Effect of the Antiarrhythmic Drug "AMIODARONE" on the Thyroid Gland of Adult Male Albino Rat and the Effect of Vitamin E Supplementation (Light and Electron Microscopic Study)

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Contents

Abstract

Amiodarone is a very effective drug , widely used for tachyarrhythmias, but it is associated with many side effects involving different organ systems . The aim of this study was to elucidate the adverse effect of amiodarone on the thyroid gland and the possible protective role of vitamin E (α-tocopherol).The present study revealed that AMD administration resulted in loss of normal follicular architecture . Many follicles appeared shrunken. Many follicles were empty or contained scanty pale colloid . Some follicles appeared lined by more than one layer of cells . Others showed interruption of their basement membrane . Thickened collagen fibers in between the thyroid follicles were observed. Strong +ve PAS reaction in the basal lamina and cells lining the follicles and negative (–ve) to weak +ve reaction in the colloid filling the follicles were noticed . Sections of the thyroid gland of this group showed positive immune reaction to caspase-3 in the cytoplasm and nuclei of many follicular cells . The apical border of the follicular cells showed few irregular detached microvilli . The nuclei of the follicular cells were almost irregular and showed chromatin condensation . The cytoplasm of most follicular cells revealed numerous dilated rough endoplasmic reticulum . Numerous lysosomes were frequently seen . Some of these changes were irreversible during the recovery period. Supplementation of vitamin E (α tochopherol) with amiodarone improved these changes .

Key words:

Amiodarone -Thyroid Glandof rat

Dysrhythmias or arrhythmias are abnormalities of electrical conduction or rhythm in the heart. They encompass a number of different disorders that range from being harmless to life threatening disorders **(Holland and Adams, 2003).**

Arrhythmias are induced by abnormal pacemaker activity or abnormal impulse propagation.Thus, the aim of therapy of the arrhythmias is to reduce ectopic pacemaker activity and modify conduction or refractoriness in reentry circuits to disable circus movement. The major mechanisms currently available for accomplishing these goals are sodium channel blockade, blockade of sympathetic autonomic effects in the heart, prolongation of the effective refractory period and calcium channel blockade **(Katzung, 2004).**

Amiodarone (AMD) is a very effective drug, widely used for tachyarrhythmias and, to a lesser extent, ischemic heart disease **(Bartalenaet al.,2004).**

Amiodarone shows noncompetitive alpha-blocking activity and inhibits the show inward calcium-ion current. Lastly, it is possible that some antiarrhythmic actions of amiodarone result from its antithyroid effect ; the drug interfere with thyroid metabolism and with the effects of thyronix on the heart**(Kopp, 2005).**

Schenck et al., (2006)emphasized that, despite amiodarone is a highly effective antiarrhythmic agent; however it is associated with many side effects involving many different organs.Amiodarone-induced tissue toxicity could be a life-threatening complication which limits its widespread use as an anti-arrhythmic agent.

Most side effects of amiodarone are not dose-related and occur only after weeks or months of therapy, as a result of its

pharmacologicproperties .Moreoverthe incidence of side effects increases over time and , therefore, many side effects may be related to the total amount of the drug that has been accumulated .Amiodarone exerts effects on many organs that may dought its therapeutic effects on the heart **.**Many adverse effects have been reported due to the use of amiodarone that include injury to the liver, thyroid, cornea, skin, and neuromuscular system **(Iskandar et al., 2006).**

The complex effects of amiodarone on the thyroid range from abnormalities of thyroid function tests to marked thyroid dysfunction, Therefore, thyroid toxicity is perhaps the significant and potentially life-threatening side effect resulting from amiodarone use thyroid dysfunction, whether hypothyrodisim or thyrotoxicosis, is common in patients on long-term AMD therapy . Hypothyrodisim may be transient or persistent with the latter almost always associated with underlying thyroid disease.On the other hand thyrotoxicosis is normally found in patients without pre-existing thyroid disease especially in areas of sufficient iodine intake and is thought to be due to a non-autoimmune form of thyroditis**(Bogazzi. 2007).**

