Nitric oxide for preventing and reducing the severity of winter infections in care (residential and nursing) homes (BEET-Winter)

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Acronym: BEET-Winter

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SYNOPSIS

Title	Nitric oxide for preventing and reducing the severity of winter infections in care (residential and nursing) homes (BEET-Winter)
Acronym	BEET-Winter
Short title	BEET-Winter
Chief Investigator	Professor Philip Bath
Objectives	**PRIMARY OBJECTIVES** 1. To assess the feasibility of recruiting and randomising care homes, identifying and treating residents with active vs placebo nitrate, to inform a definitive trial. 2. To assess the effect of nitrate on the incidence and severity of winter infections. **SECONDARY OBJECTIVES** To assess the effect of nitrate on clinical measures.
Trial Configuration	Prospective phase II cluster-randomised double-blind placebo-controlled trial assessing feasibility and proof of principle.
Setting	Social care
Sample size estimate	N=360 residents from 30 homes with 12 residents per home (range 12-17), assuming alpha 0.05, power 0.80, intra-cluster correlation (ICC) 0.01.[8] Up to six additional nursing homes, each with at least 12 participants, may be added in case some homes drop-out, or if less than 12 residents are recruited at some homes.
Number of participants	Approximately 432 adult residents from 30-36 care homes with at least 4-20 participants in each.

Eligibility criteria	**Care Home criteria** ***Inclusions*** • Ideally CQC good or outstanding rating ***Exclusions*** • Staff "live" in care home • Small homes <18 **Resident criteria** ***Inclusions*** • Age >=65 • Taking a normal / soft diet • Willing to take treatment having taste-tested a beetroot shot ***Exclusions***: • Participating in another randomised intervention trial • No consent (resident, or family if resident lacks capacity) • Using a thickener with food • Feeding tube • Using antiseptic mouthwash • Currently has an infection requiring hospitalisation • Identified by care home staff to be in last few days of life • Short-term respite care • Care home staff • Takes beetroot juice daily
Description of interventions	The intervention and placebo are foods and not investigational products. Active beetroot juice is available from supermarkets and on-line. **Intervention:** Nitric oxide (NO) in the form of 70 ml of beetroot juice containing 400 mg nitrate given once daily for 60 days. **Comparator:** Placebo in the form of 70 ml of beetroot juice containing 0 mg nitrate given once daily for 60 days.
Duration of trial	90 days for each participant: 60 days for taking juice, 30 days of follow-up)
Randomisation and blinding	Homes, and not participants, will be randomised. Randomisation will be stratified by: • Care Home type (residential vs nursing/mixed) • Prior COVID-19 in phase 1 of pandemic (yes vs no) • Size of Care Home (number of residents <32 vs >32) Residents and Care Home staff are blinded, i.e. double-blind design.

Outcome measures	**Primary endpoints** *Feasibility* • Recruitment of care homes • Recruitment of residents • Assessment of incident infection rate • Assessment of background dietary nitrate intake • Adherence to the intervention - 75% of residents take >50% • Ability to measure the ordinal outcome measure • Estimation of the intra-cluster correlation (ICC) • Incidence of death from any cause, hospitalisation for any reason, need for healthcare advice for infection, and infection *Clinical/Proof of principle* Effect of nitrate vs no nitrate on severity of worst infection (as 5 level ordinal outcome): 0 = no symptoms of infection 1 = symptoms of infection 2 = symptoms of infection needing healthcare advice (e.g. call to 111, GP) 3 = hospitalised for any reason, or intention to hospitalise but advance directive precluded this 4 = died of any cause • Use the worst level if >1 event, i.e. symptoms of cold, is trumped by symptoms of flu with confusion, is trumped by stroke needing hospitalisation, is trumped by fatal heart attack. **Secondary endpoints (at day 60 unless stated)** Clinical • Primary outcome in subgroups: age (median), sex, size of home (median), type of care home (residential, nursing, mixed), home's location (median deprivation index), infection location (respiratory tract, gastrointestinal tract, urinary tract, cutaneous, other). • Effect of nitrate vs no nitrate on severity of first infection (as 5 level ordinal outcome): 0 = no symptoms of infection 1 = symptoms of infection 2 = symptoms of infection needing healthcare advice (e.g. call to 111, GP) 3 = hospitalised for any reason, or intention to hospitalise but advance directive precluded this 4 = died of any cause • Number of infections • Time to first infection • Time to first healthcare advice • Time to first hospitalisation • Time to death • Disposition at day 60 (care home, with relative/friend, another home, hospital, died) • Clinical frailty index (CFI) [19] • Activities of daily living (Barthel index, BI) [20]

	• 6 item cognitive impairment test (6CIT) [21] • Quality of life (EQ-5D-5L, EQ-VAS) [22] • Global outcome: Primary outcome, CFI, BI, 6CIT, EQ-5D-5L, EQ-VAS [23] Safety endpoints • Death, all cause • Fatal SAEs Mechanisms • Salivary nitrite/nitrate and urinary nitrate, where feasible.[24] • Care home dietary nitrate intake (from menus and photographs of food) **Further follow-up to one month after end of intervention** • Infection after end of intervention

Statistical methods	Tabulation of Care Home and Participant characteristics, and juice consumption. Comparison of frequency and severity of infection (ordinal logistic regression), death (Cox proportional hazards).

