END OF STUDY REPORT

07/DEC/2015

The effect of probiotics on the incidence of spontaneous bacterial peritonitis in patients with cirrhosis and ascites

Protocol Number	Version No: 4.0, Date: 2012/03/14
Chief Investigator	Dr Martin James
EudraCT Number	2010-022886-92
REC Reference Number	10/H0405/81
Sponsor Reference Number	10GA021
Internal Reference No	MWJ NDDC BRU ascites study
Study Start Date	05/Dec/2012
Study End Date	17/Oct/2014
Funder(s)	NUH Charities & VSL#3 pharmaceuticals
Sponsor(s)	Nottingham University Hospitals NHS Trust

Name of Test Drug/Investigational Product	VSL#3 probiotics Co-trimoxazole 960mg once daily antibiotics
Indication Studied	Primary prevention of spontaneous bacterial peritonitis in cirrhosis

Report Author: Signature
Dr Martin James, Chief Investigator

Date: 14 – DEC – 2015
DD-MMM-YYYY

Sponsor Authorisation: Signature
Dr Maria Koufali, Deputy Director of R&I

Date: 22/12/2015
DD-MMM-YYYY

Dr. Brian Thomson
Director of Research and Innovation
Nottingham University Hospitals NHS Trust

This study was carried out in compliance with International Conference on Harmonisation (ICH) Good Clinical Practices (GCP) and Nottingham University Hospitals NHS Trust (NUH) Research and Innovation (R&I) Procedures

Contents

List of Abbreviations and Definition of Terms

AE	Adverse event
AR	Adverse reaction
BRU	Biomedical Research Unit
CHILD-PUGH class	Used to assess the prognosis of chronic liver disease
CI	Chief Investigator
CRA	Clinical Research Associate (Monitor)
CRF	Case Report Form
CRO	Contract Research Organisation
CT	Clinical Trials
CTA	Clinical Trials Authorisation
EC	Ethics Committee (see REC)
GCP	Good Clinical Practice
GP	General Practitioner
GTAC	Gene Therapy Advisory Committee
HVPG	Hepatic venous pressure gradient
IB	Investigators Brochure
ICF	Informed Consent Form
ICH	International Conference of Harmonisation
IEC	Independent Ethics Committee
IMP	Investigational Medicinal Products
IRB	Independent Review Board
MELD score	Model for end-stage liver disease
MHRA	Medicines and Healthcare products Regulatory Agency
NIHR	National Institute for Health Research
NRES	National Research Ethics Service (previously known as COREC)
NUH	Nottingham University Hospitals NHS Trust
PI	Principal Investigator
PIL	Participant/ Patient Information Leaflet
QP	Qualified Person
R&D	NHS Trust R&D Department
REC	Research Ethics Committee
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SBP	Spontaneous bacterial peritonitis
SIL	Subject Information Leaflet (see PIL)
SmPC/SPC	Summary of Products Characteristics
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reactions
TMF	Trial Master File
TSG	Oxford Radcliffe Trust / University of Oxford Trials Safety Group
UKELD	UK model for end-stage liver disease
WCC	White cell count

1. Summary of Study

Seventeen patients with cirrhosis and ascites were consented, with ten of those randomised, at Nottingham University Hospitals NHS Trust (NUH). Patients had histological, radiological or validated non-invasive tests (e.g. transient elastography) suggesting cirrhosis. Patients without prior history, but who were high risk of spontaneous bacterial peritonitis (Child-Pugh class ≥B or MELD >12 or hyponatraemia (Na<130) or ascitic albumin <15g/dL) were included. Baseline laboratory testing of ascites and blood were collected aseptically and baseline faecal bacterial quantification was performed. Patients were then randomly allocated to take 2 sachets' of VSL#3 probiotic (containing 8 types of lactic acid bacteria and bifidobacteria; Ferring® pharmaceuticals; 900 billion bacteria), co-trimoxazole 960mg or placebo each day for 12 months. Repeat sampling of ascites, blood and faeces were taken after 4, 12, 24 and 48 weeks. Demographic data was collected and the severity of hepatic dysfunction quantified by calculating MELD and Child-Pugh severity scores and by performing baseline and 6 month hepatic venous pressure measurements (HVPG). Spontaneous bacterial peritonitis (SBP) rates in untreated patients after 12 months is 60%, which is reduced to 7% with primary antibiotic prophylaxis (*Fernandez 2007*); to detect a 50% absolute reduction in clinical episodes of SBP compared to placebo, with 80% power at the α level 0.05, it was calculated that 17 patients were required in each group (51 patients in total).

