CLINICAL STUDY PROTOCOL

An Open-Label, Single-Dose, Single-Period Study Designed to Assess the Mass Balance Recovery, Metabolite Profile and Metabolite Identification of [^{14}C]-MD1003 in Healthy Male Subjects

Quotient Study Number: QSC201639
Sponsor Study Number: MD1003CT2019-03MB
EudraCT Number: 2019-003122-24

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CONFIDENTIALITY AND GCP COMPLIANCE STATEMENT

I confirm that I have read and that I understand this protocol, the Investigator's Brochure, and any other product information provided by the sponsor. I agree to conduct this study in accordance with the requirements of this protocol and also protect the rights, safety, privacy, and well-being of study subjects in accordance with the following:

- The ethical principles that have their origin in the Declaration of Helsinki.
- International Council for Harmonisation E6 (R2) Good Clinical Practice: Consolidated Guideline.
- All applicable laws and regulations, including, without limitation, data privacy laws and regulations.
- Regulatory requirements for reporting serious adverse events defined in Section 16.3 of this protocol.

Information taken from the study protocol may not be disseminated or discussed with a third party without the express consent of the sponsor.

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3 Synopsis

Sponsor:	**Drug Substance:**	**EudraCT No.:**
MedDay Pharmaceuticals	[^{14}C]-MD1003	2019-003122-24

Title of Study:

An Open-Label, Single-Dose, Single-Period Study Designed to Assess the Mass Balance Recovery, Metabolite Profile and Metabolite Identification of [^{14}C]-MD1003 in Healthy Male Subjects

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Objectives and Endpoints:

Objectives	Endpoints
Primary • To determine the mass balance recovery after a single oral dose of carbon-14 [^{14}C]-MD1003 • To perform metabolite profiling and structural identification of MD1003 metabolites from plasma, urine and faecal samples	• Mass balance recovery of total radioactivity in all excreta (urine and faeces): Ae, %Ae, Cum Ae and Cum %Ae • Metabolite profiling, structural identification, and quantification of [^{14}C]-MD1003 metabolites in plasma, urine and faeces
Secondary • To determine the routes and rates of elimination of [^{14}C]-MD1003 • To identify the chemical structure of each metabolite accounting for more than 10% of circulating total radioactivity • To further explore the oral pharmacokinetics (PK) of MD1003 and its metabolites bisnorbiotin and biotin sulfoxide • To evaluate the extent of distribution of total radioactivity into blood cells • To provide additional safety and tolerability information for MD1003	• Amount of total radioactivity excreted and amount of total radioactivity excreted as a percentage of the administered dose in urine and faeces at each time interval • Identification of the chemical structure of each metabolite accounting for more than 10% by AUC of circulating total radioactivity • Assessment of the oral PK profile for MD1003, bisnorbiotin, biotin sulfoxide and total radioactivity based on: Tlag, Tmax, Cmax, AUC(0-last), AUC(0-inf), AUCextrap, T1/2, Lambda-z, CL/F (MD1003 only), Vz/F (MD1003 only), MPR Cmax and MPR AUC(0-inf) • Evaluation of whole blood:plasma concentration ratios for total radioactivity • To provide additional safety and tolerability information for MD1003 by assessing: adverse events (AEs), vital signs, electrocardiograms (ECGs), physical examinations and laboratory safety

The metabolite profile and structural identification objectives will be reported separately.

Methodology:

This is a single-centre, open-label, non-randomised, single oral dose study in healthy male subjects. It is planned to enrol 6 subjects to ensure 4 evaluable subjects.

Each subject will receive a single oral administration of a capsule containing 100 mg MD1003 and not more than 2.22 MBq (approximately 60.0 µCi) [^{14}C], in the fasted state.

Study Design:

Subjects will be screened to participate in the study up to 28 days before dosing. Subjects will be admitted to the clinical unit on the evening of Day -1 prior to investigational medicinal product (IMP) administration.

Subjects will be dosed on the morning of Day 1 following an overnight fast and will remain resident in the clinical unit until up to 168 h after dosing (up to Day 8). It is planned that subjects will be released as a group when all subjects have achieved a mass balance cumulative recovery of >90% or if <1% of the dose administered has been collected in urine and faeces within 2 separate consecutive 24 h periods. This may result in the subjects being discharged as a group prior to completion of the planned residency period. Once the discharge criteria or the planned residency period have been achieved, collection of all samples (blood, urine and faeces) will be stopped and the subjects will undergo discharge assessments. If the mass balance criteria have not been met by all subjects by Day 8, the residency period for the subjects not achieving the release criteria may be extended up to a maximum of an additional 48 h (up to Day 10) for further collection of urine and/or faeces. If the release criteria have still not been met by Day 10, or if any additional residency is not considered appropriate or necessary, then home collections of urine and/or faeces may be requested at the discretion of the investigator for individual subjects.

Number of Subjects Planned:

It is planned to enrol 6 healthy male subjects to ensure data in 4 evaluable subjects. A subject will be considered evaluable if they have provided mass balance and PK samples for up to 72 h (up to Day 4) after IMP administration, or have demonstrated >90% mass balance recovery, or have <1% of the administered dose eliminated in excreta for 2 consecutive days, whichever is sooner. No replacement subjects are to be used in this study.

Duration of Study:

Single oral dose administration on a single occasion. The estimated time from screening until the end of the study is approximately up to 6 weeks.

Main Inclusion Criteria:

Healthy males aged 30 to 65 years.

Body mass index (BMI) 18.0 to 30.0 kg/ m^2.

Investigational Medicinal Product, Dose and Mode of Administration:

The following IMP will be used in this clinical study.

Treatment	IMP Name	Dose	Route of Administration
Single dose	[^{14}C]-MD1003 Capsule	100 mg (NMT 2.22 MBq; 60.0 µCi ^{14}C)	Oral, fasted

^{14}C: carbon 14, NMT: not more than

All doses will be administered orally with 240 mL water, following an overnight fast.

Mass Balance Assessments:

Mass balance of total radioactivity in urine and faeces will be calculated as follows:

Parameter	Definition
Ae(urine)	amount of total radioactivity excreted in urine
%Ae(urine)	amount of total radioactivity excreted in urine expressed as a percentage of the radioactive dose administered
CumAe(urine)	cumulative amount of total radioactivity excreted in urine
Cum%Ae(urine)	cumulative amount of total radioactivity excreted in urine expressed as a percentage of the radioactive dose administered
Ae(faeces)	amount of total radioactivity eliminated in faeces
%Ae(faeces)	amount of total radioactivity eliminated in faeces expressed as a percentage of the radioactive dose administered
CumAe(faeces)	cumulative amount of total radioactivity eliminated in faeces
Cum%Ae(faeces)	cumulative amount of total radioactivity eliminated in faeces expressed as a percentage of the radioactive dose administered
Ae(total)	amount of total radioactivity excreted in urine and faeces combined
%Ae(total)	amount of total radioactivity excreted in urine and faeces combined expressed as a percentage of the radioactive dose administered
CumAe(total)	cumulative amount of total radioactivity excreted in urine and faeces combined
Cum%Ae(total)	cumulative amount of total radioactivity excreted in urine and faeces combined expressed as a percentage of the radioactive dose administered

Pharmacokinetic Assessments:

The plasma concentration data for MD1003, bisnorbiotin and biotin sulfoxide, and plasma and whole blood total radioactivity concentrations provided by Pharmaron will be analysed by Quotient Sciences using appropriate non-compartmental techniques to obtain estimates of the following PK parameters:

Parameter	Definition
Tlag	Time prior to the first measurable (non-zero) concentration
Tmax	Time of maximum observed concentration
Cmax	Maximum observed concentration
AUC(0-last)	Area under the curve from 0 time to the last measurable concentration
AUC(0-inf)	Area under the curve from 0 time extrapolated to infinity
AUCextrap	Percentage of AUC(0–inf) extrapolated beyond the last measurable concentration
T1/2	Apparent elimination half-life
Lambda-z	Slope of the apparent elimination phase
CL/F (MD1003 only)	Apparent total body clearance calculated after a single extravascular administration where F (fraction of dose bioavailable) is unknown
Vz/F (MD1003 only)	Apparent volume of distribution based on the terminal phase calculated after a single extravascular administration where F (fraction of dose absorbed) is unknown
MPR Cmax	Metabolite to parent ratio based on Cmax
MPR AUC(0-inf)	Metabolite to parent ratio based on AUC(0-inf)

Metabolite Profiling and Identification

Metabolite profiling of plasma, urine and faeces will be performed using liquid chromatography-radio-detection with subsequent mass spectrometry where appropriate. Identification of the chemical structure of each metabolite accounting for greater than 10% of circulating radioactivity in plasma (“AUC pool”) and accounting for greater than 10% of the administered dose in urine and faeces (from urine pools and faeces homogenate pools) will be performed. These aspects will be reported separately from the clinical study report as a standalone document.

Safety Assessments:

The safety assessments to be conducted are:

- Adverse Events
- 12-lead ECGs
- Vital Signs
- Clinical laboratory tests (clinical chemistry, haematology and urinalysis)
- Physical examinations

Statistical Methodology:

No formal statistical analysis will be performed for the safety, mass balance or PK data. Descriptive statistics (eg mean, standard deviation, median, minimum, maximum and number of subjects with an observation) are considered adequate for a study of this type.

Sample Size and Power:

The study is exploratory and no formal sample size calculation has been made. Based on experience from previous studies of a similar design, a total of 6 subjects is to be enrolled and a minimum of 4 evaluable subjects is considered sufficient.

4 List of Abbreviations

Abbreviation	Definition
^{14}C	carbon-14
ACC	acetyl-CoA carboxylase
ADME	absorption, metabolism, distribution and elimination
AE	adverse event
ALT	alanine aminotransferase
ARSAC	Administration of Radioactive Substances Advisory Committee
ATP	adenosine triphosphate
ATUc	Autorisation Temporaire d'Utilisation de cohorte
BMI	body mass index
CA	colon arrival
CHMP	Committee for Medicinal Products for Human Use
CNS	central nervous system
CV%	coefficient of variation
DMP	data management plan
EC	ethics committee
ECG	electrocardiogram
eCRF	electronic case report form
EDSS	Expanded Disability Scale Status
EMA	European Medicines Agency
GCP	good clinical practice
GGT	gamma glutamyl transferase
GP	general practitioner
HBsAg	hepatitis B surface antigen
HCV Ab	hepatitis C virus antibody
HIV	human immunodeficiency virus
ICF	informed consent form
ICH	International Council for Harmonisation
IMP	investigational medicinal product
MCC	3-methylcrotonyl-CoA carboxylase
MedDRA	Medical Dictionary for Regulatory Activities
MHRA	Medicines and Healthcare products Regulatory Agency
MS	multiple sclerosis
NMT	not more than

ON	optic neuritis
PC	pyruvate carboxylase
PCC	propionyl-CoA carboxylase
PHE	Public Health England
PIS	Participant Information Sheet
PK	pharmacokinetic(s)
QA	quality assurance
QTcF	Corrected QT interval by Fridericia's formula
RAP	Reporting and Analysis Plan
SAE	serious adverse event
SMVT	sodium multivitamin transporter
SOP	standard operating procedure
SUSAR	suspected unexpected serious adverse reaction
TCA	tricarboxylic acid
TID	three times daily
WHO	World Health Organisation

5 Background Information

5.1 Introduction

MD1003 is the code name for high doses of pharmaceutical-grade biotin being developed by MedDay Pharmaceuticals as a treatment for patients with primary and secondary progressive multiple sclerosis (MS).