ABBREVIATIONS

AE Adverse Event

CI Chief Investigator overall

CRF Case Report Form

DMC Data Monitoring Committee

GCP Good Clinical Practice

ICF Informed Consent Form

NHS National Health Service

PI Principal Investigator at a local centre

PIS Participant Information Sheet

REC Research Ethics Committee

R&D Research and Development department

SAE Serious Adverse Event

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TRIAL BACKGROUND INFORMATION AND RATIONALE

Winter infections

Epidemic winter respiratory infections cause much morbidity and mortality in care (residential and nursing) homes. Common viral causes include influenza A/B viruses, parainfluenza virus, respiratory syncytial virus (RSV), rhinovirus and coronaviruses (CoVs: 229E, NL63, OC43, HKU1). Bacterial causes include *Chlamydia pneumoniae*, *Haemophilus influenzae*, *Legionella* spp. and *Streptococcus pneumoniae*.[1] Nursing homes also have winter outbreaks of gastrointestinal tract infections, e.g. viral gastroenteritis due to norovirus, and skin and soft-tissue infections.[1] There is increasing concern that the ongoing COVID-19 pandemic, caused by the SARS-CoV-2 virus, will worsen again during the 2020 autumn and winter (as of September, rates are already starting to rise again in parts of the UK) and so co-exist with regular winter epidemic infections. As a result, there is a triple risk this winter (and potentially in future winters) of: (i) a surge in COVID-19; (ii) standard winter infections, especially RSV and influenza; and (iii) dual or triple infections with COVID-19 and winter viruses in the same individuals. Despite the significant enhancements made to infection control procedures in care homes during the pandemic, high mortality rates internationally in long-term care facilities due to COVID-19 highlight that co-located older people with frailty are a high-risk group for outbreaks of infectious diseases, with potentially catastrophic consequences.[2] Hence, the combination of pandemic and epidemic infections may be very challenging to care homes (and the NHS) during the coming winter.

Nitric oxide

Nitric oxide (NO) and its donors, including dietary nitrate, are generic antimicrobials with much *in vitro*, and some *in vivo*, data demonstrating anti-viral (including influenza and CoVs), bacterial, protozoal and fungi/yeast activity (Bath *et al*, review in preparation [3]). The antimicrobial effects of NO and derivative molecules such as peroxynitrite are mediated by effects on DNA and protein conformation.[3] NO also improves organ blood supply and has anti-inflammatory and antithrombotic effects mediated through anti-leucocyte and anti-platelet activity, and these effects may also contribute to the antimicrobial effects. Antiviral and antibacterial activity has been demonstrated against many of the common causes of respiratory, gastrointestinal and soft-tissue infections. Phase II clinical trials have supported anti-microbial effects of acidified nitrite on cutaneous viral and bacterial infections, dietary nitrate on oral bacteria, and NO gas against some respiratory viral infections. In addition to microbial-static/cidal effects, NO has vasodilatory/pro-endothelial/anti-platelet effects [4] which may also counteract the negative vascular effects of infection. Further, NO (as beetroot juice) has been shown to improve exercise performance (as used by GB elite athletes in the 2012 Olympics) and cognition in older people,[5] potential benefits of relevance to care home residents. Although some common infections have vaccines available (e.g. influenza), many do not (e.g. RSV) and vaccinations may need to be combined with chemoprophylaxis for effective prevention.[6] So, NO donors may be particularly relevant due to their potential generic antimicrobial effects even in the presence of vaccination, and especially since resistance against NO appears to be rare [3] in contrast to many specific antibacterial agents.[1]

Cluster trials in care homes

Trials in care/nursing homes have an established history.[7] Because of the risk of confusion in delivering both active and placebo nitrate within a care home and so crossing-over treatment, it is more appropriate to randomise care/nursing homes to one or other intervention. Cluster randomised trials are relatively common in care/nursing homes and design parameters for some of these trials are given in Table 1.

Table 1. Design criteria in published cluster care home trials

	Target	N	Homes	N/home	Active	Control	RRR	ARR	ICC	A
Infection										
Baldwin 2010 [8]	MRSA	480	24	20	15.3	17.0	10	1.7	0.01	
Chami 2012 [9]	All	3,524	44	80	4.1	8.1		4.0	0.04	
Mody 2015 [10]	UTI	418	12	35			23 S			
Gravenstein 2017 [11]	Influenza	75,917	823	92	3.4	3.9	11.2	0.5	0.35	
Gravenstein 2018 [12]	Influenza	2,957	39	76	13.5	20.1	33.1	6.5	?	
Loizeau 2019 [13]	UTI/LRI	410	28	15				0.38	0.01	
Arnold 2020 [14]	UTI	1,274	22	58	0.30	0.15	50	0.15	0.07	
Teesing 2020 [15]	Hand hygiene		45	6	50	35		15	0.40	
Others, Nottingham										
OTCH 2015 [16]	OT	1042	228	4.6	5.5	5.3		0.2	0.37	
Walker 2016 [17]	Falls	52	6	8.7	1.9	4.0			-	
Logan 2020 (in prep)	Falls	1308	66	20	1.65pa	2.5pa	33%		0.10	

ICC: intra-cluster correlation; LRI: lower respiratory tract infection; MRSA: methicillin-resistant *Staphylococcus aureus*; OT: occupational therapy; UTI: urinary tract infection

Rationale
The proposed trial takes account that: (i) Respiratory epidemic and norovirus infections in care homes will increase during the coming autumn and winter; (ii) A further surge of COVID-19 may occur in parallel; (iii) The symptoms of many respiratory infections overlap so it will not be possible to distinguish clinically the causative organism (e.g. influenza virus vs. RSV vs SARS-CoV-2); (iv) Co-infections caused with two or more pathogens of concern may interact in as yet undetermined ways; (v) Most ongoing trials in care/nursing homes focus specifically on COVID-19 and their interventions are unlikely to have effects on other respiratory viral pathogens; (vi) Dietary NO has a very low risk of harm and yet may reduce infections and their severity and so potentially save lives; and (vii) Antigen/antibody tests, when done, can identify most of the common organisms that cause winter infections. As a result, a trial using a generic anti-microbial and focussing on the prevention of winter infections in care/nursing homes may reduce morbidity and mortality, including from COVID-19.

TRIAL OBJECTIVES AND PURPOSE

PURPOSE
1. To determine if it is feasible to recruit residents in care homes into a trial aiming to reduce winter infections.
2. To determine if nitric oxide given as dietary nitrate reduces winter-timed infections and their severity in care homes residents.

PRIMARY OBJECTIVE
1. To assess the feasibility of recruiting and randomising care homes, randomising, identifying and treating residents with active vs placebo nitrate, to inform a definitive trial.
2. To assess the effect of nitrate on the incidence and severity of winter infections.

