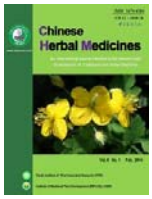

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Letter

Chemical Constituents from Barks of *Lannea coromandelica*

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ABSTRACT

Objective To study the chemical constituents from the barks of *Lannea coromandelica*. **Methods** The chemical constituents were isolated and purified by column chromatography on silica gel column. NMR spectra were used for structural identification. **Results** Thirteen compounds were isolated and identified as quercetin (**1**), (2*S*,3*S*,4*R*,10*E*)-2-[(2′*R*)-2′-hydroxytetracosanoyl amino]-10-octadecene-1,3,4-triol (**2**), aralia cerebroside (**3**), 5,5′-dibuthoxy-2,2′-bifuran (**4**), β-sitosteryl-3β-glucopyranoside-6′-*O*-palmitate (**5**), β-sitosterol palmitate (**6**), myricadiol (**7**), protocatechuic acid (**8**), *p*-hydroxybenzoic acidethyl ester (**9**), isovanillin (**10**), *trans*-cinnamic acid (**11**), palmitic acid (**12**), and stearic acid (**13**). **Conclusion** Compounds **2**−**13** are isolated from this plant for the first time.

Key words

Anacardiaceae; isovanillin; *Lannea coromandelica*; myricadiol; *trans*-cinnamic acid

1. Introduction

Lannea coromandelica (Houtt.) Merr. (Anacardiaceae) is a deciduous tropical tree widely distributed in Bangladesh, India, and some other tropical countries. In China, *L. coromandelica* could be found in Hainan, Yunnan, Guangdong, and Guangxi provinces (South China Research Institute of Plants, Chinese Academy of Sciences, 1964). The bark of *L. coromandelica* is useful in treating cuts, wounds, bruises, ulcers, gastritis, enteritis, leukorrhagia, ophthalmia, gout, ulcerative stomatitis, odontalgia, sprains, diarrhea, and dysentery, and the leaves could be used in treating elephantiasis, inflammation, neuralgia, sprains, and bruises (Islam and Tahara, 2000; Gan et al, 2007). The fruits paste of *L. coromandelica* is therapeutically used for bone fractures by tribes in eastern Ghat of Andhra Pradesh (Venkata and Venkata, 2008). The pharmacological properties of the extract from the stem barks of *L. coromandelica* were screened for anti-inflammatory (Singh and Singh, 2005), hypotensive (Islam et al, 2002), and cytotoxic effects (Rahman et al, 2008). Five dihydroflavonols have been isolated and identified from the stem barks of *L. coromandelica* (Islam and Tahara, 2000).

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To further explore and make good use of this Chinese herbal source, the chemical constituents in the barks of *L. coromandelica* were investigated and compounds **1**−**13** were isolated. Compounds **2**−**13** were isolated from this plant for the first time.

2. Materials and methods

2.1 Apparatus and reagents

The NMR spectra were recorded on a Bruker Avance−400 Instrument in deuterated chloroform and acetone with TMS as internal standard. Silica gel (200−300 mesh, Qingdao Marine Chemical, China) and Sephadex LH-20 (Amersham Pharmacia Biotech., Hongkong, China) chromatographies were used for column chromatography. Precoated silica gel GF_{254} plates and RP-18 F_{254} plates (0.25 mm, Merck, Germany) were used for TLC.

2.2 Plant material

The barks of *Lannea coromandelica* (Houtt.) Merr. were collected in June 2010 from Hainan province, China, and authenticated by Prof. Qiong-xin Zhong, College of Life Science, Hainan Normal University. A voucher specimen (201006) of *L. coromandelica* was deposited in the Key Laboratory of Tropical Medicinal Plant Chemistry, Ministry of Education, Hainan Normal University, Haikou, China. The material was then air-dried and coarsely powdered.

2.3 Extraction and isolation

The dried powdered stem barks (6.0 kg) of *L. coromandelica* were extracted with 78% ethanol for three times at room temperature. After removing the solvent under reduced pressure, an aliquot of the crude extract (0.8 kg) was suspended in H_2O and the aqueous suspension was successively extracted for three times each with petroleum ether, $CHCl_3$, and EtOAc, respectively.

