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Original Article

High dietary consumption of iodine induced thyroid cytotoxicity in diabetic intoxicated rats and oxidonitrergic stress in non-diabetic rats

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Abstract

This study aimed to investigate the role of iodine intake in thyroid function ofdiabetic rats. Twenty-four (24) male Wistar rats were placed into four groups (n=6): Group (non-diabetic without iodine), Group 2 (non-diabetic + iodine), Group 3 (diabetic without iodine) and Group 4 (diabetic + iodine). 10mg/kg bw of iodine were mixed with the feeds. Serum triodothyronine (T3), thyroxine (T4), Thyroid Stimulating Hormone (TSH), thyroglobulin and thyroperoxidase antibodies were assessed using ELISA. Serum MDA, SOD, and NO levels were assessed with spectrophotometry. In the diabetic rats, lower mean serum T4 and TSH concentrations were observed (T4: 13.16±0.55 Vs 11.75±0.21 mg/dL, TSH: 2.62±0.11 Vs 2.28±0.08 IU/mL). Iodine treatment further reduced T4 and increased TSH concentrations (T4: 11.75±0.21 vs 6.75±0.22 mg/dL, TSH: 2.28±0.08 Vs 3.08±0.15 IU/mL). Thyroglobulin and thyroperoxidase antibodies were absent in all the rats. It was also observed that iodine intake caused an increase in oxidative stress in both diabetic and non-diabetic treated rats (MDA; 18.4±1.3 Vs 22.2±2.7 µmol/l X 10-5, NO; 14.08±0.38 Vs 13.24±0.07µm/l) and increased SOD levels in diabetic rats (44.44±2.94 Vs 68.94±0.91 mg/ml); this increase could be due to the increased TSH. Consumption of excess iodine suppressed thyroid function in diabetic rats and induced oxidative stress in both diabetic and non-diabetic treated rats.

Keywords: iodine supplementation, diabetes, oxidative stress, thyroid function

1. Introduction

The two endocrinopathies that affect people most frequently are diabetes and thyroid disease. As both insulin and thyroid hormones are essential in cellular metabolism, an excess or lack of one might cause difficulties with the other's functionality (Mohamed *et al*., 2017). Thus, it is possible for diabetes and thyroid problems to coexist in individuals. Patients with type 1 diabetes experience hyperthyroid symptoms, while those with type 2 diabetes typically experience hypothyroid symptoms (Mohamed *et al*., 2017). Thyroid disorder seems to induce oxidative stress in the testis by reducing the levels of testicular enzymatic and non-

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enzymatic defenses (Mohamed *et al*., 2017).

Diabetes mellitus is a disease of metabolic dysregulation (Chijiokwu *et al*., 2022) accompanied by long-term vascular and neurological complications (Rhoades & Tanner, 2003). The prevalence of diabetes for all ages worldwide was estimated at 2.8% in 2000 and 4.4% by 2030 (Wild, Roglic, Green, Sicree, King, 2004). It has long been recognized that thyroid disorder is related with an increased prevalence of poor glucose metabolism (Kabadi & Eisenstein, 1980). Following that, numerous *in vivo* and *in vitro* tests were carried out in attempts to identify the fundamental pathophysiologic abnormalities underlying the relationship of hyperglycemia with thyrotoxicosis (Wajchenberg *et al*., 1978). In numerous reports on hyperthyroidism, fasting blood sugar levels were found to be either normal or excessive. More so, an expanded body of evidence has also shown a relationship between thyroid function and blood glucose levels (Dandan *et al*., 2021; Ogbonna *et al*., 2019; Wenhua *et al*., 2019). Notably, Arigi, Fabiyi and Fasanmade (2014) observed that both hypothyroidism and hyperthyroidism caused dysfunction in glucose tolerance and led to increased fasting blood glucose in non-diabetic rats. Macini *et al*. (2019) discovered that there was a higher risk of developing type 2 diabetes in individuals consuming high levels of iodine in their diet. Ravindra *et al*., (2011) observed that diabetic serum had a significantly lower ability to bind and transport iodine.

