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Questions and Discussion [The Scientist's Role in the Controversy Over Genetic Engineering, Regulation and Utilization of Microorganisms: A Symposium]

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Then finally, one of my pet concerns is revision of the Plant Pest Act, with which most of you probably are not familiar. The Plant Pest Act is now being interpreted by the USDA so broadly that virtually any microorganism that affects a plant or even a plant derivative and its products can be considered to be a plant pest. One of the primary illustrations that I think demonstrates there is a problem here is *Rhizobium* which now falls in that category. Farmers probably do not

know that the USDA now considers *Rhizobium* a plant pest and yet recommends it as a legume inoculant — a logical inconsistency to me.

I think that I have raised a number of issues here. It comes down to this: if we are going to be competitive and if we are going to realize the benefits, particularly of genetically engineered microorganisms, we need public support, and we need a reasonable public policy. In my view, we have a long road ahead.

Questions and Discussion

ARTHUR WEISSINGER, Moderator

(Weissinger) Thank you, Dr. Vidaver. That gave us a lot to think about. Now I'd like to move on to a couple of people who have agreed to serve as our representatives to ask questions of these biologists. The first will be Dr. Donald Huffman who is Chair of the Department of Biology at Central College in Pella, where there probably are many hundreds of millions of microorganisms residing on tulips. Dr. Huffman comes from a biological background, trained as a plant pathologist, and is, I think, an excellent person to ask questions from a biological perspective, but as a person who is not directly involved in this kind of work.

(Huffman) I don't think that most of you expect nor would you appreciate a lot of comments of my own. Instead, I would like to move directly to some questions that I would like to have addressed. I do thank our speakers for a very fine coverage of the topic. There is one question I would like to address to all three individuals.

Do we have good information on the extent to which altered genes can be transferred to other organisms besides the target organism of *Bacillus* or other genera? In other words, what is the likelihood of transfer of these genes to other natural ecosystem bacteria?

(Dean) In many cases we know that mechanisms exist, but we have no examples in the case of *Bacillus thuringiensis*, which is the major experiment I mentioned that has been conducted, of genes being transferred out of or into this organism. I might say that the genes that encode the toxins for insect toxicity are borne upon plasmids, which would make them excellent candidates for transfer into other organisms and some other bacilli which exist in nature that couldn't possibly transfer their genes, if they would be harmful, into this massive inoculum of *Bacillus thuringiensis*. I think that since we have no examples of this, we could ask, "Have we done all the experiments we need to do to find cases?" I think certainly not. The field of microbial ecology has been compared to microbiologists attempting to study their subject without microscopes. That should have caused a roar of laughter, but it didn't. At any rate, this area of microbiology has been, in fact, the least funded and most ignored, and now at least it's coming into its own light as many other subjects do in the evolution of time.

Nevertheless, scientifically we know that if genes are to be transferred and persist, there must be some selective advantage for the recipient organism to receive these genes. It is simply not a scientific response to say yes, the mechanism is known, and therefore make up your own answer. We have to perceive that there would be some selective advantage in the case of the microorganism to have the genes to open up a new niche for itself, and if this is to be the case, we have to imagine what those selective advantages might be.

(Huffman) I could speculate on what it might be if you had, let's say, endophytic organisms such as were mentioned here, and you could alter those endophytic organisms, that could presumably be an advantage to the organism harboring them.

(Dean) Which way would you alter them?

(Huffman) If you were able to take, let's say, insect resistance conferred by *Bacillus thuringiensis* and to incorporate that into one of these endophytic organisms, surely that would be of some advantage to the host plant harboring the endophytic organisms.

(Dean) Well, there would have to be an advantage to the endophyte. It would have to create a new niche for insect pathogenesis, and that involves a number of steps. It involves the fact that the microorganism would be able to maintain itself in a pathogenic interaction with that insect and detailed, subtle, and multifaceted interactions. It could not be assumed that now I have a gene and can be king of the world. The development of a pathogenic situation is very fine tuned, and I think most of us are working in this area of microbial genetics have a great sense of *deja vu*. We are asking ourselves, "Didn't we discuss these things ten years ago when recombinant DNA first come out?" When epidemiologists first indicated that *E. coli*, the gut microorganism of humans, happens to be the major experimental tool we are using in the laboratory, the reaction was, "Wow, you stick things in there and they happen to get out, and there are going to be some pathogens to humans." The epidemiologists have spoken on this more than ten years ago and have said that it was a ludicrous assumption. What is necessary is for the public to be cognizant of the terms of which they speak when they make that decision.

