Subject information for participation in medical research

Controlled hookworm infections

Research title: Study into the local immune responses in the skin after repeated hookworm infection

Introduction

Dear reader,

With this information letter we want to ask you if you want to participate in a medical study. Participation is voluntary. Here you can read what kind of research it is, what it means for you, and what the advantages and disadvantages are. It's a lot of information. Would you like to read through the information and decide whether you want to participate?

Ask your questions

You can make your decision with the information provided in this information letter. In addition, we recommend that you do this:

- Ask questions to the investigator who gives you this information.
- Talk to your partner, family or friends about this study.
- Ask questions to the independent expert, Dr. A.H.E. Roukens
- Read the information on www.rijksoverheid.nl/mensenonderzoek.

1. General information

This study is being done at the Leiden University Medical Center (LUMC). A maximum of 5 healthy volunteers are needed for this study. The study takes place in two parts: part 1 (the infection phase) and part 2 (the donor phase). We are looking for three participants for only part 1 (group A) and two participants for part 1 and part 2 (group B). The study is different in design and content for participants from group A and group B, these differences are further explained below. You can indicate your preference for the group in which you would like to participate in this study.

The Medical Ethics Review Committee Leiden The Hague Delft (MREC LDD) has approved this study. General information about the review of research can be found on the website of the Dutch government: www.rijksoverheid.nl/mensenonderzoek.

2. What is the purpose of this study?

The aim of this study is to investigate the immune response in the skin in hookworm infection. With this study, we want to find out which cells play a role in the immune response. This may allow new vaccines and treatments to be developed in the future.

In addition, we are looking for two volunteers who want to become donors for 2 years so that larvae can be grown from the faeces of these donors for follow-up study. This means that the donors will not be treated for the hookworm infection after the first phase of the study, but only at a later point in time.

3. What is the background to this study?

Worldwide, about 250 million people are infected with hookworms, mainly in Africa and Southeast Asia. Hookworm infections cause anaemia and protein loss, particularly in children, which prevents them from developing properly and can become malnourished. There is a medicine that can be used to treat hookworms well. Unfortunately, mass treatment is insufficient to eradicate the worm. This is because people often contract the infection again immediately after treatment. A vaccine can prevent this. However, it is not yet known how to induce good protection against hookworm infection. Hookworms do not occur in animals, so animal models cannot be used to test a vaccine or learn more about protection against infection. A hookworm larva infects humans through the skin and moves to the lungs, after which it is coughed up and ends up in the intestines. Research has already been done into the response of the immune system to the hookworm infection in the lungs and intestines, but little is known about what happens in the skin. This will be studied in this study. The new information provided by this study may help develop a vaccine.

In order to continue researching hookworm infections, we need healthy volunteers who can be donors of faeces from which larvae can be bred. With these larvae, new volunteers can be infected and new vaccines or medicines can be developed or tested.

4. How will the study be carried out?

How long will the study take?

The study consists of two groups. Group A will only participate in the first part of the study, of a total of 13 weeks. Group B consists of two parts: part 1 for 16 weeks and part 2 for a maximum of two years.

If you participate in the study, you will go through the first part of the study anyway. In addition, you also have the choice to participate in part 2 of the study, the group B. This will be discussed with you at the screening visit. After the screening, you will make a choice to participate in group A or group B. However, during the study you can decide not to participate in group B anymore, or you can indicate that you still want to switch from group A to group B. If there are more than two participants for group B, a choice will be made by the investigators.

First step: the screening visit

The screening visit will take about an hour and a half. We first want to know if you are suitable to participate. That is why the investigator is doing some checks:

- Conversation about your medical history, during which the investigator asks questions about your health, use of medication and health complaints relevant to the study, etc.

- Physical. For example, the investigatorr listens to your heart and lungs and measures your blood pressure and heart rate.
- Blood test. To do this, the investigator will take some blood from you. We test for HIV, Hepatitis B and Hepatitis C, among others. We will tell you if you have any of these diseases.
- Urinalysis for the use of drugs, such as cocaine, amphetamine and cannabis. We do a pregnancy test for women.
- Examination of the stool. After the screening, you will be given a jar to hand in stool.

Please note: it may happen that you are healthy, but that you are not suitable to participate. The investigator will tell you more about this.

Visits and measurements
The entire schedule of visits can be seen in Figure 1 for Group A (Part 1 of the study only), Figure 2 for Group B (Parts 1 and 2) and in Appendix C.

