Exploring the use of recombinant human thyrotropin in the diagnosis of central hypothyroidism

Abbreviated title: rhTSH in diagnosis of central hypothyroidism

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Abstract

Context: The diagnosis of central hypothyroidism (CH) is often difficult to establish as serum TSH levels may be low, normal or slightly increased.

Objective: To explore the use of recombinant human TSH (rhTSH) in the diagnosis of CH.

Design: Randomised singel-blind clinical trial.

Setting: Out-patient clinic of a tertiary care referral center.

Intervention: A single intramuscular injection of 0.1 mg and 0.9 mg rhTSH in random order with one-week’s interval.

Participants: Eighteen adult patients with pituitary insufficiency and six healthy age-, sex- and BMI-matched controls. Six patients had untreated CH (newCH), six had treated CH (CH) and six patients were TSH-sufficient (nonCH). Five weeks before TSH stimulation, levothyroxine was replaced with triiodothyronine for four weeks. One week before stimulation treatment was withdrawn.

Main outcome measures: Thyroid hormones and thyroglobulin before and 2, 3½, 7, 24, 48 and 72 hours after each injection.

Results: In the newCH group, basal FT4 levels were lower than in controls (p<0.05). After 0.9 mg rhTSH, the increases in FT4 and rT3 were less marked in the newCH group than in controls (FT4±SEM 9.2±0.5 to 19.7±1.2 vs 11.3±0.5 to 27.8±2.4 pmol/L, p<0.05). The CH group exhibited reduced basal and stimulated FT4 compared with the TSH-sufficient groups. Thyroglobulin increased similarly among all study-groups after rhTSH injection.

Conclusion: In this pilot study patients with untreated CH had lower response to 0.9 mg rhTSH in FT4 and rT3 than controls. An rhTSH test may be useful in the diagnosis of CH, but further studies are required.
Introduction

Central hypothyroidism (CH) occurs due to insufficient synthesis and secretion of biologically active thyroid stimulating hormone (TSH). It may present as part of a general hypopituitarism or as consequence of a limited pituitary damage. The diagnosis of CH may often be difficult as serum TSH levels can be both low, normal or slightly increased (1). The bioactivity of TSH (2) is also reduced because of an inadequate hypothalamic stimulation that causes the pituitary to secrete an abnormally glycosylated TSH. TSH in this form has a longer half-life than normal TSH (3), which explains the normal and sometimes slightly elevated levels of TSH seen in CH. In addition, thyroid hormone levels in mild hypothyroidism may be within the lower normal range (4-7). Because of this uncertainty of using basal thyroid hormone levels in the evaluation of CH, other tests have been developed. Patients with CH have a blunted nocturnal surge (8, 9) in the TSH circadian secretion (10-12). This may, however, be found in non-thyroidal illness (13), in postoperative patients (14, 15), during starvation (16), and in severe primary hypothyroidism (17). The thyrotropin releasing hormone stimulation test has been used in the diagnosis of CH (18, 19), but its value has been questioned (20). However, authors to a recent study of children with congenital CH claim the value of the TRH-test to differentiate between isolated CH and CH combined with multiple hormonal insufficiencies (21). Hence, the diagnosis of CH may be difficult and an additional test to clarify the diagnosis is warranted.

In some early studies, bovine TSH (bTSH) stimulation was explored for the diagnosis of CH (22). In 1949, an increased iodine uptake in the thyroid was detected in normal subjects after bTSH administration that was sustained if the subjects were treated with thyroid hormone during the stimulation (23). It was later established that an inactive gland in CH can be stimulated to resume thyroid hormone synthesis after numerous bTSH injections (22). However, bTSH usage was terminated due to commonly occurring allergic reactions (24) and the appearance of neutralising and hemagglutinating antibodies (24, 25).

