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N^b-benzoyltryptamine derivatives with relaxant activity in guinea-pig ileum

Stanley Juan C. Gutierrez [a], Fladmir de S. Claudino [a], Bagnólia A. Da Silva [a], Celso A. Câmara [a], Reinaldo N. de Almeida [a], Maria de Fátima V. de Souza [a], Marcelo S. Da Silva [a], Emídio V.L. Da-Cunha [a,b], José Maria Barbosa-Filho [a,*]

[a] *Laboratório de Tecnologia Farmacêutica, Universidade Federal da Paraíba, Cx. Postal 5009, 58051-970 João Pessoa, Paraíba, Brazil*
[b] *Departamento de Farmácia e Biologia, Universidade Estadual da Paraíba, CCBS, 58100-000 Campina Grande, Paraíba, Brazil*

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Abstract

A series of derivatives analogous to N^b-benzoyltryptamine were synthesized by the Schotten–Bauman procedure. The products obtained were: N^b-4-methoxy-benzoyltryptamine, N^b-2,4-dimethoxy-benzoyltryptamine, N^b-3,4-dimethoxy-benzoyltryptamine, N^b-3,4-methylenedioxy-benzoyltryptamine and N^b-3,4,5-trimethoxy-benzoyltryptamine. They were characterized through the usual spectrometric methods (UV, IR, ^{1}H and ^{13}C NMR) and showed non-selective relaxant activity in guinea-pig ileum pre-contracted with acetylcholine, histamine and KCl.

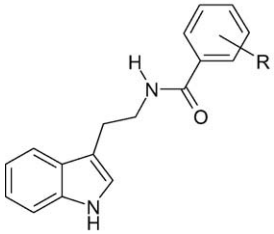
Keywords: Indole alkaloids; Tryptamine; Spasmolytic activity

1. Introduction

Because of their biological importance, and potentiality as key intermediates in the synthesis of more complicated indole alkaloids, the synthesis of indoles bearing an aminoalkyl or carboxyaryl group has attracted our interest, with the indole nucleus of tryptamine consisting of an important pharmacophore itself. Recently we have been engaged in the synthesis and anticonvulsant activity of a structurally related N^b-2-hydroxy-benzoyltryptamine (**1**) derivative [1]. The pharmacological activity found with this compound prompted us to perform the synthesis of some amide analogs (**2**) focusing on the benzoyl portion of the molecule by the suitable Schotten–Bauman procedure with available acid chlorides and tryptamine was performed (Fig. 1). These substances are structurally related to the parent compound of the series, N^b-benzoyltryptamine (*N*-[2-(1*H*-Indol-3-yl)ethyl]benzamide), that was previously isolated as a natural product from *Myrtopsis myrtoidea*, a plant of the Rutaceae family [2]. A preliminary study concerning the structure–activity relationships in these compounds was also performed.

2. Chemistry

For the synthesis of the substances with general structure (**2a–e**), Schotten–Baumann's technique was used, for the condensation of the respective benzoyl chlorides with tryptamine in a single reaction, using triethylamine as base. Following this simple methodology, it was possible to synthesize the five amide derivatives (**2a–e**) with reasonable yields. The compounds are, namely, N^b-4-methoxy-benzoyltryptamine (**2a**), N^b-2,4-dimethoxy-benzoyltryptamine (**2b**), N^b-3,4-dimethoxy-benzoyltryptamine (**2c**), N^b-3,4-methylenedioxy-benzoyltryptamine (**2d**) and N^b-3,4,5-trimethoxy-benzoyltryptamine (**2e**), and were obtained in good to reasonable yields of 53–80% in a straightforward methodology (Fig. 1).



Compound	R	Yield (%)
1	2-OH	Ref 1
2a	4-OMe	75
2b	2,4-OMe	65
2c	3,4-OMe	63
2d	3,4-OCH_2O	53
2e	3,4,5-OMe	80

Fig. 1.

* Corresponding author. Tel.: +55 83 216 7364; fax: +55 83 216 7365.
E-mail address: jbarbosa@ltf.ufpb.br (J.M. Barbosa-Filho).

