Influence of Chronic Exposure to Cold Environment on Thyroid Gland Function in Rabbits

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Key words
- cold
- perfusion
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- thyroxine (T4)
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- radionuclide
- ^{99m}Tc pertechnetate

Abstract

Chronic exposure to cold can affect the thyroid gland. However, the effect on thyroid gland perfusion images and the ratio between thyroid hormones secretion were not addressed in any previous study. The present study investigates the effects of chronic cold exposure on thyroid gland function using radionuclide tracer and thyroid hormones secretion concentration. New Zealand white rabbits weighing approximately 1.8–2 kg were kept in a cold room (4 °C) for 7 weeks. Thyroid scintigraphy was performed for cold exposed rabbits and a control rabbit group. Each rabbit was injected with 115 MBq (3.1 mCi) technetium-99m pertechnetate (99mTc pertechnetate). Studies were performed using Gamma camera equipped with a low energy, high resolution, pinhole collimator interfaced with a computer. Static images were acquired 20 min after administration of the radiotracer. Rabbits chronically exposed to cold had less body weights than control. Thyroid gland uptake is higher in rabbits chronically exposed to cold than controls using radionuclide perfusion study. The increase was proportional to the time period, so the increase after 7 weeks was greater than 5 weeks. There is also an increase in free triiodothyronine (FT3) and a decrease in free thyroxine (FT4) values. Our results indicate that thyroid gland uptake is higher in rabbits chronically exposed to cold than control and the increase was proportional to the duration. The decrease in rabbit body weights may be related to the increase in metabolism due to the increase of thyroid hormones. Chronic cold exposure also increased the conversion of T4 to T3, which is more potent in thermogenic effect.

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Bibliography
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Introduction

Acclimatization to cold is difficult to be demonstrated, due to human's success in avoiding cold stress by behavioral and insulative mechanisms [1]. Man exhibits certain metabolic adaptations during prolonged cold exposure as increased sensitivity to noradrenaline occurred in Australian men in the Antarctic [2–4]. Increased thyroid hormone turnover and elevated serum thyrotropin levels have been proved in normal men during prolonged cold exposure in the Arctic [5–10]. Thyroid gland has many functions due to its hormones. It has calorigenic and thermogenic effect. The major thermogenic effect in mammals is to induce transcript of Na^+/K^+ ATPase gene. It also increases oxygen consumption (used for ATP production). Thyroid gland has intermediary metabolism and mobilizes glucose for use (glycogenolysis to make ATP). Increased thyroid hormone concentration results in a high metabolic rate, an increase in skin temperature and sweating, and a marked sensitivity to heat. In normal subjects, the thyroid gland secretes about 110 nmol of T4 and 10 nmol of T3 each day. T3 has a greater affinity and greater efficacy than T4 for the nuclear receptor, thus although T4 is quantitatively secreted at much higher levels, to become biologically active, it requires deiodination and conversion to T3. Therefore, the present study investigates the effects of chronic cold exposure on thyroid gland function using radionuclide tracer and also the concentration of thyroid hormones secretion during this low temperature.

Materials and Methods

Twelve adult male New Zealand white rabbits weighing approximately 1.8–2 kg were used in these experiments. They were divided into 2 groups, 6 rabbits per each (1 rabbit per cage). The first group (group 1), were kept at room temperature at 25 °C and served as controls. While the second group (group 2), age-matched littermates rabbits were kept in the cold room at 4 °C for a period of 7 weeks. Rabbits in both groups were fed with the same amount of food and had access to water throughout the duration of the study. The study was approved by the institutional research committee. Rabbits in both groups, control group and chronically exposed to cold group, were weighed at the beginning of the experiments, and then each following week for a period of 7 weeks.

Radionuclide imaging

At the end of 5 weeks and 7 weeks, thyroid scintigraphy was performed for control rabbit group and for the chronically exposed to cold group. Each radionuclide study was performed using 115 MBq (3.1 mCi) 99mTc-pertechnetate. Studies were done using Gamma camera (Meridian System, T55B- 1473) equipped with a low energy, high resolution, pinhole collimator with standardized distance to the rabbit, interfaced with a dedicated computer. Rabbits were positioned after anesthesia in the supine position. Static images were acquired 20 min after administration of the radiotracer on a matrix of 256 × 256.

Thyroid function tests

Blood samples were taken from the marginal veins of rabbit ears for control group and chronically exposed to cold group (each group, n = 6). The blood was collected in syringes, transferred into heparinized tubes, and centrifuged at 3 000 rpm for 10 min. The plasma was separated and kept at −20 °C. Plasma FT4, FT3, and TSH levels were measured for the 2 groups. The measurements were made by radioimmunoassay (Immunotech and DPC kits) using Stratec SR 300 instrument.

