

enough to occupy specialists for some time. Infrared emission measurements allow the calculation of asteroid sizes and reflectivity or albedo. Without such measurements, sizes remain unknown unless an albedo is assigned on the basis of the asteroid's color. The IRAS observations show that no single albedo can be assigned to a given color; there is actually a continuous spread of albedos.

Those working with IRAS data have already reported the first detection of faint dust bands possibly produced by collisions in the asteroid belt as well as dust trails left by known and unknown comets. Next comes a distillation of 35,618 sightings of unknown asteroids.

## From Its Shape, A Look Inside Mimas

Mimas, the small inner satellite of Saturn, is the first planetary satellite other than Earth's moon to have its inner structure probed. It is not a uniform ball of ice and rock, according to Stanley Dermott and Peter Thomas of Cornell University. At some time it apparently separated into a relatively small, probably rocky core and a thick mantle of water ice. That such a small planetary body could manage to become differentiated into core and mantle may help explain how moons and planets form in the first place.

Dermott and Thomas probed the interior of Mimas by simply measuring its shape as recorded by the Voyager spacecraft. By locating the edges of the 400-kilometer moon to accuracies of 0.5 to 2 kilometers in various images, they found that it has a triaxial ellipsoid shape, a less extreme version of the shape of a slightly squashed football. Most crucially for their purpose, the relative lengths of the three axes showed that Mimas is not so small or so strong that it could effectively resist the gravity-induced tidal forces of Saturn and those of its own rotation that create the triaxial ellipsoid.

Given that Mimas does respond like a fluid to the forces shaping it, Dermott and Thomas could compare its actual shape to that predicted for a body that is uniform throughout. Mimas, it turns out, is not quite as squashed as it would be if it were homogeneous. Instead of a difference between its longest and shortest axes of  $20.3 \pm 0.3$  kilometers, Mimas has a difference of  $17.0 \pm 0.7$  kilometers. The conclusion is that Mimas is not uniform throughout but has subdivisions of differing density and thus composition.

Knowing that moons in the outer solar system are composed of ice and rock, Der-

mott and Thomas used the moment of inertia, also called the rotational inertia, calculated from Mimas's shape and the mean density calculated from earlier estimates of Mimas's mass to constrain a model of a moon with a rocky core. They found that it could have only a small core, one less than 200 kilometers in diameter and containing 14 to 28% of the mass of the satellite. The ice mantle in this model must have a mean density of about 1 gram per cubic centimeter or less, which is consistent with the density of uncompressed water ice.

Because the rock and ice of Mimas started out as thoroughly mixed dust and gas, the two must have become separated at some point. In the case of Earth, the separation presumably came after rocky debris had completely accreted to form the planet and radiogenic heat, compression, and the heat generated by impacts during accretion formed molten iron, which sank to form a core. Dermott and Thomas speculate that,

in light of the weak heating expected within Mimas, the separation of the rock to form a core may have occurred as the first step of accretion. Dragged down by the enveloping gas, rocky debris could have formed a planetesimal onto which ice later accreted as the second stage of a two-stage, heterogeneous process.

This technique of shape analysis may be applied to at least one other case, Uranus's innermost major moon Miranda. A patchwork of oddly sculpted dark and light areas, Miranda may have frozen in mid-differentiation after a huge impact broke its light-colored mantle and dark core into pieces. At the meeting Merton Davies and his colleagues at the Rand Corporation and Thomas reported preliminary measurements that suggest Miranda too is a triaxial ellipsoid in hydrostatic equilibrium. Whether it too has a core could help decide among competing theories of its formation. ■

RICHARD A. KERR

# Rice Plants Regenerated From Protoplasts

*The ability to regenerate rice protoplasts means that for the first time a major cereal will become subject to modern biotechnological methods*

THE major cereals are the mainstays of the world's food supply and therefore prime targets for efforts aimed at improving crop yields and hardiness. Nevertheless, these plants, which include rice, corn, and wheat, have so far not shared in the current flowering of plant biotechnology research. A major barrier preventing the application of modern biotechnological methods to cereal improvement has been the long-standing inability to find ways of regenerating the whole plants from the denuded cells known as protoplasts.

That barrier now appears to be falling as groups in England, France, and Japan are reporting success in rice protoplast regeneration. "It is clearly an important breakthrough in that it is the first of the major cereals for which this has been accomplished," says Gary Toenniessen, who oversees the Rockefeller Foundation's program on rice biotechnology research. The achievement opens the way to introducing desirable new traits into rice by direct gene transfer or

by "protoplast fusion," which is a nonsexual way of producing hybrid plants.

The most recent and detailed report of rice protoplast regeneration comes from Edward Cocking, John Thompson, and Ruslan Abdullah of the University of Nottingham, England, who describe their results in the December issue of *Biotechnology*. In addition, M. Y. Coulibaly and Y. Demarly at the Université de Paris-Sud in Orsay, and at least four groups in Japan have been able to recover whole plants from rice protoplasts. The Japanese groups are those of T. Fujimura at the Mitsui-Toatsu Chemicals, Inc., in Kanagawa, who published the first report in 1985; Y. Yamada at Kyoto University; K. Shimamoto at Plantech Research Institute in Yokohama; and K. Toriyama at Tohoku University in Sendai.

Protoplasts of the monocotyledonous subclass of plants, which include the cereals, have generally been much more difficult to regenerate than protoplasts of the other major plant subclass, the dicots, which in-

clude such species as tobacco, petunia, and tomato. Why this should be so is not completely understood, as protoplast regeneration is still more of an art than a science. Nor are the reasons for the current successes with rice completely clear.

