

PERSULFATE OXIDIZABLE CARBON  
AS AN ENGINEERING MEASURE OF  
ORGANIC POLLUTANTS IN WATER

by

JOHN KEITH CARSWELL

A THESIS

submitted to

OREGON STATE COLLEGE

in partial fulfillment of  
the requirements for the  
degree of

MASTER OF SCIENCE

June 1961

APPROVED:

Redacted for privacy

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Associate Professor of Civil Engineering

In Charge of Major

Redacted for privacy

---

Head of Department of Civil Engineering

Redacted for privacy

---

Chairman of School Graduate Committee

Redacted for privacy

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Dean of Graduate School

Date thesis is presented May 1, 1961

Typed by Mary Adams

## ACKNOWLEDGMENT

The author wishes to acknowledge the financial support and the invaluable assistance received from the following organizations and people: The United States Public Health Service for granting the author Traineeship No. 61-266, to finance his graduate program. The Oregon State College Engineering Experiment Station for the prompt and efficient administration of the trickling filter research project. Dr. Campbell M. Gilmour, Professor of Bacteriology, Oregon State College, for his advice and continued interest in the project. Mrs. Vera Evenson and Mrs. Betty Wullstein, both of the Department of Bacteriology, for assisting in the laboratory analyses. Mr. Thomas L. Miksch, Instructor in Civil Engineering, Oregon State College, for assisting in the laboratory analyses. Mr. Harold Sawyer and Mr. Ralph Carter, Civil Engineering students, for assisting in the preparation of this thesis. Mr. Ray Borrall, Corvallis Sewage Treatment Plant operator, and his employees for their cooperation and assistance throughout the project.

Finally, special acknowledgment and sincere thanks are given to Mr. Frederick J. Burgess, Associate Professor of Civil Engineering, Oregon State College, for suggesting the area of research, obtaining the additional personnel and financial aid, directing the project, supervising the testing program, obtaining equipment and materials, and for being the author's major professor.



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PERSULFATE OXIDIZABLE CARBON  
AS AN ENGINEERING MEASURE  
OF ORGANIC POLLUTANTS IN WATER

INTRODUCTION

Sanitary engineering has as its goal the establishment and maintenance of a healthful environment for man. One of the oldest and most formidable problems to be faced in the achievement of this goal is the effective disposal of the myriad of liquid wastes created as by-products of man's activities in this environment. Although the last one hundred years has seen great improvements and advancements in the field of waste disposal, population growth and increased industrial complexity in our modern society have made necessary the acceleration of research and development of more efficient treatment processes.

The basic tool of any science is that of measurement. Certainly only limited advancement may be made in any scientific endeavor where a means of fast and accurate measurement is lacking. This is particularly true in the field of waste disposal, where large variability of both the quantity and quality of liquid wastes makes accurate methods for their measurement extremely necessary. Of principal concern in the investigation or development of any waste treatment process is the measurement of the organic content of the waste to be treated, and the measurement of the process efficiency in its removal. These two measurements form the basis for the sanitary design and evaluation of any waste treatment process and must be known accurately.

Unfortunately, the development of methods of measurement has

not kept pace with the development and improvement of waste treatment processes. Existing methods for determination of waste strength and process efficiency are inadequate in many instances, and, indeed, have been proven nearly useless when applied to many complex industrial wastes. Because of inadequate yardsticks of measurement it is reasonable to believe that existing treatment units are being operated without the accurate knowledge of process efficiency necessary to determine if they are solving the disposal problem they were established to eliminate. Finally, the complexity of the modern waste disposal problem has made necessary an increase in knowledge of the biochemical nature of waste treatment. Here, the development of new and better methods of waste composition measurement are mandatory to the progress of needed research since many of the existing methods fail to make the fine distinctions necessary in understanding the biochemical relationships.

It is with the intent of developing and evaluating a new and promising method of waste strength measurement that the following discussion is presented. It is believed that the knowledge presented will contribute to the solution of the measurement problem.

#### Object and Scope of this Thesis

The objectives of this thesis are twofold: namely, (a) the initial engineering application of the persulfate oxidation test for organic carbon in waste water, and (b) to further investigate the relationship between persulfate oxidizable carbon and biochemical oxygen demand. It is to be noted that the original

adaptation of the test from the literature and the initial development of the persulfate oxidizable carbon-biochemical oxygen demand relationship was the work of Mr. Lorus L. Purkerson, graduate student in the Department of Bacteriology at Oregon State College. This initial stage of the test's development was conducted as Project 220, "Chemical and Physical Aspects of Deep Trickling Filters", of the Engineering Experiment Station. The author was privileged to work with Mr. Purkerson throughout the initial development period as a laboratory assistant, and thus became familiar with the test prior to making its initial engineering application. Since a graduate thesis is currently in preparation by Mr. Purkerson covering his work in the initial development and adaptation of the test, this thesis will be limited in scope to all work done with the test subsequent to that of Mr. Purkerson. Reference will be made to his findings only as is necessary to establish the basis and background for the author's work.

#### Method of Study

The initial engineering application of the test was accomplished through its use as a means of evaluating the organic loading and removal efficiency of an experimental deep trickling filter. Following an evaluation of the data obtained from this study, it was determined that additional work was needed to permit a more exact definition of the persulfate oxidizable carbon-biochemical oxygen demand relationship. This would permit a more useful interpretation



of the persulfate oxidation test's results. To this end, laboratory studies were conducted with emphasis placed on the variation with time of the carbon and biochemical oxygen demand parameters common to the relationship.

## THE PERSULFATE OXIDATION TEST

### History and Previous Studies

The earliest reference found in the literature to persulfate oxidation of carbon is the work of Franz and Lutze who utilized potassium persulfate in the quantitative oxidation of dilute solutions of certain organic compounds (3, p. 2659). Their method was later modified by Osburn and Werkman for the determination of carbon in fermented liquors (6, p. 421-423). In turn, the latter method was further modified by Katz, et al. for use in the radioassay of carbon-14-labeled compounds (5, p. 1503-1504). It was this method of Katz that was subsequently adapted by Purkerson to the measurement of organic pollutants in water. Prior to the work of Purkerson, no previous studies on this particular application of the test had been conducted.

### Initial Adaptation for the Analysis of Waste Waters

As determined by Osburn and Werkman, and also by Katz, et al., the persulfate oxidation test can be used only for water soluble organic compounds. Since it is generally assumed that the first stage biochemical oxygen demand of a waste water is due to decomposable carbonaceous materials in the waste, it logically follows that direct measurement of the carbonaceous matter can be used to determine the first stage demand. More specifically, it was thought that a measure of the carbon content of a waste water through the use of the persulfate oxidation test could be directly related to

the results of the standard five day, 20° C., biochemical oxygen demand, or BOD test. This latter test is the accepted method of measurement of the strength of organic pollutants.

To test the relation presumed above, a series of tests were initiated by Purkerson in which known amounts of organic substrates common to waste water were analyzed both individually and in mixtures by the persulfate oxidation and standard BOD methods. The successful results of this test series prompted a second series using various wastes of unknown carbon content, including samples of domestic sewage, bean canning waste, slaughterhouse waste, and others. Results of this series established the first relationship between persulfate oxidizable carbon and five day BOD for domestic sewage. At this point, it was considered that the test adaptation was sufficiently developed to warrant an actual research application.

#### Test Procedure

Step 1: Place 30 ml of the sample to be analyzed in the special oxidation flask (see Figure 1). Add, in order, 1 ml of 5N  $\text{H}_2\text{SO}_4$  and 2 ml of 4%  $\text{AgNO}_3$ . Swirl gently to mix the contents.

Step 2: Add, by use of a short funnel, 1 gram of potassium persulfate,  $\text{K}_2\text{S}_2\text{O}_8$ . Immediately fill a center well vial (see Figure 1) with  $\text{CO}_2$ -free 5N NaOH and place in the center well (see Figure 2).

Step 3: Fit a rubber serum cap (see Figure 1) to the flask and evacuate (see Figure 3). Swirl gently to distribute the



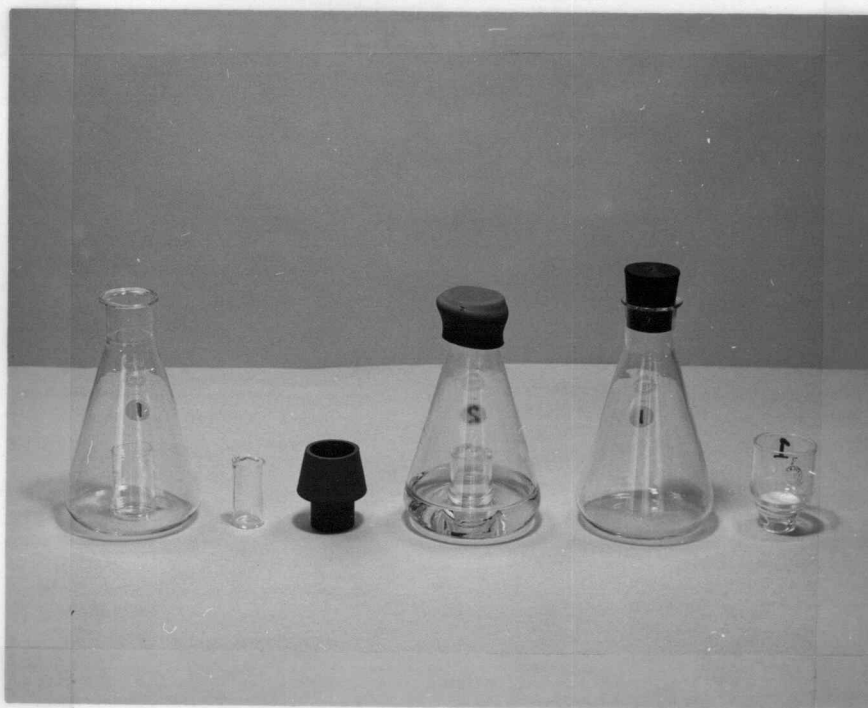


Figure 1. Special glassware for the persulfate oxidation test. Left to right: reaction flask, center well vial, serum cap, assembled reaction flask, precipitation flask, sintered glass filter.

persulfate. Place in a 75° C. oven for 2 hours to effect the oxidation.

Step 4: Remove flask from the oven, cool, and remove the center well vial. Wash the contents of the vial quantitatively to a 125 ml Erlenmeyer flask using approximately 40 ml of CO<sub>2</sub>-free distilled water.

Step 5: Add 5 ml of precipitate mixture (122.2 grams BaCl<sub>2</sub> • 2H<sub>2</sub>O plus 53.0 grams NH<sub>4</sub>Cl in one liter of CO<sub>2</sub>-free distilled water), and stopper the flask without agitation.

Step 6: Filter the flask contents quantitatively through a

tared, ultra-fine sintered glass filter (see Figures 1 and 4). Dry filter at  $102^{\circ}\text{C}$ . for 1 hour, cool, and weigh.

Step 7: Calculate the weight of carbon per unit volume of the original sample (see sample calculation in the appendix).

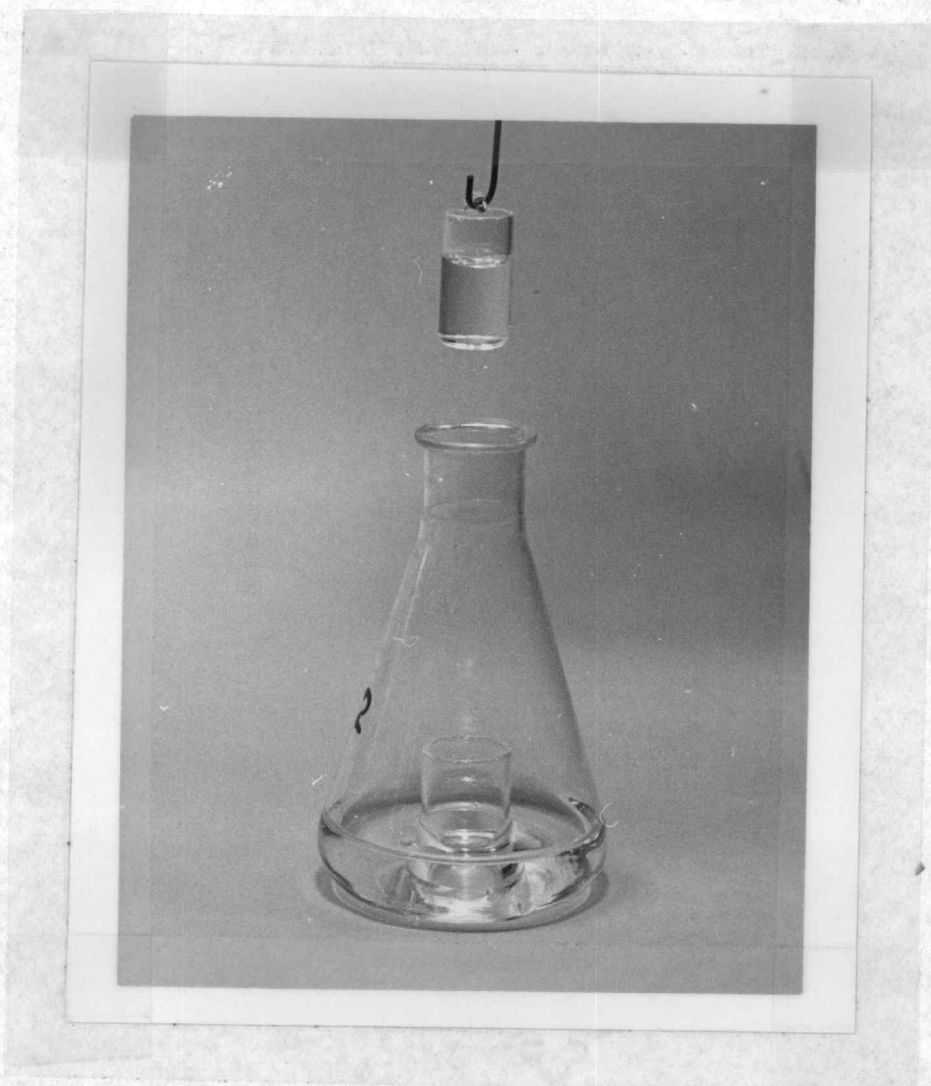


Figure 2. Method of insertion and removal of the center well vial.

