Effect of immuno-nutrition on systemic inflammation in people receiving haemodialysis: a pilot study

Version 2.0

25 February 2022

Short title:	Immuno-nutrition in haemodialysis
Acronym:	
IRAS Project ID:	309937
Study Sponsor:	University of Nottingham
Sponsor reference:	21085
Funding Source:	Internal funds

STUDY PERSONNEL AND CONTACT DETAILS

Sponsor: University of Nottingham
Ms Angela Shone
Research and Innovation
University of Nottingham, East Atrium, Jubilee Conference Centre
Triumph Road
Nottingham
NG8 1DH
Email: sponsor@nottingham.ac.uk

Chief investigator: Professor Maarten Taal
Professor of Medicine and Honorary Consultant Nephrologist
Academic Unit for Translational Medical Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 224618
Fax: 01332 789352
Email: m.taal@nottingham.ac.uk

Co-investigators: Dr Bethan E Phillips
Associate Professor of Translational Physiology
Academic Unit of Injury, Inflammation and Recovery Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 724687
Email: beth.phillips@nottingham.ac.uk

Dr Daniela Viramontes Hörner
Renal Research Fellow
Academic Unit for Translational Medical Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 788262
Email: mszdv@nottingham.ac.uk

Professor Philip Atherton
Professor of Clinical, Metabolic and Molecular Physiology
Academic Unit of Injury, Inflammation and Recovery Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 724 725
Email: philip.atherton@nottingham.ac.uk

Professor Kenneth Smith
Professor of Metabolic Mass Spectrometry
Academic Unit of Injury, Inflammation and Recovery Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 724700
Email: ken.smith@nottingham.ac.uk

Professor Nicholas Selby
Professor of Nephrology and Honorary Consultant Nephrologist
Academic Unit for Translational Medical Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 788262
Email: nicholas.selby@nottingham.ac.uk

Dr Daniel Wilkinson
Assistant Professor of Metabolic Mass Spectrometry
Academic Unit of Injury, Inflammation and Recovery Sciences, School of Medicine, University of Nottingham, Royal Derby Hospital
Uttoxeter Rd, Derby, DE22 3DT
Phone: 01332 724850
Email: daniel.wilkinson@nottingham.ac.uk

Study Statistician: Dr Daniel Wilkinson

Study Coordinating Centre: Academic Unit for Translational Medical Sciences, School of Medicine, University of Nottingham

SYNOPSIS

Title	Effect of immuno-nutrition on systemic inflammation in people receiving haemodialysis: a pilot study.
Acronym	
Short title	Immuno-nutrition in haemodialysis
Chief Investigator	Professor Maarten Taal
Objectives	1. To investigate whether provision of an immuno-nutrition supplement decreases systemic inflammatory status in people on haemodialysis. 2. To investigate the effect of an immuno-nutrition supplement on biochemical blood variables, body composition, dietary intake and muscle strength in people on haemodialysis.
Study Configuration	Single-centre, non-randomised, interventional pilot study.
Setting	Secondary care
Sample size estimate	As a pilot study power calculations are not appropriate
Number of participants	15
Eligibility criteria	*Inclusion criteria:* • CRP level >5 mg/L. • Age: ≥18 years (no upper age limit). • At least three haemodialysis sessions per week for ≥3 hours using a biocompatible dialyser. • Able to give informed consent. *Exclusion criteria:* • Treatment with drugs that cause immunosuppression. • Non-English speakers or those with special communication needs. • Pregnancy, breast feeding or intending pregnancy. • Expected survival <6 months. • Hospitalisation at the time of screening. • Known intolerance or allergy to oral nutritional supplement (or isolated ingredients). • Pre-dialysis serum potassium >5.0 mmol/L. • Unable to provide informed consent.

Description of interventions	Dietitian supervised nutritional support with an immuno-nutrition supplement (Oral Impact®, Nestle)
Duration of study	Duration of individual study participation: 6 weeks. Overall study duration: 3 months.
Methods of analysis	To compare baseline versus final evaluations, a Wilcoxon test or paired t-test will be used in the case of dimensional variables, and McNemar test in the case of categorical variables. Inter-group comparisons will be performed using a Mann-Whitney test or Student t-test for continuous variables and χ^2 test or Fisher's exact test for categorical variables. A p value <0.05 will be considered to have statistical significance.

ABBREVIATIONS

AE	Adverse Event
BMI	Body Mass Index
CI	Chief Investigator
CRF	Case Report Form
CRP	C Reactive Protein
GCP	Good Clinical Practice
HGS	Handgrip strength
NHS	National Health Service
ONS	Oral Nutritional Supplement
PIS	Participant Information Sheet
REC	Research Ethics Committee
R&D	Research and Development department
SAE	Serious Adverse Event
SAF	Skin Autofluorescence
TMF	Trial Master File

TABLE OF CONTENTS

STUDY BACKGROUND INFORMATION AND RATIONALE

Low skeletal muscle mass is a highly prevalent problem of kidney disease occurring in 25-55% of people receiving haemodialysis treatment [1]. Moreover, loss of skeletal muscle mass is one of the strongest predictors of frailty, disability and increased mortality, severely affects health-related quality of life, and increases hospital admissions and length of hospitalisations in this patient population, which results in higher hospital costs [2].

