Influence of Nutrients on the Biological Phosphorus Removal Process at High Acetate Concentrations

by

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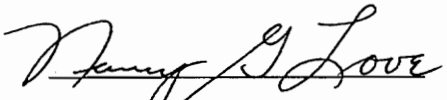
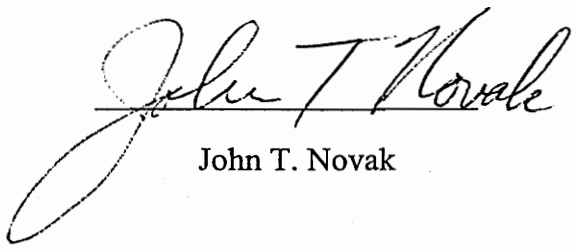
Approved :

Clifford W. Randall



Nancy G. Love



John T. Novak

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Environmental Engineering

(Abstract)

The objective of this study was to examine the influence of nutrients on the biological phosphorus removal process at high acetate concentrations. It was an extension of studies conducted by Randall and Chapin (1994), who found that industrial wastewater with high concentrations of acetate were able to inhibit the biological phosphorus removal process.

Two bench scale pilot plants were operated under controlled conditions that included synthetic wastewater as feed. The acetic acid concentrations in the feed of one system was increased in steps from 200 to 800 mg/L while the acetic acid concentrations in the feed of the other system was constantly held at 200 mg/L. Sludge from both systems was used for batch tests determining the kinetics of phosphorus release and uptake and poly-ß-hydroxybutyric acid synthesis. Furthermore, the influence of various nutrients were examined during these batch tests.

The results of this study confirmed the observations of Randall and Chapin (1994). High concentrations (600 mg/L) of acetic acid did inhibit the biological phosphorus removal process; however, this inhibition could be countered by adding calcium into the feed. The reactions of phosphorus release and uptake are described by first order kinetics.

Two hypotheses are proposed :

- A transport step across the cell wall or within the activated sludge floc is the rate determining step and limits the maximal speed of phosphorus release and uptake.
- Phosphate precipitation competes with the poly-phosphate accumulating bacteria for calcium, which is assumed to be essential for phosphorus release and uptake, thus inhibiting the phosphorus uptake.

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1. Introduction

While phosphate itself is not toxic, it may have detrimental effects on the environment by supporting the growth of algae (Cole 1988). These algae may in turn cause other problems such as taste and odor, oxygen depletion, or toxic effects (*Water Quality and Treatment*, 1990). For this reason, phosphate has to be removed from many wastewaters. This can be accomplished by two types of processes : chemical or biological phosphorus removal. Disadvantages of the chemical phosphorus removal are the increased sludge volume, high chemical costs and the possible increase of chloride or sulfate in the water (Winter 1989). Biological phosphorus removal not only avoids these problems, but can reduce the oxygen requirements too (Pokethitiyook, 1990; Wable and Randall, 1992).

A problem for the biological phosphorus removal (BPR) process is the rising need for denitrification. This process has high reaction rates with the same short chain fatty acids (SCFA) that are needed for the biological phosphorus removal process, leading to a conflict of interests for the planning engineer. While many proprietary BPR processes are trying to solve this problem by applying sophisticated ways to use several streams of return activated sludge, the amount of data to base a design on is sparse. Since the nitrification / denitrification process as well as chemical phosphorus precipitation are better researched that the BPR process, it is easier to build a conventional nitrifying and denitrifying wastewater treatment plant with chemical phosphate elimination than to build a fully biological treatment plant. On the other hand, the final disposal of sludge is increasingly becoming a problem for the operators of wastewater treatment plants. Since the BPR process reduces the amount of sludge, this process will steadily gain in importance.

Acetate is usually regarded as virtually the best substrate for biological phosphorus removal (Lötter, 1985; Abu-ghararah and Randall, 1991). On the other hand, Randall and Chapin (1994) found during experiments with industrial wastewater and mixtures of municipal wastewater and acetate that high dosages of acetate can inhibit the BPR process. Their research triggered this thesis. The purpose of the research for this thesis was:

- To set up and monitor two systems for biological phosphorus removal under controlled conditions with synthetic wastewater as feed.
- To increase the acetate load of one system to the point of breakdown.
- To operate the second system under constant conditions to have a defined source of biomass.
- To conduct parallel batch tests in order to collect additional information about the kinetics of the systems.
- To examine if nutrient deficiencies could have caused the problems observed by Randall and Chapin.

2. Literature Review

2.1 Introduction

All biological processes for the removal of conservative chemicals like heavy metals or phosphorus are based on the incorporation of the substance into the bacterial floc and the subsequent removal of it from the system together with the waste activated sludge. Therefore, when the phosphorus content of cells is increased, the mass of sludge that has to be wasted to remove the same amount of phosphorus is being decreased, making a higher mean cell residence time (MCRT) possible. All processes and mechanisms for biological phosphorus removal (BPR) discussed in the following literature review are based on this principle.

2.2 Beginnings of the BPR process

The average phosphorus content of sludge from wastewater treatment systems without enhanced biological phosphorus removal ranges from 1% to 2.5% (Metcalf & Eddy, p 365). Higher phosphorus contents were first observed independently by Srinath *et al.* (1959) and Alarcon (1961), with the phosphorus taken up during aeration (both sources as cited in Levin and Shapiro, 1965).

Levin and Shapiro (1965) succeeded in inhibiting the phosphorus uptake during aeration with 2,4-dinitrophenol. This substance selectively uncouples oxidative phosphorylation without uncoupling substrate phosphorylation (Rich and Yates, 1955). Based on this observation, they proposed the biological concept that the increased phosphorus accumulation in the sludge was "luxury uptake" by bacteria under aerobic conditions with release following under anaerobic conditions, like those encountered in a secondary clarifier.

On the other hand, Menar and Jenkins (1969) developed the theory that the luxury uptake was due to calcium precipitation of the phosphorus facilitated by hard water in connection with CO_2 stripping and subsequently increased pH, while the CO_2 equilibration in the undisturbed settling tank causes the pH to drop and the phosphorus precipitants to dissolve again.

Yall *et al.* (1970), however, traced the exchange of phosphorus and calcium between activated sludge and sewage with the help of the radioactive isotopes ^{32}P and ^{45}Ca. They observed parallel uptake and release of phosphorus by the bacteria cells under aerobic conditions after sewage and sludge were mixed. Furthermore, they noticed an increased phosphorus uptake by phosphorus depleted sludge. The calcium exchange, according to their measurements, was not correlated to the phosphorus uptake. Finally, they were able to discontinue the uptake of radioactive phosphorus by adding 2,4-dinitrophenol, a well kown oxidative phosphorylation uncoupler (Loomis and Lipmann, 1948). Based on these results, Yall *et al.* (1970) concluded that the enhanced phosphorus uptake is of mainly biological character. This statement was strengthened by Boughton *et al.* (1971), who added 2,4-dinitrophenol to enhanced phosphorus accumulating sludge from two wastewater treatment plants, thus inhibiting the phosphorus uptake. One of the examined sludges came from the same plant, on whose examination Menar and Jenkins (1969) based their precipitation theory. The final proof against the precipitation theory was given by Buchan (1981), after BPR systems had already been built. She examined the sludge from seven South African wastewater treatment plants for location and nature of the poly-phosphorus granules. The granules were located inside the cell and the calcium to phosphorus ratio in the granules of 0.2 mole/mole was too low for calcium precipitants.

Randall *et al.* (1970) examined phosphorus release under aerobic and anoxic conditions following previous phosphorus uptake and observed that the amount of phosphorus taken up is in a constant ratio to the amount released and that the anoxic release occurs at a higher rate than the aerobic release. They attributed these phenomena to bacteriolysis, further linking the phenomenon of biological phosphorus removal to bacteria.

Barnard (1975) developed in 1972/1973 a purely biological nutrient removal system (later called the Bardenpho® system) by combining anaerobic, aerobic and anoxic reactors. In addition to a high rate of nitrogen removal he observed extensive biological phosphorus removal as well. In 1975 he summarized these and other observation and proposed the following conditions as necessary for biological phosphorus removal (Barnard, 1976) :

- An anaerobic zone of sufficient size in which the influent is mixed with the return sludge for the release of phosphorus.
- Minimizing nitrification or complete denitrification to preserve the anaerobic conditions needed for phosphorus release.
- The phosphorus is taken up in the aeration basin.

2.3 Outline of the Biological Phosphorus Removal Process

2.3.1 Introduction

The basic design of a biological phosphorus removing treatment plant is based on several different, mainly proprietary, processes like A/O®, UCT®, Bardenpho® etc. (Arvin 1985). All these processes have basic elements in common. In the following text, function and peculiarities of these elements are pointed out.

2.3.2 Feed requirements

Systems for the biological removal of phosphorus need short chain fatty acids (SCFA) to induce phosphorus release or compounds that can be fermented to form SCFA (Wentzel *et al.*, 1988; Winter, 1989). The efficiency of various SCFA differs. Jones *et al.* (1987) compared several SCFA and their salts and found that sodium acetate was the substrate that triggered the lowest degree of phosphorus release, but that this phenomenon had no effect on the amount of phosphorus removal. Abu-ghararah and Randall (1991) added one to four carbon SCFA and their branched forms to the feed of an UCT® system. They found that, with the exception of the formic acid, all the examined acids enhanced the BPR process, with acetate and iso-valeric acid producing the best results. The iso-forms of the SCFA proved to yield superior results in comparison to their straight chain isomers.

In addition to SCFA, several cations are needed for the biological phosphorus removal process. Baxter and Jensen (1980) found the metals potassium, calcium, and magnesium in the polyphosphate granules of cyanobacterium cultures. The observations regarding the importance of cations for the biological phosphorus removal process can be divided between those regarding potassium / magnesium and those regarding calcium.

According to Wentzel *et al.* (1988), magnesium and potassium are necessary for the biological phosphorus removal process, while the exact role of calcium is unclear. Furthermore, Rickard and McClintock (1992) showed that potassium and magnesium

were essential for the BPR process by excluding them from the feed of a working BPR system, while excluding calcium did not upset the release / uptake cycle.

Several authors (Rickard and McClintock, 1992; Winter, 1989; Gerber *et al.*, 1987; Pattarkine, 1991) observed that the uptake and release of potassium and magnesium were parallel to the uptake and release of phosphorus with fixed ratios between these chemicals; their results are summarized in Table 2.1 while Figure 2.1 shows some of the results of Pattarkine (1991).

Regarding the importance of calcium, Pattarkine (1991) found differing correlation between P and Ca during uptake and release, with an uptake of 0.22 Ca/P [mol/mol] and a release of 0.05 Ca/P [mol/mol], but questioned the necessity of calcium for BPR other than for the general formation of cells (Figure 2.2).

Table 2.1 Observed Ratios between Phosphorus and Potassium / Magnesium

	Cations to Phosphorus Ratio during Release and Uptake	
Source	K : P [mole/mole]	Mg : P [mole/mole]
Rickard and McClintock (1992)	0.22	0.3
Gerber *et al.* (1987)	0.25	0.25
Pattarkine (1991)	0.23 to 0.43	0.25 to 0.36

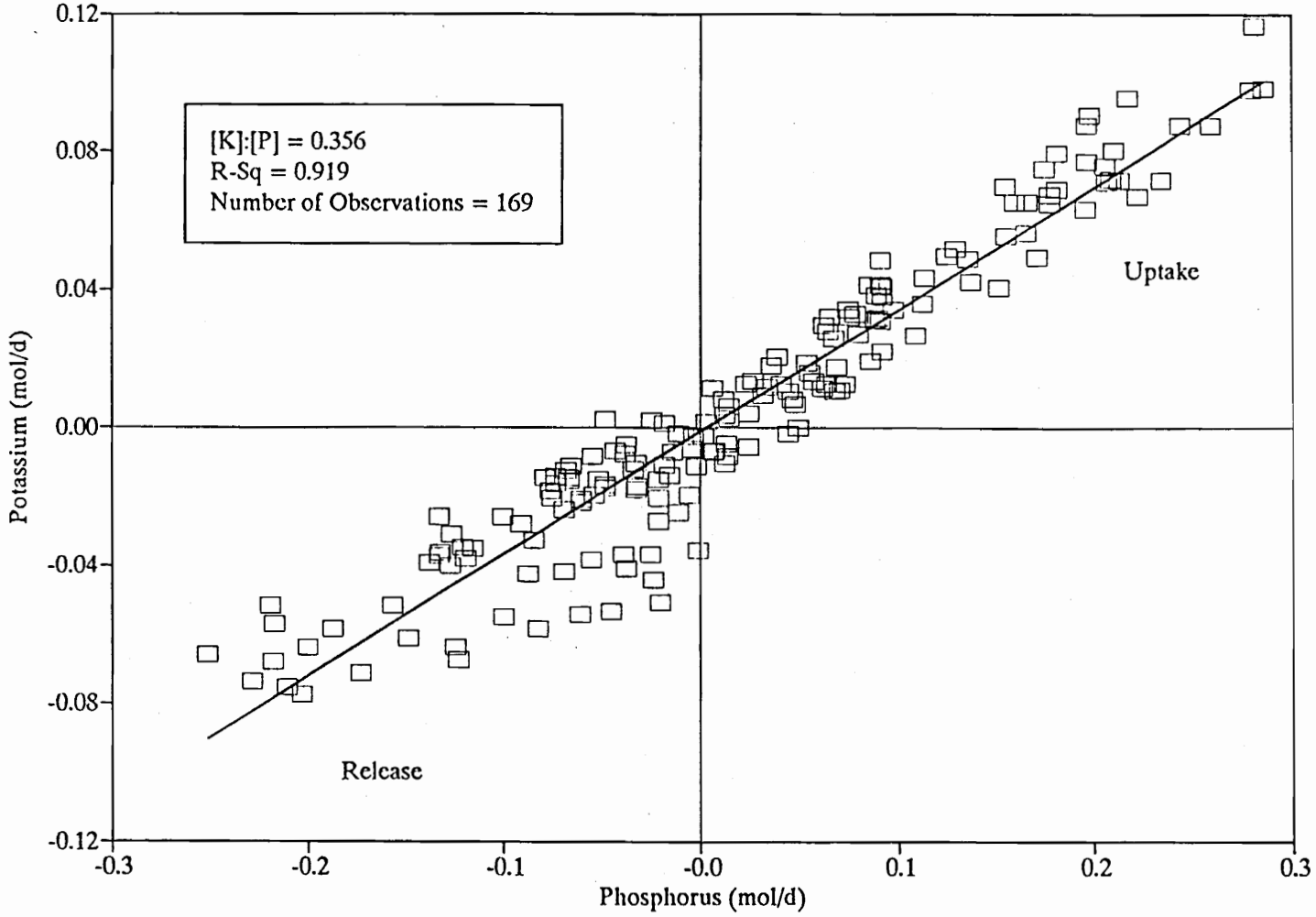

Figure 2.1 Release and Uptake of Potassium with Phosphorus, after Pattarkine (1991)

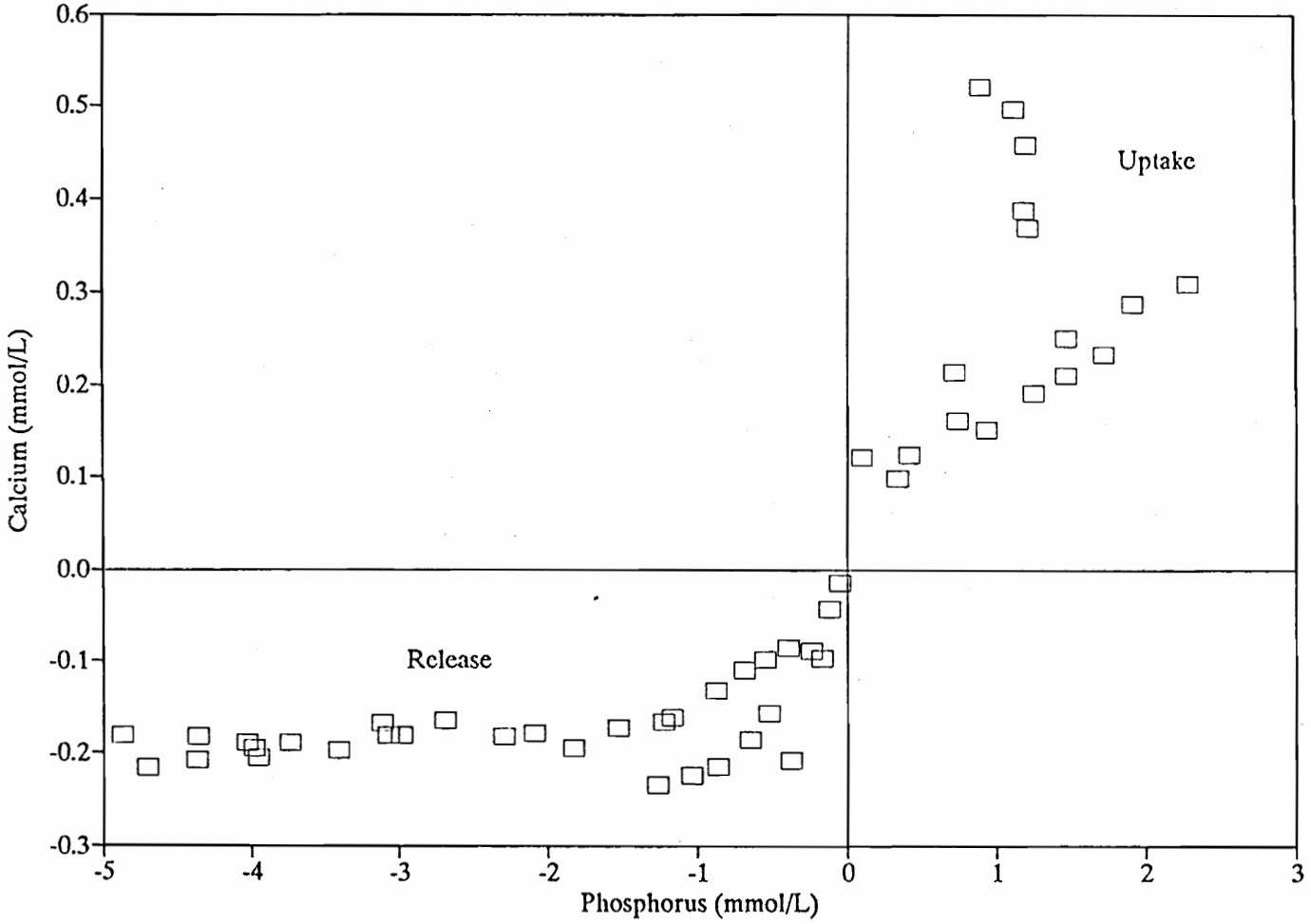

Figure 2.2 Release and Uptake of Calcium with Phosphorus during Batch Tests, after Pattarkine (1991)

2.3.3 Anaerobic Zone

In the anaerobic zone the phosphorus is released by the bacteria, while poly-ß-hydroxybutyrate (PHB) is synthesized (Fuhs and Chen, 1975; Comeau, 1986; Wentzel, 1991; Mino 1987). Gerber and Winter (1984) showed in an experiment with municipal wastewater as substrate that an extended anaerobic phase of 6 to 24 hours could improve the phosphorus removal without any negative side effects.

Furthermore, the processes in the anaerobic zone may have positive effects on the energy needed for the treatment of wastewater. Based on experiments with a lab-scale A/O® system, Wable and Randall (1992) hypothesized that "*aeration-induced stripping of reduced volatile species produced in the anaerobic zone*" causes the so called anaerobic stabilization (AnS). The oxygen requirements can be reduced by 15-20% in comparison to the theoretical oxygen requirements as a result of AnS (Wable and Randall, 1992). A more detailed description of the processes in the anaerobic zone is given in chapter 2.5.1.

2.3.4 Anoxic zone

Engineers use the term "anoxic" to describe a zone in which the electron acceptor is available in the form of nitrogen oxides. Whether the anoxic zone forms a problem for the biological phosphorus removal process or not seems to depend upon whether one visualizes the anoxic zone as an anaerobic zone contaminated by nitrates or an aerobic zone without oxygen.

If the former is the case, the main problem seems to be the effects of the nitrogen oxides on the fermenting bacteria rather than on the phosphorus accumulating bacteria, as Fuhs and Chen (1975) observed; for example, Gerber (1986) observed during batch-tests that nitrate did not block phosphorus release when one to three carbon SCFA were used as substrate. On the other hand, with other organic compounds no phosphorus release happened until all nitrate was denitrified. He concluded that this effect was due to the need of fermentation of these other compounds, with the nitrate blocking the

fermentation process. Furthermore, Gerber *et al.* (1987) observed that propionate and acetate triggered phosphorus release independent from the presence or absence of oxygen or nitrate, while glucose and ethanol depended on anaerobic conditions. Similarly, Mostert *et al.* (1988) observed that phosphorus release is possible under any (anaerobic / aerobic / anoxic) conditions if acetate or propionate are being used; but, under anoxic conditions, 50% to 66% of the acetate was used for denitrification and not for phosphorus release, thus reducing the efficiency of the whole BPR process. Lötter and Murphy (1985) found that 50% of the *Acinetobacter* spp. from a South African Bardenpho® plant were able to denitrify. This observation was generalized by Kerrn-Jespersen and Henze (1993), who observed that only a part of the poly-phosphorus bacteria were able to denitrify.

On the other hand, the anoxic zone can be used for phosphorus uptake. If this is the case, then the total oxygen requirements of the system can be reduced (Pokethitiyook, 1990).

2.3.5 Aerobic zone

In the aerobic zone the phosphorus released in the anaerobic zone is taken up while the previously stored PHB is utilized (Comeau, 1986; Wentzel, 1991; Mino 1987). However, in the presence of directly metabolizable substances like acetate or propionate (Hart, 1994; Mostert *et al.* 1988), phosphorus release parallel to the uptake may occur in the aerobic zone. This phenomenon had been observed earlier by Yall *et al.* (1970), but was not linked to specific mechanisms or substrates. Nevertheless, the insensitivity of the poly-P bacteria against aerobic conditions seems not to be without limitations. Rees *et al.* (1992) showed that the formation of PHB by strains of *Acinetobacter* could be blocked by high oxygen concentrations approaching saturation or phosphorus limitation.

Wentzel *et al.* (1985) observed a linear relationship between the amount of phosphorus released and the amount of phosphorus taken up, with the degree of the uptake being approximately 1.1 times higher than the release. In addition, Jones *et al.* (1987)

observed that while the phosphorus release was in related in a linear manner to the subsequent phosphorus uptake, the absolute removal was not. Reddy (1991) observed that the upper limits of the extent and efficiency of phosphorus uptake are controlled by neither MCRT nor the influent phosphorus loading, but by what he called the *"maximum phosphorus storage capability"*. A more detailed description of the processes in the aerobic zone is given in chapter 2.5.2.

2.3.6 Observations regarding precipitation

While the main part of the biological phosphorus removal process, as shown in the preceding text, is definitely caused by biological activity, there is some evidence that part of the enhanced phosphorus removal is caused by chemical precipitation. Chemical phosphorus removal processes precipitate phosphate with metal ions like Fe^{+3}, Al^{+3} or Ca^{+2}; for example, calcium reacts at a pH $\geq$ 10 with phosphate in a molar ratio of 10 mole Ca^{+2} per 6 mole PO_4^{-3} and forms hydroxylapatite $Ca_{10}(PO_4)_6(OH)_2$ (Metcalf & Eddy 1991, pp 306-309).

Regarding the phenomenon of phosphorus precipitation in connection with biological phosphorus removal, Lan *et al.* (1982) concluded from an experiment in which a bench scale A/O® system was fed with increasing calcium concentrations that a small part (15%-27%) of the phosphorus removal could have been due to calcium phosphate precipitation. In addition to that, Arvin and Kristensen (1983) concluded from experiments that the increase in pH caused by denitrification can lead to phosphorus precipitation in biofilms. Furthermore, Bark *et al.* (1992) examined various *Acinetobacter* spp. isolates by electron microscopy and electronspectroscopic imaging. Adsorption of phosphorus precipitants on extracellular polymers was observed for one of the strains, but the extent of adsorption could not be quantified. The possibility for partly biologically mediated phosphorus precipitation is further supported by Joko (1984), who observed that while a pH $\geq$ 9 is usually necessary to induce phosphorus removal by calcium, the presence of seed crystals can lower this border to a pH of approximately 8.

2.4 Bacteria involved in the Biological Phosphorus Removal Process

Two groups of bacteria control the BPR process: the poly-P accumulating bacteria, and the fermenting bacteria which provide the necessary SCFA if they are not present in the wastewater (Brodisch, 1985). Brodisch (1985) underlined the importance of fermenting bacteria for the BPR process by showing that the addition of acetate producing *Aeromonas* to a batch of *Acinetobacter* and settled sewage increased the phosphorus release. Similarly, Ye *et al.* (1988) observed limited phosphorus removal capabilities of pure strains of *Acinetobacter*, while mixing the cultures with aerobic bacteria improved the degree of phosphorus removal.

Ever since Fuhs and Chen (1975) were able to induce biological phosphorus removal by adding a pure culture of *Acinetobacter* to a working activated sludge system, *Acinetobacter* spp. are most often discussed as being responsible for the poly-P accumulation (Helmer, 1994). For example, Lötter (1985) and Lötter and Murphy (1985) found mainly *Acinetobacter* ssp. as the poly-P accumulating bacteria in activated sludge. Beacham (1990) found *Acinetobacter* spp. as the dominant phosphorus removing bacteria in the sludge from an UCT® plant that operated at temperatures between 18°C and 22°C.

On the other hand, Brodisch and Joyner (1983) or Cloete and Steyn (1988) questioned the predominance of *Acinetobacter* as phosphorus removing bacterium, because other bacteria seemed to be able to accumulate poly-P, too. For example, Auling *et al.* (1991) found during a survey of the sludge from eleven BPR wastewater treatment plants in Germany that in those with a high organic loading, *Acinetobacter* was not the dominant polyphosphate accumulating bacteria species. Kavanaugh (1991) found during a study of an UCT® pilot plant system at MCRT of 15 days and 20°C operating temperature that *Aeromonas* spp., *Pseudomonas* spp, and *Acinetobacter* spp. were storing excess phosphorus.

Helmer (1994) observed at operating temperatures of 15°C to 20°C in wastewater treatment plants that the *Acinetobacter* spp. no longer were the dominant species with regard to poly-P accumulation, and that at even lower operating temperatures the

Acinetobacter spp. had no influence at all. Furthermore, Wagner *et al.* (1994) examined the bacteria population in the sludge from a BPR treatment plant both with traditional plate counting methods and a method enumerating the amount of an *Acinetobacter* specific DNA section, in particular a *"16S rRNA-targeted oligonucleotide probe specific for the genus Acinetobacter"*. They found not only an unidentified gram-positive phosphorus accumulating group of bacteria in higher numbers than *Acinetobacter*, but that the plate counting method offered better growth conditions for *Acinetobacter*, skewing the results of plate counts in favor of *Acinetobacter*. These two studies may explain the contradicting observations regarding the importance of *Acinetobacter* spp. for biological phosphorus removal.

2.5 Biochemistry of the Biological Phosphorus Removal Process

In the following text, a short overview of two biochemical models of the BPR process is given. These models describe only the possible pathways with acetate as substrate, while other research (Gerber, 1987) clearly indicates that other substances as well are metabolized by a direct pathway. For a more detailed description of the two models, see Wentzel *et al.* (1991).

2.5.1 PHB accumulation and phosphorus release

Two models are most often used to explain the phosphorus release. On one hand, the model developed by Comeau (1986) and expanded by Wentzel *et al.* (1985, 1986) assumes that the energy necessary to synthesize PHB from acetate is obtained by metabolizing adenosintriphosphate (ATP) to adenosindiphosphate (ADP) with the help of some of the acetate; on the other hand, the model by Mino *et al.* (Mino *et al.*, 1987; Arun *et al.* 1988) and its adaptation by Wentzel *et al.* (1991) assumes that the necessary energy is generated by metabolizing ATP to ADP with the help of previously stored carbohydrates.

Figure 2.3 shows the Comeau/Wentzel model for phosphorus release. The transport of the acetate into the cell is assumed to be driven only by the concentration gradient across the cell wall, while the transport of cations and phosphate is facilitated by antiport protein carriers. Figure 2.4 shows the proposed mechanism for PHB synthesis in more detail. The overall stoichiometry of the anaerobic process for the PHB synthesis according to Comeau/Wentzel is shown in the following equation:

$$9\ \text{Acetate} + 9\ \text{ATP} + \text{CoASH} \Rightarrow (\text{PHB})_4\text{CoA} + 9\ \text{ADP} + 9\ \text{Phosphate-P} + 2\ CO_2$$

Figure 2.5 shows the Mino model for anaerobic phosphorus release. Its main difference is the way the NAD is reduced to NADH in order to facilitate PHB synthesis. Instead of assuming the use of acetylCoA in connection with the tricarboxylic acid (TCA) cycle, the Mino model assumes that carbohydrates are metabolized by the Embden-Mayerhof-Parnas (EMP) pathway with some of the resulting acetylCoA used as building blocks for the PHB synthesis. As a result, the number of carbons in the synthesized PHB

is higher than the number of carbons in the acetic acid utilized for the synthesis, as the following equation shows:

$$(C_6H_{10}O_5) + 6\ \text{Acetate} + 3\ \text{ATP} \Rightarrow (PHB)_4 + 3\ \text{ADP} + 3\ \text{Phosphate-P} + 2\ CO_2$$

Figure 2.6 shows the Mino model as extended by Wentzel for anaerobic phosphorus release. The only difference to the Mino model is that the carbohydrates are not transformed by the EMP pathway to pyruvate, but by the Entner-Doudoroff (ED) pathway, changing the amount of phosphate released per acetate utilized. The summarized stoichiometric equation for this model obviously contains a misprint in Wentzel (1991). Its correct form should be:

$$(C_6H_{10}O_5) + 6\ \text{Acetate} + 4\ \text{ATP} \Rightarrow (PHB)_4 + 4\ \text{ADP} + 4\ \text{Phosphate-P} + 2\ CO_2$$

The stoichiometry of the three different models is summarized and normalized with regard to acetate in Table 2.2.

Table 2.2 Theoretical molar ratios of phosphorus, PHB, and acetate during acetate uptake for different models

		Model		
		Comeau/Wentzel	Mino	Adapted Mino
P released / Acetate taken up	[mol/mol]	1	0.5	0.67
PHB formed / Acetate taken up	[mol/mol]	0.44	0.67	0.67
PHB formed / P released	[mol/mol]	0.44	1.34	1

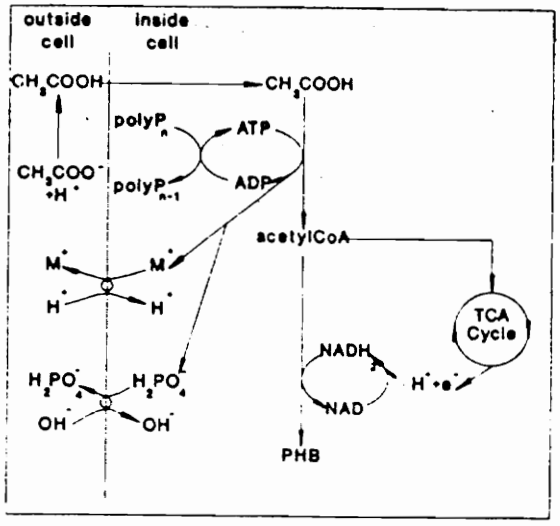

Figure 2.3 Flowchart for the Comeau/Wentzel model for anaerobic phosphorus release (from Wentzel, 1991)

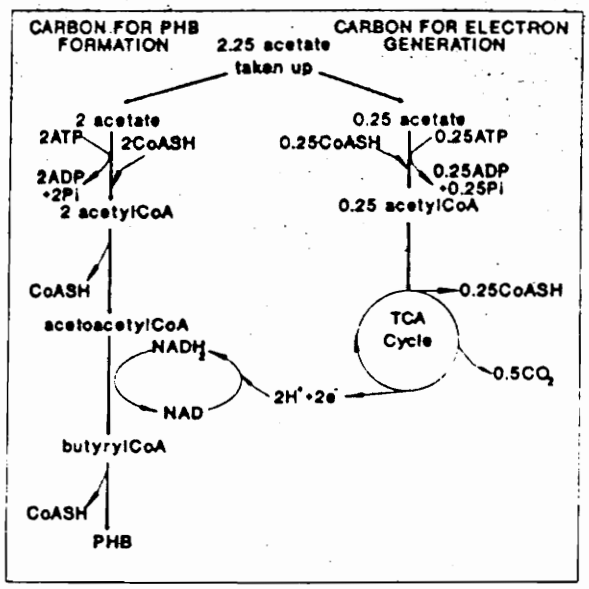

Figure 2.4 Flowchart of the PHB synthesis according to the Comeau/Wentzel model for the anaerobic phosphorus release (from Wentzel, 1991)

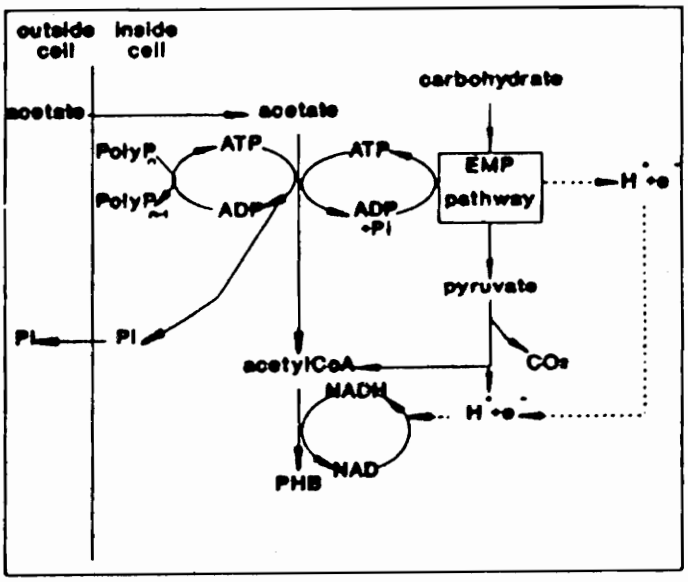

Figure 2.5 Flowchart for the Mino model for anaerobic phosphorus release (from Wentzel, 1991)

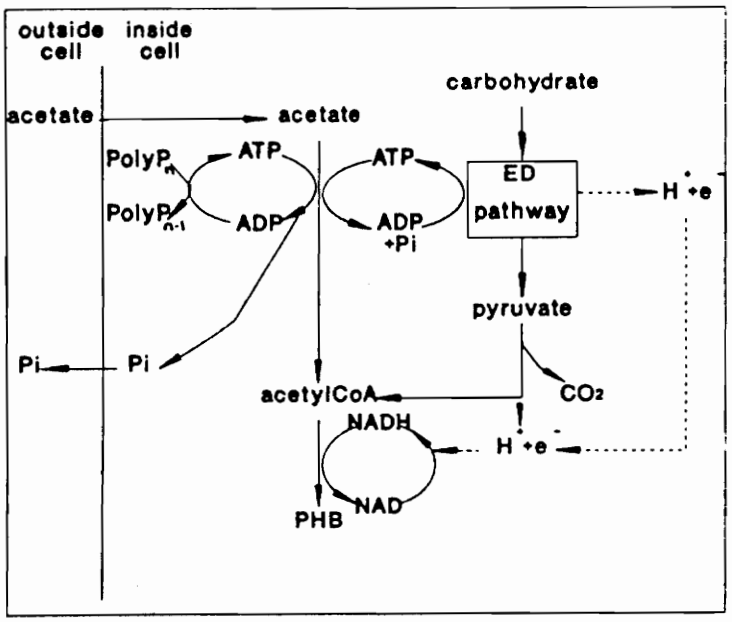

Figure 2.6 Flowchart for the Mino model for anaerobic phosphorus release as extended by Wentzel (from Wentzel, 1991)

2.5.2 Phosphorus uptake and PHB utilization

Figure 2.7 shows the Comeau/Wentzel model for aerobic phosphorus uptake. In it the PHB is either used to synthesize new cell material (anabolism) or used as an energy source (catabolism). The transfer of the cations and the phosphorus across the cell wall to build up the poly-P granula uses the same pathways as during phosphorus release. Since the PHB can be used for anabolism as well as catabolism, stoichiometric equations can be given only based on observations, but not based on theoretical considerations.

Figure 2.8 shows the Mino model for aerobic phosphorus uptake. It is identical to the Comeau/Wentzel model, with the exception that it makes no assumptions regarding cation uptake parallel to the phosphorus uptake.

