A randomised, placebo controlled first in human study to investigate the safety, tolerability, pharmacokinetics and pharmacodynamics of Y14 in adult subjects

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PROTOCOL REVISION HISTORY

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2 LIST OF ABBREVIATIONS

adm	Administration
ADR	Adverse drug reaction
AE	Adverse event
ALK	Alkaline phosphatase
ALS	Advanced life support
ALT	Alanine aminotransferase
API	Active pharmaceutical ingredient
aPTT	Activated partial thromboplastin time
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration time curve
BMI	Body mass index (kg/m^2)
CI	Confidence interval
C_{max}	Maximum plasma concentration
CNS	Central Nervous System
CRF	Case report form
CRO	Contract research organisation
CRP	C reactive protein
CRU	Clinical research unit
CV	Coefficient of variation
DEBQ	Dutch Eating Behaviour Questionnaire
DSM	Diagnostic and Statistical Manual
ECG	Electrocardiogram
EDC	Electronic data capture
EMA	European Medicines Agency
EU	European Union
FDA	Food and Drug Administration
FIH	First in human
FSH	Follicle-stimulating hormone
FT4	Free thyroxine

GCP	Good Clinical Practice
GGT	Gamma glutamyl transpeptidase
GLP	Good Laboratory Practice
GLP-1	Glucagon-like peptide-1
GMP	Good Manufacturing Practice
h	Hour
HbA1c	Haemoglobin A1c
HBsAg	Hepatitis B surface antigen
HED	Human equivalent dose
hERG	Human ether-a-go-go related gene
HIV	Human immunodeficiency virus
IB	Investigator's brochure
ICF	Informed consent form
ICH	International Conference on Harmonization
IMP	Investigational medicinal product
IMPD	Investigational medicinal product dossier
ISO	International Organisation for Standarisation
IUD	Intrauterine device
IUS	Intrauterine system
LC-MS/MS	Liquid chromatography-tandem mass spectrometry
LDH	Lactate dehydrogenase
LH	Luteinizing hormone
MABEL	Minimum anticipated biological effect level
MAD	Multiple ascending dose
Max	Maximum
MedDRA	Medical Dictionary for Regulatory Activities
MHRA	Medicines and Healthcare products Regulatory Agency
Min	Minimum
ml	Milliliter
mm	Millimeter
MRSD	Minimum recommended starting dose
MTD	Maximum tolerated dose
ND	Not determined
ng	Nanogram
nM	Nanomole
NOAEL	No observed adverse effect level
OECD	Organisation for Economic Co-operation and Development
OGTT	Oral glucose tolerance test
PD	Pharmacodynamic
PK	Pharmacokinetic
PP	Pancreatic polypeptide
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
PYY	Peptide YY
QMS	Quality management system
QP	Qualified person

R_o	Extent of accumulation in plasma
SAD	Single ascending dose
SAE	Serious adverse event
SC	Subcutaneous
SCOFF	Sick Control One stone Fat Food questionnaire
SD	Standard Deviation
SEM	Standard Error of the Mean
SGLT-2	Sodium/Glucose cotransporter 2
SI	Statutory Instrument
SUSAR	Suspected unexpected serious adverse reaction
$t_{½}$	Apparent terminal half life
TEAE	Treatment Emergent Adverse Event
t_{max}	Time to maximum plasma concentration
TP	Treatment period
TSH	Thyroid stimulating hormone
UK	United Kingdom
US	United States of America
VAS	Visual analogue scale
WHO DD	World Health Organization Drug Dictionary
Y1r	Neuropeptide Y subtype 1 receptor
Y2r	Neuropeptide Y subtype 2 receptor
Y4r	Neuropeptide Y subtype 4 receptor
Y5r	Neuropeptide Y subtype 5 receptor
λ_z	Apparent terminal rate constant

3 INTRODUCTION

A powerful and complex physiological system exists in humans to ensure that adiposity and body weight remain constant despite variations in daily food intake and energy expenditure [1]. Daily energy intake is determined not only by the composition of the food being eaten but also by an individual's appetite, which can be considered as their drive to eat. The level of appetite is controlled predominantly by peptide hormones released in the gut. In particular, pancreatic polypeptide (PP), glucagon-like peptide-1 (GLP-1), oxyntomodulin and peptide YY (PYY) increase satiety and therefore reduce food intake following a meal [2-7]. However, excessive appetite and the easy availability of highly calorific foods with enhanced hedonic properties can lead to an increase in body weight and the potential for the development of obesity.

Obesity is defined as a body mass index (BMI) of more than 30 kg/m^2. Obesity reduces life expectancy and increases the incidence of cardiovascular disease, diabetes, certain cancers and depression [8,9].

In 2014, about 26% of adults in the United Kingdom were obese [10]. The prevalence of obesity is increasing with time and a recent estimate indicated that obesity affects 35% of men and 40% of women in the US adult population [11].

Advice on dietary and lifestyle changes has been ineffective in reducing the incidence of obesity. Currently, two general approaches to treatment are available: first, the use of oral medication (orlistat) to reduce fat absorption from the diet, and, second, the use of surgical interventions. However, both approaches are subject to potential problems regarding efficacy or safety. The use of orlistat is limited by side effects and its effect on body weight is relatively short-lived [12]. Other drugs, e.g. sibutramine and rimonabant, have recently been withdrawn from marketing in Europe due to side effects [13]. Surgical interventions e.g. gastric bypass and band surgery can be effective, but also carry a risk of death to the patient, and are expensive, being dependent on the availability of specialist surgeons and support facilities.

3.1 Background Information for Y14

Y14 is a human peptide YY (PYY) analogue that is being evaluated by Imperial College London as a potential treatment for obesity.

It was selected from about 1500 PYY analogues that were screened for human neuropeptide Y2 receptor (Y2r) binding, mouse Y2r binding and anorectic and weight-reducing effect in rodents. The amino acid sequence of Y14 is based on human PYY_{1-36}, with the following features:

- Deletion of the first residue;
- Substitution of six other residues within the 35-residue peptide with the receptor active sequence of PYY being unchanged
- Composition wholly of natural L-amino acids without derivatisation or cross linking.

For further information please refer to the Investigator's Brochure (IB) [14].

3.2 Risk Assessment

Y14 has not yet been tested in humans, however the amino acid sequence of Y14 is similar to that of PYY. In addition, previous experience was gained with a similar PYY analogue (Y242 – Imperial College London QLON/2011/Y242-01). Therefore indications of potential safety issues related to the administration of Y14 can be obtained from review of the effects of PYY and Y242 in humans.

In a number of published studies when PYY_{3-36} was administered by IV infusion to healthy subjects in doses to achieve normal postprandial values (40-50 pmol/L) no adverse effects were reported [6,7].

When PYY_{3-36} was infused at a dose to raise circulating PYY to supraphysiological levels (90 to 190 pmol/L) nausea, vomiting and abdominal discomfort were reported [16 - 19]. Therefore the published data indicate that the therapeutic window for PYY_{3-36} is narrow. It is possible that nausea and vomiting may also be observed in those subjects given Y14. Nausea is a common side effect of gut hormone analogues in clinical use, such as the GLP-1 analogues exenatide and liraglutide, although this side effect tends to decline with continued use and rarely causes discontinuation of the drug [20]. As Y14 has been formulated [redacted] to produce a sustained release formulation, the potential for nausea and vomiting to occur is reduced. It was also observed that in single doses ranging from 2 to 90 mg, Y242 was well tolerated. Subjects receiving multiple doses of Y242 (90 mg) experienced nausea and vomiting as a dose related TEAE.

As nausea and vomiting are expected to be the most common AEs, anti-emetics will be available at the clinical unit. As a precaution, individuals frequently suffering from migraines will be excluded from investigational studies with Y14, so that the risk of nausea caused by migraines does not interfere with the analysis of adverse events during the study. Volunteers with other potential causes of nausea (e.g., gastrointestinal disease) will be ineligible for the study at screening. Nausea will be detected both by adverse event recording and by the use of specially designed questionnaires. Mild to moderate nausea will not be a cause for withdrawal from the study. If eligible subjects suffer from severe and persistent nausea, or severe vomiting during this study, which the Investigator believes may be due to the investigational product, they will be withdrawn from the study.

It was also observed in the Y242 study that skin reactions were a common TEAE. Dose levels or concentrations may require adjustment accordingly, and the initial doses given during the SAD phase of the study will be limited to 20 mg/ml. If necessary, the doses may be divided into multiple injections to reduce injection volumes and the amount of Y14 administered per injection site. In addition local tolerance and in particular injection site reactions will be monitored closely throughout the study.

PYY is known to cause a 30% increase in urinary sodium output when given to healthy human volunteers, but no significant change in urinary volumes [21]. In the Y242 trial, a natriuretic

effect was not observed. In the target clinical population of obese people, hypertension is a common co-morbidity, and it is anticipated that, if Y14 does indeed cause a natriuresis, this may ameliorate hypertension [21]. GLP-1 has a similar natriuretic effect in humans [22] and this effect is thought to underlie its anti-hypertensive action [23].

No other safety concerns are anticipated, but monitoring of an extensive list of safety variables will take place during this study.

The proposed study is a single and double-blind, randomised, placebo controlled Phase I FIH study to be conducted in 2 parts (Parts A and B);

As this is a FIH study a sentinel dosing strategy will be used for the first dosing group. Safety data up to and including 24 h postdose will be reviewed by the Investigator prior to dosing the remaining subjects at that dose level 48 h later. Dose escalation will only occur if the previous dose level was deemed to be safe and well tolerated.

Part B will only commence after appropriate and sufficient data from Part A are available to enable the determination of a safe starting dose for Part B. Administration of subsequent dose levels will be based on review of available safety, tolerability and PK data from previous doses. As with Part A, progression to the next higher dose will only occur if the previous dose level was deemed to be safe and well tolerated. In Part B, dose titration for an individual subject may be necessary within a dosing regimen depending on the activity profile and tolerability.

The study will be conducted in compliance with United Kingdom (UK) regulations and guidance, EU and ICH GCP, GMP and current GLP. The study requires clinical trial authorisation by Medicines and Healthcare Products Regulatory Agency (MHRA) and National Research Ethics Service approval. It does not require Ethics Advisory Group approval.

As this is a FIH study involving the administration of a peptide analogue the study will be conducted on an in-patient basis, with dosing in a Phase I unit with accreditation from the MHRA. The Principal Investigator and the investigating team will be experienced in the administration of novel molecules to man for the first time.

There is no known antidote for Y14. Full resuscitation facilities and emergency medication will be available at all times.

3.3 Urgent safety measures

In accordance with UK Law (Medicines for Human Use [Clinical Trials] as amended: Statutory Instrument [SI] 1031 Part 4 Section 30) the Sponsor and Investigator may take appropriate urgent safety measures in order to protect the subjects of a clinical trial against any immediate hazard to their health or safety. If such measures are taken the Sponsor shall immediately (no later than 3 days from the date the measures are taken) give written notice to the licensing authority and the relevant ethics committee of the measures taken and the circumstances giving rise to those measures.

