point about this selection approach taken by Li and coworkers is that the final screen for putative RNA binding dyes is indeed performed on living cells. Finally, three library components were selected in the secondary screen (E36, E144 and F22) and notably E36, which gave a positive reaction in the nucleoli staining screen, was shown to be particularly highly selective for RNA.

Why are these fluorescent RNA binding dyes particularly suitable for live cell studies? First, they are small and cell membrane permeable. Therefore, they are rapidly and efficiently taken-up by cells when added to the cell culture medium. This feature eliminates the need for rather laborious microinjection procedures. Second, because of their small size, styryl dyes are expected to have easy access to the various compartments within a cell and to the RNA molecules that might be present in those compartments. This is an important issue because there has always been some debate concerning the accessibility of RNA molecules for dyes when they are assembled in ribonucleoprotein complexes in their native cellular environment. Third, the selected compounds have their excitation and emission wavelength in the visible range, are highly fluorescent, are photostable, and show hardly any cytotoxicity and phototoxicity. These are all essential characteristics for live cell imaging studies.

No doubt, RNA-specific fluorescent styryl dyes have the potential to find wide application in research fields related to cell biology. These innovative compounds are particularly relevant in analysis of the dynamic properties of RNA localization in living cells. Furthermore, because various color variants can be selected, these dyes can be easily applied in multicolor approaches by combining them with dyes selective for other cellular components like DNA.

Future studies may provide insight in the mechanism by which styryl dyes bind to RNA. If a structural feature or specific order of nucleotide sequence in the RNA turns out to be involved in binding, it might then be possible to refine the selection procedure to select for library components that bind specific types of RNAs; for example, messenger RNAs, ribosomal RNAs, transfer RNAs or small nuclear RNAs, or even specific gene transcripts.

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Myxing It Up to Study Chondramides

Considerable progress has been achieved in elucidating the biosynthesis of natural products with interesting cellular targets. In this issue of *Chemistry & Biology*, Rachid et al. [1] provide a new example, taming genetic manipulation of the producer of chondramide, an actin-interfering compound.

Natural products (NPs) have provided a large number of existing drugs and NPs, NP-derived, or NP-inspired compounds represented over half of the drugs approved during the 1981–2002 period [2]. A large number of NP-related compounds are also undergoing clinical trials, particularly in the anti-infective and anticancer fields [3]. However, NP-based drug discovery has been progressively abandoned by the pharmaceutical industry during the last two decades. Two reasons have probably contributed to the declining interest in NPs as potential drug leads: the labor-intensive nature of this type of endeavor, and the expectation that genomics, synthetic/combinatorial chemistry, and high throughput screening can provide a sufficient number of drug leads. This expectation has not been realized, and fewer drugs are being approved despite spiraling R&D costs [4].

While large pharma was becoming disinterested in NP research, the field attracted increasing attention from many academic laboratories and biotech companies, especially for the possibility of expanding NP diversity through biology-based approaches, such as mutasynthesis [5], combinatorial biosynthesis [6], or chemoenzymatic routes [7]. These approaches can complement chemistry in expanding NP diversity. In order to

generate analogs of a particular NP, the biosynthetic gene cluster must be available and the producing strain genetically accessible.

Myxobacteria are valuable producers of secondary metabolites, and many compounds interfering with eukaryotic cytoskeleton have been identified from them [8]. While the diversity of their secondary metabolites makes myxobacteria an interesting source of drug leads, the strains are difficult to work with, and few laboratories have mastered the skills necessary to handle them [8]. In recent years, myxobacteria research has gained momentum through the work of Rolf Müller's group, applying molecular genetics to secondary metabolism within this order [9]. In the current issue of *Chemistry & Biology*, Rachid et al. [1] describe the gene cluster for the synthesis of the depsipeptide chondramide in *Chondromyces crocatus* Cm5. This compound interferes with actin, and appears to have the same binding site as phalloidin, the well-known mushroom toxin (see references in [1]). In contrast to phalloidin, chondramide effectively penetrates mammalian cells, resulting in IC_{50} values in the low nanomolar range.

Despite the swarming properties of *C. crocatus* and the paucity of selectable markers, Rachid et al. were able to genetically access this strain through intergeneric conjugation. This allowed them to establish the cluster boundaries through gene knockouts and to propose a biosynthetic pathway for the chondramides. The authors noted several rare or unusual features in the biosynthetic pathway as deduced from the chondramide cluster, such as the presence of two adjacent acyltransferase domains in the second module of the PKS, the occurrence of a likely tyrosine aminomutase, and the lack of obvious candidates for hydroxylation and *O*-methylation.

Despite the regularity of the pathways in secondary metabolism, we should not be surprised to discover "exceptions to the rules." Since the resulting compounds are not strictly necessary for growth, producing strains have probably had the opportunity to "mix and match" genes and to experiment with different enzyme systems for performing apparently identical chemical reactions. Interestingly, the chondramides bear a striking similarity to jaspamide and related compounds isolated from marine sponges, with which they also share the same actin binding site. Thus, it may be that the sponge metabolites are actually produced by bacterial symbionts, as is increasingly being appreciated [10].

The work by Rachid et al. illustrates the amazing progress that can be made by using the genetics of secondary metabolism. The authors successfully applied an almost "full package" of tools to a newly isolated gene cluster from a hard-to-work-with strain: a gene transfer system for the producing strain; gene knockouts; characterization of domain specificity after expression in *E. coli*; and production of fluorinated chondramides after feeding fluorotryptophane to the producing strain. However, further work is necessary before a suitable production system can be made available for this class of actin inhibitors. *C. crocatus* Cm5 is limited in this respect as it has a 9 hr doubling time and produces chondramides in low yield. Possibly, heterologous expression of the chondramide cluster may lead to greater accessibility of chondramide and its derivatives [11].

The current decade is witnessing the development of a growing number of NP tools and the generation of exciting information as a result. The last year has seen the description of new strategies for NP-based focused libraries [12], of novel groups of antibiotic-producing actinomycets [13], of useful tailoring steps [14], the structural elucidation of new classes of enzymes [15–16], and the application of strategies for genome mining [17], just to name a few. At the same time, new chemical classes are being discovered from relatively common microbial sources [18–19]. Therefore, despite a decreased industrial interest, NP research has never been as active as today. In addition to providing important insights into microbial ecology, physiology, and enzymology, some of these tools will eventually be applied for discovering and developing new drugs. It is unlikely that a single set of tools will prove of general applicability. Instead, success may depend on choosing the appropriate approach for the specific drug discovery objective.

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