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The Need for Nitrogen

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Narrator: Exacting a toll of death and suffering, the world's food supply trails population growth. To help bridge this gap, scientists are probing a uniquely-talented group of bacteria. This is Stuart Finley for *Men and Molecules*, presented by the American Chemical Society. Today, a report on the progress and promise of nitrogen fixation. Grim pictures tell the story. Babies dying from lack of milk their mothers are unable to supply; children with grotesquely bloated stomachs and vacant, staring eyes; adults wasted away to little more than skin and bone—all harrowing testimony to the world's shortage of food. And unfortunately, the job of supplying enough food—and especially enough protein—to erase these pictures is becoming ever more difficult.

Hardy: The population of the world is currently about 3.8 to 4 billion people; it is increasing at slightly greater than two percent per year. This means that by 2000 we will have something on the order of 7 billion people. To feed these people we are going to need more protein; to produce more protein we are going to need more nitrogen. All protein is composed of carbon, hydrogen, oxygen and nitrogen; about 16 percent of protein is nitrogen. The only new source of nitrogen that can be tapped readily is the abundant nitrogen of the atmosphere. And that's where nitrogen fixation comes into the problem of food production and feeding the world.

Narrator: To Ralph Hardy, associate director of research in the Central Research Department at the DuPont Company, nitrogen fixation and its vital link with protein supply is of real concern. Put very simply, nitrogen fixation is the process in which the abundant nitrogen in the air is "fixed," or converted to ammonia, the chemical form of nitrogen plants need to make protein. In this task, an assorted lot of special bacteria play a key role. One group—called free-living bacteria—lives in the soil, extracting nitrogen from pockets of air and releasing ammonia back into the soil ready to be taken up by plants. A second, and

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more important, group of bacteria—called rhizobia—nestle inside tiny bumps, or nodules, on the roots of certain plants, members of the bean family known as legumes. In this symbiotic relationship, the rhizobia provide fixed nitrogen for the plant, the plant provides vital nourishment for the bacteria—and neither can operate without the other. As far as nature goes, these bacteria together account for most of the fixed nitrogen available for conversion to protein. During this century, though, man has entered the picture as a nitrogen fixer—building factories that produce ammonia in great quantities by combining nitrogen with hydrogen. In fact, much of the world's agriculture now rests solidly on intensive application of fertilizer nitrogen. But today this industrial fixation, as it might be called, faces growing problems. Perhaps most pressing, there is a matter of money. Capital costs of building new fertilizer plants required to meet increasing nitrogen demands are skyrocketing, a special burden for protein-hungry developing nations already strapped for funds. A second problem, of course, is the energy crunch. Huge amounts of fossil fuels are consumed in fertilizer production, both as raw materials and for power. Entwined with these problems, says Dr. Hardy, there is still another, somewhat ironic factor.

Hardy: There has been a Green Revolution. The Green Revolution has made it much more economically attractive to grow high-yielding wheats and rices than to grow legume crops. We've upset, if you will, the legume-to-cereal-grain ratio. The disadvantage of upsetting this ratio is that the cereals have lower amounts of protein—and it is primarily protein that the world population lacks. Secondly, the cereals require added fertilizer nitrogen, whereas the legumes tend to provide a good bit of their own nitrogen by nitrogen fixation. This means that the Green Revolution may in fact have worsened our situation with respect to provision of protein and requirements for fertilizer nitrogen.

Narrator: Driven by these combined forces, there is growing interest in developing alternative nitrogen fixation methods, and we have traveled some distance toward this goal already. The 1960's were the golden decade in unravelling the mysteries of nitrogen fixation. For example, DuPont scientists extracted from bacteria the enzyme, or biological catalyst, responsible for fixing nitrogen. Called nitrogenase, this enzyme was installed in a test tube system, providing the first detailed look at how bacteria are able to convert normally unreactive nitrogen to ammonia. Equally important, the scientists developed a handy method for measuring when, and how much, nitrogen fixation takes place during plant growth. These developments, along with a host of others, have set the stage for future advancements, says Dr. Hardy, and there are three avenues of inquiry. First, is it possible to enhance nitrogen fixation in legumes, thereby increasing the yield and protein production of these crops? Dr. Hardy and his colleagues discovered that the nitrogen fixed by a legume provides only 25 percent of the plant's total nitrogen intake. Furthermore, the rate of fixation slowed down as the plant's seeds grew larger, suggesting that nitrogen fixation is determined by the amount of available photosynthate, that is, the carbohydrates and sugars produced in a plant during photosynthesis. With this as background Dr. Hardy and his colleague, Dr. Havelka, studied the soybean, the most important of the legumes because it constitutes almost half of the world's legume production. They planted the soybeans and surrounded them with plastic enclosures so they could enrich the air around the leaves with carbon dioxide, a key ingredient in photosynthesis. The results were immediate, says Dr. Hardy. The amount of nitrogen fixed increased more than four-fold—enough to almost double total nitrogen input and soybean yield.

