Experimental and model-based evaluation of the role of denitrifying polyphosphate accumulating organisms at two large scale WWTPs in northern Poland

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Abstract The capabilities of denitrifying Polyphosphate Accumulating Organisms (DPAOs) in two large-scale plants in northern Poland performing enhanced biological phosphorus removal (EBPR) were evaluated in this study. A series of batch tests with the process biomass aimed at the measurements of phosphate release (with artificial substrate and real wastewater) and subsequent phosphate uptake under anoxic/aerobic conditions. The process kinetics were predicted using ASM2d implemented in the GPS-X ver. 4.0.2 simulation package. The results from one experimental series (summer) were used for the model calibration, whereas the results from another series (spring) were used for the model validation. The model parameters were also accurately confirmed by predictions of the accompanying field measurements in the full-scale bioreactors. The experimental and simulation results revealed that a relatively small fraction of PAO could denitrify ($\eta_{NO_3,PAO} = 0.32$). The denitrification rates associated with the anoxic storage of PP and the anoxic growth of PAO only constituted 16.0–21.0% of the denitrification rates associated with the anoxic activity of "ordinary" heterotrophs.

Keywords Batch test; dynamic simulation; EBPR; full-scale WWTP; mathematical modelling; PAO

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Introduction

In the modern biological nutrient removal activated sludge (BNRAS) systems performing nitrification, denitrification and enhanced biological phosphorus removal (EBPR), the latter process is most complex and least understood. The EBPR is accomplished by microorganisms referred to collectively as the Phosphate Accumulating Organisms (PAOs). The principal requirements for building up PAO in the system are a sequence of alternating anaerobic/aerobic (or anoxic) phases and the presence of volatile fatty acids (VFA) during the anaerobic phase. Many studies have been dedicated to the behaviour of PAO under such conditions. However, most of these studies have been carried out in lab-scale sequencing batch reactor (SBR) systems with enriched PAO cultures using acetate as a sole source of VFA in the influent medium (synthetic wastewater). Batch tests aiming at the measurement of the kinetics of anaerobic P release and subsequent aerobic/anoxic P uptake by the full-scale process biomass mixed with real wastewater have seldom been reported in literature (Kuba *et al.*, 1997; Sorm *et al.*, 1998). In the mixed culture BNRAS systems fed with real wastewater, the PAOs coexist with other microbial groups such as the "ordinary" heterotrophs. The PAO behaviour in these systems may thus be different from that encountered in the enriched culture systems. Hu *et al.* (2002a) proved this hypothesis for the denitrification kinetics of denitrifying PAOs (DPAOs) which competed with other denitrifying heterotrophs for the limited nitrate under anoxic conditions. Due

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to the double function (EBPR and denitrification), this fraction of PAO may play an essential role in BNRAS systems but the actual behaviour of DPAO in full-scale plants has hardly been studied so far.

The aim of this paper was to evaluate the capabilities of DPAOs by means of lab experiments and computer simulation in two large-scale plants in northern Poland performing EBPR. A series of batch tests with the process biomass aimed to measure the rates of phosphate release and subsequent phosphate uptake under anoxic/aerobic conditions. The process kinetics in both the batch reactor and the full-scale plant were predicted using a selected biokinetic model. The contribution of DPAO to the denitrification rates was determined by coupling the experimental and simulation results.

Methods

Process configuration and performance of the WWTPs under study

Experimental studies were carried out at the plants located in two neighbouring cities of Gdansk (approx. 500,000 inhabitants) and Gdynia (approx. 330,000 inhabitants). Both plants discharge the treated wastewater to the Bay of Gdansk which is part of the Baltic Sea.

Wschod WWTP. The "Wschod" WWTP (approx. 700,000 PE) in Gdansk is one of the largest facilities located upon the Baltic Sea. The biological step consists of six parallel bioreactors and twelve circular secondary clarifiers. The bioreactors run in the MUCT process configuration with an additional deoxic zone in the internal recirculation line from the aerobic zone to the anoxic zone. The volume of a single bioreactor is 26,350 m^3.

