

The NIHR Southampton Biomedical Research Centre (BRC) has a tight quality assurance system for the writing, reviewing and updating of Standard Operating Procedures. As such, version-controlled and QA authorised Standard Operating Procedures are internal to the BRC.

The Standard Operating Procedure from which information in this document has been extracted, is a version controlled document, managed within a Quality Management System. However, extracts that document the technical aspects can be made more widely available. Standard Operating Procedures are more than a set of detailed instructions; they also provide a necessary record of their origination, amendment and usage within the setting in which they are used. They are an important component of any Quality Assurance Framework, but in themselves are insufficient and need to be used and interpreted with care.

Alongside the extracts from our Standard Operating Procedures, we have also made available here an example Standard Operating Procedure and a word version of a Standard Operating Procedure template. Using the example and the Standard Operating Procedure template, institutions can generate their own Standard Operating Procedures and customise them, in line with their own institutions.

Simply offering a list of instructions to follow does not assure that the user is able to generate a value that is either accurate or precise so here in the BRC we require that Standard Operating Procedures are accompanied by face-to-face training. This is provided by someone with a qualification in the area or by someone with extensive experience in making the measurements. Training is followed by a short competency assessment and performance is monitored and maintained using annual refresher sessions. If you require any extra information, clarification or are interested in attending a training session, please contact Dr Kesta Durkin (k.i.durkin@soton.ac.uk).

This document has been prepared from Version 2 of the BRC Standard Operating Procedure for making up triglyceride emulsion for assessing pancreatic function. It was last reviewed in September 2015 and the next review date is set for September 2017. The version number only changes if any amendments are made when the document is reviewed.

NIHR Southampton Biomedical Research Centre

Procedure for making ^{13}C MIXED TRYGLYCERIDE EMULSION

BACKGROUND

There are a number of ways of assessing pancreatic function using non-invasive methods. One of these is to orally administer a substrate that is labelled with a tracer, which when ingested will be absorbed and subsequently oxidised so that the tracer may then be detected on the breath. The amount of tracer in breath then reflects pancreatic function.

The substrate used is a type of triglyceride where one of the carbon atoms of the fatty acids is modified to contain a stable isotopic tracer (^{13}C) – in this case, **2- ^{13}C -octanoyl-1,3-distearin** or ^{13}C mixed triglyceride (^{13}C -MTG). Triglycerides are a form of fat which occur naturally in the diet. In order to reflect the normal digestion of fat in food, the ^{13}C -MTG must be incorporated into a food matrix similar to how triglyceride is found in the diet normally. The substrate is then incorporated into a lipid emulsion with other ingredients and the final product resembles a milkshake. This also makes it more palatable for the subject to consume.

PURPOSE

The purpose of this document is to ensure the safe and accurate preparation of the emulsion containing ^{13}C -MTG. Several steps which must be carried out in the correct sequence are involved in making-up the emulsion. It is important to ensure that this is done consistently so the resulting emulsion is the same each time. Changes in the proportion of ingredients, their sequence of mixing and the temperature of the emulsion each can affect the way the substrate is handled in the gut.

SCOPE

This procedure applies to all personnel involved in making up the emulsion containing ^{13}C -MTG for non-invasive assessment of pancreatic function.

RESPONSIBILITIES

It is the responsibility of the person preparing this ^{13}C mixed triglyceride emulsion drink, to ensure that it is made up and administered to the participant, according to this procedure.

PROCEDURE

3.5g Olive oil
3g Sunflower oil
33.2g Milk powder (Marvel)
9g Glucose
4.5g Caster sugar
10g Strawberry Nesquik flavouring
22g Double cream
200 ml Bottled water
10mg ^{13}C -MTG/kg bodyweight of subject
2 slices of white bread and 10g butter to serve as toast
Weighing boats (x7).
Saucepan for use on the hob
Thermometer
Food blender
Large ceramic mug
Small ceramic mug to drink the emulsion from
Weighing scales
Large (400ml) Pyrex beaker
Small (200ml) Pyrex beaker
2 pipettes
Spatula

