DEVELOPMENT OF A TEACHING MODULE ON WATER QUALITY:

DEVELOPMENT OF A TEACHING MODULE

ON WATER QUALITY

THAT INVOLVES TWO LOCAL COMMUNITIES

By

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ABSTRACT

This project describes the development of microbiology laboratory exercises to be used in the teaching of first year undergraduates.

One of the main goals of this project, is to try to make these laboratory exercises relevant to the life of local communities. Two local communities were chosen in the Hamilton area. The communities of Hamilton Beach and Dundas were involved and their water was tested for the presence of faecal coliform bacteria.

The design of these laboratory exercises includes relevant aspects of educational theory intended to stimulate the students'interests in Biology and, simultaneously, strengthen the relationship between the students and their local communities.

The main conclusions drawn from this project are:

- 1. The primary objectives established for the course seem to have been fulfilled.
- 2. The majority of the students considered these laboratory exercises to be socially and educationally relevant.
- 3. It is important to stimulate dialogue amongst the students.
- 4. It is desirable to make educational programs immediately

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relevant to the lives and the experiences of the students.

RESUME

Ce projet, destiné à des étudiants de première année universitaire, a pour but le dévelopement en laboratoire de microbiologie, d'analyses bactériologiques à des fins pedagogiques.

L'un des objectifs principaux de ce projet a tenté de rendre ces analyses bactériologiques pertinentes à la vie des communautés locales. Deux communautés ont été choisies dans la région d'Hamilton, Hamilton- Plage et Dundas, qui ont plus précisement participé à des essais de qualité de l'eau, consistant en la recherche de coliformes fécaux.

La conception de ces analyses de laboratoire a sélectionné un nombre de méthodes éducatives, destinées d'une part, à stimuler l'interêt des étudiants pour la biologie, et, d'autre part, à renforcer la relation étudiants-communautés locales.

Les conclusions principales qui peuvent être tirées de ce projet sont les suivantes:

- L'objectif principal fixé pour ce cours semble avoir été atteint.
- 2. La plupart des étudiants ont jugé ces analyses de laboratoire éducativement et socialement utiles.
- 3. La stimulation d'une discussion chez ces étudiants est

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apparue très importante.

4. Il est à souhaiter que ces programmes éducatifs soient directement pertinentes à la vie personnelle et professionnelle de ces étudiants.

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This thesis is dedicated to my mother.

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CHAPTER I

Introduction:

1. Definition of The Problem:

a. Educational Problem.

Universities in many parts of the world suffer from a lack of modern sophisticated technical and scientific infrastructure. Simultaneously, they suffer from a barrier of communication with local communities, especially the most disadvantaged members. In third world countries, the latter class represents the majority of the population. One of the consequences is that universities, although they make strenuous efforts to improve the conditions of the poor, often do not seem to be relevant to the needs of their communities.

In trying to address this problem, schools and universities must respond to the needs of the community they serve. To do this, universities need to:

- take advantage of the learning resources available in the community.
- be sufficiently well equipped to foster the learning process, thereby enabling students to seek out, reflect upon, and help resolve community problems.
- provide training in skills which allow students to play

an active role in their local communities.

These principles are sometimes followed in universities of western countries, however, they have yet to be achieved in schools of developing countries. In Algeria, for example, there is a shortage of skilled personnel in many fields. This creates a problem of technological dependency, and leads to importation of a wide variety of skilled human resources to meet the needs of the people. Although a great effort has been undertaken by Algerian authorities to establish specialized agencies and extension services designed to introduce vocational training in various fields, the university curriculum must be revised in order to make it more relevant to community issues.

When I was a biology student at the university of Constantine (East Algeria), at the conclusion of each topic we questioned the relevance of what we had just learned to everyday life. Part of our education seemed unrelated to what we were expected to achieve in the working world. Even when laboratory sessions were available, they failed to overcome this deficiency. This was due to:

- the lack of contact between the university and the local community.
- inefficient systems for introducing and financing new laboratory technology.

These problems have resulted in dissatisfaction among

students with respect to their educational needs. Many students migrate abroad to pursue advanced studies. Many of them travel to France, or other western European countries, various states of Eastern Europe, and North America. Many students do not return home, and graduates who do return become alienated from their own society when they are unable to gain access to the type of employment they desire.

Universities need to introduce more relevant programmes in order to overcome students' frustrations and encourage them to participate in the life and work of their home communities.

b. Biological Problem.

Water is a very precious resource that many people take for granted. Water contamination and pollution problems have however, caused many people to become concerned. A great deal of work has been undertaken by Canadian regulatory authorities to improve the water quality in both the rural and urban districts of North America and some of the third world countries.

Many Canadian citizens and Algerian citizens are not aware of the hazards in their water supply and do not know how to address the issue of water quality. In Algeria, both drinking and recreational waters are often not safe especially in the small isolated villages where diarrhoea, cholera, and hepatitis are widespread. Despite efforts by

health services to improve health conditions, contaminated water and its associated health problems persist.

The coliform group of bacteria has been used as one of the principle indicators in determining whether water is polluted with faecal waste. High levels of this contamination can lead to serious health problems if left unchecked, and hence it should be monitored continuously. The classical human coliform indicator is <u>E.coli</u>, which is not necessarily harmful in itself but, if its density in a sample exceeds certain limits, can indicate a potential problem.

2. Educational Objectives.

This project attempts to address a socially significant issue, namely 'water quality', by designing and developing laboratory exercises for first year undergraduates. Through the relationship between scientific research and community involvement, water purity and quality can be improved and maintained.

The educational objectives on which this project is based, are derived from educational theories of the learning environment, as well as the nature of the subject matter.

These educational goals are:

- to promote interaction between the students and their

community through discussing the existing community water problems. This interaction will hopefully create awareness among the community residents about the quality of their water, as well as creating a social awareness amongst the students.

- to help students develop personal attitudes about themselves and their environment. Some attitudes which are conducive to scientific enquiry include:

- 1) Consideration for others
- 2) Concern and care for the environment
- 3) Curiosity.
- to assist students in developing laboratory skills. These skills include:
 - 1) Communicating: receiving and expressing information.
 - Experimenting : designing and carrying out experiments to investigate the problem.
 - 3) Interpreting: evaluating data and developing conclusions on the basis of the information collected.

3. Literature Review:

a) Content should be relevant.

Interest in the relevance of content has increased in recent years. There seems to be widespread support in the literature for arranging for students to go back and forth between school and community. There are three general principles prepared by Rassekh and Vaideanu (1987) for the relevance of content. The first is that schools need to be receptive to the problems and needs of the local and national community. The scale of world problems, requires this receptiveness to be wider and better organized.

The second general principle is to adjust the content to the intellectual and physical needs and abilities of the learner. The third and final principle is that the content should be capable of stimulating and sustaining the pleasure of learning.

One of the characteristics of the community school as it is developing in America, is that the students and teachers use the resources of the community as the core of the curriculum, relating the major content of the curriculum to what is happening and can be studied in the community (Sinclair & Lillis 1980).

A more radical approach to the use of community resources is taken by Bal et al, (1973). They advocate that there should be no boundaries between school and community and that individual students must serve the community on their own terms. Thus, they are not serving the community, but doing what they can do or enjoy doing, in ways or places which provide a service for others. Rosenstein (1972) supports this idea and shows that when teachers and

administrators begin to perceive the community as a learning resource for individuals rather than the entire class they are able to exploit the local community for meaningful educational experiences.

School experiences must be fused with community and work experience. On the other hand, community participation in the school is regarded by some as an instrument of power to effect changes in the school. This attitude is derived from dissatisfaction with school performance in recent years and the assumption that increased citizen participation in school affairs would improve that performance. Based on an idea of Salisbury (1980), this implies that citizens have demands to make regarding how the schools should be run and that if those demands were more clearly heard and better needed, educational performance would improve.

b) Educational Theories:

The design of these laboratory exercises is ideologically derived from a variety of educational theories which I have encountered during my learning and which have deeply affected my personal philosophy about the learning/ teaching process.

I. Integration of knowledge:

I believe that all knowledge should be integrated into a comprehensive picture. Biology, physics and psychology

should not be viewed as three segregated types of knowledge; they are very much related to each other. In any learning situation where the learner asks questions and seeks solutions, integration of science with other subjects will happen naturally. The educational theories which I think relevant and supportive to the present study are:

1. Holistic curriculum (Miller 1988):

This curriculum perceives learning in a holistic way without restrictions that might be imposed by subject area boundaries. The aim of this approach is to help educators integrate theory and practice in order to create an orientation to curriculum that is consistent with one's personal world view.

