Thyroid Hormones and Muscle Bioenergetics

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Abstract

By controlling the protein synthesis, thyroid hormones act on the energetic state in muscle cell. They regulate the DNA expression of myosin, Ca-ATPase, and Na-K pump isoforms, as well as the activity of several key-enzymes of the main energetic pathways (glycolysis and mitochondrial metabolism). A lack or an excess of thyroid hormones therefore induce a new equilibrium between the energetic production and consumption. Hyperthyroidism results in a transition to fast myosin isoforms and in an increased metabolism dependence on glycolysis. A rise in the mitochondrial enzyme activity is theoretically observed *in-vitro*, although the hypercatabolism of protein probably counterbalances this effect *in-vivo*. Thyroid hormone defect results in a transition to slow myosin isoforms and in an inhibited mitochondrial metabolism. This is compensed by an increased glycogen consumption during exercise, despite the observed decreased enzyme activity found *in-vitro* in glycolysis. Hypothyroidism possibly affects the H^+ metabolism. Both hyper and hypothyroidism therefore induce a higher acidosis and a lower muscle performance during exercise, which could explain the exercise intolerance in patients with dysthyroidism.

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The bioenergetic state in cells is dependent on the balance between the energetic production and consumption. To a first approximation, this can be resumed as an equilibrium between synthesis and hydrolysis of ATP, which is considered as the main energetic vector in cell.

The glycolysis pathway partially ensures the ATP production in muscle. However, as in all tissues highly dependent on the energetic metabolism, ATP is essentially produced by the mitochondrial metabolism, which supplies more than 90% of the total energetic availability [3]. The ATP consumption site specific to muscle is the actin-myosin interaction, which induces the fibre contraction. The ATP hydrolysis level during the muscle contraction varies with the activity of the myofibrillar ATPase, which characterizes the fibre type. Moreover, ion exchanges through the muscle membrane are ATP dependent.

The thyroid hormones act on several steps of the energetic production and on the ATPase activity, mainly by controlling the protein synthesis.

1. Control of the energetic production by thyroid hormones

1.1. Energetic pathways

In experimentally thyroid-deprived animals, the activity of several key-enzymes in glycolysis, such as myophosphorylase, fructose 1-6 diphosphatase or lacticodehydrogenase has been found to be reduced *in vitro* in skeletal muscle [11, 28]. Conversely, these enzyme complexes appear to be activated in experimental hyperthyroidism. In fact, these modifications affect especially the type II muscle fibres, or the diaphragm (a mixed muscle), whilst the type I muscles are not or only slightly affected. These abnormalities could partially explain the decreased lactate level found after exercise in serum of hypothyroid subjects and the increased glycogen content observed in muscle biopsy in hypothyroidism [30].

A decreased free fatty acid (FFA) turnover is moreover well documented in hypothyroidism, despite an elevated FFA level in serum [30]. In addition to the frequent finding of lipid droplet accumulation in the muscle biopsy of hypothyroid subjects, this suggests an inhibition of the fat catabolism. Indeed, Baldwin and co-workers [2] have demonstrated a dramatic decrease of ^{14}C-labelled palmitate uptake into muscle homogenates. To our knowledge, similar studies have not been performed in hyperthyroid muscles, but an increased activity of ß hydroxybutyrate

tate uptake into muscle homogenates. To our knowledge, similar studies have not been performed in hyperthyroid muscles, but an increased activity of ß hydroxybutyrate dehydrogenase has been documented in hypothyroid rat muscles [39].

1.2. Mitochondrial metabolism

Several mitochondrial key-enzymes (tricarboxylic Krebs cycle, enzyme complexes of the respiratory chain) are inhibited by experimental thyroid hormone deficiency or activated by experimental thyroid excess [2, 11, 28, 29, 35, 39]. These modifications are especially observed in type I and type IIA muscle fibres, and for a few studies in heart muscle.

The *in-vitro* effects of thyroid administration have been reported in isolated mitochondria: thyroid hormones increase the amino-acid uptake, the synthesis of the mitochondrial inner membrane, the oxygen consumption, the cytochrome concentration and the cytochrome c oxydase activity [27]. Receptors specific to T_3 have been demonstrated on the mitochondrial membrane, suggesting a direct effect of this hormone on the mitochondrial metabolism [33].

