo was titrated as required. Additional guidance on dose titration was given to each trial site in a standard operating procedure, and to the patient in a dose titration instruction leaflet (see *Report Supplementary Materials 1* and *2*). A check for serious adverse events (SAEs) was performed during each telephone call. The steady dose to be taken forward for the remainder of the trial was mostly confirmed in week 2, although patients could alter this during the 12 weeks, if required, to avoid constipation.

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Visit 4

At 6 weeks on the trial treatment, patients returned for their fourth visit. Diaries were collected, and the investigator asked whether any reportable adverse events (AEs) had occurred since the last visit. A pregnancy test was taken (if required) and concurrent medication reviewed to ensure these do not interfere with the trial medication. A further 6-week patient diary, trial medication and Transit-Pellet capsules (for use 6 days prior to visit 5) were dispensed. If the patient consented to mechanistic studies, appointments were confirmed and arranged as convenient between 8 and 11 weeks of treatment. Daily text messages continued for a further 6 weeks for those patients who agreed.

Visit 5

After 12 weeks on the trial medication, patients returned for visit 5. A pregnancy test was taken (if required), concurrent medication reviewed to ensure that these did not interfere with the trial medication and the investigator asked whether any reportable AEs had occurred since the last visit. Unused medication and completed patient diaries were collected. The plain abdominal X-ray to assess whole gut transit was performed for consenting patients. Serum and stool samples were collected for consenting patients, and all patients completed the 12-week questionnaire booklet, including the IBS-SSS, SFLDQ, HADS and IBS-QOL questionnaires. Patients were issued with a follow-up diary and continued to respond to text messages for a further 4 weeks.

Visit 6

Patients returned for the sixth and final visit, where the diary was collected, and the investigator asked about any reportable AEs since the last visit.

See *Appendix 1*, *Table 34* for a breakdown of activities per visit.

Trial outcomes

Primary outcome measure

Food and Drug Administration (FDA)-defined response in relation to abnormal defaecation and abdominal pain measured over 12 weeks post randomisation by patient diary and daily text message.

Definition: A FDA responder is a patient who records **both** a reduction in pain intensity **and** an improvement in stool consistency for at least 6 weeks of the 12-week treatment period, where:

- reduction in pain intensity is defined as ≥ 30% decrease from baseline in weekly average worst daily pain;
- improvement in stool consistency is defined as ≥ 50% decrease in the number of days per week with at least one loose stool [BSFS (21) 6 or 7)].

The two components to response (pain intensity and stool consistency) are also reported as individual outcomes.

Secondary outcome measures

Irritable bowel syndrome with diarrhoea symptoms measured by patient diary and daily text message over 12 weeks post randomisation and calculated for weeks 1–12 and for weeks 9–12:

1. abdominal pain (mean daily pain score, scored 0 = no pain to 100 = worst possible pain);
2. urgency of defaecation (mean daily urgency score, scored 0 = no urgency to 100 = worst imaginable urgency);
3. stool consistency (mean number of days per week with at least one loose stool; mean daily BSFS);
4. stool frequency (mean number of daily stools);
5. use of rescue medication (loperamide) over 12 weeks of treatment;

6. satisfactory relief assessed through answers to 'Overall, have you had satisfactory relief from your IBS symptoms in the past week?' Yes or No (measured by diary at the end of each week).

Self-reported symptoms, QoL and mood at 12 weeks post randomisation:

7. IBS symptom severity [measured by the IBS Severity Scoring System (IBS-SSS)];[28]
8. dyspepsia (using the SFLDQ);[29]
9. QoL and mood (IBS-QOL);[30]
10. HADS questionnaires;[31]
11. somatic symptoms [Patient Health Questionnaire 12 Somatic Symptoms (PHQ-12SS)] questionnaire.[32]

Post-treatment (weeks 13–16 post randomisation) recorded in patient diary:

12. abdominal pain;
13. urgency;
14. stool consistency;
15. stool frequency.

Safety:

16, SAEs assessed throughout 12-week treatment period;
17. AEs assessed 4 weeks after the end of treatment to determine if there are any persisting effects.

Summary of any changes to the project protocol

A number of minor amendments were made; a summary of protocol changes can be found in *Appendix 1*, *Table 35*. There were no changes to end points or drug administration.

These minor amendments included:

1. raising the permitted calprotectin level to be < 100 ng/mg in view of new national recommendations;
2. allowing a 7-day trial of colestyramine to replace SeHCAT testing to exclude BDA to overcome the problem of long delays in SeHCAT testing;
3. simplifying the information sheets and including the new data on congenital malformations which became available during the trial;
4. adding GP surgeries within the East Midlands as Participant Identification Centres (PICs), October 2018 and January 2019;
5. allowing patients who have had a colonoscopy within 10 years to exclude microscopic colitis to be enrolled in the trial. The exclusion criteria have been amended to clarify that patients would need to stop taking high-dose TCAs to 19 July 2019;
6. allowing new adverts, July 2019;
7. both the treatment patient information sheet (PIS) and the treatment + tests PIS have been updated to reflect the new safety information regarding the use of ondansetron in pregnancy, October 2019;
8. the SMS Guidance for patients has been updated. A fully tracked document has been included for review. Changes have been made to this document to make it easier to understand and to add clarity on what is involved in this aspect of the study, October 2019;
9. an SMS messages flowchart has been added. Due to an oversight the content of the SMS messages has not been previously submitted for approval, October 2019.

Mechanistic methods

The trial aimed to evaluate the possible mechanisms underlying any changes in the primary and secondary end points. The hypothesis we wished to test was that ondansetron would slow transit,

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particularly on the left side of the colon, possibly by stimulating retrograde sigmoid contractions. We planned to measure whole gut transit at baseline and 12 weeks (*n* = 400), using radio-opaque markers and an abdominal X-ray as previously described by Abrahamsson.[33] All patients were offered the opportunity to take part in high-resolution manometry (HRM) studies to be performed at baseline and after 8–11 weeks of treatment at either Nottingham or the Royal London Hospital. We provided travel expenses and where necessary overnight stay for those who needed to travel to reach one of these centres though few required this. We also hypothesised that, like alosetron, a closely related $5HT_3RA$, ondansetron would relax the rectum as shown by increased compliance in the rectal barostat studies which were performed in Nottingham and the Royal London Hospital. Previous studies had identified abnormalities of FPs and bile acids in IBS-D which correlated with transit, so we planned to assess their correlation with symptoms at both baseline and week 12.

We attempted to collect faecal samples at baseline and week 12 for all patients to assess faecal water %, bile acids and protease.

Transit

Aim
To assess if ondansetron altered colonic transit significantly.

Methods
Patients were asked to take the transit capsules daily for Transit-Pellets method™ supplied by Medifactia AB Limited, Stockholm, Sweden. Pellets were taken starting 6 days before their planned visit 3. Patients were instructed to take one pill at approximately 9 a.m. each day apart from day 6 when there were two pills to take, one at 9 a.m. and the second pill at around 6 p.m. This adaptation of the standard Metcalf method is designed to deal with patients with very fast transit when pellets taken at 9 a.m. may well have left the colon leaving zero pills to count. This then would raise the question of whether any pills had been taken whereas taking the second later pill makes this much less likely. On visit 3 a plain abdominal X-ray was taken and used to count the number of pellets in each of three regions designated right colon, left colon and pelvic area as defined on the plain abdominal X-ray by a vertical line to tip of the fifth lumber vertebra spinal process and then tangential to either pelvic brim as shown below. Colonic transit calculations from plain X-ray (*Figure 1*) were as described by Chan *et al.*[1] and used by Garsed *et al.*[2]

Regional transit was calculated from the number of pellets in the right, left and pelvic areas × 2.4 so in the example right transit time = 2 × 2.4 = 4.8, left = 2.4 × 3 = 7.2 and pelvic = 16 × 2.4 = 38.4 hours.

High-resolution manometry

Aim
Our aim was to characterise for the first time, the motor patterns in IBS-D using high-resolution manometry (HRM). We also planned to assess if baseline readings would relate to symptoms and whether these would alter with treatment.

This was performed at baseline and after 8–11 weeks of treatment in Nottingham and the Royal London Hospital. Patients from other sites were also offered the opportunity to travel to either site; two patients accepted this offer. The methods followed those reported by Dinning *et al.*[34] and subsequently used by Corsetti *et al.*[35,36]

Methods
After a 12-hour fasting period, all patients were admitted to the Motility Unit for bowel preparation with tap water enemas. They then underwent a colonoscopy-assisted positioning of the colonic HRM catheter (UniTip High-Resolution Catheter, Unisensor AG, Attikon, Switzerland). As the colonoscope was

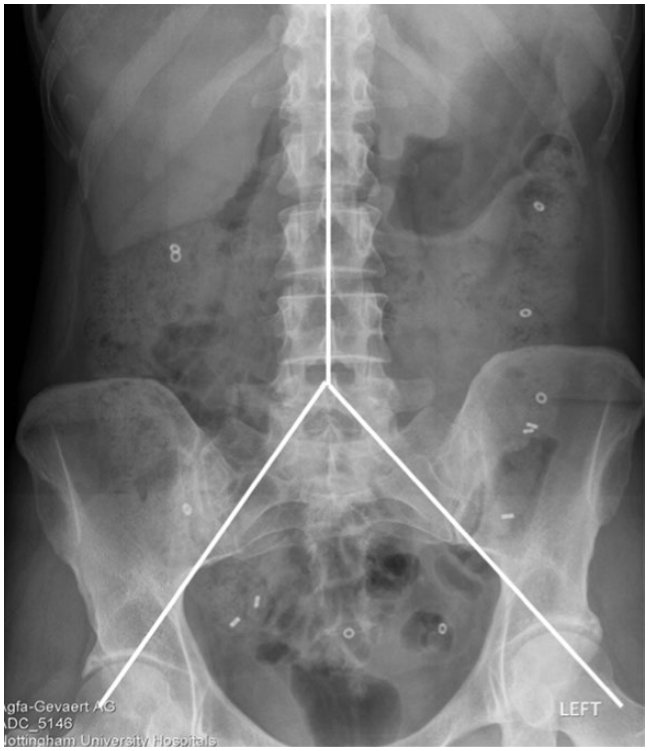


FIGURE 1 Colonic transit calculations from plain X-ray. Whole gut transit (WGTT) was calculated from total pellets ×2.4 hours, so in this example of a patient on ondansetron with 21 pellets, WGTT = 50.4 hours.

being withdrawn after having fixed the catheter with haemoclips, four biopsies were taken. The catheter consisted of 40 sensors spaced 2.5 cm apart. After allowing 1 hour for the recording to settle following the colonoscopy, colonic pressure recording was started and continued for 2 hours before consuming a standardised meal [Tomato & Basil Dolmio PastaVita (285 kcal, 3.9 g fat, 8.7 g sugars) and Ensure® TwoCal drink (399 kcal, 17.8 g fat, 9.2 g sugars)]. During the recording, the subjects were asked to score their feeling of abdominal gas, desire to evacuate gas, desire to defaecate and urgency to defaecate every 15 minutes. Both the investigator and patient were blind as to their treatment. The manometry files were sent via secure transfer to Dr Phil Dinning at the University of Adelaide for analysis using previously published methods.[21,37]

Barostat assessment

This was performed at baseline in nine subjects and after 8–11 weeks of treatment in Nottingham and the Royal London Hospital; six of whom also underwent manometry. Patients from other sites were also offered the opportunity to travel to either site but only two patients accepted this offer.

Aim

To assess if ondansetron increases rectal compliance and decreases sensitivity (manifest as increased pressure thresholds for pain and urgency) compared to placebo.

Methods

Patients were asked to fast from midnight for morning slots, or from 8 a.m. for afternoon slots prior to the assessment. They were asked to bring their usual dose of the study drug for the 8–11-week visit. After taking the dose, there was a 60-minute delay before making any measurements. On arrival patients were given instructions on how to self-administer a 500 ml body temperature tap water enema. After defaecating, patients were asked to lie down on the bed in a semi-prone left lateral decubitus position in a relaxed fashion with the foot end of the bed elevated from horizontal by 15° to decrease the effects of abdominal viscera on the bag volume. Rectal sensitivity was then assessed 60 minutes after taking the trial medication using a dual-drive barostat (Distender series II, G & J Electronics, Toronto, ON,

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Canada) and a polyethylene bag (600 ml) fixed on the end of a double-lumen barostat catheter (MUI Scientific, Mississauga, ON, Canada). The catheter was inserted into the rectum so that the middle of the bag was located approximately 10 cm from the anal verge and taped securely to the buttocks. The barostat bag was then unfolded by transiently inflating it with 75 ml of air and subsequently deflating it completely but leaving it in situ. Rectal pressure/volume relationships were assessed during a phasic isobaric, ascending method of limits distension protocol (4 mmHg steps to maximum toleration; 1 minute distension period, with 1 minute rest period between distensions). This was followed by a random phasic distension protocol distending to 8, 16, 24 and 36 mmHg with subjects rating sensation of gas, urgency, discomfort and pain on a 0–10 visual analogue scale (VAS). The maximum pressure used was adjusted depending on established pain threshold to avoid excessive pain and pressure was immediately released if patients reported >80 mm of discomfort or pain on the VAS, following which higher distensions were not administered. Once the series had been completed the bag was deflated and removed. All procedures followed the written standard operating procedure which was part of the protocol.

End points

Primary: Pain pressure threshold mmHg.

Secondary: Urgency volume threshold, urgency pressure threshold, pain volume threshold.

Stool biomarkers: faecal proteases and bile acids

Aim

Previous studies had identified abnormalities of FPs and bile acids in IBS-D which correlated with transit and urgency, so we planned to assess these along with transit to see if we could confirm these findings and see if these affected symptoms.

Methods

All patients were asked to provide stool samples to assess whether ondansetron reduces total FBA and tryptase concentrations, both active mediators which might correlate with urgency.

Faecal water was measured by simple drying of weighed faecal samples in a vacuum rota-evaporator (Jouan, RC10.22, Thermo Fisher Scientific, Waltham, MA, USA) at 400 °C until constant weight was achieved.

Proteases

Using previously reported methods, samples (reproduced under creativecommons.org/licenses/by-nc/3.0/)[23] (1 g) of stool stored at −70 °C were thawed and mixed in 5 ml 50 mM Tris buffer, 150 mM NaCl, pH7.2. Turbid suspensions were clarified using a sequential combination of centrifugation [10 minutes, 3000 relative centrifugal force (RCF)] and filtration (0.2 µm). Particulate-free supernatants were archived (−70 °C) until required for various assays or protein characterisation procedures.

Quantitation of total protein

Total protein quantitation was performed using the Bradford method,[38] modified for low volume and high throughput. Briefly, equal volumes of sample and reagent were mixed in 96-well microplates and, following 20 minutes incubation, absorbance measurements at 595 nm were used to quantitate by reference to bovine serum albumin calibrants.

Quantitation of total protease activity

Total FP quantitation was performed using the non-specific proteolysis of azo-casein. The endoproteolysis of this liberates azo-peptides into the supernatant which are quantitated by absorbance measurement at 440 nm subsequent to protein precipitation with trichloroacetic acid. Protease activity

was quantitated against bovine trypsin calibrant and expressed as trypsin units. Protein concentrations ranged from 0.08 to 1.7 mg/ml and measured protease activities were normalised against these values and expressed as units of trypsin/mg protein.

Quantification of bile acids (LC–MS)

After thawing faecal sample, 0.5 g was suspended in 2 ml of 50% (w/v) acetonitrile and extracted by vortexing and sonication for 10 minutes. The suspension was centrifuged twice at 25,000 g for 20 minutes. Supernatants were transferred to sample vials and loaded onto a liquid chromatography mass spectrometer (LC–MS) system containing online solid-phase extraction (SPE).

At the beginning of each analysis, 50 µl of the sample were transferred to SPE column at a flow of 0.1 ml/min with the loading mobile phase, aqueous 5% acetonitrile, 0.1% formic acid and 0.02% trifluoroacetic acid.

The chromatographic separation was performed using a binary system with pumps (A) and (B) (Jasco, PU-2085 Plus) connected to a degasser (Alltech, degasser, Stamford, UK). The two systems were connected using a two-position, six-port valve, used to switch automatically from loading (position 1) to injection (position 2) after 9 minutes (Valvemate®, Gilson, Dunstable. UK). Samples were analysed on a Waters Ex-bridge C18 Column (Waters, 100 × 2.1 mm; 3.5 µm particle size, Waters, Wexford, Ireland), using a gradient program. Mobile phase (A) 5 mM ammonium acetate, 0.1% ammonium hydroxide, mobile phase (B) 100% acetonitrile. Initial composition was 80% (A) was reduced to 70% (A) over 30 minutes the further reduction to 65% (A) over the next 3 minutes. The eluent composition was held at 65% for 1.5 minutes before returning to 80% (A) initial condition over the next 1.5 minutes. Flow rate 0.2 ml/min.

High-performance liquid chromatography was coupled in series with the turbo ion-spray (ESI) source of the tandem mass spectrometer (Micromass, Manchester, UK). Electrospray ionisation was performed in negative mode with nitrogen as the nebuliser gas. Detection of individual bile acids was performed using selective-ion monitoring (SIM) mode. Additional structural information was obtained via tandem MS (MS/MS) fragmentation, with collision energies ranging from 15 to 30 electron volts. Data were acquired using software MassLynX (Waters, Wexford, Ireland).

The concentration of bile acids in the samples was determined on the basis of the peak areas of individual bile acids and external standards.

End points

Primary: FP in trypsin units/mg protein, primary and secondary bile acids mmol/l stool water.

Secondary: Concentrations of individual bile acids, namely, cholic, chenodeoxycholic, deoxycholic and lithocholic acid, ratio of secondary/primary bile acids.

Mechanistic outcome measures

1. Whole gut transit time (WGTT; in hours), assessed using radio-opaque markers and an abdominal radiograph at baseline and 12 weeks.
2, Pain pressure threshold, pain volume threshold, urgency pressure threshold and urgency volume threshold, assessed using a barostat prior to starting the trial and 8–11 weeks during the trial.
3. FBAs and proteases prior to starting the trial and at 12 weeks during the trial.
4. Number of high amplitude propagated contractions (HAPCs) fasted and postprandially, assessed by HRM at baseline and at a visit at 8–11 weeks during the trial.
5. Percentage time occupied by cyclical propagated contractions, assessed by HRM at baseline and at a visit at 8–11 weeks during the trial.

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Sample size

Primary end point

Treatment of irritable bowel syndrome using titrated ondansetron trial (TRITON) planned to recruit 400 patients from up to 24 sites across England and Scotland. This would have provided 90% power at 5% significance to detect a 15% absolute difference between the randomised groups in the proportion of patients achieving the FDA-recommended end point[39] of a weekly responder for pain intensity and stool consistency for at least 6 weeks of the 12-week treatment period. This difference (15%) was considered to represent the minimum clinically important difference since in practice over the last two decades new IBS drugs with a lesser margin of efficacy are rarely prescribed in the NHS. We assumed a placebo response rate of 17%, as recently reported[12] using this end point and allowed for a 15% attrition rate.

Mechanistic studies

Whole gut transit

Our previous study using the same radio-opaque marker technique showed ondansetron increased WGTT by a mean [95% confidence interval (CI)] of 10 (6 to 14) hours.[12] Using 200 per group would have given us a 90% power to detect a change of 0.7 hours.

High-resolution left-sided colonic manometry

Previous studies with the closely related $5HT_3$RA alosetron showed an increase in motility index compared with placebo, with a mean [standard deviation (SD)] of 1.0 (1.2),[19] indicating we would have a power of 80% to detect a standardised effect size of 1 with 17 patients. We aimed for 20 patients on each treatment to allow for dropouts, that is, 40 each undergoing 2 studies, a total of 80 HRM studies.

Rectal compliance and sensitivity

Previous studies with alosetron showed an increase in compliance from 5.9 (SD 1.3) to 9.8 (SD 1.2) ml/mmHg in 22 patients.[40] We aimed to study 40 patients on each treatment to calculate correlations with symptoms, which typically require much larger numbers than just showing a change in mean values.

Randomisation

Randomisation was performed on a 1 : 1 basis to receive either ondansetron or placebo, and each patient was allocated three bottles of trial medication, each with a unique IMP kit code. Minimisation was used to ensure treatment groups were well balanced with respect to the minimisation factors of registering site and whether the patient had undergone mechanistic assessments.

Blinding

The trial was double-blind, neither the patient nor those responsible for their care and evaluation (treating team and research team) knew the allocation or coding of the treatment allocation. This was achieved by identical packaging and labelling of both the over-encapsulated ondansetron and matched placebo. Each bottle of ondansetron/placebo was identified by a unique kit code. Randomisation lists containing kit allocation were generated by the safety statistician at the Clinical Trials Research Unit (CTRU) and sent to the clinical supply company that produced the kits and the code break envelopes. Management of kit codes on the kit logistics application, which was linked to the 24-hour randomisation system, was conducted by the CTRU safety statistician in addition to maintaining the backup kit-code lists for each site.

Access to the code break envelopes was restricted to the safety statistician and designated safety team. Code breaks were permitted in emergency situations, where treatment allocation knowledge was

needed to optimise treatment of the patient. Unblinded interim reports provided to the Data Monitoring and Ethics Committee (DMEC) were provided by the CTRU safety statistician and the reports were securely password protected.

Statistical methods: general considerations

All hypothesis tests were two-sided and used a 5% significance level. Methods to handle missing data are described for each analysis. Analysis and reporting were in line with Consolidated Standards of Reporting Trials (CONSORT).[41] As TRITON was a double-blind trial, the trial statistician was blinded to treatment group allocation throughout the trial, until the database had been locked and downloaded for final analysis. Only the safety statistician, supervising trial statistician, backup safety statistician and authorised unblinded individuals at the CTRU had access to unblinded treatment group allocation prior to final analysis.

Frequency of analyses

Outcome data were analysed once only, at final analysis, although statistical monitoring of safety data was conducted throughout the trial and reported at agreed intervals to the DMEC. Final analysis took place 16 weeks post–last patient randomisation.

End-point analysis

All analyses were conducted on the ITT population, defined as all patients randomised, regardless of non-compliance with the intervention. A per-protocol (PP) analysis of the primary end point was carried out to indicate whether results were sensitive to the exclusion of patients who violated the protocol (e.g. those patients randomised but subsequently found to be ineligible). Primary and secondary analyses were blind to random allocation. Outcome measures were analysed by regression models appropriate to the data type. Such analyses adjusted for randomisation minimisation factors: site, completion of manometry assessment and barostat assessment, as well as baseline values where applicable, age and gender. Baseline characteristics were summarised by randomised group.

Primary analysis

The primary analysis compared the proportions of patients achieving the FDA-recommended end point between treatment groups at 12 weeks post randomisation using a logistic regression model adjusted for minimisation factors, age and gender. The plan was that any missing data were assumed missing at random (MAR) and imputed for the primary analysis. However, complete case analysis was undertaken and those without sufficient data (n = 4) to evaluate the primary end-point were assumed to be non-responders. The potential impact of missing data would be small with only 5% of missing responses. Odds ratios and corresponding 95% CIs are presented.

Sensitivity analysis was planned to assess the impact of missing data on the treatment effect was performed. This was planned to include complete case analysis and alternatives to multiple imputation (e.g. pattern mixture modelling), if missing patterns suggested data were missing not at random. However, given the small overall sample size and only four participants with missing data (5%), this was not undertaken.

Secondary analyses

The proportions of patients with satisfactory relief of IBS symptoms at 12 weeks post randomisation was compared between the treatment groups using logistic regression models, adjusting for minimisation, baseline values, age and gender. Odds ratios and corresponding 95% CIs are presented.

Differences between the treatment groups for the continuous secondary end points at 12 weeks post randomisation were compared using linear regression models, adjusted for the minimisation variables, baseline where applicable, age and gender. These end points were urgency of defaecation over the last month, stool frequency over the last month, number of days per week with at least one loose stool (BSFS > 5) over the last month, average stool consistency, number of days rescue medication used over

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12 weeks, abdominal pain score, HADS depression and anxiety scores, SFLDQ score, IBS-QOL score and subscales, PHQ-12 and IBS-SSS severity scores. Treatment estimates and corresponding 95% CIs are reported.

The differences between the treatment groups post treatment over weeks 13–16 post randomisation in the following end points: stool frequency, abdominal pain and urgency of defaecation were compared using linear regression model adjusting for minimisation factors, baseline values and relevant baseline factors. Treatment estimates and corresponding 95% CIs are reported. Any missing data are assumed MAR.

Exploratory analyses on the daily measurements (worst abdominal pain, loose stools, number of stools passed, consistency of stool, worst urgency and use of loperamide) were carried out, using repeated measures models, which incorporate correlation between measurements from the same patient.

SAS software version 9.4 was used in the analyses of primary and secondary end points.

Safety analyses

All patients who receive at least one dose of trial treatment were included in the safety analysis set. The number of patients reporting a SAE (up to 28 days after the last dose of treatment), and details of all SAEs, are reported for each treatment group. The number of patients withdrawing from trial treatment is summarised by treatment arm, along with reasons for withdrawal. All safety analyses performed prior to final analysis were undertaken by the safety statistician (rather than the trial statistician), thus ensuring that the trial team remain blinded.

Subgroup analyses

No subgroup analyses were planned.

Mechanistic studies

Mechanistic studies were analysed by the site research fellow, blinded to the intervention allocation under supervision of the chief investigator and local supervising PIs. The differences between treatment groups for changes in whole gut transit times, colonic motility measures (% time of cyclical retrograde contractions and HAPC frequency), rectal compliance and thresholds for urgency and pain measured using the barostat, FBA concentrations and faecal tryptase were assessed using two-way analysis of variance (ANOVA) for analysis of treatment and time effects and *t*-test, Mann–Whitney *U*-test and Wilcoxon matched pairs test for paired comparisons between normally and non-normally distributed variables, respectively. In addition, exploratory mediator analyses were planned to explore whether treatment effects, in terms of changes in urgency or pain, are mediated through changes in FBAs or protease using Spearman/Pearson correlation as appropriate.

Patient and public involvement

Both the grant application and the design of the study were assisted by our patient participation and involvement (PPI) group, which included patients with IBS-D who supported the original application and subsequently helped with the design of patient-facing documents.

Chapter 3 Clinical trial results

Site and patient recruitment

Recruitment strategies

A number of strategies were adopted during the trial to address the recruitment difficulties that were encountered. The initial plan had been to open 11 secondary care centres for recruitment, but it soon became clear that this would not be adequate to achieve the necessary numbers required to complete the trial. The source of patients varied widely by site (see *Appendix 3*, *Table 37*). As well as increasing the number of secondary care sites to 16, we utilised numerous other strategies to attract more patients to the trial.

We opened a number of secondary PICs. The secondary care PICs would identify potential participants from their clinics and refer them on to a recruiting centre for consideration for the trial. A total of six such PICs were opened.

A key issue in recruitment of patients within secondary care seemed to be changes in referral patterns over recent years. A trial in Yorkshire showed the value of screening patients with diarrhoea with calprotectin in primary care, reducing unnecessary colonoscopy and this has strengthened existing National Institute for Health and Care Excellence (NICE) guidance[42] on the use of calprotectin in primary care. Evidence from NICE suggests a substantial reduction in referral from primary care with recent onset diarrhoea (56% in Cannock Chase, Staffordshire, UK).[43] Patients are thus less likely to be referred into secondary care for investigation of diarrhoea but are being investigated and managed in primary care. We soon realised that we would need to widen our recruitment pool to include patients in primary care and also in the general population.

We set up a self-referral pathway to allow patients to refer themselves to a local recruitment centre to be considered for the trial. Advertising was used to increase awareness of the trial and interested patients could visit the TRITON trial website for further information. The website included a simple self-screening questionnaire to confirm whether the patient was broadly eligible for the trial (see *Appendix 10*, *Figure 31*). Potentially eligible patients could then contact one of the trial research fellows to obtain further information about the trial and discuss their suitability before being referred on to their local recruiting centre.

We used a number of strategies to reach patients outside the secondary care centres. We approached GP practices in the areas surrounding the recruiting centres to set them up as PICs. The practices were asked to perform a search of their patient database to identify any suitable patients for the trial. They would then mail out a letter and leaflet introducing the trial to the patient asking them to visit the trial website if they were interested. Between March 2019 and February 2020, a total of 26 GP practices were set up as PICs and mail outs were sent to a total of 1381 patients. Of all the patients screened, two were identified in this way and of these one was randomised into the trial.

Starting in October 2019, we advertised within GP practices and pharmacies using posters and leaflets. GP practices that agreed to participate as PICs were asked to display the promotional materials in their waiting rooms. We also approached the Clinical Research Networks to arrange for promotional materials (see *Appendix 10, Figures 32* and *33*) to be displayed in the waiting rooms of GP practices not involved as PICs, as well as pharmacies.

In addition, we instigated a publicity drive to increase awareness of the trial outside of clinical settings. To achieve this, we advertised in local press in the areas surrounding our recruiting centres. The first advertisement drive was in May 2019 and regular advertising continued until March 2020. This advert

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(see *Appendix 10*, *Figure 34*) consisted of a quarter page in colour in publications such as the Metro, Manchester Evening News and London Evening Standard.

In November 2019, a Communications Officer was appointed to assist with the advertising strategy for the study, being responsible for engaging with Twitter (Twitter, Inc., San Francisco, CA, USA), TV, newspaper and radio, IBS charities and other patient forums to publicise the study. The trial Twitter account was used to engage with sites and improve awareness within trial teams in the hope that it would help attract more patients to the study. We actively encouraged staff at recruiting centres and collaborators to follow and retweet as well as using appropriate hashtags and tagging any relevant organisations in our tweets. We also advertised on social media, in particular Facebook (Meta Platforms, Inc., Menlo Park, CA, USA) and Google (Google Inc., Mountain View, CA, USA).

Professor Spiller and Karen Andrews (patient and public involvement representative) did an interview for the Mail on Sunday, which was the lead article on their health page on Sunday, 23 March 2019 and produced 3000 visits to our webpage and some 430 contacts of which approximately 10 entered the trial.

ContactME-IBS is a registry run through one of the TRITON recruiting sites, County Durham and Darlington NHS Foundation Trust. ContactME-IBS holds the contact information of approximately 2766 adults interested in hearing about, and taking part in, IBS research, of which 641 were suitable for TRITON having been identified as having IBS-D and associated pain (a key eligibility criterion). A mail shot to the 641 potentially eligible registry patients was sent in November 2019. Due to the nature of the registry setup, patients are mainly centred around areas where ContactME have had a drive to recruit GP practices, with 593 of the potentially eligible registry patients located in areas affiliated with TRITON secondary care sites, and 48 in areas without a suitable TRITON site.

Summary of patient identification method and screening approach is in *Table 1*.

Screening and recruitment

A total of 1582 potential participants were screened for entry into TRITON, across 13 hospital sites, including those referred to local sites by the Clinical Research Fellows (section Self-referrals below). Of these, 295 (18.6%) were found to be eligible for the study, 173 (58.6% of eligible) consented to take part in the study, 149 (86.1% of consented) were registered to the study, and 80 (53.7% of registered) were randomised to the study. Eligibility varied widely between centres from 10% to 90% with some being more efficient than others in deciding who to approach (see *Appendix 3*, *Table 36*). A detailed flow of these patients in the form of a CONSORT diagram can be seen in the supplementary document. A large proportion, 55.8% (882/1582), of screened patients were ineligible due to not meeting the Rome IV criteria. Of those who passed screening and were registered, insufficient pain scores were the largest single cause of being ineligible for randomisation (31/149). Overall, patients were screened over 28 months from March 2018 until June 2020.

Self-referrals

A total of 620 people self-referred via the TRITON website (*Figure 2*) and were sent information sheets. Subsequently, 297 (47.9%) did not respond further. A total of 155 (25.0%) were referred onto a local site, having confirmed their interest in taking part in the TRITON study and fulfilled the initial eligibility checks.

The trial Clinical Research Fellows responded to each patient's e-mail, and 64 patients were screened through the formal screening process. Twelve patients were randomised using the self-referral route (see *Table 1*). Self-referrals were received and screened over 20 months from November 2018 until June 2020.

Screening characteristics

Characteristics of screened participants (age and gender) summarised by site are detailed in *Table 2*. One site in particular screened more potential participants than the others, 49.2% (778/1582) of the total; but most of these screening forms were missing age and gender data. The percentage who were eligible

TABLE 1 Identification method and screening approach

Identification method	Screened (*n* = 1582)[a] (%)	Clinic 41 (2.6%) (%)	Telephone call 20 (1.3%) (%)	E-mail/letter 15 (0.9%) (%)	Missing 1506 (95.2%)[b] (%)	Randomised[a] (*n* = 80) (%)	Clinic 17 (21.3%)	Telephone call 3 (3.8%) (%)	E-mail/ letter 3 (3.8%) (%)	Missing[b] 57 (71.3%)
Secondary care	485 (30.7)	41 (8.5)	18 (3.7)	11 (2.3)	415 (85.6)	58 (72.5)	17 (29.3%)	1 (1.7)	1 (1.7)	39 (67.2%)
Primary care and pharmacies	10 (0.6)	0 (0.0)	1 (10.0)	1 (10.0)	8 (80.0)	4 (5.0)	0 (0.0%)	1 (25.0)	0 (0.0)	3 (75.0%)
Self-referral via website	64 (4.0)	0 (0.0)	1 (1.6)	2 (3.1)	61 (95.3)	12 (15.0)	0 (0.0%)	1 (8.3)	2 (16.7)	9 (75.0%)
Self-referral – other means	34 (2.1)				34 (100.0)	4 (5.0)	0 (0.0%)	0 (0.0)	0 (0.0)	4 (100.0%)
Secondary PIC	4 (0.3)				4 (100.0)					
Primary care (invitation letter)	2 (0.1)				2 (100.0)	1 (1.3)	0 (0.0%)	0 (0.0)	0 (0.0)	1 (100.0%)
Patients on file	951 (60.1)				951 (60.1)					
Forwarded by CTRU	2 (0.1)				2 (0.1)					
Identified by bowel cancer charity	1 (0.1)				1 (0.1)					
Other	1 (0.1)				1 (0.1)					
Missing	28 (1.8)	0 (0.0)	0 (0.0)	1 (3.6)	27 (96.4)	1 (1.3)	0 (0.0%)	0 (0.0)	0 (0.0)	1 (100.0%)

a Column %, the remaining table is row %.
b Missing – information about the screening approach is not available.

