TREATABILITY ASSESSMENTS: BATCH VERSUS CONTINUOUS CULTURE TESTS

By

James C. Young
4190 Bell Engineering Center
University of Arkansas, Fayetteville, AR

Thomas J. Irwin
Challenge Technology,
700 West 20th Street
Fayetteville AR

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ABSTRACT

Batch and continuous culture reactors are used widely for conducting treatability assessments of industrial wastewaters. Batch tests have the advantage of producing results on a rather rapid basis – typically hours or a few days. The major disadvantage of batch tests is that they represent a short term response of a seed culture to the test wastewater and can be affected by toxicity, lack of acclimation, or initial dilution effects. The real key to successful use of batch tests alone for conducting treatability assessment is an understanding of the conditions under which the test is run. Continuous reactor tests provide more realistic measures of treatability but require operation for sufficient lengths of time to allow steady-state conditions to be approached. Continuously operated reactors have the advantage of allowing sufficient time for culture acclimation and simulate full-scale operation more closely than batch tests.

KEY WORDS

Treatability, industrial wastes, respirometers, batch reactors, continuous reactors, kinetics

INTRODUCTION AND OBJECTIVES

Treatability assessments are essential to any consideration of industrial wastewater treatment. Such assessments often involve batch respirometric tests in which a known amount of industrial wastewater is added to a culture of microorganisms obtained from a laboratory reactor, a full-scale wastewater treatment plant, or commercial sources of specialized microorganisms. The response – as measured by oxygen uptake in aerobic biological reactions and gas production in anaerobic reactions – indicates potential biodegradability or toxicity. Unfortunately, batch tests alone can be of limited value if the wastewater is toxic at the levels tested, the culture requires acclimation, or the toxicity of the wastewater is reduced by sample dilution. These characteristic features of batch tests can be offset significantly by using repetitive batch or continuous reactors to simulate long-term impacts of the wastewater on biological processes. The objective of this paper is to illustrate the relative advantages and disadvantages of batch and continuous tests for assessing the effects of acclimation and toxicity in biological treatability tests. Case studies are used to illustrate the relative merits of each alternative.

BACKGROUND

Batch And Continuously Fed Reactors

Batch tests typically fall into two classifications: high rate and low rate as illustrated in **Figure 1**. A high rate batch test is a transient, non-steady state reactor to which a substrate is added to a seed culture having a relatively large biomass concentration. Typically the ratio of initial substrate concentration to biomass is about 1:1 to 5:1 but not so high that substrate toxicity occurs. The substrate concentration decreases relatively rapidly from an initial high to an extreme low over a short period of time – typically hours **(Figure 1A).** Biomass growth and decay occur throughout the reaction but the changes are small relative to the total active biomass. The principal advantages of using high-rate batch reactors is that the tests are simple to perform, and kinetic parameters can be obtained over a relatively short period of time – typically hours. This rapid response minimizes the likelihood of major shifts in microbial populations within the test duration.

An example of a high rate batch test is the oxygen uptake rate test in which a relatively high strength mixed liquor (the culture) is placed in a bottle with subsequent measure of the dissolved oxygen depletion occuring as a result of microbial respiration and biodegradation of the wastewater constituents. High rate batch tests are less susceptible to showing toxic effects than are low rate tests because the low F/M ratio minimizes negative impacts on the seed culture.

Low rate batch tests involve adding wastewater or test chemical to a relative small, but known, amount of seed organisms so that the F/M ratio ranges from 10:1 to 100:1 (Yang and Okos, 1987; Mulchandani and Luong, 1989: Young and Tabak, 1993). Low-rate batch tests are more likely to show toxic effects because of the relatively high F/M ratio **(See Figure 1B)**. A significant disadvantage of low rate tests is that they do not represent the response that would be seen in a treatment process where the wastewater is diluted in the mixed liquor. A well-known example of a low rate test is the dilution BOD test. Here, a sample of wastewater is diluted until a very small amount of substrate – typically less than 8 mg/L – is added along with sufficient seed to provide an F/M ratio of around 10:1. A distinguishing feature of the BOD test is that the concentrations of organic constituents are so low that the oxygen uptake rate is limited to first-order biodegradation kinetics. With high strength industrial wastes, this extreme dilution causes considerable distortion of biodegradation rates relative to a treatment environment and the dilution can reduce the concentration of constituents to well below toxic levels.

