

## Auricular Acupressure May Improve Absorption of Flavanones in the Extracts from *Citrus aurantium* L. in the Human Body

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### ABSTRACT

**Objective:** To explore whether auricular acupressure could improve the absorption of flavanones in the extracts from *Citrus aurantium* L. in the human body.

**Methods:** Ten (10) healthy male volunteers were randomly divided into two groups. Group A was treated with the combination of oral administration of *C. aurantium* L. extracts and auricular acupressure. Group B was treated only with the oral administration of *C. aurantium* L. extracts in the same dosage as Group A. After 7 days of treatment, the plasma concentration of naringenin and hesperetin of the two groups were determined using high-performance liquid chromatography.

**Results:** The plasma concentration of naringenin and hesperetin in Group A were significantly higher than Group B.

**Conclusions:** Auricular acupressure at some specific acupoints can significantly improve the absorption of naringenin and hesperetin in the extracts from *C. aurantium* L. in the human body.

### INTRODUCTION

Acupuncture and Chinese medicinal herbs were sometimes used together, which was called “combined acupuncture and herbs therapy.” In most cases, acupuncture and herbs were respectively applied to treat various symptoms or syndromes. However, no research has been conducted on the effect of acupuncture on the absorption of Chinese medicinal herbs in the human body.

A special emphasis has been laid on the pharmacological properties and medicinal uses of the compounds in *Citrus aurantium* L., commonly known as sour orange, in recent years.<sup>1</sup> The present study, for the first time, aimed to explore whether auricular acupressure could

improve the absorption of plasma naringenin and hesperetin in the extracts from *C. aurantium* L. in the human body.

### MATERIALS AND METHODS

#### *Preparation of the extracts of Citrus aurantium L.*

*C. aurantium* L. extract was prepared by boiling 100 g of the dried fruit *C. aurantium* L. in 1 L of distilled water for 1 hour. The operations were repeated twice. The total extracts were condensed to obtain the extract in the form of a standard concentration of 1g/mL.

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## Subjects

Ten (10) healthy male volunteers were randomly divided into Group A with 5 cases and Group B with 5 cases.

## Group and administration

**Group A.** The subjects in group A were orally administered 200 mL of the extracts from *C. aurantium* L., once a day for 7 consecutive days and the auricular acupressure was conducted simultaneously for 7 consecutive days.

Sympathetic (AH6a); Shenmen (TF4); Adrenal gland (TG2p); Subcortex (AT4); Endocrine (CO18); Kidney (CO10); Heart (CO15); and Liver (CO12) were selected as the auricular acupoints.

**Group B.** The subjects in group B were orally administered 200 mL of the extracts from *C. aurantium* L., once a day for 7 consecutive days.

Baseline blood samples were obtained at the beginning of the study with a 12-hour fast before the first administration of *C. aurantium* L. extracts. The 10 blood samples were collected 6 hours after the last administration.

Each participant received a physical examination, routine blood examination, routine urinoscopy, liver function test, and renal function test, respectively, 2 days before the treatment and on the second day after the treatment ended.

## SAMPLE COLLECTION AND ANALYSIS

The blood samples were collected into vacuum tubes containing EDTA 6 hours after the last administration of *C. aurantium* L. extract. The plasma concentrations of naringenin and hesperetin were determined using a reversed-phase high-performance liquid chromatography (HPLC) procedure. The samples were enzymatically hydrolyzed. The supernatant was dissolved in 100  $\mu$ L methanol that was subjected to HPLC (Shimadzu LC2010A system, Japan). A diamonsil C18 column (250 mm  $\times$  4.6 mm, 5  $\mu$ m) from Dikma Technologies (Beijing, China) was equipped with pre-column (4 mm  $\times$  5 mm) C18. A linear gradient elution of A (0.5% acetic acid solution) and B (acetonitrile) was applied. The standards were treated the same way as the samples. The plot of peak area (*Y*) against the concentration (*X*,  $\mu$ g/L) for each of the compounds was evaluated by linear regression analysis.

## Statistical analysis

Analysis of variance was employed for comparison of the plasma naringenin and hesperetin concentration of the subjects from both groups. For all hypothesis tests, a 5% significance level ( $p < 0.05$ ) and two-tailed tests were used.

## RESULTS

### Analysis of the samples

The body weight and the concentrations of plasma naringenin and hesperetin in the two groups were shown as Table 1. The peak plasma concentrations of naringenin and hesperetin in Group A were respectively  $3.91 \pm 1.01$   $\mu$ g/L and  $20.96 \pm 3.07$   $\mu$ g/L. The peak plasma concentrations of naringenin and hesperetin in Group B were respectively  $2.40 \pm 0.44$   $\mu$ g/L and  $13.74 \pm 4.97$   $\mu$ g/L. The plasma concentrations can reflect the absorption of flavanones in *C. aurantium* L. in healthy volunteers. No significant difference in body weight existed between the two groups. The concentrations of plasma naringenin and hesperetin in Group A were significantly higher than Group B, indicating that auricular acupressure can improve the absorption of *C. aurantium* L. extracts in the human body.

### Adverse result

No side effects were reported in both groups.

## DISCUSSION AND CONCLUSION

The earliest record of diagnosis and treatment with auricular acupoints is in *Huangdi Neijing* (the *Yellow Emperor's Classic of Internal Medicine*).<sup>2</sup> The famous Chinese medical classics summarized a great number of acupuncture treatments, among which are the specific acupoints on the external ear for the relief of certain disorders.<sup>2</sup> Rapid progress has been made on the theory and practice of auricular acupressure since Paul Nogier, a French practitioner, discovered the inverted-fetus shape distribution pattern of auricular acupoints.<sup>3</sup> Auricular acupressure can associate the meridians of the body, regulate *Qi* and activate blood, regulate *Zang-fu* (organs), and promote the metabolism.<sup>4</sup> Each acupoint on the ear, when treated, triggers electrical impulses, from the ear, via the brain, to the specific area of the body.<sup>4,5</sup> Auricular acupressure is safe, effective, economical, and simple and it works by stimulating the central nervous system through the cranial nerves/spinal nerves on the auricle of the ear.<sup>5</sup> A meta-analysis found that auricular

TABLE 1. COMPARISON OF BODY WEIGHT, PLASMA NARINGENIN, AND HESPERETIN BETWEEN GROUP A AND GROUP B

Group	n	Weight (kg)	Plasma ( $\mu$ g/L)	
A	5	$71.20 \pm 9.04$	Naringenin	$3.91 \pm 1.01^{*(0.016)}$
			Hesperetin	$20.96 \pm 3.07^{*(0.020)}$
B	5	$70.61 \pm 9.29$	Naringenin	$2.40 \pm 0.44$
			Hesperetin	$13.74 \pm 4.97$

\* $p < 0.05$ , compared with group B.

acupuncture produced better rates of recovery and improvement than diazepam for treating insomnia.<sup>6</sup>

The result of the study demonstrated that auricular acupressure at the specific acupoints can significantly improve the absorption of naringenin and hesperetin in the extracts from *C. aurantium* L. in the human body. The result of the present study is consistent with two previous researches on rats.<sup>7,8</sup>

It may be possible to find out auricular acupoints specific to each Chinese medicinal herb, which would allow us to administer the minimum dose of the herb to treat a disease when combining it with auricular acupressure at these specific acupoints. For this hypothesis to be examined, much further research with large samples should be done. If successful, a database would be established that would include each Chinese medicinal herb and its corresponding auricular acupoints that function to improve absorption of the active compounds in the herb. Patients could then benefit greatly from the database.

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