RELATIONSHIP BETWEEN RETENTION TIME AND SPECIFIC GROWTH RATE IN A MUNICIPAL WASTEWATER UNDER CONTINUOUS FLOW CONDITIONS

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ABSTRACT

RELATIONSHIP BETWEEN RETENTION TIME AND SPECIFIC GROWTH RATE IN A MUNICIPAL WASTEWATER UNDER CONTINUOUS FLOW CONDITIONS

By

John K. Nelson

A relationship between bacterial specific growth rate (k_1) and bacterial specific respiration rate (k_r) was developed for a municipal wastewater. Determination of the specific respiration rate was accomplished by use of a continuous flow respirometer connected to a mixed bacterial culture grown under continuous flow conditions with municipal wastewater as a feed solution, using the equation:

$$k_1 = \frac{k_r - b}{d}$$

where k_1 = specific growth rate (hr^{-1})

d = mg of oxygen consumed per gram of volatile suspended solids (VSS) produced

k_r = specific respiration rate (mg O_2/hr/g VSS)

b = endogenous respiration rate (mg O_2/hr/g VSS)

The data resulted in an endogenous respiration rate of $b = 27$ mg O_2/hr/g VSS under steady state conditions.

In addition, the value of d was found to be 540 mg O_2 consumed per gram of volatile suspended solids produced.

A linear relationship between effluent soluble substrate concentration and specific growth rate k_1 was developed using the following equations:

$$COD_s = 20 + 240\ k_1$$

$$TOC_s = 9 + 45\ k_1$$

where

COD_s = dissolved chemical oxygen demand (mg/l)

TOC_s = dissolved total organic carbon (mg/l)

These equations were valid up to $k_1 = 0.167$/hr.

The linear relationship between specific growth rate and both dissolved COD_s substrate concentration and specific respiration rate became discontinuous after $k_1 = 0.167^{-hr}$. This discontinuity was attributed to a substrate limitation.

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John K. Nelson

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SYMBOLS

b	Endogenous respiration rate (mg O_2/g VSS/hr).
BOD	Biochemical oxygen demand (mg/l).
C	Concentration of dissolved oxygen (mg/l).
c	Teissier equation constant.
C°	Degrees centigrade.
COD_S	Dissolved chemical oxygen demand (mg/l).
D	Dilution rate = Q/V.
d	Weight of oxygen consumed per gram of volatile suspended solids produced.
DO	Dissolved oxygen (mg/l).
g	Grams.
k_1	Specific growth rate (grams per gram cell weight per hour).
k_m	Maximum specific growth rate
k_r	Specific respiration rate (mg O_2/g VSS/hr).
mg/l	Milligrams per liter.
ml	Milliliters.
Q	Feed rate (ml/min).
r_r	Overall respiration rate (mg/l O_2/hr).
S	Substrate concentration (mg/l).
S_n	Substrate concentration at which $k_1 = k_m/2$.
T	Temperature (°C).
t_r	Retention time = V/Q (hours).
TOC_S	Dissolved total organic carbon (mg/l).
V	Volume (liter).
VSS	Volatile suspended solids (mg/l).
x	Concentration of volatile suspended solids (grams/l).

INTRODUCTION

The design, operation and performance of a biological wastewater treatment process is largely dictated by the rate at which the bacteria are capable of assimilating or oxidizing carbonaceous matter. Assimilation of organic matter is accompanied by an increase in the number of bacteria. This increase in numbers may be defined by the specific growth rate constant (k_1) which is part of an equation used to characterize the exponential multiplication of bacteria in a culture:

$$k_1 = \frac{\ln (x/x_o)}{t} \qquad \text{(I)}$$

k_1 = specific growth rate

x = cell concentration at time t

x_o = initial cell concentration

t = time elapsed

Knowing the maximum rate of growth at which bacteria are capable of multiplying in a given wastewater is of importance in relation to design and operational parameters for wastewater treatment.

An increase in microbial mass can be determined directly by measurement of volatile suspended solids, analysis of

total organic (cell) nitrogen, assessment of DNA content or application of the firefly luminescent test for measurement of cellular ATP (1). Common indirect measurements have related bacterial density to turbidity or to the metabolic activity of cells. Oxygen uptake has been used for aerobic organisms because the rate of oxygen uptake is proportional to cell mass under certain conditions.

Experimentation with continuous flow cultures of bacteria has shown that a direct proportionality exists between the specific respiration rate (k_r) and the specific growth rate (k_1) (21):

$$k_r = \frac{1}{x} r_r = b + dk_1 \qquad \text{(II)}$$

k_r = specific respiration rate (mg O_2 per gram of cell weight per hour)

x = cell concentration (grams/l)

r_r = overall respiration rate (mg/l O_2 per hour)

b = endogenous respiration rate

d = mg O_2 utilized per gram of cell weight produced

k_1 = specific growth rate (grams per gram cell weight per hour)

Equation II states that by measuring the overall respiration rate (r_r) and dividing this value by the cell concentration (x) a specific respiration rate (k_r) may be found for a bacterial culture. Equation II also shows that plotting k_r versus k_1 should produce a straight line with the y

intercept equal to the endogenous respiration rate (b) and the slope (d) equal to the mg O_2 consumed per gram of cell weight produced. Thus determination of the specific respiration rate (k_r) would provide a means to obtain the specific growth rate (k_1), if (b) and (d) are known.

A variety of techniques are available for the measurement of overall respiration rate (r_r) (10). These techniques rely primarily on the somewhat involved manometric (Warburg) principles or the less involved oxygen electrode systems. Ideally, the closer one can approach actual steady state conditions and at the same time measure bacterial respiration in situ the better can laboratory results be related to full scale treatment plant conditions. Several techniques are available for determining overall respiration rate (r_r) of biological organisms under flow-through conditions (6-8,13-15,17,18,20,24). A unique flow-through respirometer using polarographic dissolved oxygen probes has been specifically adapted to wastewater treatment (8,17). This method uses a dissolved oxygen electrode immersed in a bacterial growth chamber with a second electrode immersed in a separate respiration cell through which the bacterial culture from the growth chamber is pumped at a pre-determined rate of flow. The bacterial cells in the respiration cell and in the pump line respire at the same rate as the bacterial cells in the growth chamber from which the culture is continuously drawn.

The bacterial culture flowing through the respiration cell is closed off from the atmosphere and therefore the dissolved oxygen level in the respiration chamber falls to a lower level than that existing in the aerated growth chamber. The oxygen level in the growth chamber and the oxygen level in the respiration cell are monitored separately with the difference between the two levels being proportional to the retention time of the bacterial suspension in the respiration cell. Measurement of the overall respiration rate (r_r) of the bacterial culture in mg/l O_2 utilized per hour can thus be obtained by the following expression:

$$r_r = \frac{(C_1 - C_2)Q}{V} \times 60 \qquad \text{(III)}$$

r_r = overall respiration rate (mg/l O_2 per hour)

C_1 = mg/l of O_2 in the aerated growth chamber

C_2 = mg/l of O_2 in the respiration cell

Q = rate of flow in ml per minute through the respiration cell

V = volume of the respiration cell and tubing (ml)

The specific respiration rate (k_r) may then be found as a result of a linear relationship which exists between the bacterial mass (x) measured as volatile suspended solids (VSS) and the overall respiration rate (r_r) as shown in equation II: $k_r = \frac{r_r}{x}$.

To provide steady-state conditions under which the respiration rate could be measured, a continuous flow culturing technique was used. The chemostat has provided a method by which a bacterial culture can be studied under continuous flow and complete mixing conditions (4,9,11-13,19,21,24). The chemostat is a device which consists of a feeding system that admits a nutrient liquid at a controlled rate to a constant volume reactor chamber. The flow of the nutrient liquid into the reactor and the resulting loss of bacterial biomass from the reactor is at a constant rate. The equation for the growth of bacteria in a batch culture can be written as (11):

$$\frac{dx}{dt} = k_1 x \qquad \text{(IV)}$$

k_1 = specific growth rate (growth per unit cell weight per unit time)

x = microbial cell concentration (grams/l)

t = time (hr)

The growth rate in a continuous flow culture then becomes:

$$\frac{dx}{dt} = k_1 x - Dx \qquad \text{(V)}$$

D = dilution rate or flow (Q) divided by volume (V) $\doteq 1/t_r$

where t_r = retention time

Equation V shows that if D is greater than k_1 the culture will be diluted out more rapidly than it can grow and will eventually be completely washed out of the growth chamber.

If D is less than k_1 the bacteria are growing more rapidly than they are being washed out and the density of the bacteria in the reactor will continuously increase. Steady state operation is possible only when the specific growth rate (k_1) is equal to the dilution rate (D). Equation V expresses this mathematically. For steady state, i.e., $\frac{dx}{dt} = 0$, we obtain

$$k_1 = D \qquad \text{(VI)}$$

Since the volume of the chemostat is constant, the dilution rate is determined by the feed rate and can be varied simply by varying the feed rate. This provided a means for establishing the relationship between specific respiration rate and specific growth rate. Decreasing the feed rate results in a proportional decrease in substrate concentration due to the increased contact time between cells and substrate and vice versa. Corresponding to a given D-value, as long as it remains below the maximum growth rate k_m, the culture will automatically establish a definite substrate concentration in the reactor and in the reactor effluent (11).

