

## Negative energy balance plays a major role in the IGF-I response to exercise training

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Submitted 24 June 2003; accepted in final form 23 August 2003

**Nemet, Dan, Peter H. Connolly, Andria M. Pontello-Pescatello, Christie Rose-Gottron, Jennifer K. Larson, Pietro Galassetti, and Dan M. Cooper.** Negative energy balance plays a major role in the IGF-I response to exercise training. *J Appl Physiol* 96: 276–282, 2004. First published August 29, 2003; 10.1152/jappphysiol.00654.2003.—Circulating IGF-I is correlated with fitness, but results of prospective exercise training studies have been inconsistent, showing both increases and decreases in IGF-I. We hypothesized that energy balance, often not accounted for, is a regulating variable such that training plus an energy intake deficit would cause a reduction in IGF-I, whereas training plus energy intake excess would lead to an increased IGF-I. To test this, 19 young, healthy men completed a 7-day strenuous exercise program in which they were randomly assigned to either a positive energy balance [overfed (OF),  $n = 10$ ] or negative energy balance [underfed (UF),  $n = 9$ ] group. IGF-I (free and total), insulin, and IGF-binding protein-1 were measured before, during, and 1 wk after the training. Weight decreased in the UF subjects and increased in the OF subjects. Free and total IGF-I decreased substantially in the UF group ( $P < 0.0005$  for both), but, in the OF group, IGF-I remained unchanged. The UF group also demonstrated an increase in IGF-binding protein-1 ( $P < 0.027$ ), whereas glucose levels decreased ( $P < 0.0005$ ). In contrast, insulin was reduced in both the OF and UF exercise-training groups ( $P < 0.044$ ). Finally, within 7 days of the cessation of the diet and training regimen, IGF-I and IGF-binding protein-1 in the UF group returned to preintervention levels. We conclude that energy balance during periods of exercise training influences circulating IGF-I and related growth mediators. Exercise-associated mechanisms may inhibit increases in IGF-I early in the course of a training protocol, even in overfed subjects.

diet; growth; insulin-like growth factor-I; growth hormone

THE GROWTH HORMONE-IGF-I AXIS is a system of mediators and binding proteins that modulates growth in many tissues and is now known to play a key role in the adaptation to exercise (1). Circulating IGF-I is higher in fitter subjects and is correlated with lean body mass (LBM) (9, 10, 22). Based on these observations, one might expect that exercise training would lead to increases in IGF-I, but the results from prospective training studies have been mixed (16, 26), with studies showing both increases and reductions in IGF-I.

These seemingly contradictory findings might be explained, in part, by the substantial role that overall energy balance plays in the regulation of circulating IGF-I. Negative energy balance, whether caused by increasing exercise energy expenditure with an exercise training program or by reducing energy intake without increasing exercise, causes a reduction in circulating IGF-I within several days (17, 28). Conversely, IGF-I increases

with overfeeding, even without a change in physical activity (11). Collectively, these previous studies suggest the following rather straightforward hypothesis: circulating IGF-I will be reduced in response to an exercise training program when energy balance is negative but increased when energy balance is positive.

To test this, we examined the combined influence of 7 days of diet alteration and strenuous exercise training on body composition, IGF-I and key binding proteins, and related growth mediators in a group of young, healthy adult men. We designed the experiment to carefully measure energy balance, weight change, and circulating IGF-I so that we could begin to examine a quantitative relationship between IGF-I and energy balance during a short period of intense exercise training. We made sure that, even in the underfed group, protein intake and hydration were sufficient so as to minimize confounding effects of either a low-protein diet or changes in body water compartments (6, 19). Finally, we measured growth mediators early in the morning, at least 18 h after any previous exercise, to minimize the potential confounding effects of acute hormonal responses to exercise.

### METHODS

#### Study Design and Sample Population

The study lasted a total of 3 wk with a 7-day exercise training and diet component (Fig. 1) during *week 2*. Subjects underwent a preparticipation examination (medical history, physical examination, ECG, and complete blood count) performed within 1 mo before the beginning of the study by a physician who was not part of the study team. Inclusion criteria were as follows: male, age 18–25 yr, peak  $\dot{V}O_2$  of 35–45 ml  $O_2 \cdot kg^{-1} \cdot min^{-1}$ , normal body mass index (BMI), and nonsmoking. Subjects with asthma; cardiorespiratory, musculoskeletal, or metabolic disease; or eating disorders were excluded. No subjects were on any medications at the time of the study.