MD1003 is a powder of biotin in hard capsule (100 mg per capsule). The therapeutic dose of MD1003 is 300 mg/day (ie between 3 and 6 mg/kg/day) [1].

Biotin is a water-soluble vitamin acting as a carboxyl (CO_2) transporter in carboxylation reactions. In mammals, biotin serves as a cofactor for four carboxylases involved in the metabolism of carbohydrates, amino acids and fatty acids: (1) pyruvate carboxylase (PC), (2) 3-methylcrotonyl-CoA carboxylase (MCC), (3) propionyl-CoA carboxylase (PCC) and (4) acetyl-CoA carboxylase (ACC) [2]. These carboxylases play a critical role in the intermediate metabolism of gluconeogenesis, fatty acid synthesis, and amino acid catabolism

Of note, in the nervous system, ACC1 and ACC2 are detected exclusively in oligodendrocytes, the cells responsible for myelin synthesis, and are found in purified myelin, suggesting that these enzymes are key regulators of myelin synthesis [3].

Besides its role as a co-factor, biotin may regulate genes encoding enzymes or transporters involved in its own metabolism or other cell function [4]. However, this physiological role of biotin remains unclear.

MD1003 is expected to be the first therapeutic approach having an impact on not active progressive MS by reversing the “virtual hypoxia” phenomenon and triggering remyelination [5],[6].

It was hypothesised that the positive effects of MD1003 that are seen in demyelinating diseases such as multiple sclerosis are linked to: (1) increased energy production through feeding the tricarboxylic acid (TCA) cycle in demyelinated neurons and in oligodendrocytes, and (2) stimulation of myelin repair through activation of the acetyl-CoA carboxylase in oligodendrocytes [6].

It is hypothesised that the treatment with MD1003 reverses the state of energy deficit through its role as a cofactor for PC, MCC, and PCC. All 3 of these enzymes are expressed in central nervous system (CNS) cells. These three biotin-dependent carboxylases generate intermediates for the TCA cycle at three different entry points: oxaloacetate, succinate and acetyl-CoA, and could be expected to increase the levels of cellular adenosine triphosphate (ATP). By increasing the available intraneuronal pool of ATP, high-dose biotin may reduce demyelinated neural dysfunction and the adverse effects of hypoxia.

A second biotin-dependent pathway that occurs in the cytoplasm through the biotin-dependent ACC1 and ACC2 may play a role in myelin repair. This pathway is key in the regulation of fatty acid synthesis [2]. ACC1 and ACC2 are key regulators of membrane lipid synthesis including myelin synthesis [3]. In the CNS, ACC1 and ACC2 are mostly expressed in oligodendrocytes [3]. Furthermore, ACC can be detected in purified myelin [3].

As other chronic demyelinating diseases and neuropathies share the pathophysiological mechanisms that are thought to be targeted by MD1003, it is expected that MD1003 would be beneficial in these diseases such as some demyelinating neuropathies and adrenomyeloneuropathy.

5.2 Investigational Medicinal Product

The following investigational medicinal product (IMP) will be used in this clinical study (Table 1).

Table 1 Investigational Medicinal Product

Treatment	IMP Name	Dose	Route of Administration
Single dose	[^{14}C]-MD1003 Capsule	100 mg (NMT 2.22 MBq; 60.0 µCi ^{14}C)	Oral, fasted

^{14}C: carbon 14, NMT: not more than

[^{14}C]-MD1003 is an un-licensed medicinal product for use only in the proposed clinical trial.

Only subjects enrolled in the study may receive study treatment and only authorised site staff may supply or administer study treatment. All study treatments will be stored in a secure, environmentally-controlled, and monitored (manual or automated) area in accordance with the labelled storage conditions with access limited to the investigator and authorised site staff.

Where Quotient Sciences is manufacturing the IMP(s), suitability of the manufacturing process will be documented in a Pharmaceutical Development and Control Strategy Report.

IMPs will be reconciled and destroyed in accordance with the study-specific quality agreement and technical addendum.

5.3 Previous Study Findings

The non-clinical and clinical studies conducted for MD1003 are summarised below; further details are provided in the Investigator's Brochure [1].

5.3.1 Non-clinical Findings

The hypothesised positive effects of MD1003 that may be seen in demyelinating or dysmyelinating diseases such as MS are thought to be linked to: (1) increased energy production through augmenting the TCA cycle in demyelinated neurons, and (2) stimulation of myelin repair through activation of the acetyl-CoA carboxylase in oligodendrocytes.

Following oral administration, biotin is absorbed in the intestine via the sodium multivitamin transporter (SMVT). At the therapeutic dose level, biotin has good oral bioavailability (~50%) and is largely not bound to plasma proteins. Following an oral administration of MD1003 in rats, the highest concentrations were found in the stomach, bladder, and kidneys with a rapid elimination with very low concentrations after 24 h and no storage of biotin observed. Biotin uptake into the CNS, liver, and most peripheral tissues also involves the SMVT. No saturation of the SMVT at the blood-brain barrier is expected at the therapeutic dose. Biotin may accumulate in the placenta and is excreted in milk in animals. Biotin is catabolized mostly in the liver via 2 pathways leading to 2 main metabolites, bisnorbiotin and biotin sulfoxide. Biotin metabolites are inactive. Following an intravenous administration of MD1003 in rats, biotin and its metabolites are rapidly excreted in the urine (>80% of elimination) with biotin being the major form excreted. Urinary clearance data obtained following administration of MD1003 in healthy volunteers suggest that biotin is actively secreted by the kidneys.

Drug interaction with MD1003 treatment involving cytochrome P450 enzymes or transporters commonly involved in the absorption, metabolism and elimination of other drugs are not expected. The expected risk of drug interaction with MD1003 administration in humans resides mainly from pharmacodynamic interaction with insulin and pharmacokinetic (PK) interactions with other substrates of the biotin transporter (SMVT) and some antiepileptic drugs (carbamazepine, oxcarbazepine, primidone, and valproate semisodium).

Based on the safety profiles of MD1003 in rats and dogs following repeated administrations, there is no identified toxicity associated with MD1003 using doses resulting in up to 8-fold the plasmatic exposure corresponding to the MS trial doses of 100 mg orally 3 times daily (TID). MD1003 did not reveal any untoward effect on the CNS, respiratory, cardiovascular, hepatic or renal functions. In several studies, MD1003 did not demonstrate genotoxic potential. This combined with the absence of genotoxicity or carcinogenicity in literature reports suggest that biotin is unlikely to present a risk for carcinogenicity. No carcinogenicity studies have been conducted with MD1003, but a 2-year study in rats and a 6-month study in transgenic rasH2 mice are ongoing. A teratogenic effect of MD1003 was observed in pregnant rabbits at a dose equivalent to twice the exposure in humans in the absence of maternal toxicity. No teratogenic effect was observed in rats. However, the risk of teratogenicity observed in rabbits cannot be excluded in humans at the MD1003 therapeutic dose. Oral doses of MD1003 did not result in epithelial irritation and is not expected to have allergic potential.

5.3.2 Clinical Findings

MD1003 is currently in Phase III of clinical development for the treatment of progressive MS. At the date of the last update of the Investigator's Brochure (cut-off date 18 Jan 2019), a total of 1,145 patients or healthy volunteers had been randomised in 11 studies (ongoing or completed) in the MD1003 clinical program. Of the 1,145 subjects/patients, a total of 493 had been exposed to MD1003 and for 642 patients with progressive MS (Phase III study), treatment groups are still blinded.

In addition, an early access program for MD1003 (ATUc; Autorisation Temporaire d'Utilisation de cohorte) is ongoing in France since July 2016 with MD1003 administered in patients with progressive MS.

A total of 9,661 patients were included in the ATUc between 13 Jul 2016 and 12 Jan 2019 representing 12,241 patients-years of exposure.

A Phase I program with 4 studies assessing the safety and PK profile of MD1003 has been completed. A total of 124 healthy volunteers have been randomised to date. MD1003 daily doses ranged from 100 to 600 mg, with treatment duration of up to 8 days. No deaths, adverse events (AEs) or serious adverse events (SAEs) leading to treatment discontinuation with MD1003 were reported in these 4 healthy volunteer studies.

Two Phase II studies were conducted in which 30 patients with amyotrophic lateral sclerosis and 15 patients with demyelinating neuropathies were included with a daily dose of 300 mg (100 mg TID) MD1003.

A total of 5 Phase IIb-III or Phase III studies are ongoing or completed with MD1003 in Europe and Lebanon at the dose of 300 mg per day (100 mg TID) in patients with progressive MS (830 patients) and adrenomyeloneuropathy (60 patients). Further details can be found in the Investigator's Brochure [1].

5.3.2.1 Safety

Two large randomised, double blind, placebo-controlled Phase IIb-III studies have been conducted in patients with MS; 65 patients in study MS-ON for up to 6 months and 103 patients in study MS-SPI received 100 mg MD1003 TID for up to 12 months. Both studies have open label extension phases which are ongoing. The most commonly reported adverse events (>5% of patients) after treatment with MD1003 in these 2 completed randomised, double-blind, placebo-controlled studies, regardless of assessment of relationship to study treatment were nasopharyngitis, urinary tract infection, worsening of signs and symptoms of MS and MS relapse. The majority of reported adverse events were mild to moderate. Reported SAEs are consistent with the conduct of a trial in an MS patient population.

In completed studies with MD1003, a total of 4 serious adverse events (SAEs), reported in 4 patients and considered by investigators as being related to the study drug were reported. These SAEs included mucocutaneous rash, hypoglycaemia, transient myopathy and laboratory test interference.

In ongoing studies, one SAE (rash maculo-papular) in one patient with amyotrophic lateral sclerosis was considered as related to MD1003 by the investigator.

No deaths, SAEs, or AEs leading to treatment discontinuation with MD1003 were reported in the 4 healthy volunteer studies with MD1003 administered at doses up to 600 mg.

Based on MD1003 clinical and non-clinical data available, as well as the corpus of scientific and medical literature related to biotin, the following safety issues have been identified in patients treated with MD1003: interference with laboratory tests based on a biotin/streptavidin interaction, for which minimisation measures are in place (trial participation card and referral letter to general practitioner/hospital stating the subject has received a high biotin dose if a subject if withdrawn early), and a potential risk of teratogenicity observed in animal studies (rabbits). Since this study involves a single dose of biotin and subjects are planned to be discharged only after the administered dose has been eliminated, risk due to interference with laboratory results is considered

to be very low and will be considered only if subjects are withdrawn early. In the case of early withdrawal, the risk minimisation measures above will be applied In addition, some local allergic skin reactions have been observed. A single case of hypoglycaemia in an insulin-dependent patient and a single case of transient myopathy have also been reported in clinical trials. Adverse drug reactions reported in clinical trials to date are provided in Table 2.

Table 2 Tabulated List of Adverse Reactions

MedDRA System Organ Class	Adverse Reaction (Preferred Term)	Frequency Category
Metabolism and nutrition disorders	Hypoglycaemia[a]	Uncommon[d]
Skin and subcutaneous tissue disorders	Mucocutaneous rash[b]	Uncommon[d]
	Blister[b]	Uncommon[d]
	Eczema[b]	Uncommon[d]
Musculoskeletal and connective tissue disorders	Myopathy[c]	Uncommon[d]

MedDRA: Medical Dictionary for Regulatory Activities
[a] One patient (0.4%) with insulin-dependent diabetes experienced hypoglycaemic episodes, which occurred in the open-label phase of the studies approximately 1 year after initiation of MD1003. Episodes ceased when MD1003 was stopped and recurred on re-challenge.
[b] During treatment with MD1003 in the double-blind placebo-controlled phases of the clinical studies, 1 (0.6%) patient developed a mucocutaneous rash, 1 (0.6%) patient cutaneous blisters and 1 (0.6%) patient eczema. No patients in the placebo group reported these events.
[c] One (0.4%) patient developed transient myopathy, which occurred in the open-label phase of the studies 5 months after initiation of MD1003. The patient fully recovered when MD1003 was stopped.
[d] Uncommon frequency category defined as ≥1/1000 to <1/100.

