

Diabetes-Ameliorating Effects of Fermented Red Ginseng and Causal Effects on Hormonal Interactions: Testing the Hypothesis by Multiple Group Path Analysis

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ABSTRACT Although diagnostic criteria for metabolic syndrome (MtS) vary among various health professionals and organizations, blood glucose dysregulation and insulin resistance are common to all definitions. Red ginseng is beneficial for glucose regulation and insulin sensitivity but the mechanism is not yet elucidated. Ginsenosides Rh1 and Rg3 act as ligands of the estrogen receptor, and Rh2 and compound K act as ligands of the glucocorticoid receptors, which may influence the diabetes markers. The objective of this study was to test the hypothesis that there are significant causal relationships among diabetes-related markers and several hormones, and assess whether or not the consumption of fermented red ginseng (FRG) influences these causal relationships by multiple group path analysis and conventional statistical analyses. The 93 postmenopausal women were randomly divided into two groups for a double-blind trial. FRG powder and placebo were provided for 2 weeks. The data were analyzed by multiple group path analysis and the mean between groups were compared. The model's goodness of fit was excellent, with a root mean square error of approximation of 0.00, and comparative fit index of 1.00. The FRG group exhibited significantly increased levels of dehydroepiandrosterone sulfate (DHEAS), growth hormone (GH), and estradiol (E2), and they exhibited decreased levels of glycosylated hemoglobin (HbA1c), insulin, and homeostatic model assessment of insulin resistance. With regard to the hypothesis, the blood glucose lowering effects of FRG were due to the negative effects of aldosterone and increased GH, which was associated with DHEAS and E2. Even though the differences of variables between both groups were small, the total effects of these variables may indicate beneficial changes for the prevention of diabetes in healthy postmenopausal women.

KEY WORDS: • blood glucose • compound K • DHEAS • estradiol • ginsenoside • HOMA-IR • insulin resistance • path model • Rg3

INTRODUCTION

THE WORLD HEALTH ORGANIZATION defines metabolic syndrome (MtS) as “glucose intolerance, impaired glucose tolerance or diabetes mellitus, and/or insulin resistance together with two or more of the following: elevated arterial pressure, elevated plasma triglycerides, central adiposity, microalbuminuria, and several other components (*e.g.*, hyperuricemia, coagulation disorders, raised PAI-1, *etc.*).”¹

Although these diagnostic criteria include a broad spectrum of definitions and points of emphasis,² several major components of MtS diagnosis are shared among various professional bodies, especially, blood glucose and insulin resistance.

Studies have reported that the incidences of MtS in postmenopausal women were as high as 35.1% in Latin America,³ 35% in Portugal,⁴ 33% in the United States,⁵ and 27.3% in China.⁶ A low level of E2 is considered as a major cause of MtS, and estrogen hormone replacement therapy (HRT) decreases this risk in postmenopausal women. For example, postmenopausal women who undergo HRT show a 12.9% decrease in insulin resistance and a 35.8% reduction in the incidence of diabetes.⁷ However, a study by the Women's Health Initiative reported that HRT increases the risk of gynecological cancer and obesity and has a negligible beneficial effect on cardiovascular diseases.⁸ Given the severity of these HRT side effects, the study of estrogen mimics—including selective estrogen receptor modulators—has emerged as an important MtS treatment area.⁹ On the other hand, many studies have shown that the rate of onset of MtS is related not only to estrogen levels, but also to the levels of other hormones,¹⁰ which may be due to the broad and complicated interrelationships of the constituents of the endocrine system.

Article received 18 September, 2012. Revision accepted 24 February, 2013.

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Ginseng is one of the most popular herbal supplements in Asia, especially Korea, China, and Japan, and records document its use back to 2000 years.¹¹ In modern times, the popularity of ginseng has grown in western countries as well. For example, ginseng ranked as one of the top-10 selling herbal supplements in the United States in 2003.¹² The primary pharmacological components in ginseng are known as ginsenosides, chemicals that have a steroid skeleton. Studies have shown that ginsenosides, Rh1 and Rg3 act as ligands of the estrogen receptor (ER), and that Rh2 and compound K (CK) act as ligands of the glucocorticoid receptor (GR).^{13,14} Many studies have reported that red ginseng is beneficial for glucose regulation and insulin sensitivity.^{15,16} One possible hypothesis would be that ginsenosides can influence the variables of diabetes, and the functioning of the endocrine system. The first objective of this study was to test the hypothesis that there are significant causal relationships among diabetes-related markers and several hormones by a path analysis. The second objective was to test the hypothesis that consumption of fermented red ginseng (FRG) influences these causal relationships by multiple group path analysis and conventional statistical analyses.

MATERIALS AND METHODS

Participants and study design

This study was approved by the Institutional Review Board of Sahmyook University (Seoul, Korea). Women aged 50–73 years were recruited from several Catholic churches (Seoul, Korea). Participants with hypertension or diabetes and those taking prescription or other drugs were excluded. Dietary supplements were not allowed during the experimental period.

The 117 volunteers were randomly divided into two double-blinded groups. FRG powder was provided by Bifido Inc. (Gangwon-do, Korea). One group (the FRG group) took FRG capsules three times a day, each time taking a dose of three capsules (2.1 g/day) for 2 weeks. The other group took a placebo containing starch. The composition of the FRG capsules was crude saponin, 258.6 mg/g; Compound K, 57.05 mg/g; Rg3, 53.85 mg/g; Rh2, 11.97 mg/g; Rg2, 5.72 mg/g; Rh1, 2.99 mg/g; and Rb1, 0.023 mg/g. Blood samples, after 8 hours of fasting, were collected before and after the 2-week intake of either the FRG or the placebo capsules. The blood samples were collected between 8:00 a.m. and 10:00 a.m. Urine samples were collected 24 hours prior to collection of blood samples. All 117 women participated in the first blood sample collection, but only 93 women participated in the second blood sample collection (Fig. 1). Ninety of the 93 participants were postmenopausal, and three were perimenopausal.

Forty subjects were selected from both groups (20 subjects per group) after matching age, height, weight, and BMI and were further analyzed for several hormones (Table 1). All biochemical components were measured by the Green Cross Reference Lab (Gyeonggi-do, Korea). The analytical methods are shown in Appendix Table A1.