4 STUDY OBJECTIVES

4.1 Primary Objective

- To investigate the safety and tolerability of single doses of Y14 in overweight/obese but otherwise healthy male subjects.
- To investigate the safety and tolerability of multiple doses of Y14 in overweight/obese male subjects with normal glucose tolerance, Type 2 diabetes or prediabetes.

4.2 Secondary Objectives

- To assess the pharmacokinetic (PK) profile of single doses of Y14 in overweight/obese but otherwise healthy male subjects.
- To assess the PK profile of multiple ascending doses of Y14 in overweight/obese male subjects with normal glucose tolerance, Type 2 diabetes or prediabetes.

4.3 Exploratory Objective

- To investigate the effects of multiple doses of Y14 on food consumption, body weight, enteropancreatic hormone changes and glucose tolerance in overweight/obese male subjects with normal glucose tolerance, Type 2 diabetes or prediabetes.
- To assess the analytical performance of the Imperial College radioimmunoassay for Y14 compared with the LC-MS/MS assay for Y14.

5 INVESTIGATIONAL PLAN

5.1 Overall Study Design

This is a single centre, randomised, placebo controlled Phase I FIH 2 part study: Part A (single ascending dose – SC administration) and Part B (multiple ascending dose – SC administration).

5.1.1 Part A (Single Ascending Dose)

Part A is a double blinded, randomised, placebo controlled, single ascending dose study (SAD). The following reflects the plan prior to execution of Part A. Approximately 28 eligible subjects will be enrolled in 5 sequential cohorts of overweight but otherwise healthy male subjects (BMI range of 25 to 38 kg/m^2). No subject will be a member of more than 1 cohort.

Cohort 1 will comprise 4 subjects (3 active and 1 placebo). In cohort 1, each subject will be dosed in three treatment periods (TPs) with three ascending dose levels (proposed doses 1 mg, 4 mg, 8 mg Y14). The subjects will be randomised to either placebo or active treatment at each of these visits, i.e. different subjects will receive placebo in each TP. There will be a minimum washout period of 1 week between Day 1 of each TP. If any subject in Cohort 1 drops out between doses, they will be replaced.

Cohort 2 onwards will comprise 6 subjects (5 active and 1 placebo) for each cohort. Each volunteer in Cohort 2 onwards will only be dosed once in one TP. Proposed doses for cohorts 2-5 are 16 mg, 32 mg, 64 mg and 96 mg. All doses, and peptide concentrations are subject to change according to safety and PK review.

Up to eight additional cohorts of up to 6 subjects each (i.e. up to 48 subjects in total) may be enrolled if recommended by the Safety Committee (see Section 7.1.4) to increase scientific value of this study, e.g. to verify PK data or further characterise PD markers.

For each dose level, subjects will be randomised to receive a single dose of Y14 or matching placebo by SC injection (with fractionation if needed). A sentinel dosing strategy will be used as outlined in Table 5.1.

Table 5.1 Dosing Schedule: Part A

Cohort	Treatment Periods	Dosing Session	Number of Subjects	
			Y14	Placebo
1	1	Sentinel subjects	1	1
		Non-sentinel subjects	2	0
1	2-3	Sentinel subjects	0	0
		Non-sentinel subjects	3	1
2 onwards	1	Sentinel subjects	0	0
		Non-sentinel subjects*	20	4

*May be increased if additional groups are dosed

In Cohort 1, TP1, the first two subjects will be randomised such that 1 subject receives Y14 and 1 subject receives placebo. This sentinel pair will be dosed first and will be observed for at least 48 h before study drug is administered to the remainder of the cohort. Safety (vital

signs, ECGs, and clinical laboratory safety tests), and tolerability (adverse event profile) data up to and including 24 h postdose will be reviewed by Covance's clinical team prior to dosing the remaining subjects at that dose level. Dosing of the subsequent subjects in cohort 1 will be at the discretion of the Principal Investigator.

Treatments will be administered double blinded (except for Cohort 1, TP1 where it will be known that the 3rd and 4th subjects will receive the active dose); subjects will be blinded with regard to treatment and clinical staff will remain blinded with regard to treatment until the Safety Committee meeting.

An interval of at least 7 days will separate the dosing of the last subject in one cohort and dosing of the first subject in the next cohort to permit a timely review and evaluation of interim safety, tolerability and PK data. Safety, tolerability and PK data will be reviewed up to 48 hours post-dose prior to the dose escalation decision. If T_{max} is significantly later than predicted, PK data to a later timepoint may be reviewed prior to the dose escalation decision. The data of up to at least 3 subjects receiving Y14 in cohort 1 and 4 subjects, (i.e. a minimum of 3 subjects receiving Y14) in cohort 2 onwards will be reviewed before dose escalation.

Following a 28-day screening period eligible subjects will be admitted to the clinical unit on Day -1 (1 day prior to dosing) for eligibility checks and baseline assessments. On Day 1 subjects will receive a single dose of Y14 or placebo by SC injection, and will remain in the clinical unit under medical supervision until 72 h postdose (Day 4) or until C_{max} is achieved, whichever is the later.

Safety monitoring will include assessment of adverse events (AEs), physical examination, 12-lead ECGs, vital signs (blood pressure pulse rate and body temperature) and clinical laboratory safety tests (serum biochemistry, haematology, coagulation, thyroid stimulating hormone [TSH], FT4 and urinalysis).

Blood samples for biomarker (tryptase) will be collected immediately after any possible allergic or anaphylactic adverse reaction, and 12 hours later. These samples will only be analysed if relevant adverse reactions occur.

The PK evaluation will include measurement of plasma Y14 levels and associated PK parameters.

Exploratory PD evaluation will include evaluation of visual analogue scales (VAS) of nausea and satiety. Subjects in Cohort 3 onwards will undergo food intake studies on Day -1 and Day 1. The timing of the food intake study may be modified according to emerging PK data.

Blood samples for immunogenicity assays will be collected prior to the first dose (Treatment Period 1) and at the final follow-up visit for the volunteers from Cohort 1 only (as these volunteers have received multiple doses of Y14).

All subjects will return for a follow up visit 14 days (± 1 day) after the final dose.

Following discharge of subjects and depending on emerging PK data, the subjects (except cohort 1, dose level 1 and 2) may return to the clinical unit for further outpatient visits. The

number of additional visits per subject will not exceed six and will not extend beyond 8 weeks after each final dosing occasion. If additional outpatient visits are scheduled the final outpatient visit will be considered to be the final follow up visit where the follow-up visit assessments will be performed.

5.1.2 Part B (Multiple Ascending Dose)

Part B will be a double blind, randomised, placebo controlled, multiple ascending dose (MAD) study in sequential groups of male subjects (BMI range 25-38 kg/m^2). These subjects can either have normal glucose tolerance, prediabetes (impaired glucose tolerance or impaired fasting glucose) or Type 2 diabetes, defined according to WHO 2006 and 2011 diagnostic criteria. If the subject is diagnosed with diabetes or prediabetes, this condition should be stably controlled either with:

i. diet only;
ii. monotherapy with a sulphonylurea, metformin, or SGLT-2 inhibitor;
iii. dual therapy with a sulphonylurea, metformin, and/or a SGLT-2 inhibitor;
iv. or triple therapy with a sulphonylurea, metformin, and SGLT-2 inhibitor.

Patients treated with other anti-diabetic treatments are excluded.

Where subjects have prediabetes or diabetes, their HbA1c at screening should be 6.0–8.5% (42–69 mmol/mol) and <±1.0% (±11 mmol/mol) from a previous HbA1c reading within the last 6 months, where available. Where a HbA1c reading within the last 6 months is not available, the subject should have HbA1c re-measured after at least 4 weeks to assure stability of glycaemia before inclusion in the study. This remeasurement may take place any time up to and including check in on Day -2.

For other subjects without known diabetics or pre-diabetes, or in whom the glycaemic status is in doubt, they may attend a pre-screening visit no more than 10 weeks before Day 1, where a fasting glucose and HbA1c will be measured, and a 75 g oral glucose tolerance test performed to classify their diagnosis according to the WHO 2006 and 2011 criteria.

It is proposed to investigate 3 dose levels of Y14 in up to 3 cohorts of 8 subjects, anticipated total of 24 subjects. In each cohort 6 subjects will receive Y14 and 2 subjects will receive placebo. Where possible, subjects of a similar glycaemic status (i.e. diabetics, prediabetics or normal glucose tolerance) will be grouped in a cohort.

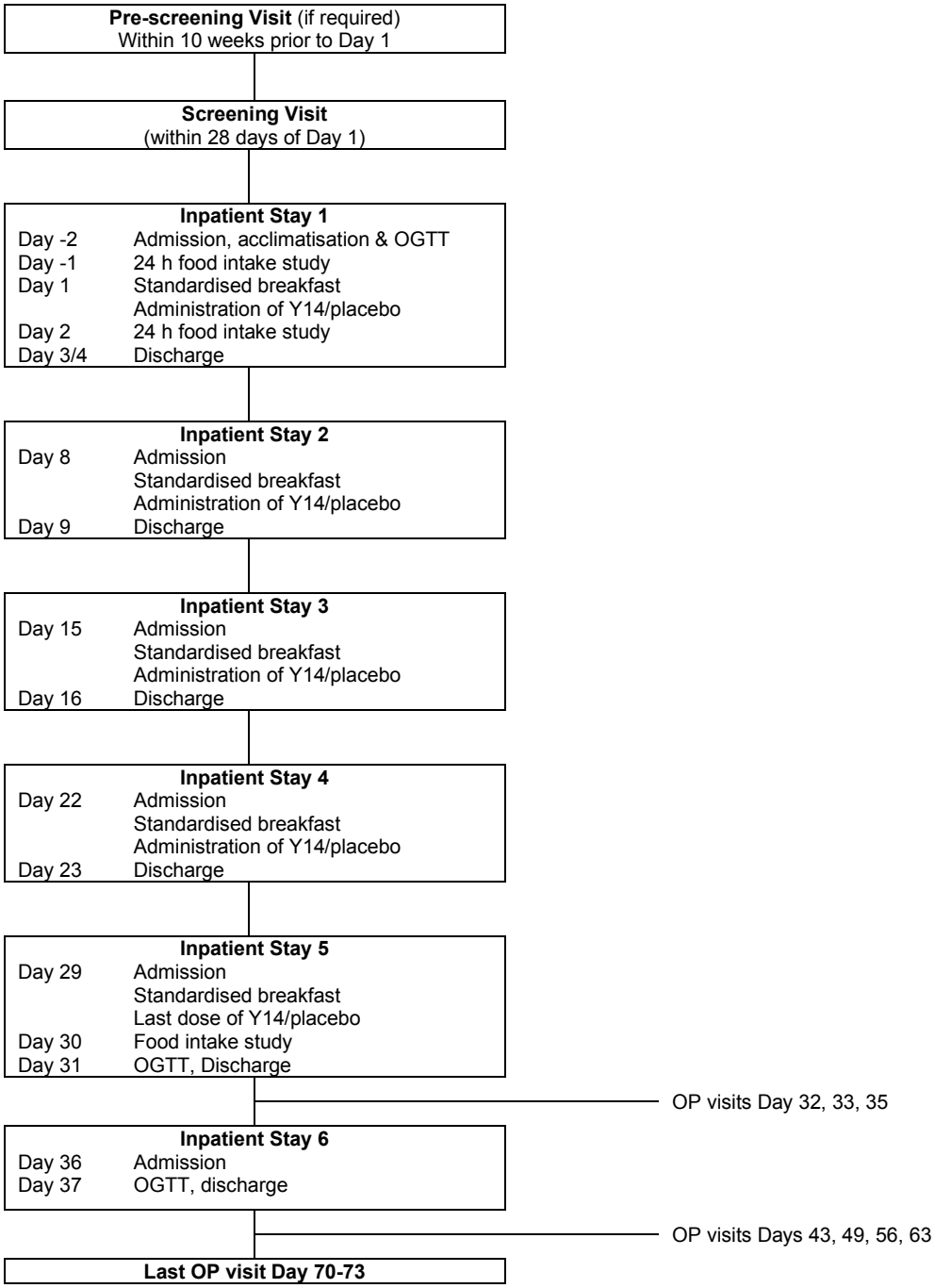
Up to 3 additional cohorts of 8 patients each, i.e. up to 24 subjects in addition, may be enrolled if recommended by the Safety Committee (see Section 7.1.4) to increase scientific value of this study, e.g. to assess additional doses of Y14 or further characterise PD markers.

