INNOVATION

Rescuing drug discovery: *in vivo* systems pathology and systems pharmacology

Jan van der Greef and Robert N. McBurney

Abstract | The pharmaceutical industry is currently beleaguered by close scrutiny from the financial community, regulators and the general public. Productivity, in terms of new drug approvals, has generally been falling for almost a decade and the safety of a number of highly successful drugs has recently been brought into question. Here, we discuss whether taking an *in vivo* systems approach to drug discovery and development could be the paradigm shift that rescues the industry.

Has target-centric drug discovery, practiced intensely by pharmaceutical companies for the past 30 years and recently buttressed by the availability of genomic data, become unproductive to the point where the economic future of the industry is questionable? Several authors have raised this issue[1–3], and indeed the well-publicized general decline of new drug approvals each year in the United States from the high-point reached in 1996 is in sharp contrast with the almost doubling of expenditures on pharmaceutical R&D during the same period[4,5].

With this issue in mind, there is growing interest in the proposition that a systems-based, rather than a target-based, orientation to human diseases and to *in vivo* pharmacology could transform drug discovery and development in a manner that will more cost-efficiently produce the right treatments for the right patients. A recent issue of *Nature Biotechnology*[6] covered a broad spectrum of topics in this area, from visions for the future to opportunities for commercialization of systems biology in drug discovery, and exposed the need for practical tools to move from ideas and future potential towards addressing today's problems and challenges in the pharmaceutical industry. Here, we focus on the importance of *in vivo* studies using a systems approach on the path to transforming the drug discovery and development process from disease diagnosis to the prescribing of drug treatments.

Background to systems thinking

The past century saw the dramatic growth of reductionism in the life sciences based on the development of methods to isolate and study molecules, cells and other components of living systems. Studying the parts of living systems in isolation has substantially advanced our understanding of the nature of life and has led to remarkable societal and economic benefits, including those derived from the development of new medicines. However, during the same period, the nature of life has also increasingly been studied from a systems perspective across different scientific disciplines, partly because of a growing realization of the difficulties of predicting the behaviour of an intact organism from the behaviour of its parts in isolation. The visionary works of von Bertalanffy[7], Sheldrake[8], Capra[9], Laszlo[10] and Rosen[11] constitute an inspirational impetus for a systems approach to biology and medicine, while the articles by Ideker *et al.*[12] and Kitano[13] provided the first practical frameworks for applying 'systems thinking' to human diseases and drug discovery.

The landscape of systems thinking about biology and medicine has been shaped by many contributors who have advanced key concepts, including life's complexity pyramid from individual molecules to interacting subsystems[14]; dynamical disease and the importance of biological rhythms[15,16]; and the robustness and multiple parallel control systems for crucial, fault-intolerant biological processes[17]. As biomedical scientists and the healthcare community embrace systems thinking as central to improving the practice of medicine, these concepts will have a profound influence on strategies and tactics in drug discovery.

Systems thinking in drug discovery

The idealized goal of current drug discovery is to create a single chemical substance that interacts specifically with a single molecular target to perturb *in vivo* biochemistry in a manner that eliminates the biochemical changes that have taken place as a disease takes hold of an organism, and reinstates healthy-state biochemistry. The reality is that some of the most specific drugs are directed towards a target that is not central to the pathophysiology of the disease and simply produce improvements in a limited number of symptoms. Moreover, many drugs designed to interact with a single target have unanticipated effects on 'off target' biochemical mechanisms and the safety implications of those unwanted effects might not be revealed until a drug candidate is in large-scale clinical trials or even on the market. Although this approach has generated highly successful medicines, especially during the past 30 years, there is concern that the target-centric approach might have 'hit the wall' of productivity[18,19] because, despite the massive increase in potential new drug targets for different diseases resulting from genetic and genomic studies from the late 1980s to