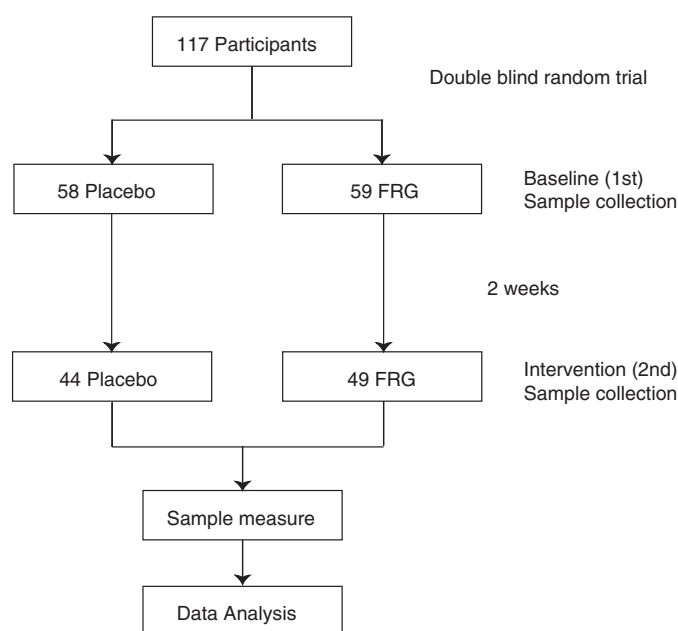


FIG. 1. Flowchart of this study.

Statistical analyses

Given that the production of the estrogen hormone slowly decreases during the first five postmenopausal years, the three perimenopausal women were included in this analysis. The baseline comparisons of both groups between the first baseline sample and the second postintervention values were compared by independent *t*-test. The means of the postintervention samples were also compared between the FRG group and the placebo group, with an analysis of covariance (ANCOVA) by SPSS 18.0 (SPSS, Inc., Chicago, IL, USA). The level of statistical significance was $P < .05$ and a statistical tendency was considered as $P < .1$. The outliers that were over three times the standard deviation were excluded in the estrogen, insulin, and homeostasis model of insulin resistance (HOMA-IR) variables. The HOMA-IR index was calculated as follows:

$$\text{HOMA-IR index} = \left[\frac{\text{fasting serum glucose (mM)} \times \text{fasting serum insulin } (\mu\text{U/mL})}{22.5} \right]$$

The unmeasured hormones of 53 participants, and some data that could not be measured due to the detection limitation of the instruments, were considered as random missing values. Following this, 10 data sets were generated using the multiple imputation method and then were analyzed by Mplus 6.11 (Muthén & Muthén, Los Angeles, CA, USA).

RESULTS

Anthropometric data

Table 2 shows the anthropometric variables of the participants. There was no significant difference between the FRG and the placebo groups in the mean values of age, weight, height, BMI, waist, and hip circumference.

TABLE 1. COMPARISON OF HORMONE AND DIABETES MARKERS BETWEEN FERMENTED RED GINSENG GROUP AND PLACEBO GROUP

Marker	Group (n)	2nd sample (mean $\pm$ SD) ^a	Δ [2nd–1st] (mean) ^b
Diabetes			
Glucose (mg/dL)	Placebo (44)	89.6 $\pm$ 6.8	2.2
	FRG (49)	87.4 $\pm$ 7.1	2.5
HbA1c (%)	Placebo (44)	5.8 $\pm$ 0.3**	0.1
	FRG (49)	5.7 $\pm$ 0.3**	0.1
HOMA-IR	Placebo (43)	1.4 $\pm$ 0.8*	0.2**
	FRG (48)	1.4 $\pm$ 0.7*	–0.1**
Insulin (μ U/mL)	Placebo (43)	6.4 $\pm$ 3.6**	1.0**
	FRG (48)	6.2 $\pm$ 2.9**	–0.2**
HPA axis			
ACTH (pg/mL)	Placebo (44)	20.4 $\pm$ 10.1	2.3
	FRG (49)	22.7 $\pm$ 11.8	0.9
ADH (pg/dL)	Placebo (20)	0.8 $\pm$ 0.2	–0.1
	FRG (20)	0.8 $\pm$ 0.2	0.0
CBG (ng/mL)	Placebo (20)	6178 $\pm$ 8294	2269
	FRG (20)	6293 $\pm$ 8469	2114
Cortisol (μ g/dL)	Placebo (44)	11.2 $\pm$ 3.5	–0.2
	FRG (49)	10.8 $\pm$ 3.8	0.9
Free cortisol (μ g/day)	Placebo (41)	22.5 $\pm$ 15.8	–5.5
	FRG (47)	19.8 $\pm$ 14.0	–2.1
CRH (ng/mL)	Placebo (20)	0.2 $\pm$ 0.1	0.0
	FRG (20)	0.5 $\pm$ 1.0	0.2
HPG axis			
E2 (pg/mL)	Placebo (19)	14.5 $\pm$ 7.3*	–7.4*
	FRG (19)	18.3 $\pm$ 7.3*	1.0*
FSH (mIU/mL)	Placebo (20)	74.0 $\pm$ 24.3	0.1
	FRG (20)	68.0 $\pm$ 18.5	2.4
LH (mIU/mL)	Placebo (20)	38.8 $\pm$ 14.8	1.0
	FRG (20)	30.3 $\pm$ 9.0	–0.3
HPS axis			
Aldosterone (ng/dL)	Placebo (20)	6.2 $\pm$ 3.5	0.2
	FRG (20)	8.4 $\pm$ 3.6	0.9
DHEAS (μ g/dL)	Placebo (44)	65.2 $\pm$ 31.4*	–6.2**
	FRG (49)	66.2 $\pm$ 29.1*	0.5**
GH (ng/mL)	Placebo (20)	0.9 $\pm$ 1.2	–0.4
	FRG (20)	1.8 $\pm$ 1.4	–0.1
IGF-1 (ng/mL)	Placebo (20)	144 $\pm$ 49	–2.3
	FRG (20)	144 $\pm$ 67	1.8

Data expressed as a mean SD.

Values are significantly different as indicated (* $P < .1$, ** $P < .05$, *** $P < .01$) by ^aanalysis of covariance or ^bindependent *t*-test.

ACTH, adrenocorticotrophic hormone; ADH, antidiuretic hormone; CBG, cortisol-binding globulin; CRH, corticotropin-releasing hormone; DHEAS, dehydroepiandrosterone sulfate; E2, estradiol; FRG, fermented red ginseng; FSH, follicle stimulating hormone; GH, growth hormone; HbA1c, glycosylated hemoglobin; HOMA-IR, homeostasis model of insulin resistance; HPA axis, hypothalamic–pituitary–adrenal axis; HPG axis, hypothalamic–pituitary–gonadal axis; HPS axis, hypothalamic–pituitary–somatotroph axis; IGF-1, insulin-like growth factor-1; LH, luteinizing hormone.

