International Development Research Centre

MANUSCRIPT REPORT

Reclamation of Nutrients, Water and Energy from Waste

A Review of Selected IDRC-Supported Research

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RECLAMATION OF NUTRIENTS, WATER AND ENERGY FROM WASTES: A REVIEW OF SELECTED IDRC-SUPPORTED RESEARCH

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FOREWORD

The urgent need in many developing countries to develop low-cost, appropriate technologies for the treatment and disposal of human, animal and industrial wastes, led to the support by the Health Sciences Division of a number of projects that focused on waste treatment and waste reuse.

This report summarizes the research findings of 10 projects and one survey that were funded by IDRC from 1977 to 1982 to study the technologies, costs and public health implications of waste reuse. Grants totalled approximately CAD \$1.5 million. The work was carried out by researchers in the following countries: Guatemala, Israel, Korea, Malaysia, Peru, Singapore and Thailand.

It was felt that a report such as this would serve at least three important functions: 1) to summarize important and relevant research findings; 2) to stimulate researchers to continue the work that still needs to be done; and 3) to inform other agencies of IDRC's research support in this field and thus avoid duplication and conserve scarce resources. It is therefore hoped that this document will complement the UNDP/World Bank Global Integrated Resource Recovery Program.

It is also hoped that this publication will serve as a focal point in the search for new approaches and solutions to waste treatment, disposal and reuse. In this regard, IDRC is seeking to collaborate with "the World Health Organization, the International Reference Centre for Waste Disposal, and the London School of Hygiene and Tropical Medicine in the development of joint projects in waste reclamation. This report also represents a contribution to the activities of the International Water Supply and Sanitation Decade (1981-1990).

At the time of their inception, IDRC-supported waste reuse research projects were breaking new ground. Not only were new technologies developed, but also the research capabilities of young scientists were enhanced and national research institutions strengthened. Some of the scientists have continued work in the broad field of resource recovery with support from their own governments or from other international agencies such as UNDP and the World Bank.

This manuscript report does not include a literature review of waste reclamation research. Instead, selected projects have been summarized to provide an overview of IDRC-supported waste reclamation projects. Since only relevant aspects of the individual projects are reproduced in the annex, readers interested in more detailed information are encouraged to consult the researchers themselves.

It must be noted that IDRC is not necessarily suggesting that the research methodologies presented are the best or the only ones to be pursued. They do represent, however, several different approaches for investigating the technical and economic feasibility of waste reuse and its effect on human health and the environment.

D. Sharp

ACKNOWLEDGEMENT

The extensive contribution made by Dr. Michael McGarry, formerly of IDRC and now of Cowater International (Ottawa, Canada), to this report is gratefully acknowledged. He was responsible for the development of many of the projects summarized in this report and was instrumental in the establishment of the IDRC wastes reclamation network. His detailed review and constructive criticism of the document are highly appreciated. As well, the contribution of Mr. Diep Tam, also of Cowater International, in the technical editing of the manuscript is gratefully acknowledged.

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SUPPLARY

Wastes have often been called "resources out of place". In many developing countries these resources have traditionally been recognized as such and are reused as important agricultural inputs. The East and West have diverged on this point. For example, during the last century in the West, human wastes (excreta) have been regarded as highly undesirable: to be flushed away for treatment and dumped elsewhere. At the same time in the East, excreta (nightsoil) has been collected, and applied to the land. As both fertilizer and soil conditioner nightsoil has been a very important element in China's food production.

By far the largest quantity of wastes from developing country populations is discharged in the form of nightsoil, wastewaters and solid wastes (garbage/refuse). Managing these wastes is very expensive. The costs of collecting and treating them can absorb as much as 30% of a city's income.

In addition to these costs, the city which does not reclaim its wastes loses the opportunity of recovering valuable byproducts from them. The collection of wastes for treatment and reuse provides two principal benefits (1) waste treatment and (2) byproduct recovery. Wastes reclamation holds considerable potential for reducing the costs of wastes management.

There are various ways in which human wastes can be treated and recovered. (1) Waste-fed aquaculture produces plants and fish in ponds fed with organic wastes. Septic tank sludge, and treated nightsoil and sewage are all forms of wastewaters which can be used for this purpose. (2) Wastewaters can also be treated in stabilization ponds before being reused as irrigation waters in agriculture. In this way both water and the remaining nutrients in the water can be reclaimed. (3) Instead of dumping or incinerating organic wastes, they can be composted, and transformed into humus for soil rehabilitation. (4) Livestock (and human) wastes can be treated anaerobically in digesters which naturally break down their organic materials into a gas called biogas composed of 60% methane, 40% carbon dioxide and nutrient rich slurry. The gas can be used as an energy source for cooking, and the slurry used as a fertilizer on the land.

However, in these forms, wastes reuse is not a panacea for all our pollution control problems. The value of the recovered byproducts only partially cover the costs of collection and treatment. Strict control over the reuse processes is necessary to ensure that public health is not endangered. Because the potential benefits from wastes reuse technologies are so great and the health implications so important, these processes have attracted a great deal of attention world wide by researchers, governments and aid agencies over the past decade.

Waste reclamation has been widely practiced in many areas of the world for decades, and even centuries. Traditional waste handling and waste reuse practices seldom protect against the dangers of disease transmission and are often neither technically nor economically efficient. At the time of initiation of the IDRC-supported wastes reclamation program, researchers in many countries had already begun to realize the urgent need for research into waste reclamation technologies that were appropriate, efficient, and above all were low cost and posed no threat to human health.

IDRC supports research of direct relevance to developing countries, especially applied research with the potential for widespread application. Such was the case with the wastes reclamation network of projects which were initiated in Thailand, Malaysia, Peru, Israel, Guatemala, Korea and Singapore in the late 1970's.

The network concept involves the promotion of inter-project collaboration, standardization of methodology and sharing of results. The waste fed aquaculture network began with a meeting of interested researchers which was later followed by mid-project and end-of-project meetings.

Research projects which were eventually funded by IDRC were designed to respond to the individual country's research needs and conditions while at the same time the researchers were encouraged to have similar objectives and overall methodologies. A network co-ordinator was posted at IDRC's Singapore office to foster communication and inter-project collaboration.

IDRC utilized both a formal and informal network concept which consisted of scientific and technical collaboration and exchange of ideas. Visits, meetings, workshops, training

courses and correspondence as well as the use of common methodologies and discussion of findings formed the basis of the networks.

This report, intended for the general professional with interest in wastes reclamation, presents the principal results of the network. Due to space limitations it does not present all the research results achieved. For this the reader is referred to the detailed final research reports which may be made available by the individual research project leaders. As well, where information was available, publications arising from the IDRC-supported waste reuse projects are listed at the end of each project summarized.

Research results have been summarized and are presented in the annex along with the objectives of the research and brief discussion of the findings. Selected tables and figures from each project's final report have also been reproduced. These abridged reports (based on final reports submitted to IDRC) have been vetted by the researchers.

Table 1 illustrates the broad coverage of research interests in wastes treatment, water reclamation, fertilizer, plant and fish production, and energy recovery from wastes. Most of the projects utilized domestic wastewater as the initial source of nutrients although septic tank sludge, nightsoil, livestock wastes and solid wastes were also employed. Further detail of the projects' end products and public health considerations is given in Table 2 at the end of this section.

In most instances, the waste treatment processes used were biological. Table 2 shows that the most common form of treatment was the stabilization pond. These ponds are well suited to the warm, sunny climates of many developing countries, are relatively inexpensive to construct, and require little maintenance. Experience shows that they are the most suitable for use in developing countries where land is readily available and inexpensive, where future expansion is possible, and where trained operating personnel may not be available.

As can be seen from the stated objectives of the individual projects (see annex), the researchers were attempting to find new approaches and solutions to the problem of waste disposal and waste reuse. Research focused on technology development and the engineering aspects of waste reuse.

Table 1: Project Activities

Project	Waste treatment	Water reclamation	Fertilizer production	Plant production	Fish production	Energy recovery
Waste reclamation, Thailand	2	2		2	2	
Waste management, Thailand	3,5	3,5	3,5	3,5	3,5	3,5
Wastewater reclamation, Israel	2	2		2	2	
Pathogen transfer/wastewater, Israel	2	2		2	2	
Wastewater reclamation, Malaysia	2	2		2	2	
Stabilization Ponds, Peru	2	2				
Nightsoil survey	1,3	1,3		1,3	1,3	
Waste reuse, Korea	3,4		3,4			
Excreta reuse, Guatemala	3		3	3	3	3
Piggery waste treatment I, Singapore	1	1		1		
Piggery waste treatment II, Singapore	1	1		1		

Notes

- 1 Agro-industrial waste
- 2 Household wastewater
- 3 Nightsoil
- 4 Solid waste
- 5 Septic tank sludge

Projects' Scope

Waste recovery processes can be designed to recover water, fertilizer and humus, plants, fish and energy. The IDRC supported projects summarized here dealt with one or more of these aspects, alone or in combination. All processes performed the common function of treating wastes to an acceptable level before being reused or discharged to the environment.

Wastewater Reclamation.....

With the exception of the Korean and Guatemalan research, all projects experimented with processes which had the potential for reclaiming the treated wastewater for irrigation; one (Thailand) carried out crop growth experiments using the reclaimed wastewater.

Fertilizer Production.....

The Thai, Guatemalan and Korean research projects all produced organic fertilizer for use in agriculture. Biogas digester slurry was researched in Thailand whereas in Korea night-soil was composted with urban refuse to produce a soil conditioner and fertilizer. In Guatemala, human wastes were turned into dry humus-like fertilizer in double vault house-hold latrines.

Plant Production.....

Plant production in the form of algae as fish feed was studied in the Thai, Israeli and Malaysian experiments. In Guatemala the humus derived from latrines was applied to vegetables, while in Thailand corn was fertilized by nutrients in the reclaimed wastewater.

Fish Production.....

Fish, mainly carp and tilapia were grown in wastewater-fed ponds in Thailand, Malaysia and Israel. Both were intended for the human food market. The effluent from biogas digesters was fed to aquaculture ponds stocked with tilapia in the highlands of Guatemala.

Energy Recovery.....

Biogas was the single form of energy recovery from wastes that was studied by the network. Both the Thailand and Guatemala projects included biogas generation aspects as part of their nightsoil and septic tank sludge reclamation research.

Reclamation of Nutrients.....

All ten projects investigated nutrient recovery in one way or another. Aerobic composting of wastes and subsequent land application of the compost or its application to fish culture ponds was investigated in Korea, Guatemala and Thailand. Nutrients in stabilization pond effluent were used for fish culture or agriculture in Thailand, Israel and Malaysia. Finally, algae were grown in high-rate algal ponds in Singapore as feed for pigs.

The projects summarized in this report were generally successful in terms of developing design and operating criteria for waste treatment and reuse systems. It was demonstrated that under prescribed conditions, waste fed aquaculture can produce acceptable fish yields. It was also shown that aerobic composting and other processes can transform human wastes into innocuous fertilizer. The land application of the compost and the treated wastewater was carried out but not thoroughly investigated.

With the exception of the Guatemala project which developed and tested latrines and biogas digesters for family use, the research conducted dealt with domestic wastes from medium-sized communities, and agricultural or industrial wastes. Stabilization ponds were shown to be very successful in treating wastes from a variety of sources but local conditions such as climate, availability of land, strength of wastes, and reuse applications strongly influenced design and operational criteria.

Composting was employed to break down domestic refuse and nightsoil in Korea. Pilot plant experiments proved composting to be a viable process, holding good potential for full scale use. A treatment system was designed and costed for a small township and found to compete favourably cost-wise with alternative treatment methods. Composting was also used to transform organic wastes such as nightsoil and water hyacinth into compost-like material which could be successfully fed to tilapia. This latter work was carried out as part of

the Thai research into alternative ways of safely treating nightsoil prior to its use as a fish feed.

Composting was used in Guatemala where fecal material with a small ash admixture was stabilized in double vault dry composting latrines. The process was both technically sound and economically feasible and the by-product compost could be applied to vegetable plots. Although acceptance of these types of latrines by the rural population was initially slow, hundreds of units have now been installed in rural areas.

Stabilization pond treatment of wastewaters was used for wastewater reclamation and/or fish production in several of the projects. As anticipated it was confirmed to be a low cost, efficient method of reducing both the organic content (800) and the pathogen load of the influent. Design and operational criteria were defined by the Peruvian project and observed to agree with those used by other projects in Malaysia, Thailand and Israel (after necessary adjustments for climate and sewage characteristics).

The high rate algal pond is a variation on the stabilization pond in that its main purpose is to produce microscopic aquatic plants (algae). Algae, normally 50% protein (dry weight basis), was produced in this way from piggery wastes in Singapore for the purpose of recycling it back to the pigs as feed. The separation of the algae from the pond water was technically possible through a belt filtration harvester. Harvesting by centrifuge was also feasible. Algae were fed "wet" to the pigs at up to 15% dietary levels with little or no adverse dietary effects.

The production of fertilizer slurry and energy was achieved in the Thai project. Operational parameters were determined for the biogas digestion of nightsoil and vegetable matter. However, the focus of the work was on use of the digested slurry to feed fish culture ponds, which proved successful.

Six of the projects studied fish culture in waste enriched ponds. In general, the research on fish production had three main goals: (1) to produce acceptable fish yields; (2) to develop sanitary engineering criteria; and (3) to study the public health aspects related to wastewater aquaculture.

Several different approaches to raising fish in a wastewater environment were tried. Fish culture was shown to be possible with extrapolated yields ranging from two to nineteen tons per hectare per year depending on the fish species stocked, stocking density, amount and quality of the wastewater applied and other factors. Table 3 indicates the potential fish yields observed in the projects with an aquaculture component.

Researchers in Israel found that the three critical wastewater quality parameters were the biochemical oxygen demand (80D), detergents and bacterial load. High 80D depleted oxygen especially in the early morning, and led to fish loss. Detergents, often present in domestic wastewater, were found to be toxic to fish. High bacterial concentrations were found to cause stress in the fish and lowered their disease resistance.

The degree of bacterial contamination of the exterior, viscera and flesh of the fish reared in wastewater was found to depend to a large extent on the bacteriological quality of the water. Therefore, raw sewage should be partially treated and diluted before being used in fish ponds. Fish culture in primary stabilization ponds receiving settled raw sewage was not successful in the Malaysian project, mainly due to a high 800. An air-breathing species was, however, successfully cultured in the secondary stabilization ponds.

Public Health Aspects

The treatment and sanitary disposal of human and animal wastes is necessitated by a need to protect both the natural environment and public health. The projects present results on the survival and persistence of human pathogens (including bacteria, protozoa, helminths and viruses) in reclaimed wastes and wastewater as well as in fish ponds and the fish themselves (see Table 4).

The various biological treatment methods employed served to decrease the concentrations of bacteria, parasites and protozoa in the effluents, but did not eliminate them. Helminth ova were removed by sedimentation in stabilization ponds. The destruction of parasite ova by composting was successful but was dependent on the temperature achieved. For example, in the Korean project, it was necessary to cover the experimental compost piles during the winter months in order to maintain an internal temperature of 55°C or more to achieve pathogen destruction.

Two projects (Malaysia and Israel) addressed the potential health problems associated with the accumulation of pesticides and PCBs in wastewater and in fish. Israeli results showed the presence of residues of several organochlorine insecticides and PCBs in fish but on a scale similar to those found in fish taken from commercial fish ponds.

In Peru, at least two stabilization pends in series was shown to be necessary for parasite and protozoa removal. Secondary pend effluent still had fecal coliform counts ranging from 10^2 to 10^7 per 100 ml.

The Israeli experiments illustrated that tilapia was the hardiest fish investigated and was best able to resist penetration of bacteria into its muscle. There existed a pattern of penetration of bacteria and bacteriophages into the organs and muscles of fish. The threshold concentration of bacteria in the water at point of penetration was lower for bacteriophages than bacteria. Bacteria were recovered in small numbers from the muscles and blood of fish tested. The numbers of micro-organisms in the pond water determined their presence and concentration in the fish.

The results of the field as well as the laboratory experiments showed that fish can be produced using domestic wastewater effluent. They also showed that the fish produced can be safe for human consumption provided that special attention and consideration is given to the critical concentration of pathogenic bacteria in the pond water and to the threshold concentration of these bacteria in the fish.

Overall the results pointed to potential public health hazards emanating from waste reuse operations. Little is known about the health risks which may accompany the reuse of treated excreta and how these risks may be minimized at least cost. Before encouraging the practice of wastewater recycling, there is a need to demonstrate that associated public health risks are within acceptable limits. Each process and each situation in which it operates should be given separate consideration.

Economic Aspects

The importance of the financial viability of their reuse processes was recognized by all researchers, however cost/benefit analyses were often relegated second priority and some-

times unexpected delays and technical difficulties associated with the research made it impossible for them to carry out adequate cost analyses within the allotted time. However, cost analyses were performed in the Singaporean, Malaysian and Korean projects. Somewhat more limited analyses were carried out by the Thai and Guatemalan projects.

Treatment of pig wastewater in the Singapore projects was found to be economical for farms with pig populations of 30,000 or greater. Proposed aerobic composting methods in Korea were also found to be economically viable, even without including the potential revenue from the sale of the compost. The double-vault composting toilet studied in Guatemala was found not only to be affordable by the rural Indian population, but to be gaining in demand and popularity.

Apart from the purely technical findings of the network projects, there have been other noteworthy accomplishments. These relate to the IDRC objectives of building research capability in developing countries. For example, in Thailand the researchers and their institution have continued and expanded their work in waste-fed fish culture. In Peru the research has been extended to include fish production. The UNDP/World Bank project on wastes reuse has utilized both researchers and experimental results from the network and expanded its investigations into the viability of waste reuse processes in developing countries.

RECOMMENDATIONS

Based on the research findings of the ten projects summarized in this report, several broad generalizations can be made regarding gaps in knowledge and future research directions.

The selection of the appropriate resource recovery technology will depend on the site, the nature of the waste to be recycled, the reuse application as well as social and cultural considerations. Research into waste reuse practices and applications for developing countries should continue to look at those technologies that are appropriate and do not require large technological, capital or energy inputs.

Recognizing that waste recycling schemes may introduce an increased risk to public health, it is important to determine the degree and exact nature of that risk, of the traditional local practices against it such as cooking methods, and to determine whether the increased economic, nutritional and environmental benefits outweigh any possible detrimental health effects.

Data are available on the ability of different waste treatment methods to remove or inactivate pathogenic bacteria and helminths from wastewater, but the fate of pathogenic viruses is not fully understood. The impact of existing waste reuse schemes on the health of the surrounding community, as well as on the health of the workers and others handling the products such as agricultural crops, fish or compost, deserves continuing examination. Subsequently, design criteria should be developed for larger-scale waste reuse systems.

Much of the existing fish production data has been based on harvests of the fish after only a short growing period. Longer term aquaculture studies need to be conducted in which the fish are harvested at market size.

Systems of wastewater recycling that include modifications to conventional stabilization ponds into ponds for combined fish culture and waste treatment deserve more study. Along with this, routine sampling regimes and methodologies for monitoring certain important wastewater quality parameters such as 80D, bacterial loads and detergents should be developed.

More information is required on the "threshold" concentrations of specific human pathogens (both bacteria and virus), such as <u>Vibrio parahaemolyticus</u>, <u>Vibrio cholerae</u>, <u>Salmonella</u>, <u>Shigella</u> and <u>Yersinia</u> amongst others. The effect of local food handling and cooking practices on the transmission of these pathogens also requires investigation.

Before large-scale waste reuse implementation projects are contemplated, it will be essential to establish their technical and economic viability by undertaking detailed technical and economic studies through pilot projects.

Table 2: SUMMARY OF SELECTED IDRC-SUPPORTED WASTE REUSE PROJECTS

PROJECT TITLE RESEARCH PERIOD	COUNTRY	MASTE REUSED	BIOLOGICAL TREATMENT	END PRODUCT	APPLICATION TESTED	PUBLIC HEALTH ASPECTS
Waste Reclamation 1977-80	Thailand	Domestic sewage	High rate algal ponda	Stabilized effluent, algae and zooplankton	1) Fish culture 2) Agriculture (maize crop)	Bacterial asssess- ment of effluents
Wastes Management 1979-82	Theiland	1) Septic tank sludge	Stabilization /fish ponda	Stabilized effluent, algae and zooplankton	Fish culture	Determination of bacteria, bacte- riophage and para- sites in compost.
		2) Nightsoil and organic refuse	a) Aerobic composting	Compost	Fish culture	Use of *ELISA test for the detection of antibodies of
		101430	b) Anaerobic digestion	Digester slurry and biogas	Fish culture	human pathogens in tilapia
Wastewater Reclamation 1977-80	Israel	Domestic sewage	Oxidation ponds	Stabilized effluent, algae and zooplankton	Fish culture	Determination of bacteria in waste-water and fish. Determination of organochlorides and PCB in fish.
Pathogen Transfer/ Wastewater 1979-82	Israel	Domestic sewage	Stabilization ponds	Stabilized effluent, algae and zooplankton	Fish culture	Determination of "Threshold Concen- tration" of bacte- ria and bacterio- phage for fish culture
Wastewater Reclamation 1977-80	Malaysia	Domestic sewage	Stabilization ponda	Stabilized effluent, algae and zooplankton	Fish culture	Determination of bacteria, parasi- tes and toxic metals in the wastewaters and in fish.

^{*} ELISA = Enzyme-linked immunosorbant assay

Table 2: SUMMARY OF SELECTED IDRC-SUPPORTED WASTE REUSE PROJECTS (continued)

PROJECT TITLE RESEARCH PERIOD	COUNTRY	MASTE REUSED	BIOLOGICAL TREATMENT	END PRODUCT	APPLICATION TESTED	PUBLIC HEALTH ASPECTS
Stabilization Ponds 1977-79	Peru	Domestic sewage	Stabilization ponds	Stabilized effluent, algae and zooplankton	Not done	Determination of bacteria, patho- genic protozoa and parasites in effluents
Waste Reuse 1978-80	Korea	Nightsoil and municipal refuse	Aerobic composting	Compost	Agricultur o	Not determined
Excreta Reuse 1978-80	Guatemala	Human excreta	a) Aerobic composting	Compost	Fish ponds	Microbiological analysis of com- post and biogas
			b) Anaerobic digestion	1) Biogas and slurry		digester effluent
				2) Effluent	Fish ponds	
Piggery Waste Treatment 1977-79	Singapore Phase I	Piggery wastes and wastewater	High rate algal pond	Treated effluent and algae	1) Algae extracted and used for animal feed	Not determined
1979-82	Phase II				2) Wastewater recycled	

Table 3: POTENTIAL FISH YIELDS FOR WASTEWATER-FED AQUACULTURE PROJECTS

RESEARCH PROJECT	FISH SPECIES	EXTRAPOLATED YIELD tons/ha/yr
Excreta Reuse (Guatemala)	Tilapia (<u>Sarotherodon</u> sp.)	2
Wastewater Reclamation (Israel)	Polyculture of Tilapia and Common Carp	2
Wastewater Reclamation (Malaysia)	Trichogaster pectoralis	5
Waste Reclamation (Thailand)	Tilapia nilotica	19 *
Wastes Management (Thailand)	Tilapia nilotica	3-6 **

^{*)} Based on a fish stocking density of 37 fish/ m^3 in 4 m^3 concrete ponds with an 84-day growth period.

^{**)} Based on a fish stocking density of 5 fish/ m^2 in 200 m^2 earthen ponds with a 170-day growth period.

Table 4: SUMMARY OF BACTERIAL QUALITY OF THE TREATED WASTES OR WASTEWATER, WASTE-FED FISH POND WATERS, AND WASTEWATER-GROWN FISH OF SELECTED PROJECTS

COUNTRY	FORM OF WASTE TREATMENT AND REUSE	BACTERIAL QUALITY OF STABILIZATION POND EFFLUENT, COMPOST, BIOGAS SLURRY OR CESSPOOL SLUDGE	BACTERIAL QUALITY OF FISH POND WATER	BACTERIAL QUALITY OF FISH
GUATEMALA	Aerobic composting Anaerobic digestion	Coliforms; MPN up to 10 ⁴ /100g Coliforms; MPN slightly lower than for composting (above)	Coliforms; MPN 10 ³ /100ml	Acceptable for human consumption from micro- biological and taste viewpoints
ISRAEL	Extended aera- tion effluent used for fish culture	-	10 ⁶ SPC/ml	Coliform and fecal coliform bacteria found in organs, flesh, blood and digestive tract
MALAYSIA	bilization	99% removal efficiency for total and fecal coliforms and fecal streptococci	Secondary pond/fish pond had total and fecal coliforms; MPN 10 ⁴ -10 ⁵ /100ml	Fish showed an increase in bacterial concentra- tion in flesh, gills and viscera
PERU	Primary and secondary sta- bilization ponds	Secondary ponds had fecal coliforms; MPN 10 ² -10 ⁷ /100ml	-	-
THAILAND	Stabilization pond effluent used in fish ponds	Fecal coliforms; MPN 10 ⁵ /100ml	Fecal coliforms; 10 ² -10 ⁵ /100ml	-
THAILAND	Compost fed fish ponds	Coliforms; 10 ³ -10 ⁸ /100g	Total and fecal coliforms; 104/100ml	No coliform bacteria and bacteriophage in blood, bile and muscle but found in intestine
	Cesspool sludge-fed fish ponds	Coliforms; 10 ³ -10 ⁷ /100ml	-	Similar to fish grown in commercial fish ponds in Thailand

WASTE RECLAMATION (THAILAND)

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Most developing countries are faced with the serious sanitation problem of human waste disposal. The costs involved in building and operating sewage treatment facilities are a burden to the economy as no financial return on the investment is provided. However, the possible reuse of such wastes for fish production could have the double advantage of improved sanitation and increased protein for the population.

The addition of human waste to water causes dense blooms of microalgae to form. These algae contain about 50% protein on a dry weight basis and could be used by phytoplankton feeding fish and converted to fish tissue. This project proposed to study the dynamics of such a relationship in sewage effluent, both raw and after treatment, in a high rate algal pond.

OBJECTIVES

The overall objective of this project was to determine the optimal operating parameters of a sewage treatment and herbivorous fish production system, taking into account the safety of the fish for human consumption and the quality of the effluent.

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The specific objectives were to:

- a) study the yield and feeding efficiency of herbivorous fish at different fish stocking and algae feeding rates.
- b) study the basic public health aspects with particular attention to monitoring pathogenic organisms present in the system at successive steps in the process of converting sewage into fish protein.
- c) determine the agricultural fertilization potential of the fish pond effluent; and
- d) carry out an economic appraisal in order to determine the system's applicability to rural and urban areas in Thailand and other tropical countries in Southeast Asia.

RESULTS

The experimental system consisted of three major components; a 200 m^2 high rate algal pond; a series of 4m^3 concrete fish ponds and earthen fish ponds with a 200m^2 water surface; and twenty-seven 4 m² maize plots (Figures 1 and 2). The high rate pond (20.0 m long, 10.0 m wide with a 0.5 m water depth) had a reinforced concrete base and walls made of cement blocks with an inner lining of concrete. To prevent shortcircuiting, baffles made of concrete blocks were constructed and the 100 m^3 pond contents were continually mixed by a paddle wheel.

five different experiments were conducted to assess the potential of raw sewage and stabilization pend effluent on fish production and on a maize production system.

Experiment 1.

<u>Tilapia nilotica</u> were cultivated in the 4 m³ concrete fish pands for 12 weeks at different fish stocking densities (1, 10, 20, 30 fish/m³) and for different detention times (40, 10, 3.3, and 2 days). The sewage was very weak with an average BOD_5 of 45mg/l and average organic loading of 75kq $BOD_5/ha/day$, making the pond nutrient and not light limited.