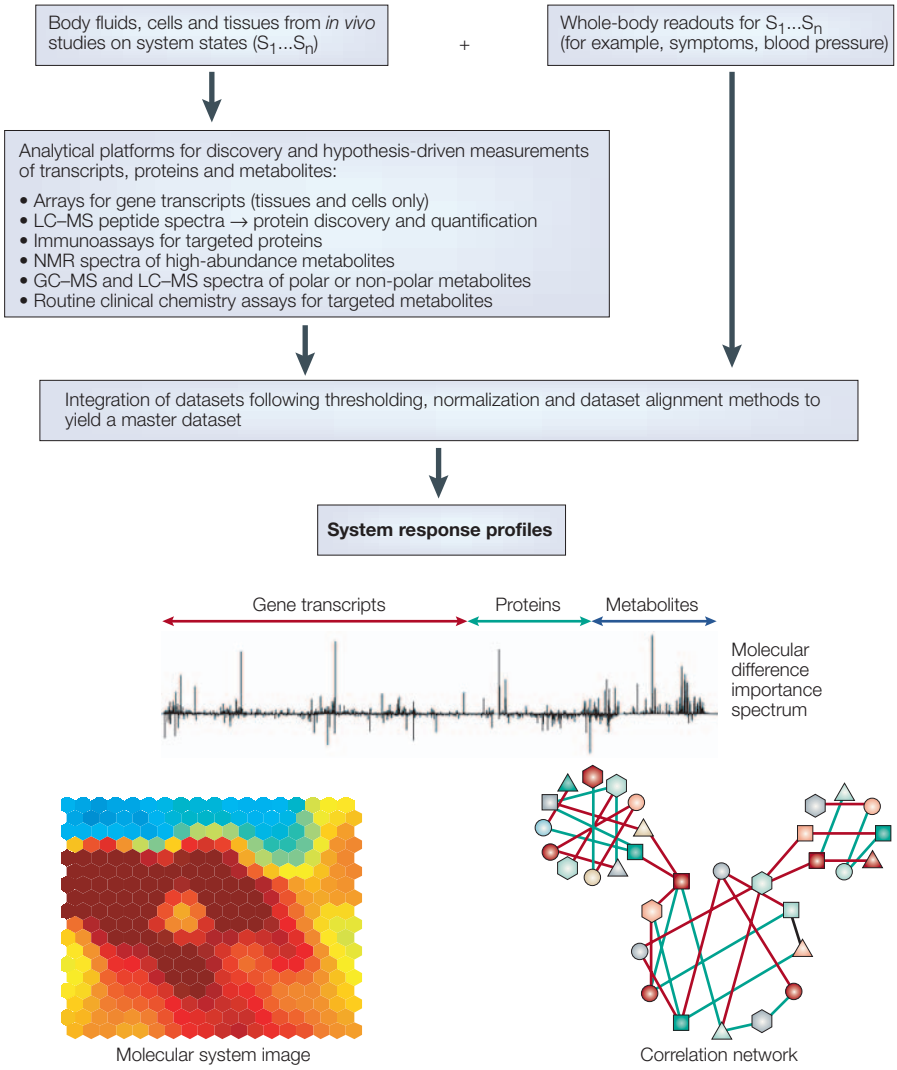

Figure 1 | **Flowchart for systems pathology and systems pharmacology.** An overview of the materials, information and analytical methods that constitute the workflows and outputs of systems pathology and systems pharmacology. Three forms of system response profiles are presented in the lower portion of the figure, each of which highlights a different aspect of the dataset for comparisons between system states, such as drug-perturbed versus unperturbed. A molecular difference importance spectrum, or factor spectrum (see REF. 26 for details), is created from the relative contribution of each individual molecule (length of vertical line) to the separation between two states determined by principal component analysis. The direction of each vertical line indicates whether the change in the molecule between the states was an increase or a decrease. A molecular systems image is a self-organizing map[41,42] created from the dataset and provides a ready colour-coded visualization of levels of molecules and the relationships between molecules in the dataset in state-to-state comparisons. A correlation network[23–26], shown here in a schematic form, provides simultaneous information about the class of molecule (symbol shape), the direction of the change in its level between states (red, higher in the displayed state than in the comparator state; green, lower in the displayed state; white, no change between states) and the associations between pairs of molecules (red line, positive correlation; green line, negative correlation). In the statistical treatment of datasets containing thousands of measurements derived from relatively small numbers of biological samples, care must be taken to avoid false discoveries[43]. LC–MS, liquid chromatography–mass spectrometry; GC–MS, gas chromatography–mass spectrometry.

the present day, and substantial increases in pharmaceutical R&D expenditures, yearly new drug approvals have not increased during this period, even when allowing for the usual time delay between R&D spending and new product launches. So, there is ample justification for an immediate effort to explore strategies for improving the cost-effectiveness of drug discovery, drug development and, ultimately, healthcare delivery. Strategies based on systems thinking represent an enticing departure from current, target-centric activities and offer the promise of opening up an entirely new, more productive, era of drug discovery and also of preventing or effectively dealing with unforeseen problems with marketed drugs following approval.

Cell-based systems biology

In the current target-centric approach to drug discovery, potential hits and leads are identified and optimized for activity against specific molecular targets. It has recently been noted that human cell-based studies with a systems orientation could be a valuable approach for generating hits and leads in a manner that is not dependent on the prior identification and validation of a particular target[1]. *In vitro* studies with a focus on systems dynamics can also provide valuable information about the different pathways through which drugs can produce similar effects[20]. However, as such approaches have been covered comprehensively in the article by Butcher[1], they will not be discussed further here, except to note that the systems-oriented approaches described in the present article could be valuable in revealing the *in vivo* correlates of the responses of *in vitro* cell systems used for lead identification and optimization in cell systems biology. Indeed, because of the lack of background target validation in cell-systems approaches, detailed systems-orientated analyses of animal responses could be particularly valuable for the development of leads generated from human cell-based screens.

Nevertheless, the results 'push' from *in vitro* studies is unlikely to be sufficient to guarantee that drug discovery will successfully leap the chasm of missing information to complex human diseases and multifaceted, system-wide drug responses. The results 'pull' from *in vivo* systems pathology and systems pharmacology studies, which are the focus of this article, will also be essential if new drug discovery is to be rescued.

Systems pathology/pharmacology

A crucial step in a systems approach to drug discovery is to describe diseases not by symptoms but in the molecular language that 'drug hunters' can act on. Additionally, to discover and develop new medicines with improved efficacy and reduced side effects for common multi-factorial, system-wide diseases, such as type 2 diabetes and cardiovascular disease, it is essential to generate comprehensive molecular descriptions of the disease and the responses to drugs so that the breadth of biochemical changes contributing to a disease or drug response can be taken into account.

In this article, 'systems pathology' is used to refer to the body-system-wide, predominantly molecular, characterization of a disease state relative to a healthy state,

and 'systems pharmacology' is used to refer to the same characterization of the drug-perturbed state relative to the unperturbed state. The resultant datasets of largely molecular changes between states of the system (diseased versus healthy or drug-perturbed versus unperturbed) are referred to as 'system response profiles' (SRPs).