No serious adverse reactions have been listed as expected so far [1].

The safety profile of MD1003 is favourable, supporting the continuation of the clinical investigations program for the treatment of MS as planned.

5.3.3 Efficacy

An open label pilot study was conducted in MS patients (n=23) using high doses of biotin and demonstrated that biotin could favourably effect disease progression in patients with progressive MS [5].

MS-SPI Trial: The Multiple Sclerosis-Spinal (MS-SPI) study (randomised double-blind study) was conducted to demonstrate the superiority of MD1003 300 mg/day over placebo in patients with progressive MS (n=154). The primary outcome measure was the proportion of patients who demonstrated improvement in a composite measure of Expanded Disability Scale Status (EDSS) and timed 25-foot walk. The primary endpoint was reached with 13 (12.6%) patients treated with MD1003 who achieved clinical improvement in MS-related disability at month 9, confirmed at month 12, compared with none of the placebo-treated patients (p=0.0051). Results of clinician and patient global impression of change measure (CGI/SGI) also showed statistically significant differences in favour of the MD1003 group. The mean change EDSS was stable in MD1003 treated patients while it progressively worsened in placebo active arms and compared to previous published studies (p=0.69) [7]. An open label extension study continues.

MS-ON Trial: The effect of MD1003 in MS patients with permanent visual loss following optic neuritis (ON) was assessed in another controlled clinical study, MS-ON, enrolling 93 patients. Following a 6-month treatment, there was no clinically significant difference between the MD1003 group and the placebo group, with no increase in the mean change of visual acuity in all groups. A positive trend was observed in the progressive optic neuropathy subgroup (n=31), which is consistent with findings of Study MS-SPI, where MD1003 showed efficacy in patients with a progressive form of MS [8].

These results support that targeting neuron/oligodendrocyte metabolism represents a novel disease modifying therapy approach in progressive MS.

Overall, results of the double-blind and extension phases of MS-SPI and MS-ON studies showed a benign safety profile and suggest a durable treatment effect of MD1003 in patients with progressive MS.

A larger international randomised placebo-controlled trial (SPI2) is ongoing in patients with progressive MS.

5.3.4 Pharmacokinetics and Product Metabolism in Animals

Full details of PK and absorption, metabolism, distribution and elimination (ADME) data can be found in the Investigator's Brochure [1].

In the GLP Study MD1003-ADME-RAT, the radioactivity was rapidly eliminated from plasma after intravenous administration of 10 mg/kg ^{14}C-MD1003. After oral administration of 30 mg/kg ^{14}C-MD1003 (human equivalent therapeutic dose), the radioactivity was rapidly absorbed and after a first phase of rapid elimination from the plasma, the radioactivity was then slowly eliminated.

At physiological doses of [^{14}C] biotin (88 nmol/kg body weight) given orally to pigs, the plasma concentration profile best fitted a tri-exponential equation [9], which corresponds to a tricompartmental open model. Disappearance curves of total ^{14}C and of [^{14}C] biotin were similar. Biotin pharmacokinetics after intravenous injection of [^{14}C] biotin was marked by a very short half-life of the initial distribution phase (7 min), followed by a slower plasma elimination phase (T1/2, β of 1.43 h) and a long apparent terminal half-life T1/2, γ estimated at 22 h. The rapid initial distribution phase was shown to correspond to the disappearance of biotin from the plasma (central compartment) to the peripheral compartments, such as the total body water pool or tissue pools [9].

The retention over many days of a substantial portion of the administered dose may occur as a consequence of the metabolic trapping of biotin into cells: conversion of biotin to intermediates such as biotinyl-adenosine monophosphate and biotinyl-CoA, or binding of biotin to proteins such as carboxylases and histones [9]. The half-life of pyruvate carboxylase was determined to be 28 h and 4.6 days for acetyl-CoA carboxylase [10].

Overall, PK data obtained with MD1003 confirmed the literature data and completed the PK properties of biotin:

- Biotin is rapidly absorbed by the oral route in animals

- At the equivalent therapeutic dose in rats (30 mg/kg), biotin is not completely absorbed
- At very high doses (up to 3000 mg/kg), plasma exposure does not significantly increase, suggesting that the transport is saturated
- The major route of elimination is urinary and most of the biotin is excreted unchanged
- There was no evidence of accumulation after once daily repeated dosing in animals

6 Rationale

6.1 Study Rationale

This study is being conducted to investigate the oral PK, mass balance and metabolite profiling and identification of [^{14}C]-MD1003.

6.2 Dose Rationale

The therapeutic daily dose of biotin is 300 mg given as 100 mg TID; therefore, a 100 mg single does is considered the appropriate dose to investigate in an ADME study. This dose is not expected to result in any significant adverse effects in a healthy volunteer population.

The dose of radioactivity has been determined following review of human dosimetry calculations provided by Public Health England (PHE). The associated radiation exposure will fall within International Commission on Radiological Protection (1992) Guidelines for Category IIa studies (0.1 to 1 mSv). The dose of radioactivity will be no more than 2.22 MBq (60.0 µCi).

To ensure that the [^{14}C] drug product(s) does not exceed the limit for radioactive dose approved by the Administration of Radioactive Substances Advisory Committee (ARSAC), the target specific activity of the drug substance will be set at 95% of 90% of the threshold radioactive dosing limit. This takes into account the allowed tolerances during the manufacture of the drug product (± 5% range and drug substance ± 10% range). This provides continued assurance for compliance with the ARSAC-approved limit for drug product radioactivity dose.

6.3 Population Rationale

As this is a Phase I study assessing the mass balance, PK and safety of MD1003, the most relevant population is healthy volunteers. Subjects who are non-smokers without a history of alcohol or drug abuse are proposed to avoid interaction on drug metabolism and to avoid non-compliance.

We acknowledge the ARSAC Notes for Guidance recommend that wherever possible, healthy subjects selected for research projects should be aged over 50 years [11]. However, the current study is designed to generate data for supporting the investigation of the human ADME of a drug, as well as generating samples for metabolite profiling and structural identification.

There are 2 main reasons for generating these data within a clinical development programme. The first is to provide human metabolite data that can be used to interpret the metabolism profiles seen in the preclinical species employed in the longer term toxicity studies, to ensure that there is adequate toxicology coverage for the safe development of the drug in patients. The second is to provide data to understand how the drug is processed in physiologically normal subjects, because understanding the routes of metabolism and elimination in a healthy population generates the appropriate data to guide the clinical pharmacology package required to fulfil the regulatory requirements of a New Drug Application.

In order to address these 2 main aims of an ADME study, investigation of the drug under development is required in a population with normal physiological function, as it is recognised that certain physiological processes eg renal function, deteriorate with age and therefore it is preferable to use as healthy as possible population, to mitigate against factors which may make interpretation of the data difficult. Also, healthy subjects as a trial population are ideal since they have a relatively stable physiological, biochemical and hormonal status, which removes any disease-related variations and variations due to concomitant medications. Therefore given the aims of this ADME study our target age range for this study will be 30 to 65 years.

Based on the above considerations, including the risk of teratogenicity (see Section 5.3.1) in rabbits that cannot be excluded in humans and risks associated with radiation exposure, and to ensure homogeneity of the population, healthy male subjects, aged 30 to 65 years are considered suitable for this study.

6.4 Risks and Benefits

Adverse drug reactions from the MS-SPI and MS-ON studies in MS patients reported in the Investigator's Brochure [1] are listed in Table 2. There are no expected serious adverse reactions for either healthy volunteers or patients administered MD1003. It is unlikely that healthy volunteers administered a single 100 mg dose of MD1003 will experience an adverse reaction. Adverse events will be treated symptomatically, if required.

The dosage form will contain a radionuclide (NMT 2.22 MBq [60.0 µCi] ^{14}C), so subjects will be exposed to ionising radiation. The effective dose that each subject will receive from one administration will not exceed 1 mSv. This is approximately 4.5 months of the average radiation exposure received in the UK each year (2.7 mSv; data obtained from PHE Ionising Radiation Exposure of the UK Population: 2010 Review). This equates to slightly more than the radiation dose that would result from 2 X-rays of the abdomen. It is believed that any increase in the amount of radiation that is received above natural radiation carries a risk of later developing serious and possibly fatal conditions. The risk associated with the maximum possible dose of radiation in this study is very small indeed and is considered to be acceptable.

Collecting a blood sample from a vein may cause pain, swelling, bruising, light-headedness, fainting, and very rarely, clot formation, nerve damage and/or infection at the site of the needle stick.

During cannulation, more than one attempt may be needed to insert the cannula in a vein of a subject and it is possible that bruising and/or inflammation may be experienced at the site of cannulation.

Electrocardiogram stickers on the subjects' chests and limbs may cause some local irritation and may be uncomfortable to remove but subjects will be closely monitored to ensure any local irritation does not persist.

There is no benefit to the subjects from taking part in this study. The development of a product to treat MS will be of benefit to the wider community/patients with this disease.

The overall risk benefit balance is therefore considered to be acceptable.

7 Objectives and Endpoints

Objectives	Endpoints
Primary • To determine the mass balance recovery after a single oral dose of [^{14}C]-MD1003 • To perform metabolite profiling and structural identification of MD1003 metabolites from plasma, urine and faecal samples	• Mass balance recovery of total radioactivity in all excreta (urine and faeces): Ae, %Ae, Cum Ae and Cum %Ae • Metabolite profiling, structural identification, and quantification of [^{14}C]-MD1003 metabolites in plasma, urine and faeces
Secondary • To determine the routes and rates of elimination of [^{14}C]-MD1003 • To identify the chemical structure of each metabolite accounting for more than 10% of circulating total radioactivity • To further explore the oral PK of MD1003 and its metabolites bisnorbiotin and biotin sulfoxide • To evaluate the extent of distribution of total radioactivity into blood cells • To provide additional safety and tolerability information for MD1003	• Amount of total radioactivity excreted and amount of total radioactivity excreted as a percentage of the administered dose in urine and faeces at each time interval • Identification of the chemical structure of each metabolite accounting for more than 10% by AUC of circulating total radioactivity • Assessment of the oral PK profile for MD1003, bisnorbiotin, biotin sulfoxide and total radioactivity based on: Tlag, Tmax, Cmax, AUC(0-last), AUC(0-inf), AUCextrap, T1/2, Lambda-z, CL/F (MD1003 only), Vz/F (MD1003 only), MPR Cmax and MPR AUC(0-inf) • Evaluation of whole blood:plasma concentration ratios for total radioactivity • To provide additional safety and tolerability information for MD1003 by assessing: AEs, vital signs, electrocardiograms (ECGs), physical examinations and laboratory safety

8 Study Design

8.1 Study Plan

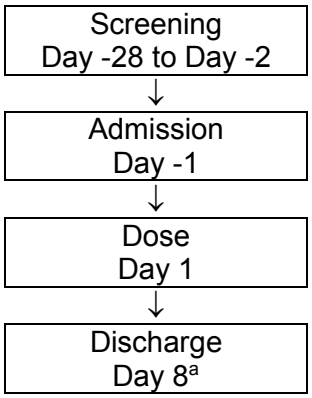
This is a single-centre, open-label, non-randomised, single oral dose study in healthy male subjects. It is planned to enrol 6 subjects to ensure 4 evaluable subjects.