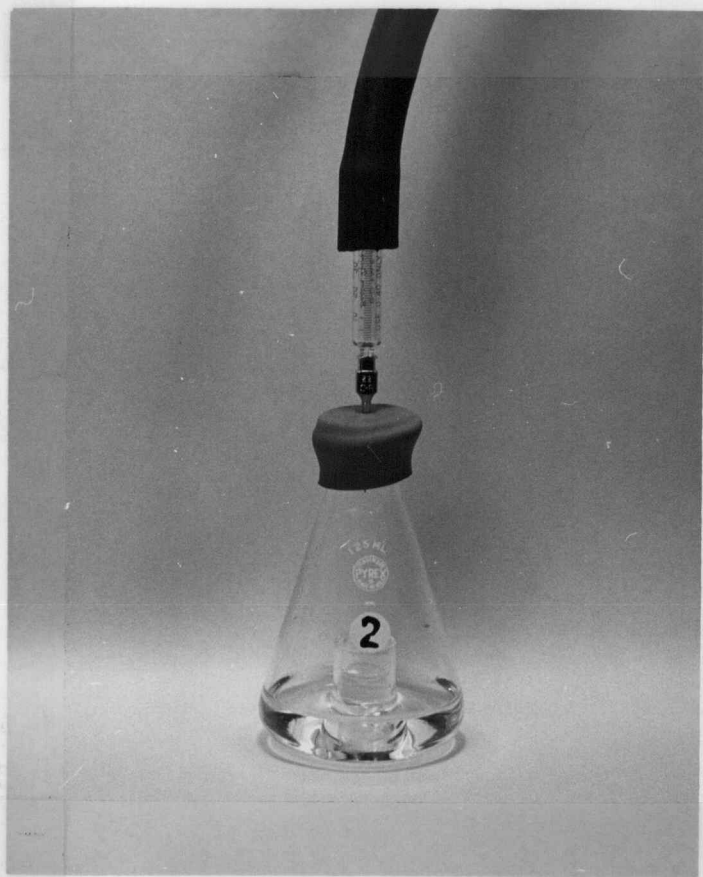


Figure 3. Method of evacuation of the reaction flask.

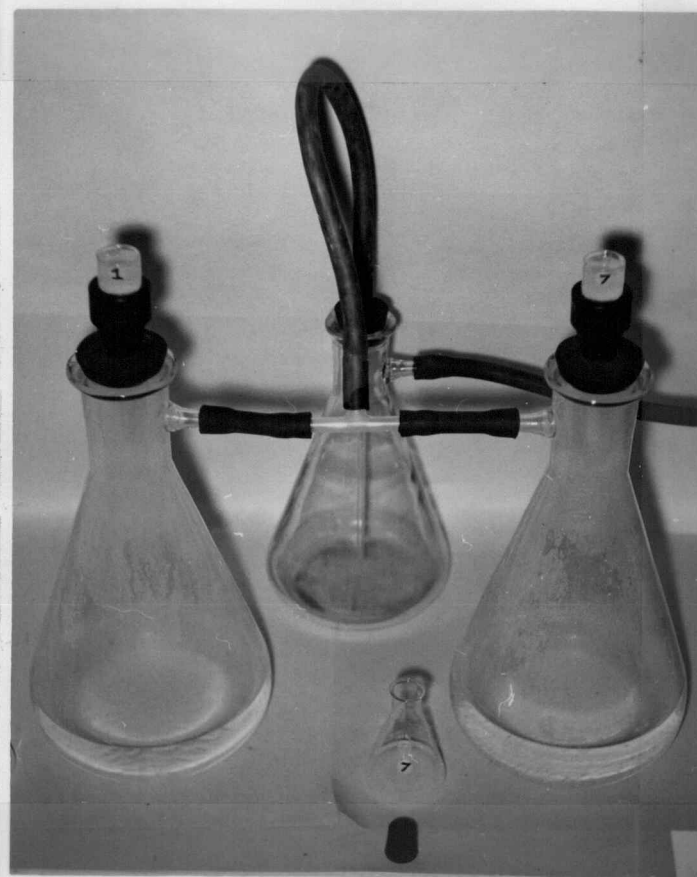


Figure 4. Filtration apparatus.



### Discussion of the Test Procedure

The measurement of the waste sample into the oxidation flask is the only critical volumetric measurement of the test. This step is normally performed using a delivery pipette with an enlarged tip to permit passage of particulate matter.

The addition of the acid is necessary to destroy any carbonate or bicarbonate alkalinity present which would contribute to the  $\text{CO}_2$  production. Neither the acid nor the silver nitrate need be measured with great accuracy as their prescribed volumes are only for the purpose of providing them in excess.

Measurement of the required amount of potassium persulfate may be accomplished with sufficient accuracy through the use of a torsion balance. Since an excess of persulfate is required in the reaction, the accuracy of the measurement is not critical and weighing may be done quite rapidly. The measured portions of salt should be stored in closed vials prior to their use.

It is important that the 5N NaOH ( $\text{CO}_2$ -free) be stored in a container having an efficient air trap to prevent absorption of  $\text{CO}_2$  from the atmosphere. While the amount of NaOH placed in the center well vials is not critical, sufficient freeboard must be maintained in the vial to prevent creep of the absorbent and allow for thermal expansion.

The evacuation of the flask provides for the removal of any free  $\text{CO}_2$  remaining in the sample and effects a tight seal of the flask. Wetting of the serum cap prior to insertion also aids in

providing a tight seal.

The temperature of the oven should be carefully controlled at 75° C. Temperatures in excess of this have caused the serum caps to be blown from the flasks. It will be noted that as the sample-reagent mixture warms in the oven it will turn quite black. This is due to the formation of silver peroxide. Upon further heating this deposit dissolves and the solution becomes clear, indicating the completion of oxidation.

When washing the cooled contents of the center well vial into the flask for precipitation, it is important to use the utmost care to perform the transfer quantitatively. Only distilled water which has been thoroughly boiled to remove CO<sub>2</sub> and cooled in air trapped containers should be used.

It is also important to add the precipitate mixture to the vial washings immediately to prevent absorption of atmospheric CO<sub>2</sub>. The precipitation flasks should be stoppered immediately without agitation which would promote additional gas transfer.

In a test series where many samples are being analyzed, it has been found very convenient to perform all weighings of the sintered glass filters on an analytical balance of the single pan direct reading type. Since four place accuracy is required to accurately determine the carbon content of the sample, the time saving by use of such a device is significant.

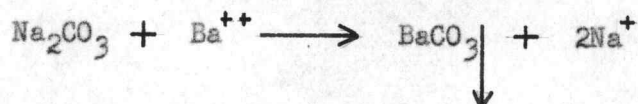
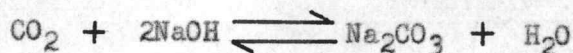
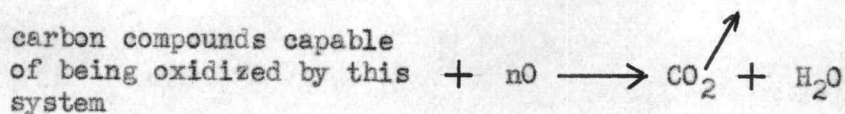
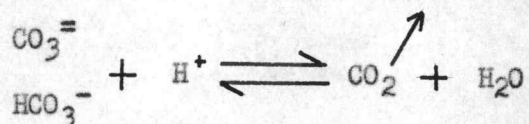
When washing the precipitate into the filters, quantitative care must be exercised. Normally, three washings, each of ten ml

of CO<sub>2</sub>-free distilled water, is considered sufficient. Recovery tests made by Purkerson on known compounds indicate that loss of precipitate through the filters is insignificant.

Every test series should be accompanied by a triplicate reagent blank using the same batch of CO<sub>2</sub>-free distilled water to be used in the analysis. Unusually high blank values (greater than 0.02 mg Carbon/ml) should be eliminated.

#### Chemistry of the Method

The chemistry of the persulfate oxidation reaction must be expressed in qualitative form when the test is applied to waste water analysis, because of the variable and complex nature of such substrates. The basic reactions below are expressed in the order of the addition of reagents.





## THE STANDARD BIOCHEMICAL OXYGEN DEMAND TEST

### Standard Method of Analysis

The biochemical oxygen demand, or BOD, test is the currently accepted method for determining the organic strength of a waste. The results of the test are expressed in terms of the weight of oxygen required to meet the aerobic decomposition requirements of the waste under controlled laboratory conditions. Essentially, the test consists of diluting a sample of the waste to be tested with an aerated dilution water, determining the dissolved oxygen content of the mixture, and after an incubation period (usually five days at 20° C.) once again determining the dissolved oxygen remaining in the mixture. From the amount of oxygen utilized during the incubation period, the BOD of the waste may be calculated. The reagents, special apparatus, and procedure for this test have been standardized (1, p.318-324). Figure 5 shows typical laboratory apparatus for BOD analysis.

### Physical Aspects of the Test

The BOD of a waste water is customarily divided into stages, each stage representing the aerobic decomposition of a different type of material. The first stage is largely attributed to the oxidation of carbonaceous matter, while the second stage represents the decomposition of nitrogenous substances. A typical curve of BOD exerted versus time is illustrated in Plate 1.

The first stage of the BOD has been subject to much mathematical

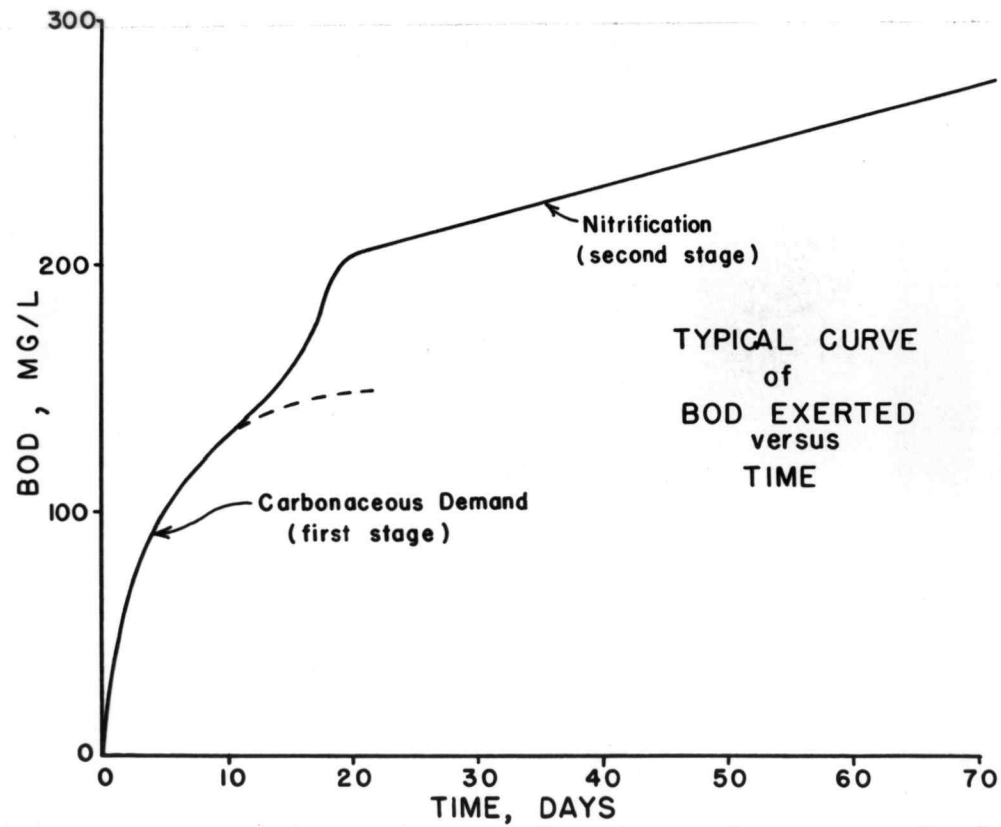


Plate 1. Typical curve of BOD exerted versus time.

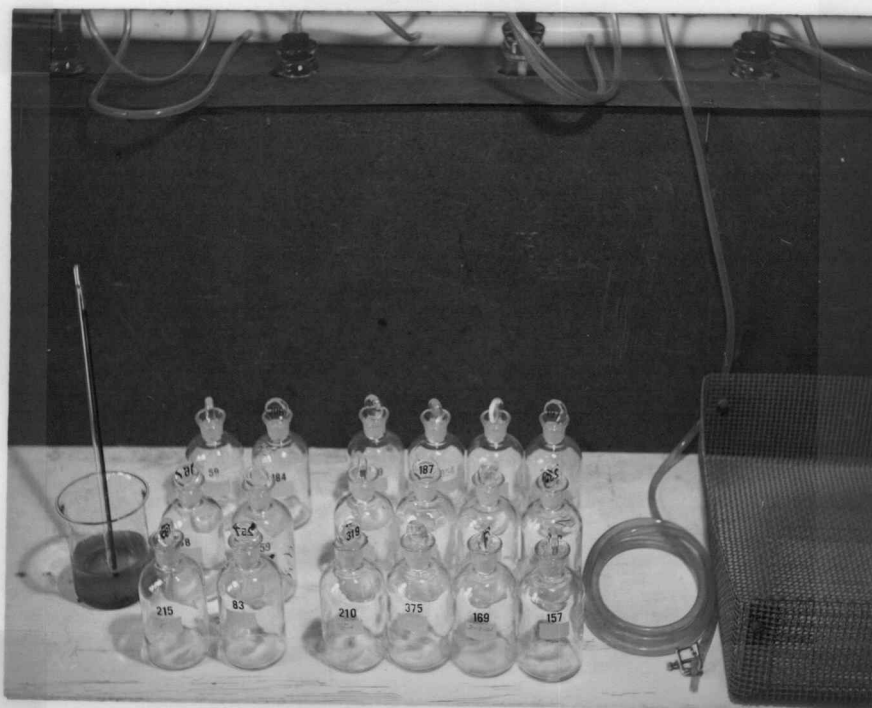


Figure 5. Typical laboratory apparatus for BOD analysis.

formulation. It is usually interpreted as a first order chemical reaction, although formulations have been proposed assuming a second order reaction. Following the classical development of Phelps, the first stage first order equation may be formulated in the following manner (9, p. 5):

$$\frac{dL}{dt} = -KL$$

Which, when integrated becomes

$$y = L (1 - 10^{-kt})$$

Where:

$y$  = BOD at any time,  $t$



Where:

$L$  = ultimate first stage BOD

$k$  = reaction velocity constant

In the above equation, the variables  $y$  and  $t$  are readily obtained from the laboratory data. The parameters  $k$  and  $L$  are more difficult to determine since there is but one equation having two unknowns. Various graphical and statistical methods have been suggested in the literature for the determination of the  $k$  and  $L$  values. The graphical method proposed by Thomas was used to analyze the BOD data presented in this thesis, since it is most suitable for data reported on non-successive days of the BOD incubation period (10, p. 123-124).

