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Ph. D. Thesis

WPŁYW SUBSTRATU WOLNOROZKŁADALNEGO NA KINETYKĘ PROCESÓW BIOCHEMICZNYCH W KOMORACH OSADU CZYNNEGO

The effect of slowly biodegradable substrate on the kinetics of biochemical processes in activated sludge bioreactors

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GDAŃSK 2012

Summary (in Polish)

Powszechnie stosowane w oczyszczalniach komunalnych układy wielofazowe, oparte na technologii osadu czynnego, umożliwiają efektywny przebieg procesów biologicznego usuwania związków organicznych, azotu i fosforu. Nowoczesne układy z biologicznym usuwaniem związków biogennych (z ang. BNR), wymagają zapewnienia odpowiedniej ilości organicznych związków rozkładalnych, w celu utrzymania prawidłowych warunków przebiegu jednostkowych procesów biochemicznych w komorze osadu czynnego, takich jak denitryfikacja czy podwyższona biologiczna defosfatacja (z ang. EBPR). W zależności od pochodzenia van Haandel i wsp. (1981) wyróżnili trzy grupy źródeł związków organicznych: wewnętrzne (obecne w dopływających ściekach), zewnętrzne (nieobecne w ściekach dopływających) i endogenne (wytwarzane w systemie w wyniku obumierania organizmów).

Związki organiczne występujące w ściekach dopływających do oczyszczalni, oprócz podziału uwzględniającego ich stan fizyczny (tzn. związki rozpuszczone, koloidalne czy zawiesinowe), można podzielić również ze względu na szybkość biodegradacji. Związki nierozkładalne (nie biorące udział w procesach biologicznego oczyszczania ścieków) oraz rozkładalne (łatworozkładalne, S_S i wolnorozkładalne, X_S), które odgrywają istotną rolę w przebiegu procesów biochemicznych, zachodzących w komorach osadu czynnego. Wpływ związków łatworozkładalnych, do których należą m.in. lotne kwasy tłuszczowe (LKT), glukoza, etanol i metanol, jest dobrze rozpoznany, natomiast wciąż niewiele wiadomo na temat wpływu frakcji wolnorozkładalnej. Wprawdzie frakcja wolnorozkładalna została zdefiniowana w modelu Dolda i wsp. (1980), jednak dopiero znacznie później stało się oczywiste, że obejmuje ona szeroki zakres związków rozpuszczonych, koloidalnych i zawiesinowych (Orhon i Cokgor, 1997). Wspólną cechą związków wolnorozkładalnych jest to, że nie mogą być one bezpośrednio metabolizowane przez mikroorganizmy. Dlatego też, związki te są rozkładane przy udziale skomplikowanych reakcji określanych ogólnie jako proces hydrolizy, które przebiegają przy udziale enzymów zewnątrzkomórkowych, wewnątrz kłaczków osadu czynnego (Henze i wsp., 1987). Dopiero wówczas produkty hydrolizy, mogą być transportowane do komórek oraz wykorzystywane do przebiegu metabolizmu wewnątrzkomórkowego (Wanner, 1994). Z ostatnich eksperymentalnych obserwacji wynika, że hydroliza jest ponadto poprzedzona flokulacją (Jimenez i wsp., 2005).

Dostępne dane literaturowe na temat charakterystyki frakcji w ściekach dopływających do oczyszczalni, podsumowane przez Mąkinię (2010) wykazały, że w różnych krajach zawartość frakcji organicznej ulegającej biodegradacji w przeliczeniu na całkowite ChZT może być bardzo zróżnicowana i wahała się od 38% (Zele, Belgia) do 91% (Stambuł, Turcja). Z kolei, na podstawie ostatnich badań przeprowadzonych w czterech oczyszczalniach komunalnych na Pomorzu Gdańskim (ścieki oczyszczone mechanicznie) można stwierdzić, że w ściekach udział frakcji ChZT (w zawiesinie i koloidalnej), zawierającej zarówno związki wolnorozkładalne (X_S) jak i nierozkładalne (X_I), jest znaczący i wynosi ponad 60%.

Występujące w ściekach związki łatworozkładalne (S_S) mogą być bezpośrednio metabolizowane przez mikroorganizmy (w przeciwieństwie do X_S), jednak ich ilość jest zwykle ograniczona, co uniemożliwia efektywny przebieg denitryfikacji i EBPR. Lotne kwasy tłuszczowe (LKT) są wykorzystywane w komorze beztlenowej (znajdującej się w przedniej części bioreaktorów), przez co odpowiadają bezpośrednio za przetrwanie i rozwój w tej strefie bakterii fosforowych (PAO). Stężenie LKT w dopływających ściekach komunalnych jest zazwyczaj niskie. Z tego względu, podczas fermentacji „złożone" związki łatworozkładalne przekształcane są do LKT przy udziale „zwykłych" mikroorganizmów heterotroficznych. Proces przyczynia się do niemal całkowitego wykorzystania łatwo-rozkładalnych substratów w komorach beztlenowych. W kolejnych strefach anoksycznych, związki wolnorozkładalne stają się dominującym źródłem węgla dla denitryfikacji, przebiegającej przy udziale fakultatywnych mikroorganizmów heterotroficznych. Według Oleszkiewicza i wsp. (2004), niedobory organicznych związków węgla, które limitują przebieg procesu denitryfikacji, są jedną z podstawowych przyczyn niezadowalających efektów usuwania związków azotu w oczyszczalniach. Potwierdzają to również wyniki badań w obu trójmiejskich oczyszczalniach, które wykazały, że podstawową barierą dla uzyskania lepszej efektywności procesu usuwania azotu jest brak łatworozkładalnych związków organicznych. Szacunkowe stosunki S_S/N wynosiły 1,7-2,4 g ChZT/g N w oczyszczalni „Gdańsk-Wschód" i 2,7-2,9 g ChZT/g N w oczyszczalni „Gdynia-Dębogórze" (Mąkinia, 2006). Wartości te są niższe niż optymalna wartość 3,4 g ChZT/g N proponowana przez Kubę i wsp. (1996) dla układów ze zintegrowanym usuwaniem azotu i fosforu.

Z tego względu, dokładne rozpoznanie wpływu związków organicznych koloidalnych i zawiesinowych, na szybkość procesów biochemicznych zachodzących w komorze osadu czynnego, jest niezbędne dla optymalnego projektowania i eksploatacji systemów BNR opartych na technologii osadu czynnego. Jak dotąd, rzeczywisty wpływ tych związków nie został dostatecznie poznany, zarówno w zakresie teoretycznych rozważań (mechanizmy metabolizowania przez mikroorganizmy), jak i praktycznych aspektów (wpływ na wydajność procesów biochemicznych). Najnowsze publikowane w literaturze wyniki badań wpływu związków organicznych koloidalnych i zawiesinowych na szybkość w/w procesów biochemicznych zrealizowanych w pełnej skali są niejednoznaczne. Na przykład, Tuncal i wsp. (2009), w badaniach prowadzonych na dwóch równoległych, wydzielonych liniach (z i bez podstawowej sedymentacji) w oczyszczalni ścieków Izmir (Turcja), wykazali podczas rocznego okresu badawczego, że średnie stężenia NO_3-N i PO_4-P w ściekach oczyszczonych były niższe dla linii bez osadnika wstępnego (0,82 g N/m^3 i 1,7 g P/m^3) w porównaniu do równoległego ciągu z osadnikiem wstępnym (1,6 g N/m^3 i 3,8 g P/m^3). Według autorów te różnice mogę być spowodowane dwoma mechanizmami. Po pierwsze, wzrost szybkości procesów przy udziale związków w postaci zawiesinowej wspiera rozwój bakterii denitryfikacyjnych PAO. Po drugie, umieszczenie frakcji zawiesinowej w komorach beztlenowych pozwala na fermentację wstępnie przetworzonych związków organicznych oraz wspomaga wydajność procesu EBPR. Z kolei, Puig i wsp. (2010) stwierdzili, że bezpośrednie wprowadzanie dopływających ścieków surowych do reaktora biologicznego,

nie skutkowało poprawą jakości ścieków oczyszczonych, a zwłaszcza skutecznością usuwania substancji biogennych w badanych oczyszczalniach. Obserwowany w dopływie wzrost stosunku C/N i C/P został przypisany wysokiemu stężeniu frakcji nierozkładalnej ChZT oraz zawiesin z niskim wskaźnikiem ChZT/s.m.o.

Pomocnym narzędziem do oceny wpływu związków organicznych koloidalnych i zawiesinowych są modele matematyczne osadu czynnego (ASM). Wśród publikacji naukowych znaleźć można szereg przykładów zastosowania modeli matematycznych do badań nad wpływem różnych związków organicznych w procesach oczyszczania ścieków. Zastosowanie takich modeli dla systemów zintegrowanego usuwania biogenów wymaga określenia frakcji ChZT w zależności od ich podatności na biodegradację oraz wyznaczenia wartości współczynników kinetycznych i stechiometrycznych w wybranym modelu. Przegląd wczesnych procedur, mających na celu określenie charakterystyki ścieków, został przedstawiony m. in. w pracach Henze (1992) oraz Orhona i Cokgora (1997). Procedury takie były i są nadal rozwijane, szczególnie w odniesieniu do frakcji łatworozkładalnej S_S. W nowej wersji holenderskich wytycznych STOWA zaleca się strącanie chemiczne lub sączenie na membranie filtracyjnej o wielkości porów 0,1 µm, w celu wydzielenia frakcji łatworozkładalnej w ściekach (Roeleveld i van Loosdrecht, 2002). Strącanie chemiczne metodą Mamaisa i wsp. (1993), w prosty sposób pozwala, w wyniku koagulacji i flokulacji, wyznaczyć stężenie frakcji rozpuszczonej. Natomiast ChZT dla próby sączonej, $ChZT_{filt.}$ powinno określać stężenie frakcji rozpuszczonej. W rzeczywistości jednak ścieki sączone zawierają pewną ilość związków koloidalnych, które traktowane są jako związki wolnorozkładalne. Na przykład, udział frakcji koloidalnej w ChZT dla próby sączonej na filtrze z włókna szklanego (Whatman GF/C) o wielkości porów 1,2 µm wynosi średnio około 45% (Henze i wsp., 1994). Nawet zastosowanie membrany filtracyjnej o wielkości porów 0,45 µm czy 0,1 µm nie zapobiega całkowicie przedostawaniu się do filtratu cząstek koloidalnych.

Dokonując przeglądu metod pomiarów frakcji wolnorozkładalnej w ściekach oraz jej modelowania, można zauważyć, że w początkowych modelach procesu oczyszczania ścieków, pojęcie „hydrolizy", było rozumiane jedynie jako przemiana substratu wolnorozkładalnego w formę łatworozkładalną (Henze i wsp., 1987; Henze 1992). W rzeczywistości proces hydrolizy uwzględnia wszystkie mechanizmy, które dotyczą podziału złożonych związków organicznych za pomocą enzymów zewnątrzkomórkowych i ich przekształcenia do prostszych związków. Identyfikacja i opis tych procesów nie jest możliwy, dlatego też są one zebrane w wygodny sposób jako jeden mechanizm hydrolizy (Insel i wsp., 2002). Stopień hydrolizy jest stosowany do wyrażania skumulowanych efektów innych kompleksowych reakcji. Począwszy od fizycznego pobierania i adsorpcji, przez okres magazynowania związków organicznych, aż do biochemicznego ich utleniania i syntezy (Orhon i wsp., 1999). Należy jednak pamiętać, że w zależności od zastosowanego modelu procesu osadu czynnego, przebieg dalszych przemian substratu łatworozkładalnego (S_S) może być różny. W ASM1 następuje bezpośredni wzrost na substracie pierwotnym (S_S), biomasy żywych bakterii (X_H), które następnie obumierają, tworząc ponownie substrat

wolnorozkładalny (X_S). Natomiast w ASM3 wzrost nowej biomasy jest poprzedzony etapem magazynowania substratu łatworozkładalnego, bez uwzględniania produkcji X_S z rozkładu obumarłej biomasy bakterii.

Istotny rozwój wiedzy w zakresie procesów usuwania związków biogennych przyczynił się do opracowania kilku kompleksowych modeli procesu osadu czynnego (oprócz w/w także ASM2 i ASM2d). Model osadu czynnego ASM2 stanowi rozszerzenie ASM1, tzn. oprócz modelowania procesów usuwania związków organicznych i azotu dodatkowo został uwzględniony proces usuwania fosforu na drodze biologicznej i chemicznej. W efekcie dalszych badań powstały modyfikacje obu modeli, tzw. ASM2d i ASM3, które z kolei są ostatnio szeroko wykorzystywane w programach symulacyjnych, służących m.in. do projektowania i optymalizacji pracy oczyszczalni ścieków. Według Orhona i wsp. (1997) modele osadu czynnego, uwzględniające usuwanie związków organicznych i biogennych, uwzględniają kinetykę procesu hydrolizy z X_S jako składnikiem modelu. W fachowej literaturze istnieje wiele równań opisujących proces hydrolizy. Są to równania począwszy od najprostszych (0 czy I rzędu) do bardziej skomplikowanych równań zawierających wiele członów Monoda.

Dotychczasowy stan wiedzy wskazuje na to, że szybkość hydrolizy jest zróżnicowana także w zależności od rodzaju akceptorów elektronów dostępnych w środowisku. Henze i Mladenovski (1991) zaobserwowali największą szybkość hydrolizy w warunkach beztlenowych, która znacznie obniżała się kolejno w warunkach tlenowych i anoksycznych. Morgenroth i wsp. (2002) stwierdzili, że inne złożone reakcje, takie jak rozkład chemiczny i procesy transportu masy, powinny być również brane pod uwagę przy ocenie szybkości hydrolizy. Najnowsze badania prowadzone w tym kierunku wskazują nawet, że proces hydrolizy jest wolniejszy niż heterotroficzny wzrost i zazwyczaj staje się etapem ograniczającym szybkość biodegradacji związków organicznych.

Celem niniejszej rozprawy doktorskiej było określenie wpływu substratu wolnorozkładalnego (w postaci związków koloidalnych i zawiesinowych) na kinetykę wybranych procesów biochemicznych, zachodzących w komorach osadu czynnego (tj. denitryfikacja, anoksyczny/tlenowy pobór fosforanów i pobór tlenu), opierając się na badaniach prowadzonych w dwóch dużych komunalnych oczyszczalniach ścieków „Wschód" w Gdańsku i „Dębogórze" w Gdyni. Z uwagi na brak metody, umożliwiającej bezpośrednie wyznaczanie stężenia substratu wolnorozkładalnego w ściekach, została opracowana i wdrożona procedura oparta na prowadzeniu pomiarów równolegle w dwóch reaktorach nieprzepływowych (RI i RII). W tym celu wykorzystano osad czynny, pochodzący ze stopnia biologicznego obu oczyszczalni oraz ścieki po oczyszczaniu mechanicznym bez podczyszczania (RI – zawierający wszystkie frakcje ChZT) oraz po koagulacji i flokulacji (RII – wyłącznie frakcje rozpuszczone bez X_S). Przygotowanie scieków w RII odbyło się metodą Mamaisa i wsp. (1993). Na podstawie porównania szybkości procesów w obu reaktorach możliwa była ocena wpływu X_S na szybkości następujących

procesów biochemicznych: denitryfikacji (NUR), poboru tlenu (OUR), uwalniania fosforanów (PRR) w fazie beztlenowej i poboru fosforanów w fazie anoksycznej. Ocenę zdolności predykcyjnych modelu ASM2d przeprowadzono w oparciu o wyniki badań laboratoryjnych i pomiarów terenowych na bioreaktorze MUCT w oczyszczalni „Wschód". Ponadto wyniki badań laboratoryjnych wykorzystane zostały do zweryfikowania mechanizmu procesu hydrolizy związków w modelu ASM2d i opracowania zmodyfikowanej wersji modelu (dwustopniowa hydroliza).

Badania przeprowadzone w ramach tej rozprawy doktorskiej pozwolą szerzej wyjaśnić czy związki X_S, które w znacznych ilościach występują w ściekach, mogą wpłynąć na efektywne usuwanie związków biogennych i zrekompensować niedobory S_S. Kontynuowanie badań w pełnej skali może również dać istotną odpowiedź na pytanie, czy strącanie chemiczne w celu usunięcia ze ścieków związków koloidalnych i zawiesinowych, jest korzystne dla prawidłowego przebiegu procesów biochemicznych w komorze osadu czynnego. Wpływ koloidalnych i zawiesinowych związków organicznych ma istotne znaczenie dla optymalizacji chemicznego strącania oraz sedymentacji w osadnikach wtórnych, tj. dla osiągnięcia właściwej równowagi, pomiędzy efektywnością usuwania związków biogennych (N, P) w bioreaktorze, a produkcją biogazu w komorze fermentacyjnej.

Metodyka badań

Do pomiarów szybkości wybranych procesów biochemicznych zastosowano zestaw doświadczalny, składający się z dwóch równoległych reaktorów nieprzepływowych o pojemności 4 dm^3 każdy, sterownika programowalnego oraz komputera. Każdy z reaktorów wyposażony był w systemy regulacji temperatury i pomiaru stężenia tlenu rozpuszczonego, a także w sondy do pomiaru pH i potencjału redox. Podczas jednego cyklu pracy reaktora laboratoryjnego, który odzwierciedla konfigurację wielofazowej komory osadu czynnego (faza beztlenowa, anoksyczna i/lub tlenowa) możliwy był pomiar on-line i zapis danych podczas wybranych testów NUR, PRR i anoksyczny/tlenowy PUR oraz OUR. Każdorazowo przed rozpoczęciem doświadczenia, ze ścieków oczyszczonych mechanicznie wydzielano pewną objętość (zazwyczaj ok. 2,5-3 dm^3), którą poddawano koagulacji i flokulacji metodą Mamaisa, w celu wyznaczenia stężenia rzeczywistej frakcji rozpuszczonej ChZT ($ChZT_{rozp.}$).

Przygotowanie próbek ścieków oczyszczonych mechanicznie ***metodą Mamaisa (zwaną też "metodą koagulacyjną")*** rzeczywistej frakcji $ChZT_{rozp}$ zakłada strącenie koloidów za pomocą $Zn(OH)_2$ przy pH=10,5. Na koniec przeprowadzono korektę pH otrzymanej cieczy nadosadowej (do stanu początkowego) przy użyciu HCl.

„Konwencjonalne" pomiary NUR

„Konwencjonalne" pomiary szybkości denitryfikacji (NUR) prowadzone były w reaktorze nieprzepływowym. Każdy z dwóch równoległych reaktorów wyposażony był w mieszadło

mechaniczne oraz elektrody do pomiaru „on-line" stężenia tlenu rozpuszczonego, potencjału redox oraz wartości pH i temperatury. Do badań wykorzystywano osad recyrkulowany ze stopnia biologicznego oczyszczalni ścieków „Wschód" i „Dębogórze", który rozcieńczany był ze ściekami oczyszczonymi mechanicznie bez podczyszczania (RI) lub podczyszczonymi metodą koagulacji i flokulacji (RII). Doświadczenia prowadzono przez okres 4 h od momentu dodania azotanu potasu (KNO_3). Próbki z reaktora pobierane były z częstotliwością 2-30 min, filtrowane i poddawane analizom laboratoryjnym w celu określenia stężeń PO_4-P, NO_3-N i ChZT.

Pomiary PRR i anoksycznego/tlenowego PUR

Na początku testu osad czynny mieszany był ze ściekami oczyszczonymi mechanicznie bez podczyszczania (RI) lub podczyszczonymi metodą koagulacji i flokulacji (RII). W trakcie fazy beztlenowej mierzona była szybkość uwalniania fosforanów (PRR), natomiast w trakcie fazy anoksycznej/tlenowej mierzona była szybkość poboru fosforanów (PUR) oraz odpowiednio NUR/AUR. Czas trwania fazy beztlenowej i anoksycznej/tlenowej wynosił odpowiednio 2,5 h i 4 h. Na początku fazy anoksycznej/tlenowej, do reaktora dodawano odpowiednio KNO_3/O_2 w celu podniesienia stężenia azotanów/tlenu w reaktorze. Próbki z reaktora pobierane były z częstotliwością 2-30 min, filtrowane i poddawane analizom laboratoryjnym w celu określenia stężeń PO_4-P i ChZT (faza beztlenowa) oraz ChZT i PO_4-P, NO_3-N/NH_4-N (faza anoksyczna/tlenowa).

Pomiar szybkości poboru tlenu (z ang. OUR)

W doświadczeniu utrzymywano stężenie tlenu rozpuszczonego na poziomie ok. 6 mg O_2/dm^3. Na początku testu dodawano ok. 30 mg inhibitora nitryfikacji (ATU), aby zatrzymać pobór tlenu przez bakterie nitryfikacyjne. Próbki do badań o objętości 50 cm^3 pobierano z częstotliwością co 5-30 min, filtrowano przez sączki typu Whatman GF/C i poddawano analizom laboratoryjnym w celu określenia stężenia ChZT. Natomiast szybkość poboru tlenu (OUR) była mierzona automatycznie (co 3 min) za pomocą sond tlenowych, umieszczonych w oddzielnych komorach, aż do zakończenia doświadczenia (tj. po czasie 5-7 h). Pomiary całkowitej szybkości poboru tlenu ($\int$OUR(t)dt) i stopnia rozkładu związków organicznych (ΔChZT), pozwoliły wyznaczyć wartość współczynnika przyrostu bakterii heterotroficznych (Y_H):

$$Y_H = \frac{\Delta ChZT - \int_{t0}^{te} OUR_{net} \cdot V dt}{\Delta ChZT} \qquad (1)$$

gdzie:

Y_H *–współczynnik przyrostu bakterii heterotroficznych, mg ChZT (biomasa)/mg ChZT (substrat)*

$\Delta ChZT$ *– różnica stężeń substratu na początku i na końcu doświadczenia, mg* O_2/dm^3

t_0/te *– początkowy/końcowy czas doświadczenia, h*

OUR_{net} *– szybkość poboru tlenu netto (bez uwzględnienia endogennej respiracji) mg* $O_2/(dm^3 \cdot h)$

V *– pojemność reaktora,* dm^3

Modelowanie matematyczne i symulacje komputerowe

Na podstawie koncepcji, uwzględniającej dwustopniową hydrolizę w warunkach beztlenowych, anoksycznych i tlenowych, opracowany został nowy model osadu czynnego jako modyfikacja modelu ASM2d. Nowy model uwzględnia jedną nową zmienną (X_{SH}) oraz trzy nowe procesy, tj. hydrolizę X_{SH} w warunkach beztlenowych, anoksycznych i tlenowych. Frakcja X_{SH} została zdefiniowana jako „pośrednia forma hydrolizy". Do obliczeń symulacyjnych wykorzystano program komputerowy GPS-X ver. 5.0.2 (Hydromantis, Kanada). Nowy model został wdrożony za pomocą specjalnego modułu o nazwie „Model Developer".

Metody analityczne

Zawiesina ogólna i organiczna była mierzona metodą grawimetryczną według Polskich Norm (PN-72/C-04559). ChZT, PO_4-P, NO_3-N, NH_4-N oznaczano metodą testów kuwetowych na spektrofotometrze Xion 500 (Dr Lange GmbH, Niemcy). Zastosowane w pracach badawczych procedury analityczne, zaadoptowane przez firmę Dr Lange GmbH, bazowały na metodach standardowych APHA (APHA, 1992).

Wyniki badań

W oczyszczalni ścieków „Wschód" w Gdańsku, w średniodobowej próbce ścieków po oczyszczeniu mechanicznym, frakcja rozpuszczona stanowiła 19-39% całkowitego ChZT. Średnia wartość całkowitego ChZT wynosiła 627 (±81) g ChZT/m^3, w tym frakcje rozpuszczone ChZT = 194 (±38) g ChZT/m^3 oraz nierozpuszczone (koloidalne i zawiesinowe) ChZT = 433 (±73) g ChZT/m^3. Dla porównania, średnie wartości całkowitego ChZT oraz frakcji rozpuszczonych z rutynowych analiz w oczyszczalni „Wschód" nieznacznie odbiegały od powyższych stężeń, tj. 594 i 172 g ChZT/m^3 (2007 rok) oraz 715 i 192 g COD/m^3 (2008 rok). Z kolei w oczyszczalni ścieków „Dębogórze" w Gdyni, rozpuszczona frakcja wyznaczona w 16 próbkach ścieków po oczyszczaniu mechanicznym stanowiła 23-46% całkowitego ChZT. W tym wypadku średnia wartość całkowitego ChZT wynosiła 533 (± 86) g ChZT/m^3, w tym rozpuszczone COD = 211 (±32) g ChZT/m^3 oraz nierozpuszczone COD = 322 (±95) g COD/m^3.

Oszacowano, na podstawie badań charakterystyki ścieków, że stosunek frakcji (S_S/(S_S + X_S)) na oczyszczalni ścieków „Wschód" wyniósł 0,32-0,40 i mieścił się w zakresie 0,3-0,5 (Sahlstedt i wsp., 2003). Natomiast zakres tego wskaźnika w oczyszczalnia „Dębogórze" (0,50-0,54) został nieznacznie przekroczony w porównaniu do powyższych danych literaturowych. Ponadto Pagilla i wsp. (2008) opublikowali wyniki wartości ChZT w ściekach w obu oczyszczalniach po filtracji na sączkach o różnej wielkości porów. Dominująca część związków organicznych była usuwana na sączkach o wielkości porów > 1,2 µm, co stanowiło odpowiednio 67 i 75% całkowitego ChZT, odpowiednio, w oczyszczalni ścieków „Wschód" i „Dębogórze".

„Konwencjonalne" pomiary NUR

W czasie testów ze ściekami po oczyszczaniu mechanicznym (RI), obserwowano podwójne szybkości denitryfikacji (NUR1 i NUR2), które były związane z wykorzystaniem rozpuszczonych, łatworozkładalnych związków organicznych (tylko NUR1) i wolnorozkładalnych (rozpuszczonych, koloidalnych i zawiesinowych) związków organicznych (NUR2). W okresie badań, średnie wartości NUR1, jakie otrzymano odpowiednio dla oczyszczalni ścieków „Wschód" i „Dębogórze", mieściły się w zakresie 3,2-5,3 i 3,2-3,6 mg N/(g s.m.o. ·h). Średnie zużycie związków organicznych (wyrażone stosunkiem Δ ChZT: Δ N) związane z NUR1 wahało się w granicach od 5,1-10,4 („Wschód") i 6,4-8,1 („Dębogórze"). Średnie wartości NUR2 w równoległych doświadczeniach, otrzymano w zakresach zbliżonych dla obu oczyszczalni, tj. 1,5-1,8 mg N/(g s.m.o. ·h) („Wschód") oraz 1,7-2,0 mg N/(g s.m.o.·h) („Dębogórze"). Obserwowane wartości NUR1 i NUR2 w obu badanych oczyszczalniach są porównywalne do zakresów przedstawionych przez Naidoo i wsp. (1998) dla podobnych doświadczeń wykonanych w ośmiu komunalnych oczyszczalniach w Europie. W badaniu tym otrzymano także podwójne wartości NUR w zakresie 3,3-5,7 (NUR1) i 1,6-3,6 mg N/(g s.m.o.·h) (NUR2).

Porównując wyniki doświadczeń (związanych z wykorzystaniem pozostałych frakcji rozpuszczonej) uzyskanych dla ścieków oczyszczonych mechanicznie po koagulacji i flokulacji (RII) zaobserwowano niższe wartości NUR. W tym przypadku wartości NUR1 wahały się w zakresie 2,4-3,4 g N/(kg s.m.o.·h) („Wschód") oraz 2,7-2,9 g N/(kg s.m.o.·h) („Dębogórze"), natomiast wartości NUR2, w analogicznym eksperymencie, w zakresie 1,0-1,2 g N/(kg s.m.o.·h) („Wschód") i 1,1-1,2 g N/(kg s.m.o.·h) („Dębogórze"). Obliczone stosunki Δ ChZT: Δ N (związane z NUR1) wahały się od 5,5 do 8,5 („Wschód") i od 7,3 do 8,6 („Dębogórze"). Usunięcie koloidalnych i zawiesinowych związków organicznych spowodowało zmniejszenie szybkości denitryfikacji odpowiednio o 21-37% i 24-28% odpowiednio dla oczyszczalni ścieków „Wschód" i „Dębogórze".

Oprócz NO_3-N i ChZT, badano także zachowanie fosforanów w czasie pomiarów NUR. W początkowej fazie, w obu reaktorach (RI i RII), występowało uwalnianie fosforanów, pomimo wysokiego stężenia azotanów (ponad 10g NO_3-N/m^3), które według powszechnej opinii, powinno hamować ten proces. Co więcej, uwalnianie P było kontynuowane aż do momentu zużycia związków Ss w reaktorze. W zależności od temperatury w danym okresie badawczym (wiosna, lato, zima), stężenia biomasy i początkowego stężenia ChZT w reaktorach, czas uwalniania P był różny (od ok. 0,5-2,5 h). Bardzo podobne zachowanie fosforanów było obserwowane przez Brdjanovica i wsp. (2000) w teście NUR przeprowadzonym z octanem jako źródłem węgla. W tym doświadczeniu znaczne ilości fosforanów zostały uwolnione (z 2 g P/m^3 do prawie 10 g P/m^3) w ciągu pierwszej godziny testu aż do całkowitego zużycia octanu. Jednocześnie stężenie NO_3-N zmniejszyło się z około 25 do 10 g N/m^3. Obserwacje te są zgodne z danymi opublikowanymi przez Yuan i

Oleszkiewicza (2008), którzy wykazali, że uwalnianie fosforanów trwa do momentu wykorzystania przez bakterie osadu czynnego łatworozkładalnych związków organicznych, niezależnie od stężenia NO_3-N w fazie beztlenowej.

Pomiary PRR i anoksycznego/tlenowego PUR

Podczas testów dwufazowych ze ściekami oczyszczonymi mechanicznie bez podczyszczania najwyższe wartości PRR wahały się w granicach 10,5-11,2 mg P/(g s.m.o.·h) w oczyszczalni „Wschód" i 7,9-9,6 mg P/(g s.m.o.·h) w oczyszczalni „Dębogórze". Proces koagulacja-flokulacja również nie miał wyraźnego wpływu na PRR. Jak wynikało z obserwacji szybkości procesów w równoległych reaktorach wahały się w zakresie około ± 20%. Średnia szybkość uwalniania P w przeliczeniu na s.m.o. była podobna w obu reaktorach, tj. 13,7-14,0 g P /g s.m.o. („Wschód") i 10,8-11,3 g P/g s.m.o. („Dębogórze"). Fosfor zgromadzony w komórkach PAO nie został całkowicie uwolniony pod koniec fazy beztlenowej. W poprzednich doświadczeniach (Mąkinia, 2006), szybkość uwalniania P w obecności octanu była wyższa o 40-70% w porównaniu do testów ze ściekami oczyszczonymi mechanicznie bez podczyszczania. Wydaje się więc, że beztlenowa hydroliza związków organicznych Xs generuje niewielkie ilości substratu dla utrzymania PAO w warunkach beztlenowych.

Otrzymane wartości PRR w obu oczyszczalniach mieściły w zakresie wartości podawanych w literaturze (4,4-18,8 mg P/(g s.m.o.·h)), dla podobnych testów przeprowadzonych w różnych systemach osadu czynnego z biologicznym usuwaniem związków biogennych, zarówno w skali technicznej (Kuba i wsp., 1997 ; Sorm i wsp., 1998;. Tuncal i wsp., 2009;. Puig i wsp., 2010), jak i pilotowej (Petersen i wsp., 1998; Tykesson i wsp., 2002). W badaniach przeprowadzonych przez Tuncala i wsp. (2009), PRR = 10,8 i 16,2 mg P/(g s.m.o.·h), odpowiednio dla próbek pobranych z dwóch równoległych linii, bez i z osadnikiem wstępnym. Puig i wsp. (2010) stwierdzili odwrotny efekt przy pominięciu wstępnej sedymentacji. Obserwowane wartości PRR zmniejszył się z 18,8 mg P/(g VSS · h) w trakcie normalnego działania do 13,6 mg P/(g s.m.o.· h) z pominięciem osadników.

Średni stosunek uwalnianego PO_4-P do wykorzystywanego ChZT przez mikroorganizmy w procesie uwalniania P (Y_{PO4}), mierzony w początkowej fazie beztlenowej, był podobny w obu reaktorach i wahał się w zakresie 0,29-0,35 g P/g ChZT („Wschód") i 0,34 -0,59 g P/g ChZT („Dębogórze"). Wartości te tylko nieznacznie różniły się od typowego zakresu (0,35-0,5) dla systemów osadu czynnego. Niskie wartości Y_{PO4} mogą wynikać z długiego wieku osadu lub obecności bakterii kumulujących glikogen (GAO) (Brdjanovic i wsp., 2000). Ponadto, rodzaj substratu mógł być kolejnym czynnikiem wpływającym na Y_{PO4}. We wcześniejszym badaniach (Mąkinia, 2006), wartości Y_{PO4} były znacznie wyższe (o około 35-40%) dla octanu w porównaniu do ścieków oczyszczonych mechanicznie bez podczyszczania. Także Ubukata (2005) stwierdził, że związki organiczne obecne w rzeczywistych ściekach przyczyniają się do różnej szybkości uwalniania fosforu niż w przypadku dodania octanu.

Podczas testów dwufazowych ze ściekami oczyszczonymi mechanicznie bez podczyszczania, najwyższe wartości PRR wahały się w granicach 6,1-11,4 i 5,6-5,9 mg P/(g s.m.o.·h) wobec 3,8-11,0 i 4,9-5,0 mg P/(g s.m.o.·h) ze ściekami po koagulacji-flokulacji, odpowiednio, dla oczyszczalni ścieków „Wschód" i „Dębogórze". Wartości PUR w warunkach anoksycznych były znacznie niższe, tj. 4.7-6.8 i 1.8-2.0 mg P/(g s.m.o.·h) w testach ze ściekami oczyszczonymi mechanicznie bez podczyszczania oraz 1.2-6.7 i 1.2-1.7 mg P/(g VSS ·h) dla ścieków po koagulacji-flokulacji. Obliczony stosunek zużycia azotanów do fosforanów był zróżnicowany w zakresie 0,51-0,78 g N/g P i 0,74-1,41 g N/g P w testach ze ściekami oczyszczonymi mechanicznie bez podczyszczania wobec 0,47-1,74 g N/g P i 0,83-1,01 g N/g P ze ściekami po koagulacji-flokulacji, odpowiednio, dla oczyszczalni ścieków „Wschód" i „Dębogórze". Dla porównania, w badaniach Tuncala i wsp. (2009) stosunek N/P wyniósł 0,4 g N/g P dla ciągu bez osadnika wtórnego i 0,5 g N/g P dla ciągu z osadnikiem wtórnym. Dla porównania, literaturowe wartości PUR w warunkach anoksycznych i tlenowych były zróżnicowane i wahały się w zakresie 1,9-13 i 3,6-30 mg P/(g VSS·h) (Kuba i wsp., 1997; Sorm i wsp., 1998; Tuncal i wsp., 2009; Puig i wsp., 2010). W badaniach Tuncala i wsp. (2009) oraz Puiga i wsp. (2010), obserwowane anoksyczne i tlenowe PUR były w przybliżeniu równe, jednak wpływ osadnika wstępnego w tych doświadczeniach był niejednoznaczny. Tuncal i wsp. (2009) zaobserwowali znaczny wzrost PUR (o 70%) dla ciągu bez osadnika wstępnego w porównaniu do ciągu z osadnikiem wstępnym. Natomiast pominięcie osadnika wstępnego w badaniu Puiga i wsp. (2010) spowodowało niewielki spadek wartości PUR (o 15%), prawdopodobnie ze względu na krótszy wiek osadu i wyższe stężenie nierozkładalnych frakcji organicznych w sciekach.

Podczas testów dwufazowych z wykorzystaniem ścieków oczyszczonych po koagulacji i flokulacji, wartości NUR były niższe o około 22-44% (dla fazy anoksycznej), a tylko o 3-20% niższe w odniesieniu do pomiarów OUR (dla fazy tlenowej). Nieznaczne obniżenie wartości w fazie tlenowej w przypadku testów z wykorzystaniem ścieków po koagulacji i flokulacji, jest wynikiem podobnego efektu nitryfikacji w obu reaktorach (proces ten nie został zakłócony przez usunięcie cząstek zawiesinowych i koloidalnych). Dla porównania, Tuncal i wsp. (2009), zaobserwowali podobny poziom (25%) redukcji NUR mierzony w ciągu z osadnikiem wstępnym w stosunku do ciągu bez osadnika (1,8 wobec 2,4 mg N/(g s.m.o.· h)).

Pomiary OUR

Maksymalna wartość OUR w pierwszej fazie wahała się w zakresie 22,8-39,5 i 20,4-21,8 g O_2/(kg s.m.o.·h), odpowiednio, w oczyszczalni „Wschód" i „Dębogórze". Różnice między pomiarami w równoległych reaktorach osiągały wartości do 20-30%. Dla porównania, Choi i Daehwan (2001) stwierdzili, że zawiesinowe frakcje ChZT, które stanowiły 65% całkowitego ChZT (z czego ok. 50% określono jako rozkładalne) przyczyniły się do wzrostu całkowitego OUR (łącznie z nitryfikacją) o około 5,5%. W innym badaniu, Kristensen i wsp. (1992) wykazali, że hydroliza osadu wstępnego może produkować związki łatworozkładalne. Autorzy stwierdzili to na podstawie obserwacji, że wartości OUR z hydrolizatem (osadem

wstępnym w postaci ciekłej, który hydrolizowano w warunkach beztlenowych) jako źródłem węgla były zazwyczaj 10-20% wyższe w porównaniu do wartości OUR dla octanu.

Wartości współczynników przyrostu bakterii heterotroficznych (Y_H), uzyskano z równania 1 w oparciu o jednoczesne pomiary ChZT i szybkości poboru tlenu. Wartości współczynnika Y_H wahały się w zakresie 0,62-0,67 i 0,68-0,71 g ChZT (biomasa)/ g ChZT (substrat), odpowiednio, w oczyszczalni „Wschód" i „Dębogórze". Sugeruje to, że skład ścieków oczyszczonych mechanicznie mógł nieznacznie różnić się w przypadku obu oczyszczalni. Typowe wartości Y_H dla ścieków komunalnych mieściły się w przedziale 0,46-0,69 g ChZT/ g ChZT (Henze i wsp. (1987)). Domyślna wartość Y_H w Modelu Osadu Czynnego nr 1 (ASM1) została przyjętą na poziomie 0,67 g ChZT/g ChZT. Wyższy zakres wartości Y_H (0,72-0,78 g ChZT/g ChZT) stwierdzono dla oczyszczalni ścieków „Wschód" w wyniku dozowania zewnętrznych źródeł węgla (octan, etanol, olej fuzlowy i destylowany alkohol surowy). Dricks i wsp. (1999) udowodnili, że różnica współczynnika Y_H rzeczywiście może wynikać z rodzaju substratu. Wyznaczyli oni współczynniki Y_H dla osadu czynnego z dwóch duńskich oczyszczalni ścieków, wykorzystując pomiary szybkości zużycia substratu i poboru tlenu. Uzyskane przez autorów wartości współczynnika Y_H wyniosły 0,71-0,72 g ChZT/ g ChZT dla octanu oraz 0,66-0,67 g ChZT/ g ChZT dla etanolu.