Several mechanisms can be put forward to explain the control of the mitochondrial metabolism by the thyroid hormones. Firstly, thyroid hormones modulate the nuclear DNA genes encoding the majority of the mitochondrial proteins, but also the mitochondrial DNA genes encoding several subunits of the respiratory chain enzyme complexes. This provides an explanation for the rise in cytochrome content and in the activity of the cytochrome c oxidase [27]. Secondly, thyroid hormones regulate the substrate incorporation by mitochondria and their availability to their oxidation, implying modifications of the $NADH_2/NAD$ ratio [27]. In addition to the above mentioned effects on the FFA metabolism, it is probable that the thyroid hormones act on the pyruvate dehydrogenase complex, since the incorporation of ^{14}C-labelled pyruvate into muscle homogenates has been found to be strongly suppressed in experimental hypothyroidism [2]. Thyroid hormones also act on the membrane synthesis, by modifying the ratio of unsaturated FFA/ saturated FFA. Since the enzyme complexes of the respiratory chain are located in the mitochondrial inner membrane, thyroid hormones therefore control indirectly their total concentrations, by modifying the functional area of the mitochondrial membrane [27].

All these modifications can be compared with the ultrastructural abnormalities of mitochondria reported in hypothyroid muscle biopsy, such as honey moon aspect, abnormal inclusions or cristae [19]. Conversely, an abnormally increased number of mitochondria has been reported in hyperthyroid heart muscle.

2. Energetic consumption

2.1. Myosin isoforms

Myosin is composed by two heavy and four light chains. Several isoforms have been described and are encoded by a multiple gene family. The expression of this gene family is regulated by nervous and mechanical stimuli, and by hormonal systems [13]. In the heart muscle, three myosin isoforms have been reported; thyroid hormone deficiency induces a transition to the V_3 isoform (α_2 dimere), and a replacement therapy in a thyroid-deficient animal increases the proportion of V_1 isoforms ($ß_2$ dimere) [6].

Similar results have been observed in skeletal muscle, in which at least five different isoforms exist in rodents [36]. Histochemical studies of experimental hypothyroidism have shown an abnormally high proportion of type I fibres in muscle biopsies [12]. Similar results have been reported in hypothyroid man [31, 38]. Moreover, a transition from type I to type IIA fibres has been observed when thyroid hormones were administered in hypothyroid rats [11, 28]. These modifications appear to be caricatural when thyroid hormones are in excess.

Myosin transition is linked to the T_3 control of the genes encoding the SM (slow myosin), IN (intermediate myosin) and FM3 (type 3 fast myosin) isoforms, so that thyroid hormones preferentially affect the myosin distribution of slow type I fibres, when fast type II fibres seem to be only minimally concerned [5, 13]. These modifications are not related to the muscle innervation, since the myosin transition obtained by repetitive stimulations is inhibited in hypo or hyperthyroid animals [29].

This thyroid-related myosin isoform distribution theoretically results in a new energetic equilibrium: myosin isoforms which are present in type I fibres (or the V_3 isoform in heart) express a "slow" ATPase activity and are therefore "economical" in their energetic consumption. Conversely, the isoforms which characterize type II fibres consume more energy and this corresponds to a faster and higher developed twitch tension.

2.2. Sarcoplasmic Ca-ATPase dependent channels

The actin-myosin interaction is controlled by the intracellular level of Ca^{++}, the nerve impulse inducing the Ca^{++} extrusion from the sarcoplasmic reticulum. Moreover, the muscle relaxation is related to the Ca^{++} uptake by the sarcoplasm. This Ca^{++} uptake is faster in type II than type I fibres [4]. Hypothyroid rat soleus (slow type I muscle) contains essentially mRNA of the type I isoform of Ca-ATPase, whilst the mRNA level of the type II isoform is 150-fold greater in the presence of T3. In hyperthyroidism, this last isoform represents 50% of the total mRNA content [32]. Fast type II muscles appear to be less affected [32].

