REMOVAL OF BACTERIA

BY REVERSE OSMOSIS METHOD

by

Emmanuel Achonna Anyahuru

A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfilment of the requirements for the degree of Master of Engineering.

Department of Civil Engineering
and Applied Mechanics
McGill University
Montreal, August 1972.

A B S T R A C T

The Removal of Bacteria by Reverse Osmosis Method.

by

E. A. Anyahuru.

Degree: Master of Engineering.

Department of Civil Engineering and Applied Mechanics.

This investigation is a continuation of previous studies which have been made at the Department of Civil Engineering and Applied Mechanics on the use of reverse osmosis for water treatment. In 1971 this Department developed a membrane which has a high flux characteristic. This investigation shows that further modification will be required for this membrane in order to produce water which meets the bacteriological Standards of drinking water.

A C K N O W L E D G E M E N T S

The author wishes to express his appreciation and gratitude to the following for the generous and painstaking assistance given to him:

Dr. S. E. McKyes of the Department of Civil Engineering and Applied Mechanics, McGill University, and his staff, for their assistance in producing the electron microscopic views of the membranes.

Mr. H. Brown, graduate student, for help in every phase of this research.

Mr. B. Cockayne and his technical staff in the Department, for help in modifying the equipments whenever it was necessary to do so.

Mrs. Diana Bouchard, who typed this thesis.

Professor Andrejs Pakalnins of the Department of Civil Engineering and Applied Mechanics, McGill University, for directions, encouragement, and helpful suggestions in all phases of this research.

This investigation was supported by a research grant No. 604-7-590, Water Treatment by Reverse Osmosis Method, of the Federal Department of National Health and Welfare, Ottawa. The author was sponsored by the Canadian Government under the Commonwealth Fellowship scheme. The East Central State of Nigerian Ministry of Works, Housing and Transport, where the author is an Executive Engineer, provided assistance by granting the author study leave.

The author is indebted to both Governments.

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I N T R O D U C T I O N

Most of the reports on reverse osmosis as a process for water treatment have been concerned with the theoretical considerations of the mechanism which result in a complete or partial rejection of ions in electrolytic solutions such as saline water, heavy metal molecules and removal of turbidity. The literature has been silent on the bacteriological aspects of the removal mechanism. Investigators have reported product waters with insignificant number of the coliform indicator organism and have often described the observations as being due to contamination of the collecting system.

With theoretical pore sizes encountered in cellulose acetate membranes being of the order of 1500 Å, it appears physically impossible for bacteria to pass it through an osmotic mechanism acting on the pores.

In 1971, the Department of Civil Engineering and Applied Mechanics, McGill University, developed a semi-permeable cellulose acetate membrane for water treatment. This membrane produced flux comparable to and even better than those of slow sand filters from fresh, heavily polluted water. This was a remarkable improvement on the membranes developed elsewhere mainly for desalination and, recently, for polishing the effluents of waste water treatment plants. It becomes necessary to determine whether this high-flux membrane would produce water which would meet the bacteriological Standards for drinking water.

Considering that industrial and domestic waste

discharges into surface waters steadily increase while the capacity of the surface waters for self-purification is limited, any water treatment process should be able to remove contaminants as their concentrations increase. This investigation also had to determine if this high-flux membrane could produce potable water from the contaminated surface waters as their bacteriological contamination increased. This defined the basic aims of this investigation.

(1) To find if the high-flux membrane could produce water which meets the bacteriological Standards of Drinking Water.

(2) To find what effects the high bacterial concentrations might have on the product water of the high-flux membrane.

CHAPTER I

THEORY

A. Reverse Osmosis in Water Treatment

If two liquids of different ionic concentrations are separated by a semi-permeable membrane, it is observed that the less concentrated liquid flows across the semi-permeable membrane into the liquid with higher concentration. If the two solutions are of identical ionic content, the flow continues either until the two concentrations balance or until a hydrostatic pressure, equal to the original driving force, p_o, is established. This original driving force is called the osmotic pressure. On the other hand, if originally an external pressure equal to p_o is imposed onto the solution with higher concentration, there will be a state of equilibrium and no flow will be observed in any direction. If a pressure P, greater than p_o, is applied, the direction of flow across the semi-permeable membrane will be reversed. The flow would proceed from the liquid with higher concentration and an external pressure, $P > p_o$, into the less concentrated liquid. This phenomenon is referred to as reverse osmosis. Figures 1 and 2 illustrate this principle. It was first observed by Abbé Nollet (1) in 1748 with a water-alcohol solution. In the nineteenth century Pfeffer (2) made the first quantitative measurement of osmotic pressure using copper ferricyanide and water, with porcelain as the membrane. Further developments on the work of Pfeffer were published by van't Hoff (3) in 1886.

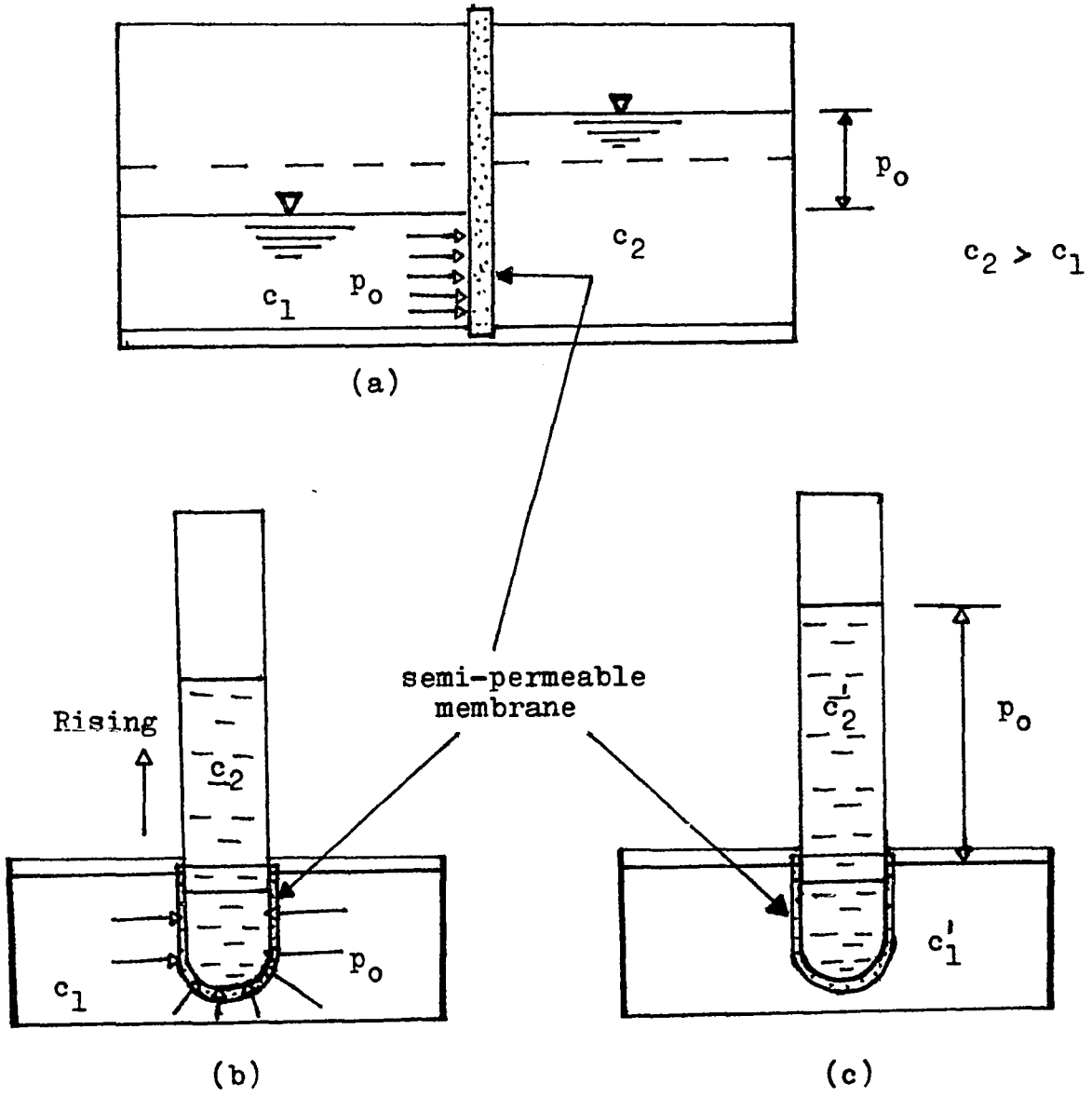

Figure 1. Principle of osmosis.

(a) Two liquids of concentrations c_1 and c_2 are separated by a vertical wall of a semi-permeable membrane. $c_2 > c_1$, and flow is from c_1 to c_2. The level of liquid c_1 falls, level of c_2 rises.

(b) Same condition as in (a); $c_2 > c_1$.

(c) Condition of osmotic equilibrium. p_o = osmotic pressure in psi.

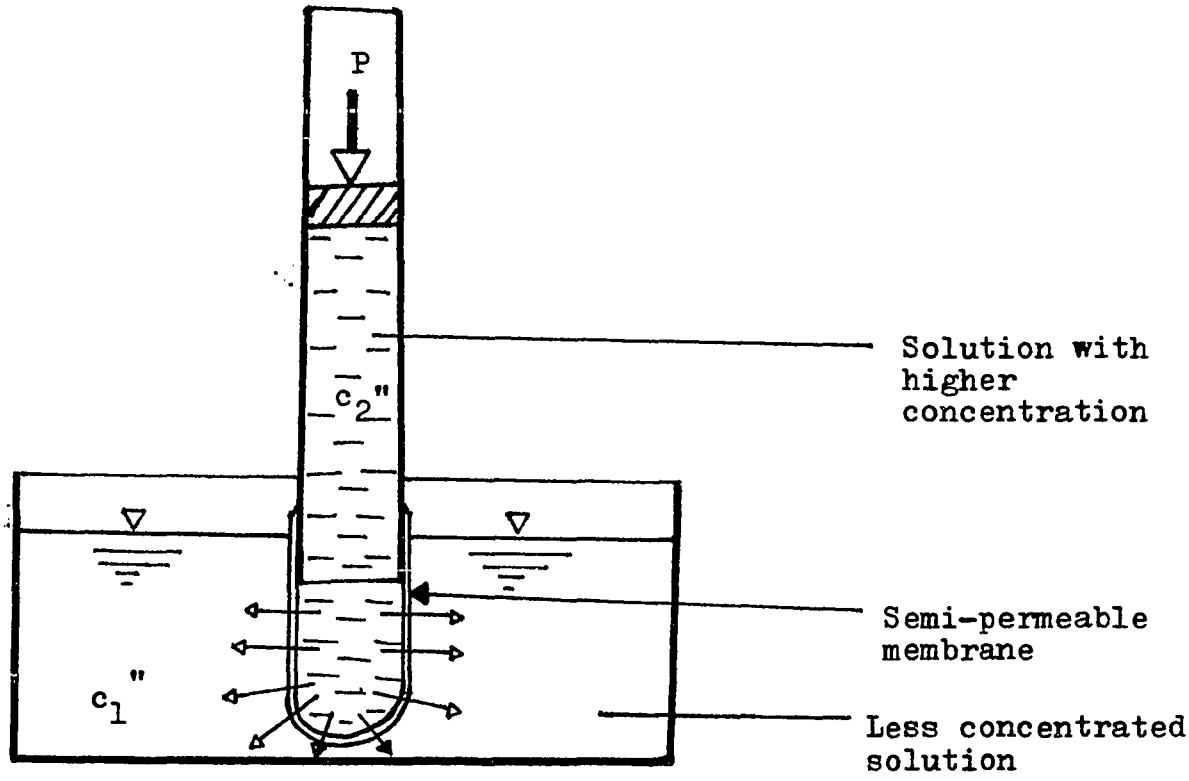

Figure 2. Principle of reverse osmosis.

P = reverse osmosis pressure in psi

$P > p_o$

The application of the reverse osmosis principle has been developed very extensively on the Pacific Coast of the United States for converting saline and brackish water into potable water. When the need for an improvement of effluent quality of waste water treatment units became high, reverse osmosis was developed as an advanced method for waste water treatment. Organizations which have made substantial contributions to the development of reverse osmosis as a treatment technique for water and waste water include, among others, Aerojet General Corporation at Azusa, California (4); General Atomic Division of General Dynamic Corporation, and the County Sanitation Districts of Los Angeles County at the Pomona Water Reclamation Plant, California (5); General Atomic in the Bergen County Sewer Authority Plant at Little Ferry, New Jersey (6); Department of Applied Chemistry, National Research Council, Ottawa (7); and Washington State University, Pullman, Washington (8).

The parameters which determine the operation of a reverse osmosis process in water treatment include

the type of membrane used in the process,

the nature of the influent water, and

the pressure of the system, P.

Various research organizations have developed membranes adapted to a specific need. Original membranes cast by Loeb and Sourirajan (9) contained the following materials, given as percentages by weight.

Cellulose Acetate	25.15%
Acetone	67.50%

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(cont.)	Water	5.37%
	Magnesium Perchlorate	1.65%
	Hydrochloric Acid	0.33%.

This membrane was later improved by Manjikian et al. (10) with a cast composed of:

Cellulose Acetate	25% by weight
Formamide	30% " "
Acetone	45% " " .

Today, many companies are evaluating several polymeric membranes for use in industrial processes such as pulp and paper, pharmaceutical, and food processing. The membrane used in this investigation corresponds to Membrane C-5 developed by Trivedi (11). Details of the casting procedures will be reviewed in a later section.

Raw water characteristics are a major factor in the use of the reverse osmosis process. High concentration of suspended solids and turbidity may clog the cells very rapidly, thereby reducing the flux. This phenomenon, however, is not uncommon on conventional water treatment plants.

Operational pressures in a reverse osmosis process are high, ranging from 500 psi to 1,400 psi, and this requires special equipment discussed later.

Numerous theories (12) have been put forward to explain the mechanism leading to the removal of both ionic and non-ionic components of the solvent through a membrane which can be classified as semi-permeable. Surface tension, sieve mechanism, distillation, ion exchange, and hydrogen ion bonding

are a few such mechanisms which have been postulated. Many investigators favour the hydrogen bonding mechanism. There are long polymer chains in a cellulose-based membrane. The polymers, of 1500 Å, are randomly stalked in the system like a pile of straw. Under an organized reorientation, crystalline regions are developed with a minimum of void space between the polymer molecules which are believed to be held together by van der Waal forces and hydrogen bonds within the crystalline lattice. In this configuration, the rate of flow is very small, but there is a high percentage of removal of particles which are trapped between the limited void spaces. The reorientation of the polymer molecules is controlled by the nature of the mixture, the method of mixing, and heat treatment. When the random array of these polymer chains is not tightly bound together, it forms an amorphous system. An amorphous system has large voids which provide a high flux with poor removal of particles. Under this configuration, the individual polymer chains are affected by Brownian motion with little or no bonding between them. Breton (13) postulated that it is through hydrogen bonding of the water molecules that the various polymer chains in the amorphous system are held together, the cross linkage depending on the size, number, and orientation of the voids.

A cast membrane has an amount of amorphousness and a degree of crystallinity. Two types of movement take place in the cellulose-water interface - ions and molecules may go through the voids, or the ions and molecules which have formed

hydrogen bondings are transported by migration from one hydrogen bonding site to another until they cross the membrane.

B. Bacteriology of Water

The three categories of bacteria which are found in natural sources of water are

(a) Natural Water Bacteria,

(b) Soil Bacteria,

(c) Animal Excremental Bacteria.

Natural water bacteria require only traces of nutritional materials for their normal life cycle. They can be found in the air where they are washed down by precipitation, and some have been isolated from distilled water with traces of ammonia. In 1906, Kohn (14) found that certain species of these bacteria could exist in water in the presence of 2×10^{-12} mg per litre of dextrose and 6×10^{-12} mg per litre each of ammonium sulphate and ammonium phosphate. Natural water sources - springs, wells, open surface waters - have traces of organic and inorganic compounds well above these limits. Water is the natural environment of many chromatic and fluorescent bacteria, some vibrio and spirochaetes, and some low-temperature coliform organisms (43). They attach themselves to solid surfaces of reservoirs, bottom sludge, plants, and any solid in contact with water. Growth of the so-called metatropic bacteria (14) does occur in lower layers of sand filters and in underdrain systems. Sudden increase

in filtration rates results in total bacterial surge at the effluent, an increase which is not due to a break-through by the raw water, but due to the bacterial growth at the lower depths of the filter system. Slimes and bacteria are found in tightly sealed pressure pipe lines. These bacteria, however, which are indigenous to water, are of no sanitary significance in water supply.

The catchment basins of surface water systems contain decaying vegetation and other sources of organic and inorganic compounds which represent nutrients for bacterial growth. These are washed off into the open waters after precipitation, thereby increasing their bacteriological density and the food potential for further bacterial growth. Most of the soil bacteria have close morphological resemblance to some of the bacteria that grow in the intestinal tract of warm-blooded animals. Examples of these are the common coliform and cocci groups of bacteria. Some of the soil bacteria can grow in very dilute nutrient solutions as do the natural water bacteria, but many require a high concentration of nutrients. These die off very rapidly except in heavily polluted waters.

Bacteria in water arising from sewage and animal excrement include the coliforms, notably Escherichia coli, streptococci, anaerobic spore-forming bacilli, and some enteric pathogens which will be discussed later. The types of bacteria due to sewage alone depend on the composition of the sewage and its stage in the process of decomposition. In all the stages their densities are quite high, ranging in millions per cubic centimeter.

Pathogenic Contamination of Water

The most important water-borne diseases of bacterial origin are:

Typhoid and paratyphoid fever,

Cholera,

Amoebic and bacillary dysentery, and

Gastro-enteritis.

The pathogenic or disease-causing bacteria are found in the body discharges of a person who has suffered from the disease or who is merely incubating the bacteria without necessarily showing signs of the disease. In 1854, Snow (43) showed how cholera was spread by drinking water, and about three years later Budd (43) established how typhoid fever was also caused by drinking contaminated water. The enteric pathogens get into surface waters by direct excretion by a carrier, through runoff discharges along contaminated catchment basins, or through poorly treated sewage systems. They reach shallow groundwaters by seepage through the soil. In water, these pathogens may thrive for a while under favourable environments, but they are quite sensitive to temperature and other environmental changes. Clark et al. (50),investigating the die-away rate of Salmonella typhosa and Entamoeba histolytica, which are the organisms responsible for typhoid fever and amoebic dysentery, respectively, obtained the following relations.

For Salmonella typhosa: $y = y_o e^{-1.1\ t}$

For Entamoeba histolytica: $y = y_o e^{-0.17\ t}$

For Escherichia coli: $y = y_o e^{-0.044\ t}$,

where y = number of bacteria remaining in the water at time t days,

y_o = number of bacteria in the water at time $t = 0$.

The coefficients were obtained assuming that Chick's law applies. The above relations show a rapid die-away rate for Salmonella typhosa compared with Escherichia coli. Fair et al. (20) found this die-away rate to be more rapid in heavily polluted streams than in clean streams, in the summer than in the winter, and in shallow streams flowing at high velocities than in deep, sluggish waters.

Another factor which accounts for the safeguard against water-borne enteric pathogens is the dilution factor of the receiving water. High dilution factor reduces the probability of contact with pathogens in the water.

In London, England, cholera continued to plague the city until 1893. In the United States of America there were over 30 cases of typhoid fever from 1946 to 1960, according to a report by Weibel et al. (15). Many countries continue to have frequent cases of water-borne diseases due to insufficient sanitary safeguards.

The pathogenic bacteria cannot be readily isolated from domestic waters, and there is no single routine procedure for their identification. It is believed that Salmonellae are the most abundant of the common enteric pathogens and there is no simple laboratory method to isolate them yet. A laboratory method for identifying pathogens directly and routinely requires further research in order to develop a

technique which will be cheap to operate, easy to handle, and versatile in application.

Every warm-blooded animal, including man, discharges large numbers of the coliform organisms. A general statistical figure estimates a per capita rate of discharge of Escherichia coli of about 300 billion daily. The streptococci form another group of enteric bacteria discharged in large numbers. Therefore, instead of trying to identify the pathogens, every attempt is now made to ensure that any potential sources of fecal contamination are eliminated in the raw water supplies. This is done by ensuring that drinking water does not contain a significant number of a group of bacteria normally present in the intestinal tract of man. By this assurance, it can be presumed that the likelihood of intestinal pathogens being present in the water is very small.

Hanes et al. (16) have shown that the coliform group of organisms does multiply in water. Their presence, therefore, does not necessarily indicate fecal contamination, and only their complete absence is a true index of the bacteriological safety of drinking water. The streptococci are not known to multiply in water. Their death rates are similar to or greater than those of the coliforms under identical laboratory conditions (16). In the investigations of the bacteriological contamination of the Richelieu River, Neelakantan (17) showed that the coliform organisms outnumbered the enterococcus organisms two- to ten-fold. Consequently, the presence of fecal streptococci may be a better indicator

of recent fecal contamination of surface waters and shallow wells than the coliform group of bacteria.

While the coliforms and streptocccci serve as an index of a possible presence of enteric pathogenic bacteria, there is another class of pathogenic entities which are known to be water-borne. These are the viruses.

Stanier et al. (18) have described a virus as an "infectious, filterable particle (usually less than 0.3μ in diameter) that can reproduce only within a specific host cell." Of the three types of viruses - animal viruses, plant viruses, and bacterial viruses (bacteriophages) - only the animal viruses that are water-borne and enteric in habitat are of importance in this discussion. Echoviruses, polio viruses, coxsackie viruses, adenoviruses, reoviruses, and the viruses of infectious hepatitis are a few water-borne viruses of enteric nature. They are discharged by "carriers" only, and it is believed that their number in wastewaters is several orders of magnitude lower than that of the coliform organisms (19). Even in the feces of infected persons, their order of discharge is much lower than those of bacteria discharged by infected persons (20).

In Edmonton in 1953 (51) there was an outbreak of poliomyelitis believed to have been due to breakdown of a sewage treatment plant located upstream from Edmonton on the Saskatchewan River. In Delhi, India, in 1955 (52), an outbreak of infectious hepatitis was believed to have followed

a flood which reversed the direction of the river Jumma, flooding the two raw water intakes at Chandrawal with raw sewage.

Viruses are small in size, ranging from 0.10 μ to 0.30 μ, and can pass through conventional slow and rapid sand filter units. Most of them, especially the viruses of infectious hepatitis, require longer contact time with, and heavier dosages of, conventional disinfectants. Their sizes can be determined only by the aid of an electron microscope or by analytical ultracentrifuge. Cliver (53) claimed that membrane filters of 0.45 μ pore sizes have been successfully used in isolating viruses by adsorption. It is not yet known what effect the membranes in a reverse osmosis process will have on their removal from water, and further research in this field is required.

The Drinking Water Standards do not list any standard examination methods for enteric viruses.

C. Bacteriological Standards of Water

Surface waters have been the main sources of water supply to communities. Most ancient, medieval, and even modern cities have been founded on river banks where, apart from other factors, the inhabitants would find water to satisfy their thirst. As the communities developed, the need to drain the cities grew. Gutters and channels were built to convey the runoff into nearby rivers, lakes, and other bodies of water. Standards of hygiene improved and domestic sewers were

introduced and connected into the existing channels draining into the nearby body of water. This provided a direct, rapid, and concentrated access of human excreta to the rivers which had previously supplied pure and adequate water to the people.

The Industrial Revolution of the nineteenth century further intensified this problem by bringing more people together into the industrial towns and cities, and later added another type of waste as a result of the industrial processes. The natural capacity of the rivers for self-purification became overtaxed by the organic and inorganic deposits from these various sources.

By the middle of the nineteenth century, a significant breakthrough in environmental sanitation was made by establishing the role of contaminated water in the transmission of cholera and typhoid fever. In England, water supply companies had to move their intakes upstream to avoid the wastewater discharges, treat the water, and generally ensure that the distribution systems were protected against contamination. In 1847, a royal commission was appointed to study and report to Parliament on the sanitary conditions of London (54). As the result of this report, the Metropolitan London Commission of Sewers was established in 1848. The continued concentration of population in England led to mandatory treatment of sewage before it was discharged into fresh water. In the United States, progress was slow until after the Civil War (55).

The pace of industrialization grows and population increases, but the quantity of natural water remains virtually

constant. Government agencies are taking over the duty of protecting water resources. Conventional water treatment processes must operate within definite economic limitations, so that the degree of contamination on the sources of supply must be controlled. Cox (21) has pointed out that the degree of removal of bacteria by any water treatment process is approximately constant for any specific plant and raw water under an efficient control. Therefore, any increase in the bacterial content of raw water resulting from pollution will reduce the degree of removal. Fixing the maximum number of bacteria remaining in the treated water will impose a restriction on the degree of contamination which may be tolerated in the raw water.

Water suitable for public supply must not contain any pathogens which may cause disease. As mentioned earlier, the coliforms and some of the streptococci are the yardstick for measuring the probability of the presence of the pathogens. Therefore, there must be a definite limit to the number of these organisms which may be present before water is considered safe to drink. It was demonstrated earlier that the streptococci are fewer in number than the coliforms and they appear to die off much faster than the coliforms. Coliforms are therefore better factors of safety in a drinking water standard.

In many countries the bacteriological characteristics of a source of water supply determine the minimum level of treatment required by the water before it can be considered safe for drinking. The Canadian Drinking Water Standards (19)

have three categories concerning raw water and specify a minimum treatment for each category. This standard distinguishes between the coliforms of fecal origin and the total coliforms as recovered by the Endo medium. Details of the Canadian Raw Water Standards are shown in Table 1. Table 2 is the Canadian Drinking Water Standards, and no distinction is made between the fecal and total coliform organisms. Any water that contains a considerable number of the coliform organisms of any origin does not meet the Drinking Water Standards. An international standard for raw water (56) which is similar to the Standards adopted in the United States, Britain, and Europe, has four categories, as given in Table 3. The International Standards for drinking water, approved by the World Health Organization, are similar to the Canadian Standards given in Table 2.