The rapid evolution of novel 'omics' tools, biostatistics and bioinformatics during the past decade has made *in vivo* systems pathology and systems pharmacology possible[22]. SRPs can be generated efficiently (and with ever-improving economics) by applying analytical techniques (see below and FIG. 1) to samples of body fluids, cells or tissues obtained from *in vivo* studies. The range of SRPs that can be generated in an investigation of a disease or of a drug response can extend from a dataset created by applying, to a single cell type, a single analytical platform that focuses on a single class of molecules (for example, RNAs or triglycerides) through to a complex dataset created from the analysis of samples from multiple tissues and body fluids with an array of analytical platforms that can capture as many biochemical changes as technically possible. SRPs can reflect the comparison of just two 'stable' states of the system or the dynamics of a transient response to a drug treatment or of the progression of a disease.

Quantitative data from both discovery and hypothesis-driven bioanalytical techniques involving multiple biochemical components from different molecular classes, such as transcripts, proteins and endogenous metabolites, can be integrated to create comprehensive SRPs of system-state differences (FIG. 1). Depending on the intended use, SRPs can be created in many forms (see FIG. 1 for examples), including human-unfriendly but computer-friendly records containing all the data generated for individual system states and state-to-state comparisons; reduced datasets created using statistical methods to find minimal subsets of molecular components that constitute practical biomarkers for classifying samples into specific categories (see later); molecular systems images (MSIs; see FIG. 1), which are a convenient tool for visualizing all the molecular changes associated with a particular state-to-state comparison (for example, disease versus healthy); and correlation networks (CNs; see FIG. 1), which provide specific information about the interconnectivity and interdependency of molecules in an SRP (see, for example, Steuer *et al.*[23,24] for studies in plants, and Clish *et al.*[25] and Oresic *et al.*[26] for CNs associated with a mouse model of atherosclerosis, the ApoE*3-Leiden mouse.

SRPs generated from cells, organs or body fluids support the concept that, at higher levels of complexity, new properties emerge within a system[26,27]. At the level of investigating blood plasma level, for example, SRPs reflect the interactive dynamics of body tissues, providing detailed information on how certain communication and control mechanisms are functioning *in vivo*[17,28]. For multi-factorial diseases, studying cells or tissues in isolation with a systems orientation can be informative, but not to generate information about the organizational level of the entire body[21]. In such diseases, the organizational level is the key level at which to understand the onset of a pathological process — namely, the initial loss of homeostasis within the body[22].

In the remainder of this article, we first expand on the role of systems pathology in drug discovery, and then the role of systems pharmacology, and, finally, highlight the potential for a combination of such systems-oriented approaches to transform each step of the drug discovery and development process.

Drug discovery: systems pathology

Systems pathology is a crucial, but currently under-represented, step on the path to successful drug discovery. For example, SRPs of the disease state relative to a healthy state, in addition to their value in drug-target discovery activities, can provide much-needed information about major biochemical subclasses of a population of patients diagnosed on the basis of symptoms. This information can enable the use of biochemically similar subclasses of patients for drug-target discovery efforts, for those who wish to optimize research in this area, and for creating primary human cell lines for the type of cell-based assays advanced by Butcher and colleagues[1,29]. Diagnostic biomarkers for patient subclasses derived from systems pathology studies also have the potential to solve the riddle of drug 'responders' and 'non-responders' and greatly facilitate the transition from drug discovery to drug development by enabling the right drug (or drug combination) to be developed for the right patient group within a population of patients defined on the basis of disease symptoms.

If drug discovery is to be aligned with the goals of general healthcare, more emphasis is needed on the early detection of diseases and on pharmacological interventions to arrest or reverse disease processes before irreversible pathologies are well established, disease symptoms are clearly evident, the prospects of a cure are low, and the cost of patient care high. The SRPs derived from systems pathology studies contain the molecular information that will enable the crossover from symptom-related research in drug discovery, focusing on late-stage disease processes, to an emphasis on discovering drugs for the initial stages of a disease. Such a re-orientation of disease diagnosis and drug discovery will also probably close the gap between nutritional and pharmaceutical research[30].

For the early detection of disease and to generate datasets that will enable the discovery of drugs for early intervention in disease processes, it will probably be necessary to use standardized system perturbations to uncover the initial loss of homeostatic mechanisms. Such studies would be considered a hybrid of systems pathology and systems pharmacology. A prototype example of such a diagnostic system perturbation is the oral glucose-tolerance test (OGTT), which is useful in revealing the initial stages of type 2 diabetes in the face of normal concentration values for fasting plasma glucose and for plasma insulin. However, in the OGTT currently practiced, the evaluation is typically limited to measuring plasma glucose and insulin as biomarkers, whereas in the context of a systems orientation, the sensitivity and specificity of the read-out could potentially be greatly improved by analysing dynamic SRPs.

Cross-species systems pathology in drug discovery. The performance of promising drug candidates in animal models of human diseases is an early gatekeeper on the path from drug discovery to clinical trials. There are currently few criteria beyond symptoms that can be used to select the best animal models for this vital role in the drug discovery and development process. If a drug candidate passes the test of an inappropriate animal model, it might be doomed to a failure that will probably not be recognized until late-stage Phase II clinical trials, by which time substantial financial capital and human resources will have been invested. We propose that selections of suitable animal models can be made by comparing SRPs from systems pathology studies on a variety of candidate animal models with the SRPs from similar studies on patients. As a general rule, the most convenient SRPs to be compared will be derived from the analyses of available body fluids, preferably blood plasma, which represents the window on disease processes across all body organs and tissues and the disordered blood-borne communication and control systems that are contributing to the disease. In cases in which biochemical subclasses of a patient population have been identified, it might be possible to select different animal models to