Mean

Table 1 shows the means of the variables from the intervention samples taken after 2 weeks, and the difference between the first baseline sample and the week 2 intervention sample. The results from the week 2 intervention sample show that the level of growth hormone (GH, 1.8 vs. 0.9 ng/mL), estradiol (E2, 18.3 vs. 14.5 pg/mL), and dehy-

TABLE 2. ANTHROPOMETRIC DATA OF PARTICIPANTS

	Control (placebo; n = 44)	FRG group (n = 49)
Age (years)	58.4 $\pm$ 5.9	58.4 $\pm$ 5.5
Weight (kg)	57.6 $\pm$ 6.6	57.1 $\pm$ 6.7
Height (cm)	156.5 $\pm$ 5.3	157.7 $\pm$ 5.3
BMI (kg/m ²)	23.6 $\pm$ 2.5	22.9 $\pm$ 2.4
Waist circumference (cm)	33.4 $\pm$ 2.3	32.8 $\pm$ 2.4
Hip circumference (cm)	37.4 $\pm$ 2.0	37.0 $\pm$ 1.9
Waist/hip ratio	0.9 $\pm$ 0.0	0.9 $\pm$ 0.0

Data are mean $\pm$ SD values.

BMI, body mass index.

droepiandrosteronesulphate (DHEAS, 66.2 vs. 65.2 μ g/dL) were significantly higher in the FRG group than in the placebo group, at $P < .1$. The mean values of insulin (6.2 vs. 6.4 μ U/mL), HbA1c (5.70 vs. 5.77%), and the HOMA-IR (1.36 vs. 1.42) indexes were significantly lower in the FRG group than in the placebo group.

When the first and second sample were compared, the values of E2 (1.0 vs. –7.4 pg/mL) and DHEAS (0.6 vs. –6.2 μ g/dL) were higher in the FRG group than in the placebo group, and the values of insulin (–0.2 vs. 1.0 μ U/mL), glycosylated hemoglobin (HbA1c, 0.06 vs. 0.09%), and HOMA-IR (–0.09 vs. 0.17) were significantly lower in the FRG group than in the placebo group, but the values of GH were not significantly different (Table 1).

Path model

In this model, all paths were established based on the results of precedents, which can be found in previously published studies, but these paths were not based on a strict statistical correlation.¹⁷ The studies that established the precedents for these path models are presented in Table 3,^{1,18–33} and the correlations are presented in Appendix Table A2.

Path analysis is a useful statistical analysis method, one that can analyze several causal relationships among several variables at the same time. Further, multiple group path analysis allows for the comparison of the path coefficients between two groups, followed by identifying statistical significances of the differences between two path coefficients. Therefore, the multiple group path analysis may be a useful tool for the analysis of the complicated interrelationship between hormonal and diabetes variables. One of the major purposes of this multiple group path model was to analyze whether there was invariance of path coefficients across two groups. The testing of equivalence was conducted in two steps. First, a baseline model was established, followed by equality constraints of the path coefficients of the two groups. The path coefficients of the baseline model are presented in Table 4, and the final path model after equality constraints test presented in Figure 2.

An unstandardized path coefficient shows that when a causative variable increases by a value of one unit, the resultant variable changes in value. In the baseline model (Table 4), when E2 increased by 1 pg/mL, GH increased to 0.160 ng/mL in the FRG group; whereas GH decreased to 0.301 ng/mL ($P = .02$) in the placebo group. To compare the

TABLE 3. DESCRIPTION OF PATHS IN PRESENT STUDY

Path	Contents	Reference
ACTH→aldosterone	When ACTH is treated to rat, the level of 11 β -hydroxylase, mRNA in the adrenal gland and in the cerebral cortex is increased, and aldosterone synthase mRNA increases in all central nerves except the cerebral cortex, and decreases in the adrenal gland.	Ye <i>et al.</i> ¹⁸
ACTH→cortisol	ACTH was released after the administration of CRH and followed by the release of GCs.	Kling <i>et al.</i> ¹⁹
ACTH→DHEAS	When ACTH is treated, the DHEA in serum and brain is increased.	Torres and Ortega ²⁰
ADH→ACTH	In a study of sheep, CRH and AVP stimulated the release of ACTH.	Matthews and Challis ²¹
Aldosterone→glucose	When aldosterone is given to mice, the level of blood glucose is increased. When aldosterone is administered to liver cells, the promoter activity of glucose-6-phosphatase is increased.	Yamashita <i>et al.</i> ²²
Cortisol→free cortisol	In normal circulatory condition, 80–90% of cortisol strongly binds with CBG, 10–15% of the cortisol binds with low affinity, and 5–10% is unbinding free form.	Lewis <i>et al.</i> ²³
Free cortisol→glucose	The gene of G6Pase has two glucocorticoid response elements. When glucocorticoid is administrated to a liver cell <i>in vivo</i> , the activity of G6Pase is increased by 40%.	Lin <i>et al.</i> ²⁴
CRH→ACTH	ACTH is released after the administration of CRH	Kling <i>et al.</i> ¹⁹
CRH→DHEAS	When CRH is treated, the DHEA in serum and brain is increased.	Torres <i>et al.</i> ²⁰
DHEAS→aldosterone	The administration of DHEAS to rats increases the level of aldosterone.	Song <i>et al.</i> ²⁵
DHEAS→cortisol	When DHEAS is treated to rats, the level of cortisol shows a significant increase	Song <i>et al.</i> ²⁵
DHEAS→E2	When DHEAS is ingested by a human, the level of serum E2 and IGF-1 is increased.	Jankowski <i>et al.</i> ²⁶
E2→GH	When the level of estrogen increases during the menstrual cycle, the level of GH also increases.	Ovesen <i>et al.</i> ²⁷
FSH→E2	FSH receptor is expressed in follicle cells only, FSH is a decisive element for estrogen production in the ovary. FSH induces aromatase activity in the granulosa cell of a hair follicle.	Fogle <i>et al.</i> ²⁸
FSH→LH	Administration of FSH suppresses the release of LH in the hypothalamus and is independently mediated by ovary dose dependently.	Gordon <i>et al.</i> ²⁹
GH→Glucose	When GH is treated, the mean level of blood glucose is increased.	Qian <i>et al.</i> ³⁰
GH→HbA1c	The administration of growth hormone increases fasting blood glucose in patient deficient in growth hormone.	Woodmansee <i>et al.</i> ³¹
Glucose→HbA1c	Glycated hemoglobin represents the mean value of blood	Alberti and Zimmet ¹
Insulin→glucose	Insulin decreases the activity of G6Pase and the level of mRNA.	Argaud <i>et al.</i> ³²
LH→E2	LH stimulates the production of estrogen in the ovary.	Macklon <i>et al.</i> ³³

several causative variables with the one resultant variable, standardized path coefficients were used. When a causative variable changed by one standard deviation, the resultant variable changed by the same value of the standardized path coefficient. In the FRG group, when aldosterone increased to one standard deviation (3.6 ng/dL), blood glucose significantly decreased to 0.147 of one standard deviation (7.1 mg/dL $\times$ 0.147 = 1.00 mg/dL), whereas free cortisol increased to one standard deviation (14.0 μ g/day), and blood glucose increased to 0.032 of one standard deviation of blood glucose (7.1 mg/dL $\times$ 0.032 = 0.227 mg/dL) (Table 4).