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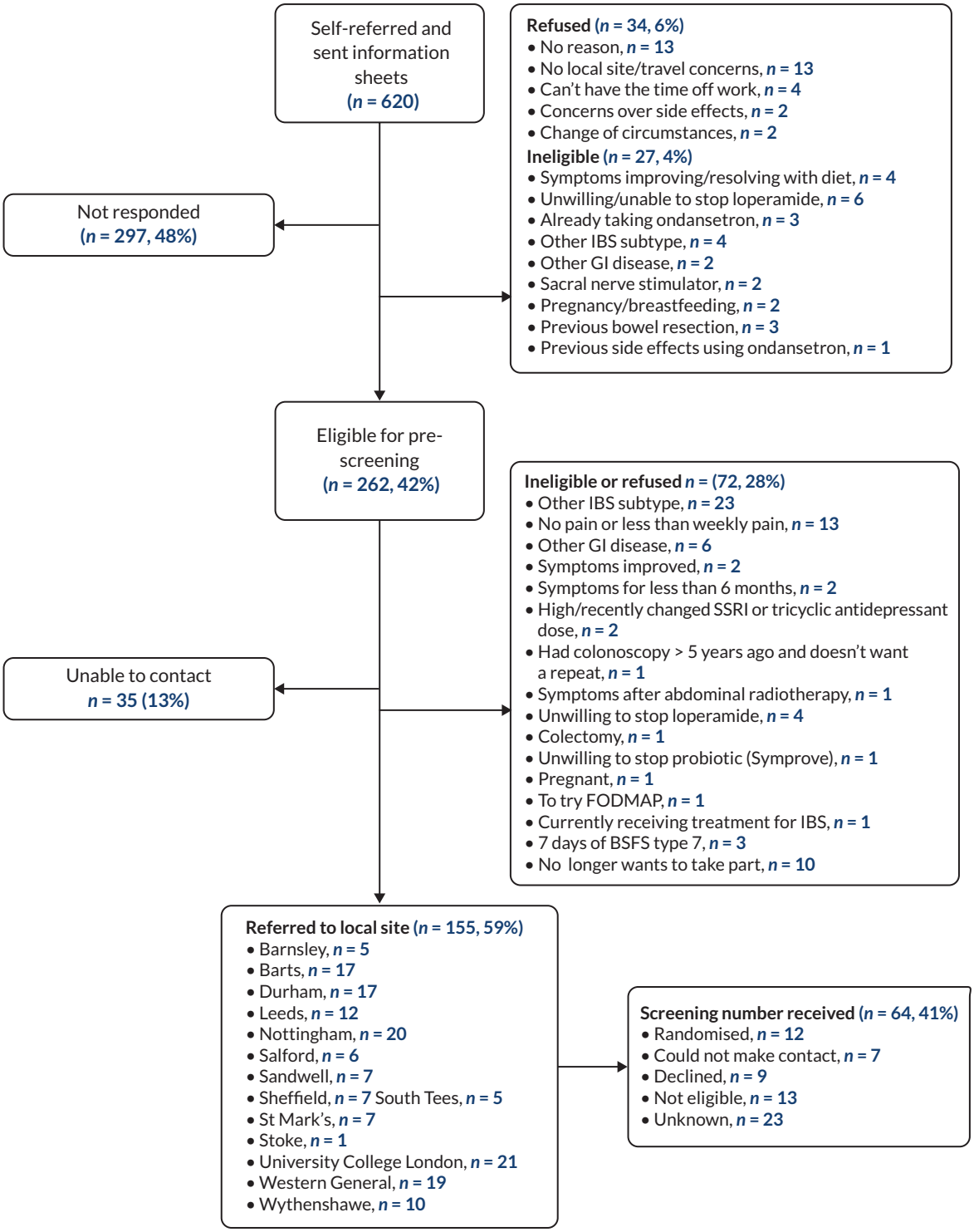


FIGURE 2 Self-referral flow of participants.

varied from 0.6% to 91.7% of those considered indicating that some sites were much more selective in who they approached. For those potential participants with screening data available, the mean age of those screened was 44.9 years old (SD 15.4), and 67.1% were female (523/780). Further screening information by site can be found in *Appendix 3*, *Tables 36*–*38*.

Randomisation and participant flow

Registration data by site are provided in *Appendix 3*, *Table 39*. Participants were randomised over 23 months from July 2018 until May 2020 (*Figure 3*). Thirty-seven (46.3%) participants were randomised

TABLE 2 Patient screening characteristics by site

Site	Total (%)	Age (years)		Gender		
		Mean (SD)	Missing	Male (%)	Female (%)	Missing (%)
James Cook, South Tees	38 (2.4)	39.6 (12.5)	0	13 (34.2%)	25 (65.8%)	0 (0.0%)
St James's University Hospital, Leeds	80 (5.1)	43.5 (17.4)	6	16 (20.0%)	63 (78.8%)	1 (1.3%)
Royal Hallamshire Hospital	51 (3.2)	48.1 (13.9)	1	23 (45.1%)	27 (52.9%)	1 (2.0%)
Sandwell Hospital	43 (2.7)	45.2 (12.5)	1	15 (34.9%)	28 (65.1%)	0 (0.0%)
University Hospital of North Durham	23 (1.5)	49.0 (11.8)	4	6 (26.1%)	16 (69.6%)	1 (4.3%)
Wythenshawe Hospital	12 (0.8)	47.4 (15.7)	0	5 (41.7%)	7 (58.3%)	0 (0.0%)
Royal Stoke University Hospital	44 (2.8)	43.6 (17.1)	0	14 (31.8%)	28 (63.6%)	2 (4.5%)
Nottingham University Hospital	58 (3.7)	45.7 (18.2)	4	25 (43.1%)	32 (55.2%)	1 (1.7%)
Barnsley District General Hospital	216 (13.7)	46.8 (15.5)	5	54 (25.0%)	161 (74.5%)	1 (0.5%)
St Mark's Hospital	11 (0.7)	56.3 (12.1)	2	6 (54.5%)	5 (45.5%)	0 (0.0%)
Western General Hospital	53 (3.4)	43.8 (14.6)	1	12 (22.6%)	40 (75.5%)	1 (1.9%)
Salford Royal Hospital	16 (1.0)	38.6 (15.5)	0	6 (37.5%)	10 (62.5%)	0 (0.0%)
Barts and London School of Medicine and Dentistry	159 (10.1)	43.0 (14.3)	67	43 (27.0%)	53 (33.3%)	63 (39.6%)
University College London Hospital	778 (49.2)	42.1 (16.1)	740	19 (2.4%)	28 (3.6%)	731 (94.0%)
Total	**1582 (100)**	**44.9 (15.4)**	**831**	**257 (32.9% of non-missing) (16.2% of total)**	**523 (67.1% of non-missing) (33.1% of total)**	**802 (50.7%)**

to ondansetron and 43 (53.8%) to placebo (see CONSORT diagram). By the end of treatment at 12 weeks, six participants discontinued treatment, four in the ondansetron arm (one of these patients withdrew from follow-up) and two in the placebo arm.

Populations

Populations are summarised in *Table 3*.

Intention-to-treat (ITT) population. All 80 randomised participants are included in the ITT population.

Per-protocol (PP) population. Four patients of the 80 randomised were removed from the PP population due to major protocol violations. One participant was taking a restricted medication (Buscopan), two participants' abdominal pain scores in the pre-treatment diary were too low compared with the inclusion criteria, and one participant became pregnant while in the trial.

Safety population. All 80 participants received at least 1 dose of trial medication, and there were no cases of incorrect treatment taken, so all 80 randomised participants are included in the safety population in accordance with randomised allocation.

Baseline

At baseline, randomised participants were of a similar age to all screened participants, mean age 43.9 and 44.9 years, respectively. Overall, a higher proportion of males were randomised compared with the

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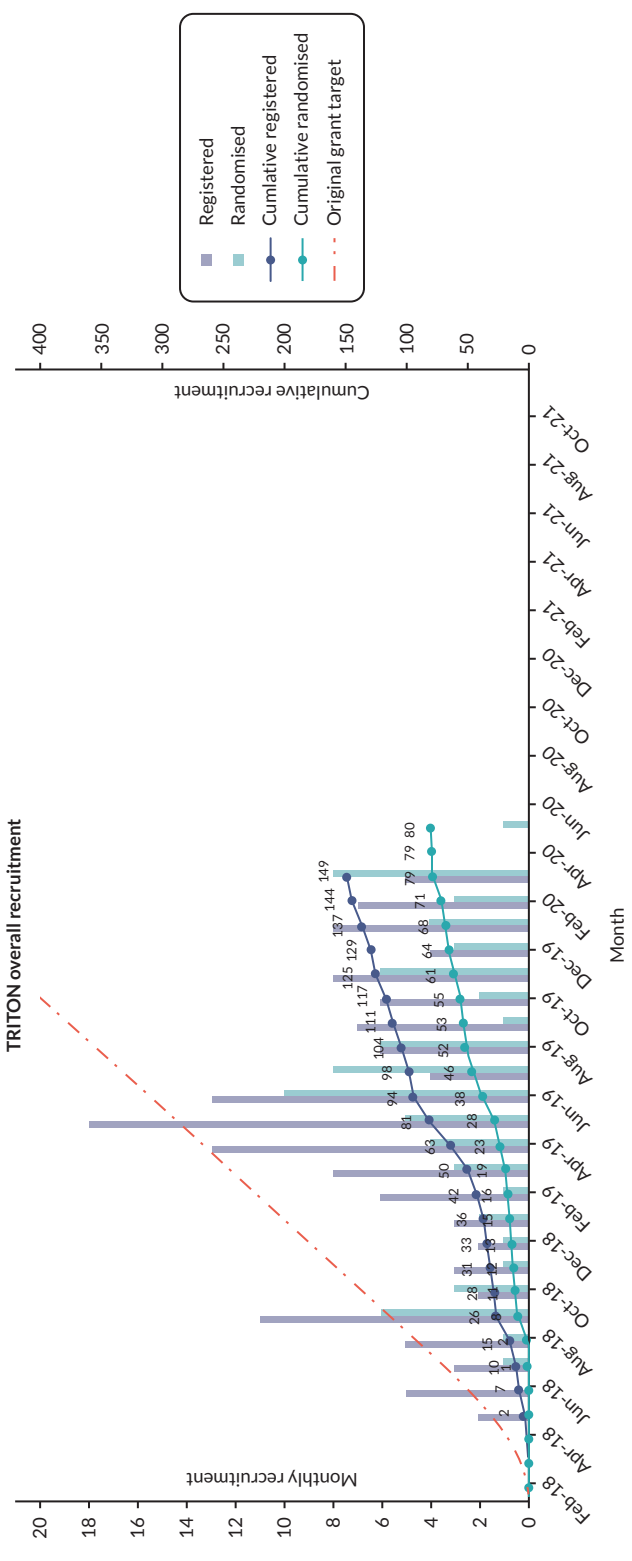


FIGURE 3 Overall recruitment.

TABLE 3 Study population by treatment arm

Population	Randomised to		
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
ITT	37	43	80
PP	36	40	76
Safety	37	43	80

TABLE 4 Participant demographic information by treatment arm

Demographics	Randomised			Screened	
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Screened (*n* = 1582)	Missing
Mean **age** (years) (SD)	45.0 (15.7)	43.0 (16.3)	43.9 (16.0)	44.9 (15.4)	831[a]
Sex, *N* (%)					802[a]
Male	16 (43.2%)	17 (39.5%)	33 (41.3%)	257 (32.9%)[b]	
Female	21 (56.8%)	26 (60.5%)	47 (58.8%)	523 (67.1%)[b]	
Ethnicity, *N* (%)					
White	34 (91.9%)	41 (95.3%)	75 (93.8%)	–	
Black	2 (5.4%)	1 (2.3%)	3 (3.8%)	–	
Asian	1 (2.7%)	1 (2.3%)	2 (2.5%)	–	
Current smoker, *N* (%)					
Missing	4 (10.8%) 1	5 (11.6%) 0	9 (11.3%) 1		
Number of years smoked/ number of cigarettes/day listing	(8/12, 10/15, 12/7, 38/9)	(5/10, 10/5, 12/7, 18/7, 37/6)			

a 740 age and 731 sex missing from one site.
b Out of non-missing records.

proportion of males screened; 41.3% and 32.9%, respectively. However, a high proportion of screening records had no age and sex data making comparisons uncertain (*Table 4*). In both arms, males and females reported similar non-gastrointestinal somatic symptoms associated with IBS measured by PHQ-12 (*Table 5*).

During the pre-treatment period, participants randomised subsequently to ondansetron reported, on average, higher abdominal pain scores (61.4 vs. 55.2 mean pain score), slightly more days per week with a loose stool (5.9 vs. 5.4 mean days per week) and a higher urgency to defaecate score (67.5 vs. 60.4 mean urgency score) than those randomised to placebo (see *Table 5*). Pre-treatment diary scores by site are provided in *Appendix 3*, *Table 40*.

Table 4 shows that the participants were well matched for demographics across randomised treatment arms. Demographic data by site are provided in *Appendix 3*, *Table 41*.

As part of the trials mechanistic substudy, participants were offered a barostat procedure and a colonic manometry. Eighteen participants underwent a barostat and 13 a colonic manometry assessment. Of these, 12 undertook both manometry assessments (see *Table 5*). Mechanistic test uptake by site is provided in *Appendix 3*, *Table 42*.

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Further baseline summaries including summaries across time points are in *Summaries and analyses of the secondary outcomes*.

Losses and exclusions after randomisation

Treatment discontinuation and withdrawals

Five participants discontinued treatment, four in the ondansetron arm (two were requested by the patient and two by the clinician) and one in the placebo arm (requested by the patient) (see *Table 6* and

TABLE 5 Pre-treatment diary scores, uptake of mechanistic substudy and PHQ-12 scores by treatment arm

Pre-treatment diary	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Mean pain score (SD)	61.4 (19.7)	55.2 (16.7)	58.0 (18.3)
Mean days per week with loose stool (SD)	5.9 (1.3)	5.4 (1.2)	5.6 (1.3)
Mean urgency score (SD)	67.5 (19.6)	60.4 (17.8)	63.8 (18.9)
Urgency score missing	0	1	1
Mechanistic test undertaken			
Barostat, *N* (%)	8 (21.6%)	10 (23.3%)	18 (22.5%)
Colonic manometry, *N* (%)	7 (18.9%)	6 (14.0%)	13 (16.3%)
Both mechanistic tests undertaken	6 (16.2%)	6 (14.0%)	12 (15.0%)
PHQ-12 score			
Male, *N*	16	17	33
Mean (SD) missing	7.5 (4.63) 0	7.5 (3.54) 0	7.5 (4.04) 0
Female, *N*	21	26	47
Mean (SD) missing	10.3 (4.32) 0	9.6 (4.63) 0	9.9 (4.46) 0

Notes
Pain and urgency scores: range 0–100.
PHQ-12 score: range 0–22 in men, 0–24 in women, high score indicates multiple somatic symptoms.

TABLE 6 Treatment discontinuation

	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
Number of discontinued treatment (% of randomised)	4 (10.8)	1 (2.3)	5 (6.2)
Requested by patient	2 (50.0)	1 (100.0)	3 (60)
Requested by clinician	2 (50.0)	0 (0.0)	2 (40)
Reason for discontinuation			
Patient decision	1 (25.0)	0 (0.0)	1 (16.7)
Patient's health compromised	1 (25.0)	0 (0.0)	1 (16.7)
Drug intolerance	1 (25.0)	1 (50.0)	2 (33.3)
Other[a]	1 (25.0)	0 (0.0)	1 (16.7)

a Patient not eligible (unable to discontinue Buscopan), withdrawn by clinician.

TABLE 7 Withdrawal of consent for data collection

Withdrawal	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
Number of withdrawn from any part of the study (% of randomised)	10 (27.0)	7 (16.3)	17 (21.3)
Number of withdrawn consent for			
Daily text messaging (% of withdrawals)	10 (100.0)	7 (100.0)	17 (100.0)
Use of previously obtained samples (% of withdrawals)	2 (20.0)	0 (0.0)	2 (11.8)
Optional main study assessments (% of withdrawals)	2 (20.0)	1 (14.3)	3 (17.6)
Trial follow-up (% of withdrawals)	3 (30.0)	0 (0.0)	3 (17.6)

Appendix 4, *Table 43*). One participant in the placebo arm withdrew due to pregnancy (see *Appendix 8*, *Table 65*).

Seventeen participants, 10 in the ondansetron and 7 in the placebo arm, withdrew from one or more parts of the study data collection processes. All 17 withdrew consent for daily text messaging, and 3 participants in the ondansetron arm also withdrew from trial follow-up (*Table 7*).

Protocol violations and deviations

There were four major protocol violations: one participant took a restricted medication (hyoscine), two had abdominal pain scores in the pre-treatment diary too low to meet the inclusion criteria and one became pregnant during the trial. All four were removed from the PP population.

There were 12 participants that deviated from the protocol in terms of taking more than 2 loperamide doses a week in the treatment period, 5 in the ondansetron arm and 7 in the placebo arm (*Table 8*). Four of those five participants in the ondansetron arm deviated from the protocol during the pre-treatment, treatment and post-treatment periods. Two of the participants in the placebo arm deviated from the protocol in the pre-treatment, treatment and post-treatment periods.

These participants were included in the ITT analysis and additional analysis was undertaken adding the use of loperamide as an independent variable in the primary analysis model, section *Supportive and sensitivity analyses*.

Clinical efficacy of the intervention

Analyses of the primary outcome

Analyses were conducted on the ITT population and all 80 participants were included in the primary analysis. There were 15/37 [40.5%, 95% CI (24.7%, 56.4%)] primary end-point responders in the ondansetron arm and 12/43 [27.9%, 95% CI (14.5% to 41.3%)] in the placebo arm. Four participants (two in each arm) did not provide sufficient data to calculate the primary end point and were therefore assumed to be non-responders. Multiple imputation was not used, as there were only 5% of missing responses the potential impact of the missing data would be small[44] and the overall sample size was small.

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TABLE 8 Participants taking more than two loperamide doses a week

	Ondansetron		
Unique participant	**Pre-treatment**	**Treatment (weeks 1–12)**	**Follow-up (weeks 13–16)**
1	1		
2	2	3	4
3	3	1	1
4			3
5		12	
6	1	2	1
7	2		3
8	2	8	2
	Placebo		
1			1
2	1	2	1
3	1		
4			1
5		5	3
6		1	
7	2	6	2
8	1		
9		5	
10	1		
11		1	
12		1	
13	2		2

Considering the individual components of the primary end point; there were 17/37 [46.0%, 95% CI (29.9% to 62.0%)] on ondansetron who were pain responders (i.e. met FDA criteria for reduction in pain intensity) and 16/43 [37.2%, 95% CI (22.8% to 51.7%)] on placebo. As regards stool consistency, 25/37 [67.6%, 95% CI (52.5% to 82.7%)] on ondansetron and 22/43 [51.2%, 95% CI (36.2% to 66.1%)] on placebo were stool consistency responders according to FDA criteria (*Tables 9* and *10*). There was no statistical evidence of a difference in the FDA-defined primary end-point responder rate between arms. The odds ratio from the logistic regression model, adjusted for the treatment group, minimisation variables (undergoing manometry or barostat), age and gender was 1.93 (95% CI 0.73 to 5.11, *p*-value 0.1869). Site was excluded from the model due to model convergence issues (see *Table 10*). Additionally, when assessing individual components of the primary end point, ondansetron had greater effect on stool consistency improvement than on pain intensity reduction when comparing between arms. However, none of these differences were statistically significant.

TABLE 9 Primary end-point data summary

Primary end point	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Number of primary end-point responders, *N* (%)	15 (40.5%)	12 (27.9%)	
Number of **non-responders**, *N* (%)	20 (54.1%)	29 (67.4%)	
Number of participants **with insufficient data** to evaluate primary end point, *N* (%) *(treated as non-responders in ITT)*	2 (5.4%)	2 (4.7%)	4 (5.0%)
Pain intensity reduction responders, *N* (%)	17 (46.0%)	16 (37.2%)	
Stool consistency reduction responders, *N* (%)	25 (67.6%)	22 (51.2%)	
Number of available data weeks			
Mean (SD)	11.2 (2.1)	11.3 (2.0)	11.3 (2.0)
Median (range)	12.0 (4, 12)	12.0 (0, 12)	12.0 (0, 12)
Number of weeks as a responder			
Mean (SD)	4.3 (3.9)	3.4 (3.9)	3.8 (3.9)
Median (range)	3.0 (0, 12)	2.0 (0, 12)	3.0 (0, 12)
Number of weeks as a non-responder			
Mean (SD)	7.0 (3.9)	7.9 (4.0)	7.5 (4.0)
Median (range)	7.0 (0, 12)	8.0 (0, 12)	8.0 (0, 12)

Supportive and sensitivity analyses

Analysis of the primary outcome on the PP population supported the primary analysis result that there is no evidence of a difference in the treatment arms (see *Table 10*). Throughout the study (pre-treatment, treatment and follow-up), participants were allowed to use rescue medication (loperamide) no more than twice per week. Similar proportions of participants used loperamide during pre-treatment and follow-up (off-treatment) across arms, but a higher proportion used loperamide in the placebo arm during treatment; 39.5% (*n* = 17) compared with 18.9% (*n* = 7) in ondansetron (see *Appendix 6*, *Table 48*). Therefore, as part of the sensitivity analyses, use of rescue medication was added as an independent variable and the primary analysis model was repeated. The outcome of this analysis is consistent with the primary analysis finding that there is no difference in the primary end point of FDA-defined responder rates between the treatment arms. Additionally, loperamide use was added as an interaction term with the treatment allocation and the findings were again similar. The number of responders using rescue medication is summarised in *Appendix 6*, *Table 49*; higher proportions of both responders and non-responders in the placebo arm used loperamide. Sensitivity analysis including HADS anxiety and depression scores as independent variables in the primary end-point model supports the conclusion of no evidence of a statistical difference between the treatment arms (see *Table 10*).

Summaries of overall responders and responders to individual components of primary outcome by week

A graphical summary of responders by week is provided in *Figures 4–6*. Weeks 1–12 are the treatment weeks and weeks 13–16 are the post-treatment follow-up weeks. Based on those summaries, participant response increased during the weeks in treatment in both treatment arms and decreased in the follow-up period. The largest difference between treatment arms is in stool consistency; a higher proportion of participants in the ondansetron arm were responders compared with those in the placebo arm (see *Figure 6*). This effect was apparent already in the first week (see *Appendix 7*, *Table 54*).

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TABLE 10 Primary end-point analysis and sensitivity analyses

Analysis	Responders				Model output	
	Ondansetron (*n* = 37)		Placebo (*n* = 43)			
	N (%)	(95% CI)	*N* (%)	(95% CI)	*p*-value	OR (95% CI)
1. **Primary end point**	**15 (40.5)**	**(24.7 to 56.4)**	**12 (27.9)**	**(14.5 to 41.3)**	**0.1869**	**1.93 (0.73 to 5.11)**
a. **PP**					0.2493	1.78 (0.67 to 4.76)
b. **Sensitivity** + loperamide use					0.3076	1.68 (0.62 to 4.57)
c. +interaction (treatment arm * loperamide use)					0.3598[a]	1.71 (0.54 to 5.40)
d. +HADS anxiety score					0.2026	1.96 (0.70 to 5.53)
e. +HADS depression score					0.0942	2.50 (0.86 to 7.31)
2. **Pain intensity reduction**	17 (46.0)	(29.9 to 62.0)	16 (37.2)	(22.8 to 51.7)	0.3222	1.61 (0.63 to 4.12)
a. **PP**					0.4229	1.47 (0.57 to 3.81)
b. **Sensitivity +** loperamide use					0.4976	1.40 (0.53 to 3.67)
c. + interaction (treatment arm * loperamide use)					0.3496[a]	1.72 (0.55 to 5.35)
3. **Stool consistency reduction**	25 (67.6)	(52.5 to 82.7)	22 (51.2)	(36.2 to 66.1)	0.0730	2.45 (0.92 to 6.52)
a. **PP**					0.1042	2.28 (0.84 to 6.16)
b. **Sensitivity** + loperamide use					0.0647	2.60 (0.94 to 7.15)
c. + interaction (treatment arm[a] loperamide use)					0.0595[a]	3.13 (0.96 to 10.26)

OR, odds ratio.
a Estimates presented from the model with the interaction terms are for ondansetron vs. placebo without loperamide use, as majority of participants did not use loperamide.

Note
Placebo is reference category.

The number and proportion of participants using loperamide as a rescue medication by week are summarised in *Figures 7* and *8*. Additionally, the proportion of participants using loperamide was added to the figures to complement the sensitivity analyses. Tables summarising number and proportion of responders by week are in *Appendix 7*, *Tables 52–56*.

Summaries and analyses of the secondary outcomes

Abdominal pain

Mean abdominal pain scores were higher in the ondansetron arm prior to randomisation compared with placebo (see *Table 5*), but were similar across the treatment arms during the treatment and showed a similar increase in the follow-up period (*Table 11*), indicating a greater reduction in pain scores in the

ondansetron arm during treatment (see *Appendix 6, Tables 44* and *53*). However, there was no evidence of a statistically significant difference in abdominal pain scores between the treatment arms during or after the treatment periods (*Tables 12–14*).

Urgency

Mean urgency of defaecation at baseline was higher in the ondansetron arm compared with placebo (see *Table 5*). During the treatment period and in the follow-up, unadjusted urgency scores were slightly higher in the placebo arm compared with ondansetron, having reduced more in the ondansetron arm

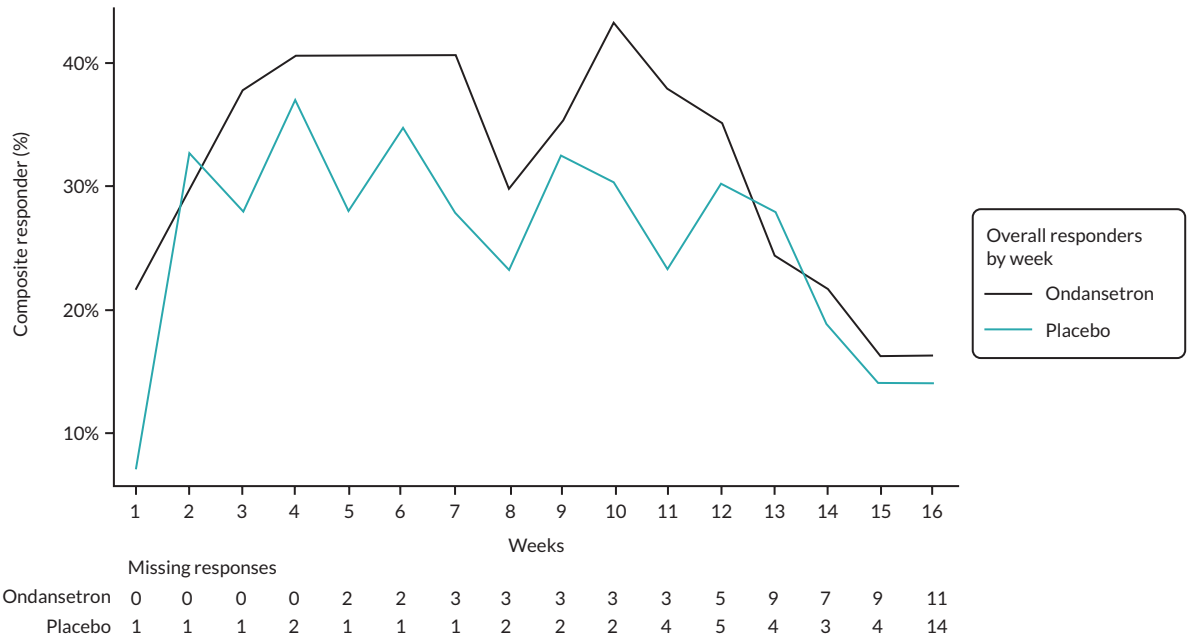


FIGURE 4 Overall responders by week.

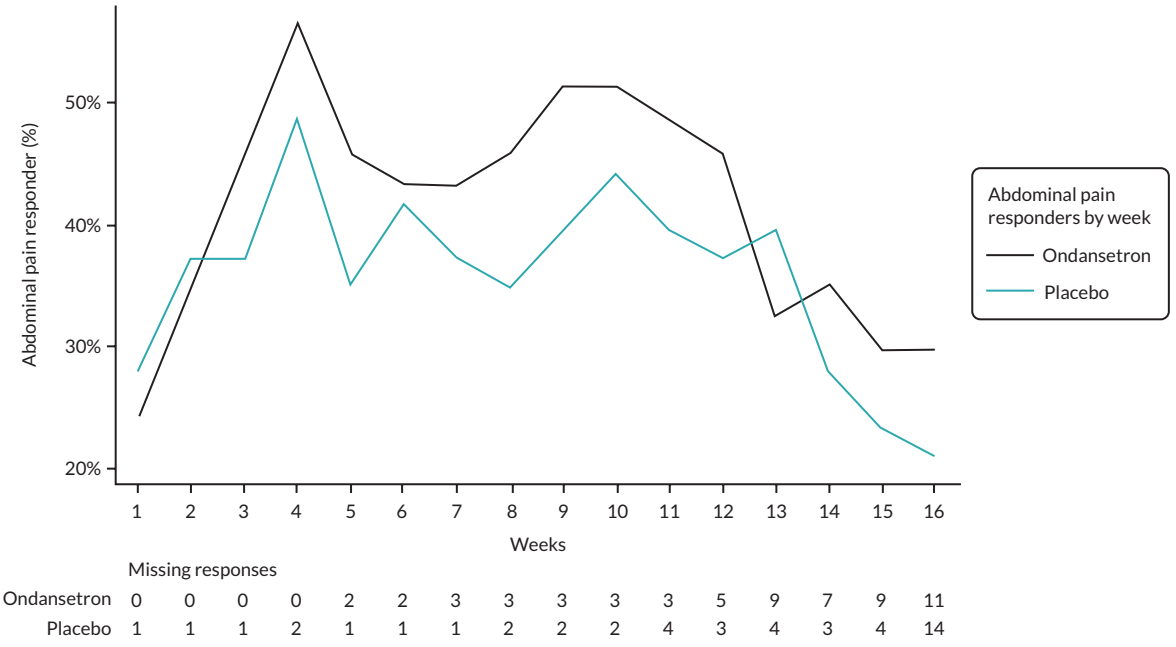


FIGURE 5 Weekly abdominal pain responders.

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FIGURE 6 Weekly stool consistency responders.

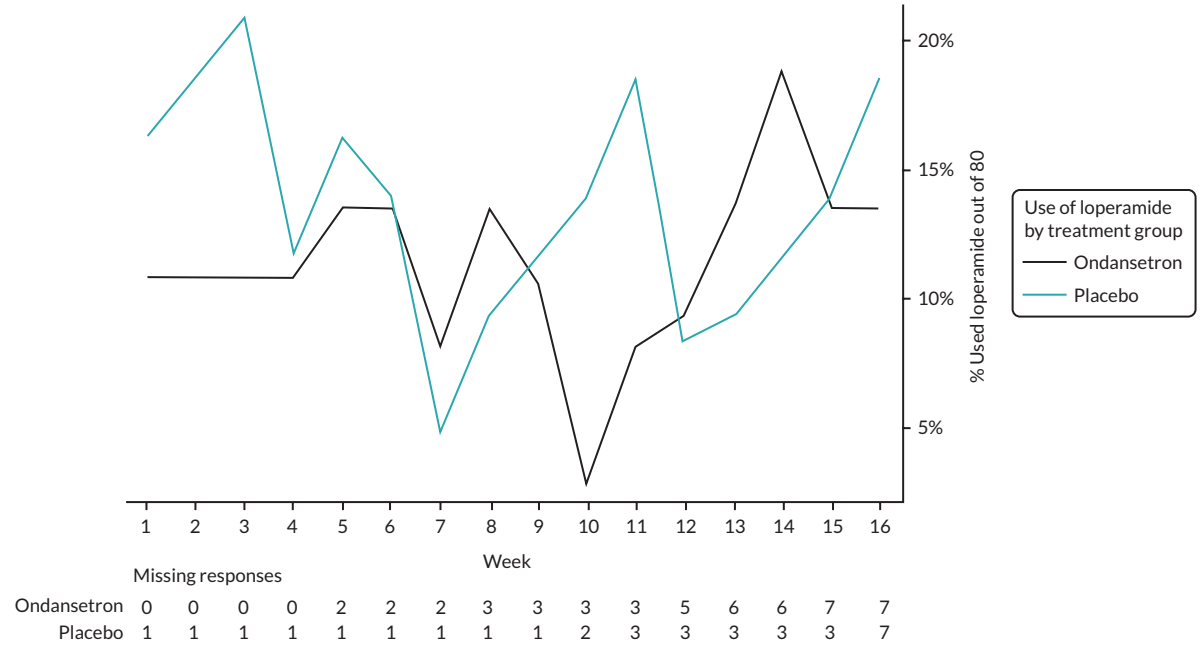


FIGURE 7 Loperamide use (proportion of participants).

from baseline (see *Table 11*). Urgency scores increased in both arms in the follow-up period compared with the treatment period. There was no evidence of a difference in urgency of defaecation scores between the treatment arms during or after the treatment periods (see *Tables 12*, *13* and *15*).