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The N^b-3,4-methylenedioxy-benzoyltryptamine is a new compound.

3. Experimental procedures

3.1. Chemistry

Melting points are uncorrected and were determined on an electrically heated metal block apparatus. UV spectra were recorded on a Vankel 50 UV–Vis spectrophotometer in MeOH as solvent. IR spectra were obtained on a Bomen Michelson spectrophotometer using KBr. ^{1}H and ^{13}C NMR spectra were recorded in C_5D_5N on a Varian-Mercury 200 spectrometer at 200 MHz for ^{1}H NMR and 50 MHz for ^{13}C NMR and 2D techniques. For the analytical TLC, precoated Merck silica gel PF_{254} plates were used with a 0.25 mm layer. For column chromatography, silica gel-60 Merck was used. The tryptamine and benzoyl chlorides were purchased from Sigma-Aldrich and used as received.

3.1.1. N^b-4-methoxy-benzoyltryptamine (**2a**)

To a solution of triethylamine (1 ml) and chloroform (20 ml), tryptamine was added until total solubilization. To this solution a solution of *p*-anisoyl chloride (1 g) dissolved in chloroform (10 ml) was added dropwise over 15 min with magnetic stirring. The reaction mixture was left at room temperature for 2 h. The solution was then washed with first 2% HCl and then H_2O, dried over anhydrous Na_2SO_4, filtered and concentrated under vacuum, yielding a white powder. This product was chromatographed on a silica gel column, eluted with $CHCl_3$/hexane (1:1 and 8:2), $CHCl_3$ and $CHCl_3$/MeOH (98:02). After TLC analysis, fractions 10–21 were reunited and crystallized from $CHCl_3$/MeOH (1:1). The resulting material (**2a**, 75% yield) was obtained as white crystals with melting point of 134 °C (Lit. 132.5–133.5 °C) [3]; IR ν_{max} (KBr) cm^{-1}: 1179, 1257, 1437, 1455, 1507, 1607, 1615, 2847, 2937, 2977, 3011; UV λ_{max} MeOH nm: 235, 247, 256; ^{1}H NMR (200 MHz, C_5D_5N) δ: 3.36 (1H, s), 3.64 (3H, s), 4.07 (2H, q, J = 8.0), 7.01 (1H, d, J = 8.8), 7.25 (1H, dt, J = 1.2; 7.0), 7.29 (1H, dt, J = 1.4; 7.0), 7.35 (1H, d, J = 2.4), 7.60 (1H, d, J = 7.2), 7.89 (1H, d, J = 8.0), 8.31 (1H, d, J = 8.8), 9.09 (1H, t, J = 6.0), 11.8 (1H, s); ^{13}C NMR (50 MHz, C_5D_5N) δ: 26.4, 41.4, 55.2, 112.0, 113.3, 113.9, 119.1, 119.2, 123.3, 128.5, 128.5, 129.7, 162.3, 167.3.

3.1.2. N^b-2,4-dimethoxy-benzoyltryptamine (**2b**)

Yield of the product was 65%, orange solid from $CHCl_3$/MeOH (1:1), with m.p. 116 °C (Lit. reported as oil) [4]; IR ν_{max} (KBr) cm^{-1}: 1206, 1437, 1466, 1497, 1604, 1634, 2850, 2975, 3016, 3111, 3228, 3369; UV λ_{max} MeOH nm: 225, 255, 287; ^{1}H NMR (200 MHz, C_5D_5N) δ: 3.25 (2H, t, J = 6.8), 3.53 (3H, s), 3.67 (3H, s), 4.08 (2H, q, J = 6.4), 6.57 (1H, d, J = 2.2), 6.68 (1H, d, J = 7.8), 7.25 (1H, dt, J = 1.2; 7.0), 7.29 (1H, dt, J = 1.4; 7.0), 7.40 (1H, d, J = 1.6), 7.62 (1H, d, J = 8.2), 8.37 (1H, t, J = 5.2), 8.61 (1H, d, J = 8.6), 11.85 (1H, sl); ^{13}C NMR (50 MHz, C_5D_5N) δ: 26.0, 40.9, 55.4, 55.5, 98.9, 105.9, 112.0, 113.1, 115.7, 119.2, 119.3, 121.9, 123.5, 128.4, 133.9, 137.7, 159.4, 163.6, 165.2.