The determination of the rate constant is extremely important in the evaluation of any BOD data. This rate constant, which has the units  $1/\text{days}$ , is a function both of the temperature of the reaction and the nature of the waste itself. Numerous investigations have shown that the magnitude of the rate constant is quite variable, even among wastes of the same general type. As noted by Fair, the five day,  $20^{\circ}\text{C}$ . BOD value is not in itself a complete measure of waste strength as is normally assumed (2, p. 525). Inspection of the BOD formulation shows that the BOD at any time may become a variable portion of the ultimate first stage BOD, depending on the rate constant used. It is therefore very important when making comparisons of BOD values of different samples to be sure that the rate constants of the samples are identical.

### Advantages and Disadvantages of the Test

The greatest single advantage to the BOD test is its relative simplicity. With a few simple pieces of laboratory apparatus and very little manipulation, much useful data can be obtained as to the strength and polluttional characteristics of a given waste. Because of its simplicity and because of the enormous quantity of research that has been conducted on it, the test has received widespread publicity and general acceptance. The test is not without some rather serious disadvantages however, particularly when application is attempted to situations where a high degree of accuracy is necessary.

A first disadvantage to the standard BOD test is the time required to obtain the test results. An incubation period equal in length of the number of days for which the BOD is desired must be used. In contrast, the time required for analysis by the persulfate test is but a few hours.

As in any volumetric test, certain errors may accrue in the measurement and transfer of liquid. Of a more serious nature is any variation which may occur in the temperature of the incubator. Such a variation may have serious consequences on the accuracy of the results because of the sensitivity of the rate constant to temperature. Both volumetric and temperature errors can be eliminated or lessened through careful manipulation.

Errors basic to the test mechanism itself, and over which little control can be exercised by the analyst, are those of a

biochemical nature. Since it is impossible to achieve equality among the bacterial populations delivered to the various bottles of a test series, the rate of the oxidative reaction, which is dependent on the magnitude of this population, may vary and cause an inherent error. Associated with this bacterial population problem is the lag time, or time required for the reaction to proceed at its normal rate. This lag time is related to the activity of the biological population present in the sample bottle. An error of a different nature is introduced by immediate oxygen demand which may be exerted when the waste has undergone partial decomposition before analysis or contains reducing substances. Such an immediate demand will obviously produce high results unless separated from the true BOD.

Another disadvantage is the matter of interpretation of results as effected by the rate constant. As may be expected, different wastes have different rate constants. Also, wastes of the same general type frequently exhibit widely varying values, even to the point where subsamples of a given sample show a rate constant variation. Because BOD values obtained from samples having different rate constants are not directly comparable, a serious limitation is placed upon the usefulness of the BOD determination. Plate 2 illustrates the effect of a variation in rate constant on the observed BOD when all other variables are held constant.

Although the above disadvantages are quite important with regard to the reliability of the BOD test, one remaining factor



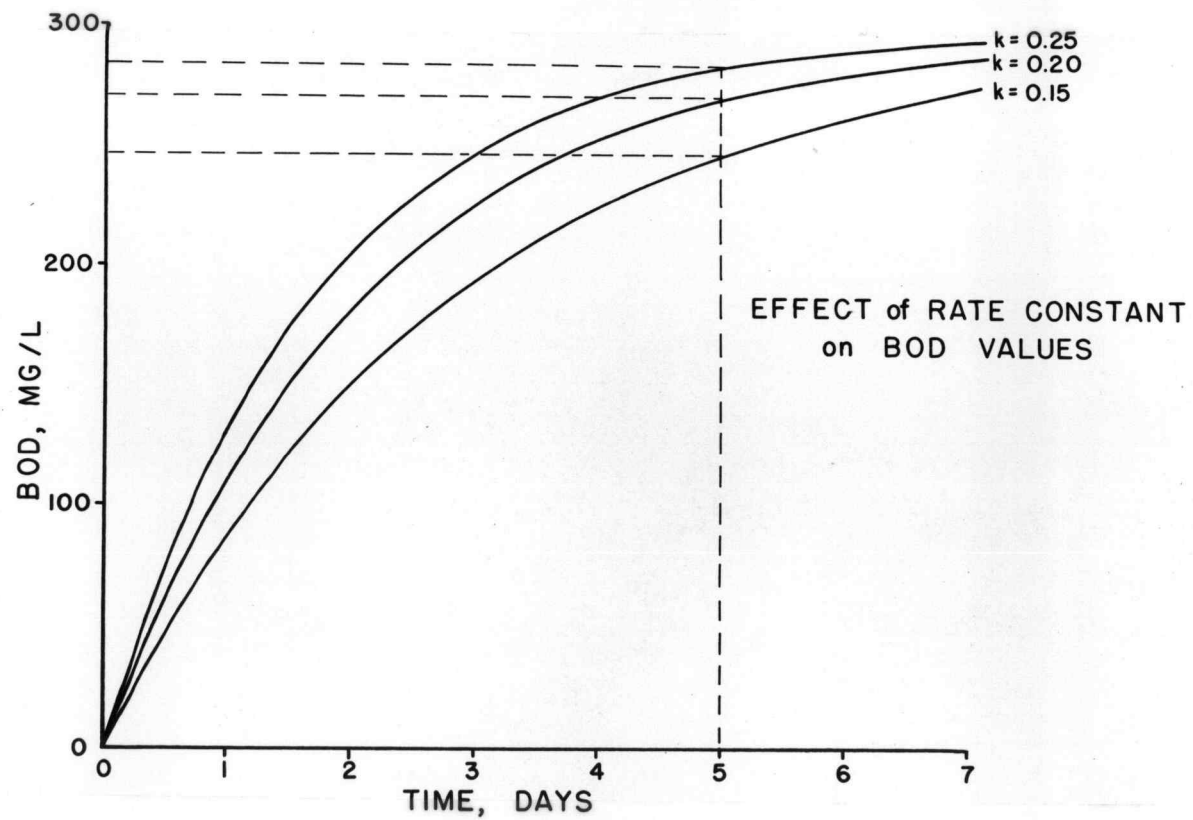


Plate 2. Effect of rate constant on BOD values.

is perhaps the most serious problem. This is the effect of nitrification upon the results of the first stage, or carbonaceous, BOD normally measured. Of principal concern is the time of onset of nitrification as compared to the time of the BOD being measured. Evidence in the literature indicates that for domestic sewage the onset may occur as early as the 1st day of incubation or as late as the 10th day (7, p. 834). That the nitrification stage can effect the results of the desired carbonaceous BOD is illustrated in Plate 3, which shows various times of onset superimposed on a typical first stage demand curve. It becomes readily apparent that high values of first stage BOD may be recorded because of nitrification.

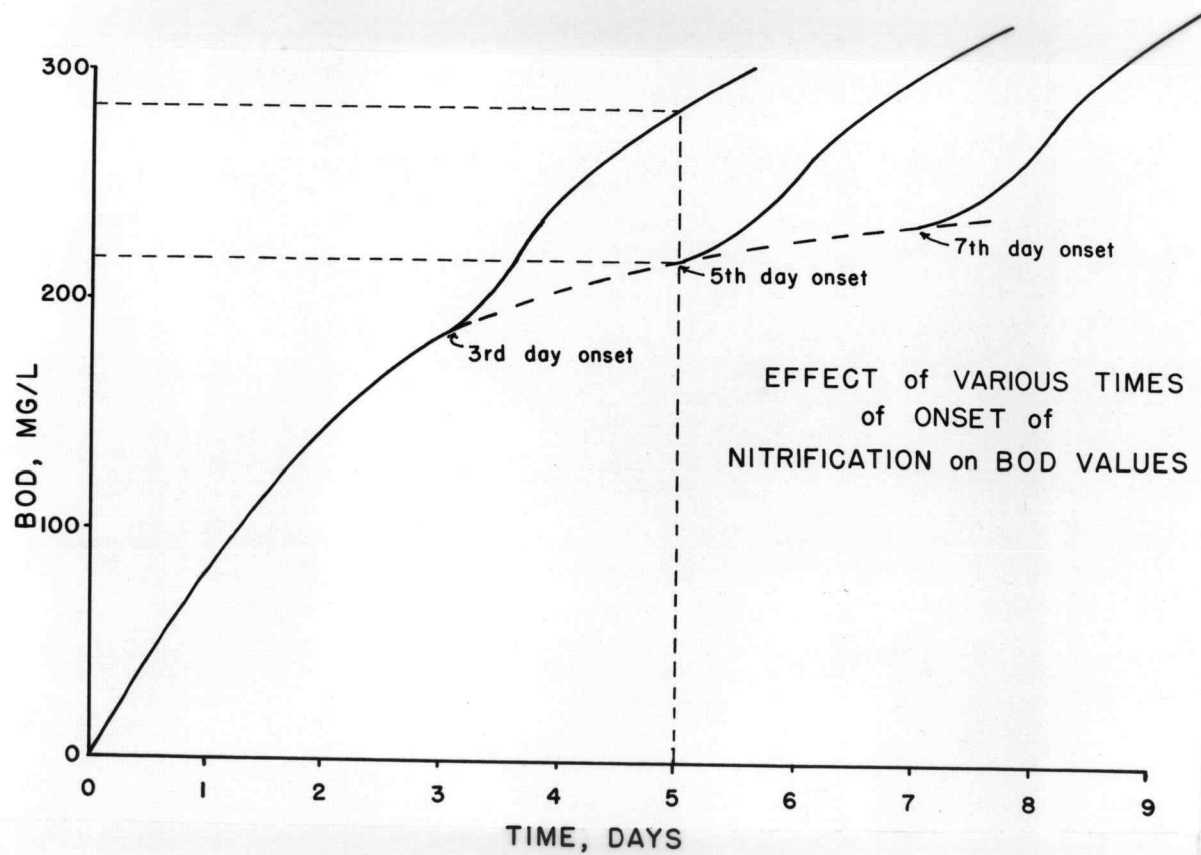


Plate 3. Effect of various times of onset of nitrification on BOD values.



## THE MANOMETRIC BIOCHEMICAL OXYGEN DEMAND TEST

### Method of Analysis

The determination of BOD by a manometric method is not a standard method of analysis for waste water. However, a procedure is offered in the literature which has received some use and tentative approval by the APHA (1, p. 396-399). The principal advantage of the method is that it permits analysis of wastes in much greater concentrations than is possible by the standard BOD method. This advantage becomes important in the study of oxidation rates of strong industrial wastes.

In essence, the method measures directly the oxygen utilization of a waste sample placed in a closed environment, under carefully controlled conditions of temperature and agitation. In the Warburg apparatus, the oxygen uptake is determined by the change in pressure occurring under conditions of constant volume.

### Test Procedure

The test procedure used in this study is essentially that which appears in the literature, with slight modification to meet certain conditions encountered. The reaction flasks, as pictured in Figure 6, were specially made from 250 ml Erlenmeyer flasks with the neck modified to fit the manometer connection. The flasks were also equipped with a center well cup to contain the CO<sub>2</sub> absorbant, and a side arm for the addition of reagents. Normal procedure was to place 50 to 60 ml of the sample in the reaction

flask, add 7 ml of 10% KOH to the center well, attach the flask to the manometer assembly and begin testing. Because of breakage of the manometer arms with the increased weight of the special reaction flasks, the rate of agitation could not be maintained at the 70 oscillations per minute recommended. However, a rate of 24 oscillations per minute was found to be satisfactory.

Determinations were made by stopping the apparatus and reading each manometer to determine the pressure. The system was then opened to the atmosphere for five minutes to insure an adequate supply of oxygen in the flask at all times. Following this exposure period, the manometers were reset, the system closed, and agitation resumed. Calculation of the oxygen uptake was accomplished using the interval uptake method described in the literature (11, p. 9). Figure 7 shows the Warburg apparatus in operation.



ADVANCE BOND

CHILLBROWN Paper

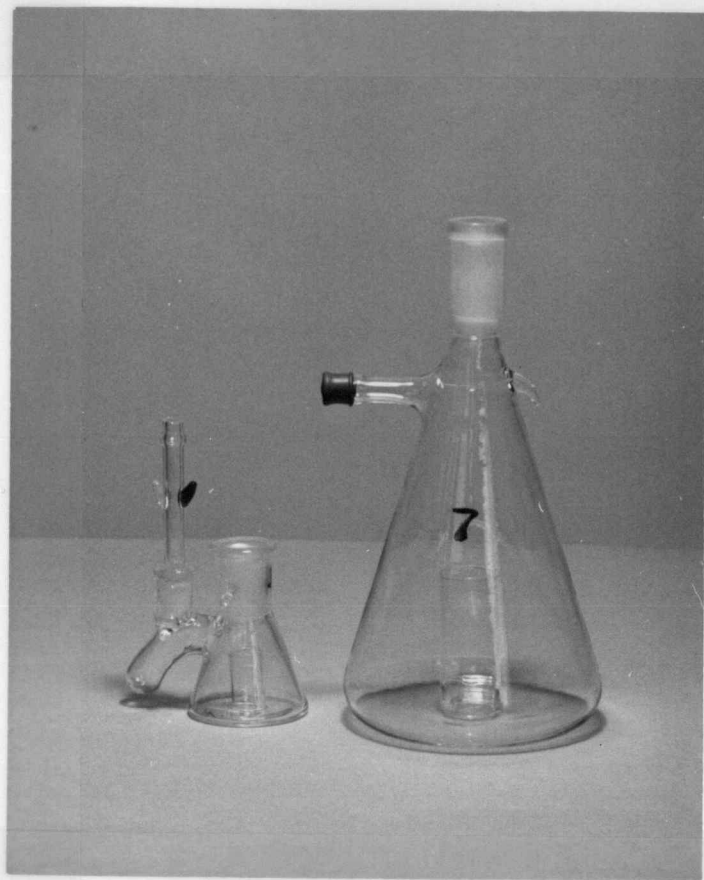


Figure 6. Special large capacity Warburg flask compared to a conventional Warburg flask.