The system can thus be operated at a series of different growth rates and for each value of k_1 the corresponding substrate concentration (S) can be experimentally determined. Several mathematical models have been proposed for the relationship between k_1 and S. The Monod equation is most often

referred to in the literature (4,9,12,13,16,19,23,27) and is expressed as:

$$k_1 = k_m\left(\frac{S}{S_n+S}\right) \qquad \text{(VII)}$$

where k_1 = specific growth rate

k_m = maximum growth rate

S = substrate concentration

S_n = substrate concentration at which $k_1 = k_m/2$

A second model uses the Teissier equation (19,21) and may be expressed as:

$$\frac{dk_1}{dS} = c(k_m - k_1) \qquad \text{(VIII)}$$

The integrated forms of equation VII is similar to a first order reaction equation:

$$k_1 = k_m(1 - e^{-cS}) \qquad \text{(IX)}$$

From equation VIII it can be seen that the growth rate increases with the increasing substrate concentration proportionally to the difference between the existing growth rate (k_1) and a maximum growth rate (k_m).

In a third model the relationship between k_1 and S has been expressed as a two phase discontinuous relationship (3,4), where the rate of growth is directly proportional to the substrate concentration up to a critical point (k_m).

Above this point of discontinuity (k_m), the growth rate is constant and independent of substrate concentration.

The experimental data obtained were then used in an attempt to establish relationships between specific growth rate (k_1), specific respiration rate (k_r) and substrate concentration (S) under actual field conditions using municipal wastewater as a feed solution.

METHODS AND PROCEDURES

The continuous flow culture apparatus is shown in Figures 1, 2, and 3. It was constructed from a 190 liter (55 gal.) barrel lined by a polyethylene container of equal size. The actual volume of the complete mix reactor was 160 liters (30 gal.). Complete mixing was maintained by a 1/3 horsepower Model S-7 Lightning Mixer running at a constant speed of 1725 rpm. Compressed air was passed through an air filter to a pressure regulator set at 6 psi. The air was then passed through an Ace Glass #1A-15-1 flow meter and dispersed from a modified Nalgene plastic aspirator for the purpose of supplying large bubble aeration. The mixing supplied by the Lightning Mixer served to break up the large air bubbles into a finer size. The dissolved oxygen range maintained during the experiment was 2.0 to 6.0 mg/l. Primary effluent from the East Lansing Wastewater Treatment Plant was obtained by tapping into a sampling pump which transferred a portion of the flow to a small stilling well that was continuously flushed by the incoming flow. From the stilling well, the primary effluent was pumped at a steady rate into the reactor using a Sigmamotor peristalic pump Model T65. The feed rate was determined by using a graduated cylinder and stopwatch. Effluent from the reactor was returned to the inplant waste.

Due to the tendency for bacterial floc to adhere to the side walls of the aeration chamber, the walls were brushed down daily.

The respiration cell as illustrated in Figures 1, 2, 3, 4 and 5 was designed by R. Knop and has been described in his thesis on the determination of the overall oxygen transfer coefficient in an operating sewage plant (6). The device was constructed of galvanized sheet metal with a top made of 0.6 cm (1/4 in.) lucite attached to the base with eight screws to permit removal for cleaning. The lucite top was slanted to allow immediate passage of air bubbles through the effluent line. An inlet piece of 1.25 cm ID (1/2 in.) lucite tubing entered to within 1.9 cm (3/4 in.) of the bottom of the cell. The outlet of the cell was constructed of 1.9 cm (3/4 in.) lucite tubing to avoid possible clogging and provide fast entrapment of rising air bubbles. Stirring within the cell was provided by a variable speed Cenco Laboratory Stirrer #18802 equipped with a chain stirrer attached to the shaft. The bacterial culture was pumped from the reactor to the respiration cell via a Cole Palmer Model 7017 peristalic pump using 0.6 cm (1/4 in.) Tygon tubing.

Monitoring of the dissolved oxygen levels in the reactor and the respiration cell was done with two dissolved oxygen meters employing polarographic dissolved oxygen probes (Model 54--Yellow Springs Instrument Co.).

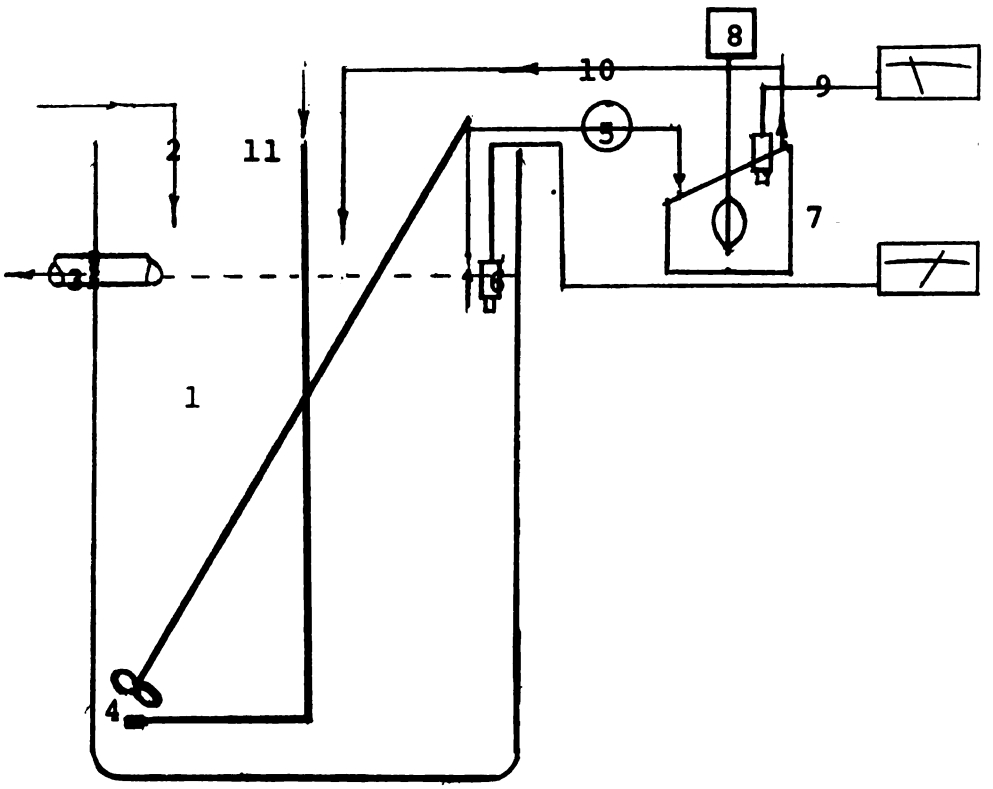

Figure 1. Chemostat and Respirometer.

1. Chemostat--complete mix reactor operated at continuous flow.
2. Influent, wastewater feed.
3. Chemostat effluent.
4. Sparger and mixer blade.
5. Variable speed peristalic pump drawing bacterial culture from chemostat to respiration cell.
6. Polarographic dissolved oxygen probe recording concentration of DO in chemostat.
7. Respiration cell.
8. Variable speed chain stirrer.
9. Polarographic dissolved oxygen probe recording concentration of DO in respiration cell.
10. Effluent from respiration cell.
11. Air supply line.

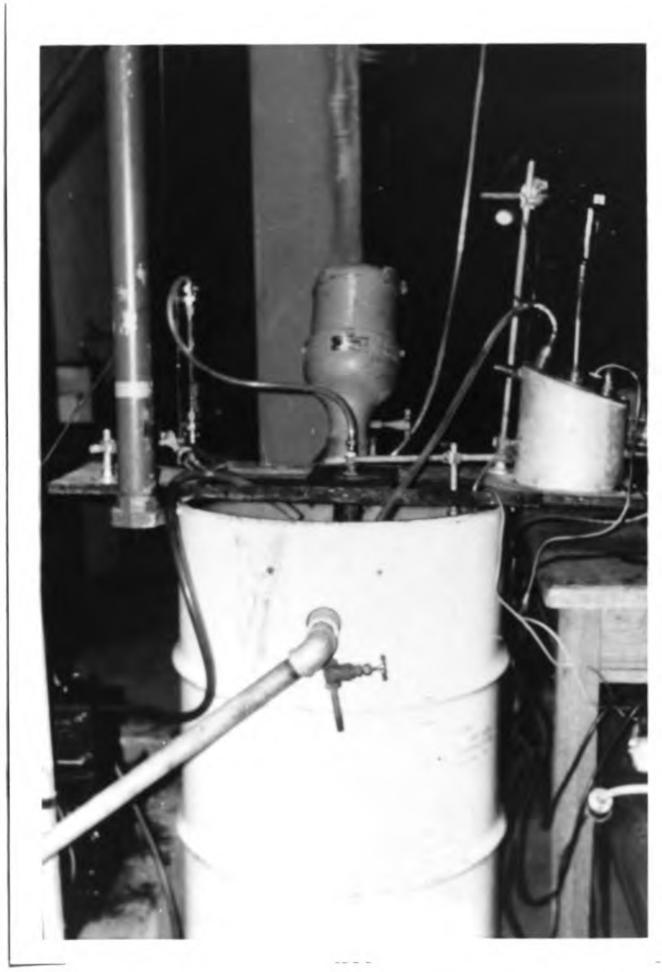

Figure 2. Continuous flow culture apparatus showing mixer and respirometer cell.

Figure 3. Flow through respirometer attached to continuous flow culture apparatus.

Figure 4. Flow through respirometer showing respiration cell, variable speed peristalic pump and DO meters.