2. The Scientific Method:

Dewey's philosophy of education is based on the scientific method. He focuses on curriculum strategies that facilitate learning within a social context. Dewey's five step method starts with the confrontation of a problematic situation. In the second step, students define exactly what the problem is, make assumptions about an attribute that has not been directly observed, followed by analysis of the factors contributing to the problem. In the fourth step the students develop a hypothesis providing a possible explanation based on what has been observed. In the fifth and final step students select one hypothesis or alternative

and implement it. Boud (1985) calls this teaching strategy Problem-Based-Learning. It is considered as one form of learning from experience, or experience-based learning in which learning is focused on problems derived from practice. In other words, experience-based learning is any form of learning which places the experience of learners as central and uses it as the organizing principle of learning. However, solving these problems is limited within the boundaries of the academic institution. This educational context would be more effective if schools expanded the students' learning experience beyond the classroom to reach the needs of their local communities. This can be accomplished through school-community interactions and having students deal with a social or scientific problem under the actual circumstances.

3. Community Connections:

Miller (1988) and Miller and Seller (1990) stated a number of connections with the subject matter so as to render it more relevant to the real world. These connections are:

i. Students/Community:

Several programs have been established to encourage contact between students and their local community. It was suggested that schools should encourage student involvement in community services. They recommended that students have

opportunity to practice caring. The student should be assigned to hospitals and to participate in social change in order to develop an ability to effect his or her social environment. Examples of these activities are letter writing, participation in meetings and community visits.

Moreover, Davis (1981) stresses the necessity of citizens' participation as a way of strengthening public commitment to the maintenance of a healthy public school system. Citizens participate because they care and they want to have a say in significant issues.

ii. Subject/Community:

Academic subject can provide a bridge to a community, thereby transferring the needs of the community into the classroom and providing work in order to meet those needs. Subsequently, students will develop a sense of wholeness of themselves and the environment.

c. <u>Teaching Methods (practice):</u>

There are many teaching strategies that can be used by the teacher in order to achieve the aims of the program. Some of these strategies are:

i. Role playing:

Application of role playing, particularly socio-drama or social simulation in the classroom, would facilitate the learning process. It enables students to explore "real"

life situations and to gain awareness as they would during the actual situation. As Hayman (1974) described it, simulation games provide a means for making the topic at hand relevant in the present. Students can take on the role of others and become very involved in these roles as they experience various social processes and realities, and examine their own reactions to them. Unfortunately, as Bourne and Eisenberg (1978) point out, students do not always take role-playing seriously, especially when they have to hold unfamiliar positions. Perhaps this means that students should try to use more imagination to project themselves into unfamiliar situations and express what they would do if they were charged with various responsibilities

ii. Cooperative Learning:

The most immediate connection to the students is the classroom where cooperative learning takes place and facilitates the learning process. Group discussion for example, is an important teaching strategy. It helps students to seek for knowledge autonomously, to improve their self-image and communication skills. Glassers (1986) elucidated that the classroom meeting should be considered as a vehicle to enhance students self-esteem as well as to help them develop a sense of community in the classroom. Schwab and Evelyn (1968) asserted that class discussion facilitates learning and strongly promotes interpersonal

relationships.

iii. Social Projection:

Social projection is an educational program which has been developed in Latin America, particularly in the National University of El Salvador. This teaching concept requires that students spend time with communities researching the needs of the people. Together, in the classroom, students and community leaders discuss issues that are relevant to the development of those communities. Eventually, they attempt to find and implement solutions to the problem. A concrete example of these activities is seen in two years of theoretical instruction of a class of nursing students. The class is divided into groups, and each group selects a community to work with. Together, the students and the community executives diagnose a health problem in the community. Then the students take the health problem to the university to analyze it. The analysis of the problem usually involves other faculties within the university. Finally, a solution to the problem is proposed and the knowledge gained is used to modify the curriculum at the university (Austin 1992). This teaching concept summarises a number of educational strategies by which connections to the community can occur. These strategies are also outlined by Miller (1988). They are the following: - cooperative learning: the discussion of the problem in

the class.

 invitational learning: participation of community leaders in solving the community problem through their involvement and interaction with the students in the classroom.

d. <u>Water Quality</u>.

Most of us could live without medicine; but, no one can live without water. In fact, over half of the human body is water. If every one of us, especially those who live on farms or in small isolated villages, made the best use of water, the amount of sickness and death could probably be cut in half. For example, correct use of water is basic in both prevention and treatment of diarrhea. Contaminated water is often part of the cause of this illness.

Water pollution's greatest danger is derived from enteric pathogens that are excreted in the faeces of disease carriers. Although determining the concentration of pathogenic organisms is possible, it is an extremely laborious and inefficient process in regular water analysis because each species of pathogens requires separate incubation conditions and enumeration procedures. For example, a negative result when testing for typhoid does not necessarily mean the water is safe for human consumption because other undetectable pathogenic species may be present. Also, a water supply may be polluted by excreted contamination but have low or zero pathogens because there are no pathogen carriers in the faeces. A water supplier testing for only pathogens would remain unaware of the contamination. In an effort to overcome these difficulties, microbiologists have adopted the use of indicator species of enteric bacteria. The presence of faecal coliforms in water is an indicator of potentially dangerous contamination from human waste. For this reason the measurement of faecal coliform in water is an important test of community water quality.

The objective of this study was to evaluate the water quality of two local communities in the Hamilton area and to increase their awareness about the quality of their water. The communities are: Hamilton Beach in the east end, and the community of Dundas in the west end of Hamilton.

A similar study was proposed by Environment Canada personnel to examine quality of drinking and recreational waters of the Cree Nation of the Split Lake community located in Northern Manitoba. The project was conducted by Dutka and his colleagues (1990) who assisted the people of the community to become self sufficient in monitoring and controlling the bacteriological quality of their water. Intensive training of community residents in simple bacteriological procedures resulted in a remarkable increase

in community involvement. Residents were trained sufficiently to maintain and report on the bacterial level of their community drinking water.

Other similar studies have also been conducted by Dutka (1991). He developed appropriate simple bacteriological procedures suitable for developing countries. He suggested a number of tests that require inexpensive material and equipment so as to reduce the technical dependency and help improve the water quality in third world countries.

e. <u>Bacteriological Tests</u>.

Studies of microorganisms in natural waters involve enumeration as a general index of activity and as a measurement of bacteria in water and sediments. Cultural methods are most commonly used even though they often yield a small percentage of the microorganisms present. Several tests are performed to determine the sanitary quality of water. According to the standard methods, the principal test used for bacteriological analysis of drinking water and recreational waters is the five tubes most probable number test using A-1 broth. This procedure is very simple and does not require sophisticated instruments. Another test called the Presence/Absence (P/A) test has been used to examine the water quality in third world countries as well as rural and isolated areas of North America (Dutka 1990). Another test which has been developed by Guelin (1948) to determine the safety of water, is the presence of bacteriophage (viruses that infect bacterial cells).

The most frequent approach is the membrane filtration technique (Appendix B). This method is applicable to all types of water and waste waters. It is also recommended for evaluating recreational waters (beaches, rivers, etc..) and testing for the presence of faecal coliform. Moreover, it has a definite advantage over other methods in that it can test large volumes of water and produces more rapid results.

The tests described above are called presumptive tests They are quantitative, but do not provide enough information to identify the type of bacteria present in the water supplies.

Confirmatory tests for faecal coliform require further examination for acid and gas production by inoculation and incubation of lactose broth fermentation tubes, or by using diagnostic media such as MacConkey's media and mFC media (Appendix B).

Complete tests are usually performed to determine the characteristics of the organism isolated. These involve the Gram Stain and additional biochemical tests for specific types of organisms (Appendix D).

i. Definitions:

• Coliform: usually refer to those bacteria that are

normal inhabitants of intestinal tracts. They are gram negative, non spore forming, facultative anaerobic, rod shaped bacteria, capable of fermenting lactose broth with the production of gas within 48 hours at 35°C.

• Faecal coliform: similar to coliform but with the additional feature of being able to ferment lactose with the production of gas within 24 hours when incubated at 44.5°C.

Generally, there is more than one type of indicator bacteria used for determining the safety of water. These bacteria are also found to be present in the water samples tested during the course of these research project. These are <u>E.coli</u>, <u>Pseudomonas aeruginosa</u>, <u>Klebsiella</u> and Citrobacter. Strep fecalis is also a faecal coliform organism, it is a gram positive coccus. Staphylococci or faecal streptococci are also indicators for faecal contamination Dutka (1990).

• <u>E.coli</u>: Is an intestinal parasite that can live free in nature, and its presence (in high concentration) in water supplies is accepted as evidence of recent faecal contamination. <u>E.coli</u> is a common cause of diarrhea, and urinary tract infections (Creager et al,1990). It uses lactose as a carbon source; resulting in a formation of individual colonies of an intense purple colour when plated on MacConkey's medium or blue colour when plated on mFC medium APHA (1985).

• <u>Pseudomonas aeruginosa</u>: This type of bacteria and others, such as Klebsiella, are often responsible for chronic infections. Pseudomonas infections, are common in swimmers because the bacteria are resistant to hypochlorite. They are introduced directly into damaged tissue created by burns, injections and bites. Pseudomonas can also cause ear infections Volk (1984).

