Similarities in the interatomic distances of some anti-inflammatory agents and inflammagenic amines: a possible insight into their common receptor(s)

SIR,—Histamine and 5-hydroxytryptamine (5-HT) both promote inflammation in small animals, with 5-HT more effective in the rat than in other animals or man (Skidmore & Whitehouse, 1967).

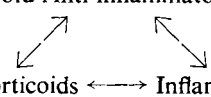
Recently Kier has postulated the preferred conformation of the (17β)-α-ketol side-chain in corticosteroids from molecular orbital calculations of the total energy of the molecule in each of several possible conformations, using the extended Hückel theory (Kier, 1968c). The distances between the hydrogen of the 11β-hydroxy group and each of the two oxo-groups in cortisol (hydrocortisone) in its preferred conformation are rather close to the inter-nitrogen distances in one of the two preferred conformations of histamine (Kier, 1968a) and in 5-HT in its preferred conformation (Kier, 1968b) (see the upper part of Table 1). Kier has therefore postulated that the 11β-hydroxy group hydrogen of cortisol, which is essential for anti-inflammatory activity, could also fit those (inflammagenic) receptor sites which bind the protonated ethylamino-group of histamine or 5-HT, while the 20-oxo- and 3-oxo-groups of cortisol could, respectively, attach to those receptor sites which normally bind the imidazole nitrogen of histamine or the indole nitrogen of 5-HT.

TABLE 1. INTERATOMIC DISTANCES IN CORTISOL, SOME NONSTEROID ANTI-INFLAMMATORY ACIDS (OR THEIR METABOLIC DERIVATIVES), AND INFLAMMAGENIC AMINES

Molecule	Inflammation activity	Distance (Å)
Cortisol	A	20-oxo to 11-oxy-H = 4·8
Histamine	P	Ring N to NH_3^+ = 4·55
Cortisol	A	3-oxo to 11-oxy-H = 6·0
5-HT	P	Ring N to NH_3^+ = 5·84
5-MeO-MIAA*	[A]	Ring N to carboxyl-H = 6·17 or 5·05
3'-Oxophenylbutazone*	A	3'-oxo to ring-ene-3(5)-ol-H = 6·0 or 4·4
N-Arylanthranilic Acids	A	N to carboxyl-H = 4·65
Salicylic Acid	A	Phenolic-O to carboxyl-H = 4·60

* See text
A = antiflammatory; P = pro-inflammatory

This postulate can be extended to include certain nonsteroid anti-inflammatory drugs which, we propose, may be related as in the diagram below. This depicts a pharmacological relation based on possible identity of receptors, the connection lines representing "receptor equivalents".



Indomethacin, phenylbutazone, the fenamic acids (*N*-arylanthranilates), and salicylates are nonsteroid drugs widely used as alternatives to cortisol (or its more potent derivatives) for treating rheumatoid diseases. They all mimic cortisol in suppressing acute inflammation in experimental animals and the adjuvant-induced arthritis in rats (reviewed by Winter, 1966; Adams & Cobb, 1967; Shen, 1967).

Indomethacin (1,4-chlorobenzoyl-5-methoxy-2-methylindol-3-ylacetic acid) is metabolized in animals, but not apparently in man, with loss of the N_1-chlorobenzoyl moiety (Shen, 1967), possibly through the action of chymotrypsin (Skidmore & Whitehouse, 1967). Molecular orbital calculations indicate that

there are two energetically preferred conformations of the acetic acid side-chain in 5-methoxy-2-methyl-indole acetic acid (MeO-MIAA). The plane of the protonated carboxyl group lies perpendicular to the plane of the ring and two rotamers (preferred conformations) arise because of two energy barriers to rotation of the methylene-carboxyl bond. The distance between the carboxyl hydrogen and the ring-nitrogen in MeO-MIAA is then either 5·05 or 6·17 Å, but this interfunctional group distance cannot be less than 5·0 Å. Therefore, the N_1 and carboxyl-H could not simultaneously bind to the two nitrogen binding sites (4·55 Å apart) of the postulated histamine receptor, but they could simultaneously bind to the two nitrogen-binding sites (5·84 Å apart) of a 5-HT receptor. It is merely coincidental that indomethacin is metabolized to MeO-MIAA in several animal species (rat, rabbit, guinea-pig) but only in the rat, which is uniquely sensitive to 5-HT as an inflammatory mediator, does it have extremely potent activity (at least 80 × phenylbutazone)? In man, indomethacin is not much more potent than phenylbutazone and MeO-MIAA has not been found as a metabolite (Shen, 1967).