Thyroid hormones **[**tri-iodo-thyronine (T3) and thyroxine (T4)**]** are known to regulate a variety of biochemical processes throughout the body, such that are required for appropriate growth, metabolism, and brain activity. Iodine is an essential component of T3 and T4, and it has unique effects on the thyroid gland and the immune system. Iodine is a non-metallic element belonging to the halogen family in Group VIIA of the periodic table (Cooper, 2007). It is a dark purple, crystalline and lustrous solid at room temperature (Cooper, 2007). Iodine is necessary to living organisms, in which it is actively concentrated in the thyroid gland for the synthesis of thyroid hormones; triiodothyronine (T_3) and thyroxine (T_4) (Haldimann, Bochud, Burnier, Paccaud, & Dudler, 2015). The recommended dietary intake (RDI) of iodine for adults is 150µg/day. It could be obtained by consuming foods such as seaweed, milk and dairy products, iodized table salt, or seafood (DOH-UK, 1995). According to Wolf and Chaikoff (1948), consumption of high amounts of iodine inhibits three steps in the synthesis of thyroid hormones; iodide trapping, thyroglobulin iodination (wolf-Chaikoff effect) and thyroid hormone release from the thyroid gland, which can lead to hypothyroidism. Excessive iodine consumption can also induce hyperthyroidism and this is known as the Jod-Basedow effect. Excess thyroid hormones cause “thyroid diabetes” (Hartoft-Nielsen *et al*., 2009), whereas hyperthyroidism causes glucose intolerance in animals and humans (Hartoft-Nielsen *et al*., 2009). Diabetes mellitus has been proven to coexist with a range of thyroid disorders. The thyroid gland regulates carbohydrate metabolism at the levels of pancreatic islets and glucose-using target tissues, raising crucial therapeutic and diagnostic challenges. There is however limited information regarding the effects of high iodine intake on the thyroid gland of diabetic rats. In the above context, this study was carried out to investigate the effects of high iodine intake on thyroid function in diabetic rats and its underlying mechanisms.

2. Materials and Methods

2.1 Animal handling

Adult male Wistar rats weighing 150 to 200 g (7-9 weeks old) used for this study were obtained from the Central Animal house, Faculty of Basic Medical Sciences, University of Ibadan, Nigeria, and were kept in plastic cages under normal standard conditions of about 25 ± 2 °C in 12:12 h day and night cycle. The animals were left to acclimatize for at least 14 days with unrestricted access to water and standard rat chow before commencing the experiments. The study protocols used in handling the animals were in line with those established by the National institutes of Health (NIH) Guideline for the Care and Use of Laboratory Animals (Publication No. 85-23, revised). Six animals per group were used in this study, based on the principle of the three Rs (3Rs: Replacement, Reduction and Refinement) by Oyovwi *et al*. (2021).

2.2 Ethical approval

The University of Ibadan's Ethics Committee for Animal Care and Use (ACUREC), reference number UI-ACUREC/19/0029, approved the use of animals in this study. The Animal Care and Use Ethics Committee (ACUREC) guarantees that all adverse events are promptly reported to ACUREC and those institutional policies and laws are followed.

2.3 Determination of iodine and caloric content of feeds

The standard rat feed was analyzed as described in the following paragraphs.

2.3.1 Determination of caloric content (using a bomb calorimeter)

The apparatus used was the Gallenkamp Ballistic Bomb Calorimeter. Reagent used for calibration was Benzoic acid. Determination: 0.25g of each sample depending on the bulkiness was weighed into the steel capsule. A 10 cm cotton thread was attached to the thermocouple to touch the capsule. The bomb was closed and charged in with oxygen up to 30 atm. The bomb was operated by depressing the ignition switch to burn the sample in an excess of oxygen. The maximum temperature rise in the bomb was measured by the thermocouple and a galvanometer system.

2.3.2 Determination of iodine content

The method of A.O.A.C (1984) was used to determine the content of iodine. A 5g sample was dissolved in approximately 100 ml water. The pH was adjusted to 2.8 using 0.6% HCl. 30 mg potassium iodide powder (KI) was added to convert all iodate present to elemental iodine. The liberated iodine was titrated with 0.005 N freshly prepared $Na_2S_2O_3$ (sodium thiosulphate solution) using 1% starch solution as indicator of the end point or equivalence point. The titer obtained at this point was used to calculate iodine concentration in the sample, in mg/kg.

2.4 Induction of diabetes mellitus

Diabetes was induced in subgroups 3 and 4 after 6 weeks with one dose by intraperitoneal administration of Alloxan monohydrate, at the dose level of 150 mg per kg rat body weight (Akinola, Gabriel, Suleiman & Olorunsogbon, 2012; Maiffo *et al*., 2019; Sikarwar & Patil 2010) after an overnight fast. The Alloxan was diluted in normal saline and administered within a few minutes. One hour after the dose, the rats were given feed *ad libitum* and 5% dextrose. After 7 days, the fasting blood glucose of the rats was assessed with the use of Accu-check glucometer and strip. The rats with fasting blood glucose levels higher than 200 mg/dl were considered diabetic.