Multiple factors contribute to skeletal muscle loss in those receiving haemodialysis, including:

• reduced appetite and therefore dietary intake [3-6], resulting in lack of dietary amino acids - a key anabolic stimulus [7],
• muscle tissue catabolism during haemodialysis [8, 9], reflected by increased urea production [10] and amino acid efflux from muscle [11],
• physical inactivity, anxiety and depression [12, 13], and the
• presence of other co-morbidities associated with muscle wasting, such as diabetes and heart disease [12].

In addition, increased systemic inflammation, a frequent complication in the haemodialysis population, has been postulated to be a primary driver of skeletal muscle loss [12, 14]. To exemplify the magnitude of this problem, it has been reported that people on haemodialysis lose an average of 6.4 kg of lean tissue mass over 20 weeks, with associated significant reductions in muscle strength. In addition, and key to the research proposed herein, systemic inflammation was shown to markedly accelerate these muscle mass losses [15].

Provision of dietetic advice and high-energy/high-protein conventional oral nutritional supplements (ONS) to people on haemodialysis has previously shown to improve postabsorptive plasma amino acid profiles, body mass index (BMI), and serum albumin and prealbumin levels [16, 17]. Despite this, conventional ONS have not been proven successful in stimulating muscle protein synthesis, and consequently in increasing skeletal muscle mass [18]. Therefore, achieving improvements in skeletal muscle mass in the haemodialysis population is challenging. In part, this appears to relate to non-adherence [18, 19]. Our previous experience, based on both research studies and clinical practice, is that the taste of some conventional ONS (often described as too sweet) is commonly cited by people on haemodialysis as a reason for non-adherence to prescribed supplementation regimes [19]. In addition to the challenge of long-term adherence to conventional ONS, the persistent systemic inflammation seen in people receiving haemodialysis plays a major role in causing “anabolic resistance”, a condition that inhibits improvements in skeletal muscle mass despite an increase in dietary energy and protein intake/availability [20].

Immuno-nutrition supplements are high in energy and protein (similar to conventional ONS), but also contain a unique combination of nutrients (i.e., omega-3 fatty acids, L-arginine and nucleotides), each acting in a complementary way to suppress inflammation, while concomitantly enhancing immune function [21]. Immuno-nutrition supplements (specifically Oral Impact®, Nestle) have been mostly used in pre-surgical settings, and previous studies conducted in cancer populations have reported a decrease in post-operative infections and length of hospitalisation, underpinned by evidence of a decrease in systemic inflammation [22].

This will be the first study to explore the potential effect of an immuno-nutrition supplement (Oral Impact®, Nestle) to address systemic inflammation in people receiving haemodialysis. The findings of this pilot study will help to design a randomised controlled clinical trial investigating whether a treatment bundle of resistance exercise training and immuno-nutrition supplementation can improve skeletal muscle mass, other markers of nutritional status and quality of life in people on haemodialysis.

STUDY OBJECTIVES AND PURPOSE

PURPOSE

The purpose of the present study is to investigate the impact of an immuno-nutrition supplement on systemic inflammation in people receiving haemodialysis.

PRIMARY OBJECTIVE

To investigate whether provision of an immuno-nutrition supplement decreases systemic inflammatory status in people on haemodialysis.

SECONDARY OBJECTIVES

To investigate the effect of an immuno-nutrition supplement on biochemical blood variables, body composition, dietary intake, skin autofluorescence (SAF) and muscle strength in people on haemodialysis.

DETAILS OF PRODUCT(S)

Description

Oral Impact® is a powdered oral nutritional supplement that contains a unique combination of ingredients with immuno-modulating properties, namely omega-3 fatty acids, arginine and nucleotides, as well as soluble fibre.

Manufacture

Oral impact® is manufactured by Nestlé Health Science, Reg. Trademark of Société des Produits Nestlé S.A. OIDC 001

Packaging and labelling

Oral impact® comes in a box that contains 5 sachets x 74g. Please see Appendix 1 for images of packaging and labelling.

Storage, dispensing and return

Oral impact® will be stored unopened in a locked room which should be cool, dry and away from light and humidity. Access to the room will be restricted to members of the research team.

Known Side Effects

No known side effects. However, Oral impact® is unsuitable for people with cow's milk protein allergy and those who are severely septic. Oral impact® should not be mixed with medication and food.

STUDY DESIGN

This will be a single-centre, non-randomised, interventional pilot study where people on haemodialysis will receive an immuno-nutrition supplement (Oral Impact®, Nestle) for 6 weeks.

STUDY CONFIGURATION

Primary outcome

Change in C reactive protein (CRP), and pro- and anti-inflammatory cytokine levels (via multiplex plasma analysis) after 6 weeks of treatment with an immuno-nutrition supplement.

Secondary endpoints

Change after 6 weeks of treatment with an immuno-nutrition supplement in:

- Biochemical blood variables including haemoglobin, urea, creatinine, potassium, phosphate, calcium, sodium, albumin, total proteins, total cholesterol and triglycerides.
- Body composition assessed with bioelectrical impedance analysis and BMI.
- Dietary intake assessed with 3-day food diaries.
- Muscle strength assessed with handgrip strength (HGS).
- SAF levels.