SECONDARY OBJECTIVES
To assess the effect of nitrate on clinical measures of function.

DETAILS OF PRODUCT(S)
The intervention and placebo are foods and not investigational products. Beetroot juice is used as the nitric oxide-donating inorganic nitrate since it offers matching fixed dose and placebo versions, as used in previous randomised controlled trials. Active beetroot juice is available from supermarkets and on-line.

Description

Intervention: Nitric oxide in the form of 70 ml of beetroot juice containing 400 mg nitrate given once daily for 60 days (Beet It Nitrate 400 shots). The dose is as used in previous trials.

Manufacture

James White Drinks Ltd

.

Packaging and labelling

Active and placebo beetroot juice is provided in 70 ml plastic drinking containers
Active and placebo are packaged in identical containers, labelled as "James White Beet It Beetroot Juice Sport Shot - 70ml, and are as sold on-line by JWD. JWD will deliver packs of shots to each Care Home directly according to a randomisation code.

Storage, dispensing and return

Both active and placebo juice are stored at room temperature and have a shelf life >1 year.

Beetroot shots (active/placebo) will be kept in Care Homes at room temperature as per their usual protocol for food supplements. Unused stock will be disposed of as kitchen waste.

Placebo

Placebo in the form of 70 ml of beetroot juice containing 0 mg nitrate given once daily for 60 days. Active and placebo containers are identical in look, and juice is identical in look, smell and taste.

Ingredients

The juice is crushed beetroot juice to which 2% lemon juice from concentrate is added. There are no other additives, preservatives or allergens, and there is no risk of cross-contamination during manufacturing. The nitrate-free juice is the same juice but with nitrate removed using a microporous selective strong base anion resin; this process too avoids the risk of allergens and other cross contamination.

Reference source: Information from the manufacturer, James White Drinks Ltd.

Known Side Effects

Juice is palatable to many but taste can be masked by dilution in other juices or consumption via a straw. Beetroot juice may be continued in hospital if the resident is admitted.

Reviews of dietary nitrate consumption indicate positive effects in older adults on physiological performance, potential benefits on cardiovascular and cerebrovascular health, mixed effects on cognitive performance, and no benefit on metabolic health.
The following side effects have been reported:

- Common: Colouration of urine red or pink (beeturia).
- Uncommon: Colouration of stools/faeces red.
- Rare with short-term use: Rash, stomach cramps
- Rare with prolonged use: Renal stones

There are no known incompatibilities or interactions.

Participants will be reassured that should the beetroot juice colour their urine, that this is normal and that the exclusion of any suspected UTI will be addressed following the usual practice of the Care Home

Reference source: Information from web.

TRIAL DESIGN

TRIAL CONFIGURATION

Prospective phase II cluster-randomised double-blind placebo-controlled trial assessing feasibility and proof of principle.

Primary endpoint

Feasibility

- Recruitment of care home
- Recruitment of residents
- Assessment of incident infection rate
- Assessment of background dietary nitrate intake
- Adherence to the intervention - 75% of participants take >50%
- Ability to measure the ordinal outcome measure
- Estimation of the intra-cluster correlation (ICC)
- Incidence of death from any cause, hospitalisation for any reason, need for healthcare advice for infection, and infection

Clinical/Proof of principle

Effect of nitrate vs no nitrate on severity of worst infection (as 5 level ordinal outcome):
0 = no symptoms of any infection
1 = symptoms of an infection
2 = symptoms of an infection needing healthcare advice (e.g. call to 111, GP)
3 = hospitalised for any reason, or intention to hospitalise but advance directive precluded this
4 = died of any cause

- Use the worst level if >1 event, i.e. symptoms of cold, is trumped by symptoms of flu with confusion, is trumped by stroke needing hospitalisation, is trumped by fatal heart attack.

Secondary endpoint

Clinical

- Primary outcome in subgroups: age (median), sex, size of home (median), type of care home (residential, nursing), home's location (median deprivation index), infection location (respiratory tract, gastrointestinal tract, urinary tract, cutaneous).
- Effect of nitrate vs no nitrate on severity of first infection (as 5 level ordinal outcome):
  0 = no symptoms of any infection
  1 = symptoms of an infection
  2 = symptoms of an infection needing healthcare advice (e.g. call to 111, GP)
  3 = hospitalised for any reason, or intention to hospitalise but advance directive precluded this
  4 = died of any cause
- Number of infections
- Time to first infection
- Time to first healthcare advice
- Time to first hospitalisation
- Time to death
- Disposition at day 60 (care home, with relative/friend, another home, hospital, died)
- Clinical frailty index (CFI) [19]
- Activities of daily living (Barthel index, BI) [20]
- 6 item cognitive impairment test (6CIT) [21]
- Quality of life (EQ-5D-5L, EQ-VAS) [22]
- Global outcome: Primary outcome, CFI, BI, 6CIT, EQ-5D-5L, EQ-VAS [23]

Safety endpoints
Safety endpoints
- Death, all cause
- Fatal SAEs

Mechanisms
- Salivary nitrite/nitrate and urinary nitrate, where feasible.[24]
- Care home dietary nitrate intake (from menus and photographs of food).[25]

Further follow-up to one month after end of intervention
- Infection after end of intervention

Stopping rules and discontinuation
Participants (or relatives if the participants lacks capacity) may withdraw from treatment (but continue follow-up), or withdraw from the trial. Since the trial will recruit participants together, final follow-up will also occur together so the trial will not stop during treatment/follow-up. There will be no interim analysis.

RANDOMISATION AND BLINDING
This is a cluster randomised trial.

Care Homes, not participants, will be randomised. We will allocate care homes dynamically as they enrol and complete consent procedures in residents, balancing for the following key characteristics using minimisation: type (residential v nursing/mixed); previous COVID-19 in home (Yes/No); size (<32/>32).

We have selected these variables because they have been shown in large scale epidemiological studies, reported in the minutes of the SAGE Social Care Working Group, to influence the risk and severity of COVID-19. We believe these factors will similarly influence the spread and severity of other airborne winter infections.