The petroleum ether extract, upon concentration under reduced pressure, afforded a Blackish green syrup (60.2 g). This syrup was subjected to column chromatography (CC, 100 mesh) on silica gel (1.28 kg) eluting with a petroleum ether-EtOAc gradient (100:0→0:100), yielding 11 fractions (Frs. A−K). Further purification was submitted to silica gel, Sephadex LH-20 chromatography, and preparative TLC. Fr. D yielded compounds **7** (15 mg), **12** (18 mg), and **13** (16 mg). From Fr. F, compound **6** (36 mg) was purified. Fr. I yielded compound **3** (40 mg) and Fr. J yielded compound **10** (13 mg).

The $CHCl_3$ extract, upon concentration under reduced pressure, afforded a Blackish green syrup (18 g). This syrup was subjected to CC (100 mesh) on silica gel (0.38 kg) eluting with a petroleum ether-EtOAc gradient (100:0→0:100), yielding Frs. A−K. Further purification was submitted to silica gel, Sephadex LH-20 chromatography, and preparative TLC. Fr. C yielded compound **4** (12 mg). Fr. G yielded compound **11** (15 mg). From Fr. I, compound **5** (30 mg) was purified.

The EtOAc extract, upon concentration under reduced pressure, afforded reddish brown syrup (33.6 g). This syrup was subjected to CC (100 mesh) on silica gel (0.62 kg) eluting with a petroleum ether-EtOAc gradient (100:0→0:100), yielding Frs. A−K. Further purification was submitted to silica gel, Sephadex LH-20 chromatography, and preparative TLC. Fr. D yielded compound **9** (16 mg). Fr. F yielded compound **8** (17 mg). Fr. G yielded compound **1** (22 mg). From Fr. I, compound **2** (23 mg) was purified. The chemical structures of compounds **2**−**7** are shown in Figure 1.

3. Results and discussion

3.1 Structure identification

Compound **1**: yellow needles crystal. ^{1}H-NMR (400 MHz, CD_3OD) δ: 7.70 (1H, d, J = 2.0 Hz, H-2′), 7.60 (1H, dd, J = 8.4, 2.0 Hz, H-6′), 6.90 (1H, d, J = 8.8 Hz, H-5′), 6.39 (1H, d, J = 2.0 Hz, H-8), 6.18 (1H, d, J = 2.0 Hz, H-6). ^{13}C-NMR (100 MHz, CD_3OD) δ: 177.3 (C-4), 165.3 (C-7), 162.1 (C-9), 158.0 (C-5), 148.6 (C-4′), 148.2 (C-2), 146.0 (C-3′), 137.1 (C-3), 124.0 (C-1′), 121.8 (C-6′), 116.3 (C-5′), 116.0 (C-2′), 104.5 (C-10), 99.3 (C-6), 94.6 (C-8). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Yao et al, 2013; Zhang et al, 2010), and compound **1** was identified as quercetin.

Compound **2**: white plate solid. ^{1}H-NMR (400 MHz, C_5D_5N) δ: 8.49 (1H, d, J = 8.8 Hz, N-H), 6.58 (1H, m, H-11), 5.43 (1H, m, H-10), 5.02 (1H, dd, J = 9.2, 4.8 Hz, H-2), 4.54 (1H, dd, J = 7.6, 3.6 Hz, H-2′), 4.42 (1H, dd, J = 10.6, 4.6 Hz, H-1a), 4.33 (1H, dd, J = 10.8, 5.2 Hz, H-1b), 4.27 (1H, m, H-3), 4.19 (1H, m, H-4). ^{13}C-NMR (100 MHz, C_5D_5N) δ: 175.6 (C-1′), 131.1 (C-11), 131.0 (C-10), 77.1 (C-3), 73.3 (C-4), 72.8 (C-2′), 62.4 (C-1), 53.3 (C-2), 36.0 (C-3′), 34.4 (C-5), 33.6 (C-12), 32.4 (C-9), 29.8-30.6 (C-13−C-16), 29.8−30.6 (C-5′−C-22′), 27.0 (C-6), 26.1 (C-4′), 23.2 (C-17, C-23′), 14.6 (C-18, C-24′). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Zhan et al, 2003), and compound **2** was identified as (2*S*,3*S*,4*R*,10*E*)-2-[(2′*R*)-2′-hydroxy-tetracosanoyl amino]-10-octadecene-1,3,4-triol.