Safety endpoints

- AEs that occur will be reported during the study.
- Participant reported tolerability of the immuno-nutrition supplement.
- Monitoring of pre-dialysis serum potassium levels.

STUDY MANAGEMENT

The Chief Investigator (CI) has overall responsibility for the study and shall oversee all study management. The study will be co-ordinated from the University of Nottingham campus at the Royal Derby Hospital. Data will be collected at the Renal Dialysis Unit of the Department of Renal Medicine at the Royal Derby Hospital (University Hospitals of Derby and Burton NHS Foundation Trust) and stored at the University of Nottingham.

The CI and Co-investigators are responsible for study planning, recruitment, consenting, data collection, handling and interpretation. The data custodian will be the CI, Professor Taal.

DURATION OF THE STUDY AND PARTICIPANT INVOLVEMENT

Participant recruitment will begin in January 2022 once all necessary approvals are in place, with an anticipated total recruitment and intervention delivery period of 3 months (i.e., end of March 2022). The study will end when the last participant completes their final study visit.

SELECTION AND WITHDRAWAL OF PARTICIPANTS

Recruitment

15 participants will be recruited from the Renal Dialysis Unit at the Royal Derby Hospital. Potential participants will be approached by the clinical care team (which may include members of the research team) with reference to the inclusion and exclusion criteria. Each eligible participant will be given a Participant Information Sheet (PIS) and a minimum of 24 hours to consider whether they wish to participate in the study. At this time, the PIS will be discussed with them, any questions answered, and informed consent will be obtained by one of the investigators if they are willing to enter the study.

It will be explained to the potential participant that entry into the study is entirely voluntary and that their treatment and care will not be affected by their decision. It will also be explained that they can withdraw at any time. In the event of their withdrawal, it will be explained that their data collected so far cannot be erased and we will seek consent to use the data in the final analyses where appropriate.

If needed, the hospital interpreter and translator services will be available to assist with discussion of the study, the PIS, consent form, and be available for the duration of the study if required. The consent form and PIS will not be available printed in other languages.

Eligibility criteria

Inclusion criteria:

- CRP level >5.0 mg/L.
- Age: ≥18 years (no upper age limit).
- At least three haemodialysis sessions per week for ≥3 hours using a biocompatible dialyser.
- Able to give informed consent.

Exclusion criteria:

- Treatment with drugs that cause immunosuppression.
- Non-English speakers or those with special communication needs.
- Pregnancy, breast feeding or intending pregnancy.
- Expected survival <6 months.
- Hospitalisation at the time of screening.
- Known intolerance or allergy to ONS (or isolated ingredients).
- Pre-dialysis serum potassium >5.0 mmol/L.
- Unable to provide informed consent.

Participant Withdrawal

Participants will be withdrawn from the study if they:

- Withdraw their consent at any time.
- Become pregnant.
- Receive a kidney transplant.
- Do not adhere to the intervention; defined as <50% of the provided immuno-nutrition supplement being consumed.

Participants do not have to provide a reason for withdrawal of consent. Participants will also be withdrawn if it is deemed unsafe or is found to be impossible to proceed in the opinion of the investigators.

Participants may be withdrawn from the study either at their own request or at the discretion of the Investigator. The participants will be made aware that this will not affect their future care. Participants will be made aware (via the PIS and consent form) that should they withdraw the data collected to date cannot be erased and may still be used in the final analysis.

Participants will not be replaced if they withdraw from the study.

Informed consent

The process for obtaining participant informed consent will be in accordance with the Research Ethics Committee (REC) guidance, Good Clinical Practice (GCP) and any other regulatory requirements that might be introduced. The investigator or their nominee and the participant shall both sign and date the consent form before the person can participate in the study.

The participant will receive a copy of the signed and dated consent form and the original will be retained in the Study records. A second copy will be filed in the participant's medical notes and a signed and dated note made in the notes that informed consent was obtained for the study. The decision regarding participation in the study is entirely voluntary. The investigator or their nominee shall emphasize to them that consent regarding study participation may be withdrawn at any time without penalty or affecting the quality or quantity of their future medical care, or loss of benefits to which the participant is otherwise entitled. No study-specific procedures will be done before informed consent has been obtained.

The investigator will inform the participant of any relevant information that becomes available during the course of the study and will discuss with them whether they wish to continue with the study. If applicable they will be asked to sign revised consent forms.

If the consent form is amended during the study, the investigator shall follow all applicable regulatory requirements pertaining to approval of the amended consent form by the REC and use of the amended form (including for ongoing participants).

STUDY REGIMEN

Once participants have given their informed consent, they will be advised to take **daily** one sachet (74 g) of the immuno-nutrition supplement (Oral Impact®, Nestle) dissolved in 125 ml of water and will be followed-up for 6 weeks.