The challenges to recruitment were the demographics and comorbidities of the target population, often having patients by their nature, with advanced liver disease, months to live and frequently with a strong or ongoing alcohol history as the cause of their liver disease. This patient group often declined involvement in the study and the study clearly under recruited, despite considering for inclusion over 330 patients with ascites (through an automated system where any patient having ascites samples analysed in the NUH laboratory were notified to the clinical PI who was part of the clinical team treating these patients).

During the conduct of the study, several issues arouse with regards IMP supply. VSL#3 pharmaceuticals changed the manufacturer of the placebo and active VSL#3 sachets used for the trial, this caused a delay in product supply and delivery which resulted in the withdrawal of 3 participants and prevented the randomisation of 2 participants. The IMP was then shipped to NUH in December 2013; however the IMP remained in quarantine in clinical trials pharmacy pending QP release certification to be issued from the manufacturer. Despite numerous requests to the manufacturer over the course of 9 months, the QP release certificate was never received at NUH; as such the decision was taken by the sponsor and CI to close the trial.

2. Objectives

Primary Objective: To investigate whether oral probiotics (VSL#3 sachets) or antibiotics (co-trimoxazole) reduce liver-related mortality and morbidity in patients with cirrhosis and ascites.
Secondary objective: To investigate the effect of probiotics or antibiotics on the incidence of SBP, non-SBP sepsis, portal pressure and variceal haemorrhage, encephalopathy. To investigate which long-term primary prevention strategies affect the incidence of *C. difficile* infection.
These objectives were not achieved as recruitment was suspended for the reasons outlined in section 1 above. No additional objectives were added during the study period.

3. Ethical Review

The study was given a favourable opinion by the Local Research Ethics Committee (LREC), NRES Committee East Midlands-Derby, and all amendments were also approved, with amendment dates listed in the front of each updated protocol.

4. Investigational Plan

Patients with cirrhosis and ascites were recruited. Patients had histological, radiological or validated non-invasive tests (e.g. transient elastography) suggesting cirrhosis. Patients without prior history, but who are high risk of SBP, (Child-Pugh class ≥B or MELD >12 or hyponatraemia (Na<130) or low ascitic albumin <15g/dL) were included.

5. Selection of Study Population

Inclusion Criteria

- Participants willing and able to give informed consent for participation in the study.
- Male or Female, aged 18 years or above.
- Diagnosed with required disease/severity/symptoms.
- Stable dose of current regular medication (e.g. diuretics, beta-blockers, vitamin supplementation) for at least 4 weeks prior to study entry.
- Female participants of child bearing potential and male participants whose partner is of child bearing potential must be willing to ensure that they or their partner use effective contraception during the study and for 3 months thereafter.
- Participants have clinically acceptable laboratory tests and ECG within 14 days of enrolment.
- Able (in the Investigators opinion) and willing to comply with all study requirements.
- Willing to allow their General Practitioner and consultant, if appropriate, to be notified of participation in the study.

Exclusion Criteria

The participant could not enter the study if ***ANY*** of the following applied:

- Female participants who were pregnant, lactating or planning pregnancy during the course of the study.
- Presence of hepatocellular carcinoma.
- Scheduled elective surgery or other procedures requiring general anaesthesia during the study.
- Participant who was terminally ill.
- Any other significant disease or disorder which, in the opinion of the Investigator, may have either put the participants at risk because of participation in the study, or may have influenced the result of the study, or the participant's ability to participate in the study.
- Use of antibiotics or probiotics in the last 2 weeks.

- Known hypersensitivity to trimethoprim, sulphonamides or any other ingredients in co-trimoxazole tablet.
- History of acute porphyria or serious haematological disorder.
- Participants who have participated in another research study involving an investigational product in the past 12 weeks.

6. Study Settings

Patients were screened in several places throughout NUH. This included in-patient general admission or specialty wards, out-patient hepatology clinics or the medical day case unit for those patients attending for therapeutic paracentesis. Patients were also screened using the facilities of the Nottingham NIHR Biomedical Research Unit. The study team consisted of chief/principal investigator (Dr Martin James) who wrote and submitted the study protocol, screened and recruited patients and conducted study procedures and study visits including physical examinations and ascites and blood sampling, and research nurses (Mina Patel, Andrea Bennett and Lisa Mannion) who co-ordinated study visits and took blood and urine samples. NUH clinical trials pharmacy dispensed the trial IMP.

7. Interventions

The study medications were:

1. VSL#3 probiotic preparations. Each packet of VSL#3® contained 450 billion live lactic acid bacteria and bifidobacteria. There are 8 different strains of bacteria contained in VSL#3: Streptococcus thermophilus, Bifidobacterium breve, Bifidobacterium longum, Bifidobacterium infantis, Lactobacillus acidophilus, Lactobacillus plantarum, Lactobacillus paracasei, Lactobacillus delbrueckii subsp. Bulgaricus. *Other ingredients included m*altose and silicon dioxide.