Figure 1. Schematic diagram of the system consisting of a high rate stabilization pond, concrete fish ponds and earthen fish ponds.

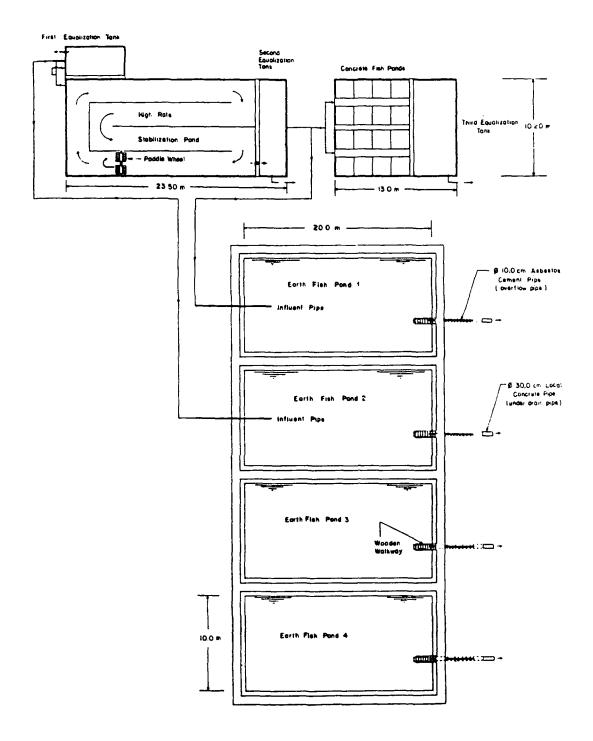
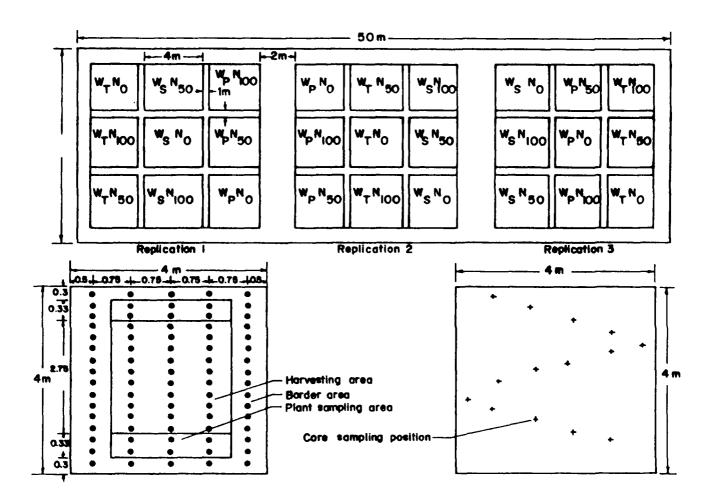


Figure 2. The layout of the maize fertilization study, Experiment 4.



Note: W represents the water treatment:

Ws = campus raw sewage

Wp = fish pond effluent

Wt = tap water

N represents the commercial fertilizer treatment:

No = 0 kg N/ha

N50 = 50 kg N/ha

N1oo = 100 kg N/ha

It was found that as the stocking density increased, the mean weight of individual fish decreased. However, at a given stocking density the mean weight of individual fish increased with a decrease in detention time since at shorter detention times there was a higher concentration of phytoplankton in the fish pond.

It was also found that as the stocking density increased, the total weight of the fish population increased. Also, as the detention time decreased from 40 to 10 to 3.3 days, the weight of the fish population increased, presumably due to the increase in phytoplankton concentration in the water. No advantage was seen in the 2 day detention time over the 3.3 day detention time.

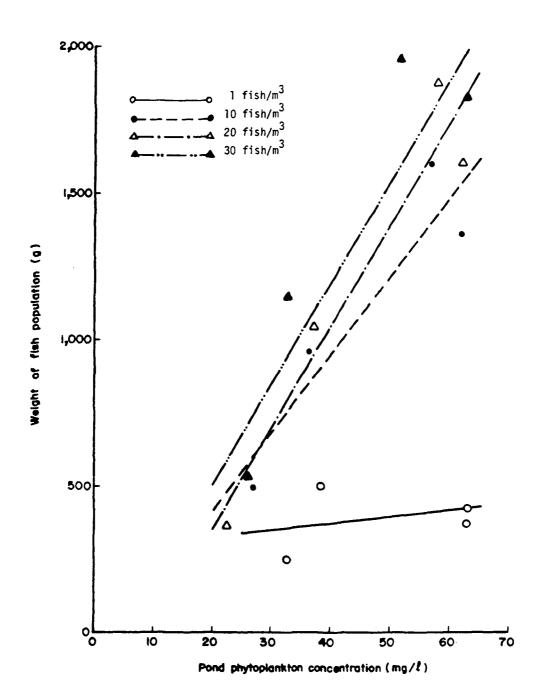
Extrapolated fish yields in the sixteen combinations of fish stocking density and detention time indicated that yields between 16 and 20 tons/ha/yr may be feasible with high fish stocking densities and short detention times (Table 1).

Table 1. Extrapolated fish yields in the 4 m^3 concrete fish ponds as a function of fish stocking density (1, 10, 20 and 30 fish/ m^3) and detention time (40, 10, 3.3 and 2 days) with a 12 week growth period.

Fish stocking density (number/m ³)	Detention time (days)	Extrapolated yield (kg/ha/yr)
1	40	2,496
1	10	5,046
1	3.3	3,725
1	2	4,460
10	40	4,940
10	10	9,604
10	3,3	16,095
10	2	13,536
20	40	3,649
20	10	10,428
20	3,3	18,789
21	2	16,096
30	40	5,312
30	10	11,534
37	3,3	19,546
28	2	18,325

A linear relationship between the mean pond phytoplankton concentration (up to 70 mg/l) and the fish yield was established (Figure 3). Since phytoplankton was the major type of food organism in the pond and in the gut of the fish, a clear indication is given that <u>Tilapia nilotica</u> thrived on phytoplankton.

Figure 3. The relationship between weight of fish population (g) per pond (4 m^3) after 12 weeks and the mean pond phytoplankton concentration (mg/1). Experiment 1.



Experiment 2.

<u>Tilapia nilotica</u> were cultivated in the 4 m^3 concrete fish ponds for 12 weeks at four stocking densities (4, 5, 6 and 7 fish/ m^3) and at different detention times (40, 10, 3.3 and 2 days). The detention times were the same as in Experiment 1, but a different range of fish stocking densities were used.

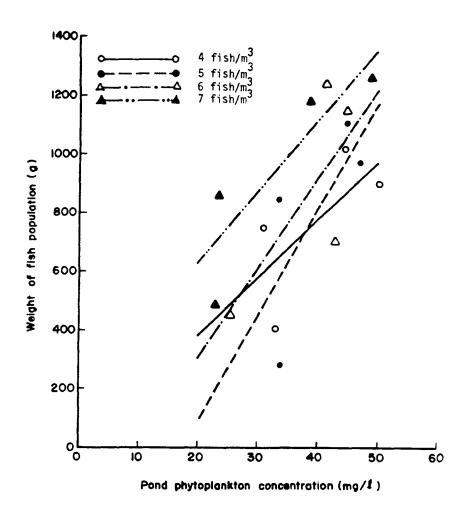
Fish growth data gave similar but less obvious trends as in Experiment 1, since a smaller range of fish stocking densities was used. However, the rates of increase of both mean weight of fish and weight of fish populations began to slow after 6-8 weeks. Extrapolated fish yields were lower than for Experiment 1, with a maximum of 10 to 12 tons/ha/year (Table 2).

Table 2. Extrapolated fish yields in the 4 m^3 concrete fish pends as a function of fish stocking density $(4, 5, 6 \text{ and } 7 \text{ fish/m}^3)$ and detention time (40, 10, 3.3, 2 days) with a 12 week growth period. Experiment 2.

Fish stocking density (number/m ³)	Detention time (days)	Extrapolated yield (kg/ha/yr)
4	40	4,009
4	10	7,536
4	3.3	10,217
4	2	9,018
5	40	2,867
5	10	8,448
5	3.3	9,757
5	2	11,129
6	40	4,515
6	10	7,067
6	3.3	12,437
6	2	11,506
7	40	4,881
7	10	8,611
7	3.3	11,858
7	2	12,682

The relationship between the weight of the fish population and the mean pond phytoplankton concentration is presented in Figure 4. The weight of the fish population increased with an increase in the pond phytoplankton concentration, but as in Experiment 1, the results were not statistically significant because of few data points.

Figure 4. The relationship between weight of fish population (g/4m³pond) after 12 weeks and the mean pond phytoplankton concentration (mg/1). Experiment 2.



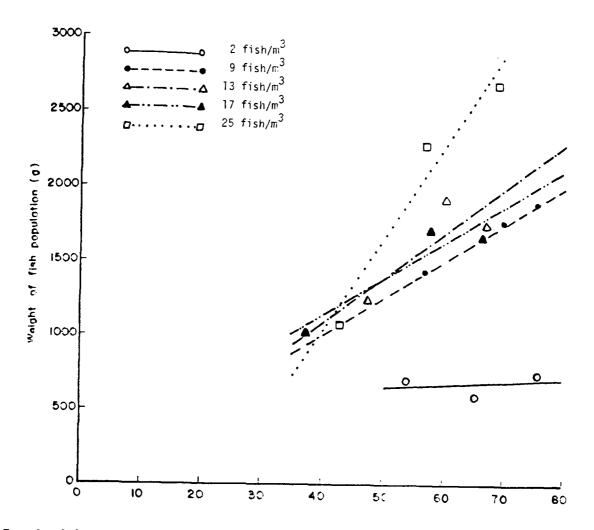
Experiment 3.

The same fish species, <u>Tilapia nilotica</u> was cultivated in the 4 m³ ponds but for a longer time, 20 weeks instead of 12 weeks. A combination of three detention times (10, 3.3 and 2 days) and five stocking densities (2, 9, 13, 17 and 25 fish/m³) were used.

Fish growth was similar to that occurring in Experiments 1 and 2, but after 12 weeks the rate of increase of both mean weight of fish and weight of fish population had slowed. After 20 weeks there was almost 100% mortality of fish in 6 of the ponds. This mortality could perhaps be attributed to low levels of dissolved oxygen, especially at dawn.

A linear relationship between the weight of the fish population and the pond phytoplankton concentration was established (Figure 5). This shows a direct response by the fish biomass to the increased food supply.

Figure 5. The relationship between the weight of the fish population (g/pond) after 12 weeks and the mean pond phytoplankton concentration (mg/l).

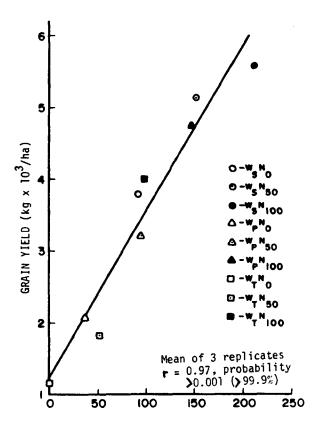


Experiment 4.

Twenty-seven 4 m^2 experimental plots, arranged in a split plot design with 3 replications, were seeded to maize. Yield assessments were performed on the treatments which consisted of three commercial nitrogen levels (0, 50 and 100 kgN/ha) and three irrigation waters (tap water, raw sewage and fish pond effluent).

A linear relationship between grain yield and total nitrogen applied to the plot was observed (Figure 6). The good yields of maize that were obtained by irrigation with raw sewage and fish pond effluent indicated that the form in which the nitrogen was applied was not important.

Figure 6. The relationship between grain yield (kg/ha) and total N applied (kg/ha).



Note: W represents water treatment: W_5 = campus raw sewage $W_P = \text{fish pond effluent}$ $W_T = \text{tap water}$

N represents commercial fertilizer treatment: N $_0$ = 0 kg N/ha N $_{50}$ = 50 kg N/ha N $_{100}$ = 100 kg N/ha

Experiment 5.

Fish were cultivated in the 200 m² earth pond system with each of the four ponds receiving a different treatment; high rate stabilization pond effluent, raw sewage, commercial fertilizer or pelleted feed.

Table 3 shows the results of the experiment. The biomass of fish after four months was quite similar in the ponds receiving raw sewage and stabilization pond effluent. The high mortality rate (72%) and low yield of fish in the pond receiving the pellets was unexplained but may have been due to the high turbidity of the water.

Bacteriological Studies

Standard plate counts/100 ml were performed on the raw sewage, stabilization pond effluent and fish pond effluent. As well, the MPN/100 ml was determined for total and fecal coliforms and fecal streptococci.

Fecal coliform counts in the raw sewage were in the range of 10^5-10^6 organisms/100 ml. Stabilization pond effluent showed some attenuation but results were still high, (about 10^5 organisms/100 ml). The fish pond effluent with a longer detention time showed a fecal coliform count of 10^2-10^3 and sometimes as high as 10^4-10^5 organisms/100 ml.

According to the WHO health standard for the reuse of wastewater for crops which are eaten cooked (such as maize and fish), the fecal coliform count should be less than 10^2 organisms/100 ml.

Economic Assessment

A cost analysis of a combined sewage treatment and water recycling system for a hypothetical city of 100,000 people was performed. A breakdown of the land requirements to treat the estimated 7000m³/day flow is as follows:

		area
high	rate stabilization pond	8.93 ha
fish	ponds	4.80 ha
land	for maize cultivation	49.21 ha
		62.94 ha

Table 3. The amount of stabilization pond effluent, raw sewage, commercial fertilizer and pellets added to the four earth ponds ($200~\mathrm{m}^3$) and summary of the fish data.

Experiment 5

Treatment	Amount added	Initial mean weight/ fish (g)	Final mean weight/ fish (g)	Initial fish stocking density (#/m ³)	Number of fish at harvest	% mor- tality	Final fish stocking density (#/m ³)	Initial biomass of fish (g)	Biomass of fish after 4 months (g)	Extrapo- lated yield (kg/ha/yr)	Weight of young fish (g)	Relative degree of spawning
High rate stabilization pond effluent	1,109 m ³	0.81	73.8	4	550	31	2.8	648	40,601	6,090	no data	++
Raw sewage	491.3 m ³	0.98	71.3	4	545	32	2.7	784	38,840	5,826	8,560	++
Commercial fertilizer	17,955 g	0.84	39.2	4	587	27	2.9	672	23,000	3,450	750	+
Pellets	11,344 g	0.76	25.5	4	222	72	1.1	608	5,670	851	-	0

The total gross return per year for the system was estimated to be:

 $$US 81,473 ext{ or } $US 1,294/ha ext{ if the fish is sold as human food} $US 76,673 ext{ or } $US 1,218/ha ext{ if the fish is sold as animal feed}$

A complete economic analysis including land, construction and operating costs of the system was not performed.

DISCUSSION

A combined wastewater treatment and nutrient recovery system was studied at the Asian Institute of Technology. The weak, raw sewage (average 800₅ 45 mg/l) passed through a high rate algal pond and into fish ponds stocked with <u>Tilapia nilotica</u>. The algal laden pond effluent served as a food supply for the fish. Crude food conversion ratios based on phytoplankton added and retained in the fish ponds were 6.2 and 2.5 respectively, at maximum fish yields. Since the lower value is similar to that obtained with some high protein supplementary feeds, the high nutritional value of the waste grown microalgae was demonstrated. Extrapolated fish yields of 10 to 20 tons/ha/yr could potentially be achieved, depending on stocking density and detention time.

A detention time of 3 days was used for sewage treatment in the high rate stabilization pond. COD reduction was 56% (COD initial concentration of = 81 mg/l COD and final concentration of 36 mg/l) which was low compared to other reports.

A great variety of phytoplankton genera became dominant in the fish ponds. This included 29 genera from the following phyla: Chlorophyta (greens), Cyanophyta (blue-greens), Bacillariophyta (diatoms), Pyrrhophyta (dinoflagellates) and Euglenophyta (euglenoids). The diversity of phytoplankton recorded as dominants was surprising when compared to other studies on high rate ponds, but was probably due to the relatively weak sewage.

An unexpected result from the earth pond experiment (Experiment 5) was the good fish production with low feeding regimes, i.e., the pond which received high rate stabilization pond effluent had a detention time of 20 days, and the pond which received raw sewage had a detention time of 46 days. Since the biomass of fish produced after four months was quite

similar for the ponds receiving raw sewage and stabilized pond effluent, respectively, a major conclusion to be drawn from the earth pond experiment was that prior stabilization of sewage in a high rate algal pond was not necessary for successful fish culture. This could possibly be due to the low 8005 of the raw sewage.

The fecal coliform counts for the earth ponds receiving both stabilization pond effluent and raw sewage frequently exceeded the WHO standard for waste water reuse despite the relatively long detention times of 20 and 46 days, respectively. Future studies should further investigate the public health aspects of waste recycling.

Publications Arising from the IDRC-supported Waste Reuse Project.

 Peter Edwards and On-Anong Sinchumpasak 1981. "The Harvest of Microalgae from the Effluent of a Sewage Fed High Rate Stabilization Pond by <u>Tilapia nilotica</u>. Part 1: Description of the System and the Study of the High Rate Pond." Aquaculture <u>23</u>: 83-105.

WASTE MANAGEMENT (THAILAND)

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The magnitude of the domestic waste disposal problem in Thailand, both in cities and in the countryside (where approximately 85% of the people live), called for a new technological approach with an emphasis on nightsoil or sludge management rather than wastewater or sewage treatment. Because of the suitability of the climate of the tropical developing world for phytoplankton and fish production, it was felt that this technology should be based on waste recycling rather than merely waste treatment and disposal.

OBJECTIVES

This project, therefore, aimed at developing bio-engineering guidelines for the construction of waste recycling schemes for both urban and rural areas in Thailand.

The specific objectives were to:

a) develop design and operational criteria for selected methods of waste treatment appropriate for rural or urban communities in Thailand;

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- b) investigate the potential reuse of wastes as feed for fish;
- c) assess pathogen transfer in the treatment and reuse processes; and
- d) determine the overall costs of the treatment and reuse processes.

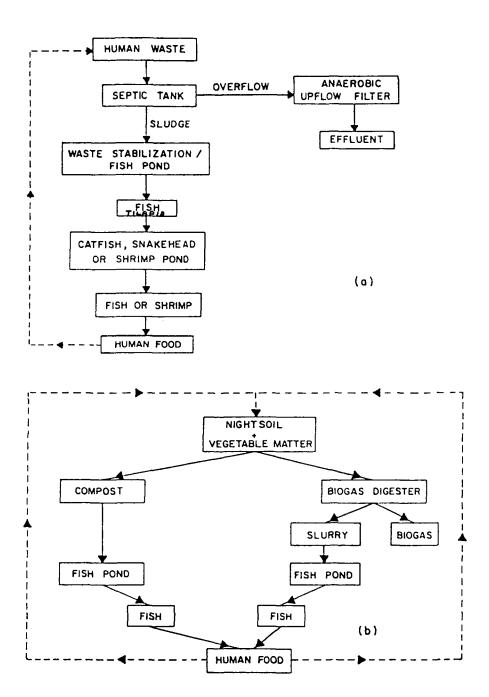
RESULTS

For rural areas, two 6-month experiments, using aerobically composted nightsoil as fertilizer for fish ponds, were performed. In one experiment the nightsoil was composted with water hyacinth and rice straw, and the compost was added to the fish ponds. In the other experiment, nightsoil and vegetable matter were used in biogas production, and the digester slurry was added to the fish ponds.

For urban areas a different experiment was performed. Septic tank sludge from Bangkok was loaded into a combined stabilization/fish pond system to grow tilapia fed on the phytoplankton produced from the nutrients in the human waste. To reduce the possibility of transmitting human pathogens along the food chain it was proposed to feed the waste-reared fish to catfish, snakehead fish or shrimp of high market value.

Figure 1 shows diagramatically the waste recycling system that was used.

Figure 1: Schemata of proposed human waste recycling system for (a) urban and (b) rural areas.



A. Rural Systems

Compost Fed Fish Pond System

A) Fish Growth Experiment 1

A simple aerobic composting method that mixed nightsoil, chopped water hyacinth and rice straw in different proportions to achieve the desired C/N ratio and moisture content was used. During the 170 day experiment, moisture content was kept at 70% and C/N ratios were kept at 20 and 30.

Four 200 m² earthen fish ponds were stocked with <u>Tilapia nilotica</u> at a rate of 5 fish/m² (1000 fish/pond). Ponds 1 and 3 received compost while ponds 2 and 4 served as control (no compost).

The fish growth data, based on monthly samplings of the fish populations, indicated marked growth in those ponds fed with compost compared to the control ponds in which relatively little fish growth took place. Table 1 shows the results achieved.

Table 1: Mean fish weight (g) at monthly intervals over a 170 day period. Ponds 1 and 3, 50 kg COD/ha/day, compost C/N 30 and C/N 20, respectively; ponds 2 and 4 controls

	Pond Number						
Date	1	2	3	4			
5 Feb 80	0.3	0.3	0.4	0.3			
5 Mar 80	4.3	4.6	7.3	4.2			
9 Apr 80	16.3	5.8	15.7	8.2			
8 May 80	21.9	5.4	52.8	9.8			
11 Jun 80	27.4	9.8	64.6	11.5			
8 Jul 80	26.0	10.1	71.4	8.5			
11 Aug 80	27.9	8.7	75.2	8.2			

The extrapolated yields for ponds receiving compost are 2,845 and 5,572 kg/ha/yr for ponds receiving compost of initial C/N ratios of 30 and 20, respectively, compared to 816 and 875 kg/ha/yr for control ponds.

B) Fish Growth Experiment 2

Similar to fish growth experiment 1, Tilapia nilotica was cultivated for 6 months in ponds receiving compost with a C/N ratio of 20. Compost loading rates were 25, 100, 0 and 50 kg COD of compost/ha/day for Pond 1, 2, 3 and 4, respectively.

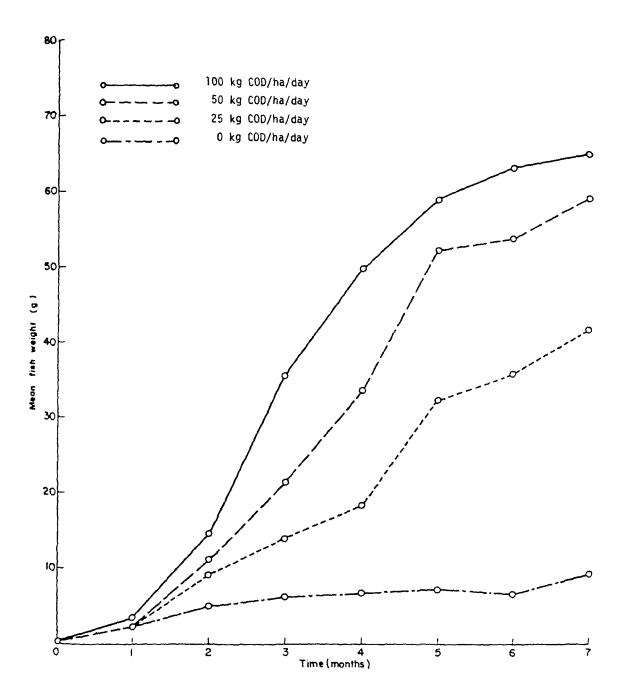
Table 2 shows the yields achieved at the end of the 190 day experiment. Relatively little growth took place in the control pond while compost fed fish ponds produced significant fish yields. Extrapolated yields in kg/ha/yr are 634, 1,507, 2,898 and 3093 for the ponds receiving 0, 25, 50 and 100 kg COD compost/ha/day. Fish breeding occurred in all ponds.

Table 2: Harvest data for the compost fed fish pond system
Growth period 187 to 193 days, mean 190 days

	Feeding regime		group) cm	Size 10 -	group 15 cm	Size	group 20 cm	[group	Total	Total	Extra- polated yield
Pond No.	1		Number of fish		Number of fish	Total wt(kg)	1		Number	fish		kg/ha/ yr
1	25	1.11	830	14.59	386	0	0	0	0	15.69	1,216	1,507
2	100	1.33	314	12.69	292	17.77	264	0.40	1	32.20	871	3,093
3	Control	6.37	1,855	0,23	12	0	0	0	0	6,60	1,867	634
4	50	3.56	1,717	14.68	327	11.93	177	0	0	30.17	2,221	2,898

Figure 2 plots the mean fish weight (at monthly intervals) during the experiment for the different treatments (organic loading rates at a stocking density of 5 fish/ m^2). A higher mean fish weight was achieved for the ponds receiving compost, as compared to the control pond. (Compost initial C/N ratio 20).

Figure 2: Nean weight of fish (g) at monthly intervals on the compost fed fish pond system at a stocking density of 5 fish/ n^2 . Compost initial C/N ratio 20.



Public Health Aspects of Fish Growth Experiments

Samples of compost and nightsoil were analyzed for the presence and densities of various enteric bacteria, bacteriophages and parasite ova. Samples of fish pond water, sediment and samples of blood, bile, meat and intestines of the fish were also analyzed for the presence and densities of enteric bacteria and bacteriophages.

Table 3 shows the results for examination of compost and nightsoil for parasite ova. Results suggest that the heat generated during composting was effective in activating the helminthic ova. However, due to the heterogeneous nature of the compost and possible uneven heat distribution, some ova could survive.

Table 3: Parasite ova in nightsoil and compost of initial C/N ratio 30.

E = empty ova, no larva inside; D = dead ova; + = positive ova;

- = negative ova; ? = not confirmed infective stage.

		First Pile					Second Pile		
Date	Sample	Hook Worm ova	Ancarls Ova	Opisthorchia viverini ova	Date	Sample	Hook Worm	ova Ascaria	Opisthorchis viverini ova
22 Nov 79	Nightsoil		-	-	20 Jan 80	Nightsoil	-	_	-
	Nighteoil	+	+	- 1		Nightaoil	-	٠ +	-
	Nightsoil	- !	-	_		Nightsoil	+	-	+
	Nightsoil	-	+	+		Nightsoil	-	+	
24 Nov 79	Nightsoil	i -	+	-	29 Jan 80	Nightaoil	-	-	-
	Nightsoil	+	-	-		Nightsoil	+	-	-
	Nightsoil	+	-	+ 1		Nightsoil	-	+	-
	Nightsoil	-	+	-		Nightsoil	-	} -	_
13 Feb 80	Compost	- 1	-	-	18 Jun 80	Compost	\ -	-	-
	Compost	i -	D	-		Compost	-	-	-
	Compost	Ε	-	- !		Compost	-	-	1 -
	Compost	-	-	-		Compost	-		-
6 Mar 80	Compost	-	+ 7	3	17 Jul 80	Compost	-	-	-
	Compost	i -	-	-		Compost	-		-
	Compost	-	-	-		Compost	-] -	-
	Compost		-	- !			i]	ļ
17 Apr 80	Compost	-	-	-		ļ	1	l .	
	Compost	-	D	-			1		1
	Compost		-	٤				1	
	Compost	j -	j -	j -					i
14 May 80	Compost	-	-	-		1		1	1
	Compost	-	-	-					1
	Compost	-	-	-		ì			ļ
	Congrest	-	J -	-		İ			1

Reductions in amounts of bacteria and bacteriophages occurred during composting. Results on the analysis of compost from fish growth experiment 2 showed the densities of coliform bacteria and bacteriophages to range from $10^3-10^{-8}/100$ ml and $10^2-10^5/100$ ml, respectively.

