

# Body Composition and Hormonal Adaptations Associated with Forskolin Consumption in Overweight and Obese Men

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## Abstract

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**Objective:** This study examined the effect of forskolin on body composition, testosterone, metabolic rate, and blood pressure in overweight and obese ( $BMI \geq 26 \text{ kg/m}^2$ ) men.

**Research Methods and Procedure:** Thirty subjects (forskolin,  $n = 15$ ; placebo,  $n = 15$ ) were studied in a randomized, double-blind, placebo-controlled study for 12 weeks.

**Results:** Forskolin was shown to elicit favorable changes in body composition by significantly decreasing body fat percentage (BF%) and fat mass (FM) as determined by DXA compared with the placebo group ( $p \leq 0.05$ ). Additionally, forskolin administration resulted in a change in bone mass for the 12-week trial compared with the placebo group ( $p \leq 0.05$ ). There was a trend toward a significant increase for lean body mass in the forskolin group compared with the placebo group ( $p = 0.097$ ). Serum free testosterone levels were significantly increased in the forskolin group compared with the placebo group ( $p \leq 0.05$ ). The actual change in serum total testosterone concentration was not significantly different among groups, but it increased  $16.77 \pm 33.77\%$  in the forskolin group compared with a decrease of  $1.08 \pm 18.35\%$  in the placebo group.

**Discussion:** Oral ingestion of forskolin (250 mg of 10% forskolin extract twice a day) for a 12-week period was

shown to favorably alter body composition while concurrently increasing bone mass and serum free testosterone levels in overweight and obese men. The results indicate that forskolin is a possible therapeutic agent for the management and treatment of obesity.

**Key words:** testosterone, DXA, fat mass, lean body mass

## Introduction

Obesity results from consuming more energy than is expended or from placing the body in a positive energy balance (1). Causes of obesity are extremely complex and multifaceted; different influences include genetic and environmental elements. Increasingly, obesity is becoming highly resistant to treatment in most individuals because of this myriad of contributing factors. While this concept of energy balance to maintain weight is easy to understand and correct in theory, the application of this in an uncontrolled environment for most individuals, especially those who are already obese, is extremely difficult, if not impossible. Also, because of advances in technology, physical activity of any kind, if not during leisure time, is almost nonexistent. Poor or no adherence to proper diet and decreased physical activity levels can be expected, especially in chronically sedentary individuals. Because of this, some form of pharmacological or supplemental treatment to aid in weight loss and/or positively alter body composition is desperately needed.

Men with hypogonadism have alterations in body composition, including increases in percentage body fat, changes in adipose tissue distribution, and reduction in muscle mass (2,3). Additionally, BMI, fat mass, waist circumference, and insulin resistance are all negatively correlated with sexual hormone levels in both men and women (2).

A potential supplemental aid for obesity and the aforementioned hormonal deficiencies is a compound containing the herbal extract forskolin. Forskolin is an extract from the

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roots of the *Coleus forskohlii* plant. *C. forskohlii* is a perennial herb with fleshy fibrous roots and is a member of the mint family of plants. It grows in the wild in warm subtropical temperate areas such as India, Burma, and Thailand. Research into the medicinal value of extracted forskolin began in the early- to mid-1980s and was primarily used as an agent to help a number of cardiovascular disease conditions, mainly through a vasodilatory effect (4–8). This effect was accomplished by increasing adenylate cyclase activity within the body.

Most of the research done with forskolin has looked at its effects either in animals in vitro or in vivo or preclinical studies with human tissue. Clinical research data are limited and are usually from an intravenous form of forskolin administered for short periods of time (9–12). However, there are several preliminary technical reports and published abstracts that have examined the effects of forskolin on body composition, primarily in women, although some men were also examined (85% of the subjects examined have been women) (13–16). Three of the four preliminary studies indicate that forskolin can have a positive effect on body composition, resulting in a significant reduction of body fat (13,14,16). The fourth study shows a significant reduction in body mass (15). No change in lean body mass is shown in any of the preliminary studies.

As a cyclic adenosine monophosphate (cAMP)<sup>1</sup> stimulator, forskolin leads to the production of the active form of hormone-sensitive lipase (HSL). HSL is directly involved in the mobilization of triglyceride stores that release free fatty acids to be used for fuel within the body. Because forskolin has a potentially favorable effect on body composition, it is important to examine its efficacy. Therefore, a study into the effects of forskolin on body compositional changes, endogenous testosterone, and any changes with regard to resting metabolic rate (RMR) is warranted.

Before forskolin, most weight loss aids used some form of adrenergic  $\alpha$ - and  $\beta$ -receptor agonists, such as ephedrine. However, compared with ephedrine and even more selective adrenergic receptor agonists, forskolin does not interact with adrenergic receptors ( $\alpha 1$ ,  $\alpha 2$ ,  $\beta 1$ , and  $\beta 2$  receptors) and, thus, does not result in excessive stimulation of cardiac tissue and does not raise blood pressure (5,17). Therefore, forskolin is not a sympathomimetic drug; it exhibits a vasodilatory effect, and a decrease in blood pressure is expected (18). Also as a postreceptor agent, adrenergic receptors should not down-regulate over time; thus, a diminished lipolytic effect should not occur. Therefore, forskolin could

potentially be used for long periods of time with no diminished lipolytic effects.