Compound **3**: white powder. ^{1}H-NMR (400 MHz, C_5D_5N) δ: 8.50 (1H, d, N-H), 5.82 (2H, H-8, H-9), 5.43 (1H, H-2), 4.96 (1H, H-1″), 4.53, 4.41, 4.34, 4.27, 4.20 (5H, H-1a, H-2′, H-1b, H-3, H-4), 0.77 (6H, $2CH_3$). ^{13}C-NMR (100 MHz, C_5D_5N) δ: 175.0 (C-1′), 130.6 (C-8), 130.5 (C-9), 107.2 (C-1″), 76.6 (C-3″, C-5″), 76.5 (C-3), 72.8(C-2″), 72.7 (C-4, C-2′, C-4″), 72.2 (C-1), 61.8 (C-6″), 52.7 (C-2), 35.5 (C-3′), 33.9 (C-5), 33.6 (C-10), 32.8 (C-7), 26.4 (C-6), 22.7−33.1 (C-11−C-17, C-4′−C-15′), 14.1 (C-18, C-16′). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Zou and Yang, 2008), and compound **3** was identified as aralia cerebroside.

Compound **4**: pale yellow amorphous powder. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 7.72 (2H, dd, J = 6.4, 3.2 Hz, H-3, H-3′), 7.53 (2H, dd, J = 6.4, 3.2 Hz, H-4, H-4′), 4.30 (4H, t, J = 6.8 Hz, H-6, H-6′), 1.72 (4H, m, H-7, H-7′), 1.44 (4H, m, H-8,

