CLINICAL STUDY PROTOCOL

MOLECULE:
Semaglutide

TITLE:
Effect of Oral Semaglutide on Epicardial and Pericoronary Adipose Tissues in Type 2 Diabetic Patients After Myocardial Infarction.

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PLACE OF STUDY:
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SUMMARY:

PROCEDURE	Diabetic patients who suffered AMI will be submitted to oral Semaglutide or placebo and temporally compared before and after 6 months of study initially for pericardial and perivascular fat.
TITLE	Effect of Oral Semaglutide on Epicardial and Pericoronary Adipose Tissues in Type 2 Diabetic Patients After Myocardial Infarction.
TARGET DISEASE	Type 2 diabetes
PRIMARY PURPOSE OF THE STUDY	To investigate the ability of oral Semaglutide to reduce pericardial and perivascular fat in type 2 diabetics after myocardial infarction.
STUDY DYNAMICS	Analysis of coronary angiotomography follow-up data before and after 6 months Initially.
STUDY DESIGN	Diabetic patients who suffered acute myocardial infarction will be divided into 2 groups, an arm that will receive medication and another arm placebo (control population). The study flow is prospective and consists of 4 phases.
DURATION	24 months
INTERVENTION	Organize the follow-up of diabetic patients who had an acute myocardial infarction and verify the patients' response to therapy with oral semaglutide. It is expected to obtain relevant data on the influence of pericardial and perivascular fat on the atherosclerotic plaque, increasing the lumen of coronary arteries.

FOLLOW-UP	The time for data collection and analysis will be 24 months.
NUMBER OF PATIENTS	88 patients
CENTERS INVOLVED	Instituto do Coração (InCor): Hospital das Clínicas da Faculdade de Medicina de São Paulo (HCFMUSP)
STUDY GOAL	We believe that a subset of diabetic patients with acute myocardial infarction may benefit from therapy with oral Semaglutide daily.
KEYWORDS	Type 2 Diabetes, Acute myocardial infarction, Coronary CT angiography, Oral Semaglutide, Prognosis.
COST OF THE STUDY	xxxxxxxx

INTRODUCTION AND HYPOTHESIS:

The LEADER [1] study showed that liraglutide significantly decreased major cardiovascular (CV) events (CV death, nonfatal acute myocardial infarction (AMI) or nonfatal stroke), compared to placebo in a group of patients with type 2 diabetes (T2D) with high CV risk.
The SUSTAIN program [2] [3] showed that subcutaneous (sc) semaglutide, a glucagon-like peptide-1 analogue once-weekly for treatment of T2D led to significant reductions in HbA1c and body weight vs all comparators across the SUSTAIN phase 3, a clinical trial programme. Patients treated with Semaglutide had a significant lower risk (26%) of major adverse CV events (MACE: a primary composite outcome of non-fatal myocardial infarction [MI], non-fatal stroke or CV death) vs those receiving placebo over 2 years.
Semaglutide might play an important role in reducing visceral cardiac fat and decreasing the risk of "Major Adverse Cardiovascular Events" (MACE).[4]
The overall safety profile of oral semaglutide was similar in all PIONEER trials and was not unexpected, considering sc semaglutide and other GLP-1 RAs. The main adverse events reported in the PIONEER program were related to the GI tract, but of mild or moderate severity, and most were transient.[5] [6] [7] [8] [9] [10]
The PIONEER 6 study [11] included 3183 T2DM patients who had atherosclerotic cardiovascular desease or who were at high risk, of whom 1591 were treated with oral semaglutide and 1592 were treated with placebo for a median time of 15.9 months. This

study was designed to evaluate the cardiovascular safety of oral semaglutide, and the results confirmed the safety.
Population pharmacokinetic analysis indicated dose-proportional pharmacokinetic profiles for both oral and sc, with body weight being the main factor infuencing exposure and efficacy for HbA1c and body weight, tolerability for nausea and vomiting.[12]
A meta-analysis showed no statistically significant difference in efficacy between the two formulations at week 26, despite the numerically higher HbA1c response and body weight with sc semaglutide [13]