2.6 Kinetics of the Biological Phosphorus Removal Process

The reactions for phosphorus uptake and release seem to follow first order kinetics; for example Wentzel *et al.* (1985, 1988) and Somiya *et al.* (1988) used first order kinetics to describe the process of anaerobic phosphorus release; Kerrn-Jespersen and Henze (1993) successfully used first order kinetics to describe the process of anaerobic phosphorus release as well as phosphorus uptake. According to these authors, the kinetic equations for phosphorus uptake and release are :

Release : $$C(t) = C_{max} \bullet [1 - e^{(-k \bullet t)}] \qquad (2.1)$$

Uptake : $$C(t) = C_{max} \bullet e^{(-k \bullet t)} \qquad (2.2)$$

where :

C(t)	=	Concentration at time t
C_{max}	=	Concentration at t=0 for uptake and t=∞ for release
k	=	Kinetic coefficient for speed of reaction

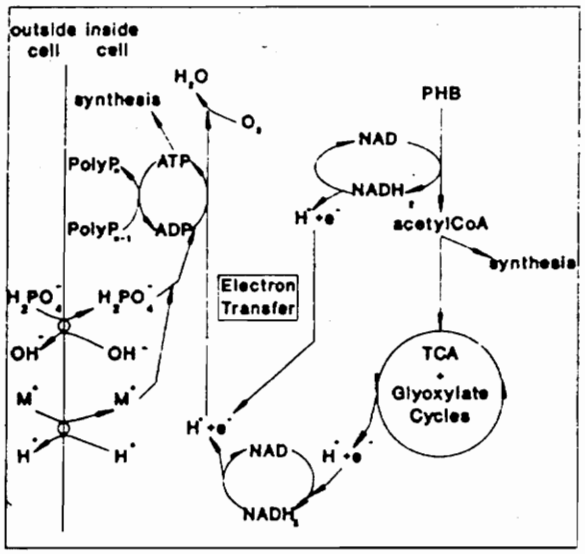

Figure 2.7 Flowchart for the Comeau/Wentzel model for aerobic phosphorus uptake (from Wentzel, 1991)

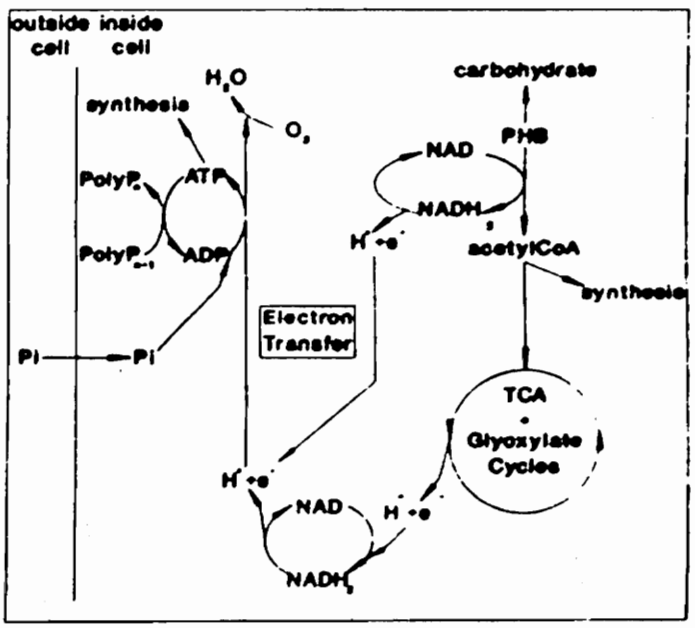

Figure 2.8 Flowchart for the Mino model for aerobic phosphorus uptake (from Wentzel, 1991)

2.7 Factors influencing the Biological Phosphorus Removal Process

2.7.1 Acetate loading

A high acetic acid load was found to be able to induce breakdown of the BPR process (Chapin 1993, Randall and Chapin, 1994). They assumed washout of the phosphorus removing bacteria as a probable cause (Figures 2.9 and 2.10). Furthermore, bulking of the sludge was observed when acetic acid entered the aerobic zone (Chapin 1993, Randall and Chapin 1994).

2.7.2 Glucose

In a study with radioactive ^{14}C, Bordacs and Chiesa (1989) found that both acetate and glucose were transformed into PHB during phosphorus release. The percentage of the acetate carbons utilized for PHB synthesis was higher than the percentage of the glucose carbons utilized, indicating different pathways and strengthening the Comeau / Wentzel model. Similarly, using varying ratios of acetate and glucose as the only carbon source in the feed of a batch reactor, Appeldoorn *et al.* (1992) found that an acetate to glucose ratio of 9:1 resulted in the highest poly-phosphorus accumulation, while the Mino model is based on a 6:1 ratio (see 2.5.1).

Cech and Hartmann (1990) discovered that competing bacteria were able to incorporate glucose without the release of phosphorus, resulting in the break down of the examined BPR system. They called these bacteria G bacteria. The glucose was probably stored as polysaccharides (Cech and Hartmann, 1993). This effect may explain the problems encountered by Okada *et al.* (1991), who were unable to accumulate poly-P bacteria at solid retention times of less than 25 days. They used a synthetic wastewater containing acetate and glucose. On the other hand, Morgan (1993) found that G Bacteria and poly-P bacteria are compatible, and that the presence of G bacteria does not necessarily cause failure of a working BPR system.

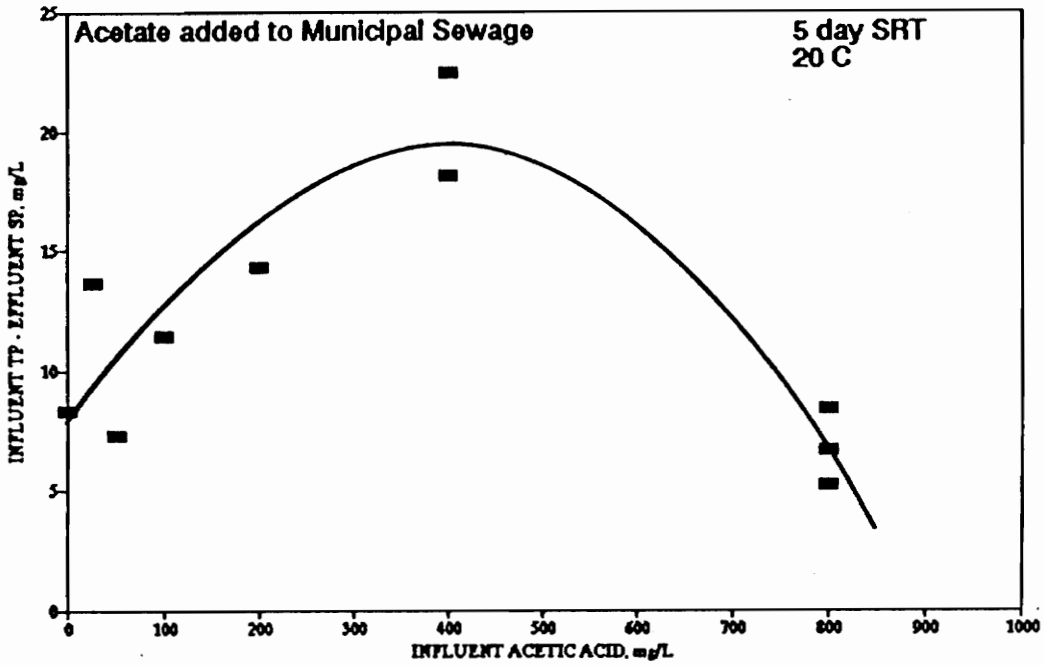

Figure 2.9 Effect of Influent Acetic Acid on BPR, after Randall and Chapin (1994)

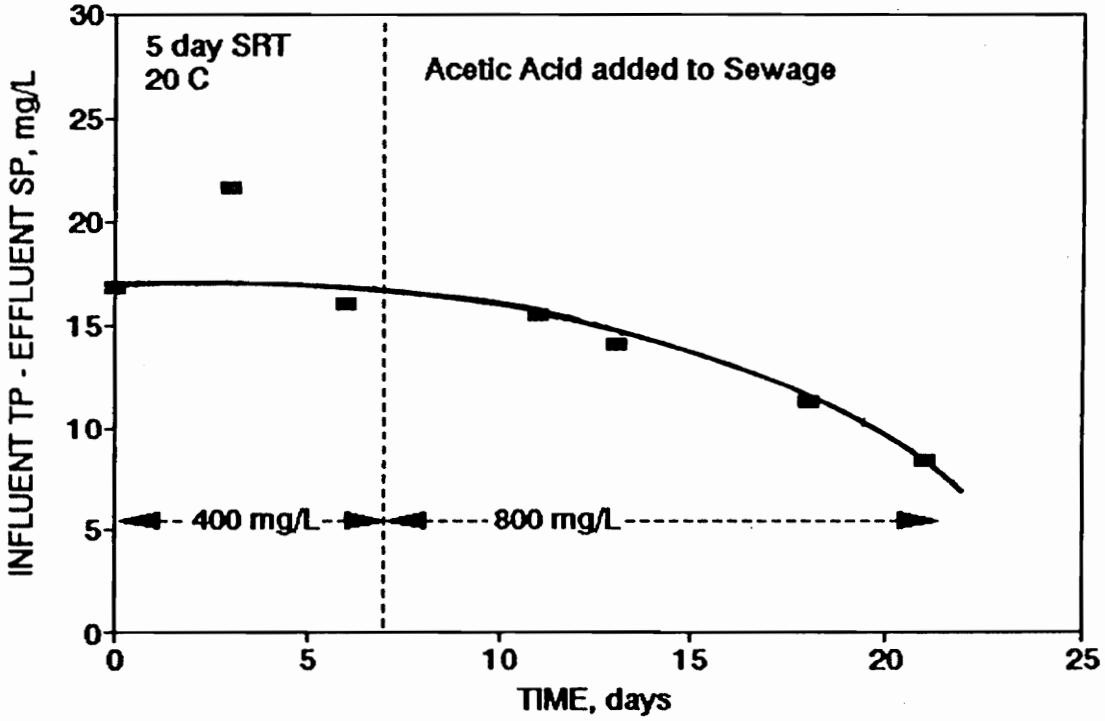

Figure 2.10 Decline of BPR with Acetic Acid Saturation, after Randall and Chapin (1994)

2.7.3 Temperature and MCRT

McClintock (1990) observed at low MCRTs an increasingly negative influence of low temperatures on the BPR process, with 5 days MCRT at a temperature of 10°C as the lower border. Furthermore, he found that complete nitrification was a reliable indicator of a sufficient MCRT for BPR. In addition, Mamais and Jenkins (1992) determined a connection between the influence of MCRT and temperature. They concluded that at lower MCRTs (below 2.9 days) the temperature controls the minimal necessary MCRT for maintaining the BPR process, while at higher MCRTs the temperature did not seem to be important. Nevertheless, the microbial composition of the sludge changes with MCRT and temperature: Hart (1994) showed that the MCRT influences the ratio of G bacteria to poly-P bacteria, and Helmer (1994) showed that the temperature influences which microorganisms are performing the phosphorus accumulation.

2.7.4 pH

The influence of the pH is more difficult to describe, since not only does the pH influence the efficiency of biological processes, but an effective BPR process also influences the pH. For example, Levin and Shapiro (1965) observed an optimal pH range for the BPR process of 7 to 8. On the other hand, Jones *et al.* (1987) observed an increase of the pH value in their system from approximately 7 in the influent to 8.1 in the effluent; Winter (1989) measured maximal pH values of up to 9 in the effluent. Wentzel *et al.* (1988) stated that this increase of the pH value did not seem to happen in the anaerobic zone, but in the aerobic zone, with pH values of up to 9.

2.8 Conclusion

The literature shows an interesting discrepancy in the behavior of biological phosphorus removal systems receiving acetic acid / acetate in the feed. On one hand, acetate seems to be without question the best substrate to enhance biological phosphorus accumulation, because it is a directly metabolizable carbon source. On the other hand, the results of Randall and Chapin show that acetate in high concentrations inhibits the biological phosphorus removal process. Therefore, the general objective of this research was to investigate this phenomenon of acetate-induced breakdown.

3. Materials and Methods

3.1 Introduction

The experiment consisted of two identical bench-scale pilot plants for biological phosphorus removal. Their purely synthetic feed contained sodium acetate and other nutrients. One pilot plant was operated under constant conditions, the other was fed with increasing sodium acetate and phosphate concentrations. The behaviors of the pilot plants were monitored. Sludge from both the pilot plants was periodically removed from the aerobic reactor and used for batch tests examining the influence of varying nutrient and acetate concentrations. Usually, the following parameters were measured during these experiments:

- Phosphate
- Nitrate and Nitrite
- Acetate
- MLSS / MLVSS
- Poly-ß-hydroxybutyric acid (PHB)

3.2 Pilot Plant Description

3.2.1 Setup of the pilot plant systems

The flow-scheme of both pilot plant systems is shown in Figure 3.1. The design loosely followed the layout of an A/O® biological phosphorus removal plant. Its layout and flow rates were changed several times during the start-up phase. The main problem that had to be addressed by these changes was the loss of biological solids in the effluent.

The feed for each system was stored in a 50 L carboy. It was pumped at a rate of approximately 0.85 L/h by a peristaltic pump to the system, where it mixed with the return sludge in a Y-connector and flowed into the anoxic reactor.

The anoxic plug flow reactor consisted of a length of Tygon® tubing (5/16 inch I.D.). It had a volume of 220 mL, resulting in a nominal hydraulic retention time (HRT) of 15 minutes and an actual HRT of 7.5 minutes. It was installed to denitrify the return sludge. This allowed the nitrogen bubbles to coalesce before the sludge entered the anaerobic rector. These larger bubbles were less able to cause floating sludge, which in turn was the main cause of spills involving the anaerobic reactor. Furthermore, the anoxic reactor might have improved the efficiency of the anaerobic reactor.

The anaerobic reactor consisted of a screw-top polyethylene jar with a volume of 1.5 L, resulting in a nominal HRT of 1.8 hours and an actual HRT of 0.9 hours. A 50 rpm paddle mixer kept the solids suspended. The outlet of the anaerobic reactor was submerged by one inch. This not only helped in keeping air out of the anaerobic reactor, but kept chunks of floating sludge from clogging the outlet tube, too.

The aerobic reactor consisted of an open top Plexiglas® tank with a volume of 5.8 L, resulting in a nominal HRT of 5.8 hours and an actual HRT of 2.9 hours. It was aerated by two diffuser stones.

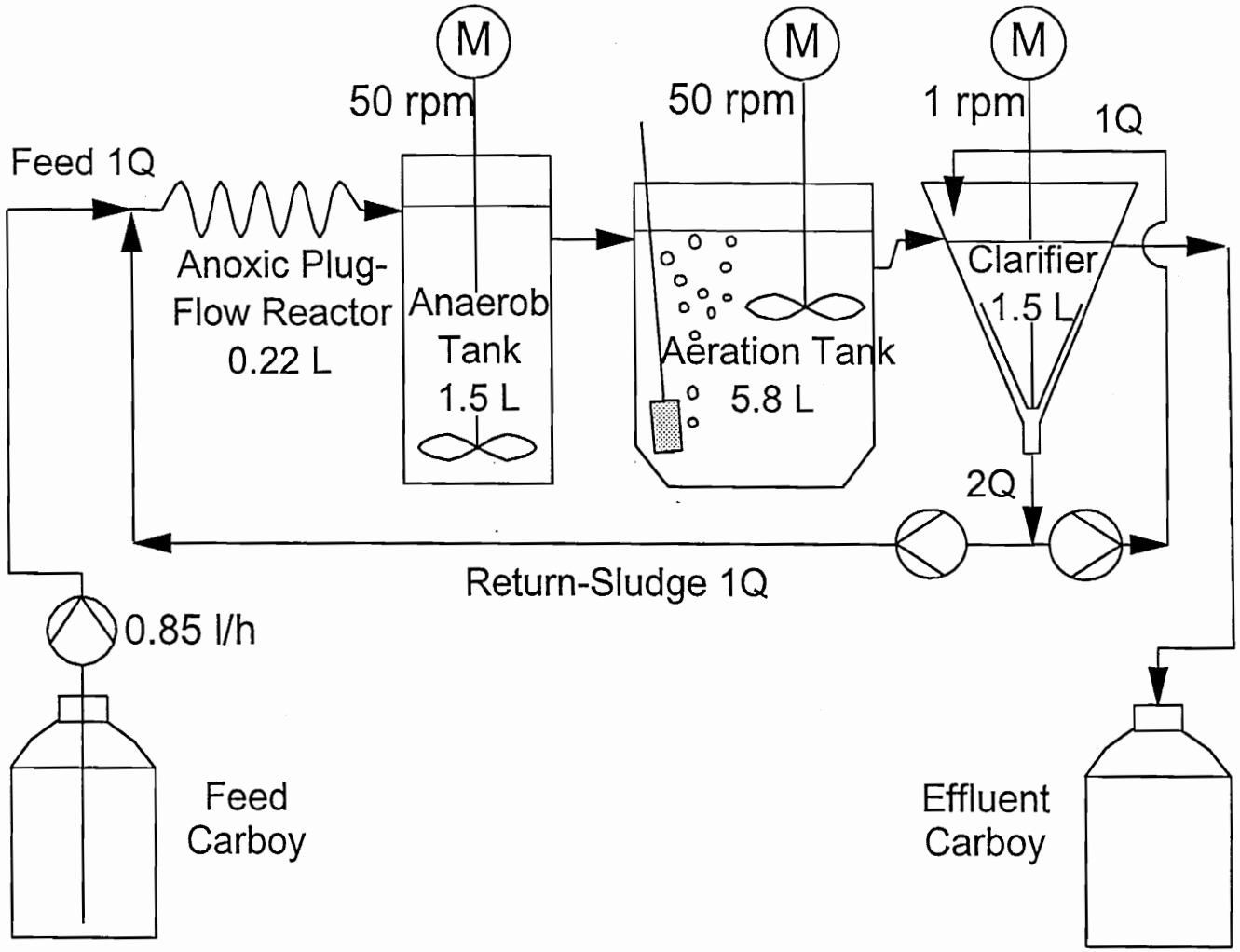

Fig. 3.1 Flow Scheme of the Pilot Plant Systems

While the eddy caused by the aeration alone kept the solids suspended, it did not extend into dead zone near the outlet opening, causing solids to be retained in the aerobic reactor. Therefore, a 50 rpm paddle mixer was installed near the outlet. The outlet of the aerobic reactor was submerged by one half inch.

The clarifier consisted of an 8.5 inch plastic funnel. Its surface area of 366 cm^2 resulted in a nominal surface loading of 0.02 m/h and a actual surface loading of 0.05 m/h. It was equipped with a 1 rpm scraper. On the axis of the scraper, a circular baffle (made from the bottom of a soda bottle) was located, extending approximately 2 cm into the water. It prevented short circuit flows effectively. The internal 1 Q recycle flow of the clarifier doubled the underflow of the clarifier. It was added because sludge bridged regularly just over the bottom outlet of the clarifier during the start up period of the experiment. This sludge blanket formed a filter, which led to a low solids concentration in the return sludge and massive sludge losses in the effluent.

The effluents of the two systems was separately collected in two 50 L carboys. This eliminated the need to be near a sink, allowing a more space efficient placement of the two systems in a constant temperature room. In addition to that, the use of carboys provided during the set-up period of the systems the possibility to collect solids lost from the clarifiers and to recycle them back into the system.

The pumps were electronically controlled peristaltic Cole Parmer Masterflex® pumps. One pump with four pumpheads handled the feed and the return sludge of both systems, while another pump with two pumpheads pumped the internal recycle of the clarifiers.

The sludge for seeding the first pilot plant came from an UCT® pilot plant operated on the Virginia Tech campus. After the first pilot plant operated with sufficient reliability, the second pilot plant was set up identical to the first. During the start up period of the pilot plants the sludges from the two systems were mixed several times to ensure that both systems developed with the same microbial population.

3.2.2 Operation and maintenance of the pilot plants

The pilot plants were located in a fully air-conditioned room. While the room temperature was constantly 20°C, the water temperature was usually 0.5°C to 1°C lower.

The systems were operated at a sludge age of approximately 10 days. As a rule, the amount of sludge wasted (usually approximately 500 mL to 600 mL) was calculated from the mixed liquor suspended solids (MLSS) and the effluent solids (SS_{eff}). When this was not possible, 500 mL of sludge were wasted from the aerobic tanks of each system.

The hydraulic retention time for both systems was 9 hours. The necessary flow rates through the pumps were checked by markings on the feed and effluent carboys.

The feed was made up every two days with tap water. On these occasions, the feed carboys were cleaned with diluted bleach. Additionally, all lines had to be cleaned once per week with diluted bleach. The contents of the feed are described in tables 3.1 and 3.2. After each increase in the feed of system I, no sludge was being wasted for two days to accelerate the acclimation of the system to the higher load.

Table 3.1 Contents of the Feed for System I

	Concentrations									
from :	5/25/94		6/6/94		7/1/94		7/21/94		8/4/94	
to :	6/5/94		6/30/94		7/20/94		8/3/94		8/12/94	
	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]
COD (theo.)	**393**		**500**		**606**		**820**		**1033**	
CH_3COONa	**273**	**3.33**	**410**	**5**	**546**	**6.66**	**820**	**9.99**	**1093**	**13.32**
as CH_3COOH	**200**		**300**		**400**		**600**		**800**	
Difco® Nutrient Broth§	180		180		180		180		180	
$CaCl_2$	104	0.94	104	0.94	104	0.94	104	0.94	104	0.94
as Ca^{++}	38		38		38		38		38	
$MgSO_4$	63	0.52	63	0.52	63	0.52	63	0.52	63	0.52
as Mg^{++}	13		13		13		13		13	
$FeCl_3$	3.2	0.02	3.2	0.02	3.2	0.02	3.2	0.02	3.2	0.02
as Fe^{+++}	1.1		1.1		1.1		1.1		1.1	
K_2HPO_4	**57**	**0.33**	**85**	**0.50**	**115**	**0.66**	**171**	**0.98**	**227**	**1.30**
as K^+	**25**	**0.66**	**38**	**0.98**	**51**	**1.30**	**76**	**1.95**	**52**	**2.60**
as PO_4-P	**10**	**0.33**	**15**	**0.50**	**20**	**0.66**	**30**	**0.98**	**40**	**1.30**
NH_4Cl	**21**	**0.39**	**31**	**0.59**	**42**	**0.78**	**63**	**1.17**	**84**	**1.56**
as NH_4-N	**5**		**8**		**11**		**16**		**22**	

§as described in *Standard Methods* (1992)

Table 3.2 Contents of the Feed for System **II**

	Concentrations	
	[mg/L]	[mmol/L]
COD (theo.)	393	
CH_3COONa as CH_3COOH	273 200	3.33
Difco® Nutrient Broth[§]	180	-
$CaCl_2$ as Ca^{++}	104 38	0.94
$MgSO_4$ as Mg^{++}	63 13	0.52
$FeCl_3$ as Fe^{+++}	3.2 1.1	0.02
K_2HPO_4 as K^+ as PO_4-P	57 25 10	0.33
NH_4Cl as NH_4-N	21 5	0.39

[§]as described in *Standard Methods* (1992)

3.2.3 Sampling

For each different feed composition profiles of the pilot plant for acetate, phosphate, COD, Nitrate, and PHB were measured. In addition to that, MLSS, MLVSS, and SS_{eff} were usually measured daily to determine the amount of sludge to be wasted. The samples from the two systems were taken at the following points:

- At the end of the tube of the anoxic reactor; called *anaerob in.*
- At the effluent tube of the ananerobic reactor; called *anaerob out.*
- Near the effluent opening of the aerobic tank; called *aerob.*
- The clear water near the effluent opening of the clarifier; called *clarifier.*

Samples for feed and effluent were taken directly out of the respective carboys.

3.3 Setup of the Batch Tests

3.3.1 General

A glass mason jar was used for the batch tests. Air or nitrogen were used to keep the sludge mixed.

First, 3 L of sludge from the aerobic tank were filled into the jar. Next, the sludge was aerated for approximately 30 minutes to remove all remaining acetate from the liquid. After that, nitrogen was bubbled through the sludge for approximately one hour to achieve anaerobic conditions. Finally, the nutrients were added in one or two spikes.

To keep the time for processing the samples as short as possible, a vacuum filter was placed beside the mason jar and the samples were transferred with a pipette directly from the jar to the filter. Subsequently, phosphate, nitrate, nitrite, and acetate in the filtered sample were measured while the batch test was ongoing. These phosphate measurements were used to determine the appropriate moment for switching from anaerobic to aerobic conditions. The gas flow was switched from nitrogen to air when the phosphate concentrations leveled off. Furthermore, the phosphate measurements were used to judge the time for ending the experiment. After the batch test was finished, the remaining sludge was poured back into the system it was taken from.

3.3.2 Batch Tests System I

The conditions of the batch tests performed with the sludge from system I are listed in the tables 3.3 to 3.6. These batch tests were performed to analyze the kinetics of systems with different acetate loads. The same feed concentrations were used in these batch tests as were used to operate system I at the time of the batch test. The first batch test (Ia) was performed with mixed sludge from both systems, collected before the two systems were operated separately.

3.3.3 Batch Tests System II with increasing acetate concentrations

Tables 3.7 to 3.10 list the batch tests performed with sludge from system II. These batch tests were performed to analyze the behavior of the sludge under higher acetate concentrations than those the system was brought to steady state with. For that purpose, one spike of nutrients with acetate concentrations differing from 200 mg/L to 600 mg/L were added.

3.3.4 Batch Tests System II with varying amounts of nutrients

Tables 3.11 to 3.15 list the batch tests performed with sludge from system II. The purpose of these batch tests was to determine the influence of some of the nutrients on the biological phosphorus removal process. These batch tests used one or two spikes of nutrients with a total of 600 mg/L acetate as acetic acid added and differing nutrient concentrations.

3.3.5 Precipitation Tests

Tables 3.16 and 3.17 list the batch tests performed with the nutrients only and without any sludge. The purpose of these batch tests was to determine the amount of phosphate precipitated in the nutrients by the cations. The first batch test used the maximal calcium concentration added during the other experiments at the highest pH value encountered during a batch test. The second batch test used the usual calcium concentration used for the feed of the two pilot plants at the highest pH value encountered in the aerobic reactors of the two systems.

Table 3.3 Parameters for Batch Test Ia

Name of batch test		**Ia**
Date		06/12/94
Length of anaerobic phase	[h : m]	1:30
Length of aerobic phase	[h : m]	2:45

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**273**	**3.33**
as CH_3COOH	**200**	
COD (theo.)	**393**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**57**	**0.33**
as K^+	**25**	**0.65**
as PO_4-P	**10**	**0.33**
NH_4Cl	**21**	**0.39**
as NH_4-N	**5**	

Table 3.4 Parameters for Batch Test Ib

Name of batch test		**Ib**
Date		06/30/94
Length of anaerobic phase	[h : m]	1:31
Length of aerobic phase	[h : m]	2:59

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**410**	**5**
as CH_3COOH	**300**	
COD (theo.)	**500**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**86**	**0.50**
as K^+	**38**	**0.98**
as PO_4-P	**15**	**0.50**
NH_4Cl	**31**	**0.59**
as NH_4-N	**8**	

Table 3.5 Parameters for Batch Test Ic

Name of batch test		Ic
Date		07/20/94
Length of anaerobic phase	[h : m]	1:31
Length of aerobic phase	[h : m]	3:29

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**546**	**6.66**
as CH_3COOH	**400**	
COD (theo.)	**606**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**115**	**0.66**
as K^+	**51**	**1.30**
as PO_4-P	**20**	**0.66**
NH_4Cl	**42**	**0.78**
as NH_4-N	**11**	

Table 3.6 Parameters for Batch Test Id

Name of batch test		**Id**
Date		08/03/94
Length of anaerobic phase	[h : m]	1:31
Length of aerobic phase	[h : m]	4:03
Nutrients added		
	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**820**	**9.99**
as CH_3COOH	**600**	
COD (theo.)	**820**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**171**	**0.98**
as K^+	**76**	**1.95**
as PO_4-P	**30**	**0.98**
NH_4Cl	**63**	**1.17**
as NH_4-N	**16**	

Table 3.7 Parameters for Batch Test II.1a

Name of batch test		**II.1a**
Date		06/24/94
Length of anaerobic phase	[h : m]	1:32
Length of aerobic phase	[h : m]	5:28

Nutrients added

	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**410**	**5**
as CH_3COOH	**300**	
COD (theo.)	**500**	
Difco® Nutrient Broth	**180**	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**85**	**0.49**
as K^+	**38**	**0.98**
as PO_4-P	**15**	**0.49**
NH_4Cl	**31**	**0.58**
as NH_4-N	**8**	

Table 3.8 Parameters for Batch Test II.1b

Name of batch test		**II**.1b
Date		06/19/94
Length of anaerobic phase	[h : m]	1:30
Length of aerobic phase	[h : m]	5:30

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**547**	**6.67**
as CH_3COOH	**400**	
COD (theo.)	**606**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**85**	**0.49**
as K^+	**51**	**1.30**
as PO_4-P	**15**	**0.49**
NH_4Cl	**31**	**0.58**
as NH_4-N	**8**	

Table 3.9 Parameters for Batch Test II.1c

Name of batch test		**II**.1c
Date		07/11/94
Length of anaerobic phase	[h : m]	2:02
Length of aerobic phase	[h : m]	5:58

Nutrients added

	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**676**	**8.24**
as CH_3COOH	**500**	
COD (theo.)	**713**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**113**	**0.65**
as K^+	**51**	**1.30**
as PO_4-P	**20**	**0.65**
NH_4Cl	**42**	**0.78**
as NH_4-N	**11**	

Table 3.10 Parameters for Batch Test II.1d

Name of batch test		**II**.1d
Date		07/07/94
Length of anaerobic phase	[h]	2:02
Length of aerobic phase	[h]	5:28

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	**820**	**9.99**
as CH_3COOH	**600**	
COD (theo.)	**820**	
Difco® Nutrient Broth	180	
$CaCl_2$	104	0.94
as Ca^{++}	38	
$MgSO_4$	63	0.52
as Mg^{++}	13	
$FeCl_3$	3.2	0.02
as Fe^{+++}	1.1	
K_2HPO_4	**171**	**0.98**
as K^+	**76**	**1.95**
as PO_4-P	**30**	**0.98**
NH_4Cl	**63**	**1.17**
as NH_4-N	**16**	

Table 3.11 Parameters for Batch Test II.2a

Name of batch test		**II**.2a		
Date		07/22/94		
Length of anaerobic phase	[h : m]	1:32		
Length of aerobic phase	[h : m]	6:58		
Addition 2nd Spike	[h : m]	1:32		
Nutrients added		1st Spike		2nd Spike
		Concentrations		
		[mg/L]	[mmol/L]	[mg/L]	[mmol/L]
CH_3COONa as CH_3COOH		410 300	5	410 300	5
COD (theo.)		500		500	
Difco® Nutrient Broth		180		180	
$CaCl_2$ as Ca^{++}		104 38	0.94	104 38	0.94
$MgSO_4$ as Mg^{++}		63 13	0.52	63 13	0.52
$FeCl_3$ as Fe^{+++}		3.2 1.1	0.02	3.2 1.1	0.02
K_2HPO_4 as K^+ as PO_4-P		85 38 15	0.49 0.98 0.49	85 38 15	0.49 0.98 0.49
NH_4Cl as NH_4-N		31 8	0.58	31 8	0.58

Table 3.12 Parameters for Batch Test II.2b

Name of batch test		II.2b
Date		07/26/94
Length of anaerobic phase	[h : m]	2:02
Length of aerobic phase	[h : m]	5:28

Nutrients added	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa	820	9.99
as CH_3COOH	600	
COD (theo.)	1180	
Difco® Nutrient Broth	540	
$CaCl_2$	313	2.82
as Ca^{++}	113	
$MgSO_4$	188	1.56
as Mg^{++}	38	
$FeCl_3$	11.4	0.07
as Fe^{+++}	3.9	
K_2HPO_4	171	0.98
as K^+	76	1.95
as PO_4-P	30	0.98
NH_4Cl	63	1.17
as NH_4-N	16	

Table 3.13 Parameters for Batch Test II.2c

Name of batch test		**II**.2c
Date		8/5/94
Length of anaerobic phase	[h : m]	2:02
Length of aerobic phase	[h : m]	6:29
Addition 2nd Spike	[h : m]	3:47

Nutrients added	1st Spike		2nd Spike	
	Concentrations			
	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]
CH_3COONa	820	9.99	0	0
as CH_3COOH	600		0	
COD (theo.)	820		360	
Difco® Nutrient Broth	180		360	
$CaCl_2$	104	0.94	209	1.88
as Ca^{++}	38		75	
$MgSO_4$	63	0.52	125	1.04
as Mg^{++}	13		25	
$FeCl_3$	3.2	0.02	8.1	0.05
as Fe^{+++}	1.1		2.8	
K_2HPO_4	171	0.98	0	0.00
as K^+	76	1.95	0	0.00
as PO_4-P	30	0.98	0	0.00
NH_4Cl	63	1.17	0	0
as NH_4-N	16		0	

Table 3.14 Parameters for Batch Test II.2d

Name of batch test		**II**.2d
Date		8/9/94
Length of anaerobic phase	[h : m]	2:01
Length of aerobic phase	[h : m]	5:44
Addition 2nd Spike	[h : m]	4:02

Nutrients added	1st Spike		2nd Spike	
	Concentrations			
	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]
CH_3COONa	820	9.99	0	0
as CH_3COOH	600		0	
COD (theo.)	820		360	
Difco® Nutrient Broth	180		360	
$CaCl_2$	104	0.94	0	0
as Ca^{++}	38		0	
$MgSO_4$	63	0.52	0	0
as Mg^{++}	13		0	
$FeCl_3$	3.2	0.02	0.0	0
as Fe^{+++}	1.1		0.0	
K_2HPO_4	171	0.98	0	0.00
as K^+	76	1.95	0	0.00
as PO_4-P	30	0.98	0	0.00
NH_4Cl	63	1.17	0	0
as NH_4-N	16		0	

Table 3.15 Parameters for Batch Test II.2e

Name of batch test		**II**.2e
Date		8/12/94
Length of anaerobic phase	[h : m]	2:02
Length of aerobic phase	[h : m]	6:43
Addition 2nd Spike	[h : m]	4:02

Nutrients added	1st Spike		2nd Spike	
	Concentrations			
	[mg/L]	[mmol/L]	[mg/L]	[mmol/L]
CH_3COONa	820	9.99	0	0
as CH_3COOH	600		0	
COD (theo.)	820			
Difco® Nutrient Broth	180		0	
$CaCl_2$	104	0.94	156	1.41
as Ca^{++}	38		57	
$MgSO_4$	63	0.52	0	0
as Mg^{++}	13		0	
$FeCl_3$	3.2	0.02	0.0	0
as Fe^{+++}	1.1		0.0	
K_2HPO_4	171	0.98	0	0.00
as K^+	76	1.95	0	0.00
as PO_4-P	30	0.98	0	0.00
NH_4Cl	63	1.17	0	0
as NH_4-N	16		0	

Table 3.16 Parameters for Batch Test P1

Name of batch test		P1
Date		9/6/94
pH	**[-]**	**8.8**
Nutrients added		
	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa as CH_3COOH	820 600	9.99
COD (theo.)	820	
Difco® Nutrient Broth	180	
$CaCl_2$ **as Ca^{++}**	**313** **113**	**2.82**
$MgSO_4$ as Mg^{++}	63 13	0.52
$FeCl_3$ as Fe^{+++}	3.2 1.1	0.02
K_2HPO_4 as K^+ as PO_4-P	1140 507 200	6.5 13 6.5
NH_4Cl as NH_4-N	63 16	1.17

Table 3.17 Parameters for Batch Test P2

Name of batch test		P2
Date		9/8/94
pH	**[-]**	**8.3**
Nutrients added		
	Concentrations	
	[mg/L]	[mmol/L]
CH_3COONa as CH_3COOH	820 600	9.99
COD (theo.)	820	
Difco® Nutrient Broth	180	
$CaCl_2$ **as Ca^{++}**	**104** **38**	**0.94**
$MgSO_4$ as Mg^{++}	63 13	0.52
$FeCl_3$ as Fe^{+++}	3.2 1.1	0.02
K_2HPO_4 as K^+ as PO_4-P	1140 507 200	6.5 13 6.5
NH_4Cl as NH_4-N	63 16	1.17

3.4 Analytical Methods

3.4.1 Acetate

The filtered acetate samples were acidified by adding 1% acid by volume and subsequently were injected into a gas chromatograph. The acid used was changed over time. At first, acetate-free formic acid was used. After the supply of that acid ran out, phosphoric acid was used. Since this resulted not only in a non-linear response of the chromatograph over the concentration range of approximately 100 to 300 mg/L, but also in a change of the chromatograph response for the blank sample, this practice was abandoned. Instead, acetate contaminated formic acid was used and greatest care was taken to keep the amount of acid added as constant as possible. This made it feasible to correct the data for the acetic acid content of the formic acid.