Modelowanie matematyczne i symulacje komputerowe

Pierwszy etap badań związanych z modelowaniem obejmował kalibrację/weryfikację oryginalnego modelu ASM2d w warunkach dynamicznych, na podstawie wyników badań laboratoryjnych w reaktorze nieprzepływowym ze ściekami oczyszczonymi mechanicznie (bez podczyszczania) oraz wyników 96-godzinnej kampanii pomiarowej w bioreaktorze MUCT w oczyszczalni „Wschód". W przypadku współczynników stechiometrycznych przyjęto wartości domyślne, za wyjątkiem współczynnika Y_H oraz współczynnika uwalniania polifosforanów na jednostkę zmagazynowanego PHA (Y_{PO4}). Wartość Y_H równą 0,65 i 0,68 mg ChZT/mg ChZT wyznaczono eksperymentalnie na podstawie testów OUR odpowiednio, dla oczyszczalni „Wschód" i „Dębogórze". Natomiast wartość Y_{PO4} równą 0,32 mg P/mg ChZT wyznaczono w oparciu o wcześniejsze pomiary beztlenowego uwalniania fosforanów (Mąkinia i wsp., 2009; Swinarski i wsp. 2009a, 2009b). Przyjęta wartość Y_{PO4} różni się od wartości domyślnej w ASM2d wynoszącej 0,40 mg P/mg ChZT, ale mieści się w typowym zakresie wartości stosowanych w modelach osadu czynnego (0,3-0,43 mg P/mg ChZT) (Mąkinia i wsp., 2006). Proces nitryfikacji został skalibrowany poprzez zmianę wartości trzech parametrów kinetycznych: stałej szybkości wzrostu bakterii nitryfikacyjnych μ_A, stałej nasycenia $K_{NH4,A}$ dla NH_4-N oraz stałej nasycenia $K_{PO4,A}$ dla PO_4-P. Oszacowane wartości dla μ_A i $K_{NH4,A}$ były wyższe od wartości domyślnych w ASM2d. Wyższe wartości $K_{NH4,A}$ wynikają z ograniczenia dyfuzji powodowanej niską turbulencją i dużymi kłaczkami osadu czynnego (Henze i wsp., 2000a). W przypadku $K_{PO4,A}$ wartość współczynnika została zmniejszona z 0,01 do 0,001 mg P/dm^3. Zmniejszenie wartości $K_{PO4,A}$ było konieczne, aby prognozować wyższe szybkości nitryfikacji przy bardzo niskich stężeniach PO_4-P, które

okresowo obserwowane są w strefie tlenowej bioreaktora w skali pełnotechnicznej (Meijer i wsp., 2001). Kalibracja procesu denitryfikacji została przeprowadzona w oparciu o wyniki „konwencjonalnych" pomiarów NUR oraz wyniki pomiarów NUR w testach dwufazowych, zmieniając stałą szybkości tempa wzrostu heterotrofów (μ_H) i stałą szybkości hydrolizy (k_{hyd}). W testach dwufazowych proces denitryfikacji skalibrowany został dodatkowo za pomocą współczynnika redukcji anoksycznego wzrostu PAO ($\eta_{NO3,PAO}$ = 0,5). Wartość tego współczynnika została zmniejszona w stosunku do wartości domyślnej (0,6). Uwalnianie fosforanów skalibrowano za pomocą pięciu parametrów: stałej szybkości magazynowania PHA q_{PHA}, stałej nasycenia dla PAO w odniesieniu do produktów fermentacji $K_{SA,PAO}$, stałej nasycenia dla PAO w odniesieniu do polifosforanów K_{PP}, współczynnika redukcji hydrolizy beztlenowej η_{fe} oraz stałej nasycenia K_X dla ChZT w zawiesinie. Za wyjątkiem K_X, wartości tych współczynników zostały zmniejszone w porównaniu do wartości domyślnych w ASM2d. Pobór fosforanów został skalibrowany przy użyciu trzech parametrów: stałej szybkości magazynowania polifosforanów q_{PP}, współczynnika inhibicji magazynowania polifosforanów K_{IPP} oraz stałej nasycenia K_{PHA} dla PHA. Należy podkreślić, że zmienione wartości q_{PHA} i q_{PP} były znacznie wyższe niż wartości domyślne tych parametrów w ASM2d. Wartość q_{PHA} wynosząca 6/d mieści się w zakresie wartości 6-8/d podawanych w literaturze (Mąkinia i wsp., 2006). W przypadku poboru fosforanów wyższa szybkość procesu, wynikająca z podwyższonej wartości q_{PP}, ograniczona została poprzez podniesienie wartości współczynnika inhibicji dla magazynowania fosforanów K_{IP} oraz stałej nasycenia K_{PHA} dla PHA. Podobnie jak w przypadku procesu nitryfikacji, ograniczenie szybkości procesu poboru fosforanów powodowane bardzo niskim stężeniem PO_4-P i NH_4-N wyeliminowane zostało poprzez ustalenie wartości stałej nasycenia $K_{NH4,PAO}$ dla NH_4-N i $K_{PO4,PAO}$ dla PO_4-P odpowiednio na poziomie 0,01 mg N/dm^3 oraz 0,001 mg P/dm^3.

Drugi etap badań związanych z modelowaniem obejmował opracowanie i wdrożenie nowego modelu (modyfikacji ASM2d) w pakiecie symulacyjnym GPS-X wersja 5.02 (Hydromantis, Kanada). Wyniki badań laboratoryjnych testów OUR ze ściekami oczyszczonymi mechanicznie (bez podczyszczania) zostały wykorzystane do wykonania optymalizacji parametrów metodą simplexu (Nelder-Mead) w zmodyfikowanym modelu ASM2d. Wyniki badań Mąkini i Czerwionki (2009) posłużyły do oszacowania ilości nowego składnika X_{SH} w zmodyfikowanym modelu ASM2d. Opierając się na podziale frakcji ChZT zaproponowanym przez Melcera i wsp. (2003) ustalono średnią wartość frakcji koloidalnej /rozpuszczonej ($X_{SH(C/S)}$ = 12% X_S) dla testów OUR ze ściekami oczyszczonymi mechanicznie (bez podczyszczania). W zmodyfikowanym modelu, symulacje po zmianie trzech wybranych parametrów (z pięciu początkowych): stałej szybkości hydrolizy (k_{hyd}) dla X_S, stałej szybkości hydrolizy ($k_{hyd,r}$) dla X_{SH}, współczynnika saturacji hydrolizy (K_x) dla X_S najlepiej odwzorowywały wyniki badań empirycznych, generując najmniejszy błąd względny. Parametry te zostały automatycznie skalibrowane za pomocą modułu „*Optimizer*" w programie GPS-X dla pozostałych serii doświadczalnych (w okresie zimowym, letnio-jesiennym, wiosennym) w obu oczyszczalniach. Ostatecznie przyjęto średnie wartości trzech w/w parametrów kinetycznych, ujednoliconych dla obu oczyszczalniach. Z kolei dla

pozostałych współczynników kinetycznych w nowym modelu (K_{xr}, $\eta_{NO3,hyd,r}$) przyjęto te same wartości, jak dla odpowiadających im współczynników w oryginalnym modelu ASM2d. Współczynniki stechiometryczne, Y_H i Y_{PAO}, zostały ustalone w oparciu o wyniki testów szybkości poboru tlenu. W przypadku próbki z podczyszczonymi ściekami metodą koagulacji-flokulacji, niezbędne było poszukiwanie wartości frakcji rozpuszczalnej. W celu jej dopasowania do wyznaczonych wcześniej współczynników kinetycznych i stechiometrycznych w zmodyfikowanym modelu ASM2d, wykonano symulacje dla testów OUR ze ściekami oczyszczonymi mechanicznie. Ostatecznie średnia wartość $X_{SH(S)}$ wyniosła 6% X_S dla wszystkich przeprowadzonych serii testów. Następnie, dokonano porównania zdolności predykcyjnych oryginalnego i zmodyfikowanego modelu ASM2d dla porównania pozostałych testów NUR, PRR i anoksycznego/tlenowego PUR.

Zdolności predykcyjne oryginalnego i zmodyfikowanego modelu ASM2d potwierdzone zostały obliczeniami średnich błędów względnych (z ang. ARD). Zmodyfikowany model ASM2d lepiej prognozował zmiany stężeń ChZT i poboru tlenu podczas testów laboratoryjnych OUR. Dla porównania dla obu oczyszczalni, wartości błędów symulacji testów OUR ze ściekami oczyszczonymi mechanicznie oraz po koagulacji i flokulacji wyniosły kolejno dla oryginalnego ASM2d (18,9-45,8% i 11,3-29,5%) i jego modyfikacji (11,8-30,3% i 9,7-15,8%). Natomiast procesy PRR, AUR i poboru ChZT wykazywały niewielkie różnice pomiędzy mierzonymi i prognozowanymi przez oba modele wynikami badań w obu równoległych próbkach ścieków. W przypadku symulacji procesu jednostkowego NUR, podczas obu testów (konwencjonalnego NUR oraz PRR i anoksycznego PUR) ze ściekami oczyszczonymi mechanicznie oraz po koagulacji i flokulacji różnica między średnimi wartościami błędu względnego ARD była nawet ponad 6% wyższa dla zmodyfikowanego modelu ASM2d.

Wnioski

Uzyskane wyniki badań pozwoliły na sformułowanie następujących wniosków:

1. Opracowano nową metodykę oceny wpływu koloidalnych i zawiesinowych związków organicznych na proces denitryfikacji oraz podwyższonej biologicznej defosfatacji (EBPR) w systemach osadu czynnego. Metoda ta jest oparta na pomiarze szybkości procesów biochemicznych w dwóch równoległych reaktorach nieprzepływowych z użyciem ścieków oczyszczonych mechanicznie (bez dalszego podczyszczenia oraz poddanych koagulacji i flokulacji).
2. Koloidalne i zawiesinowe frakcje organiczne odgrywają istotną rolę w przebiegu w/w procesów biochemicznych (denitryfikacji oraz EBPR). Za wyjątkiem procesu uwalniania PO_4–P (PRR), usunięcie tych związków ze ścieków spowodowało obniżenie szybkości pozostałych badanych procesów. Średnie redukcje mierzonych szybkości procesów były następujące:

- 24% i 35% odpowiednio dla redukcji NO_3–N w I i II fazie (NUR1 i NUR2) podczas konwencjonalnego pomiaru szybkości denitryfikacji (test NUR) oraz 30% dla redukcji NO_3–N w warunkach anoksycznych dwufazowego testu PUR;

- 32% i 25% odpowiednio dla poboru PO_4–P w warunkach anoksycznych i tlenowych (dwufazowy test PUR) ;

- 13% dla poboru tlenu (OUR) podczas drugiej fazy (tlenowej) testu PUR oraz 24% dla poboru tlenu podczas konwencjonalnego testu OUR.

3. Wyniki części doświadczalnej tych badań pozwoliły na uzyskanie kompleksowej bazy danych do kalibracji i oceny zdolności predykcyjnych modeli dynamicznych procesu hydrolizy. Wykorzystując koncepcję tzw. „dwustopniowej" hydrolizy opracowano nowy model matematyczny jako rozszerzenie ASM2d. W tym modelu zastosowano nową zmienną (substrat szybko ulegający hydrolizie, X_{SH}) i trzy dodatkowe procesy - hydroliza z X_{SH} w warunkach tlenowych, anoksycznych i beztlenowych. Wyniki uzyskane podczas optymalizacji parametrów na podstawie konwencjonalnych testów OUR wykazały, że stałe szybkości hydrolizy (k_{hyd} i $k_{hyd,r}$) w modelu dwustopniowym hydrolizy wynosiły odpowiednio 2,0 i 10 d^{-1}. Natomiast w przypadku stałej szybkości hydrolizy w modelu jednostopniowym wartość ta wynosiła 2,5 d^{-1} w ASM2d.
4. W porównaniu z oryginalnym ASM2d, nowy model lepiej prognozował szybkość poboru tlenu podczas konwencjonalnych testów OUR w obu badanych oczyszczalniach. Średnia wartości błędu względnego (ARD) wynosiła odpowiednio 17,2% i 27,6% (oryginalny ASM2d) oraz 13,8% i 20,2% (zmodyfikowany ASM2d) dla testów ze ściekami oczyszczonymi mechanicznie bez dalszego podczyszczenia oraz po koagulacji i flokulacji. Natomiast średnia wartości błędu względnego, mierzonych podczas testów OUR, stężeń związków organicznych wyrażonych w ChZT mieściła się w wąskim przedziale (4,4-5,0%) w obu badanych oczyszczalniach.
5. Zdolności prognozowania obu modeli były także porównane na podstawie mierzonych procesów nitryfikacji, denitryfikacji i biologicznego usuwania fosforu w trakcie testów z użyciem reaktorów nieprzepływowych. Nowy model wykazywał zmienny wpływ na poprawę prognozowania stężeń NH_4-N, NO_3-N i PO_4-P. Najmniejszą różnicę między średnimi wartościami błędu względnego w przypadku obu modeli, stwierdzono dla AUR i PRR odpowiednio (0,3% i 0,6%), zaś największą dla PUR i NUR (5,9% i 6,6%).

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Acknowledgements

I would like to express my appreciation to the following people, each of whom contributed to this Ph. D. thesis. There are not enough words that can be expressed to thank:

- My supervisor **dr hab. inż. Jacek Mąkinia, prof. nadzw. PG**, for his guidance, patience, assistance and encouragement during this research.
- The reviewers, **prof. zw. dr hab. inż. Piotr Kowalik** from the Faculty of Civil and Environmental Engineering, Gdańsk University of Technology and **prof. dr hab. inż. Marek Sozański**, from the Institute of Environmental Engineering, Poznan University of Technology for providing valuable remarks to this Ph.D. thesis.
- The head of the Department of Water and Wastewater Technologies, **prof. dr hab. inż. Hanna Obarska-Pempkowiak** for the opportunity to conduct research in the laboratory and the use of premises belonging to the Department.
- My colleagues, **dr inż. Krzysztof Czerwionka** from the Department of Water and Wastewater Technology, Gdańsk University of Technology, **dr Marek Swinarski**, **dr inż. Marta Chrzanowska, mgr Wojciech Fordoński** from the Department of Technology, Saur Neptun Gdańsk SA, and **dr inż. Anna Remiszewska-Skwarek** from the "Dębogórze" WWTP, PEWIK Gdynia Sp. z o.o. for their help and active engagement in accomplishing the project.
- The **management board and staff** at the "Wschód" and "Dębogórze" WWTPs for their help in providing a possibilites of carry out this project and very friendly cooperation.
- The **administration staff** of the Faculty of Civil and Environmental Engineering, Gdańsk University of Technology for their patience in carry out this project and help in performing a great number of documents.
- **My wife Małgosia** and **my Parents** for their love, patience and understanding for my absence in family life during writing this thesis.
- My **little daughter Maja** for the happiness she is giving me each day.
- And anyone whose name may have been omitted by mistake, please accept my apologies.

This research has been financially supported by **Polish Ministry of Science and Higher Education under the grant no. N-523-422-437** and the European Social Fund, the State Budget and the Pomorskie Voivodeship Budget according to the Operational Programme Human Capital, Priority VIII, action 8.2, under-action 8.2.2: 'Regional Innovation Strategy' within the system project of the Pomorskie Voivodeship **"Innodoktorant – Scholarships for PhD students, II edition".**

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List of important symbols and abbreviations

A/O – Anaerobic/Oxic (a type of the BNR process)
A^2/O – Anaerobic/Anoxic/Oxic (a type of the BNR process)
ARD – Average relative deviation, [%]
ASM – Activated Sludge Model
ATU – Allythiourea (nitrification inhibitor)
b_H – Specific lysis (decay) rate constant for heterotrophic organisms, [T^{-1}]
BNR – Biological nutrient removal
BOD – Biochemical oxygen demand, [M(BOD) L^{-3}]
$BOD_{u,in}$ – Ultimate BOD in influent, [M(BOD) L^{-3}]
$BOD_{5,in}$ – Influent BOD_5, [M(BOD) L^{-3}]
$CBOD_{5,sol,out}$ – Soluble carbonaceous BOD_5 in secondary effluent, [M(BOD) L^{-3}]
$CBOD_{sol,out}$ – Soluble carbonaceous BOD in secondary effluent, [M(BOD) L^{-3}]
COD – Chemical oxygen demand, [M(COD) L^{-3}]
$COD_{f,in}$ – COD in the filtered sample of influent, [M(COD) L^{-3}]
$COD_{f,out}$ – COD in the filtered sample of secondary influent, [M(COD) L^{-3}]
COD_{in} – Influent COD, [M(COD) L^{-3}]
COD_{out} – COD in secondary effluent, [M(COD) L^{-3}]
$COD_{sol,out}$ – Soluble COD in secondary effluent, [M(COD) L^{-3}]
$COD_{VFA,in}$ – Concentration of VFAs in (primary) influent, [M(COD) L^{-3}]
$COD_{x,in}$ – Difference between total COD and COD in the filtered sample of influent, [M(COD) L^{-3}]
DO – Dissolved oxygen, [M L^{-3}]
DWW – Domestic wastewater
EBPR – Enhanced biological phosphorus removal
EU – European Union
F/M – Substrate (food) to biomass ratio, [M(COD) M^{-1}]
f_p – Fraction of inert COD generated in biomass lysis (decay), [-]
$f_{(I)}$ – the function of inhibitor concentration, [-]
f_{SI} – Production of S_I in hydrolysis, [M(COD) $M(COD)^{-1}$]
$f_{XI,A}$ – Fraction of inert COD generated in lysis of autotrophic organisms, [M(COD) $M(COD)^{-1}$]
$f_{XI,H}$ – Fraction of inert COD generated in lysis of autotrophic organisms, [M(COD) $M(COD)^{-1}$]
$f_{XI,PAO}$ – Fraction of inert COD generated in lysis of PAOs, [M(COD) $M(COD)^{-1}$]
GAO – Glycogen accumulating organisms
HRT – Hydraulic retention time
I – Inhibitor concentration, [M L^{-1}]
i_{cv} – COD/VSS ratio, [M(COD) M^{-1}]
$i_{N,BM}$ – N content of biomass (X_H, X_{PAO}, X_{AUT}), mg N/mg COD
$i_{N,SI}$ – N content of S_I, [M(N) $M(COD)^{-1}$]
$i_{N,SF}$ – N content of S_F, [M(N) $M(COD)^{-1}$]
$i_{N,XI}$ – N content of X_I, [M(N) $M(COD)^{-1}$]
$i_{N,XS}$ – N content of X_S, [M(N) $M(COD)^{-1}$]
$i_{P,BM}$ – P content of X_H, X_{PAO}, X_{AUT}, [M(P) $M(COD)^{-1}$]
$i_{P,SF}$ – P content of S_F, [M(P) $M(COD)^{-1}$]
$i_{P,SI}$ – P content of S_I, [M(P) $M(COD)^{-1}$]
$i_{P,XI}$ – P content of X_I, [M(P) $M(COD)^{-1}$]
$i_{P,XS}$ – P content of X_S, [M(P) $M(COD)^{-1}$]

$i_{TSS,XI}$ – TSS/COD ratio for X_I, [M M(COD)$^{-1}$]
$i_{TSS,XS}$ – TSS/COD ratio for X_S, [M M(COD)$^{-1}$]
$i_{TSS,BM}$ – TSS/COD ratio for biomass (X_H, X_{PAO}, X_A), [M M(COD)$^{-1}$]
i_{VT} – MLVSS/MLSS ratio, [MM^{-1}]
IWA – International Water Association
IAWQ – International Association on Water Quality
IWW – Industrial wastewater
JHB – Johannesburg (a type of the BNR process)
$K_{A,H}$ – Fermentation products saturation coefficient for heterotrophic organisms, [M(COD) L^{-3}]
$K_{A,PAO}$ – Fermentation products saturation coefficient for PAOs, [M(COD) L^{-3}]
k_{BOD} – BOD decay coefficient, [T^{-1}]
k_{end} – Specific endogenous respiration rate constant, [T^{-1}]
K_F – Fermentable, readily biodegradable organic substrate, saturation coefficient, [M(O_2) L^{-3}]
K_{fe} – Saturation coefficient for fermentation of S_F, [M(O_2) L^{-3}]
k_{hyd} – Specific hydrolysis rate constant, [T^{-1}]
K_I - Inhibition constant, [M L^{-1}]
K_{IPP} – Inhibition coefficient for X_{PP} storage by PAOs, [M(P) M(COD)$^{-1}$]
K_{MAX} – Maximum ratio of X_{PP}/X_{PAO}, [M(P) M(COD)$^{-1}$]
$K_{NH4,A}$ – Ammonia saturation coefficient for autotrophic organisms, [M(N) L^{-3}]
$K_{NH4,H}$ – Ammonia saturation coefficient for heterotrophic organisms, [M(N) L^{-3}]
$K_{NH4,PAO}$ – Ammonia saturation coefficient for PAOs, [M(N) L^{-3}]
$K_{NO3,hyd}$ – Nitrate saturation/inhibition coefficient for hydrolysis of slowly biodegradable substrate, [M(N) L^{-3}]
$K_{NO3,H}$ – Nitrate saturation/inhibition coefficient for heterotrophic organisms, [M(N) L $^{-3}$]
$K_{NO3,PAO}$ – Nitrate saturation/inhibition coefficient for PAOs, [M(N) L^{-3}]
$K_{O,hyd}$ – Oxygen saturation coefficient for hydrolysis of slowly biodegradable substrate, [M(O_2) L $^{-3}$]
$K_{O2,A}$ – Oxygen saturation/inhibition coefficient for autotrophic organisms, [M(O_2) L $^{-3}$]
$K_{O2,H}$ – Oxygen saturation/inhibition coefficient for heterotrophic organisms, [M(O_2) L $^{-3}$]
$K_{O2,PAO}$ – Oxygen saturation/inhibition coefficient for PAOs, [M(O_2) L^{-3}]
K_{PHA} – PHA saturation coefficient for PAOs, [M(COD) M(COD)$^{-1}$]
$K_{PO4,A}$ – Phosphate saturation coefficient for autotrophic organisms, [M (P) L^{-3}]
$K_{PO4,H}$ – Phosphate saturation coefficient for heterotrophic organisms, [M (P) L^{-3}]
$K_{PO4,PAO}$ – Phosphate saturation coefficient for PAOs, [M (P) L^{-3}]
K_{PP} – Poly-phosphate saturation coefficient, [M(P) M(COD) $^{-1}$]
K_{PS} – Phosphate saturation coefficient in storage of X_{PP}, [M (P) L^{-3}]
K_S – Substrate saturation coefficient for microorganisms, [M (COD) L^{-3}]
K_X – Saturation coefficient for hydrolysis of particulate COD, [M(COD) M(COD)$^{-1}$]
$L_{N,dn}$ – Total load of denitrified N, [M(N)T^{-1}]
$L_{N,n}$ – Total load of nitrified N, [M(N)T^{-1}]
MLR – Mixed liquor recycle
MLSS – Mixed liquor suspended solids, [M L^{-3}]
MLVSS – Mixed liquor volatile suspended solids, [M L^{-3}]
MUCT – Modified University of Cape Town (a type of the BNR process)
$N_{tot.,in}$ –Influent total N concentration, [M(N) L^{-3}]

$N_{tot.,out}$ –Effluent total N concentration, [M(N) L^{-3}]
$N_{tot.,was}$ –Total N concentration in WAS, [M(N) L^{-3}]
NUR – Nitrate utilization rate, mg [M(N) L^{-3} T^{-1}]
ORP – Oxidation reduction potential, [mV]
OU_{tot} – Total oxygen uptake in bioreactor, [M(O_2) T^{-1}]
OUR – Oxygen uptake rate, [M(O_2) L^{-3} T^{-1}]
OUR_H – Heterotrophic oxygen uptake rate, [M(O_2) L^{-3} T^{-1}]
$OUR_{H,end}$ – Endogenous heterotrophic oxygen uptake rate, [M(O_2) L^{-3} T^{-1}]
OUR_w – Oxygen uptake rate obtained by performing a respirometric measurement
P_{in} – Total P concentration in (primary) influent, [M(P) L^{-3}]
P_{out} – Total P concentration in secondary effluent, [M(P) L^{-3}]
P_{was} – Total P concentration in WAS, [M(P) L^{-3}]
PAO – Phosphate accumulating organism
PHA – Poly-hydroxy-alkanoates
PRR – Phosphate release rate, [M(P) L^{-3} T^{-1}]
PUR – Phosphate utilization rate, [M(P) L^{-3} T^{-1}]
Q_{in} – Influent flow rate, [L^3T^{-1}]
Q_{out} – Effluent flow rate from secondary clarifier, [L^3T^{-1}]
Q_{was} – Waste activated sludge (WAS) flow rate, [L^3T^{-1}]
q_{fe} – Maximum specific rate for fermentation, [T^{-1}]
q_{PHA} – Specific rate constant for storage of X_{PHA} by PAOs, [T^{-1}]
q_{PP} – Specific rate constant for storage of X_{PP} by PAOs, [T^{-1}]
RBCOD – Readily biodegradable COD, [M(COD) L^{-3}]
SBR – Sequencing Batch Reactor
SBCOD – Slowly biodegradable COD, [M(COD) L^{-3}]
SCOD – Soluble COD, [M(COD) L^{-3}]
SD – Standard deviation
S – Substrate concentration, [M L^{-3}]
S_A – Concentration of soluble, readily biodegradable fermentation products, [M(COD) L^{-3}]
S_{ALK} – Concentration of alkalinity of the wastewater, [Mole (HCO_3^-) L^{-3}]
S_F – Concentration of soluble, readily biodegradable fermentable organic substrate, [M(COD) L^{-3}]
S_H – Concentration of soluble, rapidly hydrolysable organic compounds, [M(COD) L^{-3}]
S_I – Concentration of soluble inert organic material, [M(COD) L^{-3}]
$S_{I,in}$ – Influent concentration of soluble inert organic compounds, [M(COD) L^{-3}]
$S_{I,out}$ – Effluent concentration of soluble inert organic compounds, [M(COD) L^{-3}]
$S_{I,prod}$ – Concentration of soluble inert organic produced in the activated sludge system, [M(COD) L^{-3}]
S_{N2} – Concentration of nitrogen gas, [M(N) L^{-3}]
S_{NH} – Concentration of ammonia (and ammonium), [M(N) L^{-3}]
S_{NH4} – Concentration of ammonium plus ammonia nitrogen, [M(N) L^{-3}]
S_{NO} – Concentration of oxidized nitrogen, [M(N) L^{-3}]
S_{NO3} – Concentration of nitrate, [M(N) L^{-3}]
S_{O2} – Concentration of dissolved oxygen, [M(O_2) L^{-3}]
S_{PO4} – Concentration of orthophosphate, [M(P) L^{-3}]
SRT – Solids retention time, [T]
S_O – Concentration of dissolved oxygen, [M(O_2) L^{-3}]
S_S – Concentration of soluble, readily biodegradable organic substrate, [M(COD) L^{-3}]

T – Temperature, °C
TKN – Total Kjeldahl nitrogen, [M(N) L^{-3}]
TKN_{in} – Total Kjeldahl N concentration in (primary) influent, [M(N) L^{-3}]
TKN_{out} – Total Kjeldahl N concentration in secondary effluent, [M(N) L^{-3}]
TKN_{was} – Total Kjeldahl N concentration in WAS, [M(N) L^{-3}]
TN – Total nitrogen, [M(N) L^{-3}]
TP – Total phosphorus, [M(P) L^{-3}]
TSS – Total suspended solids, [M L^{-3}]
TUD – Technical University of Delft
UCT – University of Cape Town (a type of the BNR process)
UWW – Urban wastewater
WERF – Water Environment Research Foundation
WWTP – Wastewater treatment plant
VFA – Volatile fatty acids, [M L^{-3}]
VOC – Volatile organic compound
VSS – Volatile suspended solids, [M L^{-3}]
X_A – Concentration of autotrophic organisms, [M(COD) L^{-3}]
X_{asr} – Solids (MLSS) concentration in the bioreactor, [M L^{-3}]
X_{AUT} – Concentration of autotrophic organisms, [M(COD) L^{-3}]
X_H – Concentration of heterotrophic organisms, [M(COD) L^{-3}]
X_I – Concentration of inert particulate organic material, [M(COD) L^{-3}]
X_{MeOH} – Concentation of metal-hydroxide, [M L^{-3}]
X_{MePO4} – Concentation of metal-phosphate, [M($Fe(OH)_3$) L^{-3}]
X_{out} – Solids concentration in secondary effluent, [M L^{-3}]
X_{PAO} – Phosphate Accumulating Organisms, [M($FePO_4$) L^{-3}]
X_{PHA} – Concentation of a cell internal storage product of PAOs, [M(COD) L^{-3}]
X_{PP} – Concentation of poly-phosphate, [M(P) L^{-3}]
X_S – Concentation of slowly biodegradable substrates, [M(COD) L^{-3}]
X_{ras} – Solids concentration in RAS, [M L^{-3}]
X_v – MLVSS concentration of mixed liquor, [M L^{-3}]
X_{STO} – Concentration of cell internal storage product of heterotrophic organisms, [M(COD) L^{-3}]
X_{TSS} – Concentation total suspended solids, [M(TSS) L^{-3}]
Y – "*True*" growth yield coefficient for microorganisms, [MM^{-1}]
Y' – Apparent yield coefficient for degrading the matter remaining in the filtrate, [M(COD) $M(COD)^{-1}$]
Y_{BOD} – Yield coefficient for BOD, [-]
Y_H – Growth yield coefficient for heterotrophic organisms, [M(COD) $M(COD)^{-1}$]
Y_{PAO} – Growth yield coefficient for PAOs (biomass/X_{PHA}), [M(COD) $M(COD)^{-1}$]
Y_{PHA} – X_{PHA} requirement for X_{PP} storage, [M(P) $M(COD)^{-1}$]
Y_{PO4} – X_{PP} requirement for PHA storage, [M(P) $M(COD)^{-1}$]
η_{fe} – Anaerobic hydrolysis reduction factor, [-]
$\eta_{NO3,hyd}$ – Anoxic hydrolysis reduction factor, [-]
$\eta_{NO3,H}$ – Anoxic reduction factor for growth of heterotrophic organisms, [-]
$\eta_{NO3,PAO}$ – Anoxic reduction factor for growth of PAOs, [-]
μ_H – Maximum specific growth rate of heterotrophic organisms, [T^{-1}]
μ_{AUT} – Maximum specific growth rate of autotrophic organisms, [T^{-1}]
μ_{PAO} – Maximum specific growth rate of PAOs, [T^{-1}]

Abstract

The efficiency of denitrification and enhanced biological phosphorus removal (EBPR) in biological nutrient removal (BNR) activated sludge systems strongly depends on the availability of appropriate carbon (C) sources. The C sources can be divided into three groups: external (not present in the wastewater influent), endogenous (produced in the system as a result of decay) and internal (present in the influent wastewater). Due to high costs of commercial compounds (such as methanol, ethanol, acetic acid, etc.) and acclimation periods (usually) required, the effective use of internal substrates is preferred. In the wastewater characteristics, the internal C sources are described separately (as readily and slowly biodegradable) because of different rates of degradation. The role of readily biodegradable COD fraction (S_S) in biological wastewater treatment systems has extensively been investigated and reported, but it is still very little knowledge about the influence of slowly biodegradable substrate (X_S) on denitrification and EBPR. The aim of this study was to determine the effects of particulate and colloidal X_S, on major biochemical processes occuring in activated sludge systems such as denitrification, phosphate release/uptake and oxygen utilization. The essential study was divided into two parts: experimental investigation and mathematical modeling. In the first part of research a novel measurement procedure for a indirect determination of the effect of X_S was developed and implemented. The results of laboratory tests were used futher to verify the mechanism of the hydrolysis process using an existing Activated Sludge Model No 2d (ASM2d). Based on the concept "dual hydrolysis model" presented by Orhon et al. (1998) a new mathematical model for CNP activated sludge systems was developed as the modified version of ASM2d.

The study was conducted at two large biological nutrient removal (BNR) wastewater treatment plants (WWTPs) in northern Poland: "Wschód" in Gdańsk and "Dębogórze" in Gdynia. For the laboratory experiments, the process mixed liquor and settled wastewater (the average daily time-proportional samples) were collected from the "Wschód" WWTP during three study periods, termed winter, summer and spring, between December, 2007 and May, 2009. Similar samples from the "Dębogórze" WWTP were collected during two study periods (fall and spring) between September, 2009 and May, 2010. According to the developed novel procedure, the laboratory experiments were conducted in parallel batch reactors. For this purpose, four types of one- and two-phase laboratory batch tests (such as the conventional nitrate utilization rate (NUR), phosphate release (PRR) and anoxic/aerobic phosphate uptake (PUR), oxygen uptake rate (OUR) measurements) were carried out with the settled wastewater without pretreatment (reactor 1) and pretreated with coagulation-flocculation method (reactor 2). The latter sample, containing only a soluble organic fraction, was prepared according to the rapid physical-chemical method of Mamais et al. (1993).

The second part of this study comprised mathematical modeling and computer simulation of the activated sludge process with a special attention to hydrolysis process. Based on the existing mathematical model (ASM2d) a 96-hour measurement campaign in the full-scale Modified University of Cape Town (MUCT) bioreactor at "Wschód" WWTP was calibrated

under dynamic conditions. The predicted performance of the calibrated ASM2d was evaluated using steady state and dynamic simulations with input data from the 96-hour measurement campaign at "Wschód" WWTP. The same sets of model parameters were also used in "Dębogórze" WWTP in order to estimate input parameters of the ASM2d biomass components necessary to comply batch tests simulations. Finally, the calibrated ASM2d with adjusted stoichiometric and kinetic coefficients were used to validate the results of the batch tests with the settled wastewater without pretreatment and after coagulation-flocculation from both plants. Next the ASM2d was extended to describe hydrolysis as a two-step process. The modified model incorporated one new component, rapidly hydrolyzable substrate (X_{SH}) and three new processes, i.e., hydrolysis of X_{SH} under aerobic, anoxic and anaerobic conditions. The X_{SH} concentration in the settled wastewater was estimated based on a physical fractionation of actual data from "Wschód" and "Dębogórze" WWTP. It was assumed that the difference between membrane filtration (under vacuum pressure) of settled wastewater through 1.2 and 0.1 µm GF/C filter, constitutes an average value of X_{SH}. Due to the fact that the experiments were carried out with parallel samples of wastewater, the average value of X_{SH} was subdivided into the colloidal and soluble $X_{SH,(c/s)}$ for settled wastewater without pretreatment and only the soluble $X_{SH,(s)}$ for settled wastewater after coagulation-flocculation. The results of OUR batch tests with the settled wastewater without pretreatment, together with the new fractionation were used to perform the optimization method of Nelder-Mead simplex. For the optimization purpose one stoichiometric and four kinetic coefficients (Y_h, k_{hyd}, $k_{hyd,r}$, K_X, K_{Xr}) were selected, but only 3 of them were simultaneously used each time to carry out the optimization in the 10 scenarios. After the optimization of all possible scenarios for both plants, the best one, which generated the smallest average relative deviation (ARD) between measured and simulated results, was chosen. In the case of parallel samples with the settled wastewater after coagulation-flocculation, it was necessary to search value of the soluble $X_{SH,(s)}$ fraction in order to obtain the best fit between measured and calculated values of output parameters, based on the same kinetic and stoichiometric coefficients as set in the settled wastewater without pretreatment. Finally, average values of X_{SH} and optimized kinetic parameters were implemented to the new model for both parallel samples of wastewater in order to compare (NUR, PRR and anoxic/aerobic PUR, OUR) batch tests simulations to similar experiments in the original ASM2d.

In the experimental investigation, the removal of colloidal and particulate fractions resulted in explicitly reduced process rates (except for PRR). The average reductions ranged from 13% for the OUR during the second phase of the two-phase experiment (anaerobic/aerobic), 24/32% for the aerobic/anoxic PURs and up to 35% for the NUR during the second phase of a conventional NUR measurement. After the experimental part, mathematical modeling along with computer simulations were performed. First, based on the existing mathematical model (ASM2d), a 96-hour measurement campaign in the full-scale Modified University of Cape Town (MUCT) bioreactor at "Wschód" WWTP was calibrated under dynamic conditions. The calibration of denitrification process rates were carried out based on the results of two

batch tests (conventional NUR and PRR/anoxic PUR) with the settled wastewater without pretreatment from "Wschód" WWTP during summer study period. In the conventional NUR tests, model predictions were fitted to the measured NURs by adjusting two parameters: the maximum growth rate of heterotrophs (μ_H) and hydrolysis rate constant (k_{hyd}). In the PRR/anoxic PUR tests, the anoxic reduction factor for PAO growth ($\eta_{NO3,PAO}$) was adjusted to calibrate the NUR in the anoxic phase. The corresponding comparisons of adjusting above parameters were made for the parallel batch tests with the settled wastewater after coagulation-flocculation. In that case, no further modifications were needed to calibrate the results of NUR and PRR/anoxic PUR. The nitrification process based on the measured data from PRR and aerobic PUR batch tests at both plants was calibrated with kinetic parameters, including the maximum growth rate of autotrophs (μ_A), NH_4-N saturation coefficient ($K_{NH4,A}$). The PRR and PUR tests were calibrated with six parameters: the rate constant for storage of PHA (q_{PHA}), saturation coefficient for PAOs with respect to S_A ($K_{SA,PAO}$), saturation coefficient for PAOs with respect to polyphosphate (K_{PP}), anaerobic hydrolysis reduction factor (η_{fe}) and saturation coefficient for particulate COD (K_X). Next phase of modeling research was implementation of the modified ASM2d. The estimation of a new X_{SH} component for the settled wastewater without pretreatment resulted in the average value 12% of X_S in modified ASM2d. The kinetic parameters of the best optimization of OUR tests from the 10 scenarios, generating the smallest average relative deviation (ARD) between measured and simulated results, were chosen. The average value of these coefficients (k_{hyd} = 2, $k_{hyd,r}$ = 10, K_X = 0.1) for both plants were implemented in the new model. The other the default or calibrated, kinetic and stoichiometric coefficients (beside Y_H which was established based on the results of OUR tests) were adopted from the original ASM2d. In the case of parallel samples with the settled wastewater after coagulation-flocculation, estimated average value of $X_{SH,(s)}$ was 6% of X_S for all the batch tests based on the same kinetic and stoichiometric coefficients as set in the settled wastewater without pretreatment. The predictive capabilities of the original and modified ASM2d have been confirmed by ARD. In comparison with ASM2d, the new model has better predicted the dissolved oxygen behaviour in conventional OUR batch tests at both studied plants. The average ARDs were 17.2% and 27.6% (original ASM2d) vs. 13.8% and 20.2% (modified ASM2d), respectively, for the settled wastewater without pretreatment and after coagulation-flocculation. In contrast, the ARDs for COD concentrations measured during the OUR tests were very similar (4.4-5.0%) for both models. Predictions of both models have also been examined based on the nitrification-denitrification and EBPR measurments in the batch tests. The modified model has showed variable effects on the improvements in predicted behaviour of NH_4-N, NO_3-N and PO_4-P. The smallest difference between the average ARDs for both models (0.3 and 0.6%) was found for AUR and PRR, whereas the largest (5.9 and 6.6%) was found for PUR and NUR, respectively. The observed effects of colloidal and particulate organic compounds are important in terms of optimization the sedimentation and chemical precipitation in primary clarifiers, i.e. balancing between the efficiency of N, P removal in the bioreactor and biogas production in the digester.

