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THE FAST PRODUCTION OF METHANE BY ANAEROBIC DIGESTION

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OBJECTIVES

Conversion of the most abundant, renewable chemical, cellulose, to a highly preferred and greatly demanded fuel, methane gas, can be accomplished by anaerobic digestion. The supply of readily available or potentially available cellulose is sufficiently large to augment current natural gas consumption by about 20 percent. Since the productional cost of gas generated by anaerobic digestion has been on the economic borderline, and since it could be reduced by increasing the rate of the digestion process, a program of research has been initiated to verify the following concepts.

1. The step involving transfer of products from solution is rate-limiting and inhibiting in anaerobic digestion.

2. Anaerobic digesters can be optimized to further increase the rate of methane production.

3. Two-stage digestions, whereby unreactive solids are eliminated after the hydrolysis stage to lower the viscosity of the gas-producing stage, represent an effective means to achieve more rapid and economical methane production.

CONCLUSIONS

1. Numerous lines of evidence including temperature, pressure, agitation, viscosity, and viscosity-volatile acids effects support the phase transfer rate-limiting and inhibiting theory.

2. The theory predicts optimum kinetic performance under a combination of conditions including elevated temperature, reduced pressure, vigorous agitation, and reduced viscosity. As shown herein, it has led to kinetic performances superior to prior art efforts including a six-fold increase in the rate of reproduction for the methane bacteria and a two-fold increase in the volumetric rate of methane production.

3. The most important design implication of the theory is faster, more economic gas production can be realized in a two-stage digestion system where unreactive solids have been eliminated after the hydrolysis step so that the influent to the gas-producing stage possesses a low viscosity. The strengths and weaknesses of three such options are presented: (i) hydrolysis of cellulose to primarily glucose with Tv cellulase enzymes; (ii) the

hydrolysis of cellulose to carbohydrates and their conversion to volatile acids in an anaerobic step; and (iii) the inorganic acid-catalyzed hydrolysis of cellulose to glucose.

4. Since renewable biomass in the form of agricultural residues and urban refuse is readily available in quantities sufficient to have measurable impact on future gas supplies, and since productional costs are on the economic borderline, and since the promise for a technically improved process is very great; a highly aggressive program of R&D with a view toward creating faster, more economic anaerobic digestion systems is warranted at this time.

INTRODUCTION

Until very recently it was thought that two classic physicochemical problems, namely low rate and poor yield, prevented serious consideration of anaerobic digestion as a technically valid and economic option for supplementing much needed supplies of natural gas. These problems seemed to overwhelm some otherwise conspicuous advantages that the technology possesses.

A foremost advantage of anaerobic digestion is that it represents a viable natural pathway for creating the most preferred of hydrocarbon fuels, methane, from an abundant, renewable feedstock, cellulose. Methane is preferred over cellulose as fuel because of its greater energy content: methane has a combustion energy of 50 kjoules per gram while the value for cellulose is only 15 kjoules per gram. It is true that over the past two hundred years, increasingly industrialized societies have demanded more and more of the higher energy content, but less available, hydrocarbon fuels. Therefore, from both the existing crisis atmosphere and the longer range historical perspective, there is good reason to wonder whether or not the anaerobic pathway might at one point be useful in meeting the societal requirement for high grade fuels.

A second major advantage of anaerobic digestion as a conversion technology is that the substrate, cellulose, is so abundant, and the potential for contributing in a significant way to the

future need for quality fuels is thereby very great. For example, in this country major sources of supply include the organic fraction of urban refuse (100 million tons),[1] residues from agricultural crops (309 million tons),[2] and the biomass from plantations on idle and available crop, range, and forest lands (1210 million tons).[3] Assuming an average energy content of 6,500 Btu lb^{-1}, these sources of supply (a total of 1.63 billion tons) represent about 26 percent of total U.S. consumption in 1975, and it was about 80×10^{15} Btu.

However, when a 50 percent collection efficiency and a 50 percent bioconversion efficiency are imposed, the available supply in the form of gas reduces to about 6.5 percent of consumption; but since natural gas represents only about a third of total consumption this supply would still represent about 20 percent of total natural gas consumption--a quantity large enough to have prevented the 1976-77 shortfall. It should also be mentioned that unlike coal, oil, or natural gas, this potential supply is above ground and perpetually available.

The third major advantage of anaerobic digestion as a delivery technology is purely economic, and its potential impact as an industry is significant. If it is assumed that the future price of gaseous energy will be in the region above \$3.00 per 10^6 Btu, then the supply of cellulose is sufficient to support a \$16 billion per year bioconversion industry. With agricultural residues as substrate, our own pre-design estimates suggest that a \$3.22 per 10^6 Btu figure for gas of pipeline quality is

achievable. This cost would cover expenditures for the following unit processes: agricultural residue collection, storage and transportation, shredding, oxidative pre-treatment, fast processing in a single-stage digester, gas purification, and disposal of liquid effluent back onto the fields. If instead the gas were consumed locally and purification were unnecessary the cost would be reduced to $2.35 per 10^6 Btu, and energy efficiency would also be improved. These productional costs are consistent with other recent estimates.[2-5]

A fourth advantage of anaerobic digestion as a delivery technology is that its environmental impact would be minimal especially relative to other options, and the employment of this technology is entirely consistent with the movement toward clean air and water and toward responsible management and husbandry of agricultural lands. Methane is the cleanest burning, least polluting of hydrocarbon fuels because of its high energy content and because it mixes completely with air in combustion chambers. Furthermore, the liquid effluent from the digesters contains bacterial solids rich in nitrogen, phosphorous, potassium, and stabilized cellulose solids that have superior water retention and soil conditioning capacities. Therefore, it would appear that the highest use of digester effluent is as fertilizer and soil conditioner, and that it should be returned to the land. If in fact these nutrients and stabilized solids are returned to the land so that a partially

closed cycle is created, it is difficult to understand how agricultural lands would be harmed by energy farming.

Given these significant advantages including a high energy content gaseous fuel, an abundance of celluloses, economic incentives, and a favorable environmental impact, we turn now to the rate and yield problem. The problem may both be stated and understood with a typical operating example: for a conventional mesophilic digester being loaded at a standard rate of 0.15 lb ft^{-3} day^{-1} of volatile solids, and operating at 50 percent conversion efficiency, it takes about 3.5 years for a unit volume of the digester to produce an equivalent volume of liquid methane fuel (at -164°C, $\rho(CH_4)$ = 0.415 g cm^{-3}). These lackluster kinetics have automatically invoked relatively high operating and capital costs with the implication of an expensive gaseous product. Thus a reasonable approach for attacking the borderline economics of gaseous fuel production by anaerobic digestion necessarily involves a basic re-examination of process kinetics.

Work performed in the sixties and earlier, along with papers published so far in the seventies indicate that while yield performance is quite obviously governed by the rate of hydrolysis of cellulose to carbohydrates, the overall volumetric rate limitation is related to the conversion of volatile acids to methane and carbon dioxide.[6] On a representative basis the kinetic mechanism includes: (i) a phase transfer of solid phase cellulose to soluble carbohydrates such as glucose, cellobiose, and xylose via reactions moderated by cellulase enzymes; (ii) the

conversion of carbohydrates to lactic acid[13] and then to volatile acids--predominantly acetic, propionic and butyric--in reactions moderated by the acid bacteria; (iii) the transformation of these acids to methane and carbon dioxide in solution by the methane bacteria; and (iv) the physical transfer of these moderately soluble products into the gas phase of bubbles originating in the solution. In this report, we shall concentrate on the nature of the volumetric rate limitation reserving for later consideration the recent progress[11-13] that has been made on the yield question.

It is usually assumed that the third step in the mechanism, the conversion of volatile acids to methane and carbon dioxide, is rate limiting. The primary evidence supporting this contention is a buildup of the volatile acids as the mean residence time of the methane bacteria is reduced from about 15 to about 2 days.[7,9,10] A representative example of this kind of evidence published by McCarty[7] for mesophilic municipal residue digestion is shown in Figure 1.

Since the precursor of about 70 percent of the methane formed in anaerobic digestion is acetic acid,[8] the mechanism for the rate limitation is usually represented by,

$$HAc(aq) + B_m \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} HAc - B_m \xrightarrow{k_2} CH_4(aq) + CO_2(aq) + B_m \qquad (1)$$

where HAc(aq) is aqueous acetic acid, B_m is a methanogenic bacterium, and the k's are specific rate constants. With the usual steady assumption employed for enzyme kinetics this mechanism

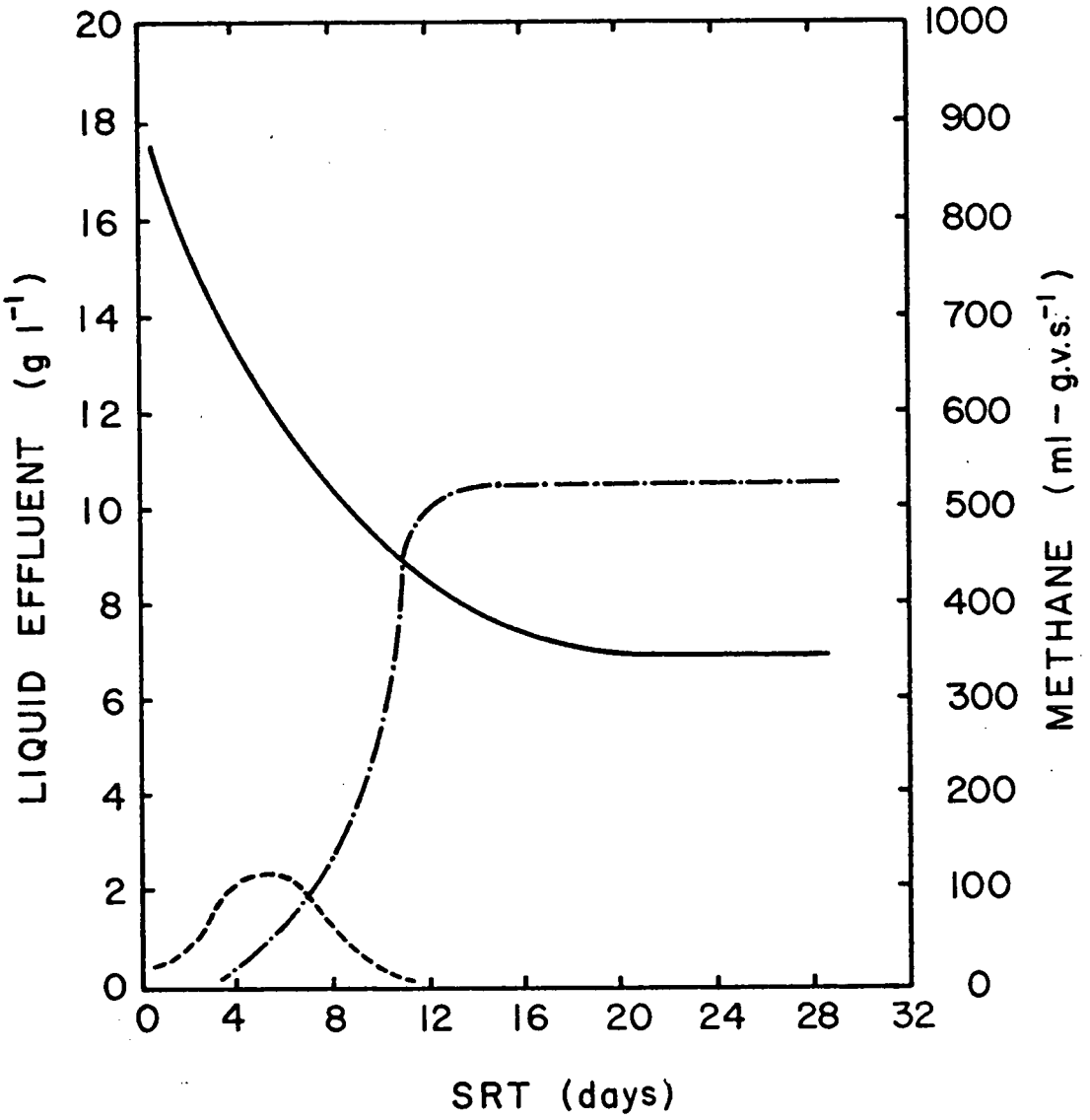

Figure 1. Relationship between retention period and effluent concentrations for sewage wastes: volatile solids ————————; methane —— – ——; and volatile acids – – – – – – (Ref. 7, Fig. 3).

leads to the familiar Monod rate expression,[14]

$$-\frac{d(HAc)}{dt} = \frac{k_2(B_m)(HAc)}{K_s+(HAc)} \qquad (2)$$

with K_s equal to $(k_2+k_{-1})/k_1$, the substrate concentration at which the rate attains its half-maximum value. It is important to remember that the mechanism in Eq. 1 and the rate expression in Eq. 2 could apply to a rate limitation related to either passage of acetic acid through the bacterial cell wall or to an enzyme process occurring inside the methane bacteria. Exactly the same kinetic treatment is appropriate for either case and the anaerobic literature does not distinguish between which is actually rate-limiting. For the former situation the bacterial concentration term in Eq. 2 would reflect surface area available for permeation of acetic acid while for the latter case the term would reflect the internal concentration of key enzymes.

Some values for the kinetic constants of Eq. 2 for acetic acid as determined by Lawrence and McCarty[15,16] are found in Table 1.

Table 1. Kinetic constants for acetic acid utilization at two loading rates (Refs 15 and 16)

Feed Concentration ($mg\ \ell^{-1}$)	k_2 (day^{-1})	K ($mg\ \ell^{-1}$)
1568	12.3	207
3135	6.5	156

These values in addition to illustrating the unfavorable kinetics of the methane bacteria (e.g. minimum regeneration period of 1.4 to 2.7 days) also demonstrate another general phenomenon of anaerobic digestion: the turnover constant, k_2, or the specific rate for methane generation decreases as loading is increased.

Andrews[17,18] has proposed that this kind of inhibition was the result of increasing concentrations of unionized volatile acids, and he added an empirical correction term to account for the exponential decline in specific rate which occurs as acid concentrations increase,

$$-\frac{d(HAc)}{dt} = \frac{k_2[B](HAc)}{K_s+(HAc)+\alpha_i(HAc)^2} \qquad (3)$$

where α_i is the inverse of Andrew's inhibition constant, K_i. Thus, Eq. 3 is a quantitative expression of the earlier suspicion by Buswell[19] that the volatile acids themselves were somehow toxic to the methane bacteria.