The term vitamin (E) describes a family of eight antioxidants, four tocopherols, alpha-, beta-, gamma- and delta-, and four tocotrienols (also alpha-, beta-, gamma- and delta-). Alpha-tocopherol is the only form of vitamin E that is actively maintained in the human body and is therefore, it is the form which found in the largest quantities in the blood and tissues **(Hong et al, 2009).**

Free radicals are formed primarily in the body during normal metabolism and also upon exposure to environmental factors such as cigarette smoke or pollutants.Fats, which are an integral part of all cell membranes, are vulnerable to destruction through oxidation by free radicals. The main function of alpha-tocopherol in humans appears to be

that of its antioxidant role.The fat-soluble vitamin, alpha-tocopherol, is uniquely suited to intercepting free radicals and preventing a chain reaction of lipid destruction. Vitamin (E) acts solely as an antioxidant agent preventing the peroxidation of polyunsaturated fatty acids, and thus preserving the integrity of cellular membranes **(Yiu*et al*,2009).**

Aside from maintaining the integrity of cell membranes throughout the body, alpha-tocopherol also protects the fats in low density lipoproteins (LDLs) from oxidation. Lipoproteins are particles composed of lipids and proteins, which are able to transport fats through the blood stream. LDL transport cholesterol from the liver to the tissues of the body. Oxidation of LDLs has been implicated in the development of cardiovascular diseases. When a molecule of alpha-tocopherol neutralizes a free radical, it is altered in such a way that its antioxidant capacity is lost. However, other antioxidants, such as vitamin C, are capable of regenerating the antioxidant capacity of alpha-tocopherol(**Esmekaya et al, 2010).**

The present study aimed to investigate:-

- Effectsof amiodarone on adult rat thyroid gland from the histological,immunohistological and electron microscopicpoint of view.
- The possible of protective role of vitamin E against amiodaroneadverseeffects.
- The possibility of recovery of thyroid after amiodaronewithdrawal.

Anatomical aspect of the thyroid gland in rat

The thyroid lobes lie ventrolateral to the upper four or five tracheal rings. Ventrally the gland is covered by the slender sternothyroid muscle and laterally by the longuscollimuscle. The pinkish triangular or rhomboid lobes are 3.9-5.5 mm long and 2-3 mmwide. Average weight of the entire gland is 15.4mg in the male and 17.9 mg in the female**.**It is 7.5 mg / 100g body weight in adult rats. The carotid arteries lie next to the lobes of the thyroid and a small nerve (recurrent laryngeal) passes between the lobes and the larynx. In about half of the animals, an isthmus can be found macroscopically. The thyroid capsule consists largely of collagenous fibers. It contains adipose cells and sinusoidal vessels with wide lumina. These are continuous with the vascular system of the stroma**(Sidhu and Jenkins, 2003)**.

The rat has two parathyroids, one laterally situated at the anterior ploe of each thyroid lobe. They are circular and paler than the thyroid**(Singh 2008).**

Histological aspect of the thyroid gland in rat

The gland proper is composed of numerous follicles. Each follicle is a ball of colloid surrounded by a single layer of follicular cells**.** The spherical, ovoid or polygonal follicles are often incompletely separated from each other by connective tissue septa of varying width. With increasing size the follicles approach a spherical shape. The largest follicles (up to 270 µ diameter) are found peripherally and the smallest (up to 120µ) are found centrally. The isthmus of the thyoid contains blood vessels with a wide lumen and isolated follicles. **(Kurnik *et al.*, 2004).**

Shape of the follicular epithelium depends on its functional state. Resting cells are low cuboidal (up to 4µ). Active cells may be columnar (about 10µ). The colloid of resting follicles is more acidophilic than that of active ones. Parafollicular cells (C cells) may be found in the stroma between epithelial cells (**Woeber, 2005).**