To ensure effective control of the numerical limits of the indicator organisms given in the Tables concerning potable water, minimum sampling frequency standards have also been set. This minimum frequency for sampling drinking water depends on the population served by a water supply system. Figure 3 , adopted from the United States Public Health Service Drinking Water Standards of 1962, is now used in many countries.

Table 1

Raw Water Fecal Coliform and Coliform Standards - Canadian (19)

Fecal Coliform		
Objective	Acceptable Limit	Maximum Permissible Limit
(a) At least 95% of the samples in any consecutive 30-day period should have a fecal coliform density of less than 10 per 100 ml. (b) Chlorination only is required.	At least 90% of the samples in any consecutive 30-day period should have a fecal coliform density of less than 100 per 100 ml. Partial treatment must include chlorination.	At least 90% of the samples in any consecutive 30-day period should have a fecal coliform density of less than 1000 per 100 ml. Complete water treatment comprising coagulation, sedimentation, filtration, and chlorination.
Total Coliform		
(c) As in (a), but coliform density to be less than 100 per 100 ml. (d) As in (b).	As in (a), but coliform density to be less than 1,000 per 100 ml. As in (b).	As in (a), but coliform density to be less than 5,000 per 100 ml. As in (b).

Table 2

Canadian Drinking Water Standards (19)

MPN Method		
Objective	Acceptable Limit	Maximum Permissible Limit
(a) No coliforms	At least 95% of the samples in any consecutive 30-day period should be "negative" for total coliform organisms	At least 90% of the samples in any consecutive 30-day period should be "negative" for total coliform organisms
(b) No coliforms	None of the samples "positive" for total coliform organisms should have an MPN index greater than 4 per 100 ml.	None of the samples "positive" for total coliform organisms should have an MPN index greater than 10 per 100 ml.
MF Method		
(c) No coliforms	Same as (a)	Same as (a)
(d) No coliforms	None of the samples "positive" for total coliform organisms should have an MF count greater than 4 per 200 ml or 10 per 500 ml portions.	None of the samples "positive" for total coliform organisms should have an MF count greater than 6 per 200 ml or 15 per 500 ml portions.

Table 3

Bacteriological Quality of Raw Waters - International Standards.

Classification		MPN/100 ml Coliform bacteria*
I	Bacterial quality applicable to disinfection treatment only	0-50
II	Bacterial quality requiring conventional methods of treatment (coagulation, filtration, and disinfection)	50-5000
III	Heavy pollution requiring extensive types of treatment	5000-50,000
IV	Very heavy pollution, unacceptable unless special treatment designed for such water is used; sources to be used only when unavoidable.	> 50,000

* When more than 40% of the number of coliform bacteria represented by the MPN index are found to be of the fecal coliform group, the water source should be considered to fall into the next higher category with respect to the treatment required.

Figure 3: Minimum Sampling Frequency for Bacteriological Analysis.

D. Methods of Analysis

There are two methods employed at the present time for the bacteriological analysis of both raw water samples and potable water samples. These are the methods of the Most Probable Number (MPN) and the Membrane Filter (MF).

D-1. Multi-Tube Method and the Most Probable Number (MPN)

This is the oldest method of estimating the number of coliform organisms present in a sample of water. This method utilizes the production of gas by the coliform organisms in lactose as given in the presumptive test; or the ability of E.coli to produce indol from tryptophan and acid in glucose fermentation; and the ability of A.aerogenes to produce a compound, acetylmethylcarbinol, in glucose-peptone medium, using sodium citrate as its sole source of carbon for cell metabolism. The above unique characteristics of E.coli and A.aerogenes define the confirmed and complete tests (22) for these coliform organisms. The presence of any of the species is marked by a positive reaction (+) on a test sample while a negative reaction (-) indicates its absence. But it must be mentioned that a positive reaction (+) can be brought about by one, ten, or any number of organisms.

In 1915 McCrady (23) propounded the Most Probable Number (MPN) technique of estimating bacterial densities from fermentation tube tests. He later prepared extensive

tabulations of the MPN corresponding to various decimal dilutions used in laboratory examination of water.

In 1917 Greenwood and Yule (24) put forward another formula also based on statistical analysis for the estimation of the MPN.

In 1925, Reed (25) revised the formula of Greenwood and Yule and adapted it for specific application to the coliform organisms. When applied to the five tubes of each of the three dilutions (N_1, N_2, N_3) to give p_1, p_2, p_3 positive results respectively, the formula is of the form (26):

$$y = \frac{1}{A}\left[(1-e^{-N_1\lambda})^{p_1}(e^{-N_1\lambda})^{5-p_1}\right]\left[(1-e^{-N_2\lambda})^{p_2}(e^{-N_2\lambda})^{5-p_2}\right]$$
$$\left[(1-e^{-N_3\lambda})^{p_3}(e^{-N_3\lambda})^{5-p_3}\right].$$

where λ is the concentration of coliform organisms per ml of sample,

y is the probability of occurrence of the particular results if the concentration from which N_1, N_2, N_3 were drawn was λ,

A is a constant for any particular set of conditions.

If a test where decimal dilutions were used gives

(i) All five of 10 ml dilution (+)

(ii) Three out of five of 1 ml dilution (+)

(iii) None of the five of 0.1 ml dilution (+),

i.e. $N_1 = 10$; $p_1 = 5$
$N_2 = 1.0$; $p_2 = 3$ } 530 Result
$N_3 = 0.1$; $p_3 = 0$

then:

$$y = \frac{1}{A}\left[(1-e^{-10\lambda})^5\right]\left[(1-e^{-\lambda})^3(e^{-2\lambda})\right]\left[e^{-0.5\lambda}\right]$$

$= \frac{1}{A} f(\lambda)$

= Probability of occurrence of the above 5.3.0 result if the concentration of the coliform organisms in the sample is λ per ml.

By plotting Ay for various assumed values of λ, it is possible to find the value of λ for which the probability of occurrence (y) is a maximum. This defines the most probable number of the organisms present in the water sample and responsible for the positive reactions.

Alternatively, by differentiating y with respect to λ and setting it to zero $\left(\frac{dy}{d\lambda} = 0\right)$, an equation in λ only can be solved to find the most probable number $\bar{\lambda}$. By multiplying $\bar{\lambda}$ by 100, the conventional MPN per 100 ml is obtained.

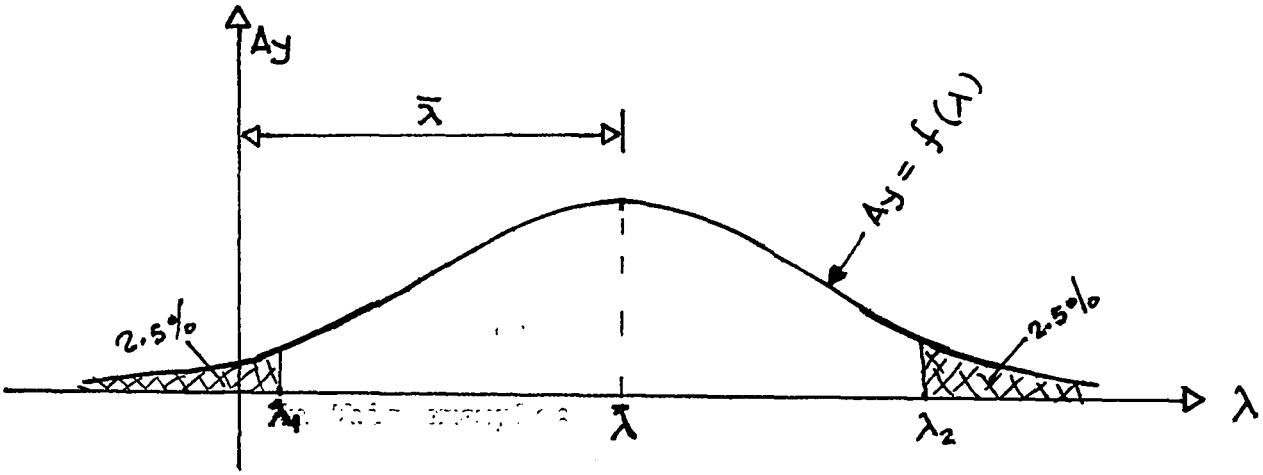

Figure 4: Probability Curve Ay=f(λ).

In this example:

$$A_y = \left(1 - e^{-10\lambda}\right)^5 \left(1 - e^{-\lambda}\right)^3 e^{-2.5\lambda}$$

For $\lambda = 0.6$; Ay = 0.0214

$\lambda = 0.8$; Ay = 0.0225

$\lambda = 1.0$; Ay = 0.0206

$\therefore \bar{\lambda}$ must lie between 0.6 and 1.0.

Actual value of $\bar{\lambda}$ = 0.79/ml of sample,

and MPN = 79 per 100 ml.

Values of $\bar{\lambda}$ expressed per 100 ml are given in tables (22) for various values of N and p.

Results obtained at very low and very large values of λ show poor agreement with expected results. This introduces the concept of <u>confidence limit</u> - the limits of λ within which the results can be considered reliable (27). As in all probability curves, the entire area enclosed by the curve in Fig. 4 sums up to unity.

$$\int_{\lambda=-\infty}^{\lambda=\infty} f(\lambda)\, d\lambda = 1$$

In defining the 95% confidence limit, the 2.5% section of the area of the curve at each end is discarded. Only λ for which

$$\lambda_2 \leq \lambda \leq \lambda_1$$

is accepted as a reasonable concentration. For the above example:

$\lambda_1 = 0.25$

$\lambda_2 = 1.90$.

Any method of enumerating the coliform organisms which gives coliform density in the same water sample between 25 and 190 per 100 ml will be accepted as correct within the 95% confidence limit.

This statistical approach, using the wide spectrum within the 95% confidence limit, becomes too flexible to be depended upon as an absolute figure.

1-D-2. Membrane Filter Technique

The first organized and systematic study of the use of so-called molecular filter membranes was made in 1929 by H. Bechhold in Germany (28), though Sanareli had studied the use of collodion membranes as far back as 1891. By 1916, the work of Zsigmondy and Bachmann led to small industrial production of these membranes in Germany. In the 1930's, membranes were produced in France and England for laboratory use only. During the Second World War, extensive use of membranes was made in Germany for bacteriological analysis of water when most of the laboratories were destroyed by air raids. The success of the use of membranes in Germany attracted both military interest in connection with their defensive applications in biological warfare, and civil sanitary interest in connection with water sanitation (29) in the United States. Before 1950, the California Institute of Technology under Dr. A. Goetz (28, 29) was producing membranes for the United States Army and other laboratories such as the Environmental Health Centre in Cincinnati. Between 1950 and 1960, numerous research groups in America (30, 31, 32, 33) developed both the membrane filters and the appropriate media for use for various cultural denominations of interest in water sanitation.

In an Executive Committee of the American Water Works Association in November 1957 (34) a resolution accepting the Membrane Filter as an alternative to the complete test for members of the coliform group of organisms was adopted, but with some reservations. It is only in the 13th edition of the Standard Methods (22) that this method was accepted without many qualifications.

The success expected from the Membrane Filter method depends to a large extent on the medium for incubating the organisms. In 1951, Clark (35) and his co-workers suggested a two-step procedure using Albimi medium and a modified Endo medium for the coliform organisms. In the same year, Goetz (28) and his co-workers produced a special indicator broth medium. In 1953, Taylor et al. (36) modified MacConkey's medium and adapted it for use with membrane filters. Others have suggested a two-stage procedure including a first stage enrichment medium. The improved medium currently recommended by the Standard Methods (22) was developed by Millipore Filter Corporation in Watertown, Massachusetts. This is the MF Endo medium which has been used for total coliform count in this investigation. Details of this and other media used in this study are given in the Tables 5 to 8 of the Appendix.

R. B. Adams (37) established that there was an agreement between the MPN and MF methods of 83.5% for raw water samples and 88.4% for prechlorinated raw water samples, using a 95% confidence limit for the MPN. G. Yee (38) and his co-workers found a good correlation between the MPN and MF methods in their investigation which focussed on the development of

media. McCarthy et al. (41) indicated that the MF technique appeared to possess greater precision than the MPN test, and better enumeration of all strains of coliform. Thomas et al. (32) obtained a correlation ranging from 75 to 95 percent. In his investigations using the Richelieu River water, Neelakantiah (17) obtained a MPN/MF ratio of 1.15 using 95 percent confidence limit for the MPN. In each case, the MF has tended to give lower coliform counts than the MPN, but the mathematical bias in the MPN concept must be well understood.

The MF gives a fast result and is most desirable when a distribution system shows signs of contamination. The MPN confirmatory test will require a minimum of 48 hours to produce results that the MF will give in less than 20 hours, so that precautions can be taken in time.

The MF is useful for enumeration of bacteria in

(a) relatively clear water with high or low bacterial densities,

(b) turbid waters with high bacterial populations where adequate dilutions will yield reasonable bacterial densities for the filter.

For turbid raw waters with low bacterial densities, the more conventional fermentation process (MPN) will yield a more reliable result.

The MF method was used throughout this investigation for both the raw water and the treated (product) water.

CHAPTER II

EQUIPMENT

The Reverse Osmosis Research Laboratory of the Department of Civil Engineering and Applied Mechanics was developed in 1957 (12). It consists of six reverse osmosis cells manufactured of stainless steel. Each cell consists of two basic parts, a high pressure upper compartment through which feed water flows, and a lower compartment in which the product water is collected. The reverse osmosis membrane is supported on a sintered metal plate with 50% porosity. Figure* 10 shows a general layout of the components. Detail sections of the cells and piping are shown in Figures 5 through 9. The raw water reservoir has been modified to provide an easier flushing of raw water after each run. The cells are connected in pairs, in parallel, as shown in Figure 5. The rest of the reverse osmosis set-up includes the back-pressure regulators, pressure gauges, a surge tank, and a pair of electro-motor driven Milton Royal Duplex Pumps. Nitrogen gas regulates the pressure in the system. The piping arrangements allow for a recycling of the excess feed water back to the raw water reservoir. All the high pressure area of the piping system is connected by Hooke Gyrolock joints.

The membrane casting equipment has three components - a freezer, a water bath, and stirrers. The freezer has three

*All the photographs are contained in the Appendix.

trays, each of which holds one casting at a time, and a free space for casting the membrane, as shown in Figure 11*. The water bath is equipped with three magnetic stirrers and has a thermostat to maintain a preset temperature. A stirrer and a Fleming-Gray model C-type paint shaker are used for mixing.

Equipment used for bacteriological analysis includes two incubators set at temperatures of 35° C and 44.5° C respectively, a refrigerator set at a temperature fluctuating between 5° and 10° C, and two ovens with temperatures up to 121° C. Disposable Petri dishes, sterilized filter papers, and, where possible, measured ampules of nutrients supplied by Millipore Company, Ltd., were used for the various cultures. Two vacuum-pressure pumps and accessories, shown in Figure 12*, provided the partial vacuum for filtering the samples.

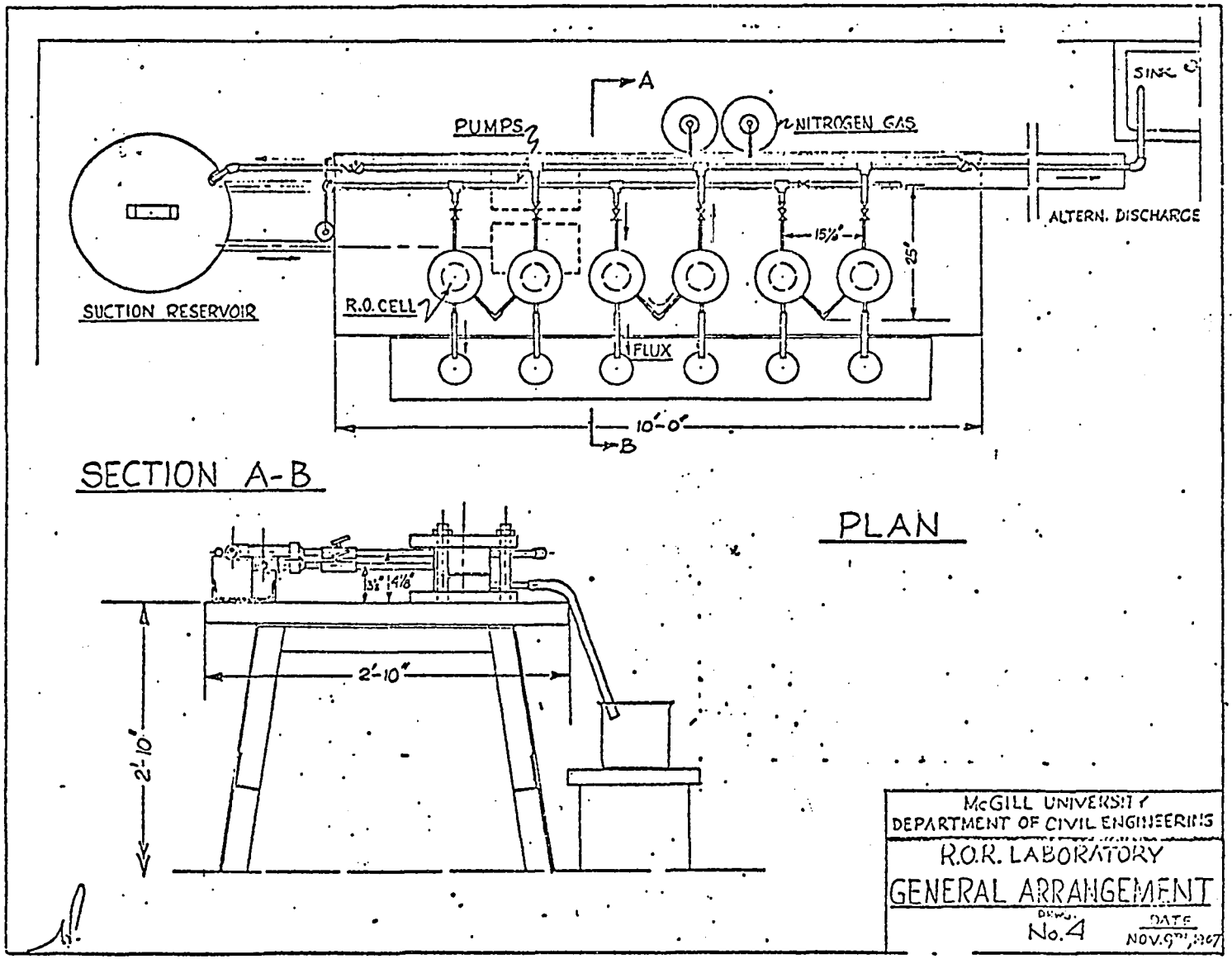

Figure 5. Plan of Reverse Osmosis Equipments.

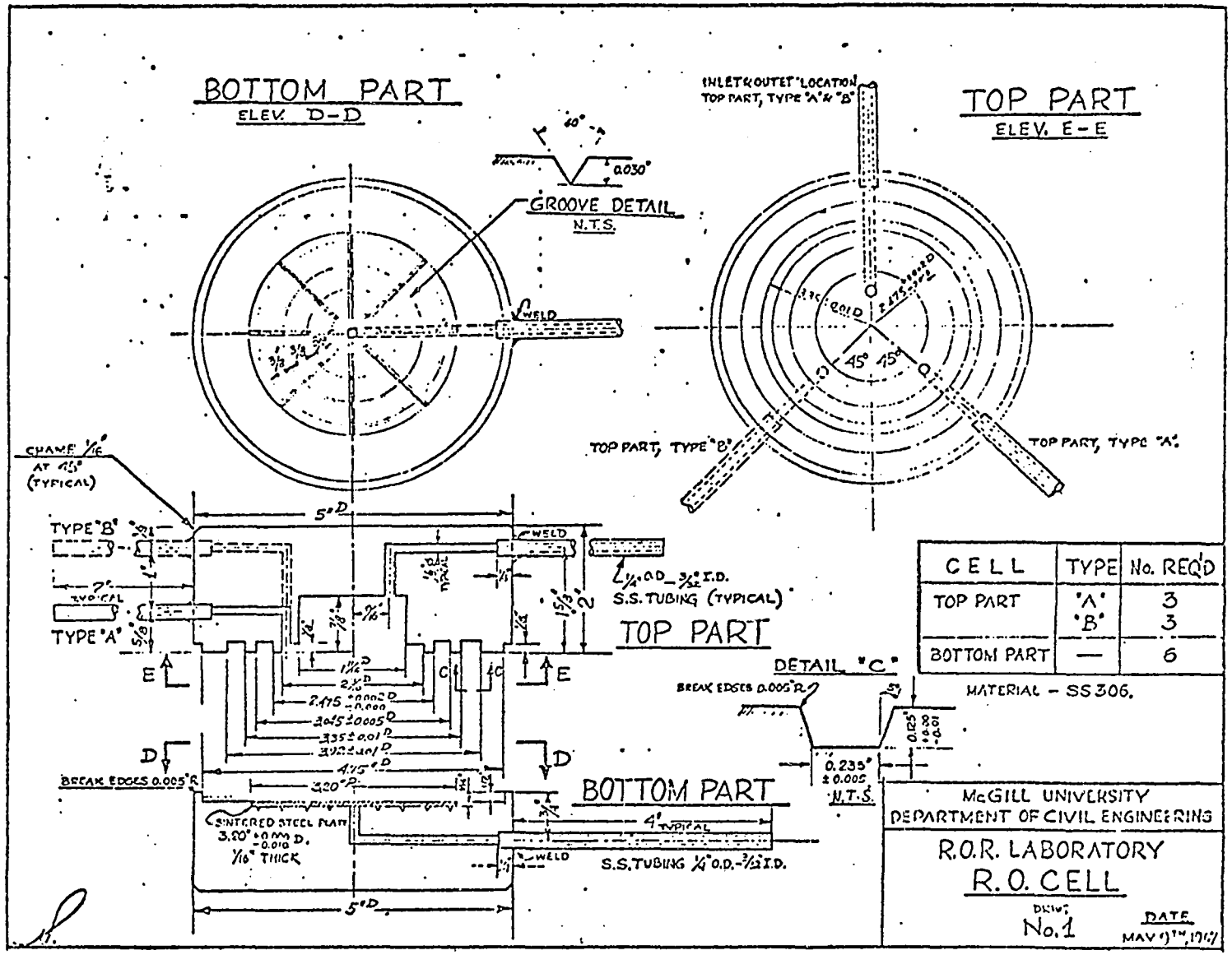

Figure 6. Details of Reverse Osmosis Equipment.

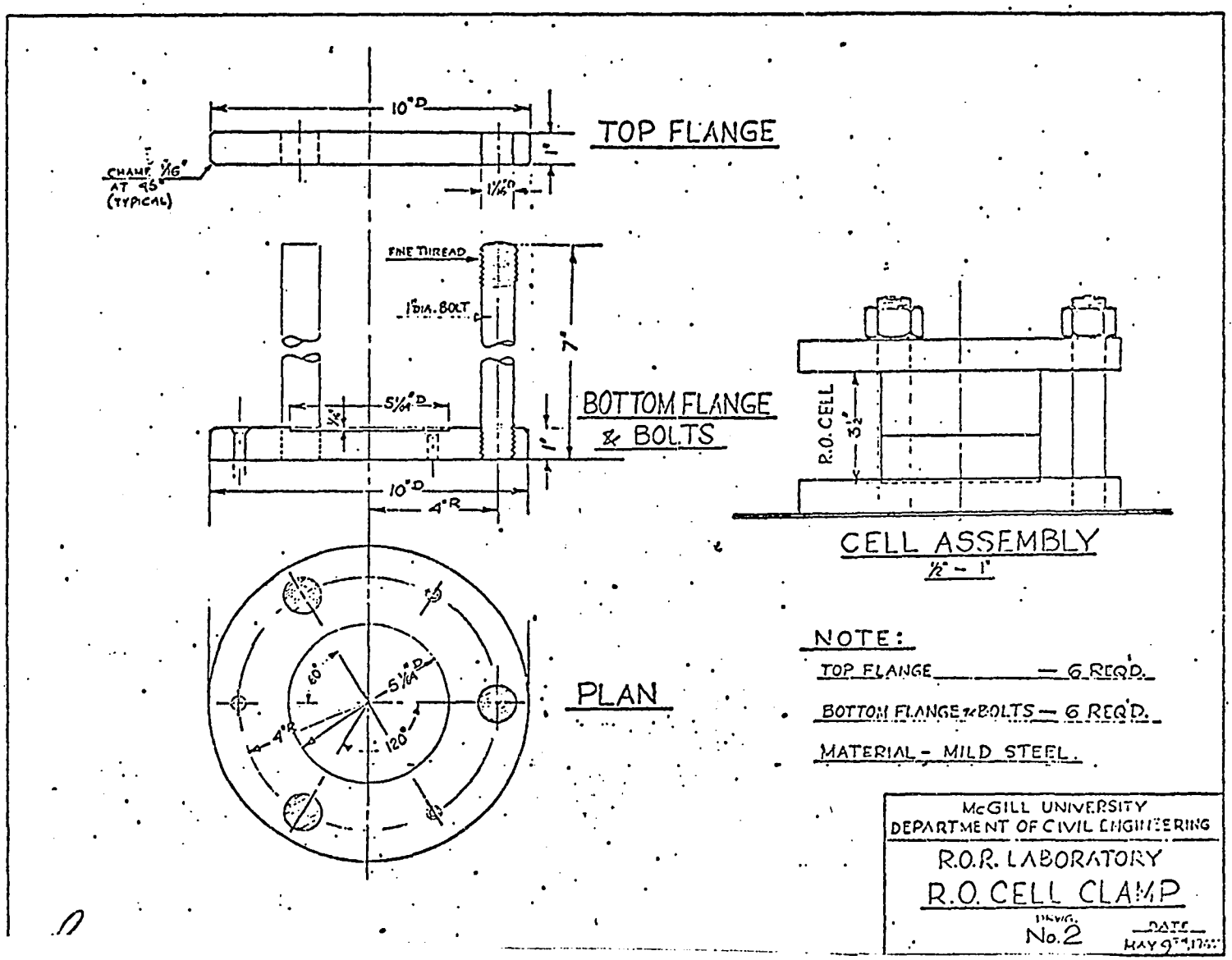

Figure 7. Details of One Reverse Osmosis Cell.

