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# THE THYROID GLAND AND OXIDATIVE STRESS; THE ROLE OF MELATONIN

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## SUMMARY

**Reactive oxygen species (ROS) and free radicals participate in physiological and pathological processes in the thyroid gland. For example, the role of hydrogen peroxide ($H_2O_2$) is crucial for thyroid hormone biosynthesis. Also other free radicals or reactive species, formed from iodine or tyrosine residues, are produced during thyroid hormone synthesis in physiological conditions. In turn, much evidence has been accumulated, showing that thyroid diseases, e.g., Graves' disease, non-toxic goitre formation or thyroid cancer, are accompanied by enhanced oxidative stress. The presence of some antioxidants has been found in the thyroid. Melatonin (N-acetyl-5-methoxytryptamine) - the main secretory product of the pineal gland is a well-known antioxidant and free radical scavenger, widely distributed in the organism. Mutual relationships between the pineal gland and the thyroid have-for a long time-been a subject of intensive research. The abundant to-date's evidence relates mostly to the inhibitory action of melatonin on the thyroid growth and function. Recently, experimental models have been developed, showing- among others-protective effects of melatonin against oxidative damage to lipids in the thyroid gland. Thus, free radicals and antioxidants, melatonin included, may participate in both physiological and pathological processes in the thyroid, what has already been partially documented.**

Key words : Antioxidants; Free radicals; Melatonin; Oxidative stress; Thyroid gland.

## INTRODUCTION

Reactive oxygen species (ROS) and free radicals are produced in living organisms, in certain amounts, under physiological conditions. An overproduction of ROS and free radicals causes oxidative stress and can lead to several diseases (1, 2). The most basic reaction of oxidative stress is Fenton reaction, in which iron [as ferrous ion ($Fe^{2+}$)] participates: $Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + {}^{\cdot}OH + OH^{-}$. Hydroxyl radical ($^{\cdot}OH$ ), produced in this reaction, is the most toxic free radical.

Melatonin (N-acetyl-5-methoxytryptamine) is produced in the pineal gland, as well as in numerous other tissues and organs (3). The indoleamine possesses properties of a hormone but also demonstrates other numerous characteristics, allowing its action in cellular compartments of all tissues and organs (4). Melatonin is a well-known antioxidant and free radical scavenger, revealing preventive action against oxidative damage in different tissues (5-9).

### Relationship between the pineal gland and the thyroid gland

In different animal species, a suppressive effect of melatonin on thyroid growth processes (10-14) and thyroid function (15-17) was shown. In contrast, thyroid hormones reveal mainly stimulatory effects on the pineal, as regards growth and secretory processes of this gland (18). Experimental evidence, concerning the relationship between the thyroid and the pineal gland, also in relation to oxidative stress (18-23) has recently been summarized.

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## The role of oxidative stress in the thyroid gland- experimental evidence

### Hydrogen peroxide ($H_2O_2$)

$H_2O_2$ is produced in the thyroid gland (24, 25) by the NADPH oxidase system of the apical membrane of thyroid follicular cells (thyrocytes) (26, 27); divalent reduction of oxygen, without superoxide anion radical ($O_2^{\cdot-}$) generation, is involved in this process (28).

$H_2O_2$ is an essential factor for thyroid hormone synthesis, acting as an electron acceptor at each step of this process, namely iodide oxidation and, next, its organification, and coupling reaction of iodotyrosines (23). This reactive species is crucial for thyroid peroxidase (TPO) activity. TPO is the key enzyme for thyroid hormone synthesis. It is a heme-dependent protein, thus it contains iron. On one hand, increased iron stores in the organism are associated with an increased risk of several diseases, cancer included (1, 29). On the other hand, however, iron is an essential element for metabolism in different tissues and organs, also in the thyroid. It has been shown in studies on animals and in human subjects that iron deficiency reduces TPO activity, enhances the consequences of iodine deficit, while iron supplementation improves the efficacy of iodine supplementation (30).