From an original pool of 24 subjects, 22 met the inclusion and exclusion requirements and were randomly assigned to either a calorie-restricted underfed (UF) or calorie-supplemented overfed (OF) group. The participants were not informed of their group assignment until the dietary manipulation began in *week 2* so as to preclude any individual changes in diet before the intervention.

The study was designed so that subjects in the OF group would experience a positive energy balance of ~15% during the 7 days of exercise training by supplying an enriched calorie diet. In the UF group, the subjects' diets were calculated to achieve a negative energy balance of ~33% during the 7 days of training. This figure was chosen because it was roughly comparable to the negative energy balance

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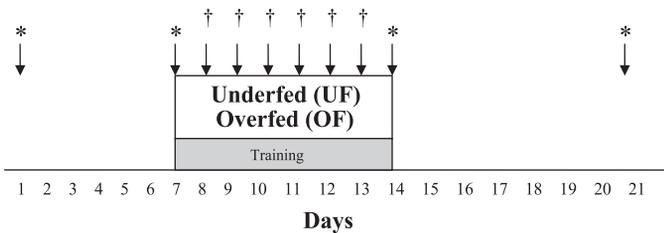


Fig. 1. Study design and assays time table. UF, underfed; OF, overfed. \*Anthropometrics, blood draw, body composition (dual-energy X-ray absorptiometry), fitness test, resting energy expenditure, dietary records. †Anthropometrics, blood draw, dietary records.

used by Smith et al. (31) in their previous study. The study was approved by the Institutional Review Board, University of California, Irvine (UCI), and written, informed consent was obtained.

#### Nutritional Protocol

**Dietary assessment.** To assess each subject's eating patterns and, thereby, to plan a palatable diet, participants completed a 3-day food record before the intervention period (32). The records were analyzed by using the Nutritionist Pro computerized database (First DataBank, San Bruno, CA).

**Dietary intervention.** During *week 2*, breakfast and lunch were provided by the UCI General Clinical Research Center (GCRC) Metabolic Kitchen. Meals were preportioned and individually provided with a known caloric content, depending on the group to which the subject had been assigned. Food items were weighed to the nearest gram before serving, and any uneaten items were evaluated after the meal.

Subjects were given specific guidelines to follow for caloric content of their evening meal. To document food and beverages consumed in the afternoons and evenings, subjects were asked to keep a log or diary of meals, as well as any additional food or beverage consumed during *week 2*. Each morning, the previous day's records were reviewed by a dietitian to clarify any questions or problems that the subjects had regarding food portions, etc.

In addition, subjects were asked to avoid all dietary supplements and caffeine for the duration of the 3-wk study. The subjects worked with the project GCRC dietitian to ensure that the diets (both UF and OF) were properly balanced and contained sufficient amounts of protein ( $>1 \text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ ), basic nutrients, and fluids. The GCRC dietitian was on call 24 h/day to answer any dietary questions.

#### Exercise Testing and Training

Subjects underwent an intense 7-day exercise training program during *week 2*, administered by the athletic trainers at the UCI student and faculty recreation center, which is a large, well-equipped sports and training facility located on the UCI main campus. Exercise prescriptions were generated individually, according to resting energy expenditure (REE) and fitness level, as determined by the maximal exercise test. The goal of the training program was to increase total energy expenditure by  $\sim 25\%$ .

Subjects exercised 3 h/day for 7 consecutive days. The training began every morning at  $\sim 9:00$  AM after baseline blood draws, measurement of postvoiding weight, and breakfast provided by the GCRC registered dietitian. The session began with low-intensity aerobic warm-up (on a treadmill, cycle ergometer, or stepping device) for  $\sim 20$  min; this was followed by an instructor-led short session of stretching. The instructor would then choose an activity for which the participants could maintain an elevated heart rate (target heart rate was predetermined for each subject) for a total of at least 1 h. These activities included stepping, spinning, cycling, or aqua-fitness classes, soccer and basketball games, or interval running. In the remainder of

the time, the subjects performed low-intensity aerobic exercise, followed by cool-down activities and stretching. The energy expenditure was planned to increase by an estimated minimum of  $20 \text{ ml O}_2\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$  for 60 min and  $10 \text{ ml O}_2\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$  for 120 min. To monitor exercise intensity, heart rates were measured both by a Polar heart rate monitor (Polar Accurex Plus, Polar Electro, Woodbury, NY) and manually every 10–20 min.