2.3. Na-K-ATPase dependent pumps

The Na-K-ATPase dependent pumps maintain the electrochemical gradient of sodium and potassium across the plasma membrane. They are composed by two subunits, three isoforms of the a subunits being known. The thyroid hormones modulate the gene expression encoding these isoforms [10]. Using labelled ouabain, which will bind sodium pumps, the pump abundance in hypothyroids has been found to be half the euthyroid values and twice in hyperthyroids [20]. In heart as well as in skeletal muscles,

physiological levels of thyroid hormones essentially control the gene expression encoding the α_2 et ß proteins, whilst thyroid hormone excess induces a rise in the α_1 protein [10].

These modifications of the Na-K-ATPase activity with the thyroid hormone level are known to be an important factor of energetic consumption in muscle and their higher (respectively lower) activity in hyperthyroidism (respectively hypothyroidism) is put forward to explain the increased (respectively decreased) thermogenesis [30].

3. In-vivo consequences of dysthyroidism

3.1. Hyperthyroidism

The causes of the cardiovascular disorders induced by thyrotoxicosis are multiple: hemodynamic modifications, sympathetic overactivity or direct alterations of the myocardium by the thyroid hormones. In thyrotoxicosis, the heart is hypertrophied and hypercontractile. The heart hypertrophy is probably due to the haemodynamic modifications. However, the transition in myosin and Ca-ATPase isoforms induced by thyroid excess explains the increased systolic cardiac volume and left ventricular systolic or diastolic functions at rest [26]. Moreover, the cardiac output and the left ventricular systolic volume increase during exercise by the same magnitude in volunteers before and after they received thyroid hormones [23, 24]. Thus, the fact that cardiac performance seems to be not compromised by thyroid excess suggests that the exercise intolerance in hyperthyroidism may be due to noncardiac mechanisms [26], which can be haemodynamic modifications or skeletal muscle alterations [23].

Martin and coworkers have recently demonstrated that the same exercise induced in subjects who voluntarily received supplementary thyroid hormones a rise in the frequency and the cardiac output, whilst the arterio-venous difference in oxygen was reduced and the serum lactate increased [23]. These modifications were attributed to a diminished oxidative capacity of hyperthyroid skeletal muscles. Taking into account the results obtained by *in-vitro* biochemical investigations, this assumption appears to be paradoxical. However, the increase per unit volume in oxidative enzyme activity reported in experimental studies may be counterbalanced by the protein hypercatabolism and subsequently the muscle atrophy documented in hyperthyroid myopathy [38]. In the same way, using phosphorus nuclear magnetic resonance spectroscopy (P.MRS), we have not found in hyperthyroid man an increased phosphocreatine synthesis rate after exercise [18], which was expected by the fact that phosphocreatine synthesis is strongly dependent on the rate of ATP synthesis by the mitochondrial metabolism. However, this result was previously reported in experimental hyperthyroidism by Argov and coworkers [1].

Moreover, we postulate that the exercise intolerance may be related to a higher recruitment of the glycolysis pathway: supporting this hypothesis, hyperthyroid subjects studied by P.MRS exhibited a higher acidosis during exercise than controls [18]. This assumption is coherent with the transition in myosin isoforms observed in thyrotoxicosis and with the increased enzyme activity in this pathway observed in thyrotoxicosis

3.2. Hypothyroidism

By symmetrical reasons as aboveexplained, the transition in myosin and Ca-ATPase isoforms induced by the lack of thyroid hormones can explain a decreased contraction and relaxation rate of muscle fibres. Indeed, hypothyroidism provokes a fall in the left cardiac output and in the left ventricular systolic fraction [9, 37].

