Effect of Chronic Exposure of Cadmium Chloride in Drinking Water on Structural and Functional Aspects of Thyroid Gland in Mature Male Rabbits[#]

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Abstract

The effect of chronic exposure to two different levels of cadmium chloride (CdCl2) 30 ppb and 40 ppb in drinking water for 12 weeks on thyroid function of mature male rabbits was studied. Eighteen mature male rabbits were randomly divided into three groups (each of six) , control group (group I): were offered ordinary tap water , and treated groups (II and III) were offered tap water containing 30ppb and 40 ppb respectively for 12 weeks .Serum concentration of thyroxin (T4) and triiodothyronine (T3) were measured every six weeks ,as an index of thyroid function , further more , section of thyroid gland were prepared for histological studies. The results showed that chronic exposure of male rabbits to two different levels of CdCl2 caused significant decrease ($p<0.05$) in serum T3 and T4 in both treated groups in compared with control group, in addition, there were histological changes in thyroid gland of treated groups manifested by hyperplasia with presence of large number of varying size of microfollicels and hypertrophic thyrocytes, small colloid with little secretion .

Key wards: Cadmium, Thyroid gland, Thyroxin, Triiodothyronin, Thyrocyt

الخلاصة

اجريت هذه الدراسة لمعرفة تأثير التعرض المزمن لمستويين مختلفين من كلوريد الكادميوم (٣٠ ، ٤٠ جزء بالبليون) في ماء الشرب ولمدة ١٢ اسبوع على وظيفة الغدة الدرقية في ذكور الارانب البالغة ٠ تم استخدام ١٨ ارنبا بالغا قسمت عشوائيا الى ثلاث مجاميع متساوية : مجموعة السيطرة (group I) استلمت ماء عادي طيلة فترة التجربة ومجموعتي المعالجة (group II) استلمت ماء الشرب المضاف اليه كلوريد الكادميوم بتركيز (٣٠، جزء بالباليون) و (group III) استلمت ماء الشرب مضافا اليه كلوريد الكادميوم بتركيز ٤٠ جزء بالبليون ولمدة ١٢ اسبوعا تم قياس هرموني الغدة الدرقية الثايروكسين T4 وتراي ايودو ثايرونين T3 كل ست اسابيع فضلا عن اخذ المقاطع النسيجية للغدة الدرقية. اظهرت النتائج ان تعرض ذكور الارانب المزمن لمستويين مختلفين من كلوريد الكادميوم في ماء الشرب قد سبب انخفاضا معنويا في تركيز هرموني T3 و T4 في مصل الدم في مجاميع المعالجة مقارنة مع مجموعة السيطرة كما اظهرت الفحوصات النسيجية للغدة الدرقية حدوث حالة فرط النسيج (hyperplasi) تمثلت بوجود عدد كبير من الخلايا الدرقية وباحجام مختلفة محاطة بطبقة من الخلايا الطلائيـــة (hypertrophic thyrocyte) فضلا عن وجود نقص واضح في كمية الغروان (colloid) داخل الجريب مع قلة في افرازاته .

Introduction

Cadmium is modern toxic metals (discovered in 1817) . Its main use in electroplating because of its anticorrosive properties, it also used as color pigment of paints, plastics and as cathode material for nickel –cadmium batteries, cadmium is an important source of environment pollution [(1)]. Hazardous waste disposal sites are large source of Cd concentration found in soil and water . Tobacco smoke is the main reason for cadmium accumulation in our body [(2)],on other hand , the major route of cadmium intake (for non smoker) is ingestion , this is largely due to the presence of Cd (2-40 ppb) in food staff of natural origin e.g. cereals beans , carrots , beverage, coffee and tea [(3)] ,or by the ingestion of contaminated food especially fish [(4, 5)] . The acute toxic effect of Cd are generally restricted to the lung ,where as the effects following chronic Cd exposure in human are multisystemic and include nephropathies emphysematous alteration in the lung , cardiovascular disease , and bone damage possibly (osteomalacia and osteoperosis) [(6)] . Wealth of evidence suggested that heavy metals including cadmium exert profound toxic effects on the activities of a number of endocrine gland including thyroid gland [(7,8)] .

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Cadmium intoxication has been reported to reduce the thyroidal iodide uptake [7] and inhibits hepatic conversion of thyroxin (T4) to triiodothyronine (T3) in rats [9] and mice [10]. It has been previously reported that subcutaneous injection of cadmium chloride (CdCl2) (mg/kg B.W) for ten weeks to male rabbits resulted in a significant reduction in serum T3 concentration and hepatic T4 production [11] . The direct relationship between heavy metals poisoning and thyroid dysfunction were studied in male rabbits by Ghosh and Battacharya ,1992 , within 24 hours of intramascular injection of CdCl2 (15mg/kg B.W) a significant increase in thyroid activity over the control with a concomitant rise in T3 titer were observed .. The toxic effect of cadmium on thyroid gland functions has been studied , yet its effect at above the permissible level and below the toxic one are questioned , to this aim the present study dedicated .