However, a question remains as to whether or not the exponential increase in volatile acids concentration with loading is and of itself a primary cause of the inhibition or whether it is only a secondary manifestation of some other more primary physical phenomenon. For example, it is true that the bacterial concentration also increases with loading. Could the bacterial mass itself be the cause of the observed inhibition--or at least a part of the cause? It is also worthwhile noting that a unique feature of the methane bacteria is that they produce a gaseous

product which must be transported away from the bacterial cell walls by gas bubbles originating in solution, and from Equations 2 and 3, the rate of gas production, or more precisely, bubble production per bacterium should increase as the volatile acids concentration increases. It is the necessary physical proximity between the bacterial cell wall and the nearby bubble membranes that initially led us to suspect that the inhibition factor in Equation 3 might be related to a reduction in the cellular surface area available for permeation processes.

HYPOTHESIS

The rapid removal of products away from the reaction site is universally recognized as a very good idea among chemists and chemical engineers. Our view of the nature of the volumetric rate limitation in anaerobic digestion as presented here and elsewhere[20] consists of two perspectives. The first is that transfer of the more soluble gaseous product, carbon dioxide, is ultimately rate-limiting in the digestion process. If this is the case then the maximum rate of gas transfer, R_t, per unit cross-sectional area across the bubble wall is

$$R_t = \frac{D}{\ell} (C_s - k_H(T) P) \qquad (4)$$

where D is the diffusion coefficient; ℓ is the width of the bubble membrane; $k_H(T)$ is Henry's constant for carbon dioxide which is a function of absolute temperature, T, and P is the carbon dioxide partial pressure.

Equation 4 is derived from Fick's First Law for the diffusion of a gas from solution to and through a bubble membrane. Our mental construct of a bubble as shown in Figure 2 is the same as the one initially developed by Lewis and Whitman.[21] The model assumes the existence of a membrane consisting of two films: on the gas side there is a relatively stationary film representing the constituents of the gas phase, and similarly on the liquid side there is a second stationary layer representing the constituents of the liquid phase. Since air bubbles in

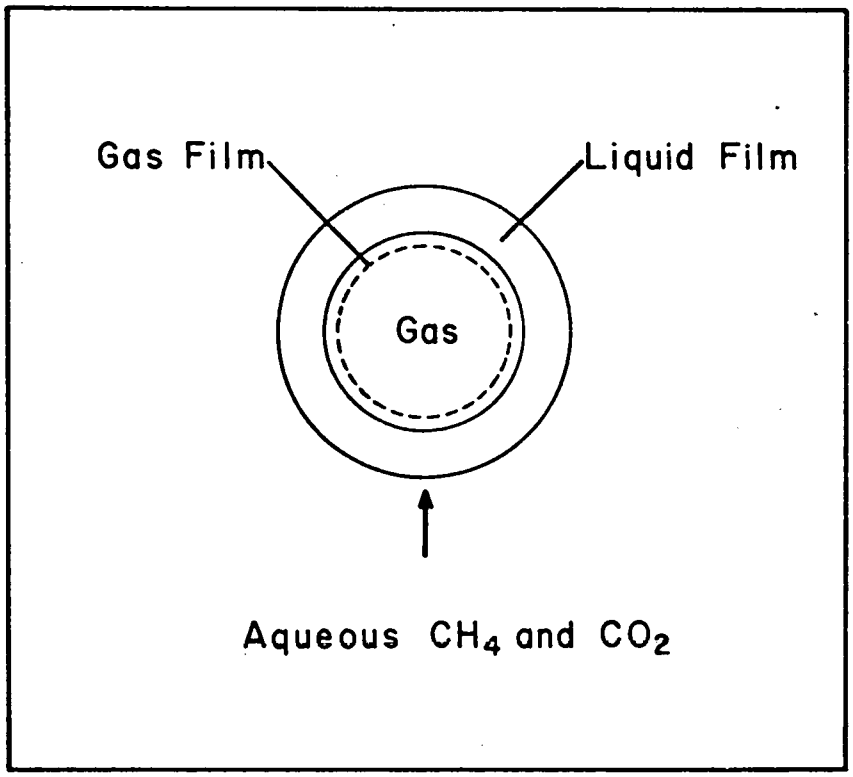

Figure 2. The Lewis and Whitman construct of absorption by a bubble (Ref. 21).

water are responsive to external electric fields, it has been suggested[22] that oriented dipoles of the water molecules account for the structure on the liquid side of the membrane.

Equation 4 predicts that gaseous transfer should be facilitated by (taken either singly or in combination) elevated temperatures which decrease $k_H(T)$; by agitation which reduces the film thickness, ℓ; and by a low carbon dioxide partial pressure which increases the magnitude of the concentration gradient across the bubble membrane. Since the diffusion coefficient is inversely proportional to viscosity, a digestion medium of low viscosity should also enhance the prospects for rapid gaseous evolution.

The second part of our hypothesis is, that under the condition of a high concentration of acid, the gas production per bacterium becomes so rapid relative to nearby transfer capacity that the methane bacteria either partially or completely smother themselves in their own bubble froth. For example, the results from one of our experimental systems[20] indicate that each methane bacterium produced an STP volume of gas equal in size to its own volume about every ten seconds. The acetic acid concentration for this system was 0.45 g ℓ^{-1} and it seems plausible, if the acid concentration were further increased, that at some point, gaseous evolution would become so profuse that the diffusion of acid to the individual bacteria would become either partially or completely jeopardized by the existence of nearby microbubbles.

There are also some good reasons to suspect that the concentration of bubbles will be especially high in the region near the bacteria. From Fick's laws of diffusion the concentration gradients of products must be greatest nearest their point of production-implying around the bacteria. Therefore, unless there is a high degree of supersaturation in the region of the bacteria--and a high degree of supersaturation seems unlikely in such impure solutions--it would appear reasonable that bubble formation will simply have a more favorable probability of occurring in this region of highest insoluble product concentration.

A second, perhaps even more compelling reason to suspect that bubbles would be formed near the bacteria is related to the fact that microspheroid particles such as bacteria and bubbles in water solution containing electrolytes are, in general, electrically charged. The electrical double layer between the microspheroid surface and the bulk of the solution is the result of selective orientation of either positive or negative ions onto or very near the microspheroid surface. For bacteria the electrical forces originate from both ionic and dipolar surface groups,[23] and for bubbles, they appear to originate entirely from the oriented water dipoles on the liquid side of the bubble wall.[22,24] Under these circumstances the charged bacterial wall would seem to be an almost ideal nucleation site since the effect that charged particles have on promoting bubble formation such as observed in bubble chambers is well established.

At this point we have a rate limitation imposed by phase

transfer and an inhibition caused from the interference of diffusion processes to bacteria because of nearby bubbles. Let us now examine the interplay between the rate limitation and the degree of inhibition. Forces acting on a bubble as it moves through the digester arise from its drag, weight, and buoyancy. By direct substitution and manipulation, it may be shown that the velocity of such a bubble moving from solution is, approximately,

$$v = \frac{2r^2 g(\rho-\sigma)}{9\eta} \qquad (5)$$

where r is the bubble radius, g is the gravitational acceleration constant, ρ is the density of the medium, and σ is the density of the bubble. Thus it is seen that the rate limitation by transfer of gases into the bubble also leads to greater inhibition because the bubble will stay in the region of the bacteria for a longer time period due to its smaller radius.

Taking this line of thought a step further and simultaneously invoking the Le Chatelier principle there is a reason to suspect that as the acid concentration is increased, and the gas transfer requirement becomes greater, the nearby product bubbles would, on the average, be smaller and thereby even more numerous and effective in promoting inhibition. Since the capacity for phase transfer is dependent upon the surface area of the bubbles, and since smaller bubbles have larger surface area-to-volume ratios, from a thermodynamic perspective it is reasonable to surmise that the system would have to react by creating a

larger number of smaller bubbles when the capacity for phase transfer is stressed, as for example, with a high acid concentration. This is especially probable because, as we have seen, supersaturation is unlikely and temperature, pressure, and agitation are all externally controlled so that the creation of smaller bubbles is virtually the only automatic mechanism for increasing the transfer capacity under a stressed condition of elevated acid concentration. An image of this phenomenon is presented in Figure 3 where facilitated transport at low acid concentrations is seen to graduate into inhibition and then to complete smothering as the acid concentration becomes progressively greater.

The value in part of a hypothesis such as the preceeding one is that it permits testing on at least three different levels. First, we may test its correctness by determining the effects of temperature, pressure, agitation, and viscosity upon observed reaction rates. The following literature review is a preliminary effort toward this end. Secondly, being reasonably well satisfied that the facts justify the hypothesis, we may proceed to use the practical implications of the theory to attempt to achieve more rapid, and thereby more economic, gas production. The details of such an experiment, whereby relatively rapid gas production has been observed, are also presented herein. Finally being reasonably confident of the theory on both basic and applied grounds, we proceed to explore the most important of the design implications. In doing all of this the authors are aware that mechanistic constructs such as the ones previously

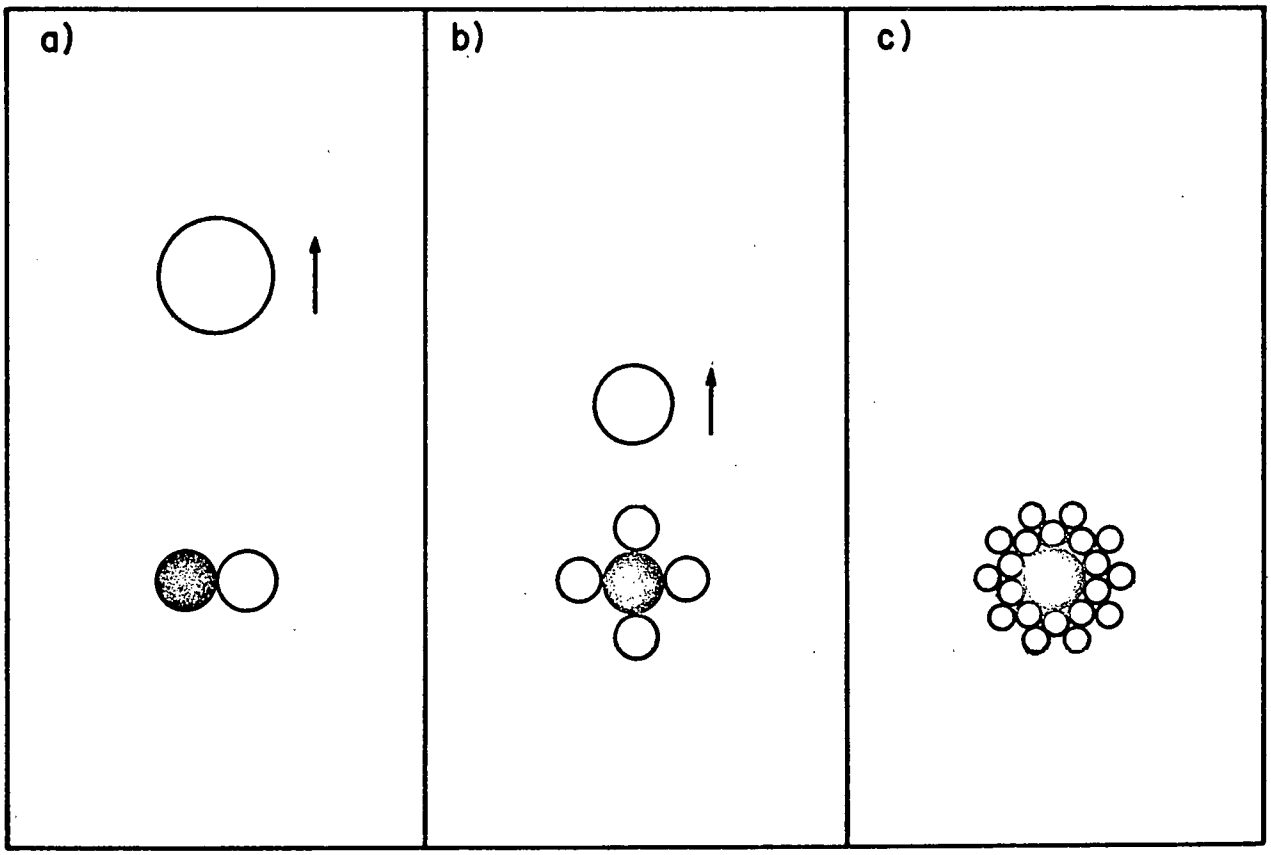

Figure 3. Facilitated transport graduating into inhibition and then smothering as the volatile acid concentration becomes greater. The trend toward inhibition b) and smothering c) is promoted at higher acid concentrations because as the system reacts to create increased gas transport capacity, the bubbles become smaller (the surface area-to-volume ratio of a bubble is equal to 3/r) and they subsequently depart from the solution more slowly.

presented can never be completely proven--they can only be shown to be reasonable or not on existing physical evidence. The usefulness of such a theory is as a means to organize thought patterns and to systematically guide an experimental effort for both pure and pragmatic ends; and we recognize that new facts, either unknown by us or to be discovered in the future, may require modification of theory or may prove it to be entirely incorrect. With this in mind, the following information seems nonetheless to be sufficiently supportive that a potentially productive avenue of investigation is suggested.

Temperature Evidence In the past most laboratory studies of the effect of temperature on digestion rates have been conducted at a total pressure of about 1 atm, the carbon dioxide partial pressure being about 1/3 atm. However, since Henry's constants and carbon dioxide concentrations for digesters are unknown, we compared the relative inverse solubility in pure water to the relative digestion rates observed by Golueke[25] for digestion of volatile solids and by Lawrence and McCarty[15,16] for acetic acid as shown in Figure 4. The use of the relative inverse solubility in this manner involves the assumption that both C_s and C_ℓ, the respective solution and membrane concentrations of carbon dioxide, decrease proportionately as the temperature is increased. Under these circumstances Fick's First Law,

$$R_t = \frac{D}{\ell} (C_s - C_\ell) \tag{6}$$

may be rearranged to give

$$R_t = \frac{D}{\ell} \frac{(C_s/C_\ell - 1)}{C_\ell} \tag{7}$$

and since the numerator is unchanging with temperature the transfer rate should be inversely proportional to the CO_2 solubility. We regard the observed correlation in the mesophilic temperature range as evidence supporting a rate limitation by gas transfer.

