Chapter 5 Ecological Considerations

"Ecology could easily provide an endless catalogue of reasons for never interfering with the biosphere. What we should be doing is to insist on adequate ecological input into the screeting of release proposals, together with a determination to extend that knowledge by case has as experience accumulates."

Bernard Dixon Bio/Technology 4:481 June 1986

"Experts, except for those operating under the banner of ecology, generally oppose the introduction of ecological questions into their problems."

> Garret Hardin Filters Against Folly, p. 56 Penguin, New York, 1985

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5-1. Economic Significance of Insect Species Introduced to the United States, 1640-1977 A Comparison of Genetically Engineered Organisms an Production and Decomposition in Six Ecosystem Types

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In 1987, a report by the National Academy of Sciences stated "an urgent need for the scientific community to provide guidance to both investigators and regulators in evaluating planned introductions of modified organisms from an ecological perspective" (39). This chapter incorporates such a perspective and focuses on a number of ecological issues relevant to the planned introduction of genetically engineered organisms into the environment.

The major concern associated with planned introductions stems from the potential for problematic ecological effects that are unintended or unforeseen. With appropriate regulatory oversight, such effects are unlikely to follow from any planned introductions in the near future. If they

occurred, however, they might be felt at any level-from the local population, through the community and ecosystem, to fundamental ecological processes -- although the probability of such disruptions decreases as their severity increases. A number of ecological questions are explored in this chapter, including the relevance of experience with introduced exotic organisms, the types of disruptions to natural populations that could take place, and the potential for effects on ecosystem processes of energy flow and nutrient cycling. Review of the data bearing on these questions suggests that there is some reason to be cautious, but no cause for alarm at the prospects of planned introductions of genetically engineered organisms into the environment.

GENETICALLY ENGINEERED ORGANISMS AND EXOTIC SPECIES

Studies of biogeography-the distributions of plants and animals-demonstrate the presence in most modern environments of nonnative or exotic organisms that evolved elsewhere. The number of these exotic species (microbes, plants, and animals) is large, as a result both of natural processes of dispersal and deliberate and accidental distribution aided by humans. No good data indicate what proportion of introduced organisms has created ecological problems, although it is generally agreed to be small. Some scientists believe that experience with exotic species provides an example of what might be expected from genetically engineered organisms in planned introductions, and that "the history of introductions is a history of disasters that should not be forgotten lest it be repeated" (23). Others argue that the best guide is the experience with past introductions in agriculture of organisms produced by hybridization and crossbreeding-a history of far more positive results, and one in which negative consequences have been met with a variety of existing control and mitigation strategies. They point out that all major crops in the United States are exotic species introduced from other regions of the world,

and that it is often important to separate the effects of planned introductions from those of accidental introductions. Although no existing database provides all the information needed to answer questions in this area, there are several different sources of relevant data.

Introductions of Exotic Species

A wide variety of reports have been published on invasion or colonization by exotic species (16, 17,23,37,57). Some of the most quantitative data available deal with insect introductions. One study reveals an exponential increase in the number of insect species introduced into the area of the 48 contiguous States from 1640 to 1977 (table 5-1) (46). No data are available on the number of potential insect introductions that failed to become established, but it is assumed to have been far larger than the number that succeeded (46, 56). Of the 1,379 species noted as having been successfully introduced, some economic importance is assigned to 1,089 of them (79 percent). This percentage is almost certain to be an overestimate since insects with no economic impact draw scant

attention and may inadvertently be excluded from the total of successful introductions. The economic significance of those introductions that were noted is further broken down in table 5-1.

One-fifth of the introductions studied had some beneficial impact; another fifth had no significant impact. Two-fifths (41 percent) of the successful introductions were of insects that turned out to be minor pests. About one in five introductions had severe, negative consequences (17 percent). The majority of these negative effects were unexpected, because no such impacts were known in the introduced species home ranges; the new environment apparently lacked constraints that operated in the original areas, If more had been known about the life histories of these insects, the kind of effects that followed their introduction to the new environment might have been anticipated.

One review of the history of plant and animal introductions (57) concluded that in nearly 80 percent of the cases examined (678 of 854), introduced species had "no effect whatsoever on species in the resident community, or on the structure and function of the community." Other, more critical reviews of the same primary data dispute this conclusion, attributing it to a peculiar interpretation and a restricted reading of the data (16,23). Although debate continues, the ecological literature is replete with examples of communities perturbed by introductions, or of introductions invading and further disrupting environments previously dis-

Table 5-1.—Economic Significance of Insect Species Introduced to the United States, 1640-1977

Estimated total native insect fauna1	04,000 species
Total insect introductions (1 percent of native fauna)	. 1,379
Beneficial introductions: Deliberate Accidental Total.	153 134 287 (20.80/o)
Economically unimportant introductions	290 (21.030/o)
Minor pests introduced	566 (41.04°/0)
Important pests introduced: Expected to be pests Not expected to be pests	80 (5.80/o) 156 (11.310/0)
Total	236 (17.1 1%)
SOURCE, R I Sailer, "Our Immigrant Insect Fauna," ESA	A Bulletin 24:3-11, 1978.

turbed. Most ecologists agree that successful introductions, while representing only a small portion of the total usually have some effect on communities in the" host environment.

Introductions into natural systems are most often successful where the host system has been previously disturbed (i.e., by human action or natural catastrophe such as volcanic eruption, severe storms, or floods) or is a "simple" or "disharmonic" system such as those found on oceanic islands. Introductions with the most serious consequences usually involve host systems or organisms that meet one or more of the following criteria (45):

- Environments lacking potential preda= tors, natural competitors, or similar checks are vulnerable to disruption. Known examples of such disruption include herbivores (e.g., goats) introduced to islands, or generalist organisms that feed or prey from high positions in a food chain, and are thus unlikely to be checked by predation themselves (e.g., mongooses, rats, or cats).
- Ecological generalists, or species that can exploit a variety of resources for differing requirements during their life cycles, are particularly capable of invading and colonizing. Examples include organisms that can produce offspring by laying eggs in many different substances, and predators or parasites that can prey upon or parasitize a variety of prey or hosts.
- Disturbed communities, or relatively "simple" or genetically homogeneous communities, are especially vulnerable to any invading species that might exploit available resources more effectively.

These principles were abstracted from studies of natural systems. Although they illustrate situations that are most likely to result in problems, and therefore to be avoided where possible, their applicability to the majority of planned introductions is quite limited.

