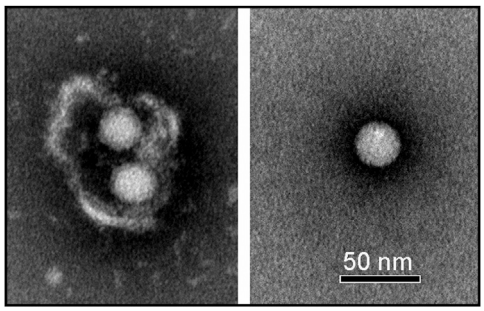
Cell

Antiviral Strategies: Building a Better Defense

Viruses are a widespread persistent threat to human health, and for the most dangerous threats, effective antiviral therapies or vaccines are often still lacking. To combat the rapid pace of viral evolution and the continual emergence of new strains, broadly neutralizing therapies or vaccines that can target multiple strains are in high demand. In this Select, we look at recent papers that are helping to advance antiviral research on several fronts.



"Enveloped" (left) and nonenveloped (right) hepatitis A virus particles released from infected hepatoma cell cultures. Image courtesy of The University of North Carolina at Chapel Hill.

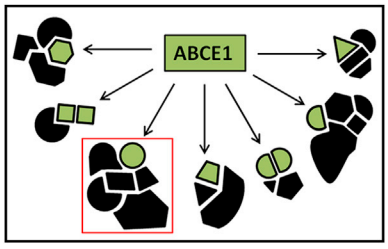
Viral Masquerade

Any student of virology knows there are two categories of viruses: those that are surrounded by a lipid bilayer that is needed for cellular entry and infectivity (enveloped), and those that are not (nonenveloped). In an interesting twist on this long-established dichotomy, Feng et al. (2013) show that the human pathogen hepatitis A virus, a nonenveloped picornavirus, is cloaked in a membrane of host lipids when released from cells, allowing it to masquerade as an enveloped virus and limiting its exposure to the immune system. Although no more infectious than nonenveloped HAV, these virus-containing, exosome-like particles (termed eHAV) are unrecognized by common antibodies against hepatitis A virus, possibly facilitating viral spread in vivo. Furthermore, knockdown of the energy-generating ATPase VPS4B or the exosome biogenesis protein Alix prevents the release of eHAV as well as the production of naked virus, suggesting that this formerly unrecognized particle may be an intermediate in the life cycle of hepatitis A. Importantly, enveloped eHAV particles are the form of virus that circulates in the blood in humans with acute hepatitis A. In yet another distinction between enveloped and nonenveloped hepatitis A, the entry of eHAV into cells relies on successful acidification of endosomes, which is effectively blocked by treatment with chloroquine. Vaccination against hepatitis A is a very successful strategy for limiting infection, and given that viral particles are not accessible to antibody recognition when circulating as eHAV, it appears that the mechanisms by which HAV is neutralized in vivo are more complicated than previously thought. How many other viruses take advantage of this stealth tactic in vivo and whether these pathways of viral escape can be targeted therapeutically are just two more of the many important and fascinating questions raised by the observations described in this report.

Feng, Z. (2013). Nature. Published online March 31, 2013. http://dx.doi.org/10.1038/nature12029.

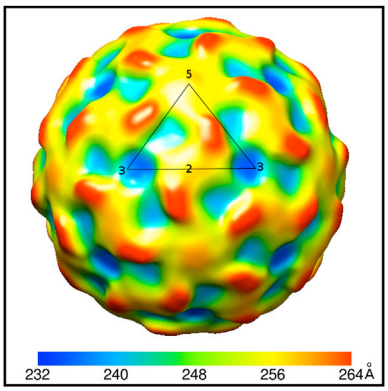
Targeting the Host-Virus Machinery

Infection with rabies virus is nearly always deadly to humans if postexposure vaccination cannot be started within a narrow time frame, and small-molecule treatments are entirely lacking for this disease. In an interesting, unbiased approach to drug discovery, Lingappa et al. (2013) target the viral and host proteins that produce virus capsid, the protein structure that encases viral nucleic acid. Although capsid formation is often viewed as a process of spontaneous self-assembly, the authors follow up on data suggesting that host factors are crucial for catalyzing the processes that generate functional capsid and create a screening method that does not rely on prior knowledge of the factors involved. Using cell-free protein synthesis to generate capsids in vitro, they design a plate-based fluorescent screen to identify small molecules that disrupt capsid production at some point on the pathway from protein synthesis to capsid assembly. Several compounds with robust antiviral activity in cell culture and promising pharmacodynamic properties are identified from this approach. Using one of the most selective compounds, they go on to isolate host-derived machinery components including ATP-binding cassette family protein E1 (ABCE1), which has been implicated in capsid assembly for HIV. In addition to the potential this screening strategy may have for targeting complex protein mixtures without the need for laborious purification, this work highlights new pathways of investigation into virus-host interactions and expands the universe of potential targets for further drug discovery.