Through its receptor, TSH regulates the expression of all gene products required for thyroid hormone synthesis (26-28). TSH receptor knockout mice are, however, able to produce and store thyroglobulin
that has a low iodine and hormone content (29). In addition, animal studies demonstrate that thyroglobulin synthesis and secretion into the follicular lumen continues in the absence of TSH (29, 30). Moreover, subsequent to the elimination of endogenous TSH, the acute endocytic response to TSH is gradually diminished due to the reduction of membrane material available for the formation of endocytotic vesicles (31). Therefore, a different response to TSH in a TSH depleted thyroid gland than under normal conditions could be expected, which is in analogy with the short ACTH stimulation test that is proven to be valid in the diagnosis of ACTH-deficiency (32).

The primary aim of the present pilot study was to investigate whether the stimulation of the thyroid gland with recombinant human TSH (rhTSH) could distinguish between patients with CH and those who are TSH-sufficient. The second aim was to investigate the physiology of a TSH depleted thyroid gland.

**Subjects and Methods**

**Subjects**

Eighteen Caucasian patients with well-defined pituitary disease and pituitary insufficiency were recruited from our endocrine clinic. These patients comprised of three groups: the CH group (n=6) was treated with levothyroxine (Levaxin®, Nycomed AB, Stockholm, Sweden), the newCH group (n=6) had a newly diagnosed CH not yet replaced, and the nonCH group (n=6) had hypopituitarism but unaffected TSH secretion, which was reflected by normal pre-study free thyroxine (FT4) levels (range 11-14 pmol/L). The consecutively recruited newCH patients had an established pituitary disease, a FT4 below the normal range (pre-study mean±SEM; (range): FT4: 7.8±0.9 pmol/L (6.6-8.9), TSH: 0.74±0.62 mU/L (0.02-1.9)) and additional pituitary insufficiencies. Six healthy Caucasian controls were also included. As one control was subnormal in FT4, he was replaced with another individual. The groups were matched for age, sex and body mass index (BMI) (Table 1). The exclusion criteria were: current thyroid disease, presence of thyroperoxidase antibodies (TPOab), cardiac disease and treatment with antiepileptic, antipsychotic or anticoagulation drugs.
**Ethics**
Subjects received oral and written information about the study and were included after written informed consent. The study protocol was approved by the Ethics Committee at Göteborg University and the Swedish Medical Products Agency, Uppsala, Sweden. The study was performed according to the Declaration of Helsinki.

**Study design**
This was a prospective, randomized single-blinded trial using two doses of rhTSH. Before the start of the study, all subjects underwent a routine clinical investigation, including an electrocardiographic registration. In the CH group, the levothyroxine substitution was changed to 20 µg triiodothyronine (Liothyronin®, Nycomed AB, Sweden) thrice daily five weeks before study-start because of its shorter half-life. If symptoms indicating over-replacement appeared, the dose was reduced by half. Triiodothyronine substitution was discontinued one week before commencement of the study and levothyroxine substitution was re-instituted after study-completion.

All participants received an intramuscular gluteal injection at 9 am of 0.1 and 0.9 mg rhTSH (Thyrogen®, Genzyme, Boston, USA) given in random order with one week in-between. Subjects were randomised in blocks by the hospital pharmacy. Before each injection, blood samples were taken for the assessment of hemoglobin, serum sodium, potassium, calcium, creatinine, alanine aminotransferase, aspartate aminotransferase, and alkaline phosphatase. Samples were collected at -45 min, immediately before and 2, 3½, 7, 24, 48 and 72 hours after each injection and a mean was calculated of the two baseline measurements. Side-effects were recorded simultaneously. The newCH patients and the replaced control were recruited after the completion of the other study groups. Based on previous experience, less frequent blood sampling was performed in the newCH group: before, and 24, 48 and 72 hours after each injection.