2.5 Distribution and treatment of animals

Twenty four (24) rats were randomly divided into four groups of six rats each. These rats received the various gavage treatments for 14 days as follows: Group 1 (normal control) received 10 mL/kg bw of distilled water; Group 2 (diabetic rats) received 10 mL/kg bw of distilled water; Group 3 (non-diabetic rats) received 2.5 mg/kg bw of the iodine; Groups 4 (diabetic rats) received the iodine at the dose of 10 mg/kg bw. The doses and routes of distilled water (Miaffo *et al*., 2019) and iodine (Kotyzováa, Eybla, Mihaljevičb, & Glattre, 2005) were selected based on previous dose-response effect and a preliminary investigation. However, normal saline (10 mL/kg, p.o.) was administered as vehicle to naïve rats in different groups that served as normal control. Notably, in groups 2 and 4, the rats were fed with feeds mixed with iodine at a concentration of 10 mg/kg, *ad libitum* throughout the experiment. The requirement was determined by comparing the weight, iodine content and dry matter content of thyroid glands from rats supplemented with various levels of iodine. All treatments were done orally between 8.00 am and 9.00 am once daily, for the period of eight (8) weeks.

2.6 Collection of blood samples and preparation

Blood was collected into plain bottles from the orbital vein with the use of plain capillary tubes. The blood was centrifuged at 3,000 revolutions per minute (rpm) for 20 minutes after which the supernatant was separated with the use of a micropipette into separate plain bottles and frozen at -20°C until the thyroid hormone assays were performed using ELISA strip reader.

2.7 Circulatory concentration of thyroid hormones analysis

The levels of free T3 (fT3), free T4 (fT4), thyroid stimulating hormone (TSH), Thyroid peroxidase (TPO) and thyroglobulin (TG) antibodies were determined in serum samples using their respective ELISA kits (diagnostic systems laboratories INC.) supplied from Monobind Inc., USA, according to the manufacturer's recommended protocol.

2.8. Determination of oxidative biomarkers

2.8.1 Determination of lipid peroxidation (Malondialdehyde-MDA)

Lipid peroxidation was determined by measuring the thiobarbituric acid reactive substances (TBARS) produced by lipid peroxidation. This was carried out by the method of Varshney and Kale (1990). An aliquot of 0.4 ml of the sample was mixed with 1.6 ml of Tris-KCl buffer, to which 0.5 ml of 30 % TCA was added. Then 0.5 ml of 0.75 % TBA was added and placed in a water bath for 45 minutes at 80 ºC. This was then cooled in ice and centrifuged at 3000 rpm for 15 minutes. The clear supernatant was collected and absorbance was measured against a reference blank of distilled water at 532 nm. The MDA level was calculated according to the method of Adam-Vizi and Seregi (1982). Lipid peroxidation in units/mg protein or gram of tissue was computed with a molar extinction coefficient of $1.56 * 10^5 M^{-1} Cm^{-1}$.

2.8.2 Determination of superoxide dismutase (SOD) activity

The level of SOD activity was determined by the method of Misra and Fridovich (1972). 1 ml of the sample was diluted in 9 ml of distilled water to make a 1 in 10 dilution. An aliquot of 0.2 ml of the diluted sample was added to 2.5 ml of 0.05 M carbonate buffer (pH 10.2) to equilibrate in the spectrophotometer and the reaction was started by adding 0.3 ml of freshly prepared 0.3 mM adrenaline to the mixture that was quickly mixed by inversion. The reference cuvette contained 2.5 ml buffer, 0.3 ml of substrate (adrenaline) and 0.2 ml of water. The increase in absorbance at 480 nm was monitored every 30 seconds for 150 seconds.

2.9 Determination of total nitrite (NO)

Nitrite determination was done using the method described by Ignarro, Buga, Wood, Byrns and Chaudhuri, (1987). The assay relies on a diazotization reaction that was originally described by Griess in 1879. The procedure is based on the chemical reaction which uses sulfanilamide and naphthylethylenediaminedihydrochlorate (NED) under acidic conditions. Sulfanilamide and NED compete for nitrite in the Griess reaction.

2.10 Statistical analysis

The results are expressed as mean ± S.E.M (Standard Error of Mean). Significance of mean values of different parameters between the groups was analyzed using one-way analysis of variance (ANOVA). Multiple comparisons were performed using the Bonferroni *post hoc* analysis. All analyses were performed using GraphPad Prism 8 and differences were considered statistically significant at probability level less than 0.05 for all tests.