Comparison of the fish pond water and sediment in the control and compost fed ponds, showed the bacteria and bacteriophages densities to be within the same ranges. The concentrations of the total and fecal coliforms in all samples of the fish pond water were below $10^4/150$ ml. Few or no bacteriophages were observed in all ponds.

Table 4 shows the results of the analysis of fish organs for the presence of microorganisms. No coliform bacteria and bacteriophages were found in blood, bile and muscle samples of fish reared in the compost fed system. The presence of bacteria and bacteriophages in the intestines is a common phenomenon.

Table 4: Bacteria and bacteriophage in fish blood, bile, meat (muscle) and intestine of fish grown in compost fed ponds.

		14 Apri	il 1981	8 Apri	1 1981
Tests	Samples	Pond 1 25 kg COD/ ha/day	Pond 3 control pond	Pond 2 100 kg COD/ ha/day	Pond 4 50 kg COD/ ha/day
Standard plate count	Blood 1	0	0	0	0
per 1 ml (or per 1 g)	Blood 2	0	0	0	0
	Bile 1	0	0	0	0
	Bile 2	0	0	0	0
	Meat 1	2.5 × 10	1.7 x 10	2.0 x 10	1.4 × 10
	Meat 2	1.5 x 10	1.4 x 10	3.1 x 10	2.0 × 10
	Intestine 1	7.2×10^{8}	3.5 x 10 ⁷	3.4 x 10 ⁶	6.5 x 10 ⁷
	Intestine 2	3.7 x 10 ⁸	4.7 x 10 ⁶	8.7 x 10 ⁵	9.4 x 10 ⁷
Standard total coliform	Blood 1	0	0	0	0
MPN index per 100 ml	Blood 2	0	0	0	0
(or per 100 g)	Bile 1	0	0	0	0
	Bile 2	0	0	0	0
	Meat 1	0	0	0	0
	Meat 2	0	0 _	0	0 _
	Intestine 1	4.3×10^6	9.2 x 10 ⁷	2.4 x 10 ⁵	9.2 x 10 ⁷
	Intestine 2	3.3 x 10 ⁶	4.3 x 10 ⁶	7.9 x 10 ⁴	3.5 x 10 ⁷
Standard fecal coliform	Blood 1	0	0	0	0
MPN index per 100 ml	Blood 2	0	0	0	0
(or per 100 g)	Bile 1	0	0	0	0
	Bile 2	0	0	0	0
	Meat 1	0	0	0	j o
	Meat 2	0	0	0	0
	Intestine 1	9.1 x 10 ⁵	4.0 x 10 ⁶	1.1 x 10 ⁴	9.2 × 10 ⁷
	Intestine 2	4.6 x 10 ⁵	4.6 x 10 ⁵	9.5 x 10 ³	3.5 x 10 ⁷
E. coli Bacteriophage	Blood 1	0	0	0	0
MPN index per 100 ml	Blood 2	0	0	0	0
(or per 100 g)	Bile 1	0	0	0	0
	Bile 2	0	0	O	0
	Meat 1	0	0	0	0
	Meat 2	0	0	0	0
	Intestine 1	3.3 x 10 ²	6.8	6.8×10^{2}	2.4 × 10 ⁴
	Intestine 2	1.3×10^{2}	0	1.3 x 10 ²	3.3 × 104

Biogas Digester/Fish Pond System

Experimental results on the influent and effluent characteristics of the biogas digesters indicated that all four digesters operated satisfactorily. Table 5 shows the biogas production and its methane content for the four digesters. The mean biogas production rate in the four digesters was similar and approximated 0.2 m³ per kg volatile solids per day with a methane content of 60%.

Table 5: The biogas production rate (m³ of gas/kg volatile solids(VS/day) and the percentage methane content of the gas from the four digesters.

	Digester Number										
Month	1		2		3		4				
	Biogas Production Rate m ³ /kg VS/ day	Methane Content %									
Dec 80	0.057	48.3	0.09	49.0							
Jan 81	0.096	60.5	0.15	61.7		39.0		ļ			
Feb 81	0.121	64.0	0.125	62.5		65.5		54.8			
Mar 81	0.270	63.10	0.270	63.7	0.225	61.3	0.350	61.25			
Apr 81	0.400	60.0	0.350	62.0	0.350	63.0	0.280	61.0			
May 81	0.163	60.5	0.173	61.8	0.179	64.0	0.162	60.3			
Jun 81	0.180	64.0	0.182	65.0	0.180	62.0	0.156	61.0			
Mean	0.184	59.3	0.191	60.8	0.207	59.13	0.199	59.67			

Fish Growth Experiment

Due to difficulties in operating the digesters, biogas digester slurry had to be loaded into the 4 ponds at lower than expected rates. Mean loadings of 0, 6.5, 14.0 and 20.0 kg COD/ha/day were achieved. However, the fish growth data (Table 6) showed a progressive increase in fish growth as the organic loading was increased. Extrapolated fish yields were 728, 1,246, 1,809 and 2,838 kg/ha/year for ponds receiving 0, 6.5, 14.0 and 20.0 kg COD/ha/day, respectively.

Table 6: Mean fish weight (g) at monthly intervals over a 177 day period. Ponds 1, 2, 3 and 4 received 6.5, 14.0, 0 and 20.0 kg COD biogas slurry/ha/day, respectively.

Date	Pond Number						
Date	1	2	3	4			
23 Dec 80	0.44	0.44	0.44	0.44			
20 Feb 81	9.40	12.98	6.20	7.50			
18 Mar 81	14.00	17.60	8.36	10.84			
17 Apr 81	17.81	23.20	9.73	20.02			
22 May 81	19.40	26.00	9.50	31.30			
18 Jun 81	18.60	23.08	10.58	45.66			

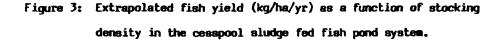
Public Health Aspects of Biogas Digester/Fish Pond System

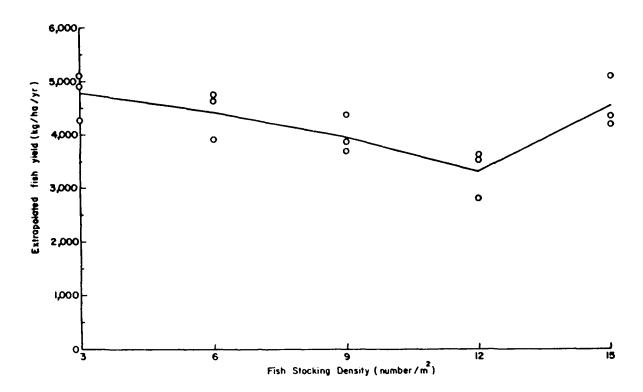
Examination of (1 to 2 litre) samples of biogas digester slurry showed only one sample to contain Ascaris ova. The slurry was found to contain high concentrations of bacteria and bacteriophages. During the fish growth experiment, however, the ponds fed with the biogas slurry had almost the same microbial concentrations in the pond water and sediment as those of the control which received no digester slurry. This suggests that there was dilution and natural die-off of the microorganisms when added to the fish ponds. Analysis of fish organs for the presence of microorganisms gave results similar to those from the compostfed fish ponds, i.e., no coliform bacteria or bacteriophages in the blood, bile and meat.

B. Urban Systems

Cesspool Sludge/Fish Pond System. Fish Growth Experiment

Sixteen 200 m² earth ponds fed with cesspool sludge were stocked with <u>Tilapia nilotica</u> at various densities. The volume of sludge fed varied according to the COD content of the sludge. The mean value for the COD was 17,306 mg/l and ranged from a low of 7,826 to a high of 44,982 mg/l.





The fish growth data, based on monthly samplings of the fish populations, showed a progressive increase in fish size with time, the increase being inversely related to fish stocking density.

Although there was considerable variation within treatments, in general, average fish size decreased with increasing stocking density of fish. However, despite the marked variation of mean fish growth as a function of stocking density, the fish harvest data from the different ponds were remarkably similar (Figure 3).

Public Health Aspects

Infective ova of hookworm, Ascaris and Opisthorchis were not found in the cesspool sludge samples. Therefore, no examination for ova in the fish pond water and sediments was conducted.

Amounts of bacteria (SPC, total and faecal coliforms) in the cesspool sludge varied from about MPN 10^3 to $10^7/100$ ml.

Fish grown in the cesspool sludge fed fish pond system frequently had a marked antibody response to Aeromonas hydrophilia whereas the antibody titres to Klebsiella pneumoniae and Salmonella typhimurium were either low or zero (Table 7).

Table 7: Antibody titres of fish grown in the cesspool sludge fed fish pond system.

All ponds were fed with sludge in a similar way, maximally but with serobic conditions at dawn. Fish were sampled at the 8th month and at the end of the experiment (9th month).

	Reciprocal antibody titre to										
Pond Aeromonas Number <u>hydrophil</u>			Klebsiel pneumoni		Salmonella typhimurium						
	8 months	9 months	8 months	9 months	8 months	9 months					
1	0	2	0	0	4	4					
3	8	0	0	0	0	0					
5	-	8	! -	0	[-	2					
6	4	4	2	0	2	0					
10	-	4	(o	0	! -	2					
11	4	8	0	0	2	0					
12	8	4	lo	lo	0	0					
13	8	4	0	8	0	2					
15	2	2	0	0	0	0					
16	0	4	0	0	0	2					

Septic Tank and Anaerobic Upflow Filter

The performance of two septic tank - anaerobic upflow filter units was evaluated. Crushed stones were employed as the filter media and each AUF unit used different sized stones and characteristics so that a comparison of the treatment efficiencies could be made.

More than 60%, 40%, 70% and 65% reductions in total COD, soluble COD, total SS and volatile SS, respectively, were observed for both AUF 1 and 2 at a one-day hydraulic detention time, and no significant advantages were observed for longer detention times.

The data for fecal coliform removal showed that an increased detention time resulted in an increase in percentage reduction of fecal coliforms, ranging from 53% to 100% removal.

The data for bacteriophage removal in the two AUF units gave results similar to those for fecal coliforms. An increase in the hydraulic detention time from 1 to 4 days gave better removal of bacteriophage.

DISCUSSION

Research results showed that significant fish yields may be obtained by the addition of compost to fish ponds containing <u>Tilapia nilotica</u>. The fish were observed to consume the compost as feed. Food conversion ratios of between 7.9 and 40.9 were obtained with compost with a high moisture content of about 60% to 70%. However, these food conversion ratios range from 3.2 to 12.2 when expressed as a compost moisture content of 8%, closer to the moisture content of supplementary feeds used in fish culture.

The major end products of anaerobic digestion are methane (CH_4) and carbon dioxide. However, the digester slurry still contains high concentrations of substrates and nutrients suitable for further reuses. The low pond loadings achieved (only a maximum of 20 kg COD/ha/day) resulted in little biological activity in the pond water. However, fish yields were surprisingly high (2,838 kg/ha/yr at the maximum loading).

The ranges of fecal coliforms found in the pond water samples in this study were within and below the threshold concentrations of 10^4 E. coli/100ml found by N. Buras to cause bacterial invasion of fish muscle (see Pathogen Transfer/Wastewater (Israel) p. 59). No fecal coliforms were found in the fish organs in this study.

Under conditions of a high water table or impervious soil, subsurface disposal of septic tank effluent is not possible. Therefore the performance of a septic tank system with completely submerged anaerobic upflow filter (AUF) was studied. Experimental results suggested that microbial removal in the AUF was primarily through adsorption, cell filtration and die-off. Adsorption of microorganisms onto biological films seemed to be the least important process in this instance. The helminthic ova which have relatively high specific gravity and larger sizes would be mostly removed by sedimentation.

Publications Arising from the IDRC-supported Waste Reuse Project.

- 1. Polprasert, C. and P. Edwards. 1981. Low-cost waste recycling in the tropics. Biocycle, J. of Waste Recycling 22(4): 30-36.
- 2. Polprasert, C., W. Kanok-nukulchai and V.S. Rajput. 1982. A ferrocement digester: Biogas and biomass production. J. Ferrocement 12(1): 25-34.
- Polprasert, C. and L.H. Hoang. 1983. Kinetics of bacteria and bacteriophages in anaerobic filters. J. Water Pollution Control Federation 55(4): 385-391.
- Edwards, P., C. Polprasert, C. Pacharaprakiti, V.S. Rajput and S. Suthirawut. 1983.
 Compost as fish feed, a practical application of detritivory for cultivation of tilapia. Aquaculture 32: 409-413.
- Polprasert, C. 1984. Utilization of composted nightsoil in fish production. Conservation and Recycling 7: 199-206.
- Polprasert, C., P. Edwards, V.S. Rajput and C. Pacharaprakiti. 1986. Integrated biogas technology in the tropics - 1. Performance of small-scale digesters. Waste Management and Research 4 (to be published).

ADDITIONAL STUDIES

Part of the research for this project was conducted by:

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The purpose of the research was to develop an enzyme-linked immunosorbant assay (ELISA) for the detection of <u>Tilapia</u> antibodies directed specifically at human bacterial and viral pathogens. This test was then applied to sera collected from fish grown in ponds exposed to treated human wastes. It was felt that evidence of antibodies against these pathogens in fish would indicate internal contamination of the fish at some time during their growth.

As an adjunct to this work, samples of fish pond sediment, water and organs were examined for the presence of <u>Salmonella</u>. In addition, samples of fish pond sediment and water were tested for the presence of enteroviruses and rotaviruses.

Antibody neutralization tests against enteric viruses commonly found in human domestic wastes were negative. Bacterial agglutination tests indicated the development of antibody titers against common enteric bacteria, but no statistically significant difference was observed between fish grown in ponds containing treated wastewater, AIT campus ponds and commercial fish ponds. This data indicates that there was no increase in incidence of antibody development among fish grown in ponds receiving treated sewage.

These results show that enteric viral or bacterial contamination of fish grown in treated domestic wastes was not any greater than that in commercial fish farms in Thailand. However, since large numbers of enteric bacteria occur in the intestines and skin of fish raised in wastewater-containing ponds, future studies should evaluate how these fish can be processed without contaminating the edible flesh.

WASTEWATER RECLAMATION (GLOBAL) ISRAEL

Dr. B. Hepher, Principal Investigator

Dr. E. Sandbank, Dr. G. L. Schroeder, A. Sukenick, A.S. Perry, Dr. J. Penciner

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Due to geography, climate and population pressure, Israel had a need to conserve its water resources. This included the recycling of wastewater along with nutrient recovery to prevent eutrophication of receiving waters. Increased industrialization along with water saving habits of the people led to the production of strong wastewaters. This wastewater, when used for fish culture had the potential for pathogen, pesticide and heavy metal transfer through the pond and by-product fish. Therefore, the public health aspects were an important part of the project.

OBJECTIVES

Specific objectives of this project were to:

- investigate the effect of wastewater loading and fish density in secondary stabilization ponds treating domestic sewage under temperate (Mediterranean) summer and winter conditions;
- assess the potential for improving the balance of nutrients in fish-feed within the pond by addition of supplemental carbohydrate sources;
- evaluate the impact of fish on effluent quality in terms of its potential re-use for irrigation;
- 4) grow Lemna (duckweed) in wastewater ponds to determine its treatment capabilities and subsequent use as fish feed; and

5) study the transfer of bacterial and viral pathogens and bioconcentration of heavy metals and pesticides through the secondary pond/fish system.

RESULTS

Four earthen ponds (400m² each) connected in series were used. Pond #1 served as the primary pond and ponds #2, #3 and #4 received effluent with a successive decrease in organic loading. On July 3, 1979, fish were introduced into ponds #2, #3 and #4 but all died within a little more than a month. On August 27, all ponds were restocked and harvested October 22 to November 4.

1. Water Treatment Aspects:

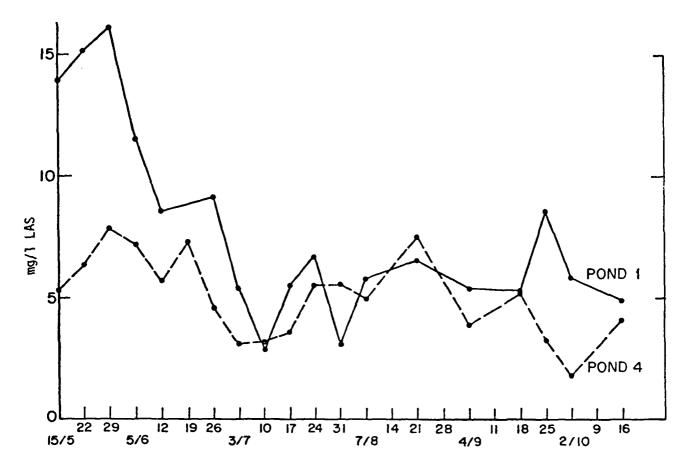
Detention times in the experimental pond system for ponds #1 (primary), #2, #3 and #4 were 8, 11, 16 and 32 days, respectively. These lower than expected detention times resulted in a lesser organic load reduction than was hoped for.

A number of the water quality parameters studied showed high fluctuations with peaks occurring, especially for COD and BOD in Pond #4. Figure 1 compares the MBAS in pond #4 (last in the series) and pond #1 (primary pond). There was a decrease in detergent concentration in pond #1 to about 5 mg/l. It seems that this was due to the degradation of the LAS, while the less degradable ABS persisted longer.

Measurements of dissolved oxygen concentration showed very low values of less than 2 ppm, occurring before dawn. An increase in the BOD such as had been observed, may have resulted in a complete depletion of 0_2 and in anoxia.

Determinations of algae and zooplankton were performed weekly. Green algae predominated although blue-green algae were often present in large numbers. The concentration of algae and rotifers seemed to be inversely related, especially in pond #4 prior to stocking with fish. After stocking, the pond became more stable with respect to phytoplakton.





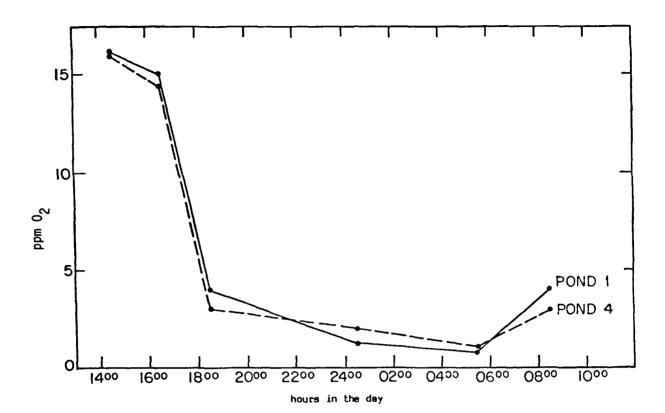


Figure 2: The daily 0_2 cycle, June 11-12, 1979.

2. Fish Growth:

Due to the sensitivity of silver carp only common carp, (the variety Israel mirror carp and a cross between the Chinese "big belly" carp and an Israeli mirror carp) and tilapia Sarotherodon aureus were introduced into the ponds. The following fish densities were achieved:

		<u>Fish/ha</u>
Pond #2	tilapia	5375
	common carp	2025
Pond #3	tilapia	6025
	common carp	2000
Pond #4	tilapia	6750
	common carp	1975

Table 1 shows the results of the sample weighings at 2 week intervals during July. Contrary to expectation, the fish did not grow at all but showed signs of stress. Mass mortality occured in all 3 ponds on August 13 and 14.

Table 1: Sample Weights of Fish in Ponds #2, #3 and #4 During the First Growth Period.

Pond	Fish species	Date	Average weight (g)	gain (g)
2	Tilapia	July 9	114.2	
		July 17	106.4	
1 :		July 30	115.0	0.8
l i	Common Carp	July 9	277.8	-50.8
L		July 17	227.3	-50.8
3	Tilapia	July 9	117.0	
1		July 17	115.8	
]		July 30	130.0	13.0
]		(Sept. 10) ^a	(160.0)	(43.0)
	Common Carp	July 9	281.0	ĺ
1		July 17	300.0	
}		July 30	266.7	!
		Aug. 13	282.3	1.3
		(Sept. 10)	(311.7)	(30.7)
4	Tilapia	July 9	123,0	
1	•	July 17	124.0	
i (July 30	125.0	2.0
		(Sept. 10)	(117.0)	(-6.0)
	Common Carp	July 9	290.0	
		July 17	266.7	<u> </u>
		July 30	261.5	[
		Aug. 8	265.2	-24.8
		(Sept. 10)	(288.5)	}
		(Oct. 22)	(295.5)	(5.5)

(a) figures in brackets are results of samplings taken after August 13 when mass mortality occurred.

The three experimental ponds as well as the high rate pond (HRP) were restocked August 27 and harvested from October 22 to November 4. No common carp survived the experiment. No tilapia survived in pond #2; 13 survived in pond #3 and 166 in pond #4 (69% survival). The latter only gained 8.3 grams during 55 days.

The tilapia and common carp in the adjacent high rate pond showed significant weight gains during the 69 day growing period. Extrapolation to a 200 day growing period gave an expected yield of about 1.87 ton/ha.

From these results, it is quite clear that the organic load in Ponds 2-4 was much too high for fish growth. Other substances such as detergents or ammonia also affected fish

growth. Where the organic loads were lower, as in the HRP, growth was rather high (considering that no supplementary feed was added) and so was the yield per pond.

Table 2: Average values of selected water quality parameters and their range in the raw sewage and experimental ponds (all figures in mg/l).

	Raw Sewage	Oxidation Pond #1	Pond #2	Pond #3	Pond #4	High Rate Pond
B00 ₅	380.4 (309-564)	207.2 (112-353)	176.8 (97-303)	156 (57–278)	151.9 (64-338)	-
Ammonia	58.6	41.8	16.1	7.77	3.4	5.6
	(43.3-76.9)	(30.3-55.7)	(5.8-28.8)	(0.32-14.2)	(0.0-16.6)	(0.9-25.0)
Detergent	9.2 (6.1-12.6)	7.5	4.7	4.5	5.0	2.35
(ABS+LAS)		(2.9-16.1)	(1.3-8.5)	(1.3-7.6)	(1.9-7.8)	(0.8-3.9)

3. Ecological Studies on Sewage Loaded Fish Ponds:

A number of studies were undertaken to better understand the pond eco-system. In spite of being rich in natural foods, as measured by volatile organic matter (VOM), the ponds did not allow for fish survival and growth.

A comparison of the respiration rates (BOD of filtered vs unfiltered samples) showed that ponds #3 and #4 had a high respiration rate (47% and 65% respectively) related to particles larger than 50 micrometers. This preponderance of phyto and zooplankton larger than 50 micrometers shows that the fish were not actively harvesting them.

Cellulose digestion (weight loss/5 days of cotton cloth hung in the ponds) gives an indication of biological activity in the ponds. Therefore, measures of cellulose digestion were taken in the experimental ponds and compared to that occurring in other ponds loaded with chicken and cow manure.

A lower rate of cellulose digestion was found in both the upper and lower water layers of the ponds. This may have been due to a steep oxygen gradient or as a result of inhibition by intense algae activity in the upper layers. As well, the substance inhibiting fish growth could also have been inhibiting the microbial activity responsible for the digestion.

4. The Effect of Fish on the Bacterial Population in Water:

Laboratory experiments were performed to study the possible reduction of pathogenic bacteria in the water by the fish. The tilapia (Sarotherodon aureus) were fasted and then introduced into 10 litre aquaria. Several aquaria remained empty of fish to serve as control. After 24 hours all the aquaria were inoculated with with an equal concentration of $\underline{E.\ coli}$.

Bacterial counts were performed on aquaria water samples taken throughout the experiment. The extinction of bacterial concentration has been observed in all aquaria, but those with fish showed a faster one than those without fish.

Figure 3 gives analyses made of \underline{E} , \underline{coli} survival at short intervals after the inoculation. It shows that without fish a lag in the extinction rate is apparent, while with fish, the extinction rate is more rapid during the first 8 hours and follows a logarithmic funtion.

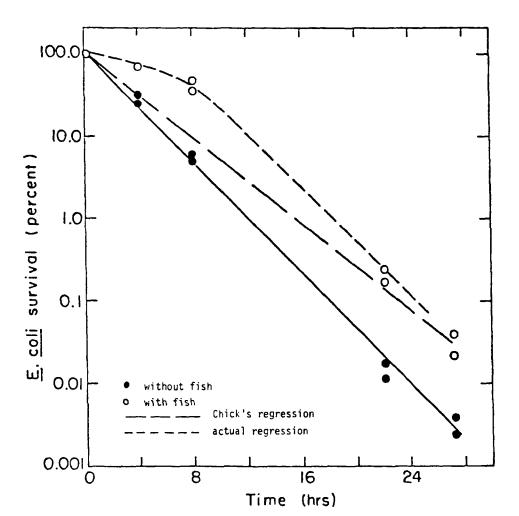


Figure 3: The Effect of Fish on Survival Rate (%) of \underline{E} , coli.

After this initial period, the rates of extinction become equal in the two treatments. It thus seems that the first hours of the experiment were critical with regard to the effect of fish on the <u>E. coli</u> population in the experimental aquaria. One cannot project, however, from this experiment to pond conditions.

Several studies were undertaken to try to explain the effect of the fish on the bacterial population. It was found that:

- a) No bacteria were taken orally by the tilapia;
- b) No flocculation of bacteria by body mucus occured; and
- c) No aglutinin activity against E. coli was found in the aguaria.

5. Determination of Pesticide Residues in Wastewater and in Three Species of Fish:

Analyses were made for total organochloride insecticides and PCBs in the wastewater and in the fish in the experimental ponds in 1979. Table 3 gives a summary of results in parts per billion on a wet weight basis.

Table 3: Organochlorine Insecticide Residues and PCBs in Three Species of Fish from the Experimental Fish Ponds [parts per billion (ppb) wet weight basis (average + standard deviation)].

Date	Species	(n)	≪ -нсн	∕& -нсн	Lindane	DDE	DDD	DDT	PCB (Avg)
30.7.79 30.7.79 30.7.79	Tilapia Carp Silver Carp	2		16.0(Avg)	13.8±5.2 12.5(Avg) 9.5(Avg)		63.5(Avg)	15.5(Avg)	1550 660 580
10.9.79 10.9.79	Tilapia Carp	6 4	20.5±9.7 13.5±7.3	8.6±3.0 10.8±8.5	18.6±11.4 8.0±3.5	28.2±9.8 53.3±14.6	13.3±6.8 9.8±7.6	9.2±3.6 7.0±4.0	875 940
20.10.79 20.10.79		9	6.0±5.8 3.8±2.0	14.4±13.4 7.8±7.2	6.2±4.7 4.8±1.5	23.3±12.9 27.8±2.2	4.3±2.3 5.5±1.0	24.3±19.2 18.0±3.3	1600 112.5

The results showed a high variability which is to be expected in a field study. They gave an accurate account of the variability between and within species as they occur in nature. Results show that there is a decrease in \propto -HCH and lindane in the fish in the 3rd series (20.10.79). This same decrease occurs in the pond waters. The greater persistence of

B-HCH is in line with its greater inertness to biodegradation.

The DDT groups (DDE, DDD, DDT) showed higher residues in water than in fish tissue. This is a surprising result since the DDT group accumulates readily in fatty tissues and it is expected that fish can actually deplete the aqueous environment of DDT residues.

Although the amounts of PCBs in the various ponds were low, bioaccumulation and biomagnification were much higher than those of the DDT group in the three fish species.

In conclusion, these results indicated the presence of residues of several organochloride insecticides and PCBs not much greater than those found in commercial fish ponds in some localities.

6. Public Health Aspects of Fish Grown in Experimental Wastewater Ponds:

To study the public health significance of pathogens in the wastewater, their concentrations in the experimental ponds as well as in the fish organs were studied. After growing fish in wastewater for one month, the effectiveness of depuration in clean water was also studied. An attempt was also made to verify the observation that the concentration of bacteria in the water determines their presence and rate of invasion into the various organs.