Number of days with loose stool, stool consistency and frequency

In all time periods reported, participants in the placebo arm had a higher mean number of days per week with loose stool and a higher mean stool consistency compared with those in the ondansetron arm. Mean daily number of stools reported by participants was similar during the treatment period

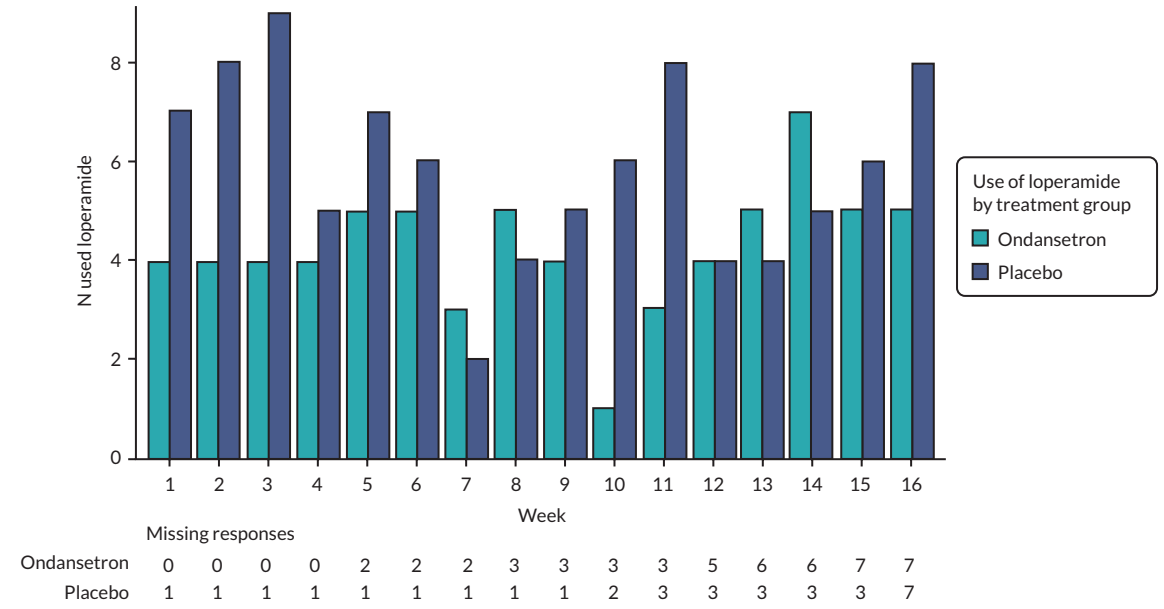


FIGURE 8 Loperamide use (number of participants).

and in the follow-up period (*Table 16*). There was a significant difference in stool consistency in favour of ondansetron between the treatment arms both in the final month of treatment, adjusted mean difference −0.5 [standard error (SE) 0.25, 95% CI (−1.0 to −0.02), $p = 0.0415$] (see *Table 12*) and during overall treatment period, adjusted mean difference −0.7 (SE 0.19, 95% CI (−1.0 to −0.3), $p = 0.0013$) (see *Table 15*). Weekly values are given in *Table 13*. There was no statistically significant difference in mean stool consistency in the follow-up period (see *Table 13*).

There was no evidence of a statistically significant difference in mean number of days per week with loose stool or mean daily stool frequency between treatment arms in the final month of treatment (see *Table 12*), in the follow-up period (see *Table 13*), or during the entire duration of treatment (see *Table 15*).

Hospital anxiety and depression scale

At baseline, participants in both treatment arms had similar HADS anxiety scores, and those in the ondansetron arms had higher depression scores. At 12 weeks, both anxiety and depression scores decreased in both arms, but were generally lower in the ondansetron arm (see *Appendix 6*, *Tables 45* and *46*). A higher proportion of participants on ondansetron reported reduction in anxiety and/or depression at 12 weeks compared with those on placebo. Anxiety and depression categories are summarised in *Appendix 6*, *Table 45*. There was no evidence of statistically significant differences between the arms at the end of the treatment period in either HADS anxiety or depression scores (see *Table 14*).

Irritable bowel syndrome severity scoring system

At baseline, participants on ondansetron reported higher IBS symptom severity scores compared with those on placebo. At the end of the treatment period, symptom severity reduced more in the ondansetron arm and a higher proportion of participants in the ondansetron arm reported symptoms that were in remission or mild compared with placebo (*Table 17*). A higher proportion of participants in the ondansetron arm 19/31 (61.2%) than in the control arm 18/43 (41.8%) reported a reduction of 50 or more points in IBS-SSS at 12 weeks compared to baseline, but this was not significant ($p = 0.15$), Fisher Exact test (see *Appendix 6*, *Table 47*). There was no evidence of statistically significant differences between ondansetron and placebo in IBS-SSS scores at the end of treatment period (see *Table 14*).

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TABLE 11 Abdominal pain and urgency of defaecation scores by time point and treatment arm

	Weeks 1–12			Weeks 9–12			Off-treatment weeks 13–16		
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Abdominal pain score (0–100)									
Mean (SD) missing	44.6 (23.13) 2	42.6 (21.25) 1	43.5 (22.00) 3	42.7 (25.28) 3	41.0 (25.02) 4	41.8 (24.98) 7	47.4 (27.08) 10	46.2 (24.06) 4	46.7 (25.14) 14
Median (range)	44.2 (5.5, 100.0)	47.3 (6.0, 81.9)	46.4 (5.5, 100.0)	44.0 (0.4, 100.0)	44.6 (1.4, 81.5)	44.5 (0.4, 100.0)	47.3 (3.2, 100.0)	48.0 (0.0, 91.5)	47.7 (0.0, 100.0)
Urgency score (0–100)									
Mean (SD) missing	44.2 (27.54) 0	45.1 (22.35) 3	44.7 (24.82) 3	40.7 (29.09) 5	43.4 (24.01) 4	42.2 (26.26) 9	48.0 (29.04) 10	50.9 (23.95) 6	49.7 (26.04) 16
Median (range)	38.9 (3.1, 100.0)	47.3 (7.1, 81.6)	44.9 (3.1, 100.0)	37.2 (0.7, 100.0)	46.2 (0.0, 78.2)	44.3 (0.0, 100.0)	45.7 (0.8, 100.0)	53.8 (0.4, 85.5)	51.5 (0.4, 100.0)

Notes
Abdominal pain score scale from 0 to 100, 0 = no pain, 100 = worst imaginable pain.
Urgency of defaecation from 0 to 100; 0 = no urgency, 100 = worst imaginable urgency.

TABLE 12 Secondary outcomes means over weeks 9–12 – treatment period (linear models)

Analysis (linear models) – weeks 9–12	Adjusted mean ondansetron	Adjusted mean placebo	Difference in adjusted means	SE	*p*-value	95% CI Lower	95% CI Upper	*N* analysed
Abdominal pain score (0–100)	40.0	41.6	−1.6	4.67	0.741	−10.9	7.8	73
Stool urgency (0–100)	35.6	42.5	−6.8	5.7	0.236	−18.2	4.6	70
Days/week with loose stool	2.4	3.0	−0.6	0.51	0.2652	−1.6	0.4	76
Stool consistency (BSFS) (1–7)	4.4	5.0	−0.5	0.25	0.0415	−1.0	0.02	71
Stool frequency (*n* stools/day)	2.5	2.5	0.1	0.28	0.8166	−0.5	0.6	73

TABLE 13 Secondary outcomes means over follow-up weeks 13–16

Analysis (linear models) follow-up weeks 13–16	Adjusted mean ondansetron	Adjusted mean placebo	Difference in adjusted means	SE	*p*-value	95% CI lower	95% CI upper	*N* analysed
Abdominal pain score (0–100)	44.4	46.0	−1.5	4.55	0.7389	−10.6	7.6	66
Stool urgency (0–100)	43.9	53.3	−9.3	4.94	0.0639	−19.2	0.6	63
Days week with loose stool	3.1	3.8	−0.7	0.54	0.2076	−1.8	0.4	70
Stool consistency (BSFS) (1–7)	5.1	5.2	−0.1	0.23	0.6166	−0.6	0.3	67
Stool frequency (*n* stools/day)	2.6	2.9	−0.2	0.3	0.4254	−0.9	0.4	70

Short-form Leeds dyspepsia questionnaire

Participants in the ondansetron arm at baseline reported higher severity of dyspeptic symptoms compared with placebo. At the end of 12 weeks, both groups reported reduction of severity of symptoms, although the reduction was greater in the ondansetron arm. The largest reduction was in symptoms of indigestion and nausea (see *Appendix 6*, *Table 50*). There was evidence of a statistically significant difference between ondansetron and placebo in the total SFLDQ score, with an adjusted mean difference in scores, −3.2 points [SE 1.43, 95% CI (−6.1 to −0.4), $p = 0.0275$], suggesting that participants in the ondansetron arm experienced lower severity of dyspeptic symptoms (see *Table 14*). However, the result needs to be considered with caution due to the small sample size and missing data.

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TABLE 14 Secondary outcomes questionnaires at 12 weeks (linear models) – end of treatment period

Questionnaire	Adjusted mean ondansetron	Adjusted mean placebo	Difference in adjusted means	SE	*p*-value	95% CI lower	95% CI upper	N analysed
HADS anxiety score (0–21)	8.4	9.4	−1	0.84	0.2464	−2.7	0.7	72
HADS depression score (0–21)	5.3	6.1	−0.8	0.85	0.3659	−2.5	0.9	72
IBS-SSS questionnaire score (0–500)	228	254.5	−26.5	32.51	0.4183	−91.5	38.5	69
SFLDQ (0–32)	6.1	9.3	−3.2	1.43	0.0275	−6.1	−0.4	66
IBS-QOL questionnaire score (0–100)	61.9	53.1	8.8	4.96	0.0817	−1.1	18.7	72

TABLE 15 Additional secondary outcomes analyses – means over whole treatment period weeks 1–12

Analysis (linear models) – weeks 1–12	Adjusted mean ondansetron	Adjusted mean placebo	Difference in adjusted means	SE	*p*-value	95% CI lower	95% CI upper	N analysed
Abdominal pain score (0–100)	41.6	43.5	−1.8	3.88	0.6391	−9.6	5.9	77
Stool urgency (0–100)	38.4	44.9	−6.5	4.8	0.1788	−16.1	3.1	76
Days/week with loose stool	2.3	3.2	−1	0.45	0.0362	−1.9	−0.1	79
Stool consistency (BSFS) (1–7)	4.4	5.0	−0.7	0.19	0.0013	−1.0	−0.3	75
Stool frequency (number of stools/day)	2.5	2.8	−0.3	0.25	0.2036	−0.8	0.2	76

Irritable bowel syndrome quality of life

At baseline, participants in the placebo arm reported better IBS-specific QoL compared with those in the ondansetron arm. At 12 weeks, participants in both arms reported improvements in QoL, those in the ondansetron arm reported a greater improvement; there was improvement reported in all subscales of the IBS-QOL (see *Appendix 6*, *Table 51*). There was no statistically significant difference in IBS-specific QoL between the treatment arms (see *Table 14*).

TABLE 16 Days/week with loose stool, stool consistency and number of stools

	Weeks 1–12			Weeks 9–12			Off-treatment weeks 13–16		
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Number of days/week with loose stool									
Mean (SD) missing	2.4 (2.13) 0	3.1 (2.06) 1	2.8 (2.11) 1	2.5 (2.38) 3	2.9 (2.18) 1	2.7 (2.26) 4	3.1 (2.52) 7	3.6 (2.17) 3	3.4 (2.32) 10
Median (range)	2.0 (0.0, 7.0)	2.7 (0.2, 7.0)	2.3 (0.0, 7.0)	1.9 (0.0, 7.0)	2.6 (0.0, 7.0)	2.4 (0.0, 7.0)	2.5 (0.0, 7.0)	3.6 (0.0, 7.0)	3.1 (0.0, 7.0)
Mean stool consistency									
Mean (SD) missing	4.4 (1.01) 3	5.1 (0.77) 1	4.8 (0.94) 4	4.4 (1.32) 4	5.0 (0.85) 4	4.7 (1.12) 8	5.0 (1.36) 8	5.2 (0.82) 4	5.1 (1.09) 12
Median (range)	4.2 (2.1, 6.5)	5.3 (3.9, 6.7)	4.9 (2.1, 6.7)	4.5 (1.6, 6.5)	5.2 (3.2, 6.8)	5.0 (1.6, 6.8)	5.1 (0.0, 6.8)	5.4 (3.4, 6.8)	5.2 (0.0, 6.8)
Mean daily number of stools									
Mean (SD) missing	2.6 (1.45) 2	2.7 (1.74) 1	2.7 (1.60) 3	2.7 (1.57) 3	2.5 (1.43) 3	2.6 (1.49) 6	2.6 (1.69) 6	2.8 (1.80) 3	2.7 (1.74) 9
Median (range)	2.5 (0.0, 7.0)	2.2 (0.1, 9.4)	2.3 (0.0, 9.4)	2.4 (0.0, 7.3)	2.0 (0.0, 8.1)	2.2 (0.0, 8.1)	2.5 (0.0, 7.6)	2.3 (0.0, 10.0)	2.4 (0.0, 10.0)

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TABLE 17 IBS-SSS summaries by treatment arm

	Baseline			12 weeks		
IBS-SSS	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Overall score						
Mean (SD) missing	387.6 (89.06) 0	336.5 (82.21) 1	360.4 (88.73) 1	258.6 (137.80) 6	263.6 (113.85) 4	261.4 (124.09) 10
Median (range)	385.0 (155.0, 700.0)	335.0 (190.0, 500.0)	350.0 (155.0, 700.0)	270.0 (0.0, 500.0)	260.0 (30.0, 475.0)	265.0 (0.0, 500.0)
Categorical						
Remission (0–74)				3 (8.1%)	3 (7.0%)	6 (7.5%)
Mild (75–174)	1 (2.7%)	0 (0.0%)	1 (1.3%)	8 (21.6%)	5 (11.6%)	13 (16.3%)
Moderate (175–299)	4 (10.8%)	11 (25.6%)	15 (18.8%)	6 (16.2%)	15 (34.9%)	21 (26.3%)
Severe (300–500)	31 (83.8%)	31 (72.1%)	62 (77.5%)	14 (37.8%)	16 (37.2%)	30 (37.5%)
Missing	1 (2.7%)	1 (2.3%)	2 (2.5%)	6 (16.2%)	4 (9.3%)	10 (12.5%)

Note
IBS symptom severity score from 0 to 500, where 0 is the best score and 500 the worst.

Satisfactory relief of IBS symptoms (weeks 1–12)

There was no evidence of a statistically significant difference between treatment arms in terms of satisfactory relief of IBS symptoms (*Table 18*).

Use of rescue medication – number of days taken loperamide over weeks 1–12

Loperamide was used as a rescue medication by only a small per cent of subjects, mostly those that were not treatment responders (see *Appendix 5*, *Table 55* and *Figures 28* and *29*). The number of days participants took loperamide over weeks 1–12 was planned to be evaluated. However, data on the number of days were right-skewed with considerable proportion of participants not using loperamide at all (see *Appendix 7*, *Table 55*). We attempted to fit two-stage model; however due to low frequencies, we were unable to fit it. Logistic regression was used instead (participant used loperamide 0 days or used it at least for 1 day during the time period). There was no evidence of statistically significant differences between the treatment groups in using loperamide during treatment or post-treatment period (see *Table 18*). Abdominal pain response and stool response according to loperamide use are detailed in *Appendix 5*, *Figures 30* and *31*).

Exploratory analyses on end points measured weekly, repeated measures

Weekly summaries

Figures 9–14 provide graphical summaries of abdominal pain, stool frequency, stool consistency, number of days with loose stool and satisfactory relief by week. The largest observed difference between the treatment arms was in mean stool consistency during the treatment period. Participants in the ondansetron arm had improved stool consistency measured by the BSFS compared with those in the placebo arm; these differences diminished in the post-treatment period in weeks 13–16 (see *Figure 12*).

TABLE 18 Outputs from logistic models; satisfactory relief of IBS symptoms, urgency responders over weeks 1–12 – treatment period and loperamide use

	Number of participants (proportions)						
	Ondansetron (*n* = 37)		Placebo (*n* = 43)		Model output		
Secondary outcome	*N* (%) missing	(95% CI)	*N* (%) missing	(95% CI)	*p*-value	OR (95% CI)	*N*
Satisfactory relief of IBS symptoms (Yes) weeks 1–12	15 (40.5%) 12	(24.7% to 56.4%)	17 (39.5%) 9	(24.9% to 54.1%)	0.5332	1.43 (0.46 to 4.41)	58
Urgency responders (Yes) weeks 1–12	18 (48.6%) 0	(32.5% to 64.8%)	16 (37.2%) 4	(22.8% to 51.7%)	0.6041	1.3 (0.49 to 3.46)	76
Loperamide use (0 days of loperamide use)							
Weeks 1–12 treatment	23 (62.2%) 7	(46.5% to 77.8%)	22 (51.2%) 8	(36.2% to 66.1%)	0.1962	0.43 (0.12 to 1.55)	61
Weeks 9–12 only	23 (62.2%) 8	(46.5% to 77.8%)	23 (56.5%) 13	(38.6% to 68.4%)	0.3619	0.48 (0.10 to 2.31)	56
Weeks 13–16 post treatment	21 (56.8%) 8	(40.1% to 72.7%)	23 (56.5%) 13	(38.6% to 68.4%)	0.6277	1.39 (0.36 to 5.35)	56

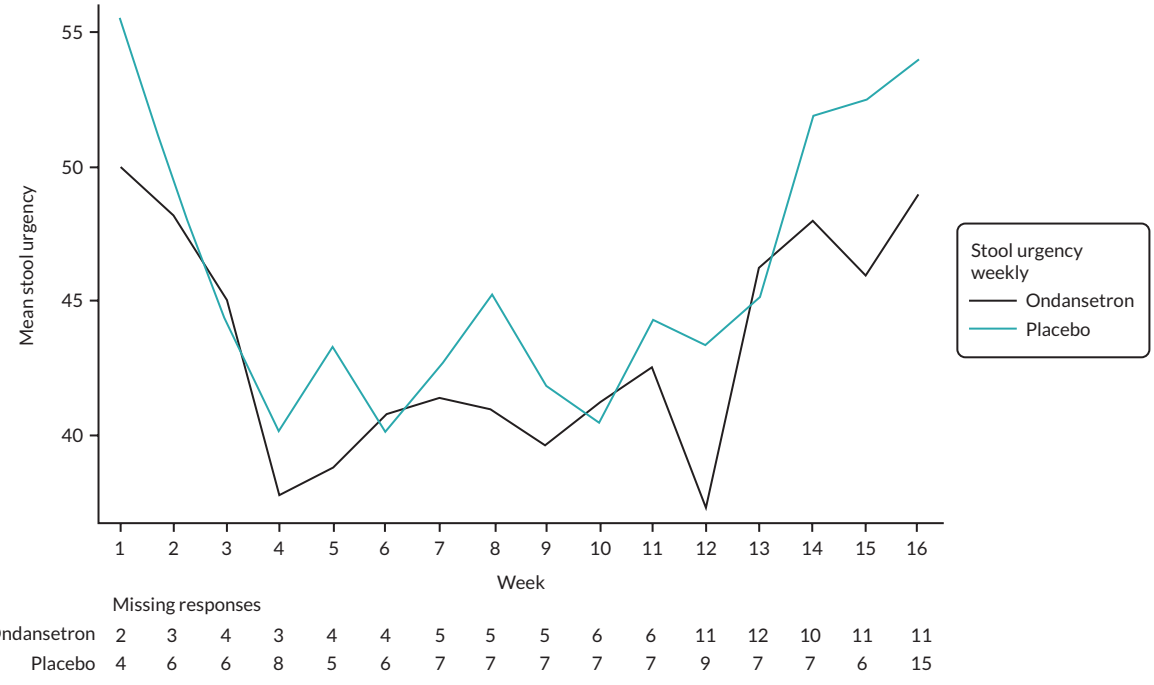
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Missing responses

Ondansetron	0	0	0	0	2	2	3	3	3	3	3	5	9	7	9	11
Placebo	1	1	1	2	1	1	1	2	2	2	4	5	4	3	4	14

FIGURE 9 Abdominal pain by week.



Missing responses

Ondansetron	2	3	4	3	4	4	5	5	5	6	6	11	12	10	11	11
Placebo	4	6	6	8	5	6	7	7	7	7	7	9	7	7	6	15

FIGURE 10 Stool urgency by week.

Tabulated summaries of numbers, proportions, means and SDs of the end points described above are in *Appendix 7*, *Tables 57–61*.

Repeated measures models

In all repeated measures models, a week was fitted as a time variable and interaction between time and treatment arm was included in the model. Unstructured covariance structure provided the best fit for models. Models were run for all 12 weeks of treatment, with numerical results presented for the last

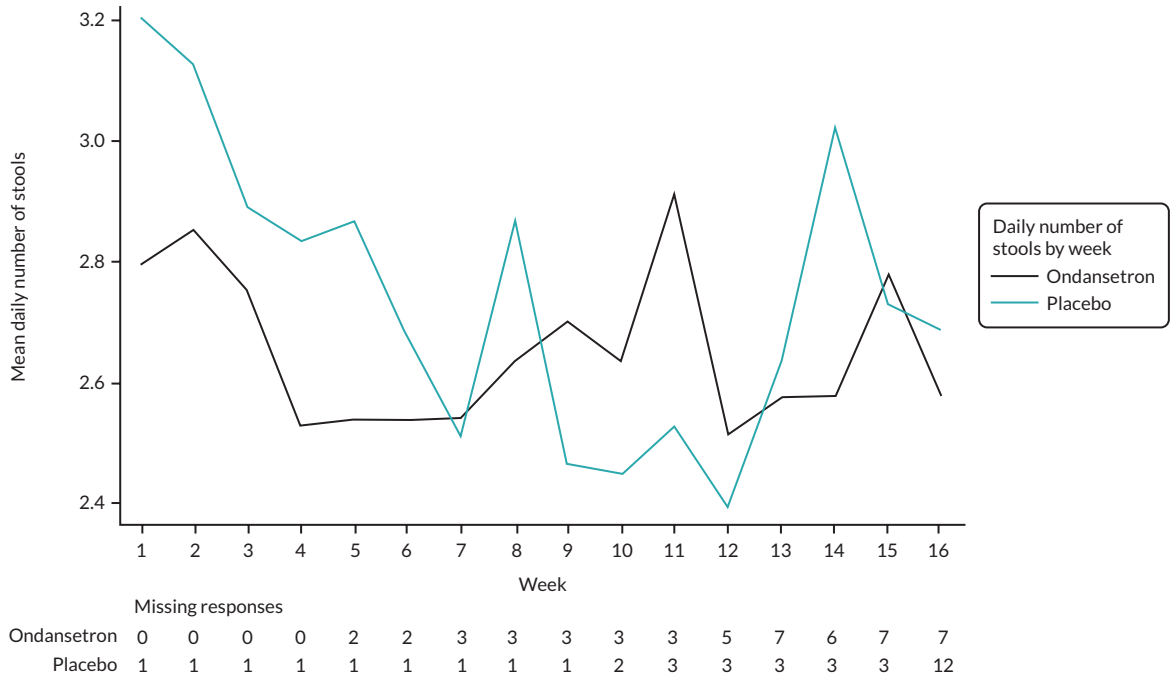


FIGURE 11 Daily number of stools by week.

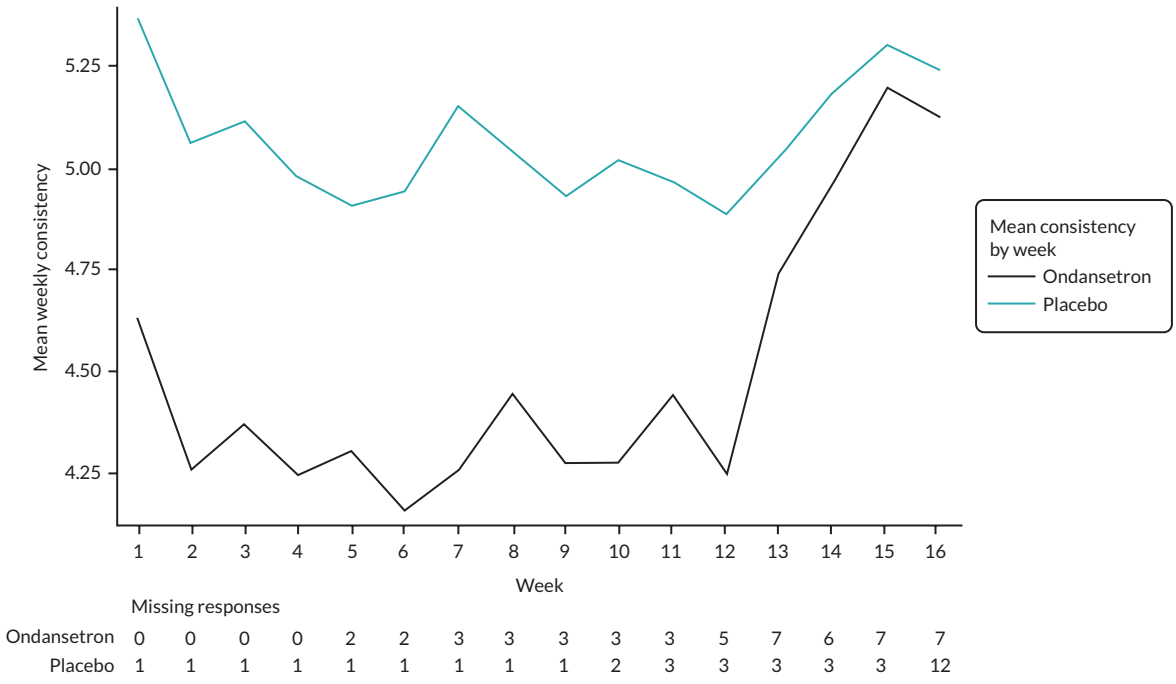


FIGURE 12 Mean stool consistency by week.

4 weeks of treatment due to space issues. The small number of participants needs to be considered in interpretation of repeated measures models described below.

Abdominal pain

There were no statistically significant differences between the treatment arms in weekly abdominal pain scores (*Table 19* and *Figure 15*).

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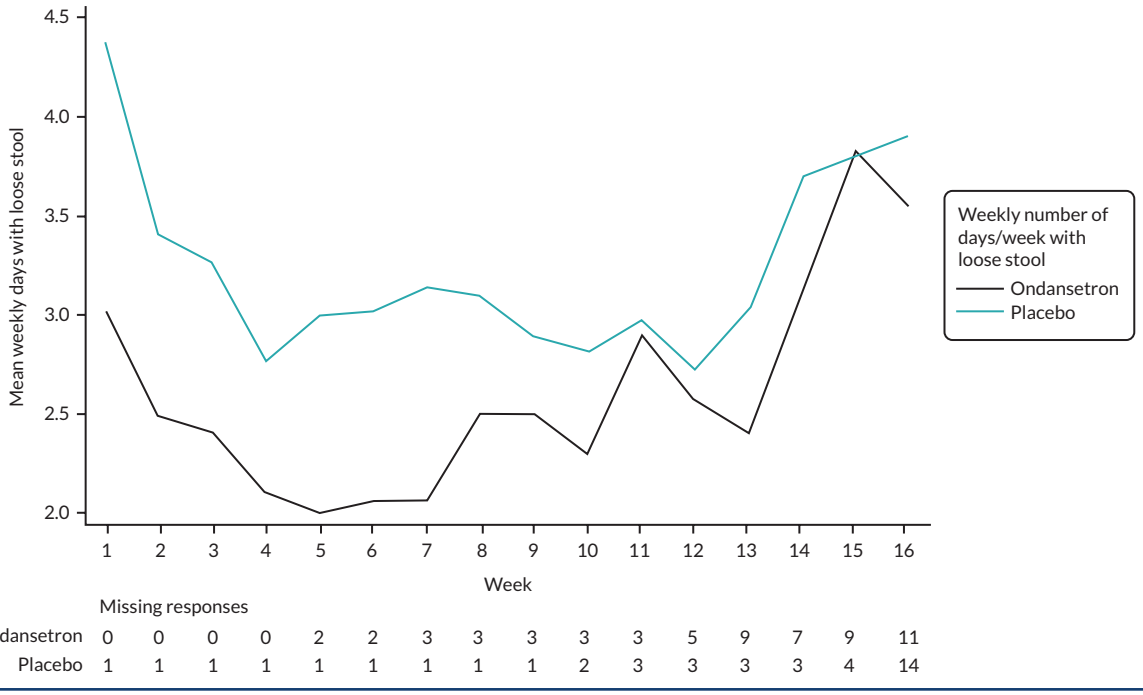


FIGURE 13 Number of days/week with loose stools by week.

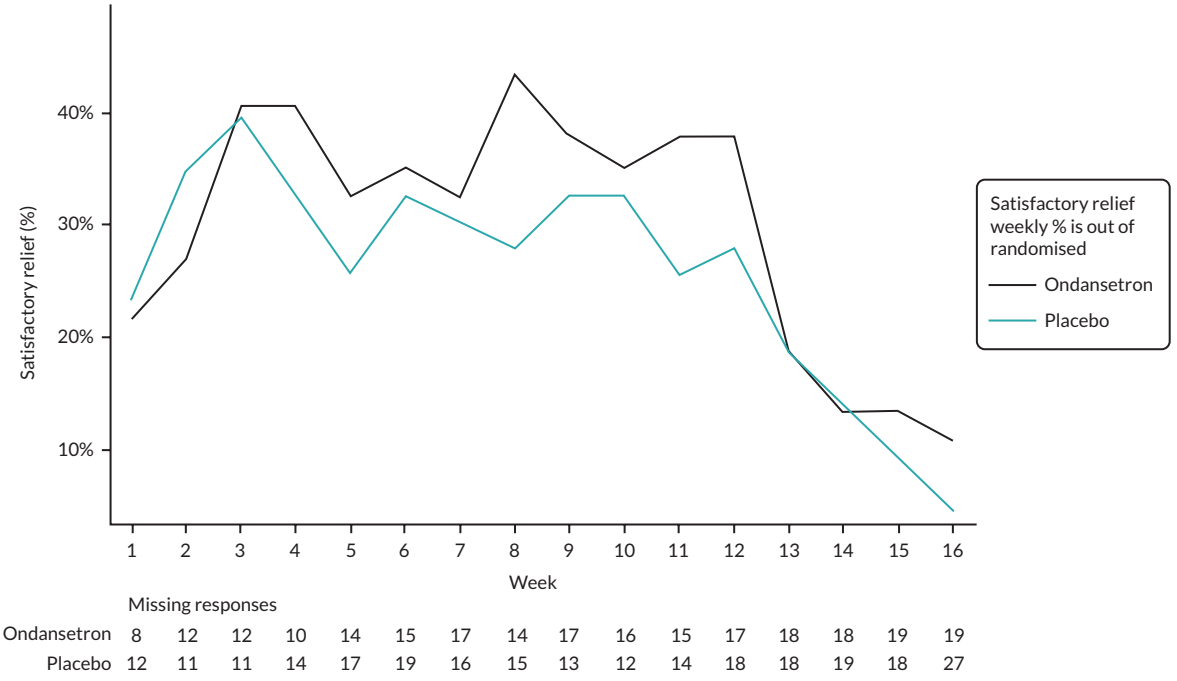


FIGURE 14 Satisfactory relief by week.

Stool urgency

There were no statistically significant differences between the treatment arms in stool urgency scores (see *Table 20* and *Figures 10* and *16*).

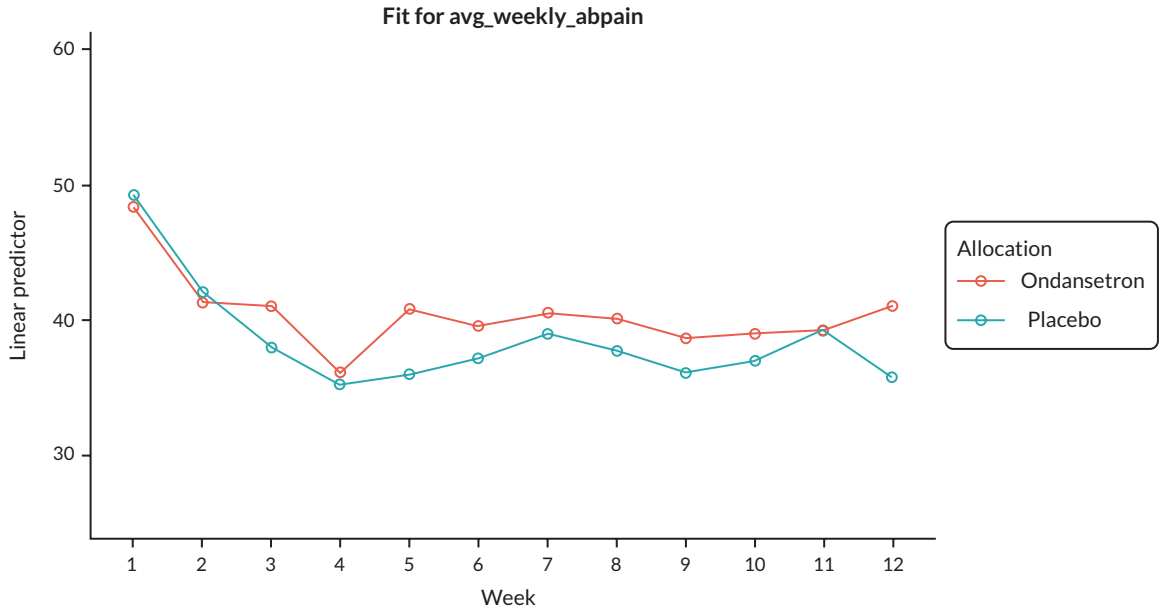
Urgency

Urgency responders were defined as participants that reported reduction in urgency score by at least 30% in at least 6 weeks of 12 weeks treatment. There was no evidence of a difference in proportions of responders between the treatment arms (see *Appendix 6*, *Table 45* and *Figure 17*).