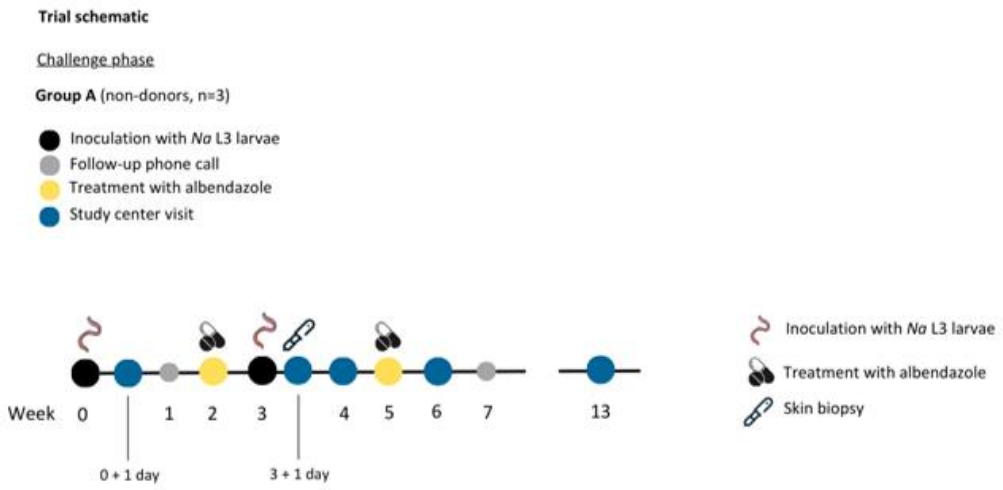


Figure 1: overview of study visits group A (non-donors)

- Participants in Part 1 of the study (Group A and Group B)
You will be exposed to hookworm larvae twice. On the day of the infection or 1 day before, you will be in the hospital and your blood will first be checked to see if you are still suitable for participation in the study. In women, a pregnancy test is taken prior to each infection and treatment.
Exposure to hookworm larvae takes place by placing a damp mesh on the skin. The hookworm larvae are on it. You cannot see these. The gauzes remain in place for an hour. After removing the gauzes, you will remain in the hospital for another half hour. Then you go home. The whole procedure of exposure takes about 1 1/2 to 2 hours.

At the first exposure, four gauzes with hookworm larvae are placed, one on each upper arm and one on each calf. In the second exposure, a lower amount of larvae is given and only one mesh is used, which is placed on the non-writing arm.
You will return to the study center one day after exposure. After the first exposure, only blood is taken. After the second exposure, two skin biopsies are also taken: one on the side where the hookworm exposure took place and one on the other side.

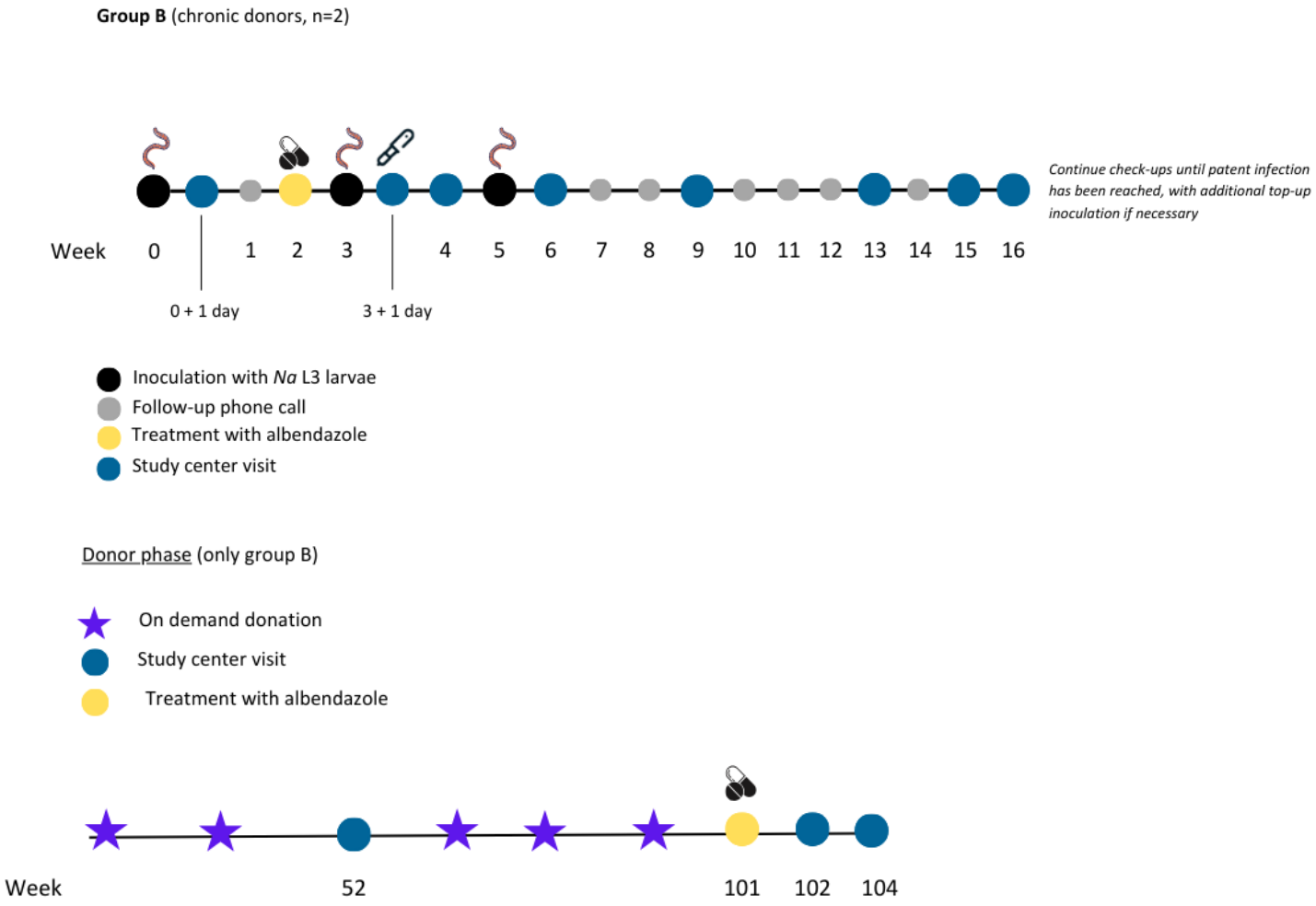
Skin biopsy procedure
Two skin biopsies will be taken from each volunteer one day after the second infection; one of each upper arm. Because the second infection only occurred on the non-writing arm, we can compare the biopsies and the biopsy of the writing arm serves as a 'control collection'. A biopsy is the taking of a piece of tissue, in this case a piece of skin. First, the skin will be locally numbed with an anesthetic shot. As a result, you will not experience any pain from taking the biopsy. This injection causes a sharp, burning pain for a short time - a few seconds. A small device ('apple corer') is pressed into the skin, after which a piece of skin of about 3 mm comes loose. In general, people have no or very little problems with this after the procedure. There is a small chance of bleeding or infection at the site of the biopsy. This can be treated well. It is possible that a small scar remains at the site of the skin biopsy that can remain visible for several months. People with darker skin types are more likely to develop a scar. In total, the skin biopsy takes about 15 minutes.