Phenylbutazone (1,2-diphenyl-4-n-butyl-pyrazolidine-3,5-dione) is metabolized in man yielding either a phenolic derivative, oxyphenbutazone, or another hydroxy metabolite with a secondary alcoholic group at C-3 in the n-butyl side-chain. This latter compound is claimed to be devoid of anti-inflammatory activity but is chemically labile (see Whitehouse, 1965). The corresponding γ-ketone, another potential metabolite, is an effective antirheumatic agent used, for example, in Czechoslovakia under the name Ketazon. It is a much weaker drug than phenylbutazone *in vitro* in test systems not related to possible amine antagonism (Whitehouse & Leader, 1967). A 3-sulphoxy analogue, sulphinpyrazone (Anturan), is also an effective antirheumatic drug and is more potent than phenylbutazone in suppressing oedema due to formalin or 5-HT in rats (Adam & Cobb, 1967). These experimental observations suggest that a 3′-carbonyl or 3′-sulphoxy group could contribute to drug activity, especially in antagonizing inflammatory amines. A molecular model 3′-oxophenylbutazone, with the 3-oxobutyl side-chain folded in the normal *trans* conformation of paraffin derivatives, shows that the interatomic distances between the hydrogen of a protonated enol group on the pyrazolidine ring and the 3′-oxo group in the benzyl side-chain can be either 4·4 or 6·0 Å, depending on whether the enolic hydrogen is cisoid or transoid with respect to the side-chain. This is remarkable, for it indicates that this one molecule, like cortisol, could possibly interact with both a histamine and a 5-HT receptor. The distances between a ring carbonyl group and the hydrogen atom of a 3-hydroxyl group in the phenylbutazone metabolite from man can be likewise either 4·6 or 5·8 Å.

The salicylate anion is stabilized by an intramolecular hydrogen bond (Baker, 1936) and, if this ion is then protonated, the distance between the phenolic oxygen and the carboxyl hydrogen is 4·6 Å. In the *N*-aryl anthranilic acids mefenamic acid, flufenamic acid, and meclafenamic acid, the distance between the secondary nitrogen and the carboxyl hydrogen is 4·65 Å, assuming a similar hydrogen bond to fix the conformation of the carboxyl group. Thus, both the unionized salicylate and anthranilates might compete with histamine for a receptor site.

These conclusions about possible agonist-antagonist relations presuppose (i) that phenylbutazone and indomethacin are metabolized *in vivo* and that the metabolites may be pharmacologically active *in situ*, even though the same molecular species supplied exogenously are reported to have little or no anti-inflammatory activity (in assays not designed necessarily to detect amine antagonism) probably because they are not similarly distributed or as long-lived *in vivo*

as the parent drugs which engender these metabolites, (ii) that the 11β-hydroxyl of cortisol, the carboxylic hydroxyl group of indole-acetic and other substituted benzoic acids, and the enol group of phenylbutazone are all equivalent to a protonated amino-group, not in any functional sense of reactivity but in a passive binding role, and (iii) it is the unionized form of these nonsteroid drugs that associates with the receptor surface for the inflammagenic amines.

These postulates are not incompatible with previous conclusions that at other receptor sites, not concerned with amine binding, these nonsteroid drugs may be ionized and bound as anions—for these drugs are certainly polyvalent and may influence more than one event in the overall inflammatory process (Whitehouse, 1968). Even though these particular nonsteroids and cortisol may have the requisite intramolecular features for displacing the inflammatory amines from certain receptors, they may not necessarily act as classic anti-5-HT or anti-histamine drugs, that is to say as competitive antagonists; rather, they may serve as false feedback inhibitors to "switch off" amine biogenesis if indeed amine formation is autoregulated by either allosteric or end-product inhibition.

To summarize: It may be significant that both anti-inflammatory steroids, related to cortisol, and nonsteroid anti-inflammatory agents, like fenamates, salicylic acid, and some metabolites or chemical modifications of phenylbutazone and indomethacin, contain oxo- and oxy-groups or oxy and imino-groups so disposed within the molecule that they could also bind to these parts of certain receptors for histamine and 5-HT, that bind the ring nitrogen and ethylamino nitrogen of these two inflammagenic amines.

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