



SYNTHESIS OF HALOGENATED/NITRATED FLAVONE DERIVATIVES AND EVALUATION OF THEIR AFFINITY FOR THE CENTRAL BENZODIAZEPINE RECEPTOR

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Abstract: A series of halogenated/nitrated flavone compounds were synthesized. Some of the products were found to be potent central benzodiazepine receptor (BDZ-R) ligands. The structure-activity relationships (SAR) analysis of the new synthetic compounds, together with that of others already described, indicates that substitutions at position 6 or 6 and 3' in the flavone nucleus are the only ones that give rise to high affinity BDZ-R ligands. © 1997 Elsevier Science Ltd.

Introduction

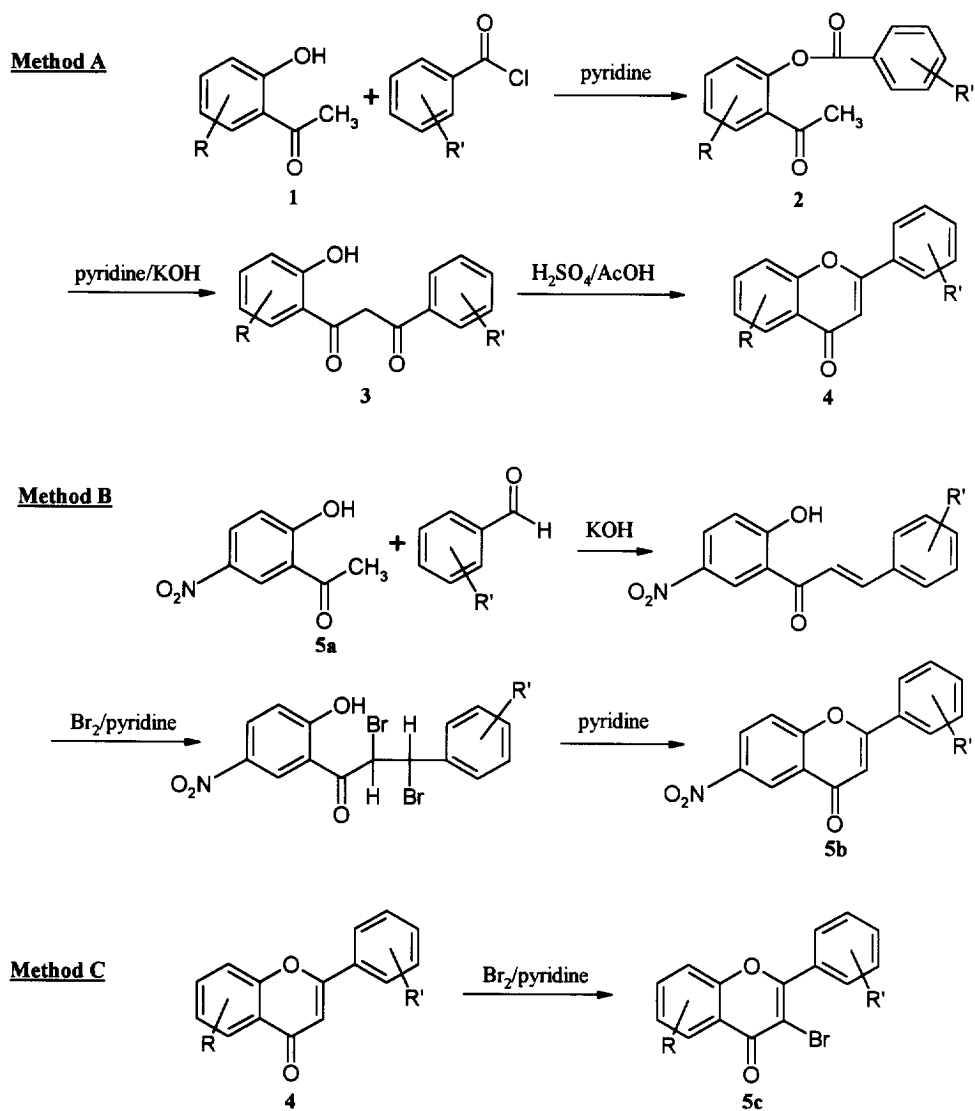
Previous work from our laboratories disclosed that some naturally occurring flavonoids, as well as the flavone nucleus itself, were ligands for the BDZ-Rs that exhibited low-medium affinity in vitro and anxiolytic activity in vivo, with minor sedative or myorelaxant effects.¹⁻³

The introduction of electronegative substituents in the flavone nucleus led to the discovery of novel high affinity ligands for the BDZ-Rs endowed with potent anxiolytic effects.⁴⁻⁷ Although it has yet to be established whether any of these compounds may originate a useful therapeutic drug, it was deemed important to synthesize a wider series of analogous compounds. In this Letter we describe the synthesis of 15 new halo/nitro derivatives of flavone and the measurement of their affinities for the BDZ-Rs. A series of SARs could be established that proved useful to define the minimum requirements for the generation of a high affinity ligand.

Chemistry

Three methods (A, B, and C), indicated in Scheme 1, were employed to prepare the analogs of interest in this study.

Scheme 1



We used the Baker-Venkataraman transformation, the most common method of synthesizing flavones.

In this process (Method A, Scheme 1) a hydroxyacetophenone (1) is first converted into a benzoyl ester (2) and this species is then treated with base, forming a 1,3-diketone (3). Treatment of this diketone with acid leads to generation of the desired flavone (4).⁸

In method B (Scheme 1), 2-hydroxy-5-nitro-acetophenone (5a) prepared according to Cushman et al.,⁹ was transformed into the corresponding B-ring substituted flavone(s) (5b) following the procedure described in reference 10.