After study termination, samples were analysed for TSH, FT4, total thyroxine (TT4), free triiodothyronine (FT3), total triiodothyronine (TT3), reverse triiodothyronine (rT3), thyroglobulin (Tg) and Tg antibodies. In addition, IGF-I and insulin levels were measured before any rhTSH injection.
**Hormonal assays**
Immuno-chemoluminometric methods (Architect, Abbot, USA) were used for the analyses of TSH (interindividual coefficient of variation (CV) 3%, reference range (refr) 0.20-4.0 mIU/L), FT4 (CV 6% at low-, 5% at high-levels, refr 10-22 pmol/L), TT4 (CV 5%, refr 56-147 nmol/L), FT3 (CV 9% at low-, 4% at high-levels, refr 2.6-5.7 pmol/L) and TT3 (CV 7% at low-, 3% at high-levels, refr 0.9-2.4 nmol/L). RT3 was determined by a radioimmunometric assay (Wallac Adaltis, Bologna, Italy, CV 5%, refr 0.14-0.54 nmol/L). Tg was analysed by an immunoflourimetric method (Delphia, Wallac Sweden AB, Finland, CV 3% at low-, 8% at medium-, 4% at high-levels, refr 2-20 µg/L). Tg antibodies (Lumitest, Brahms, Henningsdorf, Germany, CV 9%, refr <60 U/ml), and TPO antibodies (Berilux 400, Perkin Elmer BRAHMS Diagnostica, Berlin, Germany, CV 11%, refr <60 kU/L) were measured by immunoluminescence technique. IGF-I was determined with a radioimmunoassay after acid ethanolic extraction (Nicols Institute Diagnostics, San Juan Capistrano CA, USA, CV 6.3% at low-, 6.5% at medium-, 7.7% at high-levels) and insulin by a chemoluminometric method (Advia Centaur, produced by Kyowa Medex Co, Japan for Bayer Health Care, Göteborg, Sweden, CV 5% at low-, 6% at medium- and high-levels). Specimens were analysed in the same batch, except for the newCH- and the exchanged control-specimens that were analysed separately but with the same immunoassays, except, in the control, for IGF-I that was determined by Immulite 2500, DPC, USA.

**Statistical methods**
Descriptive data are presented as mean ± standard error of the mean (SEM). The effect before and after rhTSH administration were analysed with paired t-test and comparisons of hormone levels between groups were performed with unpaired t-test. Statistical significance was considered if p<0.05.

**Results**
All subjects completed the study; however, one control was unable to leave specimens twice. One patient on triiodothyronine substitution experienced headache and nausea, and one tiredness; their dose was reduced by half according to the study protocol. Symptoms after the rhTSH administrations were mild and transient. Marked symptoms of hypothyroidism (n=1) and a brief period of chest pain not related to ischemic heart disease (n=1), occurred in the CH group. Tiredness was, otherwise, the...
most frequent symptom (newCH n=1, CH n=5, nonCH n=1) followed by sensation of warmth (newCH n=1, controls n=2), nocturnal perspiration (controls n=2), slight discomfort (controls n=2), palpitation (controls n=1), pain in the calves (controls n=1), dizziness (nonCH n=1), and less nocturia (nonCH n=1).

In subjects randomised to receive 0.9 mg as first dose, the hormonal levels had not completely returned to baseline before the next injection. Baseline was, therefore, characterised as the values before any rhTSH administration and compared to peak levels after each injection. Moreover, the response in thyroid hormones from 0.1 mg rhTSH was less pronounced compared to the 0.9 mg dose response and did not discriminate newCH patients from controls. The emphasis in the result section is therefore on the 0.9 mg rhTSH dose.

**TSH** (Table 2)
Basal levels of TSH did not differ between groups. After rhTSH injection, serum TSH increased similarly in the four groups to a peak of $16.1 \pm 6.0$ mIU/L after 0.1 mg and $>100$ mIU/L after 0.9 mg within the first 24 hours, results of groups combined.

