lenge, both for experiment and theory. A central question is what the right level of description is when constructing quantitative models of large or even systemwide genetic networks (see the figure). Is coarse-graining of genetic network models possible?

A number of general building blocks identified in genetic networks at least indicate that robust simplified models are possible. Modules such as autoregulatory excitatory (positive) feedback loops (which can convert a transient signal into a sustained signal and thus serve as "storage" devices), inhibitory feedback loops (which suppress instability due to noise), or feed-forward loops (which may enhance responsiveness of a gene) represent different kinds of robust switching elements. Brandman *et al.* describe another such building block—the dual positive-feedback loop, which is frequently found in subnetworks of larger cellular and genetic networks. But why would cells have evolved two positive feedback loops when one is enough to create a switch? Brandman *et al.* find that the combination of the two loops can make genetic switching faster and, at the same time, reduce signal noise. A slow loop creates robustness in the signal, whereas a fast loop allows for switching speed. Given the quite complex cellular machinery that is needed to run this dual positive feedback circuit with biochemical means, its dynamic behavior is intriguingly simple. It functions as a particularly robust, yet fast switch that is reminiscent of the robustly designed electronic building blocks used to build modern computers.

This observation provides support for discrete models of genetic networks in which genes are modeled as switchlike dynamic elements that are either ON or OFF. The first such models, generated about 36 years ago, were random networks of discrete dynamical elements, as few data about regulatory genetic networks were available at the time (*3*). These models were long considered to be merely a speculative analogy. However, recent advances in modeling combined with the first opportunities to validate genetic network models with data from living cells show that simplified network models, such as those representing a regulatory gene as a binary (ON/OFF) switch, can indeed predict the overall dynamical trajectory of a biological genetic circuit. For example, the trajectory of the segment polarity network in the fly *Drosophila melanogaster* has been predicted solely on the basis of discrete binary model genes (*4*). Similarly, a dynamic binary model of the genetic network that controls the yeast cell cycle was constructed (*5*). In both systems, the dynamics converge to so-called attractors (states or

sequences of states of the genes) and for these, the models match the biological dynamics. These dynamical attractors seem to depend not so much on the details of the kinetic constants, as on the circuit wiring. Insensitivity to biochemical kinetic parameters indicates that for understanding the dynamics of these circuits, it's their wiring that is most important (*6*). This seems to be why large genetic networks can be represented as networks of discrete dynamic elements, without the tuning of parameters. Simplified models on even larger scales are encouraged.

Modeling of large cellular networks is often hampered by incomplete knowledge of the full circuitry, despite a wealth of data. An example of how simplification of the dynamics of single elements enables us to gain valuable information about a system's function is presented in the recent article by Ma'ayam *et al.* (*7*). Here, discrete "pseudo-dynamics" of binary states simply percolate through the known part of a 1500-node mammalian cellular network and give a rough but informative estimate of the property of the regulatory information flow through the system. The thousands of parameters required to generate a standard differential equations model of all the relevant biochemical interactions has been neglected here in favor of a statistical perspective that provides valuable information about the global architecture of a cellular network. It is not a direct representation of

the biochemical dynamics and does not allow a detailed dynamic simulation of the network. However, it is an analog of the potential propagation of a signal and therefore useful to determine the global signaling structure of an overall network. This approach is error tolerant and gives a robust picture of the overall global modular structure of a network.

The simple dynamics of the building blocks points to an interesting perspective for our further understanding of genetic networks. Distinguishing between the robust effective dynamics of a genetic or regulatory switch and the biochemical means to practically run it shows that, to understand the system, we do not have to retrace all the details of the biochemistry. Characterizing the circuit wiring seems to be the most important consideration, and when going "dynamic," a clever way to throw away details may be the most important part of model building.

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CHEMISTRY

The Renaissance of Natural Products as Drug Candidates

Ian Paterson and Edward A. Anderson

Around half of the drugs currently in clinical use are of natural product origin (*1*, *2*). Despite this statistic, pharmaceutical companies have embraced the era of combinatorial chemistry, neglecting the development of natural products as potential drug candidates in favor of high-throughput synthesis of large compound libraries (*3*). Perhaps it is time to reassess this prevailing dogma for chasing quantity over quality.

