ally inhibit photosynthes (*8*). However, in deeper water, light becomes limiting.

It is a fairly simple matter to measure the availability of the various raw materials in the ocean. Can we then predict which nutrient is limiting? In practice we cannot do this with any confidence because the interactions between the phytoplankton and their environment are so delicate and so complex. The ultimate test is simply to try adding more of a particular material and see what happens. The most spectacular example of this experimental approach has been a pair of open-ocean iron-enrichment experiments (IronExI and IronExII) (*5*). In these experiments, enough iron was added to the equatorial Pacific to raise the dissolved iron concentration to 4 nM over an area of about 70 km^2 (*5*). Remarkably, the iron enrichment caused a huge, though temporary, bloom of phytoplankton. This provided convincing evidence that iron availability is the factor that limits phytoplankton growth in this region of the Pacific. However, this conclusion cannot necessarily be extrapolated to other regions of the ocean, where nitrate or phosphate could be limiting.

Do we need an IronEx enrichment experiment (and the equivalent for all the other possible limiting nutrients) for every area of the ocean? This might be the only way to definitively determine the rate-limiting factors in a particular region. In the meantime, some provisional answers can be obtained by devising physiological tests for nutrient limitation. Behrenfeld and Kolber now report a simple physiological indicator of iron limitation in phytoplankton (*1*). Their test is based on the measurement of fluorescence—light that is absorbed by the photosynthetic pigments of the cells but subsequently escapes. Fluorescence measurements are a vital tool in photosynthesis research (*9*). They can give information on everything from the fast early steps in the chemistry of photosynthesis (by using subnanosecond laser pulses) to the global distribution of photosynthetic organisms (by using remote sensing from satellites). Fluorescence measurements can also indicate the mechanisms that photosynthetic organisms use to adjust the function of their photosynthetic apparatus. One such mechanism is the state transition, a rapid rearrangement of the photosynthetic complexes in response to a change in illumination conditions (*10*). State transitions are responsible for a characteristic diurnal variation in phytoplankton fluorescence. Behrenfeld and Kolber report that this pattern is altered specifically under conditions of iron limitation. Iron enrichment changed the state transition fluorescence signature of phytoplankton in the wild, and comparable effects could be seen in laboratory-grown pure cultures of *Synechococcus*. This provides a nice link between laboratory and field studies of the physiology of photosynthetic prokaryotes. Many photosynthetic acclimation mechanisms have been characterized almost entirely on the basis of laboratory studies. It is exciting to see these mechanisms at work on such a huge scale in the real world.

The underlying reasons for the effect on state transitions are not yet certain. Behrenfeld and Kolber give a very plausible interpretation based on the effects of iron limitation on the ratios of different photosynthetic complexes in the cell. However, the situation is remarkably complex in a lab culture of *Synechococcus*, let alone in a mixture of diverse species in the open ocean.

Whatever the physiological explanation, it appears that fluorescence measurements can provide a simple and reliable indicator of iron limitation. The results suggest that iron limitation is much more widespread than previously thought. Primary production appears to be iron-limited, not only in the equatorial Pacific but also in a huge area of the south Pacific (though not in the Atlantic). Widespread iron limitation has interesting implications, because iron is a trace nutrient—a little iron could make a great difference to primary production in vast areas of the oceans. Past episodes of climatic and environmental change may be linked to changes in iron availability in the oceans (*11*). The development of an efficient way to make iron available might even lead to large-scale iron fertilization, which in turn could make the oceans more productive. Furthermore, an iron-fertilized ocean could act as a better sink for atmospheric carbon dioxide (*12*), possibly buffering some of the effects of global warming.

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PERSPECTIVES: VIRAL RNA REPLICATION

With a Little Help from the Host

James H. Strauss and Ellen G. Strauss

Viruses are intracellular parasites that infect cells and use machinery inside the cell to multiply. During multiplication, the viral genomes replicate and these progeny genomes, together with newly synthesized viral proteins, are assembled into new virus particles. What does a virus need from a cell to make new copies of its genome other than metabolic machinery that produces such basic materials as energy, DNA and RNA precursors, and ribosomes to produce viral proteins? Viral genomes usually encode polymerases to synthesize new RNA or DNA, helicases to unwind double-strand RNA or DNA during replication, DNA-binding proteins, proteases, and capping enzymes that add a structure called a cap to the 5′ ends of messenger RNAs. Viral genomes are small, however, with limited coding capacity, so viruses must borrow host proteins to complete their replication machinery. The interactions between a virus and its host have been shaped by long stretches of coevolution, and the borrowed proteins do not necessarily serve the same function in viral replication as they serve in the host. Identification of these borrowed host proteins, which perform essential functions during viral replication, is important for understanding virus-host interactions, which influence the host range and virulence of the virus. The search for such proteins has heated up in the last few years.

