

Full Length Research Paper

Phenolic compounds and flavonoids from the fruits of *Pandanus tectorius* Soland

Xiaopo Zhang^{1,2}, Peng Guo¹, Guangli Sun^{1,3}, Shilin Chen¹, Meihua Yang^{1,4}, Naiguang Fu², Haifeng Wu¹ and Xudong Xu^{1*}

¹Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100193, China.

²Hainan Medical University, Haikou 570010, China.

³College of Pharmacy, Hebei United University, Hebei, 063000, China.

⁴Hainan-branch Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences and Peking Union Medical College, Wanning 571533, China.

Accepted 18 November, 2011

Fifteen (1-15) compounds including ten phenolic compounds and five flavonoids were isolated from the fruits of *Pandanus tectorius*. All of the compounds were isolated and purified by various column chromatographies especially by semi-preparative high performance liquid chromatography (HPLC). Their structures were determined under the aid of spectral methods. All compounds were isolated from this medicinal plant for the first time. The biological activities of some compounds were discussed according to the results of related literature.

Key words: *Pandanus tectorius* Soland, Pandanaceae, phenolic compounds, flavonoids, biological activities.

INTRODUCTION

The genus *Pandanus* of Pandanaceae family comprises of about 700 species distributed worldwide in the subtropical and tropical regions (Tan et al., 2008). Various *Pandanus* species are important commercial crops and also used as folk medicine as a treatment for leprosy, bronchitis, measles, dermatitis and urinary tract ailments (Vahirua-Lechat et al., 1996). In China, five species and one varied species belonging to the genus are spread in the subtropical and tropical regions including Hainan Province, Guangdong Province, Taiwan Province, etc. The extract of roots and stem of *Pandanus tectorius* possessing many medicinal effects in treating influenza, hepatitis, urinary tract infection, and nephritis have been recorded in standardized and benchmarked Chinese Materia Medica of Guangdong Province (Peng et al., 2010). Its fruits with the biological activity of anti-inflammatory attracted our interest. However, little information is available on the chemical constituents of the fruits of the medicinal plants. Based on these, we carried out the

studies on chemical principles of the fruits. During this course, ten phenolic compounds and five flavonoids were obtained, and the reported biological activities of some isolated compounds were summarized in this paper.

EXPERIMENTAL

General experimental procedures

The NMR data were recorded on Bruker AV 600 (600 MHz for ¹H and 150 MHz for ¹³C) in MeOD or CDCl₃ with TMS as internal standard. Chromatography was performed on silica gel column (200-300 mesh, Qingdao Haiyang) and Sephadex LH-20 column (GE Healthcare, Sweden). HPLC isolation was conducted on a lumtech K1001 analytic LC equipped with two pumps of K-501, a UV detector of K-2600, and a column of YMC-Pack ODS-A (250×10 mm, 5 μm).

Collection and preparation of plant material

The medicinal material was collected at Hainan Province in July 2011 and identified by Prof. Weiyong Lai at the School of Pharmaceutical Science, Hainan Medical University. A voucher specimen has been deposited there (Voucher specimen No. PT20110714).

*Corresponding author. E-mail: xdxu@implad.ac.cn. Tel: +86-10-57833296. Fax: +86-10-57833296.

Extraction and isolation

The fruits (20 kg) of *P. tectorius* were dried in shade and exhaustively extracted with 95% ethanol. The extract was filtered and concentrated on reduced pressure until only H₂O remained. The remaining solution was sequentially partitioned with petroleum ether (34 g), CHCl₃ (42 g), EtOAc (53 g) and n-BuOH (100 g). The CHCl₃ extract (42 g) was subjected to a column chromatography on silica gel using petroleum ether-EtOAc as the mobile phase and six fractions (Fr. 1-6) were obtained. Five fractions (Fr. 2-6) were purified by a column chromatography of Sephadex LH-20 eluting with CHCl₃-CH₃OH (1:1) and then the eluent was concentrated *in vacuo*, respectively. The obtained residues were further purified by semi-preparative HPLC using MeOH-H₂O as the eluent to yield compounds 1-15 (Figure 1).