Statistical analysis

Data are presented as mean ± SEM of number of rabbits used (n) in experiments. Differences between mean values were compared using one-way ANOVA followed by post hoc analysis and the difference was assumed to be significant when $p < 0.05$.

Results

Effect of cold exposure on rabbit's weight

The weight of the rabbits chronically exposed to cold group showed a steady loss of weight than their age-matched littermates rabbits kept at room temperature at 25 °C (control group), for the period of 7 weeks, as shown in **Fig. 1**. The cold exposure effect was observed after 1 week, and then the increase is similar in both the groups.

Radionuclide imaging

99mTc-pertechnetate is an agent which, after administration, is transported by the blood to the thyroid gland by a transporter and is distributed proportionally to regional tissue blood flow. 99mTc-pertechnetate is trapped by the thyroid gland. However, unlike iodine, it does not undergo organification and remains in the thyroid for a relatively short period. Therefore, scintigraphy is best done about 15–30 min after administration of the radiopharmaceutical reagent. The uptake of pertechnetate is the basis for 99mTc-pertechnetate scintigraphy, which has the ability as a substance to be retained within the thyroid gland. The uptake of 99mTc-pertechnetate by the thyroid gland in the cold exposed rabbits was increased compared to the control group. This increase was significant between the 2 groups (n = 6, $^{*}p < 0.05$). The increase was proportional to the duration of cold exposure, so the increase after 7 weeks was greater than 5 weeks, as shown in **Fig. 2, 3**.

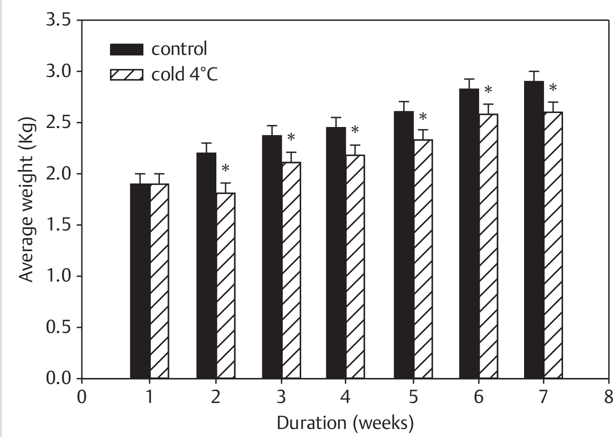

Fig. 1 Effect of chronic exposure to cold (4 °C) on the weight of rabbits, the mean ± S.E. of 6 experiments; $^{*}p < 0.05$.

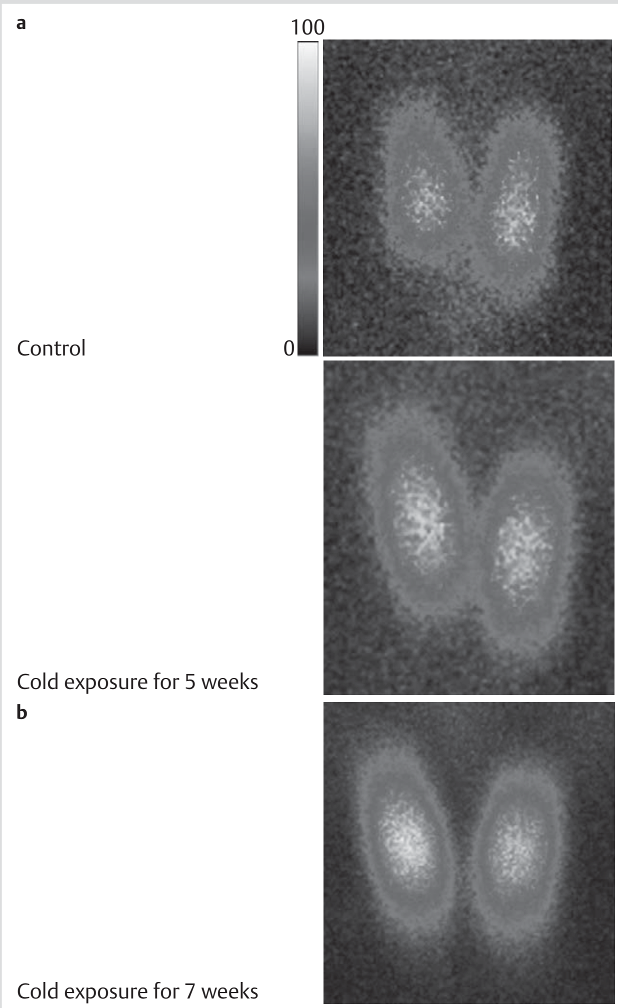

Fig. 2 Images of rabbit thyroid gland perfusion views before and after chronic exposure to cold (4 °C). Control: At room temperature; **a** Exposed to cold for 5 weeks; **b** Exposed to cold for 7 weeks.

