Hormonal response to overfeeding1,3

Gilbert B Forbes, Marilyn R Brown, Stephen L Welle, and Louis E Underwood

ABSTRACT We assessed the hormonal status of adult female volunteers before and during a 3-wk period of weight gain induced by mixed diet overfeeding. Forty-six percent of the 4.3-kg average weight gain experienced by these subjects consisted of lean body mass (LBM) and it is of interest that there were also increases in plasma Somatomedin-C/Insulin-like Growth Factor (SM-C/IGF-1) and testosterone concentrations as well as insulin. We suggest that it was the combined anabolic effect of these three hormones that facilitated the increase in LBM. Of the other assays done, increases were recorded for urinary 17-ketosteroids, 17-hydroxysteroids, epinephrine, and creatinine, whereas there were no changes in serum cortisol or triiodothyronine (T3), or urine norepinephrine; serum thyroxine (T4) fell slightly. Thus it appears that energy surfeit as well as energy deficit (reported by others) has an effect on blood hormone concentrations. Am J Clin Nutr 1989;49:608–11.

KEY WORDS Overfeeding, testosterone, somatomedin, body composition, lean body mass

Introduction

Obese individuals have a greater lean body mass (LBM) than normal weight subjects (1) and deliberate overfeeding of normal volunteers results in significant increases in LBM as well as adipose tissue (2). Although the response of the latter to energy surfeit is to be expected, the reason for the increase in lean tissue is not known. Overfeeding has been shown to enhance insulin secretion (3–5) but the possibility that other hormonal mechanisms could be involved in facilitating LBM accretion has not been explored. Among the hormonal agents that might stimulate anabolism during overfeeding, Somatomedin-C/Insulin-like Growth Factor (SM-C/IGF 1) appears to be a good candidate because its plasma concentration is reduced by fasting and undernutrition and restored by adequate dietary intake (6–9). Another candidate is testosterone, which has known anabolic properties (10). Therefore, we carried out a study in which normal female volunteers were overfed for 21 d, experienced weight gain and significant increments in LBM, and in whom changes in insulin, SM-C/IGF 1, and testosterone (substances known to stimulate anabolism) were monitored. To our knowledge there is no information available on the response of these latter two hormones to overfeeding in human subjects.

As a matter of interest, adrenal and thyroid hormone status was also monitored because changes in these hormones have been described by some but not all investigators in response to overfeeding (3, 5, 11–13).

Subjects and methods

Thirteen normal female subjects aged 18–41 y were housed in the Clinical Research Center for 28 d. They ranged in weight from 44 to 80 kg, in body fat content from 6 to 25 kg (13–35% fat), in LBM from 38 to 59 kg, and in body mass index (wt/ht2) from 17.2 to 28.8. None had a significant change in body weight during the past 2 y. All were in good health with no evidence of diabetes, cardiac or renal disease, or menstrual irregularities. The procedures were explained to the subjects, who gave their written consent. The protocol was approved by the University of Rochester's committee on human investigation.

During the first week of hospitalization the subjects were given a mixed diet (15% of energy from protein, 35–40% from fat, and 45–50% from carbohydrate) in amounts designed to maintain body weight. They were then given an additional 700 kcal/d (2.93 Mi/d) for 2 d, followed by an extra 1200–1600 kcal/d (5.02–6.69 Mi/d) for the next 19 d, which consisted of a total of 21 d of overfeeding. The excess food provided 6% of energy from nonmeat protein, 45–50% from fat, and 45–50% from carbohydrates. Sodium intake was held constant at 170 mmol/d for the entire 28 d. All meals were consumed at the Clinical Research Center.

The total excess energy consumed during the overfeeding period ranged from 18 900 to 35 400 kcal (79 to 148 MJ). The subjects were encouraged to be up and about and to take walks on the hospital grounds; some worked at part-time clerical jobs. Every effort was made to keep each individual's activity pattern

1 From the Departments of Pediatrics, Internal Medicine, and Biophysics, University of Rochester School of Medicine and Dentistry, Rochester, NY, and the Department of Pediatrics, University of North Carolina School of Medicine, Chapel Hill, NC.
2 Supported by NIH grants RR00044, HD18454, AM32562, HD08299, and AM01022.
3 Address reprint requests to GB Forbes, PO Box 777, University of Rochester Medical Center, Rochester, NY 14642.