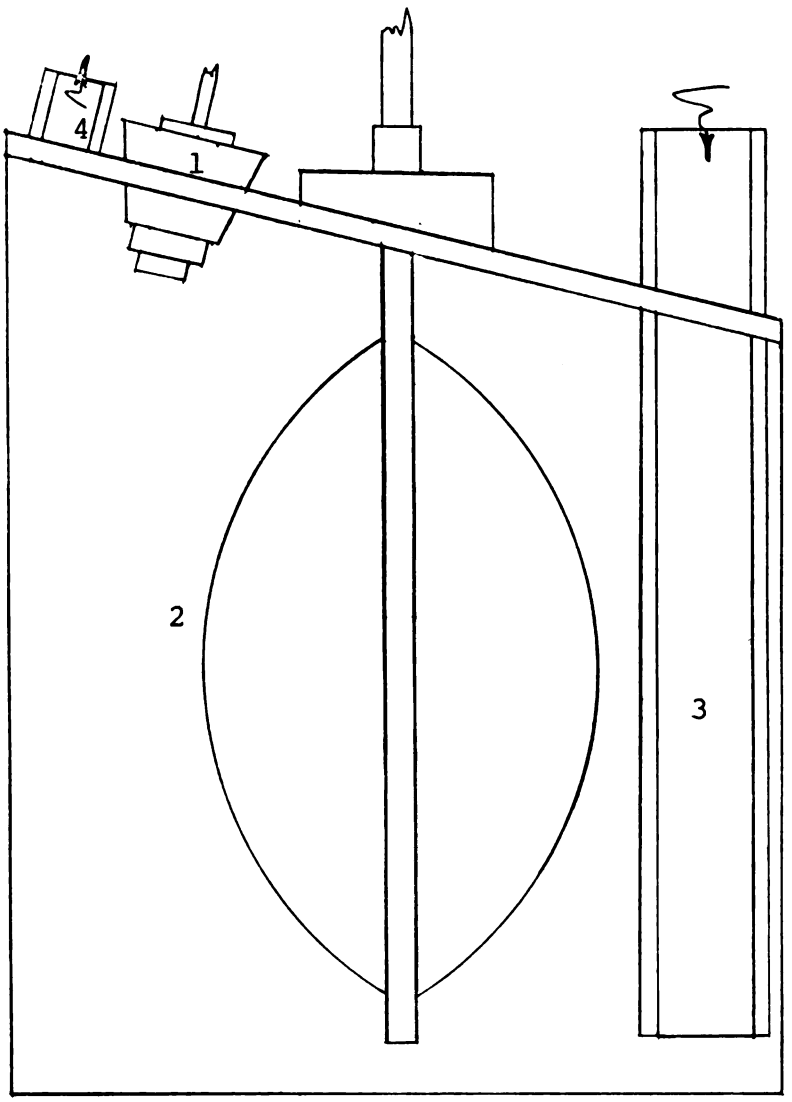

Figure 5. Respiration cell.

1. Polarographic dissolved oxygen probe
2. Variable speed chain stirrer
3. Influent from chemostat
4. Effluent from respiration cell

Using primary effluent from the East Lansing Wastewater Treatment Plant as a feed solution, a series of overall respiration rates (r_r) were measured for a pre-determined D-value. D-values ($D = 1/t_r$) or retention times were closely controlled by measuring feed rates every 4 hours and dividing these values into the volume of the reactor ($t_r = V/Q$). The average D-values and retention times obtained in this manner are shown in Table I.

For each series of D-values, the reactor was allowed to acclimate for a one-week period. This period of acclimation was then followed by a series of ten daily respiration rate and effluent substrate concentration measurements. Substrate concentration was determined as dissolved chemical oxygen demand (COD_s) and dissolved total organic carbon (TOC_s).

Prior to the measurement of each overall respiration rate, the dissolved oxygen meters were calibrated against an air-saturated sample of tap water.

Volatile suspended solids (VSS) were analyzed by filtration through a glass fiber mat and ignited at 550°C (25). Concentration of the volatile suspended solids was then designated as X and used to determine the specific respiration rate (k_r) from the overall respiration rate (r_r) as described in equation II.

Total dissolved organic carbon (TOC_s) and dissolved chemical oxygen demand (COD_s) were determined for the filtrate

TABLE I

AVERAGE D-VALUES, RETENTION TIMES AND FEED RATES

D			t_r	Q
hr^{-1}			hr	ml/min
Y	a			
	+	-		
.0207	2.8	1.9	48.3	55
.0420	2.2	2.2	23.8	112
.0833	2.8	2.2	12.0	223
.167	1.8	2.7	6.0	445
.333	2.4	1.5	3.0	889

Y = Average D value for period of experiment

a = Average variation of % of D for period of experiment

obtained from the suspended solids analysis. The filtrate sample was acidified to pH 1 with sulfuric acid, sparged with nitrogen and analyzed for total organic carbon with a Beckman organic carbon analyzer (25). Chemical oxygen demand was measured by using an acidified solution of 0.01 N potassium dichromate and refluxing for two hours (25).

DATA AND RESULTS

The oxygen utilization rates (k_r and r_r) obtained from the flow through respirometer and the dissolved substrate data (COD_s and TOC_s) for a series of D values are listed in Tables II to VII. Figures 6 to 11 represent the variation in the data for each D value. As can be seen from these graphs and the tabulated standard deviations, the variation in r_r, k_r, effluent COD_s and effluent TOC_s increased as the values of D increased. When influent COD_s values are tabulated and graphed (Tables II to VII, and Figure 8), it can be seen that as the influent COD_s increases and then declines with the last series of D values, so do the r_r, k_r and the effluent COD_s values. Figure 9, showing incoming TOC_s values, tends to follow those patterns as shown by the COD_s values although not as pronounced as with the influent COD_s values.

As can be seen from Figure 10 and Tables II to VII, the temperature fluctuations during this experiment were small, with a range of 24°C-29°C. In order to compensate for any variation in r_r due to these temperature changes, corrections were made by using the following formula (28):

TABLE II

SUBSTRATE AND RESPIRATION DATA

t_r = 48 hr. D = 0.0208

Temp °C	r_r Obs.	r_r 20°C	VSS g/l	$k_{r_{20°C}}$	COD_s In	COD_s Eff	TOC_s In	TOC_s Eff
29	4.4	2.3	0.060	38	68	24	23	7
29	4.3	2.3	0.060	38	-	27	-	12
29	3.8	2.0	0.058	35	53	26	24	11
27	3.6	2.1	0.056	38	81	27	29	11
27	4.1	2.5	0.062	40	68	27	28	11
29	4.2	2.2	0.053	41	62	28	31	11
29	3.9	2.0	0.054	38	-	24	-	11
29	3.9	2.0	0.053	38	75	28	26	9
27	4.1	2.5	0.065	38	-	24	-	8
27	4.5	2.7	0.068	40	-	27	-	7
$\bar{y}$* 28	4.1	2.3	0.059	38	68	26	27	10
σ* 1.0	0.3	0.2	0.005	1.6	9.8	1.3	3.1	3.0

*$\bar{y}$ = Average value

σ = Standard deviation

TABLE III

SUBSTRATE AND RESPIRATION DATA

t_r = 24 hr. D = 0.0417

Temp °C	r_r Obs	r_r 20°C	VSS g/l	k_r 20°C	COD_s In	COD_s Eff	TOC_s In	TOC_s Eff
27	4.1	2.4	0.052	46	91	35	32	13
26	5.4	3.5	0.067	52	79	38	27	13
26	5.4	3.5	0.065	54	79	34	27	13
27	4.8	2.9	0.061	47	86	32	31	12
27	5.0	3.0	0.062	49	86	32	31	12
27	5.2	3.2	0.066	48	86	29	27	12
27	5.2	3.1	0.068	46	86	38	27	12
26	6.1	3.8	0.081	47	77	34	26	13
26	6.2	3.9	0.097	40	73	30	29	11
26	6.9	4.4	0.087	51	110	29	42	10
$\bar{y}$ 26	5.4	3.4	0.071	48	85	33	30	12
σ 0.5	0.8	0.6	0.014	3.9	10	3.3	4.7	1.0

TABLE IV

SUBSTRATE AND RESPIRATION DATA

t_r = 12 hr. D = 0.0833

	Temp °C	r_r Obs.	r_r 20°C	VSS g/l	k_r 20°C	COD_s In	COD_s Eff	TOC_s In	TOC_s Eff
	26	8.1	5.2	0.078	67	110	35	41	11
	26	8.8	5.7	0.077	74	100	38	37	12
	26	9.6	6.0	0.074	81	100	30	37	12
	26	7.6	4.9	0.069	71	87	30	33	9
	26	8.1	5.3	0.079	67	87	31	33	9
	26	8.8	5.7	0.089	64	90	43	33	15
	26	9.6	6.2	0.079	79	120	44	38	15
	25	8.7	6.0	0.067	90	120	47	41	17
	25	8.7	6.0	0.083	72	120	42	43	14
	25	9.0	6.2	0.083	75	120	44	43	15
$\bar{y}$	26	8.7	5.7	0.078	74	100	38	38	13
σ	0.5	0.6	0.4	0.006	7.8	14	6.5	4.0	2.7

TABLE V

SUBSTRATE AND RESPIRATION DATA

t_r = 6 hr. D = 0.167

Temp. °C	r_r Obs.	r_r 20°C	VSS g/l	k_r 20°C	COD_s In	COD_s Eff	TOC_s In	TOC_s Eff
25	14	9.5	0.110	88	110	64	36	22
24	12	9.3	0.074	130	77	39	26	13
24	11	8.6	0.075	110	100	54	35	15
24	11	8.5	0.073	120	100	84	35	20
24	16	12	0.100	120	-	65	-	18
23	17	14	0.110	120	150	75	38	22
22	15	13	0.130	97	-	51	-	14
22	14	12	0.083	140	-	42	-	11
23	12	9.5	0.110	87	170	66	47	15
23	20	16	0.120	140	150	72	46	20
$\bar{y}$ 23	14	11	0.098	120	120	61	38	17
σ 1.0	1.8	2.5	0.021	20	34	14	7.2	3.9

