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# Microbiological Purification Kinetics of Wine-Distillery Wastewaters

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**Abstract:** Wine alcohol distilleries produce eight volumes of high-strength waste (known as vinasse) from every volume of ethanol. The waste has an acidic character and a high organic content. Three adequate microbiological treatments (aerobic, mesophilic anaerobic and thermophilic anaerobic) for the purification of vinasses were examined. When around 90% biodegradable chemical oxygen demand (COD) removals were achieved in every treatment, optimum operating conditions had been attained, resulting in an optimum hydraulic retention times (HRT) of 8 days for aerobic, 6 days for mesophilic anaerobic, and 4 days for thermophilic anaerobic processes. The experimental results were compared with those obtained from a substrate utilization kinetic model. The model accurately predicted the performance of these processes, except at HRTs shorter than minimum, since these systems work under transient conditions. A comparison between kinetic coefficients obtained from the model showed that a thermophilic anaerobic process was the more efficient, since the process reached the same purification level, could be energy self-maintaining and needed smaller process plants than the other two systems.

Key words: aerobic treatment, anaerobic digestion, mesophilic, thermophilic, vinasses purification, purification kinetics.

## NOTATION

| | |
|---|---|
| Alk | Alkalinity (g $CaCO_3$ $dm^{-3}$) |
| AT | Aerobic treatment |
| B | Biogas produced at STP ($dm^3$ $g^{-1}$) |
| COD | Chemical Oxygen Demand (g $dm^{-3}$) |
| $COD_b$ | Effluent biodegradable COD (g $dm^{-3}$) |
| DO | Dissolved Oxygen concentration (mg $dm^{-3}$) |
| DVS | Dissolved volatile solids concentration (g $dm^{-3}$) |
| $DVS_b$ | Effluent (DVS (g $dm^{-3}$) |
| $E$ | Biodegradable treatment efficiency (%) |
| $f$ | Substrate utilization rate (dimensionless) |
| $f_{max}$ | Maximum substrate utilization rate (dimensionless) |
| $F$ | Substrate utilization rate (g $dm^{-3}$ $day^{-1}$) |
| $F_{max}$ | Maximum substrate utilization rate (g $dm^{-3}$ $day^{-1}$) |
| $k$ | Kinetic coefficient of substrate utilization model |
| $k_s$ | Substrate saturation constant of Monod equation (g $dm^{-3}$) |
| $M$ | Biomass concentration (g $dm^{-3}$) |
| MAT | Mesophilic anaerobic treatment |
| MR | Microbiological recount (colonies $cm^{-3}$) |
| $R$ | Ratio of the non-biodegradable substrate concentration to the initial input substrate concentration (dimensionless) |
| $S$ | Effluent substrate concentration (g $dm^{-3}$) |
| $S_o$ | Input substrate concentration (g $dm^{-3}$) |
| $S_b$ | Biodegradable effluent substrate concentration (g $dm^{-3}$) |
| $S_{bo}$ | Biodegradable input substrate concentration (g $dm^{-3}$) |
| TAT | Thermophilic anaerobic treatment |
| VA | Volatile acidity (mass/volume) |
| VS | Volatile solids concentration (g $dm^{-3}$) |
| $Y$ | Growth yield coefficient (biomass/substrate mass) |
| $\beta$ | Kinetic parameter of Contois' equation (dimensionless) |

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$\Theta$ Hydraulic retention time (days)
$\Theta_f$ HRT at which $f_{max}$ occurs (days)
$\Theta_{min}$ Minimum hydraulic retention time (days)
$\mu$ Specific growth rate of microorganisms ($day^{-1}$)
$\mu_{max}$ Maximum specific growth rate of microorganisms ($day^{-1}$)

## 1 INTRODUCTION

Wine distilleries produce large volumes of wastes, known as vinasses, with an acidic character (pH = 3·4–5) and a high organic content (20–100 g $dm^{-3}$ chemical oxygen demand, COD), which varies widely according to the raw material distilled: wine, lies, pressed grapes, etc.[1] Biological treatments have proved to be the most efficient methods for purifying these wastes,[2,3] due to the high rates of organic matter removal achieved.

A particularly important factor in microbiological treatment is the operating temperature, which determines the predominant bacterial flora in the medium and their growth rate.[4] There are three significant temperature ranges within which the process may take place: cryophilic (5–15°C), mesophilic (15–45°C) and thermophilic (45–60°C).

The present work compares the results of mesophilic aerobic, mesophilic anaerobic and thermophilic anaerobic processes using vinasses as substrate. Other cases were not taken into account since degradation within the cryophilic range is too slow and thermophilic aerobic processes are not suitable for high organic strength wastes since oxygen transfer is reduced at higher temperatures.

The experimental results of vinasse purification processes were compared to theoretical results obtained from a kinetic model of biological treatment fitted to high strength organic wastes: the substrate utilization model, proposed by Chen and Hashimoto.[5] The efficiency of the model is discussed with reference to process purification performances and a comparison of reactor volumes for each process is made.