Part B may commence prior to the completion of Part A (SAD) and the total daily dose will not exceed a single dose shown to be safe and well tolerated.

Subjects will receive 5 doses of Y14 or placebo by SC injection (with fractionation if needed), each separated by 7 days (i.e., on Days 1, 8, 15, 22, 29). The starting dose level to be administered in Part B will be determined from Part A. Administration of subsequent doses will be based on review of available data on safety, tolerability and PK up to 48 hours after the last dose, gathered from previous doses in a minimum of 6 subjects. The individual doses to be given in Part B will not exceed the maximal tolerable dose identified from Part A. Dose titration for an individual subject may be necessary within a dosing regimen depending on the activity profile and tolerability.

Subjects are proposed to have 6 inpatient stays in Part B as outlined in Figure 5.1.

Figure 5.1 Study Overview: Part B



Following a pre-screening visit within 10 weeks of day 1 (if applicable), and a 28-day screening period, eligible subjects will be admitted to the clinical unit on the morning of Day -2 (2 days prior to first dosing) for the first inpatient stay. At Day -2, the volunteers will undergo an OGTT, and therefore will need to attend the visit after an overnight fast.

Subjects will receive a single dose of Y14 or placebo by SC injection(s) on Day 1. Subjects will remain in the clinical unit for at least 24 h postdose. It is the intention that subjects will be discharged on Day 4, but they may be discharged on Day 3 after the food intake study has been completed and if the subject is in good health. If volunteers are discharged on Day 3, they will return on Day 4 for collection of a pharmacokinetic sample.

For most of the subsequent inpatient stays (stays 2 to 4) subjects will be admitted to the clinical unit on the day of dosing and will remain in the clinical unit at least until 24 h postdose. In stay 5 the subjects will be admitted to the clinical unit on the day of dosing and will remain in the clinical unit until 48 h postdose. It is proposed that doses will be administered 7 days apart (i.e. on Days 1, 8, 15, 22 and 29).

The proposed duration of the inpatient stays may be altered based on emerging safety, tolerability and PK data from Part A (SAD) or previous cohorts in Part B.

After each inpatient stay, subjects will return to the clinical unit for outpatient visits as described in Figure 5.1,

Safety monitoring will include assessment of adverse events (AEs), physical examination, 12-lead ECGs, vital signs (blood pressure, pulse rate and body temperature) and clinical laboratory safety tests (serum biochemistry, haematology, coagulation, TSH, FT4, and urinalysis).

Blood samples for biomarker (tryptase) will be collected immediately and 12 hours after any possible allergic or anaphylactic adverse reaction. These samples will only be analysed if there is a relevant adverse reaction.

The PK evaluation will include measurement of plasma Y14 levels and associated PK parameters.

Exploratory PD evaluation will include evaluation of the following: glucose tolerance (as assessed by a standard 75 g oral glucose tolerance test before and after treatment), energy intake at mealtimes, body weight, visual analogue scales (VAS) of nausea and satiety and enteropancreatic hormones.

Blood samples for immunogenicity assays will be collected predose and at the final follow-up visit.

Subjects will attend a final follow-up visit on Day 70-73. If deemed necessary, unblinding will be performed as outlined in Section 6.9.

5.1.3 Safety Committee

The Safety Committee's membership will be composed of a minimum of the Sponsor team (Sponsor Senior Physician and Sponsor Senior Scientist or an appropriate delegate) and Principal Investigator (or an appropriate delegate). Additional members may be invited as needed (e.g. PK scientist, project manager).

All data reviewed at the dose escalation meeting (safety, tolerability and available PK data) will be subjected to a quality control review.

5.1.4 Stopping Rules

Dosing for any individual subject will be stopped if the subject experiences a possibly drug-related serious adverse event or a possibly drug-related significant non serious adverse event, which in the opinion of the study physician, Principal Investigator or Chief Investigator (Sponsor's medical representative), warrants discontinuation of the study for that subject's well being.

In Parts A and B, following consultation with the Sponsor, dose escalation will stop if:

- Clinically relevant signs or symptoms of similar nature, occur in 2 or more subjects within a group/cohort, which in the opinion of the Investigator warrant stopping of dose escalation.
- A serious adverse event (SAE) in one or more subjects thought to be related to the study drug or a severe AE in two or more subjects thought to be related to study drug. If this criterion is met the study will be halted. If, following internal review, it is deemed acceptable to restart the study a substantial amendment will be submitted to the Competent Authority.
- Moderate nausea or vomiting that prevent subjects from eating a meal on 3 or more occasions on 2 successive days are seen in 2 or more subjects in a group.
- Severe diarrhoea, defined as 7 or more episodes, rated as ≥ 5 on the Bristol Stool Chart, in 1 day for 2 consecutive days are seen in 2 or more subjects in a group.
- The mean systemic exposure is predicted to exceed a C_{max} of 440 ng/mL and/or AUC(0-72h) of 28,100 ng·h/mL i.e. systemic exposure will be no greater than that the lowest exposure at the NOAEL for male rats (30 mg/kg/dose). This limit is chosen on the basis that rats are expected to be the species that will most closely predict toxicity in humans.

5.1.5 Discussion of Study Design

This is a double-blinded, randomised, placebo-controlled study in two parts: a single ascending dose study and multiple ascending doses, to investigate the effects of Y14. Note, with the exception of Cohort 1 TP1 which is partially blinded as this will be a crossover design where the volunteers will return for 3 treatment periods. The time interval between each TP should be no less than 1 week. As Y14 is intended as an anti-obesity drug, overweight/obese subjects have been chosen to provide more clinically relevant PK and PD information. With regards to Part B, we propose to recruit patients with Type 2 diabetes or pre-diabetes in order to study the impact of Y14 on glycaemia.

It is the intent of Part B to dose volunteers such that steady-state is achieved and maintained for several days. Based on the available pre-clinical data it is expected that this will be achieved following 28 days once weekly dosing, however, a full review of all the safety, tolerability and pharmacokinetic data from Part A will be performed to confirm the dose regimen for Part B. If

the elimination half-life of Y14 is shorter than anticipated from the pre-clinical data, more frequent dosing (e.g. twice weekly) may be appropriate. If the elimination half-life of Y14 is longer than anticipated from the pre-clinical data, dosing every two weeks may be more appropriate (as steady-state will take longer to achieve). The dose frequency will be no more frequent than once a day and no less frequent than once every 14 days. The dosing duration will not exceed 29 consecutive days of dosing.

Details of the dosing regimen and duration used for Part B of the study will be documented in the SMF.

The sample size (approximately 28 subjects in Part A [SAD] and 24 subjects in Part B [MAD]) has been chosen to minimise the number of subjects exposed to Y14 whilst obtaining sufficient information to assess the safety, tolerability, PK and PD of Y14.

5.2 Study Population

Approximately 28 subjects will be enrolled in Part A (SAD) and 24 subjects will be enrolled in Part B (MAD).

Overweight male volunteers will be entered into this study provided that they satisfy the following inclusion/exclusion criteria.

5.2.1 Subject Inclusion Criteria

1. Adult males aged 18 to 65 years inclusive with BMI between 25.0 and 38.0 kg/m^2 inclusive;
2. (PART B only) Subjects who have normal glucose tolerance, Type 2 diabetes, impaired glucose tolerance or impaired fasting glucose according to WHO 2006 and 2011 criteria,
   a) In subjects who are identified as being prediabetic or diabetic, they should be stably treated either with:
      i. diet only;
      ii. monotherapy with a sulphonylurea, metformin, or a SGLT-2 inhibitor;
      iii. dual therapy with a sulphonylurea, metformin, and/or a SGLT-2 inhibitor;
      iv. or triple therapy with a sulphonylurea, metformin, and a SGLT-2 inhibitor.

      Patients treated with other anti-diabetic treatments are excluded.
   b) In subjects who are identified as being prediabetic or diabetic, the HbA1c at screening should be 6 - 8.5% (42 - 69 mmol/mol) and <±1.0% (±11 mmol/mol) from a previous HbA1c reading within the last 6 months, where available. Where an HbA1c reading within the last 6 months is not available, the subject should have HbA1c re-measured after at least 4 weeks to assure stability of glycaemia before inclusion in the study. This remeasurement may take place any time up to and including check in on Day -2.
   c) To allow assessment of eligibility, subjects without known diabetes or prediabetes, or in whom the glycaemic status is in doubt, may undergo a pre-screening visit no more

than 10 weeks before Day 1 for assessment of fasting glucose, HbA1c and glucose 2 hours after a 75 g oral glucose tolerance test.

3. Subjects who are otherwise healthy enough to participate, as determined by pre-study medical history, physical examination and 12-lead ECG;
4. Subjects whose clinical laboratory test results are either within the normal range or if outside this range the abnormalities are judged to be not clinically relevant and are acceptable to the Investigator;
5. Subjects who are negative for hepatitis B surface antigen (HBsAg), hepatitis C antibody and human immunodeficiency virus (HIV) I and II tests at screening;
6. Subjects who are negative for drugs of abuse and alcohol tests at screening and admissions;
7. Subjects who are non-smokers for at least 3 months preceding screening;
8. Subjects who agree to use medically acceptable methods of contraception for at least 3 months after study drug administration;
9. Subjects who agree not to donate sperm for at least 3 months after study drug administration;
10. Subjects who are able and willing to give written informed consent.

Subjects' medical history must be verified by either a personal physician or medical practitioner as appropriate.