Received February 18, 1988.
Accepted for publication April 20, 1988.

the same throughout the study. None took oral contraceptive drugs during the study period or for the week before admission to the Clinical Research Center; all had negative pregnancy tests. Data on the energy cost of the weight gain in these subjects were included as part of a general review of overfeeding

Blood samples for hormone assays were obtained during the weight maintenance week and at frequent intervals during the overfeeding period. All samples were obtained at 0730 before breakfast. Three consecutive 24-h urine collections were made on days 4, 5, and 6 of the weight maintenance week and again on days 5, 6, and 7 of the final overfeeding week; each 3-d collection was averaged for data analysis.

Plasma testosterone assays were done with a kit supplied by Radio Assay Systems Laboratory, Carson, CA; serum thyroxine (T₄) and triiodothyronine (T₃) and cortisol with kits supplied by Clinical Assays, Cambridge, MA; plasma SM-C/IGF-1 (in quadruplicate) by methods described previously (14); plasma insulin by double-antibody radioimmunoassay; urinary 17-ketosteroids by the Zimmerman reaction; urinary 17-hydroxycorticosteroids by Porter-Silber colorimetric reaction; urinary C peptide with a kit supplied by Nova Research Institute, Bagsvaard, Denmark; urinary catecholamines by radioenzymatic assay (15); and urinary creatinine in the Multistat III™ autoanalyzer (Instrumentation Labs, Lexington, MA).

Estimates of LBM were made from the ⁴⁰K counts by methods described previously (16); body fat was calculated as weight minus LBM.

Statistical analyses were done on the Clinfo Data Analysis System (BBN Software Products Corp, Cambridge, MA); the significance of differences between means was determined by one-tailed paired t tests.

Results

During the weight maintenance week the mean change in body weight was -0.15 kg (range +0.36 to -0.50 kg), which is not significantly different from 0 (p = 0.2).

From menstrual histories it was determined that five of the subjects were in the follicular phase of their menstrual cycle on admission to the Clinical Research Center, three were in midcycle, and five were in the luteal phase. The mean plasma testosterone values during the weight maintenance week were 2.14 ± 0.40 (x ± SEM), 2.42 ± 0.33, and 1.98 ± 0.24 nmol/L, respectively, for these phases of the menstrual cycle; mean SM-C/IGF-1 values were 1.89 ± 0.34, 1.93 ± 0.23, and 1.76 ± 0.22 U/ml, respectively. In neither instance is there a significant difference among subjects in different phases of the menstrual cycle, nor could we detect a menstrual phase difference in hormone levels on day 21 of overfeeding.

All subjects gained between 3.1 and 5.6 kg in weight during the 21 d of overfeeding and 12 of the 13 volunteers sustained increases in LBM (Fig 1, lower panel). For both weight gain and LBM gain the values at all points were significantly greater than preoverfeeding values. Each subject gained at a steady rate with little evidence of influence from the menstrual cycle. As estimated by ⁴⁰K counting, 46% ± 8% of the gain consisted of lean tissue and 54% was fat. The change in LBM is reflected in the increase in urinary creatinine excretion of 575 ± 194 µmol/24 h (p < 0.02) (Table 1) whereas the increase in body fat is reflected in the changes in skinfold thickness and body circumferences. Triceps skinfold thickness increased by 1.38 ± 0.86 mm, biceps skinfold by 1.87 ± 0.70 mm, and subcapular skinfold by 2.29 ± 0.48 mm; abdominal circumference increased by 2.92 ± 0.92 cm, and buttocks circumference by 2.83 ± 0.69 cm. Except for triceps skinfold, all of these changes were significant (p < 0.05).

The ratio of the mean change in LBM to the mean change in 24-h urinary creatinine excretion, namely 1.86 kg LBM/575 µmol Cr (=0.0032 kg/µmol), was precisely the ratio that had been determined in a study of weight stable normal subjects with widely differing LBM's (17).