When evaluating body composition, it is also important to focus on ways to increase lean body mass (LBM) and not just reduction in fat mass. A critical component of LBM maintenance is having a sufficient supply of endogenous testosterone. Testosterone is the end product of a number of hormonal reactions. Gonadotropin-releasing hormone is secreted by the hypothalamus and controls the pulsatile secretion of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) by the anterior pituitary (19,20). LH regulates the production and secretion of testosterone by the Leydig cells of the testes, and FSH stimulates spermatogenesis (19).

Forskolin may have a favorable effect on enhancing serum testosterone levels through the theorized potential influence on cAMP. Because LH exerts its effects on Leydig cells of the testicles (stimulating production of testosterone) through cAMP, one may expect an increased level of endogenous testosterone with the use of this compound as well (21,22). Through enhanced natural testosterone production caused by cAMP accumulation, the preservation of LBM along with concurrent reductions in body fat could be expected (23). The predominately female preliminary studies have shown that LBM can be preserved (13–16). Therefore, one of the hypotheses in our study, which only consisted of men, was that LBM would increase significantly in the forskolin compared with the placebo group because of significant increases in endogenous testosterone levels.

Forskolin has been shown to have a positive inotropic and vasodilatory effect in clinical human studies. However, no research has been conducted on forskolin's effect on endogenous testosterone levels or potential effect on RMR in human clinical studies conducted exclusively in men. It is important to determine whether a relationship exists between positive changes in body composition through changes in RMR and changes in endogenous testosterone levels through oral supplementation of a capsule containing forskolin. The primary objectives of this study were to determine whether 1) forskolin administration (250 mg of 10% forskolin extract twice a day) results in fat loss and muscle gain; 2) forskolin administration results in higher endogenous testosterone levels; 3) forskolin has a positive effect on increasing RMR; and 4) forskolin administration results in lower systemic blood pressure.

## Research Methods and Procedures

Subjects were assessed for any potential physiological changes associated with forskolin supplementation a total of three times (pre, mid, post) during a 12-week trial period. This study was a double-blind, placebo-controlled clinical trial. Baseline values consisted of a blood draw for enzyme-linked immunosorbent assay (ELISA) analysis of total testosterone and free testosterone concentration in serum,

<sup>1</sup> Nonstandard abbreviations: cAMP, cyclic adenosine monophosphate; RMR, resting metabolic rate; HSL, hormone-sensitive lipase; LBM, lean body mass; LH, luteinizing hormone; FSH, follicle-stimulating hormone; ELISA, enzyme-linked immunosorbent assay.

**Table 1.** Body composition values including body weight, LBM, and fat mass at each time-point

	Pre	Mid	Post	Change (pre – post)	Percent change (pre – post)
<b>Forskolin</b>					
Body weight (kg)	103.98 ± 14.89	104.23 ± 15.04	103.91 ± 15.06	−0.07 ± 2.39	−0.08 ± 2.44
LBM (kg)	63.61 ± 5.94		67.32 ± 8.29 <sup>†</sup>	3.71 ± 4.07	5.65 ± 6.32
Fat mass (kg)	37.43 ± 12.65		32.91 ± 11.29 <sup>†</sup>	−4.52 ± 5.74*	−11.23 ± 13.20*
Bone mass (kg)	3.41 ± 0.43		3.68 ± 0.43 <sup>†</sup>	0.27 ± 0.31*	8.63 ± 10.46
<b>Placebo</b>					
Bodyweight (kg)	100.95 ± 9.30	102.09 ± 9.75	102.15 ± 9.65	1.20 ± 2.33	1.20 ± 2.35
LBM (kg)	61.82 ± 6.44		63.39 ± 7.07 <sup>†</sup>	1.57 ± 2.56	2.56 ± 4.39
Fat mass (kg)	35.65 ± 9.99		35.14 ± 10.56	−0.51 ± 1.91	−1.73 ± 5.64
Bone mass (kg)	3.41 ± 0.55		3.60 ± 0.51	0.20 ± 0.53	7.46 ± 18.78

The actual change from pre- to post-measurement and the percent change are also included.

All values are presented as means ± SD.

\* Significant difference between groups and † significant difference within groups across time ( $p \leq 0.05$ ).

RMR measurement, body composition assessment through DXA, body circumference measurements, blood pressure assessment, and dietary recall. At the midpoint of the trial (6 weeks), a blood draw, blood pressure assessment, and dietary recall were completed again. Finally, at the end of the 12-week trial period, values were recorded using the same procedures as those used for baseline assessment.