Beside the main three steps of thyroid hormone synthesis, $H_2O_2$ also participates in autocatalytic covalent heme binding to the apoprotein of TPO molecule, thereby stabilizing the activity of the enzyme (31). It has also been found that either ROS or free radicals decrease TPO activity in the thyroid gland, without influencing TPO mRNA; the authors have speculated that TPO inactivation occurs at the heme-linked histidine residue of the enzyme molecule; this residue plays a critical role for TPO activity due to the presence of heme-derived iron, which potentially constitutes a substrate for Fenton reaction (28). Thus, while $H_2O_2$ is necessary for thyroid hormone synthesis, this ROS when being in excess may inhibit TPO activity, with a subsequent inhibition of thyroid hormone formation (28).

There are presumptions that oxidative stress may be involved in the pathomechanism of hypothyroidism and goiter formation. Using dog thyroid cells, it has been found that thyrotropin (TSH) - the main hormone, stimulating secretory and growth processes in the thyroid - enhances $H_2O_2$ generation through the cyclic adenosine 3',5'-monophosphate (cAMP) cascade (25). Thus, in any conditions of increased blood TSH concentration, mainly in case of hypothyroidism and in chronic stimulation of the thyroid gland due to iodine deficiency, an increased production of $H_2O_2$ must take place with subsequently enhanced formation of free radicals (especially $^{\cdot}OH$ ).

It is suggested that $H_2O_2$ participates in Wolff-Chaikoff 's effect, i.e. in a phenomenon which relies on the fact that iodides, when in excess, are able to inhibit iodine organification and subsequent thyroid hormone synthesis. Iodide was shown to strongly inhibit both protein iodination and $H_2O_2$ generation stimulated by TSH in dog thyroid slices (32). It is suggested that both apoptosis and necrosis occur in the thyroid gland via the mechanism involving $H_2O_2$, the former process resulting from lower, and the latter process from higher concentrations of this species under *in vitro* conditions (33). Because melatonin directly neutralizes $H_2O_2$ (34) and is the most effective scavenger of $^{\cdot}OH$ (4), the indoleamine might prevent pathological processes in the thyroid caused by an excessive amount of $H_2O_2$.

### Reactive nitrogen species

Nitric oxide synthase (NOS) catalyzes the formation of free radical-nitric oxide ($NO^{\cdot}$). An expression of mRNA for the three isoforms of NOS - brain (type I), endothelial (type III), and inducible (type II) has been detected in the rat thyroid gland (35-37). Nitric oxide has been found to inhibit cell proliferation in cultured human thyrocytes (38). Some data suggest that $NO^{\cdot}$ participates in the regulation of thyroid hormone synthesis. While $NO^{\cdot}$ inhibits TSH-stimulated iodide uptake by-probably-stimulation of guanylyl cyclase (GC) activity and cyclic GMP (cGMP) production in calf thyroid (39),

the radical has been found to stimulate thyroid peroxidase activity in monolayer cultures of primary human thyrocytes (40).

It is known that melatonin directly neutralizes $NO^{\cdot}$ (41) and peroxynitrite anion ($ONOO^{-}$) (42), the latter being a highly destructive product of the interaction between the $O_2^{\cdot-}$ and $NO^{\cdot}$. Additionally, melatonin has been demonstrated to inhibit the activity of NOS (5). There is also an indirect evidence that melatonin detoxifies $O_2^{\cdot-}$ (43).

**Other free radicals**

Numerous free radicals and reactive species are undoubtedly involved in the complex process of thyroid hormone synthesis. According to experimental evidence or suggestions, the following radicals or reactive species are of special importance: tyrosine free radical ($Tyr^{\cdot}$), diiodotyrosyl residue radical ($DIT^{\cdot}$), diiodotyrosyl residue radical in thyroglobulin ($Tg\text{-}DIT^{\cdot}$), iodine radical ($I^{\cdot}$), iodinium ion ($I^{+}$), hypoiodous acid intermediate [$IO^{\cdot}(IOH)$], and ascorbate radical ($Asc^{\cdot}$) (44).