The following measurements were performed (Fig. 1).

**Height, weight, and BMI.** Standard, calibrated scales and stadiometers were used to determine height, weight, and BMI (weight/height<sup>2</sup>).

**Assessment of body composition by dual-energy X-ray absorptiometry.** Body composition was measured by dual-energy X-ray absorptiometry with a Hologic QDR 4500 densitometer (Hologic, Bedford, MA), a well-established technique for assessing body composition. Subjects were scanned in light clothing while lying flat on their backs. On the days of each test, the dual-energy X-ray absorptiometry machine was calibrated by using the procedures provided by the manufacturer.

**Measurement of cardiorespiratory fitness.** Each subject performed a ramp-type progressive exercise test on an electronically braked, servo-controlled, cycle ergometer (Ergoline 800S SensorMedics, Yorba Linda, CA). After an initial warm-up (0-W pedaling), the work rate increased progressively (ramp function) until the limit of the subject's tolerance was reached. The ramp slope was chosen so that the subject would complete the exercise in 8–12 min. Subjects were instructed to maintain a constant pedaling frequency at 60 rpm. Subjects were vigorously encouraged during the high-intensity phases of the exercise protocol. Gas exchange was measured breath by breath (3) by using a Vmax 229 metabolic cart (SensorMedics), and the peak  $\dot{V}\text{O}_2$  was determined as previously described (7).

**Evaluation of REE.** Energy expenditure was determined by indirect calorimetry. Oxygen and carbon dioxide exchange were measured by using a metabolic cart with an open canopy system (Vmax 229 calorimeter, SensorMedics). Subjects were instructed to avoid exercise as well as stimulants, such as caffeine, tobacco, or medication, at least 24 h before the test. Measurements were performed in the morning after an overnight fast and after the patient had been recumbent for at least 30 min. After initiating calorimetry, time was allowed for measured values to stabilize, and REE was then measured over a 10-min period of steady state. The flow,  $\text{O}_2$ , and  $\text{CO}_2$  sensors were calibrated before each test.

#### Serum Measurements

Fasting blood samples were obtained by standard phlebotomy at  $\sim 8:00$  AM. Samples were placed in an ice bath and were immediately centrifuged. Aliquots of the resulting plasma were stored at  $-80^\circ\text{C}$  until analyzed. All specimens were analyzed in the same batch by technicians who were blinded to the order of the samples and grouping of subjects.

**Albumin.** Albumin levels were determined by colorimetric determination by the use of the Sigma BCP albumin procedure no. 256 (Sigma Diagnostics, St. Louis, MO).

**Hematocrit.** Hematocrit levels were determined by using a standard technique.

**IGF-I: total and free.** IGF-I was extracted from IGF-binding proteins (IGFBPs) by using the acid-ethanol extraction method (8). Serum IGF-I concentrations were determined by a two-site immunoradiometric assay by using the DSL-5600 Active kit (Diagnostic System Laboratories, Webster, TX). IGF-I interassay coefficient of variation (CV) was 3.7–8.2%, and intra-assay CV was 1.5–3.4%. Assay sensitivity was 0.8 ng/ml. Free IGF-I was determined by ELISA with the use of the DSL-10-9400 Active kit (Diagnostic System Laboratories). Intra-assay CV was 3.74–4.8%, interassay CV was 6.2–11.1%, and the sensitivity was 0.015 ng/ml. This commercially available assay measures the sum of free plus readily disso-

able IGF-I. Free IGF-I levels by ELISA are known to be slightly elevated compared with those detected by the ultrafiltration method (13) or the recently described IGF-I kinase receptor activation assay (5); both are not commercially available.