Active and placebo shotes are identical and neither Care Homes, their staff nor participants will know what intervention they are receiving.

Randomisation will be blinded to Chief Investigator, Trial Manager. The Statistician will be unblinded and not in contact with Care Homes. The Trial Coordinator will be unblinded and responsible for coordination between the Care Homes and juice provider.

The block size is defined by the number of Care Homes randomised to each of Active/Placebo, as randomised 1:1. Allocation is stratified and analyses will take account of minimisation.

Participants will be enrolled by Care Home staff and receive active or placebo shots as determined by how their Care Homes was randomised. Randomisation will be concealed to end of treatment for the final participant at 60 days.

TRIAL MANAGEMENT

The Chief Investigator has overall responsibility for the trial and shall oversee all trial management.
The data custodian will be the Chief Investigator.

The trial will be managed overall by the Trial Steering Committee, membership as listed at the top of the protocol. The Trial Management Committee (Chief Investigator, Senior Trial Manager, Trial Coordinator, Statistician) will manage the trial day-by-day. As a feasibility trial and with all participants starting randomised treatment and reaching follow-up almost all together, there is no Data Monitoring Committee.

Care Home staff will be trained in the trial (usually the Care Home manager, senior team leader or senior nursing staff) and they will approach residents and assess capacity. The Care Home Manager will be treated as the Site Investigator.

Care Home Managers will be provided with a full training manual, and together with their staff will attending a training session lead by the Chief Investigator, Trial Manager and Trial Coordinator.

DURATION OF THE TRIAL AND PARTICIPANT INVOLVEMENT

Participants

Participants will all join the trial at the start of the study and the intervention will be given for 60 days (Table 2, Figure 1). Altogether, participants will be involved in the trial for 90 days.

Table 2. Flow of participants

	Screen	Baseline	Day 1-60	Day 14	Day 60	Day 90
Eligibility	+					
Consent, can tolerate juice	+					
Baseline, past history		+				
Clinical (CFI, ADL-BI, 6CIT, EQ-5D-5L, EQ-VAS)		+			+	
Saliva/urine [Nitrate, nitrite]		+			+	+
Dietary nitrate intake from menus		+		+	+	+
Beetroot juice adherence				+		
Event 1			<=>			
Event 2			<=>			
Event 3			<=>			
End of beetroot juice					+	
End of Follow-Up						+
End of Study						+

6CIT: 6-item cognitive impairment test; ADL-BI: Activities of daily living-Barthel index; EQ-VAS: EuroQoL visual analogue scale
Events 1-3 are components of the ordinal outcome: no infection, infection, healthcare involvement, hospitalisation, death.

Study duration
The study will be designed and set-up in later summer/early autumn 2020, and the intervention given in November-January (Table 3).

Table 2. GANNT Chart

Year	2020					2021	
Month	Aug	Sept	Oct	Nov	Dec	Jan	Feb
Regulatory approvals [1]	<	=	>				
Program database	<	=	>				
Funding		<	=	=	=	>	
CC training		<	>				
Order intervention		<	>				
Identification of homes		<	>				
Training & initiation of homes			<	>			
Identify participating residents [2]			<	>			
Distribute intervention				<>			
Recruitment				FPFV LPFV			
Sites				26	26	26	
Recruitment/month [3]				360	0	0	
Recruitment, total				360	360	360	
Intervention [4]				<	=	>	
On-line data entry to day 60				<	=	>	
Final endpoint ascertainment						FPLV LPLV	
Data cleaning				<	=	=	>
Ascribe infection diagnosis [5]							
Publish SAP					<>		
Analysis & writing						<	>

CC: Coordinating Centre; FPFV: first patient first visit; FPLV: first patient last visit LPFV: last patient first visit; LPLV: last patient last visit

1. Trial approvals (ethics, HRA, Care Home approval. MHRA approval is not necessary)
2. Consent and can tolerate test beetroot juice intervention
3. Slow recruitment could mean start treatment in December and finish in February
4. Start intervention on 18/11/20
5. Ascribe infection diagnosis from investigations (if available) and/or symptoms (table 3)

End of the Trial

The end of the study will be the last follow up of the last participant.

SELECTION AND WITHDRAWAL OF PARTICIPANTS

Recruitment

Participants will be recruited from residential and nursing homes. The initial approach will be from a member of the participant's usual care team (which may include the investigator).

The investigator or their nominee, e.g. a member of the participant's usual care team, will inform the participant or their nominated representative (other individual or other body with appropriate jurisdiction), of all aspects pertaining to participation in the study.

Interpreter and translation services will not be available for this trial. Communication requirements will already have been assessed and addressed within the care home. We will use the usual support for any resident who has communication issues.

It will be explained to the potential participant that entry into the trial 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 but attempts will be made to avoid this occurrence. 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.

Eligibility criteria

Care Home criteria
Inclusions

- Ideally CQC good or outstanding

Exclusions

- Staff "live" in care home (a marker of less infection being brought into homes)
- Small homes <18

Resident criteria
Inclusions

- Age >=65 years
- Taking a normal / soft diet
- Willing to take the intervention having taste-tested a beetroot shot

Exclusions:

- Participating in another intervention trial
- Has not given consent (resident, or family if resident lacks capacity)
- Using a thickener in food/fluids
- No feeding tube
- Using antiseptic mouthwash
- Currently has an infection requiring hospitalisation
- Identified by care home staff to be in last few days of life
- Short-term respite care
- Care home staff
- Already taking beetroot juice regularly

Identification of Residents.
We will reduce bias by recruiting Residents at random from each care home. Specifically, we will give the Care Homes random numbers by which to identify Residents from an alphabetical list of their names. They will approach Residents in numerical order until reach at between 4-20 Residents.

Expected duration of participant participation
Trial participants will be participating in the trial for 90 days (60 taking juice, 30 follow-up).

Removal of participants from therapy or assessments/Participant Withdrawal

Participation in the trial is voluntary and home residents are free to stop treatment, miss follow-up visits or withdraw completely from the trial at any stage without giving a reason.

