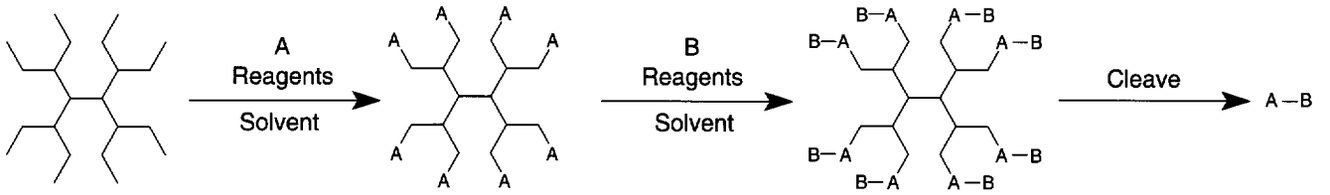
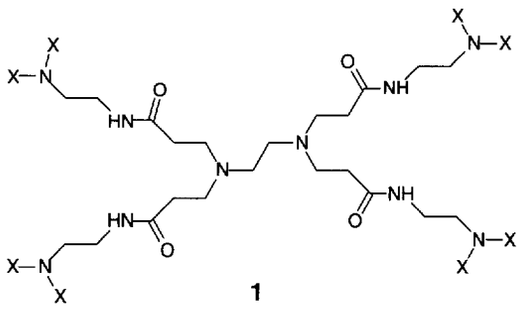
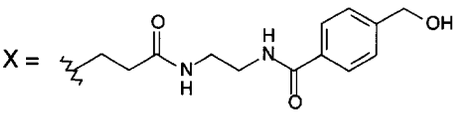
Combinatorial chemistry

Scheme 1





1

X = 

Dendrimer supports

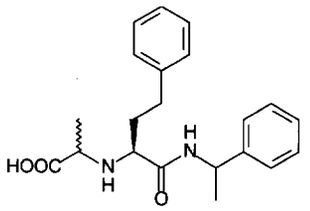
Several groups have tried to combine the reliability of solution-phase synthesis with the purification advantages offered by solid-phase. The latest contribution to this 'hybrid' library technology is the use of dendrimers to support combinatorial chemistry [Kim, R.M. *et al. Proc. Natl. Acad. Sci. U. S. A.* (1996) 93, 10012–10017] (Scheme 1). Dendrimers are branching oligomers characterized by discrete and controllable molecular architectures, and building compound libraries on the dendrimers provides several advantages. Solution-phase synthetic techniques can be used while purification may be effected by size-based separations such as size-exclusion chromatography.

The intermediates may be characterized by a variety of routinely available analytical methods such as NMR, IR, UV and mass spectrometry. Also, because of the nature of dendrimer structure, the molecules provide extremely high loading. For example, to obtain the amount of product that would be synthesized on 100 mg of a resin with loading of 0.23 mmol/g, only 7 mg of the dendrimer (**1**) is required. The dendrimer structure (**1**) has been used for the Fischer synthesis of a library of indoles, giving individual compounds in high purity and yield. As in solid-phase synthesis, a chemical cleavage step at the end of the synthesis provides the products free of the supporting dendrimer.

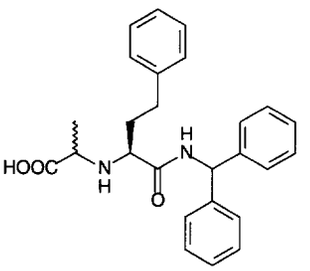
Exploiting rational drug design

A recent paper from DuPont Merck describes a successful marriage of combinatorial chemistry and rational drug design [Decicco, C.P., DeGrado, W.F. *et al. J. Am. Chem. Soc.* (1996) 118, 10337–10338]. In the search for novel inhibitors of the matrix metalloproteinases stromelysin (MMP-3) and collagenase (MMP-8) this group prepared, through solid-phase synthesis, a library of over 100 *N*-carboxyalkyl amino acid derivatives. One derivative (**2**) was weakly active (10% inhibition of MMP-3 at 100 mM), but a consideration of how the 1-phenylethyl group bound led to the rational design of benzhydrylamine derivative **3**. Introducing the benzhydrylamine derivative into a more potent hydroxamic acid template gave **4**, which inhibited MMP-3 and MMP-8 with K_i = 148 nM and 1.9 nM respectively.

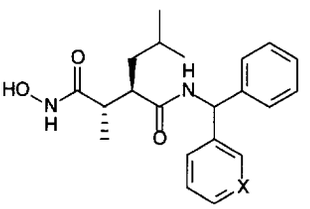
The crystal structure of **4** bound to MMP-3 was determined and revealed an unexpected conformational shift of a loop region of the protein. Examination of the crystal structure suggested that a hydrogen bond acceptor in the inhibitor would enhance binding. The pyridine derivative **5** was synthesized, and this derivative exhibited potent binding to MMP-3 (K_i = 9 nM).



