

The effect of fermented red ginseng on depression is mediated by lipids

Kwang Jo Lee¹, Geun Eog Ji^{1,2}

¹Department of Food and Nutrition, Research of Institute of Human Ecology, Seoul National University, Seoul 151-742, Korea, ²Research Center, Bifido Inc., Hongcheon, Gangwon 250-804, Republic of Korea

Objectives: The cortico-limbic hypothalamic–pituitary–adrenal axis has emerged as an important area for the cause and treatment of depression. The primary aim of this study was to test the hypothesis that hormones, energy sources, and minerals have a causal relationship with depression. The secondary aim was to test whether consumption of fermented red ginseng (FRG) would influence that causal relationship.

Methods: For this study, 93 postmenopausal women were randomly divided into two groups. One group (49 women) was supplied with FRG capsules, and the other group (44 women) with placebo capsules, for 2 weeks. Both before and after the study, the participants filled out the Beck depression inventory questionnaire, and then blood samples were collected. The structural regression model was established. The causative latent variables were hormone (adrenocorticotrophic hormone and cortisol), energy (low-density lipoprotein (LDL) cholesterol, total cholesterol, and blood glucose), mineral 1 (potassium, sodium, chloride, and iron), and mineral 2 (magnesium, calcium), and the resultant latent variables were cognitive depression (CD) and somatic depression. The goodness-of-fit statistics of the final model were good (root mean square error of approximation = 0.033, comparative fit index = 0.877, and Tucker–Lewis index = 0.870).

Results: The structural regression path of the energy factor on CD showed a significant difference between the FRG group (0.259) and the placebo group (−0.201). The factor loadings of total cholesterol (1.236) and LDL cholesterol (1.000) on the energy factor were much higher than that of glucose (0.166).

Conclusion: Based on the analysis used in this model, the effect of FRG consumption on CD occurred via the energy factor, which is mainly attributable to cholesterol.

Keywords: Cholesterol, Depression, Fermented red ginseng, Multiple imputation, Structural equation model

Introduction

The limitations of monoamine theory in depression

The World Health Organization (WHO) defines depression as ‘a common mental disorder that presents with depressed mood, loss of interest or pleasure, feelings of guilt or low self-worth, disturbed sleep or appetite, low energy, and poor concentration’.¹ Concern about the incidence of depression has grown with the increase in the number of patients being diagnosed with depression.²

The standard medical models of depression treatments involve normalizing the functions of monoamine neurotransmitters such as serotonin, norepinephrine, and dopamine.³ However, the monoamine hypothesis underlying this protocol has several limitations. These include the low efficacy rates, the long period (several weeks) needed for symptom improvement, and the side effects, which include agitation, nausea, and sexual dysfunction.⁴

Limbic HPA axis, hormones, and depression

The cortico-limbic hypothalamic–pituitary–adrenal (HPA) axis circuit has emerged as an important area of study for the elucidation of the causes of depression. The limbic system, which encloses the brain stem, includes the cingulate gyrus, amygdala, hippocampus, hypothalamus, and anterior thalamic nuclei. Some of the area of the limbic system and the HPA axis is composed of the circumventricular organs (CVOs). The CVOs are able to reduce the blood–brain barrier, and are influenced by circulating hormones, energy sources, and minerals.⁵

Several hormones have been shown to influence the function of the limbic system. For example, the amygdala and the hippocampus have a high concentration of glucocorticoid receptors (GRs),⁶ and a pathologically high level of cortisol may damage the neurons⁷ via apoptotic signals.⁸ Such a defective limbic system may induce depression. Patients with depression tend to show a decreased volume of the amygdala,⁹ and the degree of shrinkage in hippocampal volume has been shown to be proportional to the period and severity of the symptoms of depression.¹⁰

Correspondence to: Kwang Jo Lee, Department of Food and Nutrition, Seoul National University, 1 Gwanak-ro, Gwanak-gu, Seoul 151-742, Republic of Korea. Email: geji@snu.ac.kr

Cortex, energy, and depression

It is well known that the prefrontal cortex (PFC) regulates thinking, emotion, behaviour, and decision making. According to the results of a functional magnetic resonance imaging study, patients with depression showed a decrease in blood supply and energy metabolism in the dorsolateral and dorsomedial PFC regions compared with a healthy control group. Furthermore, when these areas were normalized, the depressive symptoms improved.¹¹

Minerals and depression

Iron, potassium, sodium, magnesium, chloride, and calcium are essential components for the action potential in neurons, and for other aspects including synaptic communication. Patients with depression have been shown to have a low level of serum ferritin.¹² The symptoms of iron deficiency are similar to those occurring at the onset of depression, and iron and zinc supplements with vitamin have been shown to alleviate symptoms of depression.¹³

Another example is bio-energy release from ionic Mg adenosine triphosphate (ATP). A decreased level of ATP was found in patients with depression,¹⁴ and another found that the administration of magnesium supplements to those patients showed an anti-depression effect.¹⁵

The effects of red ginseng on hormones, energy sources, minerals, and depression

In Asia, especially in Korea, China, and Japan, ginseng has been one of the most popular herbal supplements, and records of its use date back 2000 years. In recent times, the popularity of ginseng has grown in western countries. For example, ginseng was one of the top 10 herbal supplements sold in the USA in 2003.¹⁶