The use of batch tests for measurement of kinetic parameters must be accompanied by good experimental design together with appropriate mathematical techniques for analyzing the data (Dang, et al, 1989). Among other things, the experimental design must consider 1) proper balance between the initial substrate and biomass concentrations, 2) an adequate number of data points throughout the substrate uptake curve, and 3) the influence of decay if long time periods are required for substrate utilization. Method limitations often control the type of batch tests that can be run. For example, aerobic respirometers often are limited to low rate tests because of oxygen transfer limits in the reactions vessels (Li and Zhang, 1996). This limit is related to the size of the reaction vessel, the oxygen demand of the culture, and the mixing capabilities of the respirometer system.

More realistic indications of treatability can be attained by operating bench-scale reactors under continuous or semi-continuous feed conditions. These tests typically involve setting up a number of reactors each operated at different hydraulic and solids retention times. The reactors must be operated for sufficient lengths of time for steady-state conditions to occur with respect to both wastewater treatment and biomass growth. Such tests can allow for acclimation of microorganisms – which can occur slowly – and can show cumulative effects of toxic substances that may be in the wastewater or may accumulate as biodegradation products. For example, ammonia released during the biodegradation of high-strength protein wastes can inhibit nitrification when operating at low solids retention times.

Biodegradation Kinetics

One objective – but not always – of treatability testing is to determine biodegradation coefficients for use in process design and performance models. Biodegradation of organic materials typically is expressed by **Equations 1 and 2.**

The products, k_O **[k*]** and K_{SO} **[K_S*]** in **Equation 1** typically represent in situ values of k and K_S rather than intrinsic values. **Figure 2** shows a plot of this relationship and identifies critical points or regions relative to biodegradation tests and plant operations. Sensitivity analyses have indicated that the accuracy of estimating kinetic parameters is critically related to the number of points in the zero order (straight-line) part of the reaction and at low substrate concentrations especially near the break in the curve **(See Figure 1A)** (Davies-Venn, et al., 1992). Biodegradability and treatability tests are controlled by these relationships and test results must be interpreted accordingly.

Combining **Equations 1 and 2**, recognizing that the term M/(dM/dt) represents the solids retention time (SRT), and rearranging to solve for the substrate concentration – which now equals the effluent concentration, S_e, from a continuously-fed reactor – gives **Equation 3**. **Equation 4** gives an expression for estimating net yield from measures of gross or thermodynamic yield, Y_O, and SRT (Young, 1983).

Equations 1, 2 and 3 ideally apply to biodegradation of single organic compounds, when the microorganisms are acclimated, and when no toxic chemicals are present. Under such ideal conditions, **Equations 1 and 2** are used to estimate kinetic parameters – Y, k, and K_S – using non-linear numerical analysis techniques (Dang, et al, 1989; Young and Tabak, 1993). However, industrial wastewaters typically contain a number of constituents each having unique biodegradation parameters. Therefore, application of **Equations 1 - 3** to such wastewaters represent averages or composites of the biodegradation characteristics of the mixture.

CASE STUDIES

An example of the above relationship is shown in **Figure 3**. In this case, an industrial wastewater was added at four dilutions to respective respirometer vessels (AER-200 system, Challenge Environmental Systems, Fayetteville, AR). The vessels were seeded with unacclimated seed. As indicated in **Figure 3A**, the oxygen uptake reactions show at least three chemicals or chemical groups having different lag times and oxygen uptake rates. A second series of tests using the biomass from the first series as a seed culture shows significantly reduced lag times and more rapid biodegradation of the three constituent groups **(Figure 3B)**. Adding this wastewater proportionally to other streams contributing to an aerobic treatment process and using acclimated seed shows essentially no lag or negative impacts on biodegradation rates **(Figure 3C)**.

Kinetic modeling showed good fit of the composite data with the following values of kinetic coefficients: Y = 0.57 mg VSS/mg COD_f , k = 0.70 hr^{-1}, and K_S = 250 mg/L (at 22°C). The estimated biodegradable COD (bCOD) of the wastewater was 80% of the COD of the wastewater indicating that one or more constituents were not biodegradable under the test conditions. These data, if taken alone, should allow projection of net yields and effluent biodegradable COD as shown in **Figure 4**. The full-scale process receiving this mixed wastewater was operated at an SRT of 3 to 6 days and produced an effluent TCOD concentration averaging around 300 mg/L and a biodegradable COD of approximately 50 mg/L. Therefore, the batch tests produced a significant over-estimate of the effluent TCOD and bCOD. While the cause of this discrepancy is not known, it is possible that the culture was not well acclimated to the test wastewater or that there was some lingering inhibition at the levels of substrate used in the batch tests.

