Egg White, a Favourable Solvent to Extract the Active Ingredient of Mylabris phalerata

Bing-Ji Ma* and Yuan Ruan

Department of Traditional Chinese Medicine, Agronomy College of Henan Agricultural University, Zhengzhou 450002, P. R. China. E-mail: mbj123@sina.com

- * Author for correspondence and reprint requests
- Z. Naturforsch. 63c, 619-620 (2008); received October 15, 2007/February 25, 2008

To characterize the role of hen eggs used for analyzing the insect *Mylabris phalerata* a traditional Chinese medicine for curing liver cancer, egg white was separated from the whole egg and added to *M. phalerata*. The volatile compounds found in egg white when added to *M. phalerata* and therefore in *M. phalerata* itself were analyzed by GC. Cantharidin was detected both in egg white added to *M. phalerata* and *M. phalerata* itself. Egg white can be used as a favourable solvent for extracting the active ingredient of *M. phalerata*.

Key words: Egg White, Mylabris phalerata, Cantharidin

Introduction

Mylabris phalerata is an insect belonging to the family Meloidae. It has been used for the treatment of liver cancer in traditional Chinese medicine for more than 1000 years (Jiangsu New College of Medicine, 2006), and it is still used as a folk medicine. In recent studies, M. phalerata has been found to possess antitumour properties, to increase the number of leucocytes and to have irritant effects on the urinary organs. Cantharidin was isolated from M. phalerata and revealed to be the active ingredient (Wang, 1989; McCluskey et al., 2002). Cantharidin is a potent inhibitor of hepatocellular carcinoma cells (Hep 3B cells) and its IC₅₀ values on Hep 3B cells and normal Chang liver cells were found to be 2.2 and 30.2 μ M for 36 h, respectively (Efferth, 2005).

In Chinese folk medicine, the use of *M. phalerata* against liver cancer is conducted as follows: Firstly, *M. phalerata* is caught and its head, legs and wings are removed; then the body is dried for several days. Secondly, a small hole is knocked out of the eggshell, big enough for putting in a dried insect body of *M. phalerata*; then the hole is covered with clay. Finally, the egg is cooked and the patients eat the egg white.

To characterize the role of hen eggs used for the insect *Mylabris phalerata* to cure liver cancer in traditional Chinese medicine, egg white was separated from the whole egg and added to *M. phalerata*. The volatile compounds of egg white added to

M. phalerata and M. phalerata itself were analyzed by GC. Cantharidin was detected both in egg white added to M. phalerata and M. phalerata itself. Our experimental results indicate that egg white can be used as a favourable solvent for extracting the active ingredient of M. phalerata.

Results and Discussion

The main compound, namely cantharidin, was detected and identified both in egg white added to *M. phalerata* and *M. phalerata* itself by means of GC analysis.

Egg white is the common name for the clear liquid contained in eggs, and is a mixture of 85-88% water, 11–12% protein (mainly ovalbumin, ovotransferrin and lysozyme) and 1% lipids (mainly lecithin). In China, egg white has been used in traditional Chinese medicine for nearly 1000 years, and is still used today. The extraction solvent is either water or methanol/ethanol. Egg white can be seen as a two-phase solvent by the analysis of its composition; and it does not only easily dissolve the water-soluble ingredients but also easily dissolves the lipid-soluble ingredients. Our experimental results indicate that egg white can be used in solvent extraction especially in the pharmaceutical industry. It is used as a favourable solvent, with several merits; that are: it is easy to absorb by the patients, affluent in resource and not expensive.

Experimental

General

Sample analysis was conducted in a GC instrument (HP6890GC/5973MS, Agilent Technologies, Palo Alto, USA). The sample was injected into a capillary column ($30 \text{ m} \times 0.32 \text{ mm} \times 0.50 \,\mu\text{m}$ film thickness). After injection, the initial temperature ($46 \,^{\circ}\text{C}$) was increased at $30 \,^{\circ}\text{C/min}$ up to $175 \,^{\circ}\text{C}$ and held for $10 \,^{\circ}\text{min}$, thereafter at $10 \,^{\circ}\text{C/min}$ up to $210 \,^{\circ}\text{C}$ and held for $10 \,^{\circ}\text{min}$. Nitrogen was used as carrier gas, and the flow was $40 \,^{\circ}\text{ml/min}$.

For comparison, cantharidin was also analyzed by the same method as the standard.

Animal material

Fresh *M. phalerata* was collected at Zhengzhou city, Henan province, China, in September 2006. The animal identification was made by Prof. Zhi-

Ming Gao, Department of Traditional Chinese Medicine, Agronomy College of Henan Agricultural University, Zhengzhou, China. A voucher specimen (Ma 2006012) was deposited in the herbarium of Henan Agricultural University.

Analysis of volatiles of M. phalerata and egg white added to M. phalerata

A small hole was knocked out of the eggshell, thus the egg white (ca. 40 ml) flowed out of the hole was collected in a 150 ml flask. A dried insect body of *M. phalerata* was put into the flask for 24 h and then was removed from the egg white. Following extraction from the egg white with chloroform (20 ml) the samples were directly analyzed by GC. The main compound was identified as cantharidin. The same analytical method was used on *M. phalerata* itself. As a result, cantharidin was also detected at the same retention time in the gas chromatogram.

Efferth T. (2005), Microarray-based prediction of cytotoxicity of tumor cells to cantharidin. Oncol. Rep. 13, 459–463.

Jiangsu New College of Medicine (2006), The Compilation of Traditional Chinese Medicine. Shanghai Science and Technology Press, Shanghai, pp. 2280–2282. McCluskey A., Sim T. R., and Jakoff J. A. (2002), Serine-threonine protein phosphatase inhibitors: development of potential therapeutic strategies. J. Med. Chem. **45**, 1151–1175.

Wang G. S. (1989), Medical uses of *Mylabris* in ancient China and recent studies. J. Ethnopharmacol. **26**, 147–162.