ERMA

UNIVERSITY OF JYVÄSKYLÄ

INFORMATION ON THE STUDY

Estrogenic Regulation of Muscle Apoptosis (ERMA, ERMA/Core and ERMA/Physical Activity)

Gerontology Research Center (GEREC), Faculty of Sport and Health Sciences,
University of Jyväskylä

Research group:
Vuokko Kovanen, Adjunct Professor, Principal Investigator, University of Jyväskylä
Eija Laakkonen, Adjunct Professor, co-Principal Investigator, University of Jyväskylä
Pauliina Aukee, Chief Clinician, Department of Obstetrics and Gynaecology, Central Finland Central Hospital
Urho Kujala, Professor of Sports & Exercise Medicine, University of Jyväskylä
Taija Juutinen, Professor of Kinesiology, University of Jyväskylä
Eeva-Maija Palonen, Research Coordinator, University of Jyväskylä
Harri Selänne, Specialist in Sports and Exercise Medicine, LIKES Research Centre for Physical Activity and Health
Sarianna Sipilä, Professor in Exercise Gerontology, University of Jyväskylä
Tuija Tammelin, Research Director, LIKES Research Centre for Physical Activity and Health
Ina Tarkka, Adjunct Professor, Senior Researcher, University of Jyväskylä

FOR FURTHER INFORMATION AND INQUIRIES, PLEASE CONTACT:

Vuokko Kovanen
Principal Investigator, Adjunct Professor
University of Jyväskylä
Department of Health Sciences
Tel: 040 805 3566 or 040 549 1486
erma-info@jyu.fi

Eeva-Maija Palonen
Research Coordinator
University of Jyväskylä
Department of Health Sciences
Tel: 040 805 4211
erma-info@jyu.fi

ESTROGENIC REGULATION OF MUSCLE APOPTOSIS (ERMA) STUDY

We invite you to participate in a research project that focuses on menopausal estrogen deficiency and the mechanisms through which estrogen affects body composition, muscle performance and mental wellbeing in 47–55-year-old women. Estrogen deficiency in the menopause may be one of the factors that weaken muscle properties and function during ageing. An optimal physiological muscle condition is extremely important for health and wellbeing.

We have estimated that you may be suitable for this study because women of your age are likely to reach or have reached the menopause during this study. This information leaflet describes the study and your role in it. You can also discuss the details with the principal investigator or coordinator (please find the contact info on the cover page).

The study is conducted at the University of Jyväskylä, Faculty of Sport and Health Sciences, Gerontology Research Center from 2014 to 2018. The University of Jyväskylä maintains the research register, for which the contact person is principal investigator Vuokko Kovanen. The Research Ethics Committee of the Central Finland Health Care District has evaluated the research plan and provided a favourable statement on it.

VOLUNTARY PARTICIPATION AND RIGHT TO INTERRUPT

Participation in this study is voluntary, and you can withdraw at any time with no need to explain the reason. Potential withdrawal has no consequences for you**.** If you cancel your consent, you have the right to request that the information and samples collected from you so far will no longer be used in the study. If you, instead of cancelling your consent, withdraw from the study, the information and samples collected from you before the interruption will be used in the study.

PURPOSE OF THE STUDY

The purpose of this study is to determine how estrogen differences in women at different stages of menopause and changes in estrogen level affect the body composition, muscle performance and mental wellbeing of 47–55-year-old women. We examine these issues with questionnaires, laboratory tests and measurements, also including blood and tissue samples. In addition, the function of deep pelvic floor muscles is mapped with the attached symptom questionnaire. **After completing the attached forms, return them together with the signed consent form within ten days from receiving this letter.** Please return them even if you were not interested in participating in all the stages of the study. Return postage is paid for you.

PROGRESS OF THE ERMA STUDY

The invitation to participate in the study is sent to randomly selected 47–55-year-old women living in the city of Jyväskylä or its surrounding municipalities. We have received their contact information from the population information system of the Population Register Centre. After receiving your answer, we will inform you in a letter about your approval for the study. We will also send you a letter if you are not suitable for the study. In the consent form, you are also asked about your email address and your possibility to use the Internet. They are needed for using the online appointment system and for completing a

menstruation dairy. However, you can also complete the diary on paper and book appointments by phone, if you prefer to do so. We utilise a safe Internet connection maintained by the University of Jyväskylä.

