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To the Chairman, Committee on Science
and Technology, House of
Representatives

October 1985

BIOTECHNOLOGY

The U.S. Department of Agriculture's Biotechnology Research Efforts



BRIEFING REPORT



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UNITED STATES GENERAL ACCOUNTING OFFICE
WASHINGTON, D.C. 20548

RESOURCES, COMMUNITY,
AND ECONOMIC DEVELOPMENT
DIVISION

October 25, 1985

B-220899

The Honorable Don Fuqua
Chairman, Committee on Science and Technology
House of Representatives

Dear Mr. Chairman:

Your letter of March 29, 1984, requested that, among other things, we obtain information on the extent of the U.S. Department of Agriculture's (USDA) biotechnology research efforts. In discussions with your office, we agreed to identify and document all of the biotechnology research projects being funded in whole or in part by USDA, because this information was not readily available within USDA or from any other source.

USDA-funded biotechnology research is conducted primarily in USDA's own research facilities directed and operated by the Agricultural Research Service (ARS) and in facilities at state agricultural experiment stations or colleges of veterinary medicine that receive a portion of their funding from USDA's Cooperative State Research Service (CSRS). USDA's Office of Grants and Program Systems (OGPS) provides additional funding for biotechnology research that is conducted at these same institutions as well as other institutions.

Centralized data of the type your committee was interested in were not available for CSRS-funded projects. Therefore, we sent a questionnaire to each state agricultural experiment station and each college of veterinary medicine to obtain information on such things as funding and staffing levels; research objectives and results of USDA-supported biotechnology research; and whether genetically engineered organisms were expected to be released into the environment. ARS and OGPS officials agreed to provide us with similar information on biotechnology projects funded by ARS and OGPS although, as discussed with your office, the information provided by ARS and OGPS was not as comprehensive as the data provided through the questionnaire.

We found that

--At the time of our review, USDA was funding, in whole or in part, 778 biotechnology research projects. The amount of USDA funding either spent on these projects in fiscal year 1984 by CSRS and OGPS or planned to be spent in fiscal year 1985 by ARS totaled \$40.5 million.

--The state agricultural experiment stations and colleges of veterinary medicine reported to us, through use of the questionnaire, that they were conducting 495 biotechnology projects funded in whole or in part during fiscal year 1984 by USDA. The amount of such funding totaled \$10.7 million. For these projects, we learned that

-A variety of biotechnology techniques was being used in the research. The technique known as recombinant DNA (this term and other technical terms are defined in app. II) was being used in 267, or 54 percent, of the projects. This technique has caused considerable concern to people who are worried about the risks and potential negative consequences of biotechnology.

-Of the 495 projects, 87 (or 18 percent), conducted in 28 states, were expected to involve the deliberate release of genetically engineered organisms into the environment (11 within 1 year, 47 within 2 to 5 years, and 29 after 5 years). The scientists working on these projects generally stated that the planned releases will cause no problems, although in three cases they said they did not know. The scientists also stated that any problems that might result from such releases would generally be controllable. In one case, however, the principal scientist stated that "One can consider many scenarios. In the worst case (also the most improbable), the situation could not be corrected." The scientist told us later that her experiment would not involve a release as long as there was a potential danger. She said that before there would be an approval for release, the experiment will have to undergo careful scrutiny and testing according to specific federal guidelines.

--In October 1984 ARS was conducting 183 biotechnology research projects with an estimated cost in fiscal year 1985 of \$26.4 million. Information provided to us by ARS did not identify the biotechnology research techniques used or which of the projects were expected to result someday in the deliberate release of genetically engineered organisms into the environment. We were assured, though, by ARS' Assistant Administrator for Cooperative Interaction that no such release would be approved without careful scrutiny.

--OGPS funded through its competitive grants program 145 biotechnology research projects at a cost of \$4.8 million in fiscal year 1984. Of these projects, 45 (representing an annual cost of \$1.4 million) duplicated projects reported to us by state agricultural experiment stations and by ARS. With respect to the 100 projects that were not duplicates, recombinant DNA was the prevalent technique used. An OGPS official identified 4 of the 100 projects as ones expected to involve a deliberate release of

genetically engineered organisms into the environment over the next 5 or so years.

Appendixes I through XI provide more information on the results of our work. Officials from ARS, CSRS, and OGPS were given the opportunity to comment on a draft of this report. A number of changes were made to clarify information in the report on the basis of the comments received.

As arranged with your office, unless you publicly announce its contents earlier, we do not plan to distribute this report further until 14 days from its issue date. At that time we will send copies to interested parties and make copies available to others upon request.

Sincerely yours,

A handwritten signature in cursive script that reads "Brian P. Crowley". The signature is written in dark ink and is positioned above the typed name.

Brian P. Crowley
Senior Associate Director



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ABBREVIATIONS

ARS	Agricultural Research Service
CSRS	Cooperative State Research Service
DNA	deoxyribonucleic acid
EPA	Environmental Protection Agency
GAO	General Accounting Office
NASULGC	National Association of State Universities and Land Grant Colleges
OGPS	Office of Grants and Program Systems
USDA	U.S. Department of Agriculture

INFORMATION ON USDA'S BIOTECHNOLOGY RESEARCH EFFORTS

Biotechnology--broadly defined to include any technique that uses living organisms (or parts of organisms) to make or modify products, to improve plants or animals, or to develop microorganisms for specific uses--has, in fact, been in existence for a long time. Historically, the manipulation of plants and animals to benefit mankind began with primitive agricultural societies. For example, humans were unknowingly exploiting the ability of microorganisms to convert sugar in grape juice into alcohol in wine; to break down the proteins in milk to soften and flavor cheese; and to convert the starch in flour into carbon dioxide, which causes bread to rise during baking.

During the past few years, however, the term has taken on new meaning as new techniques used in genetic manipulation have been developed that greatly enhance both the rate and potential degree of innovation. The new techniques focus not on the whole plant or animal, but rather on the cellular and subcellular levels of plants, animals, and microorganisms. At least some of the new techniques--recombinant DNA¹ being one of them--bypass the sexual reproduction process and make it possible to move genes from one organism to another (related or otherwise). Such control by mankind over the fundamental characteristics of organisms raises questions about the relationships of humans to other living things and to the environment as a whole. The potential for mankind to alter genetic traits in a directed fashion is seen by some as a challenging opportunity with benefits expected to include plants with greater disease resistance; bacteria that enhance the nitrogen-fixing capability of plants; and microbes that detoxify hazardous wastes, clean up oil spills, or facilitate the recovery of minerals from the ground. Other people, however, for ethical, moral, religious, or scientific reasons, respond to the new techniques with vague unease or see such techniques as capable of producing consequences that threaten public health and/or the environment.

OBJECTIVES, SCOPE, AND METHODOLOGY

Our overall objective in this phase of our work was to inventory the biotechnology research being funded in whole or in part by USDA. Information regarding such things as the number,

¹DNA (deoxyribonucleic acid) is the genetic material found in all living organisms. Recombinant DNA techniques involve joining together pieces of DNA from different organisms or synthetic DNA in vitro (outside the living body in an artificial environment), thus producing hybrid DNA. (These and other technical terms are defined in the glossary in app. II.)

location, funding level, and objectives of specific biotechnology research products was generally not readily available within USDA.

To obtain information on the biotechnology research funded in whole or in part by CSRS in fiscal year 1984, we--in conjunction with the National Association of State Universities and Land Grant Colleges (NASULGC)--developed a questionnaire, a copy of which is included as appendix III. In appendix IV we provide a brief description about NASULGC and our joint effort in designing and administering the questionnaire.

The questionnaire was sent to all 58 state agricultural experiment stations (University of California experiment stations at Berkeley, Davis, and Riverside were counted as individual stations), 55 of which responded. USDA and NASULGC officials told us that the three stations not responding (College of Virgin Islands, American Samoa Community College, and College of Micronesia) were unlikely to be performing any biotechnology research. Of the 55 that did respond, 50 reported USDA-funded biotechnology research at their institutions; 5 (Alaska, Arkansas, District of Columbia, Guam, and Nevada) reported that they were doing no such work. We additionally contacted and/or sent the questionnaire to all 28 colleges of veterinary medicine. Five reported USDA-funded biotechnology research over and above that being reported through the state agricultural experiment stations. The others either did not respond to the questionnaire (there were 4 of these) or informed us that their biotechnology research had been reported through the experiment station or that they had no such research underway. See appendix V for a listing of all state agricultural experiment stations and colleges of veterinary medicine.

The questionnaire was pretested. Nevertheless, some questionable responses were received that were subsequently reviewed by the late Dr. F. Aloysius Wood, Chairman of the NASULGC Committee on Biotechnology and Dean for Research and Associate Director of the Florida Agricultural Experiment Station, University of Florida; or by Dr. Charles E. Hess, member of the NASULGC Committee on Biotechnology, and Dean of the College of Agricultural and Environmental Sciences and Associate Director of the California Agricultural Experiment Station, University of California (Davis). Questionnaires were excluded if the research reported did not reflect the expenditure of any USDA funds and/or the biotechnology techniques we specified (or other closely related techniques) were not being used in the research.

The questionnaire was not used to obtain information from ARS and OGPS with respect to the biotechnology research they were conducting or sponsoring. Rather, we were told by USDA officials that these agencies could provide us such information directly. ARS provided us with A Compendium of Biotechnology Research in the

Agricultural Research Service, dated February 1985. This compendium was developed to describe the extent of biotechnology research in ARS and to serve as an index and cross reference for scientists and administrators. OGPS provided us with data sheets on the biotechnology research projects it was funding through its competitive grants program. Some of these projects duplicated projects reported to us by the state agricultural experiment stations and ARS. We attempted, with help from an OGPS official, to identify the projects so as not to count them twice in our reporting.

The information we received from ARS and OGPS was not as extensive as what we received through our questionnaire at the state agricultural experiment stations and colleges of veterinary medicine for several reasons. ARS' information, for example, had been assembled for purposes other than ours. Additionally, some staffing, funding, and other information obtained from administrators and scientists with respect to specific institutions and research projects turned out not to be readily available at the headquarters offices of ARS and OGPS.

For purposes of our work we--in consultation with NASULGC and USDA officials--defined biotechnology research as

"The process of in vitro alteration of genetic material for the purpose of creating new gene combinations or modifications."

In this regard we asked for information on research involving the following biotechnology research techniques.

--Direct manipulation of the genome (the total DNA complement of a cell) using recombinant DNA, chemical synthesis of nucleic acids (production of nucleic acids by combining simple molecules rather than using whole organisms), and/or site-directed mutagenesis (the focused induction of mutation in an organism's genetic material). These techniques are often referred to as genetic engineering--although the term could apply to the other techniques as well.

--Direct manipulation of cells (altering genetic information) using microinjection, transfection, transformation, embryo transfer, and/or cell culture and protoplast fusion. (See glossary in appendix II for definitions of these terms.)

This is the definition that was generally followed by those from the experiment stations, veterinary colleges, and OGPS that provided us with biotechnology research information. ARS, however, in preparing its compendium used a slightly different definition of biotechnology research. We were assured by

Dr. Hess, representing the NASULGC, and by ARS' Assistant Administrator for Cooperative Interaction, that the difference in the two definitions was minimal and that, irrespective of this difference, projects identified as biotechnology research on the basis of either of the two definitions would have been essentially the same.

Our work was conducted during the period September 1984 through August 1985.

BACKGROUND: USDA'S RESEARCH NETWORK

Food and agricultural research has made significant contributions to a wide range of agricultural and societal needs. Such research, accomplished through a federal/state research partnership, has given our nation new and better ways to improve food production, processing, and marketing and has helped solve problems in environmental quality and human nutrition. USDA, a major contributor to the nation's public-sector agricultural research, distributes its funds in three basic ways. First, funds are allocated to USDA's ARS for in-house research. ARS, in turn, allocates these funds to its 140 research facilities on the basis of research programs and without regard to geographic dispersion. ARS research focuses on agricultural problems of regional, national, and international concern. ARS' budget for fiscal year 1985 was set at \$488 million.

Second, funds are allocated to USDA's CSRS for further distribution to states on the basis of a formula incorporating each state's farm and rural population. This research is accomplished largely in state agricultural experiment stations and colleges of veterinary medicine, which are a part of what are known as land-grant universities and whose research is directed at problems ranging from those of a local and regional nature to those of a national and international nature. CSRS' budget for fiscal year 1985 was set at \$292 million.

Third, funds are allocated to USDA's OGPS for distribution as competitive grants to a wide range of institutions. OGPS received \$17 million for such distribution during fiscal year 1984.

PRINCIPAL FINDINGS

Biotechnology research represents but a small part of the total agricultural research funded by USDA. Our efforts, for example, to inventory the biotechnology research conducted or sponsored by USDA disclosed a total of 778 projects being worked on during the 1984-1985 time frame. As shown in table I.1, USDA expended \$40.5 million for these projects, or about 6.3 percent of its total funding for agricultural research during the time frame.

Table I.1

USDA-Conducted or -Sponsored Biotechnology Research

<u>USDA agency</u>	<u>Time frame (fiscal year)^a</u>	<u>Number of biotechnology research projects</u>	<u>USDA dollars spent on these projects during related time frame (millions)</u>	<u>Total USDA dollars spent on agricultural research during related time frame (millions)</u>	<u>Percent biotechnology research represents of total USDA research</u>
ARS	1985	183	\$26.4	\$488.0	5.4
CSRS	1984	495	10.7	185.3	5.8
OGPS	1984	<u>145</u>	<u>4.8^b</u>	<u>17.0^b</u>	28.2
Totals		<u>778^b</u>	<u>\$40.5^b</u>	<u>\$688.9^b</u>	5.9

^aInformation we received from CSRS (through the questionnaire sent to state agricultural experiment stations and colleges of veterinary medicine) and OGPS related to fiscal year 1984. Information received from ARS related to fiscal year 1985.

^bFigures in each column do not add to the totals. This is because 45 of the 145 OGPS-funded projects (representing a cost of \$1.4 million in fiscal year 1984) were also reported to us by CSRS (through the questionnaire used at state agricultural experiment stations) or by ARS. To avoid double counting, we deducted these projects and their annual cost from the totals.

Source: Prepared by GAO from information supplied by ARS, OGPS, state agricultural experiment stations, and colleges of veterinary medicine.

