'A wise use of lead discovery tactics will distinguish successful drug discovery engines.'

editorial

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Critical review of the role of HTS in drug discovery

The past decade has seen the emergence and maturation of HTS. HTS evolved rapidly to enable the exploitation of genomics and combinatorial chemistry in drug discovery. Despite the current emphasis on quality and the growing number of success stories, an initial focus on quantity and speed, together with overly optimistic forecasts made at the start, have led to skepticism about the true value of these new technologies. An analysis of the output of lead discovery by HTS unveils key factors affecting success and points to some areas for improvement in the current drug discovery paradigm.

All practitioners of HTS are aware of the impressive revolution of assay technologies and methodologies in the past decade (for example, see Refs [1,2]) and the central role that HTS now occupies in drug discovery (for example, see Ref. [3]). However, this success is not fully recognized by skeptics who see HTS as the flagship of a failed new paradigm in drug discovery (for example, see Ref. [4]). Let us try to understand these contradictory views by scrutinizing the facts behind them. In the evolution of the field we can delineate two phases, described in the following paragraphs.

Phase 1: exploring a new paradigm – focus on quantity and speed

Starting at the end of the 1980s, up to the late 1990s, novelty and quantity were overemphasized. The HTS process was focused on absorbing the demand for faster and more screening of small-molecule libraries driven by genomics and combinatorial chemistry. Assay well-metrics and throughput were the main measures of success. Novel targets with unprecedented druggability records [5,6] were tested against compound collections being built in great proportions with large, impure and non-drug-like compounds [7]. Automation and assay technologies were rapidly evolving, fueled by strong investments from big pharma.

There was a cause for this influx of money. The existing paradigm for drug discovery was failing and several unsolved and new diseases demanded new ideas. High expectations were placed on the emerging efforts in genomics. These demanded taking a chance in 'engineered serendipity' because rational design approaches could not work for targets of unknown structure and/or unknown natural ligands.

This was a time of exploration of uncharted waters (new targets) and of breaking new ground (combinatorial chemistry, HTS). Coopetition (cooperation among competitors) between biochemists and engineers, vendors and users [fostered by groups such as the Society for Biomolecular Sciences (SBS)] was a key factor for success in establishing several fundamental technologies (high-density microplates, homogeneous assays, high-performance microliter and nanoliter dispensers, imaging, laboratory automation, etc).

Phase 2: perfecting HTS as a drug discovery engine – focus on quality

Combinatorial chemistry was initially driven by numbers but in the late 1990s the emphasis shifted to making single, pure drug-like compounds [8]. Around the same time, HTS specialists began

1359-6446/06/$ - see front matter doi:10.1016/j.drudis.2006.02.001

to focus on quality control measures for assays (for example, see Refs [9,10]). Now the field was poised to make progress.

HTS hype

The early 1990s were also years of hype. The genomic revolution, the power of combinatorial chemistry and ultra-high-throughput automated assay technologies – the new paradigm was oversold as a single panacea for the dearth of new chemical entities (NCEs), within the scientific community and to the outside world. New drugs lacking side effects were apparently expected to be in the pharmacy just a few months after articles were written [11]. The long time scale of drug discovery and development was poorly publicized and understood. Furthermore, the time needed to engineer, refine and successfully establish the new technologies, and to learn how to use them wisely, was never accounted for.

All these facts explain the emergence of skepticism for the HTS paradigm just as we were starting to master our craft (Phase 2). They also allow a more accurate analysis of the success rate of HTS. Clinical candidates from HTS only started entering into pharma's pipelines in the new century, a few years after we learned more about the choice of targets, compounds, technologies and quality control procedures. Because of long cycles in discovery (ca. seven years on average, according to Ref. [12]) and development (ca. eight years on average, according to Ref. [13]), the HTS drugs are still in development. There were 62 candidates from HTS, declared by HTS laboratories responding to a survey conducted in 2002 [14], 74 in a follow up survey from 2003 [15] and 104 in the most recent survey from 2005 (www.hightechdecisions.com/reports.html). These numbers are surely an underestimation because not all laboratories worldwide were interviewed and, based on personal experience, it is often hard to track the fate of HTS hits accurately. Drugs that can be tracked down to this new paradigm for drug discovery have been registered in recent years (e.g. Gleevec®).

The paucity of NCEs reaching the market is worrisome. We are challenged scientifically to understand why and we have the duty to improve our output at every step. The current paradigm has many aspects to it; let us focus on HTS and try to analyze the scientific factors that most influence its output.

Targets

Screening of corporate collections yields a diversity of results depending on the target class [5,15]. In the survey summarized in Ref. [15], 42 HTS laboratories reported success rates in finding leads in the range 8–100% of the targets tried, with an average of 56%.

As shown in Table 1, in the screening history within GlaxoSmithKline (GSK) we can distinguish some trends. Full diversity screening is successful for certain classes [e.g. family A G-protein-coupled receptors (GPCRs) and ion channels], and not so much for others (e.g. family B and family C GPCRs). In addition, current screening strategies are not limited to full diversity HTS but include focused screening, a very successful evolution of HTS (after all most of the chemotypes currently in these sets were found via random screening) for families of targets with narrow chemical connectivity, such as kinases around the ATP-binding pocket [16].