The results of another case study are illustrated in **Figure 5**. In this case, respirometer tests using unacclimated microorganisms did not show a lag in oxygen uptake, but did indicate inhibition because the oxygen uptake for the 50 ml sample – after correction for seed and dilution – was less than for the 20

mL sample **(Figure 5A)**. Continuously operated bench-scale reactors were fed this wastewater for 68 days to develop steady-state conditions. A second set of respirometer tests using the acclimated seed from the steady-state semi-continuous reactors **(Figure 5B)** indicates no lag and no toxic impacts. Kinetic analysis of these latter data using **Equations 1 and 2** produced the following parameters: Y = 0.45 mg VSS/mg COD_r, k = 1.1 mg COD_r/mg VSS-hr, and K_S = 110 mg/L **(See Figure 5C)**. Measured net yield and residual bCOD estimated from the continuously operated reactors are shown in **Figure 6** (symbols) along with estimates of the best fit model using **Equations 3 and 4** (dark lines). Estimates of the kinetic parameters from the batch tests using **Equations 3 and 4** showed reasonably good agreement with net yield values but substantial error occurred in estimates of residual bCOD. In this case, the continuously operated reactors produced the following estimates of kinetic parameters: Y = 0.55 mg VSS/mg COD_r, k = 1.4/hr, and K_S = 130 mg/L. The continuous reactor data are considered to give more reliable estimates of the kinetic parameters than are the batch tests because the data were collected over a longer period of time and do not rely on a single batch test.

Further comparison of short-term respirometric measurements to long-term continuously operated reactors is shown in **Figures 7 and 8** for anaerobic testing of an industrial wastewater. **Figure 7A** shows high-rate anaerobic respirometer test results when feeding a candidate industrial wastewater to an unacclimated culture. In this case, the feed-stock included a control plus three mixtures of control and wastewater plus one reactor receiving 100% wastewater. The ratio of feed volume to culture volume was 5% so that substantial dilution of the wastewater occurred. The data show slight reduction in methane production as the percentage of wastewater increased, but this inhibition was not considered sufficient to indicate that anaerobic treatment could not be accomplished. Therefore, a series of continuously operated bench-scale reactors were started. This test series included a control reactor, three reactors receiving mixtures of control substrate and test wastewater, and one reactor receiving 100% wastewater. All reactors initially were operated at a 20-day SRT and at an organic loading rate of 1 g COD/L-d. Cumulative methane production was measured over time of operation as shown in **Figure 8**. These data show that the wastewater immediately began to produce an increasingly negative influence as the percentage of test wastewater increased.

Batch respirometer tests on the 15th day of operation verified the substantial inhibition of methane production with the 100% wastewater showing essentially no methane production **(Figure 7B)**.

On day 17, the HRT and SRT of all reactors were changed from 20 to 40 days and feeding was continued at an organic loading rate of 0.5 g COD/L-d. The methane production rate began to increase immediately in all reactors **(Figure 8)**. The time of recovery to a constant methane production rate increased with increasing percentage of test wastewater, but after the 35th day of operation, all reactors were showing essentially the same methane production rate. The methane production rates in the test reactors at this point of operation was essentially the same as that for the control, thereby indicating essentially 100% biodegradability of the organic constituents in the wastewater.

A third set of respirometer runs using the seed culture from the continuously operated reactors after 35 days of operation showed reasonable agreement between the control and the reactors receiving test wastewater **(Figure 7C)** thereby indicating that the culture was fully acclimated and was able to degraded the wastewater constituents efficiently. This test series illustrates the benefits of both batch and continuously operated reactors for assessing treatability. While the initial respirometer tests showed no significant toxic impacts, they did not show the need for operation at a 40-day SRT. This need was identified easily from the continuously operated reactors.

CONCLUSIONS

Batch and continuously operated test reactors each make specific contributions to treatability assessments. Batch tests are good indicators of biodegradability and can provide estimates of kinetic parameters that give reasonable predictions of full-scale process performance. But batch data are reliable only if the wastewater contains nothing that is toxic to the test culture, when the culture is

acclimated to the wastewater constituents, and when no toxic wastewater constituents or by-products accumulate during treatment. However, it often is difficult to know that these conditions are met without conducting long-term tests. Also batch tests may produce misleading information if the wastewater is diluted to below toxic levels when conducting the tests. Ideally, a treatability assessment will include both batch and continuously-operated reactors. This combination provides more reliable estimates of kinetic and process parameters than can be obtained by either method alone.