Operation parameters of the gas chromatograph:

- Column: Packed column 0.3% Carbowax®; 0.1 % H_3PO_4; 60/80 Carbopack Column
- Oven temperature: 120°C
- Injection method: Hot needle, on column, 1 µL
- Injector temperature: 200°C
- Detector: Flame ionization detector
- Detector temperature: 200°C

3.4.2 Poly-ß-hydroxybutyric acid

The measurements for the PHB content of the sludge followed the method described by Hart (1994). The main differences were in the collection of the solids and the parameters of the gas chromatograph.

The relatively small size of the pilot plants made it necessary to reduce the amount of sludge lost due to sampling. Therefore, the dewatered solids from the sample preparation for the acetate and phosphate measurement were used. To collect the solids without contaminating them with pieces of the filters, the backside of loaded filters were wetted with distilled water. This caused the solids to separate from the filter and to drop into the sampling cup.

After the solids dried in the oven for one day at approximately 105°C, they were cooled down in a dessicator. Approximately 50 mg of the coarsely ground solids were measured into a pressure resistant 5 mL vial. In addition to that, two or three standards were prepared by measuring 5 to 15 mg NaPHB into vials. Afterwards, 2 mL of an extraction reagent and 2 mL of chloroform were added and the vial firmly capped. This extraction reagent was a mixture of 100 mL methanol, 3 mL concentrated sulfuric acid, and 50 mg benzoic acid as internal standard. It could be stored for approximately 2 weeks. A blank was prepared in the same way.

The filled vials were heated at approximately 105°C in an oven. After 3.5 hours, the vials were cooled to room temperature, 1 mL of distilled water was added into each vial and the vial was firmly recapped. Next, the vials were placed for 15 minutes on a shaking table. On the same day, a 1 µL sample of the bottom liquid layer was injected into a gas chromatograph.

Operation parameters of the gas chromatograph:

- Column: packed column; 2% Reoplex® 400
- Oven temperature: 130°C; constant
- Injection method: Hot needle, on column, 1μL
- Injector temperature: 160°C
- Detector: Flame ionization detector
- Detector temperature: 200°C

3.4.3 Standardized Analytical Methods

Methods that are described in *Standard Methods* (1992) are shown in Table 3.18.

Table 3.18 Standardized Analytical Methods

Parameter	Method # according to *Standard Methods* (1992)	Comments
Total Suspended Solids	2540 D	The sample volume was doubled by using filter pads. It was usually 50 mL for MLSS and 500 mL for SS_{eff}.
Volatile Solids	2540 E	
COD	similar to 5220 C	The digestion reagent contained less mercury than described in the *Standard Methods* (1992)
SVI	2710 D	
pH	4500-H^+ B	
total Phosphorus	4500-P B5 4500-P E	Digestion Method Colometric Method
Nitrate	4500-NO_2^- C	Dionex® AS4A Column
Nitrite	4500-NO_3^- C	Dionex® AS4A Column
Phosphate	4110	Dionex® AS4A Column

3.4.4 Miscellaneous

All glassware and all plasticware were washed in diluted hydrochloric acid.

4. Results

4.1 Results for continuous flow

In this section, the results for the continuous flow operation of System I are presented. In addition to that, the equivalent data for the System II are shown. To make the figures in this chapter easier to comprehend, Figure 4.1 shows the feed concentrations for System I normalized by the feed concentrations of System II.

4.1.1 Solids

Figure 4.2 shows the data for mixed liquor suspended solids (MLSS) for System I and System II. The vertical lines indicate an increase of the acetate concentration in the feed of System I. The numbers between the vertical lines show the nominal acetate concentration in mg/L as acetic acid during that time interval. The dip in the curve for System I after 6/23/94 shows the effects of a spill. The steep increase after each increase of feed was due to the intentional omittance of wasting sludge. This was done to reduce the time needed for the system to acclimate to the new conditions.

Figure 4.3 shows the percentage of volatile solids for System I and System II. Note, that the Y-axis has been spread to show the changes better. Nevertheless, for both systems similar ratios can be observed.

The data for the effluent suspended solids of the systems have been split between two figures, to make it possible to distinguish between the curves. Figure 4.4 shows the effluent suspended solids for System I. Note that one extreme value (268 mg/L due to spill) has been omitted. Figure 4.5 shows the effluent suspended solids for System II. Note that the values are usually slightly higher than those for System I; despite the fact that both systems were nearly identical.

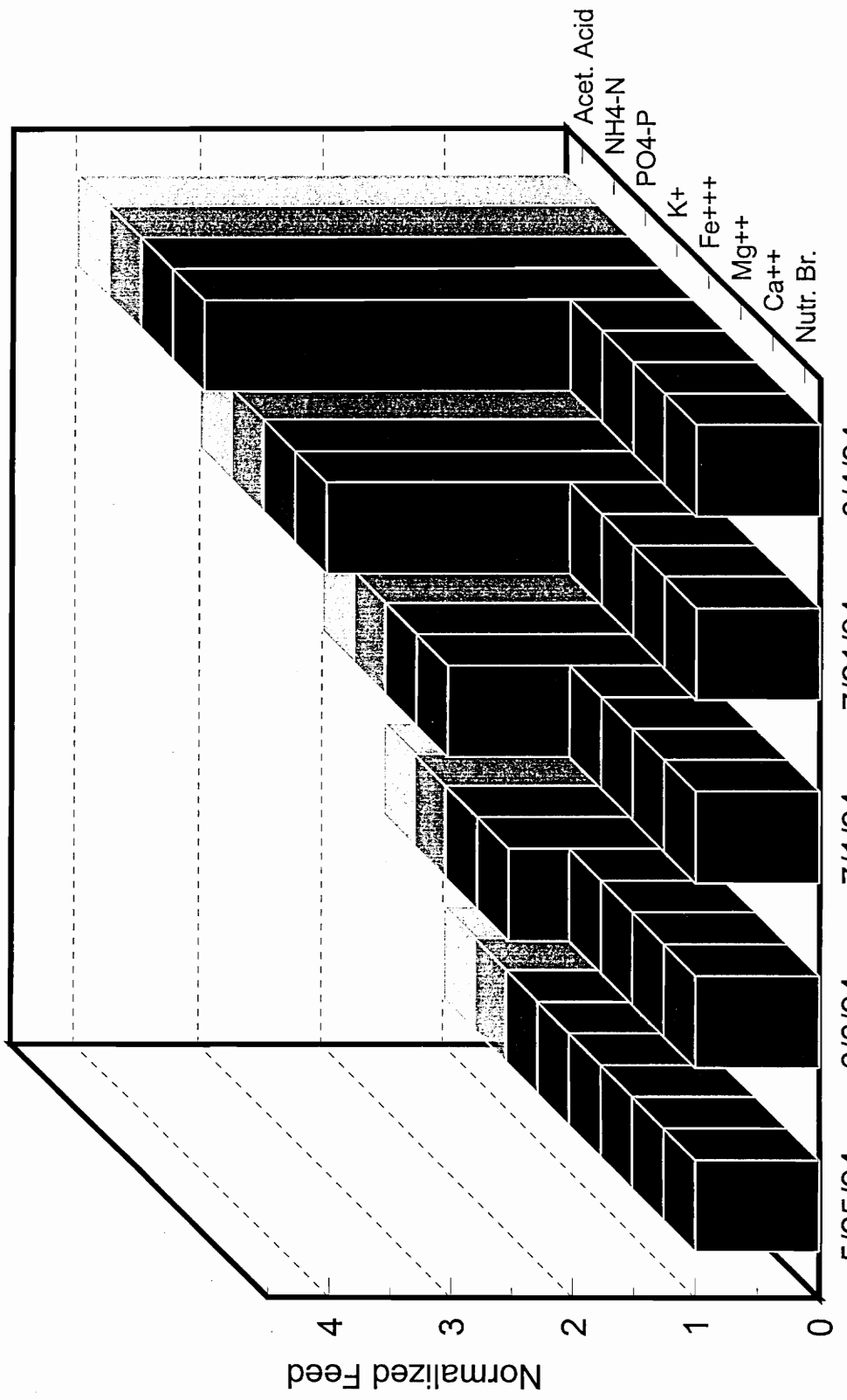

Figure 4.1 Feed for System I

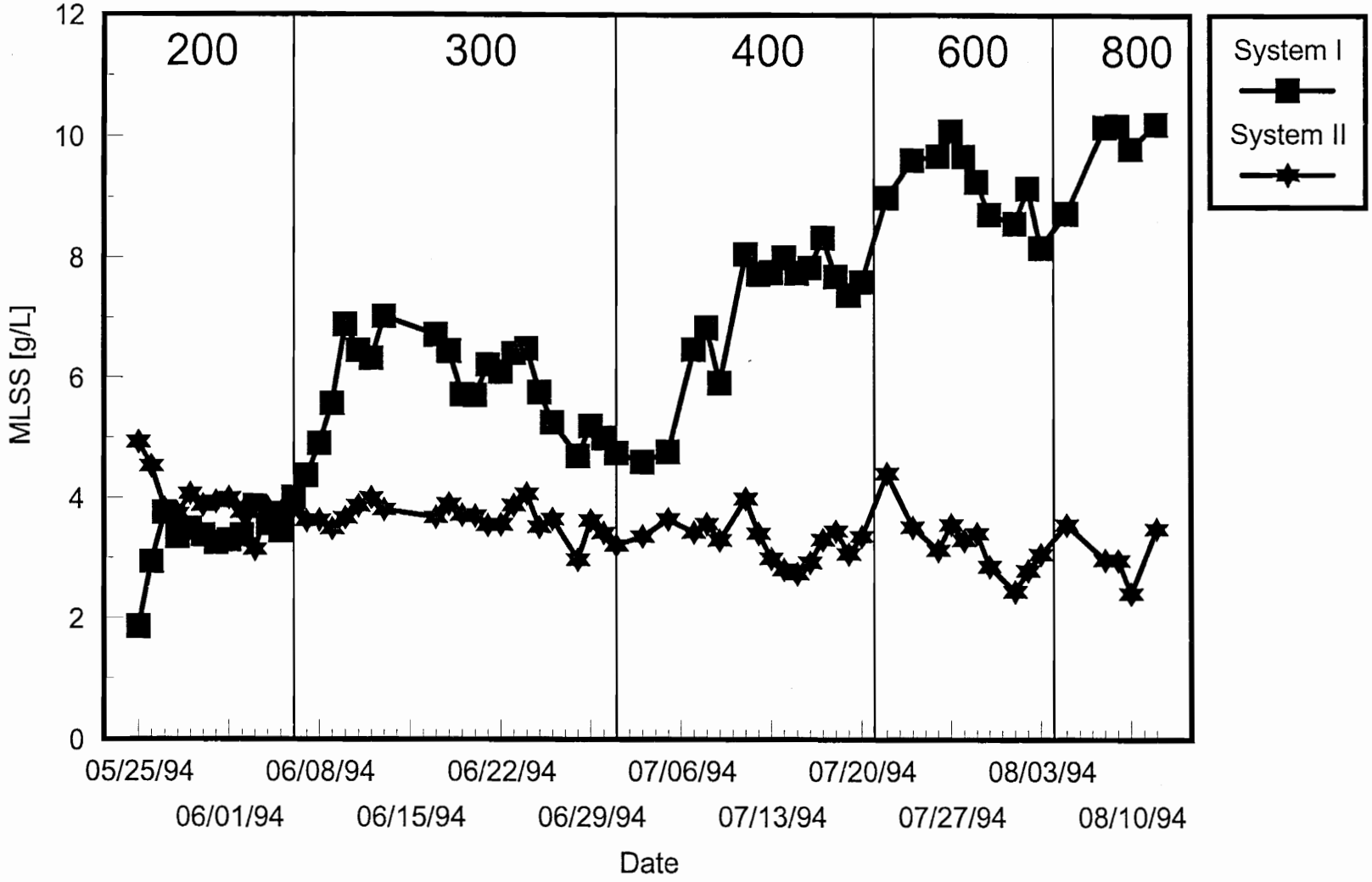

Figure 4.2 MLSS in Systems I and II versus Time

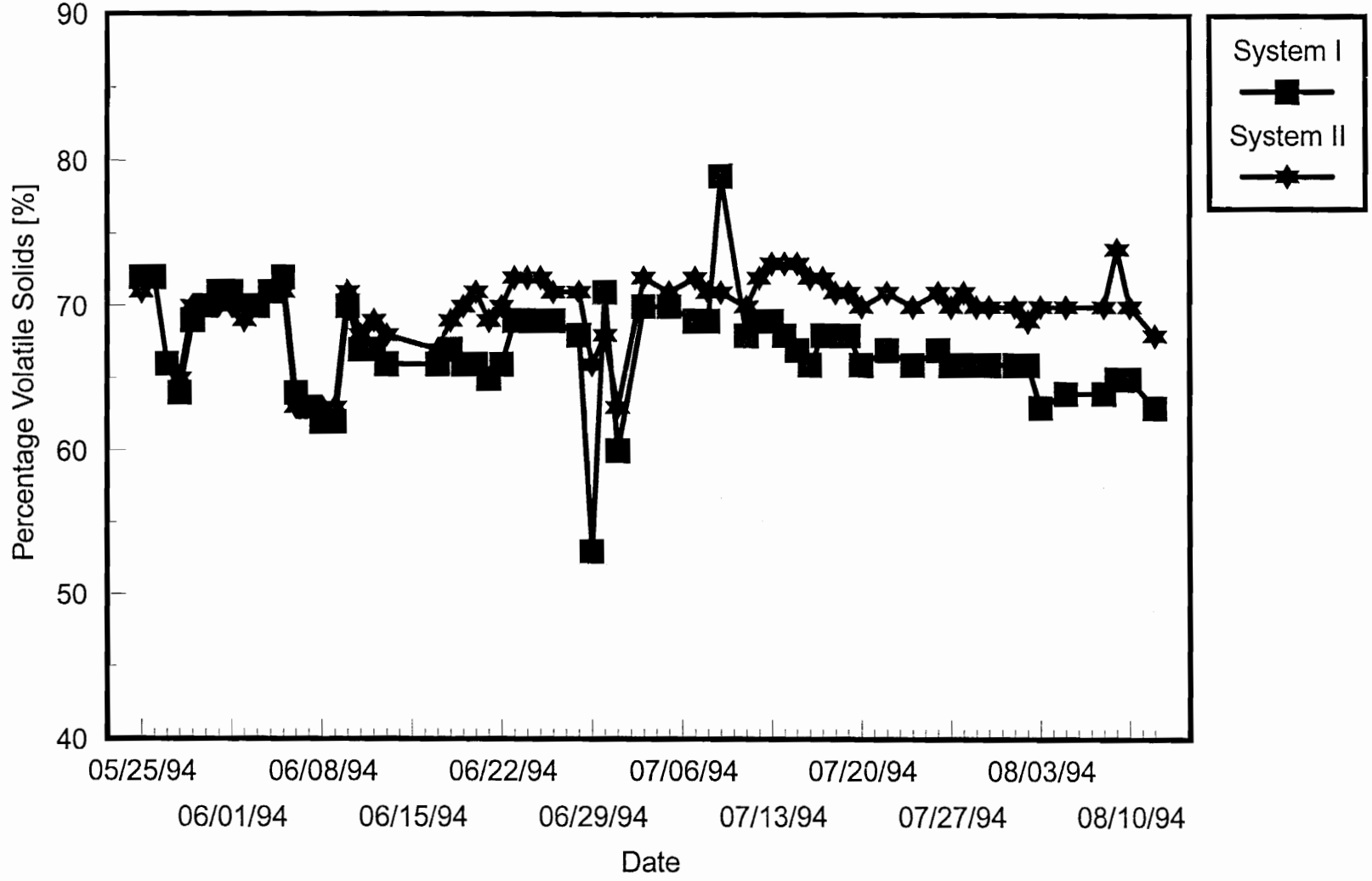

Figure 4.3 Percent VSS in Systems I and II versus Time

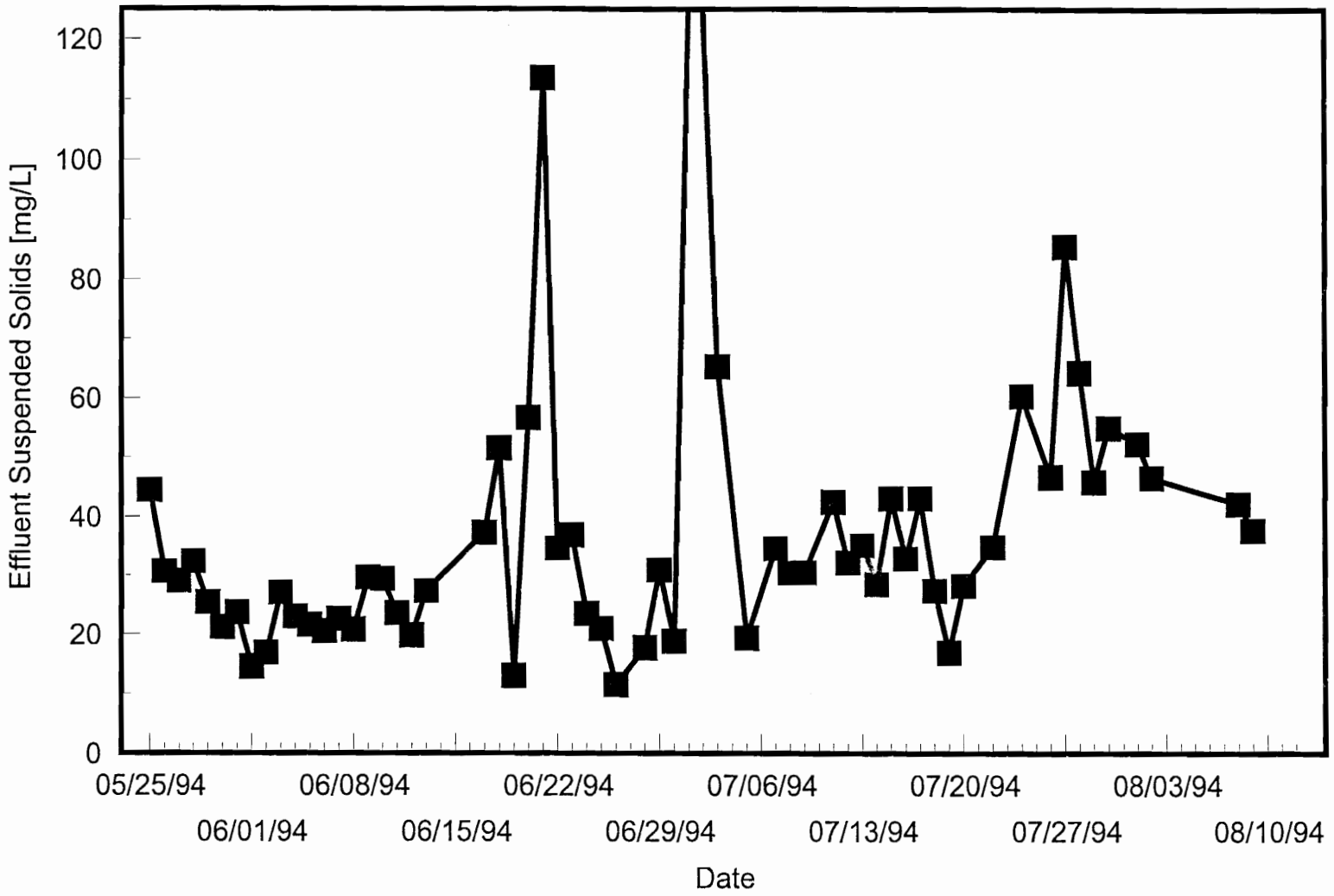

Figure 4.4 Effluent Suspended Solids from System I versus Time

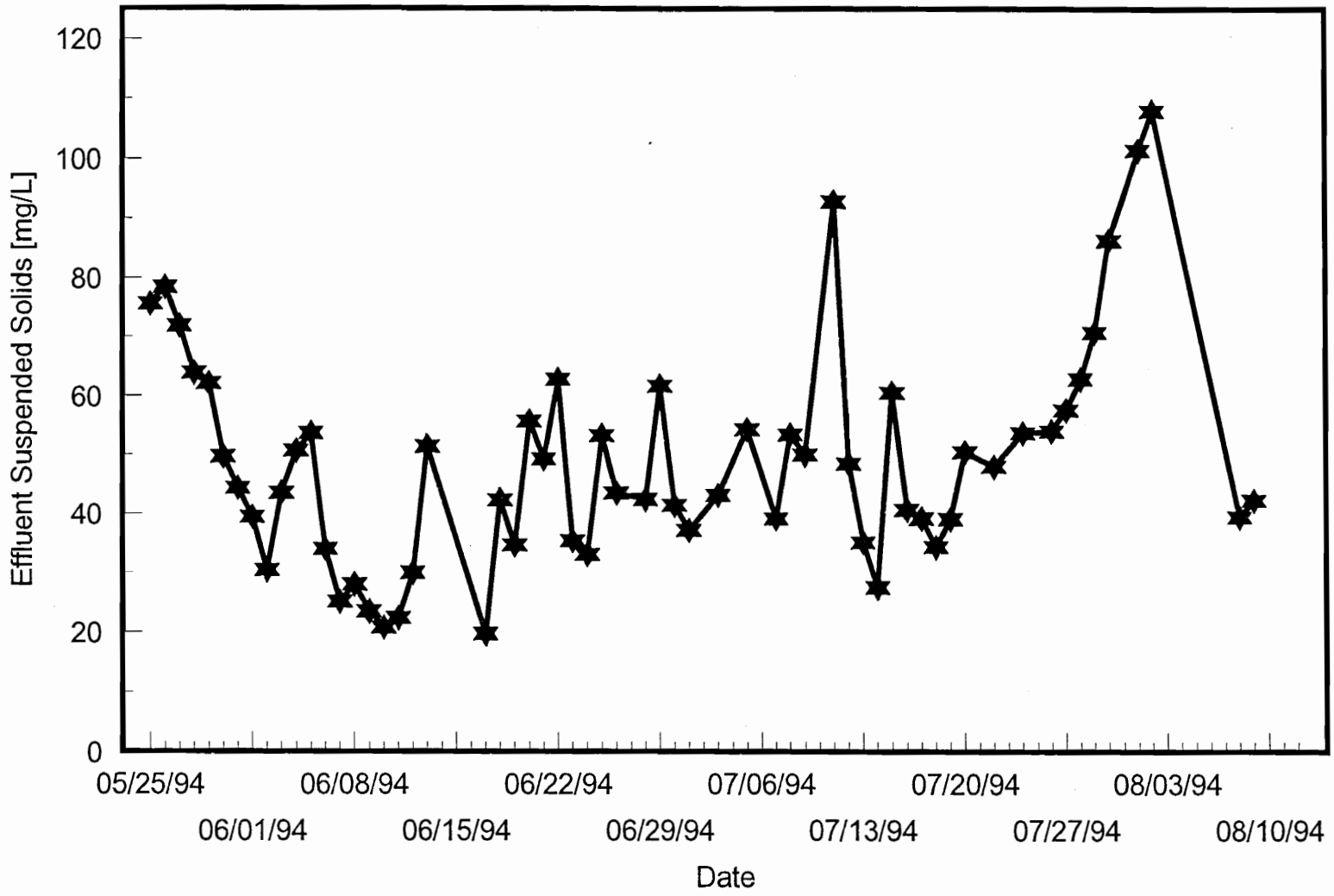

Figure 4.5 Effluent Suspended Solids from System II versus Time

4.1.2 Profiles of the systems

Figures 4.6 to 4.10 show the profiles of PO_4-P, NO_3-N, COD, Acetate and PHB for System I for different acetate loading, and the profiles of the same parameters for System II for the time when both systems were operated with the same acetate feed of 200 mg/L as acetic acid. Whenever possible, the average of several profiles measured under the same feed conditions were used.

In the phosphorus profile in Figure 4.6, note that the effluent phosphorus values were between 1.3 mg/L PO_4-P and 0.1 mg/L PO_4-P, and therefore too small to be displayed properly. Furthermore, the maximal value for released phosphorus increased nearly proportional to the increased feed. The nitrogen profile in Figure 4.7 shows a decrease of the nitrate values for increasing feed acetate concentrations. No COD profile was taken for System I at a feed of 800 mg/L, as Figure 4.8 shows. Note the similar form of the curves and the small increases of the COD concentration with increased feed. In Figure 4.9 the acetate profiles are shown. Note, that the acetate value for the feed at nominal 300 mg/L acetate was only 180 mg/L. However, this is only a single value, since acetate measurements were not possible for the other profiles. A comparison with the COD (Figure 4.8) shows, that this value was too low and not representative. No acetate profiles were measured for either of the systems for the feed acetate concentration of 200 mg/L. Note the similar form of the curves and that practically all acetate had been taken up before the aerobic zone. Judging from Figure 4.9, the acetate uptake in the anoxic reactor was higher than the uptake in the anaerobic reactor. However, the sample had to be collected by letting the sludge drip from the tube of the anoxic reactor. Therefore, the sludge was given additional reaction time in the beaker, skewing the results.

Finally, the PHB profiles are displayed in Figure 4.10. The PHB values for the feed acetate concentration of 400 mg/L show a completely different curve form than the other curves. Furthermore, the PHB values PHB values for the feed acetate concentration of 300 mg/L is lower than that for the for the feed acetate concentration of 200 mg/L. This reflects probably more the influence of the measurement method than specific biological reactions. The relatively high concentrations of PHB in the aerobic reactor are noteworthy. All the profiles show similar to identical curves for systems I and II during the initial phase of 200 mg/L feed acetate concentration.