TABLE VI

SUBSTRATE AND RESPIRATION DATA

t_r = 3 hr. D = 0.333

Temp. °C	r_r Obs.	r_r 20°C	VSS g/l	$k_{r_{20°C}}$	COD_s In	COD_s Eff	TOC_s In	TOC_s Eff
21	15	14	0.011	130	140	82	49	35
19	5.2	5.6	0.051	110	130	59	45	24
20	8.8	8.8	0.068	130	140	70	34	25
19	13	14	0.100	130	110	82	45	31
19	14	16	0.096	160	120	73	53	34
19	10	11	0.085	130	51	50	38	27
20	9.1	9.1	0.065	140	100	65	33	29
19	8.0	8.7	0.059	150	70	40	39	24
19	14	15	0.091	160	68	75	38	38
19	12	13	0.088	140	51	54	38	24
$\bar{y}$ 19	11	11	0.081	140	98	65	41	29
σ 0.7	3.2	3.3	0.088	17	35	14	6.5	5.2

TABLE VII

AVERAGE SOLUBLE SUBSTRATE AND RESPIRATION DATA

D	t_r	r_r		k_r	COD_s		TOC_s	
hr^{-1}	hr	Obs.	20°C	20°C	In	Eff.	In	Eff.
0.0208	48	4.1	2.3	38	68	26	27	10
0.0417	24	5.4	3.4	48	65	33	30	12
0.0833	12	8.7	5.7	74	100	38	38	13
0.167	6	14	11	120	120	61	38	17
0.333	3	11	11	140	98	65	41	29

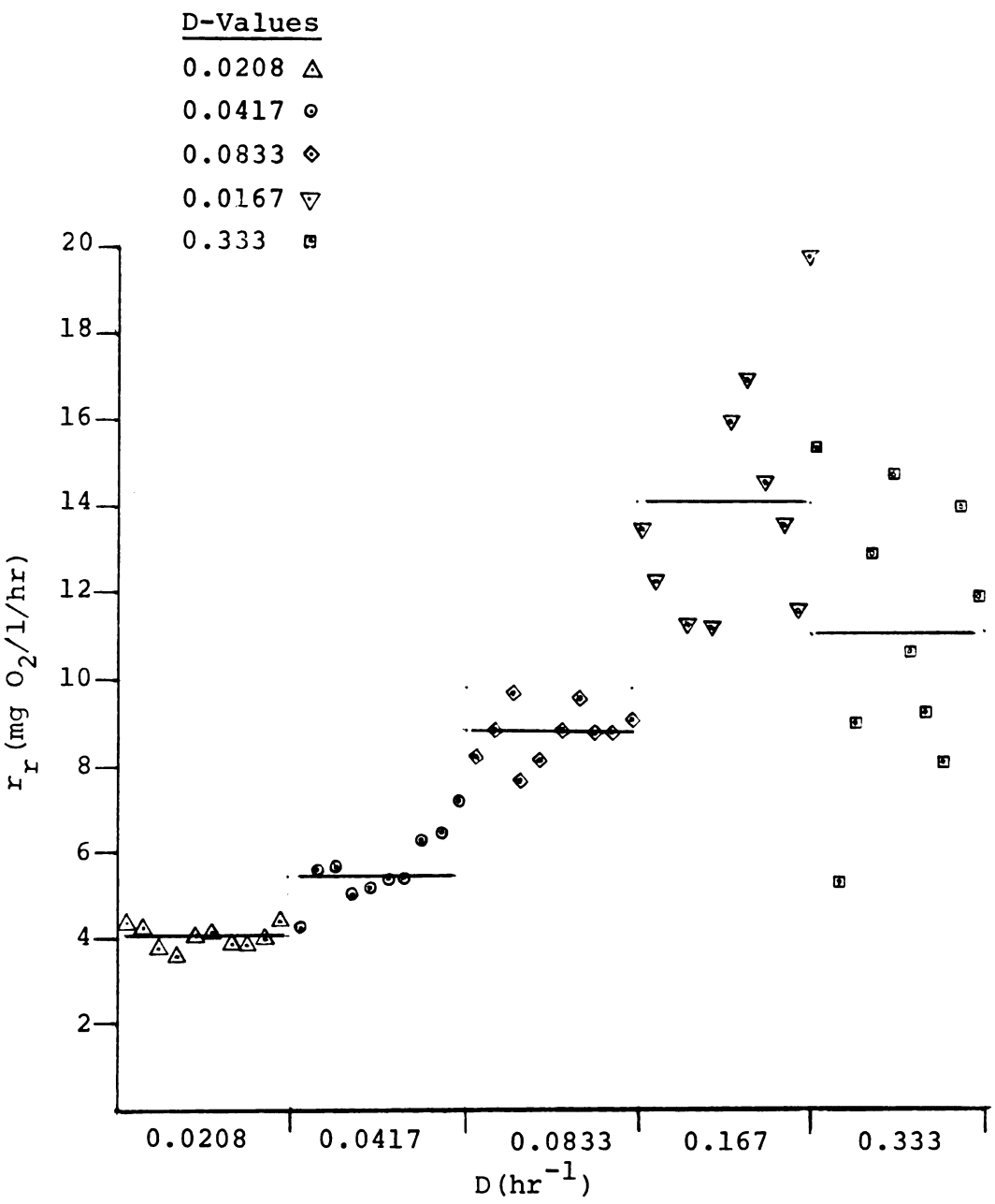

Figure 6. Variation of overall respiration rate (r_r) for a series of D-values--no temperature correction.

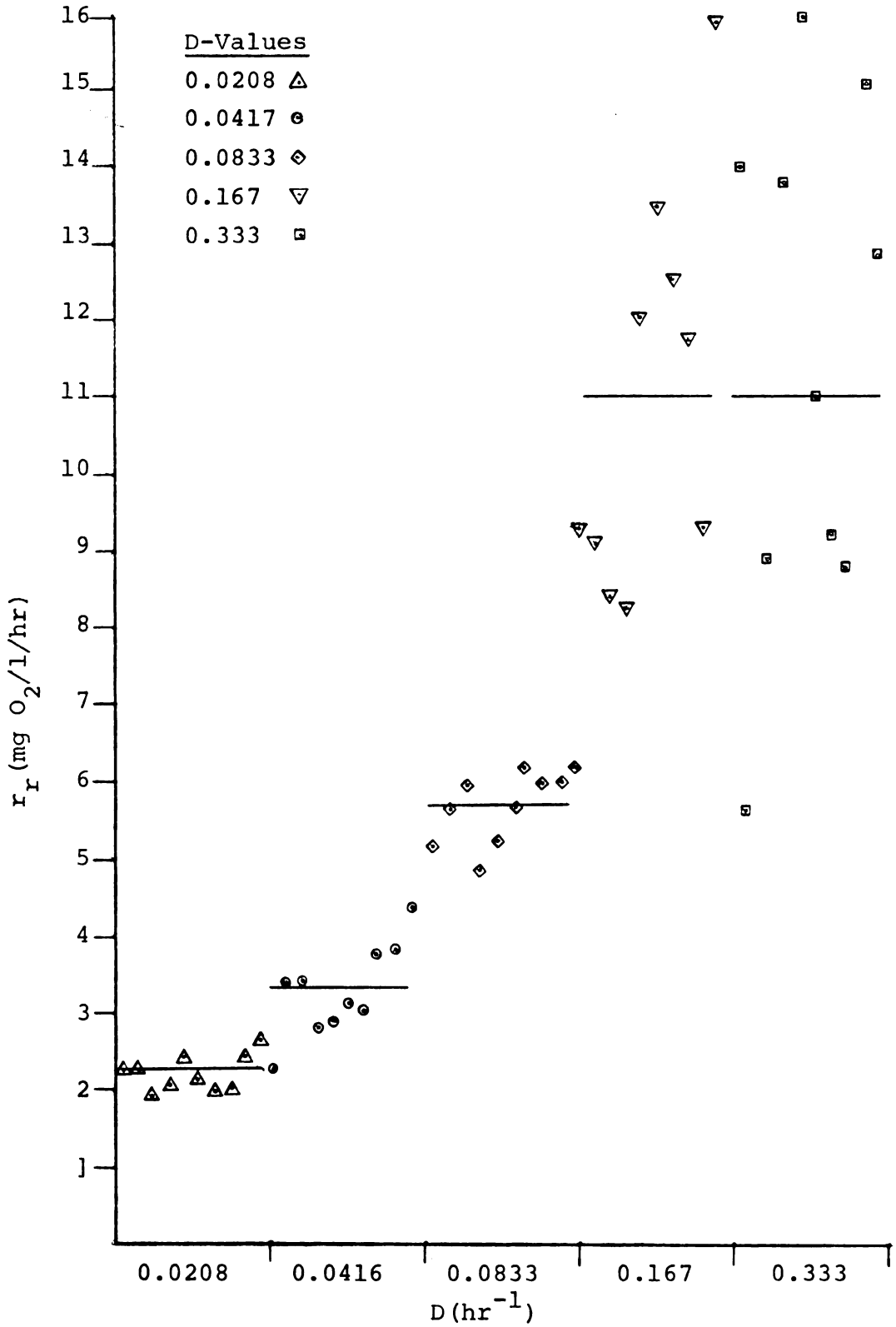

Figure 7. Variation of overall respiration rate (r_r) for a series of D-values--corrected to 20°C.