## 2 MATHEMATICAL MODEL

According to Monod's model,[6] the specific growth rate of a microorganism, $\mu$, is given by:

$$\mu = \mu_{max} S_b/(k_s + S_b) \quad (1)$$

where $\mu_{max}$ is the maximum specific growth rate of the microorganism, $S_b$ is the limiting substrate concentration and $k_s$ is the value of the $S_b$ where $\mu$ has half its maximum value, $\mu_{max}$.

However, as previously reported,[7,8] the Monod model cannot be used to predict the kinetic behaviour of high strength organic wastewater anaerobic digestion, like vinasses, because the effluent substrate concentration should not be considered independent of the input substrate concentration, $S_{bo}$.

For this reason, Contois[9] has suggested the following model:

$$\mu = \mu_{max} S_b/(\beta M + S_b) \quad (2)$$

where $M$ is the biomass concentration and $\beta$ is a dimensionless kinetic parameter, which denotes the value of $S_b/M$ at which $\mu$ is half of $\mu_{max}$.

The main disadvantage of the Contois model, at least in anaerobic digestion, lies in the difficulty in measuring accurate values of microorganism concentration. To avoid this difficulty, Chen and Hashimoto[5] proposed the substrate utilization model, which is based on the definition of a dimensionless kinetic parameter, $k$, in the form:

$$k = \beta Y \quad (3)$$

where $Y$ is the growth yield coefficient, which under steady state conditions, is defined by:

$$Y = M/(S_{bo} - S_b) \quad (4)$$

The Contois kinetic function can now be expressed as:

$$\mu = \mu_{max} S_b/[k(S_{bo} - S_b) + S_b] \quad (5)$$

showing that $\mu_{max}$ occurs when $S_b$ approaches $S_{bo}$ (at wash-out) and that $\mu$ is zero when no substrate is available ($S_b = 0$). Furthermore, this equation gives the physical significance of $k$ as the value of the ratio between the unassimilated substrate concentration, $S_b$, and the assimilated substrate concentration ($S_{bo} - S_b$) at which $\mu$ is half of $\mu_{max}$:

$$\mu = \mu_{max}/2 \quad \text{if} \quad k = (S_b/S_{bo} - S_b) \quad (6)$$

Thus, $k$ is a coefficient which indicates that some type of inhibition occurs during the process since, when $k$ increases, $S_b$ approaches $S_{bo}$ and purification does not take place.

Other characteristics of the substrate utilization model are that: continuous or semicontinuous completely-mixed flow systems without solids recirculation are used; the predominant microorganisms in the biological treatment system are not present in the influent; the yield coefficient (ratio of the biomass concentration divided by the substrate concentration) is constant; cellular lysis is not taken into account; and effluent substrate concentration, $S$, is directly proportional to the input substrate concentration, $S_o$.

## 3 MATERIALS AND METHODS

The microbiological treatments used for vinasses were aerobic purification at 25°C, mesophilic anaerobic

digestion at 35°C and thermophilic anaerobic digestion at 55°C.

### 3.1 Biological reactors

Completely mixed reactors without sludge recycling were used. The capacity of the reactors was 2 $dm^3$ and the working volume was 1·8 $dm^3$ to avoid overflow of the foam produced. During aerobic purification, the medium was stirred by bubbling air into the reactors. Air flow (at STP) was 5 $dm^3$ per reactor $dm^3$ per minute.

In this type of reactor, the retention time for solids coincides with hydraulic retention time (HRT). To maintain the desired HRT, the required amount of vinasses was supplied daily to the reactors, in one feed.

The reactors were maintained at optimum processing temperature by immersion in thermostatic baths. All the experiments were conducted in duplicate reactors.

### 3.2 Vinasses

Vinasses were from distilleries using wine and lies (a by-product of wine fermentation) as raw materials. Lies-vinasses were previously centrifuged (at 1000 *g* for 5 min) to remove suspended solids. Since the supernatant from centrifugation showed similar characteristics to those from wine-vinasses,[10] the discussion below refers only to the treatment of wine-vinasses. A descriptive study of vinasses may be found in a previous publication.[1]

Vinasses do not contain microorganisms capable of aerobic or anaerobic digestion. Hence a previous start-up stage was necessary to acclimatize bacterial flora from other wastes to this substrate.[2,11,12] This work deals with the experimental data obtained once the start-up period was finished.

### 3.3 Reactor operation

Once the start-up of the reactors was achieved, a series of experiments was conducted to obtain optimum operating conditions for the processes. In each experiment, the amount of vinasses fed daily to reactors (and hence the HRT) was different. Tests were run in a range of HRT from 2 to 20 days, except that an HRT of 2 days was not used for the mesophilic anaerobic processes.

During the start-up of the reactors, the HRT was 20 days. When start-up finished and steady-state conditions (those at which volume and composition of biogas and COD removal remained constant) were achieved, the same HRT was maintained for 25 days to assure steady state. Then the HRT was changed and the next experiment was started. Every experiment was maintained until the steady state was achieved and then continued for 25 days. Samples from the reactor effluent were then collected and analysed, first as samples, then after centrifugation at 1000 *g* for 5 min.

### 3.4 Determination of non-biodegradable organic matter

The values of both the input and effluent biodegradable substrate concentration ($S_{bo}$ and $S_b$, respectively), which are needed to determine the kinetic parameters of the substrate utilization model, were obtained by determining the total digestibility.