To assess whether or not the difference in a path between groups was significant, the Wald test with cross-group equality constraints was employed. When the difference between the original chi-square value and the cross-group equality constrained chi-square value was higher than 3.84, the hypothesis of cross-group equality was rejected, which meant the differences were statistically significant. In the path of aldosterone to blood glucose, when the path coefficients between the FRG group and the placebo group were constrained for cross-

group equality, the value of the Wald test was 6.90 and the *P* value was 0.009. Therefore, the path coefficients between both groups were significantly different. (Table 4).

Based on the results listed above, the final model was made and presented in Figure 2. The model's goodness of fit was excellent, as accounted for by the measures root mean square error of approximation (0.00) and comparative fit index (1.00). The chi-square of the model was 146.5; the degree of freedom value was 198.

DISCUSSION

Hormones

The highest percentages of ginsenosides in this FRG capsule were Rg3 and compound K, which are the ligands of ER and GR, respectively. Therefore, the hormones evaluated in this study were those on the hypothalamic–pituitary–adrenal (HPA) axis and those on the hypothalamic–pituitary–gonadal (HPG) axis; the central variables for diabetes were blood glucose, HbA1c, insulin, and HOMA-IR.

TABLE 4. PATH COEFFICIENTS OF THE BASELINE MODEL AND VALUES OF THE WALD TEST

Path	Unstandardized estimate	Standardized estimate	Wald test ^a χ^2_D	Path	Unstandardized estimate	Standardized estimate	Wald test ^a χ^2_D
ACTH → aldosterone			0.51	DHEAS → estradiol			0.33
Placebo	-0.3 ± 0.1**	-0.4 ± 0.2***		Placebo	0.2 ± 0.2	0.1 ± 0.2	
FRG	-0.1 ± 0.2	-0.1 ± 0.2		FRG	0.4 ± 0.3	0.3 ± 0.2	
ACTH → cortisol			0.03	Estradiol → GH			2.42
Placebo	0.4 ± 0.1***	0.5 ± 0.1***		Placebo	0.2 ± 0.3	-0.4 ± 0.2**	
FRG	0.4 ± 0.1***	0.5 ± 0.1***		FRG	-0.3 ± 0.1**	0.2 ± 0.3	
ACTH → DHEAS			0.69	FSH → estradiol			1.28
Placebo	0.0 ± 0.1	0.0 ± 0.2		Placebo	-0.1 ± 0.3	-0.2 ± 0.3	
FRG	0.1 ± 0.1	0.2 ± 0.1		FRG	-0.5 ± 0.2**	-0.5 ± 0.2**	
ADH → ACTH			0.06	FSH → LH			0.00
Placebo	0.0 ± 0.4	0.0 ± 0.3		Placebo	0.4 ± 0.2*	0.4 ± 0.2**	
FRG	0.1 ± 0.3	0.1 ± 0.2		FRG	0.4 ± 0.2*	0.5 ± 0.2*	
Aldosterone → glucose			6.90***	GH → glucose			0.60
Placebo	0.9 ± 0.3***	0.6 ± 0.1***		Placebo	-0.5 ± 0.4	-0.3 ± 0.2	
FRG	0.0 ± 0.3	-0.1 ± 0.3		FRG	-0.2 ± 0.3	-0.2 ± 0.2	
Cortisol → free cortisol			2.30	GH → HbA1c			7.81***
Placebo	0.4 ± 0.2**	0.3 ± 0.1**		Placebo	-0.4 ± 0.3	-0.2 ± 0.24	
FRG	0.1 ± 0.1	0.1 ± 0.1		FRG	0.7 ± 0.2***	0.6 ± 0.1***	
Cortisol → glucose			1.05	Glucose → HbA1c			0.09
Placebo	-0.1 ± 0.2	-0.1 ± 0.1		Placebo	0.6 ± 0.2***	0.5 ± 0.1***	
FRG	0.1 ± 0.2	0.1 ± 0.2		FRG	0.5 ± 0.1***	0.5 ± 0.1***	
Free cortisol → glucose			0.56	Glucose → HOMA			0.75
Placebo	0.2 ± 0.1*	0.2 ± 0.1*		Placebo	0.2 ± 0.0***	0.2 ± 0.0***	
FRG	0.0 ± 0.2	0.0 ± 0.2		FRG	0.2 ± 0.0***	0.2 ± 0.0***	
CRH → ACTH			0.04	Insulin → glucose			0.10
Placebo	0.1 ± 0.7	0.1 ± 0.3		Placebo	0.3 ± 0.2*	0.3 ± 0.2*	
FRG	0.3 ± 0.5	0.2 ± 0.3		FRG	0.2 ± 0.2	0.2 ± 0.2	
CRH → DHEAS			0.05	Insulin → HOMA			2.26
Placebo	0.4 ± 0.3	0.2 ± 0.2		Placebo	0.9 ± 0.0***	0.9 ± 0.0***	
FRG	0.3 ± 0.2*	0.3 ± 0.2*		FRG	0.9 ± 0.0***	1.0 ± 0.0***	
DHEAS → aldosterone			0.16	LH → estradiol			0.30
Placebo	0.1 ± 0.2	0.1 ± 0.2		Placebo	0.2 ± 0.2	0.2 ± 0.2	
FRG	0.3 ± 0.2	0.2 ± 0.2		FRG	-0.1 ± 0.5	-0.1 ± 0.3	
DHEAS → cortisol			0.19				
Placebo	-0.1 ± 0.1	0.0 ± 0.1					
FRG	0.1 ± 0.2	0.0 ± 0.1					

Data expressed as a mean ± SD. Values presented in boldface indicate significant difference ($P < .05$) between FRG and control groups by the Wald test.

^aWald test was performed using one unstandardized path coefficient.

Values are significantly different (* $P < .1$, ** $P < .05$, *** $P < .01$) by analysis of covariance.