The group mean plasma SM-C/IGF-1 concentrations were 1.89 ± 0.15 U/mL before overfeeding and increased significantly (p < 0.02) by day 14 of overfeeding (2.24 ± 0.13 U/mL). Mean fasting plasma insulin concentrations, which were 85.4 ± 8.8 pmol/L before overfeeding, more than doubled by day 14 of overfeeding (p < 0.04). Urinary C peptide, measured in seven subjects, increased from 18.9 ± 2.3 nmol/24 h before overfeeding to a mean value of 24.4 ± 2.6 during the last 3 d of overfeeding (p < 0.05), which reflected enhanced insulin secretion. Likewise, serum testosterone increased from

![FIG 1. Mean values (±SEM) for plasma testosterone (T), SM-C/IGF-1 (SM-C), and insulin (In). Numbers in parentheses above the vertical bars indicate numbers of subjects, those below are p values compared with the pre-overfeeding period. Bottom portion shows mean changes in body weight and LBM (±SEM).](image-url)
TABLE 1
Changes with overfeeding of normal female volunteers

<table>
<thead>
<tr>
<th>Constituent</th>
<th>Initial value*</th>
<th>Change*</th>
<th>Percent change</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma somatomedin-C (U/mL)</td>
<td>1.89 ± 0.15</td>
<td>+0.34 ± 0.14</td>
<td>21</td>
<td>&lt;0.03</td>
</tr>
<tr>
<td>Plasma testosterone (nmol/L)</td>
<td>1.94 ± 0.15</td>
<td>+0.63 ± 0.14</td>
<td>32</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Urine 17-ketosteroids (nmol/24 h)</td>
<td>37.1 ± 2.53</td>
<td>+16.6 ± 2.81</td>
<td>45</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Plasma insulin (pmol/L)†</td>
<td>85.4 ± 8.8</td>
<td>+40.8 ± 12.0</td>
<td>48</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Urine C peptide (nmol/24 h)‡</td>
<td>18.9 ± 2.3</td>
<td>+5.54 ± 2.2</td>
<td>29</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Body weight (kg)</td>
<td>61.4 ± 3.1</td>
<td>+4.34 ± 0.15</td>
<td>7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>LBM (kg)</td>
<td>47.0 ± 1.8</td>
<td>+1.86 ± 0.38</td>
<td>4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Urine creatinine (mg/24 h)</td>
<td>12.280 ± 522</td>
<td>+575 ± 194</td>
<td>5</td>
<td>&lt;0.02</td>
</tr>
<tr>
<td>Urine 17-hydroxysteroids (mg/24 h)</td>
<td>17.9 ± 1.71</td>
<td>+5.96 ± 1.96</td>
<td>33</td>
<td>&lt;0.02</td>
</tr>
<tr>
<td>Urine epinephrine (pg/24 h)</td>
<td>109 ± 12.6</td>
<td>+12.2 ± 5.1</td>
<td>11</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Serum cortisol (pg/mL)§</td>
<td>516 ± 61</td>
<td>+40.3 ± 58</td>
<td>8</td>
<td>0.5</td>
</tr>
<tr>
<td>Serum norepinephrine (pg/24 h)</td>
<td>199 ± 18</td>
<td>+10.5 ± 14.8</td>
<td>5</td>
<td>0.5</td>
</tr>
<tr>
<td>Serum T₃ (pg/mL)</td>
<td>1.78 ± 0.08</td>
<td>+0.053 ± 0.065</td>
<td>3</td>
<td>0.5</td>
</tr>
<tr>
<td>Serum T₄ (pg/mL)</td>
<td>103 ± 3.7</td>
<td>−9.5 ± 3.1</td>
<td>−9</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

* ± SEM. † n = 10. ‡ n = 7. § n = 8. Morning measurement.

basal values of 1.94 ± 0.14 to 2.67 ± 0.20 nmol/L by day 21 of overfeeding (p < 0.01) and urinary excretion of 17-ketosteroids increased (p < 0.001).