1

General Introduction

1.1. Background

Developed countries have started to take awareness of the environmental protection since 1970s, especially in the field of wastewater treatment. The international governments obligated by public awareness about importance of nutrients (N, P) removal, started to be more strict in their regulations about the wastewater discharges to the natural environment. Poland after accession to the European Union (EU) significantly reduced the permissible concentration of nutrients in effluent wastewater. The new, more stringent regulations, concerning wastewater treatment and disposal, were set in 2004 (and revised in 2006) in accordance to the recommendations of the EU Urban Wastewater Directive 91/271/EEC. In 2003, the Ministry of the Environment estimated, that only 23 such facilities of the total number of 129 were capable of meeting the limits in accordance with the EU Directive. According to Mąkinia et al. (2004), the most challenging issue for the existing wastewater treatment plants (WWTPs) in large agglomerations (>100,000 PE) would be achievement of the effluent limits for total N (Table 1.1).

Table 1.1. Polish regulations for large WWTPs vs. EU directive

Parameter	Unit	EU directive of 1991	Polish regulation of 1991	Polish regulation of 2006
TN	g N/m^3	10	30	10
NH_4-N	g N/m^3	-	6	-
NO_3-N	g N/m^3	-	30	-
TP	g P/m^3	1	1.5	1

The research on potential utilization of different carbon (C) sources for denitrification and enhanced biological phosphorus removal (EBPR) has been carried out for over 20 years. However, the topic of alternative C sources for denitrification and EBPR has recently received a special attention with the EU policy outlined in the currently running program "Cooperation Theme 6 – Environment (including climate change)". The major strategic priorities of this program is to strongly support research activities aiming at developing new environmental technologies. In this circumstances effective use of internal carbon sources, such as slowly biodegradable substrate (Xs) for denitrification and EBPR may help in realization the sustainable solution of environmental problems by achieve by WWTPs new limits in accordance with the EU Directive (1991). The next step in this issue is evaluating the kinetics of biochemical processes enhance by alternative carbon sources in full-scale activated sludge systems. This study should focus on validating, to what extent the activated sludge process rates, based on the batch tests, are comparable with the rates obtained from the simulation study. Consequently, such information can be used to optimize

the WWTP, where the tests were conducted, and to provide guidelines for designing an activated sludge reactor to upgrade existing plants. Due to the similar characteristics of municipal wastewater, it is expected that the performance of computer simulation and optimization, helped to avoid the WWTP unnecessary costly construction work during the modernization counted in millions Euro.

1.2. Motivation and problem formulation

The efficiencies of denitrification and EBPR in biological nutrient removal (BNR) activated sludge systems are strongly dependent on the availability of appropriate C sources. Denitrification is accomplished by a variety of facultative heterotrophic microorganisms, which can utilize nitrate (or nitrite) instead of oxygen as the final electron acceptor. Sufficient amounts of organic carbon (electron donor) must also be ensured to provide energy for the conversion. The energy sources can be categorized as internal (present in the influent wastewater), endogenous (self-generated within the system as a result of organism decay) and external (not present in wastewater). Many of the operating systems to enhance the denitrification process and improve the overall efficiency of N removal within the existing capacities of activated sludge systems use of latter (external) sources. Henze et al. (1994) suggested that the dominating rate limiting factor in nutrient removal processes is the organic carbon source. Their addition in pre-denitrification anoxic zones increases the denitrification rates and nitrogen removal efficiencies, while the addition of C to the anaerobic zone of EBPR systems may help to improve the process performance and stability (Figure 1.1). There is a number of effective, commercially available organic compounds (such as methanol, ethanol, acetic acid, sodium acetate and glucose) which can be categorized as the conventional C sources. Among them, methanol has been most commonly used and best documented. However, due to high costs of commercial compounds and acclimation periods (usually) required, the effective use of "alternative" C sources for denitrification is preferred. In particular, industry effluents (especially food industry such as breweries and distilleries) appear to be good candidates due to their high C/N ratios and high content of readily biodegradable organic fraction (Cappai et al., 2004; Quan et al., 2005; Sage et al., 2006). Therefore, the usefulness of alternative sources rich in organic C should be reviewed on the basis of research. Moreover, the industry effluents may sometimes reveal operating problems during production cycles, including the variation in quality and quantity, which means that it would be difficult to provide effluents for all potencial WWTPs. Finally, the possibility of use this kind of C sources is limited only to the nearest area from plant, because of the high cost of transportation.

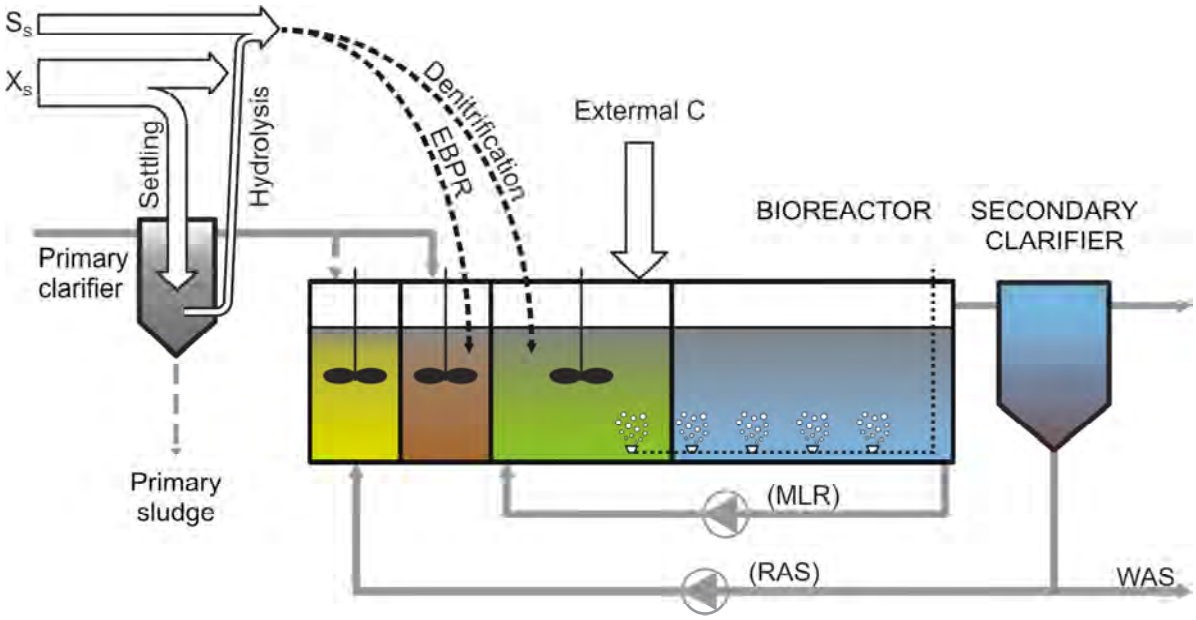


Figure 1.1. The substrates flow in BNR activated sludge systems (Mąkinia et al., 2009).

The EBPR process is accomplished by heterotrophic microorganisms collectively referred as phosphate accumulating organisms (PAOs). The principal mechanism for attaining the EBPR process is a continual circulation of the activated sludge biomass through an alternating anaerobic/aerobic (or anoxic) phases or zones, and provision of specific carbon sources during the anaerobic phase. These sources primarily include acetate and propionate which are most common volatile fatty acids (VFAs) in domestic wastewater (Pijuan et al., 2004). The contribution of other carbon sources than VFAs for proliferation of PAOs in the anaerobic phase is currently not clear, although the EBPR process also occurred successfully with other organic substrates, such a mixture of peptone and glucose, or even only glucose (Carucci et al., 1999) as well as carboxylic acids and amino acids (Mino et al., 1998). In "normal" municipal wastewater, the VFAs concentrations are usually insignificant. Therefore, in practice, fermentation of "complex" readily biodegradable substrates, accomplished by "ordinary" heterotrophs, plays an important role in EBPR allowing to sustain PAOs in the system.

In order to improve complex wastewater characteristics, different organic fractions have to be described separately. Ekama and Marais (1979) were the first, who divided the wastewater into distinct biodegradable fractions, that are degraded at two different rates. The readily biodegradable (S_S) fraction consists mainly of soluble organic compounds and the slowly biodegradable (X_S) fraction consists of large molecules, colloids and particles. According to the literature data (e.g. Orhon et al., 1997; Lagarde et al., 2005), municipal wastewater after primary treatment contains readily and slowly biodegradable compounds in the amounts of approximately 10-

30% and 40-60% of total COD, respectively. An investigation in four municipal WWTPs in northern Poland revealed that particulate and colloidal fractions constituted more than 60% of total COD (Czerwionka et al., 2008). These two physical fractions mostly contain X_S, which can be taken up and degraded by microorganisms after hydrolysis to the S_S (Levine et al., 1985).

The role of S_S in biological wastewater treatment systems has extensively been investigated and reported, but it is still very little known about the influence of X_S on denitrification and EBPR. The observed effects of colloidal and particulate organic compounds are important in terms of optimization the sedimentation and chemical precipitation in primary clarifiers, i.e. balancing between the efficiency of nutrient removal in the bioreactor and biogas production in the digester. Confirmation of suitability X_S could mean the possibility to obtain, as required by EU Directive (1991), the concentration of N, P in the effluent, without incurring significant expenditure to upgrade the plants.

1.3. Research objectives

The aim of this study was to determine the effects of particulate and colloidal X_S, on major biochemical processes occuring in activated sludge systems such as denitrification, phosphate release/uptake and oxygen utilization. The essential study was divided into two major parts: experimental investigation and mathematical modeling. In the first part of research a novel measurement procedure for a indirect determination of the effect of X_S was developed and implemented, based on the work of Goel et al. (1999). The results of laboratory tests were futher used to evaluate a mechanism of the hydrolysis process using an existing Activated Sludge Model No 2d (ASM2d). The biodegradation of X_S is initiated by hydrolysis, which is an integral part of complex activated sludge models, such as ASM2d (Henze et al., 1999). This process is slower than heterotrophic growth and thus becomes the rate-limiting step for the biodegradation of organic compounds (Insel et al., 2003). Based on the concept "dual hydrolysis model" presented by Orhon et al. (1998), a new mathematical model was developed as the modified version of ASM2d.

The study was conducted at two large biological nutrient removal (BNR) wastewater treatment plants (WWTPs) in northern Poland: "Wschód" in Gdańsk and "Dębogórze" in Gdynia. Four types of one-phase and two-phase laboratory batch tests (such as the "conventional" nitrate utilization rate (NUR), phosphate release (PRR) and anoxic/aerobic phosphate uptake (PUR), oxygen uptake rate (OUR)

measurements) were carried out with the settled wastewater without pretreatment (R1) and pretreated with coagulation-flocculation method (R2), in order to determinate the effect of X_S.

After the experimental part, this study comprised mathematical modeling and computer simulation of the activated sludge process with a special attention to hydrolysis process. The modified ASM2d incorporated one new component, rapidly hydrolyzable substrate (X_{SH}) and three new processes, i.e., hydrolysis of X_{SH} under aerobic, anoxic and anaerobic conditions. The X_{SH} concentration in the settled wastewater was estimated based on a physical fractionation of actual data from "Wschód" and "Dębogórze" WWTPs. It was assumed, that the difference between membrane filtration (under vacuum pressure) of settled wastewater through 1.2 and 0.1 µm GF/C filter, constitutes an average value of X_{SH}. Due to the fact that the experiments were carried out with parallel samples of wastewater, the X_{SH} was further subdivided into the colloidal and soluble $X_{SH,(c/s)}$ for the settled wastewater without pretreatment and only the soluble $X_{SH,(s)}$ for the settled wastewater after coagulation-flocculation. The results of OUR batch tests with the settled wastewater without pretreatment, together with the new fractionation were used to perform the optimization method of Nelder-Mead simplex. For the optimization purpose one stoichiometric and four kinetic coefficients ($Y_{H,}$ k_{hyd}, $k_{hyd,r}$, $K_{X,}$ K_{Xr}) were selected, but only 3 of them were simultaneously used each time to carry out the optimization in the 10 scenarios. After the optimization of all possible scenarios for both plants, the best one which generated the smallest average relative deviation (ARD) between measured and simulated results, was chosen to improve the modified ASM2d predictions of OUR batch tests. In the case of parallel samples with the settled wastewater after coagulation-flocculation, it was necessary to search value of the soluble $X_{SH,(s)}$ fraction in order to obtain the best fit between measured and calculated values of output parameters, based on the same kinetic and stoichiometric coefficients as set in the settled wastewater without pretreatment. Finally, average values of X_{SH} and optimized kinetic parameters were implemented to the new model for both parallel samples of wastewater in order to compare (NUR, PRR and anoxic/aerobic PUR, OUR) batch tests simulations to similar experiments in the original ASM2d.

1.4. Scope of the work

The thesis consists of five major chapters, which are further divided into several Sections. The chapters are organized in the following order: introduction (Chapter 1),

theoretical background (Chapter 2), materials and methods (Chapter 3), results and discussion (Chapter 4), and conclusions (Chapter 5). Chapter 1 presents a general background of this study with some basic definitions in the fields of different C sources, wastewater fractionation and hydrolysis. In Chapter 2, a comprehensive literature review provides insight into the approaches to present wastewater fractionation and principal processes occurring in the activated sludge systems, which are important to improve efficiency of nutrient compounds removal. The development and comparison of the most common activated sludge models are discussed in more detail in this chapter as well as the modeling of hydrolysis process and the role of slowly biodegradable substrate in full-scale biological nutrient removal activated sludge systems. Chapter 3 describes the methodology of this research including the configuration and performance of the studied WWTPs, organization of the simulation studies, containing model calibration and validation based on the results of laboratory scale batch experiments and data obtaining from studied plants. In Chapter 4, the results of this study are discussed in terms of mechanism of the hydrolysis process and verified using an existing ASM2d and its modified version. The last concluding Chapter 5, contains a summary of the main findings of this research, their significance and possible implications as well as some practical applications. Finally, a list of important and innovative studies, which have been performed in the field of hydrolysis process and activated sludge modeling, can be found in an extended bibliography. In addition, at the end of this thesis were attached four Appendices with the brief description of ASM2d as well as the results of full experimental data and mathematical modeling simulations from both studied plants.

2

Theoretical Background

2.1. Wastewater characterization

Every community produces not only gas emissions but also solid and liquid wastes. The liquid wastes according to the Council Directive 91/271/EEC are divided on three type of municipal wastewater defining as:

- ✓ "DWW – domestic wastewater" means wastewater from residential settlements and services which originates predominantly from the human metabolism and from household activities,
- ✓ "UWW – urban wastewater" means domestic wastewater or the mixture of domestic wastewater with industrial wastewater and/or run-off rain water,
- ✓ "IWW – industrial wastewater" means any wastewater, which is discharged from premises used for carrying on any trade or industry, other than domestic wastewater and run-off rain water.

Typical municipal wastewater contains a combination of carbon, oxygen, hydrogen, together with nutrients (nitrogen and phosphorus). In some cases may contain sulphur and toxic compounds (especially industrial wastewater), that potentially may be mutagenic or carcinogenic. Urea, the major constituent of urine, together with proteins, carbohydrates, lipids are important organic compounds contributing to fresh urban or domestic wastewater. The chemical composition of organic matter in municipal wastewater are presented in Table 2.1.

Table 2.1. Chemical composition of organic matter in the municipal wastewater (Morgenroth et al., 2002)

Organic matter / References	Protein	Carbohydrate	Lipid	Others
	% of total COD			
Henze (1982)	8	12	10	70
Tanaka et al. (1991)	12	6	19	63
Raunkjaer et al. (1994)	28	18	31	22
Range	**8-28**	**6-18**	**10-31**	**22-70**

These compounds, mainly in the colloidal and dissolved form, are treated by the most widely used technology for biological wastewater treatment named activated sludge process (bacterial biomass suspension). Since its was discovered in 1913 in the UK by two engineers, Edward Ardern and W.T. Lockett, conducting research for the Manchester Corporation Rivers Department at Davyhulme Sewage Works has gained-increasing importance in the treatment from classical domestic/urban to industrial wastewaters. Depending on the design and the specific application, an activated sludge wastewater treatment plant (WWTP) can achieve, besides removal of organic carbon substances, biological nitrogen (N) and phosphorus (P) reduction.

Many different activated sludge process configurations have evolved during the years in order to meet high limits, especially in the case of nutrient removal.

The main global analytical parameters such as biochemical oxygen demand (BOD) and chemical oxygen demand (COD) are used routinely to measure the organic content of wastewaters. The COD measurement was chosen as the parameter, which adequately represents the organic carbon material found in raw wastewater and mixed liquor. COD measurements allow for the calculation of mass and electron balances, which is not possible with the BOD or total organic carbon (TOC) measurements. In this balance, the amount of organic carbon is only meaningful when it is expressed in terms of various fractions with different mechanisms and rates of biodegradation (Henze, 1992). For example, hydrolysis rates determine slowly biodegradable (X_S) organic matter conversion into readily biodegradable (S_S), that can be used as a necessary carbon source for denitrification or EBPR. In turn, the X_S fraction and associated hydrolysis rates may have a direct impact on the volume of nutrient removing treatment, however, it depends on the delicate balance between the organic carbon, nitrogen and phosphorus content of the wastewater. In this respect, COD fractionation has been introduced as a very useful tool for the evaluation of biological treatment processes (Henze, 1992).

Modeling study developed by the Task Group of the International Water Association (IWA; formerly, IAWPRC and IAWQ) Henze et al. (2000a) showed, that nowadays appropriate determination of accurate COD fractionation in wastewater, together with kinetics degradation of organic matter, has a prime importance in activated sludge systems. The concentrations of all components in wastewater usually is characterised with so-called combined approach i.e. using either physical-chemical or biological methods (Petersen, 2000). This Section is devoted to the development of the wastewater COD fractionation, which is a necessary step to obtain high removal of organic matter and ability to reduce N and P in order to gain discharge limits to the surface waters (rivers and seas) by the WWTPs. Furthermore, this literature overview contains the methods employed for the determination COD fractions used in the modelling activated sludge systems, which are the key for better understanding of the role of Xs in nutrient removal treatment process.

2.1.1. Factors affecting wastewater characterization

Wastewater compounds are transformed during transportation in the municipal sewers. The nature and extent of these transformations will depend on several factors such as temperature, residence time and state of aeration within the sewer system. For example, high temperature and/or residence time could increase the biological

activity in the sewers, while low value of these parameters could reduce it. In anaerobic sewer systems with long solids retention time (SRT), sulphate reduction and acid fermentation occur, while aerobic conditions in sewers could increase COD reduction and biomass growth. Another factor, which can influence wastewater characteristics, is the use of combined or separate sewers. The combined sewers result in a lower strength of wastes due to dilutions and much higher flows with increased variability due to storm flows (Mbewe et al., 1995). One of the key factors, that influence wastewater characteristics, is the community that is served. If there is a significant input of industrial wastes into the sewer, then the wastewater characteristics can be further changed. The type of industry that is discharging to the sewer can also have a major impact on the characteristics, e.g. dairy industries may discharge compounds which are largely biodegradable, while chemical industries may discharge a larger proportion of X_S and/or unbiodegradable compounds. Sewers receiving 100% domestic wastes are also influenced by several factors. Water availability can determine whether the plant receives high or low strength wastes. The socio-economic status of the community is influential, e.g. high income communities use more water per capita and the dietary habits are different. The use of garbage grinders, detergents and wastewater treatment processes are important factors, which can influence wastewater characteristics (Naidoo, 1999). Hence, raw wastewater characteristics and particle removal efficiencies vary, resulting in a large range of particle sizes in the primary effluent. For example, Munch et al. (1980) reported, that in a primary effluent the amount of particles smaller than 1 μm was 57%, while Levine et al. (1985) found 59%. Size distributions of organic matter from different sampling points in WWTPs are presented in Table 2.2.

Table 2.2. Size distribution of organic matter samples from different technology parts of WWTP (adopted from Levine et al., 1991)

Munch et al. (1980)			Levine et al. (1985)		
Size range (μm)	**Raw wastewater**	**Primary effluent**	**Size range (μm)**	**Primary effluent**	**Secondary effluent, AS**
	% of organic matter			**% of organic matter**	
<0.001	12	9	<0.1	51	28
0.001-1	15	48	0.1-1	8	3
1-100	30	15	1-12	34	20
>100	43	28	>12	7	49

Note: AS – activated sludge

In addition, the presence of a primary settling tank can reduce the COD load up to 40%. Then, the settled wastewater has higher total nitrogen to COD (TN/COD) and total phosphorus to COD (TP/COD) ratios compared to raw wastewater (Naidoo, 1999). The other pretreatment processes which affect wastewater characteristics

include grit removal, degreasers (fats and oil removal) and dissolved air flotation. To reduce organic loading of the biological treatment step, primary treatment can be enhanced by direct filtration of raw municipal wastewater using sand filters, where iron or polymers can be used not only to remove particles but also colloidal organic matter (van Nieuwenhuijzen et al., 2001) or by coagulation using "lime-enhanced sedimentation" as proposed by Marani et al. (2004). However, primary clarification (sedimentation) and the presence of equalization tanks have a dominant effect (Mbewe et al., 1995; Henze et al., 1997). Therefore in order to sufficiently recognize unit processes occurring in activated sludge systems, it is essential to characterize the composition of the relevant substrates in the wastewater, taking into account the form of incidence and rate of compounds degradation in biological processes.

2.1.2. Wastewater COD fractionation based on the biodegradability criterias

In recent years the wastewater characterization, which allows to distinguish fractions related to the particle size and their sensitivity to the biochemical distribution, is a very important achievement. Wastewater fractions are usually characterized using physical-chemical or biological criteria. The physical-chemical characterization of wastewater is important in terms of the particle size division (e.g. soluble, colloidal and particulate fractions), whereas biological methods allow to distinguish organic compounds, according to their biodegradation rate, into biodegradable and unbiodegradable – not involved in the biological wastewater treatment process (see Section 2.1.3). Knowledge of the participation of particular wastewater influent fractions enables more exact estimation of the biological degradablity of wastewater contaminations than the generally applied COD/BOD_5 ratio (Zawilski and Brzezińska, 2009).

The COD has been adopted by Dold et al. (1980) and Ekama et al. (1986) as the main parameter to quantify organic compounds in wastewater. The soluble (S_{COD}) and particulate (X_{COD}) organic COD matter in the influent, beside microorganisms was divided, according to IWA nomenclature, into biodegradable and nonbiodegradable COD fractions. The IWA task group subdivided the total COD into the readily and slowly (S_S+X_S) biodegradable, the soluble and particulate (S_I+X_I) unbiodegradable and active biomass (X_H) fractions (Equation 2.1). The mixed liquor can also be divided in the similar way, but a distinction needs to be made between the active, endogenous, and inert sludge fraction (Naidoo, 1999).

$$Total\ COD = S_{COD} + X_{COD} = S_S + S_I + X_S + X_I + X_H \qquad \textbf{(2.1)}$$

The Ss fraction has been further divided into S_A (volatile fatty acids) and S_F (readily "fermentable" substrate). This division was made largely to improve the models EBPR systems. The distinctive feature of the readily biodegradable compounds (S_A and S_F) is that they can directly be absorbed for synthesis by microorganisms, whereas hydrolysis is required first for the utilization of X_S. The hydrolysis products, chemically similar to Ss, can be transported into the cells for intracellular metabolism (Wanner, 1994). Due to a possible significant variation in the composition of slowly biodegradable compounds (such as a large number of compounds of different size and nature) it may be difficult to characterize them only by a single hydrolysis rate (Orhon and Cokgor, 1997). This observation has been taken a step further by (e.g. Henze, 1992; San Pedro et al., 1994) and provides the basis of the recent approach to subdivide the slowly biodegradable COD into the rapidly (S_H) and slowly (X_{SH}) hydrolyzable COD fractions (Figure 2.1). A common assumption is that the high molecular weight soluble compounds are termed rapidly hydrolysable while the organic suspended solids are termed slowly hydrolysable (Wanner, 1994).

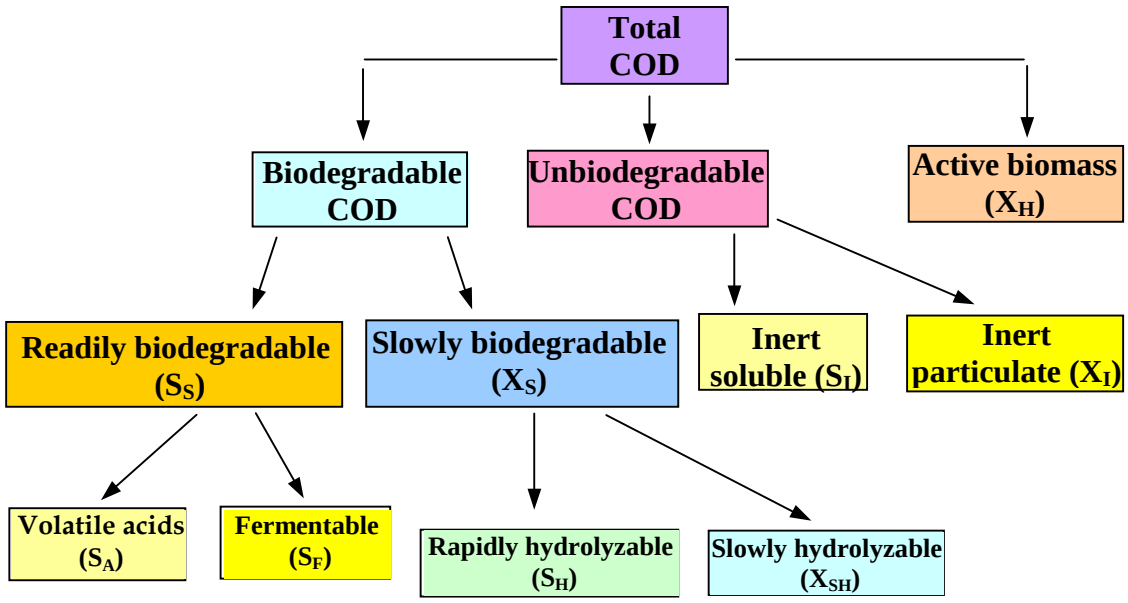


Figure 2.1. Division of influent COD into its component fractions (modified from Wentzel et al., 1995; Orhon and Cokgor, 1997).

The contribution of specific fractions may vary considerably. The problem for a COD fractionation still exist, due to the lack of definition, which clearly can distinguish between particulate and soluble COD compounds. For some authors (e.g. Henze et al., 2000b), the colloidal fraction is classified as particulate COD (X_{COD}) by the others (e.g. Kalinowska and Oleszkiewicz et al, 2001) as soluble COD (S_{COD}). The first approach assumes that the raw municipal wastewater contains the organic S_{COD} and X_{COD} fraction in the amounts of approximately 21-35% and 65-79% of total COD,

respectively. In this respect, the amount of S_S and S_I is considered between 12-25% and 8-10%, respectively, whereas X_S is approximately 50% and X_I = 15%. Moreover Łomotowski and Szpindor (1999) sugested that according to this approach the biodegradable organic compounds consist on average 61% of all organic substances in wastewater, and the remaining 39% are non-biodegradable. Supporters (e.g. Bogdańska, 2002) of the assignment of the colloidal compounds into soluble fractions indicate that, the percentage contribution of particulate fraction is 57%, while the amount of soluble and colloidal organic compounds is 43%, half (approximately 19.5%) for each, so called "readily" and "easily" biodegradable fraction and the remaining 2% for inert substances (Sadecka, 2010). The slowly biodegradable compounds in this approach constituted only 23% of total COD (Figure 2.2).

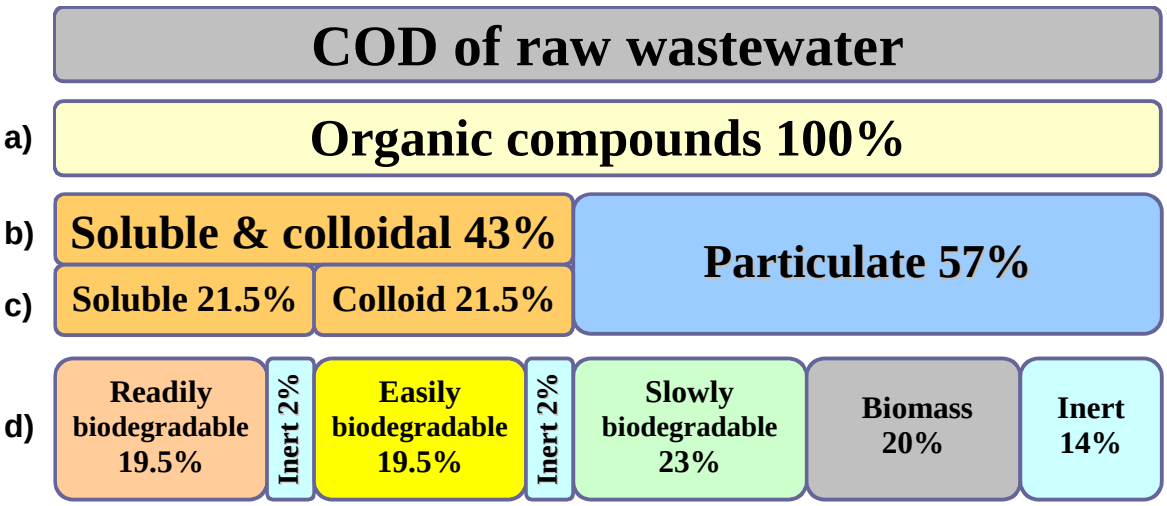


Figure 2.2. Percentage contribution of organic components to total COD in raw wastewater (a) classical model, (b) postclassical model, (c) model considering the physical state of organic compounds, (d) model considering the biodegradability of organic compounds (modified from Sadecka, 2010).

During last decades percentage contribution of organic components in total COD of municipal wastewater has been changed. According to the literature data (e.g. Ekama et al., 1986; Henze et al., 2000a; Lagarde et al., 2005), municipal wastewater after primary treatment contains readily and slowly biodegradable compounds in the amounts of approximately 10-30% and 30-60% of total COD, respectively. The investigation in four municipal WWTPs in northern Poland revealed that particulate and colloidal fractions constituted 65-70% of total COD (Czerwionka et al., 2008). In the Netherlands, obtained with the STOWA guidelines for the COD fractions (see Section 2.1.4), particulate compounds usually varied between 30 and 50% of the total COD investigated in municipal wastewater at different WWTPs (Roeleveld and van Loosdrecht, 2002), while in other countries even higher values were reported, i.e.

75% in Switzerland (Kappeler and Gujer, 1992), 77% in Turkey (Orhon et al., 1997), and 70-90% in South Africa (Casey et al., 1999). The comparison of investigation carried out by the different authors on division of COD fractions in wastewater are presented in Table 2.3.

Table 2.3. Contribution of specific fractions to total COD in raw wastewater

Total COD fractions / References	S_S	S_I	X_S	X_I	X_H
	% of $COD_{tot.}$				
Ekama et al. (1986)	20-25	8-10	60-65	5-7	13
Henze (1992)	24-32	2-11	43-49	11-20	11-20
Kappeler and Gujer (1992)	10-20	7-11	53-60	7-15	8-10
Sozen (1995)	22	9	69*		-
Orhon et al. (1997)	9	4	77*		10
Henze et al. (2000a)	12-30	5-12	30-60	10-15	5-15
Mąkinia and Wells (2000)	22-26	7-9	53-54	12-15	-
Kalinowska and Oleszkiewicz (2001)	12-25	8-10	50	15	-
Roeleveld and van Loosdrecht (2002)	9-42	3-10	10-48	23-50	-
Myszograj and Sadecka (2004)	23-29	2-3	51-56	17-19	-
Mąkinia (2006)	21-40	4-7	34-51	21-28	-
Sadecka (2010)	22-27	1-3	54-56	18-19	-
Range of fractions concentration	**9-42**	**1-12**	**10-65**	**5-50**	**5-20**
Average values ± standard deviations	**23.1±8.6**	**6.5±3.6**	**49.3±12.8**	**17.2±9.9**	**11.5±4.6**

Note: *Included X_S and X_I

2.1.2.1. Biodegradable COD fractions

The study of Stern and Marais (1974) showed that in the primary anoxic reactor, the rate of denitrification occurred in two linear phases (van Haandel et al., 1981). In the secondary anoxic reactor, there was a single linear phase due to endogenous/adsorbed organic carbon utilization. In terms of the nitrate utilization rate (NUR), it was found that the single rate in the secondary anoxic reactor is about two-third slower than the second rate in the primary anoxic reactor. It was therefore hypothesized that the two linear phases were linked to the biodegradability of the organic carbon substrate, readily and slowly biodegradable COD fraction (S_S and X_S). In the secondary anoxic reactors of plug – flow systems, the single linear phase is due to the utilization of adsorbed X_S generated from organism death i.e. endogenous respiration (see Section 2.2.4.4). Further investigations revealed, that under dynamic loading conditions e.g. plug – flow, short SRT cyclic loading and batch tests, two distinct rates of utilization were observed for either aerobic conditions (Ekama et al., 1986) or anoxic conditions (van Haandel et al., 1981; Ekama et al., 1986) (see Section 2.2.4). A subdivision of this biodegradable fraction is required if denitrification or EBPR are included in the design or the system behaviour is simulated with a

dynamic model. Unlike the soluble S_S fraction, which is exposed to biological treatment for as long as the liquid remains in the system (i.e. the time corresponding to the hydraulic residence time (HRT)), the X_S fraction is exposed to biological treatment for as long as the solids are retained in the system (i.e. the time corresponding to the SRT). Therefore, the utilization rate of X_S is about 10% in comparision with S_S, unless the SRT (as it is in most activated sludge systems) is more than 10 times longer than the HRT, and usually the X_S is completely utilized. A modelling study has shown that all the X_S is completely utilized for SRT's from 2-3 d and at temperatures of about 20 °C. At lower temperatures, longer SRT's are required (Mbewe et al., 1995).

Readily biodegradable fraction (Ss or RBCOD) consists of soluble compounds with low molecular weights and usually constitutes 10-15% of total COD in raw wastewater (Henze, 1992). Small simple molecules can pass directly through the cell wall (via passive or active uptake) for synthesis (growth) and catabolism (energy). Moreover, these compounds usually are immediately metabolized by microorganisms, i.e. transported to the cells and oxidized or converted to storage products or biomass.

The S_S can be determined by biological methods, aerobic or anoxic, continuous or batch (Ekama et al., 1986). Only readily biodegradable compounds are considered to be the substrate in heterotrophic growth, either under aerobic or anoxic conditions (Henze et al., 1987). Typical examples of Ss in raw wastewater are VFAs (especially acetic acid), alcohols (methanol, ethanol), glucose and other monosacharides or lower amino acids (Wanner, 1994). Recently according to Equation 2.2 the readily biodegradable COD was subdivided into the fermentation products (S_A) and fermentable biodegradable COD (S_F) (Mbewe et al., 1995; Henze et al., 1997 and Orhon and Cokgor, 1997), covering a wide range of compounds generally assumed, that mostly are acetate (Henze et al., 1995). As mentioned above, this division is largely required for accurate design and modelling of EBPR systems. The S_A fraction can be determined by chemical methods or gas chromatography.

$$S_S = S_A + S_F \qquad \textbf{(2.2)}$$

where:
S_S – *readily biodegradable soluble COD*
S_A – *volatile fatty acids soluble COD*
S_F – *fermentable, readily biodegradable soluble COD*

Volatile fatty acids (S_A) consists of fermentation products, such as volatile fatty acids (VFAs), constitutes 2-10% of total COD. The VFAs are present in the influent wastewater, but can also be generated in the anaerobic reactor by fermentation. The rate of VFA uptake is so rapid that it can be assumed that all VFAs in the influent can be utilized in the anaerobic reactor by (if present) polyphosphate accumulating organisms (PAOs) (Naidoo, 1999).

Readily (fermentable) biodegradable fraction (S_F or F-RBCOD) consists of fermentable, readily biodegradable organic compounds. The S_F fraction constitutes 10-20% of total COD and is considered to be directly available for biodegradation by heterotrophic organisms. It is assumed that S_F may serve as a substrate for fermentation and thus, does not include the fermentation products. The rate of the fermentation reaction is slower than the sequestration rate and the amount of S_F fermented to VFAs, which depends on the influent concentration of this fraction and wastewater treatment system design (Naidoo, 1999).

Slowly biodegradable fraction (Xs or SBCOD) constitutes 30-60% of total COD and is usually a major component of organic material in raw wastewater (Henze et al., 1995). This fraction was originally defined as particulate organics in the model of Dold et al. (1980), but later on it became evident that a wide range of compounds (soluble, colloidal and larger organic particles of complex structure) could be classified as slowly biodegradable (Orhon and Cokgor, 1997). This kind of organic matter in municipal wastewater is often removed (about 50-70% of suspended solids and 25-40% biochemical oxygen demand) in conventional primary clarification (Tchobanoglous et al., 2003).

The X_S is utilized slower and metabolized at the rates that are about 10% of the rate of S_S metabolism. This COD fraction is thought to consist of complex organic molecules which cannot pass directly through the cell wall. The utilization of this organic carbon material involves enmeshment and adsorption to activated sludge flocs. This is followed by the extracellular enzymatic breakdown of the complex compounds to simpler molecules, which are able to pass through the cell wall. The molecules are then metabolized by the microorganisms for their growth. The overall reaction rate is limited by the hydrolysis rate of the adsorbed organic carbon rather than the rate of metabolism (Ekama et al., 1986; Wentzel et al., 1992). The rate of hydrolysis of higher molecular weight compounds to S_S will limit the denitrification rate. The addition of S_S to carbon limited sludge will accelerate the denitrification rate. However, once the S_S fraction has been utilized, the denitrification rate falls back to the rate limited by the rate of hydrolysis of the X_S.

