ORIGINAL ARTICLE

TWO TRITERPENES LUPANONE AND LUPEOL ISOLATED AND IDENTIFIED FROM *TAMARINDUS INDICA* LINN.

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ABSTRACT

Tamarindus indica, a useful medicinal plant was subjected to phytochemical investigation. Two triterpenes (lupanone and lupeol) have been isolated from the leaves of this plant. Their structures were elucidated with the help of physico-chemical methods and spectroscopic techniques. The lupanone and lupeol from this plant are being reported for the first time.

Keywords: *Tamarindus indica*, caesalpiniaceae, triterpenes.

INTRODUCTION

Tamarindus is a monotypic genus distributed throughout much of the tropics. It belongs to the family Caesalpiniaceae. Different parts of the plant such as leaves, fruit and seeds have been extensively used in traditional Indian and African medicines (Gunasema and Hughas, 2000). Aqueous extract of seed reduced blood sugar level (Maiti *et al*., 2004), showed hypolipidemic effect, reduced 14-17% of plasma lipid, total lipid, cholesterol, lipoprotein and triglycerides (Yamatoya *et al*., 1966,1998). The seed coat extract has strong anti-oxidant property, used as additive to food, cosmetic-pharmaceutical preparations (Pauly, 1997). The seeds also inhibit the growth of urinary crystals and are used in the treatment of recurrent kidney stones in patients (Natarajan *et al*., 1997). The fruit also has antimicrobial and antibiotic activity (Ross *et al*., 1980).

The plant has a great phytochemical significance. On literature survey it was revealed that a variety of secondary metabolites have been reported from tamarind. The leaf oil contains thirteen components among which linonene (24.4%) and benzyl benzoate (40.6%) were most predominant (Pino *et al*., 2002). The volatile constituents of the fruit pulp were furan derivatives (44.4%) and carboxylic acid (33.3%) of the total volatiles (Wong *et al*., 1998). The major fatty acids of seeds were palmitic acid, oleic acid, linoleic acid and eicosanoic acid. Seven hydrocarbons, β-amyrin, compesterol and β-sitosterol were found in the unsaponifiable matter of seeds. The mucilage and pectin, arabinose, xylose, galactose, glucose and uronic acid were also identified (Ibrahim and Abbas, 1995).

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A new bufadienolide (Scilliphraside 3-O-β-D-glucopyranosyl-(1-2)-L-rhamnopyranoside) and cardeno-lide (uzarigenin-3-O-β-D-xylopyranosyl (1-2)-α-L-rhamnopyranoside) were identified from the seed extract (Yadara *et al*., 1999a, 1999b). The fruit pulp is the richest source of tartaric acid (8-18%) and seeds are rich of protein and valuable amino acid (Shankaryacharya, 1998). Some glycosides and other constituents like ritexin, isoritexin, orientin and iso-orientin were isolated from the leaves of tamarind (Bhatia *et al*., 1966).

No study was made on terpenic constituents of this plant. Therefore, an attempt was made to investigate the terpenes present in the leaves of tamarind.

MATERIAL AND METHODS

Extraction and isolation

The leaves of *T. indica* were collected during the month of April in 2005 from Botanical Garden, University of Karachi. The leaves were soaked in methanol for a period of one month. The extract was evaporated under reduced pressure and a gummy residue was obtained. The extract was first partitioned between hexane and water, then chloroform and water and finally between ethylacetate and water. The chloroform soluble part was concentrated and subjected to vacuum liquid chromatography. Elution was carried out using 5%, 10% and 15% chloroform in hexane as mobile phase.

The fraction eluted with 15% chloroform from vacuum liquid chromatography (VLC) was subjected to column chromatography using the various combination of hexane and chloroform mixture as mobile phase. The fraction eluted with 5% chloroform in hexane gave compound 1 and 20% chloroform in hexane gave compound 2 in pure form.

