Participant Information Sheet Part 4 (b)

Title of study: **An investigation into the effect of dietary nitrate on blood vessel function in healthy volunteers**

Simplified title: Effect of nitrate on blister formation

1. Invitation paragraph

You are being invited to take part in a clinical study. Before you decide it is important for you to understand why the research is being done and what it will involve. Please take time to read the following information carefully and discuss it with others if you wish. Ask us if there is anything that is not clear or if you would like more information. Take time to decide whether or not you wish to take part.

Thank you for reading this.

2. What is the purpose of the study?

The goal of the research is to determine whether nitrate, present in many vegetables (including beetroot juice) can change the response to inflammation. Chronic sustained inflammation is present in many diseases, including arthritis, heart disease and cancer. We are using a naturally-occurring protein called cantharidin to induce a mild and temporary inflammation in the skin. The cantharidin is applied to the skin of the forearm (or elsewhere) and a small (~1cm^2) blister forms within 24h, which we are able to study to gain insights into the effects of nitrate on inflammation.

3. Why have I been chosen?

You have been approached because you are healthy and well. 24 subjects will be recruited in the study.

4. Do I have to take part?

It is up to you to decide whether or not to take part. If you do decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still free to withdraw at any time and without giving a reason. A decision to withdraw at any time, or a decision not to take part, will have no adverse consequences (if you are a student, this will not affect your course of study). You will still receive your full payment for your participation in whichever parts of the study you have commenced, even if you withdraw before it finishes. Information and samples already gathered will be destroyed/disposed of.

A payment of £20 will be organised for each time you participate in a part of the study, totaling £120 in all. We also reimburse travel expenses within Zone 6 in London, on provision of receipts. Travel from outside this area needs to be agreed with the investigators in advance. Payment is by cheque (payable from Barts and the London School of Medicine and Dentistry): this may take 2-3 weeks; or may be made in cash on the

day, depending on the arrangements at the time. Payment is for undergoing the study and not for us taking and storing your blood. Payment is therefore not dependent on you agreeing to tissue retention (blood, urine and saliva samples).

5. **What will happen to me if I take part?**

You will be invited to visit the Centre for Clinical Pharmacology and the study will be explained to you. If you wish to take part you will be invited to sign the consent form. We will then interview you about your medical history (illnesses, medications, herbal products, smoking, alcohol, dental hygiene) to check that you are healthy and well. You will need to come to the centre on a further 6 occasions, which will be booked in at the beginning of the study. The basic study is summarized below:

Visit 1 (~1.5h)
Blood, urine and saliva sample collected.
Blood pressure measured every 15min for 1 hour.
Cantharidin-soaked 1cm^2 filter paper applied to inside of forearm, back or leg depending on preference and suitability of sites.

Visit 2 (~0.5h)
Cantharidin-soaked 1cm^2 filter paper applied adjacent to first site.

Visit 3 (~1.5h)
Blood pressure measured every 15min for 1 hour.
Blister fluid sampled (by carefully piercing of the side of the blister and collection of the fluid contained within the blister)
Blood, urine and saliva sample collected.

At this point, you will be randomised to receive a daily dose of potassium nitrate capsules (or potassium chloride placebo) that you will need to take first thing in the morning at home for at least 4 days. We can give you up to 6 days depending on when you are able to attend for your following visits.

The potassium nitrate and potassium chloride capsules appear identical and will be labelled either “Compound A for study” or “Compound B for study”. Neither you, nor the investigator performing the study, will know which is which.

After taking the capsules at home for at least 1 day, you will return for ***visits 4, 5 and 6*** that are the same as visits 1, 2 and 3 but cantharidin will be applied to the opposite arm, leg or different part of your back and you will continue to take the capsules daily in the morning at home until the end of visit 6.

Over the entire study, you will 4 small skin blisters produced by the cantharidin. Blood samples (~7.5ml, a teaspoonful) will be taken a total of 4 times. Hence ~30ml of blood will be taken during the entire study. Blood samples may be stored for up to three years to perform chemical and enzymatic studies, although

most analyses will be performed within the days and weeks of collection and once they have been completed the samples will be destroyed.

In total, the study will take a minimum of 10 days, though there are only 6 visits with the longest being only ~1.5h in duration.

By agreeing to tissue storage, you will only be giving us permission to store your blood for future research of the type described above. Any other use that we may want to make of your sample in the future will require approval by a Research Ethics Committee, which is an independent panel of experts who assess all research projects for safety, ethical acceptability and who protect volunteers' interests. None of the work that we envisage will have any direct implications for your personal health. You do not have to agree to the storage of your tissue. You are free to decide not to participate.

6. **What do I have to do?**

We need to know whether you are taking any medication or if you have had any reactions to drugs in the past as this also may exclude you from the study. If you have high blood pressure you will not be able to take part.

The day before your study visits that include the collection of blood samples (i.e. 4 times in total) we would ask you to fast after your evening meal (except water) until you are finished with us the following morning. We will provide tea / coffee and toast if you require after each study. You should avoid out of the ordinary strenuous exercise on the day before the studies. If you feel unwell (e.g. have a cold, fever, sore throat, headache) you should inform the researcher. If you are a blood donor you should refrain from giving blood from one week before you start the study until one week after you have finished the study.

