



**ASSESSMENT AND CONTROL
OF
PATHOGENS
IN
MUNICIPAL WASTEWATER
USED FOR IRRIGATION
TO
PROTECT CROPS AND GROUNDWATER**

Norma L. Duran, Microbiologist

PROJECT SUMMARY

Population growth and water shortages will increase the need to use treated wastewater effluent for irrigation, particularly in areas where fresh water resources are limited. However, there are serious concerns about the transmission of pathogens and toxic chemicals from municipal and animal wastewater to agricultural land and crops and thus to human food and to groundwater. An increase in foodborne disease in the U.S. has been attributed in part to the transmission of pathogens in the water used for irrigation of edible crops. Furthermore, there is limited knowledge on the long-term effects of irrigation with sewage effluent on soil and underlying groundwater. Thus, the aim of this research project is to assess the microbiological safety of waste water irrigation of food crops and potential environmental hazards in order to protect the public health and our future groundwater resources. Molecular biology techniques will be used to evaluate pathogen survival, regrowth, and transport in vegetated and non-vegetated soil columns, water distribution systems, and field sites with a long history of wastewater application for crop irrigation. Studies will determine the movement of pathogens through the soil column as well as the factors affecting their survival and transport. This could lead to the development of management strategies that would minimize the introduction of pathogens into the environment and thus reduce the risk to human health.

OBJECTIVES

1. Determine the fate and transport of pathogens present in treated sewage using vegetated (grass and alfalfa) and non-vegetated soil columns irrigated at various efficiencies or flooded to simulate artificial groundwater recharge conditions with chlorinated secondary sewage effluent. The columns will also be used to determine the fate of organic compounds, such as pharmaceuticals and pharmaceutically active chemicals and disinfection by products under a companion project under National Program 201, Water Quality and Management (Wastewater Irrigation and Groundwater Recharge).
2. Determine if wastewater irrigation has an effect on groundwater quality by analyzing upper groundwater samples below agricultural fields, urban irrigated areas (golf courses, parks, landscaping), and/or groundwater recharge areas with a long history of municipal wastewater application for emerging microbial pathogens including but not limited to *Escherichia coli* O157:H7, *Salmonella*, and *Campylobacter*. The samples will also be analyzed for pharmaceuticals and other chemicals under a companion project.
3. Determine if bacterial pathogens present in treated sewage can regrow in conveyance systems used to transport wastewater to fields for irrigation of fresh fruits and vegetables and conduct laboratory studies using a model system to determine the physical and chemical factors that promote/inhibit pathogen regrowth so that cost effective prevention strategies can be developed.

NEED FOR RESEARCH

Description of the problem to be solved

Increasing populations, finite water resources and increasingly stringent treatment requirements for discharge of sewage effluent into surface water is increasing the need for water reuse practices in the United States. However, due to recent foodborne outbreaks, public concern about the potential human health risks and environmental consequences of water reuse in agriculture is increasing. Thus, research is needed to increase our current knowledge on the long-term effects of wastewater irrigation on food, soil and underlying groundwater. In addition, the potential for pathogen regrowth in conveyance systems used to transport treated wastewater over long distances to the irrigated areas also deserves attention. Furthermore, proper assessment of water reuse practices will require microbial detection methods that are fast, sensitive and specific for pathogens of concern. Addressing these research needs will help assess the environmental and public health risks associated with wastewater irrigation so that future problems of food, soil and groundwater contamination can be anticipated or avoided.

Relevance to ARS National Program Action Plan

The research directly addresses national and global problems dealing with safety of food produced in fields that have been irrigated with treated sewage effluent or with effluent contaminated water. This project falls under National Program 108, Food Safety, Microbial Pathogens Component. The reduction of microbial pathogens in food products also relates to reducing environmental contamination from animal (and human) waste. This project is related to objective 1.6.1.1 "Identify sources and reservoirs of pathogens relative to on-farm and environmental situations" by determining the fate of pathogens in wastewater applied as irrigation to crops.

Potential benefits

Benefits from attaining the objectives include safe use of sewage effluents for irrigation from the standpoint of food safety and groundwater protection. Water reuse will be more common and the practices will be safer for public health.