During the first 2 wk of overfeeding there were progressive increases in mean SM-C/IGF-1, testosterone, and insulin, all in concert with the increase in LBM. However, during the final week hormone levels seemed to reach a plateau or even to decline somewhat despite a further increase in LBM.

The initial values and the changes recorded in the items noted above by day 21 of overfeeding are given in Table 1. All changes are statistically significant.

The change in SM-C/IGF-1 was significantly correlated with the change in testosterone plus the change in insulin (multiple R² = 0.74). However, neither the magnitude of the change in body weight nor the change in LBM was significantly correlated with a change in SM-C/IGF-1, testosterone, or insulin, either singly or in combination. The problem may lie in the relatively small changes in body weight and in LBM that occurred (7% and 4%, respectively, of initial values).

The influences of overfeeding on other hormones was also investigated (Table 1). There were significant increases in urinary 17-hydroxysteroids and epinephrine. There was a modest albeit significant decline in serum T₃. Serum T₄ did not change nor did serum cortisol or urinary norepinephrine.

Discussion

It is known that plasma insulin and somatomedin levels are reduced in malnutrition (18) and that both decrease when there is an energy deficit (6-9) as does plasma testosterone (19).

The present study shows that overfeeding produces the opposite effect, namely a significant increase in both plasma SM-C/IGF-1 and testosterone, as well as confirming the work of others in regard to insulin. All of these hormones have anabolic properties, therefore, the question posed is whether the increase in their blood concentrations is responsible for the accumulation of lean tissue, which we and others have observed to occur in response to energy surplus.

SM-C/IGF-1 is a potent promoter of growth (20, 21) and the physiological regulation of this hormone seems to make it a logical candidate for stimulating the accumulation of LBM during overfeeding. It is regulated tightly by dietary intake and nutritional status, being reduced to 10–20% of normal by fasting for 10 d (6) and restored promptly by refeeding diets sufficient in energy and protein (7-9).

Although the increase in serum insulin is likely to be mediated by increased substrate, it seems probable that the increment in circulating insulin of 48% (accompanied by a 29% increase in urinary C peptide) could have contributed to the accumulation of lean tissue.

It is known that androgenic-anabolic steroids are potent stimulators of nitrogen retention and their administration readily leads to an increase in LBM in both men and women (10); it is therefore of interest that both plasma testosterone and urinary 17-ketosteroid excretion increased by 32% and 45%, respectively, in our subjects. Although it is doubtful that increases of this magnitude could be solely responsible for the observed increase in LBM; they could certainly have contributed to it.

With regard to changes in other hormones during overfeeding, Katzeff et al (11) found an increase in plasma norepinephrine. Ravussin et al (5) and O'Dea et al (13) reported no change in plasma or urinary catecholamines (the latter found an increase in plasma norepi-
neprine appearance rate) whereas we found an increase in urinary epinephrine but no change in norepinephrine. Horton et al (3) found no change in serum cortisol, nor did we; they reported an increase in cortisol production rate and we found an increase in 17-hydroxycorticoster-
oid excretion. The thyroid response to overfeeding has been inconsistent. Others (11-13) report no change in
serum T₃ whereas we found a slight increase; three
groups of investigators (5, 12, 13) describe an increase in
serum T₄, another group (11) an increase only in lean
subjects, whereas we found no change.

It is unlikely that any of these hormones could facili-
tate the increase in LBM that occurs during overfeeding. Glucocorticoids are catabolic and although thyroid hormone stimulates growth in hypothyroid individuals, those with hyperthyroidism have a reduced total body K
and hence probably a subnormal LBM (22). Even
though epinephrine administration acts to reduce pro-
tein breakdown (23), we found only a small increase in
epinephrine excretion whereas others report no change.

This brings us back to the three hormones:—SM-C/
IGF-1, testosterone, and insulin—as likely candidates for facilitating the increase in LBM observed in our sub-
jects.