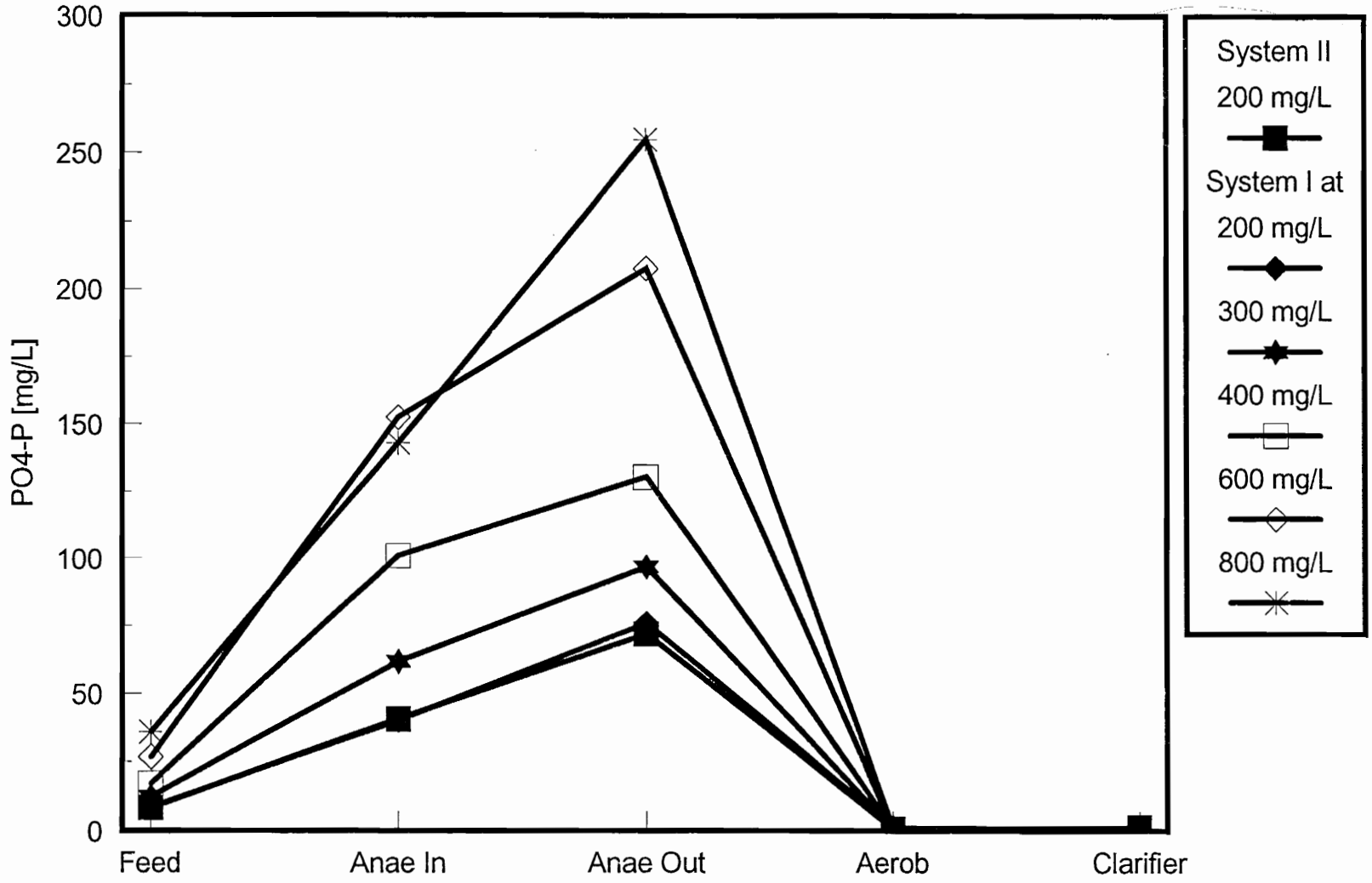

Figure 4.6 PO_4- P Profiles during Continuous Flow Experiments

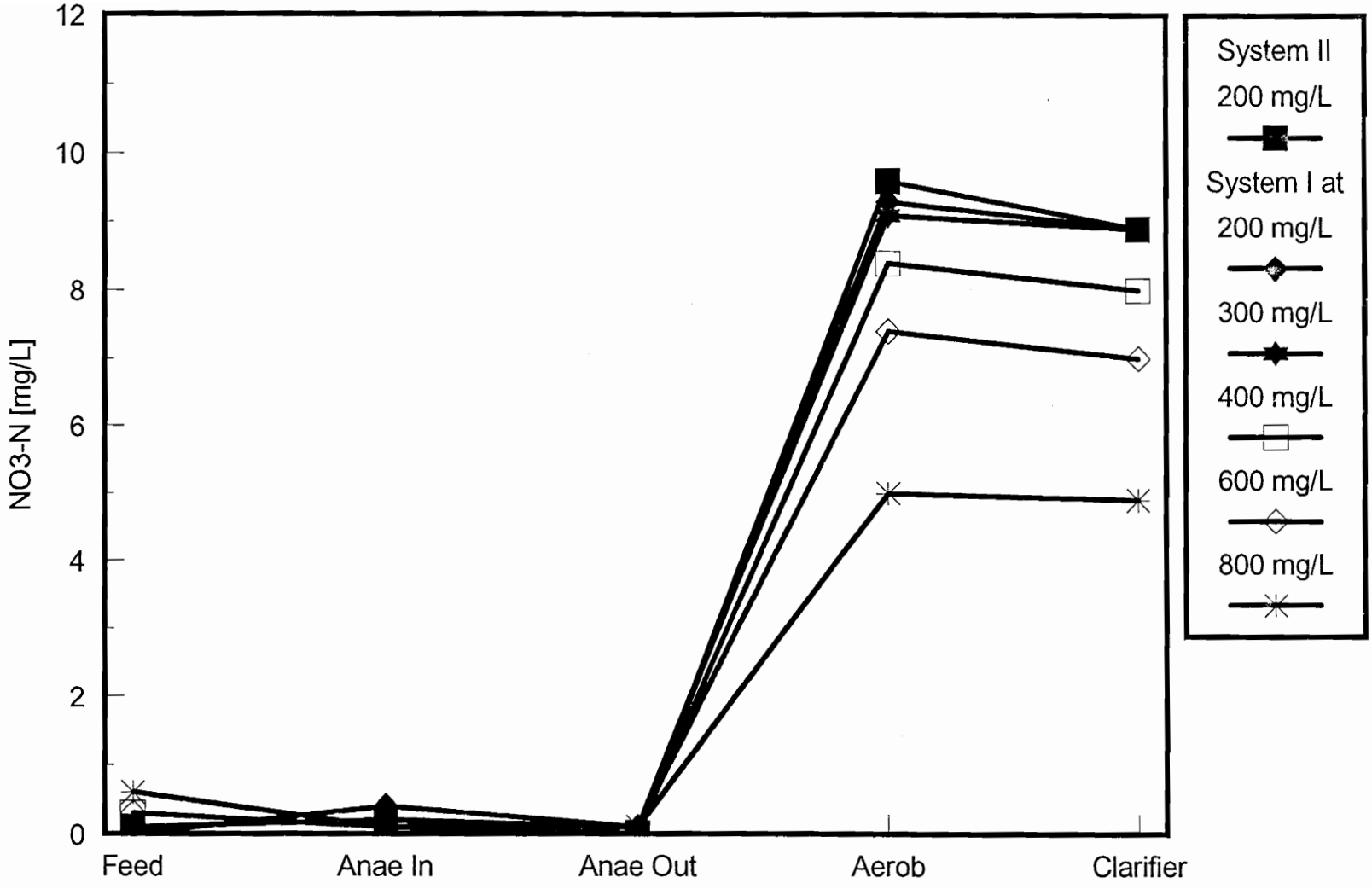

Figure 4.7 Nitrate Profiles during Continuous Flow Experiments

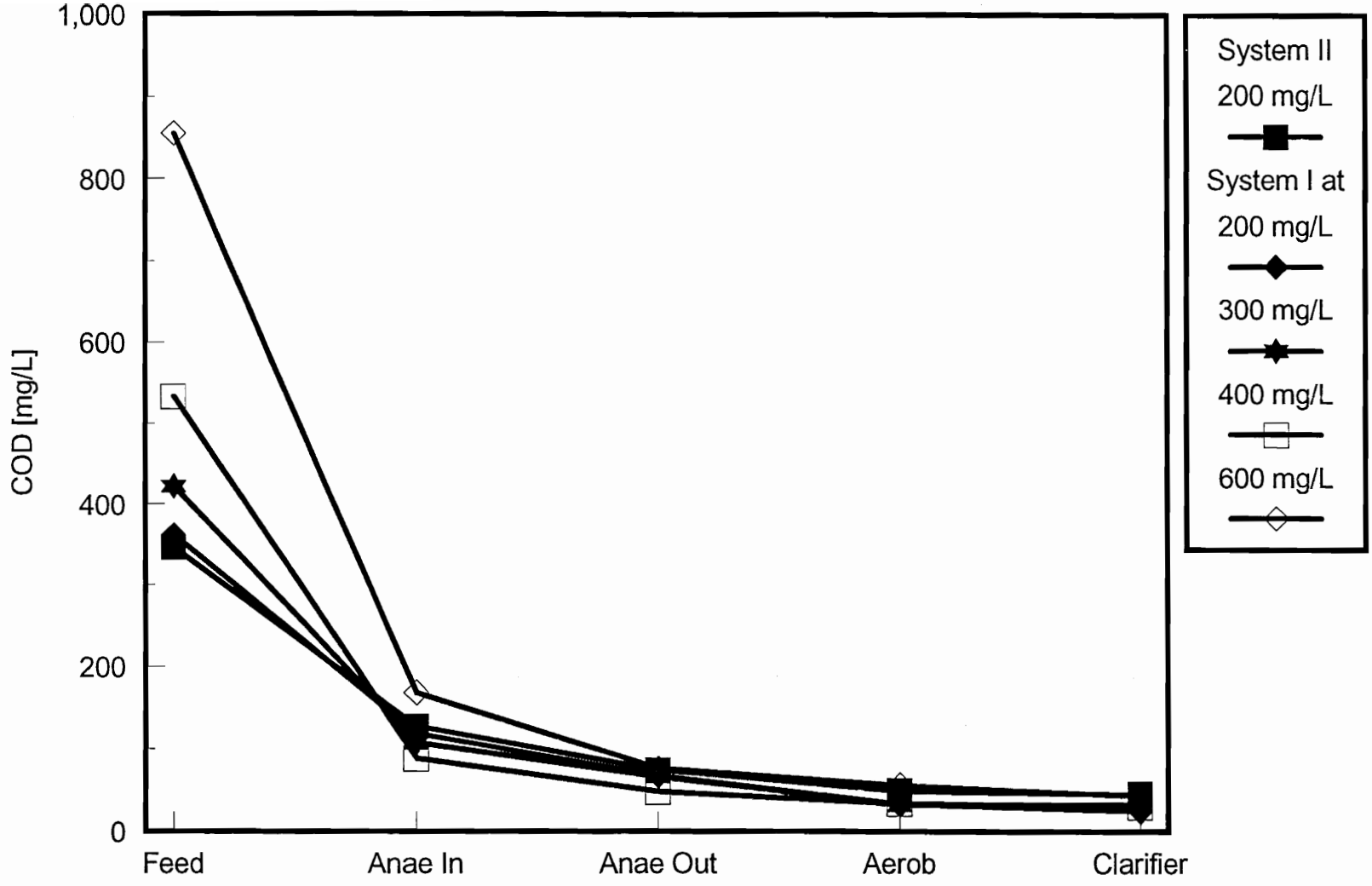

Figure 4.8 COD Profiles during Continuous Flow Experiments

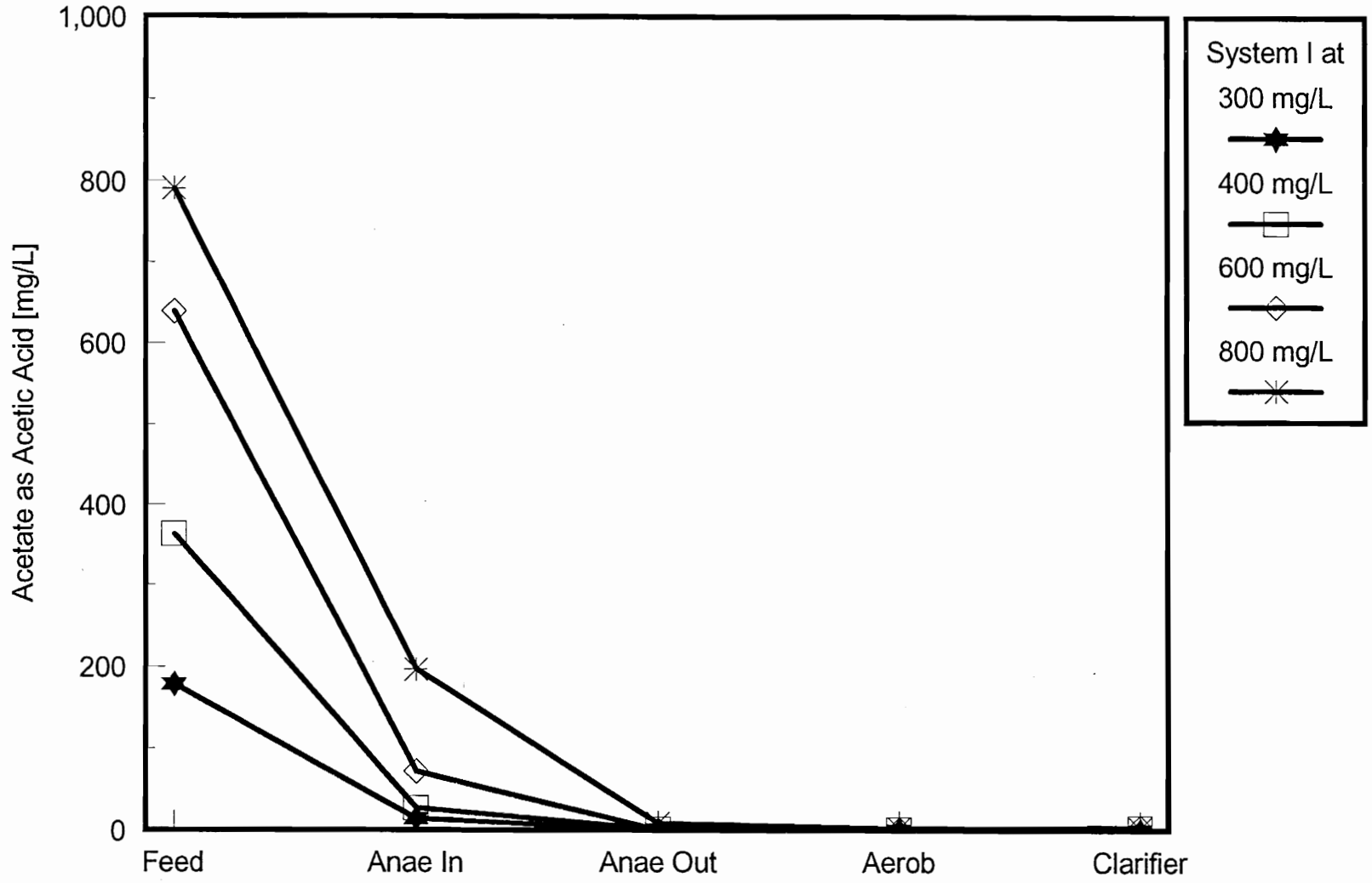

Figure 4.9 Acetate Profiles during Continuous Flow Experiments

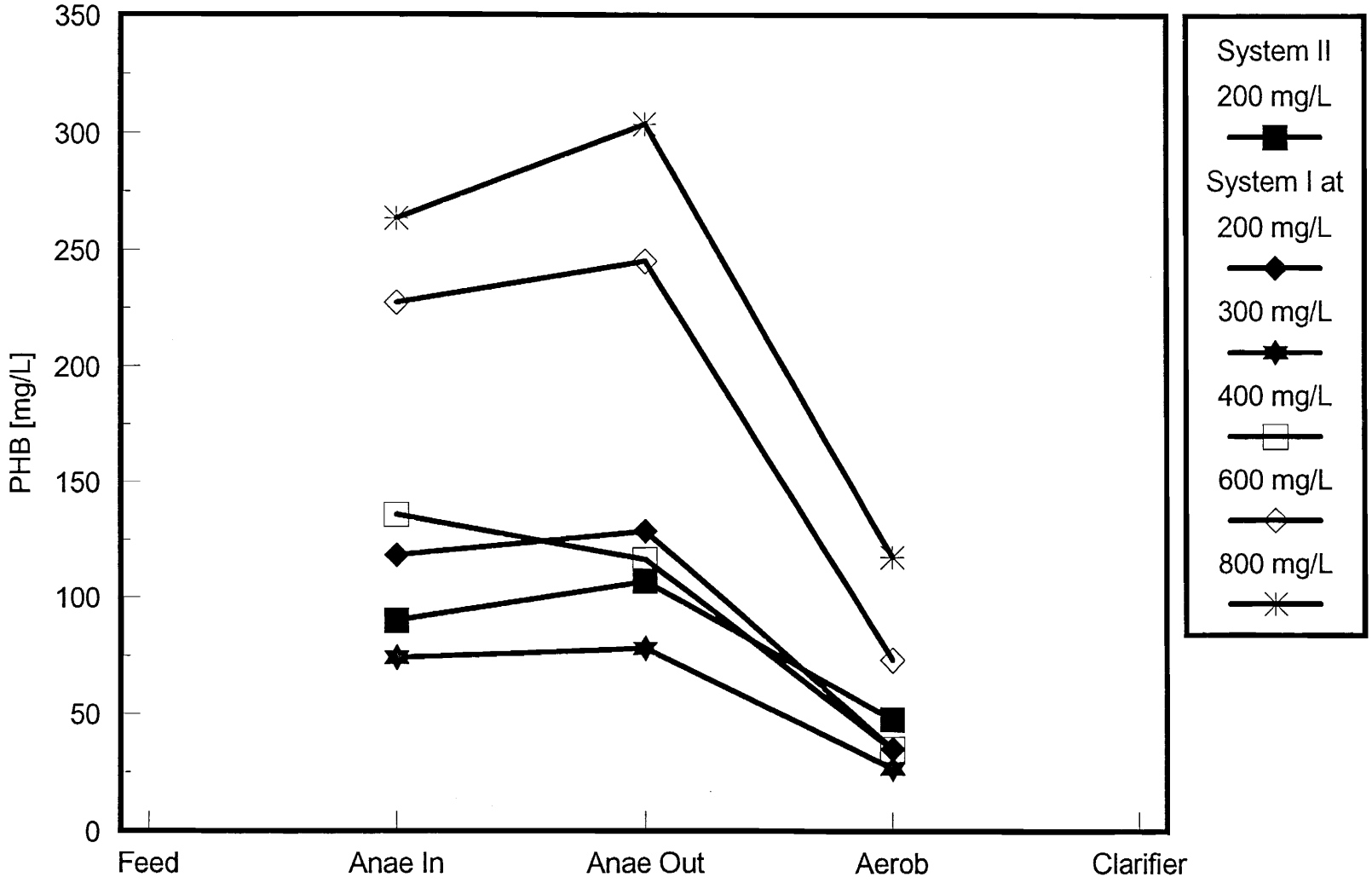

Figure 4.10 PHB Profiles during Continuous Flow Experiments

4.2 Results for batch tests with feed equal to the continuous flow feed

Figure 4.11 shows the results of four batch tests performed with the sludge of System I (see 3.3.2). The vertical line marks the start of aeration. The points show the actual raw data, while the curves represent best fits based on the assumption that the reaction followed first order kinetics. The curve fitting was done "by hand" (actually, by spreadsheet using "What/If" tables and trial & error), since the usual way of obtaining a best fit by plotting the data on a log basis allowed the data points in the asymptotic part of the curves to disproportionally influence the form of the curve. While no regression coefficients could be obtained due to the hand fitting, first order kinetics seem to describe the reactions well.

Figure 4.12 shows the results for the acetate concentrations during the batch tests. As in Figure 4.11, the vertical line marks the start of aeration. Due to the fact that only a few non-zero data points were measured, no curves were fit to the data. The rapid change of the acetate concentration would have allowed to use a large range of values for the rate constant with a near perfect fit of the regression curve.

Figure 4.13 shows the results for the PHB concentration in the biomass during the batch tests. In this case, no attempt was made to draw a curve through the raw date, since the data were too scattered. However, the PHB concentrations in the biomass seem to increase during phosphorus release and decrease during phosphorus uptake.

Figure 4.14 shows the results for the nitrate concentration during the batch tests. It shows denitrification during the anaerobic time frame and nearly linear nitrification during the aerobic time frame.

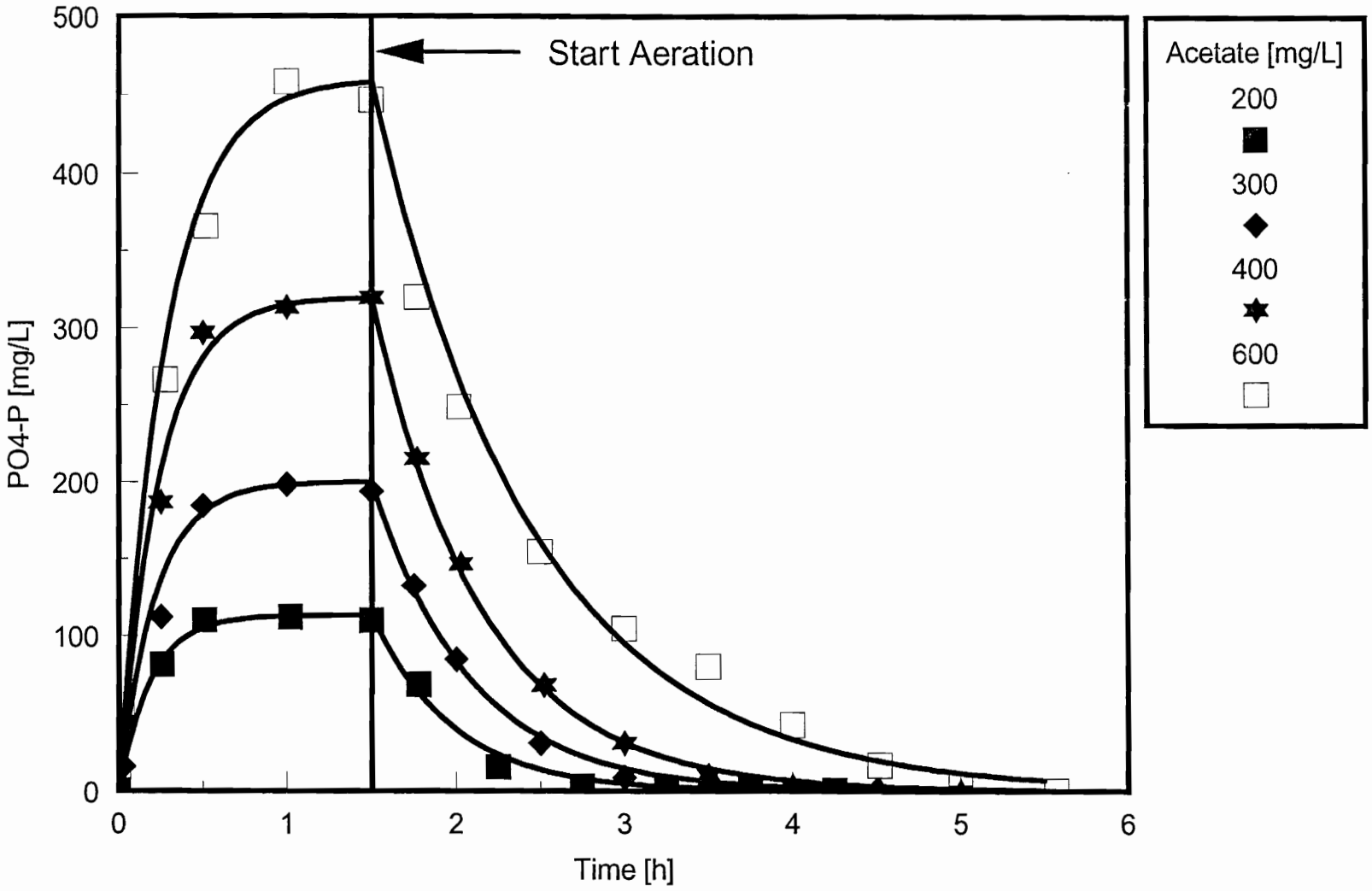

Figure 4.11 PO_4 - P Concentrations during Batch Tests of System I

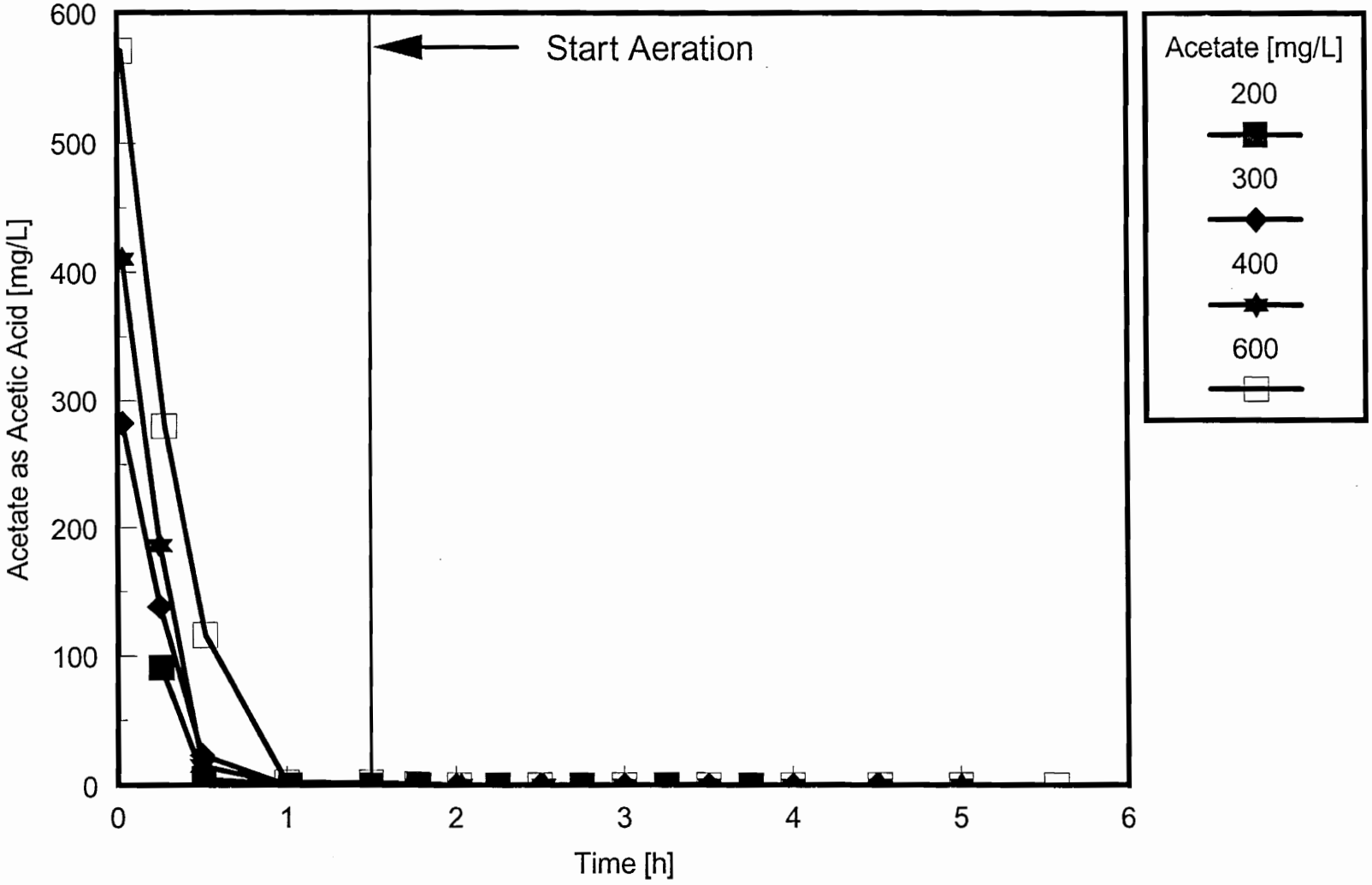

Figure 4.12 Acetate Concentrations during Batch Tests of System I

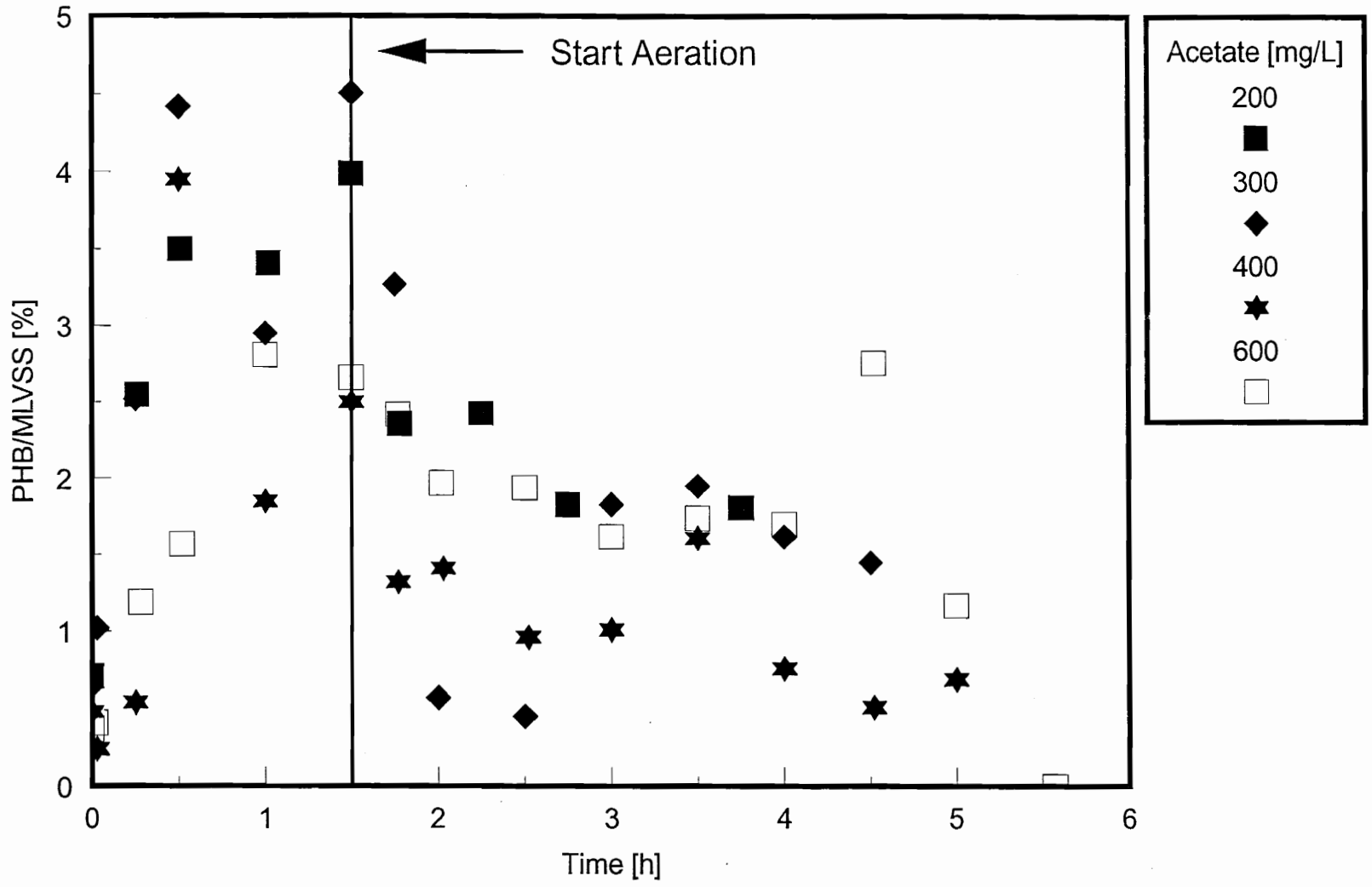

Figure 4.13 PHB Concentrations during Batch Tests of System I

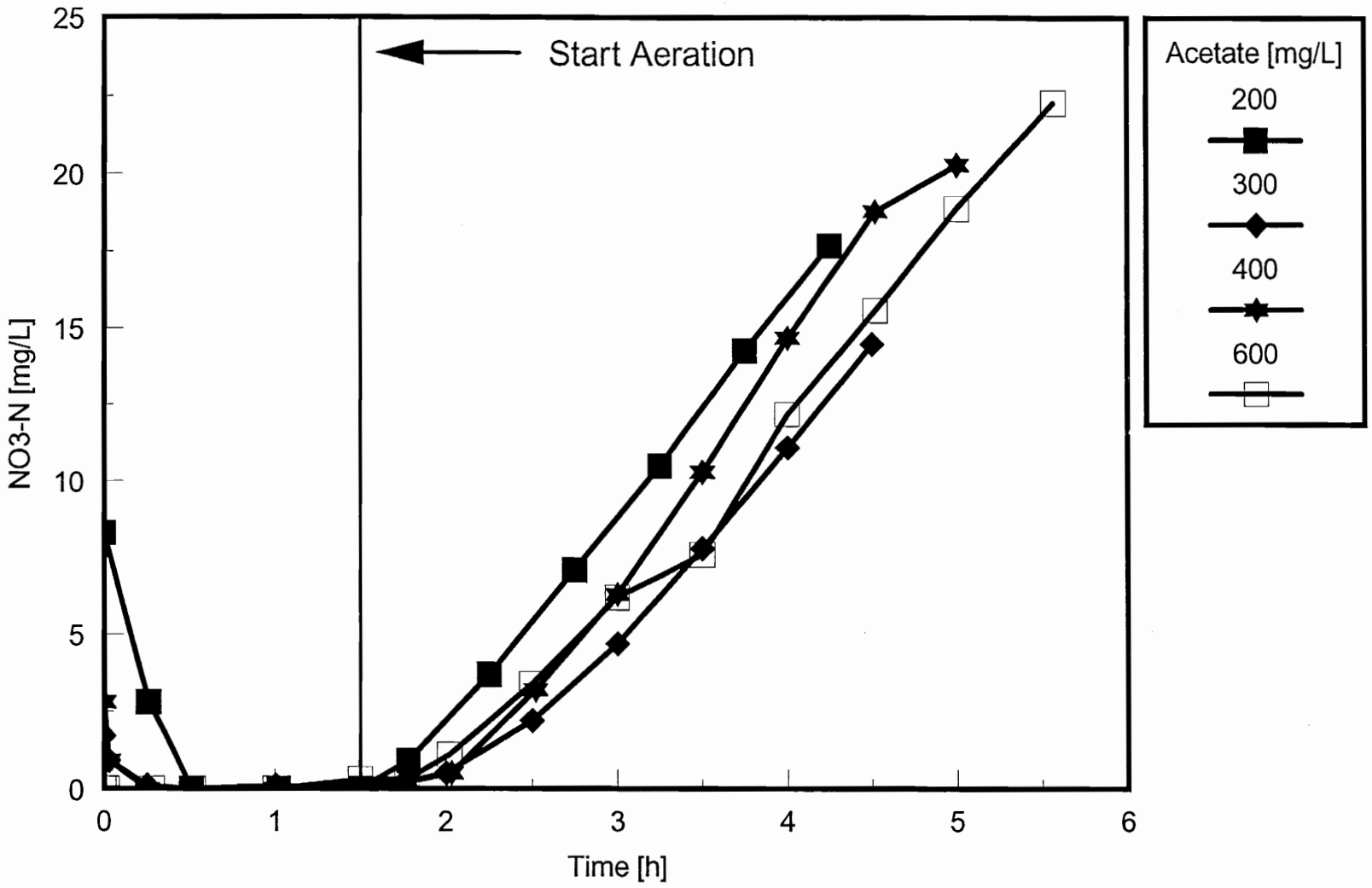

Figure 4.14 Nitrate Concentrations during Batch Tests of System I

4.3 Results for batch tests with increasing acetate concentrations

In this section, the results for the first series of batch tests with sludge from System II are shown. In that series the sludge was subjected to higher acetate concentrations than those the system was exposed to during steady state, as shown in Figure 4.15.

Figure 4.16 shows the phosphate results during the anaerobic phase of the batch tests. With the exception of the data for the experiment with 600 mg/L acetate, all data fit quite well the theoretical curves for first order kinetics. Figure 4.17 shows the results for the phosphate during the aerobic phase of the batch tests. The change in the form of the curves with increasing acetate concentration is noteworthy. No curve was fitted to the data of the experiment with 600 mg/L acetate.

The results for acetate are shown in Figure 4.18. The dotted line marks the beginning of aeration for the 200, 300, and 400 mg/L acetate experiments, and the solid line marks the start of aeration for the 500 and 600 mg/L acetate experiments. Note, that the curves for the acetate concentrations greater than 400 mg/L do not return to zero. The reason is most probably the use of phosphoric acid for the acetate measurements during those two batch tests (see chapter 3.4.1).

The data for PHB / MLVSS are split up into those for Figure 4.19 for acetate concentrations of 200 mg/L to 400 mg/L and Figure 4.20 for acetate concentrations of 500 mg/L and 600 mg/L. While the PHB concentration in the first three experiments has its maximum value during the anaerobic phase, the PHB concentration in the two other experiments has its maximum value during the aerobic phase.

The data for nitrate-N are shown in Figure 4.21. Observe, that in all experiments steady nitrification started with the beginning of aeration.

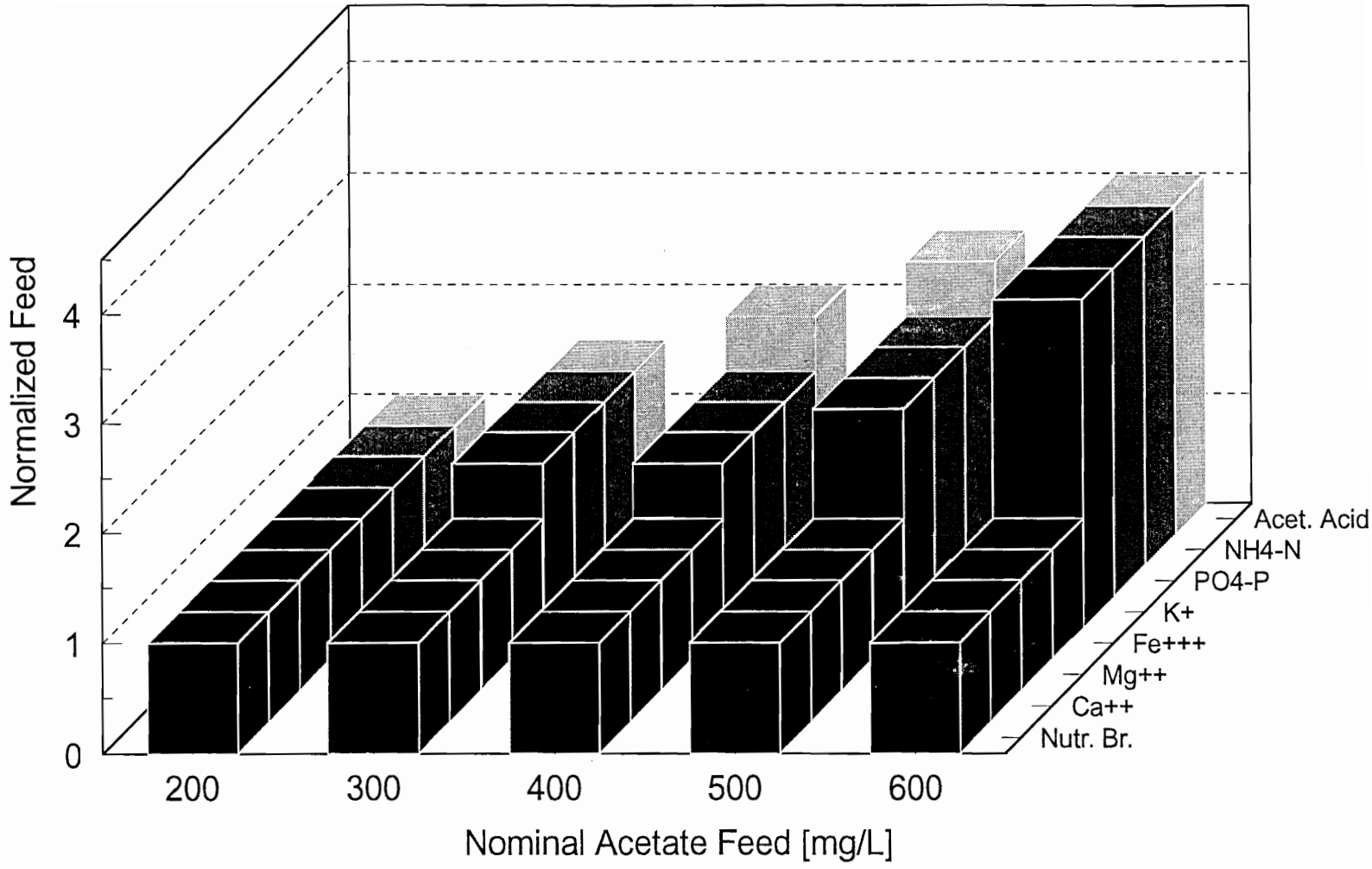

Figure 4.15 Feed for Batch Tests of System II at increasing Acetate Concentrations

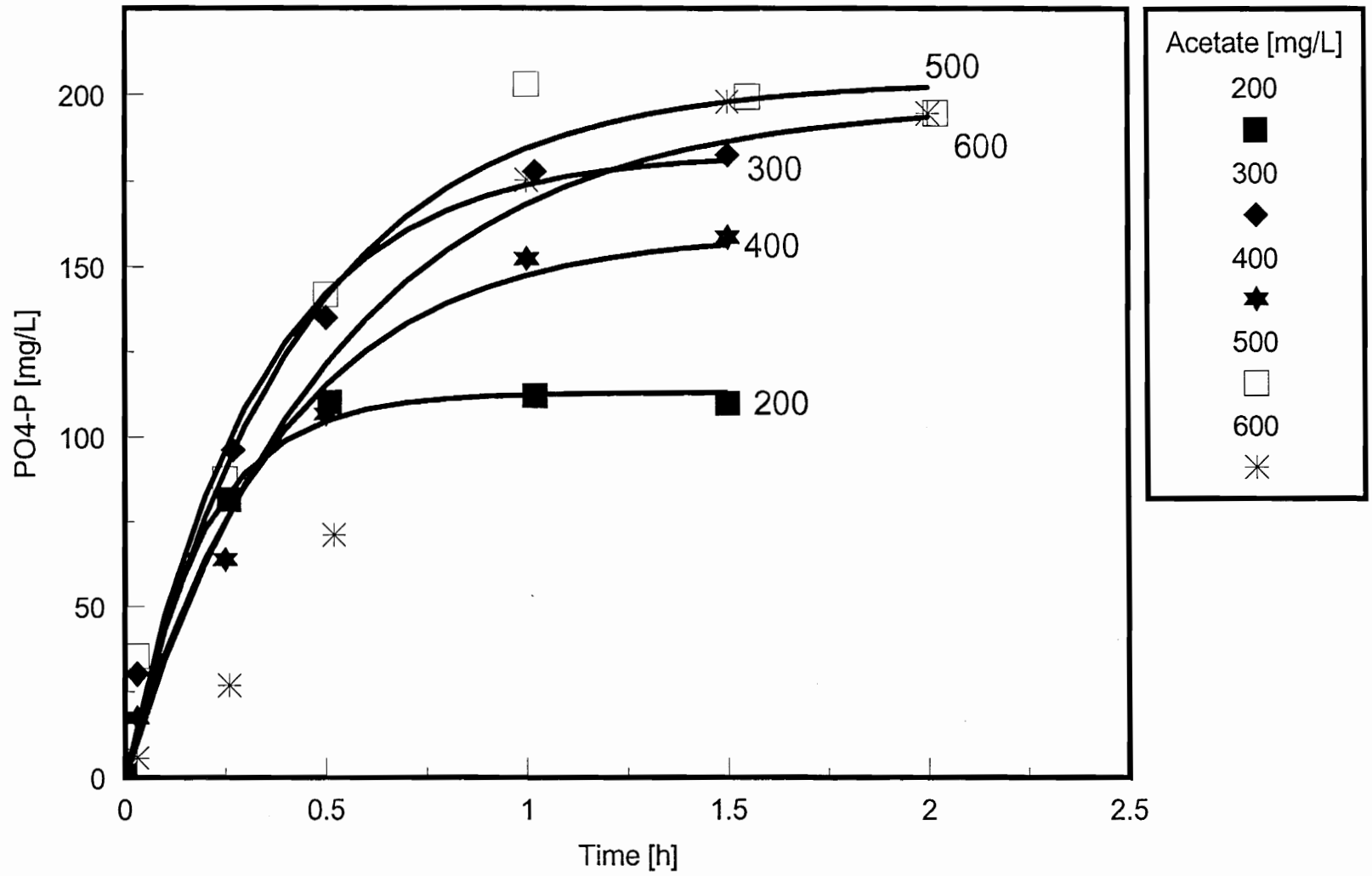

Figure 4.16 PO_4 - P Conc. during first Series of Batch Tests of System II; Anaerobic Time

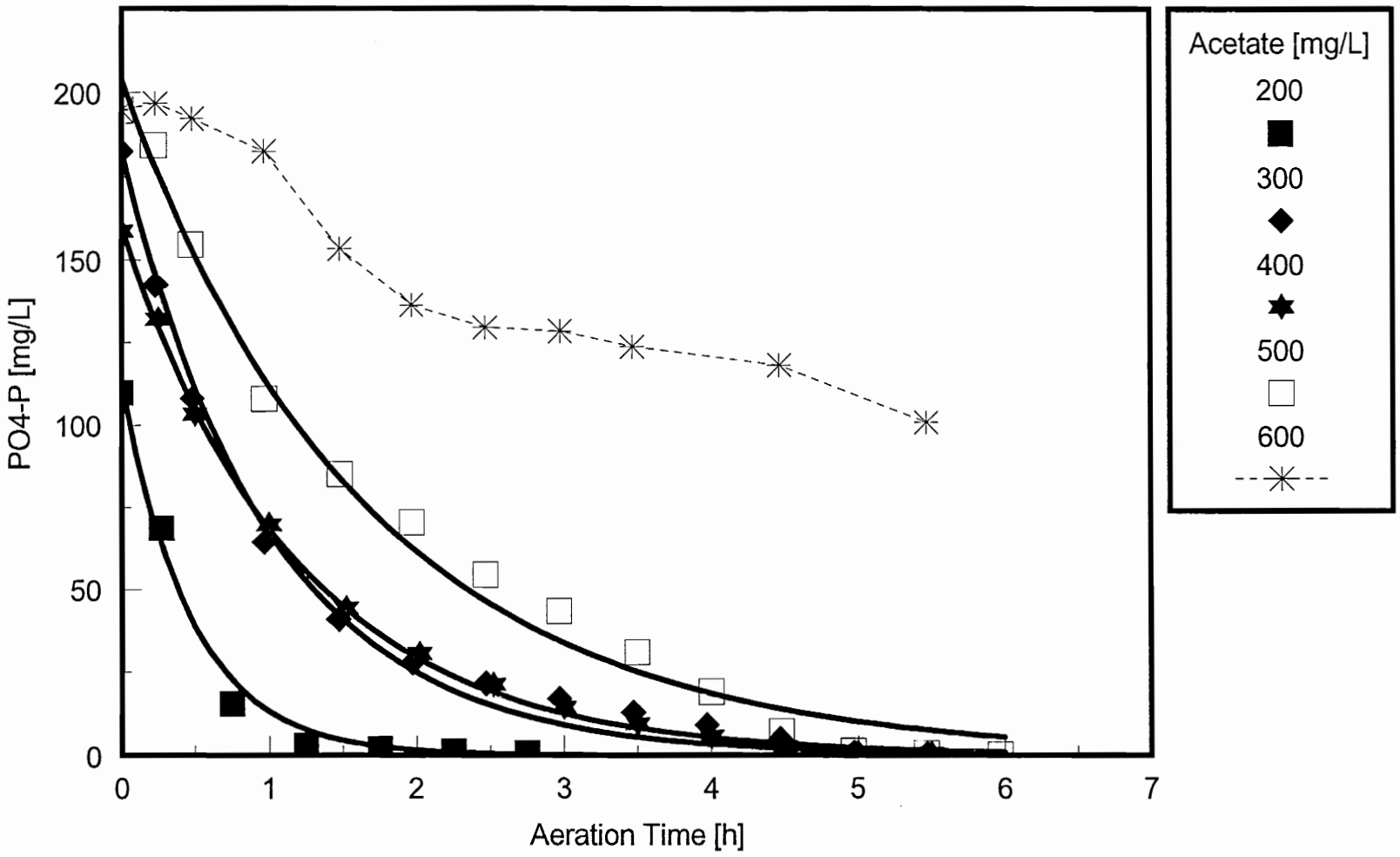

Figure 4.17 PO_4 - P Conc. during first Series of Batch Tests of System II; Aerobic Time