The major pharmaceutical components of ginseng are ginsenosides, which have a steroidal structure, and have been shown to have a relationship with

hormones, energy metabolism, and mineral concentrations in the body. The ginsenosides Rh1¹⁷ and Rg3¹⁸ function as a ligand for oestrogen receptors (ERs), and Rh2¹⁹ and Compound K²⁰ for GRs. Therefore, it is possible to hypothesize that some ginsenosides can influence the components or level of depression mediated by ERs or GRs in the neurons of the limbic HPA axis. In fact, studies have reported that Rg1 decreases the apoptosis of neuronal cells, and this decrease is mediated by ERs and GRs.²¹ Furthermore, Rb1 and Rg3 alleviate neuronal toxicity, which is associated with glucocorticoid activity.²²

Second, ginseng has been shown to have the effect of lowering blood glucose levels and of decreasing insulin resistance. When crude saponin extract of red ginseng was administered to old female rats, their levels of blood glucose decreased from 148 to 124 mg/dl.²³ In a study on a group of patients with well-controlled type 2 diabetes who consumed red ginseng, there was a decrease in fasting plasma insulin (PI) of 8–11%, and a decrease of 33–38% in the PI level measured by the 75 g oral glucose tolerance test, compared with the healthy control group.²⁴ As well as blood glucose levels, the lipid levels were also influenced by consumption of red ginseng. Lee *et al.*²⁵ reported that red ginseng stimulated lipid oxidation in an *in vitro* study.

Third, ginseng treatment decreased the level of minerals in rat bones. For example, when the crude saponin extract of red ginseng was administered to old female rats, the level of sodium ions in the femur increased from 3.0 to 5.8 mg/g.²³

Aims and hypotheses

The first aim of this study was to test the relationship between depression (as represented by the Beck depression inventory (BDI)) and hormones, energy sources, and minerals. Based on the literature, the research hypotheses of this study were that (1) hormones influence depression via ERs or GRs in the limbic HPA axis; (2) energy sources influence depression via the central nervous system (CNS); and (3) minerals influence depression via action potential or synaptic function.

The second aim was to test the hypothesis that the consumption of fermented red ginseng (FRG) influences the causal relationship between hormones, energy sources, minerals, and depression. The research hypothesis was that some ginsenosides influence depression via the effects of hormones, energy sources, and minerals on the CNS.

Methods

Participants and criteria

The study was approved by the Internal Review Board of Sahmyook University. Women aged over 50 years old were recruited to the study. All participants of this study had to be free from diseases such as

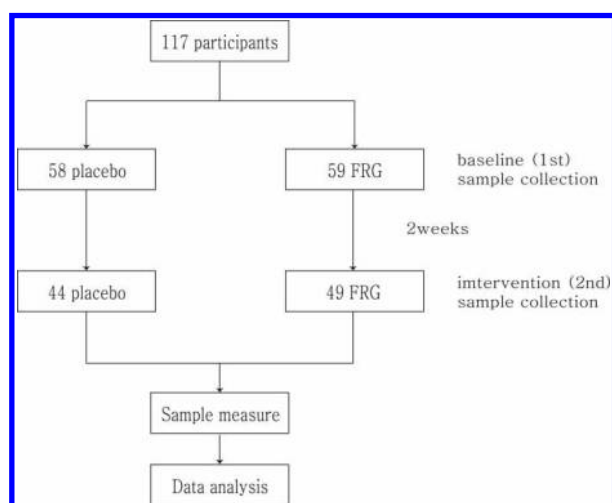


Figure 1 The flowchart of this study.

hypertension, high blood pressure, or diabetes, and were not taking any drugs or other supplements.

Procedure

In total, 117 (mean age 58.4 years, s.d. 5.6, range 50–73) participated in the first round of blood sampling and completed the BDI questionnaire. Blood samples were collected between 08:00 and 10:00 hours, following an 8-hour overnight fast. Participants completed the BDI questionnaire themselves before blood samples were taken (Fig. 1). Subsequently, these 117 women were divided into two groups by double-blind randomization (drawing lots). Appendix 1 shows the anthropometric variables of the participants. The groups were both given a 2-week supply of identical-looking capsules, with the experimental group (59 women) receiving capsules filled with lyophilized FRG powder, and the control group (58 women) receiving placebo capsules filled with edible starch.

Each FRG capsule contained 235 mg FRG powder. The components of the FRG powder were: 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. Each group consumed a total of nine capsules a day (three capsules three times a day), yielding a total consumption of 2.1 g of FRG a day for the experimental group. Given the various side effects that are possible during prolonged intake of a steroid drug,²⁶ we considered 2 weeks of FRG consumption to be both a sufficient and an advisable dosage, even though there were few side effects reported from this instance of red ginseng consumption. After 2 weeks, 24 women dropped out following the collection of the second blood sample, therefore, the final numbers analysed in this study were 49 women in the FRG group and of 44 women in the placebo group (Fig. 1).

Psychological variables

The BDI comprises 21 items, with each item measured on a four-point scale (0–3), a higher number indicates a higher frequency and greater significance of symptoms of depression. The total score for the 21 items on the BDI ranges from 0 to 63.

Biological variables

Blood samples were analysed (Green Cross Reference Lab, Gyeonggi-do, Korea), and the instruments, reagent, and methods for measurement are presented in Appendix 2.