**Thyroglobulin** (Table 2)
Basal Tg-levels were similar in all groups. In CH subjects, basal Tg levels were increased before the second injection, regardless of given dose (data not shown). RhTSH stimulation resulted in a distinct and comparable increase in Tg in all groups, with similar peak levels 48 hours after the 0.9 mg dose (Figure 1A). In the CH group, an extreme outlier (max Tg 600 µg/L) occurred and after the exclusion of this subject, the Tg analyses did not differ between groups (Figure 1A).

**FT4 and TT4** (Table 2)
FT4 values in the nonCH and control groups had similar baseline FT4 levels (range 8.6-14.5 pmol/L and 10-13.0 pmol/L, respectively), whereas FT4 levels in the CH group were low. The lowest FT4 in the nonCH group was a mean of FT4 9.8 and 7.3 pmol/L. FT4 in newCH group (range: 7.8-11.0 pmol/L) was lower than in control subjects but higher than in CH patients. After the administration of 0.9 mg rhTSH, the increase in the newCH group was less pronounced than in controls (Figures 1B, 2A, 3A). A small overlap in the peak FT4 level occurred between the two groups, range: newCH 15-
23 pmol/L, controls 20-36 pmol/L. Over-lapping values was seen in 67% and 50% of newCH patients and controls after 0.9 mg and 0.1 mg rhTSH, respectively. However, the lowest FT4 level in the controls after 0.9 mg rhTSH was from one subject unable to leave specimens on two out of three occasions where FT4 used to peak. If this patient was excluded from the over-lap analysis no overlap existed between the newCH and the control group after 0.9 mg rhTSH. The mean increase of FT4 after the high-dose-stimulation was in the newCH group 10.5 pmol/L (range 7-13.2) and in controls 16.5 pmol/L (range 10.0-23.5, p<0.05). The FT4 concentration peaked after 48 or 72 hours (n=10, n=2, respectively). A much smaller increase in FT4 was observed in the CH group and no over-lap was detected with the TSH sufficient subjects (Figure 2A, 3A). In the CH group, however, basal levels of FT4 were higher before the second than before the first injection of rhTSH, regardless of the dose administrated (data not shown). Serum TT4 concentrations displayed a similar pattern as FT4.

**FT3 and TT3** (Table 2)
Baseline serum levels of FT3 and TT3 were similar in the nonCH-patients and controls, whereas reduced levels were observed in the CH group. In the newCH group, FT3 levels were lower than in controls, but higher than in the CH group. After 0.9 mg rhTSH, a marked increase in FT3 and TT3 was seen in newCH-, nonCH-patients and controls (Figure 2B, 3B). However, there was a considerable over-lap comparing the rhTSH-response of the newCH group and the TSH sufficient patients.

**RT3** (Table 2)
NewCH-, nonCH-groups and controls did not differ in baseline rT3. The CH group had decreased levels compared with the other groups. After the high-dose-rhTSH-stimulation, a less pronounced increase was observed in the newCH patients than in controls. The CH group had a lower response than newCH (Figure 2C, 3C). Stimulated rT3 levels did not differ between TSH sufficient groups. A small over-lap was detected in the increase and the maximum rT3 levels after high-dose-stimulation: mean delta rT3 in newCH patients 0.17 nmol/L (range -0.02-0.40) and in controls 0.34 nmol/L (range 0.26-0.46), p<0.05 and peak range in newCH group 0.32-0.56 nmol/L and in controls 0.56-0.81 nmol/L. The peak occurred after 48 or 72 hours in the majority of cases.
IGF-I and insulin (Table 2)
The serum IGF-I level from the replaced control was not included in analysis as methods differed.
Serum IGF-I levels were decreased in the newCH group compared with controls and the GH replaced patients in the CH group. Fasting serum insulin levels did not differ among the groups.