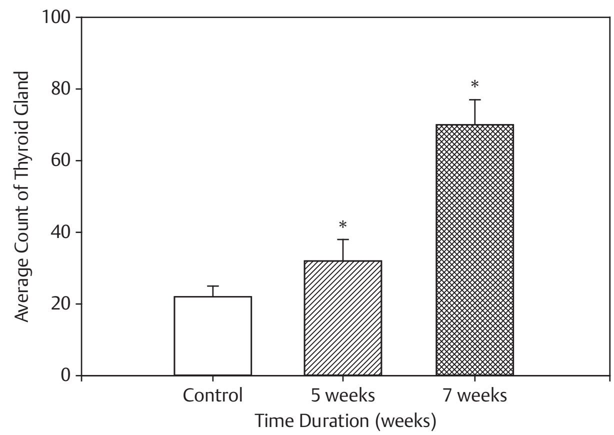

Fig. 3 Total counts for rabbit thyroid gland before and after chronic cold exposure (4 °C). Results are mean ± SEM of 6 experiments; *p < 0.05.

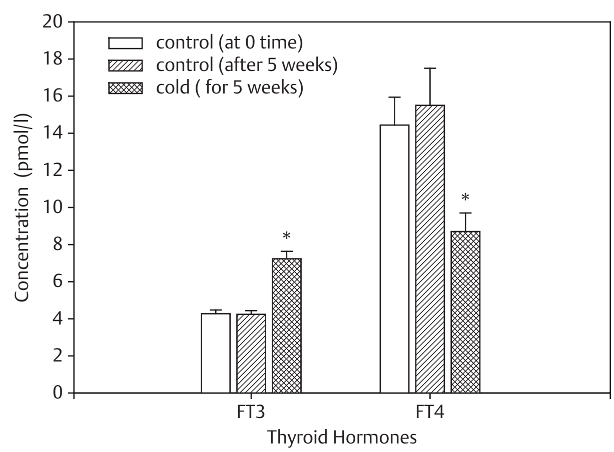

Fig. 4 Effect of chronic exposure to cold (4 °C) on the concentration of the 2 thyroid hormones FT3 and FT4 of rabbits. Notice the significant increase of FT3 than its control and the significant decrease of FT4 than its control, the mean ± S.E. of 6 experiments; *p < 0.05.

Thyroid function tests

There were changes in the values of both the FT4 and FT3 levels in rabbits chronically exposed to cold compared to controls (▶ **Fig. 4**). The FT4 value decreased while the FT3 value was increased. The high value of FT3 is indicating an increase in the uptake due to chronic exposure to cold environment. Plasma TSH levels were also increased after chronic cold exposure from ±0.01 to ±0.05 μm/l, (n = 6, *p < 0.05).

Discussion

▼

A thyroid scan can provide information about the size and shape of the gland as well as the activity of the gland, either it is overactive or underactive in manufacturing thyroid hormone. Prolonged exposure to cold is known to encourage the hypothalamus to produce thyrotropin-releasing hormone (TRH). TRH is modulating the production of thyroid stimulating hormone (TSH). Therefore changes in environmental temperature may cause alterations in TSH secretion and thyroid hormones [11, 12]. It is known that in newborn, high secretion of TSH occurs in the first few hours after birth, accompanied by an increase in thyroid hormone secretion [13, 14]. The rise in TSH secretion could be due to the postnatal cooling, accordingly they have to be maintained in a warm environment. Increases in serum TSH levels have also been observed in infants and young children during surgical hypothermia [15]. Some studies have shown prolonged arctic residence leads the increase in TSH to be associated with an increase in thyroglobulin and T3 [16]. In addition, cold exposure in animals leads to enhanced thyroid hormonal secretion, degradation, and excretion. It is associated with augmented rates of T4 and T3 deiodination, increased conversion of T4 to T3, and enhanced hepatic binding and biliary and fecal clearance of the iodothyronines [17–25]. All of these effects are presumably due to an increased need for thyroid hormones by peripheral tissues, since the calorigenic action is important as part of the response to a cold environment [26].