Appendix VI reflects the state-by-state distribution of biotechnology research projects funded during fiscal years 1984 or 1985 in whole or in part by USDA and also the USDA research dollars that relate to those projects. The appendix breaks the state-by-state totals of projects and funding down further according to USDA funding source. The five states with the largest number of projects were California, Maryland, New York, Florida, and Texas. In addition, the five states receiving the greatest number of USDA biotechnology research dollars were Maryland, New York, California, Florida, and Illinois.

Biotechnology research funded by CSRS

Responses to our questionnaire, which was sent to all state agricultural experiment stations and colleges of veterinary

medicine, disclosed 495 biotechnology research projects that received \$10.7 million in USDA funding during fiscal year 1984. The average life of these projects was estimated to be 82 months, with an average of 50 months already expended and 32 months remaining. The overall funding relating to these projects as well as the total funding relating to all agricultural research at each of the 55 state agricultural experiment stations and the 5 colleges of veterinary medicine that responded to our questionnaire is shown in table I.2.

Table I.2

Biotechnology Research Funding^a Compared To Total Agricultural
Research Funding At Experiment Stations and
Veterinary Colleges

<u>Source of funding</u>	<u>Biotechnology research</u> ----- (millions)	<u>Total agricultural research</u> -----	<u>Percent of biotechnology research to total research</u>
USDA competitive grants	\$ 2.8	\$ 11.6	23.9
All other USDA funds	7.9	173.6	4.6
Other federal agencies	13.6	109.1	12.5
State agencies	17.3	551.2	3.1
Industry	<u>5.6</u>	<u>86.3</u>	6.5
Total	<u>\$47.2^b</u>	<u>\$931.8</u>	5.1

^aRelates to the 495 research projects discussed on the previous two pages.

^bThis figure does not include an estimated \$500,000 reported to us by the North Dakota Agricultural Experiment Station or \$1,693 reported by the Ohio experiment station. The two stations, although providing us with a total figure for their biotechnology research, did not identify the specific sources of that funding and we, therefore, excluded the amounts from the table.

Table I.2 shows that USDA provides only a portion of the funding used for agricultural research (biotechnology or otherwise) at state agricultural experiment stations and colleges of veterinary medicine. From the table, for example, it can be calculated that the \$10.7 million spent by USDA on biotechnology research was about 23 percent of the total \$47.2 million spent on such research at these institutions and that the \$185.3 million USDA spent at these institutions on agricultural research as a

Table I.3

Biotechnology Research Techniques Used on the
495 Projects Reported

<u>Biotechnology research techniques</u>	<u>Number of instances in which technique was used</u>	<u>Percent of total projects (495) using techniques</u>
Direct manipulation of genome		
Recombinant DNA	267	54
Chemical synthesis of nucleic acids	95	19
Site-directed mutagenesis	91	18
Direct manipulation of cells		
Microinjection	27	6
Transfection	79	16
Transformation	148	30
Embryo manipulation and transfer	66	13
Cell culture and protoplast fusion	233	47
Other (as specified by respondents)	93	19

We also asked in our questionnaire for the scientists to identify biotechnology research projects that they expected in the future to involve the deliberate release of genetically engineered organisms into the environment. In contrast to previous attention paid to the harm that could result from the accidental escape of some new, genetically engineered organism, concern has been expressed over the possible effects from the deliberate release of one. Although a USDA official told us that there have been no deliberate releases of genetically engineered organisms to date, it appears that such a time may not be far off. The responses to our questionnaire, for example, disclosed 87 research projects that were expected to involve such releases as a part of the experimentation; 11 of these projects were expected to involve a release within 1 year from the time the questionnaire was filled out (early 1985), from 2 to 5 years was specified for 47 projects, and after 5 years was specified for the remaining 29 projects.

The 87 projects that were expected to lead to the release of genetically altered organisms into the environment cover a broad spectrum of agricultural and food-related concerns. The organisms being altered include crops such as beans, rice, corn, wheat, grapes, potatoes, and lettuce; specific types of viruses, bacteria, and fungi; and forest, fruit, and ornamental trees as well as florist-related crops. One project involved the development of a much larger variety of salmon. The objectives of

whole was about 20 percent of the total \$931.8 million spent. Appendix VII reflects the state-by-state distribution of the 495 biotechnology research projects as well as the \$10.7 million spent by USDA and the total \$47.2 spent by all sources on biotechnology research at state agricultural experiment stations and colleges of veterinary medicine.

As another means of determining the relative emphasis being placed on biotechnology research as compared to total agricultural research, our questionnaire asked the various institutions for the number of scientists each had working on a full-time equivalent basis in terms of total agricultural research and biotechnology research. The 49 respondents to this question reported a total of 6,666 scientists (full-time equivalents) at their institutions with 358, or about 5.4 percent, of them involved in biotechnology research. All of the respondents expected their efforts in biotechnology research--in terms of not only scientists but graduate students and technical support as well--to either increase, or at least stay the same, during the next 2 years. None foresaw decreases.

We asked in our questionnaire which biotechnology research techniques were used with respect to each of the 495 projects reported to us. Table I.3 shows that the techniques known as recombinant DNA and cell culture/protoplast fusion were used most frequently in 54 and 47 percent of the cases, respectively. Multiple techniques were being used in many cases.

Table I.4Scientists' Responses to Problematic Releases

<u>Will such releases into the environment cause problems or represent reason for concern?</u>		<u>What effort would it take to correct any problems that might arise?</u>	
No problem	75	Self-controlling	68
Very minor problem	9	Little effort	13
Minor problem	0	Some effort	2
Moderate problem	0	Moderate effort	3
Major problem	0	Great effort	0
Very major problem	0	Very great effort	0
Do not know	<u>3</u>	Uncontrollable	<u>1^a</u>
	<u>87</u>		<u>87</u>

^aIn this case, the research involves an attempt to improve--and facilitate commercial production of--viral pesticides through genetic engineering. The principal scientist noted in the questionnaire that she anticipated a release within 2 to 5 years. She did not know whether such a release would result in any problems. Although she thought that no problems would occur, she also recognized that a variety of "constructs" are possible and that some could have broader effects than desired. The scientist stated that risk assessment was a part of the research project and that "In the course of our studies already we have developed what we consider improved methods of assessing risks of genetically engineering viral pesticide products."

Later, by telephone, the scientist told us that biotechnology research involving microbial (e.g., viral, bacterial, or fungal) pesticides must be thoroughly tested before approval will be given to release any genetically engineered organisms into the environment. She mentioned that there are specific Environmental Protection Agency (EPA) guidelines that must be followed and that, before any such release, EPA approval must be obtained. She also said that intelligent scientists can design experiments that present no danger, and that it is extremely unlikely that anyone would want to endanger human health or the environment as a result of their experimentation.

Our questionnaire asked each experiment station and veterinary college to list its biotechnology agricultural research accomplishments since October 1, 1982. Appendix X provides such a listing. The accomplishments have covered a wide range of activities. For example, different biotechnology techniques such as cell culture, embryo transfer, and recombinant DNA have

the projects were also widely varied. Some of the objectives included the weakening of disease-causing organisms or the strengthening of resistance in plants and animals to disease and other stresses; the stimulation of growth or productivity; the improvement or preservation of quality in specific foods; and the development of more effective biocontrol agents (generally, microorganisms or insects that prey on harmful organisms). For a profile of each of the 87 projects, see appendix VIII.

The states with the greatest number of these 87 projects included North Carolina (13), California (10), Texas (8), Florida (6), and Minnesota (6). The remaining 44 projects were spread among 23 additional states (see app. IX).

We asked the scientists working on the 87 projects if they believed the releases would cause any problems and, if so, what level of effort might be needed to correct them. The following responses generally reflect a great deal of optimism and confidence on the part of the scientists.

Table I.5ARS' Biotechnology Research

<u>By geographic area</u>	<u>Planned expenditures</u> (millions)
Northeastern region	\$12.3
North central region	5.0
Western	3.8
Southern	<u>5.3</u>
Total	<u>\$26.4</u>
<u>By strategic plan code</u>	
Soil and water	\$ 0.20
Plants	10.00
Animals	8.80
Conversion (postharvest/utilization)	7.40
Human nutrition	<u>0.05</u>
Total	<u>\$26.45</u>
<u>By biotechnology research area</u>	
Genes	\$13.8
Membranes	3.5
Mediators (things that bring about a response)	6.4
Bioconversion (postharvest biology and processing)	<u>2.6</u>
Total	<u>\$26.3</u>

The compendium did not give an indication of the types of biotechnology techniques that were being used on the 183 projects, nor did it tell which of the projects were expected to someday involve a deliberate release of genetically engineered organisms into the environment. A knowledgeable ARS official who helped develop the compendium said that he did not know which of the projects might someday involve a deliberate release--that this would be very difficult to determine given the constantly changing nature of the experimentation. In commenting on this report, ARS' Assistant Administrator for Cooperative Interaction pointed out that research conducted by ARS generally involves concepts, not products, and that it is therefore unlikely that many of ARS' biotechnology research projects would be expected to someday

involved research at the cellular and molecular levels and have produced altered organisms that have survived and demonstrated desirable traits. Plants with greater resistance to herbicides, low temperatures, and salt were being developed. Chromosome maps of specific gene sequences in plants and viruses have been established. New vaccines and methods of diagnosing disease have been proved successful and a vector (e.g., a virus used for introducing DNA into an organism) has been developed for crop plants.

Biotechnology research conducted by ARS

ARS' February 1985 compendium of biotechnology research listed a total of 183 biotechnology research projects being conducted by ARS at an expected cost in fiscal year 1985 of \$26.4 million. These figures represent approximately 8.8 percent of the 2,075 total projects ARS was conducting and about 5.4 percent of the \$488 million ARS was planning to spend on agricultural research during the fiscal year.

The compendium's executive summary classified the biotechnology research as follows:

GLOSSARY OF TECHNICAL TERMS

Cell culture and protoplast fusion: cell culture involves the in vitro growth of cells isolated from multicellular organisms. These cells are usually of one type. Protoplast fusion is a technique used in joining two cells in vitro. A protoplast is a cell from which the outer cell wall has been removed.

Chemical synthesis of nucleic acids: an in vitro technique used to produce nucleic acids (the chemical basis of DNA) by combining simple molecules without having to work with whole organisms.

Deoxyribonucleic acid (DNA): the genetic material found in all living organisms.

Embryo manipulation and transfer: a technique used to work with embryos in utero (in the mother) or in vitro and to transfer embryos from mother to mother or from in vitro to mother.

Genome: the basic chromosome set of an organism or the sum total of its genes. The total DNA complement of a cell, carrying the blueprint for the cell's organization and function.

In vitro: outside the living body in an artificial environment.

Microinjection: a technique by which nucleic acids are injected directly into a cell.

Recombinant deoxyribonucleic acid (DNA): Recombinant DNA techniques involve joining together pieces of DNA from different organisms or synthetic DNA in vitro, thus producing hybrid or chimaeric DNA.

Site-directed mutagenesis: the induction of mutation at a specific point or points in the genetic material of an organism; researchers may use physical or chemical means to cause mutations.

Transfection: a technique for changing a cell's genetic information by using a vector (carrier) to introduce desired foreign DNA into host cells. Examples of vectors are plasmids, transposable elements, or viruses.

Transformation: the acquisition of new genetic information by incorporation of DNA.

Vector: an organism, such as a virus, used for introducing DNA into another organism.

involve the deliberate release of genetically engineered organisms into the environment. In the event an experiment was to involve such a release, however, he said that approval to do so would not be given without careful scrutiny.

Biotechnology research funded by OGPS

OGPS provided information on 145 biotechnology research projects that received \$4.8 million in competitive grants funding during fiscal year 1984. We determined with the help of an OGPS official that 45 of these projects valued at \$1.4 million were duplicative of projects reported to us by either ARS or a state agricultural experiment station. With respect to the remaining 100 projects, the OGPS information showed that recombinant DNA was being used in 66 instances, transformation in 8 instances, cell culture and protoplast fusion in 8 instances, and site-directed mutagenesis in 5 instances. OGPS listed techniques other than those we specified in 14 instances.

We asked the OGPS official who assembled the information how many of the projects would involve a deliberate release of genetically engineered organisms into the environment. Without checking with the scientists responsible for each of the projects, the official advised us of four projects that she expected to result in a release--one within a year, two within 2 to 5 years, and one after 5 years. The official expected no problems to develop in three of the four releases; in one case she did not know whether problems would result.

A copy of our questionnaire, containing the information we received from OGPS, is included as appendix XI.

VIEWS OF AGENCY OFFICIALS

Officials from ARS, CSRS, and OGPS were given the opportunity to comment on a draft of this report; and a number of changes were made to clarify information in the report on the basis of the comments received. An OGPS associate program manager in the Competitive Research Grants Office additionally told us of the rather significant increase in OGPS' biotechnology research effort during fiscal year 1985 that she believed should be acknowledged in this report. She said that \$20 million had been made available during the year specifically for biotechnology research (up from \$4.8 million in fiscal year 1984) and that OGPS was also administering a new forestry biotechnology program that had a budget for the year of close to \$8 million. She said that the majority of these funds had been distributed in the form of grants awarded during July through September 1985 (a time period so recent that we were precluded from obtaining and including more detailed information on these grants in this report).

Biotechnology risk assessment research—involves (1) assessing risks associated with the research and (2) developing new or improving existing methods for determining the possibility:

- 1-of the survival and growth of genetically engineered organisms beyond intended environments;
- 2-that genetically engineered organisms may be harmful to humans, the environment, or to other organisms or species they may come in contact with; or
- 3-that genetically engineered organisms may exchange genetic information with other organisms, resulting in possible harmful effects such as those alluded to in (2) above.

Non-biotechnology risk assessment research—

Although risk assessment in biotechnology may sound like a new area of research, risk assessment in the non-biotechnology area has been done for some time. The following are several on-going research programs which are examples of risk assessment that are being conducted in association with conventional breeding programs. These types of research should be reported in this questionnaire.

- Remote sensing to detect and evaluate potential problems
- Field evaluations of germplasm for performance under varied biological and physical stresses
- Evaluation of food products for potential toxic effects
- Systems science and modeling

The questions which follow should be considered in light of the above definitions. The first section of the questionnaire relates to general information about research at your state agricultural experiment station. The second section relates to specific biotechnology and risk assessment research projects. For your convenience enclosed are 20 copies of the second section which can be used to provide information on specific research projects. Please duplicate these pages if you need additional copies.