The peak period of DHEAS production is between the ages of 20–30 years, followed by a decrease of 2% every year, until the rate of release is finally only 10–20% of the peak levels at 70 years of age.³⁴ Studies reported that DHEAS, E2, and GH have serial causative relationships. Pluchino and colleagues reported that when DHEAS was administered to postmenopausal women, the level of estrogen increased,³⁵ which is consistent with the path coefficient of DHEAS to E2 (0.307) in the equality constraint state (Fig. 2). When the level of estrogen increases during the menstrual cycle, the level of GH also increases,²⁷ which is consistent with the path coefficient of E2 to GH (0.160) in the FRG group, but not consistent with the path coefficient of E2 to GH in the placebo group (-0.301; Table 4). Studies have reported that the level of GH is negatively correlated with the incidence of insulin resistance. Colao *et al.* reported that after administration of GH, subjects showed a significant decrease

in insulin resistance,³⁶ which is consistent with the path coefficient of GH to blood glucose (-0.358) in this study. Nam *et al.* reported that there was no difference in the levels of GH between a red ginseng group and a placebo group, whereas red ginseng consumption with aerobic exercise was found to significantly increase the level of GH,³⁷ which is consistent with the higher GH level in the FRG group. Salpeter and colleagues reported in a meta-analysis that HRT decreases insulin resistance (HOMA-IR) by 12.9%, and reduces the risk of diabetes by 35.8% in postmenopausal women.³⁸ Considering the report of Salpeter *et al.*,³⁸ in this study, the higher level of E2 in the FRG group should be beneficial for glucose management.

Since the levels of E2, GH, and DHEAS gradually decrease along with ageing and the postmenopausal period, even though the mean differences between groups were small, the cumulative effects of these hormonal increments

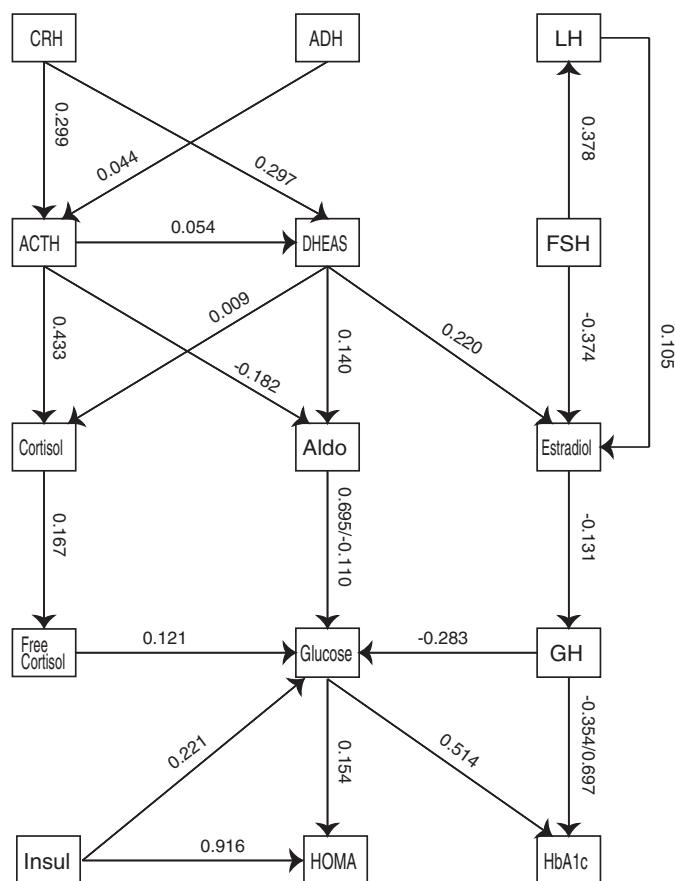


FIG. 2. The final path model of hormones and diabetes markers. Two paths showed the significant differences in the present path model. First, the path coefficient of aldosterone (Aldo) on blood glucose was significantly different across two groups ($P=.005$). Second, the path coefficient of growth hormone (GH) on blood glucose was significantly different across two groups ($P=.009$). These invariance tests were conducted by the Wald test, and the numbers in parenthesis present the unstandardized path coefficients.

in the FRG group can be interpreted as having the potential to affect the progress of diabetes.

Vuksan and colleagues reported that the consumption of red ginseng improved glucose and insulin regulation in 19 participants with well-controlled type 2 diabetes. Red ginseng consumption decreased fasting plasma insulin by 8 pM, plasma glucose during a 75 g oral glucose tolerance test (OGTT) by 8–11%, and it increased the fasting-HOMA-insulin sensitive index by 33%.¹⁵ In this study, the insulin levels and HOMA-IRs of the second sample, and the gap between the second and first sample, were both significantly lower in the FRG group than in the placebo group (Table 1). Vogeser *et al.*³⁹ reported that the distribution of HOMA-IR results was mainly determined by fasting serum levels rather than fasting glucose levels, because of the high variability of fasting serum insulin concentrations. In this path model, the contribution ratios in HOMA-IR of insulin and blood glucose were 0.918 and 0.157 respectively in the standardized path coefficients (Table 4).

Clinically, the HbA1c level reflects the average concentration of blood glucose for 2 or 3 months. The American Diabetes Association considers HbA1c as a criterion for the diagnosis of diabetes. Normally, HbA1c increases 0.1% every decade after 40 years of age.⁴⁰ In the difference between the second and first sample in this study, the level of HbA1c was significantly lower in the FRG group than in the placebo group.

In the path analysis, aldosterone was the largest single factor in the decreased blood glucose of the FRG group. Aldosterone regulates blood glucose through a mineral corticoid receptor (MR)-dependent mechanism with GLUT4 and GLUT2, and an MR-nondependent mechanism with an insulin receptor.⁴¹ GLUT4 is a glucose transporter that is highly expressed in skeletal muscle and adipose tissue.⁴² When insulin stimulates the cells, GLUT4 in the cytosol translocates to the cell membranes, and transports glucose across the cell membrane. Therefore, the impairment of GLUT4 functionality is an important cause of insulin resistance, as seen in type 2 diabetes patients.⁴³ When aldosterone is administrated to rats, the level of GLUT4 protein drops remarkably in muscle cells, whereas glucocorticoid treatments increase the expression of GLUT4.⁴⁴ Since the effects of aldosterone on GLUT4 are related to MR-MR homodimer and MR-GR heterodimer, and it is well known that cortisol is a ligand of GRs, cortisol levels can also related to the GLUT4 mechanism. Huang *et al.* reported that when CK or Rg1 was administered to adipocyte cell lines, GLUT4 mRNA and glucose uptake increased. This increase implicated GLUT4 movement from intracellular vesicles to the plasma membrane.⁴⁵ The MR-nondependent mechanism in glucose regulation of aldosterone is related to the gene expression of the insulin receptor. GREs on the promoter of insulin receptor are regulated by an MR-MR homodimer or an MR-GR heterodimer. When aldosterone was administered to MIN6 beta cells, a pancreas cell line, insulin release was suppressed.⁴⁶ Since the aldosterone and glucocorticoid share the GR and MR,⁴⁷ it is possible that ginsenosides, especially compound K, may interact with aldosterone functions.

