**SUPPLEMENTARY SECTION**

**A randomised, double-blind, placebo-controlled pilot trial of omeprazole in idiopathic pulmonary fibrosis**

Prosenjit Dutta1\*, Wendy Funston1\*, Helen Mossop2, Vicky Ryan2, Rhys Jones1, Rebecca Forbes3, Shilpi Sen4, Jeffrey Pearson5, Michael Griffin6, Jaclyn A Smith4,7, Christopher Ward1, Ian A Forrest8^, A John Simpson1,8^

**SUPPLEMENTARY METHODS**

**Study schedule**

The study schedule is shown in Table S1.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  | Days -7 to 0  (ie any 3 days in the lead up to starting study medication | | | Days 1-87 | Days 88-90  **§§** | | |
| Confirm eligibility | \* |  |  |  |  |  |  |
| Consent | \* |  |  |  |  |  |  |
| Demographics | \* |  |  |  |  |  |  |
| VC and TLco | \* |  |  |  | \* |  |  |
| 6-minute walk distance | \* |  |  |  | \* |  |  |
| Cough questionnaire | \* |  |  |  | \* |  |  |
| Reflux questionnaire | \* |  |  |  | \* |  |  |
| Commence 24h  cough monitoring | \* |  |  |  | \* |  |  |
| Commence 24h  oesophageal physiology tests |  | \* |  |  |  | \* |  |
| Randomisation |  |  | \* |  |  |  |  |
| Issue study drug |  |  | \* |  |  |  |  |
| Issue adverse event diary | \* |  |  |  |  |  |  |
| Take study medication |  |  |  | \* | \* | \* | \* |
| Bronchoscopy and BAL |  |  |  |  |  |  | \* |

**Table S1. Summary of study assessments and participant visits.**

§§ - the tests scheduled for days 88-90 were ideally performed on these days but could be performed at any point from day 76.

**Inclusion criteria**

A pragmatic clinical definition of IPF was used, in which recruited participants had to fulfil all of the following criteria:

* IPF was considered the most likely diagnosis by the regional interstitial lung disease multidisciplinary team meeting (ILD MDT)
* history of cough, with or without exertional dyspnoea
* high resolution computed tomography (HRCT) scan features of honeycombing in a predominantly basal and subpleural distribution
* bi-basal crackles on auscultation
* features of a restrictive ventilatory defect (vital capacity (VC) <90% predicted and/or diffusion factor for carbon monoxide (TLco) <90% predicted)
* age 40-85 years

Patients with radiological emphysema were eligible so long as the diagnosis of IPF was secure, ie all the features above were satisfied.

If the regional ILD MDT could not reach a clear consensus as to the diagnosis, the case would be referred to 2 experts in ILD from outside the region, and the patient eligible if both considered IPF to be the most likely diagnosis.

Patients taking a PPI during screening were potentially eligible. In these cases the indication for on-going treatment was reviewed.

* Patients taking short courses (e.g. 2 months) of PPI were considered eligible once the treatment had been discontinued for a minimum of 1 month.
* There are few licensed indications for long-term omeprazole beyond Zollinger-Ellison syndrome. Therefore, unless there was a known diagnosis of Zollinger-Ellison or a history of significant dyspepsia or gastrointestinal bleeding during a previous discontinuation of PPI, patients on long-term PPI were asked to consider a trial of supervised discontinuation. If the patient agreed and provided written consent to a trial of discontinuation, the GP was contacted. If both the patient and the GP agreed to a trial of discontinuation, the patient was eligible for the study if he/she provided written consent after at least 2 weeks without PPI.

Patients taking antacids, prokinetics or raft alginates at the time of screening were eligible if they have been off these treatments for a period of at least 2 weeks.

**Exclusion criteria** consisted of:

* known allergy to omeprazole or another PPI
* concomitant use of warfarin, diazepam, phenytoin or ketoconazole
* concomitant use of a regular PPI, antacid, prokinetic or raft alginate during the trial period
* history of upper respiratory tract infection, lower respiratory tract infection or exacerbation of IPF in the 4 weeks before starting study drugs
* active trial of treatment for IPF (e.g. pirfenidone, nintedanib, prednisolone, N-acetylcysteine) started in the 4 weeks before starting study drugs
* documented history of hepatic cirrhosis
* pregnancy or lactation
* ILD MDT considered the most likely cause of the patient’s ILD to be a condition other than IPF, for example rheumatoid lung, systemic sclerosis ILD, asbestosis, chronic hypersensitivity pneumonitis, sarcoidosis, etc.
* concurrent enrolment in a clinical trial of an investigational medicinal product (CTIMP) for IPF.

**Questionnaires**

Leicester Cough Questionnaire (LCQ). This is a fully validated 19-item cough-related questionnaire. It is divided into 3 domains (physical, social and psychological) to assess the overall impact of cough in day-to-day life and to assess any improvement after intervention. It has a 7-point Likert response scale and the total calculated score ranges from 3-21. A higher score implies better quality of life.

Reflux Symptom Index (RSI). This is a fully validated 9-item questionnaire to assess the possibility of laryngo-pharyngeal reflux. Each item is rated on a scale of 0 – 5 (0 = no problems, 5 = severe problems). Possible scores range from 0 – 45. A composite score of 10 or below is taken as normal, while scores above 10 should prompt assessment for reflux disease. Lower scores imply better health.

Gastro-Intestinal Quality of Life Index Questionnaire (GIQLI) and De-Meester Reflux Questionnaire (DeMRQ). These two questionnaires are designed to assess quality of life in patients with gastro-intestinal disease in clinical practice and in clinical studies. GIQLI is a 36-item questionnaire relating to symptoms attributable to gastro-intestinal disease. There are 5 possible options or responses to each question with 4 points allocated to the “most desirable option” and 0 points allocated to the “least desirable option”. Possible scores range from 0 – 144 with higher scores indicating better quality of life. Healthy individuals have a mean score of 122.6 +/- 8.5. The DeMRQ questionnaire is a short 3-item questionnaire to assess health-related quality of life in patients with reflux disease. Possible scores range from 0 – 9 with low scores indicating better quality of life.

**Oesophageal physiology tests**

Oesophageal manometry was performed using a water-perfused system (Medical Measurement Systems, Enschede, the Netherlands). Oesophageal peristalsis and lower oesophageal sphincter function were assessed using ten 5 ml water swallows according to the Chicago classification.[S1] The location of the lower oesophageal sphincter was defined manometrically.

24-hour ambulatory pH-impedance monitoring was performed using the Ohmega™ system (Medical Measurement Systems). Impedance was measured across six oesophageal segments and a pH probe was located 5cm above the lower oesophageal sphincter. Data were compared to published normal ranges derived from healthy European volunteers.[S2]

**Bronchoscopy and bronchoalveolar lavage (BAL)**

Continuous monitoring of oxygen saturations was employed throughout. Supplemental oxygen was provided. Participants were offered sedation with intravenous midazolam and topical anaesthesia with 1% lidocaine. Bronchoscopy and BAL were performed by an experienced operator (IAF). The bronchoscope was passed into a lobe known to be affected by IPF on HRCT. Three 60ml aliquots of sterile saline were sequentially instilled and aspirated.

An aliquot of the BAL fluid retrieved was used neat for microbiological analysis. BAL fluid was plated on microbiological culture plates and incubated at 37°C for 24 hours before colony counting. Remaining BAL fluid was centrifuged and the cell-free supernatant retrieved and frozen at -80°C for later use. The cell pellet was re-suspended for cell counting and to make cytospin preparations, which were stained with Giemsa to allow differential cell counts to be estimated.

