THE SCIENCE TEACHER - A SANITARIAN

A Thesis

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Master of Arts

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by

Oren J. Hill

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by

Oren J. Hill

APPROVED BY:

F. Ray Derrick

Chairman of Thesis Advisory Committee

Herbert Wey

Dean of Graduate School

A B Crews

Major Professor

I W. Carpenter Jr.

Minor Professor

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CHAPTER I

THE PROBLEM AND DEFINITIONS OF TERMS USED

Throughout the centuries, man has been gradually finding out how he can best adapt himself to the world's environment and to others in that environment in order to live long, happily, and well. And as soon as he has arrived at certain conclusions, these conclusions have been crystallized in the form of precepts or laws to be used as an epitome of past experiences for the guidance of the children of the family or the whole community group. In Egypt or wherever one looks in early history, he finds evidence of precepts or laws concerning such matters as health, the choice and preparation of foods, and the care of children. From the Hebrew race, posterity has the Mosaic Laws easily accessible in the book of Leviticus of the Bible; and though one finds throughout that the laws have more religious than sanitary significance; yet some of the laws as to personal cleanliness, the isolation of lepers, sex relations, and the care of pustular secretions are in a sense health precepts. In the writings of Hippocrates are many valuable precepts concerning the sanitation of air, water, soil, and occupation, and valuable observations on the seasonal incidence of certain diseases.

In the period of Greek and Roman ascendancy community health activities flourished. Health, cleanliness, beauty, and strength were qualities of life to be sought and worth community action to be achieved.

Man from the earliest known periods of history has recognized the fact that cleanliness is godliness. Water is a means to cleanliness. However, water is the universal solvent. Through the efforts of science teachers over the nation, the purity and quality of water can be determined for individuals as well as for communities.

I. THE PROBLEM

Statement of the problem. It was the purpose of this study (1) to enable science teachers to make preliminary determinations of the purity of water; (2) to describe the preliminary methods for testing water within the jurisdiction of the science teacher; and (3) to show how a kit may be constructed to serve the purpose for such duties.

Importance of the study. To determine the character and quality of water determines the utilization for which the water can be made. In the more remote sections of the country, there is the constant question, "Is this water safe to drink?" It is evident that State Departments of Public

Health supply such answers; but the lapse of time, in most cases, means a delay in the use of the vital water under examination. Utilization of the water determines what a person is interested in finding in the water. Some water may be used for washing only. Therefore, the user is interested in the hardness of the water. Some water may be used for drinking and cooking. Therefore, he is interested in the bacterial count of the water. Some water may be used for swimming and recreation. In a public pool one would be interested in the amount of residual chlorine that is present, and in the "ole swimmin hole" he would be interested in the bacteria present as well as the sedimentary materials. It is essential that his knowledge be gained before the water is used, in order that a more healthful condition may be maintained for society. In this study it has been the purpose to determine the purity of water for drinking and recreation or swimming.

Any time a new well is dug or a new spring is exposed to supply drinking water, the supply should be tested. This is a duty the community school science teacher can perform and thereby render greater service to the community.

In the trend toward outdoor education and recreation, the problem of a safe water supply becomes progressively greater. One finds that students learn best when they do

not realize they are learning. Children love the out-of-doors. They are fascinated with the sights, sounds, and follies of the out-of-doors. Therefore, the streams and water supplies should be kept safe for their enjoyment and study. The various school-camping programs, that are definitely on the increase, are a factor indicating the trend toward outdoor education. At these school camps the students have excellent opportunities to engage in easier learning and greater retention of facts.

The Scouting movement over the United States is increasing each year. It is bringing more boys and girls into a fuller realization and understanding of their surrounding. Their camping activities are numerous and varied. None of their camps should operate without a safe water supply.

There is a concentration of the population in the city or suburban areas at present. For the past quarter of a century, the population has been migrating from the rural areas. This migration has brought about a real problem of supplying these large cities with an adequate supply of safe water. The water supplies become contaminated because of the methods by which people dispose of their waste and their refuse.

A safe water supply is one of the best safeguards of the health of a community. A relatively small number

of diseases, however, are actually transmitted by impure water. All of these are diseases in which the infection atrium is the alimentary tract and in which the organisms leave the body with the excretions. The most important of these diseases are typhoid fever, amoebic dysentery, bacillary dysentery, paratyphoid, hemorrhagic jaundice, and Asiatic cholera.

A pure water supply, however, seems to affect the general health of a community and to decrease its death rate wholly apart from the decrease due to the elimination of these specific diseases mentioned. It has been noted, for example, that when a city changes from a contaminated to a pure supply, there is not only a marked decrease in the typhoid death rate, but there is also in some instances decreases noted in the number of deaths from tuberculosis, pneumonia, and other diseases. In practically every instance there is a decided decrease in the total death rate. This has been termed the Mills-Reinche phenomenon, in honor of the men who first called attention to these facts. This phenomenon must result from one of two causes. Either diseases not ordinarily regarded as being carried by water are in a certain percentage of cases so transmitted, or the use of an impure water supply so affects the average health of a community that the individuals become more readily

susceptible to other diseases than they otherwise would be. It is probable that the latter is the true explanation (Buchanan, 1940).

II. DEFINITIONS OF TERMS USED

Science teacher. Science teacher is interpreted as meaning all teachers holding a certificate to teach science. It is assumed that all certified science teachers have the educational background for the interpretation of the determinations described. However, all methods of testing have been over-simplified in order that this water testing kit may be adapted to all schools and especially those with very little equipment.

Sanitarian. A sanitarian is a person familiar with or engaged in sanitation work. Sanitation work deals with the prevention of diseases through the removal of dirt or filth (Thorndike, 1941). This report dealt with only a small phase of the work of a full-time sanitarian. This report implied sanitation through water supplies. The phase of work of a sanitarian which deals only with the detection of dirt or filth in sources of water shall be interpreted as the meaning of sanitarian.

Pure water. Pure water may be defined as water

which contains no disease-producing bacteria nor an excess of organic matter of any kind (Buchanan, 1940).

III. REVIEW OF THE LITERATURE

No easily accessible literature has been published linking the public school science teacher with the portion of a sanitarian's work dealing with water. However, much has been written in regards to the testing and determination of the quality of water.

I. Literature on the Detection of Minerals, Chlorine, and Bacteria in Water

Prescott, Winslow, and McCardy describe reliable methods of bacterial detection. Of course, their methods were limited to the public health phase of the subject, the distribution and behavior and significance of those organisms introduced into water from extraneous sources and having sanitary significance (Prescott, Winslow, and McCardy, 1947).

The chemical determinations necessary for the control and detection of chlorine and minerals in water and the analysis of polluted water were beneficially described by Theroux, Eldridge, and Mallman (1935).

The book, Standard Methods for the Examination of Water, Sewage, and Industrial Wastes, as prepared and published jointly by the American Public Health Association, The American Water Works Association, and The Federation of Sewage and Industrial Wastes Associations, was the final reference for all tests that were made in this study.

Other sources of value in this study were Fred Wilbur Tanner, Bacteriology; Charles P. Hoover, Water Supply and Treatment; P. L. Gainey, Microbiology of Water and Sewage for Engineering Students; Max Levine, Laboratory Technique in Bacteriology, Estelle D. Buchanan and Robert Earle Buchanan, Bacteriology; and Stanley E. Wedberg, Microbes and You.

II. Limitations of Previous Studies

Previous studies were limited because of the complexity of the procedures described and expensive equipment required. Procedures were difficult in progression and interpretation. This study is intended to simplify, as much as possible, the tests involved in water determinations.

This report is not intended to discover every known feasible method for testing water. Only those simplified methods deemed necessary for the science teacher to perform to determine the purity of water are

included.

III. Organization of Remainder of the Thesis

The remainder of the thesis is divided into three units of development:

1. The various determinations used for testing water. The preliminary tests that are necessary for the determination of the quality of water are described and elaborated upon. The purpose for such determinations is established.

2. The construction of a compact kit for testing water. A simple compact kit is described, with detailed instructions, as to its proper construction.

3. How the science teacher may use a kit for testing water and steps for the science teacher to take in order to use the kit to the best advantages and purposes are described.

CHAPTER II

STANDARD METHODS FOR DETERMINING THE QUALITY OF WATER

The methods set forth in the study for the determination of the purity of water for drinking and recreation are considered standard methods, approved jointly by The American Public Health Association, The American Water Works Association, and The Federation of Sewage and Industrial Wastes Associations. The purpose of these organizations has been to secure the adoption of more uniform and efficient methods of water analysis. The methods for analysis presented are believed to represent the best current practice of American Water Analysts and to be generally applicable in connection with the ordinary problems of water purification, sewage disposal and sanitary investigations (Standard Methods, 1955).

I. DETERMINATIONS TO BE MADE AT THE SOURCE OF SUPPLY

All tests to prove the purity of water can not be made at the well or beside the stream. It is mandatory that some tests be performed at the source of supply of water and that those tests that require heating and incubation be performed in the school laboratory. Water, when left standing, will deteriorate or lose some of its true characteristics; therefore, it is necessary that some

of the determinations or tests be made at the source of water and the remaining tests be made in the school laboratory. Along with the tests made at the source of water, three samples will be collected and carried to the school laboratory to complete the remaining tests.