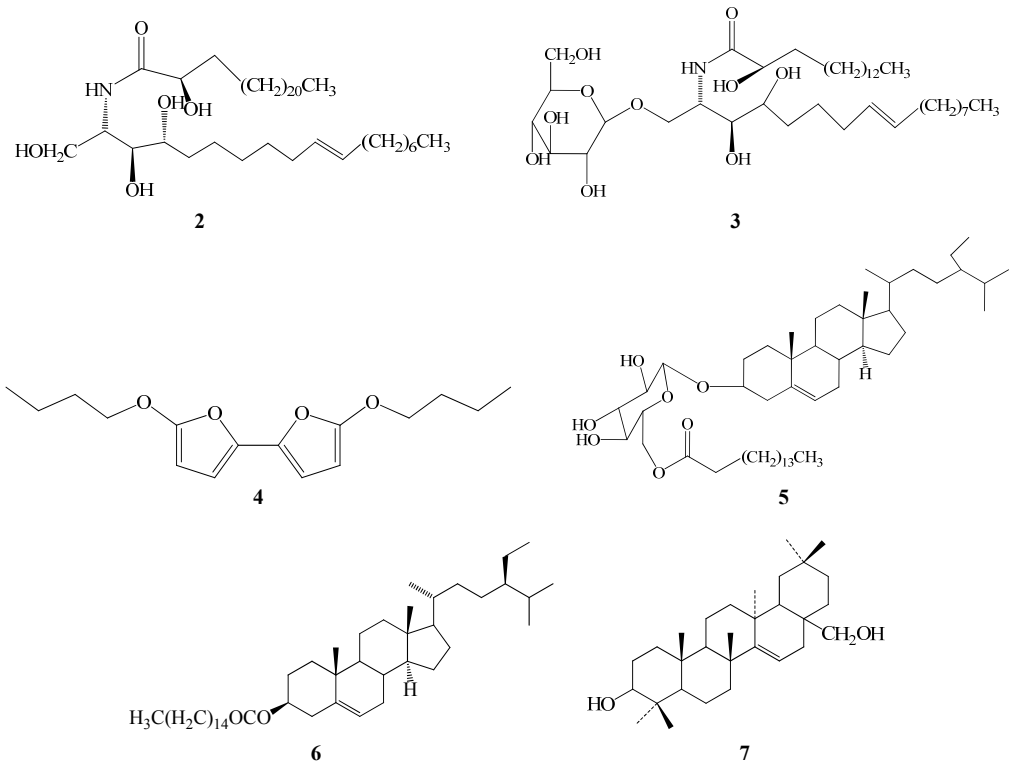

Figure 1 Chemical structures of compounds 2−7

H-8′), 0.96 (6H, t, *J* = 7.4 Hz, H-9, H-9′); ^{13}C-NMR (100 MHz, $CDCl_3$) *δ*: 167.7 (C-5), 167.7 (C-5′), 132.3 (C-2,C-2′), 130.9 (C-4, C-4′), 128.8 (C-3, C-3′), 65.6 (C-6, C-6′), 30.6(C-7, C-7′), 19.2 (C-8, C-8′), 13.7 (C-9, C-9′). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Liu et al, 2010), and compound **4** was identified as 5,5′- dibuthoxy-2,2′-bifuran.

Compound **5**: white powder. ^{1}H-NMR (400 MHz, C_5D_5N) *δ*: 0.56, 0.84 (3H each, s, CH_3-18, CH_3-19), 0.89 (3H, d, *J* = 6.4 Hz, CH_3-21), 0.76, 0.77, 0.79, 0.82 (3H each, CH_3-26, CH_3-27, CH_3-29, CH_3-16′); ^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 175.04 (C-1″), 140.69 (C-5), 121.7 (C-6), 102.4 (C-1′), 78.39 (C-3), 78.27 (C-3′), 77.88 (C-5′), 75.12 (C-2′), 71.47 (C-4′), 62.61 (C-6′), 56.61 (C-14), 56.02 (C-17), 50.12 (C-9), 45.82 (C-24), 42.26 (C-13), 39.73 (C-12), 39.12 (C-4), 37.26 (C-1), 36.71 (C-10), 36.17 (C-20), 33.99 (C-2″), 33.89 (C-22), 31.95 (C-7), 31.88 (C-14″), 31.84 (C-8), 30.03 (C-2), 29.77 (C-7″−C-12″), 29.71 (C-6″), 29.68 (C-5″), 29.39 (C-13″), 29.37 (C-4″), 29.24 (C-25), 28.32 (C-16), 26.16 (C-23), 24.29 (C-3″), 24.28 (C-15), 23.17 (C-28), 22.7 (C-15″), 21.06 (C-11), 19.76 (C-27), 19.20 (C-19), 18.99 (C-26), 18.79 (C-21), 14.04 (C-16″), 11.94 (C-29), 11.75 (C-18). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Nguyen et al, 2004), and compound **5** was identified as β-sitosteryl-3-*O*-β-6′-*O*-palmitate.

Compound **6**: colorless needles crystal. ^{1}H-NMR (400 MHz,$CDCl_3$) *δ*: 0.68, 1.00 (3H each, s, CH_3-18, CH_3-19), 0.93, 0.91, 0.84, 0.82, 0.80 (each 3H, m, CH_3-21, CH_3-16′, CH_3-26, CH_3-29, CH_3-27), 1.24 (nH, br. s), 2.34 (2H, t, *J* = 7.6 Hz), 4.16 (1H, m), 5.35 (1H, d, *J* = 5.2 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$) *δ*: 175.5 (C-1′), 140.8 (C-5), 121.7 (C-6), 71.8 (C-3), 56.8 (C-14), 56.1 (C-17), 50.2 (C-9), 45.9 (C-24), 42.3 (C-4, C-13), 39.8 (C-12), 37.3 (C-1), 36.5 (C-10), 36.2 (C-20), 34.1 (C-2′), 34.0 (C-22), 32.0 (C-7), 31.9 (C-8, C-14′), 31.7 (C-2), 29.7 (C-10′, C-11′, C-12′, C-13′), 29.6 (C-9′), 29.5 (C-8′), 29.4 (C-7′), 29.3 (C-6′), 29.2 (C-25, C-4′, C-5′), 28.3 (C-16), 26.1 (C-23), 24.9 (C-3′), 24.3 (C-15), 23.1 (C-28), 22.7 (C-15′), 21.1 (C-11), 19.8 (C-26), 19.4 (C-19), 19.1 (C-27), 18.8 (C-21), 14.1 (C-16′), 12.0 (C-29), 11.9 (C-18). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Sun et al, 2002), and compound **6** was identified as β-sitosterol palmitate.