**Estimation of inflammatory and anti-inflammatory mediators in BAL fluid**

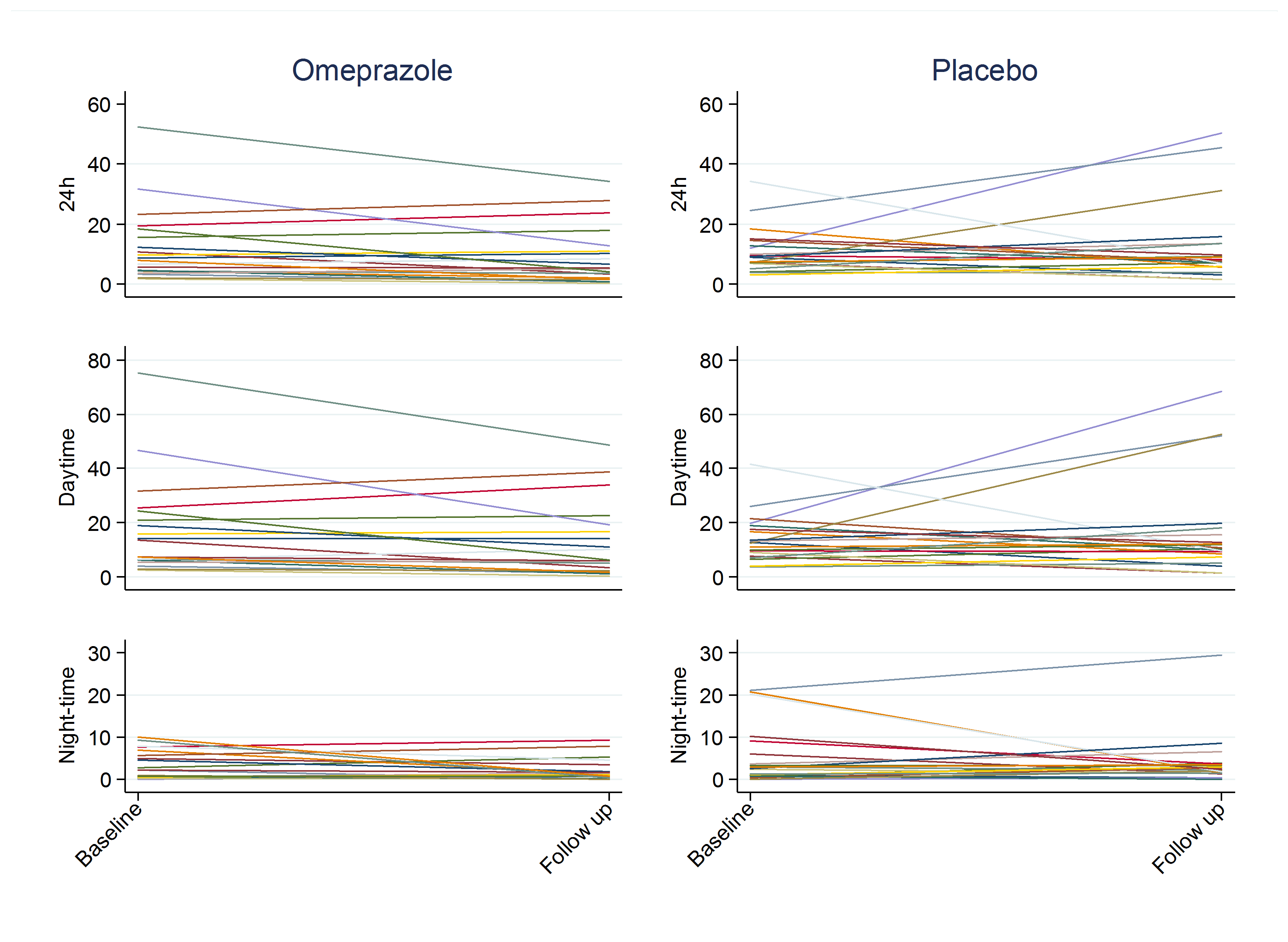
Interleukin (IL)-1 beta, IL-6, IL-8, IL-10, tumour necrosis factor (TNF) and transforming growth factor (TGF) beta were measured by enzyme-linked immunosorbent assay (ELISA, R&D Systems, Minneapolis, MN, USA) and pepsin by in-house ELISA.[S3] Urea was measured by spectrophotometry (Quantichrom, BioAssay Systems, Hayward, CA, USA) and total bile acids by colorimetric assay (DIALAB, Wiener Neudorf, Austria). Glyco- and tauro-dihydroxycholanoic (DHC) acid, as well as glyco- and tauro-trihydroxycholanoic acid, were assessed by tandem mass spectrometry.[S4]

A copy of the statistical analysis plan for the study is shown as an appendix at the end of this document.

**SUPPLEMENTARY RESULTS**

**Changes in cough frequency in individual patients**

Alterations in patients’ cough frequency over time are shown in Figure S1.



**Figure S1. Spaghetti plots of cough frequency over time in individual patients.**

**Oesophageal physiology tests**

In total, 13 participants gave consent to take part in the 24-hour oesophageal physiology study. Assessments were completed at baseline in 9 participants. Seven participants had high resolution manometry at the end of treatment, six of whom completed the 24-hour impedance pH study. High resolution manometry data are summarised in Table S2.

|  |  |  |
| --- | --- | --- |
| Treatment group | High resolution manometry | |
| Baseline | Follow up |
| omeprazole | Normal | Normal |
| Normal | Ineffective oesophageal motility |
| Oesophago-gastric junction outflow obstruction | Normal |
| Oesophago-gastric junction outflow obstruction | Normal |
| Normal | Normal |
| Ineffective oesophageal motility |  |
| Oesophago-gastric junction outflow obstruction | Distal oesophageal spasm |
| placebo | Normal | Normal |
| Normal |  |

**Table S2. High resolution manometry.**

Individual ambulatory pH/impedance testing data are listed in Table S3. Where noted the raw data have been corrected for the time on study (i.e. raw data x (time on study (in hours) / 24 hours)).

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Visit | Treatment group | Time pH<4 (%) | No. of times pH<4 | No. of long reflux episodes | Longest reflux episode (min) | No. of acid refluxes | No. of week acid refluxes | No. of proximal reflux events | DeMeester score | Total No. of reflux episodes | Bolus clearance time  (min) | Symptom associated probability |
| Baseline | Omeprazole | 6.4 | 36.0 | 4.0 | 25.2 | 35.0 | 24.0 | 10.0 | 18.4 | 73.1 | 9.0 | 0.0 |
| Follow up | 1.0 | 9.0 | 1.0 | 6.2 | 10.0 | 21.9 | 1.0 | 4.0 | 35.0 | 12.0 | 0.0 |
| Baseline | 12.8 | 71.0 | 6.0 | 33.0 | 48.0 | 17.0 | 13.0 | 46.4 | 65.9 | 10.0 | 99.9 |
| Follow up | 5.5 | 111.9 | 2.0 | 11.3 | 12.0 | 34.0 | 0.9 | 24.0 | 46.0 | 10.0 | 0.0 |
| Baseline | 8.4 | 51.0 | 7.0 | 13.1 | 36.0 | 9.0 | 20.0 | 25.4 | 46.0 | 12.0 | 0.0 |
| Follow up | 0.1 | 2.0 | 0.0 | 1.2 | 0.0 | 15.0 | 5.0 | 0.7 | 15.0 | 12.0 | 0.0 |
| Baseline | 1.0 | 13.0 | 0.0 | 3.2 | 15.0 | 38.0 | 13.0 | 3.8 | 55.0 | 8.0 | 0.0 |
| Follow up | 0.1 | 1.0 | 0.0 | 1.2 | 0.0 | 25.0 | 4.0 | 0.6 | 27.0 | 5.0 | 0.0 |
| Baseline | 4.2 | 23.0 | 2.0 | 26.3 | 29.0 | 5.0 | 21.0 | 17.6 | 36.0 | 13.0 | 0.0 |
| Follow up | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 48.0 | 22.0 | 0.2 | 52.0 | 15.0 | 0.0 |
| Baseline | 0.7 | 11.0 | 0.0 | 1.7 | 1.0 | 58.0 | 13.0 | 3.0 | 61.0 | 14.0 | 0.0 |
| Follow up |  |  |  |  |  |  |  |  |  |  |  |
| Baseline | 1.6 | 14.0 | 0.0 | 4.0 | 12.0 | 19.0 | 3.0 | 4.2 | 31.0 | 11.0 | 0.0 |
| Follow up |  |  |  |  |  |  |  |  |  |  |  |
| Baseline | Placebo | 7.6 | 54.1 | 4.0 | 14.6 | 23.0 | 15.0 | 2.0 | 22.9 | 38.0 | 13.0 | 99.9 |
| Follow up | 23.1 | 171.0 | 13.0 | 20.4 | 37.0 | 2.8 | 20.3 | 74.5 | 42.5 | 16.5 | 0.0 |
| Baseline | 22.4 | 65.0 | 8.0 | 16.9 | 18.6 | 2.7 | 17.0 | 57.8 | 30.8 | 16.0 | 0.0 |
| Follow up |  |  |  |  |  |  |  |  |  |  |  |