Statistical analyses

Data were analysed using analysis of covariance using SPSS software (version 11.5, SPSS Inc, Chicago, IL, USA), and the results are presented as mean and standard error. Significance was set at $P < 0.05$, and the

statistical tendency at $P < 0.1$. Any outliers for oestrogen and insulin were excluded. After matching for age, weight, height, and body mass index, 20 subjects were assigned to each of the two groups, and the levels for several hormones were further analysed (Table 1).

A structural regression model was used to test the causal relationship between depression (BDI items) and hormones, energy sources, and minerals. The final model was established using the serial steps of equality constraint from the baseline model. All models of this study were fitted using the weighted least squares means and variance adjusted estimation method. Weighted least square-based estimators do not require large sample sizes for categorical variables.²⁷

The unmeasured variables, missing blood samples, and missing responses on the BDI questionnaires were assigned random values, which were generated by multiple imputations ($n = 10$)²⁸ using Mplus software (version 6.11, Muthén & Muthén, CA, USA), prior to data analysis.

The data in the present report are part of a larger FRG study on endocrine, metabolism, immune, and psychology, conducted in Seoul in 2010. Only the data relevant to this area of the study have been presented in this report.

Results

Comparison of psychological variables

The mean for the total values for all BDI items was 7.53 (s.d. 5.49) in the FRG group and 6.16 (s.d. 4.26) in the placebo group, and there was no significant difference ($P = 0.244$) between the groups in total mean BDI scores. Normally, a total score of 0–9 on the BDI indicates that the subject is experiencing a minimal amount of depression.

Comparison of biological (hormones, energy sources, minerals) variables

Table 1 shows the mean comparison of the endocrine and metabolic variables between these two groups. For the hormone variables, the level of growth hormone and oestradiol were significantly higher in the FRG group than in the placebo group ($P < 0.1$). The level of dehydroepiandrosterone sulphate (DHEAS) was higher in the FRG group (66.2, s.d. 29.1 µg/dl) than in the placebo group (65.2, s.d. 31.4 µg/dl) ($P = 0.02$).

Among the energy source variables, the level of insulin, haemoglobin A1c, and the homeostasis model of assessment of insulin resistance were significantly lower in the FRG group than in the placebo group. The three outlier values of the insulin samples were excluded from the data analysis (one from the FRG group and two from the placebo group). The level of low-density lipoprotein (LDL) cholesterol

Table 1 Comparison of variables mean between the FRG group and the placebo group

Markers	Groups	Second sample	P	$\Delta 2\text{nd} - 1\text{st}$	P
		Mean $\pm$ s.d. (n)		Mean $\pm$ s.d. (n)	
ACTH (pg/ml)	Placebo	20.4 $\pm$ 10.1 (44)	0.34	2.3	0.52
	FRG	22.7 $\pm$ 11.8 (49)		0.9	
Cortisol ($\mu\text{g}/\text{dl}$)	Placebo	11.2 $\pm$ 3.5 (44)	0.52	-0.2	0.13
	FRG	10.8 $\pm$ 3.8 (49)		0.9	
Estradiol (pg/ml)	Placebo	14.5 $\pm$ 7.3 (19)	0.06*	-7.4	0.06*
	FRG	18.3 $\pm$ 7.3 (19)		1.0	
GH (ng/ml)	Placebo	0.9 $\pm$ 1.2 (20)	0.06*	-0.40	0.50
	FRG	1.8 $\pm$ 1.4 (20)		-0.06	
DHEAS ($\mu\text{g}/\text{dl}$)	Placebo	65.2 $\pm$ 31.4 (44)	0.05*	-6.2	0.03**
	FRG	66.2 $\pm$ 29.1 (49)		0.5	
Insulin ($\mu\text{U}/\text{ml}$)	Placebo	6.4 $\pm$ 3.6 (43)	0.04**	1.0	0.01**
	FRG	6.2 $\pm$ 2.9 (48)		-0.2	
Glucose (mg/dl)	Placebo	89.6 $\pm$ 6.8 (44)	0.31	2.2	0.85
	FRG	87.4 $\pm$ 7.1 (49)		2.5	
HbA1c (%)	Placebo	5.77 $\pm$ 0.29 (44)	0.04**	0.09	0.06*
	FRG	5.70 $\pm$ 0.27 (49)		0.06	
HOMA-IR	Placebo	1.42 $\pm$ 0.80 (43)	0.06*	0.17	0.03**
	FRG	1.36 $\pm$ 0.67 (48)		-0.09	
Total cholesterol (mg/dl)	Placebo	216.2 $\pm$ 31.6 (44)	0.14	8.0	0.07*
	FRG	217.7 $\pm$ 30.9 (49)		-0.2	
LDL cholesterol (mg/dl)	Placebo	139.1 $\pm$ 31.6 (44)	0.08*	9.3	0.04**
	FRG	141.3 $\pm$ 30.1 (49)		0.7	
Calcium (serum) (mg/dl)	Placebo	10.8 $\pm$ 1.6 (45)	0.03**	-1.0	0.06*
	FRG	11.4 $\pm$ 1.7 (46)		-0.2	
Iron (plasma) ($\mu\text{g}/\text{dl}$)	Placebo	26.3 $\pm$ 7.4 (41)	0.68	9.1	0.44
	FRG	26.5 $\pm$ 11.4 (46)		8.5	
Magnesium (plasma)	Placebo	265 $\pm$ 48 (41)	0.06*	0.03	0.18
	FRG	282 $\pm$ 39 (46)		0.05	
Phosphate (plasma) (mg/dl)	Placebo	3.4 $\pm$ 0.5 (41)	0.19	-5.9	0.75
	FRG	3.6 $\pm$ 0.6 (46)		-5.8	
Sodium (plasma) (mmol/L)	Placebo	141.21 $\pm$ 1.5 (41)	0.97	-4.7	0.29
	FRG	141.2 $\pm$ 1.6 (46)		-4.5	
Chloride (plasma) (mmol/L)	Placebo	90.0 $\pm$ 5.7 (41)	0.78	-9.9	0.88
	FRG	90.3 $\pm$ 4.5 (46)		-9.5	