Participants will receive individualised (to patient needs and food preferences) dietetic advice formulated and delivered by an experienced renal dietitian under an honorary NHS research contract (D.V.H), who is a member of the research team and will be supervised by an NHS renal dietitian, aiming to achieve estimated nutritional requirements for people on haemodialysis (i.e., energy: 30-35 kcal/kg/day, and protein intake: 1.0-1.2 g/kg/day) [23]. Each dietetic advice provided to participants will also be reviewed by the NHS clinical renal dietitians who are members of the participants' usual clinical care team.

Consumption of the immuno-nutrition supplement will be supervised during haemodialysis sessions (thrice weekly) and self-reported (written and counting empty sachets) on non-dialysis days. Participants will receive precise oral instructions on how and when to take the supplement with close monitoring by the renal dietitian on the research team to encourage adherence to the intervention.

Three-day food diaries (i.e., one dialysis day, one non-dialysis day and one weekend day) will be completed by the participants at baseline, 3 and 6 weeks of follow-up, and will be analysed (Microdiet, Downlee Systems Ltd, UK) to assess compliance with the advice provided and to assess energy, protein and fat intake across the study period.

<u>Data collection procedures</u>

Hospital electronic medical records will be used to collect relevant baseline participant characteristics, including age, sex, ethnicity, time since dialysis initiation, dialysis adequacy,

current medication, presence of diabetes and history of cardiovascular disease. Information regarding educational level, occupation status, and history of smoking will be obtained by direct interview with the participants at the time of the baseline assessment. A blood sample will be taken weekly pre-dialysis after commencing the immuno-nutrition supplement to monitor serum potassium. Results from routine clinical blood tests will be recorded at baseline and 6 weeks of follow-up and will include haemoglobin, urea, creatinine, potassium, phosphate, calcium, sodium, albumin, total proteins, total cholesterol, triglycerides and CRP.

Pre- and post-dialysis blood samples (10 mL) will be taken at baseline and 6 weeks of follow-up and centrifuged at 2000g for 20-min to obtain at least 2 mL of plasma which will be frozen and stored at -80°C for subsequent analysis of circulating systemic pro- and anti-inflammatory cytokines via the Luminex multiplex platform.

Handgrip strength (HGS) will be measured at baseline and 6 weeks of follow-up using the Takei 5401 handgrip digital dynamometer (Takei Scientific Instruments Co., Ltd., Tokyo, Japan) within the first hour of haemodialysis treatment. The highest value of 3 attempts in the non-fistula arm will be recorded as HGS.

Body composition will be assessed via bioelectrical impedance analysis (InBody 770, InBody, Leicester, UK) and BMI at baseline and 6 weeks of follow-up.

Skin autofluorescence (SAF) will be measured using a validated Autofluorescence Reader (AGE Reader, DiagnOptics, Groningen, The Netherlands) at baseline and 6 weeks of follow-up. For each participant, three measurements will be performed on the volar surface of the lower arm at ~10 cm below the elbow, ensuring that the area has normal skin without visible vessels, scars or other abnormalities. Only the non-fistula arm will be used, and readings will be undertaken within the first hour of haemodialysis treatment.

Criteria for terminating trial

In the event of major safety concerns, new information, or issues with study conduct (e.g., poor recruitment, loss of resources), termination of the study will be considered by the sponsor. In the event of study termination, unused study resources will be reallocated as appropriate.

TRANSPORT AND STORAGE OF THE TISSUES

Pre- and post-dialysis blood samples (10 mL) will be collected at intervals as described above and centrifuged. At least 2 mL of plasma will be stored at -80°C for future analysis at the University of Nottingham Centre of Metabolism, Ageing & Physiology (COMAP).

Samples and data will be stored in a linked anonymised format. Samples will be labelled using a combination of study reference, unique study identifier and cross referenced with location code numbers to permit accurate linkage to study data and the consent form.

The master database will be held by the CI in a password encrypted file.

Where participants do not agree to the future use of the samples, they will be destroyed in accordance with the Human Tissue Act, 2004.

LABORATORY ANALYSES

Blood samples will be taken at the Renal Dialysis Unit and transferred to the local NHS laboratory (which is serviced and managed according to national UKAS standards and governance frameworks) at the Royal Derby Hospital for routine biochemistry and

haematology testing. These samples will be labelled according to NHS requirements. No DNA or RNA extraction will occur as part of this study.

STATISTICS

Statistical analyses will be performed using SPSS version 28.0. Data will be presented as mean ± standard deviation, median (interquartile range), or percentages, as appropriate. To compare baseline versus final evaluations, a Wilcoxon test or paired t-test will be used in the case of dimensional variables, and McNemar test in the case of categorical variables. Intergroup comparisons will be performed using Mann-Whitney test or Student t-test for continuous variables and χ^2 test or Fisher's exact test for categorical variables. An alpha error of less than 0.05 will be judged to be significant. All data will be analysed on University of Nottingham computers and backed up regularly.

Sample size and justification

A formal power calculation is not appropriate for this pilot study design, and consequently, the sample size has been selected on a pragmatic basis. We will aim to recruit 15 participants in total.