**Table S3. Individual pH impedance data by visit.**

**Bronchoscopy and BAL**

In total, 13 participants gave consent to take part in bronchoscopy and BAL studies at follow up. Assessments were completed on 8 of these participants (5 in the omeprazole group). In the omeprazole group, *Staphylococcus aureus* was isolated from the BAL fluid of one participant, and Pseudomonas from another. No potential pathogens were isolated from the other 3 participants in the omeprazole group or the 3 participants in the placebo group. Mediators of lung inflammation and markers of aspiration are shown for individual participants in Table S4, and differential cell counts from BAL fluid in Table S5.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Omeprazole | | | | | Placebo | | |
| IL-1 beta (pg/ml) | ND | ND | ND | ND | ND | ND | ND | ND |
| IL-6 (pg/ml) | ND | ND | ND | ND | ND | ND | ND | ND |
| IL-8 (pg/ml) | 141.6 | 156.6 | 138.8 | 99.6 | ND | 81.0 | 52.1 | 14.4 |
| IL-10 (pg/ml) | ND | 9.0 | ND | ND | ND | ND | ND | ND |
| TNF alpha (pg/ml) | 58.9 | ND | ND | ND | ND | ND | ND | ND |
| TGF beta (pg/ml) | 157.1 | 76.1 | 21.1 | ND | ND | ND | ND | ND |
| Urea (mg/dl) | 2.28 | 1.48 | 1.37 | 0.34 | 0.99 | 0.56 | 1.43 | 0.10 |
| Pepsin (ng/ml) | 2.6 | 2.5 | 10.5 | 8.4 | ND | ND | ND | 8.3 |
| Total Bile Acids (mmol/l) | ND | ND | ND | ND | ND | ND | ND | ND |
| Volume of BAL (ml) | 40 | 46 | 45 | 75 | 90 | 88 | 40 | 82 |
| Cell Count/ (x105/ml) | 30.2 | 2.4 | 1.7 | 1.5 | 2.7 | 1.2 | 8.3 | 3.6 |
| Glyco-DHC (µmol/l) | ND | ND | ND | 1.17 | 0.03 | ND | 0.03 | ND |
| Glyco-THC (µmol/l) | ND | .34 | ND | 0.75 | ND | ND | 0.02 | ND |
| Tauro-DHC (µmol/l) | 1.77 | 2.17 | ND | 2.14 | 0.94 | 0.71 | 0.40 | 1.15 |
| Tauro-THC (µmol/l) | 11.40 | 2.39 | ND | 3.86 | 1.44 | 2.33 | 1.24 | 1.66 |

**Table S4. Markers of inflammation and aspiration – by participant.**

IL, interleukin; ND, not detected; TNF, tumour necrosis alpha; TGF, transforming growth factor; BAL, bronchoalveolar lavage; DHC, dihydroxycholanoate; THC, trihydroxycholanoate.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Omeprazole | | | | | Placebo | | |
| Macrophages | 17.2 | 39.0 | 69.0 | 92.4 | 98.4 | 98.0 | 72.0 | 98.0 |
| Neutrophils | 79.6 | 48.0 | 25.0 | 6.6 | 0.4 | 1.2 | 18.0 | 0.2 |
| Lymphocytes | 3.2 | 13.0 | 6.0 | 0.8 | 1.2 | 0.8 | 10.0 | 1.8 |
| Eosinophils | 0 | 0 | 0 | 0.2 | 0 | 0 | 0 | 0 |
|  | | | | | | | | |
| Ciliated epithelia | 0.8 | 2.0 | 1.2 | 2.9 | 0.4 | 0.2 | 4.0 | 1.0 |
| Metaplastic epithelia | 0 | 3.0 | 0.8 | 0 | 0 | 2.1 | 0.9 | 0.2 |

**Table S5. Differential cell counts in BAL fluid – by participant.**

All figures are percentages. The sum for leukocytes (%macrophages + %neutrophils + %lymphocytes + %eosinophils) adds up to 100%. Ciliated epithelia and metaplastic epithelia are shown as the number of cells relative to the total number of leukocytes, expressed as a percentage.

**Safety**

Adverse events were coded according to the latest available version of the Medical Dictionary for Regulatory Activities (MedDRA) and reported by preferred term.

For each adverse event, the worst severity grade reported during treatment is tabulated in Table S6. The overall worst grade reported (of any event) is given at the end of Table S6. Adverse events resulting in treatment discontinuation are shown in Table S7.

|  | Omeprazole (n=23) | | | | | | | | | | Placebo (n=22) | | | | | | | | | |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| None reported | | Mild | | Moderate | | Severe | | Unknown grade | | None reported | | Mild | | Moderate | | Severe | | Unknown grade | |
| No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % |
| Abdominal pain | 20 | 87.0 | 0 | 0.0 | 3 | 13.0 | 0 | 0.0 | 0 | 0.0 | 19 | 86.4 | 1 | 4.5 | 0 | 0.0 | 2 | 9.1 | 0 | 0.0 |
| Acid reflux | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Back injury | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Back pain | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Cellulitis | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Chest pain | 22 | 95.7 | 1 | 4.3 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Constipation | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Cough | 20 | 87.0 | 0 | 0.0 | 2 | 8.7 | 1 | 4.3 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Cough syncope | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Depression | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Diarrhoea | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 20 | 90.9 | 2 | 9.1 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Dizziness | 22 | 95.7 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Dyspepsia | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 19 | 86.4 | 1 | 4.5 | 1 | 4.5 | 1 | 4.5 | 0 | 0.0 |
| Dyspnoea | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 20 | 90.9 | 0 | 0.0 | 2 | 9.1 | 0 | 0.0 | 0 | 0.0 |
| Fatigue | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 20 | 90.9 | 1 | 4.5 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 |
| Gastric ulcer | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 |
| Gout | 22 | 95.7 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Headache | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 18 | 81.8 | 2 | 9.1 | 0 | 0.0 | 2 | 9.1 | 0 | 0.0 |
| Hot flushes | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 |
| Hyperglycaemia | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Influenza | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 |
| Ischemia | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Local swelling | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Lower respiratory tract infection | 17 | 73.9 | 1 | 4.3 | 0 | 0.0 | 3 | 13.0 | 2 | 8.7 | 19 | 86.4 | 1 | 4.5 | 0 | 0.0 | 2 | 9.1 | 0 | 0.0 |
| Lung neoplasm (malignant) | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Muscle spasm | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Oedema | 22 | 95.7 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Oropharyngeal pain | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 |
| Pain in extremity | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Exacerbation of pulmonary fibrosis | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 |
| Salivary gland calculus | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Sepsis | 22 | 95.7 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Sore throat | 23 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 0 | 0.0 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 |
| Urinary tract infection | 21 | 91.3 | 0 | 0.0 | 2 | 8.7 | 0 | 0.0 | 0 | 0.0 | 22 | 100.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Viral upper respiratory tract infection | 22 | 95.7 | 1 | 4.3 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 | 21 | 95.5 | 1 | 4.5 | 0 | 0.0 | 0 | 0.0 | 0 | 0.0 |
| Vomiting | 21 | 91.3 | 0 | 0.0 | 0 | 0.0 | 1 | 4.3 | 1 | 4.3 | 18 | 81.8 | 1 | 4.5 | 1 | 4.5 | 2 | 9.1 | 0 | 0.0 |
| ***Overall worst grade per participant*** | ***7*** | ***30.4*** | ***0*** | ***0.0*** | ***6*** | ***26.1*** | ***9*** | ***39.1*** | ***1*** | ***4.3*** | ***8*** | ***36.4*** | ***3*** | ***13.6*** | ***2*** | ***9.1*** | ***9*** | ***40.9*** | ***0*** | ***0.0*** |

**Table S6. Adverse events by worst reported grade.**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Treatment group | Description of event | Overall no. IMP taken | Severity | Serious | Causality |
| Omeprazole | New diagnosis of lung cancer. Clinical decision for conservative treatment of lung cancer in view of co-morbidities. | 107 | Severe | Yes (SAE) | Not related |
| Omeprazole | Participant admitted to hospital with one week of increased dyspnoea, received intravenous antibiotics; participant died. | 5 | Severe | Yes (SAE) | Not related |
| Omeprazole | Stomach pain. | 8 | Moderate | Yes (SAE) | Related |
| Placebo | Burning sensation akin to hot flushes, and exhaustion. | 1 | Moderate | No | Related |
| Placebo | Gastric ulcers: participant underwent endoscopy which showed gastric ulcers, which required regular prescription of PPI. | 72 | Unavailable | No | Not related |
| Placebo | Abdominal pain. | 26 | Severe | Yes (SAE) | Related |
| Placebo | Abdominal pain (epigastric radiating into back). | 60 | Moderate | Yes (SAE) | Related |

**Table S7. Adverse events resulting in treatment discontinuation.**

IMP, investigational medicinal product; PPI, proton pump inhibitor. SAE, serious adverse event.