Amiodarone

Amiodarone is a member of antiarrhythmic drugs with predominantly class III (Vaughan Williams' classification) effects (Table 1). Amiodarone is a benzofuranderivative: 2-buty1-3- benzofurany1 4-[2-(diethylamino)-ethoxy]-3.5-diiodopheny1 ketone hydrochloride. Its molecular formula is: C25H29I2NO3•HCl. its structural formula is as follows**:**

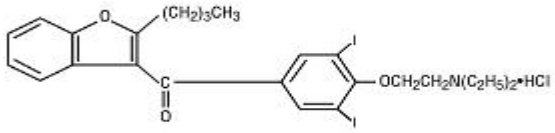

Diagram (2): formula of amiodarone

Table (1): Vaughan Williams' classification of antiarrbythmic drugs**(Bartalena et al., 2002).**

Class	Mechanism	Examples
Ia	(Na^+) channel block (intermediate association/dissociation)	Disopyramide Procainamide Quinidine
Ib	(Na^+) channel block (fast association/dissociation)	Lidocaine Phenytoin
Ic	(Na^+) channel block (slow association/dissociation)	Flecainide Propafenone
II	beta blocking	Propranolol Timolol
III	K^+ channel blocker	Amiodarone Sotalol bretylium
IV	Ca^{2+} channel blocker	Verapamil Diltiazem

Orally, it is available under the trade names Pacerone (produced by Upsher-Smith Laboratories, Inc.) and Cordarone (produced by Wyeth-Ayerst Laboratories) in 200 mg and 400 mg tablets An oral loading dose is typically a total of 10 grams, divided over one to two weeks but there are many other dosing regimens. Once an individual is loaded, a typical maintenance dose of amiodarone is 100 or 200 mg either once or twice daily**(Eaton et al. 2002).**

It is also available as intravenous ampoules and vials, typically in 150 mg increments. An intravenous loading dose is typically 300 mg in 20-30cc for cardiac arrest. The loading infusion for dysrhythmias is typically 150 mg in a 100cc bag of given over 10 minutes. Both can be followed by a 360 mg slow infusion over 6 hours then a maintenance infusion of 540 mg over 18 hours **(Surks et al.,2004).**

Its most important direct electrophysiologic effect is prolongation of the action potential duration and repolarization time; this prolongation results from inhibition of the potassium ion fluxes that normally occur during phases 2 and 3 of the action potential and from prolongation of the refractory periods. These actions represent the class III activity of amiodarone and occur in all cardiac tissue. The increase in the duration of repolarization and refractoriness reduces membrane excitability; this reduction represents the antifibrillatory property of amiodarone**(Kathofer et al., 2005).**

Amiodarone is also a weak sodium channel blocker and as a result of its class I antiarrhythmic activity, slows the upstroke velocity of phase 0 of the action potential**.** This reduces the rate of membrane depolarization and impulse conduction.Amiodarone also directly depresses the automaticity of the sinus and atrioventricularnodes**.**In addition to these direct antiarrhythmic effects,amiodarone has several

important actions, including beta-blokade, that indirectly affect the electrophysiologic properties of the myocardium. Unlike blockade using the standard beta-blocking agent.Amiodaron& blockage of the beta-adrenergic receptors is noncompetitive and results from inhibition of adenylatecyclase formation and from a reduction in the number of beta-adrenergicreceptors**.**Amiodarone also shows noncompetitive alphablocking activity and inhibits the slow inward calcium-ion current. Lastly, it is possible that some antiarrhythmic actions of Amiodarone result from its antithyroideffect; the drug interferes with thyroid metabolism and with the effects of thyroxin on the heart **(Standringet al. 2005).**

The gastrointestinal absorption of Amiodarone is slow and Incomplete; a peak serum level is achieved 4 to 7 hours after an oral dose is received**.** Only 50% of the administered dose is bioavailable because of first-pass intestinal mucosal and hepatic metabolism as well as incomplete absorption.Amiodarone is highly protein and lipidbound and is widely distributed throughout all adipose tissue**(Uzan et al. 2006).**