Cancer chemotherapy, in particular, presents an ideal opportunity for natural product–inspired drug discovery and development. Unfortunately, many of the most

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promising natural lead compounds are available only in extremely small quantities, especially those from marine organisms such as sponges. The reluctance of industry to pursue such bioactive natural products as potential drugs lies primarily in the perceived supply problem. This leaves organic synthesis as a key option for sourcing these important drug candidates for preclinical and clinical studies. However, the academic-style approach to "hot target molecules" usually results in lengthy synthetic routes owing to their often exquisitely complicated architectures, with long development times, low overall yields, and impracticality of scale-up and provision of diverse structural analogs.

An alternative approach to drug discovery, which has been embraced by the phar-

maceutical industry, lies in combinatorial chemistry and diversity-oriented synthesis (*4*). This method offers access to a pre-selected range of fairly structurally diverse molecules based around a common core, providing large compound libraries in a short time. These in turn fuel high-throughput biological assays, which have become possible through advances in biotechnology and automation. The problem with this approach lies in the relatively low hit rate of these libraries, relative to natural products, and the potential for undesired side effects due to the often less specific binding characteristics of many of these rather simple molecules.
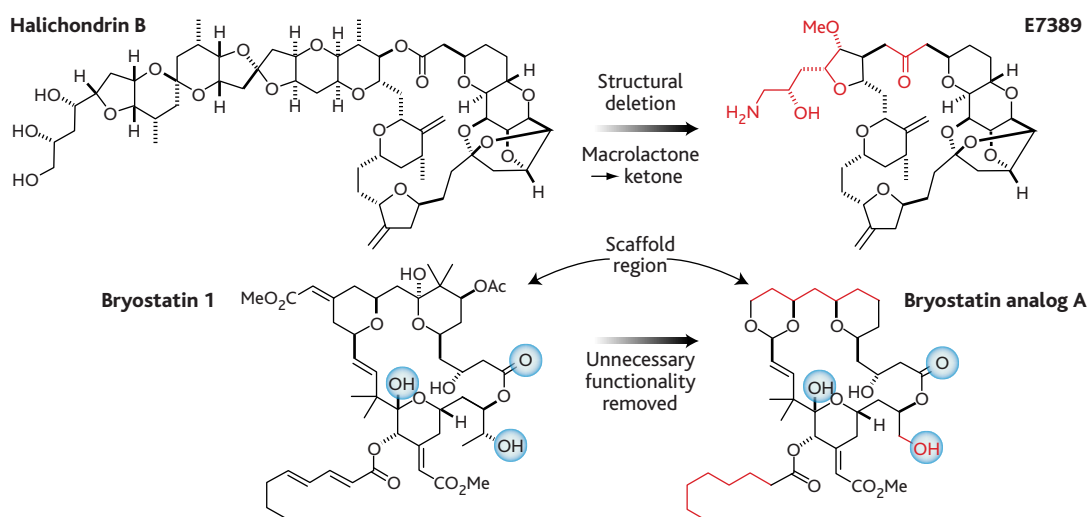
Why do natural products possess such extraordinary specificity and potency compared to artificially designed molecules? The answer lies in evolutionary selection—nature's own high-throughput screening process for the optimization of biologically active compounds. Natural products tend to possess well-defined three-dimensional structures, embellished with functional groups (providing hydrogen bond acceptor/donors, etc.), which have been fine-tuned into a precise spatial orientation. Additionally, the structures of the biological targets of such natural products (e.g., protein binding sites) are often well conserved among proteins of markedly different genetic sequences (*5*, *6*), such that secondary metabolites that have evolved for a certain purpose and mode of action by a producing organism may exert different, yet equally potent, effects in other settings. This leaves open the question of whether further fine-tuning might increase the potency of what really corresponds to a highly advanced lead compound. The preparation

of natural product analogs, which are themselves not naturally occurring, may allow humans to tailor and enhance the druglike properties (bioactivity, pharma-cokinetics, solubility, etc.) of the medi-cines that nature has provided.