RESULTS AND DISCUSSION

Compound 1, NMR data: ¹HNMR (600 MHz, MeOD) δ: 9.83 (1H, s, -CHO), 7.43 (1H, dd, *J* = 8.4, 1.8 Hz, H-5), 7.42 (1H, d, *J* = 1.8 Hz, H-2), 7.04 (1H, d, *J* = 8.4 Hz, H-5), 3.97 (-OMe). These spectral data showed basically agreement with the compound of vanillin (Xiang et al., 2011).

Compound 2, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.57 (1H, d, *J* = 15.6 Hz, H-7), 7.09 (1H, d, *J* = 1.8 Hz, H-2), 7.00 (1H, dd, *J* = 7.8, 1.8 Hz, H-5), 6.87 (1H, d, *J* = 7.8 Hz, H-6), 6.24 (1H, d, *J* = 15.6 Hz, H-8), 4.25 (2H, q, *J* = 6.6 Hz, -CH₂), 1.34 (3H, t, *J* = 6.6 Hz, -CH₃). These spectral data showed basically agreement with the reported literature of *trans*-ethyl caffeate (Xiang et al., 2009).

Compound 3, NMR data: δ: 7.32 (1H, d, *J* = 1.8 Hz, H-2), 7.02 (1H, dd, *J* = 8.4, 1.8 Hz, H-6), 6.77 (1H, d, *J* = 12.6 Hz, H-7), 6.72 (1H, d, *J* = 8.4 Hz, H-5), 5.73 (1H, d, *J* = 12.6 Hz, H-8), 4.16 (2H, q, *J* = 6.6 Hz, -CH₂), 1.26 (3H, t, *J* = 6.6 Hz, -CH₃). These spectral data showed basically agreement with the reported literature of *cis*-ethyl caffeate (Bunzel et al., 2005).

Compound 4, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.60 (1H, d, *J* = 16.2 Hz, H-7), 7.45 (2H, d, *J* = 8.4 Hz, H-2, 6), 6.80 (2H, d, *J* = 8.4 Hz, H-3, 5), 6.31 (1H, d, *J* = 16.2 Hz, H-8), 4.21 (2H, q, *J* = 6.6 Hz, -CH₂), 1.31 (3H, t, *J* = 6.6 Hz, -CH₃). These spectral data showed basically agreement with the reported literature of ethyl coumarate (Kaewamatawong et al., 2007).

Compound 5, NMR data: ¹HNMR (600 MHz, MeOD) δ: 9.58 (1H, d, *J* = 7.8 Hz, -CHO), 7.58 (1H, d, *J* = 15.0 Hz, H-7), 7.00 (2H, s, H-2, 6), 6.87 (1H, dd, *J* = 15.0, 7.8 Hz, H-8), 3.89 (6H, s, 2x-OCH₃). These spectral data showed basically agreement with the reported literature of sinapaldehyde (Wen et al., 1996).

Compound 6, NMR data: ¹HNMR (600 MHz, MeOD) δ: 9.59 (1H, d, *J* = 7.8 Hz, -CHO), 7.11 (1H, dd, *J* = 8.4, 1.8 Hz, H-6), 7.06 (1H, d, *J* = 1.8 Hz, H-6), 6.95 (1H, d, *J* =

8.4 Hz, H-6), 6.59 (1H, dd, *J* = 15.0, 7.8 Hz, H-8). These spectral data showed basically agreement with the reported literature of *trans*-3, 4-dihydroxycinnamaldehyde (Demin et al., 2004).

Compound 7, NMR data: ¹HNMR (600 MHz, MeOD) δ: 6.77 (1H, d, *J* = 1.8 Hz, H-2), 6.69 (1H, d, *J* = 8.4 Hz, H-5), 6.61 (1H, dd, *J* = 8.4, 1.8 Hz, H-6), 3.86 (3H, -OMe), 3.55 (2H, t, *J* = 6.6 Hz, H-9), 2.59 (2H, t, *J* = 6.6 Hz, H-7), 1.80 (2H, m, H-8). These spectral data showed basically agreement with the reported literature of dihydroconiferyl alcohol (Saracoğlu et al., 2002).