Anticipated products of the research

Anticipated products of the research include (1) improved techniques of sewage treatment and system management for safe and sustainable water reuse with minimum adverse health effects and in environmentally acceptable ways, and (2) new guidelines for irrigation with wastewater to protect groundwater and surface water quality and for control measures of pathogen regrowth in water distribution systems.

Customers of the research and their involvement

Customers of the research include the public, farmers and farm workers, water planners and managers, government regulators, consulting engineers, water districts and municipalities, wastewater treatment plant operators and water managers.

SCIENTIFIC BACKGROUND: Refer to 2002 Annual Report

APPROACH AND PROCEDURES: Refer to 2002 Annual Report

PHYSICAL AND HUMAN RESOURCES: Refer to 2002 Annual Report

MILESTONES AND OUTCOMES

By the end of FY2002, the initial screening of pathogens in sewage and column effluents will be completed and should determine the presence of specific pathogens of highest concern to groundwater contamination. By the end of FY2003, we expect results regarding the fate and transport of pathogens from field studies as well as the completion of pathogen regrowth assessment in distribution systems. Studies on the effects of irrigation and groundwater recharge with sewage effluent will continue until dynamic equilibrium or end conditions are reached. If adverse effects are observed, procedures for mitigating these effects will be developed and tested on the columns by FY2004 (Table 5).

Table 5. Milestones and outcomes

Research Study-Component	Months of Study			
	11	22	33	44
Pathogen Transport/Column Studies	Operation and management for irrigation and groundwater recharge procedures, development of sampling and DNA extraction protocols completed	Operation continued, establish PCR procedures for selected pathogens, initial screening of pathogens going into and out of the columns completed	Operation continued, evaluation of fate and transport of pathogens completed	Final reports and manuscript prepared, develop recommendations and future studies
Pathogen Transport/Field Studies	Site characterization and sample collection completed	Optimization of DNA extraction and analysis protocols completed	Detail sampling to valuate fate of selected pathogen(s), analysis by PCR completed	Interpretation of results, final reports and manuscript prepared, develop recommendations for future studies
Pathogen Regrowth/Laboratory and Field Studies	Field site characterization, operation and management of annular reactor completed	Operation and sampling of annular reactor continued, sampling at different points in the water distribution completed	Molecular analysis of samples completed	Interpretation of results, final reports and manuscript prepared, develop recommendations for the control of pathogen regrowth

PROGRESS:

SOIL COLUMNS

Laboratory studies have been accomplished using soil column in collaboration with the USGS to identify the organic wastewater contaminants including pharmaceuticals and pathogens that can persist under groundwater recharge conditions. The results from this study have led to an additional project involving the testing of microbial resistance to the antibiotics, which were identified to persist in the soil column studies during recharge. This project is in collaboration with the USGS and the ARS Food and Feed Safety Research Unit. The information obtained from this study will help evaluate the degree by which water reuse practices will influence the transmission of antimicrobial resistance

determinants to groundwater when wastewater is used for irrigation and/or groundwater recharge.

FIELD STUDIES

Field studies are in progress to study impacts of land application of wastewater on groundwater quality. A project was initiated in collaboration with the USGS to study the spatial and temporal distributions of wastewater contaminants in the surface water and in the underlying groundwater at an effluent-dependent stream, the Santa Cruz River, Tucson, AZ. This study will help to better understand the processes affecting bacterial concentration in effluent-dependent streams as well as address potential effects of incidental ground-water recharge in the Santa Cruz River on groundwater quality. Other field sites are currently being evaluated for further fieldwork in conjunction with the USGS.

PATHOGEN REGROWTH

Field sites are being evaluated to study a real-time distribution system where effluent is used for turf irrigation and also to compare the efficiency of different disinfection processes (UV and chlorine disinfection) on preventing regrowth. Results from this research project will greatly add to our understanding of the microbiological safety of water reuse practices as well as their potential environmental hazards and will ultimately help protect public health and our future groundwater resources.

Describe the major accomplishments over the life of the project, including their predicted or actual impact.

