applying a large magnetic field, however, it is possible to lift the spin degeneracy, thereby splitting the modes and giving rise to conductance plateaus at integer multiples of e^2/h. In an early experiment by Thomas *et al*. (*3*), the 0.7 feature was shown to evolve smoothly toward e^2/h with increasing magnetic field, leading the authors to suggest that this feature may be associated with spin polarization of electrons in the constriction, which persists even in the absence of any external magnetic field.

The 0.7 feature has now been observed in many experiments, and our understanding of its origin has recently advanced considerably. Cronenwett *et al*. (*5*) have shown that the 0.7 feature exhibits many characteristics in common with the Kondo effect, a well-known phenomenon in solid-state physics that arises from the interaction of the conduction electrons in a metal with a magnetic impurity. This experiment suggests that electrons in the point contact play the role of the magnetic impurity in the usual Kondo effect, although how such a local magnetic moment might form in an open constriction was unclear at the time. Recent theoretical studies have cast light on this problem, however, indicating that the strong exchange interaction between electrons can give rise to a spin polarization, with an associated magnetic moment comparable to the Bohr magneton, as the electron density in the constriction is reduced (*6*, *7*). Consistent with these ideas, Reilly *et al*. have suggested that the results of different experiments can be explained by an intuitive phenomenological model, which assumes that a density-dependent spin gap opens in the constriction as its conductance drops below $2e^2/h$ (*8*).

More recently, we have detected the spin-polarized state by studying the interaction between coupled quantum point contacts (see the figure) (*9*). By sweeping one point contact through the regime of expected spin polarization, we observe a resonant peak in the conductance of the upper point contact. Starting from the assumption that a spin-polarized state is formed in the swept point contact, Puller *et al*. (*10*) have formulated a model that accounts quantitatively for the features of our experiment. Within this model, the conductance resonance in the detector point contact is understood to result from its tunnel coupling to the spin-polarized system of electrons.

Although there is a growing consensus that spin polarization can indeed occur when electrons are strongly confined in a nanoscale constriction, much remains to be understood about the microscopic origins of this novel nanomagnetic system. Nonetheless, in future it will be of great interest to exploit these nanoscale constrictions as a controlled experimental system for the study of exotic spin-transport phenomena (such as spin-charge separation). The lessons learned from these studies may ultimately be important for the development of future spintronic devices.

References

1. B. J. van Wees *et al*., *Phys. Rev. Lett.* **60**, 848 (1988).
2. D. A. Wharam *et al*., *J. Phys. C* **21**, L209 (1988).
3. K. J. Thomas *et al*., *Phys. Rev. Lett.* **77**, 135 (1996).
4. A. Kristensen *et al*., *Physica B* **249**, 180 (1998).
5. S. M. Cronenwett *et al*., *Phys. Rev. Lett.* **88**, 226805 (2002).
6. K.-F. Berggren, I. I. Yakimenko, *Phys. Rev. B* **66**, 85323 (2002).
7. K. Hirose, Y. Meir, N. S. Wingreen, *Phys. Rev. Lett.* **90**, 26804 (2003).
8. D. J. Reilly *et al*., *Phys. Rev. Lett.* **89**, 246801 (2002).
9. T. Morimoto *et al*., *Appl. Phys. Lett.* **82**, 3952 (2003).
10. V. Puller *et al*., *Phys. Rev. Lett*., **92**, 096802 (2004).

OCEANOGRAPHY

Microbes, Molecules, and Marine Ecosystems

Farooq Azam and Alexandra Z. Worden

Antonie van Leeuwenhoek (1632–1723), the first observer of bacteria, would be surprised that over 99% of microbes in the sea remained unseen until *after* Viking Lander (1976) set out to seek microbial life on Mars. Through much of the 19th and 20th centuries, microbiologists focused on life-threatening pathogenic microbes or microbes as models of how life "works" at the molecular level. But heightened concerns for ocean health and biodiversity, highlighted in the 2003 Pew Oceans Commissions report (*1*), prompted microbial explorations of the sea with state-of-the-art tools such as satellite and laser-based imaging, as well as massive genomic surveys rivaling the human genome project. This has led to a gold rush of discoveries underscoring critical influences of microbes on marine ecosystems and carrying significant implications for sustainability, global climate, and human health. The challenge is integrating these discoveries at the systems level to elucidate microbial roles in overall system resilience. Understanding the role of microbes in structuring healthy and stressed marine ecosystems will provide the mechanistic basis for prognostic models. Accomplishing this may require a new and unifying framework for conceptualizing these ecosystems.