In conclusion, in the conventional average comparison, the FRG group significantly increased the levels of DHEAS, GH, and E2, and decreased the levels of HbA1c, insulin, and HOMA-IR. In the hypothesis of this path model, DHEAS, E2, GH, blood glucose, and HbA1c established a causal relationship. The blood glucose lowering effects of the FRG group came from two causal effects. One line was the negative effects of aldosterone and the other line was GH, which was connected with DHEAS and E2. Considering the participants were healthy postmenopausal women who may be assumed to have a healthy level of homeostasis, the small difference of several variables may be reasonable and somewhat desirable, and the cumulated summation of these variable changes in the FRG group cannot be ignored as a potential preventive effect for postmenopausal women at risk of developing diabetes.

The fermentation process of ginseng transforms the inactive state of the ginsenosides, Rb1, Rc, Rb2, Rb3, and Rd, to a bioactive state, rendering the ginsenosides into an easily absorbable structure such as CK.⁴⁸ The effect of red ginseng

can vary depending on the type, ratio, period, and ginsenoside dose. Therefore, for a better understanding of the effects of ginseng, a study with a single type of ginsenoside and unhealthy participants, including those with diabetes or cardiovascular disease, may prove instructive.

ACKNOWLEDGMENTS

We thank Mr. John Mensing (PhD Candidate, Department of Pali and Buddhist Studies, University of Peradeniya, Peradeniya, Sri Lanka), who assisted with the proofreading of the article. This work was supported by the Next-Generation BioGreen 21 Program (No. PJ009543), Rural Development Administration, and by the Small and Medium Business Administration (SA114187), Republic of Korea.

AUTHOR DISCLOSURE STATEMENT

No competing financial interests exist.

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(Appendix follows) →

APPENDIX TABLE A1. REAGENTS AND INSTRUMENTS IN THIS STUDY

Test name [units of measurement]								
	ACTH [pg/mL]	ADH (RIA) [pg/mL]	Aldosterone [ng/dL]	Cortisol [μg/dL]	Cortisol, free [μg/dL]	CRH [ng/dL]	DHEA-S [μg/dL]	Estrogen (E1) [pg/mL]
Test code	E416	E418	E436	E435	E439	—	E431	E010
Test method	IRMA	RIA	RIA	RIA	RIA	EIA	RIA	RIA
Storage method of sample	Cold storage	Cold storage	Cold storage	Cold storage	Cold storage	Cold storage	Cold storage	Cold storage
Reagent								
Kit name	ACTH IRMA	Vasopressin 1251 RIA kit	Coat-A-Count Aldosterone	Coat-A-Count Cortisol	Coat-A-Count Cortisol	CRH EIA kit	Coat-A-Count DHEA-Sulfate	Total Estrogen
Kit company, nationality	Brahms, Germany	DiaSorin, USA	Siemens, USA	Siemens, USA	Siemens, USA	Phoenix, USA	Siemens, USA	ICN, USA
Analytical instrument								
Instrument name	R-counter	R-counter	R-counter	R-counter	R-counter	EIA reader	R-counter	R-counter
Model name	Cobra 5010 series Quantum	Cobra 5010 series Quantum	Cobra 5010 series Quantum	Cobra 5010 series Quantum	Cobra 5010 series Quantum	E max presion	Cobra 5010 series Quantum	Cobra 5010 series Quantum
Company, nationality	Packard, USA	Packard, USA	Packard, USA	Packard, USA	Packard, USA	Molecular Devices, USA	Packard, USA	Packard, USA
Reference range	8–10 a.m.: 10–60 8–10 p.m.: 6–30	≤4.7	Supine: 1.0–16.0 Standing: 4.0–31.0	a.m.: 5–25 p.m.: half of a.m. values	20–90	Standard range in kit: 0.04–25	M: 80–560 F: 35–430	M, adult: 40–115 M or F, prepubertal: ≤40 F, follicular: 61–394 midcycle: 122–437 luteal: 156–350 postmenopausal: ≤40
(continued)								

(continued)

APPENDIX TABLE A1. (CONTINUED)

Test name								
	Estrogen (E2) [pg/mL]	FSH [mU/mL]	Glucose (S) [mg/dL]	HbA1c [%]	HGH (S) [ng/mL]	IGF-1 [ng/mL]	Insulin [μU/mL]	LH [mU/mL]
Test code	C001	E421	C123	C431	E417	E455	E441	E422
Test method	CLIA	CLIA	Enzymatic method	HPLC	CLIA	CLIA	ECLIA	CLIA
Storage method of sample	Refrigeration	Cold storage	Refrigeration	Refrigeration	Cold storage	Cold storage	Cold storage	Cold storage
Reagent								
Kit name	Advia Centaur Estradiol	Advia Centaur FSH	Glucose Hexokinase	Variant II HbA1c T.	Immulite 2000 GH	IGF-1	Insulin	Advia Centaur FSH
Kit company, nationality	Bayer, USA	Siemens, USA	Bayer, USA	Bio-Rad, Germany	DPC, USA	DPC, USA	Roche, Germany	Siemens, USA
Analytical instrument								
Instrument name	Centaur	Advia Centaur	Advia	Variant	Immulite	Immulite 2000	Modular Analytics	Advia Centaur
Model name	Advia Centaur	Advia Centaur	Advia 1650	Variant II Turbo	Immulite 2000	Immulite 2000	E170	Advia Centaur
Company, nationality	Bayer, USA	Siemens, USA	Bayer, Japan	Bio-Rad, Germany	DPC, USA	DPC, USA	Roche, Germany	Siemens, USA
Reference	M: < 52 F, follicular: 11–165 midcycle: 146–526 luteal: 33–196 postmenopausal: <37	M, 13–70 years: 1.4–18.1 F, follicular: 2.5–10.2 midcycle: 3.4–33.4 luteal: 1.5–9.1 pregnant: <0.3 postmenopausal: 23.0–116.3	70–110	3.5–6.5	M, adult: ≤1.0 F, adult: ≤10.0	1–5 years: 49–327 6–8 years: 52–345 9–11 years: 74–551 12–13 years: 143–850 14–17 years: 193–996 18–25 years: 116–584 26–40 years: 109–329 41–55 years: 87–267 >55 years: 55–225	2.6–24.9	M, 20–70 years: 1.5–9.3 >70 years: 3.1–34.6 F, follicular: 1.9–12.5 midcycle: 8.7–76.3 luteal: 0.5–16.9 pregnant: <1.5 postmenopausal: 15.9–54.0

CLIA, chemiluminescence immunoassay; ECLIA, electrochemiluminescence immunoassay; EIA, enzyme immunoassay; HPLC, high-performance liquid chromatography; IRMA, immunoradiometric assay; RIA, radioimmunoassay.