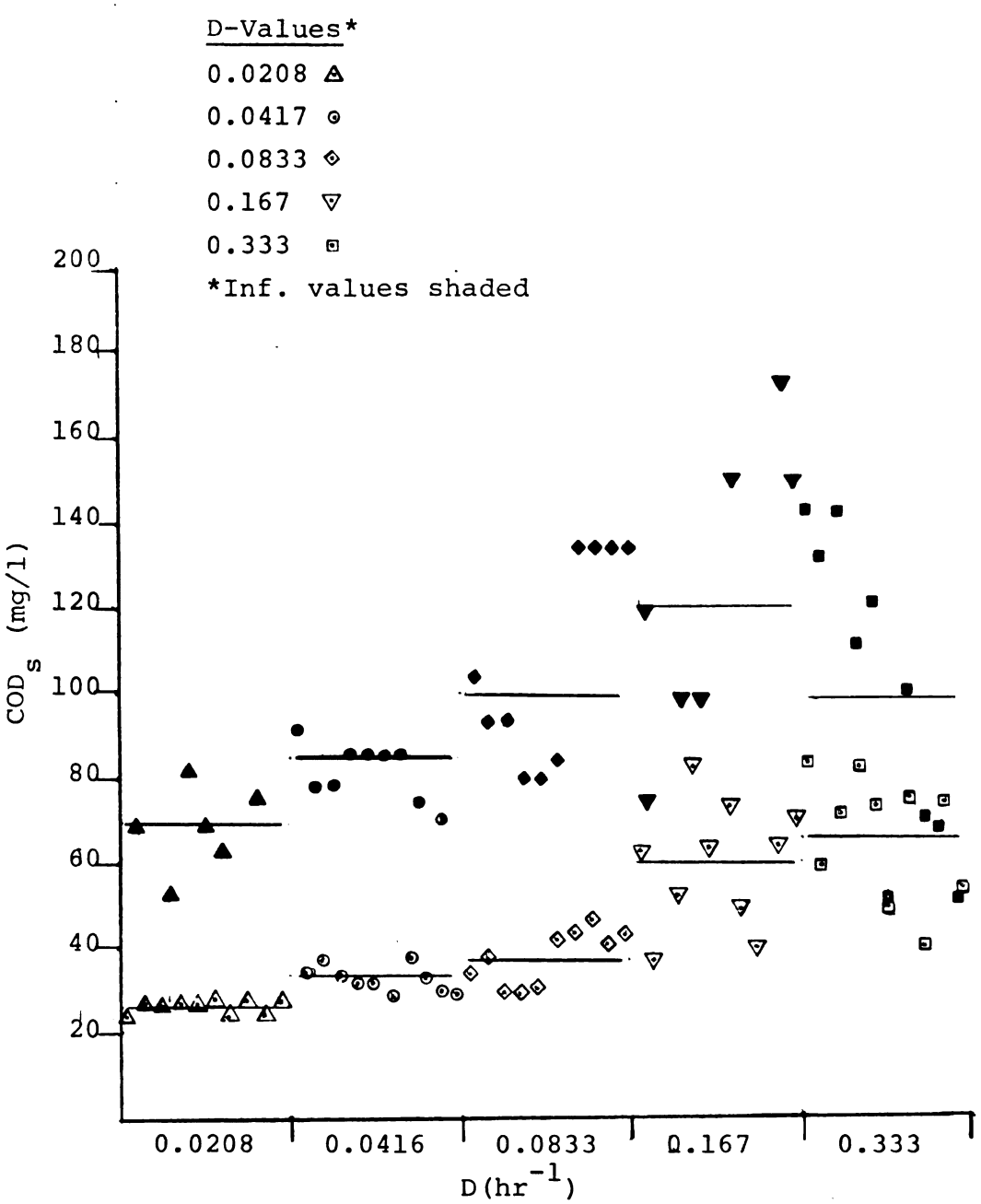

Figure 8. Variation of influent and effluent dissolved COD_S for a series of D-values.

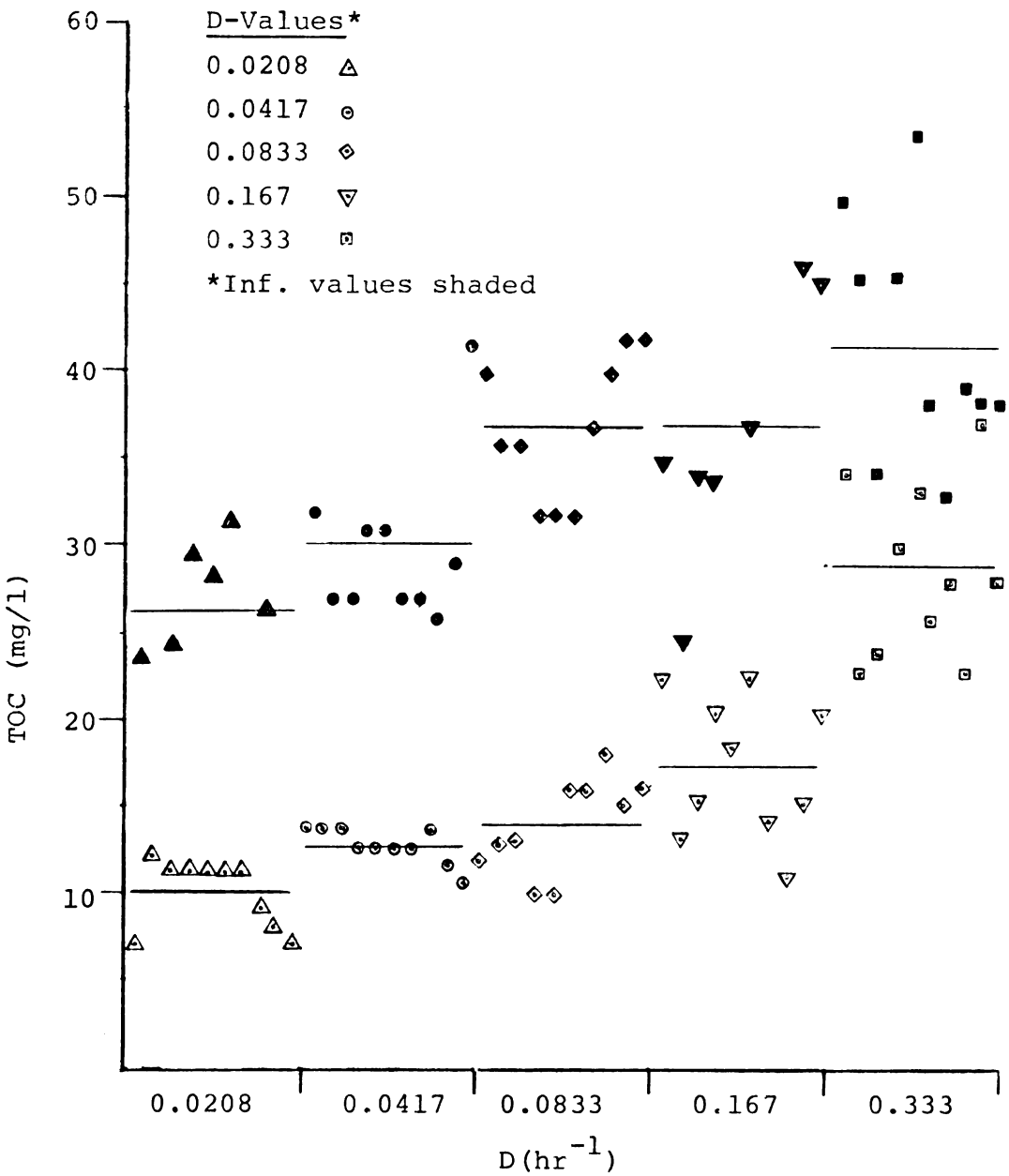

Figure 9. Variation of influent and effluent dissolved TOC_S for a series of D-values.

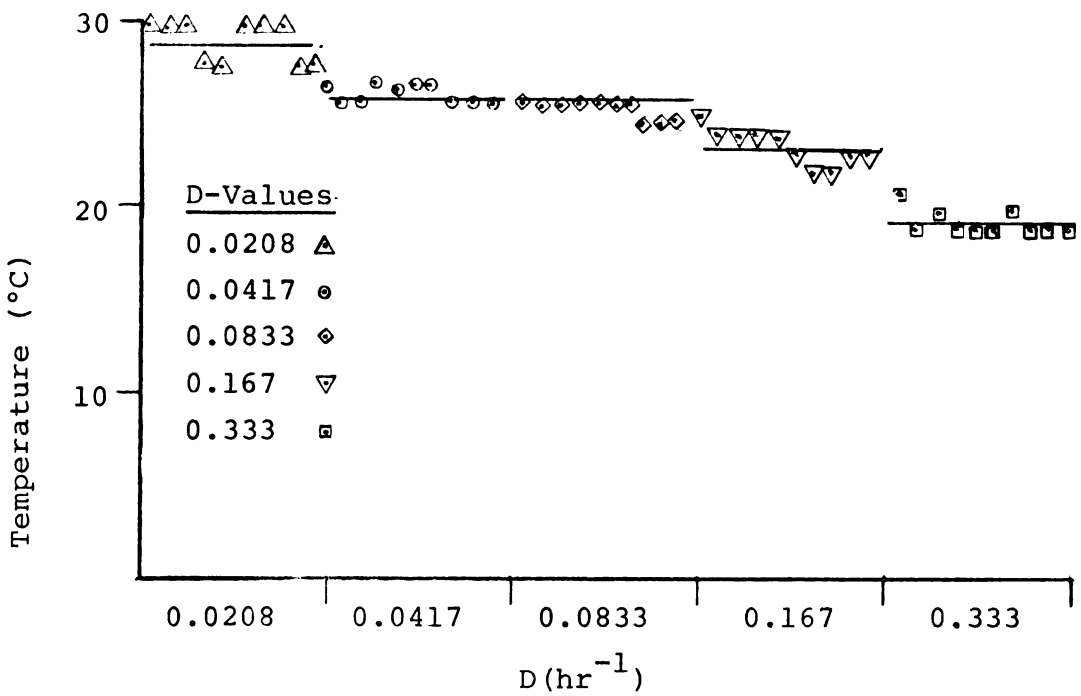

Figure 10. Variation of temperature (°C) for a series of D values.