   The prescribed dose was 2 sachets (containing 900 billion bacteria) orally each day. Sachets were plain white and indistinguishable from placebo sachets, but labelled with a study/code number, and provided by VSL#3 pharmaceuticals. Probiotics and placebo sachets should have been opened and the contents stirred into cold water or another non-fizzy cold drink (but not with hot drinks or hot food). Study patients were prohibited from taking any other probiotics during the study (such as Yakult®).

2. Cotrimoxazole 960mg orally each day (two 480mg tablets). Cotrimoxazole is a combination of sulfamethoxazole (800mg) and trimethoprim (160mg). This was supplied by NUH clinical trials pharmacy and is the standard dose for prophylaxis of spontaneous bacterial peritonitis in clinical practice. Cotrimoxazole antibiotics were to be taken with food or any drink.

3. VSL#3 Placebo. This was two placebo sachets identical to VSL#3 active sachet.

The study medications were supplied as matched appearance sachets for active and placebo VSL3 and as open label cotrimoxazole tablets. Tablets and sachet counts were confirmed with patients at each study visit to document compliance.

8. Changes in the Protocol from Initial Approval

The amendment history was detailed at the start of each protocol and is outlined in the table below.

Amendment No.	Protocol Version No.	Author(s) of changes	Details of Changes made
20100512	1	MW James	Original
20100811	2	MW James	Updated study flow chart and dosing schedule.
20110408	3	MW James	Updated trial duration 48 weeks Refined study design (placebo/VSL#3/co-trimoxazole) Updated discontinuation criteria
20120314	4	MW James	**7 & appendix B** Adjustment to blood volume collection at each visit (20ml stored). Inclusion of baseline& 3 month urine test **9.1.5** Clarification of expected adverse events/reactions

9. Protocol Deviations

The deviation log can be found in appendix 1 and mostly relate to the problems encountered with ongoing supply of the IMP from VSL#3 pharmaceuticals and failed QP certificate release. This meant that any product that had been supplied to the study team at NUH could not safely be prescribed to the three patients who remained active on the study. Patients were therefore subsequently withdrawn and treated according to best clinical care in the hepatology out-patients.

10. Patient Information & Consent

Written informed consent was obtained prior to trial procedures occurring. Each participant was requested to personally sign and date the latest approved version of the informed consent form (appendix 2: participant consent form) before any study specific procedures were performed.

Written and verbal versions of the participant information (appendix 3: participant information sheet) and informed consent were presented to the participants and detailed no less than: the exact nature of the study; the implications

and constraints of the protocol; the known side effects and any risks involved in taking part. It was clearly stated that the participant was free to withdraw from the study at any time for any reason without prejudice to future care, and with no obligation to give the reason for withdrawal.

The participant was allowed as much time as wished to consider the information, and the opportunity to question the Investigator, their GP or other independent parties to decide whether they would participate in the study. Written Informed Consent was then obtained by means of participant dated signature and dated signature of the person who presented and obtained the informed consent. The person who obtained the consent was suitably qualified and experienced, and authorised to do so by the Chief Investigator. A copy of the signed Informed Consent Form was given to the participants and the original signed form was retained at the Investigator Site File at the study site.

11. Randomisation

Subject numbers were assigned sequentially as each subject entered the study. The subjects were assigned study drug through a randomisation schedule based on the randomisation plan. The study drug was labelled with the study number and unique identification number. The three treatments: VSL#3 probiotics, antibiotics and placebo with tablets or sachets were administered as appropriate as follows:

Study medication	Sachets	Tablets
VSL#3	2 active sachets*	-
Cotrimoxazole 960mg	-	960mg (2 x 480mg)
Placebo	2 dummy sachets*	-

* All probiotic/placebo sachets appeared exactly the same

In the event of an emergency, the investigator was to decide the necessity of unblinding the subject's treatment assignment. The blinded treatment assignments were accessible to the investigator should a subject need to be unblinded in an emergency using the unblinding envelopes provided to the hospital pharmacy. Unblinding did not occur, however if unblinding was to occur, the investigator or study pharmacist would have recorded the reason for unblinding, as well as the date and time of the event. Corresponding information would have been recorded on the CRF by the investigator.

12. Safety Reporting

All adverse events (AEs) occurring during the study observed by the investigator or reported by the participant, whether or not attributed to study medication, were recorded on the CRF.

The following information was recorded: description, date of onset and end date, severity, assessment of relatedness to study medication, other suspect drug or device and action taken. Follow-up information was collected as necessary.

All AE's reported are listed in appendix 4. AEs considered related to the study medication as judged by a medically qualified investigator or the sponsor were followed until resolution or until the event was considered stable. All

related AEs that resulted in a participant's withdrawal from the study or were present at the end of the study, were followed up until resolution.

