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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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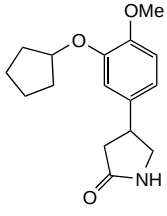
Molecules

Selective phosphodiesterase 4 inhibitors

The cyclic nucleotide phosphodiesterases (PDEs) are hydrolases that are responsible for the degradation of the second messengers cAMP and cGMP. Evidence suggests that cAMP plays a central role in regulating the function of airway smooth muscle, inflammatory cells, and immune cells. In particular, the cAMP-specific PDE4 is the predominant isoenzyme found in pro-inflammatory cells associated with several airway disorders [1,2]. It is, therefore, suggested that selective PDE4 inhibitors could be good therapeutic agents for the treatment of asthma, chronic obstructive pulmonary disease (COPD) and other inflammatory conditions [3]. However, the use of the first generation of PDE4 inhibitors (e.g. rolipram, **i**) has been severely curtailed because of dose-limiting side effects such as nausea and vomiting [4].

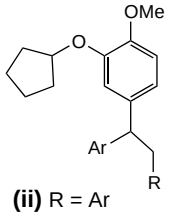
Alexander and co-workers [5] have started a project to identify a novel, potent and selective series of inhibitors devoid of these side effects. In particular, exploring the structure–activity relationship (SAR) around rolipram provided them with several series of PDE4 inhibitors, which ultimately led to the triarylethanes (**ii**). All of the compounds were tested for their potency against PDE4 and for their ability to inhibit lipopolysaccharide (LPS)-stimulated tumour necrosis factor-α (TNF-α) production in human whole blood, a measure of cellular efficacy of the inhibitor [6]. Among several other potent derivatives, compound CDP840 (**iia**) has an IC_{50} value against PDE4 of 4 nM. However, its ability to inhibit LPS-induced TNF-α production in human whole blood is 8.5 μM, representing a 2125-fold reduction of activity.



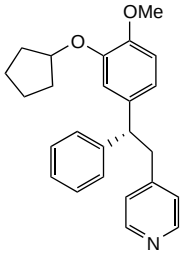
(i) Rolipram

CDP840 proved to be an exceptionally selective PDE4 inhibitor, having an IC_{50} value against PDE subtypes 1, 2, 3, 5 and 7 of >10,000 nM. Its distomer (enantiomer showing lower activity; **iib**) is >30-fold less active against PDE4, although this differential is reduced to threefold when inhibition of LPS-stimulated TNF-α production in human whole blood is considered (IC_{50} = 148 nM against PDE4 and 27.6 μM against TNF-α production).

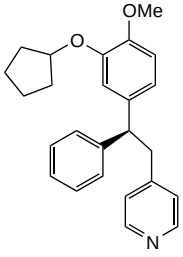
When profiled in animal models of pulmonary eosinophilic inflammation and antigen-induced bronchoconstriction, CDP840 (**iia**) was shown to reduce both inflammation and bronchoconstriction after oral administration. In addition,



(ii) R = Ar



(iia) CDP840



(iib)

CDP840 attenuated the late-phase asthmatic response to allergen challenge in patients. Finally, CDP840 was non-emetic in ferrets at doses up to 30 mg kg^{-1} perorally. These results suggest that the triarylethane series could be the basis of an orally prophylactic treatment for asthma or other inflammatory diseases.

Guay and coworkers have recently published [7] the results of their SAR

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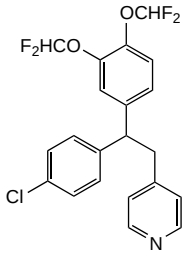
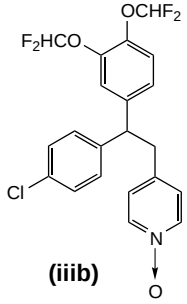
studies directed towards improving the potency and metabolic stability of CDP840. CDP840 suffers from extensive metabolism *in vitro* [8] and this translates into a short half-life *in vivo*. In the rat, the major metabolic pathway of CDP840 is the *para*-hydroxylation on the pendant group, whereas in human hepatocytes the major metabolite is the pyridinium glucuronide, which is not detected in rat hepatocytes.

The goal of this project was to increase both the potency and the metabolic stability of CDP840. After trying many structural modifications of the cathecol ring, which led to a constant loss of potency, the group obtained a significant improvement with the *bis*-difluoromethoxy analogue of CDP840 (compound **iiia**) and its corresponding *N*-oxide (compound **iiib**), which showed an increased potency for the inhibition of LPS-induced TNF-α in human whole blood (IC_{50} = 4.5 μM, 3.8 μM and 16 μM for **iiia**, **iiib**, and CDP840, respectively).

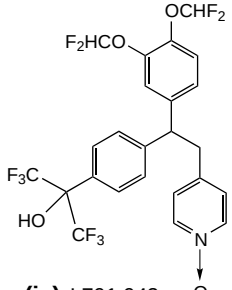


(iiia)



(iiib)

Having discovered the metabolically stable cathecol, the group focused on potency. They synthesized a few hundred analogues in this series, which showed that substitution in the *para*-position was in general better tolerated. Compound L791,943 (**iv**) was selected for *in vitro* evaluation of its metabolism in rat hepatocytes and compared with CDP840. The results indicated that, in standard incubation conditions, >98% of the parent drug remained in the case of L791,943 whereas only 11% of CDP840 was left intact.



(iv) L791,943

Additional assays showed that L791,493 was active in blocking the ovalbumin-induced bronchoconstriction in conscious guinea pig by 58% at a dose of 1 mg kg^{-1} (intraperitoneally 4 h pretreatment). L791,493 showed good *in vivo* activity in the anesthetized squirrel monkey and in the conscious sheep models of ascaris-induced bronchoconstriction. Finally, the potential of L791,943 for causing hemesis was assessed. Ferrets could be dosed orally up to 30 mg kg^{-1}, with plasma concentrations reaching 14 μM, without causing emesis.

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Identification of a protein tyrosine phosphatase 1B inhibitor with cellular activity

A characteristic of non-insulin dependent diabetes mellitus (NIDDM) is the resistance of cells to insulin. The cellular response to insulin binding to its receptor is dependent upon the auto-phosphorylation of the insulin receptor on a tyrosine residue and the subsequent phosphorylation of downstream effector proteins. In insulin-resistant cells the signalling cascade is attenuated most probably as a result of a defect in the insulin receptor itself.

Protein tyrosine phosphates (PTPs) in conjunction with protein tyrosine kinases (PTKs) regulate the level of protein phosphorylation. The protein tyrosine phosphate (PTP1B) has been shown to negatively regulate insulin signalling and thus inhibitors of PTP1B would be expected to prolong the activated state of the insulin receptor and be a potential novel treatment for NIDDM.

Most progress towards specific phosphatase inhibitors has been to mimic the natural substrate; a phosphotyrosine containing polypeptide. As part of a collaboration between scientists at Biovitrum (http://www.biovitrum.com) and Pharmacia (http://www.pharmacia.com), the peptide Ac-NH-Asp-Tyr(SO_3H)-Nle-NH_2 was identified as a competitive inhibitor of PTP1B (K_i = 5 μM) [9].