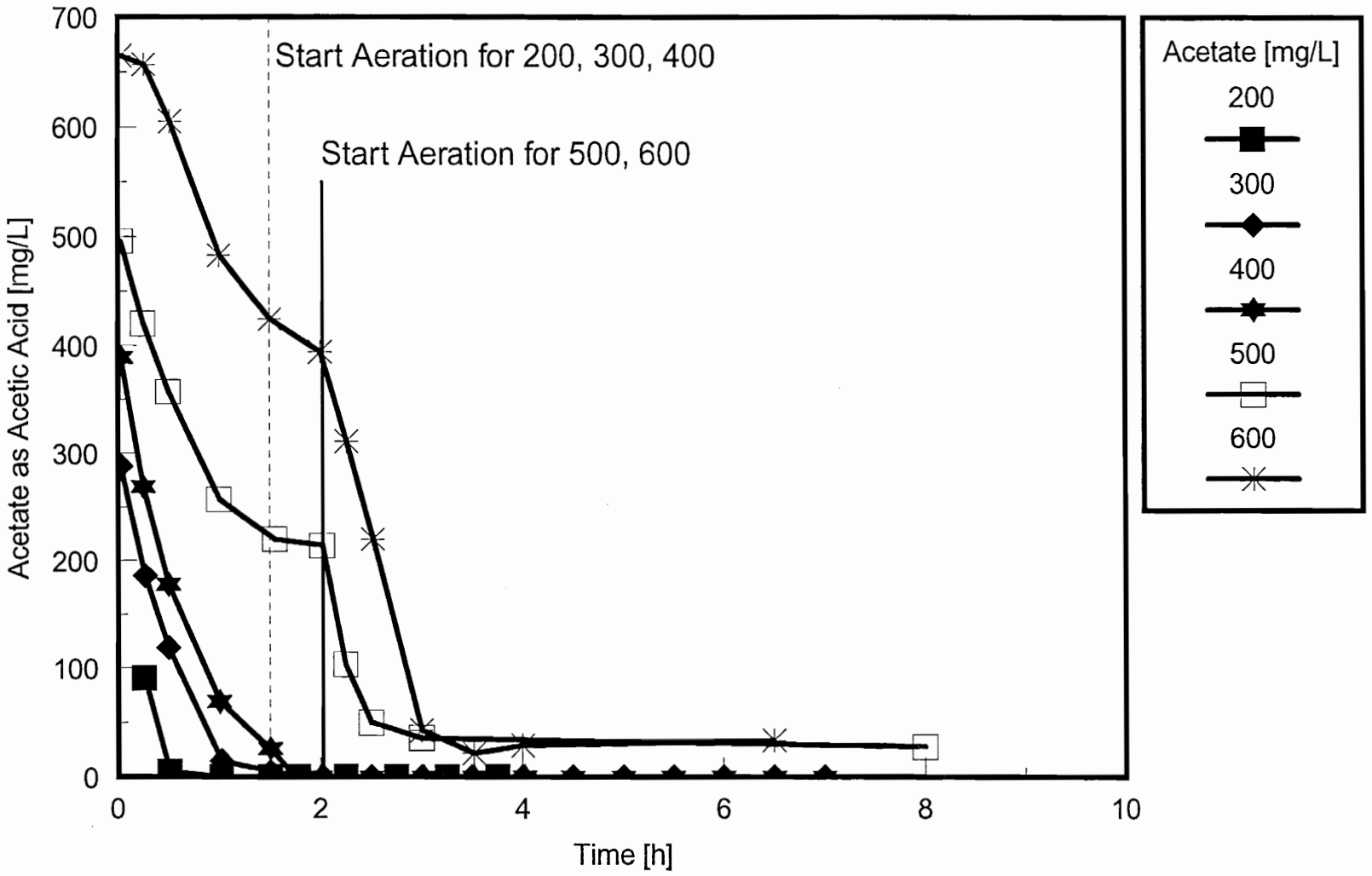

Figure 4.18 Acetate Concentrations during first Series of Batch Tests of System II

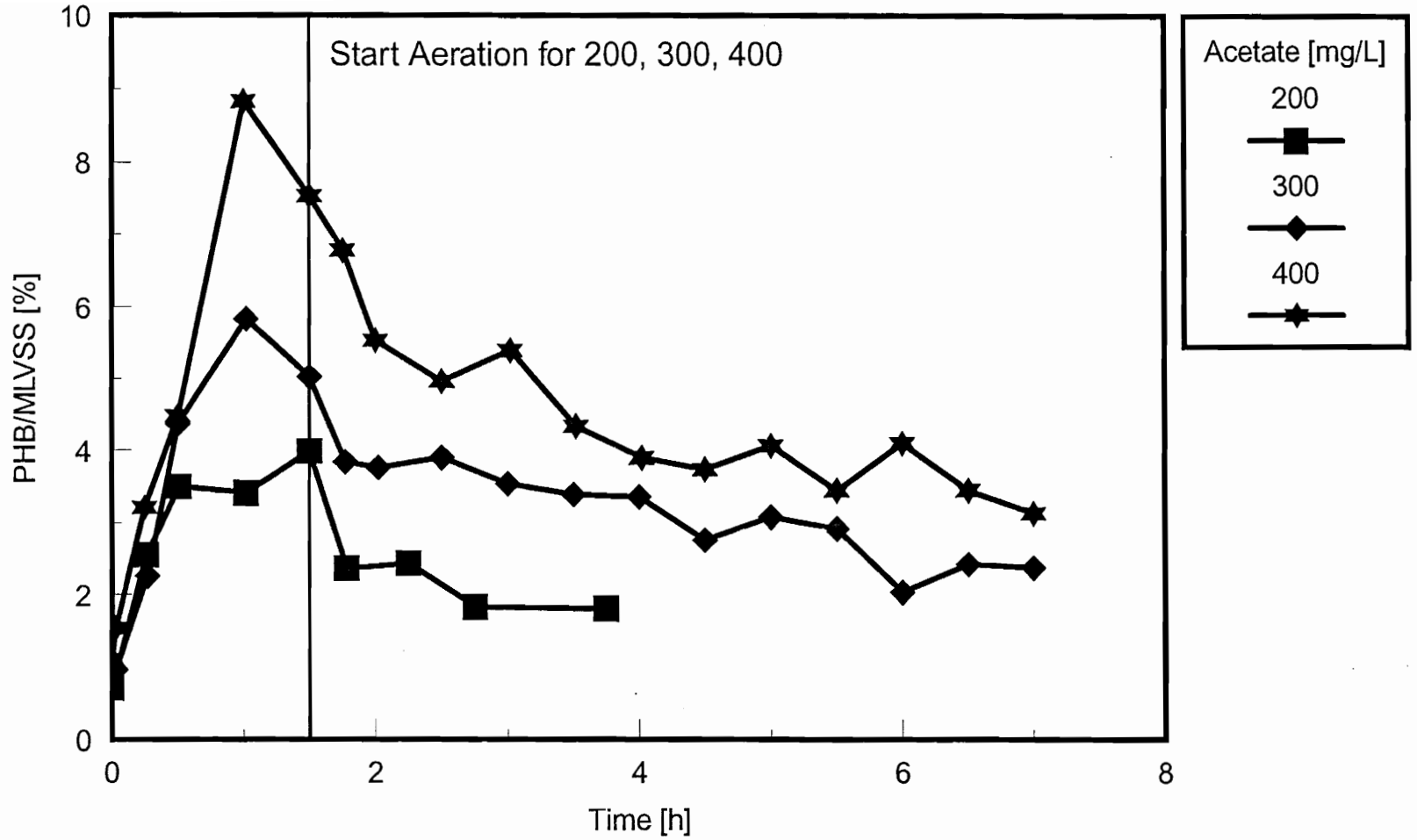

Figure 4.19 PHB Conc. during first Series of Batch Tests of System II (200 to 400 mg/L Acet.)

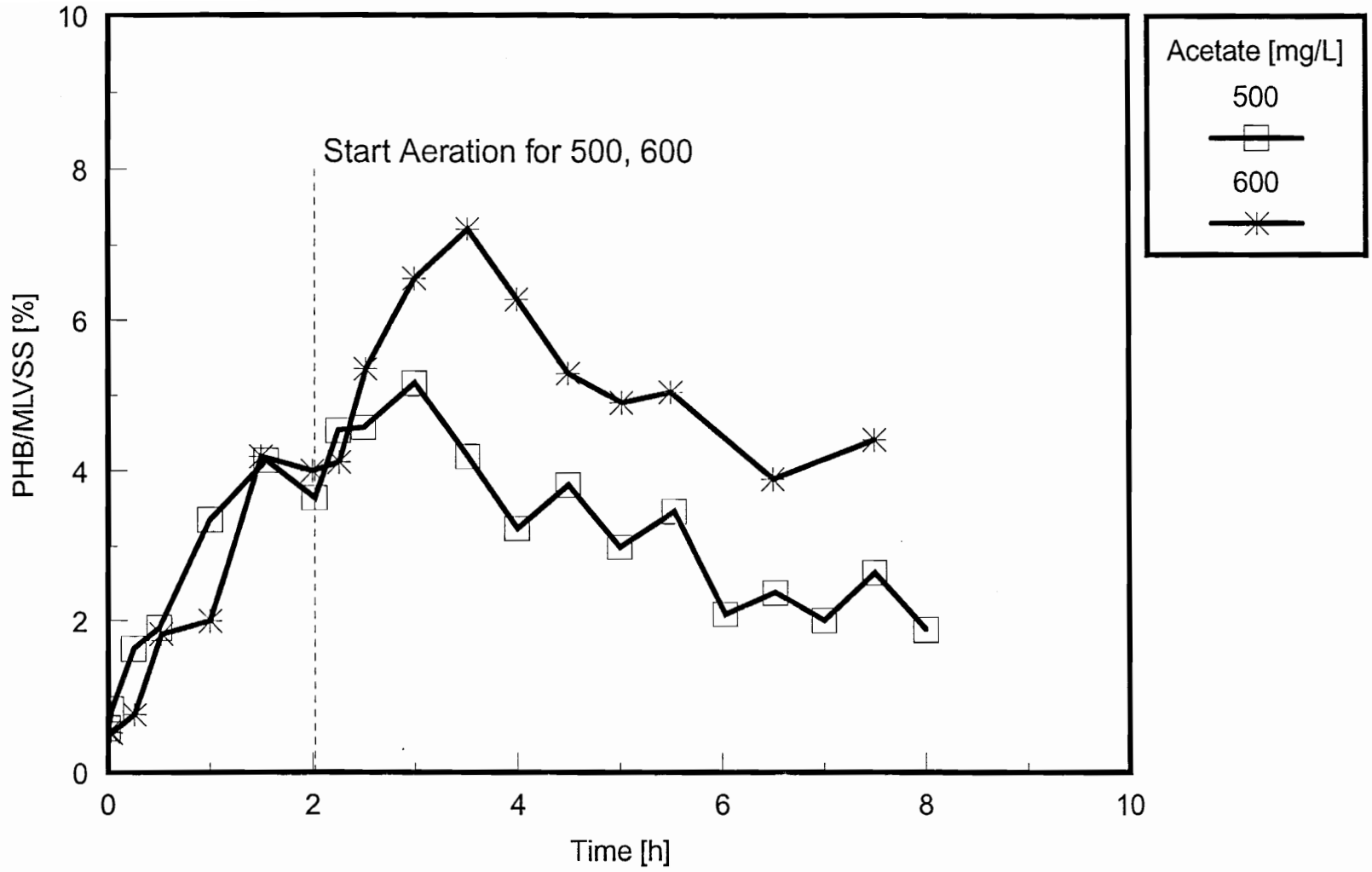

Figure 4.20 PHB Conc. during first Series of Batch Tests of System II (500 and 600 mg/L Acet.)

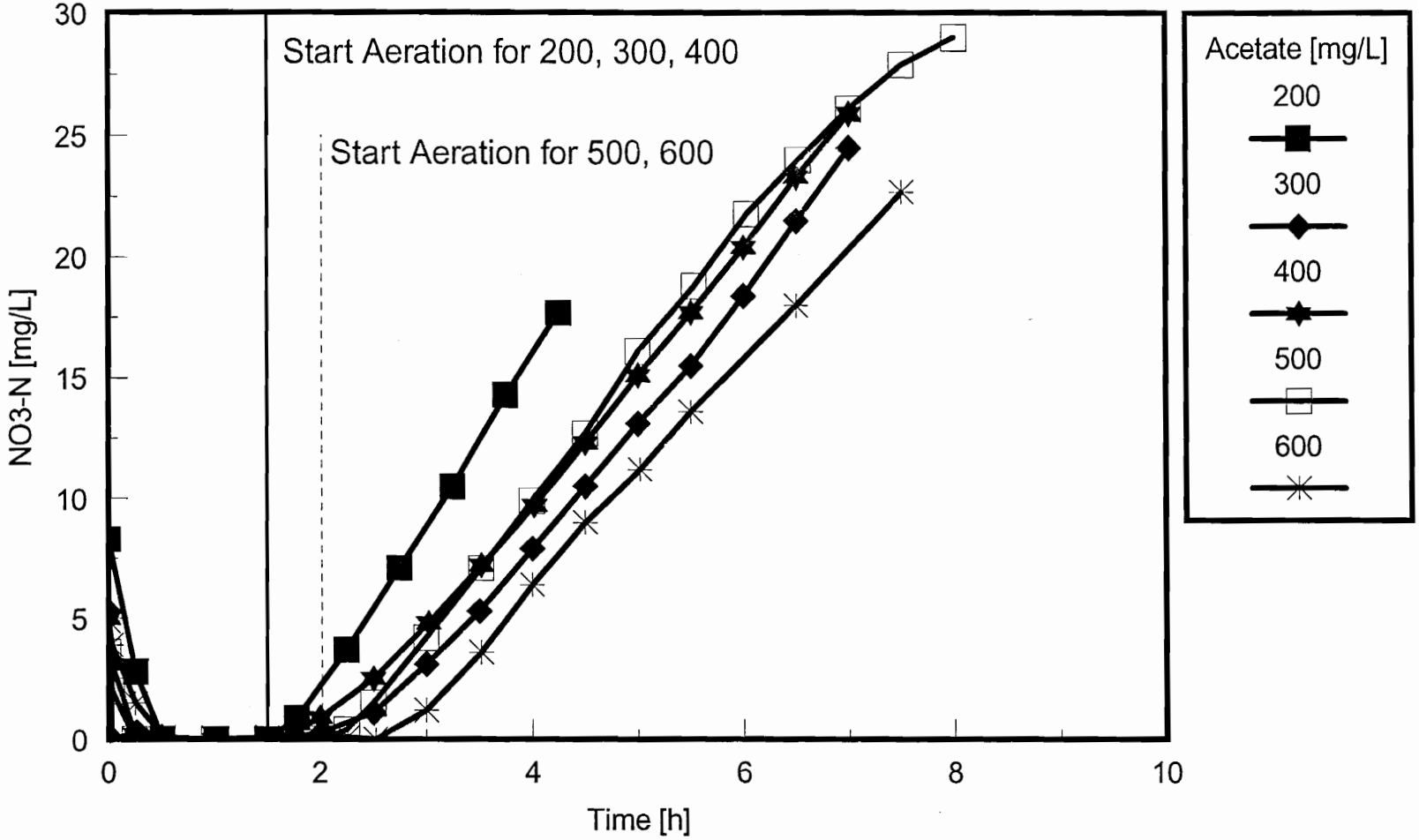

Figure 4.21 Nitrate Concentrations during first Series of Batch Tests of System II

4.4 Results for batch tests with varying amounts of nutrients

In this section, the results for the third set of batch tests with a total acetate concentration added of 600 mg/L are shown. Since each batch test was performed under different conditions for nutrients, time of anaerobic and aerobic phase, and time of the addition of a second spike, the results for the three most important parameters : phosphate, PHB, and acetate, are shown together in a normalized form for each batch test. Furthermore, the bar - charts for the feed are included in the graphs. These bar - charts are normalized for the feed under steady state conditions.

Figure 4.22 shows again the results for the batch test with 600 mg/L acetate, i.e. the same shown in the previous section. The vertical line marks the beginning of aeration. The correlation between acetate and PHB is obvious. Furthermore, while the acetate was consumed during the aerobic phase, the PHB concentration increased proportionally.

Figure 4.23 shows the results for batch tests II.2.a, during which the acetate was added in two equal spikes. This experiment was conducted to determine if the occurrence of acetate in the aerobic zone caused the failure of BPR that was observed in the previous batch test (II.1d). The phosphorus was removed much better than in the experiment shown in Figure 4.22. Still, the concentrations for PHB and acetate were reciprocal.

Figure 4.24 shows the results of batch test II.2.b, during which the concentration for all nutrients were raised proportional to the acetate concentration. This experiment was conducted to determine if nutrient deficiencies caused the failure of BPR. The PHB did not decrease much during aeration, as happened in the experiments mentioned above. However, the phosphate was sufficiently removed from the mixed liquor.

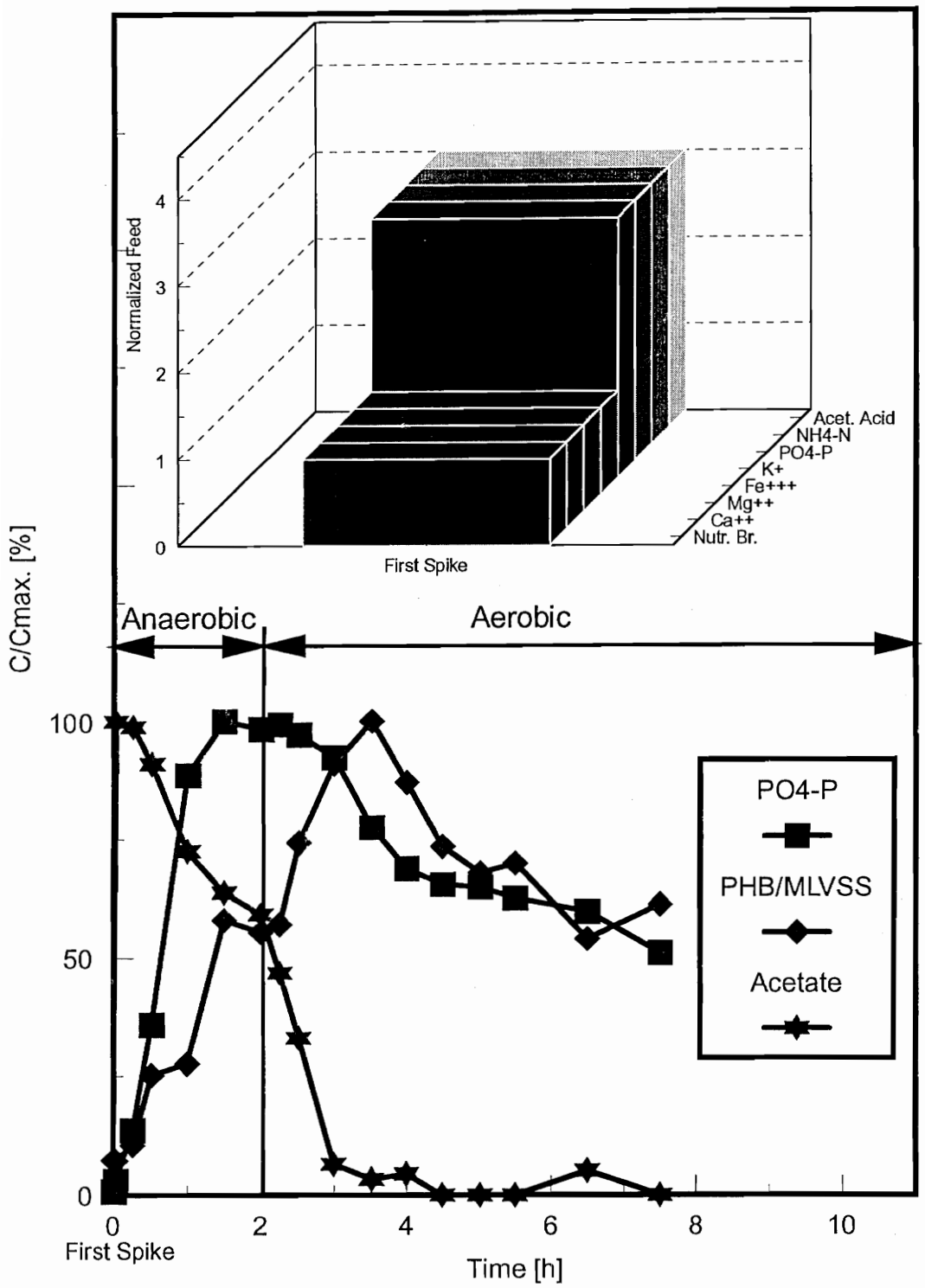

Figure 4.22 Batch Test II.1.d (High Acetate; Normal Nutrients)

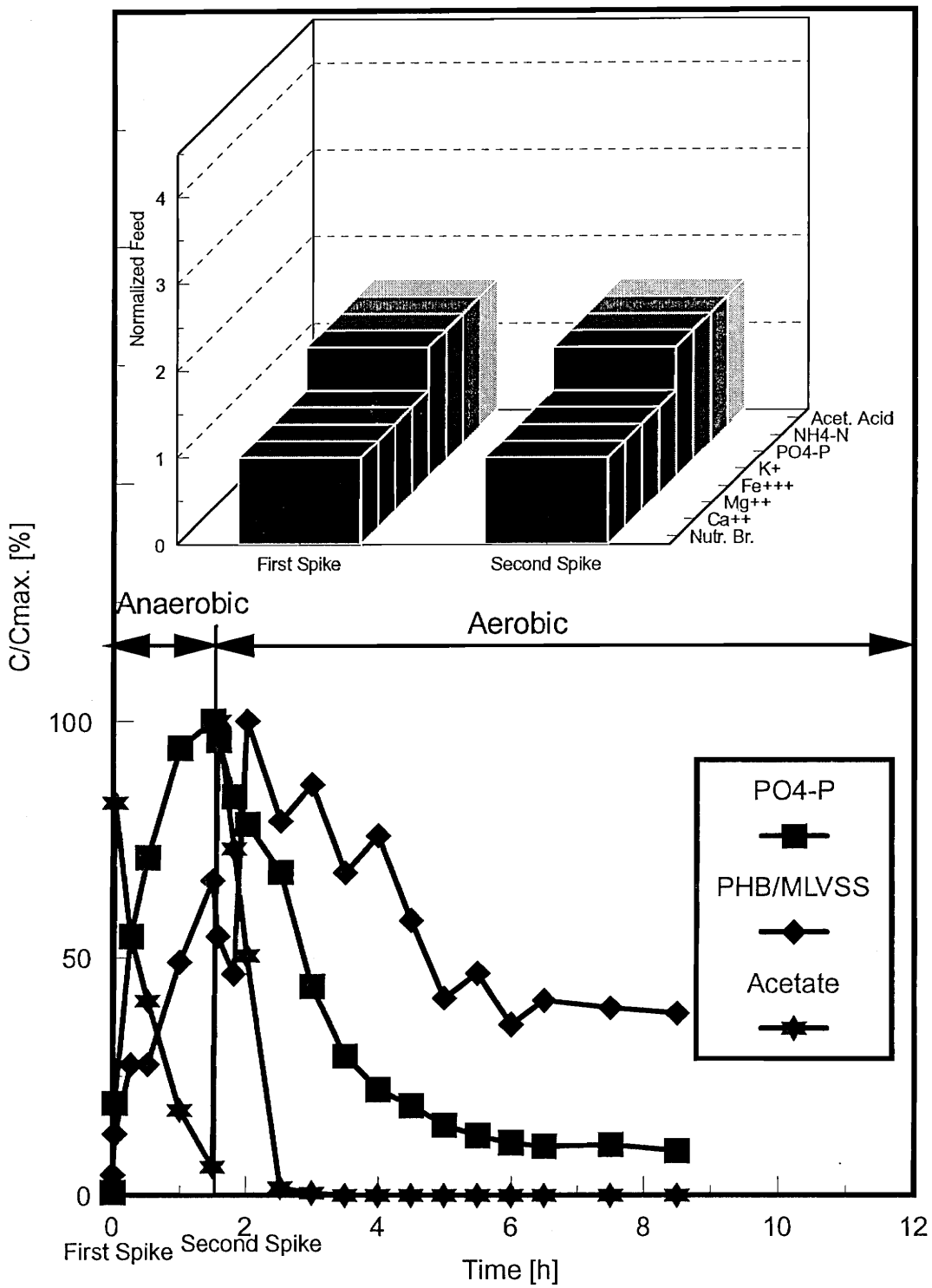

Figure 4.23 Batch Test II.2.a (Two Acetate Spikes)

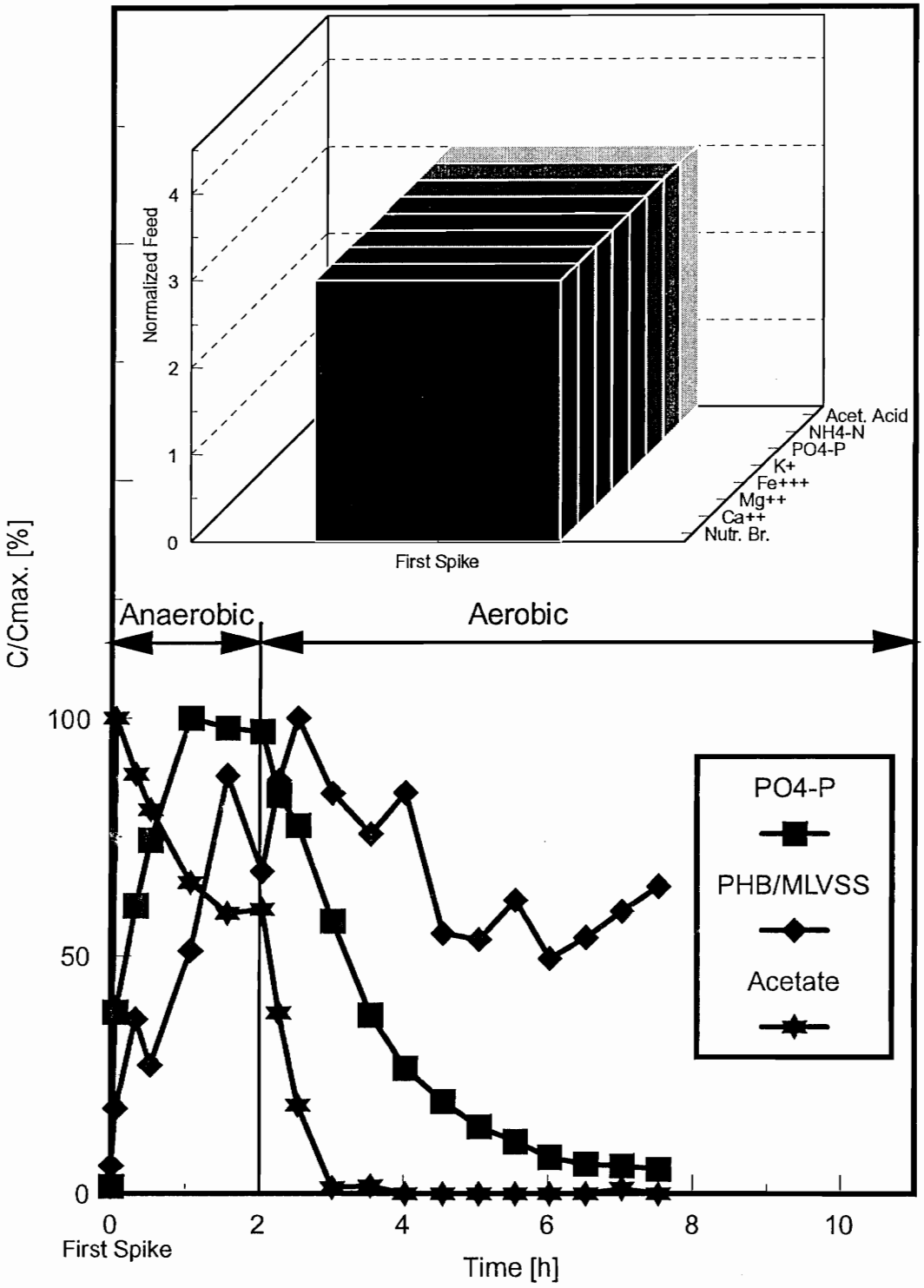

Figure 4.24 Batch Test II.2b (One Spike, Everything increased)

Figure 4.25 shows the results for batch test II.2.c. The first vertical line marks the beginning of aeration and the second vertical line marks the point at which the second spike, in this case the difference in nutrients between the experiment shown in Figure 4.22 and the one shown in 4.24, was added. This experiment was conducted to determine if nutrient deficiencies in the anaerobic or the aerobic zone caused the failure of BPR. Up to the point where the second spike was added, the conditions were the same as in the experiment shown in Figure 4.22, and, the results were similar. But, the the phosphorus cpncentrations dropped faster and further after the nutrients were added, compared to the former experiment. This experiment established that the possibility of nutrient deficiencies in the aerobic zone needed further research.

Figure 4.26 shows the results for batch test II.2.d. The first vertical line marks the beginning of aeration and the second vertical line marks the point at which the second spike was added, in this case nutrient broth. This experiment was conducted to determine if a lack of vitamins, whose only possible source could be the nutrient broth, caused the failure of BPR. Up to that point where the second spike were added the conditions were the same as in the experiment shown in Figure 4.22, and the results were similar. But, the drop in the phosphorus concentration after the second spike was not nearly as fast as it was in Figure 4.24, and the efficiency of phosphorus removal was low.

Finally, the experiment shown in Figure 4.27 was conducted to examine if the additional calcium caused the good phosphorus removal shown in Figure 4.24. Therefore, only calcium chloride was added with the second spike. The decrease in phosphorus was similar to the one in the experiment shown in Figure 4.24, suggesting an important role of the calcium.

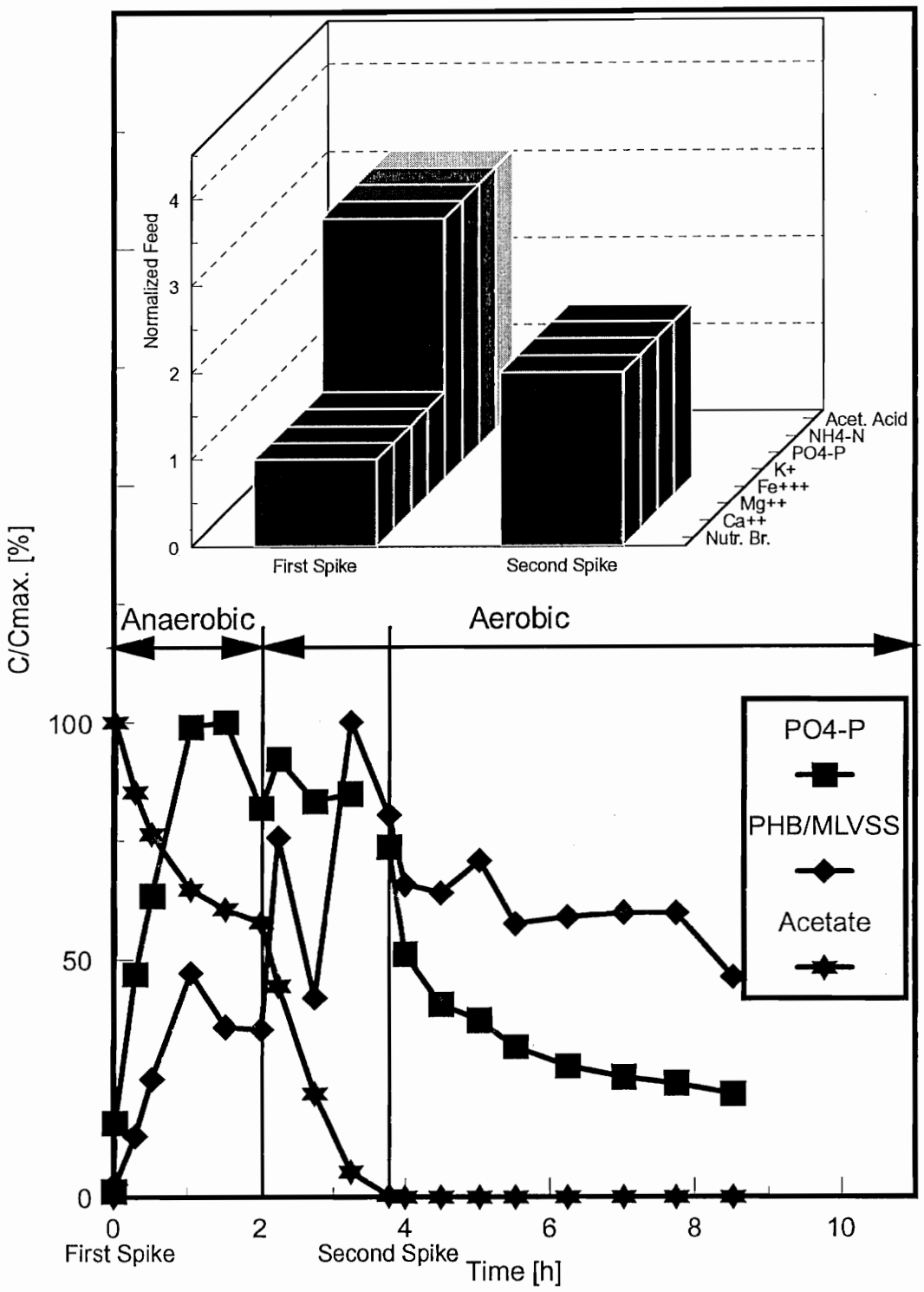

Figure 4.25 Batch Test II.2c (Additional Nutrients in second Spike)

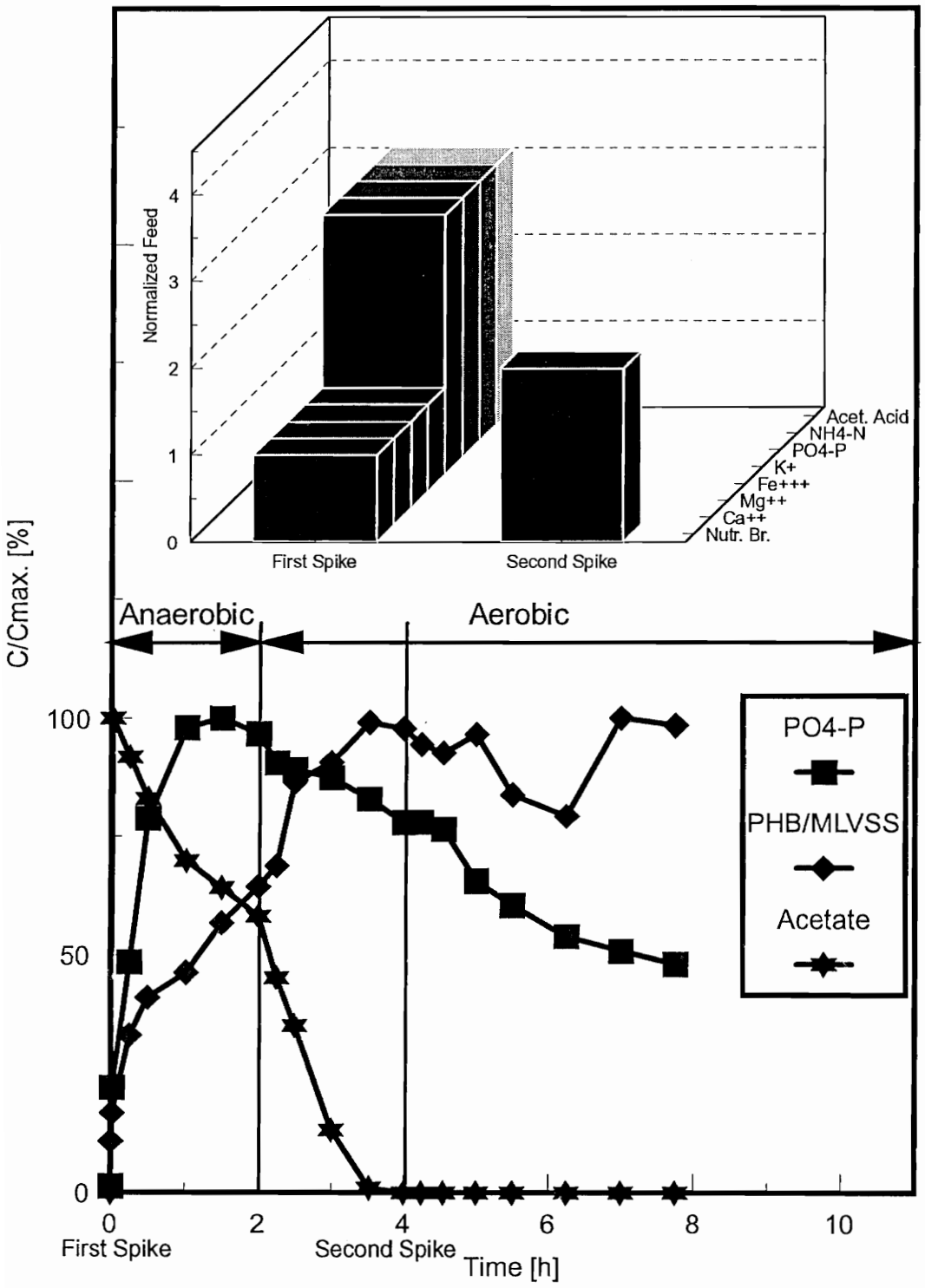

Figure 4.26 Batch Test II.2d (Nutrient Broth in second Spike)