1. Determination of PH of the water. The pH is the logarithm of the reciprocal of the hydrogen-ion concentration - more precisely, of the hydrogen ion activity- in moles per liter. The pH enters into the calculation of carbonate, bicarbonate, and carbon dioxide; into the calculation of the corrosion or stability index; and into the control of water treatment. The practical pH scale extends from 0, very acid, to 14, very alkaline, with the middle value of pH 7, corresponding to exact neutrality at 25°C. Whereas "alkalinity" and "acidity" express the total reserve or buffering capacity of a sample, the pH value represents the instantaneous concentration of hydrogen-ion (Standard Methods, 1955). Thus, the pH of the water must be checked immediately after the sample is collected because a change in temperature of the water will cause a change in the pH factor. The most important determination is that of the actual pH in the field; a pH determination on a sample which has been exposed to temperature change, storage, agitation, or other

conditions which can cause pH change is of negligible value.

The pH can be measured either colorimetrically or electrometrically. The more accurate of the two is the electrometric which involves a rather expensive piece of equipment. The other technique, and the more commonly employed method, depends upon indicator dyes or dye solutions, which change color as the pH is altered. However, for most pH determinations, the colorometric techniques lend themselves well for use in bacteriological laboratories, in spite of the sacrifice of some accuracy (Wedberg, 1954).

Pure water dissociates very slightly into equal numbers of hydrogen ions and hydroxyl ions, and hence such water is neutral in reaction. At 22°C the hydrogen ion concentration of pure water is 1×10^{-7} gram ions per liter. For convenience this is expressed as a positive number and called pH 7.0. To picture graphically degrees of sourness as they correlate with pH, Table I lists common substances on page 14. Table II, on page 15, correlates the hydrogen ion concentration to the pH factor.

Nitrazine paper is an indicator in acidimetry and alkalimetry and is suggested for use by this study. Nitrazine (sodium dinitrophenyl-azo-naphthol disulfonate)

is an indicator dye showing a series of sharply defined color changes in the pH range 4.5 to 7.5. Nitrazine paper is a very convenient means of determining pH values in this range with an accuracy of a few tenths of a pH unit and is more useful than that of indicators such as litmus, methyl orange, and phenolphthalein.

Nitrazine paper is more broadly useful and more sensitive than litmus. Litmus gives information with respect to one pH point only; there is only one color change with it - the red-blue change at pH 7 -, and it serves only to indicate the relationship of the liquid to that one pH. Moreover, with litmus paper color change occurs sharply only if the pH being determined is outside the range 6-8; between 6 and 7 the pink develops slowly, and between 7 and 8 the blue develops slowly. Obviously, therefore, litmus is not primarily for the determination of pH but rather to establish whether the reaction is acidic or alkaline and whether it is slightly or strongly so. Nitrazine with its series of color changes gives definitive results sharply and quickly at any pH in the range from the moderately acid pH of 4.5 to the slightly alkaline pH of 7.5 in which the majority of the clinical interest lies. Nitrazine, of course, also indicates whether a liquid is stongly acid (pH lower than 4.5) or more alkaline than pH 7.5.

COMMON SUBSTANCES	PH
Hydrochloric Acid	1.0
Human gastric contents	2.0
Ginger ale	3.0
Wines	3.0
Sour pickles	3.2
Tomatoes	4.2
Beans	5.5
Human saliva	7.0
Human blood plasma	7.4
Sea Water	8.2

TABLE I

THE PH OF COMMON MATERIALS
(FROM WEDBERG, 1954)

Grams of Hydrogen ions per liter		pH	Number of times acidity or alkalinity exceeds that of pure water.	
1.0	(10^{-0})	0.0	10,000,000	
0.1	(10^{-1})	1.0	1,000,000	
0.01	(10^{-2})	2.0	100,000	Acid
0.001	(10^{-3})	3.0	10,000	
0.000,1	(10^{-4})	4.0	1,000	
0.000,01	(10^{-5})	5.0	100	
0.000,001	(10^{-6})	6.0	10	
0.000,000,1	(10^{-7})	7.0	0	Neutral (pure H_2O)
0.000,000,01	(10^{-8})	8.0	10	
0.000,000,001	(10^{-9})	9.0	100	
0.000,000,000,1	(10^{-10})	10.0	1,000	
0.000,000,000,01	(10^{-11})	11.0	10,000	Alkaline
0.000,000,000,001	(10^{-12})	12.0	100,000	
0.000,000,000,000,1	(10^{-13})	13.0	1,000,000	
0.000,000,000,000,01	(10^{-14})	14.0	10,000,000	

TABLE II

HYDROGEN ION CONCENTRATION AND PH
(FROM WEDBERG, 1954)

Methyl orange and phenolphthalein are like litmus in that they show only one color change and, therefore, permit only the establishment of the relationship of the pH to their color change-point: methyl orange indicates whether the reaction is lightly or strongly acidic and phenolphthalein whether it is slightly or strongly alkaline. They are useful for titration purposes but not for pH determination.

The accuracy of all colorimetric methods is affected to some extent by salts, proteins, temperature, and certain conditions. The deviation may, under extreme conditions, be as great as 0.5 pH unit or more (Squibb, 1957).

Procedure. To determine pH of a solution one transfers a drop of the fluid to the surface of the paper by means of a glass rod and spreads it evenly by stroking. It is also permissible to dip the Nitrazine paper into the water. After one minute, one compares the color of the paper with the color chart which is supplied. Record the pH of the matched color.

2. Detection of Chlorine in Water. Chlorine in aqueous solution is not stable, and the chlorine content of samples or solutions, particularly weak solutions, will rapidly decrease. Exposure to sunlight or other

strong light or agitation will further reduce the quantity of chlorine present in such solutions. Therefore, it is recommended that chlorine determination shall be started immediately after sampling, avoiding excessive light and agitation. Samples to be analyzed for chlorine cannot be stored.

The treatment of water to make it potable involves the physical removal of enteric pathogens or their chemical destruction. The use of disinfectants goes back to at least 1897 when Jewell added bleaching powder to the water supply of Adrian, Michigan. Since this date the treatment of public water supplies to make them safe has become widespread. Chlorine gas, liquid chlorine, and various chlorine compounds have been employed since Jewell's early work in the field, and sharp declines in the typhoid rate were recorded upon the introduction of this practice. Rates of twenty deaths or more per hundred thousand population have dropped to less than one death per hundred thousand at the present time.

The concentration required to disinfect water satisfactorily is usually not more than one part of chlorine per million parts of water (abbreviated P. P. M.), with a residual of one tenth to three tenths P. P. M. at distant points throughout the distribution system (Wedberg,

1954).

Seven tenths P. P. M. residual chlorine is recommended for swimming pool water. It has been shown that higher concentrations of free residual chlorine can be maintained without bodily irritation as long as the pH of the water is held at about 8.0, and that such concentrations produce a purer water. As the chlorine dosage is increased the residual chlorine concentration also increases but not necessarily at the same rate. Finally a point is reached beyond which no rise in chlorine residual occurs and frequently a decrease in residual takes place. This is known as the breakpoint and is the point at which the chlorine concentration is high enough to break down many of the taste-and-odor producing impurities (Taylor & Company, 1955).

Properly chlorinated water ordinarily cannot be detected organoleptically by the average consumer. Occasional gastro-intestinal outbreaks of non-bacterial origin might arise from filtered and chlorinated water. These outbreaks are probably caused by viruses which are unharmed by the physical and chemical treatment (Wedberg, 1954).

The following method is intended for use: (a) on polluted water; (b) on water in process of purification;

(c) at the end of the purification process; (d) incidental to the distribution of water prepared for human consumption; (e) on swimming pool water; (f) on condenser water; and (g) on industrial process water. Highly polluted waters may require other methods for satisfactory analysis.

Residual chlorine of the water may be determined by the Starch-Iodide Method or the Orthotolidine Method (Theroux, Eldridge, and Mallmann, 1935). Because of the expense of the chlorimeter used in the Orthotolidine Method and the fading of the colors in the Chlorimeter, the Starch Iodide Method will be used. The Chlorimeter costs $26.50, and the color standards carry an unlimited guarantee against fading according to Taylor & Company (1955), the manufacturer. However, the Orthotolidine Method, if Chlorimeter is available, could be used as a reference. Because of the lack of funds of most schools and because the results of the Starch Iodide Method are just as accurate for both drinking water and swimming pool water, the Starch-Iodide Method is more practical at the present.

Preparation of Reagents:

Starch Indicator: Make a thin paste of about 2 grams of starch in cold water. Pour into 200 ml. of boiling water and stir. When cool add a few drops of chloroform.

Standard Sodium Thiosulfate (0.001N): Dilute the

calculated amount of stock Standard sodium thiosulfate solution to one liter with distilled water. Make up fresh every few days.

$$\frac{1}{\text{Normality of stock standard } Na_2S_2O_3} = \text{ml. stock standard } Na_2S_2O_3 \text{ required to make 1 liter of 0.001 N } Na_2S_2O_3.$$ (Theroux, Eldridge, and Mallmann, 1935).

Procedure. Starch-Iodide Method.

1. Place 200 ml. of the sample in an Erlenmeyer flask. Chill to 20°C or lower by immersing the flask in a bath of cold water.
2. Add about 1 gram or a small crystal of Potassium Iodide crystals and 1 ml. of concentrated hydrochloric acid.
3. Add 1 ml. of starch solution.

When the starch solution is added, the presence of a blue color indicates residual chlorine.

4. For a quantitative estimation, titrate the solution with 0.001 N sodium thiosulfate solution until the blue color just disappears. Record the ml. of thiosulfate used. (The formula for an accurate calculation of the amount of residual chlorine is found in the appendix.)