The infections will occur at week 0 and week 3 of the study. Two weeks after the initial infection, all participants will receive treatment with albendazole tablets. This consists of a 3-day course, taking one tablet each day. Participants in group A will also receive treatment with albendazole two weeks after the second infection. For participants in group B, the study proceeds differently from week 5, this procedure is in the next paragraph. Group A participants will come to the study center at weeks 4, 6, and 13 of the study for controls. The investigator will contact you by phone for check-ups at week 1 and week 7. If necessary, you may be asked to visit the study centre in between.
Blood will be drawn at all visits and you will be asked to bring stool with you at each visit. If you only participate in part 1 of the study, the study will be completed after the thirteenth week.

During the infection phase of the study, all participants will be asked to keep a paper diary in which you write down whether you experience any physical symptoms. This diary will be issued to you on the day of the first infection. You fill this in at home if you experience complaints and take it with you on every subsequent visit. With the diary, the investigators check your symptoms and copy them into the electronic study system. You will return the diary to the investigators at the end of the study.

- Participants in Part 2 of the study (Group B only)
For the two donors, the course of the study is different from week 5 onwards (see Figure 2).

Figure 2. Overview of study visits for chronic donors (group B)



In total, you will be exposed to hookworm larvae three times. Where the participants from group A are treated after the second infection, you will be exposed for the third time at that time to increase the risk of chronic infection. With the third infection, you will receive the same dose of larvae as with the first infection, which are administered to the arms and calves. Then you will come to the study center on weeks 6, 9 and 13 for check-ups where blood is drawn and stool must be submitted. You will also come to the hospital in week 15 and week 16 of the examination, with a total of five stool submissions during these two weeks. In the intervening weeks, the investigator will contact you by telephone.

After week 16, the infection part of the study is over and the donor part starts.

At that time, you have already had an examination for inclusion in the hookworm study. If your health has not changed during this study, another short examination will take place during which blood tests will be done. The HIV, hepatitis B and hepatitis C tests are repeated. The

only other condition for participating in the donor part of the study is production of sufficient worm eggs.

After week 16, the two donors will be asked to donate feces regularly (5-10 times a year). It is possible that the study doctor will also take blood for checking for HIV, hepatitis B and C, but this will not be more than once a month. The timing of these donations has not yet been determined and depends on the planned experiments. The study doctor will contact you at least one week in advance. Your donation will be used to breed larvae from the faeces.
After a year, the donors come for a check-up where we check the blood and stool. After two years or if you stop earlier, you will be treated with albendazole. After the treatment, you will come back twice more to check whether the infection has been cured.

5. What agreements do we make with you?

In order for the investigation to run smoothly and for your own safety, it is important that you adhere to the following agreements. The agreements are that you:

- contact the investigation team in case of complaints (available 7 days a week, 24 hours a day);
- does not also participate in another medical-scientific study;
- keeps all appointments for check-ups at the hospital;
- maintain good hygiene during toilet visits and always use a toilet that can be flushed with tap water;
- you do not travel to an area where hookworm occurs during the study (the coloured areas of Figure 2);
- you do not donate blood during the study.
- For female participants: that you do not become pregnant during the study. If you do want to become pregnant during the study, you can indicate this to the investigator, so that you can be treated if desired. The study will then end for you.