Introductions of Agrcultural Varieties

Most of the introductions of genetically engineered organisms planned for the foreseeable future are intended for agricultural settings. In such systems—with a long history of introductions of new crop cultivars, animal varieties in husbandry, and microbes either for biocontrol of plant pathogens or inocculants for nitrogen fixation—a significant body of experience exists that is more directly relevant, with better developed control measures available, than is the case for introductions of exotic species.

The Agricultural Research Service of the U.S. Department of Agriculture (USDA) maintains a Plant Introduction Office that compiles data on all plant material brought into the United States. Since its inception in 1898, the office has recorded over 500,000 different plant samples (species and varieties). From 1982 to 1986, an average of 8,270 new accessions per year were added to USDA records (65). Over 90 percent of these were of foreign origin, and the majority (over 80 percent) had no measurable environmental or economic impact, About 10 percent of the introductions have proven to be valuable crops. A small number have become serious pests, such as Johnson grass, water hyacinth, and veronica.

Agricultural introductions of plants are susceptible to a variety of control measures, such as herbicides, tilling, or crop rotation, in cases where field performance is not acceptable. A plant is not likely to escape, and the propensities for hybridization with adjacent weedy species are well known (10), easily reviewed, and often subject to existing methods of mitigation and control (7,8).

Genetically engineered agricultural organisms are more likely to differ from parental strains in only one or a few genes, and to be introduced into a familiar environment, than they are to resemble introduced exotics, which usually consist of a completely new genome in a novel environment (see table 5-2). The small number of genes changed in most planned introductions should not be overinterpreted as grounds for reassurance, however, because in some cases single gene changes are known to have affected the virulence or host range of a parasite or pathogen (22)28) 32,56). Although this is of concern, such changes are not common, and engineered organisms are no more likely than nonengineered organisms to be susceptible to them. Indeed, recombinant DNA techniques enable engineered organisms to be changed from parental strains with respect to the genes controlling only one trait at a time. They are therefore likely to differ from parental strains less, and more precisely, than new varieties or cultivars produced by historical methods of hybridization and crossbreeding. This should permit a quicker understanding of any such shifts in host range or virulence affecting engineered organisms than was possible before, permitting in turn the earlier application of mitigation or control measures.

The introduction of selected microbes for agricultural purposes has been carried out for nearly a century (18,31). Substantial literature exists on the ecology of microbial plant pathogens and their introduction for biological control purposes (5,24, 48,62,64,67). Although uncertainties remain, this experience gives good reason to expect that planned introductions of genetically engineered microbes can be carried out safely.

All this does not mean, however, that there will be no problems with planned introductions of genetically engineered organisms. Indeed, in the long term (10 to 50 years), unforeseen ecological con-

Exotic organism [®]	Engineered organism ^b			
4,000 to >20,000	1 to 10			
All genes have evolved to work together in a single package	Organism has several genes it may never have had before, Most likely to impose a cost or burden.			
Foreign	Familiar, with possible exception of new genes			
	Exotic organism ^a 4,000 to >20,000 All genes have evolved to work together in a single package Foreign			

Table 5-2.—A Comparison of Genetically Engineered Organisms and Exotic Species

^b "Engineered organism" is used here to mean one new to the nabitat.
 ^b "Engineered organism" is used here to mean a slightly modified (usually, but not always Via recombinant DNA techniques) form of an organism already present in the habitat.

SOURCE: Office of Technology Assessment, 19S8.

sequences of using recombinant organisms in agriculture are not only likely, they are probably inevitable. But it is crucial to put this into perspective: It is difficult to describe a credible scenario that will lead to a problem that is different in kind from the problems created by, and grappled with, in past agricultural practicees. And while the adequacy of current regulatory policies in dealing with existing agricultural practice may deserve examination, planned introductions of genetically engineered organisms do not appear to bring with them such potentially new problems that they require entirely new

regulatory approaches or more stringent review.

In summary, for the majority of planned introductions of genetically engineered organisms presently being contemplated, the experience with new agricultural varieties is a better model of what can be expected than is experience from introduced exotic species. While there are grounds for caution, at least in the near term this experience offers more reason to be reassured than alarmed.

POTENTIAL IMPACTS ON POPULATION OR COMMUNITY STRUCTURE OR INTERACTIONS

Much of the concern over planned introductions of genetically engineered organisms stems from the difficulty of predicting reliably the consequences of any particular ecological perturbation. In all cases, the ability to predict depends on the degree to which the organisms and the initial conditions involved are understood, and in some cases this is considerable. But the complexity of ecosystem processes and the numbers of different species even in simple systems means that there will always be uncertainties. Nevertheless, laboratory studies, greenhouse and small scale fieldtests, and historical experience all suggest it is possible to anticipate the likely consequences of introductions planned for the near future (over the next 5 years).

The potential ecological impacts most likely to be noticed early involve effects on indigenous organisms. Such effects are more likely to be seen at the population or community level than at the level of ecosystem process. Engineered organisms that might cause such effects may be grouped into four categories (29):

- 1. slightly modified forms of native organisms;
- organisms existing naturally in the target environment but requiring continual supplemental support;
- 3. organisms that exist naturally elsewhere in the environment, but that previously have not reached the target environment (or have

reached it only at low levels); and 4. genuine novelties.

Slightly modified' forms of indigenous organisms have a relatively high (but still low) probability of competing effectively with natives and thus of perturbing populations of closely related or conspecific individuals. Most deliberate releases for the foreseeable future are likely to be of this sort. Despite some exceptions, it remains true that the majority of mutations or genetic changes are more likely to have negative than positive consequences for the competitive abilities of engineered organism& Thus researchers working to produce organisms for planned introductions are generally more concerned about the problems of enhancing the competitive abilities of engineered organisms so that they will survive to perform their intended functions. When genetic changes are directly aimed at enhancing competitive or survival abilities the review process should explicitly recognize this, analyze the selective forces involved, and review the potential implications.

Organisms existing naturally in the target environment, if they require continual supplemental support, are even less likely to cause negative effects. The need for continual supplementation provides an effective means of control: stop the supplementation, and growth or function of the introduced organism ceases. It would be logical for planned introductions of this sort to be most common in agricultural settings, but few are anticipated. Researchers and farmers are trying to minimize the need for continual replenishment (e.g., fertilizers for crop plants), not increase it. This desire is one of the factors that has driven agriculture to exploit biotechnology, in the hope that products can be engineered to be easily managed and self-sustaining once introduced. The need for continual supplementation might be exploited, however, to help control certain applications. But it must be realized that natural selection would be continually operating in ways that would decrease the effectiveness of this type of control, favoring genetic variants free of the imposed limitation.