**Antioxidative defence mechanisms in the thyroid gland**

An antioxidative defence system in the thyroid gland comprises both antioxidative enzymes and free radical scavengers. The presence of the following antioxidative enzymes in the thyroid gland has been documented: superoxide dismutase (SOD) (45, 46), glutathione peroxidase (GSH-Px) (47), and catalase (CAT) (48). It should be stressed here that melatonin has been documented to stimulate the activity of SOD, GSH-Px, and CAT (5, 6). Moreover, the presence of glutathione (GSH), a well known intracellular antioxidant, alpha- and gamma-tocopherols, and coenzyme Q has been found in human thyroid (49), and of ascorbic acid in hog thyroid (50). Ascorbic acid becomes a free radical itself in the process of scavenging free radicals in the thyroid gland (50).

Peroxiredoxins (Prxs), antioxidative proteins involved in the regulation of cell differentiation and proliferation, have been found in human thyroid follicular cells and in FRTL-5 cells (51). Using FRTL-5 cells, it has been documented that Prxs are involved in the process of $H_2O_2$ elimination, when this oxygen species is produced in response to TSH, and that they protect thyroid cells from $H_2O_2$-induced apoptosis (51).

A positive immunostaining with antibodies against melatonin has been found in C cells in the rat thyroid gland (3, 52). It has not been examined till now whether melatonin is produced in thyroid follicular cells. However, because melatonin is able to reach any tissue or organ within very short time (5, 6), it is highly probable that the indoleamine is also available for the thyroid gland, when it is required to reveal antioxidative effects.

**Oxidative changes in the thyroid gland under pathological conditions**

The results obtained in humans and in animal models suggest that oxidative damage in the thyroid is accompanied by increased activities of antioxidative enzymes or increased production of antioxidants, what probably represents the defence mechanism (46, 47, 53). In case of human thyroid tissue - non-toxic nodular goitre, carcinoma (follicular and papillary) and follicular adenoma - the highest level of lipid peroxidation products was found in carcinomas and it was still increased in adenomas, when compared to the level of lipid peroxidation products in control tissue and in nodular goitre; those changes were accompanied by increased activities of antioxidative enzymes, such as SOD, GSH-Px, and CAT, especially in carcinomas (47).

A much higher amount of Prx I, measured by immunoblotting, has been found in follicular thyroid adenomas and carcinomas than in histopathologically unchanged thyroid tissue; additionally,

a significantly higher amount of Prx I has been found in thyroids collected from patients with Graves' disease than in normal thyroid tissue (54).

The presence of immunochemical staining of SOD type II (inducible), and of mRNA for this enzyme was found in the human papillary thyroid carcinoma but not in normal human thyroid tissue (55). On the contrary, a decreased expression of mRNA encoding for CAT, and copper and zinc SOD were found in anaplastic thyroid carcinoma, when compared to histopathologically unchanged thyroid tissue and to differentiated thyroid tumors (56).

Thus, in case of more advanced tumors or, generally, more serious diseases of the thyroid gland, more free radicals are generated, followed by an increased production of antioxidants or increased activities of antioxidative enzymes. However, at extremely advanced stages of diseases, like anaplastic carcinoma, the defence mechanisms remain inactive.

Regarding the protective effects of melatonin against oxidative stress in the thyroid gland, experimental data have unfortunately been very scarce. However, we have found recently a protective action of melatonin against lipid peroxidation in porcine thyroid. Because iron is present in TPO, and $H_2O_2$ is essential for TPO activity, the thyroid gland may - under pathological conditions - be exposed to excessive amounts of either $Fe^{2+}$ or $H_2O_2$, or both. Ferrous iron and $H_2O_2$ constitute substrates for Fenton reaction.

Using an *in vitro* model of Fenton reaction, we have induced oxidative damage to lipids in homogenates of porcine thyroids; we have found that $Fe^{2+}$ plus $H_2O_2$ increased - in the $Fe^{2+}$ concentration - dependent manner - the level of lipid peroxidation products [malondialdehyde + 4-hydroxyalkenals (MDA+4-HDA)]; at the same time, melatonin - in a concentration-dependent manner - decreased lipid peroxidation induced by Fenton reaction, with the lowest effective concentration of 0.25 mM (57). We would like to indicate that the above study is the first one showing lipid peroxidation induction by Fenton reaction and the protective influence of melatonin against oxidative damage in the thyroid tissue.