3. Results

3.1 Mean iodine and caloric content of normal rat feed

Table 1 shows the mean iodine and caloric content of normal rat chow. The iodine content of the feed, measured in mg/kg, is about 5.68 mg/kg, while the caloric content has been shown to be about 3.97 kcal/g of rat feed. However, the iodine content (5,680.01) in the rat chow was significantly ($p<0.05$) higher than the caloric content (3.97 ± 0.001).

Table 1. Mean iodine and caloric content of normal rat feed

Sample	Iodine (mg/kg)	Gross energy (Kcal/g)
Normal rat feed	5.68 ± 0.01*	3.97 ± 0.001

3.2 Effects of high consumption of iodine supplement on thyroid hormonal function in diabetic rats

The mean T4 levels of rats with and without diabetes were not significantly different. Additionally, diabetic rats' serum T4 concentrations were shown to be lower. As seen in Figure 1, a one-way ANOVA and *post hoc* analysis revealed that excessive iodine supplementation considerably (p less than 0.05) decreased T4 levels and increased TSH levels in rats, as compared to diabetic groups. In contrast to the control group, neither the non-diabetic nor the iodine-supplemented groups alone showed any appreciable alterations in T3 (Figure 1c).

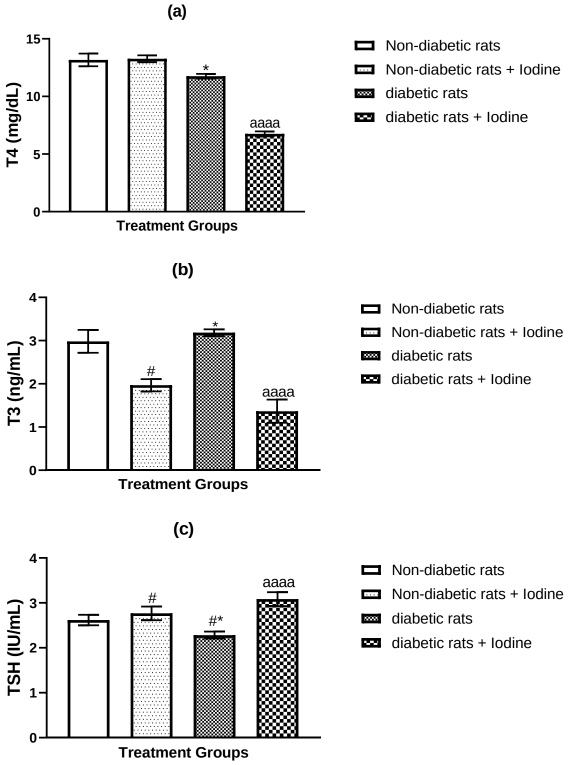

Figure 1. Effects of high consumption of iodine supplement on thyroid hormonal function in diabetic rats

3.3 Effects of high consumption of iodine supplement on antibody index for thyroperoxidase (TPO) in diabetic rats

As shown in Table 2, excess iodine supplementation shows negative level of thyroperoxidase antibody among diabetic and diabetic treated with iodine supplementation groups, when compared to the control group. This is an indication that iodine intake did not cause the development of thyroperoxidase antibody.

Table 2. Effects of high consumption of iodine supplement on antibody index for thyroperoxidase (TPO) in diabetic rats

Groups	1	2	3	4
	0.1	0.1	0.1	0.1

3.4 Effects of high consumption of iodine supplement on antibody index for thyroglobulin (TG) in diabetic rats

As shown in Table 3, excessive iodine supplementation shows a negative value for the antibody index for thyroglobulin in diabetics and diabetics treated with iodine supplementation, when compared to the control group. This is an indication that iodine intake did not lead to the formation of thyroglobulin antibodies.

Table 3. Effects of high consumption of iodine supplement on antibody index for thyroglobulin (TG) in diabetic rats

Groups	1	2	3	4
	0.1	0.1	0.1	0.1

3.5 Effects of high consumption of iodine supplement on oxidative status in diabetic rats

Rats given iodine alone showed a considerable rise in MDA levels compared to the controls (Figure 2a), although there were no discernible differences between the rats given iodine alone and the control group in terms of their SOD levels (Figure 2b). A one-way ANOVA and *post hoc* test revealed that excessive iodine supplementation in diabetic rats increased MDA and SOD significantly (p less than 0.05) in comparison to diabetic rats. In contrast to the control group, neither the diabetes nor the iodine-supplemented groups showed any appreciable alterations in SOD (Figure 2b).

