DDT Vol. 6, No. 13 July 2001

Monitor: molecules and profiles

Monitor provides an insight into the latest developments in drug discovery through brief synopses of recent presentations and publications together with expert commentaries on the latest technologies. There are two sections: *Molecules* summarizes the chemistry and the pharmacological significance and biological relevance of new molecules reported in the literature and on the conference scene; *Profiles* offers commentary on promising lines of research, emerging molecular targets, novel technology, advances in synthetic and separation techniques and legislative issues.

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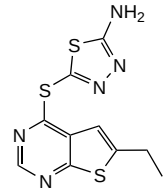
Molecules

Inhibition of cell adhesion molecule expression

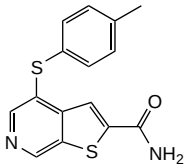
Inflammatory processes involve the migration of circulating leukocytes to a site of tissue injury. This migration requires the induction of cell adhesion molecules on the surface of vascular endothelial cells. The cell adhesion molecules include E-selectin, intercellular adhesion molecule-1 (ICAM-1) and vascular adhesion molecule-1 (VCAM-1), and serve to slow and ultimately arrest leukocytes, facilitating their movement along a chemotactic gradient to the site of tissue injury.

In chronic inflammatory diseases, the natural response is exaggerated to such a degree that it is harmful. Pharmaceutical agents that reduce the induced expression of one or more of the cell adhesion molecules would be expected to attenuate the inflammatory process.

A group at Abbott Laboratories (Abbott Park, IL, USA) set out to discover compounds that inhibit cytokine-induced expression of one or more of E-selectin, ICAM-1 and VCAM-1 (Ref. 1). A high-throughput transcriptional screen identified compound **(i)** as a low micromolar inhibitor of the expression of all three adhesion molecules. In an effort to improve potency, different substituents and heterocyclic core structures were prepared and tested in whole-cell ELISA assays. Compound **(ii)** was identified as a potent inhibitor of E-selectin and ICAM-1 expression (IC_{50} = 0.02 and 0.025 μM, respectively) but not VCAM-1 (IC_{50} = >1 μM). The position of the nitrogen atom was crucial because the other possible pyridine isomers were inactive. The molecule was shown to have little cellular toxicity and to be an effective inhibitor of cell–cell adhesion in an *in vitro* flow model.



(i)



(ii)

Pharmacokinetic analysis showed that the two major routes of metabolism are rapid amide hydrolysis to the acid, and oxidation of the sulfide to the sulfoxide. Both compounds are inactive and the lack of *in vivo* stability has limited the use of compound **(ii)** for proof-of-principle animal studies.

1 Stewart, A.O. *et al.* (2001) Discovery of inhibitors of cell adhesion molecule expression in human endothelial cells. 1. Selective inhibition of ICAM-1 and E-selectin expression. *J. Med. Chem.* 44, 988–1002

Novel ibutilide analogues for the treatment of re-entrant cardiac arrhythmias

Cardiac arrhythmias continue to represent a major source of human morbidity and mortality. From a historical standpoint, therapy for cardiac arrhythmias began with the identification of quinidine as the active component of a cinchona alkaloid preparation[2]. Three decades later, electrophysiology studies indicated that quinidine significantly prolonged the effective refractory period (ERP) in ventricular myocardium, as a consequence of its ability to slow conduction by depressing the maximal rate of rise of the cardiac action potential. On this basis, a series of class I antiarrhythmic agents (e.g. procainamide, lidocaine, disopyramide, encainide and flecainide) were investigated for their ability to reduce the rate of death from arrhythmia in patients that had suffered a myocardial infarction (MI). However, a Cardiac Arrhythmia Suppression Trial (CAST) showed that the death rate in the treated group exceeded that in the control group, which led to a diminished interest in this class[3]. Studies with sotalol and amiodarone suggested that the ERP of cardiac tissue could also be increased by a different mechanism, which led to the development of the class III antiarrhythmic agents. Unfortunately, class III antiarrhythmic activity can be associated with a polymorphic ventricular

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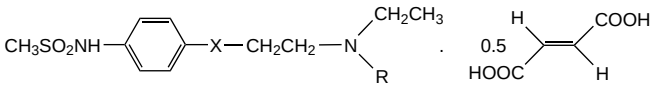

(iii) X = $CH(OH)CH_2$ R = $(CH_2)_6CH_3$

(iv) X = (*S*)-$CH(OH)CH_2$ R = $(CH_2)_5C(CH_3)_2F$

tachyarrhythmia (PVT), known as *torsade de pointes*.