Characterization of Compound 1

IR: ($CHCl_3$) ν_{max}: 3080, 1700 (C=O), 1648 (C=C) and 890 cm^{-1}.

EIMS: m/z 424 (M^+, $C_{30}H_{48}O$), 409, 313, 218, 205 (100), 189, 161, 149 and 135.

^{1}HNMR ($CDCl_3$, 300 MHz): 84.66-4.54 and 2.40 (each 2H, m, H-29 and 2), 1.65, 1.53, 1.04, 0.99, 0.92, 0.89 and 0.76 (each 3H, δ, 7x CH_3).

^{13}C-NMR ($CDCl_3$ 125 MHz): δ 217.9 (C-3), 150.7 (C-20), 109.2 (C-29), 55.0 (C-5), 49.8 (C-9), 48.3 (C-18), 47.9 (C-19), 47.3 (C-4), 42.9 (C-17, 14), 40.9 (C-8), 40.0 (C-22), 39.6 (C-1), 38.2 (C-13), 36.9 (C-20), 35.6 (C-16), 34.1 (C-2), 33.6 (C-7), 29.9 (C-21), 27.4 (C-15), 26.2 (C-23), 25.2 (C-12), 21.5 (C-11), 21.0 (C-24), 19.6 (C-6), 19.3 (C-30), 18.0 (C-28), 15.9 (C-26), 15.8 (C-25) and 14.4 (C-27).

Characterization of compound 2

The isolated pure compound was characterized and identified by spectral analysis.

$[\alpha]_D$: +26.0° (C=0.80, $CHCl_3$)

IR: (KBr) ν_{max}:3235, 1640, 1490, 1382, 1185, 1105, 1040, 984 and 943 cm^{-1}.

EIMS m/z 426 (M^+,$C_{30}H_{50}O$), 411 (4.45%, M^+-CH_3), 296 (9%), 278 (8.9%), 218 (39%), 207 (28%), 203 (26%), 189 (27%), 175 (10%), 135 (27%), 95 (100%).

^{1}H-NMR ($CDCl_3$), 400 MHz): δ4.69 and 4.56 (each 1H, m, H-29), 3.18 (1H, dd, H-3), 2.39 and 1.93 (each 1H, m, H-19, 21A), 1.71 (1H, t, H-15A), 1.69 (3H, s, H-30), 1.68 (2H, d, H-12A, 1A), 1.67 (1H, t, H-13), 1.61 (1H, d, H-2A), 1.54 (1H, q, H2B), 1.54, 1.49 and 1.42 (each 1H, d, H-64, 16A, 11A), 1.42 (1H, m, H-22A), 1.41 (2H, m, H-7), 1.39 (1H, q, H-6B), 1.38 (1H, t, H-16A), 1.37 (1H, t, H-18), 1.33 (1H, m, H-21B), 1.28 (1H, d, H-9), 1.29 (1H, q, H-11B), 1.20 (1H, m, H-22B), 1.07 (1H, q, H-12A), 1.04 (3H, s, H-23), 1.01 (1H, d, H-15A), 0.98 (3H, s, H-23), 0.97 (3H, s, H-27), 0.91 (1H, t, H-18), 0.27, 0.84, 0.79 (each 3H, s, H-25, 28, 24) and 0.69 (1H, d, H-5).

^{13}CNMR ($CDCl_3$, 100 MHz): δ 150.8 (C-20), 109.3 (C-29), 78.9 (C-3), 55.2 (C-5), 50.3 (C-9), 48.2 (C-18), 47.9 (C-19), 42.9 (C-17), 42.7 (C-14), 40.7 (C-8), 39.9 (C-22), 38.8 (C-4), 38.6 (C-1), 38.0 (C-13), 37.1 (C-10), 35.5 (C-16), 34.2 (C-7), 29.8 (C-21), 27.9 (C-23), 27.4 (C-15), 27.3 (C-2), 25.0 (C-12), 20.9 (C-11), 19.2 (C-30), 18.2 (C-6), 17.9 (C-28), 16.1 (C-25), 15.9 (C-26), 15.3 (C-24) and 14.5 (C-27).