Similarly, the activation energy from the rate data in Figure 4 also supports a diffusion controlled, as opposed to a chemical reaction, rate-limiting process. When the temperature

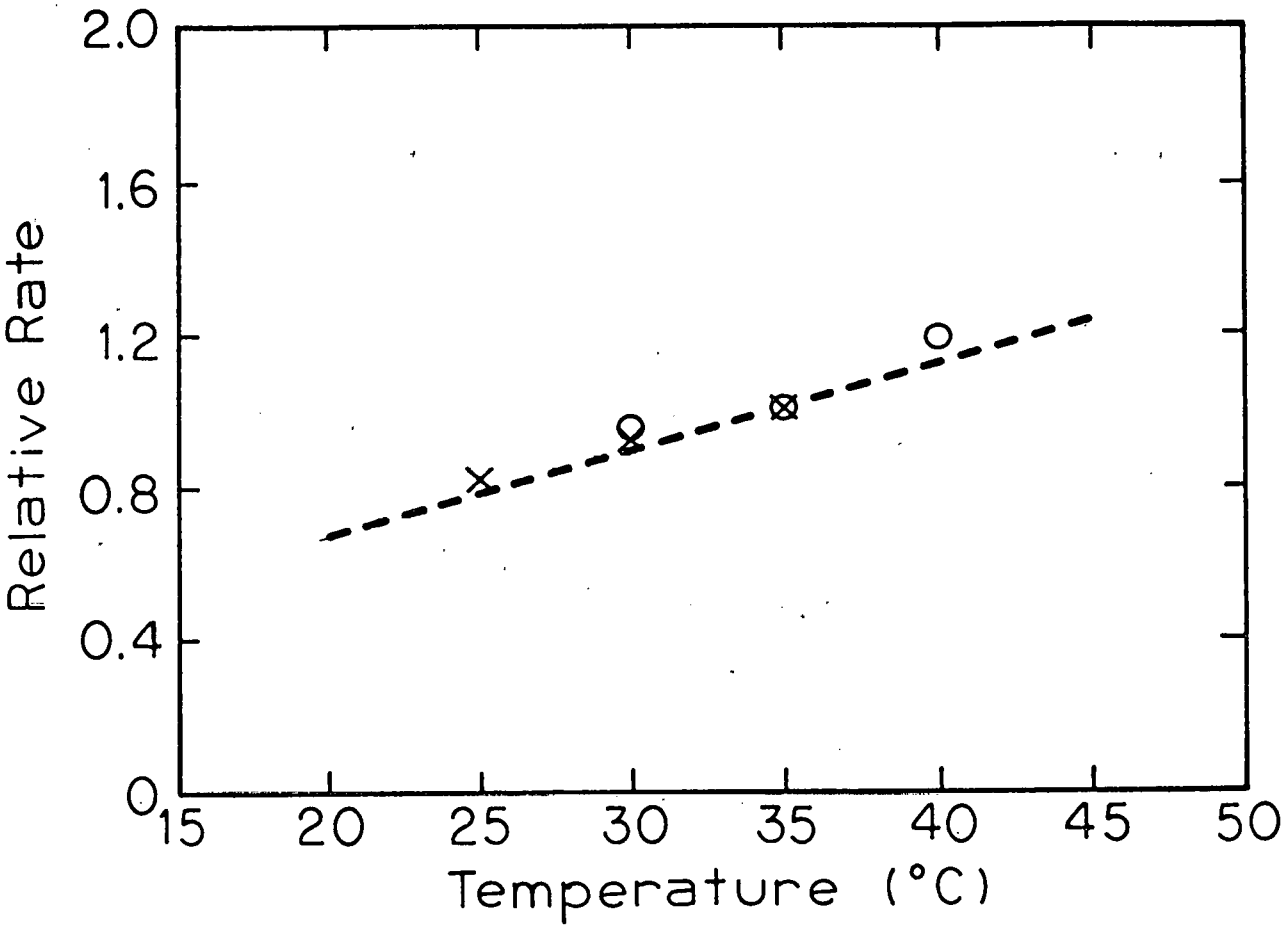

Figure 4. Relative digestion rates and inverse carbon dioxide solubility as a function of temperature normalized at 35°C: (dashed line) $1/CO_2$; (X) data of Lawrence and McCarty (refs 15 and 16); and (O) data of Golueke (ref 25). Lawrence's data for effluent acetate concentrations are extracted for digesters 2, 4+5, and 6+7 at SRT=12 days. Data on total gas production are extracted from Golueke's Table 1.

range is small, the activation energy may be calculated using the empirical Arrhenius equation[26],

$$k = A\, e^{-E_a/RT} \tag{8}$$

where k is the rate constant, A is the pre-exponential factor, E_a is the activation energy, and R is the gas constant. Differentiating Equation 8 with respect to temperature,

$$\frac{d(\ell nk)}{dT} = \frac{E_a}{RT^2} \tag{9}$$

and integrating between limits followed by rearrangement gives,

$$E_a = 2.303R \left(\frac{T_1 T_2}{T_2 - T_1}\right) \log \left(\frac{k_2}{k_1}\right) \tag{10}$$

If it is now assumed that the relative rates at 40°C and 25°C in Figure 4 are proportional to the respective rate constants at those temperatures, the activation energy is 4.5 kcal mol^{-1}. This value is consistent with that expected for a physical process exhibiting diffusive resistance such as transfer of carbon dioxide molecules across the potential barrier of a bubble wall,[27] but the value is much lower than expected for biochemical reactions occurring either within or without the bacteria ($E_a \geq 10$ kcal $-mol^{-1}$).

Pressure Evidence In a phase transfer, rate-limited system, the carbon dioxide partial pressure would play a critical role because its rate of transfer should be proportional to the concentration difference across the bubble wall. On the grounds that the authors carefully controlled temperature at 20°C, pH

at an acceptable value of 6.7, and substrate concentrations, the best study on the effects of pressure upon the rate of digestion is an early batch-type experiment by Whipple, Fair, and Klein at Harvard.[28] As shown in Table 2 and Figures 5 and 6, the authors measured gas yields as a function of pressure at 460-, 760-, and 1385-mm of mercury absolute. The values in parentheses in Table 1 are normalized yields computed by dividing the value of respective gas yields at seven weeks into the values at other times. This internal normalization helps to eliminate any spurious differences between experiments due to changing solubility of gas or inconsistencies of substrate.

A comparison of the plots in Figures 5 and 6, however, indicates that such differences were probably not important. Both sets of plots indicate a pressure effect in the direction indicated for a gas transfer rate limitation. An examination of the initial rates by a comparison of the slopes of the first three data points at each pressure indicates that methane production was about 1.5 times faster at 460 mm than it was at 1385 mm. Since the pressure differed by a factor of three there is obviously less than a one-to-one dependence of rate upon pressure; however, this is not surprising since from Equation 1, Henry's constant also contributes to the slopes.

Agitation Evidence Anaerobic digesters are heterogeneous systems incorporating an aqueous phase that contains dissolved inorganic and organic materials, a solid phase consisting of both reactive and unreactive celluloses, a solid phase consisting of bacteria,

Table 2. Methane and carbon dioxide production as a function of pressure in unmixed batch experiments with fresh organic solids seeded with anaerobic bacteria as substrate. The temperature was 20°C and the pH was controlled at 6.7. The columns with parentheses represent the normalized gas yields from the columns directly to the left (ref 28).

Elapsed Time	Gas Yield (ℓ-kg^{-1} of organic material)					
	460 mm		760 mm		1385 mm	
(weeks)	CH_4	CO_2	CH_4	CO_2	CH_4	CO_2
1	47 (0.11)	56 (0.27)	45 (0.10)	23 (0.15)	40 (0.09)	26 (0.18)
2	114 (0.27)	104 (0.51)	115 (0.25)	52 (0.35)	92 (0.21)	55 (0.38)
3	194 (0.46)	136 (0.67)	185 (0.40)	78 (0.52)	140 (0.32)	75 (0.52)
4	275 (0.65)	159 (0.78)	258 (0.56)	99 (0.66)	207 (0.48)	94 (0.65)
5	350 (0.82)	182 (0.89)	342 (0.74)	119 (0.79)	279 (0.65)	112 (0.78)
6	397 (0.93)	195 (0.96)	423 (0.91)	138 (0.92)	369 (0.85)	132 (0.92)
7	426 (1.00)	204 (1.00)	464 (1.00)	150 (1.00)	432 (1.00)	144 (1.00)

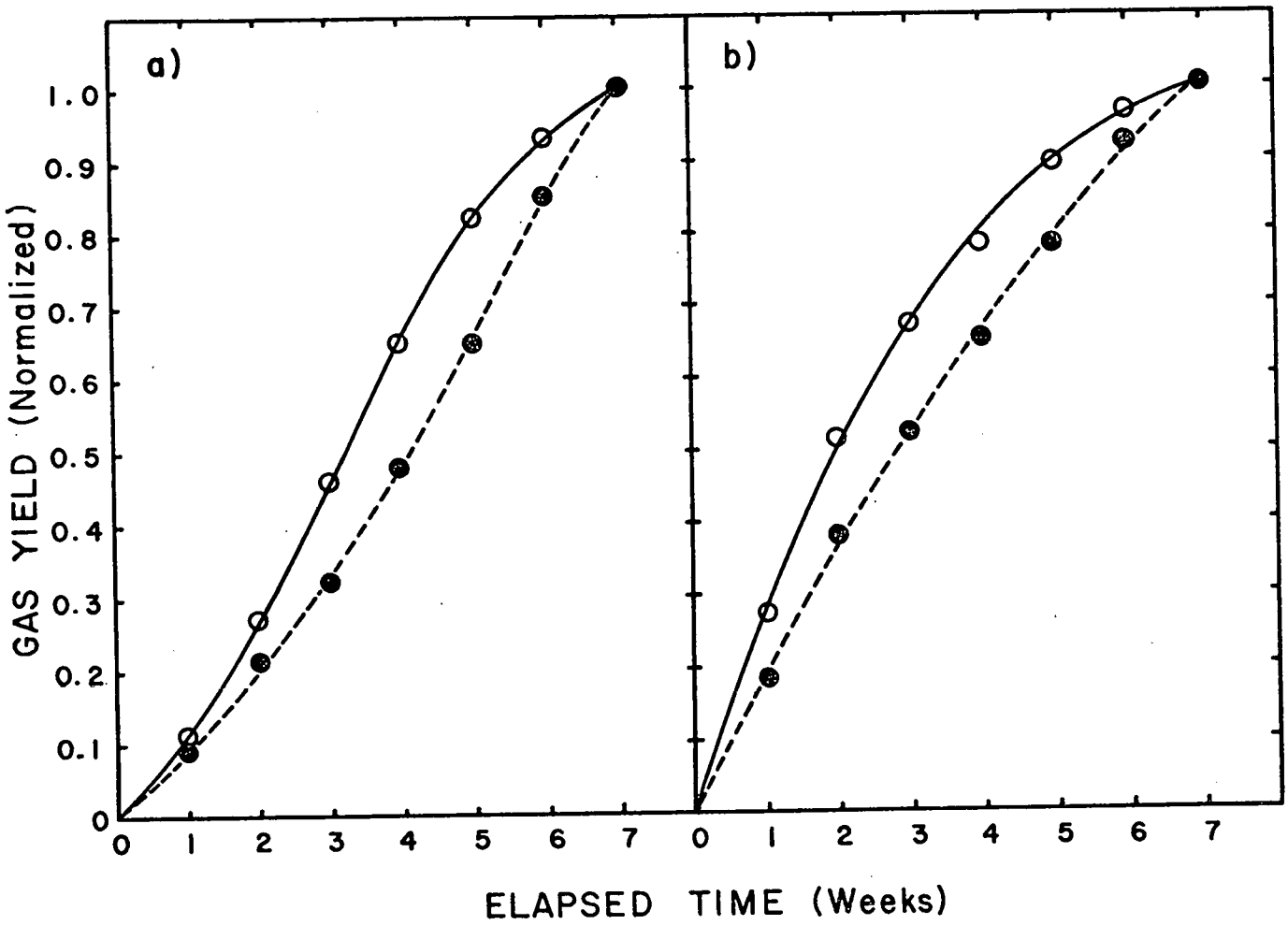

Figure 6. Normalized gas yields as a function of pressure: a) methane; b) carbon dioxide (ref. 28). Open circles are P = 460 mm and closed circles are P = 1385 mm.

