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The Producing Area of Chinese Medicine and Famous Region Drug Research – Magnolia Officinalis

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1. Introduction

1.1 Producing area

Under the guidance of the theory of the Traditional Chinese Medicine, many plants, animals and minerals in China can be medicines to treat the human diseases. Synchronously, the capacious land, sea and rich mineral deposits supply resources for them. Before they become medicines, they need grow and be gathered in some areas. But in these areas the weather, geographic features and biologic distributions are very different such as the different natural zones. Therefore these comprehensive factors endow Chinese medicines with different biological features and effects. After long time use, the medical practitioners gradually found the quantity or quality of some Chinese medicines which were produced in different areas were also different. An appellation came into being, which was called famous region drug.

1.2 Famous region drug

This appellation means those medicines which have good breeds and higher quality grow in some characteristic regions and feasible growing environments. They are cultivated and processed reasonably and output is great.

One thousand and five hundred years ago, in (Annotated Shen Nong's Herbal), Jinghong Tao had discussed the producing areas of Chinese Medicine. But at Yuan dynasty, famous region drug were first written in (Peony Pavilion) Xianzu Tang wrote. Until Tang dynasty, the government divided the country into ten areas according natural form. So Simiao Sun recorded famous region drugs producing in these areas in(a supplement to the essential prescriptions worth a thousand gold). (commentaries on the illustrations)written by Song Su and (Compendium of Materia Medica)written by Shizhen Li both recorded the regions and quality of Chinese Medicine. Now some places in China are famous regions of Chinese Medicines, such as Sichuan province, Guangdong province, Shandong province where respectively produced Rhizoma Chuanxiong, Fructus Amomi, Equus asinus L.

1.3 Magnolia officinalis

Magnolia officinalis are from the barks of Magnolia officinalis Reha. et Wils. and Magnolia officinalis Reha. et Wils.var. biloba. Reha. et Wils which belong to Magnoliaceae [1]. Its

producing areas mainly include Sichuan, Hubei and Jiangxi province [2]. The characters of its medicinal materials usually showed single or double drum, grey outer surface with vertical wrinkles, purple brown inter surface with wiped oil mark and section with fine crystallization. Microstructure of its medicinal materials usually showed stone and oil cells or starch grains. The modern research also showed that its main effective component is magnolol and honokiol.

Fig. 1. Medicinal plant of Magnolia officinalis

Fig. 2. Medicinal materials of Magnolia officinalis

It is wide used in clinic due to the effect of eliminating dampness and phlegm, promoting qi and removing distention. But the resource of Magnolia officinalis is decreasing and the

imbalance between supply and demand is outstanding in recent years. It has been involved in natural protected Chinese medicine. Its variety research seems very urgent for looking for its substitute.

2. Purpose

To explore the exact substitute and the quality evaluation, optimization methods of Magnolia officinalis, the comprehensive varieties researches based on its adulterants and substitutes and quality evaluation methods were reviewed.

3. Methods

The appearance characters, microstructure, physical and chemical(including Thin-Layer Chromatography and high performance liquid chromatography) and randomly amplified polymorphic DNA(RAPD) identification, 1H Nuclear magnetic resonance spectroscopy, powder X-ray diffraction fourier fingerprint, polyamide chromatography of different adulterants and substitutes were applied according to the records. And they were reviewed. The results showed that they totally contained ten families and forty - three varieties. The influence factors on the quality of Magnolia officinalis and the other quality evaluation methods were also reviewed.