5.2.2 Subject Exclusion Criteria

1. Subjects who do not conform to the above inclusion criteria;
2. Subjects who have a clinically relevant history or presence of gastrointestinal (especially associated with vomiting), respiratory, renal, hepatic, haematological, lymphatic, neurological (especially if associated with balance disorders or vomiting e.g. migraine or labyrinthitis), cardiovascular, psychiatric, musculoskeletal, genitourinary, immunological, dermatological, connective tissue diseases or disorders;
3. Subjects who have a clinically relevant surgical history;
4. Subjects who are currently taking any of the following classes of diabetes medications: thiazolidinediones, dipeptidyl peptidase IV inhibitors ('gliptins'), GLP-1 analogues, and insulin;
5. Subjects who have a history of relevant and severe atopy e.g. asthma, angioedema requiring emergency treatment, severe hayfever requiring regular treatment (i.e. taking antihistamines and/or glucorticoids more regularly than 3 times a week), severe eczema requiring regular treatment (i.e. taking antihistamines and/or glucocorticoids more regularly than 3 times a week);
6. Subjects who have a history of relevant drug hypersensitivity;

7. Subjects who have a history of alcohol abuse or alcohol dependence according to DSM-IV criteria within the last 2 years;
8. Subjects who have a history of drug or substance abuse according to DSM-IV criteria within the last 2 years;
9. Subjects who have a history of clinically significant migraine as judged by the Investigator. Subjects can be included if they have not had a migraine for the last 3 years;
10. Subjects with a history of pancreatitis or pancreatic cancer;
11. Subjects who consume more than 21 units of alcohol a week (unit = 1 glass of wine (125 mL) = 1 measure of spirits = ½ pint of beer);
12. Subjects who have a significant infection or known inflammatory process on screening;
13. Subjects who have acute gastrointestinal symptoms at the time of screening or admission (e.g. nausea, vomiting, diarrhoea, heartburn);
14. Subjects who have an acute infection such as influenza at the time of screening or admission;
15. Subjects who have used prescription drugs within 2 weeks of first dosing. For Part B, patients are allowed to be treated for their diabetes with monotherapy with a sulphonylurea, metformin, or a SGLT-2 inhibitor, dual therapy with any two of the following drug types: a sulphonylurea, metformin, and/or a SGLT-2 inhibitor; triple therapy with a sulphonylurea, metformin, and a SGLT-2 inhibitor. In addition, patients in Part B are allowed to take hypolipidaemic and/or antihypertensive treatments, provided that the doses have not been altered within the 4 weeks prior to entering the study. Other medications may be allowed if the Investigator and Sponsor both agree that they will not affect the outcome of the study or the safety of the subject.
16. Subjects who have used over the counter medication excluding routine vitamins and paracetamol but including megadose (intake of 20 to 600 times the recommended daily dose) vitamin therapy within 7 days of first dosing, unless agreed as not clinically relevant by the Principal Investigator and Sponsor;
17. Subjects who have donated blood within 3 months prior to screening;

    Subjects who have donated plasma within the 7 days prior to screening;

    Subjects who have donated platelets within the 6 weeks prior to screening
18. Subjects who have used any investigational drug in any clinical trial within 3 months of their first admission date;
19. Subjects who have received the last dose of investigational drug greater than 3 months ago but who are on extended follow-up;
20. Subjects who have previously received Y14;

21. Subjects who are vegans, vegetarian or have any dietary restriction (unless agreed as not clinically relevant by the PI and Sponsors);
22. Subjects who cannot communicate reliably with the Investigator;
23. Subjects who are unlikely to co-operate with the requirements of the study;
24. History or evidence of abnormal eating behaviour, as observed through the Dutch Eating Behaviour (DEBQ) and SCOFF questionnaires [24,25] at screening.

5.2.3 Restrictions

In Part A (SAD) subjects will be admitted to the clinical unit on Day -1 and will remain in the clinical unit under supervision until discharge on Day 4 (at least 72 h postdose) or when C_{max} is achieved, whichever is later. Subjects will be required to attend outpatient visits as described

In Part B (MAD) subjects will be admitted to the clinical unit for 6 inpatient stays: Day -2 (admission) to Day 4 (discharge), Day 8 (admission) to Day 9 (discharge), Day 15 (admission) to Day 16 (discharge), Day 22 (admission) to Day 23 (discharge), Day 29 (admission) to Day 31 (discharge), and Day 36 (admission) to 37 (discharge). Subjects will return to the clinical unit for outpatient visits as described in Table 17.2.

For both Part A and Part B, the inpatient stays and outpatient visits may be amended upon review of safety, tolerability and PK data from previous cohorts or study part.

Subjects will receive a standard diet whilst resident in the clinical unit and will only consume foodstuffs provided by the clinical unit. Subject's energy intake at each meal, along with fluid intake and output will be recorded during the inpatient stays. For Part B, study participants will be provided with a dietary advice sheet providing information with regards to a healthy eating diet which they are recommended to follow when they have been discharged from the clinical unit.

For Part A and Part B subjects should refrain from the following:

- Subjects should refrain from strenuous exercise for 48 h before screening, prior to each admission, throughout each inpatient stay and for 48 h prior to each outpatient visit.
- Subjects should refrain from alcohol and/or xanthine containing products (e.g. caffeine) for 48 h prior to screening and admission, during the inpatient stay, and for 48 h prior to each outpatient visit.
- Subjects should refrain from smoking for the duration of the study (until after the final follow-up visit).
- Subjects should refrain from eating foodstuffs that contain poppy seeds for 7 days prior to screening and 7 days prior to each admission, as these may result in a positive urine test for opiate consumption.
- Subjects should refrain from taking medications during the study as described in Section 6.5.

5.2.3.1 Avoidance of Pregnancy

Instructions for Male Subjects

There is no information about effects that Y14 could have on the development of the foetus in humans. Therefore, it is important that the partners of male subjects do not become pregnant during the study and for a total period of 3 months after the male subject has taken the last dose of Y14.

As a precaution, all male subjects should avoid fathering a child by either true abstinence or the use of 2 effective means of contraception (see Section 5.2.3.2).

As there is no information about Y14 being secreted in the ejaculate, male subjects (including men who have had vasectomies) whose partners are currently pregnant should use barrier methods for the duration of the study and for a suitable time afterwards (e.g. 1 month). This is to ensure that the foetus is not exposed to the investigational medicinal product (IMP) in the ejaculate.

5.2.3.2 Acceptable Forms of Contraception

Highly effective methods of birth control are defined as those which result in a low failure rate (i.e. less than 1% per year) when used consistently and correctly.

These include male vasectomy with confirmation of surgical success, female sterilisation (i.e. documented bilateral tubal ligation), hormonal methods of contraception (oral, implanted or transdermal) or an intrauterine device (IUD).

Where there is a possibility of pregnancy one of the above highly effective methods should be used in combination with male condoms.

True abstinence is acceptable when this is in line with the preferred and usual lifestyle of the subject. Periodic abstinence (e.g., calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.

5.2.3.3 Time Period for the Collection of Pregnancy Information

All pregnancies in female partners of male subjects receiving at least one dose of IMP will be recorded from first dose to 3 months after the final dose.

5.2.3.4 Follow-up in the Event of a Pregnancy

If the female partner of a male subject, who has received Y14 becomes pregnant the pregnancy will be recorded. The ethics committee and the Sponsor will be informed. The subject will be asked provide information on the outcome of the pregnancy, including premature termination should the case arise.

Spontaneous miscarriage and congenital abnormalities will be reported as SAEs.

The follow-up period will be deemed to have ended when the health status of the child has been determined on its birth.

5.2.4 Subject Withdrawals

A subject may withdraw for any reason. The Investigator will advise the Sponsor of the withdrawal of any subject.

A subject may be withdrawn in any of the following circumstances:

- Adverse events;
- Protocol violation;
- Withdrawal of consent;
- Termination of the study by the Investigator or Sponsor.

Subjects who voluntarily withdraw are termed dropouts. Dropouts may be replaced following discussion with the Investigator and Sponsor. Subjects withdrawn due to an adverse event which is thought to be related to the study drug will not be replaced. This statement will not apply to Cohort 1 in the event that a subject is withdrawn prior to dosing in Treatment Periods 2 or 3. If a subject is withdrawn from Cohort 1, that subject may be replaced in order to allow a full cohort of 4 subjects to be dosed in the subsequent treatment period.

If a subject is withdrawn or withdraws during the inpatient stay the discharge procedures should be performed and follow-up assessments completed prior to them leaving. If a subject is withdrawn or withdraws after the inpatient stay is completed a follow-up visit should be scheduled.

If a subject is withdrawn due to an adverse event, appropriate medical care should be provided and the adverse event should be followed to resolution. Follow-up procedures should be conducted as scheduled.

6 STUDY TREATMENT

6.1 Investigational Product(s)

Y14 is a 35 amino acid linear peptide. Y14 does not contain any products of human or animal origin.

The bulk drug product (20 mg/vial) is manufactured as a 2 mL ISO clear Type 1 tubular glass vial with a 13mm crimp neck. The vials are sealed with a fluoro-coated lyophilisation stopper and an aluminium crimp seal. Y14 will be reconstituted in sterile diluent supplied in a glass vial with a fluoro-coated rubber stopper and an aluminium crimp seal. The details of the diluent and reconstitution steps will be provided in the IMP Preparation Information.

The placebo is sterile 0.9% (w/v) saline.

6.1.1 Supply, Packaging and Labelling

Y14 bulk Drug Product and Diluent will be supplied to Covance Clinical Research Unit by AMRI (Glasgow).

A release document signed by a legally authorised Qualified Person (QP) at Covance Clinical Research Unit will be placed in the appropriate section of the Trial Master File to document labelling and dispensing of the study drug to the subject. A technical agreement between Covance Clinical Research Unit and Sponsor will be in place to cover all pharmacy related activities, detailing roles and responsibilities prior to receipt of the IMPs at Covance Clinical Research Unit.

These supplies will be packaged in accordance with Annex 13 of "The Rules Governing Medicinal Products in European Community, Volume IV Good Manufacturing Practice for Medicinal Products".

All documents required to perform GMP activities at Covance Clinical Research Unit will be supplied as per the Technical Agreement. The Technical Agreement will outline the roles and responsibilities between the contract giver and the contract acceptor.

The placebo (sterile 0.9% [w/v] saline) will be commercially sourced by Covance Clinical Research Unit.

6.1.2 Storage and Handling Procedures

The Y14 lyophilised powder will be stored at -15 to -25°C. The diluent will be stored at +2 to +8°C. All study medication will be stored in the CRO Pharmacy.

Details of how the IMP will be prepared for administration will be included in a separate IMP Preparation Information document.

The Sponsor will be permitted upon request to audit the supplies, storage, dispensing procedures and records provided that the blind of the study is not compromised.

6.1.3 Accountability

In accordance with GCP, the clinical unit will account for all supplies of Y14 and placebo. Details of receipt, storage, assembly and return will be recorded.