Batch reactors were loaded with vinasses of known substrate concentration. These were fermented using adapted flora corresponding to every process, until biogas production was not detected (or COD was not removed). Then, incubation was continued for two weeks, after which analyses were made of the parameter quantifying organic matter, whose value corresponded to the amount of non-biodegradable organic matter.

### 3.5 Analytical procedures

All analytical determinations were carried out according to Standard Methods.[13] The parameters analysed in both the input and effluent of the reactors were: pH; alkalinity (Alk); volatile acidity (VA); COD; volatile solids (VS) and dissolved volatile solids (DVS); biogas produced at STP (B), carbon dioxide ($\%CO_2$) and volumetric methane ($\%CH_4$) content in the biogas; dissolved oxygen (DO).

The biomass of aerobic bacteria was determined following the Standard Plate Count procedure, also described in the Standard Methods[13] and expressed as microbiological recount (MR).

## 4 RESULTS AND DISCUSSION

### 4.1 Determination of non-biodegradable organic matter

Table 1 shows the values of dissolved non-biodegradable COD and dissolved non-biodegradable VS, obtained from the digestibility study. As observed, the average value of dissolved non-biodegradable COD (8%) was

**TABLE 1**
Results from the Digestibility Study with Different Treatments of Vinasses

| *Purification treatment* | *Dissolved non-biodegradable COD* | | *Dissolved non-biodegradable VS* | |
|---|---|---|---|---|
| | ($g\ dm^{-3}$) | (%) | ($g\ dm^{-3}$) | (%) |
| AT | 1·77 | 8·79 | 1·65 | 10·1 |
| MAT | 1·90 | 7·96 | 2·26 | 14·2 |
| TAT | 1·08 | 7·10 | 1·48 | 14·1 |

**TABLE 2**
Results Obtained on Optimization of the Operating Conditions for the Biological Reactors

| *Treatments/parameters* | *Hydraulic retention time (days)* | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | *20* | *10* | *12* | *8* | *7* | *6* | *5* | *4* | *3* | *2* |
| *Aerobic treatment* | | | | | | | | | | |
| pH | 8·41 | 8·15 | — | 6·93 | — | 6·61 | 5·53 | 5·14 | 4·96 | 4·43 |
| DO (mg $O_2$ $dm^{-3}$) 2·45 | 2·35 | — | 1·90 | — | 1·30 | 0·50 | 0·50 | 0·50 | 0·60 | — |
| MR (col. $\times 10^8$ $cm^{-3}$) 16·0 | 9·60 | — | 13·0 | — | 9·10 | 8·90 | 8·20 | 6·40 | 3·60 | — |
| *Anaerobic mesophilic treatment* | | | | | | | | | | |
| pH | 7·72 | 7·59 | 7·69 | 7·40 | 7·53 | 7·57 | 7·53 | 7·52 | 6·32 | — |
| Alk (g $CaCO_3$ $dm^{-3}$) | 7·90 | 8·25 | 8·01 | 8·14 | 9·16 | 9·27 | 10·1 | 9·61 | 9·73 | — |
| VA (g AcH $dm^{-3}$) | 0·93 | 0·93 | 0·85 | 0·78 | 0·83 | 0·72 | 2·83 | 3·29 | 7·28 | — |
| B ($dm^3$ $CH_4$ at STP $g^{-1}$ COD) | 0·24 | 0·24 | 0·23 | 0·24 | 0·24 | 0·24 | 0·22 | 0·21 | 0·07 | — |
| B ($dm^3$ $CH_4$ at STP $g^{-1}$ DVS) | 0·35 | 0·36 | 0·34 | 0·35 | 0·34 | 0·34 | 0·34 | 0·33 | 0·12 | — |
| $CO_2$ (%) | 23·6 | 25·2 | 23·4 | 29·1 | 29·4 | 27·3 | 34·6 | 35·6 | 59·9 | — |
| $CH_4$ (%) | 73·3 | 72·0 | 74·1 | 68·5 | 67·7 | 69·7 | 62·0 | 61·0 | 32·7 | — |
| *Anaerobic thermophilic treatment* | | | | | | | | | | |
| pH | 7·69 | 7·60 | 7·48 | 7·31 | 7·50 | 7·60 | 7·46 | 7·54 | 7·62 | 7·14 |
| Alk (g $CaCO_3$ $dm^{-3}$) | 6·18 | 6·01 | 4·64 | 4·90 | 5·39 | 5·47 | 6·19 | 6·10 | 5·54 | 5·65 |
| VA (g AcH $dm^{-3}$) | 0·32 | 0·43 | 0·73 | 1·02 | 1·16 | 0·54 | 1·95 | 1·50 | 1·51 | 3·78 |
| B ($dm^3$ $CH_4$ at STP $g^{-1}$ COD) | 0·40 | 0·36 | 0·23 | 0·20 | 0·30 | 0·33 | 0·27 | 0·25 | 0·17 | 0·07 |
| B ($dm^3$ $CH_4$ at STP $g^{-1}$ DVS) | 0·53 | 0·47 | 0·45 | 0·39 | 0·39 | 0·43 | 0·37 | 0·34 | 0·24 | 0·12 |
| $CO_2$ (%) | 23·0 | 27·2 | 28·9 | 30·9 | 30·7 | 28·6 | 31·8 | 30·1 | 32·1 | 47·0 |
| $CH_4$ (%) | 67·1 | 62·0 | 63·7 | 61·6 | 61·6 | 62·3 | 59·9 | 61·0 | 58·1 | 44·4 |