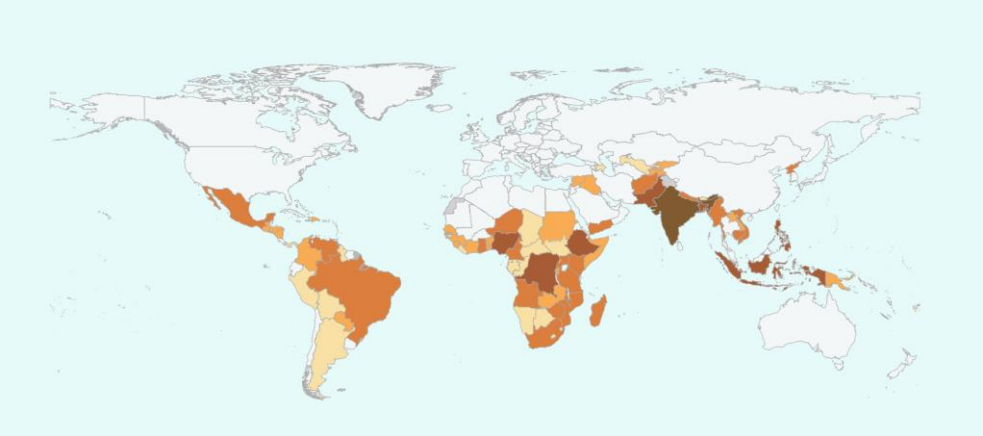


Figure 2. Areas where hookworm infections occur (WHO 2019)

It is important that you contact the investigator:

- before taking any other medicines.
- if you are admitted to hospital or treated;

- if you suddenly develop health problems;
- if you no longer wish to participate in the study;
- if your contact details change.

6. Which side effects, adverse effects or inconveniencies may you encounter?

The symptoms after a hookworm infection can be split into symptoms during the first 16 weeks and the symptoms thereafter. There is no risk of spreading the infection in the Netherlands, because the climate is too cold and flush toilets are used.

Side effects of hookworm infection first 16 weeks

A hookworm infection can cause symptoms. These side effects are expected to occur with some regularity.

Immediately after the infection:

- Itching and rashes at the site of infection: in people with darker skin tones, this can lead to skin discoloration.

From 3 to 4 weeks after infection:

- Diarrhoea
- Abdominal cramps
- Nausea
- Flatulence

The amount of complaints and severity of the complaints can vary greatly from person to person. In a previous study, 4 out of 10 volunteers suffered from severe abdominal pain, 6 out of 10 volunteers had little or no complaints. The severe abdominal pain lasted less than a day in most cases. The abdominal complaints can last from a few days to a few weeks. Abdominal pain can be treated with painkillers. If this does not work sufficiently, you will be treated with albendazole, which will make the symptoms disappear after a few days.

Side effects of chronic hookworm infection for donors (after 16 weeks)

It is expected that you will not experience any symptoms during the chronic infection (after week 16): in our previous study, no one had any symptoms. However, there is a chance that you may suffer from

- bellyache;
- diarrhoea;
- flatulence;
- nausea;
- weight loss;
- anemia.

Discomfort of skin biopsies

Skin biopsies can lead to discomfort, such as:

- Pain;

- Hemorrhage;
- Wound infection;
- Scarring;
- Local discoloration of the skin (hypo- or hyperpigmentation).

Side effects of albendazole

Albendazole is the most effective drug against hookworm infections. Side effects that are common (more than 1 in 10 people) are headaches and a slight to moderate increase in liver enzyme levels. Occasional side effects (more than 1 in 100 people) include dizziness, fever, and temporary hair loss.

Other inconveniences

Blood draws: We will take blood from you during the scheduled visits (8 times in group A and 9 times in group B). In group B, we also sometimes take blood samples with a donation and at weeks 52, 101 and 102. Blood tests can hurt or cause bruising. All in all, we no longer take 500 ml of blood from you every four months. This amount does not cause a problem in adults. In comparison, 500 ml of blood is taken at a time at the blood bank.

Stool collection: For most visits to the study center, you will need to bring some stool that you will need to collect at home. For this you will receive materials that make it easier to collect.

Study Center Visits: You must come to the study center a total of 9 (Group A) or 12 (Group B) times for the screening, scheduled check-ups, and the moment of infection. Donors also come 5-10 times a year for a donation and have four extra check-up appointments during the two years. Checks/donations take about 15 minutes. You will be present for about 2 hours for the moment of infection, the visit in which the skin biopsies are taken takes about 45 minutes. It is important that you are present on time for the check-ups.

7. What are the advantages and disadvantages of participating in this study?

Participating in the study can have advantages and disadvantages. We list them below. Think about this carefully, and talk about it with others.

You do not benefit from participating in this study. But with your participation, you can contribute to the development of new vaccines and medicines against hookworm infections.

Disadvantages of participating in the study can be:

- possible side effects of the hookworm infection;
- possible inconveniences of the measurements in the study;
- possible discomfort from the collection of the skin biopsy;
- possible discomfort of treatment with albendazole;
- you are not allowed to travel to areas where hookworms occur during the study;
- You are not allowed to donate blood during the study.
- For female participants: you must not become pregnant during the study

Participation in the study also means:
- that you lose extra time;
- that you have agreements that you must keep.

All these matters are described above under points 4, 5 and 6.

