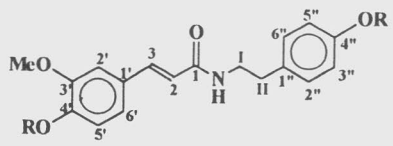
Indian Journal of Chemistry
Vol.36B, September 1997, pp.837 - 839

N-trans-Feruloyltyramine from *Tinospora cordifolia*

V D Gangan*[a], P Pradhan[b], A T Sipahimalani[b], V G Bhave[c] & A Patil[c]
[a]Hindustan Ciba Geigy Research Centre, Chemical Development, Aarey Road, Goregaon(E), Bombay 400 063, India
[b]Bio-Organic Division, Bhabha Atomic Research Centre, Bombay 400 085, India
[c]Dept. of Chemistry, Bhavan's College, Andheri(W), Bombay 400 058, India
Received 11 February 1997; accepted 12 June 1997

N-trans-Feruloyltyramine **1** has been isolated as diacetate from the *n*-BuOH fraction of the methanolic extract of *T. cordifolia* stems. Its structure has been elucidated by extensive 1D and 2D NMR studies.

Tinospora cordifolia Miers (Menispermaceae), an important medicinal plant cultivated throughout Indian subcontinent, has been used in Ayurvedic preparations for treatment of various ailments[1,2]. More recently, Thatte and Dahanukar[3] have shown that the aqueous extracts of the plant were associated with the stimulation of phagocytic and bactericidal capacity of neutrophils and macrophages. Chemical investigations of the plant have indicated the presence of several diterpene furan lactones[4], phenolic lignan[5], etc. Our previous studies, with the polar fraction of the plant, dealt with the isolation and characterisation of two phenylpropane glycosides[6], five norditerpene furan glycosides[7,8] and two diterpene furan glycosides[9]. This communication describes the isolation of *N- trans*-feruloyltyramine **1**. It was obtained as the minor component and was characterised by high resolution 1D and 2D NMR spectroscopy as well as by comparison of physical and spectral data with that of authentic sample. Although known from *Tinospora tuberculata*[10], this constitutes the first report about its occurrence in *Tinospora cordifolia*. In the previous reports[10], no attempts were made to assign the attachment of protons to appropriate carbons, and also some of the ^{13}C chemical shifts assigned to feruloic acid ring were not proper. Therefore, 2D NMR techniques were applied extensively to assign



1; R = H
1a; R = Ac

the positions of all protons and carbons unequivocally which is depicted in Table I.

Following the procedure mentioned in our earlier publication[7], compound **1** was isolated as its diacetate **1a** from the butanol extract fraction of the methanolic extract of the stem of *T. cordifolia*.

Compound **1a**, m.p. 155°, was a colourless solid and showed bluish fluorescence under long UV after spraying with $SbCl_3$ ($CHCl_3$) and heating. The IR spectrum exhibited several strong absorptions at 1700-1770 and 1190-1263 cm^{-1}, indicating the presence of acetoxy carbonyls and α,β-unsaturated ketonic carbonyl. The medium intensity bands at 2936, 1509-1609 and around 1653-1671 cm^{-1} were characteristic of aromatic ring and olefinic bond, respectively. The presence of > CONH group was inferred from the presence of bands at 3384 (br), 3256, 1767 (sh) and 1540 cm^{-1}. The UV spectrum showed bands at 276 nm (ε 3.33) and 315.8 nm (ε 3.12) indicating the presence of α,β-unsaturated carbonyl system with extensive conjugation. The FAB MS $[Na]^+$ of compound **1a** showed a molecular ion at 420 $[M+Na]^+$, thus indicating a molecular weight of 397 $[M]^+$. The ^1H NMR spectrum displayed seven aromatic protons (δ_H 7.04-7.30) and a methoxyl group (δ_H 3.85) substituted to one of the aromatic rings. A pair of *trans*- coupled doublets at δ_H 6.27 and 7.59 with J = 16 Hz is typical of a α,β conjugated carbonyl system. In addition, an amide (>CONH) triplet, one methylene triplet and one methylene doublet of a triplet were observed. These data indicated the compound **1a** to be identical with *N-trans*-feruloyltyramine isolated by Fukuda *et al.*[10] from *Tinospora tuberculata*. The unambiguous assignments, of various C and H positions were achieved on the