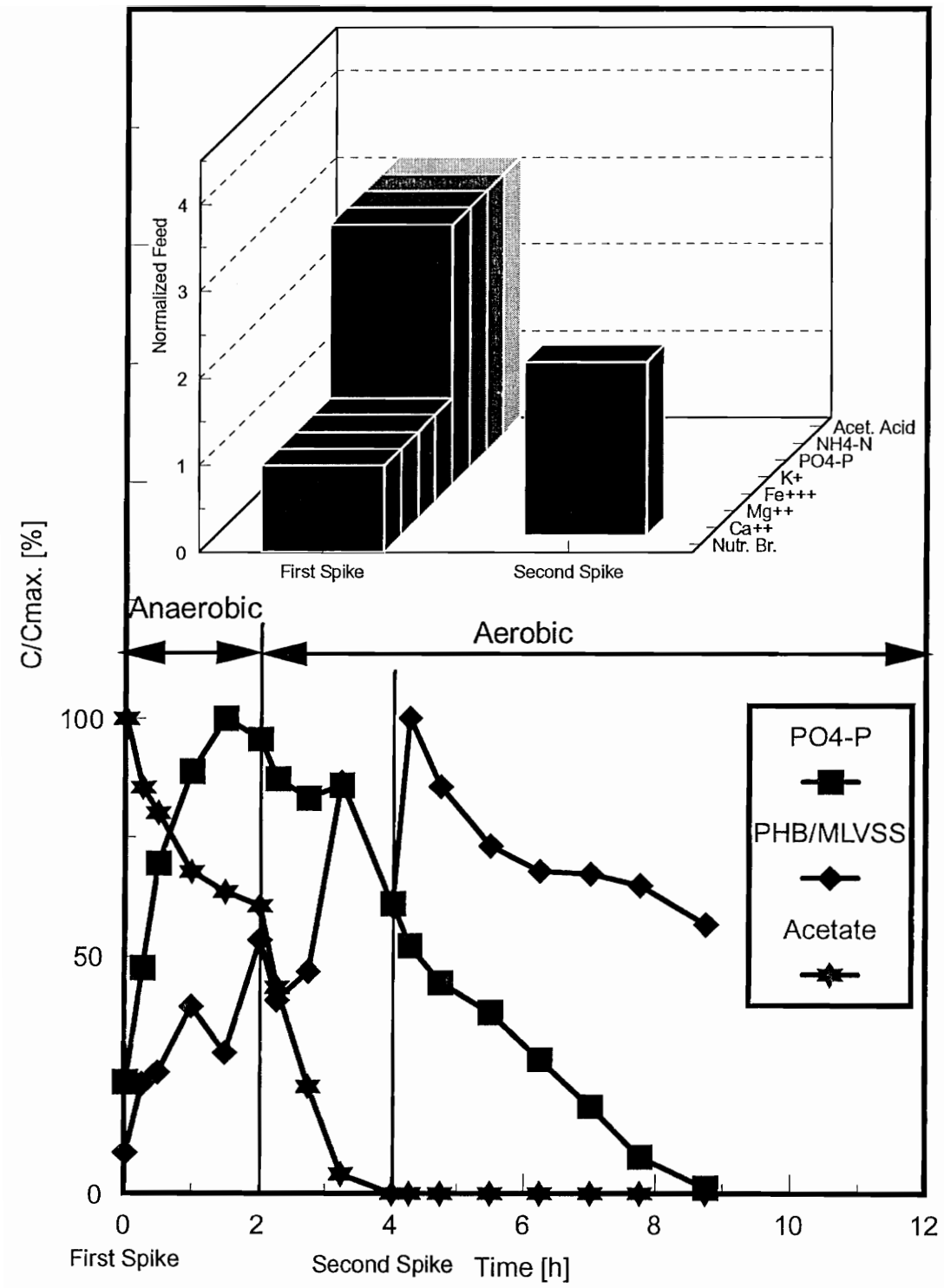

Figure 4.27 Batch Test II.2e (Calcium Chloride in second Spike)

4.5 Results for precipitation experiments

The results for the two precipitation experiments are shown in Figure 4.28. These experiments were conducted with the feed, but without bacteria, at an increased pH to determine if calcium phosphate precipitation caused the effects shown in Figure 4.24 and 4.27. Most of the phosphorus removal happened during the first moments of the experiment, and only up to 40 mg/L of phosphorus were removed.

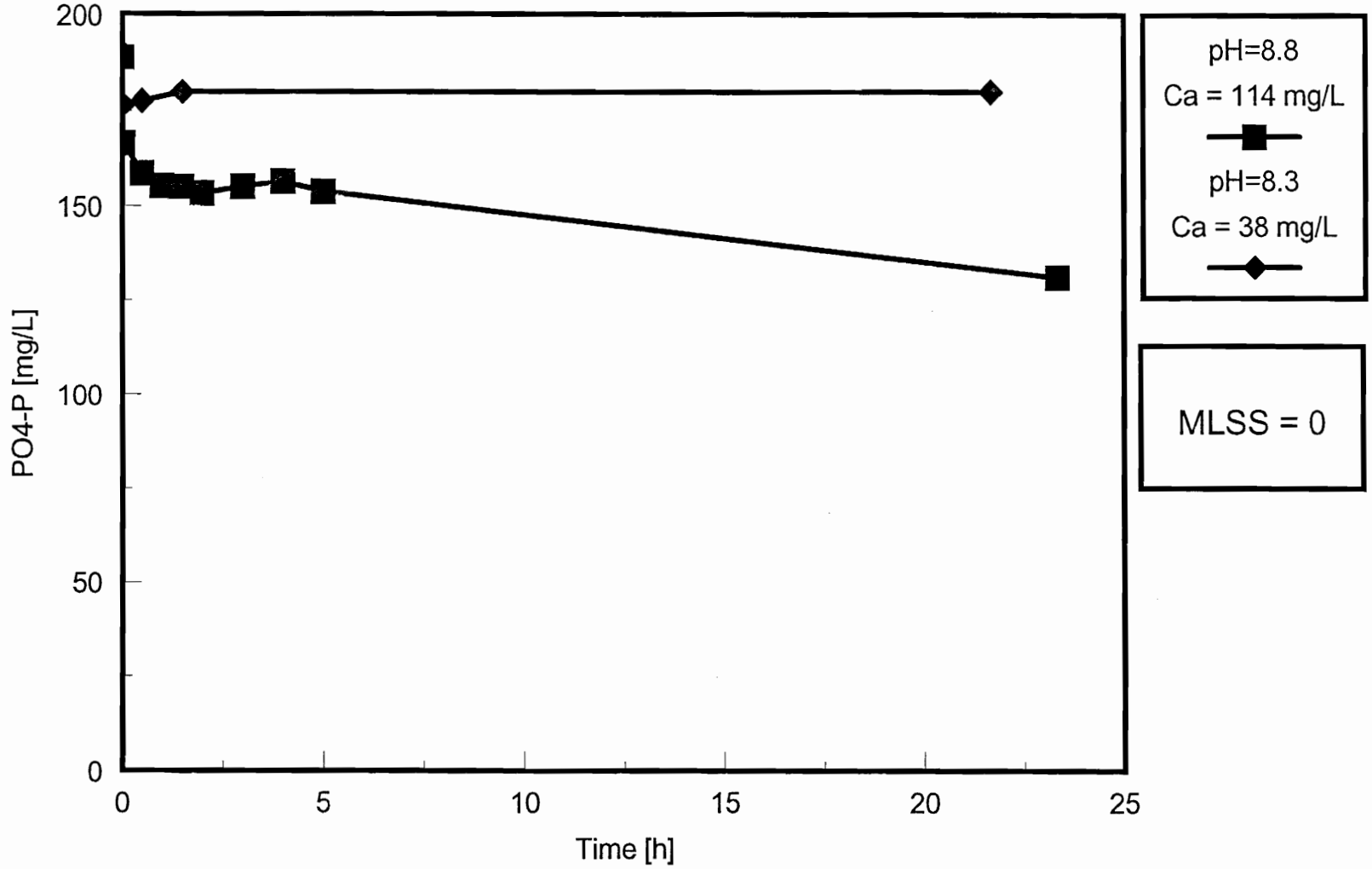

Figure 4.28 Precipitation Experiments

5. Discussion

5.1 Discussion of the continuous flow data

The profiles for System I and System II show that both systems performed very similarly, which is not very surprising, since both systems were started with the same microbial composition. With the exception of the values for PHB, all parameters were nearly identical. However, it is not clear why System II lost more solids into the effluent than System I.

The profiles of System I show that the intended breakdown of the system did not happen. Instead, the system adapted to high acetate concentrations without any problems. It removed most of the phosphorus and no acetate broke through to the aerobic zone (Figures 4.6 and 4.9). Nevertheless, it is possible that at an acetate concentration of 800 mg/L the system would have eventually failed due to the inability to maintain such a high (up to 10 g/L) MLSS concentration. The observation that the maximum concentration of phosphorus released increased nearly proportionally for acetate concentrations from 200 mg/L to 600 mg/L, but not for 800 mg/L (Figure 4.6), is a possible sign of the beginning of breakdown. But, System I was shut down at that time to allow more time for batch tests. Therefore, this part of the experiments could neither confirm nor reject the observations made by Randall and Chapin (1994) regarding the influence of acetate on washout of bacteria and sludge bulking. However, the main difference in the system configuration of Chapin (1993) and the configuration of the system used for this research is that the former reached a maximum MLSS of only 4 g/L while the latter reached a maximum MLSS of 10 g/L. In other words, the system used for this research adapted much better to high acetate loadings with the consequence of a lower acetate to MLSS ratio. This might explain why no breakdown of BPR occurred under continuous flow conditions.

Figure 5.1 shows the summarized data for phosphate released versus acetate taken up. The figure shows both the values measured at the end of the anoxic zone and the values measured at the end of the anaerobic zone. The ratio of P released to acetate utilized is 1.06 mol/mol ($r^2 = 0.95$). This is in near perfect accordance with the model by Comeau/Wentzel (1991). Additionally, the ratio of PHB synthesized to acetate utilized is 0.49 mol/mol ($r^2 = 0.69$) as shown in Figure 5.2, 15 % higher than the 0.44 proposed by Comeau/Wentzel (1991) in their model and 30 % lower than the 0.67 proposed by Mino *et al.* (1987). In Figure 5.2 the values for PHB over COD (r^2=0.57) are plotted also, with the COD transformed into acetate equivalents (1 mole acetate = 64 g COD). The incline of the regression lines differs by 25%; however, for some of the data points only COD data were available and for some only acetate data. Therefore, the reason for the difference between the observed and theoretical ratios of PHB synthesized to acetate utilized may or may not be the utilization of COD from the nutrient broth.

Figure 5.3 shows the results for PHB synthesized versus phosphorus released. The regression line ($r^2 = 0.61$) does not intercept the origin; the incline is 0.39 mol PHB / mol P released. This value is relatively near to the value of 0.44 mol/mol proposed by Comeau/Wentzel (1991). The failure to intercept the origin can be explained with the observation that not all PHB had been metabolized at the end of the aerobic zone (Figure 4.10), leading to the accumulation of PHB in the bacteria. To summarize, the data for the measurements under continuous flow conditions follow the theoretical model proposed by Comeau/Wentzel (1991).

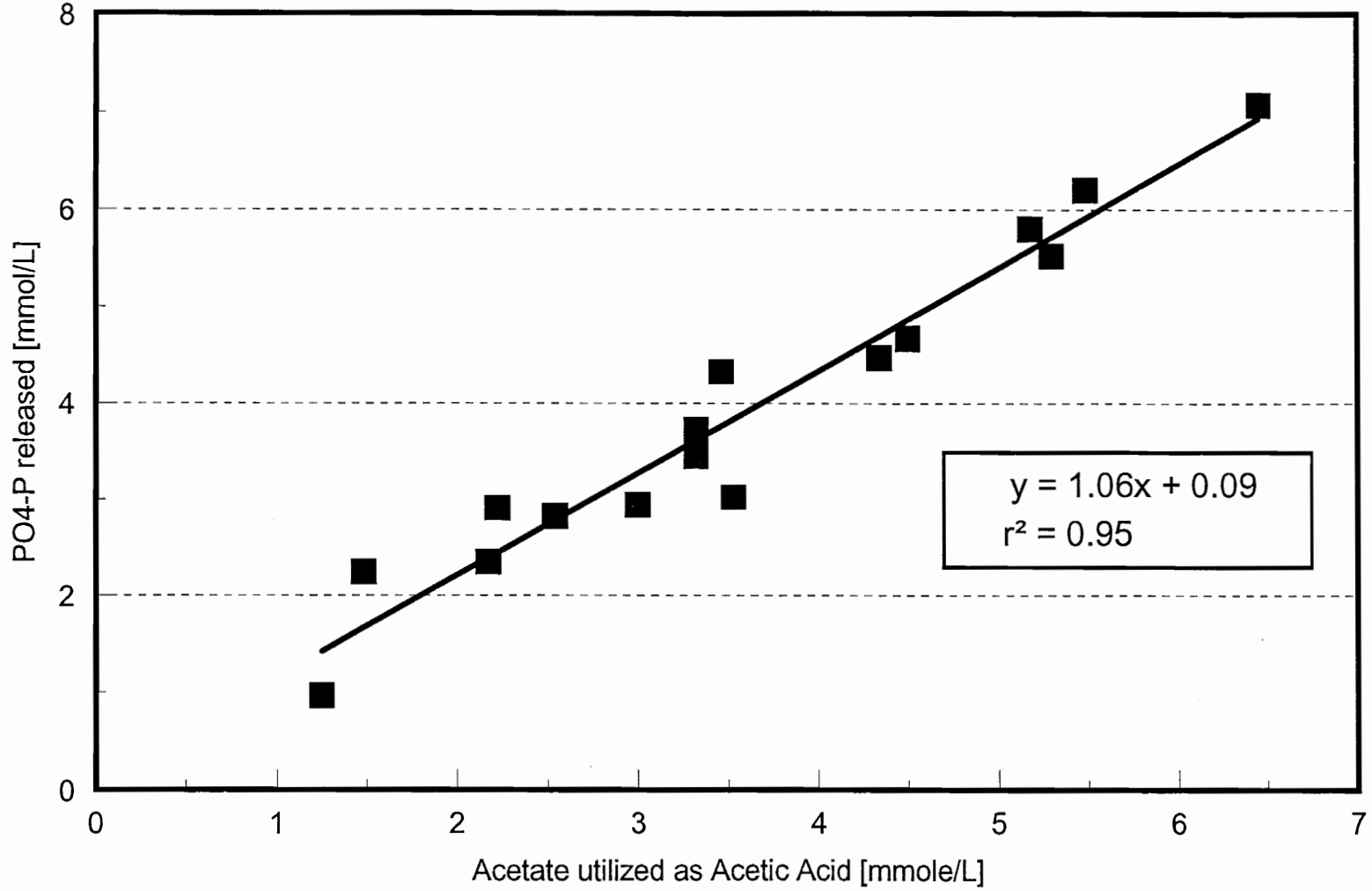

Figure 5.1 PO_4- P released versus Acetate utilized during Continous Flow Experiments

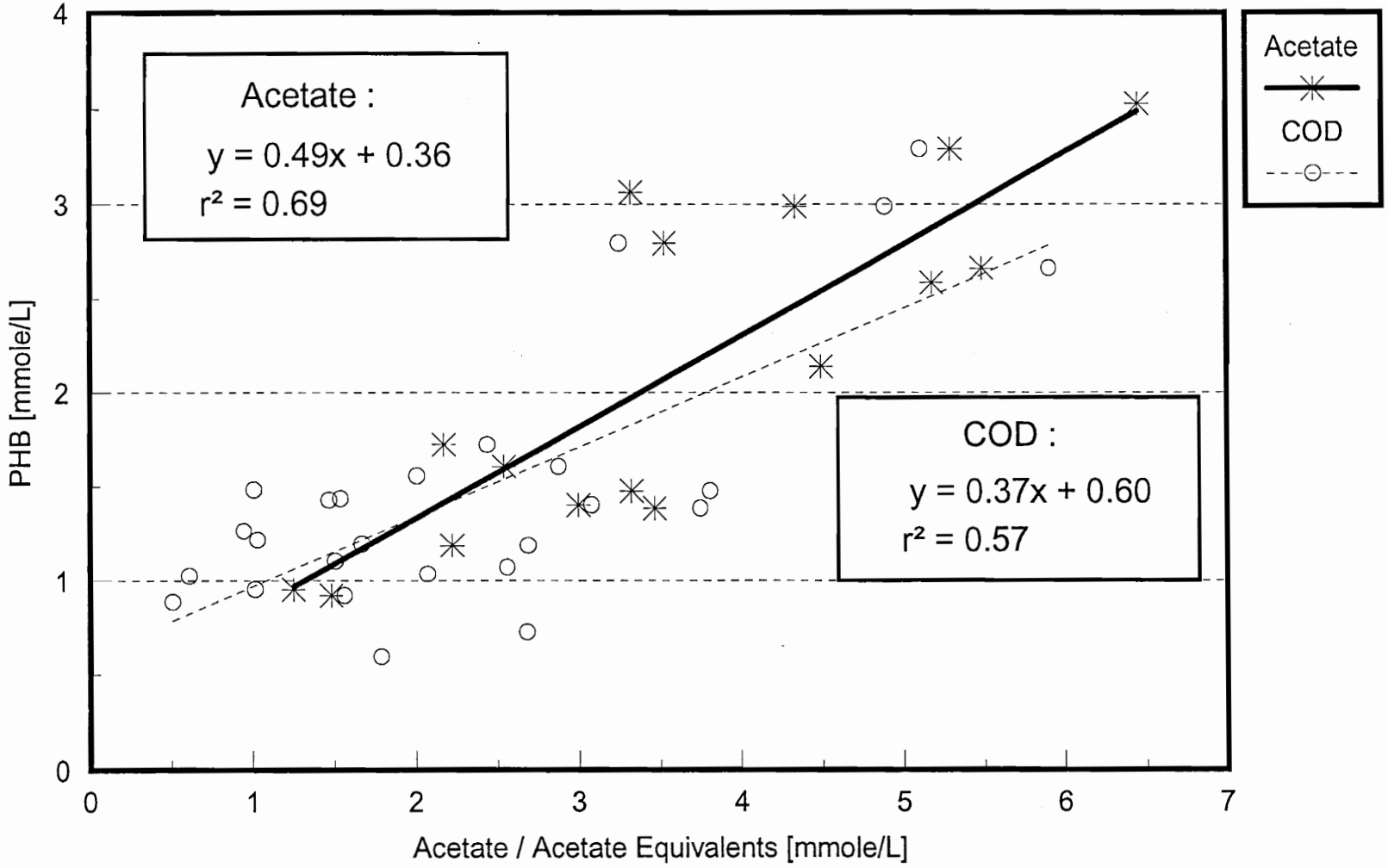

Figure 5.2 PHB synthesized versus Acetate utilized during Continuous Flow experiments

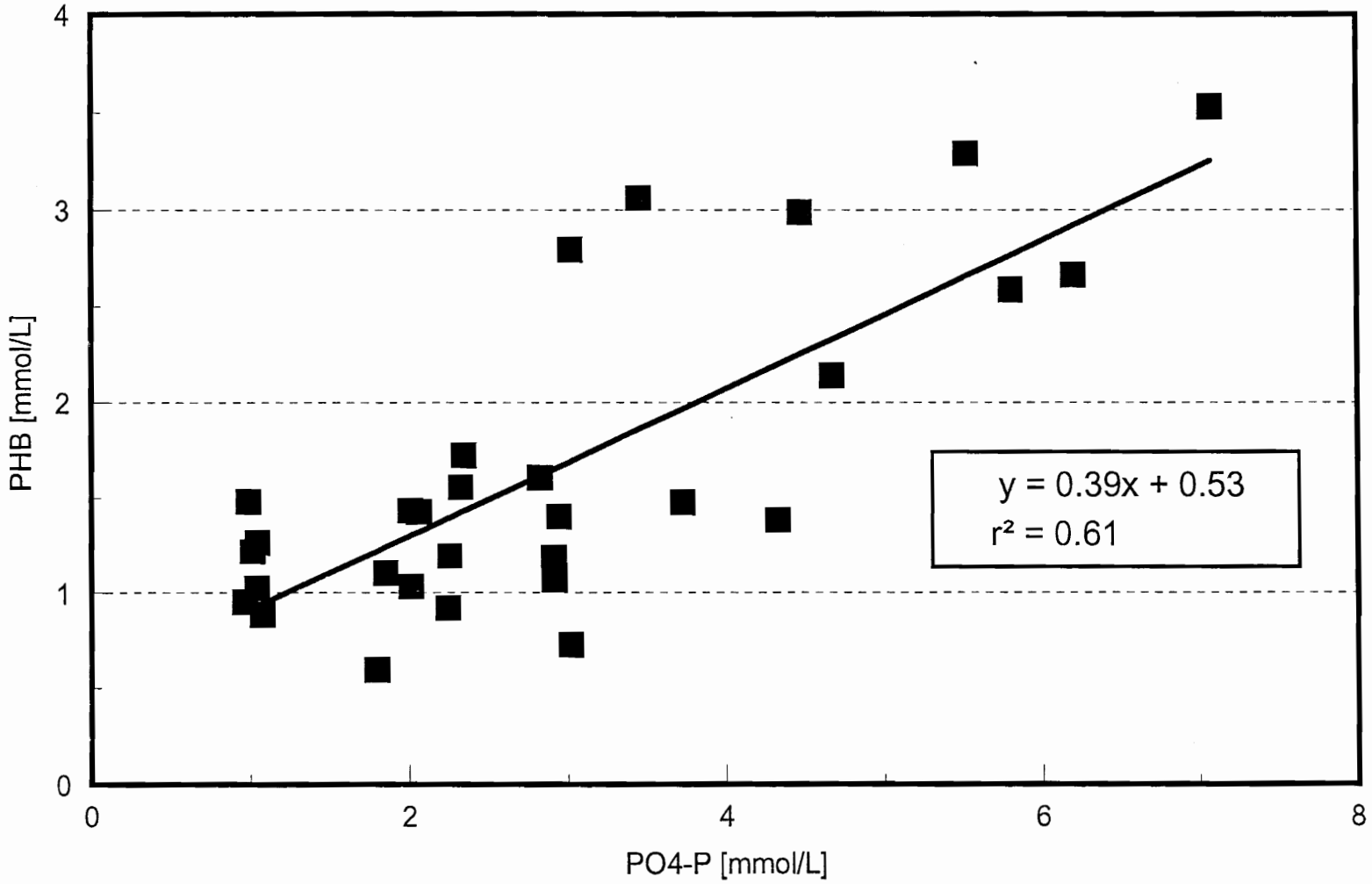

Figure 5.3 PHB synthesized versus PO_4 - P released during Continuous Flow Experiments

The batch tests with the sludge from System I did confirm that first order kinetics (Figure 4.11) are a good way to describe the process of phosphorus release and uptake. However, in connection with the above stated observations regarding the ratios of PHB, acetate, and phosphorus, it is not possible to deduct from the kinetics of these batch tests alone which of these substances controlled the speed of the reactions. However, the observation that phosphorus release stopped while acetate was still present and the PHB content of the cell was lower than its maximum value (Figures 4.18-4.20; 4.22-4.27), indicates that the kinetics were controlled by the phosphorus concentration.

A plot of phosphorus released vs. acetate utilized for the batch tests of System I is shown in Figure 5.4. The P / acetate ratio is 1.43 mol/mol (r^2 of 0.97). This value does not correspond to the value of 1.06 mol/mol obtained from the profiles and is higher than the theoretical value of 1 mol/mol for the Comeau/Wentzel model. The actual reason for this phenomenon is unclear. However, batch tests and continuous flow systems are different. For example, dispersed bacteria are washed out in a continuous flow system and retained in a batch test, it is easier to keep oxygen from entering the anaerobic "zone" during a batch test and the growth conditions for the bacteria in general are better in a batch test.

The plot for PHB vs. acetate utilized (Figure 5.5) shows the limitations of the method of PHB measurement employed. The scatter is obvious, and the regression line (dotted line) had a r^2 of only 0.4. The statistically much better results for the PHB measurements shown in Figure 5.2 ($r^2 = 0.69$) are most probably caused by the fact that each profile was measured up to three time and the values averaged. However, with a processing time of at least 20 minutes per PHB sample, the use of triplicate samples during batch testing and triple injections without an automated injector was not feasible (avg. 20 sampling points = 60 vials = 180 injections = 60 hours).

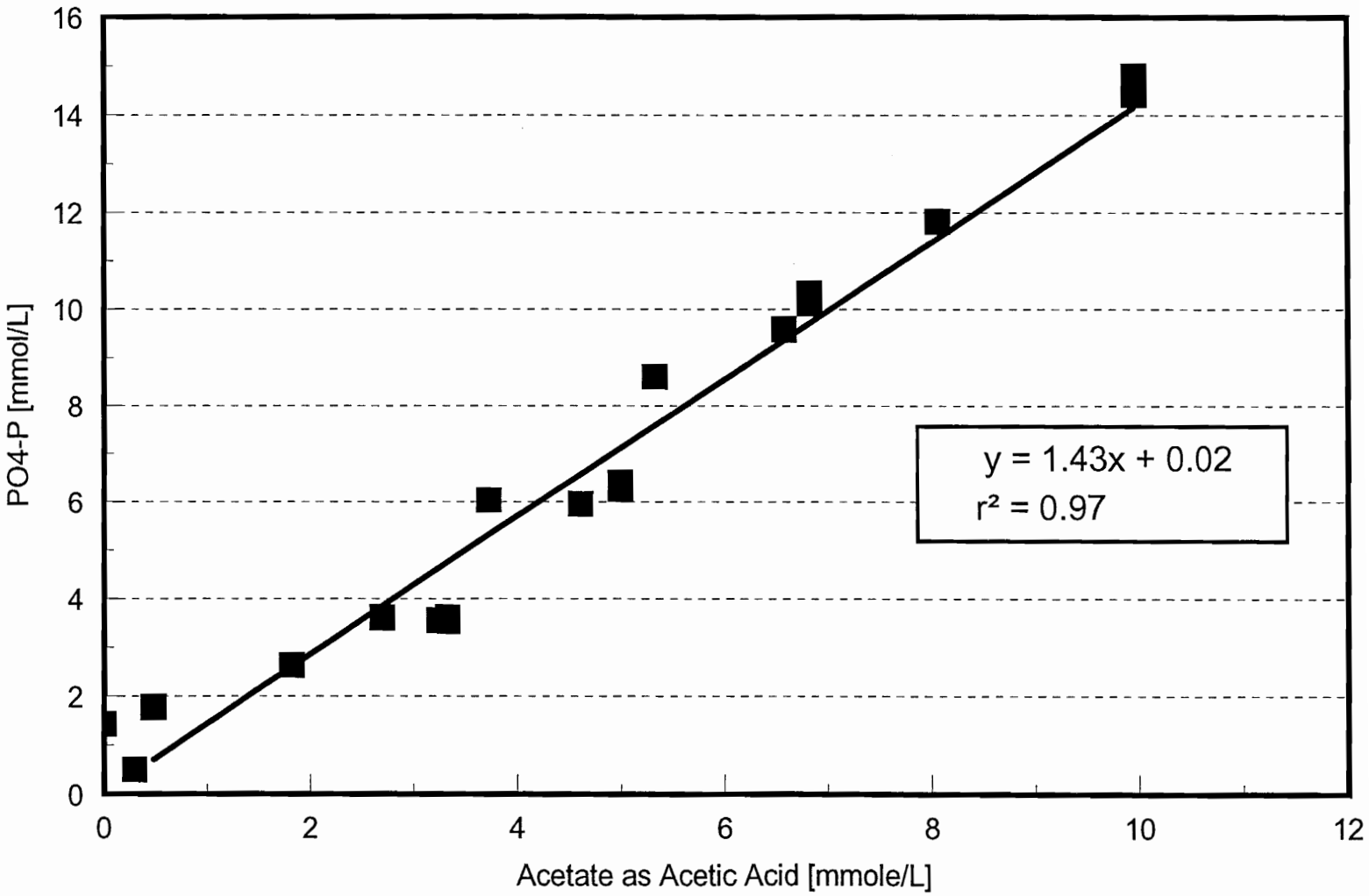

Figure 5.4 PO_4 - P released versus Acetate utilized during Batch Tests for System I

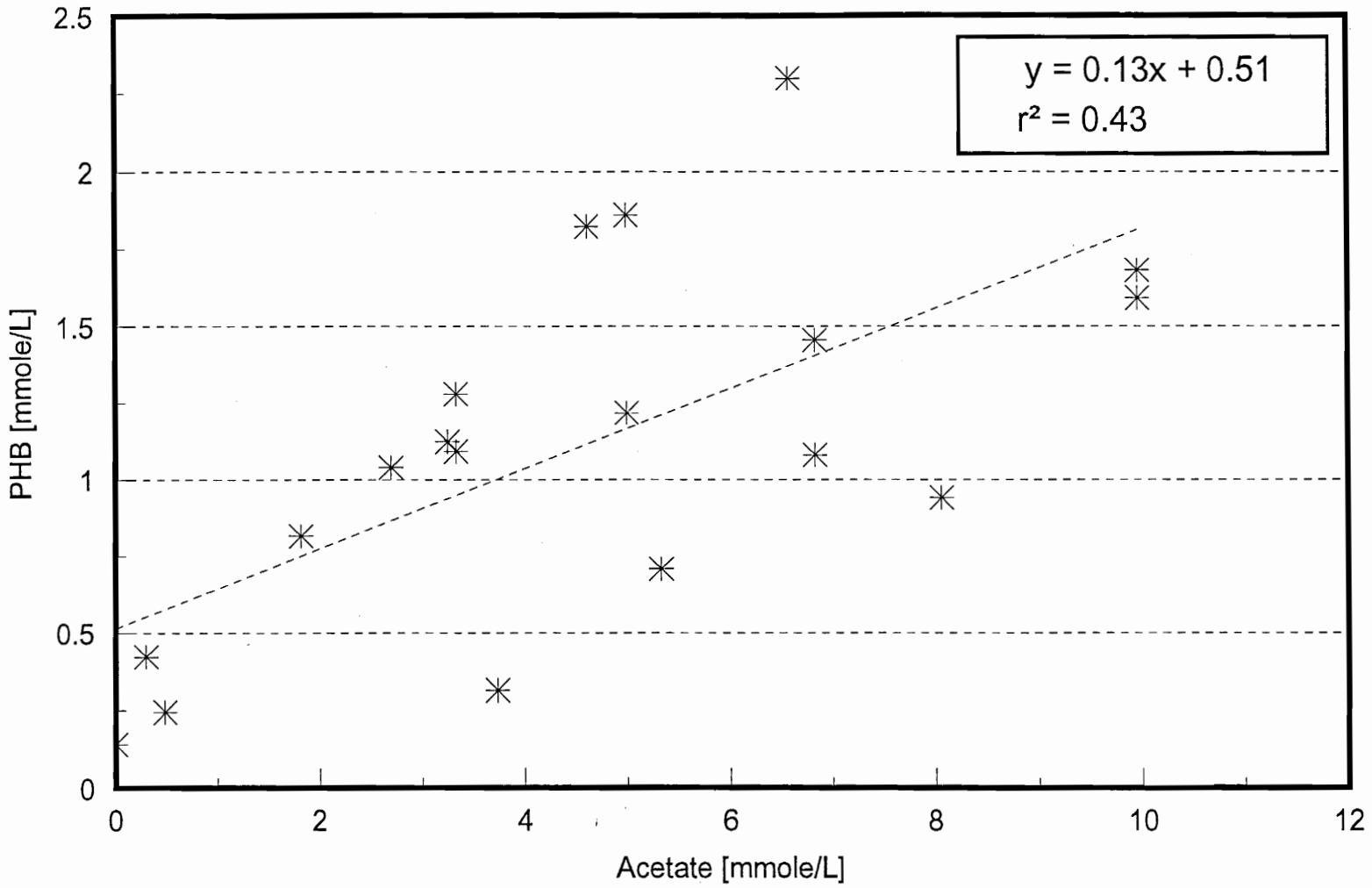

Figure 5.5 PHB synthesized versus Acetate utilized during Batch Tests for System I

Figure 5.6 shows the kinetic parameters obtained from the batch test of System I and used in Figure 4.11. The regressions fit quite good; the lowest r^2 is 0.96, the highest 0.998. In judging these data, it is important to remember that the absolute speed of the reaction is determined by the maximal possible concentration **and** the kinetic coefficient. Nevertheless, the relative speed of phosphorus uptake and release seems to drop with increasing acid concentrations. However, the model of Comeau/Wentzel proposes that acetate is transported into the cell by concentration gradient driven diffusion only. This means that the effect of high acetate concentrations on the cells inside a bacteria floc is lower than the effect on dispersed bacteria. As a result, a possible hypothesis for the effect of decreasing reaction rates at increasing acetate concentrations is that the transport step across the cell membrane and inside the bacteria floc slows down the overall reaction at higher absolute speeds. Other effects like enzyme inhibition seem less likely, because the effect of the high acetate concentrations seems to be limited as shown in Figure 5.7. A possible way to test this hypothesis is to destroy the bacterial flocs and to disperse the bacteria as evenly as possible in the mixed liquor without damaging the cell walls in the process. If the above stated hypothesis is true, the effect of the acetate concentration on the kinetic coefficient would be reduced, since the bacteria would expose their entire surface to the liquid, thus maximizing any passive transport step.