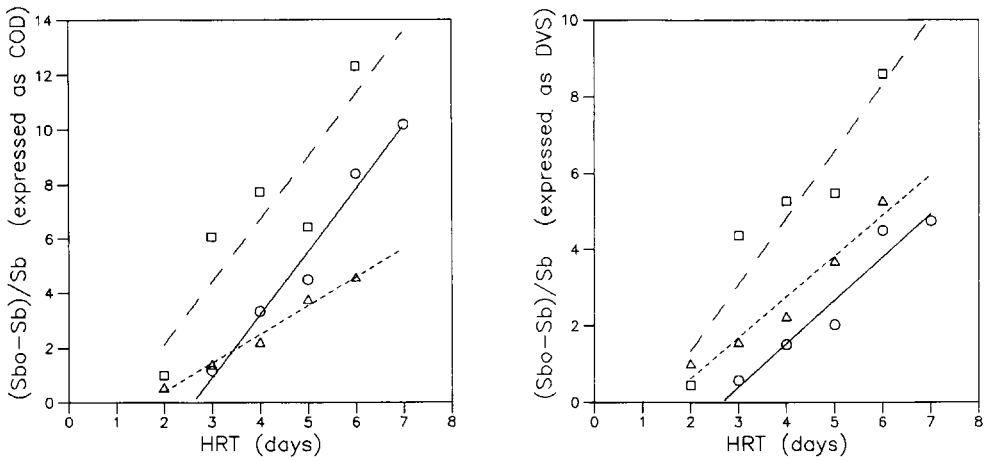


**Fig. 1.** Ratio of non-biodegradable substrate concentration (expressed as $COD_b$ or $DVS_b$) versus hydraulic retention time (○, MAT; □, TAT; △, AT). Lines are adjusted using the least squares method (——, MAT; — — —, TAT; ----, AT).

very low. The average value of dissolved non-biodegradable VS was only slightly higher (13%).

Some authors have indicated that this residual non-biodegradable organic matter is due to the presence of compounds like polyphenols, which are very difficult for the flora to break down.[14,15]

### 4.2 Optimization of processes

At the beginning of the optimization processes, after reactor start-up, with an HRT of 20 days, the process data obtained gave 72% COD and 42% VS removal for the mesophilic anaerobic process,[11] 63% COD and 55% VS removal for the thermophilic anaerobic process[12] and 70% COD and 47% VS removal for the aerobic process.[2]

The analysis results of the reactor effluents after the steady state was assured, are reported in Table 2 and Figs 1–4.

In the aerobic process, pH values fell as retention time decreased because of acidity of the unneutralized vinasses used. Biodegradable COD and DVS removals reached a maximum with 8 days HRT and maintained that level

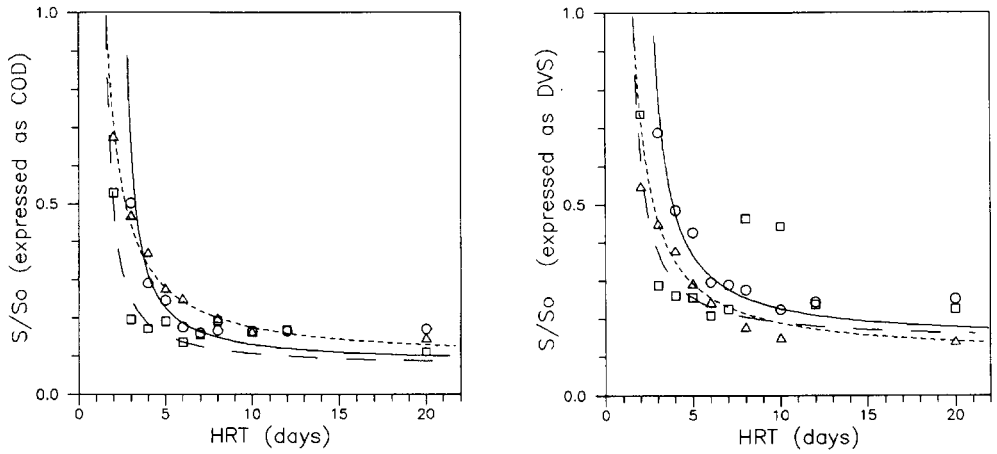


**Fig. 2.** Experimental values of the ratio between input and effluent total substrate concentration (expressed as COD or DVS) versus hydraulic retention time (○, MAT; □, TAT; △, AT). Lines denote the theoretical curves obtained from eqn (9) (——, MAT; — —, TAT; ---, AT).