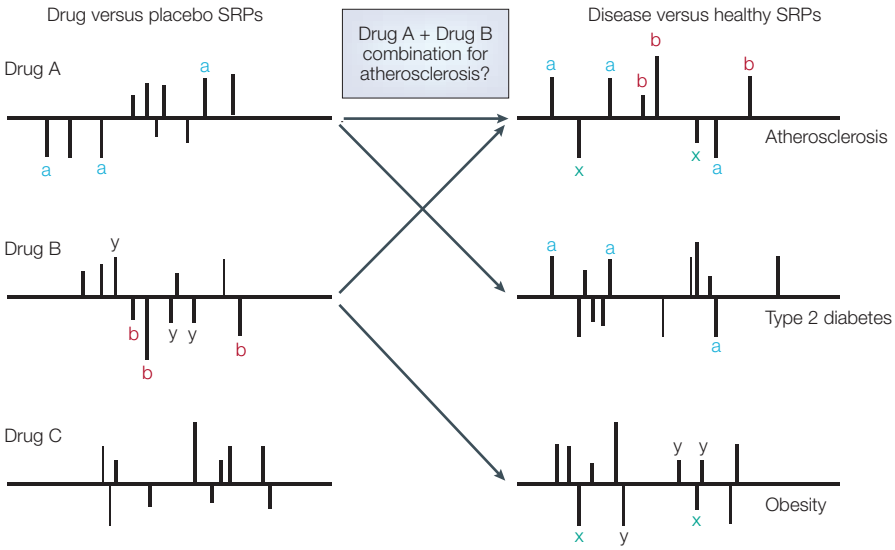

Figure 2 | **Potential use of systems pathology and systems pharmacology to identify potential drug combinations for treating a disease.** Idealized system response profiles (SRPs) in the form of molecular difference importance spectra (FIG. 1) derived from the analysis of plasma samples obtained from healthy subjects for three drugs (each versus placebo) are shown on the left of the figure, and SRPs in the same form derived from the analysis of plasma samples from patients with three diseases (versus healthy subjects) are shown on the right. The arrows connecting drug SRPs to disease SRPs indicate the potential for individual drugs to antagonize a portion of the biochemical changes associated with each of the diseases based on the opposite polarity of certain features of the drug and disease SRPs (contrast with features labelled 'a' in both Drug A and atherosclerosis SRPs or 'y' in both Drug B and obesity SRPs). By inspecting the disease SRPs and the drug response SRPs, it is clear that combining Drug A and Drug B would lead to broader coverage of the biochemical changes that occur in atherosclerosis than either drug alone would generate.

mimic the different subclasses or different stages of the human disease. Furthermore, where approved drugs are already available to treat the human disease, the selection of the best animal models for specific diseases can be further enabled by comparisons of SRPs derived from systems pharmacology studies on the candidate animal models and from drug-treatment studies in patients.

Drug discovery: systems pharmacology

Systems pharmacology is the key to understanding the breadth of drug action *in vivo* and should be a crucial activity on the path to rescuing drug discovery. This novel strategic approach can also open up entirely new paradigms in drug discovery and development, as highlighted in the two examples below.

Comparative reverse systems pharmacology. The current strategy for the discovery of second-generation candidate compounds, in a class of drugs designed to interact with a specific molecular target, is to seek ever-more selective compounds for the target by differential *in vitro* screening of molecules in an array of available 'on-target' and 'off-target' assays. This approach usually produces a few improved follow-on drugs until the areas for additional improvement in drug performance based on the efficacy and side effects of the drugs in patients are found to be unrelated to the drug properties measured in the screening assays. In parallel, or subsequently, a new target for drug discovery soon becomes fashionable and the 'first-in-class followed by improved second-generation drugs' cycle repeats itself until a disconnect is again reached between the effects of the second-generation drug candidates in patients and the early-stage screening assays. This situation arises because beyond the primary and secondary outcome measures, and a handful of conventional vital signs and clinical chemistries assessed in late-stage clinical trials, there is generally no useful information fed back from clinical trials to early-stage drug discovery to aid the process of designing improved drugs.

Systems pharmacology could enable dramatic improvements on marketed drugs of a structural or mechanistic class by establishing a role for SRPs as the system-wide activity measure for chemical structure–activity studies. Features of the SRPs obtained from studies in patients with marketed drugs or late-stage drug candidates could be correlated with efficacy and side-effect measures in the same patients. If the features of the SRPs obtained in patients can also be identified in the best animal model, irrespective of whether the relationship of those features to the disease or drug response can be understood, then drug discoverers will be able to use animal model SRPs that reflect human efficacy and safety as criteria for selecting the next generation of development candidates. Such comparative reverse systems pharmacology would constitute a radical departure from current drug-improvement practices.

Combination drug discovery guided by SRPs. Combination drug therapy has undergone several stages of acceptance and utility in the past, from undesirable to acceptable, and from a compliance perspective to an innovative activity. The commercial benefits of including a blockbuster drug going off patent into a new combination product with extended patent protection are now well recognized. On the other hand, an appreciation of the system-wide nature of diseases and an insight into the regulation of homeostasis via multiple biochemical mechanisms and multi-compartment interactions could unlock the potential for a totally new perspective on the discovery of combination drug products. For example, many of the drug candidates that have failed in clinical development on the basis of limited efficacy, despite clear evidence that their targets have some role in a particular disease mechanism, could be revived in combination with marketed drugs or other failed drug candidates. Similar revival opportunities exist for compounds that have failed due to safety issues that were revealed at the efficacious doses: as components of combination drug products, it might be possible to administer those compounds at doses below the threshold at which the safety issues arose.