**IGFBPs.** IGFBP-1 was measured by coated-tube immunoradiometric assays with the use of the DSL-10-7800 Active kit (Diagnostic System Laboratories). For IGFBP-1, interassay CV was 1.7–6.7%, and intra-assay CV was 2–4%. Assay sensitivity is 0.33 ng/ml (results for IGFBP-1 were not obtainable for one subject). IGFBP-2 serum concentrations were determined by RIA with the use of the DSL-7100 kit (Diagnostic System Laboratories). Intra-assay CV was 4.7–8.5%, interassay CV was 7.2–7.4%, and the sensitivity was 0.5 ng/ml. IGFBP-3 serum concentrations were determined by ELISA with the use of the DSL 10-6600 Active kit (Diagnostic System Laboratories). Intra-assay CV was 7.3–9.6%, interassay CV was 8.2–11.4%, and the sensitivity was 0.04 ng/ml. IGFBP-4 serum concentrations were determined by ELISA with the use of the DSL 10-7300 Active kit (Diagnostic System Laboratories). Intra-assay CV was 2.8–6.4%, interassay CV was 2.3–6.7%, and the sensitivity was 1 ng/ml. IGFBP-6 serum concentrations were determined by RIA with the use of the DSL-9800 kit (Diagnostic System Laboratories). Intra-assay CV was 6.4–10.7%, interassay CV was 6.1–9.5%, and the sensitivity was 1.1 ng/ml.

**Glucose.** Serum glucose levels were determined by quantitative enzymatic measurements with the use of Sigma diagnostic kit no. 510 (Sigma Diagnostics).

**Insulin.** Insulin serum levels were determined by ELISA with the use of the DSL-10-1600 Active kit (Diagnostic System Laboratories). Intra-assay CV was 1.3–2.6%, interassay CV was 5.2–6.2%, and the sensitivity was 0.26  $\mu$ U/ml.

#### Statistical Analysis

Two-way (time  $\times$  group) ANOVA for repeated measures was used to determine the effect of exercise on all variables. Repeated measures over time, as well as groups (i.e., low calorie vs. high calorie), were accounted for in the covariance structure of the ANOVA models.

Pairwise *t*-test comparisons of interest (2-tailed) were made for each outcome variable when main effects were found to be significant. Standard linear regression analysis was used to determine correlation coefficients and parameter estimates. Data are presented as means  $\pm$  SE. Statistical significance was set at  $P < 0.05$ .

## RESULTS

Nineteen subjects completed the study. Subject characteristics are presented in Table 1. No differences were found between the two groups at baseline.

Table 1. Subject characteristics

	High Calorie-Overfed	Low Calorie-Underfed
<i>n</i>	10	9
Age, yr	20.7 $\pm$ 0.68	20.33 $\pm$ 0.44
Height, cm	172.5 $\pm$ 2.78	171.6 $\pm$ 1.50
Weight, kg	70.05 $\pm$ 2.27	73.68 $\pm$ 3.29
BMI, kg/m <sup>2</sup>	23.59 $\pm$ 0.77	24.98 $\pm$ 0.97
Peak $\dot{V}O_2$ , l/min	2.64 $\pm$ 0.12	2.49 $\pm$ 0.12
Peak $\dot{V}O_2$ , ml·min <sup>-1</sup> ·kg <sup>-1</sup>	37.92 $\pm$ 1.584	34.86 $\pm$ 1.257
Resting respiratory exchange ratio	0.85 $\pm$ 0.01	0.83 $\pm$ 0.03
Resting energy expenditure, kcal/24 h	1,826.90 $\pm$ 61.70	1,859.44 $\pm$ 76.15

Values are means  $\pm$  SE; *n*, no. of subjects. BMI, body mass index;  $\dot{V}O_2$ ,  $O_2$  uptake.

Table 2. Dietary intake and energy expenditure during intervention week

	High Calorie-Overfed	Low Calorie-Underfed
<i>n</i>	10	9
Dietary intake, kcal/kg	57.95 $\pm$ 1.734	23.41 $\pm$ 0.768
Protein intake, g·kg <sup>-1</sup> ·day <sup>-1</sup>	1.925 $\pm$ 0.06	1.161 $\pm$ 0.09
Daily dietary intake, kcal/day	4,021 $\pm$ 131	1,714 $\pm$ 78
Total energy expenditure without exercise, kcal/day	2,612 $\pm$ 88	2,659 $\pm$ 108
Energy expenditure in exercise session, kcal/3 h	1,055 $\pm$ 61	1,034 $\pm$ 77
Total energy expenditure, kcal/day (TEE + exercise session)	3,689 $\pm$ 106	3,797 $\pm$ 168
Energy balance, kcal/day	393 $\pm$ 150	-2,052 $\pm$ 83

Values are means  $\pm$  SE; *n*, no. of subjects. TEE, total energy expenditure.

