CLINICAL EVALUATION OF GUILU ERXIAN JIAO IN TREATING PERIMENOPAUSAL SYNDROME

Pin-Han Wang[1], Yi-Chiu Li[1], Yi-Hui Wu[1], Jiun-Liang Chen[1,2], Jian-Tai Qiu[3,4], Sien-Hung Yang[1,2,*]

[1]Center for Traditional Chinese Medicine, Chang Gung Memorial Hospital, Taoyuan, Taiwan

[2]School of Traditional Chinese Medicine, Chang Gung University, Taoyuan, Taiwan

[3]Department of Obsterics and Gynocology, Chang Gung Memorial Hospital, Taoyuan, Taiwan

[4]Department of Biomedical Sciences, School of Medicine, Chang Gung University, Taoyuan, Taiwan

(Received 29th February 2012, accepted 30th July 2012)

In Taiwan, approximately 46% of women during perimenopause have experienced perimenopausal syndrome, which mostly includes insomnia, hot flushes and sweating, and palpitation. Hormonal replacement therapy (HRT) is the mainstream treatment of perimenopausal syndrome. However, many women suffering from perimenopausal syndrome would prefer to take Guilu Erxian Jiao (GEJ) rather than receive HRT. Therefore, the aim of this study is to determine whether GEJ helps relieve perimenopausal syndrome. We treated perimenopausal women who suffered from perimenopausal symptoms and divided them into three groups, administering GEJ at 200 mg per day, 100 mg per day, and a placebo (GEJ-free starch powder), all of which were in the same capsule form with one capsule administered per day for 2 months. Serum estradiol and follicle-stimulating hormone (FSH) levels were tested at three stages: pre-treatment, 1-month post-treatment, and 2-month post-treatment. The clinical symptoms were estimated using the questionnaire of physical and mental symptoms during perimenopause before and after 2 months of treatment. After 2 months of treatment, serum estradiol levels were significantly elevated in the high-dose group (200 mg/day) ($P < 0.05$). However, no significant differences of serum FSH levels were observed among the three groups after GEJ treatment. Although improvements were observed in both the high-dose and low-dose groups, more amelioration in clinical symptoms was noted in the low-dose group (100 mg/day) than in the high-dose group, especially for hectic sweats and palpitation. GEJ is effective for relieving perimenopausal syndrome, and taking high-dose GEJ once per day for 2 months elevates more serum estradiol levels than does taking a low dose. Therefore, we concluded that GEJ is beneficial for treating perimenopausal syndrome.

Key words: traditional Chinese medicine, perimenopause, Guilu Erxian Jiao, estradiol

*__Correspondence to:__ Sien-Hung Yang, Division of Chinese Internal Medicine, Center for Traditional Chinese Medicine, Chang Gung Memorial Hospital, No. 123, Dinghu Rd., Guishan Township, Taoyuan County 33378, Taiwan, Tel: +886-3-3196200 ext. 2611, Fax: +886-3-3298995, E-mail: dryang@adm.cgmh.org.tw

Introduction

Menopause occurs when the oocytes in a woman's ovaries deplete and ovulation ceases[1]. Women experience a hormonal change at approximately this time. Because of decreasing hormone secretion, women suffer from physical and mental discomfort. This is called perimenopausal syndrome[2], which can be generally classified into two categories. The first is physical symptoms, such as hot flushes, night sweats, headaches with dizziness, chest tightness, palpitation, joint and bone pain, and a dry sensation in the vagina. The second is psychological symptoms, such as insomnia, irritability, depression or a sense of loss, and general weakness[3,4]. In recent decades, researchers have classified physical perimenopausal syndrome further into specific categories according to general, to vasomotor , and to genitourinary properties[5-8].

In Europe and the United States, 80% of perimenopausal women experienced hot flushes[9], and the symptom lasted from half a year to five years[10]. In Asia, the symptom of hot flushes in perimenopausal women has been less common but has still occurred, ranging from 17.6%[11], 24.2%[12], to 38%[13], in different studies. A multinational study in Asia, including 22 investigational centers in China, Hong Kong, Indonesia, South Korea, Malaysia, Pakistan, the Philippines, Singapore, Taiwan, Thailand, and Vietnam, revealed a 62.7% prevalence rate of hot flush symptoms[14]. Although perimenopausal syndrome ceases after a few years, numerous diseases such as urethritis, urine incontinence, osteoporosis, atherosclerosis, and cardiovascular disease may be induced by insufficient hormone secretion after menopause[15-17], greatly influencing women's health and life quality.

Menopausal transition is the period from variable menstrual cycle length to skipped cycles and then to amenorrhea[1]. According to recent studies, the average age of female menopause in Taiwan ranges from 49.5 to 49.8 years old[18-20]. Among the cases in these studies, 46% of self-described menopausal women have experienced perimenopausal syndrome, and the most common symptoms include insomnia, hot flushes, sweating, palpitation, irritable temper, depression, dyspareunia, lumbago, and general weakness[13,18,20].

Hormone replacement therapy (HRT) has been frequently used since the 1970s for alleviating perimenopausal syndrome and preventing osteoporosis in menopausal woman[10,21,22]. Nevertheless, increased adverse or even harmful effects were reported by a large-scale randomized controlled trial performed by the Women's Health Initiative. The report showed that after 5.2 years of follow-up of hormone therapy, combined an oral estrogen plus progestin supplement resulted in harms rather than benefits in menopausal women. The treatment was found to increase the risk of coronary heart disease and invasive breast cancer. However, the study also revealed that combined an oral estrogen plus progestin supplement could reduce the risk of osteoporosis, hip fracture, and colorectal cancer[17].

According to the Inner Cannon of the Yellow Emperor, the most classical book in traditional Chinese medicine (TCM), the essence of Ren and Taichong Meridian, and the essence of the kidney are exhausted during a female's late 40s. The physiological function of the perimenopausal phase is characterized by ceased menstruation and reproductive function. The basis of the pathophysiology of menopausal syndrome results from the emptying

of the yin, yang, and qi of the kidney. Therefore, treatment strategies rely on nourishing the essence of the kidney and refilling the vitality of the kidney. The prescription Guilu Erxian Jiao (GEJ) originates from *Yi Fang Ji Jie* (*The Collected Explanation of Medical Prescription*). The four major components of GEJ are *Cornu Cervi, Carapax et Plastrum Chrysemys, Lycii Fructus,* and *Ginseng Radix*. According to the traditional Chinese records on GEJ, the efficacies involve supplying the essence and marrow, replenishing vital energy, and refreshing the mind. GEJ is one of the best prescriptions for deficiency of both yin and yang in the kidney, and exhausted essence and blood in Ren and Du Meridians, emaciation of the body. Clinically, GEJ has been commonly used to treat general weakness, fatigue, wilting and weak waist and knees, blurred vision, neurasthenia, erectile disorders, anaphrodisia, spermatorrhea, semen abnormality[23], edema, dysuria, urinary incontinence, menstrual disorders[24], menopause, and postpartum urinary retention[25]. GEJ has also been used to treat anti-aging[26], chronic renal disease[27], diabetes mellitus, osteopenia[28], and stroke sequel[29].