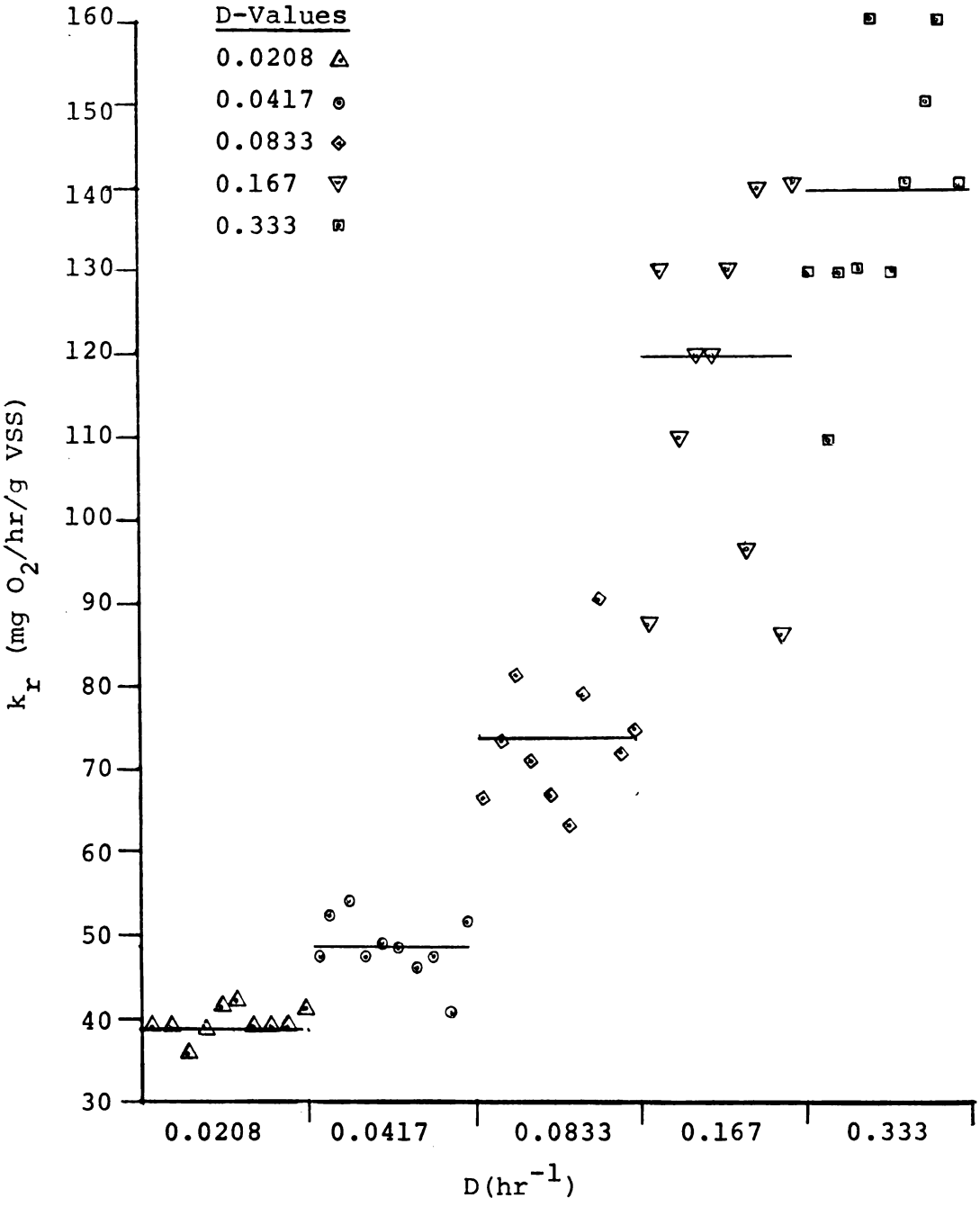

Figure 11. Variation of specific respiration rate (k_r) for a series of D-values--corrected to 20°C.

$$r_{20} = 10(r_t - 0.0315\Delta T) \qquad (X)$$

r_{20} = r_r at 20°C

r_t = the observed value of r_r at temperature T

ΔT = difference in temperature between observed value and 20°C

Plotting the average corrected values of k_r against the associated D values produced a curve as shown in Figure 12. It becomes evident that there exists a direct relationship between D and k_r as shown by the straight line. Thus only one point, that obtained for D = 0.33 clearly does not fit the straight line relationship. Equations V and VI have shown that for steady state conditions k_1 = D. The data presented in Figures 8, 9 and 11 suggest that steady state existed up to D = 0.167 i.e., up to a retention time of t_r = 6 hr. At D = 0.33, t_r = 3 hr., the performance of the reactor became very irregular.

It is therefore assumed that up to a value of 0.167/hr. D = k_1 so that up to D = 0.167 the D values represent the specific growth rate at which the continuous flow mixed culture was operating.

The linear portion of the curve shown in Figure 12 follows the equation

$$k_r = b + dk_1$$

as previously discussed (Equation II).

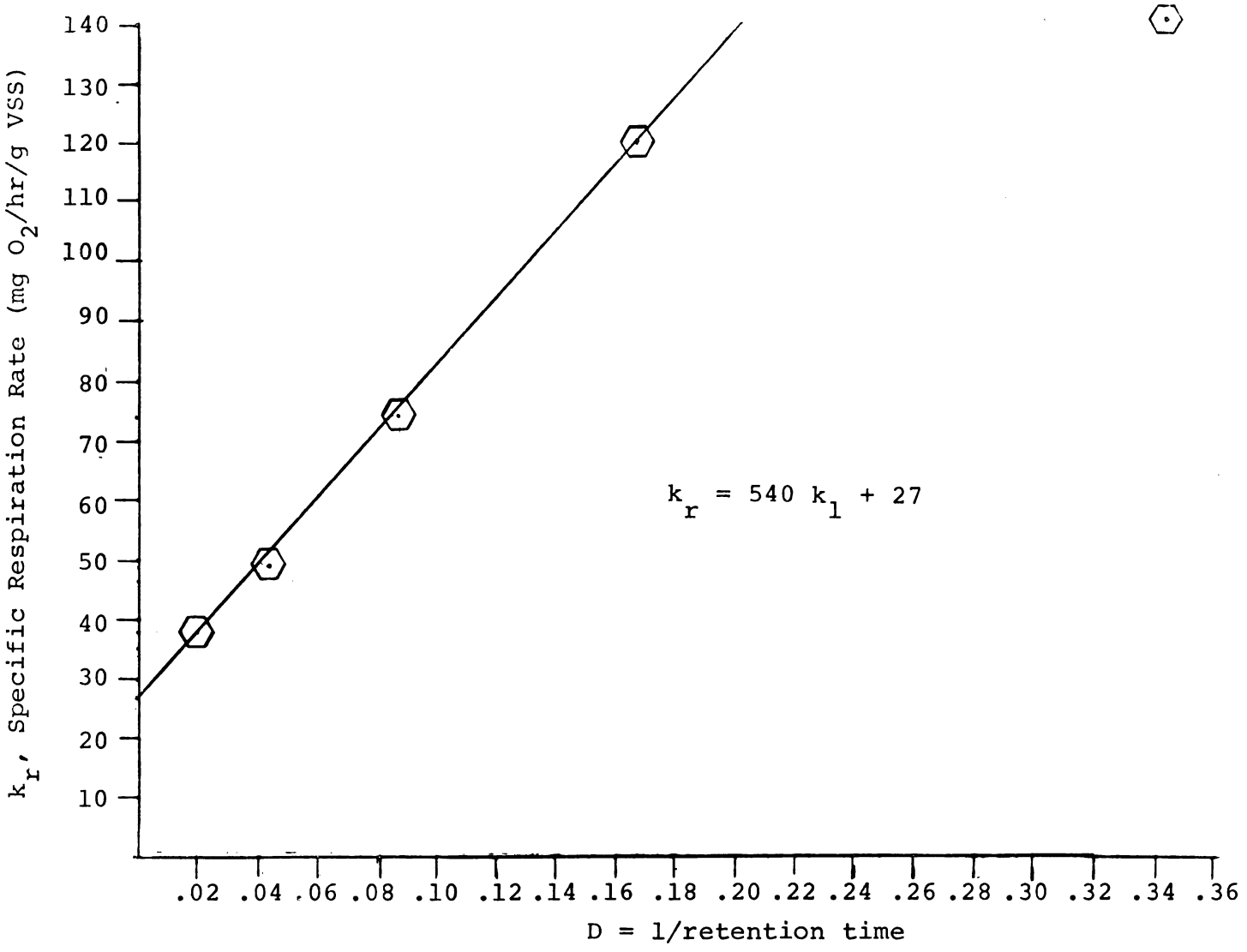

Figure 12. Plot of specific respiration rate (k_r) at 20°C vs D.

Using the least squares method, values of b = 27 mg O_2/hr/gram VSS and d = 540 mg O_2/gram VSS produced were obtained from the experimental data.

Thus,

$$k_r = 27 + 540\ k_1 \qquad \text{(XI)}$$

$$k_1 = \frac{k_r - 27}{540} \qquad \text{(XII)}$$

In general, the data demonstrate that at 20°C, 0.540 grams of oxygen were consumed per gram of volatile solids produced and that the endogenous respiration rate of the bacterial cells was 27 mg O_2 per gram of volatile suspended solids per hour when the specific growth rate (k_1) equals zero. For k_1 values up to 0.167 the curve shows that the specific respiration rates (k_r) were directly proportional to the specific growth rate (k_1).

