UNIVERSITI TEKNOLOGI MARA

EFFECT OF ANTIOXIDANTS ON CORONARY RISK MARKERS IN HYPERCHOLESTEROLAEMIA

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MSc

December 2009

CANDIDATE'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and the result of my own, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any other degree qualification.

In the event that my thesis is found to violate the conditions mentioned above, I voluntarily waive the right of conferment of my degree and agree to be subjected to the disciplinary rules and regulations of Universiti of Teknologi MARA.

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ABSTRACT

Introduction: Antioxidant vitamins have been suggested to play a role in preventing atherosclerosis. Several vitamins with anti-oxidant properties, such as vitamin E and C, are thought to act cooperatively and possibly synergistically *in vivo*. However, the efficacy of tocotrienol, a potent vitamin E molecule as compared to α-tocopherol, in reducing risk of heart disease has not been fully explored. The optimal dose of tocotrienol in atherogenesis and the synergism of tocotrienol with vitamin C remain unclear.

Objectives: (i) To study the effects of tocotrienol-rich fraction (TRF) supplement on fasting lipid profiles (FSL), oxidative stress, inflammation and atherosclerotic lesions in hypercholesterolaemic (HC) rabbits, (ii) To determine the dose response relationship of TRF treated HC rabbits, (iii) To examine the effects of antioxidants (TRF plus vitamin C) on the inflammatory markers and endothelial dysfunction (ED) in statin treated HC patients with high coronary risk.

Materials and Methods:

(i) Animal model experiment: Twenty-eight male New Zealand white rabbits were given 1% cholesterol diet for 5 months and randomised from the second month onwards into 5 groups: Placebo (n=7), TRF 15 mg/kg (n=5), TRF 30 mg/kg (n=6), TRF 60 mg/kg (n=5) and TRF 90 mg/kg (n=5) daily. Serum FSL, C-reactive protein (CRP), malondialdehyde (MDA) and 8-Isoprostane levels were measured at baseline (BL), 1 and 2 months post-HCD, 1, 2 and 3 months post-intervention. Aortic vessels were obtained to assess the atherosclerotic lesions and immunohistochemical studies for Intercellular Adhesion Molecule-1 (ICAM-1) and Nuclear Factor Kappa-B (NFκ B) were performed.

(ii) Clinical trial: Twenty-nine HC patients were identified in high risk category according to the National Cholesterol Education Programme Adult Panel Treatment III and treated with atorvastatin to achieve low density lipoprotein (LDL-c) target (LDL-c < 2.6 mmol/L) before being randomised into a double-blinded placebo-control clinical trial with 2 groups: Placebo and combined tocotrienols-vitamin C supplement (TRF-160mg plus C-500mg daily) for 3 months. FSL, high sensitivity CRP (hsCRP), interleukin-6 (IL-6), tumor necrosis factor alpha (TNF-α) levels and ED by brachial artery flow mediated Dilatation (FMD) were measured at entry, BL, 2 weeks and 3 months post-intervention.

Results:

(i) Animal model experiment: There were no differences in percentage changes of FSL, MDA, 8-Isoprostane, CRP levels and extent of atherosclerosis between the placebo and TRF groups. Reduced tissue expressions of ICAM-1 (57% vs. 20%, 29%, 50%, 25%: placebo vs. TRF 15, 30, 60, 90 mg/kg) and NFκB (67% vs. 40%, 50%, 25%: placebo vs. TRF 15, 30, 90 mg/kg) were found with significant decrease in area of ICAM-1 expression (Mean SEM; 3.8 2.0 % vs. 25.1 19.5 %, $p<0.05$) between the placebo and TRF 15 mg/kg groups.

(ii) Clinical trial: There were no differences in percent changes of FSL, hsCRP, IL-6, sICAM-1, sVCAM-1, E-selectin levels and percentage of FMD between the placebo and combined supplement groups. Significantly higher endothelial-dependent FMD were found between the antioxidant and placebo group at 3 months (Mean SEM; 8.8 1.6 % vs. 4.7 1.0 %, $p<0.05$).

ACKNOWLEDGEMENT

With great thanks to God and His Blessings, I have been able to experience lots of difficulties and new things during this degree of Master programme.

After all the difficult time, tears and disappointments, finally, I have completed this study and come up with this thesis. None of the project can be completed without the help, cooperation and support of many individuals. I am deeply indebted to all who have directly or indirectly assisted me towards the completion of this study.

First and foremost, I would like to express my most sincere appreciation and deepest gratitude to my supervisor, Prof Dr Hapizah Mohd Nawawi, for her encouragement, guidance, advice, constructive criticisms and suggestions. Without her, I would not be able to present this work as it is today.

I wish to extent my heartiest thanks to my dear parents, Ir Dr A. Razak Yaacob and Mrs Sarifah Abdullah and my entire siblings for their endless prayers, support, encouragement and sacrifice. I also wish to express my gratitude to my beloved auntie, Prof Dr Maimon Abdullah for making me appreciate my examiners on their precious comments and proof read my draft of the corrected thesis, even in the very nick of time.

Sincere thanks to all closed researchers, Prof Dato' Dr Khalid Yusoff, Dr Tengku Saifudin Tengku Ismail, Associate Prof Dr Zaiton Zakaria, Dr Effat Omar, Mrs Rafezah Razali and Mrs Suhaila Abd Muid, for your great dedication in the joint research collaboration, kindness in thoroughly scrutinized this write up and supportive efforts. To Prof Dr Nafeeza Mohd Ismail, Dr Muhammad Huzaimi Haron, Dr Sushil Kumar, Associate Prof Dr Normalina Mansor and Dr Anis Safura Ramli, special appreciation is extended for your committed cooperation in running the animal model experiment and clinical trial.

Beside that, I am really grateful for all the sweet and sour memories in conducting research with all the staff of the animal house, clinical trial centre and laboratories. I truly hope that this study will benefit greatly to the research community and enlighten the future direction in the related research area.

TABLE OF CONTENTS