3. Collecting Samples. Among the many tests that have been proposed for ascertaining the biological purity of water, the one that has received almost universal attention, and the value of which is recognized in all water laboratories, is that of the number of bacteria per unit volume of water - not that large numbers of bacteria are within themselves necessarily dangerous, but

it has been definitely established that the most desirable from the point of view of human consumption, is water containing very few bacteria. Therefore, encountering relatively high numbers of bacteria in water is indicative of some abnormal condition, possible of an undesirable nature (Gainey, 1939).

The principal objective of sampling water for bacteriological or chemical examination consists in securing a sample that is representative of the supply and in preserving the bacterial content of the sample, as nearly as possible, in its original state until the water is examined. Since, however, the bacteriological and chemical results obtained may be worthless and perhaps even misleading unless the sample, when examined, is reasonably representative of the supply, a special effort should be made to select a fair sample, to collect it properly, and to prevent an excess alteration of its bacterial content until it reaches the place of examination (Prescott, Winslow, and McCrady, 1947).

Sample Bottles. Samples of water for bacterial or chemical analysis should be collected in clean, sterile, glass bottles with rubber stoppers. Wide-mouth bottles are preferable with a capacity of at least 100 ml.

Samples from Water Systems. The tap and piping

of a water system should be thoroughly flushed before collecting the sample. The amount of flushing will depend upon the time the water has been standing in the piping and the time elapsed since the system was last used. Avoid sampling from taps which might be subject to contamination. If flaming is used to reassure sterility, sufficient heat must be applied to heat the tap to boiling-water temperature. If the surfaces of the tap are dry there is little danger of contaminating the sample, provided the water is allowed to flow several minutes. Avoid sampling from wet taps and temporary attachments which may be fastened to the tap.

Samples from pools, lakes, and rivers. When samples are collected from pools, lakes, or rivers, care should be taken to obtain the sample from a point which represents average conditions of the supply. In all cases where samples are collected from standing water, one should remove the stopper aseptically and plunge the bottle beneath the surface, mouthdown, to a depth of several inches. The hand and bottle should make a wide arc as they rapidly pass into and out of the water; by this procedure water that has been contaminated by the hand is prevented from entering the bottle. One should discard an amount of the water and replace the stopper immediately (Prescott, Winslow, and McCrady, 1947).

Pools. In a swimming pool, the sample should be collected at the side of the pool at a point near the deepest part during periods of use, preferably at the time of the heaviest bathing load of the day. The samples should not be taken in the absence of bathers and the chlorine residuals should be made at the pool side (Prescott, Winslow, McCrady, 1947).

Lakes. Lake samples should be taken from a boat at a distance of twenty-five feet or more from the shore. The water should have a depth of at least four feet and preferably more at the sampling point. A sampling rod is an extremely useful aid. It consists of a rigid or jointed rod of almost any sturdy material, from bamboo to steel, provided with a right-angled base to which the sample bottle is fastened. When the bottle is lowered by means of the rod to the desired sampling point, the stopper can be jerked out by pulling an attached cord; or a mechanism, actuated by a spring, which will remove the stopper and replace it after the bottle has been filled, can be fixed near the lower extremity of the rod (Prescott, Winslow, and McCrady, 1947).

Rivers. Samples from a river should represent flowing water and not stagnant pools. Samples may be collected in a straight stretch of the river, about four

feet from the banks. In a meandering stream the samples should be collected at the point of greatest depth or from the center of the stream.

All samples should be closed and tested as soon as possible after they have been collected, because of the biological and chemical changes that may occur. In warm weather, if the transportation period exceeds more then one hour, the sample should be iced. The time of transportation and storage should not exceed six hours for impure waters and not more than twelve hours for relatively pure waters. Samples should be stored at a temperature between 6 and 10°C. Samples stored longer than twenty-four hours should be discarded, or the results obtained should be judged accordingly (Theroux, Eldridge, and Mallmann, 1935).

Unless the supply to be examined is sampled at frequent intervals, more than one sample should be collected in order that results reasonably representative of the quality of the water may be assured. Particular care should be taken to reassure the proper labeling of each sample. Gummed labels are not adequate because moisture collects on the bottles and will cause the labels to become detached. If the bottles do not have frosted labels or permanent numbers, tags attached with wire are best.

II. DETERMINATIONS TO BE MADE IN THE SCHOOL LABORATORY

The following standard test methods are best performed in the school laboratory because of the amount of equipment and source of heating required. Conditions at a source of water supply are not stable, and the samples may become contaminated.

1. Detection by Odor Quality - hot and cold.

The olfactory sense is a most sensitive means for detecting small concentrations of odoriferous substances; but, unfortunately, it lacks precision. There is no absolute odor value. Different persons react differently to a given odor concentration, and even the same individual may report a different intensity for a given concentration of odoriferous substance at different times. Therefore, it follows that an odor value obtained on a certain day should not be compared with a value obtained on another day, unless there is some common basis for comparison. There are two ways of making odor values more precise, and both are recommended. The first is to compare the unknowns continually with a freshly prepared solution of a known concentration of an odorous substance, such as phenol or butyl alcohol. The second is to average the results of as many observers as possible; for instance, as many as fifteen persons are used in evaluating the

flavor of food in commercial testing laboratories. The method to be used depends upon the purpose for which the test is intended. The first, Odor Quality, is best suited for the interpretation of consumer complaints and for assistance in deciding when odors caused by microscopic organisms are excessive; the second, Threshold Odor, is best suited for use in the treatment of plants in determining the odor-removal treatment required, or for water systems in which variable proportions of two or more separate supplies may be mixed (Standard Methods, 1955). Of course, for the purpose of this study the Odor Quality method will be used.

Apparatus. Odor-free glassware is best prepared by scouring powder, by steaming it out with clean steam, or by soaking it in chromic-sulfuric cleaning mixture. Glassware should be cleaned just before use, because it often develops a slight earthy odor on storage. Soaps or detergents may leave persistant odors on the glassware. The final rinsing of glassware should be made with odor-free water. Wide-mouth bottles with glass stoppers or wide mouth Erlenmeyer flasks are suitable. Rubber stoppers and corks should be avoided; if glass-stopper vessels are not available, watch glasses may be used as covers.

Reagents. Odor-free water is obtained by passing

tap water through a column of granular activated carbon at a slow rate. It is best to prepare odor-free water only as and when needed, because such water may develop odor upon standing (Standard Methods, 1955).

Procedure. Certain conditions are required to obtain consistent results. Some degree of practice and experience is a necessity. The analyst should avoid smoking or the consumption of highly seasoned food prior to testing. The odor-free water, when prepared, should be truly free of all detectable odor. All glassware must be odor-free. All dilutions when examined for odor should be at the same temperature (within 1°C.). Each dilution should be compared with an odorless standard. This simplifies the work of the analyst because his task then is to decide only whether an odor is present in the dilution. The odor-free standard checks judgment and minimizes reliance upon memory. A sudden change in the character of the odor during the testing procedure should be considered a warning that there may be interference from outside odors or that the dilution water is not odor-free. The character of odor should always be described for future consideration. The test should be conducted in a room free from odors (Standard Methods, 1955).

Cold Odor Quality:

1. Shake about 125 ml. sample at 20°C in a 250 ml. wide mouth Erlenmeyer flask. Lightly sniff the odor. Avoid vigorous or repeated shaking as the odor will be dissipated.

Hot Odor Quality:

1. Pour about 125 ml. sample into a 250 ml. Erlenmeyer flask. Close the mouth of the flask with a watch glass. Heat the water to approximately 58° to 60°C., agitate it with a rotary movement; slip the watch glass aside; and sniff the odor.

The odor is reported as to both intensity and type. Results are reported on both hot and cold samples. The intensity and type are recorded in the form "VDp", which would represent a water unfit to drink because of excessive Anabaena. Results may be determined from Table III and Table IV on page 29.

2. Detection of Water Hardness. Water that is safe for drinking purposes is not always suitable for laundry purposes or boilers in heating systems or factories. When one adds soap to water, the desired reaction is the formation of immediate suds, as soap does not cleanse unless suds are present. Usually, however, a sticky scum or curd is first obtained and increased amounts of soap are needed to form a lather. Water which at first forms a "curd" is called hard water; while water which immediately forms a rich lather with a small amount of hardness, and the ingredients causing the hardness are

TABLE III

INTENSITY EXPRESSION

Intensity	Definition
0	No odor detectable.
I	Odor would not be detected by an average consumer but would be detected by experienced observer.
II	Odor might be detected by an average consumer but only if his attention were called to it.
III	Odor would be readily detected and might cause the water to be regarded with disflavor.
IV	Odor would force itself upon the attention and might make the water unpalatable.
V	Odor of such intensity that the water is absolutely unfit to drink.

TABLE IV

ODOR TYPE

Abbreviation	Nature of Odor	Description
A	Aromatic (spicy)	Camphor, cloves, lavender, lemon.
Ac	Cucumber	Synura.
B	Balsamic(flowery)	Geranium, violet, vanilla.
Bg	Geranium	Asterionella.
Bn	Nasturtium	Aphanizomenon.
Bs	Sweetish	Coelosphaerium.
Bv	Violets	Mallomonas.
C	Chemical	Industrial wastes or chemical treatment.
Cc	Chlorinous	Free Chlorine.
Ch	Hydrocarbon	Oil refinery wastes.
Cm	Medicinal	Phenol and iodoform.
Cs	Sulfuretted	Hydrogen sulfide.
D	Disagreeable	(Pronounced unpleasant Odors)
Df	Fishy	Uroglenopsis and Dinobryon.
Dp	Pigpen	Anabaena.
Ds	Septic	Stale sewage.
E	Earthy	Damp earth.
Ep	Peaty	Peat.
G	Grassy	Crushed grass.
M	Musty	Decomposing straw.
Mm	Moldy	A damp cellar.
V	Vegetable	Root vegetables.