Many previous studies have mentioned the effects of the four gradients in GEJ. *Ginseng Radix* helps delay the aging process[30,31], excites the central nervous system[32], improves immunity[33], alleviates hemopoiesis[34], and suppresses cancer cell proliferation[35]. *Lycii Fructus* enhances immunity function and anti-aging[36]. *Carapax et Plastrum Chrysemys* can modulate immunity[27], prevent apoptosis of certain neurons in the central nervous system[37], and promotes mesenchymal stem cell differentiation into osteoblasts[38]. *Cornu Cervi* is beneficial for increasing hemoglobin and elevating the concentration of platelets, erythrocytes, and leukocyte counts[39]. However, few studies have investigated the effects of GEJ on menstrual regulation and perimenopausal disorder treatment[40].

Because of rapid developments in marketing and advertising, traditional Chinese prescriptions are considered as healthy food or medication treatment for climacteric women. Therefore, instead of receiving hormone preparations, many women choose GEJ as an alternative medicine. Because numerous patients suffering from perimenopausal syndrome purchase and receive GEJ as an over-the-counter (OTC) drug, we considered it is necessary to study the efficacy of GEJ in climacteric women treatment. Therefore, the objective of this study was to evaluate whether GEJ alleviates perimenopausal syndrome and modulates hormones in women.

Materials and Methods

I. Participants

We recruited 65 healthy female volunteers, aged 40 to 59 years old who had regular menstruation but with any one of the menopausal symptoms such as hot flushes, palpitation, insomnia, irritable moods, depression, general weakness, fatigue, low back pain, joints pain, and a dry sensation in the vagina; or who had already suffered from irregular menstruation or amenorrhea. The participants were excluded if they had abnormal liver and renal function, uncontrolled diabetes and hypertension, major infections, cancer diseases, during radiotherapy or chemotherapy, taken immunosuppressive agents, or gotten pregnant. All of the participants were required not to receive hormone supplements or any health foods that were proven to have hormonal effects one week before participating in the study and during the 2-month treatment.

II. Herb preparation

The herbs and placebo were prepared by the Chuang Song-Zong pharmaceutical factory, a TCM Good Manufacturing Practice (GMP) manufacturer certified in Taiwan. The batch number was RD-EG689-701. The ingredients of the medicinal herbs in the GEJ were *Cornu Cervi , Carapax et Plastrum Chrysemys, Lycii Fructus,* and *Ginseng Radix et Rhizoma Rubra.* The weight proportion of these medicinal herbs was 10:5:1.1:0.9, sequentially. The preparation procedure of GEJ in this study was as follows. The raw herbs, *Cornu Cervi* and *Carapax et Plastrum Chrysemys*, were stewed using gentle heat for around one week, and *Lycii Fructus* and *Ginseng Radix et Rhizoma Rubra* were then added. The mixture was stewed further, then filtered and concentrated into a paste. The concentrated paste was later mixed with the starch and finally dried to a fine GEJ powder. The total weight of the original raw herbs when compared to the concentrated paste was 5.67:1. The high-dose group was composed of 266.8 mg of the concentrated paste and the low-dose group was composed of 133.4 mg of the concentrated paste. A GEJ-free powder was also made, mainly of corn starch, microcrystalline cellulose, caramel, and ginseng perfume oil (so that the appearance and smell resembled those of the GEJ powder). It was used as the padding in the high-dose, low-dose, and placebo groups. Therefore, all of the capsules in the three groups were of equal weight.

High performance liquid chromatography (HPLC) was used to ensure the quality (standard effective components) of the GEJ.

III. Study design

Sixty-five participants were recruited and each was randomly assigned to three groups: 22 participants in the high-dose group (200 mg/day); 22 participants in the low-dose group (100 mg/day); and 21 participants in the placebo group. All of the participants received the treatment for 2 months. The medicine in all 3 groups was administered orally in the form of capsules; the dosage was one capsule taken once per day. All participants were evaluated according to serum follicle-stimulating hormone (FSH) levels, estradiol concentration, and the questionnaire of 25 clinical menopausal symptoms. The evaluation of serum FSH and estradiol concentration were performed before and immediately after the treatment. The questionnaire evaluation was performed at the beginning of the treatment, at one month during the treatment, and at the end of the treatment. The concentration of serum hormone levels were entrusted to and tested by the Department of Laboratory Medicine, Chang Gung Memorial Hospital (CGMH).

IV. Questionnaire

The questionnaire was completed by the participants orally with instruction and assistance, and was administered 3 times, at pre-treatment, one month during the treatment, and two months during the treatment (treatment completion). The questionnaire in the study was based mainly on the Menopause-Specific Quality of Life (MENQOL) Questionnaire[41], Greene Climacteric Scale[6], Menopause Rating Scale (MRS)[5], and study of menopausal symptoms in Taiwan[13]. Some clinical symptoms frequently observed in TCM were also added to the questionnaire (Table 1). The items of the questionnaire constituted four major domains of menopausal symptoms: vasomotor, physical, urogenital, and psychological (Table 2).

Table 1. Questionnaire of physical and mental symptoms during perimenopause.

Physical and mental symptoms during perimenopause	Severity				
	None	Mild	Moderate	Severe	Very Severe
1. Abnormal Hot Sensation (Hot Flushes)	0	1	2	3	4
2. Insomnia	0	1	2	3	4
3. Palpitation	0	1	2	3	4
4. Tiredness	0	1	2	3	4
5. Headache, Dizziness	0	1	2	3	4
6. Night Sweats	0	1	2	3	4
7. Depressed Moods or a Sense of Loss	0	1	2	3	4
8. Vaginitis	0	1	2	3	4
9. Tends to Get Urinary Tract Infection	0	1	2	3	4
10. Pain during Sexual Intercourse	0	1	2	3	4
11. Urinary Incontinence	0	1	2	3	4
12. Allergies	0	1	2	3	4
13. Dry Mouth	0	1	2	3	4
14. Hectic Sweats	0	1	2	3	4
15. Constipation	0	1	2	3	4
16. Diarrhea	0	1	2	3	4
17. Chest Tightness	0	1	2	3	4
18. Elevated Blood Pressure	0	1	2	3	4
19. Decreased Blood Pressure	0	1	2	3	4
20. Facial Flushing	0	1	2	3	4
21. Pale Appearance	0	1	2	3	4
22. Joint And Bone Pain	0	1	2	3	4
23. Being Depressed And Suspicious	0	1	2	3	4
24. General Weakness	0	1	2	3	4
25. Irritable	0	1	2	3	4

V. Statistical analysis

To test the significance of changing, statistical analysis for serum FSH and estradiol concentration was applied using a paired-sample t-test because there were two time points of investigation in this study. Three groups were in this study and each group was evaluated by administering the questionnaire three times (before treatment, during treatment, and post treatment). The varying original statistics of the 3 groups (high-dose, low-dose, and placebo groups) made comparing the symptom improvements among the 3 groups difficult. Hence, we analyzed the statistics of the questionnaire at 3 different time points (before treatment, during treatment, and post treatment) for each group by performing one-way ANOVA.

