

Histological And Immunohistochemical Study On The Effect Of Green Tea On Thyroid Gland Of Rat In Overconsumption And Amiodarone Induced Damage

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*I would like to
present this work to*

My Mother

&

My Family

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Abstract

The present study aimed at investigating the possible protective effect of green tea on thyroid damage. This was accomplished by the model of amiodarone induced thyroid damage in albino rat. The possible thyroid changes secondary to green tea overconsumption were also determined.

Twenty four adult male albino rats were divided into 5 groups: Group 1 (Control Group) of 4 rats. Group 2 (Green Tea Group) of 5 rats, each given 50mg/kg of green tea extract tablets dissolved in 0.5 ml distilled water orally. Group 3 (Overconsumption Group) of 5 rats, each given 1000mg/kg of green tea extract dissolved in 2.5ml distilled water orally. Group 4 (Amiodarone Group) of 5 rats, each given 30mg/kg daily orally. Group 5 (Amiodarone and Green Tea Group) of 5 rats, each given amiodarone 30mg/kg and green tea extract 50mg/kg daily orally. Thyroid function tests were performed. Rats were sacrificed after 6 weeks. Thyroid sections were subjected to histological, histochemical, immunohistochemical and morphometric studies. A serological study and statistical analysis were also performed.

Group 3 demonstrated some follicles with cellular masses extending into the lumen, some other distorted follicles, few follicles with dislodged nuclei or cellular debris into the lumen, few follicles containing crescents of colloid and some follicles devoid of colloid. Group 4 recruited multiple follicles with crescents of colloid and multiple follicles exhibiting colloid with large peripheral vacuolations. Some follicles were focally lined by more than one layer of nuclei and occasional follicles were lined by more than one layer of nuclei. Vacuolated cytoplasm and dark nuclei were observed in multiple lining follicular cells in both group 3 and 4. In group 5 multiple lobules contained few follicles with large peripheral vacuolations of colloid. Few lobules demonstrated some follicles containing crescents of colloid. Increased interlobular and interfollicular collagen fibers were found in groups 3 and 4. PAS reaction was confirmative in groups 3 and 4. In addition, group 4 revealed discontinuous strong PAS+ve reaction at foci of disrupted basement membrane. In group 3, few PCNA +ve nuclei of follicular cells, some +ve nuclei in masses extending into the lumen and few +ve dislodged nuclei were found. In group 4, multiple PCNA +ve nuclei were seen in follicular cells and in follicles partially lined by more than one layer of follicular cells. In group 5, some PCNA +ve nuclei were found in follicular cells. A significant increase in the mean area of connective tissue fibers and that of PCNA +ve nuclei were recorded in groups 3 and 4. The mean area of colloid indicated a significant decrease in both. A significant decrease in the mean value of TSH was found in groups 3, 4 and 5.

In conclusion, green tea proved to have an ameliorating effect on the amiodarone induced cytotoxic and proliferative changes of thyrocytes. In addition, regression of fibrotic changes in the gland was recorded. On the other hand, overconsumption of green tea resulted in proliferative and cytotoxic effects on thyrocytes, even more marked in comparison to amiodarone cytotoxic effect.

Key Words: Amiodarone, green tea, high dose, thyroid.

List of Abbreviations

Ab: Antibody
ABC: Avidin-Biotin Complex
Ag-Ab: Antigen-antibody
AIH: Amiodarone-induced hepatotoxicity
AIT: Amiodarone-induced thyrotoxicosis
Al: Aluminium
AM: Amiodarone
ANOVA: Analysis-of-variance
ARDS: Acute respiratory distress syndrome
AV: Atrio-ventricular
DAB: Diaminobenzidine
DEA: Desethylamiodarone
DNA: Deoxy-ribonucleic acid
DPC: Diagnostic Products Corporation
EC: (-)-epicatechin
ECG: (-)-epicatechin-3-gallate
EGC: (-)-epigallocatechin
EGCG: (-)-epigallocatechin-3-gallate
F: Fluoride
Fc: Fraction Crystallizable
Fe: Iron
GA: Gallic acid
GT: Green tea
GTC: Green tea catechins
GTE: Green tea extract
GTPs: Green tea polyphenols
H&E: Hematoxylin and Eosin
HIER: Heat-Induced Epitope Retrieval
HIV: Human immunodeficiency virus
HPF: High power fields
IRMA: Immunoradiometric assay
LBPA: Lysobisphosphatidic acid
LDL: Low density lipoprotein
mM: millimolar
MNG: Multinodular goiter
OC: Overconsumption
PAS: Periodic acid Schiff's
PBS: Phosphate buffer saline
PCNA: Proliferating cell nuclear antigen

RER: Rough endoplasmic reticulum
RIA: Radio-immunoassay
ROS: Reactive oxygen species
RT: Room temperature
SD: Standard Deviation
SPSS: statistical package for social sciences
T3: Tri-iodothyronin
T4: Tetra-iodothyronin
TFCs: Thyroid follicular cells
THs: Thyroid hormones
TSH: Thyroid stimulating hormone
µg: microgram
+ve: Positive
-ve: Negative

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*Introduction
And
Aim of work*

Introduction

Amiodarone (AM) is used to manage ventricular and atrial arrhythmias (**Souney et al, 2010**). AM remains one of the most commonly prescribed antiarrhythmics effectively treating atrial and ventricular fibrillation (**Morris and Cameron, 2011**). AM is frequently associated with thyroid dysfunction. Identifying predictors for amiodarone-associated thyroid dysfunction may aid clinicians in daily practice (**Ahmed et al, 2011**).

It was hypothesized that green tea extract (GTE) supplementation would elevate antioxidant potential and attenuate exercise-induced oxidative stress and muscular damage (**Jówko et al, 2011**). Recently it was suggested that GTE and tamoxifen, individually and in combination, have potential for chemoprevention of breast cancer (**Sakata et al, 2011**). Green tea is associated with beneficial health effects mainly because of its body fat-reducing and hypocholesterolemic activities. It improves cardiovascular risk indicators such as body weight, visceral fat content and atherogenic index of plasma (**Bajerska et al, 2011**).

The potential toxicity of green tea on various organs when administered at high doses via concentrated extracts has not been completely investigated (**Hsu et al, 2011**). Green tea was proved to be associated with various beneficial health effects, but several cases of hepatotoxic side effects have been reported. The first case of toxic hepatitis

was presented following the consumption of 4-6 cups of green tea per day for six months (**Rohde et al, 2011**).

Aim of work:

The present study aimed at investigating the possible protective effect of green tea on thyroid damage. This was accomplished by the model of amiodarone induced thyroid damage in albino rat. The possible thyroid changes secondary to green tea overconsumption were also determined.

Review of Literature