Debogorze WWTP. The "Debogorze" WWTP (approx. 500,000 PE) treats wastewater originating from the city of Gdynia and four surrounding smaller towns. The existing biological step, run in the Johannesburg process configuration, consists of four parallel bioreactors and six secondary clarifiers. The volume of a single bioreactor is 12,000 m^3.

More information about the "Wschod" and "Debogorze" WWTPs, operational parameters, effluent standards and N removal efficiency can be found elsewhere (Makinia *et al.*, 2004). The principal parameters characterizing the operating conditions at these plants are summarized in Table 1. It should be noted that in both cases the settled wastewater contains a high fraction of soluble organic material. The average ratios of COD in samples pre-treated with $Zn(OH)_2$, referred to as "soluble COD", to total COD were 37.7% and 44.0% at the "Wschod" and "Debogorze" WWTP, respectively. The ratio of VFA to soluble COD was similar in both cases and equal to approx. 0.5. The procedure of detailed wastewater characterization was based on the standard Dutch guidelines (Roeleveld and Van Loosdrecht, 2002) and adapted to the existing measurements at the plants under study (Makinia, 2006).

Procedures of lab-scale experiments

Lab experiments were carried out in a batch reactor with the maximum volume 3.7 dm^3. The reactor was equipped with electrodes for a continuous monitoring of pH, ORP, temperature and dissolved oxygen (DO). An automated control system allowed us to keep a desired DO concentration in the reactor and temperature in a water jacket of the batch reactor. Moreover, an automatic measurement of oxygen uptake rate (OUR) was conducted in a cyclic mode (every 3 minutes) in a small chamber connected with the main unit. During the period November, 2001 – September, 2002 three series of various batch tests were carried out at the "Wschod" and "Debogorze" WWTPs. Two types of these

Table 1 Characteristics of the operating conditions at the two WWTPs under study

Parameter	Unit	Wschod WWTP		Debogorze WWTP	
		Summer period	Spring period	Summer period	Spring period
Operating parameters:					
Influent flow rate	m^3/d	21,189	23,503	13,568	13,948
Process temperature	^oC	19.6	14.1	21.2	18.6
Sludge Retention Time	d	14.6	14.2	15.0	15.5
Concentrations in settled wastewater:					
COD	$gCOD/m^3$	546	609	571	590
P_{tot}	gP/m^3	14.6	15.0	17.7	16.3
$P\text{-}PO_4^-$	gP/m^3	9.0	9.1	13.4	12.7
N_{tot}	gN/m^3	70.0	76.7	79.0	78.7
Concentrations in secondary effluent:					
COD	$gCOD/m^3$	33.0	52.8	36.6	33.1
P_{tot}	gP/m^3	0.39	0.50	0.89	1.61
$P\text{-}PO_4^-$	gP/m^3	0.09	0.03	0.77	1.43
N_{tot}	gN/m^3	9.9	11.9	12.7	13.6
Biomass characteristics:					
MLSS	g/m^3	3110	3630	4570	4710
MLVSS/MLSS ratio (i_{VT})	–	0.712	0.747	0.690	0.689
P content of MLSS (i_{PT})	gP/g	0.055	0.053	0.049	0.046

tests aiming at the determination of denitrification rates were previously outlined by Makinia *et al.* (2004). Below are described the tests that allowed us to determine the rates of P release/uptake and evaluate the DPAO capabilities in the plants under study.

Phosphorus release in the anaerobic conditions and phosphorus uptake in the aerobic conditions. The test was carried out with two different sources of VFA: settled wastewater and sodium acetate (948 mg dissolved in the denitrified treated wastewater). Fresh returned activated sludge and the substrate were mixed in such proportions to obtain MLSS at approx. $3\,kg/m^3$ in the reactor. At the beginning of the test, the aeration system supplying gaseous nitrogen was turned on and the on-line monitoring of temperature, DO, pH and ORP was initiated. The temperature in the water jacket was set at a desired value. The duration of the anaerobic and aerobic phases was 150 min and 180 min, respectively. At the end of the anaerobic phase, the aeration system was switched from gaseous nitrogen to air. During the aerobic phase, the DO set point was controlled at $6\,g\,O_2/m^3$. Samples of $50\,cm^3$ were withdrawn with the frequency of 10–30 min over the duration of the entire test, filtered under vacuum pressure on the Whatman GF/C filter and analysed for $P\text{-}PO_4^{3-}$ and COD. The actual MLSS concentration in the reactor was measured at the beginning and at the end of the test.

