HOSPITAL DAS CLÍNICAS OF THE UNIVERSITY OF SÃO PAULO MEDICAL SCHOOL – HCFMUSP

INFORMED CONSENT FORM

RESEARCH DATA

Study Title – "**ATTR Amyloid Cardiomyopathy: Characterization of Extracellular Vesicles as Possible Stratifiers and Prognostic Biomarkers of the Disease"**

Principal Investigator – Prof. Dr. Felix José Alvarez Ramires

Department/Institute – Clinical Unit of Cardiomyopathies and Aortic Diseases

Invitation to Participate – You are cordially invited to participate in a study that aims to quantify and characterize serum extracellular vesicles (EVs), which are small particles found in the blood, in patients with different forms of cardiac amyloidosis related to abnormalities in a protein called transthyretin (TTR-CA). The main objective is to investigate the role of these EVs in the development and progression of the disease. Your participation will not only enrich scientific research in this field but may also offer valuable insights for advancing treatments and clinical care for individuals affected by this challenging medical condition.

1. Rationale and Objectives of the Study - This project arises from the urgent need to find more effective ways to identify cardiac amyloidosis early, a condition that often goes unnoticed until symptoms become evident. Early detection is crucial to prevent severe complications.

One promising approach is to analyze information contained in the small particles released by cells, known as extracellular vesicles. These "messengers" carry signals that reflect cellular health and play an essential role in intercellular communication.

Our goal is to identify specific signals within these vesicles that may indicate the presence of cardiac amyloidosis at early stages. We believe this approach will support more effective treatments and significantly improve the quality of life of individuals affected by this often underdiagnosed condition.

Study Group Composition

The study will include three distinct groups of participants: a group of patients diagnosed with TTR amyloid cardiomyopathy with myocardial dysfunction **(TTR-CA + MD Group),** a group of patients diagnosed with TTR amyloidosis with neurological involvement **(TTR-A Group)**, and a group composed of healthy individuals **(CTL Group).**

You will be assigned to one of these groups based on your health condition, as determined by the results of cardiac imaging tests that will assess your heart function and overall cardiovascular health.

2. Procedures and Methods to be Used

During your participation in this study, there will be no changes to your current medical treatment. Exams will be conducted to better understand your clinical condition and determine in which group you fit. These include:

- **Electrocardiogram (ECG):** This test checks how your heart is beating. It shows whether there is any electrical problem in the heart that could affect your heartbeat.
- **Echocardiography with Tissue Doppler and Strain Analysis:** This exam allows us to see how your heart is functioning. It verifies whether the heart muscle is very thick, whether the heart chambers are the appropriate size, and whether all parts of the heart are moving correctly.
- **Technetium Pyrophosphate Scintigraphy:** This exam helps detect whether there is anything abnormal in your heart muscle. It can show whether a substance called amyloid is accumulating in your heart, which may indicate problems.
- **Cardiac Magnetic Resonance Imaging (MRI):** This exam provides a detailed image of your heart. It shows whether the walls of your heart are too thick, whether there is excess fluid around the heart, and whether there are damaged areas in the heart muscle.
- **Blood Collection for Laboratory Analysis and Biomarker Identification in Vesicles:** During the research, we will also collect a small amount of blood for laboratory analysis.

This will help us search for specific substances, called biomarkers, that may be present in extracellular vesicles. These biomarkers act as signals that can tell us more about your health condition and how the disease is progressing.

During the inclusion visit, if you have recently undergone any of the exams required for the study, you will be exempt from repeating those additional tests. In such cases, only your data will be collected to ensure accuracy and reliability of the information. However, it is important to note that blood collection will be performed on all participants for laboratory analysis as part of the essential study procedures. These measures aim to ensure the quality and reliability of the data collected.

4. Explanation of Possible Discomforts and Risks Resulting from Participation

- **Electrocardiogram (ECG):** You may feel slight adherence on the skin due to electrode placement. Although generally safe, there is a very small risk of skin irritation from the adhesive. Rarely, allergic reactions may occur, especially with short-term use.
- **Echocardiography with Tissue Doppler and Strain Analysis:** The conductor gel may feel cold or wet on the skin, but the procedure is painless. This exam is considered safe and non-invasive, with minimal risks. In rare cases, an allergic reaction to the gel may occur.
- **Technetium Pyrophosphate Scintigraphy:** The procedure is usually painless, although the injection of the radiopharmaceutical may cause mild discomfort. This exam is safe, but there is a very low risk of allergic reactions to the radiopharmaceutical. Radiation exposure occurs, but at very low levels that do not cause harm to health.
- **Cardiac Magnetic Resonance Imaging:** The exam may be uncomfortable for individuals with claustrophobia due to the enclosed environment. It is generally safe, but risks may arise with gadolinium contrast use in patients with kidney failure. People with certain medical implants may be restricted due to interaction with magnetic fields.
- **Blood Collection for Laboratory Analysis and Biomarker Identification in Vesicles:** You may feel mild discomfort during needle insertion and a sensation of pressure in the arm. This procedure is safe but carries a small risk of bruising or post-collection dizziness.