**Serious adverse events**

In total, 6 serious adverse events (SAEs) and 3 serious adverse reactions (SARs) were reported in 8 participants, 4 allocated to omeprazole and 4 to placebo. Assessment of causality was blind to treatment allocation. All SAE/SARs are described in Table S8.

| Treatment group | Days from IMP start | Description | Seriousness | Severity | Causality | Expected | Event type | Outcome |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Omeprazole | 55 | New diagnosis of lung cancer. | Resulted in death | Severe | Not related | NA | SAE | Death |
| Omeprazole | 16 | Hospital admission - cellulitis +/- lower respiratory tract infection | Hospitalisation | Severe | Not related | NA | SAE | Condition improved |
| Placebo | 63 | Hospital admission - Ischaemic right leg | Hospitalisation | Severe | Not related | NA | SAE | Condition unchanged |
| Omeprazole | 81 | Hospital admission with sepsis- likely related to skin infection | Hospitalisation | Severe | Not related | NA | SAE | Condition improved |
| Placebo | 34 | Hospital admission - exacerbation of pulmonary fibrosis and lower respiratory tract infection | Hospitalisation | Severe | Not related | NA | SAE | Condition improved |
| Omeprazole | 2 | Participant admitted to hospital with one week of increased dyspnoea; received intravenous antibiotics; participant died. | Hospitalisation | Severe | Not related | NA | SAE | Death |
| Placebo | 0 | Abdominal pain | Other significant medical event | Severe | Related | Expected | SAR | Condition unchanged |
| Omeprazole | 0 | Stomach Pain. | Other significant medical event | Moderate | Related | Expected | SAR | Condition unchanged |
| Placebo | 28 | Abdominal pain (epigastric area, radiating to back). | Other significant medical event | Severe | Related | Expected | SAR | Condition unchanged |

**Table S8: Serious adverse events / serious adverse reactions.**

IMP, investigational medicinal product; NA, not applicable; SAE, serious adverse event; SAR, serious adverse reaction.

**SUPPLEMENTARY REFERENCES**

S1 Bredenoord AJ, Fox, M, Kahrilas PJ, et al. Chicago classification criteria of esophageal motility disorders defined in high resolution esophageal pressure topography. *Neurogastroenterol Motil* 2012;24(Suppl 1):57-65.

S2 Zerbig F, des Varannes SB, Roman S, et al. Normal values and day-to-day variability of 24-h ambulatory oesophageal impedance-pH monitoring in a Belgian-French cohort of healthy subjects. *Aliment Pharmacol Ther* 2005;22:1011-1021.

S3 Hunt EB, Ward C, Power S, et al. The potential role of aspiration in the asthmatic airway. *Chest* 2017;151:1272-1278.

S4 Parikh S, Brownlee IA, Robertson AG, et al. Are the enzymatic methods currently being used to measure bronchoalveolar lavage bile salt levels fit for purpose? *J Heart Lung Transplant.* 2013;32:418-423.

Statistical Analysis Plan for the PPIPF Trial

**A randomised placebo-controlled pilot trial of omeprazole in idiopathic pulmonary fibrosis (IPF) (PPIPF Study)**

Version 1.0 [13/06/2017]

This statistical analysis plan is based on protocol version 5.0 [07/03/2016]

ISRCTN Number: ISRCTN 07139948

EudraCT Number: 2013-003301-26

REC Reference: 13/YH/0284

Sponsor: The Newcastle upon Tyne Hospitals NHS Foundation Trust

Sponsor protocol number: IAFIPF001

Funder: British Lung Foundation

Funder reference number: IPFPSG12-7

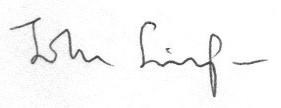
**Trial Statisticians: Vicky Ryan and Helen Mossop**

**Reviewed and Approved by:**

Senior Trial Statistician: Ms Vicky Ryan

Date: 13.06.17 

\*See the file note in the Statistics TMF Section 3: SAP 130617

Chief Investigator: Professor John Simpson

Date: 14.6.17

This statistical analysis plan (SAP) details the analysis for presentation and publication of the PPIPF trial. This plan, along with all other documents relating to the analysis of this trial, will be stored in the ‘Statistical Documentation’ of the Trial Master File (TMF) and the final signed SAP will be stored in section 16 of the TMF (16. Statistics / 16.1 Final signed Statistical Analysis Plan). Any deviations from the statistical analysis plan will be documented in the end of study report.

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# **trial summary**

## Trial design

PPIPF is a prospective, single-centre, double-blind, randomised, pilot trial of omeprazole in patients with idiopathic pulmonary fibrosis (IPF). Patients will be randomised 1:1 to omeprazole 20mg twice daily or matching placebo, to be taken orally before food for 90 days.

The primary objective of this pilot trial is to test the hypothesis that omeprazole reduces cough in IPF. The results of this trial may be used to inform the design of definitive multi-centre trials in IPF. To this end, a focus of the pilot trial, which seeks to provide proof of concept with respect to reduction in cough, will be on rates of participant eligibility, recruitment and retention and on the yield and quality of data in respect of the proposed secondary outcomes.

The primary analysis will take place when all patients have follow-up data available, 90 days following the start of trial treatment.

## Patient population

The target population is patients with IPF. Patients will be recruited from one centre in the UK (the Royal Victoria Infirmary, Newcastle upon Tyne). Patients can also be identified and referred for participation from Participant Identification Centres (PICs) by their treating clinicians.

*Inclusion criteria*

A pragmatic clinical definition of IPF will be used, in which recruited patients must fulfil all of the following criteria

* IPF is considered the most likely diagnosis by the regional interstitial lung disease multidisciplinary team meeting (ILD-MDT)
* history of cough, with or without exertional dyspnoea
* high resolution computed tomography (HRCT) scan features of honeycombing in a predominantly basal and sub-pleural distribution
* bi-basal crackles on auscultation
* features of a restrictive ventilatory defect (vital capacity (VC) <90% predicted and/or transfer factor of the lung for carbon monoxide (TLco) <90% predicted)
* aged 40-85 years

*Exclusion criteria*

* known allergy to omeprazole or other proton pump inhibitor (PPI)
* concomitant use of warfarin, diazepam, phenytoin or ketoconazole
* Concomitant use of a regular PPI\*, antacid, prokinetic or raft alginate† during the trial period.
* history of upper respiratory tract infection, lower respiratory tract infection or exacerbation of IPF in the 4 weeks before starting study drugs
* active trial of treatment for IPF (e.g. prednisolone, pirfenidone, nintedanib, N-acetylcysteine) started in the 4 weeks before starting study drugs
* documented history of hepatic cirrhosis
* pregnancy or lactation
* ILD-MDT considers the most likely cause of the patient’s ILD to be a condition other than IPF, for example rheumatoid lung, systemic sclerosis ILD, asbestosis, chronic hypersensitivity pneumonitis, sarcoidosis, etc.
* concurrent enrolment in a Clinical Trial of an Investigational Medicinal Product(CTIMP) for IPF

\*Patients taking a PPI during screening will potentially be eligible. In these cases the indication for on-going treatment will be reviewed and patients may be enrolled if they agree to a trial without PPI and consent to participation after that trial is tolerated and completed.

†Patients taking antacids, prokinetics or raft alginates at the time of screening will be eligible if they have been off these treatments for a period of at least 2 weeks.

For full details of inclusion and exclusion criteria please refer to sections 4.5 and 4.6 of the study protocol (version 5, 07/03/2016).

## Trial objectives

**Primary efficacy outcome**

To assess whether omeprazole can lead to a reduction in cough frequency in patients with IPF.

**Primary feasibility objectives**

To assess the feasibility and acceptability of trial procedures.