(STANDARD METHODS, TENTH EDITION, 1955)

determined, the water can then be treated with a sufficient amount of certain softeners to counteract its hardness. It is only through this action that bacteria of dirt and filth can effectively be removed from clothes through washing. However, this hardness may effect the taste and purity of the water.

Water acquires its hardness from contact with mineral bearing substances in the earth's crust. Both hard and soft water are clear and colorless. Therefore, the hardness of water must be due to certain substances, salts, dissolved in water. The hardness of water is caused principally by the elements calcium and magnesium and sometimes by iron and aluminum. Most of the calcium and magnesium is present in natural waters as bicarbonates, sulfates, and sometimes chlorides and nitrates (Luros and Oram, 1943). Hardness producing substances react with soaps, forming insoluble compounds before a lather is produced. There are two types of water hardness, temporary and permanent. Temporary hardness is caused principally by bicarbonates of calcium, magnesium or iron, and can be softened by boiling. Permanent hard water contains sulfates and chlorides of calcium, magnesium or iron, and cannot be softened by boiling. Permanent hard water can be softened by such chemicals as sodium carbonate, or washing soda, sodium tetraborate, household ammonia,

lye, or a mixture of calcium hydroxide and sodium carbonate (Luros and Oram, 1943).

There are on the market softeners which exchange the sodium in their salts for the calcium, magnesium, or iron in the hard water.

The amount of hardness is always expressed in terms of calcium carbonate; however, a report showing one hundred parts per million does not signify just what compounds cause the hardness but only that the hardness is equivalent to that produced by one hundred parts per million of calcium carbonate. In calculating the determination, the results are obtained in parts per million of the element as found in the appendix of this thesis.

Water having less than fifty to seventy-five parts per million of hardness is generally considered as sufficiently soft for the ordinary uses of water. Water having seventy-five to one hundred and fifty parts per million of hardness may be considered moderately hard but still not sufficiently hard to interfere seriously with its use for most purposes or to cause much demand for water softening. Hardness above one hundred and fifty parts per million is noticed by most persons; and, if the hardness is above two hundred parts per million,

many homes will have to be provided with household softners or cisterns (Luros and Oram, 1943).

Procedure. An approximate determination of total hardness may be made with a soap solution of known strength. Total hardness may also be obtained by the soda-reagent method. This soda-reagent method is more accurate and gives quantative results.

Reagents. The soda reagent is made by dissolving about two grams of sodium hydroxide and about 2.65 grams of anhydrous sodium carbonate in distilled water and made up to one liter (Theroux, Eldridge, and Mallmann, 1935).

Total Hardness, Soda-Reagent Method.

1. Pipette 200 ml. of the sample into a 500-ml. Erlenmeyer flask.
2. Add 3 drops of methyl orange.
3. Add 0.02 N sulfuric acid from a burette until the first permanent color change is observed.
4. Place 200 ml. of distilled water into a second flask.
5. Boil each for 5 minutes.
6. Add exactly 25 ml. of soda reagent to each flask and boil for 10 minutes until the volume is reduced to about 150 ml.
7. Cool and pour into 200-ml. volumetric flasks.
8. Rinse the solutions into the volumetric flasks with small quantities of hot distilled water.
9. Make up to the mark with boiled distilled water and mix.
10. Filter each solution, rejecting the first 50 ml. of each filtrate.
11. Pipette a 50-ml. portion of each filtrate into an Erlenmeyer flask, add 3 drops of methyl orange and titrate each portion with 0.02 N sulfuric acid. Record the ml. of acid used. (The

calculations for p. p. m. of total hardness as Calcium carbonate is illustrated in the appendix.)

3. Determination by Color. The expression "color" shall be defined to mean "true color" - that is, the color that is due only to substances which are actually in solution, and not to suspended matter. The "apparent color" shall include not only the color due to substances in solution, but also that due to suspended matter. Apparent color is determined on the original sample without filtration or centrifugation.

Color is determined by visual comparison of the sample with known concentrations of colored solutions. The platinum-cobalt method of measuring color shall be the standard method, and the unit of color shall be that produced by one milligram of platinum, in the form of the chloroplatinate ion, per liter. The use of liquids as standards for laboratory work is permissible only if these have been individually calibrated against platinum-cobalt standards. Samples for the color determination should be representative and taken in clean glassware. The color determination should be made within a reasonable period of time, as biological changes occurring in storage may affect the color.

Preparation of standards. If a reliable supply of

potassium chloroplatinate cannot be purchased, it may be replaced by chloroplatinic acid, which the analyst can prepare from metallic platinum.

1. Dissolve 1.245 g. potassium chloroplatinate, K_2PtCl_6, equivalent to 0.500 g. metallic platinum and 1 g. crystallized cobaltous chloride, $CoCl_2 \cdot 6H_2O$, equivalent to about 0.25 g metallic cobalt in distilled water with 100 ml. concentrated hydrochloric acid, and dilute to 1 liter with distilled water. This stock standard has a color of 500 units.
2. If potassium chloroplatinate is not available, dissolve 0.500 g. pure metallic platinum in aqua regia with the aid of heat; remove HNO_3 by repeated evaporation with fresh portions of concentrated HCl. Dissolve this product together with 1 gram crystallized cobaltous chloride.
3. To prepare standards having colors of 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 60, and 70 dilute 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, 4.5, 5.0, 6.0, and 7.0 ml. stock color standard with distilled water to 50 ml. in standard tall-form 50 ml. Nessler tubes. Protect these standards against evaporation and contamination when not in use (Standard Methods, 1955).

Procedure. The color of a sample shall be observed by filling a matched Nessler tube with the water to be examined to the 50-ml. mark and comparing it with the standards. The observation shall be made by looking vertically downward through the tubes toward a white or specular surface placed at such an angle that light is reflected upward through the columns of liquids. If turbidity is present, the true color shall be determined after the removal of turbidity by centrifuging. The sample shall be placed in a suitable centrifuge tube or

COLOR UNITS	RECORD TO NEAREST UNITS
1 - 50	1
51 - 100	5
101 - 250	10
251 - 500	20

TABLE V

EXPRESSION OF COLOR DETERMINATIONS

(STANDARD METHODS, TENTH EDITION, 1955)

tubes and centrifuged until the supernatant is clear. The time required will depend upon the nature of the sample, the speed of the motor, and the radius of the centrifuge; but rarely will more than one hour be necessary. The centrifuged sample shall be compared in a Nessler tube with distilled water to insure the elimination of all turbidity. If clear, the sample is then compared with the standards. If the color exceeds 70 units, the sample shall be diluted with distilled water in known proportions until the color is within the range of the standards, and the results shall be multiplied by the appropriate dilution factor (Standard Methods, 1955).

In thin layers water appears colorless, but in thick layers it has a bluish-green color. Therefore the water could not be considered safe if it had a determination above 1 (Briscoe, 1945). The results of color determination shall be expressed in whole numbers and recorded as shown in Table V on page 35.

4. Determination by Total Residue. Residue is the general term used for the residue left after a sample is evaporated at a definite temperature, usually 103° to 105°C. Total residue includes both suspended and dissolved residue.