ACKNOWLEDGEMENTS

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EQUATION 1

$$\frac{dS}{dt} = \frac{k_O [k^*] S M_a}{K_{SO} [K_S^*] + S}$$

EQUATION 2

$$dM_a/dt = Y\ (dS/dt) - K_d M_a$$

where:

S	=	Substrate concentration, mg/L
k_O	=	maximum specific substrate uptake rate, mg S/mg VSS-d
K_{SO}	=	half-saturation coefficient, mg/L
M	=	Active biomass concentration, mg/L
k^*	=	$(1 - I/I^*)^n$ for toxicant inhibition
K_S^*	=	$(1 - I/I^*)^m$ for toxicant inhibition
K_S^*	=	$(1 + K_S S/K_H)$ for substrate inhibition
I^*, K_H	=	inhibition coefficients
K_d	=	decay coefficient, day^{-1}

EQUATION 3

$$S_e = \frac{K_S [1 + K_d SRT]}{SRT (Yk - K_d) - 1}$$

EQUATION 4

$$Y_n = Y_O \frac{(1 + 0.2 K_d SRT)}{(1 + 1.2 K_d SRT)}$$

Figure 1 - Graphical illustration of the progress of substrate conversion and biomass concentration changes during high-rate (A) and low-rate batch (B) batch culture tests.

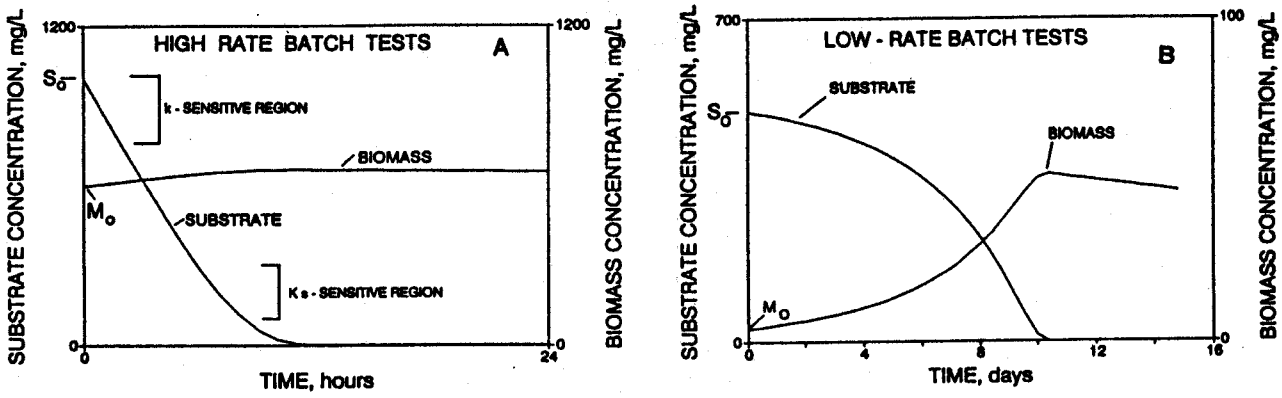


Figure 2 - Graphical illustration of the relationship of substrate conversion rate to substrate concentration as related to kinetic modeling.