DNA viruses use facets of the host cell machinery that replicate and transcribe DNA—hardly a surprising fact, because any DNA virus that replicates in the nucleus has access to such machinery. The intimacy of these interactions is illustrated by differences between the closely related tumor viruses SV40 (a monkey virus) and

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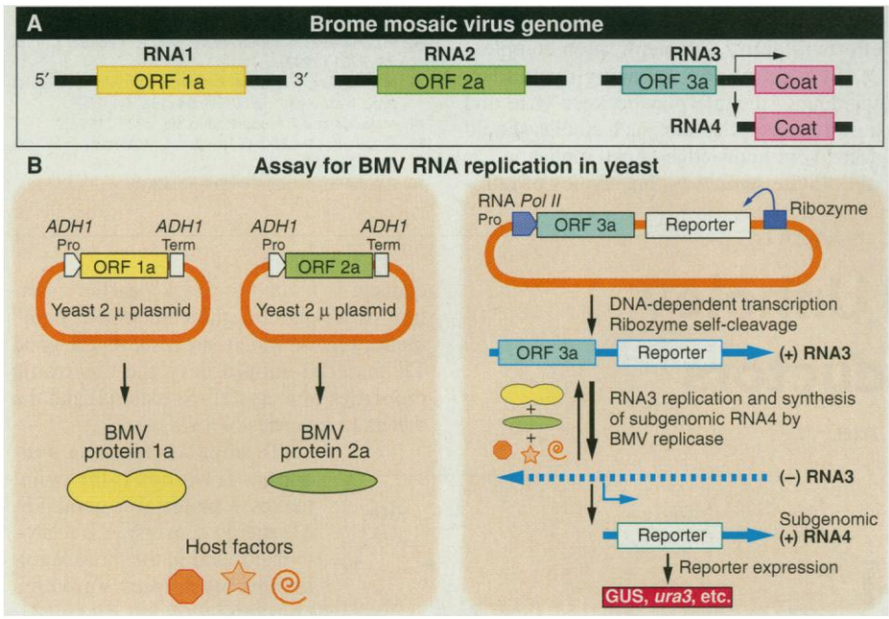
polyoma (a mouse virus). Complexes containing the virally encoded T antigen plus host cell DNA polymerase α-primase (among other constituents) are required for replication of their genomes. α-Primase initiates host or viral DNA synthesis by synthesizing short RNA-DNA primers that are extended by other DNA polymerases that carry out the bulk of DNA replication. Because SV40 requires the primate enzyme to form a stable complex and polyomavirus requires the mouse enzyme, SV40 will replicate only in primate cells and polyomavirus will replicate only in mouse cells (*1*).

of active replicases has proven difficult, and few studies with purified replicases have been reported. As an alternative, many laboratories have turned to the identification of cell proteins that bind to the viral RNA (with mobility shift assays) or to virally encoded replicase components (with cross-linking or two-hybrid system assays) to identify participating host proteins. Cell proteins identified by these various means (*3*) comprise a diverse lot. Most exciting has been the identification of plant and animal elongation factors EF-1α, β, and γ as potential components of replicases. The strongest evidence for the



Yeast reveals host proteins that contribute to RNA replication. (**A**) The BMV genome, which contains four open reading frames (ORFs) on three genome segments. The coat protein is expressed from a subgenomic RNA called RNA4. (**B**) Yeast have been modified to express BMV proteins 1a and 2a (**left**). When RNA3 is added (**right**), it will replicate because suitable host factors are present in addition to 1a and 2a, resulting in the expression of a reporter gene that has been substituted for the coat protein. A ribozyme immediately downstream of RNA3 cleaves to produce RNA3 whose 3′ end is identical to that of RNA2 in the virion. Pro, promoter.