Each subject will receive a single oral administration of a capsule containing 100 mg MD1003 and NMT 2.22 MBq (approximately 60.0 µCi) [^{14}C], in the fasted state.

The study design is presented in Figure 1. Subjects will be screened to participate in the study up to 28 days before dosing. Subjects will be admitted to the clinical unit on the evening of Day -1 prior to IMP administration.

Subjects will be dosed on the morning of Day 1 following an overnight fast and will remain resident in the clinical unit until up to 168 h after dosing (up to Day 8). It is planned that subjects will be released as a group when all subjects have achieved a mass balance cumulative recovery of >90% or if <1% of the dose administered has been collected in urine and faeces within 2 separate consecutive 24 h periods. This may result in the subjects being discharged as a group prior to completion of the planned residency period. Once the discharge criteria or the planned residency period has been achieved, collection of all samples (blood, urine and faeces) will be stopped and the subjects will undergo discharge assessments. If the mass balance criteria have not been met by all subjects by Day 8, the residency period for the subjects not achieving the release criteria may be extended up to a maximum of an additional 48 h (up to Day 10) for further collection of urine and/or faeces. If the release criteria have still not been met by Day 10, or if any additional residency is not considered appropriate or necessary, then home collections of urine and/or faeces may be requested at the discretion of the investigator for individual subjects.

Figure 1 Study Sequence



[a] Subject may be discharged as a group earlier if a cumulative recovery of >90% has been achieved or if <1% of the dose administered has been collected in urine and faeces within 2 separate consecutive 24 h periods. If the criteria are not met by Day 8, this may result in the extension of the residency period for the subjects not achieving the release criteria up to a maximum of an additional 48 h (up to Day 10) for further collection of urine and/or faeces. If the criteria are still not met by Day 10, or if additional residency is not considered appropriate or necessary, then home collections of urine and/or faeces may be requested at the discretion of the investigator for individual subjects.

8.2 Criteria for In-Study Decisions

Not applicable for this study.

8.3 Subject Withdrawal

If a subject wishes to leave the study at any time, they will be permitted to do so. Every reasonable effort will be made by Quotient Sciences to complete a final assessment/discharge procedures. Quotient Sciences will advise the sponsor of the withdrawal of any subject from the study.

Early withdrawal is defined as the date of the decision to withdraw the subject from the study. Subject completion is defined as the date of the last procedure conducted or last contact (ie phone call) for that subject.

If a subject requests to leave the clinical unit earlier than the planned discharge time eg due to unforeseen personal circumstances, but aims to return to the clinical unit to complete the study, this will be documented as a subject self-discharge and a protocol deviation. The subject must complete the planned assessments/discharge procedures before discharge from the clinical unit and will return for the next study period/assessments, as planned.

Subjects will be withdrawn from the study drug(s) for the following reasons:

- Experiencing a serious or severe AE including but not limited to:
    - corrected QT interval by Fridericia's formula (QTcF) of >500 msec or increase in QTcF interval of >60 msec from baseline (confirmed following a repeat ECG)
    - alanine aminotransferase (ALT) concentration >3 × the upper limit of the reference range (confirmed following a repeat ALT blood test)
- Experiencing a 25% decrease in creatinine clearance (CLcr)
- Termination of the study
- Upon the subject's request (withdrawal of consent)
- Significant deviation from the protocol
- Concurrent illness that would compromise subject safety of study data interpretation or requirement for prohibited medication
- At the discretion of the investigator

For the purpose of withdrawal criteria, baseline will be considered as pre-dose Day 1 measurement.

For a subject who withdraws because of an IMP-related AE, every effort will be made to ensure the subject completes follow-up procedures.

Early termination of the study will be distinguished from withdrawal of consent by the subject to participate in any further activities.

8.4 Subject Replacement

No replacement subjects are to be used in this study.

A subject will be considered evaluable if they have provided mass balance and PK samples for up to 72 h (up to Day 4) after IMP administration, or have demonstrated >90% mass balance recovery, or have <1% of the administered dose eliminated in excreta for 2 consecutive days, whichever is sooner.

8.5 Stopping Criteria

The study will be halted, and the risk to other subjects evaluated if any of the following criteria are met:

- A serious adverse reaction (ie a serious AE considered at least possibly related to the IMP administration) in one subject.

- Severe non-serious adverse reactions (ie severe non-serious AE considered as, at least possibly related to the IMP administration) in two subjects in the same cohort, independent of within or not within the same system organ class.

Relatedness will be determined by the investigator.

If the study is halted, a temporary halt will be submitted to the Medicines and Healthcare products Regulatory Agency (MHRA) and ethics committee (EC) in the form of a substantial amendment. The study may be resumed or terminated; however, it will not be resumed until a further substantial amendment to resume the study is submitted and approved by MHRA and EC.

The ARSAC Practitioner will also be informed of the temporary halt.

8.6 Study Termination

After the start of protocol activities but prior to the commencement of dosing, the study may be terminated by the sponsor and investigator without consultation with the MHRA, EC and ARSAC practitioner or ARSAC. The end of the trial must be notified to the MHRA and EC immediately and at the latest within 15 days after the study is terminated, clearly explaining the reasons. A description of any follow-up measures taken for safety reasons if applicable, will also be provided. The ARSAC Practitioner and ARSAC will also be notified within an appropriate timeframe.

If the study is abandoned prior to commencement of any protocol activities, the PI or sponsor must notify the EC, MHRA, ARSAC practitioner and ARSAC (if ARSAC research application has been submitted or approved) in writing outlining the reasons for abandonment of the trial.

Once exposure to ionising radiation has begun, the study will be completed as planned unless the following criteria are satisfied that require a temporary halt or early termination of the study.

- The occurrence of serious or severe adverse event(s), as defined in Section 8.5, if considered to be related to the IMP, as defined in Section 14.2.
- New information regarding the safety of the IMP that indicates a change in the risk/benefit profile for the compound, such that the risk/benefit is no longer acceptable for subjects participating in the study.
- Significant violation of Good Clinical Practice (GCP) that compromises the ability to achieve the primary study objectives or compromises subject safety.

If any of the above occurs, the study will be terminated if careful review of the overall risk/benefit analysis described in Section 6.4 demonstrates that the assumptions have changed and that the overall balance is no longer acceptable. In these circumstances termination can only take place with the agreement of the investigator and sponsor. The MHRA, EC, ARSAC practitioner and ARSAC will be informed of study termination.

If it becomes necessary to consider termination of the study on the dosing day, dosing may be suspended pending discussion between the investigator, sponsor and ARSAC practitioner. Dosing will be stopped immediately on safety grounds.

The study may be terminated or suspended at the request of the MHRA or EC.

8.7 Treatment Allocation

This is an open-label, non-randomised study, therefore a randomisation schedule will not be produced. A treatment allocation will be produced prior to dosing with IMP, which will dictate the order in which the treatments should be administered to each subject. The treatment allocation will be retained in the Investigator Site File.

8.7.1 Subject Numbers

Subject numbers will be allocated on the morning of dosing according to the code 001 to 006 using the lowest number available.

8.7.2 Blinding

This is an open-label, non-randomised study and therefore blinding is not required.

9 Selection of Subjects

Quotient Sciences must have a full medical history from each subject's general practitioner (GP) within the last 12 months, prior to enrolment for the study.

Subjects will be recruited from the Quotient Sciences panel or by direct advertising to the public.

Before subjects are admitted to the clinical unit, The Over Volunteering Prevention System will be checked to ensure that each subject has not participated in a study at another site within at least 90 days of the dosing date.

9.1 Informed Consent

Subjects will be provided with a written explanation of the study at least the day before the screening visit. A physician or nurse will explain to each subject the nature of the study, its purpose, expected duration and the benefits and risks involved in study participation. Subjects will be informed that, for safety reasons, brief details of their involvement in the study may be revealed to other units and companies that carry out clinical studies in the local area. Subjects will then be given the opportunity to ask questions and will be informed of their right to withdraw from the study without prejudice. After this explanation and before entering the study, the subject will voluntarily sign an informed consent form (ICF). Until written consent has been obtained from the subject no study specific procedure or investigation will be performed. If an amendment is made to the participant information sheet (PIS), participants will be re-consented to the most current version of the ICF(s) where appropriate.

9.2 Inclusion Criteria

1. Healthy males
2. Age 30 to 65 years of age at the time of signing informed consent
3. Body mass index (BMI) of 18.0 to 30.0 kg/m^2 as measured at screening
4. Must be willing and able to communicate and participate in the whole study
5. Must have regular bowel movements (ie average stool production of ≥1 and ≤3 stools per day)
6. Must provide written informed consent
7. Must agree to adhere to the contraception requirements defined in Section 9.4

Inclusion criteria 1, 4, 5, 6 and 7 from the list above will be re-assessed at admission/pre-dose.

9.3 Exclusion Criteria

1. Subjects who have received any IMP in a clinical research study within the 90 days prior to Day 1
2. Subjects who are study site employees, or immediate family members of a study site or sponsor employee
3. Subjects who have previously been enrolled in this study
4. History of any drug or alcohol abuse in the past 2 years
5. Regular alcohol consumption in males >21 units per week (1 unit = ½ pint beer, or a 25 mL shot of 40% spirit, 1.5 to 2 Units = 125 mL glass of wine, depending on type)
6. A confirmed positive alcohol breath test at screening or admission
7. Current smokers and those who have smoked within the last 12 months. A confirmed breath carbon monoxide reading of greater than 10 ppm at screening or admission
8. Current users of e-cigarettes and nicotine replacement products and those who have used these products within the last 12 months
9. Subjects with pregnant or lactating partners
10. Radiation exposure, including that from the present study, excluding background radiation but including diagnostic X-rays and other medical exposures, exceeding 5 mSv in the last 12 months or 10 mSv in the last 5 years. No occupationally exposed worker, as defined in the Ionising Radiation Regulations 2017, shall participate in the study
11. Subjects who do not have suitable veins for multiple venepunctures/cannulation as assessed by the investigator or delegate at screening
12. Clinically significant abnormal clinical chemistry, haematology or urinalysis as judged by the investigator (laboratory parameters are listed in Appendix 1). Subjects with Gilbert's Syndrome are allowed
13. Confirmed positive drugs of abuse test result (drugs of abuse tests are listed in Appendix 1)
14. Positive hepatitis B surface antigen (HBsAg), hepatitis C virus antibody (HCV Ab) or human immunodeficiency virus (HIV) results
15. Evidence of renal impairment at screening, as indicated by an estimated creatinine clearance of <80 mL/min using the Cockcroft-Gault equation
16. History of clinically significant cardiovascular, renal, hepatic, chronic respiratory or gastrointestinal disease, neurological or psychiatric disorder, as judged by the investigator
17. Serious adverse reaction or serious hypersensitivity to any drug or the formulation excipients
18. Presence or history of clinically significant allergy requiring treatment, as judged by the investigator. Hay fever is allowed unless it is active
19. Donation or loss of greater than 400 mL of blood within the previous 3 months
20. Subjects who are taking, or have taken, any prescribed or over-the-counter drug (other than 4 g of paracetamol per day), herbal remedies, vitamin B5 or dietary supplements containing lipoic acid in the 14 days before IMP administration (see Section 11.4). Exceptions may apply on a case by case basis, if considered not to interfere with the objectives of the study, as determined by the PI
21. Subjects who have had any intake of biotin (including as a nutritional supplement) in the 14 days before IMP administration
22. Failure to satisfy the investigator of fitness to participate for any other reason

Exclusion criteria 6, 7, 9, 10, 12, 13, 18, 19, 20, 21 and 22 from the list above will be re-assessed at admission/pre-dose.

