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In-Depth Laboratory Review October 19-21, 1994

BYO

SOIL MICROBIAL SYSTEMS LABORATOR

USDA-ARS-NRI

Soil Quality Sustainable Agriculture Composted Waste Arbuscular Mycorrhizae Pesticide Metabolism Bioremediation

Biocontrol Management System

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AGENDA

October 19, 1994 Theater, Log Lodge, BARC-East

8:15	PRE REVIEW "BREAK"	GET ACQUAINTED SESSION	AND) SNACKS	
8:30	P. KEARNEY	INTRODUCTORY REMARKS			
8:40	P. MILLNER	LAB OVERVIEW AND ISSUES	6		
9:20		EXECUTIVE SESSION			
10:20		BREAK			
10:30		PATRICIA MILLNER		U.S.D.A., IVAL	
11:30		CHLOE RINGER		NCT 2 3 1998	
12:00		LUNCH	- E		

Received

Lincoln Room, Log Lodge, BARC-East

1:00	LAWRENCE SIKORA
2:00	DONALD KAUFMAN
3:00	BREAK
3:15	LAURA LENGNICK
4:00	REVIEW TEAM DISCUSSION

October 20, 1994 Lincoln Room Log Lodge, BARC-East

8:00	JEFFREY BUYER
9:00	WALTER MULBRY
10:00	BREAK
10:15	JEFFREY KARNS
11:15	SARA WRIGHT
12:15	LUNCH
1:15	TOUR FACILITIES
2:30	MEETING WITH SUPPORT STAFF
3:15	REVIEW TEAM DISCUSSION AND REPORT DRAFTING

October 21, 1994 Lincoln Room, Log Lodge, BARC-East

8:30	EXECUTIVE SESSION
10:30	END OF REVIEW



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Jeffrey S. Karns	41
Sara F. Wright	44

REVIEW TEAM

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Dr. William Brinton, Jr. Director, Woods End Research Laboratory, Inc. 1860 Rome Road Mr. Vernon, ME 04352 Phone: 207-293-2488

DEFINITION OF SOME TERMS, ABBREVIATIONS AND ACRONYMS USED IN ARS

<u>ARS</u>: Agricultural Research Service. An agency in the Science and Education branch of USDA. ARS has about 8,000 employees, including about 2,500 senior scientists. The Agency conducts research at 127 locations in the U.S. ARS is led by an Administrator and is divided geographically into eight Areas, which are led by Area Directors.

BA: The Beltsville Area includes the Beltsville Agricultural Research Center, the Beltsville Human Nutrition Research Center, the U.S. National Arboretum, and the Glenn Dale Plant Distribution Station. The Beltsville Area, at 6,600 acres, is the smallest Area geographically, but the largest in terms of personnel and budget. About 1,450 employees, including about 440 scientists, work in the BA.

<u>NPS</u>: National Program Staff. Members are called National Program Leaders and each is a subject matter specialist. NPS serves the Administrator of ARS in developing and coordinating research plans and strategies on a national basis. NPS sets National program-directions, establishes priorities, allocates resources, including this review, and acts as a clearing house for decision making. Considerable interaction between Area managers and NPS is required to fulfill our respective roles.

<u>INSTITUTES</u>: The Beltsville Agricultural Research Center is composed of four Institutes (see Appendix): the Plant Sciences Institute, the Livestock and Poultry Sciences Institute, the Natural Resources Institute, and the Product Quality and Development Institute.

<u>LABORATORIES</u>: Laboratories are units located in the Institutes. Laboratories are led, both scientifically and administratively, by Research Leaders. Typically, a Laboratory is comprised of 8-10 scientists, a scientific and clerical support staff and several temporary student and postdoctoral employees. The program and mission of a Laboratory of this size must obviously be limited. In reviewing a Laboratory, bear in mind that what appear to be discipline or program gaps are often filled by collaboration with other Laboratories in the BA or elsewhere.

<u>CRIS</u>: Current Research Information System. This is an electronic system for the filing and retrieval of information about individual agricultural research projects. In ARS, the terms "CRIS Work Unit" or the acronym "CRIS" are used <u>synonymously with "research project" or "project."</u> New projects are planned in coordination with NPS and are subjected to peer-review. The normal life of a project in ARS is 3 to 5 years.

SY: Scientist Year. This is the effort of a research scientist for 1 year. Fractional efforts (e.g., 0.5 SY) in a given project are possible when a scientist works in more than one project during the course of a fiscal year. The term is also used in ARS as a synonym for a research scientist [e.g., "I have six SYs (research scientists).in my Laboratory'].

<u>OTHER KINDS OF SCIENTIFIC PERSONNEL:</u> Research scientists are responsible for all phases of research. ARS also employs research associates ("postdocs"), support scientists (who have responsibility for some portion of a project), technicians, students, and in some operations nonresearch scientific personnel who perform work involving service to the public or to other government agencies.

AM: Administrative Management. This branch of ARS manages support activities, such as procurement, facilities, fiscal allocations and personnel operations at all levels in ARS.

NOTE: The organizational scheme described above is presented graphically on the following pages.



[*Each Area is led by an Area Director and an Associate Area Director]



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USDA Organizational Chart

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USDA, AGRICULTURAL RESEARCH SERVICE GELTSVILLE AREA

............ AREA DIRECTOR K. D. MURRELL ASSOCIATE AREA DIRECTOR G. C. MARTEN ----..... ______ BELTSVILLE HUMAN NUTRITION NATURAL RECOURCES PLANT SCIENCES INSTITUTE RESEARCH CENTER INSTITUTE DIRECTOR DIRECTOR J. B. St. John P. C. Kearney ------............. CARBOHYDRATE NUTRITION CLINATE STRESS J. G. Hallfrisch S. Britz ENERGY & PROTEIN NUTRITION ELECTRON HICROSCOPE V. P. Vergin LIPID MUTRITION ENVIRONMENTAL CHEMISTRY R. J. Wright NUTRIENT COMPOSITION HYDROLOGY G. R. Beecher W. J. Rawls (Acting) VITANIN & NINERAL NUTRITION REMOTE SENSING RESEARCH G. F. Hart -----SOIL MICROBIAL SYSTEMS P. D. Hillner SYSTEMS RESEARCH J. C. Ritchie (Acting) ------FACILITIES MANAGEMENT AND PRODUCT QUALITY AND OPERATIONS DIVISION DEVELOPMENT INSTITUTE -----...................... DIRECTOR DIRECTOR

D. D. Bills

F. Schuenk

QUALITY

SENSING

Y. R. Chen

NEAT SCIENCE RESEARCH

M. B. Sologon PESTICIDE RESEARCH

> STAFF D. D. Bills

A. E. Watada

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............... ASSOCIATE DIRECTOR J. J. Kenn BEE RESEARCH H. Shimanuki -----BIOCONTROL OF PLANT DISEASES R. D. Lumsden FRUIT R. F. Korcak INSECT BIOCONTROL J. L. Vaughn INSECT CHENICAL ECOLOGY 8. Leonhardt INSECT NEUROBIOLOGY & HORHONE J. A. Svoboda HOLECULAR PLANT PATHOLOGY R. E. Davis NATIONAL GERHPLASH RESOURCES A. K. Stoner **NEKATOLOGY** D. J. Chitwood PLANT HOLECULAR BIOLOGY A. K. Hattoo SOYBEAN AND ALFALFA RESEARCH D. L. Keister SYSTEMATIC BOTANY & MYCOLOGY EAHILY ECONONICS A. Y. Rossman RESEARCH GROUP SYSTEMATIC ENTOHOLOGY H. B. Stoetzel (Acting) HORTICULTURAL CROPS VEGETABLE S. L. Sinden WEED SCIENCE INSTRUMENTATION AND J. D. Anderson

----LIVESTOCK AND POULTRY SCIENCES INSTITUTE DIRECTOR T. J. Sexton ANIMAL INPROVEMENT PROGRAMS H. D. Norman BIOSYSTEMATIC PARASITOLOGY J. R. Lichtenfels GENE EVALUATION AND HAPPING C. E. Rexroad GERKPLASH & GAKETE PHYSIOLOGY L. A. Johnson RELNINTHIC DISEASES J. F. Urban-LIVESTOCK INSECTS D. K. Hayes HILK SECRETION & MASTITIS R. H. Hiller HONRUMINANT ANIMAL MUTRITION H. C. Steele PROTOZOAN DISEASES K. D. Ruff RESEARCH ANIMAL SERVICES K. Hummet RUHINANT NUTRITION T. S. Rumsey VETERINARY SERVICES 8. Stroud

U.S. NATIONAL

ARBORETUN

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T. E. Elias

ADHINISTRATIVE

C. Kosberger

EDUCATION UNIT

E. A. Neumann

GARDENS UNIT

S. G. March

GERHPLASH UNIT

E. J. Garvey

FLORAL & MURSERY

PLANTS RES. UNIT

R. H. Lawson

..............

FACILITIES NGR.

DIRECTOR

P. W. Noe

J. T. Judd

J. C. Smith

W. G. Horner

......

ADMINISTRATIVE BRANCH

C. Bowie (Acting)

BRANCH

R. D. Little

FARM OPERATIONS BRANCH

H. T. Badger

FACILITY ENGINEERING

J. Spence

DIRECTOR

ZOONOTIC DISEASES R. Faver



USDA-ARS SOIL MICROBIAL SYSTEMS LABORATORY

BACKGROUND

The Soil Microbial Systems Laboratory was established in 1984; it was previously known as the Biological Waste Management and Organic Resources Laboratory. The change in name was accompanied by a change in research direction away from waste utilization in agriculture to very basic soil microbiology - plant - rhizosphere investigations. In 1993, the Laboratory acquired staff, space, equipment, funds and projects from the former Pesticide Degradation Laboratory. The molecular biology experience of these new colleagues has been as welcome and productive addition.

PROBLEM

Public and private sectors demand that agricultural and other systems operate sustainably, i.e., so they meet the needs and aspirations of the present without compromising the ability of future generations to meet their own needs. Development of alternative agricultural practices that balance the needs to conserve soil and water quality, profitability, food safety and quality will address concerns about the impacts of traditional and modern farming practices on sustainability and negative environmental impacts. Such new approaches require more internalized nutrient cycling and fewer offfarm inputs to increase system efficiency. Soil quality conservation and improvement and use of organic matter will involve microbial processes. Improved agricultural system functioning will require a better understanding of the critical biological components, interactions, and nutrient transformations involved. How can crop/soil/nutrient plans be managed so that optimal benefits of microbial activities are realized? Which microbial processes are critical to the transition from conventional to alternative agricultural practices need to be used to ease this transition.

MISSION

The mission of the laboratory is to conduct research on soil microbiological processes that influence soil quality, especially the biological activity of soil. Emphasis is placed on the development and application of basic principles and technology of organic matter recycling, including composting, environmental, biochemical, and genetic factors that influence interactions of soil-plant microbes and microbiological processes critical in nutrient availability, agroecosystem sustainability, and biodegradation of complex chemical compounds. The resultant improvements in microbial cycling of plant nutrients which are derived from soil, soil organic matter, crop residues, and animal manures will enhance soil quality, conservation, and productivity by minimizing agricultural chemical inputs as well as losses from erosion, nutrient runoff/leaching, and pest damage.

RESEARCH ACCOMPLISHMENTS

SOIL QUALITY AND MICROBIAL DIVERSITY

An assay was developed for pseudobactin, the siderophore of plant growth-promoting *Pseudomonas putida* B10, using monoclonal antibodies. This is the most sensitive assay for any siderophore yet reported. A method was also developed to extract pseudobactin from soil quantitatively; this is the first report of such for siderophores from soil. These two methods were combined to measure pseudobactin in the rhizosphere of barley colonized with *P. putida* B10. This is the first directly measured concentration of a siderophore produced by an introduced organism in a natural ecosystem. For the first time monoclonal antibodies to phytosiderophores (iron chelators) secreted by graminaceous plants were produced.

A protein from the membrane that surrounds poly- β -hydroxybutyrate (PHB) granules was isolated. This protein is missing in the bacteroid of a Fix⁻ mutant of *Bradyrhizobium japonicum*. This research suggests for the first time that PHB formation is important in N₂ fixation conducted by Bradyrhizobia.