* $P < 0.1$, ** $P < 0.05$, *** $P < 0.01$.s.d., standard deviation; $\Delta 2\text{nd} - 1\text{st}$, second sample minus first sample; (n), number of sample; ACTH, adrenocorticotrophic hormone; GH, growth hormone; DHEAS, dehydroepiandrosterone; HbA1c, glycated haemoglobin; HOMA-IR, homeostatic model assessment insulin resistance.

was higher in the FRG group (141.3, s.d. 30.1 mg/dl) than in the placebo group (139.1, s.d. 31.6 mg/dl) ($P = 0.08$).

In the mineral variables, the level of serum calcium was significantly higher in the FRG group (11.4, s.d. 1.7 mg/dl) than in the placebo group (10.7, s.d. 1.6 mg/dl) ($P = 0.03$). The level of plasma magnesium was higher in the FRG group (282, s.d. 39 $\mu\text{g}/\text{dl}$) than in the placebo group (265, s.d. 48 $\mu\text{g}/\text{dl}$) ($P = 0.06$).

The multiple group structural regression models Factor analysis for construction of the model

Factors (latent variables) were ascertained from the observed variables. In this study, the causative latent variables (independent latent variables) were hormone, energy, mineral 1, and mineral 2, and the resultant latent variables (dependent latent variables) were somatic depression (SD) and cognitive depression (CD). The criteria for determining a finding of CD and SD were based on the literature²⁹ and current factor analysis.

The consecutive steps for the final model

There were three major invariance tests between the groups in this study: factor loading, structural regression paths, and latent variable means. For these tests, a model analysis was conducted in consecutive steps as follows.

Model 1 (the configural model) was taken to be the baseline model, which was constrained by the mean of the continuous observed variables (hormones, energy sources, and minerals), and was constrained by the categorically observed variables (BDI items) as 0 across both groups. Studies have reported that the goodness of fit has ranged from about 0.7 to 0.9 in factor analysis of BDI only.³⁰ Given the range of the goodness of fit for BDI factor analysis in the literature, the goodness-of-fit statistics of the present model 1, which included BDI and four causative latent variables (root mean square error of approximation (RMSEA) = 0.033, comparative fit index (CFI) = 0.888, and Tucker–Lewis index (TLI) = 0.875), were considered acceptable (Appendix 3).

Model 2 was a measurement model. All factor loadings were constrained equally for both groups. The goodness-of-fit statistics were good (RMSEA = 0.034, CFI = 0.883, TLI = 0.871), and these statistics did not show a significant difference from the baseline model 1 described above.

In model 3, the intercepts of the continuous observed variables were constrained equally for both groups and tested. The goodness-of-fit statistics were good (RMSEA = 0.034, CFI = 0.882, TLI = 0.872), and did not show a significant difference from the values of model 2.

In model 4, the factor loading and threshold of the categorical variables for each BDI item were constrained. The goodness-of-fit statistics of model 4 were good (RMSEA = 0.034, CFI = 0.879, TLI = 0.872). The difference in goodness-of-fit statistics between models 3 and 4 was not significant (Appendix 2).

In model 5, the structural regression paths between both groups were equal, the individual structural regression paths were equality constrained, and the model was analysed by the Wald test. The structural regression path estimate of the energy factor on CD was 4.495 ($P = 0.034$); therefore, there was a significant difference between the two groups, and thus the equality hypothesis was rejected.

Estimates derived from the final model

Based on the results, a final model was created (Fig. 2). The structural regression path of the energy factor on CD was freely estimated; and the other remaining structural regression paths and factor loadings were constrained for both groups. The structural regression estimate for the path of the energy factor on CD was significantly higher in the FRG group (0.259) than in the placebo group (-0.201).

When the factor-loading estimate for LDL cholesterol on the energy factor was 1, then the factor-loading estimate of total cholesterol on the energy factor was 1.246, and the factor-loading estimate of glucose on the energy factor was very low (0.166).

When a baseline of 0 was taken as the mean for each factor for the FRG group, a comparison with the means in the placebo group showed values of -0.238 for hormone, -0.062 for energy, -0.232 for mineral 1, and -0.217 for mineral 2, respectively.

For the BDI items, the squared multiple correlations (SMC; R^2_{smc}) of variables were the percentages that could be explained by a latent variable factor. For example, the SMC of the 'sadness' item (D1) was higher in the FRG group (0.754) than in the placebo group (0.252), thus the variance in this item can be explained by the variance in the CD factor in 75.4% of the FRG group, and in 25.2% of the placebo group (Fig. 2).