Table 2. Questionnaire analysis of physical and mental symptoms during perimenopause.

Physical and mental symptoms during perimenopause	vasomotor	physical	urogenital	psychological
1. Abnormal Hot Sensation (Hot Flushes)	V			
2. Insomnia				V
3. Palpitation		V		
4. Tiredness		V		
5. Headache, Dizziness		V		
6. Night Sweats	V			
7. Depressed Moods or a Sense of Loss				V
8. Vaginitis			V	
9. Tends to Get Urinary Tract Infection			V	
10. Pain during Sexual Intercourse			V	
11. Urinary Incontinence			V	
12. Allergies		V		
13. Dry Mouth		V		
14. Hectic Sweats	V			
15. Constipation		V		
16. Diarrhea		V		
17. Chest Tightness		V		
18. Elevated Blood Pressure		V		
19. Decreased Blood Pressure		V		
20. Facial Flushing	V			
21. Pale Appearance		V		
22. Joint And Bone Pain		V		
23. Being Depressed And Suspicious				V
24. General Weakness		V		
25. Irritable				V

Table 3. Average age in each group.

Group	High-dose (200 mg/day)	Low-dose (100 mg/day)	Placebo
Age (years old)	50.0 ± 3.79	49.7 ± 5.28	48.4 ± 5.69

Results

Sixty-five female participants were recruited. Among whom, the high-dose group contained 22 participants, but 3 participants withdrew during the experiment; 1 participant withdrew in the first month because she could not sustain the discomforts arising from holding the HRT, which she received previously; and 2 participants withdrew in the second month because one held the treatment by herself and the other lost of follow up. The low-dose group contained 21 participants, but 1 participant withdrew during the

experiment in the second month because she was too busy to receive the treatment regularly (Figure 1). The placebo group contained 21 participants and no one withdrew. Table 3 lists the average age of the participants in each group. The menstruation states and number of participants in each state are listed in Table 4.

According to the experimental results and those of the paired t-test (used to determine the significant differences between pre-treatment and post-treatment conditions in each group), after 2 months of treatment, for the high-dose group, the serum estradiol concentration was significantly increased when compared to the placebo group ($P < 0.05$) (Table 5). For the low-dose

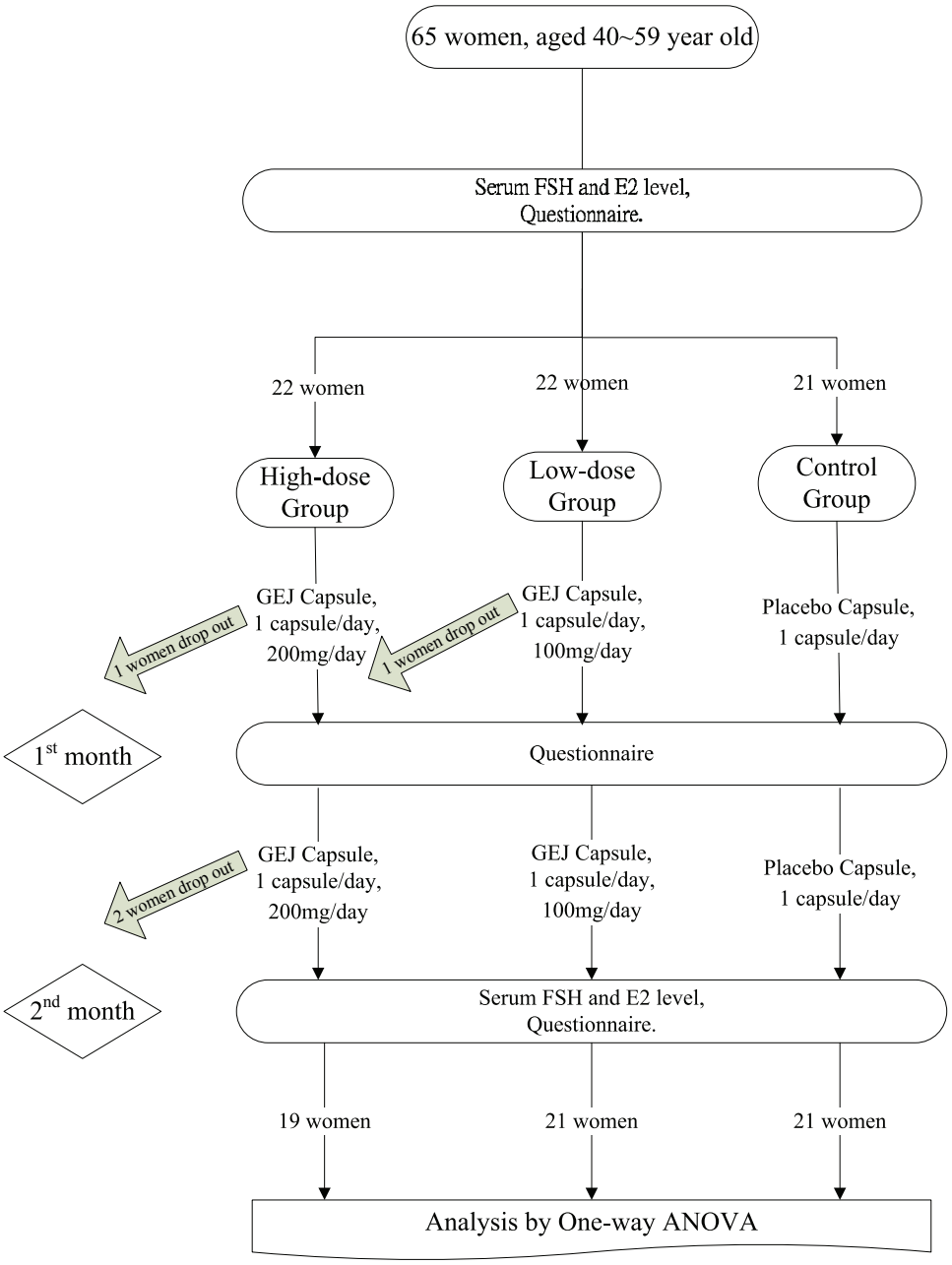

Fig. 1. Study framework. GEJ: Guilu Erxian Jiao, FSH: follicle-stimulating hormone, E2: estradiol.

Table 4. State of menstruation.

	State	Numbers of participants
1	Still having menstrual cycles	19
2	Irregular menstrual cycles	14
3	Last natural final menstruation period in one year	5
4	Last natural final menstruation period at one year before, or longer	19
5	Menopause due to surgery, radiation therapy, or chemotherapy	4

Table 5. Pre-treatment and post-treatment of estradiol (E2) and follicle-stimulating hormone (FSH) level in groups. For each group, a paired t-test was conducted to test the significant difference between pre-treatment and post-treatment. The asterisk (*) shows the significance with *P*-value < 0.05.