It was left to the investigator's clinical judgment whether or not an AE was of sufficient severity to require the participant's removal from treatment however a participant could also voluntarily withdraw from treatment due to what he or she perceived as an intolerable AE. If either occurred, the participant was required to undergo an end of study assessment and be given appropriate care under medical supervision until symptoms ceased or the condition became stable.

The severity of events was assessed on the following scale: 1 = mild, 2 = moderate, 3 = severe.

The relationship of AEs to the study medication was assessed by a medically qualified investigator.

Any pregnancy occurring during the clinical study and the outcome of the pregnancy was to be recorded and followed up for congenital abnormality or birth defect. There were no reported pregnancies during the study.

Reporting Procedures for Serious Adverse Events

This was according to Nottingham University Hospitals NHS Trust (sponsor) R&I reporting requirements, and in accordance with Directive 2001/20/EC. The SAEs reported during the study period are listed appendix 5.

13. Statistical Analysis

Patient outcomes were planned to be compared between placebo and active therapy (either VSL#3 probiotics or co-trimoxazole antibiotics). Variables would be compared using Students *t*-test or Chi-square test with Yates correction where appropriate. Probability curves would be generated using Kaplan-Meier methods and compared using log-rank testing.

14. Main Findings of the Study

Patients who were having ascitic samples taken to investigate the cause or to exclude infection (SBP) were considered for entry into the study. 334 patient's records were reviewed by the PI or research nurse to consider eligibility. Of those that were approached or given the patient information sheet, seventeen consented to be screened for the study. Ten were eligible and randomised to start treatment; however one patient had a baseline ascitic sample that demonstrated active infection so was withdrawn on the day of randomisation and the infection treated appropriately.

A summary of the visit schedule for participants that provided written informed consent for the trial is outlined in appendix 6 and includes the date of consent, date of randomisation, date of each study visit attended, date of withdrawal and reason for withdrawal.

- Of the 17 patients who provided written informed consent, only 10 were randomised.
- Of the 10 patients randomised;
  - 1 participant demonstrated active infection so was withdrawn on the day of randomisation.
  - 1 participant reached visit 2 (baseline).
  - 2 participants reached visit 3 (week 4).
  - 2 participants reached visit 4 (week 12).
  - 4 participants reached visit 5 (week 48).

- 0 participants reached visit 6.

Appendix 7 details participant demographics (age and sex), treatment group, CHILD-PUGH Class, MELD score and White Cell Count (WCC) at each study visit attended by the participant and whether the participant met the primary or secondary endpoints.

Of the 10 patients randomised to study treatment;

- 4 participants met the primary end point criteria.
  - Death from liver failure.
  - Hospital admission for liver related complications; admission for encephalopathy (intracerebral haemorrhage) and ascites.
  - Hospital admission for liver related complication; admission for liver transplant.
  - Hospital admission for liver related complication; admission for episode of melaena and haematemesis. Liver disease deterioration-for palliative care only.
- 1 participant met the secondary end point criteria.
  - Patient diagnosed with *C. difficile* infection and pleural effusion.
- 3 participants were withdrawn due to a lack of IMP.
- 1 participant was withdrawn as they did not attend their final appointment and informed the study team that they had not been taking the study medication.
- 1 participant demonstrated active infection so was withdrawn on the day of randomisation.
- No participants attended for the final visit (week 48).

The study was terminated before completion; this was primarily due to a lack of QP release certificate being issued by the manufacturer and despite numerous attempts by the sponsor and chief investigator to acquire the documentation from the manufacturer the decision was taken to close the study in October 2014. These issues were beyond the control of the principal investigator and study team. Insufficient patients were recruited and the participants that were still ongoing at the time these problems occurred were withdrawn due to a lack of IMP supply on 12-Dec-2013 and were treated according to best clinical practice.

15. Conclusions

Only 10 of the required 51 participants were randomised, and only 5 of the 10 recruited met the primary of secondary endpoints and therefore completed the trial. Consequently given the split between the 3 treatment groups (VSL#3, placebo and contrimoxazole) no conclusions can be drawn regarding strategies for primary prevention of spontaneous bacterial peritonitis in the light of this study, which was terminated prematurely with too few patients randomised, resulting in insufficient power. This study was terminated for the reasons explained, and did not recruit the numbers required to complete or write up the study in full. There are no immediate plans to conduct further studies in this area.

16. Arrangements for Disseminating Findings

If the study had continued and recruited and completed successfully, the study findings would have been disseminated through submission of study outcomes to national and international hepatology conferences, followed by submission of the manuscript to relevant peer-reviewed hepatology journals.