**Secondary objectives**

The following objectives will be explored for suitability as potential secondary clinical outcome measures for future trials

* To assess change in patient-reported symptoms of cough and reflux
* To assess change in functional status, assessed using the 6 minute walk test
* To assess change in lung function tests (forced vital capacity (FVC) and TLco)
* To assess change in acid and non-acid reflux
* To investigate markers of lung inflammation and lung infection in bronchoalveolar lavage (BAL) fluid

## Sample size and power

Due to the nature of this pilot study no formal sample size calculations have been performed as analyses of outcome data will be, by definition, exploratory in nature. However, recommendations for good practice suggest that 20-30 patients per treatment group should provide sufficient data to assess the feasibility of a trial, investigate the distribution of outcome measures and estimate with adequate precision standard deviations of key study parameters.1, 2

The study therefore aims to randomise 60 patients. The anticipated attrition rate of patients in this study is not determined (and is part of the feasibility assessment), however randomising 60 patients will allow for up to 33% attrition while achieving the minimum recommended sample size of 20 patients per treatment group with complete follow-up.

# **recruitment and Randomisation**

## Recruitment

The original recruitment target was to randomise 3 patients per month over 21 months starting in August 2013 and ending in May 2015. Due to a delay in obtaining the required approvals, recruitment opened on 1st March 2014 and the first patient was randomised on 28th March 2014. At a data monitoring committee (DMC) meeting in February 2015 recruitment was running below target at an average rate of 1.8 patients per month. The Committee recommended that an application for a no cost extension be made to the funders, the British Lung Foundation (BLF). An extension to recruitment to the end of August 2016 was agreed by the BLF.

The trial closed to recruitment on 19th August 2016 with 45 patients randomised. The last patient to enter the trial was randomised on 4th July 2016.

## Randomisation

Randomisation was through a secure password-protected web-based system administered centrally by the Newcastle Clinical Trials Unit (NCTU). Participants were randomised to omeprazole or placebo in a 1:1 ratio, using random permuted blocks to ensure concealment of allocation. Assignment to either omeprazole or placebo was blinded to both the participant and the research team (double-blinded). Randomisation generated two numbers: a unique 3-digit “Study ID” number for each participant and a unique 2-digit “Bottle” number (which matched a “Trial patient no. (Pack number)” on a medication pack held in Pharmacy at the Royal Victoria Infirmary).

# **TIMING AND REPORTING of interim AND FINAL ANALYSES**

The end of the study is defined as last patient, last visit (90 day follow-up visit).

Final analyses will be carried out when all participants have been followed up and all study data have been entered into MACRO (apart from the cough data which will be provided (quality assured) by Manchester University in an Excel spreadsheet).

DMC meetings were held on:

* 23rd June 2014
* 13th October 2014
* 27th February 2015
* 16th July 2015
* 18th March 2016

# **defintion and calculation of outcome measures**

## Primary clinical outcome

The primary clinical outcome measure is cough frequency determined using 24-hour cough monitoring, once before starting treatment and again at the end of treatment. Further information on the methods of assessment is given in Section 6 of the protocol.

A report documenting the number of coughs in each 1 hour time period and a calculation of cough frequency (the average number of coughs per hour: total number of coughs/total time recording in hours) will be generated for statistical analysis.

The primary clinical outcome is the mean difference between treatment groups in the change in cough frequency from baseline to follow-up.

## Primary feasibility outcomes

Feasibility of recruitment

The following data will be presented:

* + Eligibility (number of patients eligible/total number of patients approached);
  + Recruitment (number of patients recruited/total number eligible);
  + Study completion (number of patients completing trial/total number randomised);
  + Recruitment rate (average number of patients recruited per month)

Reasons for ineligibility and non-acceptance of recruitment will be summarised.

Acceptability of trial procedures

* + Consent to 24-hour oesophageal physiology assessment (number consenting to procedure/number randomised)
  + Consent to bronchoscopy assessment (number consenting to procedure/number randomised)
  + Acceptance of 24-hour oesophageal physiology assessment
    - number undergoing procedure at baseline/number consented
    - number undergoing procedure at follow-up/number consented
  + Acceptance of bronchoscopy assessment (number undergoing procedure/number consented)

## Secondary outcomes

Leicester Cough Questionnaire

The Leicester Cough Questionnaire consists of 19 questions covering 3 domains; physical, psychological and social. Domain scores will be calculated according to the scoring manual as the sum of scores from items in the domain/number of items in domain (each with range 1-7). The total score will then be calculated as the sum of domain scores (range 3-21). The change in the total score from baseline to follow-up will be calculated (follow-up-baseline).

Gastrointestinal Quality of Life Index (GIQLI)

The GIQLI consists of 36 questions with responses rated on a 5-point scale from 0-4. The total score is calculated as per the scoring manual as a sum of all responses (range 0-144). The change in total score from baseline to follow-up will be calculated (follow-up-baseline).

The Reflux Symptoms Index (RSI)

The RSI consists of 9 questions with responses rated on a 6-point scale from 0-5. The total score is calculated as per the scoring manual as a sum of all responses (range 0-45). The change in total score from baseline to follow-up will be calculated (follow-up-baseline).

De Meester reflux-associated symptoms questionnaire

The questionnaire consists of three questions assessing heartburn, regurgitation and dysphagia. Each symptom is scored on a scale of 0-3. A total symptomatic score is calculated as the sum of the three responses (range 0-9). The change in total score from baseline to follow-up will be calculated (follow-up-baseline).

Six minute walk distance

This is measured as the distance a patient can walk in a period of 6 minutes. The change in distance from baseline to follow-up will be calculated (follow-up-baseline).

Lung function

FVC and TLco will be measured at baseline and follow-up. For each, change from baseline will be calculated (follow-up-baseline).

Oesophageal manometry

Oesophageal manometry will be performed at baseline and follow-up for those consenting to the procedure. Results of the manometry study will be in a narrative form.

pH-impedance monitoring

24-hour pH-impedance monitoring will be performed at baseline and follow-up for those consenting to the procedure. Results of the pH-impedance monitoring will be adjusted for the total length of time the impedance monitoring was performed over and will include:

* Duration of pH-impedance monitoring (hh:mm)
* Proportion of time pH<4 (%)
* Number of times pH drops to <4 in 24 hours
* Number of long reflux episodes (> 5 minutes) in 24 hours
* Longest reflux episode (min)
* Number of acid refluxes (pH<4) in 24 hours
* Number of weak acid refluxes (pH≥4 to <7) in 24 hours
* Number of proximal reflux events in 24 hours
* DeMeester score
* Total number of reflux episodes in 24 hours
* Bolus clearance time (s)
* Symptom-associated probability (%)

All the above measurements will be treated as continuous variables. For each measurement, change from baseline will be calculated (follow-up-baseline) and summarised descriptively.

Bronchoscopy and BAL

Bronchoscopy and BAL are performed at the end of treatment. Key mediators of lung inflammation, markers of aspiration and growth of potentially pathogenic bacteria and fungi in BAL fluid will be reported and will include:

* Pepsin
* Bile acids
* Cytokines (including IL-1β, IL-6, IL-8, IL-10, TNFα, TGFβ)
* Bacterial culture
* Fungal culture