There is a need to understand the environmental fate of microorganisms and the potential for bacterial regrowth in reclaimed water used for crop irrigation so that future problems of food contamination via wastewater irrigation can be prevented. A laboratory study was conducted at the U.S. Water Conservation Laboratory to assess the survival and regrowth potential of bacteria present in tertiary-treated effluent used for crop irrigation and surface-water discharge as it passed through a model distribution system at bench scale (annular reactor). The results demonstrated that total coliforms and heterotrophic bacteria increased by three to four orders of magnitude, respectively, and that *E. coli* remained viable during the extent of the experiment (11 days). This research has established that although the reclaimed water met EPA standards for irrigation at the wastewater treatment plant, there is great potential for bacteria regrowth during transport that could place the water out of compliance at the point of intended use.

By FY04 we will have completed the column study looking at microbial resistance to antibiotics present as wastewater contaminants under groundwater recharge conditions.

By FY05 we will have completed fieldwork on fate and transport of wastewater contaminants.

with the USGS and the ARS Food and Feed Safety Research Unit. The information obtained from this study will help evaluate the degree by which water reuse practices will

influence the transmission of antimicrobial resistance determinants to groundwater when wastewater is used for irrigation and/or groundwater recharge.

FIELD STUDIES

Field studies are in progress to study impacts of land application of wastewater on groundwater quality. A project was initiated in collaboration with the USGS to study the spatial and temporal distributions of wastewater contaminants in the surface water and in the underlying groundwater at an effluent-dependent stream, the Santa Cruz River, Tucson, AZ. This study will help to better understand the processes affecting bacterial concentration in effluent-dependent streams as well as address potential effects of incidental ground-water recharge in the Santa Cruz River on groundwater quality. Other field sites are currently being evaluated for further fieldwork in conjunction with the USGS.

PATHOGEN REGROWTH

Field sites are being evaluated to study a real-time distribution system where effluent is used for turf irrigation and also to compare the efficiency of different disinfection processes (UV and chlorine disinfection) on preventing regrowth. Results from this research project will greatly add to our understanding of the microbiological safety of water reuse practices as well as their potential environmental hazards and will ultimately help protect public health and our future groundwater resources.

Describe the major accomplishments over the life of the project, including their predicted or actual impact.

There is a need to understand the environmental fate of microorganisms and the potential for bacterial regrowth in reclaimed water used for crop irrigation so that future problems of food contamination via wastewater irrigation can be prevented. A laboratory study was conducted at the U.S. Water Conservation Laboratory to assess the survival and regrowth potential of bacteria present in tertiary-treated effluent used for crop irrigation and surface-water discharge as it passed through a model distribution system at bench scale (annular reactor). The results demonstrated that total coliforms and heterotrophic bacteria increased by three to four orders of magnitude, respectively, and that *E. coli* remained viable during the extent of the experiment (11 days). This research has established that although the reclaimed water met EPA standards for irrigation at the wastewater treatment plant, there is great potential for bacteria regrowth during transport that could place the water out of compliance at the point of intended use.

By FY04 we will have completed the column study looking at microbial resistance to antibiotics present as wastewater contaminants under groundwater recharge conditions.

By FY05 we will have completed fieldwork on fate and transport of wastewater contaminants.

By FY04 we will have finished evaluating field sites to examine microbial regrowth in real-time distribution systems and by FY05 we will have completed the sampling and assessment of the real-time distribution system.

By FY06, mitigation procedures for microbial regrowth in distributions systems will be developed and final reports and manuscripts will be prepared.

Publications:

Cordy, G., Duran, N.L., Bouwer, H., Rice, R., Kolpin, D., Furlong E., Zaugg, S., Meyer, M., Barber, L. Persistence of pharmaceuticals, pathogens, and other organic wastewater contaminants when wastewater is used for ground-water recharge. 2003. Proceedings of 3rd International Conference on Pharmaceuticals and Endocrine Disrupting Chemicals in Water. p. 195-196.

Jerman, J., N.L. Duran. Deterioration in Microbial Quality of Treated Effluent Used for Crop Irrigation During Distribution. ASM 103rd General Meeting, Washington, D.C. 2003. Abstract p. 582.

Spencer, K.L., N.L. Duran. Effects of Effluent-Dependent Stream on Microbial Loads and Groundwater Quality. ASM 103rd General Meeting, Washington, D.C. 2003. Abstract p. 609.