Hydrolysis of the X_S is assumed to be catalyzed by extracellular enzymes (see Section 2.2.2). There are two hypotheses with regard to the location of these extracellular enzymes:

- some authors (e.g. Henze et al., 1987; Wanner, 1994) suggested, that these large molecules are adsorbed to the surface of the biomass where hydrolysis is mediated by the cell bound extracellular enzymes. This was accepted and adopted in the UCT Model i.e. these hydrolysis products pass directly to the microorganism (Naidoo, 1999);
- however, according to the IWA Models (e.g. ASM1) the organics are hydrolyzed by extracellular enzymes and are released in the bulk liquid (Dold et al., 1991).

Rohold and Harremoes (1993) futher Larsen and Harremoes (1994) investigated this phenomenon in biofilm reactors with molasses and starch, respectively, as X_S substrates. They reported that the extracellular enzymatic breakdown of non-diffusible organics occurs in the bulk liquid and that the enzymes are washed out of the system when the HRT is decreased. However, San Pedro et al. (1994) found, during the first phase of experiment, that starch disappeared from the bulk liquid solution within a 2 hour period in suspended growth systems. This indicated a rapid adsorption to the biomass, which suggests that the X_S becomes adsorbed to the biomass before hydrolysis. Moreover, San Pedro et al. (1994) found that the second phase was characterized by a gradual decrease in the OUR profile and this was attributed to the metabolism of intracellular glycogen. The authors further presented, that the intracellular glycogen was metabolized after the exhaustion of hydrolyzable starch. The third phase in this OUR profile was attributed to an endogenous respiration phase. San Pedro et al. (1994) suggested that the difference in the rates for starch and intracellular glycogen was due to differences in the hydrolysis rates of these compounds. Such observations have resulted in some researchers suggesting that the X_S can be subdivided into smaller fractions according to their rate of hydrolysis (Henze, 1992). Although this fraction was originally defined as particulate COD (Dold et al., 1991; Dold and Marais, 1986), nowadays usually cover a wide range of particle sizes, from soluble to colloidal and larger organic particles (see Section 2.2).

Endogenous respiration, also provides a source of X_S, which occurs when the organic substrate concentration is low or absent. Consequently, the bacterial cells die and release cell materials, which are unbiodegradable and biodegradable. The biodegradable fraction becomes part of the X_S in the liquid and thus, the same cycle of adsorption, hydrolysis and utilization occurs (Randall et al., 1992; Wentzel et al.,

1992). The endogenous denitrification rate is dependent on the respiration rate of the bacteria, using the stored substratesor substrate released from endogenous decay (Randall et al., 1992). The denitrification rate depends on whether S_S or X_S serves as electron donor (substrate) and the relative proportion of these two materials will influence the amount of nitrogen removed. Phosphorus removal, however, is dependent on the available VFA (S_A) and S_F fraction.

2.1.2.2. Unbiodegradable COD fractions

The unbiodegradable (inert) COD can be divided into soluble (S_I) and particulate (X_I) fractions. The inert organic compounds can be present in raw wastewater, but cannot be further degraded in the WWTPs under normal operating conditions (Naidoo, 1999). The soluble unbiodegradable compounds (S_I) pass unchanged through any biological treatment process. The particulate non-biodegradable organic compounds (X_I) are incorporated in the activated sludge flocs and removed from the system with waste activated sludge (WAS) (Wanner, 1994). The concentration of inert particulates in primary effluent varies in the range of 15-40 g/m^3 (Stensel, 1992) and they constitute 35-40% of organic particulates in domestic wastewater (Grady et al., 1999). According to Xu and Hultman (1996), the contribution of X_I usually ranges 8-18% of total COD in raw wastewater, however Table 2.3 contains more data.

Additionally, unbiodegradable organic compounds are generated during biological treatment processes and hence the chemical composition of this fraction in the activated sludge bioreactor may significantly differ from that in the raw wastewater. The soluble inerts are produced as metabolism by-products during decay or hydrolysis (Orhon et al., 1989; Boero et al., 1991). Their amount may be even greater than the S_I in the influent (Henze, 1992). The particulate inerts are a fraction of the net biomass decay (Henze et al., 1987). Kappeler and Gujer (1992) found that the coefficient for the production of X_I from endogenous respiration equals to approximately 0.2 g COD/g COD consumed. For models using the death-regeneration concept of Dold et al. (1980), this fraction is less than 20% (Henze et al., 1987). Alternatively, a simplified approach can be used in terms of a "rough guess" wastewater concentration, which includes both generated and present inert compounds in the influent (Henze, 1992).

Unbiodegradable soluble fraction (S_I) contained in raw wastewater, constitutes certain proportion (see Table 2.3). The total soluble COD in effluent includes the unbiodegradable organic compounds, which originate from the wastewater and soluble residual COD fractions generated as soluble metabolic ($S_{I,P}$) products (Equation 2.3). Therefore, the effluent generally contains more soluble

unbiodegradable residual fraction ($S_{I,R}$) than the raw wastewater. The generation of soluble metabolic products is modelled by means of growth- or decay- associated processes (Orhon and Cokgor, 1997):

$$S_I = S_{I,R} - S_{I,P} \qquad \textbf{(2.3)}$$

where:
S_I – *unbiodegradable soluble COD*
$S_{I,R}$ – *unbiodegradable (residual) soluble COD*
$S_{I,P}$ – *unbiodegradable (metabolic products) soluble COD*

The soluble unbiodegradable materials pass out in the secondary effluent as the COD effluent. This is done by accepting that no soluble unbiodegradable organics are generated during biological treatment in the bioreactor. It is therefore, assumed that the effluent S_I (<0.45 µm filtered COD) is equal to the influent S_I (Dold et al., 1991). The nature of the soluble products is not very well recognized, i.e. if they are indeed residuals or they undergo biodegradation at a much lower rate than the biodegradable compounds in the influent wastewater (Orhon and Cokgor, 1997).

Unbiodegradable particulate fraction (X_I) becomes enmeshed in the sludge and settles out in the secondary clarifier. The microbial metabolic activity during the endogenous decay or death-regeneration phase may generated X_I, which is retained in the system to accumulate as unbiodegradable organic settleable solids (VSS). At steady state, the mass of X_I entering to the system is balanced by the mass of unbiodegradable particulates removes by sludge wastage. The X_I is generated by the bacteria during the treatment process. This material is referred to as "endogenous residue" (Ekama et al., 1986; Henze et al., 1995).

2.1.2.3. Active heterotrophic biomass fraction (X_H)

The active heterotrophic biomass is included as wastewater COD fraction in the influent (Naidoo, 1999). In the raw wastewater the X_H may vary in the range of 5–20% of the total organic matter as presented in Table 2.3 or constitute 10-80% of the volatile suspended solids (Henze, 1987). According to Henze, 1989 and Kappelar and Gujer, 1992, European wastewaters may contain a significant heterotrophic active mass fraction i.e. up to 20% of the total COD. Some of these organisms can grow under aerobic or/and anoxic conditions and others may be active anaerobically. The biomass is responsible for hydrolysis of particulate X_S and utilization of the soluble biodegradable organic compounds (Henze et al., 1995).

Naidoo (1999) suggested that in South Africa, the sewers have generally short retention time (<6 h) and it is therefore, considered unlikely to support X_H

generation. However, most of the X_H in the primary effluent may have its origin in the recycled activated sludge (RAS), which is usually transport to the inlet of the WWTP (Kappeler and Gujer, 1992). Seeding of this fraction into the activated sludge system may have a significant impact on modelling and design.

2.1.3. Wastewater characterization methods

The accuracy of the input data is dependent on the methods used to determine the wastewater fractions (Table 2.4). The biodegradability of a specific compound is in general associated with the readily biodegradable COD fraction. Since the Ss is modelled as a simple soluble compounds, physical-chemical methods have been tested in order to find procedures, which are as comparable and reliable as the biological respirometric tests. The biological methods are based either on oxygen uptake rate (OUR) measurements, or on nitrate utilization rate (NUR) measurements and their time profile (NUR) in anoxic batch tests. The physical-chemical methods are instead based on sample filtration, preceded with or without a flocculation step. These methods do not give any useful information on biomass behavior, but can give an estimate of the readily biodegradable COD fraction (Onnis-Hayden and Gu, 2008). In practice, the characterization of wastewater, typically ends up with a combined approach using both physical-chemical and biological methods to obtain an estimate of the all influent components concentrations. The features and limitations of major methods to accomplish this task are summarised and discussed in the following Sections.

Table 2.4. Summary of the major methods proposed in the literature to determine the wastewater characterization (adapted from Onnis-Hayden and Gu, 2008)

Method / Fraction	Continous flow	Physical-chemical	OUR	Anoxic batch	Aerobic batch
Total COD	X	X	X	X	Wentzel et al. 1999
Inert Particulate	Ekama et al. (1986)	X	X	X	X
Inert Soluble	X	Mamais et al. (1993)	X	X	X
Readily biodegradable	Ekama et al. (1986) Sollfrank and Gujer (1991)	Mamais et al. (1993)	Ekama et al. (1986 Kappeler and Gujer (1992) Xu and Hasselblad, (1997)	Ekama et al. (1986)	Ekama et al. (1986) Kappeler and Gujer (1992) Wentzel et al. (1995)
Slowly biodegradable	X	X	X	Ekama et al. (1986)	X

2.1.3.1. Physical-chemical vs. biological wastewater characterization

The physical-chemical method of wastewater characterization can be a relatively simple manner to separate different components in wastewater. The difference in molecular size can give an indication on biodegradability, because small molecules can be taken up directly over the cell membranes, whereas larger molecules need to be broken down prior to uptake (e.g. Mamais et al., 1993; Naidoo, 1999).

The general classification of compounds, due to their physical state (soluble and suspended) is the particle diameter (Bever et al., 1997). According to Henze et. al. (2002) there is no clear definition, of what are suspended particles and soluble compounds. A fractionation can be made by filtering through filters of various pore size: 1.6; 1.2; 1.0; 0.45; 0.1 µm. In early studies, the wastewater components were separated physically into four size-depending fractions by successive sedimentation, centrifugation, and filtration. The fractions were classified as settleable, supracolloidal, colloidal, and soluble (Rickert and Hunter, 1971), and were analysed for COD. An important conclusion from these early studies was that particles smaller than 1.0 µm were approximated to be the true soluble fraction. Moreover, the particles smaller than 1.0 µm were observed to be more rapidly degradable than particles larger than 1.0 µm. Later on Levine et al. (1985) studied the size distribution of the organic matter in wastewater and the relationship with different wastewater treatment processes. The authors concluded that separation over a membrane with a pore size of 0.1 µm was valid for a differentiation between the soluble and particulate organic fractions. The organic particles smaller than 0.1 µm are typically cell fragments, viruses, macromolecules and miscellaneous debris. The major groups of macromolecules in wastewater are polysaccharides, proteins, lipids and nucleic acids. The fraction measured by the standard test for suspended solids (1.2 µm) includes protozoa, algae, bacterial flocs and single cells. However, some bacterial cells, cell fragments, viruses and inorganic particles have a size from 0.1 to 1.2 µm and will thus also pass through the more typically applied filter size of 0.45 µm for separation between soluble and suspended fractions (Levine et al., 1985). The size of colloidal matter is typically in the range of 0.1-0.45 µm are called “low”, whereas material with a size larger than 0.45 µm usually settles are called “high” colloidal (Czerwionka and Mąkinia, 2009). The division of organic compounds due to the pore size in the approach of “true” and conventional fractionation shown in Figure 2.3.

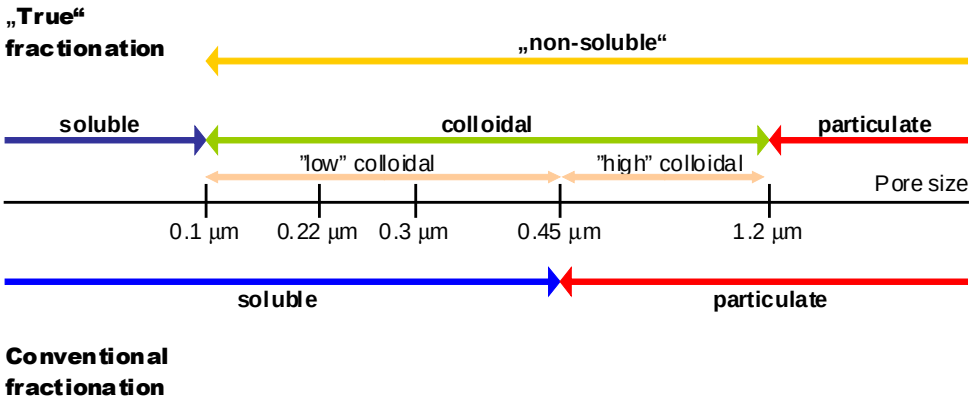


Figure 2.3. The division of organic compounds based on the filter pore size in the approach of "true" and conventional fractionation (Czerwionka and Mąkinia, 2009).

Physical characterization, important for estimating the potential efficiency of the primary clarifiers (Water Research Commission, 1984), involves the separation of wastewater into soluble, colloidal, and particulate fraction. The average concentrations of various components in the Danish municipal wastewater are presented in Table 2.5.

Table 2.5. Average composition of domestic wastewater based on the physical characterization (Henze and Harremoes, 1992)

Component / Fraction	COD	BOD_5	Total N	Total P
	g COD/m³	g BOD_5/m³	g N/m³	g P/m³
Particulate	200	85	4	1
Colloidal	65	35	6	1
Soluble	85	40	20	6
Total	**350**	**160**	**30**	**8**

Chemical characterization of wastewater comprises identification and measurement of the following constituents (Water Research Commission, 1984):

- carbonaceous and nitrogenous constituents including biomass;
- inorganic constituents, principally alkalinity, acidity, pH, and phosphorus.

Organic constituents in wastewater can be further categorized, based on the characterization of specific groups of chemical compounds, such as lipids, proteins, carbohydrates, etc. or fractionation of the organic constituents, according to their rate of biodegradation (Wanner, 1994). The latter approach (organic fractionation) is primarily related to modeling activated sludge systems and has its origin in the multi-substrate models of Dold et al. (1980) and Henze et al. (1987). The major difference between the organic fractions is the rate of their utilization by microorganisms (see Section 2.1.2). Expressed in the "oxygen demand" units, the organic compounds can be divided into three major categories: readily biodegradable, slowly biodegradable, and non-biodegradable (inert) organic

compounds (Henze et al., 1987; Henze, 1992; Wanner, 1994; Barker and Dold, 1997a; Orhon and Cogkor, 1997). Conventional analytical parameters, such as BOD and COD, routinely used to quantify collectively the concentrations of organic compounds in wastewater, cannot directly differentiate between the individual categories (Orhon and Cokgor, 1997). The advantage of using COD is that it provides a consistent basis for the description of the activated sludge process including relationships between substrate, biomass and electron acceptor (DO and nitrate) (Barker and Dold, 1997a).

Methods used for separating wastewater fractions include: sedimentation, centrifugation, filtration and precipitation. Filtration methods with several pore sizes have been investigated, because it has been suggested that the difference in biokinetic response to the S_S and X_S is due to differences in molecular size. These fractions consists of small molecules, which can easily pass into the microbial cells (S_S) or comprises complex molecules which require extracellular breakdown before cell utilization (X_S). It has been found that membranes with a molecular weight limit of less than 10 000 daltons (Da) gave S_S concentrations similar to that determined in biological respirometric tests. However, it has also been reported that with textile wastewater, these membranes gave a S_S concentration lower (13% of total COD) in comparision with (20% of total COD) derived in batch test (Bortone et al., 1994; Wentzel et al., 1995)

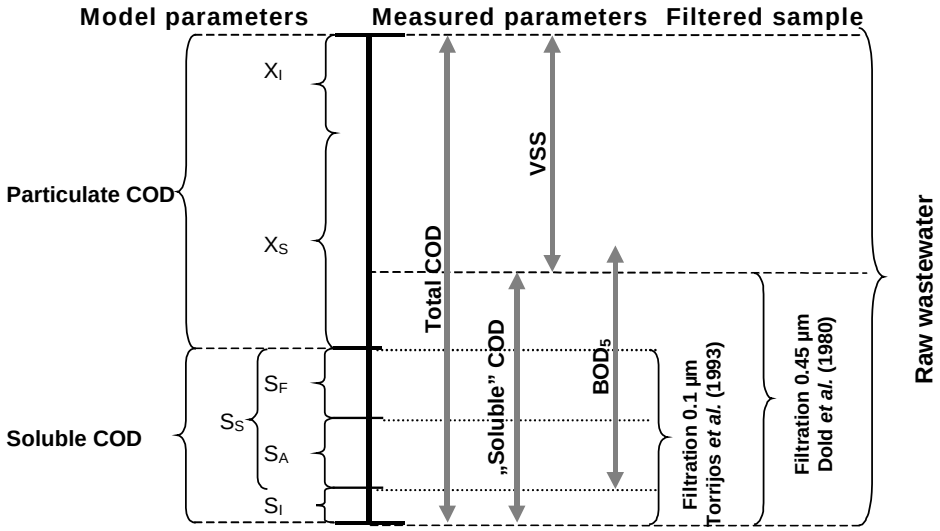


Figure 2.4. The COD fractionation in raw wastewater and tipical analytical techniques for measuring parts of total COD (adapted from Mąkinia, 2006).

In most popular mathematical models of activated sludge process (e.g. ASM1, ASM2, ASM2d, ASM3) in order to separate soluble and particulate compounds, filtration through a membrane 0.45 µm pore size has been recommended. Dold et al. (1980)

assessed 0.45 µm filters and found that a small fraction of the X_S of domestic wastewater passed through the filter, which resulted in an overestimation of S_S fraction. Evidently, the activated sludge models (ASMs) do not differentiate on the filtered sample between colloidal and settleable wastewater fractions. Torrijos et al. (1994) found that wastewater passed through a 0.1 µm filter gave a true indication of the S_S fraction (Figure 2.4). Several other researchers have attempted to classify the soluble fraction, but their results and methods vary. The cut-off utilized for the characterization of the soluble fraction has varied from < 0.001 µm (Pouet and Grasmick, 1994); <0.01 µm or <0.03 µm (Henze and Harremoes, 1990) and <0.45 µm (Henze et al., 1995). This soluble fraction constitutes approximately 24-30% of COD in the raw wastewater (e.g. Pouet and Grasmick, 1994; Henze and Harremoes, 1990). Henze and Harremoes (1990) determined colloidal particle sizes between 0.01 and 10 µm, while Pouet and Grasmick (1994) or Klimiuk and Łebkowska (2004) classified this fraction in the range of 0.001-1 µm. In addition, another fraction called the supracolloids was considered by Henze and Harremoes (1990) as the size >10 µm and by Pouet and Grasmick (1994) as 1-100 µm size range, while Klimiuk and Łebkowska (2004) did not consider this fraction. Therefore, these differences in the sizes resulted in variations in the percentage of these fractions found in wastewater (Table 2.6).

Table 2.6. Size distribution of different components in raw wastewater

References / Fraction	Henze and Harremoes (1990)		Pouet and Grasmick (1994)		Klimiuk and Łebkowska (2004)	
	Size range (µm)	(%)	Size range (µm)	(%)	Size range (µm)	(%)
Soluble	< 0.01	24	<0.001	30	<0.001	24
Colloidal	0.01-10	19	0.001-1	35	0.001-1	18.5
Supracolloidal	>10	57	1-100	c.f.	n.c.	n.c.
Settleable	n.c.	n.c.	>100	35	>1	57.5

Note: n.c. – not considered; c.f. – included in colloidal fraction

Mamais et al. (1993) proposed a physical-chemical method based on coagulation-flocculation of the suspended and colloidal material in wastewater. With this method, a sample of wastewater is flocculated by adding 1 cm^3 of a solution of $ZnSO_4$ (100 mg/dm^3) to 100 cm^3, mixing vigorously for approximately 1 min, and adjusting the pH with NaOH (6M solution). The sample is then allowed to settle quiescently before clear supernatant is withdrawn and filtered through a 0.45 µm membrane filter. Based on this rationale Mamais et al. (1993) showed that a filtrate contained only "truly" soluble (SCOD) organic matter: both biodegradable (Ss) and inert (S_I). This method makes use of an equation proposed by Ekama et al. (1986) and the IWA Task Group (Henze et al., 2000a), which relates the influent Ss to the "truly" soluble influent COD (Equation 2.4).

$$S_S = SCOD - S_I \qquad \textbf{(2.4)}$$

where:
S_S – *influent readily biodegradable soluble COD, M(COD)L^{-3}*
$SCOD$ – *influent truly soluble COD i.e. after coagulation-floculation, M(COD)L^{-3}*
S_I – *influent inert COD, M(COD)L^{-3}*

The difference between the SCOD and S_I measurments allows to estimate the Ss concentration (Table 2.7). Results of S_S, which were obtained from the physico-chemical (flocculation) and the biological method were highly comparable (Mamais et al., 1993).

Table 2.7. Comparison of the readily biodegradable COD concentrations from the physico-chemical (c-f) and biological method for different wastewater sources (Mamais et al., 1993)

Wastewater source	SCOD (c-f)	S_I (c-f)	S_S (c-f)	S_S (biological)
	mg O_2/l			
Raw wastewater	63	41	23	22
Primary effluent 1	99	37	62	65
Primary effluent 2	84	52	32	32
Primary effluent and acid digester centrate	163	53	110	119

A disadvantage of this method is the necessity for quantifying the S_I independently, which is a time consuming procedure (Wentzel et al., 1995). In addition, the S_I fraction may also contain soluble microbial products generated in the result of biomass decay (these products are biodegradable). Mamais et al. (1993) hypothesized that the soluble fraction of wastewater contains only S_S and S_I, while Orhon and Cokgor (1997) found that the soluble fraction consists of S_S, S_I and S_H. In this case, the S_S calculated by the method presented above would result in an overestimation of the actual S_S concentration.

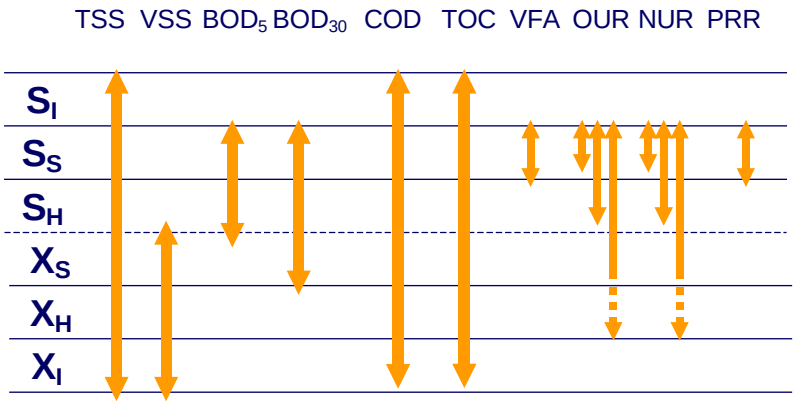


Figure 2.5. The physical-chemical and biological procedures for determining various organic fractions in wastewater (Henze, 1992).

Henze (1992) presented an overview of the early methods for determining wastewater fractions (Figure 2.5). These procedures have been continuously developed and evaluated (e.g. Spanjers and Vanrolleghem, 1995; Kristensen et al., 1998; Orhon and Cokgor, 1997; Spanjers et al., 1998; Vanrolleghem et al., 1999; Petersen et al., 2002). The most common methods for determining of each wastewater components nowadays, is discussed in this Section more comprehensively.

2.1.3.2. Readily biodegradable fraction (S_S)

Most methods used for determining S_S rely on respirometric measurements conducted in continuous or batch reactors, under either aerobic or anoxic conditions (Orhon et al., 1997). It should be noted, however, that these methods are based on the approach proposed in the ASM1 (Henze et al., 1987) that the S_S is directly used for growth of microorganisms without considering any storage phenomena. In the presence of intracellular storage polymers, the interpretation of OUR profiles becomes confusing (see Section 2.2.3).

The fraction of S_S can be measured through the OUR in the continuous reactors (Ekama et al., 1986; Sollfrank and Gujer, 1991; Witteborg et al., 1996) as well as in the batch reactors (Kappeler and Gujer, 1992; Kristensen et al., 1992; Orhon et al., 1994; Xu and Hasselblad, 1997) (see Table 2.4). Similarly, a NUR in the batch reactors can be used to determine the concentration of S_S (Ekama et al., 1986; Kristensen et al., 1992; Orhon et al., 1994).

The S_S concentration can also be estimated by measuring the change in OUR in a single completely mixed reactor operated at a very short SRT of 2 days under a daily cyclic square feeding pattern (12 h with and without feed) as presented in Figure 2.6. A rapid drop in the OUR profile (ΔOUR) following feed termination, associated only with utilization of S_S, can be used to find its concentration (Ekama et al., 1986):

$$S_S = \frac{V \cdot \Delta OUR}{Q_{in} \cdot (1 - Y_H)} \qquad \textbf{(2.5)}$$

where:

S_S – *soluble, readily biodegradable COD,* $M(COD)L^{-3}$
ΔOUR – *rapid drop in the oxygen uptake profile following feed termination,* $M(O_2)L^{-3}T^{-1}$
Y_H – *heterotrophic yield coefficient,* $M(COD)\ M(COD)^{-1}$
V – *volume of completely mixed reactor,* L^3
Q_{in} – *inflow rate,* $L\ h^{-1}$

Witteborg et al. (1996) proposed another continuous flow method, based on the measurement of OUR under different wastewater loading conditions (high,

intermediate and endogenous) applied to a continuously operated respirometer. In this approach the concentration of S_S is calculated by solving a set of mass balance equations corresponding to the different loading conditions (Mąkinia, 2006).

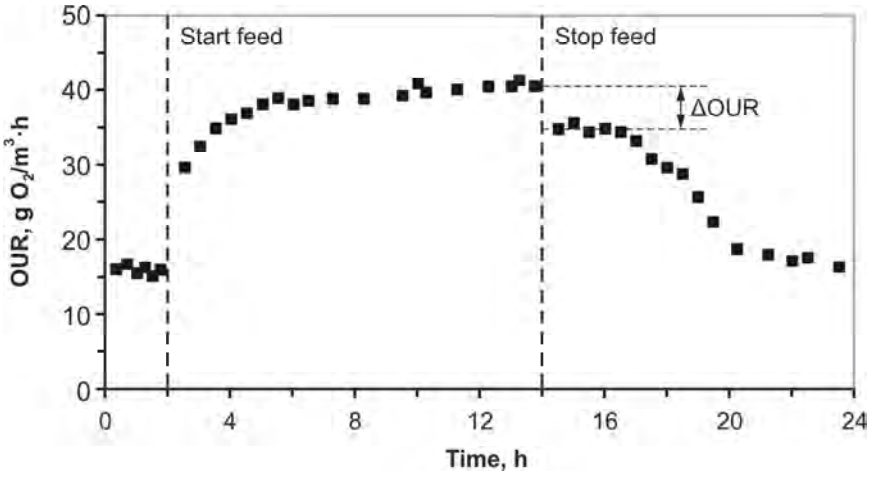


Figure 2.6. The OUR measurements in a completely mixed reactor fed continuously under a daily cyclic square wave feeding pattern (Ekama et al., 1986).

The batch test results for determining S_S concentration, was originally proposed by Ekama et al. (1986) and developed further by Kappeler and Gujer (1992). In the standard test a sample of wastewater is mixed with endogenous biomass and carried out under aerobic conditions with the monitoring of respiration rate until it reaches the endogenous level (Figure 2.7a). At the beginning of experiment nitrification has to be stopped by addition of an inhibitor, usually allylthiourea (ATU). The selection of an appropriate initial F/M (substrate/biomass) ratio provides to make a distinction between the heterotrophic OUR (OUR_H) related to the S_S and X_S utilization. Carucci et al. (2001) suggested that in order to separate these two phases the initial F/M ratio of 0.05-0.3 g COD/g VSS should be used, whereas Kappeler and Gujer (1992) recommended to use a volumetric ratio of activated sludge and wastewater in approximately amount 1/2. The initial high OUR_H, as suggested Ekama et al. (1986), remains constant over a 1-3 h period. Then after depleting S_S it rapidly drops to a lower level associated with hydrolysis of the remaining X_S, finally reaches the endogenous level, $OUR_{H,end}$. The concentration of S_S in wastewater can be calculated from the following relationship (Ekama et al., 1986; Kappeler and Gujer, 1992; Orhon et al., 1994):

$$S_S = \frac{\int OUR_H \cdot dt - \int OUR_{H,end} \cdot dt}{1 - Y_H} \qquad \textbf{(2.6)}$$

where:

S_S – *soluble readily biodegradable COD, M(COD)L^{-3}*
OUR_H – *heterotrophic oxygen uptake rate, M(O$_2$)L^{-3}T^{-1}*
$OUR_{H,end}$ – *endogenous heterotrophic oxygen uptake rate, M(O$_2$)L^{-3}T^{-1}*
Y_H – *heterotrophic yield coefficient, M(COD) M(COD)$^{-1}$*

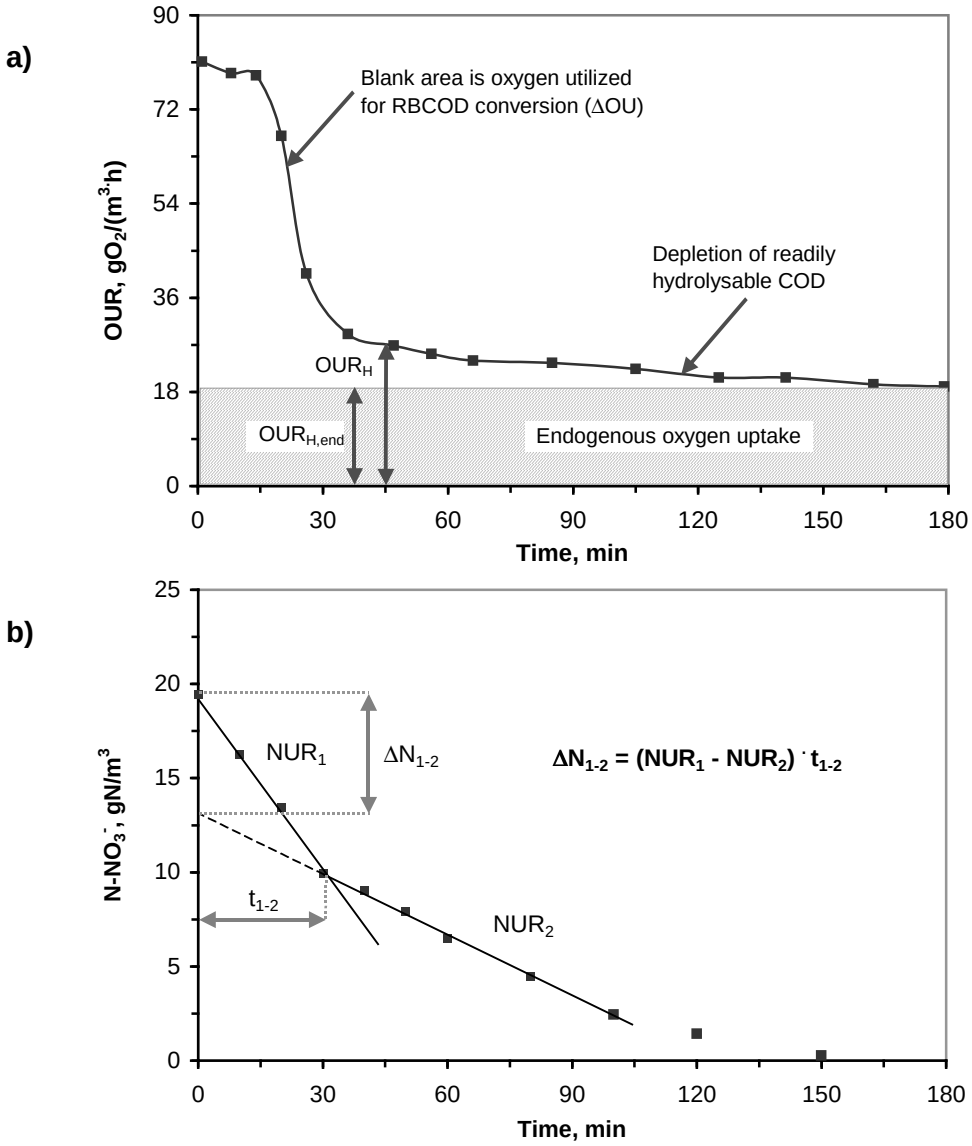


Figure 2.7. Interpretation of the batch test results for determining S_S (a) under aerobic conditions based on the method of Kappeler and Gujer (1992) and (b) under anoxic conditions based on the method of Orhon et al. (1994).

In a similar way based on the NO_3-N profile during the anoxic batch test, the concentration of S_S can also be determined (Figure 2.7 b). The initial nitrate utilization rate (NUR_1), in this experiment, is faster due to oxidation of the S_S. During this period the amount of nitrate (ΔN_{1-2}) consumed could be used to quantify the S_S concentration from the following relationship (Orhon et al., 1994):

$$S_S = \frac{2.86}{1 - Y_H} \cdot \Delta N_{1-2} \qquad \textbf{(2.7)}$$

where:

ΔN_{1-2} – amount of nitrate consumed due to oxidation of the readily biodegradable substrate, $M(N)L^{-3}$

Cokgor et al. (1998) evaluated both anoxic and aerobic respirometric batch tests to calculate the value of S_S in municipal wastewater. The authors found that the S_S concentrations in wastewater obtained from the anoxic reactors were higher by approximately 14% than the values from the parallel aerobic reactors, when equal values for anoxic/aerobic yield coefficients for heterotrophic biomass were assumed. It was concluded that differently (lower) anoxic yield for biomass compared to the corresponding aerobic value were needed.

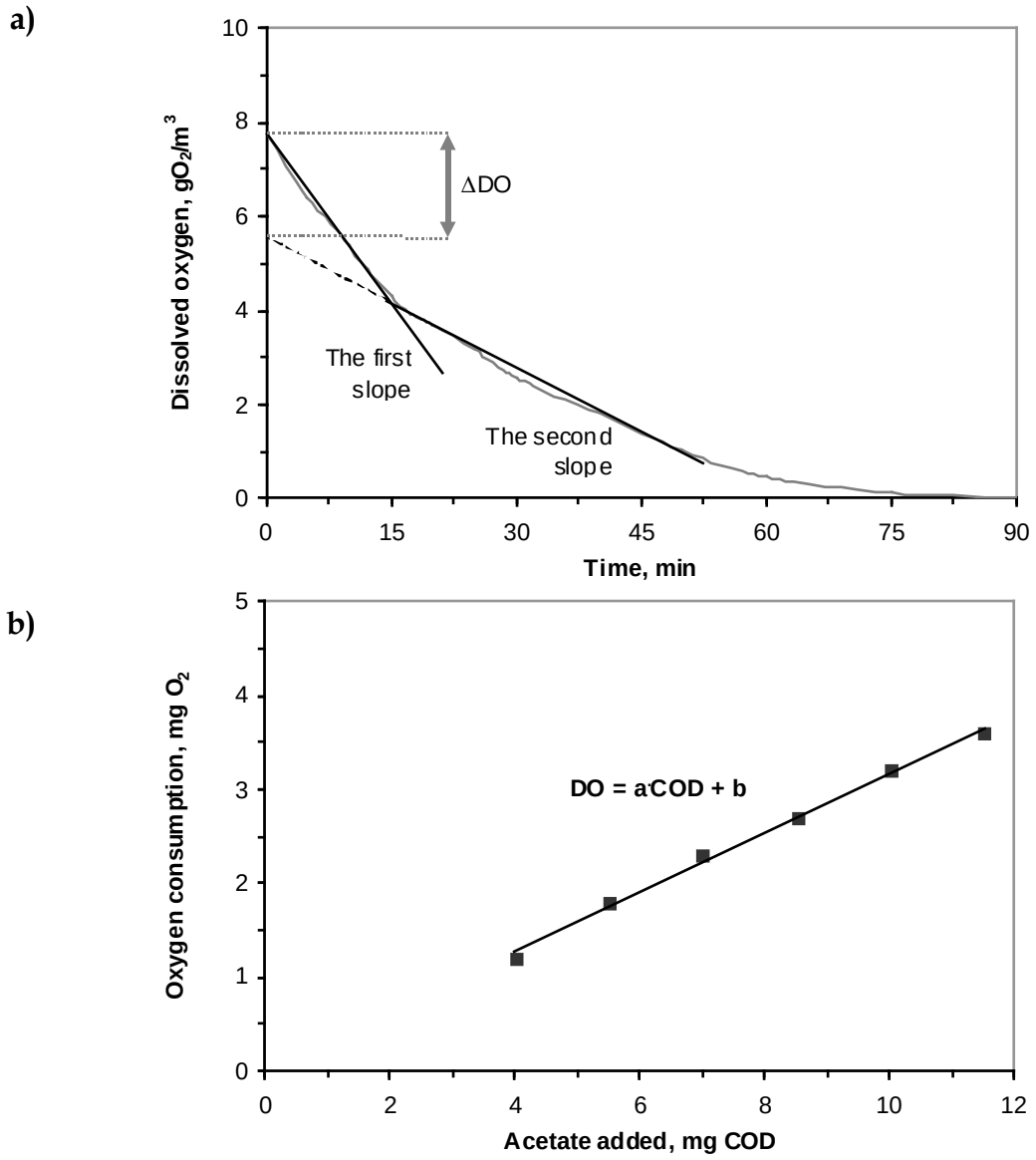


Figure 2.8. Estimation of the S_S according to the single-OUR method (Xu and Hasselblad, 1997).

A simple method for determining the S_S concentration, called the "single-OUR" (Xu and Hasselblad 1997), assumes only the monitoring of a single depletion curve of DO concentration (Figure 2.8 a). At the beginning of test the DO concentration decreases rapidly, due to the utilization of S_S (the first slope) and along with this process a

change in the OUR profil can be observed (the second slope). Then dissolved oxygen demand (ΔDO) that corresponds to the amount of S_S can be determined graphically from the DO profile. Finally, the ΔDO could be compared with a calibration curve to quantify the S_S concentration (Figure 2.8 b). This approach was experimentaly performed earlier by conducting several "single-OUR" tests with a substrate (e.g. acetate) having a known value of COD and oxygen demand for its removal. The relatively short time (approx. 30 min), compared to the traditional "multi-OUR" tests of Ekama et al. (1986) and Kappeler and Gujer (1992) which require 2-3 hours to determine the concentration of S_S is an advantage of this method. Ziglio et al. (2001) validated the "single-OUR"experiment by comparing it with the traditional batch method of Ekama et al. (1986) at a large full-scale WWTP (100,000 PE). During 4 measurements the comparison between both methods revealed only minor difference of 2% (±1.5 g COD/m^3).