Figure 7. Warburg apparatus.



## THE PERSULFATE OXIDIZABLE CARBON- BIOCHEMICAL OXYGEN DEMAND RELATIONSHIP

### Purkerson's Initial Relationships

Since first stage BOD is largely due to the oxidation of carbonaceous matter, any method which will detect that portion of the total carbon content of a waste liquid which is readily oxidizable should relate to first stage BOD. On this basis, Purkerson initiated studies of such a relationship using the persulfate oxidation test. After first establishing that the recovery efficiency of the test was satisfactory on both individual compounds and mixtures common to domestic sewage, he proceeded to determine what relation might exist between persulfate oxidizable carbon and the five day 20° C. BOD. His method consisted of testing solutions of known carbon content, using a compound or compounds present in domestic sewage, by both the persulfate and five day BOD tests. When the results of these investigations were analyzed, it was shown that a linear relationship existed between the persulfate carbon level and the five day BOD, for any one known solution over the range in carbon level tested. The culmination of this phase of testing was the analysis of a mixture containing fifteen different carbohydrates, fatty acids, and amino acids all common to domestic sewage. Evaluation of the data, which showed a correlation coefficient of 0.998, resulted in the equation:

$$\text{BOD}_5 \text{ at } 20^\circ \text{ C.} = (\text{mg Carbon/ml}) (1807.0)$$

Following the successful determination of relationships based

on known compounds, attention was turned to the analysis of unknown waste samples. Using methods similar to those described above, samples of domestic sewage, cannery wastes, slaughterhouse waste, and paper mill waste were examined. The trends for these unknowns were found to be non-linear. However, in the case of domestic sewage, a good approximation of the relationship could be made using the statistically derived linear equation:

$$\text{BOD}_5 \text{ at } 20^\circ \text{ C.} = (\text{mg Carbon/ml}) (1383.2)$$

The data on which this equation was based also showed a correlation coefficient of 0.998.

#### Application of the Initial Relationship

Following the apparent success of the formulation of the initial persulfate carbon-BOD relationship, it was decided to further examine its merits by subjecting it to an actual research application. Such an application was conducted in conjunction with Engineering Experiment Station Project 220, "Chemical and Physical Aspects of Deep Trickling Filters". This work was undertaken by the author as a part of his research program. Figure 8 shows the experimental trickling filter located at the Corvallis Sewage Treatment plant.

The persulfate carbon test was applied both to the measurement of filter loading rates and the determination of the efficiency of the treatment process. Before critical evaluation could be made of these applications, it was necessary to check the initial

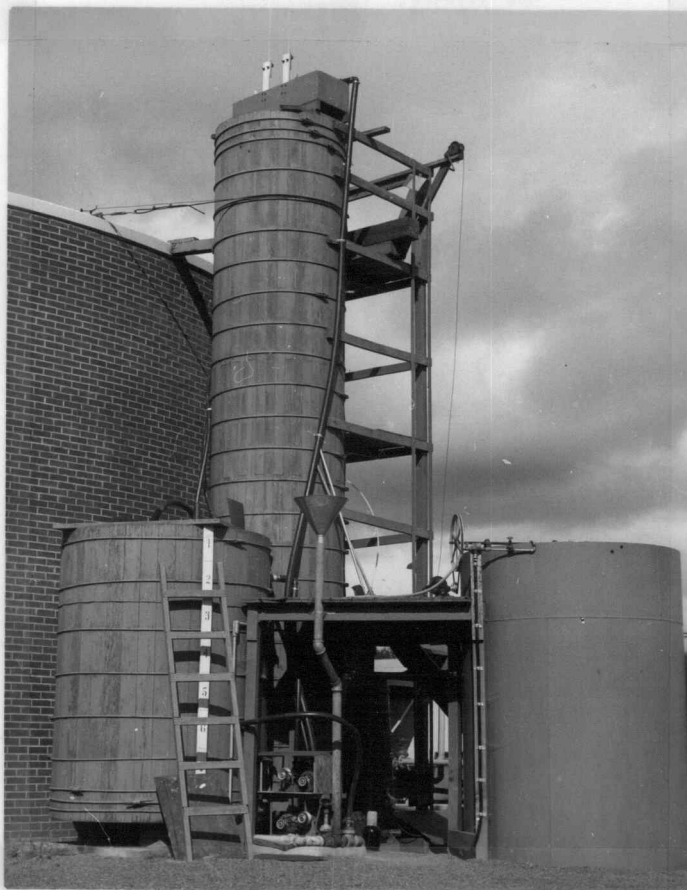


Figure 8. Experimental deep trickling filter unit.

relationship using data obtained from the filter. Statistical analysis of the  $BOD_5$  and persulfate oxidizable carbon values gave the relationship:

$$BOD_5 \text{ at } 20^\circ \text{ C.} = (\text{mg Carbon/l}) (0.835) - 2$$

This relationship is significantly different from that of Purkerson and shows a definite bias factor in favor of the carbon. The data also showed a correlation coefficient of 0.946 and a standard error



of 7.25 mg/liter. A plot of the data appears in Plate 4, which also indicates the 90% confidence limits.

Since twenty day BOD values were determined to permit calculation of rate constant and ultimate BOD values for the various filter test series, a statistical analysis was also made on the data to determine if any correlation with the soluble carbon values existed. The relationship obtained was:

$$\text{BOD}_{20} \text{ at } 20^{\circ} \text{ C.} = (\text{mg Carbon/l}) (2.124) + 79$$

The data showed a correlation coefficient of 0.89 and a standard error of 26 mg/liter. The low value of the correlation coefficient is to be expected as the twenty day BOD measures other oxygen demands in addition to that of carbonaceous material. A plot of the data for the above relationship appears in Plate 5.

In addition to the above direct comparisons of soluble carbon and BOD, a study was made on the possible determination of treatment process efficiency using soluble carbon values. Using  $\text{BOD}_5$  and soluble carbon values for the filter influent and effluent at various rates of hydraulic and organic loading, the statistical analysis yielded the equation:

$$Y = 1.28X - 2.7$$

where Y is the percent removal through the filter based on  $\text{BOD}_5$ , X the percent removal based on soluble carbon, and 2.7 a bias factor. The efficiency data yielded a correlation coefficient of 0.905 and a standard error of 5.8% based on the BOD values. A plot of the data appears in Plate 6.

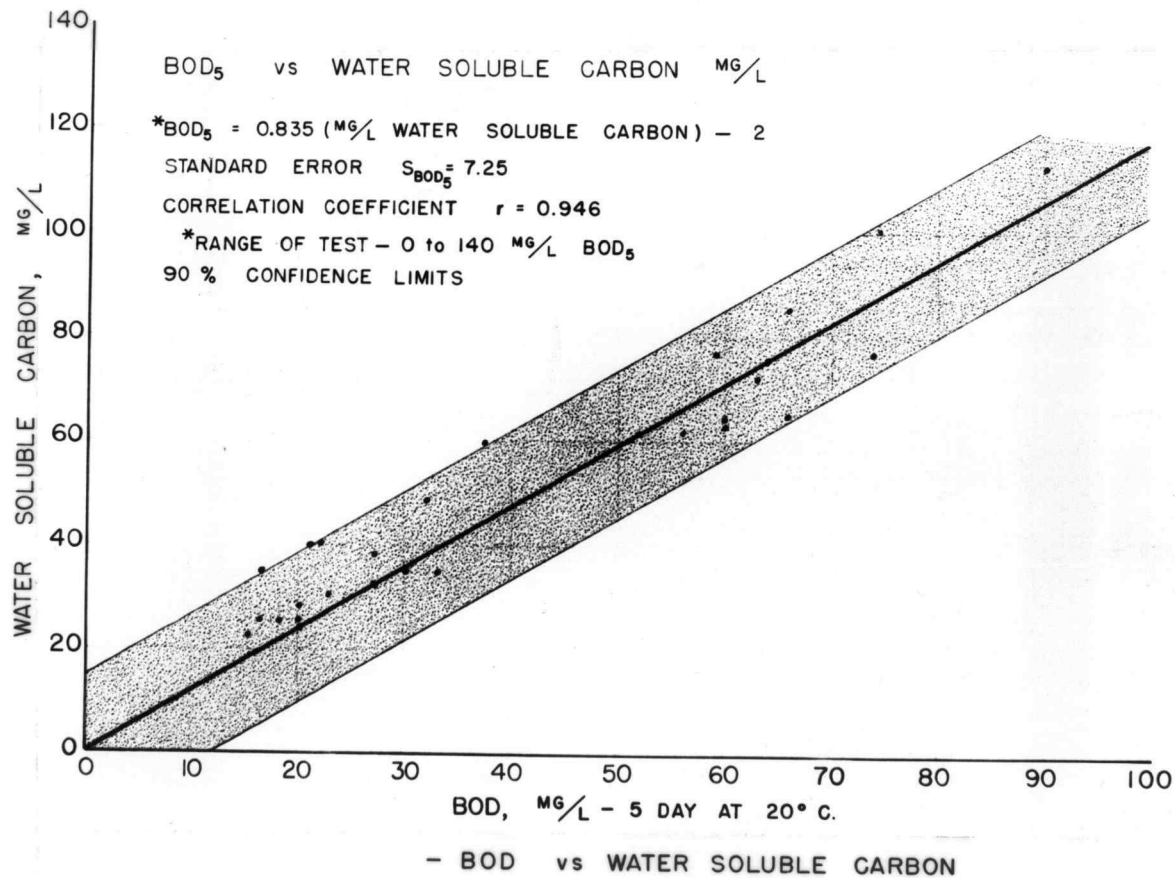


Plate 4. BOD<sub>5</sub> versus water soluble carbon (filter data).

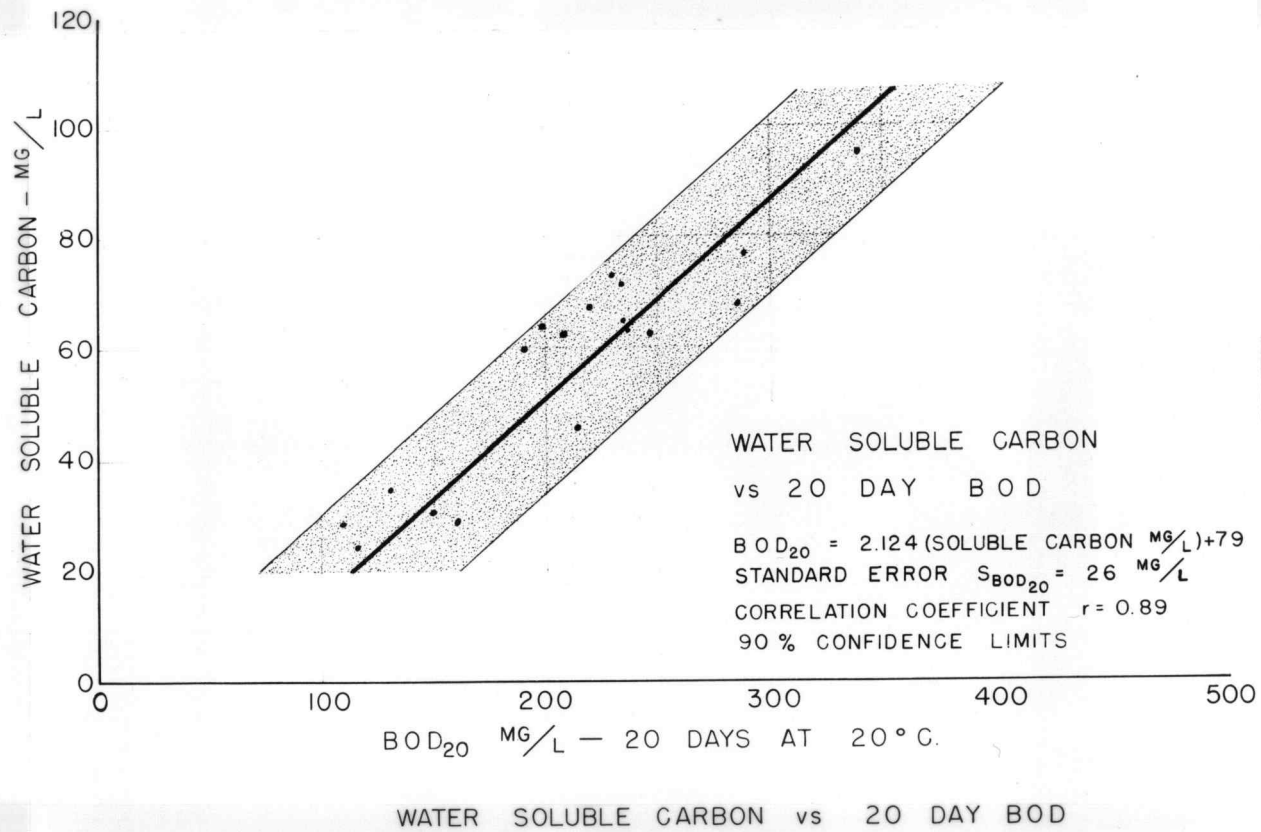
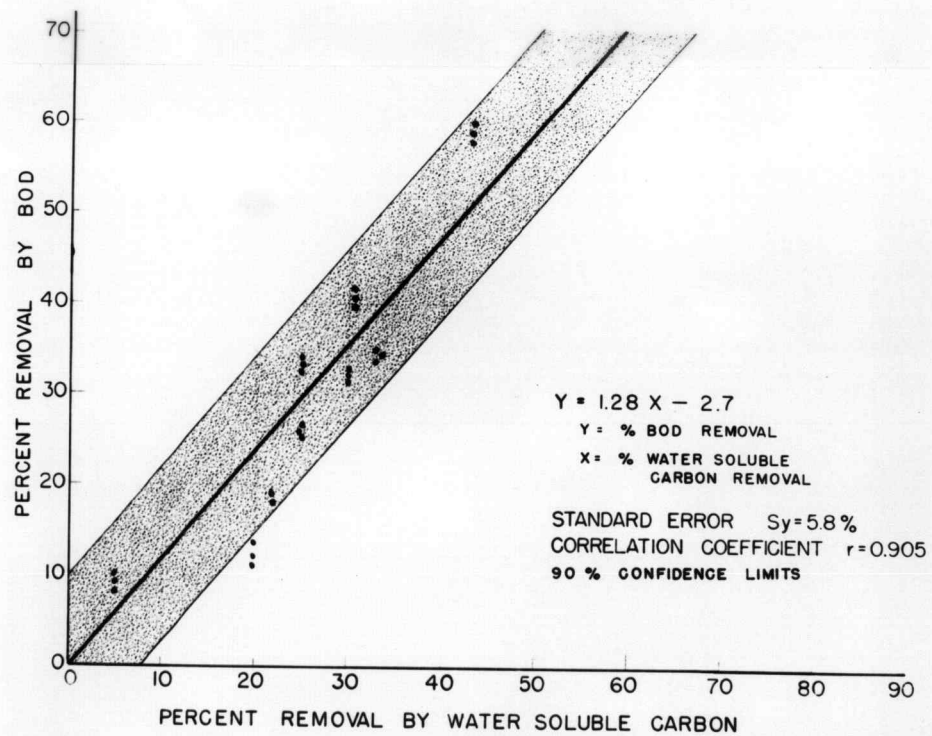


Plate 5. BOD<sub>20</sub> versus water soluble carbon (filter data).