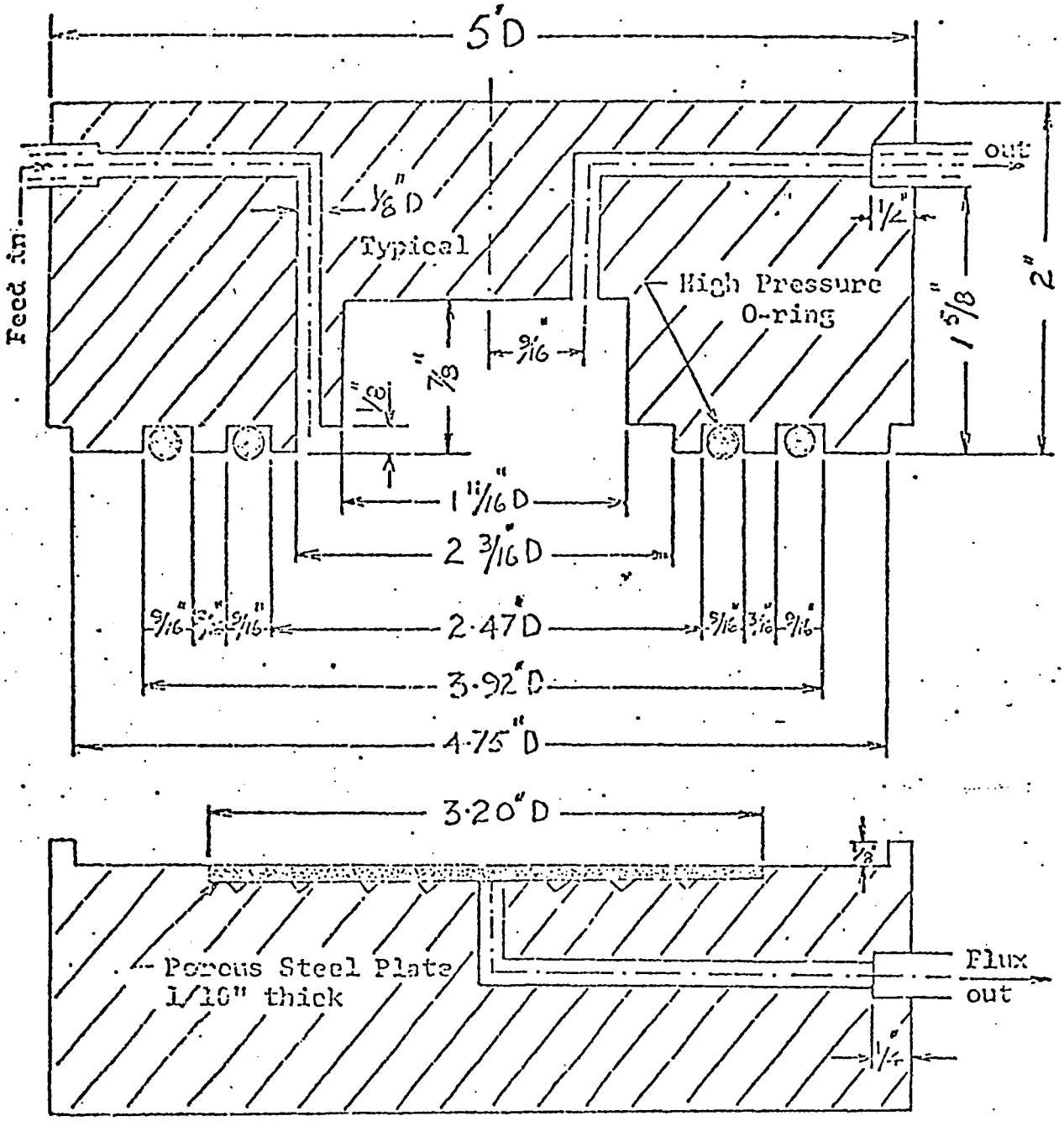

Figure 8. Section through One Reverse Osmosis Cell.

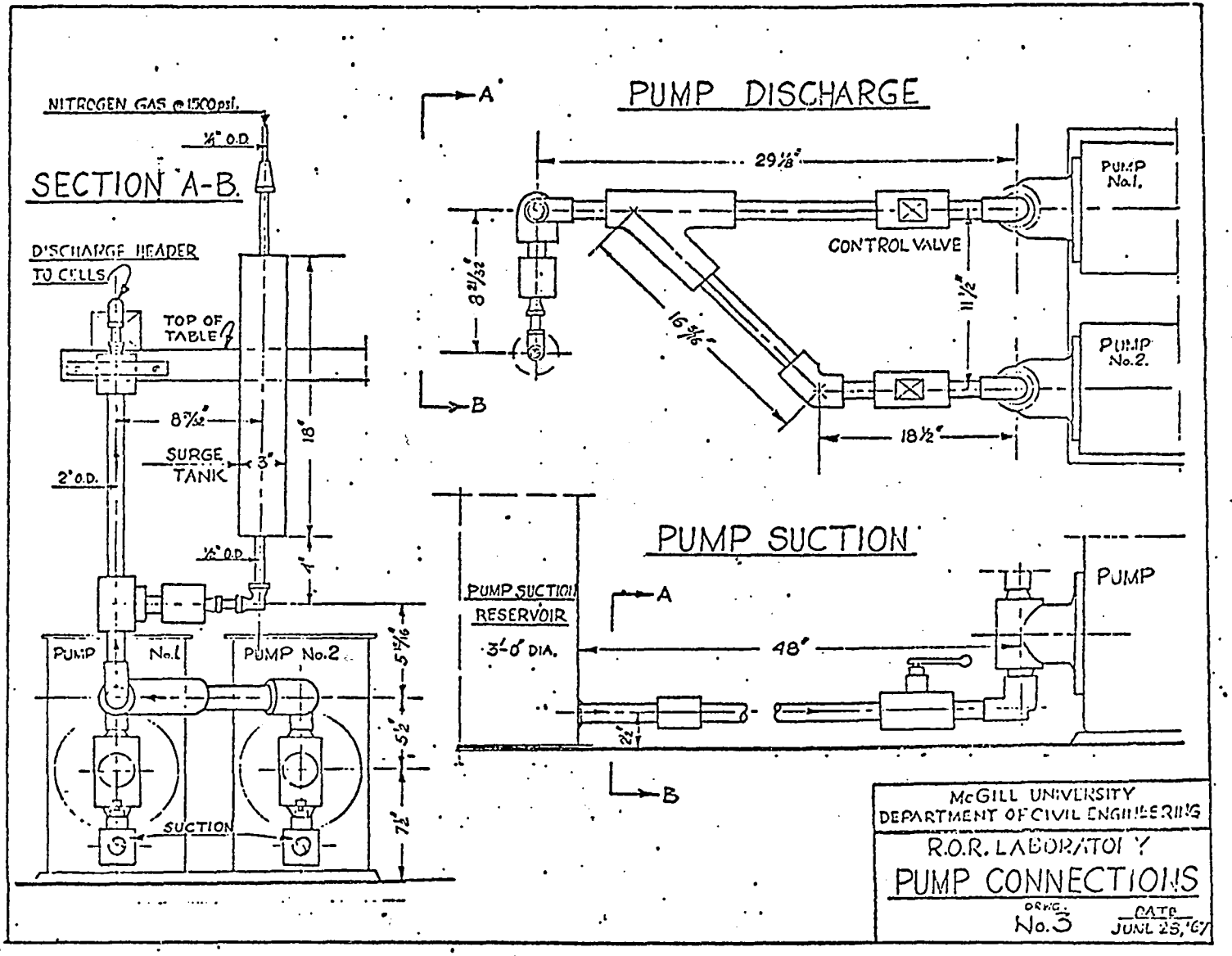

Figure 9. Pump Assembly.

CHAPTER III

PROCEDURES

A. Review of Membrane Casting Procedures

The membrane C-5, developed by C. S. Trivedi (11) in 1971, was used in all the tests. The composition of the membrane is as follows:

i.	Cellulose Acetate	17% by weight
ii.	Acetone	40% " "
iii.	Formamide	23% " "
iv.	Pyridine	20% " " .

No other additives were used.

The liquid components of the casting solution are measured into a jar for each casting as follows:

Acetone	170 ml
Formamide	70 ml
Pyridine	70 ml.

Corresponding to these volumes of the liquid components, 56 gm of cellulose acetate powder is added.

A stirrer is placed into the jar containing liquid mixture so as to have the stirrer blade ½" to 1" below the liquid surface. With the stirrer in motion, the solid powder of cellulose acetate is added slowly. The jar is mixed for 15 minutes.

A rubber band 3/8" wide is wrapped around a jar cover without a hole in it to seal the space between the jar

and cover. The covered jar is agitated in a paint shaker for three hours. Table 25 in the Appendix is a typical membrane casting record.

The mixed solution is placed in a freezer overnight to minimize the loss by evaporation of the volatile acetone component. A glass casting plate is cooled in the freezer for a minimum of 30 minutes to bring its temperature to that of the casting solution. Aluminum trays with about 1" of water are cooled inside the freezer to a temperature of 4° C or less using ice blocks if necessary. The glass rod for membrane casting is also stored in the freezer.

About one inch of the casting solution spread over three quarters of the casting plate width is rolled with the glass rod over the entire length of the plate. The rod rests on the side runners, creating a membrane corresponding to the thickness of the runners. The cast membrane is left for 5 minutes to evaporate in the evaporation medium, which consists of air saturated with the vapour of the solution at the temperature of the freezer, about 4° C.

The casting plate with the membrane is immersed into aluminum trays located in the freezer at a temperature of 0° to 4° C for a period of 1 hour. Then the membrane is removed from the casting plate and cured for 5 minutes in the water bath set at a predetermined temperature. For the preliminary investigations, curing temperatures of 50°, 60°, 70°, and 80° C were used for a number of membranes. Final runs were made with membranes cast at 70° C.

The membrane is now ready for use. It is cut into the required circular shape of 4.75" diameter with a cutter and trimmed at the edges with scissors.

It is believed that membranes so prepared and stored in tap water can be used at any time. In this investigation, the maximum storage time never exceeded four days and no deterioration was observed in the membrane properties. To minimize possible contamination, membranes were stored in distilled water.

In the casting process, precautions were taken to ensure that the membrane was not turned over, as it has been observed (11) that the removal characteristics are almost zero if the casting surface of the membrane is reversed.

A finished membrane has a thickness of about 0.01 inches and an effective flux diameter of 2¼ inches corresponding to a flux area of 0.0276 square feet.

B. Bacteriological Analysis

In this study, series of tests were run to enumerate the following bacterial types from samples of river waters:

(a) Plate Count or total bacteria count which can grow on the medium provided.

(b) Total Coliform Bacteria (T.C.).

(c) Fecal Coliform Organisms (F.C.).

(d) Fecal Streptococci Organisms (F.S.).

Series of six-hour runs were conducted on the reverse

osmosis system to determine the behaviour of the membranes at 50°, 60°, 70°, and 80° C curing temperatures. The flux of the various membranes was determined during the first hour of operation by observing discharges at intervals of five minutes. Results are presented in Table 14.

Final runs were made with raw water samples collected from the St. Lawrence River at Pointe-aux-Trembles, located on the Montreal bank of the river.

B-1. Total Bacteria

There are three major types of bacteria - the rod-shaped types generally classified as bacillus, to which the coliform organisms belong; the spherical types classified as coccus, such as streptococcus; and the spiral-shaped, classified as spirillum.

The bacilli vary in size from 0.5 to 1.0 μ in width and 1.5 to 3.0 μ in length. The cocci range from 0.5 to 1.0 μ in diameter, while the spirilli vary from 0.5 to 5 μ in width and 6 to 15 μ in length.

Like a colloid in water, these bacteria can occur dispersed as individual cells, or they can agglomerate in clusters as chains, tetrads, cubes, and palisades. Shaking and agitation break up most of the clusters, and a completely uniform distribution in a sample is practically impossible. Two aliquot samples of water from the same source, taken under identical conditions, can hardly give identical bacterial counts. The Standard Methods (22) estimate a difference of about 37% under ideal sampling conditions. This condition is

much more pronounced when bacterial densities are very high.

Bacteria are the most numerous organisms in nature. With adequate food supply, they grow and multiply by binary fission. In drinking water the amount of substrate present is so small that the theory of bacterial growth is of no practical consequence except in culture media. But as the level of organic pollution in streams increases, more nutrients become available for richer and more populous bacterial development. Bacterial densities become so high that the idea of a total plate count loses any practical significance. Table 4 illustrates bacterial densities in grab samples from some local surface waters, most of which are being used as sources of public water supply. The rivers show total bacterial densities in millions per cubic centimeter. Bacteria enumerated in the total or plate count include all the natural water bacteria which can grow on agar media. To this group belong also the pseudomonas, acetobacta, and xanthomonas which occur both in air and in water. H. G. Neumann (28) noted the undesirable results which may be produced by the presence of atypical organisms, spore-formers, and noncoliform organisms in water samples. With non-inhibiting media, spore-formers produce an ill-defined mat of growth and confluent colonies over the entire surface of a membrane filter. It appears that total or plate count has no meaning for raw waters from streams in their present levels of pollution. Their reduction, however, after a water sample has been passed through a treatment unit helps to assess the efficiency of the unit.

The medium used for total or plate count throughout

this investigation was Millipore 2 ml ampule for Total Count No M000-000-2T, Difco 01-730, corresponding to Standard Methods (22), Section 404C, No. 6, and the procedure given in Section 406 was adapted for membrane filters. Details of the medium are given in Table 5 of the Appendix.

Table 4

Bacterial Counts from Random Water Samples

	Source	Plate Count	Total Coliform Organisms	Fecal Coliform Organisms	Fecal Strepto-cocci
		per cc	per 100 ml	per 100 ml	per 100 ml
1	Tap Water	1.2	1.0	1.0	1.0
2*	Ottawa River at Hawksbury	3×10^6	13,000	1,200	150
3	Richelieu River at Beloeil	3×10^6	85,000	33,000	2,000
4*	St. Lawrence River at Varennes	3×10^6	25,000	2,000	750
5	St. Lawrence River at Pointe-aux-Trembles	3×10^6	300,000	29,000	3,600
6*	St.Maurice River at Three Rivers	3×10^6	1,100	800	100

* Water Works Sites.

B-2. Total Coliform Bacteria (TC)

The coliform group of organisms consists of all aerobic and facultative, gram-negative, nonsporulating bacilli which produce dark colonies within 24 hours on Endo-type medium with lactose at 35° C. When the multi-tube incubation is employed, the distinguishing characteristic is the decomposition of lactose with gas and acid formation within 48 hours at 35° C.

Two predominant groups of the coliform organisms are of importance in Water Supply Engineering practice - the Escherichia coli and Aerobacta aerogenes. The former is found in high concentrations in the intestines of warm-blooded animals, while the latter is frequently found on grains, plants, and soil. There are intermediate groups which according to Pelczar and Reid (39) are also found in the urinogenital tract of man. In this study, the Membrane Filter technique has been used.

The medium of incubation was Millipore's 2 ml Endo ampule No. M000-000-2E, Difco 01-730, corresponding to Standard Methods (22), Section 404C, No. 13. The procedure was as given in Section 408. A list of the composition of the medium is given in the Appendix, Table 6.

B-3. Fecal Coliform Organisms (FC)

These are the coliform group of organisms originating from the intestines of warm-blooded animals and excreted with their feces.

In 1884, Escherich (14), working with the feces of

a cholera patient, isolated a group of bacteria which he identified as Bacterium coli. These strains of organisms later became known as Escherichia coli (E.coli). There are various sub-groups of E.coli and intermediate strains including Escherichia fruendi and Aerobacta cloacae which exist in relatively small numbers in the intestinal tract of warm-blooded animals.

The main objective of conducting a bacteriological examination of raw surface water is to estimate the hazards due to fecal pollution. This enables the designer to select a supply source with lowest contamination.

The use of elevated temperature reactions where coliform organisms of other than fecal source would die has been the main test to distinguish the fecal coliform bacteria in water analysis. Even this method has been restricted to the confirmatory multi-tube Most Probable Number (MPN) tests. In the Membrane Filter technique, the fecal coliforms have only been vaguely distinguished by the "characteristic sheen" of the colonies on the membranes. Metallic sheens, however, differ in intensity, and intermediate coliform groups are hard to distinguish. Counting the wet colonies helps, but the results are not very satisfactory. It is only in the 13th Edition of the Standard Methods (22) that a procedure has been given for the specific isolation of fecal coliforms. Details of the media specifications are included in Table 7 in the Appendix.

Millipore's 2 ml FC ampule No. M000-000-2F, Difco

01-730, corresponding to Standard Methods (22), Section 404C, No. 12, was used according to the procedures of Section 404B. Colonies in the M-Endo media which showed characteristic metallic sheen provided a check on counts obtained by this method. Sample readings are shown in Tables 9 to 13 in the Appendix.

B-4. Fecal Streptococci Organisms (FS)

The group of organisms classified as streptococci can be divided into three major sub-groups - milk streptococci, enterococci, and the streptococci of the mouth and respiratory tract. The commonest milk streptococcus is Streptococcus lactis which is responsible for normal milk souring. The streptococci of the mouth and respiratory tract include a few pathogenic species, and are of no sanitary significance except in swimming pools and beaches.

The enterococci can be further divided into two main species - Streptococcus fecalis, a normal intestinal parasite of man; and Streptococcus bovis, a normal intestinal parasite of cattle and other mammals. These two major species constitute the fecal streptococci. The work of Mallman et al. (40) shows that these intestinal organisms are excreted in relatively smaller numbers per capita per day than E.coli. Their presence in a source of water supply is indicative of fresh contamination.

The medium used for their incubation is M-Enterococcus

Agar - Difco No. 0746-01-8, corresponding to Standard Methods (22), Section 404C, No. 18. Details of the media specifications are listed in Table 8 in the Appendix. The laboratory procedure is as given in Section 409B (22).

C. Preliminary Investigation of River Waters

Raw water samples were taken from the following rivers at the points indicated below:

(i) Ottawa River at the low lift pumps of the Water Filtration Plant, Hawksbury, Ontario.

(ii) Richelieu River at Beloeil Yacht Club, Quebec.

(iii) St. Lawrence River at the Water Filtration Plant, Varennes, Quebec.

(iv) St. Lawrence River at a wharf located on 55th Avenue, Pointe-aux-Trembles, Quebec.

(v) St. Maurice River at the low lift pumps of the Water Filtration Plant, Three Rivers, Quebec.

The samples were analysed for the various bacteria discussed previously.

Total Bacterial Count continued to be difficult to filter out (MF) in dilutions reasonable enough to give reliable results. For instance, making one litre of dilution from 1 ml of raw water (0.001 per ml) could not give distinct colonies even if 0.1 ml from the dilution was filtered through. Typical results are presented in Tables 9 to 13 in the Appendix.

It was observed that of the various sampling points, the St. Lawrence River at Pointe-aux-Trembles consistently gave the highest bacterial densities. Final runs were therefore

made with samples collected from this point. This observation is, however, not surprising in view of the numerous trunk sewer discharges scattered all along the Montreal bank of the river.

D. Experimental Procedures

Raw water for the several experimental runs was collected in 5-gallon plastic containers from rivers and locations listed for each run. The test runs were started without delay so as to obtain the natural bacteriological conditions similar to those in the rivers. Before a run, the collected water was analysed for the coliforms, fecal coliforms, and fecal streptococci. Temperature in the raw water tank was recorded.

Dilutions were made with buffered dilution water prepared with potassium dihydrogen phosphate, KH_2PO_4, and sodium hydroxide, NaOH, as specified in the Standard Methods (22). Distilled water alone was not used directly for dilution or for rinsing the funnels.

Coliform analyses of raw water samples were made by filtering two samples containing 0.01 ml of raw water per 100 ml, and two samples containing 0.005 ml of raw water per 100 ml. This inoculum was established in the preliminary studies to give convenient colony counts after incubation. By means of a pair of sterile forceps, a sterile membrane filter was placed over the porous plate of the filter assembly with the grid side facing up. A sterile, seamless glass funnel was clamped onto the porous plate to hold the filter tightly in place. This tight fitting ensured that all the liquid passed through the filter without any mechanical damages to the pad and without any leakage losses. When the test sample was poured into the funnel, the electrically-driven vacuum pump sucked it into a 1-litre receptacle which

held the filtrate. A vacuum of the order of 20 to 25 inches of mercury gave a most effective, smooth, and rapid filtration. Lower pressures were observed to cause ridges and wrinkles on the filter. Such wrinkles occasionally caused clusters of colonies at the edge, thereby introducing errors in the count.

The funnel was rinsed with buffered dilution water to ensure that no bacteria were left on the walls of the funnel. In this way, it was usually not necessary to re-sterilize the funnel between runs. The filter was removed with sterile forceps and placed in a disposable Petri dish with pad soaked in 1.8 ml of the Endo broth described earlier. Placement of the filter paper onto the pad required some care in order to avoid entrapping air bubbles between the interfaces. Air bubbles were observed to interrupt uniform distribution of the broth on the filter, resulting in poor colony development in the dish. Dishes were inverted, marked, and incubated immediately.

Two dilutions used for fecal streptococci in the raw water were 1 No, 10 ml, and 1 No 1 ml. This range of dilutions usually gave good colony distributions. The agar medium already poured into the Petri dish solidified very readily. Any visible air bubbles were removed by flame. The filter paper required gentle rolling also to avoid air entrapment. Dishes were inverted, marked, and incubated without delays.

All red and pink colonies in the agar dish after

48 hours at $35^{\circ} \pm 0.5^{\circ}$ C. were counted as fecal streptococci. All dark colonies appearing on the Endo media with a metallic-appearing surface lustre after 20 to 24 hours at $35^{\circ} \pm 5^{\circ}$ C. were counted as coliform colonies. Distinctive colonies with brilliant metallic sheen were tentatively enumerated as fecal coliforms. Confirmatory enumeration of the fecal coliforms was made as described earlier.

Colonies were counted with a Unitron MSF stereoscopic microscope with 10X eyepieces and two objective lenses of 1X and 2X magnifications fitted on the objective slideway. This made it possible to obtain magnifications of 10 and 20 times which are adequate for well-developed colonies. Illumination was provided by means of a spotlight illuminator with an adjustable focussing condenser to give a brilliant, concentrated light spot on the white base plate.

Colonies were expressed in terms of coliform or streptococci per 100 ml, as follows.

$$\text{Bacteria per 100 ml} = \frac{\text{No. of colonies counted}}{\text{ml sample filtered}} \times 100$$

100 ml of product water from the reverse osmosis cells was filtered directly through the MF filter and incubated for coliform count. Since the standards of drinking water are fixed from the number of the more resistant and abundant coliform organisms, the product water from the process was analysed for the coliform organisms only. However, samples were occasionally analysed for fecal streptococci. Samples of the product water were collected with sterile

flasks from sterilized 1-litre graduated cylinder receptacles every two hours unless otherwise stated in the recorded observations. The MF filter funnel was always sterilized either by immersion in boiling distilled water for five minutes or in 95% ethyl alcohol for the same period of time. This special aseptic precaution is not usually required with MF filter procedures, but was considered necessary to ensure that any coliform organisms in the filter would come from the product water only.

Raw water samples and temperatures from the tank were taken simultaneously with the product water samples. As mentioned earlier, raw water samples were analysed for both the coliforms and fecal streptococci. The yield or flux from each test membrane was read every hour and discarded. This method ensured that the quality of the product water was that of the previous hour only, and that bacterial counts were those which passed through the membrane within the hour. Graduated cylinder receptacles were also sterilized either with boiling distilled water or with ethyl alcohol.

CHAPTER IV

EXPERIMENTAL WORK AND RESULTS

A. Effects of Membrane Characteristics and Raw Water on Flux.

In conventional water treatment plants, both the nature of the raw water and the characteristics of the treatment media control the rate and quality of the treated water. Slow sand filters with effective sand size of 0.3 mm have an average depth of about 3 ft. and yield flux of the order of 50 Igpd/sq.ft. Rapid sand filters after extensive chemical pretreatment of the raw water yield flux of the order of 2,880 Igpd/sq.ft., and up to 10,000 Igpd/sq.ft. in modern multi-media filters. Laboratory investigations conducted on reverse osmosis for water treatment by various research organizations mentioned earlier have yielded flux not exceeding about 80 gpd/sq.ft. Municipal supplies processed by reverse osmosis have as their supply sources brackish ground waters where the level of turbidity is relatively low (44). When surface waters are used, special pretreatment units have been required to reduce the turbidity, and even under these conditions the flux has been low.

This investigations was conducted using surface waters without any pretreatment except prechlorination applied to some of the samples at the sample collection source. The prechlorination was of the order of 0.5 to 1.0 ppm which reduced the bacterial density of the raw water. Laboratory tests

always showed a zero chlorine residue before the beginning of each run. Turbidity, colour, suspended matter, and dissolved solids remained at the original levels of raw water.

Flux obtained varied from 30 to 130 Igpd/sq.ft. Results for short runs of six hour durations are shown in Table 14. Figure 13 shows the results for short runs of six hours for membranes cast at 50° C, 60° C, and 80° C. Figure 14 shows plots of extended run results varying from one day to two and a half days. These plots are made on semi-logarithmic scales.

Table 14

Relationship of Flux, Temperature, and Time

Duration of Run - 6 hrs.
Raw Water Source - St. Lawrence River at Pointe-aux-Trembles.