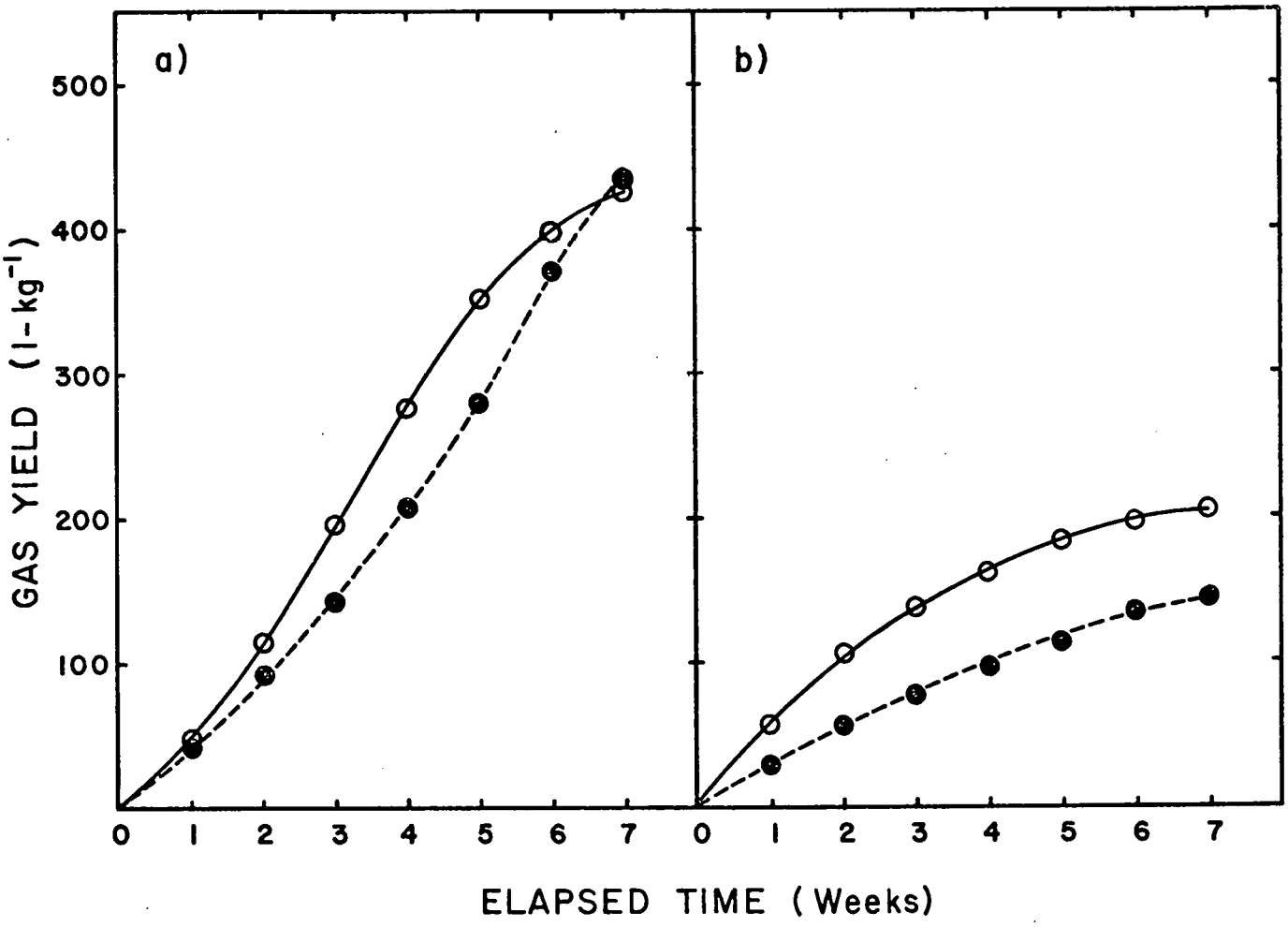

Figure 5. Experimental gas yields as a function of pressure: a) methane; b) carbon dioxide (ref. 28). Open circles are P = 460 mm and closed circles are P = 1385 mm.

and a gas phase in the form of bubbles. Starting in about 1955, when it was shown by Sawyer and Roy[29] along with Torpey[9] and others that loading in agitated, mesophilic digesters could be increased to 3-8 g ℓ^{-1} day^{-1} of volatile sewage solids from only 0.6-1.1 g ℓ^{-1} day^{-1} for quiescent mesophilic systems[30] agitation was widely implemented. The usual reasons given for employing agitation are to provide maximum contact between phases and a uniform thermal environment, and to prevent concentration stratifications of nutrients, products, and inhibitors. Although recommended agitation levels are about 0.2 hp per thousand cubic feet, there does not appear to be a very extensive study determining specific rate and/or loading as a function of agitation level.

However, the increases in loading that have been observed in the past are consistent with the phase transfer rate-limiting model since agitation is thought to reduce diffusive resistance by decreasing the thickness of the bubble walls.[21,31] The effect of mixing toward reducing diffusive resistance in anaerobic digestion has recently been stressed by Gaddy et al.,[3] and although their interpretation of the rate-limiting step is different from ours, it is seen that their results are entirely consistent with our model.

Starting with Fick's second law and the assumption that the rate limitation is diffusion of substrate into a microbial floc, they have developed a first order rate expression,

$$r_g = k_g C_g \qquad (11)$$

where: r_g is the rate of substrate consumption, C_g is the concentration of substrate in the reactor, and k_g is a combined rate constant including reactive and diffusive influences. Using typical agricultural residues as substrates they found that the digestion was indeed first order, and specific rates were 0.086 day^{-1} and 0.054 day^{-1} for continuously-stirred and daily-stirred systems, respectively. They concluded that their results were consistent with a rate limitation by diffusion of substrate to microbial flocs; and "high diffusive resistance is further indicated by the increase in rate constant with agitation."

We should note that the temperature and activation energy evidence from a previous section is also consistent with their interpretation. Summarizing; temperature, activation energy, and mixing evidence would all tend to favor Gaddy's substrate diffusion proposal. However, the observed dependence of rate upon pressure from the previous section is not explained by this proposal. Since substrates are all non-gaseous materials such as solid cellulose, glucose, or volatile acids, a pressure dependence would not be expected if any of these substrates (or some combination) were rate-limiting. We conclude, therefore, that the available temperature, activation energy, pressure, and agitation evidence all support a rate limitation imposed by phase transfer of products.

Pressure-Agitation Evidence In the following we compare the rate performance of a phase transfer-assisted digester operating at mesophilic temperature, reduced pressure, and vigorous agitation

against one operating at mesophilic temperature, atmospheric pressure, and very mild agitation. The former is our work[20] which was reported in 1975, while the latter is that of Lawrence and McCarty[15,16] reported in 1967. The only significant differences in the two experiments is the lower absolute pressure (180 mm vs. 740 mm) and the more vigorous agitation (3.1 liters of fluid recycled at 11.4 liter/min by an external centrifugal pump vs agitation by gas recirculation) that we employed.

Acetic acid was the substrate used in both studies. Conversion of substrate into cells and bacterial decay may be described by

$$\frac{d(B_m)}{dt} = Y \frac{d(HAc)}{dt} - k_d(B_m) \qquad (12)$$

where Y is the bacterial yield or fraction of substrate converted to cells and k_d is the first-order bacterial decay constant. For a steady state digester the biological solids retention time (SRT) is the reciprocal of the specific bacterial growth rate $[d(B_m)/dt]/(B_m)$. Accordingly, with Eqn 12 and Eqn 2, the Monod expression, the dynamic constants (Y, k_2, K_s and k_d) may be evaluated when effluent bacterial masses and acetic acid concentrations are measured as a function of SRT. Finally, a comparison of process chemical efficiencies, E_c, as a function of residence time is a more practical, direct method for determining rate performance. E_c is defined by

$$E_c = \frac{(HAc)_i - (HAc)_e}{(HAc)_i} \times 100 \qquad (13)$$

where i and e denote influent and effluent acid concentrations.

The data and conditions for our constant loading study are found in Table 3. Plots of our chemical efficiencies and those of Lawrence and McCarty are found in Figure 7. It is seen in the figure that the lower pressure and more vigorous agitation we employed resulted in much higher conversion efficiencies at the lower residence times, and this fact is indicative of higher specific rates under the conditions of our experiment.

Least-squares fitting of the data in Table 2 to Equations 2 and 12 gave Y=0.044, K_s=250 mg/liter, k_2=56 day^{-1} and k_d=0.44 day^{-1}. In contrast, for their digester operating at a constant influent feed concentration of 3135 mg/liter, Lawrence and McCarty determined Y=0.040, K_s=166 mg/liter, k_2=9.6 day^{-1} and k_d=0.019 day^{-1}.

The close agreement in yield constants suggests that an irregularity in the experimental procedure, chemical analysis, or data treatment does not account for the significantly different values determined by k_2 and k_d. The yield constant should be invariant because it is an internal cellular metabolic constant, the manifestation of which is the mass of cells produced per unit mass of acetic acid consumed.

The observation of an increased k_2, the turnover constant, with decreased pressure and more vigorous agitation is of importance. It is difficult to imagine how this specific rate could increase by about 600 percent if the kinetic treatment actually applies to internal cellular enzymatic processes since

Table 3. Constant loading SRT study with acetic acid as substrate. The conditions were: liquid temperature, 35±1°C; gas temperature, 21° to 27°C; liquid volume, 3.10 liters; gas volume, 25.5 liters; initial pressure, 180 torr; and pH, 6.4 to 7.4.

SRT (days)	Feed frequency (per day)	Feed concentration (mg/l)	Effluent acetic acid (mg/l)	Effluent organic nitrogen x 11.4 (mg/l)	Pressure increment (Torr. per day)	Chemical efficiency (%)
2.00	2	8580	271	211	217	96.8
1.50	3	6430	146	153	207	97.7
1.00	4	4290	451	77.6	192	89.5
0.788	4	3380	584	58.5	181	82.7
0.394	8	1690	356	30.4	142	78.9

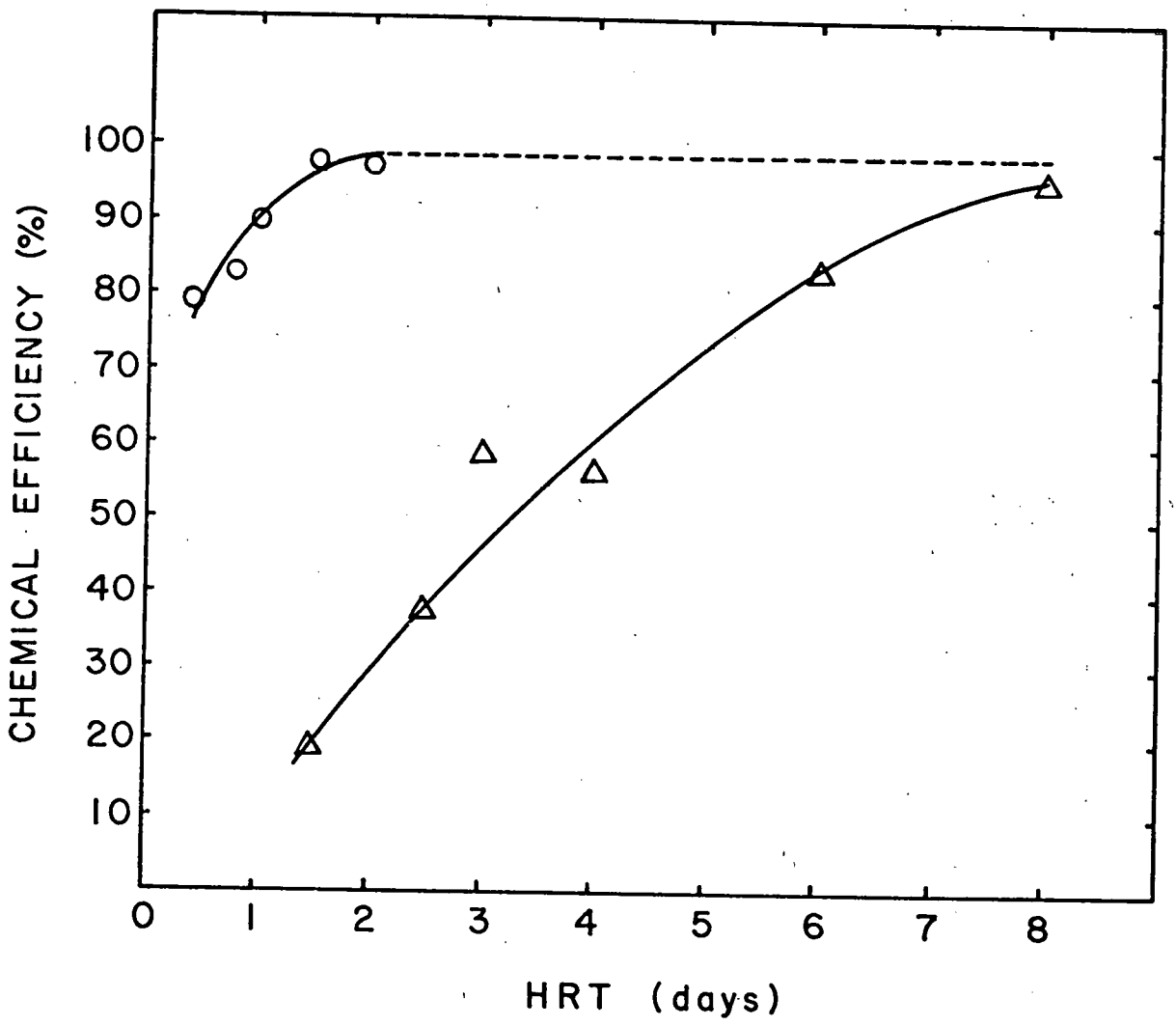

Figure 7. Chemical efficiencies with acetic acid as substrate. Open circles represent our data at 180 mm with liquid recycle at 4290 mg ℓ^{-1} day^{-1}. The triangles represent the data of Lawrence and McCarty (Refs. 15 and 16) at 760 mm with gaseous recycle at 3135 mg ℓ^{-1}.

neither pressure nor agitation should directly effect the rate of these internal processes. Instead, we believe that since the same mechanism and rate expression (Eqs. 1 and 2) are applicable for transport of substrate across the bacterial cell wall, entrained gases in the form of bubbles were acting to decrease the bacterial surface area (and thereby k_2) in the Lawrence and McCarty work. Viewed from the opposite perspective, our greater k_2 is the result of more rapid transfer of products to gas bubbles promoted by low pressure and vigorous agitation thereby eliminating inhibition by entrainment because the bubbles can grow to their critical size and escape from solution more rapidly.

Viscosity-Volatile Acids Evidence. The viscosity of a fluid is a measure of its resistance to flow. From Equation 4, the rate of phase transfer is directly proportional to the diffusion coefficient which, in turn, for ideal solutions is inversely proportional to viscosity,[32]

$$D = \frac{RT}{6\pi rN\eta} \tag{14}$$

where R is the gas law constant, T is the absolute temperature, r is the radius of the diffusing molecule, N is Avogadro's number and η is the coefficient of viscosity of the liquid. It would be expected, therefore, that the greatest volumetric rates would be achieved in low viscosity anaerobic systems.