Compound **7**: white amorphous powder. ^{1}H-NMR (400 MHz, C_5D_5N) *δ*: 0.81, 0.87, 0.90, 0.90, 0.92, 0.98, 1.03 (3H each, 21H, s), 3.34 (1H, dd, *J* = 10.2, 5.8 Hz, H-3), 5.53 (1H, dd, *J* = 7.8, 3.0 Hz, H-15); ^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 158.5 (C-14), 117.1 (C-15), 78.2 (C-3), 62.2 (C-28), 56.0 (C-5), 49.6 (C-18), 49.2 (C-9), 41.7 (C-19), 39.4 (C-4), 39.3 (C-8), 38.3 (C-13, C-17), 38.0 (C-10), 37.8 (C-1), 36.9 (C-7), 34.0 (C-29), 33.8 (C-21), 33.4 (C-16), 32.1 (C-12), 30.0 (C-26), 29.6 (C-20), 29.0 (C-22), 28.7 (C-23), 28.0 (C-2), 26.2 (C-27), 21.5 (C-30), 19.2 (C-6), 17.8 (C-11), 16.4 (C-24), 15.7 (C-25). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Sakurai et al, 1986), and compound **7** was identified as myricadiol.

Compound **8**: colorless needles crystal. ^{1}H-NMR (400 MHz, CD_3OD) *δ*: 7.41 (1H, d, *J* = 2.0 Hz, H-2), 6.80 (1H, d, *J* = 8.0 Hz, H-5), 7.44 (1H, s, H-6). ^{13}C-NMR (100 MHz, CD_3OD) *δ*: 170.4 (-COOH), 151.5 (C-4), 146.0 (C-3), 123.9 (C-6), 123.2 (C-1), 117.8 (C-2), 115.8 (C-5). The ^{1}H-NMR and

^{13}C-NMR data were in agreement with those given in literature (Wu et al, 1999), and compound **8** was identified as protocatechuic acid.

Compound **9**: white crystal. ^{1}H-NMR (400 MHz, CD_3COCD_3) δ: 9.17 (1H, s), 7.95(2H, d, J = 8.4 Hz, H-3, 5), 6.97 (2H, d, J = 8.8 Hz, H-2, 6), 4.31 (2H, q, J = 7.2 Hz), 1.34 (3H, t, J = 7.0 Hz); ^{13}C-NMR (100 MHz, CD_3COCD_3) δ: 167.1 (-C=O), 162.8 (C-4), 132.7 (C-2, C-6), 122.9 (C-1), 116.3 (C-3, C-5), 61.3 (-OCH_2-), 15.0 (-CH_3). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Wang et al, 2005; Yang et al, 2011), and compound **9** was identified as *p*-hydroxybenzoic acid ethyl ester.

Compound **10**: pale yellow needles crystal. ^{1}H-NMR (400 MHz, CD_3COCD_3) δ: 10.62 (1H, s, -CHO), 10.06 (1H, s, -OH), 7.28 (1H, d, J = 8.0 Hz, H-4), 7.22 (1H, d, J = 7.6 Hz, H-6), 6.94 (1H, t, J = 7.8, 15.6 Hz, H-3), 3.84 (3H, S, -OCH_3); ^{13}C-NMR (100 MHz, CD_3COCD_3) δ: 196.8 (-CHO), 152.4 (C-5), 149.3 (C-2), 124.4 (C-1), 122.4 (C-4), 120.4 (C-3), 119.0 (C-6), 56.6 (-OCH_3). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Qiao et al, 2000), and compound **10** was identified as isovanillin.