17. Appendices

1. Log of protocol deviations
2. Participant information sheet
3. Participant consent forms
4. Log of adverse events
5. Log of serious adverse events
6. Participant visits
7. Participant Data

Appendix 1: Log of Protocol Deviations

Breach Number	Date Breach Identified	Subject Identifier *(if applicable)*	Brief Description of Breach	Date Breach Reported to Sponsor
1	12/Dec/2013	AD0006 TD0007 DC0008	Three participants that were on active treatment had their treatment terminated early due to a lack of drug supply.	18/Dec/13
2	30/May/2013	RW0003	Clinical care blood results that were taken on 14/Jun/2012 and used to screen for study inclusion were outside of the defined time of 14 days as stated on the protocol.	11/Aug/2015
3	12/Oct/2015	N/A	Deviation to sponsor SOP-RES-019_Adverse Event Reporting in Clinical Trials of Investigational Medicinal Products, as adverse events have been logged within the CRF rather than the AE Record.	17/Nov/2015
4	17/Nov/2015	DC0012	SAE report 5000-09 was not submitted within 24 hrs of identification by the site	26/Nov/2015
5	21/Aug/2014	LH0015	Patient was consented to the study but did not commence treatment and was withdrawn from the study due to a lack of IMP.	26/Nov/2015
6	21/Aug/2014	AM0016	Patient was consented to the study but did not commence treatment and was withdrawn from the study due to a lack of IMP.	26/Nov/2015

Appendix 2: Participant Consent Form

Nottingham University Hospitals NHS
NHS Trust

NDDC Nottingham Digestive Diseases Centre
BRU Biomedical Research Unit

Digestive Diseases and Thoracics Directorate
Queen's Medical Centre Campus
Derby Road
Nottingham
NG7 2UH

Tel: 0115 924 9924 ETX: 841147
Email: martin.james@nuh.nhs.uk

CONSENT FORM
(Version 4.1 Date 15/05/12)

Title of Study: A trial of probiotics on the incidence of spontaneous bacterial peritonitis (*infection in abdominal fluid*) in patients with cirrhosis and ascites

REC ref: **10/H0405/81**
Eudract ref : **2010-022886-92**

Name of Researcher: Dr Martin James

Name of Participant: **Please initial box**

1. I confirm that I have read and understand the information sheet version number 4.0 dated 14.03.2012 for the above study and have had the opportunity to ask questions. ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time, without giving any reason, and without my medical care or legal rights being affected. I understand that should I withdraw, information collected may still be used in the project analysis. ☐

3. I understand that relevant sections of my medical notes and data collected in the study may be looked at by authorised individuals from the University of Nottingham, the research group and regulatory authorities where it is relevant to my taking part in this study. I give permission for these individuals to have access to these records and to collect, store, analyse and publish information obtained from my participation in this study. I understand that my personal details will be kept confidential. ☐

4. I understand and agree at 4, 12, 24 and 48 weeks during my course of treatment I will return to Queen's Medical Centre where further samples will be taken for analysis. ☐

5. I agree to my GP being informed of my participation in this study. ☐

6. I agree to take part in the above study. ☐

Nottingham University Hospitals NHS
NHS Trust

Name of Participant	Date	Signature
Name of Person taking consent (if different from Principal Investigator)	Date	Signature
Name of Principal Investigator	Date	Signature

3 copies: 1 for participant, 1 for the project notes and 1 for the medical notes

Nottingham University Hospitals NHS
NHS Trust

NDDC Nottingham Digestive Diseases Centre
BRU Biomedical Research Unit

Digestive Diseases and Thoracics Directorate
Queen's Medical Centre Campus
Derby Road
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Tel: 0115 924 9924 ETX: 63443
Email: martin.james@nuh.nhs.uk

Patient Information Sheet

A trial of probiotics on the incidence of spontaneous bacterial peritonitis (*tummy fluid infection*) in patients with cirrhosis and ascites

You are being invited to take part in a research study. Before you decide it is important for you to understand what it is about and what it will involve. Please read this sheet carefully and discuss it with friends, relatives or your GP if you want to. You can ask any questions about bits that you don't understand.

What is the purpose of this study?

Cirrhosis (scarring) of the liver can lead to tummy swelling from fluid. This fluid can become infected, and may be serious (even fatal) in some patients.

We would like to invite you to help us test a new way of stopping infection in this fluid, by using "probiotics".

When people take probiotics ("good" or "healthy" bugs) less harmful bugs are inside them. This may stop infection in the surrounding fluid.

We want to test whether these probiotics work as well as antibiotics. If they do, they may be safer to use than antibiotics.

Why have I been chosen?

You are being invited to take part because you have liver scarring and tummy fluid.

Do I have to take part?