The ERMA study is implemented in **two stages**. In addition, some of the participants also participate in the *ERMA core study group* later referred as ERMA+ study, in which we carry out more detailed muscle tests and analyse cognitive function. Depending on which research group you will be included in and whether you participate in the ERMA+ study, the project can take a minimum of about three months (two visits) and a maximum of about 3.5 years (APPENDIX 1). The number of visits (about 2–20) depends on the group and the development of menopausal status.

STAGE 1 OF THE ERMA STUDY

The first stage of the study includes two visits. You will begin by keeping a menstruation diary.

At the beginning of the first visit, we will ask for your consent to participate in the study, and you can complete a health status form. In addition, we will take a venous blood sample from the antecubital vein in order to determine sex hormones. The blood sample is taken on one of the initial days of your menstrual cycle (bleeding days 1–5). If you no longer have menstrual periods, the sample can be taken on any working day. The blood sample may cause mild pinching pain and a bruise in the sample area. Based on hormone analyses and the regularity of your periods, you will be placed in one of the four research groups: premenopause (Group 1), early perimenopause (Group 2), late perimenopause (Group 3) or postmenopause (Group 4). The blood sample is also used to analyse common health factors such as blood sugar, cholesterol, genes (DNA) and their manifestation. The sample is taken after overnight fasting. After the sample, we will offer you a light breakfast. Please reserve about half an hour for the visit.

On the second visit, we perform measurements to analyse your body composition and physical condition. This visit takes about three hours (in the ERMA+ study: four to five hours). The tests are performed after overnight fasting. We will offer you a light breakfast during the morning. The tests are described more closely below.

Health examination: weight, height, waist–hip ratio, blood pressure, blood sample for small blood count. In addition, the questionnaire you have completed in advance is used to find out your health status, potential medications and health habits (e.g. physical activity, nutrition, smoking, alcohol consumption).

Body composition analysis: analysis of the proportional share of fat and non-fat tissue in your body as well as bone properties using the bioimpedance method and DXA device. The functioning of a DXA device is based on the use of X-radiation. APPENDIX 2 of this information leaflet provides more information on X-radiation and its potential negative effects.

Muscle strength measurements: The maximal strength of knee extensor muscles and handgrip strength are measured. The explosive strength production of foot extensor muscles is assessed with a jump test. The individual tests take a few seconds, and there is a break between them.

Maximal walking pace is measured during a ten-metre walk. In addition, a six-minute walking test is performed. During this test, you are expected to walk as long a distance as you can. Your heart rate and experienced load are simultaneously measured.

The women who, based on hormone analysis, belong to Group 2 or Group 3 participate in Stage 2 of the study. These analyses and the research conducted in Stage 2 are described in detail below.

STAGE 2 OF THE ERMA STUDY

The development of menopause status in women within Groups 2 and 3 will be followed based on the regularity of their menstrual periods and hormone analyses on blood samples. When the menopausal transition stage is over, the participants will be invited to final measurements. The follow-up in Stage 2 will last until the end of 2017, at the latest, after which we will perform the final measurements of the study.

Follow-up of menstrual cycle and related blood samples: Venous blood samples will initially be taken from Group 2 women every six months and from Group 3 every three months. The regularity of their menstrual cycle will be followed during the entire study with a menstruation diary. The follow-up will continue until the FSH value is higher than 30 IU/L in two successive samples, taken at an interval of a month, and until at least six months have passed from the last periods. In the follow-up visits, body composition is also measured with a bioimpedance device and questionnaires are completed.

Final measurements of the study: After you have passed the menopausal transition, final measurements will be performed for you. They are precisely the same measurements as in Stage 1 of the ERMA study.

ERMA+/CORE STUDY (see APPENDIX 1)

The ERMA+ study is part of the ERMA study. About 140 randomly selected women from Groups 2 and 3 participate in closer analyses of muscle condition and cognitive function. The analyses are performed during stages 1 and 2 of the ERMA study. Please find detailed descriptions of the ERMA+ measurements below.