### Subjects

The subject population consisted of 30 men who were overweight/obese ( $BMI \geq 26 \text{ kg/m}^2$ ). Of the 30 individuals recruited, 15 were randomly assigned to receive forskolin supplementation for the 12-week trial period ( $n = 15$ ), whereas the other 15 subjects were assigned a placebo ( $n = 15$ ). The average age, BMI, and body fat percent were  $24.4 \pm 5.9$  years,  $32.5 \pm 4.1 \text{ kg/m}^2$ , and  $35.2 \pm 8.3\%$  for the forskolin group and  $28.7 \pm 8.6$  years,  $32.6 \pm 3.8 \text{ kg/m}^2$ , and  $35.0 \pm 7.3\%$  for the placebo group, respectively (other subject demographics are presented in Table 1). To meet the eligibility requirements for this study, individuals had to be overweight/obese ( $BMI \geq 26 \text{ kg/m}^2$ ), be non-active sedentary individuals, taking no antihypertensive and/or antiasthmatic medication, and must not have asthma, low blood pressure, or gastric ulcers. Before participating in the study, participants were informed of all potential risks and procedures involved with the study. Subjects signed an informed consent in concordance with the requirements of the University of Kansas Human Subjects Committee Review Board.

### Determining Body Composition

**DXA Body Fat Percent Evaluation.** Body composition was evaluated using DXA scans (Lunar Prodigy; GE Lunar,

Madison, WI). Subjects laid motionless for ~30 minutes while the DXA machine emitted alternating high and low energy X-rays over their entire bodies. DXA data acquisition is based on the differences between bone and soft tissue attenuation at the high and low X-ray levels. The raw scan data, which includes tissue and bone, were captured and sent to a computer. An algorithm interpreted each pixel and created an image and quantitative measurement of mass for both bone and other body tissues. Fat mass, LBM, and bone mass were collected directly from DXA.

**Body Circumference Measurement.** Body composition was determined by measuring two sites on the subject using an anthropometric tape measure. These measurements were done at the neck and abdomen and were repeated three times to increase reliability. These numbers were evaluated, along with the subject's height, using a known equation [ $\text{percent body fat} = (0.771 \times \text{abdominal} - \text{neck circumference in cm}) - (0.132 \times \text{height}) + 4.29$ ] to determine body fat percentage.

### Blood Pressure Determination

Blood pressure was determined using a stethoscope, a manometer, and inflatable cuff. All readings were taken on the subject's left arm after the subject had remained seating for at least 5 minutes.

### Blood Draw

Five milliliters of whole blood was collected from a vein in the forearm of each subject and was used to measure serum total and free testosterone levels. This blood was collected in a 5-mL glass Vacutainer tube. Serum was separated from blood using a centrifuge. Serum was placed

in a cryotube and stored at a temperature of  $-80^{\circ}\text{C}$  until the samples underwent ELISA analysis.

#### **Determining Endogenous Testosterone Levels**

**Total Testosterone ELISA.** Total testosterone levels were quantified using an ELISA procedure developed by ALPCO Diagnostics (catalog 020-DR-1559; Windham, NH). Approximately  $25\ \mu\text{L}$  of standards and serum samples were placed into each of the 96 antibody-coated wells (all values were measured in triplicate). Next,  $200\ \mu\text{L}$  of enzyme-conjugate solution was placed into each well. The plate was mixed thoroughly for 10 seconds and allowed to incubate for 60 minutes at room temperature uncovered. The contents of the well were briefly shaken out and washed three times with diluted wash solution ( $400\ \mu\text{L}/\text{well}$ ) using an automated plate washer (EL  $\times 405$  Automated Plate Washer; Bio-Tek Instruments, Winooski, VT). Then,  $200\ \mu\text{L}$  of substrate solution was placed into each well. After that was completed, the plate was allowed to incubate at room temperature for 15 minutes. Then,  $100\ \mu\text{L}$  of stop solution was placed into each well. Finally, absorbency was measured at 450 nm using a 96-well spectrophotometer microplate reader ( $\mu\text{Quant}$  Universal Microplate Spectrophotometer; Bio-Tek Instruments).

**Free Testosterone ELISA.** Free-testosterone levels were quantified using an ELISA procedure developed by ALPCO Diagnostics (020-DR-2924). First,  $50\ \mu\text{L}$  free testosterone standards, controls, and samples were dispensed into appropriate wells. Then,  $50\ \mu\text{L}$  of enzyme conjugate was dispensed into each well and mixed thoroughly for 10 seconds. The plate was incubated for 60 minutes at  $37^{\circ}\text{C}$ . After incubation, the contents of the wells were shaken out and rinsed five times using an automated plate washer (EL  $\times 405$  Automated Plate Washer; Bio-Tek Instruments). Next,  $100\ \mu\text{L}$  of substrate solution was added to each well and allowed to incubate for 15 minutes at room temperature while covered. Then,  $100\ \mu\text{L}$  of stop solution was added to each well, and absorbance was measured at 450 nm using a 96-well spectrophotometer microplate reader ( $\mu\text{Quant}$  Universal Microplate Spectrophotometer; Bio-Tek Instruments).