In order to determine the value of S_S, the physical-chemical (i.e. flocculation-coagulation) method developed by Mamais et al. (1993) could be used (see Section 2.1.3.1), alternatively to respirometric measurements. Hu et al. (2002a) confirmed that the COD calculated with this method corresponded closely with the low (<1000 Da) molecular weight fraction. Grady et al. (1999) confirmed, that the physical-chemical method for domestic wastewater, gives results remaining in well correlation with the traditional batch test of Ekama et al. (1986). This statement is in contradiction to the observations from other studies, which revealed that the filtered COD is equal to the "truly" soluble SCOD rather than only to the S_S (Xu and Hultman, 1996). The SCOD is a sum of three fractions:

$$SCOD = S_S + S_H + S_I \qquad \textbf{(2.8)}$$

where:

S_H – *soluble, rapidly hydrolysable organic compounds, M(COD)L^{-3}*

It should be noted, that different respiration rates may occur for the same concentration of SCOD, due to the presence of S_H in the membrane filtrate (Sollfrank and Gujer, 1991). Ginestet et al. (2002) characterized samples of raw, settled and coagulated (i.e. settled and precipitated with $FeCl_3$) wastewater by respirometric measurements, originatin from seven French WWTP. The pretreated wastewater predominantly consisted of the S_H fraction (37-90%), whereas the S_S and S_I accounted for 2-27% and 2-47% of soluble COD, respectively. Moreover Naidoo et al. (1998) examined the raw wastewater from eight WWTPs in different European countries (Table 2.8). The total COD (COD_{tot}), COD after centrifugation (COD_{cent}), COD after filtration through a 0.45 μm (COD_f) and COD after coagulation and centrifugation

($COD_{coag+cent}$) were determined. Beside these four parameters, additionally the concentration of S_S, ($RBCOD_{NUR}$) was calculated from Equation 2.7 based on the specific NURs. The estimated S_S ranged between 7 and 19% of total COD and these values were significantly lower than the SCOD determined by the other methods, i.e. physical (filtration) or physical-chemical (coagulation-centrifugation). According to Carrette et al. (2001) significant deviations between the respirometric measurements and physical-chemical methods may be observed for samples with a high content of industrial wastewater.

Table 2.8. Results of raw wastewater characterization at eight European municipal WWTPs (Naidoo et al., 1998)

WWTP	COD_{tot}	COD_{cent}	COD_f	$COD_{coag+cent}$	$RBCOD_{NUR}$
	g COD/m³	% of COD_{tot}	% of COD_{tot}	% of COD_{tot}	% of COD_{tot}
Crespieres (France)	549	57	39	31	7
Morainvilliers (France)	344	49	48	41	15
Boran (France)	707	65	57	50	10
Plaisir (France)	691	32	30	25	11
Rostock (Germany)	953	30	27	24	19
Berwick (England)	913	n.d.	51	41	n.o.
Orense (Spain)	407	n.d.	32	17	14
Brno (Czech Republic)	250	n.d.	40	32	10

Note: n.d. – not determined, n.o. – not observed

2.1.3.3. Slowly biodegradable fraction (Xs)

The slowly biodegradable substrate in the particulate form is a major fraction of organic material in municipal wastewater and has to be hydrolyzed, before it can be taken up and be degraded by bacteria (Levine et al.,1985). In wastewater treatment systems, the process of hydrolysis summarizes all mechanisms that make X_S available for bacterial growth (Gujer et al., 1999). The hydrolysis rate is lower than the utilization rate of S_S, which makes hydrolysis the limiting step in heterotrophic growth only on the X_S (Henze et al., 1987).

The OUR measurements were undertaken by Scheumann (2010) to determine the growth and decay rate of X_H, as well as the hydrolysis rate. The evaluation of kinetic data for greywater treatment with a submerged membrane sequencing batch reactor was performed with different municipal wastewater. The Figure 2.9 gives an example of the measurements by the OUR batch test with five different phases. The first phase shows the exponential OUR increase, due to the growth of heterotrophic active biomass. In the second phase, the OUR drops instantly, due to depletion (see t_d in Figure 2.9) of S_S. At the third phase, the OUR shows the growth of microorganism,

due to X_S depletion (phase IV). The growth on X_S becomes only evident when the S_S concentration becomes limiting (see Section 2.2.3). Despite the fact that wastewater models do not show the evident growth on X_S, the OUR in phase I is assumed to be the sum of S_S and X_S utilization (Wentzel et al. 1995). Ultimately, the OUR decreases slowly and this corresponds to endogenous respiration and decay (phase V).

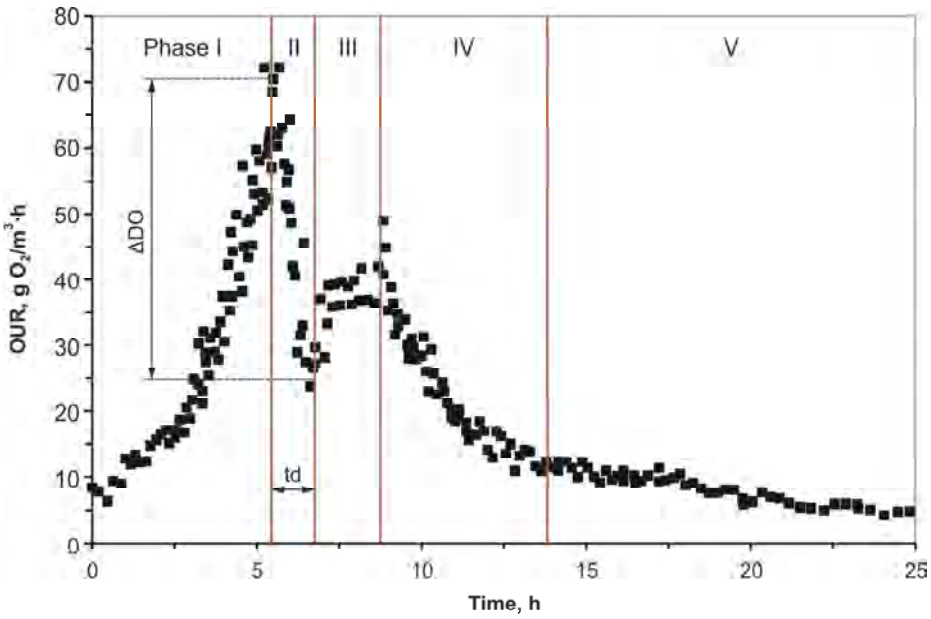


Figure 2.9. Example of the fractination method based on results of the OUR batch test (Scheumann, 2010)

The "optimization approach" proposed by Vanrolleghem et al. (1999) can be used to estimate the value of X_S together with k_{hyd} constant, based on iterative curve fitting according to Kappeler and Gujer (1992) study. The X_S can also be determined by rearranging Equation 2.11 after estimating the concentration of X_I using the method of Lesouef et al. (1992) (Equations 2.12-2.14). Due to the uncertainty of the BOD analysis, the fractions X_S and X_I should be further calibrated. Meijer et al. (2001) demonstrated a proper way to evaluate the estimated X_S/X_I ratio using mass balances over the activated sludge system.

2.1.3.4. Unbiodegradable soluble fraction (S_I)

The S_I fraction is considered to be equal to the effluent soluble COD of an activated sludge system, treating the influent in activated sludge systems (HRT > 3 days). In such a case, the concentration of S_I can be estimated by measuring filtered SCOD in the effluent from a WWTP (Ekama et al., 1986; Sollfrank et al., 1992; Stensel, 1992). Then the value of S_I may be assumed as 90–95% of the effluent SCOD (Henze et al., 1994). However, more precise method of the S_I estimation was proposed by Henze (1992) and Petersen et al. (2002):

$$S_I = COD_{sol,out} - CBOD_{sol,out} = COD_{sol,out} - 1.5 \cdot CBOD_{5,sol,out} \quad \textbf{(2.9)}$$

where:
$CBOD_{5,sol,out}$ – *soluble carbonaceous* BOD_5 *in secondary effluent,* $M(BOD)L^{-3}$
$CBOD_{sol,out}$ – *soluble carbonaceous BOD in secondary effluent,* $M(BOD)L^{-3}$
$COD_{sol,out}$ – *soluble COD in secondary effluent,* $M(COD)L^{-3}$

Henze et al. (1987) proposed a different method for determining the S_I fraction. An aliquot sample was removed from the continuously fed reactor operating with completely mixed liquor at a solids retention time (SRT) in excess of 10 days. Then the sample was aerated in a batch reactor until the SCOD remained constant. The S_I fraction can also be estimated from a batch test after continuous aeration of a filtered sample of the influent for several days, usually 1-2 weeks (Lesouef et al., 1992). Hence, the major disadvantage of both methods is the inability to differentiate between the S_I in the influent and the soluble residual fraction of microbial products, which may partly be biodegradable. This approach is acceptable for the characterization of municipal wastewater, but not recomended for industrial wastewater especially these strong concentrated. (Orhon and Cokgor, 1997). The effluent soluble inert organics, in such a case, should be assumed as the influent ($S_{I,in}$) and produced ($S_{I,prod}$) fractions:

$$S_{I,out} = S_{I,in} + S_{I,prod} \quad \textbf{(2.10)}$$

where:
$S_{I,in}$ – *influent soluble unbiodegradable organic compounds,* $M(COD)L^{-3}$
$S_{I,out}$ – *effluent soluble unbiodegradable organic compounds,* $M(COD)L^{-3}$
$S_{I,prod}$ – *soluble unbiodegradable organic produced in the activated sludge system,* $M(COD)L^{-3}$

2.1.3.5. Unbiodegradable particulate fraction (X_I)

The X_I should be estimated in wastewater by comparing the observed and predicted sludge production in a real WWTP as a function of time (Henze et al., 1987) This approach allows to tune the model to the specific wastewater and compensates any error made in the Y_H or b_H estimation. However, exist other possibility, i.e. batch test should be carried out only with wastewater. The initial S_S, X_S, X_H concentrations can be determined by curve fitting, while the total COD and the S_I can be measured directly in sample of wastewater. Then the value of X_I can be calculated as follows (Kappeler and Gujer, 1992):

$$X_I = Total\,COD - S_S - S_I - X_S - X_H \quad \textbf{(2.11)}$$

where:

X_I – *particulate unbiodegradable organic compounds,* $M(COD)L^{-3}$

In parallel batch reactors with non-filtered and filtered wastewater Lesouef et al. (1992) determined the X_I based on the long-term aeration. The final COD of the non-filtered sample consists of both fractions the S_I, X_I, and the produced biomass:

$$Final\ COD = S_I + X_I + Y' \cdot (Initial\ total\ COD - S_I - X_I) \quad \textbf{(2.12)}$$

where:

Y' – apparent yield coefficient for degrading the matter remaining in the filtrate, M(COD)[M(COD)]$^{-1}$

Rearranging Equation 2.12 the X_I can be estimated from the following relationship:

$$X_I = \frac{Final\ COD - S_I - Y' \cdot (Initial\ total\ COD - S_I)}{1 - Y'} \quad \textbf{(2.13)}$$

$$Y' = \frac{Particulate\ COD\ formed}{COD\ degraded} = \frac{Final\ COD\ (filt.sample) - Final\ SCOD\ (filt.sample)}{Initial\ SCOD\ (non\ filt.sample) - S_I} \quad \textbf{(2.14)}$$

2.1.3.6. Active heterotrophic biomass fraction (X_H)

The X_H concentration in the influent wastewater was determined by Xu and Hultman (1996) based on comparing the measured OUR values with a conversion factor of 150 mg O_2/(g VSS·h) assumed to correspond to the maximum OUR for X_H (Henze, 1986). The wastewater should be aerated until DO concentration reaches 6–8 g O_2/m^3 and before the measurement, the nitrification process should be stopped by the addition of ATU. The X_H concentration is then given by the following equation:

$$X_H = \frac{OUR_w}{150} \cdot 1000 \cdot i_{CV} \quad \textbf{(2.15)}$$

where:

OUR_w – *oxygen uptake rate obtained by performing a respirometric test with wastewater only, M(O_2)$L^{-3}T^{-1}$*
i_{CV} – *COD/VSS ratio, M(COD)M^{-1}*

The X_H can be also estimated, along with the concentrations of S_S and X_S by curve fitting against the experimental data from a batch test performed only with wastewater (Kappeler and Gujer, 1992). It should be noted, that in some activated sludge models, where the presence of X_H is not considered, the heterotrophic biomass fraction is included into the X_S. This assumption does not affect the general modeling significantly, when the Y_H coefficient is increased by approximately 10% (Henze, 1992). In this circumstances, the value of Y_H could be diverse, depending on activated sludge models 0.67 (ASM1) and 0.63 (ASM2d).

2.1.4. Integrated methods of wastewater characterization

The integrated methods, based on a review of different techniques of wastewater characterization, were formulated by Dutch Foundation of Applied Water Research (STOWA) along with their practical applicability (Roeleveld and Kruit, 1998). Roeleveld and van Loosdrecht (2002) discussed experiences with the guidelines including modifications to the original version, based on a physical-chemical method to characterize the sum (S_I+S_S) of the soluble COD fractions, combined with a measuring of the BOD as a function of time for determining the both (S_S, X_S) influent biodegradable fraction (Table 2.7). The wastewater characterization studies at the early stages assumed that the use of a 0.45 µm membrane filter allows to obtain filtrate SCOD sample without the fine colloidal matter (e.g. Stensel, 1992). Originally the STOWA recommended to use such a filter for the division between both COD fractions: soluble and particulate. The analysis at six full-scale WWTPs showed that the concentration of S_S in pre-precipitated samples was reduced up to 29% compared to the traditional membrane filtrate. Similar reduction (up to 25%) was obtained in pretreated samples by flocculation with $Zn(OH)_2$ during further studies. The STOWA guidelines, due to these differences, advised to use flocculation with $Zn(OH)_2$ or a 0.1 µm membrane filter instead of the 0.45 µm (see Section 2.1.3.1). At seven full-scale WWTPs, filtration using 0.1 µm filter and flocculation method generated comparable results with the difference of approximately 1%. Moreover the updated STOWA guidelines presents, that variation in the X_S/X_I ratio is very sensitive to almost every modeled process (e.g. the sludge production) and should be determined indirectly by the measurement of biodegradable COD via the k_{BOD} coefficient. Equations for determining the influent components according to the STOWA guidelines for wastewater characterization can be found in Table 2.9.

In Table 2.10 are summarized wastewater characterization results, obtained with the STOWA guidelines for the COD fractions at 21 Dutch WWTPs. Based on different methods the characterization was performed in order to determine the soluble COD fraction, including: filtration (0.45 µm) of a pre-treated sample with $Zn(OH)_2$, only filtration (0.1 or 0.45 µm) or only flocculation (Roeleveld and van Loosdrecht, 2002). The original STOWA guidelines (Roeleveld and Kruit, 1998) was used by Carrette et al. (2001) to perform the characterization of the settled wastewater containing a high proportion of textile industry (up to 41% of COD). For comparison, the fractionation was also performed with a combination of biological methods, i.e. the method of Orhon et al. (1992) for the inert fractions and respirometric technique of Spanjers and Vanrolleghem (1995) for S_S. The contribution of S_S determined using the STOWA guidelines was extremely high, during the activation of industrial discharge,

accounting for 44% of total COD. Furthermore, this result did not allow for a good calibration of the sludge mass balance. In contrast, the biological methods of the COD fractionation allowed to fit both the observed measurement data and sludge mass balance after a minor adjustments of the X_S/X_I ratio. Temporary stop of the industrial discharge allowed to characterize influent composition with the STOWA guidelines. Finally, the estimated S_S was considerably lower (12%) and the sludge mass balance fitted accurately.

Table 2.9. Equations for determining the influent components according to the STOWA guidelines for wastewater characterization (cited in Brdjanovic et al., 2000)

Particulate components	Soluble components	Additional calculations
$X_S = \alpha \cdot COD_{X,in}$	$S_A = COD_{VFA,in}$	$\alpha = \frac{\frac{BOD_{u,in}}{1-Y_{BOD}} - S_S}{COD_{X,in}}$
$X_I = (1-\alpha) \cdot COD_{X,in}$	$S_I = 0.9 \cdot COD_{f,out}$	$BOD_{u,in} = \frac{BOD_{5,in}}{1-e^{-5 \cdot k_{BOD}}}$
$X_A = 0.1-1.0$	$S_S = COD_{f,in} - S_I$	$COD_{X,in} = COD_{in} - COD_{f,in}$
$X_{PAO} = 0.1-1.0$	$S_F = S_S - S_A$	
$X_H = 0$	$S_{NH} = TKN - \sum_{i=1}(i_{N,Si} \cdot S_i + i_{N,Xi} \cdot X_i)$	
$X_{PHA} = 0$	$S_{PO4} = P_{tot.} - \sum_{i=1}(i_{P,Si} \cdot S_i + i_{P,Xi} \cdot X_i)$	
$X_{PP} = 0$	$S_A = COD_{VFA,in}$	

In a case of simulation studies, it would be useful to convert traditional data available in the records of WWTPs into a form that can be used with the complex biokinetic models (Grady et al., 1999). The GPS-X influent advisor developed by Hydromantis (Canada) allows to perform the complete organic fractionation of the influent wastewater using only a few input parameters, i.e. TSS and COD or BOD_5, and several stoichiometric coefficients. The calculation procedure based on the GPS-X influent advisor was presented by Mąkinia et al. (2002), who introduced minor modification. The authors estimated initially of X_I concentration (instead of X_S originally proposed in GPS-X) as a particulate COD fraction. This approach was made based on the Grady et al. (1999) experimental evidence, which assumed that 35-40% of the particulate organic matter in domestic wastewater is non-biodegradable. Moreover, another assumption of Grady et al. (1999) was adapted to convert biodegradable COD (BCOD) to ultimate BOD (BOD_U).

Table 2.10. Average results of the wastewater characterization performed at 21 different WWTPs in the Netherlands (Roeleveld and van Loosdrecht, 2002)

Fractions / WWTP	S_I	S_S	X_S	X_I	Measurements					
	% of total COD_{in}				COD_{in}	$COD_{f,in}$	$COD_{f,out}$	$BOD_{5,in}$	k_{BOD}	BCOD
Geestmerambacht[1)]	4.89	17.71	27.66	49.75	593	134	32	189	0.35	269
Niedorpen[1)]	8.91	29.26	38.68	23.16	393	150	39	199	0.42	267
Stolpen[1)]	4.47	20.56	43.65	31.32	827	207	41	373	0.35	531
Ursem[1)]	4.81	27.01	34.09	34.09	748	238	40	330	0.38	457
Wieringermeer[1)]	6.08	17.57	33.78	42.57	592	140	40	227	0.42	304
Wieringen[1)]	5.54	30.39	37.87	26.20	668	240	41	330	0.38	457
Deventer[2)]	3.50	12.14	34.17	50.19	799	125	31	257	0.34	370
Nijmegen[2)]	4.66	24.07	39.18	32.09	536	154	28	238	0.35	339
Franeker[2)]	7.91	10.71	47.45	33.94	607	113	53	251	0.36	354
Gouda[2)]	4.68	22.51	30.51	42.30	662	180	34	254	0.38	351
Venlo[3)]	3.73	33.06	13.45	49.92	617	222	25	220	0.47	286
Apeldoorn[4)]	6.41	35.26	9.62	48.72	468	195	33	173	0.70	210
Boxtel[4)]	6.27	27.45	20.59	45.69	510	172	36	181	0.41	244
Nieuwgraaf[4)]	6.36	42.09	14.03	37.52	613	297	43	277	0.59	344
Groote Lucht[4)]	6.49	28.86	22.82	41.83	447	158	32	174	0.43	232
Nieuwe Waterweg[4)]	6.28	20.00	25.12	48.60	430	113	30	148	0.46	194
Haarlem–WP[4)]	6.40	29.57	20.43	43.60	328	118	23	120	0.39	165
Alphen KenZ[5)]	5.27	23.05	26.95	44.73	512	145	30	176	0.33	256
Groesbeek[5)]	6.31	35.75	24.77	33.18	428	180	30	180	0.34	259
Hardenberg[5)]	6.13	33.33	19.82	40.72	555	219	38	217	0.40	295
Papendrecht[5)]	10.4	42.32	19.09	28.22	241	127	28	119	0.60	147
Average value	6	27	28	39	551	173	35	221	0.42	301

Note:

1) Average values of 24-h composite samples from five days; soluble analysis after filtration (0.45 µm) of pre-treated sample with $Zn(OH)_2$;
2) Average values of 24-h composite samples from seven days; soluble analysis after filtration (0.45 µm) of pre-precipitated wastewater;
3) Grab sample; soluble analysis after filtration (0.1 µm);
4) Average values of six grab samples; soluble analysis after flocculation;
5) Average values of 24-h composite samples from 4 two-days measuring campaigns; soluble analysis after filtration (0.45 µm).

2.2. Hydrolysis of slowly biodegradable substrate

The particulate organic fraction mostly consist of X_S that have to be hydrolyzed prior it can be taken up and be degraded by bacteria (Levine et al., 1985). That is a general consensus of the necessity to differentiate between both biodegradable substrates (S_S and X_S) in municipal wastewater (Dold et al. 1980, Henze et al. 1987, Orhon and Artan 1994). Immediately, after bringing biomass into contact with wastewater in a batch reactor, the activity in the reactor will be dominated by growth of heterotrophs on S_S, whereas the subsequent activity will be predominantly, due to use of substrate released from hydrolysis of X_S (Ekama et al., 1986).

Takahashi et al. (1969) studied the hydrolysis of various fractions of suspended solids and demonstrated that soluble substances were produced during hydrolysis. The extent to which the particles are removed in primary treatment, are utilized by bacteria under anaerobic, anoxic or aerobic conditions. There are also enmeshed in the sludge and removed with the WAS depends on treatment plant design and operation. Particulate organic matter that is hydrolyzed and used under anaerobic or anoxic conditions is a valuable carbon source for biological nutrient removal — a process that in many cases is limited by the availability of readily biodegradable organic matter (Henze, 1992). In addition, particulate organic matter oxidized under aerobic conditions is not beneficial and increases the overall energy demand for treatment (van Loosdrecht et al., 1997a). Those particles that are not hydrolyzed within the system increase the sludge concentration in the reactors and the production of WAS — both are associated with the increasing costs.

Nutrient removal are in many cases limited by the extent and kinetics of hydrolysis processes and particulate organic matter can be important for the selection of specific bacterial populations (Frigon et al., 2001). In the design of reactors in BNR plants, the hydrolysis rates determine the conversion of X_S into S_S that can serve as a carbon source for denitrification or EBPR. Particulate organic matter attaches rapidly enmeshed in the sludge flocs and then is either degraded or removed with the WAS. In both cases, the treatment objective of removing the organic matter from the effluent is achieved. The particulate organic fraction and associated hydrolysis rates has a direct impact on the volume of bioreactors in BNR WWTPs (Morgenroth et al., 2002). The following Section contains an overview over mechanisms involved in hydrolysis, mathematical models to describe hydrolysis, and experimental tools to determine model parameters.

2.2.1. Definition and types of hydrolysis

Hydrolysis is an important process in biological wastewater treatment and has been known as the rate limiting step in organic carbon removal from municipal or industrial wastewater. The strict definition of hydrolysis is "the breakdown of a polymer into smaller units by addition of water" (Brock and Madigan, 1991). In wastewater treatment applications, the process of hydrolysis summarizes all mechanisms and complicated reactions inside the microorganism flocs by means of extracellular enzymes that make X_S available for bacterial growth (Gujer et al., 1999) Simply, hydrolysis is usually understood as a conversion of X_S into S_S form (Henze et al., 1987).

With this broader definition of hydrolysis, other processes such as chemical dissolution and mass transport processes have to be also considered, when evaluating hydrolysis rates (Morgenroth et al., 2002). The utilization of X_S is defined by means of a surface-limited reaction kinetics (Dold et al., 1980; Henze et al., 1987), but not only (see Table 2.14). On the other hand, considering the effluent quality, hydrolysis mechanism also plays a dominant role in delicate balance of electron donor/electron acceptor ratios in BNR activated sludge systems as an important carbon source. In addition, the WAS production is also affected by the nature of X_S (Insel et al., 2005).

Two types of hydrolysis processes can be differentiated (Morgenroth et al., 2002):

- hydrolysis of primary substrate where organic substrate present in the original wastewater is broken down;
- hydrolysis of secondary substrate that refers to the breakdown of substrate that has been produced by the bacteria e.g. hydrolysis of internal storage products, substances released by the bacteria during the normal metabolism, or particles produced during decay of bacteria (Bryers and Mason, 1987, van Loosdrecht and Henze, 1999).

In ASM1 (Henze et al., 1987) and ASM3 (Gujer et al., 1999) the term “hydrolysis” is used differently. In case of the ASM1, the process of hydrolysis combines primary and secondary hydrolysis (Henze et al., 1987). The death-regeneration concept was utilized to describe the reactions that occurred during the "endogenous" or "death phase" of microbial growth: hydrolysis (enzymatic degradation of degradable cell protoplasm), growth (oxidation of the decay products and synthesis of new organisms) and decay (lysis of the cell membrane) (Figure 2.10 a). This approach has a distinct advantage over "endogenous respiration" in modeling systems incorporating aerobic, anoxic and anaerobic conditions (Dold et al., 1980). The decay of a portion of alive organisms rise to the formation of X_I and X_S. The generated X_S originating from the influent are transformed by hydrolysis to S_S. Compared to ASM3, the degradation of external substrate and the degradation of cellular products is decoupled and the process of hydrolysis refers to primary hydrolysis only (Gujer et al., 1999) (Figure 2.10 b).

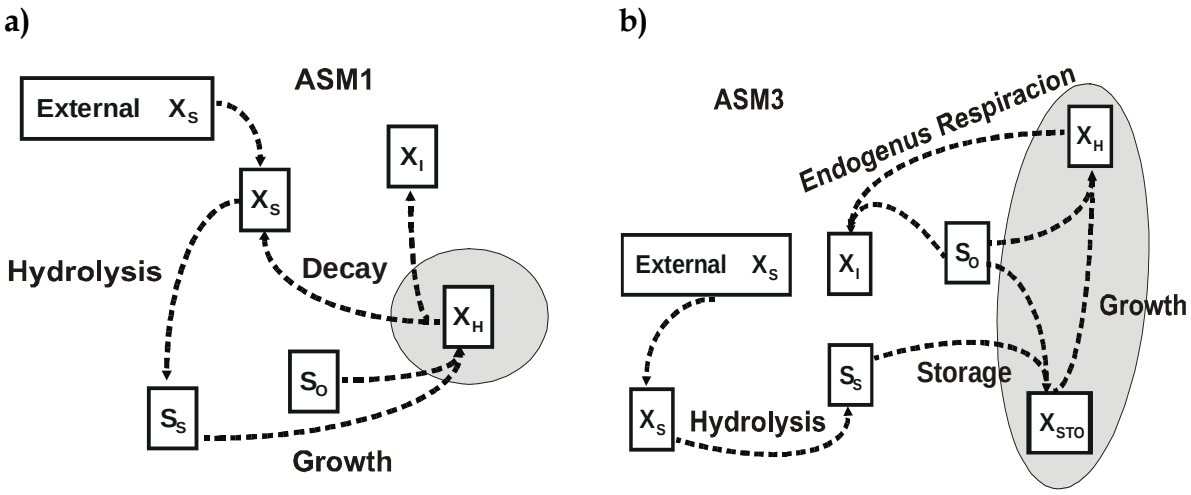


Figure 2.10. Comparison of the hydrolysis concept (a) ASM1 – combines primary and secondary hydrolysis (b) ASM3 – primary hydrolysis only. The shaded area marks the components considered as part of the bacterial cell (Gujer et al., 1999)

Hydrolysis of secondary substrate refers to the breakdown of particles that has been produced during decay of bacteria was called "decay" in ASM1 (Henze et al., 1987) or "lysis" in ASM2 (Henze et al., 1995) and ASM2d (Henze et al., 1999), although both processes are equivalent in terms of the mathematical description. Under the term "decay", van Loosdrecht and Henze (1999) classified all processes that reduce the number of microorganisms and/or the weight and specific activity of biomass. The authors assumed that the decay can be caused by cell external factors, e.g. predation (external decay) or mechanisms inside the microbial cell, such as death and self-oxidation of cell components (internal decay), whereas the term "lysis" can refer to solubilization of biomass, causing the release of secondary substrates into the liquid phase. The concept of lysis was introduced to reflect more complex transformations of the PAO cell components (biomass, polyphosphate and PHA) in contrast to the single decay process of heterotrophic and autotrophic biomass included in ASM1. The process of decay (or lysis) is followed by growth (so called "cryptic growth") on the secondary substrate generated during the cell decay/lysis. Several studies have suggested that hydrolysis of secondary substrate such as the formation of internal storage products polyphosphates, glycogen and poly-β-hydroxyalkanoates inside cells, may be more significant than microbial kinetics (Kohno et al., 1991; Majone et al., 1996; van Loosdrecht et al., 1997a; Goel et al., 1998c; Krishna and van Loosdrecht, 1999).

2.2.2. Mechanisms of hydrolysis

Hydrolysis of particulate organic matter, acording to Morgenroth et al. (2002), seems to be as diverse as the particles and organisms that are involved in the process. Evaluating mechanisms for hydrolysis or even kinetics using particles extracted from real wastewater is complicated, because of different sizes range and types of organic matter (see Section 2.1.3) or significant amounts of heterotrophic biomass (such as bacteria and also higher organisms) involved in the overall process (Henze, 1992; Morgenroth et al., 2002). Therefore, a mathematical model has to rely on simplified mechanisms for hydrolysis. The most important aspects of these mechanisms, such as direct degradation of particles by protozoa and enzymatic hydrolysis, were presented by Morgenroth et al. (2002).

2.2.2.1. Direct degradation of particles by protozoa

Acording to Alberts et al. (1994) protozoa can directly take up particles in the micrometre range and degrade them intracellulary through phagocytosis. Thus, for the majority of particles in wastewater protozoa can metabolize them without any prior extracellular hydrolysis step. The significance of protozoa on the overall conversion of particulate organic matter in a wastewater treatment system are still not well determined. In the common opinion, a large fraction of the organic particles during entering to a WWTP is captured by protozoa. It could be possible that most captured particles are not completely metabolized and protozoa may release organic material to futher degradation. However, the protozoa belong to aerobic organisms and their activity would be significantly reduced during anoxic and anaerobic conditions. In this context, protozoan activity could have possible effect for measured changes of electron acceptor on overall hydrolysis in activated sludge systems (Henze and Mladenovski, 1991).

2.2.2.2. Enzymatic hydrolysis

Traditionally, degradation of slowly biodegradable matter in wastewater systems has been related to the hydrolysis process, due to the fact that most depolymerisation reactions are catalysed by this process and only a few through lyase reactions (Morgenroth et al., 2002). Depolymerisation is generally carried out by extracellular hydrolases and/or lyases. According to the Enzyme Handbook (Schomburg et al., 1997) from all 197 extracellular enzymes that have been identified, only 11 are lyases, while most of them (145) are hydrolytic. Hydrolytic cleavage is characterised by the addition of a water molecule (hydroxyl and proton), while lyase forms an unsaturated and protonated end products.

Two mechanisms of depolymerisation can be differentiated, which are mediated by different types of enzymes: exo- and endo- enzymes. The exo-enzymes act on a specific bond above the end (normally the non-reducing end), while the endo-enzymes act randomly on internal polymer bonds. Therefore, degradation of polymers normally involves a range of endo/exo reactions. As an example, the most important reactions involved in the degradation of the polysaccharide Dextran are shown in Figure 2.11 (Morgenroth et al., 2002).

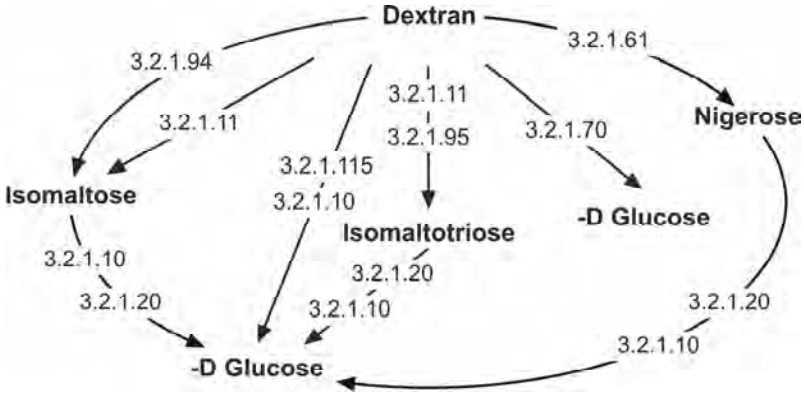


Figure 2.11. Major routes for enzymatic hydrolysis of Dextran. Each process is indicated by the enzyme involved (EC No) and ends by hydrolysis product (Morgenroth et al., 2002)

The degradation of organic particles and polymers by extracellular hydrolysis depends on several other factors. As it was mentioned above (see Section 2.2.2.1), the cell wall disables most bacteria from uptake and degradation of particles and aqueous polymers by phagocytosis. Thus, the degradation depends on the extracellular depolymerisation, followed by cellular uptake and subsequent metabolisation (Chrost, 1991). Microorganisms benefit from the soluble products and produce the corresponding hydrolytic enzymes. Contrariwise to bacterial hydrolysis, protozoa competing for particulate organic substrate (Ratsak et al., 1996). Some eukaryotes, such as fungi and yeast, excrete depolymerisation enzymes, which take only one special substrate without any additional cofactors or prosthetic groups. The enzymatic degradation converts particulate carbohydrates, proteins and lipids into monosaccharides, amino acids, VFAs or glycerol, but the local concentration of enzymes, location of the enzymes and product/intermediate transport mechanisms all influence the rate of reaction (Morgenroth et al., 2002; Vavilin et al., 2008). Most of extracellular enzymes reported in enzyme databases (such as BRENDA) have been studied "in vitro".

2.2.3. Experimental approaches to evaluate the influence of hydrolysis

Morgenroth et al. (2002) provided an overview of experiments that have been designed to evaluate mechanisms or location of hydrolysis and to quantify the influence of hydrolysis on substrate utilization. Four experimental approaches can be differentiated, according to measured parameters:

- measurement of specific hydrolytic enzymes;
- measurement of specific hydrolytic intermediates or specific end products;
- overall mass balances for bulk organic parameters;
- measurement of respiration rates to quantify bacterial activity.

The first two approaches allow to study specific mechanisms involved in hydrolysis but are often restricted to specific substrates (e.g. starch), while two last approaches allow to evaluate the overall processes, but may not allow to study specific mechanisms involved in the hydrolysis process (Morgenroth et al., 2002). The most common, from presented above experimental approaches, used for example to mathematical modeling of activated sludge systems is a measurement of bacterial respiration. Dynamic experiments measuring respiration rates, as it was mentioned before, were the basis for introducing two biodegradable X_S and S_S organic fractions (Ekama and Marais, 1979; Dold et al., 1980). However, the model parameters for hydrolysis (stoichiometry and kinetics) and the component concentration of X_S cannot be easily estimated, based on the respiration rate experiments, because this fraction in wastewater is composed of a heterogeneous mixture of colloids and particles with variable chemical composition (Spanjers and Vanrolleghem, 1995; Sophonsiri and Morgenroth, 2004). Hence, other research has aimed at evaluating specific mechanisms of hydrolysis using substrates such as starch (Larsen and Harremoes, 1994), dextran (Haldane and Logan, 1994; Confer and Logan, 1997b), dextrin (Confer and Logan, 1997b), bovine serum albumin (BSA) (Confer and Logan, 1997a), and fats (Sprouse and Rittmann, 1990). These commercially available substrates are well defined, but are only representative of macromolecular or colloidal X_S. Therefore, during last decades only a limited number of experiments measuring respiration rates in "real" wastewater has been carried out to determine the amount of X_S and evaluate of the hydrolysis rate coefficients (Henze and Mladenovski, 1991; Orhon et al., 1998).

Henze et al., 1987 proposed to measure or estimate all other COD fractions and then determine the X_S, based on a mass balance. Kappeler and Gujer (1992) proposed curve fitting of the concentration of X_S and the first order hydrolysis rate constant, which was assumed to be comparable to hydrolysis kinetics in ASM1 only for low concentrations of X_S. Spanjers and Vanrolleghem (1995) also estimated the

concentration of X_S from respiration rate measurements. However, they divided the X_S into two fractions. The curve fitting allowed to estimate only the S_H, for which the process kinetics was described by the first order expression (Table 2.15, Model No I). Vollertsen and Hvitved-Jacobsen (1999) divided the X_S into three fractions: slowly, intermediate and rapidly biodegradable organic matter that are hydrolyzed in parallel (Table 2.14, Model No 4). Subsequently, they used batch experiments with respiration rate measurements to quantify all biodegradable organic matter fractions and hydrolysis rates for the three parallel hydrolysis processes.

For the estimation of parameters describing the surface saturation type hydrolysis, Ekama et al. (1986) proposed to use OUR measurements in a completely mixed reactor fed continuously under a daily cyclic square wave feeding pattern. In the second phase, after an immediate depletion of the S_S, the accumulated X_S continues to be used at the same rate for a time period. For the batch experiments, the authors recommended that the value of k_{hyd} could be estimated at a high X_S/X_H ratio or performing simulations for curve fitting of OUR profiles. Sollfrank and Gujer (1991) determined the first order hydrolysis rate constant from the slope of the OUR_H plot vs. the concentration of degradable matter in a batch experiment. For the time $t>t_1$ (t_1 denotes the time when a nearly linear relation is reached), it was assumed that hydrolysis was the limiting process in the degradation of filtered wastewater. The value of k_{hyd} was determined from the slope after the net respiration rate becomes proportional to the concentration of biodegradable matter by the following equation:

$$OUR_{Hnet}\ (t>t_1) = (1 - Y_H) * k_{hyd} * X_S(t) \qquad \textbf{(2.16)}$$

where:

k_{hyd} – *specific hydrolysis rate constant,* T^{-1}
OUR_{Hnet} – *net heterotrophic oxygen uptake rate,* $M(O_2)L^{-3}T^{-1}$
X_S – *slowly biodegradable substrates,* $M(COD)L^{-3}$
Y_H – *"true" growth yield coefficient for heterotrophic organisms,* $M(COD)M(COD)^{-1}$

In general, the accepted procedure for the experimental assessment of k_{hyd} and K_X under aerobic conditions involves model-based evaluation and curve fitting of OUR profiles in laboratory scale batch or semi-continuous reactors using wastewater containing total COD fractions and heterotrophic biomass (Insel et al., 2003). Experimental results are then evaluated using a mathematical model to determine both the COD fractions in the wastewater and hydrolysis kinetics (Sollfrank and Gujer, 1991; Insel et al., 2002). Such an experimental approachs have been extensively discussed in literature. Insel et al. (2002 and 2003) demonstrated that the model-based evaluation and curve fitting allows to generate not a unique set, but a relatively large combinations of different pairs of k_{hyd} and K_X coefficients equally

applicable to the experimental data. Mogenroth et al. (2002) concluded that there is a large uncertainty dealing with model parameters estimated from respirograms.