**Involvement of free radicals and reactive species in thyroid enlargement**

Several experimental data, as well as results of studies in humans, support the hypothesis that oxidative stress is involved in goitre formation. It is supposed that $NO^{\cdot}$ participates in vascular control during goitre formation. In animal model of thiouracil - and low iodine diet - induced goitre, the increased vascularization of the gland was accompanied by an induction of genes encoding for NOS I and NOS III; during goiter involution, the expression for these genes returned to basal values (37).

In another study, low iodine diet resulted as expected in an increased thyroid weight, increased DNA and protein content, and increased $^3$H-thymidine incorporation into DNA of thyroid follicular cells; all those changes were reduced by an administration of an antioxidant - vitamin E (58). It is worth stressing that the changes in parameters of growth processes after vitamin E treatment were not accompanied by any changes in either TSH or thyroid hormone concentrations (58). These results suggest that vitamin E has a direct antigoitrogenic effect.

Further studies, performed by the same group of authors, support the above suggestion. They have observed that vitamin E deficiency resulted in a significant increase in epithelial cell necrosis during goitre development and involution, and in increased lipid peroxidation and decreased GSH-Px activity in the thyroid (59).

Studies in humans suggest that iodine deficiency is associated with an increased oxidative stress and decreased antioxidative defence. A decreased SOD activity was observed in endemic

goiter tissue (45). In iodine-deficient children with goitre, the activities of antioxidative enzymes (GSH-Px, CAT and SOD) in erythrocytes, and selenium concentration in plasma and erythrocytes, have been found to be significantly lower than their respective values in non-goiter and non iodine-deficient control subjects, and even, what is most important, than in non-goiter but iodine-deficient children (60). Thus, it seems plausible that low activities of antioxidant enzymes and low selenium concentrations contribute to thyroid enlargement in iodine-deficient subjects.

In another study, increased damage to genomic DNA (evaluated by the measurement of the concentration of different oxidized bases in peripheral blood) was found in children (aged 15-8 years) with goitre, living in the area of severe or moderate iodine deficiency; DNA damage was accompanied by decreased blood concentration of thyroid free hormones, decreased concentration of selenium,

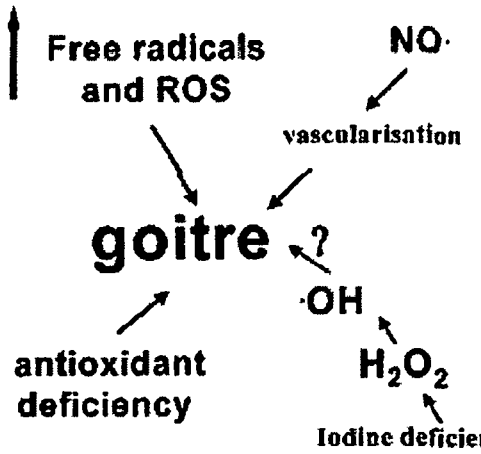


Fig. 1 The proposed mechanism - related to oxidative *stress - of thyroid enlargement (goitrogenesis).* $H_2O_2$, hydrogen peroxide; NO·, nitric oxide; ·OH, hydroxyl radical; ROS, reactive oxygen species; TSH, thyrotropin

decreased activity of GSH-Px and SOD in blood erythrocytes, and by decreased ioduria (61). The observation that iodine deficiency, resulting in goitre, leads to oxidative DNA damage, supports the view on an essential relationship between iodine deficiency and the increased incidence of thyroid cancer.

It has recently been found that melatonin concentration, measured at night, was significantly higher after than before the operation performed in patients with very large non-toxic nodular goitre (62). These results, for some reasons - unexpected, could be explained as follows: melatonin might actively be taken up by an enlarged thyroid with a subsequent decrease in blood concentration of the indoleamine before surgery.