In this structural regression model, the SMC of the dependent (resultant) latent variable shows the percentage of direct effects of independent (causative) latent variables on the dependent (resultant) latent variable.³¹ The SMCs of CD and SD were higher in the FRG group (0.395 and 0.550, respectively) than in the placebo group (0.108 and 0.151, respectively). The standardized correlation coefficients between CD and SD were higher in the FRG group (0.829, $P = 0.005$) than in the placebo group (-0.143 , $P = 0.629$).

Discussion

Several studies have reported that consumption of ginseng or red ginseng improves the symptoms of depression. However, such changes have been shown by studies on blood flow³² or a forced swimming test in mice.³³ They have not been shown by the emotional state of humans. Depression is normally measured by questionnaires such as the self-report BDI. The BDI is one of the most widely used instruments for evaluation of depressive symptoms, both in healthy people and in those who have been diagnosed with an emotional disorder.³⁰

Hormone factor and BDI factors

In this study, the factor labelled hormone comprised measurement of adrenocorticotrophic hormone and cortisol. Even though it is well known that patients with depression have a high level of cortisol, the level of cortisol in this study showed a negative causal relation with depression. These results may have been due to the relative health of the participants of this study, who all seemed to be healthy women without depression, as reflected in the low BDI values. Therefore, it is possible that cortisol levels in the normal range may act as a co-reliever of the symptoms of depression.

Energy factor and BDI factors

Studies have reported that a low level of either LDL or total cholesterol has a positive correlation with depression.³⁴ These reports are connected with several hypotheses: serotonin uptake in the serotonin receptors of cell membranes,³⁵ the supply of polyunsaturated fatty acids, especially omega 3 fatty acid,³⁶ the endocannabinoid receptors in the lipid raft,³⁷ and the myelination of neurons.³⁸

Bouwman *et al.* reported that the symptoms of depression had a positive correlation with defectiveness in glucose metabolism.³⁹ However, in the current study, the factor loading of total and LDL cholesterol on the energy factor (1.236 and 1.000) was higher than the factor loading of blood glucose on the energy factor (0.166). A possible explanation is that sampling occurred after 8 hours of overnight

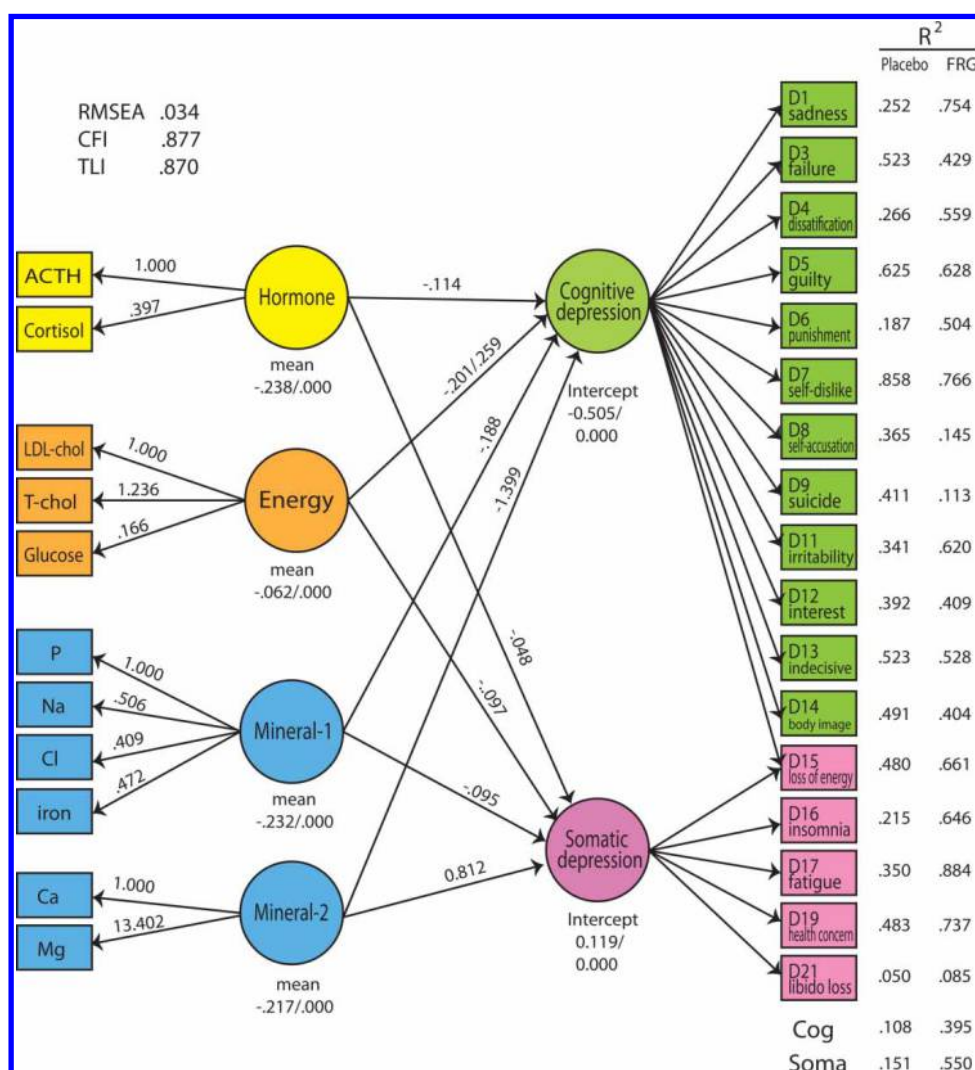


Figure 2 Final model. The structural regression path of the energy factor on the CD factor was freely analysed and the other structural regression paths were constrained for equality between FRG group and placebo group. The numbers on the arrow line represent the estimates of structural regression paths and factor loadings. R^2 , squared multiple correlations.

fasting in this study. Postprandially, blood glucose, which is regulated by insulin, is the major energy source, whereas after fasting, blood lipids are the major factor for energy metabolism. One of the major regulators of blood lipids during fasting is growth hormone.⁴⁰ However, because circulating free fatty acids cannot be used in the brain as an energy source, the level of lipids cannot explain their relation with depression.