Compound **11**: white powder. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 10.71 (1H, -COOH), 7.81 (1H, d, J = 16.0 Hz, H-β), 6.48 (1H, d, J = 16.0Hz, H-α), 7.56 (2H, m, H-2, 6), 7.41 (3H, m, H-3, 4, 5); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 172.6 (-COOH), 147.0 (β-C), 133.9 (C-1), 130.7 (C-4), 128.9 (C-2, C-6), 128.3 (C-3, C-5), 117.3 (α-C). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Xu et al, 1999), and compound **11** was identified as *trans*-cinnamic acid.

Compound **12**: white wax solid. EI-MS *m/z*: 256 [M^+], 227, 213, 199, 185, 171, 157, 143, 129, 115, 101, 87, 73, 57, 43, 29. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 2.32 (2H, t, J = 7.4 Hz, H-2), 1.62 (2H, m, H-3), 1.25 (24H, br s), 0.87 (3H, t, J = 6.6 Hz, H-16); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 180.2 (C-1), 34.1 (C-2), 31.9 (C-14), 29.0−29.7 (C-4−C-13), 24.6 (C-3), 22.6 (C-15), 14.0 (C-16). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Wang et al, 2003; Zhao, 2013), and compound **12** was identified as palmitic acid.

Compound **13**: white wax solid. EI-MS *m/z*: 284 [M^+], 185, 129, 85, 73, 60, 57, 43, 29. ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 2.32 (2H, t, J = 7.4 Hz, H-2), 1.62 (2H, m, H-3), 1.25 (28H, br s), 0.87 (3H, t, J = 6.6 Hz, H-16); ^{13}C-NMR (100 MHz, $CDCl_3$) δ: 180.2 (C-1), 34.1 (C-2), 31.9 (C-16), 29.0−29.7 (C-4−C-15), 24.6 (C-3), 22.6 (C-17), 14.0 (C-18). The ^{1}H-NMR and ^{13}C-NMR data were in agreement with those given in literature (Lu et al, 2009), and compound **13** was identified as stearic acid.

3.2 Chemotaxonomic significance

In this study, 13 compounds are isolated and purified from the barks of *L. coromandelica.* To the best of our knowledge, the occurrence of compounds **2**−**13** is reported for the first time in this plant. Furthermore, compounds **2**−**8** and **10**−**13** have not been reported in any species in *Lannea* A. Rich. Related protocatechuic acid was already known from the *Lannea nigritana* (Sc. Ell.) Key (Kapche et al, 2007). On the other hand, the species in *Lannea* A. Rich. seem to have a closer chemotaxonomy relationship to the investigated species, as some compounds containing long side-chain were also reported to be typical in some species of this genus (Queiroz et al, 2003; Amiram et al, 1997; Kapche et al, 2007).

In addition, it should be noted that ceramide (compound **2**) and glycosphingolipid (compound **3**) have not so far been reported to occur in other species of *Lannea* A. Rich., nor in other genera of the Anacardiaceae, and could serve as the chemosystematic marker for *L. coromandelica.*

The above information may give some chemotaxonomic support for the treatment of the investigated species as *L. coromandelica*.

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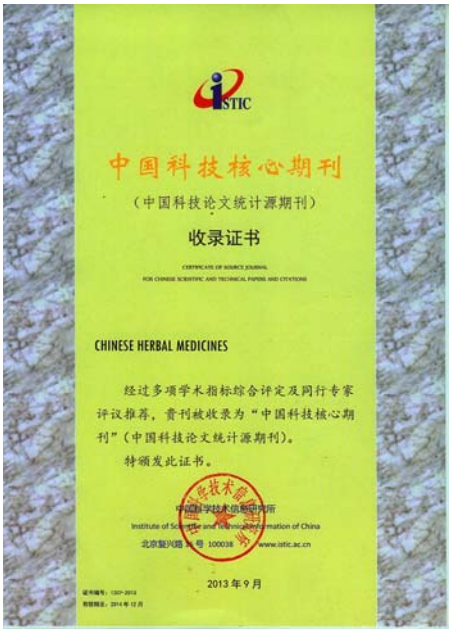

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