#### Effect of Diet and Exercise Intervention

Dietary intake and energy expenditure during the intervention week are presented in Table 2.

#### Body Weight, Fitness, and Body Composition

The UF group had an average weight loss of  $-1,448 \pm 246$  g ( $P < 0.0001$ ) during the training week, whereas the OF group gained  $890 \pm 270$  g ( $P < 0.009$ , Fig. 2). LBM increased in the OF group ( $1,001 \pm 403$  g,  $P < 0.035$ ), whereas no significant change in LBM was found in the UF group ( $-147 \pm 275$  g, not significant). Body fat decreased significantly in the UF group ( $-1,260 \pm 206$  g fat;  $P < 0.0005$ ), whereas the decrease in body fat in the OF group ( $-271 \pm 146$  g fat) was not significant. Interestingly, despite the decrease in LBM, fitness ( $\dot{V}O_2$ ; ml·min<sup>-1</sup>·kg<sup>-1</sup>) increased only in the UF group ( $9.4 \pm 3.4\%$ ,  $P < 0.024$ ) (Table 3). Neither group demonstrated significant changes in REE.

#### Serum Measurements

**Albumin.** No significant changes were noted in albumin level throughout the intervention.

**Hematocrit.** No significant changes were noted in hematocrit levels throughout the intervention.

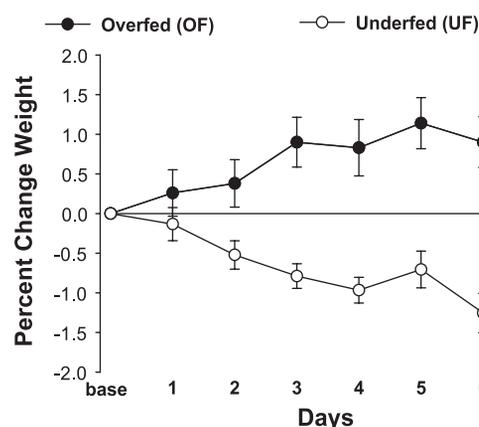


Fig. 2. Percent change in weight after the intervention week. Values are means  $\pm$  SE. base, Baseline. The UF group lost a significant amount of weight ( $P < 0.0001$ ), whereas the OF had a significant weight gain ( $P < 0.009$ ).

Table 3. Change in fitness during intervention week

	Peak $\dot{V}O_2$							
	l/min		ml·min <sup>-1</sup> ·kg <sup>-1</sup>		ml·min <sup>-1</sup> ·kg LBM <sup>-1</sup>		Power Output, W	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post
Low calorie-underfed	2.49±0.12	2.71±0.11*	33.87±1.26	37.76±1.22*	41.58±1.24	45.39±1.05*	222.1±7.6	235.3±8.4*
High calorie-overfed	2.64±0.12	2.72±0.15	37.79±1.51	38.57±1.62	45.72±1.94	46.12±1.99	235.6±10.8	250.7±12.3*

Values are means ± SE. LBM, lean body mass; Pre, before intervention; Post, after intervention. \* $P < 0.05$ .

**Total and free IGF-I.** The effect of diet and exercise on serum levels of total and free IGF-I is shown in Fig. 3. A decrease in both total and free IGF-I levels was noted only in the UF group ( $P < 0.0005$ ). After recovery, total and free IGF-I levels returned to baseline.

We examined the correlation between change in body weight and percent change in total IGF-I (Fig. 4). A significant correlation was found ( $r = 0.65$ ,  $P < 0.02$ ). Moreover, a significant, negative  $y$ -intercept was found ( $-15.7 \pm 3.2\%$ ,  $P < 0.0001$ ). A similar analysis was performed using percent change energy balance and percent change in total IGF-I in all 19 subjects. Again, a significant correlation was found ( $r = 0.77$ ,  $P < 0.0005$ ) with a significant negative  $y$ -intercept ( $-8.9 \pm 3.6\%$ ,  $P < 0.0024$ ).

**IGFBP-1.** There was a significant increase in IGFBP-1 during the exercise week (week 2) only in the UF group ( $P < 0.027$ , Fig. 5). IGFBP-1 returned to baseline levels after recovery.