No, taking part is up to you. If you do decide to take part we will ask you to sign a consent form and give you a copy of this information sheet to keep.

If you decide to take part you can withdraw at any time.

If you decide not to take part you do not have to give a reason, nobody will be upset and the care we still give to you will not be affected.

What will I be asked to do if I take part?

To begin with, we will check you over carefully to make sure it is safe for you to take part in this study. This will include blood pressure tests and a heart trace.

Samples of your blood, tummy fluid, faeces and urine will be taken. Your doctor may also have planned to take a liver biopsy sample as part of your clinical care.

You will receive either probiotics ("good bugs"), antibiotics (called co-trimoxazole) or placebo (a dummy sachet) for one year. You have an equal chance of each of these three treatments.

After 4, 12, 24 and 48 week's treatments, you will return to Queens's Medical Centre. We will let you know when to come. More samples (about 20mL, or four-teaspoons-full) of your blood, tummy fluid, faeces and urine will be taken. Urine collection for 24 hours will also take place at baseline and after 3 months.

You may need to come to hospital for other visits, to help look after you.

What are my responsibilities?
During the study you would need to take the treatment every morning. If you do forget, take it at lunchtime or later in the day.

Probiotics should be drunk after stirring the contents of 2 sachets into a cold drink such as water or orange juice (but not fizzy or hot drinks and not with hot food).

The antibiotics should be taken with any drink.

You should not be taking any other probiotics during the study (such as Yakult®). If you are prescribed any other tablets during the study then the study doctor should be told.

When you come back for your study visits please bring your study medication with you so that we can check it and give you a fresh supply.

What other treatments are available?
Treatment for tummy fluid can be with water tablets (diuretics) and by being careful with salt. These may still be used even if you agree to take part.

What are the possible side-effects of taking part?
The antibiotic co-trimoxazole can cause headache, rash, diarrhoea, pain in the muscles and feeling sick. Any new problems with treatment such as rash should be reported to the doctor at once.

Side effects from probiotics are rare, but if they do occur they tend to be mild and digestive like gas or bloating.

What are the possible disadvantages and risks of taking part?
You will come for regular hospital visits after 4, 12, 24 and 48 weeks during your treatment, which will take time out of your day. In patients like you, we would be seeing you regularly anyway.

This medicine should not be used during pregnancy or breastfeeding as it may be harmful to the baby. If you are pregnant or plan to become pregnant during the study you should not take part. To be sure, we will do a pregnancy test. You should also use effective contraception (like condoms) while taking part. If you become pregnant during the study you must tell the study doctor.

If you have private medical insurance, it may be affected by taking part and you should check with your insurance company before taking part.

If we discover that you have a medical condition that you did not know about before, we will tell you about it and discuss how this should be treated.

What are the possible benefits in taking part?
You will help us to understand better how to look after patients with liver scarring and tummy fluid better in the future.

What happens when the research study is finished?
If we feel that you should carry on with medication after the study ends, this will be prescribed for you.

What if something goes wrong?
There are no special compensation arrangements for this study. However, if you are harmed as a result of someone's negligence, then you may have grounds for a legal action for compensation, but you may have to pay your legal costs.

If you have concerns about any aspect of the way you have been treated during the study you can contact the hospital's Patient Advice and Liaison Service (PALS) on 0115 924 9924 EXT 65412 or 62301, minicom 0800 183 0204 or write to PALS, NUH NHS Trust, c/o PALS, Freepost, NEA 14614, Nottingham, NG7 1BR. If you wish to make a formal complaint, please write to Chief Executive, Dr Peter Homa at NUH NHS Trust, QMC campus, Derby Road, Nottingham, NG7 2UH.

What if there is an emergency?
If there is an emergency you should seek medical attention in the usual way; either through NHS direct, the emergency department or by contacting one of the study doctors (Dr Martin James 0115 9249924 Ext 63443).

Will my taking part in this study be kept confidential?
Your medical records may be examined by an authorized person from the clinical trial team or the regulatory authorities. This is to check the study is being carried out correctly. Your name will not be disclosed outside the hospital although with your permission, your GP will be informed you are taking part.

What will happen to the results at the end of the research study?
The results of this study will not be known until some time after the last patient has finished (in about one year's time). The doctor will let you know the results, and which treatment you were taking. The results may be reported in medical journals or meetings, but you will not be identified by name.

Who is organizing and funding the research?
This study is being designed and run by the Nottingham NIHR Biomedical Research Unit and funded by a charity research grant from Nottingham University Hospitals. The probiotic study medication and placebo (dummy) sachets are being funded by VSL#3 pharmaceuticals.

Who has reviewed the study?
The Nottingham Research Ethics Committee has approved this study.