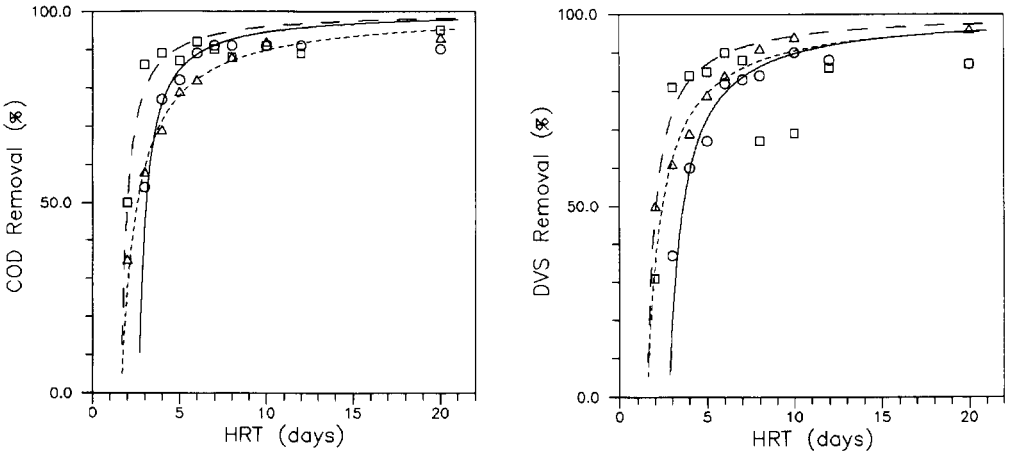


**Fig. 3.** Biodegradable treatment efficiencies (expressed as COD or DVS) versus hydraulic retention time (○, MAT; □, TAT; △, AT). Lines denote the theoretical curves obtained from eqn (10) (——, MAT; — —, TAT; ---, AT).

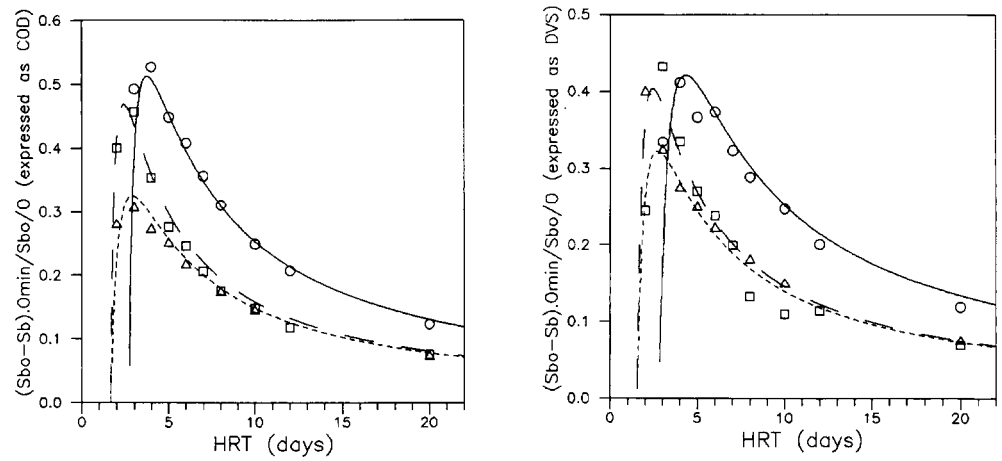


**Fig. 4.** Experimental values of substrate utilization rate (according to whether $S$ is expressed as COD or DVS) versus hydraulic retention time (○, MAT; □, TAT; △, AT). Lines denote the theoretical curves obtained from eqn (12) (——, MAT; — —, TAT; ---, AT).

over longer HRTs (Fig. 3) due to the presence of compounds such as polyphenols.[14,15] For HRTs of between 8 and 20 days, the $COD_b$ and $DVS_b$ removals were 90–93% and 91–95%, respectively, after the effluents had been centrifuged. These values are comparable to those obtained by other authors for aerated

lagoons[16] and for activated sludges,[17] in both cases with HRTs of 15–20 days.

The DO level in the medium fell as HRT was decreased; that is, the organic load (g COD $dm^{-3}$ $day^{-1}$) increased, due to the higher oxygen demand of the microorganisms when degrading organic matter. However, Table 2 shows that for HRTs lower than 5 days the DO value was maintained at 0·5–0·6 mg $dm^{-3}$, indicating that process purification performance may have been limited by the available DO. The fall in oxygen levels occurred simultaneously with a reduction in the number of the microorganisms in the medium, also confirmed by the microbiological recount. If HRT had decreased further, a point would have been reached where the regeneration rates of the flora and that of output of microorganisms were equal. At that point, the effect known as 'wash-out' would occur and consequently the purification capacity of the reactors would have ceased. The experimental minimum HRT for the aerobic process was estimated to be 2 days.

Table 2 and Figs 1–4 show two clearly differentiated areas in the case of the mesophilic anaerobic process. The first, between 6 and 20 days HRT, is characterized by a good link between the acidogenic and methanogenic phases of the anaerobic process, so that degraded products from the first were purified in the second. In this area, alkalinity, volatile acidity and pH remained constant. The second area, located between 3 and 6 days HRT, was unstable. Here, pH decreased and volatile acidity increased as a consequence of the unstable equilibrium between acidogenic and methanogenic flora. Alkalinity increased as a consequence of the need to add large amounts of alkali to maintain constant pH in the reactors. Under these conditions, small disturbances of the process conditions caused large disturbances in the performance of the reactors. For HRTs less than 3 days, wash-out ensued, nullifying the reactors' purification capacity.