However, for any digestion system the viscosity increases as a function of loading because of the concomitant increasing

concentrations of reactive celluloses unreactive celluloses, and bacteria. Ideally, the increase in viscosity would be represented by the Einstein Equation,[33]

$$\eta = \eta_L(1 + 2.5\ C_x) \tag{15}$$

where η_L is viscosity of the pure liquid and C_x is the volume-fraction of solids. However, Pfeffer has demonstrated[33] as shown in Figure 8, that for digestion systems the viscosity increases exponentially with suspended solids concentration. Presumably the Bingham plastic behavior and the more rapid increase in viscosity than expected from the Einstein equation are due both to the fibrous nature of the substrate (urban refuse) and flocculation of the organisms. It is seen from Figure 8 that the data would fit an empirical expression of the form

$$\eta = \eta_L + b(S) + c(S)^2 \tag{16}$$

where S is the suspended solids concentration in grams per liter. From Figure 8, η_L is seen to be 0.9 cp so that a least-squares fitting of the data in Figure 8 to Equation 16 gives

$$\eta = 0.90 + 0.070(S) + 0.050(S)^2 \tag{17}$$

Substitution of Equation 17 into 14 followed by substitution into 4 gives

$$R_t = \frac{RT}{6\pi rN\ell} \frac{(C_s - K_H(T)P)}{(0.90 + 0.070(S) + 0.050(S)^2)} \tag{18}$$

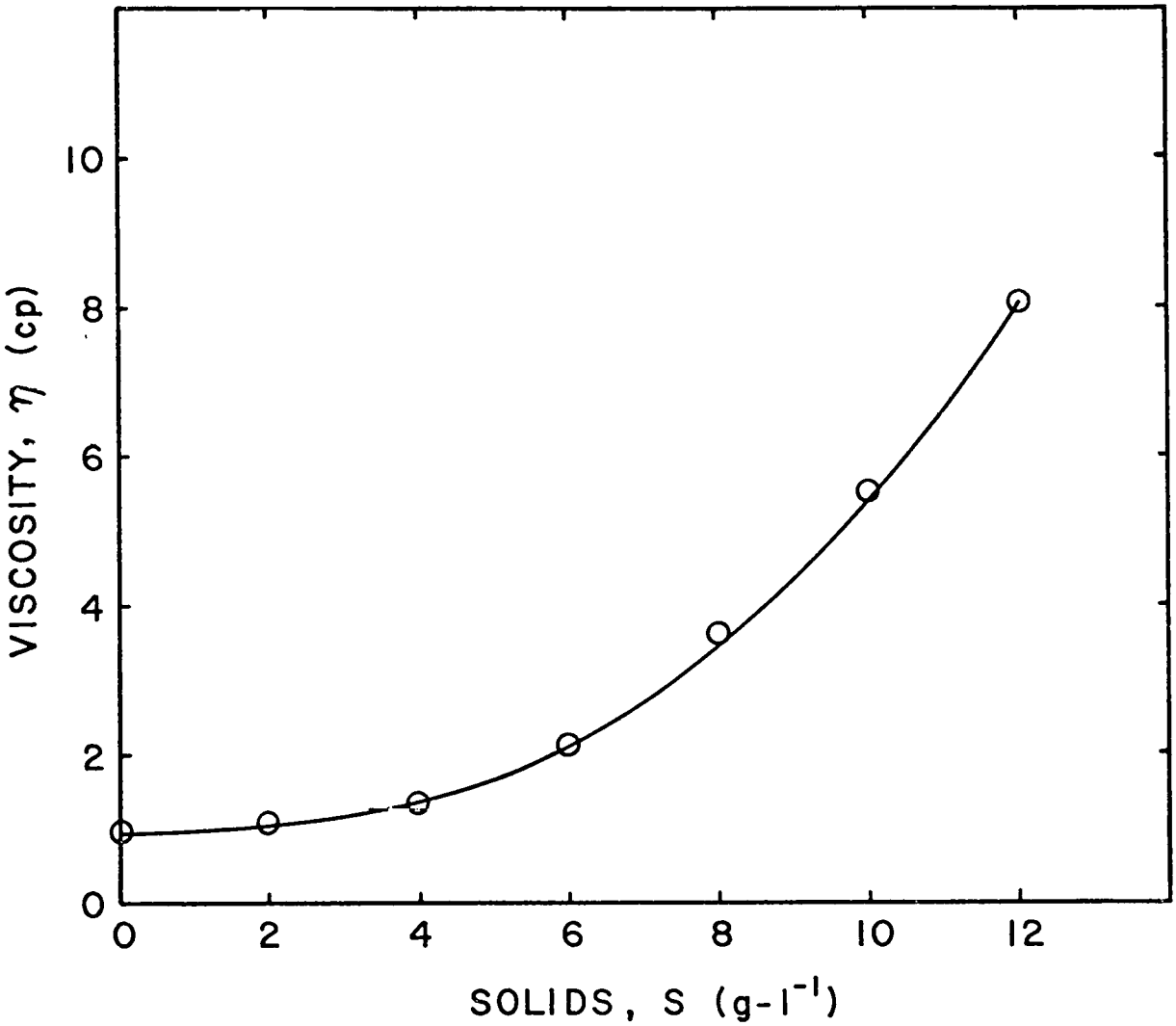

Figure 8. The exponential increase in viscosity with slurry solids concentration (Ref. 33).

Importantly it is seen that the viscosity term in Equation 18 is precisely the same form as the denominator of Andrew's expression (Equation 3) for the inhibited rate loss of acetic acid. This similarity provides at least a potential link between inhibited acid consumption and a slowdown in the rate of bubble growth.

There is also a reasonably direct correlation in the literature between increasing viscosity and decreasing specific rate. In his study of the thermophilic decomposition of urban waste by single-step anaerobic digestion, Pfeffer[11] determined two first-order rate coefficients for each temperature; one for short residence times and another for longer residence times as shown in Table 4. Pfeffer has made the following comment regarding the duality of rate constants:

> "As the temperature increases, the rate constant increases as would be expected. However, there appears to be two rate constants, one at short retention times and another one for the longer retention times. These two constants are not pronounced at 35°C, but increasing the temperatures show a definite break in the curve. The significance of this break is not known at this time except that the low rates at the longer retention times may be nothing more than endogenous respiration."

Table 4. Pfeffer's thermophilic, first-order rate coefficients for the anaerobic digestion of urban refuse (ref. 11).

Temperature (°C)	Initial (day^{-1})	Final (day^{-1})	Transition residence time (days)
50	0.117	0.030	10
55	0.623	0.042	6
60	0.990	0.040	6

At first glance, Pfeffer's speculation that the lower rates at longer residence times were due to respiration seems plausible. Since the rate expression usually employed for bacterial growth and death is,

$$\frac{d(B)}{dt} = Y \frac{d(S_g)}{dt} - k_d(B) \tag{12}$$

where (S_g) is now a generalized cellulose substrate. Assuming, as Pfeffer did, that substrate utilization is first order, we have,

$$- \frac{d(S_g)}{dt} = k(S_g) \tag{19}$$

Because the loss of bacteria from the effluent of a steady state digester is just Q(B), where Q is the effluent flow rate and V is the volume, we have as a steady state expression,

$$\frac{d(B)}{dt} V = VYk(S_g) - Vk_d(B) - Q(B) \tag{20}$$

Letting $\frac{d(B)}{dt} = 0$, and since the residence time, θ, is equal to V/Q, we have as an expression for the bacterial concentration,

$$(B) = Yk(S_g) \left(\frac{\theta}{k_d\theta + 1}\right) \tag{21}$$

Assuming, for the moment, that the substrate concentration is held constant as the residence time is varied, plots of θ versus $\theta/(k_d\theta + 1)$ as shown in Figure 9 give the relative increase in bacterial concentrations with residence time.

It is seen in Figure 9, that even when the specific death rate is quite high, the very sharp falloff in specific rate

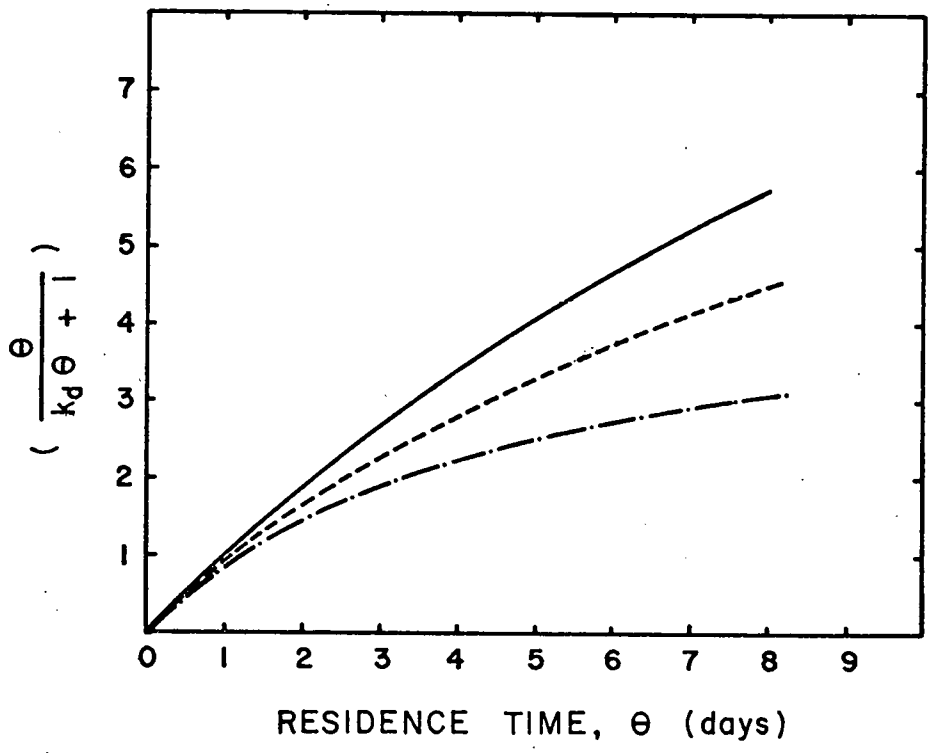

Figure 9. Relative bacterial concentrations as functions of residence time and death constants: k_d = 0.05 day^{-1} ————————; k_d = 0.10 day^{-1} – – – – – –; and k_d = 0.20 day^{-1} —— – ——.

observed by Pfeffer cannot be attributed to a rapid increase in respiration rate. Furthermore, neither can the decline in rate be attributed to a decreasing substrate concentration. Table VI of Reference 11 shows that, for the thermophilic digestions, the concentrations of volatile acids actually increased as a function of residence time.

An alternative explanation of these phenomena is that the viscosity increased at the longer residence times thereby imposing a phase transfer rate limitation. We note, at the outset, that the final rate constants in Table 4, for residence times greater than 6 to 10 days, exhibit a very weak temperature dependence as would be expected for a phase transfer, rate-limiting process.

Thus, our purpose here is to show the reasonableness of the explanation that at short residence times Pfeffer's digesters were rate-limited by the hydrolysis reaction, while at longer retention times they were rate-limited by phase transfer of gaseous products. Further, the transition between the two rate-limitations is the result of an increasing digester viscosity at the longer residence times. An understanding of this phenomenon is instrumental in approaching the later question of how best to optimize two stage anaerobic digesters.

It has been shown[34] that urban refuse consists of fixed solids (25 Percent); digestible cellulose, oils, and waxes (65 percent); and lignin (10 percent). Assuming the stoichiometry,

$$(C_6H_{10}O_5)_n + nH_2O \rightarrow 3n\ CH_4 + 3n\ CO_2 \qquad (22)$$

for the digestible cellulose fraction, then at 100 percent conversion efficiency of this fraction, each pound of total solids would produce about 8.6 SCF of gas (13.3 SCF lb^{-1} for the cellulose fraction or 11.5 SCF lb^{-1} of volatile solids). For solids that were not pre-treated, Pfeffer observed a gas production rate of about 5.0 SCF lb^{-1} of volatile solids or 3.7 SCF lb^{-1} of total solids indicating that the conversion efficiency from total solids was only about 28 percent.

To accomplish our purpose here, where we wish to calculate viscosity as a function of residence time, we shall make the simplifying assumption that only two kinds of solids enter the digester; these are reactive and unreactive and the reactive portion is 28 percent of total solids.

A mass balance for the reactive cellulose, (S_r), is given by

$$-\frac{d(S_r)}{dt} V = Q(S_r)_i - Vk_h(S_r) - Q(S_r) \tag{23}$$

where $(S_r)_i$ is the concentration of reactive celluloses in the influent and k_h is the hydrolysis rate constant.

For a digester in a steady state, $d(S_r)/dt = 0$, and since $V/Q = \theta$, solution for (S_r) gives,

$$(S_r) = \frac{(S_r)_i}{(\theta k_h + 1)} \tag{24}$$

so that the reactive cellulose concentration in the digester may now be computed as a function of θ.

The concentration of completely unreactive solids is just the loading rate in g ℓ^{-1} day^{-1} of this species times the residence time, θ. Values for influent and effluent solids concentrations,

digester viscosity, and hydrolysis efficiences can be found in Table 5. A hydrolysis constant of 0.5 day^{-1}, about equal to Pfeffer's initial rate constant at 55°C, has been assumed. The hydrolysis efficiency in percent is defined by

$$E_h = \frac{(S_r)_i - (S_r)}{(S_r)_i} \times 100 \qquad (25)$$

and it is assumed that once the reactive celluloses are hydrolyzed, they no longer significantly contribute to viscosity. A constant loading rate of 3.33 g ℓ^{-1} day^{-1} of total solids has been used. This value is equal to that employed by Pfeffer.[11]

Table 5. Influent and effluent total solids concentrations, effluent viscosity, and hydrolysis efficiency as functions of residence time with $k_h = 0.5$ day^{-1} for reactive celluloses with a constant loading of 3.33 g ℓ^{-1} day^{-1} of total solids and 28 percent reactive solids and 72 percent unreactive solids.