APPENDIX TABLE A2. CORRELATION COEFFICIENTS OF HORMONES IN THIS STUDY—TWO-SIDED TEST WITH PEARSON CORRELATION COEFFICIENTS

	ACTH	ADH	Aldosterone	CRH	DHEAS	E2	FSH	GCs	fGCs	GH	Glucose	HbA1c	IGF1	Insulin	LH
ACTH	1	-0.051	-0.368**	0.008	0.119	-0.093	-0.304	0.513***	-0.022	-0.366**	0.050	-0.040	0.022	0.140	-0.357**
Pearson coefficient															
Significance		0.754	0.020	0.959	0.255	0.568	0.057	0.000	0.837	0.020	0.637	0.707	0.833	0.187	0.024
(two tail)															
Sum of square	11,165.430	-3.661	-543.483	0.407	3625.281	-278.712	-2640.150	1892.192	-325.209	-201.538	353.358	-391.634	1522.510	325.886	-1847.679
Covariance	121.363	-0.094	-13.935	0.011	39.405	-7.146	-67.696	20.567	-3.654	-5.168	3.841	-4.257	16.549	3.662	-47.376
n	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40
ADH	-0.051	1	-0.111	-0.388**	-0.330**	0.026	-0.152	0.043	-0.023	-0.031	-0.078	0.041	-0.022	0.048	-0.138
Pearson coefficient															
Significance	0.754		0.497	0.015	0.037	0.873	0.350	0.794	0.894	0.850	0.632	0.801	0.893	0.772	0.397
(two tail)															
Sum of square	-3.661	1.229	-2.805	-0.323	-72.670	1.345	-22.663	1.011	-2.605	-0.291	-3.446	2.492	-8.888	0.818	-12.254
Covariance	-0.094	0.032	-0.072	-0.009	-1.863	0.034	-0.581	0.026	-0.072	-0.007	-0.088	0.064	-0.228	0.022	-0.314
n	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40
Aldosterone	-0.368**	-0.111	1	0.248	0.229	0.269	-0.017	-0.159	-0.141	0.369**	0.230	0.118	-0.210	0.153	0.126
Pearson coefficient															
Significance	0.020	0.497		0.129	0.155	0.093	0.919	0.326	0.404	0.019	0.153	0.468	0.194	0.352	0.437
(two tail)															
Sum of square	-543.483	-2.805	524.054	4.256	1040.763	285.357	-51.440	-77.965	-329.270	72.127	209.888	147.518	-1750.614	54.235	232.321
Covariance	-13.935	-0.072	13.437	0.112	26.686	7.317	-1.319	-1.999	-9.146	1.849	5.382	3.783	-44.888	1.427	5.957
n	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40
CRH	0.008	-0.388**	0.248	1	0.188	-0.117	-0.047	-0.212	-0.035	0.303	0.133	0.239	0.018	0.166	0.028
Pearson coefficient															
Significance	0.959	0.015	0.129		0.251	0.477	0.775	0.195	0.838	0.061	0.418	0.143	0.915	0.318	0.864
(two tail)															
Sum of square	0.407	-0.323	4.256	0.567	25.750	-4.044	-4.731	-3.405	-2.738	1.916	3.944	9.819	4.825	1.884	1.705
Covariance	0.011	-0.009	0.112	0.015	0.678	-0.106	-0.124	-0.090	-0.078	0.050	0.104	0.258	0.127	0.051	0.045
n	39	39	39	39	39	39	39	39	36	39	39	39	39	38	39
DHEAS	0.119	-0.330**	0.229	0.188	1	0.142	0.230	0.067	-0.077	-0.049	0.183	0.133	0.169	0.125	0.162
Pearson coefficient															
Significance	0.255	0.037	0.155	0.251		0.381	0.154	0.522	0.469	0.764	0.080	0.203	0.106	0.241	0.318
(two tail)															
Sum of square	3625.281	-72.670	1040.763	25.750	82918.040	1309.486	6135.009	676.283	-3082.100	-83.025	3551.608	3596.090	31,632.259	788.628	2578.632
Covariance	39.405	-1.863	26.686	0.678	901.283	33.577	157.308	7.351	-34.630	-2.129	38.604	39.088	343.829	8.861	66.119
n	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40

(continued)

APPENDIX TABLE A2. (CONTINUED)