• <u>Klebsiella</u>: These bacteria are commonly found in the human digestive tract. Klebsiella pneumoniae, can cause severe pneumonia, it can cause diarrhea in infants, abscesses and urinary tract infections Dutka (1989) and Creager et al (1990).

• Citrobacter: These bacteria are not considered to be pathogenic (disease causing) even though, they have been isolated from individuals with diarrhea. They could be also isolated from urinary tract infections and superficial wounds Volk (1984).

• Bacteriophage: Are viruses infecting bacteria. By itself, a phage can persist, but cannot replicate except within the bacterial cell. Best studied of these are the bacteriophages infecting the human intestinal bacterium E.coli, known as "T even viruses" (T2, T4, T6). When T4 virus initiates an infection upon a layer of host cells growing on a nutrient agar, a zone of a growth inhibition may occur resulting in a clear area called "Plaque" Wolfe

(1983).

• Membrane Filtration Technique: This technique is being used to detect the presence of bacteria in water. The membrane filter is generally made of Cellulose Acetate or Cellulose Nitrate with a mean pore size of 0.45 microns. It is used by water bacteriologists to collect and enumerate the total coliform and faecal coliform bacteria in water and wastewater APHA (1985) and Dutka (1989).

ii. Growth Media.

The optimum growth of different bacterial organisms depends on the type of growth media. Many studies using total counts obtained from various synthetic media have indicated these counts to be much less then the total microscopic counts. This inconsistency may be due to the fact that many organisms are non viable, and that there is no single growth medium which would be suitable for growth of all organisms in a sample. For this reason selective media are used routinely in environmental microbiology. For example, on mFC agar (medium for faecal coliform), faecal coliforms produce typically blue colonies. On MacConkey medium faecal coliform produce red- pink colonies (Appendix B).

CHAPTER II

Methods:

The technical aspects of these laboratory exercises are of great importance. In the summer of 1991 laboratory equipment and material to be used by the students were prepared. All procedures were tested in advance so we knew what to expect in terms of results.

Equally important, the social aspects of the program had be developed. We contacted a testing agency (see below) to learn the bacteriological tests. We also contacted local communities to find out which ones were interested in the tests. The sequence of activities is summarized in Table 1.

METHODS

Table 1: Sequence of activities.

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HAMILTON BEACH (first term)	DUNDAS (second term)
 Contact with the testing agency. 	1. Contact with the testing agency was already arranged in the first term.
2. Design of laboratory exercises.	2. Design of the lab exercises was done in the first term.
3. Contact with the community.	3. Contact with the community.
4. Contact with Saint Mary's High School.	4. Contact with the Biology Department.
5. Teaching lab skills.	5. Teaching lab skills.
 Meeting with the community was more than once. 	 Meeting with the community only at the end of the project.
7. Collection of water samples.	7. Collection of water samples.
8. Bacteriological examination of water.	8. Bacteriological examination of water.
9. Role playing was not performed.	9. Role playing (Socio- drama) was performed.
10. Presentation of results to the community.	10. Presentation of results to the community.

Phase 1 of the project: Preparations.

1. Contact with the Testing Agency:

The Public Health Laboratory in Hamilton was contacted first. This allowed me to identify existing community water problems and to determine how they are routinely diagnosed, tested and evaluated.

2. Designing the Labs: (Appendices A, B, C,D)

With information from this contact, a suitable experimental design for the students was developed and tested. This included preparation of bacteriological media suited to the general growth of bacteria (Luria broth), as well as selective media specific for identification of groups of faecal microorganisms (MacConkey's medium, and medium for faecal coliform (mFC)). A membrane filtration technique was also adapted for our use.

The laboratory manual was designed to introduce the students to basic microbiology as it is practiced in the McMaster Department of Biology, with slight modifications in order to meet the project's educational objectives. Each lab included questions related to particular experiments to facilitate the students' participation in the class.

3. Initial contact with a Local Community:

A target community that has a perceived water problem

was contacted. The Beach Boulevard is part of the Hamilton Beach located in Ward 5. The president of The Beach Preservation Committee, Mr. Bob Jaggard, was contacted. Arrangements for meetings with the interested members of this community were made. At these meetings a water sampling schedule was agreed upon.

4. Contact with Students:

Since laboratory sessions for first year undergraduates could not be offered by the Biology Department during the fall, these students could not participate in the research project. Therefore, we contacted Saint Mary's High School in order to find some student volunteers. After they were introduced to the topic, the principal, the biology teacher, and the guidance counsellor were very enthusiastic. However, they did not show as much interest in the social aspects of the project as in the scientific aspects. They were mainly interested in having the students attend the laboratory setting within the university. The students were told about the project by their science teacher, and five students volunteered to participate.

In the second term, the program was implemented with first year Biology students from McMaster University. A Dundas community was selected for this term through contacts with Mrs E. Shannon, leader of a Dundas group of

environmentally concerned citizens. Contacts with the testing agency and both communities were established through letters (Appendix F) and community visits.

Phase 2 of the Project: Work With Students.

After developing the laboratory procedures, establishing contact with the local community and finding the high school students volunteers, the next steps were to train the students and introduce them to the community representatives. During this first meeting, I attempted to generate an atmosphere conducive to dialogue by encouraging the students to participate actively in this inquiry. Two out of the five students displayed serious interest in both social and scientific approaches. They became even more enthusiastic upon meeting members of the local community. In the second term, twenty five Biology students joined the research project as part of their scheduled course. The students were required to write two reports: the first included the testing procedures and the second one included the actual water testing experiments combined with the social interaction. The standard method of writing a scientific report was also introduced.

1. Laboratory Skills:

The students were made aware of laboratory safety
regulations and procedures, such as the proper handling of bacterial samples, the sterilization of filtration units, pouring plates and disinfecting and sterilizing tubes, bottles, etc.. (Appendix A). They learned how to make and use general bacteriological media as well as selective media suited for the diagnosis of certain groups of microorganisms. The students also learned how to perform calibrations of the tests, which included two parts.

In the first part, students learned how to count <u>E.coli</u> in samples specifically prepared to teach this skill. Through this test, dilutions were made which, when plated, gave rise to 20-50 colonies. Students learned the usefulness of the test and how to do dilutions, to plate samples from their dilutions, to count colonies and to extrapolate the results to initial sample concentration.

In the second part of the laboratory, students learned how to identify <u>E.coli</u> using growth tests on selective media (mFC & MacConkey's medium). They also learned to use the very specific virus T4 that infects a very restricted group of <u>E.coli</u> by lysing them. Finally, students learned to calibrate the test they were going to use in the field, by mixing a known number of <u>E.coli</u> cells with a standard sample volume of water and filtering the sample through a membrane that traps bacteria. The filter is then placed on top of solid medium (mFC). Each trapped bacterium grows into one

colony, which, with this selective medium, has a characteristic colour and appearance. The purpose of the above experiment was to examine the recovery of bacteria from the filtration procedures. In this way students learned how to count and identify <u>E.coli</u>, as well as how to use a calibrated test to detect the microorganism in a water sample (Appendix B).

2. Field Work: Meetings With the Local Community.

The purpose of this part of the project was to have students establish a rapport with members of local communities and gain awareness of their existing water problems.

The municipal election campaign was in progress and we were invited to a meeting in which candidates running for alderman, for mayor, and for regional chairman were present. This made it interesting for the students as they could listen to the community problems and solutions suggested by the candidates. However, most of the discussion was focused on another issue (a proposed highway through the Red Hill Creek Valley). This discussion caused one of the students to raise questions about school/community involvement and how to initiate contacts. The water quality issue was also raised, and a few members of the community were highly concerned. At the conclusion of the meeting, students had the opportunity to speak with several community members who provided them with more information about the various existing local problems. The beach area residents at the meeting supported the idea of the project. They said they were willing to cooperate and seemed eager to learn more about the water quality in the area.

In the second term the Biology students did not have an opportunity to meet with the Dundas community prior to the initiation of the research. They met with the community members only at the end of the project when they had to present and discuss the results.

3. Collection of Water Samples in Field:

Sampling of the first body of water was undertaken jointly by community members and students. The former indicated what and where they wanted to sample, the latter indicated how this was to be done scientifically. It was possible to sample four times in the Beach Boulevard area Fig (1). The temperature during this period of time (Oct-Dec) ranged from -4 to 8 degrees centigrade. Samples were taken from various points at about 20 feet from the shore. A total of 24 samples of 100 mL were collected in sterile bottles. Finally, the students took the samples to the university to analyze them using the knowledge they had acquired in the earlier laboratory exercises (Appendix B).

In the second term, water samples were collected from two locations: Olympic Drive (At the Dundas end of the body of water parallel to Cootes Drive) and two sites in Sydenham Creek. Site A was on Alma Street. Site B was on King Street next to Little Caesar's restaurant Fig (2). Students filled sterile 200 mL bottles with stream water at each of these locations and brought eight samples to the lab at McMaster University. Samples of 100 mL, ten, one and one tenth of a mL of the original water collected, were mixed with distilled water to a final volume of 100 mL and filtered through membrane filters of a mean pore size of 0.45 u. The filters were then placed on MacConkey's medium or mFC plates and incubated overnight at 44.5°C. Standard methods were employed in all analytical work. The bacteriological examination of the water samples was started immediately after their collection.

