by surface science techniques. To overcome this limitation, model oxide catalysts consisting of sufficiently thin, crystalline oxide films have been prepared by epitaxial growth on metallic substrates (*7*).

In a model study, Over *et al.* (*3*) verify the concept of coordinatively unsaturated sites on oxides on an atomic scale. The authors prepared an epitaxially grown RuO_2 (110) film on a Ru single crystal by oxidation of the metal at 700 K. The film thickness was typically about 1 to 2 nm. The films were fully structurally characterized by quantitative LEED and STM measurements. The atomic positions in the RuO_2 (110) surface were fully consistent within experimental error with those known for the (110) plane of bulk RuO_2. The electronic structure of the surface oxide phase was elucidated by density-functional theory (DFT) calculations.

The RuO_2 (110) surface is characterized by rows of bridging oxygen atoms that are running along the [110] direction. These rows were clearly imaged by STM, and the plane of the bridging O atoms was found to be located 0.115 nm above the topmost plane of Ru atoms. This surface also exposes Ru atoms not capped by oxygen atoms, in rows parallel to those of the bridging oxygens. The coordination number of these Ru atoms is 5, and they were therefore identified as cus Ru atoms. According to DFT calculations, these cus Ru atoms maintain their bulk hybridization, and they are therefore expected to expose dangling bonds at the surface and to create high reactivity. Adsorbed CO molecules, which could be imaged by STM because of their low mobility on the oxide surface, were shown to be bonded to the Ru atoms, consistent with their coordinative unsaturation. The pronounced localization of the chemisorption at dangling bond sites distinguishes oxide surfaces from metal surfaces because of the different nature of neighboring surface atoms.

Over *et al.* (*3*) also showed that the chemisorbed CO molecules reacted with bridging oxygen atoms upon brief heating to 600 K, leading to the formation of CO_2 that readily desorbs into the gas phase. The resulting oxygen vacancies were also imaged by STM; reoxidation with O_2 led to healing of the surface. Obviously, in this case, the CO molecule undergoes bonding interactions with both cus Ru atoms and surface O atoms (pair sites), in close similarity to the formation of CO_2^- on the MgO surface mentioned above, the crucial difference being that the latter species cannot easily be desorbed as CO_2.

The work of Over *et al.* (*3*) represents a key step toward an understanding of the nature of chemisorption and active sites on oxide surfaces. The results provide direct evidence for the existence of cus surface sites on oxides. The imaging of chemisorbed CO, and of the O defects created by the oxidation of the former, permits an interpretation of elementary steps of an important catalytic reaction at an atomic scale. Such studies are key for driving surface science research further toward investigations into dynamic processes occurring on oxide surfaces.

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PERSPECTIVES: MICROBIOLOGY

Mice Are Not Furry Petri Dishes

James Bull and Bruce Levin

It is not as though microbiologists really believe that what is true in vitro is also true in vivo. Like most other scientists, they realize that progress depends on developing model systems that, although not faithful replicas of the in vivo environment, enable the easy and repeatable study of biological phenomena. Microbial culture methods have facilitated the isolation and selective propagation of microbes and control of their genetics and metabolism. Unfortunately, it has become clear that interactions between microbes and the complex environment of their hosts cannot be gleaned from in vitro studies alone. The genes expressed by microbes inhabiting a mammalian host are different from those expressed by microbes living in a petri dish (*1*). On page 1479 of this issue, Björkman *et al.* (*2*) now show that the processes of mutation and selection (the basic elements of evolution) in bacterial populations differ depending on whether the bacteria grow in vivo or in vitro.

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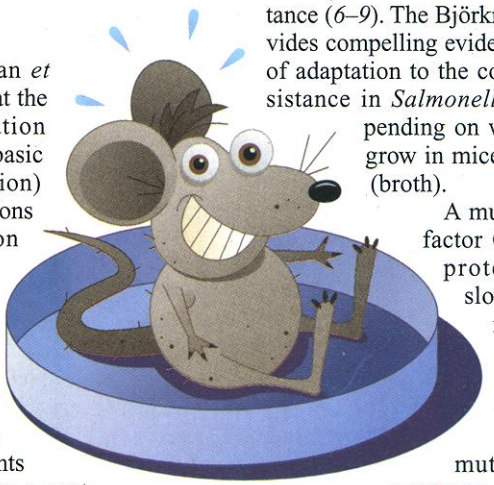

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Mutations that enable bacteria and other microbes to grow in the presence of antimicrobial agents commonly engender a cost that is manifest as a reduced growth rate (competitive disadvantage) in environments where the drug is absent. The cost incurred by drug resistance has been touted as a route to combatting the ever-increasing numbers of drug-resistant pathogenic microbes. The rationale is that because resistance incurs a cost, its incidence will wane if we administer antimicrobial drugs more prudently (*3–5*). Unfortunately, bacteria and viruses adapt to the cost of drug resistance through secondary mutations that compensate for the loss of fitness but usually do not reduce the level of resistance (*6–9*). The Björkman *et al.* study provides compelling evidence that the process of adaptation to the costs of antibiotic resistance in *Salmonella* are different depending on whether the bacteria grow in mice or culture medium (broth).