TABLE 19 Abdominal pain repeated measures model – last 4 weeks of treatment displayed

	Ondansetron, adjusted abdominal pain score		Placebo, adjusted abdominal pain score		Difference		
Week	Est (SE)	(95% CI)	Est (SE)	(95% CI)	Est (SE)	(95% CI)	*p*-value
9	39.7 (3.51)	(32.7 to 46.7)	42.3 (3.37)	(35.6 to 49.0)	−2.6 (4.40)	(−11.4 to 6.2)	0.5607
10	40.6 (3.59)	(33.5 to 47.8)	42.6 (3.45)	(35.7 to 49.4)	−1.9 (4.53)	(−11.0 to 7.1)	0.6680
11	42.9 (4.03)	(34.9 to 50.9)	42.8 (3.83)	(35.2 to 50.4)	0.1 (5.16)	(−10.2 to 10.4)	0.9849
12	39.4 (4.06)	(31.4 to 47.5)	44.7 (3.85)	(37.1 to 52.4)	−5.3 (5.19)	(−15.6 to 5.0)	0.3105

Allocation*week interaction: *p*-value 0.9241.



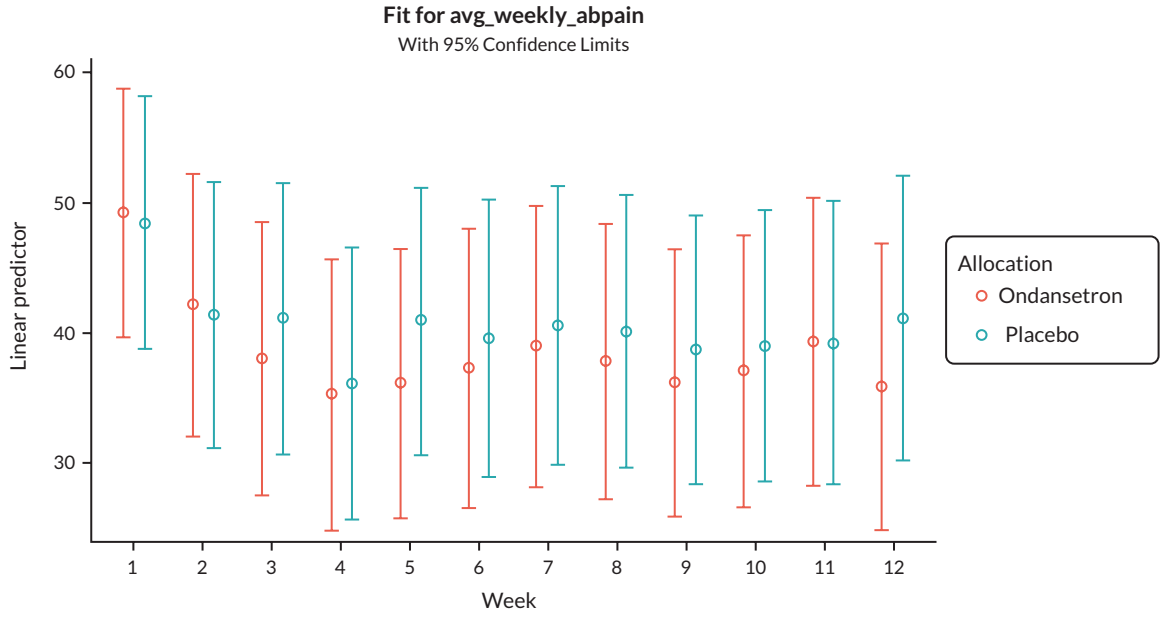


FIGURE 15 Abdominal pain, output from repeated measures model, adjusted scores and 95% CI by week.

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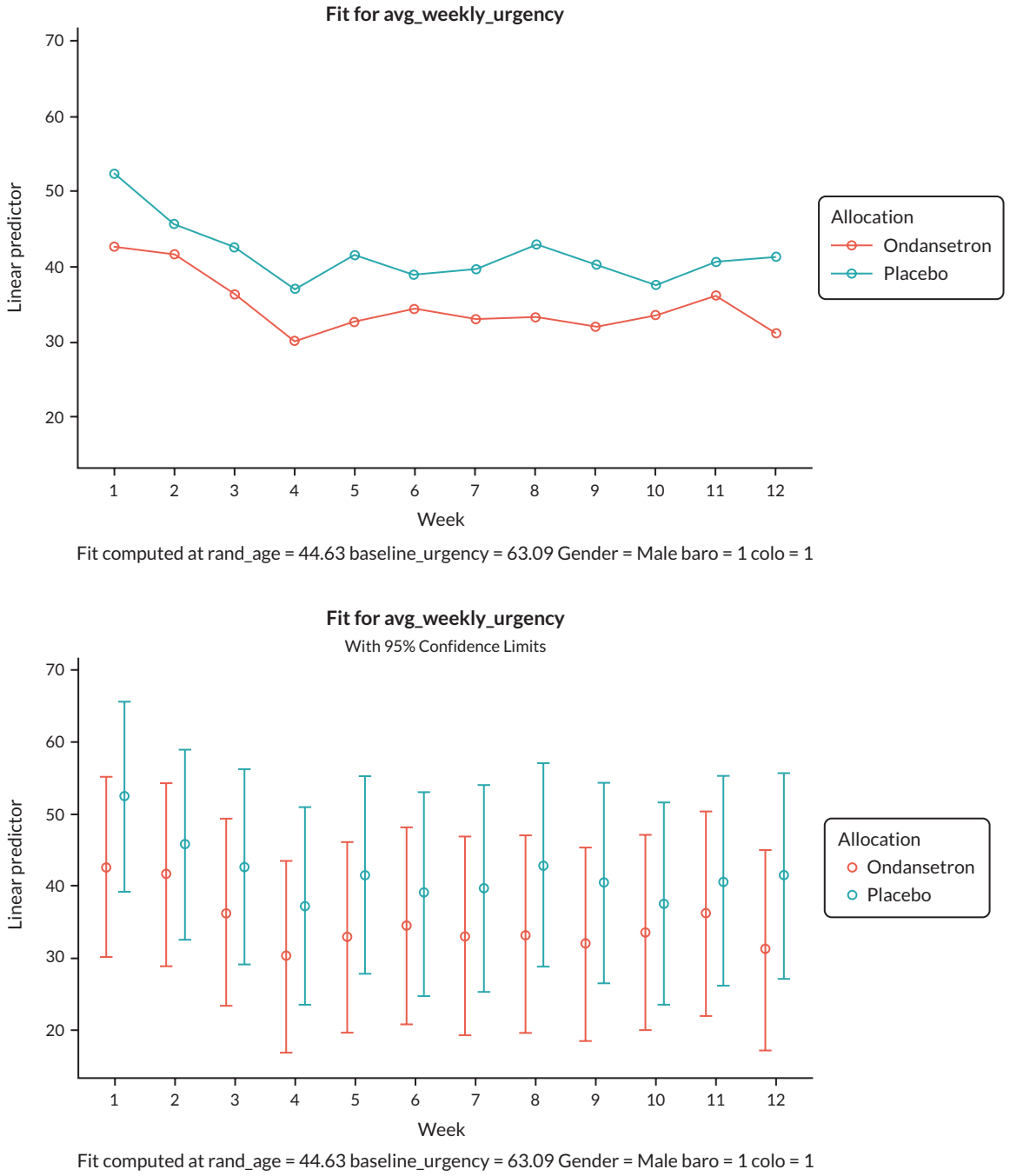


FIGURE 16 Stool urgency, output from repeated measures model, adjusted scores and 95% CI by week.

Stool frequency

We were unable to fit the Poisson model due to dispersion, and a negative binomial model did not converge, so stool frequency was fitted using a linear regression model instead as stool frequency was fitted as continuous data. There were no statistically significant differences in daily stool frequency between participants in the ondansetron and placebo arms (see *Table 21*, *Appendix 7*, *Table 58*, *Figures 11* and *18*).

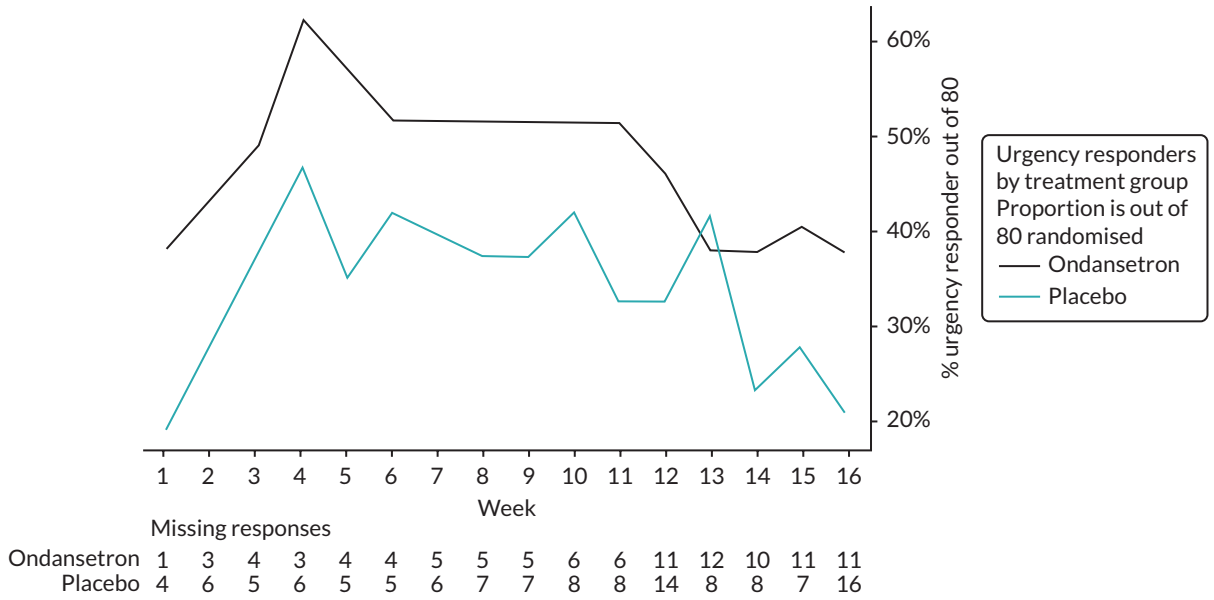


FIGURE 17 Urgency responders by treatment arm.

TABLE 20 Stool urgency repeated measures model – last 4 weeks of treatment displayed

	Ondansetron, adjusted stool urgency score		Placebo, adjusted stool urgency score		Difference		
Week	Est (SE)	(95% CI)	Est (SE)	(95% CI)	Est (SE)	(95% CI)	*p*-value
9	36.1 (4.35)	(27.5 to 44.8)	44.5 (4.42)	(35.7 to 53.3)	−8.4 (5.51)	(−19.4 to 2.6)	0.1326
10	37.7 (4.51)	(28.7 to 46.7)	41.8 (4.54)	(32.7 to 50.8)	−4.1 (5.74)	(−15.5 to 7.4)	0.4806
11	40.4 (5.02)	(30.4 to 50.4)	44.9 (4.99)	(34.9 to 54.8)	−4.5 (6.48)	(−17.4 to 8.4)	0.4915
12	35.3 (4.74)	(25.9 to 44.7)	45.6 (4.72)	(36.2 to 55.0)	−10.3 (6.06)	(−22.4 to 1.8)	0.0930
Allocation*week interaction, *p*-value 0.7387.							

TABLE 21 Stool frequency, repeated measures model – last 4 weeks of treatment displayed

	Ondansetron, adjusted stool frequency		Placebo, adjusted stool frequency		Difference		
Week	Est (SE)	(95% CI)	Est (SE)	(95% CI)	Est (SE)	(95% CI)	*p*-value
9	2.72 (0.238)	(2.25 to 3.20)	2.96 (0.240)	(2.49 to 3.44)	−0.24 (0.294)	(−0.83 to 0.35)	0.4155
10	2.62 (0.259)	(2.10 to 3.13)	2.70 (0.257)	(2.19 to 3.21)	−0.08 (0.325)	(−0.73 to 0.57)	0.8066
11	2.93 (0.278)	(2.38 to 3.48)	2.76 (0.272)	(2.22 to 3.30)	0.17 (0.352)	(−0.54 to 0.87)	0.6364
12	2.60 (0.268)	(2.06 to 3.13)	2.74 (0.264)	(2.22 to 3.27)	−0.14 (0.337)	(−0.82 to 0.53)	0.6693
Allocation*week interaction, *p*-value for interaction 0.5825.							

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Stool consistency

Due to the small number of participants, we were not able to fit a proportional odds model for stool consistency measured by BSFS. A linear model was fitted instead. Values from weeks 9–12 are shown in *Table 22* and *Figure 19*. There was no evidence of statistically significant differences in mean daily stool consistency between participants in the ondansetron and placebo arms, Allocation*week interaction, *p*-value 0.3988.

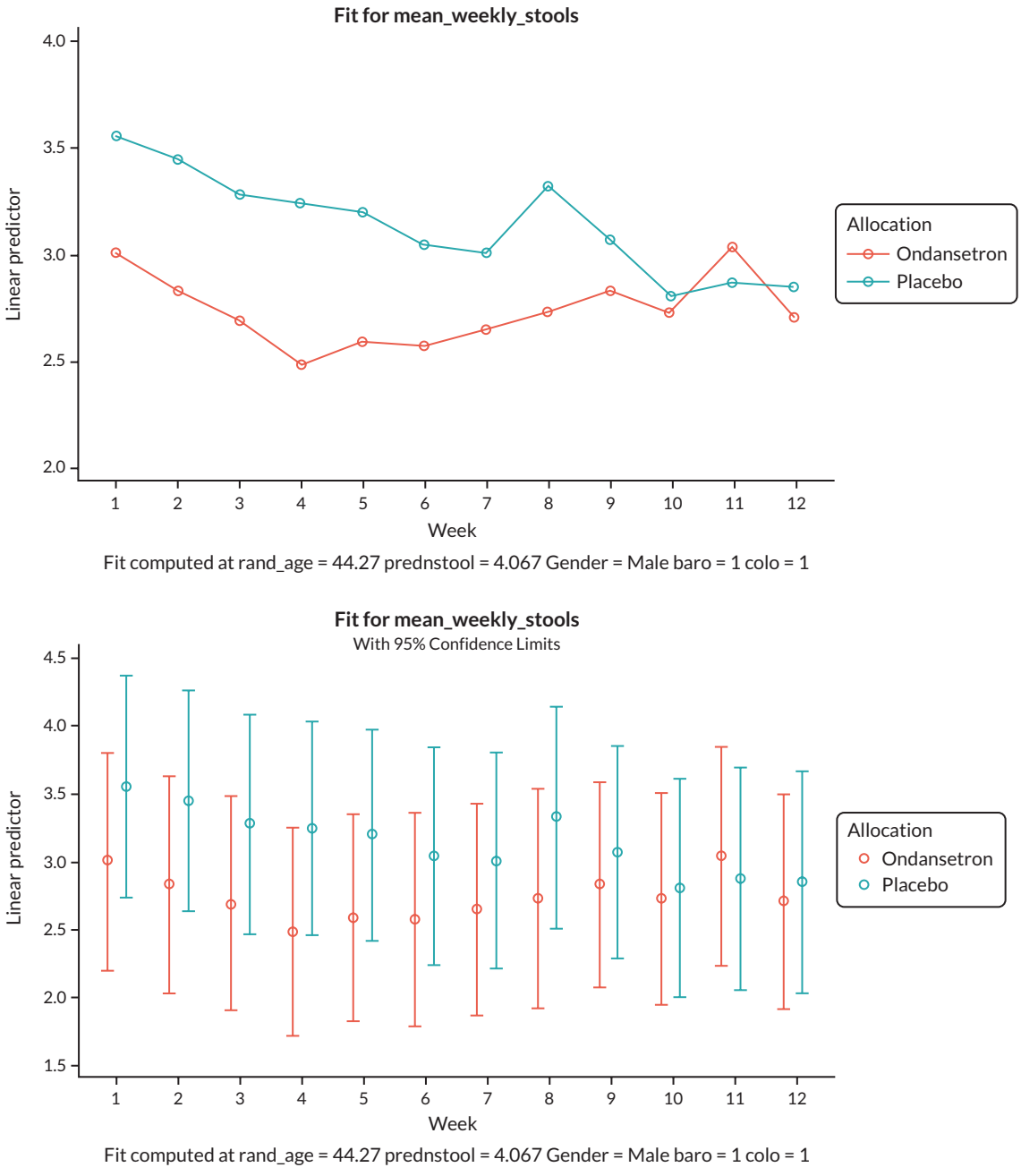


FIGURE 18 Stool frequency, output from repeated measures model, adjusted scores and 95% CI by week.

TABLE 22 Stool consistency, repeated measures model – last 4 weeks of treatment displayed

	Ondansetron, adjusted mean stool consistency		Placebo, adjusted mean stool consistency		Difference		
Week	Est (SE)	(95% CI)	Est (SE)	(95% CI)	Est (SE)	(95% CI)	*p*-value
9	4.09 (0.257)	(3.57 to 4.60)	4.64 (0.254)	(4.14 to 5.15)	−0.56 (0.323)	(−1.20 to 0.09)	0.0891
10	3.95 (0.264)	(3.42 to 4.47)	4.45 (0.259)	(3.94 to 4.97)	−0.51 (0.332)	(−1.17 to 0.16)	0.1319
11	4.14 (0.285)	(3.57 to 4.70)	4.49 (0.277)	(3.94 to 5.04)	−0.36 (0.362)	(−1.08 to 0.37)	0.3292
12	4.01 (0.302)	(3.41 to 4.61)	4.31 (0.292)	(3.73 to 4.89)	−0.30 (0.387)	(−1.07 to 0.47)	0.4409
Allocation*week interaction, *p*-value 0.3988.							

TABLE 23 Number of days with loose stool/week repeated measures model – weeks 9–12 displayed (difference)

	Ondansetron adjusted *n* of days/week with loose stool		Placebo adjusted *n* of days/week with loose stool		Difference		
Week	Est (SE)	(95% CI)	Est (SE)	(95% CI)	Est (SE)	(95% CI)	*p*-value
9	6.97 (0.133)	(6.71 to 7.24)	6.83 (0.122)	(6.59 to 7.07)	0.15 (0.177)	(−0.20 to 0.49)	**0.4048**
10	7.01 (0.133)	(6.75 to 7.27)	6.85 (0.122)	(6.61 to 7.09)	0.16 (0.177)	(−0.18 to 0.51)	0.3566
11	6.98 (0.133)	(6.72 to 7.24)	6.87 (0.122)	(6.63 to 7.11)	0.11 (0.177)	(−0.24 to 0.45)	0.5521
12	6.42 (0.135)	(6.16 to 6.69)	6.68 (0.123)	(6.44 to 6.93)	−0.26 (0.179)	(−0.61 to 0.09)	0.1478
Allocation*week interaction, *p*-value 0.4025.							

Number of days with loose stools

Number of days/week with loose stool was fitted as a model outcome. A Poisson regression model did not converge and so a linear regression model was used instead. There was no evidence of a statistically significant difference in the number of days with loose stools between the treatment arms (see *Table 23*, *Appendix 7*, *Table 60*, *Figures 13* and *20*).

Relief model

A logistic regression model was fitted; however odds ratios between the treatment arms need to be interpreted with caution due to missing data. Estimated treatment odds ratio varied by week (i.e. below and above 1) and there was no evidence of a statistically significant difference between arms.

Loperamide use model

A logistic regression model was fitted to the data and similarly to the relief model, odds ratios need to be interpreted with caution due to missing data and the small number of participants. Estimated treatment odds ratio varied by week and there was no evidence of a statistically significant difference between treatment arms.

Safety

There were no SAEs, serious adverse reactions or suspected unexpected serious adverse reactions reported. Comparison between treatment arms needs to be considered with caution as there were

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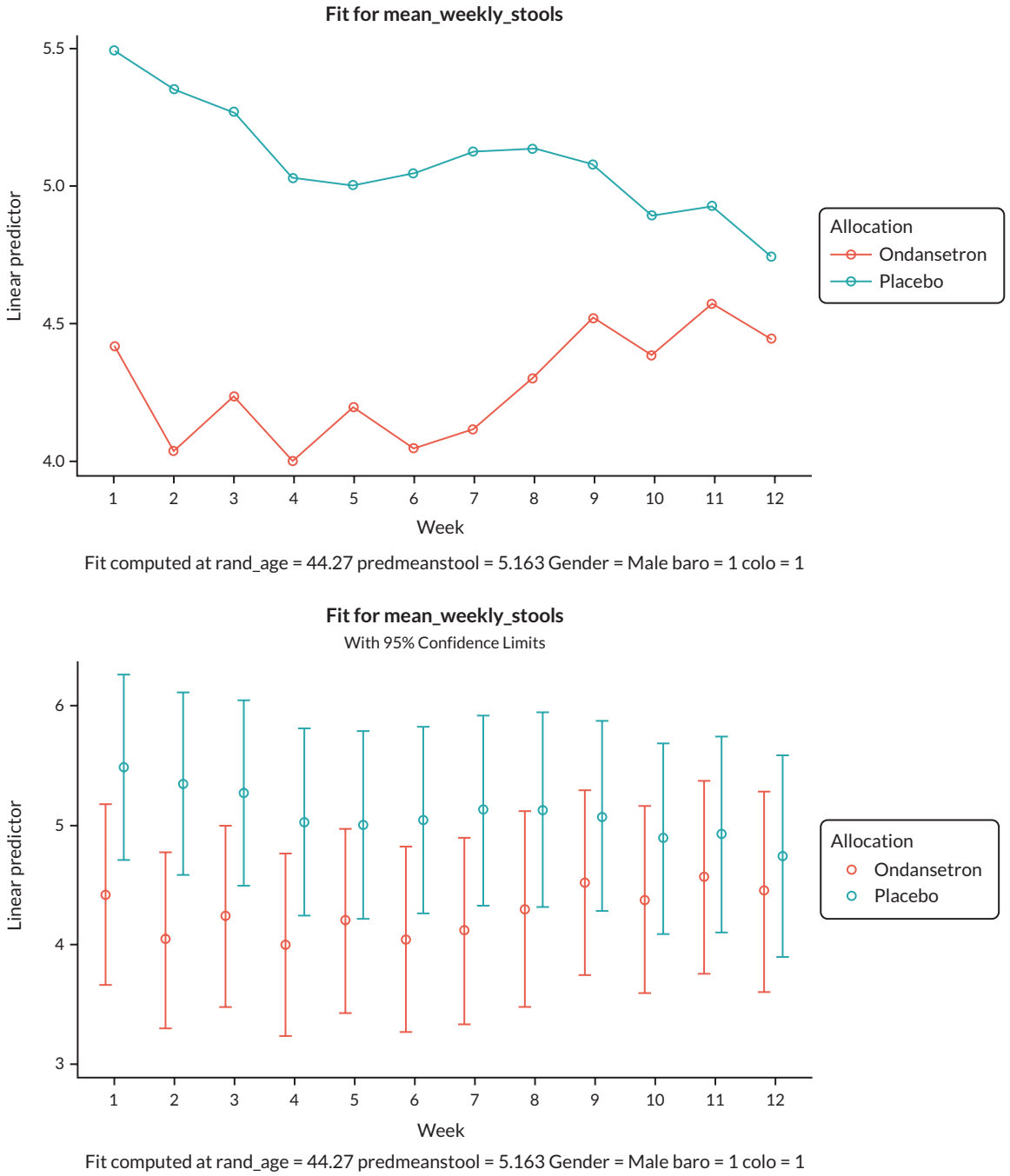


FIGURE 19 Stool consistency, output from repeated measures model, adjusted scores and 95% CI by week.

small numbers of participants in both arms (see *Appendix 8*, *Table 62*). During treatment at both 6 and 12 weeks, a higher proportion of participants in the ondansetron arm reported constipation compared with placebo. In the majority of cases, constipation was mild. Severe constipation was rare, 5% on ondansetron and 0% on placebo (see *Appendix 7*, *Table 61*).

At all time points (including follow-up), a higher proportion of participants in the placebo arm reported abdominal pain or bloating. However, the severity of abdominal pain/bloating was mild or moderate in the majority of participants (> 80%) at both 6 and 12 weeks. During follow-up, 17 (27.9%) participants overall reported severe abdominal pain/bloating.

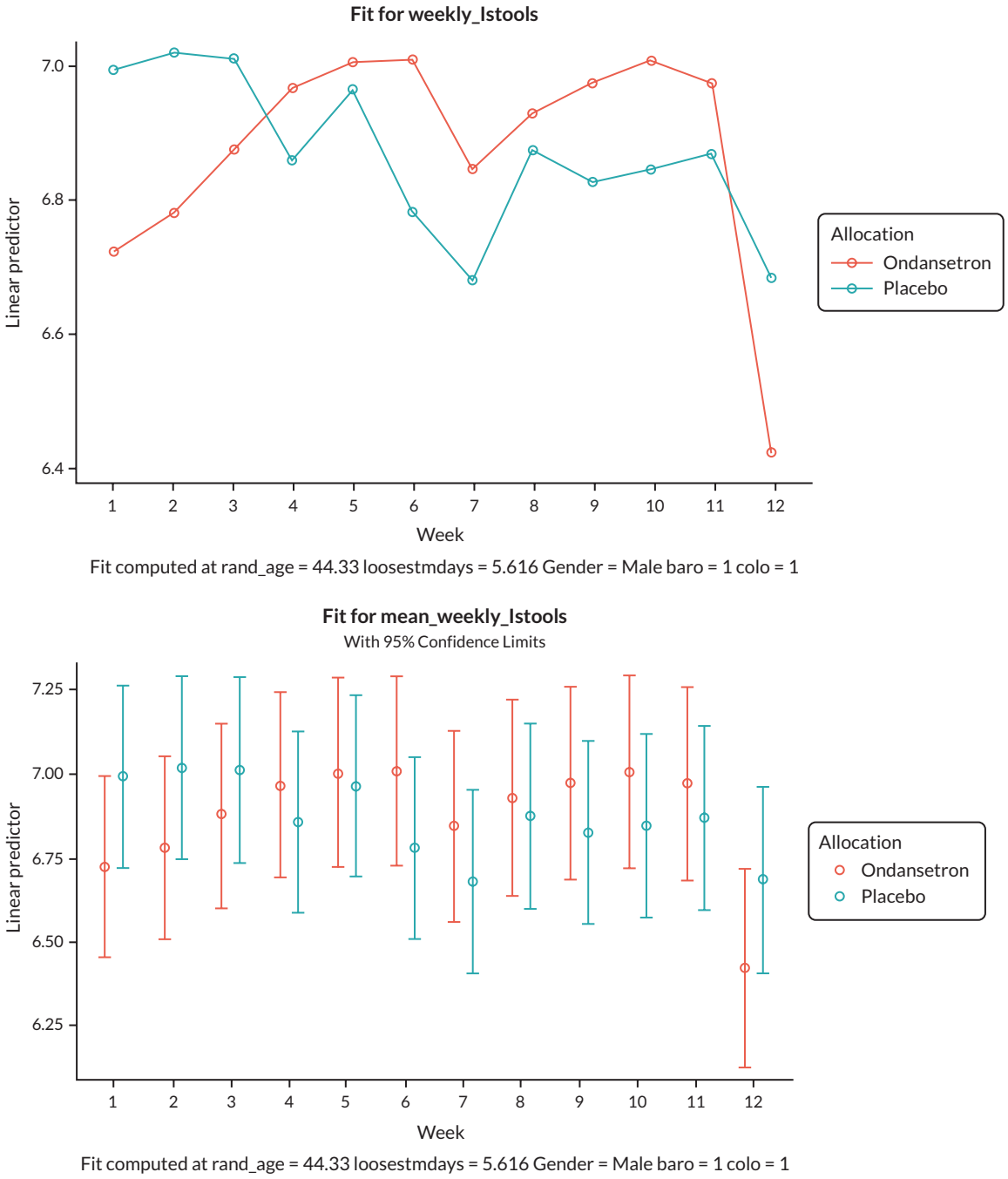


FIGURE 20 Number of days with loose stool/week, output from repeated measures model, adjusted scores and 95% CI.

At 6 weeks, a higher proportion of participants in the ondansetron arm reported headache, but at 12 weeks and during follow-up, proportions of participants with headache between arms were similar. The majority of headaches were mild or moderate with few rated severe.

A higher proportion of participants in the placebo arm reported nausea at 6 and 12 weeks. The majority of cases of nausea were mild or moderate. Less than 10% of participants reported vomiting during the treatment period and only one participant on ondansetron reported severe vomiting at that time.

A slightly higher proportion of participants in the placebo arm reported rectal bleeding at all time points. The proportion of participants with rectal bleeding was small, 12.5% overall. One participant in the placebo arm had severe rectal bleeding; the remaining participants had only minor rectal bleeding. At

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6 weeks, all but 1 of the 10 participants with rectal bleeding had a clinical review; none were thought to need colonoscopy or flexible sigmoidoscopy performed to exclude ischaemic colitis (see *Appendix 8*, *Table 63*).

At 12 weeks, 60 (75%) participants overall had pulse and BP reported. All those with data had pulse within the normal range and two participants (one in each arm) recorded BP outside the normal range (see *Appendix 8*, *Table 64*).

There was one pregnancy reported in the placebo arm, with a positive pregnancy test confirmed after the treatment period, 96 days post randomisation; the outcome was a healthy live birth (see *Appendix 8*, *Table 65*).

At 6 weeks, five participants had commenced a new medication (two in the ondansetron and three in the placebo arm), all of which were considered by the PI as not restricted or prohibited and the participants were considered suitable for continuation in the trial (see *Appendix 8*, *Table 66*).

Additional summaries

Dose titration

Participants were allowed to self-titrate during the treatment period. Based on clinical experience, it was considered unlikely that participants would need to alter their dose after 6 weeks. The recommended titrated dose at 2 weeks, the requested dose at 6 weeks and the number and percentage of patients taking a constant dose from 6 weeks onwards are reported in *Appendix 8*, *Table 67*. A higher proportion of participants in the placebo arm required higher titrated doses of IMP after 2 weeks of treatment. Consequently, a higher number of bottles was requested for placebo participants at 6 weeks of replenishment.

In the majority of cases 54/80 (72%) overall, the number of capsules returned at 12 weeks was not consistent with the dose at 6 weeks (see *Appendix 8*, *Table 62*).

Unplanned trial drug usage

No incorrect bottles were provided to participants and there was one replacement of two damaged bottles.

Blinding and exit poll

There were no unblindings reported.

An exit poll completed by the study nurse is summarised in *Table 24*. There are differences in blinding index between the treatment arms. The blinding index[45] ranges from −1 to +1 and a blinding index of 0 indicates compete blinding. The blinding index from the study nurse perspective was 0.40 [95% CI (0.17 to 0.63)] in the ondansetron arm compared with 0.10 [95% CI (−0.15 to 0.35)] in the placebo arm. As the lower limit of the 95% CI for the ondansetron arm is above zero; this suggests failure in masking in the ondansetron arm.

An exit poll completed by participants is in *Table 25*. There are differences in blinding index in participants' responses. The participant blinding index in ondansetron was 0.42 [95% CI (0.18 to 0.67)] and in placebo 0.05 [95% CI (−0.18 to 0.28)], suggesting unblinding in the ondansetron arm.

TABLE 24 Exit poll by study nurses (site perspective)

Randomised allocation	Exit poll response			Missing response	Total	Blinding index	(95% CI)
	Ondansetron	Placebo	Don't know				
Ondansetron	22	8	5	2	37	0.40	(0.17 to 0.63)
Placebo	17	21	2	3	43	0.10	(−0.15 to 0.35)

TABLE 25 Exit poll by patient responses

Randomised allocation	Exit poll response			Missing response	Total	Blinding index	(95% CI)
	Ondansetron	Placebo	Don't know				
Ondansetron	22	8	3	4	37	0.42	(0.18 to 0.67)
Placebo	16	18	7	2	43	0.05	(−0.18 to 0.28)

Possible explanations for this occurrence in terms of response could be that participants in the ondansetron arm were more likely to experience (and/or report to the study nurse) some therapeutic effects. However, results of the exit poll should be treated with caution due to the small number of participants.

Certainty of choice on the scale 1–10 and reasons for choice which were completed by the site study nurse are summarised in *Appendix 9*, *Tables 68* and *69*.