No preexisting cellular machinery for replication of RNA genomes exists in host cells, however. All RNA viruses thus encode an RNA polymerase. Nevertheless, it is increasingly clear that cellular proteins form part of the replicase complex. Early studies of RNA bacteriophage Qβ identified three cellular proteins as components of its replicase, translation elongation factors (EFs) Ts and Tu, and ribosomal protein S1 (*2*). These three cellular proteins function in protein synthesis, but the virus has appropriated them for its RNA replicase.

Abundant genetic evidence suggested that cellular proteins were also components of the replicases of plant and animal RNA viruses, but the identification of such proteins has been slow. Purification

participation of EF-1 in replication comes from Das *et al.* (*4*) who have shown that the polymerase of vesicular stomatitis virus, which replicates in both insects and mammals, binds EF-1α very tightly and that the resulting complex binds EF-1β and γ. All three EF-1 proteins are required for replicase activity in an in vitro system. EF-1α is functionally homologous to bacterial Tu, and EF-1β and γ to Ts. It is not known whether these proteins are true homologs that have descended from common ancestors (although the sequences are known, no sequence identities between the bacterial and animal proteins are apparent), but if they are it would suggest that the participation of these translation factors as components of viral RNA replicases is ancient and has coevolved with the viruses and their hosts.

Other cell proteins implicated in more than one virus system include the polypyrimidine tract binding protein, the protein hnRNP A1, and the autoantigen La (*3*). La associates with the RNAs of many viruses, but it is a known RNA-binding protein; it has been difficult to show that the binding is functional. There is evidence that La binding to poliovirus RNA (*5*) and HIV RNA (*6*) is required for optimal efficiency and fidelity of their translation, but in other viruses it may be involved in RNA replication (*7*).

A very different approach to identification of cellular proteins involved in replication of viral RNAs is being used by Ahlquist and his colleagues. They have developed systems in which the RNAs of two RNA viruses, the plant virus brome mosaic virus (BMV) (*8*) and the animal virus flock house virus (*9*), will replicate in yeast, and are using the power of yeast genetics to identify yeast genes required for this replication. The BMV system and method is further developed (see figure). BMV, which is related to animal alphaviruses such as Sindbis virus and Semliki Forest virus, has a genome consisting of three RNA segments called 1, 2, and 3. Segment 1 encodes protein 1a, which contains an amino-terminal capping activity and a carboxyl-terminal helicase activity that are homologous to the corresponding proteins of alphaviruses (*10*), and segment 2 encodes an RNA polymerase that is homologous to the RNA polymerase of alphaviruses (*11*). RNA3 encodes two proteins, a movement protein in the 5′ region and the coat protein in the 3′ region, which have no homologs in the alphaviruses. Whereas 1a, 2a, and the movement protein are translated directly from the genome segments, the coat protein is translated from a subgenomic RNA produced during replication of RNA3, and thus replication of this genomic segment is required before the coat protein (or a reporter gene substituted for it) can be expressed.

In the yeast system, 1a and 2a are expressed from plasmids stably present in the organism. When replication-competent RNA3 is introduced into these yeast cells, either by electroporation of the RNA or by its expression from a plasmid (*12*), it will replicate when proteins 1a and 2a are present. Several reporter genes have been used to assay for replication of RNA3. One that allows a very sensitive assay is *ura3*, whose product is required for biosynthesis of uracil, and it is possible to select either for or against the ability to synthesize uracil.

Mutagenesis of yeast followed by identification of strains unable to replicate RNA3 has shown that expression of several yeast genes is required for its replication. To date genes in three complementation groups, called mab-1, 2, and 3 (for maintenance of BMV functions), which presumably encode three different proteins, have been partially characterized (*13*). Because the mutant yeast are temperature sensitive, it is clear that these gene products are also required for normal cell growth. In the yeast cell, protein 1a stabilizes 2a, but the mab-1 and mab-2 products are required for this stabilization, suggesting that the 1a, 2a, mab-1, and mab-2 proteins may directly interact (*13*). 1a also leads to a dramatic stabilization of RNA3, suggesting that it may bind directly to RNA3 (*14*). Because assembly of active replicase complexes requires the presence of one of the promoters in RNA3 (*15*), one model consistent with these data is that the active replicase complex consisting of host factors (including mab-1 and -2) as well as proteins 1a and 2a assembles on the promoter.