Highly heterogeneous proteins like ABCE1 may be part of "next generation" drug targets, provided the implicated subset can be distinguished, as done by Lingappa et al. for rabies capsid assembly (red box). Image courtesy of U.F. Lingappa.

Lingappa, U., et al. (2013). Proc. Natl. Acad. Sci. USA 110, E861–E868.



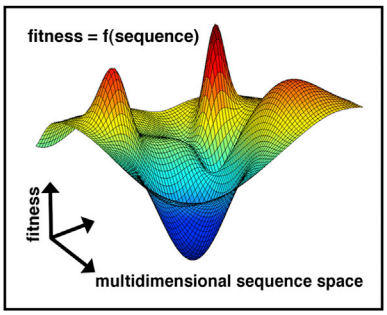
A cryoelectron microscopy reconstruction of the high-temperature dengue virus "bumpy" surface, colored according to the radial distance of the surface from the viral center. Image courtesy of M. Rossmann.

Visualizing the Target

A key aspect of successful antibody targeting is the exposure of any given epitope for binding an antibody, and one recent article provides a new view of the surface architecture of dengue virus obtained using cryoelectron microscopy. Transmitted to humans via mosquito bites, dengue virus usually causes no symptoms or a relatively mild disease known as dengue fever, which is endemic in many countries worldwide. However, a small fraction of cases progress to the more dangerous diseases dengue hemorrhagic fever or dengue shock syndrome. No effective vaccine is currently available, and available therapies target the symptoms but not the virus itself. The structure of the mature dengue virus is known, and it has a smooth viral surface, unless induced to change structure by a reduction in pH. In contrast to the smooth surface observed previously, Zhang et al. (2013) describe a novel "bumpy" viral structure after incubation at temperatures greater than approximately 33°C, i.e., at temperatures similar to those of human hosts. An apparently irreversible structural transition occurs between 31°C and 35°C, a transition that corresponds with a modest increase in infectivity. Zhang et al. propose that the "bumpy" structure is the predominant form of the virus in humans and represents an intermediate capable of fusing with cells. This suggests that future vaccine design should take into account the arrangement of surface proteins observed at higher temperatures and may pave the way for more substantial progress in developing an effective vaccine.

Zhang, X., et al. (2013). Proc. Natl. Acad. Sci. USA. Published online April 8, 2013. http://dx.doi.org/10.1073/pnas.1304300110.

Designer Antibodies and T Cells



A cartoon of a viral fitness landscape inferred by the approach of Ferguson et al.; Image courtesy of A. Chakraborty.

For HIV, the rapid rate of viral mutation and immune escape is one of the biggest challenges to successful vaccine development. Two recent papers present two methods to help understand the effects of mutations on interactions between the immune system and viral particles. In a report that sheds light on the coevolution of viruses and antibodies, Liao et al. (2013) sequence a series of viruses and antibodies isolated from a single host over an extended time period. By following the mutational progression of both the viral envelope (ENV) protein and the host antibodies, the authors find that recognition of the founder virus by the ancestor antibody appears to drive viral epitope diversification and selection, which is then followed by the evolution of a broadly neutralizing antibody, suggesting an ordered sequence of coevolution. They also find that the pathway by which broadly neutralizing antibodies develop is surprisingly less complex than has been previously shown and identify viral protein sequences (both initial and mutated) that might form the basis of an effective vaccination strategy. In a second report, Ferguson et al. (2013) present a computational approach to infer quantitative HIV viral fitness landscapes from sequence databases. This allows the identification of regions for which mutations (single and multiple) impart a high fitness cost, thus they are attractive as immunogens for vaccine development. Although developed in the context of HIV, similar models may be helpful for vaccine design in the context of other rapidly mutating viruses or even in the design of vaccines against cancer. Both reports provide significant insight into the sequences most likely to produce robust, effective immune responses after vaccination for HIV.

Liao, H.-X., et al. (2013). Nature. Published online April 3, 2013. http://dx.doi.org/10.1038/nature12053.
Ferguson, A. L., et al. (2013). Immunity 38, 606–617.

Laurie Gay
Editor, *Trends in Molecular Medicine*

Nicole Neuman
Editor, *Trends in Biochemical Sciences*