PERCENT REMOVAL OF WATER SOLUBLE CARBON  
 vs PERCENT REMOVAL OF BOD BY FILTER

Plate 6. Percent removal of water soluble carbon versus percent removal of BOD (filter data).

### Discussion of the Results of the Initial Application

Significant differences exist between the soluble carbon-BOD<sub>5</sub> relationships obtained by Purkerson and the field application to the filter data which require additional investigation and explanation. Further analysis of the filter test procedures and data revealed two important factors which at that time had not been considered in the persulfate oxidizable carbon-BOD relationship.

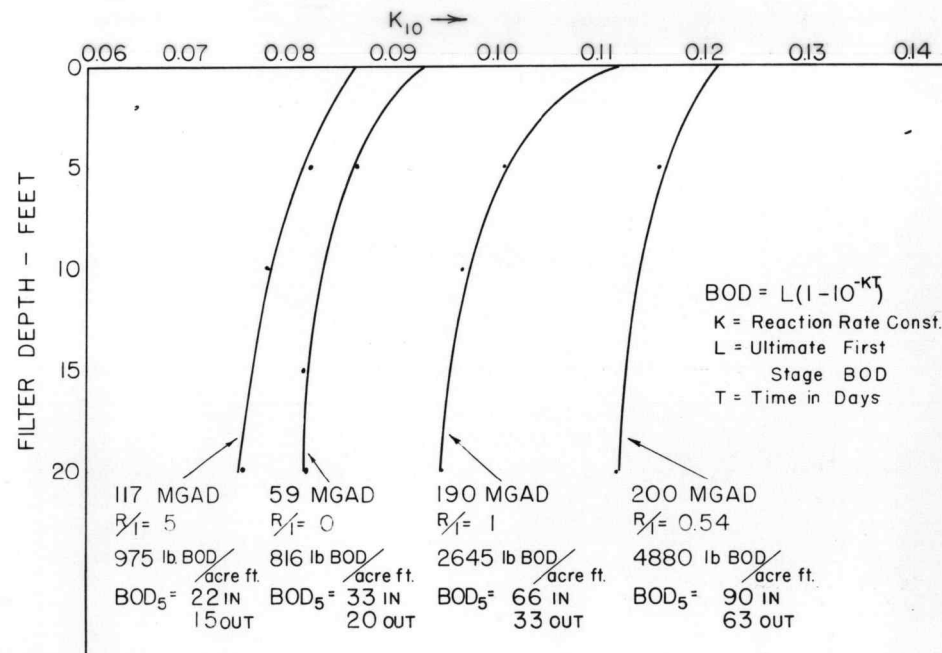
The first of these factors is that of sample pretreatment prior to laboratory analysis. All previous work with domestic and industrial wastes was done on samples that had been filtered through coarse filter paper. The intention was to make the sample more homogeneous and reduce the error inherent in taking aliquot portions of a non-homogeneous solution. Based on the literature, it was believed at that time that the persulfate test would measure only carbon in aqueous solution, and thus such sample pretreatment would have no effect on the final relationship obtained.

It was found during the field application of the test that filtration of the large sample volumes was too time consuming. Further, a search of the literature revealed that such sample pretreatment could have a serious effect on the BOD values (8, p. 779-786). For these reasons, pretreatment of the samples was eliminated from the filter test program. It was concluded that further study was necessary on the effect of sample pretreatment on the soluble carbon-BOD relationship.

The second new factor considered to have an effect on the

relationship was that of the variation in the BOD rate constant values of the samples. Calculation from the filter data showed that for any given rate of hydraulic and organic loading, there was a significant change with depth in the rate constant of the waste passing through the filter. Plate 7 shows this variation for four selected loading rates. Since BOD values based on samples having different rate constants are not directly comparable, the change of rate constant with depth in the filter makes determination of the true efficiency very difficult. It was concluded that more information was necessary as to the variation with time of soluble carbon levels during the BOD incubation period.





BOD<sub>5</sub> - REACTION RATE CONSTANT "K" vs FILTER DEPTH

Plate 7. BOD<sub>5</sub> reaction rate constant versus filter depth.

## FURTHER INVESTIGATION OF THE PERSULFATE OXIDIZABLE CARBON-- BIOCHEMICAL OXYGEN DEMAND RELATIONSHIP

### Effect of Sample Pretreatment

In order to determine the effect of sample pretreatment upon the values obtained by the persulfate oxidation test, the following test series was conducted. Using raw domestic sewage in the analysis, a sample was separated into fractions using the basic separation scheme of Sorrels and Zeller (8, p. 780). A minor modification of the scheme was the use of a one hour settling time in the Imhoff cone, as is standard procedure in the settleable solids test, rather than the 45 minutes used by the authors. Plate 8 shows a diagram of the separation procedure used.

The entire separation procedure was carried out as rapidly as possible to prevent significant sample decomposition. As the various liquid fractions were obtained, they were placed in a 4° C. refrigerator until needed. Immediately upon completion of the separations, duplicate values of the POC level were determined as were triplicate values of the five day BOD. The results of these analyses appear in Table 1.

Letting the values obtained for the original sample represent 100 percent, Table 1 also shows the percent of the original sample value remaining after each successive separation. It is apparent from the data that such separations have about equal effect on either test. Further evidence of this fact may be seen in Plate 9, which shows a plot of the values obtained. The definite linear trend

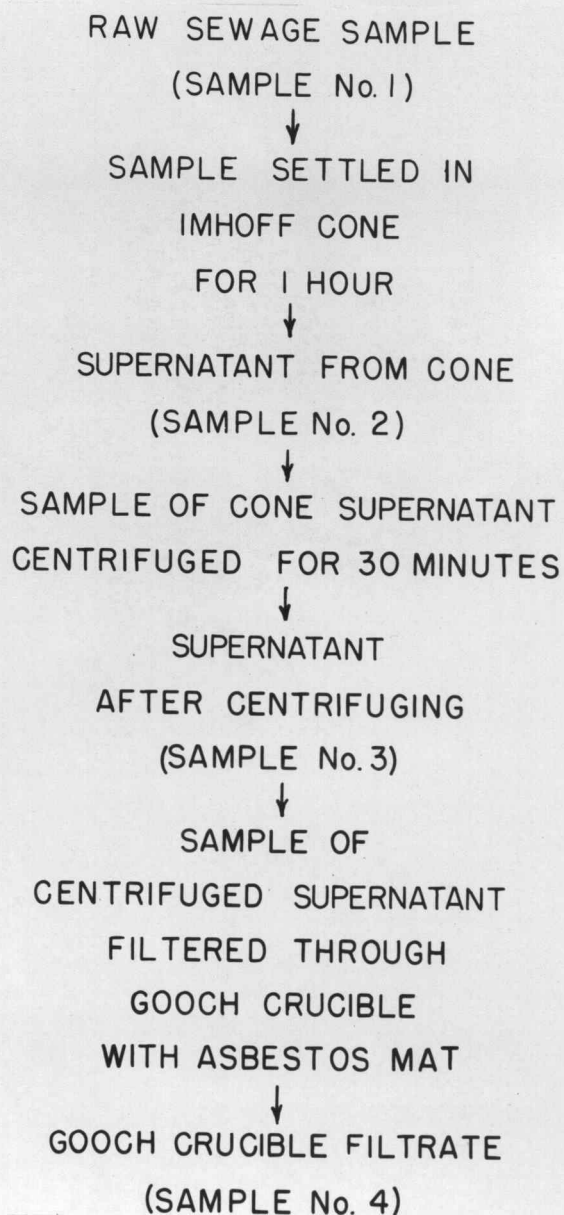


Plate 8. Diagram of separation procedure.



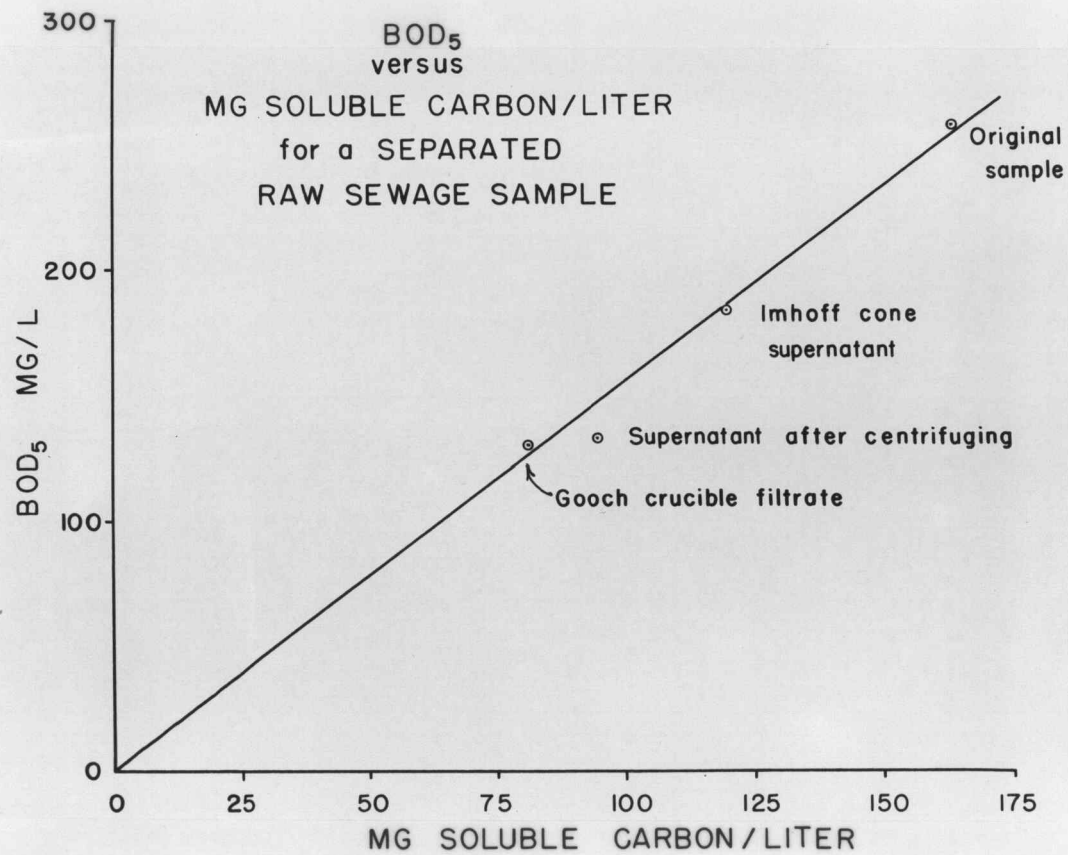


Plate 9. BOD<sub>5</sub> versus mg Carbon per liter for a separated raw sewage sample.

shown by the data illustrates the equal effect of the separations on each test, and further demonstrates the existence of a valid relationship between soluble carbon and five day BOD.

It may also be observed from the data that the persulfate oxidation test measures carbon initially in forms other than that of true aqueous solution. The data shows that as more of the non-soluble fractions of the original sample were removed, the soluble carbon level dropped accordingly. Thus, particulate, suspended, and colloidal compounds of carbon must therefore be oxidized in whole or in part by the persulfate reaction.

Based on analysis of these data, it appears that while sample pretreatment effects the magnitude of the exact values, it has little effect upon the persulfate oxidizable carbon-BOD relationship providing that similar treatment is given to samples for either form of analysis. Since minimizing pretreatment reduces the time spent in laboratory analysis, use of untreated samples would be the best and simplest procedure. Such a procedure would also give values more truly representative of the conditions being measured.

Since the persulfate test measures forms of carbon in addition to those in solution form, the term soluble carbon may lead to misunderstanding when applied to waste water analysis. In its place, the term persulfate oxidizable carbon, abbreviated POC, will be used.

### Time Relationships of BOD and POC

Prior to this phase of the testing program, all attempts to establish a POC-BOD relationship had been based on the measurement of the initial POC level of a waste sample followed by a comparison of this value with the five day BOD determined for the same sample. Because BOD is a time function dependent to a great extent upon the reaction rate constant, it was necessary to determine how the POC level might vary with time during the course of the BOD reaction.

A major obstacle is presented in determining the time variation of POC level by the dilution of 50 to 300 fold of the sample to permit aerobic conditions to obtain in the BOD test at all times during the incubation period. Thus, a sample removed from a BOD bottle for purposes of determination of POC level would contain  $1/50$  to  $1/300$  of the POC level of the undiluted original sample. This reduction is due only to the effects of dilution and is further reduced by utilization of carbon in the BOD reaction. As a result, the amount of POC remaining in the BOD dilution is too small to be measured accurately by the persulfate test.