The introduction of organisms that exist naturally elsewhere, but that have not previously reached the target environment, presents the highest probability of disrupting a community or constellation of species in a given area. This category bears the most similarity to the introduction of exotics, and is most likely to result in what have been called "cascade effects" (discussed in the section on insect communities). And although most introductions do not result in such severe effects, dramatic and far-reaching disruptions of community structure can take place. Especially here, though, perspective is important. Although some planned introductions are indeed intended to allow growth in environments new to the organism, appropriate regulatory oversight should minimize risks. One example of a beneficial introduction of this kind would be specific insect predators as biocontrol measures aimed at severe pests. In the past, biocontrol introductions that have been preceded by critical review have often been remarkably successful, with negative consequences rare, but sometimes substantial (25,44).

The effects of genuine novelties are the most difficult to predict because directly relevant experience is lacking. One of the chief problems here is the lack of agreement on what would constitute a genuine novelty. Some contend that inserting a gene into an organism that would thus obtain an entirely new function or property, as for example, Monsanto's *Bacillus thuringiensis (BT)* **toxin containing pseudomonas, would constitute a novelty. There is no general agreement on this, however, worthy arguments being raised by both** sides. Fortunately, few planned introductions envisioned in the near future fall into such disputed categories and it is broadly agreed that, for the foreseeable future, most engineered organisms will not be truly novel.

A clearer picture of the potentially negative consequences most likely from planned introductions can be gained from a review of the types of introductions anticipated indifferent categories of communities.

Plant Communities

The largest number of modifications to plants planned for introduction in the foreseeable future involves the insertion of genes to provide herbicide resistance. Most of the major companies working in agricultural biotechnology are mounting efforts in this direction, studying crops for which herbicides are useful in restricting competition from weeds. Such crops include tobacco (more valuable, however, as a well understood and malleable experimental system than as an end product for agricultural use in and of itself), tomatoes, corn, rape, soybeans, and cotton.

Introducing herbicide resistance genes into plants may bring ecological as well as economic benefits by increasing the use of safer herbicides and allowing their more precise administration. Success could lead to significant increases in market share for particular herbicides, some of which, however, are associated with significant environmental disadvantages.

Most of the herbicides to which resistance is being engineered (e.g., glyphosate or sulfonylurea) are environmentally "soft" -in other words, they are not highly toxic to humans or vertebrates. In many cases they affect only plants, since their target sites are metabolic pathways unique to plants. Some of these herbicides degrade within days or weeks to benign end products such as carbon dioxide and water. Work is also being done on inducing resistance to longer lasting, or more toxic, herbicides, such as atrazine. Successful *research* in such programs might well lead to increased use of some herbicides with less desirable characteristics. The particular cost/benefit estimates will vary with each herbicide and its qualities. Consideration of such factors might be more appropriate in reviews for product licensing than in the regulation of field tests, but could be included at any stage.

The major concern with herbicide resistance genes is that they might spread into related, weedy species, most likely through sexual reproduction (10). The potential drawbacks are obvious: Worldwide economic losses to weeds are still measured in billions of dollars per year. While this is a greater problem outside the United States, it is neither a new problem nor one unique to genetically engineered plants. In many cases such existing problems are managed by rotating either crops or herbicides.

Researchers are also trying to insert pest resistance genes directly into plants, which could reduce the need for pesticides. Several companies are pursuing such applications, but the principles are exemplified by the efforts of Rohm & Haas. Successful field tests have already taken place (see ch. 3 and app. A) with tobacco plants engineered to carry the delta-endotoxin gene from BT.

Different varieties of the toxin (derived from different strains of B. *thuringiensis*) are poisonous to the larvae of some herbivorous insects, primarily certain Lepidoptera (moths and butterflies) and Coleoptera (beetles). The toxin's action depends on fundamental biochemical characteristics (pH, membrane permeability, etc.) of the larval digestive tract in these insects. As the larvae mature, and their digestive tracts develop, they become less sensitive to the toxin. Young larvae are most sensitive.

The field test showed that the delta-endotoxin gene is expressed in engineered tobacco plants. The toxin produced in plant tissues conferred essentially complete protection against predation by the tobacco hornworm *Manduca sexta*. This caterpillar is a common agricultural pest on a variety of crops, and several companies are working on protecting different crop species by this or a similar technique.

There appears to be little, if any, cause to be alarmed at the direct consequences of this compound being present in large quantities in the environment. The BT toxin has been used in various formulations since the first product was licensed in 1962 (66). As indicated in chapter 4, it is presently available in 13 different formulations (e.g., powder or soluble concentrate) in over 410 different registered products. Total quantities that have been administered are in the hundreds of thousands of tons, with ill effects on humans, vertebrates, or nontarget organisms virtually unknown. (For one of the rare citations of a human problem associated with BT use, see (47).)

Yet there is still some reason for concern. Largescale applications of toxin+ obtaining plants or novel microbial delivery vehicles might promote the evolution of insect resistance to this currently safe and effective agent by increasing the distribution and persistence of the toxin in the environment. Delta-endotoxin is extremely effective against its target organisms. Evolutionary biologists would say it exerts a strong selection pressure. This means that given persistent exposure to the toxin, particularly in sublethal concentrations, target organisms could be expected eventually to evolve tolerance or resistance to deltaendotoxin through natural selection.

Because the toxin is so unstable, and sporadically used, it generally does not persist long enough for resistance to evolve. The BT toxin is a biodegradable protein, sensitive to temperature, moisture, and especially to ultraviolet (UV) light, which causes its rapid decomposition. Biotechnology could change this by packaging the toxin in new vehicles (e.g., the Mycogen product; see app. A) placing it in new locations (e.g., inside host plant tissues, as in the Rohm & Haas product) or both (e.g., the Monsanto product) where it would be protected from degradation. The toxin could persist in the environment long enough to allow natural selection to produce resistant insects. This scenario is supported by at least one report of evolved resistance to BT toxin (34), in which seeds stored in silos are dusted with BT. In this setting, where the toxin is protected from UV degradation, resistance in the insect pest has been observed to evolve rapidly, and in more than one instance.

This problem is less one of product safety, however, than of useful product life. The declining agricultural market for BT in recent years, as it has been replaced by promising new compounds like the synthetic pyrethroids, makes it unlikely that loss of BT as an agricultural pesticide would lead to an increased reliance on older, more dangerous chemical pesticides. But a consideration of possible responses to a potential problem such as this is illustrative.