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Chapter 4 Mechanistic studies results

Transit

A total of 65 patients successfully completed whole gut transit time (WGTT) measurements at baseline, 28 on ondansetron and 37 on placebo. Their results are shown in *Table 26*. There were no significant differences between treatments at baseline; however when differences in WGTT between baseline and week 12 were compared, ondansetron tended to show a bigger increase (*Figure 21*), mean (SD) being 3.8 (9.1) on ondansetron versus a decrease −2.2 (10.4) on placebo, $p = 0.011$. Comparing the changes in regional transit times (right, left and rectosigmoid colon) between ondansetron and placebo (*Table 27*), only the increase in rectosigmoid transit was statistically significant.

Barostat

Data are available on just eight patients allocated to ondansetron and 10 on placebo of whom only seven and six, respectively, also underwent a second study on treatment, meaning we were seriously underpowered to assess any effect. See *Tables 28* and *29* below for further details at baseline.

Thus, despite showing a substantial change in the mean volume to reach urgency threshold (*Figure 22*), which on ondansetron rose by 84 (SD 61) ml while on placebo rose from 38 to 76, mean difference 38 (SD 48) ml, there was wide variability in this effect, which was non-significant, $p = 0.26$.

Thus, using these figures, we calculate that to detect the difference in volume threshold urgency with 90% power would require 16 subjects per group showing as suspected, we were significantly underpowered. However, using a crossover design and paired test in the same individual we would need to study only 10 individuals, which could easily be done separate from any long-term trial.

TABLE 26 Whole gut transit time in hours at baseline and change at week 12 on ondansetron and placebo

	Baseline WGTT		Week 12 WGTT: change from baseline	
Treatment	***n***	**Mean (SD)**		
Ondansetron	28	6.8 (6.3)	27	3.8 (9.1)*
Placebo	37	10.4 (11.7)	37	−2.2 (10.4)

Ondansetron vs. placebo. * $p = 0.011$.

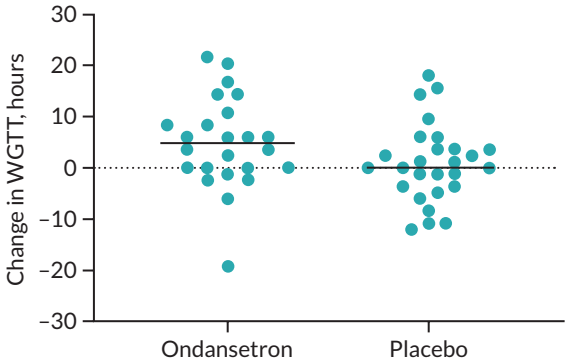


FIGURE 21 Change in WGTT in hours. This shows the change in WGTT in hours, which increased significantly more on ondansetron compared to placebo, $p = 0.011$.

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TABLE 27 Regional gut transit time in hours at baseline and change at week 12 on ondansetron and placebo median (interquartile range)

	Baseline				Week 12: change from baseline			
	WGTT	Right	Left	RS	WGTT	Right	Left	RS
Ondansetron	4.8 (2.7, 9.3)	2.4 (0.0, 6.0)	1.2 (0.0,3.3)	0.0 (0.0, 1.2)	3.6** (−2.4, 8.4)	0.0 (−2.7, 3.6)	0.0 (−1.2, 2.7)	1.2* (0.0, 5.1)
Placebo	7.2 (0.0, 4.8)	3.6 (0.6, 5.4)	1.2 (0.0, 4.2)	1.2 (0.0, 4.8)	−1.2 (−7.8, 3.0)	−1.2 (−2.4, 0.0)	0.0 (−1.2, 2.4)	0.0 (−1.2, 1.2)

* $p = 0.048$, ** $p = 0.01$.

TABLE 28 Demographics and barostat values at baseline

	Ondansetron, *n* = 8	Placebo, *n* = 10	*p*-value
Age, median (min–max)	50 (23–75)	57 (22–72)	0.76
Gender, M/F	3/4	1/5	0.6
Baseline anxiety (mean ± SD)	7 ± 3	11 ± 5	0.15
Baseline urgency volume threshold, ml (mean ± SD)	204 ± 76	196 ± 89	0.86
Baseline pain threshold, mmHg (mean ± SD)	29 ± 8	29 ± 12	0.98

TABLE 29 Change in thresholds for pain and urgency from baseline to week 12 on ondansetron vs. placebo

	Ondansetron	Placebo	*p*-value
n	7	6	
Pr1/2 (mmHg, mean ± SD)	3.5 ± 5.2	2 ± 5.6	0.64
Change in urgency threshold pressure (mmHg, mean ± SD)	9 ± 8	5 ± 6	0.27
Change in urgency threshold volume (ml, mean ± SD)	84 ± 61	38 ± 48	0.16
Change in pain threshold pressure (mmHg, mean ± SD)	5 ± 9	−1 ± 15	0.43
Change in pain threshold volume (ml, mean ± SD)	57 ± 70	24 ± 65	0.39

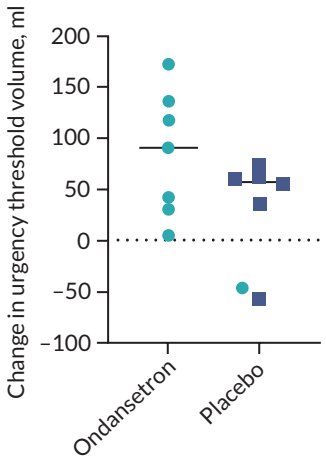


FIGURE 22 Change in urgency threshold volume by treatment. The change from baseline on treatment was assessed by barostat and showed a non-significant increase in volume on ondansetron.

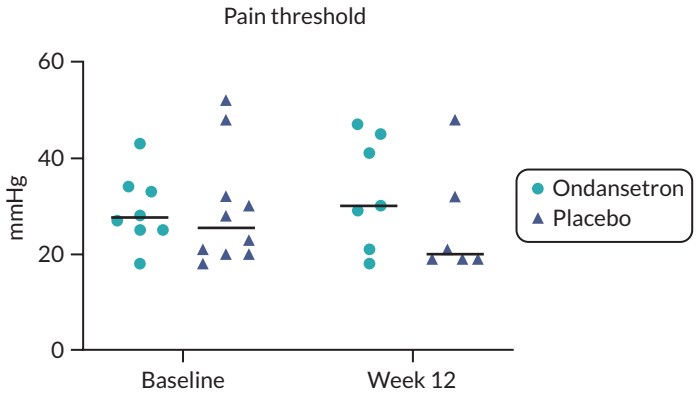


FIGURE 23 Pain threshold in mmHg. This was assessed from the barostat using the ascending method of limits at baseline and after 12 weeks treatment showing no significant effect of either time, treatment or interaction, p = 0.89, 0.4 and 0.4, respectively.

The difference in the change in pain threshold pressure (*Figure 23*) was −3.500 ± 7.0 mmHg, which would however have required 85 per group to achieve 90% power to detect such a difference emphasising the small effect size on pain. Unlike alosetron there was an even smaller effect size for compliance, which increased just −1.5 ± 3.1, to detect such a difference would require 97 in each group suggesting that ondansetron does not affect compliance. Given the very minimal effect on pressure pain threshold, it seems likely that it acts on the local reflexes to reduce urgency rather than a purely sensory effect.

High-resolution colonic manometry

Regrettably very few subjects completed the two manometry visits before and after treatment. Nine completed baseline assessment (5 allocated to ondansetron, 4 to placebo) and there was 1 dropout on ondansetron at week 12 assessment, leaving 4 and 4 to compare. Another complication was the wide variability in the depth of insertion of the manometry catheter. Some were only recording rectal activity [*Figure 24*, Panel (a)], while others reached the ascending colon [see *Figure 24*, Panel (b)].

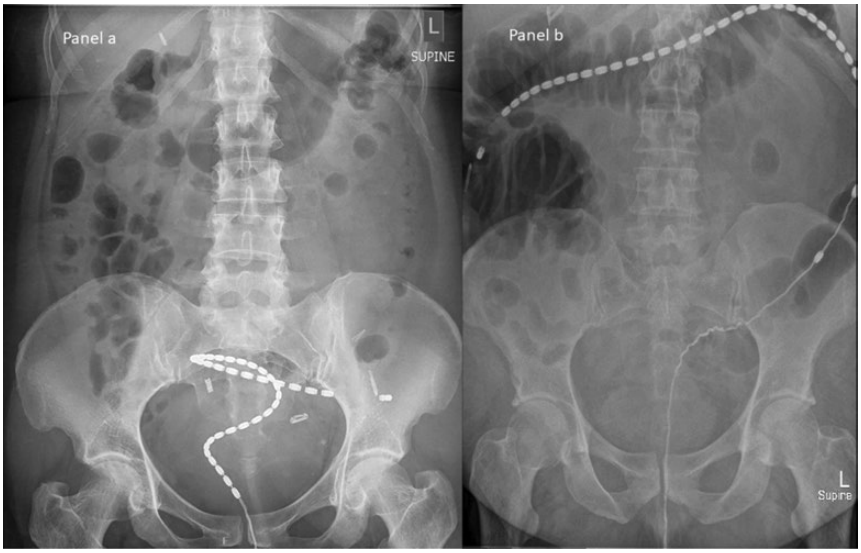


FIGURE 24 Plain X-ray at end of recording showing a very variable positioning achieved. Panel (a) shows recording ports mostly in the rectosigmoid region, while panel (b) shows recording ports in transverse colon missing the rectosigmoid altogether.

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Obviously, these data can only be used to develop hypotheses to be tested by further studies but nevertheless there were some interesting results.

We did look at the frequency of HAPCs in the 4 hours of recording, which at baseline (n = 8) were (median, range) 1 (0–3) and on ondansetron (n = 4) were 0, 1, 11, 6 and on placebo, (n = 4) 0, 0, 2, 0. With such small numbers there were no significant differences between treatments, p = 0.25, Mann–Whitney.

Subject 78 showed marked clinical improvement on ondansetron with large falls in urgency (80.7 falling to 10.0), stool frequency (4.9 falling to 2.1) and BSFS (6.5 falling to 3.0). The results are displayed in *Figures 25* and *26*. There is a striking change in patterns of motility with the baseline showing many rapid moving, almost simultaneous contractions which would be predicted to cause urgent defaecation, which fits perfectly with the patient's symptoms that were extreme.

Subject 58 also had good traces at both baseline and on ondansetron. *Figure 27* shows an increase in postprandial contraction of ondansetron and a shift in the phase spectrum indicating an increase in retrograde propagated contractions. These would push material from the rectum into the sigmoid, which would reduce urgency induced by liquid reaching the sensitive anorectal margin.

Thus, manometry is inconclusive. However, it does provide pilot data and a plausible hypothesis, namely that ondansetron stimulates retrograde activity in the rectosigmoid region, and this could be tested in future studies.

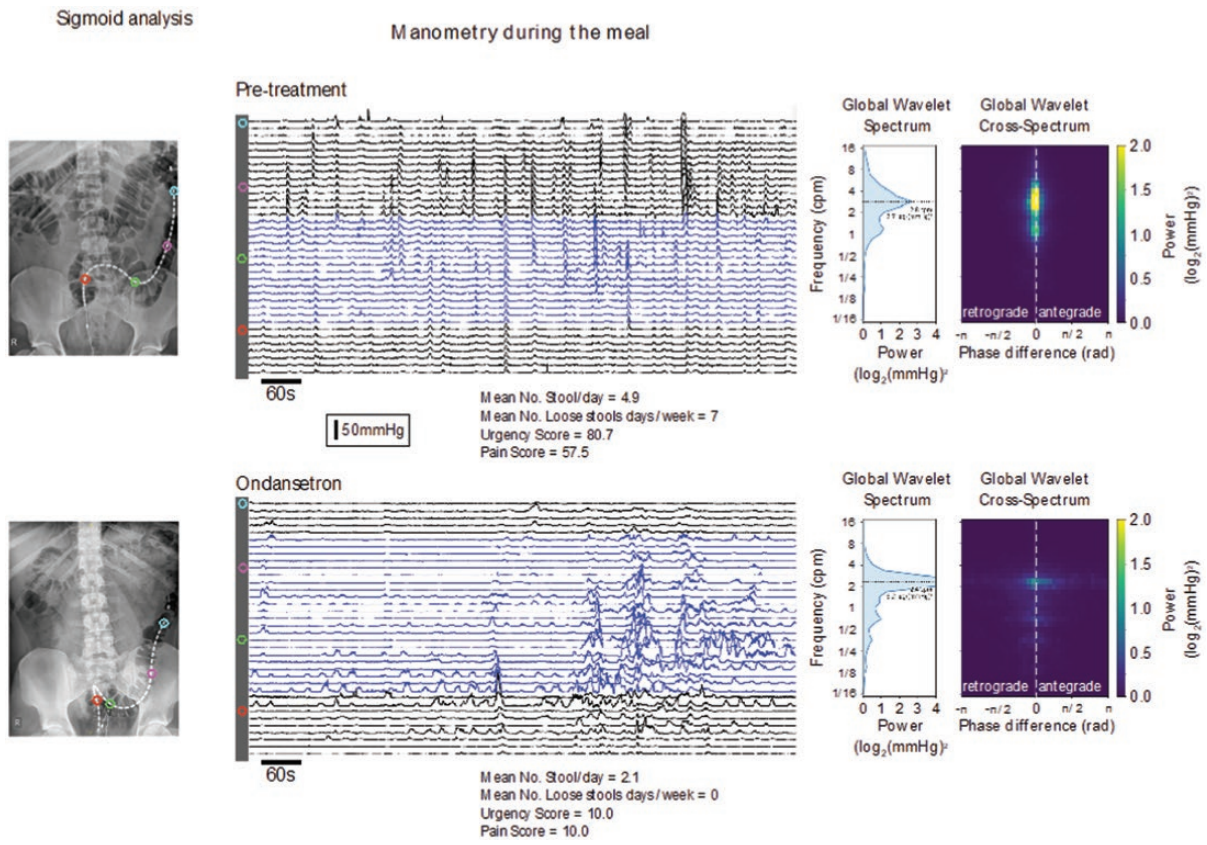


FIGURE 25 Manometry: example of response to a meal. Left-hand image shows position of manometry catheter, and the upper tracing shows pressure waves before ondansetron during a meal when contractions appear to rapidly traverse the sigmoid colon and rectum. In contrast, the lower trace made at the same time during a meal while taking ondansetron shows a very different, more normal pattern, with retrograde movements in both sigmoid and rectum. The right-hand panels depict the power spectrum of the pressure waves showing most are around 2–3 contractions per minute with greater power at baseline compared to after treatment.

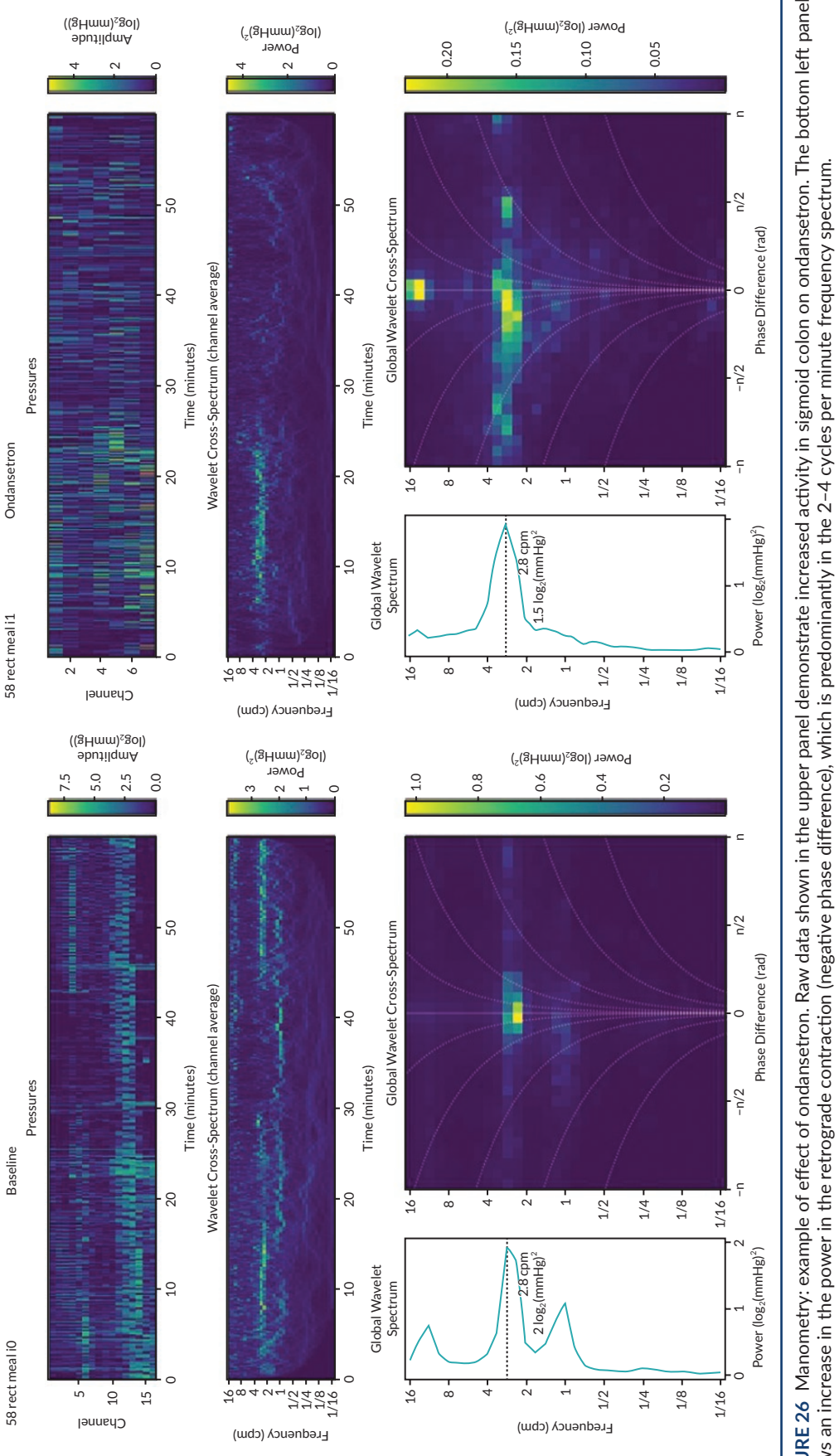


FIGURE 26 Manometry: example of effect of ondansetron. Raw data shown in the upper panel demonstrate increased activity in sigmoid colon on ondansetron. The bottom left panel shows an increase in the power in the retrograde contraction (negative phase difference), which is predominantly in the 2–4 cycles per minute frequency spectrum.

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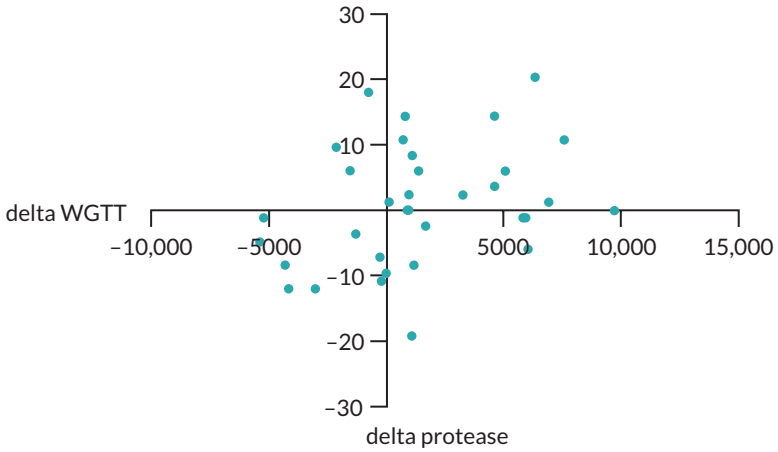


FIGURE 27 Change in WGTT in hours.

Stool % water

Combining all participants' stools water content at baseline was mean (SD) 72.4 (6.5) %, *n* = 48. There were no significant differences between ondansetron or placebo groups, either at baseline or week 12 (*Table 30*). There was no significant correlation with transit time, *r* = 0.02, *p* = −0.88, *n* = 48.

Loose stool responders to ondansetron (defined by < 1 day reduction in days per week with loose stools) tended to have less watery stool % water = 71.3 (6.3), *n* = 17 versus non-responders, stool % water = 75.5 (3.3), *n* = 4, but the difference was not significant, *p* = 0.16.

At baseline, there was no correlation between stool % water and urgency score, Pearson's *r* = 0.11, *n* = 52, *p* = 0.42, nor between stool % water and days with loose stool, stool consistency nor number of stools per day, *p* = 0.8, 0.8 and 0.2, respectively.

Faecal proteases

These were assessed at baseline and week 12 and the results are shown in *Table 31*. There was wide variability but no significant change by visit or treatment, *p* = 0.8, two-way ANOVA.

Contrary to our previous findings,[12] we found no correlation between baseline protease and WGTT, Spearman's *r* = −0.0037, *p* = 0.98, *n* = 39. However, when looking at the change from baseline in FP and WGTT at 12 weeks, Spearman's *r* = mean (95% CI) 0.41 (0.03 to 0.69), there is a statistically significant correlation *p* = 0.03 (*Figure 27*).

TABLE 30 Stool % water by visit and allocation

	Baseline		Week 12	
	n	Mean (SD)	*n*	Mean (SD)
Ondansetron	20	72.0 (6.5)	16	72.83 (5.7)
Placebo	27	72.9 (6.6)	16	74.20 (6.4)

TABLE 31 Faecal proteases in trypsin units/mg protein

	Baseline			Week 12		
	Mean	SD	*n*	Mean	SD	*n*
Placebo	2849	2620	23	2867	2600	17
Ondansetron	4017	2685	16	3753	2867	13

Change in WGTT in hours [delta WGTT plotted against associated change in FP (delta protease) in trypsin units/mg protein for all subjects]. Spearman's r = mean (95% CI) 0.32 (0.02 to 0.60), n = 34, p = 0.03.

Faecal bile salts

Faecal bile acids were assessed at baseline as shown in *Table 32*. Stools with more watery stool had lower total bile acid concentration, correlation with stool % water, Spearman's r = 0.56, N = 48.

As *Table 33* shows, at baseline there was wide variability. After 11–12 weeks of treatment, there was a numerical fall in primary bile acids on both ondansetron and placebo with a non-significant rise in the secondary/primary ratio.

Using the total sample of 51 at baseline, there was no correlation between the secondary/primary bile acids ratio and WGTT, Spearman's r = –0.08 (0.39–0.75), p = 0.9.

TABLE 32 Faecal bile acids concentration at baseline mean (SD)

Bile acids	CA (mM)	CDCA (mM)	DCA (mM)	LA (mM)	Total bile acids (mM)
Baseline	0.22 (0.13)	0.14 (0.17)	1.55 (1.05)	2.06 (1.5)	3.9 (2.1)

Notes
Primary bile acids: CA, cholic acid; CDECA, chenodeoxycholic acid.
Secondary bile acids: DCA, deoxycholic acid; LA, lithocholic acid.

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TABLE 33 Effect of ondansetron vs. placebo on primary and secondary bile acids in µM per litre mean (SD)

	Baseline			Week 11–12			
	P	S	S/P ratio	P	S	S/P ratio	*p**
	N = 23			*N* = 17			
Ondansetron	0.35 (0.24)	3.4 (1.77)	9.72 (7.08)	0.18 (0.09)	3.44 (1.88)	21.4 (32.9)	0.85
	N = 30			*N* = 18			
Placebo	0.36 (0.26)	3.40 (2.29)	22.84 (58.23)	0.25 (0.18)	3.435 (1.80)	28.61 (31.42)	0.56

**p*-value for paired comparison of difference in S/P ratio baseline vs. week 11–12, Wilcoxon matched pairs signed-rank test.

Note
P = Primary, S = secondary bile acids, S/P ratio = secondary/primary bile acid ratio.

Chapter 5 Discussion

Recruitment failure and lessons learnt

This study was ended prematurely because of failure to recruit the required number of patients, despite strenuous efforts by the research team as documented in *Chapter 6 Conclusions*. We had previously recruited well in secondary care,[12,46] so the current failure is most likely due to a substantial shift in referral practice, which markedly reduces the numbers of IBS-D patients referred to secondary care. Previously many patients suffering from pain and diarrhoea in primary care would have been referred to secondary care to exclude inflammatory bowel disease (IBD), particularly Crohn's disease. However, this leads to extensive negative investigations and high healthcare costs, particularly those associated with colonoscopy. Current evidence-based advice is for primary care practitioners to screen such patients with a faecal calprotectin and not to refer to secondary care for those that have normal values. This care pathway is associated with a substantial reduction in costs while still ensuring that inflammatory bowel disease (IBD) cases are appropriately referred.[42]

Although we responded to this reduction in referral by spreading our recruitment net wider, the alternative routes we used via advertising in media and screening via primary care patient lists proved very much less efficient. Of 620 self-referred patients who approached the trial staff for information, only 12 (2%) were ultimately randomised compared to 58/485 (12%) of referrals from secondary care. This emphasises that patients being referred to secondary care are already highly selected and well-motivated to take part in trials as their symptoms are severe enough that they want treatment. By contrast, given the fluctuating nature of IBS-D severity, our impression was that many patients approached using GP lists are often either not motivated or fail to meet severity criteria currently.

Given our small numbers, our ability to generalise is weak and the CIs for estimates are large. As such, we lacked power to demonstrate the efficacy of ondansetron in the treatment of IBS-D. Like many other trials, we found a strong placebo effect with substantial improvements in pain and stool consistency in the first 4 weeks such that, overall, in the present study around 30% met FDA criteria for response. The size predicted for this placebo effect led to our power calculation indicating we would need to recruit 400 patients to prove superiority over placebo.

We found many with a diagnosis of IBS-D did not suffer abdominal pain ≥ 1 days per week and, therefore, using Rome IV criteria to define IBS, they were not eligible. Although this change in criteria was intended to raise the bar for diagnosis so that patients meeting criteria would warrant treatment, it has changed the nature of the population and made recruitment more difficult.

Impact on primary end point

We were unable to demonstrate a significant increased response rate in the ondansetron arm, as defined by the FDA composite end point compared with placebo. As already mentioned, our data support our initial power calculations suggesting we needed > 400 patients to achieve adequate power (80%). As in our previous pilot study,[12] and the recently reported Bekinda trial using modified release ondansetron,[47] the effect on responder rate for stool consistency was higher than for pain. Although the current trial result did not confirm the efficacy of ondansetron, the size and direction of the effect were very similar, suggesting there is a genuine effect.

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Impact on stool consistency, days with loose stool and stool urgency

Despite our lack of statistical power, we did show statistically significant differences between treatment arms with improvement in stool consistency as assessed by the BSFS and reduction in days with loose stool in the ondansetron arm.

Ondansetron treatment in our trial was associated with a lower average BSFS over weeks 1–12 with a mean difference from placebo of 0.7 (0.19), $p = 0.0013$. It was also associated with fewer days per week with loose stool, a mean difference of −1.0 day (0.45), $p = 0.03$, compared with placebo. Although we showed a reduction in stool urgency in the ondansetron arm, there was no evidence of a statistically significant difference between the arms.

Dose titration

Our trial differed from most IBS trials in mimicking clinical practice and using dose titration, which prior clinical experience had suggested as optimum. We had previously reported that when patients were allowed to adjust the dose, they choose a wide range of optimum doses. A subgroup who needed very small doses were more sensitive to the drug as shown by a larger increase in transit time despite the lower dose.[12] The same study had a much-reduced incidence of constipation, just 9% compared with 33% in a recent study of alosetron, where fixed dosing was used.[48] Evidence from our study supports this with only 1/37 (2%) of our patients on ondansetron discontinuing treatment because of constipation, no different from placebo in whom one patient also discontinued because of constipation. We suggest that future trials should also use the titration method as it avoids early dropout due to unacceptable constipation.

Time course of effect

Analysis of the weekly scores showed that the effect of ondansetron on stool form was rapid, being seen in the first week in keeping with long clinical experience from prescribing this drug. Thus, 51.4% were stool consistency responders on ondansetron at week 1 compared with 16.3% on placebo. This difference was maximal in week 1 but over time differences lessened; there were no statistically significant differences between the arms over weeks 1–12. Interpretation of this was complicated by loperamide use, which was greater in the placebo group, a not unexpected feature that would however have tended to minimise treatment differences. The option of not allowing loperamide use was considered, but rejected, as it was felt that this would prove a severe barrier to recruitment and would lead to extensive dropouts from the placebo arm. Furthermore, when we performed a sensitivity analysis including loperamide use in the model, we found it made very little difference. Nearly 80% of subjects did not use loperamide and its use does complicate interpretation. A simpler alternative would be to prohibit loperamide and treat dropouts with uncontrolled diarrhoea as treatment failures.

Mode of action

Our transit studies confirmed earlier reports that ondansetron significantly slowed transit, an effect particularly noticeable in the rectosigmoid region. This was reflected clinically in the significant increase in the proportion of stool consistency responders. Unexpectedly, the water content of stool did not correlate with either the objective measure of WGTT or any of the expected symptoms, such as urgency and days with loose stool. This was despite quite reasonable numbers, especially at baseline. One problem is the erratic nature of the IBS bowel habit, so perhaps it would have been necessary to average several stools to get a more meaningful value, as has been suggested with analysis of the microbiota. The other conclusion could be that other factors such as sensitisation of pelvic nerves or central

sensitisation are more important than stool % water in determining symptoms. Regrettably for most of the remaining studies, we had far too few patients to be able to reliably define the mode of action.

The barostat study was underpowered and there was a large placebo effect but there was a tendency for ondansetron to increase the volume eliciting a sense of urgency. We calculated that we would only need 16 per group to show this difference significant at 90% power or even 10 using a crossover design, so this should probably be tested further.

Protease results showed no treatment effect, nor did we confirm our original finding that baseline protease correlated negatively with transit, although we did show that changes in transit correlated with changes in protease, supporting the concept that fast transit increases FP levels by reducing the time for protease degradation, as we had previously shown using an osmotic laxative to accelerate transit.[23] We performed an individual patient data meta-analysis of the change in transit and change in FP combining our data with the first arm of the Garsed crossover trial, which comes to a similar conclusion (Spearman's $r = 0.25$, $n = 60$, $p = 0.048$).

Sensitivity as assessed by baseline barostat pain threshold, however, did not correlate significantly with baseline FPs [Pearson's $r = 0.23$ (–0.7–0.8), $p = 0.65$], which gives no support for the idea that proteases are critical targets for future therapies.

Recognising the high prevalence of BAD in patients thought to have IBS-D, we deliberately excluded those meeting conventional criteria for BAD. Studies at the Mayo Clinic have previously reported on 74 IBS-D patients in whom BAD was excluded and showed a weak correlation with transit measured as geometric centre after 24 hours. We found a similar tendency, but this was not significant, likely due to our smaller numbers. Our values for total bile acids of 3.9 (2.1) mM were in the lower range of those reported by the Mayo group of 9.6 (7.1) mM for normal subjects.[49] None of our patients' values for concentration of total bile acids were above the mean + 2SD reported by Peleman and colleagues,[49] suggesting that we had effectively excluded BAD.

Equality, diversity and inclusion

We attempted to include all patients presenting with IBS-D using a wide range of approaches. However, the distribution of patients with 93.8% white people and only 3.8% black people and 2.5% Asian suggests failure to recruit from minorities. Future studies should explore the use of alternative ways of reaching these groups.

Recommendations for future trials

Our experience suggests that the change in referral practice over the last decade has substantially reduced referrals of IBS-D patients to secondary care. This means that, to improve recruitment, future trials should recruit mainly from primary care. We have recently had good experience using enthusiastic primary care doctors to successfully recruit 463 IBS patients to a the HTA-funded ATLANTIS trial of amitriptyline, despite the ongoing COVID pandemic.[50] This was achieved using 56 general practices across 3 regions of England. The trial was adapted to the pandemic, with as many remote processes (including data entry) as possible, other than screening bloods and informed consent, which required face-to-face visits. The barriers to recruitment could be reduced by adopting national guidelines using recommended simple screening to exclude IBD and coeliac disease [calprotectin, full blood count (FBC) and coeliac serology]. The threshold for entry could be the Rome IV clinical practice modifications with loose stools 'usually' or > 25% of the abnormal stools passed.[51] The symptom threshold would be ≥ 1 day per week with pain but not requiring an average pain > 3 out of 10, since this is not part of the Rome criteria and may exclude the moderate cases who might do best. A 1-week trial of colestyramine should

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be used to exclude BAD and the trial should use titrated ondansetron starting with 4 mg daily. Titration undoubtedly accounts for the low rate of constipation and the fact that dropouts due to constipation were very low (2%) compared to other studies on $5HT_3$RAs. The primary end point should be the responder rate, defined clinically. We followed recent commercial trials and used the FDA composite end point responder with stool consistency, frequency, urgency and abdominal pain scores as secondary end points. Using our own data, we can calculate that using the FDA responder rate, we would need 222 per arm to achieve 80% power with type I error of 5% and allowing 15% attrition would require 522 to be recruited in total. However, there are alternatives to the FDA end points. Several recent trials, particularly for treatments which do not directly affect bowel habit such as diets, have used the IBS-SSS responder rate, defining responders as those with a > 50[52] or 75[53]-point fall in IBS-SSS score, which is a more global assessment of IBS impact on QoL. Using IBS-SSS score as an end point and a responder definition of a fall > 50 points in IBS-SSS [which the originators of the IBS-SSS give as the minimal clinically important difference (MCID)], we could achieve a 90% power to detect the difference we observed (61.8% vs. 41.8%) using 306. Allowing for 15% dropout, the study would need to recruit a total of 360 patients, which might be considered more feasible for a non-commercial study. Loperamide use complicated interpretation of results with more taken on placebo likely to have reduced differences observed in stool consistency and frequency. However, at any one time only 10–15% took loperamide and if we had excluded loperamide use and taken its use as a failure of treatment, it would have made the study more powerful to detect such differences, accepting that some patients might have refused to take part.