- ***Study intervention***: This may be stopped at any time by the participant, or by home staff or relatives (if the participant lacks capacity) if deemed in the participants' best interest. The intervention will be given in addition to care perceived as best practice. Stopping study intervention does not constitute withdrawal and follow-up should be continued as per this protocol.
- ***Follow-ups***: These may be missed but this does not constitute withdrawal from the trial and the remaining follow-up visits (e.g. days 14, 60 and 90) should be continued as per this protocol.
- ***Withdrawal from trial***: Participants may withdraw from the trial either at their own request or at the discretion of the home staff or relatives (if the participant lacks capacity). The participants will be made aware that this will not affect their future care. Participants will be made aware (via the information sheet) that should they withdraw, data collected to date cannot be erased and may still be used in the final analysis.

Informed consent*

Care Home staff, trained by the research trial team, will approach eligible care home residents/relatives if the resident lacks capacity.

We will approach the resident for consent, or a consultee for proxy consent for residents who lack capacity.

The consultee will be a relative as identified from the Care Home records and will be approached by the Care Home. If a relative is not available or does not respond within 3 days of contact, the Care Home manager may give consent by proxy, as per the FinCH trial (HTA Journal 2020 in press).

The research team are experienced in recruiting those who lack capacity to trials and have previously trained care home staff to conduct these assessments in a research context (FINCH, HTA Report in press, 2020)

Capacity will be assessed using the '3 question approach' used in the RIGHT-2 trial (Lancet 2019), as delivered by paramedics. Nominated Care Home staff trained in the trial (usually the Manager, senior team leaders or senior nursing staff) will first tell residents 3 pieces of information:

- We want to **prevent infections** and reduce their severity;
- We will ask you to drink **beetroot juice** every day for 2 months;
- We will ask you **questions** at 2 months to see if you have developed any infections.

The care home staff will then ask the resident to feed this information back. If all the key underlined/bold information is correctly given, then capacity is demonstrated. Otherwise the resident is assumed to lack capacity (which may apply to approximately 60% of residents).

After assessing capacity, the care home investigator or their nominee will explain the trial to the resident/relative, and answer any questions the resident/relative may have. Following the assessment of capacity, the Resident will be asked to test taste the juice to see if they can take the beetroot juice regularly. Consent or consultee advice will then be sought and the Resident will enter the trial. This will be recorded in the resident's notes/records that they have been recruited into the trial and that they have completed the taste test.

The decision regarding participation in the trial is entirely voluntary. The care home investigator or their nominee shall emphasize to them that consent regarding trial 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 trial-specific interventions will be done before the trial has been explained to the resident/relative and they have had the opportunity to decline consent.

For residents/relatives who do not give consent, it will be recorded in the resident's notes/records that they have declined to enrol in the trial.

TRIAL INTERVENTION AND REGIMEN

The intervention and placebo are foods and not investigational products. Active beetroot juice is available from supermarkets and on-line.

Intervention: NO in the form of 70 ml of beetroot juice containing 400 mg nitrate given once daily for 60 days.

Comparator: Placebo in the form of 70 ml of beetroot juice containing 0 mg nitrate given once daily for 60 days.

Questionnaires will be completed online by Care Home staff, with help from the resident as appropriate, at designated timepoints:

- Screening – completed by Care Home staff to confirm eligibility
- Baseline – completed by/on behalf of the resident
- Nitrate/nitrite levels – a sample of saliva and urine will be dipstick tested by the Care Home staff. All samples and test strips will be immediately destroyed as per Care Home policy

- Food intake
- Symptoms/Infection diagnosis – completed when/if the participant develops an infection
- Food intake
- Nitrate/nitrite levels as above
- End of treatment – completed after 60 days of taking beetroot juice
- End of follow up – completed 30 days after the end of juice
- End of study – completed at the end of the study

Nitrate/Nitrite level assessment mechanisms

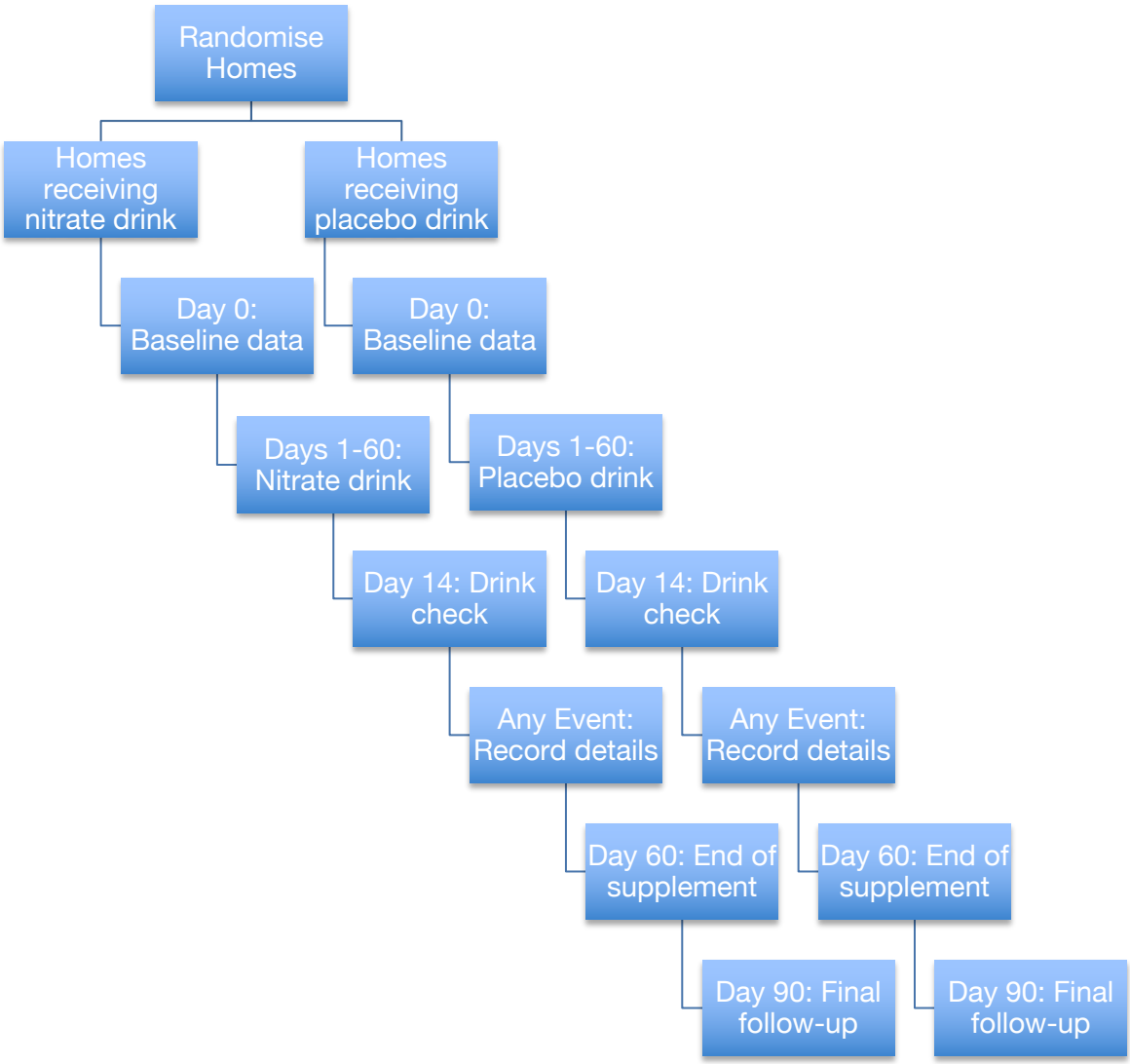
Salivary nitrite/nitrate and urinary nitrate