Even though ketone bodies were not analysed in this study, they are one possible solution to the enigma of the relationship between lipids and depression, because ketone bodies are a major source of energy in the brain.⁴¹ Normally, the concentrations of ketone bodies have a positive correlation with free fatty acids, and the level of blood ketone bodies increases from a postprandial state (6 mmol/l) to a fasting state (25 mmol/l).⁴²

In this study, the energy factor had a negative causal relation with CD in the placebo group (-0.169), but the relationship was positive in the FRG group

(0.259), which may imply an increase in the efficiency of energy metabolism in the FRG group.

Conclusion

The factor coefficients of the energy factor on CD showed a significant difference between the FRG group and the placebo group. The factor loading of LDL and total cholesterol on the energy factor were higher than that of glucose on the energy factor. Cholesterol can be interpreted as the representation of lipids and lipid derivatives, including ketone bodies.

Even though the SMCs for CD and SD in the placebo group were small, the hormones, energy, and mineral factors showed different effects on some BDI factors. Some of the remaining portions of the SMCs can be explained by other variables, including prostaglandin, E2, and DHEAS.

The ginsenosides used in this study were fermented so that there would be more bioactivation and ease of absorption. Since the effects of steroidal ginsenoside depend on the type, dose, ratio, and time of the

ginsenoside treatment, a further study with a single type of ginsenoside would be helpful, as well as a comparative study between patients with depression and a healthy control group.

Acknowledgment

We thank Mr John Mensing, 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.