Residence Time ($days^{-1}$)	Influent Solids (g ℓ^{-1})			Effluent Solids (g ℓ^{-1})			Visc. (cp)	Eff. (%)
	Total,	Reactive,	Unreactive	Unreactive,	Reactive,	Total		
1	3.33	0.93	2.40	2.40	0.62	3.02	1.57	33
3	9.99	2.80	7.19	7.19	1.12	8.31	4.93	60
5	16.6	4.66	12.0	12.0	1.35	13.3	10.7	71
7	23.3	6.53	16.8	16.8	1.44	18.2	18.8	78
9	30.0	8.39	21.6	21.6	1.51	23.1	29.2	82
11	36.6	10.3	26.4	26.4	1.54	27.7	41.8	85

Results from the table are also plotted in Figure 10, where it is seen that the viscosity increases very rapidly in the 6-10 day region. As seen from the hydrolysis efficiency curve about 75 percent of the reactive cellulose has hydrolyzed at a 6-day residence time so that the high viscosity results almost entirely

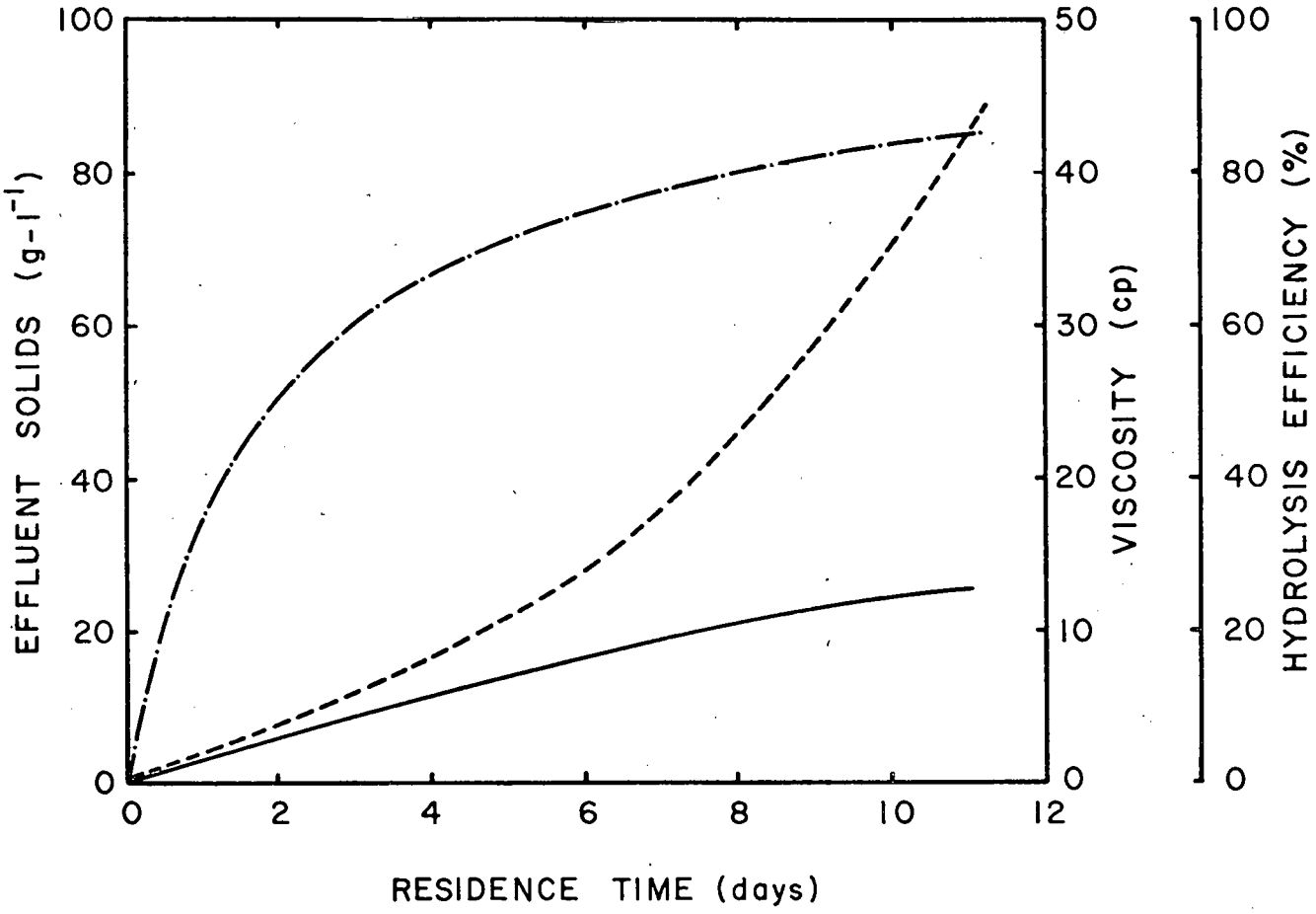

Figure 10. Effluent solids ————————, viscosity – – – – – –, hydrolysis efficiency ——— – ——— as a function of residence time with 28 percent reactive celluloses at a constant loading of 3.33 g ℓ^{-1} day^{-1} with k_H = 0.5 day^{-1}.

from the 72 percent contribution of unreactive solids. Thus it is our belief that the slowdown in specific rate that Pfeffer observed was the result of a phase transfer limitation imposed by increasing viscosity. This view is further corroborated by the increasing volatile acids concentrations and by the weak temperature dependences of the rate coefficients at longer residence times.

In summary, the evidence supporting the proposition that phase transfer of products is rate-limiting includes: (i) a weak dependence of rate upon temperature leading to a low activation energy which is consistent with that expected for diffusion-controlled process across the bubble wall; (ii) an experimental dependence of rate upon pressure also consistent with the hypothesis indicating that a greater concentration gradient across the bubble wall is favorable to rate performance; (iii) improved rates in agitated systems signifying that decreased wall thickness is favorable for enhancing rate; (iv) the observation of significantly improved reproduction rates in a low pressure, vigorously agitated digester; and (v) the observation of a transition to lower digestion rates at longer residence times which is consistent with the hypothesis since viscosity increases exponentially with residence time.

EXPERIMENTAL

A practical implication of the preceeding line of thought is that faster gas production might be observed in a phase transfer-assisted digester. A one-liter pyrex digester in a standard mixing configuration was placed in a water bath at 60±1°C. Mixing was accomplished with an indirect drive, magnetically coupled impeller operating continuously at 100 and pulsed to 400 rpm for a fifteen second period every five minutes. The purpose of the pulse was to insure the uniform distribution of heavy particulate matter and to minimize the accumulation of bacteria on digester parts. Effluent gases flowed to a 303 liter reservoir which was maintained at about 250 torr by periodic evacuation. Feeding and effluent withdrawal were accomplished with tubing pumps governed by timers.

Components of the feed solution are given in the caption of Figure 11. Enough lime and sodium hydroxide was added to maintain the digester pH between 6.8 and 7.2, and at the 31 g/ℓ-day feed rate these values were, respectively, 1.50 and 1.22 grams per liter of feed. Also, 25 percent of the effluent cells were lysed in boiling water for 30 minutes, and the water soluble protein extract was recycled into the feed solution. It has been shown that this soluble component has a stimulatory effect upon the methanogenic bacteria at high loading rates.[35,36] To minimize the bacterial concentration, and thereby viscosity, a residence time of one day was selected.

Standard analytical procedures were used for determining

the carbon and nitrogen balances. Analyses for carbon in the forms of glucose, bacterial cells, and volatile acids were performed using the COD and spectrometric volatile acids tests. The Kjeldahl analysis and an ion specific electrode were used to measure organic and ammonium nitrogen, respectively. The rate of gas production was determined by monitoring the pressure increase in the 303 liter reservoir. Mass spectrometric analysis of the gas phase gave 46 and 54 percent, respectively, for methane and carbon dioxide.

The system was initially seeded with thermophilic bacteria ($60^{o}C$) obtained from an experimental system operating at the University of Illinois.[34] Glucose was substituted for urban refuse as substrate upon seeding. After an initial equilibration at a residence period of 8 days, the residence time and pressure were gradually reduced to the values given in Figure 11. Microscopic observations performed after the equilibration period revealed that the predominant faunae were single, curved rod-shaped bacteria and chains of straight rod-shaped bacteria. The experiment took about nine (9) months to perform.

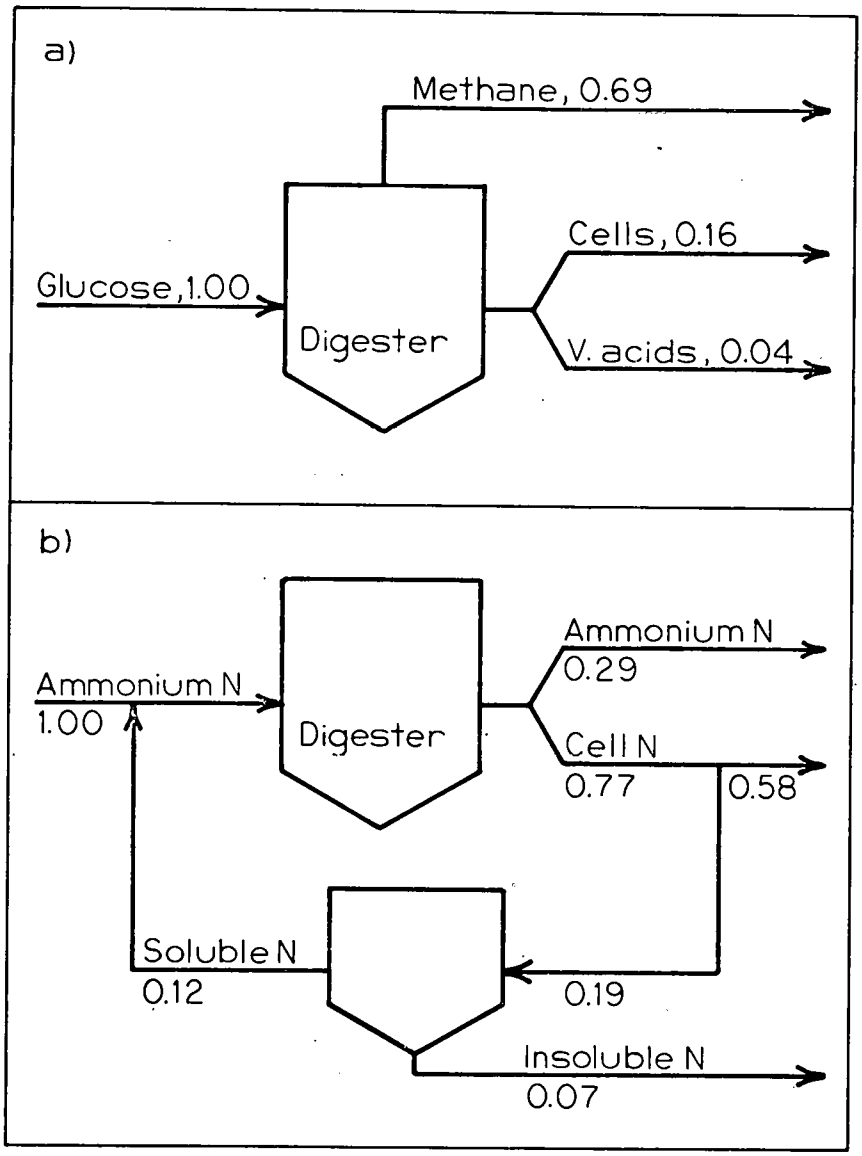

Figure 11. Experimental carbon and nitrogen balances: a) basis; 1.00 g ℓ^{-1} day^{-1} COD. b) basis; 1.00 g ℓ^{-1} day^{-1} ammonium nitrogen. Components of the feed solution were D-$C_6H_{12}O_6$, NH_4Cl, $(NH_4)_2HPO_4$, $FeCl_3$, K_2SO_4, $CoCl_2 \cdot 6H_2O$ and thiamine hydrochloride present in the weight ratios, 100 : 6.0 : 1.6 : 0.75 : 0.39 : 0.15 : 0.15, respectively. The rate of loading was increased in one gram increments with a minimum of a three-day interval between increases. Experimental conditions were: hydraulic residence time, 1.0 day; pressure, 250 mm of mercury absolute; agitation, 100 rpm pulsed to 400 rpm for 15 seconds every five minutes; temperature, 60°C.

RESULTS AND DISCUSSION

The detailed carbon and nitrogen balances in Figure 11 give a reasonable overview of the experiment. The data for the balances were accumulated in the 20-22 g ℓ^{-1} loading range. The absolute value of organic nitrogen (cell N) in Figure 11b was multiplied[37] by 11.4 to give the absolute value of cell COD for use in Figure 11a. It is seen from Figure 11 that 89 and 95 percent, respectively of the glucose COD and nitrogen mass are accounted for. It is felt the lower figure for recovery of COD is probably the result of an experimental artifact--perhaps related to the greater pumping speed for methane compared to that for carbon dioxide. The true value of the methane COD in this loading range was probably closer to 0.80.

This latter conclusion is reinforced by defining the system chemical efficiency, E(COD) as

$$E(COD) = \frac{(COD)_{inf} - (COD)_{eff}}{(COD)_{inf}} \times 100 \qquad (26)$$

where the subscripts inf and eff denote liquid phase influent and effluent COD, respectively. It is seen in Figure 12d that in the 15-24 g/ℓ-day range, the value for E(COD) is consistently 79-80 percent.

Some insight into the nature of the loading limit is also provided in Figure 12 where effluent organic nitrogen, volatile acid and COD concentrations are plotted against glucose loading. It is worthwhile focusing on the fact that the volatile acids concentration increases with loading in a fashion similar to the

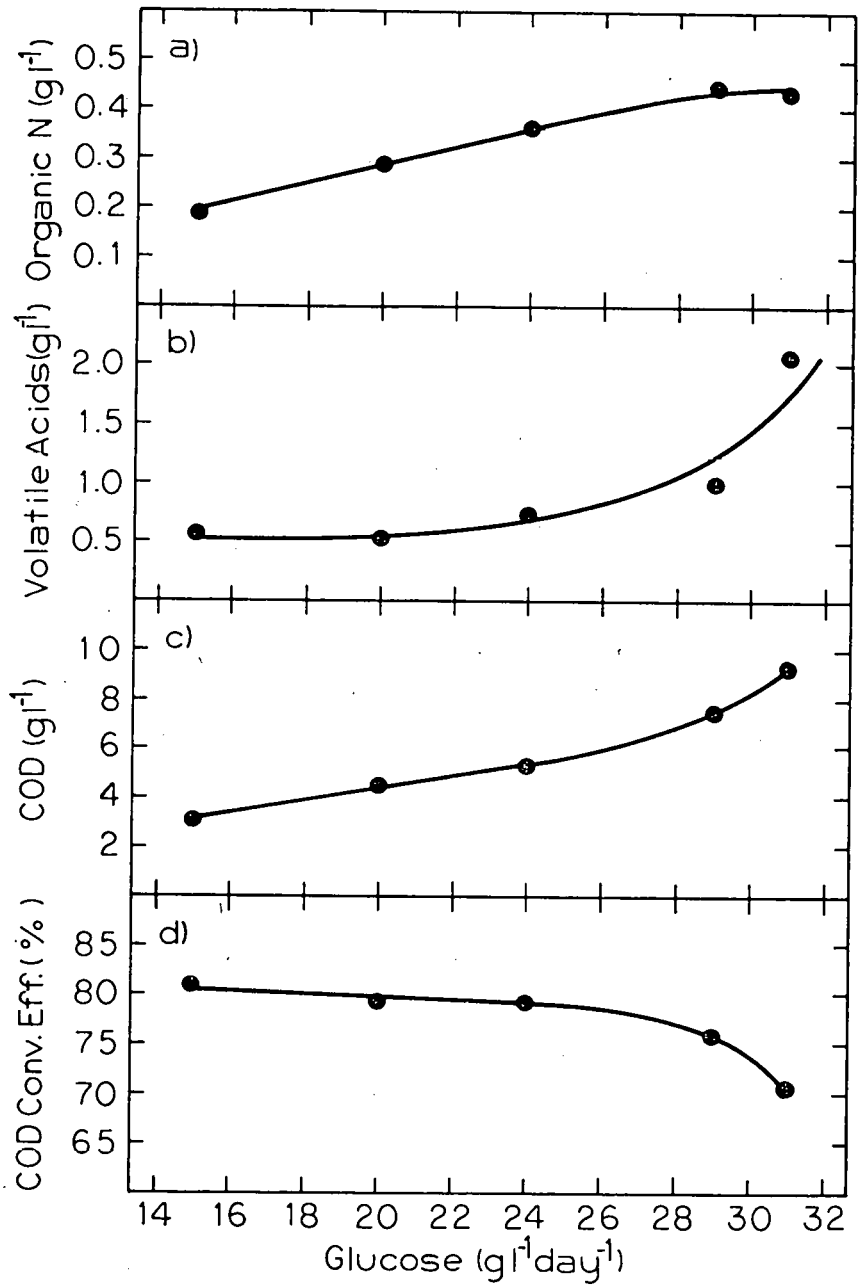

Figure 12. The effect of glucose loading on a) organic nitrogen, b) volatile acids, c) COD, and d) COD conversion efficiency.

increase in viscosity with slurry solids.