Please return each completed questionnaire in the enclosed pre-addressed postage-paid envelope by February 15, 1985. In the event the envelope is misplaced, the return address is:

Mr. Ralph W. Lamoreaux
U.S. General Accounting Office
441 G Street, N.W., Room 4476
Washington, DC 20548

If you have any questions about the survey, please call either Mr. Ralph W. Lamoreaux on (202) 275-5405 or Dr. Charles E. Hess of the NASULGC's Committee on Biotechnology on (916) 752-1605. We appreciate your participation and cooperation.



NASULGC

JOINT GAO AND NASULGC QUESTIONNAIRE

JOINT SURVEY SPONSORED BY THE
U.S. GENERAL ACCOUNTING OFFICE AND THE
NATIONAL ASSOCIATION OF STATE UNIVERSITIES AND LAND GRANT COLLEGES
(DIVISION OF AGRICULTURE-COMMITTEE ON BIOTECHNOLOGY)



Biotechnology and Risk Assessment Research

INTRODUCTION

At the request of the Chairman, U.S. House of Representative's Committee on Science and Technology, the U.S. General Accounting Office (GAO) is examining the U.S. Department of Agriculture's (USDA's) role in the biotechnology/genetic engineering area. One part of this examination involves documenting USDA's research and development activities in biotechnology/genetic engineering. USDA's Agricultural Research Service has been requested to provide GAO with information relating to the research USDA is doing "in-house." USDA's Office of Grants and Program Systems has likewise been asked to provide GAO with information relating to the research being done as a result of the competitive grants program. This questionnaire will provide GAO with information relating to the research being done at state agricultural experiment stations which receive funding from USDA's Cooperative State Research Service.

The National Association of State Universities and Land Grant Colleges (NASULGC) is cooperating with the GAO in conducting this survey. Questions have been incorporated which will permit the NASULGC's Division of Agriculture-Committee on Biotechnology to update its 1982 survey of involvement of state agricultural experiment stations in biotechnology research. The results of this survey will be presented at the land grant meeting scheduled for November 1986.

This inquiry is interested in the overall research effort at this state agricultural experiment station, but more particularly in the biotechnology and risk assessment (biotechnology and non-biotechnology) research efforts which this institution has underway. To aid those filling out the questionnaire, definitions of these two terms are provided as follows.

Biotechnology research--the process of in vitro alteration of genetic material for the purpose of creating new gene combinations or modifications.

In this regard, this inquiry is limited to research involving the following biotechnology research techniques:

- 1-Direct manipulation of the genome using recombinant DNA, chemical synthesis of nucleic acids, and/or site-directed mutagenesis (these techniques are often known as genetic engineering).
- 2-Direct manipulation of cells (altering genetic information) using microinjection, transfection, transformation, embryo transfer, and/or cell and protoplast culture and fusion (i.e., using other biotechnology research techniques).

5. Since plant and animal breeding research helps translate biotechnology advances in agricultural research into practice, how many scientist FTEs are involved in such breeding programs at this institution? FTEs should be reported to the nearest tenth. (FOR THE TWO TYPES OF BREEDING PROGRAMS WRITE IN THE NUMBER OF SCIENTIST FTEs.)

Number of plant breeding scientist FTEs 425 (N=48)

Number of animal breeding scientist FTEs 144 (N=48)

6. For each of the following funding sources, please answer the following two questions as they relate to this agricultural experiment station.
- a. In Column A, for each funding source, indicate how much money was spent (to the nearest dollar) between October 1, 1983 and September 30, 1984 on all research. (INCLUDE BOTH NON-BIOTECHNOLOGY AND BIOTECHNOLOGY AGRICULTURAL RESEARCH. IF NONE ENTER 0.)
- b. In Column B, for each funding source, indicate how much money was spent (to the nearest dollar) between October 1, 1983 and September 30, 1984 on all biotechnology agricultural research. For research projects which combine both biotechnology and conventional procedures, report only the funds devoted to the biotechnology part of the project. (IF NONE ENTER 0.)

<u>FUNDING SOURCES(S)</u>	<u>COLUMN A</u>	<u>COLUMN B</u>
	TOTAL FUNDS SPENT ON ALL RESEARCH	TOTAL FUNDS SPENT ON BIOTECHNOLOGY AGRICULTURAL RESEARCH
1. USDA competitive grants	\$ 11,676,549 (N=45)	\$ 2,803,651 (N=46)
2. USDA (all other)	\$ 173,585,939 (N=48)	\$ 7,906,627 (N=48)
3. Other federal agencies	\$ 109,083,115 (N=46)	\$ 13,634,943 (N=47)
4. State agencies	\$ 551,193,485 (N=48)	\$ 17,235,007 (N=48)
5. Industry	\$ 86,288,800 (N=46)	\$ 5,615,759 (N=47)
TOTALS	\$ 923,571,065 (N=47) ^a	\$ 47,697,680 (N=50) ^b

^aThis is the figure reported to us by the questionnaires. It does not include \$8,882,010 total reported by Maryland. It does include \$625,177 overstatement of Colorado's total. Correct figure, therefore, is \$931,827,898.

^bThis is the figure reported to us by the questionnaires. It includes \$500,000 reported to us by North Dakota, and \$1,693 reported to us by Ohio, both of which were not broken down (or reported to us) by funding source.

SECTION IGeneral Questions Relating To Overall Research
At This State Agricultural Experiment Station

1. What is the name of this state agricultural experiment station?
PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 10 WORDS OR LESS.
State Agricultural Experiment Stations (N=50) and Colleges of
Veterinary Medicine (N=5) [See GAO Note]
2. During fiscal year (FY) 1984 [10/1/83-9/30/84], how many scientist full time equivalents (FTEs) in the agricultural experiment station were at this institution? Please include all research activities, do not limit your answer to biotechnology and risk assessment research. FTEs SHOULD BE REPORTED TO THE NEAREST TENTH.

Number of scientist FTEs 6666 (N=49)

3. During FY 1984, how many scientist FTEs at this institution are involved in biotechnology agricultural research? Consider only the time each scientist worked on biotechnology research, and then report the total scientist FTEs to the nearest tenth. Enter estimates from .05 through .14 as .1; enter .15 through .24 as .2, etc. PLEASE SEE INTRODUCTION FOR DEFINITIONS.
- Number of scientist FTEs in
biotechnology agricultural research 358 (N=48)
4. During the next two years, does this institution expect to increase or decrease (through reallocation or attrition) its scientist FTEs in biotechnology agricultural research? FTEs should be reported to the nearest tenth. (UNDER INCREASES IN FTEs CHECK ALL THAT APPLY AND SPECIFY THE INCREASE. UNDER DECREASES OF FTEs CHECK ALL THAT APPLY AND SPECIFY THE DECREASE.)

A. We will neither increase nor decrease any FTEs (N=2)

B. INCREASES IN FTE

1. Yes, we will increase faculty FTEs by 162 (N=47) FTEs
2. Yes, we will increase graduate student FTEs by 277 (N=47) FTEs
3. Yes, we will increase technical support staff FTEs by 191 (N=45) FTEs

C. DECREASES IN FTE

1. Yes, we will decrease faculty FTEs by 0 FTEs
2. Yes, we will decrease graduate student FTEs by 0 FTEs
3. Yes, we will decrease technical support staff FTEs by 0 FTEs

[GAO Note: N = the number of responses to a specific question. A total of 50 State Agricultural Experiment Stations and 5 Colleges of Veterinary Medicine reported USDA-funded biotechnology research directly to us through this questionnaire. The stations and colleges did not always respond to all questions. The value of N, therefore, varies with respect to many questions.]

SECTION I

(To be filled out by Director, State Agricultural Experiment Station)

SECTION II

QUESTIONS RELATING TO SPECIFIC BIOTECHNOLOGY RESEARCH PROJECTS

For each biotechnology research project funded in whole or in part by USDA at this agricultural experiment station, please answer the following 18 questions. IF NECESSARY, PLEASE REPRODUCE THESE QUESTIONS SO THAT YOU CAN PROVIDE ANSWERS FOR EACH OF YOUR ONGOING RESEARCH PROJECTS.

PART A. SPECIFIC PROJECT INFORMATION

- 1. What is the project's title? PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

- 2. Does the project have a CRIS identification number?

- 1. Yes...WHAT IS THE CRIS IDENTIFICATION NUMBER? _____

- 2. No....IS THERE ANY OTHER IDENTIFICATION NUMBER?
IF SO, PLEASE PROVIDE. _____

- 3. Are there keywords reported for this project in the CRIS?

- 1. No...Keywords are not reported/Project is not in the CRIS

- 2. Yes...(SPECIFY UP TO 10 KEYWORDS AND PLEASE PRINT CLEARLY.
EACH KEYWORD SHOULD BE NO LONGER THAN 50 CHARACTERS.)

- a. _____
- b. _____
- c. _____
- d. _____
- e. _____
- f. _____
- g. _____
- h. _____
- i. _____
- j. _____

7. For each of the following funding sources, please answer the following two questions as they relate to risk assessment research conducted at this agricultural experiment station. See the introduction for a description and examples of risk assessment research.
- a. In Column A, for each funding source, indicate how much money was spent (to the nearest dollar) between October 1, 1983 and September 30, 1984 on biotechnology research devoted primarily to risk assessment. (IF NONE ENTER 0.)
- b. In Column B, for each funding source, indicate how much money was spent (to the nearest dollar) between October 1, 1983 and September 30, 1984 on non-biotechnology research devoted primarily to risk assessment. (IF NONE ENTER 0.)

<u>FUNDING SOURCES(S)</u>	<u>COLUMN A</u>	<u>COLUMN B</u>
	TOTAL FUNDS SPENT ON BIOTECHNOLOGY RISK ASSESSMENT RESEARCH	TOTAL FUNDS SPENT ON NON-BIOTECHNOLOGY RISK ASSESSMENT RESEARCH
1. USDA competitive grants	\$ 53,156 (N=39)	\$ 790,712 (N=42)
2. USDA (all other)	\$ 645,651 (N=41)	\$ 7,244,415 (N=42)
3. Other federal agencies	\$ 128,512 (N=38)	\$ 2,674,456 (N=41)
4. State agencies	\$ 1,610,921 (N=41)	\$ 20,540,575 (N=42)
5. Industry	\$ 450,062 (N=41)	\$ 3,977,843 (N=41)
TOTALS	\$ 2,907,602 (N=41)	\$ 34,618,001 (N=42)

8. Since October 1, 1982, what biotechnology agricultural research accomplishments (if any) have occurred at this institution? Please describe. PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 100 WORDS OR LESS.

7. For how many months has this project been funded? (WRITE IN NUMBER OF MONTHS.)

Number of months Average is 50 months. (N=480)

8. How many months longer is this project expected to run? (WRITE IN NUMBER OF MONTHS.)

Number of months Average is 32 months. (N=466)

9. Is it expected that this project will involve the release of genetically engineered organisms into the environment? (CHECK ONE.)

1. Yes...CONTINUE TO QUESTION 10 N=87

2. No...SKIP TO QUESTION 14 N=406

10. When will this project involve the release of genetically engineered organisms into the environment? (CHECK ONE.)

1. Within 1 year N=11

2. In 2 to 5 years N=47

3. After 5 years N=29

11. Will the National Institutes of Health's Recombinant DNA Advisory Committee's approval for the deliberate release into the environment of a genetically engineered organism be sought? (CHECK ONE.)

1. Yes, it is applicable and will be sought N=52

2. No, it is not applicable and will not be sought N=33

3. No, it is applicable and will not be sought...PLEASE EXPLAIN WHY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

4. If we need additional information, who should be contacted regarding this particular project?

Scientist's Name _____

Title _____

Telephone Number () _____
 Area Code

State (PLEASE PROVIDE TWO-LETTER POSTAL ABBREVIATION) _____

5. Briefly, what are the project's major objectives? PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

6. Which of the following genetic engineering techniques are being used in this project? (CHECK ALL THAT APPLY.) GAO Note: Some projects involve more than one technique.

1. Recombinant-DNA N=267

2. Chemical synthesis of nucleic acids N=95

3. Site-directed mutagenesis N=91

4. Microinjection N=27

5. Transfection N=79

6. Transformation N=148

7. Embryo manipulation and transfer N=66

8. Cell culture and protoplast fusion N=233

9. Other (SPECIFY PLEASE LIMIT YOUR RESPONSE TO 50 WORDS OR LESS) N=96

14. Is risk assessment, as defined in the introduction, a part of this research project?

1. Yes...CONTINUE TO QUESTION 15 N=62

2. No....SKIP TO QUESTION 16 N=422

15. Is the risk assessment part of this research project expected to result in (CHECK ALL THAT APPLY AND EXPLAIN AS APPROPRIATE).....

1. an assessment of the risks associated with this experimentation? N=45

2. new risk assessment methods or techniques? (PLEASE EXPLAIN AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.) N=9

Examples included new methods or techniques such as (1) simulation of crop growth effects resulting from genetic engineering, (2) new methods of risk assessment for genetically engineered viral pesticides, and (3) a quick test to evaluate exposure and residue clearance involving a certain toxin in animals.

3. improvement of existing risk assessment methods or techniques? (PLEASE EXPLAIN AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.) N=15

Examples included (1) a faster method to assay nutrient composition and pathogen resistance, (2) special genetic markers to detect foreign genome introduction, and (3) use of trout for cancer tests to reduce dependency on expensive rodents.

12. In your opinion, if the genetically engineered organisms produced by this project were released into the environment, how much of a problem would or could such action be to the environment? (CHECK ONE.)

1. No problem N=76
2. Very minor problem N=9
3. Minor problem N=0
4. Moderate problem N=0
5. Major problem N=0
6. Very major problem N=0
7. Don't know N=3

13. In your opinion, how much effort would it take to correct any such problems which might result from releasing into the environment genetically engineered organisms produced by this project? (CHECK ONE.)

1. Situation could not be corrected (situation would be uncontrollable) N=1
2. Very great effort N=0
3. Great effort N=0
4. Moderate effort N=3
5. Some effort N=2
6. Little effort N=13
7. No effort (situation would be self-controlling) N=69

PART B. STAFFING AND FUNDING INFORMATION

16. How many paid researchers worked on this project during FY 1984? If this research project combined both biotechnology and conventional procedures, then report only the FTEs devoted to the biotechnology part of the project. FTEs should be reported to the nearest tenth.

a. Number of faculty FTEs 279 (N=472)

b. Number of graduate students FTEs 418 (N=437)

c. Number of technical support staff FTEs 308.8 (N=439)

17. To the best of your knowledge, how many FTEs are expected to be expended on this project (biotechnology only) over its entire life? (INCLUDE FACULTY, GRADUATE STUDENTS, AND TECHNICAL SUPPORT STAFF.)