Detailed results outlining the public health aspects of fish grown in treated wastewater can be found on p. 59 to p. 72.

DISCUSSION

It was found that the fish were extremely sensitive to the wastewater conditions in which they were reared. All three species, tilapia, common carp and silver carp showed signs of stress that were related to amount of wastewater loading on the experimental ponds. Tilapia got darker (sometimes with distinct striped patterns) and excreted less mucous; common carp excreted more mucous than usual and silver carp were seen to produce skin appendices near the mouth.

During the first growing season, high concentrations of bacteria in the wastewater caused bacterial invasion of fish organs as well as the muscles. During the second growing season,

the pond was found to contain a high concentration of PCB and pesticides. Under these conditions, the fish did not feed, did not gain weight and had low concentrations of bacteria in the organs.

The above experiments showed that undiluted or only slightly diluted wastewater was not suitable for fish production. Since different fish species show different sensitivites to wastewater, it was important to stock the ponds with a suitable species. Silver carp was found to be more sensitive than common carp which in turn was more sensitive than tilapia.

A relationship was established between fish survival and several wastewater components from which maximum concentrations of certain parameters in the fish culture ponds were established:

COD 200-300 ppm BOD₅ 35-40 ppm

NH₄+N 1-1.5 ppm

detergents 1-1.25 ppm LAS + ABS

SPC/ml 1x10

Future studies should establish more precise limits of these variables.

Best experimental results (in terms of yield) were obtained using a stocking rate of 2000 common carp and 6000 tilapia per hectare. It can be expected that a yield of 10-15 kg/ha/day (2 to 3 ton/ha/year for 200 days) be obtained from fish ponds fed with wastewater stocked by the three fish species under Israeli conditions. Best results from the point of view of fish production were obtained using the high rate pond (HRP) effluent.

PATHOGEN TRANSFER/WASTEWATER (ISRAEL)

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The use of domestic wastewater for fish production presents potential hazards for fish handlers and consumers because of the presence of a variety of pathogens in these waters.

The common bacterial pathogens of warm-blooded animals do not cause diseases in fish. Some, however, such as vibrios, clostridia, streptococci, shigella and various serotypes of salmonella, may survive and multiply in the gut, mucus, and other fish tissues. As they are not the normal flora of the fish, they reflect the bacteriological condition of the waters in which the fish are grown. Human enteric viruses present a special problem in wastewater-fed fish ponds as they have been isolated, sometimes in large numbers, from domestic wastewater, and, as their infective dose is very low, their presence in the pond water even in small numbers may constitute a public health problem.

OBJECTIVES

The overall objective of this project was to study public health aspects of fish grown in wastewater-fed ponds in order to develop operational criteria for the wastewater treatment/fish culture process, so as to produce, from the public health viewpoint, fish suitable for human consumption.

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Specific objectives were to:

- a) determine the rate of sewage loading which can be sustained by the stabilization pond while ensuring that no pathogens accumulate in the organs and muscles of the fish;
- identify the species of fish which is least susceptible to invasion by bacteria and viruses when cultured in the sewage stabilization ponds;
- c) correlate sewage loading parameters, supplemental fish feeding rates and pond water bacterial concentrations with levels of contamination in the product fish;
- d) quantify, as far as possible, the balance between materials in the sewage-fed fish culture ponds and thereby determine the effects of sewage loading and fish populations on the quality of the effluent in terms of both its nutrient and pathogen concentrations; and
- e) develop operational criteria for the process of sewage-grown fish depuration in clean water ponds.

RESULTS

The project initially included only field experiments. These were designed to study the behaviour of the fish in ponds containing treated wastewater and follow the bacterial penetration into the fish.

At the end of the first growing season, it became clear that some of the findings needed elucidation and laboratory experiments were set up. In the field experiments fish were introduced into various experimental ponds, and the water and fish were examined periodically during the growing season for the presence of bacteria, bacteriophages and human enteric viruses. Laboratory experiments were designed to evaluate results obtained in the field experiments and to test assumptions based on observations made during the field experiments. In these experiments fish were inoculated with various microorganisms usually present in domestic wastewater and their incorporation into the fish organs was determined.

Field Experiments

Field experiments were carried out in four earthen ponds 400 m^2 each. The first pond received the extended aeration effluent and was used as an equalizer pond. The others were operated in parallel as fish culture ponds. The evaporation water in the experimental ponds was replaced by extended aeration effluent at a flow rate of 20 m^3 per day (5.0 cm/day).

The fish studied were tilapia aurea (Sarotherodon aureus), common carp (Cyprinus carpio), and silver carp (Hypophthalmichthys molitrix).

Examination of fish before their introduction into the experimental ponds, showed that fish reared in clean water do not harbor bacteria in their blood or muscles. Bacteria in small numbers usually are found in the spleen, liver and kidneys.

High concentrations of bacteria were recovered only from the digestive tract content (DTC) (Table 1). When carp were reared in treated wastewater in which the concentration of bacteria was 10⁶ SPC/ml, the numbers of bacteria recovered from the various organs increased and bacteria were found also in the muscles (Table 1). In one instance when human enteric viruses were present in the pond water, they were recovered also from the kidneys of the fish (Table 1). The results obtained for tilapia at the beginning and end of the growing season are summarized in Table 2.

Some differences were observed in the incorporation of bacteria into the organs in the three species studies. In common carp the highest concentration was recovered from the kidneys, followed by pronephros, spleen and liver. In tilapia it was from the pronephros, followed by kidneys and liver. And in silver carp the highest concentration was observed in the pronephros, followed by liver and kidneys.

When stressful conditions such as low dissolved oxygen or high organic matter concentration occurred in the pond, tilapia appeared to be less affected than carp.

The water level in the experimental ponds was maintained by daily addition of extended aeration effluent. Whenever the bacteriological quality of the extended aeration effluent

Table 1. Recovery of bacteria from common carp at the beginning and end of the first growing season.

Organ	SPC /g	Coliform MPN/ 100 g	Fecal Coliform MPN/ 100 g	E. coli phages MPN/ 100 g	Salmonella MPN/ 100 g	Vibrio parahaem.	Human enteric Viruses PFU/g
Beginning							
Blood*		0		0	0	-	NF
Muscle				0	0	+	NF
Liver		0		0	0	+	NF
Kidney		2		0	O	+	NF
Spleen		2	ļ	0	0	+	NF
DTC		1.4x10 ⁶		0	ο	+	NF
Bile		0		0	0	+	NF
End of Season							
Blood	3.5x10 ⁵	2.2×10 ⁴	7.0×10 ³	0	0	-	NF
Muscle	1.0x10 ⁵	1.3x10 ³	790	0	(0	-	NF
Liver	1.5x10 ⁵	7.9x10 ³	4.9x10 ³	0	0	-	NF
Kidney	7.5x10 ⁴	1.1x10 ³	1.3x10 ³	0	0	++	2
Spleen	3.5x10 ⁶	1.3x10 ⁵	3.3×10 ⁴	0 _	0	-	NF
DTC	6.2x10 ⁸	7.9x10 ⁹	2.2×10 ⁹	2.3×10 ³	0	++	NF
Bile	2.3x10 ⁷	2.2×10 ⁶	1.7×10 ⁴	0	0	-	

^{*} Values for blood are given per ml.

NF = not found

SPC = standard plate count

DTC = digestive tract contents

PFU = plaque-forming unit

deteriorated, the bacteriological quality of the fish deteriorated, and bacteria were recovered also from the fish muscles. The quality of the fish was always dependent on the quality of the water in which they were reared. As fluctuations in the water quality influenced the quality of the fish, whenever the quality of the water is relatively constant throughout the experiment it can be expressed as an average value, but if high fluctuations in the water quality occur, the extreme values, i.e., high concentrations of bacteria in the water persisting for some weeks during the experiment, should be given proper consideration because of their impact on the bacteriological quality of the fish.

In all the fish examined, bacteria were found in the muscles only after they were present in high numbers in all the organs. It seemed therefore appropriate to use the presence of

Table 2. Recovery of bacteria from tilapia at the beginning and end of the first growing season.

Organ	SPC /g	Coliform MPN/ 100 g	Fecal Coliform MPN/ 100 g	E. coli phages MPN/ 100 g	Salmonella MPN/ 100 g	H.E. Viruses PFU/ g
Beginning						
Blood*	63	0	0	0	0	0
Muscle Liver	0 75	0	0	0	0	0
Kidney	0	0	0	0	0	0
Spleen	1.0×10 ²	0	0	0	0	0
DTC	5.0×10 ⁴	ō	ō	ō	o	0
End of Season						
Blood	100	20	0	0	0	0
Muscle	6.5x10 ⁴	1.3x10 ⁴	2.1×10 ²	9	0	0
Liver	8.0×10 ⁴	2.4x10 ³	2.7×10 ²	9	0	0
Kidney	5.5x10 ⁵	1.7x10 ⁴	5.3x10 ⁴	9	0	0
Spleen	3.0x10 ⁵	2.4×10 ⁶	1.3x10 ⁶	9	0	0
DTC	3.5x10 ⁷	4.9x10 ⁷	1.1x10 ⁷	1.3x10 ⁴	0	0
Bile	5.0x10 ⁴	1.3x10 ³	1.4×10 ²	0	0	0

^{*} Values for blood are given per ml.

SPC = standard plate count

DTC = digestive tract contents

bacteria in the muscles of fish as a measure of the bacteriological quality of fish grown under these special conditions. The concentrations of bacteria that caused their appearance in the muscles of fish has been named the "threshold concentration".

In all three fish species studied, bacteria were recovered from the muscles only if their concentrations in the water exceeded a certain critical level. In our experiments the critical concentration of bacteria in the pond water was between 1.0 and 5.0×10^4 SPC/ml.

Depuration Experimenta

Depuration experiments were carried out on common carp and tilapia. The results are summarized in Tables 3 and 4 where it can be seen that when carp (Table 3) were heavily polluted depuration even for six weeks was ineffective, although some reduction in the number of bacteria recovered from the various organs was observed. When the concentration of

Table 3. Recovery of bacteria from common carp before and and after depuration in clean water for 6 weeks.

Organ	SPC /g	Coliform MPN/ 100 g	Fecal Coliform MPN/ 100 g	E. coli phages MPN/ 100 g	Salmonella	Vibrio
Before depuration						
Blood* Muscle Liver Kidney Spleen DTC Bile After depuration	9.5x10 ⁵ 1.0x10 ⁵ 1.5x10 ⁵ 7.5x10 ⁴ 3.5x10 ⁶ 6.2x10 ⁸ 2.3x10 ⁷	2.2×10 ⁴ 1.3×10 ³ 7.9×10 ³ 1.1×10 ³ 1.3×10 ⁵ 7.9×10 ⁹ 2.2×10 ⁶	17.0x10 ³ 790 4.9x10 ³ 1.3x10 ³ 3.3x10 ⁴ 2.2x10 ⁹ 1.7x10 ⁴	0 0 0 0 0 0 2.3×10 ³	NF NF NF NF NF NF	- - + + -
Blood* Muscle Liver Kidney Spleen DTC Bile	0 2.8×10 ⁴ 2.1×10 ⁵ 5.0×10 ³ 1.2×10 ⁴ 2.2×10 ⁵ 1.3×10 ⁸ 8.2×10 ⁶	0-20 3.3×10 ³ 1.3×10 ³ 200 1.1×10 ³ ND 2.4×30 ³	0 3.3x10 ³ 1.3x10 ³ 0 1.1x10 ³ ND 2.4x10 ³	0 0 0 0 0 93	NF NF NF NF NF	

^{*} Values for blood are given per ml.

NF = not found; ND = not done

SPC = standard plate count

DTC = digestive tract contents

bacteria was smaller, the depuration was more effective. The effectiveness of the depuration process increased as the concentration of bacteria in the fish decreased (Table 4).

E. coli as Bacteriological Indicators for Fish Quality

Since the main purpose of the project was to assess the public health aspect of fish grown in the presence of wastewater, special emphasis was placed on the recovery of coliform and fecal coliform bacteria from the fish samples examined. It had been observed during the various growing seasons that although coliform and fecal coliform bacteria were present in the pond water, they were not always recovered from the organs and very rarely from the muscles of fish. Other bacteria present in the pond water in much smaller concentrations

Table 4. Recovery of bacteria from tilapia aurea before and after depuration for 2 weeks in clean water.

Organ	SPC /g	Coliform MPN/ 100 g	Fecal Coliform MPN/ 100 g	Fecal Streptococci /g
Before depuration				
Blood* Muscle Liver	65 65 2.0×10 ³	0 52 2.3×10 ³	0 52 2.3×10 ³	0 0 32
After depuration			}	
Blood* Muscle Liver	0 30 10	0 0 0	0 0 0	0 0 0

^{*} Values for blood are given per ml.

SPC = standard plate count

were recovered in higher numbers from the same samples. As one of the conditions of an indicator organism is to be present and easily detectable in the sample, it seems that in this case E. coli was not performing the role of an appropriate indicator organism.

Field Experiments Conclusions

Of the three fish species studied, tilapia was the hardiest and able to survive at higher concentrations of pollutants, both chemical and bacteriological. The recovery of bacteria from the organs and muscles of fish was lower in tilapia than in common carp. Silver carp was the most sensitive and usually perished first.

It has been observed repeatedly that healthy, clean fish do not harbor bacteria in the blood or muscles. Bacteria in small numbers were recovered from the various organs and in high numbers $(10^6-10^7/g)$ from the digestive tract. The numbers of the microorganisms in the pond water determined their presence and concentration in the fish.

When salmonella were present in the pond water, they were recovered from the digestive tract content but were not detected in any of the fish organs. Coliform and fecal coliform

bacteria, although present in the water in high numbers, were not always detected in the muscles of the fish, whereas other bacteria were recovered. It seems, therefore, that in the case of fish grown in treated wastewater the role of fecal coliform bacteria as indicators for the presence of various bacteria originating from wastewater is questionable.

Laboratory Experiments

The laboratory experiments were designed to validate and clarify results obtained in the field experiments and test assumptions based on observations made during the field experiments.

The problems to be elucidated were:

- 1) The possible existence of different patterns of penetration for various organisms, bacteria, bacteriophages and animal viruses, into the fish organs and muscles.
- 2) The existence of a threshold concentration over which the various organisms might be able to penetrate freely into the organs and muscles of fish.
- 3) The existence of different threshold values for different organisms.
- 4) The effect of depuration of polluted fish.

The experiments comprised three parts: 1) inoculation of fish with microorganisms present in wastewater, 2) exposure to microorganisms in the water, and 3) fish depuration.

Tilapia aurea (Sarotherodon aureus) and common carp (Cyprinus carpio) were used in this study. The fish weighed between 60 and 100 g.

Inoculation Experiments

Fish were administered the test organisms directly into the esophagus by means of a capillary cannula. The volume inoculated was 0.1 ml, and it contained the number of the organism tested. The results obtained showed the existence of a threshold dose which differed from one organism to another.

The threshold dose has been defined as the number of bacteria that inoculated into the fish causes their appearance in the muscles within 2 hours after inoculation.

Table 5 summarizes the threshold doses found from the experiments. The thresholds for tilapia were lower for the two bacteriophages tested than for bacteria. Polio 1 LSc viruses were inoculated at a dose of 2.0×10^4 PFU/fish. Two 10^2 PFU/g were recovered from the muscles. These results indicate that the threshold for poliovirus is much lower than the dose inoculated.

As for Polio 1 viruses, when 2.0×10^4 PFU were inoculated to common carp, $5.0 \times 10^2/g$ were recovered from the muscles.

Although there were small differences between the values obtained in tilapia and in common carp, the pattern seemed to be similar. In all the fish tested high numbers of the organisms inoculated were recovered from the digestive tract content. The retention of inoculated organisms in the digestive tract is due mainly to the high concentration of phagocytic cells localized in the intestines of these fish. These phagocytic cells constitute the first barrier to foreign organisms invading the fish.

The lymphoid cells and macrophages localized in these organs express the potential of the defense mechanism of the fish. The threshold values show the limits of the system over which microorganisms, among them pathogens, might invade the muscles.

Table 5. Summary of Threshold Values found from experiments.

Organism	Threshold Values* In tilapia In common car					
E. coli Citrobacter freundii Streptococcus faecalis Salmonella montevideo T2 phage T4 phage	2.5×10 ⁶ 9.3×10 ³ 1.9×10 ⁴ 1.8×10 ⁴ 4.0×10 ³ 1.8×10 ⁴	1.5×10 ⁶ - 4.0×10 ⁴ 3.7×10 ⁴ 4.6×10 ³				

^{*} Bacteris and phage threshold values are expressed in numbers and plaque-forming units (PFU), respectively, inoculated per fish.

Exposure of Fish to Water Containing Bacteria or Bacteriophages

Common carp were introduced into 40 litre aquaria containing a mixture of sterile extended aeration effluent and tap water at a concentration of 1:100 to which the tested organisms were added. The organisms tested were: Salmonella montevideo and T_2 bacteriophages of E. coli B. During the exposure of carp to microorganisms in the water, both bacteria and bacteriophages penetrated into the fish and were recovered in various organs and in the muscles of the fish. It was interesting to note that after eight to nine days the concentration of both organisms in the digestive tract content was equal to or higher than the concentration in the water (Table 6). T_2 bacteriophages were recovered from the digestive tract content in all fish exposed. They were recovered from the peritoneal fluid when their concentration in the water was $2.6 \times 10^2/\text{ml}$ (Table 6). When fish were exposed to T_2 bacteriophages for 7 and 30 days, their concentration in the digestive tract content was higher after 30 days. At a concentration of $7.0 \times 10^4/\text{ml}$, T_2 bacteriophages were recovered from all the organs after seven days exposure (Table 7).

When common carp were exposed to salmonella, the bacteria were recovered first in the digestive tract content followed by peritoneal fluid, spleen, kidney and liver. Salmonella were recovered from the muscles when their concentration in the water was $5.0 \times 10^5/\text{ml}$ (Table 8).

By comparing the results obtained in the laboratory experiments to the ones in the field experiments (Table 9), it can be seen that when the concentration of bacteria in the water was 4.5 x 10⁴ SPC/ml, at the end of the season the concentration of bacteria in all the organs was high in both carp and tilapia. It can be stated therefore that exposure for a long time (120 days) to concentrations of bacteria above the threshold will result in a higher concentration of bacteria in the fish. The threshold values obtained in the laboratory give a good indication of what the critical concentrations of bacteria in the pond water should be.

Table 6. Recovery of T_2 bacteriophages from common carp organs after exposure to water containing the T_2 phages.

Concentration in water /ml	Time of exposure days	8lood /ml	Muscle /g	Kidney /g	Liver /g	Spleen /g	Peritoneal Fluid /ml	DTC /ml
20	9	0	0	0	0	0	0	1.6x10 ²
2.6x10 ²	8	lo	0	0	0	0	6	5.4×10 ³
8.0x10 ³	8	0	0	2	1	1	2.4×10 ²	1.3×10 ⁴
7.0×10 ⁴	9	5	2	22	16	18	5.0×10 ³	5.5x10 ⁵

Table 7. Recovery of bacteriophage T_2 from fish organs after exposure to T_2 bacteriophages.

Phage concentration in water/ml	Time days	Blood /ml	Muscle /g	Kidney /g	Liver /g	Spleen /g	DTC /ml
20	7	0	0	0	0	0	1.6×10 ²
	30	0	0	O	0	0	2.1x10 ³
3.0x10 ²	7	0	0	0	0	0	5.0x10 ³
	30	0	0	lo	0	0	1.0x10 ⁴
7.0×10 ⁴	7	4	2	20	15	17	4.0x10 ⁵
	30	5	2	22	17	16	1.7x10 ⁶

Table 8. Recovery of Salmonella from common carp organs after exposure to the becteria in water.

Concentration in water /ml	Time of exposure days	Blood /ml	Muscle /g	Kidney /g	Liver /g	Spleen /g	DTC /ml
2.5×10 ³	8	0	a	0	0	a	1.2×10 ³
2.8x10 ⁴	9	0	0	2	1	3	1.8×10 ⁴
5.0x10 ⁵	9	0	2	23	20	31	6.0x10 ⁵

Table 9. Comparison of recovery of Salmonella in common carp organs following inoculation and immersion in the presence of the bacteria.

Treatment	Concentration /fish or in water	Exposure days	Muscle /g	Kidney /g	Liver /g	Spleen /g	Peritoneal Fluid /ml	DTC /ml
Inoculation	3.7x10 ⁴		2	2.1×10 ²	5.8x10 ²	 5.5x10 ²	1.4x10 ³	1.4x10 ³
Immersion Inoculation Immersion	2.8×10 ⁴ 1.2×10 ⁵ 5.0×10 ⁵	9	0 18 2	2	1	3	1.0x10 ²	1.0×10 ⁴ 5.0×10 ⁴ 6.0×10 ⁵

Depuration Experiments

After being exposed for eight days to <u>S. montevideo</u> at a concentration of $5.0 \times 10^5/\text{ml}$, common carp were transferred to clean water tanks and kept there for one week. The water was changed daily. The depuration process was quite effective. No <u>S. montevideo</u> were recovered from the organs after the fish were kept for eight days in clean water. In the digestive tract content, although a big reduction was recorded, the bacteria recovered were still at a concentration of $3.0 \times 10^2/\text{ml}$. It should be noted that the concentrations of salmonella in the various organs prior to depuration were relatively low.

Laboratory Experiments Conclusions

The laboratory experiments established the existence of a pattern of penetration of bacteria and bacteriophages into the organs and muscles of fish and helped define and establish threshold values for various organisms prevalent in domestic wastewater under the conditions tested. The threshold concentrations proved to be different for different organisms, being lower for bacteriophages than for bacteria $(10^2 \text{ and } 10^3/\text{fish for } \text{T}_2 \text{ and } \text{T}_4$ bacteriophages, and $10^6 \text{ and } 10^4/\text{fish for } \text{E. coli} \text{ and } \text{S. montevideo}$, respectively). The experiments performed with Polio 1 LSc viruses suggested a very low threshold. As viruses have a low infective dose, their presence in the wastewater and in the fish pond water should be given serious attention.

The inoculation experiments showed also that the threshold concentration for $\underline{E.~coli}$ was higher (2.5 x 10^6 /fish) than for $\underline{C.~freundii}$ (9.3 x 10^3 /fish) or for Salmonella montevideo (1.8 x 10^4 /fish).

Salmonellae were recovered first in the digestive tract, followed by peritoneal fluid, spleen, kidney, and liver. They were recovered from the muscles when the concentration in the water was 5.0×10^5 per ml.

Depuration proved effective only when the concentrations of bacteria in the organs was low.

DISCUSSION

Of the three fish species grown in wastewater treated ponds, tilapia was the hardiest and was able to survive at higher concentrations of pollutants, chemical as well as bacteriological. The recovery of bacteria from the organs and muscles of fish was lower in tilapia than in common carp. Silver carp was the most sensitive.

It has been observed repeatedly that healthy, clean fish do not harbor bacteria in their blood or muscles. Bacteria in small numbers were recovered from the various organs and in high numbers $(10^6-10^7/g)$ from the digestive tract. The presence and concentration of the microorganisms in the fish was determined by their concentration in the pond water.

In our experiments when salmonella were present in the pond water, they were recovered from the digestive tract content. They were not detected in any of the organs of the fish. Coliform and fecal coliform bacteria, although present in the water in high numbers, were not always detected in the muscles of fish, while other bacteria present in the water were recovered. It seems therefore that in the case of fish grown in treated wastewater the role of fecal coliform as indicators for the presence of various bacteria originating from wastewater is questionable.

As a) these fish have been exposed for a long time to treated wastewater which contains various pathogens, bacteria as well as viruses, it seems that the standard which expresses the bacteriological quality should be more stringent than in the case of fish grown in clean water where the pollution occurs usually during the handling and processing, and as b) the coliform and especially the fecal coliforms have proven during our research project not to be adequate indicators, it is proposed that in the case of fish grown in treated wastewater the bacteriological quality should be determined by the examination of the fish muscles for the presence of bacteria.

If the "critical concentration" of the bacteria in the water (between 1.0 and 5.0 \times 10⁴ SPC/ml) and the "threshold concentration" of bacteria in the fish are considered during the growth period, the fish produced should be, from the bacteriological aspect, suitable for human consumption. The bacteriological quality of these fish used for human consumption should be very good, 0-10 bacteria/g; medium quality, 10-30 bacteria/g; more than 50 bacteria/q, not adequate.

Depuration was not effective when bacteria were present in the fish muscles. It was effective when the concentration of bacteria in the organs was low and running water was used.

Although pesticides and heavy metals were tested only in some of our field experiments, their determination should be included in future studies.

The results of the field as well as the laboratory experiments showed that fish can be produced using domestic wastewater effluent. They also showed that the fish produced can be safe for human consumption provided that special attention and consideration is given to the critical concentration of bacteria (not coliform) in the pond water and to the threshold concentration of bacteria in the fish.

Publications arising from the IDRC-supported Waste Reuse Project.

- Buras, N., L. Duek and S. Niv. 1985. Reactions of Fish to Microorganisms in Wastewater. Applied and Environmental Microbiology 50(4): 989-995.
- Buras, N. Microbial Aspects of Fish Grown in Treated Wastewater. Water Research (to be published).

WASTEWATER RECLAMATION (GLOBAL) MALAYSIA

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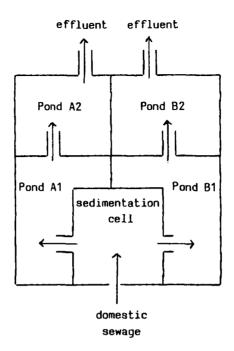
This project was concered with the treatment of domestic sewage and nightsoil in oxidation pends as well as fish production in the wastewater treatment system.

OBJECTIVES

The specific objectives were to:

- study the nutrients and population dynamics of an existing oxidation pond system treating domestic sewage with concurrent production of fish;
- investigate the treatment of nightsoil in series-operated stabilization ponds and the potential for transfer of certain pathogens, heavy metals and pesticides through the system; and
- 3. survey current practices of using nightsoil and other household wastes as feed for small fish culture ponds in rural areas of Malaysia and Indonesia.*

^{*} These survey results are summarized in "The Use of Nightsoil with Household and Agricultural Wastes for Fish Culture in Indonesia, Taiwan, Malaysia, Thailand and Bangladesh", pages 97 to 101.



RESULTS

Research was carried out at an existing stabilization pond system, the Taman Tun Dr. Ismail (TTI) site in Kuala Lumpur which served to treat domestic sewage from a housing development. The pond system consisted of an anaerobic sedimentation cell connected to two primary facultative ponds in parallel (Ponds A1 and B1). The partially stabilized wastewater then flowed into two secondary ponds (Ponds A2 and B2).

The area of each pond was 0.48 ha with a total pond area of 1.92 ha. Water depth for each pond ranged from 120-150 cm.

The project experienced delays in obtaining the ponds for research purposes and problems with flows in the system further decreased the time available for experiments and fish culture.

Section 1. Primary Productivity

Parameters studied included temperature, light intensity, dissolved nitrogen (ammonia, organic-nitrogen, nitrite and nitrate), dissolved phosphorus (orthophosphate and organic phosphorus), dissolved oxygen, chlorophyll "a", the phytoplankton population, suspended solids, the photoautotrophic productivity and respiration.

The pond system was found to have a diurnal limnological cycle with a period of thermal stratification in the day followed by mixing and a period of isothermy at night. During the period of stratification, a high pH was obtained at the surface of the water. The high pH was beneficial in that it aided in removal of phosphorus and nitrogen from solution. During the day, oxygen supersaturation occured in the top half of the pond with levels below saturation in the bottom half of the pond.