Different terms have been attributed to describe adipose tissue deposited around the heart, they are epicardial, pericardial and paracardiac. [14]
The pericardial fat, the ectopic fat that surrounds the heart, consists of two different compartments of fat separated by the pericardium: intrapericardial fat, that is, Epicardial Adipose Tissue (EAT), located between the myocardium and the visceral serous pericardium; and Extrapericardial Adipose Tissue (XAT), which lies externally to the fibrous parietal pericardium. These fatty tissues differ in their embryonic origin and blood supply. The accumulation of EAT is associated with an increased risk of cardiovascular diseases, cardiac dysfunction, and atrial fibrillation. Extrapericardial Adipose Tissue is associated with obesity-related diseases such as metabolic syndrome, dyslipidemia, and T2D. The modulation of these adipose tissues after weight loss and its relation to cardiometabolic alteration and the risk of cardiovascular disease have gained scientific interest .[15]
EAT can be considered an endocrine organ because it is capable of synthesizing bioactive molecules that are transported to the adjacent myocardium. [16] It is an exclusive compartment of fat between the myocardium and the visceral pericardium that shares a common embryological origin with the deposit of visceral fat. EAT volume has a relatively strong genetic dependence, similar to other visceral fat deposits.
Epicardial fat deposition is associated with increased cardiometabolic risk and coronary events, regardless of the traditional CV risk. Reduction of fat volume in epicardium may be associated with faster improvement and decreased risk of new CV events.
The role of EAT on heart diseases such as coronary artery disease, ventricular dysfunction, arrhythmias, and epicardium has been studied recently. [17 18]
Under physiologic conditions, EAT has a cardioprotective biochemical property, secreting antiatherosclerotic substances. [7]
EAT has several specific roles and its local effects on cardiac function are incorporated into the complex pathological mechanism of coronary artery disease and can produce bioactive substances such as adipocytokines and chemokines, influencing, through paracrine and vasocrine effects, the development and progression of atherosclerosis coronary artery. [19]
The thickness of epicardial fat can be measured easily and accurately and can serve as a marker of visceral adiposity and changes in visceral fat during weight loss interventions and fat-targeted drug treatments.
The potential of modulating epicardial fat with targeted pharmacological agents may open new avenues in the pharmacotherapy of endocrine and metabolic diseases.
EAT coordinates the activation of immune cells, granulopoiesis after myocardial infarction.[20]
Insulin resistance is highly prevalent with atherosclerosis and is associated with an increased risk of AMI and stroke. The IRIS [21 22] study evaluating the insulin resistance intervention with pioglitazone in non-diabetic patients after a recent stroke and AMI, where there was a reduction in the risk of a fatal or non-fatal compound and AMI. Epicardial fat in patients with type 2 diabetes also decreased with SGLT2 (Inhibitor of co-transporter enzyme 2 sodium and glucose) luseoglyphosine. [23]

HOMA-IR (Homeostasis Model Assessment-Insulin Resistance) is evaluated by the following Formula: (Fasting Insulin, PU / mL X Fasting Glycaemia, mmol / L / 22.5) and is a simple way to check for the presence of insulin resistance. [24]
Cystatin C is a non-glycosylated cationic protein which is synthesized as a pre-protein and is produced at a constant rate by all nucleated cells and is present in biological fluids. Cystatin C has been used as a practical alternative to assess renal function [25] and is high in human obesity and produced by adipose tissue. [26]
The accumulation of EAT and the elevation of cystatin C have been independently considered as risk factors that influence atherosclerosis. The strong association between EAT and cystatin C indicates that EAT accumulation may play an important role in cystatin C secretion, possibly contributing to cardiometabolic risk in patients with T2D. [27]
For the quantification of EAT by cardiac computed tomography (CCT) it is more reproducible and correlates better with the extension of Coronary Artery Disease (CAD) than manual two-dimensional measurements. [28]
CCT is a method that can accurately measure the volume of pericardial fat, quantifying the volume of XTA and EAT. [29]
The estimated mean normal volume of EAT observed in CCT is 73.0 mL in man and 64.8 mL in the woman, corresponding to autopsy studies. [30]
The quantification of EAT by CCT and the model based on iterative reconstruction can improve image quality and decrease measurement variability. [31]
Accurate quantification of the EAT volume by an automated algorithm can be determined from non-contrast-enhanced CT. [32]
Most studies suggest that the quantification of EAT is significantly associated with clinical outcomes and provides an incremental prognostic value on coronary artery calcium score. [33]

The epicardial adipose tissue is composed of two compartments. The outermost one – called epicardial adipose tissue and located between the myocardium and the visceral layer of the pericardium – reflects the individual nutritional status.[34] On the other hand, perivascular fat, also called perivascular adipose tissue, is closely related to atherogenesis through the production of inflammatory mediators that act directly on the coronary vessel, such as leptin and resistin.[35, 36] Perivascular adipose tissue both mediates the production of inflammatory cytokines and is expressed as a marker of change in the vascular inflammatory state.[37] Because it changes structurally in response to a greater coronary vascular inflammatory state (lipid x aqueous composition), it is currently easily quantified by software coupled to CT angiography – adipose attenuation index (FAI). [38]

Coronary Computed Tomography Angiography (CTA) is increasingly adding information through the quantification of adipose attenuation index.