Biodegradable COD and DVS removals reached a maximum with 6–7 days HRT, and maintained that level over longer HRTs.[14,15] For HRTs between 6 and 20 days, $COD_b$ and $DVS_b$ removals were 89–91% and 85–90% respectively, when the effluents were centrifuged (Fig. 3). These values are comparable to those obtained by sludge recycling[18] or anaerobic filters.[14] For 6 days HRT and longer, biogas and methane reached volumes of 0·34 $dm^3$ and 0·24 $dm^3$ (at STP) respectively per gram of COD fed to the reactors. These volumes agree with those found by other authors[14,18] for the same type of wastes. For HRTs under 5 days, the $CH_4$ content of the biogas decreased (and $CO_2$ increased) since the equilibrium between acidogenic and methanogenic flora shifted towards the former.

In the case of thermophilic anaerobic processes, Table 2 and Figs 1–4 show that all the studied parameters of the thermophilic anaerobic processes evolved similarly to the mesophilic process. The process stabilized for HRTs between 4 and 20 days, became unstable for HRTs between 2 and 4 days, with wash-out ensuing for HRTs less than 2 days. When the system was at steady conditions, $COD_b$ and $DVS_b$ removals were 89–92% and 85–95% respectively for the centrifuged effluents (see Fig. 3) and the biogas and methane produced reached volumes of 0·34 $dm^3$ and 0·25 $dm^3$ (at STP) respectively per gram of COD fed to the reactors.

For the same HRTs, both the volume of methane and COD and DVS removal were always greater for the thermophilic anaerobic process than for the mesophilic (except for 8 and 10 days' HRT, because the vinasses used were more dilute). On the other hand, volatile acidity and alkalinity were lower in all cases. This indicates that, for similar HRT's, the thermophilic process was more efficient for vinasses purification than the mesophilic processes.

If the experimental optimum HRT is defined as the minimum for which stable conditions are maintained and provided acceptable purifying performances are reached, then the optimum can be determined from the reported experimental data. For biodegradable COD removals of around 90%, the experimental optimum HRTs were 8 days for aerobic, 6 days for mesophilic anaerobic and 4 days for thermophilic anaerobic processes.

These experimental optimum HRTs provided the smaller laboratory scale reactor volumes for each feed flow rate and can be used for scale-up and pilot plant design.

### 4.3 Kinetics of microbiological treatments

From a mass-balance applied to microorganism concentration in a Continuous Stirred Tank Reactor, $\mu = 1/\Theta$ and as a consequence $\mu_{max} = 1/\Theta_{min}$. Equation (5) can therefore be transformed to the following linearized equation:

$$\Theta = 1/\mu = (\Theta_{min}) + (k\Theta_{min})[(S_{bo} - S_b)/S_b] \qquad (7)$$

Thus, $\Theta_{min}$ and $k$ can be graphically determined by plotting $(S_{bo} - S_b)/S_b$ versus $\Theta$, where the intercept is equal to $\Theta_{min}$ and the slope is equal to $k\Theta_{min}$.

Figure 1 shows the experimental substrate concentration values found in both the reactor input and effluent at the different retention times. The parameters of eqn (7) determined by fitting the equation to experimental data by linear regression using the least squares method,[19] are reported in Table 3. Whereas $\mu_{max}$ (and hence the inverse $\Theta_{min}$) does not depend on the form in which substrate concentration is expressed, $k$ does. $k$, however, is not dependent on the values of $S_{bo}$ for vinasses,[20] since their VS content was always less than 20 g $dm^{-3}$.[1] Only retention times less than 8 days have been taken into account to determine the kinetic parameters values, since at greater retention times the slope of the straight lines tended towards infinity. This is

**TABLE 3**
Kinetic Coefficients in Eqns (5) and (7). Average Values are Used in Eqns (16)–(18)

| *Purification treatment* | *Substrate concentration expressed as COD* | | | *Substrate concentration expressed as DVS* | | | *Average values (calculated from COD and DVS values)* | | |
|---|---|---|---|---|---|---|---|---|---|
| | $k$ | $\mu_{max}$ | $\Theta_{min}$ | $k$ | $\mu_{max}$ | $\Theta_{min}$ | $(k)_A$ | $((1/\Theta)_{max})_A$ | $(k)_A(\Theta_{min})_A$ |
| AT | 0·571 | 0·607 | 1·647 | 0·579 | 0·650 | 1·538 | 0·575 | 0·629 | 0·915 |
| MAT | 0·158 | 0·374 | 2·675 | 0·292 | 0·356 | 2·806 | 0·225 | 0·365 | 0·616 |
| TAT | 0·213 | 0·608 | 1·644 | 0·329 | 0·648 | 1·544 | 0·271 | 0·628 | 0·432 |