The proposed mechanism - related to oxidative stress - of thyroid enlargement (goitrogenesis) is presented in Figure 1.

**Thyroid autoantigens and oxidative stress**

Thyroglobulin (Tg) and TPO belong to the main thyroid autoantigens. Thyroglobulin is stored extracellulary in the thyroid follicle lumen and is essential for thyroid hormone biosynthesis. During thyroid hormone synthesis, resulting from several oxidative reactions, the multimerized inactive form of Tg (mTg) is formed (63). It has recently been found that experimentally-induced oxidative stress participates in mTg fragmentation, producing Tg molecules with a recovered ability to store thyroid hormones; at the same time, the fragmentation of mTg precludes its excessive accumulation in the thyroid and, consequently, the thyroid enlargement (63).

Under experimental conditions of Fenton reaction or in the presence of ferric ions ($Fe^{3+}$) - both causing oxidative stress - a production of immunoreactive C-terminal fragments (40 kDa) of Tg

was observed and that process was accompanied by thyroid hormone synthesis (64). Using human thyroid cells, $H_2O_2$-being in excess - was shown to produce the same 40 kDa immunoreactive fragments of Tg; it is worth stressing that both $H_2O_2$ and iodide have been indispensable to cause Tg fragmentation (65), what suggests that Tg cleavage occurs during thyroid hormone synthesis. Of great importance is the fact that immunoreactive Tg fragments have been found only in dead cells but not in living ones and that those fragments are able to enter living thyrocytes (65), what would start autoimmune response.

Concerning another thyroid autoantigen - TPO - worth mentioning are studies on oxidative stress and myeloperoxidase (MPO), which is very similar in structure and properties to TPO. Whole body ionizing radiation (800 cGy) resulted in a decreased level of glutathione, increased levels of lipid peroxidation products and increased levels of MPO (as the index of neutrophil infiltration) in different tissues, the changes which were reduced by a co-administration of melatonin (66). It is highly probable that melatonin is also able to modify the level of TPO protein under conditions of excessive oxidative stress. No studies have till now been performed, showing that oxidative stress is involved in autoimmunity directed against TPO. Some other results are worth mentioning, concerning the relationship between oxidative stress and thyroid autoimmunity.

It has been documented that free radicals are involved in interleukin - 1β - induced glycosaminoglycan production by retro-ocular fibroblasts and their accumulation in patients with. Graves' disease (67). Furthermore, it has been shown that $NO^{\cdot}$ is involved in interleukin - 1α - induced cytotoxicity in polarized human thyrocytes, suggesting that this free radical may promote the exposure of autoantigens to the immune system (68).

The relationship between oxidative stress and thyroid autoimmunity suggests a potential protective role of antioxidants in autoimmunological diseases. However, the application of antioxidants in patients with autoimmunological diseases or patients, predisposed genetically to these disorders, is still the point of discussion. As far as the influence of melatonin on the immune system is concerned, divergent results exist (69, 70). However, the precise effect, caused by the indoleamine in patients with autoimmunological diseases, as well as the potential indications and contraindications for the treatment with this substance in such patients remain to be determined.

**Effects of thyroid hormones on oxidative processes**

Thyroid hormones regulate energy metabolism, among others, by their effects on mitochondria (71), in which free radicals and ROS are produced. Under *in vitro* conditions, thyroid hormones may act as antioxidants; thyroid hormones [3,5,3'-triiodothyronine, $T_3$; and 3,5,3',5'-tetraiodothyronine (thyroxine), $T_4$] and their structural analogues (L-thyronine, $T_0$; 3,5,3'-triiodothyroacetic acid, $TA_3$; 3,5,3',5'-tetraiodothyroacetic acid, $TA_4$) were found to reveal antioxidative properties (72-74). For example, using different models of oxidative stress, it was shown that all the above mentioned hormones or their analogues revealed a capacity to scavenge free radicals, $^{\cdot}OH$ and $O_2^{\cdot-}$ included (shown for $T_3$), and that $T_0$, $T_3$, and $TA_3$ effectively prevented the formation of lipid peroxidation products - conjugated dienes and thiobarbituric acid reactive substances - during LDL oxidation (74). It was documented in the same study that the 4'-hydroxydiphenylether structure of thyroid compounds was necessary for their free radical scavenging activity. This has been confirmed by antioxidant effects of $T_0$, which has the basic structure of thyroid hormones but does not possess iodine atom; conversely, monoiodotyrosine (MIT), which possesses the mono-iodosubstituted phenolic ring of $T_3$, does not reveal any antioxidative effects (74).