4. Result and conclusion

4.1 Families and varieties of plants identified with Magnolia officinalis and identified methods

The difference between Magnolia officinalis and these adulterants and substitutes were significant through the research. Firstly the morphological and(or) histological characters of some varieties were different from Magnolia officinalis, such as sikimmi, aleurites montana, Magnolia rostrata w w smith, Manglietia szechuanica Hu., Neolitsea levinei Merr., phyllanthi fructus, White yulan Magnolia, bigleaf magnolia bark, Wudang yulan magnolia, Magnolia wilsonii, camphortree bark , M.szechuanica Hu, M.insignis(Wal1.)Bl, Manglietia chingii Dandy, Manglietia yuyuanensis Law, sprenger magnolia bark, sargent magnolia bark, Magnolia wilsonii , Magnolia campbellii Hook.f.et Thoms., Mountain yulan Magnolia, M.szechuanica Hu, Manglietia chingii Dandy [3-14]. Secondly some physical and chemical identification showed significant results between Magnolia officinalis, Wudang magnolia bark, Magnolia campbellii Hook.f.et Thoms., albiziae , thinleaf machilus and Ormosia balansae. [15-17].The thin-Layer Chromatography analyzing also showed Magnolia officinalis were very different from the following adulterants and substitutes: (1)Wudang magnolia bark, Xikang magnolia bark and sargent magnolia bark were identified with cyclohexane-chloroform-ethanol (7:3:1) as developer and 5% vanillin sulfuric acid solution as coloration using silica gel G and observed at 254 nm using ultraviolet lamp [18]. (2)White yulan Magnolia, biond magnolia flower, shikimmi, red nanmu, aleurites montana, schima root-bark, Albizziakal Kora(Roxb).Prain., arbutus were identified with benzene-methanol (27:1) as developer and vanillin sulfuric acid solution as coloration using silica gel G and observed at 365 nm using ultraviolet lamp [19]. (3)Magnolia campbellii Hook.f.et Thoms.was identified with benzene-methanol (9:1) as developer and 1% vanillin sulfuric acid solution as coloration using silica gel G [20]. (4) Guangxi Manglietia was identified with benzene-methanol (8:2) as developer and 5% vanillin sulfuric acid solution as coloration

families	varieties numbers	varieties
Magnoliaceae	26	sprenger magnolia bark、sargent magnolia bark、Magnolia campbellii Hook.f.et Thoms.、Xikang magnolia bark 、Wudang yulan magnolia、white yulanMagnolia、Mountain yulan Magnolia、Magnolia wilsonii、Tianmu mountain magnolia immature flower、bigleaf magnolia bark、Magnolia rostrata w．w．smith、purple yulan Magnolia、magnolia bioudii、Mt. Huang magnolia bark、M.szechuanica Hu、M.insignis(Wall.)Bl、Manglietia chingii Dandy、Manglietia yuyuanensis Law、 Guangxi Manglietia、liriodendron 、biond magnolia flower、 shikimmi 、Michelia maudiae、Qinshi Manglietia、Magnolietia patungensis、Michelia champaca L..
Euphorbiaceae	2	phyllanthi fructus、 aleurites montana
lauraceae	4	red nanmu、 thinleaf machilus 、 camphortree bark 、 Neolitsea levinei Merr.
Juglandaceae	3	Engelhardia roxburghiana Wall 、 wild walnut 、 Juglans mandshurica Maxim.
myricaceae	1	arbutus
araliaceae	1	Manglietia szechuanica Hu.
Scrophulariaceae	1	Paulownia tomentosa (Thunb.) Steud.
Theaceae	1	schima root-bark
Leguminosae	3	Albizziakal Kora(Roxb).Prain.、 Ormosia balansae 、 albiziae
Oleaceae	1	Fraxinus rhynchophylla Hance

Table 1. Families and varieties of plants identified with Magnolia officinalis

using silica gel G [21]. (5) Juglans mandshurica Maxim.,Wild walnut, Paulownia tomentosa (Thunb.) Steud.and Mountain yulanMagnolia were identified with benzene-methanol (27:1) as developer and 1% vanillin sulfuric acid solution as coloration using silica gel G [22-25]. (6) Tianmu mountain magnolia immature flower was identified with benzene-ethyl acetate-methanol (27:1) as developer using silica gel G [26]. (7)Mt. Huang magnolia bar k, Manglietia yuyuanensis Law, Michelia maudiae were identified with benzene-methanol (27:1) as developer and 5% vanillin sulfuric acid solution as coloration using silica gel G [27]. (8) Qinshi Manglietia was identified with chloroform-methanol as developer and 5% vanillin sulfuric acid solution as coloration using silica gel G [28]. (9)Engelhardia roxburghiana Wal1 was identified with benzene-methanol (9:1) as developer and 1% vanillin sulfuric acid solution as coloration using silica gel G [29]. High performance liquid chromatography(HPLC) detection showed the total amount of magnolol and honokiol in Magnolia wilsonii, sprenger magnolia bark, and sargent magnolia bark were corresponding with PRC codex 2005 which was on Permaphase ODS, under the condition of mobile phase of benzene:phosphate(70:30) with the flow rate of 1.0ml/min. The calibration curves showed linear regression $r > 0.9989$. The recoveries ranged from 99.04% to 105.45%. [30]. But there