According to Cocking, his group finally succeeded because the researchers took the time to induce the plant cells to grow vigorously in suspension culture before using them to prepare protoplasts, which is done by digesting away the rigid cellulose walls that surround plant cells. Most researchers, he notes, make their protoplasts with cells just as they come from the plant.

The long-term culture step—it takes 6 to 7 months—is the major disadvantage of the Nottingham group's method. Toshio Mirashige of the University of California at Riverside, a 30-year veteran of plant cell culture research who is helping the Rockefeller Foundation to keep track of the Japanese work in that area, suggests that such lengthy culture periods may ultimately prove to be unnecessary. They do not appear to be required by the other groups' methods for rice protoplast regeneration. According to Mirashige, protoplast regeneration is regulated by genes, and a plant's genetic constitution is therefore the most important factor in determining whether its protoplasts can be regenerated.

Although the Nottingham group's procedure for regenerating rice protoplasts is time-consuming, it is efficient. Plants were obtained from up to 20% of the protoplast colonies that were eventually cultured. The plantlets developed from somatic embryos that formed from the protoplast colonies. As least as far as the published figures indicate, the other groups have not obtained such high regeneration frequencies.

The high frequency obtained by Cocking and his colleagues also helps to rule out the possibility that the regenerated plants came from contaminating cells that still retained their cell walls, instead of from protoplasts. The contamination problem has long plagued efforts to demonstrate regeneration of monocot protoplasts. In the current Nottingham work, however, the degree of contamination with whole cells was much less than 20% regeneration frequency. Moreover, the Nottingham workers showed that those cells in the protoplast preparation that retained their cell walls were dead. "Cocking has established once and for all that rice protoplasts can be regenerated," Mirashige says.

Cocking suggests that the method may be generally applicable, perhaps even to other cereals. The procedure has worked for several japonica rice varieties, which are grown in temperate climates. It has not yet worked,

however, for indica rice varieties, which are grown in the tropical areas where rice is the staple food and the need for higher yields is greatest.

The ability to regenerate plants from rice protoplasts removes the last barrier to genetic engineering in this species. Researchers have developed highly successful methods for introducing new genes into dicot plants. With these methods they have been able to engineer herbicide resistance into petunia plants and resistance to tobacco mosaic virus into tobacco and tomato plants. A group from the University of California at San Diego has even put the gene for the light-generating luciferase enzyme into tobacco plants, thereby producing plants that light up like fireflies when they are watered with a solution containing the substrate for luciferase.



**Regenerated rice plants:** These rice plants were regenerated from protoplasts by the Nottingham group. [Credit: John Thompson, Ruslan Abdullah, and Edward Cocking]

The methods used for these gene transfers are not applicable to the cereals because the techniques rely on the bacterium *Agrobacterium tumefaciens* to introduce genes into the plants, and this bacterium does not infect cereals and other monocots. However, researchers have shown that cell wall-free protoplasts, including those of rice and maize, can be induced to take up new genes directly, without the aid of a bacterial vector. Further progress has been blocked, however, by the inability to recover whole plants from the protoplasts. The two steps, gene transfer and regeneration, have yet to be put together, but for rice, and perhaps for the other cereals as well, it is now only a matter of time.

Rice protoplast regeneration also means that hybrid rice varieties can be produced by fusing protoplasts. Many of the crosses between plant varieties that breeders would

like to make are not possible because the plants cannot be mated sexually. In the case of rice, for example, wild strains often show better resistance to drought, salty conditions, or disease than cultivated strains, but the wild and cultivated strains are sexually incompatible. This problem can be circumvented by fusing protoplasts of the two strains, but this, of course, will not be of much value unless whole plants can subsequently be regenerated from the hybrid protoplasts.

The newer techniques in plant biotechnology are not intended to replace conventional breeding methods, but rather to facilitate and supplement them. Gene transfer and protoplast fusion permit the introduction of desired traits from any source without the limitations imposed by the genetic incompatibilities that preclude the production of fertile plants by sexual crosses. But once a new trait has been introduced into a plant, further development of that strain will proceed by more standard breeding methods.

Among the improvements that researchers would like to see in rice are improved tolerance for the various environmental stresses such as heat, cold, drought, and salinity and also increased resistance to insect pests and diseases. The tungro virus, for example, is an important rice pathogen. Researchers would like to improve the resistance of rice to this virus by applying methods similar to those used by Roger Beachey and his colleagues at Monsanto Corporation to increase the resistance of tomato and tobacco plants to the tobacco mosaic virus. The Monsanto workers introduced a gene from the virus into the plants that renders them less susceptible to subsequent infection by interfering with tobacco mosaic virus replication.

The varieties of rice that have benefited the most from the improvements of the "Green Revolution" during the past few decades are those that must be grown under irrigation. Efforts to use conventional breeding methods to improve unirrigated rice, such as the upland and "rain-fed" varieties that account for about one-third of the total rice planted worldwide, have been much less successful, Toenniessen says.

Moreover, cultivation of the high-yielding strains of irrigated rice is expensive in that it depends heavily on commercial fertilizers and pesticides. Having built-in resistances to insects and pathogens and more stress-tolerant plants would be of particular advantage in the poorer countries of the tropics where farmers may not be able to afford the necessary chemicals. The ability to regenerate rice protoplasts will aid in the efforts to introduce new traits into all rice varieties. ■ **JEAN L. MARX**