3.6 Effects of high consumption of iodine supplement on nitric oxide in diabetic rats

In diabetic rats, excessive iodine supplementation increased NO levels significantly (p less than 0.05) compared to untreated diabetic rats, as demonstrated in Figure 3, from a one-way ANOVA and *post hoc* test. However, neither the diabetic nor the iodine supplemented groups showed any discernible differences in NO levels from the control group.

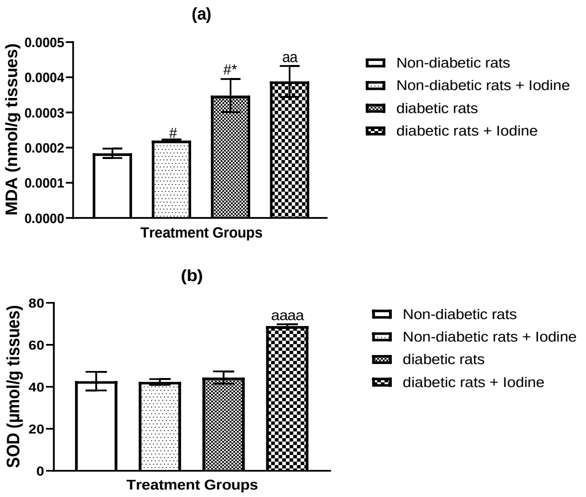

Figure 2. Effects of high consumption of iodine supplement on oxidative status in diabetic rats

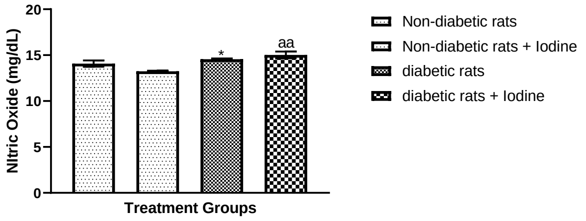

Figure 3. Effects of high consumption of iodine supplement on nitric oxide in diabetic rats

3.7 Effects of high consumption of iodine supplement on body weights of rats

Figure 4 depicts the results of high iodine supplementation. As shown in Figure 4, the body weight of diabetic rats alone increased significantly more than those of the iodine-treated group alone, as well as of the control group.

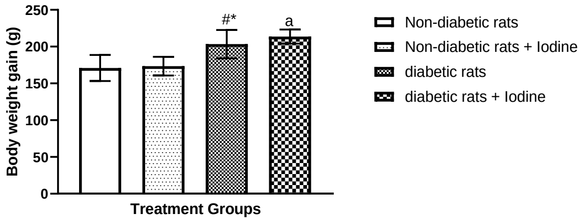

Figure 4. Effects of high consumption of iodine supplement on body weights of rats

4. Discussion

The amount of iodine found in the analysis of typical rat food was based on data from Halverson, Zepplin and Hart (1949). The standard rat food had an appropriate amount of iodine, according to the rodents' estimated daily iodine needs. Therefore, excessive extra iodine (10 mg/kg feed) was given. It is well-known that hypothyroidism makes people gain weight. When compared to diabetic rats not given supplementary iodine (group III), group IV's diabetic rats gained more weight, which may be attributable to the lower serum -T4 concentrations in that group. Diabetes affects the pituitary-thyroid axis, increasing the occurrence of thyroid abnormalities. Diabetes appears to alter hypothalamic thyrotropin-releasing hormone (TRH) secretion and pituitary thyrotropin (TSH) release. Excess thyroid hormones have been investigated to cause “thyroid diabetes” (Hartoft-Nielsen *et al*., 2009), whereas hyperthyroidism causes glucose intolerance in animals and humans (Hartoft-Nielsen *et al*., 2009). Diabetes mellitus has been proven to coexist with a range of thyroid disorders in humans, as also indicated in our animal study. In accordance with this, Wolff and Chaikoff (1948) reported that an iodine injection in rats almost completely inhibited organification (iodide oxidation) in the thyroid gland, which lasted for about ten days, and was then followed by an escape phenomenon known as adaptation and restoration of normal organization of iodine and normal peroxidative function of the thyroid. In free-living populations, Katagiri, R., Yuan, X., Kobayashi, S., and Sasaki, S. (2017) observed that salts with poor control or continuous exposure to excessive iodine from water are risk factors for hypothyroidism.