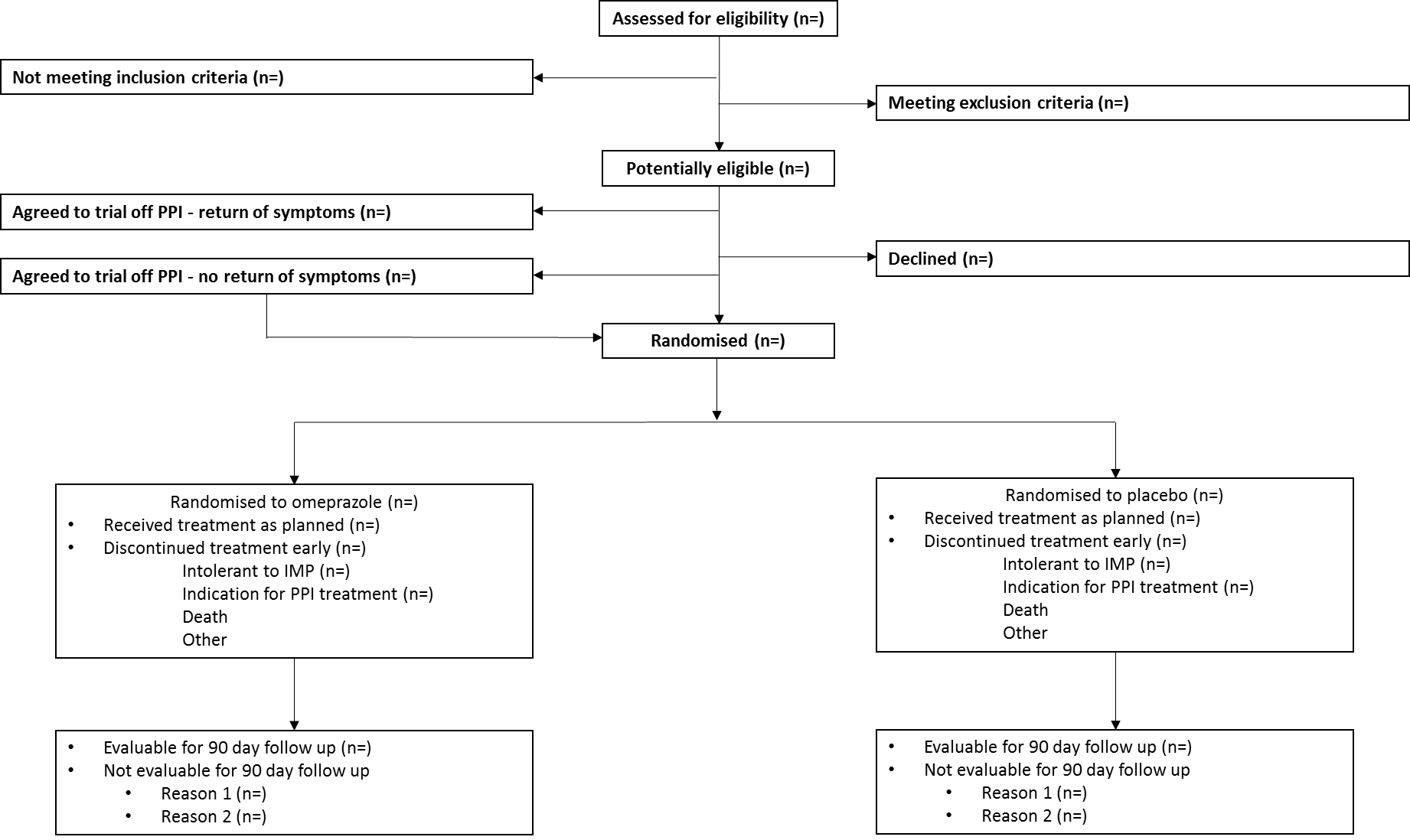
These measures will be summarised descriptively.

# **STUDY POPULATION**

Patient flow through the trial will be presented using a CONSORT diagram. Information will be provided on numbers and reasons (where appropriate) for: approached patients not being eligible; eligible patients not being randomised; patients found to be ineligible after randomisation; patients deviating from allocated treatment; patients not evaluable for the primary endpoints; withdrawal from follow-up; withdrawal of consent and all protocol violations.

The number of ineligible patients and reasons for ineligibility will be reported.

**Example Figure 1: CONSORT flow diagram**



## Enrolment

The representativeness of the study sample will be described with the following data:

* The number of patients identified at screening
* The number of patients excluded at screening due to ineligibility (with reasons where available)
* The number of eligible patients identified at screening
* The number of eligible patients not taking part in the study (with reasons where available)
* The number of eligible patients randomised into the study.

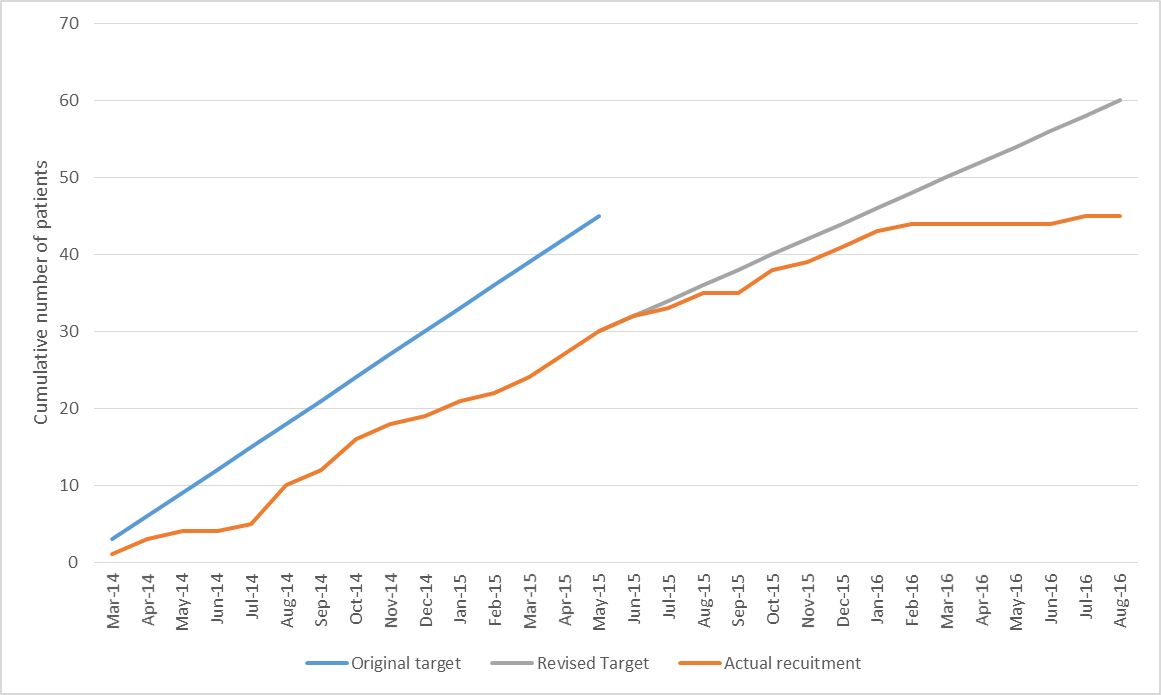
As feasibility of recruitment is an endpoint of this study, further detail on description of these data is given in Section 6.2.

The number of patients taking PPI, antacids, prokinetics or raft alginates at screening will be presented, along with the number agreeing to a trial off treatment and the number subsequently randomised. This data will also be presented in the CONSORT diagram.

|  |  |  |  |
| --- | --- | --- | --- |
|  | No. taking treatment at screening | No. agreeing to trial off treatment | No. randomised |
| PPI | n (%) | n (%) | n (%) |
| Antacid | n (%) | n (%) | n (%) |
| Prokinetics | n (%) | n (%) | n (%) |
| Raft alginates | n (%) | n (%) | n (%) |

Duration of recruitment will be presented graphically as cumulative recruitment per month. Target and actual recruitment will be described.

**Example Figure 2: Cumulative target and observed recruitment per month**

****

## Baseline characteristics

Demographic and clinical characteristics at baseline will be reported across the two randomised treatment groups descriptively. We will report the number and percentage in each group for all categorical variables (e.g. gender) and mean, SD or median, IQR/range, as appropriate for all continuous variables. The distribution of the data will be explored graphically. No significance testing will be carried out on baseline characteristics.

**Example Table 1: Patient demographics at baseline**

|  | Omeprazole (n=) | | Placebo (n=) | |
| --- | --- | --- | --- | --- |
| ***Demographics*** | | | | |
| Gender |  |  |  |  |
| Male | n | % | n | % |
| Female | n | % | n | % |
| Age (years) |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| Age categories 40-64 | n | % | n | % |
| (for EudraCT report) 65-84 | n | % | n | % |
| 85 | n | % | n | % |
| Ethnicity |  |  |  |  |
| Caucasian | n | % | n | % |
| Smoking history |  |  |  |  |
| Never smoked | n | % | n | % |
| Ex-smoker | n | % | n | % |
| *Estimated pack years* | Median | IQR/Range | Median | IQR/Range |
| Current smoker | n | % | n | % |
| *Estimated pack years* | Median | IQR/Range | Median | IQR/Range |
| Estimated number of comorbidities |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
| ***Concomitant medications*** | | | | |
|  | n | % | n | % |
| Other 1 | n | % | n | % |
| Other 2 | n | % | n | % |
| Other | n | % | n | % |
| ***Physical examination*** | | | | |
| BMI (kg/m2) |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| Blood pressure (mmHg) |  |  |  |  |
| Systolic | Mean | SD | Mean | SD |
| Diastolic | Mean | SD | Mean | SD |
| Heart rate (beats per minute) | Mean | SD | Mean | SD |
| Respiratory rate (per minute) | Mean | SD | Mean | SD |
| ***Lung function*** | | | | |
| FEV1 (% predicted) | Mean | SD | Mean | SD |
| FVC(% predicted) | Mean | SD | Mean | SD |
| FEV1 / FVC (%) | Mean | SD | Mean | SD |
| TLco(% predicted) | Mean | SD | Mean | SD |
| Kco(% predicted) | Mean | SD | Mean | SD |
| ***Six minute walk test*** | | | | |
| Distance (m) | Mean | SD | Mean | SD |
| ***24h cough monitor*** | | | | |
| Coughs per hour |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| ***Patient-reported*** | | | | |
| LCQ |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| RSI |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| GIQLI |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |
| DeMRQ |  |  |  |  |
|  | Median | IQR/Range | Median | IQR/Range |
|  | Mean | SD | Mean | SD |

Data are n; %, mean; SD or median (IQR); range, unless otherwise stated

***Note: if any assessment is not performed for a patient the number with data will be included as a footnote.***

## Treatment received

Patients are randomised to receive omeprazole 20mg (1 tablet) twice daily (i.e. 2 tablets per day) or matching placebo for 90 days. The final study visits can be performed at any point in the 2 weeks prior to completing 90 days of treatment (ie ≥ 76 days from the start of treatment). The final tablet count will take place at the end of trial treatment. The number of tablets returned will be reported and the number of tablets taken will be calculated as (190 - tablets returned), accounting for the 10 extra tablets dispensed.