Healthy subjects who do not meet the inclusion/exclusion criteria for the study will not be enrolled.

9.4 Contraception

Male subjects who are sexually active with a partner of child bearing potential must use, with their partner, a condom plus an approved method of effective contraception from the time of informed consent until 90 days after IMP administration.

The following methods are acceptable:

- Partner's use of combined (oestrogen and progestogen-containing) hormonal contraception associated with inhibition of ovulation:
  - oral
  - intravaginal
  - transdermal
- Partner's use of progestogen-only hormonal contraception:
  - oral
  - injectable/implantable
  - intrauterine hormone-releasing system
- Partner's use of intrauterine device
- Surgical sterilisation (for example, vasectomy or partner's bilateral tubal occlusion)
- Partner's use of female cap or diaphragm or sponge with spermicide (double barrier)

Alternatively, sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study treatments. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the subject.

These contraception requirements are aligned with guidance issued by the Heads of Medicines Agency: Clinical Trials Facilitation Group [12].

9.4.1 Exposure to Sexual Partners During the Study

There is a significant risk of drug exposure through the ejaculate (which also applies to vasectomised males) that might be harmful to sexual partners (both male and female), including pregnant partners of male subjects. Therefore, a condom should be used by all male subjects throughout the study and for 90 days after IMP administration.

9.4.2 Sperm Donation

Male subjects should not donate sperm for the duration of the study and for 90 days after IMP administration.

9.5 Pregnancy

Subjects will be instructed that if their partner becomes pregnant during the study this should be reported to the investigator. The investigator should also be notified of pregnancy occurring during the study but confirmed after completion of the study. In the event that a subject's partner is subsequently found to be pregnant after the subject is included in the study, then consent will be sought from the partner and, if granted, any pregnancy will be followed and the status of mother and/or child will be reported to the sponsor after delivery.

A pregnancy notification form and follow-up will be completed.

9.6 Additional Study Restrictions

The following additional restrictions will be in place for the duration of the study:

1. Subjects must abstain from alcohol during the 24 h prior to screening and the 24 h prior to admission until discharge from the study
2. Subjects must not drink liquids or eat food containing grapefruit, cranberry, caffeine or other xanthines from 24 h prior to admission until discharge from the study
3. Subjects should refrain from eating food containing poppy seeds for 48 h prior to screening and for 48 h prior to admission until discharge from the study
4. Subjects must not take part in any unaccustomed strenuous exercise from the 72 h before the screening visit and then from 72 h prior to admission until discharge from the study

The additional restrictions above are not exclusion criteria; if non-compliance occurs, a protocol deviation will be completed.

10 Study Procedures

Study procedures will be performed as detailed in the study schedule of assessments in Appendix 2, and in accordance with Quotient Sciences standard operating procedures (SOPs) unless otherwise stated in this protocol.

10.1 Screening

Within the 28 days preceding first dose, all subjects will be required to undergo a screening visit. Screening procedures will be carried out in accordance with the study flow chart in Appendix 2.

If the start of the study is delayed for any reason so that the interval between screening and first dose exceeds 28 days, all or part of the screening procedures may be repeated at the discretion of the investigator.

Subjects previously screened generically may participate in this study provided they meet the subject selection criteria. Procedures required by this protocol will only be done if they were not performed during generic screening. All screening data must be obtained within 28 days prior to administration of study medication, as stipulated above.

Screening safety procedures such as safety bloods, ECGs, vital signs, carbon monoxide breath tests, alcohol breath tests and urinalysis can be repeated as clinically indicated under the discretion of the investigator or sub-investigator if there is a concern regarding a subject's safety or eligibility to participate in the trial.

10.1.1 Subject Re-Screening

This study permits the re-screening of a subject who has discontinued the study as a pre-treatment failure (ie subject has not been treated); the reason for failure must be temporary and expected to resolve. If re-screened, the subject must be re-consented.

10.2 Admission and Pre-dose Procedures

The admission and pre-dose procedures are presented in Appendix 2.

The identity of the subjects will be confirmed at admission and pre-dose.

In addition, the ongoing eligibility of subjects will be re-assessed at admission/pre-dose, as described in Sections 9.2 and 9.3.

Admission safety procedures such as safety bloods, ECGs, vital signs, urinalysis and drugs of abuse tests can be repeated as clinically indicated under the discretion of investigator or sub-investigator if there is a concern regarding a subject's safety or eligibility to participate in the clinical trial.

The subjects will be admitted to the clinical unit on the evening before dosing (Day -1).

10.3 Study Day Procedures

10.3.1 Blood Volume

The total blood volume for each subject will not exceed 550 mL in a 4 week period.

The first 0.5 mL of blood withdrawn via cannula will be discarded.

10.3.2 Timing of Procedures

There are times where the protocol requires more than one procedure to be completed at the same time point. In these instances the following will apply to post-dose time points:

PK samples should take priority over other procedures scheduled at the same time point.

As guidance, the preferred order of assessments is:

ECGs should be taken prior to vital signs when both measurements are scheduled at the same time point. Other assessments, eg physical examinations, will be performed within the required time windows.

All safety assessments will be timed and performed relative to the start of dosing.

10.3.3 Discharge from the Clinical Unit

A subject will be allowed to leave the premises without additional investigator or delegate review, following completion of study-specific procedures at discharge, providing that:

- no AEs have been reported during the study visit
- the subject responds in the affirmative when asked if they are feeling well

Subjects will be dosed on the morning of Day 1 following an overnight fast and will remain resident in the clinical unit until up to 168 h after dosing (Day 8). It is planned that subjects will be released as a group when all subjects have achieved a mass balance cumulative recovery of >90% or if <1% of the dose administered has been collected in urine and faeces within 2 separate, consecutive 24 h periods. This may result in the subjects being discharged as a group prior to completion of the planned residency period. Once the discharge criteria or the planned residency period has been achieved, subjects will undergo discharge assessments and collection of all samples (blood, urine and faeces) will be stopped. If mass balance criteria have not been met by all subjects on Day 8, the residency period for the subjects not achieving the release criteria may be extended up to a maximum of 48 h post-dose (Day 10) for further collection of urine and/or faeces. If the criterion is still not met by Day 10, or if additional residency is not considered appropriate or necessary, then home collections of urine and/or faeces may be requested at the discretion of the investigator for individual subjects.

If any of these conditions are not met, then the subject will only be allowed to leave the clinical unit with the authorisation of the investigator or appropriately qualified delegate.

There will be no continued provision of the study intervention or treatment for subjects as this study involves healthy volunteers only.

10.3.4 Medical Supervision

A physician will be responsible for the clinical aspects of the study and will be available at all times during the study. In accordance with the current Association of the British Pharmaceutical Industry guidelines [13], each subject will receive a card stating the telephone number of the investigator and the 24/7 contact details of the Quotient on-call physician.

11 Dosing of Subjects

11.1 Food and Fluid Intake

The calorie/fat content of meals is not required to be controlled during this study, with the exception of meals on Day 1. Meals on subsequent days will be provided at nominal times.

Subjects will be allowed water up to 1 h before the scheduled dosing time and will be provided with 240 mL of water at 1 h post-dose. Water will be allowed ad libitum after 1 h post-dose. Decaffeinated fluids will be allowed ad libitum from lunch time on the day of dosing.

If, for technical reasons, dosing is delayed for more than 2 h beyond the expected dosing time, subjects will receive 200 mL of Lucozade Sport at the originally scheduled dosing time, or earlier if possible.

Subjects will be provided with a light snack and then fast from all food and drink (except water) for a minimum of 8 h the day prior to dosing until approximately 4 h post-dose, at which time lunch will be provided. An evening meal will be provided at approximately 10 h post-dose and an evening snack at approximately 14 h post-dose. On subsequent days, meals will be provided at appropriate times.

If an individual subject has not experienced a bowel movement in any 36-h period post-dose, fluid intake should be increased and administration of a mild laxative (eg prune juice or a mild stool softener) should be implemented.

11.2 Administration of Test Preparations

Specific details of IMP(s) and doses to be administered are provided in Section 5.2 and Section 8.1, respectively. Subjects will be dosed on the morning of Day 1.

The exact time of dosing will be decided based on logistics and will be documented in the source.

Each subject will receive a single oral administration of a capsule containing 100 mg [^{14}C]-MD1003 and NMT 2.22 MBq (60.0 µCi) on a single occasion in the fasted state.

11.3 Dosing Compliance

During all clinical phases of the study, subjects will be observed by study staff to assure compliance to all study procedures, including dose administration.

Mouth and hand checks will be conducted after dosing to ensure the capsule has been swallowed.

The date and time that each subject is dosed will be recorded in the subject's source data. Any violation of compliance will require evaluation by the investigator and sponsor to determine if the subject can continue in the study.

11.4 Prior and Concomitant Medications

No prescribed, over-the-counter medication, herbal remedies, vitamin B5 or dietary supplements containing lipoic acid will be permitted from 14 days before IMP administration until discharge from the study except 4 g per day of paracetamol and those deemed necessary by the investigator to treat AEs (see also Section 9.3). Any medications used will be recorded in the source.

Emergency equipment and drugs will be available within the clinical unit as per current standard procedures. In the unlikely event that they are required, their use will be documented.

12 Assessment of Efficacy

Not applicable for this Phase I study.

13 Assessment of Mass Balance, Pharmacokinetics, Metabolite Profiling and Identification, and Pharmacodynamics

13.1 Assessment of Mass Balance, Pharmacokinetics and Metabolite Profiling and Identification

13.1.1 Blood Sampling

Venous blood samples will be collected from the subjects by a trained member of the clinical team. Consent will be collected from the subjects for use of these samples for the purposes of the proposed study.

Plasma and whole blood samples are sent for laboratory testing in linked anonymised form (subject number only). This information is able to be linked directly to the volunteer by the Quotient research team and study monitor, however not by the laboratory staff or Sponsor.

Venous blood samples will be withdrawn via an indwelling cannula or by venepuncture according to the time schedule presented in Appendix 2.

The acceptable deviations from the nominal blood sampling times are as follows:

- The pre-dose samples will be taken ≤1 h before dosing
- 0 to 1 h post-dose samples will be taken within ± 2 min of the nominal post-dose sampling time
- >1 to 12 h post-dose samples will be taken within ± 10 min of the nominal post-dose sampling time
- >12 h post-dose samples will be taken within ± 30 min of the nominal post-dose sampling time

Samples will be collected into appropriate tubes as specified by the bioanalytical laboratory. Details of sample tubes and processing will be contained in the Clinical Sample Processing Manual.

Samples will be shipped to Pharmaron for the analysis of total radioactivity in plasma and whole blood, and for metabolite profiling and identification in plasma. Samples will be shipped to Atlanbio for the analysis of MD1003, bisnorbiotin and biotin sulfoxide in plasma.

13.1.2 Urine Sampling

Urine samples will be collected according to the time schedule presented in Appendix 2.

A single urine sample will be taken at pre-dose (the first void of the day). Where a sample is not provided, this will not be considered a deviation. All individual urine voids will be collected and shipped to the metabolism laboratory for analysis, according to Quotient Sciences SOPs, unless indicated otherwise by the sponsor.