Excitement over the "microbial ocean" was roused by the 1977 seminal discovery by Hobbie (*2*) and later studies that pelagic bacteria are hugely abundant (10^6 ml^{-1}), accounting for most oceanic biomass and metabolism (*3*). They had eluded detection because most are extremely small—only a few percent of *Escherichia coli* in volume—and uncultivable. Thus, in order to measure bacterial biodiversity and in situ metabolism, microbial oceanographers had to devise cultivation-independent methods. Results show that bacteria are a major biological force in the oceanic carbon cycle and ecosystem structure.

Photosynthetic bacteria *Prochlorococcus* and *Synechococcus* are the most abundant oceanic primary producers, and their sequenced genomes provide new ecological insights (*4*–*6*). Other discoveries confront oceanographers and conservation biologists with the probability that many functional capabilities and mechanisms remain unknown. For instance, a bacterial gene for proteorhodopsin was discovered 3 years ago (*7*). This pigment, thought to exist only in Archaea, converts light energy directly into an electrical gradient across the cell membrane. The gene occurs in divergent marine bacterial taxa and diverse environments (*8*); hence, inclusion of proteorhodopsin activity may change oceanic energy budgets. Another important example is the discovery of oceanwide distribution of anoxygenic phototrophic bacteria that contribute to oceanic energy and carbon budgets (*9*). Nitrogen budgets, a regulating force of carbon processing and sequestration, must also be reconsidered given new evidence for microbial N_2 fixation as a common feature of oceanic systems (*10*, *11*). Despite tremendous diversity, a single bacterial clade can dominate numerically. The α-proteobacterial clade SAR11 constitutes ~30% of Sargasso Sea surface cells (*12*), and a newly discovered cluster within the *Roseobacter* clade, found

F. Azam is in the Marine Biology Research Division, Scripps Institution of Oceanography, University of California, San Diego, La Jolla, CA 92093, USA. E-mail: fazam@ucsd.edu A. Z. Worden is at the Marine Biology and Fisheries Division, Rosenstiel School of Marine and Atmospheric Science, University of Miami, Miami, FL 33149, USA. E-mail: aworden@rsmas.miami.edu

throughout temperate and polar regions, makes up ~20% of Southern Ocean bacteria (*13*). Finally, massive marine prokaryotic metagenome sequencing is providing a tremendous database for discovering metabolic capabilities and new ways to conceptualize and study prokaryotic biodiversity (*14*).

Environmental genomics is also revealing bacterial interactions with marine animals, and their influences on animal populations and ecosystem function. Corals offer an excellent example. A recent study discovered 430 novel bacterial ribotypes associated with three coral species (*15*), and shifts in bacterial species composition appear to underlie coral health and disease (*16*, *17*). Appreciating this diversity in conservation efforts is important because functional redundancy cannot be assumed.