8. If you do not want to participate or want to stop the study

You decide whether to participate in the study. Participation is voluntary. If you do participate, you can always change your mind and still stop, even during the study. You don't have to say why you are quitting. However, you must report this immediately to the investigator. If you want to stop after you have been infected, you will still need to take albendazole and visit the hospital several times for your own safety and to monitor the infection. The data collected up to the moment you indicate that you want to stop will be used for the study. The data from the subsequent visits (treatment and safety tests) will only be collected to monitor your safety, but will not be used for the study. If there is any new information about the study that is important to you, the investigator will let you know.

9. End of the study

Your participation in the study will stop if:
- all visits are over;
- you choose to stop;
- the investigator thinks it is better for you to stop;
- The Safety Monitor, the government or the assessing medical ethics review committee decides to stop the study.

The entire study is over when all participants have finished. After processing all the data, the investigator will inform you about the most important results of the study. This will take place after the last visit of all participants.

10. Use and storage of your data and body material

Will you participate in the survey? Then you also give permission for your data and bodily material to be collected, used and stored.

What data do we keep?
We store this data:
- your name
- your gender
- your address
- your date of birth
- data about your health
- (medical) data that we collect during the study

In addition, your account number and BSN are required to pay the compensation. This data will not be stored after payment of the compensation.

What body material do we store?
We collect, use and store tubes of blood, urine and faeces.

Why do we collect, use and store your data and body material?
We collect, use and store your data and your bodily material in order to be able to answer the questions of this study and to publish the results.

How do we protect your privacy?
To protect your privacy, we provide a code for your data and your body material. We only put this code on all your data and body material. We keep the key to the code in a secure place at the LUMC. When we process your data and body material, we only use that code. Even in reports and publications about the investigation, no one can recall that it was about you.

Who can see your data?

However, some people can see your name and other personal information without a code. This can be data collected specifically for this study, but also data from your medical file. These are people who check whether the investigatorss are conducting the study properly and reliably. These people can access your data:

- The monitor that monitors the safety of the investigation.
- An inspector who works for the client.
- National supervisory authorities.

These people keep your data confidential. We ask you to give permission for these persons to inspect it. The Health and Youth Care Inspectorate can view your data without your permission.

How long do we keep your data and body material?
We store your data at the LUMC for 15 years. The paper diaries are also kept at the LUMC for 15 years.
We store your body material at the LUMC. It will be kept for 15 years in order to be able to make new determinations related to this research in the course of this study. As soon as this is no longer necessary, we destroy your body material.

What happens in the event of unexpected discoveries?
During the examination, we may accidentally find something that is not directly important for the examination but is important for your health. The investigator will then contact your general practitioner or specialist. You will then discuss with your general practitioner or specialist what needs to be done. The costs of this are covered by your own health insurance. With the form, you give permission to inform your general practitioner or specialist.

Can you withdraw your consent to the use of your data?

You can revoke your consent to the use of your data at any time. Then tell the investigator. But beware: do you withdraw your consent, and have investigatorss already collected data for a study? Then they may still use this data. For your body material, the investigator will destroy it after you withdraw your consent. But have measurements already been taken with your body material? Then the investigator may continue to use the results.

Want to know more about your privacy?

- Would you like to know more about your rights regarding the processing of personal data? Then take a look at www.autoriteitpersoonsgegevens.nl.
- Do you have questions about your rights? Or do you have a complaint about the processing of your personal data? Please contact the person responsible for processing your personal data. For your study, this is:
    - The Data Protection Officer of the LUMC. See Appendix A for contact details, and website.
- If you have any complaints about the processing of your personal data, we recommend that you first discuss them with the investigation team. You can also go to the Data Protection Officer of the LUMC. Or you can file a complaint with the Dutch Data Protection Authority.

Where can you find more information about the study?
You can find more information about the study on www.clinicaltrials.gov, under number NCT06478498. After the study, the website can show a summary of the results of this study.

11. Subject insurance

Insurance has been taken out for everyone who participates in this study. The insurance covers damage caused by the investigation. Not all damage is covered. You can find more information about the insurance in Appendix B. It also states to whom you can report damage.

12. Informing the GP

We always send your GP (and possibly specialist) a letter to let them know that you are participating in the study. This is for your own safety. If you do not agree with this, you cannot participate in this study. You cannot participate in the study if you do not have a primary care physician.

13. Compensation for participation

For participating in this study, you will receive an expense allowance including travel expenses. This amount consists of an amount per visit. The compensation is different for participants from group A and B. Participants from group A receive a total of €1250 for all visits,-. Participants from group B will receive €2050 plus €50 extra for each stool donation. Of this, you will receive €1700 after completing the infection phase, and the remaining €350 after completing the entire study. If you miss visits, you may not be paid for the missed visit. The allowance must be reported to the Tax and Customs Administration as income.

14. Do you have any questions?

If you have any questions, please contact the study team. For independent advice on participating in this study, please contact the independent doctor, A.H.E. Roukens. She is an internist-infectiologist at the LUMC and knows a lot about the study, but is not directly involved. Its details can be found in **Appendix A.**

If you have any complaints about the study, you can discuss this with the investigator or your attending physician. If you prefer not to do this, you can contact the complaints officer. All details can be found in **Appendix A**: Contact details.