Concentrations of orthophosphate, ammonia and organic nitrogen increased with depth while the concentrations of organic phosphorus, nitrite and nitrate decreased. As the water flowed through the pond system the concentrations of orthophosphate and ammonia decreased while concentrations of nitrite and nitrate increased.

A study of the phytoplankton revealed <u>Scenedesmus</u> to be the dominant genus. Other genera present were <u>Oscillatoria</u>, <u>Chlamydomonas</u> and <u>Euglena</u>.

The productivity at the surface was very high and it decreased with depth. Gross productivity measured from 10 to 12 am represented about 32% of the diurnal productivity. Maximum productivity at the surface was 3.9 mg carbon/1/hr. The photosynthetic efficiency of the pond system in the conversion of solar energy to heat energy was 1.34%.

A study of the community respiration showed that zooplankton contributed 36.2%, phytoplankton 15.6% and the bacteria, 48.2%.

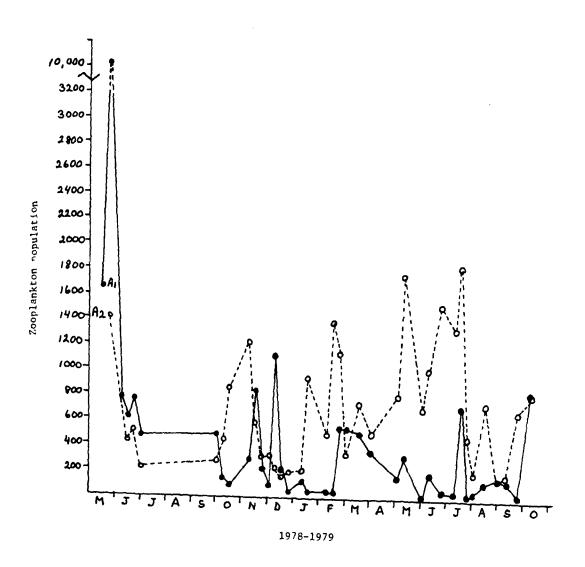
There was a low linear correlation between the concentrations of suspended solids and chlorophyll "a" and no correlation between the concentration of chlorophyll "a" and gross productivity or between the concentration of chlorophyll "a" and the total number of phytoplankton units.

An attempt to culture Chinese carp in the Pond A series was unsuccessful. From 960 fingerlings stocked in Pond A1 and 4800 stocked in Pond A2, only 9 were harvested after 20 weeks. Several reasons accounted for this including oxygen deficiency, high ammonia concentration, escape of fish, poaching and difficulty in harvesting.

It was concluded that since the intensity of the water stratification varies greatly, depending on the thermal stratification which in turn depends on the light intensity,

diurnal depth studies are more suitable for studying oxidation ponds in tropical countries than is taking regular samples at a fixed time and place.

Figure 1: Seasonal variation of the zooplankton population in Ponds A1 and A2.



Section 2. Secondary Productivity

The zooplankton population was sampled by means of a modified Van Dorn Sampler. It was found that seasonal variations occured in the zooplankton population in all four ponds. Figure 1 shows that population peaks occured in Pond A1 in July 1978, November-December 1978, February-March 1979 and July 1979. The secondary ponds, A2 and B2 showed a higher overall zooplankton population than the primary ponds.

Four main species were present in the ponds, i.e. Moina, Cyclops, Brachionus and Asplancha. When a percentage composition analysis was done, it was found that Moina dominated the system. Although this was the case, the percentage composition of the other species was higher in the secondary ponds than in the primary ponds.

Comparison of the dry weight of the zooplankton population gave lower values for the primary ponds than for the secondary ponds, showing that the secondary ponds were more productive than the primary ponds.

A regression equation was developed to estimate the standing crop of zooplankton in the ponds (Figure 2).

$$\hat{w} = 0.8921 + 0.0011N$$

where $\hat{\mathbf{w}} = d\mathbf{r}\mathbf{y}$ weight of zooplankton (mg) per litre of water

N = no. of zooplankton per litre of water

The benthos population of the ponds was found to consist mainly of chironomid larvae. Using boards lowered into the water, a vertical stratification was found to exist with most of the larvae settled in the first 90 cm of board below the water surface. There were none on the bottom of the pond. When Ponds A1, A2 and B2 were stocked with fish, there was a breakdown of stratification with larvae distributed evenly on the plank to the pond bottom. Also, the fish population had an effect on the numbers of chironomid larvae present in the pond.

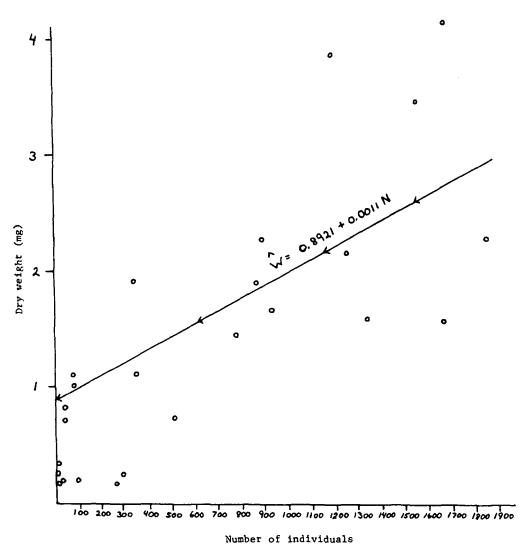


Figure 2: Dry weight-number correlation of the zooplankton population.

Section 3. Fish Farming

Four species of carp, Ctenopharyngodon idella (grass carp), Hypophthalmichthys molitrix (silver carp), Aristichthys nobilis (bighead carp) and Cyprinus carpia (common carp) were stocked in the ponds. Primary pond A1 received 960 fingerlings and secondary pond A2 received 4800 fingerlings. Average size was 1-3g, depending on the species. The fish, however, did not survive.

The raw sewage loading varied from 150 kg BOD_5/day during December 1979, a wet spell, to 334 kg BOD_5/day during May 1979, a dry spell. The mean sewage strength was 242 kg BOD_5/day .

A 6 hour detention time of the raw sewage in the sedimentation cell reduced sewage strength by 51%. Thus the average daily flow of presettled sewage into the primary stabilization ponds was 124 kg BOD₅/ha/day.

Average values for 800_5 (mg/l) in the primary ponds (A₁ and B₁) and in the secondary ponds (A₂ and B₂) of samples taken monthly for 15 months (1978-1979).

	Pond A ₁	Pond A ₂	Pond B ₁	Pond B ₂
80D ₅ (mg/1)				
Maximum :	121	108	126	114
Minimum :	11	11	12	6
Overall Mean:	48	36	49	31

Monoculture of <u>Trichogaster pectoralis</u>, an airbreathing species, was initiated between April and December 1979, in the A series ponds (A1 and A2). Stocking rate was 1 fish/m² for both ponds. Actual number stocked in each .48 ha pond was 4248 in pond A1 and 4692 in pond A2. Survival rate was 14.3% in A1 after 187 days and 71.1% in the secondary pond after 190-252 days. Gross yield in the secondary pond was calculated to be 1368.1 kg/ha/year. These results showed that the primary pond was not suitable for culture of <u>T. pectoralis</u>.

A trial with varying stocking densities of <u>I. pectoralis</u> in the secondary pond B2 showed a rate of 3 fish/m² to be optimum in terms of survival, biomas production and size of fish at harvest. Table 1 gives the results of this trial.

Table 1: Maximum biomass estimation of <u>I. pectoralis</u> cultured in secondary pond effluent at TII.

area	.48 ha
growth period	134 days
planned stocking rate	$3/m^2$
cumulative stocking/acre	12,000
average stocking size (g)	1.0
average size at harvest (g)	81.4
total estimated mortality/acre	2815
percent survival	76.5%
gross yield (kg/ha/yr)	5091.3

Section 4. Bacteriological Study

There was an increase in indicator organisms, i.e. total and fecal coliforms and fecal streptococci between October 1978 and August 1979. This was due to a population increase at the housing estate which was served by the ITI oxidation ponds.

As wastewater flowed through the stabilization pond system, the bacterial population densities were reduced. There was a 25-63% bacterial reduction in the sedimentation cell, a 19-70% (except for fecal coliform) reduction in the primary ponds and about 59-90% decrease in bacterial population in the secondary ponds. For the whole TTI stabilization pond system, the overall efficiency in lowering the standard plate count (SPC) was 96% and removal efficiency for total and fecal coliforms and fecal streptococci was 99%. This reduction was achieved by a 4.3 day detention time for the whole system and at a daily range of water temperatures of 27-34°C.

Fish rearing in the ponds did not have a significant effect in reducing the bacterial population in the wastewater. This is in contrast with the laboratory experiment in Israel where fish introduced into 10 1 aquaria were shown to have a significant effect on the extinction rate of \underline{E} , coli (p. 55).

Examination of <u>I. pectoralis</u> reared in the secondary pond A2 showed that there was an increase in bacterial population densities in the flesh, gills and viscera. The increase in bacterial population densities in the flesh indicated systemic infection.

Depuration of the fish in clean tap water for a 2 to 3 week period reduced most of the population densities in the four bacterial types (SPC, TC, FC and FS) in the flesh, gills and viscera. However, fecal coliform contamination of the flesh increased during depuration, indicating that possibly FC had multiplied in the flesh.

Section 5. Parasitological Study

A total of 90 water samples were taken from the sewage ponds at ITI for analysis of helminth parasites. Results showed that helminth ova were present in the raw sewage, the sedimentation cell and the primary ponds. As well, primary pond A1 was found to contain ova of Ancylostoma and Trichuris. The secondary ponds and their effluent were negative for helminth ova.

Results also showed that some of the viscera from fish grown in the primary ponds were positive for ova of <u>Ascaris</u>, <u>Taenia saginata</u>, <u>Enterobius</u> and <u>Hymenolepis</u> and one specimen contained an <u>Acanthocephala</u> worm. Although helminth ova were not found in the water of the secondary ponds, examination of fish from these ponds revealed the presence of parasite ova.

Section 6. Toxic Metals

From August to November 1979 a total of 31 wastewater samples were collected from the raw sewage, primary and secondary ponds and the effluent and analyzed for selected inorganics. Four of the metals studied, arsenic, cadmium, lead and mercury have a health significance

Table 2: Concentration of selected inorganics in wastewater samples.

Metal	Range	Mean	Recommended levels in drinking water, WHO 1984 (mg/l)
Inorganics wit	h health significance		
Arsenic	D.M.* 0.008-0.025 ppm	0.019 ppm	0.05
(As)	T.M.* 0.015-0.143 ppm	0.061 ppm	
Cadmium	D.T. 0.001-0.006 ppm	0.002 ppm	0.005
(Cd)	T.M. 0.016-0.038 ppm	0.026 ppm	
Lead	D.M. 0.004-0.030 ppm	0.012 ppm	0.05
(Pb)	T.M. 0.01 -0.17 ppm	0.06 ppm	
Mercury (Hg)	T.M. 0.01 -1.80 ppb	0.26 ppb	0.001
Inorganics aff	ecting aesthetic quality		
Copper (Cu)	T.M. 0.012-0.310 ppm	0.059 ppm	1.0
Iron	D.M. 0.31 -7.36 ppm	1.08 ppm	0.3
(Fe)	T.M. 0.31 -8.10 ppm	1.19 ppm	
Zinc	D.M. 0.06 -9.0 ppm	0.26 ppm	5.0
(Zn)	T.M. 0.08 -2.20 ppm	0.27 ppm	

^{*} D.M. = dissolved metal

^{**} T.M. = total metal

and three, copper, iron and zinc affect mesthetic quality. Results of the heavy metals analysis are shown in Table 2 along with recommended levels for these metals in drinking water taken from the WHO 1984 "Guidelines for Drinking Water Quality" for comparison.

Generally, the amounts of toxic and non-toxic metals in the wastewater are considered low and within acceptable levels. Levels for iron and for zinc in some samples were high. The highest concentration of the metals was found in the primary pond, followed by levels in the raw sewage.

The flesh and internal organs of four fish samples taken on two different dates were snalyzed for the presence of mercury, cadmium, zinc, copper, iron and lead. Tables 3 and 4 show the results of the heavy metals analysis.

Table 3: Total mercury in fish samples.

Sample	Sample Tissue (ug Hg/g)		Average fish size (g)	
No. 32 Aug/79	0.094	0.130	-	
Pond A1 Jan/80	0.078	0.080	60	
Pond A2 Jan/80	0.148	0.135	100	
Pond B2 Jan/80	0.137	0.115	75	

Table 4: Selected heavy wetals in fish muscles and in internal organs.

Cu nd*	Cd	Zn
	İ.	
no-	nd	5.9
nd 0.23	nd nd	5.4
1.02	nd	6.0
0.26 0.62 0.96	0.62 1.02	0.62 1.02 nd
	0.23	0.23 nd 1.02 nd

*nd = not determined

With the exception of iron and zinc, levels are generally low.

DISCUSSION

The results presented in the report cover a relatively short sampling period due to delays in project startup. As well, they show the difficulties experienced when using an existing wastewater oxidation pond for fish culture as manipulation in BOD loading could not be performed and harvest was difficult as the ponds could not be emptied and were deep. This points out the need for integrating the design of waste treatment with the needs of aquaculture if the two are to function together. Due to delays and difficulties in obtaining the vinylon fish tanks for the nightsoil experiment, only a preliminary report was available and results are not reported here.

The study showed that the TTI oxidation pond system was efficient in the removal of orthophosphate (75%) and total nitrogen (70-80%) from solution. These nutrients are converted to phytoplankton and not completely removed from the pond effluent.

The secondary ponds were found to be more productive (in terms of biomass) than the primary ponds and exhibited a lower total bacterial population. Water samples taken from secondary ponds were free of parasite ova and had lower concentrations of heavy metals than the primary ponds.

The primary ponds were found not to be suitable for fish culture. The secondary ponds were suitable for culture of an air-breathing species, <u>Trichogaster pectoralis</u>. However, losses occured with the escape of fish through the outlet drain and by poschers. Harvesting was difficult.

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EVALUATION OF THE SAN JUAN STABILIZATION PONDS (PERU)

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In the semi-arid coastal areas of Peru, water shortages exist and the situation is expected to worsen in the future. Peruvian authorities recognized the need to consider domestic wastewater as a water resource and looked to the reuse of treated sewage for agricultural purposes along the desert coast of Peru as a necessary solution to the problem.

From a survey of sewage irrigation sites in Peru it was found that the most used treatment process for wastewater prior to irrigation was the facultative stabilization pond. The prevalence of enteric diseases in the area, along with the appearance in the environment of enterobacteria such as <u>Salmonella</u> serotypes which were resistant to antibiotics, pointed to the need for a thorough evaluation of the stabilization ponds.

OBJECTIVES

Although the San Juan stabilization ponds have been in use in Lima for over 15 years, their performance had not been properly evaluated. Therefore the present study was designed to:

 a) evaluate the performance of the San Juan stabilization ponds under a variety of loading conditions (including variations in climatic conditions and waste water quality) in

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order to develop design criteria for this type of process,

- b) study the health implications of reusing pond-treated sewage in irrigation, through analysis of the survival of enteric pathogens,
- c) use the design criteria developed to study the economic feasibility of treating Lima's domestic wastewater discharged to the southern interceptor, as well as to determine the economic impact of large-scale fish farming in sewage lagoons.

RESULTS

1. Sewage Characteristics and Design Criteria

Of the San Juan Lagoon complex, eight waste stabilization ponds (four primary and four secondary) were isolated for study. Figure 1 shows the arrangement of the four primary lagoons, P7, P8, P9 and P10 which were set to receive loads of 400, 600, 800 and 1000 kg B00/ha/day, respectively and the four secondary ponds (S5, S6, S7 and S8). Table I shows the area, depth and B00 loadings of the experimental ponds. Total raw sewage flow was measured continuously by a Parshall flume and flow distribution was controlled by three calibrated rectangular weirs. Nine points were selected for wastewater sampling - raw sewage and the eight pond effluents.

Figure 1: Pond Arrangement.

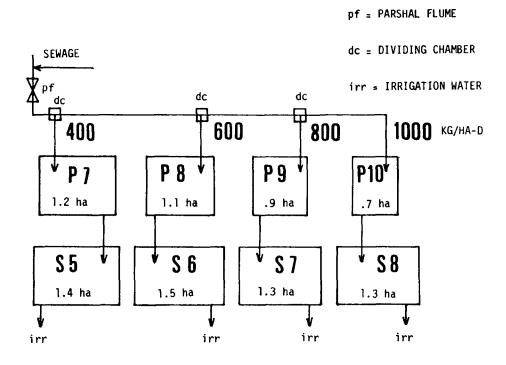


Table 1: Area, depth and BOD loading of ponds.

POND	AREA Ha	AVERAGE WATER DEPTH meter	ESTIMATED 80D LOADING kg/ha/day
P7	1.20	1.30	400
P8	1.10	1.30	600
P9	0.88	1.30	800
P10	0.69	1.30	1000
S 5	1.44	1.60	
S6	1.49	1.30	
S7	1.30	1.30	
58	1,30	1.30	

The experimental phase of the study lasted 21 weeks and included the coldest months, April to August. A large number of physical, physico-chemical, biochemical and microbiological parameters were selected for study. BOD and COD were the main parameters used for performance evaluation. The BOD of the raw sewage ranged from a low of 112 mg/l to a high of 197.2 mg/l with an average of 158.5 mg/l, which is a somewhat weak domestic sewage.

Table 2 summarizes the applied and removed BOD loads for the four primary and four secondary ponds.

Several correlations of removed BOD load versus applied BOD load were determined. These are important for the development of design criteria for waste stabilization ponds for the critical climatic conditions along the Peruvian coast. The coastal area of Peru is semi-arid with average yearly precipitation of about 15 mm. During the months of the study (April, May, June, July and August, 1979), rainfall averaged 1.4 mm/month and temperature averaged about 17.2°C.

Regression equation for primary ponds: Lr = 7.67 + 0.8063 La

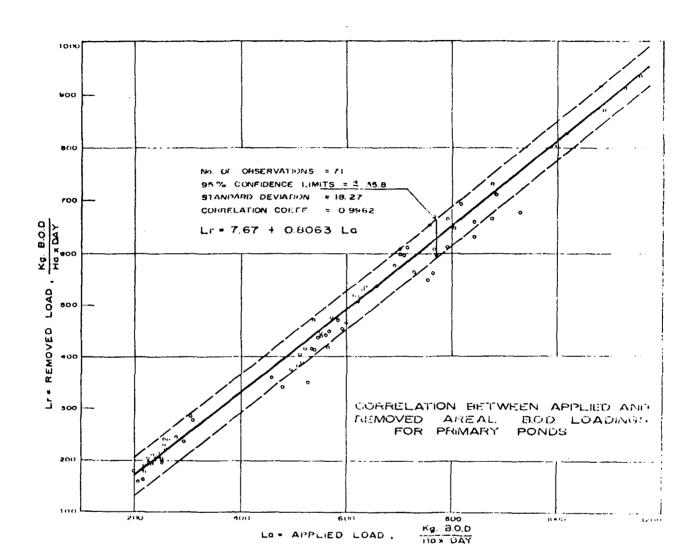
where: Lr = aereal BOD load removed

La = aereal BOD load applied

Table 2: Applied Loads (Based on Total BOD) and Removed Loads (Based on Soluble BOD) (kg BOD/ha/day).

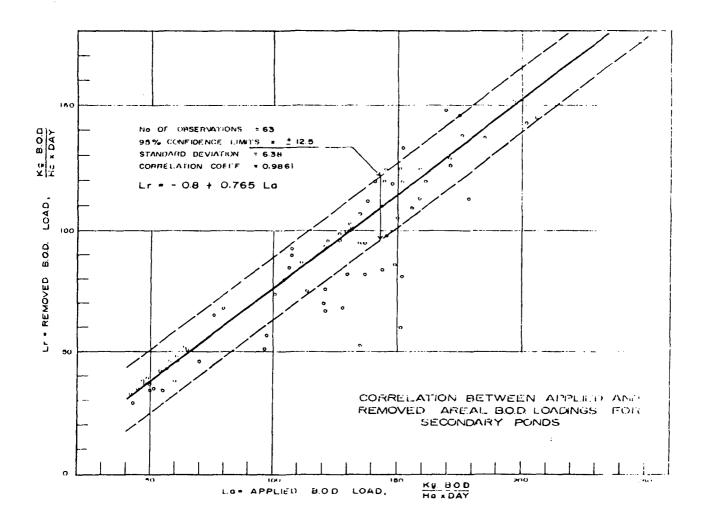
	Kaw Sewage	APPLIED LOADS					KEMOVEL IDANS										
Week No.	Kg BOD	r	RTMARY	PONDS		SE	CONDAR	Y POND			PRIMA	RY PONI);;	SH	CONTAK	a rom	95
	Day	۲7	P8	1'9	P10	55	sc	57	58	17	1'8	119	P10	55	86	57	:.8
1	1364.4	200	257	459	635	54	58	107	152.	171	231	36.1	535	42	46	85	! 133.
2	1525.2	224	287	513	710	47	70	108	145.	203	244	400	1597	38	46	(41)	(134)
3	1641.7	240	309-	553	764+	80	108	96	121.	201	281-	447	564+	68	93	52.	67.
4	2283.2	265-	516	758-	1081-	64	121	152	122	243-	389	655-	918+	52	94	120	96
5	1 1		1	ł		1	{	i			1	}	1	i .	i	•	•
6	2387.7	279	539-	792	1130	65	127	120.	171	248	474-	665	912	51	94	70.	124
7	2546.3	295	692	772	1090	60	144.	135+	149.	23/	580	639	872	38	84.	5.14	1 800
8	2092.7	252	569	629	882	55	145	114	150	201	452	510	1/12	34	120	11.	105
9	1862.7	221	507	563.	792	61	159	156	151-	163	385	422.	613	46	1113	1 1000	į 100 s
10	2063.7	246	562	622	874+	45	136	105	152+	209	44.4	518	668+	35	95	101	. 81.
11	1910,9	230	521	573	804	48	127	97.	137.	196	416	476	649	37	96	57.	H2.
12	2072.4	209	481+	534	751+	61	159	135	185	159	343+	417	うらいチ	46	125	45	1.07
13	1981.8	235	541	600	844	52	132	130.	161	194	416	466	666	35	101	H2.	120
14	2339.8	261	633	715-	1017	43	151	12H.	197	723	533	612-	826	29	125	lin.	152
15	2159.7	249	586	655	927+	50	135	121.	171	206	472	537	680+	34	107	76.	126
16	1818.3	21H	495	546	767	50	131	112	137	179	378	438	610	3/	103	87	45
17	2029.8	235	550	616	871	42	141	101	131	212	446	516	/33	33	120	14	100
18	2675.6	306-	728		1158	57	175	144	202	286-	566		1936	4)	146	110	143
19	2289.3	249	617	-	1004	61	138	130	178.	217	515	607-	1119	48	112	100	113.
20	2700.5	25/-	622	703-	1000	49	169.	148	206	242-	50R	600-	803	39	148.	1110	145
21	1944.3	220	526+	592	840+	76	176	146	248	191	353+	455	636+	65	138	98	1/"
VERAGE.	2064.8	244.6	526.6	635.7	897.1	56.0	135.1	124.2	163.3	209.1	 421.3	522.1	715.0	42.5	105.6	85	114

Figure 2: Correlation Between Applied and Removed
Aereal BOD Loadings for Primary Ponds.



Regression equation for secondary ponds: Lr = -0.8 + 0.765 La

Figure 3: Correlation Between Applied and Removed Areal 80D Loadings for Secondary Ponds.



Regression equation for combined BOD performance data:

Lr = -7.81 + 0.8193 La

Figure 4: Correlation Between Applied and Removed Areal BOD Loadings for Primary and Secondary Ponds.

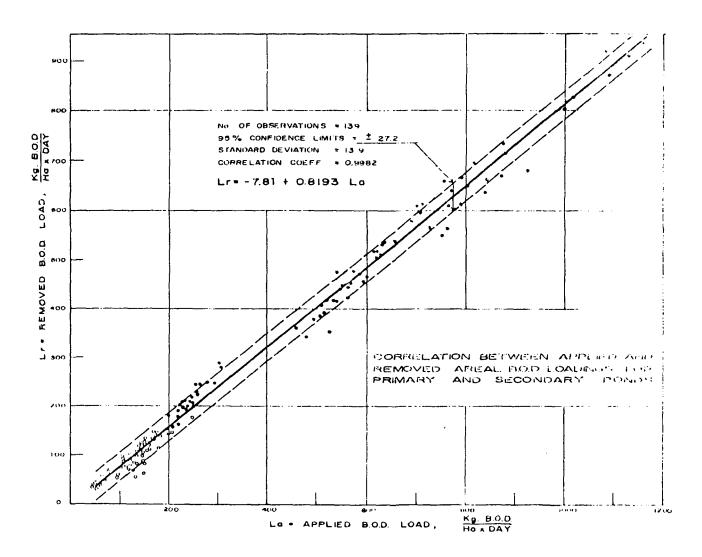
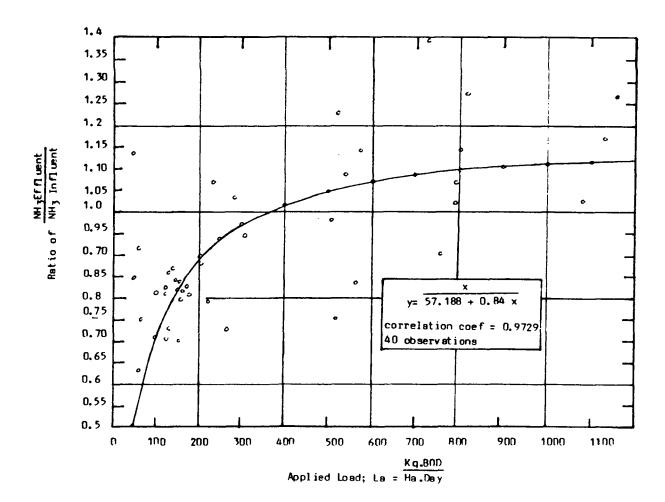


Figure 5 shows the correlation of the ratio of effluent to influent ammonia vs. applied load (at 20°C). The ammonia fraction is less than one for loading rates up to about 350 kg B00/ha/day, thus this load represents an upper limit between facultative lagoons and pre-

dominantly anaerobic lagoons. Subsequent data analysis yielded the following design equation for maximum loading of facultative lagoons as a function of water temperature:

$$La = 357.4 \times 1.085^{T-20}$$

Figure 5: Fraction Ammonia Present in Effluent Vs. Load



Correlations between the ratio of total to soluble 80D in lagoon effluents against applied load were developed in primary lagoons. For loads ranging from 450 to 1150 kg 80D/ha/day, the relationship has a high correlation coefficient and indicates an estimated constant ratio of 1.7. At lower loading rates in both primary and secondary ponds this ratio tended to be consistently higher, due to the greater and more diversified algal populations, but no significant correlation was found with applied loadings.

It was found that the ratio of soluble COD to BOD increased with increased degree of treatment, i.e. 1.75 for raw sewage; 3.0-4.5 for primary pond effluent and 3.3-6.0 for secondary pond effluent.

Counts of the main algal species in the primary and secondary pond effluents were determined. It was found that in general, the higher the load on the ponds, the more reduced the number of genera present. The presence of <u>Chlamydomonas</u> was associated with higher loads. Table 3 shows the algae counts in the primary and secondary lagoons for the 21 weeks of the study.