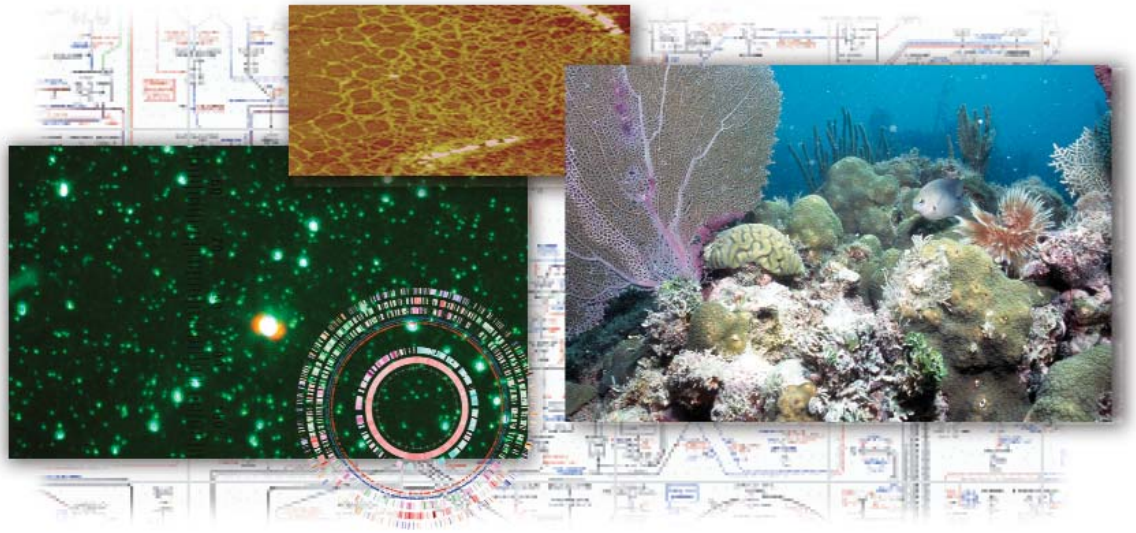
Marine bacteria are but one microbial realm in the ocean for which discoveries with ecosystem consequences abound. Viruses were not studied until 1989 yet are the most abundant biological entities in the sea (10^7 ml^{-1}). Bacteriophages induce bacterial mortality, creating a futile carbon cycle in which dissolved organic matter assimilated by bacteria is released via bacterial lysis and metabolized by other bacteria, enhancing upper-ocean respiration (*18*). Species specificity and host density–dependence lead phage to "kill the winner," maintaining bacterial diversity, with implications for organic matter decomposition (*19*, *20*). Phage diversity studies were initially restricted by the requirement for cultivated hosts. Now, cultivation-independent genomics reveal enormous diversity, including a picornalike superfamily implicated in phytoplankton mortality, even of toxic bloom–forming algae (*21*, *22*). Furthermore, specific cyanophage distributions vary from coastal to open ocean, likely influencing *Prochlorococcus* or *Synechococcus* host distributions differentially (*23*).

In view of microbial abundance, diversity, dynamics, and influence on ocean chemistry, ecosystem-based conservation models must explicitly include microbes, to develop a functional view that integrates microbes, macrobes, and abiotic ecosystem components. This requires going beyond biodiversity assessment, because functional diversity depends on the environmental context of microbial expression. The task concerns an age-old theme in ecology: scale, both spatial and temporal, and the integration of scales (see the figure). Microbes are no different from larger organisms in this sense—one must study them at habitat scales relevant to their adaptive strategies to determine how their metabolism influences larger-scale ecosystem dynamics. For microbes this spatial scale is miniscule, from micrometers to millimeters. Because microbes influence ecosystems through molecular interactions, for example, involving cell surface receptors, permeases, or enzymes, the scale reduces to macromolecular, or nanometers. Thus, microbes' ecosystem activities, whether they involve carbon cycling or pathogenesis toward marine animals, should be modeled as molecular events.

Studies of biochemical interactions and fluxes have yielded valuable insights. The oceanic silicon cycle is being revised following the discovery that colonizing bacteria cause postmortem dissolution of silica from diatom cell walls (frustules). These bacteria secrete proteases that denude the silica shell of its protective protein layer and enhance silica dissolution rates (*24*, *25*). Silicon cycling, in turn, regulates diatom productivity, which is important, for example, in carbon cycling and fisheries.

Genomics opens another dimension for integrating cell biochemistry and ecosystem dynamics. Genomic exploration of two *Prochlorococcus* ecotypes has not only identified features with "…obvious roles in the relative fitness of the ecotypes in response to key environmental variables…" but has also inferred biochemical bases for ecotype-specific environmental impacts (*5*). The *Silicibacter pomeroyi* genome is yielding ecophysiological insights into bacteria producing dimethyl sulfide (DMS), a volatile compound that influences cloud formation (*26*, *27*). *S. pomeroyi* degrades dimethylsulfoniopropionate (DMSP) via several pathways, but only one leads to DMS; thus, the relative expression of particular DMSP-degrading enzymes regulates climate. Overarching the microbial activities is the physical organization of organic matter in seawater, allowing the visualization of ecosystems as molecular architectures. Seawater is structured with cross-linked polymers, colloids, and nano- and microgels, creating an organic matter continuum and a wealth of surfaces displaying activity and biodiversity hot-spots (*3*). This structure provides a spatial context for microbial interactions and adaptations, and permits ecosystem analysis in terms of molecular architecture and biochemical fluxes.