3.1.3. N^b-3,4-dimethoxy-benzoyltryptamine (**2c**)

Yield of the product was 63% as a white powder from $CHCl_3$/MeOH (1:1), with m.p. 182 °C (Lit. 182 °C) [5]; IR ν_{max} (KBr) cm^{-1}: 1230, 1416, 1441, 1461, 1504, 1605, 2839, 2873, 2930, 3011, 3080, 3219, 3390; UV λ_{max} MeOH nm: 220, 260, 290; ^{1}H NMR (200 MHz, C_5D_5N) δ: 3.36 (2H, t J = 7.4), 3.65 (3H, s), 3.72 (3H, s), 4.06 (2H, q, J = 8.8), 6.96 (1H, d, J = 8.2), 7.24 (1H, dt, J = 1.2; 7.0), 7.32 (1H, dt, J = 1.4; 7.0), 7.60 (1H, d, J = 7.2), 7.60 (1H, d, J = 2.2), 7.87 (1H, sl), 7.93 (1H, m), 7.99 (1H, d J = 2.0), 9.13 (1H, t J = 5.8); ^{13}C NMR (50 MHz, C_5D_5N) δ: 26.0, 41.5, 55.8, 56.9, 111.4, 111.8, 113.4, 119.1, 119.2, 121.0, 121.7, 123.4, 128.5, 128.7, 137.6, 149.5, 152.2, 167.4.

3.1.4. N^b-3,4-methylenedioxy-benzoyltryptamine (**2d**)

Yield of the product was 53% as a white powder from $CHCl_3$/MeOH (1:1) with m.p. 150 °C; IR ν_{max} (KBr) cm^{-1}: 1443, 1457, 1485, 1501, 1618, 1640, 2787, 2918, 2976, 3118, 3342, 3401, 3416; UV λ_{max} MeOH nm: 220, 260, 290; ^{1}H NMR (200 MHz, C_5D_5N) δ: 3.35 (2H, t J = 8.0), 4.04 (2H, q J = 6.0), 5.95 (2H, s), 6.91 (1H, d J = 8.6), 7.24 (1H, td J = 1.2; 7.0), 7.29 (1H, td J = 1.4; 7.0), 7.35 (1H, d J = 2.4), 7.59 (1H, d J = 1.2), 7.86 (1H, m), 9.06 (1H, t J = 6.0); ^{13}C NMR (50 MHz, C_5D_5N) δ: 26.3, 41.5, 102.5, 108.1, 108.4, 112.0, 113.2, 119.1, 119.2, 121.7, 122.6, 128.5, 137.6, 148.2, 149.7, 167.0.

3.1.5. N^b-3,4,5-trimethoxy-benzoyltryptamine (**5e**)

Yield of the product was 80%, white powder from $CHCl_3$/MeOH (1:1), with a m.p. 214 °C (Lit. 165 °C [6]; 200–202 °C [7]); IR ν_{max} (KBr) cm^{-1}: 1127, 1233, 1413, 1458, 1503, 1586, 1635, 2855, 2938, 2959, 3013, 3297, 3363; UV λ_{max} MeOH nm: 230, 260, 270; ^{1}H NMR (200 MHz, C_5D_5N) δ: 3.35 (2H, t J = 8.0), 3.64 (3H, s), 3.89 (3H, s), 4.05 (2H, q J = 6.0), 7.24 (1H, td J = 1.2; 7.0), 7.29 (1H, td J = 1.4; 7.0), 7.36 (1H, d J = 2.0), 7.61 (1H, m), 7.62 (1H, s), 7.88 (1H, d J = 2.0), 9.25 (1H, t J = 6.0); ^{13}C NMR (50 MHz, C_5D_5N) δ: 26.2, 41.6, 55.9, 60.5, 105.7, 112.0, 113.3, 119.1, 119.2, 121.7, 123.4, 128.6, 131.5, 137.6, 141.2, 153.6, 167.3.