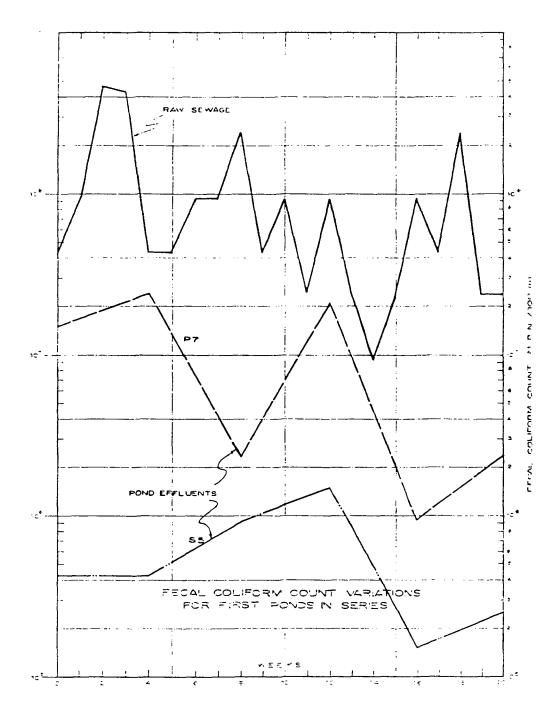
Table 3: Total Algae Population in Cells/ml x 10⁻³

Week		Primary E	ffluents		Secondary Effluents					
WEEK	Р7	Р8	Р9	P10	S 5	56	\$7	58		
1	79.718	-	-	-	114.299	_	_	-		
2	_	31.387	- '	-	-	32.081	-	-		
3	127.076	37.498	2.5	2.778	213.603	68.206	15.277	10.138		
4	140.146	33.609	65,136	31.943	34.86	43.749	36.386	67.357		
5	40.415	2.361	102.910	81.939	147.075	50.552	73.051	68.33		
6	11.805	25.744	39.025	7.917	425.668	41.437	125,409	51.941		
7	4.306	122.354	44.442	54.024	132.492	21.665	462.9	56.663		
8	18.610	50.691	86.522	86.939	59.023	17.777	311.507	72.217		
9	44.858	92.217	86.078	94.438	187.627	91.522	390.808	41.248		
10	67.218	24.303	69.579	9.166	154.573	99.577	259.289	27.36		
11	85.828	90.550	115.410	54.858	165.685	132.631	414.706	43.47		
12	144.713	83.328	119.176	37.498	166.516	121.103	310.536	27.138		
13	103.743	90.411	27.082	90.689	33.470	372.476	113.743	127.631		
14	131.797	66.551	78.606	64.301	76.522	171.933	101.798	83.328		
15	644.542	68.051	135.547	2.222	189.016	98.864	62.496	18.055		
16	124.199	79.717	65.274	6.250	179.85	106.382	67.357	16.943		
17	1708.224	0.417	81.106	39.720	126.798	562.464	247.762	64.856		
18	1486.016	19.443	139.297	32.081	128.742	206.793	6883.872	88.327		
19	2250.134	50.691	224.430	35.414	133.186	471.359	413.863	120.548		
20	1736.278	765.501	151.379	40.414	167.212	336.368	521.772	134.297		
21	1139.372	120.687	139.019	58.885	209.212	450.11	690.373	137.769		

2. Public Health Aspects

a) Fecal coliform counts: A decrease in fecal coliform counts was generally observed from the raw sewage to primary pond effluent to secondary pond effluent. However, secondary pond effluents still had fecal coliform counts in the range of 10² to 10⁷/100ml. Figure 6 shows the fecal coliform counts in the effluents from the primary pond P7 and secondary pond S5 series.

Figure 6: Fecal coliform count variations for the first ponds in the series.

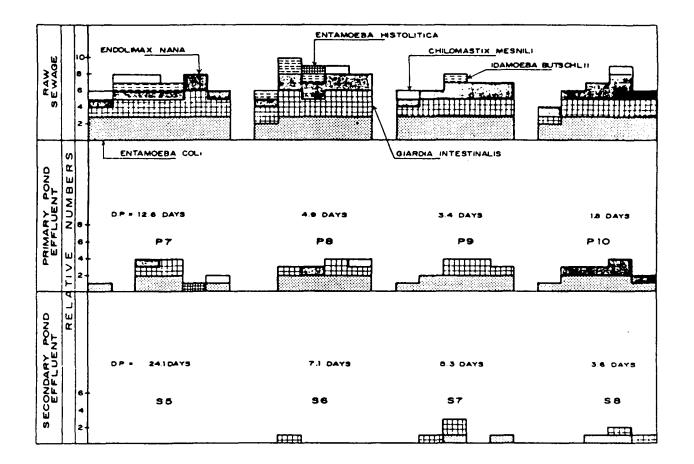


b) Pathogenic protozoa and helminths: Although the information is largely qualitative, it can be concluded that for complete removal of protozoa and helminths in ponds with loads in the range of 200-300 kg 80D/ha/day, more than a primary pond is required. Complete protozoa removal was only achieved in the pond series P7/S5 which had a maximum B0D loading on lagoon P7 of around 310 kg B0D/ha/day. Complete removal of helminths was achieved in the secondary ponds for all the load ranges encountered in the study. A pond series with a total detention period of 36.7 days was satisfactory for complete removal of protozoa under the Peruvian conditions.

TRICHURIS STRONGYLOIDES STERCORALIS HYMENOLEPSIS NANA RAW SEWAGE ASCARIS LUMBRICOIDES NUMBERS PRIMARY POND D.P . 12.6 DAYS 4.9 DAYS 3.4 DAYS 1.8 DAYS P7 29 P 10 TIVE ٥ SECONDARY POND A L α 5.3 DAYS 3.6 DAYS 24.1 DAYS 7.1 DAYS COMPLETE REMOVAL **S**7 **S**B **S**5 96

Figure 7: Presence of Helminths in Raw Sewage and Pond Effluents.

Figure 8: Presence of Pathogenic Protozoa in Raw Sewage and Pond Effluents.



c) Salmonella Species: Nine serotypes of <u>Salmonella enteritidis</u> were isolated. All primary and secondary lagoon effluents tested were positive for <u>Salmonella</u> showing that stabilization pond treatment does not completely destroy <u>Salmonella</u> species. Paratyphi B and Derby were the serotypes most often isolated and these have been found to have developed antibiotic resistance.

DISCUSSION

The need to reuse treated domestic wastewater for agricultural purposes in coastal Peru led to the study of the San Juan stabilization ponds. Performance evaluation based on the BOD and COD of four primary ponds and four secondary ponds for a 21 week period and under a variety of loading conditions allowed the development of design criteria for the treatment process.

Three correlations for the removed BOD load versus applied BOD load were developed for primary and secondary ponds and a combined pond system. The correlation for primary ponds covered a range of BOD loading from 200 to 1158 kg BOD/ha/day. The correlation for the secondary pond covered a range from 40 to 210 kg BOD/ha/day. In the above equations, the applied load was based on total BOD and the amount removed was based on dissolved BOD. A maximum loading rate was found for facultative ponds based on water temperature.

Fecal coliforms decreased as raw sewage was treated in the primary and secondary ponds but removal was not complete. Helminth removal was achieved in the secondary ponds while protozoa removal was not consistently achieved, particularly at higher BOD loadings. The detention time in the ponds under study was about three times those reported by other investigators.

The two-stage treatment did not destroy Salmonella species. Their persistence during stabilization pond treatment pointed to a need to reassess pond loading conditions, detention time and pond effluent use.

Publications arising from the IDRC-supported Waste Reuse Projects.

- Yanez, F. 1982. "Manual de métodos experimentales. Evaluación de Lagunas de Estabilización". Serie Técnica de HPE No. 24, 181 p. CEPIS, OPS-OMS, Lima.
- Yanez, F. 1982. "Avances en el tratamiento de aguas residuales por lagunas de estabilización". Serie de Documentos Técnicos No. 7, 58 p. CEPIS, OPS-OMS, Lima.

Additional Note

The research described here constituted phase one of a two part study. Additional important work was done on the San Juan Stabilization Ponds after the IDRC-supported project was completed. The main purpose of the second phase of research was to investigate the reduction of pathogenic organisms through a series of waste stabilization ponds with the objective of developing practical information for the design of ponds in tropical conditions and for the large-scale agricultural reuse of treated effluents. The results of this phase are reported in the following publications:

- Yanez, F. "Indicator and Pathogen Organisms Die Off in Ponds and Design Under Tropical Conditions", paper submitted to W.H.O./Geneva for publication, April 1984.
- Yanez, F. "Reduccion de organismos patógenos y diseño de lagunas de estabilización en países en desarrollo". Paper presented at the XIX AIDIS Congress, Santiago, Chile, 1984.
- 3. Bartone, C. "Reuse of Waste Water at the San Juan Stabilization Ponds Public Health, Environmental and Socioeconomic Implications". Paper presented at the A.W.W.A. Water Reuse Symposium III, San Diego, California, August 1984.

THE USE OF NIGHTSOIL WITH HOUSEHOLD AND AGRICULTURAL WASTES FOR FISH CULTURE IN INDONESIA, TAIWAN, MALAYSIA, THAILAND AND BANGLADESH

M.G. McGarry* and Lee Kam Wing**

As a part of the IDRC-supported Wastewater Reclamation (Global) project, surveys of night-soil use along with household and agricultural wastes in fish culture were conducted. Countries included in the survey were: Bangladesh, Indonesia, Malaysia, Taiwan and Thailand. Below is a brief summary of the findings.

The objectives of the survey were to:

- a) determine the extent of nightsoil use for fish culture;
- b) assess fish culture operations in terms of yield, costs and benefits;
- c) identify potential problems in the use of nightsoil in fish ponds.

The study was carried out by investigators from institutions of the five countries. Researchers from the following departments or institutes were involved:

- a) Inland Fisheries Research Institute, Indonesia
- b) National Inland Fisheries Institute, Thailand
- c) Directorate of Fisheries, Ministry of Fisheries, Banqladesh
- d) Taiwan Institute of Environmental Sanitation, Taiwan
- e) Botany Department, University of Malaya, Malaysia

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The researchers used a common survey instrument to gather information on fish farm practices. They also gathered information by direct observation of the fish farm facilities and waste reuse systems, interviews with local leaders and persons involved in local fishery and sanitation programmes and by informal conversations with fish dealers and sellers.

Indonesia

Fifty-six thousand tonnes of fish are produced yearly in freshwater ponds throughout Indonesia. Nightsoil fed fish ponds are mainly small scale, backyard or village ponds, often fertilized by overhanging latrines.

Agricultural or household wastes are often added to the fish ponds. These ponds are also frequently used as a source of water for washing, and, in a more limited number of instances, as a source of water for cooking and drinking.

Most fish farmers practice polyculture. Common species used are tilapia, nile carp, java carp, common carp, kissing gouramy, giant gouramy and a sepat siam. Sometimes common carp is raised alone.

Placing latrines directly over the fish ponds is a common practice in rural Indonesia. Usually the wastes are discharged directly into the pond, without the benefit of pretreatment or storage. Ideally the system should be upgraded, perhaps by using a Chinese 3-tank system which allows for a storage time of the excreta prior to discharge to the fish pond. Any improvement should, however, be appropriate, readily acceptable and affordable by the local population.

Taiwan

The technique of waste reuse for fish culture has been relatively well developed in Taiwan. Nightsoil is used extensively in both brackish water and freshwater ponds. Often, treated or composted nightsoil is used, showing a regard for public health. During winter, when the ponds are drained, nightsoil, alone or in combination with agricultural wastes, is spread over the bottom of the ponds, providing some detention time prior to the introduction of water.

Over 30,000 tonnes per year of tilapia and carp are produced in the freshwater ponds. The natural food (phytoplankton) produced by the ponds is frequently supplemented by traditional feeds to increase fish production. Indeed, many farmers now combine fish farming with the raising of pigs or ducks which supply excreta to fertilize the ponds.

In the past decade there has been a general decline in nightsoil reuse because, as income rises, farmers purchase inorganic fertilizer.

Malaysia

Fish farmers in West Malaysia, mainly ethnic Chinese, have adopted the traditional concepts of fish culture from their country of origin, i.e. polyculture, utilization of farm wastes and nightsoil and a low level of capital and technological input. Indian and Malay fish farmers rarely fertilize their fish ponds, and do not use human or piggery wastes due to religious and health reasons.

Most traditional fish farms, as opposed to government assisted fish farms, are family owned and operated. Due to their low capital input and recurrent costs, production is low and little desire exists to optimize the system.

The species reared are bighead carp, grass carp, mudcarp, <u>Puntius javanicus</u> and <u>Oxyeleotris</u> <u>marmoratus</u>. For most farms, the addition of nightsoil is unplanned and incidental with no attempt to treat it.

In recent years, the reuse of nightsoil in agriculture and aquaculture has been decreasing due to discouragement by local health authorities, the low supply of nightsoil as a result of better waste collection and disposal systems, the ability to purchase inorganic fertilizer and the availability of sufficient piggery wastes.

Thailand

The direct use of nightsoil for fish culture in Thailand has declined significantly through the years. Human excreta does, however, find its way into the fish ponds by a more indirect route via the waterways. Some of the nightsoil collected in Bangkok is composted with agricultural wastes and used as compost fertilizer by vegetable farmers.

At the time of the survey, the Department of Fisheries was making a significant effort to develop aquaculture, especially freshwater pond culture. A major problem, however, had been insufficient fish feed.

Bangladesh

Improper sewage disposal was found to be a serious problem in Bangladesh. Wastes are indiscriminately discarded, leading to insanitary conditions and serious health problems.

The direct application of nightsoil to fish ponds is not practiced due to social, religious and practical reasons. However, human wastes find their way into fish ponds indirectly and in an unplanned way, either during flooding in the rainy season or as a convenient place to dispose of household wastes.

Most ponds serve the dual function of producing fish and water for domestic purposes such as bathing and washing. Many small, backyard ponds are stocked by wild fish that find their way into the ponds. Larger farms stock purchased fries of mainly the following species: Labeo calbasu, Labeo robita, Catla catla, Cirrhina mirigila and Cyprinus carpio.

Ninety percent of the farms don't use fish feeds but fertilize their ponds with agricultural wastes. The small, backyard ponds are harvested continuously. As a result, production is low, up to 600 kg/ha/yr in unfertilized ponds and up to 1625 kg/ha/yr in fertilized ponds.

It seems that due to the rising costs of fertilizers and fish feed, farmers are increasingly interested in recycling their household wastes, provided that some treatment process is included.

Discussion and Conclusions

Traditional fish farming has often been used as a means of supplementing the family protein intake as well as an income supplement. Often fish ponds are small and fish raising is not the only activity. The ponds may be used for other domestic activities besides the raising of fish.

In the five countries surveyed, yields varied from 200 kg/ha/yr to 2000-3000 kg/ha/yr. The use of nightsoil for fish culture is extensive in both Taiwan and Indonesia and more haphazard and incidental in other countries.

The use of nightsoil in aquaculture is closely tied up with religious, cultural and social customs and beliefs. Any attempt to introduce recycling schemes should be conscious of these factors and should incorporate some form of treatment but bear in mind that treatment usually means added cost.

Since there was a trend observed towards decreased nightsoil use with increases in income, it is likely that countries with relatively low per capita incomes would be more receptive to low-cost reuse technologies.

WASTE REUSE (KOREA)

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Korea's population increase coupled with the growth of cities due to urbanization and industrialization called for a need to reexamine both solid waste and nightsoil disposal. This had to be done with a view to safeguarding public health and the environment in such a way as to make safe treatment economically feasible.

OBJECTIVES

The IDRC-sponsored project carried out by researchers from the Department of Sanitary Engineering at Dong-A University in Busan, Korea, proposed to study the composting process of a mixture of domestic solid waste and nightsoil to form an acceptable and reusable humus product for use in agriculture. To achieve the general objective, the following specific objectives were set:

- a) to quantify and analyse typical South Korean urban and rural community nightsoil and refuse.
- b) to conduct composting tests of several variables under controlled conditions,

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- c) to carry out prototype experiments under field conditions,
- d) to perform agricultural trials using the night soil/refuse,
- e) to design a full-scale collection, treatment and reuse scheme for the rural town of Gimhae, and
- f) to conduct a cost-effectiveness analysis of the overall system.

RESULTS

1. Solid Waste Characteristics

Both the quantity and quality of the solid waste generated by the city of Busan were determined, as noted in Tables 1-4 below.

Table 1. Specific Refuse Weight, Density and Volume for the cities of Busan and Kimhae, Korea and Tubingen, Germany.*

	Season		Busa	an**	Kimhae**	Tuebingen	
_	Season	1972	1979	1982	1984	1979	1970
Refuse Weight kg/person/day	Winter Summer Average	0.694	0.880 0.720 0.800		1.500 1.274 1.387	0.610	0.40 0.30 0.35
Refuse Volume litre/person /day	Winter Summer Average	1.521	2.056 1.748 1.905		3.947 4.110 4.028	1.466	0.79 0.73 0.76
Refuse Density kg/litre	Winter Summer Average	0.456	0.428 0.412 0.420		0.380 0.310 0.345	0.416	0.51 0.41 0.46

^{*} The IDRC-supported project covered the period 1977-79. The researcher provided additional data for 1982 and 1984.

^{**} Included anthracite ash.

Table 2. Seasonal Composition of the domestic refuse of the city of Busan for 1972-1984.*

Year		1972	1977	1978	1979	1984
Investigated		by PEB	by Y00	by KIM	by KIM	by KIM
Composition	Season	%	%	%	%	%
Ashes	Winter	85.52		83.60	L	l '
	Summer Average	75.22 80.37	80.80	68.00 75.80	65.20 73.64	44.10 58.25
Iron	Winter	0.40		0.30	1	
	Summer Average	0.57 0.49	0.40	0.30 0.30	0.32 0.30	0.80 1.00
Glass	Winter Summer	0.52		0.50 0.70	0.51 0.61	0.80
	Average	0.58	0.50	0.60	0.56	0.90
Stones	Winter	2.15 3.36		2.10 2.70	i .	1
	Summer Average	2.76	-	2.40	2.50 2.30	1.20 1.55
Compostable	Winter	9.88		11.50		18.20
	Summer Average	17.05 13.46	15.90	24.30 17.90	27.57 20.10	44.80 31.50
Combustible	Winter	1.53		2.00	2.40	5.50
	Summer Average	3.15 2.34	2.40	4.00 3.00	3.80 3.10	8.10 6.80
Total	Winter	100.00		100.00		
	Summer Average	100.00	100.00	100.00 100.00	100.00	100.00

^{*} The IDRC-supported project covered the period 1977-79.
The researcher provided additional data for 1984.

Table 3. Percent composition of domestic refuse for Busan and Kimhae in 1972-1984.*

City	Year	Ash, Soil	Glass	Rubber, Vinyl and other Combustibles	Iron	Compostable materials such as paper, vegetables, etc.	Total
	1972	83.13	0.58	2.34	0.49	13.46	100.00
Busan	1977	80.80	0.50	2.40	0.40	15.90	100.00
busan	1978	78.20	0.60	3.00	0.30	17.40	100.00
1	1979	75.94	0.56	3.10	0.30	20.10	100.00
{	1984	59.80	0.90	6.80	1.00	31.50	100.00
	1978	79.00	0.55	2.80	0.30	17.35	100.00
Kimhae	1979	77.50	0.57	2.90	0.30	18.73	100.00
	1984	57.70	0.90	8.70	1.00	31.70	100.00

^{*} The IDRC-supported project covered the period 1977-79. The researcher provided additional data for 1984.

Table 4. Chemical properties of urban refuse collected in 1978-79 in the cities of Busan and Kimhae, respecting the composting of nightsoil and urban refuse.

	Summer	Winter	Spring/Fall
Busan			
рН	7.5	8.5	7.8
Water content (%)	24.5	14	18
Ignition loss rate p. TS(%)	20.0	10	14
TOC p. TS(%)	9.9	4.7	6.7
Total N per TS(%)	0.183	0.157	0.18
C/N	54	30	37
Kimhae			
рН	7.5	8.7	7.9
Water content (%)	24	14	18
Ignition loss rate p. TS(%)	21	10	13.5
TOC p. TS(%)	10.4	4.7	6.5
Total N per TS(%)	0.179	0.152	0.181
C/N	58	31	36

The water content of Korea's urban solid wastes varied from about 15% in winter to about 25% in summer. Its high density was due to a greater proportion of ashes. In winter, the low organic matter content made it unsuitable for composting.

2. Nightsoil Characteristics

Korean adults were found to produce an average of 1,370 g/d or 1.35 l/d of nightsoil. However, amounts collected were lower, ranging from 0.4 l/d to 0.6 l/d due to incomplete collection. Table 5 compares the chemical composition of nightsoil in Japan and Korea.

3. Composting Experiments Under Controlled Conditions

To find the most effective composting method, aerobic and anaerobic experiments were carried out under controlled conditions. A mixture of nightsoil and solid waste equivalent to the amount generated per person was used.

It was found that in terms of temperature increase, pathogen removal and odour control, the aerobic method was superior to the anaerobic method. A water content of less than 60% was necessary to maintain sufficient heat in the compost pile. The lower the water content, the more rapid the initial heat generation.

A C/N ratio of about 30 is desirable to maintain high enough temperatures for the required time. This value was generally achieved when nighsoil and refuse were mixed. An experiment was performed comparing crushed and non-crushed refuse and the results showed that it was not necessary to crush the refuse.

Composting performed during spring, summer and autumn was able to maintain an internal temperature of 55°C or greater for more than 10 days to ensure destruction of pathogens. However, during the winter months, it was difficult to maintain a temperature of 55°C. The addition of organic materials (2% by weight) in the form of agricultural wastes such as rice or barley straw, rice bran, etc. or the covering of the compost pile by glass or ripened compost (as was done under laboratory conditions), allowed sufficiently high temperatures to be maintained.

Table 5. Chemical composition of community nightsoil in Korea and Japan.

					Busan				
	Item		Japan	Korea	1978 Summer	1978 Autumn	1978 Winter	1979 Spring	Range
1	рН		7-9	7-8.5	8.5	7.5	8.6	7.7	7.5-8.6
2	Water Content	%	79	96-98	96.04	95.46	96.98	96.24	95.46 -96.98
3	Residue on Evaporation	ppm	30000	20000 -30000	39600	35400	30200	37200	30200 -45400
4	Residue on Ignition	ppm	12000	7000 -15000	14200	11200	11800	13800	11200 -14200
5	Total Organic Carbon (TOC)	ppm			14000	16500	15200	15800	14000 -16500
6	Total Nitrogen	ppm	5520	6250	4960	5300	5920	5850	4960-5920
7	C/N Ratio		1.7	1.6	2.82	3.11	2.57	2.71	2.57-3.11
8	C1-	ppm	5500	5000	4130	3400	4650	4230	3400-4650
9	P04 ⁻³	ppm	1000	_	715	1450	780	815	715–1450
10	B 0 D	ррт	13500	20000	32000	28700	25900	26000	25900-32000
11	COD	ppm	4000	4750	8620	14100	10600	9600	8620-14100

Table 6. Comparison of Aerobic and Anserobic Composting.

Composting Method	Experiment Period	Initial Water Content %	1	Atmospheric Temperature (°C)	
Aerobic	1979, 3.16-4.10	47.68	12-63	3-22	Nightsoil: Waste = 2,340(Kg):4,214(Kg) maintenance days more than 50°C= 8 days
Anaerobic	"	37.68	12-43	11	N : W = 2,340(Kg): 4,214(Kg) maintenance days more than 40°C= 2 days
Aerobic	1979, 5.25-6.21	33.85	40-79	17-36	N : W = 1,430(Kg): 2,100(Kg) m. d. m. t. 50°C= 30 days
Anaerobic	10	33.85	17-51	11	N : W = 1,430(Kg): 2,100(Kg) m. d. m. t. 40°C= 20 days
Aerobic	1979, 9.11-10.1	36.25	25-65	17-38	N : W = 1,400(Kg): 2,400(Kg) m. d. m. t. 50°C= 16 days
Anaerobic	11	36.25	29-45	11	N : W = 1,400(Kg): 2,400(Kg) m. d. m. t. 40°C= 5 days
Aerobic	1979, 1.24-2.13	37.13	10-55	-5-15	N : W = 1,665(Kg): 2,337(Kg) m. d. m. t. 50°C= 7 days
Anaerobic	п	37.13	7-35	11	N : W = 1,665(Kg): 2,337(Kg) m. d. m. t. 40°C= 4 days

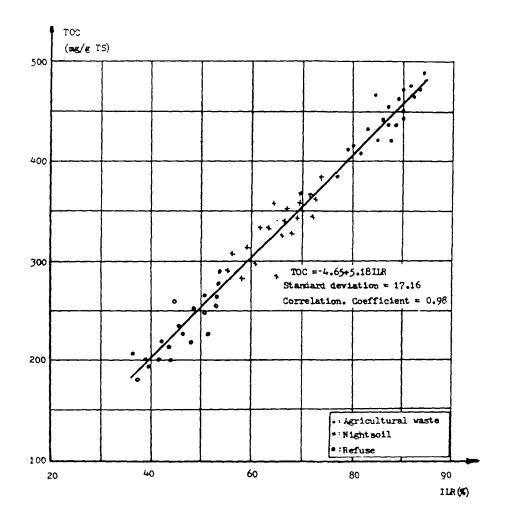
The C/N ratio has been shown to be an important variable that needs to be maintained at about 30 for successful aerobic composting. The quantitative determination of organic carbon in the wastes to be composted can be achieved by measuring the ignition loss rate (or % ash content). A dry sample is ignited to drive off combustible material and weighed after cooling. The percent ash content is determined according to the following formula:

% ash = $\frac{dry \text{ weight before ignition - weight after ignition}}{dry \text{ weight before ignition}} \times 100$

Figure 1 shows the correlation regression for the total organic carbon TOC (mg/g) and the ignition loss rate ILR(%). Specifically, it was found that

TOC = 5.18 ILR - 4.65

Figure 1. Correlation between total organic carbon and ignition loss rate.



4. Composting Experiments Performed Under Field Conditions

Under field conditions, samples of night soil mixed with solid wastes were distributed among five different composting methods, shown below. The effectiveness of the methods was evaluated in terms of temperature maintained, odour control and destruction of microorganisms. It was found that eggs of <u>Ascaris</u> were mostly inactivated when a temperature of 55°C or more was maintained for 7 days.

Figure 2. Comparison of Different Aerobic Composting Methods.

It was shown that such composting is feasible under Korean conditions, except for the winter months when the amount of the compostable material in the refuse is less than 10%. However, the addition of agricultural wastes can solve that problem. It is desirable, moreover, to incorporate a method of separation of inorganic material such as glass, iron, PVC, etc. from the solid waste before composting.

5. Agricultural Trials Using the Ripened Compost

The compost produced was stored on the field for more than 2 months before being passed through a 4 cm sieve and applied to the experimental plots at various rates. The chemical composition of the compost is shown in Table 7.

Table 7. Chemical Composition of the Compost.