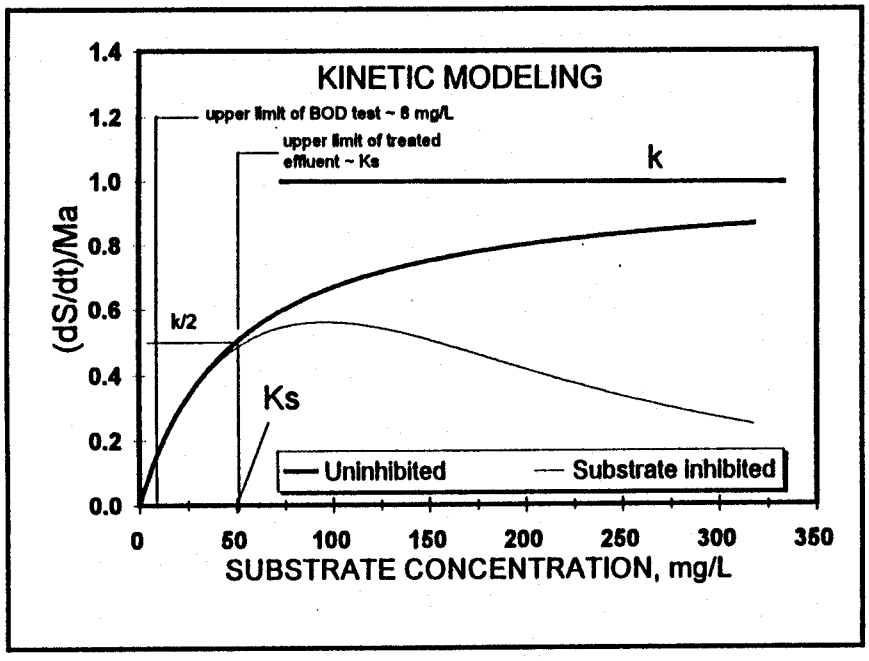


Figure 3 - **Batch oxygen uptake test results for an industrial wastewater when using an unacclimated culture (A and B) and an acclimated culture (C) including results of kinetic modeling of the acclimated culture data.**

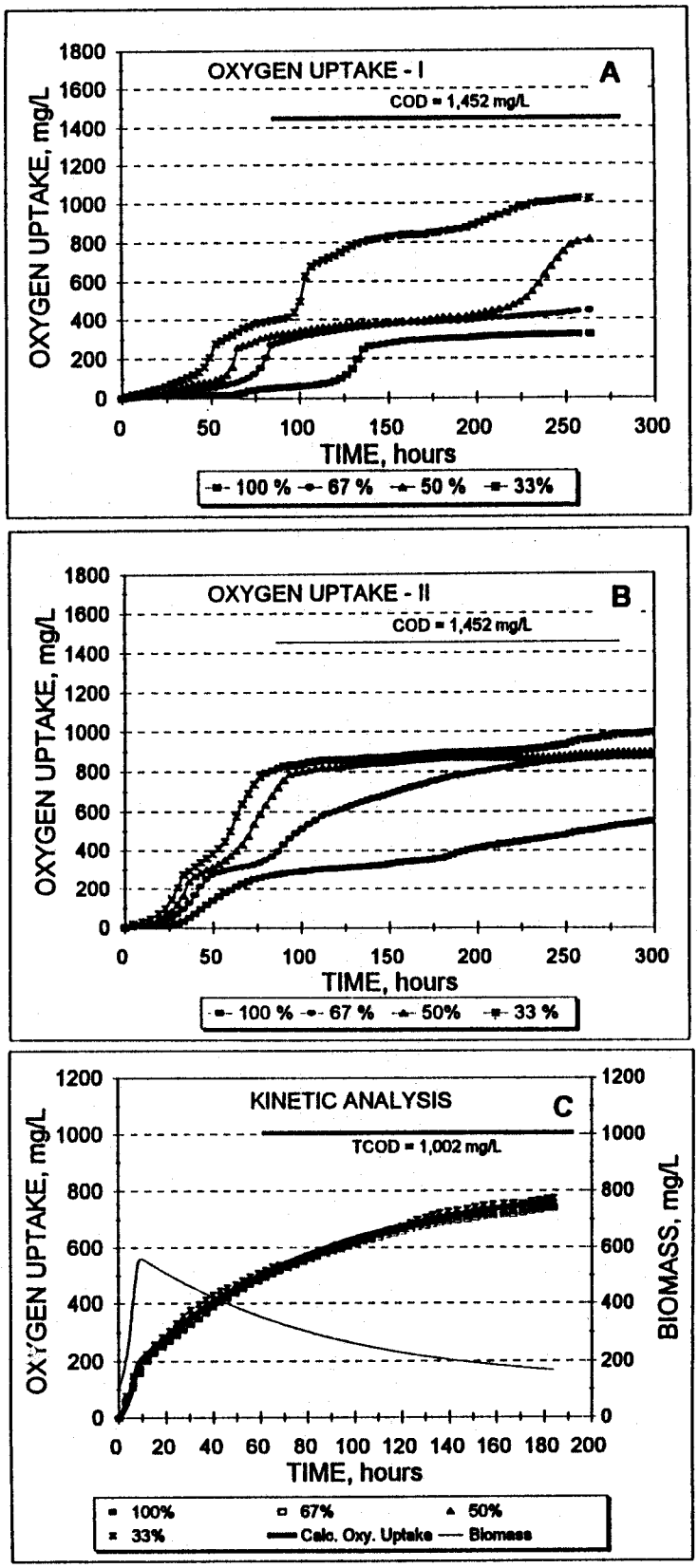


Figure 4 - Net yield and effluent biodegradable COD concentrations as estimated from the data shown in Figure 3C.