**Effects of thyroid hormone excess**

Changes in the values of oxidative parameters in different tissues were observed, due to thyroid hormone excess resulting from either thyroid diseases or treatment with thyroid hormones. In blood samples, collected from hyperthyroid patients suffering from Graves' disease, the levels of thiobarbituric acid-reacting substances and conjugated dienes (both being parameters of oxidative stress), and the activities of main antioxidative enzymes (SOD, CAT and GSH-Px) were significantly higher than those in healthy subjects: all those changes were - to a differential degree - restored in result of antithyroid drug treatment (75).

Similarly, a significant increase in malondialdehyde (MDA) concentration and a slight increase in Schiff's bases and conjugated dienes concentration (all being parameters of oxidative damage to lipids) were found in blood serum collected from hyperthyroid patients with Graves' disease; treatment with methimazole (an antithyroid drug) brought about a decrease in the values of indices of oxidative stress, accompanied by normalization of thyroid hormone and TSH concentration (76).

*In mononuclear cells, collected from hyperthyroid patients with Graves' disease, an increased* level of 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxo-dGuo) (the parameter of DNA oxidative damage) was found; additionally, 8-oxo-dGuo level correlated significantly with free $T_4$ concentration (77). It is presumed that untreated hyperthyroidism contributes to oxidative damage to DNA.

An induction of oxidative stress was also observed in experimental models of hyperthyroidism. The treatment of rats with thyroid hormones caused a production of $O_2^{\cdot-}$ and $H_2O_2$ in liver mitochondria (78) and of $NO^{\cdot}$ in liver cytosol (79). Expectedly, $T_3$-injection increased lipid peroxidation and protein oxidation in rat liver; it is worth stressing that the damage to lipids preceded the damage to protein molecules in response to thyroid hormone, what suggests different susceptibility of target molecules to excessive amounts of thyroid hormones (80).

In animal models of hyper- and hypothyroidism, $T_3$- treatment in rats brought about a significant increase in lipid peroxidation in the liver, the heart, and skeletal muscles, whereas methimazole administration (causing hypothyroidism) did not influence the level of lipid peroxidation products in those organs; at the same time, a decrease in the whole antioxidant capacity of tissues and an increase in GSH-Px and glutathione reductase (GSH-Rd) were observed in only some examined organs in both hyper- and hypothyroid rats (81).

In other animal models of hyper- and hypothyroidism, L-$T_4$ treatment in *OFI* female mice resulted in an increased sensitivity to lipid peroxidation, an increased level of oxidized GSH (GSSG), and an increased ratio GSSG/GSH, as well as increased oxidative damage to mitochondrial DNA in the heart; interestingly, a treatment with an antithyroid drug - propylthiouracil (PTU) - did not influence oxidative damage to lipids and decreased GSSG concentration and the GSSG/GSH ratio, as well as reduced oxidative damage to mitochondrial DNA (82). The latter finding is of great importance, suggesting protective effects of PTU against oxidative stress. In agreement with this, L-$T_4$, but not PTU, treatment in *OFI* female mice increased lipid peroxidation in the skeletal muscle (83).