Given that the experimental period was longer than ten days and that Wolf and Chaikoff (1948) observed the escape phenomenon over a longer period of time, the results in non-diabetic rats are compatible with their findings. Additionally, the lack of thyroid autoimmune antibodies such as the thyroid peroxidase antibody (TPOAb) and the thyroglobulin antibody (TgAb) suggests that high iodine intake had no appreciable impact on thyroid function in non-diabetic rats fed standard rat food. According to findings by Ravindra *et al.* (2011), diabetic serum has lower iodine uptake. They claim that elevated blood sugar levels may be the root of the lower iodine intake, since they can modify the structure of biomolecules through glycation, which reduces the iodine binding sites. Since less iodine is available for the production of thyroid hormones, this may be the cause of the markedly lower mean serum content of T4 seen in diabetic rats fed standard rat food. However, it is unclear what caused the mean serum TSH to be considerably lower.

Lower T4 levels and higher TSH levels compared to diabetic rats not given supplementary iodine orally were indications that iodine consumption at a dose of 10 mg/kg food had the ability to further depress thyroid function in diabetic rats. The mean serum T3 levels, however, did not differ significantly from one another. TSH is released more frequently by the anterior pituitary gland when thyroid hormone levels are lower (Hall, 2008). The higher serum TSH values seen in this study are therefore likely caused by reduced blood T4 levels. Although thyroid peroxide antibodies (TPOAb) and thyroglobulin antibodies (TGAb) were noticed, they have already been characterized by Lindberg, Ericsson, Ljung and Ivarsson (1997) and Otken *et al.* (2006). In this investigation, they were not seen in patients with type 1 diabetes.

MDA levels have been observed to rise in both hypo- and hyperthyroidism (Chakrabarti, Ghosoh, Banerjee, Mukherjee & Chowdhury, 2016; Cheserek, Wu, Ntazinda, Shi, Shen, & Le, 2015; Dumitriu, Bartoc, Ursu, Purice, Ionescu, 1988; Mancini *et al*., 2016). Notably, the considerably elevated serum MDA levels and serum NO levels found with iodine intake show that excessive iodine consumption in rats caused oxidative stress via a mechanism that did not impact thyroid function. According to Verma *et al*. (1991), injection of TSH led to a marked decrease of SOD in the adrenal gland. The serum SOD concentrations likely rose as a result of the adrenal gland's depletion of SOD. Given that their mean TSH levels were noticeably raised, this may be the cause of the elevated blood SOD levels seen in diabetic rats fed iodine and standard rat food. The creation of ROS, which can result in thyroid gland oxidative damage and put a diabetic patient at risk for thyroid disease, is one of the factors contributing to the thyroid cytotoxicity seen in diabetic rats (Mohamed *et al*., 2017). According to the findings of Mohamed *et al*. (2017), DM cases are more prone to experience problems when an abnormality is present. According to the current study, increased iodine intake promotes thyroid cytotoxicity and changes the antioxidant defense system, leading to an increase in oxidative stress in both normal and diabetically poisoned rats.

This study supports Messarah, Saoudi, Boumendjel, Boulakoud and Feki (2010) regarding the findings that the antioxidant system was altered in hypo-/hyperthyroidism-induced rats through an increase in the activities of catalase, glutathione peroxidase, and superoxide dismutase, as well as a decrease in glutathione (GSH) concentration. In addition, extra iodine dramatically raised MDA and antioxidants in rats with normal and hypothyroid function, according to Hussein, Abbas, Wakil, Elsamanoudy and El Aziz (2012). The high iodine intake tested in this study is particularly notable for the increased accumulation of oxido-nitrogen stress indicators like MDA and NO as well as the alteration in antioxidant function. To avoid oxido-nitrogen stress-induced thyroid cytotoxicity and enable more effective therapeutic intervention, it is crucial to moderate iodine intake in the management of the diabetic condition.

5. Conclusions

The regular rat food's iodine content was adequate to provide the daily dose. Thyroid function was reduced by diabetes mellitus. The thyroid function of diabetic rats was further inhibited by excessive iodine consumption, and both diabetic and non-diabetic rats had an increase in oxidative stress. Iodine needs for diabetics are higher than for non-diabetics, although excessive iodine consumption should be avoided as it can harm thyroid function.

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References

Association of Official Analytical Chemist. (1984). *Official methods of analysis* (14th ed.). Arlington, TX: Author.