(Vidaver) I will comment just briefly on that endophytic question. It turns out there is a company using a similar approach that wants to put out an endophytic bacterium similar to mine with precisely that toxin in it. The proposal is being evaluated by the EPA. Experimentally the difficulty with that organism is to have that toxin expressed long enough for it to be effective. The probability of transfer is extremely low, even in experimental situations. People who have not worked with microbes might need to know that you need literally millions and sometimes billions of cells in order to find a single transfer. You have to recognize also that there are probably at least a million microorganisms catalogued throughout the world, and we think that we don't even know about half of them yet. They are all distinct, and they remain distinct. Obviously if we had easy genetic transfer from one microorganism to another, we would have only one or two of them. So, it isn't easy, but that does not mean it can't work.

(Huffman) To me, this represents a very good situation in which one cannot extrapolate, let's say, from antibiotic resistance which does appear to be of some concern, to a situation like this.

(Vidaver) That is correct, and the common thing about that is, that typically that works under selective conditions.

(Lindow) I was going to add that we can basically assume that some transfer would almost inevitably occur in almost all organisms. This can't be demonstrated in natural environments. Some transfer does

occur, but the big question is, what trait would that transfer gain confer, and under what situations would it be selected? I think, in most cases, it would not be significantly different from those characteristics that would have been part of that living organism, and that this truly shouldn't have been treated differently.

(Huffman) Perhaps this is just pursuing the same question, but maybe a bit differently. Do you believe that one can extrapolate freely from the organisms you have investigated so far to assume that this will be, in fact, the expectation in most organisms that will become involved in experiments? The ice-minus bacterium and *Bacillus* look to be fairly readily controlled under natural conditions. Do you expect that to be true for nearly every instance? How far can you extrapolate?

(Dean) We talked a little bit about Martin Alexander today. Prior to the subject of release of genetically engineered microorganisms, Dr. Alexander published a book on microbial ecology, and it has been my duty to educate myself in the theory. I don't know that much about it, but it is interesting that Dr. Alexander made a statement to the effect that the introduction of an alien organism into the soil rarely leads to its establishment. The fact that the species introduced is scarce or absent indicates that the habitat is unfavorable for the microorganism's development. He goes on to say that the ecological axiom is that the community reflects a habitat. He has a long statement about how alien microorganisms (a good example of these are the pathogens that are used in the soil) really have a very poor survival mechanism in that soil. That is the general rule, and we really don't know of any exceptions to that at the present time. I might say that he is doing some very interesting experiments currently, looking at different microorganisms and their survivability in nature. I don't think there is any need for us to make *Bacillus thuringiensis* a paradigm for all possible genetically released microorganisms. I feel fairly confident that this is true for all bacilli, but some other microorganisms may have other special survival advantages, and they may be more persistent. As Dr. Lindow pointed out, there are certain circumstances where we might want to have organisms with better resistance in the environment. For example, I mentioned how the mosquito control agent of *Bacillus thuringiensis israelensis* has such a short survival. It would be very important in Africa to have a microorganism that could last one month, for example, in nature. I don't think they would mind if it lasted forever, because their interest is in controlling malaria and some of the most devastating diseases known to man, which are transmitted by mosquitoes. They want to get rid of the mosquitoes; that's their basic interest. In that case, engineering in more persistence would bring an advantage.

(Lindow) A point was raised earlier that we have a great diversity of microorganisms existing in the world, and one reason they all exist is that they all persist; they persist indefinitely. But the reason they can persist is because they don't all appear at the same place. So we really have to consider the environmental context under which we are going to be looking at them. My *Pseudomonas syringae* didn't persist in the soil, but I wouldn't expect it to because that's not its habitat. Had we left it on leaves, it probably would still be there. So I would guess that the big question is, "Are you going to be interested in making an introduction in the proper environmental context?" What would be the organisms or types of organisms that your strain might be competing with or likely to exchange genes with? This is important since it will persist within the proper or natural environmental context with probably a lot of other organisms.

(Weissinger) Our next questioner will be the Honorable Paul Johnson who is State Representative from Decorah, Iowa. Paul Johnson was invited as an informed lay person, but I should give you some information about his background. He is trained as a forester and is a farmer by profession. I think that he represents a very well-informed lay person. I would like to allow him to question our speakers about some concerns he might have.