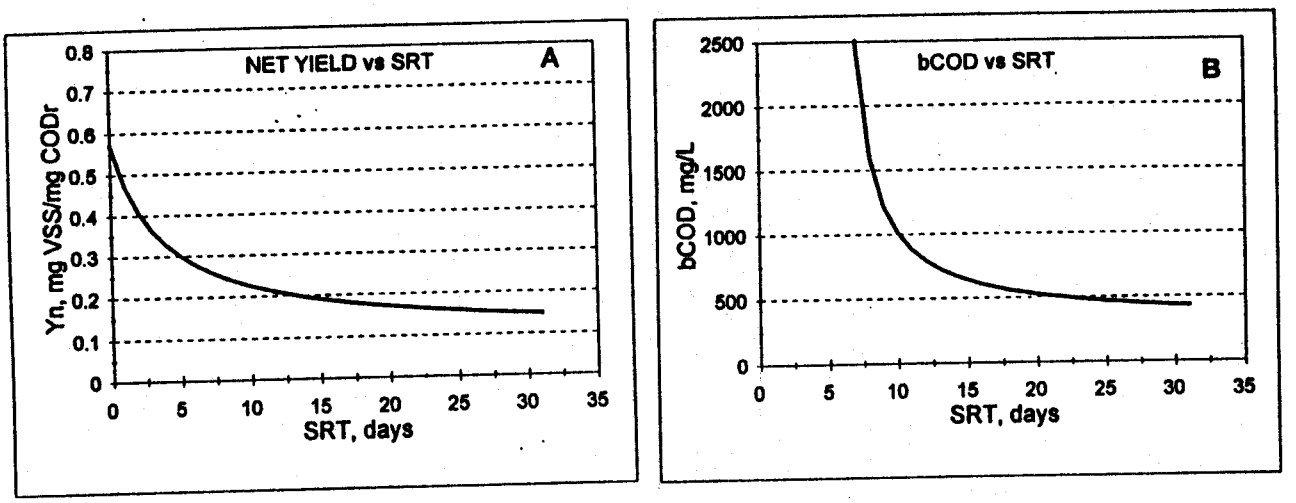


Figure 6 - Net yield and effluent biodegradable COD concentrations as estimated from continuously operated reactors including estimates from the data shown in Figure 5C

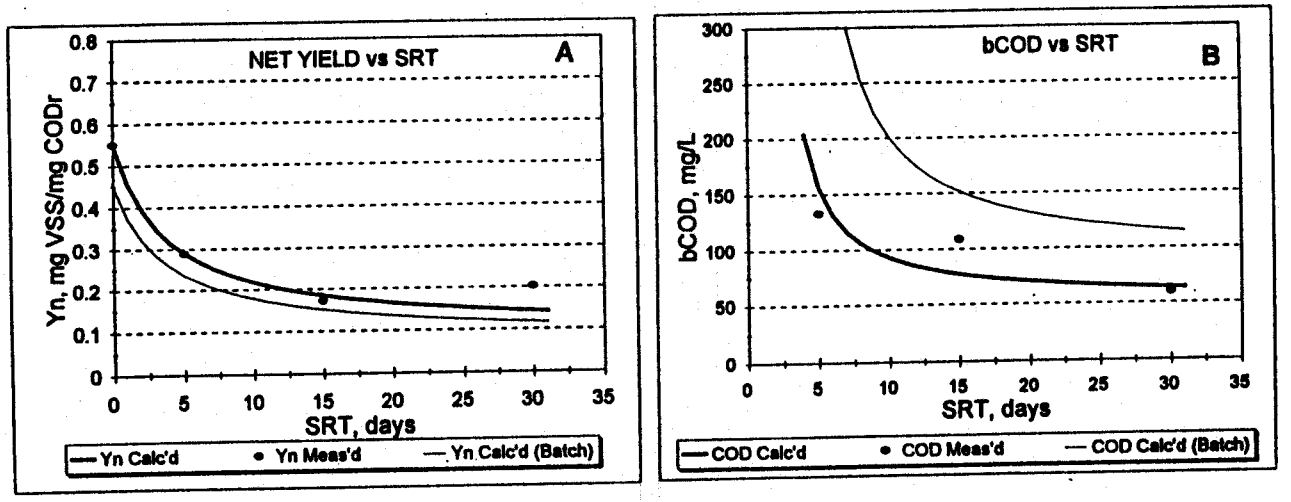


Figure 5 - Batch oxygen uptake test results for an industrial wastewater when using an unacclimated culture (A and B) and an acclimated culture (C) including results of kinetic modeling of the acclimated culture data.