	ACTH	ADH	Aldosterone	CRH	DHEAS	E2	FSH	GCs	fGCs	GH	Glucose	HbA1c	IGF1	Insulin	LH
E2															
Pearson coefficient	-0.093	0.026	0.269	-0.117	0.142	1	-0.320**	-0.178	-0.171	-0.025	0.184	0.071	0.164	0.247	-0.179
Significance (two tail)	0.568	0.873	0.093	0.477	0.381		0.044	0.273	0.311	0.879	0.256	0.664	0.311	0.130	0.269
Sum of square	-278.712	1.345	285.357	-4.044	1309.486	2149.515	-2000.403	-175.953	-801.559	-9.824	339.385	179.371	2775.361	175.552	-665.779
Covariance	-7.146	0.034	7.317	-0.106	33.577	55.116	-51.292	-4.512	-22.266	-0.252	8.702	4.599	71.163	4.620	-17.071
<i>n</i>	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40
FSH															
Pearson coefficient	-0.304	-0.152	-0.017	-0.047	0.230	-0.320**	1	0.032	0.387**	0.075	0.027	-0.224	-0.048	-0.238	0.618***
Significance (two tail)	0.057	0.350	0.919	0.775	0.154	0.044		0.845	0.018	0.646	0.868	0.164	0.770	0.145	0.000
Sum of square	-2640.150	-22.663	-51.440	-4.731	6135.009	-2000.403	18,129.884	91.615	4626.337	85.990	145.429	-1648.293	-2344.634	-494.963	6675.971
Covariance	-67.696	-0.581	-1.319	-0.124	157.308	-51.292	464.869	2.349	128.509	2.205	3.729	-42.264	-60.119	-13.025	171.179
<i>n</i>	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40
GCs															
Pearson coefficient	0.513***	0.043	-0.159	-0.212	0.067	-0.178	0.032	1	0.179	-0.086	0.089	-0.146	-0.046	0.122	0.089
Significance (two tail)	0.000	0.794	0.326	0.195	0.522	0.273	0.845		0.091	0.598	0.396	0.163	0.665	0.252	0.583
Sum of square	1892.192	1.011	-77.965	-3.405	676.283	-175.953	91.615	1217.652	887.288	-15.674	209.940	-477.044	-1034.772	92.957	153.336
Covariance	20.567	0.026	-1.999	-0.090	7.351	-4.512	2.349	13.235	9.970	-0.402	2.282	-5.185	-11.248	1.044	3.932
<i>n</i>	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40
fGCs															
Pearson coefficient	-0.022	-0.023	-0.141	-0.035	-0.077	-0.171	0.387**	0.179	1	0.088	0.210**	-0.033	0.010	0.136	0.462***
Significance (two tail)	0.837	0.894	0.404	0.838	0.469	0.311	0.018	0.091		0.604	0.047	0.754	0.922	0.210	0.004
Sum of square	-325.209	-2.605	-329.270	-2.738	-3082.100	-801.559	4626.337	887.288	20,332.205	78.186	2000.389	-442.690	960.588	421.727	3834.771
Covariance	-3.654	-0.072	-9.146	-0.078	-34.630	-22.266	128.509	9.970	228.452	2.172	22.476	-4.974	10.793	4.904	106.521
<i>n</i>	90	37	37	36	90	37	37	90	90	37	90	90	90	87	37
GH															
Pearson coefficient	-0.366**	-0.031	0.369**	0.303	-0.049	-0.025	0.075	-0.086	0.088	1	-0.139	0.181	0.203	-0.171	0.265
Significance (two tail)	0.020	0.850	0.019	0.061	0.764	0.879	0.646	0.598	0.604		0.391	0.265	0.209	0.298	0.098
Sum of square	-201.538	-0.291	72.127	1.916	-83.025	-9.824	85.990	-15.674	78.186	72.841	-47.288	84.225	632.069	-22.672	181.619
Covariance	-5.168	-0.007	1.849	0.050	-2.129	-0.252	2.205	-0.402	2.172	1.868	-1.213	2.160	16.207	-0.597	4.657
<i>n</i>	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40

(continued)

APPENDIX TABLE A2. (CONTINUED)

	ACTH	ADH	Aldosterone	CRH	DHEAS	E2	FSH	GCs	fGCs	GH	Glucose	HbA1c	IGF1	Insulin	LH
Glucose															
Pearson coefficient	0.050	-0.078	0.230	0.133	0.183	0.184	0.027	0.089	0.210**	-0.139	1	0.445***	0.068	0.194	0.121
Significance (two tail)	0.637	0.632	0.153	0.418	0.080	0.256	0.868	0.396	0.047	0.391		0.000	0.519	0.067	0.457
Sum of square	353.358	-3.446	209.888	3.944	3551.608	339.385	145.429	209.940	2000.389	-47.288	4557.118	2818.065	2978.642	286.648	386.082
Covariance	3.841	-0.088	5.382	0.104	38.604	8.702	3.729	2.282	22.476	-1.213	49.534	30.631	32.377	3.221	9.900
<i>n</i>	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40
HbA1c															
Pearson coefficient	-0.040	0.041	0.118	0.239	0.133	0.071	-0.224	-0.146	-0.033	0.181	0.445***	1	0.076	0.119	-0.019
Significance (two tail)	0.707	0.801	0.468	0.143	0.203	0.664	0.164	0.163	0.754	0.265	0.000		0.471	0.266	0.907
Sum of square	-391.634	2.492	147.518	9.819	3596.090	179.371	-1648.293	-477.044	-442.690	84.225	2818.065	8794.933	4615.574	238.352	-83.798
Covariance	-4.257	0.064	3.783	0.258	39.088	4.599	-42.264	-5.185	-4.974	2.160	30.631	95.597	50.169	2.678	-2.149
<i>n</i>	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40
IGF1															
Pearson coefficient	0.022	-0.022	-0.210	0.018	0.169	0.164	-0.048	-0.046	0.010	0.203	0.068	0.076	1	0.015	-0.001
Significance (two tail)	0.833	0.893	0.194	0.915	0.106	0.311	0.770	0.665	0.922	0.209	0.519	0.471		0.885	0.997
Sum of square	1522.510	-8.888	-1750.614	4.825	31,632.259	2775.361	-2344.634	-1034.772	960.588	632.069	2978.642	4615.574	423,959.630	222.087	-17.970
Covariance	16.549	-0.228	-44.888	0.127	343.829	71.163	-60.119	-11.248	10.793	16.207	32.377	50.169	4608.257	2.495	-0.461
<i>n</i>	93	40	40	39	93	40	40	93	90	40	93	93	93	90	40
Insulin															
Pearson coefficient	0.140	0.048	0.153	0.166	0.125	0.247	-0.238	0.122	0.136	-0.171	0.194	0.119	0.015	1	-0.156
Significance (two tail)	0.187	0.772	0.352	0.318	0.241	0.130	0.145	0.252	0.210	0.298	0.067	0.266	0.885		0.344
Sum of square	325.886	0.818	54.235	1.884	788.628	175.552	-494.963	92.957	421.727	-22.672	286.648	238.352	222.087	492.369	-193.254
Covariance	3.662	0.022	1.427	0.051	8.861	4.620	-13.025	1.044	4.904	-0.597	3.221	2.678	2.495	5.532	-5.086
<i>n</i>	90	39	39	38	90	39	39	90	87	39	90	90	90	90	39
LH															
Pearson coefficient	-0.357**	-0.138	0.126	0.028	0.162	-0.179	0.618***	0.089	0.462***	0.265	0.121	-0.019	-0.001	-0.156	1
Significance (two tail)	0.024	0.397	0.437	0.864	0.318	0.269	0.000	0.583	0.004	0.098	0.457	0.907	0.997	0.344	
Sum of square	-1847.679	-12.254	232.321	1.705	2578.632	-665.779	6675.971	153.336	3834.771	181.619	386.082	-83.798	-17.970	-193.254	6440.390
Covariance	-47.376	-0.314	5.957	0.045	66.119	-17.071	171.179	3.932	106.521	4.657	9.900	-2.149	-0.461	-5.086	165.138
<i>n</i>	40	40	40	39	40	40	40	40	37	40	40	40	40	39	40

Values are significantly different as indicated (** $P < .05$, *** $P < .01$) by analysis of covariance (SPSS v. 18).