All exams required for this research are standard hospital procedures. There are no additional risks from participation. Taking part in this study does not imply any changes in your treatment. As in any clinical study, there may be indirect psychological, moral, intellectual, social, or cultural risks—for example, feeling uncomfortable answering a question. If this occurs, you may refuse to answer without any negative consequences for your follow-up in the study.

To protect your personal data, you will be identified in the study file by a numerical code. All collected information will be handled in a coded manner. Participation records will be kept confidential, with access restricted to individuals directly involved in the study. These professionals will be responsible for transferring clinical information to the forms and ensuring the proper conduct of the study.

Collected data may be used in future research related to the topic. However, these data will not be shared with third parties and will be kept confidential, in compliance with the Brazilian Data Protection Law of August 14, 2018.

Expected Benefits for the Participant – By participating in this study, you will have access to a comprehensive evaluation of your heart health through a series of advanced cardiac exams. These exams will allow us to analyze the functioning of your heart in detail, enabling early identification of any abnormalities related to cardiac amyloidosis or other heart conditions.

Additionally, by taking part in this research, you will directly contribute to the advancement of scientific knowledge regarding cardiac amyloidosis and its early diagnosis. Our investigation of extracellular vesicles as potential disease biomarkers may lead to the development of new diagnostic approaches that are more sensitive and specific, as well as more targeted and effective therapies for treating this challenging medical condition. Your participation is fundamental to improving the understanding and management of cardiac amyloidosis—not only for you but also for others affected by this condition..

5. Clarification Regarding Follow-Up and Assistance Rights:

- ✔ All information obtained in this study will be completely confidential, private, and protected.
- ✔ If you experience any physical harm as a direct result of the procedures performed, you will receive all necessary medical care provided by the Heart Institute. If the Institute is unable to provide such care, you will be reimbursed by the party responsible for the harm, based on usual and reasonable medical expenses incurred in treating the injury. By participating, you agree to cooperate with any health or medical insurance available to you regarding such medical care.
- ✔ If you suffer physical harm as a result of study-required procedures, you will be reimbursed for usual and reasonable medical expenses that are not covered by hospital insurance or other third-party coverage, provided the harm was not caused by your failure to follow instructions in this informed consent form or given by the study team. These medical services must be obtained in the same way you normally receive medical care. No other type of financial compensation (such as lost wages, missed workdays, or discomfort) has been arranged; however, by signing this form, you are not waiving any of your legal rights.

6. Participant Guarantees:

1. **Full Freedom to Refuse or Withdraw:** You have the right to refuse participation or to withdraw consent at any stage of the research, without any penalty or loss of benefits to which you are entitled.
2. **Reimbursement of Expenses:** Any expenses arising from your participation in the research will be duly reimbursed. Details regarding the reimbursement process will be available for your review.

At any stage of the study, you will have access to the professionals responsible for the research to clarify any questions. The principal investigator is Prof. Dr. Félix José Alvarez Ramires, who can be found at Avenida Dr. Enéas de Carvalho Aguiar 44, 5th floor – Block II. Phone: +55 11 99484-9265 and +55 11 2661-5931. If you have any concerns or questions about the ethics of the research, please contact the Research Ethics Committee (CEP) – Rua Ovídio Pires de Campos, 225 – 6th floor – ZIP 05403-905, Phone: (11) 2661-7585, (11) 2661-1548, from 7 a.m. to 4 p.m., Monday through Friday, or by email: cappesq.adm@hc.fm.usp.br.

I have been sufficiently informed about the study "ATTR Amyloid Cardiomyopathy: Characterization of Extracellular Vesicles as Possible Stratifiers and Prognostic Biomarkers of the Disease." I have discussed the above information with the Responsible Researcher (Prof. Dr. Félix José Alvarez Ramires) and/or the person delegated by him (Dr. Camila Rodrigues Moreno) regarding my decision to participate in this study. The objectives, procedures, potential discomforts and risks, and guarantees have been clearly explained to me. I voluntarily agree to participate in this study, sign this informed consent form, and receive a copy initialed by the researcher.

---------- Date____/____/_____

Participant / Legal Representative Signature

---------- Date____/____/_____

Participant / Legal Representative Name

---------- Date ____/____/_____

Study Responsible Signature