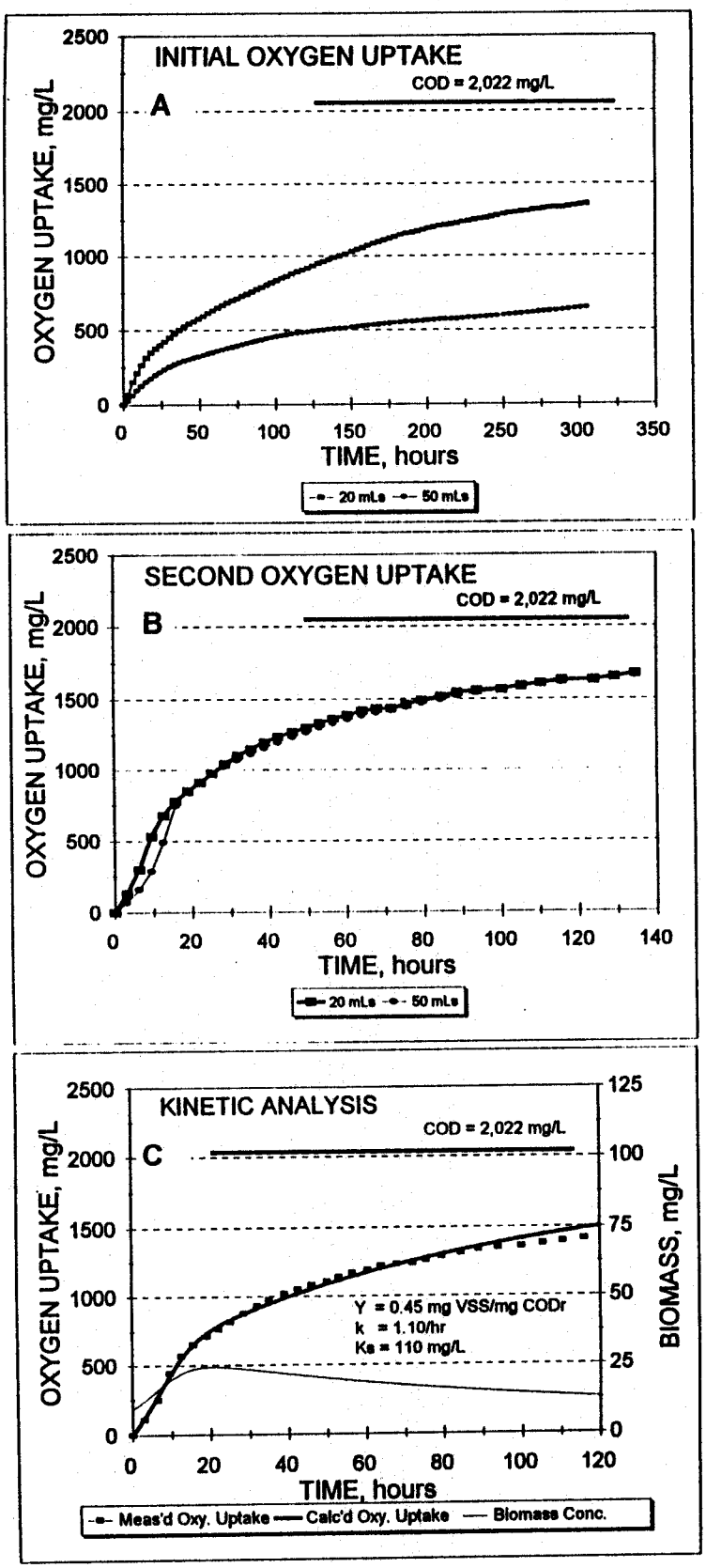
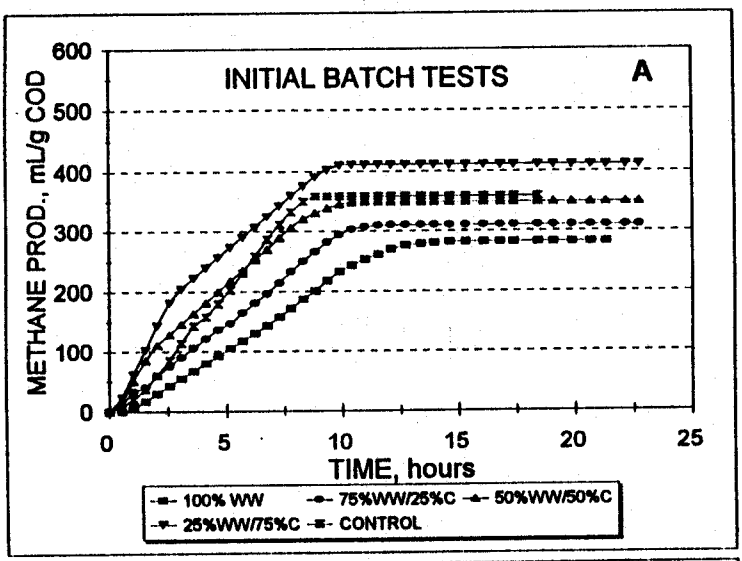
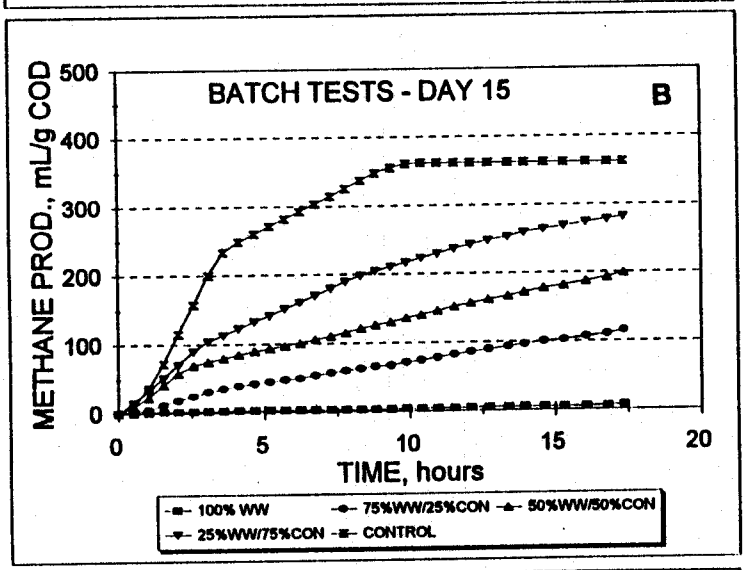