New studies have shown that adipose tissue, specifically coronary perivascular adipose tissue, has an additional role in the atherosclerotic process, through the production of pro and anti-inflammatory cytokines in response to vascular inflammation. Quantification of the degree of attenuation of perivascular adipose tissue (PVAT) through CTA is considered an inflammatory biomarker with an important prediction of future cardiovascular events.

Nomura et al.[39] evaluated the correlation between the degree of attenuation of coronary adipose tissue (FAI) with the measurement of coronary flow reserve (CFR) by PET, showing an independent association between high values of FAI (> - 70.1 HU) with a fall in CFR. Furthermore, in patients with coronary calcium score (CCS) < 100, high FAI values were also associated with lower CFR (2.47 ± 0.95 vs. 3.13 ± 0.89, P = 0.003) (21). This finding confirms the viability of this marker as a potential restratifier of cardiovascular risk.

Iacobellis et al [40] showed by ultrasound that EAT thickness measured at baseline and at the 12-week follow-up, that epicardial adipose tissue thickness decreased significantly in both semaglutide and dulaglutide groups ($P < 0.001$) after 12 weeks, accounting for a 20% reduction. Epicardial adipose tissue thickness reduction was significantly greater ($P < 0.01$) with the higher doses of semaglutide (1 mg) and dulaglutide (1.5 mg), respectively,

RATIONALE AND JUSTIFICATION FOR THIS ESTUDY:

This prospective study of diabetic patients with acute myocardial infarction intent to domonstrate the effects of oral semaglutide on pericardial and perivascular coronary fats and the benefit of this study in the development of a future model that can assess the risks of new myocardial infarction with understanding of the long-term prognosis.

RESEARCH LINE:

The project is included in the thematic line of research – Treatment.

SUMMARY OF THE CLINICAL STUDY:

MOLECULE: Semaglutide

TITLE: Effect of Oral Semaglutide Under the Epicardial and Pericoronary Adipose Tissues in Type 2 Diabetic Patients After Myocardial Infarction.

OBJECTIVE: This randomized placebo-controlled study has the primary objective of demonstrating the effect of oral semaglutide on pericardial and pericoronary adipose tissues, atherosclerotic plaque and the vascular lumen in patients with T2D after myocardial infarction through the analysis of CCT, CTA and secondary objective in the analysis of anthropometric markers, cardiac markers and insulin resistance.

MATERIAL AND METHODS:

Equipments: Cardiac Computed Tomography (CCT) and Echocardiography: A standard CTA protocol using a 320 detector-row scanner (Aquillion ONE, Canon Medical Systems, Ottawa, Japan), including coronary calcium score (CCS) and CTA, will be performed. To reach a heart rate < 65 b.p.m. during acquisition, patients will receive either oral (50–100 mg) or intravenous metoprolol (up to 15 mg in 5 mg increments). Fast-acting sublingual nitrate (2.5–5 mg) will also giving to all patients prior to scanning. After CCS acquisition, electrocardiogram-triggered CTA will be performed with 70 mL of non-ionic contrast (Iopromide 370 mg iodine/mL, Bayer Schering Pharma, Berlin, Germany), injected intravenously at 5.0 mL/s, followed by 30–40 mL of saline. CTA parameters as follows: collimation 0.5 mm, rotation time 400 ms, tube voltage and current were 100–120 kV and 250–550 mA, adjusted to body mass index.

To be carried out on Visits 1 and 2:

- Coronary Computed Tomography Angiography (CTA) in the proximal segment of the right coronary and/or anterior descending coronary artery.
- Heart Markers: Troponin I, CK-MB m, Reactive Protein C , Interleucin 6
- Metabolic Markers: The measure of neck circumference, hepatic steatosis (Abdominal USG), Fasting Glycaemia, HbA1c, Basal Insulin and HOMA-IR, Cystatin C, Total Cholesterol and Fractions, Uric Acid, TSH and Free T4.
- Anthropometric and Clinical Markers: Body Weight, Body Mass Index (BMI) = weight / height², Abdominal Waist (AW), AW / Height Ratio, Blood Pressure, and Heart Rate.