Phosphorus release in the anaerobic conditions and phosphorus uptake in the anoxic conditions. Until the end of the anaerobic phase the procedure was identical with the previous one. At the beginning of the anoxic phase, 693 mg of potassium nitrate (KNO_3) were added to raise the $N\text{-}NO_3^-$ concentration by $30\,g\,N/m^3$. Samples were withdrawn with the frequency of 10–30 min, filtered under vacuum pressure on the Whatman GF/C filter and analysed for $P\text{-}PO_4^{3-}$ and COD (anaerobic phase) and $P\text{-}PO_4^{3-}$, $N\text{-}NO_3^-$ and COD (anoxic phase). The actual MLSS concentration in the reactor was measured at the beginning and at the end of the test.

Full-scale measurement campaigns

Each series of lab experiments was accompanied by an additional 48-hour "continuous" measurement campaign conducted in the full-scale reactors. Grab samples were withdrawn every two hours at the following locations: reactor inlet (denoted as INF), effluent from the anaerobic zone (ANA), effluent from the anoxic zone (ANOX), middle of the aerobic zone (AER-M), reactor effluent (AER-E). The samples were analysed for several parameters including the P-PO_4^{3-} concentrations. In addition, "on-line" recordings of flow rates, temperature and DO concentrations in the aerobic compartments were also collected for the simulation study.

Simulator environment and model calibration procedure

The simulation study was accomplished using Activated Sludge Model No. 2d (ASM2d) (Henze *et al.*, 1999) implemented in the GPS-X ver. 4.0.2 simulation package (Hydromantis, 2002). Simulations were initially run with the model default values, but the obtained predictions were inaccurate for both plants and the model required further calibration. The iterative step-wise calibration procedure including steady-state and phased-dynamic simulations of the full-scale reactor performance as well as dynamic simulations of the batch tests with real wastewater was continued until one set of model parameters could be used in all of the cases. The batch tests from another (spring) study period and 48-hour "continuous" measurements in the full-scale bioreactors provided data for the ASM2d validation. The entire calibration/validation procedure was discussed in detail by Makinia (2006).

Results and discussion

Kinetics and stoichiometry of the P release and uptake

The measured specific process rates are listed in Table 2. These results are compared with the literature data reporting similar experiments carried out with the biomass originating from full-scale BNRAS systems (Kuba *et al.*, 1997; Sorm *et al.*, 1998) or full-scale pilot plants performing EBPR (Petersen *et al.*, 1998; Tykesson *et al.*, 2002). The anaerobic P release rates at the two plants under study were comparable with the upper range of the literature data. The ratio of P released to COD utilized (Y_{PO_4}) measured during the initial 60 min of the anaerobic phase ranged from 0.32 to 0.39 gP/gCOD ($r^2 = 0.92–1.0$) for the "Wschod" WWTP and from 0.39 to 0.50 gP/gCOD ($r^2 = 0.88–1.0$) for the "Debogorze" WWTP. For comparison, the same coefficients calculated for the results of the tests with acetate varied within a similar range (0.36–0.68 gP/gCOD, $r^2 = 0.93–1.0$) at the "Debogorze" WWTP, and were higher (0.48–0.66 gP/gCOD, $r^2 = 0.92–0.99$) at the "Wschod" WWTP. In the latter case, the discrepancy may imply that acetate is not a representative substrate for the PAO. These results are in accordance

Table 2 Comparison of the measured process rates at the WWTPs under study with the literature data