P.MRS studies in experimental or human hypothyroidism have demonstrated in skeletal muscle a decreased phosphocreatine resynthesis rate after exercise, related to the functional inhibition of the oxidative phosphorylation [1, 17]. Moreover, measurements of the intracellular pH during exercise have shown a more pronounced acidosis in hypothyroid than in euthyroid muscles [16, 17]. These results contrast with those of biochemical investigations which have demonstrated a glycolysis inhibition *in-vitro*. In fact, according to this glycolysis inhibition, the pH decrease during exercise has been found to be delayed in hypothyroid in comparison with euthyroid muscles [34]. However, the importance of the functional inhibition of the mitochondrial metabolism implies an enhanced glycolysis recruitment in order to compensate the mitochondrial failure so, an increased glycogen consumption during exercise has been found in hypothyroid rats [2], as well as a lactate accumulation in hypothyroid dogs [14], suggesting an alteration of the pyruvate oxidation. In addition, the abnormal acidosis found in hypothyroid muscle is probably related to an abnormal H^+ metabolism, since the pH recovery after exercise appears abnormally slow [16, 34]. The H^+ exchanges across the cell membrane depend on the lactate expulsion and Na^+/H^+ antiports. The lactate transport through the muscle membrane is probably affected by the lack of thyroid hormones, since a decreased lactate level in serum after exercise is well documented, contrasting with the previously mentioned rise in intracellular lactate content [14, 21]. Moreover, the defect in sodium pumps tends to decrease the intracellular Na^+ level, which can interfere with the Na^+/H^+ antiport functioning [34].

4. Muscle efficiency in dysthyroidism

The twitch tension developed during a tetanic stimulation has been found to be similar in hypothyroid and in euthyroid muscles, whilst the ATP turnover appeared reduced [8, 22, 38]. Conversely, hyperthyroid muscles in rodents and humans exhibited a decreased twitch tension with an increased ATP turnover. The laws of thermodynamics therefore imply that in hypothyroidism (respectively in hyperthyroidism), the muscle efficiency is higher (respectively lower) than in euthyroidism [7, 22, 38], and that the calorific production by muscle must be lower (respectively higher). As these results are mainly related to the transition of the myosin isoforms, one can postulate that thermogenesis is dependent on this transition.

However these modifications of the muscle efficiency are not sufficient to improve the muscle performance. The treatment of the thyroid disease in man increases the muscular volume in hyperthyroids and decreases it in hypothyroids, whilst a rise in the muscle strength is observed in both cases [40]: this suggests an improvement of the muscle performance per unit volume, at least in hypothyroidism. In the same way, the energetic cost of an aerobic exercise, as estimated by the depletion of phosphocreatine, is higher in hypo and hyperthyroid than in euthyroid muscle [1, 17, 18]. As in both cases, an abnormal acidosis is demonstrated and implies a deplacement of the dissociation equilibrium of the inorganic phosphate, this provokes an increased monovalent ion $H_2PO_4^-$ [15], which had been implicated in alteration of the actin-myosin coupling [25].

Conclusion

The thyroid hormones modulate the energetic balance in muscle by modifying the energy production and the energy consumption. Theoretically, the transition to slow (respectively fast) myosin isoforms in hypothyroidism (respectively hyperthyroidism), characterized by a gain (respectively a loss) of energy is counterbalanced by a reduction (respectively an enhancement) of the ATP production in the glycolysis and especially the mitochondrial metabolism. Nevertheless, in both hypo and hyperthyroidism, the muscles are less performant, maybe due to a high recruitment of glycolysis, which induces acidosis, by disorders of the ion exchanges through the plasma membrane and by alteration of the actin-myosin coupling during contraction.

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References

[1] Argov Z, Renshaw PF, Boden B, Winokur A, Bank WJ: Effects of thyroid hormones on skeletal muscle bioenergetics: in vivo phosphorus-31 magnetic resonance spectroscopy study of humans and rats. J *Clin Invest* 1988; 81: 1695-1701.

[2] Baldwin KM, Hooker AM, Herrick RE, Schrader LF: Respiratory capacity and glycogen depletion in thyroid-deficient muscle. J *Appl Physiol* 1980; 49 (1): 102-106.

[3] Brand MD, Murphy MP: Control of electron flux through the respiratory chain in mitochondria and cells. *Biol Rev* 1987; 62: 141-193.

[4] Burke RE, Levine DN, Tsairis P, Zajac FE: Physiological types and histochemical profiles in motor units of the cat gastrocnemius. J *Physiol (London)* 1973; 234: 723-748.

[5] Caiozzo VJ, Herrick RE, Baldwin KM: Influence of hyperthyroidism on maximal shortening velocity and mysosin isoform distribution in skeletal muscles. *Am J Physiol* 1991; 261 (30): C285-C295.

