Simultaneous Analysis of Ingredients in Chinese Medicinal preparations by High Performance Liquid Chromatography

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Merits of Chinese medicine products depend on the quality and efficacy of its preparation materials. To extend the development of Chinese medicine, the quality control of Chinese herbal medicines (CHM) must be promoted previously since the pharmacologic efficacy of CHM can be influenced by the uneven quality of CHM. Department of Health, Executive Yuen, announced that certification of validity period extension of new drugs detection registration and drugs permission, no matter indigenously made or imported concentrated preparations, the attached data must follow the act of “Marker components quantification methods and specs notification of concentration preparations of Chinese medicine” and 33 preparations include Ger- Gen-Tang, Siao-Ching-Lung-Tang, Jia-Wei-Siao-Yao-San, Qwei-Je-Tang, Gan-Lu-Yin, Ma-Sing-Gan-Seg-Tang, Bu-Chung-Yi-Chi-Tang, Lieu-Wei-Di-Hung-Wan, Huang-Lian-Jei-Du-Tang, Du-Ho-Gi-Sen-Tang (Jan.1st, 2001), Ze-Bor-Di-Hunag- Wan, Lung-Dan-Sei-Gan-Tang, Sin-Yi-Ching-Fe-Tang, Sui-Fu-Ju-Yu-Tang, Chi- Jui-Di-Hung-Wan, Siao-Fun-San, Ching-Sin-Liang-Zu-Yin, Si-Ni-San, Ding-Chuang- San, Chai-Ger-Jei-Ji-Tang (Feb. 1st, 2003), Che-Gan-Tsau-Tang, Ba-Wei-Di-Hung- Wan, Chuan-Chung-Cha-Tiau-San, Siao-Yao-San, Huo-Siang-Cheng-Chi-San, Siang- Sa-Lieu-Jun-Zu-Tang, Jing-Fang-Bai-Du-San, Su-Jing-Huo-Sei-Tang, Ze-Ker-San, Ji-Seng-Sen-Chi-Wan, Fang-Fung-Tung-Seng-San, Err-Cheng-Tang, Lieu-Jun-Zu- Tang (Aug. 23rd, 2006) were announced. If the data did not follow the rule of the act, new drugs can not be registered and certification of validity period extension will not be allowed. According to the act, every prescription should quantify at least two marker components chosen from different crude materials. However, only 33 preparations including Ger-Gen-Tang were announced that they must do marker component quantification. It will be the trend of the future Chinese herbal medicine development.

In our laboratory committed developing HPLC analysis methods for different Chinese medicine preparations and dosage formulations. The analysis techniques will be established by first using column chromatography of various packing materials (including silica gel, sephadex LH-20), or preparative thin layer chromatography to purify and to identify marker components. Meanwhile, HPLC analysis methods of marker components were developed and were tested by inter-day, intra-day, and recovery validation. The development of high stability, high reproducibility, and trusty technology of simultaneous multi marker components analysis may reduce the analysis period of materials and preparations, enhance efficacy of quantification, and setup standard operation procedure and criteria of detection to reach the goal of quality control and assure the efficacy and safety of traditional or commercial Chinese medicine preparations. The results of the HPLC analysis methods developed on relative preparations were listed and elucidated below:

1 Simultaneous Analysis of Eight Components in “Pin-Wei-San” by High Performance Liquid Chromatography

Simultaneous determination of eight marker substances was established for the quality control of Chinese medicinal preparation “Ping-Wei-San” by HPLC. These substances were glycyrhrizin in Glycyrrhizae Radix, hesperidin, nobiletin, 3’,4’,3,5,6,7,8-heptamethoxy flavone, tangeretin, 5-hydroxy-3’,4’,6,7,8- pentamethoxy flavone in Citri Leiocarpae Exocarpium, honokiol, magnonol in Magnoliae Coatex. Extracted sample were run through the HPLC column (Inertsil ODS-80A 5μm, 4.6 mmφ×250 mm) at 30°C and the column was developed with a mixture of 20% acetonitrile (pH 2.5) and 70% acetonitrile (pH 2.5) aqueous solution employing linear gradient elution method at a flow-rate of 1.0 ml/min. The detection wavelength varied with time. It was 275 nm during 0–19 min, 250 nm during 19–80 min.
Simultaneous determination of five marker substances was established for the quality control in traditional Chinese medicine preparation of "Tzyy-Yun-Gau". These substances included shikonin, deoxyshikonin, β,β-dimethylacryl shikonin, and acetylschizandrin in Schizandrae Fructus; baicalin and baicalein in Scutellariae Radix. Different rice wine extraction volume and extraction temperature conditions were performed to evaluate quality of "King-Mon-Long-Fong-Jyo". Extracted samples were analyzed with reversed-phase column (Inertsil ODS-2, 4.6 mm I.D. × 250 mm) at 30°C and the column was developed with a mixture of 20% acetonitrile and 70% acetonitrile aqueous solution and then employed linear gradient elution method at a flow-rate of 1.0 mL/min. An UV 250 nm was used for the detection of the marker substances.

Simultaneous determination of seven marker substances was established for the quality control in tonic wine preparation of "King-Mon-Long-Fong-Jyo". These marker substances were gomisin A and schizandrin from Schizandrae Fructus, loganin from Corni Fructus, cinnamic acid and cinnamaldehyde from Cinnamomi Cortex, and scopoletin and ferulic acid from Angelicae Radix. Different rice wine extraction volume and extraction temperature conditions were performed to evaluate quality of "King-Mon-Long-Fong-Jyo". Extracted samples were run through the HPLC column (Inertsil ODS-2, 4.6 mm I.D. × 250 mm) at 30°C and the column was developed with a mixture of 20% acetonitrile and 70% acetonitrile aqueous solution and then employed linear gradient elution method at a flow-rate of 1.0 mL/min. An UV 250 nm was used for the detection of the marker substances.

Simultaneous determination of nine marker substances was established for the quality control of "Byi-Liang-Tang" by HPLC. These marker substances included berberine from Phellodendri Cortex, curcumin (Curcumae Rhizoma), imperatorin (Angelicae Dahuricae Radix), magnolol (Magnoliae Cortex), hesperidin (Citri Leiocarpae Exocarpium), glycyrrhizin (Glycyrrhizae Radix), and emodin, sennoside A, sennoside B (Rhei Rhizoma). The ingredients in the formula for water-base and oil-base patches from different manufactures were also analyzed for quality evaluation. Extracted samples were analyzed with reversed-phase column (Inertsil 5 ODS-2, 4.6 I.D. × 250 mm) at 30°C and eluted with a mixture of 20% acetonitrile and 70% acetonitrile aqueous solution in gradient manner at a flow-rate of 1.0 mL/min, and detected at 230 nm.

Simultaneous determination of seven marker substances was established for the quality control in patch formula preparation of Ru-Yi-Jin-Huang-San by HPLC. These marker substances included berberine (Phellodendri Cortex), curcumin (Curcumae Rhizoma), imperatorin (Angelicae Dahuricae Radix), magnolol (Magnoliae Cortex), hesperidin (Citri Leiocarpae Exocarpium), glycyrrhizin (Glycyrrhizae Radix), and emodin, sennoside A, sennoside B (Rhei Rhizoma). The ingredients in the water-based and oil-based patches of the formula from different manufactures were also analyzed for quality evaluation. Extracted samples were analyzed with reversed-phase column (Inertsil 5 ODS-2, 4.6 I.D. × 250 mm) at 30°C and eluted with a mixture of 20% acetonitrile and 70% acetonitrile aqueous solution in gradient manner at a flow-rate of 1.0 mL/min. The detection wavelength varied with time, which was 275 nm during 0–72 min, 250 nm during 72–105 min, and lastly 220 nm during 105–145 min.

REFERENCES