Number of FTEs 4,051 (N=442)

18. For each of the following funding sources, please answer the following three questions as they relate to the specific biotechnology research project covered by this questionnaire. If this research project combined both biotechnology and conventional procedures, then report only the funds devoted to the biotechnology part of the project. Funds should be reported to the nearest dollar.

a. In Column A, for each funding source, indicate how much money was spent before October 1, 1983 on this biotechnology research project.

b. In Column B, for each funding source, indicate how much money was spent between October 1, 1983 and September 30, 1984 on this biotechnology research project.

c. In Column C (to the best of your knowledge), for each funding source, indicate how much additional money is expected/needed to be spent on this biotechnology research project.

<u>FUNDING SOURCE(S)</u>	<u>COLUMN A</u> TOTAL FUNDS SPENT TO 9/30/83 ON THIS SPECIFIC BIOTECHNOLOGY RESEARCH PROJECT	<u>COLUMN B</u> FUNDS SPENT 10/1/83-9/30/84 ON THIS SPECIFIC BIOTECHNOLOGY RESEARCH PROJECT	<u>COLUMN C</u> TOTAL ADDITIONAL FUNDS EXPECTED/ NEEDED OVER THIS PROJECT'S LIFE
1. USDA competitive grants	\$ 3,402,711(N=255)	\$ 2,722,622(N=277)	\$ 22,291,518(N=290)
2. USDA (all other)	\$ 7,058,461(N=329)	\$ 5,329,372(N=390)	\$ 23,886,482(N=369)
3. Other federal agencies	\$ 9,803,204(N=301)	\$ 8,116,391(N=354)	\$ 28,002,186(N=332)
4. State agencies	\$14,511,612(N=331)	\$11,790,723(N=409)	\$ 37,872,390(N=384)
5. Industry	\$ 5,621,530(N=295)	\$ 4,260,580(N=338)	\$ 14,836,961(N=310)
TOTALS	\$38,038,682(N=350)	\$30,656,378(N=438)	\$123,188,558(N=421)

JOINT DEVELOPMENT OF QUESTIONNAIRE WITH NASULGC

To obtain information on the biotechnology research funded in whole or in part by CSRS, we--in conjunction with NASULGC-- developed a questionnaire (see app. III). NASULGC is an association of 69 land grant colleges and 78 state universities, formed in 1963. Its Division of Agriculture's Committee on Biotechnology was appointed in April 1982 and charged with advising the Division on biotechnology matters. In November 1983 the Committee reported on its assessment of the investment in biotechnology by state agricultural experiment stations and the ARS.¹ At the time we began our work, we learned of the Committee's desire to update its earlier findings. To avoid duplication, we therefore approached and reached agreement with NASULGC to jointly proceed with a questionnaire to be sent to all state agricultural experiment stations and colleges of veterinary medicine, which would provide the information needed by both the NASULGC and us. Questions were incorporated that would permit the NASULGC Division of Agriculture's Committee on Biotechnology to update its earlier findings as well as allow us to be responsive to the needs of the House Committee on Science and Technology.

¹The report was entitled Emerging Biotechnologies In Agriculture: Issues and Policies, Progress Report II, November 1983.

STATE AGRICULTURAL EXPERIMENT STATIONS AND
COLLEGES OF VETERINARY MEDICINE

Alabama

Agricultural Experiment Station, Auburn University
School of Veterinary Medicine, Auburn University
School of Veterinary Medicine, Tuskegee Institute

Alaska

Agricultural Experiment Station, University of Alaska

Arizona

Agricultural Experiment Station, University of Arizona

Arkansas

Agricultural Experiment Station, University of Arkansas

California

Agricultural Experiment Station, University of
California-Berkeley
Agricultural Experiment Station, University of California-
Riverside
Agricultural Experiment Station, University of California-
Davis
School of Veterinary Medicine, University of California-Davis

Colorado

Agricultural Experiment Station, Colorado State University
College of Veterinary Medicine and Biomedical Sciences,
Colorado State University

Connecticut

Agricultural Equipment Station, University of Connecticut

Delaware

Agricultural Experiment Station, University of Delaware

Washington, DC

Agricultural Experiment Station, University of the District
of Columbia

Florida

Agricultural Experiment Station, University of Florida
College of Veterinary Medicine, University of Florida

Georgia

Agricultural Experiment Station, University of Georgia
College of Veterinary Medicine, University of Georgia

Hawaii

Agricultural Experiment Station, University of Hawaii at
Manoa

Idaho

Agricultural Experiment Station, University of Idaho
W.O.I. Regional Program in Veterinary Medicine, University of
Idaho

Illinois

Agricultural Experiment Station, University of Illinois
College of Veterinary Medicine, University of Illinois

Indiana

Agricultural Experiment Station, Purdue University
School of Veterinary Medicine, Purdue University

Iowa

Agricultural and Home Economics Experiment Station, Iowa
State University
College of Veterinary Medicine, Iowa State University

Kansas

Agricultural Experiment Station, Kansas State University
College of Veterinary Medicine, Kansas State University

Kentucky

Agricultural Experiment Station, University of Kentucky

Louisiana

Agricultural Experiment Station, Louisiana State University
and A&M College
College of Veterinary Medicine, Louisiana State University

Maine

Agricultural Experiment Station, University of Maine

Maryland

Agricultural Experiment Station, University of Maryland

Massachusetts

Agricultural Experiment Station, University of Massachusetts
School of Veterinary Medicine, Tufts University

Michigan

Agricultural Experiment Station, Michigan State University
College of Veterinary Medicine, Michigan State University

Minnesota

Agricultural Experiment Station, University of Minnesota
College of Veterinary Medicine, University of Minnesota

Mississippi

Agricultural and Forestry Experiment Station, Mississippi
State University
College of Veterinary Medicine, Mississippi State University

Missouri

Agricultural Experiment Station, University of Missouri
College of Veterinary Medicine, University of Missouri

Montana

Agricultural Experiment Station, Montana State University

Nebraska

Agricultural Experiment Station, University of Nebraska

Nevada

Agricultural Experiment Station, University of Nevada

New Hampshire

Agricultural Experiment Station, University of New Hampshire

New Jersey

Agricultural Experiment Station, Rutgers University

New Mexico

Agricultural Experiment Station, New Mexico State University

New York

Agricultural Experiment Station, Cornell University
New York Veterinary College, Cornell University

North Carolina

North Carolina Agricultural Research Service,
North Carolina State University
School of Veterinary Medicine, North Carolina State
University

North Dakota

Agricultural Experiment Station, North Dakota State
University

Ohio

Ohio Agricultural Research and Development Center, Ohio State
University
College of Veterinary Medicine, Ohio State University

Oklahoma

Agricultural Experiment Station, Oklahoma State University
College of Veterinary Medicine, Oklahoma State University

Oregon

Agricultural Experiment Station, Oregon State University
School of Veterinary Medicine, Oregon State University

Pennsylvania

Agricultural Experiment Station, Pennsylvania State
University
School of Veterinary Medicine, University of Pennsylvania

Rhode Island

Agricultural Experiment Station, University of Rhode Island

South Carolina

Agricultural Experiment Station, Clemson University

South Dakota

Agricultural Experiment Station, South Dakota State University

Tennessee

Agricultural Experiment Station, University of Tennessee
College of Veterinary Medicine, University of Tennessee

Texas

Agricultural Experiment Station, Texas A&M University
College of Veterinary Medicine, Texas A&M University

Utah

Agricultural Experiment Station, Utah State University

Vermont

Agricultural Experiment Station, University of Vermont

Virginia

Agricultural Experiment Station, Virginia Polytechnic
Institute and State University
Virginia-Maryland Regional College of Veterinary Medicine,
Virginia Polytechnic Institute and State University

Washington

Agricultural Research Center, Washington State University
College of Veterinary Medicine, Washington State University

West Virginia

The West Virginia Agricultural and Forestry Experiment
Station, West Virginia University

Wisconsin

Agricultural Experiment Station, University of Wisconsin
College of Veterinary Medicine, University of Wisconsin

Wyoming

Agricultural Experiment Station, University of Wyoming

Guam

Agricultural Experiment Station, University of Guam

Puerto Rico

Agricultural Experiment Station, University of Puerto Rico

Virgin Islands

Agricultural Experiment Station, College of the Virgin
Islands

American Samoa

Agricultural Experiment Station, American Samoa Community
College

Eastern Caroline Islands

Agricultural Experiment Station, College of Micronesia

USDA FUNDING OF BIOTECHNOLOGY RESEARCH
PROJECTS DURING FISCAL YEAR 1984 (CSRS
and OGPS) OR FISCAL YEAR 1985 (ARS)

<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Alabama	CSRS	4	\$ 32,516
	OGPS	(1) ^a	(4,000) ^a
	ARS	<u>4</u>	<u>269,748</u>
Total		<u>8</u>	<u>\$302,264</u>
Alaska	(No biotechnology research)		
Arizona	CSRS	7	\$25,500
	OGPS	2	70,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>9</u>	<u>\$95,500</u>
Arkansas	(No biotechnology research)		
California	CSRS	45	\$ 354,755
	OGPS	23	810,000
	ARS	(2) ^a	(53,000) ^a
	ARS	<u>19</u>	<u>2,539,583</u>
Total		<u>87</u>	<u>\$3,704,338</u>
Colorado	CSRS	11	\$ 646,408
	OGPS	1	55,000
	ARS	<u>5</u>	<u>486,689</u>
Total		<u>17</u>	<u>\$1,188,097</u>
Connecticut	CSRS	3	\$ 76,848
	OGPS	1	75,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>4</u>	<u>\$151,848</u>

^aFigures in parentheses throughout this appendix represent OGPS-funded biotechnology projects that were also reported to us by either CSRS through the questionnaire used at state agricultural experiment stations or ARS. Although they are shown here for information purposes, they are not counted twice in the totals.

APPENDIX VI

APPENDIX VI

<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Delaware	CSRS	3	\$44,410
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
	Total	<u>3</u>	<u>\$44,410</u>
District of Columbia	(No biotechnology research)		
Florida	CSRS	34	\$1,604,477
	OGPS	1	50,000
		(6) ^a	(146,000) ^a
	ARS	<u>12</u>	<u>1,878,611</u>
Total	<u>47</u>	<u>\$3,533,088</u>	
Georgia	CSRS	5	\$ 6,000
	OGPS	1	12,000
	ARS	<u>8</u>	<u>991,229</u>
	Total	<u>14</u>	<u>\$1,009,229</u>
Guam	(No biotechnology research)		
Hawaii	CSRS	1	\$60
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
	Total	<u>1</u>	<u>\$60</u>
Idaho	CSRS	7	\$300,000
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
	Total	<u>7</u>	<u>\$300,000</u>
Illinois	CSRS	9	\$ 170,598
	OGPS	3	95,000
		(1) ^a	(3,000) ^a
	ARS	<u>9</u>	<u>2,757,343</u>
Total	<u>21</u>	<u>\$3,022,941</u>	
Indiana	CSRS	25	\$712,276
	OGPS	5	62,000
		(4) ^a	(143,000) ^a
	ARS	<u>0</u>	<u>0</u>
Total	<u>30</u>	<u>\$774,276</u>	

APPENDIX VI

APPENDIX VI

<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Iowa	CSRS	20	\$723,886
	OGPS	(2) ^a	(56,000) ^a
	ARS	<u>0</u>	<u>0</u>
Total		<u>20</u>	<u>\$723,886</u>
Kansas	CSRS	9	\$124,090
	OGPS	2	16,000
		(2) ^a	(38,000) ^a
	ARS	<u>2</u>	<u>154,525</u>
Total		<u>13</u>	<u>\$294,615</u>
Kentucky	CSRS	9	\$247,921
	OGPS	1	33,000
		(2) ^a	(87,000) ^a
	ARS	<u>0</u>	<u>0</u>
Total		<u>10</u>	<u>\$280,921</u>
Louisiana	CSRS	19	\$248,975
	OGPS	0	0
	ARS	<u>1</u>	<u>640,606</u>
Total		<u>20</u>	<u>\$889,581</u>
Maine	CSRS	3	\$250,000
	OGPS	2	19,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>5</u>	<u>\$269,000</u>
Maryland	CSRS	7	\$ 427,500
	OGPS	(6) ^a	(244,000) ^a
	ARS	<u>56</u>	<u>\$8,030,270</u>
Total		<u>63</u>	<u>\$8,457,770</u>
Massachusetts	CSRS	9	\$156,000
	OGPS	5	281,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>14</u>	<u>\$437,000</u>

APPENDIX VI

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<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Michigan	CSRS	19	\$ 289,840
	OGPS	1	50,000
		(4) a	(144,000) a
	ARS	<u>4</u>	<u>710,007</u>
Total		24	\$1,049,847
Minnesota	CSRS	17	\$449,297
	OGPS	2	96,000
		(1) a	(2,000) a
	ARS	<u>0</u>	<u>0</u>
Total		19	\$545,297
Mississippi	CSRS	5	\$ 65,962
	OGPS	0	0
	ARS	<u>5</u>	<u>423,166</u>
Total		10	\$489,128
Missouri	CSRS	7	\$120,000
	OGPS	5	151,000
		(1) a	(20,000) a
	ARS	<u>5</u>	<u>546,012</u>
Total		17	\$817,012
Montana	CSRS	1	\$60,142
	OGPS	(1) a	(3,000) a
	ARS	<u>1</u>	<u>6,677</u>
Total		2	\$66,819
Nebraska	CSRS	6	\$ 17,000
	OGPS	2	38,000
		(2) a	(65,000) a
	ARS	<u>3</u>	<u>84,656</u>
Total		11	\$139,656
Nevada	(No biotechnology research)		
New Hampshire	CSRS	4	\$5,000
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		4	\$5,000