About 93 % of the metabolically active hormones secreted by the thyroid gland is T4 and 7 % is T3, however almost all T4 is eventually converted to T3 in the tissues, so that both are functionally important. Conversion of T4 to T3 was observed in tumor tissues studied and was markedly enhanced in Graves' thyroid tissues compared to that of normal thyroid tissues. It is concluded that T4 is enzymatically converted to T3 in normal and Graves' thyroids and differentiated thyroid neoplasm [27, 28]. Most antithyroid drugs decrease hormone synthesis and/or conversion of T4 to T3. Most of the circulating thyroid hormones are bound to plasma proteins, the free fraction comprising 0.03 % of T4 and 0.3 % of T3. Only the free hormone has metabolic effects, and measurement of free hormone generally is more reliable than measurement of the total circulating hormone, since the latter varies with levels of binding proteins [29]. Therefore in this study, we measured the free fraction of the 2 hormones (FT3, FT4).

The thyroid hormones regulates metabolism in humans and animals. Increased metabolism rate could lead to body weight loss. Our findings suggest that prolonged cold exposure is a potent stimulus to increase circulating thyroid hormone levels, which is the reason for the difference between the animal body weights at the end of these experiments. The body weight of the rabbit group chronically exposed to cold was steadily less than the control group kept at room temperature (at 25 °C), for the period of 7 weeks as shown in ▶ **Fig. 1**.

The thyroid gland has a rich blood flow, which equals to about 5 times the weight of the gland each minute. This will make the measurement for blood perfusion very valuable and important to know the effect of cooling on this gland, as we did in this investigation. Increased iodine uptake by the whole gland is the indication of overactivity. The uptake of pertechnetate is the basis for ^{99m}Tc-pertechnetate scintigraphy, which has the ability as a substance to be retained within the thyroid gland, but unlike iodine, it does not undergo organification. In the present study, the uptake of ^{99m}Tc-pertechnetate was observed in the thyroid gland of control rabbits. The uptake of ^{99m}Tc-pertechnetate by the thyroid gland in the cold exposed rabbits had significantly increased after 5 weeks as well as after 7 weeks, as shown in ▶ **Fig. 2, 3**. This is a clear evidence that exposure to chronic cold proportionally increases the perfusion to thyroid gland.

The present investigations show the different responses on the thyroid hormones level due to cold. The high value of FT3 indicates an increase in blood perfusion to the gland due to chronic exposure to cold environment. Plasma TSH levels were also increased after chronic cold exposure. Therefore, it is clear that the conversion of T4 into T3 is enhanced in several situations, and the result of our study is the first to supply data on the chronic exposure of thyroid gland to cold in such conditions.

Conflict of Interest

The authors declare that they have no conflicts of interest in the authorship or publication of this contribution.