Structural modification of natural prod-ucts can be approached in several ways. The first and possibly simplest (although least diverse) is to chemically modify the natural product itself by simple functional-group transformations. This semisynthetic approach has the benefit of providing analogs rapidly, but it is fairly limited in terms of variety. Another possibility is to use genetic engineering to reconstruct biosynthetic pathways (combinatorial biosynthesis) leading to natural product–like structures, but again this has limitations with respect to the extent of possible modi-fication and diversity. Arguably the most versatile approach to analog preparation is the design of a synthetic path to a given nat-ural product that allows for the introduction of deep-seated structural variations en route to the targeted molecule, so-called diverted total synthesis (*7*).

A fine example of a natural pro-duct–inspired drug candidate with its devel-opmental roots in total synthesis is the potent oncolytic (cancer cell–killing) agent E7389 (see the figure), currently in phase I clinical trials. E7389 arose from extensive studies toward the total synthesis of hali-chondrin B (*8*), a highly cytotoxic and com-plex marine natural product. Zheng *et al.* modified the existing route to halichondrin B for analog synthesis (*9*) and discovered that deletion of a large region of the mole-cule did not adversely effect its antimitotic properties. Furthermore, increased stability

in vivo was realized by replacing the readily cleaved lactone linkage with a ketone. The overall yield for the preparation of E7389 was around 1%; however, as with other highly potent and low-dosage drugs, multi-gram quantities should be sufficient for clinical development.

A key point from this exercise is that the analog is easier to synthesize than the parent natural product. In fact, this is one of the main aims of analog synthesis—the design of an unnatural relative that maintains or even improves biological activity, while removing unnecessary molecular complex-ity. The implications from a commercial and practical scale-up viewpoint are obvious. A further compelling illustration of the power of natural product–inspired drug design, embodying the concept of structural simplification, is the development of bryo-statin analogs by Wender *et al.* (*10*). Consideration of the mode of action led to a hypothesis that a large part of the bryostatin structure acted as a framework to position three oxygen atoms (highlighted in blue in the figure), within the pharma-cophore region, in a certain orientation for binding to the target protein. Simplifi-cation of the so-called scaf-fold region led to analog A, which was found to be more potent than bryostatin (cur-rently in phase II clinical tri-als as an anticancer agent) in in vitro assays, yet could be prepared by a sufficiently practical route for consider-ation for large-scale synthesis, which would be extremely challenging for the natural bryostatin structure.

The microtubule-stabilizing agent dis-codermolide is available only in minute amounts from its natural origin (a marine sponge). Through the continued evolution and optimization of synthetic strategies, discodermolide has been prepared by increasingly practical routes (*11*, *12*). Moreover, recent reports highlight the ease with which certain analogs, which would not be accessible by direct modification of discodermolide (i.e., semisynthesis) but are again more potent than the parent natu-ral product in preliminary biological assays, can be synthesized efficiently and rapidly (*13*). The ease of analog preparation relies entirely on a sound synthetic route toward the original natural product and underscores the importance of careful plan-ning in total synthesis. Furthermore, an amalgamation of the best features of syn-

Nature's medicine cabinet. Structural modifications of natural product templates can lead to biologically effective drug candidates. Permutations include structural simplification with the removal of unneeded functional groups and stereochemistry, facilitating chemical synthesis. (Blue circles highlight proposed drug-target interaction sites.)

thetic routes from several academic groups (by Novartis process chemists) has resulted in an almost combinatorial-style synthesis of discodermolide, readily adaptable to analog preparation, that has provided more than 60 g of active pharmaceutical ingredient to enable its clinical development as an anticancer drug (*14*).

Of course, the opportunities for total synthesis are not restricted to the discovery of anticancer drug candidates. In the case of anti-infectives, analog design may allow us to circumvent drug resistance, in a manner that again cannot be matched by standard methods for antibiotic development. The recent report of a general synthetic route to tetracyclines and analogs shows the potential that lies in this area (*15*). From the outset, this synthesis was designed to access multiple analogs of tetracycline and could be achieved in consistently high overall yield (5 to 7% over 14 steps).