Compound 8, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.00 (1H, d, *J* = 2.4 Hz, H-2), 6.85 (1H, dd, *J* = 8.4, 1.8 Hz, H-6), 6.72 (1H, d, *J* = 8.4 Hz, H-5), 6.50 (1H, d, *J* = 15.6 Hz, H-7), 6.19 (1H, dt, *J* = 15.6, 6.0 Hz, H-8), 4.19 (2H, d, *J* = 6.0 Hz, H-9), 3.86 (3H, -OMe). These spectral data showed basically agreement with the reported literature of coniferyl alcohol (Kim et al., 2011).

Compound 9, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.58 (1H, dd, *J* = 8.4, 2.4 Hz, H-6), 7.54 (1H, d, *J* = 2.4 Hz, H-2), 6.86 (1H, d, *J* = 8.6 Hz, H-5), 3.94 (2H, t, *J* = 6.0 Hz, H-9), 3.16 (2H, t, *J* = 6.0 Hz, H-8). These spectral data showed basically agreement with the reported literature of 3-hydroxy-1-(4-hydroxy-3-methoxyphenyl)propan-1-one (Li et al., 2011).

Compound 10, NMR data: ¹HNMR (600 MHz, MeOD) δ: 9.89 (1H, s, -CHO), 8.15 (1H, d, *J* = 8.4 Hz, H-6), 8.10 (1H, s, -OH), 7.48 (1H, d, *J* = 8.4 Hz, H-2), 7.28 (1H, t, *J* = 8.4 Hz, H-3), 7.28 (1H, t, *J* = 8.4 Hz, H-4). These spectral data showed basically agreement with the reported literature of salicylaldehyde (Janes et al., 2008).

Compound 11, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.98 (2H, d, *J* = 8.4 Hz, H-2', 6'), 7.12 (2H, d, *J* = 8.4 Hz, H-3', 5'), 6.68 (1H, s, H-3), 4.11 (s, -OMe), 4.03 (s, -OMe), 3.93 (s, -OMe), 3.90 (s, -OMe), 3.89 (s, -OMe). These spectral data showed basically agreement with the reported literature of tangeretin (Hamdan et al., 2007).

Compound 12, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.58 (2H, d, *J* = 8.4 Hz, H-2', 6'), 6.82 (2H, d, *J* = 8.4 Hz, H-3', 5'), 6.05 (1H, d, *J* = 2.4 Hz, H-8), 6.04 (1H, d, *J* = 2.4 Hz, H-6), 5.38 (1H, dd, *J* = 13.2, 3.0 Hz, H-2), 3.94 (3H, s, -OMe), 3.15 (1H, dd, *J* = 16.8, 13.2 Hz, H-3a), 2.73 (1H, dd, *J* = 16.8, 3.0 Hz, H-3b). These spectral data showed basically agreement with the reported literature of sakuranetin (Freitas et al., 2008). **Compound 13, NMR data:** ¹HNMR (600 MHz, MeOD) δ: 8.00 (2H, d, *J* = 8.4 Hz, H-2', 6'), 7.58 (3H, m, H-3', 4', 5'), 6.50 (1H, d, *J* = 1.8 Hz, H-8), 6.24 (1H, d, *J* = 1.8 Hz, H-6). These spectral data showed basically agreement with the reported literature of chrysin (Subramanian et al., 1972).

Compound 14, NMR data: ¹HNMR (600 MHz, MeOD) δ: 7.30 (2H, d, *J* = 8.4 Hz, H-2', 6'), 6.82 (2H, d, *J* = 8.4 Hz,