**TABLE 4**
Values of Maximum Substrate Utilization Rates, and HRT Values at which these were Reached

| *Purification treatment* | *Expressed as:* | $f_{max}$ | | $\Theta_f$ | |
|---|---|---|---|---|---|
| | | *From model* | *Experimental* | *From model* | *Experimental* |
| AT | COD | 0·33 | 0·32 | 2·90 | 3 |
| | DVS | 0·32 | 0·39 | 2·71 | 2 |
| MAT | COD | 0·51 | 0·52 | 3·73 | 4 |
| | DVS | 0·42 | 0·42 | 4·33 | 4 |
| TAT | COD | 0·47 | 0·47 | 2·41 | 3 |
| | DVS | 0·40 | 0·42 | 2·43 | 3 |

due to the fact that, for 8 days or greater HRT, the effluent substrate concentration was low and close to non-biodegradable organic matter because of the high microorganism concentration and the low feed flow rate. Under these conditions, the microorganisms have enough time to remove all the biodegradable substrate and $S_b$ tends towards zero. Consequently, the expression $(S_{bo}-S_b)/S_b$ tends towards infinity.

To compare the experimental and theoretical results obtained from the model, eqn (7) can be expressed as a function of the biodegradable substrate concentration:

$$S_b/S_{bo} = k/[(\Theta/\Theta_{min})+(k-1)] \quad (8)$$

or as a function of the total substrate concentration:

$$S/S_o = R+(1-R)k/[(\Theta/\Theta_{min})+(k-1)] \quad (9)$$

In Fig. 2, the experimental values of $S/S_o$ (expressed as COD or DVS, respectively) versus $\Theta$ are plotted, together with the theoretical curves obtained from model eqn (9). In this figure, the values obtained from the model (parametrized using only experimental data for HRTs of less than 8 days) agree with the experimental values at retention times between 3 and 20 days for the aerobic process and 4–10 days for both the mesophilic and the thermophilic anaerobic processes, which indicates the accuracy of the model in these intervals. However, at retention times shorter than those minima, the experimental and theoretical values are different as the systems lie in unsteady zones close to microorganism wash-out, in which small changes in the operating conditions lead to great fluctuation in purifying levels.

The biodegradable treatment efficiency, $E$, defined as a percentage of biodegradable substrate utilization of the input stream by the treatments, is given by:

$$E = 100(S_{bo}-S_b)/S_{bo} \quad (10)$$

The experimental values of $E$ are shown in Fig. 3, together with the curves obtained from eqn (10). In these figures, similar variations can be observed to those described earlier.

The volumetric substrate utilization rate of the treatment systems, $F$, defined as the organic matter degraded by microorganisms per unit time and reactor volume, is given by:

$$F = (S_{bo}/\Theta)[1-k/(\Theta/\Theta_{min}+k-1)] \quad (11)$$

where $S_{bo}/\Theta$ is the loading rate of the biodegradable substrate. Dividing $F$ by $S_{bo}/\Theta_{min}$, the following expression is obtained for the non-dimensional loading rate of the biodegradable substrate, $f$:

$$f = (F/S_{bo}/\Theta_{min}) = [1-k/(\Theta/\Theta_{min}+k-1)](\Theta_{min}/\Theta) \quad (12)$$

In Fig. 4, the experimental values of $f$ are plotted versus $\Theta$, together with the theoretical curves obtained from eqn (12). These values are expressed in two forms, according to whether substrate concentration has been expressed as COD or as DVS. Here, the theoretical and

experimental values agree throughout the retention times studied, except at those close to the minima.

Theoretical maximum substrate utilization rates, $F_{max}$, are determined by taking the derivative of $F$ in eqn (11) with respect to $\Theta$ and equating it to zero:

$$F_{max} = (S_{bo}/\Theta_{min})(1+k^{\frac{1}{2}})^2 \quad (13)$$

The dimensionless value, $f_{max}$, is:

$$f_{max} = (F_{max}/S_{bo}/\Theta_{min}) = 1/(1+k^{\frac{1}{2}})^2 \quad (14)$$

which occurs at:

$$\Theta = \Theta_f = (1+k^{1/2})\Theta_{min} \quad (15)$$

Both the experimental and theoretical values of the maximum substrate utilization rates found and the respective retention times at which they were reached, are reported in Table 4.

### 4.4 Comparison of biological reactor volumes

The volume of a biological reactor can be determined from the HRT process as a function of feed flow rate. In order to compare the volume of biological reactors for every purifying treatment used, an average specific growth rate of microorganisms, $\mu_A = 1/\Theta_A$, must be defined in the form:

$$\Theta_A = 1/(k)_A + [(k)_A(\Theta_{min})_A](S_{bo}-S_b)/S_b \quad (16)$$

where $(\Theta_{min})_A$ and $(k)_A$ are, respectively, the average values of $\Theta_{min}$ and $k$ calculated from the model (both from COD and DVS data). In Table 3, values of $(1/\Theta_{min})_A$ and $(k)_A$ are included.