(Johnson) Thank you, and it's good to be here. What I'm hearing here today is that policy-makers are in disarray and that we've got to get our act together and become competitive, that we have experiments that we have done for a long time, and that we know that we are probably being over-cautious here.

The question I have is, in the real world, we will not continue to control these as you have. We're entering into a new area, an area of genetically altered organisms. They are not going to be controlled as scientists now control them. They are going to be in the hands of people, and they're going to be moved throughout the world at will and used at will, and abused. We have over 6,000 pesticides in the State of Iowa today. We have 11,000 being used in California. We have very few controls over those, over who uses them and how they are used. Are there concerns here, beyond just the initial licensing for use, in terms of distribution and use throughout the world? We know, for example, that larger organisms escape and cause a great deal of harm. Have we no concerns in the real world with microorganisms that we genetically alter?

(Dean) My initial impression and response would be that the data necessary to address a lot of those questions would result from the larger scale tests. In other words, the way it's supposed to work is that after you get a product and you have some initial idea what is effective, you go through extensive environmental testing in natural situations where you do large-dose response curves and those sorts of things to get a feel for how it behaves and how it might be abused. Those sorts of things are all taken into account in making the final decision for registration of this product before it is legal and commercial use can be made. I guess I'm a natural optimist in that respect, in that if given a chance to put it through the paces under natural situations, we will be getting information that would suggest situations where it might be abused and that if those situations are deemed likely enough to happen, it might pull the plug on such a product. I think that those sort of things should become obvious during some of the larger scale testing that will result.

(Johnson) They are obvious right now in the use of some of our pesticides. We know that some should be pulled, but it's not that easy to do. How about the rest of you?

(Lindow) One of the reasons I research microbial pesticides is because I feel it is a very good ecological/biological approach, which I perceive to have a lot of safety margins over the unusual organic compounds that are being created. I would say that in answer to your question, I have no hesitation to advocate turning to microbial pesticides from a safety standpoint. I couldn't guarantee the effectiveness with our current products, but I think from a safety standpoint I would have a lot of confidence in recommending that we start going in that direction now. In terms of genetic improvements that we would make, I can't imagine that this adds a new attribute of risk or fear. If there are particular questions about it, I would be glad to address them, but I don't believe that these are realistic questions.

(Vidaver) I would tend to agree with the previous commenters in that one can imagine various scenarios, I suppose, but even I (and others) when you get down to specifics, have difficulty imagining a real scenario in which something bad would happen with genetically engineered organisms, again, taking into account our experiences with the customary modified organisms that we have had experience with these many years. That's not to say that it can't happen. For people who are, for example, unfamiliar with plants — plants eventually emerge in production. Most of these originate from a single seed. A plant breeder does testing and then multiple testing and then has favorable consequences, and so on. There are elimination points at many stages. The same thing is true for microorganisms, only we have had less experience. It certainly has been the case with microbial pesticides. So, the possibility is there, but I think we have to take a look at the organisms themselves in our collective experience, and ask

if that is really realistic. Again, I would remind you, one of the problems that we have had with the use of these microorganisms is their specificity. In that regard, we have a great deal of safety built into these organisms, at least in terms of human health and, in fact, in terms of most of the environments in which we would put these microorganisms. There are legitimate questions that have to do with scale-up, but based on a meeting that Steve and I attended last week on the release of genetically engineered microorganisms, I think that over 500 scientists ended up concluding that we should move ahead, though cautiously. One of the concerns was to have a slow scale-up, compared to organisms that had not been so treated, and I think that we can do that. I think that we would minimize any potential problems.

(Johnson) I see the track record for our release of pesticides and what has happened, although I know that this is not exactly the same. As a farmer, I have used these pesticides in the past, being told that they were absolutely safe.

(Vidaver) Have you used atrazine?

(Johnson) Yes, I do, as a matter of fact. I have for a number of years. There is concern from the public's point of view, and I think you certainly must understand that. One other quick question, and then I think the audience ought to ask questions since they have some that are more important than mine. Where should regulation be? Should the State be involved at all in the regulation of genetically altered organisms, or should it be entirely on the federal level?

(Dean) I think that the regulation should be consistent. I don't mind the State having the responsibility, but it should be consistent throughout the states. I think we should be cognizant of some realistic expense for conducting these experiments, but nevertheless, consistency is the major issue. If one state requires undue regulations, that would certainly dampen applications for beneficial agents.