These discoveries illustrate that nanoscale biochemical bases of ecosystem function are tractable, informative, and in-



Microbes in marine ecosystems. Highly abundant and diverse bacteria and viruses (green dots) interact with biotic and abiotic ecosystem components at the molecular, or nanometer, scale yet critically influence the functioning of complex, large-scale marine ecosystems [e.g. endangered coral reefs (**right**)]. An atomic force microscope image of marine polysaccharides (**top**), a biochemical pathways chart (**background**) and genome map of *Prochlorococcus* provide examples of the context for analysis of complex marine ecosystems as dynamic molecular architectures.

PHOTO CREDITS: (CORAL REEF) D. KLINE; (EPIFLUORESCENCE MICROGRAPH) J. FUHRMAN; (ATOMIC FORCE MICROGRAPH) V. SVETLICIC & E. BALNOIS; (BIOCHEMICAL PATHWAYS) BOEHRINGER MANNHEIM

deed essential, if we are to develop mechanistic models of ocean-basin biogeochemical dynamics. Microbial oceanography has made remarkable strides toward system inventory, exposing tremendous diversity of microbial capabilities, and spatial and temporal dynamics. Although they reveal the incredible capacity of microbes to interact with ocean systems, these studies do not address system structure or organizational processes. Biosystems exhibit variability at all organizational scales, from gene expression to groups of diverse individuals, but all display molecular interconnectedness. Analysis of marine ecosystems as biochemical matrices is complex, but not unlike, for example, that of neurobiology and systems biology. Modeling efforts must incorporate interaction mechanisms and variability at all ecosystem scales, including the nanometer-to-millimeter realm of microbes and molecules. Such an analysis can now begin, but it will require a convergence of genomics and the nascent field of microscale biogeochemistry, addressing spatially explicit biochemical interactions and their ecosystem consequences. This approach will lead to new testable hypotheses and prognostic models.

We propose consideration of concepts that are driving systems biology with its goal of elucidating all significant molecular interactions underlying the structure and functioning of a cell or an organism. The approach can be extended to marine ecosystem analysis ("ecosystems biology"), in essence treating the ecosystem as dynamic molecular architecture and appreciating real-time expression as the mechanistic basis of ecosystem dynamics. Although the goal of treating organisms and the environment as a molecular continuum is huge, the rate of progress in genomics and proteomics and its integration into oceanography—a field rich in computational talent—promises success. Ecosystems biology offers a framework for integrating genomic, biochemical, and environmental data. This framework will also unify efforts for biodiversity conservation and conservation of desirable biogeochemical states of the ocean.

References and Notes

1. Pew Foundation Ocean Commissions report, www.pewoceans.org/oceans/oceans_report.asp (2003).
2. J. E. Hobbie *et al.*, *Appl. Environ. Microbiol.* **33**, 1225 (1977).
3. F. Azam, *Science* **280**, 694 (1998).
4. A. Dufresne *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **100**, 10020 (2003).
5. G. Rocap *et al.*, *Nature* **424**, 1042 (2003).
6. B. Palenik *et al.*, *Nature* **424**, 1037 (2003).
7. O. Beja *et al.*, *Science* **289**, 1902 (2000).
8. J. R. de la Torre *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **100**, 12830 (2003).
9. Z. S. Kolber *et al.*, *Science* **292**, 2492 (2001).
10. D. G. Capone, J. P. Zehr, H. W. Paerl, B. Bergman, E. J. Carpenter, *Science* **276**, 1221 (1997).
11. J. P. Zehr *et al.*, *Nature.* **412**, 635 (2001).
12. R. M. Morris *et al.*, *Nature* **420**, 806 (2002).
13. N. Seije, M. Simon, T. Brinkhoff, *Nature*, **427**, 445 (2004).
14. J. C. Venter *et al.*, *Science*, published online 4 March 2004 (10.1126/science.1093857).
15. F. Rohwer, V. Seguritan, F. Azam, N. Knowlton, *Mar. Ecol. Prog. Ser.* **243**, 1 (2002).
16. K. Patterson *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 8725 (2002).
17. J. Frias-Lopez, A. L. Zerkle, G. T. Bonheyo, B. W. Fouke, *Appl. Environ. Microbiol.* **68**, 2214 (2002).
18. J. A. Fuhrman, *Nature* **399**, 541 (1999).
19. T. F. Thingstad, R. Lignell, *Aquat. Microbiol. Ecol.* **13**, 19 (1997).
20. J. A. Fuhrman, M. Schwalbach, *Biol. Bull.* **204**, 192 (2003).
21. M. Breitbart *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 14250 (2002).
22. A. I. Culley, A. S. Lang, C. A. Suttle, *Nature* **424**, 1054 (2003).
23. M. B. Sullivan, J. Waterbury, S. W. Chisholm, *Nature* **424**, 1047 (2003).
24. K. Bidle, F. Azam, *Nature* **387**, 508 (1999).
25. K. Bidle, M. Manganelli, F. Azam, *Science* **298**, 1980 (2002).
26. J. M. Gonzalez *et al.*, *Int. J. Syst. Evol. Microbiol.* **53**, 1261 (2003).
27. M. A. Moran, J. M. Gonzalez, R. P. Kiene, *Geomicrobiol. J.* **20**, 375 (2003).