All unused supplies of Y14 and placebo will either be destroyed by CRO or returned to the study Sponsor at the end of the study in accordance with instructions by the Sponsor.

6.2 Dosage and Administration

Y14 or placebo will be administered by SC injection into the abdomen. The maximum individual injection volume will not exceed 1.0 mL, though multiple injections (up to 5) may be given for a single dose. The injection site(s) should be circled with indelible marker (or the position recorded in some other suitable manner) and inspected at each AE check during Day 1. Reactions should be described according to the grading table (Table 7.4). Where multiple doses of Y14 are given to the volunteer, the injection sites must rotate, i.e. given at different sites in the abdomen.

In the event of injection site reactions thought to be related to the volume of the injection, the dose of Y14 or placebo may be fractionated and up to 5 injections may be given.

Subjects will receive Y14 or placebo, whilst in the supine/semi-supine position, said dose to be given 1 hour after the subject has completed eating a standardised breakfast. Subjects will remain confined to their beds for a period of 4 h after injection, but will be permitted to visit the bathroom to use the toilet if needed.

In Part A (SAD) the proposed doses are: 1 mg, 4 mg, 8 mg, 16 mg, 32 mg, 64 mg and 96 mg. Depending on emerging safety, tolerability and PK data, the doses given may be modified. Dose levels administered in Part B (MAD) will be selected following review of the data from Part A. In Part B, dose titration for an individual subject may be necessary within a dosing regimen depending on the activity profile and tolerability, but will not exceed the maximum dose tested in the SAD trial.

Subjects will not eat for 4 h postdose after which time lunch will be served. Dinner will be served 10 h postdose.

6.3 Treatment Strategy

The clinical staff at Covance Clinical Research Unit, Leeds, are responsible for the ongoing safety and well-being of the volunteers while they are in the clinical unit. There is a paging system to alert the clinical staff to any area in the unit where a subject may need medical attention. In the case of an emergency, cardiac resuscitation trolleys are found in the main ward areas of the clinical unit. These trolleys contain drugs, equipment for airway insertion, circulation lines, defibrillation etc, together with oxygen cylinders and portable suction machines. There is a physician on-call 24 hours a day (this can be off site) and all physicians are Advanced Life Support (ALS) trained. In addition, the clinical staff can contact further on-call physicians or public emergency services, including the ambulance service, in the event of

a serious medical event. Equipment and emergency drugs are available to treat common medical emergencies that might occur in a Phase I study.

6.4 Warnings and Precautions

As this is the first administration of Y14 to man, all effects cannot be reliably predicted. The preclinical data suggest an acceptable safety margin. Facilities and staff for resuscitation and the treatment of other medical emergencies will be provided.

6.5 Prior and Concomitant Medication

The participants are not allowed to take any prescription medication within 2 weeks of first dosing apart from those required for treatment of their diabetes, hypercholesterolaemia and hypertension. Subjects are not allowed to take any non-prescription medication (excluding paracetamol) or megadose (intake of 20 to 600 times the recommended daily dose) vitamins within 7 days of first dosing prior to entrance into the clinical research facility and for the duration of the study.

In the interests of subject safety and acceptable standards of medical care the Investigator will be permitted to prescribe treatment(s) at his/her discretion. All treatments must be recorded in the subjects' case report form (CRF) (medication, dose, treatment duration and indication).

6.6 Method of Assigning Subjects to Treatment Groups

Following confirmation of eligibility, at study drug administration, subjects will be assigned a subject number in the order in which they are enrolled in the study, as shown in Table 6.1. The subject number will determine the allocation of treatment.

Table 6.1 Assignment of Subjects to Treatment

Study Part	Dose Level(s)	Subject Numbers
A	1, 2, 3	[redacted]
	4	[redacted]
	5	[redacted]
	6	[redacted]
	7	[redacted]
	Additional[1]	[redacted]
B	1	[redacted]
	2	[redacted]
	3	[redacted]
	Additional[1]	[redacted]

[1] eight additional cohorts of up to 6 subjects in Part A (up to an additional 48 patients) and three additional cohorts of up to 8 subjects in Part B (up to an additional 24 patients) may be enrolled in each part if agreed by the Safety Committee.

Any replacement subjects will receive the same treatment allocation as those whom they replace.

Subjects who are replaced (dropouts or replacements for subjects who are withdrawn from Cohort 1) will be allocated the same treatment number with the number 1 in the second digit position, e.g., if subject number [redacted] is replaced then the replacement subject's number will be [redacted].

6.7 Randomisation Procedures

Allocation to treatment will be according to a predetermined random order. Randomisation of Y14 or placebo will take place for each group separately. In Part A (SAD), each pair of sentinel subjects in Cohort 1 will be randomised such that 1 subject receives Y14 and 1 receives placebo.

The randomisation list will be generated by Covance using the statistical analysis system (SAS®) computer package.

6.8 Maintenance of Randomisation Codes

Randomisation codes will be provided to the pharmacist and bioanalyst in a list. Individual treatment disclosure envelopes (code break) will also be provided to the clinical unit. The pharmacist will use the randomisation code list for preparing subject doses throughout the study. The individual disclosure envelopes will be used if it is necessary to break the blind for an individual subject.

6.9 Blinding

Part A (SAD) will be conducted double-blind (with the exception of Cohort 1 where the volunteers taking the 3rd and 4th injections within a treatment period are guaranteed to be given Y14); subjects and clinical staff will remain blinded until the Safety Committee meeting.

Part B (MAD) will be conducted double-blind. In the event of a medical emergency when management of a subject's condition requires knowledge of the trial medication, the sealed "code-break" envelope provided may be opened by personnel authorised by the Principal Investigator to determine the nature of the trial medication dispensed. If possible, such emergencies should be discussed with the study monitor and the Sponsor's safety manager prior to disclosure of the treatment allocation. Reasons for breaking a code must be clearly explained and justified in the CRF. The date on which the code was broken together with the identity of the person responsible must also be documented.

The bioanalyst and the PK scientist who will be unblinded, will be required to provide coded data to the Principal Investigator and the Sponsor so that the blind is maintained for all other personnel connected with Part B of the study.

With the exception of the CRO pharmacy department, the statistician preparing the randomisation, the bioanalytical assay group and the quality assurance auditors where necessary, all clinical and non-clinical staff, with the exception of the PK scientist and PK/PD statistician, will remain blinded to the treatment allocation (where applicable) until after the database is locked unless there is a medical event that requires code break.

7 STUDY PROCEDURES

The blood volumes to be collected from each subject are detailed in Table 7.1 for Part A (SAD) and Table 7.2 for Part B (MAD). The total volume to be collected per subject will be no greater than 600 mL, staggered over the duration of the study. No more than 15% of the estimated blood volume of a subject will be taken on any one occasion as per JPAC guidelines (http://www.transfusionguidelines.org/).

Laboratory sample handling details will be presented in a separate laboratory manual.

Table 7.1 Blood Volumes: Part A (SAD)

Cohort 1

Assessment	Sample Volume (mL)	No. of Samples	Total Volume (mL)
Safety			
Serum Biochemistry, Thyroid Function	3.5	11	38.5
Serology	3.5	1	3.5
Coagulation	1.8	11	19.8
Haematology	4.0	11	44
Tryptase[1]	3.5	2	7
Bedside blood glucose	0.1	Up to 23	2.3
Pharmacokinetic			
Y14	6.0	Up to 17	102
Pharmacodynamics			
Immunogenicity[3]	6.0	2	12.0
Total[2]			**229.1**

[1] In the event of an allergic reaction additional samples will be collected.
[2] Sample volumes are based on direct venepuncture
[3]Immunogenicity samples from cohort 1 only

Cohort 2 onwards

Assessment	Sample Volume (mL)	No. of Samples	Total Volume (mL)
Safety			
Serum Biochemistry, Thyroid Function	3.5	5	17.5
Serology	3.5	1	3.5
Coagulation	1.8	5	9.0
Haematology	4.0	5	20.0
Tryptase[1]	3.5	2	7
Bedside blood glucose	0.1	Up to 11	1.1
Pharmacokinetic			
Y14	6.0	Up to 17	102.0
Total[2]			**160.1**

[1] In the event of an allergic reaction additional samples will be collected.
[2] Sample volumes are based on direct venepuncture.

Table 7.2 Blood Volumes: Part B (MAD)

Assessment	Sample Volume (mL)	No. of Samples	Total Volume (mL)
Safety			
Serum Biochemistry including Thyroid function (TSH, FT4)	3.5	16	56.0
Serology	3.5	1	3.5
Coagulation	1.8	16	28.8
Haematology	4.0	16	64.0
Tryptase[1]	3.5	2	7.0
Bedside glucose monitoring	0.1	54	5.4
Pharmacokinetic			
Y14	6.0	37	222.0
Pharmacodynamics			
Immunogenicity	6.0	2	12.0
Oral glucose tolerance test glucose and insulin levels	3.5	21	73.5
Enteropancreatic hormones (GLP-1, PYY, ghrelin, leptin)	5.0	21	105
Total[2]			577.2

[1] In the event of an allergic reaction additional samples will be collected.
[2] Sample volumes are based on direct venepuncture

7.1 Pharmacokinetic Assessments

7.1.1 Plasma Samples

Blood sample collection times are included in the schedule of events.

Details of how samples will be collected, processed and stored will be included in a separate laboratory manual.

Bioanalysis

The bioanalysis will be performed by [redacted]. They will be responsible for the GLP validated bioanalysis, which will utilise a high performance liquid chromatography-tandem mass spectrometry (LC-MS/MS) assay in human plasma.

Any remaining plasma may be sent to the sponsor for analysis of Y14 using an exploratory non-GLP compliant, non-validated radioimmunoassay.

7.2 Pharmacodynamic/Efficacy Assessments

Time points for collection of PD variables are included in the schedule of events.

7.2.1 Visual Analogue Scales

Assessments of nausea and satiety will be performed using VAS. Subjects will be requested to rate their responses to a series of questions on a VAS from 0 to 100 mm.

7.2.2 Energy Intake

Energy intake will be recorded for Part A and Part B.

A formal 24 h food intake study will be carried out on Part A from Cohort 3 onwards, on Day -1 (measuring lunch and dinner on Day -1 and breakfast on Day 1) and Day 1 (measuring lunch and dinner on Day 1 and breakfast on Day 2).

For Part B, the food intake studies will be carried out on Days -1, 2, and 30. The food intake study on Day -1 will measure the ad libitum intake of food during breakfast, lunch and dinner on Day -1. The food intake study on Day 2 will measure the ad libitum intake of food during breakfast, lunch and dinner on Day 2. The food intake study on Day 30 will measure the ad libitum intake of food during breakfast, lunch and dinner on Day 30. The timing of the food intake study will be altered according to emerging PK data, if the t_{max} is delayed beyond 24 hours. If the timing of the food intake studies are changed the times of the OGTT samples may also be changed to ensure they do not happen on the same day.