Because the evolution of resistance is a product of selection pressure created by the chronic presence of the toxin, either when or where it is not needed (e.g., in roots or stems, in addition to the leaves that are eaten), the problem might be solved by limiting toxin distribution to the times and places it is really needed. One way to do this would be to limit the expression of the BT gene to only the tissues subject to damage by herbivorous insects. The system could be refined further by making gene expression inducible, so that toxin production in the plant tissues is triggered by damage caused by herbivorous insects (27). This strategy would require the insertion of precisely controlled regulatory sequences along with the BT toxin gene, an approach that is beyond the reach of current techniques. But with increased understanding of the mechanisms of gene regulation, it may be possible in the near future (21).

A second approach, while not so obvious, employs tactics that can be used effectively and immediately, though the logistics of this approach may complicate existing methods of planting and harvesting somewhat. It draws upon studies in game theory of evolutionary stable strategies (12,13,14,58). Some biologists who have studied the relationships between pathogens or pests and their hosts feel that pathogens or pests may adapt more quickly to the defenses of agricultural crops than to those of hosts in the natural environment (1,32). The key seems to lie in the differences in genetic variation in the two populations. Variation is higher in natural than in agricultural populations, because the latter often involve huge areas of genetically homogeneous host plants. The selection pressures in agricultural systems are more often even and continual, in marked contrast to the spatial, temporal, and other variables that complicate selection pressures in the natural environment. These even, continual selection pressures are much easier to adapt to. Thus, anything that could be done to increase the genetic variation (at least in the genes controlling pest defenses) of the host population would likely lead to a slower adaptive response in the pest.

Increasing genetic variation in agricultural populations could be accomplished by mixing genetically pest-resistant strains of crop plants with strains not so protected. Modeling studies show that as simple a change as planting a mixture of 50 percent resistant and 50 percent unprotected seed in place of 100 percent resistant seed would substantially extend the time it takes for the pest to mount an adaptive response (32). The same principle also applies to other host/pathogen relationships, such as wheat/rust or corn/blight, or integrated pest management in general. Indeed, the relationship could be extended to planting crops that vary with respect to their tolerance to environmental factors (drought, cold) in areas where wide annual fluctuations in these parameters occur. The net effect could well be an increase in crop yields or productivity, although most often there is a trade-off between yield and the degree of resistance or tolerance. Just as complex communities seem best to resist the perturbing effects of introduced species (17), so might genetically complex agricultural systems better resist many types of potential environmental challenge.

Other types of genetic modifications to plants, such as altering photosynthetic pathways to increase efficiency and production rates, or modifying seed components to resist insect predation and loss during storage, may well have environmental effects of an unforeseen nature. But much larger changes in photosynthetic rates or biomass productivity can be achieved more easily by planting different crops, something that takes place constantly without review for potential environmental impacts. It hardly seems logical, therefore, to review engineered plants for such an effect when traditional agricultural crops are not subjected to such review.

Insect Communities

A great deal of research in geneticall, *altered* plants and microbes focuses on ways to combat insect pests—a major destroyer of both field and stored crops. Relatively little work is presently being done with insects per se. Some plant modifi-

cations for controlling insects were discussed in the preceding section. This section describes two representative modifications to insect-targeted microbes: those intended to reduce crop losses to the black cutworm, and modifications to viruses that parasitize certain insect species.

Significant corn crop losses each year are attributable to one pest species—the black cutworm. This lepidopteran larva feeds on the roots of corn plants. To combat this pest, researchers are exploiting both its sensitivity to the delta-endotoxin from B. thuringiensis and its narrow host range of corn roots. Their work involves engineered versions of the common bacterium *Pseudomonas fluorescent*.

Some strains of P. *fluorescent* live in the rhizosphere formed by the interaction between corn roots and associated filamentous fungi. Scientists at Monsanto have used a specific transposable element, Tn5, to insert the delta-endotoxin gene into the chromosome of this bacterium. This transposable element is not common in nature (6,41). Monsanto scientists have disarmed it in two independent ways to make further movement after insertion unlikely (41). In addition, frameshift mutations were inserted to further decrease the possibility of horizontal gene transfer due to reversion or complementation.

Greenhouse tests suggest that if corn seeds are coated with the altered *Pseudomonas*, the bacterium will colonize the emerging roots upon germination. Preliminary results also indicate that the engineered bacteria do not disperse significantly beyond the rhizosphere habitat in which they are naturally found, nor do they seem to persist beyond the end of the corn growing season. All these factors, coupled with the known, safe record of the BT toxin, suggest that this environmental application is safe enough to be field tested on a small scale. Such a test would measure survival, dispersal, and efficacy under realistic field conditions, and could be expected to provide useful lessons for other microbial applications.

Another early microbial application aimed at insect pests uses the host specificity of a class of viruses known as baculoviruses. These have relatively narrow host ranges, usually one or a few closely related insect species. Different viruses are specific for different pests, including the cabbage looper (a moth larva) and the pine sawfly, both of which threaten agriculture and forestry in the United Kingdom.

During 1986, researchers at the Institute of Virology in Oxford concluded a preliminary field test of a cabbage-looper virus. To track the released virus, the researchers inserted specific, marker DNA sequences into the engineered organism. Similar tests are planned for a virus specific for the pine sawfly.

Scientists hope that learning more about the genetics of host specificity in such viruses will enable them to target viruses precisely to specific pests. But the difficulties of tracking a virus in the environment, the possibilities for dispersal, the ability of viruses to remain infective for long periods, and the possibility of mutations disrupting the host range of released forms must be considered before wholesale applications are undertaken. When asked about the degree of risk attending such work, one researcher responded,

My guess is no problem, but there are a variety of constructs possible, and some could have broader effects than desired . . . One can consider many scenarios. In the worst case (also the most improbable), the situation could not be corrected (36).

One of the least predictable effects of environmental alterations has been termed the "cascade effect ." Such effects are not likely to be common consequences of planned introductions, and quite unlikely to be associated with small-scale fieldtests. But they are known to have been associated with some large-scale environmental activities in the past, so a brief description is in order. The term '(cascade effect" describes a community disruption in which a perturbation in numbers (or even loss) of one species triggers reverberations throughout the community, These effects could alter predator/prey relationships or significantly change community structure even if ecosystem processes themselves are not altered.

Cascade effects can be both surprising and counterintuitive. One example can be found in the case of a World Health Organization program to control insect pests. The program used large-scale applications of DDT in Borneo to kill house flies. The local lizard population, however, could not distinguish between flies that died of natural causes, and were safe to eat, and those that succumbed to the spraying program, and therefore were not. Large numbers of geckos ate the flies and died, as did cats that ate the poisoned geckos. The reduction in the cat population allowed rat numbers to rise, Stored food supplies suffered, and the increased numbers of rats brought an increase in the numbers of rat pest species. The result: an outbreak of bubonic plague, a public health problem that would not normally be expected to result from a program of insect eradication (30.