A new solid medium was developed for enumeration and isolation of soil and rhizosphere microorganisms without interference from the rapidly spreading bacterium *Bacillus mycoides* This medium was used successfully in soil/rhizosphere studies to suppress the growth of *B. mycoides* while enumerating and isolating other gram-positive bacteria.

Preliminary studies of the effects of the cropping systems on soil microbial community diversity which was determined for bacteria by fatty acid methyl ester analysis, with calculations of evenness and diversity, showed that total counts were not significantly different for the three cropping systems studied (Rodale Institute Research Center's Farming Systems Trial). The bacterial population isolated from the low-input animal system had the fewest genera and species, and the lowest diversity and evenness. The bacterial populations from the conventional and low-input legume systems had similar diversity and evenness. The fungal population from the animal manure system had lower diversity and evenness than the other two systems. These preliminary results suggest that conventional agricultural practices may maintain high indices of microbial diversity in the rhizosphere. The functional significance of this needs to be investigated.

Arbuscular-Mycorrhizae

A significant accomplishment was achieved when the site of reaction of a monoclonal antibody on vesicular-arbuscular mycorrhizal fungal hyphae was identified as a transient protein. All tests to date indicate that the protein is present on actively growing, young hyphae and absent on older, lysing, or melanized hyphae and after pot cultures have been allowed to dry at room temperature. Thus, the presence of the protein is an indicator of activity of hyphae. Because the probe for the protein is a monoclonal antibody, it is predicted that rapid, easily performed assays can be developed to quantify the protein in soils. Also, preliminary tests have indicated that the protein may be important in stabilization of soil aggregates which would be another breakthrough in defining elements that contribute to soil quality.

Farming Systems and Compost Utilization Trials

Relevant historical data from RIRC's Farming systems Trial (FST) was reviewed; determined that



new statistical approaches were needed to evaluate the data because of variability in soil and vield factors. A substantial library of aerial photographs of the RIRC area which covers the period from 1948-1981 at irregular intervals of 4-7 years, and at least annual coverage of the years 1981-1989 was compiled. Such photographs have been essential in developing more accurate soil maps of FST. Through computer digitization of soil reflectance, as recorded by aerial photography, a new, more accurate soil map of RIRC Farming Systems Trial (FST) was generated in cooperation with Dr. Eileen Perry. This map is currently used to accurately define and select sampling sites for on-going data collection. Research was initiated to collect data needed to characterize measurable variables such as soil reflectance, rooting depths, chlorophyll content, plant height, rock fragment content, soil moisture, bulk density and crop yield. These data will be used to develop a multivariate statistical procedure needed to analyze treatment effects in FST. These procedures and results are being used by agronomists, soil scientists, extension agents and farmers who are concerned with use, yield, and systems responses of sustainable agriculture practices. Procedures developed in this investigation are being used successfully to delineate the design of new, long-term experiments in such a way as to reduce variability due to soil chemical/physical characteristics.

Cooperative investigations were conducted by Dr. M. Wander, Ohio State University, (presently at University of Illinois, Urbana, Illinois) Dr, Bob Dudley, Eastern regional Research Center, and Dr. Kaufman, on metabolism of C¹³-labeled acetate, benzoate, and catechol in FST conventional and sustainable cropped soils. Treatment difference were apparent among treatments in C¹³O₂ evolution, which indicates a potentially different microbial utilization of these substrates by microbes in the different cropping systems. NMR analysis of the treated/incubated soils is being used to characterize the distribution of C¹³ products in soils.

A major collaborative long-term investigation was designed and initiated in 1992 by USDA-SMSL and Rodale Institute Research Center scientists which involves composting of various combinations of urban wastes (leaves, brush, newspapers, cafeteria wastes) and rural wastes (animal manures), and subsequent utilization of the composted wastes in rotational field trials which include both field and vegetable crops. The experiment is designed to last 15-20 years and active research components of the project include formation, maintenance, and analysis of the compost pile and process, utilization of the compost in field trials, effects of composts on soil quality, crop yield and quality, nutrient cycling, leaching, and other environmental parameters.

Biodegradation of several starch-based plastic materials (golf tees, votive candle cups, film, and pellets) were examined with microbial cultures in the laboratory and sequentially in compost piles and field soils. Although most materials were still readily recognizable by shape, substantial loss of composition did occur. Carbon-13 NMR spectra of the biodegradable materials showed significant losses in starch contents, but few changes in composition of the petroleum-based plasticizer in each item. Despite extensive degradation of the starch component, the votive candle cups, and pellets were still readily identifiable in field soils after 11 weeks of compositing and 52 weeks in cropped field soil.

Working collaboratively with Penn state and RIRC, SMSL scientists have participated in development of efficient methods to evaluate root architecture and dynamics in field and vegetable cropping systems and to obtain data on interaction of sustainable agricultural practices on root architecture and dynamics. Corn and bell pepper roots were excavated from several depths in field plots where different cropping practices (permanent bed, moldboard plowed, or minimum till time-fixed or variable traffic patterns) were used. Observed that all root forms were dynamic within each treatment over time.

Bioremediation and Pesticide Metabolism

A total management system for coumaphos-containing cattle-dipping vats used in APHIS-VS's Tick eradication Program was field tested at CFTRL in Mission, TX. Acidification of the vat contents prevented accumulation of a toxic metabolite, formed in untreated vats due to the action of anaerobic microbes, without using a biocide. The contents of the vat were then successfully biodegraded by a consortium of bacteria. This allows maximum use of the coumaphos in the vat accomplishing waste minimization, and allows for environmentally-sound disposal of vat contents.

Biodegradation studies were conducted using soils from cattle dipping waste pits that were contaminated with high levels of coumaphos. Results showed that the coumaphos in all of the soils could be rapidly biodegraded in soil slurries using indigenous soil microorganisms.

A cloning strategy based on DNA amplification to generate gene specific probes was used to isolate, sequence and over-express the parathion hydrolase gene adpB from a *Nocardia* strain in *E. coli*. This strategy was also used to isolate and characterize an s-triazine dechlorination gene (*trzA*) from the *Rhodococcus corallinus*. A collaborator has used the *trzA* gene to construct a recombinant bacterial strain capable of dealkylating and dechlorinating atrazine and simazine.

Several genes were cloned from bacterial strains that degrade simpler s-triazine compounds likely to be intermediates in the degradation of the major herbicides, atrazine, simazine, and cyanazine. These studies demonstrated the importance of transmissible DNA elements and DNA/DNA recombination in the evolution and spread of genes encoding the degradation of xenobiotic compounds. The cloned genes may be of use in future construction of bacterial strains that completely degrade triazine herbicides, which are typically relatively resistant to microbial degradation.

A special 2-year project of a headquarters supported post doc resulted in production of a monoclonal antibody which reacted with alachlor and diethyl analine, the simplest residue of this herbicide. The antibody is being used in a flow immunosensor by the Environmental Chemistry Laboratory to detect metabolites of alachlor in meat.

Sustainable Agriculture

Collected baseline measurements and uniformity trial data in preparation for designing a long-term field crop exeriment on alternative agricultural practices. Established a pilot field study of utilization of urban and rural wastes in sustainable agriculture. The study compares six composts, two manures, and five inorganic wastes to each other and to fertilizer.

Demonstrated that one third compost N and two thirds fertilizer N equaled 100% fertilizer N in the growth chamber. These data suggest that farmers could reduce fertilizer costs by using composts. Determined that addition of compost to soil did not stimulate the mineralization of soil organic matter. Developed a new microbial biomass determination based on rehydration of dried soils. Results were similar to the fumigation technique but used chemicals that are less hazardous than chloroform. Proposed an indicator of soil quality based on the use efficiency of N from organic and inorganic sources.

Alternatives to Methyl Bromide Soil Fumigation

New CRIS proposal was written and sent out for review.

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SMSL VISITING SCIENTISTS SINCE 1990

Suichi Mihashi, University of Tokyo

Vladimir Yakovchenko, Kiev State University, Ukraine

Paulette Royt, George Mason University

Sergei Chernikov

Vitaly Nebesny, Ukranian Academy of Science

Wolfgang Hordt, Univ. Hohenbeim, Stuttgart, Germany

Thierry Heulin, C.N.R.S., Nancy, France

Steve Bentivenga, University of West Virginia

Chang Lu, Peoples Republic of China

Keng-Yeang Lum, Malaysian Agricultural Research and Development Institute, Kuala Lumpor, Malaysia

COOPERATIVE RESEARCH AGREEMENTS

Soil and Agronomic Factors III Con-University of Maryland Agronomy Department (1270-12000-010-Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Rodale Institute Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Rodale Institute Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Rodale Institute Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Rodale Institute Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Rodale Institute Root Biology in Sustainable Agriculture - Specific Cooperative Agreement - Related From ARS Last year field work @ Rodale Pennsylvania State University (1270-12000-014-02S) compost use Penn State will continue composting work



SUMMARY SOIL-MICROBIAL SYSTEMS LABORATORY CRIS PROJECTS

CWU NO.	TITLE	NET TO CRIS	DURATION
1270-12000-013-00D	Role of Vesicular Arbuscular Mycorrhizae (VAM in Low Input Agricultural Systems	155,157	2/91 - 2/96
1270-12000-014-00D	Production and Use of Rural/Urban Waste Co-Compost: Microbial Processes	265,211	6/94 - 6/99
1270-12000-015-00D	Integrated Soil-Nutrient-Crop- Microbial-Pest-Waste Management Strategies for Sustainable Agriculture	296,095	8/94 - 8/99
1270-12000-016-00D	Soil Quality and Soil Ecology in Sustainable Agriculture	351,108	8/94 - 8/99
1270-12220-001-00D	Integrated Soil/Crop Biocontrol Management System: Sustainable Alternatives to Methyl Bromide	232,162	8/94 - 8/99
1270-12130-005-00D	Biochemistry and Molecular Biology of Pesticide Metabolism in Soil and Water	269,414	89 - 12/94
0500-00026-034-00D	Bioremediation of Contaminated Sites to Protect Water Quality	60,000	10/94 - 9/95

TOTAL

1,629,147

OLD CRIS PROJECTS

CWU NO.	TITLE	DURATION
1270-12000-009-00D	Analysis of Soil Microbiological Processes Which Affect Low-Input Sustainable Crop/Livestock System	3/89 - 2/94
1270-12000-010-00D	Fertilizer and Cultural Management Effects on Crop Nutrient Quality and Bioavailability	4/89 - 4/94
1270-12000-011-00D	Management of Beneficial Plant Microbe Soil Interactions for Low Input Sustainable Agriculture	4/89 - 4/94
1270-12000-012-00D	Effects of Residue Decomposition in Low Input Sustainable Agricultural Systems	5/89 - 5/94



DISTRIBUTION OF SCIENTIFIC PERSONNEL AMONG CRIS PROJECTS

Scientist	1270- 12000- 013-00D	1270- 12000- 014-00D	1270- 12000- 015-00D	1270- 12000- 016-00D	1270- 12220- 001-00D	1270- 12130- 005-00D	0500- 00026- 034-00D
Buyer		.20	.20	.50	.10		
Karns				.25		.75	
Kaufman		.65	.20	.15			
Lengnick			.70		.30		
Millner	.35	.10	.10	.10	.35		
Mulbry	.10			.10	.10	.70	
Ringer		.50			.50		
Sikora		.10	.50	.30		.10	
Wright	.55		.10	.05	.30		

SOIL MICROBIAL SYSTEMS LABORATORY

SCIENTIFIC PERSONNEL AND EXPERTISE

Dr. Jeffrey Buyer	Research Chemist	biochemistry, siderophore-metal interactions, microbial ecology
Dr. Jeffrey Karns	Research Microbiologist	molecular biology, bioremediation, biochemistry, microbial ecology
Dr. Donald Kaufman	Research Microbiologist	sustainable agriculture, composting, pesticide metabolism
Dr. Patricia Millner	Research Microbiologist & Research Leader	soil microbiology, composting, bioaerosols, immunology, mycology/mycorrhizae, molecular systematics
Dr. Walter Mulbry	Research Microbiologist	molecular biology, bioremediation, biochemistry
Dr. Låwrence Sikora	Research Microbiologist	immunology, compost, science, soil science, nutrient cycling, soil quality
Dr. Sara Wright	Research Microbiologist	immunology, mycorrhizae

