monitor

MOLECULES

Manipulating molecular architecture in parallel

'Credit card' libraries for inhibition of HIV-1 gp41

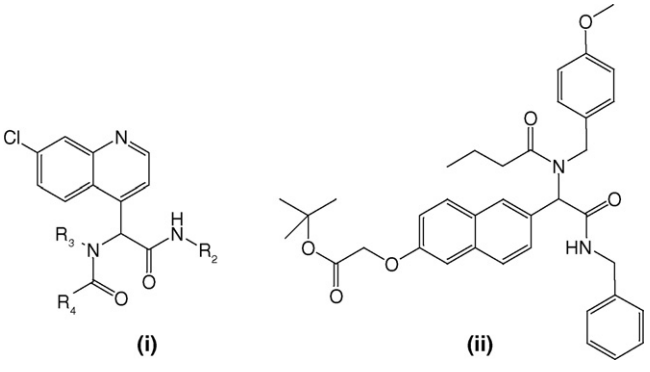
The desire for therapeutic small-molecule inhibitors of HIV-1 infection remains strong. During the past decade, increased knowledge of viral entry mechanisms has facilitated the design of novel clinical tools and marketed drugs that target several entry points in the viral life cycle. One of these approaches has been the design of HIV-1 entry inhibitors. Such molecules can intercept the virus before it invades the cell, unlike most current anti-HIV-1 drugs, which act only after infection occurs. In principle, HIV-1 entry inhibitors could also be used as prophylactic agents to assemble a barrier against the initial infection. The viral transmembrane glycoprotein gp41 is of interest in this regard owing to its role in viral fusion [1].

The HIV-1 virus enters a target cell by fusion of the viral envelope and the cell membrane, followed by release of viral genetic material into the cell. This process is mediated by viral envelope (Env) glycoproteins gp120 and gp41, both derived from precursor protein gp160. Fusion of the virion with the target cell is triggered by gp120 binding to the CD4 cell surface protein and then to one of a group of chemokine co-receptors on CD4+ target cells, such as CCR5 or CXCR4 [2]. This ligand–receptor binding induces a conformational change in gp120 that converts gp41 to its fusogenic form. These conformational changes position the gp41 fusion peptide near the target cell membrane, leading to viral entry. Therefore, the gp41 subunit of the HIV-1 Env glycoprotein represents an interesting target for the development of viral fusion inhibitors.

A gp41 C-terminal peptide, Fuzeon™, is marketed for treating HIV-1 infection. Although the search for peptide-based fusion inhibitors has been successful, the identification of small-molecule HIV viral fusion inhibitors remains a challenge. This search has been given impetus by the observation that small-molecule inhibitors usually exhibit improved pharmacokinetic profiles, oral bioavailability and simpler synthesis scale-up on a manufacturing scale relative to synthetic peptide-based therapeutics. In an effort to discover small-molecule inhibitors targeting gp41 fusogenic activation, Janda and colleagues applied their 'credit card' library approach to gp41 research. Here, this approach aims to disrupt protein–protein interactions of biological relevance. The chemical structures of these libraries are based upon flat rigid scaffolds, decorated with functionalities that span a range of size, polarity, aromaticity and H-bonding capability.

The rationale for the design of the library scaffold is based on the concept of the 'hot spot', - a region in the protein–protein interfaces that is rich in aromatic residues and contributes to the stability and overall quaternary structure [3]. Through the binding of a planar aromatic residue, one might expect disruption of protein–protein interactions. Applying this logic, Janda and colleagues [4] demonstrated that two credit card libraries inhibit the HIV-1 gp41 fusogenic core formation and HIV-1 replication. In the present work, credit card libraries were based upon scaffolds displaying planar, aromatic core structures, such as naphthalene and quinoline. The Ugi four component condensation (4CC) reaction was used to introduce functional diversity to the general α-acylaminoamide core **(i)**, whereas a wide range of structural elements were introduced through variation of size, polarity, aromaticity and H-bonding capability. From this approach, a total of 285 compounds were synthesized in solution as singletons.