15. Signing consent form

You can first think calmly about this study. Then you tell the investigator whether you understand the information and whether or not you want to participate. Would you like to participate? Then you must fill in the consent form that you will find with this information letter during the screening visit. If you want to participate in the donor sub-study, you must sign an additional consent form. You and the investigator will both receive a signed version of the consent form.

Thank you for your attention.

16. Attachments to this information

A. Contact details
B. Insurance information
C. Schedule of study actions participants group A
D. Schedule of study actions participants group B
E. Consent form participants group A
F. Consent form group B participants

Appendix A: contact details

Researchers
Leiden University Medical Center
Prof. Dr. M. Roestenberg, internist-infectiologist
Dr. M. Hoogerwerf, physician-researcher
Department LUCID
Phone: 06-20942061
Email: vaccinonderzoek@lumc.nl

Independent doctor
Leiden University Medical Center
Dr. A.H.E. Roukens, internist-infectioloog
Infectious Diseases Department
Tel: 071-5262613
Email: A.H.E.Roukens@lumc.nl

Complaints:
In case of complaints about the study, you can contact the LUMC Complaints Team via email: patiëntenservicebureau@lumc.nl. You can also contact the Patient Service Office by telephone (071-5262989; during office hours). They will handle the complaint in accordance with the applicable agreements.

Contact details patient service office
LUMC Patient Service Office
P.O. Box 9600
2300 RC Leiden
Phone: 071-5262989

Data Protection Officer of the institution:
If you have any questions about the protection of your privacy, you can contact the data protection officers of the LUMC (DPO) via infoavg@lumc.nl

For more information about your rights:
Contact details LUMC
Albinusdreef 2
2333 FOR Leiden
Central telephone number: 071-5269111
For more information about your rights, see the LUMC website
www.lumc.nl/over-het-lumc/privacy/

Appendix B: information about the insurance

The Leiden University Medical Center has taken out insurance for everyone who participates in this study. The insurance covers damage caused by participation in the study. This applies to damages during the study or within four years after the end of your participation in the study. You must have reported damage to the insurer within those four years.

The insurance does not cover all damages. At the bottom of this text you can briefly see which damage is not covered.
These provisions are contained in the 'Decree on compulsory insurance for medical-scientific study involving human subjects 2015'. This decree is in the Government Laws Bank (https://wetten.overheid.nl).

In the event of damage, you can contact the insurer directly by phone or e-mail

The insurer of the study is:	
Name:	Centramed
Address:	Maria Montessorilaan 9, 2719 DB Zoetermeer
Telephone number:	070-3017070
Email:	info@centramed.nl
Policy number:	624.530.305

The insurance provides coverage of €650,000 per test subject and €5,000,000 for the entire study and €7,500,000 per year for all studies from the same client.

The insurance does not cover the following damages:

- damage due to a risk that you have been informed of in the written information. This does not apply if the risk is more severe than anticipated or if the risk was very unlikely;
- damage to your health that would have occurred even if you had not participated in the study;
- damage due to not (fully) following directions or instructions;
- damage to your descendants, as a result of a negative effect of the study on you or your descendants;
- damage caused by an existing treatment method in research into existing treatment methods.

Appendix C: Schedule of study actions participants group A

Day	Location/time & duration	Activity
Screening visit	LUMC (by appointment), duration: 1 – 1.5 hours	The study will be explained and questions from the candidate subject will be answered. Medical examination: the consent form will be signed and the medical history of candidate subjects will be requested and recorded. The following tests and assessments are performed: - Temperature, blood pressure, heart rate; -Physical; - Blood sampling and blood tests; - Urine collection for a drug test; Pregnancy test for women - Submit stool sample material (after the screening appointment)
Study Day 1 (Week 0) Day of Hookworm Exposure	LUMC (by appointment), duration: 1.5 – 2 hours in the morning	The test subjects are exposed to hookworms through infection with 50 larvae The following tests and assessments are performed: -Temperature; - Blood sampling and blood tests - Pregnancy test for women
Study day 2 (week 0)	LUMC, between 8:00 and 12:00 in the morning, duration: 15 minutes	The following tests and assessments are performed:- Blood sampling and blood tests
Study day 8 (week 1)	Subjects will be called, duration: 2-15 minutes	The following tests and assessments are performed: - Discussion of side effects; - Discussion of the study diary.
Study Day 15 (Week 2) Treatment	LUMC, between 8:00-12:00 in the morning,,duration: 15-30 minutes	All subjects will be treated with albendazole The following tests and assessments are performed: - blood sampling and blood tests - Pregnancy test for women
Study Day 22 (Week 3) Second Exposure to Hookworms	LUMC (by appointment), duration: 1.5 – 2 hours in the morning	The test subjects will be exposed to hookworms through infection with 10 larvae The following tests and assessments will be performed: -Temperature; - Blood sampling and blood tests - Submit a stool sample - Pregnancy test for women - Skin reaction assessment
Study day 23 (week 3) Day of taking biopsies	LUMC (by appointment, duration: 45 min in the morning	Two biopsies will be taken from the test subjects. The following tests and assessments also take place:- Blood sampling and blood tests - Skin reaction assessment
Study day 29 (week 4)	LUMC (by appointment), duration: 15 min in the morning	Check-up appointmentThe following tests and assessments will be performed: - Skin reaction assessment - Discussion of side effects; - Discussion of the study diary.
Study Day 36 (Week 5) Second Treatment	LUMC, between 8:00-12:00 in the morning, duration: 15-30 minutes	The subjects will be treated with albendazole. The following tests and assessments are performed: - Skin reaction assessment - Blood sampling and blood tests