Process	Wschod WWTP		Debogorze WWTP		Literature data	
	Process rate mgP/(gVSS·h)	Temp. °C	Process rate mgP/(gVSS·h)	Temp. °C	Process rate mgP/(gVSS·h)	Temp. °C
Anaerobic P release	15.1–21.4	15.9–21.9	12.8–21.8	18.1–20.3	4.4–18.0*	20 ± 1
Aerobic P uptake	7.0–9.6	16.1–22.2	5.8–6.5	18.2–20.5	13.0–30.0**	20 ± 1
Anoxic P uptake	2.2–3.0	15.4–18.7	2.1–3.1	17.7–20.1	1.9–13.0***	20 ± 1

*Kuba *et al.* (1997), Petersen *et al.* (1998), Sorm *et al.* (1998), Tykesson *et al.* (2002)
**Kuba *et al.* (1997), Sorm *et al.* (1998)
***Kuba *et al.* (1997), Sorm *et al.* (1998)

with the observations of Ubukata (2005) who demonstrated that acetate and compounds present in real wastewater could cause different P release kinetics.

In contrast to the P release, the specific rates of aerobic/anoxic P uptake at the two plants remained in the lower range (or even below the range) of the values reported in the literature. It should be noted, however, that the rates presented in this study refer to the entire duration of the uptake phase. In the case of the aerobic tests, the initial process rates were considerably higher than the average values listed in Table 2. The maximum aerobic P uptake rates, measured during the first 60 min of the aerobic phase, ranged from 10.0 to 15.3 mgP/(gVSS·h) ($r^2 = 0.99$) at the "Wschod" WWTP and from 11.2 to 11.9 mgP/(gVSS·h) ($r^2 = 0.99$) at the "Debogorze" WWTP. The anoxic P uptake rates correlated closely with the denitrification rates measured in this phase of the experiment, i.e. 2.53–3.11 mgN/(gVSS·h) at the "Wschod" WWTP and 2.05–3.72 mg N/(gVSS·h) at the "Debogorze" WWTP (Makinia *et al.*, 2004). The calculated ratios of utilized nitrate and phosphate ($\Delta N - NO_3^- / \Delta P - PO_4^{3-}$) were 1.03–1.17 gN/gP ($r^2 = 0.97$–$0.98$) and 1.07–1.20 gN/gP ($r^2 = 0.84$–$0.87$) at the "Wschod" and "Debogorze" WWTP, respectively. Even though both processes appeared to be strongly related, the actual contribution of denitrifying PAO to the utilization of $N\text{-}NO_3^-$ was not significant. This observation was confirmed by the follow-up simulation studies.

Simulation of the P release and uptake in the batch reactor

Table 3 contains a list of kinetic coefficients adjusted at both plants during the model calibration. The default values were assumed for stoichiometric coefficients except for the polyphosphate (PP) requirement for PHA storage, Y_{PO_4}, which was the average value of the direct measurements during the anaerobic P release with real wastewater (i.e. $Y_{PO_4} = 0.34$ gP/gCOD and 0.43 gP/gCOD for the "Wschod" WWTP and "Debogorze" WWTP, respectively). It should be noted that the modified values of the rate constants for storage of PHA, q_{PHA}, and for storage of PP, q_{PP}, are considerably higher than the

Table 3 List of the kinetic parameters in ASM2d adjusted during model calibration

Symbol	Unit	Default value[1]	Calibrated value	
			Wschod	Debogorze
Hydrolysis:				
K_h	d^{-1}	3.0	4.0	4.0
η_{NO_3}	–	0.6	0.8	0.9
η_{fe}	–	0.4	0.1	0.15
Heterotrophic organisms:				
μ_H	d^{-1}	6.0	4.0	4.0
η_{NO_3}	–	0.8	default	0.9
K_{fe}	$gCOD/m^3$	4.0	default	8.0
Autotrophic (nitrifying) organisms:				
μ_{AUT}	d^{-1}	1.0	1.2	1.0
K_{NH_4}	gN/m^3	1.0	1.2	1.3
Phosphorus Accumulating Organisms:				
q_{PHA}	d^{-1}	3.0	10	10
q_{PP}	d^{-1}	1.5	8	8
η_{NO_3}	–	0.6	0.32	0.32
b_{PAO}	d^{-1}	0.2	default	0.14
b_{PP}	d^{-1}	0.2	default	0.14
b_{PHA}	d^{-1}	0.2	default	0.14
K_A	$gCOD/m^3$	4.0	1.0	default
K_{IPP}	gP/gCOD	0.02	0.3	0.3
K_{PHA}	gP/gCOD	0.01	0.3	0.3