Since sensitivity of the test did not permit measurement of the POC level in a BOD dilution directly, an attempt was made to concentrate a waste dilution to the volume required for the persulfate test by using a low temperature vacuum condenser of the rotary type. It was found that at  $50^{\circ}\text{C}$ ., two hours was required to concentrate the sample volume from 300 ml to approximately 30 ml.

Comparison of the known amount of carbon in a control sample



to the recovered amount of POC determined by the concentration method showed a recovery of approximately 145 percent. This high value, in addition to the cumbersome nature of the condensation procedure, gave cause for rejection of the method. It was further concluded that excessive decomposition of the sample during testing was promoted by the 50° C. temperature required to attain concentration within a practical time limit.

Determination of BOD by a manometric method was next investigated. It was found that use of the special large capacity flasks with the Warburg apparatus permitted obtaining an undiluted sample at any time during the BOD reaction. While the literature states that an exact correlation does not exist between the results of the Warburg and standard BOD tests, the mechanisms of oxidation are similar in both methods (4, p. 1276). This fact permits the formulation of general conclusions as to the rate of POC uptake during aerobic oxidation.

Based on experimental trials, the following procedure was determined to be satisfactory. Using raw domestic sewage as the test substrate, a 50 ml sample was placed in each of a number of special Warburg flasks equal to the number of days of the BOD reaction to be investigated. Each flask was designated with a day for analysis in the reaction period. Starting all flasks in the Warburg apparatus at the same time, the reactions proceeded until the first 24 hour period had passed. At this point, the flask designated for the first day was removed from the apparatus and a

sample from it analyzed for POC. The manometer readings for this flask determined the one day Warburg BOD. An identical procedure was followed for flasks designated for the remaining days of the test period.

Upon completion of the desired test period, data would then be available on the Warburg BOD and corresponding POC for any day of the period. By assuming that the reactions taking place in the various flasks were essentially identical, a curve could then be plotted of Warburg BOD and POC versus time. Such a curve permits a qualitative knowledge of POC variation with time during the BOD reaction. Determination of the standard BOD of the sample for the same increments of the test period would permit a plot of standard BOD versus time to be added to the POC and Warburg BOD plot. Because of the similarity between the Warburg and standard BOD curves, it could then be implied that a similar trend in POC existed in the BOD bottle as in the Warburg flask. It should be noted that the reliability of this approach is at best questionable and severely limited by problems of experimental measurement.

For the initial test series, a five day test period was chosen. Table 2 shows the results obtained using the described procedure. It will be noted that the POC is reported both as carbon remaining and utilized. The POC remaining is the value obtained by the direct analysis of the Warburg flask contents, while the POC utilized is a calculated value obtained by subtracting the POC remaining at any time from the initial POC level. Since the BOD

curves represent oxygen utilized, it was felt that a more significant comparison to them could be made by plotting POC utilized.

A plot of the BOD and POC values appears in Plate 10. Similar trends of the curves illustrate a relationship exists between the amounts of oxygen and POC utilized at any stage of the reaction. Plate 11 shows the plot of BOD values versus POC utilized. These curves exhibit a strong bias in favor of the BOD values. That the two BOD curves shown are not more nearly identical is explained by the difference existing between the two types of BOD values at any time. This latter fact is presented graphically in Plate 12, which shows a plot of standard BOD versus Warburg BOD.

Before any sound conclusions could be drawn from the data obtained, it was deemed necessary to attempt a duplication of the results of the initial test series and to extend the test period to ten days in an effort to show the variation in POC level under conditions of nitrification in the BOD reaction. Utilizing an analytical procedure similar to that of the first series, the Warburg and standard BOD's and POC utilized were determined for selected intervals of the ten day test period. Table 3 contains the results obtained. A graphical presentation of the data is given in Plate 13, which shows a trend very similar to that of the initial series. The upward trend in the BOD curves after the eighth day is caused by the onset of nitrification.

The upward trend shown by the POC utilized curve after the eighth day was totally unexpected. A basic assumption in the



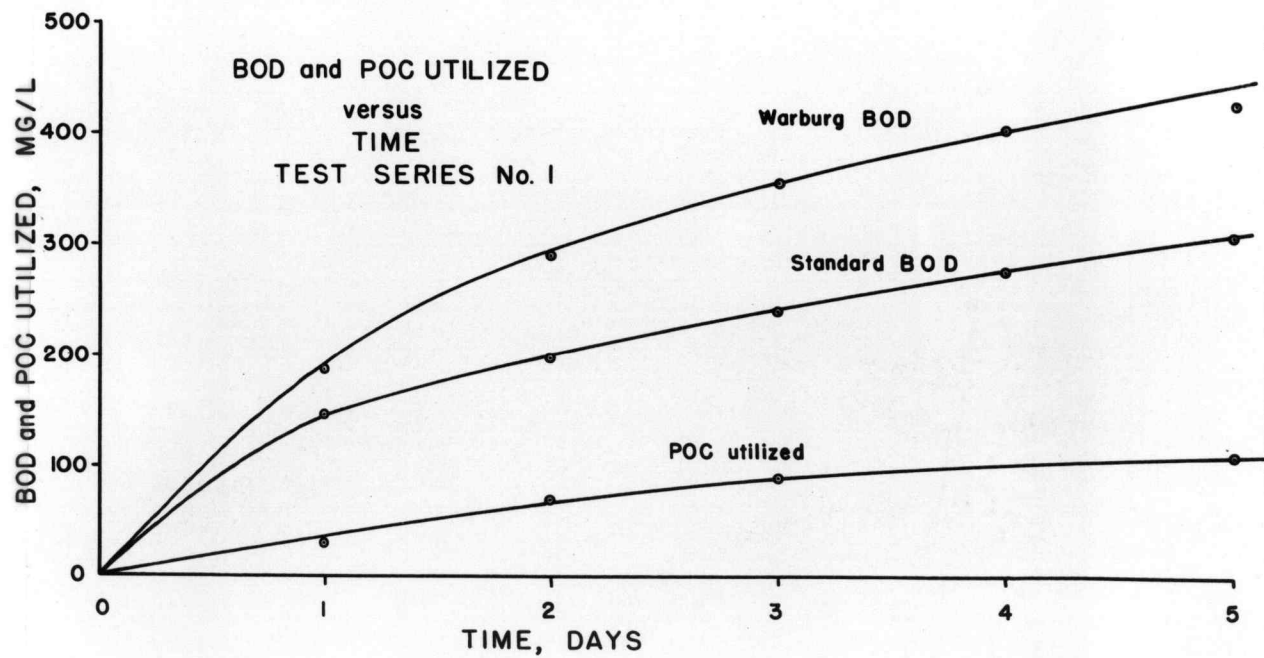


Plate 10. BOD and POC utilized versus time - Test Series No. 1.

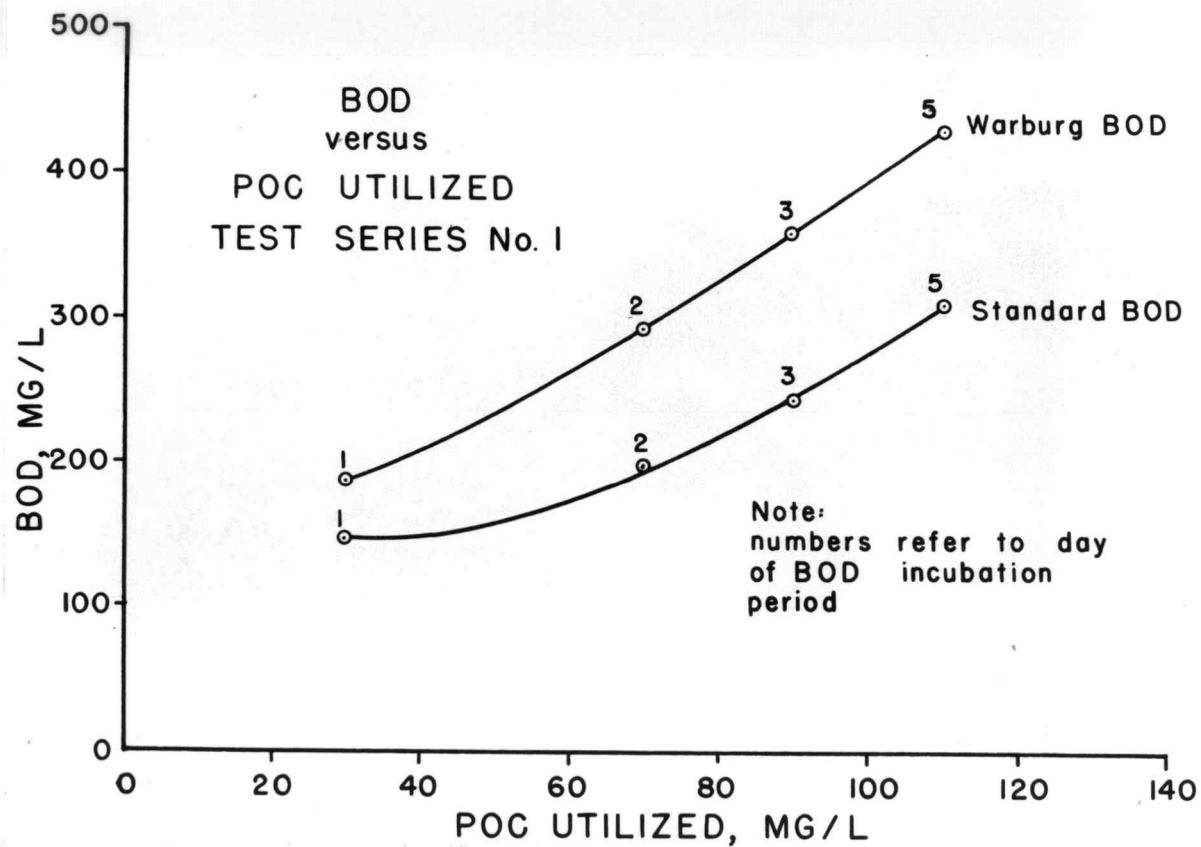


Plate 11. BOD versus POC utilized - Test Series No. 1.

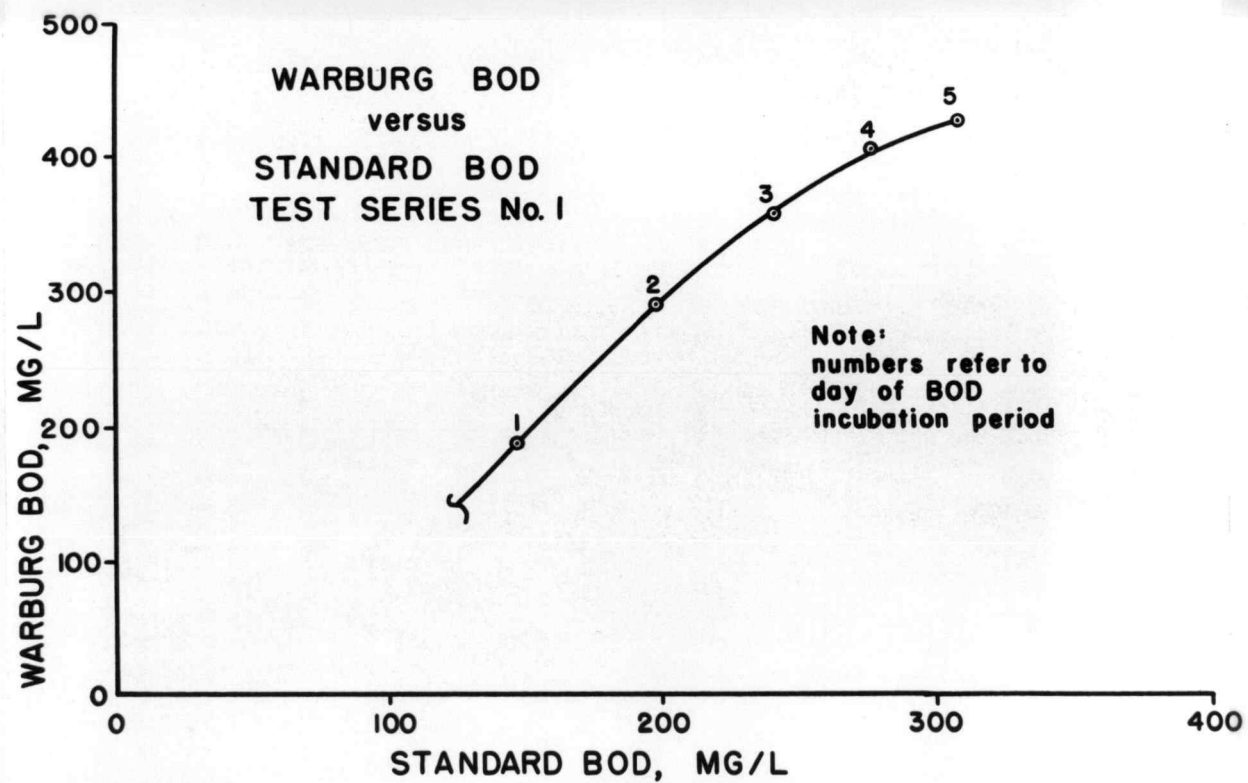


Plate 12. Warburg BOD versus standard BOD - Test Series No. 1.