		- Pregnancy test for women - Skin reaction assessment
Study day 43 (week 6)	LUMC (by appointment), duration: 15 min in the morning	Check-up appointment The following tests and assessments will be performed:- Skin reaction assessment - Blood sampling and blood tests - Discussion of side effects - Study diary discussion
Study day 50 (week 7)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study day 92 (week 13) Submission of stool sample, last study appointment	LUMC (by appointment), duration: 10 min in the morning	This is the end of the investigation. The following tests and assessments will be performed: - Submit stool sample (two samples)- Discussion of adverse events; - Discussion of the study diary.

Appendix D: Schedule of study actions participants group B

NB. The stool donations are not listed in this schedule, because the date is not fixed. The investigator will call at least 1 week in advance to schedule an appointment for this. You can count on about 5-10 additional visits per year for the donation of stool. Every time you donate stool you will be checked for HIV, hepatitis B and C but no more than once a month. During this period, you decide when and if you want to donate.

Day	Location/time & duration	Activity
Screening visit	LUMC (by appointment), duration: 1 – 1.5 hours	The study will be explained and questions from the candidate subject will be answered. Medical examination: the consent form will be signed and the medical history of candidate subjects will be requested and recorded. The following tests and assessments are performed: - Temperature, blood pressure, heart rate; -Physical;

		- Blood sampling and blood tests; - Urine collection for a drug test; Pregnancy test for women - Submit stool sample material (after the screening appointment)
Study Day 1 (week 0) Day of exposure to hookworms	LUMC (by appointment), duration: 1.5 – 2 hours in the morning	The test subjects are exposed to hookworms through infection with 50 larvae The following tests and assessments are performed: -Temperature; - Blood sampling and blood tests
Study day 2 (week 0)	LUMC, between 8:00 and 12:00 in the morning, duration: 15 minutes	The following tests and assessments are performed:- Blood sampling and blood tests
Study day 8 (week 1)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 15 (Week 2) Treatment	LUMC, between 8:00-12:00 in the morning, duration: 15-30 minutes	All subjects will be treated with albendazole The following tests and assessments will be performed: - Taking albendazole - blood sampling and blood tests - Pregnancy test for women
Study day 22 (week 3) Second exposure to hookworms	LUMC (by appointment), duration: 1.5 – 2 hours in the morning	The test subjects will be exposed to hookworms through infection with 10 larvae The following tests and assessments will be performed: -Temperature; - Blood sampling and blood tests - Submit a stool sample - Pregnancy test for women - Skin reaction assessment
Study day 23 (week 3) Day of taking biopsies	LUMC (by appointment, duration: 45 min in the morning	Two biopsies will be taken from the test subjects. The following tests and assessments also take place:- Blood sampling and blood tests - Skin reaction assessment
Study day 29 (week 4)	LUMC (by appointment), duration: 15 min in the morning	Check-up appointmentThe following assessments will be performed: - Skin reaction assessment- Discussion of side effects; - Discussion of the study diary.
Study day 36 (week 5) Third exposure to hookworms	LUMC (by appointment), duration: 1.5 – 2 hours in the morning	The test subjects are exposed to hookworms through infection with 50 larvae The following tests and assessments are performed: -Temperature; - Blood sampling and blood tests - Pregnancy test for women
Study day 43 (week 6)	LUMC (by appointment), duration: 15 min in the morning	Check-up appointment The following tests and assessments will be performed:- Skin reaction assessment - Blood sampling and blood tests - Discussion of side effects - Study diary discussion
Study day 50 (week 7)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.