Duration hrs.	Flux Igpd/ft^2			
	50°C	60°C	70°C	80°C
1/12	109	99	120	35
1/6	103	103	130	35
1/4	100	99	128	33
1/3	115	96	126	31
1/2	-	-	125	33
2/3	-	-	120	32
3/4	-	-	119	33
5/6	-	-	116	33
1	115	105	115	33
2	95	90	91	35
3	94	83	69	31
4	72	67	65	34
5	78	66	60	33
6	61	63	57	33

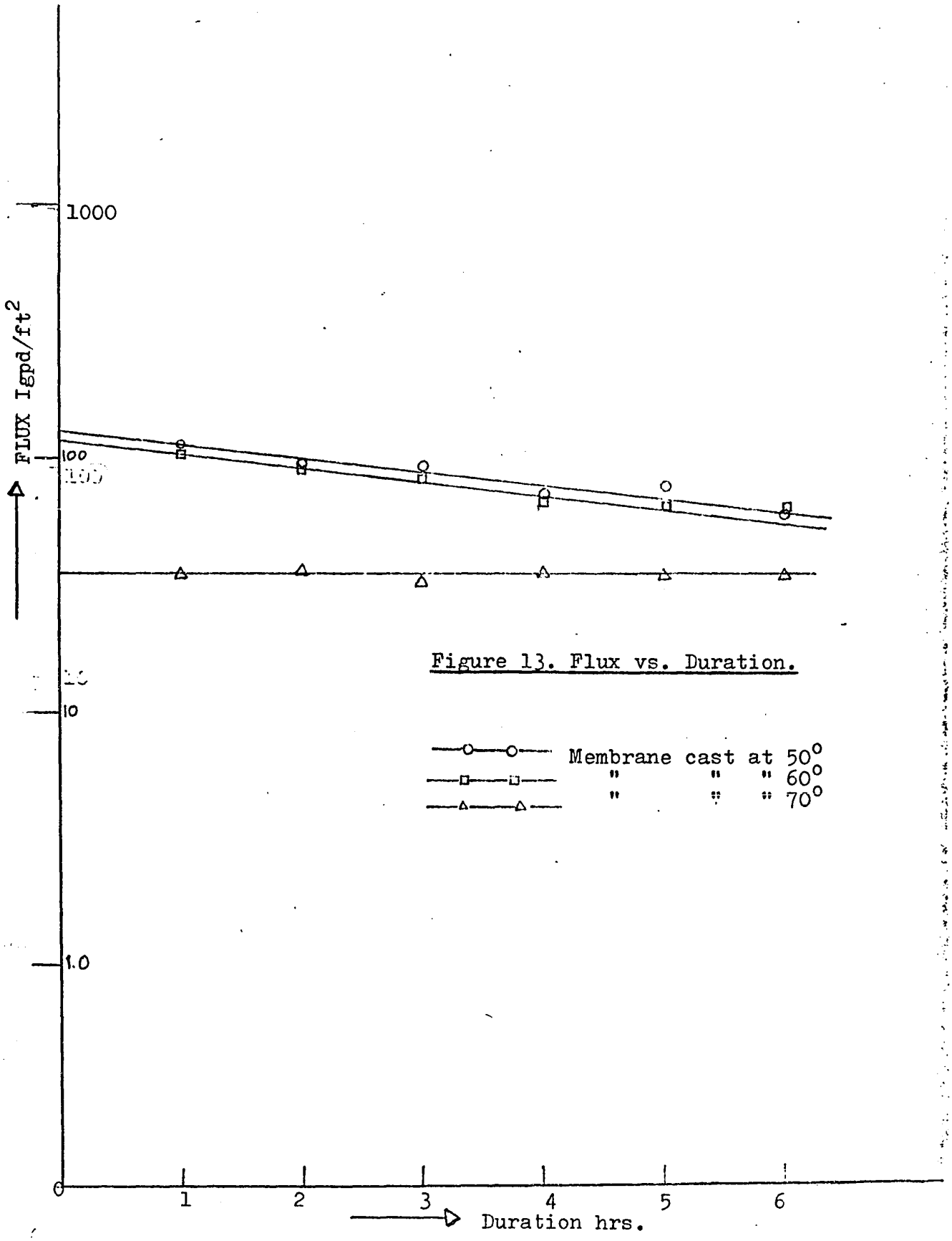

Figure 13. Flux vs. Duration.

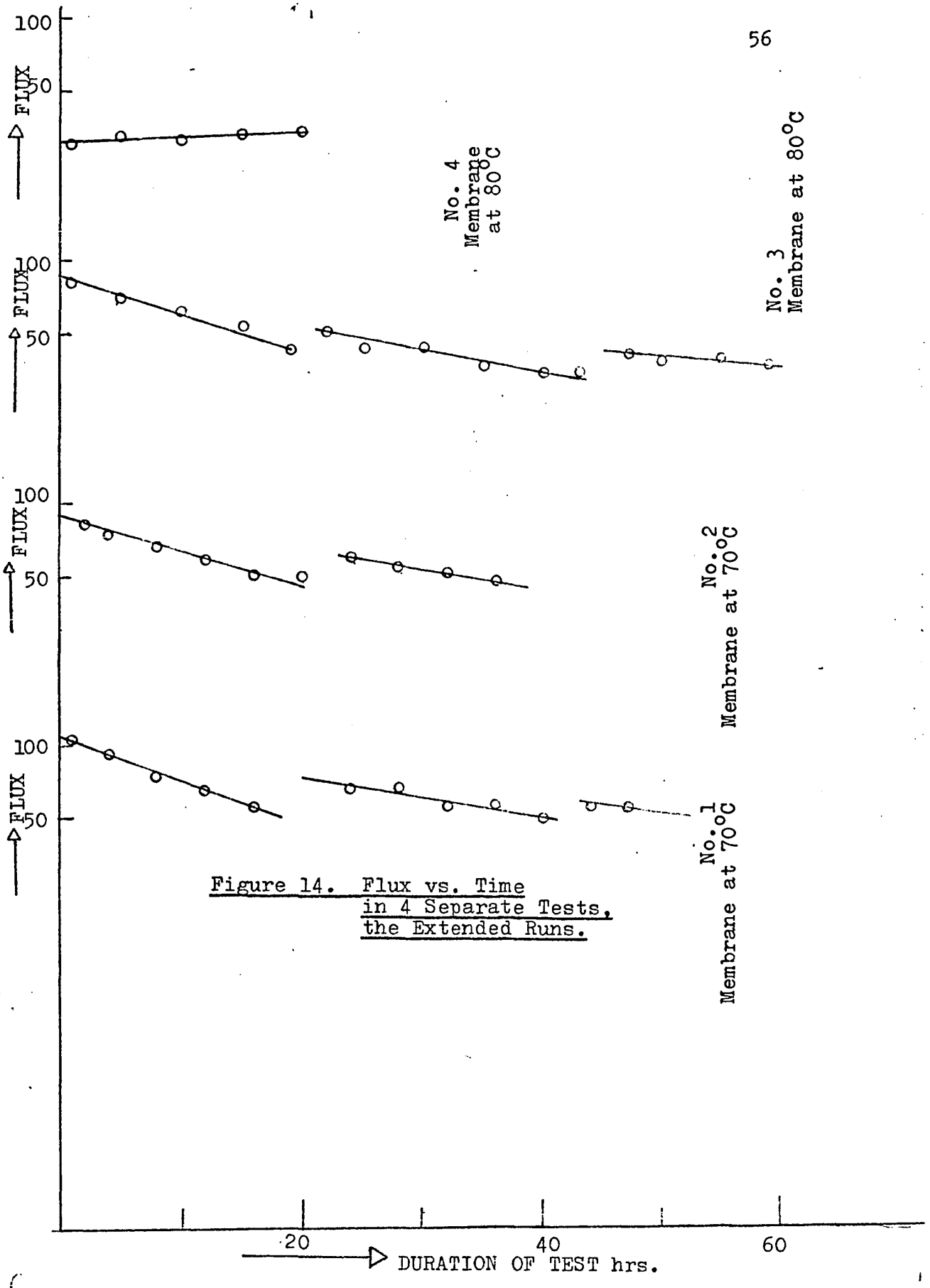

Figure 14. Flux vs. Time in 4 Separate Tests, the Extended Runs.

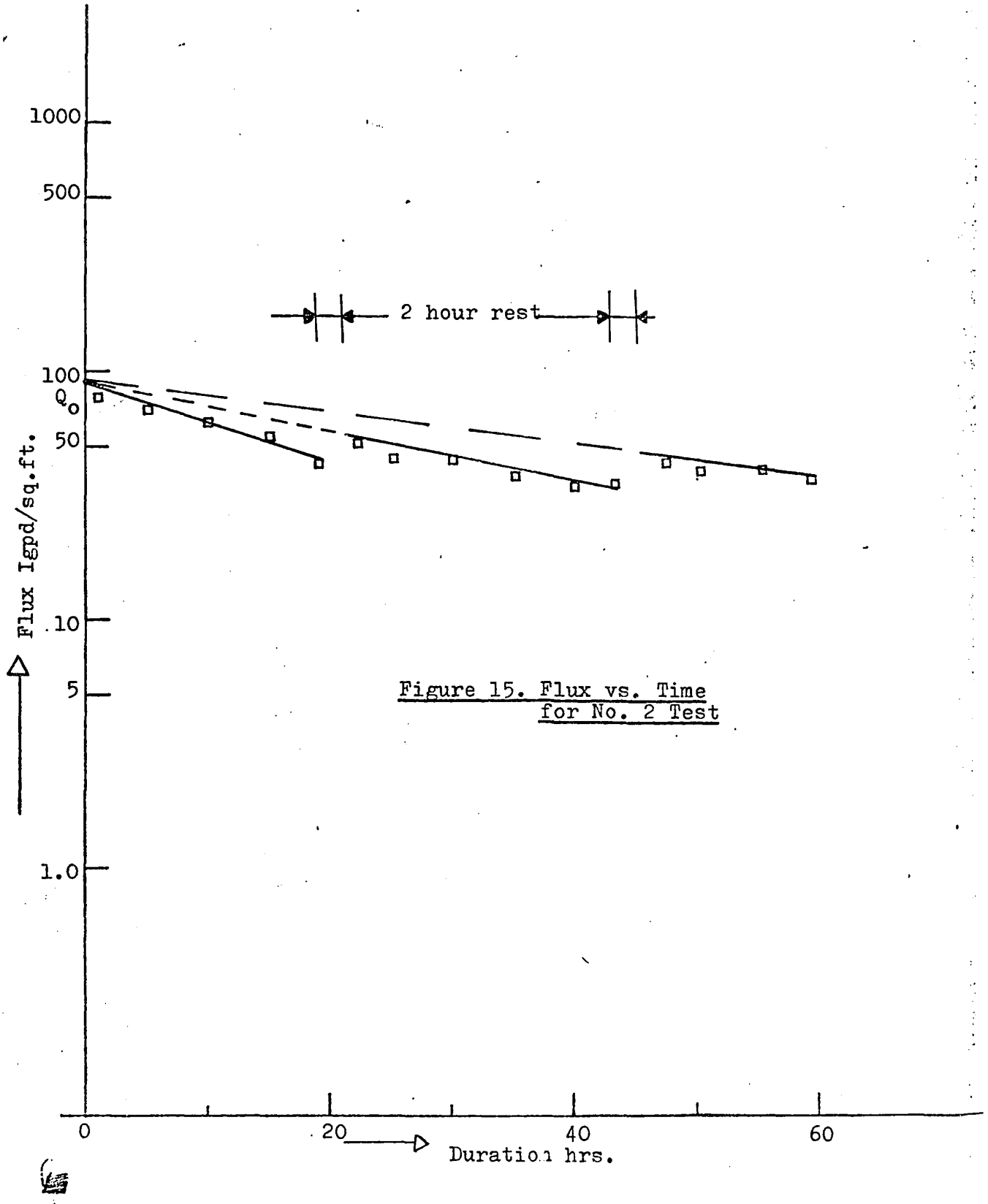

Figure 15. Flux vs. Time for No. 2 Test

B. Temperature and Raw Water Bacteria

Temperature variations in the raw water were obtained by returning excess water within the system to the raw water tank. This recirculation resulted in a temperature rise of nearly 20° C within ten hours of operation with temperature gradients reaching even 6° C per hour during the short runs with a small quantity of water in the tank. Recirculation returned the organisms to the tank by washing off the surface of the membranes. It may be assumed that this effect is negligible when the volume of water in the tank and the flux across the membranes are considered.

The temperature range within which the growth of living organisms takes place is between -5° C and about 80° C. The lower limit of temperature depends on the freezing point of water which forms a large percentage of cell content of living organisms. The upper limit depends on the response of the chemical components of the organism to higher temperatures. Most of the chemical components, the proteins and nucleic acids, are destroyed at temperatures between 50° C and 90° C. The spore-forming bacteria have cysts which can withstand much higher temperatures, while coliforms, as defined earlier, do not form spores.

The coliform group of organisms belongs to the psychrophilic and mesophilic bacteria with temperature optima between 10°C and 45°C. Fecal coliforms, typified by E.coli, have their optimum temperature around the temperature of the normal human body, which is between 35°C and 40°C. The nonfecal and

intermediate groups, typified by Aerobacta aerogenes and E.fruendii, are each distinguished by its entire temperature range for growth. This individual range depends on the normal natural habitat of the individual strain. The cause of these variations in temperature for growth for various types of organisms is not quite clear, but it has been postulated that the enzymes of the psychrophils and mesophils are very unstable at high temperatures and cannot metabolise nutrients required for normal life activities and growth.

The temperature changes in the raw water helped to vary the density of the organisms. This made it possible to study the effects of the variation of these organisms on the removal mechanism. The effects of other physical variables such as the pH, nutrients, sedimentation, adsorption, and competitive life in the water were assumed to be constant.

In one test, the temperature of the raw water was kept fairly constant after the first cycle of an extended run. The results are shown in Table 18 and plotted in Figure 19.

The coliform variation is plotted against time and shown in Figures 16 to 21. The temperature variation is also plotted against time in the same figures. Figures 16 and 17 show the variations obtained in the six hour runs. There is an initial drop of the count in the sample due to prechlorination which was also observed in the extended run shown in Figure 19. The unchlorinated sample shown in Figure 17 showed an initial increase of bacterial count with temperature. This observation repeated in the unchlorinated sample during

the extended run as shown in Figure 18. The increase in count in the extended run with unchlorinated water was followed by a decrease in the count between the temperatures of 36°C and 39°C.

Figure 22* shows the variation of raw water coliform colonies on membrane filters during one of the extended runs.

Tables 19 to 21 show the variation of the fecal coliforms and fecal streptococci with temperature and time. These have also been plotted in Figures 23 to 26. Figure 27* shows the variation of raw water fecal streptococci colonies on membrane filters during one of the extended runs.

Table 15: Total Coliform Survival in Storage Tank.

Membrane Curing t^{o} - $60^{o}C$
Source of Supply - Richelieu River at St. Hilaire.

Test Duration hrs.	Water Tempt. °C	Total Coliforms per 100 ml
0	27	TNTC
1	33	30,700
2	35	37,000
3	37	66,000
4 1/2	39	84,000
5 2/3	41	TNTC

Table 16: Total Coliform Survival in Storage Tank.

Membrane Curing t^{o} - $70^{o}C$
Source of Supply - St. Lawrence River at Pointe-aux-Trembles.

Test Duration hrs.	Water Tempt. ^{o}C	Total Coliforms per 100 ml
0	25	150,000
3/4	31	240,000
1 3/4	34	260,000
2 3/4	35	205,000
3 1/4	36	200,000
4 1/4	37	170,000
5 1/4	38	165,000
6	39	140,000

Table 17: Total Coliform Survival in Storage Tank.

Membrane Curing t^{o} - $80^{o}C$
Source of supply - St. Lawrence River at Pointe-aux-Trembles.
No prechlorination of raw water.

Duration hrs.	Raw Water Temp. ^{o}C	Total Coliform Remaining x 10^{-6} per 100 ml
0	24	0.185
1		
2	28	0.31
3		
4	33.5	0.34
5		
6	36	0.35
7		
8	37.5	0.283
9		
10	38.5	0.47
11		
12	39	-
13		
14	39.5	-
15		
16	39.5	0.41
17		
18	39.5	0.67
19	39.5	1.02
20		
21	35	1.60
22	36	2.93
23		
24	38	2.93
25		
26	38.5	2.85
27		
28	39	1.13
29		
30	39.5	2.20
31		
32	40	1.02
33		
34	40	1.00
35		
36	40	0.42
37		
38	40	0.37
39		
40	40	0.185

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Table 17 cont.

Duration hrs.	Raw Water Temp. ^{o}C	Total Coliform Remaining x 10^{-6} per 100 ml
41		
42	40	0.055
43	40	0.068
44		
45		
46	35	0.058
47	36.5	0.058
48	37	0.023
49		
50	38	0.070
51		
52	38.5	0.068
53		
54	39	0.093
55		
56	39.5	0.185
57		
58	39.5	0.20
59		

Table 18: Coliform Organism Variation with Raw Water Temperature.
Raw Water Source - St. Hilaire filtration plant,
Richelieu River, P.Q.

Duration hrs.	Raw Water Tempt. °C	Total Coliform Remaining x 10^{-6} per 100 ml
0	22.5	0.048
1	25	0.038
2	27	0.053
3	-	
4	30	0.038
5	-	
6	33	0.063
7	-	
8	34	0.223
9	-	
10	36	0.358
11	-	
12	36.5	0.385
13	-	
14	38	0.373
15	-	
16	38	0.380
17	-	
18	39	0.435
19	-	
20	39	0.438
21	-	
22	36	0.463
23	36	0.525
24	36	0.520
25	-	
26	36.5	0.518
27	-	
28	36.5	0.470
29	-	
30	37	0.365
31	-	
32	37	
33	-	
34	37	0.32
35	-	
36	37	0.30

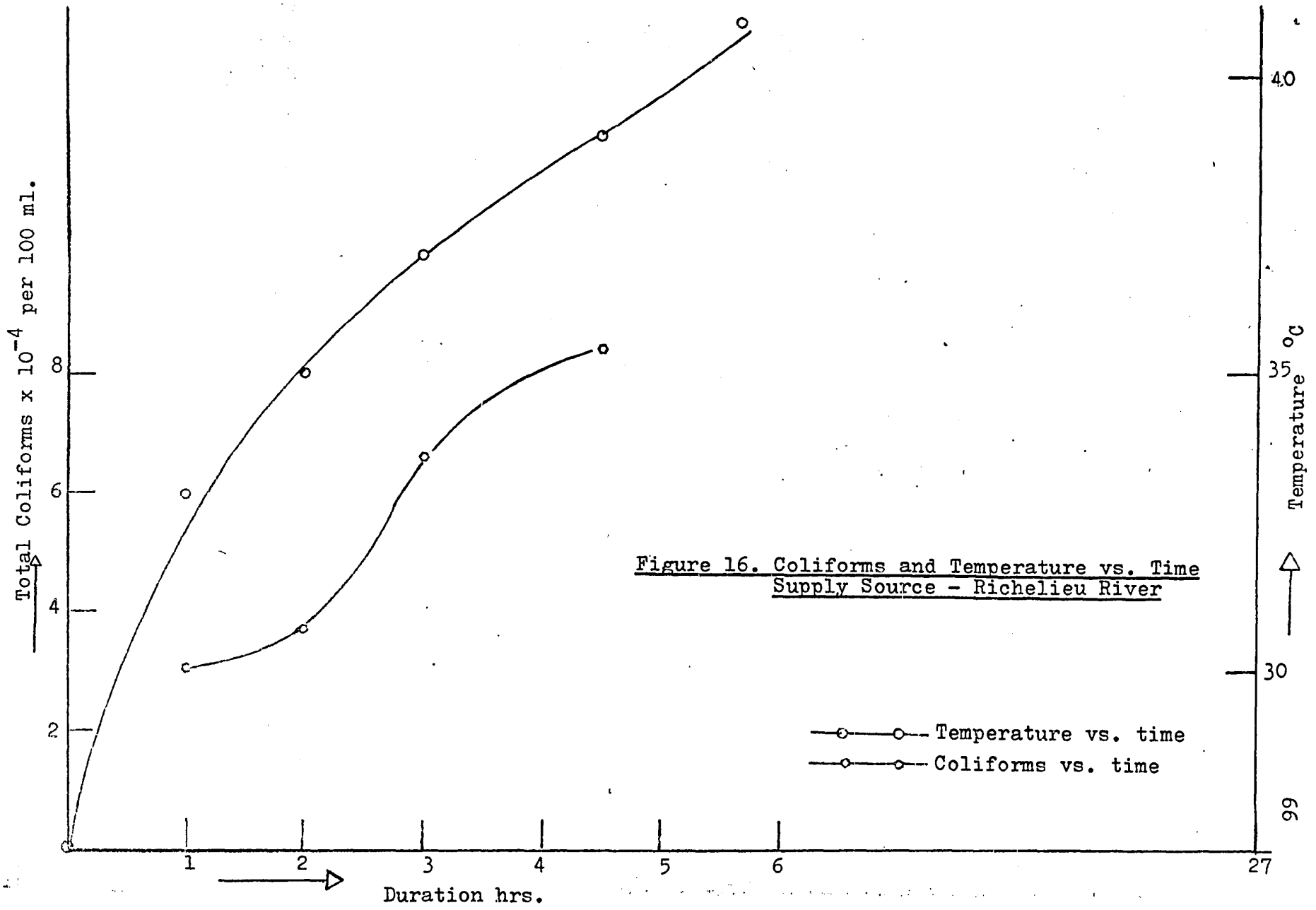

Figure 16. Coliforms and Temperature vs. Time
Supply Source - Richelieu River

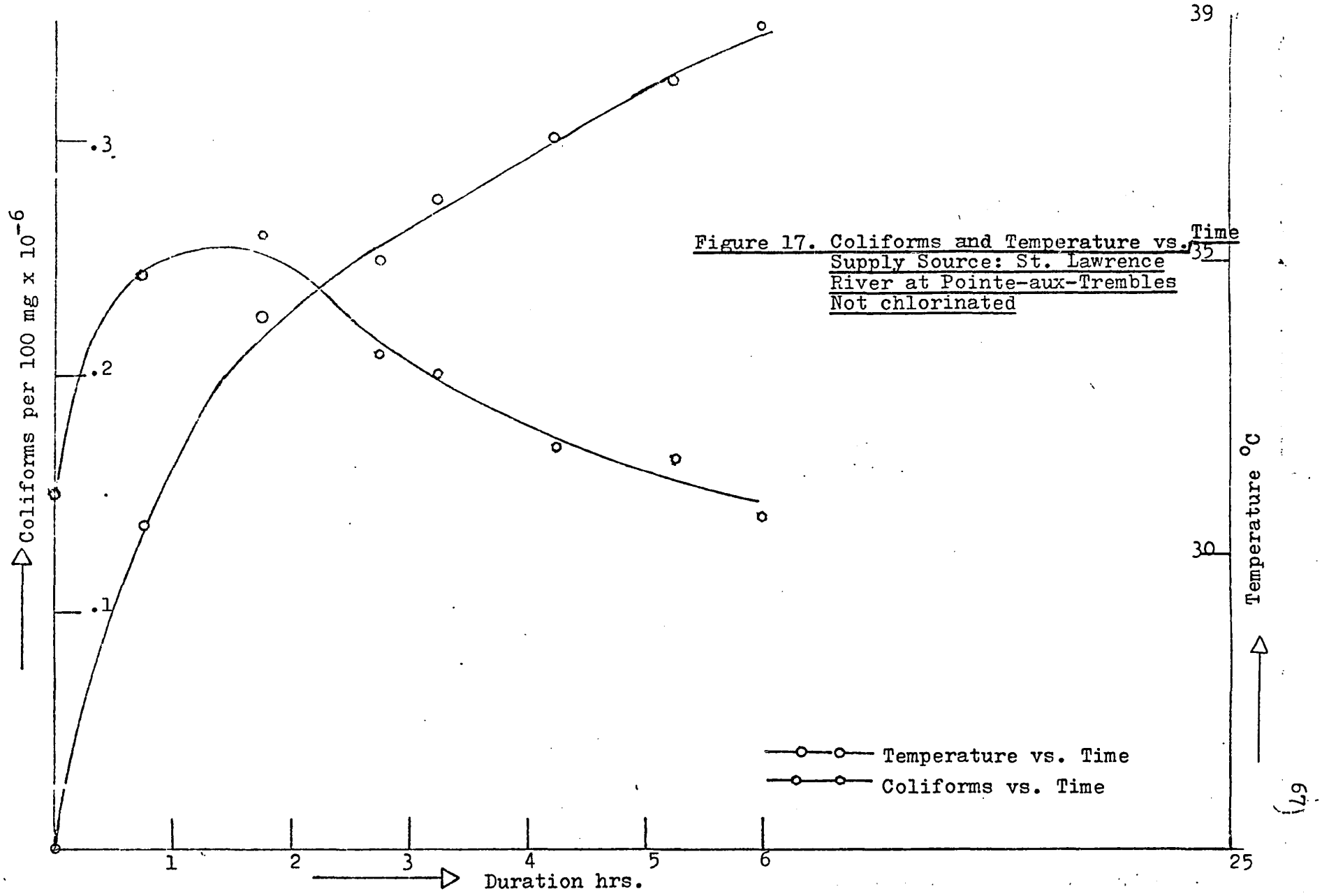

Figure 17. Coliforms and Temperature vs. Time
Supply Source: St. Lawrence River at Pointe-aux-Trembles
Not chlorinated

