Authentication of *Aucklandia lappa* Decne. (Compositae) by Automated Multiple Development System 2 (AMD 2)

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Introduction

Common Aucklandia Root, the dried root of *Aucklandia lappa* Dence.(Compositae) included in Chinese Pharmacopoeia(Ch.P) 2005 Ed. The TLC identification of Common Aucklandia Root in Ch.P 2005 gave too poor quality of TLC image to differentiate this herbal drug from the adulterants such as Common ladimiriae root and Inulae root. To develop a new TLC method to increase the separation and enhance the specialty is ecessary for the purpose of authentication. Automatic multiple development (AMD) was chosen for optimization of experiment of this herbal drug, and a satisfactory result was available.

Appearance of Common Aucklandia Root

Appearance of Common Vladimiriae Root

Experimental

Add Chloroform to 2g of the powdered sample in a 50mL flask, ultrasonicate two times, each 30mL solvent and 15 min, filter, combine the filtrate, and evaporate to dryness. Dissolve the residue in 3mL absolute ethanol as the test solution. Dissolve Costunolide and Dehydro costuslactone reference substances separately in methanol to produce a solution containing 1mg per mL as the reference solution. Apply separately Test solution 4 μL and reference solution 2 μL on Silica gel 60 pre-coated HPTLC plate (Merck). Solvent system and developing distance were listed in table 1, Develop within AMD2 (CAMAG) at 19℃. After developing, dry the plate with hair-dryer, spray with 2% solution of ρ dimethylaminobenzaldethyde in 20% sulfuric acid and 20% phosphoric acid, heat at 105℃ until the color of the bands clear. Examine in daylight and under ultraviolet light at 365nm.

Table 1 Solvent system and developing distance

step	Toluene (%)	Chloroform (%)	Ethyl acetate (%)	Methanol (%)	Distance (mm)	Drying time (min)
1	20	50	20	10	15	10
2	64	20.4	14	1.6	28	10
3	71.2	17	10.6	1.2	41	10
4	78.4	13.6	7.1	0.9	55	10
5	85.6	10.2	3.7	0.5	70	10
6	92.8	6.8	0.3	0.1	80	10
7	98.6	1.4	0	0	90	10
8	98.6	1.4	0	0	90	10

TLC images of CH.P's mobile phase

S1-2：Costunolide(down) and Dehydro costuslactone（up）;lane1～6：Common Aucklandia Root; lane7～9：Common Vladimiriae root .Plate: TLC silica gel 60 (Merck); mobile phase :Chloroform-cyclohexane (5:1) Derivatization and Observation: spraying 2% solution of ρ-dimethylaminobenzaldethyde in 20% solution of sulfuric acid and 20% solution of phosphonic acid onto the developed and solvent-free plate, heat at 105℃ to the bands clear. Examine under ultraviolet light at 365nm.Documentation:prepare the TLC image photo with Digistore device .

TLC fluorescent images by means of AMD

S1:Costunolide; S2: Dehydro costuslactone; lane1～6: Common Aucklandia Root; lane7～9：Common Vladimiriae root

Discussion

AMD system is suitable in particular for the development of the non-polar constituents like the terpene lactones contained in Common Aucklandia root. Multiple steps development increase the resolution of the chromatogram drastically than conventional development in a twin trough chamber.

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