A plot showing the relationship between D and dissolved substrate concentration remaining (COD_s and TOC_s) in the reactor indicated that the substrate concentration increased with increasing values of D (Table VII and Figures 13 and 14). In Figure 13 the dissolved COD_s concentration increases in direct proportion to D up to a D-value of 0.167. Just as in Figure 12 the COD_s concentration for D = 0.33 does not follow the straight line relationship. In fact, COD_s at D = 0.33 is much lower than would be expected by extending the straight line. The data support the previously made assumption that

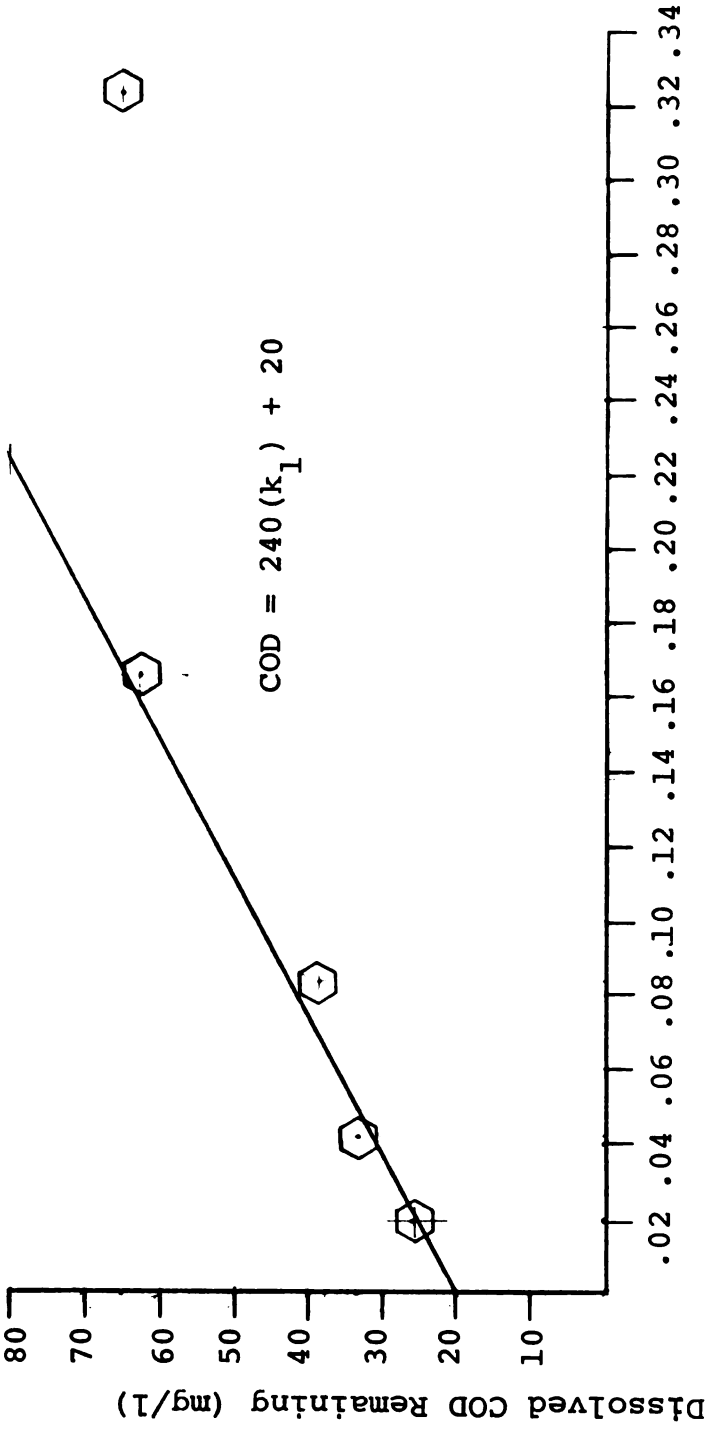

Figure 13. Plot of effluent dissolved COD vs D.

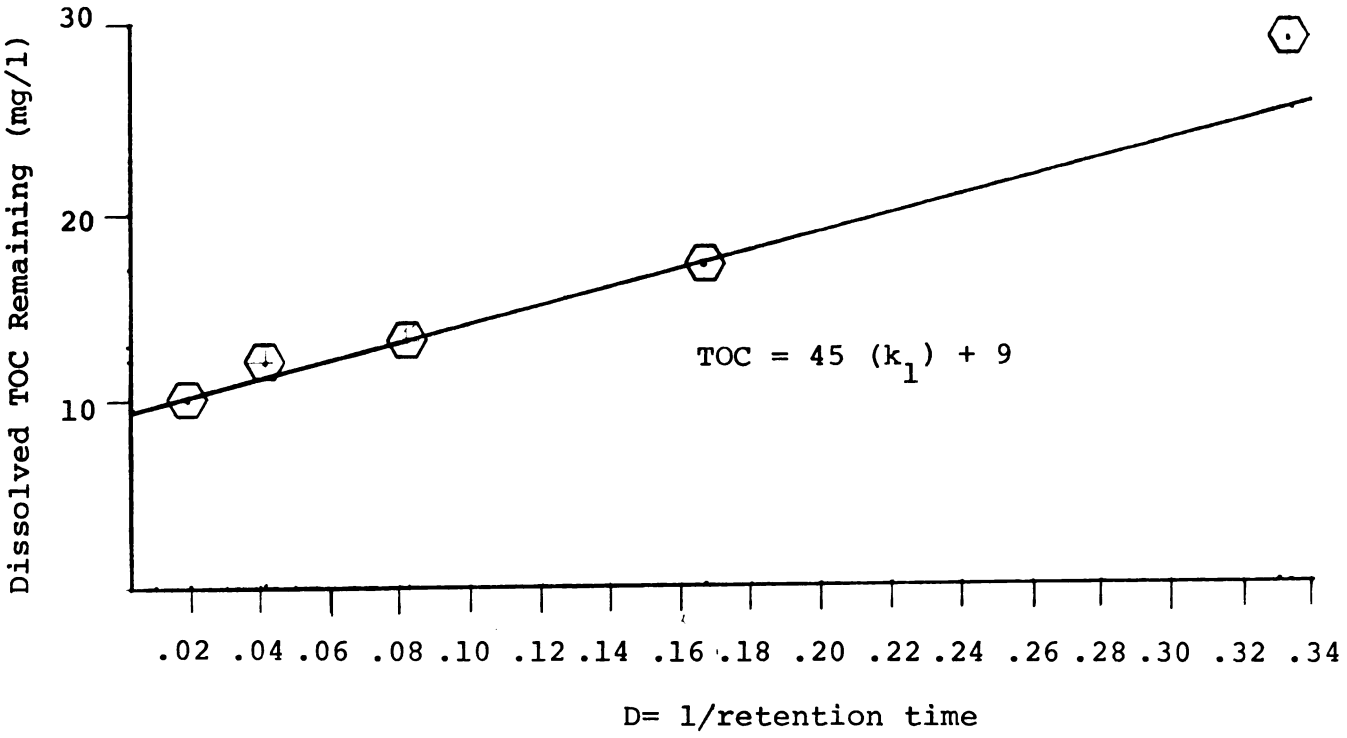

Figure 14. Plot of effluent dissolved TOC vs D.

steady state existed up to D = 0.167, so that D = k_1 up to D = 0.167.

The TOC_s data in Figure 14 show a similar relationship to D, except that in this case the TOC_s value for D = 0.33 is somewhat higher than the straight line extension would indicate.

As mentioned before, the relationship between k_1 and substrate concentration remaining has been expressed either as a direct proportionality up to a maximum value or as a hyperbola by the Monod and Teissier equations. The applicability of these concepts to the experimental data was tested.

Treating the relationship between specific growth rate and remaining substrate concentration as linear gave the equation

$$COD_s = 20 + 240\ (k_1)$$

as shown in Figure 13 for k_1 values up to 0.167.

For the TOC_s data shown in Figure 14, the following relationship was observed:

$$TOC_s = 9 + 45\ (k_1).$$

The Monod equation in rectified form is:

$$S/k_1 = s_n/k_M + (1/k_m)S \qquad \text{(XIII)}$$

Plotting S/k_1 against S produced a curve as shown in Figure 15. According to Equation XII, a plot of S/k_1 versus 5 should