Actually, the combination drug concept is very old — it has been the basis for herbal medicines for several thousand years, but mechanistic information for rationally optimizing drug combinations has been very limited. A prime example of how strong synergistic effects can be has been generated in a study of the growth inhibition effects on *Staphylococcus aureus* by berberine[31]. Berberine in combination with 5′-methoxyhydnocarpin, a multi-drug-pump inhibitor that has no significant effect on growth inhibition, has a substantially greater efficacy than berberine alone. In recent studies on ginseng extracts, Sengupta *et al.*[32] convincingly demonstrated the functions of opposing principles in the extracts and provided new insights into the mechanisms of a combinatorial intervention. Of course, the tight compositional control of complex mixtures, such as found in herbal medicines, is a challenge in itself; however, in principle, this challenge

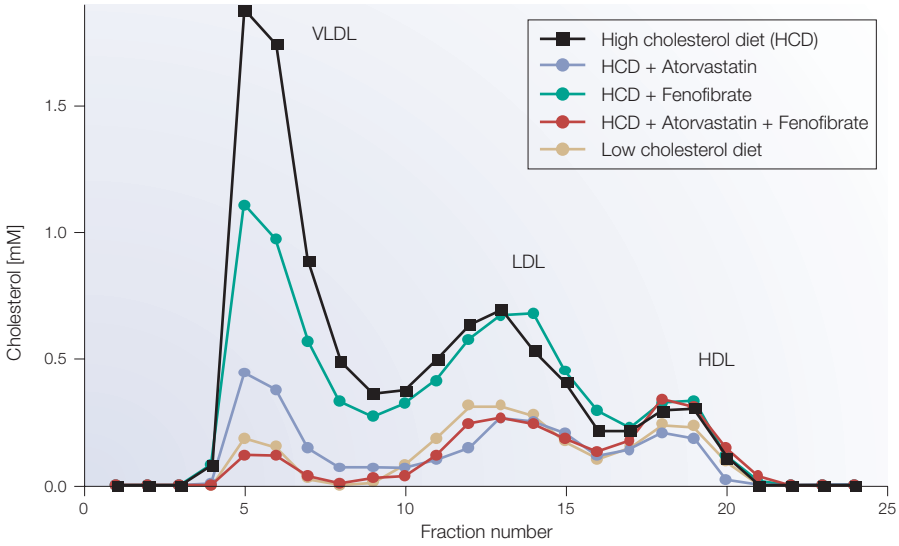

Figure 3 | **Assessing the properties of a system in response to combination drug therapy.** The effects of atorvastatin (0.004% weight/weight (w/w) in diet) and fenofibrate (0.003% w/w in diet) alone and in combination on plasma lipoprotein profiles in the high cholesterol diet, ApoE*3-Leiden mouse model of atherosclerosis. Both atorvastatin (blue symbols and line) and fenofibrate (green symbols and line) lower total cholesterol; however, the combination (red symbols and line), while further lowering very-low-density lipoprotein (VLDL) cholesterol, actually raises high-density lipoprotein (HDL) cholesterol modestly above the level achieved by exposure to atorvastatin alone. See REF. 44 for methods.

can be met from a systems pharmacology perspective[33] using SRPs as a quality-control tool. A randomized, controlled, double-blind, non-inferiority clinical trial for a hypericum extract versus paroxetine for the indication of depression is a good illustration of the performance of such complex mixtures[34]. Furthermore, a recent report suggested that the combination of an anti-T-cell antibody and an insulinotropic hormone could substantially cure type 1 diabetes in an animal model in which the antibody alone produced less than 50% remission of the disease and the hormone alone had essentially no beneficial action on the disease[35].

Beyond the examples above, based largely on natural products, combination drug products for treating HIV/AIDS, cancer, cardiovascular disease, type 2 diabetes and other diseases are now on the market based largely on serendipity or trial-and-error clinical experience. The medical need for rationally designed, combination drug products certainly exists, the precedents are well established, and the scientific and commercial environment is receptive. The challenge is now to design a drug discovery strategy that can generate the product candidates.

Impressive results have been published for drug-combination studies at the cellular level[36,37]. Ultimately, one needs to demonstrate similar impressive results in animal studies using a strategy that is practically realizable. The systems pathology and systems pharmacology approach in combination offer this opportunity. Drug-induced SRPs can be used to reveal unknown mechanisms of action, as experimentally demonstrated by Tas[16] and discussed previously as a viable approach[30].

The essential elements for combination drug discovery guided by SRPs are knowledge of SRPs for many human diseases; the availability of SRP-qualified animal models; and SRPs for compounds in control animals. The comparison of a disease SRP with the SRP for a drug will reveal, at a molecular level, the 'unmet need' in the disease profile and the 'unwanted effects' in the drug-response profile. The biochemical pathways related to the 'unmet need' and 'unwanted effects' are the starting point for exploring ways to improve the activity spectrum of compounds in drug discovery or to combine drugs according to their molecular response profiles to achieve more 'coverage' of the biochemical mechanisms contributing to the disease, or to eliminate the undesirable actions of a single drug. FIGURE 2 illustrates an approach to discovering candidate combination drug products that achieve more coverage of the biochemical mechanisms contributing to a disease.

The approach outlined in FIG. 2 does not take into account possible emergent properties of the system response to the combination drug therapy that are not predicted from the simple linear addition of the responses to the individual drugs. Such an emergent property is exemplified in FIG. 3 for a study performed with hypolipidaemic drugs in monotherapy and combined therapy on the regression of atherosclerosis in the ApoE*3-Leiden transgenic mouse. FIGURE 3 illustrates the improved reduction of plasma cholesterol levels by a drug combination (atorvastatin plus fenofibrate) based on previous established SRPs for the disease and the effects of the individual drugs. However, in addition to the improved reduction of cholesterol generated by the combination, an emergent, beneficial effect is observed on the ratio between very-low-density lipoprotein cholesterol and high-density lipoprotein cholesterol.