#### **RMR Testing**

For RMR testing, subjects were required to report to the laboratory between 6:00 AM and 10:00 AM after a 12-hour fast and a 48-hour abstention from physical activity. Subjects rested quietly for 30 minutes in a darkened room. Subsequently, they were connected to a metabolic measurement system (CardioO@/CP/Max System; MedGraphics, Minneapolis, MN) using a mask for 30 minutes. Expired air was collected by the metabolic measurement system and analyzed for oxygen and carbon dioxide concentrations. Carbon dioxide and oxygen concentrations were analyzed to determine calories burned and estimate overall caloric expenditure at rest (RMR).

#### **Forskolin Administration**

Subjects reported to the laboratory on a weekly basis to receive the forskolin (Forslean; Sabina Corp., Piscataway, NJ) supplement ( $n = 15$ ; 250 mg of 10% forskolin extract twice a day) or placebo ( $n = 15$ ).

#### **Statistical Analysis**

The differences between body composition, RMR, blood pressure, and total and free testosterone concentrations in serum associated with subjects ingesting the forskolin and those taking a placebo were tested with ANOVA. A repeated measures ANOVA was conducted to determine any significant interactions between group and time. A one-way ANOVA was used to assess changes across time. Follow-up analysis was performed to determine differences among groups at each time-point. Means and SDs were calculated for all variables (body composition, total and free testosterone levels, blood pressure, and RMR). The overall significance level was set at 5% ( $\alpha = 0.05$ ). The SPSS software program version 11.0 (SPSS, Chicago, IL) was used to perform the statistical analysis.

## **Results**

#### **Body Composition Analysis**

The reproducibility of measurements was determined through the quality control measurements obtained from the phantom calibration standard provided by GE Medical (Madison, WI). Over the course of the study, the coefficient of variation trend summary for the quality assurance measurements was 0.07%.

**DXA Body Fat Percent.** The forskolin group had a significant decrease in body fat percent from baseline ( $35.17 \pm 8.03\%$ ) to final measurement ( $31.03 \pm 7.96\%$ ). The placebo group showed no significant difference in body fat percent from baseline to final measurement. The actual change in body fat percent from before to after the study ( $-4.14 \pm 4.47\%$  vs.  $-0.96 \pm 1.66\%$  for forskolin vs. placebo, respectively) was shown to be significantly different among groups ( $p \leq 0.05$ ).

**Fat Mass.** After the 12-week trial period, fat mass decreased significantly in the forskolin group ( $p \leq 0.05$ ) with no change occurring in the placebo group. Additionally, the actual change in fat mass from before to after showed a significant difference among groups ( $p \leq 0.05$ ; Table 1).

**LBM.** After the 12-week trial period, LBM increased significantly in both groups ( $p \leq 0.05$ ). No significant differences were shown among groups at either pre- or post-time-points. However, the actual change in LBM from baseline to final measurements revealed a trend toward significance among groups ( $p = 0.097$ ; Table 1).

**Bone Mass.** No significant differences were shown among groups at either pre- or post-time-points. Follow-up analysis showed a significant increase from pre- to post-

**Table 2.** Total testosterone and free testosterone values at each time-point

	Pre	Mid	Post	Change (pre – post)	Percent change (pre – post)
<b>Forskolin</b>					
Total testosterone (ng/mL)	5.06 ± 1.21*	5.27 ± 1.03*	5.75 ± 1.50*	0.69 ± 1.26	16.77 ± 33.77
Free testosterone (pg/mL)	15.90 ± 13.39	15.67 ± 13.68	16.36 ± 13.32	0.46 ± 0.86*	3.47 ± 8.10
<b>Placebo</b>					
Total testosterone (ng/mL)	4.12 ± 0.82	3.97 ± 0.85	4.00 ± 0.89	–0.11 ± 0.95	–1.08 ± 18.35
Free testosterone (pg/mL)	13.28 ± 7.26	12.28 ± 7.44	12.77 ± 7.30	–0.51 ± 1.04	–4.11 ± 11.48

The actual change from pre- to post-measurement and the percent change are also included.

All values are presented as means ± SD.

\* Significance between groups ( $p \leq 0.05$ ).

values for total BM in the forskolin group ( $p \leq 0.05$ ). The actual change in BM from baseline to final measurements was significantly different among groups ( $p \leq 0.05$ ; Table 1).

**Body Circumference Analysis of Body Fat Percent.** There was a trend toward a significant interaction across time among groups ( $p = 0.089$ ). Follow-up analysis revealed no significant difference within groups across time, but a trend toward significance existed from pre- and post-time-points within the forskolin group for a decrease in percentage body fat ( $p = 0.061$ ). The coefficient of variation was 1% for test-retest reliability values.

**Body Weight.** No significant differences were found for the actual change in body weight from pre- to post-measurements. Overall, the forskolin group lost  $0.07 \pm 2.39$  kg of body weight compared with the placebo group, which actually gained  $1.20 \pm 2.33$  kg (Table 1).