Materials and Methods

Number of mature male rabbits 800-1000 g of local breed were acclimated to holding facilities for one week prior to commencement of dosing . Animals in all stages of experiment were housed in clear plastic cages in conditioned room (22-25 c◦) with controlled lightening ,eighteen rabbits were randomly and equally divided into three groups and were treated for three months as fallow : group I, rabbits in this group were received tap water and served as a control group , the group II were received 30 ppb cadmium chloride in drinking water , while the animals of group III were received 40 ppb of cadmium chloride in drinking water . Fasting blood samples were collected at zero, 6, and 12th weeks of experiment via cardiac puncture technique , then serum was separated and frozen at – 20 c◦ for hormonal analysis ,tetraiodothyroninT4 and triiodothyronin T3 were determined by using immuno assay detection [12]. For histological studies rabbits were killed and a portion of trachea with intact thyroid gland was dissected free of connective tissue and preserved in 10% formaline buffer solution till the preparation of histological sections . Tissues were embedded in paraffin and several tissues sections were prepared, and stained with Hematoxylin- Eosin stain [13].Statistical analysis of data was done on the basis of two – way analysis of variance (ANOVA) , using a significant level of $p < 0.05$ [14] .

Results

The effect of the two different levels of cadmium chloride on mean values of serum T3 concentration of male rabbits are shown in table (1) , serum T3 concentration showed a significant decrease ($p< 0.05$) at the 6th week of exposure to cadmium chloride in group III and at the 12th week of experiment in group II as compared with control . In cadmium treated groups , In table(2) thyroxin serum concentration significantly decreased ($p<0.05$) compared to control , such decrement was observed at the 6th –12th week of experiment. Within groups , the values tended to decrease significantly ($p<0.05$) with time (at 6th-12th week) as compared with the pretreated period . The histological structure of thyroid gland of un treated rabbits was shown in figure (1) , the thyroid follicles containing colloid of uniform color , oval in shape , lined by cuboidal thyroidal follicular cells , figures 2,3 illustrated the histological changes in thyroid gland of rabbits following 30 and 40 ppb of CdCl2 (group II and group III),the figures showed a case of hyperplasia manifested by presence of thyroid microfollicels of varying size lined by high columnar thyroidal follicular cells (hypertrophic thyrocyte) , small colloid with little secretion in follicular lumen (figure 2) , while the figure 3 showed a large area of irregular and varying size microfollicels with hypertrophic thyrocyte with little colloid secretion .

Table 1 : Serum triiodothyronine (T3) concentration (nmol/l) in rabbits treated with two different levels of cadmium chloride in drinking water.

Groups / Time (weeks)		I	II	III
Pre-treatment	0	0.43± 0.01 A a	0.44±0.56 A a	0.44±0.01 A a
During treatment	6	0.45±0.01 A a	0.43±0.01 A a	0.41±0.02 B b
	12	0.44±0.01 A a	0.40±0.02 B b	0.40±0.02 B b

Values are expressed as mean ±SE n = 6/group
Groups: I = control,
II = rabbits received 30 ppb of cadmium chloride in drinking water,
III = rabbits received 40 ppb of cadmium chloride in drinking water.
Capital letters denote between group differences, P< 0.05 vs. control.
Small letters denote within same group differences, P<0.05 vs. pretreated values.

Table 2: Serum tetraiodothyronine (T4) concentration (nmol/l) in rabbits treated with two different levels of cadmium chloride in drinking water.

Groups / Time (weeks)		I	II	III
Pre-treatment	0	58.17±0.7 A a	57.42±0.56 A a	57.12±0.42 A a
During treatment	6	58.5±1.25 A a	51.0±2.4 B b	49.2±1.6 B b
	12	58.35±1.3 A a	44.5±1.9 B c	41.7±0.4 B c

Values are expressed as mean ± SE n = 6/ group
Groups: I = control,
II = rabbits received 30 ppb of of cadmium chloride in drinking water,
III = rabbits received 40 ppb of cadmium chloride in drinking water.
Capital letters denote between group differences, P< 0.05vs. control.
Small letters denote within group differences, P< 0.05 vs. pretreated values.