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<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
New Jersey	CSRS	3	\$5,500
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		<u>3</u>	<u>\$5,500</u>
New Mexico	CSRS	3	\$26,655
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		<u>3</u>	<u>\$26,655</u>
New York	CSRS	36	\$ 184,021
	OGPS	11	362,000
		(1) ^a	(35,000) ^a
	ARS	<u>10</u>	<u>3,728,906</u>
Total		<u>57</u>	<u>\$4,274,927</u>
North Carolina	CSRS	27	\$404,748
	OGPS	1	8,000
		(2) ^a	(98,000) ^a
	ARS	<u>0</u>	<u>0</u>
Total		<u>28</u>	<u>\$412,748</u>
North Dakota	CSRS	2	Not provided
	OGPS	0	0
	ARS	<u>9</u>	<u>\$734,730</u>
Total		<u>11</u>	<u>\$734,730</u>
Ohio	CSRS	2	0
	OGPS	3	\$89,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>5</u>	<u>\$89,000</u>
Oklahoma	CSRS	9	\$112,034
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
		<u>9</u>	<u>\$112,034</u>
Oregon	CSRS	12	Not provided
	OGPS	6	\$227,900
		(1) ^a	(55,000) ^a
	ARS	<u>3</u>	<u>309,047</u>
Total		<u>21</u>	<u>\$536,947</u>

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<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Pennsylvania	CSRS	8	\$111,120
	OGPS	2	100,000
	ARS	<u>4</u>	<u>\$384,058</u>
Total		<u>14</u>	<u>\$595,178</u>
Puerto Rico	CSRS	3	Not provided
	OGPS	<u>0</u>	0
Total		<u>3</u>	Not provided
Rhode Island	CSRS	2	\$ 80,330
	OGPS	1	45,000
	ARS	<u>0</u>	<u>0</u>
Total		<u>3</u>	<u>\$125,330</u>
South Carolina	CSRS	5	\$57,862
	OGPS	(1) ^a	(18,000) ^a
	ARS	<u>1</u>	<u>40,889</u>
Total		<u>6</u>	<u>\$98,751</u>
South Dakota	CSRS	3	\$46,000
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		<u>3</u>	<u>\$46,000</u>
Tennessee	CSRS	9	\$ 41,175
	OGPS	1	20,000
	ARS	(1) ^a	(41,000) ^a
		<u>1</u>	<u>52,123</u>
Total		<u>11</u>	<u>\$113,298</u>
Texas	CSRS	24	\$ 732,038
	OGPS	3	124,000
	ARS	<u>13</u>	<u>974,367</u>
Total		<u>40</u>	<u>\$1,830,405</u>
Utah	CSRS	17	\$278,683
	OGPS	0	0
	ARS	<u>1</u>	<u>108,768</u>
Total		<u>18</u>	<u>\$387,451</u>

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<u>State</u>	<u>USDA funding source</u>	<u>Number of biotechnology projects</u>	<u>Amount of USDA funding</u>
Vermont	CSRS	3	\$61,440
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		<u>3</u>	<u>\$61,440</u>
Virginia	CSRS	9	\$109,925
	OGPS	1	8,000
	ARS	(1) ^a	(30,000) ^a
Total		<u>0</u>	<u>0</u>
Total		<u>10</u>	<u>\$117,925</u>
Virgin Islands	(No response)		
Washington	CSRS	15	\$ 990,000
	OGPS	7	307,000
	ARS	<u>4</u>	<u>365,358</u>
Total		<u>26</u>	<u>\$1,662,358</u>
West Virginia	CSRS	3	\$ 7,000
	OGPS	0	0
	ARS	<u>3</u>	<u>150,155</u>
Total		<u>6</u>	<u>\$157,155</u>
Wisconsin	CSRS	7	\$250,323
	OGPS	7	221,000
	ARS	(3) ^a	(100,000) ^a
Total		<u>0</u>	<u>0</u>
Total		<u>14</u>	<u>\$471,323</u>
Wyoming	CSRS	4	\$51,963
	OGPS	0	0
	ARS	<u>0</u>	<u>0</u>
Total		<u>4</u>	<u>\$51,963</u>
American Samoa	(No response)		
Eastern Caroline Islands	(No response)		
TOTAL		<u>778</u>	<u>\$40,502,701</u>

BIOTECHNOLOGY RESEARCH AT STATE AGRICULTURAL
EXPERIMENT STATIONS/COLLEGES OF VETERINARY MEDICINE

<u>State</u>	<u>Number of projects</u>	<u>USDA funding</u> -----thousands-----	<u>Total funding</u>
Alabama	4	\$ 32.5	\$ 102.6
Alaska	0	0	0
Arizona	7	25.5	113.2
Arkansas	0	0	0
California	45	354.8	3,722.6
Colorado	11	646.4	1,747.1
Connecticut	3	76.8	284.7
Delaware	3	44.4	71.9
District of Columbia	0	0	0
Florida	34	1,604.5	6,023.1
Georgia	5	6.0	194.1
Guam	0	0	0
Hawaii	1	.006	49.6
Idaho	7	300.0	650.0
Illinois	9	170.6	256.5
Indiana	25	712.3	4,129.5
Iowa	20	723.9	2,893.0
Kansas	9	124.1	612.2
Kentucky	9	247.9	607.8
Louisiana	19	249.0	1,526.2
Maine	3	250.0	450.0
Maryland	7	427.5	617.4
Massachusetts	9	156.0	381.0
Michigan	19	289.8	1,063.3
Minnesota	17	449.3	1,415.0
Mississippi	5	66.0	150.3
Missouri	7	120.0	343.7
Montana	1	60.1	581.7
Nebraska	6	17.0	215.0
Nevada	0	0	0
New Hampshire	4	5.0	7.0
New Jersey	3	5.5	7.5
New Mexico	3	26.7	161.3
New York	36	184.0	2,031.0
North Carolina	27	404.8	2,146.6
North Dakota	2	a	500.0
Ohio	2	a	1.7
Oklahoma	9	112.0	484.5
Oregon	12	a	a
Pennsylvania	8	111.1	179.6
Puerto Rico	3	a	a
Rhode Island	2	80.3	896.3

^aFunding information was not provided.

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South Carolina	5	57.9	167.6
South Dakota	3	46.0	52.3
Tennessee	9	41.2	351.0
Texas	24	732.0	4,554.0
Utah	17	278.7	869.9
Vermont	3	61.4	188.9
Virginia	9	109.9	624.1
Virgin Islands	a	a	a
Washington	15	990.0	2,510.0
West Virginia	3	7.0	32.0
Wisconsin	7	250.3	3,475.4
Wyoming	4	52.0	255.7
	<u>495</u>	<u>\$10,710.2</u>	<u>\$47,697.9</u>

APPENDIX VII

EXPERIMENT STATION AND VETERINARY COLLEGE BIOTECHNOLOGY RESEARCH
PROJECTS EXPECTED TO RESULT IN ENVIRONMENTAL RELEASE

This appendix summarizes the important aspects of each of the 87 projects expected to result in the release of new, genetically engineered organisms into the environment. The projects are presented by state. The information for each project includes the title, objectives, and time frame for the release. It also reports (1) the expected scope of any problem resulting from the release, (2) the degree of effort that might be required to control the problem, and (3) whether or not risk assessment was a part of the research project. In the first project, for instance, these risk-related data are reported as "No problem/no effort/no risk assessment." The genetic engineering techniques involved in each project are also listed, followed by the GAO number assigned to each project.

ALABAMA

Title: Cellular and Molecular Genetics for Crop Improvement.
Objectives: Develop genetic vectors for use in crop plants; improve photosynthetic bacteria's capacity/resistance to herbicides after basic genetic analysis; develop strategy for control of aflatoxin biosynthesis on the basis of suppression of toxic synthesis by viral determinants.
Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transformation, and cell culture and protoplast fusion. (102)

ARIZONA

Title: Cytoplasmic Diversity and the Inheritance of Mitochondrial DNA.
Objectives: Investigate the flow of mitochondrial genes in plant populations.
Release expected in 2 to 5 years. Degree of risk unknown/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and cell culture and protoplast fusion. (306)

Title: Recombinant DNA Vectors for Gene Transfer in Crop Plants.
Objectives: Design chimaeric genes that are transformed into plants wherein the gene is expressed and the protein transported into chloroplasts.
Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transformation, and cell culture and protoplast fusion. (309)

CALIFORNIA

Title: Leaf Surface Bacterial Ice Nuclei as Incitants of Frost Injury in Plants.
Objectives: Determine the basis for ice nucleation in epiphytic bacteria; develop control measures to enhance plant supercooling and thus to avoid frost injury.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves use of recombinant DNA, site-directed mutagenesis, and transformation. (501)

Title: Genetic Improvement of Beans (*Phaseolus vulgaris* L.) for Yield, Pest Resistance, and Food Value.

Objectives: Investigate the genetic, biochemical, and physiological basis of bacteria-legume interaction and ways to improve disease resistance or affect pest resistance.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (503)

Title: Comparative Biology of Plant Pathogenic Bacteria.

Objectives: Investigate the physiological, biochemical, and genetic basis for pathological specialization, biological diversity, taxonomic classification; develop practical pathogen identification tests and improve disease diagnosis procedures.

Release expected in 2 to 5 years. Very minor problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transfection, and transformation. (504)

Title: Development of Biological Control Agents for Bacterial Diseases of Plants by Genetic Manipulations.

Objectives: Construct attenuated bacterial pathogens by recombinant DNA technique for use as biological control agents in diseases.

Release expected in 2 to 5 years. No problem/moderate effort/no risk assessment. The project involves recombinant DNA, transfection, and transformation. (603)

Title: Development of Effective, Competitive Strains of *Rhizobium leguminosarum* (Brady) and *Rhizobium japonicum*.

Objectives: Produce strains of rhizobia that will successfully compete with rhizobia for legume inoculation; inoculate fields with strains of rhizobia with higher nitrogen fixation rates and increase plant productivity.

Release expected in 2 to 5 years. No problem/little effort/risk assessment. The project involves recombinant DNA and transfection. (605)

Title: Genetics and Epigenetics of Variants Selected in Plant Cell Cultures.

Objectives: Select and characterize (genetically, physiologically, and biochemically) mutants resistant to aluminum toxicity and phosphorus deficiency from plant cell cultures.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (5801)

Title: Grape Somatic Cell Genetics.

Objectives: Develop somatic cell genetic techniques by which new grape genotypes may be obtained from cell cultures and select new grape genotypes with improved resistance to pests, diseases, and environmental stresses.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (5802)

Title: Improvement of Lettuce Through Breeding.

Objectives: Produce germplasm of lettuce with improved horticultural performance and disease resistance; broaden the genetic base of the crop in both practical and theoretical terms; develop alternative methods of crop improvement through genetic engineering techniques; reduce the time needed to respond to developing needs in the lettuce industry.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and transformation. (5806)

Title: Applications of Standard and Innovative Genetic Techniques to Rice Germplasm Improvement.

Objectives: Improve rice germplasm, using both conventional and new biotechnologies.

Release expected in 2 to 5 years. No problem/no effort/risk assessment. The project involves cell culture and protoplast fusion. (5826)

Title: The Pathogenicity and Control of Nematodes Parasitizing Grapevines in California.

Objectives: Control of nematode pathogens of grapevines and the nematode virus disease complex (X index fanleaf) by nematicide applications for replants and treatments of established vines; evaluation of genetic stock assembled at U.C. Davis for rootstock resistance to nematodes and virus.

Release expected within 1 year. No problem/no effort/no risk assessment. No information on techniques was provided. (5827)

COLORADO

Title: Stress-Tolerant Crop Plants Derived from Plant Cell Cultures.

Objectives: Derive salt-tolerant and drought-tolerant oat and wheat plants from tissue culture; investigate tolerance limits and inheritance patterns of these plants to determine physiological mechanisms of tolerance.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (702)

Title: Cell and Tissue Culture of Economically Important Species.

Objectives: Develop techniques for cell and tissue culture, protoplast fusion, and protoplast culture; develop and improve

plant regeneration methods; develop and utilize protoplast and embryo culture techniques for use in hybridization. Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves site-directed mutagenesis, microinjection, and cell culture and protoplast fusion. (703)

DELAWARE

Title: Study of Deciduous Forest Tree Tissue Cultures For Resistance to *Nectria galligena* (Bres).

Objectives: Generate somaclonal variants of deciduous forest tree species; investigate host-pathogen interaction in vitro; correlate in vitro resistance to field resistance; induce in vitro selection pressures for the production of resistant varieties of susceptible species.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (901)

FLORIDA

Title: Breeding Selection Agronomic and Grazing Evaluation of Tropical Forage Legumes.

Objectives: Screen tropical legume germplasm for winter survival, nematode resistance, soil stress tolerance, and N₂-fixation; incorporate desirable traits into adapted cultivars; evaluate selected lines for yield under grazing.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. No techniques were listed. (1103)

Title: Selection of Improved Strains of Entomopathogenic Fungi via Protoplast Fusion.

Objectives: Develop methods to produce viable protoplasts, fuse selected fungal pathotypes, and assess biological activity of regenerated fusion products.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (1109)

Title: Genetic Recombination in Baculoviruses to Analyze Host Range and Virulence.

Objectives: Expand host range and increase virulence of insect pathogenic virus from *Spodoptera frugiperda* and *Anticarsia gemmatialis*; genetic recombination and molecular manipulation of the viral DNAs will be used.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transfection, transformation, and cell culture and protoplast fusion. (1110)

Title: Genetics and Physiology of Sweet Corn Quality, Pest Resistance, and Yield.

Objectives: Determine rate-limiting steps and the genetics of these biochemical reactions for sweet corn productivity.

Release expected after 5 years. Very minor problem/little effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, and site-directed mutagenesis. (1122)

Title: Cellular and Molecular Genetics for Crop Improvement.
Objectives: Determine at the molecular level those DNA sequences that are important in gene expression in the maize endosperm.

Release expected after 5 years. Very minor problem/little effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, and transformation. (1123)

Title: Cellular and Molecular Genetics for Crop Improvement.
Objectives: Induce regeneration and plant development in selected crop plants from organs, tissue, and cells; select variant cell lines and appropriately regenerate plants for pathogen, pesticide, stress tolerance, and biochemistry characteristics.
Release planned after 5 years. No problem/no effort/risk assessment. The project involves embryo manipulation and transfer and cell culture and protoplast fusion. (1128)

IDAHO

Title: Organization and Expression of a Baculovirus Genome.
Objectives: Improve efficiency of viral pesticides by genetic engineering; facilitate commercial virus production.
Release expected in 2 to 5 years. The degree of risk is unknown. According to the researcher, "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." Regarding risk assessment the researcher says: "In the course of our studies already we have developed what we consider improved methods of assessing risks of genetically engineering viral pesticide products. . . Our methods of assessing viral gene expression in non-target hosts are extremely sensitive." The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, and transformation. (1504)

Title: Genetic Manipulation of Fungal Insect Pathogens.
Objectives: Create *B. bassiana* with enhanced control potential for insect pests by fusion or transformation.
Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transfection, and cell culture and protoplast fusion. (1505)

ILLINOIS

Title: Propagation of Perennial Plants by In Vitro Culture.