In the study, performed at our laboratory, a 2-week-treatment with L-$T_4$ (100 μg/kg B.W., for 14 days) resulted in an increased concentration of free fractions of both thyroid hormones; interestingly, a co-treatment with melatonin (5 mg/kg B.W., for 7 days) completely prevented the increase in free $T_3$concentration (84). Thus, melatonin has abolished the increase in the concentration of this thyroid hormone, which is directly responsible for tissue effects of hyperthyroidism. At the same time, it was observed that melatonin injection resulted in decreased basal values of conjugated dienes and Schiff's bases in rat kidney (84). It has recently been found that $T_3$ causes mutagenic action through induction of oxidative stress (85).

**Effects of thyroid hormone deficiency**

It has been observed that hypothyroidism, resulting from either PTU-treatment or thyroidectomy, causes a decrease in $H_2O_2$ generation (86). Additionally, hypothyroidism resulted from thyroidectomy, prevented post-ischemic lipid peroxidation in rat kidney (87).

Thus, it is still unclear, if the above-mentioned protective effects of PTU against oxidative stress (82) are really related to direct antioxidant action of the drug or to hypothyroidism, or if they result from both mechanisms. Indeed, PTU has been shown to react directly with ·OH and, to a lesser extent, with $O_2^{\cdot-}$, protecting against lipid peroxidation (88), and to prevent the formation of hypochlorous acid (another oxygen species) (89).

On the other hand, however, it should be expected that hypothyroid status, which constitutes a pathological condition, leads, sooner or later, to oxidative damage of biological molecules. In agreement with the last assumption, progressive hypothyroidism, observed early in postnatal rats, enhanced oxidative processes; increased SOD and CAT activities, and decreased GSH level were accompanied by an increased amount of ·OH, enhanced protein carbonylation and lipid peroxidation in rat brain (90).

It is known that in aging humans, the function of several endocrine glands declines progressively, resulting in decreased concentrations of different hormones, among others of melatonin and of thyroid hormones. The study, performed in perimenopausal and menopausal women, with initial low levels of blood melatonin, revealed that melatonin treatment for 3-6 months resulted in a significant increase in thyroid hormone concentrations (91). Thus, melatonin reveals a recovery effect on thyroid function towards a more juvenile pattern of regulation (91); it is not excluded that the described effect is a direct one because a similar treatment with melatonin in aging patients did not result in any changes of TSH concentration (92).

**Radioiodine ($^{131}I$) - induced oxidative stress**

Radioiodine ($^{131}I$) therapy is a treatment of choice in differentiated thyroid cancer (after total thyroidectomy) and in hyperthyroidism. In one of the studies, the parameters of oxidative stress were evaluated in patients subjected to total thyroidectomy because of thyroid carcinoma, before (at that time patients were hypothyroid) and after $^{131}I$ treatment (93). The level of lipid peroxidation products, measured in blood erythrocytes, significantly increased after thyroidectomy, comparing to values in the controls; a further increase in lipid peroxidation was found after $^{131}I$ treatment. Whereas GSH level and GSH-Px and GSH-Rd activities were lower in thyroidectomized patients than those in the control subjects, the values of those parameters significantly increased after $^{131}I$ treatment.

In another study, a treatment with $^{131}I$, using either higher (in case of thyroid cancer) or lower (in case of hyperthyrodism) activities, resulted in increased levels of isoprostanes (products of oxidative injury to lipids) in blood plasma, serum and urine; the damaging effect of $^{131}I$ was significantly higher and longer lasting after higher-activity therapy (94). The above findings are in agreement with other numerous data, indicating that radiation causes oxidative damage to tissues, while antioxidants, like melatonin, may prevent it (8). Because of the potential role of ionizing radiation in the pathogenesis of thyroid cancer, the studies on protective effects of melatonin against radiation-induced oxidative stress and thyroid cancer seem to be of special value. It is worth mentioning here that melatonin effectively reduced histoenzymological changes in the rat thyroid gland caused by exposure to $\gamma$-radiation (95).

Summing up, oxidative stress plays a significant role in the thyroid gland under both physiological and pathological conditions. Melatonin, as an antioxidant and widely distributed molecule, may constitute a modulator of physiological processes in the thyroid, and under pathological

conditions may protect against oxidative damage in the gland and in other tissues. This assumption requires further experimental evidence.

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