The following method may be used with all types of

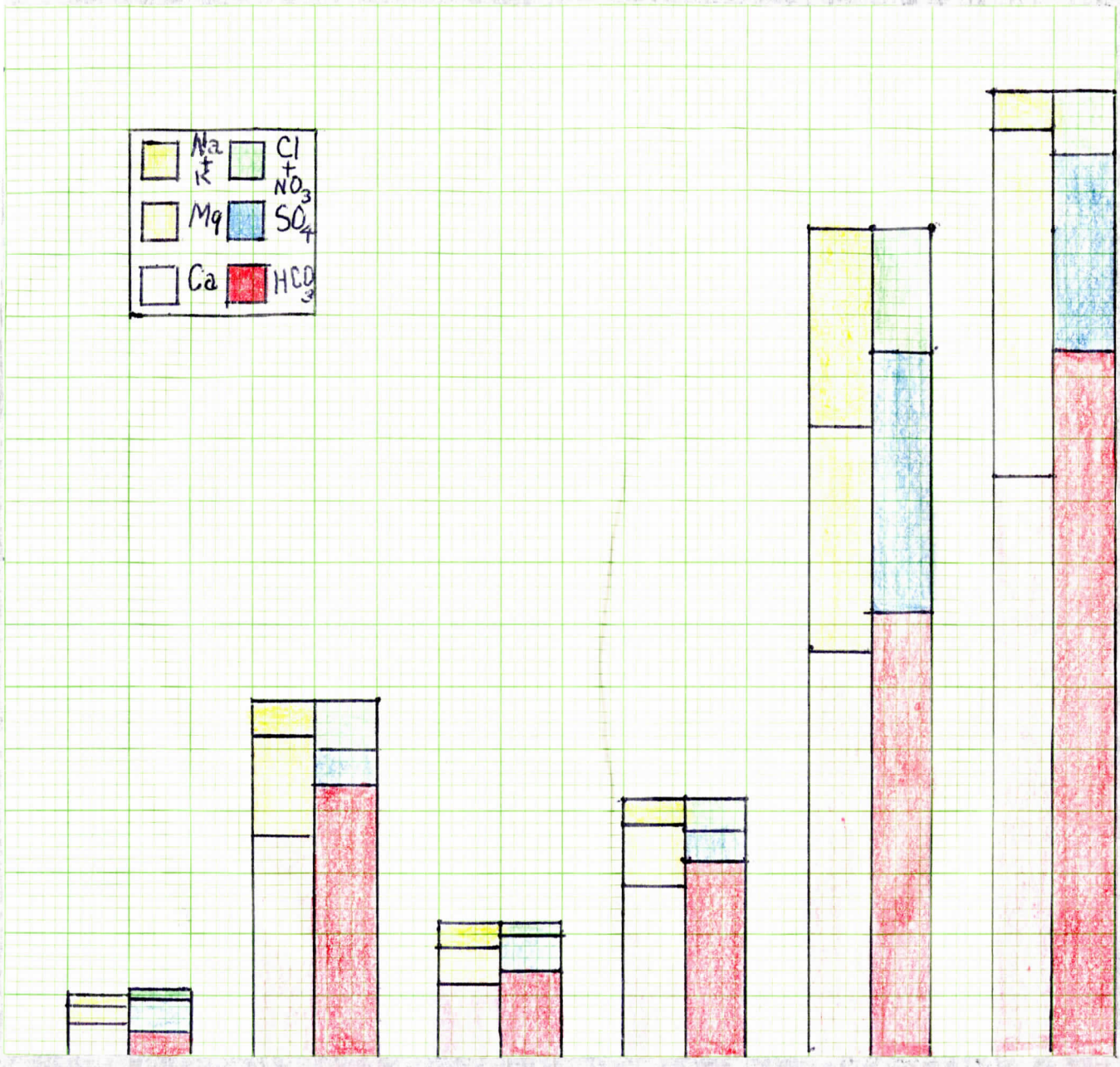


TABLE VI

COMPOSITION OF WATER

(Composition of water at the six largest cities in the United States and the largest public well supply in Ohio.)

(FROM HOOVER, 1946)

waters, but care should be exercised in the interpretation of the results. Since the weight of the residue will vary with the temperature of drying, a definite temperature must be used to obtain consistent results. The results are best verified by testing duplicate portions. See Table VI, page 37, for the composition of water.

Procedure. Place a 100 ml. sample of water, thoroughly shaken and unfiltered, in a weighed evaporating dish. The dish should preferably be made of platinum; but silica, porcelain, or pyrex dishes may be used. Place the dish containing the sample in an oven maintained at 103° to 105°C and evaporate the sample to dryness. If more convenient, evaporation may be carried out on a steam bath, but the final drying should be done in the oven. After all of the liquid has evaporated, continue to dry the residue to constant weight. If time is no obstacle, allow the residue to dry overnight. After drying, allow the warm dish to cool in a desiccator and weigh. Report the increase in weight as total solids or residue on evaporation. An accurate calculation of the total residue is found in the appendix of this thesis.

The U. S. Public Health Service Drinking Water Standards (1946) suggests that "total solids" (total residue on evaporation) shall not exceed 500 mg./liter in

waters of good chemical quality. However, if such water is not available, a "total solids content of 1,000 mg./liter may be permitted.

5. Determination by Bacterial Analysis. Sterile water is rarely found in nature. Water obtained even from deep wells and springs usually contains a few bacteria. Surface waters (those of streams, ponds, lakes, and the water of the ocean) always contain bacterial in greater or smaller numbers. These organisms may be designated as the normal water flora, as they are constantly present and in no case are capable of producing disease in man. For the most part, they are organisms that grow best at the temperature of the water in which they are found.

Cocci are common in water. They are largely chromogenes, either micrococci or sarcinae. Probably the commonest of these is Sarcina lutea, a form developing lemon-yellow colonies when grown upon artificial media. Sarcina aurantiaca, which produces a golden yellow pigment, is also common, as are other cocci producing pink, red, yellow, and orange pigments. Certain non-chromogenic cocci, as Micrococcus candicans, may also be found.

Chromogenic bacilli such as Serratia marcescens (red pigment), Flavobacterium aurantiacum (orange pig-

ment), and Chromobacterium volaceum (violet pigment) - are common in surface waters. Bacilli such as Pseudomonas fluorescens, which form fluorescent colonies upon media, are also commonly present. Whenever water receives a considerable amount of surface drainage, the normal soil bacteria, such as various members of the Bacillus subtilis group, are practically always present (Buchanan, 1946).

The presence of considerable quantities of organic matter leads to the multiplication of many putrefactive bacteria, such as the spore-bearing anaerobes. While these are not necessarily injurious to health, they are undesirable.

The organisms of importance in impure water may be either pathogenic or non-pathogenic. Among the pathogenic microorganisms which may gain entrance to water supplies through contamination with sewage are the bacteria causing typhoid fever, paratyphoid, bacillary dysentary, Asiatic cholera, and the protozoan of amoebic dysentery.

An impure water is generally defined as one which received more or less sewage, one, therefore, that is not fit for human consumption because of the possible presence of pathogenic bacteria. Relatively few species of organisms are held to be indicative of sewage pollution.

The most important of these are Escherichia coli, Aerobacter aerogenes, Streptococcus pyogenes, and Clostridium Welchii.

From the standpoint of human health, the most important test conducted on water is the bacteriological analysis. While people are interested in knowing the numbers of living organisms present in water to be used for drinking, they are more interested in the source of these bacteria.

The standard technique calls for the determination of the presence of pollution; and if sewage organisms are found, the water is condemned. One of the easiest ways to detect sewage is to look for Escherichia coli. This organism is found in the excreta of man and other warm blooded animals. If Escherichia coli is present, there exists the possibility that enteric pathogens might also be expected. However, by using the index organism, Escherichia coli, it can be determined with a relatively high degree of accuracy whether a water supply is polluted. If this organism can be detected in the water, feces can be said to have found their way into the supply, and such water is potentially dangerous.

Escherichia coli is a short, gram negative, non spore-forming rod that ferments lactose with both acid and gas production. This is a unique set of reactions

reserved for a limited number of organisms, and its uniqueness provides a simple screening technique for determining the presence of sewage.

Preparation of Nutrient Agar. In order to detect the presence of bacteria, it is necessary to have something in or upon which they will grow. There is no limit to the variety of substances used as culture media; some are liquid, some solid, some semi-solid; whereas a few are liquid at one temperature and solid at another and are termed liquefiable-solid media. The most extensively used consists of nutrient broth to which 1-2% agar-agar is dissolved. The medium then forms a firm jel at temperatures below 42°C and is called nutrient agar. The most commonly employed medium in water bacteriology is nutrient broth to which has been added 0.5% of the sugar lactose (Standard Methods, 1955).

Procedure.

1. Add 3 grams of beef extract (0.3%) and 5 grams of peptone (1%) to 1000 cc of distilled water in a double boiler.
2. Heat in a double boiler to 65°C, stirring until dissolved. This will require approximately one-half hour.
3. Make up the lost volume with distilled water and adjust the reaction with litmus paper so that the final pH is between 6.4 and 7.0.
4. Bring to a boil over a free flame, cool to 25°C., make up the lost volume with distilled water.

5. If the Media is to be used in the Durham Fermentation tube, add 0.5% by weight, of lactose, C. P. Otherwise, omit this step.

6. Filter through absorbent cotton or filter paper until clear.

7. Distribute in test tubes, 10 cc to each tube, or in other desired containers and place the remainder in a flask.

8. Sterilize in a pressure cooker at 15 lbs pressure for 15 minutes (Levine, 1947).

Broth is best stored at room temperature and should be in such quantities that evaporation is not excessive before the entire batch is used. Tubes containing gas bubbles in the inserts should be discarded. Tubes should be plugged with sterile cotton to prevent contamination and slight evaporation.

Standard Plate Count.

Dilution. Dilution bottles shall be sterilized in the pressure cooker at 121°C (15 lbs.) for Fifteen minutes after the temperature reaches 121°C. Dilution bottles or tubes shall be filled with the proper amount of water so that after sterilization they shall contain the quantity desired, with a tolerance of 2 per cent. The exact amount of water to be placed in the bottles may be determined only by experiment with the particular pressure cooker. The sample bottle shall be shaken vigorously, twenty-five times, and then 10 ml., 1 ml., or 0.1 ml. withdrawn with a standard sterile pipette and added to

the proper dilution bottle, tube, or petri dish as required. Each dilution bottle or tube after the addition of the portion of the sample shall be shaken vigorously, twenty-five times, before a second dilution or sample is removed.

Plating. One ml., 0.5 ml., or other appropriate volume, of the sample or dilution shall be used for plating and shall be placed in the petri dish first. In the examination of sewage or highly turbid waters 0.5 ml. inoculum of the original sample should not be measured directly, but an appropriate dilution should be used. The cover of the petri dish shall be lifted just enough for the introduction of either the pipette or culture medium, and the lips of all test tubes or flasks used for pouring the medium shall be flamed. The medium and sample in the petri dish shall be thoroughly mixed and uniformly spread over the bottom of the petri dish by tilting and rotating the dish. All plates shall be solidified as rapidly as possible after pouring and then be placed immediately in the appropriate incubator or oven. Not more than twenty minutes shall elapse between plating and pouring (Standard Methods, 1955).

Incubation. Agar medium may be used for counts made either at 20°C plus or minus 0.5°; or 35°C plus or

minus 0.5°. The time for incubation at the 20°C temperature shall be 48 plus or minus three hours and that at the 35°C temperature shall be 24 plus or minus two hours. Glass-covered petri dishes shall be inverted in the incubator or oven (Standard Methods, 1955).