When tested in clinical studies, ibutilide fumarate **(iii)**, a marketed class III representative, showed an incidence of *torsade de pointes* lower than 8%. However, the drug suffered by a rapid first-pass metabolism and could only be given by intravenous administration. On these bases, Hester and coworkers[4] have recently published a series of ibutilide analogues, modified at the heptyl side chain, which is the first responsible for metabolic degradation in the model compound. Modifications included the incorporation of alkyl, cycloalkyl, hydroxy, acetoxy and fluoro substituents. Their metabolic stability was initially evaluated *in vitro* by incubating the compounds with human liver microsomes. Preliminary results on rabbit heart preparations indicated the importance of the fluorine substitution. In particular, compound **(iv)** was devoid of proarrhythmic activity in a rabbit proarrhythmia model and was chosen for further evaluation in two canine models of re-entrant arrhythmias. The compound showed interesting activity and is currently undergoing clinical evaluation for the treatment of atrial arrhythmias.

2 Morgan, P.H. *et al.* (1976) Arrhythmias and antiarrhythmic drugs: mechanism of action and structure–activity relationship. *J. Pharm. Sci.* 65, 467–482

3 The Cardiac Arrythmia Suppression Trial (CAST) Investigators (1989) Preliminary report: effect of encainide and flecainide on mortality in a randomized trial of arrhythmia suppression after myocardial infarction. *New Engl. J. Med.* 321, 406–412

4 Hester, J.B. *et al.* (2001) Progress towards the development of a safe and effective agent for treating re-entrant cardiac arrhythmias: synthesis and evaluation of ibutilide analogues with enhanced metabolic stability and diminished proarrhythmic potential. *J. Med. Chem.* 44, 1099–1115

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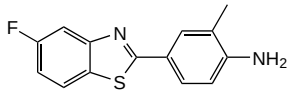
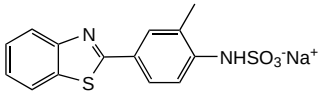
Novel antitumour molecules

Potent and selective antitumour benzothiazoles

The 3′-substituted 2-(4-aminophenyl) benzothiazole series possess, in a consistent pattern, potent antitumour activity (GI_{50} = < 1nM) in certain sensitive human cancer cell lines only (e.g. breast MCF-7, MDA-468; renal TK-10; ovarian IGROV-1). Other notable features of this class of agent include the unique biphasic dose–response relationship in sensitive cell lines, with cell kill occurring at low nanomolar concentrations, followed by a proliferative response (second growth phase) at low micromolar concentrations. It has been postulated that the second growth phase might be elicited by a metabolite that inactivates the bioactivating enzyme, cytochrome P450 CYP1A1; for the 3′-methyl substituted member of the series, the corresponding 6-hydroxylated metabolite has been identified to fulfil this role. To thwart deactivating metabolism, a series of fluorinated 2-(4-aminophenyl)benzothiazoles have been synthesized and evaluated as antitumour agents by Stevens and coworkers (Cancer Research Laboratories, University of Nottingham, UK)[1]. The fluorinated analogues retained the potency and selectivity *in vitro* of the parent non-fluorinated analogues, whereas in certain cases (5- and 7-fluorination) abolished the biphasic dose–response relationship. The most potent broad-spectrum agent in this series, 2-(4-amino-3-methylphenyl)-5-fluorobenzothiazole **(i)** is now the focus of pharmaceutical and preclinical development.



(i)



(ii)

A potential problem concerning the clinical use of these agents is posed by their lipophilicity, which presents limitations on drug formulation and bioavailability. This is because an aqueous intravenous formulation is desired to minimize the possibility of first pass deactivating metabolism. A further paper by Stevens and coworkers[2] describes the synthesis and physicochemical properties of sulfamate salt derivatives, (e.g. **ii**), of 2-(4-aminophenyl)benzothiazoles as potential (water-soluble) prodrugs for parenteral administration. These novel salts were found to be sparingly soluble under aqueous conditions (pH 4–9), and degradation to the free bioactive amine occurred under strongly acidic conditions.

1 Stevens, M.F.G. *et al.* (2001) Antitumor benzothiazoles. 14. Synthesis and *in vitro* biological properties of fluorinated 2-(4-aminophenyl)benzothiazoles. *J. Med. Chem.* 44, 1446–1455

2 Stevens, M.F.G. *et al.* (2001) Antitumour benzothiazoles. 15. The synthesis and physicochemical properties of 2-(4-aminophenyl)benzothiazole sulfamate salt derivatives. *Bioorg. Med. Chem. Lett.* 11, 1093–1095

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