Although such chains of events are almost wholly irrelevant to considerations of field-test safety, they should be kept in mind when the largescale applications that will follow are contemplated. It should also be noted that the negative consequences of the house-fly eradication program resulted from an accumulation in the food chain of an enduring, toxic compound. Widespread applications of genetically engineered organisms are generally expected to reduce reliance on compounds of this sort. Also, most other examples of cascade effects involve perturbations of natural, not agricultural systems. Great care must be taken in extrapolating from nature to the artificial environments found in agriculture. Nevertheless, large-scale applications of insecticides have sometimes increased the numbers of insect pests when beneficial insects are eliminated along with the pests (35).

Examples of cascade effects resulting from use of a chemical pesticide bear little relation to the problems most likely with planned introductions of genetically engineered organisms. They do, however, illustrate that complex relationships govern the interplay of organisms in ecosystems relationships that might not be comprehended with cursory review, and that are not always immediately apparent.

Microbial Communities

Although some concern is raised over possible consequences of introducing engineered animals or plants, greater public apprehension is associated with possible uses of engineered microbes (ch. 3). Planned applications include microbes altered to prevent plant damage by herbivorous insects (as just discussed), protect crops from frost damage, increase nitrogen available to plants, and, eventually, to degrade toxic wastes (covered later in this chapter). These introductions may take place either in limited agricultural settings or in broadcast environmental applications.

The greater concern over planned microbial introductions has less to do with a higher probability of risk than with greater uncertainty about some factors in microbial ecology and the requirements of monitoring or tracking that are peculiar to microbes. Extensive experience with past microbial introductions does not suggest that potential problems are worse than those associated with plants or animals. But few of the microbes living in soils can be cultured in the lab, and little is known about them or their relationships beyond the broad outlines of morphology and apparent function. Microbial ecologists do know, however, that there is a high degree of functional redundancy among members of microbial communities, a redundancy that should act to mitigate any general consequences of perturbing a particular microbial population, although some limited communities (e.g., degraders of lignin) may be less protected by such redundancy. Furthermore, a substantial literature suggests that microbial systems are resilient in the face of perturbations, and that it is often difficult to produce a measurable impact, even by design, on such populations.

Developing environmental applications with microbes affords certain advantages over working with plants or animals. Enormous populations (numbering in the billions) can be studied over hundreds of generations on a rapid time scale. On the other hand, microbes present some unique problems. They are microscopic, and the techniques for tracking their movements, distribution, and numbers are often more involved, or more tedious and expensive, than for plants or animals.

Microbes, particularly those in soil, can be difficult to detect. The most sensitive available techniques for sampling populations, based on detecting microbial DNA, are difficult or impossible to apply if the DNA to be tested is taken from fewer than a thousand cells, Smaller numbers—as low as 100 cells per cubic centimeter—may be assayed if bacterial colonies are cultured from naturally occurring individuals and screened with techniques that rely upon resistance to specific antibiotics, or other biochemical markers. This assumes, however, that the species can be grown in the laboratory.

Bacteria can persist in a microbial community at levels undetectable by any existing sampling technique. These levels can rise rapidly under favorable conditions, so the inability to detect a microbe does not mean that it is absent from an environment. Microbial species are enormously abundant—upwards of a thousand different species may be found per square yard of land surface (9). A single species may encompass from 5,000 to 20,000 genetically different strains or varieties, varying in their adaptive qualities and ecological requirements.

Microbes are also ubiquitous, found in every terrestrial environment and habitat in which they have been sought, including such hostile and unlikely sites as hot springs and subterranean aquifers (11,20). Perhaps 90 percent of these species cannot presently be cultured in the laboratory, making them difficult to study. Despite such obstacles, the ecological questions raised by the environmental release of genetically engineered microbes are not intractable, and a great deal of relevant historical experience can be studied.

Genetic alterations to microbes for specific environmental purposes are many and various. This section outlines two not covered elsewhere in this report. The first—the application of "ice-minus" bacteria to crop plants to protect against frost damage–involves what was one of the more contentious of the early applications for permission to field test, though for reasons other than the scientific questions involved (see ch. 3). The second, involving the inoculation of crops with enhanced nitrogen-fixing bacteria, has been less controversial.

The technical details of the ice-minus application are simple. Some bacteria contain, as a component of the cellular membrane, a protein encoded by a single gene in the bacterial chromosome. This protein can act as an efficient nucleus for the formation of ice crystals on the surfaces of plant leaves or blossoms where the bacteria live. With-



Photo credit: Peter Forde, Advanced Genetic Sciences

Advanced Genetic Sciences Researcher Julianne Lindemann spraying "ice-minus" bacteria on strawberry plants in test plot on April 24, 1987. Protective clothing was required by the California Department of Health Services. Note reporters and onlookers in immediate background where coffee and donuts were consumed without hesitation.

out such nuclei, water does not generally freeze at 32 "F. Rather, it supercools to between 5 and 100 below the "normal" freezing point before the formation of ice crystals begins to take place.

Agricultural losses to frost damage each year are variable but significant. Some estimates place the average annual loss at \$1.6 billion in the United States, and \$14 billion worldwide (15). Most of this loss results from frosts that take place near harvest time, or near flower-budding time in spring. Part of it might be avoidable if some protection could be provided against snap frosts, as opposed to hard freezes, because hard freezes are uncommon during the growing seasons of most crops.



protection could be afforded by deleting the "icenucleating" gene (or an essential part of it) from the major bacterial strains living on the surfaces of particular plants (potatoes and strawberries, in the first fieldtests). These altered bacteria would then be inoculated on the plant surfaces to be protected, in the hope that they would colonize them and replace the naturally occurring bacteria (designated ice plus, or INA +, for ice nucleation active).

Naturally occurring populations of ice-nucleating bacteria contain low numbers of individuals that are ice-minus. The phenotype can be produced by any mutation that inactivates the production of the critical protein, and thousands of such mutations are possible. Many could produce ice-minus bacteria in natural populations. The primary genetic difference between these naturally occurring ice-minus bacteria and those produced with recombinant DNA techniques is that the latter are produced by a specific technique that yields a consistent, precisely characterized, identifiable genetic deletion, Such consistency and identity is not a characteristic of the naturally occurring strains. These and other factors led both the National Institutes of Health and the Environmental Protection Agency to approve applications for preliminary, small-scale field tests of the bacteria, which were successfully completed in 1987.