Peroneal muscle activity and strength are measured based on voluntary maximal ankle extension and electric stimulation. In the latter, an ankle nerve is activated with a short-term electric impulse. The activation is first done on a relaxed muscle and then during maximal ankle extension. The electrodes used in activation are attached to the skin at the back of your knee and on the knee bone. The points where the electrodes are attached are shaved and the skin is cleaned. The correct places of the electrodes are determined by using a weak electric impulse. The impulse may feel a little uncomfortable for a moment, and you may feel a slight pinching pain that lasts less than a second. The same electrodes are used to examine the role of the brain in regulating peroneal activity. The analysis method is called transcranial magnetic stimulation (TMS). It is a method that has been used for over 20 years to measure nerve activity: an external magnetic field produces an autonomous muscle contraction. The study aims at finding out how fast a message (a nerve fibre impulse) travels from the brain to the muscle. The arrival of the message is visible as a reflexive tensioning of the peroneal muscle. In this test, the brain is activated during three different strength levels generated by the peroneal muscle. On each strength level, six to ten activations are performed. The activation impulse is very short and does not cause pain, but the reflexive tensioning of the muscle may startle you. The intervals between individual activations are 12 seconds, and they last for six seconds.

Cognitive function is analysed using commonly accepted paper-based tests. The tests assess, for example, your linguistic fluency, memory, attention, psychomotor speed, and how methodical and flexible your actions are.

Blood and muscle tissue samples. You can have a light breakfast at home before the visit. A venous blood sample is collected for hormone analyses. The tissue sample is taken by an experienced doctor who participates in the research. The sample cannot be taken if you are allergic to anaesthetics, have a haemorrhagic disease or take anticoagulants (e.g. Marevan or aspirin). The muscle sample is taken from the thigh muscle on the external side of your thigh, from a depth of about 1.5 centimetres. Your skin is cleansed and anaesthetised for this procedure (Lidocain c. adrenalin). A short, sterile cut of 0.5 to 1 centimetres is made on the skin and fasciae, through which the sampling needle is taken into the muscle. The anaesthetic needle can cause slight pain. You can feel the collection of the sample mainly as compression in the thigh muscle. The wound is closed with a butterfly bandage (no need for stitches), wound compress and an elastic adhesive bandage, which are removed on the following day, at the latest. Sampling may cause slight venous bleeding, which is minimised using an anaesthetic mixture, a support bandage and cold therapy. Sampling usually causes a small bruise and redness or a burning sensation in the sample area. This is part of normal wound healing process. Wound infections or bleedings within the muscle, instead, are very uncommon, but treatable as well. The muscle sample can leave a small 5-millimetre scar. Very seldom, a branch of a superficial nerve may be injured so that tactile sensations in a small area of the thigh skin become weaker for a few months. Even more seldom, one can

have a small local atrophy of the lower part of the muscle, which will heal on its own in about six months. For the days immediately after the sample collection, you will receive separate instructions for moving, washing and pain treatment, as well as the doctor's phone number for potential questions. The collection of the muscle sample takes about ten minutes, but you should reserve about 90 minutes for the entire visit.

The size and composition of muscles in the thigh and leg area are analysed with computer tomography (CT) scanning. CT scanning is based on X-radiation (see APPENDIX 2). Radiation is targeted at a limited area in the thigh and leg. The CT scanning is performed at the Central Finland Central Hospital, and the scanning itself takes about 15 minutes.

ERMA/PHYSICAL ACTIVITY STUDY (see APPENDIX 1)

ERMA/Physical Activity is part of the ERMA study. The measurements are performed during stages 1 and 2 of the ERMA study. No additional visits are needed. The ERMA/Physical Activity measurements are described in detail below.

Measuring physical activity with a physical activity meter (accelerometer). An accelerometer is a small, easy-to-use device, attached to your hip with an elastic band. The meter will be given to you on the second laboratory visit of the ERMA study and again when you come to the follow-up blood tests in Stage 2 of the ERMA study. You can return the meter to us by mail. During the visit, you will receive detailed instructions on how to attach and use the meter. The meter does not influence your normal life, and you only need to let it stay in the right place for the agreed period. Remove the meter before going to the sauna, washing and water sports. While using the meter, you also keep a diary of your physical activity. In the ERMA/Physical Activity study, the meter is worn during two to three periods, one period lasting seven successive days.