Figure 7 - Methane production during anaerobic batch tests of an industrial wastewater when using an unacclimated culture (A and B) and an acclimated culture (C)





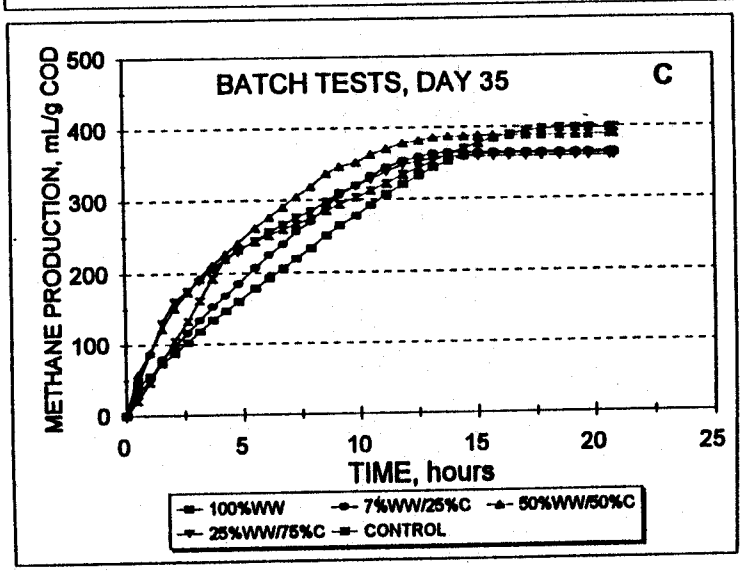


Figure 8 - Methane production during continuously operation anaerobic reactor tests of an industrial wastewater