As a result of the drug's affinity for adipose tissue, the estimated volume of distribution is 50 L and approximately 15 g are necessary to saturate these large body stores. For these reasons, a long and variable loading period is necessary before antiarrhythmic activity is apparent **(Ghosseinet al., 2006)**

Amiodarone is metabolized principally in the liver into desethylamiodarone (DEA), the major active metabolite of Amiodarone. DEA serum concentrations greater than 0.05 mg/L are not usually seen until after several days of continuous infusion but with prolonged therapy

reach approximately the same concentration as amiodarone**(Killion et al., 2006).**

Clearance of Amiodarone involves deiodination, but the major route of elimination is by hepatic metabolism to one principal metabolite, desethyl-amiodarone, which has antiarrhythmic activity equivalent to that of the parent drug**.** There is 10 – 50% transfer of amiodarone and DEA in the placenta as well as presence in breast milk**.** It has been reported that a serum concentration of at least 1 to 2 micrograms /ml is necessary for drug efficacy, but there is wide interpatient variability in the dose-concentration relation and in levels associated with efficacy or toxicity. Therefore,amiodarone levels in the blood have limited clinical usefulness. The elimination half-life of amiodarone is also highly variable but long ranging from 16 to 180 days (mean, 52days), because of drug's extensive storage and avid binding to poorly perfused adipose tissue **(Kondo, 2006).**

Because of the unique pharmacokinetic properties of amiodarone, the onset of the drug's action is delayed and may not be apparent for as long as 3 months. When administered orally, a loading dose of the drug is required. The dose used depends on the arrhythmia being treated, the need to achieve efficacy more quickly, and the occurrence of side effects, some of which are associated with higher doses **(Klein andDanzi, 2007).**

There is no well-established relationship of plasma concentration to effectiveness, but it does appear that concentrations much below 1 mg/L are often ineffective and that levels above 2.5 mg/L are generally not needed. Within individuals dose reductions and ensuing decreased plasma concentrations can result in loss of arrhythmia control. Plasma-concentration measurements can be used to identify patients whose levels are unusually low, and who might benefit from a dose increase, or

unusually high, and who might have dosage reduction in the hope of minimizing side effects**(Singh et al.,2007).**

Amiodarone is an iodine-rich drug widely used for the management of ventricular arrhythmias, paroxysmal supaventricular tachycardia and atrial fibrillation and flutter**.** The drug is, to a lesser extent, also employed for severe congestive heart failure because of its minimal negative inotropic action**.** In addition, it may decrease cardiac-related mortality after myocardial infarction, although the latter effect has been questioned**,** decrease the immediate and late complications of atrial fibrillation in patients undergoing elective cardiac surgery**,** prevent recurrence of atrial fibrillation and decrease cardiac arrhythmia death when given to patients upon arrival of emergency medical technicians (EMTs) **(Tschuppert et al.,2007).**

Amiodaronefrequently interacts with other drugs. One of the most common interactions is with warfarin; amiodarone can potentiate the effect of this drug by interfering with hepatic metabolism. The prothrombin time is elevated, necessitating a reduction in the warfarin dose. This interaction may be erratic; hence, frequent measurement of the prothrombin time, especially during the first few weeks of loading, is essential. Because amiodarone can elevate the serum digoxin level, reduction of the digoxin dose may be necessary. Amiodarone may also increase the serum level of other antiarrhythmic drugs, including quinidine, procainamide, mexiletine, and propafenone. When combined with class IA drugs,amiodarone may cause an exaggerated QT prolongation. Potentiation of the effects of anesthetic agents, including hypotension and bradycardia, has been reported. The combined use of amiodarone and beta-blockers or calcium channel blockers may result in marked depression of sinus or atrioventricular nodal function and