ELSEVIER

feature

Filtering promiscuous compounds in early drug discovery: is it a good idea?

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The use of computational filters for excluding supposedly nonspecific and promiscuous compounds from chemical libraries is a controversial issue, because many drugs used in clinics today would never reach the market if these filters were applied. In part, this conflict could be caused by the paradigm: one-drug–one-target, even though it is widely agreed that drug action is a result of a complex network of biomolecular interactions. Therefore, the so-called pan assay interference compounds (PAINS) or promiscuous compounds could be in fact assay artifacts, false positives or, simply, bright chemical matter (BCM) composed of privileged scaffolds, as we propose here. Despite apparent promiscuity, BCM can be tailored into new and safe drugs after overcoming selectivity criteria.

Introduction

HTS is considered the workhorse for drug discovery in the pharmaceutical industry and its implementation in academia has been increasing over the past decades. The identification of new molecular entities (NMEs) by HTS relies on robust biological assay systems and good quality chemical libraries with diverse chemical scaffolds. There are several ways to improve the quality of chemical libraries for screening campaigns in early steps of drug discovery [1]. One strategy consists of using computational filters, such as ADMET predictors, Lipinski's Rule of Five and others, to predict drug-like features. Some researchers advocate the use of substructure filters to recognize nonspecific or promiscuous compounds, the so-called pan assay interference compounds (PAINS; see Glossary), when screening libraries [2,3]. According to this line of thought, PAINS should be excluded early from drug discovery campaigns because they are frequently found as hits in a variety of target-based HTS assays (four out of six assays in the original work of Baell and Holloway) [2]. This reductionist point of view implies ultimately that truly active and potentially druggable hits must have molecular mechanisms of action (MoA) based on the compound interaction with a single macromolecular target. However, many drugs available in the pharmaceutical market exert their biological activities by acting on multiple molecular targets. Indeed, active drugs with few side effects are much more important characteristics than the identification of molecular targets to reach the pharmaceutical market.

The concept one-drug–one-target is biased by an increasingly outdated paradigm. Currently, disciplines such as systems biology and areas such as polypharmacology already consider the action of a drug in a living organism as an integrated result of a range of perturbations to a complex network of biomolecular interactions [4]. Polypharmacology is supported by growing evidence that clinical effects are often mediated by drug action on multiple targets [5]. This evidence motivated the concept of molecular master keys for a target family with similar structure or functionality [6]. This concept was coined in analogy to the seminal lock and key model, proposed by Erlich in 1894. Furthermore, there is a thin line between the therapeutic and toxic effects of a drug, which reinforces the complexity that underlies the interaction with the biological system [7,8]. The applicability of the PAINS filter should be restricted to libraries used in biochemical assays (molecular target-based), and the latter is just one of the two main drug screening strategies currently adopted. The other strategy, phenotypic-based screening, is steadily increasing in its use by the

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GLOSSARY

Activity cliff unexpected abrupt changes in biological activity as a result of small changes in molecular structure [18].
Bright chemical matter a frequent hitter compound with biological activity in diverse assays and must have specificity improved to be a drug candidate.
Dark chemical matter a nonfrequent hitter (nonpromiscuous) that significantly and unusually lacks biological effects on hundreds of diverse assays [17].
Molecular master keys privileged structures that act in a target (gene) family, sharing structural or functional similarities [6].
PAINS assay artifacts that can mask biological activity in an assay, by acting in a non-drug-like mechanism [2,3].
Privileged structures scaffolds or frameworks that with judicious structural modifications provide useful ligands for more than one biological target, owing to presenting some particular structural properties favorable for the molecular recognition by many different bioreceptors [19–21].

pharmaceutical industry and constitutes a return to a more physiological approach in drug assays, which in the past has yielded the majority of the drugs that are marketed nowadays [9]. Even though it can be argued that the reason for this fact is the relatively recent introduction of target-based approaches in the context of the long timeframes of drug development, the importance of phenotypic screening cannot be minimized [10].

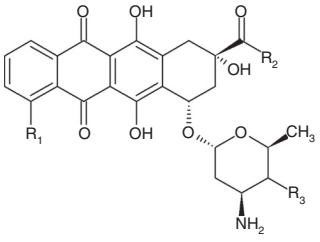
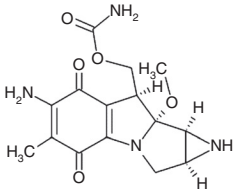
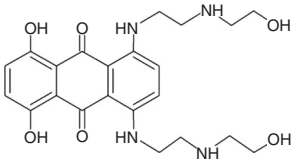
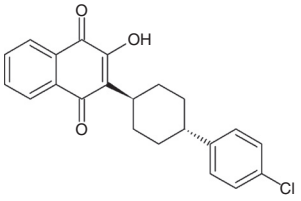
Besides, by providing a more physiological context, phenotypic screening adds value to the drug discovery process by its potential to identify novel biological mechanisms for drug action while decreasing the numbers of false positives. Finally, by monitoring functional endpoints in a target-agnostic manner, phenotypic screening is potentially more effective than target-based approaches in the discovery of NMEs. Target identification and validation is still useful for many aspects of drug development (e.g. structure-based optimization of binding affinities) but it can be undertaken as a parallel process. Computational approaches to investigate the biological and chemical space are growing in the literature proportionally with the increase in the number of biomolecular 3D structures known [5,11]; but the complete topography of the chemical and biological activity spaces is still far from being known. Indeed, methods were developed for virtual screening and visualization of very large *in-silico*-generated databases of over a hundred billion small molecule structures showing the incredible depth of chemical space [12,13]. Furthermore, computational methods to predict molecular binding pockets, at this moment, are not reliable enough to inspire absolute confidence [14]. Therefore, we must take a holistic approach when trying to understand the huge amount of data coming from screening campaigns.

Catch-22 of library filtering

Although the PAINS filter can be of use in biochemical assays, its employment could be highly detrimental if applied to a library to be screened by phenotypic assays. An argument in favor of this identifies examples of compounds used in the clinic today that would be eliminated by this filter (doxorubicin and others, see below). By filtering out PAINS, one is potentially eliminating molecules with higher chance of being hits in the phenotypic screening (privileged structures) and therefore will not be able to optimize them to be more specific (less promiscuous) causing a catch-22 situation. Compound promiscuity might not be desirable but it is almost certainly unavoidable for the discovery of modern drugs. The challenge of medicinal chemistry is not only to improve potency but also to improve selectivity with a tolerated toxicity.

The apparent promiscuity of certain chemical groups does not impair their use in clinics. One typical example involves quinone-based drugs, approved by the FDA and available in the pharmaceutical market (https://www.accessdata.fda.gov/scripts/cder/drugsatfda/) (Table 1). These drugs are prescribed as antineoplastic, immunosuppressant and antiprotozoal agents and have been used in the treatment of different types of cancer (leukemia, breast, lung and stomach), pneumocystis pneumonia and multiple sclerosis (http://www.nlm.nih.gov/medlineplus/). Although the exact MoA of quinones is still unknown it is thought to involve redox reactions, at least in part. Quinone compounds undergo redox cycles *in vivo* generating semiquinone radicals and oxygen reactive

TABLE 1
Quinone-based drugs prescribed in clinics

Drugs	Chemical structure	Clinical use
Anthracyclines (doxorubicin, daunorubicin, epirubicin and idarubicin)		Antineoplastic
Mitomycin		Antineoplastic
Mitoxantrone		Antineoplastic Immunosuppressant
Atovaquone		Antiprotozoal