Impact and cost-effectiveness

Systems pathology and systems pharmacology, although poised to substantially impact drug discovery as outlined above, have the potential to affect every stage of the drug discovery and development process (TABLE 1). If the vision of a molecular systems re-orientation of drug discovery and development is realized, a number of things will follow. Diseases will be diagnosed earlier and more precisely than is possible by symptoms. Preclinical toxicology will be facilitated by the knowledge of system-wide biochemical changes induced by drugs, which might not be immediately associated with pathologies but might provide clues to prevent or deal effectively with unanticipated adverse events later in drug development. Phase I clinical studies will be improved because biomarkers will be available to assess drug action on volunteers for comparison with preclinical efficacy and safety studies. Phase II and Phase III clinical studies will be enabled by biomarker criteria that can be used to select the most appropriate patients for inclusion in a trial and to monitor the system-wide biochemical impact of drug treatments, especially when a Phase II trial cannot be designed so that definitive outcome measures can be used in dose-ranging studies to find the most appropriate dosing regimen for a pivotal clinical trial. Finally, following approval, all the SRPs generated in the entire drug discovery and development programme will be available to assist in the interpretation and resolution of unanticipated severe adverse events that might arise when thousands of patients are exposed to the marketed drug.

With all this potential for a major impact of systems pathology and systems pharmacology on the productivity of the drug discovery process, why isn't there more activity in this area within the pharmaceutical industry and what will be the cost–benefit reality?

With regard to the uptake of some of these concepts within the industry, there is

Table 1 | **Systems pathology/pharmacology solutions to potential problems in pharmaceutical R&D**

Stage in pharmaceutical value chain	An example problem	Systems pathology/pharmacology solution
Disease diagnosis	Symptom-driven diagnoses identify late-stage disease and biochemically diverse patients	Biochemical detection of disease enables early and appropriate drug treatment
Target discovery and validation	Difficulty of selecting the most appropriate drug target based on genetic or genomic studies	Broad-spectrum information on disease biochemistry enables multi-target approach
Drug discovery and optimization	Difficulty of confidently selecting an animal model	Comparison of SRPs for disease and animal models enables confident selection
Preclinical development	Preclinical efficacy and safety studies do not fully enable translation to clinical trials	Development compound SRPs biomarkers to translate efficacy and safety into man
Clinical trials	Symptom-based selection of patients for trials leads to unpredictable drug response	Selection of patients based on systems pathology SRPs lowers non-response rates
Marketing and prescribing	Unanticipated and unexplained rare AEs lead to drug withdrawal	SRPs for AE patients provide predictive biomarkers and market rescue information

AE, adverse event; SRP, system response profile.

certainly activity in the systems-biology area within pharmaceutical companies, especially in the areas of target validation and biomarkers[38]. However, despite the call from the FDA in the 'Critical Path Initiative'[39] for an increase in science-based drug development and the recognition within the industry of the productivity challenges, there is only sporadic activity directly in line with the systems thinking presented here. Proof-of-principle studies are currently being undertaken, but no critical path success has yet emerged.

From a cost–benefit point of view, it would be inappropriate to give the impression that the full incorporation of systems thinking into the pharmaceutical value chain will provide immediate cost savings. The implementation of such a new concept over the entire process can only take place gradually, given the existing infrastructures that might need to be changed, the current development pipelines and the regulatory constraints. The analytical platforms necessary to undertake systems pathology and systems pharmacology are not inexpensive, nor trivial, to implement. Furthermore, the effort to establish quality-controlled methods to acquire, store, integrate and interpret datasets from different analytical platforms is substantial and the task of obtaining high-quality biological samples from animal and clinical studies is time- and human-resource consuming.

It is probable that the real impact of the systems-based approach will start at the late-stage development or the market end of the process and move towards compound discovery. An example of a late-stage impact would be the use of biomarkers to select subclasses of probable responders for drug-rescue programmes with compounds that failed to achieve efficacy endpoints in clinical trials with heterogeneous patient populations using responder versus non-responder evaluation in the clinical stage. Moreover, combination drug strategies built around disease SRPs and SRPs for existing drugs might be fast and attractive endeavours with an unprecedented development speed and reward structure. Furthermore, the use of SRPs to validate animal models through cross-species studies will create major assets for all drug discovery programmes to follow in the same disease domains.

Incorporating the systems approach across the whole drug discovery process will, in the short term, substantially increase the cost of any particular drug discovery and development programme. The cost-effectiveness to pharmaceutical companies of incorporating the systems approaches outlined here will ultimately be derived from the productivity of their pipelines, because the majority of the expenditures rolled into the average cost of each successful drug approval comes from the failed projects[40]. Rescuing drug discovery and development is initially more about changing the cost to the industry of failed programmes than lowering the cost of successful projects. Leveraging the results from the first steps in entering the era of high-quality systems-based medicine will be of crucial importance. If, with partial incorporation of a systems approach reflected by increasing R&D expenditures for each drug discovery project of, say, 20%, a pharmaceutical company could guarantee a 20% reduction in the incidence of projects that fail in late-stage clinical development, the cost-effectiveness and the societal benefits would be very substantial because, currently, only 8% of compounds entering preclinical development reach the market[40]. In the current business environment worldwide, it will take bold decision-makers in the pharmaceutical industry to increase R&D costs at the expense of a company's bottom line in order to increase the cost-effectiveness of healthcare worldwide and the commercial success of the company over a longer period.