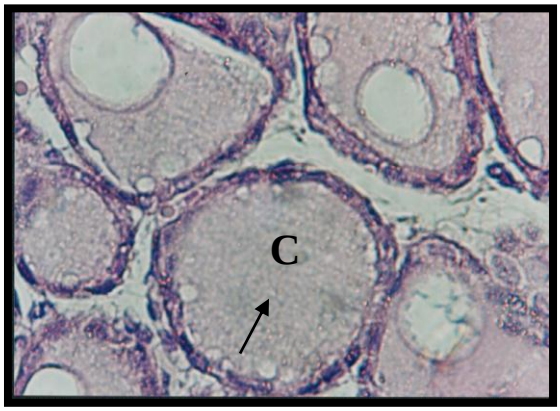

Figure 1:.Section in thyroid gland of untreated rabbit (group I), note thyroid follicles with thin epithelial lining cells (thyrocyte) (arrow), filled within colloid secretion (c). H&E, 40x

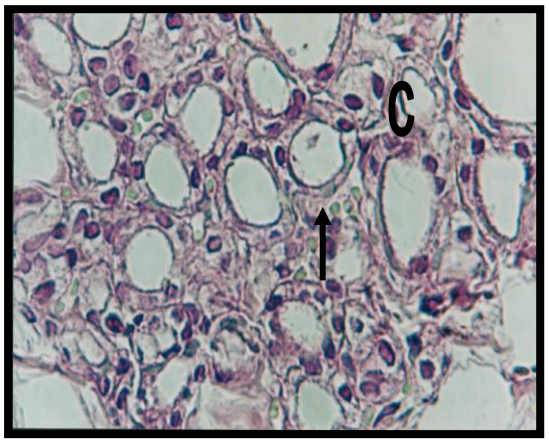

Figure 2: Section in thyroid gland from cadmium treated rabbit (group II), note follicular- cells of varying size, high hypertrophic thyrocytes (arrow), with a little area of micro colloid secretion.(C) H &E. 40x

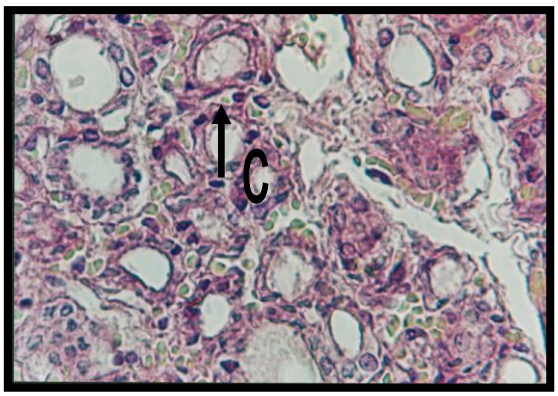

Figure 3 :Section in thyroid gland from cadmium treated rabbit (group III), showing large area of microfollicels of varying size with hypertrophic thyrocyte (arrow), and hardly little secretion in the lumen. (C) H&E. 40x

Discussion

This study showed that exposure of rabbits to cadmium bring the animal to the case of hypothyroidism manifested by significant decrease in serum T3 and T4 concentration associated with biological changes . To the best of our knowledge , the effects of chronic exposure to CdCl2 at level 30 and 40 times above the permissive level on thyroid gland feature , has not been studied yet , however considerable body of evidence how exist attesting the involvement at heavy metals including cadmium in the toxicity of a number of endocrine glands including thyroid gland [8,9] . Many studies suggested the mechanisms including affecting hepatic thyroxin metabolism through reduction in the activity of hepatic " Outer Ring Deiodinase ORD" an

enzyme which responsible for conversion of major circulating form of thyroid hormone (T4) to more biologically active form(T3) with disruption of T3 signaling leading to reduction in T4 conversion [15]. In addition cadmium may cause reduction of thyroidal I uptake by damages the structure and function of both follicular and parafollicular cells of thyroid [9,10]. Long term exposure to cadmium may induce the activity of hepatic microsomal enzyme especially UDP-GT (Uridine DiPhosphate Glucournyl Tranferase) [16] and phenol sulftransferase [17] resulting in rapid clearance of T3 and T4 , on other hand , accumulation of cadmium in mitochondria of thyroid follicular epithelial cells might disturb the oxidative phosphorylation of this organelle with subsequent loss of energy supply leading to inhibition in the synthesis and release of thyroid hormone [18] . The suspected selenium deficiency caused by cadmium treatment may lead to histological changes by activation fibrotic process in which inflammatory reaction and excess transforming growth factor B play a role in thyroid gland morphology . [19] Gupta P and Kar A (1999) study the relation between cadmium and selenium , vitamin E and thyroid dysfunction in chicken, in this study they suggested that cadmium decrease T3, probably by inhibiting hepatic 5-monodeiodinase (5D-I)activity, which is a selenium dependent function. Cadmium is known selenium antagonist while vitamin E facilitate selenium metabolism .Vitamin E was shown to protect against cadmium toxicity and maintain 5D-I activity and T3 levels, while the experimenters concluded that the metal – induced inhibition in hepatic 5 D-I activity is mediated through lipid piroxidation my conclusion is that the cadmium inhibited 5D-I activity by decreasing selenium.While vitamin E does decrease lipid peroxidation it does this by facilitating selenium metabolism and selenium is the key metal in glutathione peroxidase which is a potent inhibitor of lipid peroxidation [9] Accordingly we can speculate that selenium deficiency may occur in this study following cadmium exposure resulting in the damage of thyroid gland , decrease the concentration of both (T3 and T4) and inhibit monodeiodinase activity leading to state of hypothyroidism .

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