Group	High-dose (200 mg/day)			Low-dose (100 mg/day)			Placebo		
	Pre-treatment	Post-treatment	*P*-value	Pre-treatment	Post-treatment	*P*-value	Pre-Treatment	Post-treatment	*P*-value
1. E2 (pg/ml)	35.3 ± 54.2	64.1 ± 70.9	0.048*	34.0 ± 30.7	54.3 ± 40.9	0.21	36.2 ± 26.1	40.3 ± 31.5	0.07
2. FSH (mIU/ml)	39.2 ± 10.2	41.9 ± 12.7	0.37	33.6 ± 29.4	35.5 ± 30.0	0.7	39.7 ± 32.1	36.9 ± 33.1	0.45

group, after 2 months of treatment, the estradiol increased as well; but there was no significant difference when compared to the placebo group. Regarding serum FSH level changes, no significant difference was found in the three groups.

The results of physical and mental symptoms of menopause are shown in Table 6. The patients receiving high-dose GEJ had significant improvement in facial flushing, allergies, chest tightness, general weakness, joint and bone pain, insomnia, and being depressed and suspicious ($P < 0.05$). The patients receiving low-dose GEJ had significant improvement in hectic sweats, palpitation, allergies, chest tightness, general weakness, joint and bone pain, and depressed moods or a sense of loss ($P < 0.05$). Regarding physical symptoms, both the high-dose and low-dose groups had improved allergies, chest tightness, general weakness, and joint and bone pain; and the low-dose group also had alleviated palpitations. Regarding vasomotor symptoms, there was a significant difference in facial flushing in the high-dose group, and in hectic sweating in the low-dose group ($P < 0.05$), but both dosages could not alleviate hot flushes. Regarding urogenital symptoms, both dosages in this study could not improve them. Regarding psychological symptoms, the more meaningful finding is that high dosages could alleviate insomnia.

Further analyzing the symptoms significantly improved ($P < 0.05$), as shown in Figures 2 and 3, in both the high-dose and low-dose groups, reduction of joint and bone pain after 2 months of treatment was higher than after 1 month of treatment. Comparing both groups, the reduction in the high-dose group was more obvious than in the low-dose group; one half degree of pain reduction in the first month and one third of pain reduction was noted in the second month. This was compatible with the general clinical practice of using GEJ for relieving osteoarthritis and other

Table 6. Statistics of 25 common physical and mental symptoms around menopause. For each group, one-way ANOVA was used to test the significance of difference between the three different times (before treatment, during treatment, and post treatment) within the same group. The asterisk (*) shows the significance with $P < 0.05$.

Group	High-dose (200mg)				Low-dose (100mg)				Placebo			
	Pre-treatment	1-month Post-treatment	2-month Post-treatment	*P*-value	Pre-treatment	1-month Post-Treatment	2-month Post-treatment	*P*-value	Pre-treatment	1-month Post-treatment	2-month Post-treatment	*P*-value
1. Abnormal Hot Sensation (Hot Flushes)	0.58 ± 0.51	0.33 ± 0.49	0.58 ± 0.67	0.109	0.94 ± 1.2	0.82 ± 1.2	0.72 ± 1.1	0.27	1 ± 1.22	0.59 ± 1.18	0.88 ± 1.32	0.68
2. Insomnia	1 ± 0.85	0.17 ± 0.39	0.42 ± 0.67	0.047*	0.17 ± 0.98	0.53 ± 0.7	0.94 ± 0.94	0.49	1.06 ± 1.34	0.76 ± 1.44	1 ± 1.32	0.81
3. Palpitation	0.67 ± 0.65	0.17 ± 0.39	0.5 ± 0.8	0.56	0.89 ± 0.8	0.5 ± 0.85	0.35 ± 0.86	0.03*	0.7 ± 0.59	0.24 ± 0.44	0.41 ± 0.62	0.06
4. Tiredness	1 ± 0.95	0.33 ± 0.5	0.67 ± 0.65	0.3	1.44 ± 0.7	0.94 ± 0.66	1.1 ± 0.8	0.07	1.12 ± 0.85	0.65 ± 0.78	0.82 ± 0.81	0.19
5. Headache, Dizziness	0.75 ± 0.97	0.25 ± 0.45	0.5 ± 1.0	0.56	1.17 ± 1.0	0.65 ± 0.78	0.83 ± 0.7	0.28	0.65 ± 0.6	0.18 ± 0.39	0.59 ± 0.71	0.84
6. Night Sweats	0.33 ± 0.49	0.17 ± 0.39	0.33 ± 0.5	1	0.78 ± 1	0.53 ± 0.8	0.5 ± 0.85	0.2	0.53 ± 0.71	0.18 ± 0.73	0.41 ± 0.8	0.62
7. Depressed Moods or a Sense of Loss	0.33 ± 0.65	0.17 ± 3.4	0.33 ± 0.5	0.375	1 ± 0.97	1.18 ± 2.56	0.55 ± 0.28	0.049*	0.94 ± 1.2	0.59 ± 1.0	0.53 ± 0.87	0.03*
8. Vaginitis	0.33 ± 0.65	0.25 ± 0.45	0.58 ± 0.8	0.44	0.44 ± 0.7	0.29 ± 0.59	0.33 ± 0.59	0.58	0.35 ± 0.6	0.12 ± 0.33	0.23 ± 0.56	0.5
9. Tends to Get Urinary Tract Infection	0.42 ± 0.79	0.17 ± 0.58	0.33 ± 0.79	0.75	0.41 ± 1	0 ± 0	0.18 ± 0.39	0.5	0.25 ± 0.58	0 ± 0	0.06 ± 0.25	0.25
10. Pain during Sexual Intercourse	0.83 ± 1.27	0.5 ± 1.17	0.91 ± 1.38	0.75	0.7 ± 0.92	0.43 ± 0.62	0.64 ± 0.86	0.87	0.5 ± 0.8	0.5 ± 1.16	0.44 ± 1.1	0.63
11. Urinary Incontinence	0.75 ± 0.62	0.58 ± 0.51	0.67 ± 0.65	0.75	0.47 ± 0.8	0.19 ± 0.4	0.24 ± 0.43	0.5	0.37 ± 0.5	0.38 ± 0.5	0.25 ± 0.45	0.38
12. Allergies	0.5 ± 0.8	0.17 ± 0.39	0.25 ± 0.45	0.046*	0.64 ± 0.7	0.5 ± 0.63	0.23 ± 0.44	0.03*	0.81 ± 1.05	0.38 ± 0.8	0.38 ± 0.5	0.16
13. Dry Mouth	0.18 ± 0.75	1.27 ± 1.1	0.9 ± 0.7	0.25	1.06 ± 0.82	0.63 ± 0.72	0.88 ± 0.93	0.47	1 ± 0.97	0.75 ± 1	0.87 ± 1	0.73
14. Hectic Sweats	0.45 ± 0.52	0.46 ± 0.69	0.54 ± 0.69	1	1.18 ± 1.3	0.88 ± 0.9	0.56 ± 0.9	0.02*	1.25 ± 1.29	0.69 ± 1.25	0.88 ± 1.26	0.19
15. Constipation	0.82 ± 0.9	0.56 ± 1.0	0.73 ± 0.9	0.8	0.71 ± 1.2	0.38 ± 1.02	0.4 ± 1	0.13	0.56 ± 0.72	0.38 ± 0.8	0.5 ± 0.9	0.75
16. Diarrhea	0.17 ± 0.4	0 ± 0	0.1 ± 0.3	1	0.35 ± 0.49	0.16 ± 0.52	0.12 ± 0.33	0.16	0.13 ± 0.34	0.06 ± 0.25	0 ± 0	0.5
17. Chest Tightness	0.82 ± 0.75	0.18 ± 0.4	0.34 ± 0.9	0.047*	0.82 ± 1.0	0.38 ± 1.0	0.29 ± 0.59	0.04*	0.73 ± 0.96	0.43 ± 0.9	0.43 ± 0.9	0.06
18. Elevated Blood Pressure	0.45 ± 0.69	0.1 ± 0.3	0.18 ± 0.4	0.5	0.35 ± 0.61	0.44 ± 0.81	0.24 ± 0.44	0.62	0.47 ± 0.74	0.46 ± 0.25	0.4 ± 0.5	0.81
19. Decreased Blood Pressure	0.18 ± 0.4	0 ± 0	0 ± 0	0.5	0.18 ± 0.39	0 ± 0	0 ± 0	0.25	0.2 ± 0.41	1 ± 0	0.07 ± 0.26	0.5