Salivary/urine nitrate/nitrite concentrations will be measured from samples provided by the resident immediately after collection using a commercial test strip. These strips will then be discarded into clinical waste containers. No other samples will be taken.

Care home dietary nitrate intake

We will collect meal plans and photographs of meals pre and post consumption on 4 occasions: Day 0 (baseline), Day 14, Day 60 (end of juice) and Day 90 (end of study). These will be assessed for dietary nitrate content.

Figure 1. Flow chart



Compliance
Ideally all 60 shots should be taken; in the event of a dose being missed, this should be recorded in the resident's notes/records and on the electronic data form.

Risks
Competing trials: There are no directly competing trials testing nitrate supplementation in care-homes whether for prophylaxis/treatment of COVID-19 or other infections. However, there are three anti-COVID-19 trials that are targeting nursing homes: PRINCIPLE, AVID-CC and CONDOR-CH. The uptake of PRINCIPLE is improving and AVID-CC will start in the Autumn. Neither of these trials will predominate in the care home sector to the extent that it would render recruitment to the proposed study challenging. CONDOR-CH is a point of care study and could run in parallel with the proposed study. Importantly, there are 420,000 care home residents in England so there should be sufficient care homes to approach..
Low Infection rate: Although many experts believe that the usual winter flu season will accompany the current surge in COVID-19, effective social distancing and other measures might mean that infection rates will be low. Assessment of the underlying infection rate is one of the feasibility aims.
Background nitrate intake: The study assumes that background consumption of dietary nitrate such as lettuce, rocket, spinach, beetroot etc is only moderate in care homes so that active shots will substantially increase nitrate intake. Unfortunately there is no literature on this and a feasibility aim is to assess nitrate intake using menu cards from the participating homes.
Tolerance: Although inorganic nitrates are not considered to exhibit tolerance (in contrast to organic nitrates) this remains to be demonstrated in a large study

Criteria for terminating trial
As this trial is non-medical and is a dietary supplement it is unlikely that we would need to terminate the trial. It is imperative that the trial continues during the second wave of coronavirus infection so that data can be analysed to determine the effect of beetroot juice in preventing infection/limiting the effects of the infection.

TRANSPORT AND STORAGE OF THE TISSUES

There will be no collection of samples. The urine and saliva samples will be measured using a test stick, and both samples and test sticks will then be disposed of in the usual way as per Care Home policies.

STATISTICS

Methods
Tabulation of Care Home and Participant characteristics, and juice consumption.
Comparison of frequency and severity of infection (ordinal logistic regression), death (Cox proportional hazards).

Sample size and justification
This phase II trial: N=360 residents from 30 homes with 12 residents per home (range 12-17), assuming alpha 0.05, power 0.80, intra-cluster correlation (ICC) 0.01.[8] Up to six

additional nursing homes, each with between 4-20 participants, may be added in case some homes drop-out, or if less than 12 residents are recruited at some homes.

Assessment of efficacy
Efficacy will be the number and severity of infections

Assessment of safety
Number of deaths in the trial

Procedures for missing, unused and spurious data
Missing data will be reported. The investigation of this data and methods implemented to address the missing data, if appropriate, will be detailed in the SAP.

Definition of populations analysed
Safety set: All randomised participants who receive at least one treatment.
Full Analysis set: All randomised participants, who participated in at least one treatment 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

Adverse events will not be collected in this trial.

Side effects include:
- Common: Colouration of urine red or pink (beeturia).
- Uncommon: Colouration of stools/faeces red.
- Rare with short-term use: Rash, stomach cramps
- Rare with prolonged use: Renal stones

There are no known incompatibilities or interactions.

Participant removal from the trial due to adverse events
Participants are not expected to suffer adverse events as this is a non-medical intervention.

ETHICAL AND REGULATORY ASPECTS

ETHICS COMMITTEE AND REGULATORY APPROVALS
The trial will not be initiated before the protocol, informed consent forms and participant information sheets have received approval / favourable opinion from the Research Ethics Committee (REC). The local CCG and Director of Social Services will be made aware of the trial.. 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 forms and participant information sheets (if appropriate) have been reviewed and received approval / favourable opinion from the REC and R&D departments. A protocol amendment intended to eliminate an apparent immediate hazard to participants may be implemented immediately providing that the REC are 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 trial will be conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki, 1996; the principles of Good Clinical Practice, and the UK Department of Health Policy Framework for Health and Social Care, 2017.