STUDY DESIGN:

Single-center, prospective, dubleblinded, placebo-controlled, randomized study with 4 phases:

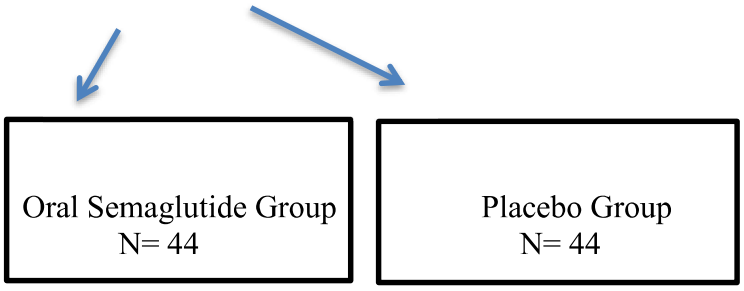
First step:
Screening of chronic patients who had AMI with T2D for more then 1 month and less than 6 months at InCor data base, according to inclusion criteria.
Call the patient for Visit 1 (inclusion). If screening not fail, the patient will be fully informed about this study and read and sign free the TCLE. After that, the investigator will make the clinical consultation, anthropometric data, blood pressure, heart rate will be measured and order the exams of CCT, CTA, Abdominal USG, Echocardiography and other markers described to enter the 1st. study phase and receive randomized oral semaglutide / placebo-controlled treatment at the day of visit 1.

Randomization:
We consider the start of randomization as the first day. The other phases are counted as the days of the start of randomization.

AMI (> 1 month and < 6 months):
1st. Phase ---------- 2nd. Phase----------3rd. Phase----------4th. Phase

1o. day ----------30 days ----------90 days ---------- 180 days
(Visit 1: randomization) (*Phone call) (*Phone call) (Visit 2)

CCT, CTA and ---------- CCT, CTA and
other markers other markers



- Phone calls at the 2nd and 3rd. phases to ask the following questions:

1) Is the patient using the medicine correctly?
   If no, we´ll invite the patient for extra visit.
2) Is the patient having any Adverse Event (AE) ?
   If yes, we'll invite the patient for extra visit.

4th. Phase (Visit 2): The investigator will ask if the patient used correctly the medication/placebo, if occoured AE, make the clinical consultation and order the exams of CCT, CTA, Abdominal USG, Echocardiography and other markers described.

Adverse Event Expectation:

Possibilities with increasing doses:
Nausea, vomiting, rare headache, hypoglycemia, abdominal pain, constipation and diarrhea.
The patient who has an adverse event due to study medication will contact the principal investigator or sub-investigator for instructions and depending on the degree of adverse event (mild, moderate or severe), may be called for extra visit.

Inclusion Criteria:

1. AMI >1 month and <6 months.
2. Signed free and informed consent signed.
3. Female and Male patients and aged> = 18 years.
4. BMI> = 20 and <35
5. Among T2D patients, those with a pre-established diagnosis of fasting glycaemia > 126 mg/dL, and or Hba1c > 6.4% more than 3 months and undergoing treatment.

Exclusion Criteria:

1- Type I diabetics
2- Type II diabetics using inhibitors of SGLT2 or DPP4 or GLP-1 agonist receptors or systemic corticosteroids within 3 months prior this study
3- Chronic Renal Disease defined by GFR <60.
4- Patients who did not participate in the second phase of the study for any reason.
5- Not Treated Hypothyroidism.
6- Cardiac Congestive Insufficiency degree III or IV.
7- AMI > 6 months
8- Any package insert contraindication for the use of AR GLP1/ Oral Semaglutide.
9- Cancer or Chemotherapy
10- Breastfeeding, or the intention of becoming pregnant.

SAMPLE CALCULATION:

Based on the bibliography [40] , patients who used Semaglutide after 12 weeks showed a mean EAT of 7.5 mm (SD = 2 mm), considering that the treatment reduce EAT by at least 15% compared to placebo, with 95% confidence and 80% of power, the sample required to carry out the study is 36 patients in each group. [41]
Assuming that there are losses to follow-up in the order of 20%, 44 patients should be included in each group. The calculation was performed considering the two-tailed test.

STATISTICAL ANALYSIS:

Qualitative characteristics will be described according to groups using absolute and relative frequencies and the association between groups will be verified at visit 1 using chi-square tests or exact tests, quantitative characteristics will be described according to groups using summary measures (mean, standard deviation, median, minimum and maximum) and compared at baseline using Student's t-test or Mann-Whitney tests according to the probability distribution of the data.
The characteristics of interest will be described according to groups throughout the follow-up using summary measures and compared between groups and evaluation moments using generalized estimation equations with adequate distributions and linkages and followed by Bonferroni multiple comparisons when necessary.
The IBM-SPSS for Windows version 22.0 software will be used for the analysis and the Microsoft Excel 2010 software will be used for data tabulation. The tests will be performed with a significance level of 5%.