The basic assumption of those experiments using respiration rate measurements to quantify hydrolysis is that hydrolysis determines respiration rates, when the readily biodegradable substrate is depleted. However, in the presence of storage of internal polymers the interpretation of OUR profiles (see Section 2.1.3) becomes confusing and extracellular hydrolysis of slowly biodegradable COD and intracellular degradation of storage polymers cannot be distinguished from the OUR profile (Goel et al., 1998c). In the fallowing study, Goel et al. (1999) suggested an experimental approach to separate hydrolysis from storage by performing and analyzing two parallel OUR measurements: one with filtered wastewater (including soluble COD) and the other with non-filtered wastewater (including total COD). On the other hand, batch respirometric tests were proposed by adjusting the appropriate wastewater/biomass mixtures with different (either high or low) initial ratios between wastewater and biomass (S_T/X_V) (Orhon et al., 1999; Sperandio and Paul, 2000). They estimated wastewater composition and the hydrolysis rate based on simultaneous parameter estimation from two batch experiments. The substrate observed in short-term respirometric experiments was classified as a "readily hydrolysable" COD fraction and could be modeled by a first order reaction. The hydrolysis of "slowly hydrolysable" COD fraction was correctly modeled with a sequential two-step process (adsorption followed by surface-limited hydrolysis).

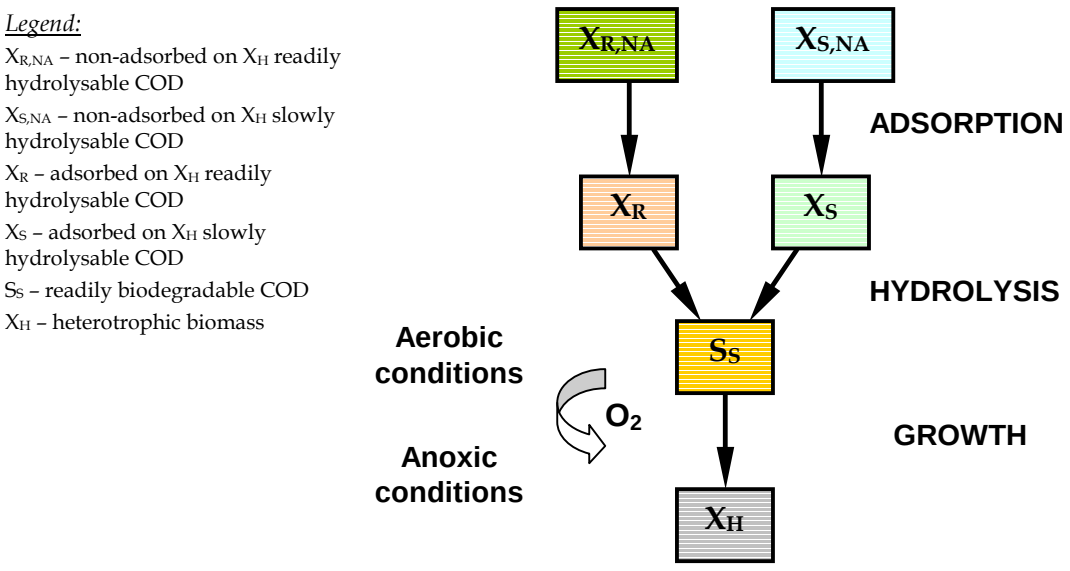


Figure 2.12. Diagram of the mechanisms of COD degradation according to the model of Lagarde et al. (2005)

Lagarde et al. (2005) used analysis of the experimental data of not settled samples, relied on a three substrate model derived from Sollfrank and Gujer (1991) and Spanjers and Vanrolleghem (1995), with an additional distinction between adsorbed and free substrates according to Sperandio and Paul (2000). In this model, as illustrates Figure 2.12, the fractions were: readily (S_S) biodegradable COD; readily (X_R) hydrolysable COD (adsorbed on biomass); slowly (X_S) hydrolysable COD (adsorbed on biomass); non-adsorbed ($X_{R,NA}$ and $X_{S,NA}$) COD that were readily and slowly hydrolysable, respectively, and two other variables: dissolved oxygen (O_2) and heterotrophic (X_H) biomass concentrations.

Henze et al. (1987) used that model as a reference for the respirograms simulations in ASM1. The initial COD was considered as not adsorbed ($X_{R,NA}$ and $X_{S,NA}$) or under S_S, i.e. fractions X_R and X_S are initially equal to zero. Both substrates $X_{R,NA}$ and $X_{S,NA}$ progressively adsorb on the biomass and only the resulting fractions, X_R and X_S, respectively, can be hydrolysed, assuming the mediation of bound exoenzymes. For this fraction, the hydrolysis rate constants, $k_{hyd,X,}$ and entrapment saturation coefficients, $K_{X,X}$, ranged 0.25-1.05 d^{-1} and 0.33-0.95 g COD/g COD, respectively. In the study of Orhon et al. (1999), the results for municipal wastewater yielded average $k_{hyd,S}$ and $K_{X,S}$ values of 3.1 d^{-1} and 0.2 g COD/g COD, respectively, associated with the hydrolysis of S_H and significantly lower values of $k_{hyd,X}$ = 1.2 d^{-1} and $K_{X,X}$ = 0.5 g COD/g COD associated with hydrolysis of X_S. It was observed, however, that the discrepancy between measured and predicted OURs was reduced considerably by shifting from a single hydrolysis model to a dual hydrolysis model. It should be noted, that the hydrolysis rate (especially of nitrogenous compounds) depends on electron-acceptor conditions. For example, Henze and Mladenovski (1991) found a significant reduction of hydrolysis rates under anoxic and anaerobic conditions compared to aerobic conditions. The rate at 20°C is high under aerobic conditions (0.12 d^{-1}) medium under anaerobic conditions (0.06 d^{-1}) and low under anoxic conditions (0.03 d^{-1}). The ratio between the hydrolysis rates under aerobic and anoxic conditions are very similar to the respiration rates measured as electron equivalents. This could indicate that the biomass activity has significant impact upon the hydrolysis rate. However, there is no direct explanation for the high rate under anaerobic conditions, but it could be caused by fermentation processes performed by the heterotrophic microorganisms (Henze and Mladenovski, 1991). On the other hand, indirect evidence suggests that the rate under anaerobic conditions should not be high. If the anaerobic hydrolysis rate was significant, it would make a substantial contribution to EBPR, which is contrary to experimental observations strongly relating EBPR to the S_S in influent (Ekama and Wentzel, 1999a). A subject of ongoing

debate, according to Morgenroth et al. (2002), has been reduced to whether and how hydrolysis rates are influenced by electron acceptor conditions.

The basic assumption of experiments using respiration rate measurements to quantify hydrolysis is that hydrolysis is the rate limiting step that determines respiration rates (Dold et al., 1980; Arvin and Harremoes, 1990). Experimental evidence would appear to indicate that hydrolysis may be the rate controlling step even at 20°C. In order to check this hypothesis Tian et al. (1993) investigated the effect of temperature on sludge accumulation. The parallel reactors were run at a sludge age of 10 days under identical conditions with the exception of temperature. The reactor, which run at 12°C produced 12-20% more sludge than the corresponding reactor run at 20°C. The authors reasoned that the excess sludge accumulation resulted from hydrolysis being appreciably slower than synthesis or decay. Overall, respirometry is a valuable tool to estimate wastewater composition and reaction rates, however, there is a large uncertainty associated with hydrolysis parameters extracted from parameter estimation (Vanrolleghem et al., 1999). Differences between hydrolysis in traditional activated sludge systems and other treatment technologies (membrane activated sludge system, granular sludge system, biofilm reactors) should be also evaluated (Henze et al., 1995; Barker and Dold, 1997a; Ekama and Wentzel, 1999a, Morgenroth et al., 2002).

2.2.4. Mathematical modeling of hydrolysis

Municipal wastewater is composed of a complex mixture of organic substrates, which have to be described separately in order to allow an adequate model under dynamic conditions. The distinction between these fractions was made not only on physical separation, but also on biological response for use it to mathematical modeling purpose to optimize overall WWTP operation (Ekama et al., 1986). Commonly used IWA Task Group Activated Sludge Models (Henze et al., 1987 and 1995; Gujer et al., 1999) all consider biodegradable organic compounds as separate fractions, where e.g. X_S is hydrolyzed using a surface limited process (see Section 2.2.4.1). The overall process of hydrolysis is described by combining of both models stoichiometry and kinetics. Morgenroth et al. (2002) noted that modeling would have to rely on simplified process mechanisms and pseudo parameters, because it is questionable how stoichiometric and kinetic coefficients could be represented in solving "practical problems" reflecting e.g. real conditions at a WWTP. Hence, selection of a model structure should be based on the necessary detail for the specific simulation and the availability of data for model calibration (Morgenroth et al., 2002). In the following Section, development of complex hydrolysis activated sludge models and the kinetic expressions associated with model stoichiometry for

hydrolysis process are discussed. Finally, the comparison between single and dual hydrolysis models are presented.

2.2.4.1. Development of complex hydrolysis activated sludge models

Many researchers, for more then three decades, proposed a wide range of models of biological treatment by activated sludge method. The first important studies with respect nitrification were published even earlier by Downing et al., (1964) and Knowles et al., (1965). However, an important step in the modeling of activated sludge, as well as optimal design, operation and control of WWTP, was new approach to wastewater characterization (see Section 2.1.2). During last decades, several attempts, to describe major wastewater components in mathematical modeling of activated sludge systems, have been presented as biomass, readily-, slowly- and non-biodegradable COD (Dold et al., 1980; Henze et al., 1987; Gujer et al., 1995; Gujer et al., 1999). The International Association on Water Quality (IAWQ, formerly IAWPRC) formed a task group in 1983 to promote development of practical models for design and operation of biological wastewater treatment systems. The first goal of this task group was to review existing models and the second was to reach a consensus concerning all-important mathematical models (Jeppsson, 1996). In IAWQ models, the main parameters associated with removal rates are presented in Table 2.11.

Table 2.11. Main parameters in IAWQ models associated with removal rates (Henze et al., 1987)

Parameter	Fraction name	Residence time	Removal mechanisms	Removal rates
S_I	Inert soluble	HRT	None	-
S_S	Readily biodegradable	HRT	Oxidation, growth	$\frac{\mu \cdot X_H \cdot S_S}{Y_H \cdot (S_S + K_S)}$
X_I	Inert particulate	SRT	Settling	-
X_S	Slowly biodegradable	SRT	Hydrolysis, settling	$k_{hyd} X_H \frac{X_S / X_H}{K_X + X_S / X_H}$
X_H	Heterotrophic biomass	SRT	Decay, settling	$\mu \cdot X_H \frac{S_S}{S_S + K_S} - b \cdot X_H$

In 1987, the mathematical modelling of activated sludge processes was introduced as the Activated Sludge Model No 1 called ASM1. This model was developed to simulate organic carbon removal, nitrification and denitrification processes with simultaneous consumption of oxygen or nitrate as electron acceptors. The ASM1 may also be used to predict a sludge production in activated sludge systems treating municipal wastewaters. Dissemination in last decades at WWTP with EBPR led to further development of the mathematical modeling. In 1995, the IAWQ Task Group

presented ASM2, which has been extended in order to include a description of biological phosphorus removal. Besides biological processes, ASM2 encompassed modelling of chemical phosphorus precipitation (Henze et al., 1995). Finally in 1999, the IAWQ Task Group presented ASM2d (see Appendix 1), which was an extantion of ASM2 in part of accumulating phosphorus organisms (PAOs). The prime innovation of the model resulted from the introduction all different types of microorganisms capable of accumulating phosphorus in the form of polyphosphates (Henze et al., 1999). Prior to the publication of ASM2 (Henze et al., 1995) and ASM2d (Henze et al., 1999), researchers developed own models, which combined the ideas formulated in ASM1 and by Wentzel et al. (1989) in "first" general model for biological nutrient removal. The ASM1 and later ASM2/2d components are divided into particulate and soluble components, Barker and Dold (1997b) following Wentzel et al. (1989), defined biomass components and substrate concentrations.

The use of ASM1, in research and practice, revealed many defects, which were corrected in a new model termed ASM3. This model consisted of the same biological processes as ASM1, and the major difference was in the hypothesis of COD transformation (see Figure 2.10). It was assumed that heterotrophic biomass grow on Ss is indirect. This fraction of COD is first taken up and stored before utilization by bacterial growth. The storage concept was based on the recognition that in activated sludge could be observed different oxygen consumption rates. The rapid rate associated with the utilization of Ss, slow rate associated with the utilization of Xs and the slowest endogenous oxygen uptake rate. Moreover, in ASM3 the biomass decay process has been replaced by endogenous respiration, which eliminates growth-decay-growth cycling in the COD (Henze et al., 2000a). The recent developments of activated sludge models, mainly focus on the family of activated sludge models, presented by IAWQ Task Group (nowadays IWA), and the metabolic model developed at the Delft University of Technology (TUDP model). Table 2.12 summarises essential features of the most popular activated sludge models.

At beginning of the 1990's the activated sludge models have been implemented in the computer software as simulator environments or simulation platforms. In recent years dynamic models, including commercial software, have increasingly been used for optimization of activated sludge systems, process control (Coen et al., 1997; Hvala et al., 2002) or as a decision and detection tool (Mąkinia et al., 2005). The most popular commercial simulator environments include AQUASIM (Switzerland), BioWin (Canada), EFOR (Denmark), GPS-X (Canada), SIMBA (Germany), STOAT (United Kingdom) and WEST (Belgium) (Mąkinia, 2006). In the literature was reported that mathematical modeling and computer simulation are a valuable tool

for searching optimal operating conditions for full-scale applications, e.g. Hulsbeek et al. (2002) mentioned over 100 Dutch WWTPs examined by dynamic simulation. Furthermore, practical applications of the mathematical modeling can be qualified to optimization of the performance and upgrade of existing plants as well as design or development of new treatment concepts and facilities (Andrews, 1992; Orhon and Artan, 1994). An overview of practical experiences with full-scale WWTP model calibration is presented in Table 2.13.

Table 2.12. Overview of the most popular activated sludge models

MODEL References	Nitr./ Denitr.	Heterotrophic/ Autotrophic decay	Hydrolysis	BioP/ Den. PAOs	Lysis of PAO/PHA	Fermentation/ Chemical P removal	Reactions	State variables
ASM1 Henze et al. (1987)	+/+	DR, Cst	EA	-/-	-	-/-	8	13
ASM2 Henze et al. (1995)	+/+	DR, Cst	Cst	+/-	Cst	+/+	19	19
ASM2d Henze et al. (1999)	+/+	DR, Cst	Cst	+/+	Cst	+/+	21	19
B&D Baker & Dold (1997)	+/+	DR, Cst	Cst	+/+	EA	+/-	36	19
ASM3 Gujer et al. (1999)	+/+	ER,EA	Cst	-/-	-	-/-	12	13
TUDP Brdjanovic et al. (2000)	+/+	DR, Cst	Cst	+/+	EA	+/-	21	17
ASM3-bioP Rieger et al. (2001)	+/+	ER,EA	EA	+/+	EA	-/-	23	17

Note: Den. PAO – denitrifying PAO activity included in the model; DR – death regeneration concept; EA – dependent on electron acceptor; ER – endogenous respiration concept; Cst – independent on electron acceptor; Nitr./Denitr. – nitrification /denitrification; +/- process concidered/not concidered

The concept of hydrolysis process, in complex models, may vary. In ASM1 the hydrolysis process (termed as conversion X_S to S_S) is modelled on the basis of surface reaction kinetics $(X_S/X_H)/(K_x+X_S/X_H)$ and occurs only under aerobic and anoxic conditions. The rate of hydrolysis is reduced under anoxic conditions compared with aerobic conditions by a factor $\eta_{NO3} < 1$ (see Section 2.2.4.3). The rate is also first-order with respect to the heterotrophic biomass present, but saturates as the amount of entrapped substrate becomes large in proportion to the biomass. The hydrolysis of particulate, biodegradable organic nitrogen is included as a separate process in ASM1 (not in other ASMs) at a rate defined by the hydrolysis reaction for entrapped organic matter described above. This process is necessary if the nitrogen content of X_S is variable. For comparation in order to simplify ASM2/2d, it is assumed that X_S contains a constant fraction of nitrogen (i_{NXs}) and phosphorus (i_{PXs}). Without this

simplifying assumption, six more hydrolysis processes and two more particulate components would be required. Moreover, the proposed rate equations for the hydrolysis processes at both ASM2/2d are similar to those of ASM1: hyperbolic switching functions for S_{O_2} and S_{NO_3} consider the environmental conditions; a surface-limited reaction $(X_S/X_H)/(K_x+X_S/X_H)$ is assumed for the hydrolysis process itself. Furthermore, it is proposed that only heterotrophic organisms may catalyse hydrolysis. Typically hydrolysis at both ASM2/2d is slower under denitrifying or anaerobic (fermentation) than under aerobic conditions. The rate for anaerobic and anoxic hydrolysis is therefore reduced by the factors η_{fe} and $\eta_{NO3,}$ respectively (see Section 2.2.4.3).

Table 2.13. Overview of papers presenting detailed practical experiences with full-scale WWTP model calibration (Gernaey et al., 2004)

Reference	Model	Data			Phase		Remarks
		Batch exp.	Static	Dynamic	Calibration	Validation	
Coen et al. (1996)	**ASM1**	+	+	+	D	S	Dynamic influent profile was designed based on the measurement campaign
Cinar et al. (1998)	**ASM2**	-	+	-	S	S	-
van Veldhuizen et al. (1999a)	**TUDP**	-	+	+	S	D	-
Brdjanovic et al. (2000)	**TUDP**	+	+	+	S	S	Use of batch experiments mainly during model unfalsification phase
Koch et al. (2000)	**ASM3**	+	+	+	D	D	Use of batch experiments before the full-scale model calibration phase
Meijer et al. (2001)	**TUDP**	-	+	+	S	-	-
Rieger et al. (2001)	**ASM3P**	+	+	+	S/D	S/D	Use of batch experiments during calibration; iterative calibration procedure
Hulsbeek et al. (2002)	**ASM1/ general**	-	+	+	S/D	S/D	Type of data depending on model purpose
Petersen et al. (2002)	**ASM1/ general**	+	+	+	S/D	S	Type of data depending on model purpose; use of batch experiments for calibration (wastewater characterization) and unfalsification

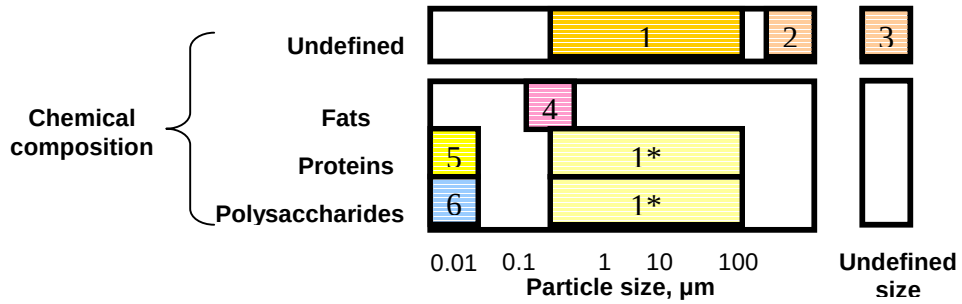
Note: D – dynamic, S – steady state

In ASM3, hydrolysis similar to ASM1 is assumed to be active independently of electron donor on the contrary to other complex models. The importance of hydrolysis has been reduced for the rates of oxygen consumption and denitrification as compared with ASM1. Degradation of soluble and particulate organic nitrogen in ASM3 have been integrated into the hydrolysis decay and growth process. Moreover, Gujer et al. (1999) proved that there was no difference between hydrolysis rates

under aerobic or anoxic conditions and introduced this modification in ASM3 by "eliminating" reduction factor. Rieger et al. (2001) extended ASM3 with a bio-P module. In comparison with ASM2d, there are several differences in the new model structure of ASM3P. The main distinction can be summarized that the hydrolysis rate under anaerobic and anoxic conditions is not reduced in comparison with aerobic conditions and process accomplished only by "ordinary" heterotrophs. Contrary to ASM3P the TUDP models hydrolysis are similar to ASM2d. The hydrolysis rate in the latest modification of TUD model by Meijer (2004) is a function of the heterotrophic biomass, being the total of both "ordinary" heterotrophs and PAOs. This adaptation was made in accordance with the observation of Goel et al. (1998c and 1999) that the hydrolysis rate is associated with the total (active) biomass and not solely with X_H. However hydrolysis was identified as a less reliable model process. Finally, New General models are aerobic, anoxic and anaerobic hydrolysis accomplished only by "ordinary" heterotrophs. The rate is a surface−limited reaction (X_S/X_H) / (K_H+X_S/X_H) and under anoxic and anaerobic conditions is reduced by $\eta_{S,ANOX}$ and $\eta_{S,ANA}$. In addition, two hydrolysis efficiency factors, E_{ANOX} and E_{ANA}, are included to allow for the possibility of COD loss.

In mathematical modeling, the process of hydrolysis must be adequately described in order to predict spatial and temporal availability of organic substrate for nutrient removal (such as denitrification and EBPR) processes. Currently, available models of hydrolysis may well predict for typical applications in wastewater treatment. However, for new treatment processes, or if the composition of the wastewater is not typical, current modeling approaches may not be applicable any more (Morgenroth et al., 2002). At high organic loading inhibition of hydrolysis should be considered, when analysis of the effciency of anaerobic digestion of complex substrates is carried out. The first-order kinetics has traditionally been used to describe the hydrolysis process in anaerobic digestion, but it may be inaccurate to describe the hydrolysis of certain complex substrates. In such cases, methanogenesis or acetogenesis can be the rate-limiting steps in anaerobic digestion. To describe these phenomena numerically, a more complex structured model should be used, but nowadays it is still not possible to adopt a general model applicable under all circumstances (Vavilin et al., 2008). This follows from the fact that many mechanisms of hydrolysis in wastewater treatment applications are not well understood. Moreover, these mechanisms as well as hydrolysis rates and locations depend on type/concentration of X_S substrate and microbial population (Goel et al., 1998a). Caution should be employed, when applying results from experiments with a simple substrate (e.g. starch) and/or a pure culture to wastewater treatment applications. Experimental investigations often use

well defined substrates that allow to trace intermediates and enzyme activity. Many studies have used starch (as a model compound that can be easily quantified) because the enzymes involved in hydrolysis are known. Results of experiments using starch, conducted with pure cultures (Mino et al. 1995) and activated sludge (Goel et al. 1998c) provided the argument that a bulk-reaction kinetics, similar to that proposed by Monod, may be applicable for the hydrolysis process (see Table 2.15). Moreover, Goel et al., 1998c, showed that even though hydrolysis of starch was limiting the overall reaction rate, because the accumulation of glycogen as a storage product was observed. Thus, it was concluded that the respiration rate cannot directly be linked to substrate hydrolysis.



where:

*1 – Filtered wastewater; *note: proteins and polysaccharides were measured (Guellil et al., 2001)*
2 – Filtered, settled or centrifuged wastewater or sewer solids (Eliosov and Argaman, 1995; Janning et al., 1998; Vollertsen and Hvitved-Jacobsen, 1999)
3 – Raw wastewater (Henze and Mladenovski, 1991)
4 – Fats (Sprouse and Rittmann, 1990)
5 – Bovine Serum Albumin (Ubukata, 1992; Confer and Logan, 1997a)
6 – Starch, dextrin (Ubukata, 1992; Larsen and Harremoes, 1994; Haldane and Logan, 1994; San Pedro et al., 1994; Goel et al., 1997/1998a,c; Confer and Logan, 1997b)

Figure 2.13. Particle size and composition used for hydrolysis experiments (Morgenroth et al. 2002)

Many studies measured hydrolysis rates using either raw wastewater or large particles extracted from raw wastewater by physical-chemical processes such as sedimentation or filtration. Bovine serum albumin and starch have been frequently used as the substrates representing protein or polysaccharide, respectively. A common conclusion is that the rate of hydrolysis decreases with the increasing particle size (Balmat, 1957). In such cases, first-order kinetics should be corrected by taking into account the hardly biodegradable material. However, it is still disagreement how far particle size is affecting hydrolysis rates, because only a few experiments has been done using defined synthetic or particles extracted from wastewater in the range from 0.1 to 100 µm (Figure 2.13). The fact is that above range of the fraction size contains the majority of particulate organic matter in settled wastewater (Levine et al., 1991).

The rate of hydrolysis not only could decrease from particle size or be slower than either death or synthesis from low temperature, but might even be inhibited. Inhibitory studies have mainly been focused on acetoclastic methanogens and acetogens, while less attention has been paid to inhibition of hydrolysis. Hydrolysis can be inhibited by the accumulation of amino acids and sugars (Sanders, 2001; Kadam et al., 2004). For example, during cellulose degradation, cellobiose as the intermediate product may be a stronger inhibitor than glucose (Duff and Murray, 1996). Volatile fatty acids (VFA) are other possible inhibitors (De Baere et al., 1985; ten Brummeler et al., 1991), however, some controversy can be found in the literature about the inhibitory effect of VFA. Veeken et al. (2000) designed a set of experiments to elucidate the mechanisms of VFA inhibition. It was concluded that no inhibition by VFA or by non-ionized VFA can be measured at pH values between 5 and 7, and that acidic pH was the inhibitor factor. They proposed a linear function of pH inhibition in the interval between 5 and 7:

$$k_{hyd} = 0.048\ pH - 0.172 \qquad \textbf{(2.17)}$$

where:

k_{hyd} – *first-order hydrolysis constant,* T^{-1}

For manure digestion, Angelidaki et al. (1993 and 1999) proposed a structured model, where hydrolysis of particulate matter was represented by a first-order reaction affected by a non-competitive reversible inhibition due to VFA. This model has been used by Keshtkar et al. (2003) with satisfactory results. A generalization of the non-competitive inhibition function, multiplying the rate coefficient, has been used by Vavilin and Angelidaki (2005) for expressing inhibition of hydrolysis by VFA (Eq. 2.18).

$$f(I) = [1 + (I/K_I)^n]^{-1} \qquad \textbf{(2.18)}$$

where:

$f(I)$ – *the function of inhibitor concentration, -*

I – *inhibitor concentration,* $M\ L^{-1}$

K_I – *the inhibition constant,* $M\ L^{-1}$

n – *a degree index, -*

The effects of pH and acetate on the hydrolysis of carbohydrate differed from those on the hydrolysis of protein (He et al., 2006). Specifically for the hydrolysis of proteins, the study of the possible effect of VFA has received special attention. While Breure et al. (1986), and Yu and Fang (2003) concluded that VFA do not inhibit protein degradation. Using gelatine as substrate, Gonzales et al. (2005) clearly showed that acetic acid reduced the gelatine hydrolysis rate in a mesophilic saline

environment, with the inhibition constant value (0.229 g COD-Ac/l) for a non competitive inhibition affecting a first-order hydrolysis. In contrast, Flotats et al. (2006) showed that no inhibition by VFA occurred during gelatine hydrolysis.

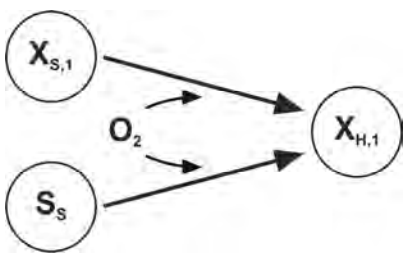
Low pH and high lipid concentration can also affect the hydrolysis (Palenzuela-Rollon, 1999). It has been stated that lipid hydrolysis hardly occurs without methanogenic bacteria that keep pH at non-acidic levels and VFA at non-toxic concentrations. Lokshina et al. (2003) showed a temporary inhibition of the hydrolysis of household waste (HSW) at acidic pH and of the slaughterhouse solid waste (SSW) degradation at neutral and alkaline pH. Simulations showed that it was most likely that hydrolysis was inhibited by high VFA concentrations in the case of HSW, lowering the pH, and by high long-chain fatty acids (LCFA) concentrations in the case of SSW. These compounds increased during hydrolysis and acidogenesis of the easily degradable fractions. Both systems recovered after an increase of the methanogenic biomass. It is difficult to distinguish the inhibitory effects caused by pH or VFA. Previous studies indicate that VFA accumulation induces a pH decrease, lowering the hydrolysis rate and making pH the effective inhibitor factor. Gradients of pH around the hydrolysable particles, which might not be measurable in the bulk liquid, could explain the different results found in the literature. Hence, the level of homogenization or mixing, and the concentration of methanogenic biomass lowering VFA, even at micro niche level, would both be factors affecting the hydrolysis rate (Vavilin et al., 2008).

2.2.4.2. Stoichiometry model concepts of hydrolysis process

Morgenroth et al. (2002) performed an overview of different hydrolysis stoichiometries, which are grouped in Table 2.14. The most important conclusions graphic concepts of models are presented below:

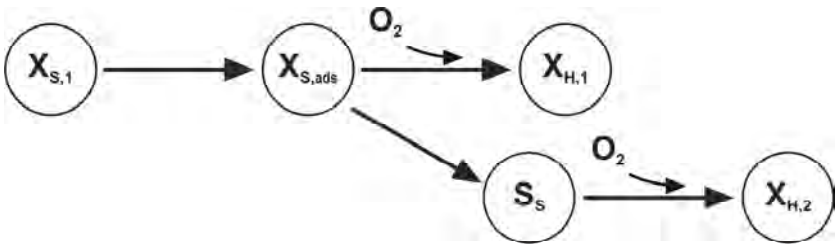
- in early study (Model 1), hydrolysis was not interpreted as a separate process. It was considered that one group of microorganisms grows directly on both biodegradable (X_S and S_S) substrates;

Model 1



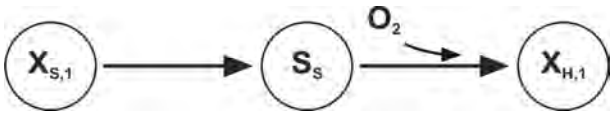
– in the Model 2 of Ekama and Marais (1979) a two biomass system was proposed, in which X_S is adsorbed and two groups of microorganisms grow separately on soluble (S_S) or adsorbed (X_S) substrate, respectively. Frigon et al., 2001 supported this hypothesis by presenting experimental evidencies from measuring rRNA levels of two bacterial populations degrading either predominantly X_S or S_S fraction;

Model 2



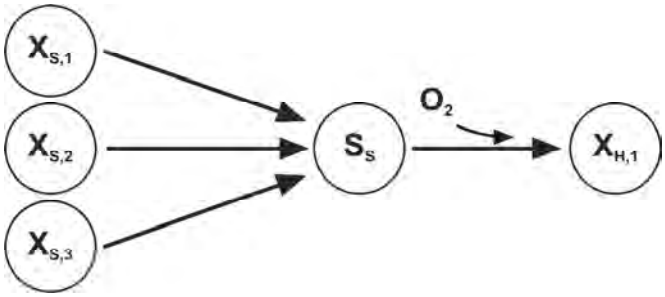
– the idea of two separate populations has been discarded in ASM1 (Henze et al., 1987). The next series of the ASMs (i.e. No 2 and 3) developed by the IWA Task Group (Gujer et al., 1999; Henze et al., 2000a) assumed the one step hydrolysis approach, in which X_S is converted into S_S (Model 3);

Model 3

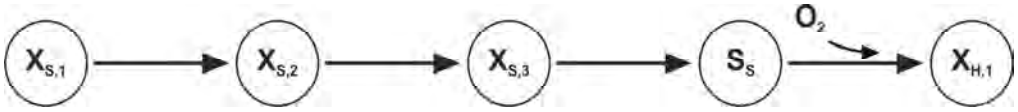


– multiple X_S fractions in parallel hydrolysis (Model 4) or sequential hydrolysis (Model 5) enclose more complicated hydrolysis stoichiometries. Parallel hydrolysis enables more flexibility in model calibration by making the utilization of different fractions independent of each other, whereas sequential hydrolysis should be considered when the accumulation of intermediate hydrolysis products is principal;

Model 4



Model 5



– hydrolysis involves no energy utilization in all of the reviewed model stoichiometries, and therefore there is no utilization of electron acceptor associated with this process.

Table 2.14. Review of hydrolysis and growth stoichiometries in various model concepts (Morgenroth et al., 2002)

Processes	$X_{S,1}$	$X_{S,2}$	$X_{S,3}$	$X_{S,ads}$	S_S	S_{O2}	$X_{H,1}$	$X_{H,2}$
Model No 1: Direct growth on both soluble and particulate organic matter (e.g. Stenstrom, 1975)								
Growth on $X_{S,1}$	$-1/Y_H$					$-(1-Y_H)/Y_H$	1	
Growth on X_S					$-1/Y_H$	$-(1-Y_H)/Y_H$	1	
Model No 2: Two biomass system (e.g. Ekama and Marais, 1979, Dold et al., 1980, Frigon et al., 2001)								
Adsorption of hydrolysable COD ($X_{S,1}$)	–1			1				
Direct growth on adsorbed COD				$-1/Y_H$		$-(1-Y_H)/Y_H$	1	
Growth on soluble					$-1/Y_H$	$-(1-Y_H)/Y_H$		1
Model No 3: One step hydrolysis (e.g. Henze et al., 1987 (ASM1); Henze et al., 1995 (ASM2), Henze et al., 1999 (ASM2d), Orhon et al., 1999 (ASM3), Sollfrank, 1988; Larsen, 1992; Spanjers and Vanrolleghem, 1995)								
Hydrolysis of hydrolysable COD ($X_{S,1}$)	–1				1			
Growth					$-1/Y_H$	$-(1-Y_H)/Y_H$	1	
Model No 4: Parallel hydrolysis (e.g. Sollfrank and Gujer, 1991; Janning, 1998; Orhon et al., 1998, Vollertsen and Hvitved-Jacobsen, 1999, Vollertsen, 1998)								
Hydrolysis of slowly hydrolysable COD ($X_{S,1}$)	–1				1			
Hydrolysis of intermediate hydrolysable COD ($X_{S,2}$)		–1			1			
Hydrolysis of rapidly hydrolysable COD ($X_{S,3}$)			–1		1			
Growth					$-1/Y_H$	$-(1-Y_H)/Y_H$	1	
Model No 5: Sequential hydrolysis (e.g. Bjerre, 1996; Confer and Logan, 1997a; Sperandio and Paul, 2000)								
Hydrolysis of slowly hydrolysable COD ($X_{S,1}$)	–1	1						
Hydrolysis of intermediate hydrolysable COD ($X_{S,2}$)		–1	1					
Hydrolysis of rapidly hydrolysable COD ($X_{S,3}$)			–1		1			
Growth					$-1/Y_H$	$-(1-Y_H)/Y_H$	1	

Note: $X_{S,1}$, $X_{S,2}$, $X_{S,3}$ – slowly biodegradable organic matter (in models with multiple X_S fraction $X_{S,1}$ is slowly and $X_{S,3}$ is rapidly hydrolysable; $X_{S,ads}$ – adsorbed X_S; S_S – readily biodegradable organic matter; S_{O2} – oxygen; $X_{H,1}$, $X_{H,2}$ – separate heterotrophic bacterial populations.

These mentioned above more complicated hydrolysis stoichiometries (Model 4 and 5) have been suggested on further investigation for three reasons:

– model predictions did not match experimental results and only were increasing the number of slowly biodegradable fractions allowed for an

improved simulations of measured data (Sollfrank and Gujer, 1991; Orhon et al., 1998);

- an accumulation of hydrolysis products was measured during the experiment, where accumulating products were still too large to be directly metabolized by bacteria (Confer and Logan, 1997a);
- in biofilm reactors, particle size has to be considered in addition to hydrolysis kinetics because mass transport of slowly biodegradable organic matter within the biofilm strongly depends on size (Janning, 1998).

The current understanding of hydrolysis is insufficient to judge which of the proposed model stoichiometries is most appropriate. Even in the relatively simple hydrolysis process of dextran to glucose (see Figure 2.11) could have multiple pathways and be a mixture of sequential and parallel processes with multiple enzymes, intermediates and diferent hydrolysis rates (see Section 2.2.2.2).

2.2.4.3. Kinetic rate expressions of hydrolysis process

Morgenroth et al. (2002) provided an overview of the kinetic rate expressions for hydrolysis (Table 2.15). Simple expressions of the hydrolysis kinetics have been divided into three groups: zero, first order or saturation type kinetics with a Monod term (Models 0-IV), whereas more complex expressions incorporate a surface-limited term (Model V) or multiple surface-limited and Monod terms with correction factors for different electron acceptor conditions (Model VI). Many authors have shown, that under specific conditions (i.e. very high or very low substrate amount to microorganism ratios), the surface limited hydrolysis rate expression in the original Model V can be simplified into first order substrate kinetics. For example, below Equation for very low substrate concentration to microorganism ($X_S << X_H$), can be approximated to Model I with the assumption of $k_{hyd,I} = k_{hyd,IV}/K_X$. In situations when $X_S >> X_H$, Model V can be approximated in terms of the biomass concentration (Model II, with $k_{hyd,II} = k_{hyd,IV}$). Likewise Equation IV also simplifies to Model III ($X_S << X_H$) and Model II ($X_S >> X_H$). On the other hand, based on experimental results with variable SRT and sludge concentration Dold et al. (1980) suggested that hydrolysis kinetics could not be described with simple Monod kinetics (such as Model IV). Then, the same authors developed a kinetic expression for hydrolysis according to the assumption of Stenstrom (1975), that X_S is adsorbed to the surface of microorganisms and is degraded by extracellular enzymes. Such cases should be sufficiently well described by surface limited hydrolysis kinetics, e.g. Model V and Model VI.