What do I do now?
Thank you for considering taking part in this research. You will be contacted in a few days by one of the study investigators (doctor or nurse) when you can ask any questions you have and let us know whether or not you would like to take part

Nottingham University Hospitals NHS
NHS Trust

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Appendix 4: Log of Adverse Events

Participant Identifier	Event Summary	Date of Onset	Relationship to IMP
RW0003	Paracentisis*	25-Sep-12	Not related
RW0003	Paracentisis*	29-Aug-12	Not related
RW0003	Paracentisis*	28-Dec-12	Not related
RW0003	Paracentisis*	14-Dec-12	Not related
SK0008	Paracentisis*	15-Jan-13	Not related
SK0008	Paracentisis*	28-Dec-12	Not related
SK0008	Paracentisis*	18-Mar-13	Not related
AD0011	Paracentisis*	06-Feb-13	Not related
AD0011	Paracentisis*	02-May-13	Not related
AD0011	Tooth Extraction	05-Jul-13	Not related
DC0012	Acid Reflux	UNK	Possibly related
TD0013	Constipation	15-Feb-13	Unlikely related
JH0014	Abdominal discomfort and flatulence	12-Mar-13	Possibly related
DC0008	Blood transfusion	19-Dec-13	Not related

* Please note paracentesis was initially reported as an AE but given the frequent occurrence of this procedure in this population of patients, during the trial the decision was made by the sponsor and chief investigator that paracentesis would not need to be reported as an AE

Appendix 5: Log of Serious Adverse Events

Participant Identifier	Event Summary	Criteria	Date of Onset - Date Resolved	Relationship to IMP	Expected	Date Reported to Sponsor	Sponsor Reference	No Further Follow Up Required
MS0002	Jaundice	Hospitalisation or prolongation of hospitalisation	15/Jun/2012 to ongoing	Not related	☐ Yes ☐ No ☒ N/A	10/Jul/2012	5000-01	☒
MS0002	Hepatic encephalopathy	Hospitalisation or prolongation of hospitalisation	02/Jul/2012 to ongoing	Not related	☐ Yes ☐ No ☒ N/A	10/Jul/2012	5000-02	☒
MS0002	Alcoholic liver disease	Life threatening and Hospitalisation or prolongation of hospitalisation	29/Jul/2012 to ongoing	Not related	☐ Yes ☐ No ☒ N/A	01/Aug/2012	5000-03	☒
RW0003	Vasovagal Syncope	Hospitalisation or prolongation of hospitalisation	28/Jul/2012 to 29/Jul/2012	Not related	☐ Yes ☐ No ☒ N/A	15/Aug/2012	5000-04	☒
MA0004	Cerebral haemorrhage	Hospitalisation or prolongation of hospitalisation	21/Sep/2012 to ongoing	Not related	☐ Yes ☐ No ☒ N/A	28/Sep/2012	5000-05	☒
SK0005	Pleural effusion	Hospitalisation or prolongation of hospitalisation	22/Apr/2013 to 22/May/2013	Not related	☐ Yes ☐ No ☒ N/A	03/May/2013	5000-06	☒
AW0016	Hematemesis	Hospitalisation or prolongation of hospitalisation	04/Jul/2013 to ongoing	Unlikely	☐ Yes ☐ No ☒ N/A	09/Jul/2013	5000-07	☒