Some scientists suggested a worst-case scenario that sounds like a cascade effect (42). According to that scenario (which presupposed large-scale applications, not small-scale field tests), large-scale agricultural applications of ice-minus bacteria might reduce the atmospheric reservoir of ice nuclei. These atmospheric nuclei are critical for precipitation, providing particles around which ice crystals can form so that droplets can grow large enough to fall as snow or rain. Dirt and dust can act as sources of ice nuclei, but vegetable or plant material is a better source (53,54). Another major source for ice nuclei is marine phytoplank ton, living in the top layers of oceanic waters (50,51). The sources of ice nuclei important to local precipitation are generally local (52,55), though long-distance dispersion of bacteria is known to be possible, if not common (19), and less likely for good nucleators than nonnucleators.

Some scientists asked if reducing atmospheric concentrations of ice nuclei could affect local rainfall, and perhaps even global weather, reducing or redistributing patterns of precipitation. They pointed to data suggesting that overgrazing in the Sahel may have exacerbated drought conditions in Sahelian Africa (43,49). Overgrazing reduces plant biomass in an ecosystem, thus reducing the suitable habitat for ice-nucleating bacteria, and reducing the numbers of bacteria. Such a cascade effect may have contributed to decreases in rainfall downwind of deforested areas.

Seeking to assess the likelihood of this scenario, OTA commissioned two analytical studies by groups taking slightly different approaches to the problem (4,60). Both groups made assumptions to produce a worst case scenario. Both concluded it is unrealistic to expect any significant negative impact on global climatological patterns from large-scale agricultural applications of ice-minus bacteria. The likelihood of local changes in precipitation patterns or densities is perhaps slightly higher, they concluded, but still extremely low. Indeed, to put the question in the appropriate context, the potential for negative environmental consequences from large-scale applications of iceminus bacteria should be compared with the potential for such consequences from other strategies for reducing or eliminating INA + bacteria in agriculture, such as the use of chemical bactericide. Several significant uncertainties in these analyses could be resolved, however, by further research and experimental studies.

In another planned introduction of engineered microbes, rhizobial bacteria are being studied in an effort to increase nitrogen fixation in soil, and thus to decrease reliance on costly fertilizers. Scientists at BioTechnica International (Cambridge, MA) have engineered a strain of *Rhizobium meliloti*, symbiotic on alfalfa roots, to increase its production of nitrogen. This has been accomplished by increasing the number of promoter sequences that regulate the activity of genes involved in nitrogen fixation metabolism, Greenhouse tests suggest possible increases of available nitrogen as high as 17 percent, and though field tests were originally planned for 1987, they were later postponed until 1988. A long history of agricultural

inoculations with different varieties of nitrogenfixing bacteria suggests that far-reaching or negative environmental consequences are quite unlikely, though this question is explored in more detail later in this chapter.

Some individuals believe other approaches to nitrogen fixation may lead even more quickly to practical improvements. This feeling stems partly from the fact that nitrogen fixation is an energyintensive process. Sufficient energy may not be available under most conditions to sustain appreciable increases in nitrogen fixation by microorganisms residing on or around root surfaces. It is also true that bacteria that are good modulators or colonizers of roots also tend not to be the most efficient fixers of nitrogen, though there is tremendous variation among naturally existing strains.

Improving the energy efficiency of these bacteria is, of course, one object of research. But re-



search efforts might profitably be invested in improving the ability of nitrogen-fixing microbes to withstand the stresses encountered in their rhizobial environments and in the processes of storage and agricultural inoculation, Most bacteria now marketed as inoculants fare poorly under these conditions, especially in the tropics, with the result that most attempted inoculations deliver only a small fraction of the intended microbes to the application site. Research in these areas might dramatically improve the success of inoculation techniques while effectively exploiting the existing reservoir of variation among naturally occurring rhizobial bacteria in different environments (2,3).

Aquatic Communities

Work on altering aquatic community species is being pursued in a number of areas. Potentially fruitful lines of investigation involve fisheries biology and aquiculture. At least two different research groups are exploring methods to enhance the production of "trophy" salmon in the Great Lakes region and the Pacific Northwest (see app. A). Under a flexible definition of biotechnology, this research may qualify for consideration in this report, or it may not be considered to differ significantly from past fisheries practice.

The technique used to try to produce the trophy salmon is called "heat shock." During an early, sensitive stage of embryonic development, salmon eggs are briefly subjected to unusually high temperatures (38 degrees Celsius): temperatures high enough to disrupt some metabolic processes, but not so high as to kill the developing fish embryo. This environmental insult disrupts normal cell division, causing a spontaneous doubling of the chromosomal number in each cell of the developing embryo.

Chromosomal doubling triggers a series of physiological effects, starting with a disruption of the

POTENTIAL IMPACTS ON ECOSYSTEM PROCESSES

Ecosystems are enormously complex, and to reach a clear understanding of how they function is commensurately difficult. Their complexity stems from the many and diverse interactions hormonal system. The fish fail to mature sexually and remain sterile throughout their lives. This frees them from the normal cycle of growth, maturation, sexual reproduction (spawning), and death that usually limits their lifespan. Because these fish fail to spawn and die, they are expected to continue to live and grow, eventually reaching record sizes.

Salmonid fish are among the top predators in whatever community they inhabit. So initially it may appear that the planned introduction of these fish would violate a cardinal principle mentioned earlier: Do not introduce polyphagous species, particularly if they occupy a high position in a food chain (45). But these fisheries are already managed by humans. Even under generous assumptions about the survival of released fingerlings, the resulting adults are unlikely to number more than several hundred among stocked populations that number in the millions (33). Furthermore, the same property that causes these fish to reach such large sizes prevents them from reproducing. It is impossible that any environment will ever contain these fish except through planned stocking. Careful monitoring of the fish's release and growth rates should help to avoid or correct serious unanticipated effects.

Other work with aquatic communities focuses on marine algae, to enhance the rates at which they produce substances useful in food preparation, such as carrageenan or agar (see app. A). Some aquatic algae have also been considered as potential agents to extract toxic compounds or heavy metals from aqueous solution (59). It might even be possible to exploit algae for mining or mineral recovery operations. But most of these applications involve aquatic plants that function in their environments as primary producers. Genetic alterations to improve or adjust their rates of activity could well affect other members of aquatic communities. Such possible effects are discussed in the following section.

within the numerous populations of any given species, and between the great numbers of different species that form any biological community. It is also due to the links between the physiological processes of organisms and the geological processes by which minerals are derived, distributed, and moved in the physical world. For example, the availability of minerals liberated from rocks is affected by living things (especially plants or microbes) as they influence rates of erosion.