2



3



4 X=CH
5 X=N

Emerging molecular targets

Intracellular receptor for LTB_4

Dihydroxy fatty acid leukotriene B_4 (LTB_4) is an important regulator of the inflammatory response. Its role was thought to be confined to that of a chemotactic factor that recruits and directs leukocytes to initiate an inflammatory reaction. To accomplish this, LTB_4 binds to specific receptors on the surface of immune cells and triggers a complex cascade of intracellular events that result in chemotaxis. Recent research by Dr Pallavi Devchand and coworkers at the Lausanne University (Switzerland) and the National Institutes of Health (Bethesda, MD, USA) has now also implicated the involvement of an intracellular receptor, the peroxisome proliferator-activated receptor α (PPARα) to further explain the role of LTB_4 in mediating the inflammatory response [*Nature* (1996) 384, 39–43].

PPARα is a transcription factor that controls the expression of a group of enzymes essential for general lipid catabolism, including the degradation of LTB_4. The enzymes that are synthesized in response to activation of PPARα include those involved in microsomal ω- and peroxisomal β-oxidation. Devchand's group have established that LTB_4 binds to PPARα in a saturable and specific manner, competes for binding to PPARα with a well-established hypolipidaemic factor (Wy14643; Campro Scientific), and upregulates the lipid catabolic enzyme acyl CoA oxidase, the gene for which is regulated by PPARα. LTB_4 does not bind to the other subtypes of the PPAR transcription factors, PPARβ and PPARγ; this further suggests that its binding to PPARα is specific.

Interaction of LTB_4 with PPARα is believed to act as a feedback mechanism leading to the degradation of LTB_4 and the termination of its chemotactic signal, thus providing a means by which the inflammatory response is self-limiting. The interesting observation that LTB_4 competes with Wy14643 for the same binding site on PPARα suggests that it may also have a role in the control of general lipid metabolism or possibly may play a part in the control of obesity. These findings will certainly fuel discussion and debate between scientists studying inflammation and those interested in intermediary metabolism of lipids and the development of anti-obesity drugs.

According to sources within the industry, the major companies with LTB_4 programs are likely to take this report seriously, and they will probably initiate or extend existing programs to see if the newly identified receptor for LTB_4 can be exploited therapeutically. The report by Devchand's team should also prompt groups seeking anti-obesity drugs to turn to the LTB_4 programs for antagonists to test for anti-obesity or general lipid-lowering effects, and it may provide a new spark to existing LTB_4 anti-inflammatory programs that are currently in decline or being phased out.

Histone deacetylase and apicomplexan parasites

Malaria, cryptosporidiosis and toxoplasmosis are major apicomplexan parasitic diseases that are a significant threat to humans. The organisms that cause malaria are now resistant to many of the traditional therapeutic agents, outbreaks of cryptosporidiosis occur even in developed countries and large numbers of immunocompromised AIDS patients are highly susceptible to both toxoplasmosis and cryptosporidiosis. Novel targets are desperately needed in order to discover new drugs to control the human diseases caused by the these parasites. These same organisms also cause numerous diseases in livestock and poultry with a consequent economic toll on their respective industries.

Dr Sandra J. Darkin-Rattray and coworkers (Merck Research Laboratories, Rahway, NJ, USA) have isolated a novel cyclic tetrapeptide metabolite, apicidin, from a fungal sample that was collected in Costa Rica. Apicidin has broad-spectrum activity against the apicomplexan parasites. According to their recent report [*Proc. Natl. Acad. Sci. U. S. A.* (1996) 93, 13143–13147], the new antiprotozoa agent acts on a novel target, histone deacetylase, and it is active against drug-resistant forms of the protozoa parasites that cause malaria.

Histone deacetylase regulates the level of acetylation of histones in eukaryotic cells and thereby plays an important role in the regulation of transcription. Darkin-Rattray's team clearly demonstrates that apicidin increases the level of acetylation of histones from *P. falciparum*, and that the inhibition of histone deacetylase is the likely mechanism by which apicidin blocks the proliferation of the malaria-causing organism. Unfortunately, the naturally occurring compound also blocks the proliferation of human cells, suggesting that it inhibits the human version of histone deacetylase. Clearly, the key to exploiting the new-found compound, or other compounds that act on the same target, will require selective action on protozoa. This report may prompt a careful comparison of the molecular structure of the human and protozoa histone deacetylases in an effort to discover a rationale to design selective agents.

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HTS in *Monitor*

From the April issue, a new regular column on high-throughput screening contributed by Dr Mark Rogers (Group Leader, Cellular Bioassay Design, Glaxo Wellcome, Stevenage, UK) will complement our established features on combinatorial chemistry (Dr Nick Terrett, Head of New Leads Chemistry, Pfizer, Sandwich, UK) and emerging molecular targets (Dr Bob Wallace, Wallace & Associates, New Milford, CT, USA). Guest contributions to the *Profiles* section on any topic relating to drug discovery are also welcomed. Scientists wishing to contribute such articles (approximately 1,000 words) should contact the Monitor Editor, Dr Andrew Lloyd (for contact details, see page 79).