SUMMARY OF SOIL-MICROBIAL SYSTEMS LABORATORY FINANCIAL RESOURCES

	FY '93	FY '94	FY '95
Net to Location	1,401,531	1,927,508	1,959,864
Indirect Research Cost	275,135	382,474	392,322
Adjustments			
Post Doc	40,690	50,650	9,550
Water Quality Korms Mulbry		60,000	60,000
BARD	43,651	47,261	
Net to MU	1,126,396	1,545,034	1,567,542
Salary	811,737	762,106	1,024,618
All-Other	311,380	595,919	604,530
Ratio of Salary to All-Other	2.60	1.27	1.69
Percent Salaries	58.05	37.97	50.69
Total dollar per SY	199,750	286,734	288,781
Discretionary Funds per SY	39,626	103,677	62,219
Percent Discretionary	19.84	36.16	21.55
SOIL-MICROBIAL SYSTEMS LABORATORY 1991 - 1994 PRODUCTIVITY SUMMARY

Scientist	Peer Rev 1st Author	iewed Pub. Co-Author	Other Publications	Abstract/Sci. Presentations
Buyer	3	2	4	2
Karns	2	7		23
Kaufman	1	3	3	12
Lengnick	2	1	1	3
Millner	4	3	2	15
Mulbry	5	1		10
Ringer	1			
Sikora	5	4	2	10
Wright	4	2		3
Hornick*	1	2	4	4

*Resigned 2/94

PEER REVIEWED PAPERS: 53 TOTAL PUBLICATIONS: 69

PUBLICATIONS PER SY PER YEAR

	PEER REVIEWED	ALL PUBLICATIONS
1991-1994	5.3	6.9

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SUPPORT STAFF

Ms. Dorothy Talmud	Secretary, Office Automation
Ms. Claire Reese	Office Automation Assistant
Ms. Nancy Enkiri	Microbiologist
Ms. Sara Reynolds	Microbiologist
Mr. Tim Prickett	Biological Laboratory Technician
Mr. Stanley Tesch	Biological Laboratory Technician
Dr. Cecil Tester	Chemist
Ms. Heidi Hartman	Cooperative Student - University of Maryland
Ms. Tracy Butler	Cooperative Student - University of Maryland
Mr. Pedro Del Valle	University of Maryland - Biological Lab. Technician
Dr. Laura Lengnick	Agronomist - Research Associate
Dr. Chloe Ringer	Plant Pathologist - Research Associate
Ms. Mary Jane Letaw	University of Maryland -Agricultural Technician
Ms. Leanne Teerlinck	RIRC- Research Assistant
Ms. Elizabeth Lyman	RIRC - Research Assistant

SAFETY AND HEALTH REPORT

All items identified by the Safety and Health Office have been corrected except for installation of new stairs and railing along the path leading to the Chemical Storage bunker, Building 318 B. This has been noted on every inspection since February 1990 and each time a work request was submitted to FMOD for action; none has yet been taken. The lab safety program is ongoing with each problem identified and corrective action taken by the Lab safety officer with cooperation from every lab member.

Biominillers - with Kame + MUlbry CO2 - Sikona, et.al.

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RESPONSE TO RECOMMENDATIONS FROM PREVIOUS REVIEW (1993)

Concern 1: It is recommended that all present work on the decomposition of inoculated corn residues be terminated when current experiments are completed.

Response: All work was completed and published; no new work undertaken.

Concern 2: it is recommended that Drs. Kaufman, Payne, Perry and other cooperators publish peer-reviewed manuscripts on the large amount of research conducted by ARS scientists at the Rodale Research Center.

Response: To date, 3 papers have been published on completed work; analysis of data has been hampered by the termination of temporary ARS employees, Drs. Payne and Perry, and resignation of the participating Research Director at RIRC, and change in assignments of other cooperators. Dr. Kaufman has been negotiating with ARS and RIRC staff involved in the work for an agreeable plan for publication. He has determined that the data will need specialized statistical handling, with modern georeferencing and mapping to account for spatial variability. Dr. Millner has recommended and Dr. Kaufman has welcomed the suggestion that Dr. Lengnick become involved in this project. It is the lab's intention to provide Dr. Lengnick with a full-time field assistant to manage the daily matters at the BARC Sustainable Agriculture site so that she will have time to undertake the statistical analysis of the RIRC/ARS data necessary for publication.

Concern 3: It is recommended that Dr. Laura Lengnick be asked to assume the coordinating role in SMSL for the sustainable project.

Response: Dr. Lengnick was hired as a Research Associate in 1993 and has coordinated the field crops research project of the BARC Sustainable Agriculture Program.

Concern 4: It is recommended that Dr. Millner continues to work with Dr. Kemper and Dr. Walker on establishing a Federal Center on Sustained Agricultural Use of Urban and Rural Wastes.

Response: In 1993, Dr. Millner conducted a special briefing session for the Beltsville Area Director and staff regarding the research program, proposed operational plan, and funding streams for the Center. NPS's requests for information and statement of proposed work were promptly supplied. In addition, Dr. Millner provided information to The Composting Council to assist them in establishing a working coalition of interested stakeholders. Thus far, no additional funding has been received to support a Center. Dr. Millner has recently submitted a multi-agency grant proposal to USEPA to conduct research on establishing sampling and analytical standards for compost in the US. This project will be the first one in which a research unit at BARC would take the major coordinating lead as envisioned in the plans for the Center.

Concern 5: It is recommended that Dr. Sikora continue research to more clearly define what is a measure of soil quality. He is making good progress on his nitrogen flux concepts.

Response: Dr. Sikora has continued and expanded his research on measuring soil quality through the help of Dr. Vladamir Yakovchenko, Visiting Scientist from University of Kiev, and an University of Maryland cooperative student from agronomy. In addition, the expiring CRIS project covering this research has been rewritten to include development of soil biological quality indicators.

CONTRIBUTIONS FROM SCIENTISTS

I. Patricia D. Millner, Microbiologist

II. CRIS Project:

CRIS Project 1270-12000-013D: Role of Vesicular Arbuscular Mycorrhizae (VAM) in Low Input Agricultural Systems

Objective: Determine relatedness of arbuscular mycorrhizal fungi (AMF) at the molecular level, and use this to develop molecular probes useful for identification of taxa that have colonized field samples of crop roots. Apply the technique to farming system field trials to determine changes in AMF root colonization and soil networking by plant growth effectiveness taxa in comparison with "weedy" non-beneficial taxa. Determine the relative effects of various alternative low-input treatments on AMF crop/soil colonization such that taxa which facilitate transition from conventional to alternative farming practices can be recognized and managed through cropping practices rather than inoculation.

Progress: Methods have been developed to cleanly produce spores in soilless condition and to reliably extract, amplify, and finally sequence rDNA from as few as one to ten spores of identified AMF isolates. Using areas of the variable regions of the internal transcribed spacers and the conserved 5.8s regions, it has been possible to confirm some species complexes and generic groupings relatedness. This adds support to the previously proposed concepts based on morphological, ontological, and fatty acid methyl ester patterns of our collaborators. It also provides the basic data needed to proceed directly to construction and development of the taxon specific probes.

Plans: Refine PCR primers to be selective for AMF DNA by generic and/or species groupings; adapt the extraction and PCR technique to colonized roots of corn. Construct taxon-specific oligonucleotide probes (TSOPs)for *Glomus mosseae, G. etunicatum, G. occultum, Gigaspora rosea, Gi. gigantea, Gi. albida, Acaulospora mellea, Entrophospora* (species to be determined), and other dominant AMF taxa present in the BARC long-term sustainable field crops research study site (as determined in the baseline species survey). Verify probe specificities with standard operating conditions. Use TSOPs to determine AMF colonization by crop and field location over time and relate to cropping practices, and changes in biotic and abiotic soil factors.

Old CRIS Project 1270-12000-009-003: Analysis of Soil Microbiological Processes Which Affect Low-Input Sustainable Crop/Livestock System

New CRIS Project 1270-12000-014-00D: Production and Use of Rural/Urban Waste Cocompost: Microbial Processes

Objective: Develop microbial consortia that can be used to reliably produce disease suppressive composts for control of certain soilborne plant diseases.

Progress: Worked with Dr. Ringer in planning preliminary growth chamber studies in which she has found 6 rural/urban waste co-composts suppress <u>Pythium</u>, which causes damping-off disease of cucumber seedlings.

Plans: These composts and others will be further tested (see p.24). The physical, chemical, and biological quality status of compost that is found suppressive will be characterized to increase our understanding of the substrate factors that favor development and expression of suppressiveness in compost. Effective microorganisms from these composts eventually will be used along with other biocontrol microbes to develop consortia of plant beneficial microorganisms for production of disease-suppressive compost.

New CRIS 12700-12222-001-00D Integrated Soil/Crop Biocontrol Management System: Sustainable Alternatives to Methyl Bromide

Old CRIS 1270-12000-010-00D Fertilizer and Cultural Management Effects on Crop Nutrient Quality and Bioavailability (Drs. Hornick and Chaney, previous members of SMSL staff worked on this CRIS exclusively)

Objectives: Develop an integrated group of practices for conserving and improving soil and crop health and productivity by combining the use of biocontrol agents, organic matter amendments, microbial competition, alternate cover crops, low toxicity/residual pesticides/fungicides, and low-cost field bed preparation techniques for high-value crops such as strawberries, asparagus, and tomatoes.

Progress and Plans: This project is designed to work in concert with and to move the results of other projects to the field by working toward the solution of a very urgent need. The project proposal is currently under review. Meanwhile, compost quality assessments and disease suppressive studies on CRIS 1270-12222-014-00D are proceeding so that we can move directly along. Collaborators who have good linkages with commercial enterprises that are willing to participate in field trials of promising alternatives to methyl bromide have been identified and are ready to help with field trials.

III. Cooperators:

ARS:

All staff members of Soil Microbial Systems Laboratory, BARC Dr. Jim Locke, Floral Crops Research Unit, National Arboretum, BARC Mr. T. Badger, Farm Manager, BARC Dr. Stan Nemec, Citrus & Subtropical Products Lab, Winter Haven, FL Dr. David Douds, Plant & Soil Biophysics Research, Eastern Regional Res. Ctr., Philadelphia, PA

Other:

Dr. Joseph Morton, Dept. of Soil and Plant Sciences, West Virginia University, Morgantown Dr. Raymond Weil, Department of Agronomy, University of Maryland, College Park, MD Dr. F.R. Gouin, Department of Horticulture, University of Maryland, College Park, MD Dr. John Haines, New York State Museum, Albany, NY Dr. John Walker, Office of Solid Waste, USEPA, Washington, D.C. The Composting Council, Alexandria, VA. Dr. T. Heulin, CNRS, Nancy, France

IV. Curriculum Vitae:

Education:

1966-1970 Univ. of Maryland; major, Microbiology; minor, Chemistry; B.S. 1970.
1970-1975 Univ. of Maryland; major, Botany (Mycology); M.S. 1975.
1981-1984 Univ. of Maryland; major, Environmental Science; minors, Microbiology, Biochemistry; Ph.D. 1984.

Professional Employment:

1969-1970	Biological Aid, Mycology Laboratory, Plant Protection Institute, USDA-ARS, Beltsville, MD.
1970-1978	Microbiologist, Mycology Laboratory, USDA, ARS, Beltsville, MD.
1977-1978	Microbiologist, Biological Waste Management and Soil Nitrogen Laboratory, Agricultural
	Environmental Quality Institute, USDA, ARS, Beltsville, MD.
1978-1984	Microbiologist, Biological Waste Management and Organic Resources Laboratory, USDA,
	ARS, Beltsville, MD.
1985-1991	Microbiologist, Soil-Microbial Systems Laboratory (formerly Biological Waste Management
	Laboratory), ARS, USDA, Beltsville, MD.
1991-	Research Leader, Soil-Microbial Systems Laboratory, ARS, USDA, Beltsville, MD.