As a way of reducing this daunting complexity to manageable proportions, some ecologists devote more study to the distribution and movement of a small number of vital elements and minerals than to the activities and attributes of living creatures. The most important of these vital elements are carbon, nitrogen, phosphorous, and sulfur. These elements are "rate-limiting." In other words, the amount of these elements in biologically accessible forms determines the total tissue of living organisms (biomass) that can be assembled in the ecosystem. (Phosphorous and sulfur are covered in detail in 9,22.) Carbon and nitrogen are discussed in this section.

Ecosystem processes are the vehicles by which these rate-limiting elements and their different molecular forms move, or cycle in ecosystems. The two major ecosystem processes are nutrient cycles and energy flow. These processes can be, and sometimes are, considered separately. This report will consider them together, and deal with them primarily in the language of mineral or nutrient cycles.

Every time an element is altered from one molecular form to another, or exchanged between different organisms, some chemical bonds are broken and others formed. Each of these actions either consumes or liberates energy. The integration of countless individual events of this sort is the immediate mechanism by which nutrients cycle and energy flows through an ecosystem.

The major factors driving ecosystem processes are the production of energy and the conversion of carbon into biological forms by photosynthesis, or carbon fixation. Where carbon is ratelimiting, it places an absolute upper limit on the biomass a system can support. Under such circumstances, competition for carbon is intense, as for example among soil microbes. Plants produce most of the biologically accessible forms of carbon.

Decomposers-organisms that degrade plant material—play the major role in moving carbon from one reservoir into another (e.g., from one plant, through decay, into another). Most decomposes are invertebrates (insects, nematodes, and so on) or microbes (bacteria and fungi), living primarily near the soil surface in what is termed the "litter layer" of detritus that accumulates from above, or just below, in the topsoil. Larger herbivores, or vertebrates, play a smaller role, usually assisted by an intestinal flora of symbiotic microbes.

Physical parameters, such as climate, soil quality, and available moisture, affect the distribution of both plants and decomposes. As a result, a range of ecosystems—different communities of plants and decomposes, associated with particular ecological qualities—has evolved. Table 5-3 illustrates several different terrestrial ecosystems and presents data indicating the patterns of carbon storage and movement in them.

Nitrogen is the nutrient most often cited as ratelimiting in terrestrial ecosystems. Although one form of nitrogen makes up nearly 80 percent of

	Table	5-3.—	Production	and	Decom	position	in 🗄	Six	Ecosy	/stem	Types
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	Mean NPP tons/ha/yr	Mean biomass tons/ha	Litterfall tons/ha/yr	Litter accumulation tons/ha	Decomposition k/yr
Tundra	1.5	10	1.5	44	0.03
Boreal forest	7.5	200	7.5	35	0.21
Deciduous forest	11.5	350	11.5	15	0.77
Grassland	7.5	18	7.5	5	1.50
Savannah	9.5	45	9.5	3	3.20
Tropical forest	30.0	500	30.0	5	6.00

Abbreviations:NPP -= net primary productivity, the total amount of living material produced by the organisms in an ecosystem; ha -hectare; yr = year; k = fractional weight loss of litter material (a value of 1 means the rates of production and decomposition are equal).

SOURCE: J.R.Gosz, C.N.Dahm, and P.W. Flanagan, "Ecological Impact of Genetically Engineered Organisms on Ecosystems," contract report prepared for the Office of Technology Assessment, U.S. Congress, 19S7.

the atmosphere, biologically accessible forms are more limited.

The nitrogen cycle—illustrated in figure 5-l is the most complex major nutrient cycle because the element can exist in so many qualitatively different molecular forms, Abiotic factors and the actions of living things influence the production and persistence of these forms.

While plants can absorb gaseous nitrogen or nitrogen dissolved in water through their roots, microbes provide the major pathway for nitrogen incorporation into living material. Specifically, rhizobial bacteria are responsible for nitrogen fixation in agriculture. These microbes live in very close association with the roots of certain plants (legumes), often in small granules or nodules attached to the roots. In essence, they deliver fixed nitrogen directly to the host plant. The biochemical pathway resulting in nitrogen fixation is large, complex, and energetically expensive to operate, involving at least 17 different structural genes, with associated regulatory sequences. This genetic package exists in similar form in all rhizobial bacteria, an evolutionary conservation suggesting that the pathway is quite important to the bacteria. Scientists trying to enhance nitrogen fixation face the formidable task of improving the performance of a genetic sequence that has been refined by natural selection for billions of years. Nevertheless, researchers are making the attempt, even trying to transfer the complex of nitrogen fixa-



The nitrogen cycle: processes in terrestrial and aquatic systems. The microbially mediated processes include mineralization, vitrification, denitrification, immobilization, and N-fixation. SOURCE: Office of Technology Assessment, 1988.

Figure 5-1 .— The Nitrogen Cycle

If the nitrogen-fixing abilities of rhizobial bacteria were enhanced in such a way as to make the resultant increase in available nitrogen general throughout the host environment, rather than restricted to the host plant per se, a variety of environmental consequences might result. The potential consequences of such a successful application are listed here (22), in order of decreasing probability.

- I. a) Increased leaf and shoot production in target plants
 - b) Decreased root growth and biomass
 - c) Increased decomposition rates in some systems
 - d) Increased ammonification and vitrification in soil
 - e) Increased competition on nonnitrogenfixing plants and alteration of plant community structure, with reverberations throughout the associated ecological community
- 2. a) Decreased decomposition rates in some systems
 - b) Increased potential for other element limitations, especially if 2a results
 - c) Altered chemical exchange capacities and pH shifts
- 3. a) Decreased mycorrhizal associations (dependent on 2b)
 - b) Shift from carbon-based plant defense to nitrogen-based defense (or from immobile to mobile defense compounds)
 - c) Altered foliage tissue palatability and herbivory (dependent on 3b)
 - d) Increased terrestrial denitrification rates and increased emissions of Nitrogen gases (dependent on 1d and 2c)
- 4. a) Increased leaching loss of nitrate
 - b) Increased eutrophication of drainage water
 - c) Increased algal production in receiving waters
 - d) Increased carbon enrichment of aquatic sediments
 - e) Increased anaerobic conditions in aquatic habitats

 f) Increased aquatic vitrification and denitrification rates and gaseous emissions to the atmosphere.

Some data suggest such consequences are not purely hypothetical, that under very special conditions they might actually be likely (63). Generally, however, such a concatenation of consequences rests on a series of assumptions. For most of them to occur, the introduced rhizobia would have to move beyond agricultural lands. They would then have to successfully nodulate, or colonize, wild plants, and provide substantially more nitrogen to those wild plants than is already provided by native rhizobia, This also requires that the host specificity of the introduced rhizobia would have to change genetically so they could infect the wild plants, and the introduced bacteria would have to be successful in modulating the native plants in competition with native bacteria. As cited above, considerable literature and experience demonstrates the remote likelihood of each of these assumptions; to achieve all of them at once, the probability is exponentially less likely. Indeed, a review of these assumptions and potential events leads many experts on nitrogen fixation to believe it would be difficult to conceive of a planned introduction less likely to have negative consequences than introducing engineered varieties of nitrogen-fixing bacteria.