Table 6. Statistics of 25 common physical and mental symptoms around menopause. For each group, one-way ANOVA was used to test the significance of difference between the three different times (before treatment, during treatment, and post treatment) within the same group. The asterisk (*) shows the significance with $P < 0.05$. (Continued)

Group	High-dose (200mg)				Low-dose (100mg)				Placebo			
	Pre-treatment	1-month Post-treatment	2-month Post-treatment	*P*-value	Pre-treatment	1-month Post-Treatment	2-month Post-treatment	*P*-value	Pre-treatment	1-month Post-treatment	2-month Post-treatment	*P*-value
20. Facial Flushing	0.73 ± 0.78	0.27 ± 0.65	0.44 ± 0.5	0.04*	0.59 ± 0.93	0.31 ± 0.48	0.41 ± 0.62	0.44	0.73 ± 1.1	0.53 ± 1.1	0.53 ± 1.2	0.5
21. Pale Appearance	0.31 ± 0.48	0 ± 0	0.15 ± 0.38	0.5	0.18 ± 0.39	0.13 ± 0.34	0.19 ± 0.33	0.75	0.13 ± 0.52	0.27 ± 0.71	0.33 ± 0.72	0.5
22. Joint And Bone Pain	1.54 ± 1.2	0.69 ± 0.85	0.53 ± 0.87	0.03*	1.47 ± 1.2	1.12 ± 1.16	1 ± 1	0.048*	1.2 ± 1.26	0.73 ± 1.28	0.92 ± 1.1	0.07
23. Being Depressed And Suspicious	0.84 ± 0.9	0.76 ± 0.3	0.38 ± 0.5	0.047*	0.65 ± 0.86	0.31 ± 0.7	0.47 ± 0.62	0.58	0.73 ± 1.1	0.27 ± 0.49	0.47 ± 0.9	0.19
24. General Weakness	0.85 ± 1.2	0.23 ± 0.44	0.46 ± 0.5	0.046*	0.88 ± 0.99	0.37 ± 0.62	0.35 ± 0.6	0.043*	0.8 ± 1.2	0.33 ± 0.81	0.47 ± 0.92	0.19
25. Irritable	0.08 ± 1.51	0.46 ± 0.78	0.69 ± 0.63	0.31	0.7 ± 0.98	0.37 ± 0.8	0.59 ± 0.62	0.8	0.93 ± 0.96	0.34 ± 0.49	0.53 ± 0.83	0.09

degenerative bone and joint symptoms.

Moreover, among the seven significantly improved symptoms in the low-dose group (Figures 2 and 3), reduction of most of the symptoms after 2 months of treatment was more obvious than after 1 month of treatment except the depressed moods and a sense of loss, which may have been because of the placebo effect. In the high-dose group, significantly improved symptoms were not in proportion to treatment time. All of the symptoms rebounded in the second month, except for joint and bone pain. These results indicated that low-dose GEJ could alleviate the perimenopausal symptoms more effectively.

Discussion

Although the p-value in the test of serum estradiol difference is 0.048 (which is close to 0.05), the average serum estradiol level is almost twice the initial serum level. Therefore, the value is still statistically significant.

This result reveals that GEJ can elevate serum estradiol levels as well as relieve perimenopausal symptoms. In the improvements of the perimenopausal symptoms, the most crucial are facial flushing and hectic sweats. Both of these symptoms are related to those hot flushes and may be relieved by performing HRT, implying that GEJ may have the same mechanism as estradiol[4]. Based on previous studies, the *Panax ginseng C.A. Meyer* in GEJ is related to binding or activating the estrogen receptor[42-46]. Among all ingredients in the *Panax ginseng C.A. Meyer*, the most frequent referred ingredient is ginsenoside Rb1[45,46]. This may be why the effects of GEJ resemble those of estrogen.

The results of this study indicate significant

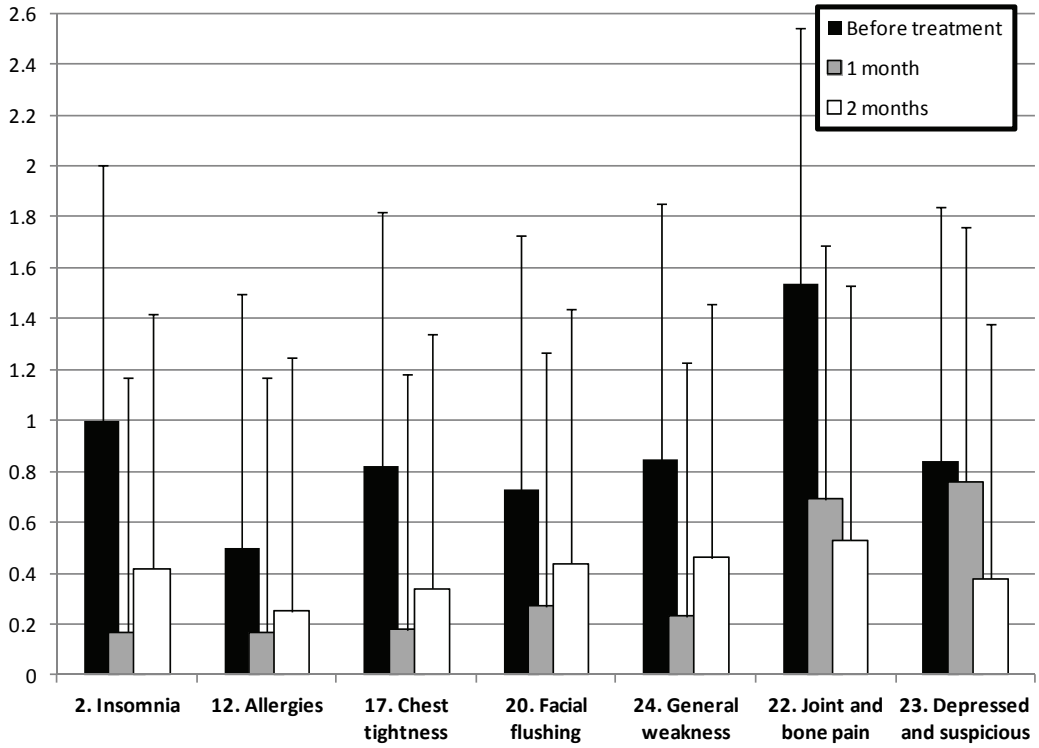

Fig. 2. Significant improvement of symptoms in the high-dose group (200 mg/day).