Membership in Professional Societies:

Mycological Society of America American Society for Microbiology American Phytopathological Society American Society of Agronomists Soil Ecology Society Soil and Water Conservation Society

Offices and Committee Assignment Held in Professional Societies:

Member, Sustaining Membership Committee, 1980-1983, Mycological Society of America. Member, Mycorrhizae Committee, 1990-1993, American Phytopathological Society. Member, Advisory Committee, 1990-Present, International Collection of Vesicular and Arbuscular-Vesicular Mycorrhizal Fungi, West Virginia University, Morgantown (NSF funded culture collection)

Honors and Awards:

- 1967 Alpha Lambda Delta, Elected Member, Freshman Woman's Honorary
- 1969 Sigma Alpha Omicron, Elected Member, Microbiology Honorary
- 1969 Mortar Board Scholarship recipient
- 1970 B.S. Degree with honors
- 1977 USDA Superior Service Award, Team member of the Sewage Sludge Land Utilization Research Group "in recognition for outstanding team effort and response to an urgent national need for research information on safe and beneficial use of sewage sludge on agricultural land."
- 1984 Nominated to receive the Outstanding Dissertation Award from the University of Maryland (College Park). Regents decided to forgo all awards that year for fiscal reasons.
- 1991 Nominated for WISE (Women in Science Engineering) award
- 1992 ARS Kinney Post-Doctoral Research Associate for proposed work on "Microbial Enhancement of Farm/Urban Waste Compost" for FY-93.

Referred Publications Since 1991:

Millner, P. D. Characterization and use of vesicular-arbuscular mycorrhizae in agricultural production systems. Plant and Soil 32:335-342. 1991.

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Lewis, J. A., Lumsden, R. D., Millner, P. D. and Keinath, A. P. Suppression of damping-off of peas and cotton in the field with composted sewage sludge. Crop Protection 11:260-266. 1992.

Millner, P. D. and Kitt, D. G. The Beltsville method for soilless production of V-A mycorrhizal fungi. Mycorrhiza 2:9-15. 1992

Wright, S. F. and Millner, P. D. Dynamic processes of vesicular-arbuscular mycorrhizae: A mycorrhizosystem within the agroecosystem. pp. 29-59. IN: J.L. Hatfield, et al. (eds.) Soil Biology: Effects on Soil Quality. Advances in Soil Science. Lewis Publ., Boca Raton, FL. 1993

Syliva, D.M., D.O. Wilson, J. H. Graham, J.J. Maddox, P.D. Millner, J.B. Morton, H.D. Skipper, S. F. Wright and A.G. Jarstfed. Evaluation of vesicular-arbuscular mycorrhizae in diverse plants and soils. Soil Biol. Biochem. 25:705-713. 1993

Millner, P.D. and D. G. Kitt. Mycorrhizal colonization of corn co-inhabited by biopesticidalrecombinant Clavibacter xyli subsp. cynodontis. Microb. Releases 2:81-84. 1993.

Millner, P. D. et al. Bioaerosols Associated with Composting. J. Regulatory Toxicol. Pharmacol. (Accepted for Publication as a supplement). 69 pp.

Other:

Millner, P. D. "VA Mycorrhizae, Soil Fertility and Crop Production in Africa", World Bank, Proceedings of Workshop on Managing The Fertility of Soils in Africa, Washington, D.C. 1992,

Millner, P. D. "Inoculum Production and Use of VA Mycorrhize in Soils", Proceedings of OECD Workshop on Establishment of Microbial Inocula in Agriculture, Maui, Hawaii (accepted for publication in a section of J. Alternative Agriculture). 1994.

Millner, P.D. <u>Book Review</u>: "Mycorrhizal Fungi in Sustainable Agriculture". J. Alternative Agriculture 8(4): 1993.



I. Chloe E. Ringer, Postdoctoral Research Associate

II. CRIS Project:

Old CRIS Project 1270-12000-009-00D: Analysis of Soil Microbiological Processes Which Affect Low-Input Sustainable Crop/Livestock System

New CRIS Project 1270-12000-014-00D: Production and Use of Rural/Urban Waste Cocompost: Microbial Processes

Objective: Determine the potential usefulness of composts and compost extracts in controlling specific soilborne and foliar plant pathogens.

Progress: Conducted preliminary growth chamber studies on the usefulness of various rural/urban waste co-composts for suppression of <u>Pythium</u> damping-off disease of cucumber seedlings in soilless potting mixes composed of 1:1 peat:perlite and 10 or 30% concentrations of the following waste co-composts:

broiler carcasses/broiler manure/sawdust/straw broiler litter/leaves dairy manure/leaves/straw/newspaper horse manure/cow manure/straw/sawdust/clay broiler litter/food waste/biodegradable plastic/leaves leaves/lawn waste

All six composts were highly suppressive to <u>Pythium</u> damping-off of cucumber. Severe disease losses in sterile, inoculated controls provided evidence that the suppression was due to the presence of beneficial microorganisms in the composts. Further, plant growth in most nonsterile compost mixes was comparable to growth in a popular, commercial soilless mix. These results will strengthen interest in on-farm composting of rural/urban wastes and contribute to the future development of a marketable, value-added farm product, while reducing the use of peat and chemical inputs.

Plans: These composts and others will be further tested to evaluate their effectiveness in suppressing other soilborne plant pathogens, such as <u>Phytophthora</u> spp. and <u>Rhizoctonia solani</u>, on cucumber seedlings and other horticultural crops. Additionally, compost extracts will be evaluated for suppression of foliar plant pathogens. Effective microorganisms from these composts eventually will be used to develop consortia of plant beneficial microorganisms for the commercial production of reliably high-quality, disease-suppressive compost.

III. Cooperators:

Dr. K.P. Hebbar, Biocontrol of Plant Diseases Lab, ARS, Beltsville, MD

Dr. R.L. Chaney, Environmental Chemistry Lab, ARS, Beltsville, MD

Dr. L.E. Carr, Agricultural Extension Service, University of Maryland, Princess Anne, MD

Dr. F.R. Gouin, Department of Horticulture, University of Maryland, College Park, MD

Rodale Institute Research Center, Kutztown, PA

Mr. G. Leidig, Autrusa Compost Consulting, Blue Bell, PA

Mr. P. Boop, Briar Patch Organic Farms, Mifflinburg, PA

IV. Curriculum Vitae:

Education:

- 1975 B.S. Botany, University of Maryland, College Park, MD
- 1985 M.S. Plant Pathology, University of Maryland, College Park, MD
- 1992 Ph.D. Plant Pathology, University of Maryland, College Park, MD

Professional Employment:

1976-1981	Plant Pathologist, Biocontrol of Plant Diseases Lab, USDA-ARS, Beltsville, MD
1981-1985	Graduate Research Assistant, Department of Botany, University of Maryland, College Park, MD
1985-1986	Graduate Teaching Assistant, Department of Botany, University of Maryland, College Park, MD
1987-1992	Graduate Student, Department of Botany, University of Maryland, College Park, MD
4/93-9/93	Acting Director, Plant Diagnostic Lab, Department of Botany, University of Maryland, College Park, MD
1/94-	Postdoctoral Research Associate, Soil Microbial Systems Lab, USDA-ARS, Beltsville, MD

Membership in Professional Societies:

American Phytopathological Society

Publications:

Ringer, C.E., and Grybauskas, A.P. 1994. Infection cycle components and disease progress of gray leaf spot on field corn. Plant Disease (In press).

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Lengnick - Research plats (being?) devised by committee

I. Lawrence J. Sikora, Microbiologist

II. CRIS Project: Old CRIS Project 1270-12000-012-00D: Effects of residue decomposition in low-input sustainable agriculture

sustainable agriculture New CRIS Project 1270-12000-015-00D: Integrated Soil-Nutrient-crop-microbial-Pest-Waste Management Strategies for Sustainable Agriculture. Compost Use essut for

Objective: Determine the effects of compost-fertilizer combinations on the yield and nutrient * Und feldstocks content of crops.

Progress: Adding a mixture of compost and fertilizer equalling to the N requirement for wheat showed that compost N can substitute for one third of the inorganic N requirement. These data suggest that farmers can save on their fertilizer costs when using composts in this manner and benefit further from the additional organic matter in compost. Similar studies using fescue showed that a refuse-sewage sludge co-compost could substitute for one third of the inorganic N requirement but sewage sludge compost did not demonstrate the same benefit. The use of ammonium nitrate or urea as the fertilizer source did not effect these results. Addition of compost to soil did not increase the mineralization of soil organic matter.

Plans: Continue to evaluate mixtures or blends of composts with fertilizers to maximize the benefits of composts while minimizing the additions of non-nutrients such as heavy metals from composts made from urban wastes. Develop control strategies for composting that will maximize benefits and remove hindrances of composts and composting such as product instability and odor generation.

Old CRIS Project 1270-12000-009-00D: Analysis of Soil Microbiological Processed which Affect Low-Input Sustainable Crop/Livestock System

New CRIS Project 1270-12000-014-00D: Production and Use of Rural/Urban Waste Cocompost;

Objective: Determine the effects of low-input agricultural practices on soil microbial C and N components and transformations.

Progress: The Farming systems Trial at the Rodale Institute Research Center has been studied extensively to determine the effect of manures or legumes as compared to inorganic fertilizer as the source of nitrogen for crop growth. Decomposition studies indicated a higher level of respiration for both legume and manure systems. Other components that were higher in the manure or legume low-input systems were microbial biomass, N mineralization and N flux. Yield data for the 13 year study did not show yield responses (decreases or increases) to the low-input treatments possible due to the organic matter variation of greater than 100% across the field . Efforts to map the area by sampling and by aerial photography indicated that soil brightness measurements of aerial photos could predict soil organic matter content with reasonable accuracy. Normalizing the yield data for organic matter levels is being used to determine

treatment effects. A soil quality indicator based on nitrogen efficiency was proposed - flending compusts - pot tests - Sikora () with quarterly 3 d N (manput) synergion) on this.

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Plans: Establish the principles of co-composting using laboratory scale composters. Determine through field utilization trials the environmental safety and agronomic/soil quality benefits of composted rural/urban waste materials on crop lands. Continue pilot field project evaluating the effects of urban and rural composts on field crops. Test the effects and survival of microbial inoculant for composting to enhance the compost as a soil biofertilizer.

Old CRIS; 1270-12000-011-00D: Management of beneficial plant microbe soil interactions for low input sustainable agriculture.

NEW CRIS 1270-12000-016-00D: Soil Quality and Soil Ecology in Sustainable Agriculture.

Objective: Analyze *Bradyrhizobium japonicum*-soybean interaction to improve the N₂ fixation capacity.

Progress: A mutant of *Bradyrhizobium japonicum* which does not fix N₂ was found to have low levels of poly- β -hydroxybutyrate (PHB) in the bacteroid form. A monoclonal antibody was raised which reacts with a 14 kDa component that resides on the membrane that surrounds PHB and is missing in the mutant. Other scientists hypothesize that this component is a membrane stabilizer which separates the hydrophilic cytoplasm from the hydrophobic granule. Without PHB, N₂ fixation in *B. japonicum* may be reduced or eliminated especially during seed filling. This research demonstrates the possible importance of PHB in N₂ fixation in Bradyrhizobia.

Plans: Extensive sampling of the FST plots at Rodale allowed preliminary evaluation of soil factors which may be used to determine soil quality. Studies will be continued at Rodale and at two other long term sites in Maryland to test a biological indicators of soil quality such as respiration, biomass content and biomass turnover of nitrogen. Guidelines will be developed for uses of composts with fertilizer to reduce the total amount of costly external nutrient inputs. Determine the effect of composts on the microbial diversity of soils and rhizosphere.

II. Cooperators

<u>ARS:</u>

Other:

Dr. Laurie Drinkwater, Rodale Institute

Dr. Vladimir Yakovchenko, Kiev, Ukraine

Research Center, Kutztown, PA

Mr. Jerry Pearson, Aurelia Corp.