	рН (H ₂ O)	pH KC1	Humus (%)	P ₂ O ₅ (ppm)	K (me/100g)	Ca (me/100g)	Mg (me/100g)	N (%)	Date
1	7.23	6.25	8.12	715	0.49	5.5	2.2	0.34	August 1, 1978
2	8.25	7.15	8.22	835	0.63	5.5	1.04	0.28	August 1, 1978
3	7.9	6.95	7.46	1210	0.58	3.55	1.65	0.42	August 1, 1978
4	7.55	6.5	7.76	380	2.11	8.31	1.95	0.38	September 1, 1979
5	8.05	7.07	7.84	430	2.45	4.65	1.32	0.35	September 1, 1979
6	7.76	6.62	8.17	778	0.56	5.40	1.63	0.37	September 1, 1979
er- ige	7.79	6.76	7.93	725	1.14	5.49	1.63	0.36	

The compost applied to crops such as radish, cabbage, barley, wheat, spinach, eggplant, red pepper, salvia and celosia was found to be beneficial but actual yield increases were not reported. The average humus content of the compost was 5% as compared to an average value of 3% for Korean soils. Inorganic materials should be removed from the solid waste before composting when the compost is to be used for agriculture.

6. Economic Aspects of Composting

Presently in Korea, solid wastes are disposed of by dumping and nightsoil is treated either by the oxidation or digestion methods before being released into the environment. The composting method for a combined treatment of nightsoil and solid waste has similar construction costs to the other two methods but substantially lower running costs (Table 8).

Table 8. Cost Comparison of the Three Different Treatment Methods.(1)

			_ , , , ,
Treatment Method	Composting	Biological aerobic oxidation	Biological anaerobic digestion
Treating Capacity	Nightsoil 48m ³ /d Refuse 200m ³ /d	Nightsoil 48m ³ /d	Nightsoil 48m ³ /d
Total Cost	341,000 Thousand Won	330,000 Thousand Won	363,000 Thousand Won
Construction costs			
Construction and Architecture	184,760 Won	152,000 Won	185,000 Won
Machine	85,000 Wan	110,000 Wan	95,000 Won
Electricity	12,000 Won	14,000 Won	12,000 Won
Additional works	5,000 Wan	5,000 Won	15,000 Won
Misc. expenses	22,440 Won	19,000 Won	23,000 Won
Added value tax	31,000 Won	30,000 Won	33,000 Won
Annual running cost	102,392 Won	125,000 Wan	95,000 Won
Refuse disposing cost (2)	40,000 Won	100,000 Won	100,000 Won
Total maintaining cost	142,392 Won	225,000 Wan	195,000 Won
Advantage	1. Low operation cost 2. Waste reusing 3. Low construction cost 4. Complete refuse treating	1. Easy operation 2. Low odour	 Low running cost Digesting-gas reusing
Disadvantage	1. Large space requirement for composting treatment plant	1. High running cost 2. Water pollution 3. Separate treating of refuse 4. Soil pollution	 High construction cost Operation trouble (in case of small city) Water pollution Separate treating of refuse Soil pollution

Notes: (1) Population - 80,000; (2) Sanitary landfill method

DISCUSSION

To combat Korea's growing solid waste and nightsoil disposal problems, better techniques for treatment and reuse of the wastes were needed. Both public health and costs had to be considered.

Physical and chemical analyses on both the refuse and nightsoil were performed, after which composting of the wastes was studied under various conditions. Due to the nature of the household fuel used, domestic refuse from several cities was found to have a high ash content (75% to 85%). This necessitated the addition of organic matter such as agricultural wastes for the biological degradation to take place, especially during the three winter months. However, due to the yearly increase of 0.5% or more of compostable materials in Korean urban and rural refuse, this may no longer be necessary.

Aerobic composting was found to be the preferred method under Korea conditions. For most of the year, a high enough internal temperature of the compost pile was achieved for effective pathogen destruction. For the winter months, however, the compost pile needed to be covered to maintain a sufficiently high internal temperature.

Preliminary agricultural trials showed the compost to be beneficial, although more testing was needed. Cost analysis showed that the proposed treatment method was economically viable, even without including the proposed revenue from the sale of the compost.

Publications arising from the IDRC-supported Waste Reuse Project.

- Gemeinsame Kompostierung Von Kommunalen Faekalien und Alfaellen in der Republik Korea.
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- Waste Reuse in Korea. Proceedings of the 4th National Congress for Solid Waste Utilization and Management, Korea Solid Waste Engineering Society, November 1985, Dong-A University, South Korea.

EXCRETA REUSE (Guatemala)

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Experiments on several appropriate technologies for the sanitary treatment and reuse of rural domestic wastes have been conducted. These include, among others, the Chinese-type biogas digester which produces biogas (mainly a mixture of methane and carbon dioxide), slurry and liquid effluent and the DAFF latrine (Dry Alkaline Fertilizer Familiar) that produces a dry humus product that can be used in agriculture and aquaculture. However, evaluation of these technologies was not complete. For the technology to be adopted by local populations it must be affordable, able to be maintained by the users and be aesthetically and culturally acceptable.

OBJECTIVES

This project was designed to develop just such economical and easily maintainable technologies for the recycling of domestic wastes by rural Indian communities in Guatemala. The specific objectives were to study three different systems:

- a) dry alkaline latrine
- b) Chinese-type biogas plant
- c) fish pond receiving wastes with and without pretreatment by the Chinese biogas digester

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These were then to be evaluated with regards to:

- -- community acceptance
- -- community use and maintenance
- -- treatment efficiency
- -- production of reusable by-products
- -- public health aspects
- -- cost efficiency

RESULTS

1. DAFF Latrine (also called LASF)

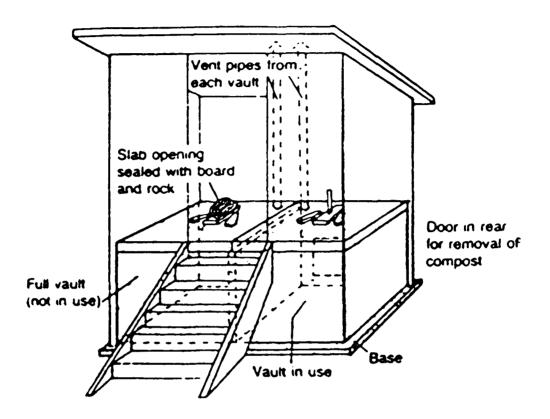
The dry composting latrine consisted of a two-chamber vault constructed above ground. The upper slab had a separate hole for each vault into which fecal matter was deposited. To maintain dryness inside the vault and to achieve a proper C/N ratio, urine was collected separately and after defecation, the user sprinkled ash to completely cover the feces. After three to six months of use, the vault was sealed with cement to allow composting to continue and the family put the second adjoining vault into use. Thus the two-vault toilet operated continuously.

Figure 1 below shows the DAFF latrine diagramatically. For privacy, the latrine is surrounded by walls made of bamboo, matting or other locally available material.

Thirty-one latrines were initially installed in rural areas in Guatemala. Of these, 14 were in use, benefiting a total of 62 people. The performance of the 14 functioning units was evaluated.

Six of the 14 latrines were found to be functioning properly. This meant that they did not contain water or insects and insect larvae and were odour free. The eight latrines that were functioning poorly contained water, insects and larvae and had a bad odour. The difference between the two groups seemed to be in the amount of ash used. For the latrines to function properly and yield a dry, innocuous product, sufficient ash must be used after defecation to completely cover the feces. Either sufficient amounts of ash were unavailable or the users needed health education in the proper use of the latrine.

Figure 1: Double-vault composting toilet.



NOTE: Outlet for urine collection not shown. DAFF latrine now in use in Quatemala has a raised seat with rubber tubing leading to a urine collection bottle.

Microbiological analyses of the end products (after a digestion period of about 2 months) from 8 latrines were performed (Table 1). Coliform counts (MPN/q) ranged from 5 to 3,800 and the number of helminth eggs per gram ranged from 0 to 8,500 with a viability of 0-30%.

No polio or spiral viruses were detected in 21 samples taken from DAFF latrine vaults in use, their compost or the biogas digesters although viruses were detected in samples taken from conventional earth latrines and sedimentation tanks for community wastewaters.

For the products of the DAFF latrines to be considered safe healthwise, counts below 10,000 coliforms/q and 10,000 helminth eqqs/q with a viability below 30% should be achieved. Thus the dry organic matter produced in the properly functioning latrines could be considered safe for use in agriculture.

Table 1: Results of microbiological analysis of fertilizers produced in eight DAFF latrines.

	TOTAL			
LATRINE	COLIFORMS MPN/q	EGGS/q	VIABILITY %	pН
C-1	210	8,500	6.0	8.46
C -2	5	Ð	0	10.00
C-3	2,400	1,750	30.0	6.61
C-4	40	1,000	5.0	8.45
C-9	1,200	250	0	9.37
P-1	110	2,800	14.3	8.60
P-2	40	0	0	9.40
T-4	3,800	5,000	15.0	8.50

2. Chinese-type Biogas Digester

During the period 1979 to 1980, eleven prototypes of biogas plants were constructed, six of which were a round-type Chinese model fed by latrines. The fixed-top, $12m^3$ Chinese-type digester was more economical to build than other models and had lower maintenance requirements. However, problems were experienced with irregularities in gas production, perhaps due to the high altitude (over 1,500 metres above sea level) and low temperatures.

Generally, it was found that methane production was directly related to digester temperatures. Often, the amount of gas produced was not enough to make the system economical. It was found, however, that extra heating improved performance and increased methane gas production. Although some problems were encountered with the gas guages, these were later solved.

The underground Chinese-type digester, when used as a latrine, had as its end products, biogas, solids and a relatively clear effluent. Both the solids and the effluent can be used as fertilizer. The digester effluent was found to be high in nitrogen, phosphorus and potassium.

Microbiological analysis was performed on the digestion products. For the 6 digesters studied, a total of 28 samples of effluents and sludges were analyzed in different phases of operation. Table 2 gives the average values of the coliform and parasite egg counts as well as percent viability of the eggs.

Table 2: Laboratory findings on effluents and sludges from Chinese-type biogas digesters.

Sample		Coliform	Eggs	Viability	
Digester	No.	type	MPN/g	No./g	*
DC-CHB-1	4	effluent	1222+1361*	0	0**
DC-CHB-2	3	effluent	103+81	0	0
DC-CQ-1	6	effluent	5571+9192	0	០
	1	sludge	2400	0	0
DC-SA-1	1	effluent	23	0	0
DC-SLT-1	5	effluent	710+1055	350+339	6.6+14.8
	2	sludge	1750+919	750+707	0
DC-SLT-2	6	effluent	1405+1154	365+175	8.3+13.3

- * Average and standard deviation of the MPN of coliforms by dilution count in 9 MacConckey tubes.
- ** % of viable helminth eggs by supravital staining and observation of the microscopic morphology.

The coliform counts were somewhat lower than those of the DAFFs, with the exception of DC-CQ-1, which gave higher counts, both in the effluents and in the sludges. The number of helminth eggs was low and their viability ranged from 0 to an average of 8.3%

To prevent storage problems, the time of unloading of the digesters should be coordinated with peak demand for fertilizer. The use of machinery for loading and unloading the digesters was found to be essential since it was heavy work.

Although the total cost of the biogas unit (about US \$1,200) was high for a rural Indian family, it was possible for a well-organized group to afford such a unit.

3. Fish Culture Experiment

Experiment 1.

Three plastic tanks of 1,000 litre capacity were set up for fish culture using carp (<u>Cyprinus carpio</u>) and tilapia (<u>Sarotherodon sp.</u>). Due to the high altitude (1500 m above sea level) and cool climate, the tanks were equipped with a thermal greenhouse type plastic dome to mainain a temperature of 23-25°C. The fish ponds were fertilized with effluents from the biogas digesters and with humus from the DAFFs. This caused abundant growth of phytoplankton and zooplankton which served as food for the alque-harvesting fish.

The pond water and sediment were examined for the presence of parasite ova and coliform bacteria. No ova were found and a coliform count of MPN=10/100 ml was reported in the water.

Three dilutions of the wastes from the DAFF latrines, corresponding to three different levels of fertilization, were used for fish culture. These dilutions were: 1:10,000 1:20,000 1:40,000

The lower levels of dilution resulted in a too strong BOD for fish culture as shown by low fish survival rates, and the 1:40,000 dilution proved to be the best.

The growth of tilapia in the three tanks was also studied using three different levels of application of effluent from the Chinese-type biogas digester (Table 3).

Table 3: Growth of tilapia during 37 days in 1000 litre tanks receiving 3 levels of fertilization from a Chinese-type biogas digester.

Tank	Fertilizer Rate	Ave. length of	Ave. length of	Rate of
No.	m1/m ³ /day	fish stocked	fish harvested	growth
		(mm)	(mm)	(mm/day)
1	800	27.3	93.0	1.78
2	400	25.7	86.5	1.64
3	200	22.3	82.0	1.61

Note: Three fish were stocked per tank but only 2 fish were harvested in tank 2 and 1 in tank 3 because of mortality due to small size of fingerlings.

The best biomass conversion from fertilizer to phytoplankton to fish was obtained in tank 1 and since no adverse effects were noted, fertilizer effluent could perhaps be applied at a higher rate of 1 litre/m³/day or more.

In tests using residue from the DAFF latrines, it was sufficient to apply 38 ml/m 3 /day, while with the biogas digester effluent, rates of up to 800 ml/m 3 /day could be used.

Experiment 2.

Two cement fish tanks covered with a plastic dome to maintain a sufficiently high temperature for the carp and tilapia were used for fish culture. Table 4 below gives the details of the tanks and numbers of fish stocked.

Table 4: Tank characteristics and fish stocking density experiment 2.

	Tank #1	Tank #2	Totals
width (m)	4	4	
length (m)	16	16	
Max. depth (m)	1.4	1.4	
surface area (m²)	64	64	128
volume * (m ³)	53	75	128
No. of tilapia	65	245	310
No. of carp	85	0	85
total/tank	150	245	395 fish
tilapia stocking density	1.2/m ³	3.3/m ³	
carp stocking density	1.7/m ³	0	
total per tank	$2.9/m^{3}$	3.3/m ³	

^{*} the difference in volume is due to internal profile of the tanks.

Table 5 shows the fish production obtained when compost from the DAFF latrines was added to the fish tanks.

Table 5: Fish yields obtained in tank 1 and tank 2 with the addition of composted human wastes from DAFF latrines.

	Tank #1		Tank #2		
	carp	tilapia	tilapia	<u>Total</u>	
No. of fish	85	65	245	395	
total initial weight (g)	1445	325	735	2505	
ave. initial weight (g)	17	5	3	-	
ave. initial length (mm)	94	55	54	-	
total final weight (g)	1185	5376	11,760	18,321	
ave. final weight (g)	15	84	48	-	
ave. final length (mm)	103	175	155	-	
net gain (kg)	26	+5.1	+11.0	16.4	
potential gain (kg/ha/yr)		79 7	1719		
amt. of fertilizer added		litres	154 litres		
added to kg fish per litre	15.	.1	14		

Experimental results showed that fish production was possible using either effluent from a biogas plant or humus from a composting toilet. No viable parasite eggs or high coliform counts were found in either surface or bottom waters or in the sediment in the fish tanks. The fish produced was acceptable for human consumption from both a microbiological and taste point of view.

Although the plastic dome cover for the fish tanks can be constructed using purchased plastic and galvanized wire and locally-grown bamboo, it was considered too expensive for even several rural families together. An alternative would be to culture fish species better adapted to the cool climate of the high plain.

4. The Use of Compost, Sludge and Effluent for Agriculture

The compost produced by the DAFF latrines was analyzed for its fertilizer value. Table 6 shows the results of the analysis for moisture content, phosphorus, carbon, nitrogen and organic matter.

Table 6: Laboratory analysis of agricultural fertilizers from dry alkaline latrines.

LATRINE	WATER 	PHOSPHORUS PPM	ORGANIC CARBON %	ORGANIC MATTER %	NITROGEN %
C-1	52.3	196.0	4.0	6.8	0.28
C-2	52.0	70.0	2.0	3.4	1.07
C-3	46.6	74.0	6.4	11.0	0.95
C-4	76.9	94.0	5.0	8.6	0.78
C-5	48.2	96.0	2.7	4.7	0.28
C-7	44.8	120.0	5.0	8.6	0.43
C-8	35.4	214.0	6.4	11.0	0 .9 5
C-9	52.7	450.0	3.9	6.7	0.56
C-11	54.8	178.0	4.8	8.2	0.74
T-1	59.0	86.0	1.2	2.0	0.28
T-2	54.0	113.0	1.8	3.1	0.73
T-4	47.8	300.0	2.4	4.2	0.88
_ ×	52.0	165.9	3.8	6.5	0.66
± DE	9.9	113.2	1.8	3.1	0.29

Moisture content ranged from 35.4 to 76.9% and organic matter from a low of 2.0% to a high of 11.0%. A nitrogen content of up to 1.07%, carbon to 6.4% and phosphorus to 450 ppm was found.

A preliminary experiment was performed growing vegetables in 14m² plots with 5 different fertilizer regimes, i.e., the use of compost alone, the use of compost and liquid biogas digester effluent, chemical fertilization, the use of the digester effluent alone and a control which did not receive any form of fertilizer. Best results were achieved with the use of compost and digester effluent, chemical fertilizer and effluent alone. The use of compost alone and the control gave lower yields. (Information as to application rate of nitrogen, phosphorus and potassium for the different fertilizer treatments was not presented.)

DISCUSSION

Researchers were successful in building and evaluating a number of DAFF latrines and Chinese-type biogas plants. Their use by members of several rural Indian communities in Guatemala was studied and their performance in transforming human excreta into a safe, reusable product was evaluated.

It was found that with proper education and supervision of the users, the latrines functioned well and produced an odourless, dry organic product that could be applied as fertilizer to crops or used in fish ponds. The use of compost alone on vegetable crops was found to be important for the improvement of soil structure and consequently, water retention, but results were inconclusive as to its ability to improve crop yield.

Problems were initially encountered with the six Chinese-type, fixed top digesters but gas production improved after some design modifications. Due to lower temperatures at higher elevations, an outside source of heat was found to be necessary to maintain the methane-forming bacteria.

The digester effluent was found to be safe from an excessive bacterial and parasite egg load and was thus acceptable for use in fish culture and in agriculture. Fish culture was shown to be feasible, using the recycled organic wastes as a nutrient source for algae production. Unfortunately, the temperature in the Guatemala highlands was too low for the tilapia to thrive without the added protection of thermal covers on the fish tanks, which added to the cost of the technology.

This project showed that the DAFF latrine was an acceptable, appropriate technology for rural communities in Guatemala. Even though pit latrines were available free via the official latrinization campaign, family groups have requested loans to build the latrines (costing about US\$ 35) along with an improved woodstove to provide the required ash. Since 1980, about 500 DAFF latrines have been built in Guatemala.

Publications arising from the IDRC-supported Waste Reuse Project.

- Van Buren A., J. McMichael, A. Caceres and R. Caceres. 1984. Dry Composting Latrines in Guatemala. Ambio 13 (4): 274-277.
- Cáceres, A. and R. Cáceres. 1984. Control Sanitario de Bioabonos y Effluentes de Latrinas Aboneras Secas Familiares y Digestores de Biogas. 18 pp. Guatemala.
- CEMAT. 1981. Los Sistemas Bioenergéticos Instrumentos del Ecodesarrollo. Nairobi, UNCNRSE.

PIGGERY WASTE TREATMENT - PHASE I (SINGAPORE)

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Singapore, despite a shortage of agricultural land, has developed a highly successful pig and poultry production system to meet domestic demands. However, the wastes generated by the population of 0.8 million pigs and 12 million poultry amount to 14 million litres of pig wastewater and 1,000 tons of poultry excreta per day. The inadequate treatment and disposal of these wastes presented serious public health and environmental problems to the authorities. Organic pollution of farm origin represented an added threat to the island's limited supplies of potable water.

To try and solve the water shortage, 60% of the island was designated a catchment area for water collection. This resulted in the relocation of the majority of pig farms into a confined area around the Ponggol and Serangoon Basins.

Treatment by conventional techniques, such as activated sludge plants or aerated lagoons, without reclamation of nutrients and reuse of water, would be expensive and nonproductive.

The reclamation of nutrients from wastewater by algae culture is a productive alternative. Recovered algae can then serve as animal feed to make the system economically attractive to Singapore farmers who have to import all their supply of animal feed.

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OBJECTIVES

The general objectives were to establish (a) a high rate pond system for purification of pig wastewater, water reclamation and nutrient recovery through algae growth, and (b) to evaluate technical efficiency and economic feasibility of this system for treatment of pig wastewater.

The specific objectives were to:

- a) design and construct a pig waste collection and transportation system for a 1,500 pig unit to channel the wastes to primary treatment facilities;
- b) construct primary treatment facilities for the collected waste in order to achieve preliminary treatment and to discharge a uniform effluent into the high rate ponds for further treatment;
- design and construct high rate ponds required for efficient photosynthetic treatment of the effluent;
- d) determine the optimal level of waste loading rates, retention periods, mixing periods and other related factors with respect to high rate pond operation;
- e) optimize algae productivity in the ponds with respect to yield, composition of algal flora and related algae biological factors;
- f) design and install equipment which could be used for small scale algae harvesting and processing;
- g) provide detailed plans for accurate costing of chemical and mechanical water reclamation and algae harvesting systems; and
- h) conduct small scale preliminary animal feeding trials with recovered algae.

RESULTS

Pig wastewater, composed of feces, urine, spilled feed and wash water was fed into sedimentation tanks with a 45 minute detention time prior to entry into the high rate ponds. Table I shows the important wastewater parameters of the raw and settled wastewater.

Table 1: 12 months weekly average of characteristics of raw wastewater, settled wastewater and percent removal at over flow rate of 3.5m/hr (June 1978-June 1979).

Wastewater Parameter	Symbol	Raw Wastewater mg/1 (±S.E.)*	Settled Wastewater mg/1 (±S.E.)	Percent Removal
Total Solida	TTS	6456 (253)	1977 (76)	69
Total Suspended Solids	TSS	4475 (227)	560 (45)	87
Biochemical Oxygen Demand	BOD	2892 (152)	1400 (79)	51
Chemical Oxygen Demand	COD	7334 (353)	1911 (80)	74
Total Kjeldahl Nitrogen	TKN	-	269 (13)	-
Ammoniacal Nitrogen	NH3-N	-	185 (8)	-

^{*} standard error

High Rate Algae Ponds

High rate ponds are defined as ponds designed to grow algae at a shallow depth for maximum light penetration, operated on a continuous basis with fixed detention time and equipped with agitators to provide low-velocity mixing.

Four pilot ponds with a surface area of 125 m^2 each and two demonstration ponds with a surface area of 1230 m^2 each were constructed. The ponds were built with racecourse configuration using precast corrugated asbestos cement sheets embedded vertically in a concrete base and jointed with silicone sealant.

The ponds were equipped with a slow speed paddle wheel mixer. The ideal mid-channel velocity for intermittent mixing was found to be 18-20 cm/sec while continuous mixing was best at 10-12 cm/sec.

Operating depth was kept at 200 mm with loading rates ranging from 175-250 kg BOD/ha/day. Optimal operating depth was found to be between 200 and 300 mm.

Table 2 summarizes the algal flora of the pilot ponds from start up, May 1978 to March 1980.

Table 2: Algal Population and Relative Dominance in the Pilot Ponds.

Month	Dominant Species	Others
May 1978 Jan 1979	Micractinium spp.	-
Feb 1979 Mar 1979	Non-filtrable blue-green "Synechocystis"	-
Apr 1979 Jul 1979	<u>Docystis</u>	Micractinium, Ankistrodesmus, Chlorella, Oscillatoria
Aug 1979 Jan 1980	Oscillatoria	Chlorella, Oocystis, Micractinium, Ankistrodesmus
Jan 1980 Mar 1980	Micractinium + Oscillatoria	<u>Chlorella</u>

It can be seen that the <u>Micractinium</u> was dominant initially until predation by <u>Moina</u> reduced its numbers. <u>Micractinium</u> was replaced by a very small non-filtrable blue-green algae for several months until larger algal species reappeared in the ponds.

1. Predators and Predator Control

Initially lime was used to increase the pH of the ponds to 9.6, killing off the predator Moina and rotifers. However, the rotifers are desirable as they graze on smaller algae species. The absence of rotifers caused an increase in the small blue-green algae population. These passed through the Gelman glass fibre filter used for total suspended solids determination which was a measure of pond productivity. As a result, pond productivity fell to very low levels.

Continuous mixing by the paddle wheel mixer at 10-12 cm/sec was found to be adequate to prevent Moina from reaching damaging levels. Continuous mixing at night drowned the predators in their attempts to compete for dissolved oxygen at the pond surface. An additional benefit of the continuous low-speed mixing was control of insect propagation in the ponds as egg-deposition was discouraged.

2. Biomass Productivity

Biomass productivity for the pilot ponds was calculated on the basis of the total suspended solids (TSS mq/1) by

$$Pg = \underline{C} \times \underline{V} \times \underline{1}$$

$$1000 \times t \quad A$$

where: Pg = gross biomass productivity in $g/m^2/day$

C = TSS in mg/l

V = nominal pond volume, i.e. 25,000 liters

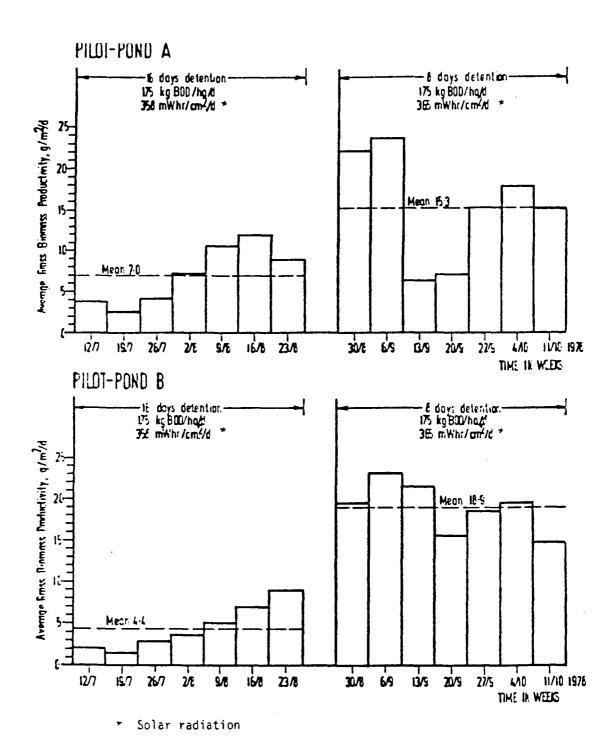
t = detention time in days

A = pond surface area = 125 m²

Detention times of 16, 8 and 4 days were tested. Figure 1 compares the average gross biomass productivity of the pilot ponds at 175 kg BOD/ha/day loading and at different detention times (8 and 16 days).

The optimum detention times under Singapore conditions were found to be 4 to 8 days. At a 4 day detention time with an increased organic loading of 200 kg BOD/ha/day, the biomass productivity ranged from $8.5g/m^2/day$ to 33 $g/m^2/day$.

Figure 1: Estimated biomass productivity from pilot ponds at 16 and 8 days detention time.



3. Pond Effluent Quality and Treatment Performance

Pond performance was assessed by comparing filtered (using Whatman No. 1 filter paper) effluent to the influent water quality. The percent removal of BOD, COD and NH₃-N were measured under various operating conditions. Table 3 shows the percent removal for these parameters during July 1978 to July 1979.

Table 3: Percent Removal of BOD, COD and NH3-N.