Synthetic developments have thus enabled the designed modification of natural product templates in ways that cannot be

readily achieved by biosynthetic means, yet potentially allow large-scale and commercial syntheses. However, despite important advances in synthetic methodology, the typical time scale for the development of truly practical synthetic routes toward complex natural products, and therefore useful derivatives, is still rather lengthy. At present, the development of new drugs seems limited not by our ability to synthesize a given natural product, nor to make analogs, but rather to do so with efficiency and flexibility, and within the short time scale required to compete with high-throughput synthesis and combinatorial chemistry. Despite the challenges that researchers face in the development of such rapid and scalable natural product syntheses, the unbeatable potencies associated with natural molecules selected by evolution should secure their future as a mainstream source of therapeutic agents for many years to come. Furthermore, the continual isolation of an increasing range of novel bioactive secondary metabolites suggests that we have

barely scratched the surface of nature's vast library of small-molecule ligands.

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Molecular Orbitals Tell the Story

James N. O'Shea

To understand the rich physics of molecular nanostructures and solids, there are times when high-resolution photoemission data are all we need to build a detailed picture of the electronic structure. At other times, structural information from x-ray diffraction or scanning tunneling microscopy (STM) can reveal precisely what is going on at the molecular level. But the most intriguing questions often leave us wishing that we could simply get in there and take a good look at the single-molecule level. On page 468 of this issue, Wachowiak *et al.* describe how they have done precisely this in order to observe the molecular distortion in an insulating monolayer of K_4C_{60} by using a combination of topographic and spectroscopic STM at low temperature (*1*).
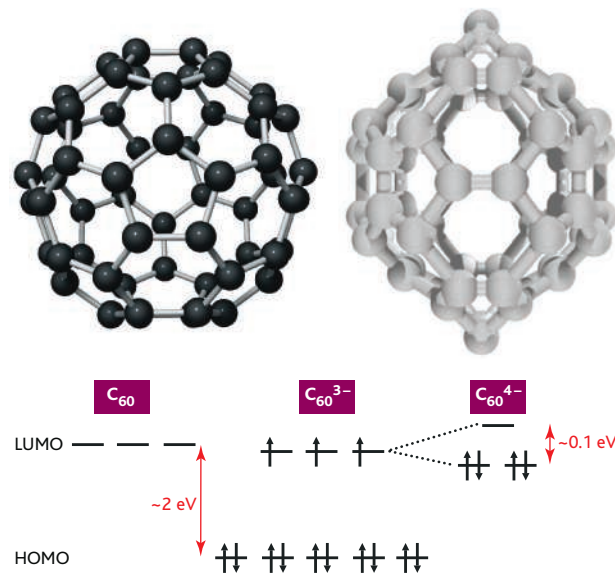
The particular distortion in question results from the Jahn-Teller (JT) effect, a phenomenon with a long history. JT distortions arise when a system is degenerate—that is, it exhibits two or more distinct states with exactly the same energy. Nature tries

to avoid this situation if there is an energy saving to be made by a molecule undergoing a physical distortion so as to split the energy levels apart. JT distortions are thought to play a key role in the electronic

Squeezed fullerenes. Geometric and electronic structure of doped C_{60} molecules. (**Top left**) Undoped and undistorted insulating C_{60}. (**Top right**) JT distorted C_{60}^{4-}. (**Center**) The addition of electrons into the threefold degenerate LUMO of C_{60} and C_{60}^{3-} and (**center right**) the JT splitting of the LUMO for distorted C_{60}^{4-}.

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properties of the alkali metal (A) fullerides A_nC_{60}, which range from insulating to metallic (*2*) and even high-temperature superconductivity (*3*).

There are technological considerations as well. C_{60} is an ideal building block for molecular devices because electrons can easily be donated to the fullerene cage from other molecules, atoms, and surfaces. In the case of A_nC_{60}, about one electron is transferred from each alkali-metal atom that sits in the interstitial sites of a C_{60} crystal or monolayer. So where do these electrons go?

Pure C_{60} is insulating. Its highest occupied molecular orbital (HOMO) is a fivefold degenerate band with a full complement of 10 electrons, whereas the lowest unoccupied molecular orbital (LUMO), some 2 eV above it, is a threefold degenerate band that could hold 6 electrons but is in fact completely empty. C_{60} is therefore a band insulator (see the figure). Additional electrons donated from the alkali-metal atoms are transferred into the LUMO, and on this basis we can intuitively understand why K_3C_{60} is metallic (because it has a half-filled conduction band). Perhaps the more compelling question, then,