### Endogenous Testosterone

**Total Testosterone.** There was a significant difference among groups at pre-, mid-, and post-time-points for total testosterone ( $p \leq 0.05$ ). A trend toward a significant increase in total testosterone existed within the forskolin group from pre- and post-time periods ( $p = 0.051$ ). The percentage change for total testosterone for all time-points can be seen in Table 2. Finally, a trend toward a significant increase for the forskolin group existed in regard to the actual change in total testosterone from pre- to post-time-points ( $p = 0.057$ ). The coefficient of variations was 7% for intra-assay and 3% for inter-assay.

**Free Testosterone.** There was a significant change across time for free testosterone in the forskolin group ( $p \leq 0.05$ ). There was no significant difference among groups for free testosterone for all time-points. When the actual change in free testosterone was examined, there was a significant increase in the forskolin group from pre- to post-time-

points ( $p \leq 0.05$ ). The coefficient of variations was 9% for intra-assay and 8% for inter-assay.

### RMR

The mean RMR at baseline for the forskolin group was  $2167.40 \pm 681.64$  vs.  $1680.13 \pm 330.72$  kcal/d for the placebo group. The final values for RMR increased to  $2182.13 \pm 610.50$  kcal/d for forskolin and increased to  $1789.07 \pm 402.40$  kcal/d for placebo. No significant difference was observed among groups or across time within each group. The coefficient of variation was 9% for test-retest reliability values.

### Blood Pressure

**Systolic Blood Pressure.** Systolic blood pressure changes were not shown to be statistically significant across time, and there were no significant differences among groups at either pre-, mid-, or post-measurements. All systolic blood pressure values are shown in Table 3. The coefficient of variation was 2% for test-retest reliability values.

**Diastolic Blood Pressure.** Mean diastolic blood pressure values obtained during the 12-week trial period are shown in Table 3. No significant differences or trends toward significance were observed for diastolic blood pressure or in the actual change in diastolic blood pressure over the 12-week period. The coefficient of variation was 2% for test-retest reliability values.

### Dietary Recall

The mean daily caloric intake, obtained through dietary recall, was  $2353.87 \pm 500.12$  kcal/d for forskolin vs.  $2461.43 \pm 471.29$  kcal/d for placebo. The post-values for daily caloric intake were  $2386.92 \pm 483.69$  kcal/d for forskolin vs.  $2558.09 \pm 579.83$  kcal/d for placebo. There were no significant differences across time or among groups for daily caloric intake as obtained with the dietary recall.

**Table 3.** Systolic and diastolic blood pressure values at each time-point

	Pre	Mid	Post	Change (pre – post)
<b>Forskolin</b>				
Systolic (mm Hg)	132.73 ± 13.83	125.87 ± 7.99	126.47 ± 5.25	-6.27 ± 12.83
Diastolic (mm Hg)	82.47 ± 6.33	83.40 ± 5.25	84.07 ± 4.25	1.60 ± 7.60
<b>Placebo</b>				
Systolic (mm Hg)	129.87 ± 11.69	123.60 ± 11.61	125.20 ± 9.48	-4.67 ± 9.47
Diastolic (mm Hg)	83.33 ± 6.94	83.33 ± 6.26	84.27 ± 7.91	0.93 ± 7.92

The actual change from pre- to post-measurement is included.  
All values are presented as means ± SD.

### Discussion

The purpose of this study was to examine the effects of oral forskolin consumption on body composition, serum testosterone concentration, RMR, and blood pressure in an overweight/obese male population. The results of this study show that forskolin promotes favorable changes in body composition by significantly decreasing percentage body fat and fat mass and increasing bone mass, as determined by DXA ( $p \leq 0.05$ ). The forskolin group lost  $4.52 \pm 5.74$  kg of fat mass while concurrently gaining  $3.71 \pm 4.07$  kg of LBM. The placebo group lost  $0.51 \pm 1.91$  kg of fat mass and gained  $1.57 \pm 2.56$  kg of LBM. The increase in bone mass was significantly different among groups for forskolin ( $0.27 \pm 0.31$  kg forskolin vs.  $0.20 \pm 0.53$  kg placebo). Forskolin also evoked a significant increase with regard to the change in free testosterone concentration among groups over the 12-week trial period ( $p \leq 0.05$ ). A significant difference also existed among groups for total testosterone at baseline, mid-, and final time-points ( $p \leq 0.05$ ).

#### Body Fat

This is the first study into forskolin's direct effect on body composition in vivo exclusively in men (other studies have looked primarily at women); however, forskolin has been studied for its lipolytic effect in vitro. Litosch et al. (17) showed that forskolin stimulated lipolysis in adipose tissue to a maximum of  $30.01 \pm 3.42$  (expressed as micromoles of glycerol per  $10^7$  cells per 2 hours) in obese men and  $50.50 \pm 6.85$  in obese women before weight loss was induced using a surgical gastric binding technique. After weight loss, forskolin increased lipolysis in adipose tissue to  $28.43 \pm 4.49$  and  $26.44 \pm 3.25$  in obese men and women, respectively. As noted before, this study was in vitro, making comparisons with the current study limited. The current study revealed that forskolin induced fat loss as measured by DXA. The decrease in body fat percent that occurred