Dr. Dave Kuykendall,Beltsville, MD Dr. Ron Korcak, FL, Beltsville, MD Dr. Jeff Buyer, SMSL, Beltsville, MD Dr. Pat Millner, SMSL, Beltsville, MD Dr. Don Kaufman, SMSL, Beltsville, MD

IV. Curriculum Vitae:

Educational Background:

1962-1966 Wayne State University; B.S. Biology 1966
1969-1971 North Dakota State University; M.S. Bacteriology; Biochemistry minor 1971
1971-1973 University of Idaho; Ph.D. Bacteriology 1973

Research Experience:

1966-1969Research Assistant, Worcester Foundation for Experimental Biology, Shrewsbury, MA1969-1971Graduate Research Assistant in Bacteriology, North Dakota State University, Fargo, ND1971-1973Graduate Research Assistant in Bacteriology, University of Idaho, Moscow. ID



- 1973-1975 Post Doctoral Research Assoc., Soil Science Dept., University of Wisconsin, Madison, WI
- 1976-1978 Research Chemist, Biological Waste Management& Nitrogen Lab, USDA-ARS, Beltsville MD
- 1978-1994 Microbiologist, Soil-Microbial Systems Lab., USDA-ARS, Beltsville, MD

Professional Society Activities:

American Society of Agronomy, Membership Committee, 1984- present

American Society of Microbiology

Sigma Xi

- Soil Science Society of America representative to Planning Committee for the Third National Symposium on Individual and Small Community Sewage Treatment sponsored by American Society of Agricultural Engineering, held Dec. 14-15, 1981
- Served on committee that organized research plot field trips at 1983 Northeast regional meeting of American Society of Agronomy

Honors and Awards:

- USDA Superior Service Award, 1977 Team member of the Sewage Sludge Land Utilization Research Group, "In recognition for outstanding team effort and response to urgent national need for research information on safe and beneficial uses of sewage e sludge on agricultural land."
- USDA Certificate of Merit, 1990 Recognition in assuming responsibility for maintaining and further developing the monoclonal antibody technology in SMSL.
- USDA Certificate of Appreciation, 1990 Recognition of committee participation for 1st BARC Orientation and Poster Day.
- USDA Certificate of Appreciation, 1992 Service on 1992 BARC Poster Day planning Committee.
- USDA Certificate of Appreciation, 1994 Serve as Mentor in Partners in Education Program.

Referred Publications Since 1991:

Buyer, J. S., L. J. Sikora and M. G. Kratzke. 1991. Development of a detection system for ferric pseudobactin using monoclonal antibodies. Plant and Soil 130:243-247.

Mihashi, S, L. J. Sikora, J. S. Buyer, S. Fushiya and S. Mori. 1992 Development and characterization of a monoclonal antibody to phytosiderophres. Plant Cell Physiol. 33:151-156.

Buyer, J. S., M. G. Kratzke and L. J. Sikora. 1993. A method for detection of pseudobactin, the siderophore produced by a plant-growth-promoting Pseudomonas strain, in the barley rhizosphere. Appl. Environ. Microb. 59:677-681.

Sikora, L. J. 1993. Effect of recombinant endophyte containing *Bacillus thuringiensis* delta endotoxin on some rhizosphere populations. Microb. Releases 2:109-112.

Buyer, J. S., M. G. Kratzke and L. J. Sikora. 1994. Microbial siderophores and rhizosphere ecology. IN Biochemistry of Metal Micronutrient in the Rhizosphere. Ed. J. A. Mathey, D. E. Crowley and D. G. Luster. Lewis Publishers, Boca Raton. LA, pp. 67-80.

Sikora, L. J. and M. I. Azad. 1993. Effect of compost-fertilizer combinations on wheat yields. 1993 Comp. Sci. & Util. 1:93-96.

Sikora., L. J., L. D. Kuykendall, R. S. Dwivedi, E. M. Herman and N. K. Enkiri. 1994. Characterization of a 14 kDa component with low expression in a unique Nod⁺Fix⁻Bradyrhizobium japonicum. Microbiol. 140:000-000.

Sikora, L. J., V. Yakovchenko, and D. D. Kaufman. 1994. A proposed soil quality indicator. Proc. The 3rd Wye International Conf. on Sustainaable Agriculuture. Ed. H. Cook and H. Lee.

Sikora, L. J., V. Yakovchenko, and D. D. Kaufman. 1994. Comparison of the rehydration method for biomass determination to fumigation-incubation and substrate-induced respiration method. Soil Biol. Biochem. 26:000-000.

Bilgers, L. J., V. Yakovetenko, and O. D. Kautman, 1994. A proposed soil quality indicative, Pro-The 3rd Wys International Conf. on Sustaina bits Agriculture. Ed. H. Cook. and Physics.

Sikom, L. J., V. Yakavetimikit and D. D. Kautmin, 1994. Companian of the retry free of for biomass date minotion to turnigation-inscharen and substrate-induced respiration of mino-Soil Biot Biothem, 26:008-000.

I. Donald D. Kaufman, Microbiologist

II. CRIS Project:

Old CRIS Project 1270-12000-009-00D: 1270-12000-009-00D: Analysis of Soil Microbiological Processes Which Affect Low-Input Sustainable Crop/Livestock System

New CRIS Project 1270-12000-014-00D - Production and Use of Rural/Urban Waste Co-Compost: Microbial Processes.

Objective: (a) To develop improved methods for assessing, predicting, and monitoring compost quality based on changes in chemical, physical and microbiological properties; (b) to assess the effects of specific microbial inoculants during the composting process in facilitating development of a more consistently uniform compost product, and enhancing compost performance as a soil biofertilizer; and (c) to determine through field utilization trials the environmental safety and agronomic/soil quality benefits of composting rural/urban waste materials on croplands, or as a farm marketable product.

Progress: (a) Methods for assessing compost pH, bulk density, cation exchange capacity, NO₂, NH₄/NH₃ determinations, C:N ratio, O₂ and CO₂ respiration, water holding capacity, phytotoxicity, and weed seed population were examined on samples of several rural/urban waste mixtures throughout the 1992-1993 and 1993-1994 composting seasons. These methods, and others being developed are important for development of in-field assessments of compost quality for consistent production of high quality composts for specific agricultural production systems; (b) Experiments have been conducted with specific compost inoculants on specific substrates to assess their efficacy in degrading such components in composting materials. Development of more effective inoculants will facilitate more consistent production of high quality composts; (c) A long term compost utilization trial was initiated in collaboration with Rodale Institute Research Center Scientists, to assess the impact of rural/urban composts on environmental safety and agronomic benefits of these materials in rotational cropping systems. Cropping systems selected include both field and vegetable crops.

Plans: New and improved tests will be developed and applied to characterization of compost quality. Results from existing and new improved tests will be correlated with results from field tests where compost utilization occurs. By matching compost production with utilization it will facilitate development of a more consistent uniform compost product which when used in field will further enhance compost performance as a soil biofertilizer.

III. Cooperators:

<u>ARS:</u>

- Dr. John Doran, Soil and Water Conservation Research Lab., Lincoln, NE
- Dr. Robert Dudley, Core Research Applications, Western Regional Res. Ctr., Philadelphia, PA
- Dr. David Douds, Plant & Soil Biophysics Research, Eastern Regional Res. Ctr., Philadelphia, PA
- Dr. Jeffrey Buyer, Soil-Microbial Systems Lab., BA, Beltsville, MD
- Dr. Lawrence Sikora, Soil-Microbial Systems Lab., BA, Beltsville, MD
- Dr. Vladimir Yakovchenko, Soil-Microbial Systems Lab., BA, Beltsville, MD
- Dr. Marion Simpson, Soil-Microbial Systems Lab., BA, Beltsville, MD
- Dr. Chloe Ringer, Soil-Microbial Systems Lab., BA, Beltsville, MD
- Dr. Patricia Millner, Soil-Microbial Systems Lab., BA, Beltsville, MD

- Dr. Richard Zobel, U.S. Plant, Soil and Nutrition Research Lab., Ithaca, N.Y.
- Dr. Jim Power, Soil and Water Conservation Research Lab., Lincoln, NE
- Dr. Sam Smith, Water Quality Research Lab., Durant, OK
- Dr. Doral Kemper, National Program Staff, USDA, Beltsville, MD
- Dr. Ben Coffman, Weed Science Lab., BA, Beltsville, MD

Other:

- Dr. Eileen Perry, Batelle N.W., Richland, Washington
- Dr. Michelle Wander, Dept. Agronomy, Univ. Illinois, Urbana, IL
- Dr. Laurie Drinkwater, Rodale Institute Research Center, Kutztown, PA
- Dr. Carolyn Reider, Rodale Institute Research Center, Kutztown, PA
- Dr. Peggy Wagoner, Rodale Institute Research Center, Kutztown, PA
- Dr. Steve Peters, Rodale Institute Research Center, Kutztown, PA
- Dr. Kim Kroll, Rodale Institute Research Center, Kutztown, PA

IV. Curriculum Vitae:

Education:

1951-52	Kent State Univ. Kent, OH; Biology Department (Biology, Chemistry, B.Sc.)
1955-58	Kent State Univ., Kent, OH; Biology Department (Microbiology); M.Sc.
1958-62	Ohio State Univ., Columbus, OH; Plant Pathology; Ph.D. 1962.

Professional Employment:

1952-1955	Laboratory Assistant, Kent State Univ., Kent, OH
1955-1956	Graduate Assistant, Kent State Univ., Kent, OH
1957-1958	Graduate Assistant, Kent State Univ., Kent, OH
1958-1962	Research Assistant, Ohio State Univ., Columbus, OH, and Ohio Agricultural Experiment
	Station, Wooster, OH
1962-1963	Research Microbiologist, Pesticide Degradation Laboratory, USDA, ARS, Beltsville, MD
1963-1965	Research Plant Pathologist, Pesticide Degradation Laboratory, USDA, ARS, Beltsville, MD
1965-1970	Research Microbiologist, Pesticide Investigations - Behavior in soils, Crops Research
	Division, USDA, ARS, Beltsville, MD
1967-1968	Fulbright-Hays Lecturer in Soil Microbiology, Plant Pathology and Environmental
	Microbiology, Khon Kaen Univ., Khon Kaen, Thailand (leave of absence)
1970-1977	Research Soil Microbiologist, Pesticide Degradation Laboratory, USDA, ARS, Beltsville, MD
1978-1984	Research Soil Microbiologist, Pesticide Degradation Laboratory, USDA, ARS, Beltsville, MD
1984-1991	Research Leader, Soil-Microbial Systems Laboratory, USDA, ARS, Beltsville, MD
1991-	Senior Scientist, ARS, Soil-Microbial Systems Laboratory, Rodale Institute Research
	Center, Kutztown, PA

Membership in Professional Societies:

Soil Ecological Society of America	American Chemical Society: Pesticide Chemistry Division
American Society of Microbiology	Sigma Xi
International Society of Plant Pathology	American Chemical Society: Pesticide Chemistry Division

Offices and Committee Assignments held in Professional Societies:

Member of Plant Science Seminar Committee, Plant Industry Station, USDA, Beltsville, MD, 1965-67 Chairman, Pesticide Degradation Laboratory Seminar Committee, 1972-74 Chairman, Pesticide Degradation Laboratory, Manuscript Review Committee, 1972-75 Chairman, Agricultural Environmental Quality Institute Seminar Committee, 1973-74 Member, Biological Control Committee, American Phytopathological Society, 1975-78



Member, Committee on Chemical Control, International Society for Plant Pathology, 1975-present Founding and Elected Board Member, Society of Environmental Toxicology & Chemistry (SETAC), 1978-83 Member, Publications Committee of SETAC; Member, Editorial Board of Environmental Toxicology and Chemistry, SETAC Journal; and Founding Editor, SETAC News, 1980-82

Honors and Awards

Gamma Sigma Delta (Nat. Hon. Soc. Agric.), 1960

Sigma Xi, Member, 1961

Fulbright-Hays Lectureship in Soil Microbiology and Plant Pathology, Khonkaen University, Khon Kaen, Thialand, 1967-68

- American Chemical Society, nominated for and awarded full membership under Bylaw I, Section 3(a)(7) for "significant achievement in chemistry", 1977
- Distinguished Alumni Award, The Ohio State University, "for recognition of his significant contributions to soil microbiology", 1980

Certificate of Merit: for "Outstanding coordination of ARS scientists with researchers and their collaborators at Rodale Institute Research Center on Sustainable Agriculture projects. \$1000, December 1992.

Publications Since 1991:

Reichel, H., H. D. Sisler and D. D. Kaufman. Inducers, substrates, and inhibitors of a propanoldegrading amidase of <u>Fusarium oxysporum</u>. Pesticide Biochem. and Physiol. 39:240-250. 1991.

Perry, E. M., Kaufman, D. D., Hart, G. F., and Payne, B. Spatial analysis of remotely-sensed and groundbased data to explain location effects in crop yields. 1993 Proceedings ASPRS/ACSM Annual Convention: Agricultural Remote Sensing Applications. Vol II., New Orleans, LA. pp. 110-114. 1993.

Young, A. L., S. A. Tolin, J. N. Rutger, D. D. Kaufman and M. B. Steinbock. 1994. Plant Biotechnology in China: Trip Report, Sept. 16-30, 1993. Prepared for: Research and Scientific Exchange Division, Office of International Cooperation and Development, USDA, Washington, D.C. 34 pp.