Procedures for missing, unused and spurious data

All missing data will be explained. If a space on the Case Report Form (CRF) is left blank because the procedure was not done or a question was not asked, "N/D" will be recorded. If the item is not applicable to the individual case, "N/A" will be recorded. All entries will be printed legibly in black ink. If any error is made in data entry, to correct the error, a single straight line will be drawn through the incorrect entry and the correct data entered above it. All such changes will be initialled and dated. Errors will not be erased or altered by any other method. For clarification of illegible or uncertain entries, the clarification will be printed above the item, initialled and dated. Correction fluid will not be used.

Definition of populations analysed

The study will analyse the following populations:

- Safety set: All participants who underwent at least one study session.
- Full Analysis set: All participants, who participated in at least one study session and for whom at least one post-baseline assessment of the primary endpoint is available.
- Per protocol set: All participants in the Full Analysis set who are deemed to have no major protocol violations that could interfere with the objectives of the study.

ADVERSE EVENTS

Definitions

An adverse event (AE) is any unfavourable and unintended sign, symptom, syndrome or illness that develops or worsens during the period of observation in the study.

An AE does include a/an:

1. Exacerbation of a pre-existing illness.
2. Increase in frequency or intensity of a pre-existing episodic event or condition.
3. Continuous persistent disease or symptoms present at baseline that worsen following the start of the study.

The following are recognised complications related to chronic haemodialysis and will be recorded on the CRF, but do not need to be reported separately on an AE/Serious Adverse Event (SAE) form:

- Related to dialysis: fistula bruising, dizziness, cramps, hypotension, minor bleeding.
- Hospital admission for vascular access, acute coronary syndromes, arrhythmias, stroke, pneumonia and infections.
- Death.

An AE does not include a/an:

1. Medical or surgical procedure (e.g., surgery, endoscopy, tooth extraction, transfusion); however, the condition that led to the procedure is an AE.

2. Pre-existing disease or conditions present or detected at the start of the study that did not worsen.

3. Situations where an untoward medical occurrence has not occurred (e.g., hospitalisations for cosmetic elective surgery, social and/or convenience admissions).

4. Disease or disorder being studied, or sign or symptom associated with the disease, or disorder unless more severe than expected for the participant's condition.

5. Overdose of concurrent medication without any signs or symptoms.

A SAE is any AE occurring following study mandated procedures, directly related to the treatment or intervention that results in any of the following outcomes:

1. Death.
2. A life-threatening AE.
3. Inpatient hospitalisation or prolongation of existing hospitalisation.
4. A disability/incapacity.
5. A congenital anomaly in the offspring of a participant.

Important medical events that may not result in death, be life-threatening, or require hospitalisation may be considered a SAE when, based upon appropriate medical judgment, they may jeopardize the patient or participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

All AEs will be assessed for seriousness, expectedness and causality:

A distinction is drawn between serious and severe AEs. Severity is a measure of intensity, whereas seriousness is defined using the criteria above. Hence, a severe AE need not necessarily be serious.

Causality

Not related or improbable: a clinical event including laboratory test abnormality with temporal relationship to study intervention administration which makes a causal relationship incompatible or for which other treatments, chemicals or disease provide a plausible explanation. This will be counted as "unrelated" for notification purposes.

Possible: a clinical event, including laboratory test abnormality, with temporal relationship to study treatment/intervention administration which makes a causal relationship a reasonable possibility, but which could also be explained by other interventions, chemicals or concurrent disease. This will be counted as “related” for notification purposes.

Probable: a clinical event, including laboratory test abnormality, with temporal relationship to study treatment/intervention administration which makes a causal relationship a reasonable possibility, and is unlikely to be due to other interventions, chemicals or concurrent disease. This will be counted as “related” for notification purposes.

Definite: a clinical event, including laboratory test abnormality, with temporal relationship to study treatment/intervention administration which makes a causal relationship a reasonable possibility, and which can definitely not be attributed to other causes. This will be counted as “related” for notification purposes.

With regard to the criteria above, medical and scientific judgment shall be used in deciding whether prompt reporting is appropriate in that situation.

Reporting of adverse events

Participants will be asked to contact the study site immediately in the event of any SAE. All AEs will be recorded and closely monitored until resolution, stabilisation, or until it has been shown that the study intervention is not the cause. The CI shall be informed immediately of any SAEs and shall determine seriousness and causality in conjunction with any treating medical practitioners.

In the event of a pregnancy occurring in a study participant or the partner of a study participant, monitoring shall occur during the pregnancy and after delivery to ascertain any study related AEs in the mother or the offspring. Where it is the partner of a study participant, consent will be obtained for this observation from both the partner and her medical practitioner.

All intervention related SAEs will be recorded and reported to the REC as part of the annual reports. Unexpected SAEs will be reported within the timeframes to the REC as stated below. The CI shall be responsible for all AEs reporting.

Study Intervention Related SAEs

A SAE that is unexpected in its severity and seriousness and deemed directly related to or suspected to be related to the study intervention shall be reported to the REC that gave a favourable opinion as stated below.

The event shall be reported immediately of knowledge of its occurrence to the CI.

The CI will:

- Assess the event for seriousness, expectedness and relatedness to the study intervention.
- Take appropriate medical action, which may include halting the study and inform the Sponsor of such action.