The only solids in our system were the bacteria themselves as manifested by the organic nitrogen concentration (Figure 12a). By using a cell weight to organic nitrogen ratio[37] of 9.4 it is then possible to compare bacterial solids with volatile acids as seen in Table 6. The table shows that the acids concentration, A, increases exponentially with suspended solids concentration, and a suitable empirical equation is

$$A = A_o + b'(S) + c'(S)^2 \tag{27}$$

and a least-squares fitting of the data in Table 6 to Equation 27 gives where A and S are both in units of g ℓ^{-1},

$$A = A_o + 0.051(S) + 0.073(S)^2 \tag{28}$$

and since A=0 when S=0, then A_o=0 giving

$$A = 0.051(S) + 0.073(S)^2 \tag{29}$$

as plotted in Figure 13. We have also included in Figure 13, a plot of viscosity at our suspended solids concentration using Equation 17 but neglecting the zero point contribution for comparison purposes.

It is seen in Figure 13 that the divergence between the two curves is never great being a maximum of 25 percent at high solids concentration. Thus, in addition to having the same general exponential shape, we conclude that when experimental error is considered, the expressions, 17 and 29, seem to be essentially the same.

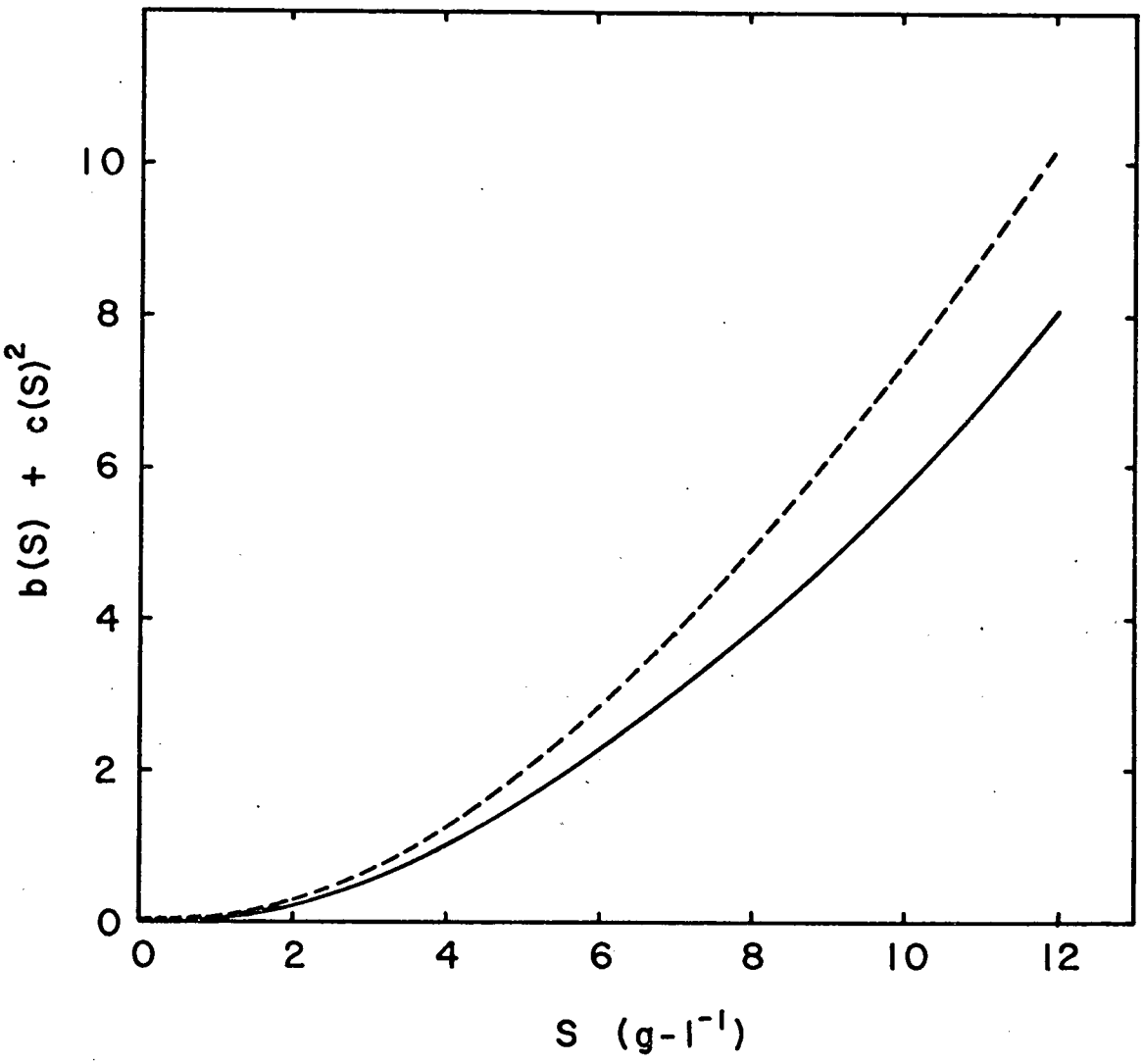

Figure 13. The least squares curves, neglecting intercepts, for Pfeffer's viscosity ———— and our volatile acids - - - - - - as a function of suspended solids.

Table 6. The conversion of organic nitrogen to bacterial solids, and corresponding values for volatile acids (data from Figure 12a).

Glucose loading ($g\ \ell^{-1}\ day^{-1}$)	Organic nitrogen ($g\ \ell^{-1}$)	Suspended Solids (Org. N x 9.4) ($g\ \ell^{-1}$)	Volatile Acids ($g\ \ell^{-1}$)
15.0	0.19	1.79	0.57
20.0	0.29	2.73	0.52
24.0	0.36	3.38	0.75
29.0	0.44	4.14	1.01
31.0	0.43	4.04	2.09

This is a rather important result. The similarity of Equations 17 and 29 indicates that the buildup of acids was probably due to the increased viscosity, which in turn, was caused by the greater bacterial solids concentration. Since in our system, the glucose-to-volatile acids conversion was unimpaired by the increased viscosity, it is probable that the gas transfer step was impaired by the increased viscosity and accordingly gas transfer was rate-limiting.

The exponential dependence of viscosity upon suspended solids concentration would lead one to suspect that the highest level of gas production might be achieved in low viscosity systems, and this suspicion is corroborated by facts in the literature. In Table 7 it is seen that, for both mesophilic and thermophilic digestions, higher rates have been achieved where low viscosity, soluble feed solutions have been employed. It is further noted

Table 7. Comparison of loading rates and conditions.

Investigator	Ref.	Subs.	Visc.	Temp.	Press. (atm)	Agitation	Res. Time (days)	Volatile Solids ($lb\text{-}ft^{3}\text{-}day^{-1}$) Added	Reacted
Torpey	9	Sewage Sludge	High	35°C	1	Liquid Recycle	3.2	0.87	0.36
Sawyer & Roy	29	Sewage Sludge	High	33-40°C	1	Gas Recycle	6	0.35	0.18
McCarty & Vath	35	Acetic Acid	Low	35°C	1	Gas Recycle	30	1.38*	1.37*
Pfeffer	33	Urban Refuse	High	60°C	1	Turbine	7.5	0.48	0.29
Buswell & Boruff	38	Butyl-acetate	Low	50-55°C	1	Slow Pumping & Recycle	2	0.94	0.54
This work	-	Glucose	Low	60°C	1/3	Turbine	1	1.93	1.37

*Unstable, one-day peak values

that the highest, stable loading and gas production rate was achieved in the system that was most optimized from the gas transfer perspective--low viscosity feed, short residence time, low pressure, vigorous agitation, and elevated temperature.

IMPLICATIONS FOR DESIGN

The preceeding discussion particularly as it relates to viscosity leads quite logically to an interesting technical paradox with implications for systems design: a rapid hydrolysis reaction requires a high concentration of cellulose and an accompanying high viscosity medium while rapid phase transfer requires a low viscosity medium. Therefore, a separation of reactions is indicated. It would make possible the elimination of fixed solids and unhydrolyzed cellulose after the hydrolysis stage so that the contribution to the viscosity by these materials is eliminated entirely in the second-stage, gas-producing reactor. Although the two-stage technology is more complicated, it is felt that the potential for increased productivity and control of individual reactions outweighs the disadvantage of added complexity.

Three separation schemes are possible and they differ primarily in the nature of the first stage. They include: (i) hydrolysis of cellulose to primarily glucose with Tv cellulase enzymes; (ii) the hydrolysis of cellulose to carbohydrates and their subsequent conversion to volatile acids in an anaerobic stage; and (iii) the inorganic acid-catalyzed hydrolysis of cellulose to glucose.

The first approach,[39-46] employing Tv cellulase enzymes is illustrated in Figure 14. It involves, in a first step, the production of enzymes from a radiation-induced mutant fungus of Trichoderma viride. The enzymes from this fungus have been

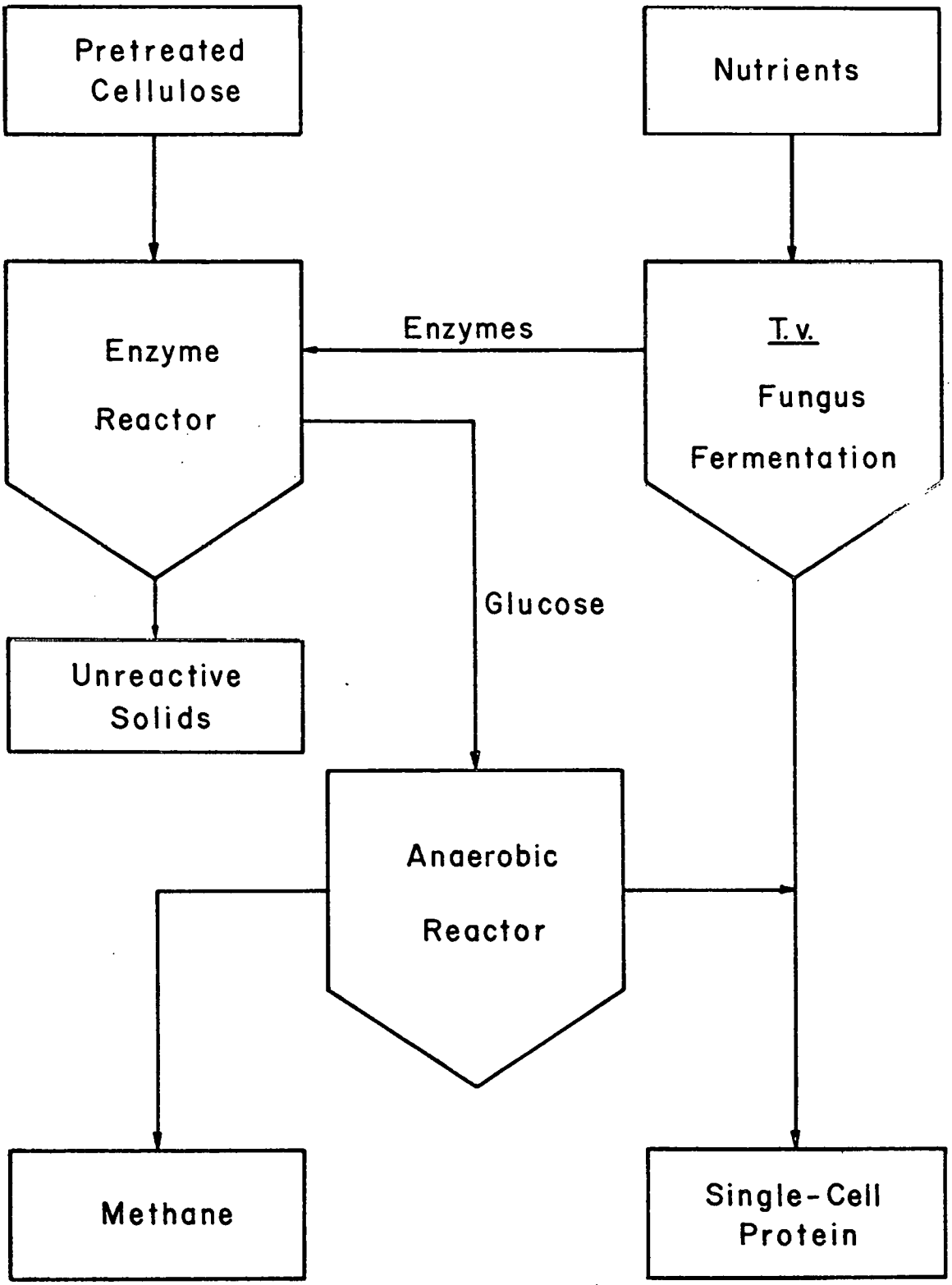

Figure 14. An advanced system for the fast production of methane using Tv cellulase enzymes.

shown to exhibit a very high degree of hydrolytic activity on more resistant substrates such as cotton. It has also been shown that cobalt irradiation of the fungus leads to mutants that are capable of producing two to four times as many cellulase enzymes as the wild strain. Enzymes produced in the first-stage are separated from the fungus and discharged into a stirred, heated reactor containing finely ground, cellulose waste materials in concentrations up to about 30 percent. The separation of enzyme from fungus is necessary to prevent the fungus from metabolizing the product glucose in the enzyme reactor.