For each individual participant, the potential treatment duration (ie the number of days the patient could have potentially taken the IMP) will be calculated from the day after their IMP was issued (as patients were instructed to start medication the next day) to the date of their last visit (when IMP should be returned), or the date of withdrawal or death if treatment was discontinued early. This duration will be summarised as the median, IQR and range in each randomised group.

The number of patients discontinuing trial treatment early (withdrawn from trial treatment by the investigator, or participant withdrew from trial treatment, or participant died prior to 76 days), will be reported as frequency and percentage in each group, with reasons. Compliance with the IMP dosing protocol will be calculated as number of tablets taken/(2 x treatment duration) and will be reported separately for participants with treatment duration of 76 days or more and for those who discontinued trial treatment prior to 76 days.

Compliance with the IMP dosing protocol will be summarised in each randomised group by the median, IQR and range and also tabulated as a binary variable; compliance ≥80% and compliance <80%.

**Example Table 2: Summary of allocated treatment received**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Omeprazole (n=) | | Placebo (n=) | |
| Completed treatment  (treatment duration ≥76 days) | n | % | n | % |
| Discontinued treatment |  |  |  |  |
| Reasons for discontinuation of treatment |  |  |  |  |
|  |  |  |  |  |
| *Intolerance to IMP* | n | % | n | % |
| *Death* | n | % | n | % |
| *Other\** | n | % | n | % |
| Treatment duration (days) |  |  |  |  |
| Median | Median | IQR/Range | Median | IQR/Range |
| <76 | n | % | n | % |
| 76-90 | n | % | n | % |
| Dose intensity (%)  (treatment duration ≥76 days) |  |  |  |  |
| Median | Median | IQR/Range | Median | IQR/Range |
| ≥80% | n | % | n | % |
| <80% | n | % | n | % |
| Dose intensity (%)  (treatment duration <76 days)  Median | Median | IQR/Range | Median | IQR/Range |
| ≥80% | n | % | n | % |
| <80% | n | % | n | % |
|  |  |  |  |  |

Data are n; % or median (IQR); range, unless otherwise stated

\*Other reasons will be given in a line listing (see example line listing below)

**Example Line listing: Reasons for treatment discontinuation**

|  |  |  |  |
| --- | --- | --- | --- |
| Trial ID | Randomised group | Category | Details |
|  | Omeprazole/Placebo | Intolerance to IMP, death, other |  |
|  |  |  |  |
|  |  |  |  |

## Follow-Up

Participants are scheduled to return for their final study assessments from day 88 on study medication to day 90, but the final study visit window opens 2 weeks prior to completing study medication.

Completeness of assessments at the 90 day visit will be tabulated for each outcome measure as frequency and percentage in each randomised group.

The number of participants completing 24 hour oesophageal physiology and bronchoscopy assessments will be presented as a percentage of the total number of patients consenting to these individual procedures at the start of the study period. Reasons for non-completion will be listed.

**Example Table 3: Follow-up assessments completed**

|  | Omeprazole (n=) | | Placebo (n=) | |
| --- | --- | --- | --- | --- |
| ***Timelines*** | | | | |
| Time from treatment day 1 to final follow-up visit (days) |  |  |  |  |
|  |  |  |  |  |
|  |  |  |  |  |
|  |  |  |  |  |
| ***Lung function*** | | | | |
| FEV1 |  |  |  |  |
| FVC |  |  |  |  |
| TLco |  |  |  |  |
| Kco |  |  |  |  |
| ***Six minute walk test*** | | | | |
| Six minute walk test |  |  |  |  |
| ***24h cough monitor*** | | | | |
| 24h cough monitor |  |  |  |  |
| ***Patient reported*** | | | | |
| LCQ |  |  |  |  |
| RSI |  |  |  |  |
| GIQLI |  |  |  |  |
| DeMRQ |  |  |  |  |
| ***24h oesophageal physiology*** | | | | |
| Consented |  |  |  |  |
| Completed |  |  |  |  |
| ***Bronchoscopy*** | | | | |
| Consented |  |  |  |  |
| Completed |  |  |  |  |

Data are n; %, mean (SD) or median (IQR); range, unless otherwise stated

***Note: reasons for not completing assessments will be given in a line listing***

## Protocol deviations

Protocol deviations will be reported overall and by randomised groups. Protocol deviations will include; deviations from the treatment protocol, deviations from visit schedule or withdrawal from trial specific follow-up, losses to follow-up and ineligible patients.

**Example Table 4: Line listing of protocol deviations**

|  |  |  |  |
| --- | --- | --- | --- |
| Trial ID | Randomised group | Deviation type | Details |
|  |  | Ineligible/withdrawn from treatment/complete withdrawal/lost to follow-up or died/deviation from visit schedule |  |
|  |  |  |  |
|  |  |  |  |

## Defining Populations for Analysis

Intention to treat: Analyses will follow the ITT principle, which includes all participants in the group to which they were randomised, regardless of the intervention that they received. The number of participants who did not receive the intervention to which they were randomised will be reported.

Complete-case: For the analyses of change data (change from baseline to follow-up) the analyses will be carried out on a complete-case basis - that is where we have data at both baseline and the follow-up visit (timing as per Section 5.4).

Safety population: For the analysis of safety data all randomised patients who received at least one dose of randomised intervention will be included.

# **AnalysIs methods**

## Primary clinical outcome

The distribution of change in cough frequency will be explored graphically using histograms, box-plots and dot-plots. In the event that data are approximately normally distributed change in cough frequency will be summarised in each treatment group by the mean and standard deviation. In the event that the difference in cough frequency change is not normally distributed, non-parametric statistics (median, IQR) will be used to summarise the data. Data transformations using ladder powers may be explored if necessary. Change in cough frequency from baseline to follow-up will also be presented graphically, for example using spaghetti plots.

The treatment effect of the mean difference between treatment groups in the change in cough frequency from baseline to follow-up will be estimated using analysis of covariance (ANCOVA), with treatment group as the independent variable and adjusting for baseline cough frequency.3

The assumptions of the ANCOVA model will be explored using the standardised residuals, leverage and Cook’s distance.

An exploratory hypothesis test for the unpowered comparison of the change in cough frequency between randomised groups will also be provided as requested by the funder. Data will be interpreted taking into account the caveats (a) that this is a pilot study4, and (b) that sample size was not based on power calculations. All results will be treated as preliminary and in keeping with this being a pilot RCT.

***Raw data at follow-up will also be presented as per Example Table 1.***

**Example Figure 3: Spaghetti plot of baseline and follow-up cough frequency**



***Note: this is not genuine cough frequency data***

**Example Table 5: Mean difference in cough frequency between treatment groups**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Change in cough frequency from baseline | Mean change from baseline | | Difference in change (Omeprazole - Placebo) | |
| Omeprazole | Placebo | Mean | 95% CI |
| Adjusted for baseline cough frequency |  |  |  |  |
| Adjusted for baseline cough frequency and PPI nativity |  |  |  |  |

Alternative summary measures of 24-hour cough data may be explored in order to investigate the most appropriate summary measure to take forward into potential definitive trials. For example, the burden of cough frequency may be explored; the per-patient area under the 24-hour frequency curve will be calculated at baseline and follow-up and the change calculated (follow-up-baseline). The sum of all changes will be calculated in each treatment group and summarised descriptively using mean (SD) or median (IQR) as appropriate.