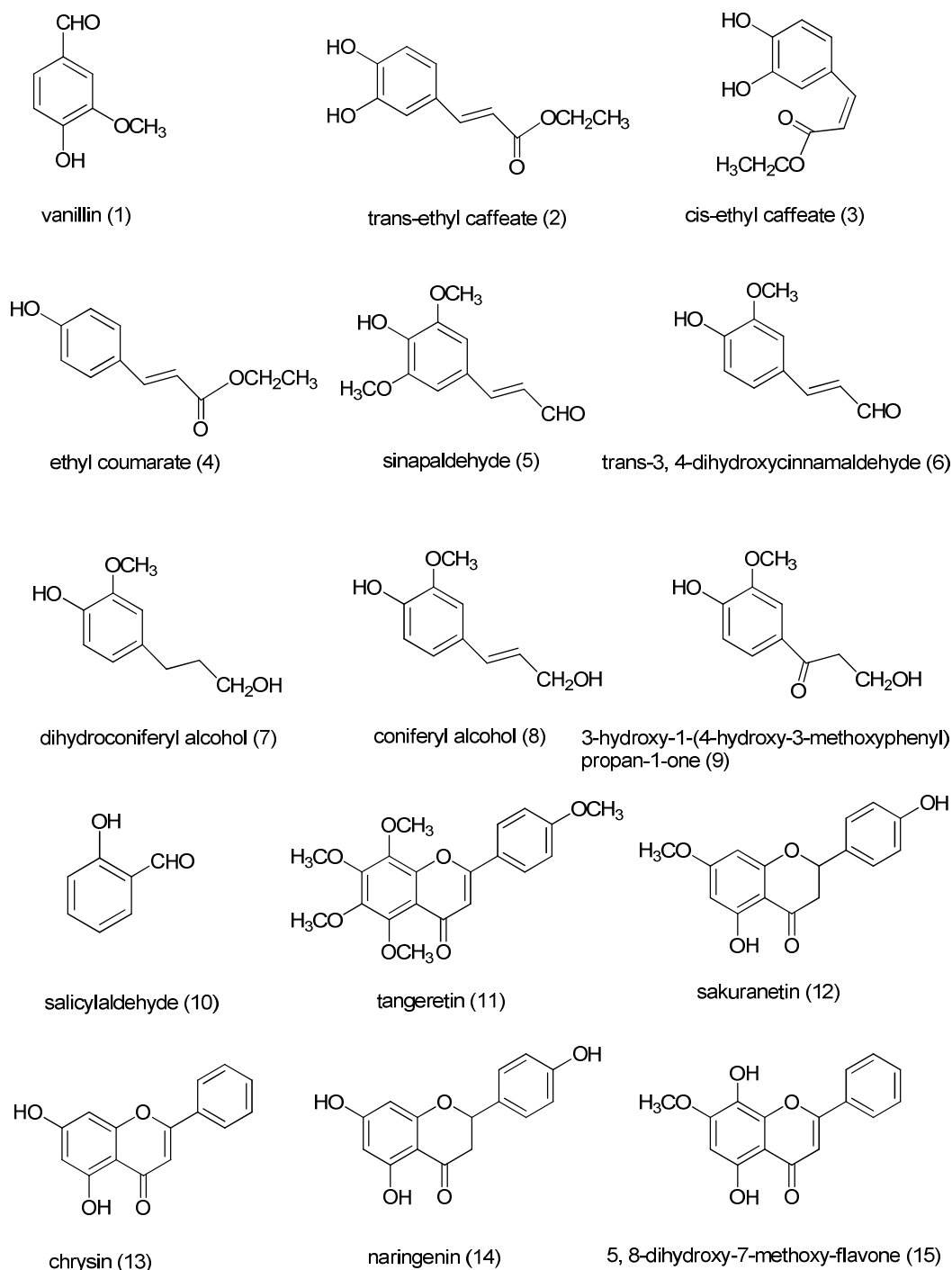


Figure 1. Structures of compounds 1–15.

H-3', 5'), 5.84 (1H, d, $J = 2.4$ Hz, H-8), 5.83 (1H, d, $J = 2.4$ Hz, H-6), 5.32 (1H, dd, $J = 13.2, 3.6$ Hz, H-2), 3.15 (1H, dd, $J = 17.4, 13.2$ Hz, H-3a), 2.73 (1H, dd, $J = 17.4, 3.6$ Hz, H-3b). These spectral data showed basically agreement with the reported literature of naringenin (Wilcox et al., 1999).

Compound 15, NMR data: ^1H NMR (600 MHz, CDCl_3) δ : 12.52 (1H, brs, 5-OH), 7.91 (2H, d, $J = 7.8$ Hz, H-2', 6'), 7.55 (3H, m, H-3', 4', 5'), 6.69 (1H, s, H-6), 6.45 (1H, s, H-3), 4.03 (3H, s, -OMe). These spectral data showed basically agreement with the reported literature of 5, 8-dihydroxy-7-methoxyflavone (Prabir et al., 1992).

Table 1. Reported biological activities of some isolated compounds.