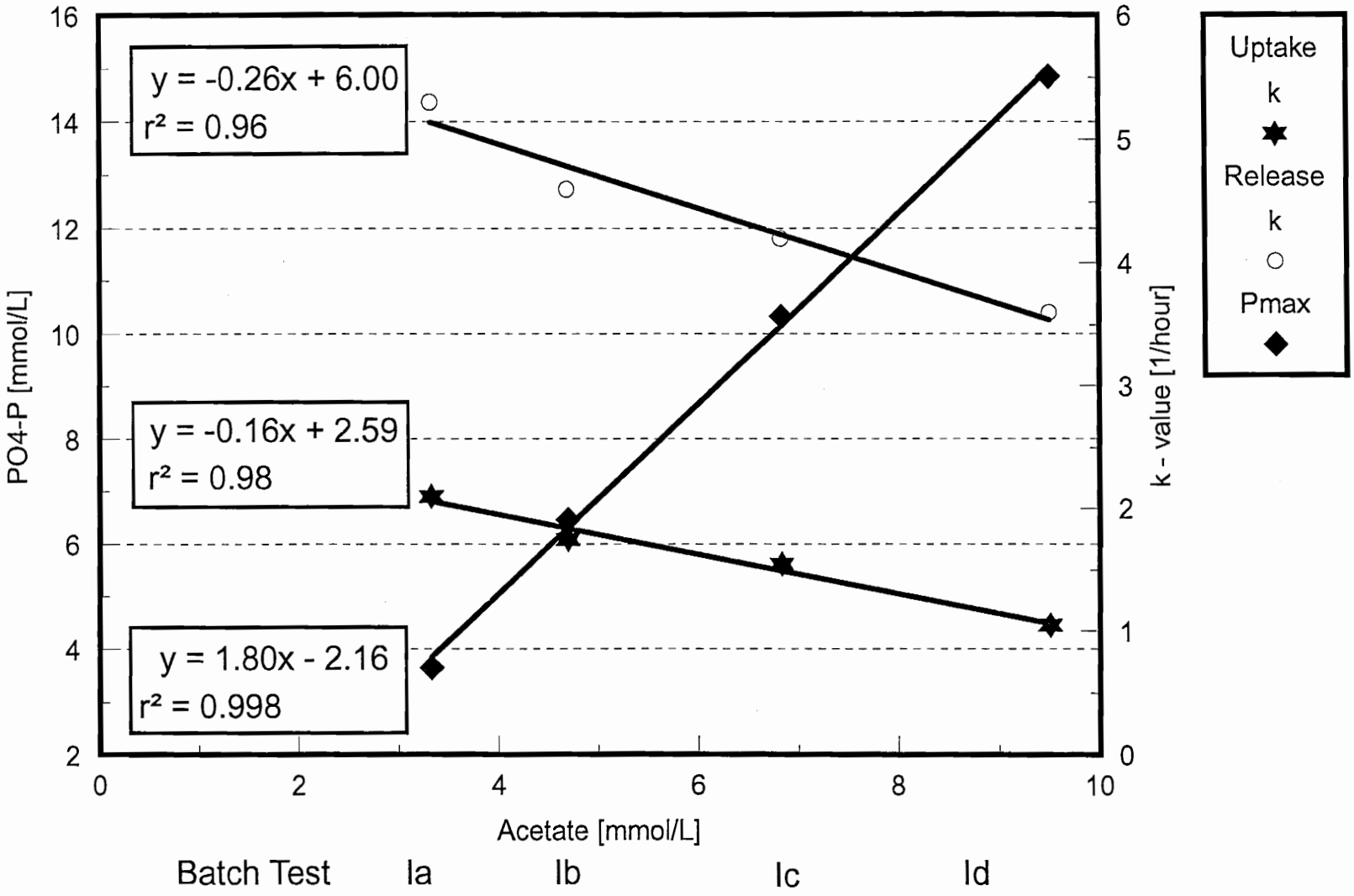

Figure 5.6 Kinetic Parameters for the Results of the Batch Tests for System I

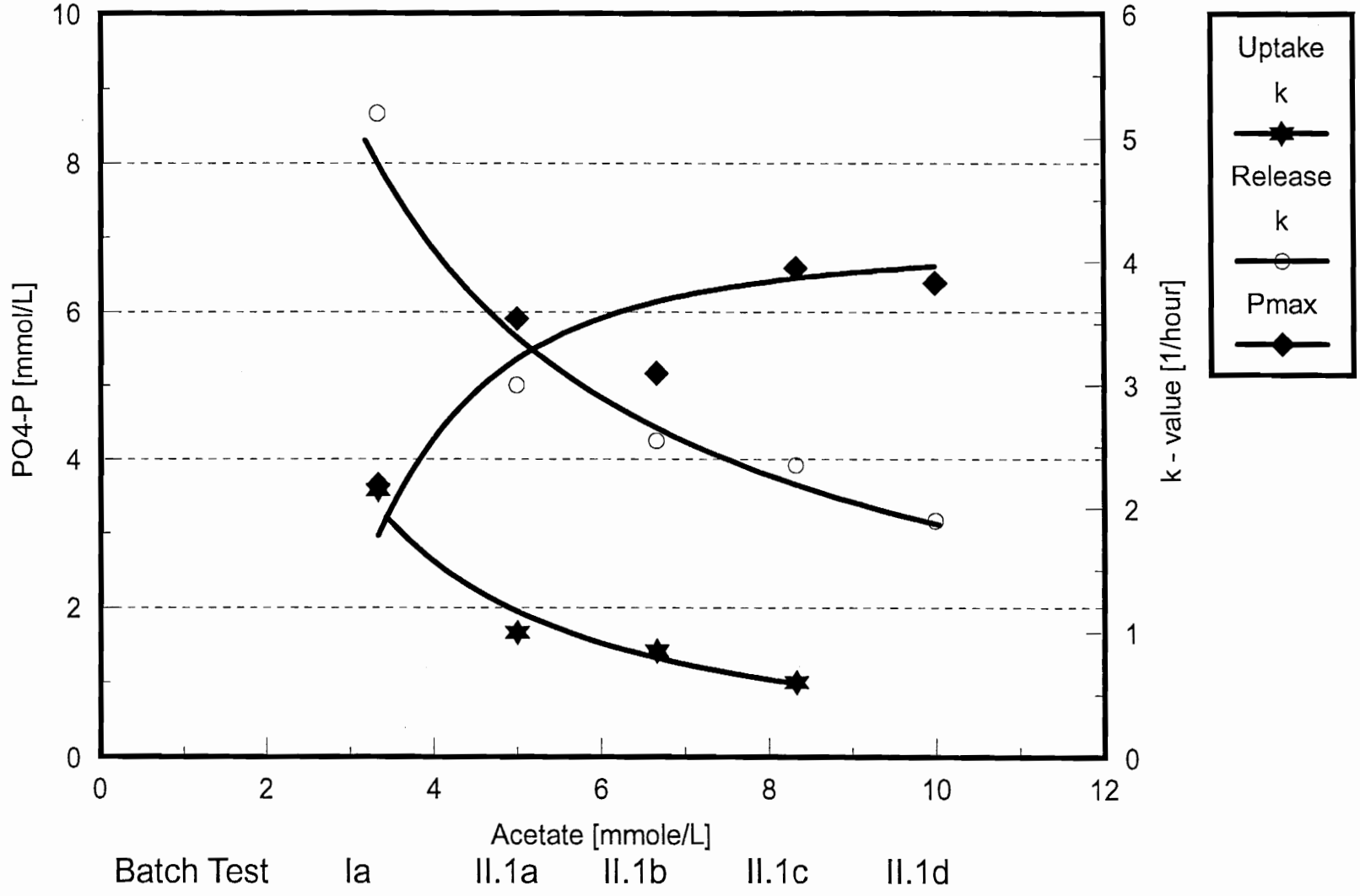

Figure 5.7 Kinetic Parameters for the Results of the Batch Tests for System II

5.2 Discussion of the Results of the Batch Tests with System II and Increasing Acetate Concentration

It is important to keep in mind that the MLSS concentration of System II stayed roughly the same. As a result, the ratio of acetate to biomass increased with higher acetate feeds in the batch tests. Therefore, direct comparisons of the results for System II and System I are not reasonable.

Judging from the Figures 4.16 and 4.17, it is evident that with higher acetate concentrations the biological phosphorus removal process slows down, thus supporting the observation by Randall and Chapin (1994). The picture is clearer if the kinetic parameters are plotted versus the acetate feed (Figure 5.7). On one hand, the values for maximal phosphorus release are asymptotically approaching an absolute maximum of about 200 mg/L, thus supporting the "maximum phosphorus storage capability" theory (Reddy, 1991). Accordingly, the kinetic coefficients asymptotically approach a minimum. When this information is extrapolated, the logical result is that:

- there is an absolute minimum reaction rate coefficient for the release and uptake of phosphorus, and
- this minimum reaction rate coefficient is reached at high acetate concentration, but the rate determining step involved is dependent upon the average acetate concentration in the feed at steady state but independent of the actual acetate concentration.

Regarding a theory for the cause of the apparent breakdown of the biological phosphorus removal at high acetate concentration, a general toxic effect of the acetate on bacteria can be ruled out, since the nitrification rate is apparently not influenced by the acetate (Figure 4.21).

5.3 Discussion of the Results of the Batch Tests with System II and varying Nutrient Concentrations

Various combinations of nutrients were tried to see if they had an influence on the phenomenon of acetate-induced breakdown of biological phosphorus removal. The test with the addition of acetate during aeration (Figure 4.23) confirmed other observations (Yall *et al.*, 1970; Mostert *et al.*, 1988; Hart, 1994; see chapters 2.3.4 and 2.3.5) that phosphorus is released in the aerobic zone if the right substrate is present, but apparently this will only slow down the uptake, not cause it to break down. Increasing the calcium concentration, however, improved the phosphorus uptake drastically (Figures 4.24 and 4.27). Apparently the calcium helped to reduce the phosphorus levels in the mixed liquor. The curve for the phosphorus concentrations under these high - calcium conditions indicate a primarily biological process. In addition to that, precipitation experiments were conducted to rule out calcium phosphate precipitation as the sole cause for the improved behavior of the biological phosphorus removal process. The results (Figure 4.28) show clearly that precipitation could not cause the removal of 200 mg/L of phosphorus from the solution. However, they show that calcium precipitation occurs very fast. Based on these observation and observations regarding phosphorus precipitation (see 2.3.6), the following chain of hypotheses for acetate-induced breakdown of biological phosphorus removal is proposed:

- Calcium is essential in small amounts for the BPR bacteria during uptake.
- When the calcium or the phosphorus concentration is increased, the amount of precipitation is increased (law of chemical equilibrium).
- Precipitation and bacteria are competing for calcium.
- The precipitation occurs inside or near to the cell of biological phosphorus removing bacteria. This assumption is supported by the observations of Arvin and Kristensen (1983) and Bark *et al.* (1992) (see chapter 2.3.6).
- If a very high phosphorus release occurs, calcium phosphate precipitation makes calcium unavailable specifically to the poly-P bacteria.

- The calcium deprived bacteria are subsequently unable to take up phosphorus.

This hypothesis would not only explain the observations made in this study, but also the observations by Pattarkine (1991), who found during batch tests that the ratio of calcium uptake to phosphorus uptake is higher than the ratio of calcium release to phosphorus release (Figure 2.2).

6. Conclusions

The objectives of this study, as listed in chapter 1, were mostly met:

- Two practically identical systems with synthetic wastewater as feed were successfully set up.
- Unlike the experiments reported by Randall and Chapin (1994), it was not possible to cause an acetate - induced breakdown of a system that could adjust its MLSS according to the load; however, acetate in high concentrations inhibited biological phosphorus removal during batch tests.
- The phosphorus release and uptake could be described by first order kinetics, and the rate coefficients decreased with higher acetate loads.
- The inhibiting effect of high acetate concentrations could be countered by increasing the calcium in the feed.

Furthermore, two hypotheses are proposed:

- The decrease of the rate coefficient with increasing acetate concentrations is caused by a transport step across the cell membrane or inside the bacterial floc that limits the flow of chemicals.
- Calcium is an essential cation for the uptake phase of the BPR process. At high acetate concentrations with ensuing rapid release of phosphorus in high concentrations, competing phosphorus precipitation can reduce the amount of calcium available to the poly-P bacteria and thus reduce the rate of phosphorus uptake.

The results of this study are significant for engineers in two ways:

- The observation of the decrease of the rate coefficients for phosphorus release and uptake at higher acetate concentrations means that reactors in series are not as effective as they would be if the coefficients were constant. Nevertheless, this observation does not allow the conclusion that reactors in series are less effective than a single reactor.
- Acetate in high concentrations may cause breakdown of biological phosphorus removal. Therefore it may be advisable to split up the feed and reduce the amount of acetate by aeration before using it for biological phosphorus removal, as shown in Figure 6.1. However, this does not address the possible problem of growth of filamentous bacteria.

The following points warrant further research:

- To investigate the behavior of calcium and magnesium during batch tests with poly-P bacteria at high acetate concentrations. No calcium measurements were conducted during this study, and due to time constraints the role of magnesium during phosphorus release and uptake was not investigated at all.
- To test methods to counter the problem of acetate-induced breakdown of biological phosphorus removal.

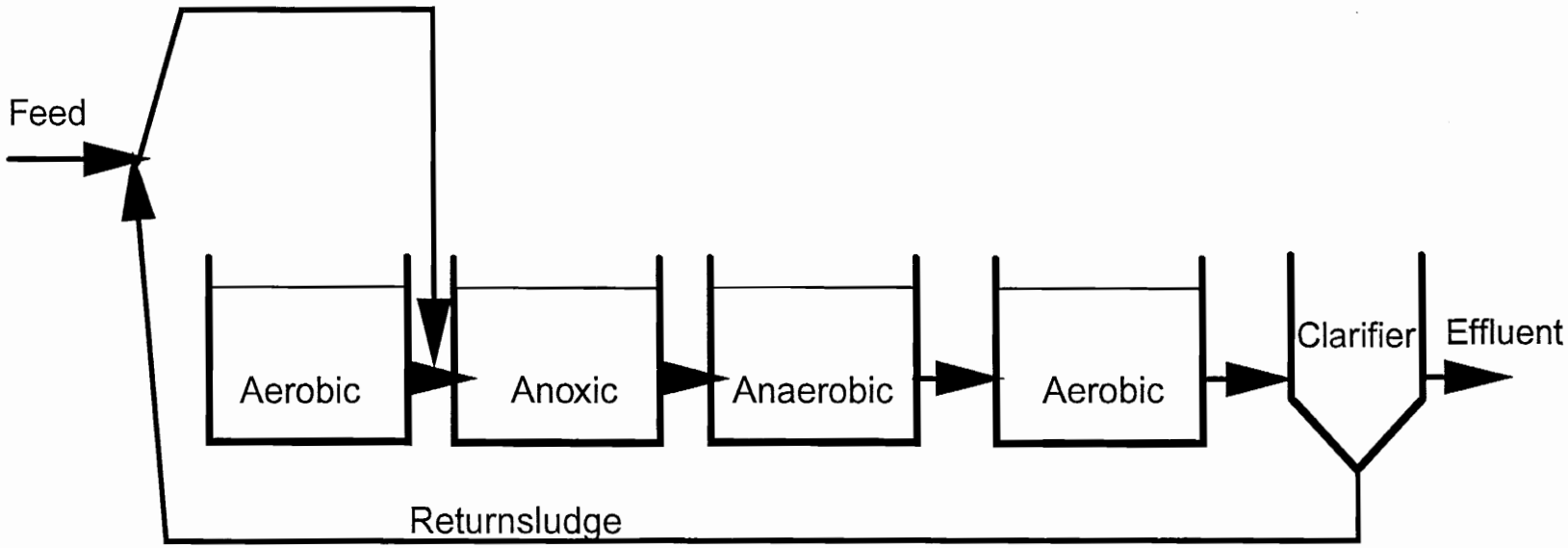

Figure 6.1 Proposed Treatment System for High Acetate Concentrations

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8. Appendix

	Sys. I Aerobic Tank			Sys. I Effluent			Sys. II Aerobic Tank			Sys. II Effluent		
Date	MLSS [g/l]	MLVSS [g/l]	% VSS	MLSS [mg/l]	MLVSS [mg/l]	% VSS	MLSS [g/l]	MLVSS [g/l]	% VSS	MLSS [mg/l]	MLVSS [mg/l]	% VSS
05/25/94	1.86	1.34	72%	44.50	34.0	76%	4.93	3.50	71%	75.60	58.0	77%
05/26/94	2.94	2.11	72%	30.72	23.7	77%	4.53	3.24	72%	78.40	60.0	77%
05/27/94	3.76	2.47	66%	29.07	22.2	76%	3.83	2.54	66%	71.88	53.0	74%
05/28/94	3.35	2.13	64%	32.41	24.8	77%	3.74	2.44	65%	63.89	47.9	75%
05/29/94	3.49	2.39	69%	25.40	18.6	73%	4.07	2.83	70%	62.09	47.2	76%
05/30/94	3.38	2.36	70%	21.22	16.3	77%	3.88	2.72	70%	49.70	38.2	77%
05/31/94	3.26	2.31	71%	23.69	18.7	79%	3.94	2.77	70%	44.38	33.8	76%
06/01/94	3.30	2.34	71%	14.57	11.3	78%	4.00	2.81	70%	39.43	31.0	79%
06/02/94	3.38	2.36	70%	16.87	12.1	72%	3.76	2.61	69%	30.36	22.9	75%
06/03/94	3.86	2.69	70%	27.09	21.0	77%	3.14	2.21	70%	43.53	32.6	75%
06/04/94	3.57	2.54	71%	23.00	19.0	83%	3.84	2.72	71%	50.67	39.1	77%
06/05/94	3.45	2.47	72%	21.58	17.6	82%	3.76	2.68	71%	53.65	41.4	77%
06/06/94	4.00	2.56	64%	20.56	16.7	81%	3.84	2.40	63%	33.96	25.8	76%
06/07/94	4.37	2.75	63%	22.59	17.2	76%	3.62	2.27	63%	25.11	18.6	74%
06/08/94	4.91	3.03	62%	20.77	16.1	77%	3.63	2.29	63%	27.96	21.4	77%
06/09/94	5.57	3.43	62%	29.69	23.7	80%	3.49	2.19	63%	23.46	19.1	82%
06/10/94	6.89	4.83	70%	29.40	21.7	74%	3.67	2.60	71%	20.75	14.5	70%
06/11/94	6.46	4.35	67%	23.59	18.5	79%	3.87	2.63	68%	22.34	18.0	80%
06/12/94	6.33	4.24	67%	19.80	14.5	73%	4.00	2.75	69%	29.96	21.3	71%
06/13/94	7.03	4.67	66%	27.36	21.1	77%	3.81	2.58	68%	51.41	38.1	74%
06/14/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
06/15/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
06/16/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
06/17/94	6.72	4.42	66%	37.37	25.5	68%	3.68	2.48	67%	19.67	14.3	73%
06/18/94	6.45	4.34	67%	51.63	36.0	70%	3.90	2.71	69%	42.31	29.8	70%
06/19/94	5.72	3.80	66%	13.06	10.2	78%	3.72	2.60	70%	34.62	25.3	73%
06/20/94	5.71	3.76	66%	56.77	37.8	67%	3.70	2.63	71%	55.67	40.6	73%
06/21/94	6.22	4.07	65%	113.62	74.6	66%	3.55	2.44	69%	49.29	36.6	74%
06/22/94	6.10	4.05	66%	34.67	25.9	75%	3.56	2.51	70%	62.75	44.5	71%
06/23/94	6.41	4.40	69%	36.95	26.3	71%	3.88	2.80	72%	35.38	25.4	72%
06/24/94	6.49	4.47	69%	23.55	17.0	72%	4.07	2.94	72%	33.06	24.4	74%
06/25/94	5.76	3.96	69%	20.98	15.1	72%	3.52	2.52	72%	53.25	38.0	71%
06/26/94	5.26	3.63	69%	11.54	9.3	81%	3.65	2.61	71%	43.52	32.6	75%
06/27/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
06/28/94	4.70	3.21	68%	17.74	12.9	73%	2.97	2.11	71%	42.45	31.0	73%
06/29/94	5.20	2.74	53%	30.94	22.2	72%	3.63	2.40	66%	61.60	43.8	71%
06/30/94	5.00	3.57	71%	18.84	13.8	73%	3.42	2.31	68%	41.43	29.8	72%
07/01/94	4.75	2.87	60%	268.90	164.9	61%	3.21	2.02	63%	37.20	27.4	74%
07/02/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/03/94	4.60	3.23	70%	65.32	45.4	69%	3.36	2.43	72%	43.12	31.2	72%
07/04/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/05/94	4.77	3.34	70%	19.38	13.7	71%	3.65	2.59	71%	54.27	39.7	73%
07/06/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/07/94	6.48	4.48	69%	34.68	25.4	73%	3.43	2.48	72%	39.16	28.7	73%
07/08/94	6.84	4.74	69%	30.46	21.4	70%	3.57	2.53	71%	53.42	37.3	70%
07/09/94	5.92	4.70	79%	30.49	21.7	71%	3.30	2.33	71%	50.00	35.5	71%
07/10/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/11/94	8.06	5.51	68%	42.57	31.1	73%	3.99	2.79	70%	92.84	66.3	71%
07/12/94	7.73	5.31	69%	32.20	22.6	70%	3.41	2.45	72%	48.50	36.0	74%
07/13/94	7.76	5.36	69%	35.20	26.3	75%	2.99	2.17	73%	35.14	26.8	76%
07/14/94	8.00	5.43	68%	28.54	20.7	73%	2.81	2.04	73%	27.50	20.6	75%
07/15/94	7.75	5.21	67%	43.15	30.7	71%	2.74	2.00	73%	60.50	45.2	75%
07/16/94	7.84	5.18	66%	32.92	23.6	72%	2.93	2.11	72%	40.70	29.2	72%
07/17/94	8.33	5.69	68%	43.15	29.0	67%	3.31	2.38	72%	39.20	28.1	72%
07/18/94	7.69	5.26	68%	27.40	19.0	69%	3.45	2.44	71%	34.34	24.3	71%
07/19/94	7.38	4.99	68%	16.87	12.3	73%	3.07	2.19	71%	39.12	27.7	71%
07/20/94	7.60	5.04	66%	28.23	20.6	73%	3.36	2.36	70%	50.46	35.6	71%
07/21/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/22/94	9.00	6.01	67%	34.88	25.6	73%	4.41	3.11	71%	48.01	34.2	71%
07/23/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/24/94	9.61	6.37	66%	60.42	45.9	76%	3.52	2.45	70%	53.71	39.5	74%
07/25/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
07/26/94	9.68	6.49	67%	46.72	33.6	72%	3.13	2.23	71%	54.02	38.8	72%
07/27/94	10.09	6.64	66%	85.59	42.8	50%	3.56	2.48	70%	57.59	39.3	68%
07/28/94	9.67	6.38	66%	64.29	46.1	72%	3.30	2.33	71%	62.80	46.1	73%
07/29/94	9.26	6.08	66%	45.89	34.3	75%	3.41	2.40	70%	70.72	51.4	73%
07/30/94	8.72	5.71	66%	55.02	40.4	73%	2.86	2.01	70%	86.23	62.6	73%
07/31/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
08/01/94	8.57	5.67	66%	52.37	38.4	73%	2.44	1.70	70%	101.36	70.7	70%
08/02/94	9.15	6.07	66%	46.61	35.5	76%	2.79	1.93	69%	107.92	75.7	70%

	Sys. I Aerobic Tank			Sys. I Effluent			Sys. II Aerobic Tank			Sys. II Effluent		
Date	MLSS [g/l]	MLVSS [g/l]	% VSS	MLSS [mg/l]	MLVSS [mg/l]	% VSS	MLSS [g/l]	MLVSS [g/l]	% VSS	MLSS [mg/l]	MLVSS [mg/l]	% VSS
08/03/94	8.17	5.16	63%	NA	NA	NA	3.06	2.13	70%	NA	NA	NA
08/04/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
08/05/94	8.74	5.57	64%	NA	NA	NA	3.56	2.48	70%	NA	NA	NA
08/06/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
08/07/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
08/08/94	10.16	6.54	64%	42.21	31.2	74%	2.97	2.06	70%	39.46	29.5	75%
08/09/94	10.18	6.57	65%	37.69	28.2	75%	2.96	2.19	74%	42.27	30.4	72%
08/10/94	9.80	6.36	65%	NA	NA	NA	2.41	1.70	70%	NA	NA	NA
08/11/94	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
08/12/94	10.21	6.47	63%	NA	NA	NA	3.49	2.37	68%	NA	NA	NA

Date	05/28/94		ml		ml
		Cold Blank	6.00	Hot Blank	6.11
Normal. FAS	0.050	Cold Blank	6.10	Hot Blank	6.12
Sample-Size [ml]	5.00	Average	6.05	Average	6.115

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	1.62	1.0	357	Feed (filtered)	2.10	1	319
Feed (unfiltered)		1.0		Feed (unfiltered)		1	
Anaerobic Tank In	4.68	1.0	114	Anaerobic Tank In	4.51	1	127
Anaerobic Tank Out	5.48	1.0	50	Anaerobic Tank Out	5.32	1	63
Aerobic Tank	5.70	1.0	33	Aerobic Tank	5.41	1	56
Clarifier	5.70	1.0	33	Clarifier	5.40	1	57

Date	05/31/94		ml		ml
		Cold Blank	6.20	Hot Blank	6.38
Normal. FAS	0.048	Cold Blank	6.20	Hot Blank	6.10
Sample-Size [ml]	5.00	Average	6.2	Average	6.24

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	2.47	1.0	289	Feed (filtered)	1.76	1	346
Feed (unfiltered)		1.0		Feed (unfiltered)		1	
Anaerobic Tank In	4.72	1.0	111	Anaerobic Tank In	4.43	1	134
Anaerobic Tank Out	5.60	1.0	41	Anaerobic Tank Out	5.28	1	66
Aerobic Tank	5.65	1.0	37	Aerobic Tank	5.63	1	38
Clarifier	5.62	1.0	39	Clarifier	5.75	1	29

Date	06/04/94		ml		ml
		Cold Blank	5.92	Hot Blank	5.95
Normal. FAS	0.051	Cold Blank	5.92	Hot Blank	5.96
Sample-Size [ml]	5.00	Average	5.92	Average	5.955

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	1.51	1.0	365	Feed (filtered)	1.43	1	372
Feed (unfiltered)		1.0		Feed (unfiltered)		1	
Anaerobic Tank In	4.58	1.0	122	Anaerobic Tank In	4.60	1	120
Anaerobic Tank Out	5.05	1.0	84	Anaerobic Tank Out	4.95	1	92
Aerobic Tank	5.70	1.0	33	Aerobic Tank	5.46	1	52
Clarifier	5.90	1.0	17	Clarifier	5.48	1	50

Date	06/10/94		ml		ml
		Cold Blank	6.05	Hot Blank	6.05
Normal. FAS	0.050	Cold Blank	6.05	Hot Blank	6.00
Sample-Size [ml]	5.00	Average	6.05	Average	6.025

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	1.65	1.0	354	Feed (filtered)		1	
Feed (unfiltered)		1.0		Feed (unfiltered)		1	
Anaerobic Tank In	4.70	1.0	112	Anaerobic Tank In		1	
Anaerobic Tank Out	5.15	1.0	77	Anaerobic Tank Out		1	
Aerobic Tank	5.55	1.0	45	Aerobic Tank		1	
Clarifier	5.55	1.0	45	Clarifier		1	

Date	06/13/94			ml		ml	
			Cold Blank	6.18	Hot Blank		6.14
Normal. FAS	0.049		Cold Blank		Hot Blank		
Sample-Size [ml]	5.00		Average	6.18	Average		6.14

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	0.67	1.0	432	Feed (filtered)		1	
	3.09	2.0	480				
Feed (unfiltered)	0.53	1.0	443	Feed (unfiltered)		1	
	3.40	2.0	431				
Anaerobic Tank In	4.80	1.0	104	Anaerobic Tank In		1	
Anaerobic Tank Out	5.52	1.0	47	Anaerobic Tank Out		1	
Aerobic Tank	6.00	1.0	9	Aerobic Tank		1	
Clarifier	5.98	1.0	11	Clarifier		1	

Date	06/18/94			ml		ml	
			Cold Blank	6.05	Hot Blank		6.08
Normal. FAS	0.050		Cold Blank	6.05	Hot Blank		
Sample-Size [ml]	5.00		Average	6.05	Average		6.08

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	0.70	1.0	430	Feed (filtered)		1	
	3.15	2.0	470				
Feed (unfiltered)	0.40	1.0	453	Feed (unfiltered)		1	
	3.00	2.0	494				
Anaerobic Tank In	4.81	1.0	104	Anaerobic Tank In		1	
Anaerobic Tank Out	5.20	1.0	73	Anaerobic Tank Out		1	
Aerobic Tank	5.60	1.0	41	Aerobic Tank		1	
Clarifier	5.58	1.0	42	Clarifier		1	

Date	06/23/94		ml		ml
		Cold Blank	6.06	Hot Blank	6.00
Normal. FAS	0.050	Cold Blank	6.06	Hot Blank	6.06
Sample-Size [ml]	5.00	Average	6.06	Average	6.03

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	2.90	2.0	510	Feed (filtered)		1	
Feed (unfiltered)	2.85	2.0	518	Feed (unfiltered)		1	
Anaerobic Tank In	4.65	1.0	116	Anaerobic Tank In		1	
Anaerobic Tank Out	5.28	1.0	66	Anaerobic Tank Out		1	
Aerobic Tank	5.60	1.0	41	Aerobic Tank		1	
Clarifier	5.65	1.0	37	Clarifier		1	

Date	07/14/94		ml		ml
		Cold Blank	6.10	Hot Blank	6.08
Normal. FAS	0.049	Cold Blank		Hot Blank	6.00
Sample-Size [ml]	5.00	Average	6.1	Average	6.04

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	2.38	2.0	593	Feed (filtered)		1	
Feed (unfiltered)	2.41	2.0	588	Feed (unfiltered)		1	
Anaerobic Tank In	4.73	1.0	110	Anaerobic Tank In		1	
Anaerobic Tank Out	5.48	1.0	50	Anaerobic Tank Out		1	
Aerobic Tank	5.75	1.0	29	Aerobic Tank		1	
Clarifier	5.77	1.0	27	Clarifier		1	

			ml		ml
Date	07/16/94	Cold Blank	6.11	Hot Blank	6.10
		Cold Blank	6.10	Hot Blank	6.08
Normal. FAS	0.049				
Sample-Size [ml]	5.00	Average	6.105	Average	6.09

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	2.45	2.0	582	Feed (filtered)		1	
Feed (unfiltered)	2.42	2.0	586	Feed (unfiltered)		1	
Anaerobic Tank In	4.90	1.0	96	Anaerobic Tank In		1	
Anaerobic Tank Out	5.45	1.0	53	Anaerobic Tank Out		1	
Aerobic Tank	5.68	1.0	35	Aerobic Tank		1	
Clarifier	5.75	1.0	29	Clarifier		1	

			ml		ml
Date	07/18/94	Cold Blank	6.09	Hot Blank	6.00
		Cold Blank	6.06	Hot Blank	6.07
Normal. FAS	0.049				
Sample-Size [ml]	5.00	Average	6.075	Average	6.035

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	3.54	2.0	409	Feed (filtered)		1	
Feed (unfiltered)	3.43	2.0	426	Feed (unfiltered)		1	
Anaerobic Tank In	5.40	1.0	57	Anaerobic Tank In		1	
Anaerobic Tank Out	5.60	1.0	41	Anaerobic Tank Out		1	
Aerobic Tank	5.65	1.0	37	Aerobic Tank		1	
Clarifier	5.70	1.0	33	Clarifier		1	

			ml		ml
Date	07/28/94	Cold Blank	6.10	Hot Blank	6.00
		Cold Blank		Hot Blank	6.06
Normal. FAS	0.049				
Sample-Size [ml]	5.00	Average	6.1	Average	6.03

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)		2.0		Feed (filtered)		1	
Feed (unfiltered)	1.20	2.0	780	Feed (unfiltered)		1	
Anaerobic Tank In	3.82	1.0	182	Anaerobic Tank In		1	
Anaerobic Tank Out	5.32	1.0	63	Anaerobic Tank Out		1	
Aerobic Tank	5.50	1.0	49	Aerobic Tank		1	
Clarifier	5.80	1.0	25	Clarifier		1	

			ml		ml
Date	07/30/94	Cold Blank	6.00	Hot Blank	6.07
		Cold Blank	6.08	Hot Blank	6.02
Normal. FAS	0.050				
Sample-Size [ml]	5.00	Average	6.04	Average	6.045

System I	ml		COD mg O2/L	System II	ml		COD mg O2/L
Feed (filtered)	2.53	3.3	948	Feed (filtered)		1	
Feed (unfiltered)	2.59	3.3	932	Feed (unfiltered)		1	
Anaerobic Tank In	4.19	1.0	153	Anaerobic Tank In		1	
Anaerobic Tank Out	5.01	1.0	88	Anaerobic Tank Out		1	
Aerobic Tank	5.32	1.0	63	Aerobic Tank		1	
Clarifier	5.36	1.0	60	Clarifier		1	

System 2
Date; from: 05/28/94 to: 06/04/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	200	3.33
	Nutr. Broth	180.0	-
	COD	393	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	25.4405	0.65069
	PO4-P	10.0185	0.32534
	NH4-N	5.45521	0.38947

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
05/28/94	3.74	2.44	65%
05/31/94	3.94	2.77	70%
06/04/94	3.84	2.72	71%
Average	3.84	2.64	69%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	319	127	63	56	57
05/31/94	346	134	66	38	29
06/04/94	372	120	92	52	50
Average	346	127	74	49	45

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94					
05/31/94					
06/04/94					
Average	ERR	ERR	ERR	ERR	ERR

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
05/28/94		2.04	2.54	0.92	
05/31/94		2.24	2.61	1.92	
06/04/94		2.73	3.20	0.86	
Average		2.34	2.78	1.23	

SYSTEM 2

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94		76.3	95.0	34.4	
05/31/94		88.3	102.8	75.6	
06/04/94		104.8	122.9	33.0	
Average		89.8	106.9	47.7	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	7.8	41.2	65.1	0.7	3.1
05/31/94	8.9	41.2	78.8	0.7	0.3
06/04/94	8.3	39.8	72.2	1.1	0.4
Average	8.3	40.7	72.0	0.8	1.3

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0.0	0.2	0.0	10.3	9.0
05/31/94	0.0	0.2	0.0	9.3	9.5
06/04/94	0.3	0.3	0.0	9.1	8.3
Average	0.1	0.2	0.0	9.6	8.9

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0	0.1	0	0.1	0
05/31/94	0.5	0.1	0	0.2	0.1
06/04/94	0.2	0.1	0	0.2	0.1
Average	0.2	0.1	0.0	0.2	0.1

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0.0	0.9	0.0	35.5	30.8
05/31/94	1.1	0.9	0.0	32.3	32.8
06/04/94	1.5	1.3	0.0	31.6	28.7
Average	0.9	1.0	0.0	33.2	30.8

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	346	ERR	0	8.3	0.1	0.2	0.9
Anae In	127	ERR	89.795	40.7	0.2	0.1	1.0
Anae Out	74	ERR	106.903	72.0	0.0	0.0	0.0
Aerob	49	ERR	47.693	0.8	9.6	0.2	33.2
Clarifier	45	ERR	0	1.3	8.9	0.1	30.8

System 1
Date; from: 05/28/94 to: 06/04/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	200	3.33
	Nutr. Broth	180.0	-
	COD	393	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	25.4405	0.65069
	PO4-P	10.0185	0.32534
	NH4-N	5.45521	0.38947

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
05/28/94	3.35	2.13	64%
06/04/94	3.57	2.54	71%
Average	3.46	2.34	67%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	357	114	50	33	33
06/04/94	365	122	84	33	17
**********					
Average	361	118	67	33	25

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94					
06/04/94					
**********					
Average	ERR	ERR	ERR	ERR	ERR

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
05/28/94		3.81	4.00	1.12	
06/04/94		3.05	3.46	0.91	
**********					
Average		3.43	3.73	1.01	

SYSTEM 1

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94		127.6	134.0	37.5	
06/04/94		108.9	123.5	32.3	
**********					
Average		118.3	128.8	34.9	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	8.5	39.2	80.6	0.9	0.2
06/04/94	8.7	41.1	70.9	1.4	0.7
**********					
Average	8.6	40.2	75.8	1.2	0.5

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0.0	0.3	0.1	10.0	9.7
06/04/94	0.0	0.5	0.1	8.6	8.0
**********					
Average	0.0	0.4	0.1	9.3	8.9

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0	0.1	0	0.2	0
06/04/94	0.5	0.2	0	0.2	0.2
**********					
Average	0.3	0.2	0.0	0.2	0.1

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
05/28/94	0.0	1.3	0.3	34.7	33.2
06/04/94	1.1	2.2	0.3	29.9	27.9
**********					
Average	0.6	1.7	0.3	32.3	30.6

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	361	ERR	0	8.6	0.0	0.3	0.6
Anae In	118	ERR	118.260	40.2	0.4	0.2	1.7
Anae Out	67	ERR	128.761	75.8	0.1	0.0	0.3
Aerob	33	ERR	34.914	1.2	9.3	0.2	32.3
Clarifier	25	ERR	0	0.5	8.9	0.1	30.6

System 1
Date; from: 06/10/94 to: 06/18/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	300	5.00
	Nutr. Broth	180.0	-
	COD	500	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	38.2	0.98
	PO4-P	15	0.49
	NH4-N	8.2	0.58

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
06/10/94	6.89	4.83	70%
06/13/94	7.03	4.67	66%
06/18/94	6.45	4.34	67%
Average	6.79	4.61	68%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	354	112	77	45	45
06/13/94	437	104	47	9	11
06/18/94	473.5	104	73	41	42
Average	422	107	66	32	33

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	178	14	0	0	0
06/13/94					
06/18/94					
Average	178	14	0	0	0

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
06/10/94		1.19	1.15	0.58	
06/13/94		0.73	0.89	0.22	
06/18/94		1.38	1.43	0.38	
Average		1.10	1.16	0.39	

SYSTEM 1

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94		82.0	79.2	40.0	
06/13/94		51.3	62.6	15.5	
06/18/94		89.0	92.2	24.5	
Average		74.1	78.0	26.6	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	12.1	42.1	81.6	0.4	0.2
06/13/94	12.7	68.3	106.2	0.4	0.3
06/18/94	12.3	74.6	102.6	0.5	0.3
Average	12.4	61.7	96.8	0.4	0.3

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	0.0	0.3	0.2	9.6	9.2
06/13/94	0.3	0.1	0.0	9.1	9.0
06/18/94	0.0	0.1	0.1	8.7	8.4
Average	0.1	0.2	0.1	9.1	8.9

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	0	0.1	0	0.1	0.2
06/13/94	0.4	0	0	0	0
06/18/94	0	0	0	0	0
Average	0.1	0.0	0.0	0.0	0.1