- If the event is deemed related to the study treatment or intervention, shall inform the REC using the reporting form found on the HRA web page within 7 days of knowledge of the event.
- Shall, within a further eight days, send any follow-up information and reports to the REC.
- Make any amendments as required to the study protocol and inform the REC as required.

Participant removal from the study due to adverse events

Any participant who experiences an AE may be withdrawn from the study at the discretion of the Investigator. The overall risk of an AE directly connected to the study is low.

ETHICS COMMITTEE AND REGULATORY APPROVALS

The study will not be initiated before the protocol, consent form and PIS have received approval/favourable opinion from the REC, and the respective NHS Research & Development (R&D) department. Should a protocol amendment be made that requires REC approval, the changes in the protocol will not be instituted until the amendment and revised informed consent form and PIS have been reviewed and received approval/favourable opinion from the REC and R&D department. A protocol amendment intended to eliminate an apparent immediate hazard to participants may be implemented immediately providing that the REC is notified as soon as possible, and an approval is requested. Minor protocol amendments only for logistical or administrative changes may be implemented immediately, and the REC will be informed.

The study will be conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki, 1996, the principles of GCP, and the UK Policy Framework for Health and Social Care Research 2017.

RECORDS

Case Report Forms

Case Report Forms (CRFs) will be the primary data collection instrument. All data requested on the CRF will be recorded.

Each participant will be assigned a study identity code number, for use on CRFs, other study documents and the electronic database.

CRFs will be treated as confidential documents and held securely in accordance with regulations. The investigator will make a separate confidential record of the participant's name, date of birth, local hospital number or NHS number, and Participant Study Number, to permit identification of all participants enrolled in the study, in case additional follow-up is required. This shall be kept securely in the Trial Master File (TMF).

CRFs shall be restricted to those personnel approved by the Chief or local Investigator and recorded as such in the study records.

All paper forms shall be filled in using black ballpoint pen. Errors shall be lined out but not obliterated by using correction fluid and the correction inserted, initialled and dated.

The Chief or local Investigator shall sign a declaration ensuring accuracy of data recorded in the CRF. Electronic data will identify participants by study ID only. Records will be kept password protected and subject to regular back up procedures.

Sample Labelling

Each participant will be assigned a study identity code for use on the CRF, other study documents and the electronic database. Samples for NHS pathology analysis will be labelled in accordance with local NHS procedures.

Source documents

The source documents for each participant will consist of the demographic, co-morbidities and concomitant medication data collected from the participant's health care records. This will be transferred into a CRF for that particular participant. Paper copies of haematological and biochemical laboratory test results from the electronic results system will be retained in the TMF as source documents.

Source documents shall be filed at the investigator's site and may include but are not limited to consent forms, study records. A CRF may also completely serve as its own source data. Only study staff shall have access to study documentation other than the regulatory requirements listed below.

Direct access to source data / documents

The CRF and all source documents, including progress notes and copies of laboratory and medical test results shall made be available at all times for review by the CI, Sponsor's designee and inspection by relevant regulatory authorities (e.g., Health Research Authority, Human Tissue Authority).

DATA PROTECTION

All study staff and investigators will endeavour to protect the rights of the study's participants to privacy and informed consent and will adhere to the Data Protection Act 2018. The CRF will only collect the minimum required information for the purposes of the study. CRFs will be held securely, in a locked room, or locked cupboard or cabinet. Access to the information will be limited to the study staff and investigators and any relevant regulatory authorities (see above). Computer held data, including the study database, will be held securely and password protected. All data will be stored on secure UoN systems. Access will be restricted by user identifiers and passwords (encrypted using a one-way encryption method).

Information about the study in the participant's medical records/hospital notes will be treated confidentially in the same way as all other confidential medical information.

Electronic data will be backed-up every 24 hours to both local and remote media in encrypted format.

QUALITY ASSURANCE & AUDIT

INSURANCE AND INDEMNITY

Insurance and indemnity for clinical study participants and study staff is covered within the NHS Indemnity Arrangements for clinical negligence claims in the NHS, issued under cover

of HSG (96)48. There are no special compensation arrangements, but study participants may have recourse through the NHS complaints procedures.

The University of Nottingham as research Sponsor indemnifies its staff with both public liability insurance and clinical trials insurance in respect of claims made by research subjects.

STUDY CONDUCT

Study conduct may be subject to systems audit for inclusion of essential documents; permissions to conduct the study; CVs of study staff and training received; local document control procedures; consent procedures and recruitment logs; adherence to procedures defined in the protocol (e.g., inclusion / exclusion criteria, timeliness of visits); accountability of study materials and equipment calibration logs.

STUDY DATA

Monitoring of study data shall include confirmation of informed consent; source data verification; data storage and data transfer procedures; local quality control checks and procedures, back-up and disaster recovery of any local databases and validation of data manipulation. The Study Coordinator or where required, a nominated designee of the Sponsor, shall carry out monitoring of study data as an ongoing activity.

Entries on CRFs will be verified by inspection against the source data. A sample of CRFs (10% or as per the study risk assessment) will be checked on a regular basis for verification of all entries made. In addition, the subsequent capture of the data on the study database will be checked. Where corrections are required, these will carry a full audit trail and justification.