GROUPS OF STUDY

(Selection of patients): N= 88 patients

A- Semaglutide Group (44 patients):

1- Patients with T2D:
Patients will receive oral semaglutide at the following doses:
3 mg/day/4 weeks (1 box of 3 mg). Then 7 mg/day/+ 4 weeks (1 box of 7 mg). Then 14 mg/day or the dose of patient supported until the end of the study (4 boxes of 14 mg).

B- Placebo Group (44 patients):

1- Patients with T2D:
Patient will receive oral placebo the following presumed doses:
3 mg/day/4 weeks (1 box of 3 mg). Then 7 mg/day/+ 4 weeks (1 box of 7 mg). Then 14 mg/day until the end of the study (4 boxes of 14 mg).

PHASES OF THE STUDY

First Phase (Visit 1)	Chronic patients who have suffered AMI who meet the inclusion criteria and will perform CCT, CTA and collection of cardiac, metabolic, anthropometric and clinical markers. (Basal). The first day of randomization is the begining of the first phase.
Second Phase	Phone call 30 days after randomization.
Third Phase	Phone call 90 days after randomization.
Fourth Phase (Visit 2)	Evaluation of these patients in the long term (180 days) with the study of the same parameters evaluated in the first phase by the same medical doctor.

STUDY DURATION

Period	Activity
July 2022 to July 2023	Recruitment of participants
July 2022 to July 2023	Selection and inclusion of patients
July 2022 to Julho 2023	Randomization of patients
July 2022 to July 2023	Data collection
July 2023 to December 2023	Data Analysis
January 2024 to December 2024	Writting the scientific article
January 2025 to March 2025	Sending to publication of the scientific article

Interruption Criteria:

1- Important side effects from the use of oral semaglutide such as diarrhea or vomiting not overcome after 7 days of side effect.
2- A higher incidence of MACE in the semaglutide group in relation to the placebo group.

3- Interruption of financing

PROJECT WORKPLAN

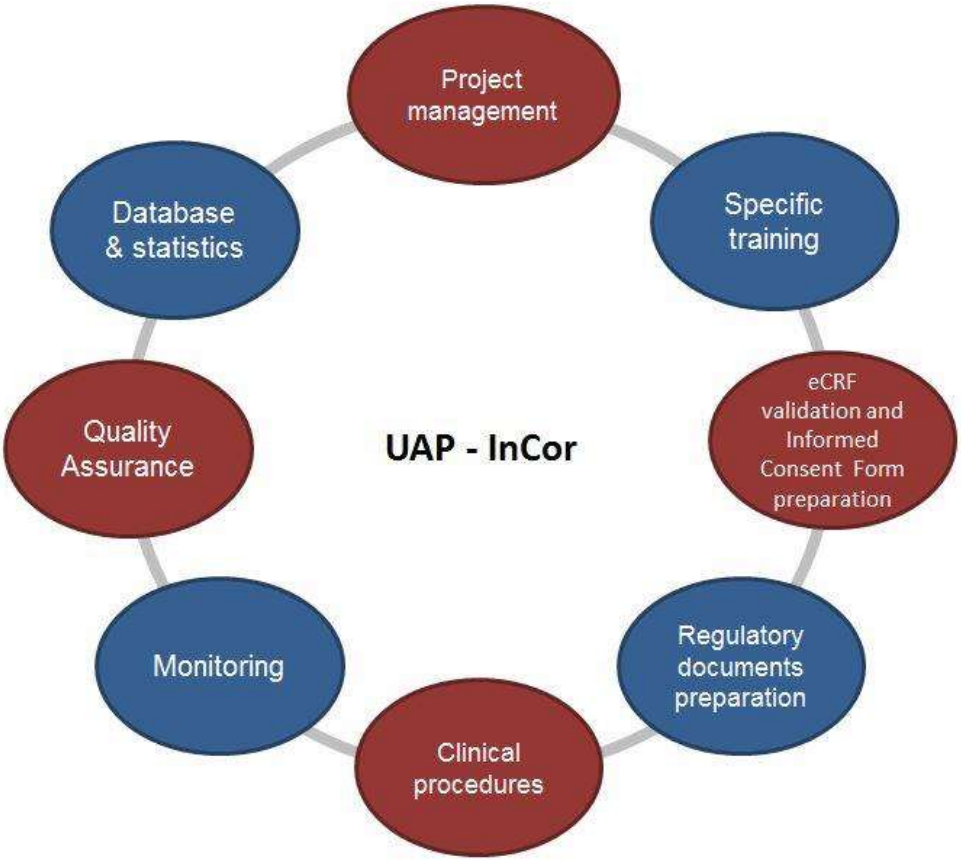
A. RESEARCH SUPPOR UNIT

The Research Support Unit (UAP) is a supporting unit from the Heart Institute of University of Sao Paulo Medical School (InCor), created to act on the assistance and development of research protocols, by providing support to researchers from various specialties and/or interested parties regarding the project structuring, submission, conduction, monitoring and close-out processes. The UAP operation aims the proper development of the Clinical Research protocols, including the Management, Planning, Execution, and Monitoring of Clinical Trials at national and international levels, providing support and advice to the contracting entity regarding the development of studies within the current regulatory norms.