Because the yeast genome has been entirely sequenced and the technology for dealing with yeast genetics is well advanced, it will be relatively straightforward to identify the mab-1, -2, and -3 proteins, and progress in this direction is being made (*16*). Ultimately, it should be possible to identify all of the host proteins required for replication of BMV RNA in yeast and, by extension, proteins required for the replication of the RNAs of the Sindbis-like viruses in plant and animal cells. Elucidation of the functions of these proteins will require further study and will be complemented by efforts to purify viral replication complexes. In addition to increasing our understanding of the interplay between virus and host during replication, such studies should deepen our knowledge of cell replication as well, in the same way that studies of retroviral oncogenes led to a new understanding of eukaryotic cellular control functions.

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PERSPECTIVES: THERMOELECTRIC MATERIALS

Holey and Unholey Semiconductors

Terry M. Tritt

Cooling electronic devices can often be necessary to optimize performance. Traditional approaches to cooling are typically based on thermodynamic cycles involving compression and expansion of refrigerant gases (such as Freon). The heat required for vaporization is drawn from the materials or the volume (for example, a refrigerator) that is to be cooled. Thermoelectric (TE) materials, in contrast, do not rely on chemicals or gases but rather on a special physical phenomena called the Peltier effect, which is explained in detail below (*1*). In TE materials, heat is not primarily transported by the lattice but by the electrical charge carriers—electrons or holes. Applying an electrical current through a TE material cools one end and transports the heat to the other end of the material or device. This has distinct advantages in that it is solid-state refrigeration, without moving parts and vibrations, and with quiet performance and the ability for localized "spot" cooling. This can be very important for many semiconducting and other electronic devices. But until recently, the efficiency of TE devices has been frustratingly low, and applications remain limited.

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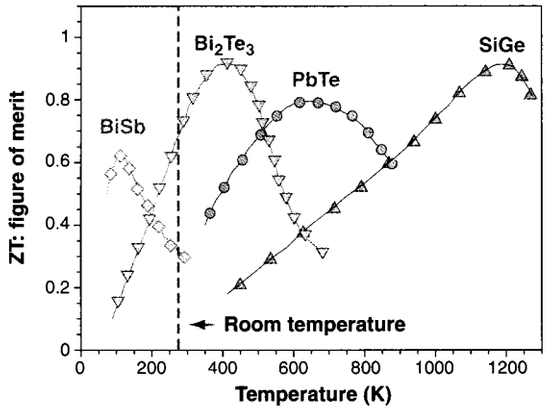

The dart board. The figure of merit, ZT as a function of temperature for current state-of-the-art thermoelectric materials.

Now, new TE materials aimed primarily at refrigeration applications have suddenly become "hot." Several promising approaches were discussed in a symposium at the 1998 Fall Materials Research Society (MRS) meeting held in Boston (*2*). In his introductory talk, Glen Slack (Rensselaer Polytechnic Institute) characterized the materials as "holey" or "unholey," depending on how good electrical conductivity and poor thermal conductivity were achieved in the same material—the key to a good TE material. This concept is related to Slack's earlier assertion of a "phonon-glass/electron-crystal" model (*3, 4*), which suggests that a good TE material should have the electronic properties of a crystalline material and the thermal properties of a glass.

To understand these concepts, first consider what makes a material a good TE. TE energy conversion is related to that found in the more familiar thermocouples, in which contacts between dissimilar metals or semiconductors result in a voltage difference (ΔV) that depends on the temperature gradient (ΔT) (this is called the Seebeck effect). When an electric current passes through a TE material, the Peltier heat transported by the charge carriers leads to a temperature gradient, with heat being absorbed on the cold side and rejected at the heat sink: The result is electronic refrigeration. Conversely, an imposed ΔT will result in a voltage or current and thus in small-scale power generation (*5*). The potential of a material for TE applications is determined by the material's dimensionless figure of merit, $ZT = (\alpha^2\sigma/\lambda)T$, where α is the Seebeck coefficient or thermopower ($\alpha \sim \Delta V/\Delta T$), σ is the electrical conductivity, and λ is the total thermal conductivity ($\lambda = \lambda_L + \lambda_E$, where λ_L and λ_E are the lattice and electronic contributions, respectively). The electronic component or power factor, $\alpha^2\sigma$, is typically optimized as a function of carrier concentra-