The fungus is grown in a standard nutrient medium at a temperature of about 29°C in the mesophilic range. Other important conditions include pH control at around 3.0, an aeration rate of about 0.2 liters of air per liter of culture per minute, an agitation speed around 100 rpm for the turbine impeller, and a residence time of two or more days. Also, Mitre and Wilke[45] have recently shown that fungus growth and enzyme elaboration may be separated into stages with residence times of 4.75 hours and 50 hours respectively.

In the hydrolysis stage a conversion of waste cellulose such as ball-milled newspapers to glucose at up to 50 percent conversion efficiency can be accomplished in one or two days. Reaction conditions include 50°C temperature, continuous stirring and the control of pH at about 4.8. As expected for a surface reaction, the rates and conversions have been found to be sensitive to particle size and so ball-milling of the waste cellulose to 50-150 μ

has been found to be effective in facilitating more rapid and complete reactions.

The strengths of this technology include the relatively high degree of control that can be exercised over the enzyme, glucose, and gas production stages, and the fact that glucose has been obtained in concentrations in the range of 50-150 g ℓ^{-1} and these concentrations will be required for the influent to the gas-producing step. Existing, but potentially resolvable, weaknesses include: (i) the extensive and expensive size reduction of cellulose to 50-250 micron particle size; (ii) the contamination by aerobic organisms of the hydrolysis phase preventing efficient operation on a continuous basis; (iii) the expense of producing the enzymes and the fact that their immobilization is still not demonstrated on a practical basis; and (iv) more efficient operation of the hydrolysis at elevated thermophilic temperatures much beyond 50°C is not possible due to enzyme denaturization.

The potential for a two-stage technology based upon an anaerobic hydrolysis phase, as seen in Figure 15 has been the subject of a recent publication.[47] The technology consisted of a mesophilic, acid-producing reactor operating at a residence time of 0.47-1.20 days followed by a methane digester operating at a residence time of 6.46 days. The respective loadings for the acid-producing reactor and the methane digester were 1.54 - 2.67 and 0.18 lb v.s. cft^{-1} day^{01}.

Although the physiological basis of this technology is understood, the conditions employed are about opposite those

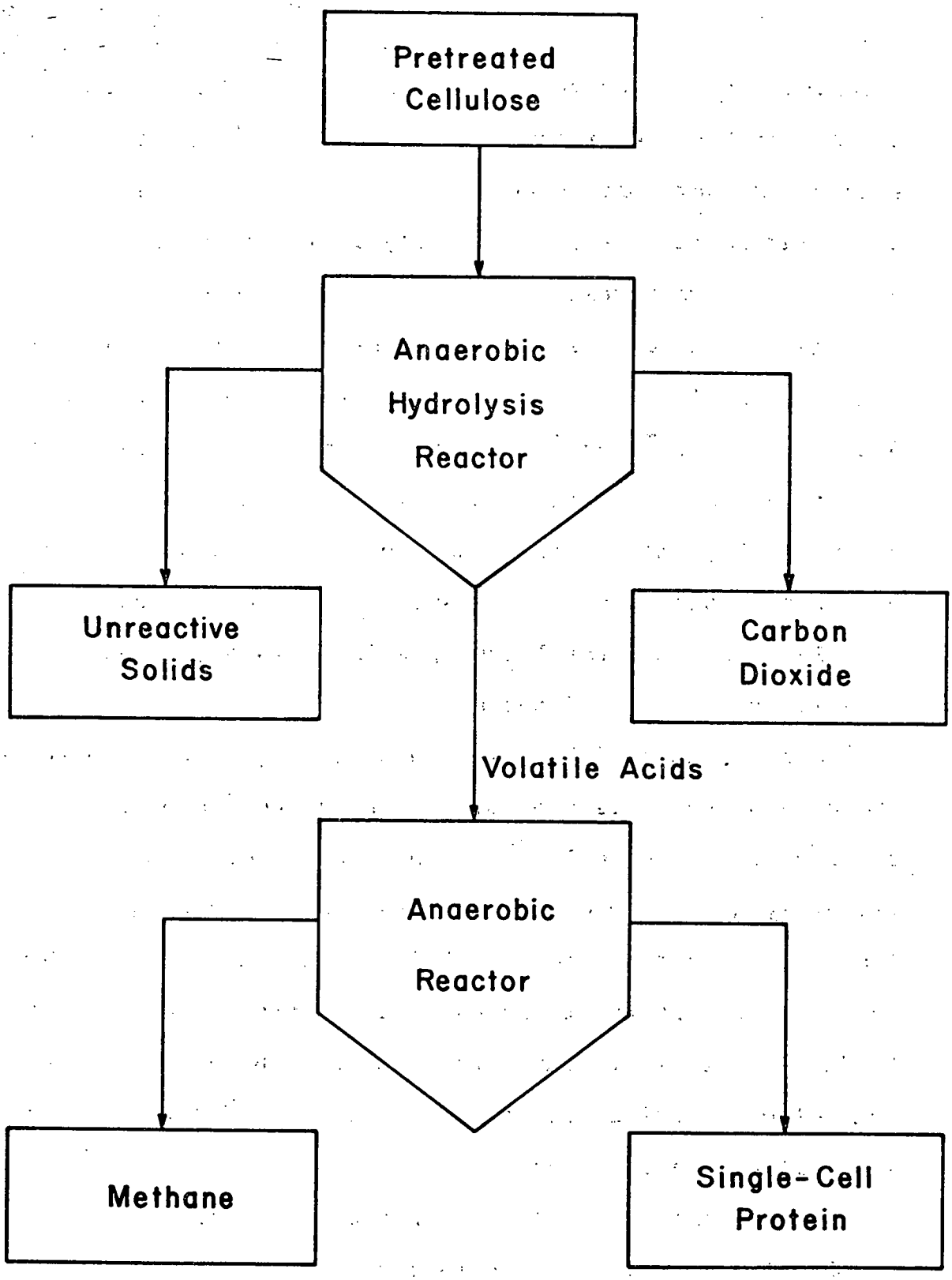

Figure 15. An advanced system for the fast production of methane using an anaerobic hydrolysis reactor.

required for optimum performance using our phase transfer approach. In the first place, recent work[5,11,13,33,34] has shown, that even under thermophilic conditions, the minimum residence time for a successful anaerobic hydrolysis is about 4.5 days. We have also shown herein that loadings of glucose up to 1.93 lb vs. ft^{-3} day^{-1} can be achieved in the methane digester at a residence time of only 1-day. It is noted that their average loading of 0.20 lb v.s. cft^{-1} day^{-1} and the volatile solids reduction efficiency of 40 percent are well within the range expected for a single-step, mesophilic digestion system (Table 7). Finally, of course, unreactive solids should be separated out after the hydrolysis so that the lowest possible viscosity feed is available for the methane digester.

A more successful two-stage technology with an anaerobic hydrolysis step will probably be based upon an expired series of patents by Langwell.[48-51] In this series, methods to obtain primarily acetic acid in concentrations up to 21 g ℓ^{-1} were described. These methods included: (i) mashing, steaming, or pulping agricultural residues; (ii) control of pH between 5 and 9; (iii) control of temperature between 60 and 70°C; and (iv) control of cellulose concentration to 6 or 7 percent during the 7- to 14-day batch reaction.

The primary strengths of an anaerobic first stage are simplicity of design and contamination-free, open operation. The weaknesses of this design concept are largely unknown due to the incomplete nature of previous investigations. Thus it is of

particular interest to learn the hydrolysis efficiency, the maximum, volumetric rate of acetic acid production, and whether or not inhibition by the acid product is evident.

A third technology for producing a lower viscosity, glucose substrate is the sulfuric acid catalyzed conversion of cellulose as illustrated in Figure 16. Brenner, Rugg and Rogers have shown that glucose yields of up to 50 percent could be achieved from pretreated newspapers in reaction periods of only 10-20 seconds. The sulfuric acid concentration was one percent and it was injected into the one-liter autoclave only after reaction temperature (220-230°C) had been reached. The newspaper was hydropulped and irradiated with a 5-10 megarad dose in a 3 MeV electron beam.

The strengths of this approach include a pretreatment process that is more effective and cheaper than ball milling and the demonstration of exceedingly rapid hydrolysis rates. The thrust of current research toward the development of a fully continuous process is also fundamentally sound. On the weakness side, the contribution to the cost of gas with the hydropulping-irradiation pretreatment is still very high. For example, if it is assumed that the substrate is 100 percent cellulose of which 50 percent undergoes acid hydrolysis, and if 80 percent of the hydrolyzed fraction is converted to gas of which 27 percent by weight is methane, and if the pretreatment cost is $0.015 per pound as suggested by Brenner, Rugg, and Roggers, then the pretreatment cost alone is $6.19 per 10^6 Btu produced as methane. While it is

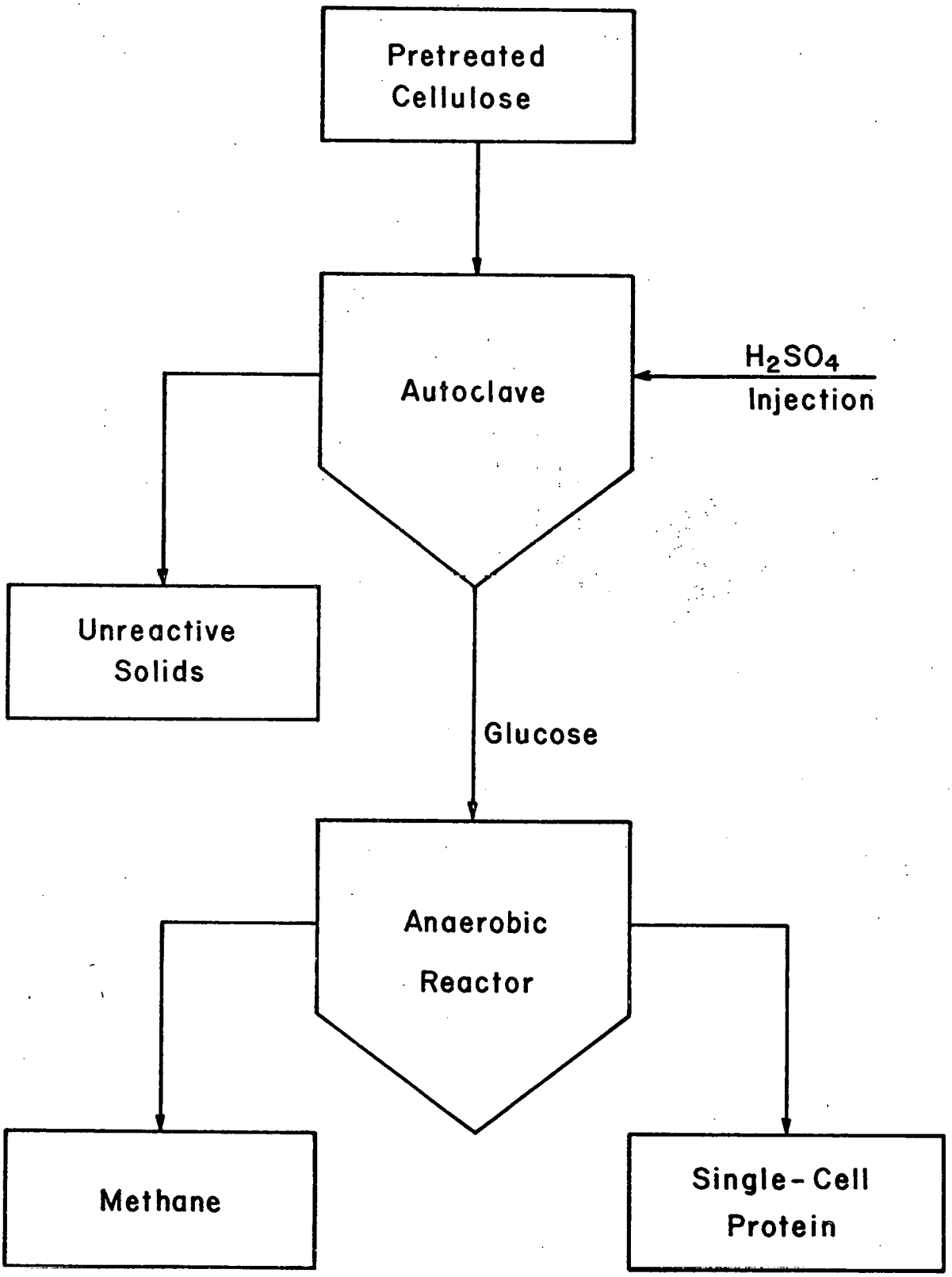

Figure 16. An advanced system for the fast production of methane using an inorganic acid-catalyzed hydrolysis reactor.

true that this cost is about a factor of four less than that for ball-milling to 50-micron particle size, this single contribution to the cost of gas is far greater than either interstate ($1.50 per 10^6 Btu) or intrastate ($2.00-$3.00 per 10^6) compensation for producers.

The usual procedure for reducing gas costs to a more competitive level involves the assumption of a credit for waste disposal. Although this economics is understood, it seems intuitively desirable to create through R&D more advanced technologies that do not require this kind of credit. At the very least it is axiomatic that the lower the credit, the more likely the success of the venture. Toward this end, it has been shown[5] that the pretreatment cost for shredding and alkali oxidation of waste cellulose is only about $1.00 per 10^6 Btu produced as methane in a single-stage digester. While the conditions of the two experiments were vastly different (10-20 sec vs. 4-6 day residence time, for example), the economic implication is quite clear.

In a similar vein, a real weakness of the acid hydrolysis may manifest itself in problems associated with the disposal of unreacted effluent and acidic liquor from the reactor. Key questions here relate to environmental impacts and costs of disposal.

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