Project Management and data collection, as well as monitoring of clinical studies, in accordance with the guidelines of Good Clinical Practices (ICH-GCP), is necessary due to the need to supervise the progress of the research, ensuring that it is conducted, recorded and reported in accordance to the national and international established procedures.

Figure 1. Activities developed by UAP-InCor in the project assistance and monitoring

Schedule of activities



The information presented in Figure 1 is related to the tasks assigned to the UAP-InCor team, which will be vigorously applied during the project, aiming to guide, advise and monitor the actions of the research teams of the clinical research center for the proper conduct of the study. The UAP-InCor will perform the following activities to ensure that the development and execution of the trial are conducted properly.

B. SCOPE OF THE WORK PLAN

This Work Plan is designed to determine the essential requirements for the development of the activities foreseen in this document, taking into account the specific period of execution of the study stages, always using the UAP-InCor practices record. Therefore, it is established in this planning, execution and monitoring process that the UAP-InCor team is responsible for the follow-up of clinical studies in order to guarantee the essential quality standards in conducting the research.
Therefore, UAP-InCor is committed to the following designations:

- Establish essential requirements for the development of the activities foreseen in this Work Plan;
- Follow the schedule to execute the planned steps;
- Record the activities performed by UAP-InCor as well as conducting the study at its site;
- Elaborate relevant records that prove the performance of the monitoring and ensure the traceability of the actions taken.

C. STUDY RESPONSABILITIES

C.1. Clinical Research Monitor - The Clinical Research Monitor is the professionally qualified and responsible for ensuring that the clinical study is conducted in accordance with the clinical protocol approved by the appropriate regulatory bodies, the GCP-ICH guidelines, the involved POPs, and local and international regulations.
The Clinical Research Monitor should ensure that the study is conducted and documented correctly through the performance of the activities, whenever relevant and necessary to the study and the Clinical Research Center (CPC).
The Clinical Research Monitor, in addition to overseeing the overall conduct of the study, is also responsible for managing and verifying the data and preparing the study report providing the necessary support to the Unit's activities. It is the responsibility of this professional to link all the stages of the study and ensure compliance with the stipulated deadlines.

C.2. Clinical Research Doctor - The clinical research physician is the professional qualified to conduct clinical visits and discuss cases in accordance with the Protocol and Good Clinical Practices. In addition to contributing to the evaluation and classification of adverse events and medical doubts, they provide support and perform each procedure and/or visits according to the specific study and research project.

D. UAP - DESCRIPTION OF ACTIVITIES

D.1. Review of the Clinical Protocol - Adequacy and formatting of the final Clinical Protocol according to Good Clinical Practices and relevant legislation, if necessary.

D.2. Elaboration of ICF – Development of the Informed Consent Form specific for the study , ensuring that is presented in a clear, objective and complete form according to the pertinent legislation, prior to ethics evaluation.

D.3. Development of regulatory documents – Development and elaboration of documents that comprise the regulatory dossier, which will highlight the importance and clarify critical details of the study for the Ethics Committee.

D.4. Preparation of study control documents - Prepare the forms, spreadsheets, and logs that will register the activities within the good clinical practices during the course of the study.

D.5. Data monitoring - The scheduling of data monitoring will be established according to the prediction of the inclusion of participants to ensure that the information collected during the execution of the study is trustworthy and reflects the facts.

D.6. Study Close-out - The close-out visit is performed when all the research subject have successfully complete all follow-up activities predicted in the study execution schedule and the developments are communicated to the Ethics Committee. A final report must be issued in 7 (seven) business days.

References:

[1] Marso S, Nauck M, Monk Fries T, Rasmussen S, Treppendahl M, Buse J. Myocardial Infarction Subtypes in Patients With Type 2 Diabetes Mellitus and the Effect of Liraglutide Therapy (from the LEADER Trial). The American Journal of Cardiology. 2018;121(12):1467-1470

[2] Bain S, Rea R, Warren M, Holst A, Vrazic H, Madsbad S. P2859 Semaglutide consistently reduces cardiovascular risk in patients with type 2 diabetes regardless of baseline cardiovascular risk level: post hoc analyses of the SUSTAIN trial programme. European Heart Journal. 2018;39(suppl_1).