Finally, we assert that the value of systems pathology and systems pharmacology can only be fully realized by combining the results of discovery science and hypothesis-driven science (respectively, the pursuit of the previously unknown and the targeted measurement of the known based on presumed participation in a particular process). Ideker *et al.*[12] propose that the integration of these two scientific approaches is one of the "mandates of systems biology". Realizing the full benefits of a systems approach to drug discovery and development might take 10 years, given the infrastructures to be changed and the need to complete ongoing programmes. So, the likely way forward is stepwise implementation based on business-driven opportunities from the clinic to discovery coupled with improvements in aspects of the process that can have widespread benefits across different therapeutic areas. Systems-based approaches provide new flexible steps into the future for improving the efficiency of drug discovery.

Jan van der Greef is at TNO Systems Biology, Netherlands Organization for Applied Scientific Research, Utrechtseweg 48, PO BOX 360, 3700 AJ Zeist, The Netherlands; Centre for Medical Systems Biology/Leiden Amsterdam Centre for Drug Research, Einsteinweg 55, PO BOX 9502, 2300 RA Leiden, The Netherlands; and BG Medicine, Inc., 610 North Lincoln Street, Waltham, Massachusetts 02451, USA.
Robert N. McBurney is at BG Medicine, Inc., 610N Lincoln Street, Waltham, Massachusetts 02451, USA.
Correspondence to J.v.d.G. and R.N.McB.
e-mails: vanderGreef@voeding.tno.nl; RMcBurney@bg-medicine.com
doi:10.1038/nrd1904

1. Butcher, E. C. Can systems biology rescue drug discovery? *Nature Rev. Drug Discov.* **4**, 461–467 (2005).
2. Grabowski, H. Are the economics of pharmaceutical research and development changing?: productivity, patents and political pressures. *Pharmacoeconomics* **22** (Suppl. 2), 15–24 (2004).
3. Booth, B. & Zemmel, R. Prospects for productivity. *Nature Rev. Drug Discov.* **3**, 451–456 (2004).
4. CenterWatch. Drugs approved by the FDA (2005) [online], <http://www.centerwatch.com/patient/drugs/druglist.html> (2005).
5. Pharmaceutical Research and Manufacturers of America. *Pharmaceutical Industry Profile 2005* [online] <http://www.phrma.org/publications/publications//2005-3-17.1143.pdf> (PhRMA, Washington DC, 2005).
6. *Nature Biotechnol.* [whole issue] **22** (October, 2004).
7. von Bertalanffy, L. *General Systems Theory: Foundations, Development, Application* (George Braziller, New York, 1968).
8. Sheldrake, R. *The Rebirth of Nature* (Bantam Books, New Jersey, 1991).
9. Capra, F. *The Web of Life* (Anchor Books, New York, 1996).
10. Laszlo, E. *The Systems View of the World: A Holistic View of Our Time* (Hampton, New Jersey, 1996).
11. Rosen, R. *Life Itself* (Columbia Univ. Press, New York, 1991).
12. Ideker, T., Galitski, T. & Hood, L. A new approach to decoding life: systems biology. *Annu. Rev. Genomics Hum. Genet.* **2**, 343–372 (2001).
13. Kitano, H. Computational systems biology. *Nature* **421**, 573 (2002).
14. Oltvai, Z. N. & Barabasi, A. L. Systems biology: life's complexity pyramid. *Science* **298**, 763–764 (2002).
15. Glass, L. & Mackey, M. C. *From Clocks to Chaos: The Rhythms of Life* (Princeton, New Jersey, 1998).
16. Tas, A. C. *et al*. Pyrolysis-direct chemical ionization mass spectrometry of the dimorphic fungus *Candida albicans* and the polymorphic fungus *Ophiostoma ulmi*. *J. Anal. Appl. Pyrol.* **14**, 309–321 (1989).
17. Kitano, H. *et al*. Metabolic syndrome and robustness tradeoffs. *Diabetes* **53** (Suppl. 3), S6–S15 (2004).
18. US FDA. FDA Drug and Device Product Approvals [online], <http://www.fda.gov/cder/da/ddpa.htm> (1996).
19. Nightingale, P. & Martin, P. The myth of the biotech revolution. *Trends Biotechnol.* **22**, 564–569 (2004).
20. Huang, S., Eichler, G., Bar-Yam, Y. & Ingber, D. Cell fates as high-dimensional attractor states of a complex gene regulatory network. *Phys. Rev. Lett.* **94**, 128701 (2005).
21. Morel, N. M. *et al*. Primer on medical genomics. Part XIV: Introduction to systems biology — a new approach to understanding disease and treatment. *Mayo Clin. Proc.* **79**, 651–658 (2004).
22. van der Greef, J., Stroobant, P. & van der Heijden, R. The role of analytical sciences in medical systems biology. *Curr. Opin. Chem. Biol.* **8**, 559–565 (2004).
23. Steuer, R., Kurths, J., Fiehn, O. & Weckwerth, W. Interpreting correlations in metabolomic networks. *Biochem. Soc. Trans.* **31**, 1476–1478 (2003).
24. Steuer, R., Kurths, J., Fiehn, O. & Weckwerth, W. Observing and interpreting correlations in metabolomic networks. *Bioinformatics* **19**, 1019–1026 (2003).
25. Clish, C. B. *et al*. Integrative biological analysis of the APOE*3-Leiden transgenic mouse. *OMICS* **1**, 3–13 (2004).
26. Oresic, M. *et al*. Phenotype characterization using integrated gene transcript, protein and metabolite profiling. *Appl. Bioinformatics* **3**, 205–217 (2004).
27. Bhalla, U. S. & Iyengar, R. Emergent properties of networks of biological signaling pathways. *Science* **283**, 381–387 (1999).
28. Davidov, E. *et al*. Methods for the differential integrative omic analysis of plasma from a transgenic disease animal model. *OMICS* **8**, 267–288 (2004).
29. Butcher, E. C., Berg, E. L. & Kunkel, E. J. Systems biology in drug discovery. *Nature Biotechnol.* **22**, 1253–1259 (2004).
30. van der Greef, J. *et al*. in *Metabolic Profiling: Its Role in Biomarker Discovery and Gene Function Analysis* (eds Harrigan, G. G. & Goodacre, R.) 170–198 (Kluwer Academic, Boston, 2003).
31. Stermitz, F. R., Lorenz, P., Tawara, J. N., Zenewicz, L. A. & Lewis, K. Synergy in a medicinal plant: antimicrobial action of berberine potentiated by 5′-methoxyhydnocarpin, a multidrug pump inhibitor. *Proc. Natl Acad. Sci. USA* **97**, 1433–1437 (2000).
32. Sengupta, S. *et al*. Modulating angiogenesis: the yin and the yang in gensing. *Circulation* **110**, 1219–1225 (2004).
33. Wang, M. *et al*. Metabolomics in the context of systems biology: bridging traditional Chinese medicine and molecular pharmacology. *J. Phytother Res.* **19**, 173–182 (2005).
34. Szegedi, A., Kohnen, R., Dienel, A. & Kieser, M. Acute treatment of moderate to severe depression with hypericum extract WS 5570 (St John's wort): randomised controlled double blind non-inferiority trial versus paroxetine. *BMJ* **330**, 503–507 (2005).
35. Ogawa, N., List, J. F., Habener, J. F. & Maki, T. Cure of overt diabetes in NOD mice by transient treatment with anti-lymphocyte serum and exendin-4. *Diabetes* **53**, 1700–1705 (2004).
36. Borisy, A. A. *et al*. Systematic discovery of multicomponent therapeutics. *Proc. Natl Acad. Sci. USA* **100**, 7977–7982 (2003).
37. Keith, C. T., Borisy, A. A. & Stockwell, B. R. Multicomponent therapeutics for networked systems. *Nature Rev. Drug Discov.* **4**, 71–78 (2005).
38. Henry, C. M. Systems biology. *C&E News* **83**, 47–55 (2005).
39. US Department of Human and Health Services. Food and Drug Administration. *Innovation or Stagnation, Challenge and Opportunity on the Critical Path to New Medicinal Products* [online], <http://www.fda.gov/oc/initiatives/criticalpath/whitepaper.html> (2004).
40. Bain & Co. *Rebuilding Big Pharma's Buisness Model* (Bain & Co., Boston, 2003).
41. Kohonen, T. *Self-Organizing Maps* 3rd edn (Springer, Berlin, 2001).
42. Tamayo, P. *et al*. Interpreting patterns of gene expression with self-organizing maps: Methods and application to hematopoietic differentiation. *Proc. Natl Acad. Sci. USA* **96**, 2907–2912 (1999).
43. Benjamini, Y. & Hochberg, Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J. Roy. Stat. Soc. B.* **57**, 289–300 (1995).
44. Delsing, D. J. *et al*. Rosuvastatin reduces plasma lipids by inhibiting VLDL production and enhancing hepatobiliary lipid excretion in ApoE*3-Leiden mice. *J. Cardiovasc. Pharmacol.* **45**, 53–60 (2005).