across the 12-week trial period was shown to be significant in the forskolin group. These findings are important because they show that forskolin can induce whole body reductions in body fat percent. Previous research, such as that done by Litosch et al., focused on lipolysis at a subcellular level. This study shows that forskolin's lipolytic effect can be generated within the whole organism as well. The change in fat mass for the forskolin group could be explained by the lipolytic effects shown by Litosch et al. (17). Also, work done by Ho and Shi (24) and Lichey et al. (25) showed that forskolin could induce lipolysis and free fatty acid release from stored triglycerides in rat adipose tissue. In both of these studies, the primary mechanism for lipolysis was the ability of forskolin to dramatically increase cAMP levels within adipose tissue in vitro. More specifically, as cAMP accumulated within the tissue, protein kinase A was activated. This in turn activates HSL, which breaks down stored triglycerides for energy and increases free fatty acid release. Therefore, the change in fat mass as shown by the forskolin group ( $-4.52 \pm 5.74$  kg) could be induced by direct free fatty acid release within adipose tissue through cAMP accumulation and directly induce fat loss.

When evaluating body composition, it is critical to explore the actual change in fat mass and LBM that comprises any change in body fat percent. The average change in weight for the subjects treated with forskolin was  $-0.07 \pm 2.39$  kg. However, the forskolin group lost  $4.52 \pm 5.74$  kg of fat mass ( $p \leq 0.05$ ) while concurrently gaining a non-significant  $3.71 \pm 4.07$  kg of LBM. There was no change in the subjects' diet as indicated by dietary recall analysis of caloric intake. In contrast to the forskolin group, the placebo group showed an average weight gain of  $1.20 \pm 2.33$  kg when the actual change across the 12-week trial period was evaluated (Table 1). The placebo group gained an average of  $1.57 \pm 2.56$  kg of LBM but only lost  $0.51 \pm 1.91$  kg of fat mass (both non-significant changes). The change in body

fat percent and fat mass was significantly different among groups during the 12-week trial ( $p \leq 0.05$ ).

Results of the RMR testing would suggest that forskolin directly activated free fatty acid release, and there was a minimal non-significant increase in RMR through increased thyroid hormone activity. Follow-up tests revealed no significant changes in RMR within either group, and there was no significant interaction seen across time among groups. This would refute the claims that forskolin can increase metabolic rate through increased thyroid hormone levels or at least not to the point where a significant increase in metabolic rate could be quantified (26). Therefore, the reductions in fat mass as shown in the forskolin group may be attributed more toward forskolin's ability to directly activate adenylate cyclase within adipose tissue, resulting in a greater release of free fatty acids.

### **Bone Mass**

In this study, forskolin was shown to increase bone mass. A significant interaction existed for bone mass across time among groups ( $p \leq 0.05$ ). Follow-up analysis showed a significant increase from pre- to post-values for total bone mass in the forskolin group ( $p \leq 0.05$ ). Finally, the actual change in bone mass from baseline to final measurements was significantly different among groups ( $p \leq 0.05$ ).

Direct comparative literature is nonexistent regarding forskolin's effect on bone mass; this study is the first to explore forskolin's effect on bone mass with humans in vivo. However, it is known that parathyroid hormone increases cAMP and calcium levels in bone cells, which can be used by osteoclasts in bone restoration and bone formation through osteoblast activation (27). The drug teriparatide has been shown to directly activate adenylate cyclase and increase cAMP and calcium similar to parathyroid hormone. In fact, a clinical study conducted by Neer et al. (28) showed that teriparatide increased bone mineral density in the lumbar spine and hip by 9.7% and 2.6%, respectively. Therefore, because of forskolin's ability to stimulate adenylate cyclase in a variety of tissues, the increase in bone mass that occurred during this study shows that forskolin can improve bone mass in overweight and obese men.

### **Testosterone and LBM**

Testosterone, as a hormone, is involved in the promotion of muscle mass and reducing fat mass (3,8). Men with hypogonadism have negative alterations in body composition, such as decreased muscle mass, increased percent body fat, and alterations in body fat distribution (8). Both groups significantly increased LBM over the course of the study ( $p < 0.05$ ); however, there was no significant difference ( $p = 0.097$ ) between the forskolin group ( $3.71 \pm 4.07$  kg increase) and the placebo group ( $1.57 \pm 2.56$  kg increase; Table 1). This is the first study to examine forskolin's effect on serum total testosterone levels in vivo. Serum total

testosterone levels were shown to be significantly different among groups for measurements taken before, during, and after the trial ( $p \leq 0.05$ ). Because the forskolin group had significantly higher serum total testosterone levels at baseline ( $p \leq 0.05$ ), it is critical to examine the percent difference that occurred between initial and final measurements for both groups to account for this difference in initial values. The percentage difference among groups that occurred between the initial and final measurements for total testosterone was  $16.77 \pm 33.77\%$  for forskolin vs.  $-1.08 \pm 18.35\%$  for placebo (Table 2).