INFORMED CONSENT AND PARTICIPANT INFORMATION

The process for consent/consent by proxy by either the resident/relative or Care Home manager (if no named relative on record) will be in accordance with the REC guidance, and Good Clinical Practice (GCP) and any other regulatory requirements that might be introduced.

The decision regarding participation in the trial is entirely voluntary. The investigator or their nominee shall emphasize to them that consent regarding trial 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 trial-specific interventions will be done before the trial has been explained to the resident/relative and they have had the opportunity to decline consent.

The investigator will inform the participant of any relevant information that becomes available during the course of the trial, and will discuss with them, whether they wish to continue with the trial.

If the consent process is amended during the trial, the investigator shall follow all applicable regulatory requirements pertaining to approval of the amended consent forms the REC.

RECORDS

Case Report Forms

Each participant will be assigned a trial identity code number, allocated at randomisation if appropriate, for use on CRFs other trial documents and the electronic database. The documents and database will also use their name and date of birth (dd/mm/yyyy). Identifiable data will only be available for access to the staff at the corresponding Care Home. Identifiable data will not be available to the trial staff, including Trial Manager, Trial Coordinator or Trial Statistician.

Electronic CRFs will be treated as confidential documents and held on a secure database 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 Trial Number (the Trial Recruitment Log), to permit identification of all participants enrolled in the trial, in accordance with regulatory requirements and for follow-up as required
CRFs shall be restricted to those personnel approved by the Chief or local Principal Investigator and recorded on the 'Trial Delegation Log.'

Sample Labelling

The salivary/urine nitrate/nitrite concentrations will be measured immediately after collection using a commercial test strip. The strips and concentrations will then be discarded into clinical waste containers. No other samples will be taken and so nothing will be stored.

Source documents

Data will be collected either directly from the participant or Care Home staff and uploaded onto a secure database. Source documents shall be filed at the investigator's site and may include but are not limited to, current medical records, laboratory results and records. A CRF may also completely serve as its own source data. Only trial staff as listed on the Delegation Log shall have access to trial 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 Chief Investigator, Sponsor's designee and inspection by relevant regulatory authorities.

DATA PROTECTION

All trial staff and investigators will endeavour to protect the rights of the trial's participants to privacy and informed consent, and will adhere to the Data Protection Act, 2018. The e-CRF will only collect the minimum required information for the purposes of the trial. Access to the information will be limited to the trial staff and investigators and relevant regulatory authorities (see above). Computer held data including the trial database will be held securely and password protected. All data will be stored on a secure dedicated web server. Access will be restricted by user identifiers and passwords (encrypted using a one way encryption method).
Information about the trial 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
The University of Nottingham as research Sponsor indemnifies its staff, research participants and research protocols with both public liability insurance and clinical trials insurance. These policies include provision for indemnity in the event of a successful litigious claim for proven non-negligent harm.

TRIAL CONDUCT
Trial conduct may be subject to systems audit of the Trial Master File for inclusion of essential documents; permissions to conduct the trial; Trial Delegation Log; CVs of trial 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, correct randomisation, timeliness of visits.

The Trial Coordinator or where required, a nominated designee of the Sponsor, shall carry out a site systems audit at least once during the trial. The audit may be conducted remotely or in person as appropriate. An audit report shall be made to the Trial Steering Committee.

TRIAL DATA
Monitoring of trial data shall include confirmation of opt out consent; source data verification; data transfer procedures; local quality control checks and procedures, back-up and disaster recovery of any local databases and validation of data manipulation. The Trial Coordinator,

or where required, a nominated designee of the Sponsor, shall carry out monitoring of trial data as an ongoing activity.

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

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

RECORD RETENTION AND ARCHIVING

In compliance with the ICH/GCP guidelines, regulations and in accordance with the University of Nottingham Research Code of Conduct and Research Ethics, the Chief or local Principal Investigator will maintain all records and documents regarding the conduct of the trial. 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 trial records, a second person will be nominated to take over this responsibility.

The Trial Master File and trial documents held by the Chief Investigator on behalf of the Sponsor shall be finally archived at secure archive facilities at the University of Nottingham. This archive shall include all trial databases and associated meta-data encryption codes.

DISCONTINUATION OF THE TRIAL BY THE SPONSOR

The Sponsor reserves the right to discontinue this trial at any time for failure to meet expected enrolment goals, for safety or any other administrative reasons. The Sponsor shall take advice from the Trial Steering Committee and Data Monitoring Committee as appropriate in making this decision.

STATEMENT OF CONFIDENTIALITY

Individual participant medical information obtained as a result of this trial are considered confidential and disclosure to third parties is prohibited with the exceptions noted above. Participant confidentiality will be further ensured by utilising identification code numbers to correspond to treatment data in the computer files.

Such medical information may be given to the participant's medical team and all appropriate medical personnel responsible for the participant's welfare.

If information is disclosed during the trial 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 trial will be available for inspection on request by the participating physicians, the University of Nottingham representatives, the REC, local R&D Departments and the regulatory authorities.

PUBLICATION AND DISSEMINATION POLICY

Reporting, dissemination and notification of the results

Trial results will be published in a peer reviewed academic journal. Reporting will be in compliance with CONSORT recommendations. The focus of that article will be to discuss

the feasibility of using beetroot juice in Care Home and the proof of concept. Findings will also be presented at relevant conferences.

Policy for publication and authorship
The main and secondary trial results will be published by named members of the trial team.

USER AND PUBLIC INVOLVEMENT

The study and protocol was discussed with our PPI representative. Participant information Sheets / Relative Information Sheets and Consent forms have been discussed with the PPI representative.

TRIAL FINANCES

Funding source
This trial is funded by an NIHR Senior Investigator award (£50K) and the Nottingham Biomedical Research Centre (£20K)

Participant stipends and payments
Participants will not be paid to participate in the trial.