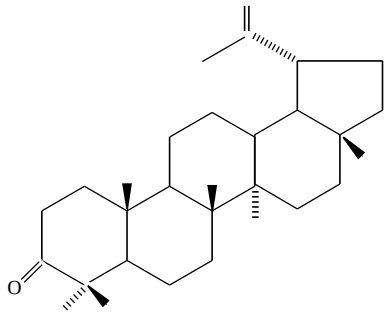
RESULTS AND DISCUSSION

The phytochemical examination of the methanolic extract of the leaves of *Tamarindus indica* afforded two triterpenes i.e., lupanone and lupeol. Both compounds (metabolites) have been isolated for the first time from this plant.

Compound 1

The compound 1 was obtained from the methanolic extract of the leaves of Tamarind. The crude extract was first semi-separated by VLC. As a result of this, the fraction eluted with 15% chloroform in hexane was subjected to column chromatography with 5% chloroform in hexane gave compound 1 as a white powder.

The infra-red spectrum of compound **1** showed the presence of a carbonyl function and an olefinic moiety which showed their presence in the spectrum at 1700 and 1648 cm^{-1}, respectively. The EI spectrum of the same compound gave an idea that compound 1 may have m/z 424 a.m.u. while the molecular weight which was confirmed by FDMS (Filled Desorption Mass Spectrometry), consequently the formula was deduced by HRMS (High Resolution Mass Spectrometry), as $C_{30}H_{48}O$.



(1)

The molecular formula depicted the presence of seven degrees of unsaturation, out of them two can be identified as (satisfied by) carbonyl and olefinic functions.

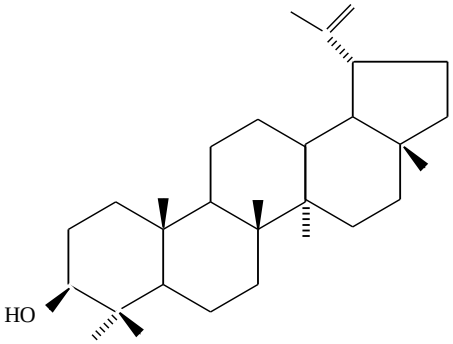
The presence of seven methyl singlets and olefinic function in the H-NMR spectrum revealed that compound **1** may be pentacyclic triterpenoidal type in nature. The comparison of H-NMR chemical shifts with that of the reported data of similar type of compounds has led to the conclusion that compound **1** is lupanone which was further confirmed by carbon spectrum having thirty carbon signals (Prajapati *et al.*, 2003; Shang-Jiang *et al.*, 1986 and Rofi and Pomilio, 1987). This is a very common compound/metabolite found in higher plants.

Compound 2

The compound 2 has been isolated from the same column and source. The fraction obtained from column

chromatography with 20% chloroform in hexane gave compound 2 as a white powder.

The infra-red spectrum of this compound showed the presence of a hydroxyl function and an olefinic moiety which showed their presence in the spectrum at 3235 and 1640 cm^{-1}, respectively. The EI spectrum of the same compound gave an idea that compound 2 may have m/z 426 a.m.u. while the molecular weight which was confirmed by FDMs, consequently the formula was deduced by HRMs as $C_{30}H_{50}O$. The molecular formula depicted the presence of six degrees of unsaturation, out of them one can be satisfied by an olefinic function.



(2)

The presence of seven methyl singlets and an olefinic function in the H-NMR spectrum revealed that **2** may be pentacylic tri-terpenoidal type in nature. The comparison of H-NMR chemical shifts with that of the reported data similar type of compounds and lupeol has led to the conclusion that compound **2** is lupeol which was further confirmed by carbon spectrum having thirty carbon signals (Prajapati *et al*., 2003; Shang-Jiang *et al*., 1986 and Pomilio, 1987). This is also a very common chemical compound found in higher plants.

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