Table 2.15. Review of kinetic rate expressions for hydrolysis in various model concepts (Morgenroth et al., 2002)

No, type of expression	Kinetic expression	Reference
0 order	$k_{hyd,0}$	(e.g. Larsen, 1992; Cliff, 1980; Andrews and Tien, 1977; Dennis and Irvine, 1981; Tsuno, 1978)
I, first order *with respect to* X_S	$k_{hyd,I} \cdot X_S$	(e.g. Gujer, 1980; Henze and Mladenowski, 1991; Sollfrank and Gujer, 1991; Janning, 1998; Kappeler and Gujer, 1992; San Pedro et al., 1994; Sperandio and Paul, 2000; Spanjers and Vanrolleghem, 1995; Goronszy and Eckenfelder, 1991)
II, first order *with respect to* X_H	$k_{hyd,II} \cdot X_H$	(e.g. Goel et al., 1997)
III, first order *with respect to* X_S *and* X_H	$k_{hyd,III} \cdot X_S \cdot X_H$	(e.g. Eliosov and Argaman, 1995; Argaman, 1995; Sollfrank, 1988; Mino et al., 1995)
IV, first order *with respect to* X_H *with a Monod term (saturation type kinetics) with respect to* X_S	$k_{hyd,IV} \cdot \frac{X_S}{K_{X,IV} + X_S} X_H$	(e.g. Larsen, 1992; Mino et al., 1995; Goel et al., 1998a)
V, first order *with respect to* X_H *with a surfacelimited term for the entrapment of* X_S	$k_{hyd,V} \cdot \frac{X_S/H_H}{K_{X,V} + X_S/X_H} X_H$	(e.g. Stenstrom, 1975; Mino et al., 1995; Janning, 1998; Gujer et al., 1999 - ASM3)
VI, first order *with respect to* X_H *with multiple surface-limited and Monod terms with correction factors for different electron acceptor conditions*	$k_{hyd,VI} \cdot \left(\frac{X_S/H_H}{K_{X,VI} + X_S/X_H} \cdot \frac{S_O}{K_{O,hyd} + S_O} X_H + \eta_{NO3,hyd} \cdot \frac{X_S/X_H}{K_{X,VI} + X_S/X_H} \cdot \frac{K_{O,hyd}}{K_{O,hyd} + S_O} X_H \right)$	(e.g. Dold et al., 1980; Henze et al., 1987 - ASM1; Henze et al., 1995 - ASM2; Henze et al., 1999 - ASM2d)

Note: *X_S – slowly biodegradable substrate, $M^{-1}\ L^{-3}$; X_H – biomass of heterotrophic organisms, $M^{-1}\ L^{-3}$; k_{hyd} – hydrolysis rate constant, T^{-1}; K_X – hydrolysis saturation constant; η – hydrolysis reduction factor*

However, a surface-limited term is somewhat misleading as X_S and X_H are usually expressed as mass concentrations and their available surface area will depend on a particle size and shape. Orhon et al. (1999) evaluated the applicability of Models I, IV, V to both municipal wastewater and a number of industrial wastewaters (see Section 2.2.4.4). The authors concluded that the surface-limited rate expression (Model V) proved to be the appropriate model for the description of hydrolysis of slowly biodegradable COD in both types of wastewater. It should be noted that Mino et al. (1995) also observed that the hydrolysis of artificial substrate (starch) in the activated sludge process followed the same surface-limited reaction kinetics.

The rate of hydrolysis depends upon the magnitude of two kinetic coefficients including k_{hyd} and the adsorption saturation constant for hydrolysis, K_X. Numerical values of these coefficients, which can be found in the literature, are usually not universal and reveal a significant variation, especially for industrial wastewaters (Insel et al., 2002). Table 2.16 summarizes a wide range of values of the first-order rate of hydrolysis kinetic coefficients, that can be seen for simpler organic materials including carbohydrates, lipids, proteins and other complex mixture such as municipal solid waste or raw, settled and coagulated wastewater.

Table 2.16. Kinetic coefficients of the first-order rate of hydrolysis (Vavilin et al., 2008)

Substrate	k_{hyd} (d^{-1})	Temp. (°C)	Reference
Carbohydrate	0.5-2.0	-	Garcia-Heras (2003)
Proteins	0.25-0.8	-	Garcia-Heras (2003)
Lipids	0.76	-	Shimizu et al. (1993)
	0.1-0.7	25	Masse et al. (2002)
	0.63	-	Garcia-Heras (2003)
Municipal solid waste	0.1	15	Bolzonella et al. (2005)
Raw wastewater	7-11	-	Ginestet et al. (2002)
Settled wastewater	9-15		
Coagulated wastewater	11-21		

This wide range of values can be explained by different experimental conditions, different hydrolytic biomass to substrate ratios and effect of hydrolysis (Vavilin et al., 2008). Moreover, the biochemical pathways of different organic materials may interact between each other. According to Breure et al. (1986), a complete degradation of protein in the presence of carbohydrates often cannot be achieved in anaerobic wastewater treatment. Another important finding is that the substrate hydrolysis rate could depend strongly on the origin and the previous acclimation of the anaerobic culture (Gavala et al., 1999).

Orhon et al. (1999) showed that hydrolysis parameters can be determined from the oxygen uptake rate (OUR) profiles generated in lab-scale aerated batch reactors, but this kind of test does not allow for identification in hydrolysis kinetics. For example, a literature review performed by Ginestet et al. (2002) revealed high values (1.5-25 d^{-1}) of the first order hydrolysis rate constants estimated from respirometric experiments shorter than 24-h with raw and settled wastewater. The authors confirmed these high ranges of hydrolysis kinetic coefficients (7-21 d^{-1}) during similar tests with raw, settled and coagulated wastewater (see Table 2.16). Morover,

Ginestet et al. (2002) demonstrated that lower values (0.12-0.20 d^{-1}) were associated with long-term experiments (over the period of several days) using just the particulate matter in wastewater.

Table 2.17. The hydrolysis reduction factor for the first-order kinetic rate expression

Model	Anaerobic hydrolysis reduction factor	Anoxic hydrolysis reduction factor	Reference
	η_{fe}	$\eta_{NO3,hyd}$	
ASM1	not included	0.4	Henze et al. (1987)
ASM2	0.1	0.6	Henze et al. (1995)
ASM2d	0.4	0.6	Henze et al. (1999)
ASM3	not included	1.0	Gujer et al. (1999)
ASM3P	1.0	1.0	Rieger et al. (2001)
B&D	0.5*	1.0*	Barker and Dold (1997a)

Note: **solubilization factors are equivalent to the ASM hydrolysis reduction factors*

The hydrolysis rates could be influenced by electron-acceptor conditions as can be seen by changes in the activated sludge models. Some complex activated sludge models, which are discussed in detail (Section 2.3), incorporate two reduction factors to provide flexibility in accounting for reduced rates under anaerobic and anoxic conditions (Table 2.17). In ASM1 (Henze et al., 1987), the anoxic hydrolysis reduction factor, $\eta_{NO3,hyd}$, was set to 0.4, whereas the anaerobic hydrolysis was not considered. In ASM2 (Henze et al., 1995), the respective values of $\eta_{NO3,hyd}$ and η_{fe} defaults were 0.6 and 0.1 in temperature 20 °C. The latter coefficient in ASM2d was increased by Henze et al. (1999) to 0.4 while the value of $\eta_{NO3,hyd}$ remained unchanged. Barker and Dold (1997a) introduced in their model two (anaerobic and anoxic) solubilization factors which were equivalent to the ASM hydrolysis reduction factors. This value 1.0 presented above indicates that the hydrolysis rate under anoxic conditions is not reduced compared to aerobic conditions. Obviously, in some specific situations modifications are possible, especially when Goel et al. (1998b) demonstrated that electron-acceptor conditions slightly affected enzyme activity. Finally, Gujer et al. (1999) proved that there was no difference between hydrolysis rates under aerobic or anoxic conditions and introduced this modification in ASM3 by "eliminating" reduction factor. Rieger et al. (2001) extended ASM3 with a bio-P module and also assumed, that the anaerobic conditions have no influence on the hydrolysis rate (η_{fe} = 1.0).

2.2.4.4. Comparison of single and dual hydrolysis models

Two models of hydrolysis, single and dual, can be differentiated, because it may be difficult and sometimes misleading to characterize X_S fraction by only a single hydrolysis rate (Figure 2.15). The observed rate of hydrolysis would be dependent on both the number of viable organisms present and the maximum specific rate of hydrolysis, which can not be determined directly by analytical measurements in activated sludge systems. Identification and description all rates of particulate fraction in municipal wastewater is impossible, due to a large spectrum of compounds with different nature, properties and biodegradation patterns. The researchers (e.g. Sollfrank and Gujer, 1991; Orhon et al., 1999) argued that it might be difficult and sometimes misleading to characterize the entire X_S fraction complexity by a single hydrolysis rate. Moreover, the single hydrolysis model was observed to involve a significant risk of overestimating the biodegradation of particulate COD. Acording to Orhon et al. (1998) estimation of the single hydrolysis kinetics by evaluation of the OUR profile yielded values are very close to constants associated with the soluble (S_H) portion of the X_S. The OUR profile was evaluated by means of Equation 2.19, which reflects the oxygen utilization rate due to microbial growth induced by substrate generated through hydrolysis to endogenous respiration corrected for the generation of soluble and particulate microbial products (Orhon et al., 1998).

$$\frac{dS_o}{dt} = \frac{1-Y_H}{Y_H} \cdot \frac{Ss}{K_S + Ss} \cdot \mu_H X_H - (1 - f_{EX} - f_{ES}) \cdot b_H X_H \qquad \textbf{(2.19)}$$

where:

b_H – *endogenous decay coefficient for active biomass,* T^{-1}
f_{ES} – *fraction of endogenous mass converted into soluble inert products, -*
f_{EX} – *inert fraction of biomass, -*
Ks – *half saturation coefficient,* $M(COD)L^{-3}$
So – *oxygen,* ML^{-3}
Ss – *readily biodegradable substrate,* $M(COD)L^{-3}$
X_H – *active heterotrophic biomass, M cell* $(COD)L^{-3}$
Y_H – *heterotrophic yield coefficient, M cell* $(MCOD)^{-1}$
μ_H – *maximum heterotrophic growth rate,* T^{-1}

Endogenous respiration is modeled as a one-step process of biomass loss coupled with a direct utilization of the electron acceptor (oxygen or nitrate) (van Loosdrecht and Henze, 1999). It is assumed that during the respiration of biomass inert residues are formed from a portion (f_P) of the disappearing biomass (Grady et al., 1999):

Biomass(COD)+$(f_P-1)O_2$ *equivalents of electron acceptor* $\rightarrow f_P$ *inert residue(COD)* **(2.20)**

where:

f_P – *fraction of inert COD generated in biomass lysis (decay), -*

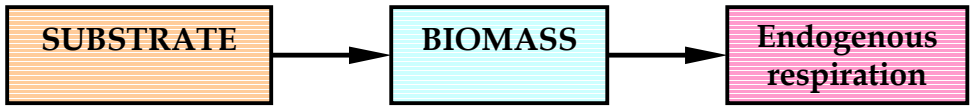


Figure 2.14. Schematic representation of endogenous respiration concepts for modeling the formation and disappearance of biomass (van Loosdrect and Henze, 1999)

The hydrolysis rate of particulate (X_S) fraction occurred at such a slow rate that could significantly interfere with endogenous decay (Grady et al., 1999). Consequently, the authors concluded that a dual hydrolysis approach is needed and justified on the basis of significantly different rate constants. The set of differential equations equally apply to single or dual hydrolysis concepts, simply by correcting Equation 2.21 and omitting or including Equation 2.22 as appropriate.

$$\frac{dS_s}{dt} = -\frac{\mu_H}{Y_H} \cdot \frac{Ss}{K_S + Ss} \cdot X_H + k_{hyd,s} \frac{S_H / X_H}{K_{XS} + S_H / X_H} \cdot X_H + k_{hyd,X} \frac{X_S / X_H}{K_{XX} + X_S / X_H} \cdot X_H \qquad \textbf{(2.21)}$$

$$\frac{dS_H}{dt} = -k_{hyd,s} \frac{S_H / X_H}{K_{XS} + S_H / X_H} \cdot X_H \qquad \textbf{(2.22)}$$

The concept of dual hydrolysis has been proposed and tested before, using continuously fed lab-scale reactors and first order kinetics (Sollfrank and Gujer, 1991), giving a totally different interpretation of the hydrolysis mechanism as compared to surface-limited reaction kinetics (Equation 2.19). Nowadays, it is commonly accepted that the soluble, rapidly hydrolysable substrate and the particulate, slowly hydrolysable fraction should be collect into an overall dual hydrolysis mechanism characterize two substrate groups by different rate coefficients (Orhon et al. 1998).

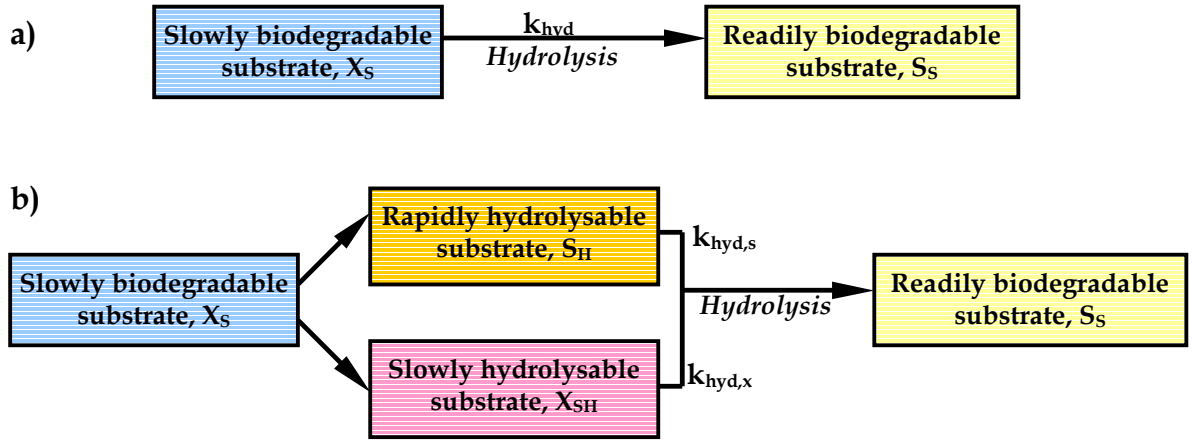


Figure 2.15. Transformation of slowly biodegradable substrate in wastewater according to (a) single hydrolysis model, (b) dual hydrolysis model

Experimental assessment of the hydrolysis rate coefficients for both domestic and a number of industrial wastewater was performed by Orhon et al. (1998) with emphasis on two different hydrolysis mechanisms associated with the rapidly and slowly hydrolyzable COD fractions. The representative hydrolysis kinetics ($k_{hyd,s}$ and K_{XS}) on the soluble S_H portion and reevaluating the total OUR profile with simultaneous dual hydrolysis approach was tested in order to assess the coefficients ($k_{hyd,x}$ and K_{XX}) related to the X_{SH}. The results of this experiments on 5 domestic wastewater samples, are summarized in Table 2.18, yielded average $k_{hyd,s}$ and K_{XS} values of 3.1 d^{-1} and 0.2 g COD/g COD associated with the hydrolysis of S_H and much lower values of 1.2 d^{-1} and 0.5 g COD /g COD for X_S. Figure 2.16 illustrates the appreciable improvement of the discrepancy between model and experimental OUR profiles obtained with one domestic wastewater sample, shifting from a single hydrolysis ($k_{hyd,s}$=2.3d^{-1}; K_X=0.4 g COD /g COD) to dual hydrolysis mechanism ($k_{hyd,s}$=3.5 d^{-1}; K_{XS}=0.1; $k_{hyd,x}$=1.8 d^{-1}; K_{XX}=0.4 g COD /g COD).

Table 2.18. Parameter estimation for domestic wastewater sample using dual hydrolysis model (Orhon et al., 1998)

Dual hydrolysis model for domestic wastewater				
Run No	**$k_{hyd,s}$**	**K_{XS}**	**$k_{hyd,x}$**	**K_{XX}**
	d^{-1}	**g COD/g COD**	**d^{-1}**	**g COD/g COD**
I-II	3.5-2.1	0.1-0.13	1.75-0.75	0.4-0.18
III-IV	3.0-2.5	0.2	1.5-0.75	0.5
V	4.5	0.3	1.0	0.8
Range	2.1-4.5	0.1-0.3	0.75-1.75	0.18-0.8
Average values	3.1	0.2	1.2	0.5

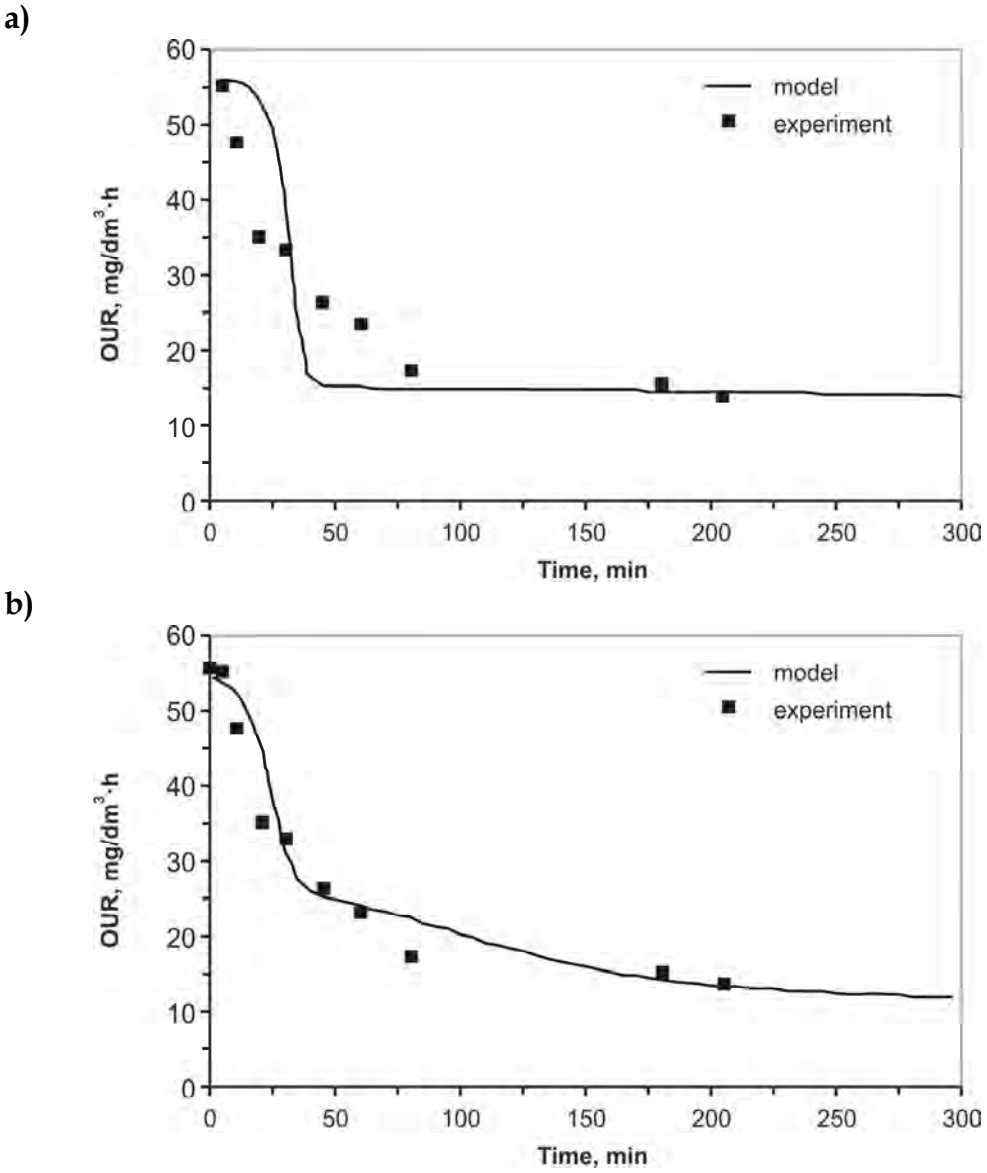


Figure 2.16. Model verification of experimental data for domestic wastewater, using (a) a single hydrolysis model, (b) a dual hydrolysis model (Orhon et al., 1998)

Acording to Okutman et al. (2001), model evaluation with the adjusted initial X_S and X_H values yielded good calibration of the OUR curve, as shown in Figure 2.17, for $k_{hyd,X}$ = 1.2 d^{-1} and K_{XX} = 0.1 g COD/g COD. The evaluation was validated by the authors with using the same kinetic coefficients for the OUR profile associated with the parallel test run at a different initial food/biomass ratio of 0.4 g COD/g VSS.

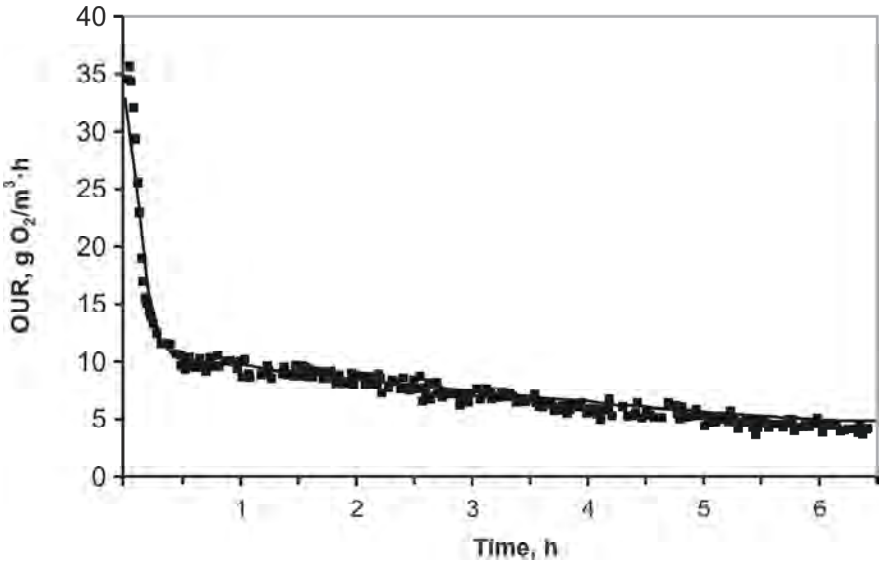


Figure 2.17. Model calibration of the OUR profile with adjusted X_S and X_H values for $k_{hyd,X}$ = 1.2 d^{-1} and K_{XX} = 0.1 g COD/g COD. Experimental data vs. model (-) for settled wastewater with food/biomass (0.2 g COD/g VSS) ratio (Okutman et al., 2001)

Henze et al. (1987 and 1995) demonstrated that values for the coefficients outlined in Table 2.19 should be evaluated within the framework of the current understanding of hydrolysis in domestic and industrial wastewater, when the entire X_S, considered as a one component in a single hydrolysis model, is not enough for validated model vs. experimental data. However, if soluble and particulate X_S components are differentiated with a dual hydrolysis model, two different hydrolysis rate coefficients should be defined (Orhon et al., 1998 and 1999). In the study of Okutman et al. (2001), k_{hyd} values were similarly determined as 3.8 d^{-1} and 1.9 d^{-1} for the soluble S_H and particulate X_{SH} of the raw wastewater, respectively. The same rate of 3.8 d^{-1} was still measured for rapid hydrolysis after settling, a physical process with no effect on the soluble COD fractions in wastewater. The rate for slow hydrolysis slightly improved to 2.1 d^{-1} in the primary effluent (supernatant), as it contained only the suspended fraction of the particulate organic matter in wastewater. Settled particulate COD was characterized by a much slower hydrolysis rate coefficient of 1.2 d^{-1}. This significantly slower hydrolysis rate makes it necessary to consider settleable COD as a separate model component in the evaluation of nutrient removal potential of wastewater (Okutman et al., 2001).

Table 2.19. Single hydrolysis rate coefficients for domestic and industrial wastewater

Source	X_S		Reference
	k_{hyd}	K_X	
	d^{-1}	g COD/g COD	
Model default values			
ASM 1	3.0	0.03	Henze et al. (1987)
ASM 2	3.0	0.1	Henze et al. (1995)
ASM 3	3.4	1.0	Gujer et al. (1999)
Experimental assesment			
Domestic wastewater	2.6	0.45	Orhon et al. (1999)
Settled particulate	1.2	0.1	Okutman et al. (2001)
Tannery wastewater			
Settling effluent	0.9	0.2	Orhon et al. (1997)
Chemical settling effluent	1.1	0.2	
Textile wastewater			
Denim processing	1.0	0.15	Orhon et al. (1998)
Predominantly textile	2.0	0.35	

Table 2.20. Dual hydrolysis rate coefficients for domestic and industrial wastewater

Source	S_H		X_{SH}		Reference
	$k_{hyd,s}$	K_{XS}	$k_{hyd,x}$	K_{XX}	
	d^{-1}	g COD/g COD	d^{-1}	g COD/g COD	
Experimental assesment					
Domestic wastewater	3.1	0.2	1.2	0.5	Orhon et al. (1999)
Raw wastewater	3.8	0.2	1.9	0.18	Okutman et al. (2001)
Settled wastewater			2.1	0.3	
Tannery wastewater					
Settling effluent	1.1	0.2	0.3	0.2	Orhon et al. (1997)
Textile wastewater					
Predominantly textile	2.5	0.40	0.1	0.5	Orhon et al. (1998)
Cotton knit fabric	3.0	0.05	1.0	0.5	
Cotton and polyester knit fabric				0.2	
Denim processing	0.8	0.05	0.15	0.15	

For comparison, the results of Orhon et al. (1998) study of single and dual hydrolysis kinetics for domestic and industrial wastewaters are summarized in Table 2.19 and 2.20. The experimental basis of the dual hydrolysis mechanism was obtained by repeating OUR batch test in duplicate on soluble and total portions of the same tannery or textile wastewater sample. A significant observation, during this experiments, was that the magnitude of the maximum specific hydrolysis rates for both S_H and X_{SH} portions were appreciable lower than their counterparts for domestic wastewater. In fact, the average values of $k_{hyd,s}$ and $k_{hyd,x}$ for tannery wastewaters were calculated as 1.1 d^{-1} and 0.3 d^{-1} respectively. On the other hand, the important feature of the results related to textile wastewater was their specific character and their variation from one textile category to another. It was found that $k_{hyd,s}$ vary from 0.8 d^{-1} for denim processing to 3.0 d^{-1} for cotton finishing. A similar variation was also depicted for $k_{hyd,x}$ ranging from 0.15 to 1.0 g COD/g COD, within the same categories (Orhon et al., 1998).

The study of Orhon et al. (1998) provided strong indication that the wide array of organics within X_S fraction could not be represented by a single hydrolysis model. The rate constants for hydrolysis of S_H were observed to be significantly higher than the ones characterizing X_S. In the case of the majority industrial effluents, the maximum specific hydrolysis rate of X_S was estimated to be at the same level as the endogenous decay constant of the biomass, generally reported as 0.24 d^{-1} (Henze et al., 1987). The observation (i.e. Orhon et al. 1998, 1999; Okutman et al., 2001) challenges the validity of all rate constants values for hydrolysis, because routine aerobic digestion tests used for the measurement of the endogenous decay constant, likely to be seriously affected by the interference of the X_S (Avcioglu et al., 1998).

3

Material and Methods

3.1. Process description and performance of the studied plants

The "Wschód" WWTP is the largest biological nutrient removal (BNR) facility in northern Poland (Figure 3.1). This is also one of the largest plant located upon the Baltic Sea, serving mainly a population of Gdańsk, Sopot and other small towns and communities such as Pruszcz Gdański, Żukowo, Kolbudy and Juszkowo. The second largest BNR facility in northern Poland upon the Baltic Sea is "Dębogórze" WWTP (Figure 3.1). This plant is serving mainly the population of Gdynia and surrounding smaller towns and communities such as Reda, Rumia and Wejherowo. The treated wastewater is discharged into the Bay of Puck through a 9-km open channel. At a distance of more than 2.3-km from the coastline underwater collector (below the seabed) is completed a set of diffusers mounted at a depth of about 8 m.

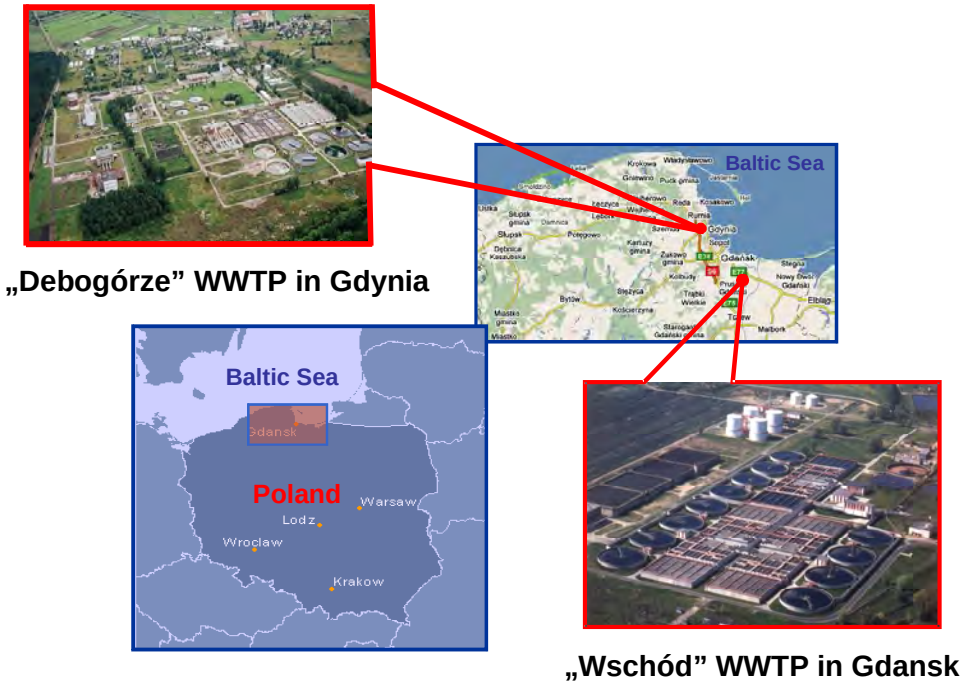


Figure 3.1. Location and view of the "Wschód" and "Dębogórze" WWTP.

3.1.1. Wschód WWTP in Gdańsk

The total pollutant load to the "Wschód" WWTP corresponds to approximately 570,000 PE although the various industrial wastewater (mostly from primarily food industry and shipyard) discharges contribute not more than 11% of the total wastewater inflow quantity. The average daily influent flow rate to the plant was 81,000 m^3/d during the study period (December, 2007 – May, 2009). Currently, the average daily flow entering the plant is 94,000 m^3/d. Although a separate sewerage system exists in the entire catchment, under rainfall events the influent flow rate

occasionally increases from 20 to 70% compared to the dry-weather conditions. Hourly flows vary in a wide range from almost 0 m^3/h to 13,000 m^3/h. The storm water and sanitary sewer systems are separated in the catchment area. The treated wastewater is directly discharged to the Bay of Gdańsk in the distance almost 3 km from the coast through the underwater tube, which was constructed at the bottom of the Baltic Sea.

The "Wschód" WWTP went into operation in the early 1970's, but only mechanical treatment was provided until 1998. The biological step, completed in 1998-1999, consists of six parallel bioreactors and twelve circular secondary clarifiers and its designed hydraulic capacity is equal to 180,000 m^3/d. Due to the low actual influent flow rates only three or four bioreactors were under operation during the study period. The bioreactors run in the MUCT (Modified University of Cape Town) process configuration (Figure 3.2). The total volume of a single bioreactor is 26,350 m^3 of which the aerobic zone occupies 11,700 m^3.

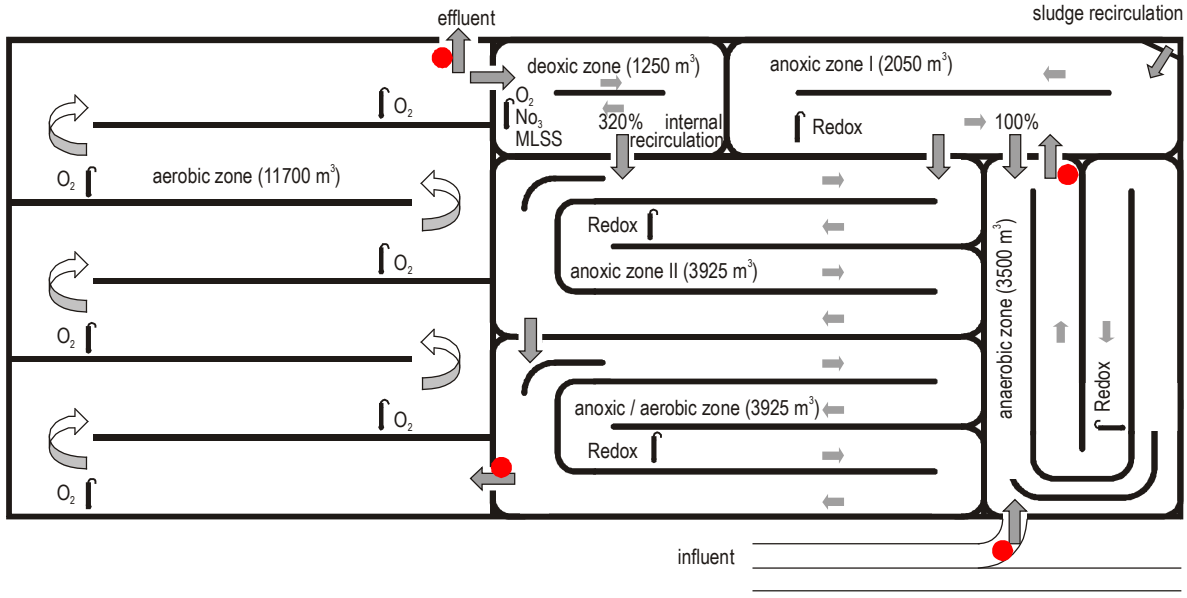


Figure 3.2. Layout of the bioreactor at the "Wschód" WWTP and location of the sampling (red) points during the measurement campaign (Swinarski, 2011).

The deoxic zone (1,250 m^3) in the internal recirculation line from the aerobic to anoxic zone is a minor modification to the original MUCT configuration. The aerobic zone has been designed as a plug flow reactor with six compartments, whereas the other zones, i.e., one anaerobic (3,500 m^3) and two anoxic (2,050 m^3 and 7,850 m^3), have been designed as carrousel systems. The internal mixed liquor anoxic recycle (MLR1 from the first anoxic zone to the anaerobic zone) and nitrified recycle (MLR2 from the last aerobic compartment to the second anoxic zone) are set to constant flow rates equal to 30,240 m^3/d and 92,544 m^3/d, which is 112% and 343%, respectively, of the

average influent flow rate. The returned activated sludge (RAS) recirculation from the bottom of the secondary clarifiers to the first anoxic zone is accomplished by 2 or 3 pumps with the capacity 10,056 m^3/d each. The mixed liquor from each bioreactor flows to two circular secondary clarifiers with the surface area of 1510 m^2 (Ø = 44.0 m) and the side water depth of 3.0 m.

Redox electrodes are installed in the both anaerobic and anoxic zones. The deoxic zone is equipped with instruments for "on-line" measurements of DO, NO_3-N, NH_4-N, PO_4-P and MLSS concentrations. The air is supplied to the aeration zone by means of a diffused aeration system. The DO concentrations and air supply in that zone are controlled based on the readings of DO probes installed in each aerated compartments. The set points are set independently in each compartment of the aeration zone (6 points). Chemical precipitation is used rarely due to very high efficiency of biological phosphorus removal.

From December, 2007 to May, 2009 (including the study period), the activated sludge system at the "Wschód" WWTP operated at temperatures varying within the range of 11.8 °C (January) - 20.5 °C (July). The MLSS concentrations were maintained at 5,450 kg/m^3 and the corresponding SRTs were equal to 21.3 d. The average concentrations of conventional parameters for the settled wastewater were as follows: COD = 669 g COD/m^3, BOD_5 = 298 g BOD_5/m^3, TSS = 285 g/m^3, TP = 14.9 g P/m^3, PO_4-P = 9.8 g P/m^3, TN = 79.8 g N/m^3 and NH_4-N = 58.9 g N/m^3. The monthly average COD/TN ratios in the settled wastewater ranged from 6.8 to 11.3 and the wastewater contained a significant soluble fraction (SCOD), i.e., 21% to 39% of total COD, which was determined according to the rapid physical-chemical method of Mamais et al. (1993). More details concerning the monthly average composition of wastewater characteristics and operating conditions of the "Wschód" WWTP during the period 2007-2009 can be found in Table 3.1.

The occurrence probabilities of the effluent concentrations and reduction efficiency of TN and TP in a 1.5-year study period (December, 2007 to May, 2009) are presented in Figure 3.3. During last years, the annual average concentrations of the total nitrogen discharged from the plant varied between 10-11 g N/m^3. From March 2011, the effluent limit of total nitrogen (15 g N/m^3) should be reduced to 10 g N/m^3 in accordance with the Regulation of the Ministry of Environment (ME Regulation, 2006).