was no magnolol and honokiol in white yulan Magnolia, purple yulan Magnolia, magnolia bioudii and Wudang yulan magnolia on Permaphase ODS，under the condition of mobile phase of benzene:water (74.5:25.5) with the flow rate of 1.0ml/min. Most of manglietia and Mountain yulan Magnolia contain small amounts of magnolol and honokiol [31]. The randomly amplified polymorphic DNA was also used to identify the certified products and the adulterants and substitutes of Magnolia officinalis like yulan magnolia, magnolia bioudii, Wudang yulan magnolia, purple yulan Magnolia, Mountain yulan Magnolia, Manglietia chingii Dandy, M.insignis(Wal1.)Bl and liriodendron.The result showed that the DNA Fingerprinting of Magnolia officinalis was very different from that of these adulterants and substitutes [32]. 15 samples of cortex Magnolia officinalis from different origin, 1 standard sample of Magnolia officinalis and 1 Wudang yulan magnolia were identified by 1H Nuclear magnetic resonance spectroscopy. The fingerprint showed that 1H-NMR could be an accurate and feasible method for the quality control of Magnolia officinalis [33]. Powder X-ray diffraction fourier fingerprint pattern was developed to identify and analyze Magnolia officinalis. Experiments and analysis were carried out on 3 samples of cortex Magnoliae officinals, 3 samples of cortex Magnolia bilobae, 14 samples of substitute and one counterfeit of cortex Magnolia officinals. It was found that this method can be used for identification on Chinese medicinal material Cortex Magnolia officinals [34].The characteristic chromatogram for cortex Magnolia officinalis was established by using polyamide chromatography (PC) to identify the cortex Magnolia officinalis from different origin and its substitute and false products. The result showed that this method could be used for the identification and quality evaluation for cortex Magnolia officinalis [35].

4.2 Influence factors and determinative methods of quality of Magnolia officinalis

The content of magnolol and honokiol of Magnolia officinalis collected from 7 provinces, 11 counties and Jingning provenance testing forest of Zhejiang province were detected as quality standards by using HPLC. And the correlation analysis between the factors which would affect the quality of Magnolia officinalis and the content of magnolol and honokiol was applied. The result showed that many factors would affect the quality of Magnolia officinalis significantly, such as provenance, producing area, blade profile, DBH, tree height, crown width, tree age, bar thickness, powder color, oiliness, grindability, bark type and position of sampling, etc. Among them, provenance, blade profile, powder color, bark thickness, DBH and position of sampling were more significant factors, especially the variety [36]. However some other researches showed tree age and length of storage period would be two factors which affected the quality of Magnolia officinalis [37-38]. After analysis, geographical position and Climate, blade profile and variety firstly maybe would affect the quality of Magnolia officinalis, especially the provenances with a sharpened leaf tip from Hubei Province has a highest content of phenols, and that with a concave leaf tip from the Lushan Mountain has a lowest content of phenols [39-40]. Secondly, there was positive correlation between the growth factors of Magnolia officinalis including tree age, DBH, tree height, crown width and content of phenols of Magnolia officinalis, especially the DBH [41]. Thirdly, appearance characters including bark thickness, powder color, grindability and bark roughness were regarded as the main traditional basis for the quality evaluation of Magnolia officinalis, which accorded with the research [42]. Position of