(Lindow) In my idealized, optimistic world, since I would envision these to be a global consequence once used, it should be primarily federal oversight, but with important input from the State because of particular local needs. For example, the State of California had input in our release in California since there were special circumstances in the community or the State that might not have been appreciated by a distant and detached body somewhere in Washington, D.C. Without the burden of numerous local reviews, which may not have the same expertise as can be garnered by a federal organization, I would see the need for federal oversight with local input from either the State or local communities, but with the primary disposition being at the federal level.

(Vidaver) I agree. The option should certainly be there for the states, but they have to realize that there are consequences to anything that they would suggest or do. I would agree that the primary oversight should be federal. I would even go so far as to say that, in the best of all worlds, it should actually be international. Whether or not we can achieve that remains to be seen.

(Weissinger) Thank you very much, Paul. I should mention in passing that the ISU Agricultural Bioethics Committee is responsible for funding this symposium, and in a very direct fashion, Paul Johnson is responsible for the Bioethics Committee. He is the person who introduced the legislation that committed a portion of ISU's biotechnology funding for the development of a bioethics component. That's a very important thing, and I really appreciate the existence of such a body. I would now like to open the session for questions from the audience.

(Question #1) I would like to pose this question in light of the comments that were made on historical records. *Agrobacterium* is very broadly distributed around the world, and it has a very convenient partner, its tumor-inducing capability. Is there any record of that

capability ever being picked up by another bacterium?

(Vidaver) None that I am aware of.

(Question #2) I'm not familiar with the action of Bt bacillus. I have used it in the garden for years and don't have any problem with its use in that kind of circumstance. What about the kind of situation where it is used to control gypsy moths where a whole group of non-target lepidopterans also are eating?

(Dean) The commercial strain at the present time makes three different toxins but, unfortunately, none of them is all that active against the gypsy moth. It usually has to be applied with two treatments and at the present time, its application against the gypsy moth is localized at best, I think. It is used mostly in communities. I think that almost all state agencies still spray chemicals. In answer to your question, there are a number of other non-target insects that may be susceptible, but usually they are not bothered because they are not feeding upon oak trees or are not in the zone where the application is taking place. It is possible that some other particular lepidopterans, but not other insects in general, might be susceptible to a broad scale treatment with *Bacillus thuringiensis*, but I understand that the total ramifications do not have great impact on other lepidopterans. I think this is something that is of concern to the Ohio lepidopterists. I think they would prefer to see the State doing what they are doing, which is much more deleterious against insects in general, not just lepidopterans but all kinds of insects. The kinds of chemical agents used certainly are far worse in affecting other insects, non-target insects, and I speak broadly in that category, than is *Bacillus thuringiensis*. In choosing between the lesser of two evils, current strains of *Bacillus thuringiensis* are far and away better than any chemicals that are used. If you are speaking about what the future may hold, I think the aim is to make more specific toxins against gypsy moths, so the future would really look brighter on protecting non-target moths.

(Question #3) You and your colleagues are all obviously working for a better world, and have goals which are admirable. If there are people with your skills working for the Department of Defense of our country or organizations of other countries, in a worst case situation, could you imagine doing work of this type which would not be free of risks?

(Vidaver) Obviously, anything can be misused. I presume the person who invented the wheel thought about the possibility of running over somebody. This technology is so prevalent that if one wanted to use it in some deleterious way, it could be done, frankly, by the equivalent of a high school student at the present time. If, however, you are in not even the best of all possible worlds, but in the real world (at least in academics, public employment, or private employment), there are very stringent regulations about what can be done at the present time.

(Dean) To reiterate in speaking to just the global potential that your question implies, I think that all technologies have the possibility of being misused as well as used to benefit man. I think that this technology probably has less opportunity for creating unusual human pathogens, for example. I don't think that in order to block off one obscure and possibly unsuccessful attempt to develop germ warfare, we should not use the technology that could have much broader benefits. I don't think that this particular technology has more inherent risks than any other. I see it as being much more specific in terms of what we do. It's much less likely to create mistakes by chance.

(Question #4) I have a comment. I don't think you should be quite so hard on the public. They have been misled by the scientific community on numerous occasions, and they have become skeptical. I think in a democracy it is healthy that they now realize that the scientific community is not neutral in its attitude on many of these issues. The question is this. You have a kind of general argument for

the safety of this technology. You have employed a principle that sounds like this: small changes, small effects. That does not seem to me to be very plausible in principle, but in *Pseudomonas* it was the case. But we're not going to be able to generalize. That did seem to be the principle used. That seems to be false, so now I don't know where you get the generality.