Parr, J. F., S. B. Hornick, and D. D. Kaufman. 1994. Use of microbial inoculants and organic fertilizers in agricultural production. Proc. Intl. Conf. on Use of Microbial and Organic Fertilizers in Agriculture. June 13-14, Suweon, Korea. In Press.

Kaufman, D. D., R. L. Dudley, C. R. Reider, L. M. Teerlinck, and B. W. Lyman. 1994. Biodegradation of several starch-based plastic materials in microbial cultures, compost pile, and field soil. Journal of Biodegradation. Approved for publication August 5, 1994.

Wander, M. M., D. S. Hedrick, D. Kaufman, S. J. Traina, B. R. Stinner, S. Kehermeyer and D. C. White. 1994. The functional significance of the microbial biomass and biologically-active soil organic matter in organic and conventionally managed soils. Plant and Soil, Special Issue on Soil Biodiversity (In Press).

Wander, M. M., D. S. Hedrick, D. Kaufman, S. J. Traina, B. R. Stinner, S. Kehermeyer and D. C. White. 1994. The functional significance of the microbial biomass and biologically-active soil organic matter in organic and conventionally managed soils. Soil Ecological Society Special Issue on Soil Biodiversity (In Press).

I. Laura Lengnick, Research Agronomist

II. CRIS Project:

Old CRIS Project 1270-12000-012-00D: Effect of Residue Decomposition in Low Input Sustainable Agricultural Systems

New CRIS Project 1270-12000-015-00D: Integrated Soil-Nutrient-Crop-Microbial-Pest-Waste Management Strategies in Sustainable Agriculture

Objective: The field crops research project will study the short- and long-term effects of sustainable and conventional production practices on biological, environmental, and economic aspects of field crop production in the Middle Atlantic states. Experimental plots are scheduled to be established in the spring of 1995 following a two year period of site characterization and experimental design.

Progress: Based on the geostatistical analysis of an initial soil survey conducted at the start of the project in May 1993, an intensive soil survey of the site on a 25 m square grid (300 points) was planned and completed this past spring. The first two of three planned uniformity crops (corn/winter wheat/soybean) were unsuccessful as a result of severe climatic conditions in the summer of 1993 and late winter of 1994. No-till corn was planted in May 1994 and conditions during the growing season have been very favorable for corn production. Data on crop growth and development have been collected on a 25 m square grid (300 points) throughout the growing season and biomass and grain yield data will be collected on this grid at crop maturity. Other data have also been acquired during the characterization period: color infrared aerial photography of the crop canopy during the 1993 and 1994 growing seasons; a digital elevation model of the site; and, black and white aerial photography of previous land use on the site during the period of 1930 to 1960. Development of a geographic information systems (GIS) data base to support comprehensive geostatistical analyses of spatial variation in soil characteristics and crop productivity is underway. All photography has been scanned into the GIS data base. Soils point data have been entered into the GIS data base and geostatistical analyses are currently being conducted on these data. Planning of the specific experimental treatments got underway with the meeting of a Farmer/Extension planning group in August 1994. This group proposed 9 potential farming systems to be considered as treatments for the project.

Plans: Additional soil physical and microbiological characterization will be completed by soil series for N mineralization, soil moisture relations, hydraulic conductivity, infiltration, aggregation, particulate organic matter, total microbial biomass C and N, nematode diversity and populations, and microbial biomass diversity (major groups). Additional uniformity crop data will be collected on a no-till hairy vetch cover crop that will be planted following corn harvest. All data collected at the site will be compiled in an ARC/INFO GIS data base and analysis of spatial characteristics will be used to aid in the design of homogeneous blocks across the 16 ha site. A BARC research planning group will meet in November 1994 to refine the systems proposed by the Farmer/ Extension planning group. Experimental treatments will be finalized in January 1995, following final review by the Farmer/Extension planning group. In early 1995, a field manager will be hired to manage the final phase of site preparation and all field operations once the experiment is underway.

III. Cooperators:

<u> ARS:</u>

Mr. G. Hart and Mr. W. Dulaney, Remote Sensing Research Laboratory Dr. J. Teasdale, Weed Science Laboratory Dr. D. Fravel and Dr. R. Lumsden, Biocontrol of Plant Diseases Laboratory Dr. Z. Handoo, Nematology Laboratory Mr. R. Hoover, Farm Branch-West

<u>Other</u>:

Dr. R. Weil, University of Maryland, College Park, MD Dr. R. Forney, Remington Farms Project, Chestertown, MD

IV. Curriculum Vitae:

Education:

1992	Ph.D. Agronomy, The Pennsylvania State University, University Park
1985	M.S. Soil Science, North Carolina State University, Raleigh
1981	B.S. Agronomy, University of Maryland, College Park

Professional Employment:

1993 -	Project Coordinator, Sustainable Agriculture-Field Crops, USDA-ARS, Beltsville, MD
1992	Congressional Science Fellow, American Society of Agronomy, Washington, DC
1988 -1991	Graduate Research Asst., Dept. of Agronomy, Pennsylvania State Univ., University Pk., PA
1987	Statistical Analyst, Dept. of Forestry, Pennsylvania State University, University Park, PA
1985 -1987	Agricultural Research Technician I, Crop Science Dept., NC State University, Raleigh, NC
1985	Graduate Teaching Assistant, Soil Science Dept., NC State University, Raleigh, NC
1982 -1984	Graduate Research Assistant, Soil Science Dept., NC State University, Raleigh, NC

Professional Activities:

Contributor, Agronomy News, American Society of Agronomy, Madison, WI, 1992.

Review Panel Member. National Academy of Sciences, 1992.

Graduate Representative to Faculty, Dept of Agronomy, Pennsylvania State Univ, University Park 1989-90 Contributing Editor, Stewardship News. Carolina Farm Stewardship Assoc., Carrboro, NC 1984 to 1987. Board Member, Carolina Farm Stewardship Association. 1985, 1986.

NC State Univ Repr, Coordinating Committee,1st Ann. No. Carolina Alternative Farming Field Days, 1985. Coach. NC State University Soil Judging Team. 1983.

Refereed Publications Since 1991:

Lengnick, L.L. and R.H. Fox. Simulation by NCSWAP of seasonal nitrogen dynamics in corn: I. Soil Nitrate. Agronomy Journal 86 (1): 167-175, 1994.

Lengnick, L.L. and R.H. Fox. Simulation by NCSWAP of seasonal nitrogen dynamics in corn: II. Corn growth and yield. Agronomy Journal 86 (1): 176-181, 1994.

Jabro, J.D., J.M. Jemison, Jr., L.L. Lengnick, R.H. Fox and D.D. Fritton. Field validation and comparison of LEACHM and NCSWAP models for predicting nitrate leaching. Trans American Soc Agri Eng 36(6): 1651-1657, 1993.
I. Jeffrey S. Buyer, Research Chemist

II. CRIS Project:

Old CRIS Project 1270-12000-011-00D: MANAGEMENT OF BENEFICIAL PLANT MICROBE SOIL INTERACTIONS FOR LOW INPUT SUSTAINABLE AGRICULTURE

New CRIS Project 1270-12000-016-00D: Soil Quality and Soil Ecology in Sustainable Agriculture

Objective: Determine the occurrence of microbial and plant siderophores in the rhizosphere and the role of regulating iron in the rhizosphere as a management tool for controlling harmful microbial populations.

Progress: We have developed immunoassays to pseudobactin, the siderophore produced by plant growth promoting *Pseudomonas* B10, and to the plant siderophore mugineic acid, and measured pseudobactin in the rhizosphere of barley inoculated with B10. We have developed monoclonal antibodies to chrysobactin, the siderophore produced by plant pathogenic *Erwinia chrysanthemi*, and are currently using these antibodies to develop a quantitative assay.

Plans: In order to study the impact of siderophores on microbial populations we are planning to study the effect of inoculated B10 on microbial diversity. Diversity will be measured by fatty acid methyl ester (FAME) analysis of isolates. Community-level diversity will be analyzed by phospholipid analysis or DNA analysis of soil extracts. Isolates will be screened for their ability to use pseudobactin and identified by FAME analysis. This experiment will allow us to determine if inoculation with a biocontrol organism affects microbial diversity and if any impact on diversity is due to siderophore production.

Objective: Evaluate the impact of soil microbial diversity on soil quality and sustainable agriculture.

Progress: Rhizosphere bacteria, actinomycetes, and fungi were enumerated and isolated from the Farming Systems Trial, Rodale Institute Research Center. Isolates are being identified by fatty acid methyl ester analysis. Preliminary results from 1993 indicate that diversity of culturable bacteria and fungi is lowest in the rhizosphere of plants treated with animal manure as the only nitrogen input.

Plans: Isolates will be examined for particular biological functions important in sustainable agriculture, such as cellulose decomposition, phosphate solubilization, and organic nitrogen mineralization. Community structure will be determined by phospholipid analysis, DNA analysis, and carbon source utilization patterns. A similar study will be carried out at the Sustainable Agriculture Field Site at Beltsville in order to follow diversity during the transition from conventional to low-input methods.

Objective: Determine relationships among selected VAM fungal isolates.

Progress: A method for fatty acid analysis of spores, at 100 times the sensitivity of previous methods, was developed.

Plans: Selected VAM fungal isolates will be grown and spores collected for fatty acid analysis. Numerical taxonomy of fatty acid composition of spores will be used to study VAM taxonomy. The great sensitivity of our method may allow us to analyze single spores of certain genera, thus allowing us to study clonal diversity.

III. Cooperators:

<u>ARS:</u>

Dr. L.J. Sikora, Soil Microbial Systems Laboratory, Beltsville, MD Dr. D.R. Kaufman, Soil Microbial Systems Laboratory, Beltsville, MD Dr. J.S. Karns, Soil Microbial Systems Laboratory, Beltsville, MD Dr. P.D. Millner, Soil Microbial Systems Laboratory, Beltsville, MD Dr. S.J. Wright, Soil Microbial Systems Laboratory, Beltsville, MD

Other:

Dr. M.J. Miller, University of Notre Dame, Notre Dame, IN Dr. Y. Chen, Hebrew University of Jerusalem, Rehovot, Israel Dr. Y. Hadar, Hebrew University of Jerusalem, Rehovot, Israel Mr. Wolfgang Hordt, Universitat Hohenheim, Stuttgart, Germany Dr. T. Heulin, CNRS, Nancy, France

IV. Curriculum Vitae:

Education and Experience:

1973 to 1977	State University of New York, Stony Brook, NY; Biochemistry Department, B.S. 1977
1977-1978	Research Assistant, New York Ocean Sciences Laboratory, Montauk, N.Y.
1978-1980	Technical Specialist, Marine Sciences Research Center, S.U.N.Y. at Stony Brook, N.Y.
1980-1985	University of California, San Diego, CA; Chemistry Department, Ph.D. 1985. Title of thesis:
	Siderophores of Plant Growth Promoting and Plant Deleterious Pseudomonads
1985-1987	Visiting scholar, Laboratory of J. Neilands, Department of Biochemistry, U C Berkeley, CA
1987-1990	Research Chemist (Postdoctoral), Soil-Microbial Systems Laboratory, USDA, ARS, .
1990-	Research Chemist, Soil Microbial Systems Laboratory, USDA, ARS, Beltsville, MD.

Professional Societies

American Association for the Advancement of Science American Chemical Society American Society for Microbiology

Invited Seminars

"Iron and Ecology", University of Maryland, College Park, 1991. "Siderophores and Rhizosphere Ecology", University of Delaware, Newark, 1991. "Microbial Siderophores and Rhizosphere Ecology", George Mason University, 1992. "Microbial Siderophores and Rhizosphere Ecology", American Chemical Society National Meeting, 1992. "Rhizosphere Siderophores, Microbial Ecology and Plant Uptake of Iron", Amer Soc Agron Meeting, 1993. "Siderophore Production by Pseudomonas B10 in the Rhizosphere", Amer Soc Microbiology 1994.

Honors and Awards

Individual National Research Service Award, 1985-87.

Peer Reviewed Publications since 1991

Buyer, J. S., de Lorenzo, V., and J. B. Neilands (1991) Production of the siderophore aerobactin by a halophilic pseudomonad. Appl. Environ. Microbiol. 57:2246-2250.