**IGFBP-2, -3, -4, and -6.** There were no significant changes in IGFBP-2, -3, -4, and -6 during the intervention.

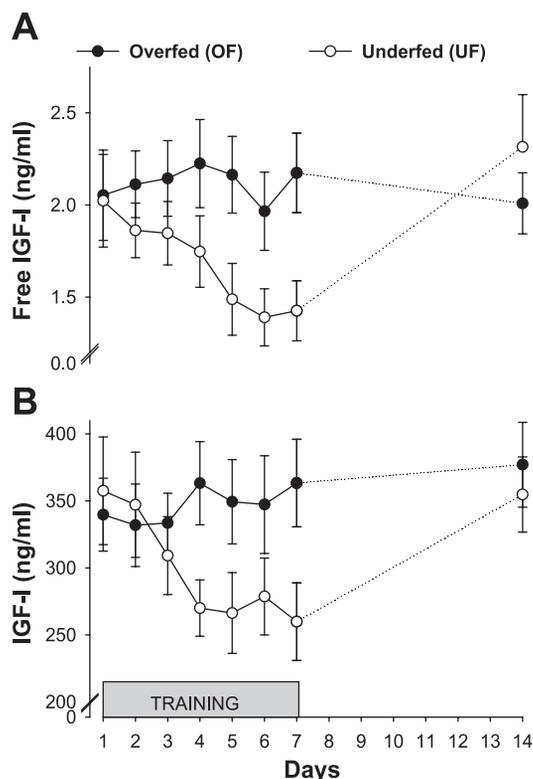


Fig. 3. Effect of diet and exercise on circulating free (A) and total (B) IGF-I. During intervention, free and total IGF-I levels significantly decreased only in the UF group ( $P < 0.0005$  for both).

**Insulin.** There was a significant effect of time on insulin levels in both UF and OF groups during the exercise week (week 2) ( $P < 0.044$ , Fig. 5). No difference was found between the groups. Insulin levels returned to baseline after recovery.

**Glucose.** There was a significant decrease in glucose levels during the exercise week (week 2) only in the UF group ( $P < 0.005$ , Fig. 5). Glucose levels returned to baseline after the recovery week.

## DISCUSSION

The data only partially supported our original hypothesis: we did find a substantial reduction in IGF-I in the UF exercise-training subjects, and this rapidly returned to baseline levels after the intervention ceased. However, despite an increase in LBM, weight gain, and peak work rate, we were unable to demonstrate an increase in circulating IGF-I in the trained OF subjects. Two additional observations emerged from our data that might help explain disparate IGF-I results from exercise training that have appeared in the recent literature (16, 26). First, IGF-I can either increase or decrease within days in response to changes in energy balance and/or levels of physical activity; thus the timing of sampling relative to the intervention must be accounted for. Second, our data would predict that, even under conditions in which subjects are weight stable and/or maintain energy balance, exercise training may, in the short term, prevent an increase in circulating IGF-I.

We did achieve a major goal of our study design to contrast the effects of exercise training on clearly UF with OF subjects, while protein intake and hydration were maintained. The weight changes in the two groups were consistent with the planned alterations in energy balance (Fig. 2), and no significant change in circulating albumin or hematocrit occurred

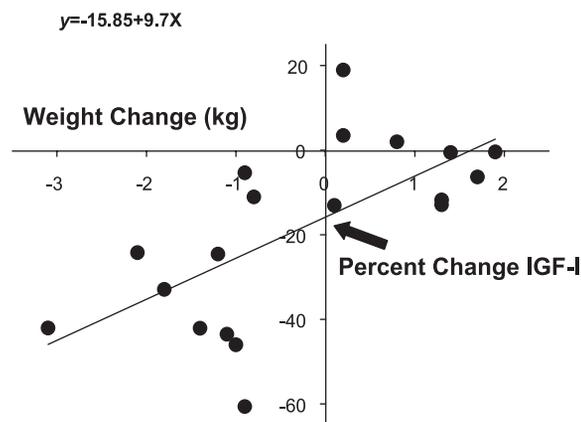


Fig. 4. Individual changes in IGF-I associated with exercise training as a function of change in body weight. Changes in IGF-I were correlated to weight change with a negative  $y$ -intercept (arrow).