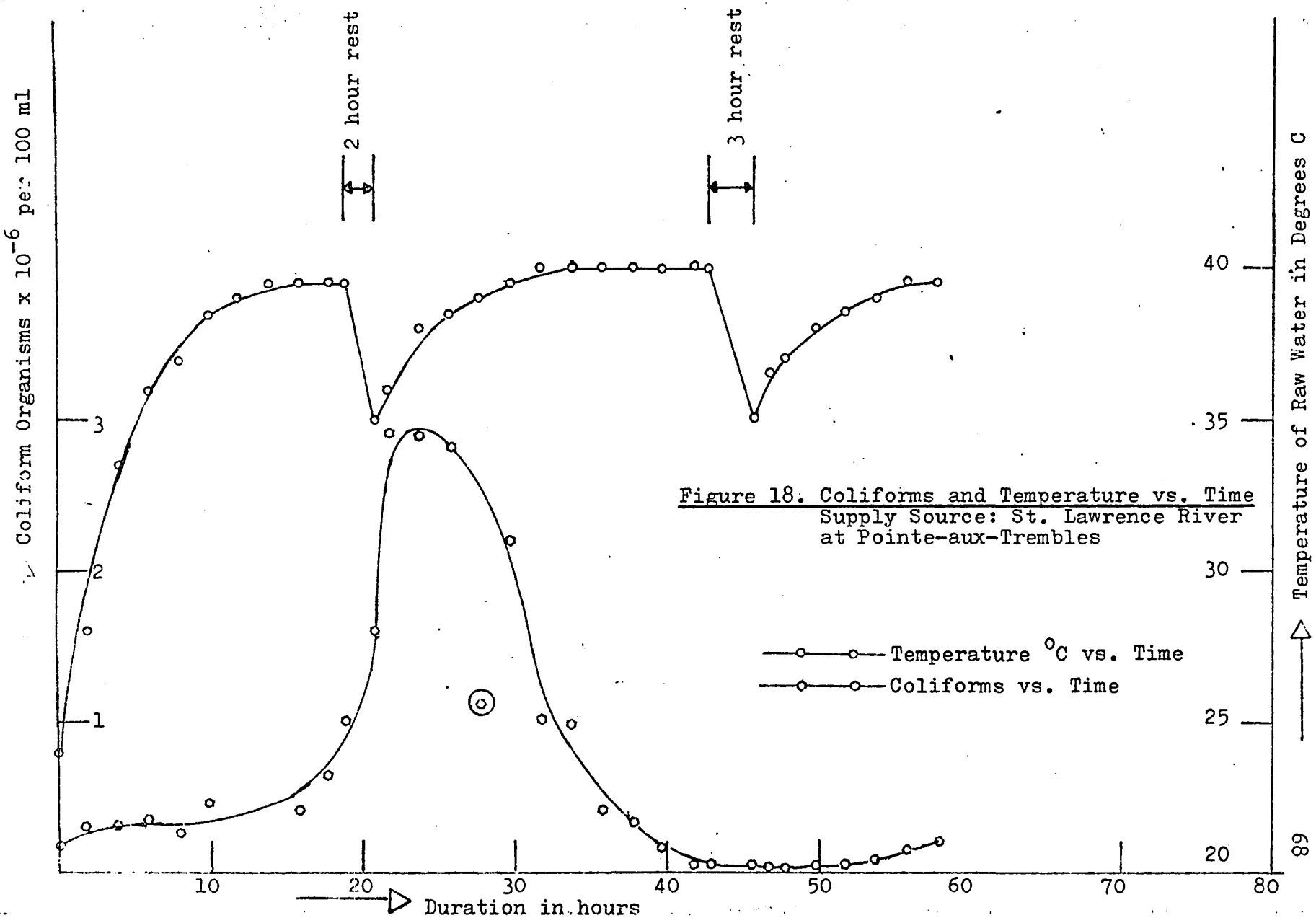

Figure 18. Coliforms and Temperature vs. Time
Supply Source: St. Lawrence River
at Pointe-aux-Trembles

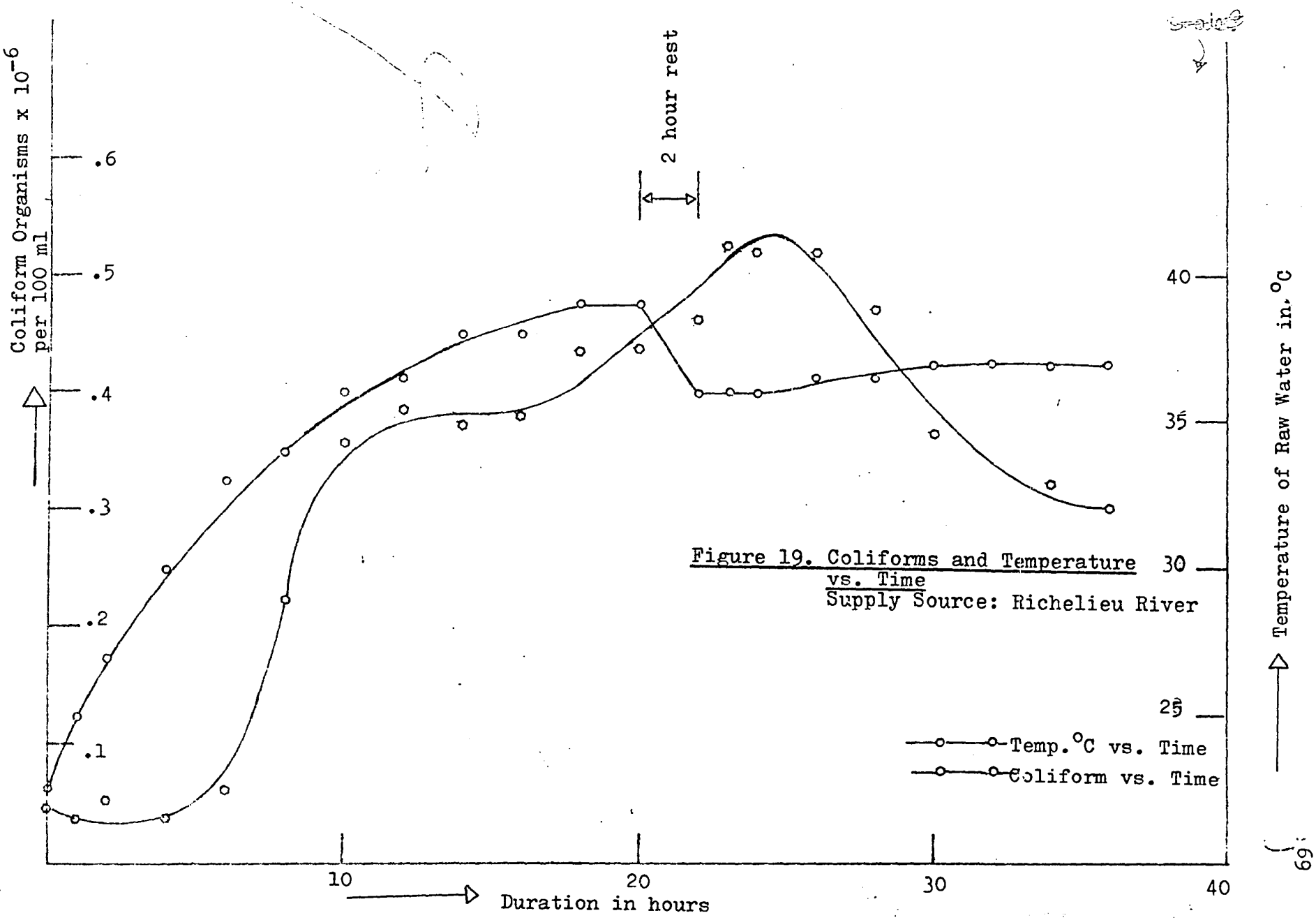

Figure 19. Coliforms and Temperature vs. Time
Supply Source: Richelieu River

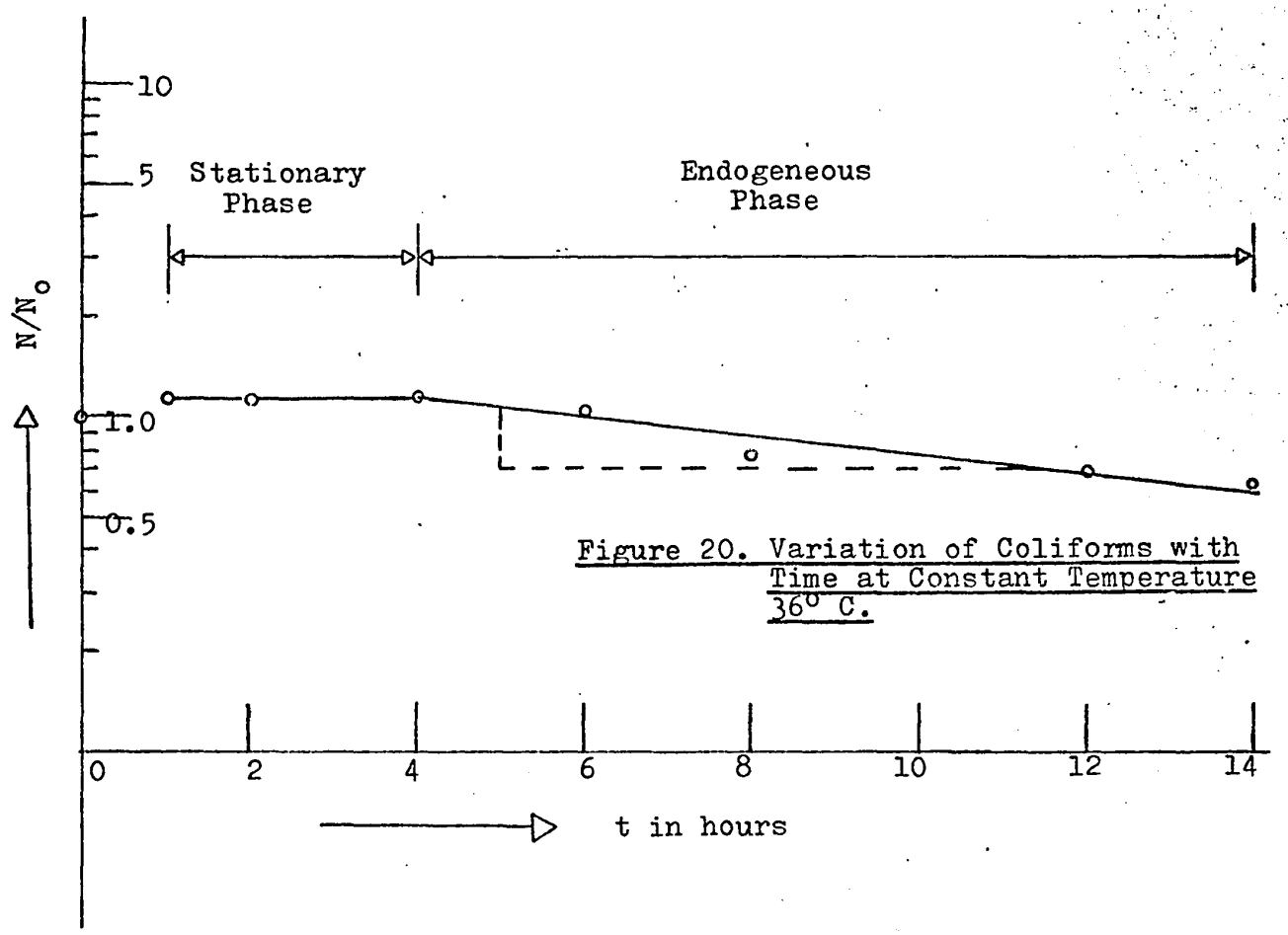

Figure 20. Variation of Coliforms with Time at Constant Temperature 36° C.

Figure 21. Total Coliforms vs. Temperature
Supply Source: St. Lawrence River
at Pointe-aux-Trembles

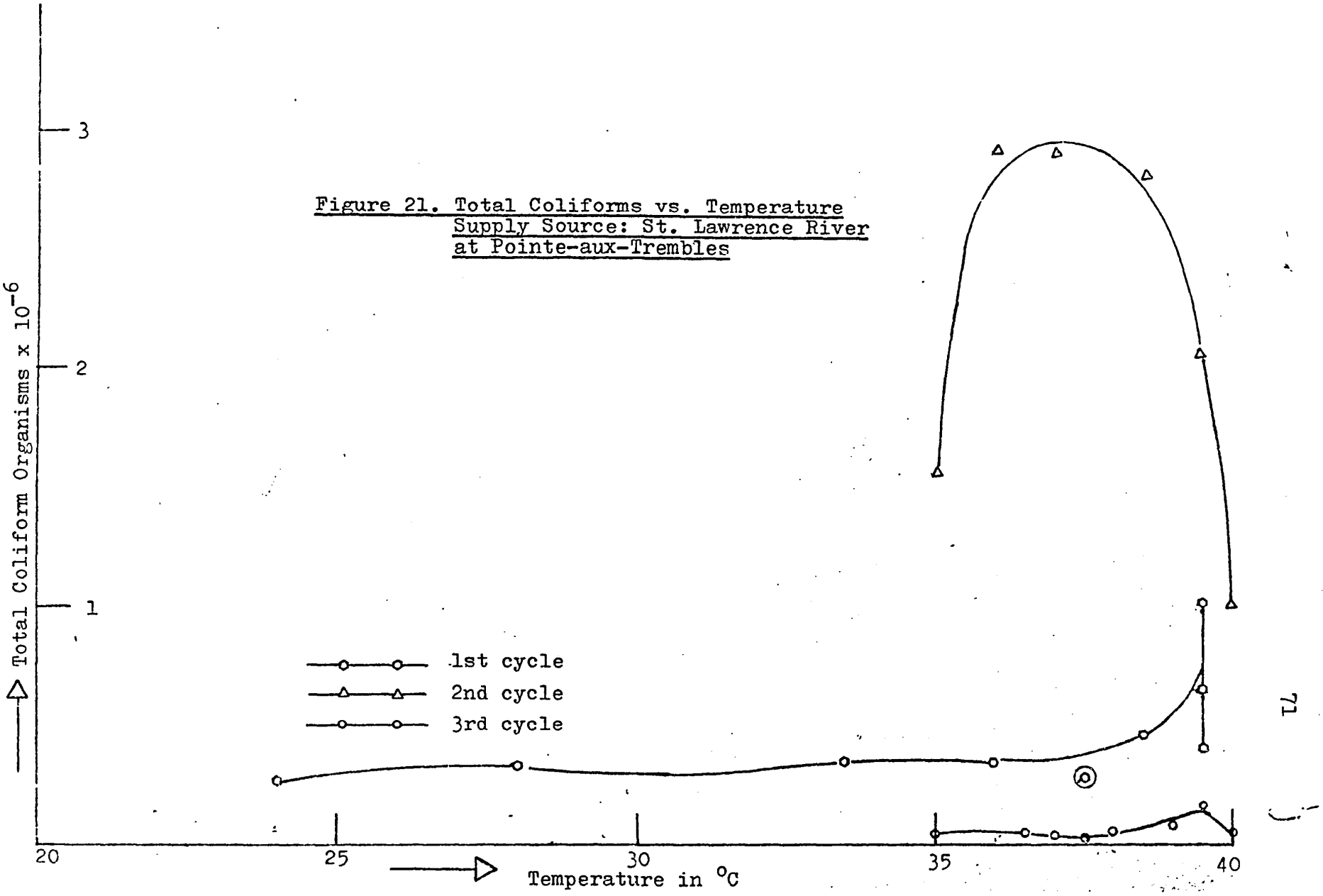

Table 19: Fecal Coliform Survival in Storage Tank.

Membrane Curing t^{o} - $80^{o}C$
Source of supply - St. Lawrence River at Pointe-aux-Trembles.

Duration hrs.	Raw Water Temperature ^{o}C	Fecal Coliforms Remaining x 10^{-3} per 100 ml
0	24	52.0
1		
2	28	87.5
3		
4	33.5	67.5
5		
6	36	65.0
7		
8	37.5	47.5
9		
10	38.5	32.5
11		
12	39	
13		
14	39.5	
15		
16	39.5	20.0
17		
18	39.5	23.0
19	39.5	10.0
20		
21	35	5.0
22	36	12.5
23		
24	38	120.0
25		
26	38.5	110.0
27		
28	39	77.5
29		
30	39.5	90.0
31		
32	40	25
33		
34	40	27.5
35		
36	40	100.0
37		
38	40	12.5
39		
40	40	5.0

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Table 19 cont.

Duration hrs.	Raw Water Temperature °C	Fecal Coliforms Remaining x 10^{-3} per 100 ml
41		
42	40	5.0
43	40	2.5
44		
45		
46	35	2.5
47	36.5	2.5
48	37	20
49		
50	38	15
51		
52	38.5	12.5
53		
54	39	2.5
55		
56	39.5	10.0
57		
58	39.5	10.0
59		

Table 20: Fecal Streptococci Survival in Storage Tank.

Membrane Curing t° - 80°C
Source of supply - St. Lawrence River at Pointe-aux-Trembles.

Duration hrs.	Raw Water Temperature °C	Fecal Streptococci Remaining per 100 ml
0	24	110
1		
2	28	165
3		
4	33.5	205
5		
6	36	280
7		
8	37.5	230
9		
10	38.5	85
11		
12	39	45
13		
14	39.5	175
15		
16	39.5	325
17		
18	39.5	270
19	39.5	200
20		
21	35	100
22	36	105
23		
24	38	150
25		
26	38.5	600
27		
28	39	185
29		
30	39.5	520
31		
32	40	300
33		
34	40	250
35		
36	40	195
37		
38	40	180
39		
40	40	25

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Table 20 cont.

Duration hrs.	Raw Water Temperature °C	Fecal Streptococci Remaining per 100 ml
41		
42	40	10
43	40	15
44		
45		
46	35	55
47	36.5	125
48	37	100
49		
50	38	30
51		
52	38.5	15
53		
54	39	15
55		
56	39.5	10
57		
58	39.5	5
59		

Table 21: Fecal Streptococci Survival in Storage Tank.

Membrane Curing t^{o} - $70^{o}C$
Source of supply - Richelieu River at the low lift pump of St. Hilaire Filtration Plant.

Duration hrs.	Raw Water Temperature °C	Fecal Streptococci Remaining per 100 ml
0	22.5	440
1	25	465
2	27	75
3		
4	30	60
5		
6	33	10
7		
8	34	-
9		
10	36	225
11		
12	36.5	400
13		
14	38	330
15		
16	38	255
17		
18	39	120
19		
20	39	175
21		
22	36	425
23	36	455
24	36	-
25		
26	36.5	2050
27		
28	36.5	375
29		
30	37	270
31		
32	37	220
33		
34	37	125
35		
36	37	110

Table 22. Ratios of Coliform Densities at a Constant Storage Tank Water Temperature.
Derivation from Table 18 by setting t=22 hrs.=0.

Time t hrs.	No. of Coliform Bacteria per 100 ml x 10^{-3}	N/No %
0	463	100
1	525	114
2	520	112
4	518	112
6	470	102
8	365	79
12	320	69
14	300	65

Table 23: Raw Water Bacterial Counts and Temperature.
Raw water from St. Lawrence River at Pointe-aux-Trembles.

Test Duration hrs.	Raw Water Temperature °C	Raw Water Bacterial Counts per 100 ml Total Coliforms x 10^{-3}	Fecal Coliforms x 10^{-3}	Fecal Streptococci
0	24.0	185	52	110
2	28.0	310	87.5	165
4	33.5	340	67.5	205
6	36.0	350	65	280
8	37.5	283	47.5	230
10	38.5	470	32.5	85
12	39.0			45
14	39.5			175
16	39.5	410	20	325
18	39.5	670	23	270
19	39.5	1,020	10	200
20	REST	-	-	-
21	35.0	1,600	5	100
22	36.0	2,930	12.5	105
24	38.0	2,930	120	150
26	38.5	2,853	110	600
28	39.0	1,130	77.5	185
30	39.5	2,200	90	520
32	40.0	1,020	25	300
34	40.0	1,000	27.5	250
36	40.0	420	100	195
38	40.0	370	12.5	180
40	40.0	185	5	25
42	40.0	55	5	10
43	40.0	68	2.5	15
44	REST	-	-	-
46	35.0	58	2.5	55
48	37.0	23	20	100
50	38.0	70	15	30
52	38.5	68	12.5	15
54	39.0	93	2.5	15
56	39.5	185	10	10
58	39.5	200	10	5

Table 24: Raw Water Bacterial Counts and Temperature.
Raw water from the low lift pump of St. Hilaire Filtration Plant on the Richelieu River.

Test Duration hrs.	Raw Water Temperature °C	Raw Water Bacterial Counts per 100 ml		
		Total Coliforms x 10^{-3}	Fecal Coliforms x 10^{-3}	Fecal Streptococci
0	21.5	130	25	2,500
1	24.0	160	35	2,650
2	26.0	230	35	2,300
4	29.0	200	25	1,400
6	31.5	200	15	1,450
8	33.5	190	10	1,250
10	35.0	130	5	950
12	36.0	100	5	900
14	37.0	-	5	620
16	37.5	120	15	295
18	38.0	140	10	220
20	38.5	165	15	140
22	35.0	65	15	155
23	36.0	145	5	130
24	36.0	150	5	125
26	37.0	170	5	50

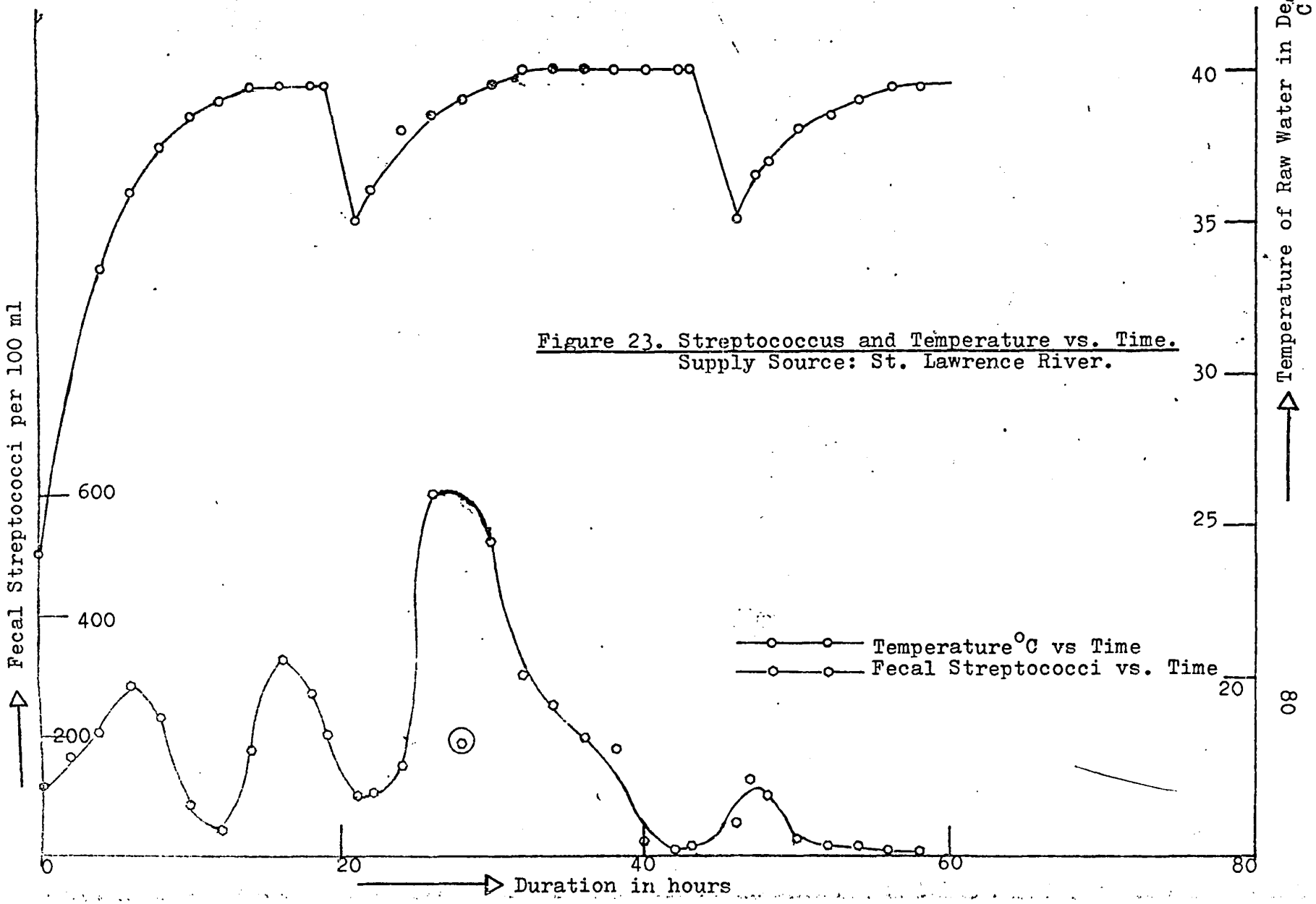
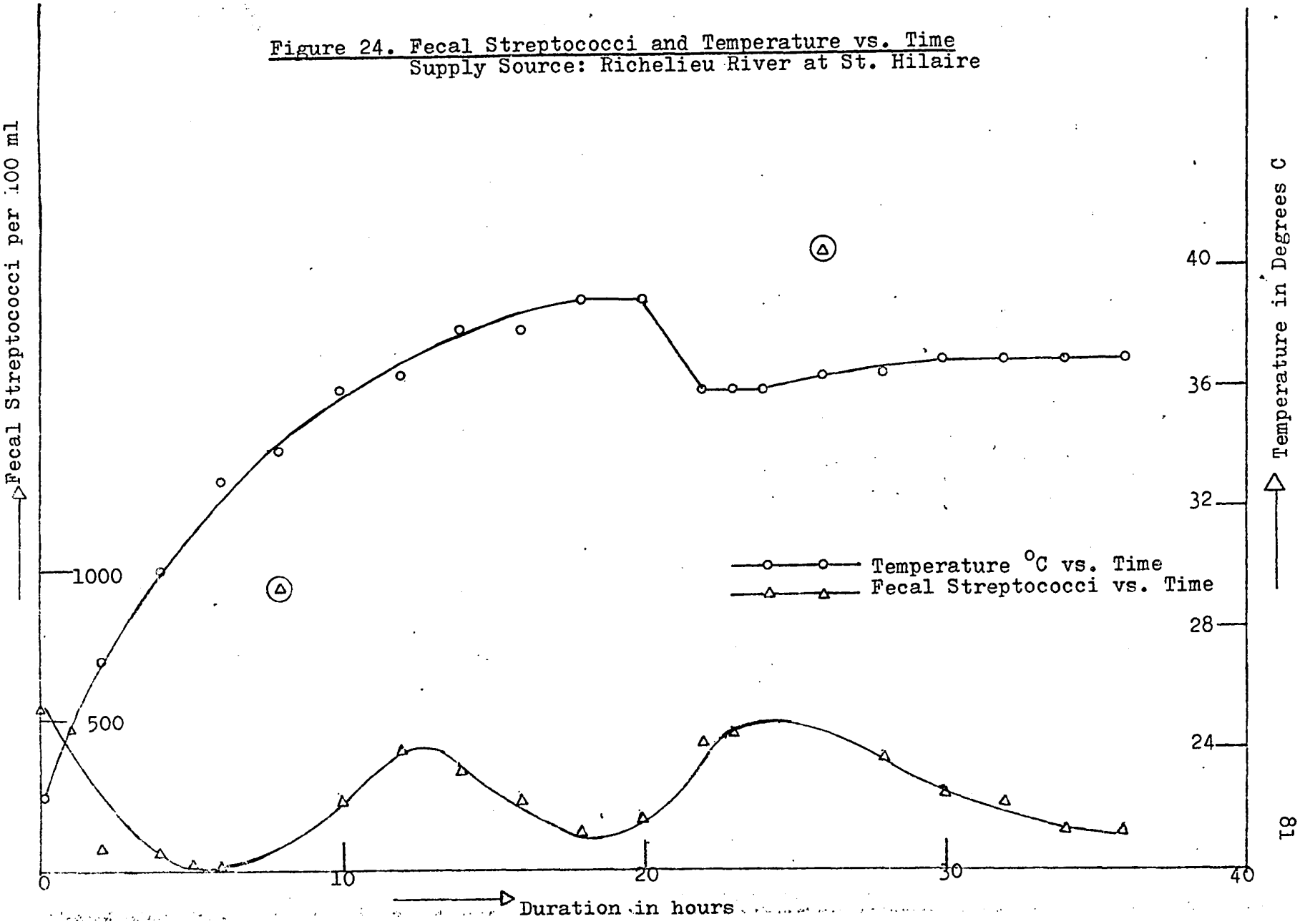


Figure 23. Streptococcus and Temperature vs. Time.
Supply Source: St. Lawrence River.