References

- WHO. What is depression? WHO Available from: http://www.who.int/mental_health/management/depression/definition/en/, 2012.
- DEPRESSION. A Global Crisis http://www.who.int/mental_health/management/depression/wfmh_paper_depression_wmhd_2012.pdf.
- Booij L, Van der Does AJ, Riedel WJ. Monoamine depletion in psychiatric and healthy populations: review. *Mol Psychiatry* 2003;8(12):951–73.
- Ruhé HG, Mason NS, Schene AH. Mood is indirectly related to serotonin, norepinephrine and dopamine levels in humans: a meta-analysis of monoamine depletion studies. *Mol Psychiatry* 2007;12(4):331–59.
- Gold PW, Chrousos GP. Organization of the stress system and its dysregulation in melancholic and atypical depression: high vs low CRH/NE states. *Mol Psychiatry* 2002;7(3):254–75.
- Fanselow MS. Contextual fear, gestalt memories, and the hippocampus. *Behav Brain Res* 2000;110(1–2):73–81.
- Manji HK, Quiroz JA, Sporn J, Payne JL, Denicoff KA, Gray N, *et al.* Enhancing neuronal plasticity and cellular resilience to develop novel, improved therapeutics for difficult-to-treat depression. *Biol Psychiatry* 2003;53(8):707–42.
- Noguchi KK, Lau K, Smith DJ, Swiney BS, Farber NB. Glucocorticoid receptor stimulation and the regulation of neonatal cerebellar neural progenitor cell apoptosis. *Neurobiol Dis* 2011;43(2):356–63.
- Hamilton JP, Siemer M, Gotlib IH. Amygdala volume in major depressive disorder: a meta-analysis of magnetic resonance imaging studies. *Mol Psychiatry* 2008;13(11):993–1000.
- Sheline YI, Gado MH, Kraemer HC. Untreated depression and hippocampal volume loss. *Am J Psychiatry* 2003;160(8):1516–8.
- Bonne O, Krausz Y, Shapira B, Bocher M, Karger H, Gorfine M, *et al.* Increased cerebral blood flow in depressed patients responding to electroconvulsive therapy. *J Nucl Med* 1996;37(7):1075–80.
- Vahdat Shariatpanaahi M, Vahdat Shariatpanaahi Z, Moshtaaghi M, Shahbaazi SH, Abadi A. The relationship between depression and serum ferritin level. *Eur J Clin Nutr* 2007;61(4):532–5.
- Nguyen PH, Grajeda R, Melgar P, Marcinkevage J, DiGirolamo AM, Flores R, *et al.* Micronutrient supplementation may reduce symptoms of depression in Guatemalan women. *Arch Latinoam Nutr* 2009;59(3):278–86.
- Iosifescu DV, Bolo NR, Nierenberg AA, Jensen JE, Fava M, Renshaw PF. Brain bioenergetics and response to triiodothyronine augmentation in major depressive disorder. *Biol Psychiatry* 2008;63(12):1127–34.
- Eby GA, Eby KL. Rapid recovery from major depression using magnesium treatment. *Med Hypotheses* 2006;67(2):362–70.
- Mintel. Complementary and alternative medicines-US-July 2008. Chicago, IL: Mintel Intl; 2008.
- Lee Y, Jin Y, Lim W, Ji S, Choi S, Jang S, *et al.* A ginsenoside-Rh1, a component of ginseng saponin, activates estrogen receptor in human breast carcinoma MCF-7 cells. *J Steroid Biochem Mol Biol* 2003;84(4):463–8.
- Hien TT, Kim ND, Pokharel YR, Oh SJ, Lee MY, Kang KW. Ginsenoside Rg3 increases nitric oxide production via increases in phosphorylation and expression of endothelial nitric oxide synthase: essential roles of estrogen receptor-dependent PI3-kinase and AMP-activated protein kinase. *Toxicol Appl Pharmacol* 2010;246(3):171–183.
- Niu CS, Yeh CH, Yeh MF, Cheng JT. Increase of adipogenesis by ginsenoside (Rh2) in 3T3-L1 cell via an activation of glucocorticoid receptor. *Horm Metab Res* 2009;41(4):271–6.
- Yang CS, Ko SR, Cho BG, Shin DM, Yuk JM, Li S, *et al.* The ginsenoside metabolite compound K, a novel agonist of glucocorticoid receptor, induces tolerance to endotoxin-induced lethal shock. *J Cell Mol Med* 2008;12(5A):1739–53.
- Wu J, Pan Z, Wang Z, Zhu W, Shen Y, Cui R, *et al.* Ginsenoside Rg1 protection against beta-amyloid peptide-induced neuronal apoptosis via estrogen receptor alpha and glucocorticoid receptor-dependent anti-protein nitration pathway. *Neuropharmacology* 2012;63(3):349–61.
- Kim SO, You JM, Yun SJ, Son MS, Nam KN, Hong JW, *et al.* Ginsenoside rb1 and rg3 attenuate glucocorticoid-induced neurotoxicity. *Cell Mol Neurobiol* 2010;30(6):857–62.
- Kwak Y-S, W. J-J, Hwang S-Y, Kyung J-S, Kim S-K. Effect of crude saponin fraction from Korean red ginseng on physiological functions of old female rat. *J Korean Soc Food Sci Nutr* 2000;29(3):460–5.
- Vuksan V, Sung MK, Sievenpiper JL, Stavro PM, Jenkins AL, Di BM, *et al.* Korean red ginseng (*Panax ginseng*) improves glucose and insulin regulation in well-controlled, type 2 diabetes: results of a randomized, double-blind, placebo-controlled study of efficacy and safety. *Nutr Metab Cardiovasc Dis* 2008;18(1):46–56.
- Lee HJ, Lee YH, Park SK, Kang ES, Kim HJ, Lee YC, *et al.* Korean red ginseng (*Panax ginseng*) improves insulin sensitivity and attenuates the development of diabetes in Otsuka long-evans Tokushima fatty rats. *Metabolism* 2009;58(8):1170–7.
- Schacke H, Docke WD, Asadullah K. Mechanisms involved in the side effects of glucocorticoids. *Pharmacol Ther* 2002;96(1):23–43.
- Muthén B. Latent variable structural equation modeling with categorical-data. *J Econometrics* 1983;22(1–2):43–65.
- Rubin DB. Illustrating the use of multiple imputations to handle nonresponse in sample surveys. *Proceedings of the 42nd Session of the International Statistical Institute*; 1979. p. 517–32.
- Poole H, Bramwell R, Murphy P. Factor structure of the Beck depression inventory-II in patients with chronic pain. *Clin J Pain* 2006;22(9):790–8.
- Beck AT, Steer RA, Carbin MG. Psychometric properties of the Beck depression inventory: twenty-five years of evaluation. *Clin Psychol Rev* 1988;8(1):77–100.
- Kline RB. Principles and practice of structural equation modeling. NY: The Guilford Press; 2005. p. 219.
- Choi HK, Seong DH, Rha KH. Clinical efficacy of Korean red ginseng for erectile dysfunction. *Int J Impot Res* 1995;7(3):181–6.
- Kim NH, Kim KY, Jeong HJ, Kim HM. Antidepressant-like effect of altered Korean red ginseng in mice. *Behav Med* 2011;37(2):42–6.
- Aijanseppa S, Kivinen P, Helkala EL, Kivela SL, Tuomilehto J, Nissinen A. Serum cholesterol and depressive symptoms in elderly Finnish men. *Int J Geriatr Psychiatry* 2002;17(7):629–34.
- Alvarez JC, Cremniter D, Lesieur P, Gregoire A, Gilton A, Macquin MI, *et al.* Low blood cholesterol and low platelet serotonin levels in violent suicide attempters. *Biol Psychiatry* 1999;45(8):1066–9.
- Sinclair AJ, O'Dea K, Dunstan G, Ireland PD, Niall M. Effects on plasma lipids and fatty acid composition of very low fat diets enriched with fish or kangaroo meat. *Lipids* 1987;22(7):523–9.
- Freund TF, Katona I, Piomelli D. Role of endogenous cannabinoids in synaptic signaling. *Physiol Rev* 2003;83(3):1017–66.
- Merklér D, Klinker F, Jurgens T, Glaser R, Paulus W, Brinkmann BG, *et al.* Propagation of spreading depression inversely correlates with cortical myelin content. *Ann Neurol* 2009;66(3):355–65.
- Bouwman V, Adriaanse MC, van't Riet E, Snoek FJ, Dekker JM, Nijpels G. Depression, anxiety and glucose metabolism in the general dutch population: the new Hoorn study. *PLoS One* 2010;5(4):e9971.
- Moller N, Jorgensen JO. Effects of growth hormone on glucose, lipid, and protein metabolism in human subjects. *Endocr Rev* 2009;30(2):152–77.