4. Examination of Water Samples (Appendix C):

The third laboratory session was a follow up study of the previous lab. Bacterial colonies from the water samples were counted, and a series of diagnostic tests were conducted. These included the Gram Stain procedure. Results were then discussed jointly with the testing agencies (The Public Health Laboratory, and the Microbiology Department of McMaster University Hospital). The students

returned to the community with their results, charts, petri dishes...etc, and discussed these with interested community members.

5. Calculation of bacterial densities:

After the appropriate incubation period, the bacterial colonies were ready to count.

Bacterial densities were calculated as colonies per unit volume of samples.

Bacterial Density= <u>Number of Colonies x Reporting Volume</u> Sample Volume Filtered X Dilution

6. Social Simulation (Socio-drama):

Having obtained the results, and prior to presenting the findings to the community, a useful teaching technique called 'Socio-drama' was used. The purpose of this technique was to enable the students to simulate real life situations and to gain awareness of some of the social issues present in the actual situation. The class was divided into two groups: one group took on the role of the community members, while the other group learned how to present and discuss the data in a concise way, so that the community members could understand easily the technical part of the study. This teaching approach was applied only in the second term with the undergraduate Biology students.

7. Evaluation of the Project: (* Also see chapter IV)

At the end of the study, questionnaires were distributed (Appendix E). The students were asked to respond to ten questions. Sixteen students responded and nine did not. The results of the questionnaire are summarized in table 5.

8. Evaluation of the Students' Achievement:

The evaluation of the students' learning was based on how well they performed the experiments and whether they gained some knowledge about the water quality. This was accomplished by evaluation of their reports.







CHAPTER III

Results:

1. Biological Results.

a. Beach Boulevard area:

Most of the water samples showed bacterial colonies in high numbers and in a wide variety (Table 2). The numbers of bacterial cells ranged between 74-150 colonies per 100 mL, which exceeds the acceptable range (60 cells per 100 mL), indicating water contamination with faecal coliforms. These results seemed to be representative because the sampling was frequent at the same location and at different times. In each of the tests we found a large number of non faecal bacteria in the samples. These were not counted or characterized.

Further tests for identification of bacteria were limited to the Gram Stain method, which showed a mixture of three types of bacteria. By itself, the Gram Stain is not enough to provide final identification of organisms. It would have required a series of biochemical tests to examine the bacteria in more detail. However, one bacterial colony was identified as <u>Pseudomonas aeruginosa</u> when plated on ordinary growth medium (ie Luria broth). As it grows, this organism produces two compounds, Pyocyanin and Fluorescein,

which colour the culture blue-green. The plates were taken to the Department of Microbiology for further identification and the organism was confirmed to be <u>Pseudomonas</u> <u>aeruginosa</u>.

These results were only preliminary and we did a follow up study jointly with the Dundas community for further examination of water quality.

<u>**Table 2</u>:** Recreational water quality study of one area in Hamilton Beach: Beach Boulevard.</u>

N° of samples	Date	Volume (mL)	[*] Counts/100 mL
6	Oct 18th 1991	100 (mL)	Too many cells
6	Nov 5th 1991	100 (mL)	150 (170-69)
6	Dec 6th 1991	100 (mL)	74 (89-58)
6	Dec 12th 1991	100 (mL)	77 (96-67)

* Number of colonies per filter (highest-Lowest) Each number represents the mean of six water samples. Recommended maximum countable colonies for faecal coliform (60) APHA (1985).

It can be seen in Table 2 that the numbers of faecal coliforms (blue colonies) are comparable in the four different sample sets. The number of cells in the two first samples were higher than in the last two samples. This might be due to the fact that the water temperature of the beach during the first two sampling times was high. (Bacterial counts increase with temperature).

b. Dundas area:

Testing water for faecal coliform in Dundas was done at two locations indicated by Mrs E. Shannon.

Location 1: Olympic Drive at the Dundas end of the body of water parallel to Cootes Drive.

Location 2 A&B: Sydenham Creek. Site A) opposite to #3 Alma Street in Dundas. Site B) next to Little Caesar's restaurant at 149 King Street East in Dundas. The results are shown in Table 3. There is obviously a great deal of variability. However, all the samples tested exhibited a larger number of faecal coliform than the acceptable number (60 colonies per 100 mL).

It was not possible to derive very meaningful conclusions from the samples taken from location 1 because the water fowl population there was very high at the time of sampling. The samples from sites 2A & 2B, however, are probably representative. In addition to faecal coliform there were a large number of other bacteria in the samples. They were not counted or analyzed further.

<u>Table 3</u>. Water quality study of three locations in Dundas: Olympic Dr, Little Caesar's, and Alma St. Number of faecal coliform colonies (blue colonies on mFC) per filter (highest, lowest count).

Volume (mL)	Olympic Dr.	Little Caesar's	Alma St
100	too many to count	too many to count	too many to count
10	=	68 (108-41)	95 (129-72)
1	=	1	6
0.1	38 (80-17)	1	1

The numbers are the average class results. The range of these results is shown in parentheses. Standard methods were carried out as described in APHA (1985).

Ten faecal coliform colonies from one of the plates containing a filtered sample from 10 mL of water from site 2B were tested further in order to identify the organisms more precisely. The results are shown in Table 4.

It can be seen in Table 4 that we observed two colonies that contained E.coli, two that contained Klebsiella and four that contained Citrobacter. Our observations are complicated by the fact that the colonies tested were not purified before testing and may thus contain more than one kind of cell.

We performed a biochemical test that is specific for <u>Pseudomonas aeruginosa</u> on the 10 colonies mentioned above and on one of the colonies from the Olympic Drive sample. Samples were plated on Christens's modified acetamide agar (Appendix D). The 10 colonies described in Table 4 were negative in this test but the Olympic Drive sample was positive, indicating the presence of <u>Pseudomonas aeruginosa</u> in it.

From our results, it would appear that the level of faecal coliform in Sydenham Creek is above the acceptable limit for recreational waters (of 60 per 100 mL (APHA 1985). Our sampling was very limited. We sampled 3 locations on one day. It would be desirable to sample more locations and at different times to confirm these results.

We realize that our sampling and identification were

very limited and tentative, but these results are indicative of a faecal contamination problem which should be examined in more detail.

Sample	Indole	Simmons' Citrate	Inositol	Gram Stain	Possible Identity
1	-	+	+	(-) Rods	Unknown
2	+	+	+	(-) Rods	Klebsiella
3	+	-	+	(-) Rods	Unknown
4	+	-	-	(-) Rods	E.coli
5	-	+	-	(-) Rods (+) Cocci	Citrobacter Unknown
6	-	+	-	(-) Rods	Citrobacter
7	-	+	-	(-) Rods	Citrobacter
8	+	-	-	(-) Rods (+) Cocci	E.coli Unknown
9	-	+	-	(-) Rods	Citrobacter
10	+	+	÷	(-) Rods	Klebsiella

Table 4: Identification of bacteria found in 10 colonies from one of the filtered samples of site 2B.

2. Results of the Social Interaction.

a. Hamilton Beach:

Throughout the duration of this research project, a concerted effort was made to involve this community through representation at regularly scheduled community meetings. These meetings provided a forum to raise citizens' awareness level of the importance of water management and the overall progress of this study. The majority of community involvement was derived from active members of the Hamilton Beach Clean-Up Association. A great deal of contact was made with Mr. Jaggard who is very active in the Beach Clean-Up program. He was extremely interested in the tests because he lives on Beach Blvd. and is very concerned with the water quality which affects many facets of life within the community. At the end of the project, another appointment was made to discuss the results. The community members were very concerned by the presence of high levels of <u>Pseudomonas aeruginosa</u> and the staphylococci organisms. They agreed that further experiments should be conducted through a continuous monitoring of bacterial levels.

b. Dundas community:

Even though the interaction between the students and the community was limited, there was concern and interest expressed by both of groups. A meeting was organized in the

lounge of the Biology Department. Mrs Shannon and five other members of the community were present. Interest and concern about water quality were shown by the various questions they asked about the bacteria found in the water samples and the health problems that they might cause. The Dundas community members seemed to be well informed about their environmental problems and they were quite aware of the harm that their contaminated water could cause if not controlled.

Overall, both communities were made aware of the quality of their water and felt that, as citizens they should have a say in what happens in their environment.

c. Students' involvement:

Generally, students seemed to have a positive reaction towards the course. They were interested in meeting with the Dundas community through their spontaneous involvement in the discussion. When presenting the results, the students expressed a great deal of concern. Some of them were so interested in the tests that they wanted to proceed further with them.