Buyer, J. S., Sikora, L. J., and M. G. Kratzke (1991) Development of a detection system for ferric pseudobactin using monoclonal antibodies. Plant and Soil 130:243-247. (republished after peer review from invited book chapter)

Mihashi, S., Sikora, L. J., Buyer, J. S., Fushiya, S., and S. Mori (1992) Development and characterization of a monoclonal antibody to phytosiderophores. Plant Cell Physiol. 33:151-156.

Buyer, J. S., Kratzke, M. G., and L. J. Sikora (1993) A method for detection of pseudobactin, the siderophore produced by a plant growth *Pseudomonas*, in the barley rhizosphere. Appl. Environ. Microbiol. 59:677-681.

Persmark, M., Pittman, P., Buyer, J. S., Schwyn, B., Gill, P. R., and J. B. Neilands (1993) Isolation and structure of Rhizobactin 1021, a siderophore from the alfalfa symbiont *Rhizobium meliloti* 1021. J. Am. Chem. Soc. 115:3950

Invited Book Chapters

Buyer, J. S., Sikora, L. J., and M. G. Kratzke (1991) Development of a detection system for ferric pseudobactin using monoclonal antibodies. IN Y. Chen and Y. Hadar (eds), Iron Nutrition and Interactions in Plants, Kluwer Academic Publishers, The Netherlands, pp. 283-287.

Buyer, J. S. and L. J. Sikora (1991) Rhizosphere interactions and siderophores. IN D. L. Keister and P. B. Cregan (eds), The rhizosphere and plant growth, Kluwer Academic Publishers, The Netherlands, pp. 263-269.

Buyer, J. S., Kratzke, M. G., and L. J. Sikora (1994) Microbial siderophores and rhizosphere ecology. IN D. Crowley, D. Luster, and J. Manthey (eds), The biochemistry of metal micronutrients in the rhizosphere, Lewis Publishers, Chelsea, Michigan, pp. 67-80.

<u>Reviews</u>

Loper, J. E. and J. S. Buyer (1991) Siderophores in microbial interactions on plant surfaces. Molecular Plant-Microbe Interactions 4:5-13.

I. Walter W. Mulbry, Microbiologist

II. CRIS Project:

1270-12130-005-00D: Biochemistry and Molecular Biology of Pesticide Metabolism in Soil and Water

Objective: To isolate and characterize microorganisms that metabolize agricultural chemicals of environmental concern and to determine the biochemical and genetic basis of pesticide metabolism in microorganisms; to isolate and characterize pesticide degradation genes; to develop microbially based waste disposal technologies that protect the environment and groundwater, and; to determine the effects of environmental factors on rates of pesticide metabolism to develop predictive models of *in situ* metabolism.

Progress: A novel cloning strategy was developed to isolate biodegradation genes from Grampositive bacteria using DNA amplification to generate gene specific probes. This strategy has broad applicability for isolating previously "unclonable" genes from diverse organisms. This strategy was used to clone, sequence and over-express the new parathion hydrolase gene adpB from a *Nocardia* strain in *E. coli*. More recently, this strategy was used to isolate and characterize an s-triazine dechlorination gene (*trzA*) from the organism *Rhodococcus corallinus*. A collaborating Canadian scientist has used this gene to construct a recombinant bacterial strain that is capable of dealkylating and dechlorinating atrazine and simazine.

DNA fragments from the cloned organophosphate degradation gene adpB have been used to characterize spontaneous mutants of *Nocardia* that have lost the ability to produce the *adp*B hydrolase. The goal of this research is to gain insight into the general mechanisms by which environmental organisms quickly change their metabolic characteristics.

Plans: Use the *s*-triazine degradation gene *trzA* as a gene probe to isolate other s-triazine degradation genes. Introduce this gene into other organisms having N-dealkylation activities (such as *Streptomyces* sp PS1/5) in order to construct microorganisms capable of constituitively degrading *s*-triazines in contaminated soils. Use the organophosphate degradation gene *adp*B to monitor the occurrence and spread of this gene in the microbial community associated with organophosphate contaminated soils. Further characterize and manipulate the genetic elements responsible for the loss of the *adp*B gene in *Nocardia* to construct bacterial suicide vectors.

0500-00026-034-00D: Bioremediation of Contaminated Sites to Protect Water Quality

Objective: To develop safe, effective and economical methods to eliminate potential point sources of ground and surface water contamination resulting from sites that have been contaminated by agrochemicals. More specifically, to refine the use of specific microbial isolates and consortia to bioremediate soils and waters from pesticide contaminated sites.

Progress: Biodegradation studies were conducted using soils from eight cattle dipping waste pits that were contaminated with high concentrations of the organophosphate insecticide coumaphos. The results showed that the coumaphos in all of the soils could be biodegraded in soil slurries using...indigenous soil microorganisms.

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Plans: Determine coumaphos degradation rates in moist buffered soils and evaluate whether this strategy would be practical for the remediation of contaminated soils. Determine the extent and rate of coumaphos degradation in dip waste applied to trickling gravel filters. Evaluate different filter matrices with regard to effectiveness and cost. Determine optimal operating parameters and scale-up factors necessary for demonstration scale experiments using trickling gravel filters.

III. Cooperators:

<u>ARS</u>:

Mr. E.H. Ahrens, Cattle Fever Tick Research Lab, Mission, TX.

Dr. R.B. Davey, Cattle Fever Tick Research Lab, Mission, TX.

Dr. Daniel Shelton, Environmental Chemistry Lab, Beltsville, MD

Dr. Larry Sikora, Soil Microbial Systems Lab, Beltsville, MD

Dr. Patricia Milner, Soil Microbial Systems Lab, Beltsville, MD

<u>Other</u>:

Dr. Ram Behki, Agriculture Canada, Ottawa, Canada Dr. B.M. Pogell, Center for Agricultural Biotechnology, Univ. of MD, College Park, MD

IV. CURRICULUM VITAE

Education

- 1975-79 Duke University; Major, Zoology; B.S. 1979.
- 1980-82 University of Utah; Major, Biology.
- 1984-87 University of Maryland; Marine, Environmental, and Estuarine SciencesProgram; Ph.D. 1988.

Professional Employment

- 1989- Microbiologist; USDA/ARS Agricultural Research Center, Natural Resources Institute, Pesticide Degradation Laboratory, Beltsville, MD. Promoted to GS-13 8/92. Transferred to Soil Microbial Systems Laboratory 10/93.
- 1988-89 Research Associate, Dept. of Biology, University of Maryland Baltimore County.
- 1987-88 Research Affiliate, GS-9; USDA/ARS Agricultural Research Center, Natural Resources Institute, Pesticide Degradation Laboratory, Beltsville, MD.

Honors and Awards

- 1992 Appointed to the Interational Union of Pure and Applied Chemistry (IUPAC) Task Force on Scientific Aspects of the Destruction of Chemical Warfare Agents.
- 1990 Research grant from Ciba Geigy Corporation for project "Isolation and characterization of <u>s</u>-triazine dechlorination genes from <u>Rhodococcus corallinus</u>."
- 1987 Research grant from Maryland Center for Agricultural Biotechnology for project "Genetic Analysis of Parathion Hydrolase Genes."

Membership in Professional Societies

American Chemical Society American Society for Microbiology Air and Waste Management Association Union of Concerned Scientists ,

Refereed Publications since 1991:

Lovett, P. S., Ambulos, N. P., Mulbry, W. W., Noguchi, N., and Rogers, E. J. UGA can be decoded as tryptophan at low efficiency in *Bacillus subtilis*. J. Bacteriol. 173:1810-1812. 1991.

Mulbry, W. W. and Kearney, P. C. Degradation of pesticides by micro-organisms and the potential for genetic manipulation. Crop Protection <u>10</u>:334-346. 1991.

Mulbry, W. W. and Eaton, R. W. Purification and characterization of the N-methylcarbamate hydrolase from *Pseudomonas* strain CRL-OK. App. Environ. Microbiol. <u>57</u>:3679-3682. 1991.

Mulbry, W. W. The aryldialkylphosphatase-encoding gene *adp*B from *Nocardia* sp. strain B-1: cloning, sequencing and expression in *Escherichia coli*. Gene 121:149-153. 1992.

Mulbry, W. W. Purification and characterization of an inducible s-triazine hydrolase from *Rhodococcus corallinus* NRRL B-15444R. App. Environ. Microbiol. 59:3533-3538. 1993.

Mulbry, W. W., Del Valle, P., Karns, J. S., and Shelton, D. R. Biodegradation of the organophosphate insecticide coumaphos in contaminated soils. Proceedings of the 1994 Annual Meeting of the Air and Waste Management Assocation. In press.

Muldoon, M.T., Nelson, J.O. Pesticide waste treatment monitoring of s-triazines uising immunoassy. J. Agric. Food Chem. 42:1686-1692. 1994.**

Muldoon, M.T., Nelson, J. O. Evlauation of the effects of selected agricultural materials on an ELISA for s-triazines. Food Agric. Immunol. 1994. (In press).**

Muldoon, M.T., Nelson, J.O. Immunochemical approach for pesticide waste treatment monitoring of s-triazines. ACS Sym. Ser. 1994 (In press).**

** Dr. M. T. Muldoon complete his doctoral research in Dr. Mulbry's laboratory in 1993-94.

I. Jeffrey S. Karns, Microbiologist

II. CRIS Project:

1270-12130-005-00D: Biochemistry and Molecular Biology of Pesticide Metabolism in Soil and Water

Objective: To isolate and characterize microorganisms that metabolize agricultural chemicals of environmental concern and to determine the biochemical and genetic basis of pesticide metabolism in microorganisms; to isolate and characterize pesticide degradation genes; to develop microbially based waste disposal technologies that protect the environment and groundwater, and; to determine the effects of environmental factors on rates of pesticide metabolism to develop predictive models of *in situ* metabolism.

Progress: The nucleotide sequence of a unique repeated element associated with s-triazine degradation genes in Pseudomonas was determined. The sequence revealed that the element has the structural characteristics of a bacterial insertion element including 27 base-pair inverted repeats at the ends of the element. Comparison with sequences in DNA databanks revealed that this element shared some degree of homology with known insertion elements but represents a heretofore unreported element. The genes encoding the enzymes for the degradation of the striazine compounds ammelide and cyanuric acid were shown to be plasmid encoded in a strain of Klebsiella pneumoniae that used these compounds as a nitrogen source. Cyanuric amidohydrolase, the enzyme responsible for cleavage of the s-triazine ring, was purified to homogeneity and characterized. Barbituric acid was found to be a potent competitive inhibitor if this enzyme reaction. A total-vat management system for the preservation and disposal of coumaphos-containing cattle-dips generated by the APHIS-Veterinary Services tick eradication program in Texas was field tested. It was shown that acidifying the vats with triple superphosphate fertilizer prevented formation of toxic potasan allowing maximum use of the coumaphos suspension in the vat. Neutralization of the vat contents and introduction of air allowed extensive biodegradation of the coumaphos in the spent cattle-dip. A screening study was conducted which showed that the organism Streptomyces sp. PS1/5 was able to metabolize a wide spectrum of herbicides in broth culture.

Plans: Continue to work with ARS scientists at the Cattle Fever Tick Research Lab in Mission, TX on monitoring the performance of the total vat management system in several working cattledipping vats. Aid them in research into chemical alternatives to coumaphos. To clone the gene encoding the enzyme biuret amidohydrolase (the product of enzymatic ring-cleavage of *s*triazines) and to characterize the enzyme.

0500-00026-034-00D: Bioremediation of Contaminated Sites to Protect Water Quality

Objective: To develop safe, effective and economical methods to eliminate potential point sources of ground and surface water contamination resulting from sites that have been contaminated by agrochemicals. More specifically, to refine the use of specific microbial isolates and consortia to bioremediate soils and waters from pesticide contaminated sites.

Progress: Collected soils from several coumaphos contaminated waste pits along the border between Texas and Mexico. Initiated studies on the biodegradation of atrazine in soil by *Streptomyces* sp. PS1/5.

Plans: Develop methods for the use of microorganisms of the genus *Streptomyces* for the bioremediation of pesticide contaminated soils through solid-state fermentation. Optimize parameters for the degradation of atrazine then look at metabolism of other pesticides. Characterize the products formed from pesticides by *Streptomyces* and isolate microorganisms to further degrade those products. Continue to support Dr. Mulbry's research into methods for the practical biodegradation of coumaphos in contaminated soils.