Figure 24. Fecal Streptococci and Temperature vs. Time
Supply Source: Richelieu River at St. Hilaire



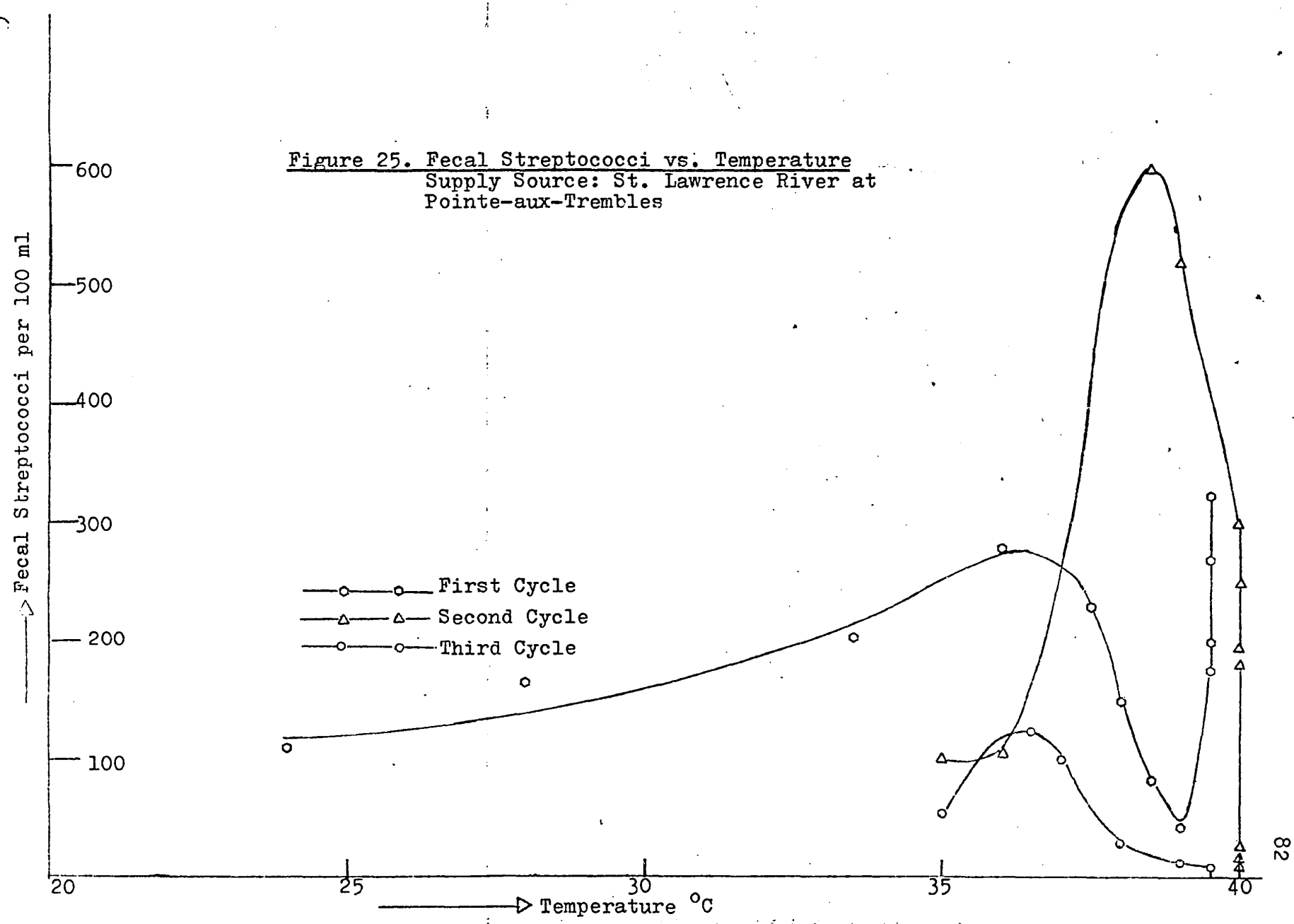


Figure 25. Fecal Streptococci vs. Temperature
Supply Source: St. Lawrence River at Pointe-aux-Trembles

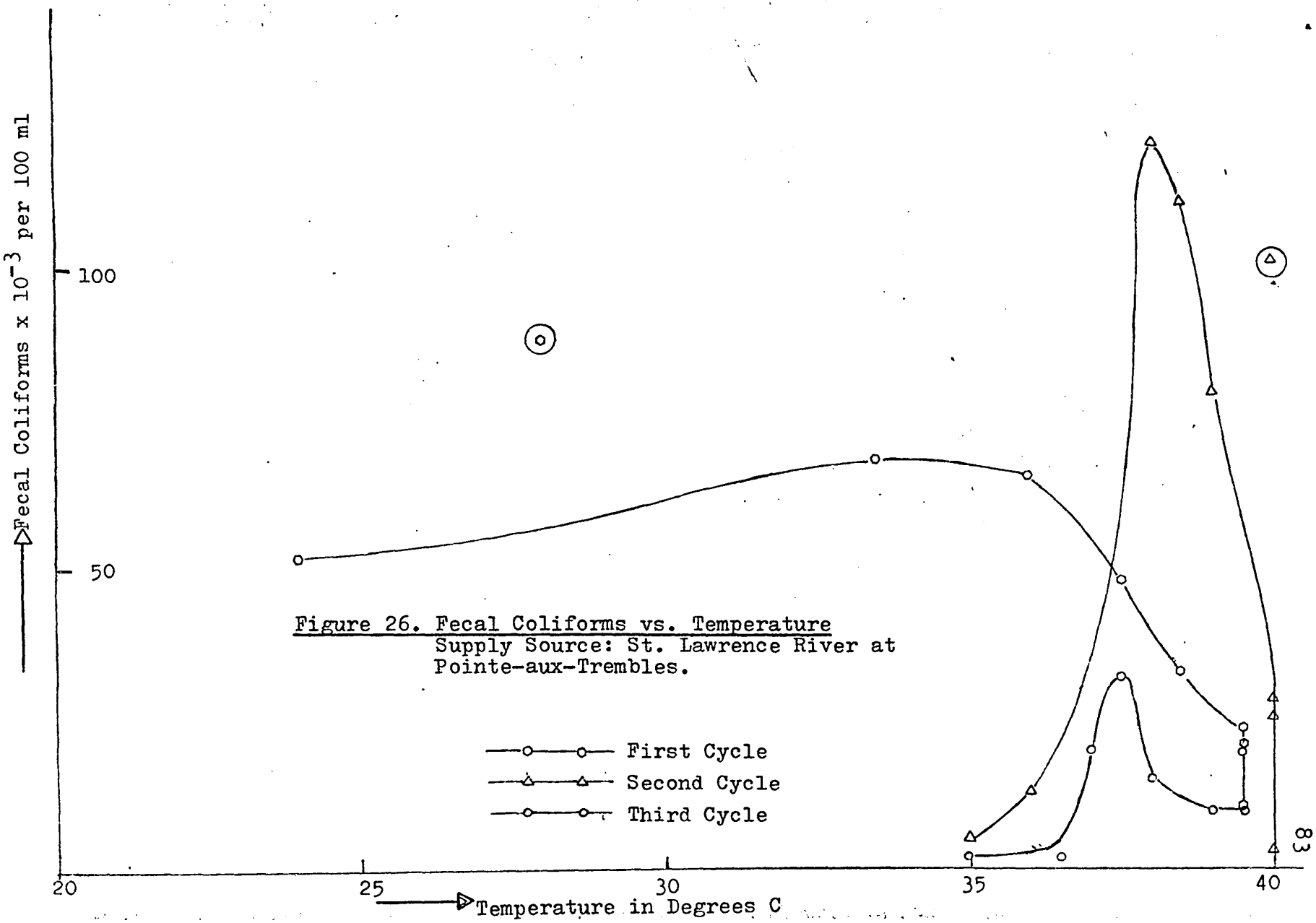


Figure 26. Fecal Coliforms vs. Temperature
Supply Source: St. Lawrence River at Pointe-aux-Trembles.

C. Removal of Bacteria

In this investigation, 100 ml of product water was filtered through the membrane filter and incubated for the total coliform count. Filters which did not show any colonies after incubation were designated as having counts "less than one" (<1) in the results. Filters that showed more than 300 colonies were designated as having counts "too numerous to count" (TNTC). Filters where the colonies overlapped in such a way that individual colonies became indistinguishable were designated as "confluent."

Results of the tests of six-hour duration are shown in Tables 26 to 33. The temperature of membrane curing is indicated in each case, and was 50°C, 60°C, 70°C, or 80°C.

Series of extended runs lasting from 36 to 60 hours were made. Samples of product water were taken at 2-hour intervals, filtered, and incubated for coliform organism count. It was intended to conduct all the extended runs at membrane curing temperature of 70°C but as the removals for 70°C membranes were established, changes were indicated, and further investigations were made using 80°C membranes. Table 34 shows the duration of test, flux, and the total coliforms in the product water in one of the runs which lasted sixty hours. Table 35 has been prepared from Table 34 to obtain graphic representation of the results, and shown in Figure 28. In Table 36, the raw water coliform counts for the same run are matched with those of the product water.

Table 37 shows the flux and coliforms of the product

water from a membrane cured at 70°C. Attempts were made to represent this graphically in Figure 29. The total coliforms in the raw water were matched with those of the product water, as given in Table 28, and shown in Figure 30.

Table 39 shows complete removal of fecal streptococci in a membrane cured at 70°C. The raw water counts of fecal streptococci and those from the two experimental cells are shown in this table as well as the respective flux for the cells. Random samples were also tested with 100 ml product water in other runs to confirm this observation.

Table 41 shows the results in an extended run which had to be terminated as the results of the incubated samples from the previous day became known. These cells were also cured at 70°C and the test lasted for two days.

In the tests shown in Table 42, two samples from each of the cells were filtered through the MF filter for total coliform count. They contained 100 ml and 1 ml of the product water. This made it possible to obtain counts from samples where 100 ml would give colonies too numerous to count. Thus it was possible to estimate the efficiency of coliform removal in the two cells during one of the extended runs as shown in Table 43.

Raw water samples for the extended runs were collected from two points for this investigation. Samples from the Richelieu River were taken from the low lift pumps of the filtration plant in St. Hilaire, Quebec. This plant prechlorinates its water in dosages mentioned earlier, but this level of pre-

chlorination leaves no measurable residue in the samples when they are delivered to the laboratory. The other samples for the extended runs were collected from the St. Lawrence River at a jetty in Pointe-aux-Trembles, Quebec. These sources of water are indicated in the headings of the various tables.

The following photographs are included in the Appendix:

i. Figure 31 shows the contrast between coliform organisms in 1 ml of raw water sample and 100 ml of product water.

ii. Figure 32 shows some samples of the product water and the nature of growth on coliform Endo media.

iii. Figures 33 and 34 show clumps of micro-organisms on top of a membrane magnified using an electron microscope.

iv. Figures 35 and 36 show sections through two membranes cast at the temperatures indicated. One of the membranes is freshly cast, the other has been stressed at the test pressure of 900 psi for a period of over 24 hours during one of the extended runs.

Table 26: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=50^{o}$
Supply Source: St. Lawrence River at Pointe-aux-Trembles. Not chlorinated.

Test Duration hrs.	Raw Water Total Coliforms per 100 ml x 10^{-3}	Product Water Total Coliforms per 100 ml Cell D	Cell E	Cell F
2	420	< 1	< 1	5
4	420	< 1	< 1	1·0
6	420	< 1	< 1	11

Table 27: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=60^{o}$
Supply Source: St. Lawrence River at Pointe-aux-Trembles. Not chlorinated.

Test Duration hrs.	Raw Water Total Coliforms per 100 ml x 10^{-3}	Product Water Total Coliforms per 100 ml Cell A	Cell B	Cell C
2	420	< 1	< 1	< 1
4	420	< 1	< 1	< 1
6	420	< 1	< 1	< 1

Table 28: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=50^{o}$,
Supply Source: Ottawa River at Hawksbury. Not chlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml $\times 10^{-3}$	Product Water Coliforms per 100 ml Cell A	Cell B	Cell C
2	12	< 1	9	14
4	12	< 1	<1	9
6	12	< 1	<1	50

Table 29: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=60^{o}$
Supply Source: Ottawa River at Hawksbury. Not chlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml $\times 10^{-3}$	Product Water Coliforms per 100 ml Cell D	Cell E	Cell F
2	12	120	< 1	< 1
4	12	160	< 1	< 1
6	12	1100	< 1	<1

Table 30: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=70^{o}$
Supply Source: St. Lawrence River at Pointe-aux-Trembles. Not chlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml $\times 10^{-3}$	Product Water Total Coliforms per 100 ml Cell A	Cell B	Cell C
2	600	< 1	16	< 1
4	600	< 1	33	< 1
6	600	< 1	< 1	< 1

Table 31: Total Coliforms of Product Water.
Results of 3 cells,
Membrane curing $t^{o}=80^{o}$
Supply Source: St. Lawrence River at Pointe-aux-Trembles. Not chlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml $\times 10^{-3}$	Product Water Coliforms per 100 ml Cell D	Cell E	Cell F
2	600	< 1	< 1	< 1
4	600	< 1	< 1	< 1
6	600	1.0	< 1	< 1

Table 32: Total Coliforms of Product Water.
Results of 2 cells,
Membrane curing $t^{o}=60^{o}$
Supply Source: Richelieu River at St. Hilaire.
Prechlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml Cell A	Cell B
1	30	2	2
2	37	1.0	22
3	66	< 1	2
4 1/2	84	7	1.0
5 2/3	TNTC	32	4

Table 33: Total Coliforms of Product Water.
Results of 2 cells,
Membrane curing $t^{o}=70^{o}$
Supply Source: St. Lawrence River at Pointe-aux-Trembles.

Test Duration hrs.	Raw Water Coliforms per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml Cell A	Cell B
3/4	240	< 1	< 1
1 3/4	260	< 1	< 1
2 3/4	205	< 1	< 1
3 1/4	200	< 1	2
4 1/4	170	< 1	< 1
5 1/4	165	< 1	< 1
6	140	< 1	< 1

Table 34. Test Duration, Flux, and Coliform Density/100 ml.Flux.
Membrane curing t^{o}=80°C
Raw Water Source: St. Lawrence River at Pointe-aux-Trembles.
Unchlorinated.

Test Duration hrs.	Flux Igpd/ft^2	Total Coliforms in 100 ml of Product Water
1	84	-
2	79	<1
3	76	-
4	74	<1
5	72	-
6	70	<1
7	68	-
8	67	2
9	64	-
10	62	2
11	61	-
12	59	10
13	57	-
14	55	12
15	55	-
16	52	14
17	50	-
18	50	35
19	44	-
20	REST	-
21	REST	-
22	52	180
23	48	-
24	46	TNTC
25	45	-
26	44	3
27	41	-
28	43	3
29	43	-
30	45	1
31	41	-
32	40	2
33	40	-
34	39	<1
35	39	-
36	38	1
37	41	-
38	36	TNTC
39	38	-
40	36	200
41	38	-
42	36	<1
43	37	-

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Table 34 cont.

Test Duration hrs.	Flux Igpd/ft^2	Total Coliforms in 100 ml of Product Water
44	REST	-
45	REST	-
46	REST	-
47	43	<1
48	42	15
49	41	-
50	40	3
51	40	-
52	40	100
53	40	-
54	40	11
55	41	-
56	40	1
57	39	-
58	38	<1
59	39	-

Table 35: Test Duration, Flux, and Total Coliform/100 ml Flux.

Test Duration hrs.	Flux Igpd/sq.ft.	Total Coliform* in 100 ml of Product Water
2	79	0
4	74	0
6	70	0
8	67	10
10	62	10
12	59	10
14	55	100
16	52	100
18	50	100
22	52	200
24	46	>300
26	44	10
28	43	10
30	45	10
32	40	10
34	39	0
36	38	10
38	36	>300
40	36	200
42	36	0
47	43	0
48	42	100
50	40	10
52	40	100
54	40	100
56	40	10
58	38	0

*Obtained from Table 34 by setting
(a) Counts less than 1 = 0
(b) Counts 1 to 10 = 10
(c) Counts 11 to 100 = 100
(d) Counts 101 to 200 = 200
(e) Counts 201 to 300 = 300
(f) Counts too numerous to count = > 300.

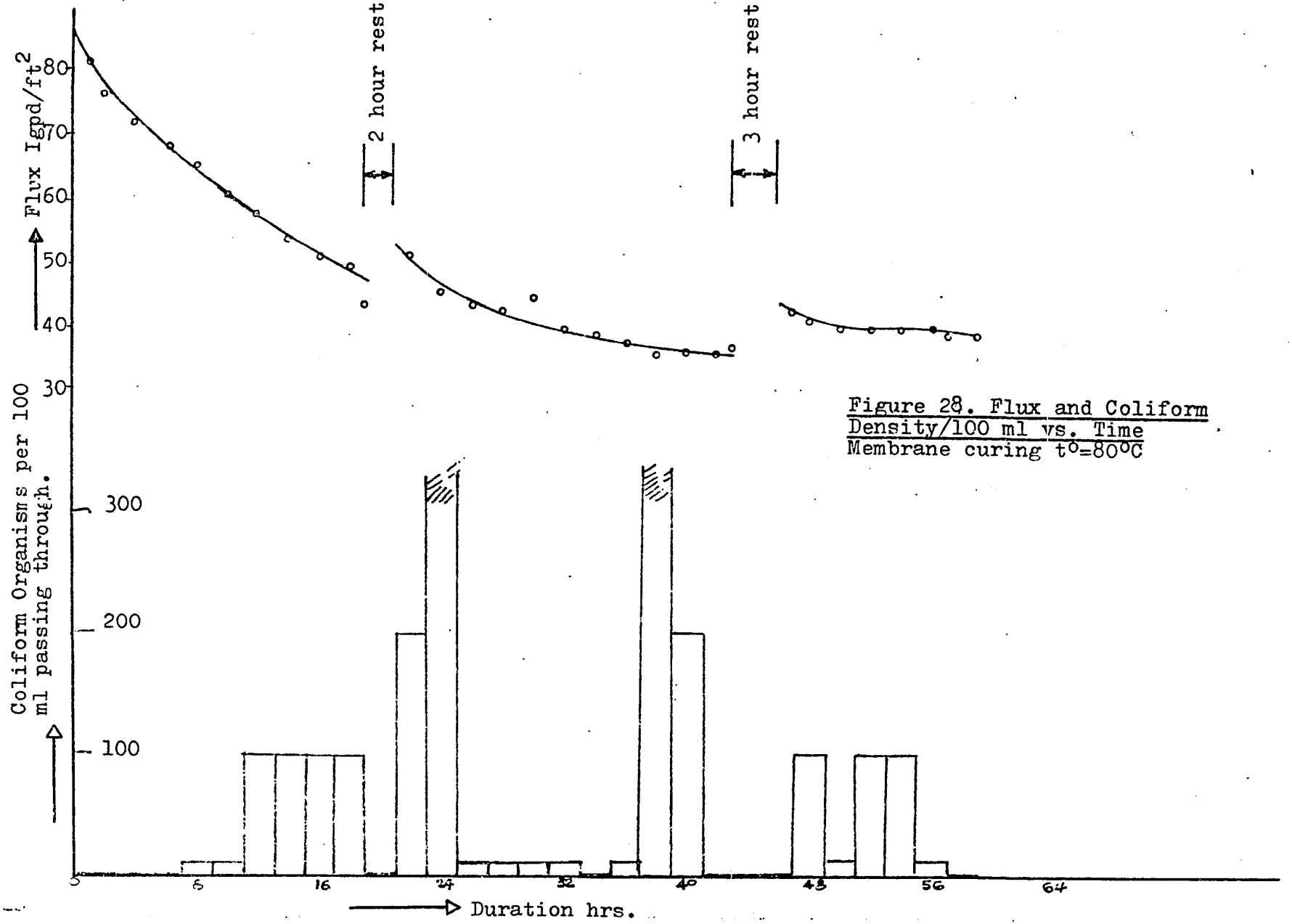

Figure 28. Flux and Coliform Density/100 ml vs. Time
Membrane curing t°=80°C

Table 36: Raw Water and Product Water Coliform Counts.
Membrane curing t^{o}=80°C.
Raw Water Source: St. Lawrence River at Pointe-aux-Trembles. No prechlorination.

Duration hrs.	Raw Water Coliforms per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml
1		
2	310	< 1
3		
4	340	< 1
5		
6	350	< 1
7		
8	283	2
9		
10	470	2
11		
12	-	10
13		
14	-	12
15		
16	410	14
17		
18	670	35
19	1,020	
20		
21		
22	2,930	180
23		
24	2,930	> 300
25		
26	2,850	3
27		
28	1,130	3
29		
30	2,200	1
31		
32	1,020	2
33		
34	1,000	< 1
35		
36	420	1
37		
38	370	> 300
39		
40	185	< 1

(cont.next page)

Table 36 cont.

Duration hrs.	Raw Water Coliforms per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml
41		
42	55	
43	68	
44		
45		
46	58	
47	58	<1
48	23	15
49		
50	70	3
51		
52	68	100
53		
54	93	11
55		
56	185	1
57		
58	200	<1
59		

Table 37: Test Duration, Flux, and Coliform Organisms Passing Through the Reverse Osmosis Membrane.
Membrane curing $t^{o}=70^{o}C$
Raw Water Source: Richelieu River at St. Hilaire.
Prechlorinated.