Study Day 57 (Week 8)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 64 (Week 9)	LUMC, between 8:00-12:00 in the morning, duration: 15-30 minutes	The following tests and assessments are performed: - Skin reaction assessment - Blood sampling and blood tests-Discussion of side effects; - Discussion of the study diary.
Study day 71 (week 10)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 78 (Week 11)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 85 (Week 12)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 92 (Week 13)	LUMC, between 8:00-12:00 in the morning, duration: 10 minutes	Check-up appointment The following assessments will be performed:- Discussion of adverse events; - Discussion of the study diary.
Study Day 99 (Week 14)	Subjects will be called, duration: 2-15 minutes	The following assessments are carried out: - Discussion of side effects; - Discussion of the study diary.
Study Day 106 (Week 15)	LUMC, between 8:00-12:00 in the morning, duration: 10 minutes	The following tests and assessments will be performed:-Submission of stool samples (two) - Discussion of side effects; - Discussion of the study diary.
Time between day 106 - 113	LUMC, Monday-Friday from 8:00 -13:00, duration: 5 minutes	- Submit a stool sample
Study Day 113 (Week 16)	LUMC, between 8:00-12:00 in the morning, duration: 10 minutes	Last visit of the infection phase of the investigation. The following tests and assessments will be performed:-Submission of stool samples (two) - Discussion of side effects; - Discussion of the study diary.
Study Day 365 (Week 52)	LUMC (by appointment), duration: 25 min	Check-up appointmentThe following tests are performed: - Blood sampling and blood tests - Submit stool sample -Discussion of side effects; - Discussion of the study diary.
Study Day 708 (Week 101) Treatment	LUMC (by appointment), duration: 30 min	The subjects will be treated with albendazole. The following tests and assessments are performed: - Inname albendazole - blood sampling and blood tests - Submit a stool sample - Pregnancy test for women - Discussion of side effects; - Discussion of the study diary.
Study Day 715 (Week 102)	LUMC (by appointment), duration:15 min	Check-up appointment The following tests and assessments will be performed: - blood sampling and blood tests - Discussion of side effects; - Discussion of the study diary.

Study Day 729 (Week 104)Stool Sample Submission, Final Study Visit	LUMC (by appointment), duration: 15 min	This is the end of the investigation. The following tests will be performed: - Submit stool sample (two samples)- Discussion of side effects; - Discussion of the study diary.

Appendix E: Consent form subject, group A

Investigating the local immune responses in the skin after repeated hookworm infection (HiBiSki)

- I have read the information letter. I was also able to ask questions. My questions have been answered well enough. I had enough time to decide whether to participate.
- I know that participating is voluntary. I also know that I can decide at any time not to participate in the study. Or to stop it. I don't have to say why I want to stop.
- I give the investigator permission to let my GP know that I am participating in this study.
- I give the investigator permission to give my GP information about unexpected findings from the examination that are important for my health.
- I give the investigators permission to collect and use my data and bodily material. The investigators are only doing this to answer the study question of this study.
- I know that for the purpose of checking the study, some people can see all my data. Those people are listed in this information letter. I give these people permission to view my data for this check.
- I know that I am not allowed to travel to an area where hookworm occurs during the study.
- I know that I am not allowed to donate blood at Sanquin during the study.
- For women: I know that I am not allowed to get pregnant during the study.
- For women: The investigator discussed with me how best to prevent myself from getting pregnant.

Would you like to tick yes or no in the table below?

I give permission to keep my data for 15 years to use it for other research, as stated in the information letter.	**Yes** ☐	**No** ☐
I give permission to ask me if I want to participate in a follow-up study after this study.	**Yes** ☐	**No** ☐

- I want to participate in this study as a participant in group A.

My name is (test subject):

Signature:

Datum (DD/MMM/JJJJ): ___ / _____ / _____ Time __ : __ hours (24h format)

I declare that I have fully informed this subject about the said study.

Is there any information that may influence the subject's consent during the study? Then I let this test subject know in time.

Name of investigator (or his representative):

Signature:

Datum (DD/MMM/JJJJ): ___ / _____ / _____ Time __ : __ hours (24h format)

The subject will be provided with a complete information letter, together with a certified copy of the signed consent form.

Appendix F: Consent form subject, group B

Investigating the local immune responses in the skin after repeated hookworm infection (HiBiSki)

- I have read the information letter. I was also able to ask questions. My questions have been answered well enough. I had enough time to decide whether to participate.
- I know that participating is voluntary. I also know that I can decide at any time not to participate in the study. Or to stop it. I don't have to say why I want to stop.
- I give the investigator permission to let my GP know that I am participating in this study.
- I give the investigator permission to give my GP information about unexpected findings from the examination that are important for my health.
- I give the investigators permission to collect and use my data and bodily material. The investigators are only doing this to answer the research question of this study.
- I know that for the purpose of checking the research, some people can see all my data. Those people are listed in this information letter. I give these people permission to view my data for this check.
- I know that I am not allowed to travel to an area where hookworm occurs during the study.
- I know that I am not allowed to donate blood at Sanquin during the study.
- For women: I know that I am not allowed to get pregnant during the study.
- For women: The investigator discussed with me how best to prevent myself from getting pregnant.

Would you like to tick yes or no in the table below?

I give permission to keep my data for 15 years to use it for other research, as stated in the information letter.	**Yes** ☐	**No** ☐
I give permission to ask me if I want to participate in a follow-up study after this study.	**Yes** ☐	**No** ☐

- I want to participate in this study as a participant in group B.

My name is (participant):

Signature:

Datum (DD/MMM/JJJJ): ___ / _____ / _____ Time __ : __ hours (24h format)

I declare that I have fully informed this subject about the said study.

Is there any information that may influence the subject's consent during the study? Then I let this test subject know in time.

Name of investigator (or his representative):

Signature:

Datum (DD/MMM/JJJJ): ___ / _____ / _____ Time __ : __ hours (24h format)

The subject will be provided with a complete information letter, together with a certified copy of the signed consent form.