Conditions	BOD	% Removal COD	NH3-N
12 Jul - 23 Aug 78 Fine weather, no predation, 16 days detention	95	88	89
30 Aug – 11 Oct 78 Fine weather, no predation, 8 days detention	89	76	64
25 Oct - 29 Nov 78 Wet weather, low predation, 8 days detention	78	63	70
25 Oct - 29 Nov 78 Wet weather, low predation, 4 days detention	65	63	49
27 Dec 78 – 31 Jan 79 Wet weather, heavy predation, 8 days detention	60	47	58
27 Dec 78 - 31 Jan 79 Wet weather, heavy predation, 4 days detention	42	22	30
6 Jan 79 – 25 Jul 79 Fine weather, moderate predation, 6 days detention	87	74	55

From August 1979 to March 1980 the ponds were receiving a BOD loading of 250 kg/ha/day. Mean percent removal was:

B0D 83-85% C0D 74-78% NH3-N 70-71% Pond performance during this period was greatly improved due to better predator control by continuous mixing. Moina predation was found to be directly related to treatment performance of the ponds which affected effluent quality.

4. Algae Harvesting and Processing

Harvesting of algae from the high rate ponds represented one of the major technological obstacles to the application of the wastewater treatment and utilization system. A pilot scale harvester based on the design of Dr. J. C. Dodd was developed.

This harvester is essentially a continuous filtration device, achieving filtration by a belt device consisting of a fine weave polyester fabric having a nominal mesh opening size of 5-16 u. The belt is supported on a perforated cylindrical wall of the filtration drum (2m dia. x 1m nominal width). The filtration drum is immersed in the influent tank with flow passing radially inward so that algae are deposited on the outer surface of the fabric.

After leaving the filtration drum the belt passes around a 0.3 m dia. separation drum in the reverse direction so that algal solids are in contact with the perforated wall of the separation drum. A suction box positioned within the separation drum is used to suck off the layers of algae on the belt, into a cyclone where the algal slurry is collected and periodically drawn off. The belt is then washed by water showers and returned over a series of rollers to the filtration drum. Figure 2 gives a schematic diagram of a Fine Weave Belt Filter Harvester.

5. Nutritional Evaluation and Feeding Trials

The algae biomass harvested for the feeding trials was made up of different species. It was either steam boiled or drum dried before analysis.

The major component of the algae was protein which constituted more than 50% on a dry matter basis. The level of crude protein in the algae was similar to soybean meal but the ash content was higher. Calcium and phosphorus were the main mineral components. Though inferior to soybean meal protein, the amino acids profile of algal protein was comparable with the exception of lysine, isoleucine and valine.

Suction box

Fine Weave Belt

Relt Guides

Screen

Influent

Tank

Filtration drum

Figure 2: Fine Weave Belt Filter Schematic Diagram.

A: Pig Feeding Trials

A pig feeding trial using steam boiled algae was performed. Three groups of 6 pigs were provided the following diets:

- Diet A standard 16% corn-soya grower diet
- Diet B diet with 52% of its soybean meal replaced by steam boiled algae
- Diet C diet with 100% of its soybean meal replaced by steam boiled algae

Initial body weights/pig were 32 kg and final body weights were 73.0 kg, 70.5 kg and 64.7 kg for Diet A, B and C, respectively. The results of the feeding trial demonstrated that steam boiled algae could replace 52% of the soybean meal in the diet with little reduction in growth rate and feeding efficiency.

Another pig feeding trial was conducted to determine if lysine supplementation would improve the growth performance of pigs fed 9% drum dried algae in the diet. Table 4 shows the chemical composition of the diets.

Table 4: Components and chemical composition of control, algae and algae + lysine diets.

Characteristics	Control	Algae	Algae + Lysine
Drum Dried Algae	0.00	9.00	9.00
Ground maize	74.50	74.50	74.50
Soybean meal (solv.)	18.00	9.00	9.00
Rice bran (solv.)	2.50	2.50	2.50
Fishmeal	2.50	2.50	2.50
Mineral Supplement	1.50	1.50	1.50
Microingredients	1.00	1.00	0.92
Lysine	0.00	0.00	0.08
Chemical composition			
Crude protein %	14.8	16.0	16.2
Crude fat %	2.9	3.0	3.1
Crude fibre %	2.7	2.8	2.6
Ash %	4.3	4.7	5.2
Lysine %	0.84	0.76	0.84
Methionine + Cystine %	0.56	0.64	0.64

The results of the feeding trial, conducted over 63 days are shown in Table 5.

These results showed that centrifuge harvested and drum dried <u>Oscillatoria</u> (dominant species) did not have any adverse pathological effects on the pigs. It also showed that lysine supplementation had no significant growth promotion effects. Possibly, lysine was not limiting in the predominantly <u>Oscillatoria</u> biomass harvested during this period. The high ash content and low carbohydrate digestibility of the algae made it a low energy feed.

B: Chicken Feeding Trials

Four groups of day old broiler chickens received diets varying in amounts of drum dried algae added to the feed. The diets were formulated so that 1/3, 2/3 and all of the soyabean meal was replaced by drum dried algae.

Table 5: Summary of performances of pigs fed with control, algae and algae + lysine diets.

		Diet Tre	atment
Characteristics	Control	Algae	Algae + Lysine
No of pigs Av. initial body-weight/pig (kg)	6 35.0	6 35.3	6 35.2
Av. body-weight/pig at the end of expt. (kg)	77.2	78.8	77.2
Av. body-weight gain/pig (kg)	42.2	43.5	42.0
Daily body-weight gain (kg)	0.67	0.69	0.67
Feed consumed/pig (kg)	122.4	118.9	121.0
Feed Conversion Ratio	2.90	2.73	2.88
Crude protein consumed/pig (kg)	18.1	19.0	19.6
Protein Efficiency Ratio	2.33	2.29	2.14

Results showed a direct relationship between reduced growth rate and amount of algae added to the diet. Twelve percent, 21 % and 30% reduction in growth rate occurred with increasing amounts of dried algae replacing soyabean meal. At the higher levels (2/3 and 3/3 algae replacement) reductions in feed efficiency were observed as well as increases in the mortality rate. At the 1/3 replacement rate, however, there was no decrease in feed efficiency or mortality rate.

DISCUSSION

A collection and transportation system for piggery wastes as well as high rate stabilization pends for waste treatment were built. The system was manipulated to determine the operating conditions that yielded the best waste treatment. It was found that the degree of treatment of the primpary effluent depended on algae productivity which in turn was dependent on design and operating criteria of the system.

Optimal operating depth for the high rate ponds was found to be 200-300 mm, optimum detention time was 4 to 8 days with a continuous slow speed mixing by paddle wheel of 10 to 12 cm/sec. Percent removal of BOD ranged from 42 to 95%. Higher algae productivities can

be expected at shorter detention times, at the expense of thinner cultures, higher costs for processing the extra water, and higher sensitivity to factors such as shock loads causing reduced stability.

Initial pig feeding trials using harvested dried algae to partially replace soybean meal in the diet showed promising results. More research is needed on harvesting and dewatering techniques, use of algae in animal feed and cost evaluation of the system. Continued IDRC support for a second project phase looked further at these important aspects.

Publications arising from the IDRC-supported Waste Reuse Project.

 "Wastewater Treatment and Resource Recovery: report of a workshop on high-rate algae ponds", Singapore, 27-29 February 1980. IDRC-154-e, 47 p. International Development Research Centre, Ottawa, Canada.

PIGGERY WASTE TREATMENT - PHASE II (SINGAPORE)

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The importance of algae for waste treatment lies in its dual function of absorbing organic nutrients from the wastewater and its production of oxygen during its photosynthetic phase. For the system to be economical and effective, the algae must be harvested efficiently to achieve high quality effluent and the harvested algae used as animal feed. The Phase I project undertook initial experiments in developing a harvesting technology and concluded that additional research was required in this area.

OBJECTIVES

For Phase II of the project, the overall objectives were, therefore, to develop equipment and methods for harvesting micro-algae grown in pig wastewater in high-rate ponds and to evaluate the technical efficiency and economic feasibility of the overall system for nutrient recovery and waste treatment. The specific objectives were to:

- a) design and fabricate a pilot-scale continuous system for harvesting micro-algae from high-rate ponds treating pig wastewater;
- operate this harvesting system for a sufficient length of time to fully evaluate its efficiency in removing algal solids;

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- c) design, fabricate and operate a pilot-scale chemical flocculation and dissolved air flotation algal harvesting system;
- d) study chemical flocculant recovery and reuse by acidification of algal concentrate, including the process' technical and economic feasibility;
- e) conduct small-scale studies on the potential of auto-flocculation as a harvesting method;
- f) study the recycling of high-rate pond effluent, after algae removal, as a washwater for pig housing and dilution water for high-rate pond operation;
- g) conduct trials on the post-harvest processing of algae by drum-drying and the use of biogas obtained from anaerobic digestion of pig waste solids (obtained during primary treatment) as fuel for the process;
- h) conduct feeding trials using processed algae as pig, poultry and fish feed to fully determine algal nutritional values and limitations of waste-grown algae as animal feeds, and
- carry out detailed economic analysis of the overall treatment and nutrient and water recovery processes.

RESULTS

The second phase of the Piggery Waste Treatment (Singapore) project concentrated on developing an efficient algae harvesting procedure. Centrifugation was found to be effective but very costly due to high energy consumption. Biological harvesting was also investigated but to a lesser degree since it was not as efficient. Research emphasis was, therefore, on the three following methods:

- -- continuous filtration harvester
- -- dissolved air flotation after chemical flocculation
- -- autoflocculation

Post-harvest handling was also important since the initial slurry obtained (containing 1-5% solids) was too wet to handle and deteriorated rapidly. It was also necessary to achieve rupture of the algae cell wall prior to feeding to non-ruminants (such as pigs and poultry).

Raw wastewater was stored overnight in holding tanks and then discharged into 4 fibreglass reinforced sedimentation tanks which had an overflow rate of 1.3 m/hr and a detention time of 1.5 hours. Table 1 gives the summarized values of several parameters for the raw wastewater and settled wastewater in 1981.

Table 1: Summary of Raw and Settled Wastewater Characteristics for 1981.

Parameter	Raw Wastewater	Settled Wastewater	% Removal
TTS mg/l	12955	5308	59
TSS mg/l	10713	3414	68
VSS mg/l	7594	2654	65
BOD mg/1	4026	2696	33
COD mg/1	8585	4096	52

The removal rates for the parameters were quite consistent over the years. Average removal of solids was below that reported for Phase I due to:

- a) no dilution water was added so the level of suspended solids was higher and
- b) enforcement of zero discharge by the government necessitated the recirculation of old wastewater.

1. High Rate Pond Operation, Productivity and Waste Treatment

Pand Depth

Experiments carried out to establish the optimal pond operating depth found 200 to 300 mm to be the best. Dissolved oxygen measurements peaked at levels of 15-18 ppm during maximum sunshine but remained near zero during the night. The dissolved oxygen level was very sensitive to periods of cloud cover.

Mixing

A regime of intermittent mixing during the day was compared to continuous mixing at a speed of 12 cm/sec mid-pond velocity. For predator control, the pond was continuously mixed at night. Power consumption was 30% less with the intermittent mixing. However, it was not recommended as some algae (Micractinium and Oocystia) tended to settle during the 3 hour quiescent period. This allowed the filamentous algae, Oscillatoria, to float on the surface and block out the penetration of sunlight to sub-surface strata.

Detention Time

Further experiments were conducted to compare 4 day and 6 day detention times. No distinct advantage could be attributed to the shortening of the detention time to four days. Biomass concentration was initially lower in the ponds with a 4 day detention time, although productivity improved towards the end of the year. However, at a 4 day detention, the algal culture became more prone to infestation with smaller algae species making harvesting more difficult.

рΗ

Together with dissolved oxygen, the pH was very important in determining the general status of the pond. Sudden changes in pH usually indicated shifts in algal culture and also an impending massive algal die-out.

High pH (> 7.5-9.0) tended to favour <u>Spirulina</u>, <u>Chlorella</u> and a very small species, <u>Synecohocystis</u>. pH values ranging from 6.90-7.5 favoured <u>Micractinium</u>, <u>Oocystis</u>, Oscillatoria and Scenedesmus.

Loading Rates

Since loading rates were important in determining treatment efficiency, these were manipulated and studied. Total solids (TTS) analysis on the settled waste in the morning was used to determine the feeding time required for the ponds

$$T = \frac{L_r \times 50,000}{V_c (112.0 + 0.395)}$$

where T = time in minutes

 $L_r = loading rate in kg BOD /ha/day$

V = feed pump rate (1/min)

S = total solids (TTS) in mg/1

Figure 1 shows an example record of the loading rate for pond D in 1981. Variability in BOD loading from nearly 0 to nearly 600 kg/ha/day can be observed. It was found that the ponds can take high shock loads of even up to 800 kg BOD/ha/day for a short period without adverse effects.

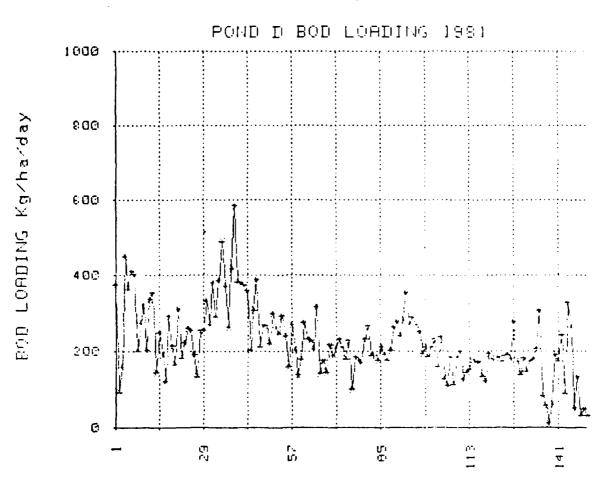


Figure 1: Pond D 800 Loading Rate 1981

SAMPLES

Generally, under normal operating conditions, the ponds can be loaded between 300-400 kg BOD/ha/day. Higher loading rates seemed to favour the faster growing algae like <u>Chlorella</u> and <u>Micractinium</u> while lower loading rates favoured <u>Ankistrodesmus</u> and <u>Raphidium</u>.

Treatment Efficiency

Treatment level of the high rate ponds was dependent on the harvesting method employed.

Table 2 gives the mean comparison of the TSS, 80D and COD levels of the different methods of harvesting.

Table 2: Effluent Quality Employing Different Techniques of Harvesting.

	TSS mg/L	BOD mg/L	COD mg/L
Pond Water	820	-	-
Centrifuge	350	136	291
Removal %	57	-	-
Pond Water	405	_	-
Filtration Harvester	106	138	276
Removal %	74	-	-
Pond Water	864	-	-
100 mg/l Alum/DAF	116	70	129
Removal %	86.3	-	-

The different harvesting techniques gave slightly different effluent results, with coagulation and flocculation with alum achieving a higher harvesting efficiency and a clearer effluent.

Pond Productivity

Productivity levels in the ponds varied substantially during the study period (1980 and 1981). Levels of up to 40 $g/m^2/day$ were achieved during sunny weather whereas during the rainy season, the productivity level dropped to below 10 $g/m^2/day$. Therefore pond design must take the large variation into account. Loading rates should be lowered during adverse periods in order to maintain acceptable levels of wastewater treatment.

Productivity also depended on the species dominance in the pond as well as on the harvesting technique.

Harvesting

- 1. Centrifuging: the disc centrifuge (Alfa Laval Model BRPX207SGT/P) was found to be effective in harvesting algae with recovery in exess of 95% and a slurry concentration of 4-5% solids. However, high power consumption (1.3 kwh/m³ pond water), high maintenance cost and the need for constant attention made this method uneconomical and not readily applicable.
- 2. Dissolved Air Flotation: the process consisted of chemical treatment to cause flocculation of the algal cells and then the introduction of fine air bubbles which adhered to the algae masses causing them to float to the surface. The algal float was then slowly scraped into a collection trough.

A pilot scale dissolved air flotation unit was constructed. Different factors such as loading rate, air to solids ratio, hydraulic detention time, etc. can be manipulated to optimize operating performance.

Different methods to achieve flocculation were tried, i.e., alum, polyacrylamide polymers, chitosan and pH adjustment. The most promising method was found to be chitosan, a substance easily prepared by acid hydrolysis of the chitin from the crustacean exoskeleton.

- 3. Autoflocculation: this is a term used for the settling of algae with the help of natural coagulation or bioflocculation without addition of chemicals. Algae appear to autoflocculate under stress conditions. Intensive sunlight, high pH, and low nitrogen levels have been associated with autoflocculation. However, attempts to achieve autoflocculation proved unsuccessful and were not continued.
- 4. Filtration Harvester: The filtration harvester, designed by Dr. Dodd has been described in Phase I of the Piggery Waste Treatment project. Performance evaluation experiments were conducted during 1980.

It was found that performance depended on a number of operating parameters such as belt speed, washing technique, water pressure and nozzle type and also the algae population dominant in the pond.

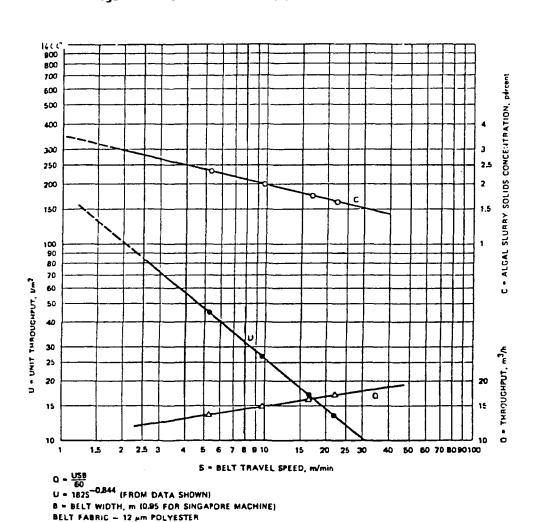
The ease of harvesting depended upon the species present in the high rate pond. With Micractinium and Oscillatoria dominant, a filter fabric with a 12 um nominal pore size could be used. When smaller species, such as Chlorella and Oocystis were dominant, a 5 um filter fabric was used.

Energy requirements per cubic meter of pond effluent harvested were compared for the different harvesting systems:

	energy requirement	slurry solids
		concentration
	kwh/m ³	%
disk centrifuge	1.4	4-5
belt filter	0.3	1-3
dissolved air flotation	1.2	4-4.5

Figure 2 shows some of the performance relationships of the mechanical harvester.

Figure 2: Performance of the Mechanical Harvester.



5. Biological Harvesting: Biological harvesting of algae using natural predators such as Moina and Daphnia was investigated. These predators are valuable as wet feed for fish in the aquarium fish industry. A small experiment was conducted in a 100 m² pond area with a 4-6 day detention time. Average production achieved was 4-5 kg/day of Moina on a wet weight basis. However, the stability of the culture over a long period is unknown.

Post-Harvest Processing and Utilization

Harvested algae deteriorated within 24 hours so it had to be used or processed quickly. Several different processing methods were tried.

Drum Drying -- the algae slurry was sprayed on a rotating, heated (110-120°C) drum and then dry algae flakes were scraped off. A final desired moisture content of 8% or less was achieved.

Results from 3 years of operation showed that the drum drier was not a viable method of algae processing due to the high energy consumption and the slow and labour intensive process.

Steam Cooking -- this alternative method was explored and the algae used in wet feeding trials. However, follow-up feeding experiments showed a lower digestibility and growth rate of steam-cooked algae compared to the drum dried algae.

Feeding trials with pigs showed that algae replacement in the diet did not cause pathological problems to the animals or inhibit growth. However, the use of algae derived by different harvesting techniques showed marked differences in feed conversion ratios.

Feeding trials with poultry showed that high level supplementation (20% or more) affected the growth rate and the feed conversion ratio. Centrifuged or filtration harvested algae followed by drum drying gave best results. Residual aluminum from chemical flocculation of algae adversely affected growth rates and feed conversion ratios.

Fish feeding trials showed encouraging results, especially for prawn farming.

Economic Analysis and Viability of the Algae System

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At the end of the four years of research (phase I and II) an economic analysis based on actual 1981 quotations in Singapore dollars was performed. The analysis was based on a hypothetical 30,000 standing pig population producing 600 m³ wastewater/day (Figure 3).

With an investment of S \$4.4 million (US \$2.1 Million) on a 6.3 ha land area, the total waste can be treated for 15 years with an internal rate of return of 18%. Figure 4 shows diagramatically the primary treatment system and Table 7 shows the costs and benefits of the high rate algae pond system.

Figure 3: Design data for a hypothetical 30,000 pig farm.

30,000

Setting and anaerobic digestion

Standing pig population (SPP)

bedridaring pag population (o)	,
Total live weight (TLW) per pig, kg	54
Total volatile solids (TVS) per day per 100 kg TLW, kg	0.54
TVS production for 30,000 pigs, kg/day	8,750
TSS removal by settling, percent	85
TVS to digester, kg/day	7,440
Biogas production, m ³ /kg TVS added	0.4
Total biogas production, m ³ /day	2,980
High rate ponds (HRP)	
BOD production per day per 100 kg TLW, kg	0.20
BOD production for 30,000 SPP, kg/day	3,240
BOD removal by settling, percent	45
BOD to HRP, settled wastewater, kg/day	1,780
BOD to HRP, digested sludge dewatering sidestream, kg/day	440
Total BOD to HRP, kg/day	2,220
Loading on HRP, kg BOD/hectare/day	350
HRP area, hectares	6.3
Biomass productivity, g/m²/day (dry weight)	20
Biomas production kg/d (dry weight)	1,260
Predominant genera	Micractinium
HRP effluent suspended solids, mg/1	500
HRP effluent flow, m ³ /hr	100

Figure 3: Design data for a hypothetical 30,000 pig farm. (con't)

Fine-weave belt filter

Belt width, m 3
Filtration drum diameter, m 2.3
Number of units 3 (including one spare)
Flow per unit, (based on two units operating continuously), m³/hr 55
Algae slurry solids, percent 2
Total algae slurry flow, m³/hr 2.5

Centrifugal dewatering

Type solid bowl decanter

Number of units 1 (plus spare components)

Flow, m³/hr (continuous operation) 2.5

Algae cake solids, percent 16

Cake production, wet kg/day 7,100

Scale drying

Type Pulse jet, Sonodyne, single engine Fuel sources Digester gas (65 percent methane)
Number of units 1 (plus spare components)
Capacity, wet kg/day (16 percent cake) 21,800
Scars of operation/day 8
Algae production, kg/day (10% moisture) 1,200
Digester gas requirement, m³/day 1,100

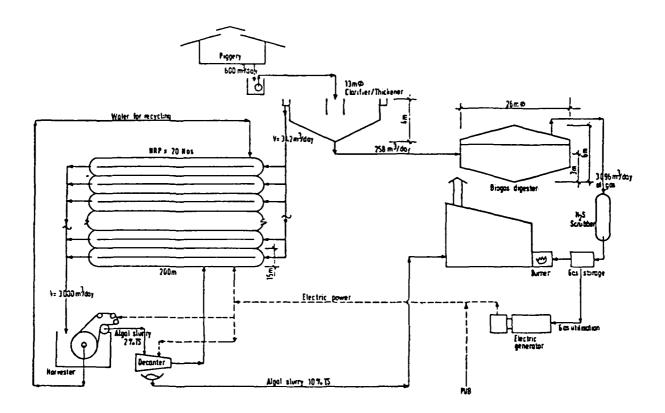


Figure 4: 30,000 pig farm-algae production system flow chart.

The bulk of the capital costs was for pond construction (36.4%) and for the purchase of harvesting equipment (37.9%). Other significant costs included labour, vehicles, power and land rental.

The benefits to be derived were the value of the algae produced, the treatment of the pig wastes and the recycling of the treated water. Algae yields varied considerably over the year so the analysis assumed an algae biomass productivity of 20g/ m/day and that 88% of this was harvested and processed. The 88% yield level of 17.6 g/m /day was equivalent to a yield of 64.24 tons of dried algae/ha/year.

Sensitivity analysis showed the important functions to be the value of the waste treatment, algae and power generation. Any adverse effect on these variables will have a negative impact on the economic viability of the system.

Table 7: Cost and Benefit of a High Rate Algae Pond.

	Year										
	1	2	3	4	5	6	7	8	9	10	11-15
<u>COST</u> (S\$1000)							1				
Capital investment	2,204	2,204	_	-	-	-	-	-	-	-	-
Maintenance	-	-	95	190	190	190	190	190	190	190	190
Labour	78	156	111	246	246	246	246	246	246	246	246
Vehicle, maintenance	1	\		,		1	ļ	1	1	1	ļ
and equipment	60	26	147	42	82	42	42	42	42	187	42
Power	-	\ - '	21	42	42	42	42	42	42	42	42
Land Rental	40	40	40	40	40	40	40	40	40	40	40
Total Cost	2,382	2,426	525	560	600	560	560	560	560	705	560
BENEFIT (S\$'000)											
Algae (tonnes)	_	-	192.7	385.4	385.4	385.4	385.4	385.4	385.4	385.4	385.4
Value of algae	i - '	_	164	328	328	328	328	407	407	407	407
Value of sewage	ĺ	<u> </u>							1	ţ	ļ
treatment	1,500	1,500	270	540	540	540	540	540	540	540	540
Value of treated water	-	-	80	160	160	160	160	160	160	160	160
Total benefit	1,500	1,500	514	1,028	1,028	1,028	1,028	1,107	1,107	1,107	1,107

Assumptions: Yields of 20 g/m²/day for 365 days. Assuming harvesting rate of 88% of total algae. Construction in years 1 and 2. Plant operating at 50% of capacity in Year 3. Value of sewage treatment - \$100 per SPP investment cost in years 1 and 2 and \$18 per SPP annual operating cost. Annual vehicle maintenance cost at 15% of value of vehicle.

DISCUSSION

The treatment and nutrient recovery from wastewater produced by a population of about 1,300 pigs was achieved using a high rate stabilization pond system. The project gave special emphasis to the design and construction of facilities for the collection of the wastes and recycling of the treated wastewater as well as the production, harvesting and processing of the algal biomass.

The dynamics of the algae population in the high rate pond was found to be complex and difficult to control. At the same time, pond performance and the characteristics of the pond effluent greatly affected the harvesting and processing of the recovered solids.

The project developed an efficient and economical harvesting method - the belt filtration harvester. Since harvested algae deteriorate rapidly, they must be used or processed within a day. Different post-harvesting treatments were investigated. Either drum-drying the algae or steam cooking it was acceptable but more work is needed to determine which treatment best improved digestibility and feed value. Pig feeding trials showed algae replacement in the diet at 15% or less did not inhibit growth. Results of poultry feeding trials were not as promising.

Cost analysis showed the system to be viable for treatment of wastewater generated by a population of 30,000 pigs or more. However, rate of return on the capital investment for the high rate pond treatment and nutrient recovery system depended to a large extent on the value of the harvested algae as pig feed as well as the value of the waste treatment and recycled water.

Publications arising from the IDRC-supported Waste Reuse Project.

 Numerous technical reports on the facilities established and tested have been prepared by the Primary Production Department, Singapore.

LIST OF ABBREVIATIONS

ABS	-	sodium alkyl benzene sulfonate
AUF	-	anaerobic upflow filter
B00 ₅	-	five day biochemical oxygen demand
COD	-	chemical oxygen demand
C/N	-	ratio of carbon to nitrogen
DDT	-	dichlorodiphenyltrichloroethane
FC	-	fecal coliform
FS	-	fecal streptococci
нсн	-	hexachlorocyclohexane
HRP	-	high rate pond
LAS	-	linear alkylate sulfonate
MBAS	-	methylene blue active substances
MPN	-	most probable number
NH4-N	-	ammonia nitrogen
02	-	molecuar oxygen
PCB	-	polychlorinated biphenyl
PFU	-	plaque forming unit
p pm	-	parts per million
SS	-	suspended solids
SPC	-	standard plate count
TC	-	total coliform
TS	-	total solids
VS	-	volatile solids