(Lindow) I guess I was not bothered by that principle; instead I suggested that very small changes lead to predictable changes. When you have such a specific change, you should in most cases have a fairly good idea of what that change should bring about. If you're concerned about some toxic effect or some new ecological activity, it should give you some clues at least as to where to start to look for effects that might have been brought about by the changes that have occurred. When you start looking at much larger possible changes such as the introduction of entire species, which commonly are used as an analogy in discussing introduced microorganisms, then indeed, where can you start looking? There are many, many possible unexpected attributes that would have been carried by an introduced foreign organism. Small change means more predictable effects that we could start to look for and adequately address.

(Questioner #4) That begs the question when you go for 'predictable'. The theory is that we're just going to tinker with it slightly so we shouldn't be very careful, but that's not a good argument. Now you have to say, we're just going to tinker with it in a way that you know how to predict your goal, and then that begs the question. Right?

(Dean) No. The problem is that you want to pose a question to us but not allow us to use scientific methods to analyze it. We are proposing in our genetic engineering approach to make specific changes, and then we propose to test the product that we produce in order to test its benefit. None of us advocate not having guidelines and test procedures. We are happy to respond to any questions that you'd like to raise, because we are trying to do something that will benefit mankind. Then we want to test, if you have questions about this organism that you think need to be tested. You need to know what its persistence properties are and what effect it has on non-target organisms. Our goal as scientists is to perform these tests. The title of the symposium is "What is the Scientist's Role" in this controversy, and our role, as I see it, is to respond to your questions and to answer questions from experience. The real difference between science and philosophy is that you must take the philosophy into the real world and kick the rock, as Johnson says, to determine whether it's there, to ascertain the size and shape of the rock and what happens when you kick it. It's our job to go out and kick the rock and find out whether it is safe where it has been put or whether it's a nice object to have around in a rock garden or whatever. So I think that's our role — to respond and from experiments to test the logic of whatever arguments are raised.

(Question #5) I'm bothered by that fact — that, theoretically, such and such would happen, that most of the time the organism doesn't move through the soil or doesn't spread. Is there a sufficient understanding of microbial ecology, where you can speak the lay truth? That is, to assure us that it's mostly, at least, all right?

(Dean) I mentioned an example of what I thought was a fairly global experiment that has been done with the deliberate release of a microorganism. I think the facts are in on that, and I have to say in that case it is apparent that there are no remarkable side effects to the deliberate release of *Bacillus thuringiensis*.

(Vidaver) Again, I would say that we have a whole body of scientific literature in many different fields that indicates variant microorganisms, but I think the answer still would be that we could say this would be something that we could control adequately. It does not mean that we would never have a potentially harmful effect. It does not mean that you could not imagine something could occur. It simply means that based on past experience, we would be able to function, again coming back to prediction and knowledge of specific situations in which we are going to be using microorganisms in the environment in which they are. It doesn't go any further than that.

(Questioner #5) Unfortunately, it isn't us who controls and us who manages. It's the organism in the environment.

(Dean) That's why we're scientists, and that's why we perform experiments. We are not so confident of our results that we just create them and release them, asking for no regulation of the process. We are asking for the right to perform the experiments to answer the questions that you've raised. We want to do the experiments, and we want to respond. I really believe history shows that recombinant DNA research is more responsive as a technology than any other technology I can think of. Perhaps you can think of a technology or field of endeavor — philosophy, theology or any other human endeavor — that has been as responsive to public criticism as recombinant DNA.

(Questioner #5) My comment is that you are not calling on the discipline that is most responsive to the public's request — that is, ecology. The emphasis must be looking to the field of ecology to understand.

(Dean) We are!

(Questioner #5) You are?

(Dean) Definitely, very definitely.

(Vidaver) I don't think that you can equate naturally occurring microorganisms that do harm and that are not controlled by scientists with scientific experimentation. We will always have problem organisms on a natural scale throughout the world, whether we are dealing with plants, humans, or animals, that cannot, in my view, be equated with doing scientific experiments in the field. I think we have to distinguish that. We may or may not ever be able to control the Brazilian coffee rust, for example. We certainly will learn more about how it behaves and so forth, but it is a naturally-occurring disease and is not something that scientists had anything to do with, to my knowledge. The same thing is true of many human diseases that are still among us. I think we need to make those kinds of distinctions.

(Weissinger) I think that this level of discussion brings out the main point that there is a lot of controversy. I hope that the discussion this morning has helped us to understand how scientists involved intimately with the work are approaching the questions that are being asked by those outside of science.