SRTP - Project Description Form #234

PART I:

Name of Schulich faculty member who will supervise the project	Mark Chandy
Supervisor's Schulich, Western, Hospital or Lawson Email	mchandy2@uwo.ca
Schulich Department	Medicine

PART II - Project Description

Title of Project	Investigating the Effects of Novel Cannabinoids on the Cardiovascular System using iPSC Disease Modeling

Background

Epidemiological studies reveal that cannabis use increases the risk of atherosclerosis. Delta-9-tetrahydrocannabinol (Δ9-THC) binds cannabinoid receptor 1 (CB1) in the vasculature and is implicated in atherosclerosis. Delta-8-tetrahydrocannabinol (Δ8-THC), hexahydrocannabinol (HHC), and cannabidiol (CBD) are frequently used and circumvent regulation because they are derived from hemp. Due to regulatory restrictions, the cardiovascular effects of these cannabinoids are unknown, but like Δ9-THC, they function via the CB1 receptor and are likely to cause cardiovascular disease. The cardiovascular effects of novel cannabinoids might not be evident in epidemiological studies for decades. Induced pluripotent stem cells (iPSCs) are a novel disease model that captures an individual's genetic information and can be differentiated into a limitless supply of cardiovascular tissue. Using iPSC-derived endothelial cells (iPSC-ECs) and iPSC-derived vascular smooth muscle cells (iPSC-VSMCs), we will elucidate the effects of novel cannabinoids on the cardiovascular system.

Hypothesis

We hypothesize that novel cannabinoids will cause endothelial dysfunction, atherosclerosis, and cardiovascular disease.

Proposed Methodology

To investigate the effects of novel cannabinoids in iPSC-derived vascular cells, we will recruit cannabis users and obtain blood samples (n=5). Peripheral blood mononuclear cells will be reprogrammed into iPSCs. Environmental insults trigger changes in cellular phenotype that are measured by functional assays, epigenetic changes, and transcriptomic changes. We will assess the effects of Δ8-THC, CBD, and HHC using iPSC-ECs (n=5). We will measure the dose-dependent effects of each cannabinoid on iPSC-ECs using cell viability, reactive oxygen species (ROS) production, and bulk RNA sequencing analysis for markers of inflammation and oxidative stress. Using the electrical cell-substrate impedance sensing (ECIS) platform, the effects of each cannabinoid on iPSC-ECs will be assessed for endothelial cell permeability and wound healing. In addition, we will expose iPSC-ECs to serum from cannabis users and perform qPCR for markers of inflammation.

Expected Outcomes

The comprehensive functional and transcriptomic analysis will reveal the cardiovascular effects of novel cannabinoids on the cardiovascular system.

Research Environment – Description of the number of research personnel, primary location of research, size of lab, etc

The project will be carried out in the Cardiology and Critical Care Research Program (C3RP) laboratory (www.c3rp.org) at Robarts Research Institute under the supervision of Dr. Mark Chandy and the co-supervision of Dr. Aleksandra Leligdowicz. There will be opportunities to interact with lab managers, lab technicians, graduate students, postdoctoral

fellows, and other scientists at Robarts, as well as in the Department of Microbiology and Immunology and the Department of Physiology and Pharmacology. The Robarts Research Institute is a rich academic environment and is located next to the University Hospital, providing an ideal location for translation biology studies. The experiments will be carried out in the biosafety level-2 laboratory and all required training will be provided but theC3RP laboratory team (https://c3rp.org/team).

Names and titles of other individuals who will be involved with the research project?
Dr. Mark Chandy, MD PhD, Cardiology, LHSC University Hospital
Dr. Aleks Leligdowicz, MD PhD, Critical Care Medicine, LHSC University Hospital
Dr. Kerry-Ann Nakrieko, Robarts Research Institute, C3RP laboratory manager

Can this project be done remotely?	No
Duration of Project	Two Summers

Expected Objectives/Accomplishments for Student for Year 1?
1) Learn how to isolate peripheral blood mononuclear cells (PBMCs) from whole blood
2) Learn how to generate induced pluripotent stem cells (iPSC)
3) Learn how to differentiate endothelial cells from iPSC
4) Learn how to use the electric cell-substrate impedance sensing (ECIS) platform
5) Optimize the ECIS platform with healthy-control plasma

Expected Objectives/Accomplishments for Student for Year 2?
1) Propagate iPSC-derived endothelial cells from 5 cannabis users.
2) Test the effects of novel cannabinoids on iPSC-derived endothelial cells.
3) Investigate the effect of cannabis serum exposure to autologous iPSC-derived endothelial cells.
4) Compare responses of iPSC-derived endothelial cells to novel cannabinoids and serum.
5) Disseminate findings in meetings and prepare a manuscript.

PART III - Certifications

If the project will require any certification approvals from one or more of the following offices, please check the appropriate box below.	- Human Ethics - Biohazard
Human Ethics: If you have the protocol information, please enter it below (or enter the status of the approval).	WREM number 122440. Approved (R-23-462).
Biohazard: If you have the protocol information, please enter it below (or enter the status of the approval).	Biosafety Approval Number: BIO-RRI-0085

Note: certification approval should be obtained prior to the start of the summer. Projects without this approval will not be a priority for funding.