Duration hrs.	Flux Igpd/ft^2	Coliforms passing through per 100 ml of Product Water
1	81	
2	83	< 1
3	81	
4	77	1
5	74	
6	72	39
7	69	
8	68	50
9	66	
10	64	40
11	61	
12	60	24
13	56	
14	59	22
15	60	
16	52	150
17	56	
18	54	200
19	52	
20	51	250
21		
22		
23	63	>300
24	61	>300
25	58	
26	57	>300
27	57	
28	56	>300
29	55	
30	53	>300
31	53	
32	53	>300
33	52	
34	50	>300
35	50	
36	50	20,000

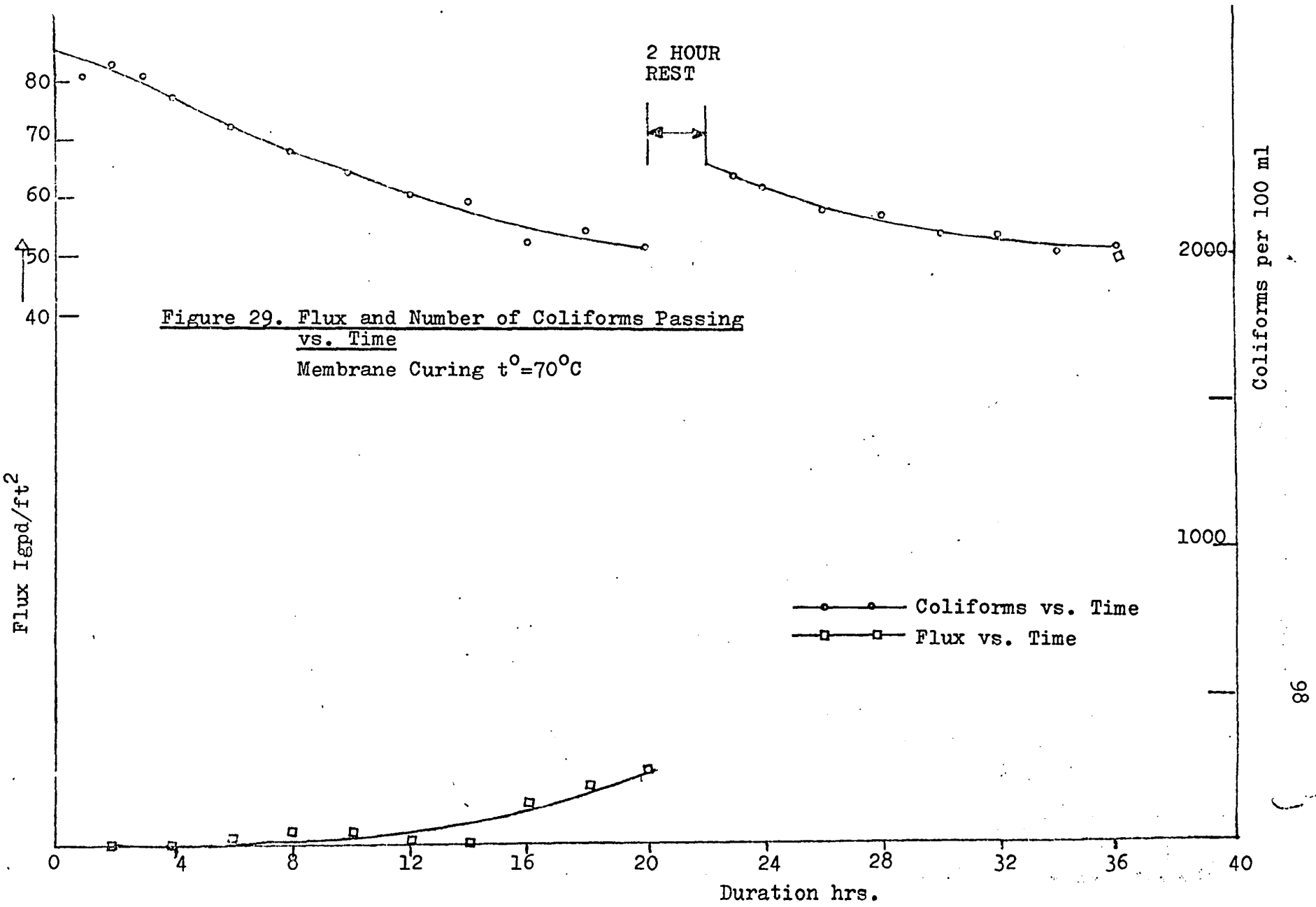

Figure 29. Flux and Number of Coliforms Passing vs. Time

Membrane Curing $t^{o}=70^{o}C$

Table 38: Raw Water and Product Water Coliform Counts.

Membrane curing $t^{o}=70^{o}C$
Source of Supply: Richelieu River at St. Hilaire.
Prechlorinated.

Test Duration hrs.	Raw Water Coliforms per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml
1	38	
2	53	< 1
3		
4	38	1
5		
6	63	39
7		
8	223	50
9		
10	358	40
11		
12	385	24
13		
14	373	22
15		
16	380	150
17		
18	435	200
19		
20	438	250
21		
22	463	
23	525	>300
24	520	>300
25		
26	518	>300
27		
28	470	>300
29		
30	365	>300
31		
32		>300
33		
34	320	>300
35		
36	300	20,000

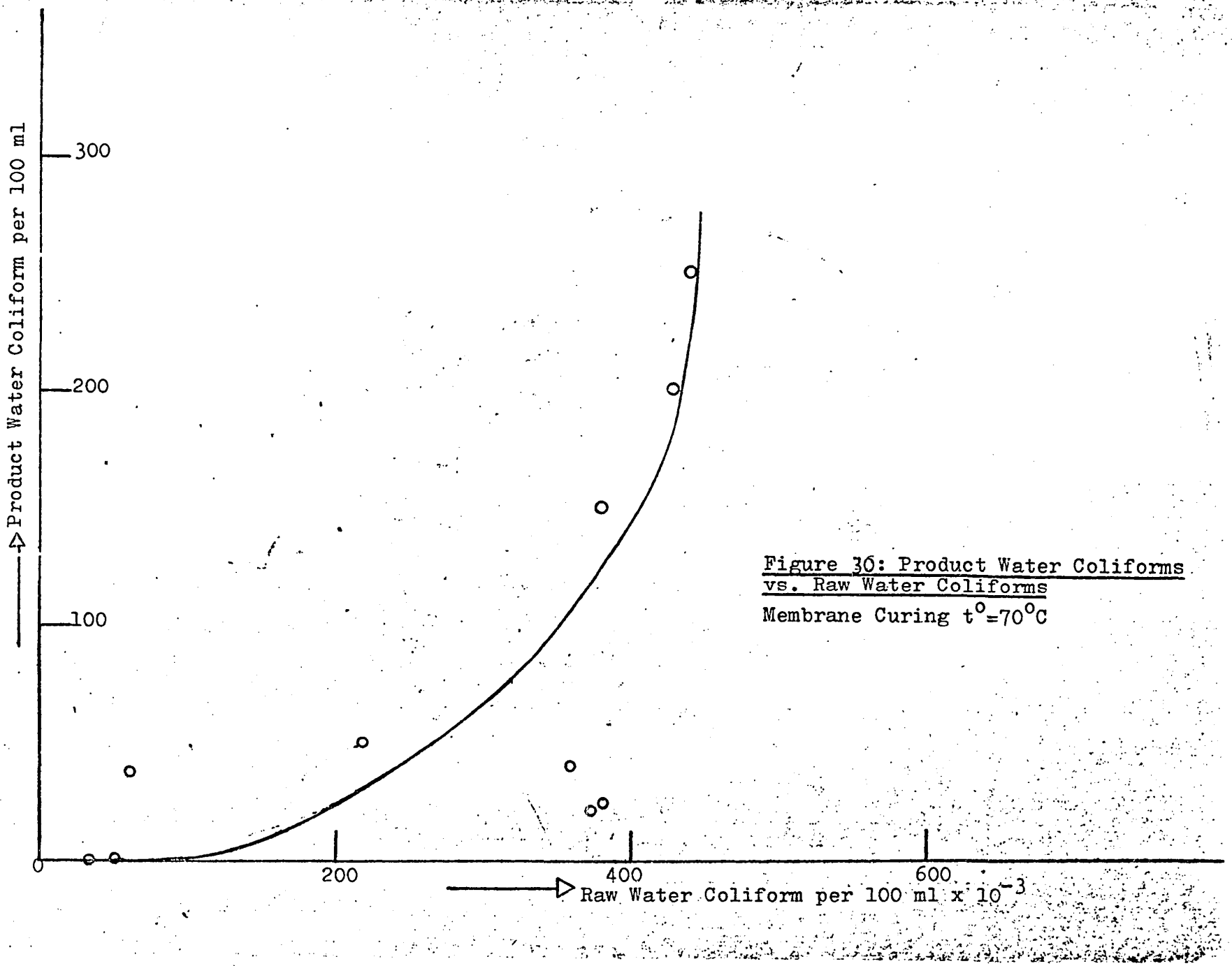

Figure 30: Product Water Coliforms vs. Raw Water Coliforms
Membrane Curing $t^{o}=70^{o}C$

Table 39: Removal of Fecal Streptococci.

Membrane Curing $t^{o}=70^{o}C$
Supply source: Richelieu River at St. Hilaire.

Flux in gpd/ft^2		Fecal Streptococci Density per 100 ml		
		Raw	Product Water	
Cell A	Cell B	Water	Cell A	Cell B
81	100	270	0	0
75	89	68	0	0
69	81	35	0	0
66	76	490	0	0
61	72	595	0	0
59	61	1120	0	0
58	59	1210	0	0
55	55	320	0	0
50	51	110	0	0

Table 40: Duration, Flux, Raw and Product Water Total Coliforms.

Membrane Curing $t^{o}=70^{o}C$
Source of supply: Richelieu River at St. Hilaire.
Prechlorinated.

Test Duration hrs.	Flux Igpd/ft^2 Cell A	Cell B	Raw Water Coliform Counts per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml Cell A	Cell B
2	83	109	53	>300*	0
4	77	93	38	>300	1
6	72	85	63	>300	39
8	68	80	223	>300	50
10	64	74	358	Confluent	40
12	60	70	385	"	24
14	60	71	373	"	22
16	52	60	380	"	150
18	46	56	435	"	200
20	51	60	438	"	250
22	REST		463	"	>300
23	62	64	525	"	>300
24	61	62	520	"	>300
26	58	59	518	"	>300
28	57	57	470	"	>300
30	53	54	365	"	>300
32	53	53		"	>300
34	50	51	320	"	>300
36	50	52	300	32,000**	20,000

* Over 300 colonies in 100 ml

** Dilutions of 0.1, 0.01 were filtered out to obtain actual counts.

Table 41: Flux, Duration, and Total Coliform Counts in Product Water.

Membrane Curing $t^{o}=70^{o}C$
Supply Source: Richelieu River at St. Hilaire.
Prechlorinated.

Test Duration hrs.	Flux Igpd/ft^2 Cell A	Cell B	Product Water Coliform Counts per 100 ml Cell A	Cell B
2	100	110	10	5
4	94	96	50	6
6	83	85	180	21
8	77	77	29	23
10	72	72	52	24
12	67	66	65	63
14	62	62	31 *	350
16	58		>3,000*	320
18	REST			
20	75	75	180	>3,000
22	71	70	180	>3,000
24	70	70	>3,000	>3,000
26	65	64	>3,000	>3,000
28	63	66	Confluent	>3,000
30	62	63	>3,000	>3,000
32	60	61	>3,000	Confluent
34	61	60	320	340
36	58	56	170	340
38	54	52	150	340
40	52	50	200	320
42	REST			
44	63	60	>3,000	>3,000
46**	58	56	>3,000	>3,000
47	57	54	>3,000	>3,000

* Over 300 colonies in 10 ml.

** Test was stopped in view of persistent results from the previous day's incubation.

Table 42: Duration, Flux, Raw and Product Water Coliforms.

Membrane curing $t^{o}=80^{o}C$
Source of water: Richelieu River at St. Hilaire.
Prechlorinated.

Test Duration hrs.	Flux Igpd/ft^2		Raw Water Coliform Counts per 100 ml x 10^{-3}	Product Water Coliforms per 100 ml	
	Cell A	Cell B		Cell A	Cell B
1	31	33	160	4,500	1,800
2	31	33	230	1,400	5,000
3	32	33	-	-	-
4	32	35	200	5,000	450
5	33	35	-	-	-
6	32	35	200	2,000	1,100
7	33	36	-	-	-
8	32	36	190	700	100
9	32	36	-	-	-
10	32	36	130	5,000	250
11	33	36	-	-	-
12	33	36	100	400	1,900
13	33	36	-	-	-
14	34	37	-	-	-
15	34	36	-	-	-
16	34	36	120	>30,000**	10,000
17	34	36	-	-	-
18	33	35	160	>30,000	20,000
19	33	35	-	-	-
20	35	37	165	>30,000	>30,000
21	REST				
22	REST				
23	32	35	145	>30,000	>30,000
24	31	34	150	>30,000	>30,000
25	32***	35	-	-	-
26	29	33	170	200	10,000
27	27	27			

** Over 300 colonies in 1 ml.

*** Line pressure started to fall below 900 psi due to leakage of gas. Test was discontinued.

Table 43: Percentage of Coliform Removal.
Raw Water Prechlorinated.
Membrane curing $t^{o}=80^{o}C$
Supply Source: Richelieu River at St. Hilaire.

Test Duration hrs.	Flux Igpd/ft^2		Raw Water Total Coliform per 100 ml x 10^{-3}	Percentage Coliform Removal	
	Cell A	Cell B		Cell A	Cell B
1	31	33	160	97.17	98.88
2	31	33	230	99.39	97.83
4	32	35	200	97.50	99.77
6	32	35	200	99.00	99.44
8	32	36	190	99.63	99.95
10	32	36	130	96.16	99.81
12	33	36	100	99.60	98.10
16	34	36	120	75.00	91.67
18	33	35	160	80.00	87.50
20	35	37	165	80.00	80.00
23	32	35	145	79.00	79.00
24	31	34	150	80.00	80.00
26	29	33	170	98.82	94.14

CHAPTER V

DISCUSSIONS AND CONCLUSIONS

A. DISCUSSIONS

A-1. Membrane Characteristics, Raw Water, and Flux.

Flux obtained in this investigation agreed very closely with that obtained by Trivedi (11) who developed this particular type of membrane. The results of all tests showed that the flux and test duration exhibited an approximately linear relationship when plotted using semi-logarithmic scales. Empirically these results can be expressed as:

$$F(t) = Q_o e^{-\alpha t}$$

where F(t) is the flux at time t hours expressed in gallons per unit time, and

Q_o and α are constants.

The constant Q_o for this composition of membrane is the intercept of the line on the t=0 line, and is a function of the membrane curing temperature, the pressure of the system, and general membrane casting technique. The most important of these is the rolling process of the casting mass on the casting plate, and the viscosity of the mass. As can be seen from Figure 14, Q_o for two membranes cast at 80°C were 90 gpd/sq.ft. and 31 gpd/sq.ft. for results numbered 3 and 4, respectively, even though the raw water samples came from a common source.

Results numbered 1 and 2 of the same Figure with membranes cured at 70°C were 90 gpd/sq.ft. and 110 gpd/sq.ft., respectively. The viscosity of the liquid membrane material depends on the effect of ambient temperatures on the volatile components of the casting medium. The effects of this viscosity variation on the flux require further investigations. Mechanizing the casting procedure will also help to ensure a reliable prediction of the maximum flux Q_o for membranes cured at any temperature. The constant α is the slope of the line. Experimental results shown on Figures 13 and 14 seem to indicate that α depends on the nature of the raw water. Loeb and Manjikian (45) have also observed that the deposition of fouling organic materials from the raw water on the membrane surface is a major factor affecting its performance. An important observation made from the results of the extended runs with intermittent rest periods is that in each case the slope of the line α changed after every interruption. This change in α was such that each line segment, when extrapolated, passed through the Q_o point for the particular test. This fact is illustrated more clearly in Figure 15 which is reproduced from test result No. 3 of Figure 14. An advantage of this is that it can be used to predict the flux and its subsequent rate of variation in a reverse osmosis system following a rest in operation as a result of a breakdown in the system or a cooling period for the mechanical parts.

A-2. Coliform Organisms and Temperature

In all the tests conducted, it was observed that the density of the coliform organisms in the tank increased with temperature. The increase reached a peak during and immediately after the rest periods in the extended runs as shown in Figure 18. This observation is similar to those of Velz (46) who demonstrated the effect of temperature on the seasonal variation of fecal coliforms. Figure 21, showing the variation of the coliforms with temperature in the three cycles of the extended run, is in agreement with the observations of Rahn as shown by Lemanna and Mallette (65). The influence of temperature on fecal coliforms as plotted in Figure 26 is in agreement with the results of Barber for E.coli as shown by Stainer et al. (18). In the total coliform variations of Figure 21, the peak in the first cycle is not distinct due to the presence of the various strains of coliform organism. The peak of the second cycle occurs over a wider range of temperature for the same reason.

The effect of prechlorination of raw water on growth rate with increasing temperature is illustrated in Figure 16, and in the first cycle of the curve of Figure 19. As mentioned earlier, the prechlorination level of 0.5 to 1.0 mg/l left no chlorine residues two hours later when the tests were started. The coliform density was observed to decrease as the initial raw water temperature increased. This drop may be attributed to the following:

(a) A breakdown of the clumps and clusters of the

micro-organisms by the chlorine which was not sufficient to kill those organisms that were embedded within the cluster. Chang (47) has given a comprehensive account of the effects of this clumping phenomenon. Chlorine also breaks up organic materials which release organisms which were occluded in the organic matter in heavily polluted waters. These organisms require time for adjustment to a new and perhaps more favourable environment.

(b) The remaining groups of bacteria which were not killed by the level of chlorine applied must be very resistant, but as the temperature gradually rises, they either die off or take time to adjust to normal growth.

The behaviour of the coliform organisms at a relatively constant and high temperature has been represented in Figure 19 and partly in Figure 20. The temperature was maintained between 36°C and 37°C from the 22nd hour. This followed immediately after the rest period and lasted for 14 hours. Nutrients appeared to become the limiting factor for growth with a peak occurring two hours later. The graph of Figure 20 was plotted using the 22nd hour of the run as the zero hour. This semi-logarithmic plot produced two straight lines - a horizontal line indicating a stationary phase, followed by a declining growth or endogeneous growth phase. Velz (46) has shown that the survival of pathogens and nonpathogens of special interest in stream sanitation approximates Chick's law:

$$\log\left(\frac{N}{N_o}\right) = -kt$$

where N is the number of the organisms remaining in the water at time t, and N_o is the number of the organisms at t=0; k is a constant.

This observation gives a value of k=0.055 per hour at 36°C which agrees closely with a value of 0.044 per hour obtained by Frost and Strater for a large stream as noted by Fair et al. (20). These artificial variations in temperature have been achieved successfully due to extended periods of pumping and steady recirculation. In the river, no such changes would take place. Even the changes from winter to summer conditions are much more gradual, giving the organisms time to adjust.

A-3. Fecal Organisms and Temperature

Fecal coliforms and fecal streptococci are examples of mesophilic bacteria. As their natural habitat is the intestinal tract of warm-blooded animals, they usually grow most rapidly at temperatures between 35°C and 40°C.

In this investigation the temperature optima for fecal streptococci in the raw, unchlorinated water were observed four times in the three cycles of one extended run. For the first cycle, the two optima were 36.3° C and 39.5° C. In the second cycle, which showed a much more rapid rate of bacterial increase, a higher peak, and a narrow range for the optimum temperature, the optimum was observed at 38.5°C. The optimum for the final cycle was observed at 36.5° C. These are illustrated in Figures 23 and 25. Temperature optima for fecal streptococci compare very closely with the four optima which were observed for fecal coliforms in the same run as shown on Figure 26. The latter were observed at temperatures of 34.5° C in the first cycle, 38° C in the second cycle, and

37.5 and 39.5° C in the third cycle. In general, these observations have illustrated the fact that the optimal growth temperature for any bacteria outside its natural habitat occurs at or close to the temperature of the natural environment which supports its normal life.

With samples of chlorinated water, the growth pattern of fecal streptococci with increasing temperature was similar to those observed for total coliforms mentioned earlier. The period of adjustment to increasing temperatures is marked by a fall in the density of the organisms, as illustrated in Figure 24. After this lag, the growth reverts to the pattern of the unchlorinated sample with two temperature optima at 37.2° C and 36° C.

With the temperature of raw water maintained almost constant for a period of 14 hours, the fecal streptococci density showed a steady decrease. Unlike the observations with the coliforms where growth continued for about two hours in the steady temperature state, the fecal streptococci density dropped immediately. This observation is in agreement with the fact that fecal streptococci do not increase in number in streams where temperatures are relatively constant and generally below those of their natural environment.

A-4. Removal of Bacteria

Lonsdale (57), by making some basic assumptions about the phenomenon of reverse osmosis, developed expressions governing the transportation of both solvent and solute across a semi-permeable membrane. Some of his assumptions include:

(a) The validity of Henry's Law applied to solvents and solutions in a reverse osmosis process.

(b) The irreversibility of flow across a reverse osmosis membrane.

(c) The independence of permeability and effective membrane thickness of applied pressure.

(d) The chemical potential of each component is continuous across the membrane-solution interface.

The membrane must be semi-permeable so that the concentration of the solute on the low pressure side of the membrane will be approximately zero in contrast to the solute concentration at the high pressure side of the membrane. To maintain flow of water across the various layers of the membrane, it is necessary that the chemical potential of water change continuously through the layers it passes through.

Sourirajan (58) conducted a number of experiments to determine the semi-permeability of cellulose acetate to various aqueous solutes. He showed that the ability of cellulose acetate membranes to reject ions in electrolytes decreased with their order in the lyotropic series where citrates and sulphates are the most highly rejected while the iodides and thiocyanates are the least rejected. This shows that ions with the higher valences are most readily rejected. He observed some trend of rejection with nonelectrolytic solutes where larger solutes like sucrose, dextrose, and sorbitol were well rejected in contrast with monohydroxy alcohols up to propyl alcohol which passed rather readily. Blunk (59) observed that solute species which are larger than glucose molecules with a

radius of about 3.6 Å will be at least partially rejected by a cellulose acetate membrane fabricated for the desalination of seawater. Sourirajan (58) confirmed this observation with a complete removal of sucrose with molecules of a radius of 4.4 Å, and predicted that other nonelectrolytes of larger sizes would be rejected regardless of their chemical nature.

These various works have been developed to account for the removal of materials of molecular and ionic sizes and concentrations. Observations reported by some investigators on the behaviour of bacteria in reverse osmosis process, however, using cellulose acetate membranes, have created doubts concerning the efficiency of this process in removal of micro-organisms from water. In his investigation, using heavily polluted river waters, Allick (12) observed that one coliform bacterium per 100 ml passed through the membranes yielding a flux of 26 gpd/sq.ft. The raw water coliform density was of the order of 4,800 per 100 ml. In his report on reverse osmosis pilot plant studies with the Potomac River water, Sieveka (61) attributed the coliform counts in the first samples taken to apparent contamination by residual materials in the system. All the other product waters gave average coliform bacteria less than 3 per 100 ml with General Atomic high selective type A membranes. The raw water coliform bacteria count was of the order of 1500 to 10,000 per 100 ml.

In this investigation some of the osmosis cells reported in Tables 26 to 33 gave completely sterile water with zero bacterial counts for the total coliforms, fecal coliforms, fecal streptococci, and standard plate counts, each tested with

100 ml of product water. This confirms the observations of many investigators whose membranes have produced waters which meet and even tend to exceed Drinking Water Standards. The bulk of the results of the extended runs in this investigation, however, produced very erratic results concerning the removal of coliform bacteria.

Matz (62) has observed the formation of large cellular cavities in cellulose acetate membranes. These cavities have been thought to be gas or aqueous occlusions entrained in the process of casting the membrane. He showed that the bubbles are equeous intrusions which develop during the gelling stages of membrane formation. It has been suggested by Saltonstall (64) and other investigators that the presence of these aqueous intrusions can be modified or even inhibited by the use of additives in the casting solutions or gelling medium, or control of the conditions of gelation. The formation of these intrusion cells appears to be related to the hydrodynamic instabilities connected with changes in the surface tension and viscosity of the casting mass.

In this investigation, the C-5 membrane developed by Trivedi (11) has been tested for the removal of bacteria. The emphasis in the development of this membrane has been on the amount of flux which the membrane could produce with raw water from surface sources with present and, possibly, future levels of industrial and domestic pollution. No additives have been used in the casting and the flux has been relatively high, exceeding the flux of conventional slow sand filters. The membrane, when inspected with an electron microscope, seems

to have an asymmetric structure in which a very thin dense layer overlies a porous bottom layer. Figures 35 and 36 are sections through two of the membranes. Riley and his co-workers (48) have shown that the dense layer is about 0.2 to 2.5 μ thick. The mean pore size of the supporting structure is of the order of 100 to 3000 Å. The C-5 membrane is initially rolled to a thickness of 0.021" (approximately 500 μ). Gelation may result in a shrinkage of the film by about 50%, giving a final membrane thickness of about 250 μ.

In a similar cellulose acetate membrane, Matz (49) observed high porous structures on the freshly cast membranes which still maintained normal flux and salt rejection characteristics. This porous structure is said to give rise to abnormal cavities with characteristic conical shapes originating from the dense surface of the membrane. He further observed that these membranes often developed pinholes when pressurized. Formation of the pinholes or bubbles, as they are called, is not uncommon in commercial membranes. In the manufacture of the C-5 membranes used in this investigation, a few membranes were discarded when these pinholes were visible.

Flow across a membrane through the pinholes contaminates the product water. Pinholes which are created by pressure can be so tiny that they may not affect the flux volume very appreciably. This result cannot be negligible when considering the effect of the breakthrough on the bacterial contamination of the product water. For instance, a change in flux volume of 0.1% resulting from this source will show a volume increase of 0.5 ml/hr on a membrane producing 96 Igpd/sq.ft. This

volume difference cannot be observed in the equipments used to collect the product water in this investigation. For a raw water with a coliform density of 5×10^5 per 100 ml, the above contamination will give rise to a product water coliform count of 500 per 100 ml. This observation points to the following phenomenon as responsible for the erratic coliform counts in the product water:

(a) Formation in the membrane of weak points which break on pressure. These weak points are not ordinarily visible except by means of powerful microscopes. When the weak points finally break under pressure, the pinholes may not produce a significant variation in the flux volume.

(b) The density of bacteria in the raw water. For raw water with low bacterial densities, there is a probability that small infiltration across the membrane through a pinhole will not yield a significant bacterial count in the product water. This is especially true where small volumes of the product water are tested for the bacteria. In this way, it will seem as if the number of organisms appearing in the product water is proportional to the number of the organisms in the raw water. This observation is illustrated in Figure 25.

It is possible that as the membrane becomes clogged by fouling materials, these pinholes may become plugged. This

will account for the fluctuation in the coliform counts observed throughout the extended runs. The low density of fecal streptococci accounts for its apparent complete removal observed throughout the investigation.

Adequate dosage of chlorine added to the raw water can reduce the density of bacteria in surface waters to a level which will minimize the effect of these pinhole infiltrations into the product water. Dosages of chlorine at such a level may leave chlorine residures in contact with the membrane. It is not yet investigated whether a prolonged exposure of cellulose acetate membranes to free chlorine residuals at concentrations up to 2 mg/l will cause deterioration of the membranes. A laboratory experience at Pomona (63) using 2 mg/l combined chlorine residuals produced satisfactory results for a period of over six months.