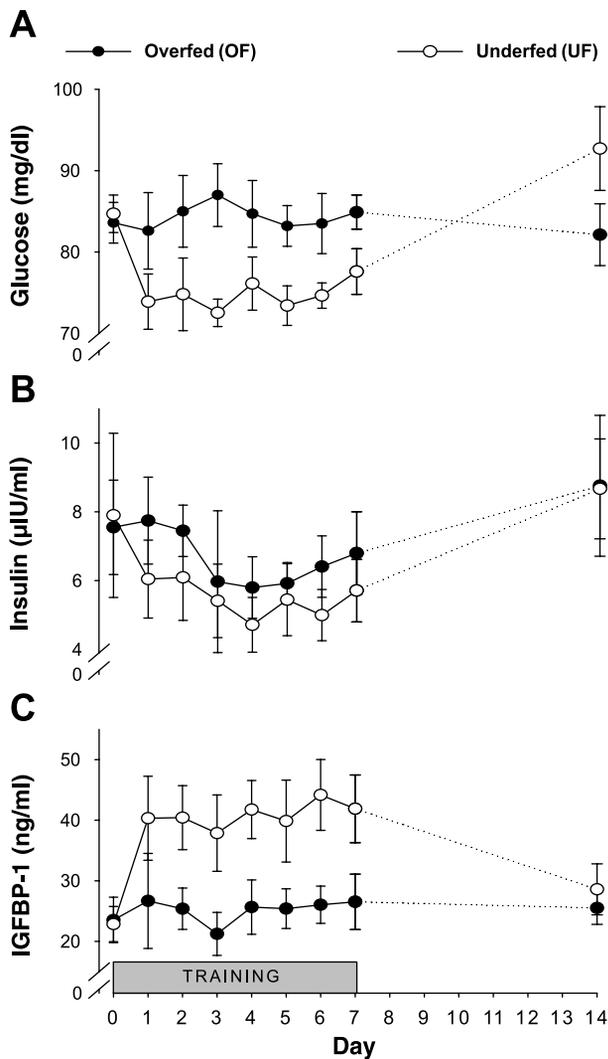


Fig. 5. Effects of diet and exercise intervention on levels of glucose (A), insulin (B), and IGF-binding protein-1 (IGFBP-1; C). Values are means  $\pm$  SE. Glucose decreased ( $P < 0.005$ ), whereas IGFBP-1 increased, during intervention week only in the UF group ( $P < 0.027$ ). There was a significant decrease in insulin in both UF and OF groups ( $P < 0.04$ ).

(note: we did not, however, formally measure protein turnover or body water distribution). The weight gain in the OF group was due mainly to an increase in LBM, whereas the loss in the UF group was predominantly body fat, and this is in contrast to what is typically found in fasting or severe caloric restriction without exercise (23). In both groups, subjects significantly increased peak work rate; however, peak  $\dot{V}O_2$  normalized to body weight or to LBM increased in the UF but not the OF group. This latter finding suggests that the increased fitness in the UF subjects occurred not primarily because of increases in muscle mass, but, rather, was due to alterations in other factors [e.g., muscle blood flow, neuromuscular efficiency, mitochondrial density (4)] that are also known to accompany exercise training.

We measured both free and bound IGF-I. Because the bulk of circulating IGF-I is bound in a ternary complex [IGF-I, IGFBP-3, and an acid-labile subunit (24)] too large to cross the capillary membranes, it has been postulated that the free, unbound IGF-I may be more physiologically active. Moreover,

free and bound IGF-I do not always respond to exercise stimuli in parallel. For example, our laboratory recently demonstrated preservation of free IGF-I levels while total IGF-I levels decrease after an acute strenuous bout of exercise (20). However, in the present study, the decrease in free and total IGF-I found only in the UF group paralleled one another, and both were attenuated by overfeeding (Fig. 3). Thus overfeeding likely mitigated any physiological consequence of exercise training on circulating IGF-I.

This study is the first to clearly demonstrate that the IGF-I response to exercise training can be altered by manipulations of diet. The reduction in IGF-I can occur in a matter of days, whether induced by starvation or by energy imbalance associated with exercise, as demonstrated previously (31) and in the present study. We wondered whether or not the present data, in which we carefully measured IGF-I levels and energy balance, could corroborate earlier observations on IGF-I and exercise training (29). To do this, we plotted the individual changes in IGF-I associated with exercise training as a function of change in energy balance and body weight (Fig. 4). The regression analysis showed a significant, negative  $y$ -intercept, suggesting that, even when energy intake equals energy expenditure and/or no weight change occurs, IGF-I will fall.