References

1 *Edholm OG*, *Lewis HE*. Terrestrial animals in cold: man in polar regions. In: Dill DB, Adolph EF, Wilber CG (ed.). Handbook of Physiology, Section 4: Adaptation to the Environment. Washington: American Physiological Society, 1964; 435–446
2 *Budd GM*, *Warhaft N*. Cardiovascular and metabolic responses to noradrenaline in man, before and after acclimatization to cold in Antarctica. J Physiol 1966; 186: 233–242
3 *Budd GM*, *Warhaft N*. Body temperature, shivering, blood pressure and heart rate during a standard cold stress in Australia and Antarctica. J Physiol 1966; 186: 216–232
4 *Budd GM*. Skin temperature, thermal comfort, sweating, Clothing and activity of men sledging in Antarctica. J Physiol 1966; 186: 201–215
5 *Andersen S*, *Kleinschmidt K*, *Hvingel B*, *Laurberg P*. Thyroid hyperactivity with high thyroglobulin in serum despite sufficient iodine intake in chronic cold adaptation in an Arctic Inuit hunter population. Eur J Endocrinol 2012; 166: 433–440
6 *Ingbar SH*, *Bass DE*. The effect of prolonged exposure to cold on production and degradation of thyroid hormone in man. J Endocrinol 1967; 37: II–III
7 *Rudas P*, *Pethes G*. Acute changes of the conversion of thyroxine to triiodothyronine in hypophysectomized and thyroidectomized chickens exposed to mild cold (10 degrees). Gen Comp Endocrinol 1986; 63: 408–413
8 *Raud HR*, *Odell WD*. The radioimmunoassay of human thyrotropin. Br J Hosp Med 1969; 2: 1366–1376
9 *Eastman CJ*, *Ekins RP*, *Leith IM*, *Williams ES*. Thyroid hormone responses to prolonged cold exposure in man. J Physiol 1974; 241: 175–181
10 *Eastman A*. Augmentation of the peripheral metabolism of L-triiodothyronine and L-thyroxine after acclimation to cold. J Clinc Invest 1974; 53: 980–987
11 *Hefco E*, *Krulich L*, *Illner P*, *Larsen PR*. Effect of acute exposure to cold on the activity of the hypothalamic-pituitary-thyroid system. Endocrinology 1975; 97: 1185–1195
12 *Levine M*, *Duffy L*, *Moore DC*, *Matej LA*. Acclimation of a non-indigenous sub-Arctic population: seasonal variation in thyroid function in interior Alaska. Comp Biochem Physiol A Physiol 1995; 111: 209–214
13 *Fisher DA*, *Odell WD*. Acute release of thyrotropin in the newborn. J Clin Invest 1969; 48: 1670
14 *Fisher DA*, *Oddie TH*. Neonatal thyroidal hyperactivity. Response to cooling. Am J Dis Child 1964; 107: 574–581
15 *Wilber JF*, *Baum D*. Elevation of plasma TSH during surgical hypothermia. J Clin Endocrinol Metab 1970; 31: 372–375
16 *Van Do N*, *Mino L*, *Merriam G*, *LeMAr H*, *Case HS*, *Palinkas LA*, *Reedy K*, *Reed HL*. Elevation in serum thyroglobulin during prolonged Antarctic residence: Effect of thyroxine supplement in the polar 3,5,3′-triidothyronine syndrome. J Clin Endocrinol Metab 2004; 89: 1529–1533
17 *Laurberg P*, *Andersen S*, *Karmisholt J*. Cold adaptation and thyroid hormone metabolism. Horm Metab Res 2005; 37: 545–549
18 *Dunn JT*, *Dunn AD*. Update on intrathyroidal iodine metabolism. Thyroid 2001; 11: 407–414
19 *Dentice M*, *Salvatore D*. Deiodinases. the balance of thyroid hormone: local impact of thyroid hormone inactivation. J Endocrinol 2011; 209: 273–282
20 *Williams GR*, *Bassett JH*. Deiodinases: the balance of thyroid hormone: local control of thyroid hormone action: role of type 2 deiodinase. J Endocrinol 2011; 209: 261–272
21 *Balsam A*, *Sexton FC*. Increased metabolism of iodothyronines in the rat after short-term cold adaptation. Endocrinology 1975; 97: 385
22 *Bernal J*, *Escobar del Rey F*. Effect of the exposure to cold on the extrathyroidal conversion of L-thyroxine to triiodo-L-thyronine, and on intramitochondrial glycerophosphate dehydrogenase activity in thyroidectomized rats on L-thyroxine. Acta Endocrinol 1975; 78: 481
23 *Tsukahara F*, *Uchida Y*, *Ohba K*, *Nomoto T*, *Muraki T*. Defective stimulation of thyroxine 5′-deiodinase activity by cold exposure and norepinephrine in brown adipose tissue of monosodium glutamate-obese mice. Horm Metab Res 1997; 29: 496–500
24 *Balsam A*, *Leppo LE*. Augmentation of the peripheral metabolism of L-triiodothyronine and Lthyroxine after acclimation to cold. Multifocal stimulation of the binding of iodothyronines by tissues. J Clin Invest 1974; 53: 980
25 *Galton VA*, *Nisula BC*. Thyroxine metabolism and thyroid function in the cold-adapted rat. Endocrinology 1969; 85: 79–86
26 *Silvestri E*, *Schiavo L*, *Lombardi A*, *Goglia F*. Thyroid hormones as molecular determinants of thermogenesis. Acta Physiol Scand 2005; 184: 265–283
27 *Ishii H*, *Inada M*, *Tanaka K*, *Mashio Y*, *Naito K*, *Nishikawa M*, *Imura H*. Triiodothyronine generation from thyroxine in human thyroid: enhanced conversion in Graves' thyroid tissue. J Clin Endocrinol Metab 1981; 52: 1211–1217
28 *Salvatore D*, *Tu H*, *Harney JW*, *Larsen PR*. Type 2 iodothyronine deiodinase is highly expressed in human thyroid. J Clin Invest 1996; 98: 962–968
29 *Bartalena L*, *Bogazzi F*, *Brogioni S*, *Burelli A*, *Scarcello G*, *Martino E*. Measurement of serum free thyroid hormone concentrations. an essential tool for the diagnosis of thyroid dysfunction. Horm Res 1996; 45: 142–147