In method C (Scheme 1), 3-bromine substituted flavones (5c) were obtained by treatment of the corresponding flavone (4) with bromine in pyridine.

Results and Discussion

Application of the chemical methods A, B, and C led to the synthesis of the majority of the flavone derivatives listed in Table 1, which also includes several similar compounds prepared and studied previously by us.^{4,5,7}

Binding affinities to BDZ-Rs were determined by the ability of the compounds to inhibit ³H-flunitrazepam binding to extensively washed rat cerebral cortical membranes. The methodology for the preparation of the membranes and the performance of the binding assays has already been described.¹¹

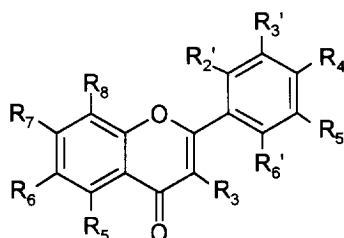
The following SARs were obtained by analysis of the information collected in Table 1 and are expressed as flavone substitutions that produce the following changes in the binding affinity of the mother compound (flavone, 6).

(A) Great increases (40 to 1000 times):

- A bromine or chlorine atom, or a nitro group in carbon 6 plus a halogen or nitro group in carbon 3' (22, 25, 27, 29, 30).

(B) Moderate increases (3.6 to 13 times):

- A bromine, or a chlorine atom, or a nitro group in carbon 6 (8, 9, 10).
- A bromine atom in carbon 6 plus a nitro group in carbon 2' (26), or in carbon 4' (28).
- A nitro group, or a bromine, or a chlorine atom in carbon 3' (12, 13, 14).
- A fluorine atom in carbon 6 plus a nitro group in carbon 3' (24).

Table 1: Binding affinity of flavone derivatives for the BDZ-Rs

Compound ^a	R ₃	R ₆	R ₂ '	R ₃ '	R ₄ '	R ₅ '	K _i ^b (μM)
6	H	H	H	H	H	H	1 ^c
7	H	F	H	H	H	H	4.5
8	H	Cl	H	H	H	H	0.164
9	H	Br	H	H	H	H	0.075 ^c
10	H	NO ₂	H	H	H	H	0.275
11	H	H	NO ₂	H	H	H	>20
12	H	H	H	NO ₂	H	H	0.285
13	H	H	H	Cl	H	H	0.614
14	H	H	H	Br	H	H	0.413
15	H	H	H	H	NO ₂	H	>20
16	H	H	H	H	Br	H	>20
17	H	H	H	NO ₂	H	NO ₂	>20
18	Br	H	H	H	H	H	>75 ^c
19	Br	Br	H	H	H	H	>75 ^c
20	Br	H	H	NO ₂	H	H	>20
21	H	NO ₂	NO ₂	H	H	H	>10
22	H	NO ₂	H	NO ₂	H	H	0.026 ^d
23	H	NO ₂	H	H	NO ₂	H	>10 ^d
24	H	F	H	NO ₂	H	H	0.180
25	H	Cl	H	NO ₂	H	H	0.008
26	H	Br	NO ₂	H	H	H	0.208 ^e
27	H	Br	H	NO ₂	H	H	0.001 and 0.016 ^e
28	H	Br	H	H	NO ₂	H	0.200 ^e
29	H	Br	H	Br	H	H	0.019
30	H	NO ₂	H	Br	H	H	0.025

^a All described compounds were fully characterized including spectroscopic and elemental analysis.

^b K_i ± SEM values are means of 3 to 5 independent determinations. For values ranging from 0.001 to 4.5, the SEM varied between 6 to 13% of the absolute values listed. The low affinity values are the result of duplicate measurements.

^c Ref. 5.

^d Ref. 4.

^e Ref. 7.

(C) Great decreases:

- A fluorine atom in carbon 6 (7).
- A nitro group in carbon 2' (11); a nitro group, or a bromine atom in carbon 4' (15, 16).
- Nitro groups in carbons 3' and 5' (17); in carbons 6 and 2' (21); or in carbons 6 and 4' (23).
- A bromine atom in carbon 3 (18) or in carbons 6 and 3 (19), a bromine atom in carbon 3 plus a nitro group in carbon 3' (20).

The effectiveness of the different substituents in carbon 6 is as follows: Br > Cl > NO₂ > F.

The influence of halogen atoms or nitro groups in carbons 5, 7, or 8 has not been established. However, some information on the relevance of these positions on the affinity for the BDZ-Rs of this kind of compounds can be obtained from the properties of several natural or synthetic flavonoids as reported by us.⁵ For instance, the presence of hydroxy groups in carbon 5 and 7 or 5, 7 and 4' is compatible with the occurrence of low to medium affinity properties. Further, the addition of halogen atoms in carbons 6 and 8, in one of those molecules (see compounds 23 and 24 in reference 5) does not appreciably modify the affinity of the mother molecule.

In conclusion, substitution with halogen atoms, or nitro groups, in carbon 6 or carbon 6 and 3' of flavone gives rise to fairly active compounds, both in vitro and in vivo.⁴⁻⁷ Similar substitutions in other carbons, with the limitations indicated before, are less effective or, alternatively, originate inactive compounds. Compound 9 behaves as a full BDZ-R agonist⁴, while compounds 22 and 27 have a partial agonist profile at the BDZ-R^{6,7} with potent anxiolytic properties, without evidencing myorelaxant or sedative actions. Biochemical and pharmacological experiments are now in progress to characterize the properties of compounds 25, 29, and 30.

Acknowledgements

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