Adam Vizi, V., & Seregi, M. (1982). Receptor dependent stimulatory effect of noradrenaline on Na+/K+ ATPase in rat brain homogenate role of lipid peroxidation. *Biochemical Pharmacology*, *31*, 2231-2236. Retrieved from http://dx.doi.org/10.1016/0006-2952(82)90106-X

Akinola, O., Gabriel, M., Suleiman, A., & Olorunsogbon, F. (2012). Treatment of alloxan-induced diabetic rats with metformin or glitazones is associated with amelioration of hyperglycaemia and neuro protection. *The Open Diabetes Journal*, *5*, 8-12

Arigi, Q. E., Fabiyi, T. D., & Fasanmade, A. A. (2014). Thyroidectomy and thyroxine replacement caused impaired oral glucose tolerance in rat world. *Journal of Medical Sciences, 11*(*3*)*, 348-352*.

Chakrabarti, S. K., Ghosoh, S, Banerjee, S., Mukherjee, S., Chowdhury, S. (2016). Oxidative stress in hypothyroid patients and the role of antioxidant supplementation. *Indian Journal of Endocrinology and Metabolism*, *20*(5), 674-78. doi:10.4103/2230-8210.190555. PMCID: PMC5040049.

Cheserek, M. J., Wu, G., Ntazinda, A., Shi, Y, Shen, L., & Le, G. (2015). Association between thyroid hormones, lipids and oxidative stress markers in subclinical hypothyroidism. *Journal of Medical Biochemistry, 34*(3), 323-331.

Chijiokwu, E. A., Nwangwa, E. K., Oyovwi, M. O., Naiho, A. O., Emojevwe, V., Ohwin, E. P., . . . Ogheneyoma, O. O. (2022). Intermittent fasting and exercise therapy abates STZ-induced diabetotoxicity in rats through modulation of adipocytokines hormone, oxidative glucose metabolic and glycolytic pathway. *Physiological Reports, 10,* e15279. Retrieved from https://doi.org/10.14814/phy2.15279

Cooper, R. A. (2007). Iodine revisited. *International Wound Journal*, *4*(2), 124-37.

Department of Health, the United Kingdom. (1995). Dietary reference value for food energy and nutrients for the United Kingdom. Report on health and social subjects No. 41. London, England: Author.

Dumitriu, L., Bartoc, R., Ursu, H., Purice, M., & Ionescu, V. (1988). Significance of high levels of serum malonyldialdehyde (mda) and ceruloplasmin (cp) in hyper- and hypothyroidism. *Endocrinologie*, *26*(1), 35-38.

Haldimann, M., Bochud., M., Burnier, M., Paccaud, F., & Dudler, V. (2015). Prevalence of iodine inadequacy in Switzerland assessed by the estimated average requirement cut-point method in relation to impact of iodized salt. *Public Health Nutrition, 18*(8), 1333.

Hall, J. E. (2008). Guyton and hall textbook of medical physiology (11th ed.). Amsterdam, the Netherlands: Elsevier.

Halverson, A. W., Zepplin, M., & Hart, E. B. (1949). Relation of iodine to the goitrogenic properties of soybeans: Four figures. *The Journal of Nutrition*, *38*(2), 115.

Hartoft-Nielsen, M.-L., Rasmussen, A. K., Bock, T., Feldt-Rasmussen, U., Kaas, A., & Buschard, K. (2009). Iodine and tri-iodo-thyronine reduce the incidence of type 1 diabetes mellitus in the autoimmune prone BB rats. *Autoimmunity*, *42*(2), 131–138. doi:10.1080/08916930802438774

Hussein, Ael-A., Abbas A. M., El Wakil, G. A., Elsamanoudy, A. Z., & El Aziz, A. A. (2012). Effect of chronic excess iodine intake on thyroid function and oxidative stress in hypothyroid rats. *Canadian Journal of Physiology and Pharmacology*, *90*(5), 617-25. doi:10.1139/y2012-046. Epub 2012 May 2. PMID: 22550940.

Ignarro, L. J., Buga, G. M., Wood, K. S., Byrns, R. E., & Chaudhuri, G. (1987). Endothelium-derived relaxing factor produced and released from artery and vein is nitric oxide. *Proceedings of the national academy of sciences of the United States of America 84*, 9265-9269.

Kabadi, U. M., & Eisenstein, A. B. (1980). Glucose intolerance in hyperthyroidism: role of glucagon. *Journal of Clinical Endocrinology and Metabolism, 50*, 392-96

Katagiri, R., Yuan, X., Kobayashi, S., & Sasaki, S. (2017). Effect of excess iodine intake on thyroid diseases in different populations: A systematic review and meta-analyses including observational studies. *PLos ONE, 12*(3): e0173722.

Kotyzováa, D., Eybla, V., Mihaljevičb, M., & Glattre, E. (2005). Effect of long-term administration of arsenic(iii) and bromine with and without selenium and iodine supplementation on the element level in the thyroid of rat. *Biomedical Papers of the Medical Faculty of the University Palacky, Olomouc, Czech* Republic, *149*(2), 329–33.