One of the most potent compounds isolated was **(ii)**, which had an IC_{50} of 38.26 μM for fusogenic core formation inhibition. Specifically, an ELISA assay was used in which the six-helix bundle(6-HB) formed by N36 and C34 was captured by rabbit polyclonal antibodies and detected by a mouse monoclonal antibody termed NC-1, which specifically recognizes the discontinuous epitopes on this quaternary complex. This assay specifically identifies small-molecule HIV-1 inhibitors that target gp41. Thus, this work has identified a novel series of aromatic-based HIV-1 gp41 fusogenic core formation inhibitors that could serve as a starting point for the clinical development of a potential treatment for HIV-1, and further work in this field is therefore warranted.



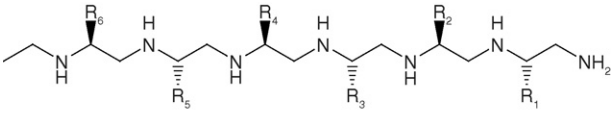
1359-6446/06/$ - see front matter doi:10.1016/j.drudis.2006.12.005

Chiral tri-amine and tetra-amine μ opioid receptors ligands

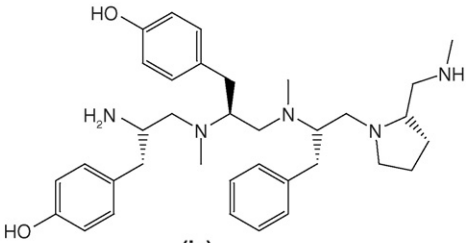
Solid-phase parallel synthesis is used to generate libraries of small organic compounds, often with the intention of accelerating the discovery of new medicines. Much of the chemistry performed on solid-phase is well understood and it is possible to obtain excellent synthetic purity and yields during, for example, the solid-phase synthesis of peptides. The chemistry of amino acids is also well understood and chemical modification of peptide libraries has enabled the generation of peptidomimetic libraries and low molecular weight, small-molecule organic libraries. Peptide libraries have, for example, been successfully modified by exhaustive alkylation of their amide bonds to yield peptidomimetics and/or by reduction of the backbone amide carbonyl groups to yield polyamines.

The amine functionality is a crucial factor in H-bonding and folding of enzymes. Amines are also key components of ligands having specificity for the opioid receptors μ, γ, and κ, for example. The opioid receptors themselves represent an important system that could be used in combination with combinatorial libraries to identify different ligands for related receptors, such as μ, γ, and κ [5]. All three receptors have recently been cloned and belong to the seven-transmembrane G-protein-coupled family of receptors, having ~60% amino acid sequence homology to each other. Screening of the same combinatorial library in separate assays selective for each of the three receptors provides new ligands for these receptors, but also yields insights into the ability of combinatorial libraries to discriminate between closely related receptors.

Recently, Houghten and co-workers reported on the synthesis and screening results of a positional scanning mixture-based library of 34,012,070 chiral hepta-amines [general structure **(iii)**] in a radio-receptor assay for the μ opioid receptor [6]. In this work, a hepta-amine library was generated following exhaustive reduction of a hexapeptide library synthesized from 18 naturally occurring amino acids. For its generation, the methodology relied on the positional scanning of one of each of the single sub-libraries derived from each variable position in the hepta-amine. In the case of a single-position defined positional scanning library, each compound in a given mixture has a common individual building block at a given position, whereas the remaining positions are composed of mixtures of all of the building blocks used in the library. A common single building block defines each relevant mixture. The sub-libraries for each position represent the same collection of individual compounds and they differ only by the location of the defined position. The activity found for a mixture upon screening is caused by the presence of specific active compound(s) within the mixture and not the individual functionalities as separate independent entities.



(iii)



(iv)

The library generated in this way was screened in a μ opioid receptor specific assay. Several active mixtures were identified upon screening. Deconvolution was undertaken to synthesize compounds as single entities that then underwent re-screening this time as single compounds. From this approach, one of the most potent isolated was **(iv)** with an IC_{50} of 13 nM against the μ opioid receptor. This work is of interest because the authors have used mixture-based library generation approaches to generate highly active compounds against the μ opioid receptor. The establishment of further work aimed at generating new SAR within this series is desirable because it could enable the identification of new, potent and selective tri- and tetra-amines that maintain a balanced pharmacokinetic and pharmacodynamic profile.

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