On the basis of observations of membrane performance it may be postulated that an adequate chlorination of the product water is imperative.

B. Conclusions

This investigation has provided data and observations concerning the behaviour of bacteria of sanitary significance in a closed recirculation system with variable temperatures. It has further demonstrated the effects of the high levels of bacterial contaminations of supply sources on public water supply and the quality of finished waters, and the need for disinfecting treated surface waters in order to comply with Drinking Water Standards.

On the basis of experimental work and theoretical studies the following may be concluded:

1. Reverse osmosis method of water treatment may produce sterile product water if the quality of semi-permeable membranes is uniformly defectless.

2. The performance of C-5 membrane employed in these investigations, while providing high flux values, has shown considerable variations of its properties.

3. The experimental work has shown that fecal streptococci and fecal coliforms have consistently been removed.

4. The removal of total coliforms varied within wide limits depending on bacterial density in the raw water and uniformity of the membrane structure.

5. The experimentally derived casting technique of the membrane resulted in pinhole formation leading to an expressed flux contamination.

6. The observed quality of product water has clearly indicated that its disinfection is imperative.

7. In order to obtain membranes with uniformly satisfactory characteristics it is recommended that the membrane casting technique should be modified, fully standardized, and mechanized.

8. Further investigations of the composition of this type of membrane and the technique for casting it may improve its structure, minimize or even inhibit the formation of pinhole cavities, and still retain its present flux volume.

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APPENDIX

A. Tables of media for bacteriological analysis.

Table 5 (Ref. 22; Section 404C, No. 6)

Plate Count Agar medium for total bacteria (adapted)

Peptone-trypton	5.0 gm
Yeast extract	2.5 gm
Glucose	1.0 gm
Agar	15.0 gm
Distilled Water	1 litre

pH should be 7.0 $\pm$ 0.1 after sterilization.

Table 6 (Ref. 22; Section 404C, No. 13)

Endo medium for Total Coliform Count.

Tryptone or polypeptone	10.0 gm
Thiopeptone or thiotone	5.0 gm
Casitone or trypticase	5.0 gm
Yeast extract	1.5 gm
Lactose	12.5 gm
Sodium chloride	5.0 gm
Dipotassium hydrogen phosphate K_2HPO_4	4.375 gm
Potassium dihydrogen phosphate KH_2PO_4	1.375 gm
Sodium lauryl sulphate	0.050 gm
Sodium desoxycholate	0.10 gm
Sodium sulphite	2.10 gm
Basic fuchsia	1.05 gm.
Distilled water	1 litre.

Final pH should be between 7.1 and 7.3. Medium stored in the dark at 2 to 10° C. Unused medium discarded after 96 hours.

Table 7 (Ref. 22; Section 404C, No. 12)

M-FC Broth for fecal coliform.

Tryptose or biosate	10.0 gm
Peptcose peptone No. 3 or polypeptone	5.0 gm
Yeast extract	3.0 gm
Sodium chloride	5.0 gm
Lactose	12.5 gm
Bile Salts No. 3 or bile salt mixture	1.5 gm
Aniline blue	0.1 gm
Distilled water	1 litre

Dehydrated medium to be rehydrated in distilled water containing 10 ml of 1% rosolic acid in 0.2 N NaOH.

Final pH should be 7.2.

Finished medium should be stored at 2 to 10° C and unused medium discarded after 96 hours.

Table 8 (Ref. 22: Section 404, No. 18)

M-Enterococcus Agar for fecal streptococcus.

Tryptose	20.0 gm
Yeast extract	5.0 gm
Glucose	2.0 gm
Dipotassium hydrogen phosphate K_2HPO_4	4.0 gm
Sodium azide	0.4 gm
Agar	10.0 gm
2,3,5-triphenyltetrazolium chloride	0.1 gm
Distilled water	1 litre

After sterilizing by boiling, final pH should be 7.2. Poured plates may be stored in the dark up to 30 days at 2 to 10° C.

APPENDIX B

Results of Raw River Water Samples tested in the Laboratory.

Table 9

Raw Water Samples. Results from Ottawa River. June 1972.

i.Total Bacteria:

2 Nos 10^{-3} ml : Confluent.
2 Nos 10^{-4} ml : Confluent. } ⇒ > 3 x 10^6/cc.

ii. Total Coliforms

Raw Water ml	Count	Average	No per 100 ml	Special Colonies	Remarks
0.1	12	13	13,000	1. FC	1000 FC/100 ml.
0.1	14			< 1. FC	
0.01	1	1.5	15,000	< 1	
0.01	2			< 1 *	
Average			14,000		1,000

ii. Fecal Coliform

1.0	30	22	2,200		
1.0	14				
0.1	2	1.5	1,500		
0.1	1				
Average			1,800		

iv. Fecal Streptococcus

1.0	2	1.5	150		
1.0	1				
0.1	< 1				
0.1	< 1				
			150		

Summary: Total bacteria over 3 million per cc.
Total Coliform 14,000/100 ml
Fecal Coliform 1,800/100 ml
Fecal Streptococcus 150/100 ml.

Table 10

Raw Water Samples. Results from Richelieu River. May 1972.

i. Total Bacteria:
2 Nos 10^{-3} ml : Confluent
2 Nos 10^{-4} ml : Confluent
$\Rightarrow$ $>3 \times 10^6$/cc.

ii. Total Coliform

Raw Water ml	Count	Average	No per 100 ml	Special Colonies	Remarks
0.1	90	100	100,000	26Fc	24,000Fc/100ml
0.1	110			22Fc	
0.01	6	7	70,000	2Fc	25,000Fc/100ml
0.01	8			3Fc	
Average			85,000		24,500Fc/100ml

iii. Fecal Coliform

1.0	TNTC	-		
1.0	TNTC			
0.1	40	33	33,000	
0.1	26			
Average			33,000	

iv. Fecal Streptococcus

1.0	24	20	2,000	
1.0	16			
0.1	3	2	2,000	
0.1	1			
Average			2,000	

Summary:
Total bacteria over 3 million per cc
Total Coliform 85,000/100 cc
Fecal Coliform 33,000/100 cc
Fecal Streptococcus 2,000/100 cc

Table 11

Raw Water Samples. Results from St. Lawrence River (Varennes). May 1972.

i. Total Bacteria: 10^{-3} ml : Confluent; 10^{-4} ml : Confluent } $> 3 \times 10^6$/cc

ii. Total Coliform

Raw Water ml	Count	Average	No per 100 ml	Special Colonies	Remarks
0.1	27	25	25,000	4Fc	3,500Fc/100ml
0.1	23			3Fc	
0.01	3	3	30,000	-	
0.01	3			-	
Average			27,500		3,500Fc/100ml

iii. Fecal Coliform

1.0	20	21	2,100		
1.0	22				
0.1	2	2	2,000		
0.1	< 1.0				
Average			2,050		

iv. Fecal Streptococcus

1.0	8	5	500		
1.0	2				
0.1	< 1.0	1	1,000		
0.1	1				
Average			750		

Summary: Total Bacteria over 3 million per cc.
Total Coliform 28,000/100 ml.
Fecal Coliform 2,000/100 ml.
Fecal Streptococcus 750/100 ml.

Table 12

Raw Water Samples. Results from St. Lawrence River (Pointe-aux-Trembles). June 1972

i. Total Bacteria:
2 Nos 10^{-3} ml : Confluent
2 Nos 10^{-4} ml : TNTC (Too numerous to count).

ii. Total Coliform

Raw Water ml	Count	Average	No per 100 ml	Special Colonies	Remarks
0.1	260	290	290,000	52Fc	55,000Fc/100ml
0.1	320			58Fc	
0.01	34	32	320,000	56Fc	50,000Fc/100ml
0.01	30			48Fc	
Average			300,000		

iii. Fecal Coliform

1.0	250	275	28,000		
1.0	300				
0.1	19	30	30,000		
0.1	51				
Average			29,000		

iv. Fecal Streptococcus

1.0	40	38	3,800		
1.0	36				
0.1	7	4	4,000		
0.1	1				
Average			3,900		

Summary: Total Bacteria over 3 million per cc.
Total Coliform 300,000/100 ml.
Fecal Coliform 29,000/100 ml.
Fecal Streptococcus 3,900/100 ml.

Table 13

Raw Water Samples. Results from St. Maurice River. June 1972.

i. Total Bacteria:
10^{-3} ml : TNTC (Too numerous to count)
10^{-4} ml: 240 colonies = 2.4×10^6/ml.

ii. Total Coliform

Raw Water ml	Count	Average	No per 100 ml	Special Colonies	Remarks
1.0	11	12	1,200	5Fc	500Fc/100 ml
1.0	13			5Fc	
0.1	1	1	1,000	1Fc	1,000Fc/100ml
0.1	<1.0			-	
Average			1,100		750

iii. Fecal Coliform

1.0	8	6	600		
1.0	4				
0.1	<1.0	1	1,000		
0.1	1				
Average			800		

iv. Fecal Streptococcus

1.0	1	1	100		
1.0	<1.0				
0.1	<1.0	-	-		
0.1	<1.0				
Average			100		

Summary:

Total Bacteria	2.4×10^6/ml
Total Coliform	1,100/100 ml
Fecal Coliform	800/100 ml
Fecal Streptococcus	100/100 ml.

Table 25: Typical Membrane Casting Records.

Date	6 Nos Membranes at 80°C				
	Operation	Time in	Duration mins.	Time out	Remarks
29/7/72	Stirring	15.12	15 ± 1	15.27	Casting solution
	Mixing	15.30	180 ± 5	18.30	
30/7/72	Evaporation	11.5900	5±0.5	12.0400	
	Cooling	12.04	60 ± 5	13.04	Membrane No. 1
	Curing	13.0530	5±0.5	13.1030	
	Evaporation	12.0600		12.1100	
	Cooling	12.11		13.11	No. 2
	Curing	13.1300		13.1800	
	Evaporation	12.1720		12.2220	
	Cooling	12.23		13.28	No. 3
	Curing	13.3000		13.3500	
	Evaporation	13.0700		13.1200	
	Cooling	13.12		14.12	No. 4
	Curing	14.1400		14.1900	
	Evaporation	13.1500		13.2000	
	Cooling	13.20		14.20	No. 5
	Curing	14.2130		14.2630	
	Evaporation	13.3020		13.3520	
	Cooling	13.35		14.35	No. 6
	Curing	14.3800		14.4300	

APPENDIX C

PHOTOGRAPHS

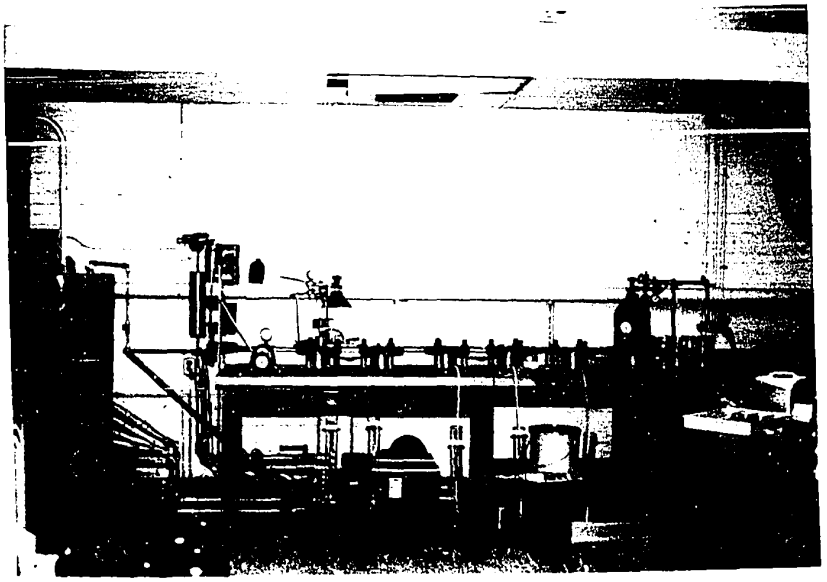

Figure 10: General Layout of Cells.

Figure 11: Membrane Casting Chamber.

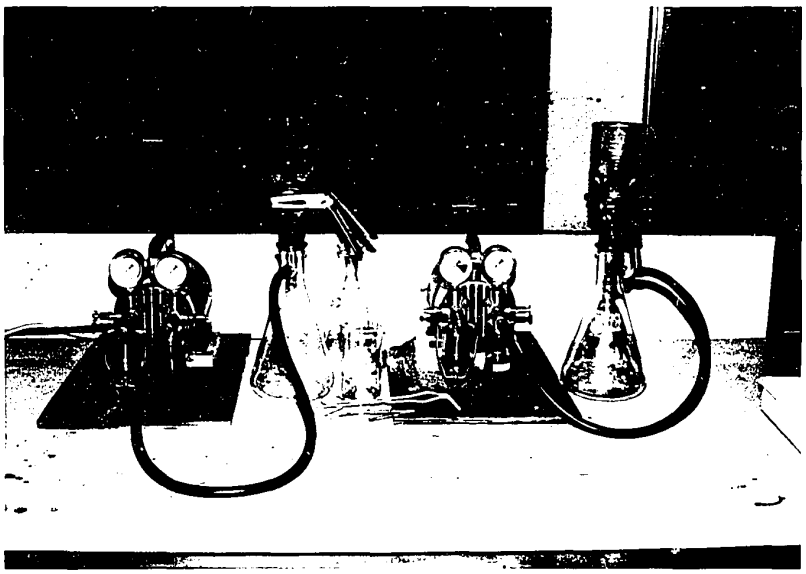

Figure 12: MF Vacuum Pump Assembly

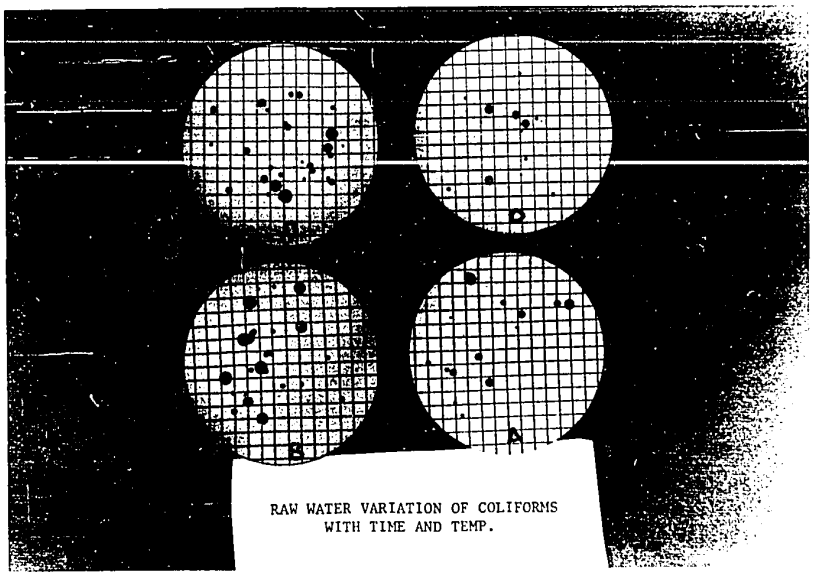

Figure 22.

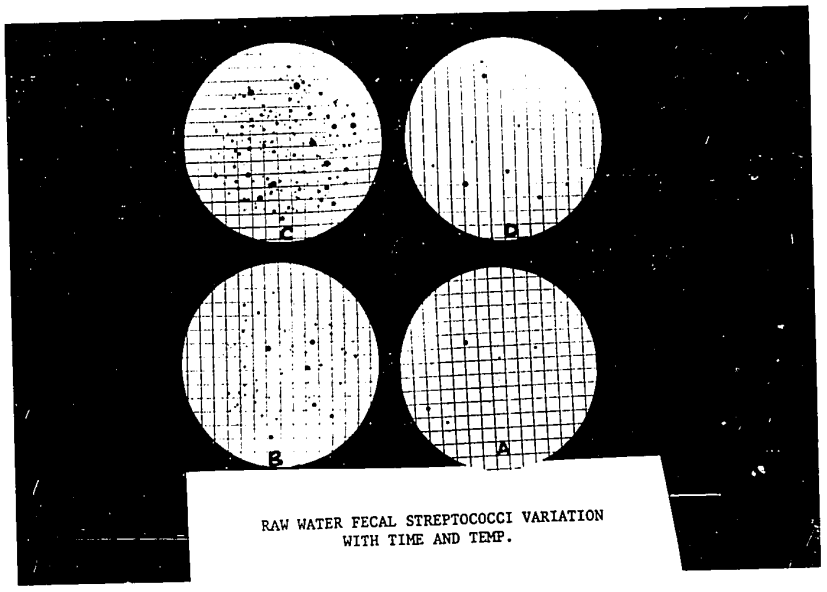

Figure 27.

	A	B	C	D
Time(hrs.)	0	6	12	18
Temp.°C	24	36	39	39.5

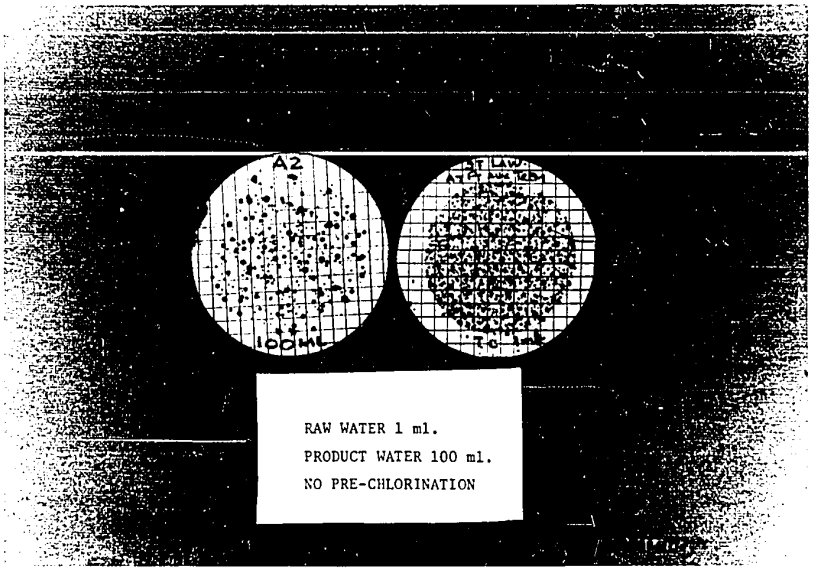

Figure 31.

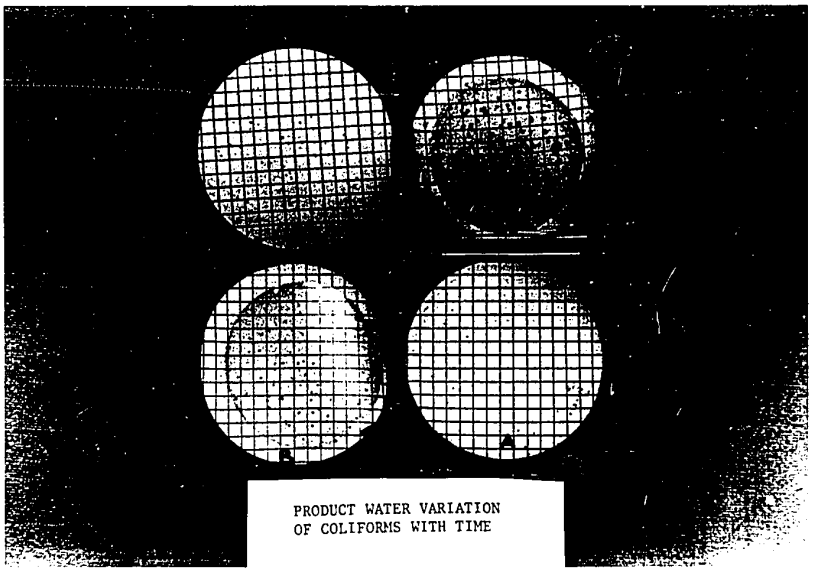

Figure 32.

	A	B	C	D
t (hrs)	2	6	12	18

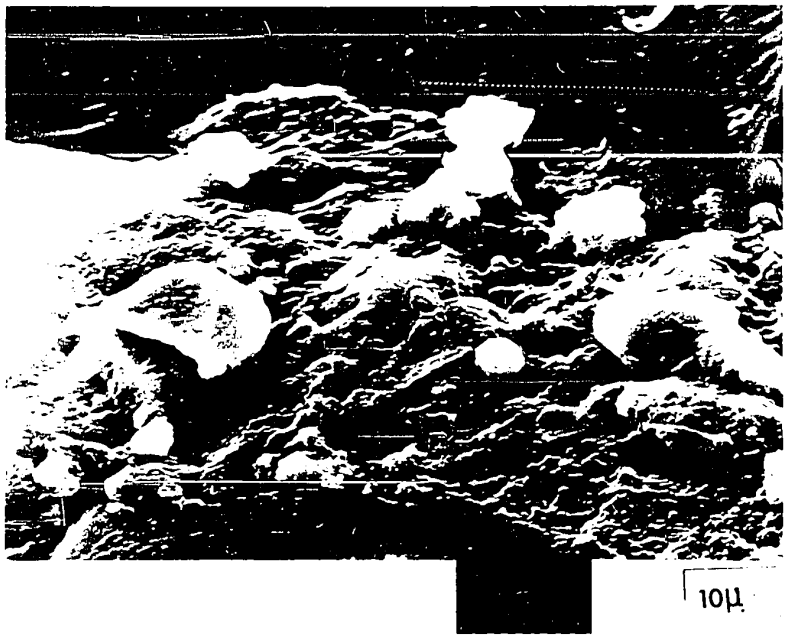

Figure 33: Clusters of organic, inorganic, and bacterial deposits on top of a reverse osmosis membrane (2000x Magnification)

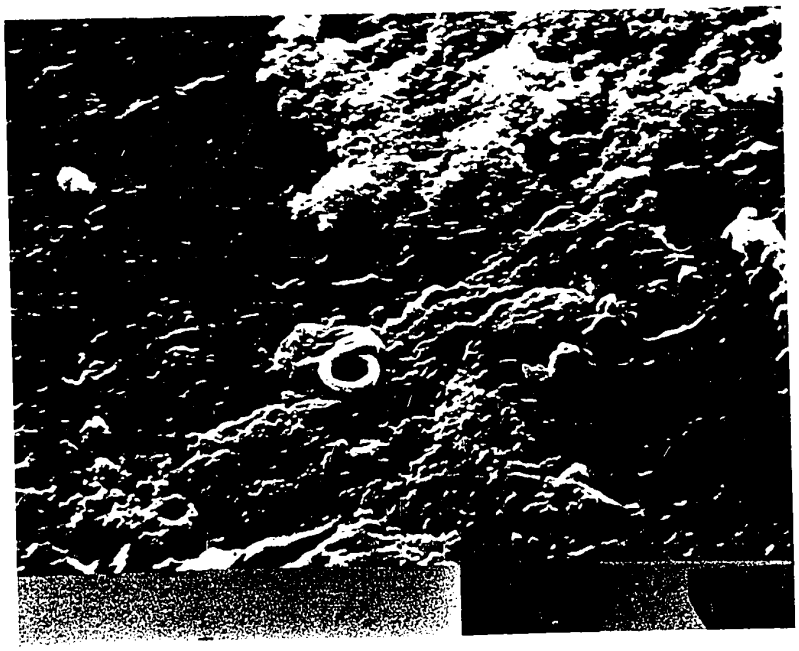

Figure 34: Surface deposit on membrane (5000x Magnification)

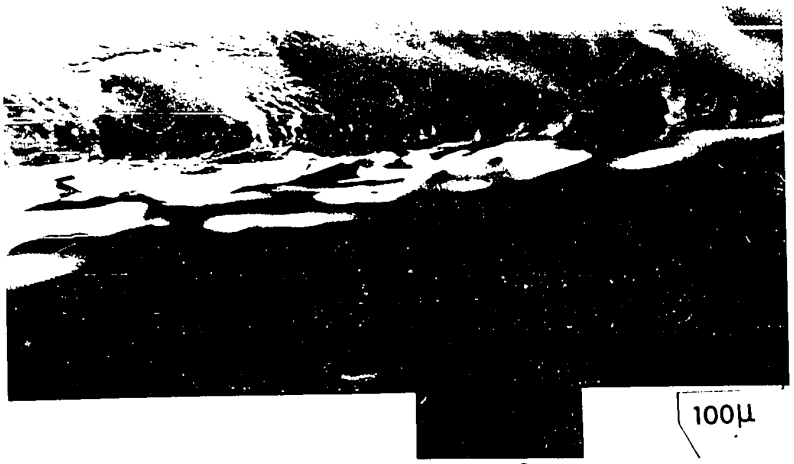

Figure 35: Edge view of an 80°C Membrane tested at 900 psi for 48 hours (200x Magnification)

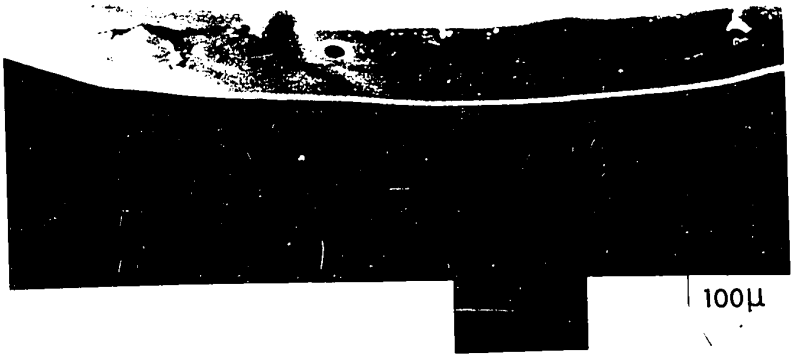

Figure 36: Edge view of a 70°C unused membrane tested at 900 psi for 48 hours (200x Magnification)