Discussion
This pilot study has explored the response of the thyroid gland to rhTSH in adults with and without TSH deficiency. Patients with newly diagnosed CH and no previous levothyroxine treatment exhibited a less pronounced increase of thyroid hormone levels after administration of 0.9 mg rhTSH than controls. An rhTSH test may, therefore, become useful in the diagnosis of CH. Notably, patients with and without TSH-deficiency displayed a similar increase in serum thyroglobulin levels in response to rhTSH.

In multiple pituitary hormone deficiency, the determination of peripheral thyroid hormone is usually enough in the diagnosis of CH. Nevertheless, some of the newCH subjects exhibited baseline FT4 values within the normal range. This is probably best explained by “regression to the mean” as their low thyroid hormone levels was used for selection into the study. This is also illustrated by the nonCH patient with a baseline FT4 of 8.6, which was a mean of two analyses: FT4 9.8 pmol/L and 7.3 pmol/L. In addition, the six controls had mean FT4 concentrations in the lower part of the normal range by chance. The newCH group had significantly lower baseline FT4 levels than controls, although there was some over-lap.

By including well-characterised hypopituitary patients with sufficient TSH production, we explored the possibility of subjects having partial CH. The results in baseline and stimulated thyroid hormone levels in the nonCH group were, however, consistent with controls; hence, no evidence of partial CH existed. Nonetheless, in clinical work, newly diagnosed CH patients may exhibit FT4 levels in the low normal or subnormal range (7). This is explained by the small intra-individual variation, commonly ± 25%, of thyroid hormone levels (4). Subsequently, a CH patient may have considerably reduced
circulating thyroid hormone levels, but still exhibit FT4 levels within the lower normal reference interval (5, 6).

The aims of this study were to investigate whether the rhTSH test was sufficient as a diagnostic device and to evaluate the response of the thyroid gland to rhTSH in CH patients. The decreased thyroid hormone response to rhTSH was most pronounced in the CH group. These results are, however, difficult to interpret because of previous levothyroxine replacement. Therefore, it is more appropriate to investigate newly diagnosed untreated CH patients. Although the response to 0.9 mg rhTSH in FT4 and rT3 was reduced in newCH patients, the test did not entirely discriminate patients with CH from controls. However, this shall be interpreted with caution as two peak data were missing on one control.

The discriminative value may therefore be better than observed in this study. In addition, under the circumstances of unreplaced GHD FT4 may be higher than under GH treatment because of a decreased peripheral deiodination of T4 to T3. NewCH patients with untreated GHD may therefore have higher FT4 levels than they would have if GH treated. Therefore, the CH of the newCH patients may be more severe than what is illustrated from FT4 levels. Nonetheless, the rhTSH test may identified patients with CH if rT3 <0.56 nmol/L or FT4<20 pmol/L 48-72 hours after 0.9 mg of rhTSH (exact figures assay specific). Moreover, no newCH patient had an increment of FT4>14 pmol/L.

Consequently, in patients with suspected CH the rhTSH test may be used as an adjuvant diagnostic tool.

The low dose rhTSH (0.1 mg) was not as sensitive as the 0.9 mg dose to detect CH, still, the experience from the ACTH-stimulation test indicates that a low dose may be more useful in ACTH-deficiency than in primary adrenal failure (33, 34). However, rhTSH stimulations in patients with nodular goiter demonstrated that a dose of 0.3 mg dose is as potent as 0.9 mg to increase the iodine uptake (35). Therefore, 0.3 mg rhTSH may be an option in CH to minimize the risk of cardiac side effects, even though the 0.9 mg dose was well tolerated in this study.

Before the availability of rhTSH, an increase I^{131} thyroid uptake was observed after multiple repeated bTSH injections in patients with CH (22), demonstrating that a dormant gland could be activated. However, these studies were performed before the introduction of sensitive immuno-methods to determine thyroid hormones and, therefore, no radioiodine uptake was performed in this study.
However, patients with severe CH (CH group) produced higher thyroid hormones level after the second rhTSH injection, regardless of dose. This is most likely due to the activation of the thyroidal cellular system by TSH increasing the iodine content of the thyroid gland and thereby making it more responsive to the next stimuli (22).