[3] Aroda VR, et al. Comparative efficacy, safety, and cardiovascular outcomes with once-weekly subcutaneous semaglutide in the treatment of type 2 diabetes: Insights from the SUSTAIN 1–7 trials. Diabetes Metab. 2019;45(5):409–18.

[4] Marso SP, Bain SC, Consoli A, Eliaschewitz FG, Jódar E, Leiter LA, et al. Semaglutide and cardiovascular outcomes in patients with type 2 diabetes. N Engl J Med. 2016;375:1834–44.

[5] Rodbard HW, et al. Oral semaglutide versus empagliflozin in patients with type 2 diabetes uncontrolled on metformin: the PIONEER 2 trial. Diabetes Care. 2019;42(12):2272–81.

[6] Rosenstock J, et al. Effect of additional oral semaglutide vs sitagliptin on glycated hemoglobin in adults with type 2 diabetes uncontrolled with metformin alone or with sulfonylurea: the PIONEER 3 randomized clinical trial. JAMA. 2019;321(15):1466–80.

[7] Pratley R, et al. Oral semaglutide versus subcutaneous liraglutide and placebo in type 2 diabetes (PIONEER 4): a randomised, double-blind, phase 3a trial. Lancet. 2019;394(10192):39–50.

[8] Mosenzon O, et al. Efficacy and safety of oral semaglutide in patients with type 2 diabetes and moderate renal impairment (PIONEER 5): a placebo-controlled, randomised, phase 3a trial. Lancet Diabetes Endocrinol. 2019;7(7):515–27.

[9] Pieber TR, et al. Efficacy and safety of oral semaglutide with flexible dose adjustment versus sitagliptin in type 2 diabetes (PIONEER 7): a multicentre, open-label, randomised, phase 3a trial. Lancet Diabetes Endocrinol. 2019;7(7):528–39.

[10] Zinman B, et al. Efficacy, safety, and tolerability of oral semaglutide versus placebo added to insulin with or without metformin in patients with type 2 diabetes: the PIONEER 8 trial. Diabetes Care. 2019;42(12):2262–71.

[11] Husain M, Birkenfeld AL, Donsmark M, Dungan K, Eliaschewitz FG, Franco DR, et al. Oral semaglutide and cardiovascular outcomes in patients with type 2 diabetes. N Engl J Med. 2019;381:841–51.

[12] Overgaard RV, Navarria A, Hertz CL, Ingwersen SH. Similar eficacy and gastrointestinal tolerability versus exposure for oral and subcutaneous semaglutide. Diabetologia. 2019;62:S375.

[13] Nuhoho S, Gupta J, Hansen BB, Fletcher-Louis M, Dang-Tan T, Paine A. Orally administered semaglutide versus GLP-1 RAs in patients with type 2 diabetes previously receiving 1–2 oral antidiabetics: systematic review and network meta-analysis. Diabetes Ther. 2019;10(6):2183–99.

[14] Patel V, Shah S, Verma S, Oudit G. Epicardial adipose tissue as a metabolic transducer: role in heart failure and coronary artery disease. Heart Failure Reviews. 2017;22(6):889-902.

[15] Tsaban G, Wolak A, Avni-Hassid H, Gepner Y, Shelef I, Henkin Y et al. Dynamics of intrapericardial and extrapericardial fat tissues during long-term, dietary-induced, moderate weight loss. The American Journal of Clinical Nutrition. 2017;106(4):984-995.

[16] Iacobellis G. Epicardial adipose tissue in endocrine and metabolic diseases. Endocrine. 2013;46(1):8-15.

[17] Lazaros G, Antonopoulos A, Antoniades C, Tousoulis D. The Role of Epicardial Fat in Pericardial Diseases. Current Cardiology Reports. 2018;20(6).

[18] Russo R, Di Iorio B, Di Lullo L, Russo D. Epicardial adipose tissue: new parameter for cardiovascular risk assessment in high risk populations. Journal of Nephrology. 2018;31(6):847-853.

[19] Nagy E, Jermendy A, Merkely B, Maurovich-Horvat P. Clinical importance of epicardial adipose tissue. Archives of Medical Science. 2017;4:864-874.

[20] Horckmans M, Bianchini M, Santovito D, Megens R, Springael J, Negri I et al. Pericardial Adipose Tissue Regulates Granulopoiesis, Fibrosis, and Cardiac Function After Myocardial Infarction. Circulation. 2018;137(9):948-960.

[21] Young L, Viscoli C, Curtis J, Inzucchi S, Schwartz G, Lovejoy A et al. Cardiac Outcomes After Ischemic Stroke or Transient Ischemic Attack. Circulation. 2017;135(20):1882-1893.