Samples will be collected into appropriate containers as specified by the metabolism laboratory. Details of sample containers and processing will be contained in the Clinical Sample Processing Manual.

Samples will be shipped to Pharmaron for the analysis of total radioactivity and for metabolite profiling and identification.

13.1.3 Faecal Sampling

Faecal samples will be collected according to the time schedule presented in Appendix 2.

The pre-dose faecal sample will be taken from admission until pre-dose. If a pre-dose faecal sample cannot be obtained, the subject will still be dosed. Where a sample is not provided, this will not be considered a deviation.

Samples and toilet paper will be collected into appropriate pots/containers as specified by the metabolism laboratory. Details of sample containers and processing will be contained in the Clinical Sample Processing Manual.

Samples will be shipped to Pharmaron for the analysis of total radioactivity and for metabolite profiling and identification.

13.1.4 Unexpected Sources of Elimination

During the study, other accidental sources of elimination will be collected as voided (eg emesis).

Samples will be shipped to Pharmaron for the analysis of total radioactivity.

13.2 Assessment of Pharmacodynamics

Not applicable for this Phase I study.

14 Assessment of Safety

14.1 Definition and Classification of Adverse Events

An AE is any untoward medical occurrence in a subject that occurs either before dosing (referred to as a pre-dose AE) or once a medicinal product has been administered, including occurrences which are not necessarily caused by or related to that product.

An adverse drug reaction is any AE where a causal relationship with the IMP is at least a reasonable possibility (possibly related or related).

AEs will be monitored from the time the subject signs the ICF until discharge from the study. The severity of AEs should be assessed as follows:

Mild An AE that is easily tolerated by the subject, causes minimal discomfort and does not interfere with everyday activities

Moderate An AE that is sufficiently discomforting to interfere with normal everyday activities; intervention may be needed

Severe An AE that prevents normal everyday activities; treatment or other intervention usually needed

14.2 Assessment of Causality

Every effort should be made by the investigator to try to explain each AE and assess its relationship, if any, to the IMP. The temporal relationship of the event to IMP administration should be considered in the causality assessment (ie if the event starts soon after IMP administration and resolves when the IMP is stopped).

Causality should be assessed using the following categories:

Unrelated: Clinical event with an incompatible time relationship to IMP administration, and that could be explained by underlying disease or other drugs or chemicals or is incontrovertibly not related to the IMP

Possibly related: Clinical event with a reasonable time relationship to IMP administration, and that is unlikely to be attributed to concurrent disease or other drugs or chemicals

Related: Clinical event with plausible time relationship to IMP administration and that cannot be explained by concurrent disease or other drugs or chemicals

The degree of certainty with which an AE is attributed to IMP administration (or alternative causes, eg natural history of the underlying disease, concomitant therapy) will be determined by how well the experience can be understood in terms of one or more of the following:

- known pharmacology of the IMP
- reactions of a similar nature have been previously observed with the IMP or this class of drug
- the experience being related by time to IMP administration, terminating with IMP withdrawal or recurring on re-challenge
- alternative cause

14.3 Recording Adverse Events

AEs will be recorded from the time of providing written informed consent until discharge from the study. During each study visit the subject will be questioned directly regarding the occurrence of any adverse medical event according to the schedule in the source. All AEs, whether ascribed to study procedures or not, will be documented immediately in the source. This will include the date and time of onset, a description of the AE, severity, duration, actions taken, outcome and an investigator's current opinion on the relationship between the study drug and the event. A diagnosis and final opinion on the relationship between the study drug and the event will be provided at the end of the study by the investigator.

Any subject who withdraws from the study due to an AE will be followed up until the outcome is determined and written reports provided by the investigator.

14.4 Serious Adverse Events

14.4.1 Definition of Serious Adverse Events

A serious adverse event (SAE) is defined as any untoward medical occurrence that at any dose:

- results in death
- is life-threatening
- requires hospitalisation or prolongation of existing hospitalisation
- results in persistent or significant disability or incapacity
- consists of a congenital anomaly or birth defect
- an important medical event as recognised by the PI

SAEs must be immediately (without any delay) reported to the sponsor and in any case no later than 24 h of becoming aware of the event.

14.4.2 Definition of Suspected Unexpected Serious Adverse Reactions

Suspected unexpected serious adverse reactions (SUSARs) are AEs that are believed to be related to an IMP and are both unexpected (ie the nature or severity is not expected from the information provided in the Reference Safety Information (RSI) section of the Investigator's Brochure Edition No 10) and serious. SUSARs are subject to expedited reporting to the MHRA, European Medicines Agency (EMA) and EC (see Section 16.3.2 for details on reporting SUSARs).

14.5 Laboratory Measurements

Venous blood, urine and faecal samples will be collected from the subjects by a trained member of the clinical team. Consent will be collected from the subjects for use of these samples for the purposes of the proposed study.

Blood, urine and faecal samples are sent for laboratory testing in linked anonymised form (subject number, initials and the subjects' gender and date of birth for analytical reasons). This information is able to be linked directly to the volunteer by the Quotient research team and study monitor, however not by the laboratory staff or sponsor.

Safety laboratory tests and virology will be carried out on blood samples, and drugs of abuse tests and urinalysis will be carried out on urine samples. The research will not involve analysis or use of human DNA.

Blood and urine samples results will be reviewed by a physician and acted upon before the subject is dosed or receives their next dose, or is released from the study, as is appropriate. A list of the laboratory parameters measured is presented in Appendix 1.

14.5.1 Haematology and Clinical Chemistry

Laboratory tests will be performed by The Doctors Laboratory according to the time schedule presented in Appendix 2. Blood samples will be collected and processed as detailed in the Clinical Sample Processing Manual. Scheduled blood samples will be taken following an 8 h fast.

The acceptable deviations from the nominal blood sampling time points for laboratory assessments are:

- The pre-dose blood sample will be taken ≤2 h before dosing
- Post-dose blood samples will be taken ± 1 h from the nominal blood sampling time

Creatinine clearance (CLcr) will be calculated at screening by The Doctors Laboratory using the Cockcroft-Gault equation and body weight for eligibility purposes:

$$\text{CLcr (mL/min)} = \frac{(140\text{-age [years]}) \times (\text{body weight [kg]}) \ (\times 1.23)}{\text{serum creatinine (µmol/L)}}$$

14.5.2 Urinalysis

Urinalysis will be performed on-site using a dipstick according to the time schedule presented in Appendix 2. Urine samples will be collected and processed as detailed in the Clinical Sample Processing Manual. If microscopy is required, a urine sample will be sent to The Doctors Laboratory. Any urine used for urinalysis will be added back to the full sample for total radioactivity analysis. If microscopy is required, a urine sample (approximately 20 mL) will be taken from the urine collection, and sent to The Doctors Laboratory after the void has been weighed.

The acceptable deviations from the nominal urine sampling time points for urinalysis are:

- The pre-dose urine sample will be taken ≤3 h before dosing or the first void of the day
- Post-dose urine samples will be taken ± 2 h from the nominal urine sampling time

14.5.3 Drug Screen

A urine drug screen will be performed on-site using a dipstick method according to the time schedule presented in Appendix 2. The sample will be collected and processed as detailed in the Clinical Sample Processing Manual. Subjects will be screened for the drugs of abuse listed in Appendix 1.

14.5.4 Alcohol Breath Test

An alcohol breath test will be performed according to the time schedule presented in Appendix 2. A positive result will exclude the subject from dosing during that admission.

14.5.5 Carbon Monoxide Breath Test

A carbon monoxide breath test will be performed according to the time schedule presented in Appendix 2. A result of greater than 10 ppm will exclude the subject from the study.

14.5.6 Abnormal Laboratory Findings

In cases where laboratory findings are outside the normal range and the investigator believes that the results may be of clinical significance, repeat sampling may be requested as clinically indicated. If the abnormal finding is clinically significant, appropriate actions will be taken eg the subject will not be entered into the study or the subject may be withdrawn from the study. The subject will be referred to their GP or other appropriate provider for further care. The same will apply if the results of the HBsAg, HCV Ab or HIV test are positive and in addition the investigator will ensure that adequate counselling is available if requested.

Abnormal results at follow-up assessments will also require repeat testing if the investigator believes the results may be of clinical significance.

Any clinically significant abnormality, including changes from baseline, must be reported as an AE.

Additional blood and/or urine samples may be taken for safety tests. Furthermore, additional assays outside those specified in the protocol may be performed for safety reasons as requested by the investigator or sub-investigator.

14.6 Vital Signs Measurements

Blood pressure and heart rate will be measured by an automated recorder after the subject has been in a supine position for a minimum of 5 min according to the time schedule presented in Appendix 2. Oral temperature will also be measured. The acceptable deviations from the nominal vital signs measurement time points are:

- The pre-dose vital signs measurements will be taken ≤2 h before dosing.
- Post-dose vital signs measurements will be taken ± 15 min from the nominal post-dose time points.
- Discharge vital signs measurements will be taken ± 1 h from the nominal time point.

If a subject shows an abnormal assessment at any stage, repeat measurements may be made and the abnormality followed to resolution if required. Additional measurements may be taken as deemed necessary by the investigator or sub-investigator.

Any clinically significant abnormality, including changes from baseline, must be reported as an AE.

14.7 ECG Measurements

Single 12-lead ECGs will be measured after the subject has been in the supine position for a minimum of 5 min according to the time schedule presented in Appendix 2. The acceptable deviations from the nominal ECG measurement time points are:

- The pre-dose ECG measurements will be taken ≤2 h before dosing
- Post-dose ECG measurements will be taken ± 15 min from the nominal post-dose time point.
- Discharge ECG measurements will be taken ± 1 h from the nominal time point.

If a subject shows an abnormal assessment at any stage, repeat measurements may be made and the abnormality followed to resolution if required. Additional measurements may be taken as deemed necessary by the investigator or sub-investigator.

Any clinically significant abnormality, including changes from baseline, will be reported as an AE.

14.8 Body Weight and Height

The subject's body weight and height will be measured as detailed in Appendix 2.

14.9 Physical Examination

Subjects will undergo a physical examination as detailed in Appendix 2.

14.10 Additional Safety Procedures

Additional non-invasive procedures that are already specified in the protocol may be performed, if it is believed that an important effect of the IMP(s) is occurring or may occur at a time when no measurements are scheduled, or if extra procedures are needed in the interests of safety.

Additional blood samples for safety assessments may be taken if required by the investigator at any point.

15 Statistics and Data Analysis

15.1 Sample Size Justification

The study is exploratory and no formal sample size calculation has been made. Based on experience from previous studies of a similar design, a total of 6 subjects is to be enrolled and a minimum of 4 evaluable subjects is considered sufficient.

15.2 Data Management

Data management will be performed by Quotient Sciences.

Study data will be managed using a validated electronic case report form (eCRF) database system and subjected to data consistency and validation checks. Data queries will be raised within the study eCRF database by data management staff and resolved with the assistance of clinical staff.

AEs and medications will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) (v22.0 or a more recent version) and the World Health Organisation (WHO) Drug Dictionary Global Drug Reference (2019 or a more recent version), respectively. An independent coding review will also be performed within the Data Sciences department.

Clinical chemistry and haematology data (and other safety laboratory data) will be collected by a central laboratory (The Doctors Laboratory) and stored electronically in their clinical pathology system. The data will be transferred electronically to Quotient Sciences and all demographic details and sample dates will be cross-referenced with the corresponding data on the study database. All queries will be resolved with the assistance of laboratory staff, or if necessary, clinical staff.

The database will be closed after all queries have been resolved. The database will be locked when all criteria listed in the Data Management Plan (DMP) are met.