CHAPTER IV

Critical Evaluation:

Scriven (1967) and Weiss (1972) distinguish between two forms of evaluation, formative and summative evaluation. When assessing is done as an ongoing aspect of learning activities, it is referred to as formative evaluation. When done on completion of a program, it is referred to as summative evaluation. In both cases it provides information about one's effectiveness and success in reaching the desired objectives. In the context of the present project, both types of evaluations were performed.

a. Evaluation to determine the worth of the programme:

At the end of the project questionnaires with ten questions were distributed (Appendix E). Sixteen students responded and nine did not.

The results of the questionnaire are presented in Table 5, from which we drew the following conclusions:

For question 1, six students strongly agreed and seven agreed that they had enjoyed the experience and gained a better understanding of scientific methods. Students were generally positive.

Students reported that the objectives and introductions

prior to each lab session helped them to understand the purpose of the experiment.

According to the answers to question 4, one student strongly agreed and seven agreed that the social interaction was useful. These are the responses of the students who came to the meeting. This does not mean that students who did not come were not interested, because we can see from students' reaction to question 5 that fifteen students had a positive reaction towards the community aspects of the project. For question 6, a similar number of students feel that the university/community interaction is an important part of science education. The answers to this question include those of the students who did not come to the meeting and were only informed about it.

From the students' comments in response to the question "what did not work well in the course?", it is clear that the majority of the students were not satisfied with the equipment. In fact, only one membrane filtration unit was available for the whole class and that made it inconvenient for the students to run their tests individually and in a brief time period.

The most effective laboratory exercise, according to the students, was the last one when they went outdoors and collected water samples, then presented their results to the community. However, some of them mentioned that this step

needs more organization since the students were not told about the time and the location of the sampling until the actual day of the tests. Table 5: Evaluation by students to determine the worth of the programme.

Item	Strongly	Agree	Neutral	Disagree	Strongly
	Agree			_	Disagree
The laboratory skills I learned					
in these labs have given me	6	9	1	0	0
a better understanding of what					
scientific experiments are like.					
The objectives and intro-					
duction at the begining of	7	7	0	2	0
each lab were helpful to me					
My impressions of the use-					
fulness of these labs have	3	11	2	0	0
improved since joining this					
research project					
For those who interacted with					
the local community	1	7	0	0	0
As a student my interaction					
with the community was useful					
I feel that the project has					
given me an understanding of	5	10	1	0	0
the community needs for water					
quality					
I feel that the university					
and the community interaction	7	8	1	0	0
is a valuable part of science					
education					

b. Assessing Progress and Outcomes:

Generally most of the students found that these lab exercises raised their interest in Biology and increased their awareness about water problems in the local community. Regardless of the lack of equipment and the limited social interaction, the students' curiosity about water quality increased. In fact some of them brought water samples from their homes to test. This shows a continued interest in the subject related to their everyday lives. The objective of developing personal attitudes about themselves and their environment was met quite well.

The aim of developing laboratory skills was achieved in that students learned some basic procedures of a microbiology lab, for example, preparation of culture media, pouring plates, counting bacteria and preparation of their own glass spreader. By the end of the course, the students mastered several different techniques. This was indicated by the quality of their plates and the way they conducted their experiments during the course of the project.

In terms of communications, and as mentioned above, the social interaction was limited but the students were interested in this aspect and they suggested that it needs to be developed in the future through splitting the class into groups and having each group meet with one community.

c. Evaluation of students' achievement:

The students' knowledge was tested by asking them to write lab reports during and after the completion of the project. The learning objectives were somewhat better achieved with the Biology 1G6 students than with the high school students. This was mainly due to the project being part of a credit course for the former, while the latter were volunteers. Only one student displayed a continuing interest and in fact made it part of her science fair project.

Evaluation of the students learning was based upon two criteria, 1)Knowledge 2)Interest and involvement. Based on their reports, the majority of students seemed to gain a fair amount of knowledge about basic bacteriology. They learned how to count the bacterial cells in a sample and how to use diagnostic bacteriological media. Three students (those received B- and C. Refer to table 6) did not seem to understand the basis of the diagnostic test we used, which uses lactose to study bacteria metabolism. The average class score in the first report was (83.92 %) and in the second report was (75.52 %). Grades ranged from C to A+ refer to table 6.

Grade	A ⁺	A	A-	B*	В	B	С
Students	1	6	7	5	3	2	1

<u>Table 6</u>: Students learning achievement.

Each grade represents the mean score of the two laboratory reports .

CHAPTER V

Conclusion:

Undergraduate laboratories tend to be a major university expense, however, they rarely evoke great enthusiasm among undergraduates, particularly in large introductory courses. This is due to the lack of social relevance of these exercises and not because the laboratories are insignificant in teaching experimental sciences. Very rarely is the social relevance of a teaching laboratory discussed in class, and seldom is its relevance to the wider non-academic community discussed.

In this study we attempted to design and develop laboratory exercises for first year undergraduate Biology students and we tried to make these exercises relevant to the needs of two local communities. A teaching module was prepared and implemented. Students were trained on the required procedures and they learned the necessary tests for water quality. Students' contact with the local communities was arranged before and after the testing.

The students collected water samples from the Beach Boulevard in Hamilton Beach and from two different locations in the Dundas area. Enumeration and isolation procedures of bacterial organisms were performed. Using the membrane

filtration technique and selective media specific for faecal coliform, quantitation and identification of bacterial organisms was done. The main conclusions drawn from this research project showed that the water quality of both communities is unsatisfactory due to the high bacterial counts, which are higher than the acceptable limit. Identification of individual organisms using Gram Stain and a series of biochemical tests (Indole test, Simmon' citrate and Inositol test) showed the presence of a number of bacteria. Those organisms are: <u>E.coli</u>, <u>Pseudomonas</u> <u>aeruginosa</u>, <u>Klebsiella</u> and <u>Citrobacter</u>. Even though the sampling and identification procedures were limited, it is indicative of a faecal contamination which should be examined in further detail.

Throughout the duration of the project, the students showed a great deal of interest and they were highly involved. From the way they handled the bacterial samples and used sterile techniques, it was clear that they learned about bacterial species and the various techniques used to isolate and identify them. The students' interest shows the important effect of hands-on experience and involvement of the students in both educational and sociological activities. This supports the following quotation of Forest (1980):

" Tell me, I'll forget, Show me, I may remember; But involve me and I'll understand..."

At the end of the project, the students met with community representatives to present and discuss their findings. Several students explained the testing techniques and equipment used to test the water. Then they explained to the community members that there is a real problem with the quality of the water sources studied, and they made them aware of the danger of ingestion or contact with the water.

Based on their evaluation of the project, the students felt that the lab experience was more effective than the regular biology labs as they presently exist in the 1G6 course outline. They gained initial experience in dealing with the public and obtained practical experience of an applied problem.

The study of the Hamilton and Dundas water sources grew out of concern from both communities about the quality of their environment. The community members posed several questions concerning the testing techniques and the results. The possible side effects of drinking the water were also discussed. Dundas community members were also concerned with possible pollution coming from decomposed industrial waste. Therefore, they were concerned with much more than faecal contamination and went on to express their interest

in continued cooperation with McMaster in studying the water quality and in obtaining help in solving the problem of water pollution. Overall, community members saw how science can serve them, through direct interaction with the research going on at the University. Also they were impressed by the students' involvement and interest in the environment. Even though the social interaction was limited, the objective of learning science in a socially meaningful way seems to have been achieved.

CHAPTER VI

Recommendations:

The general picture emerging from these laboratory exercises is that these exercises have met their objectives. However, some technical and educational aspects need to be further developed.

• Technical Aspects:

1. The water sampling should be more frequent.

2. It would be desirable to run other tests so that we could have a better picture of the type of organisms present in the water sample.

The coliphage test should be used, it is an effective test for examining water pollution. It is known to be inexpensive and easy to perform with the <u>E.Coli</u> host being the most sensitive in all the waters tested.
Since the present study is preliminary, further studies are recommended in the future to obtain more accurate and conclusive results.

• Social Interaction:

Important recommendation made by the community members: 1. Determine the sources of the water contamination. According to them, the Dundas farms upstream may have

been contributing to the water contamination in Dundas making animal waste the focus of concern as opposed to human waste.

2. Analyze water supplies for toxins. The principal concern of the Dundas community members was to analyze their water supplies for any toxic substances that might be drifting down from the old Stately-Brown guarry. There is a proposed housing development on the old guarry, and the ground streams are being closed with land fill. The quarry is right on top of the escarpment, and there is fear that the toxins of the ground fill are leaking down into community streams. The guarry has served as a dump site of mixed industrial waste for years, and it is likely that more industrial waste will be added to close off the ground water. The community is interested in what happens when this industrial waste decomposes, and what types of toxic substances may be in the streams and ground water around the Stately dump. Analyzing the presence of toxins in the water will give the community evidence to argue for closing down the landfill project.