1. In the Nutrition kitchen on level C in the WTCRF, weigh out the required mass of **sugar (4.5g)**, **glucose (9g)**, **Nesquik (10g)**, **double cream (22g)** and **milk powder (33.2g)** to the nearest 100mg or 0.1g in separate weighing boats.
2. Transfer the oils using a plastic pipette (pastette). Weigh **olive oil (3.5g)** into a weighing boat and then press the TARE button and add **sunflower oil (3g)** into the same boat.
3. Place the weighing boat on the scales DO NOT PRESS TARE. Calculate the mass of ^{13}C -MTG required according to the subject's body weight and add this mass to the weighing boat. It should be 10mg ^{13}C -MTG/kg body weight. Record the final weight to the nearest mg. Do not touch/switch the scales off.
4. Fill up the saucepan from the cupboard in the kitchen with water from the hot water system by the sink and put onto the hob to heat until the water reaches a gentle boil. Try to keep at this state throughout the process.

5. Fill the larger of the two ceramic mugs with hot water from the hot water system and place the blender attachment in there to warm up.
6. Measure out 70ml of bottled water and put into the smaller (200ml) of the two Pyrex beakers then put this beaker with water in, into the saucepan to heat up. Make sure that the amount of water in the saucepan is appropriate so that the beaker can sit stably.
7. Measure out 100ml of bottled water and put into the larger (400ml) Pyrex beaker and put into the water bath.
8. Add the smaller of the two ceramic mugs into the pan (with the 400ml beaker in) and transfer the ^{13}C -MTG into the smaller ceramic mug in the water bath. It will melt. Reweigh the weighing boat by putting back on the scales and record the weight of the empty boat. The amount of ^{13}C -MTG dispensed will be the difference between the weight of the boat with ^{13}C -MTG boat before weighing out the ^{13}C -MTG and the empty weighing boat after tipping the ^{13}C -MTG into the mug in the pan.
9. Add the sugar and glucose to the smaller (200ml) Pyrex beaker
10. When the ^{13}C -MTG has melted, add the oils into this smaller ceramic mug, using a pastette.
11. Add the double cream and wait for the temperature of the mixture to increase.
12. Add the milk powder to the smaller (200ml) beaker and mix with the blender whilst keeping the beaker in the pan.
13. Replace the blender to the insulated mug with the hot/warm water in. Do not worry about loss of mixture on the blender.
14. Add the mixture in the smaller (200ml) Pyrex beaker into the ceramic mug still in the pan, and blend for 2 minutes with the blender, keeping the mug immersed in the hot water.
15. Lift the blender out and hold over the mug whilst you put in the Nesquik.
16. Blend in the beaker for 30 seconds, whilst the mug is kept the pan.
17. Rinse the remainder from the blender into the emulsion with approximately 50ml of bottled water from the larger (400ml) Pyrex beaker (which should be in the pan and should contain 100ml of bottled water). This should be achieved by pouring the 50ml of water onto the blender attachment whilst held over the mug. Allow time for the contents to cool slightly (see point 19).
18. During this period, make two slices of white toast spread with 10g butter.
19. Allow time for the contents of the small ceramic mug to cool slightly – do not allow the temperature of the drink to fall too much. It should be taken to the participant at a temperature for them to sip the drink (as if you would a tea, coffee or hot chocolate – no lower than 70°C. A yellow thermometer can be found in the cupboard should you wish to check this).
20. Take the emulsion, toast and remaining 50ml hot water in the larger (400ml) Pyrex beaker to the subject. (The larger Pyrex beaker should now contain the remaining, approximately 50ml of bottled water).
21. Ask the participant to eat one of the slices of toast.

22. Ask them then to drink the Nesquik-flavoured drink
23. Using a stopwatch, keep a note of the time it takes the subject to drink the emulsion.
24. Pour the remaining 50ml hot water from larger (400ml) Pyrex beaker into the mug and swirl around.
25. Give the washings to the subject to drink.
26. When all the emulsion has been ingested, give the subject the second piece of toast to eat.