We also need to know whether you are pregnant. There may be a risk that taking part in this trial might harm an unborn child. Women of childbearing age in which the possibility of being pregnant cannot be otherwise excluded must have a negative pregnancy test before their participation begins and this will be done by the investigator. Women who are not pregnant must use an effective form of barrier contraception. If you suspect that you may have become pregnant during the study in which you are taking part, you must immediately inform the investigator who will advise and help you. If necessary, you will be referred to other clinicians and counsellors for specialist care.

7. **What is the drug that is being tested?**

You will be randomised to receive either the test substance, potassium nitrate or potassium chloride placebo for ~5 days. Nitrate is present in our foods (especially in vegetables) and we also produce it ourselves in our body. We are able to metabolise nitrate, in our mouths, to nitrite. It has been shown that nitrite is able to lower blood pressure and reduces inflammation in the kidney and blood vessels in animal models of human diseases. The placebo control in this case is potassium chloride, the same as the main ingredient in 'Lo-Salt' table salt.

8. **What are the possible disadvantages and risks of taking part?**

The techniques used in this study are essentially safe and widely practised. You may find the capsules cause mild stomach upset in taken without food and hence we will give you breakfast and lunch on the days when you attend the clinic and will advise you to take the capsules with food. You may have pain and / or bruising after blood-taking.

The cantharidin blisters are similar to those caused by heat or friction (such as from wearing shoes). Cantharidin may cause some mild discomfort or itch whilst the blister is forming. Once the blister is formed and has had the fluid inside sampled, all care will be taken to keep the blister-roof (as healing is aided by its presence) and the blisters will be dressed by trained healthcare professionals (a doctor or a nurse). We will provide advice on how to look after your skin after the end of the study and provide any further dressings as needed. In previous studies using this technique, there have been **no** permanent consequences, such as scars, and any pigmentation that has occurred after healing has usually disappeared by 16 weeks.

It is possible that if these treatments are given to a pregnant woman they will harm the unborn child. Pregnant women must not therefore take part in this study; neither should women who plan to become pregnant during the study. Women who are at risk of pregnancy will be asked to have a pregnancy test before taking part to exclude the possibility of pregnancy. Women who could become pregnant must use an effective contraceptive during the course of this study. Any woman who finds that she has become pregnant while taking part in the study should immediately tell her research doctor.

Future insurance status (e.g. for life insurance or private medical insurance), could be affected by taking part in this study (if for example we find that your blood pressure is raised.) If you have private medical insurance you should check with the company before agreeing to take part in the trial. You will need to do this to ensure that their participation will not affect your medical insurance.

If we find that you have a high blood pressure reading, we will tell you this and what it could mean and offer to write a letter to your GP and suggest you go to see your GP for this to be followed-up.

9. **What are the possible benefits of taking part?**

This study will not benefit you directly.

10. **What if something goes wrong?**

The chances of any harm befalling you in this study are very small indeed. However, Queen Mary University London (QMUL) have provided indemnity/insurance against negligent harm and if you are harmed through fault on the part of the investigators, you will be entitled to compensation.

Queen Mary University London (QMUL) cannot provide indemnity/insurance for non-negligent harm. If you are harmed by taking part in this study and it is not the fault of the Investigators, then you may have grounds for a legal action but you may have to pay for it.

Regardless of this, if you wish to complain, or have any concerns about any aspect of the way you have been approached or treated during the course of this study, then you should first of all discuss this with Dr Kapil, Dr Khambata, or Prof Ahluwalia. If you would rather discuss any problems with individuals independent of the study then at QMUL / Barts and the London NHS Trust you can contact Professor Mark Caulfield on 020 7882 3402.

11. **Will my taking part in this study be kept confidential?**

All the information that is collected about you during the course of the study will be kept strictly confidential and will be kept in a locked room and stored on a password-protected computer. Only the investigators will have access to the data. Any information about you that leaves the centre will have your name and address removed so that you cannot be recognised from it.

Your GP will be notified of blood pressure readings and blood results, if you agree.

12. **What will happen to the results of the research study?**

The results from each volunteer will be pooled to obtain average results, so that no individual or their results will be identifiable in any form of publication. If the research study yields interesting/positive results these will be presented as a talk (or poster) at a local meeting within the University of London, or a national meeting, or possibly a European or international meeting. The results would also be written up as a full paper and be submitted for publication in an international journal. These events would be most likely to occur 3-12 months after your participation in the study. If you would like a copy of the paper you can contact the researcher to send you one when it becomes available.

13. **Who is organising and funding the research?**

The research is being organised by the Centre of Clinical Pharmacology of Barts & The London NHS Trust / Queen Mary University London. It is being performed by doctors at this Hospital / University who have some time allocated to undertake research. The study is funded by the British Heart Foundation. None of the doctors will be paid for including you in this study.

14. **Who has reviewed the study?**

This study has been reviewed by the local Research Ethics Committee.

15. **Contacts for Further Information**

You will always be able to contact an investigator to discuss your concerns and/or to get help:

Dr Vikas Kapil, Clinical Pharmacology, Charterhouse Square, London EC1M 6BQ
Telephone number: 020 7882 5720 / 8931; email: v.kapil@qmul.ac.uk

or

Dr Rayomand Khambata, Clinical Pharmacology, Charterhouse Square, London EC1M 6BQ
Telephone number: 020 7882 5720

Alternatively, you may wish to speak someone who is independent of the study (not actively involved), but knows about the study and the techniques involved. We suggest:

Dr David Collier, Clinical Pharmacology, Charterhouse Square, London EC1M 6BQ
Telephone number: 020 7882 3415

Thank you for considering participating in this study!

You will be given a copy of the information sheet and a signed consent form to keep