A mutation in elongation factor G (which decreases protein synthesis and slows growth) confers resistance to fusidic acid on *Salmonella*. The authors recovered 26 independent compensatory mutations and two revertants when *Salmonella* with this drug-resistance mutation were grown in culture broth. (Revertants are bacteria in which the drug-resistance mutation has been lost and the original wild-type DNA sequence has been restored.) In contrast, 11 compensatory mutations and 14 revertants were obtained when bacteria with the drug-resistance mutation were grown in mice. The amino acid substitutions in the 11 mouse-derived compensatory mutations differed from those in the

J. Bull is at the Section of Integrative Biology and the Institute of Cellular and Molecular Biology, University of Texas, Austin, TX 78712, USA. B. Levin is in the Department of Biology, Emory University, Atlanta, GA 30322, USA. E-mail: bull@bull.biosci.utexas.edu; blevin@biology.emory.edu

26 broth-derived compensatory mutations. In general, most of the compensatory mutations in bacteria grown in mice provided only partial recovery of fitness, explaining the preponderance of drug-sensitive revertants. These results are somewhat surprising because it would be expected that the effects of compensatory mutations (which correct the defects in protein synthesis that accompany fusidic acid resistance) would be independent of the bacteria's environment.

The investigators also looked at streptomycin resistance conferred by a mutation in the *rpsL* gene, which encodes ribosomal protein S12. Although broth and mice were not treated with streptomycin, the adaptation to the costs of streptomycin resistance was solely through compensatory mutations and not through reversion to a drug-sensitive phenotype. Intriguingly, in broth bacteria all 14 compensatory changes were located in the *rpsD* and *rpsE* genes (extragenic), and not in the *rpsL* gene. In contrast, in all 10 mice studied, the compensatory mutations were located in *rpsL* (intragenic), within the same codon. The original *rpsL* drug-resistance mutation was a substitution (AAA to AAC) at the 42nd codon; two base changes converting AAC to AGA (which maintained drug resistance) compensated for the effects of this substitution. Unlike the case for fusidic acid resistance compensatory mutations, all of the streptomycin resistance compensatory mutations were accompanied by relatively high bacterial fitness regardless of whether bacteria were grown in broth or in mice. This led the authors to conclude that the differences between mice and broth *Salmonella* in the evolution of streptomycin resistance compensatory mutations lay in the mutation process itself, rather than in selection of mutants. An immediate implication of this finding is that making predictions about the evolution of drug-resistant pathogens in vivo requires that at least some experiments be performed in vivo. Despite the benefits of in vitro experiments, we cannot yet abandon animal models.

Regarding the problems of drug resistance, the results of the Björkman study cannot be interpreted in an optimistic light. In the case of streptomycin, at least, all of the adaptations to the cost of resistance were through amelioration of the drug-resistant mutations rather than by reversion to drug sensitivity. These findings also have implications beyond drug resistance. They suggest that in vivo the mutants generated are quite different and the mutation rate is higher than in vitro. Do compensatory mutations contribute to both acquired resistance in drug-treated hosts and the virulence of infecting microbes (*10*)? Evolution of a bacterial population in an infected host may be completely different from that taking place in a habitat outside of the host. For example, the same gene may be favored in one habitat and selected against in the other. The important findings of Björkman and co-workers raise a number of questions about why mutation and selection, the fundamental elements of bacterial evolution, are different in vivo and in vitro. Answering those questions should keep microbiologists deliciously occupied for some time to come.

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PERSPECTIVES: ASTROPHYSICS

Galaxy-Scale Mergers and Globular Clusters

François Schweizer

The formation of globular clusters and the origin of galaxy shapes, long-standing mysteries in astrophysics that were long viewed as disjoint, have recently turned out to be delightfully intertwined. The conceptual breakthrough came from Hubble Space Telescope observations of colliding and merging galaxies.

Globular clusters are densely packed aggregates of 10^5 to 10^7 stars (see the figure). How so many stars may have formed nearly simultaneously in a sphere only ~100 light-years in diameter has long been a mystery. Our Milky Way Galaxy features about 150 of these magnificent clusters, all nearly as old as the universe itself (10 to 14 billion years). In the 1960s, astronomers postulated that globular clusters in other galaxies were similarly old and may have formed even before their host galaxies (*1*). But problems with this view soon arose. The chemical abundances of globulars seemed to correlate with those of their hosts rather than having universally low metallicity, as one would expect of primordial objects (*2*). And some nearby galaxies were found to possess both old and young globular clusters. The image of globulars as primordial objects thus became tarnished.

Enter NASA's Hubble Space Telescope, among whose early successes were the discoveries of dozens of young globular clusters in a peculiar elliptical galaxy and in two pairs of merging spirals (*3*). Since then, systems of 100 to 1000 freshly minted clusters have been found in a variety of galaxies, often involved in collisions and mergers (see the figure). These observations have shed new light on the cluster-formation process.

Globular clusters apparently form from massive gas clouds in galaxies that get seriously perturbed. Within their rotating disks of ~10^{11} stars, spiral galaxies like the Milky Way or neighboring Andromeda contain a layer of dilute atomic hydrogen interspersed with denser clouds of molecular hydrogen. The most massive of these H_2 clouds, called giant molecular clouds (GMCs), contain 10^5 to 10^7 times as much gaseous mass as our sun and are only marginally stable against gravitational collapse. The 1000 to 2000 GMCs orbiting in a spiral galaxy tend to slowly condense and form stars, but things turn catastrophic when two spirals collide and merge, causing the pressure of the dilute atomic hydrogen to increase rapidly. This results in widespread star birth and shocks the GMCs into prolific star formation on a globular-cluster scale (*4*). Evidence for this process is that the newborn clusters have a mass distribution closely resembling that of the GMCs themselves (*5*).

What does this process have to do with the origin of elliptical galaxies? Ever since Edwin Hubble arranged galaxies into a morphological sequence, astronomers have wondered why galaxies at one end of the sequence are disk-shaped and those at the other end are ellipsoidal. Elliptical galaxies were long thought to have formed shortly after the big bang through

The author is at the Observatories of the Carnegie Institution of Washington, 813 Santa Barbara Street, Pasadena, CA 91101–1292, USA. E-mail: schweizer@ociw.edu