[22] Kernan W, Viscoli C, Furie K, Young L, Inzucchi S, Gorman M et al. Pioglitazone after Ischemic Stroke or Transient Ischemic Attack. New England Journal of Medicine. 2016;374(14):1321-1331.

[23] Bouchi R, Terashima M, Sasahara Y, Asakawa M, Fukuda T, Takeuchi T et al. Luseogliflozin reduces epicardial fat accumulation in patients with type 2 diabetes: a pilot study. Cardiovascular Diabetology. 2017;16(1).

[24] Ascaso J, Pardo S, Real J, Lorente R, Priego A, Carmena R. Diagnosing Insulin Resistance by Simple Quantitative Methods in Subjects With Normal Glucose. Metabolism Diabetes Care. 2003;26(12):3320-3325.

[25] Gabriel I, Nishida S, Kirsztajn G. Cistatina C sérica: uma alternativa prática para avaliação de função renal?. Brazilian Journal of Nephrology. 2011;33(2):261-267.

[26] Guerre-Millo M. Serum Cystatin C Concentrations Are Increased in Human Obesity in Relation to Over-Production by the Adipose Tissue. Obesity. 2012;20(9):1755-1755.

[27] Murai T, Takebe N, Nagasawa K, Todate Y, Nakagawa R, Nakano R et al. Association of epicardial adipose tissue with serum level of cystatin C in type 2 diabetes. PLOS ONE. 2017;12(9):e0184723.

[28] Bastarrika G, Broncano J, Schoepf U, Schwarz F, Lee Y, Abro J et al. Relationship Between Coronary Artery Disease and Epicardial Adipose Tissue Quantification at Cardiac CT. Academic Radiology. 2010;17(6):727-734.

[29] Lazaros G, Antonopoulos A, Antoniades C, Tousoulis D. The Role of Epicardial Fat in Pericardial Diseases. Current Cardiology Reports. 2018;20(6).

[30] Hindsø L, Jakobsen L, Jacobsen C, Lynnerup N, Banner J. Epicardial adipose tissue volume estimation by postmortem computed tomography of eviscerated hearts. Forensic Science, Medicine and Pathology. 2017;13(4):468-472.

[31] Oda S, Utsunomiya D, Funama Y, Yuki H, Kidoh M, Nakaura T et al. Effect of iterative reconstruction on variability and reproducibility of epicardial fat volume quantification by cardiac CT. Journal of Cardiovascular Computed Tomography. 2016;10(2):150-155.

[32] Ding X, Terzopoulos D, Diaz-Zamudio M, Berman D, Slomka P, Dey D. Automated pericardium delineation and epicardial fat volume quantification from noncontrast CT. Medical Physics. 2015;42(9):5015-5026.

[33] Spearman J, Renker M, Schoepf U, Krazinski A, Herbert T, De Cecco C et al. Prognostic value of epicardial fat volume measurements by computed tomography: a systematic review of the literature. European Radiology. 2015;25(11):3372-3381.

[34] Iacobellis G. Local and systemic effects of the multifaceted epicardial adipose tissue depot. Nat Rev Endocrinol. 2015;11:363–371

[35] Antoniades C, Shirodaria C, Warrick N, et al. 5-Methyltetrahydrofolate rapidly improves endothelial function and decreases superoxide production in human vessels. Circulation. 2006;114:1193–1201.

[36] Margaritis M, Sanna F, Lazaros G, et al. Predictive value of telomere length on outcome following acute myocardial infarction: evidence for contrasting effects of vascular vs. blood oxidative stress. Eur Heart J. 2017;38:3094–3104.

[37] Oikonomou EK, Antoniades C. The role of adipose tissue in cardiovascular health and disease. Nat Rev Cardiol. 2018:1.

[38] Antonopoulos AS, Sanna F, Sabharwal N, Thomas S, Oikonomou EK, Herdman L et al. Detecting human coronary inflammation by imaging perivascular fat. Sci Transl Med 2017;9:eaal2658.

[39] Nomura CH., Assuncao-Jr AN.., Guimaraes PO., Liberato G., Morais T. et al. Association between perivascular inflammation and downstreammyocardial perfusion in patients with suspected coronary artery disease. European Heart Journal - Cardiovascular Imaging 2020 (21), 599–605.

[40] Iacobellis, G, Villasante Fricke, AC. Effects of semaglutide versus dulaglutide on epicardial fat thickness in subjects with type 2 diabetes and obesity. J Endocr Soc. 2020;4(4):bvz042.

[41] Kirkwood, B. R. and Sterne, J. A. C (2006). Essential medical statistics. 2nd ed. Blackwell Science: Massachusetts, USA. p.502.