Study data and evidence of monitoring and systems audits will be made available for inspection by the REC as required.

RECORD RETENTION AND ARCHIVING

In compliance with GCP guidelines and in accordance with the University of Nottingham Code of Research Conduct and Research Ethics, the Chief or local Principal Investigator will maintain all records and documents regarding the conduct of the study. These will be retained for at least 7 years or for longer if required. If the responsible investigator is no longer able to maintain the study records, a second person will be nominated to take over this responsibility.

The TMF and study documents held by the CI on behalf of the Sponsor shall be finally archived at secure archive facilities at the University of Nottingham. This archive shall include all study databases and associated meta-data encryption codes.

DISCONTINUATION OF THE STUDY BY THE SPONSOR

The Sponsor reserves the right to discontinue this study at any time for failure to meet expected enrolment goals, for safety or any other administrative reasons. The Sponsor shall take advice as appropriate in making this decision.

STATEMENT OF CONFIDENTIALITY

Individual participant medical and/or personal information obtained as a result of this study are considered confidential and disclosure to third parties is prohibited. Such medical information may be given to the participant's medical team and all appropriate medical personnel responsible for the participant's welfare.

Participant confidentiality will be further ensured by utilising identification code numbers to correspond to treatment data in the computer files.

If information is disclosed during the study that could pose a risk of harm to the participant or others, the researcher will discuss this with the CI and where appropriate report accordingly.

Data generated as a result of this study will be available for inspection on request by the participating physicians, the University of Nottingham representatives, the REC, local R&D Department and the regulatory authorities.

PUBLICATION AND DISSEMINATION POLICY

The results of this study will be submitted to peer-reviewed journals for publication as soon as data analysis is completed. The results will also be presented at conferences. Participants will not be identified in any publications. However, participants will be informed of the results of the study via a lay summary written by members of the research team.

This study will be registered with a publicly accessible database before recruitment of the first subject.

USER AND PUBLIC INVOLVEMENT

The Centre for Kidney Research and Innovation work closely with a Royal Derby Hospital patient focus group. All research studies are discussed with the group. The group provides advice with regards to studies acceptability and feasibility from a patient's perspective. We also ensure that results from studies are fed back to participants.

STUDY FINANCES

Funding source

This study is funded by internal funds from the University of Nottingham and Department of Renal Medicine, Royal Derby Hospital.

Participant spends and payments

Participants will not be paid to participate in the study. Travel expenses will be offered for any hospital visits in excess of usual care. All study assessments will take place at the time of routine haemodialysis treatment.

SIGNATURE PAGES

Signatories to Protocol:

Chief Investigator: (name)_____

Signature:_____

Date: _____

REFERENCES

1. Slee A, McKeaveney C, Adamson G, Davenport A, Farrington K, Fouque D, et al. Estimating the prevalence of muscle wasting, weakness, and sarcopenia in hemodialysis patients. J Ren Nutr. 2020;30(4):313-21. Erratum in: J Ren Nutr. 2021;31(2):e5.
2. Carrero JJ, Johansen KL, Lindholm B, Stenvinkel P, Cuppari L, Avesani CM. Screening for muscle wasting and dysfunction in patients with chronic kidney disease. Kidney Int. 2016;90(1):53-66.
3. Ikizler TA, Cano NJ, Franch H, Fouque D, Himmelfarb J, Kalantar-Zadeh K, et al. Prevention and treatment of protein energy wasting in chronic kidney disease patients: a consensus statement by the International Society of Renal Nutrition and Metabolism. Kidney Int. 2013;84(6):1096-107.
4. Bergstrom J. Mechanisms of uremic suppression of appetite. J Ren Nutr. 1999;9(3):129-32.
5. Bossola M, Muscaritoli M, Tazza L, Panocchia N, Liberatori M, Giungi S, et al. Variables associated with reduced dietary intake in hemodialysis patients. J Ren Nutr. 2005;15(2):244-52.
6. Oliveira EA, Zheng R, Carter CE, Mak RH. Cachexia/Protein energy wasting syndrome in CKD: Causation and treatment. Semin Dial. 2019;32(6):493-9.
7. Mitchell WK, Wilkinson DJ, Phillips BE, Lund JN, Smith K, Atherton PJ. Human skeletal muscle protein metabolism responses to amino acid nutrition. Adv Nutr. 2016;7(4):828S-38S.
8. Stenvinkel P, Carrero JJ, von Walden F, Ikizler TA, Nader GA. Muscle wasting in end-stage renal disease promulgates premature death: established, emerging and potential novel treatment strategies. Nephrol Dial Transplant. 2016;31(7):1070-7.
9. Lim VS, Ikizler TA, Raj DS, Flanigan MJ. Does hemodialysis increase protein breakdown? Dissociation between whole-body amino acid turnover and regional muscle kinetics. J Am Soc Nephrol. 2005;16(4):862-8.
10. Lim VS, Kopple JD. Protein metabolism in patients with chronic renal failure: role of uremia and dialysis. Kidney Int. 2000;58(1):1-10.
11. Bohe J, Rennie MJ. Muscle protein metabolism during hemodialysis. J Ren Nutr. 2006;16(1):3-16.
12. Carrero JJ, Stenvinkel P, Cuppari L, Ikizler TA, Kalantar-Zadeh K, Kaysen G, et al. Etiology of the protein-energy wasting syndrome in chronic kidney disease: a consensus statement from the International Society of Renal Nutrition and Metabolism (ISRNM). J Ren Nutr. 2013;23(2):77-90.
13. Cukor D, Cohen SD, Peterson RA, Kimmel PL. Psychosocial aspects of chronic disease: ESRD as a paradigmatic illness. J Am Soc Nephrol. 2007;18(12):3042-55.
14. Kalantar-Zadeh K. Recent advances in understanding the malnutrition-inflammation-cachexia syndrome in chronic kidney disease patients: What is next? Semin Dial. 2005;18(5):365-9.
15. Visser WJ, Egmond A, Timman R, Severs D, Hoorn EJ. Risk factors for muscle loss in hemodialysis patients with high comorbidity. Nutrients. 2020;12(9).
16. Malgorzewicz S, Galezowska G, Cieszynska-Semenowicz M, Ratajczyk J, Wolska L, Rutkowski P, et al. Amino acid profile after oral nutritional supplementation in hemodialysis patients with protein-energy wasting. Nutrition. 2019;57:231-6.
17. Liu PJ, Ma F, Wang QY, He SL. The effects of oral nutritional supplements in patients with maintenance dialysis therapy: A systematic review and meta-analysis of randomized clinical trials. PLoS One. 2018;13(9):e0203706.
18. Hendriks FK, Kooman JP, van Loon LJC. Dietary protein interventions to improve nutritional status in end-stage renal disease patients undergoing hemodialysis. Curr Opin Clin Nutr Metab Care. 2021;24(1):79-87.
19. Viramontes Hörner D, Willingham FC, Selby NM, Taal MW. Impact of dietetic intervention on skin autofluorescence and nutritional status in persons receiving dialysis: a proof of principle study. J Ren Nutr. 2020;30(6):540-7.