[1]Henze *et al.* (1999)

model defaults. In the case of P uptake, the increased process rate resulted from the higher value of q_{PP} was compensated by the higher values of the inhibition coefficient for X_{PP} storage, K_{IPP}, and the saturation coefficient for PHA, K_{PHA}. The results of simulation of the P release and uptake in the batch reactor are presented in Figures 1 and 2. At the "Wschod" WWTP, the average relative deviations (ARDs) between the measured data and model predictions for the calibration study period ranged from 2.6% (anaerobic P release/anoxic P uptake) to 4.6% (anaerobic P release/aerobic P uptake). The corresponding values at the "Debogorze" WWTP were 3.5% and 8.8% (the calibration period). The values of ARD for both tests from the validation study period varied within the range 3.4–13.7% (Wschod) and 4.8–10.4% (Debogorze).

The value of anoxic reduction factor for anoxic activity of PAO ($\eta_{NO_3,PAO} = 0.32$) implies that the capabilities of DPAO are not significant at both plants in terms of the contribution to the utilization of $N\text{-}NO_3^-$. The simulation results revealed that the denitrification rates associated with the anoxic storage of PP and the anoxic growth of PAO only constituted 16.0–16.7% ("Wschod" WWTP) and 16.7–21.1% ("Debogorze" WWTP) of the denitrification rates associated with the anoxic activity of "ordinary" heterotrophs. These values are closely related to the literature data reported by Hu *et al.* (2002b) who found that the specific denitrification rate of PAOs on internally stored PHB was only about 20% of the rate of the "ordinary" heterotrophs on slowly biodegradable COD. In another paper (Hu *et al.*, 2002a), the same authors concluded that the main factor stimulating the anoxic P uptake by DPAO would be a sufficient nitrate load to the main anoxic reactor. The other factors influencing the occurrence of PAO include the system aerobic mass fraction, sequence of reactors and frequency of sludge alternation between the aerobic and anoxic states. In this study, a similar behaviour of PAO was observed at both plants even though the above mentioned factors deviated from each other. At the "Wschod" WWTP, the average effluent concentrations of $N\text{-}NO_3^-$ from the anoxic zone ranged from 0.1 to 0.7 gN/m^3 during the "continuous" measurement campaigns, whereas these concentrations at the "Debogorze" WWTP were higher,