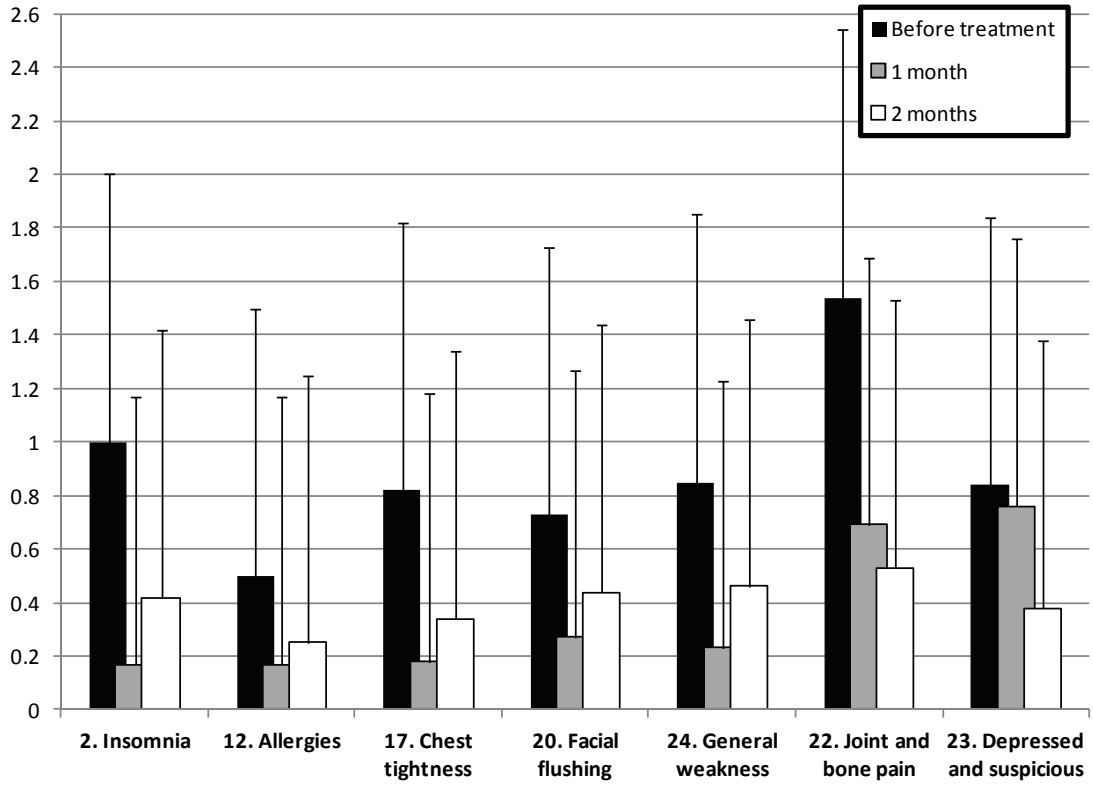

Fig. 3. Significant improvement of symptoms in the low-dose group (100 mg/day).

promotion of serum estradiol levels in the high-dose GEJ group, as compared to the low-dose GEJ group; however, the low-dose GEJ group experienced more effective relief of perimenopausal symptoms such as chest tightness, palpitation, hectic fever and sweats, allergies, and general weakness. This may be because of other components in GEJ such as *Cornu Cervi, Carapax et Plastrum Chrysemys*, and *Lycii Fructus* playing vital roles in reducing the perimenopausal symptoms. Because the clinical symptoms of menopause were not all directly related to hormone changes, the autonomic nervous system also contributes to menopause symptoms, such as palpitation, hectic fever and sweats, and insomnia. These symptoms may be reduced by other components in GEJ besides ginseng. Therefore, low-dose GEJ treatment can reduce some clinical symptoms more effectively than high-dose GEJ treatment can.

In this study, 33 women still had menstruation, and 28 women were diagnosed with amenorrhea. In both the high-dose group and low-dose group, 11 women still had menstruation, and 9 women had amenorrhea. Hence, in this study, there were more women who still had menstruation than those with amenorrhea in each group (high-dose, low-dose, and placebo). So the serum E2 elevation post 2 month-treatment may not due to the surge of menopause but is rather due to the high dose GEJ taken.

As the results indicated, in the high-dose group, treatment of GEJ could significantly reduce facial flushing but not in the low-dose and placebo groups. One probability is that the effect of *Carapax et Plastrum Chrysemys* in nourishing yin and suppressing hyperactive yang was reflected in the high-dose group more easily. The other reasons may be due to the dose dependent estradiol effect of GEJ[4]. Of the significantly improved symptoms, insomnia was noted in the high-dose group, whereas palpitation was noted in the low-dose group. Further study may be required to determine the dosage dependent effects on sympathetic and parasympathetic tone.

Regarding symptom allergies, both experimental groups attained significant improvement. The results would work in concert with a previous study in which CD4+ T lymphocyte counts reduced significantly ($P < 0.05$) after receiving GEJ for 2 months at 300 mg/day and 450 mg/day[47].

Although no significant reduction of the symptom of dry mouth occurred in either GEJ group ($P > 0.05$), the reduction in the low-dose group was in proportion to time increases. Instead, increases were observed in the high-dose group, possibly implying that the high-dose group may contribute to dry mouth and also explaining the hot property of the elevating yang energy of GEJ. Further study is required to determine whether the high percentage of the elevating yang energy ingredient, *Cornu Cervi*, caused the hectic sweats and other hot property, for example, thirst, rather than that in the low-dose group.

Regarding the results of significant improvement of general weakness and chest tightness attained in both GEJ groups ($P < 0.05$), the reason is that GEJ is clinically considered to nourish the kidney and replenish the essence, and is suitable for treating aging, weak health, severe sickness, and patients recovering from a major surgery. Therefore, either high or low GEJ dosage may treat general weakness by recovering body energy and possibly improving chest tightness by strengthening the contractility of respiratory muscle.

Significant improvement of depressed moods or a sense of loss in the placebo group and depressed moods in both GEJ groups (depressed and suspicious

in the high-dose group, depressed moods and or a sense of loss in the low-dose group) may be considered psychological effects, which require further study.

Conclusion

Based on the results of this study, perimenopausal syndromes can be relieved by the treatment of either low-dosage (100 mg/day) or high-dosage (200 mg/day) GEJ. High-dose GEJ treatment helps raise serum estradiol concentrations, but low-dose treatment can reduce perimenopausal symptoms more effectively. We conclude that GEJ treatment benefits women diagnosed with perimenopausal syndrome.