Using Rome III criteria, and so including those with less pain or abdominal discomfort alone, would facilitate recruitment. From registration to randomisation, we lost 31/149 (20.1%) of those willing to take part because of this requirement. The logic of including those with lesser or absent abdominal pain is that the drug appears most effective at controlling loose stools and urgency. We found many patients actually had only minor pain and for them control of diarrhoea would be ample gain to justify the use of ondansetron, now that it is available in an inexpensive generic form throughout the world.

Chapter 6 Conclusions

This trial closed prematurely owing to failure to recruit. However, it did show that titrated ondansetron is well tolerated in IBS-D and does slow whole gut transit and improve stool consistency. We were underpowered to prove that ondansetron improves IBS symptoms using the FDA-recommended composite end point, but our results were consistent with other trials of ondansetron which did so. We believe the treatment should be more widely available but ensuring that this is accepted by regulatory authorities will require a further larger trial in primary care.

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Acknowledgements

We would like to thank all the members of the various committees listed below who gave their time and effort to support the trial.

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Patient and public involvement

Karen Andrews and Peter Rutherford.

Participating sites

With thanks to all the research staff at the participating centres who provided and cared for trial participants and collected trial data:

Barnsley Hospital NHS Foundation Trust (Dr Kapil Kapur), Barts and The London School of Medicine and Dentistry (Prof Qasim Aziz), County Durham and Darlington NHS Foundation Trust (Prof Yan Yiannakou), Doncaster and Bassetlaw Teaching Hospitals NHS Foundation Trust (Dr Anurag Agrawal), Leeds Teaching Hospitals NHS Trust (Prof Alex Ford), London North West Healthcare Trust (Dr Ayesha Akbar), Manchester University NHS Foundation Trust (Prof Peter Whorwell), NHS Lothian (Dr Maria Eugenicos), Nottingham University Hospitals NHS Trust (Dr Maura Corsetti), Salford Royal NHS Foundation Trust (Prof John McLaughlin), Sandwell and West Birmingham NHS Trust (Dr Nigel Trudgill), Sheffield Teaching Hospitals NHS Foundation Trust (Prof David Sanders), South Tees NHS Trust (Dr Arvind Ramadas), St Georges University Hospital NHS Trust (Dr Jamal Hayat), University College London Hospitals NHS Foundation Trust (Prof Anton Emmanuel) and University Hospitals of North Midlands NHS Trust (Dr Adam Farmer).

Participants

We are grateful to all the trial participants for their essential contribution to the trial.

All data requests should be submitted to the corresponding author for consideration and would be subject to review by a subgroup of the trial team, which will include the data guarantor, Professor Farrin. Access to anonymised data may be granted following this review. All data-sharing activities would require a data-sharing agreement.

Ethical approval

Ethical approval for the study was given by Leeds West Research Ethics Committee in November 2017 (reference number 17/YH/0262). Confirmation of capacity and capability was obtained from the recruiting centres as well as the participant identification centres in primary and secondary care trusts. The trial was registered with the International Standard Randomised Controlled Trial Register (ISRCTN) under the reference number 17508514. A summary of the changes made to the original protocol is provided in *Table 35*.

Funding

This project was funded by the National Institute for Health and Care Research (NIHR) Efficacy and Mechanism Evaluation programme (project number 15/74/01) and will be published in full in *Efficacy and Mechanism Evaluation*; Vol. 10, No. 9. See the NIHR Journals Library website for further project information.

Nottingham University Hospitals NHS Trust agreed to act as sponsor for the research and the study was adopted by the CRN.

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References

1. Sperber AD, Bangdiwala SI, Drossman DA, Ghoshal UC, Simren M, Tack J, *et al.* Worldwide prevalence and burden of functional gastrointestinal disorders, results of Rome foundation global study. *Gastroenterology* 2020;**160**:99–114.e3. https://doi.org/10.1053/j.gastro.2020.04.014
2. Hobbs FDR, Bankhead C, Mukhtar T, Stevens S, Perera-Salazar R, Holt T, Salisbury C; National Institute for Health Research School for Primary Care Research. Clinical workload in UK primary care: a retrospective analysis of 100 million consultations in England, 2007–14. *Lancet* 2016;**387**(10035):2323–30. https://doi.org/10.1016/S0140-6736(16)00620-6
3. Tillisch K, Labus JS, Naliboff BD, Bolus R, Shetzline M, Mayer EA, Chang L. Characterization of the alternating bowel habit subtype in patients with irritable bowel syndrome. *Am J Gastroenterol* 2005;**100**(4):896–904.
4. Atarodi S, Rafieian S, Whorwell PJ. Faecal incontinence-the hidden scourge of irritable bowel syndrome: a cross-sectional study. *BMJ Open Gastroenterol* 2014;**1**(1):e000002. https://doi.org/10.1136/bmjgast-2014-000002
5. Lavo B, Stenstam M, Nielsen AL. Loperamide in treatment of irritable bowel syndrome: a double-blind placebo controlled study. *Scand J Gastroenterol* 1987;**22**(Suppl. 130):77–80.
6. Hovdenak N. Loperamide treatment of the irritable bowel syndrome. *Scand J Gastroenterol* 1987;**22**(Suppl. 130):81–4.
7. Andresen V, Montori VM, Keller J, West CP, Layer P, Camilleri M. Effects of 5-hydroxytryptamine (serotonin) type 3 antagonists on symptom relief and constipation in nonconstipated irritable bowel syndrome: a systematic review and meta-analysis of randomized controlled trials. *Clin Gastroenterol Hepatol* 2008;**6**(5):545–55.
8. Fukudo S, Ida M, Akiho H, Nakashima Y, Matsueda K. Effect of ramosetron on stool consistency in male patients with irritable bowel syndrome with diarrhea. *Clin Gastroenterol Hepatol* 2014;**12**(6):953–9. https://doi.org/10.1016/j.cgh.2013.11.024
9. Fukudo S, Kinoshita Y, Okumura T, Ida M, Akiho H, Nakashima Y, *et al.* Ramosetron reduces symptoms of irritable bowel syndrome with diarrhea and improves quality of life in women. *Gastroenterology* 2016;**150**(2):358–66. https://doi.org/10.1053/j.gastro.2015.10.047
10. Gore S, Gilmore IT, Haigh CG, Brownless SM, Stockdale H, Morris AI. Colonic transit in man is slowed by ondansetron (GR38032F), a selective 5-hydroxytryptamine receptor (type 3) antagonist. *Aliment Pharmacol Ther* 1990;**4**(2):139–44.
11. Talley NJ, Phillips SF, Haddad A, Miller LJ, Twomey C, Zinsmeister AR, *et al.* GR 38032F (ondansetron), a selective 5HT3 receptor antagonist, slows colonic transit in healthy man. *Dig Dis Sci* 1990;**35**(4):477–80.
12. Garsed K, Chernova J, Hastings M, Lam C, Marciani L, Singh G, *et al.* A randomised trial of ondansetron for the treatment of irritable bowel syndrome with diarrhoea. *Gut* 2014;**63**(10):1617–25. https://doi.org/10.1136/gutjnl-2013-305989
13. Gunn D, Garsed K, Lam C, Singh G, Lingaya M, Wahl V, *et al.* Abnormalities of mucosal serotonin metabolism and 5-HT3 receptor subunit 3C polymorphism in irritable bowel syndrome with diarrhoea predict responsiveness to ondansetron. *Aliment Pharmacol Ther* 2019;**50**(5):538–46. https://doi.org/10.1111/apt.15420
14. Lembo AJ, Olden KW, Ameen VZ, Gordon SL, Heath AT, Carter EG. Effect of alosetron on bowel urgency and global symptoms in women with severe, diarrhea-predominant irritable bowel syndrome: analysis of two controlled trials. *Clin Gastroenterol Hepatol* 2004;**2**(8):675–82.

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15. Spiegel B, Strickland A, Naliboff BD, Mayer EA, Chang L. Predictors of patient-assessed illness severity in irritable bowel syndrome. *Am J Gastroenterol* 2008;**103**(10):2536–43.

16. von der Ohe MR, Camilleri M, Kvols LK. A 5HT3 antagonist corrects the postprandial colonic hypertonic response in carcinoid diarrhea. *Gastroenterology* 1994;**106**(5):1184–9.

17. von der Ohe MR, Hanson RB, Camilleri M. Serotonergic mediation of postprandial colonic tonic and phasic responses in humans. *Gut* 1994;**35**(4):536–41.

18. Stacher G, Gaupmann G, Schneider C, Stacher-Janotta G, Steiner-Mittelbach G, Abatzi TA, Steinringer H. Effects of a 5-hydroxytryptamine3 receptor antagonist (ICS 205-930) on colonic motor activity in healthy men. *Br J Clin Pharmacol* 1989;**28**(3):315–22. https://doi.org/10.1111/j.1365-2125.1989.tb05432.x

19. Clemens CH, Samsom M, Van Berge Henegouwen GP, Fabri M, Smout AJPM. Effect of alosetron on left colonic motility in non-constipated patients with irritable bowel syndrome and healthy volunteers. *Aliment Pharmacol Ther* 2002;**16**(5):993–1002. https://doi.org/10.1046/j.1365-2036.2002.01252.x

20. Stacher G, Weber U, Stacher-Janotta G, Bauer P, Huber K, Holzäpfel A, *et al.* Effects of the 5-HT3 antagonist cilansetron vs placebo on phasic sigmoid colonic motility in healthy man: a double-blind crossover trial. *Br J Clin Pharmacol* 2000;**49**(5):429–36.

21. Lin AY, Du P, Dinning PG, Arkwright JW, Kamp JP, Cheng LK, *et al.* High-resolution anatomic correlation of cyclic motor patterns in the human colon: evidence of a rectosigmoid brake. *Am J Physiol Gastrointest Liver Physiol* 2017;**312**(5):G508–15. https://doi.org/10.1152/ajpgi.00021.2017

22. Lin AY, Dinning PG, Milne T, Bissett IP, O'Grady G. The 'rectosigmoid brake': review of an emerging neuromodulation target for colorectal functional disorders. *Clin Exp Pharmacol Physiol* 2017;**44**(7):719–28. https://doi.org/10.1111/1440-1681.12760

23. Tooth D, Garsed K, Singh G, Marciani L, Lam C, Fordham I, *et al.* Characterisation of faecal protease activity in irritable bowel syndrome with diarrhoea: origin and effect of gut transit. *Gut* 2014;**63**(5):753–60. https://doi.org/10.1136/gutjnl-2012-304042

24. Gecse K, Roka R, Ferrier L, Leveque M, Eutamene H, Cartier C, *et al.* Increased faecal serine protease activity in diarrhoeic IBS patients: a colonic lumenal factor impairing colonic permeability and sensitivity. *Gut* 2008;**57**(5):591–9.

25. Edwards CA, Brown S, Baxter AJ, Bannister JJ, Read NW. Effect of bile acid on anorectal function in man. *Gut* 1989;**30**(3):383–6.

26. Bajor A, Tornblom H, Rudling M, Ung K-A, Simrén M. Increased colonic bile acid exposure: a relevant factor for symptoms and treatment in IBS. *Gut* 2015;**64**(1):84–92. https://doi.org/10.1136/gutjnl-2013-305965

27. Shiotani A, Kusunoki H, Ishii M, Imamura H, Manabe N, Kamada T, *et al.* Pilot study of Biomarkers for predicting effectiveness of ramosetron in diarrhea-predominant irritable bowel syndrome: expression of S100A10 and polymorphisms of TPH1. *Neurogastroenterol Motil* 2015;**27**(1):82–91. https://doi.org/10.1111/nmo.12473

28. Francis CY, Morris J, Whorwell PJ. The irritable bowel severity scoring system: a simple method of monitoring irritable bowel syndrome and its progress. *Aliment Pharmacol Ther* 1997;**11**(2):395–402.

29. Fraser A, Delaney BC, Ford AC, Qume M, Moayyedi P. The Short-Form Leeds Dyspepsia Questionnaire validation study. *Aliment Pharmacol Ther* 2007;**25**(4):477–86. https://doi.org/10.1111/j.1365-2036.2006.03233.x

30. Hahn BA, Kirchdoerfer LJ, Fullerton S, Mayer E. Patient-perceived severity of irritable bowel syndrome in relation to symptoms, health resource utilization and quality of life. *Aliment Pharmacol Ther* 1997;**11**(3):553–9.

31. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. *Acta Psychiatr Scand* 1983;**67**(6):361–70.

32. Spiller RC, Humes DJ, Campbell E, Hastings M, Neal KR, Dukes GE, Whorwell PJ. The Patient Health Questionnaire 12 Somatic Symptom scale as a predictor of symptom severity and consulting behaviour in patients with irritable bowel syndrome and symptomatic diverticular disease. *Aliment Pharmacol Ther* 2010;**32**(6):811–20.

33. Törnblom H, Van OL, Sadik R, Abrahamsson H, Tack J, Simrén M. Colonic transit time and IBS symptoms: what's the link? *Am J Gastroenterol* 2012;**107**(5):754–60. https://doi.org/10.1038/ajg.2012.5

34. Dinning PG, Wiklendt L, Maslen L, Gibbins I, Patton V, Arkwright JW, *et al.* Quantification of in vivo colonic motor patterns in healthy humans before and after a meal revealed by high-resolution fiber-optic manometry. *Neurogastroenterol Motil* 2014;**26**(10):1443–57. https://doi.org/10.1111/nmo.12408

35. Corsetti M, Thys A, Harris A, Pagliaro G, Deloose E, Demedts I, Tack J. High-resolution manometry reveals different effect of polyethylene glycol, bisacodyl, and prucalopride on colonic motility in healthy subjects: an acute, open label, randomized, crossover, reader-blinded study with potential clinical implications. *Neurogastroenterol Motil* 2020;**33**(5):e14040. https://doi.org/10.1111/nmo.14040

36. Corsetti M, Pagliaro G, Demedts I, Deloose E, Gevers A, Scheerens C, *et al.* Pan-colonic pressurizations associated with relaxation of the anal sphincter in health and disease: a new colonic motor pattern identified using high-resolution manometry. *Am J Gastroenterol* 2017;**112**(3):479–89. https://doi.org/10.1038/ajg.2016.341

37. Wiklendt L, Costa M, Scott MS, Brookes SJH, Dinning PG. Automated analysis using a Bayesian functional mixed-effects model with Gaussian process responses for wavelet spectra of spatio-temporal colonic manometry signals. *Front Physiol* 2020;**11**:605066. https://doi.org/10.3389/fphys.2020.605066

38. Bradford MM. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem* 1976;**72**:248–54.

39. FDA. *Irritable Bowel Syndrome – Clinical Evaluation of Products for Treatment*. Silver Spring, MD: FDA; 2012.

40. Delvaux M, Louvel D, Mamet JP, Campos-Oriola R, Frexinos J. Effect of alosetron on responses to colonic distension in patients with irritable bowel syndrome. *Aliment Pharmacol Ther* 1998;**12**(9):849–55.

41. Schulz KF, Altman DG, Moher D. *CONSORT* 2010 statement: updated guidelines for reporting parallel group randomised trials. *PLOS Med* 2010;**7**(3):e1000251. https://doi.org/10.1371/journal.pmed.1000251

42. Turvill J, O'Connell S, Brooks A, Bradley-Wood K, Laing J, Thiagarajan S, *et al.* Evaluation of a faecal calprotectin care pathway for use in primary care. *Prim Health Care Res Dev* 2016;**17**(5):428–36. https://doi.org/10.1017/S1463423616000049

43. National Institute for Health and Care Excellence. *Implementing Faecal Calprotectin Testing in Primary Care 2015*. 2022. URL: https://www.nice.org.uk/sharedlearning/implementing-faecal-calprotectin-testing-in-primary-care (accessed 3 August 2022).

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44. Jakobsen JC, Gluud C, Wetterslev J, Winkel P. When and how should multiple imputation be used for handling missing data in randomised clinical trials – a practical guide with flowcharts. *BMC Med Res Methodol* 2017;**17**(1):162. https://doi.org/10.1186/s12874-017-0442-1

45. Bang H, Park JJ. Blinding in clinical trials: a practical approach. *J Altern Complement Med* 2013;**19**(4):367–9. https://doi.org/10.1089/acm.2012.0210

46. Lam C, Tan W, Leighton M, Hastings M, Lingaya M, Falcone Y, *et al.* A mechanistic multicentre, parallel group, randomised placebo-controlled trial of mesalazine for the treatment of IBS with diarrhoea (IBS-D). *Gut* 2016;**65**(1):91–9. https://doi.org/10.1136/gutjnl-2015-309122

47. Plasse TF, Barton G, Davidson E, *et al.* Bimodal release ondansetron improves stool consistency and symptomatology in diarrhea-predominant irritable bowel syndrome: a randomized, double-blind, trial. *Am J Gastroenterol* 2020;**11**:1466–73.https://doi.org/10.14309/ajg.0000000000000727

48. Olden KW, Chey WD, Shringarpure R, Nicandro JP, Chuang E, Earnest DL. Alosetron versus traditional pharmacotherapy in clinical practice: effects on resource use, health-related quality of life, safety and symptom improvement in women with severe diarrhea-predominant irritable bowel syndrome. *Curr Med Res Opin* 2019;**35**(3):461–72. https://doi.org/10.1080/03007995.2018.1533456

49. Peleman C, Camilleri M, Busciglio I, Burton D, Donato L, Zinsmeister AR. Colonic transit and bile acid synthesis or excretion in patients with irritable bowel syndrome-diarrhea without bile acid malabsorption. *Clin Gastroenterol Hepatol* 2017;**15**(5):720–7. https://doi.org/10.1016/j.cgh.2016.11.012

50. Alderson SL, Wright-Hughes A, Ford AC, Farrin A, Hartley S, Fernandez C, *et al.* Amitriptyline at low-dose and titrated for irritable bowel syndrome as second-line treatment (The ATLANTIS trial): protocol for a randomised double-blind placebo-controlled trial in primary care. *Trials* 2022;**23**(1):552. https://doi.org/10.1186/s13063-022-06492-6

51. Drossman DA, Tack J. Rome foundation clinical diagnostic criteria for disorders of gut-brain interaction. *Gastroenterology* 2022;**162**(3):675–9. https://doi.org/10.1053/j.gastro.2021.11.019

52. Goyal O, Batta S, Nohria S, Kishore H, Goyal P, Sehgal R, Sood A. Low fermentable oligosaccharide, disaccharide, monosaccharide, and polyol diet in patients with diarrhea-predominant irritable bowel syndrome: a prospective, randomized trial. *J Gastroenterol Hepatol* 2021;**36**:2107–15. https://doi.org/10.1111/jgh.15410

53. Serrano-Falcon B, Delgado-Aros S, Mearin F, Ciriza de Los Ríos C, Serra J, Mínguez M, *et al.* Clinical response to linaclotide at week 4 predicts sustained response in irritable bowel syndrome with constipation and improvements in digestive and extra-digestive symptoms. *Therap Adv Gastroenterol* 2019;**12**:9857358. https://doi.org/10.1177/1756284819857358

Appendix 1 Activities per visit

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TABLE 34 Participant timeline

Time point	Week 12	Week 10	Weeks 8–11	Week 0	Week 6	Weeks 8–11	Week 12	Week 16
Activity/assessment	**Visit 1: Registration**	**Visit 2: Eligibility confirmation**	**Mechanistic assessments**	**Visit 3: Randomisation**	**Visit 4: 6 week visit**	**Mechanistic assessments**	**Visit 5: 12 week visit**	**Visit 6: Follow-up visit**
	Enrolment							
Informed consent	X							
Demographics[a]	X							
Vital signs	X						X	
Haematology and biochemistry[b]	X						X	
12 Lead ECG	X							
Pregnancy test		X		X	X		X	X
Confirmation of eligibility		X						
Randomisation				X				
	Interventions							
Daily ondansetron or placebo[c]								
	Assessments							
Symptom diary start[d]	X							
SMS messages start[e]	X							
Questionnaires[f]				X			X	
Colonic transit assessment				X			X	
Research biopsies[g]				X				
Research blood[h]				X			X	
Stool samples[i]				X			X	

TABLE 34 Participant timeline (*continued*)

Time point	Week 12	Week 10	Weeks 8–11	Week 0	Week 6	Weeks 8–11	Week 12	Week 16
Activity/assessment	Visit 1: Registration	Visit 2: Eligibility confirmation	Mechanistic assessments	Visit 3: Randomisation	Visit 4: 6 week visit	Mechanistic assessments	Visit 5: 12 week visit	Visit 6: Follow-up visit
High-resolution colonic manometry[j]			X			X		
Barostat assessment[k]			X			X		
Exit poll								X

CRP, C-reactive protein; LFTs, liver function tests; QMUL, Queen Mary University of London; UCL is University College London; UEs, urea and electrolytes; WBC, white blood count
a Date of birth, NHS number, address, telephone number (if consented to text messages), smoking history, height, weight.
b WBC, FBC, LFTs, UEs, CRP.
c Titrated to optimum dose during the first 2 weeks.
d Recording worst abdominal pain (0–100), worst urgency (0–100), number of IMP taken, use of loperamide, stool consistency of each stool passed and relief from IBS symptoms at the end of each week.
e Asking if they have passed a stool type 6 or 7, and what their worst abdominal pain score was that day.
f At baseline only: PHQ-12; at baseline and visit: IBS-SSS, SFLDQ, HADS and IBS-QOL.
g Six high rectal biopsies taken either during mechanistic assessment or at visit 3.
h 5 ml whole blood at visit 3; 5 ml serum at visit 3 and 5.
i Four aliquots, taken and frozen at home prior to visit.
j Performed at University of Nottingham and QMUL only.
k Performed at Nottingham/Leeds/QMUL/UCL only.

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TABLE 35 Summary of protocol changes

	Date
To allow all participants to consent to optional biopsy collection not just those consenting to mechanistic studies	22 February 2018
Including patients with a prolonged QT interval or taking drugs known to cause this with caution	
Change of eligibility criteria regarding QTc interval from > 420 milliseconds to > 450 milliseconds for men and > 470 milliseconds for women	12 June 2018
Change of maximum allowed time period between consent and the eligibility confirmation visit from 4 to 8 weeks	
Change from requirement to use double contraceptives to conventional contraception	6 November 2018
Addition of a trial of bile acid binding agent as an alternative to a SeHCAT scan to exclude BDA as an alternative diagnosis	
Allow patients with minor rises in ALT (< 2 × upper limit of normal) to be included	
Allow patients assessed as ineligible to be rescreened at a later date	
Increase the time since colonoscopy required for eligibility from 5 to 10 years with stable symptoms and a normal faecal calprotectin	19 July 2019
Patients taking high-dose TCAs need to stop taking them to participate in the trial	
Extension of the recruitment period from 18 to 20 months	
Change to allow patients with pulse, BP or laboratory blood values that are out of normal range but deemed not clinically significant to enter the trial	25 September 2019
Addition of information about new safety information on the use of ondansetron in pregnancy and the risk of orofacial malformations	October 2019

Appendix 2 Quantification of bile acids (LC–MS)

After thawing, 0.5 g of the faecal sample was suspended in 2 ml of 50% (w/v) acetonitrile and extracted by vortexing and sonication for 10 minutes. The suspension was centrifuged twice at 25,000*g* for 20 minutes. Supernatants were transferred to sample vials and loaded onto a LC–MS system containing online SPE. At the beginning of each analysis 50 µl of the sample were transferred to SPE column at a flow of 0.1 ml/min with the loading mobile phase, aqueous 5% acetonitrile, 0.1% formic acid and 0.02% trifluoroacetic acid. The chromatographic separation was performed using a binary system with pumps (A) and (B) (Jasco, PU-2085 Plus, Cremella, Italy) connected to a degasser (Alltech, degasser, Stamford, UK). The two systems were connected using a two-position, six-port valve, used to switch automatically from loading (position 1) to injection (position 2) after 9 minutes (Valvemate, Gilson, Dunstable, UK). Samples were analysed on a Waters Ex-bridge C18 column (Waters, 100 × 2.1 mm; 3.5 µm particle size), using a gradient program. Mobile Phase (A) 5 mM ammonium acetate, 0.1% ammonium hydroxide, mobile phase (B) 100% acetonitrile. Initial composition was 80% (A), which was reduced to 70% (A) over 30 minutes with further reduction to 65% (A) over the next 3 minutes. The eluent composition was held at 65% for 1.5 minutes before returning to 80% (A) initial condition over the next 1.5 minutes. Flow rate was 0.2 ml/min.

High-performance liquid chromatography was coupled in series with the turbo ion-spray (ESI) source of the tandem mass spectrometer (Micromass, Manchester, UK). Electrospray ionisation was performed in negative mode with nitrogen as the nebuliser gas. Detection of individual bile acids was performed using SIM mode. Additional structural information was obtained via tandem MS (MS/MS) fragmentation, with collision energies ranging from 15 to 30 electron volts. Data were acquired using software MassLynX (Waters, Wexford, Ireland).

The concentration of bile acids in the samples was determined on the basis of the peak areas of individual bile acids and external standards.

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Appendix 3 Screening summary

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TABLE 36 Screening identification method by site

Site	Total *N* (%)	Identification method							
		Secondary care (%)	Primary care and pharmacy (%)	Self-referral via website (%)	Self-referral other (%)	Secondary PIC (%)	Primary care invitation (%)	Other identification method (%)	Missing (%)
James Cook, South Tees	38 (2.4)	26 (68.4)	0 (0.0)	3 (7.9)	0 (0.0)	0 (0.0)	0 (0.0)	9 (23.7)	0 (0.0)
St James's University Hospital, Leeds	80 (5.1)	19 (23.8)	0 (0.0)	1 (1.3)	8 (10.0)	0 (0.0)	0 (0.0)	51 (63.8)	1 (1.3)
Royal Hallamshire Hospital	51 (3.2)	20 (39.2)	0 (0.0)	1 (2.0)	0 (0.0)	0 (0.0)	0 (0.0)	29 (56.9)	1 (2.0)
Sandwell Hospital	43 (2.7)	18 (41.9)	0 (0.0)	4 (9.3)	3 (7.0)	0 (0.0)	0 (0.0)	16 (37.2)	2 (4.7)
University Hospital of North Durham	23 (1.5)	9 (39.1)	0 (0.0)	12 (52.2)	0 (0.0)	0 (0.0)	0 (0.0)	2 (8.7)	0 (0.0)
Wythenshawe Hospital	12 (0.8)	3 (25.0)	3 (25.0)	4 (33.3)	2 (16.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Royal Stoke University Hospital	44 (2.8)	29 (65.9)	0 (0.0)	0 (0.0)	0 (0.0%)	0 (0.0)	0 (0.0)	14 (31.8)	1 (2.3)
Nottingham University Hospital	58 (3.7)	23 (39.7)	5 (8.6)	10 (17.2)	6 (10.3)	0 (0.0)	2 (3.4)	11 (19.0)	1 (1.7)
Barnsley District General Hospital	216 (13.7)	195 (90.3)	0 (0.0)	0 (0.0)	7 (3.2)	0 (0.0)	0 (0.0)	1 (0.5)	13 (6.0)
St Mark's Hospital	11 (0.7)	5 (45.5)	0 (0.0)	5 (45.5)	0 (0.0)	0 (0.0)	0 (0.0)	1 (9.1)	0 (0.0)
Western General Hospital	53 (3.4)	42 (79.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	9 (17.0)	2 (3.8)
Salford Royal Hospital	16 (1.0)	2 (12.5)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	14 (87.5)	0 (0.0)
Barts and London School of Medicine and Dentistry	159 (10.1)	67 (42.1)	2 (1.3)	17 (10.7)	1 (0.6)	4 (2.5)	0 (0.0)	67 (42.1)	1 (0.6)
University College London Hospital	778 (49.2)	28 (3.6)	0 (0.0)	7 (0.9)	6 (0.8)	0 (0.0)	0 (0.0)	731 (94.0)	6 (0.8)
Total	**1582 (100)**	**486 (30.7)**	**10 (0.6)**	**64 (4.0)**	**33 (2.1)**	**4 (0.3)**	**2 (0.1)**	**955 (60.4)**	**28 (1.8)**

TABLE 37 Eligibility status of screened patients

Site	Total *N*	Eligibility status			
		Eligible (%)	Not eligible (%)	Further investigation required (%)	Missing (%)
James Cook, South Tees	38	10 (26.3)	12 (31.6)	16 (42.1)	0 (0.0)
St James's University Hospital, Leeds	80	15 (18.8)	56 (70.0)	9 (11.3)	0 (0.0)
Royal Hallamshire Hospital	51	12 (23.5)	35 (68.6)	1 (2.0)	3 (5.9)
Sandwell Hospital	43	7 (16.3)	24 (55.8)	9 (20.9)	3 (7.0)
University Hospital of North Durham	23	4 (17.4)	3 (13.0)	16 (69.6)	0 (0.0)
Wythenshawe Hospital	12	11 (91.7)	0 (0.0)	1 (8.3)	0 (0.0)
Royal Stoke University Hospital	44	20 (45.5)	24 (54.5)	0 (0.0)	0 (0.0)
Nottingham University Hospital	58	41 (70.7)	15 (25.9)	1 (1.7)	1 (1.7)
Barnsley District General Hospital	216	29 (13.4)	176 (81.5)	0 (0.0)	11 (5.1)
St Mark's Hospital	11	4 (36.4)	3 (27.3)	3 (27.3)	1 (9.1)
Western General Hospital	53	5 (9.4)	20 (37.7)	28 (52.8)	0 (0.0)
Salford Royal Hospital	16	1 (6.3)	15 (93.8)	0 (0.0)	0 (0.0)
Barts and London School of Medicine and Dentistry	159	38 (23.9)	105 (66.0)	14 (8.8)	2 (1.3)
University College London Hospital	778	5 (0.6)	761 (97.8)	0 (0.0)	12 (1.5)
Total	**1582**	**202 (12.8)**	**1249 (79.0)**	**98 (6.2)**	**33 (2.1)**

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TABLE 38 Consent status by site

Site	Total *N*	Consented to study			If not consented, reasons for non-consent			
		Yes (%)	No (%)	Status missing (%)	Refused consent (%)	Cannot get hold of patient (%)	Other reason (%)	Missing (%)
James Cook, South Tees	38	10 (26.3)	16 (42.1)	12 (31.6)	9 (56.3)	1 (6.3)	5 (31.3)	1 (6.3)
St James's University Hospital, Leeds	80	15 (18.8)	9 (11.3)	56 (70.0)	1 (11.1)	5 (55.6)	3 (33.3)	0 (0.0)
Royal Hallamshire Hospital	51	12 (23.5)	1 (2.0)	38 (74.5)	0 (0.0)	0 (0.0)	1 (100.0)	0 (0.0)
Sandwell Hospital	43	9 (20.9)	7 (16.3)	27 (62.8)	5 (71.4)	1 (14.3)	1 (14.3)	0 (0.0)
University Hospital of North Durham	23	11 (47.8)	9 (39.1)	3 (13.0)	4 (44.4)	4 (44.4)	1 (11.1)	0 (0.0)
Wythenshawe Hospital	12	12 (100.0%)	0 (0.0)	0 (0.0)				
Royal Stoke University Hospital	44	18 (40.9)	2 (4.5)	24 (54.5)	0 (0.0)	0 (0.0)	2 (100.0)	0 (0.0)
Nottingham University Hospital	58	42 (72.4)	0 (0.0)	16 (27.6)				
Barnsley District General Hospital	216	8 (3.7)	21 (9.7)	187 (86.6)	8 (38.1)	9 (42.9)	4 (19.0)	0 (0.0)
St Mark's Hospital	11	1 (9.1)	6 (54.5)	4 (36.4)	5 (83.3)	0 (0.0)	1 (16.7)	0 (0.0)
Western General Hospital	53	7 (13.2)	26 (49.1)	20 (37.7)	7 (26.9)	2 (7.7)	17 (65.4)	0 (0.0)
Salford Royal Hospital	16	1 (6.3)	0 (0.0)	15 (93.8)				
Barts and London School of Medicine and Dentistry	159	23 (14.5)	21 (13.2)	115 (72.3)	13 (61.9)	2 (9.5)	6 (28.6)	0 (0.0)
University College London Hospital	778	4 (0.5)	1 (0.1)	773 (99.4)	1 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)
Total	**1582**	**173 (10.9)**	**119 (7.5)**	**1290 (81.5)**	**53 (44.5)**	**24 (20.2)**	**41 (34.5)**	**1 (0.8)**

TABLE 39 Registration status by site

Site	Total *N*	Registered to study			Reasons for non-registration	
		Yes (%)	No (%)	Status missing (%)	No longer eligible (%)	Other reason (%)
James Cook, South Tees	38	7 (15.8)	4 (10.5)	28 (73.7)	1 (25.0)	3 (75.0)
St James's University Hospital, Leeds	80	13 (16.3)	2 (2.5)	65 (81.3)	2 (100.0)	0 (0.0)
Royal Hallamshire Hospital	51	12 (23.5)	0 (0.0)	39 (76.5)		
Sandwell Hospital	43	6 (14.0)	3 (7.0)	34 (79.1)	2 (66.7)	1 (33.3)
University Hospital of North Durham	23	4 (17.4)	7 (30.4)	12 (52.2)	5 (71.4)	2 (28.6)
Wythenshawe Hospital	12	12 (100.0)	0 (0.0)	0 (0.0)		
Royal Stoke University Hospital	44	18 (40.9)	0 (0.0)	26 (59.1)		
Nottingham University Hospital	58	39 (67.2)	3 (5.2)	16 (27.6)	0 (0.0)	3 (100.0)
Barnsley District General Hospital	216	7 (3.2)	1 (0.5)	208 (96.3)	1 (100.0)	0 (0.0)
St Mark's Hospital	11	1 (9.1)	0 (0.0)	10 (90.9)		
Western General Hospital	53	6 (11.3)	1 (1.9)	46 (86.8)	0 (0.0)	1 (100.0)
Salford Royal Hospital	16	1 (6.3)	0 (0.0)	15 (93.8)		
Barts and London School of Medicine and Dentistry	159	19 (11.9)	2 (1.3)	138 (86.8)	1 (50.0)	0 (0.0)
University College London Hospital	778	4 (0.5)	0 (0.0)	774 (99.5)		
Total	**1582**	**149 (9.4)**	**23 (1.5)**	**1411 (89.2)**	**12 (52.2)**	**10 (43.5)**