Ecosystem processes reflect the sum total of the actions of all living things in the system. Individuals concerned about planned introductions of genetically engineered organisms fear applications that might affect such fundamental processes as nitrogen or phosphorous cycles. And although basic changes to an ecosystem process would be difficult to accomplish, it might be easier to affect changes in rates of flow of some components through a portion of the process. By the time such effects were noticed, however, a community might already have been substantially altered. According to two researchers in this field,

... the quality of an environment can change markedly with no significant change in gross measures of the rates of processes. Fish production in two lakes may be the same (as lbs./year) when one produces mostly rainbow trout and the other mostly carp, but no human would say that the two lakes are therefore indistinguishable with regard to fish (9). Attempts to detect such changes in ecosystems have led to the study of so-called indicator species, species more sensitive than other organisms to environmental changes (40).

The use of indicator species requires the identification of species that are particularly sensitive to the environmental parameter of interest—for example, the presence of toxic compounds or the abundance of specific nutrients. Indicator species can give early warning of an impending change, allowing preemptive measures to be taken. Without such early warnings and preemptive action, the task of restoring a disrupted ecosystem is difficult, expensive, and slow, if even possible (38).

A better understanding of community relationships should make indicator species increasingly useful, especially in risk assessment and management. Nevertheless, any planned introduction that is likely to alter a fundamental ecosystem process should undergo careful scrutiny. None are planned in the foreseeable future.

Possibly of eventual concern, however, are applications intended to correct existing serious, generally recognized environmental problems, such as the presence of toxic chemicals. Naturally occurring microbes exist that can degrade many different, complex, toxic compounds—herbicides and pesticides, industrial solvents, wood preservatives, plasticizers, dyes, etc. (22,26). Biotechnology researchers are trying to enhance these natural degradative abilities in some microbes and introduce them into others. Success in such efforts could help significantly in decreasing the toxic waste problem that promises to be so difficult and expensive to resolve (61).

Cascade effects might be triggered by the structural similarities between naturally occurring plant materials and some of the complex organic compounds found among toxic wastes. If degraders were engineered and introduced without sufficient constraints upon their specificity, they could function as ecological generalists, breaking one of the foremost rules for introducing organisms into new environments. Communities of decomposers may also be more vulnerable to perturbations than most microbial communities, because the decomposition of lignin and cellulose (the structural components that make up from 50 to 90 percent of the biomass of higher plants) is carried out by a relatively narrow range of microorganisms. A disruption of the population dynamics of one species of decompose, in a limited community of decomposes made up of only a small number of species, could conceivably have a significant impact on the community.

In contrast, many other microbially mediated processes are carried out by the members of much larger, more diverse, and therefore more buffered, microbial communities. Indeed, no existing evidence indicates that populations even of microbial degraders are subject to cascade effects following perturbation. Such scenarios are purely speculative. In fact, some researchers in this area feel the introduction of engineered microbes to help in the degradation of organic pollutants is sufficiently distant that it need play no role in current deliberations over the safety of planned introductions, although similar introductions of naturally occurring organisms are not at all uncommon.

Finally, it is important to keep the possible consequences of planned introductions of genetically engineered organisms in perspective. As one group of ecologists has concluded, "Our analyses and remarks to this point may give the impression that we are against the release of any genetically altered organisms into the environment, This is not our view. In fact, we are enthusiastic about the prospects offered by biotechnological solutions to environmental problems ranging from increased productivity of agricultural systems, to the control of pests and pathogens, and the removal of many chemical toxins from the earth's soils and waters. We further expect that many, if not most, environmental applications of biotechnology can be made into safe and productive ventures" (28). Risk assessment and management, discussed in chapter 6, are crucial to that undertaking.

The major concern of some scientists over the planned introduction of genetically engineered organisms stems from the potential for unexpected or unforeseen consequences; other scientists believe that the likelihood of such consequences is no different, or even lower, for engineered organisms than for varieties or cultivars produced by widely accepted methods. A variety of disruptions to local populations or to more general ecosystem processes could result, but do not seem likely to result from any of the planned introductions now contemplated. Experiences with past introductions of organisms into new environments provide some limited clues to the nature of those disruptions, but a much better analogy for planned introductions of engineered organisms likely in the near future is that with new crops or cultivars in agriculture.

Only a small fraction of introduced organisms have become pests or have significantly affected their new environments. These species-called colonizing species, or weeds-differ significantly in their adaptive capabilities from most genetically engineered organisms intended for planned introductions. For the near future, engineered organisms generally will differ from nonproblematic parental strains by only one or a few structural genes. There are, however, several examples of single gene changes affecting the virulence or host range of a parasite or pathogen. Although engineered organisms are no more likely than nonengineered ones to be susceptible to such changes, until sufficient experience is gathered caution is indicated.

Environmental disruptions stemming from the deliberate release of engineered organisms might take place at any level—from the local population to ecosystem processes of energy flow or nutrient cycles. Plant introductions seem least likely to **result** in problems because of the low mobility of plants and restricted horizontal gene flow between them. Animal communities, because of the greater mobility and higher propensity for gene transfer, seem slightly more vulnerable to disruption, though the probability still seems quite low. Furthermore, most anticipated animal applications involve livestock, suggesting that the potential for disruptions of natural communities is not great.

Most difficult to assess are applications involving microbes. Microbial mobility is low but dispersibility is relatively high, as is the potential for gene transfer. Much **remains to be learned about the composition and relationships among natu**rally occurring microbial **communities (of which only a small portion of the members can be cultured** in laboratories) before general methods of risk assessment will be available (see ch. 6). But a substantial body of experience suggests that microbial introductions are not likely to produce problems.

The least likely, but potentially most serious ecological impacts involve the disruption of ecosystem processes, such as energy flow and nutrient cycling. Such disruptions could be difficult to reverse and far-reaching in their effects. They could result as a consequence of engineering microbes to increase their capabilities as degraders, e.g., to enhance the decomposition of woody tissues containing lignin or cellulose, or to help in the cleanup of toxic organic wastes. In the latter case, however, effective control might well be exerted by managing the available supplies of carbon, which are likely to constrain degradation rates. The potential problems associated with such enhanced degraders must be weighed against the existing, serious problems associated with toxic wastes and the chemical technologies that are their source.

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