Hardy: The results of this experiment were very clear: nitrogen fixation can be dramatically increased, total nitrogen input into soybean plants can be increased, and the way to cause this to occur is to increase photosynthate production by the plant. One may say, "Fine. Can you practically enrich fields with carbon dioxide?" And the answer has to be, I think, a fairly clear "No." The experiment is important because it tells us of the possibilities that can occur in modifying the soybean plant, and where to seek those modifications.

Narrator: One promising approach, for example, is linked to photorespiration—a unique form of respiration in many crops that occurs only during the light and, to our great disadvantage, actually hinders plant growth. The problem here is that the same enzyme that fixes carbon dioxide during photosynthesis turns around during photorespiration and reacts with oxygen instead—and this limits the amount of photosynthate produced.

Hardy: In the carbon dioxide enrichment experiments we've altered the ratio of carbon dioxide to oxygen in the atmosphere surrounding soybean leaves. We've made it so that carbon dioxide is more successful than oxygen, experimentally decreasing photorespiration, and we have shown that by decreasing photorespiration you can dramatically increase nitrogen input and yield. Therefore, one of the approaches in seeking ways to increase biological nitrogen fixation is to seek—via chemical growth regulators or by genetic means—ways of making the carbon-dioxide-fixing enzyme have greater affinity for carbon dioxide and less affinity for oxygen. This area I think is the one in nitrogen fixation research that is farthest along as far as showing potential impact.

Narrator: Let's turn now to the second opportunity for improvement, the cereal crops. Since wheat, corn, rice and the other cereals cannot fix their own nitrogen, we must provide fertilizer to obtain maximum production. But is it possible to find bacteria that will grow in cereals and provide fixed nitrogen just as in legumes, thus reducing our dependence on fertilizer factories? A glint of hope here came when Johanna Doberiner, a Brazilian scientist, discovered a peculiar nitrogen-fixing bacterium that lives snuggled up to the roots of certain tropical grasses, taking nutrients from the plants and fixing nitrogen in return.

Hardy: Work needs to be done in this area to find out how many such associations now exist in nature that we do not already know about. If sufficient of these undiscovered types of associations can be found, then possibly we can domesticate these bacteria and apply them to the cereals the same as we now apply bacteria to the legume crops. Another approach one may suggest is to take the symbioses which already exist in the legumes and cause them to exist in the cereal crops. Work in Australia led to the discovery of elm-type plants which had nodules on their roots, and examination showed that these nodules had bacteria inside and these turned out to be rhizobial bacteria just like legume plants. So there is at least one example that has been found in nature now which indicates that the *legume rhizobial system is not just restricted to legumes,* but can occur in plants slightly removed genetically from legumes.

Narrator: Still other approaches here spring from modern genetics. It may be possible, for example, to transfer the nitrogen-fixing or nif genes to other strains of bacteria that *will* take up residence in cereal crops. And through even greater genetic shuffling, researchers hope to transfer the nif genes directly into plant cells, eliminating the bacterial middlemen entirely. But though work on these projects is moving forward, success still lies some years in the future. Moving from the biological realm now, we come to Dr. Hardy's third area of promise, the chemical approach to nitrogen fixation. Over the past few years there has been some limited success in developing systems for generating ammonia from atmospheric nitrogen under normal, or ambient, conditions—rather than the terrific temperatures and pressures needed in conventional fertilizer production. Many of these new systems rely on special metal catalysts that mimic the nitrogenase enzyme used by legumes. By passing an electric current through the catalyst—and given other required conditions—nitrogen is converted, or as the chemist says, reduced to ammonia. While these systems do work, to date they are not terribly efficient. But suppose they can be improved, we asked Dr. Hardy, how might they be put into practical use?

Hardy: One such approach might be the following: if we could take a membrane that had selectivity for nitrogen versus oxygen, then we could take air and enrich it in molecular nitrogen so that oxygen could not compete with the catalytic reduction process. We might then take this enriched air and, using an electrode-type catalyst that would reduce nitrogen to ammonia under mild conditions and that was functional in aqueous-type or water-based systems, we would insert this electrode into an irrigation stream, and thereby at the time of need of the crop convert nitrogen to ammonia, which would then flow directly to be utilized by the crop.

Narrator: These possibilities sketch only a portion of the entire nitrogen fixation picture; work on a variety of other fronts continues apace, both in the U.S. and in foreign countries. But even with this concerted effort, Dr. Hardy cautions, alternative schemes for nitrogen fixation will by no means provide an immediate solution to the world's food shortages.

Hardy: Thus I think it's very important that you be left with the concept that nitrogen fertilizer production *is* going to be essential, that we will need to build full-throttle nitrogen fertilizer plants during at least the next decade or more to meet the growing nitrogen needs of the increasing population. Yet at the same time, I hope we've kindled in you an intriguing hope and an interest that alternate technologies and alternate approaches may well arise from the very diverse number of approaches that are being taken to increase utilization of air nitrogen.