Counting. In preparing plates, such amounts of the water under examination shall be planted as that which will give from 30 to 300 colonies on a plate. The aim should always be to have at least two plates giving colonies between these limits, except as provided below.

Ordinarily, it is not desirable to plant more than 1 ml. water in a plate; therefore, when the total number of colonies developing from 1 ml. is less than 30, it is obviously necessary to record the result as observed, disregarding the general rule given above.

In determining the standard plate count, only such plates should be considered as show 30 to 300 colonies except as provided in the preceding paragraph. If the same amount of water has been planted in two or more replicate plates and of these one shows colonies within these limits while others show less than 30 or more than 300 colonies, the results recorded should be the average of all the plates planted with this volume of sample except as provided in the preceding paragraph.

Counting shall be done with an approved counting aid, such as the Quebec colony counter. If such equipment is not available, counting may be done with a magnifying glass. In order to insure uniformity of counting conditions, illumination equivalent to that provided by the Quebec colony counter shall be employed. Most any direct lighting will be satisfactory.

In order to avoid fictitous accuracy and yet express the numerical results by a method consistent with the precision of the technique employed, the recorded number of bacteria per ml. shall include not more than two significant figures. For example, a count of 142 is recorded as 140, and a count of 145 is recorded as 150; whereas a count of 35 is recorded as such. Counts shall be designated as the "standard plate count at 20°C" or the "standard plate count at 35°C."

Presumptive Test.

Durham Fermentation tube. Put the media with lactose in the Durham tube. Inoculate the tube and let it stand for twenty-four to forty-eight hours at 37°C. Check the development at the end of twenty-four hours and forty-eight hours. Bubbles indicate bacteria. Most intestinal bacteria will produce gas because of their action with the lactose in the media. Bubbles are an indication of contamination of the water supply and should be condemned. The Durham Technique

is used very widely because it is simple. It is an excellent method for indication of intestinal bacteria but does not measure how many. Formation within forty-eight plus or minus three hours of gas in any amount in the fermentation tube constitutes a positive presumptive test.

The appearance of an air bubble must not be confused with actual gas production. If the gas formed is a result of fermentation, the broth medium will become cloudy and active fermentation may be shown by continued appearance of small bubbles of gas throughout the medium outside the fermentation tube when gently shaken.

The absence of gas formation at the end of forty-eight plus or minus three hours incubation constitutes a negative test. An arbitrary limit of forty-eight hours observation doubtless excludes from consideration occasional members of the coliform group which form gas very slowly, but for the purpose of a standard test the exclusion of these occasional slow gas-forming organisms is generally satisfactory (Standard Methods, 1955).

Confirmed Test. All primary fermentation tubes showing any amount of gas at the end of twenty-four or forty-eight hours incubation shall be subjected to the confirmed test. All tubes producing gas in twenty-four hours that have not been submitted to the Confirmed Test

shall be recorded as containing organisms of the coliform group, even though all the confirmed tests made yield negative results. Submit to the Confirmed Test all tubes of all dilutions of the original sample in which gas is produced only at the end of forty-eight hours incubation.

Procedure with Violet Red Bile Agar. Violet Red Bile Agar is purchased ready for rehydration. it does not have to be autoclaved or sterilized in the pressure cooker due to the presence of bile.

1. To rehydrate the medium, suspend 41.5 grams in 1000 ml. of cold distilled water.
2. Heat to boiling to dissolve the medium completely.
3. Cool to 40°C to 44°C and pour 15 ml. of the medium into petri dishes containing the inoculum.
4. After the inoculated medium has solidified, pour 4 ml. of the medium over the surface to form a thin covering film.
5. The inoculated plates are incubated at 37°C. for 18 to 24 hours and then observed by transmitted light.
6. Members of the coliform group form purplish-red colonies surrounded by a zone of precipitated bile salts (Difco Laboratories, 1956).

If typical coliform colonies, purplish-red surrounded by a zone of precipitated bile salts, have developed on the plate within the incubation period, the result

of the Confirmed Test may be considered positive and the water supply condemned. If only negative colonies have developed within the incubation period, the results of the Confirmed Test may be considered negative.

GENERAL PROCEDURE FOR BACTERIAL ANALYSIS

1. Shake the sample thoroughly to insure even distribution of the organisms, then open the bottle, taking care not to contaminate the sample with the fingers or equipment. This same procedure is followed for each sample representing the water supply.

2. Place 10 cc of the sample in a Durham Fermentation tube, discard the pipette.

3. Place 1 cc of the sample into:
   a. each of two petri dishes.
   b. a Durham Fermentation tube.
   c. a bottle of dilution water.

4. Place 0.1 cc of sample (1 cc of the diluted sample) into:
   a. each of two petri dishes.
   b. a Durham Fermentation tube.

5. Pour into each Petri dish a tube of melted agar which has been cooled to 42°C to 45°C. Tip the dish back and forth to insure good distribution and put aside to solidify.

6. Place the agar plates in the 37°C incubator for 24 hours; also incubate the lactose broth fermentation tubes at the body temperature.

7. Count the number of colonies on each, at the end of 24 hours incubation and record in tabular form. See Table VII on page 50.

8. Record the gas formation in the Durham Fermentation tubes after 24 hours and 48 hours.

1	to	50	colonies,	record	as	found
51	to	100	"	"	to nearest	5
101	to	250	"	"	" "	10
251	to	500	"	"	" "	25
501	to	1,000	"	"	" "	50
1,001	to	10,000	"	"	" "	100
10,001	to	50,000	"	"	" "	500
50,001	to	100,000	"	"	" "	1,000
100,001	to	500,000	"	"	" "	10,000
500,001	to	1,000,000	"	"	" "	50,000
1,000,001	to	10,000,000	"	"	" "	100,000

TABLE VII

EXPRESSION OF PLATE COUNTING

(FROM GAINEY, 1939)

9. If gas is present, complete the Confirmed Test for the presence of Coliform bacteria.

Interpreting Results. In general, the greater the variety of colony types on a plate, the more unfavorable is the indication, for polluted waters usually contain a multiplicity of bacterial species.

The ratio of the 37° to the 20° plate count should be noted. If this ratio is low - below 10% - and particularly if the body temperature count is low for the type of water examined, the indication is favorable. Low counts at both temperatures are very reassuring. Not infrequently extremely low counts in the presence of somewhat doubtful coliform results, especially in the examination of ground waters, furnish conclusive confirmation of favorable sanitary-survey findings. On the other hand, an increase in the count of a water that usually contains few bacteria may afford the first indication that something is wrong.

The first days gas indications are particularly significant, for the production of gas in twenty-four hours in the enrichment tube usually means that normal coliforms are present. When tubes of lactose broth are inoculated with water and the tubes are incubated at body temperature, the production of acid and gas through the breakdown of lactose leads the analyst to presume that coliform

bacteria are present in the water. The term coliform refers to all species of the genera Escherichia and Aerobacter. The Aerobacter is generally of soil or plant origin, but as high as 10% of fecal samples may be found to harbor these organisms. Escherichia Coli is considered to be primarily of fecal origin. The interaction of two or more species of bacteria, however, may yield acid and gas from lactose because of synergism. For this reason, it is important that positive presumptive tests be carried at least one step further before the water can be said to contain sewage (Wedberg, 1954).

The interpretation of results is much simplified if regular and frequent bacteriological examination of a water supply is practiced. A single analysis of a water reveals only its bacterial content at the moment of sampling. Repeated examination, however, furnishes a conception of the normal variations to which the bacterial population of the supply may be subject, and thereby greatly facilitates evaluation of the significance of departures from the normal that otherwise might be overlooked or misconstrued (Prescott, Winslow, McCrady, 1947).

In the very early years of water examinations Sternberg (1892), in a conservative fashion, stated that a water containing less than 100 bacteria per ml.

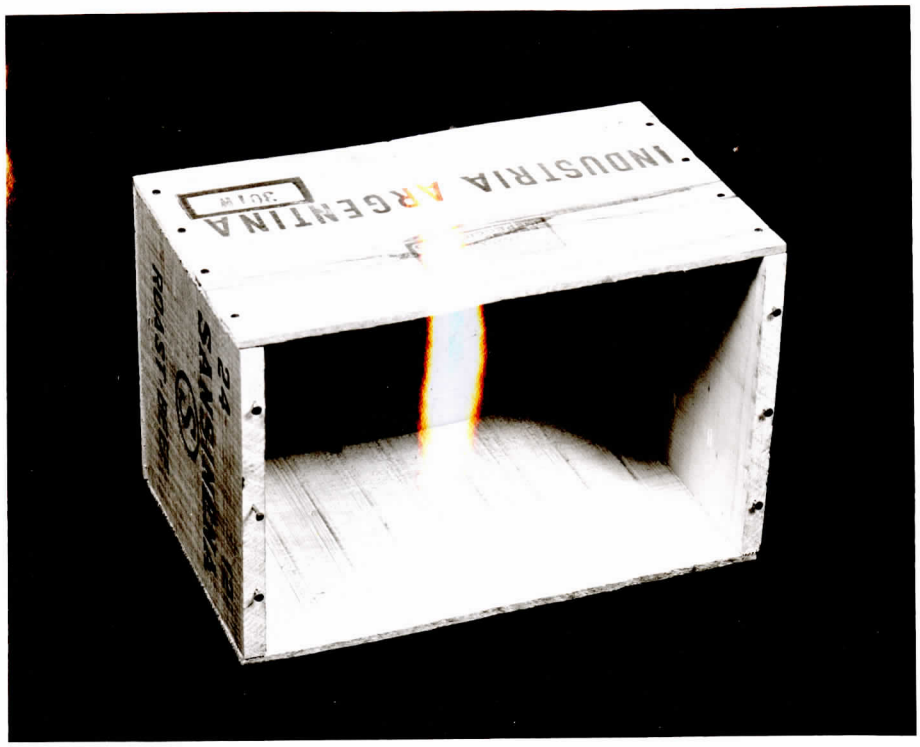
is presumably from a deep source and uncontaminated by surface drainage, that one with 500 bacteria is open to suspicion, and that one with over 1,000 bacteria is presumably contaminated by sewage or surface drainage. This is probably as satisfactory an arbitrary standard as could be devised, but any such standard must be applied with great caution. The source of the sample is of vital importance in the interpretation of analysis; a bacterial count which would excite suspicion in a spring might be quite normal for a lake; only figures in excess of those common to unpolluted waters of the same type give an indication of danger. Furthermore, the bacteriological tests are far more delicate than any others at one's command; very minute additions of food material cause an immense multiplication of the microscopic flora. It is necessary, then, in the first place, to exercise the greatest care in allowing for possible error in the collection and handling of bacteriological samples; in the second place, only well-marked differences in numbers should be considered significant (Prescott, Winslow, and McCrady, 1947).