POTENTIAL BENEFITS FROM THE ERMA STUDY

It is possible that you will not personally benefit from participation in this study. However, your participation can help us find out issues that are important for the wellbeing of all women. You will also receive a wide variety of information on your own health through the health examination and other tests.

POTENTIAL HARM AND DISCOMFORT FROM THE STUDY

In the aforementioned sections, we have described the discomfort and/or harm caused by each situation or procedure, as well as clarified that the study requires several laboratory visits. The aim is to make the appointments as suitable for your schedules as possible. As we stated above, DXA and CT scanning is based on X-rays. In compliance with the legislation on radiation, Appendix 2 of this leaflet provides you with information on radiation.

CONFIDENTIALITY, DATA PROTECTION AND STORING

The study is carried out in accordance with good ethical and scientific practice, as stipulated in Finnish legislation. The information and results received on participants are handled confidentially, based on the Personal Data Act. Each participant is given a study ID code, and the information is stored coded in the research file. The results are analysed and reported coded on the group level, so that individual participants cannot be identified without the code key. The code key is stored by the principal investigator, and no identifying information is given to external parties. The material including personal data is stored in a locked space at the University of Jyväskylä, Faculty of Sport and Health Sciences. The DNA samples (blood and tissue samples) collected in the study are stored in a locked freezer at the University of Jyväskylä, Faculty of Sport and Health Sciences. The samples are stored and handled confidentially. The research files and samples are preserved until the data have been analysed. If the collected data or samples are needed for further studies later, you will be asked for a new consent. In international research cooperation, research data or samples sometimes need to be transferred to international researchers. In such cases, all the data and samples are coded so that they cannot be identified or linked to the participants of the study.

COSTS AND FUNDING OF THE STUDY

All examinations, samples and measurements are free of charge for you. No compensation is paid for participation in the study. You may be compensated for actual travel costs caused by your participation. The ERMA study is financed by the Academy of Finland, the Päivikki and Sakari Sohlberg Foundation and the Juho Vainio Foundation, and it is planned to last for four years.

PARTICIPANTS' INSURANCE COVERAGE

The University of Jyväskylä has insurance that covers the participants' unexpected, acute accidents resulting from external causes. A muscle or tendon strain injury directly caused by the participant's motion or exertion, which has been given medical treatment within 14 days of the occurrence of the injury, is also regarded as resulting from an accident. Compensation is paid for a maximum of six weeks from the occurrence of the injury. Magnetic resonance imaging or surgery procedures are not compensated for by the insurance. Illnesses and injuries gradually resulting from strain are not accidents, and they are thus not covered by the insurance. The accident insurance covers measurements carried out in the research facilities during the study, as parts of the research programme, and in commuting immediately related to the measurements. We are prepared to give first aid on accidents and acute illnesses. There is first-aid equipment in the laboratories, and the research staff are familiar with its use. The University has also taken liability insurance for the researchers who perform the measurements. The insurance against treatment injury covers procedures (local anaesthesia, muscle sampling) carried out by a doctor.

INFORMING PARTICIPANTS ABOUT THE RESULTS

You will receive the results of your hormone analyses when you come to the second visit. At the end of the visit, you will also receive the results of your blood sugar and lipid values, haemoglobin, height, weight, BMI and resting blood pressure, as well as of your muscle strength and walking rate, if you want. In accordance with the guidelines of the Ministry of Social Affairs and Health, the results of the DNA analyses are not given to the participants because, in the light of current knowledge, the results are difficult to interpret and most probably insignificant for research participants' health.

END OF THE ERMA STUDY

The measurements end in December 2018, at the latest, after which we will continue analysing the results. The study may end earlier if we manage to collect sufficient research data more quickly than anticipated. The participation of an individual participant is terminated if we discover during the study that her health could be at risk due to the study. In such a case, the participant will be referred to further examinations at the health centre or by her personal doctor.

