Reciprocal changes in serum free thyroxine fraction and thyrotropin level after administration of thiocyanate to rats

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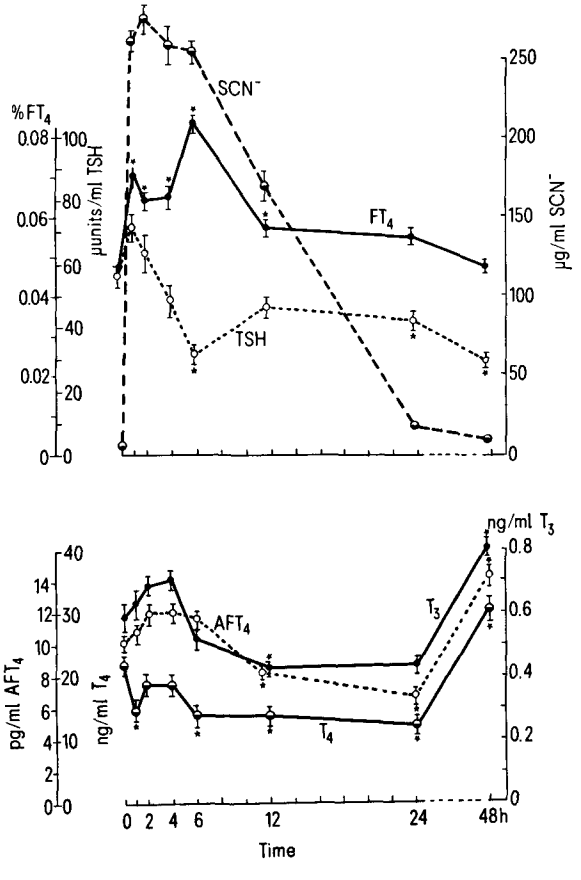
Summary. The administration of thiocyanate to rats caused a significant increase of serum free thyroxine fraction, which coincided with the significant decrease of TSH level. The other components (AFT_4, T_4, T_3) in serum at this time were decreased or unchanged. The finding suggests the role of free thyroxine fraction in feed-back regulation of TSH secretion.

The free thyroxine fraction (FT_4) can penetrate into the tissues and represents the active pool of circulating hormone, whereas the remaining protein bound thyroxine serves as a metabolically inert reservoir[2]. Recent papers raised the importance of FT_4 by the distribution and transcapillary movement of hormone[3-6]. Especially significant is some evidence on the role of FT_4 in the feed-back regulation of the pituitary thyroid axis[7-9]. As we have recently shown, the administration of thiocyanate (SCN^-) to rats caused a significant increase of serum FT_4[10]. In this study, the time course changes of serum FT_4 and TSH level, as well as of total thyroxine (T_4) and triiodothyronine (T_3) in serum after similar SCN^- administration to rats were observed. The findings confirmed the role of FT_4 in feedback regulation of TSH secretion.



Time course changes of free thyroxine fraction (FT_4) and levels of TSH, T_4, T_3, AFT_4 and SCN^- in serum after administration of thiocyanate to the groups of rats. Mean ± SE. Upper part: ◒—◒ concentration of SCN^-; ●—● percentual free thyroxine fraction (FT_4); ○····○ TSH level. Lower part: ○····○ AFT_4; ◒—◒ T_4; ●—● T_3. *Significant changes from the control (values see in text).

Material and methods. 8 groups of male Wistar rats (SPF colony) weighing about 250 g and fed standard laboratory diet were used (8 animals in each group). The groups of rats were force-fed with 3 ml of KSCN solution in amount of 12 mg SCN^- per 100 g of b.wt. The control group received 3 ml of water. The individual groups of animals were exsanguinated and sacrificed 1, 2, 4, 6, 12, 24 and 48 h after SCN^- administration.

The estimation of FT_4 fraction in serum was carried out according to Sterling and Brenner equilibrium dialysis method[11] *in our modification using 1.5 ml of serum*[12]. The serum TSH[1], T_4 and T_3[1] levels were estimated by radioimmunoassay (T_4 antibody was raised in this institute). The radioactive ^{125}I-T_4 and ^{125}I-T_3 were obtained from Institute of Nuclear Research, Swierk near Otwock, Poland. The absolute level of free thyroxine (AFT_4) was calculated as the product of serum T_4 and FT_4 fraction. The level of serum thiocyanate was estimated according to our modification of Aldridge's photocolorimetric method[13,14].