Objectives: Develop in vitro techniques to propagate and introduce new and superior perennial plants; develop techniques for rapid transfer from in vitro culture to the field.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (1604)

Title: Breeding and Genetics of Commercially Important Characters in the Apple.

Objectives: Establish a genomic library of the apple; identify and isolate genes for disease resistance, isozyme systems, and other traits; regenerate whole plants from protoplast; introduce genes of interest into plant cells.

Release expected after 5 years. No problem/little effort/no risk assessment. The project involves chemical synthesis of nucleic acids and cell culture and protoplast fusion. (1605)

Title: Exogenous Gene Transfer in Maize (Zea mays) Using DNA-Treated Pollen.

Objectives: Develop techniques to introduce individual genes into genotypes of maize without upsetting genetic balance of cultivars.

Release expected in 2 to 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, transformation, and embryo manipulation and transfer. (1606)

IOWA

Title: Gene Transfer and Mapping in Rhizobium japonicum.

Objectives: Carefully characterize genes involved in symbiotic nitrogen fixation for eventual strain construction and improvement.

Release expected in 2 to 5 years. No problem/moderate effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (1807)

Title: Nodulation and Nitrogen Fixation of PRC Rhizobium japonicum.

Objectives: Study the genetics of the response between PRC R. japonicum strains and North American soybean cultivars and do molecular genetic analysis of the nodulation genes in R. japonicum.

Release expected in 2 to 5 years. No problem/moderate effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (1812)

KANSAS

Title: Tissue Culture as a Method of Alien Gene Transfer in Wheat.

Objectives: Identify and describe the use of tissue cultures as a tool in introgression and manipulation of alien genes into wheat.

Release expected within one year. No problem/no effort/no information provided on risk assessment. The project involves cell culture and protoplast fusion. (1906)

Title: Genetic Stocks and Cytogenetic Analysis of Disease Resistance Genes in Common Wheat.

Objectives: Develop facilitator stocks for rapid genetic transfer from wild species into wheat; identify, transfer, and genetically map disease- and insect-resistance genes; develop improved germplasm wheat breeding.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves recombinant DNA. (1908)

Title: Wheat-Agropyron Hybrids: Cytogenetic Analysis of Genome in Polyploid Agropyron Species.

Objectives: Genetic analysis and evolutionary relationships of agropyron with wheat; transfer of useful traits from agropyron into wheat.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and embryo manipulation and transfer. (1909)

KENTUCKY

Title: Genetic Engineering of Tobacco Plants for Improved Health Characteristics.

Objectives: Modify chemical composition of cured tobacco leaf by altering metabolic processes to increase desirable components and decrease undesirable, hazardous components.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transformation, cell culture and protoplast fusion, and microinjection. (2002)

Title: Development of Virus-based Vector for Gene Transfer in Higher Plants.

Objectives: Same as title.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transfection, transformation, and cell culture and protoplast fusion. (2006)

LOUISIANA

Title: Pesticide Degradation and Mode of Action in Microorganisms.

Objectives: Identify the products of various pesticide metabolism; this also includes cloning the genes having to do with glyphosate resistance.

Release expected in 2 to 5 years. Very minor problem/little effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, and transformation. (2104)

Title: Construction and Expression of Genetically Modified Zein Genes.

Objectives: Construct modified zein genes in order to overcome the well-known essential amino acid deficiencies of zein.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transformation, and cell culture and protoplast fusion. (2112)

MAINE

Title: Molecular Genetics of Potatoes.

Objectives: Develop genetically improved potato varieties by direct biochemical methods.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, microinjection, transfection, transformation, and cell culture and protoplast fusion. (2202)

Title: Isolation and Characterization of Potato Virus RX-1 Protein Product.

Objectives: Isolate and characterize gene RX; elucidate the mechanism of extreme resistance to PVX conferred by gene RX.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transformation, and cell culture and protoplast fusion. (2203)

MASSACHUSETTS

Title: Development of Tissue Culture Techniques for the Genetic Improvement of Turf Grasses and Forage Grasses.

Objectives: Same as title.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves site-directed mutagenesis, cell culture and protoplast fusion, somaclonal variation, protoclinal variation, and cell selection. (2403)

Title: Control of Postharvest Decay of Fruits and Vegetables

Objectives: Determine etiology and epidemiology of pathogens; elucidate mechanisms of tissue breakdown; develop new methods of control of postharvest diseases.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (2405)

MICHIGAN

Title: Nutritional Requirements for Fishes Cultured in Michigan.

Objectives: Develop triploidy in Pacific salmon; such fish should be sterile and thus should not mature sexually, resulting in exceptionally large sizes.

Release expected within 1 year. Very minor problem/no effort/risk assessment. The project involves transformation. (2508)

Title: Forest Tree Improvement Through Genetic Engineering and Tissue Culture.
Objectives: Develop faster growing tree lines better adapted to the sites where they are planted.
Release expected in 2 to 5 years. No problem/little effort/no risk assessment. The project involves cell culture and protoplast fusion. (2514)

MINNESOTA

Title: Genetic Biotechnological Development, Characterization, and Preservation of Poultry Germ Plasm.
Objectives: Research in cytogenetics, molecular genetics, cell culture, reproductive biology, and genome evaluation for the identification, location, and transfer of useful genes to improve efficiency of poultry production.
Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, microinjection, transfection, transformation, embryo manipulation and transfer, and cell culture and protoplast fusion. (2601)

Title: Biochemical and Developmental Genetics of Higher Plants.
Objectives: Determine molecular basis of cytoplasmic male sterility in corn; select and characterize mutants with improved amino acid nutritional quality in corn; develop nonconventional methods for gene transfer in corn.
Release expected in 2 to 5 years. No problem/little effort/no risk assessment. The project involves recombinant DNA, transformation, and cell culture and protoplast fusion. (2604)

Title: Cell and Tissue Cultures for Plant Improvement.
Objectives: Develop and improve cell and tissue culture methods in corn and other crops.
Release expected in 2 to 5 years. No problem/little effort/no risk assessment. The project involves cell culture and protoplast fusion. (2605)

Title: The Control Regions of Zein Genes.
Objectives: Cloning and sequencing of the control regions of the zein genes; comparison of the primary structure of the 5' flanking region from different subfamilies of zein genes.
Release expected in 2 to 5 years. No problem/no effect/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transfection, transformation, and cell culture and protoplast fusion. (2606)

Title: Characterization and Transferability of Plasmid DNA in Dairy Starter Cultures.
Objectives: Apply biotechnology for strain construction strategies, for improving bacteria used in dairy, meat, and vegetable fermentation processes.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (2609)

Title: Gene Action in Angiosperms.

Objectives: Generate information concerning gene expression in higher plants by elucidating genetic, biochemical, and physiological mechanisms by which an organism actively controls growth and differentiation in an intimate relationship.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves transformation, embryo manipulation and transfer, and cell culture and protoplast fusion. (2610)

MISSISSIPPI

Title: Overcoming Factors Limiting Biological Nitrogen Fixation by Leguminous Plants.

Objectives: Determine factors controlling legume infection and nodule development to enhance effectiveness of inoculation of seeds or soil; isolate and characterize indigenous and important rhizobium species tolerant to stress factors.

Release expected in 2 to 5 years. Degree of risk unknown/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (2705)

MISSOURI

Title: Inception of Symbiotic and Tumorigenic Plant-Microorganism Associations.

Objectives: Identify mechanisms of rhizobium cell entering into symbiosis with legume roots; agrobacterium cell transfer DNA to plant cells.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transformation, and cell culture and protoplast fusion. (2804)

Title: The Study of the Mechanism of Heredity in Corn.

Objectives: Understand the mechanisms of heredity in corn through study of chemical mutagenesis; study the controlling elements and the genetic control of embryo lethality, development, and disease symptoms.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves site-directed mutagenesis. (2806)

NEBRASKA

Title: Corynebacterium Pathogens of Corn and Wheat: Serology and Genetics.

Objectives: Obtain gene transfer system(s) in phytopathogenic corynbacter, especially corn and wheat pathogen; genetic mapping would be contingent on successful gene transfer and expression.

Release expected after 5 years. No problem/little effort/risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transfection, and transformation. (3003)

NEW JERSEY

Title: Molecular Biology of Pathogen-Induced Chlorosis.

Objectives: Develop a model system suitable for studying toxin-induced chlorosis at the molecular level; determine the biomedical basis for toxin-induced chlorosis; identify and isolate genes that confer resistance to phytotoxins.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transformation, and cell culture and protoplast fusion. (3301)

Title: Resistance to Photosystem II Herbicides.

Objectives: Identify the specificity-determining domains of the herbicide binding site; transfer resistance to several different structural classes of photosystem II herbicide to cyanobacteria and higher plants.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, transformation and cell culture and protoplast fusion. (3302)

NEW MEXICO

Title: Cellular and Molecular Genetics in Crop Improvement.

Objectives: Improve methods for plant modification, selection, regeneration, and propagation through cell and tissue culture; identify agriculturally important genetic systems.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves transformation and cell culture and protoplast fusion. (3401)

Title: Tissue and Cell Culture Methods in the Improvement of New Mexico Crops.

Objectives: Interface with breeding programs aimed at New Mexican commodities.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (3402)

Title: Experimental Use of Isozymes in Applied Plant Genetics Research.

Objectives: Construct a chromosomal linkage map for genes coding for enzymes; use those mapped genes to find and track other genes of economic importance.

Release expected within 1 year. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (3403)

NORTH CAROLINA

Title: Development of New Biodegradable Insecticides from Studies of Juvenile Hormone Esterase Regulation.

Objectives: Isolate and assess timing and prioritization of biotic induction factors for juvenile hormone esterase (JHE) biosynthesis for use in tissues of larval trichoplusia NI and Manduca Sexta; synthesize selective, irreversible inhibitors of

JHE, purify JHE; obtain specific antibody of JHE for enzyme-linked immunosorbent assay and probe for regulation study of JHE concentration.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, and cell culture and protoplast fusion. (3601)

Title: Plasmid-like DNAs in Maize Mitochondria.

Objectives: Investigate several plasmid-like DNA species found associated with the mitochondria of maize; characterize the plasmid-like DNAs with regard to organization, genetic information, and their transpositional activity; study of transposition useful in developing transfer vectors for genetic engineering of maize.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, and transformation. (3602)

Title: Molecular Biology of the Homofermentative Lactic Acid Bacteria.

Objectives: Develop a genetic transfer system for the homofermentative lactobacilli and pediococci; analyze the molecular genetics and metabolism of these organisms; develop recombinant DNA methods in construction of strains better suited for food preservation and other beneficial uses.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, transformation, and cell culture and protoplast fusion. (3603)

Title: Development of Nonsexual Techniques for Genetic Engineering of Zea mays (L).

Objectives: Identify, isolate, and characterize mitochondrial genes of corn and tobacco; develop a transformation system for mitochondrial genes.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transformation, and cell culture and protoplast fusion. (3605)

Title: Cellular and Molecular Genetics for Crop Improvement.

Objectives: Regulation of gene expression and the delivery of genetic material to higher plants and associative microorganisms; somatic cell genetic and plant development--modify, select, regenerate, and propagate plants through cell and tissue culture.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and cell culture and protoplast fusion. (3607)

Title: In Vivo and In Vitro Comparison of Membranes of Regenerative and Nonregenerative Protoplasts.

Objectives: Characterize membrane of protoplast that readily regenerates into callus and undergoes fusion; compare these characterizations with those of nonregenerative protoplasts; manipulate chemically the membranes of nonregenerative protoplasts to enhance regeneration.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (3609)

Title: Cultured Plant Cells and Tissues for the Study of Solute Regulation and Morphogenesis.

Objectives: Clonal propagation of forest trees (pine)--regenerate A-plant from callus of mature tree and evaluate clone fidelity in greenhouse/field; in vitro study of fusiform rust and blister rust; modulate cell differences in cell cultures; fundamental processes of organogenesis (soybean, cotton); plant regeneration from callus; nutrient medium effects on organogenesis, cell differentiation, and cell metabolism.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (3610)

Title: Mechanisms of Membrane Fusion in Fusogenic Carrot Protoplasts.

Objectives: Develop method to routinely obtain fusogenic protoplasts; fuse spontaneously and with calcium at greater than 50% efficiency; determine why these protoplasts are so fusogenic and elucidate the mechanism of membrane fusion.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves cell culture and protoplast fusion. (3614)

Title: The Evolution and Systematics of Organelle DNAs in Relation to Systematics in Higher Plants.

Objectives: Characterize variability of organelle DNAs; use differences in organelle DNAs to devise a taxonomic hierarchy; compare evolutionary relationships determined by organelle DNAs with those derived by conventional systematic methods.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, and transformation. (3621)

Title: Genomic Expression and Replication of Polycistronic Plant Viruses.

Objectives: Strategy of plant virus genomic expression will be analyzed using potyviruses as a model system; three phenotypically distinct strains of tobacco etch virus will be analyzed to correlate phenotypic differences at the transcriptional level of expression.

Release expected within 1 year. No problem/no effort/risk assessment. The project involves recombinant DNA and chemical synthesis of nucleic acids. (3623)

Title: In Vitro Selection of Attenuated Virus Strains.

Objectives: Investigate the molecular basis for attenuation; determine molecular differences in wild-type sindbis virus (SB) and attenuated SB mutant, selected for rapid growth in vitro to apply a direct selective pressure for rapid penetration of tissue culture cells; isolate other attenuated strains of SB and attenuated mutants of western equine encephalitis and bovine viral diarrhea virus.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA; chemical synthesis of nucleic acids; transformation, and cell culture and protoplast fusion. (3625)

Title: Structure, Function, and Evolution of DNA Sequences in Eukaryotes: An Approach to Genetic Engineering.

Objectives: Study of structure, organization, and evolution of DNA sequences in maize and loblolly pine; specific genes are isolated and DNA sequence is determined to learn organization, regulation, and evolution of DNA sequences concentrating on transposable elements and on ribosomal RNA genes.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, transformation, and cell culture and protoplast fusion. (3627)

Title: Genetics of Nitrogen Fixation in *Azotobacter Vinelandii*.