The rhTSH-induced Tg response in CH patients indicates that the diminished response in thyroid hormones was not due to unresponsiveness of the TSH receptor of thyroid follicular cells. As endocytosis and, consequently, the removal of Tg from the lumen is diminished, the net result is a gradual accumulation of poorly iodinated Tg (29). This low-iodinated Tg has a low hormone content; hence, a smaller amount of hormones are released from the thyroid after TSH stimulation. This is probably the main reason for the blunted thyroid hormone response to rhTSH found in the two patient groups with CH. Individuals within the CH group probably had a less iodinated Tg compared to newCH individuals, most of whom apparently had a partial stimulation of the thyroid with low – but detectable levels of thyroid hormones. Therefore, the sensitivity of this suggested diagnostic tool may be lower in mild cases of central hypothyroidism.

The normal Tg levels in CH contradicts, however, a reduction in the endocytosis of Tg. As the amount of Tg increases in the absence of TSH, the high concentration in the lumen may allow a large amount of Tg to be taken into the cell by endocytosis, in spite of the reduction in the volume of the endocytotic compartment. Moreover, although the major regulator of Tg synthesis is TSH (27, 28), insulin and IGF-I are able to stimulate Tg synthesis in the absence of TSH (30, 36). Insulin levels did not differ between groups and normal serum IGF-I levels were found in all groups except for the newCH group, where IGF-I levels were low, reflecting untreated growth hormone deficiency. A reduction of basal Tg levels could, therefore, be the suspected in the newCH group. Hence, in addition to accumulation of Tg in the lumen, some other factor, yet unknown, may contribute to the normal Tg production in TSH-insufficiency.

In conclusion, patients with central hypothyroidism have an attenuated increase in serum FT4 and rT3 in response to 0.9 mg rhTSH compared with healthy controls. This observation may be useful in the diagnostic procedure of patients with suspected central hypothyroidism. However, further studies are
needed to establish optimal dose and cut-off levels before implementation of the rhTSH test can occur in clinical practice.
Acknowledgements

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Footnotes

**Table 1**

\(^a\) 2 males with unreplaced hypogonadism, 2 postmenopausal-aged women without oestrogens.

\(^b\) One postmenopausal woman on oestrogens. \(^c\) One postmenopausal woman without oestrogens.

N=number.

**Table 2**

Data are presented as mean ± SEM

n=number of patients. \(^a\) p<0.05, \(^b\) p<0.01, \(^c\) p<0.001 newCH-, CH- and nonCH-groups vs controls.
Legends

**Figure 1**

High-dose-rhTSH response in six patients with previous treated central hypothyroidism (CH), six newly diagnosed CH (newCH), six patients with hypopituitarism without CH (nonCH) compared to six healthy controls in thyroglobulin (A) (without an extreme outlier in the CH group), and FT4 (B). **p<0.05, **p<0.01 newCH- or CH-group vs controls. Values are mean. Error bars=SEM. The horizontal broken lines represent normal range.

**Figure 2**

Baseline and peak levels after 0.9 mg rhTSH in six patients with central hypothyroidism (CH), six patients with newly diagnosed CH (newCH), six patients with hypopituitarism but regarded TSH-sufficient (nonCH) and six healthy controls for FT4 (A), FT3 (B) and rT3 (C). The broken line represents the normal range.*p<0.05, **p<0.01, ***p<0.001 non-CH-, newCH- or CH-group vs controls. § p<0.01 CH- vs nonCH-group, # p<0.05 newCH- vs nonCH-group. ¶ p<0.05 CH- vs newCH-group.