3. Both communities, Hamilton Beach and Dundas, should maintain a continuous contact with the Department of Biology at McMaster university which can assist them in improving the water quality. Therefore, a program of water quality control should be implemented.

4. Community awareness was stressed above all. It is suggested that the community members should regularly examine the quality of their water as such knowledge is a form of self-empowerment, and will perhaps force action.

5. It is suggested that Hamilton beach residents should be provided with materials necessary to conduct water tests. Since these tests are simple and require inexpensive equipment, residents could regularly monitor the quality of water in "minilabs" set up in their own homes. This was also suggested in the study of water quality of the Cree Nation of Split Lake conducted by Dutka (1990).

6. The social interaction should be more frequent, to keep community involved in research that they support through taxes and this would also create social awareness amongst the students regarding the communities they live in.

7. Development and implementation of such educational projects should be assigned for the students as part of their scheduled course work. This will ensure their participation and persistence during the length of the program.

In conclusion, the solution for the future of water quality in both Hamilton Beach and Dundas, will rest on the responsible and active participation of these communities. A strong link between the local communities and the university will contribute to increasing awareness and improving the status of the water quality. This study is hopefully a beginning of a joint effort of the local communities and McMaster University to improve the local environment.

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APPENDICES
APPENDIX A

General Guide For Students And Basic Instructions

Although the microorganisms we are working with are <u>not</u> <u>considered to be pathogenic</u> (non-disease producing), use the same precautions you would use with pathogenic organisms.

- Read the laboratory instructions carefully before you start the experiment.
- Working with bacteria, it is important to keep exposure and contamination of culture media and utensils at a minimum.
- In general, all media, culture tubes, pipettes and other glassware are autoclaved to kill any residual organisms.
- Containers should be opened for as little time as possible when material is being transferred.
- If any culture spills on hands, clothing or benches,
 wipe it up with disinfectant.
- Do not put pencils or other articles in your mouth during laboratory work.
- Before leaving the lab autoclave all tubes and glassware, and wash hands.
- It will be often necessary to come in for short period of time during the week to examine your plates or to inoculate fresh cultures for the next class.

Lab 1

References: Any Introductory Microbiology book.

Objectives:

When you have completed this lab you should be able to:

 Know safety regulations e.g., handling bacterial samples and laboratory equipment.

2. Prepare the culture media.

3. Calibration test: a) determine the number of living cellsin a sample.b) identification of the living cells.

Bacteria are single-celled organisms, much smaller than the yeast, algae, and protozoa. Their identification depends on morphological features that are visible such as shape and arrangement into groups as well as the appearance of colonies and chemical properties. Reproduction in bacteria occurs by binary fission. As the bacterium takes in food from its surrounding medium, and converts it into its own material, it grows up at a point which it splits into two cells each of which continue to grow in the same In order to grow rapidly, the bacteria have to be manner. suspended in broth and incubated at required temperatures. The most common way of determining the growth rate is to estimate the number of organisms in the original culture, and then the" growth" is any increase in numbers (cell/mL) subsequent to the original determination.

Coliform usually refers to a group of bacteria that are normal inhabitants of the intestinal tract, with certain exceptions, these do not cause gastrointestinal-diseases, and using this definition the coliform can be separated into two groups:

1) Esherichia, Shigella and Salmonella.

2) Klebsiella, Enterobacter, Serratia.

<u>E-Coli</u> is an intestinal parasite that can live free in nature, and its presence (In high concentrations) in water supplies is therefore, evidence of recent faecal contamination.

pure cultures are usually isolated by spreading out a sample of cells on the surface of a suitable medium. Growth media have been developed for the isolation or identification of many specific types of bacteria, some media are selective, meaning that they contain ingredients that inhibit the growth of certain other bacteria that are of no interest to the investigator. For example bile salts, and basic dyes are often included for the identification of bacteria of interest.

2. Preparation of culture media:

The culture media must contain a source of carbon (e.g.,carbo-hydrates), nitrogen (amino acids, protease, peptone), oxygen, minerals, water and have an appropriate pH. Sometimes special suplements are also needed.

Luria Broth (LB)

Bacto-Tryptone 10.0 g

Yeast Extract 5.0 g

Sodium Chloride 10.0 g

Water 1.0 L

For plates 15.0 g agar

- Dissolve ingredients and sterilyze by autoclaving.

- Take 25 mL of LB broth and pour it into a small flask.
- With a sterile stick or a loop, touch a colony on an LB plate containing E- coli.
- Dip the stick into the flask containing the LB broth.
- Plug the flask and place it in the shaker at 37°C, to grow over night. This is your concentrated stock culture.

3. <u>Pouring the plates:</u>

- Pour at least 15 to 20 mL of media into each plate by gently lifting the cover of the petri dish just high enough to pour the medium.
- Carefully avoid spilling the medium on the outside of the container or on the inside of the plate lid when pouring.
- Allow the plates to solidify. After the medium solidifies, invert the plates open and place them upside down in the incubator to dry for 20 min.
- 4. Calibration test: The number of living cells can be

determined by plating dilutions of the original culture onto the surface of the agar medium.

Calibration of drops of solution using a pasteur pipette:

- take a 10 mL graduate cylinder
- with a pasteur pipette, add distilled water to the cylinder and count the number of drops added to make 1 mL.
- from the above, calculate how many drops make 0.1 mL.
- 4.1 <u>Serial_dilutions:</u> Work individually.
- set up a dilution series A with 5 tubes labelled A1,
 A2,....A5.
- pour sterile nutrient broth 3 mL of LB into the 5 sterile culture tubes. Need one more tube containing 1 mL of LB to make the final dilution.
- with a Pasteur pipette, transfer 2 drops (0.1 mL) from the stock culture flask into the A1 tube, swirl the content gently to mix thoroughly.
- transfer 2 drops from A1 to A2, swirl the content to mix.
- continue in this manner each time using a fresh pipette.
- finally you have a serie of 5 tubes each of which has 1/30 as many cells as the one before it.

4.2 Inoculation of Agar Plates:

Growing on an agar surface, a bacterium divides. The number of the progeny bacteria increases so much that a visible cluster of bacteria appears. This cluster is called a bacterial colony. One bacterial colony is developed from one cell.

The objective when streaking plates, is to obtain well separated individual colonies of bacteria. When streaking, hold the petri plate in your left hand partially covering the agar to prevent contamination.

- obtain five plates and label them A1 to A5 correspondingly to the culture tubes.
- transfer 2 drops from the tube A1 to the plate A1.
- dip the pasteur pipette into alcohol, flame it to make an
 L shaped instrument and spread the culture on the plate.
 continue in the same manner with the rest of the plates.
- place the plates in the incubator bottom side-up (why?) for 24 to 48 hours.

- after 24 to 48 hours, the plates are ready to be counted.

5. Diagnosis of E.coli using T4 bacteriophage:

The viruses infecting bacteria are called bacteriophage, best studied of these are the bacteriophage infecting the human intestinal bacterium 'Escherichia coli'. A single virus will form a clear plaque, or clearing on a continuous surface layer (lawn) of susceptible bacteria. Each plaque has originated from one virus particle. Procedure: Work individually.

1. Obtain one plane agar plate.

- Take an individual colony and streak several times across the plate.
- 3. Place a small drop of T4 on each streak.
- 4. Incubate the plate over night.
- 5. Examine the presence of plaques.

6. Diagnosis of E.coli using MacConkeys' medium.

A variety of special growth media are used to determine the type of bacteria. The most common media are the colour indicator plates. These agar plates incorporate the colour (dyes) into the medium. The colour of these dyes is sensitive to PH. The media also contains sugar, for example lactose, as a carbon source and a complete mixture of amino acids. Both lac⁺ and lac⁻ are able to form individual colonies on their medium.

- <u>E coli</u> cell utilizes the lactose (lac+) and in so doing produces a local decrease in PH that causes the dyes to stain the colony with an intense purple.
- Salmonella and Shigella cannot utilize the lactose
 (lac-) and instead uses some of the amino acids as carbon
 sources. one of the product of amino acids metabolism, is

ammonia which increases the local PH, decolorizes the dyes, and causes the colonies to be white.

Supplies required:

- 1) MacConkey media
- 2) LB plate containing bacterial colonies.

MacConkey medium: This medium is commercially available.

Bacto-peptone	20.0	g
Bacto-Lactose	10.0	g
Bile salts	1.5	g
Sodium chloride	5.0	g
Agar	15.0	g
Neutral red	0.03	g
Crystal violet	0.001	g
Water	1.0	L

- dissolve 51.5 g of commercial mixture in litre distilled water.
- keep boiling to dissolve completely.
- PH of medium should be approximately 7.1 after autoclaving at 121°C for 15 min.
- plates may stored up for 30 days at 4°C.

Procedure:

- Carefully plate individual colonies on MacConkey's medium, by streaking from an individual colony.
- 2. Incubate the plates at 35 to 37°C for 24 hours.