While resident in the clinical unit during 24 h food intake assessment days, subjects will be individually provided meals in a designated area with individual cubicles per subject. Meals will be presented with an excess of food and subjects will be asked to eat until they feel "comfortably full" within a period of 30 minutes. Should a volunteer require more time to eat, an extra 15 minutes will be allowed, but such an event will be recorded as a deviation from protocol. The energy value of each component of the meal will be determined. Food will be weighed pre- and post-meal to determine consumption.

For other days in Part A and Part B, energy intake will be carried out semiquantitively i.e. clinical research unit staff will record the timing, types and amounts of food eaten by subjects. For the semiquantitative measurement, at the end of each meal the percentage of food left will be assessed to be either <10%, 10-<25%, 25-<50% or ≥50%. Snack boxes will also be provided at certain timepoints whilst resident in the clinical unit (to be specified in the Meal Manual).

In Part B, subjects will be admitted on Day -2 (2 days prior to dosing) in order to acclimatise to the clinical research unit and obtain baseline food consumption measurements.

7.2.3 Body Weight

In Part A (SAD) and Part B (MAD), body weight will be recorded AM and PM each day of the inpatient stay and once on every outpatient visit (Part B).

Body weight for each subject should be measured on the same calibrated scale in underwear, after voiding urine, before breakfast (AM) and before dinner (PM), with the exception of days where subjects are having an OGTT without having breakfast, where they will be weighed prior to the OGTT.

At each timepoint the weight will be measured in triplicate and the mean calculated. The weight should be measured in kg to one decimal place.

7.2.4 Fluid Intake/Output

In Part A (SAD) and B (MAD) fluid intake will also be measured on the days when the subjects are undergoing food intake studies as per the Study Schedules.

7.2.5 Immunogenicity

For assessment of immunogenicity, plain tube serum samples (6 mL blood volume) will be collected prior to the first dose and at the final follow-up visit for SAD Cohort 1 and the MAD study, as subjects will be receiving multiple doses of the same peptide in these studies. Samples will be analysed using a GLP compliant and validated assay method utilising an ELISA (bridging assay format) using rabbit positive control antibodies raised against Y14.

7.3 Safety Assessments

7.3.1 Adverse Events

Definitions:

Adverse Event (AE)

Any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

An AE can therefore be any clinically significant sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Adverse Drug Reaction (ADR)

Any untoward and unintended response in a subject to an investigational medicinal product which is related to any dose administered to that subject.

Serious Adverse Event (SAE)

An adverse reaction is 'serious' if it:

25. Results in death;
26. Is life-threatening;
27. Requires hospitalisation or prolongation of existing hospitalisation;
28. Results in persistent or significant disability or incapacity;
29. Consists of a congenital anomaly or birth defect;
30. Is a medically important event.

Unexpected Adverse Reactions

An adverse reaction is 'unexpected' if its nature and severity are not consistent with the information about the medicinal product in question set out:

31. In the case of a product with a marketing authorisation, in the summary of product characteristics for that product;
32. In the case of any other investigational medicinal product, in the Investigator's Brochure relating to the trial in question.

7.3.2 Reporting of Adverse Events

All adverse events must be fully recorded in the adverse event book throughout the entire study period and will be transcribed into the subjects' CRF, whether or not they are considered to be drug-related. Signs and symptoms of each AE should be described in detail: onset time and date, offset time and date, description of event, severity, relationship to investigational product, action taken and outcome.

Adverse events should be followed until recovery to the normal state has been achieved. In the event of a subject not returning to the clinical unit, the outcome of this event will be recorded as lost at follow up.

Reporting of SAEs and SUSARs

SAEs occurring on this study will be reported to the Sponsor's medical representative Dr Tricia Tan within agreed timelines. The Investigator will be requested to complete a separate SAE reporting form in addition to the information on the CRF and in the AE book.

The Medicines for Human Use (Clinical Trials) Regulations 2004 (Statutory Instrument 1031) and subsequent amendments define the following terms:

An adverse reaction is any untoward and unintended response in a subject to an investigational medicinal product which is related to any dose administered to that subject.

A suspected unexpected serious adverse reaction (SUSAR) which is fatal or life-threatening must be reported to the competent authority and ethics committee immediately (within 7 days) after the Sponsor became aware of the event. Any additional information must be reported within eight days of sending the first report.

A SUSAR which is not fatal or life-threatening must be reported to the competent authority and ethics committee as soon as possible (within 15 days) after the Sponsor becomes aware of the event.

If a clinical trial is being conducted at a trial site in a third country in addition to sites in the United Kingdom, the Sponsor of that trial shall ensure that all SUSARs occurring at that site are entered into the European database established in accordance with Article 11 of the EU Clinical Trials Directive.

As soon as practicable after the end of the reporting year, a Sponsor shall, in relation to each investigational medicinal product tested in clinical trials in the United Kingdom for which he is the Sponsor provide the licensing authority and the relevant ethics committees with -

1. A list of all the suspected serious adverse reactions which have occurred during that year in relation to:
    - those trials, whether at trial sites in the United Kingdom or elsewhere;
    - or any other trials relating to that product which are conducted outside the United Kingdom and for which he is the Sponsor;

    including those reactions relating to any investigational medicinal product used as a placebo or as a reference in those trials; and
2. A report on the safety of the subjects of those trials.

7.3.3 Categorisation of Adverse Events

The intensity of an AE will be categorised as follows:

Mild:	Mild events are those which are easily tolerated with no disruption of normal daily activity.
Moderate:	Moderate events are those which cause sufficient discomfort to interfere with daily activity.
Severe:	Severe events are those which incapacitate and prevent usual activity.

7.3.4 Causal Relationship Assessment

Causal relationship assessment to drug treatments is required for purposes of reporting AEs. To promote consistency, the following guidelines should be taken into considerations along

with good clinical and scientific judgment when determining the relationship of drug treatments to an AE:

Definitely Related:	A clinical event, including laboratory test abnormality, occurring in a plausible time relationship to the medication administration, and which cannot be explained by concurrent disease or other drugs or chemicals. The response to withdrawal of the drug should be clinically plausible.
Possibly Related:	A clinical event, including laboratory test abnormality, with a reasonable time sequence to the medication administration, but which could also be explained by concurrent disease or other drugs or chemicals. Information on the drug withdrawal may be lacking or unclear.
Unlikely Related:	A clinical event, including laboratory test abnormality, with little or no temporal relationship to medication administration, and which other drugs, chemicals or underlying disease provide plausible explanations.
Not Related:	A clinical event, including laboratory test abnormality that has no temporal relationship to the medication or has more likely alternative aetiology.

7.3.5 Action Taken

Action taken will be defined as:

- None;
- Medication given;
- Dosing interrupted (this applies to Part B only as it involves multiple dosing);
- Dosing stopped (this applies to Part B only as it involves multiple dosing).

7.3.6 Outcome

Outcome will be defined as:

- Resolved;
- Ongoing;
- Lost to follow up.

7.3.7 Coding of Adverse Events

All adverse events will be coded using the current version of Medical Dictionary for Regulatory Activities (MedDRA).

7.4 Clinical Laboratory Safety Tests

Sample collection times are included in the schedule of events.

Additional and repeat testing may be performed at the discretion of the Principal Investigator. Safety laboratory variables to be assessed are presented in Table 7.3.

Table 7.3 Safety Laboratory Variables

Haematology	White blood cells	Haematocrit
	Red blood cells	Neutrophils (absolute and %)
	Mean corpuscular volume	Lymphocytes (absolute and %)
	Mean corpuscular haemoglobin	Monocytes (absolute and %)
		Eosinophils (absolute and %)
	Mean corpuscular haemoglobin concentration	Basophils (absolute and %)
		Platelets
	Haemoglobin	
Biochemistry	Sodium	Total bilirubin
	Potassium	Calcium
	Urea	Chloride
	Creatinine	Total protein
	Alkaline phosphatase	Globulin
	Alanine aminotransferase	Cholesterol
	Creatine kinase	Triglycerides
	Gamma-glutamyltransferase	Uric acid
	Lactate dehydrogenase	Phosphate
	Aspartate aminotransferase	Albumin
	Glucose	Bicarbonate
	Lipase	C-reactive protein[1]
	Amylase	
Coagulation	Prothrombin time	Activated partial thromboplastin time
	International normalized ratio	
Thyroid Function	TSH, FT4	
Biomarkers	Histamine	Tryptase
Urinalysis	Protein	Leukocytes
	Bilirubin	Red blood cells
	Urobilinogen	pH
	Ketones	Nitrite
	Glucose	Specific gravity
Urine Microscopy	Microscopic investigation of sediment in case of pathological findings [performed if clinically indicated]	
Viral Serology	HIV I and II	Hepatitis C virus
	HBsAg	
Drugs of Abuse and Alcohol Screen	Amphetamine / Ecstasy	Benzodiazepines
	Cannabinoids	Methadone metabolites
	Cocaine	Barbiturates
	Opiates	Cotinine
Alcohol Screen (Breath test)	Alcohol	

[1] At screening only

Unless otherwise specified in the schedule of assessments all clinical laboratory tests will be performed by CRO Pathology Lab, which is a GLP Accredited Laboratory. Details of all methodology and reference ranges are provided in the Trial Master File.

7.5 Clinical Safety Assessments

Assessment times are included in the schedule of events. Time points of assessments may be amended based on emerging safety, tolerability and PK data.

7.5.1 Vital Signs

Blood pressure and pulse rate will be measured using an automated instrument with the subject in the supine position after resting comfortably for 10 minutes. Body temperature will be measured orally in degrees Celsius using an automated thermometer. Additional vital signs measurements may be added for safety of the subjects.

Measurements will be reported in the subject's CRF.

7.5.2 12-Lead ECG

Computerised 12-lead ECG recordings will be obtained. after 5 minutes supine rest. Each lead shall be recorded for at least 3 beats at a speed of 25 mm/sec.

The following parameters will be recorded: ventricular rate, PR interval, QRS duration, QT and QTc.

7.5.3 Medical History

A complete medical history will include evaluation (past or present) of the following: general, head and neck, eyes, ears, nose throat, chest/respiratory, heart/cardiovascular, gastrointestinal/liver, gynecological/urogenital, musculoskeletal/extremities, skin, neurological/psychiatric, endocrine/metabolic, haemotologic/lymphatic, allergies/drug sensitivities, past surgeries, substance abuse or any other diseases or disorders.

7.5.4 Physical Examination

Physical examinations will be performed by a physician and will include the examination of the following: general appearance, eyes, ears, nose, throat, chest/respiratory, heart/cardiovascular, gastrointestinal/liver, musculoskeletal/extremities, dermatological/skin, thyroid/neck, lymph nodes, neurological/psychiatric.

7.5.5 Bedside glucose testing

Where indicated in the Study Schedules, bedside glucose testing will be carried out.

7.5.6 Local Injection Site Reactions

Injection sites will be monitored while the subject is resident in the clinical unit for redness, swelling, pain, tenderness and bruising. Reaction grades are provided in Table 7.4.