[6] Chizzonite RA, Everett A, Clark WA, Jakovcic S, Rabinowitz M, Zac R: Isolation and characterization of two molecular variants of myosin heavy chain from rabbit ventricle. Change in their content during normal growth and after treatment with thyroid hormone. J *Biol Chem* 1982; 257: 2056-2065.

[7] Everts ME, Van Hardeveld C, Ter Keurs HEDJ, Kassenaar AAH: Force development and metabolism in perfused skeletal muscle of euthyroid and hyperthyroid rats. *Horm Metabol Res* 1983; 15: 388-393.

[8] Everts ME, Van Hardeveld C, Ter Keurs HEDJ, Kassenaar AAH: Force development and metabolism in skeletal muscle of euthyroid and hypothyroid rats. *Acta Endocrinol (Copenh)* 1985; 97: 221-225.

[9] Gay RG, Raya TE, Lancaster LD, Lee RW, Morkin E, Goldman S: Effects of thyroid state on venous compliance and left ventricular performance in rats. *Am J Physiol* 1988; 254: 81-88.

[10] Horowitz B, Hensley CB, Quinterio M, Azuma KlC, Putnam D, McDonough AA: Differential regulation of Na,K-ATPase α1, α2, and ß subunit mRNA and protein levels by thyroid hormone. J *Biol Chem* 1990; 265: 14308-14314.

[11] Ianuzzo CD, Chen V, O'Brien P, Keens TG: Effect of experimental dysthyroidism on the enzymatic character of the diaphragm. J *Appl Physiol* 1984; 56 (1): 117-121.

[12] Ianuzzo CD, Patel P, Chen V, O'Brien P, Williams C: Thyroidal trophic influence on skeletal muscle myosin. *Nature* 1977; 270: 74-76.

[13] Izumo S, NadalGinard B, Mahdavi V: All members of MHC multigene family respond to thyroid hormone in a highly tissue-specific manners. *Science* 1986; 231: 597-600.

[14] KaciubaUscilko H, Brzezinska Z, Kruk B, Nazar K: Thyroid hormones deficiency and muscle metabolism during light and heavy exercise in dogs. *Pflügers Arch* 1988; 412: 336-337.

[15] Kaminsky P, Klein M, Duc M: La myopathie hypothyroïdienne. Approche physiopathologique. *Ann Endocrinol* 1992; 53 :125-132.

[16] Kaminsky P, Klein M, Robin-Lherbier B, Walker P, Escanyé JM, Brunotte F, Robert J, Duc M: A 31P NMR study of different hypothyroid states in rat leg muscle. *Am J Physiol* 1991; 261 (24): E706E712.

[17] Kaminsky P, RobinLherbier B, Brunotte F, Escanyé JM, Walker P, Klein M, Robert J, Duc M: Energetic metabolism in hypothyroid skeletal muscle, as studied by phosphorus magnetic resonance spectroscopy. J *Clin Endocrinol Metab* 1992; 74 :124-129.

[18] Kaminsky P, Robin-Lherbier B, Walker P, Brunotte F, Escanyé JM, Klein M, Forrett MC, Robert J, Duc M: Muscle bioenergetic impairment in hyperthyroid man: A study by 31P NMR spectroscopy. *Acta Endocrinol (Copenh)* 1991;124: 271-277.

[19] Khaleeli ALIA, Gohil K, McPhail G, Round JM, Edwards RHT: Muscle morphology and metabolism in hypothyroid myopathy: effects of treatment. J *Clin Path* 1983; 36: 519-526.

[20] Kjeldsen K, Everts ME, Clausen T: The effects of thyroid hormones on ^{3}H-ouabain binding site concentration, Na,K-contents and ^{86}Rb-efflux in rat skeletal muscle. *Pfluegers Arch* 1986; 406: 529-535.

[21] Kruk B, Brzezinska Z, Kaciuba-Uscilko H, Nazar K: Thyroid hormones and muscle metabolism in dogs. *Horm Metab Res* 1988; 20: 620-623.