3. Observe the colour of the colonies.

Questions

- Q1. Why did you need to do serial dilutions?
- Q2. Why would you streak the inoculating pasteur pipette back and forth on the agar surface?
- Q3. Why it is important to flame the pasteur pipette when you spread the culture ?
- Q4. What is a phage? Describe the mechanism of bacteriophage invasion.
- Q5. Draw the phage particle and label the parts.

APPENDIX B

Lab 2 : Membrane Filtration Technique.

Objectives:

When you have completed this lab you should be able to:

- 1. prepare the medium for faecal coliform mFC.
- learn the membrane filtration technique using distilled water.

1. Preparation of Faecal Coliform media:

The medium is commercially available in a dehydrated form. It consists of the following ingredients:

Tryptone 10.0 g Proteose peptone#3 5.0 g Yeast extract 3.0 q Sodium chloride 5.0 g Lactose 12.0 q Bile salt #3 1.5 q Aniline blue 0.1 g Aqar 15.0 g Distilled water 1 L

To rehydrate the medium, dissolve 52 grams of the above mixture in one litter of distilled water and heat to boiling continue heating for 1 minute. determine the pH at 25°C. Adjust if necessary, with 1 N Hcl to final 7.4. Do not autoclave cool to 50°C and pour plates.

2. Membrane Filtration Technique (MF):

A membrane filter is a thin, micropourous, non- fibrous filtration medium for liquids and gases, having a mean pore size of 0.45 micrometers in diameter. Such membrane acts as a sieve to filter microorganisms from any fluid. On mFC medium, faecal coliform, produce typically blue colonies.

Apparatus:

- sterile membrane filter unit (filter base and funnel), glass, plastic or stainless steel;
- filtering apparatus which should contain a vacuum source,
 and a filtering flask and a flask for water trap;
- forceps, 95% alcohol in small beaker, and a bunsen burner. Procedure:
- a) set up a dilution serie A. refer to lab# 1.
- b) number and label all petri dishes
- c) transfer 0.1 mL of culture from A5 into 100 mL of D H2O.
- d) mix thoroughly the sample (shaking 25 times).
- e) with a sterile forceps take the membrane filter and place it on the filter holder. Place the funnel onto the assembly.
- f) pour the water sample into the funnel of filtration unit being careful to minimize splashing on membrane holder walls.
- g) turn on the vacuum.

- h) using flamed forceps remove the membrane from holder and place on mFC media. Avoid entrapment of air between the surface of the medium and the membrane.
- i) incubate the petri dish at 37°C for 24 to 48 hours.
- j) after incubation , examine the membrane and describe the colonies

Bacterial densities are calculated as colonies per unit volumes of samples.

Identification of microorganisms:

 Any microbial growth needs a certain type of culture media that contain various carbohydrates, chemicals, etc. that serve to aid in identification of bacteria.

Supplies required for the diagnosis:

1. MacConkey plates.

2. mFC plates containing isolated colonies.

Procedure:

- 1) Carefully streak the MacConkey plate with a single colony from the mFC plate.
- 2) Incubate at 35 to 37°C for 24 to 48 hrs.
- 3) Examine the isolated colonies.
- 4) Record the results.

APPENDIX C

Lab 3: The Actual Water Testing Experiment.

Objectives:

This laboratory session will:

- 1. Enable you to learn the sampling technique.
- 2. Enable you to quantitate and identify the microorganisms present in communities' water sample.

Contamination of water supplies by human body waste can be dangerous to consumers. Experience has established the significance of coliform group densities as a criteria of the degree of pollution and thus of the sanitary quality of the sample under examination.

1. <u>Sampling Technique:</u>

In collecting samples directly from a river, stream, lake, reservoir, spring, or shallow well, samples can often be taken as follows:

- a) Obtain sterile bottles.
- b) Remove the cap (do not touch the lip of the bottle with fingers)
- c) Hold the bottle near its base in the hand and plunge it, neck downward, below the surface allowing it to fill.
- d) The bottle should then be turned until points slightly upward.

E) Replace the cap.

Size of the sample:

The volume of a sample should be sufficient to carry out all the tests required, preferably not less than 100 mL of water for samples intended for bacteriological examination.

The bacteriological examination of a water sample should be started promptly after collection to avoid unpredictable changes.

<u>Note</u> : The time and temperature of all samples should be recorded and should be considered in the interpretation of data.

1. Membrane Filtration Technique (MF):

Supplies required:

1) MFC plates

2) MacConkey plates (refer to sec. 4.3.2 lab#1)

3) Filtration unit

4) Water samples.

Apparatus: & Procedure (Appendix B)

- work in pairs.

- after incubation, count and describe the colonies.

- prior to initial sample being tested, a negative control using wash water should be processed. After all samples have been processed, another negative control should be run. Why?

Bacterial densities are calculated as colonies per unit

volumes of samples. For faecal coliform, if the number of colonies per 100 mL exceeds 60 colonies, that means the water is not sanitary.

1. First control test using a diluted and a concentrated culture.

<u>Purpose</u>: To see the effectiveness of the membrane filter technique and wether the filter is not being harmful to the organisms.

Procedure: Work individually

- with a pasteur pipette add 2 drops of E.coli culture, containing 50 cells to 100 mL of distilled water.
- swirl gently and pour the water into the membrane filtration unit.
- with a sterile forceps, remove the membrane and place it onto the surface of the mFC medium.
- take 0.1 mL of culture containing 50 cells and add it directly to the membrane filter already placed on the mFC medium.
- incubate the two plates for 24 to 48 hours. This test
 should give you a comparable number of bacterial colonies
 in both dilutions.
- after 24 hours your plates are ready to be counted.

2. <u>Second control test using community water samples</u>: <u>Purpose</u>: To find out wether growth of E.coli is prevented by other existing organisms, or due to the toxicity of chemicals in the water sample.

Procedure:

- collect water samples as directed.
- add 2 drops of E.coli from dilution factor (50 cells) to one water sample.
- filter the water sample and place the membrane filter onto the mFC medium.
- take 2 drops from dilution factor (50 cells) and add it to the filtered water.
- this will in turn, be filtered and the filter be placed on the mFC medium.
- incubate the plates at 44.5°C for 24 hours.
- after 24 hours, examine your plates and record you results.

Questions

- Q1. What conclusions can you draw relative to water source and bacterial counts?
- Q2. Can you explain the colour change produced in the mFC medium?
- Q3. Does the filtration procedure have an effect on E.coli?

APPENDIX D

Lab 4:

Identification of microorganisms:

Objectives: When you have accomplished this lab you should be able to:

- Master the use of diagnostic procedures
 (Mackonkeys'medium and the T4 phage) of microorganisms.
- Learn the Gram stain procedure, and partially identify the bacterial organisms present in water samples of Hamilton Beach and Dundas communities.

1. <u>Partial identification of bacterial colonies using</u> <u>MacConkeys'medium:</u>

Supplies required for the diagnosis:

- 1. MacConkey plates.
- MFC plates containing isolated colonies from the previous water tests.

Procedure: See Lab 1.

- 2. Identification of bacterial colonies using T4 phage: Supplies required:
- 1. LB agar.
- 2. A diluted sample of T4 phage.
- 3. mFc plates containing isolated colonies.

Procedure: See Lab 1.

3. Identification of bacterial colonies using Gram Stain:

To be properly observed under the microscope, bacteria must be stained. Three basic shapes of bacteria are present in nature: round or coccoid, rods, and curved or spiral.

In the process of Gram staining, a fixed bacterial preparation is stained with crystal violet, a basic dye which stains the cell purple. The slide is then subjected to iodine which increases the affinity of the cell for the stain. the bacterial preparation is then decolorized with 95% ethanol, which tends to destain the cell from the initial dye. Gram positive are not decolorized by ethanol and so remain purple. Gram negative bacteria are decolorized by ethanol. Finally the bacterial preparation is subjected to a counterstain; safranine which stains the gram negative cells pink. The gram positive remain purple. Supplies required:

1. Different colonies from the mFC plate.

2. Crystal violet, iodine, 95% ethanol, and safranine.

3. Two slides

Procedure for smears: work individually.

- Take the slide and draw small circles on the undersurface with a pencil. So a number of smears can be prepared on a single slide.
- 2. With a sterile pasteur pipette touch one of the bacterial colonies and rub the bacterial material on one of the circles you drew.

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- 3. In the same manner, transfer bacteria from another colony on the mFC plate to the other circle.
- 4. Make sure to sterilize your pasteur pipette each time you transfer bacterial colonies. (Be sure the sterilization of your pasteur pipette becomes a habit).
- 5. Allow the smear to air dry.
- 6. Pass the slide quickly across the top of the flame of your bunsen burner. This will fix the bacterial organisms to the slide so that the staining will not wash the organisms off the slide.

Staining procedure:

- Cover smear with crystal violet for one minute and then wash gently with water.
- 2. Cover with Gram's iodine for one minute and wash off.
- 3. Flood slide with 95% ethanol for 10 to 20 seconds, then drain the slide.
- Rinse with water and then cover with safranine for 30 seconds. Drain, wash with water and dry slide by pressing between paper towels.
- 5. Examine the slide under the oil immersion lens.