CHAPTER III

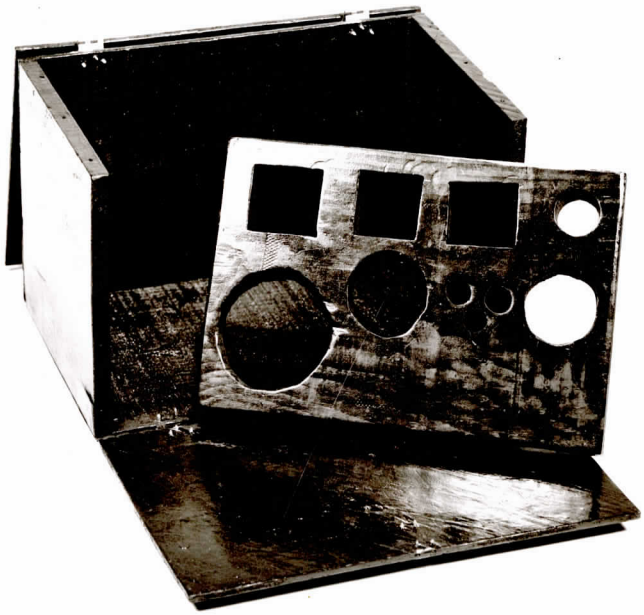
CONSTRUCTION OF AND NEEDED MATERIALS FOR A WATER TESTING KIT

There are no rigid or special requirements or specifications which should be adhered to in order to construct a water testing kit suitable for performing the tests mentioned in Chapter II. Better kits can be made than the one that has been built for this study. To encourage teachers to construct such a kit and to project this idea to the interested students of science, in the building of this kit, over simplification has been emphasized to show the value that can be derived from such a simple construction. The only objective necessary is that the water testing kit serve its purpose.

Picture I on page 55 is one taken at the beginning of the construction of the water testing kit, made for this thesis. The pictured box was obtained from a local A & P Grocery store at no charge. As shown, one side was absent and could not have been used because of its destruction when the box was opened at the store. The box was made of pine wood, 13 and 1/2 inches long, 9 inches wide, and 9 inches high. Ideally, a box of hardwood would be better to withstand the rough treatment that such



PICTURE I



PICTURE II

a kit must endure. However, the softer woods, with a good finish, may be used satisfactorily. The wooden box was first sanded, and a piece of plywood 13 and 1/2 inches in length and 9 inches wide was fitted to the fourth side of the box, in order to enclose the box. The plywood was fitted to the box with small hinges purchased at the 5 & 10 cent store for twenty-nine cents. The hinges were fitted two inches in from either side at the back of the box. The plywood side was intended as the top side of the box.

All nails were removed from the front side of the box, and the front panel was fitted to the box with hinges similar to those used to hinge the top side. The box was then painted, inside and outside, with two coats of black paint. One may use any other color that is desired. Black was used on this construction to help cover up the acid drops which burn into the wood. Picture II on page 55 shows how the sides were fitted to the box. The hinges should work well and be mounted with screws instead of nails. Nails have a tendency to pull out of the wood; while the screws will hold more securely in the wood.

A 1-inch thick poplar board 12 and 1/4 inches long and 8 and 1/4 inches wide was cut to fit snugly in the bottom of the box. Necessary placement of the equipment

in the box was determined, and holes were cut in the poplar board to correspond to each chemical or piece of glassware that would be fitted in the kit. The writer's arrangement included three holes, 2 inches square; one hole, 3 and 1/2 inches in diameter, one hole, 1 and 1/4 inches in diameter, two holes, 2 and 1/4 inches in diameter, and three holes, 3/4 of an inch in diameter. The holes were cut out, the poplar board was then sanded and painted. Upon drying, the poplar board was nailed to the bottom of the box to prevent the chemicals and glassware from capsizing while moving or carrying the water testing kit.

As shown in Picture III on page 58, three sampling bottles, one 100 ml. Nessler Tube, one 250 ml. Erlenmeyer flask, one 100 ml. bottle of concentrated Hydrochloric Acid, one 100 ml. of 0.001 N Sodium Thiosulfate, one vial of Nitrazine paper, one vial of Potassium Iodide, and one vial of starch solution with a dropper were placed in their respective place in the bottom of the kit. On the inside surface of the top of the kit, two pieces of 1-inch plastic strips were tacked eight inches apart to hold one 10 ml. pipette, one graduated 1 ml. pipette, and one ungraduated 1 ml. pipette. The Nitrazine paper color standards can be tacked below the pipettes on the inside surface of the top of the kit. The analysis sheet, as

PICTURE III

DESCRIPTION OF SOURCE

and

RESULTS OF ANALYSIS OF

SAMPLE OF WATER

Sample No.

PHYSIGAL DESCRIPTION:

1. Collected By:
2. Collected at:
3. Collection: Date Hour
4. Source of Supply: River, lake, spring, well, pool.
5. Kind of Supply: Private, public, school, municipal (community)
6. Sampling point: pump, pressure tank, tap, reservoir, fire hydrant, fountain, spring.
7. Gradient: Depth:
8. County: Township:
9. Post Office
10. Name of owner:
11. Send Results to:
12. Amount of Growth in and around Source:
13. Result: Water supply is:

Acceptable Condemned More samples Necessary

CHEMICAL ANALYSIS:

Test		Results	Acceptable
Color			1
Total Solids			500 mg/liter
Odor	Hot		0-I
	Cold		0-I
Hardness			50-100 PPM
PH			7
Chlorine			0.1 to 0.4 (ppm)

Bacterial Analysis:

Plate Count		300 per CC

Presumptive Test: Durham Fermentation tube

Hours	Gas in Lactose Broth	Negative				
24						
48						
Sample in Ml.	10	1	.1	.01	.001	

Confirmation Test: Coliform Detection.

Violet Red Bile Agar	None

illustrated on page 59, to be used for each sample, may be placed in the kit when one leaves to test the water supply. The inside surface of the front side of the kit may be used as a work table. It is a level surface and will prevent spilling or over-turning of the chemicals while performing tests.

PICTURE IV

As shown in Picture IV, above, a handle has been attached to the outside of the top panel of the box.

The handle, in this location, will facilitate handling and keep the equipment on the inside in an upright position. A hasp has been placed in the center of the top and front surface edges to hold these two panels together while the water testing kit is in transit.

Picture IV shows how compact the water testing kit is when it is ready for use in rendering a public health service. It is easily handled and occupies very little space. The weight of the stocked kit is ten pounds and four ounces. Four hours of labor is required, and the stocked kit costs approximately four dollars ($4.00).

On page 62 is a list of equipment needed to perform all tests outlined or described in this thesis. The approximate cost, to carry out all test required by this study, is listed for supplies needed in the water testing kit and in the school laboratory. The approximate cost includes the purchase of a pressure cooker and a small oven for incubation in the bacterial analysis. However, a fire proof box with a suspended light bulb can be regulated to serve the purpose of an incubator or oven. Because of the lack of funds in most of the science departments of schools, this approximate cost may seem expensive. The approximate cost has included all equipment necessary. Whereas, the flasks, pipettes, etc. may not have to be purchased.

NEEDED MATERIALS

For Water Testing Kit

1. Sample bottles (3)
2. Nitrazine paper and color chart
3. One 250 ml. Erlenmeyer Flask
4. Small Bottle of KI crystals
5. Small Bottle of Conc. HCl
6. Small Bottle of Starch Solution
7. Bottle of 0.001 N Sodium Thiosulfate Solution
8. Pipettes, 10 ml., two 1 ml.