Acknowledgments

This research was supported by National Research Institute of Chinese Medicine (plan number: NRICM-9603). We greatly appreciate all the assistance from the staff.

References

1. Soules MR, Sherman S, Parrott E, Rebar R, Santoro N, Utian W, Woods N. Executive summary: stages of reproductive aging workshop (STRAW). *Fertil. Steril.*, 76:874-878, 2001.
2. Berg G, Gottqall T, Hammar M, Lindgren R. Climacteric symptoms among women aged 60–62 in Linköping, Sweden, in 1986. *Maturitas*, 10:193-199, 1988.
3. Greene JG. Constructing a standard climacteric scale. *Maturitas*, 29:25-31, 1998.
4. Stearns V, Ullmer L, Lopez FJ, Smith Y, Isaacs C, Hayes FD. Hot flushes. *Lancet*, 360:1851-1861, 2002.
5. Heinemann LAJ, Potthoff P, Schneider HPG. International versions of the menopause rating scale (MRS). *Health Qual. Life Outcomes*, 1:28, 2003.
6. Greene JG. A factor analytic study of climacteric symptoms. *J. Psychosom. Res.,* 20:425-430, 1976.
7. Hunter M, Battersby R, Whitehead M. Relationships between psychological symptoms, somatic complaints and menopausal status. *Maturitas*, 8:217-228, 1986.
8. Holte A, Mikkelsen A. The menopausal syndrome: a factor analytic replication. *Maturitas*, 13:193-203, 1991.
9. Voda AM. Climacteric hot flash. *Maturitas*, 3:73-90, 1981.
10. Bachmann GA. Vasomotor flushes in menopausal women. *Am. J. Obstet. Gynecol.*, 180:S312-S316, 1999.
11. Chim H, Tan BHI, Ang CC, Chew EMD, Chong YS, Saw SM. The prevalence of menopausal symptoms in a community in Singapore. *Maturitas*, 41:275-282, 2002.
12. Haines CJ, Yim SF, Chung TKH, Lam CWK, Lau EWC, Ng MHL, Chin R, Lee DTS. A prospective, randomized, placebo-controlled study of the dose effect of oral oestradiol on menopausal symptoms, psychological well being, and quality of life in postmenopausal Chinese women. *Maturitas*, 44:207-214, 2003.
13. Pan HA, Wu MH, Hsu CC, Yao BL, Huang KE. The perception of menopause among women in Taiwan. *Maturitas*, 41:269-274, 2002.
14. Haines CJ, Xing SM, Park KH, Holinka CF, Ausmanas MK. Prevalence of menopausal symptoms in different ethnic groups of Asian women

and responsiveness to therapy with three doses of conjugated estrogens/medroxyprogesterone acetate: the Pan-Asia menopause (PAM) study. *Maturitas*, 52:264-276, 2005.

15. Iosif CS, Bekassy Z. Prevalence of genito-urinary symptoms in the late menopause. *Acta. Obstet. Gynecol. Scand.*, 63:257-260, 1984.
16. Colditz GA, Willett WC, Stampfer MJ, Rosner B, Speizer FE, Hennekens CH. Menopause and the risk of coronary heart disease in women. *N. Engl. J. Med.*, 316:1105-1110, 1987.
17. Rossouw JE, Anderson GL, Prentice RL, LaCroix AZ, Kooperberg C, Stefanick ML, Jackson RD, Beresford SAA, Howard BV, Johnson KC, Kotchen JM, Ockene J. Risks and benefits of estrogen plus progestin in healthy postmenopausal women: principal results from the Women's health initiative randomized controlled trial. *J. Am. Med. Assoc.*, 288:321-333, 2002.
18. Lin YH, Chang HJ, Chen TL. Prevalence of Climacteric Symptoms among Taiwanese Woman and the Health Related Behavior in Taiwan: Result of the 2002 National Health Promotion Knowledge, Attitude, and Behavior Survey. *2004 Annu. Meet.Popul. Assoc. Taiwan Popul. Fam. Natl. Health Policy Rev. Prospect.*, 2004. (in Chinese)
19. Chang C, Chow SN, Hu Y. Age of menopause of Chinese women in Taiwan. *Int. J. Gynaecol. Obstet.,* 49:191-192, 1995.
20. Chow SN, Huang CC, Lee YT. Demographic characteristics and medical aspects of menopausal women in Taiwan. *J. Formos. Med. Assoc. (Taiwan Yi Zhi)*, 96:806-811, 1997. (in Chinese)
21. O'Connell D, Robertson J, Henry D, Gillespie W. A systematic review of the skeletal effects of estrogen therapy in postmenopausal women. II. An assessment of treatment effects. *Climacteric*, 1:112-123, 1998.
22. Bjarnason NH, Hassager C, Christiansen C. Postmenopausal bone remodelling and hormone replacement. *Climacteric*, 1:72-79, 1998.
23. Guo HL, Li XY, Gao XW. Male semen abnormality treated by Guilu Erxian Jiao. *Shaanxi J. Tradit. Chin. Med.*, 26:213-214, 2005. (in Chinese)
24. Lu SL, Kao HL. The 103 examples of adolescent menorrhagia treated with Modified Guilu Erxian Jiao. *Shaanxi J. Tradit. Chin. Med.*, 19:247, 1998. (in Chinese)
25. Lu HT. A survey of meaning and application of Guilu Erxian Jiao. *J. Hubei Coll. Tradit. Chin. Med.*, 1:77, 1999. (in Chinese)
26. Cho S, Won CH, Lee DH, Lee MJ, Lee S, So SH., Lee SK, Koo BS, Kim NM, Chung JH. Red ginseng root extract mixed with Torilus Fructus and Corni Fructus improves facial wrinkles and increases type I procollagen synthesis in human skin: a randomized, double-blind, placebo-controlled study. *J. Med. Food*, 12:1252-1259, 2009.
27. Huang HC, Ma CC. Effect of Gui Ban Dihuang Decoction to the amount of proteinurea of chronic nephritis. *Pract. J. Integr. Chin. Mod. Med.*, 10:476, 1997. (in Chinese)
28. Yao CH, Liu BS, Liu CG, Chen YS. Osteogenic potential using a malleable, biodegradable composite added traditional Chinese medicine: in vitro and in vivo evaluations. *Am. J. Chin. Med.*, 34:873-886, 2006.
29. Cheng PT, Lo TW. Pharmacological studies on Guilu Erxian Jiao. *Chin. Tradit. Pat. Med.*, 22:860-861, 2000. (in Chinese)
30. Lee J, Jung E, Lee J, Huh S, Kim J, Park M, So J, Ham Y, Jung K, Hyun CG, Kim YS, Park D.

Panax ginseng induces human Type I collagen synthesis through activation of Smad signaling. *J. Ethnopharmacol.*, 109:29-34, 2007.