In the event that there are injection site reactions thought to be related to the volume of the injection, subsequent doses may be split into a maximum of 5 smaller injections.

Injection site reactions with a Grade ≥1 will be recorded as an AE.

Table 7.4 Injection Site Reaction Grading Scheme

Reaction	Grade 0	Grade 1	Grade 2	Grade 3	Grade 4
Redness[1]	0–24 mm	25–50 mm	51–100 mm	More than 100 mm	Requires medical intervention greater than analgesia
Swelling[2]	0–24 mm	25–50 mm and does not interfere with activity	51–100 mm or interferes with activity	More than 100 mm and prevents daily activity	Requires medical intervention greater than analgesia
Pain	None	Does not interfere with activity	Interferes with activity or repeated use of non-narcotic pain reliever	Prevents daily activity or repeated use of narcotic pain reliever	Requires medical intervention greater than analgesia
Tenderness	None	Mild pain to touch	Moderate pain to touch	Severe pain to touch	Requires medical intervention greater than analgesia
Bruising	None	Present			
Ulceration ± tissue necrosis	None	None	None	None	Present

1 Assessed by estimating the size of the red patch at the injection site across its widest point
2 Assessed by estimating the size of the raised area around the injection site across its widest point; also take into account how much it affects the subject in their routine daily activities.

8 DATA COLLECTION

An electronic data capture system will be used in this trial. Data will be captured onto source data documents (Workbooks) and will be entered into the Remote Data Capture system by staff at the clinical site. Following data entry, the data will undergo quality control checks. Any discrepancies will be resolved in the database.

Following all data validation steps, the Principal Investigator, or designee, will electronically sign each eCRF prior to database lock.

9 EVALUATION OF STUDY DATA

9.1 Evaluation of Pharmacokinetic Parameters

Pharmacokinetic parameters of Y14 will include: maximum observed concentrations (C_{max}), time of occurrence of C_{max} (T_{max}), area under the plasma concentration-time curves ($AUC_{0-\infty}$, AUC_{0-t}, $AUC_{0-\tau}$), the apparent terminal rate constant (λ_z), the apparent terminal half-life ($t_{½}$) and the extent of accumulation in plasma (R_O). Pharmacokinetic parameters will be calculated, where appropriate, by non-compartmental analysis using WinNonlin Pro Version 5.2.1 (or higher version). Full details of the pharmacokinetic analysis, associated statistics and reporting will be documented in a separate Pharmacokinetic Analysis Plan which will be distributed to the Sponsor and Principal Investigator for approval.

9.2 Evaluation of Pharmacodynamic Measures

9.2.1 Visual Analogue Scales

Change from baseline values will be calculated using Day -1 values as baseline.

9.2.2 Energy Intake

The energy value per unit of weight will be determined for each individual component of a meal. Each component of the meal will be weighed pre and post-meal and the weight consumed will be calculated (i.e. weight pre meal - weight post meal = weight consumed). Total energy intake will be calculated from the weight of each component consumed.

9.2.3 Body Weight

Change from baseline values will be calculated using the mean of Day -1 (AM and PM) and Day 1 (predose) values as baseline.

9.2.4 Immunogenicity

Immunogenicity data will be presented as 'yes/no' based on a cut-off for binding to labelled Y14 in the immunogenicity assay (Section 7.2.5).

9.3 Evaluation of Safety

The safety evaluation will include blood pressure, pulse rate, ECG parameters, clinical laboratory tests (haematology, serum biochemistry, coagulation, urinalysis and urine microscopy). Histamine and tryptase determinations will only be performed after relevant adverse events and analysed retrospectively to inform possible mechanism of AEs.

10 STATISTICAL METHODS

10.1 Primary and Secondary Target Variables

Primary Target Variables

- Safety and tolerability as assessed by adverse events, vital signs, physical examination, clinical laboratory safety assessments, and ECG parameters.

Secondary Target Variables

- Pharmacokinetics of Y14 as measured by plasma concentrations and derived pharmacokinetic parameters.

Exploratory Target Variables

- The effects of multiple doses of Y14 on food consumption.
- The effects of multiple doses of Y14 on VAS (nausea and satiety).
- The effects of multiple doses of Y14 on body weight.
- The effects of multiple doses of Y14 on enteropancreatic hormone levels.
- The comparative analytical performance of an Imperial College radioimmunoassay versus the LC-MS/MS assay for Y14.
-

10.2 Sample Size Determination

This is a Phase I study to investigate the safety and tolerability of a novel compound, an appropriate sample size cannot be statistically determined and hence the sample size chosen was based on previous experience in Phase I studies.

10.3 Subject Population for Analyses

Safety Population

All subjects who receive study medication (Y14 or placebo) will be included in the safety population.

Pharmacokinetic Population

All subjects who receive Y14 and have data from at least 1 PK blood draw will be included in the PK population.

Pharmacodynamic Population

All subjects who receive study medication and have data from at least 1 PD blood draw or measurement, as appropriate will be included in the PD population.

10.4 Pharmacokinetic Analysis

[redacted] will be responsible for the pharmacokinetic analysis. The pharmacokinetic analysis will be conducted at a Good Laboratory Practice compliant facility which operates a Quality Management System (QMS). The QMS has been designed to be compatible with the Organisation for Economic Co-operation and Development (OECD) Principles of GLP and GCP requirements. The pharmacokinetic analysis will be reported at [redacted] and the report will be included as an appendix to the clinical report. All data generated at [redacted] will be dispatched to the Sponsor and archived according to their archiving procedures.

A review of dosing information will be performed to consider excluding data in any period or on any day where a subject was judged to have received <80% or >120% of the scheduled dose of the investigational product; resulting exclusions will be documented in advance of issuing the draft report and agreed with the Sponsor. Plasma concentration data will be excluded for the affected period or day if concentrations are extremely low relative to other subjects' data; in these cases, plasma concentrations will be excluded from all or part of the profile, as appropriate.

PK parameters will be excluded from the analysis and summary statistics, where there are insufficient plasma concentration data available.

Plasma concentration data will be summarised by sampling time, dose level and Day, as appropriate; pharmacokinetic parameters (e.g. $AUC_{0-\infty}$, AUC_{0-t}, $AUC_{0-\tau}$, C_{max}, t_{max}, $t_{½}$ and R_O) will be summarised by dose level and Day, as appropriate.

All individual plasma and pharmacokinetic parameter estimates will be listed and summarised. Mean and individual plasma concentration versus time profiles will be illustrated using both linear-linear and logarithmic-linear scales.

Summary statistics will include number of subjects (n), arithmetic mean and standard deviation (SD). Summaries for the pharmacokinetic parameters will also display the median, minimum and maximum. In addition, the geometric mean and geometric coefficient of variation

($CV = \sqrt{\exp(SD_{ln}^{\ 2}) - 1} * 100$, where SD_{ln} is the standard deviation of the natural logarithmically transformed data) will be reported for all pharmacokinetic parameters except t_{max}.

Between-subject variability will be based on geometric mean coefficients of variation (CVs).

A non-linear power model will be used to assess dose-proportionality. The proportional relationship between each parameter and dose is written as a power function:

$y = a \times dose^b$ (Equation 1)

where 'a' is a constant, 'b' is the proportionality constant and 'y' is the parameter of interest ($AUC_{0-\infty}$, AUC_{0-t}, $AUC_{0-\tau}$ or C_{max}). $AUC_{0-\tau}$ (after repeated dosing) $AUC_{0-\infty}$ (or AUC_{0-t} if AUC_{0-}

$_{\infty}$ could not be reliably estimated in all subjects) and C_{max} will be each plotted against dose. The exponent, b, will be estimated by performing a linear regression of the logged parameter on log dose. The exponent, b, is the estimated slope of the resulting regression line since taking logs of Equation (1) gives the linear relationship, log y = log a + b×log dose. The relationship is dose-proportional when b = 1. The exponents and 95 % confidence intervals (CIs), b_{lower} (b_l) and b_{upper} (b_u), are presented. There would be evidence of non dose-proportionality if this CI excludes one. The estimate of the fold increase in exposure for a doubling in dose (with 95 % CI) will also be presented. The increase in exposure expected for a doubling in dose will be calculated as 2^b (95 % CI: 2^{bl}, 2^{bu}).

10.5 Pharmacodynamic Analysis

Observed values for all pharmacodynamic parameters will be listed and where appropriate, summarised with descriptive statistics.

Change-from-baseline may be calculated if appropriate.

Immunogenicity data will be presented as 'yes/no' based on a cut-off of binding to labelled Y14 in the immunogenicity assay (Section 7.2.5).

10.6 Safety Data Analysis

Safety data analysis will be performed by Covance Clinical Research Unit. Individual and summary blood pressures, pulse rate, ECG parameters and clinical laboratory data (haematology, serum biochemistry, urinalysis and coagulation) will be presented in tabular form with mean, median, standard deviation and range (min and max) as appropriate. Continuous variables will be presented in tabular form with mean, median, standard deviation and range (minimum and maximum) as appropriate. Categorical variables will be summarised in frequency tables (frequency and proportion).

Histamine and tryptase values will be listed if the analysis has been performed and data is available.

For the laboratory safety data, out of range values will be flagged in the data listings and a list of clinically significantly abnormal values will be presented.

Adverse events will be tabulated and summarised according to the current version of Medical Dictionary for Regulatory Activities (MedDRA).

11 REGULATORY AND ETHICAL ISSUES

11.1 Regulatory and Ethics Review and Approval

The study will be submitted to the MHRA (UK) for review and approval and to an Independent Ethics Committee for ethical review and approval. The documents submitted will include but are not limited to:

MHRA:

3. The final protocol;
4. The Investigators Brochure (IB);
5. The Investigational Medicinal Product Dossier (IMPD); and
6. Annex 1: Clinical Trial Application Form (Request for authorisation of a clinical trial on a medicinal product for human use to the competent authorities and for opinion of the ethics committees in the community).

Ethics:

7. The final protocol;
8. The Investigators Brochure (IB);
9. The ethics application form; and
10. The Informed Consent Form (ICF)

The study will not commence unless the following conditions are satisfied:

11. An ethics committee has given a favourable opinion in relation to the clinical trial; and
12. The clinical trial has been authorised by the licensing authority (MHRA).

11.2 Informed Consent

Subjects will sign the pre-screening/generic ICF in the presence of a suitably trained CRU clinical operations staff member prior to the pre-screening procedures being performed (if applicable).

For the main study, informed consent will be given freely after the subject has been informed of the nature, significance, implications and risks of the trial; and consent is evidenced in writing, dated and signed, or otherwise marked, by that person so as to indicate his/her consent, prior to the start of the study. The nature of the informed consent will comply with the current version of the Declaration of Helsinki, the current requirements of GCP (CPMP/ICH/135/95) and local regulation (The requirements are set out in Schedule 1 of Statutory Instrument 1031 and amendments) which ever provides the greater subject protection.