Appendix 6: Participant Visits

Participant Screening Number	Participant Randomisation Number	Date of Consent	Date of Randomisation	Visit 1 (screening)	Visit 2 (baseline)	Visit 3 (week 4)	Visit 4 (week 12)	Visit 5 (week 24)	Visit 6 (week 48)	Date withdrawn	Reason for withdrawal
JB0001	JB0001	13-Mar-12	13-Mar-12	13-Mar-12						13-Mar-12	Failed screening (ascitic sample diagnosed spontaneous bacterial peritonitis)
MS0002	MS0002	14-May-12	14-May-12	14-May-12	21-May-12	21-Jun-12				06-Aug-12	Death (06-Aug-2012) due to progression of decompensated liver disease
RW0003	RW0003	11-Jul-12	11-Jul-12	11-Jul-12	17-Jul-12	15-Aug-12	10-Oct-12	22-Jan-13		09-Jul-13	Withdrawn- patient was due to attend final (week 48) visit, however the patient was unable to attend. Patient stated that he has not been taking trial medication for the last 3 months.
PM0004	N/A	12-Jul-12	N/A	12-Jul-12						18-Jul-12	Failed screening (ascitic sample diagnosed spontaneous bacterial peritonitis)
MA0005	MA0004	11-Sep-12	12-Sep-12	11-Sep-12	11-Sep-12					12-Nov-12	Death (12-Nov-2012) due to intracerebral haemorrhage
WH0006	N/A	03-Oct-12	N/A	03-Oct-12						03-Oct-12	Failed screening (ascitic sample diagnosed spontaneous bacterial peritonitis)
SK0007	N/A	10-Oct-12	N/A	10-Oct-12						10-Oct-12	Unable to obtain ascitic sample, re-booked but participant forgot to attend
SK0008	SK0005	19-Dec-12	19-Dec-12	19-Dec-12	19-Dec-12	23-Jan-13	18-Mar-13			15-May-13	Withdrawn- met secondary endpoint
TH0009	N/A	16-Jan-13	N/A	16-Jan-13						16-Jan-13	Failed screening (ascitic sample diagnosed spontaneous bacterial peritonitis)
DB0010	N/A	23-Jan-13	N/A	22-Jan-13						22-Jan-13	Unable to proceed as patient was recruited onto a second interventional study
AD0011	AD0006	28-Jan-13	28-Jan-13	28-Jan-13	28-Jan-13	06-Mar-13	02-May-13	26-Jul-13		12-Dec-13	Withdrawn due to lack of drug supply
DC0012	DC0008	27-Feb-13	27-Feb-13	27-Feb-13	27-Feb-13	28-Mar-13	22-May-13	30-Aug-13		12-Dec-13	Withdrawn due to lack of drug supply
TD0013	TD0007	12-Feb-13	12-Feb-13	12-Feb-13	12-Feb-13	12-Mar-13	03-May-13	19-Jul-13		12-Dec-13	Withdrawn due to lack of drug supply
JH0014	JH0009	06-Mar-13	12-Mar-13	06-Mar-13	06-Mar-13	09-Apr-13	10-Jun-13			25-Jun-13	
AW0025	AW0010	08-May-13	03-Jun-13	08-May-13	03-Jun-13	01-Jul-13				08-Jun-13	Patient for palliative care
LH0015	N/A	21-Nov-13	N/A	21-Nov-13						12-Dec-13	Withdrawn due to lack of drug supply
AM0016	N/A	09-Dec-13	N/A	09-Dec-13						12-Dec-13	Withdrawn due to lack of drug supply

Appendix 7: Participant Data

Participant Screening Number	Participant Randomisation Number	Sex	Age	Treatment	Visit 1/2			Visit 3			Visit 4			Visit 5			Visit 6			Did the participant meet the primary endpoint?	Did the participant meet the secondary endpoint?
					CHILD – PUCH Class	Meld Score	WCC	CHILD – PUCH Class	Meld Score	WCC	CHILD – PUCH Class	Meld Score	WCC	CHILD – PUCH Class	Meld Score	WCC	CHILD – PUCH Class	Meld Score	WCC		
JB0001	JB0001	F	54	VSL#3	Failed screening															No	No
MS0002	MS0002	M	65	VSL#3	C	15	0.2	C	16	0.3										Yes- death from liver failure	
RW0003	RW0003	F	71	Cotrimoxazole	B	10	0.2	B	8	0.2	B	8	0.1	B	8	0.1				No	No
PM0004	N/A	M	51	N/A	Failed screening															No	No
MA0005	MA0004	M	47	Placebo	B	17	0.2													Yes-hospital admission for liver related complications; admission for encephalopathy (intracerebral haemorrhage) and ascites	No
WH0006	N/A	F	67	N/A	Failed screening															No	No
SK0007	N/A	F	63	N/A	Failed screening															No	No
SK0008	SK0005	M	70	Cotrimoxazole	B	12	0.2	B	13	0.1	B	14	0.1							No	Yes- patient diagnosed with C difficile infection and pleural effusion
TH0009	N/A	M	62	N/A	Failed screening															No	No
DB0010	N/A	F	59	N/A	Unable to proceed past screening															No	No
AD0011	AD0006	F	37	Placebo	C	16	0.2	C	13	0.2	C	8	0.2	C	6	N/D				No	No
DC0012	DC0008	M	49	VSL#3	B	11	0.3	B	9	0.4	B	11	0.2	B	10	0.3				No	No
TD0013	TD0007	M	53	Placebo	B	9	0.4	B	7	N/D	A	7	N/D							No	No
JH0014	JH0009	F	54	VSL#3	A	13	0.2	A	13	0.2	A	12	0.2							Yes- hospital admission for liver related complication; admission for liver transplant	
AW0025	AW0010	F	50	Placebo	C	16	0.1	C	UNK	0.1										Yes- hospital admission for liver related complication; admission for	

Appendix 7: Participant Data

												episode of malaena and haematemosis. Liver disease deterioration-for palliative care only	
LH0015	**N/A**	M	46	N/A	N/A	N/A	N/D					No	No
AM0016	**N/A**	M	57	N/A	C	15	0.3					No	No