Analysis of chlorogenic acid in traditional Chinese medicines by capillary electrophoresis

Application Note

Pharmaceutical

Authors

Carsten Buhlmann,
Gordon Ross,
Agilent Technologies,
Waldbronn, Germany

Abstract

Chlorogenic acid (CA) (figure 1) is an ester of caffeic acid and quinic acid. All three of these substances naturally occur in many plants. CA which is present in the surface skin of peaches, inhibits the cutin-digesting enzyme of the brown rot fungus, *Monilinia fructicola*, demonstrating its antifungal activity. It has also shown antioxidant activity. CA is an active consituent of *Flos Lonicerae*. CZE has been used for its analysis in the plant and in some traditional medicines which contain this plant as a constituent[1]. Although HPLC has been used for the analysis of traditional Chinese medicines (TCM), the "dirty" matrix of the prepared sample can proove problematic for the LC column. Since CE operates using small bore unpacked tubing, it is much more tolerant of complex matrices and more robust when used for their analysis. Here we describe how CA can be identified and quantified in some Chinese traditional medicines using capillary electrophoresis.

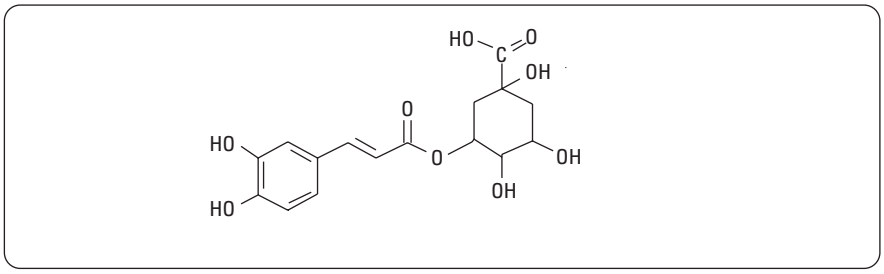


Figure 1
Structure of chlorogenic acid.

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Experimental

All analyses were performed using an Agilent CE system equipped with diode array detection and controlled via a PC running the Agilent ChemStation software. Traditional medicine samples and standards were the kind gift of Professor H. Liu, Peking University, Beijing, PR China. Other reagents were supplied by Sigma.

Extraction

The indicated amounts of pulverized samples were soaked with 7 mL 50 % ethanol/water overnight and extracted by strirring for 30 minutes. After centrifugation (4000 rpm, 10 minutes) the extraction was repeated two more times. The combined extraction volume was made up to 25 mL and filtered through 0.45 µm. Liquid samples were simply diluted and filtered before measurement. The analysis of extracts using the above method gives a complex electropherogram (figure 2). Identification of CA is more problematic. When injected individually as a standard it migrates in an area occupied in the sample injection by three peaks, all of which have similar spectra. By spiking samples with pure CA it can be unequivocally identified in the prepared TCM extract (figure 3). The table below shows the calculated amounts of CA in *Flos Lonicerae*, in some TCMs and in a coffee extract.

Reference

1.
Long, H, Yang, J, Liu, H, Wang, T, Huang, A and Sun, Y, *Journal of Chinese Pharmaceutical Sciences, 8, 152-157,* **1998.**

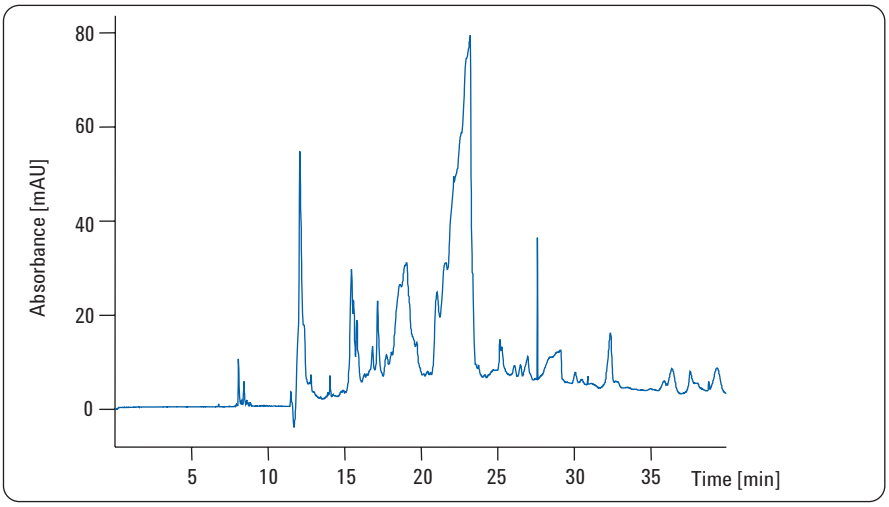


Figure 2
Extract of zhi zi jin hua wan TCM.

Chromatographic conditions

Injection:	10 sec @ 50 mbar
Capillary:	L= 64.5 cm, l= 56 cm, 50 µm id
Buffer:	40 mM phosphate, 80 mM boric acid containing 5 %, ethanol with apparant pH adjusted to 7.0
Voltage:	20 kV:
Temperature:	20°C
Detection:	254 nm

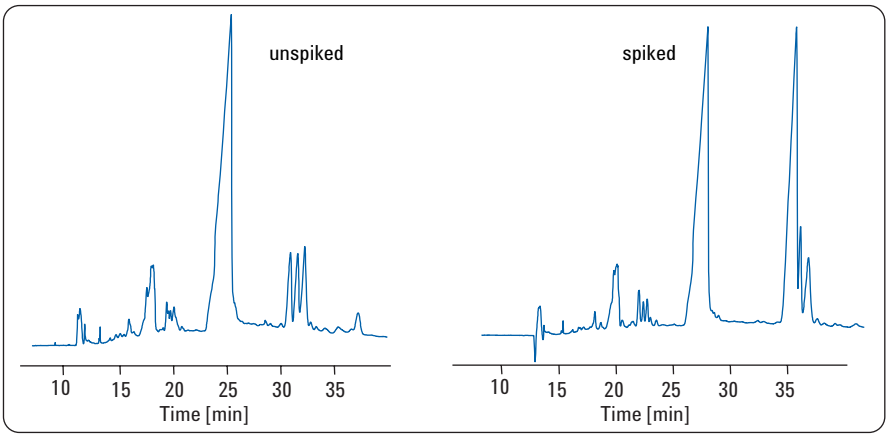


Figure 3
Identification of chlorogenic acid in shung huang lian by spiking.

Medicine	Chlorogenic acid [mg/L]	Powder weight [g]	Content [mg/g] or [mg/mL]
Flos Lonicerae	670	0.946	17.83
Zhi zi jin hua wan	61.18	3.34	0.49
Vc yin qiao pain	104.1	2.83	0.85
Yin qiao jie du pain	542.9	2.9	3.50
Xiao er qing re jie du kou fu ye	31.14	liquid	0.39
Shung huang lian	187.8	liquid	2.10

Table 1
Calculated CA content.

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