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
06/10/94	0.0	1.3	0.7	33.1	32.0
06/13/94	1.9	0.3	0.0	31.2	30.8
06/18/94	0.0	0.3	0.2	29.8	28.8
Average	0.6	0.6	0.3	31.4	30.5

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	422	178	0	12.4	0.1	0.1	0.6
Anae In	107	14	74.107	61.7	0.2	0.0	0.6
Anae Out	66	0	78.012	96.8	0.1	0.0	0.3
Aerob	32	0	26.646	0.4	9.1	0.0	31.4
Clarifier	33	0	0	0.3	8.9	0.1	30.5

System 1
Date; from: 07/14/94 to: 07/19/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	400	3.33
	Nutr. Broth	180.0	-
	COD	606	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	50.9	1.3
	PO4-P	20	0.65
	NH4-N	10.9	0.78

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
07/14/94	8.00	5.43	68%
07/16/94	7.84	5.18	66%
07/18/94	7.69	5.26	68%
Average	7.84	5.29	67%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	588	110	50	29	27
07/16/94	586	96	53	35	29
07/18/94	426	57	41	37	33
Average	533	88	48	34	30

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	401	48	1	4	4
07/16/94	420	30	2	0	5
07/18/94	267	3	0	0	0
Average	363	27	1	1	3

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
07/14/94		1.73	1.59	0.50	
07/16/94		1.54	1.52	0.39	
07/18/94		1.93	1.33	0.44	
Average		1.73	1.48	0.44	

SYSTEM 1

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94		138.4	127.2	40.0	
07/16/94		120.7	119.2	30.6	
07/18/94		148.4	102.3	33.8	
Average		135.9	116.2	34.8	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	17.8	105.3	133.3	0.4	0.1
07/16/94	18.0	109.2	152.1	0.3	0.1
07/18/94	15.5	88.2	105.7	1.1	0.1
Average	17.1	100.9	130.4	0.6	0.1

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	0.5	0.1	0.0	7.5	7.1
07/16/94	0.5	0.0	0.1	9.3	8.8
07/18/94	0.0	0.2	0.0	8.3	8.2
Average	0.3	0.1	0.0	8.4	8.0

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	0	0	0	0.2	0
07/16/94	0	0	0	0	0
07/18/94	0	0	0	0	0
Average	0.0	0.0	0.0	0.1	0.0

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/14/94	1.7	0.2	0.0	26.2	24.3
07/16/94	1.7	0.0	0.3	31.9	30.2
07/18/94	0.0	0.7	0.0	28.4	28.1
Average	1.1	0.3	0.1	28.8	27.5

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	533	363	0	17.1	0.3	0.0	1.1
Anae In	88	27	135.851	100.9	0.1	0.0	0.3
Anae Out	48	1	116.215	130.4	0.0	0.0	0.1
Aerob	34	1	34.804	0.6	8.4	0.1	28.8
Clarifier	30	3	0	0.1	8.0	0.0	27.5

System 1
Date; from: 07/21/94 to: 08/03/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	600	3.33
	Nutr. Broth	180.0	-
	COD	820	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	76.4	79.4
	PO4-P	77.4	80.4
	NH4-N	78.4	81.4

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
07/28/94	9.67	6.38	66%
07/30/94	8.72	5.71	65%
08/01/94	8.57	5.67	66%
Average	8.99	5.92	66%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	780	182	63	49	25
07/30/94	932	153	88	63	60
08/01/94					
Average	856	168	76	56	43

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	636	106	0	1	7
07/30/94	659	69	0	2	2
08/01/94	622	41	0	1	3
Average	639	72	0	1	4

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
07/28/94		2.49	2.93	0.93	
07/30/94		2.95	2.63	0.79	
08/01/94		2.15	2.60	0.71	
Average		2.53	2.72	0.81	

SYSTEM 1

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94		240.8	283.3	89.9	
07/30/94		257.2	229.3	68.9	
08/01/94		184.3	222.8	60.8	
Average		227.4	245.2	73.2	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	26.7	120.3	197.8	0.6	0.1
07/30/94	28.2	166.6	220.3	0.5	0.1
08/01/94	25.4	170.2	205.2	0.2	0.3
Average	26.8	152.4	207.8	0.4	0.2

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	0.5	0.1	0.2	7.3	7.4
07/30/94	0.5	0.1	0.1	8.1	7.6
08/01/94	0.0	0.0	0.1	6.9	5.9
Average	0.3	0.1	0.1	7.4	7.0

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	0	0.1	0	0	0
07/30/94	0	0	0	0	0
08/01/94	0	0	0	0	0
Average	0.0	0.0	0.0	0.0	0.0

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
07/28/94	1.7	0.6	0.7	25.0	25.4
07/30/94	1.7	0.3	0.3	27.8	26.0
08/01/94	0.0	0.0	0.3	23.6	20.2
Average	1.1	0.3	0.5	25.5	23.9

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	856	639	0	26.8	0.3	0.0	1.1
Anae In	168	72	227.426	152.4	0.1	0.0	0.3
Anae Out	76	0	245.162	207.8	0.1	0.0	0.5
Aerob	56	1	73.222	0.4	7.4	0.0	25.5
Clarifier	43	4	0	0.2	7.0	0.0	23.9

System 1
Date; from: 08/04/94 to: 08/12/94

Composition of Feed:		[mg/L]	[mmol/L]
	Acetate	800	3.33
	Nutr. Broth	180.0	-
	COD	1033	-
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	101.8	2.6
	PO4-P	40	1.3
	NH4-N	21.8	1.56

Date	MLSS [g/L]	MLVSS [g/L]	% volatile [%]
08/10/94	9.80	6.36	65%
Average	9.80	6.36	65%

COD Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94					
**********					
**********					
Average	ERR	ERR	ERR	ERR	ERR

Acetate Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94	791	196	8	2	2
**********					
**********					
Average	791	196	8	2	2

PHB/SS % Date	Feed [mg/mg]	Anae In [mg/mg]	Anae Out [mg/mg]	Aerob [mg/mg]	Clarifier [mg/mg]
08/10/94		2.69	3.10	1.20	
**********					
**********					
Average		2.69	3.10	1.20	

SYSTEM 1

PHB Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94		263.6	303.8	117.6	
**********					
**********					
Average		263.6	303.8	117.6	

PO4-P Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94	35.8	142.9	255.0	1.0	0.4
**********					
**********					
Average	35.8	142.9	255.0	1.0	0.4

NO3-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94	0.6	0.1	0.1	5.0	4.9
**********					
**********					
Average	0.6	0.1	0.1	5.0	4.9

NO2-N Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94	0	0	0	0	0
**********					
**********					
Average	0.0	0.0	0.0	0.0	0.0

NOx-O Date	Feed [mg/L]	Anae In [mg/L]	Anae Out [mg/L]	Aerob [mg/L]	Clarifier [mg/L]
08/10/94	2.1	0.3	0.3	17.1	16.8
**********					
**********					
Average	2.1	0.3	0.3	17.1	16.8

Averages	COD [mg/L]	Acetate [mg/L]	PHB [mg/L]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx-O [mg/L]
Feed	ERR	791	0	35.8	0.6	0.0	2.1
Anae In	ERR	196	263.620	142.9	0.1	0.0	0.3
Anae Out	ERR	8	303.800	255.0	0.1	0.0	0.3
Aerob	ERR	2	117.600	1.0	5.0	0.0	17.1
Clarifier	ERR	2	0	0.4	4.9	0.0	16.8

Date 06/30/94

Description System I at steady state with 300 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	300	5.00
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	38.2	0.98
	PO4-P	15	0.50
	NH4-N	8.25	0.59
Misc.	MLSS	[g/L]	5.00
	MLVSS	[g/L]	3.57
	S/X	[mg/mg]	0.08
	tot. P	[mg/L]	426
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	01:31
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	1.8	1.7	0.2	6.3	0.46%	0.65%	0
	2	00:02:00	0.03	15.8	0.9	ND	3.1	0.73%	1.02%	282
	3	00:15:00	0.25	112.1	0.1	ND	0.4	1.79%	2.52%	138
	4	00:30:00	0.50	184.6	0.0	ND	0.0	3.14%	4.42%	23
	5	01:00:00	1.00	198.4	0.1	ND	0.4	2.10%	2.95%	0
	6	01:30:00	1.50	193.9	0.0	ND	0.2	3.20%	4.51%	0
	7	01:45:00	1.75	132.4	0.2	0.3	1.4	2.32%	3.27%	0
	8	02:00:00	2.00	84.8	0.5	0.9	3.6	0.41%	0.57%	0
	9	02:30:00	2.50	30.9	2.2	1.6	11.4	0.32%	0.45%	0
	10	03:00:00	3.00	8.8	4.7	2.1	20.9	1.30%	1.83%	0
	11	03:30:00	3.50	3.5	7.8	2.5	32.4	1.38%	1.95%	0
	12	04:00:00	4.00	1.5	11.1	3.1	45.2	1.15%	1.62%	0
	13	04:30:00	4.50	0.7	14.5	3.5	57.6	1.03%	1.45%	0

Date 07/20/94

Description System I at steady state with 400 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	400	6.66
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	50.9	1.30
	PO4-P	20	0.66
	NH4-N	11	0.78

Misc.	MLSS	[g/L]	7.60
	MLVSS	[g/L]	5.04
	S/X	[mg/mg]	0.08
	tot. P	[mg/L]	641
	PH Start	[-]	8.12
	PH Ende	[-]	8.70
	Start Aerob	[h:m]	01:31
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	2.6	2.8	0.2	10.1	0.31%	0.48%	1
	2	00:01:30	0.03	44.5	0.9	0.0	3.2	0.16%	0.24%	410
	3	00:15:00	0.25	186.9	0.0	0.0	0.0	0.36%	0.54%	186
	4	00:30:00	0.50	297.1	0.0	0.0	0.0	2.61%	3.95%	15
	5	01:00:00	1.00	313.6	0.0	0.3	0.7	1.22%	1.85%	0
	6	01:30:00	1.50	319.7	0.1	0.0	0.2	1.65%	2.50%	0
	7	01:46:00	1.77	215.2	0.2	0.4	1.5	0.87%	1.32%	0
	8	02:01:30	2.03	146.7	0.5	1.0	4.1	0.93%	1.41%	0
	9	02:31:00	2.52	68.2	3.2	1.7	14.6	0.63%	0.96%	0
	10	03:00:00	3.00	30.9	6.3	2.0	26.1	0.67%	1.01%	0
	11	03:30:00	3.50	10.2	10.3	2.2	40.2	1.06%	1.61%	0
	12	04:00:00	4.00	3.1	14.7	2.3	55.6	0.50%	0.76%	0
	13	04:31:00	4.52	0.4	18.8	0.0	64.4	0.33%	0.51%	0
	14	05:00:00	5.00	0.1	20.3	0.0	69.4	0.46%	0.69%	0

Date 08/03/94

Description System I at steady state with 600 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	76.3	1.95
	PO4-P	30	0.98
	NH4-N	16.4	1.17

Misc.	MLSS	[g/L]	8.17
	MLVSS	[g/L]	5.16
	S/X	[mg/mg]	0.12
	tot. P	[mg/L]	913
	PH Start	[-]	8.48
	PH Ende	[-]	8.81
	Start Aerob	[h:m]	01:31
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	8.0	0.0	0.0	0.1	0.24%	0.37%	5
	2	00:01:00	0.02	55.4	0.0	0.0	0.0	0.26%	0.41%	571
	3	00:17:00	0.28	266.6	0.0	0.0	0.0	0.75%	1.19%	280
	4	00:31:00	0.52	365.9	0.0	0.0	0.0	0.99%	1.57%	116
	5	01:00:00	1.00	458.6	0.0	0.0	0.0	1.77%	2.81%	2
	6	01:30:00	1.50	446.9	0.3	0.0	1.1	1.68%	2.66%	2
	7	01:46:00	1.77	320.3	0.3	0.1	1.5	1.52%	2.42%	1
	8	02:01:00	2.02	249.0	1.1	1.3	6.5	1.24%	1.97%	0
	9	02:30:00	2.50	154.7	3.4	2.1	16.4	1.22%	1.94%	0
	10	03:00:00	3.00	104.7	6.2	2.8	27.5	1.02%	1.62%	0
	11	03:30:00	3.50	80.1	7.6	3.5	34.0	1.09%	1.74%	0
	12	04:00:00	4.00	42.8	12.2	3.8	50.7	1.07%	1.70%	0
	13	04:31:00	4.52	16.7	15.6	3.5	61.6	1.74%	2.76%	0
	14	05:00:00	5.00	4.1	18.9	0.0	64.9	0.74%	1.17%	0
	15	05:34:00	5.57	0.2	22.3	0.1	76.6	0.79%	1.26%	0

Date 06/12/94

Description System II at steady state with 200 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	200	3.33
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	25.4	0.65
	PO4-P	10	0.33
	NH4-N	5.5	0.39
Misc.	MLSS	[g/L]	4.00
	MLVSS	[g/L]	2.75
	S/X	[mg/mg]	0.07
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	01:30
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	0.5	8.3	0.5	29.6	0.49%	0.71%	0
	2	00:15:30	0.26	81.6	2.8	0.0	9.6	1.76%	2.55%	91
	3	00:30:30	0.51	110.2	0.0	0.0	0.0	2.42%	3.50%	5
	4	01:01:00	1.02	112.2	0.0	0.0	0.0	2.35%	3.41%	0
	5	01:30:00	1.50	109.9	0.0	0.0	0.0	2.75%	3.99%	0
	6	01:46:30	1.78	68.7	0.9	0.5	4.2	1.63%	2.36%	0
	7	02:15:00	2.25	15.5	3.7	0.8	14.5	1.68%	2.43%	0
	8	02:45:00	2.75	3.1	7.1	1.1	26.8	1.26%	1.83%	0
	9	03:15:00	3.25	1.8	10.5	1.4	39.2			0
	10	03:45:00	3.75	1.2	14.3	1.7	52.9	1.25%	1.81%	0
	11	04:15:00	4.25	0.6	17.7	1.9	65.0			

Date 06/19/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with 400 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	400	6.67
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	50.9	1.30
	PO4-P	15	0.49
	NH4-N	8.2	0.58

Misc.	MLSS	[g/L]	3.72
	MLVSS	[g/L]	2.60
	S/X	[mg/mg]	0.15
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	01:30
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	1.2	5.0	0.6	18.3	0.77%	1.10%	0
	2	00:02:00	0.03	17.5	3.1	1.3	13.6	1.08%	1.54%	389
	3	00:15:00	0.25	63.8	0.2	0.0	0.5	2.25%	3.21%	269
	4	00:30:00	0.50	106.7	0.0	0.0	0.0	3.14%	4.48%	178
	5	01:00:00	1.00	152.4	0.0	0.0	0.0	6.18%	8.83%	69
	6	01:30:00	1.50	158.7	0.0	0.0	0.0	5.27%	7.53%	26
	7	01:45:00	1.75	132.0	0.3	0.6	2.4	4.75%	6.78%	0
	8	02:00:00	2.00	103.4	0.9	1.0	5.2	3.87%	5.53%	0
	9	02:30:00	2.50	69.8	2.5	1.7	12.4	3.47%	4.96%	0
	10	03:01:00	3.02	44.2	4.8	2.3	21.8	3.77%	5.39%	0
	11	03:31:00	3.52	30.4	7.2	3.0	31.4	3.04%	4.34%	0
	12	04:01:00	4.02	21.2	9.7	3.4	41.1	2.73%	3.90%	0
	13	04:30:00	4.50	14.4	12.3	3.8	51.0	2.62%	3.74%	0
	14	05:00:00	5.00	9.4	15.1	4.4	61.6	2.85%	4.07%	0
	15	05:30:00	5.50	5.6	17.7	4.9	71.7	2.41%	3.45%	0
	16	06:00:00	6.00	2.5	20.4	4.9	81.3	2.87%	4.10%	0
	17	06:30:00	6.50	0.8	23.3	2.9	86.4	2.42%	3.45%	0
	18	07:00:00	7.00	0.7	25.9	1.2	91.5	2.19%	3.13%	0

Date 06/24/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with 300 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	300	5.00
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	38.2	0.98
	PO4-P	15	0.49
	NH4-N	8.2	0.58

Misc.	MLSS	[g/L]	4.07
	MLVSS	[g/L]	2.94
	S/X	[mg/mg]	0.10
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	01:32
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	2.4	5.3	0.3	18.9	0.65%	0.91%	7
	2	00:02:00	0.03	30.2	0.0	0.8	1.8	0.69%	0.96%	288
	3	00:16:00	0.27	96.1	0.3	0.5	1.9	1.62%	2.25%	186
	4	00:30:00	0.50	135.0	0.1	0.0	0.3	3.15%	4.37%	119
	5	01:01:00	1.02	177.7	0.0	0.0	0.1	4.19%	5.83%	15
	6	01:30:00	1.50	182.6	0.0	0.0	0.1	3.61%	5.02%	5
	7	01:46:00	1.77	142.5	0.1	0.3	1.1	2.77%	3.84%	0
	8	02:01:00	2.02	108.2	0.3	0.8	2.6	2.70%	3.76%	0
	9	02:30:00	2.50	64.4	1.1	1.8	8.0	2.81%	3.90%	0
	10	03:00:00	3.00	40.9	3.1	2.8	16.8	2.55%	3.54%	0
	11	03:30:00	3.50	27.8	5.3	3.3	25.9	2.44%	3.39%	0
	12	04:00:00	4.00	21.5	7.9	4.0	36.3	2.42%	3.36%	0
	13	04:30:00	4.50	17.0	10.5	4.4	45.9	1.99%	2.76%	0
	14	05:00:00	5.00	12.8	13.1	4.9	56.2	2.22%	3.08%	0
	15	05:30:00	5.50	9.1	15.5	5.4	65.5	2.10%	2.92%	0
	16	06:00:00	6.00	4.9	18.4	5.4	75.5	1.46%	2.03%	0
	17	06:30:00	6.50	0.8	21.5	3.6	82.1	1.74%	2.42%	0
	18	07:00:00	7.00	0.4	24.5	1.2	86.7	1.71%	2.37%	0

Date 07/07/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 600 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	76.3	1.95
	PO4-P	30	0.98
	NH4-N	16.4	1.17
Misc.	MLSS	[g/L]	3.43
	MLVSS	[g/L]	2.48
	S/X	[mg/mg]	0.27
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	8.53
	Start Aerob	[h:m]	02:02
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	1.3	4.9	0.5	17.8	0.38%	0.53%	14
	2	00:02:00	0.03	5.7	3.9	1.0	15.7	0.37%	0.52%	665
	3	00:15:30	0.26	26.9	1.5	2.9	11.9	0.55%	0.76%	657
	4	00:31:00	0.52	71.0	0.1	3.6	8.4	1.31%	1.62%	605
	5	01:00:00	1.00	175.3	0.0	0.0	0.2	1.43%	1.99%	483
	6	01:30:00	1.50	198.0	0.1	0.0	0.2	3.01%	4.19%	424
	7	02:00:00	2.00	194.7	0.0	0.0	0.2	2.88%	4.00%	394
	8	02:15:30	2.26	196.7	0.1	0.0	0.2	2.97%	4.12%	311
	9	02:31:00	2.52	192.3	0.0	0.9	2.2	3.86%	5.36%	220
	10	03:00:00	3.00	182.7	1.2	2.3	9.1	4.73%	6.56%	43
	11	03:31:00	3.52	153.4	3.6	2.8	18.8	5.20%	7.22%	22
	12	04:00:00	4.00	136.4	6.4	3.2	29.0	4.52%	6.28%	29
	13	04:30:00	4.50	129.6	9.0	3.8	39.7	3.82%	5.30%	
	14	05:01:00	5.02	126.5	11.2	5.0	49.9	3.54%	4.91%	
	15	05:30:00	5.50	123.8	13.6	5.5	59.4	3.64%	5.05%	
	16	06:30:00	6.50	118.3	18.0	7.0	77.8	2.80%	3.89%	34
	17	07:30:00	7.50	100.9	22.7	7.5	94.8	3.18%	4.42%	

Date 07/11/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 400 mg/L acetic acid

		[mg/L]	[mmol/L]
Feed:	Acetate	400	6.66
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	50.9	1.30
	PO4-P	20	0.65
	NH4-N	10.9	0.78

Misc.	MLSS	[g/L]	3.99
	MLVSS	[g/L]	2.79
	S/X	[mg/mg]	0.18
	tot. P	[mg/L]	316
	PH Start	[-]	-
	PH Ende	[-]	8.82
	Start Aerob	[h:m]	02:02
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time, dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	3.9	3.6	0.6	13.7	0.41%	0.58%	64
	2	00:01:30	0.03	35.4	2.0	1.2	9.6	0.58%	0.83%	496
	3	00:15:00	0.25	87.6	0.0	0.0	0.0	1.14%	1.63%	420
	4	00:30:00	0.50	141.9	0.0	0.0	0.0	1.33%	1.90%	357
	5	01:00:00	1.00	203.1	0.0	0.0	0.0	2.34%	3.35%	257
	6	01:33:00	1.55	199.5	0.0	0.0	0.0	2.90%	4.14%	220
	7	02:01:00	2.02	194.8	0.0	0.0	0.0	2.55%	3.64%	214
	8	02:15:00	2.25	184.5	0.4	0.5	2.6	3.18%	4.54%	103
	9	02:30:00	2.50	154.7	1.5	0.9	7.4	3.21%	4.58%	50
	10	03:00:00	3.00	108.1	4.2	1.2	17.3	3.62%	5.17%	36
	11	03:31:00	3.52	85.2	7.1	1.7	28.4	2.93%	4.19%	
	12	04:00:00	4.00	70.5	9.9	2.3	39.2	2.26%	3.23%	
	13	04:30:00	4.50	54.4	12.7	2.8	49.8	2.67%	3.81%	
	14	05:00:00	5.00	43.4	16.1	3.5	63.2	2.09%	2.98%	
	15	05:32:00	5.53	31.0	18.8	4.0	73.5	2.42%	3.46%	
	16	06:02:00	6.03	19.1	21.8	4.8	85.8	1.46%	2.08%	
	17	06:31:00	6.52	7.1	24.0	5.3	94.2	1.66%	2.37%	
	18	07:00:00	7.00	1.4	26.1	4.0	98.6	1.40%	2.00%	
	19	07:30:00	7.50	0.6	27.9	2.2	100.6	1.85%	2.64%	
	20	08:00:00	8.00	0.3	29.0	0.0	99.3	1.31%	1.88%	28

Date 07/22/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with two spikes of ca. 300 mg/L acetic acid each

		[mg/L]	[mmol/L]
Feed:	Acetate	300	5.00
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	38.2	0.98
	PO4-P	15	0.49
	NH4-N	8.2	0.58

		[mg/L]	[mmol/L]
2nd Spike	Acetate	300	5
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	38.2	0.98
	PO4-P	15	0.49
	NH4-N	8.2	0.58

Misc.	MLSS (est.)	[g/L]	3.44	(4.41 at End of Batch-Test)
	MLVSS (est.)	[g/L]	2.41	(3.11 at End of Batch-Test)
	S/X	[mg/mg]	0.15	
	tot. P	[mg/L]	347	
	PH Start	[-]	-	
	PH Ende	[-]	-	
	Start Aerob	[h:m]	01:32	
	2nd Spike	[h:m]	01:32	

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	1.4	1.5	2.1	9.9	0.27%	0.38%	4
	2	00:01:30	0.03	42.2	0.0	0.0	0.1	0.79%	1.11%	305
	3	00:16:00	0.27	118.7	0.0	0.0	0.2	1.68%	2.36%	201
	4	00:31:00	0.52	154.8	0.0	0.0	0.2	1.68%	2.36%	151
	5	01:00:00	1.00	205.3	0.1	0.1	0.4	3.00%	4.22%	66
	6	01:30:00	1.50	217.8	0.1	0.3	1.0	4.04%	5.69%	22
	7	01:35:00	1.58	208.8	0.1	0.0	0.5	3.33%	4.69%	369
	8	01:49:00	1.82	183.0	0.4	0.0	1.3	2.84%	4.01%	269
	9	02:01:00	2.02	170.2	1.0	0.8	5.0	6.11%	8.60%	186
	10	02:31:00	2.52	148.1	3.1	1.2	13.6	4.81%	6.78%	6
	11	03:00:00	3.00	95.7	5.4	1.4	21.8	5.28%	7.44%	2
	12	03:30:00	3.50	63.8	7.6	1.6	29.9	4.15%	5.84%	
	13	04:00:00	4.00	48.6	10.4	0.0	35.6	4.62%	6.51%	
	14	04:30:00	4.50	41.1	14.1	0.0	48.5	3.53%	4.97%	
	15	05:00:00	5.00	32.2	16.1	0.0	55.2	2.54%	3.57%	
	16	05:30:00	5.50	27.5	18.8	0.0	64.5	2.85%	4.02%	
	17	06:00:00	6.00	24.2	21.4	0.0	73.3	2.19%	3.09%	
	18	06:30:00	6.50	22.6	23.7	0.0	81.4	2.51%	3.53%	
	19	07:30:00	7.50	23.3	26.8	0.0	91.8	2.41%	3.39%	
	20	08:30:00	8.50	20.4	29.3	0.0	100.4	2.33%	3.28%	

Date 07/26/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 600 mg/L acetic acid; all Nutrients tripled
Strong foaming at first

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	540	
	Ca	112.9	2.82
	Mg	37.9	1.56
	Fe	3.9	0.07
	K	76.3	1.95
	PO4-P	30.1	0.98
	NH4-N	16.4	1.17

Misc.	MLSS	[g/L]	3.13
	MLVSS	[g/L]	2.23
	S/X	[mg/mg]	0.27
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	02:01
	2nd Spike	[h:m]	-

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	2.3	3.5	1.7	15.7	0.24%	0.34%	8
	2	00:03:30	0.06	58.9	0.0	0.0	0.0	0.73%	1.03%	555
	3	00:20:00	0.33	93.5	0.0	0.0	0.0	1.50%	2.11%	489
	4	00:32:00	0.53	114.8	0.0	0.0	0.0	1.10%	1.56%	448
	5	01:04:00	1.07	154.4	0.0	0.0	0.0	2.08%	2.93%	363
	6	01:34:00	1.57	151.2	0.1	0.0	0.3	3.58%	5.04%	327
	7	02:03:00	2.05	149.9	0.1	0.0	0.4	2.76%	3.89%	332
	8	02:17:00	2.28	129.2	0.1	0.0	0.3	3.54%	4.99%	211
	9	02:33:00	2.55	119.5	0.1	0.0	0.2	4.08%	5.74%	103
	10	03:02:00	3.03	88.4	3.8	1.5	16.5	3.43%	4.83%	7
	11	03:33:00	3.55	57.9	6.0	2.2	25.6	3.08%	4.34%	9
	12	04:02:00	4.03	40.8	9.0	2.3	36.0	3.43%	4.84%	
	13	04:33:00	4.55	30.0	12.3	0.1	42.3	2.23%	3.14%	
	14	05:03:00	5.05	21.7	14.6	0.0	50.0	2.18%	3.07%	
	15	05:33:00	5.55	16.9	16.9	0.0	58.0	2.51%	3.54%	
	16	06:02:00	6.03	11.9	19.1	0.0	65.6	2.01%	2.84%	
	17	06:32:00	6.53	9.6	21.4	0.0	73.3	2.19%	3.09%	
	18	07:02:00	7.03	9.2	23.4	0.0	80.3	2.42%	3.40%	6
	19	07:32:00	7.53	8.1	24.8	0.0	85.1	2.63%	3.70%	

Date 08/05/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 600 mg/L acetic acid
Nutrients added in two spikes

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	76.3	1.95
	PO4-P	30	0.98
	NH4-N	1.64	1.17

		[mg/L]	[mmol/L]
2nd Spike	Acetate	0	0
	Nutr. Broth	360	
	Ca	75.2	1.88
	Mg	25.2	1.04
	Fe	2.6	0.05
	K	0	0
	PO4-P	0	0
	NH4-N	0	0

Misc.	MLSS	[g/L]	3.56
	MLVSS	[g/L]	2.48
	S/X	[mg/mg]	0.27
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	-
	Start Aerob	[h:m]	02:02
	2nd Spike	[h:m]	03:47

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	2.4	1.1	2.3	8.8	0.21%	0.30%	0
	2	00:01:15	0.02	30.8	0.1	2.7	6.4	0.26%	0.37%	658
	3	00:17:30	0.29	92.7	0.1	0.1	0.4	1.21%	1.73%	561
	4	00:31:00	0.52	125.7	0.0	0.0	0.2	2.35%	3.36%	503
	5	01:03:00	1.05	195.8	0.0	0.0	0.0	4.48%	6.40%	426
	6	01:32:00	1.53	197.8	0.1	0.0	0.3	3.40%	4.86%	399
	7	02:01:00	2.02	162.0	0.1	0.0	0.4	3.34%	4.78%	380
	8	02:15:00	2.25	182.3	0.8	0.3	3.6	7.16%	10.23%	292
	9	02:45:00	2.75	164.7	2.7	0.0	9.1	3.97%	5.68%	143
	10	03:15:00	3.25	167.5	4.7	1.2	19.0	9.48%	13.55%	35
	11	03:47:00	3.78	145.7	7.0	1.7	27.6	7.62%	10.89%	2
	12	04:00:00	4.00	101.1	7.2	1.6	28.3	6.23%	8.90%	
	13	04:30:00	4.50	80.2	9.0	1.9	35.1	6.07%	8.67%	
	14	05:02:00	5.03	73.5	12.2	0.0	41.7	6.71%	9.59%	
	15	05:32:00	5.53	62.5	14.2	0.1	48.9	5.46%	7.79%	
	16	06:15:00	6.25	54.5	17.2	0.1	59.0	5.58%	7.97%	
	17	07:01:00	7.02	49.9	19.9	0.1	68.2	5.67%	8.10%	
	18	07:45:00	7.75	47.3	22.8	0.1	78.2	5.66%	8.08%	
	19	08:31:00	8.52	43.0	24.6	0.1	84.6	4.38%	6.25%	

Date 08/09/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 600 mg/L acetic acid
Nutrients added in two spikes; 2nd Spike only Nutrient Broth

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02.
	K	76.3	1.95
	PO4-P	30	0.98
	NH4-N	1.64	1.17

		[mg/L]	[mmol/L]
2nd Spike	Acetate	0	0
	Nutr. Broth	360	
	Ca	0	0
	Mg	0	0
	Fe	0	0.00
	K	0	0
	PO4-P	0	0
	NH4-N	0	0

Misc.	MLSS	[g/L]	2.96
	MLVSS	[g/L]	2.19
	S/X	[mg/mg]	0.29
	tot. P	[mg/L]	-
	PH Start	[-]	-
	PH Ende	[-]	8.60
	Start Aerob	[h:m]	02:01
	2nd Spike	[h:m]	04:02

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
	1	00:00:00	0.00	2.6	3.0	1.8	14.4	0.67%	0.96%	0
	2	00:01:20	0.02	39.0	1.5	2.6	11.2	1.04%	1.49%	631
	3	00:15:00	0.25	85.5	0.0	1.2	2.8	2.05%	2.93%	580
	4	00:30:00	0.50	138.6	0.0	0.0	0.1	2.54%	3.62%	524
	5	01:01:00	1.02	172.4	0.0	0.0	0.0	2.86%	4.09%	441
	6	01:30:00	1.50	175.9	0.0	0.0	0.0	3.50%	5.00%	405
	7	02:00:00	2.00	170.0	0.0	0.0	0.0	3.97%	5.67%	367
	8	02:15:00	2.25	159.3	0.5	0.4	2.6	4.24%	6.06%	286
	9	02:30:00	2.50	156.7	1.5	0.6	6.7	5.33%	7.61%	223
	10	03:00:00	3.00	153.6	3.4	1.3	14.4	5.59%	7.98%	84
	11	03:31:00	3.52	145.8	4.3	1.8	19.0	6.11%	8.72%	6
	12	04:00:00	4.00	137.0	6.3	0.0	21.7	6.03%	8.61%	0
	13	04:15:00	4.25	137.1	6.7	2.8	29.4	5.82%	8.32%	
	14	04:33:00	4.55	134.6	8.0	3.1	34.4	5.71%	8.16%	
	15	05:00:00	5.00	115.4	8.8	3.4	38.1	5.95%	8.50%	
	16	05:30:00	5.50	106.4	10.1	3.7	43.1	5.16%	7.38%	
	17	06:15:00	6.25	95.0	11.9	4.5	50.9	4.88%	6.97%	
	18	07:00:00	7.00	89.6	14.0	5.1	59.5	6.16%	8.80%	
	19	07:45:00	7.75	84.6	15.8	5.6	66.8	6.06%	8.66%	

Date 08/12/94

Description System II brought to steady state with 200 mg/L acetic acid
Batch-Test with ca. 600 mg/L acetic acid
Nutrients added in two spikes; 2nd Spike only CaCl2

		[mg/L]	[mmol/L]
Feed:	Acetate	600	9.99
	Nutr. Broth	180	
	Ca	37.6	0.94
	Mg	12.6	0.52
	Fe	1.3	0.02
	K	76.3	1.95
	PO4-P	30	0.98
	NH4-N	1.64	1.17

		[mg/L]	[mmol/L]
2nd Spike	Acetate	0	0
	Nutr. Broth	0	
	Ca	56.4	1.41
	Mg	0	0
	Fe	0	0.00
	K	0	0
	PO4-P	0	0
	NH4-N	0	0

Misc.	MLSS	[g/L]	3.49
	MLVSS	[g/L]	2.37
	S/X	[mg/mg]	0.25
	tot. P	[mg/L]	305
	PH Start	[-]	-
	PH Ende	[-]	8.80
	Start Aerob	[h:m]	02:02
	2nd Spike	[h:m]	04:02

Comments	Sample #	Time [h:m]	Time; dez. [hours]	PO4-P [mg/L]	NO3-N [mg/L]	NO2-N [mg/L]	NOx - O [mg/L]	PHB/MLSS [%]	PHB/VSS [%]	Acetate as Acetic Acid [mg/L]
no Zero Sample										
	1	00:01:00	0.02	40.9	0.1		0.2	0.58%	0.86%	592
	2	00:16:00	0.27	82.6	0.2		0.5	1.57%	2.31%	506
	3	00:30:00	0.50	120.8	0.0		0.1	1.74%	2.55%	474
	4	01:00:00	1.00	154.3	0.0		0.0	2.68%	3.94%	401
	5	01:30:00	1.50	173.9	0.0		0.1	2.01%	2.96%	376
	6	02:01:00	2.02	166.2	0.0		0.2	3.62%	5.32%	358
	7	02:16:30	2.28	151.6	0.1		0.2	2.76%	4.06%	257
	8	02:45:00	2.75	144.6	1.7		5.9	3.17%	4.66%	134
	9	03:15:00	3.25	149.3	3.6		12.3	5.87%	8.63%	24
	10	04:01:00	4.02	106.0	5.2		17.7	4.11%	6.04%	0
	11	04:17:00	4.28	90.8	7.2		24.6	6.78%	9.97%	
	12	04:45:00	4.75	77.2	8.9		30.5	5.81%	8.54%	
	13	05:30:00	5.50	66.5	13.1		45.0	4.96%	7.29%	
	14	06:15:00	6.25	49.1	16.2		55.5	4.60%	6.76%	
	15	07:01:00	7.02	32.0	19.3		66.3	4.55%	6.70%	
	16	07:46:00	7.77	13.3	23.1		79.2	4.39%	6.46%	
	17	08:45:00	8.75	2.1	26.1		89.3	3.84%	5.64%	

Curriculum Vitae

Alexander Gotthard Heinz Seyfried was born on June 14, 1962, in Hannover, Germany. After graduating from the Kaiser-Wilhelm-Gymnasium, Hannover, in December 1980, he had for six months an internship at Aqua Consult Ingenieur GmbH, Hannover. From July 1981 to September 1982 he completed his military service. From October 1983 to March 1985 he was enrolled at the University of Hannover (Technische Hochschule) in Civil Engineering, where he received in October 1984 his *Vordiplom* (comparable to Bachelors degree). In April 1985 he enrolled at the University of Karlsruhe in Civil Engineering. In May 1991 he completed his studies and earned the degree of *Diplomingenieur für Bauingenieurwesen* (comparable to Masters Degree in Civil Engineering). During the same time (April 1985 to August 1991) he worked as a scientific auxiliary worker at the Institut für Siedlungswasserwirtschaft (Department for Sanitary and Environmental Engineering), University of Karlsruhe. Furthermore, he completed internships at Bilfinger & Berger, AG and Phillip Holzmann, AG. Since September 1991 he is employed as a planning and design engineer by Aqua Consult Ingenieur GmbH, Hannover.

In the summer of 1993 he enrolled at the Virginia Polytechnic Institute and State University as a graduate student in Environmental Engineering. He completed his studies in December 1994 with the degree of Master of Science in Environmental Engineering.