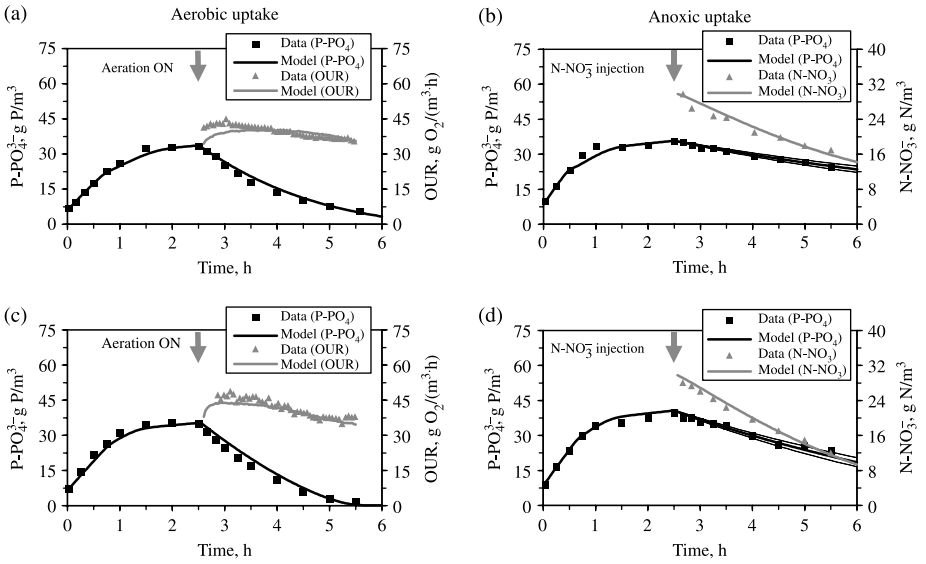

Figure 1 Measured and predicted $P\text{-}PO_4^{3-}$ concentrations during the P release/uptake batch tests carried out at the "Wschod" WWTP during the calibration period (a-b) and validation period (c-d)(dash line during the anoxic P uptake – results of the sensitivity analysis with $\eta_{NO3,PAO} = 0.32 \pm 10\%$)

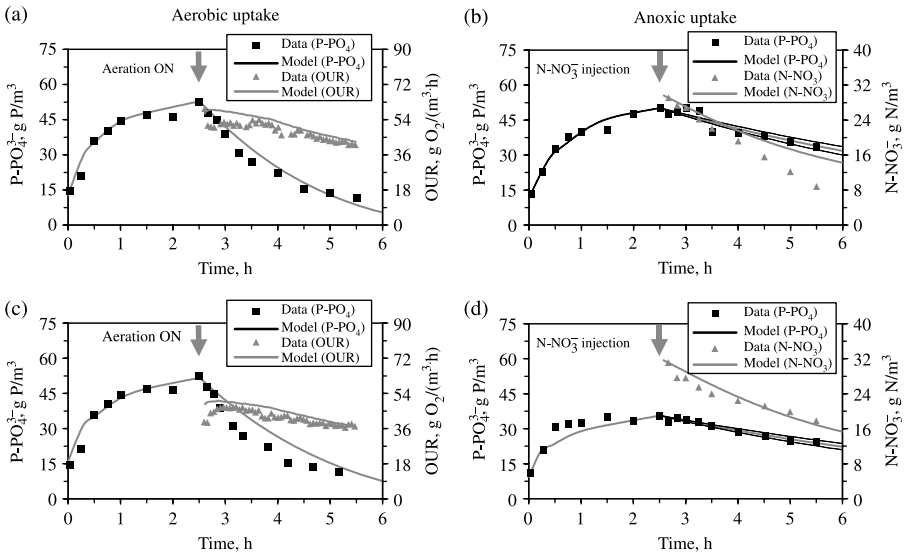

Figure 2 Measured and predicted P-PO_4^{3-} concentrations during the P release/uptake batch tests carried out at the "Debogorze" WWTP during the calibration period (a-b) and validation period (c-d))(dash line during the anoxic P uptake – results of the sensitivity analysis with $\eta_{NO3,PAO} = 0.32 \pm 10\%$)

i.e. 1.9–3.0 gN/m^3. The aerobic mass fraction was approx. 47% in the MUCT system (Wschod) and approx. 64% in the JHB system (Debogorze), respectively.

Simulation of the EBPR process in the full-scale bioreactors

The selected simulation results of the full-scale bioreactor performance in the summer study period are presented in Figures 3 and 4. At the "Wschod" WWTP, the ASM2d accurately predicted the longitudinal profile of P-PO_4^{3-} except for the concentrations in the anoxic zone (predicted 9.8 gP/m^3 vs. measured 7.8 gP/m^3). Moreover, a lower variability of the predicted concentrations (std = ±2.0 gP/m^3) compared to the measured values (std = ±4.5 gP/m^3) was found in the anaerobic zone. At the "Debogorze" WWTP, the model more accurately predicted the variability of P-PO_4^{3-} concentrations. The std values in the anaerobic zone were ± 9.5 gP/m^3 and ± 8.2 gP/m^3 for the model predictions and measured data, respectively. The corresponding values in the anoxic zone were ± 3.8 gP/m^3 and ± 3.7 gP/m^3. However, the average predicted concentrations in that zone and the middle of the aerobic compartment were overestimated by a relatively constant value of 2.2–2.3 gP/m^3.