Dividing expression $(1/\Theta)$ in eqn (16) (corresponding to the different processes studied when the purifying efficiencies are similar), the following are obtained:

$$(1/\Theta)_{A,TAT}/(1/\Theta)_{A,MAT} = [2{\cdot}740+0{\cdot}616(S_{bo}-S_b)/S_b]/[1{\cdot}592+0{\cdot}432(S_{bo}-S_b)/S_b] \quad (17)$$

$$(1/\Theta)_{A,TAT}/(1/\Theta)_{A,AT} = [1{\cdot}591+0{\cdot}915(S_{bo}-S_b)/S_b]/[1{\cdot}592+0{\cdot}432(S_{bo}-S_b)/S_b] \quad (18)$$

$$(1/\Theta)_{A,MAT}/(1/\Theta)_{A,AT} = [1{\cdot}591+0{\cdot}915(S_{bo}-S_b)/S_b]/[2{\cdot}740+0{\cdot}616(S_{bo}-S_b)/S_b] \quad (19)$$

where $(1/\Theta)_{A,AT}$, $(1/\Theta)_{A,MAT}$ and $(1/\Theta)_{A,TAT}$ are the average specific growth rates derived from eqn (16) for aerobic (AT), mesophilic anaerobic (MAT) and thermophilic anaerobic treatment (TAT), respectively. As HRT is the inverse of the average specific growth rate, for every feed flow rate, the ratio of the average specific growth rates between two purifying processes is the inverse of the reactor volume ratio. Thus, eqns (17, (18) and (19) can be transformed and rearranged, substituting $(S_{bo}-S_b)/S_b$ by the biodegradable treatment efficiency, $E/100$:

$$V_{MAT}/V_{TAT} = (2{\cdot}740-2{\cdot}124\,E)/(1{\cdot}592-1{\cdot}160\,E) \quad (20)$$

$$V_{AT}/V_{TAT} = (1{\cdot}591-0{\cdot}676\,E)/(1{\cdot}592-1{\cdot}160\,E) \quad (21)$$

$$V_{AT}/V_{MAT} = (1{\cdot}591-0{\cdot}676\,E)/(2{\cdot}740-2{\cdot}124\,E) \quad (22)$$

where $V_{AT}$, $V_{MAT}$ and $V_{TAT}$ are the required biological reactor volumes for aerobic, mesophilic anaerobic and thermophilic anaerobic treatment, respectively.

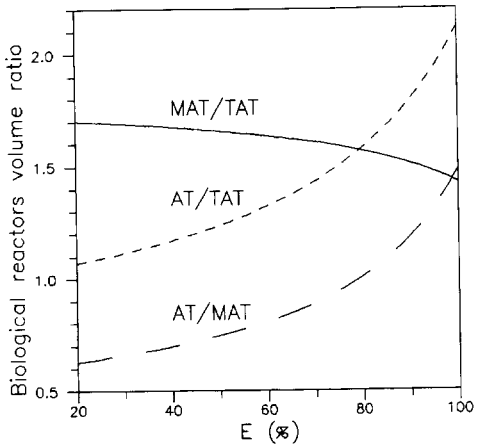


**Fig. 5.** Biological reactor volume ratios calculated from eqns (20), (21) and (22) versus purifying efficiency of treatments (——, $V_{MAT}/V_{TAT}$; ——, $V_{AT}/V_{TAT}$; ---, $V_{AT}/V_{TAT}$).

Figure 5 shows the variation of the different reactor volume ratios, defined by eqns (20), (21) and (22), as a function of the process purifying performance. For all the biodegradable efficiency ranges, $V_{AT} > V_{TAT}$ and $V_{MAT} > V_{TAT}$, which means that AT and MAT reactors are always larger than TAT. For biodegradable efficiency percentages up to 80%, $V_{MAT} > V_{AT}$, whereas for higher percentages $V_{MAT} < V_{AT}$. When the biodegradable treatment efficiency increases, $V_{AT}$ increases with respect to $V_{TAT}$ and $V_{MAT}$, and $V_{MAT}$ decreases with respect to $V_{TAT}$. Thus, for 90% purifying efficiency, biological reactor volume ratios are:

$$V_{MAT}/V_{TAT} = 1{\cdot}512 \quad V_{AT}/V_{TAT} = 1{\cdot}793 \quad V_{AT}/V_{AT} = 1{\cdot}186$$

and:

$$V_{AT} = 1{\cdot}186V_{MAT} = 1{\cdot}793V_{AT}$$

## 4 CONCLUSIONS

The three microbiological processes studied (aerobic, mesophilic anaerobic and thermophilic anaerobic) reach the same purification level when working at their optimum HRT, i.e. 90% COD removal. The optimum HRTs are 8 days for aerobic, 6 days for mesophilic anaerobic and 4 days for thermophilic anaerobic. Consequently, the thermophilic anaerobic process requires smaller reactors than the other processes since the

optimum HRT is shorter. Hence, fixed-capital investment for the installed process equipment is smaller for this treatment method than for the others.

Both anaerobic processes produce 0·24 $dm^3$ (at STP) of methane per gram of COD added to the reactor, which implies they are self-maintaining in terms of energy requirements and are more economical, unlike the aerobic process which requires additional expenditure due to aeration.

An increase in operating temperature from 35°C to 55°C (for mesophilic and thermophilic methods respectively) involves only a low additional energy cost as the vinasses leave the distilleries at 90–95°C.

For these reasons, the thermophilic anaerobic digestion has proved to be the best of the three vinasses purification processes.

The substrate utilization model gave accurate kinetics for every purification process studied, except for those retention times which make the systems unsteady.

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