APPENDIX 1. Progress of the ERMA study

Visits	Length of the visit	Procedures	Who participates	Test place
ERMA Stage 1				
1	30 min	Consent, questionnaire and blood test, on which hormones, blood sugar and lipids are analysed	All the women accepted for the study	Liikunta- ja terveyslaboratorio (Research laboratory of the University of Jyväskylä), Rautpohjankatu 8, Jyväskylä
2	about 3 hours	Health examination Checking of questionnaires Body composition Muscle strength measurements Walking tests *ERMA/Physical Activity* – physical activity meter is introduced: registration on 7 days	All the women accepted for the study	Liikunta- ja terveyslaboratorio, Rautpohjankatu 8
ERMA/CORE				
2.1 (during the 2nd visit)	30–40 min	Cognition	All the women in Groups 2 and 3 (also ERMA+)	Liikunta- ja terveyslaboratorio, Rautpohjankatu 8
	45–60 min	Muscle activation, TMS	Only the women selected for ERMA+	
3	60–90 min	Blood sample, muscle tissue sample	Only the women selected for ERMA+	Liikunta- ja terveyslaboratorio, Rautpohjankatu 8
4	15–30 min	Computer tomography (CT) scanning on thigh and leg	Only the women selected for ERMA+	Central Finland Central Hospital
ERMA Stage 2				
5->\ Visits at intervals of 3–6 months	about 30 min	Blood sample Questionnaires Body composition (bioimpedance) *ERMA/Physical Activity* – physical activity meter is introduced: registration on 7 days	All the women in Groups 2 and 3 (also those in ERMA+)	Liikunta- ja terveyslaboratorio, Rautpohjankatu 8
Final measurement	about 3 ½ hours (ERMA+: 4 ½ h)	Includes the measurements of visit 2 (ERMA+: in addition, 2.1 measurements and separate visits for the measurements of visits 3 and 4)	All the women in Groups 2 and 3 (also those in ERMA+)	Liikunta- ja terveyslaboratorio, Rautpohjankatu 8 (ERMA+: also Central Finland Central Hospital)

APPENDIX 2 TO THE INFORMATION LEAFLET

Information for research participants on ionising radiation. About one fourth (1.1 millisieverts, mSv) of the annual radiation dose of a Finn comes from background radiation in nature. This exposure cannot be influenced by us. The amount varies only a little according to location. Natural background radioactivity includes external radiation from soil and building materials as well as internal radiation caused by natural radioactive substances present in our organs. Radiation in nature also includes cosmic rays from space. People living high above sea level receive more space radiation than those living at low altitudes. A flight exceeding an altitude of 10 kilometres exposes the traveller to space radiation. The radiation dose on a flight from Helsinki to Shanghai is about 0.05 mSv. Indoor air radon is the source of more than half of Finns' annual radiation dose (average 2.0 mSv). The radon gas from natural uranium in the soil accumulates in the air of homes and workplaces, and we inhale it.
The medical use of radiation provides the largest dose of radiation coming from artificial sources. Finns get an average dose of 0.5 mSv a year from X-ray examinations. For example, a lung X-ray exposes the patient to 0.02 mSv. Due to nuclear tests and the Chernobyl accident, there is still a small amount of radioactive elements in nature, but their share of Finns' average annual radiation dose is very small. The average dose of radiation originating in Chernobyl has been 0.02 mSv per person in the past years – that is, only one percent of a Finn's annual radiation dose.

Limits have been set for the doses of people exposed to radiation in their work (Radiation Decree / Säteilyasetus 1512/1991, §3). The dose from work in, for instance, a hospital or nuclear power plant must not exceed the average annual level of 20 mSv over five years, and it must not exceed 50 mSv in any year.

Links between radiation and cancer have been discovered after nuclear explosions. Some cancer forms are also slightly more common among other groups exposed to radiation. At the individual level, no direct connection can yet be shown between the small extra radiation doses received from X-ray or isotope examinations and potential later cancers in the subjects.

All of us are exposed to ionising radiation from both natural and manmade sources. The dose of an average Finn is calculated to be 3.7 mSv per year.

This scientific research (ERMA) includes using a DXA device for a full body composition scan and for scanning the neck of the hip bone. The participant is exposed to a small radiation dose that corresponds to a maximum of a few days of natural background radiation or the dose received during a few hours' flight.

The radiation dose you receive from CT scanning (about 0.25 mSv) is about one fourth of estimated yearly natural background radiation.