Name of compounds	Activities (Reported)
Vanillin	Inhibition of tyrosinase (Gong et al., 2006); Anti-oxidant (Burri et al., 1989)
<i>trans</i> -ethyl caffeate	Anti-inflammatory (Chiang et al., 2005)
Tangeretin	Cholesterol-lowering activity (Kurowska et al., 2004); Anti-tumor (Chaumontet et al., 1997)
Chrysin	Aromatase inhibitor (Van Meeuwen et al., 2007); Apoptotic effects (Khoo et al., 2010)
Naringenin	Antiviral (Nahmias et al., 2008); Cholesterol-lowering activity (Lee et al., 1999)

Natural compounds obtained from the genus of *Pandanus* were mainly alkaloids, terpenoids, organic acids, and lignans according to the literature. Meanwhile, most studies have focused on the chemical constituents from the leaves and roots and few researches concerning the compositions of the fruits of genus *Pandanus* have been investigated. The 15 compounds were isolated from *P. tectorius* for the first time and have not been obtained from any other fruits belonging to genus *Pandanus*. Among these compounds, the content of *trans*-ethyl acetate was the highest one in the fruits with approximately 0.1% of the dry material.

Most of these compounds possessed anti-oxidative activities and some anti-inflammatory activities corresponding to the activity of the fruits of *P. tectorius*. Chiang et al. (2005) studied the anti-inflammatory functions and mechanisms of ethyl caffeate, and they found that ethyl caffeate markedly suppressed the LPS-induced nitric oxide production and exerted an inhibitory effect on COX-2 transcriptional activity. Chrysin has been shown to induce an anti-inflammatory effect, most likely by inhibition of COX-2 expression and via IL-6 signaling (Woo et al., 2005). The biological activities of some isolated compounds were listed in Table 1.

ACKNOWLEDGEMENTS

The authors gratefully acknowledged the financial support from Postdoctoral Foundation of Chinese Academy of Medical Sciences and Peking Union Medical College, the technological large platform for comprehensive research and development of new drugs in the Eleventh Five-Year "Significant New Drugs Created" Science and Technology Major Projects (No. 2009ZX09301-003), the Technological Large Platform for Comprehensive Research and Development of New Drugs in the Twelfth Five-Year "Significant New Drugs Created" Science and Technology Major Projects (No. 2012ZX09301-002-001-026), and national project of creating reference substance base from medicinal plants (No. 1310981201).

REFERENCES

Bunzel M, Ralph J, Steinhart H (2005). Association of non-starch polysaccharides and ferulic acid in grain amaranth (*Amaranthus caudatus* L.) dietary fiber. *Mol. Nutr. Food Res.*, 49(6): 551-559.