III. Cooperators:

<u>ARS</u>:

Mr. E.H. Ahrens, Cattle Fever Tick Research Lab, Mission, TX.

Dr. R.B. Davey, Cattle Fever Tick Research Lab, Mission, TX.

Dr. Cathleen Hapeman, Environmental Chemistry Lab, Beltsville, MD

Dr. Daniel Shelton, Environmental Chemistry Lab, Beltsville, MD

Dr. Larry Sikora, Soil Microbial Systems Lab, Beltsville, MD

Dr. Jeffrey Buyer, Soil Microbial Systems Lab, Beltsville, MD

Other:

Dr. B.M. Pogell, Center for Agricultural Biotechnology, Univ. of Maryland, College Park, MD Dr. Alan Sexstone, Dept. of Soil Microbiology, West Virginia University, Morgantown, WV

IV. CURRICULUM VITAE

Education:

1975 B.S. Medical Technology, The Pennsylvania State University, University Park, PA1981 Ph.D. Microbiology, Virginia Commonwealth University, Richmond, VA

Professional Employment:

- 1981-1983 Post-Doctoral Fellow, Dept. of Microbiology and Immunology, University of Illinois at Chicago, Health Sciences Center, Chicago, IL.
- 1983-1984 Postdoctoral Research Associate, Dept. of Entomology, Univ. of Maryland, College Park
- 1984-1993 Microbiologist, Pesticide Degradation Lab, USDA, ARS, Beltsville, MD.

1993- Microbiologist, Soil Microbial Systems Lab, USDA, ARS, Beltsville, MD.

Membership in Professional Societies:

American Society for Microbiology (ASM) American Chemical Society (ACS) American Society for the Advancement of Science (AAAS)

Editorial Board Appointments:

Member of editorial board of Applied and Environmental Microbiology, 1988-1994

Refereed Publications since 1991:

Karns, J.S. and P. H. Tomasek. Carbofuran hydrolase- Purification and properties. J. Ag. Food Chem. <u>39</u>:1004-1008. 1991.

Eaton, R.W. and J.S. Karns. Cloning and Analysis of <u>s</u>-Triazine Catabolic Genes from <u>Pseudomonas</u> NRRLB-12227. J. Bacteriol. <u>173</u>:1215-1222. 1991.

Eaton, R. W. and J. S. Karns. Cloning and Comparison of the DNA Encoding Ammelide Aminohydrolase and Cyanuric Acid Amidohydrolase from Three <u>s</u>-Triazine-Degrading Bacterial Strains. J. Bacteriol. <u>173</u>:1363-1366. 1991.

Smith, J. M., G. F. Payne, J. A. Lumpkin, and J. S. Karns. Enzyme Based Strategy for Toxic Waste Treatment and Waste Minimization. Biotechnology and Bioengineering <u>39</u>:741-752. 1992.

Karns, J.S. Biotechnology in Bioremediation of Pesticide Contaminated Sites: Past, Present, and Future. <u>In</u>: <u>Pesticide Waste Management</u>. Bourke, et al., (Eds.). ACS Symposium Series 510, Chapter 12. 1992.

Shelton, D.R., J.S. Karns and C.J. Hapeman-Somich. Biological methods for the disposal of cattle-dip waste. <u>In</u>: <u>Pesticide Waste Management</u>. Bourke, et al., (Eds.). ACS Symposium Series 510, Chapter 12. 1992.

Davey, R.B., E.H. Ahrens, J.E. George, and J.S. Karns. Efficacy of freshly mixed coumaphos suspensions adjusted to various pH levels for treatment of cattle infested with *Boophilus annulatus* (Say) (Acari: Ixodidae). Manuscript accepted for publication in *Preventative Veterinary Medicine* (8/8/94)

McClung, G., W. A. Dick, and J. S. Karns. Degradation of EPTC by Isolated soil microorganisms. Manuscript accepted for publication in Jnl. Agric. Food Chem. (Sent to Journal)

Mulbry, W. W., Del Valle, P., Karns, J. S., and Shelton, D. R. Biodegradation of the organophosphate insecticide coumaphos in contaminated soils. Proceedings of the 1994 Annual Meeting of the Air and Waste Management Assocation. In press.

Additional work completed by cooperators working on this project as part of M.T. Muldoon's dissertation research while this program was still affliated with Pesticide Degradation Lab.

Muldoon, M.T., Fries, G.F., Nelson, J.O. Evaluation of ELISA for the multianalyte analysis of <u>s</u>-triazines in pesticide waste and rinsate. J. Agric. Food Chem. 41:322-328. 1993.

Muldoon, M.T., Huang, R.N. Hapeman, C.J., Fries, G.F., Ma, M., Nelson, J.O. Hapten synthesis and immunoassay development for the analysis of chlorodiamino-<u>s</u>-triazine in treated pesticide waste and rinsate. J. Agric. Food Chem. 42:747-755. 1994.

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I. Sara F. Wright, Soil Scientist

II. CRIS Project:

1270-12000-013-00D: Role of Vesicular Arbuscular Mycorrhizae (VAM) in Low Input Agricultural Systems

Objective: Determine the effect of soil and crop management practices in model sustainable agriculture systems on the activity of indigenous mycorrhizal fungi. Utilize fatty acid profiles to identify fungal species contributing to the hyphal network.

Progress: An immunological test for activity of VAM fungal hyphae has been developed. A monoclonal antibody (MAb) was developed against a glycoprotein present on actively growing, young hyphae but not on older, melanized, or lysing hyphae. This is the first potentially useful tool to assess activity of VAM fungal hyphae, the most important VAM fungal contribution to an agroecosystem.

Fatty acid profiles for identification of VAM fungal isolates are being tested by Jeff Buyer. This method of identification is still in the preliminary stage of method development. We are currently supplying Jeff with single and multiple spore samples of different isolates collected from successive generations of pot cultures.

Plans: The nature of the protein will be further defined and an assay will be developed to utilize the MAb to quantify the protein in field soils. The assay will be used to quantitatively assess activity of VAM fungi in model sustainable agriculture systems at Beltsville at sites chosen for coordination with other soil tests.

Fatty acid profiles or conventional morphology will be used for identification of spores from trap cultures of different treatments in the sustainable agriculture plots at Beltsville.

Old CRIS Project 1270-12000-012-00D: Effects of Residue Decomposition in Low Input Sustainable Agricultural Systems

New CRIS Project 1270-12000-015-00D: Integrated Soil-Nutrient-Crop-Microbial-Pest-Waste Management Strategies for Sustainable Agriculture

Objective: Define the effects of isolated inputs to a sustainable system on VAM fungi as measured by activity of hyphae.

Progress: The recently discovered antibody to measure activity of VAM fungal hyphae is being used to develop an enzyme-linked immunosorbent assay (ELISA) to quantify the targeted protein under field conditions.

Plans: Assay activity of VAM hyphae under controlled conditions in greenhouse conditions using field soils to predict changes due to crops, fertilizer nutrients, and composed waste additions.

Old CRIS Project 1270-12000-011-00D: Management of Beneficial Plant Microbe Soil Interactions for Low Input Sustainable Agriculture

New CRIS Project 1270-12000-010-00D: Soil Quality and Soil Ecology in Sustainable Agriculture

Objective: To determine the role of VAM fungi in formation of water stable soil aggregates.

Progress: Preliminary tests have indicated that the protein found on active hyphae of VAM fungi is on the surface of water stable aggregates. This is the first indication that these fungi contribute a definable proteinaceous compound to aggregates, although there has been speculation on that VAM fungi contribute compounds to aggregates.

Plans: Cooperative work with Mike Miller at Argonne National Labs will be initiated to further define the presence of the immunologically reactive protein detected on water stable aggregates. Time-course studies and comparisons among genera and species of mycorrhizae for their contribution of the protein to soil aggregates will be made.

Old CRIS Project 1270-12000-010-00D: Fertilizer and Cultural Management Effects on Crop Nutrient Quality and Bioavailability

New CRIS Project 1270-12220-001-00D: Integrated Soil/Crop Biocontrol Management System: Sustainable Alternatives to Methyl Bromide

Objective: To determine the usefulness of VAM fungi in biocontrol of pathogens on selected crops currently produced after methyl bromide fumigation.

Progress: Project Outline is undergoing review.

Plans: Assess the contribution of VAM fungal isolates to biocontrol of pathogens of strawberries under greenhouse conditions using pasteurized soils. Test superior isolates under field conditions.

III. Cooperators:

<u>ARS:</u>

Dr. D. Douds, Eastern Regional Research Center, Philadelphia, PA Dr. J. S. Buyer, Soil Microbial Systems Lab., Beltsville, MD Dr. S. J. Lehotay, Environmental Chemistry Lab., Beltsville, MD

<u>Other:</u>

Dr. Frank Dazzo, Michigan State University, East Lansing, MI Dr. R. M. Miller, Argonne National Laboratory, Argonne, IL Dr. J. B. Morton, West Virginia University, Morgantown, WV Members of the Mycorrhizae section of the S-226 Regional Project

IV. Curriculum Vitae:

Education:

B.S. Medical Technology, University of Texas, Austin, TX
Ph.D. Soil Science, Texas A&M University, College Station, TX

Professional Employment:

1981-1982 Assistant Professor, West Virginia University, Morgantown, WV
 1982-1984 Research Scientist, Cooperative Agreement with West Virginia University,

- Appalachian Soil & Water Conservation Research Lab., Beckley, WV
- 1982-1991 Research Scientist, Appalachian Soil & Water Research Lab., Beckley, WV

1991-Present Research Scientist, Soil Microbial Systems Lab., Beltsville, MD

Membership in Professional Societies:

American Society for Microbiology Soil Science Society of America American Society of Agronomy

Offices and Committee Assignment Held in Professional Societies:

Soil Science Society of America, Program Session Chairperson, 1983 American Society of Agronomy, Fellows Selection Committee, 1985-1986 West Virginia Association of Professional Soil Scientists, President-elect and President, 1985-1987 American Society for Microbiology, Councilor for the Allegheny Branch, 1987-1988 American Society for Microbiology, Allegheny Branch President-elect and President, 1988-1990 American Society for Microbiology, Allegheny Branch Newsletter editor, 1990 Southern Regional Project (S-226) Program chairperson, 1990 International Collection of Vesicular and Arbuscular-Vesicular Mycorrhizal Fungi, Advisory Committee, 1990-Present

Honors and Awards:

USDA Certificate of Merit, 1989 West Virginia Celebrate Women Award for outstanding achievement in science, 1990

Refereed Publications Since 1991:

Wright, S. F., and S. K. Zeto. 1991. Effects of pH and Al³⁺ on survival of *Rhizobium leguminosarum* bv. *trifolii* in a simple solution and on nodulation of red clover in acid soils, pp. 603-609, <u>In</u> R. J. Wright, V. C. Baligar, and R. P. Murrman (eds.) Plant-Soil Interactions at Low pH. Kluwer Academic Publishers, Boston.

Wright, S. F., 1992. Immunological techniques for detection, identification, and enumeration of microorganisms in the environment, pp. 45-63. <u>In</u> M. A. Levin, R. J. Seidler, M. Rogul (eds.) Microbial Ecology Principles, Methods and Applications. McGraw-Hill, Inc., New York.

Wright, S. F. and P. D. Millner. 1993. Dynamic processes of vesicular-arbuscular mycorrhizae: A mycorrhizosystem within the agroecosystem, pp. 29-59. In J. L. Hatfield and B. A. Stewart (eds) Advances in Soil Science, Soil Biology: Effects on Soil Quality. Lewis Publishers, Boca Raton, FL.

Sylvia, D. M., D. O. Wilson, J. H. Graham, J. J. Maddox, P. Millner, J. B. Morton, H. D. Skipper, S. F. Wright and A G. Jarstfer. 1993. Evaluation of vesicular-arbuscular mycorrhizal fungi in diverse plants and soils. Soil Biology & Biochemistry 25:705-713.

Olsen, P., S. Wright, M. Collins, and W. Rice. 1994. Patterns of reactivity between a panel of monoclonal antibodies and forage *Rhizobium* strains. Applied and Environmental Microbiology 60:654-661.

Wright, S. F. 1994. Serology and conjugation of antibodies, pp. 593-645. <u>In</u> R. W. Weaver, et al. (eds.) Methods of Soil Analysis, Part 2. Microbiological and Biochemical Properties. Soil Science Society of America, Inc., Madison, WI.