Further details are addressed in the DMP.

15.3 Mass Balance Data Analysis

Urine and faeces will be collected for the analysis of total radioactivity. Graphical and tabular representation of whole blood and plasma total radioactivity concentrations and ratios at selected time points will be presented. The following mass balance parameters will be calculated by Quotient Sciences for urine, faeces and combined urine and faeces (ie total radioactivity from all excreta).

Parameter	Definition
Ae(urine)	Amount of total radioactivity excreted in urine
%Ae(urine)	Amount of total radioactivity excreted in urine expressed as a percentage of the radioactive dose administered
CumAe(urine)	Cumulative amount of total radioactivity excreted in urine
Cum%Ae(urine)	Cumulative amount of total radioactivity excreted in urine expressed as a percentage of the radioactive dose administered
Ae(faeces)	Amount of total radioactivity eliminated in faeces
%Ae(faeces)	Amount of total radioactivity eliminated in faeces expressed as a percentage of the radioactive dose administered
CumAe(faeces)	Cumulative amount of total radioactivity eliminated in faeces
Cum%Ae(faeces)	Cumulative amount of total radioactivity eliminated in faeces expressed as a percentage of the radioactive dose administered
Ae(total)	Amount of total radioactivity excreted in urine and faeces combined
%Ae(total)	Amount of total radioactivity excreted in urine and faeces combined expressed as a percentage of the radioactive dose administered
CumAe(total)	Cumulative amount of total radioactivity excreted in urine and faeces combined
Cum%Ae(total)	Cumulative amount of total radioactivity excreted in urine and faeces combined expressed as a percentage of the radioactive dose administered

15.4 Pharmacokinetic Data Analysis

The plasma concentration data for MD1003, bisnorbiotin and biotin sulfoxide, and plasma and whole blood total radioactivity concentrations provided by Pharmaron will be analysed by Quotient Sciences using Phoenix WinNonlin v8.0 or a more recent version (Certara USA, Inc., USA).

Calculated MD1003 concentrations may be corrected for specific activity of the administered radiolabelled oral dose by the Data Sciences department at Quotient Sciences.

Plasma concentration data will be tabulated and plotted for each subject for whom concentrations are quantifiable. PK analysis of the concentration time data obtained will be performed using appropriate non-compartmental techniques to obtain estimates of the following PK parameters:

Parameter	Definition
Tlag	Time prior to the first measurable (non-zero) concentration
Tmax	Time of maximum observed concentration
Cmax	Maximum observed concentration
AUC(0-last)	Area under the curve from 0 time to the last measurable concentration
AUC(0-inf)	Area under the curve from 0 time extrapolated to infinity
AUCextrap	Percentage of AUC(0–inf) extrapolated beyond the last measurable concentration
T1/2	Apparent elimination half-life
Lambda-z	Slope of the apparent elimination phase
CL/F (MD1003 only)	Apparent total body clearance calculated after a single extravascular administration where F (fraction of dose bioavailable) is unknown
Vz/F (MD1003 only)	Apparent volume of distribution based on the terminal phase calculated after a single extravascular administration where F (fraction of dose absorbed) is unknown
MPR Cmax	Metabolite to parent ratio based on Cmax
MPR AUC(0-inf)	Metabolite to parent ratio based on AUC(0-inf)

Further details of the PK data analysis will be included in the Reporting and Analysis Plan (RAP).

15.5 Metabolite Profiling and Identification

Metabolite profiling of plasma, urine and faeces will be performed using liquid chromatography-radio-detection with subsequent mass spectrometry where appropriate.

Identification of the chemical structure of each metabolite accounting for greater than 10% of circulating radioactivity in plasma (“AUC pool”) and accounting for greater than 10% of the administered dose in urine and faeces (from urine pools and faeces homogenate pools) will be performed.

These aspects will be reported separately from the clinical study report as a standalone document.

15.6 Statistical Data Analysis

Production of summary tables, figures and listings for this study will be performed by Quotient Sciences using the statistical package SAS (v9.4 or more recent version).

No formal statistical analysis will be performed for this study. Descriptive statistics (eg mean, median, standard deviation, minimum, maximum and number of subjects with an observation [n]) are considered adequate for a study of this type. Additional statistics will be provided for PK-related data, including coefficient of variation (CV%), geometric mean, geometric CV% and geometric n (ie number of subjects with an observation that are included in the natural logarithmic transformation).

Populations and analysis sets will be determined for safety, mass balance and PK data after database lock using the criteria defined in the RAP; the RAP will be signed off prior to database lock.

Further details relating to the statistical analysis will be included in the RAP including the following:

- Criteria to be used to define each of the populations and analysis sets
- Additional detail covering the analyses and/or description of primary and secondary analyses and safety data
- Handling of missing data, unused or spurious data
- Handling of data from withdrawn subjects

All safety, mass balance recovery and PK data will be listed.

15.7 Interim Analysis

No formal interim analyses are planned for this study.

16 Safety Reporting to Ethics Committees and Regulatory Authorities

16.1 Events Requiring Expedited Reporting

SUSARs (Section 14.4.2) are subject to expedited reporting to the MHRA, EMA and EC.

In addition to SUSARs, other safety issues may qualify for expedited reporting where they might materially alter the current benefit-risk assessment of an IMP or that would be sufficient to consider changes in the IMPs administration or in the overall conduct of the study, for instance:

- an increase in the rate of occurrence or a qualitative change of an expected serious adverse reaction, which is judged to be clinically important
- SAEs that occur after the subject has completed the clinical study where the sponsor considers them to be a SUSAR
- new events related to the conduct of the study or the development of the IMPs and likely to affect the safety of the subjects, such as:
    - an SAE which could be associated with the study procedures and which could modify the conduct of the study
    - a major safety finding from a newly completed animal study (such as carcinogenicity)
    - any anticipated end or temporary halt of a study for safety reasons and conducted with the same IMPs in another country by the same sponsor

16.2 Urgent Safety Measures

If Quotient Sciences or any of its staff or contractors becomes aware of an actual or potential urgent safety issue, then the sponsor must be immediately contacted so that appropriate urgent safety measures can be agreed. An urgent safety issue is defined as:

- An immediate hazard to the health or safety of subjects participating in a clinical study
- A serious risk to human health or potentially a serious risk to human health

An urgent safety issue may include issues with an investigational drug or comparators, study procedures, inter-current illness (including pandemic infections), concomitant medications, concurrent medical conditions or any other issues related to the safe conduct of the study or that pose a risk to study subjects.

In exceptional circumstances of imminent hazard and in order to safeguard the health or safety of individuals, Quotient Sciences may take urgent safety measures before informing the sponsor, but the sponsor must be informed immediately after the hazard has resolved.

Quotient Sciences will take responsibility for informing appropriate competent authorities, and the EC.

16.3 Reporting

16.3.1 Reporting Serious Adverse Events

The investigator is required to notify the study sponsor if appropriate immediately and not later than 24 h of becoming aware of the occurrence of an SAE or serious adverse reaction. A copy of the written report of the event should promptly be sent to the study sponsor for information purposes, in accordance with International Council for Harmonisation (ICH) guidelines for GCP.

16.3.2 Reporting of Suspected Unexpected Serious Adverse Reactions (SUSARs)

It is the responsibility of the sponsor to determine whether a reported SAE fits the classification of a SUSAR and to notify the investigator of their decision as soon as possible.

16.3.3 Expedited Reporting of Events

It is the responsibility of the sponsor to determine whether an event requires expedited reporting and to notify the investigator of their decision as soon as possible.

Where expedited reporting is required, the following procedures should be followed.

Fatal or life-threatening SUSARs

It is the responsibility of the sponsor to report fatal or life-threatening SUSARs to the MHRA and EMA as soon as possible, but no later than 7 calendar days after they first became aware of the reaction. Any additional relevant information should be sent within 8 days of the report. This responsibility may be delegated to the pharmacovigilance provider.

The investigator is required to notify the EC of any SUSAR as soon as possible, but no later than 7 calendar days after they first became aware of the reaction. Any additional relevant information should be sent within 8 days of the report.

The ARSAC Practitioner will be notified of any SUSAR that is considered related to the exposure to radioactivity.

Other SUSARs

It is the responsibility of the sponsor to report other SUSARs to the MHRA and EMA as soon as possible, but no later than 15 calendar days after they first became aware of the reaction. This responsibility may be delegated to the pharmacovigilance provider.

The investigator is required to notify the EC of other SUSARs as soon as possible, but no later than 15 calendar days after they first became aware of the reaction.

The ARSAC Practitioner will be notified of any SUSAR that is considered related to the exposure to radioactivity

16.3.4 Reporting of Urgent Safety Issues

Quotient Sciences is required to inform the appropriate competent authorities and the EC within 3 calendar days of the urgent safety issue.

16.4 Serious Breaches

It is the responsibility of the sponsor to notify the licensing authority of any serious breach, which is likely to affect, to a significant degree, the safety or mental integrity of the subjects of the study or the scientific value of the study.

All serious breaches will be notified to the MHRA within 7 days. The reporting will be performed by the party who suspects the serious breach.

17 Protocol Amendments and Deviations

17.1 Amendments

After the protocol has been submitted to the MHRA and/or EC, any amendment must be agreed by the investigator after discussion with the sponsor and will be formally documented.

All substantial amendments will be submitted to the MHRA and/or EC for an opinion as required by current regulations. Any amendments relating to the administration of radioactive substances will be reviewed by the ARSAC practitioner prior to submission to ARSAC as required by the current ARSAC Notes for Guidance. The ARSAC practitioner will also be notified of any substantial amendments to the PIS and ICF and/or protocol.

If the PIS and ICF are updated as a result of the substantial amendment, the new approved versions will be used to re-consent currently enrolled subjects and must be provided to additional/replacement subjects prior to their entry into the study.

17.2 Protocol Deviations

The study must be conducted in accordance with the Clinical Protocol. Should a protocol deviation occur, it must be promptly assessed in order to decide whether any of these non-compliances should be reported to the MHRA as a serious breach of GCP and the Clinical Protocol.

Protocol waivers are not acceptable.

Deviations from the protocol will be recorded in the source as noted by the clinical staff. If necessary, the sponsor will be informed of the deviation.

Any protocol deviations assessed as major will be discussed with the sponsor in order to determine if the withdrawal criteria stated in Section 8.3 have been met.

18 Regulatory

18.1 Compliance

This study will be conducted in accordance with the protocol and with the following legislation:

- International Council for Harmonisation Good Clinical Practice (GCP) Guidelines approved by the Committee for Medicinal Products for Human Use (CHMP) on 17 Jul 1996, which came into force on 17 Jan 1997, updated Jul 2002, Integrated Addendum E6 (R2) dated 09 Nov 2016 [14]
- The Medicines for Human Use (Clinical Trials) Regulations. Statutory Instruments 2004 No. 1031 [15]

- The Medicines for Human Use (Clinical Trials) Amendment Regulations. Statutory Instruments 2006 No. 1928 [16]
- The Medicines for Human Use (Clinical Trials) Amendment (No. 2) Regulations. Statutory Instruments 2006 No. 2984 [17]
- The Medicines for Human Use (Clinical Trials) Amendment Regulations. Statutory Instruments 2008 No. 941 [18]
- Health and Safety. The Ionising Radiations Regulations 2017. Statutory Instrument 2017 No. 1075 [19]
- Health and Safety. Ionising Radiation (Medical Exposure) Regulations 2017. Statutory Instrument 2017 No. 1322 [20]

In addition, the study will be performed according to the ethical principles outlined in the World Medical Association Declaration of Helsinki and its amendments [21].