[22] Leijendekker WJ, Van Hardeveld C, Kassenaar AAH: The influence of the thyroid state on energy turnover during tetanic stimulation in the fast-twitch (mixed type) muscle of rats. *Metab Clin Exp* 1985; 32: 615621.

[23] Martin WHI, Spina RJ, Korte E, Yarasheski KE, Angelopoulos TJ, Nemeth PM, Saffitz JE: Mechanisms of impaired exercise capacity in short duration experimental hyperthyroidism. J *Clin Invest* 1991; 88: 2047-2053.

[24] Massey DG, Becklade MR, McKenzie M, Bates DV: Circulatory and ventilatory response to exercise in thyrotoxicosis. N *Engl J Med* 1967; 276: 1104-1112.

[25] Miller RG, Boska MD, Moussavi RS, Carson PJ, Weiner MW: 31P nuclear magnetic resonance studies of high energy phosphates and pH in human muscle fatigue. J *Clin Invest* 1988; 81: 1190-1196.

[26] Mintz G, Pizzarello R, Klein I: Enhanced left vetricular diastolic function in hyperthyroidism: Noninvasive assessment and response to treatment. J *Clin Endocrinol Metab* 1991; 73: 146-150.

[27] Nelson BD: Biogenesis of mammalian mitochondria. *Curr Top Bioenergetics* 1987; 35: 221-272.

[28] Nicol CJ, Johnson IA: Energy metabolism of fast- and slow-twitch skeletal muscle in the rat: thyroid hormone induced changes. J *Comp Physiol* 1981; 142: 465-472.

[29] Nwoye L, Mommaerts WFHM, Simpson DR, Seraydarian K, Maruisich M: Evidence for a direct action of thyroid hormone in specifying muscle properties. *Am J Physiol* 1982; 242 (11): R401-R408.

[30] Ruff RL: Endocrine myopathies (hyper and hypofunction of adrenal, thyroid, pituitary and parathyroid glands and iatrogenic steroid myopathy). in: Engel AG, Banker BQ (eds): *Myology*. McGraw-Hill Book Co., New York, 1986, pp 1871-1906.

[31] Salviati G, Zeviani M, Betto R, Nacamulli D, Busnardo B: Effects of thyroid hormones on the biochemical specialization of human muscle fibers. *Muscle Nerve* 1985; 8: 363-371.

[32] Simonides WS, Van Der Linden GC, Van Hardeveld C: Thyroid hormone differentially affects mRNA levels of Ca-ATPase isozymes of sarcoplasmic reticulum in fast and slow skeletal muscle. *FEBS Lett* 1990; 274: 73-76.

[33] Sterling K, Lazarus JH, Milch PO, Sakurada T, Brenner MA: Mitochondrial thyroid hormone receptor: localization and physiological significance. *Science* 1978; 201: 1126-1129.

[34] Taylor DJ, Rajagopalan B, Radda GK: Cellular energetics in hypothyroid muscle. *Eur J Clin Invest* 1992; 22: 358-365.

[35] Terjung RL, Koerner JE: Biochemical adaptations in skeletal muscle of trained thyroidectomized rats. *Am J Physiol* 1976; 230: 1194-1197.

[36] Tsika RW, Herrick RE, Baldwin KM: Subunit composition of rodent isomyosins and their distribution in hind-limb skeletal muscles. J *Appl Physiol* 1987; 63: 2101-2110.

[37] Wieshammer S, Keck FS, Waitzinger 1, Kohler J, Adam W, Stauch M, Pfeiffer EF: Left ventricular function at rest and during exercise in acute hypothyroidism. *Br. Heart* J 1988; 60: 204-211.

[38] Wiles CM, Young A, Jones DA, Edwards RHT: Muscle relaxation rate, fibre-type composition and energy turnover in hyper- and hypothyroid patients. *Clin Sci* 1979; 57: 375-384.

[39] Winder WW, Holloszy JO: Response of mitochondria of different types of skeletal muscle to thyrotoxicosis. Am J *Physiol* 1977; 232: C180-C184.

[40] Zürcher RM, Horber FF, Grünig BE, Frey FJ: Effect of thyroid dysfunction on thigh muscle efficiency. J *Clin Endocrinol Metab* 1989; 69: 1082-1086.