Questions

- Q1. What can you deduce from your observation concerning the shape of different bacteria.
- Q2. Which of the stains allows you to distinguish different

organisms.

Q3. Based on your findings, is the water safe for use?

Appendix D

part 2: Biochemical Tests:

1. Indole test: Tryptone broth.

In this test a type of tryptone which has a high content of tryptophan is used to test the ability of organisms to produce indole. Tryptophan is the only amino acid containing the indole ring. After the bacterium has grown for 1-2 days in tryptone broth, add 8 drops of Kovac's reagent. A dark reddish colour developing on the surface of Amyl alcohol is indication of the presence of indole. The reddish colour results from condensation of indole with the aldehyde.

Kovac's reagent: For <u>indole Test</u>: Paradimethyl-amino- benzaldehyde 5 g Isoamyl Alcohol 75 mL Concentrated hydrochloric acid 24 mL Dissolve the P- dimethylamino -benzaldehyde in the isoamyl alcohol and then add the concentrate hydrochloric acid. The final colour should be light yellow.

2. Simmons citrate medium:

Magnesium sulfate	0.2	g
Monoamonium phosphate	1	g
Dipotassium phosphate	1	g
Sodium chloride	5	g
Bromthymol blue (0.08	a

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Mix ingredients in cold distilled water and heat to boiling to dissolve the medium completely. distribute in tubes and autoclave at 121°C for 15 minutes.

The source of carbon is citrate. The initial PH is 6.8 and the indicator dye will be green but if the bacterium can metabolize the citrate, this will be removed from the medium which will become more alkaline and deep blue in colour.

3. <u>Inositol medium:</u>

Proteose peptone	10	g
Beef extract	1	g
Sodium chloride	1	g
Bacto brom cresol purple	0.02	g
Inositol	5	g
Distilled water	1	L

Mix ingredients in water to dissolve completely. dispense into tubes and autoclave for 15 minutes at 121°C. Final PH should be 6.8 at 25°C. Production of gas is evidence for the presence of certain type of faecal coliform organisms.

4. Christensen's modified acetamide agar, for <u>Pseudomonas</u> <u>aeruginosa</u> confirmation.

Sodium chloride5gAcetamide10gDipotassium phosphate1.4gMagnesium sulfate0.75g

Phenol red	0.012	g
Agar	15	g
Distilled water	1	\mathbf{L}

Mix ingredients to dissolve then bring to boil. Autoclave for 15 minutes at 121°C. Final PH is 6.8 pour into sterile plates and leave them for 20 minutes to solidify. After inoculation of plates a colour orange red, if it occurs, should be evident in 1-2 days.

APPENDIX E

QUESTIONNAIRE

As part of the research project for MSc(Teaching) we have designed this questionnaire. It will be used to evaluate the effectiveness of the project.

Please circle the number which most closely corresponds with your opinion.

- The Laboratory skills I learned in these labs have given me a better appreciation of what scientific experiments are like.
 - 1. strongly agree 2. agree 3. neutral 4. disagree
 - 5. strongly disagree
- The objectives and introduction at the beginning of each lab were helpful to me.
 - 1. strongly agree 2. agree 3. neutral 4. disagree
 - 5. strongly disagree
- 3. My impressions of the usefulness of these labs have improved since joining this research project.
 - strongly agree 2. agree 3. neutral 4. disagree
 strongly disagree
- 4. For those who interacted with the local community: As a student my interaction with the local community was useful.
 - strongly agree 2. agree 3. neutral 4. disagree
 strongly disagree

- 5. I feel that the project has given me an understanding of the community needs for water quality
 1. very good 2. good 3. adequate 4. inadequate
 5.not at all
- I feel that university/community interaction is a valuable part of science education
 - very valuable 2. valuable 3. adequate 4. not valuable 5.not at all
- 7. Please give your comments about the instructor which could help improve her effectiveness.
- 8. What worked well in the course?

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9. What did not work so well?

10. Do you have any suggestions to improve the course?

APPENDIX F

Letters to the Testing Agencies and the Local Communities.

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McMASTER UNIVERSITY Department of Biology 1280 Main Street West Hamilton, Ontario, Canada L8S 4K1 Telephone (416) 525-9140 Ext. 4400 FAX: (416)521-2955 E-Mail: WHITEBN@McMaster.CA

September 24, 1991

To Whom It May Concern:

This is to confirm that Ms. Zaimi Ouahida is a graduate student in the Department of Biology, McMaster University. As a part of her thesis project she would like to observe water testing procedures. I would be grateful if you would allow her to see these procedures in your organization.

She might also wish to have a small group of students observe the process. I would appreciate any help you could extend to her.

Yours sincerely,

Ladey N white

Bradley N. White Professor and Chair

BNW/klg

SAINT MARY'S HIGH SCHOOL 1400 MAIN STREET WEST HAMILTON, ONTARIO L8S 1C7

MR. C. J. FOX, PRINCIPAL MR. J. D'ANGELA, VICE-PRINCIPAL TELEPHONE (416) 528-021 (416) 528-021

October 31, 1991

Dr. J. Richardson, Chairman, M.Sc.(T) Program, McMaster University, 1280 Main St. W. Hamilton, Ontario

Dear Dr. Richardson:

re: Miss Zaimi Ovahida Graduate Student

The following students of this school's OAC Biology

A State of the sta

have volunteered to help Miss Zaimi with her project:

- 1. Beth Hall
- 2. Tracey McIntosh
- 3. Giuliana Casimirri
- 4. Sujoy Majumdar
- 5. Sasha Spycher

We will be able to provide support for these students, thus enabling Miss Zaimi to collect the necessary data for her research proposal.

Yours sincerely,

Mr. . J. Principal

CJF/ms

October 6, 1991

To : Mr. BoB JAGGARD From: Miss. OUAHIDA ZAIMI Graduate student at Mac Master University.

We are presently conducting a project, which is in support of a Master's thesis in Biology/Education.

The project involves water quality testing. One aim of the study is to develop laboratory units that have some social relevance; in which students can interact with the community and discuss the existing community problems.

Would you be willing to cooperate with us in this venture? The end results will hopefully increase the awareness among students about their community. And on the other hand increase awareness in the community about how science can serve them.

Also, we will be very grateful if you allow me to attend one of your community meetings.

Sincerely, fay



HAMILTON BEACH PRESERVATION COMMITTEE INC 663 Beach Blvd L8H 6X8 President : Bob Jaggard-544-6518 Treasurer/Social Convenor: Ernestine Massignani-547-2038 TEMPORARY SEC. ALICE SELBIE.

RESIDENT'S MESSAGE - cont'd - 2 - October 1991

Please note that at the recent meeting at City Hall, Mr. David Godley from the lanning Dept , asked that if Beach residents have any suggestions and/or comments concern ng Beach development, please send your written suggestions, by OCTOBER 31st, 1991, to:-

> Mr Alex Georgieff, Director of Local Planning, Planning and Development Department, City Hall, 71 Main St. West, Hamilton, Ontario, L8N 3T4

I recently received a phone call from Dr. George Sorger at the Biology Department of Master University, asking if our Committee would co-operate with a young student from Igiers, Africa who is taking a course on Micro-Biology and Community Water Testing, and Iso to observe the socializing of our Community. This student will then be taking her earning experiences back to Algiers when she returns. IF ANY OF THE YOUTH WISH TO ASSIST HIS YOUNG GIRL IN HER STUDIES, PLEASE CONTACT MYSELF OR ERNESTINE MASSIGNANI.

Finally, there will be an ALL CANDIDATES MEETING on TUESDAY, NOVEMBER 5th, 1991, at the FIRE HALL, at 7:00 p.m.. ALL residents are encouraged to attend this meeting to voice their questions and concerns to these Candidates. This meeting will take the place of our agular monthly meeting.

EE YOU ALL AT OUR HAMILTON BEACH PRESERVATION COMMITTEE MEETINGS !

Bob Jaggard

Bob Jaggard, President, HAMILTON BEACH PRESERVATION COMMITTEE INC.



.S.

CHRISTMAS CRAFT SALE



Sunday, DEC. 1st, 1991 - 11:00-4:30 Bell Cairn Staff Development Centre

March 3,1992

To: Mrs E. Shannon, 19 parkside Avenue Dundas Hamilton,Ont.

Dear Friends

Enclosed is a copy of our project outline plus the results of the application of phase 1 of our project to McMaster course Biology 1G6 students and a Dundas community.

As you can see phase 1 did work in this case. Phase 2 and 3 need to be developed and, in the case of phase 2 research, we have applied to the Ministry of Education for funds to do this, as shown in the enclosed outline. If you agree that this is worthwhile please support us by making this known to your local MPP.

We are hopping that this approach to higher education and community outreach will empower communities and increase the social awareness of our students. We appreciate your input, we would like to stay in touch and to hear from you.

Sincerely,