For School Laboratory

1. Six Petri dishes
2. Pressure Cooker
3. Sterile pipettes in case (1 ml. and 10 ml.)
4. Dilution bottles - 4
5. Agar, Peptone, Lactose
6. 3 Durham Fermentation tubes.
7. Ten 100 ml. N essler tubes
8. Three 250 ml. Erlenmeyer Flasks
9. Two 500 ml. Erlenmeyer Flasks
10. Filter paper - standard
11. Litmus paper
12. 100 ml. pipette
13. Funnel
14. Oven (Incubator)
15. Soda Reagent
16. Potassium Chloroplatinate
17. Methyl Orange
18. 0.02 N Sulfuric Acid
19. Burner and gas jet
20. 1000 ml. distilled water
21. Violet Red Bile Agar

Approximate Cost $44.55

TABLE VIII

RESULTS OF SAMPLES

ANALYZED WITH WATER TESTING KIT

Samples	1	2	3	4	5	6	7	8	9	10	12	13	14	15	16	17	18	19	20	21	22
pH	7	6	7	6	6	7	7	7	7	7	6	6	5	5	5	7	6	7	6	8	8
Chlorine Residual	0	0	0	.2	.1	.1	.1	.1	.3	.1	.4	.4	.6	1.0	2.0	.1	.2	.2	.1	.1	.2
Hardness	80	70	152	82	102	100	82	96	72	54	100	84	100	40	37	63	48	50	50	160	210
Color	I	I	I	I	I	5	5	5	I	I	I	I	5	5	5	I	I	I	I	5	5
Odor Hot	0	0	0	0	0	I	I	I	I	0	I	0	VCs	VCs	VCs	I	0	0	I	VE	VE
Odor Cold	0	0	0	0	0	I	I	I	I	0	I	0	VCs	VCs	VCs	0	0	0	I	VE	VE
Total Solids	250	222	190	252	290	450	525	440	220	272	470	480	950	1578	1612	350	272	382	420	12,000	3,200
Plate Count	200	150	100	180	180	1500	1250	800	100	100	50	200	60	100	180	200	200	250	275	800	3,000
Presumptive	0	0	0	0	0	P	P	P	0	0	0	0	0	0	0	0	0	0	P	P	P
Confirmed Test	0	0	0	0	0	P	P	P	0	0	0	0	0	0	0	0	0	0	P	P	P
Results	A	A	A	A	A	C	C	C	A	A	A	A	C	C	C	A	A	A	C	C	C

P- positive, 0 - negative, A - acceptable, C - condemned.

On page 63, Table VIII, can be found a series of results obtained from samples of water which have been analyzed with the water testing kit. The purpose of this information is to indicate how results can be compiled compactly and the sort of results which can be expected for the various tests as well as for various kinds of water. Of the many samples that have been analyzed with this water testing kit, the 25 samples on page 63 are typical examples of all the other samples.

Samples numbered 6, 7, and 8 were collected from a stream running through a pasture. The high plate count should be noted. Samples numbered 14, 15, 16 were collected from a stream into which industrial wastes were dumped. The results from the odor, pH, and color are indicative of such pollution. Samples numbered 21 and 22 were collected from a stream after three days of rain. The results from the plate count, color and odor are typical for such conditions.

CHAPTER IV

HOW THE SCIENCE TEACHER MAY USE THE WATER TESTING KIT

A water testing kit can be of great value to the teacher, the school, and the community. The science teacher in any school has vast amounts of work included in the daily schedule. However, the water testing kit can serve to supplement the regular class work. Testing water can also become a hobby for the teacher. Nevertheless, such a kit can serve to increase the public relations factor between a school and its community. Public relations is one of the most powerful forces in any community and therefore, can not be overlooked (Zwoll, 1948). By improving the sanitary conditions in the community, the patrons have a greater respect for the school and the science teacher. It can establish the science teacher as the source of aid in case of sanitary troubles. Schools have the responsibility of serving the people of the local school district through a constant orientation of institutional activities to the particular problems and needs of the community and through a continuous program of information which will interpret the school and its activities to the people. This dual activity is part of school public relations or social interpretation. Frequently the people are not sufficiently conscious of their own needs, problems, or

powers to act; and responsibility does not rest solely with the pople or even with their immediate representatives to whom they have delegated certain powers. Responsibility for an effective operating, dynamic school also rests with the professional agents at the school (Zwoll, 1948).

The public relations responsibilities of the school employee are basically the same as those of the public. As both a member of the community and as a school employee, each has the unequivocal obligation to interpret the schools to the community. His position with the schools place him in a particularly advantageous position with respect to this phase of a school's public relations program. In this capacity, the school employee serves to bring to the attention of the school authorities the attitudes, opinions, problems, and needs of the community as these factors come to his attention (Zwoll, 1948).

The outlook for such a kit is very bright and encouraging. It is quite possible that such a kit can be integrated into a food testing kit to check food for contamination. Each year numbers of incidences hit the newspapers of people suffering from food poisonings of various kinds. In various schools there are outbreaks of food poisoning from food served in local cafeterias. A routine check of water in schools and the detection

of food which has become contaminated in cafeterias is the best use to which a kit of this sort can be applied.

Teachers may use this kit advantageously to account for individual differences in the classroom. The more interested and challenging students may find delight in making routine checks of water in the school. The teacher may use the project method to challenge the better students to build a similar water testing kit for their own use or use in the school. Child study naturally leads to "doing what that study shows to be desirable and necessary." Obviously a guidance-directed curriculum should provide each child with a series of experiences through which he can discover, without undue discouragement, his strengths and his limitations and can gain knowledge and skill in meeting the problem of school and of life. By dealing with problems of daily living, appropriate to his capacity and need, he learns to live in his environment and improve it. Thus the present and the past contribute to developing personality of each child. Especially rich in value are those social situations through which the child acquires a sense of belonging to the group and of working for common worthy purposes and the good of all concerned.

This kit offers an excellent opportunity for teach-

ing the Scientific Method. The Scientific Method or approach to problems, must be learned to properly evaluate our experiments and research in science. However, that is not the ultimate goal. It is desired that the student might apply the Scientific Method to his own daily problems as well as in evaluating things that are seen and read. In establishing truth from untruth, through the scientific method and attitudes, the student is more capable of becoming a well-adjusted personality.

The Boy Scouts of America organization has been increasing at the rate of 500,000 new memberships each year since 1935 to a total of approximately 15,000,000 memberships at present (Boy Scouts of America, 1940). The Girl Scouts organization has been increasing at the rate of 70,000 new memberships each year since 1935 to a present total of approximately 573,255 memberships (Girl Scouts, Inc., 1941). Boys and girls who are members of Scout troops in the community may be challenged to design such a kit for their use on camping trips.

Another purpose seen would be making the students more community minded and having more appreciation for the work of our health departments and their services to man. The necessary sanitation for proper healthful living should be the ultimate goal expected to be instilled within each student. Likewise, the teacher will likely

become concerned with the welfare of the community and can stress proper cleanliness to the students. The detrimental effects of impure water will become vivid in the minds of the students who participate in the testing procedure. Personal hygiene deals with the facts and principles that enable the individual to live at his best. It includes not only the prevention of disease in the person but also the achievement of best standards of living possible for him. To this end, the sanitation of the home for which the individual is responsible may greatly contribute; or public health conditions, such as pure water supply, may influence profoundly the quality of his health. On the other hand, the practice of sanitation in a community is a reflection of the desires of the people of the community to have wholesome conditions; and hence all measures of public health are dependent upon the will of the persons of the social group or groups concerned (Williams, 1928).

Progress made in the public health sciences during the past half century has made it clear that the heavy burdens of disease can, in large measure, be lifted by the application of scientific knowledge already available; and each year the results of public health research are broadening the areas of possible control.

The most dramatic results have been attained in the sphere of environmental sanitation, particularly through improvement in water supply and waste disposal and in the control of the arthropod vectors of disease.

A substantial part of the reduction of mortality rates in Western Europe and North America has been due to purification of water supplies and improvement in the disposition of fecal wastes. In these areas, not only cholera and typhoid fever, but also the various forms of dysentery, have almost disappeared; and the rare cases of intestinal infection which occur are due to personal contact or food-handling by healthy carriers (Winslow, 1951).

In Latin America striking results have been achieved in various countries in cooperation with the national health agencies and the Institute of Inter-American Affairs. Recently, in the Amazon Valley of Brazil there was an area two-thirds the size of the United States in which only two cities had adequate and safe public water supplies. The incidence of typhoid fever and other water-borne diseases was extremely high, as was the infant mortality rate and intestinal parasite infection rate. One of the most important parts of the engineering and sanitation phase of the work has been the

construction of safe public water supplies in the smaller communities, ranging in size from 500 to 10,000 inhabitants. In one of these towns, where there had been 20-30 cases of typhoid fever each year, not a single case occured after the installation of a small and economical water-supply. Water supplies and storage services in cities of Mexico considered that 22% of their general mortality rate was caused by waterborne diseases. In 1940 less than 1% of the cities and towns in Mexico had water supply systems and only five of these cities and towns had really potable water (Winslow, 1951).

Mankind suffers from many grave preventable diseases which involve not only human suffering but also impose heavy burden on the economic resources of the regions involved. Data quoted from reports on progress in many countries indicate that such conditions can, in large measure, be controlled and that such control has frequently resulted in substantial betterment of general economic status. The realization of this fact should prove of major importance in assisting the efforts of public health officials and other interested personnel.

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APPENDIX

APPENDIX

I. Calculations of Residual Chlorine from the Starch-Iodide Method.

Milliliters of 0.001 N Thiosulfate X 0.1773 equals Parts Per Million of Residual Chlorine.

II. Calculations of Total Hardness from the Soda-Reagent Method.

Example:

Milliliters of 0.02 N Sulfuric Acid (distilled water) . . .	31.2
Milliliters of 0.02N Sulfuric Acid . . .	19.6
Difference	11.6

11.6 X 20 = 232 p. p. m. total hardness as Calcium Carbonate.

III. Calculations of Total Residue.

Example:

Sample portion	No. 1	No. 2
Wt. of dish and residue	48.2982 gms.	43.8646 gms.
Wt. of dish	48.2540 gms.	43.8210 gms.
Wt. of Residue	0.0442 gm.	0.0436 gm.