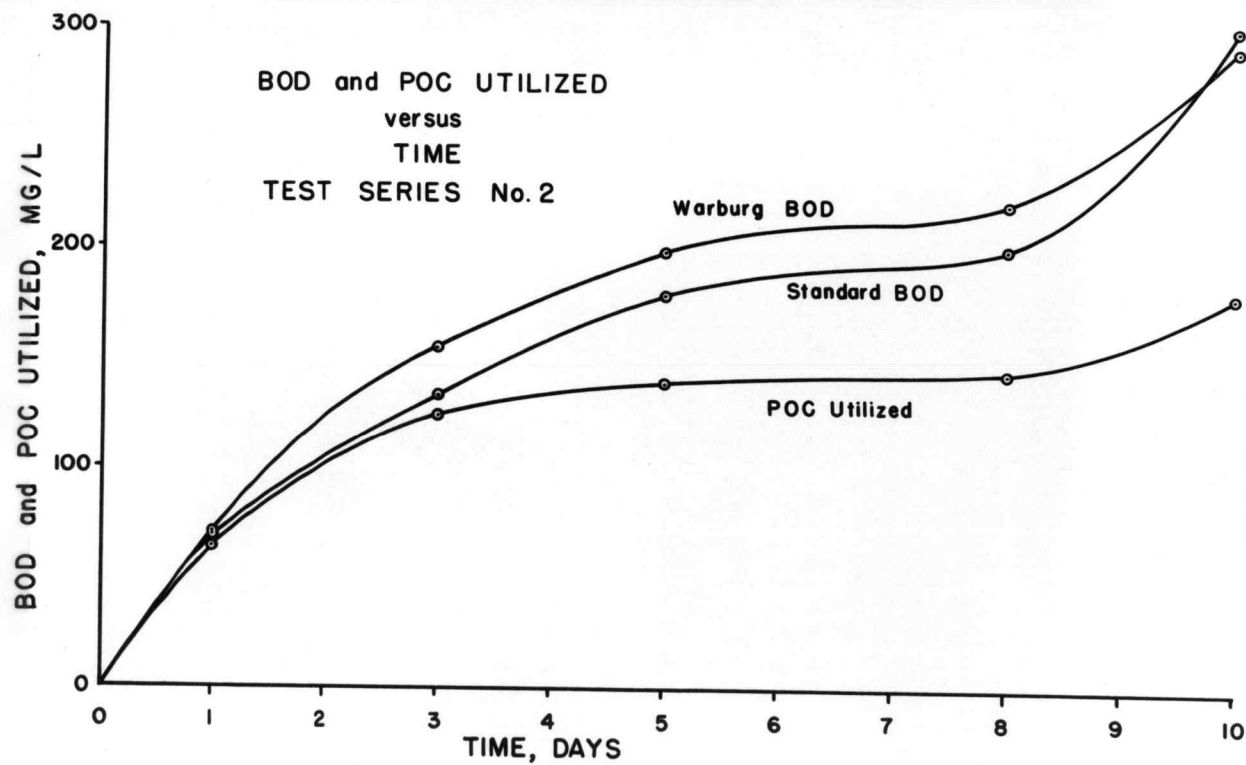


Plate 13. BOD and POC utilized versus time - Test Series No. 2.

application of the persulfate test to waste water analysis is that it measures only material which contributes to the carbonaceous or first stage BOD. The upward trend of the POC curve is in direct contrast to this assumption as it implies that the test measures material being utilized in the nitrification stage of the BOD.

When the data was replotted in Plate 14 to show the BOD-POC relationship, the same general trends were noted as those of the initial series. A similar bias toward BOD was also noted, though of lesser intensity. As before, the lack of similarity in the BOD curves was attributed to inherent differences in the methods of analysis. Plate 15 shows this lack of exact correlation.

Two interpretations of the situation with regard to nitrification are possible. If the situation is as the data from the second test series would indicate, then it would appear that POC utilized bears a relation to BOD, regardless of the presence of nitrification in the BOD reaction. On the other hand, if it were assumed that the POC utilized value for the tenth day shown in Plate 13 was in error, an entirely different situation would exist. If the amount of POC utilized actually remained unchanged during the nitrification stage of the BOD reaction, then a valid BOD-POC relationship would not be possible during that stage. It would then become impossible to predict a BOD in a situation known to be affected by nitrification from a POC measurement on the original sample, as the two values would have no constant relation to one another. This latter fact is further evidenced by the variable time of onset of nitrification

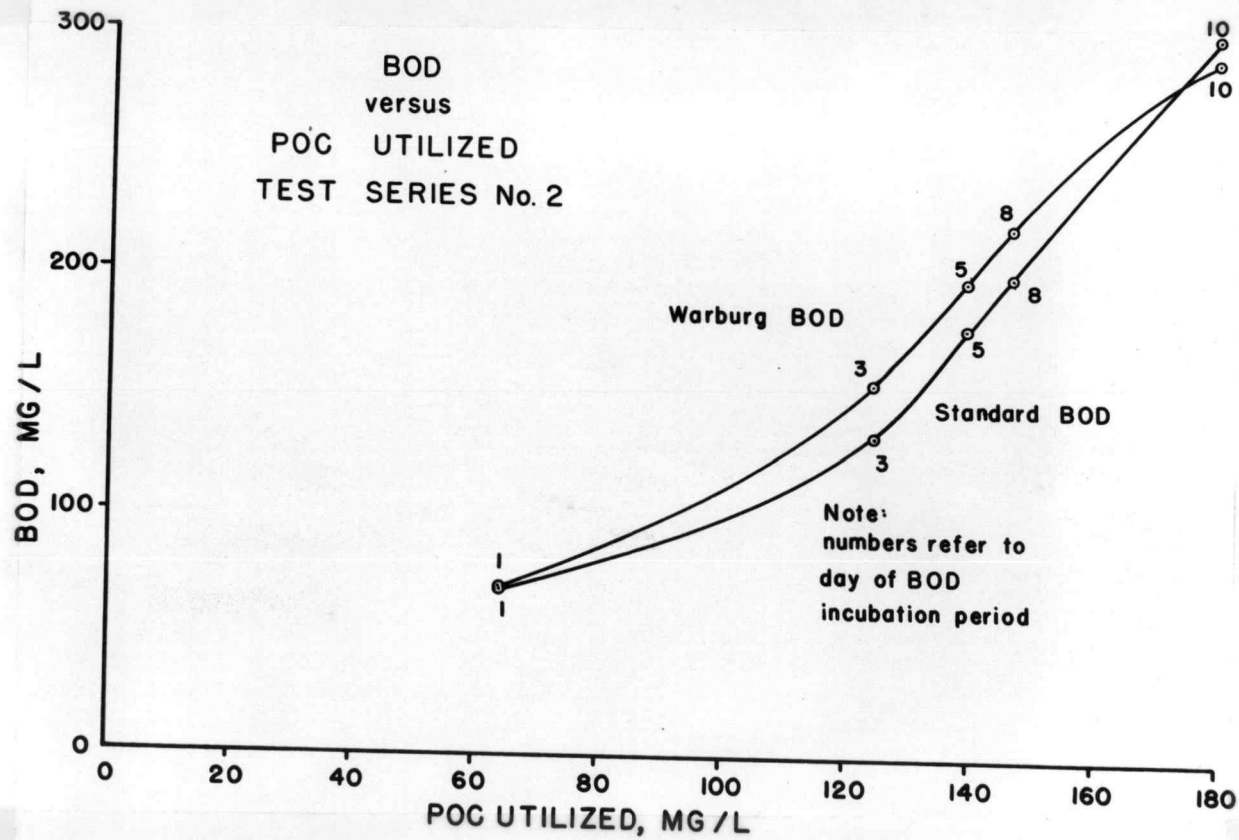


Plate 14. BOD versus POC utilized - Test Series No. 2.



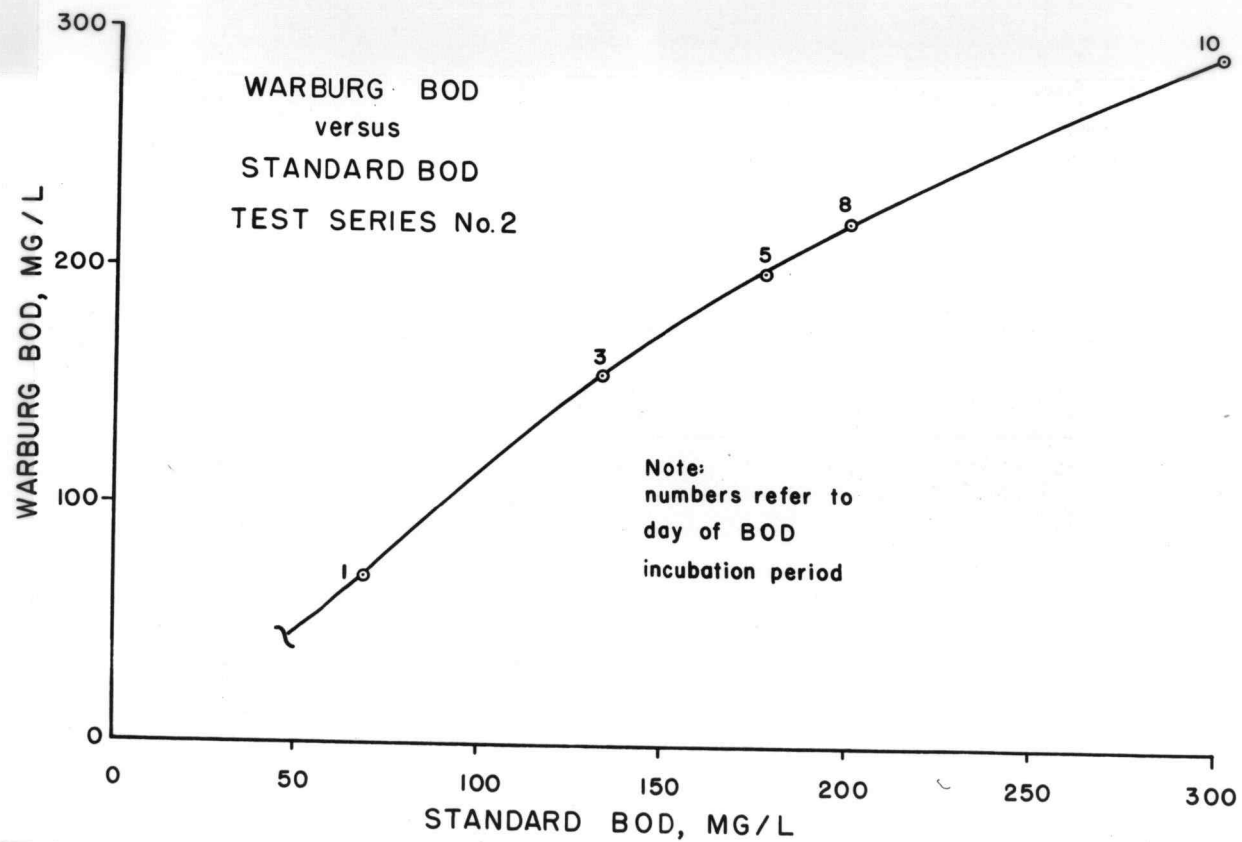


Plate 15. Warburg BOD versus standard BOD - Test Series No. 2.

in the BOD reaction. That the time of onset is variable implies independence between the nitrogenous and carbonaceous stages of the BOD reaction. Since it has been shown that the POC utilized and the carbonaceous BOD are closely related, the implication follows that POC utilized and nitrogenous BOD are independent.

In order to investigate further these effects of nitrification on the POC level, a third and final test series was organized. Since, in the second test series, the effects of nitrification had just begun to appear at the end of the test period, a longer test period of twelve days was selected in order that nitrification effects could fully develop. In addition to the Warburg and standard BOD tests and the POC test, several additional analyses were performed. In order to demonstrate that nitrification was taking place, tests for nitrate and nitrite nitrogen were conducted on samples from the Warburg flasks. Further, to make sure that the upswing in the BOD curves was not due to a sudden population increase in the organisms responsible for decomposition, total bacterial counts at 37° C. were determined on the Warburg flask contents at selected time intervals.

The results of this test series are given in Table 4. When the BOD and POC utilized values were plotted versus time in Plate 16, an important fact is noted in that the POC utilized curve gives good agreement with the Warburg BOD curve only up to the beginning of the nitrification phase. At this point, at about the seventh day, the BOD curve swings upward while the POC utilized

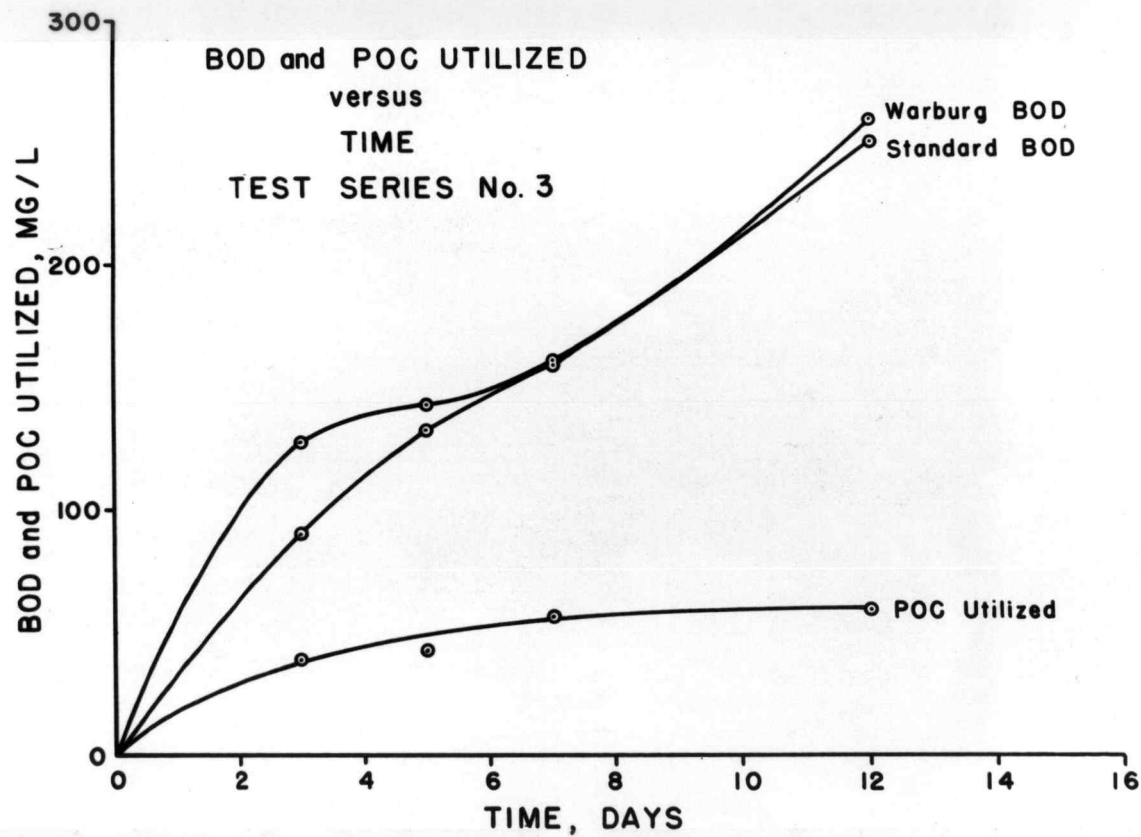


Plate 16. BOD and POC utilized versus time - Test Series No. 3.



curve levels off. This latter trend supports the supposition that POC utilized and nitrogenous BOD are independent, and in effect answers the question posed by the results of the second test series.

