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**THE INFLUENCE OF**

**DIETARY FATTY ACIDS**

**ON CARDIAC FUNCTION**

A thesis submitted for the degree of

**DOCTOR OF PHILOSOPHY**

by

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**November**

**1991**

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## ABBREVIATIONS

$\delta P/\delta t$	Pressure-Time Integral
$\mu\text{mol}$	micro moles
$\Sigma$	sum of
AA	arachidonic acid
ADP	adenosine diphosphate
AL	afterload
ATP	adenosine triphosphate
bpm	beats per minute
C	control group
CCCP	carbonyl cyanide- <i>m</i> -chlorophenyl hydrazone
CHD	ischaemic coronary heart disease
CPP	coronary perfusion pressure
DHA	docosahexaenoic acid
DMA	dimethyl acetate
dw	dry weight
E-C	excitation-contraction
ECG	electrocardiogram
EPA	eicosapentaenoic acid
FO	polyunsaturated fish oil
g	gram
Hz	hertz
J	joules
K-H	Krebs-Henseleit solution
Kg	kilogram

## A B S T R A C T

The aim was to study the direct effects of dietary fatty acids on myocardial function in rats. In particular, the effects of dietary fat intake on cardiac function stressed with myocardial ischaemia and reperfusion were investigated. Although dietary fat can influence the development of atherosclerosis, thrombosis, cardiac ischaemia and myocardial infarction, little is known of any direct effects by dietary fat on cardiac performance. It has also been clearly demonstrated that the fatty acid profile of myocardial phospholipids are predominantly dependent on the qualitative properties of dietary lipid intake. Such alterations in the cellular lipid environment may be associated with a direct dietary fatty acid influence on cardiac function.

Hooded-Wistar rats (4months old) were placed into 3 dietary groups: REF, a reference base diet or a 12% (w/w) addition of either saturated fatty acid rich sheep fat (SAT) or fish oil rich in polyunsaturated marine n-3 fatty acids (FO). Animals were maintained on the diets for a minimum of four months prior to experimental use. In order to directly study cardiac function and precisely control electrolyte and metabolic substrate availability, neural and humoral factors, preload, workload, humidity and temperature, the isolated working heart method was selected. To overcome limitations which reduce the suitability of most isolated working heart models of global ischaemia for the study of the progression of ischaemic injury, a new model of low flow global ischaemia was developed. This method did not cause total cessation of ventricular function or coronary flow and thus permitted investigation of ischaemic processes as they occurred, by simultaneous

measurement of ventricular function, oxygen uptake and metabolite release in venous outflow. This new model utilised a novel placement of two valves to permit coronary perfusion pressure reductions with maintained afterload, to provide a greater ischaemic insult yet allowing simultaneous functional and metabolic evaluation. In addition, a buffer with washed porcine erythrocytes at 40% haematocrit in a modified Krebs-Henseleit/dextran solution was utilised for improved oxygen delivery, viscosity, colloid osmotic pressure (to reduce oedema) and improved mechanical performance on which to impose the ischaemic insult.

Under control conditions, compared to REF hearts, SAT hearts demonstrated an elevated  $MVO_2$  with no performance dividend while FO hearts had reduced  $MVO_2$  with no performance deficit. The higher oxygen delivery in SAT hearts was achieved by intrinsically raised coronary flow. Ischaemic production of lactate, cellular efflux of  $K^+$ , creatine kinase, development of venous acidosis and increased arrhythmia vulnerability were enhanced in SAT hearts and reduced in FO hearts. Better post-ischaemic recovery of functional performance was evident in reperfused FO than in SAT hearts. A paradoxical increase in  $MVO_2$  (despite reduced coronary flow, contractility and external work) was observed during ischaemia and reperfusion in all groups except SAT hearts. However,  $MVO_2$  remained higher in SAT hearts during ischaemia and reperfusion compared to REF and FO hearts. The dietary differences in  $MVO_2$  were still evident following equalisation of coronary flow with hydralazine but were abolished in  $K^+$  arrested hearts. Maintenance of a constant diet-related  $MVO_2$  differential



despite work related increases and during contractile inhibition by ryanodine suggest an activation-dependent mechanism not linked to contraction. Rather the abolition of the high  $MVO_2$  in SAT hearts by ruthenium red indicates a role of mitochondrial  $Ca^{++}$ .

This thesis study has demonstrated the advantages of utilising an isolated working heart method which uses an erythrocyte buffer that allows oxygenation in the physiological range and a method of global ischaemia that is more appropriate for the study of the progression of ischaemic injury. The new model enhanced the capacity to control and directly monitor experimental ischaemic events in progress in a manner that may be more physiologically relevant than previous models and permitted observations that would have not been possible by alternative methods. Although definitive identification of the link between myocardial membrane fatty acid composition and intracellular functional changes was not provided by this study, the results confirm and provide a possible basis for the widely reported antiarrhythmic or proarrhythmic actions of fish oil or fatty acids respectively.