When evaluating testosterone levels, it is also important to measure free testosterone levels. Free testosterone is the component of total testosterone that is not bound to either albumin or sex hormone bound globulin. Of the two, albumin bound testosterone may dissociate from albumin and, thus, be able to elicit a physiological effect at a cellular level within tissue such as muscle (29). Testosterone bound to sex hormone bound globulin cannot dissociate, and, thus, its active properties are nullified (30). Free testosterone is the most potent or most readily active component of total testosterone, and, therefore, quantifying it within serum is critically important to show the physiological active component on the hormone testosterone. In this study, the change in free testosterone that occurred across the 12-week trial period was shown to be significantly different among groups ( $p \leq 0.05$ ; Table 2). It is important to understand that an increase in testosterone can elicit reductions in fat mass (3,8). In this study, the increase shown for serum total testosterone and free testosterone may be another and/or a contributing factor for the reductions in body fat percent and fat mass seen in the forskolin-treated group.

### **Blood Pressure**

Systolic blood pressure changes were not shown to be statistically significant across time, and there were no significant differences among groups at either initial, mid-, or final measurements. In fact, within both groups, a trend toward a significant reduction in systolic blood pressure was shown ( $p = 0.079$  for forskolin vs.  $p = 0.077$  for placebo). No significant difference or trends toward significance were observed for diastolic blood pressure or in the actual change in diastolic blood pressure at any time-point.

The findings of this study are in contrast to early work examining forskolin's effect on the cardiovascular system, and more specifically, its potential hypotensive effects. Lindner et al. (7) showed that forskolin could increase the force of heart contraction and could lower blood pressure in animal models. Studies conducted by Dubey et al. (31) showed that coleonol (a diterpene isolated from the extract *C. forskohlii*) produced a well-marked and sustained hypotension in anesthetized cats in the dose range of 0.1 to 1.0 mg/kg. It is important to note that all of these hypotensive effects occurred within animal models. This study did not

show a significant difference among groups for reductions in either systolic or diastolic blood pressure within humans. This could be caused by any number of variables such as physiological variations within animal models compared with humans, differences in the dose of forskolin, and differences in the form of forskolin used (i.e., colforsin daropate vs. pure *c. forskohlii* extract).

In summary, this study shows that forskolin causes positive changes in body composition in overweight and obese adult men. One of the potential explanations for the decrease in fat mass and body fat percentage may have occurred through adenylate cyclase activation and, thus, cAMP accumulation within adipose tissue, which stimulated free fatty acid release and lipolysis. RMR did not significantly change across time during this study, implying that, in a 12-week study, forskolin did not increase metabolic rate.

These findings are extremely important because of forskolin's mechanism of action. The majority of previous weight loss aids worked through adrenergic receptor activation. Adrenergic receptor activation can down-regulate over time and result in diminished lipolytic effects. Forskolin bypasses the adrenergic activation step and increases cAMP levels by either stimulating adenylate cyclase or by increasing the cAMP accumulating properties of catecholamines (32,33). Therefore, forskolin could possibly be used for long periods of time without diminished lipolytic effects in conjunction with increasing LBM. Further research into the long-term effects of forskolin is needed to accurately assess this theory.