Plate 17 shows a plot with time of the nitrogen and total count data. The increase in nitrate and nitrite concentrations with time prove that the indicated rise in the BOD curves is due to nitrification. This fact is further supported by the rapid decrease in the bacterial population at the time of upswing of the BOD curves. Since the organisms responsible for the nitrification phase grow poorly, if at all, on the nutrient agar used in the total count determination, the decrease in bacterial population shown indicates the change from organisms decomposing carbonaceous material to those decomposing nitrogenous material.

Further evidence of the independence of POC and nitrogenous BOD is shown in Plate 18, where a plot of the respective BOD values versus the POC utilized is given. The abrupt rise in the curves at the seventh day onset of nitrification gives qualitative evidence that POC and BOD are no longer closely correlated in this region.

Because of the above qualitative evidence showing independence of POC and nitrogenous BOD, previous attempts to relate all POC and BOD values obtained from the experimental trickling filter must be regarded with suspicion. Effluent from a trickling filter is known to exhibit a greater degree of nitrogenous BOD activity than the untreated influent, and thus the POC-BOD relationships for the influent and effluent would not be comparable. In addition, efforts

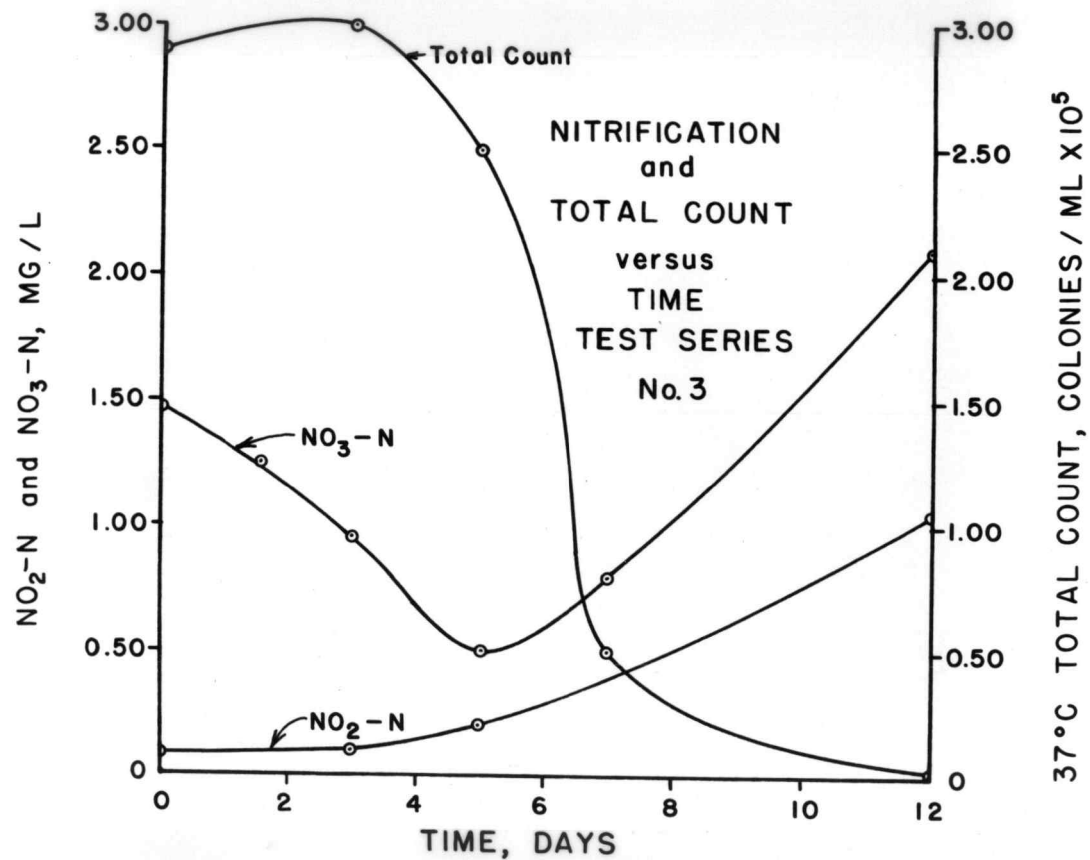


Plate 17. Nitrification and Total count versus time - Test Series No. 3.

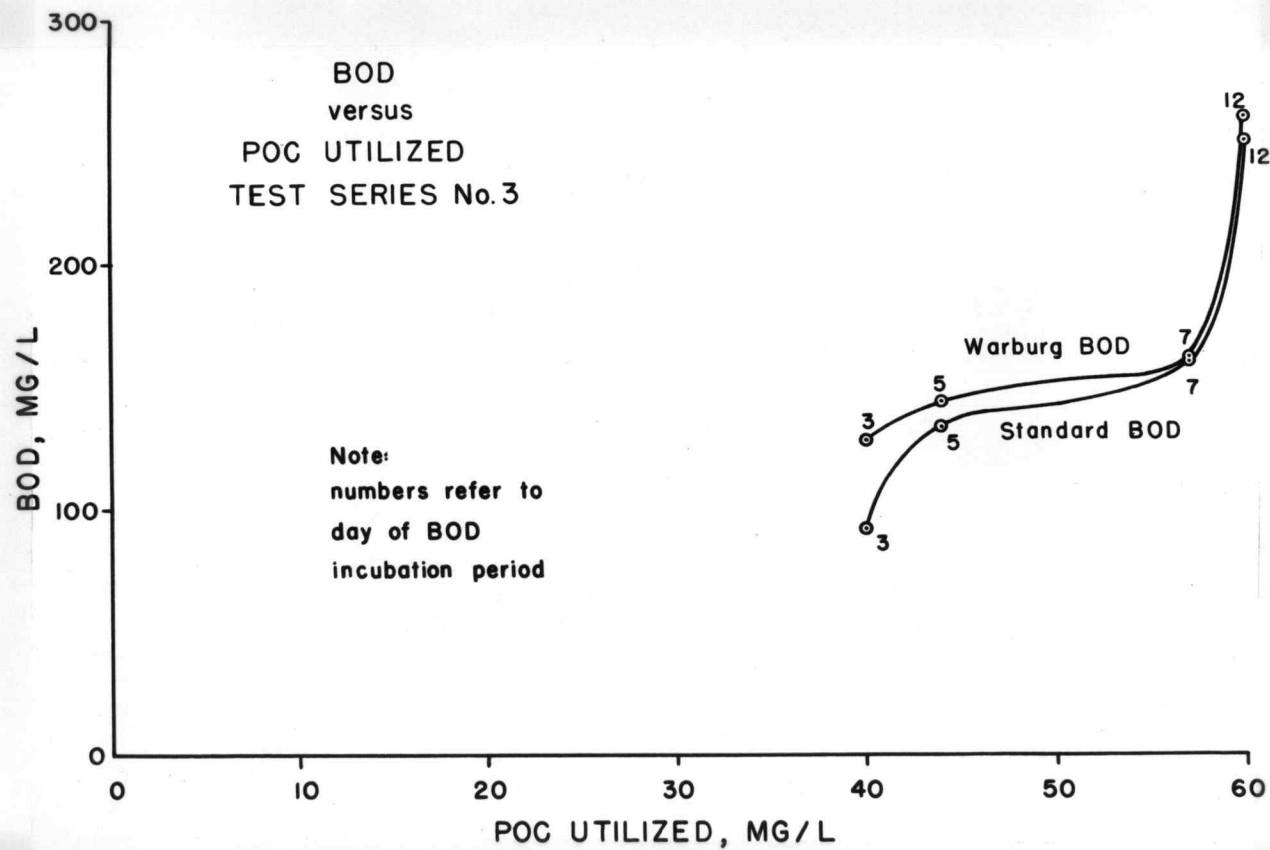


Plate 18. BOD versus POC utilized - Test Series No. 3.



to establish a valid comparison between BOD and POC removal efficiencies would also fail because of the effect of nitrification on both the BOD-POC relationship and the BOD reaction rate constants.

Since the BOD removal efficiencies for the experimental filter are also doubtful because of the known variation in the influent and effluent rate constants, the question remains as to what is the correct measure of filter performance. Because of the errors inherent in BOD removal efficiency determination, the POC removal efficiency seems to be a more correct evaluation of the existing conditions. The fact that the initial POC level is a valid representation of the carbonaceous BOD, as established by Purkerson and supported by data in this thesis, certainly permits its use as a measure of the carbonaceous BOD removal. It cannot be stated with absolute certainty, however, that measurement of the carbonaceous BOD removal is the only correct means to evaluate removal efficiency because of the existing controversy over the need to include the effects of nitrification.

One final point should be noted. In any situation where a valid application of a POC-BOD relationship can be used, there exist two alternatives as to an evaluation procedure. If it is felt to be absolutely necessary to express the results in terms of a BOD calculated from the initial POC level, then a calibration of the POC-BOD relationship applicable to the particular situation must be made in a manner similar to that of Purkerson. Such a calibration is always necessary because the resulting relationship

is of linear form derived by statistical methods from non-linear data. On the other hand, if it is accepted that for a given situation such a calibration could be made, but need not be, the POC values alone can be used to evaluate the pollutional situation. This latter approach is certainly to be recommended, as it eliminates the lengthy analytical and statistical procedures involved in establishing the required POC-BOD relationship.

## CONCLUSIONS

1. The term "persulfate oxidizable carbon", abbreviated POC, is preferable when referring to the results of the persulfate oxidation test applied to waste water analysis.
2. Sample pretreatment effects the magnitude of the values but has little effect upon a BOD-POC relationship, providing similar pretreatment is given to samples for either form of analysis.
3. In raw domestic sewage, carbon in forms other than true solution is measured by the persulfate oxidation test.
4. Direct measurement of the POC level in a standard BOD dilution is impossible due to sensitivity limits of the persulfate oxidation test.
5. Limited general conclusions as to the rate of POC uptake during a standard BOD reaction may be made from the POC uptake during a Warburg BOD reaction because of similarity of oxidative mechanisms.
6. Graphs of Warburg and standard BOD versus POC utilized for raw domestic sewage show a definite bias in favor of BOD.
7. BOD and POC utilized values for raw domestic sewage show similar trends with time during the carbonaceous stage of the BOD reaction.
8. BOD and POC utilized values for raw domestic sewage become independent during the nitrification stage of the BOD reaction.
9. BOD data used to establish a valid relationship between



BOD and the initial POC level of raw domestic sewage must be limited to values from the carbonaceous stage of the BOD reaction.

10. A valid comparison of BOD and POC removal efficiencies for the experimental trickling filter cannot be made because of the effects of nitrification.

11. In a polluttional situation where a valid BOD-POC relationship is known to exist, POC values alone may be used to evaluate the situation.

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## APPENDIX



## SAMPLE CALCULATION FOR THE PERSULFATE OXIDATION TEST

Weight of filter plus precipitate ..... 14.2733 g  
 Tare weight of filter ..... 14.1543 g  
 Difference ..... 0.1190 g  
 Difference ..... 119.0 mg  
 Blank Correction (average weight of  
 precipitate from triplicate determination) ..... 5.9 mg  
 Corrected difference ..... 113.1 mg

mg BaCO <sub>3</sub> per mg of carbon in the BaCO <sub>3</sub>	=	$\frac{\text{mol. wt. BaCO}_3}{\text{mol. wt. Carbon}}$
" "	=	$\frac{197.370}{12.010}$
" "	=	16.432
Total mg of Carbon in the sample	=	$\frac{\text{Corr. Difference}}{16.432}$
" "	=	$\frac{113.1}{16.432}$
" "	=	6.9 mg
mg Carbon per unit volume of the original sample	=	$\frac{6.9 \text{ mg}}{30.0 \text{ ml}}$
" "	=	0.23 mg Carbon/ml
" "	=	230.0 mg Carbon/l

TABLE 1  
RAW SEWAGE SEPARATION DATA

Sample	BOD <sub>5</sub> mg/l	Soluble carbon mg/l	% of original BOD	% of original soluble carbon
Original sample	257	163	100%	100%
Imhoff cone supernatant	183	119	71%	73%
Supernatant after centrifuging	133	94	52%	58%
Gooch crucible filtrate	130	81	51%	50%

TABLE 2  
TEST SERIES NO. 1 - RAW DATA

Day	Warburg BOD mg/l	Standard BOD mg/l	POC remaining mg/l	POC utilized mg/l
0	---	---	230	---
1	188	148	200	30
2	291	199	160	70
3	358	242	140	90
4	407	277	140*	90*
5	427	308	120	110

\* Value in error - reaction flask left in the oven for 22 hours.

TABLE 3  
TEST SERIES NO. 2 - RAW DATA

Day	Warburg BOD mg/l	Standard BOD mg/l	POC remaining mg/l	POC utilized mg/l
0	---	---	224	---
1	70	69	160	64
3	155	133	100	124
5	198	178	85	139
8	220	200	80	144
10	292	300	45	179



TABLE 4

## TEST SERIES NO. 3 - RAW DATA

Day	Warburg BOD mg/l	Standard BOD mg/l	POC remaining mg/l	POC utilized mg/l	NO <sub>2</sub> - N mg/l	NO <sub>3</sub> - N mg/l	Total count, 37°C. colonies/ml
0	---	---	110	---	0.09	1.48	$2.9 \times 10^5$
3	128	91	70	40	0.11	0.95	$3.0 \times 10^5$
5	144	133	66	44	0.21	0.50	$2.5 \times 10^5$
7	161	160	53	57	---	0.80	$5.0 \times 10^4$
12	259	250	50	60	1.05	2.10	$2.0 \times 10^3$