In a series of prospective training studies in children and adolescents who were energy balanced, we found reductions in circulating IGF-I ranging from 3 to 14% that fit well within the predictive range of Fig. 4 (29). Thus, in the absence of frank overfeeding, our data suggest that exercise training will lead to a decrease in IGF-I, even in weight-stable subjects. The mechanism of this exercise-training, IGF-I-suppressing effect is not yet known.

Our data show how easily IGF-I responses to exercise training can be confounded by a number of key factors. First, as seen in Fig. 4, the reduction in IGF-I is quite sensitive to energy balance: there may be small reductions when energy balance is maintained, but small alterations in energy balance in either direction could substantially influence the IGF-I levels.

Second, when the intervention of training and diet ceases, the return to preintervention levels occurs quite quickly (Fig. 3). In addition, some investigators have found a rebound in IGF-I levels after a period of heavy training in which the "posttraining" IGF-I exceeds the pretraining levels (12, 25). These insights might explain the apparent discrepancy between cross-sectional and some prospective studies of exercise training on IGF-I. During training, in the absence of overfeeding, IGF-I is likely to be depressed. In contrast, in cross-sectional studies, subjects are usually excluded if they are undergoing vigorous training at the time of the study; consequently, the fitter subjects are those who might already be in the "rebound" phase after periods of heavy training. The time course that such a putative increase in IGF-I in response to exercise occurs is not known.

We also examined key regulators of IGF-I in an effort to gain greater insight into the effect of energy balance on the IGF-I response. Of the five IGFBPs that we measured, only IGFBP-1 changed significantly. IGFBP-1 is found predominantly in tissues, not in circulating blood, and acts primarily to inhibit physiological effects of IGF-I (24). Circulating IGFBP-1 likely originates in the liver, is inversely related to levels of circulating insulin (24), and is acutely elevated with

exercise in which insulin decreases as well (20). Moreover, a number of investigators have noted increased IGFBP-1 with prolonged exercise training, which is also characterized by reduced insulin (14). We also found an increased IGFBP-1 with exercise training, but this effect was eliminated by overfeeding, despite the fact that insulin remained low in the OF, exercise-trained group.

The absence of the inverse relationship between insulin and IGFBP-1 in response to acute exercise was noted recently by several investigators in experiments that dissociated glucose and insulin from IGFBP-1 (2, 15). Thus the regulation of IGFBP-1 during exercise training and its relationship to energy balance is still not well understood. One possible explanation for our results might be found in the fact that overfeeding prevented the decrease in glucose that occurred in the UF, exercise-trained subjects. In the UF subjects, regulatory mechanisms likely existed to limit diet-induced reductions in blood glucose [note: reduced blood glucose is not typically found in training studies without dietary restriction (21)]. Elevated IGFBP-1 might be one of the mechanisms that acts to conserve circulating glucose by inhibiting the insulin-like, hypoglycemic effects of IGF-I (18). By contrast, in the OF subjects, glucose was not reduced, and, consequently, there was no metabolic stimulation of additional glucose sparing mediators, like IGFBP-1.

In summary, we did not find increased IGF-I with overfeeding in the context of exercise training, increased LBM, and increased body weight. Our data demonstrate the sensitive relationship between energy balance and training in the regulation of circulating IGF-I and suggest some as yet unknown mechanisms associated with exercise that tend to reduce IGF-I, even when energy balance and weight stability are maintained. Whether or not these relationships hold for training periods that last longer than 7 days is not known.

Controversy exists as to the long-term benefits of environmentally induced changes in circulating IGF-I. Clearly, adverse conditions associated with great morbidity and mortality, such as sepsis, burns, or trauma, are accompanied by reductions in IGF-I. In contrast, low IGF-I induced, for example, genetically or by dietary manipulation in otherwise healthy animals, may be associated with increased longevity (30, 33). The physiological meaning or role of alterations in circulating IGF-I in the context of exercise training have yet to be fully elucidated.

#### ACKNOWLEDGMENTS

The authors thank Mike Puritz and the University of California Anteater Recreation Center trainers and staff for their assistance in the study. Last but not least, we thank the participants for their cooperation.

#### GRANTS

This work was supported by National Institutes of Health Grants MO1-RR-00827 and HD-23969. D. Nemet is a Postdoctoral Research Fellow of the Joseph W. Drown Foundation.

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