- Burri J, Graf M, Lambelet P, Löliger J (1989). Vanillin: More than a flavouring agent—a potent antioxidant. *J. Sci Food Agric.*, 48 (1): 49-56.
- Chaumontet C, Droumaguet C, Bex V, Heberden C, Gaillard-Sanchez I, Martel P (1997). Flavonoids (apigenin, tangeretin) counteract tumor promoter-induced inhibition of intercellular communication of rat liver epithelial cells. *Cancer Lett.*, 114(1-2): 207-210.
- Chiang YM, Lo CP, Chen YP, Wang SY, Yang NS, Kuo YH, Shyur LF (2005). Ethyl caffeate suppresses NF- κ B activation and its downstream inflammatory mediators, iNOS, COX-2, and PGE2 *in vitro* or in mouse skin. *Br. J. Pharmacol.*, 146(3): 352-363.
- Demin P, Rounova O, Grunberger T, Cimpean L, Sharfe N, Roifman CM (2004). Tyrenes: synthesis of new antiproliferative compounds with an extended conjugation. *Bioorg. Med. Chem.*, 12(11): 3019-3026.
- Freitas MO, Ponte F AF, Lima M AS, Silverira ER (2008). Flavonoids and triterpenes from the nest of stingless bee *Trigona spinipes*. *J. Braz. Che. Soc.*, 19(3): 532-535.
- Gong SC, Yang ZY, Chen J (2006). Inhibition of tyrosinase activity by vanillin. *J. South China Univ. Technol.*, 34: 53-57.
- Hamdan D, El-Readi MZ, Tahrani A, Herrmann F, Daufmann D, Farrag N, El-SHazly A, Kaewamatawong R, Ruangrunsi N, Likhitwitayawuid K (2007). Chemical constituents of *Polyalthia parviflora* stem. *J. Nat. Med.*, 61(3): 349-350.
- Janes D, Kreft S (2008). Salicylaldehyde is a characteristic aroma component of buckwheat groats. *Food Chem.*, 109(2): 293-298.
- Khoo BY, Chua SL, Balaram P (2010). Apoptotic effects of chrysin in human cancer cell lines. *Int. J. Mol. Sci.*, 11(5): 2188-2199.
- Kim JA, Son JH, Yang SY, Kim YH (2011). Isoconiferoside, a new phenolic glucoside from seeds of *Panax genseg*. *Molecules*, 16(8): 6577-6581.
- Kurowska EM, Manthey JA (2004). Hypolipidemic effects and absorption of citrus polymethoxylated flavones in hamsters with diet-induced hypercholesterolemia. *J. Agric. Food Chem.*, 52(10): 2879-2886.
- Lee SH, Park YB, Bae KH, Bok SH, Kwon YK, Lee ES, Choi MS (1999). Cholesterol-lowering activity of naringenin via inhibition of 3-hydroxy-3-methylglutaryl coenzyme A reductase and acyl coenzyme A: Cholesterol acyltransferase in rats. *Ann. Nutr. Metab.*, 43(3): 173-180.
- Li L, Seeram NP (2011). Further investigation into maple syrup yields 3 new lignans, a new phenylpropanoid, and 26 other phytochemicals. *J. Agric. Food Chem.*, 59(14): 7708-7716.
- Nahmias Y, Goldwasser J, Casali M (2008). Apolipoprotein B-dependent hepatitis C virus secretion is inhibited by the grapefruit flavonoid naringenin. *Hepatology*, 47(5): 1437-1445.
- Peng LH, Cheng JL, Zhan RT, Li JR (2010). Research progress on the chemical constituents and biological activities of genus *Pandanus*. *Chin Med. Mater.*, 33, 640-643.
- Prabir KG, Anjan B (1992). 5, 8-dihydroxy-7-methoxyflavone from the immature leaves of *didymocarpus Pedicellata*. *Phytochemistry*, 31(5): 1833-183
- Saracoğlu I, Harput ÜŞ, Caliş I (2002). Phenolic constituents from *Phlomis lycia*. *Turk. J. Chem.*, 26(1): 133-142.
- Subramanian SS, Nair AGR (1972). Flavonoids of the stem bark of *Oroxylum indicum*. *Curr. Sci.*, 41(2): 62-63.
- Tan MA, Takayama H, Aimi N, Kitajima M, Franzblau SG, Nonato MG (2008). Antitubercular triterpenes and phytosterols from *Pandanus tectorius* Scoland. var. *laevis*. *J. Nat. Med.*, 62: 232-235.
- Vahirua-Lechat I, Menut C, Roig B, Bessiere JM, Lamaty G (1996).

- Isoprene related esters, significant components of *Pandanus tectorius*. *Phytochem.*, 43(6): 1277-1279.
- Van Meeuwen JA, Korthagen N, de Jong PC, Piersma AH, van den Berg M (2007). Anti-estrogenic effects of phytochemicals on human primary mammary fibroblasts, MCF-7 cells and their co-culture. *Toxicol. Appl. Pharmacol.*, 221(3): 372-383.
- Wen DX, Chen ZL (1996). A dimeric sinapaldehyde glucoside from *Ilex rotunda*. *Phytochemistry*, 41(2): 657-659.
- Wilcox LJ, Borradaile NM, Huff MW (1999). Antiatherogenic properties of naringenin, a citrus flavonoid. *Cardiovasc. Drug Rev.*, 17(2): 160-178.
- Woo KJ, Jeong YJ, Inoue H, Park JW, Kwon TK (2005). Chrysin suppresses lipopolysaccharide-induced cyclooxygenase-2 expression through the inhibition of nuclear factor for IL-6 (NF-IL6) DNA-binding activity. *FEBS Lett.*, 579: 705-711.
- Xiang MX, Su HW, Hu JY, Yan YJ (2011). Isolation, identification and determination of methyl caffeate, ethyl caffeate and other phenolic compounds from *Polygonum amplexicaule* var. *sinense*. *J. Med. Plants Res.*, 5(9): 1685-1691.