TABLE 40 Pre-treatment diary scores by site

Site	Total *N* (%)	Abdominal pain score	Days with loose stool	Urgency score	
		Mean score (0–100) (SD)	Mean days (per week) (SD)	Mean urgency score (0–100) (SD)	Urgency score missing
James Cook, South Tees	4 (5.0)	66.2 (23.4)	6.1 (1.0)	69.9 (18.8)	0
St James's University Hospital, Leeds	10 (12.5)	66.3 (19.5)	5.8 (1.3)	76.0 (17.5)	0
Royal Hallamshire Hospital	4 (5.0)	48.4 (11.9)	4.9 (1.1)	55.5 (11.6)	0
Sandwell Hospital	3 (3.8)	61.6 (4.6)	5.1 (1.6)	49.3 (12.9)	0
University Hospital of North Durham	2 (2.5)	71.4 (7.1)	6.3 (1.1)	76.4 (17.2)	0
Wythenshawe Hospital	6 (7.5)	43.5 (5.8)	6.2 (0.6)	60.7 (13.9)	0
Royal Stoke University Hospital	11 (13.8)	59.6 (19.1)	6.2 (0.8)	63.4 (18.9)	1

continued

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TABLE 40 Pre-treatment diary scores by site (*continued*)

Site	Total *N* (%)	Abdominal pain score	Days with loose stool	Urgency score	
		Mean score (0–100) (SD)	Mean days (per week) (SD)	Mean urgency score (0–100) (SD)	Urgency score missing
Nottingham University Hospital	21 (26.3)	54.4 (10.2)	5.2 (1.4)	58.7 (15.5)	0
Barnsley District General Hospital	2 (2.5)	55.4 (20.7)	6.3 (1.1)	65.9 (6.4)	0
St Mark's Hospital	1 (1.3)	98.6 (.)	7.0 (.)	96.1 (.)	0
Western General Hospital	6 (7.5)	56.3 (29.4)	6.0 (1.4)	60.0 (22.3)	0
Barts and London School of Medicine and Dentistry	9 (11.3)	63.0 (21.4)	5.1 (1.6)	68.6 (27.6)	0
University College London Hospital	1 (1.3)	21.7 (.)	5.0 (.)	39.2 (.)	0
Total	**80 (100)**	**58.0 (18.3)**	**5.6 (1.3)**	**63.8 (18.9)**	**1**

TABLE 41 Demographic data by site

Site	Total *N* (%)	Age	Gender		Ethnicity		
		Mean age (years) (SD)	Male (%)	Female (%)	White (%)	Black (%)	Asian (%)
James Cook, South Tees	4 (5.0)	38.3 (12.1)	1 (25.0)	3 (75.0)	4 (100.0)	0 (0.0)	0 (0.0)
St James's University Hospital, Leeds	10 (12.5)	37.3 (12.6)	3 (30.0)	7 (70.0)	10 (100.0)	0 (0.0)	0 (0.0)
Royal Hallamshire Hospital	4 (5.0)	36.0 (15.7)	2 (50.0)	2 (50.0)	4 (100.0)	0 (0.0)	0 (0.0)
Sandwell Hospital	3 (3.8)	44.3 (6.5)	2 (66.7)	1 (33.3)	2 (66.7)	0 (0.0)	1 (33.3)
University Hospital of North Durham	2 (2.5)	53.0 (4.2)	0 (0.0)	2 (100.0)	2 (100.0)	0 (0.0)	0 (0.0)
Wythenshawe Hospital	6 (7.5)	43.5 (15.4)	3 (50.0)	3 (50.0)	6 (100.0)	0 (0.0)	0 (0.0)
Royal Stoke University Hospital	11 (13.8)	42.9 (19.4)	3 (27.3)	8 (72.7)	11 (100.0)	0 (0.0)	0 (0.0)
Nottingham University Hospital	21 (26.3)	45.0 (19.4)	10 (47.6)	11 (52.4)	20 (95.2)	1 (4.8)	0 (0.0)
Barnsley District General Hospital	2 (2.5)	50.0 (12.7)	1 (50.0)	1 (50.0)	2 (100.0)	0 (0.0)	0 (0.0)
St Mark's Hospital	1 (1.3)	55.0 (.)	0 (0.0)	1 (100.0)	1 (100.0)	0 (0.0)	0 (0.0)
Western General Hospital	6 (7.5)	39.5 (13.8)	3 (50.0)	3 (50.0)	5 (83.3)	1 (16.7)	0 (0.0)
Barts and London School of Medicine and Dentistry	9 (11.3)	52.9 (13.6)	5 (55.6)	4 (44.4)	7 (77.8)	1 (11.1)	1 (11.1)
University College London Hospital	1 (1.3)	61.0 (.)	0 (0.0)	1 (100.0)	1 (100.0)	0 (0.0)	0 (0.0)
Total	**80 (100)**	**43.9 (16.0)**	**33 (41.3)**	**47 (58.8)**	**75 (93.8)**	**3 (3.8)**	**2 (2.5)**

TABLE 42 Mechanistic test uptake by site

Site	Total *N* (%)	Barostat undertaken (%)	Colonic manometry (%)
James Cook, South Tees	4 (5.0)	0 (0.0)	0 (0.0)
St James's University Hospital, Leeds	10 (12.5)	0 (0.0)	0 (0.0)
Royal Hallamshire Hospital	4 (5.0)	0 (0.0)	0 (0.0)
Sandwell Hospital	3 (3.8)	1 (33.3)	1 (33.3)
University Hospital of North Durham	2 (2.5)	0 (0.0)	0 (0.0)
Wythenshawe Hospital	6 (7.5)	0 (0.0)	0 (0.0)
Royal Stoke University Hospital	11 (13.8)	3 (27.3)	3 (27.3)
Nottingham University Hospital	21 (26.3)	9 (42.9)	5 (23.8)
Barnsley District General Hospital	2 (2.5)	0 (0.0)	0 (0.0)
St Mark's Hospital	1 (1.3)	0 (0.0)	0 (0.0)
Western General Hospital	6 (7.5)	0 (0.0)	0 (0.0)
Barts and London School of Medicine and Dentistry	9 (11.3)	4 (44.4)	3 (33.3)
University College London Hospital	1 (1.3)	1 (100.0)	1 (100.0)
Total	**80 (100)**	**18 (22.5)**	**13 (16.3)**

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Appendix 4 Treatment discontinuation

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TABLE 43 Treatment discontinuation listing

Patient	Allocation	Who requested discontinuation	Last dose (days post randomisation)	Reason for discontinuation	Further information	Withdrawn from trial follow-up
1	Ondansetron	Clinician	18	Other (patient not eligible)	Patient not eligible as per section 8.5 of protocol as unable to discontinue Buscopan	Yes
2	Ondansetron	Patient	26	Drug intolerance	Became very constipated and found it intolerable to continue	Yes
3	Ondansetron	Patient	27	Patient decision	Side effects	Yes
4	Ondansetron	Clinician	28	Patient's health compromised		No
5	Placebo	Patient	12	Drug intolerance	Severe constipation. The participant discontinued treatment but did not withdraw from the trial	No

Appendix 5 Responders versus loperamide use by week

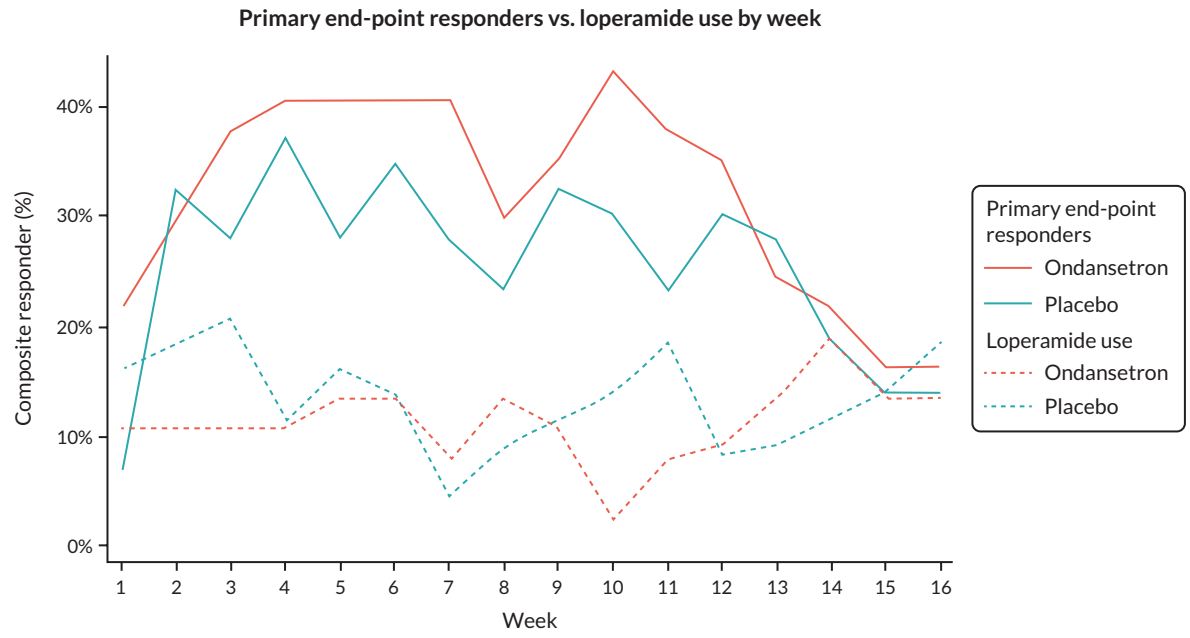


FIGURE 28 Overall responders vs. loperamide use by week.

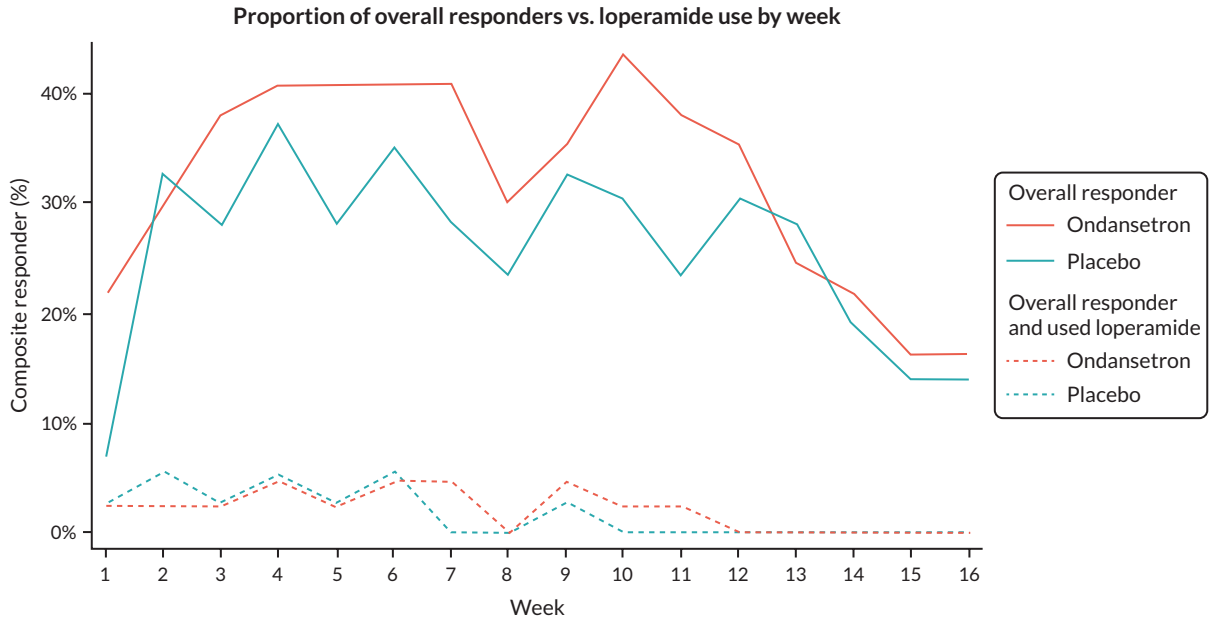


FIGURE 29 Proportion of responders vs. loperamide use by week.

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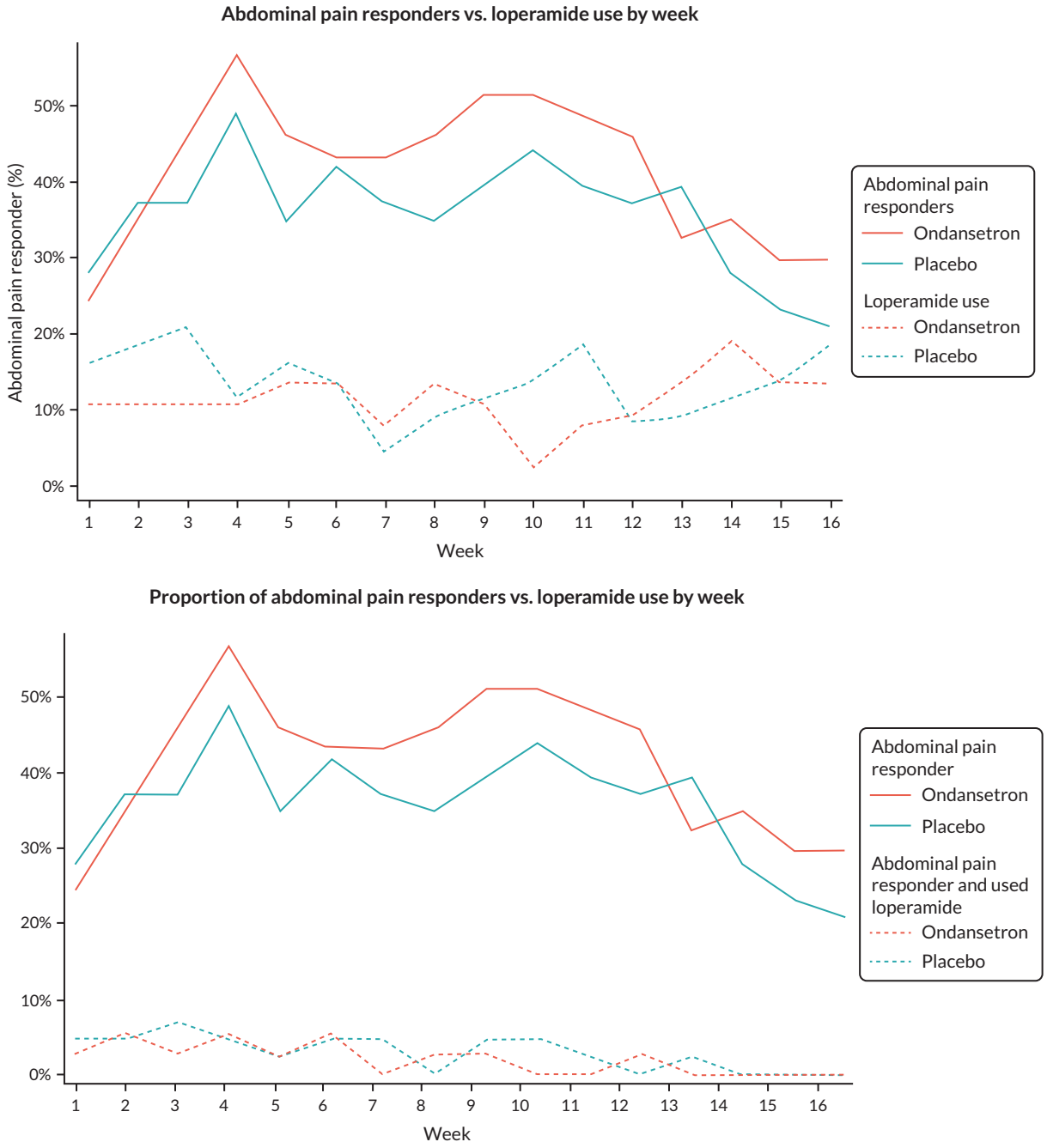


FIGURE 30 Weekly abdominal pain responders and loperamide use.

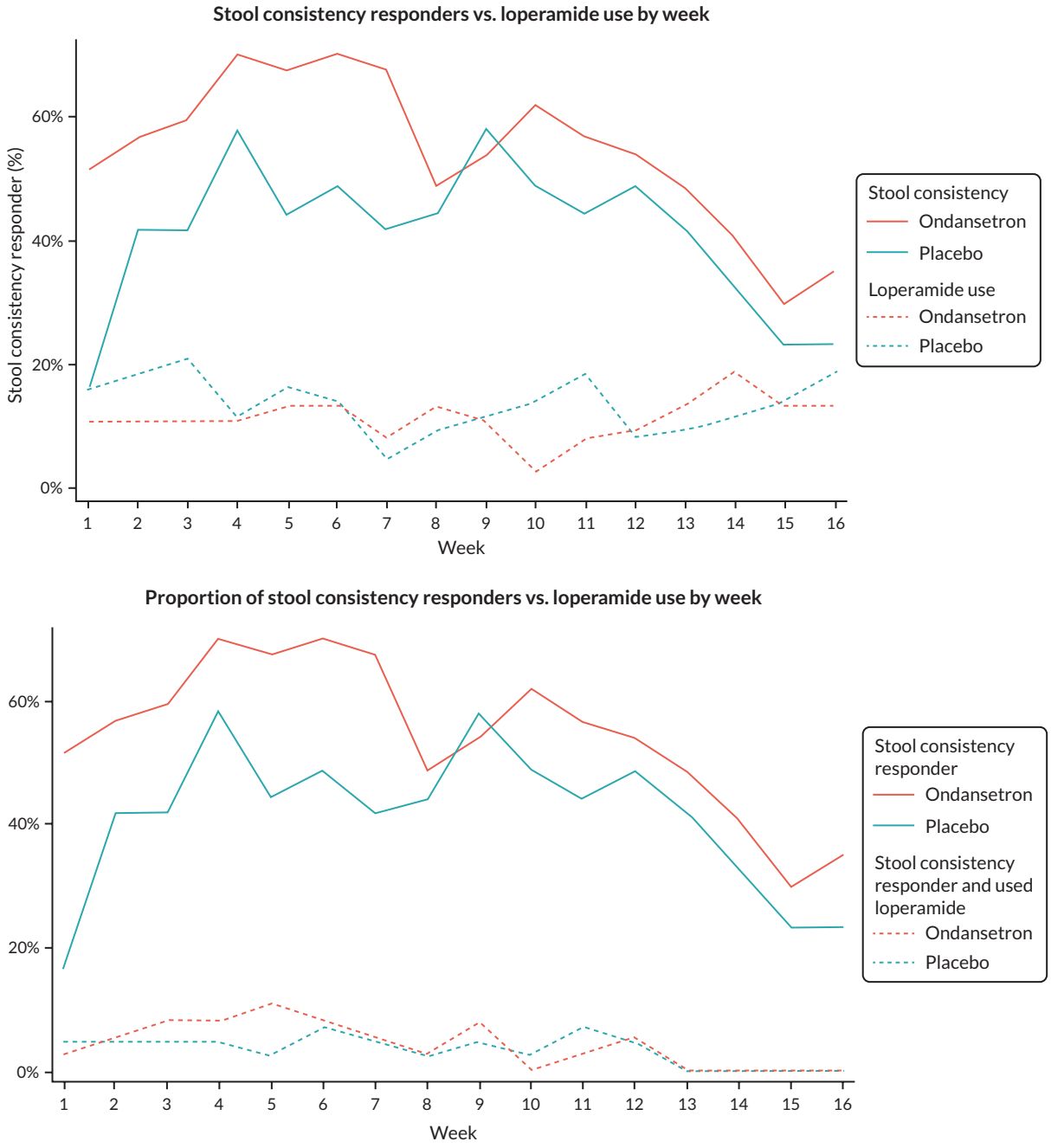


FIGURE 31 Weekly stool consistency responders vs. loperamide use.

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Appendix 6 Additional outcome summaries

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TABLE 44 Changes in mean scores compared to baseline

	Weeks 1–12			Weeks 9–12			Off-treatment weeks 13–16		
Change in pain score	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Mean (SD) missing	−15.6 (17.30) 2	−12.7 (16.44) 1	−14.0 (16.78) 3	−17.3 (19.56) 3	−14.8 (19.16) 4	−16.0 (19.26) 7	−12.1 (18.55) 10	−9.7 (17.88) 4	−10.7 (18.06) 14
Mean (95% CI)	−15.6 (−21.3, −9.8)	−12.7 (−17.6, −7.7)	−14.0 (−17.7, −10.2)	−17.3 (−23.9, −10.8)	−14.8 (−20.8, −8.8)	−16.0 (−20.4, −11.5)	−12.1 (−19.1, −5.1)	−9.7 (−15.3, −4.1)	−10.7 (−15.0, −6.3)

Note
Abdominal pain score scale from 0 to 100, 0 = no pain, 100 = worst imaginable pain.

TABLE 45 Hospital anxiety and depression scale summaries by time point and treatment arm

	Baseline			12 weeks		
HADS scores	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Anxiety score (continuous)						
Mean (SD) missing	9.9 (5.01) 0	10.2 (4.54) 0	10.1 (4.74) 0	8.4 (5.40) 5	9.3 (4.83) 3	8.9 (5.08) 8
Median (range)	9.0 (0.0–19.0)	10.0 (2.0–19.0)	9.5 (0.0–19.0)	8.0 (1.0–19.0)	9.0 (1.0–18.0)	8.5 (1.0–19.0)
Depression score (continuous)						
Mean (SD) missing	8.0 (4.16) 0	6.9 (4.00) 0	7.4 (4.09) 0	6.0 (5.43) 5	5.7 (4.19) 3	5.8 (4.74) 8
Median (range)	9.0 (1.0–17.0)	6.0 (1.0–15.0)	8.0 (1.0–17.0)	5.0 (0.0–18.0)	5.0 (0.0–14.0)	5.0 (0.0–18.0)
Anxiety category						
Normal	12 (32.4%)	15 (34.9%)	27 (33.8%)	15 (40.5%)	17 (39.5%)	32 (40.0%)
Borderline abnormal	11 (29.7%)	7 (16.3%)	18 (22.5%)	5 (13.5%)	6 (14.0%)	11 (13.8%)
Abnormal	14 (37.8%)	21 (48.8%)	35 (43.8%)	12 (32.4%)	17 (39.5%)	29 (36.3%)
Missing	0	0	0	5 (13.5%)	3 (7.0%)	8 (10.0%)
Depression category						
Normal	15 (40.5%)	23 (53.5%)	38 (47.5%)	22 (59.5%)	25 (58.1%)	47 (58.8%)
Borderline abnormal	13 (35.1%)	11 (25.6%)	24 (30.0%)	4 (10.8%)	9 (20.9%)	13 (16.3%)
Abnormal	9 (24.3%)	9 (20.9%)	18 (22.5%)	6 (16.2%)	6 (14.0%)	12 (15.0%)
Missing	0	0	0	5 (13.5%)	3 (7.0%)	8 (10.0%)

Note
HADS score 0–21; 0 indicates no anxiety or no depression, 21 indicates high anxiety or high depression. Categorical score: normal (0–7), borderline abnormal (8–10), abnormal (11–21).

TABLE 46 Hospital anxiety and depression scale category change at 12 weeks compared to baseline

	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
Anxiety			
Reduction/better by two categories	1 (2.7)	2 (4.7)	3 (3.8)
Reduction/better by one category	7 (18.9)	5 (11.6)	12 (15.0)
No change	22 (59.5)	30 (69.8)	52 (65.0)
Increase/worse by one category	1 (2.7)	2 (4.7)	3 (3.8)
Increase/worse by two categories	1 (2.7)	1 (2.3)	2 (2.5)
Missing	5 (13.5)	3 (7.0)	8 (10.0)

continued

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TABLE 46 Hospital anxiety and depression scale category change at 12 weeks compared to baseline (*continued*)

	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
Depression			
Reduction/better by two categories	1 (2.7)	1 (2.3)	2 (2.5)
Reduction/better by one category	12 (32.4)	6 (14.0)	18 (22.5)
No change	17 (45.9)	30 (69.8)	47 (58.8)
Increase/worse by one category	2 (5.4)	3 (7.0)	5 (6.3)
Missing	5 (13.5)	3 (7.0)	8 (10.0)

Note
HADS categories: normal (0–7), borderline abnormal (8–10), abnormal (11–21).

TABLE 47 IBS SSS reduction in score at 12 weeks by 50 or more points from baseline

Reduction by	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
< 50 points	12 (32.4)	20 (46.5)	32 (40.0)
≥ 50 points	19 (51.4)	18 (41.9)	37 (46.3)
Missing	6 (16.2)	5 (11.6)	11 (13.8)

TABLE 48 Number and per cent of randomised participants using loperamide by treatment period using loperamide by treatment period

Used loperamide *N* (%) missing	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Pre-treatment	8 (21.6%) 0	9 (20.9%) 2	17 (21.3%) 2
Treatment (weeks 1–12)	7 (18.9%) 0	17 (39.5%) 1	24 (30.0%) 1
Weeks 9–12 only	5 (13.5%) 3	11 (25.6%) 1	16 (20.0%) 4
Follow-up	8 (21.6%) 6	9 (20.9%) 3	17 (21.3%) 9

TABLE 49 Responder status and loperamide use

Responder?		Used loperamide?	Primary end-point responder			Abdominal pain responder			Stool consistency responder		
			Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)	Ondansetron (*n* = 37) (%)	Placebo (*n* = 43) (%)	Total (*n* = 80) (%)
Yes	AND	Yes	2 (5.4)	3 (7.0)	5 (6.3)	2 (5.4)	5 (11.6)	7 (8.8)	5 (13.5)	10 (23.3)	15 (18.8)
Yes	AND	No	13 (35.1)	9 (20.9)	22 (27.5)	15 (40.5)	11 (25.6)	26 (32.5)	20 (54.1)	12 (27.9)	32 (40.0)
No	AND	Yes	5 (13.5)	13 (30.2)	18 (22.5)	5 (13.5)	11 (25.6)	16 (20.0)	2 (5.4)	7 (16.3)	9 (11.3)
No	AND	No	15 (40.5)	16 (37.2)	31 (38.8)	13 (35.1)	14 (32.6)	27 (33.8)	8 (21.6)	13 (30.2)	21 (26.3)
Missing			2 (5.4)	2 (4.7)	4 (5.0)	2 (5.4)	2 (4.7)	4 (5.0)	2 (5.4)	1 (2.3)	3 (3.8)

Note
Loperamide used at least once in a treatment period.

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TABLE 50 Short-form Leeds dyspepsia questionnaire by time point and treatment arm

SFLDQ	Baseline			12 weeks		
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
SFLDQ score (0–32)						
Mean (SD) missing	14.9 (7.75) 1	11.7 (6.91) 1	13.2 (7.43) 2	7.5 (8.27) 8	9.4 (6.84) 4	8.6 (7.49) 12
Median (range)	15.0 (0.0–31.0)	13.0 (0.0–24.0)	14.0 (0.0–31.0)	4.0 (0.0–28.0)	8.0 (0.0–21.0)	6.0 (0.0–28.0)
SFLDQ missing items						
0	36 (97.3%)	42 (97.7%)	78 (97.5%)	29 (78.4%)	39 (90.7%)	68 (85.0%)
1	1 (2.7%)	1 (2.3%)	2 (2.5%)	2 (5.4%)	0 (0.0%)	2 (2.5%)
All questionnaire missing				6 (16.2%)	4 (9.3%)	10 (12.5%)
Most troublesome in last 2 months						
Heartburn	2 (5.4%)	5 (11.6%)	7 (8.8%)	3 (8.1%)	3 (7.0%)	6 (7.5%)
Regurgitation	2 (5.4%)	4 (9.3%)	6 (7.5%)	1 (2.7%)	2 (4.7%)	3 (3.8%)
Indigestion	9 (24.3%)	10 (23.3%)	19 (23.8%)	6 (16.2%)	6 (14.0%)	12 (15.0%)
Nausea	9 (24.3%)	15 (34.9%)	24 (30.0%)	7 (18.9%)	9 (20.9%)	16 (20.0%)
None of these have troubled me	5 (13.5%)	3 (7.0%)	8 (10.0%)	10 (27.0%)	12 (27.9%)	22 (27.5%)
Missing	10 (27.0%)	6 (14.0%)	16 (20.0%)	10 (27.0%)	11 (25.6%)	21 (26.3%)

Note
Short-form Leeds Dyspepsia Questionnaire ranges from 0 to 32; 0 is the best possible score and 32 is the worst.

TABLE 51 Irritable bowel syndrome quality of life overall and subscale summaries by time point and treatment arm

IBS-QOL	Baseline			12 weeks		
	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Overall score (0–100)						
Mean (SD) missing	39.7 (19.20) 0	43.6 (20.69) 0	41.8 (19.98) 0	57.7 (28.61) 5	53.3 (25.35) 3	55.2 (26.74) 8
Median (range)	41.9 (5.1–77.2)	39.7 (6.6–89.7)	40.8 (5.1–89.7)	58.1 (4.4–97.8)	50.0 (14.0–98.5)	52.2 (4.4–98.5)
Subscale: Mean score (SD)						
Dysphoria	38.9 (25.28)	39.2 (24.56)	39.1 (24.74)	61.5 (32.65)	52.0 (30.03)	56.2 (31.37)
Inference with activity	29.2 (19.06)	34.9 (22.63)	32.2 (21.12)	50.9 (33.81)	50.2 (28.07)	50.5 (30.53)
Body image	37.0 (22.60)	43.2 (23.62)	40.3 (23.21)	51.6 (32.18)	51.3 (28.03)	51.4 (29.73)
Health worry	57.9 (22.99)	60.1 (21.86)	59.1 (22.27)	67.4 (22.24)	67.7 (19.08)	67.6 (20.39)

TABLE 51 Irritable bowel syndrome quality of life overall and subscale summaries by time point and treatment arm (*continued*)

	Baseline			12 weeks		
IBS-QOL	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Food avoidance	23.4 (21.86)	33.3 (29.10)	28.8 (26.32)	45.6 (34.26)	39.6 (31.51)	42.2 (32.66)
Social reaction	41.4 (24.75)	43.6 (26.92)	42.6 (25.80)	59.4 (30.66)	51.1 (30.48)	54.8 (30.63)
Sexual	49.0 (35.89)	63.1 (33.40)	56.6 (35.07)	58.6 (35.70)	59.7 (35.41)	59.2 (35.29)
Relationship	60.1 (30.12)	56.4 (25.51)	58.1 (27.62)	70.8 (25.84)	64.8 (28.27)	67.5 (27.20)

Note
IBS-QOL transformed into a 0–100; 100 indicates good IBS-specific QoL.

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Appendix 7 Additional end-point summaries by week

TABLE 52 Primary end-point responders by week

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Composite responder			Composite responder		
Week	Yes	%	Missing	Yes	%	Missing
1	8	21.6	0	3	7.0	1
2	11	29.7	0	14	32.6	1
3	14	37.8	0	12	27.9	1
4	15	40.5	0	16	37.2	2
5	15	40.5	2	12	27.9	1
6	15	40.5	2	15	34.9	1
7	15	40.5	3	12	27.9	1
8	11	29.7	3	10	23.3	2
9	13	35.1	3	14	32.6	2
10	16	43.2	3	13	30.2	2
11	14	37.8	3	10	23.3	4
12	13	35.1	5	13	30.2	5
13	9	24.3	9	12	27.9	4
14	8	21.6	7	8	18.6	3
15	6	16.2	9	6	14.0	4
16	6	16.2	11	6	14.0	14

TABLE 53 Abdominal pain responders by week

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Abdominal pain responder			Abdominal pain responder		
Week	Yes	%	Missing	Yes	%	Missing
1	9	24.3	0	12	27.9	1
2	13	35.1	0	16	37.2	1
3	17	45.9	0	16	37.2	1
4	21	56.8	0	21	48.8	2
5	17	45.9	2	15	34.9	1
6	16	43.2	2	18	41.9	1
7	16	43.2	3	16	37.2	1

continued

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TABLE 53 Abdominal pain responders by week (*continued*)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Abdominal pain responder			Abdominal pain responder		
Week	Yes	%	Missing	Yes	%	Missing
8	17	45.9	3	15	34.9	2
9	19	51.4	3	17	39.5	2
10	19	51.4	3	19	44.2	2
11	18	48.6	3	17	39.5	4
12	17	45.9	5	16	37.2	5
13	12	32.4	9	17	39.5	4
14	13	35.1	7	12	27.9	3
15	11	29.7	9	10	23.3	4
16	11	29.7	11	9	20.9	14

TABLE 54 Stool consistency responders by week

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Stool consistency responder			Stool consistency responder		
Week	Yes	%	Missing	Yes	%	Missing
1	19	51.4	0	7	16.3	1
2	21	56.8	0	18	41.9	1
3	22	59.5	0	18	41.9	1
4	26	70.3	0	25	58.1	1
5	25	67.6	2	19	44.2	1
6	26	70.3	2	21	48.8	1
7	25	67.6	3	18	41.9	1
8	18	48.6	3	19	44.2	1
9	20	54.1	3	25	58.1	1
10	23	62.2	3	21	48.8	2
11	21	56.8	3	19	44.2	3
12	20	54.1	5	21	48.8	3
13	18	48.6	9	18	41.9	3
14	15	40.5	7	14	32.6	3
15	11	29.7	9	10	23.3	4
16	13	35.1	11	10	23.3	14

Note
Participants needed to have at least four entries in the diary for it to be considered a response in that week.