20. van Vliet S, Skinner SK, Beals JW, Pagni BA, Fang HY, Ulanov AV, et al. Dysregulated handling of dietary protein and muscle protein synthesis after mixed-meal ingestion in maintenance hemodialysis patients. Kidney Int Rep. 2018;3(6):1403-15.
21. Grimble RF. Basics in clinical nutrition: Immunonutrition – Nutrients which influence immunity: Effect and mechanism of action. e-SPEN. 2009;4:e10-e3.
22. Adiamah A, Skorepa P, Weimann A, Lobo DN. The impact of preoperative immune modulating nutrition on outcomes in patients undergoing surgery for gastrointestinal cancer: A systematic review and meta-analysis. Ann Surg. 2019;270(2):247-56.
23. Ikizler TA, Burrowes JD, Byham-Gray LD, Campbell KL, Carrero JJ, Chan W, et al. KDOQI Clinical Practice Guideline for Nutrition in CKD: 2020 Update. Am J Kidney Dis. 2020;76(3 Suppl 1):S1-S107. Erratum in: Am J Kidney Dis. 2021;77(2):308.

APPENDICES

Appendix 1

NUTRITION INFORMATION – TROPICAL FLAVOUR

Typical values	Per 100ml	Per 74g (Tropical flavour*)
General		
Energy kJ/kcal	1763/418	1304/309
Fat (22% kcal) g	10	7.4
of which saturates g	5.1	3.8
of which monounsaturates g	2.1	1.5
of which polyunsaturates g	3.2	2.4
Carbohydrate (54% kcal) g	56	41
of which sugars g	30	22
of which lactose g	1.2	0.89
Fibre (2% kcal) g	4.1	3
Protein (23% kcal) g	24	18
Salt g	1.1	0.81
Vitamins		
A µg	520	385
D µg	4.2	3.1
K µg	27	20
C mg	105	78
Thiamin (B1) mg	0.49	0.36
Riboflavin (B2) mg	0.80	0.59
B6 mg	0.70	0.52
Niacin mg/mg NE	1.7/6.5	1.3/4.8
Folic acid µg	140	104
B12 µg	2.5	1.8
Pantothenic acid mg	4	3
Biotin µg	20	15
E mg TE	12	8.9
Minerals		
Sodium mg/mmol	434/19	321/14
Chloride mg/mmol	608/17	450/13
Potassium mg/mmol	543/14	402/10
Calcium mg/mmol	324/8.1	240/6.0
Phosphorus/Phosphate mg/mmol	292/9.4	216/7.0
Magnesium mg/mmol	70/2.9	52/2.2
Iron mg	4.9	3.6
Zinc mg	5	3.7
Copper mg	0.69	0.51
Iodine µg	61	45
Selenium µg	25	18
Manganese mg	0.81	0.6
Chromium µg	41	30
Molybdenum µg	50	37
Fluoride mg	0.54	0.4
Other nutrients		
Choline mg	165	122
L-arginine g	5.1	3.8
Omega-3 g	1.3	0.96
MCT g	3	2.2
Nucleotides g	0.6	0.4