- ▶41 Fukao T, Lopaschuk GD, Mitchell GA. Pathways and control of ketone body metabolism: on the fringe of lipid biochemistry. *Prostaglandins Leukot Essent Fatty Acids* 2004;70(3):243–51.
- ▶42 Owen OE, Felig P, Morgan AP, Wahren J, Cahill GF, Jr. Liver and kidney metabolism during prolonged starvation. *J Clin Invest* 1969;48(3):574–83.

Appendix 1: Anthropometric data of participants

	Mean ± S.E.		P
	FRG (n = 49)	Placebo (n = 44)	
Age (years)	58.4 ± 5.5	58.4 ± 5.9	1.00
Weight (kg)	57.1 ± 6.7	57.6 ± 6.6	0.80
Height (cm)	157.7 ± 5.3	156.5 ± 5.3	0.26
BMI (kg/m ²)	22.9 ± 2.4	23.6 ± 2.5	0.17
Waist circumference (cm)	32.8 ± 2.4	33.4 ± 2.3	0.10
Hip circumference (cm)	37.0 ± 1.9	37.4 ± 2.0	0.30
Waist/hip	0.89 ± 0.03	0.89 ± 0.04	0.31

Appendix 2: Reagent and instrument

Test name		Glucose (S)	Insulin	Cholesterol, total	LDL cholesterol	Na (Sodium)
Test method		Enzymatic method	ECLIA	Enzymatic, colorimetry	Enzymatic, colorimetry	Ion-selective electrodes
Reagent	Kit name	Glucose Hexokinase	Insulin	Cholesterol reagent	LDL-cholesterol	Na ion selective electrode
	Company, nationality	Bayer, USA	Roche, Germany	Bayer, USA	Bayer, USA	Bayer, USA
Analytical instrument	Instrument name	ADVIA	MODULAR ANALYTICS	ADVIA	ADVIA	ADVIA
	Model name	ADVIA 1650	E170	ADVIA 1650	ADVIA 1650	ADVIA 1650
	Company, nationality	Bayer, Japan	Roche, Germany	Bayer, Japan	Bayer, USA	Bayer, Japan
Test name		Iron	Cl	K (Potassium)	Phosphorus	Calcium
Test method		Colorimetric assay	ion-selective electrodes	ion-selective electrodes	Phosphomolybdate & UV	Colorimetry
Reagent	Kit name	Fe	ion selective electrode	ion selective electrode	Inorganic phosphorus reagents	CALCIUM reagents
	Company, nationality	Roche, Germany	Bayer, USA	Bayer, USA	SIEMENS, USA	SIEMENS, USA
Analytical instrument	Instrument name	Modular Analytics	ADVIA	ADVIA	ADVIA	ADVIA
	Model name	P	ADVIA 1650	ADVIA 1650	ADVIA 1650	ADVIA 1650
	Company, nationality	Roche, Germany	Bayer, Japan	Bayer, Japan	SIEMENS, USA	SIEMENS, USA
Test name		Cortisol	ACTH	DHEA-S	oestrogen (E2)	HGH (S)
Test method		RIA	IRMA	RIA	CLIA	CLIA
Reagent	Kit name	Coat-A-count Cortisol	ACTH IRMA	Coat-A-Count DHEA-sulphate	ADVIA centaur oestradiol	Immulate 2000 GH
	company, nationality	SIEMENS, USA	BRAHMS, Germany	SIEMENS, USA	Bayer, USA	DPC, USA
Analytical instrument	Instrument name	r-counter	r-counter	r-counter	Centaur	Immulate
	Model name	COBRA 5010 QUANTUM	COBRA5010 Quantum	COBRA5010 Quantum	ADVIA Centaur	Immulate 2000
	Company, nationality	PACKARD, USA	PACKARD, USA	PACKARD, USA	Bayer, USA	DPC, USA

Appendix 3: Equality constrained of models and goodness-of-fit statistics

Model	Continuous variables	Categorical variables	RMSEA	CFI	TLI
Model 1	1.Configural invariance: (1) freely analysed factor loading, intercept, variance (2) factor mean = 0 across group	(1) Freely analysed factor loadings and thresholds. (2) Fix the scale factors at 1 across group	0.033	0.888	0.875
Model 2	2.Metric invariance: (1) freely analysed intercept, variance fix factor mean at 0 across group (3) test equality constrained of factor loading across group	(3) Equality constrained mean at 0 across group	0.034	0.883	0.871
Model 3	4.Intercept scalar invariance: (1) freely analysed factor mean across group (2) equality constrained factor loading, intercept across group		0.034	0.882	0.872
Model 4	4.Intercept scalar invariance: (1) freely analysed factor mean across group (2) equality constrained factor loading, variance, intercept across group	(1) Equality constrained factor loadings and thresholds across group (2) Freely analysed scale factors and means in second group	0.034	0.879	0.872
Model 5	5. Test structural regression path estimate across group		0.033	0.883	0.877