TABLE 55 Weekly loperamide use (participant used loperamide at least once in a week)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
Week	*N*	%	Missing response	*N*	%	Missing response
1	4	10.8	0	7	16.3	1
2	4	10.8	0	8	18.6	1
3	4	10.8	0	9	20.9	1
4	4	10.8	0	5	11.6	1
5	5	13.5	2	7	16.3	1
6	5	13.5	2	6	14.0	1
7	3	8.1	2	2	4.7	1
8	5	13.5	3	4	9.3	1
9	4	10.8	3	5	11.6	1
10	1	2.7	3	6	14.0	2
11	3	8.1	3	8	18.6	3
12	4	9.3	5	4	8.3	3
13	5	13.5	6	4	9.3	3
14	7	18.9	6	5	11.6	3
15	5	13.5	7	6	14.0	3
16	5	13.5	7	8	18.6	7

TABLE 56 Urgency responders (30% reduction in urgency score weekly by treatment arm)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Urgency responders			Urgency responders		
Week	Yes	%	Missing	Yes	%	Missing
1	14	37.8	2	7	16.3	5
2	16	43.2	3	11	25.6	7
3	18	48.6	4	14	32.6	7
4	23	62.2	3	19	44.2	8
5	21	56.8	4	16	37.2	6
6	19	51.4	4	18	41.9	7
7	19	51.4	5	16	37.2	8
8	18	48.6	5	16	37.2	8
9	19	51.4	5	15	34.9	8
10	19	51.4	6	17	39.5	8
11	19	51.4	6	13	30.2	8
12	17	45.9	11	14	32.6	10
13	14	37.8	12	18	41.9	8

continued

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TABLE 56 Urgency responders (30% reduction in urgency score weekly by treatment arm) (*continued*)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Urgency responders			Urgency responders		
Week	Yes	%	Missing	Yes	%	Missing
14	14	37.8	10	10	23.3	8
15	15	40.5	11	12	27.9	7
16	14	37.8	11	9	20.9	16

TABLE 57 Weekly abdominal pain score by treatment period and treatment arm

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Pain score (0–100)			Pain score (0–100)		
Week	Mean	SD	Missing	Mean	SD	Missing
1	56.8	23.0	0	50.3	17.8	1
2	49.5	25.5	0	44.5	20.8	1
3	45.7	26.7	0	42.8	24.5	1
4	42.7	25.1	0	39.1	20.7	2
5	42.6	26.4	2	43.2	22.2	1
6	43.6	26.8	2	41.8	23.8	1
7	44.1	26.9	3	42.1	25.0	1
8	42.9	26.5	3	43.2	25.3	2
9	41.8	25.5	3	41.0	23.4	2
10	42.9	25.4	3	40.6	24.4	2
11	44.4	27.5	3	41.5	26.6	4
12	41.3	27.9	5	42.3	28.3	5
13	43.4	28.2	9	42.5	26.0	4
14	45.5	28.0	7	47.8	25.2	3
15	45.7	28.1	9	48.5	24.7	4
16	47.9	29.7	11	47.4	27.4	14

TABLE 58 Weekly mean number of stools/day by treatment arm

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Number of stools			Number of stools		
Week	Mean	SD	Missing	Mean	SD	Missing
1	2.8	1.6	0	3.2	2.3	1
2	2.9	1.8	0	3.1	2.1	1
3	2.8	2.0	0	2.9	2.2	1
4	2.5	1.9	0	2.8	2.1	1

TABLE 58 Weekly mean number of stools/day by treatment arm (*continued*)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Number of stools			Number of stools		
Week	Mean	SD	Missing	Mean	SD	Missing
5	2.5	1.6	2	2.9	2.1	1
6	2.5	1.8	2	2.7	2.0	1
7	2.5	1.8	3	2.5	2.1	1
8	2.6	1.7	3	2.9	2.3	1
9	2.7	1.6	3	2.5	1.7	1
10	2.6	1.6	3	2.4	1.7	2
11	2.9	1.9	3	2.5	1.7	3
12	2.5	1.5	5	2.4	1.8	3
13	2.6	1.6	7	2.6	1.8	3
14	2.6	1.7	6	3.0	2.1	3
15	2.8	1.8	7	2.7	1.8	3
16	2.6	1.8	7	2.7	1.6	12

TABLE 59 Weekly mean stool consistency by treatment arm

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Mean stool consistency			Mean stool consistency		
Week	Mean	SD	Missing	Mean	SD	Missing
1	4.6	1.2	0	5.4	0.7	1
2	4.3	1.3	0	5.1	1.0	1
3	4.4	1.3	0	5.1	1.0	1
4	4.2	1.3	0	5.0	0.9	1
5	4.3	1.2	2	4.9	1.0	1
6	4.2	1.3	2	4.9	1.1	1
7	4.3	1.2	3	5.2	0.8	1
8	4.4	1.2	3	5.0	0.9	1
9	4.3	1.4	3	4.9	1.0	1
10	4.3	1.4	3	5.0	0.8	2
11	4.4	1.5	3	5.0	0.9	3
12	4.2	1.4	5	4.9	1.1	3
13	4.7	1.4	7	5.0	1.0	3
14	5.0	1.4	6	5.2	0.8	3
15	5.2	1.2	7	5.3	0.8	3
16	5.1	1.0	7	5.2	0.9	12

Note
Desirable stool consistency 3–5.

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TABLE 60 Weekly mean number of days/week with loose stool by treatment arm

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Number of days with loose stool			Number of days with loose stool		
Week	Mean	SD	Missing	Mean	SD	Missing
1	3.0	2.6	0	4.4	2.0	1
2	2.5	2.4	0	3.4	2.6	1
3	2.4	2.5	0	3.3	2.4	1
4	2.1	2.2	0	2.8	2.4	1
5	2.0	2.4	2	3.0	2.4	1
6	2.1	2.6	2	3.0	2.6	1
7	2.1	2.5	3	3.1	2.5	1
8	2.5	2.3	3	3.1	2.5	1
9	2.5	2.8	3	2.9	2.6	1
10	2.3	2.5	3	2.8	2.3	2
11	2.9	2.6	3	3.0	2.4	3
12	2.6	2.8	5	2.7	2.5	3
13	2.4	2.6	9	3.0	2.5	3
14	3.1	2.8	7	3.7	2.5	3
15	3.8	2.7	9	3.8	2.3	4
16	3.5	2.8	11	3.9	2.4	14

TABLE 61 Satisfactory relief by week and by treatment arm

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Satisfactory relief			Satisfactory relief		
Week	Yes	%	Missing	Yes	%	Missing
1	8	21.6	8	10	23.3	12
2	10	27.0	12	15	34.9	11
3	15	40.5	12	17	39.5	11
4	15	40.5	10	14	32.6	14
5	12	32.4	14	11	25.6	17
6	13	35.1	15	14	32.6	19
7	12	32.4	17	13	30.2	16
8	16	43.2	14	12	27.9	15
9	14	37.8	17	14	32.6	13
10	13	35.1	16	14	32.6	12
11	14	37.8	15	11	25.6	14

TABLE 61 Satisfactory relief by week and by treatment arm (*continued*)

	Ondansetron (*n* = 37)			Placebo (*n* = 43)		
	Satisfactory relief			Satisfactory relief		
Week	Yes	%	Missing	Yes	%	Missing
12	14	37.8	17	12	27.9	18
13	7	18.9	18	8	18.6	18
14	5	13.5	18	6	14.0	19
15	5	13.5	19	4	9.3	18
16	4	10.8	19	2	4.7	27

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Appendix 8 Safety

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TABLE 62 Adverse event reporting at different time points

	6 weeks			12 weeks			Follow-up		
Experienced *N* (%)	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**
Constipation									
Yes	17 (45.9%)	11 (25.6%)	28 (35.0%)	12 (32.4%)	10 (23.3%)	22 (27.5%)	5 (13.5%)	8 (18.6%)	13 (16.3%)
Missing	2 (5.4%)	3 (7.0%)	5 (6.3%)	3 (8.1%)	3 (7.0%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	11 (64.7%)	6 (54.5%)	17 (60.7%)	9 (75.0%)	8 (80.0%)	17 (77.3%)	4 (80.0%)	3 (37.5%)	7 (53.8%)
Moderate	5 (29.4%)	5 (45.5%)	10 (35.7%)	2 (16.7%)	2 (20.0%)	4 (18.2%)	0 (0.0%)	5 (62.5%)	5 (38.5%)
Severe	1 (5.9%)	0 (0.0%)	1 (3.6%)	1 (8.3%)	0 (0.0%)	1 (4.5%)	1 (20.0%)	0 (0.0%)	1 (7.7%)
Abdominal pain/bloating									
Yes	26 (70.3%)	36 (83.7%)	62 (77.5%)	23 (62.2%)	33 (76.7%)	56 (70.0%)	25 (67.6%)	36 (83.7%)	61 (76.3%)
Missing	2 (5.4%)	2 (4.7%)	4 (5.0%)	3 (8.1%)	3 (7.0%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	10 (38.5%)	20 (55.6%)	30 (48.4%)	12 (52.2%)	14 (42.4%)	26 (46.4%)	7 (28.0%)	9 (25.0%)	16 (26.2%)
Moderate	10 (38.5%)	11 (30.6%)	21 (33.9%)	7 (30.4%)	15 (45.5%)	22 (39.3%)	11 (44.0%)	17 (47.2%)	28 (45.9%)
Severe	6 (23.1%)	5 (13.9%)	11 (17.7%)	4 (17.4%)	4 (12.1%)	8 (14.3%)	7 (28.0%)	10 (27.8%)	17 (27.9%)
Headache									
Yes	17 (45.9%)	8 (18.6%)	25 (31.3%)	11 (29.7%)	14 (32.6%)	25 (31.3%)	10 (27.0%)	12 (27.9%)	22 (27.5%)
Missing	2 (5.4%)	4 (9.3%)	6 (7.5%)	3 (8.1%)	3 (7.0%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	11 (64.7%)	7 (87.5%)	18 (72.0%)	3 (27.3%)	8 (57.1%)	11 (44.0%)	6 (60.0%)	8 (66.7%)	14 (63.6%)
Moderate	5 (29.4%)	1 (12.5%)	6 (24.0%)	5 (45.5%)	5 (35.7%)	10 (40.0%)	3 (30.0%)	4 (33.3%)	7 (31.8%)
Severe	1 (5.9%)	0 (0.0%)	1 (4.0%)	3 (27.3%)	1 (7.1%)	4 (16.0%)	1 (10.0%)	0 (0.0%)	1 (4.5%)

TABLE 62 Adverse event reporting at different time points (*continued*)

	6 weeks			12 weeks			Follow-up		
Experienced *N* (%)	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**	**Ondansetron (*n* = 37)**	**Placebo (*n* = 43)**	**Total (*n* = 80)**
Nausea									
Yes	6 (16.2%)	12 (27.9%)	18 (22.5%)	10 (27.0%)	18 (41.9%)	28 (35.0%)	9 (24.3%)	12 (27.9%)	21 (26.3%)
Missing	2 (5.4%)	4 (9.3%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	4 (66.7%)	9 (75.0%)	13 (72.2%)	7 (70.0%)	10 (55.6%)	17 (60.7%)	3 (33.3%)	6 (50.0%)	9 (42.9%)
Moderate	2 (33.3%)	3 (25.0%)	5 (27.8%)	1 (10.0%)	5 (27.8%)	6 (21.4%)	4 (44.4%)	3 (25.0%)	7 (33.3%)
Severe				2 (20.0%)	3 (16.7%)	5 (17.9%)	2 (22.2%)	3 (25.0%)	5 (23.8%)
Vomiting									
Yes	3 (8.1%)	3 (7.0%)	6 (7.5%)	1 (2.7%)	4 (9.3%)	5 (6.3%)	4 (10.8%)	6 (14.0%)	10 (12.5%)
Missing	2 (5.4%)	4 (9.3%)	6 (7.5%)	3 (8.1%)	3 (7.0%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	2 (66.7%)	2 (66.7%)	4 (66.7%)				0 (0.0%)	1 (16.7%)	1 (10.0%)
Moderate	1 (33.3%)	1 (33.3%)	2 (33.3%)	0 (0.0%)	4 (100.0%)	4 (80.0%)	3 (75.0%)	2 (33.3%)	5 (50.0%)
Severe				1 (100.0%)	0 (0.0%)	1 (20.0%)	1 (25.0%)	3 (50.0%)	4 (40.0%)
Rectal bleeding									
Yes	3 (8.1%)	7 (16.3%)	10 (12.5%)	4 (10.8%)	6 (14.0%)	10 (12.5%)	4 (10.8%)	6 (14.0%)	10 (12.5%)
Missing	2 (5.4%)	2 (4.7%)	4 (5.0%)	3 (8.1%)	3 (7.0%)	6 (7.5%)	3 (8.1%)	2 (4.7%)	5 (6.3%)
If yes: Severity									
Mild	3 (100.0%)	6 (85.7%)	9 (90.0%)	4 (100.0%)	6 (100.0%)	10 (100.0%)	4 (100.0%)	6 (100.0%)	10 (100.0%)
Moderate									
Severe	0 (0.0%)	1 (14.3%)	1 (10.0%)						

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TABLE 63 If the patient experienced rectal bleeding at 6 weeks

Patient	Clinical opinion obtained[a]	Colonoscopy/flexi sigmoidoscopy performed	Allocation
1	Yes	No	Ondansetron
2	Yes	No	Ondansetron
3	Yes	No	Ondansetron
4	Yes	No	Placebo
5	Yes	No	Placebo
6	Yes	No	Placebo
7	Yes	No	Placebo
8	Yes	No	Placebo
9	Yes	No	Placebo
10	No	No	Placebo

a No patient had ischaemic colitis diagnosed.

TABLE 64 Pulse and BP at 12 weeks

	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Pulse within normal range			
Yes	29 (78.4%)	31 (72.1%)	60 (75.0%)
Missing	8 (21.6%)	12 (27.9%)	20 (25.0%)
BP within normal range			
Yes	28 (75.7%)	30 (69.8%)	58 (72.5%)
No	1 (2.7%)	1 (2.3%)	2 (2.5%)
Missing	8 (21.6%)	12 (27.9%)	20 (25.0%)

TABLE 65 Pregnancy

Treatment allocation	Details	Pregnancy outcome	Number of days between randomisation and date of positive pregnancy test
Placebo	Trial participant is pregnant	Live birth	96

TABLE 66 Patient starting any new medications at 6 weeks

Patient	New medications since previous visit	PI confirmation of medications not being restricted or prohibited	Allocation
1	Yes	Yes	Ondansetron
2	Yes	Yes	Placebo
3	Yes	Yes	Placebo
4	Yes	Yes	Placebo
5	Yes	Yes	Ondansetron

TABLE 67 Recommended titrated dose at 2 and 6 weeks and changes in dose

	Ondansetron (*n* = 37)	Placebo (*n* = 43)	Total (*n* = 80)
Recommended titrated dose after 2 weeks of treatment			
One capsule (4 mg) every 3 days	1 (2.7%)	2 (4.7%)	3 (3.8%)
One capsule (4 mg) every 2 days	7 (18.9%)	3 (7.0%)	10 (12.5%)
One capsule (4 mg) daily	6 (16.2%)	3 (7.0%)	9 (11.3%)
Two capsules (8 mg) daily	8 (21.6%)	10 (23.3%)	18 (22.5%)
Three capsules (12 mg) daily	2 (5.4%)	5 (11.6%)	7 (8.8%)
Four capsules (16 mg) daily	2 (5.4%)	3 (7.0%)	5 (6.3%)
Five capsules (20 mg) daily	5 (13.5%)	3 (7.0%)	8 (10.0%)
Six capsules (24 mg) daily	3 (8.1%)	12 (27.9%)	15 (18.8%)
Missing	3 (8.1%)	2 (4.7%)	5 (6.3%)
Number of bottles requested 6 weeks post randomisation			
One bottle (one capsule or less per day)	7 (18.9%)	6 (14.0%)	13 (16.3%)
Two bottles (two capsules per day)	8 (21.6%)	9 (20.9%)	17 (21.3%)
Three bottles (three capsules per day)	3 (8.1%)	5 (11.6%)	8 (10.0%)
Four bottles (four capsules per day)	3 (8.1%)	5 (11.6%)	8 (10.0%)
Five bottles (five capsules per day)	3 (8.1%)	2 (4.7%)	5 (6.3%)
Six bottles (six capsules per day)	5 (13.5%)	10 (23.3%)	15 (18.8%)
Missing	8 (21.6%)	6 (14.0%)	14 (17.5%)
Number of bottles requested 6 weeks post randomisation consistent with the recommended titrated dose after 2 weeks?			
Yes	19 (51.4%)	29 (67.4%)	48 (60.0%)
No	8 (21.6%)	6 (14.0%)	14 (17.5%)
Missing	10 (27.0%)	8 (18.6%)	18 (22.5%)
Dose at 2 weeks compared with 6 weeks *N* participants	10	7	17
Participant on a lower dose at 6 weeks compared with 2 weeks	2 (20.0%)	2 (28.6%)	4 (23.5%)
Participant on a higher dose at 6 weeks compared with 2 weeks	8 (80.0%)	5 (71.4%)	13 (76.5%)
Number of capsules returned at 12 weeks consistent with dose at 6 weeks *N* participants	33	42	75
Yes	7 (21.2%)	7 (16.7%)	14 (18.7%)
No	22 (66.7%)	32 (76.2%)	54 (72.0%)
Missing	4 (12.1%)	3 (7.1%)	7 (9.3%)
Dose at 12 weeks compared with 6 weeks *N*	24	32	56
Participant on a higher dose at 12 weeks compared with 6 weeks	9 (37.5%)	20 (62.5%)	29 (51.8%)
Participant on a lower dose at 12 weeks compared with 6 weeks	15 (62.5%)	12 (37.5%)	27 (48.2%)

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Appendix 9 Exit poll

TABLE 68 Certainty of choice and reasons for choice of treatment – site completed

Certainty of choice	Reasons for choice of those that guessed ondansetron						
	Ondansetron (*n* = 22)	Treatment benefited the patient (*n* = 21)	AE(s) consistent with ondansetron (*n* = 1)	Placebo (*n* = 17)	Treatment benefited the patient (*n* = 16)	Other[a] (*n* = 1)	Total (*n* = 39)
4				5 (29.4%)	5 (31.3%)		5 (12.8%)
5	1 (4.5%)	1 (4.8%)		2 (11.8%)	1 (6.3%)	1 (100.0%)	3 (7.7%)
6	2 (9.1%)	2 (9.5%)		2 (11.8%)	2 (12.5%)		4 (10.3%)
7	5 (22.7%)	5 (23.8%)		3 (17.6%)	3 (18.8%)		8 (20.5%)
8	6 (27.3%)	6 (28.6%)		3 (17.6%)	3 (18.8%)		9 (23.1%)
9	2 (9.1%)	2 (9.5%)		1 (5.9%)	1 (6.3%)		3 (7.7%)
10	6 (27.3%)	5 (23.8%)	1 (100.0%)	1 (5.9%)	1 (6.3%)		7 (17.9%)
	Reasons for choice of those that guessed placebo						
	Ondansetron (*n* = 8)	**Treatment had no benefit (*n* = 8)**		**Placebo (*n* = 21)**	**Treatment had no benefit (*n* = 21)**		**Total (*n* = 29)**
4	1 (12.5%)	1 (12.5%)		2 (9.5%)	2 (9.5%)		3 (10.3%)
5				1 (4.8%)	1 (4.8%)		1 (3.4%)
6	1 (12.5%)	1 (12.5%)		2 (9.5%)	2 (9.5%)		3 (10.3%)
7	2 (25.0%)	2 (25.0%)		3 (14.3%)	3 (14.3%)		5 (17.2%)
8	1 (12.5%)	1 (12.5%)		3 (14.3%)	3 (14.3%)		4 (13.8%)
9	2 (25.0%)	2 (25.0%)		7 (33.3%)	7 (33.3%)		9 (31.0%)
10	1 (12.5%)	1 (12.5%)		3 (14.3%)	3 (14.3%)		4 (13.8%)
	Reasons for choice of those that responded 'Don't know'[b]						
	Ondansetron (*n* = 5)			**Placebo (*n* = 2)**			**Total (*n* = 7)**
0				1 (50.0%)			1 (14.3%)
5	2 (40.0%)						2 (28.6%)
6				1 (50.0%)			1 (14.3%)
7	1 (20.0%)						1 (14.3%)
9	2 (40.0%)						2 (28.6%)

a Other: discussion with patient.
b Those that responded 'Don't know' did not have to complete reasons for choice.

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TABLE 69 Certainty of choice and reasons for choice of treatment – patient completed

Certainty of choice	Reasons for choice of those that guessed ondansetron						
	Ondansetron (*n* = 22)	**Treatment worked (*n* = 18)**	**I had a side effect (*n* = 3)**	**Other (*n* = 1)[a]**	**Placebo (*n* = 16)**	**Treatment worked (*n* = 16)**	**Total (*n* = 38)**
2	1 (4.5%)		1 (33.3%)				1 (2.6%)
5	0 (0.0%)				1 (6.3%)	1 (6.3%)	1 (2.6%)
6	4 (18.2%)	3 (16.7%)	0 (0.0%)	1 (100.0%)	1 (6.3%)	1 (6.3%)	5 (13.2%)
7	2 (9.1%)	2 (11.1%)	0 (0.0%)		3 (18.8%)	3 (18.8%)	5 (13.2%)
8	4 (18.2%)	3 (16.7%)	1 (33.3%)		2 (12.5%)	2 (12.5%)	6 (15.8%)
9	1 (4.5%)	1 (5.6%)	0 (0.0%)		3 (18.8%)	3 (18.8%)	4 (10.5%)
10	10 (45.5%)	9 (50.0%)	1 (33.3%)		6 (37.5%)	6 (37.5%)	16 (42.1%)
	Reasons for choice of those that guessed placebo						
	Ondansetron (*n* = 8)	**Treatment didn't work (*n* = 6)**	**Treatment worked (*n* = 1)**	**Other (*n* = 1)[b]**	**Placebo (*n* = 18)**	**Treatment didn't work (*n* = 18)**	**Total (*n* = 26)**
4	2 (25.0%)	2 (33.3%)					2 (7.7%)
5	1 (12.5%)		1 (100.0%)		1 (5.6%)	1 (5.6%)	2 (7.7%)
6	1 (12.5%)			1 (100.0%)	1 (5.6%)	1 (5.6%)	2 (7.7%)
7					2 (11.1%)	2 (11.1%)	2 (7.7%)
8					6 (33.3%)	6 (33.3%)	6 (23.1%)
9					5 (27.8%)	5 (27.8%)	5 (19.2%)
10	4 (50.0%)	4 (66.7%)			3 (16.7%)	3 (16.7%)	7 (26.9%)
	Reasons for choice of those that responded 'Don't know'[c]						
	Ondansetron (*n* = 3)				**Placebo (*n* = 7)**		**Total (*n* = 10)**
2	1 (33.3%)						1 (10.0%)
4					1 (14.3%)		1 (10.0%)
5					3 (42.9%)		3 (30.0%)
6					1 (14.3%)		1 (10.0%)
7	1 (33.3%)				2 (28.6%)		3 (30.0%)
10	1 (33.3%)						1 (10.0%)

a Other: First 6 weeks fine then no good.
b Other: Felt it was inconsistent on how it worked.
c Those that responded 'Don't know' did not have to complete reasons for choice.

Appendix 10 Advertisement

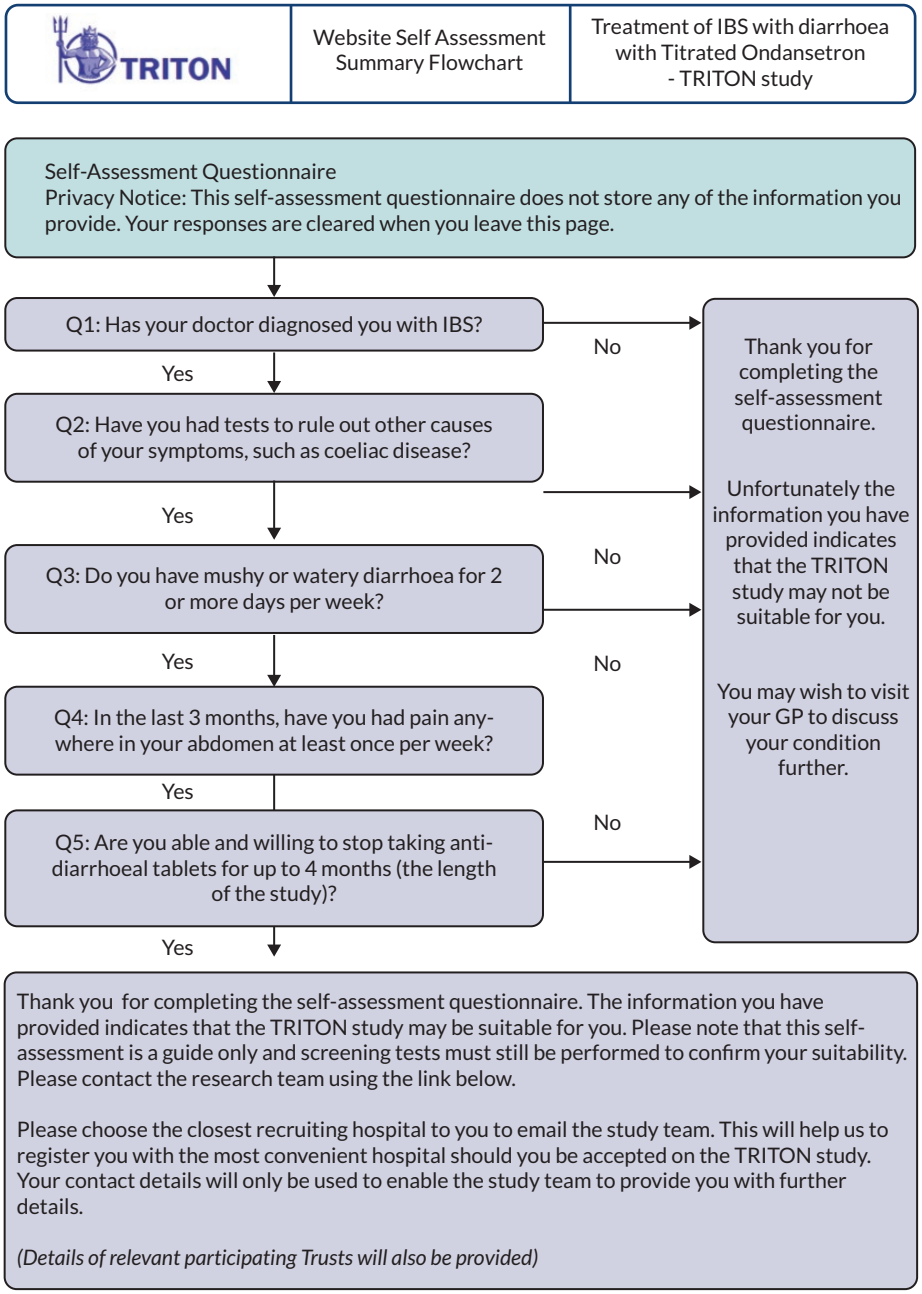


FIGURE 32 Website self-assessment summary flowchart.

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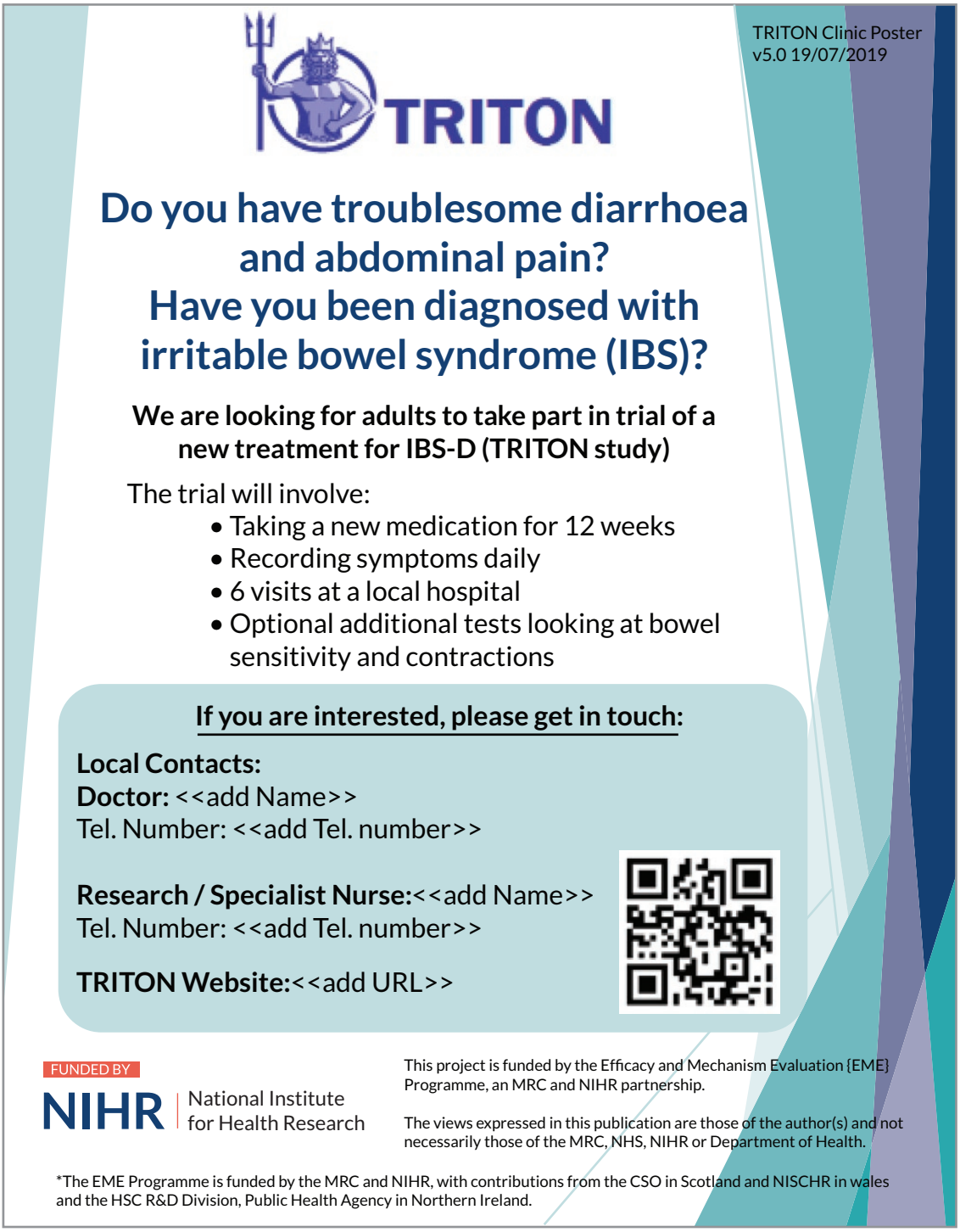


FIGURE 33 Clinic poster.

Do you suffer from IBS with Diarrhoea (IBS-D)?

- Has your doctor diagnosed you with IBS?
- Have you had tests to rule out other causes of your symptoms, such as coeliac disease?
- Do you have mushy or watery diarrhoea for 2 or more days per week?
- In the last 3 months, have you had pain anywhere in your abdomen at least once per week?
- Are you able and willing to stop taking anti-diarrhoeal tablets for up to 4 months (the length of the study)?

We are looking for adults to take part in a study investigating treatment for IBS-D (the TRITON study).

See overleaf for more details.

TRITON

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Version 3.0 28/06/2019

This project is funded by the Efficacy and Mechanism Evaluation {EME} Programme, an MRC and NIHR partnership. The views expressed in this publication are those of the author(s) and not necessarily those of the MRC, NHS, NIHR or the Department of Health. The EME Programme is funded by the MRC and NIHR, with contributions from the CSO in Scotland and NISCHR in wales and the HSC R&D Division, Public Health Agency in Northern Ireland.

FIGURE 34 Clinic leaflet page 1; clinic leaflet page 2.

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Do you suffer from IBS with Diarrhoea (IBS-D)?

TRITON

We are looking for adults to take part in a study investigating treatment for IBS-D (the TRITON trial).

The study will involve:

- Taking a study medication for 12 weeks
- Recording symptoms daily
- 6 visits to a local hospital
- Optional tests looking at bowel sensitivity and contractions
- Inconvenience payments so you are not out of pocket

If you are interested, please contact the study researchers Dr David Gunn or Dr Rabia Lalani. They will answer your questions and direct you to the nearest hospital running the TRITON study.

Email:

Telephone:

Website: https://ctru.leeds.ac.uk/triton

University of Nottingham UK | CHINA | MALAYSIA

ctru University of Leeds

Queen Mary University of London

NHS Nottingham University Hospitals NHS Foundation Trust

FIGURE 34 Clinic leaflet page 1; clinic leaflet page 2. (*Continued*)



FIGURE 35 Press advertisement.

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