11.3 Indemnity and Compensation

In accordance with Statutory Instrument 1031 and amendments section 15 (5i, j) and the EU Clinical Trials Directive 2000/20/EC Article 3 (2f), provision is to be made for:

1. The indemnity or compensation in the event of injury or death attributable to the clinical trial; and
2. Insurance or indemnity to cover the liability of the Investigator or Sponsor.

Therefore the Sponsor, Imperial College, will indemnify the Investigator, Covance Clinical Research Unit, from all and any claims arising out of this study except for their negligence or malpractice and providing that the study is conducted according to the standards established by the protocol.

In the event that it can be demonstrated that a subject suffers any significant deterioration in health or well-being or any harmful susceptibility or toxicity as a direct result of their participation in this study then Imperial College will agree to abide by the current Association of the British Pharmaceutical Industry Guidelines with regard to compensation payable to the subject. The amount of compensation will be calculated by reference to the level of damages commonly awarded in the UK for similar injuries at the time when such injury occurred.

The Investigator, Covance Clinical Research Unit, declare to having insurance cover for the malpractice and/or negligence of their employees and agents.

12 STUDY MANAGEMENT

12.1 Quality Assurance and Quality Control

In accordance with the guideline for ICH GCP, the Sponsor has responsibility for implementing and maintaining quality assurance and quality control systems, and the ultimate responsibility for the quality integrity of the trial data resides with the Sponsor.

Authorised representatives of the Sponsor may perform audits or inspections during the study. The purpose of an audit is to independently assure that the study is conducted and that data is collected and reported according to the protocol, Good Clinical Practice (GCP), guidelines of the International Conference on Harmonisation (ICH), and any applicable regulatory requirements.

12.2 Protocol Adherence

The protocol must be read thoroughly and the instructions followed exactly. Any deviations should be agreed by both the Sponsor and the Investigator, with the appropriate written and approved protocol amendments made to reflect the changes agreed upon. Where the deviation occurs for the well-being of the subject, the Sponsor must be informed of the action agreed upon.

12.3 Documents Necessary for Initiation of Study

The following documents will be available prior to the first administration of the drug to the first subject:

1. Regulatory authorisation;
2. Copy of current Investigator's Brochure;
3. Risk assessment report;
4. Completed and signed investigator agreement/contract;
5. Signed original of the final protocol;
6. Ethics Committee approval;
7. Copy of the constitution of the Research Ethics Committee;
8. A list of members of the Ethics Committee;
9. A copy of the consent form and subject information to be used;
10. The curriculum vitae of all Investigators;
11. The Qualified Person's certification for the release of each batch of IMP;
12. A technical agreement between the Sponsor and Covance defining the responsibilities of the Sponsor, Covance Clinical Research Unit, and any third parties significantly involved in the supply chain of the IMP, where applicable;

13. The product specification file, where applicable; and

14. A list of laboratory reference range values for parameters measured in the study.

12.4 Study Monitoring

In accordance with ICH GCP, the Sponsor will be responsible for monitoring the conduct of the study. An independent study monitor will be appointed before the study begins.

The study monitor, will conduct pre-study and start-up meeting site visits and in addition will visit the site during the conduct of the study, review the study data and conduct a post-study visit. Study data recorded on source documents will be made available to the study monitor for the purpose of source document verification. The monitor will verify that the:

- data are authentic, accurate, and complete.
- safety and rights of subjects are being protected.
- study is conducted in accordance with the currently approved protocol and any other study agreements, GCP, and all applicable regulatory requirements.

The Investigator will allow trial-related monitoring audits, IEC review, and regulatory inspection allowing direct access to the source data/documents.

12.5 Study Closure

The Sponsor will be responsible for the close out visit which will be conducted on clinical completion of the study.

12.6 Study Record Retention

In accordance with SI 1928, the Sponsor and the Principal Investigator shall ensure that the documents contained, or which have been contained, in the trial master file are retained for at least 15 years after the conclusion of the trial and that during that period are:

- Readily available to the licensing authority on request; and
- Complete and legible.

All data derived from the study will remain the property of the Sponsor. The study will be the subject of a final clinical study report compiled by, or by order of the Sponsor.

All correspondence (e.g. with the Sponsor, ethics committee) relating to this study should be kept in the appropriate file folders.

Records of subjects' source documents, CRFs, IMP inventory, pertaining to the study must be kept on file. Records must be retained according to the current ICH Guidelines on GCP.

The Sponsor and Principal Investigator shall ensure that the medical files of trial subjects are retained for at least 15 years after the conclusion of the trial. The Sponsor shall appoint named individuals within his organisation to be responsible for archiving the documents which are, or have been, contained in the trial master file. Access to those documents shall be restricted to

those appointed individuals. If there is transfer of ownership of data or documents connected with the clinical trial:

- The Sponsor shall record the transfer; and
- The new owner shall be responsible for data retention and archiving in accordance Statutory Instrument 1031 and amendments.

If the Investigator moves, withdraws from an investigation or retires, the responsibility for maintaining the records may be transferred to another person who will accept responsibility. Notice of transfer must be made to and agreed by the Sponsor.

12.7 Publication Policy

After completion of the study, the Investigator may prepare a joint publication with the Sponsor. The Investigator must undertake not to submit any part of the individual data from this protocol for publication without prior consent of the Sponsor at a mutually agreed time.

13 REFERENCES

1. Wynne K, Stanley S, McGowan B, Bloom S. Appetite control. J Endocrinol. 2005 Feb;184(2):291-318.
2. Dakin CL, Gunn I, Small CJ, Edwards CM, Hay DL, Smith DM, et al. Oxyntomodulin inhibits food intake in the rat. Endocrinology. 2001 Oct 1;142(10):4244-50.
3. Dakin CL, Small CJ, Batterham RL, Neary NM, Cohen MA, Patterson M, et al. Peripheral oxyntomodulin reduces food intake and body weight gain in rats. Endocrinology. 2004 Jun 1;145(6):2687-95.
4. Cohen MA, Ellis SM, Le Roux CW, Batterham RL, Park A, Patterson M, et al. Oxyntomodulin suppresses appetite and reduces food intake in humans. J Clin Endocrinol Metab. 2003 Oct 1;88(10):4696-701.
5. Baggio LL, Huang Q, Brown TJ, Drucker DJ. Oxyntomodulin and glucagon-like peptide-1 differentially regulate murine food intake and energy expenditure. Gastroenterology. 2004 Aug 1;127(2):546-58.
6. Batterham RL, Cohen MA, Ellis SM, Le Roux CW, Withers DJ, Frost GS, et al. Inhibition of food intake in obese subjects by peptide YY3-36. N Engl J Med. 2003 Sep 4;349(10):941-8.
7. Batterham RL, Cowley MA, Small CJ, Herzog H, Cohen MA, Dakin CL, et al. Gut hormone PYY(3-36) physiologically inhibits food intake. Nature. 2002 Aug 8;418(6898):650-4.
8. The Comptroller and Auditor General. Tackling Obesity in England. Report by the Auditor and Comptroller General (HC 220 Session 2000-2001). London: National Audit Office; 2001.
9. Naslund E, Gryback P, Hellstrom PM, Jacobsson H, Holst JJ, Theodorsson E, et al. Gastrointestinal hormones and gastric emptying 20 years after jejunoileal bypass for massive obesity. Int J Obes. 1997 May;21(5):387-92.
10. The NHS Information Centre. Statistics on obesity, physical activity and diet: England, 2016. http:// content.digital.nhs.uk/catalogue/PUB20562/obes-phys-acti-diet-eng-2016-app.pdf
11. Flegal KM, Kruszon-Moran D, Carroll MD, Fryar CD, Ogden CL. Trends in Obesity Among Adults in the United States, 2005 to 2014. JAMA. 2016 Jun 7;315(21):2284–91.
12. Finer N. Pharmacotherapy of obesity. Best practice & research. Clinical endocrinology & metabolism. 2002 Dec;16(4):717-42.
13. European Medicines Agency. European Medicines Agency recommends suspension of marketing authorisation for sibutramine. 2010 [cited 2011 28 Feb]; Available from:

http://www.ema.europa.eu/ema/index.jsp?curl=pages/news_and_events/news/2010/01/news_detail_000985.jsp&murl=menus/news_and_events/news_and_events.jsp&mid=WC0b01ac058004d5c1.

14. Y14 Investigator's Brochure. Edition 01, 7th Feb 2017
15. US Department of Health and Human Services Food and Drug Administration, Center for Drug Evaluation and Research (CDER). Guidance for Industry: Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult healthy Volunteers. July 2005.
16. le Roux CW, Borg CM, Murphy KG, Vincent RP, Ghatei MA, Bloom SR. Supraphysiological doses of intravenous PYY3-36 cause nausea, but no additional reduction in food intake. Annals of clinical biochemistry. 2008 Jan;45(Pt 1):93-5.
17. Degen L, Oesch S, Casanova M, Graf S, Ketterer S, Drewe J, et al. Effect of peptide YY3-36 on food intake in humans. Gastroenterology. 2005 Nov 1;129(5):1430-6.
18. Sloth B, Holst JJ, Flint A, Gregersen NT, Astrup A. Effects of PYY_{1-36} and PYY_{3-36} on appetite, energy intake, energy expenditure, glucose and fat metabolism in obese and lean subjects. Am J Physiol Endocrinol Metab. 2007 Apr 1;292(4):E1062-8.
19. Sloth B, Davidsen L, Holst JJ, Flint A, Astrup A. Effect of subcutaneous injections of PYY1-36 and PYY3-36 on appetite, ad libitum energy intake, and plasma free fatty acid concentration in obese males. Am J Physiol Endocrinol Metab. 2007 Aug;293(2):E604-9.
20. Astrup A, Rossner S, Van Gaal L, Rissanen A, Niskanen L, Al Hakim M, et al. Effects of liraglutide in the treatment of obesity: a randomised, double-blind, placebo-controlled study. Lancet 2009; 374 (9701): 1606-16.
21. Playford RJ, Mehta S, Upton P, Rentch R, Moss S, Calam J, et al. Effect of peptide YY on human renal function. Am J Physiol. 1995 Apr;268(4 Pt 2):F754-9.
22. Gutzwiller J-P, Tschopp S, Bock A, Zehnder CE, Huber AR, Kreyenbuehl M, et al. Glucagon-like peptide 1 induces natriuresis in healthy subjects and in insulin-resistant obese men. J Clin Endocrinol Metab. 2004;89(6): 3055-61.
23. Crajoinas RO, Oricchio FT, Pessoa TD, Pacheco BP, Lessa LM, Malnic G, Girardi AC. Mechanisms mediating the diuretic and natriuretic actions of the incretin-hormone glucagon-like peptide-1. AJP: Renal Physiology. 2011; epub ahead of print (doi: 10.1152/ajprenal.00729.2010).
24. Van Strien T, Rookus MA, Bergers GP, Frijters JE, Defares PB. Life events, emotional eating and change in body mass index. Int J Obes. 1986; 10:29-35.
25. Morgan JF, Reid F, Lacey JH. The SCOFF questionnaire: assessment of a new screening tool for eating disorders. BMJ, 1999;319:1467-8.