sampling was also a basis for the quality evaluation of Magnolia officinalis which included dry hide, root bark, shoot cortex, etc. Content of phenols of root bark from Magnolia officinalis was 3-5 times higher than that of shoot cortex from Magnolia officinalis [41].
Besides the TLC and HPLC, amplified fragment length polymorphism analysis was applied as a method for further study with molecular markers in the field of genetic diversity, for breeding new cultivars, and for genetic relationships with Magnolia officinalis [43]. First derivative of UV spectrophotometry was also used to analyze compositions of Magnolia officinalis. The result showed that this method could eliminate interference of other impurity and make all samples exhibit maximum absorbance at 300nm [44]. A gas chromatography/mass spectrography was developed to identify the compositions of Magnolia officinalis, including magnolol, honokiol, δ-selinene and β-eudeomol, etc. And the content of magnolol and honokiol were determined [45]. The second order derivative synchronous fluorescence spectra of magnolol, honokiol and their mixture in 0.08%-0.16% methanol solution were studied. The experiment results indicated that their second order derivative synchronous fluorescence spectra were separated absolutely, which eliminated the disturbance between them [46]. To detect the contents of six trace elements (Fe ,Cu ,Zn ,Mn ,Ca ,Mg) in cortex Magnolia officinalis by flame atomic absorption spectroscopy, the contents of six trace elements (Fe ,Cu ,Zn , Mn ,Ca and Mg) were determined by calibration curve method. The result showed that the Cortex Magnolia officinalis is rich in trace elements which are necessary for people [47]. Quantitative Measurement of magnolol and honokiol was tested by using excitation–emission Matrix Fluorescence coupled with second–order calibration algorithm. It showed that the second-order calibration methods could quantify the analysis of interest from overlapped chromatographic profiles and give the accurate predicted results by utilizing mathematical-separation instead of physics-separation [48]. Fluorescent determination of magnolol has been effected employing the sensitivity- and stability-enhancing action of the non-ionic surface active emulsifier OP. As a result, the accuracy of determination was raised by 2 orders of magnitude as compared to that of ultraviolet spectrophotometry [49]. A highly sensitive and selective method was developed for the determination of honokiol and magnolol by HPLC-electrochemical detection, using a microbore column. The result showed that this method could be proposed for the determination of honokiol and magnolol in traditional Chinese medicines and human plasma sample [50].
All above-mentioned researches showed there were four kinds of factors mainly affecting the quality of Magnolia officinalis. And many methods had been used to determinate the content or compositions of Magnolia officinalis for the identification or quality control. Making and following the comprehensive and well-considered plans to develop famous region drugs would benefit protecting the environment, rare Chinese Medicine and highlighting characteristic of Chinese Medicines significantly.

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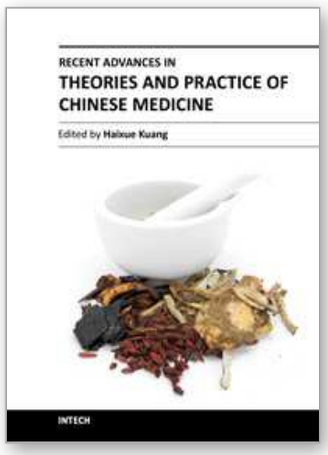

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During the recent years, traditional Chinese medicine (TCM) has attracted the attention of researchers all over the world. It is looked upon not only as a bright pearl, but also a treasure house of ancient Chinese culture. Nowadays, TCM has become a subject area with high potential and the possibility for original innovation. This book titled Recent Advances in Theories and Practice of Chinese Medicine provides an authoritative and cutting-edge insight into TCM research, including its basic theories, diagnostic approach, current clinical applications, latest advances, and more. It discusses many often neglected important issues, such as the theory of TCM property, and how to carry out TCM research in the direction of TCM property theory using modern scientific technology. The authors of this book comprise an international group of recognized researchers who possess abundant clinical knowledge and research background due to their years of practicing TCM. Hopefully, this book will help our readers gain a deeper understanding of the unique characteristics of Chinese medicine.

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