**Figure 3**

The increase of FT4 (A), FT3 (B) and rT3 (C) after 0.9 mg rhTSH in six patients with central hypothyroidism (CH), six patients with newly diagnosed CH (newCH), six patients with hypopituitarism but considered TSH-sufficient (nonCH) and six healthy controls. *p<0.05, **p<0.01, ***p<0.001 non-CH-, newCH- or CH-group vs controls.

§ p<0.05, §§ p<0.01 CH- vs nonCH-group.

&& p<0.01, &&& p<0.001 CH- vs newCH-group.
Table 1  Demography of the study population, presented as mean and range, of patients with central hypothyroidism (CH), patients with newly diagnosed CH (newCH), patients with pituitary insufficiency but intact secretion of TSH (nonCH) and controls.

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<th>Subjects</th>
<th>NewCH (n=6)</th>
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<th>Non-CH (n=6)</th>
<th>Controls (n=6)</th>
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<td><strong>Age (years) mean (range)</strong></td>
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Table 2 Basal and peak serum levels of thyroid-related hormones and thyroglobulin after 0.9 mg rhTSH in patients with treated central hypothyroidism (CH), patients with newly diagnosed CH (newCH), patients with pituitary insufficiency but intact secretion of TSH (nonCH) and controls. In addition, fasting serum insulin and IGF-I levels are presented.

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<th>Analytes</th>
<th>NewCH (n=6)</th>
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<th>NonCH (n=6)</th>
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<tr>
<td>basal</td>
<td>3.7 ± 0.5</td>
<td>5.7 ± 1.1</td>
<td>7.9 ± 2.0</td>
<td>3.4 ± 0.5</td>
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<tr>
<td>peak</td>
<td>68.3 ± 15.2</td>
<td>164 ± 89.6</td>
<td>79.3 ± 12.3</td>
<td>53.2 ± 8.9</td>
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<td><strong>FT4 (pmol/L)</strong></td>
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<td>9.2 ± 0.5a</td>
<td>&lt;5.2 ± 0.0c</td>
<td>11.6 ± 0.9</td>
<td>11.3 ± 0.5</td>
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<tr>
<td>peak</td>
<td>19.7 ± 1.2a</td>
<td>6.4 ± 0.6c</td>
<td>30.0 ± 4.3</td>
<td>27.8 ± 2.4</td>
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<td><strong>TT4 (nmol/L)</strong></td>
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<td>basal</td>
<td>57.0 ± 5.2a</td>
<td>19.6 ± 2.9c</td>
<td>77.8 ± 7.0</td>
<td>72.6 ± 3.3</td>
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<tr>
<td>peak</td>
<td>140.0 ± 9.3</td>
<td>38.7 ± 7.3c</td>
<td>178.3 ± 20.6</td>
<td>161.7 ± 14.3</td>
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<td>1.9 ± 0.2c</td>
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<td>peak</td>
<td>8.4 ± 0.8</td>
<td>3.7 ± 0.7c</td>
<td>10.0 ± 0.7</td>
<td>10.4 ± 1.0</td>
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<td><strong>TT3 (nmol/L)</strong></td>
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<td>1.5 ± 0.0</td>
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<tr>
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<td>1.4 ± 0.3c</td>
<td>3.4 ± 0.2</td>
<td>3.4 ± 0.2</td>
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<td><strong>rT3 (nmol/L)</strong></td>
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<td>0.01 ± 0.01c</td>
<td>0.37 ± 0.05</td>
<td>0.33 ± 0.01</td>
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<tr>
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<td>0.10 ± 0.04c</td>
<td>0.77 ± 0.14</td>
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<td>15.1 ± 3.0</td>
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<td><strong>IGF-I (µg/L)</strong></td>
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<td>98 ± 18b</td>
<td>242 ± 21</td>
<td>189 ± 38</td>
<td>186 ± 12</td>
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</table>
Figure 1A
Figure 2A
Figure 2C
Figure 3A
Figure 3B
Figure 3C