**Example Figure 4: Per-patient longitudinal plots of cough frequency per hour at follow-up**



***Note: this is not genuine cough frequency data***

## Primary feasibility outcomes

Feasibility and acceptability outcomes will be summarised by frequency and percentages or line listings in the entire study population (i.e. not by randomised group).

**Example Table 6: Feasibility of recruitment and acceptability of trial procedures**

|  |  |  |
| --- | --- | --- |
|  | N | % |
| Screened for eligibility |  |  |
| Eligible |  |  |
| Ineligible |  |  |
| Unable to stop current PPI |  |  |
| Allergy or known intolerance to omeprazole or other PPI |  |  |
| Concomitant use of prohibited medication\* |  |  |
| No features of honeycombing |  |  |
| Reason 1 |  |  |
| Reason 2 |  |  |
| Reason 3 |  |  |
| Other |  |  |
| Randomised |  |  |
| Not randomised |  |  |
| Participant declined |  |  |
| Died during screening |  |  |
| Reason 1 |  |  |
| Other |  |  |
| Mean recruitment rate per month |  |  |
| Consent to 24-hour oesophageal physiology assessment |  |  |
| Acceptance (baseline) |  |  |
| Acceptance (follow-up) |  |  |
| Consent to bronchoscopy assessment |  |  |
| Acceptance (follow-up) |  |  |
| **\*** e.g.X were on warfarin, X were on phenytoin and X on diazepam | |  |

## Secondary outcomes

All analyses will be conducted in the complete-case population.

**Patient-reported symptoms**

For each patient reported symptom questionnaire, the change of score will be summarised descriptively using the mean and standard deviation or median and IQR in the event of non-normally distributed data in the two treatment groups. The distribution of the data may also be summarised graphically using histograms or boxplots, as appropriate. Change in score from baseline to follow-up will also be presented graphically using spaghetti plots. Assumptions of the model will be assessed as for the primary clinical endpoint.

The difference in the change in score between randomised groups will be estimated using analysis of covariance with treatment group as the independent variable and adjusting for baseline score. 95% confidence intervals for the difference between groups will be presented.

**Six minute walk distance**

The change in distance walked will be summarised descriptively using the mean and standard deviation or median and IQR in the event of non-normally distributed data in the two treatment groups. The distribution of the data may also be summarised graphically using histograms or boxplots, as appropriate. Change in distance from baseline to follow-up will also be presented graphically using spaghetti plots.

The difference in the change in distance between randomised groups will be estimated using analysis of covariance with treatment group as the independent variable and adjusting for baseline score. 95% confidence intervals for the difference between groups will be presented. Assumptions of the model will be assessed as for the primary clinical endpoint.

**Lung function**

For each measure of lung function, the change in measurement will be summarised descriptively using the mean and standard deviation or median and IQR in the event of non-normally distributed data in the two treatment groups. The distribution of the data may also be summarised graphically using histograms or boxplots, as appropriate. Change from baseline to follow-up will also be presented graphically using spaghetti plots. Assumptions of the model will be assessed as for the primary clinical endpoint.

The difference in the change between randomised groups will be estimated using analysis of covariance with treatment group as the independent variable and adjusting for baseline score. 95% confidence intervals for the difference between groups will be presented.

**Example Table 7: Mean difference in secondary outcomes between treatment groups**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Change from baseline, adjusted for baseline | Mean change from baseline | | Difference in change (Omeprazole - placebo) | |
| Omeprazole  (n=) | Placebo  (n=) | Mean | 95% CI |
| LCQ |  |  |  |  |
| RSI |  |  |  |  |
| GIQLI |  |  |  |  |
| DeMRQ |  |  |  |  |
| Six minute walk test |  |  |  |  |
| FVC |  |  |  |  |
| TLco |  |  |  |  |

***Raw data at follow-up will also be presented as per Example Table 1.***

pH Impedance monitoring

Change in pH Impedance monitoring variables after treatment will be summarised descriptively using the mean and standard deviation or median and IQR in the event of non-normally distributed data. The distribution of the data may also be summarised graphically using histograms, boxplots or dot-plots, as appropriate. Change in baseline and follow-up data may also be displayed graphically, for example using spaghetti plots.

**Bronchoscopy and BAL**

Markers of lung inflammation and aspiration in BAL fluid will be summarised using frequency and percentage for categorical variables and mean and standard deviation or median and IQR for normally and non-normally distributed continuous variables respectively. Transformations of non-normally distributed continuous marker variables may be explored if required and if suggested by the literature at the time of analysis.

## Missing data

Given this is a pilot study with only two measurement occasions, baseline and 90 days, there will be no data imputation where outcome data at the 90 day visit is missing.

Missing outcome data due to participant loss to follow-up will not be imputed.

Missing items from a partially completed validated questionnaire will be handled as described in the scoring manual.

# **SAFETY data**

Toxicities will be coded according to the latest version of the Medical Dictionary for Regulatory Activities (MedDRA) available. Severity, seriousness and relationship to study treatment will be reported.

All AEs will be tabulated by preferred term and (worst reported) severity, graded on a three-point scale (mild, moderate, severe), in each randomised treatment group. In addition, the data will be presented in the same way but only including those AEs recorded as related to study treatment. A chronological listing of serious adverse events (SAEs) will be presented, within allocated treatment group. The number of SAEs and the number of patients reporting at least one SAE will be reported in each group. The worst reported severity of all AEs (and similarly for only those related to study treatment) will be tabulated by randomised group.

**Example Table 7: Adverse events (worst reported)**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Adverse event** | **Grade** | **Omeprazole (N=)** | | **Placebo (N=)** | |
| **N** | **%** | **N** | **%** |
| Cough | None |  |  |  |  |
| Mild |  |  |  |  |
| Moderate |  |  |  |  |
| Severe |  |  |  |  |
| Abdominal pain | None |  |  |  |  |
| Mild |  |  |  |  |
| Moderate |  |  |  |  |
| Severe |  |  |  |  |
| Fatigue | None |  |  |  |  |
| Mild |  |  |  |  |
| Moderate |  |  |  |  |
| Severe |  |  |  |  |

**Example Line listing: SAEs, SARs and SUSARs**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ID | SAE no. | Tx. group | Tx. start | Tx. end | Onset date | Description | Severity(1) | Causality(2) | Seriousness(3) | Expected(4) | Type(5) | Outcome(6) | Outcome date |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |

(1) Mild/ moderate/ severe

(2) Life threatening / hospitalisation or prolongation of hospitalisation / persistent or significant disability or incapacity / other significant medical event / congenital anomaly or birth defect

(3) Unrelated/unlikely/possibly/probable/definite

(4) Expected /Unexpected

(5) SAE/SAR/SUSAR

(6) Recovered with sequelae, condition improving, condition still present & unchanged, condition deteriorated, death

# **Statistical Software**

Data will be downloaded directly from MACRO into the statistical software package requested by the Trial Statisticians (e.g. SAS, Stata, R). Statistical analyses will be carried out by the Trial Statisticians. All programs will be stored in the School Statistics folder on the IHS server. A paper master copy of all analysis reports will be stored securely in the statistical section of the trial master file held in a locked fire-proof cupboard with restricted access.

# **StORAGE AND ARCHIVING**

Trial data are entered by individual site staff into a MACRO database held and maintained by the Newcastle Clinical Trials Unit, Newcastle University. Access to the database is limited to authorised personnel with specific access levels. All systems are backed-up on regular basis in accordance with current SOPs. Cough data will be collected by Manchester University and transferred to the Trial Statistician securely via email in an Excel spreadsheet.

The Database Manager will release study data to the Trial Statisticians at time points agreed by the TMG in accordance with current SOPs. Any snapshots of the database taken are kept on the NCTU server, which is backed up daily.

At the end of the study, permissions for the database will be removed for data entry personnel and the status of the MACRO database will be changed to “Closed to Follow-up”, ensuring that no further data can be entered or changed. The data used for the final analysis will be archived according to current SOPs. The Chief Investigator will receive a CD containing data from MACRO in CSV (comma-separated values) format, including a full download of all participants’ data (with audit trail) in PDF and HTML formats. An additional CD/DVD with all the study data will be archived with the TMF.

**References**

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3. Vickers and Altman, Analysing controlled trials with baseline and follow up measurements

BMJ 2001; 323: 1123-1124.

4. Eldridge SM et al. CONSORT 2010 statement: extension to randomised pilot and feasibility trials. *BMJ* 2016; 355: i5239