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Identification of the Chinese Herbal Formulas by HPTLC – Mahuang & Apricot Seed Combination and Mahuang & Coix Combination

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Background

Traditional Chinese Herbal Medicine (TCM) combines several herbs in a prescription in order to enhance the desired action and to minimize side effects. Several formulas show only slight differences. For example, the two formulas Mahuang & Apricot Seed Combination (Ma Xing Shi Gan Tang) and Mahuang & Coix Combination (Ma Xing Yi Gan Tang) (Figures 1&2). To distinguish these similar formulas Thin-Layer Chromatography (TLC) is not always easy. In this report, a simple High-Performance Thin-Layer Chromatography (HPTLC) procedure with specific mobile solution for identification of the formulas is described.

Materials and Methods

Chromatography was performed on a HPTLC silica gel 60 F₂₅₄ plate as stationary phase while four different solutions

1. Dichloromethane: ethyl acetate: methanol: dest. water = 15:40:22:10 (V/V/V/V)
2. Petroleum ether: ethyl acetate: acetic acid = 10:3:0.1(V/V/V)
3. Toluene: ethyl formate: formic acid: dichloromethane = 10:12:6:12 (V/V/V/V)
4. N-butanol: acetic acid: dest. water = 7:1:2 (V/V/V)

were treated and compared as mobile phase. The chromatogram was then examined at UV 254 nm/UV 365 nm and in white light/UV 365 nm after derivatization with 10% of sulfuric acid in ethanol/molybdophosphoric acid reagent.

Results and Discussion

In this report, solution containing petroleum ether: ethyl acetate: acetic acid (10 : 3 : 0.1 V/V/V) with derivatization by 10% sulfuric acid in ethanol was shown to be the best mobile phase for identification of the two Formulas – Mahuang & Apricot Seed Combination and Mahuang & Coix Combination. Because Mahuang & Coix Combination are composed of Semen Coicis that can distinctly distinguish between Mahuang & Apricot Seed Combination and Mahuang & Coix Combination (Figure 3).

Taken together, due to original nature of herbal drugs, it is meaningful to develop an adequate method suitable for individual analysis distinguishing the existing variations among many similar formulas, such as extraction methods, mobile phases and derivatization systems. TLC method is simple, rapid, cheap, robust and applicable at a larger scale than any other analytical technique.



Figure 1. Mahuang & Apricot Seed Combination is composed of Herba Ephedrae (middle/up), Gypsum fibrosum (left), Semen Armeniacae (middle/down), and Radix Glycyrrizae (right). In TCM, the Formula act for releasing the exterior.

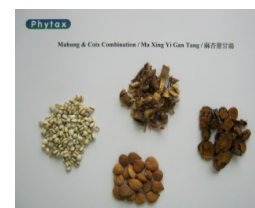


Figure 2. Mahuang & Coix Combination is composed of Herba Ephedrae (middle/up), Semen Armeniacae (middle/down), Semen Coicis (left), and Radix Glycyrrizae (right). In TCM, the formula act for clearing heat from organs.

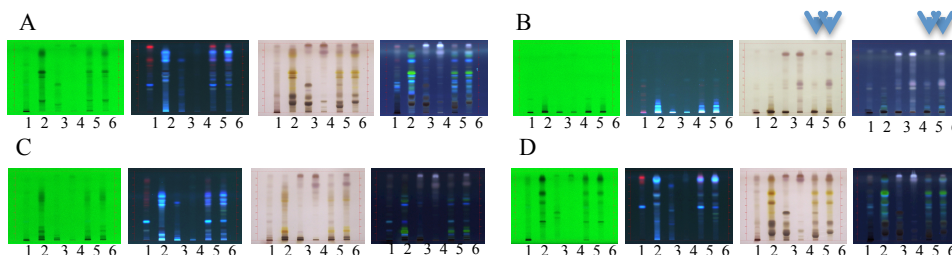


Figure 3. HPTLC fingerprints of comparison of Herba Ephedrae (Track 1), Radix Glycyrrizae (Track 2), Semen Armeniacae (Track 3), Semen Coicis (Track 4), Mahuang & Apricot Seed Combination (Track 5), and Mahuang & Coix Combination. Separation on HPTLC plate. Mobile phase: dichloromethane: ethyl acetate: methanol: dest. water = 15:40:22:10 (A), petroleum ether: ethyl acetate: acetic acid = 10:3:0.1 (B), toluene: ethyl formate: formic acid: dichloromethane = 10:12:6:12 (C), N-butanol: acetic acid: dest. water = 7:1:2 (D), derivatization with sulfuric acid/ethanol reagent. Detection in UV 254 nm, UV 365 nm, white light after derivatization, UV 365 nm after derivatization (left to right).

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