SIGNATURE PAGES

Signatories to Protocol:

Chief Investigator: (name)_____

Signature:_____

Date: _____

Co- investigator: (name) _____

Signature:_____

Date: _____

Trial Statistician: (name)_____

Signature:_____

Date: _____

REFERENCES

1. Strausbaugh LJ, Sukumar SR, Joseph CL. Infectious disease outbreaks in nursing homes: an unappreciated hazard for frail elderly persons. *Clin Infect Dis* 2003; **36**(7): 870-6.
2. Comas-Herrera A, Zalakaín J, Litwin C, et al. Mortality associated with COVID-19 outbreaks in care homes: early international evidence. *LTCcovidorg, International Long-Term Care Policy Network, CPEC-LSE*, 26 June, 2020. (accessed.
3. Bath PM, Coleman CM, Lim W-S, Webb AJ. Nitric oxide for preventing and treating viral (including coronaviruses/COVID-19), bacterial, parasitic and other infections - a neglected generic antimicrobial. *To be submitted* 2020.
4. Mills CE, Khatri J, Maskell P, Odongerel C, Webb AJ. It is rocket science - why dietary nitrate is hard to 'beet'! Part II: further mechanisms and therapeutic potential of the nitrate-nitrite-NO pathway. *Br J Clin Pharmacol* 2017; **83**(1): 140-51.
5. Stanaway L, Rutherfurd-Markwick K, Page R, Ali A. Performance and Health Benefits of Dietary Nitrate Supplementation in Older Adults: A Systematic Review. *Nutrients* 2017; **9**(11).
6. Bradley SF. Prevention of influenza in long-term-care facilities. Long-Term-Care Committee of the Society for Healthcare Epidemiology of America. *Infect Control Hosp Epidemiol* 1999; **20**(9): 629-37.
7. Gordon AL, Logan PA, Jones RG, et al. A systematic mapping review of randomized controlled trials (RCTs) in care homes. *BMC Geriatr* 2012; **12**: 31.
8. Baldwin NS, Gilpin DF, Tunney MM, et al. Cluster randomised controlled trial of an infection control education and training intervention programme focusing on meticillin-resistant Staphylococcus aureus in nursing homes for older people. *J Hosp Infect* 2010; **76**(1): 36-41.
9. Chami K, Gavazzi G, Bar-Hen A, et al. A short-term, multicomponent infection control program in nursing homes: a cluster randomized controlled trial. *J Am Med Dir Assoc* 2012; **13**(6): 569.e9-17.
10. Mody L, Krein SL, Saint S, et al. A targeted infection prevention intervention in nursing home residents with indwelling devices: a randomized clinical trial. *JAMA Intern Med* 2015; **175**(5): 714-23.
11. Gravenstein S, Davidson HE, Taljaard M, et al. Comparative effectiveness of high-dose versus standard-dose influenza vaccination on numbers of US nursing home residents admitted to hospital: a cluster-randomised trial. *Lancet Respir Med* 2017; **5**(9): 738-46.
12. Gravenstein S, Davidson HE, Han LF, et al. Feasibility of a cluster-randomized influenza vaccination trial in U.S. nursing homes: Lessons learned. *Hum Vaccin Immunother* 2018; **14**(3): 736-43.
13. Loizeau AJ, D'Agata EMC, Shaffer ML, et al. The trial to reduce antimicrobial use in nursing home residents with Alzheimer's disease and other dementias: study protocol for a cluster randomized controlled trial. *Trials* 2019; **20**(1): 594.
14. Arnold SH, Jensen JN, Kousgaard MB, Siersma V, Bjerrum L, Holm A. Reducing Antibiotic Prescriptions for Urinary Tract Infection in Nursing Homes Using a Complex Tailored Intervention Targeting Nursing Home Staff: Protocol for a Cluster Randomized Controlled Trial. *JMIR Res Protoc* 2020; **9**(5): e17710.
15. Teesing GR, Erasmus V, Petrignani M, et al. Improving Hand Hygiene Compliance in Nursing Homes: Protocol for a Cluster Randomized Controlled Trial (HANDSOME Study). *JMIR Res Protoc* 2020; **9**(5): e17419.
16. Sackley CM, Walker MF, Burton CR, et al. An occupational therapy intervention for residents with stroke related disabilities in UK care homes (OTCH): cluster randomised controlled trial. *BMJ* 2015; **350**: h468.

17. Walker GM, Armstrong S, Gordon AL, et al. The Falls In Care Home study: a feasibility randomized controlled trial of the use of a risk assessment and decision support tool to prevent falls in care homes. *Clin Rehabil* 2016; **30**(10): 972-83.
18. British Geriatrics Society. Managing the COVID-19 pandemic in care homes, 2020.
19. Rockwood K, Song X, MacKnight C, et al. A global clinical measure of fitness and frailty in elderly people. *Canadian Medical Association Journal* 2005; **173**(5): 489-95.
20. Mahoney FI, Barthel DW. Functional evaluation: The Barthel Index. *Maryland State Medical Journal* 1965: 61-5.
21. Brooke P, Bullock R. Validation of a 6 item cognitive impairment test with a view to primary care usage. *Int J Geriatr Psychiatry* 1999; **14**(11): 936-40.
22. Euroqol Group. EuroQol--a new facility for the measurement of health-related quality of life. The EuroQol Group. *Health Policy* 1990; **16**(3): 199-208.
23. Lachin J. Applications of the Wei-Lachin multivariate one-sided test for multiple outcomes on possibly different scales. *PLoS One* 2014; **9**(10).
24. Clodfelter WH, Basu S, Bolden C, Dos Santos PC, King SB, Kim-Shapiro DB. The relationship between plasma and salivary NOx. *Nitric Oxide* 2015; **47**: 85-90.
25. Jackson JK, Patterson AJ, MacDonald-Wicks LK, et al. Dietary Nitrate and Diet Quality: An Examination of Changing Dietary Intakes within a Representative Sample of Australian Women. *Nutrients* 2018; **10**(8).