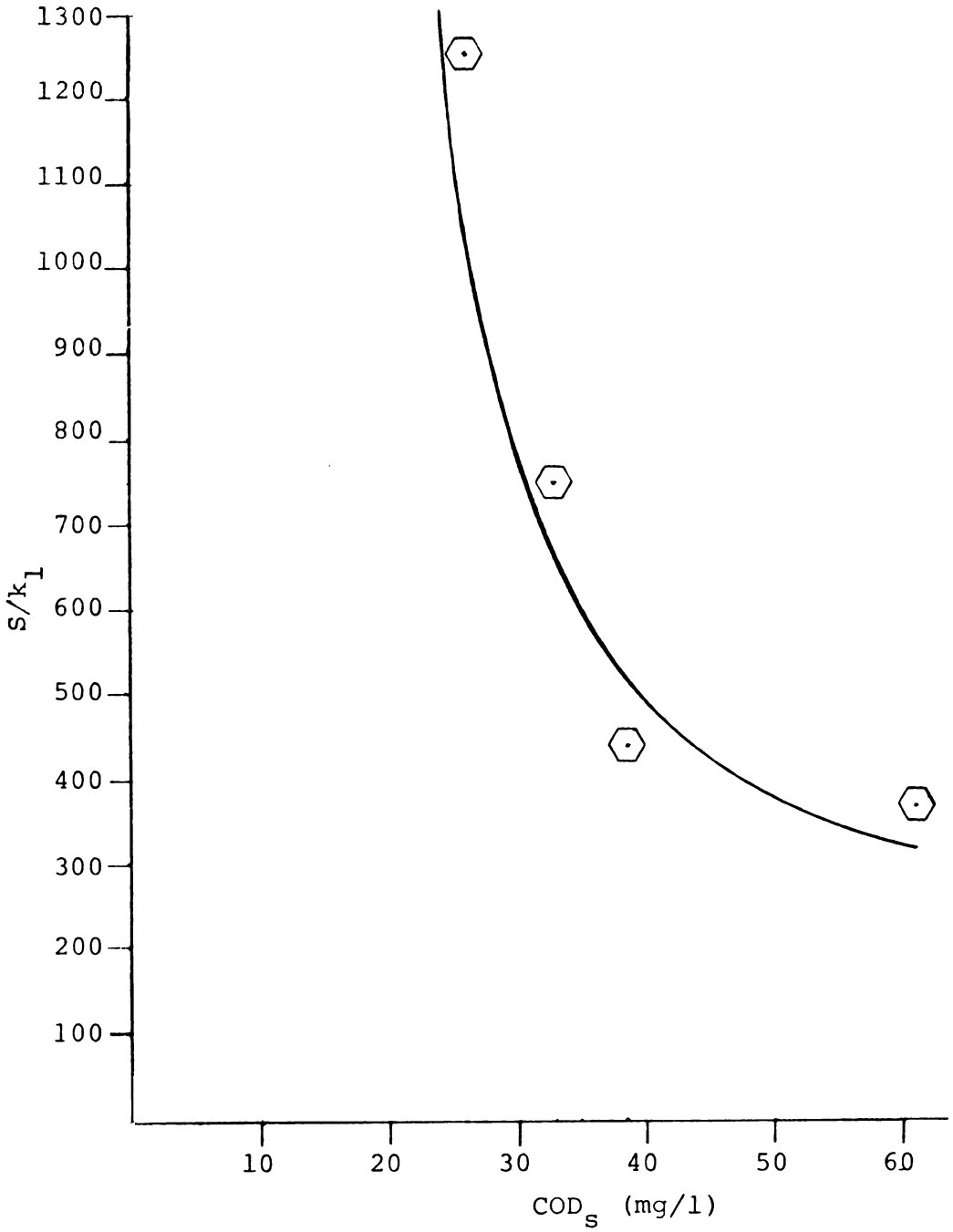

Figure 15. Plot of S/k_1 vs S.

produce a straight line where the y-intercept would be equal to Sn/k_m and the slope would be equal to $1/k_m$. Figure 15 shows that the experimental data do not agree with Equation XII.

In applying the Teissier equation it is necessary to obtain the two constants k_m and c from the data. In an attempt to use the Slope Method (26) for this purpose, it was found that the four sets of experimental data for k_1 and S were insufficient.

The data would thus indicate that using East Lansing wastewater, steady state conditions existed up to a retention time of 6.0 hours corresponding to a specific growth rate of $k_1 = 0.167^{-hr}$ and a specific respiration rate of 120 mg O_2/ hr/g VSS at an effluent substrate concentration of 61 mg/l as dissolved COD_s and 17 mg/l as TOC_s.

DISCUSSION

Experimentation has shown that under field conditions and constant flow, relationships between specific growth rate, specific respiration rate and substrate concentration existed for a domestic wastewater. Measurement of the soluble substrate (TOC_s and COD_s) and specific respiration rate (k_r) of a continuous flow mixed bacterial culture provided a linear relationship with the specific growth rate (k_1) under steady state conditions.

When overall variations of influent COD_s and TOC_s values are compared with the overall variations of r_r, k_r and effluent COD_s and TOC_s (Figures 6, 7, 8 and 9), it would appear that there is a direct relationship between these parameters, e.g., as the incoming COD_s and TOC_s increased so did r_r, k_r and effluent COD_s and TOC_s values. However, when the variation for an individual series of influent COD_s and TOC_s values are compared with those of r_r, k_r and effluent COD_s and TOC_s it can be seen that for the four series of D values (0.0208, 0.0416, 0.0833 and 0.0167) no such direct relationship can be found. Large fluctuations of incoming COD_s and TOC_s values could not be correlated with large fluctuations in values for r_r, k_r, and effluent COD_s and TOC_s. Such were also the

findings of a recent study (13) where a threefold increase of influent COD_s did not increase the effluent COD_s of an aerated system using artificial substrate and mixed culture operating at a 6-hour mean residence time ($k_1 = 0.167$). The observed data support the theory of steady state effluent COD_s and TOC_s values, regardless of incoming COD_s and TOC_s values up to D values of 0.167 ($t_r = 6.0$).

As mentioned before, Figure 12 shows that specific respiration rate is directly proportional to specific growth rate up to the values of $k_1 = 0.167$ and $k_r = 120$. At D = 0.33 the value of $k_r = 140$ is much lower than would be expected by extending the straight line. In fact, according to equation XI the specific consumption rate should have been

$$k_r = 27 + 540 \times 0.33 = 205 \text{ mg } O_2/\text{g VSS/hr}$$

at $D = k_1 = 0.33$. Apparently the mixed culture in the aeration tank was not capable of multiplying at a rate of $k_1 = 0.33$ per hour and therefore steady state operation was not possible at D = 0.33. The explanation may be found by going back to Figure 13 where the average substrate concentration in the reactor effluent was 65 mg/l measured as COD_s. According to equation XIII, at $D = k_1 = 0.33$ COD_s should be equal to $20 + 240 \times 0.33 = 100$ mg/l. Table VII indicates that at D = 0.33 even the incoming average COD_s reached only 98 mg/l. Thus it appears that in this case the substrate concentration in the incoming feed solution (primary effluent) was too low

to sustain a specific growth rate of k_1 = 0.33 per hour or a specific oxygen consumption rate of k_r = 205 per hour. The attainment of steady state conditions at D = 0.33 was therefore prevented by the existing limitation in the substrate concentration.

It would appear that the observed values of k_1 = 0.167/hr at a specific respiration rate of 120 mg O_2/hr/g VSS and a reactor substrate concentration of COD_s = 61 mg/l represent maximum values for East Lansing Wastewater.

The relationship between specific growth rate and dissolved TOC substrate concentration in Figure 14 was linear up to a D-value of 0.167 yet did not indicate a discontinuity beyond this point.

The decline of the incoming COD_s concentration as shown in Figure 8 at k_1 = 0.33 was probably due to the decline of the population served by the East Lansing Wastewater Treatment Plant. The final phase of this experiment was conducted at a time when the student body of Michigan State University had left the community for Christmas recess.

The problem of limited substrate measured as COD_s in sewage has been documented in an experiment where it was necessary to concentrate a domestic sewage prior to developing a curve showing the relationship between k_1 and S (5).

The fact that domestic wastewater is a relatively poor growth medium in comparison to prepared nutrient solutions became evident under field conditions where the highest

specific growth rate was k_1 = 0.167 at a specific respiration rate of 120 mg O_2/hr/g VSS and an effluent dissolved COD_s substrate concentration of 61 mg/l.

Data previously obtained from East Lansing primary effluent by manometric measurement of the overall respiration rate (r_r) found the highest specific growth rate to fall in the range of 0.28 to 0.33/hr. (22). The experimentally observed value of k_1 = 0.167 falls within the range of values used by Wuhrmann (27) and Pearson (16). These authors also attributed their lower k_1 values to the poor growth characteristics of sewage (Table VIII).

Information pertaining to specific growth rates in an actual domestic wastewater is limited (Table VIII). To make a comparison between the values obtained from the data in this paper and those discussed in Table VIII may not be warranted in light of the varying techniques and composition of nutrient substrates used. The data gained from this experimental work represent specific growth rates obtained under continuous monitoring of the overall respiration rate using a continuous flow respirometer and directly relating these values to specific respiration rates while other techniques relied on batch processes, manometric measurements or theoretical applications of reaction kinetics. A review of the literature has failed to provide additional data relating *in situ* respiration values to specific growth rates for

TABLE VIII

COMPARISON OF EXPERIMENTAL SPECIFIC GROWTH RATES (k_1)

Author and Reference	k_1 hr^{-1}	Temp. °C	Substrate	Type of Study
Pearson (16)	0.125	-	Sewage	Assumed operating cond. for activated sludge
Downing (2)	0.20	-	-	Assumed
Gaudy (5)	0.5-0.6	25	Artif.	Laboratory
Garrett (4)	0.20	20	Artif.	Laboratory
Schultz (22)	0.28-0.33	20	Sewage	Laboratory
Smith (23)	0.20	-	-	Assumption used for plug flow model
Wuhrmann (27)	0.126	-	Sewage	Assumed operating cond. for activated sludge

municipal wastewaters. It would appear that additional studies are needed to develop more reliable estimates of the kinetic characteristics of an actual wastewater source so that it will be possible to predict performance and offer guidelines to process design on a rational basis.

CONCLUSIONS

1. In situ continuous, polarographic measurement of the overall respiration rate (r_r) of a municipal wastewater with conversion to specific respiration rate (k_r) provided a relationship with the specific growth rate (k_1) under steady state conditions as described by the formula:

$$k_1 = \frac{k_r - b}{d}$$

Where b is equal to the endogenous respiration rate and d equals the amount of oxygen utilized per unit of volatile suspended solids produced.

2. At 20°C an endogenous respiration rate of 27 mg O_2 per hour per gram of volatile solids present was found with d = 540 mg O_2 consumed per gram of volatile suspended solids produced.

3. Specific growth rate (k_1) was found to be directly proportional with specific respiration rate (k_r) and effluent dissolved COD_s concentration up to a value of $k_1 = 0.167^{-hr}$ at a specific respiration rate of 120 mg O_2/hr/gram volatile suspended solids and COD_s = 61 mg/l. This maximum value for

k_1 was attributed to a substrate limitation in the incoming feed solution.

4. Specific growth rate (k_1) was found to be directly proportional with dissolved TOC_s concentration up to a value of $k_1 = 0.333^{-hr}$ at a TOC_s value of 29 mg/l.

5. Steady state conditions were in all probability established for retention times of 48 hr ($k_1 = 0.0208$), 24 hr ($k_1 = 0.0417$), 12 hr ($k_1 = 0.0833$) and 6 hr ($k_1 = 0.167$).

6. A relationship between specific growth rate and dissolved substrate concentration could not be established through the use of the Monod equation.

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