Table I—1D and 2D NMR data of *N-trans*-feruloyltyramine diacetate **1a**

	δ_C	δ_C	DEPT	δ_H*	COSY	NOESY	COLOC
	Rev.	Rep		Feruloyl			
1	165.6	165.5	> C=O				
2	120.8	139.5	> CH-	6.27(d,16.0)	H-3	H-6é,H-2′,NH	
3	140.4	111.1	> CH-	7.59(d,16.0)	H-2		
1′	133.7	148.8	> C <				H-5″
2′	111.2	121.0	> CH-	7.08(br s)	-OCH_3	-OCH_3,H-2	
3′	151.4	133.6	> C <				H-5′,-OCH_3
4′	141.0	140.4	> C <				H-2′
5′	122.8	120.1	> CH-	7.04(br s)	H-6′		
6′	121.8	122.8	> CH-	7.12(br s)	H-5′	H-2	
-OCH_3	55.8	55.7	–CH_3	3.85(s)	H-2′	H-2′	
> NH				5.71(t,5.8)	H-1	H-2	
				Tyramine			
I	40.6	40.7	-CH_2-	3.67(dt,5.8,7.7)	H-II, > NH	H-II	
II	34.9	34.8	-CH_2-	2.89(t,7.7)	H-I	H-I,H-2″,6″	
1″	136.4	136.3	> C <				H-I,H-II
							H-3″,5″
2″,6″	129.4	129.4	2 x > CH-	7.25(d,8.5)	H-3″,5″	H-II,H-3″,5″	H-II
3″,5″	121.3	121.3	2 x > CH-	7.06(br s)	H-2″,6″	H-2″,6″	
4″	149.3	150.9	> C <				H-2″,6″
				Acetoxy			
2 x O*CO*CH_3	20.6	20.5					
	21.1	21.0	2 x -CH_3	2.32(br s)			
2 x O*CO*CH_3	168.8	168.4					
	169.8	169.3					

*Proton assignments are based on ^{13}C-^1H HECTOR and ^1H COSY experiments.

basis of 2D experiments involving ^{13}C-^1H COLOC, ^1H-^1HCOSY and ^1H-^1H NOESY experiments.

The ^1H NMR signal at δ_H 5.71 was assigned to > NH proton as it had no corresponding carbon in HETCOR spectrum. This proton showed COSY interaction with a methylene at δ_H 3.67 which in turn had interaction with the second methylene at δ_H 2.89. On the basis of COSY, the signals at δ_H 7.25 (H-2″,6″) and 7.06 (H-3″,5″) were found to constitute an A_2B_2 pair and therefore these protons belong to the para-substituted aromatic ring. The rest three aromatic protons belong to the other ring with 1, 3, 4-substitution pattern. The *ortho*-pair appeared at δ_H 7.04 (H-5′) and 7.12 (H-6′), thus assigning the remaining proton to H-2′(δ_H 7.08). The corresponding carbons were assigned accordingly on the basis of $^1J_{CH}$ in HETCOR (Table 1).

The methoxyl resonance showed COLOC interaction with a carbon at δ_C 151.4 and was assigned to C-3′. The former showed nOe with H-2′ whereas the latter carbon showed COLOC interaction with H-5′, thus confirming their assignments. Similarly, peaks at δ_C 133.7 and 141.0 were assigned to C-1′ and C-4′ on account of the COLOC interactions with protons at H-5′ and H-2′ respectively. In the second aromatic (tyramine) ring, the peak at δ_C 136.4 showed COLOC interactions with two methylene protons at δ_H 3.67, 2.89 as well as with H-3″, 5″ (δ_H 7.06), whereas the other carbon at δ_C 149.3 was involved in the COLOC interaction with H-2″, 6″ (δ_H 7.25). This completes the unequivocal assignments of the carbons.

As described previously, the olefinic bond had a *trans* (*E*)-configuration on the basis of *J* value ($J_{2,3}$ = 16 Hz). H-2 showed nOe with H-2′ and H-6′ protons. This confirmed that H-2 is *cis*-disposed to the aromatic ring and the aromatic ring is probably perpendicular to the plane passing through C-1, C-2 and C-3.

Experimental Section

General. As per reference No.7.