31. Metori K, Furutsu M, Takahashi S. The preventive effect of ginseng with du-zhong leaf on protein metabolism in aging. *Biol. Pharm. Bull.*, 20:237-242, 1997.
32. Zhao J, Wang D, Duan S, Wang J, Bai J, Li W. Analysis of fuzhisan and quantitation of baicalin and ginsenoside Rb_1 by HPLC-DAD-ELSD. *Arch. Pharmacal. Res.*, 32:989-996, 2009.
33. Jang HI, Shin HM. Wild *Panax ginseng* (*Panax ginseng* C.A. Meyer) protects against methotrexate-induced cell regression by enhancing the immune response in RAW 264.7 macrophages. *Am. J. Chin. Med.*, 38:949-960, 2010.
34. Son CG, Han SH, Cho JH, Shin JW, Cho CH, Lee YW, Cho CK. Induction of hemopoiesis by saenghyuldan, a mixture of Ginseng Radix, Paeoniae Radix Alba, and Hominis Placenta extracts. *Acta. Pharmacol. Sin.,* 24:120-126, 2003.
35. Chui CH, Wong RSM, Cheng GYM, Lau FY, Kok SHL, Cheng CH, Cheung F, Tang WK, Teo ITN, Chan ASC, Tang JCO. Antiproliferative ability of a combination regimen of crocodile egg extract, wild radix ginseng and natural *Ganoderma lucidum* on acute myelogenous leukemia. *Oncol. Rep.*, 16:1313-1316, 2006.
36. Xiao PG, Xing ST, Wang LW. Immunological aspects of Chinese medicinal plants as antiageing drugs. *J. Ethnopharmacol.*, 38:159-165, 1993.
37. Deng R, Li Y, Chen D, Li H, Zhang S, Zhao D, Meng L, Zhou J, Meng D, Zhang R, Hailing Z. Protective effect of Plastrum Testudinis on the apoptosis of dopamine neurons of rats with Parkinson's disease. *Chin. J. Neuroanat.*, 24:301-306, 2008. (in Chinese)
38. Hou QK, Wu J, Yi XH, Chen DF, Zhou JH, Li YW. Induction of Plastrum Testudinis extract to mesenchymal stem cells differentiation into osteoblast through up-regulating expression of vitamin D receptor. *Chin. Tradit. Herb. Drugs*, 41:607-612, 2010. (in Chinese)
39. Hijikata Y, Kano T, Xi L. Treatment for intractable anemia with the traditional Chinese medicines Hominis Placenta and Cervi Cornus Colla (deer antler glue). *Int. J. Gen. Med.*, 2:83-90, 2009.
40. Yan, DY, Duan XZ. Clinical Study on menopausal women's unstable angina pectoris treated by supplemented Guilu Erxian Jiao. *J. Anhui Tradit. Chin. Med. Coll.*, 19:20-22, 2000. (in Chinese)
41. Hilditch JR, Lewis J, Peter A, van Maris B, Ross A, Franssen E, Guyatt GH, Norton PG, Dunn E. A menopause-specific quality of life questionnaire: development and psychometric properties. *Maturitas*, 24:161-175, 1996.
42. Pearce PT, Zois I, Wynne KN, Funder JW. *Panax ginseng* and *Eleuthrococcus senticosus* extracts--in vitro studies on binding to steroid receptors. *Endocrinol. Jpn.*, 29:567-573, 1982.
43. Attele AS, Wu JA, Yuan CS. Ginseng pharmacology: multiple constituents and multiple actions. *Biochem. Pharmacol.*, 58:1685-1693, 1999.
44. Furukawa T, Bai CX, Kaihara A, Ozaki E, Kawano T, Nakaya Y, Awais M, Sato M, Umezawa Y, Kurokawa J. Ginsenoside Re, a main phytosterol of *Panax ginseng*, activates cardiac potassium channels via a nongenomic pathway of sex hormones. *Mol. Pharmacol.*, 70:1916-24, 2006.
45. Lee Y, Jin Y, Lim W, Ji S, Choi S, Jang S, Lee S. A ginsenoside-Rh1, a component of ginseng saponin, activates estrogen receptor in human

breast carcinoma MCF-7 cells. *J. Steroid Biochem. Mol. Biol.*, 84:463-468, 2003.

46. Hao K, Gong P, Sun SQ, Hao HP, Wang GJ, Dai Y, Liang Y, Xie L, Li FY. Beneficial estrogen-like effects of ginsenoside Rb1, an active component of *Panax ginseng*, on neural 5-HT disposition and behavioral tasks in ovariectomized mice. *Eur. J. Pharmacol.,* 658:15-25, 2011.
47. Lin YH, Chen HY, Li YC, Chiu JC, Yang SH. Adverse events assessment of traditional Chinese herbal product, Guilu Erxian Jiao, in healthy volunteers. *J. Chin. Med.*, 22:65-75, 2011.

龜鹿二仙膠治療更年期綜合症之臨床評估

王品涵[1]、李怡秋[1]、吳奕慧[1]、陳俊良[1,2]、邱健泰[3,4]、楊賢鴻[1,2,*]

[1]桃園長庚紀念醫院中醫部，桃園，台灣

[2]長庚大學中醫學系，桃園，台灣

[3]長庚紀念醫院婦產部，桃園，台灣

[4]長庚大學生物醫學系，桃園，台灣

（101 年 02 月 29 日受理，101 年 07 月 30 日接受刊載）

在台灣，有46%絕經前後女性經歷過更年期綜合症。常見的症狀有失眠、熱潮紅、潮熱盜汗及心悸。目前治療的主流為賀爾蒙替代療法。但許多患有更年期綜合症的婦女會自行選用龜鹿二仙膠而不願使用賀爾蒙替代療法。因此，本研究的主要目標在於研究龜鹿二仙膠是否能緩解更年期綜合症。本研究將患有更年期綜合症的婦女分為三組，分別為每天服用龜鹿二仙膠200毫克、100毫克、及安慰劑（澱粉）之三組。各組每日均服用一顆膠囊一次，內含上述三組成分，共連續服用兩個月。分別在治療前、治療中（治療後一個月）、及治療後（治療後兩個月）測量三組婦女血中雌激素、促濾泡成熟素濃度。臨床症狀部分，則以問卷方式分別在治療前及治療後（治療後兩個月）評估婦女生理及心理等不同面向的更年期症狀。結果顯示，經歷兩個月的治療，血中雌激素濃度在高劑量組（每日服用200毫克龜鹿二仙膠）有顯著上升（$p < 0.05$）。但是在血中促濾泡成熟素濃度方面，三組均無顯著差異。臨床症狀方面，雖然高劑量組及低劑量組分別比安慰組在許多面向均有改善（$p < 0.05$），但整體看來，低劑量組（每日服用100毫克龜鹿二仙膠）的治療效益較高，尤其表現在改善潮熱盜汗及心悸上。因此，我們的結論為龜鹿二仙膠對於治療更年期綜合症是有效的。

關鍵字：中醫藥、更年期、龜鹿二仙膠、雌激素

*聯絡人：楊賢鴻，桃園長庚紀念醫院中醫部，33378桃園縣龜山鄉頂湖路123號，電話：03-3196200分機2611，傳真：03-3298995，電子郵件信箱：dryang@adm.cgmh.org.tw