18.2 Ethical Approval

Prior to the initiation of the study, the protocol and associated documentation must be given a favourable opinion by an EC. A copy of this written approval and any correspondence with the EC will be provided to the sponsor.

18.3 MHRA Approval

Prior to the initiation of the study, the Clinical Trial Authorisation application must be approved by the MHRA. A copy of this approval and any correspondence with the MHRA will be available at the clinical and sponsor sites. A copy of the MHRA approval will be provided to the EC.

18.4 Administration of Radiation

Dr Stuart Mair will be the ARSAC practitioner for this study, which includes the administration of radiation at Quotient Sciences. Administration will be conducted in accordance with Dr Mair's current ARSAC practitioner licence and Quotient's current ARSAC Employer licence. Additionally a research application will be submitted to ARSAC to obtain approval for the conduct of the study before dosing.

Before submitting to the ARSAC, a summary of available nonclinical tissue distribution and excretion information on [^{14}C]-MD1003 will be submitted to the Radiation Protection Division of PHE for human dosimetry calculations in order to facilitate the selection of the dose of radioactivity to be administered. The final report from the PHE will be included in the application to the ARSAC.

The protocol will be reviewed and the final version will be approved by the ARSAC practitioner, Dr Stuart Mair.

18.5 Source Data

A study-specific source document identification list will be finalised with the sponsor prior to the start of the clinical phase of the study. The document will identify what data should be considered source data for this study.

For this study, electronic data capture will be used where possible and data will be automatically recorded into an eCRF. In instances where paper source documents are used, data to be transcribed into the eCRF will be identified using a Source Document Identification List, as governed by Quotient Sciences SOPs.

18.6 Declaration of the End of the Study

The end of the study is defined as the last visit of the last subject (eg discharge). Any changes to this definition will be notified as a substantial amendment (see Section 17.1).

The EC and MHRA should be notified in writing of the conclusion of the study within 90 days of the end of the study, or within 15 days if the study is terminated early, clearly explaining the reasons for the termination.

ARSAC and the ARSAC Practitioner will also be notified of the end of trial or early termination of the trial in writing within an appropriate timeframe.

18.7 Document Storage and Archiving

All documentation and correspondence pertaining to the study (source data, raw data, letters etc) will be kept in accordance with the ICH guidelines for Good Clinical Practice 1996, updated 2002, Integrated Addendum E6 (R2) dated 09 Nov 2016 (ICH GCP Section 4.9.5) [14], The Medicines for Human Use (Clinical Trials) Regulations 2004 [15] and The Medicines for Human Use (Clinical Trials) Amendment Regulations 2006 [16],[17].

All study related documents will be retained for a minimum period of 25 years. After this time, the sponsor will be contacted to ascertain whether continued storage or destruction is required in accordance with current regulations.

18.8 Protection of Personal Data and Confidentiality

Personal data are securely stored to prevent unauthorised access, disclosure, dissemination, alteration or loss of information and unauthorised personal data processing. Access to personal information is restricted so that only personnel who are required to access personal data as part of their job role can do so. All personnel who access personal information are bound by a duty of confidentiality.

Technical arrangements surrounding the electronic storage and use of data are as follows:

- Computers storing electronic personal data are protected by antivirus software and the network on which computers are linked are protected by industry grade firewalls
- Off-site personnel can only access networked computers through a virtual private network
- Electronic access of data is limited according to user roles
- All data are stored on password protected computers

Organisational arrangements are as follows:

- All buildings are secured by key-card access
  Manual files of personal data are stored within locked cabinets that can only be accessed by authorised personnel
- Data security and/or confidentiality provisions are utilised in agreements with third parties
- Documented Back-up and disaster recovery procedures are in place
- Internal audit and compliance functions provide regulatory oversight

The personal data of volunteers will be pseudonymised in that they will only include health, initials, date of birth and demographics (gender and ethnicity) and cannot be linked back to the individual by the recipient. The sponsor shall be the data controller in respect of the personal data of the study subjects collected in connection with the study, and shall act in accordance with the relevant data protection laws in relation to the collection and processing of those personal data. The study subjects' pseudonymised personal data shall be collected and processed for the purposes of the study and may also be added to research databases and used in the future by the sponsor and its affiliates for certain additional clinical research, for product regulation and safety reporting purposes and for ensuring compliance with legal requirements. The study subjects' pseudonymised personal data may be processed for such purposes by other parties including: the sponsor's affiliates and licensing partners, its business partners, regulatory agencies and other health authorities, and ECs. The study subjects' authorisation for such use and disclosure shall be obtained by the study subjects signing the ICF for the study.

Additionally, Quotient personnel are contractually bound by a duty of confidentiality and receive training in this matter.

18.9 Data Security Breach

Quotient has a comprehensive process in place for identifying, assessing, resolving and reporting any potential data security breach. All staff are trained in the identification of potential data security breaches. Potential breaches are managed by appropriately trained quality assurance personnel in accordance with Quotient Sciences standard operating procedures. After robust assessment of data breaches, those deemed serious will be reported to the sponsor and Information Commissioner's Office, as applicable.

19 Quality Control and Quality Assurance

Quality control of all data collected from this study will be performed in accordance with Quotient SOPs. This study (or elements thereof) may be subject to Quotient quality assurance (QA) audit, in line with current internal auditing procedures. Similarly, the study (or elements thereof) may be subject to sponsor QA audit.

19.1 Monitoring

GCP requires that studies are adequately monitored. The sponsor should determine the appropriate extent and nature of monitoring. A study monitor, independent of Quotient Sciences, will be appointed to verify that the study is conducted in accordance with current GCP, regulatory requirements, the protocol and that the data are authentic, accurate and complete.

The investigator agrees to receive visits from a study monitor and provide assistance to verify protocol implementation, source completion and transcription of data into the eCRF, document storage and AE reporting.

Quotient Sciences will extend the professional privilege of access to the subjects' clinical source documents to the study monitor, EC, regulatory bodies or other authorised personnel (eg auditor) for the purposes of source data verification.

Following completion of the study both study related documents and subject data may be sent to the sponsor at a location outside of the UK where data protection laws differ. In the interests of confidentiality, subjects will not be identified on any such documents or data, and specific subject consent for such a disposition will be obtained.

20 Finance and Insurance

The sponsor (MedDay Pharmaceuticals) has funded this study. A no-fault clinical trials insurance has been obtained by the sponsor. The sponsor insurance will compensate subjects in accordance with the Association of the British Pharmaceutical Industry Guidelines for Phase I Clinical Trials 2018 edition [13].

21 Publication

Please refer to the Master Services Agreement for information on publication.

Quotient Sciences shall have the right to publish the results of the research, subject to the sponsor's prior written consent, which shall not be unreasonably withheld or delayed. Following the receipt of such consent, Quotient Sciences shall submit a copy of the proposed publication to the sponsor who shall have 30 days in which to request amendments thereto which, to the extent that such proposed amendments are reasonable, Quotient shall be obliged to incorporate prior to such publication.

The sponsor undertakes that, prior to publication of any information, article, paper, report or other material concerning the research, it will submit a copy of such publication to Quotient Sciences who shall have 90 days in which to request amendments thereto which, to the extent that such proposed amendment are reasonable, the sponsor shall be obliged to incorporate prior to such publication.

22 References

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Appendix 1 Clinical Laboratory Parameters

Haematology	Clinical Chemistry	Virology	Urinalysis	Drugs of Abuse
Basophils Eosinophils Haematocrit (Packed Cell Volume- PCV) Haemoglobin Lymphocytes Mean Cell Haemoglobin (MCH) Mean Cell Haemoglobin Concentration (MCHC) Mean Cell Volume (MCV) Monocytes Neutrophils Platelet Count Red Blood Cell (RBC) Count White Blood Cell (WBC) Count	Alanine Aminotransferase (ALT) Albumin Alkaline Phosphatase Aspartate Aminotransferase (AST) Bicarbonate Bilirubin (Total) Bilirubin (Direct) (only if Total is elevated) Calcium Chloride Creatine Kinase (CK) Creatinine[a] Gamma Glutamyl Transferase (GGT) Glucose Glucose (Fasting) Potassium Phosphate (Inorganic) Protein (Total) Sodium Urea	Hepatitis B Surface Antigen Hepatitis C Antibody HIV Antibody	Bilirubin Blood Glucose Ketones Leukocytes Nitrites pH Protein Specific gravity Urobilinogen **At discretion of investigator based on urinalysis results** Microbiology Urine Microscopy	Amphetamines Barbiturates Benzodiazepines Cocaine Marijuana/Cannabis Methadone Methamphetamine/ Ecstasy Morphine/Opiates Phencyclidine Tricyclic Antidepressants

[a] Creatinine clearance will be calculated at screening using the Cockcroft-Gault equation for eligibility purposes

Appendix 2 Study Flow Chart

Study Day	-28 to -2	-1	1											2		3	4	5	6	7	8
				Times After Dosing (h)																	
	Screening	Admission	Pre-dose	0	0.5	1	1.5	2	3	4	6	8	12	18	24	36	48	72	96	120	144	168[a]
General Assessments																						
Informed Consent	X																				
Medical History	X	X																			
Weight and Height	X																				
Vein Assessment	X																				
Carbon Monoxide Breath Test	X	X																			
Drug Screen	X	X																			
Alcohol Breath Test	X	X																			
IMP Administration				X																	
Safety Assessments																					
Physical Examination	X													X[b]							X[b]
Safety Labs[c]	X		X											X							X
Urinalysis	X		X											X							X
ECG	X		X			X				X				X							X
Vital Signs[d]	X		X			X				X				X			X				X
Adverse Events	←																				→
Prior and Concomitant Medication	←																				→
Mass Balance and PK Assessments																					
Plasma Samples for MD1003, bisnorbiotin and biotin sulfoxide			X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Plasma Samples for TR			X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Whole Blood Samples for TR			X			X				X		X	X		X		X	X	X			X
Plasma Samples for Metabolite Profiling and ID			X			X		X		X		X	X								
Urine Samples for TR and Metabolite Profiling and Identification[e]			←																		→
Faecal Samples for TR and Metabolite Profiling and Identification[f]			←																		→

ECG: Electrocardiogram; ID: Identification; IMP: Investigational Medicinal Product; TR: total radioactivity

[a] Discharge from clinical unit; subjects may be discharged earlier if a cumulative mass balance recovery of 90% % has been achieved or if <1% of the dose administered has been collected in urine and faeces within 2 separate consecutive 24 h periods. If the criteria are not met by Day 8, this may result in the extension of the residency period for the subjects not achieving the release criteria up to a maximum of an additional 48 h (up to Day 10) for further collection of urine and/or faeces. If the criteria are still not met by Day 10, or if additional residency is not considered appropriate or necessary, then home collections of urine and/or faeces may be requested at the discretion of the investigator for individual subjects

[b] Targeted (symptom driven) physical examination

[c] Haematology and clinical chemistry at each time point including virology at screening (see Appendix 1 for a full list of clinical laboratory parameters). Creatinine clearance will be estimated at screening from plasma creatinine using the Cockcroft-Gault equation for eligibility purposes

[d] Blood pressure and heart rate. Oral temperature will be measured at screening, pre-dose and any unscheduled assessments as required

[e] A single urine sample will be collected at pre-dose (the first void of the day) and then at the following collection periods: 0 to 12 h, 12 to 24 h, and then daily (24 h intervals) until discharge

[f] Faeces will be collected pre-dose (sample to be taken between admission and pre-dose) and then daily (24 h intervals) until discharge