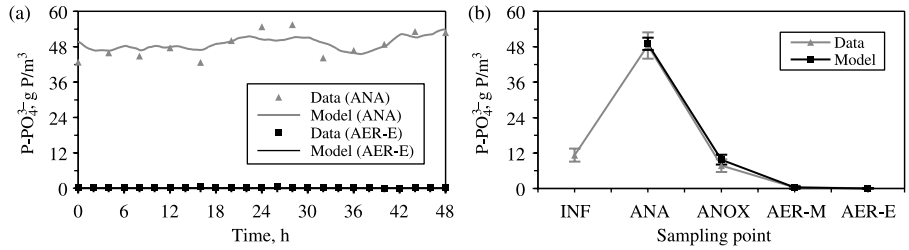

Figure 3 Measured and predicted P-PO_4^{3-} concentrations in the "Wschod" WWTP including temporal variations in the anaerobic zone and in the reactor effluent (a) and longitudinal profile of the average concentrations along the bioreactor (b)

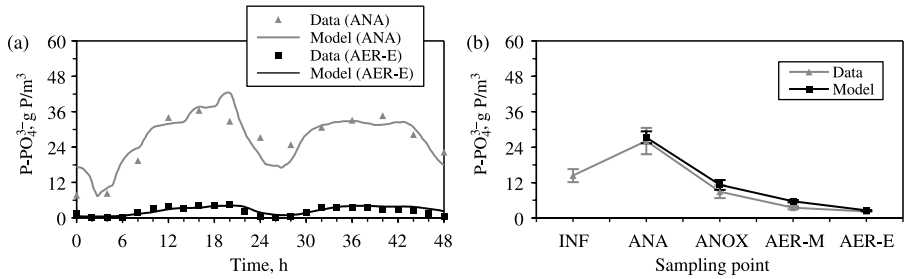

Figure 4 Measured and predicted P-PO_4^{3-} concentrations in the "Debogorze" WWTP including temporal variations in the anaerobic zone and in the reactor effluent (a) and longitudinal profile of the average concentrations along the bioreactor (b)

Table 4 Normalized sensitivity coefficients for $\eta_{NO3,PAO}$ calculated using the calibrated ASM2d

WWTP	Batch experiments						Full-scale measurements			
	P rel./aerobic P uptake			P rel./anoxic P uptake						
	Max. P release	Max. P uptake	OUR	Max. P release	Max. P uptake	NUR	P-PO_4^{3-} anaerobic	P-PO_4^{3-} anoxic	P-PO_4^{3-} effluent	N-NO_3^- effluent
Wschod	0.13	0.06	0.01	−0.03	1.13	0.14	−0.06	−0.71	−4.39	−0.17
Debogorze	0.04	0.03	0.01	−0.02	1.05	0.19	−0.06	−0.30	−0.30	−0.11

Sensitivity analysis

The normalized sensitivity coefficient, $S_{i,j}$, represents the percentage change in the output variable resulting from a 10% change in the input variable. A list of the sensitivity coefficients, $S_{i,j}$, calculated with the calibrated model for η_{NO_3},PAO with respect to the most important output variables considered in this study is presented in Table 4.

The sensitivity analysis revealed that only the impact of η_{NO_3},PAO on the maximum anoxic P uptake turned out to be influential, i.e. $S_{i,j} > 1$ (a high $S_{i,j}$ for the effluent P-PO_4^{3-} concentrations at the "Wschod" WWTP was caused by extremely low values of these concentrations). These results also confirm that the measurements of maximum anoxic P uptake rate are most suitable for estimating the value of η_{NO_3},PAO (see: Figures 1 and 2).