For those targets with a poor record of hit discovery there are two possible causes of failure: the target is not druggable (e.g. most protein–protein interactions) or the compounds are not there. GSK had an experience shortly after the merger of GlaxoWellcome (GW) and SmithKline Beecham (SB), which indicated that for some targets it is simply a matter of not having the right compounds. For the first two years post-merger, some targets were only screened against the GW collection and others were only screened against the SB collection. Once the two collections were merged into a larger, unified GSK collection, some of the unsuccessful targets were rescreened to sample additional diversity. Tractable hits were found for a significant proportion (30%) of the previously unsuccessful targets, suggesting that some targets thought to be less tractable can be solved with the right chemical diversity.

TABLE 1

Success rates in finding tractable hits via full diversity HTS at GSK

Target class		Success rate: HTS to tractable hit (%)
G-protein-coupled receptors (GPCRs)	Family A	54
	Family B	0
	Family C	0
Ion channels		73
Kinases*		7*
Nuclear receptors		72
Other enzymes		50
Other targets		33

Data shown correspond to the period 2001–2004. The tractable hit (often called 'lead' at other companies) is a confirmed hit with activity in a biologically relevant assay with a tractable chemical structure and an initial indication of SAR such that a chemical optimization effort can begin. *Kinase success rate only reflects the contribution of full diversity HTS to identify series not found via focused screening. Overall success rate in finding ATP-competing tractable hits from all screening tactics is 85%.

Compounds

The coverage of the chemical space (even if all compounds ever made or isolated were combined) is very limited. Typically, a drug discovery lab tests 10^5–10^7 compounds versus 10^{40}–10^{100} possible compounds in the small-molecule universe [17]. Therefore, the reported success rates (for example, see Table 1 or Ref. [15]) are indicative of a fantastic enrichment of screening collections with privileged motifs. In other words, despite a poor coverage of the chemical space, screening collections are a good representation of the 'protodrug' space – a smaller section of the chemical space. Nevertheless, we can expand the druggable genome if we find the same thread for other areas of chemical diversity poorly covered in today's corporate collections. Lipinski's rules have had a profound beneficial impact in drug discovery but it is time to exploit the often overlooked fifth rule – the first four rules don't apply to natural products or other compounds that benefit from active transport into target cells or tissues [8].

Natural products were a main source of drugs and a starting point for synthetic drugs until the industry, in the middle of the hype of new technologies in the 1990s, performed a cost–benefit analysis and decided virtually en masse to abandon this approach for lead discovery. New ways of exploiting this diversity (that provides complementary scaffolds not amenable to standard synthetic chemistry) have recently been developed [18].

Furthermore, analysis of previous success and failure is based on our short history of chemical exploration. The journey might be long and difficult once we get into unexplored areas of chemical

space but the value of solving the remaining 50% of biological targets, including many known to be highly validated, is huge. For example, for many targets that transform hydrophilic substrates (e.g. sugars), less-hydrophobic chemicals are likely to be a better source of leads [19].

Alternatively, targets with poor results in HTS but amenable to other lead discovery tools, such as design of transition state analogues or protein structure-based design, should be exploited in addition to or instead of HTS.

Screening platforms

How compounds are screened against targets has been a matter of major discussion. The key goal of HTS as an isolated activity is to discover as many hits from the compound collection as reasonably achievable (ideally all hits would be discovered although practical constraints, such as experimental errors or compound concentrations, among others, impose obvious limitations) and to deliver accurate data packages to guide decision making. This can be done if, and only if, the following three conditions are met:

(i) the assay chosen is predictive in the appropriate sensitivity range of the degree and nature of interaction of an unknown ligand to the protein of interest
(ii) compounds are properly managed and tracked
(iii) screening processes are the subject of proper quality control

It is irrelevant if the plate density is lower or higher, the process is fully automated or purely conducted by hand, or if you trust the screener. Many useless hours have been spent scrutinizing HTS laboratories when the underlying cause of a lack of success lies, for the most part, in the poor druggability of the target or in the lack of the right chemotype in the collection tested.

Conclusion

After the hype of this revolution in drug discovery it is important to review what has worked well and what needs improvement.

Today, the tools at hand for lead discovery are expanding and complementing each other: focused screening for well connected target classes, virtual screening and access to large external chemistry sources (including isolated natural products), structure-based design and cellular assays of increasing value in understanding compound effects. All of these tools are important complementary approaches to HTS for providing entry points for lead optimization.

A wise use of lead discovery tactics will distinguish successful drug discovery engines. Different engines might be able to afford different options in the menu. As long as we keep learning how to best select and validate targets linked with human disease and wisely exploit these recently refined technologies (in addition to and not instead of those previously established) we should expect an increase in drugs in the market that help people to live healthier and longer lives.

Acknowledgements

The author would like to thank Drs Thomas Meek and Robert Hertzberg for their support and mentorship throughout this journey and for their critical review of this critical review. Data were provided by many scientists from GSK, in particular from the HTS groups led by Dr Emilio Diez, Dr Michael Snowden, and myself. Their contributions and those of many other colleagues at GSK are also gratefully acknowledged for inspiring discussions and ideas.

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