Objectives: Develop in vivo and vitro mutagenic process (transposon-mediated and localized mutagenesis; plasmid-mediated conjugative gene transfer); study the genetics of nitrogen fixation in *A. vinelandii*.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and transformation. (3631)

OREGON

Title: Viral and Chemical Interactions with Cells.

Objectives: Determine safety of biological pesticides and increase efficiency of the biological agents that can be genetically engineered.

Release expected after 5 years. Very minor problem/little effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, transfection, and transformation. (4002)

Title: Identification, Characterization, and Transfer of Hydrogen Uptake Genes Between Different Rhizobial Strains and Species to Increase Nitrogen Fixation in Agriculturally Important Leguminous Plants.

Objectives: Transfer hydrogen uptake genes among different rhizobia bacteria that form nitrogen-fixing nodules on these plants.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (4009)

SOUTH CAROLINA

Title: Bacterial Extrachromosomal Factors Controlling Rhizobium japonicum Soybean Symbiosis.

Objectives: Determine the genetic basis for initial attachment of Rhizobium japonicum to soybean roots; transfer extrachromosomal element responsible for rhizobia attachment to soybean roots between different serotypes; transferral will be used to enhance N-fixation; identify DNA gene segment(s) responsible for specific attachment and construction of a symbiotic gene probe.

Release expected in 2 to 5 years. Very minor problem/some effort/no risk assessment. The project involves recombinant DNA, site-directed mutagenesis, and transformation. (4404)

Title: Live Mutants Pasteurellas Multocide Vaccine for Prevention of Fowl Cholera in Turkeys.

Objectives: Mutate CU strain of Pasteurella to less pathogenic strain; maintain or elevate immune response to organism.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. No information provided on techniques to be employed. (4406)

TENNESSEE

Title: Chemical Control of Plant Growth in Florists' Crops.

Objectives: Test growth regulating chemical effects on floriculture crops; assess application to growth, water-use efficiency and transpiration rates during production.

Release expected in 2 to 5 years. No problem/no effort/risk assessment. The project involves cell culture and protoplast fusion. (4603)

Title: Propagation of Ornamental Plants.

Objectives: Develop methods for efficient production of high quality vegetative propagules; in vitro cloning to develop better methods of propagating ornamental plants and producing new strains and types.

Release expected in 2 to 5 years. No problem/no effort/risk assessment. The project involves cell culture and protoplast fusion. (4604)

TEXAS

Title: Studies of Insect Neurohormones for their Applied Potential.

Objectives: Identify, isolate, and structurally characterize peptidic neurohormones of insects; use neurohormone structure to isolate neurohormone gene for cloning into insect baculovirus

cloning-expression vectors; improve the viral pathology as a biocontrol agent.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and chemical synthesis of nucleic acids. (4702)

Title: Embryo Transfer in Domestic and Laboratory Animals.

Objectives: Improve efficiency and usefulness of embryo transfer, gene transfer, and related technologies in mammals; use to reduce animal disease, produce food and fiber, and preserve wildlife.

Release expected after 5 years. No problem/no effort/risk assessment. The project involves recombinant DNA, microinjection, and embryo manipulation and transfer. (4704)

Title: Bovine Brucellosis Research.

Objectives: Identify and employ differential diagnostic antigens; improve synthetic or recombinant vaccine; elucidate mechanism of molecular pathogenesis.

Release expected in 2 to 5 years. Very minor problem/no effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, microinjection, transfection, embryo manipulation and transfer, and cell culture and protoplast fusion. (4706)

Title: Development of Insect Viruses as Pest Control Agents.

Objectives: Develop baculovirus pesticides; use recombinant baculovirus and insecticides as viral expression vector for proteins of medical and agricultural importance.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transfection, transformation, and cell culture and protoplast fusion. (4709)

Title: Use of Molecular Biology for Potato Improvement.

Objective: Same as title.

Release expected in 2 to 5 years. No problem/little effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transfection, and transformation. (4710)

Title: Insect Parasite-Host Relationships.

Objectives: Develop methods for genetic engineering of beneficial insect parasitoids using symbiotic parasitoid viruses.

Release expected after 5 years. No problem/little effort/no risk assessment. The project involves chemical synthesis of nucleic acids, embryo manipulation and transfer, and cell culture and protoplast fusion. (4711)

Title: Direct Genetic Manipulation in Higher Plant:
Extrachromosomal Gene Amplification-Cloning Vehicle for
Genetic Engineering in Plant Tissue Culture.

Objectives: Identify and characterize gene or gene systems with importance to agriculture as positive selection markers; identify gene systems for direct agricultural technologies; identify gene systems as model systems for constructing hybrid enzymes and chimeric genetic systems.

Release expected in 2 to 5 years. No problem/little effort/risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transfection, transformation, and cell culture and protoplast fusion. (4712)

Title: Analysis of the Structure and Action of a Flavin Hydroxylase by Recombinant DNA Technology.

Objectives: Develop understanding of how interactions of flavoprotein with flavin moiety can regulate chemical activity of flavin; general area is protein structure and function.

Release expected in 2 to 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA, chemical synthesis of nucleic acids, site-directed mutagenesis, transfection, and transformation. (4715)

UTAH

Title: Characterization of Plasmid DNA in *Streptococcus cremoris* for Genetic Engineering of Dairy Starters.

Objectives: Develop genetic engineering techniques in group N streptococci for the improvement of starter cultures; will use these cultures in industrial dairy fermentations.

Release expected after 5 years. No problem/no effort/no risk assessment. The project involves recombinant DNA and transformation. (4811)

WASHINGTON

Title: Ecological Factors Influencing the Persistence of *Rhizobium* in Soil and Competition in the Rhizosphere.

Objectives: Determine the mechanism of host-strain specificity in legumes and the nature of competition for nodulating sites.

Release expected in 2 to 5 years. Very minor problem/some effort/no risk assessment. The project involves recombinant DNA. (5201)

Title: Physiological Studies on Vegetables and Vegetable Seed Crops.

Objectives: Develop methods for selecting superior yielding lines of processing peas from cultivars and breeding lines by utilizing physiological stresses that normally occur in western Washington crop production.

Release expected in 2 to 5 years. No problem/no effort/risk assessment. The project involves cell culture and protoplast fusion. (5203)

STATE BREAKDOWN OF 87 BIOTECHNOLOGY RESEARCH PROJECTS
EXPECTED TO RESULT IN ENVIRONMENTAL RELEASE

<u>State</u>	<u>Number of projects</u>
Alabama	1
Arizona	2
California	10
Colorado	2
Delaware	1
Florida	6
Idaho	2
Illinois	3
Iowa	2
Kansas	3
Kentucky	2
Louisiana	2
Maine	2
Massachusetts	2
Michigan	2
Minnesota	6
Mississippi	1
Missouri	2
Nebraska	1
New Jersey	2
New Mexico	3
North Carolina	13
Oregon	2
South Carolina	2
Tennessee	2
Texas	8
Utah	1
Washington	<u>2</u>
Total states	28
Total projects	<u>87</u>

BIOTECHNOLOGY RESEARCH ACCOMPLISHMENTS SINCE OCTOBER 1, 1982,
AT STATE AGRICULTURAL EXPERIMENT STATIONS AND
COLLEGES OF VETERINARY MEDICINE

ALABAMA

1. Atrazine resistant mutants of photosynthetic bacteria were characterized.
2. Vector for crop plants was developed using geminivirus replicative DNA.
3. Monoclonal antibodies were developed for Mycoplasma gallisepticum and synoviae characterization of these is in progress.

ARIZONA

1. First single strand DNA plant virus sequenced was the bean golden mosaic virus by our plant pathologists in late 1984.
2. Our biochemists established a reliable plant transformation system with stable chromosome integration of new genes.
3. Our biochemists have patented a shuttle vector system for the modification of chloroplast metabolism.

ARKANSAS

1. Some tissue culture of various plant material. A little monoclonal antibody work.

CALIFORNIA

1. Cloning and transfer of genes from malolactic bacteria into yeast, allows for wine fermentation to proceed simultaneously.
2. First transfer of a gram positive procaryote gene into a eucaryote.
3. Selection of low temperature and salt tolerant genetics in pollen survival studies.
4. Regeneration celery tissue culture; discovery of somaclonal variants; selecting disease resistant celery using tissue culture.
5. Use tissue cultures in lettuce improvement including somaclonal variation from protoplast regenerants and other cultures.
6. Develop technique to identify livestock sex before birth; selection of males for meat production and females for milk production.
7. Convert solar energy more efficiently into food/fiber; more efficient nitrogen fixation in legumes; nitrogen transfer fixation into crops.
8. Isolation and transfer of osmotic tolerance gene from salt tolerant bacteria into nitrogen-fixing bacterium.
9. Clone plant pathogen bacteria genes interacting with plant disease resistance genes; code/clone genes of pectate lyases of Erwinia chrysanthemia.

COLORADO

1. Sponsor three different types of research using biotechnology techniques which emphasize improvement of cereal grains.
2. Chromosomes of several barley strains are being mapped to associate specific traits to particular gene action.
3. Salt tolerance of major cereals--rice, oats, millet, wheat--is researched by subjecting cell cultures to salt concentrations. These cultures are then tested for inheritable salt tolerance traits.
4. Salt protoplast fusion and cell culture of several economically important plants is used to refine protoplast fusion techniques. These techniques produce desirable germplasm that can be used for breeding research.

CONNECTICUT

1. Growth of plant cells on solid medium dependent entirely on photosynthesis was demonstrated.
2. Genetic changes of photosynthesis can now be studied in plant cell culture.
3. A mutation conferring resistance to isonicotinic acid hydrazide (INH) was characterized in plant cells. The mutant enzyme correlated with growth of cells with INH, and the mutation transmitted to plants. This was one of the first examples showing production of biochemical mutants of higher plants from plant cells.
4. Plants have been selected with resistance to oxygen stress using plant cells.

DELAWARE

No accomplishments listed.

FLORIDA

The shrunken-1 gene of corn has been cloned and sequenced. cDNA clones of potyvirus express viral proteins in E coli. Potyvirus genome mapped. Soybean gene for small heat shock protein transferred into sunflower tumor by t-DNA based vector. The lactose operon has been introduced into *Zymomonas mobilis* with expression of both the permease and galactosidase genes. A cDNA has been prepared for uteroferrin, the progesterone-induced iron transport protein secreted by the swine uterus. Other cDNAs involved in reproductive physiology have been prepared, including a cDNA probe for pro-oxyphysin from cattle. Monoclonal antibody developed for use as better serologic test for brucellosis & genetically engineered interferon for bovine viral disease. Somatic embryogenesis regenerated mango, eugenia & other trees of the myrtaceae tree

family. Somatic embryogenesis technique used to regenerate citrus species & variety of cereals & related grass species. Maize-sorgum-peanut protoplast isolated & divided & produced callus & transposon-like sequence & cDNA libraries cloned from maize. Production of a live calf following embryo transfer in water buffalo. Monoclonal antibodies for improved brucellosis immunodiagnosis. Use genetically engineered interferons to treat bovine viral disease.

GEORGIA

1. Progress in synthesizing complementary DNA to several strains of peanut mottle virus. Will make nucleic acid hybridization studies to determine relationships among viral strains.
2. Develop lines of pearl millet with high ability to support bacterial acetylene reduction activity (ARA) in seedling agar.
3. Develop lines of pearl millet with high ability to support bacterial ARA in soil N₂-fixing microbes.
4. Development of technique which may be useful in enhancing N₂-fixing associations between soil microbes and grasses.
5. Development of an inexpensive, portable container and procedure for freezing embryos.

GUAM

No accomplishments listed.

HAWAII

1. Yeast complementary DNA (cDNA) cloning vector previously constructed has been tested and found to express cDNA inserts.
2. A small library of cDNA has been produced from total mRNA extracted from maize seedling. cDNA clone of approximately 1,500 base pairs was isolated from partial library construction of the maize seedling mRNA.

IDAHO

Organization of baculovirus genome, use of baculoviruses as genetic vectors. Bioconversion of lignin to useful biochemicals. Biomass & alcohol production via yeasts. Development of fungi as biological pesticides. Hybridoma and monoclonal antibody techniques for disease diagnosis. Conventional breeding of wheat, barley, oats, pulse, and vegetable crops. Protoplast fusion for genetic hybridization. Biocontrol of plant disease & insect pests (mosquitoes) via bacteria.

ILLINOIS

No accomplishments listed.

INDIANA

1. Capability to regenerate corn plants from small groups of embryonic cells.
2. Identification of the number of genes that code for corn storage proteins.
3. Identification of sites in zein proteins where amino acid substitutions might be made to increase lysine content. This increase occurs without seriously altering protein function.

IOWA

1. Solid phase radioimmunoassay (spira) developed to detect virus in mosaic soybean, lettuce, & maize dwarf seeds. A single monoclonal antibody was developed for the spira system which differentiates infected & uninfected seeds. The approximate level of the pathogen in the infected samples could be measured. Spira system used to eliminate seed lots of soybean, lettuce, & maize that are too highly infected with virus for commercial use.
2. Subunit transmissible gastroenteritis of swine vaccine was developed.
3. Production of milk antibody following intramuscular injection of 23000 d unit was demonstrated.
4. Pseudorabies subunit vaccine & complementary negative diagnostic subunit antigen developed & are being field evaluated.
5. Regeneration of age atrophied canine thymus by bovine anterior pituitary growth hormone has been demonstrated.
6. Isolate cytoplasmic vesicle membrane fusion inhibit factor isolate from bacteria which initiate intracellular infection.
7. Technique for microinjection of mitochondria into 2 cell stage mammalian embryos.
8. Cryopreservation techniques for mammalian embryos are being extended. Hormonal control of parturition mechanism, hormonal control of cervical softening. Interdependence of support cells & neuronal development using brain tissue transplant. Basic pain transmission mechanism under study. Development of brain as influenced by androgens being defined.

KANSAS

1. Embryo culture used to extend the wheat hybridization range was successful in getting two new wheat xagrophyron hybrids. Wheat x rye amphidiploids formed callus from immature embryos and contained translocations, deletions, and amplifications. Method increases instability of chromosomes in tissue culture and represents a useful tool in introgressing alien genes or chromosomes into wheat.

KENTUCKY

No accomplishments listed.

LOUISIANA

1. Production of beef cattle offspring from 1/4 embryos.
Production of pigs from split embryos.
2. Development of short-statured "saturn" rice through tissue culture.
3. Development of preliminary vaccine for control of anaplasmosis.

MAINE

No accomplishments listed.