Lindberg, B., Ericsson, U. B., Ljung, R., & Ivarsson S. A. (1997). High prevalence of thyroid antibodies at diagnosis of insulin-dependent diabetes mellitus in swedish children. *Journal of Laboratory and Clinical Medicine, 130*(6) 585-9.

Macini, A., Segni, C., Raimondo, S., Olivieri, G., Silvestrini, A., Meucci, E., & Curro, D. (2016). thyroid hormones, oxidative stress and inflammation. *Journal of Meidators of Inflammation, 2016*, 6757154.

Macini, F. R., Rajaobeli, K., Dow, C., Habbal, T., Affret, A., Balkau, B., . . . Fagherazzi, G. (2019). High iodine intake is associated with type 2 diabetes among women of the E3N-EPIC cohort study. *Journal of Clinical Nutrition, 38*(2019), 165.

Messarah, M., Saoudi M, Boumendjel, A., Boulakoud, M. S., & Feki, A. E. (2010). Oxidative stress induced by thyroid dysfunction in rat erythrocytes and heart. *Environmental Toxicology and Pharmacology, 31*(1), 33-41.

Misra, H. P., & Fridovich, I. (1972). The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. *Journal of Biology and Chemistry, 247*(10), 3170-3175.

Mohamed, N. A., & Gawad, H. S. A. (2017). Taurine dietary supplementation attenuates brain, thyroid, testicular disturbances and oxidative stress in streptozotocin-induced diabetes mellitus in male rats. *Beni-Suef University Journal of Basic and Applied Sciences, 6*(3), 247–252. doi:10.1016/j.bjbas.2017.04.006

Otken, A., Akcay, S., Cakir, M., Girisken, I., Kosucu, P., & Deger, O. (2006). Iodine Status, thyroid function, thyroid volume and thyroid autoimmunity in patients with type 1 diabetes mellitus in an iodine-replete area. *Diabetes and Metabolism, 32*(4):323-9.

Oyovwi, M. O., Ben-Azu, B., Tesi, E. P., Oyeleke, A. A., Uruaka C. I., Rotu, R. A., & Aya-Ebi., E. O. (2021) Repeated endosulfan exposure induces changes in neurochemicals, decreases ATPase transmembrane ionic-pumps, and increased oxidative/nitrosative stress in the brains of rats: Reversal by quercetin. *Pesticide Biochemistry and Physiology, 175*, 104833. Retrieved from https://doi.org/10.1016/j.pestbp.2021.10483

Ravindra, M., Vivek, R. J., Ayaz, K. M., Manjunath, G., Jeevan K. S., Prakash, M., . . . Shivaraj, B. (2011). A comparative study on iodination of normal and diabetic serum. *International Journal Applied Biological and Pharmaceutical Technology, 2*(1)

Rhoades, R. A., & Tanner, G. A. (2004). *Medical physiology* (2nd ed.). Philadelphia, PA: Lippincott Williams and Wilkins.

Sikarwar, M. S., & Patil, M. B. (2010). Antidiabetic activity of crateva nurvala stem bark extracts in alloxan-induced diabetic rats. *Journal of Pharmacy and Bioallied Sciences, 2*(1), 18.

Varshney, R., & Kale, R. K. (1990). Effects of calmodulin antagonists on radiation-induced lipid peroxidation in microsomes. *International Journal of Radiation Biology, 58*(5), 733-743.

Verma, S., Pradeep, K. G., Laloraya, M., Nivsarkar, M., & Singh, A. (1991). Superoxide dismutase activation in thyroid and suppression in adrenal novel pituitary regulatory routes. *FEBS Letters, 282*(2), 310-312.

Wajchenberg, B. L., Cesar, F. P., Leme, C E., Souze, I. T., Pieroni, R. R., & Mattar, E. (1978). carbohydrate metabolism in thyrotoxicosis: studies on insulin secretion before and after remission from the hyperthyroid state. *Hormonal and Metabolic Research, 10*, 294-99.

Wild, S., Roglic, G., Green, A., Sicree, R., & King, H. (2004). Global prevalence of diabetes: estimates for 2000 and projections for 2030. *Diabetes Care, 27*(5), 1047–1053. doi:10.2337/diacare.27.5.1047.PMID 15111519.

Wolff, J., & Chaikoff, I. L. (1948). Plasma inorganic iodide as a homeostatic regulator of thyroid function. *Journal of Biology and Chemistry, 174*(2), 555-64.