Conclusions

The experimental and simulation results revealed that a relatively small fraction of PAO could denitrify ($\eta_{NO_3,PAO} = 0.32$) at two large scale WWTPs in northern Poland. The denitrification rates associated with the anoxic storage of PP and the anoxic growth of PAO only constituted 16.0–21.0% of the denitrification rates associated with the anoxic activity of "ordinary" heterotrophs. The same set of ASM2d parameters (except for the rate constants for lysis of X_{PAO}, X_{PP} and X_{PHA}) revealed that the PAO processes could be used at both plants under study, even though the process conditions deviated from each other in terms of the system configuration, nitrate limitation in the anoxic zone and fraction of the aerobic biomass. The calibrated model accurately predicted the longitudinal profiles of P-PO_4^{3-} except for the concentrations in the anoxic zone. In this zone, the model predictions were overestimated by 2.0–2.3 gP/m^3 compared to the measurements at both studied plants. This discrepancy could be associated with the local disturbances, i.e. uncontrolled oxygen penetration to the anoxic zone from the air or aerobic compartments, and will be subject to further studies.

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References

Henze, M., Gujer, W., Mino, T., Matsuo, T., Wentzel, M.C., Marais, G.v.R. and Van Loosdrecht, M. (1999). Activated Sludge Model No. 2d. *Wat. Sci. Tech.*, **39**(1), 165–182.

Hu, Z.-R., Wentzel, M.C. and Ekama, G.A. (2002a). Anoxic growth of phosphate accumulating organisms (PAOs) in biological nutrient removal activated sludge systems. *Wat. Res.*, **36**, 4927–4937.

Hu, Z.-R., Wentzel, M.C. and Ekama, G.A. (2002b). The significance of denitrifying polyphosphate accumulating organisms in biological nutrient removal activated sludge systems. *Wat. Sci. Tech.*, **46**(1–2), 129–138.

Hydromantis, Inc. (2002). GPS-X 4.0.2 – User's Guide and Technical Reference, Hydromantis, Inc, Hamilton, Ontario.

Kuba, T., van Loosdrecht, M.C.M. and Heijnen, J.J. (1997). Biological dephosphatation by activated sludge under denitrifying conditions: pH influence and occurrence of denitrifying dephosphatation in a full-scale waste water treatment plant. *Wat. Sci. Tech.*, **36**(12), 75–82.

Makinia, J. (2006). Mathematical modelling and computer simulation as tools for the optimisation of biological nutrient removal activated sludge systems. *Veröffentlichungen des Institutes für Siedlungswasserwirtschaft und Abfalltechnik der Universität Hannover* (in print).

Makinia, J., Dobiegala, E. and Swinarski, M. (2004). The Polish perspective on adopting EU standards for nitrogen removal at large WWTP's – case studies. *Wat. Sci. Tech.*, **50**(7), 27–34.

Petersen, B., Temmink, H., Henze, M. and Isaacs, S. (1998). Phosphate uptake kinetics in relation to PHB under aerobic conditions. *Wat. Res.*, **32**, 91–100.

Roeleveld, P.J. and Van Loosdrecht, M.C.M. (2002). Experience with guidelines for wastewater characterisation in the Netherlands. *Wat. Sci. Tech.*, **45**(6), 77–87.

Sorm, R., Bortone, G., Wanner, J. and Tilche, A. (1998). Behaviour of activated sludge from a system with anoxic phosphate uptake. *Wat. Sci. Tech.*, **37**(4–5), 563–566.

Tykesson, E., Aspegren, H., Henze, M., Nielsen, P.H. and Jansen, J.L. (2002). Use of phosphorus release batch tests for modelling an EBPR pilot plant. *Wat. Sci. Tech.*, **45**(6), 99–106.

Ubukata, Y. (2005). Role of particulate organic matter and acetic acid for phosphate removal in anaerobic/aerobic activated sludge process. *Preprints of the IWA Specialized Conference "Nutrient Management in Wastewater Treatment Processes and Recycle Streams*, *19–21 September 2005"*, Lemtech Konsulting, Cracow (Poland), pp. 183–192.