Acknowledgements

The authors thank A. Adourian and P. Stroobant for helpful suggestions on the manuscript and for assistance with the figures.

Competing interests statement

The authors declare competing financial interests: see Web version for details.

Online links

FURTHER INFORMATION

Systems Biology: http://www.systemsbiology.org
SBI: http://www.sbi.jp
BG Medicine: http://www.bg-medicine.com
Access to this interactive links box is free online.

OPINION

NK_3 receptor antagonists: the next generation of antipsychotics?

Will Spooren, Claus Riemer and Herbert Meltzer

Abstract | Although current antipsychotic drugs are effective at treating the psychotic (positive) symptoms of schizophrenia, they have one or more serious side effects, including extrapyramidal symptoms, weight gain, cardiovascular liabilities and type II diabetes. However, recent data from clinical trials of selective neurokinin 3 (NK_3) receptor antagonists in schizophrenia — osanetant and talnetant — have shown significant improvement in positive symptoms, with no major side-effects reported as yet. Here we discuss the preclinical and clinical evidence that indicates that NK_3 receptor antagonists might represent a new approach to the treatment of schizophrenia and possibly other neuropsychiatric disorders.

Schizophrenia is a severe, disabling and lifelong condition that affects 1% of the population. It is traditionally characterized by positive (psychotic) symptoms, such as delusions, hallucinations and paranoia, and negative symptoms, such as anhedonia, anergia, avolition, flat affect and loss of spontaneity. However, cognitive impairment (for example, attention deficits, working memory deficits and deficits in executive function) is now also recognized as a key hallmark of the disease[1,2]. The aetiology of schizophrenia is not known, but it is generally accepted that both genetic and environmental factors are important in the development and clinical manifestation of this disorder[1].

A range of pharmacological treatments are now available that are relatively effective in providing symptomatic relief. Although diverse in nature and chemical structure, all currently approved antipsychotic drugs share the trait of reducing dopaminergic function by at least two mechanisms — either dopamine D_2 receptor antagonism or partial agonism[3]. Indeed, it has been hypothesized that dopamine has a key role in schizophrenia[4] because stimulants such as amphetamine that enhance the availability of dopamine in the limbic system induce paranoid psychoses, and in light of the relationship between blockade of D_2 receptors by antipsychotic drugs (for example, haloperidol and chlorpromazine) and average clinical dose[4].

First-generation antipsychotic drugs are most effective in improving positive symptoms, but the newer agents, which are called atypical antipsychotic drugs because of their diminished extrapyramidal side effects, are effective in treating some components of cognition deficits and negative symptoms.