Results and discussion. The time course changes of all of these parameters after the administration of SCN^- to the rats are presented in the figure. The level of SCN^- in serum after its administration rapidly increased more than 50fold with a maximum at 2 h, then decreased slowly and reached the control values after 48 h. FT_4 (percentual free thyroxine fraction) increased significantly ($p < 0.001$), but with a retardation to the SCN^- level, reached maximum at the 6th h, at the 12th h remained significantly increased ($p < 0.01$), then decreased to the control value at the 48th h. The level of TSH showed

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mainly changes reciprocal to these of FT_4. At the beginning, up to the 4th h, the changes were not significant, but at 6 h (maximal FT_4) the TSH level decreased significantly ($p < 0.001$) to 53% of control value, then slowly, but significantly ($p < 0.01$) increased to 80% at 12–24 h and again significantly decreased ($p < 0.05$) after 48 h to 50% of control (the change to the contol value $p < 0.001$). The level of serum total thyroxine was from the beginning of the experiment mainly significantly ($p < 0.001$ at 1, 6, 12, 24 h) decreased to 60% of control, but at 48 h it rapidly increased up to 135% ($p < 0.001$). The levels of T_3 and of AFT_4 from the beginning to the 6th h showed insignificant increase or decrease from the control values, at 12–24 h they were significantly ($p < 0.05$) decreased, but at 48 h significantly ($p < 0.01$) increased.

The maximal changes of TSH and FT_4 temporally coincide at 6 h after SCN^- administration. The levels of other parameters at this time were significantly decreased (T_4) or insignificantly changed (T_3, AFT_4). This suggests that in this period of experiment only the elevated FT_4 was responsible for the inhibition of TSH secretion. In the next period (12–24 h), TSH level increased, but it still did not attain the control level. This increase of TSH could follow from the low concentration of T_4, T_3 and AFT_4, but most probably from rapid decrease of FT_4, which remained, however, increased in comparison with controls. Persistent lower level of TSH could be ascribed particularly to this increased level of FT_4. Reciprocal changes of FT_4 and TSH levels during 24 h are in accordance with the similar changes in the man[7,8]. Moreover, some authors[15,16] also found that in contrast to the inconsistent relationship of AFT_4 and AFT_3 to TSH levels in patients, drug-induced increases in FT_4 and FT_3 appeared consistently to be related temporally to decreases in serum TSH values[16], or to the reduced peak elevation of TSH after TRH[15]. All these findings support the idea that FT_4 may be the important factor regulating the feedback mechanism of the TSH secretion.

48 h after SCN^- administration FT_4 attained control level, while T_4, T_3 and AFT_4 increased vehemently. This unexpected rise of thyroid hormone level could be explained by the increase of hormone biosynthesis (after withdrawal of antithyroid effect of SCN^-) and also by intake of thyroid hormone from intracellular to intravasal space after restoration of binding of protein carriers in blood. Significant decrease of TSH level at 48 h suggests that the other components of circulating thyroid hormones also participate on the feedback regulation of TSH secretion.

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Ovarian serotonin content in relation to ovulation[1]

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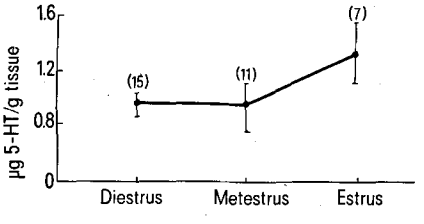
Summary. In the mature cyclic female rat, analysis for ovarian serotonin content reveals comparatively high serotonin content. Fluctuation of serotonin content was observed; peak for serotonin was observed at estrus. In gonadotropin-treated immature rats, there was no detected ovarian serotonin using this procedure. It was concluded that ovaries from gonadotropin-treated immature rats are physiologically different from ovaries taken from mature cyclic rats.

More of the work in the past in relation to serotonin (5-hydroxytryptamine, 5-HT) has focused primarily on the neuroendocrine control of the anterior pituitary function in controlling ovulation. It has been assumed that alterations in the metabolism of amines in discrete parts of the brain influence the secretion of gonadotropines and hence the process of ovulation[4].

It was reported that ovulation can be inhibited in mature adult rats using antagonists to 5-HT such as cyproheptadine, mianserin, and methysergide or LSD[5]. It was also observed physiological and pharmacological differences between the spontaneous ovulating mature rats and the pregnant mare serum gonadotropin (PMS)-induced ovulating immature rats. The present investigation was designed to relate ovarian 5-HT level to ovulation in spontaneously ovulating mature rats and PMS-induced ovulating immature rats.



Levels of ovarian serotonin content in the mature cyclic spontaneously ovulating rats. Each point represents the serotonin content of ovaries ± SE of the mean. Number of animals used to determine each point is indicated in parentheses.

Methods. In the 1st experiment, mature cyclic female Sprague-Dawley rats were used. They were maintained under controlled light (14 h light: 10 h dark) and temperature (23 °C ± 1 °C) in an environmental chamber. Food and water were provided ad libitum. Vaginal smears were taken daily with the establishment of 2 consecutive regular estrus cycles. Animals were sacrificed by over-exposure to ether vapor at stages of estrus, metestrus, and diestrus. The ovaries were removed immediately, dissected free from the bursa and extraneous tissues, weighed and analyzed.

In the 2nd experiment, 56 immature female Sprague-Dawley rats were used. They were maintained under the same experimental conditions as in the 1st experiment.

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