Because the increases in SM-C/IGF-1, testosterone, and insulin were found to be intercorrelated in our sub-
jects, it is difficult to assign priority to any one of them
in connection with the observed increase in LBM. Given the known anabolic activity of these three hormones, our
data suggest that their combined action could have been
responsible for the increment in lean tissue sustained by
our subjects. However, because of the lack of a correla-
tion between the magnitude of the increase in hormone
levels and the extent of the increase in LBM, this conclu-
sion must be regarded as tentative. Taken together with
the findings of others in regard to underfeeding, our dem-
stration of an increase in anabolic hormones in re-
sponse to overfeeding adds strength to the concept that
nutritional status can influence hormonal function.

We thank Cheryl Porta for doing the ⁴⁰K assays and the staff of the Clinical Research Center for their gener-
ous cooperation.

References

1. Forbes GB, Welle SL. Lean body mass in obesity. Int J Obesity
2. Forbes GB, Brown MR, Welle SL, Lipinski BA. Deliberate over-
feeding in women and men: energy cost and composition of the
3. Horton ES, Danforth E Jr, Sims EAH, Salans LB. Endocrine and
metabolic alterations in spontaneous and experimental obesity. In:
Bray GA, ed. Obesity in perspective. Washington, DC: DHEW,
1975. (DHEW Publication [NIH] 75-708.)
4. Olefsky J, Crapo PA, Ginsburg H, Reaven GM. Metabolic effect
5. Ravussin E, Schutz Y, Acheson KJ, Dusmit M, Bourquin L,
Jéquier E. Short term, mixed diet overfeeding in man; no evidence
6. Clemons DR, Kilbanski A, Underwood LE, et al. Reduction of
immunoreactive somatomedin-C during fasting in humans. J Clin
7. Clemons DR, Underwood LE, Dickerson RN, et al. Use of So-
matomediC-Inulin-like Growth Factor I measurements to
monitor nutritional repletion in malnourished patients. Am J Clin
8. Isley WL, Underwood LE, Clemons DR. Changes in plasma so-
matomediC-in response to ingestion of diets with variable protein
9. Isley WL, Underwood LE, Clemons DR. Dietary components
that regulate serum somatomedin-C concentrations in humans. J
10. Forbes GB. The effect of anabolic steroids on lean body mass: the
11. Katzeff HL, O’Connell M, Horton ES, Danforth Jr E, Young JB,
Landsberg L. Metabolic studies on human obesity during overnu-
trition and undernutrition: thermogenic and hormonal responses
to norepinephrine. Metabolism 1986;35:166-75.
alterations in thyroid hormone metabolism during overnutrition.
J Clin Invest 1979;64:1336-47.
turnover during undereating and overeating in normal weight sub-
14. Copeland KC, Underwood LE, Van Wyk JJ. Induction of immu-
noreactive somatomedin-C in human serum by growth hormone:
dose response relationships and effect on chromatographic profiles.
15. Welle SL, Campbell R. Effect of overeating on plasma and urinary
concentration of norepinephrine. J Clin Endocrinol Metab
1984;59:531-4.
16. Forbes GB, Schultz F, Cafarella C, Amirhakimi GH. Effects of
body size on potassium-40 measurement in the whole body
17. Forbes GB, Bruining GJ. Urinary creatinine excretion and lean
18. Torin B, Viteri FE. Protein-energy malnutrition. In: Shils ME,
Young VR, eds. Modern nutrition in health and disease, 3rd ed.
19. Nair KS, Woolf PD, Welle SL, Matthews DE. Leucine, glucose,
and energy metabolism after 3 days of fasting in healthy human
20. van Buul-Offers S, van den Brande JL. Effect of growth hormone
and peptide fractions containing somatomedin activity on growth
and cartilage metabolism in Snell dwarf mice. Acta Endocrinol
21. Schoenle E, Zepf J, Humbel RE, Sprueche RE. Insulin-like growth
factor I stimulates growth in hypophysectomized rats. Nature
1982;296:252-3.
22. Edmonds CJ, Smith T. Total body potassium in relation to thyroid
23. Miles JM, Nissen SL, Gerich JE, Haymond MW. Effects of epi-
nephrine infusion on leucine and alanine kinetics in humans. Am