28. We thank T. Hollibaugh, J. Fuhrman, and C. Beardsley for critical reading of the manuscript; V. Svetlicic, E. Balnois, and D. Kline for sharing unpublished images. Work supported by NSF (OCE132677) and NIH (RO1 A146600).

IMMUNOLOGY

Keeping the Gut Microflora at Bay

Jean-Pierre Kraehenbuhl and Max Corbett

The gastrointestinal tract of human adults contains more than 400 different species of bacteria. Indeed, the total weight of microflora in the human gut has been estimated to be about 1 kg. Most of these bacteria are commensals that coexist peacefully with their host and remain harmless provided that they do not stray beyond the gut lumen. Some commensals may even confer health benefits upon their host by helping to digest dietary carbohydrate and by maintaining the appropriate balance among the different types of gut bacteria. The correct microbial balance helps to stimulate gut immunity and to prevent colonization by pathogenic bacteria that cause diarrhea and other intestinal disorders (*1*).

The authors are at the Swiss Institute for Experimental Cancer Research, CH1066 Epalinges, Switzerland. E-mail: jean-pierre.kraehenbuhl@isrec.unil.ch

The intestinal epithelium provides a barrier against the invasion of host tissues by both pathogenic and nonpathogenic bacteria (*2*). However, pathogenic bacteria are able to breach the gut epithelium and the host's innate immune system in a number of ways, in some cases destroying gut epithelial cells (*3*). In contrast to bacterial pathogens, commensals have been presumed to reside in the gut lumen with little or no direct interaction with the epithelium. Recent studies, however, indicate that gut commensals do interact with the gut epithelium and can trigger innate and adaptive immune responses. In addition, commensals can influence epithelial cell proliferation, for example, by injecting factors into the gut epithelium that block β-catenin degradation (*4*). These microbes also enhance the nuclear export of NF-κB, the transcription factor that controls the production of proinflammatory chemokines by gut epithelial cells (*5*). On page 1662 of this issue, Macpherson and Uhr (*6*) reveal that the gut mucosa—comprising the gut epithelium, connective tissue layer (lamina propria), and gut-associated lymphoid tissues (GALT)—has developed an elegant system for keeping commensals in check. They report that commensals are prevented from breaching the gut mucosal barrier by an immunoglobulin A (IgA) antibody–mediated mucosal immune response. This immune response is triggered by presentation of commensal antigens to B cells in GALT by intestinal dendritic cells (DCs).

Cells of the innate immune system, such as dendritic cells and macrophages, provide broad nonadaptive (innate) protection against microorganisms that are newly encountered by the host. Activation of innate host defense depends on specific recognition of microbial signature molecules called pathogen-associated molecular patterns (PAMPs) (*7*). In the gut mucosa, monocytes and particularly DCs in the intraepithelial and subepithelial layers are specialized for detecting microbial pathogens. Both cell types recognize PAMPs through pattern-recognition receptors that are either secreted or expressed on the immune cell surface (*8*). These molecules include the Toll-like receptor (TLR) family in animals and the disease resist-