3.2. Pharmacology

The tissues were suspended in 6 ml organ baths under a resting load of 1.0 g at 37 °C. Force generation was monitored using an isometric transducer (7003-UGO BASILE) coupled to a polygraph (7070-UGO BASILE). The modified Krebs solution: NaCl (117.0), KCl (4.7), $MgSO_4{\cdot}7H_2O$ (1.3), $NaH_2PO_4{\cdot}H_2O$ (1.2), $CaCl_2{\cdot}H_2O$ (2.5), glucose (11.0), $NaHCO_3$ (25.0) was bubbled with a 95% O_2 and 5% CO_2 gas mixture before use. Drugs—$NaHCO_3$, KCl, $MgSO_4{\cdot}7H_2O$ (Reagen); acetylcholine, $CaCl_2{\cdot}H_2O$, NaCl, $MgCl_2{\cdot}6H_2O$, $NaH_2PO_4{\cdot}H_2O$, glucose (Merck) and histamine (Sigma-

Table 1
Percentages values for relaxation of derivates (**2a**–**e**) in guinea-pig ileum

Substance (10^{-4} M)[a]	Acetylcholine (1 μM)[a]	Histamine (1 μM)[a]	KCl (40 mM)[a]
2a	77.6 ± 11.7	92.9 ± 4.1	88.7 ± 6.4
2b	100 ± 0	86.7 ± 1.0	100 ± 0
2c	73.0 ± 15.1	100 ± 0	64.0 ± 2.6
2d	75.3 ± 2.6	78.1 ± 1.4	54.6 ± 6.7
2e	100 ± 0	84.3 ± 2.0	78.3 ± 5.6

[a] $n = 4$ for all the tests; values are given in percentage of relaxation of the tonic contractions induced by 1 μM of acetylcholine or histamine, and KCl 40 mM.

Aldrich). Data analysis—unless otherwise stated, all values were expressed as mean ± S.E.M. Statistical analysis were performed by Student's "*t*"-test, and was considered significant when probability (P) was < 0.05. All data were analyzed with the software package GraphPad Prism version 3.02 (GraphPad Software Incorporated, San Diego, CA 92121, USA).

4. Results and discussion

The pharmacological effects on smooth muscle of isolated guinea-pig ileum for the derivatives synthesized in present study are listed in Table 1, with the percentages relaxation tested at 10^{-4} M. The most important finding is that all the substances tested presented spasmolytic action. The tests were performed measuring the percentage of relaxation of the contractions induced by acetylcholine (1 μM), histamine (1 μM) or KCl (40 mM).

These preliminary results showed a correlation between the substitution patterns in the tested compounds. The 4- and 3,4-substitution drops the activity (**2a**, **2c** and also **2d**, respectively), against acetylcholine-, histamine- and KCl induced contractions. An extra alkoxy-substitution at C-5 is also tolerated without significant increase of activity (**2e**), but an *ortho* group shows an increase in the tested activity. These findings suggest that the observed effect is probably from stereoelectronic nature.

The fact that all the substances were active reflects probably the presence of the indole core nucleus. The substances tested relaxed the guinea-pig ileum pre-contracted by acetylcholine, histamine and KCl in a non-selective manner. Independently of the contraction being evoked by either pharmacomechanical (acetylcoline and histamine) or electromechanical (KCl) coupling, the maintenance of tonic contraction involves activation of the voltage-operated Ca^{2+} channels (VOCC's) [8,9]. Since the substances tested relaxed the guinea-pig ileum pre-contracted with used contractile agents, it is suggestive that this effect is probably due to blockade of Ca^{2+} influx through VOCC'S, however further studies will be necessary to reinforce this hypothesis.

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