gates of MTX and anti-tumour monoclonal antibodies (Baldwin et al 1986).

Overall, these and other related studies (Corvalan et al 1987) indicate that modification of drug biodistribution with monoclonal antibodies and/or tissue specific targeting with complexes formed with hybrid antibodies may be valid approaches to prolongation of drug survival and/or site-specific targeting. The degree of drug loading of monoclonal antibodies, with theoretical maxima of two molecules per molecule with anti-drug antibody and one molecule per molecule of hybrid antibody, might be a limitation, since the amount of antibody required to carry conventional therapeutic doses of drug would be very high. In addition the extent of extravasation of antibody is much smaller than that of drugs. But, as this study shows, the survival of drug is markedly prolonged simply with the anti-drug antibody, and the additional site targeting effect possible with hybrid antibodies might mean the possible use of greatly reduced doses of drug.

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Letter to the Editor

The influence of aromatic substituents on the binding of substituted benzamides to dopamine D-2 receptors: congruent QSAR and MEP analyses

B. Testa*, N. El Tayar, P. A. Carrupt, H. Van de Waterbeemd, G. J. Kilpatrick†, P. Jenner†, C. D. Marsden†
School of Pharmacy, University of Lausanne, CH-1005 Lausanne, Switzerland, †MRC Movement Disorders Research Group, University Department of Neurology and Parkinson's Disease Society Research Centre, Institute of Psychiatry and King's College Hospital Medical School, Denmark Hill, London SE5 8AF, UK

Substituted benzamides (orthopramides) are a group of atypical neuroleptics principally acting as selective antagonists of dopamine D-2 receptors. Besides the essential carboxamide group in position 1 and alkoxy group in position 2, these compounds exhibit various other ring substituents, mainly in positions 4 and 5. A few years ago, we demonstrated the critical role of adequate aromatic substitution at positions 4 and 5 for binding to the D-2 receptors, analogues of metoclopramide unsubstituted in these positions being fully devoid of in-vitro D-2 receptor affinity (Anker et al 1983).

In an effort to unravel the structure-activity relationships (SAR) of the 4- and 5-substituents, some of us (Testa et al 1986; Van de Waterbeemd et al 1986a,b) have calculated the molecular electrostatic potential (MEP) maps of a number of variously ring-substituted orthopramides using ab initio (STO-3G basis set) molecular orbital (MO) calculations. Consistent results were obtained which led us to propose a stereo-electro-

* Correspondence

static pharmacophore characterizing orthopramides and featuring the following pharmacophoric elements (Fig. 1): a plane of zero-potential cutting the aromatic region and separating a sector of positive potential (on the right-hand side) from a sector of negative potential; a region of maximum positive potential located close to the methoxy oxygen atom; two regions of minimum negative potential located near the carbonyl oxygen and the 5-substituent. From such a model, it can be deduced that the 5-substituent must be electron-rich in order to generate the appropriate strong negative potential, while the 4-substituent, being in a region where the potential might be slightly positive, cannot be electron-rich.

Recently (El Tayar et al, unpublished), we have measured the affinity of twenty orthopramide derivatives to the rat striatal dopamine D-2 receptor (displacement of [^{3}H]spiperone as expressed by K_i values). The compounds were a group of eleven analogues of

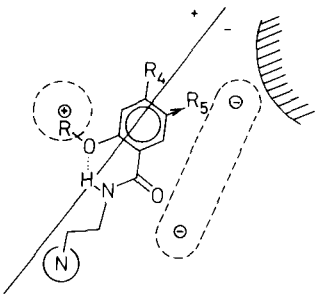
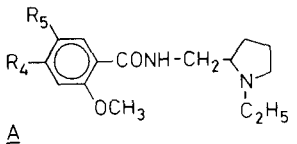
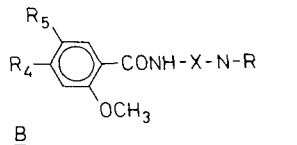


FIG. 1. Stereo-electrostatic pharmacophore of orthopramides (Van de Waterbeemd et al 1986a,b) as confirmed in the present study. This pharmacophore is juxtaposed with a region of steric hindrance in the D-2 receptor as rationalized from equations 1 and 2.



A



B

FIG. 2. General structure of sulpiride analogues (A) and other benzamide derivatives (B).

sulpiride (Fig. 2A), and a group of nine other benzamide derivatives (Fig. 2B; X = 2-pyrrolidylmethyl, 4-piperidinyl or 4-tropanyl; R = alkyl or benzyl). The compounds exhibited a large variety of aromatic substituents in the 4- and 5-position, the steric and electron-withdrawing properties of which were expressed by the molar refractivity (MR) and the sigma (σ_{meta} and σ_{para}) parameters, respectively. Quantitative SAR (QSAR) analysis lead to two significant multiple regression equations, presented here in their normalized form (Mager & Barth 1979) to evaluate better the contribution of each parameter to the explained variance. For the 11 sulpiride analogues, affinity is correlated with the steric and electronic properties of the 5-substituent (95% confidence limits in parentheses):

$$pK_i = -0{\cdot}66\ (\pm 0{\cdot}44)\ MR_5 + 0{\cdot}73\ (\pm 0{\cdot}42)\ \sigma_{m5}$$
$$n = 11;\ R = 0{\cdot}866;\ s = 0{\cdot}496;\ F = 12{\cdot}2 \qquad (1)$$

For the complete data set, a better correlation equation was obtained which also included the electronic effect of the 4-substituent:

$$pK_i = -0{\cdot}42\ (\pm 0{\cdot}22)\ MR_5 + 0{\cdot}24\ (\pm 0{\cdot}21)\ \sigma_{m5} - (0{\cdot}60)\ (\pm 0{\cdot}23)\ \sigma_{p4}$$
$$n = 20;\ R = 0{\cdot}928;\ s = 0{\cdot}491;\ F = 33{\cdot}1 \qquad (2)$$

When interpreted in molecular terms, equations 1 and 2 indicate the following: (a) an electron-withdrawing substitutent in the 4-position is detrimental to affinity; (b) in contrast, an electron-withdrawing substituent in the 5-position enhanced affinity; (c) the bulkier the 5-substituent, the smaller the affinity. Conclusions (a) and (b) are in full agreement with, and offer independent validation of, the pharmacophore shown in Fig. 1, particularly the electrostatic contributions of the 4- and 5-substituents. In addition, conclusion (c) is a novel finding indicating that large 5-substituents are repelled by a poorly defined and presumably flexible zone of steric hindrance (see Fig. 1).

The congruence of MEP calculations and QSAR analysis is a noteworthy asset in topographical studies of dopamine D-2 and other receptors.

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