Isolation N-*trans*-feruloyltyramine diacetate 1a. The plant material[11] (5.8kg) was collected from Trombay campus, Bhabha Atomic Research Centre, Bombay. Fresh stems were subjected to cold extraction with MeOH through percolation. The non-polar compounds were removed by extraction with petrol and EtOAc. It was then extracted with *n*-BuOH and subjected to column chromatography over silica gel, using increasing polarities of MeOH in $CHCl_3$. The fractions, with similar polarities, were pooled together (five fractions). The crude fractions 2 and 3 were acetylated using Ac_2O and pyridine at room temperature for 48 hr. Further fractionation was achieved by repeated radial chromatography of the acetylated crude fractions as well as by preparative TLC. This resulted in the isolation of one more compound, in addition to previously reported cordifolisides A, B, C[7], cordifolisides D, E[8] and palmatosides C, F[9]. It was named as *N-trans*-feruloyltyramine **1** and was characterised as *N- trans*-feruloyl tyramine diacetate (**1a**).

Chromatographic system

Thin layer chromatography: TLC plates were prepared using silica gel G (Acme, Bombay), $CHCl_3$-MeOH (99:1) was used as the solvent system. Detection was done by spraying with 20% $SbCl_3$ ($CHCl_3$) and heating (100′, 10 min, bluish fluorescence under long UV). For *radial chromatography*, circular glass plates (thickness 1mm) were prepared by using silica gel (PF_{254}, E. Merck, 50 g) in cold distilled water (105 mL), and gradually increasing concentrations of MeOH in $CHCl_3$ were employed for elution.

***N-trans*-Feruloyltyramine diacetate (1a)** Colourless solid (7 mg), R_f (TLC, 0.57, $CHCl_3$:MeOH, 99:1); $C_{22}H_{23}NO_6$; m.p. 155′; IR:3384, 3256, 2936, 2861, 1767, 1670, 1653, 1609, 1540, 1509, 1368, 1264, 1190, 1161, 1117 cm^{-1}, UV 276 nm (ε 2177), 315.8 nm (ε 1344); for ^1H NMR (200 MHz, $CDCl_3$) and ^{13}C NMR (50 MHz, $CDCl_3$), see Table I; FAB MS: m/z 420 $[M+Na]^+$, 398 $[M+H]^+$, 397 $[M]^+$, 355, 251, 235, 219, 207, 193, 181, 177, 121, 77. Anal. Found C, 66.53; H, 5.83; N, 3.49 Calcd. for $C_{22}H_{23}NO_6$: C, 66.49, H, 5.79; N, 3.52 %.

Acknowledgement

We thank Dr O Seligmann and Prof. Dr H Wagner of the Institute of Pharmaceutical Biology, University of Munchen, Germany for recording the FABMS. One of the authors (V D G) thanks the Department of Atomic Energy for the award of a senior research fellowship.

References

1. Kirtikar K R & Basu B D in:*Indian medicinal plants*, Vol.I, edited by E Blatter, J R Cauis and K S Mhaskar (Lalit Mohan Basu, Allahabad, India) **1933**, p.77.
2. Nayampalli S, Ainapore S S & Nadkarni P M, *Indian J Pharmacol*, 14, **1982**, 64.
3. Thatte U M & Dahanukar S A, *Phytotherapy Res*, 3, **1989**, 43.
4. Bhatt R K & Sabata B K, *Phytochemistry*, 28, **1989**, 2419.
5. Hanuman J B, Mishra A K & Sabata B K, *J Chem Soc, Perkin Trans*, 1, **1986**, 1181.
6. Sipahimalani A T, Norr H & Wagner H, *Planta Medica*, 60, **1994**, 596.
7. Gangan V D, Pradhan P, Sipahimalani A T & Banerji A, *Phytochemistry*, 37, **1994**, 781.
8. Gangan V D, Pradhan P, Sipahimalani A T & Banerji A, *Phytochemistry*, 39, **1995**, 1139.
9. Gangan V D, Pradhan P, Sipahimalani A T & Banerji A, *Indian J Chem*, 35B, **1996**, 630.
10. Fukuda N, Yonemitsu M & Kimura T, *Chem. Pharm. Bull*, 31, **1983**, 156 and the references cited therein.
11. Identified by Dr V Abraham of Nuclear Agriculture Division, BARC, Bombay.