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### References

1. **Van Itallie TB.** Obesity: adverse effects on health and longevity. *Am J Clin Nutr.* 1979;32:2723–33.
2. **McCann D, Kirkish L.** Evaluation of free testosterone in serum. *J Clin Immunoassay.* 1985;8:234–6.
3. **Waters DL, Yau CL, Montoya GD, Baumgartner RN.** Serum sex hormones, IGF-1, and IGFBP3 exert a sexually dimorphic effect on lean body mass in aging. *J Gerontol A Biol Sci Med Sci.* 2003;58:648–52.
4. **Adnot S, Desmier M, Ferry N, Hanoune J, Sevenet T.** Forskolin (a powerful inhibitor of human platelet aggregation). *Biochem Pharmacol.* 1982;31:4017–4.
5. **Bristow MR, Ginsberg R, Strosberg A, Montgomery W, Minobe W.** Pharmacology and inotropic potential of forskolin in the human heart. *J Clin Invest.* 1984;74:212–23.
6. **de Souza NJ, Dohadwalla A, Reden J.** Forskolin: a labdane diterpenoid with antihypertensive, positive inotropic, platelet aggregation inhibitory, and adenylate cyclase. *Med Res Rev.* 1983;3:201–19.
7. **Lindner E, Dohadwalla AN, Bhattacharya BK.** Positive inotropic and blood pressure lowering activity of a diterpene derivative isolated from *Coleus forskohlii*: forskolin. *Arzneimittelforschung.* 1978;28:284–9.
8. **McNicholas TA, Dean JD, Mulder H, Carnegie C, Jones NA.** A novel testosterone gel formulation normalizes androgen levels in hypogonadal men, with improvements in body composition and sexual function. *BJU Int.* 2003;91:69–74.
9. **Hakkinen K, Kraemer W, Pakarinen A, et al.** Effects of heavy resistance training/power training on maximal strength, muscle morphology, and hormonal response patterns in 60–75-year-old men and women. *Can J Appl Physiol.* 2002;27:213–31.
10. **Hibino N, Kawai A, Uchikawa S, et al.** Cardiovascular effects of colforsin daropate hydrochloride for acute heart failure after open heart surgery. *Kyobu Geka.* 2001;54:1016–9.
11. **Iranami H, Okamoto K, Kimoto Y, Kakutani T, Hatano Y.** Use of colforsin dalopate following cardiac surgery in a neonate. *Anesthesiology.* 2002;97:503–4.
12. **Paulson JD, Keller DW, Wiest WG, Warren JC.** Free testosterone concentration in serum: elevation is the hallmark of hirsutism. *Am J Obstet Gynecol.* 1977;128:851–7.
13. **Bhagwat AM, Joshi B, Joshi AS, Jain A, Sawant N.** A Randomized Double-blinded Multicenter Phase III Clinical Trial to Investigate the Efficacy and Safety of ForsLean in Increasing Lean Body Mass. Mumbai, India: Patel Research Center for Chemistry and Biological Sciences; 2004.
14. **Badmaev V, Majeed M, Conte AA, Parker JE.** Diterpene forskolin (*Coleus forskohlii* Benth.): a possible new compound for reduction of body weight by increasing lean body mass. *NutraCos.* 2002;1:6–7.
15. **Kreider R, Henderson S, Magu B, et al.** Effects of *Coleus forskohlii* supplementation on body composition and markers of health in sedentary overweight females. *FASEB J.* 2002;16:59.
16. **Tsuguyoshi A.** *Clinical Report on Root Extract of Perilla Plant (Coleus forskohlii) ForsLean in Reducing Body Fat.* Tokyo, Japan: Asano Institute; 2004.
17. **Litosch I, Hudson TH, Mills I, Li SY, Fain JN.** Forskolin as an activator of cyclic AMP accumulation and lipolysis in rat adipocytes. *Molec Pharmacol.* 1982;22:109–15.
18. **Wagner DR, Heyward VH.** Techniques of body composition assessment: a review of laboratory and field methods. *Res Qtlly Exerc Sport.* 1999;70:135–49.
19. **Barry Z, Chen H.** Regulation of leydig cell steroidogenic function during aging. *Biol Reproduction.* 2000;63:977–81.
20. **Bondanelli M, Ambrosio M, Margutti A, et al.** Activation of the somatotrophic axis by testosterone in adult men: evidence for a role of hypothalamic growth hormone-releasing hormone. *Neuroendocrinology.* 2003;77:380–7.
21. **Valenti S, Guido R, Giusti M, Giordano G.** In vitro acute and prolonged effects of melatonin on purified rat Leydig cell steroidogenesis and adenosine 3',5'-monophosphate production. *Endocrinology.* 1995;136:5357–62.

22. **Valenti S, Fazzuoli L, Giordano G, Giusti M.** Changes in binding of iodomelatonin to membranes of Leydig cells and steroidogenesis after prolonged in vitro exposure to melatonin. *Int J Androl.* 2001;24:80–6.
23. **Marcus GJ, Durnford R.** Estradiol assay by microtitre plate enzyme immunoassay. *J Steroid Biochem.* 1988;29:207–12.
24. **Ho R, Shi Q.** Forskolin as a novel lipolytic agent. *Biochem Biophys Res Commun.* 1982;107:157–64.
25. **Lichey I, Friedrich T, Priesnitz M, Biamino G, Usinger P, Huckauf H.** Effect of forskolin on methacholine-induced bronchoconstriction in extrinsic asthmatics. *Lancet.* 1984;2:167.
26. **Kasai K, Suzuki Y, Hiraiwa M, et al.** Forsolin stimulation of adenylate cyclase in human thyroid membranes. *Acta Endocrinol.* 1985;108:200–5.
27. **Watrous DA, Andrews BS.** The metabolism and immunology of bone. *Semin Arthritis Rheum.* 1989;19:45–65.
28. **Neer R, Arnaud CD, Zanchetta JR, et al.** Effect of parathyroid hormone (1–34) on fractures and bone mineral density in postmenopausal women with osteoporosis. *N Engl J Med.* 2001;344:1434–41.
29. **Pardridge WM.** Serum bioavailability of sex steroid hormones. *Clin Endocrinol Metab.* 1986;15:259–78.
30. **Selby C.** Sex hormone binding globulin: origin, function and clinical significance. *Ann Clin Biochem.* 1990;27:532–41.
31. **Dubey MP, Srimal RC, Nityanand S, Dhawan BN.** Pharmacological studies on coleonol, a hypotensive diterpene from *Coleus forskohlii*. *J. Ethnopharmacol.* 1981;3:1–13.
32. **Hoffstedt J, Arner P, Schalling M, et al.** A common hormone-sensitive lipase i6 gene polymorphism is associated with decreased human adipocyte lipolytic function. *Diabetes.* 2001;50:2410–3.
33. **Lang JH, Zhu L, Sun ZJ, Chen J.** Estrogen levels and estrogen receptors in patients with stress urinary incontinence and pelvic organ prolapse. *Int J Gynecol Obstet.* 2003;80:35–9.