MARYLAND

1. Seventeen maternally derived dihaploids for increased tobacco lodging resistance have been developed.
2. Monoclonals produced against aflatoxin b1 and afb-diolthase for radio-immunoassay and enzyme-linked immunosorbent assay.
3. Genes encoding pectate lyase (pl) were cloned from *Erwinia chrysanthemi* into *Escherichia coli*.
4. Plantlet regeneration and embryogenesis of *Rubus* sp. (blackberries) and plantlet regeneration of *Fragaria* (strawberry).
5. Determined plantlet screening for herbicide resistance unspecific.
6. Determined resistance of sweet potato cultivars to excess soil aluminum was not at the cellular level.
7. Determined human interferon @>10 I.U. inhibited tobacco mosaic and potato m viruses, ineffective with other potato viruses.
8. Generated and demonstrated viability of strawberry protoplasts.

MASSACHUSETTS

Three tissue culture laboratories were established with programs for turfgrass, ornamentals, and plant virus tissue. Pectic enzyme genes cloned into *e. coli*. Apple viruses cultured in n and 2n isolated protoplast cultures. Monoclonal antibodies for Mareks disease and brucellosis.

MICHIGAN

1. Regenerated tomato leaf protoplast into whole plants and created somatic hybrid plants between tomatoes and wild species.

2. Somatic cell culture systems have been developed for several tree species.
3. Antibodies for immunochemical assays of mycotoxins in foods and feeds have been produced.
4. Hypovirulant strains of chestnut blight virus have been used to control canker diseases of chestnut.
5. Characterize genetic regulatory mechanism in *Rhizobium japonicum* (Brady), soybean symbiont.
6. Can manipulate genome to increase N-fixation levels.
7. Photosynthesis gene CO₂-fixation cloned and structure changed leading to possible genetic improvement of photosynthesis.
8. Gene giving resistance to a new class of herbicides was identified and ongoing work to transfer this gene to crop species.

MINNESOTA

No accomplishments listed.

MISSISSIPPI

No accomplishments listed.

MISSOURI

Embryo transfers. Mutants produced maize. Chromosomes mapped and used in triticale and wheat research. Tomato fruits grown in tissue culture. DNA transfer agrobacterium of plant cells.

MONTANA

In process of developing/patenting *Rhizobium meliopi* transconjugate. It will be useful in the study of plant pathogenicity.

NEBRASKA

Establishment of tissue culture systems. Development of genetic transfer system (bacteria). Isolation and characterization of potential cloning vector (plant).

NEVADA

No accomplishments listed.

NEW HAMPSHIRE

Micropropagation by cell culture has been worked out for several plants.

NEW JERSEY

Selected for spontaneously occurring cyanobacterial mutants resistant to structurally different herbicides. These mutants inhibit photosynthetic electron flow by binding to the qb apoprotein. These mutants are characterized by examining qb protein & gene encoding this electron transport component. Herbicide resistance conferred on wild-type cells by transformation with DNA isolated from various mutants. Transfer chloroplast PSB, a gene from triazine resistant higher plant weel, to cyanobacteria. Select most amenable system to study molecular biology of pathogen-induced chlorosis. Using tentoxin; have chosen several species within genus Nicotiana as host plants. Obtained seeds of several pairs of closely related species of Nicotiana (sensitive/non-sensitive to tentoxin), protoplast & cell culture of these plants, & isolate genes for a & b subunits of cfl to compare nucleotide sequence.

NEW MEXICO

1. State legislature created a center for research and development in the Rio Grande research corridor.
2. Chromosome map of tomato has been enhanced.
3. Chromosome map of chili has been initiated.
4. Differentiation of onion and chili from callus.

NEW YORK

Studies of regulating nutrient partitioning show administration of bovine growth hormone increases milk production 41%. There were no effects on milk quality and composition and no ill effects on animal health. Bovine growth hormone increases mammary development in young dairy animal with increases to 38% of mammary secretory tissue. Porcine growth hormone injected daily into sows for 5 weeks before & after farrowing greatly decreases baby pig mortality.

NORTH CAROLINA

1. Development of new procedure that uses calcium to facilitate plant protoplast fusion.
2. Development of in vitro procedure for selection for resistance to fusiform rust in pines.
3. Use of maternal haploid plants for rapid screening for resistance to viruses and nematodes in tobacco.

NORTH DAKOTA

Potato clone systems have been significantly improved. A mechanism for disease resistance transfer is being explored.

OHIO

No accomplishments listed.

OKLAHOMA

1. Cauliflower mosaic virus genome was characterized re: transcription and translation. CMV shows good potential as a "gene transfer vehicle."
2. Bacterial avirulence genes in cotton identified. Products elicited by them in characterization of cultivar specificity to cotton bacterial blight.
3. Tissue culture used successfully in studying biochemical nature of bacterial disease resistance.
4. Tissue culture used successfully in studying embryo rescue of wide crosses in peanuts and wheat.
5. Tissue culture used successfully in studying presence of somoclonal variations.

OREGON

No response.

PENNSYLVANIA

1. 225 new mushroom lines, one 40% larger than either parent.
2. Exchanged genetic material between *Escherichia coli* and blue-green algae.
3. Screened cotton for salt and herbicide resistance using tissue culture methods.
4. Isolated protein and cloning gene for glutathione-s-transferase isoenzymes in mammals (rats).
5. Implemented DNA probe detecting *E. coli* enterotozigenic genes using colony blot hybridization.
6. Measured effect of recombinantly derived growth hormone. Detailed mode of action using cell culture.
7. Determined effect of recombinantly derived products of mammary cells in culture. Whole animal studies to follow.
8. Regenerated cotton plant from callus cultures.

PUERTO RICO

No response.

RHODE ISLAND

1. Patented process for more efficient cloning of grapes, producing vines superior to parent material. This is half of system for genetically engineering grapes. We now have suitable gene receptor for delivery of gene transformation vector, which is next half of system to develop.

2. In vitro regeneration of maize has been demonstrated to the small seedling stage. The conditions of regeneration will be defined and somatic variation is being investigated.

SOUTH CAROLINA

No accomplishments listed.

SOUTH DAKOTA

Creation by genetic engineering of high-ethanol-yielding yeast strains that can ferment whey.

TENNESSEE

Two significant developments in orchardgrass in vitro culture system were accomplished. These accomplishments include the production of embryos directly from mesophyll cells. A second accomplishment includes the full development of embryos directly in liquid suspension culture. Cereal and grass species research allow for studies on embryo development, mutant selection, and, maybe, genetic engineering.

TEXAS

Microinjection technique introduces genetic materials into previously fertilized embryo. Potential-to impact new traits, faster progress. Vector system-baculoviruses for rapid production of new genetically expressed materials (100 X more efficient). Plant propagation-selection and increase of dioecious (female, fruit-bearing) trees via tissue culture (date palms). Protein enhancement-improved quality and quantity of proteins in potato tubers. Made library of monoclonal antibodies-brucellosis against major Brucella abortus antigens used in diagnostics and vaccines.

UTAH

No accomplishments listed.

VERMONT

Using recombinant DNA methods, have data which suggest several approaches to controlling plant pathogenic fungi.

VIRGINIA

No accomplishments listed.

WASHINGTON

No accomplishments listed.

WEST VIRGINIA

1. American chestnut has been cloned by stem culture for the first time. Media was developed where the tissue from 4 major chestnut species was grown at same rate for disease-resistance studies.
2. A healthy, normal calf was born to an ovariectomized cow by embryo transfer.

WISCONSIN

1. Basic knowledge on use of tissue culture in plant breeding.
2. Use cloned embryos to facilitate more precise testing of gene-environment interaction allowing increased livestock production.
3. Use of micropropagation permits high degree of control on disease and pests.

WYOMING

No accomplishments listed.

Institutions Receiving Competitive Grants

SECTION I

(To be filled out by Director, State Agricultural Experiment Station)

SECTION II

QUESTIONS RELATING TO SPECIFIC BIOTECHNOLOGY RESEARCH PROJECTS

For each biotechnology research project funded in whole or in part by USDA at this agricultural experiment station, please answer the following 18 questions. IF NECESSARY, PLEASE REPRODUCE THESE QUESTIONS SO THAT YOU CAN PROVIDE ANSWERS FOR EACH OF YOUR ONGOING RESEARCH PROJECTS.

PART A. SPECIFIC PROJECT INFORMATION

1. What is the project's title? PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

2. Does the project have a CRIS identification number?

1. Yes...WHAT IS THE CRIS IDENTIFICATION NUMBER? _____

2. No....IS THERE ANY OTHER IDENTIFICATION NUMBER?
IF SO, PLEASE PROVIDE. _____

3. Are there keywords reported for this project in the CRIS?

1. No...Keywords are not reported/Project is not in the CRIS

2. Yes...(SPECIFY UP TO 10 KEYWORDS AND PLEASE PRINT CLEARLY.
EACH KEYWORD SHOULD BE NO LONGER THAN 50 CHARACTERS.)

- a. _____
- b. _____
- c. _____
- d. _____
- e. _____
- f. _____
- g. _____
- h. _____
- i. _____
- j. _____

Note: This summary does not include 45 projects OGPS cosponsors with CSRS and ARS.

4. If we need additional information, who should be contacted regarding this particular project?

Scientist's Name _____

Title _____

Telephone Number () _____
 Area Code

State (PLEASE PROVIDE TWO-LETTER POSTAL ABBREVIATION) _____

5. Briefly, what are the project's major objectives? PLEASE PRINT CLEARLY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

6. Which of the following genetic engineering techniques are being used in this project? (CHECK ALL THAT APPLY.)

1. Recombinant-DNA N = 66
2. Chemical synthesis of nucleic acids N = 0
3. Site-directed mutagenesis N = 5
4. Microinjection N = 0
5. Transfection N = 0
6. Transformation N = 8
7. Embryo manipulation and transfer N = 0
8. Cell culture and protoplast fusion N = 8
9. Other (SPECIFY PLEASE LIMIT YOUR RESPONSE TO 50 WORDS OR LESS) N = 14

7. For how many months has this project been funded? (WRITE IN NUMBER OF MONTHS.)

Number of months Average 26.3 (N = 100)

8. How many months longer is this project expected to run? (WRITE IN NUMBER OF MONTHS.)

Number of months Average 16.5 (N = 100)

9. Is it expected that this project will involve the release of genetically engineered organisms into the environment? (CHECK ONE.)

1. Yes...CONTINUE TO QUESTION 10 N = 4

2. No....SKIP TO QUESTION 14

10. When will this project involve the release of genetically engineered organisms into the environment? (CHECK ONE.)

1. Within 1 year N = 1

2. In 2 to 5 years N = 2

3. After 5 years N = 1

11. Will the National Institutes of Health's Recombinant DNA Advisory Committee's approval for the deliberate release into the environment of a genetically engineered organism be sought? (CHECK ONE.)

1. Yes, it is applicable and will be sought N = 1

2. No, it is not applicable and will not be sought N = 3

3. No, it is applicable and will not be sought...PLEASE EXPLAIN WHY AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.

12. In your opinion, if the genetically engineered organisms produced by this project were released into the environment, how much of a problem would or could such action be to the environment? (CHECK ONE.)

1. No problem N = 3
2. Very minor problem
3. Minor problem
4. Moderate problem
5. Major problem
6. Very major problem
7. Don't know N = 1

13. In your opinion, how much effort would it take to correct any such problems which might result from releasing into the environment genetically engineered organisms produced by this project? (CHECK ONE.) Not provided

1. Situation could not be corrected (situation would be uncontrollable)
2. Very great effort
3. Great effort
4. Moderate effort
5. Some effort
6. Little effort
7. No effort (situation would be self-controlling)

PART B. STAFFING AND FUNDING INFORMATION

16. How many paid researchers worked on this project during FY 1984? If this research project combined both biotechnology and conventional procedures, then report only the FTEs devoted to the biotechnology part of the project. FTEs should be reported to the nearest tenth.

a. Number of faculty FTEs Not provided

b. Number of graduate students FTEs Not provided

c. Number of technical support staff FTEs Not provided

17. To the best of your knowledge, how many FTEs are expected to be expended on this project (biotechnology only) over its entire life? (INCLUDE FACULTY, GRADUATE STUDENTS, AND TECHNICAL SUPPORT STAFF.)

Number of FTEs Not provided

18. For each of the following funding sources, please answer the following three questions as they relate to the specific biotechnology research project covered by this questionnaire. If this research project combined both biotechnology and conventional procedures, then report only the funds devoted to the biotechnology part of the project. Funds should be reported to the nearest dollar.

- a. In Column A, for each funding source, indicate how much money was spent before October 1, 1983 on this biotechnology research project.
- b. In Column B, for each funding source, indicate how much money was spent between October 1, 1983 and September 30, 1984 on this biotechnology research project.
- c. In Column C (to the best of your knowledge), for each funding source, indicate how much additional money is expected/needed to be spent on this biotechnology research project.

<u>FUNDING SOURCE(S)</u>	<u>COLUMN A</u>	<u>COLUMN B</u>	<u>COLUMN C</u>
	TOTAL FUNDS SPENT TO 9/30/83 ON THIS SPECIFIC BIOTECHNOLOGY RESEARCH PROJECT	FUNDS SPENT 10/1/83-9/30/84 ON THIS SPECIFIC BIOTECHNOLOGY RESEARCH PROJECT	TOTAL ADDITIONAL FUNDS EXPECTED/ NEEDED OVER THIS PROJECT'S LIFE
1. USDA competitive grants	\$ 5,527,000 (N=100)	\$ 3,424,900 (N=100)	\$ 6,495,800 (N=100)
2. USDA (all other)	\$	\$	\$
3. Other federal agencies	\$	\$	\$
4. State agencies	\$	\$	\$
5. Industry	\$	\$	\$
TOTALS	\$	\$	\$

(097709)

14. Is risk assessment, as defined in the introduction, a part of this research project?

1. Yes...CONTINUE TO QUESTION 15 N = 27

2. No....SKIP TO QUESTION 16

15. Is the risk assessment part of this research project expected to result in (CHECK ALL THAT APPLY AND EXPLAIN AS APPROPRIATE).....

1. an assessment of the risks associated with this experimentation? N = 6

2. new risk assessment methods or techniques? (PLEASE EXPLAIN AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.) N = 21

3. improvement of existing risk assessment methods or techniques? (PLEASE EXPLAIN AND LIMIT YOUR RESPONSE TO 50 WORDS OR LESS.) N = 0

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