Table 3.1. Characteristics of the "Wschód" WWTP in Gdańsk during the period 2007-2009 (monthly average values)

Parameter	Unit	Minimum	Maximum	Average	Std. Deviation
Influent characteristics					
COD	**g COD/m³**	536	876	669	± 82
SCOD	**g COD/m³**	153	245	194	± 25
BOD_5	**g BOD_5/m³**	216	385	298	± 35
TN	**g N/m³**	69.7	94.5	79.8	± 4.9
NH_4-N	**g N/m³**	52.3	65.9	58.9	± 3.4
TP	**g P/m³**	8.9	18.5	14.9	± 2.6
PO_4-P	**g P/m³**	5.6	12.0	9.8	± 1.8
TSS	**g/m³**	227	339	285	± 30
VSS	**g/m³**	143	212	176	± 20
Alkalinity	**val/m³**	9.0	10.3	9.7	± 0.3
Effluent characteristics					
COD	**g COD/m³**	41.2	60.1	47.6	± 4.2
SCOD	**g COD/m³**	34.6	43.6	38.5	± 2.1
BOD_5	**g BOD_5/m³**	3.7	13.1	6.7	± 2.1
TN	**g N/m³**	9.4	14.3	11.0	± 1.1
NH_4-N	**g N/m³**	0.5	3.7	1.2	± 0.7
NO_3-N	**g N/m³**	6.1	8.5	7.4	± 0.6
TP	**g P/m³**	0.4	1.1	0.6	± 0.1
PO_4-P	**g P/m³**	0.1	0.5	0.2	± 0.1
TSS	**g/m³**	8.2	23.8	12.6	± 3.5
VSS	**g/m³**	2.4	15.4	5.8	± 2.8
Alkalinity	**val/m³**	5.2	5.8	5.5	± 0.2
Operating parameters					
Influent flow rate to a single reactor (Q)	**m³/d**	21,640	32,780	26,900	± 2,790
RAS flow rate (Qras)	**m³/d**	20,110	30,170	25,140	± 7,111
MLR1 flow rate (Qmlr1)	**m³/d**	30,240	30,240	30,240	± 0
MLR2 flow rate (Qmlr2)	**m³/d**	92,540	92,540	92,540	± 0
MLSS concentration	**g/m³**	4,430	6,610	5,450	± 560
SRT	**d**	16.2	28.3	21.3	± 2.9
Process temperature	**°C**	11.8	21.0	16.5	± 3.0

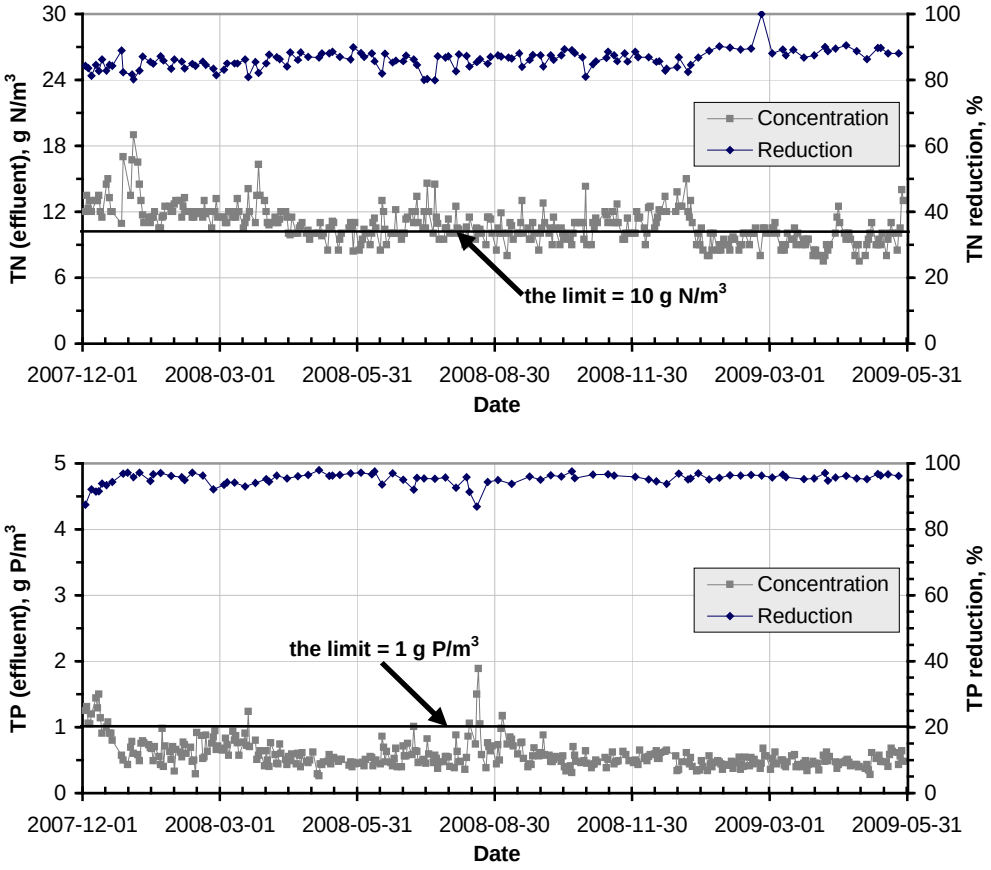


Figure 3.3. Efficiency of nutrient removal in the biological part of the "Wschód" WWTP during the study period (December, 2007 to May, 2009)

3.1.2. Dębogórze WWTP in Gdynia

The "Dębogórze" WWTP provided only mechanical treatment until the beginning of the 1990's. The first biological step (a conventional two-stage activated sludge process) was completed in 1994, but even then the effluent quality did not meet the Polish standards of 1991. Therefore, a further expansion towards biological nutrient removal was carried out in the years of 1994-1997. Until 2009, the treatment line has consisted of four parallel bioreactors run in the Johannesburg (JHB) process configuration and six secondary clarifiers. After the expansion and modernization completed in June, 2009, the plant meets the stringent requirements on the quality of treated wastewater (TN = 10 g/m^3, TP = 1.0 g/m^3) in accordance with the Regulation of the Ministry of Environment (ME Regulation, 2006). The existing biological step was expanded, just before studied periods at "Dębogórze" WWTP, with new treatment lines giving the total volume of 104,000 m^3. The JHB process configuration was designed with simultaneous nitrification-denitrification in a carrousel system.

The by-pass of settled wastewater was constructed in order to bring sewage directly to the denitrification/nitrification zone.

The total volume of a single bioreactor line is 12,000 m^3, of which the aerobic zone occupies 4,625 m^3. A layout of the new bioreactor, including the volumes of individual compartments and the locations of on-line instruments, is shown in Figure 3.4. The anaerobic and pre-denitrification (Pre DN) zones have been constructed as completely mixed reactors. Plug flow reactors were designed for the following compartments: anoxic, intermediate (anoxic/aerobic) and four aerobic. The air is supplied to the aerobic zone with a diffused aeration system, which is controlled by on-line measurements of DO in the third compartment of the aerobic zone at the set point of 2-4 g O_2/m^3. In the bioreactor, could be also found several other probes and sensors such as a DO and ORP, that have been installed in the first aerobic compartment and in the anaerobic (Bio P) or anoxic zone, respectively (Figure 3.4).

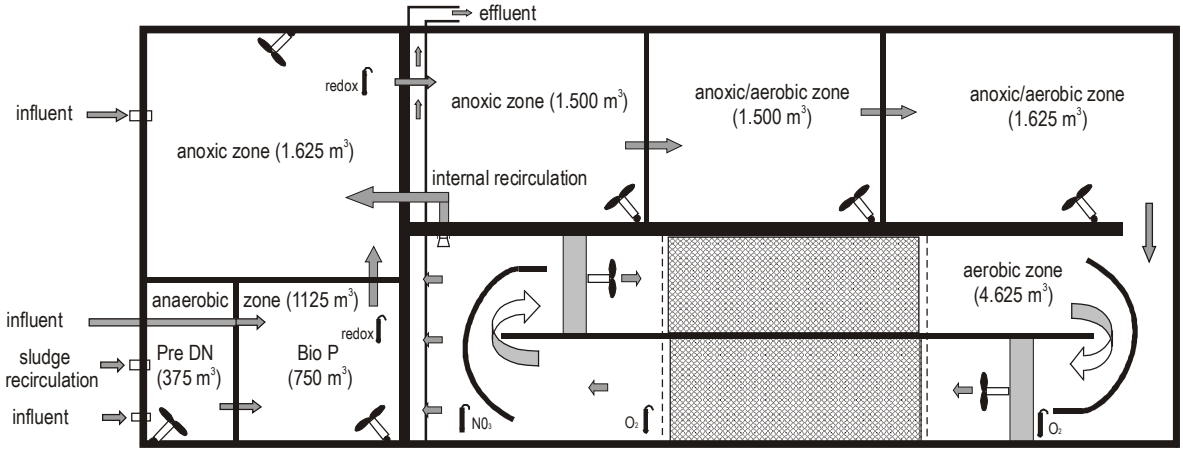


Figure 3.4. Layout of the bioreactor after modernization at the "Dębogórze" WWTP.

The mixed liquor from all of the bioreactors flows to eight circular secondary clarifiers having the surface area of 1370 m^2 (Ø = 41.7 m) and the total depth of 4.6 m. The old aeration tanks of the first stage were adapted as the primary sludge fermenters. Occasionally, in order to ensure an adequate level of phosphorus removal, iron sulfate (PIX) can be added to the bioreactor effluent to precipitate most of the remaining soluble phosphorus (simultaneous precipitation).

Currently, the average pollutant load corresponds to approx. 420,000 PE due to a significant contribution of industrial wastewater discharges, even though in terms of volume they account for only 16% of the total amount. The average daily influent wastewater flow rate was equal to 55,000 m^3/d during the fall and spring study

periods (September-November 2009 and May, 2010). However, during heavy rainfall events, the peak hourly influent flow rates were reaching even 9000 m^3/h.

Table 3.2. Characteristics of the "Dębogórze" WWTP in Gdynia during the period 2009-2010 (monthly average values)

Parameter	Unit	Minimum	Maximum	Average	Std. Deviation
Influent characteristics					
COD	**g COD/m^3**	541	1,230	856	± 116
VFA	**g/m^3**	68	299	167	± 59
BOD_5	**g BOD_5/m^3**	140	500	319	± 61
TN	**g N/m^3**	33.7	110	86.5	± 10.6
NH_4-N	**g N/m^3**	29.6	81.8	65.1	± 7.45
TP	**g P/m^3**	5.6	23.1	12.2	± 2.46
PO_4-P	**g P/m^3**	2.43	10.2	7.6	± 1.4
TSS	**g/m^3**	120	620	255	± 70
VSS*	**g/m^3**	210	320	255	± 32
Alkalinity	**val/m^3**	5.8	10.6	9.3	± 0.8
Effluent characteristics					
COD	**g COD/m^3**	15.2	31.9	25.4	± 4.7
BOD_5	**g BOD_5/m^3**	3.2	5.3	4.2	± 0.6
TN	**g N/m^3**	7.0	10.6	8.4	± 2.1
NH_4-N	**g N/m^3**	0.5	1.7	0.9	± 0.3
NO_3-N	**g N/m^3**	4.8	7.7	6.0	± 0.7
TP	**g P/m^3**	0.2	0.8	0.5	± 0.2
PO_4-P	**g P/m^3**	0.1	0.6	0.3	± 0.2
TSS	**g/m^3**	5.0	7.2	5.5	± 7.7
Alkalinity	**val/m^3**	4.0	4.8	4.5	± 0.3
Operating parameters					
Influent flow rate (Q)	**m^3/d**	46,250	98,815	54,688	± 7,579
RAS flow rate (Qras)	**m^3/d**	11,556	126,041	55,559	± 28,853
MLSS concentration	**g/m^3**	2,790	6,610	4,750	± 900
SRT	**d**	16.1	49.3	29.2	± 7.8
Process temperature	**°C**	15.4	17.8	16.6	± 1.2

Note: * Data compatible with own (not from WWTP's database) laboratory analysis during the fall, 2009 & spring, 2010 sessions

During the fall and spring study periods from September-November 2009 and May, 2010, the activated sludge system at the "Dębogórze" WWTP operated at temperatures varying within the range of 17.8 °C (September) - 15.4 °C (May). The average MLSS was 4750 g/m^3 and the corresponding SRTs were equal to 29 d. The average concentrations of conventional parameters for the settled wastewater were as follows: COD = 856 g COD/m^3, BOD_5 = 319 g BOD5/m^3, TSS = 255 g/m^3, TP =

12.2 g P/m^3, PO_4-P = 7.6 g P/m^3, TN = 86.5 g N/m^3 and NH_4-N = 65.1 g N/m^3. More details concerning wastewater characteristics and operating conditions from the study period can be found in Table 3.2. Variations in the effluent concentrations of nutrients (TN and TP) and their removal efficiency during the study period from September, 2009 to June, 2010 are presented in Figure 3.5.

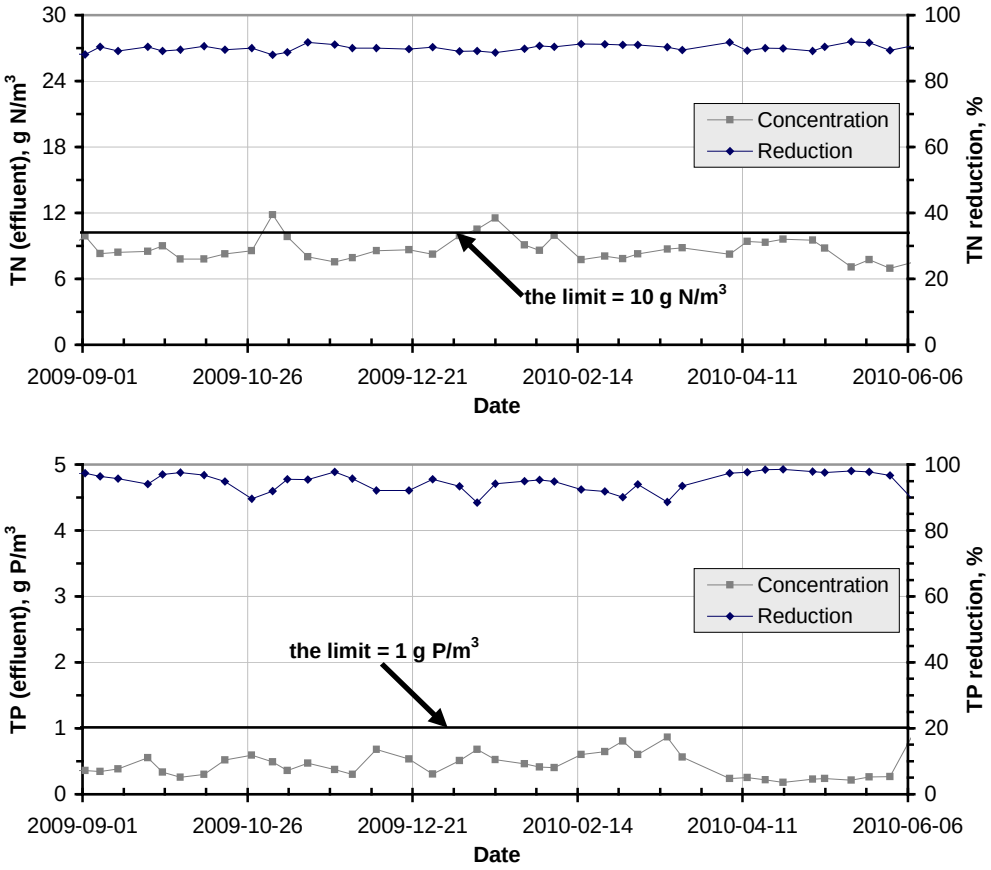


Figure 3.5. Efficiency of nutrient removal in biological part of the "Dębogórze" WWTP during the study period (September, 2009 to June, 2010).

3.2. Description of the laboratory experiments

During the period from December, 2007 to May, 2009 and September, 2009 to June, 2010 three and two experimental series of various batch tests were carried out at "Wschód" and "Dębogórze" WWTP, respectively (Figure 3.6). Within the framework of each series termed winter, spring, summer ("Wschód" WWTP) and fall, spring ("Dębogórze" WWTP) the experiments were repeated.

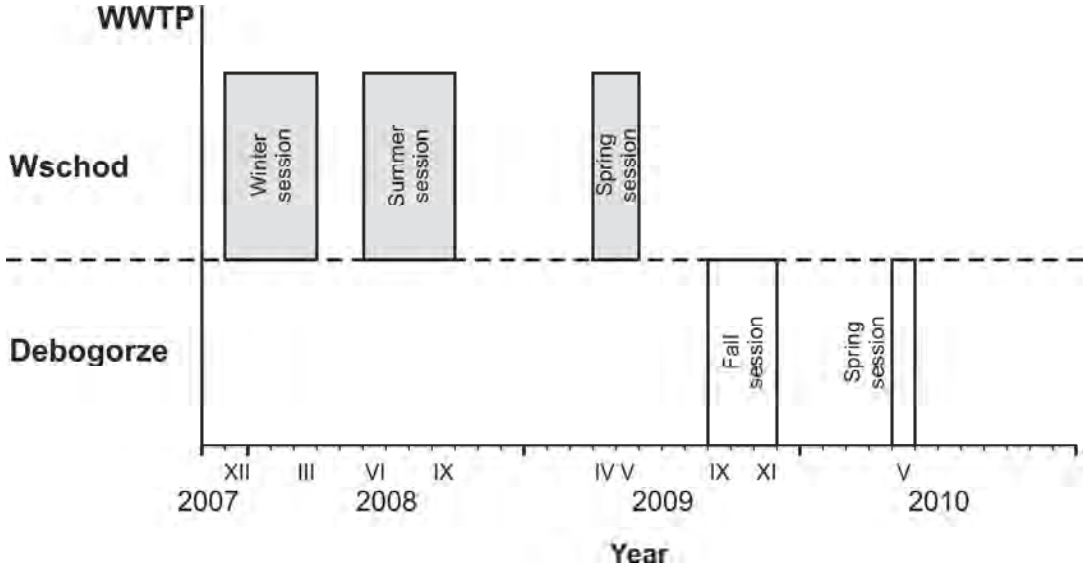


Figure 3.6. Schedule of the laboratoty experiments at the "Wschód" and "Dębogórze" WWTP

3.2.1. Experimental set-up

Laboratory experiments were carried out in a specially designed and constructed experimental set-up consisting of two parallel, plexiglass batch reactors (max. volume of 4.0 dm^3), automated monitoring, control box and computer (Figure 3.7 a-b). The reactors were equipped with electrodes for a continuous monitoring of pH, ORP, temperature and dissolved oxygen (DO) probes (WTW Cellox 325). The automated control system were introduced from the computer and maintained a desired set point DO for concentration and temperature in the reactors. The DO concentration was controlled at the selected set point between 0-6 g/m^3 during anaerobic/anoxic and aerobic conditions, respectively. The temperature set point in a water coat of both batch reactors was controled by refrigerated/heating circulators system (JULABO F32-EH) and was set constant close to the actual process temperature in the full-scale bioreactors, i.e. 12-14°C, 15-17°C and around 19-20°C, respectively, during the winter, spring and summer session. In addition, a small chamber designed for continuous measurements of OUR was connected with the main unit. Its operation in a cyclic mode was initiated automatically through opening an electromagnetic valve. One cycle of the OUR measurement (3.5 min) consisted of the following phases: emptying, rinsing, filling and measuring (3 min). The oxygen depletion was measured using a DO probe with a stirrer (WTW Stirrox G), placed in the OUR chamber. The experimental data (pH, DO, T, ORP) were collected on-line by computer from control box and electrodes/probes installed in each batch reactor. All of the measured parameters were recorded with the interval of 10 s and stored in the computer memory (Figure 3.8 a-b).

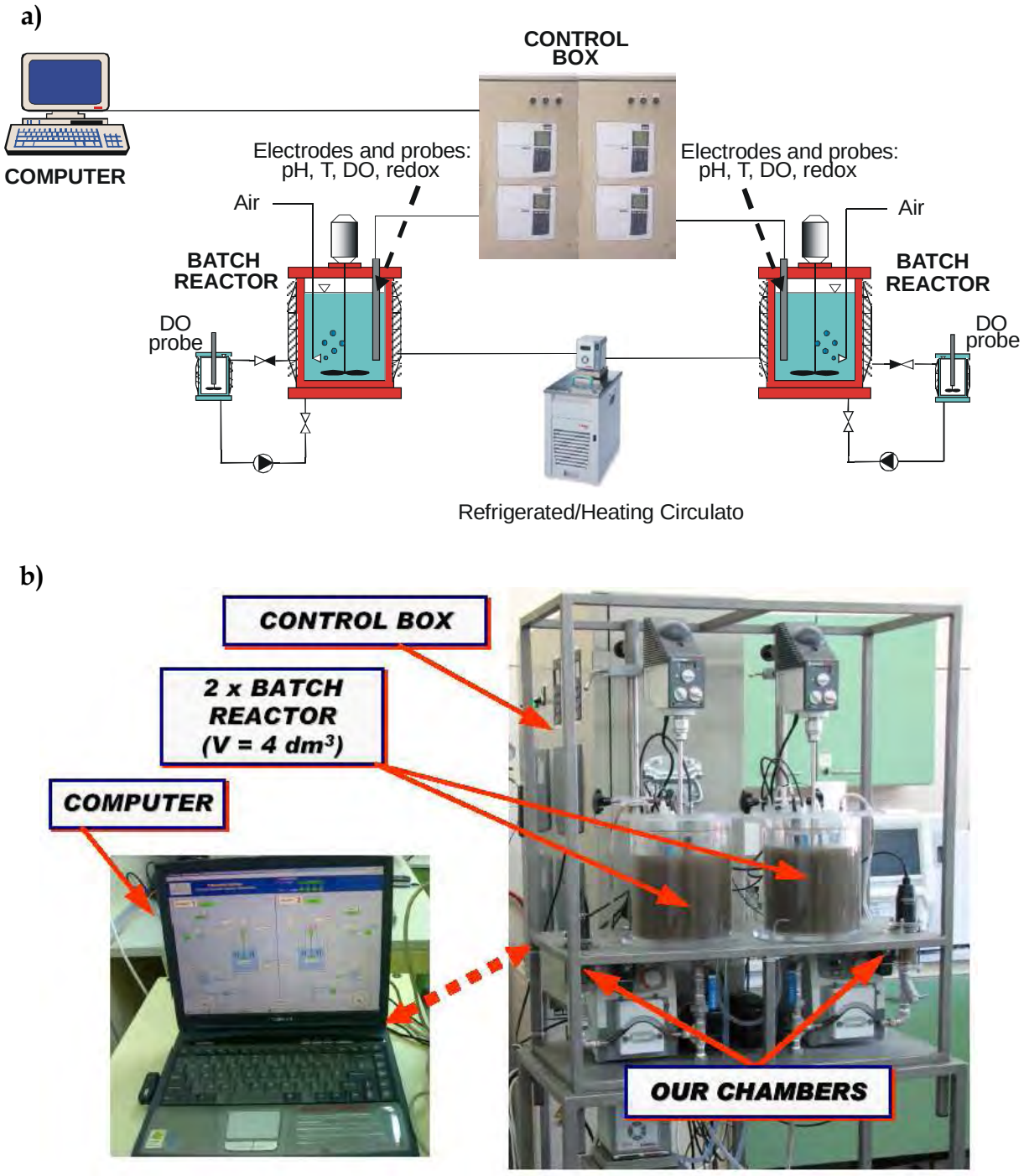


Figure 3.7. Schematic diagram (a) and actual view (b) of the laboratory apparatus with accompanying equipment for determination biochemical process rates during batch tests.

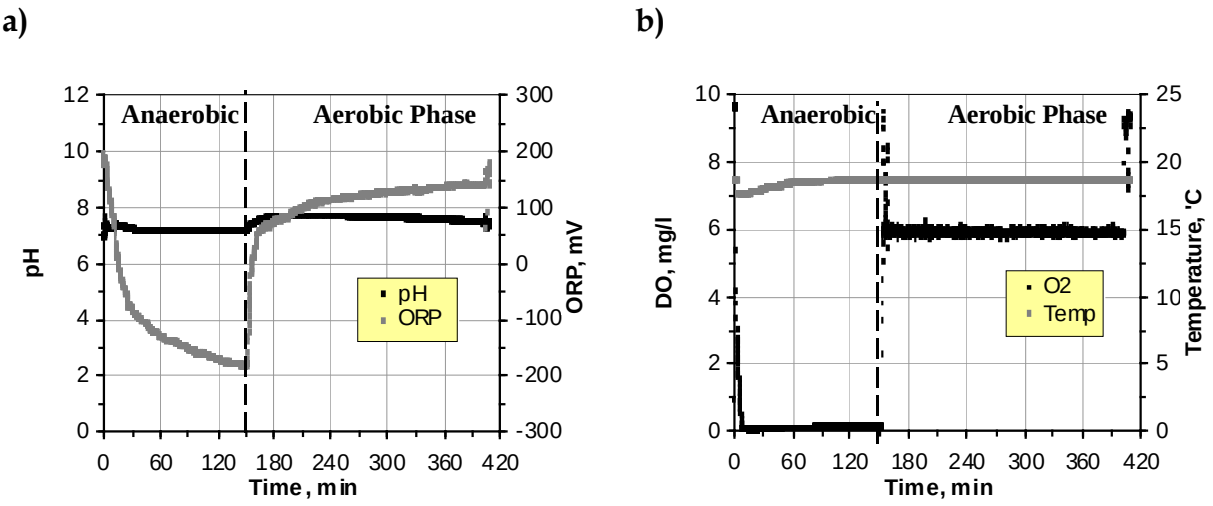


Figure 3.8. Example of the functioning of the monitoring system in the batch reactor (PRR and aerobic PUR test): (a) pH and redox (ORP), (b) dissolved oxygen (DO) and temperature.

3.2.2. Method for determination of the effect of particulate and colloidal compounds

The 24-hour composite samples of the settled wastewater entering the full-scale bioreactors were used in the experiments. The laboratory experiments were conducted in two parallel batch reactors in order to distinguish the slowly biodegradable substrate from wastewater indirectly (Figure 3.9). In the first reactor, the process biomass (fresh returned activated sludge) was mixed with the settled wastewater without pretreatment (containing soluble, colloidal and particulate organic fractions), whereas the settled wastewater after coagulation-flocculation was used in the second reactor. The latter sample of wastewater, containing only a soluble organic fraction, was prepared according to the rapid physical-chemical method of Mamais et al. (1993).

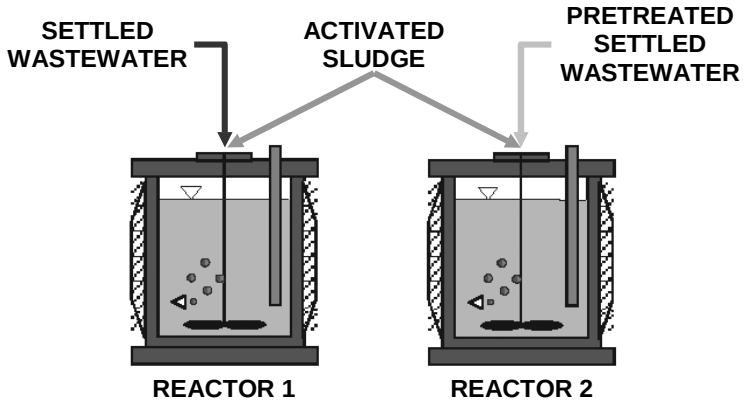


Figure 3.9. Diagram illustrating the experimental procedure with two parallel batch reactors.

That method is based on the rationale that membrane filtration of sample that has been flocculated (by precipitating with $ZnSO_4$ at pH = 10.5) will remove the colloidal particles from a filtrate leaving only truly soluble organic matter in the filtrate. After removing colloids and particulates, the pH was adjusted to its original value by adding 6M HCl. For comparison, Goel et al. (1999) proposed a similar experimental procedure to separate hydrolysis from storage by performing and analyzing two parallel oxygen uptake rate (OUR) measurements: one with filtered wastewater (including soluble COD) and the other with non-filtered wastewater (including total COD).

3.2.3. Procedures of laboratory experiments

Each experimental series comprised different types of batch tests (conventional oxygen uptake rate, OUR and nitrate utilization rate, NUR; anaerobic phosphate release and anoxic/aerobic phosphate uptake rate, PRR and anoxic/aerobic PUR), referred as:

One-phase batch tests

- ✓ ***Conventional oxygen uptake rate (Conventional OUR)*** – at the beginning fresh returned activated sludge and 30 mg of nitrification inhibitor (ATU) were prepared in a similar way for both parallel batch reactors. At the same time both samples were diluted together with settled wastewater (reactor 1) and settled wastewater after coagulation-flocculation (reactor 2) to obtain 3 dm^3 of mixed liquor with the MLSS concentration about 2.5 kg/m^3. After dissolving the components, the wastewater was combined with the sludge in each reactor in order to measure the actual MLSS and MLVSS concentration at the beginning of each batch test. The mixer device, equipped with a mechanical stirrer and aeration system, were turned on. The automated measurement of OUR and on-line monitoring of temperature, DO, pH and ORP were also initiated. Temperature in the water coat of the batch reactor was set at a desired value. The DO set point was controlled at 6 g O_2/m^3. The measurement of OUR was conducted for 6-8 hours. The samples of the mixed liquor (V = 50 cm^3) were withdrawn with the frequency of 5-60 min, filtered under vacuum pressure on the Whatman GF/C filter and analyzed for COD. The OUR measurement in one cycle was conducted for 3 min. Based on the measurements of total OUR (OUR_H), endogenous OUR (OUR_{Hend}) and degraded COD (S_S), the Y_H coefficient was determined after rearranging Equation 2.6.

- ✓ ***Conventional nitrate utilization rate (Conventional NUR)*** – a sample of fresh returned activated sludge and settled wastewater was prepared in a similar way to the OUR test for both batch reactors except that a source of nitrate (KNO_3) was injected at the beginning of the test (20 mg N/dm^3). After adding the mixture to each parallel reactor, the mixer device equipped with a mechanical stirrer and aeration system were turned on. The automated measurement and on-line monitoring of temperature, DO, pH and ORP were also initiated. Temperature in the water coat of the batch reactor was set at a desired value. The test was run for 4 hours and the samples were withdrawn with the frequency of 10-30 min, filtered under vacuum pressure on the Whatman GF/C filter and analyzed for NO_3-N, PO_4-P and COD. The actual MLSS and MLVSS concentrations in both reactors were measured at the beginning of each batch test. The dilution rate of process biomass was adjusted to obtain MLSS at approx. 2.5 kg/m^3 in the reactors.

<u>Two-phase batch tests</u>

- ✓ ***Phosphate release rate under anaerobic conditions and phosphate uptake rate under anoxic/aerobic conditions (PRR and anoxic/aerobic PUR)*** – the sample of fresh returned activated sludge and settled wastewater was prepared in a similar way to the NUR test. After adding the mixture to each parallel reactor, the mixer device equipped with a mechanical stirrer and aeration system were turned on. The automated measurement and on-line monitoring of temperature, DO, pH and ORP were also initiated. Temperature in the water coat of the batch reactor was set at a desired value. The test was run for 6.5-hour: a 2.5-hour anaerobic phase was followed by an anoxic or aerobic phase, both lasting 4-hours. In the anoxic PUR test, a source of nitrate (KNO_3) was injected at the beginning of an anoxic phase (20 g N/m^3). In the aerobic PUR test, the air supply system was turned on at the beginning of the aerobic phase (DO set point = 6 g O_2/m^3) and the OUR measurement was initiated. The samples were withdrawn with the frequency of 10-30 min, filtered under vacuum pressure on the Whatman GF/C 1.2 m pore size filter and analyzed for PO_4-P, COD and NO_3-N/NH_4-N (anoxic/aerobic phase). The actual MLSS and MLVSS concentrations in both reactors were measured at the beginning and at the end of each batch test. The dilution rate of process biomass was adjusted to obtain MLSS at approx. 2.5 kg/m^3 in the reactors.

3.2.4. Analytical methods

The total or soluble COD, PO_4-P, NO_3-N and NH_4-N were analyzed by Hach "test-in-tube" using Xion 500 spectrophotometer (Dr Lange GmbH, Germany). The TN concentrations were measured using TOC/TN analyzer (SHIMADZU Corporation, Japan). The analytical procedures, which were adapted by Dr Lange GmbH (Germany), followed the Standard Methods (APHA, 1992). Total suspended solids (TSS) and volatile suspended solids (VSS) analyses were performed in accordance with the Polish Standards (PN-72/C-04559).

3.3. Mathematical modeling and computer simulation

3.3.1. Conceptual and mathematical models of hydrolysis

The hydrolysis concept in the original ASM2d and its modification (dashed arrow) considering two-step hydrolysis process with a new variable (rapidly hydrolyzable substrate, X_{SH}), is illustrated in Figure 3.10. Based on this concept, a mathematical model for CNP activated sludge systems was developed and referred further to Table 3.3 as an "modified" ASM2d. The new model incorporates one new component, X_{SH}, and three new processes, i.e., hydrolysis of X_{SH} under aerobic, anoxic and anaerobic conditions. The new variable, X_{SH}, is termed "readily hydrolyzable organic compounds" in order to implement two-step hydrolysis process and denote, that in comparision with X_S, this kind of substrate is hydrolyzed faster than X_S under aerobic, anoxic and anaerobic conditions. The modified ASM2d was implemented using the Model Developer module in the GPS-X simulation platform (see Section 3.3.2). The original ASM2d is described in Appendix 1.

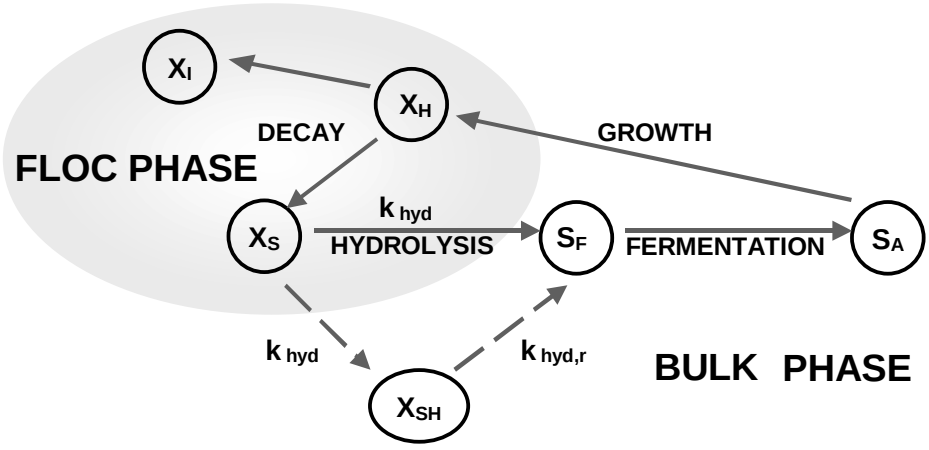


Figure 3.10. The hydrolysis concept of the original ASM2d and its modification (dashed arrows) considering two-step hydrolysis process with new variable (X_{SH}).

Table 3.3. Stoichiometric matrix and process rates for the modified ASM2d including the new variable X_{SH} and three new hydrolysis processes under aerobic, anoxic and anaerobic conditions

Process \ Variable	S_F	S_{NH4}	S_{PO4}	S_I	S_{ALK}	X_{SH}	X_S
Aerobic hydrolysis of X_S		$\nu_{1,NH4}$	$\nu_{1,PO4}$		$\nu_{1,ALK}$	1	-1
Anoxic hydrolysis of X_S		$\nu_{2,NH4}$	$\nu_{2,PO4}$		$\nu_{2,ALK}$	1	-1
Anaerobic hydrolysis of X_S		$\nu_{3,NH4}$	$\nu_{3,PO4}$		$\nu_{3,ALK}$	1	-1
Aerobic hydrolysis of X_{SH}	$1-f_{SI}$	$\nu_{22,NH4}$	$\nu_{22,PO4}$	f_{SI}	$\nu_{22,ALK}$	-1	
Anoxic hydrolysis of X_{SH}	$1-f_{SI}$	$\nu_{23,NH4}$	$\nu_{23,PO4}$	f_{SI}	$\nu_{23,ALK}$	-1	
Anaerobic hydrolysis of X_{SH}	$1-f_{SI}$	$\nu_{24,NH4}$	$\nu_{24,PO4}$	f_{SI}	$\nu_{23,ALK}$	-1	

Process	Process rate, ρ_j
Aerobic hydrolysis of X_S	$k_{hyd} \frac{S_{O2}}{K_{O2}+S_{O2}} \frac{X_S/X_H}{K_X+X_S/X_H} X_H$
Anoxic hydrolysis of X_S	$k_{hyd} \eta_{NO3} \frac{K_{O2}}{K_{O2}+S_{O2}} \frac{X_S/X_H}{K_X+X_S/X_H} X_H$
Anaerobic hydrolysis of X_S	$k_{hyd} \eta_{fe} \frac{K_{O2}}{K_{O2}+S_{O2}} \frac{K_{NO3}}{K_{NO3}+S_{NO3}} \frac{X_S/X_H}{K_X+X_S/X_H} X_H$
Aerobic hydrolysis of X_{SH}	$k_{hyd,r} \frac{S_{O2}}{K_{O2}+S_{O2}} \frac{X_{SH}/X_H}{K_{Xr}+X_{SH}/X_H} X_H$
Anoxic hydrolysis of X_{SH}	$k_{hyd,r} \eta_{NO3} \frac{K_{O2}}{K_{O2}+S_{O2}} \frac{X_{SH}/X_H}{K_{Xr}+X_{SH}/X_H} X_H$
Anaerobic hydrolysis of X_{SH}	$k_{hyd,r} \eta_{fe} \frac{K_{O2}}{K_{O2}+S_{O2}} \frac{K_{NO3}}{K_{NO3}+S_{NO3}} \frac{X_{SH}/X_H}{K_{Xr}+X_{SH}/X_H} X_H$

3.3.2. Simulation platform

The General Purpose Simulator (GPS-X) ver. 5.0.2 (Hydromantis, 2007) was used in this study as a simulator environment for implementing the examined models, running simulations as well as performing parameter estimation and optimization. GPS-X consist of several modules:

- ✓ ***Simulator*** is module that allows to run simulations of more than 50 preconfigured models included with GPS-X, as well as own custom-designed layouts.

- ✓ ***Builder module*** uses GPS-X's graphical interface to custom-design defined plant layouts or modify existing process flow diagrams.

- ✓ ***Model Developer (MD)*** is an utility module that allows to create and edit user-defined biokinetic models or make changes to the models existing in the GPS-X libraries. MD is built on the Microsoft Excel spreadsheet and consists of series of spreadsheet pages that contain information on model structure in the Petersen matrix format, parameters and GPS-X variables.

- ✓ ***Analyzer module*** is used to conduct sensitivity analysis on process layouts. The objective of a sensitivity analysis is to determine the sensitivity of the model output variables (dependent variables) to changes in its parameters (independent variables). This provides insight into the model's behavior and helps identify the parameters that have the greatest impact on the model.

- ✓ ***Optimizer module*** is a flexible, dynamic optimization package for evaluating important model parameters. After the important parameters have been identified, GPS-X optimizer can be used to fit a model to measured data or to optimize process performance. The procedure of fitting a model to measured data is called parameter estimation and involves adjusting selected model parameters to achieve the best possible fit between the model responses and the measured data. Optimizer may be used in optimizing a WWTP operations (e.g., determine the best air flow distribution in an activated sludge tank to optimize effluent quality and minimize aeration costs) and also to automate the model calibration process, enabling fine-tuning of one or more parameters so that the predicted data fitted the measured ones more closely. Optimization involved adjusting certain model parameters to maximize or minimize an objective function, and it is carried out by using the Nelder-Mead simplex method (Press et al., 1986) in the optimizer module.

The GPS-X program has a large number of pre−compiled models supplied in libraries, which covers many unit processes found in WWTPs. The user, depending upon processes to be modeled, may introduce modifications to the existing GPS−X models or even build completely new models. A process layout is built by manipulating graphical icons on a program drawing board and assigns the appropriate descriptive models with default parameter values. After placement graphic objects in GPS- X simulation platform, model types have to be specified in order to retrieve them from the library. When the connections between objects are specified, a dynamic model of the entire process layout may be generated. Olsson and Newell (1999) classified GPS−X as a very powerful tool dedicated for dynamic simulation of municipal and industrial wastewater treatment systems.

A schematic layouts in the GPS-X platform of the MUCT and JHB bioreactor from "Wschód" and "Dębogórze" WWTP, respectively are presented in Figure 3.11 (a-b). The batch reactors also in the GPS-X simulator, reflecting lab-scale tests, are shown in Figure 3.12.

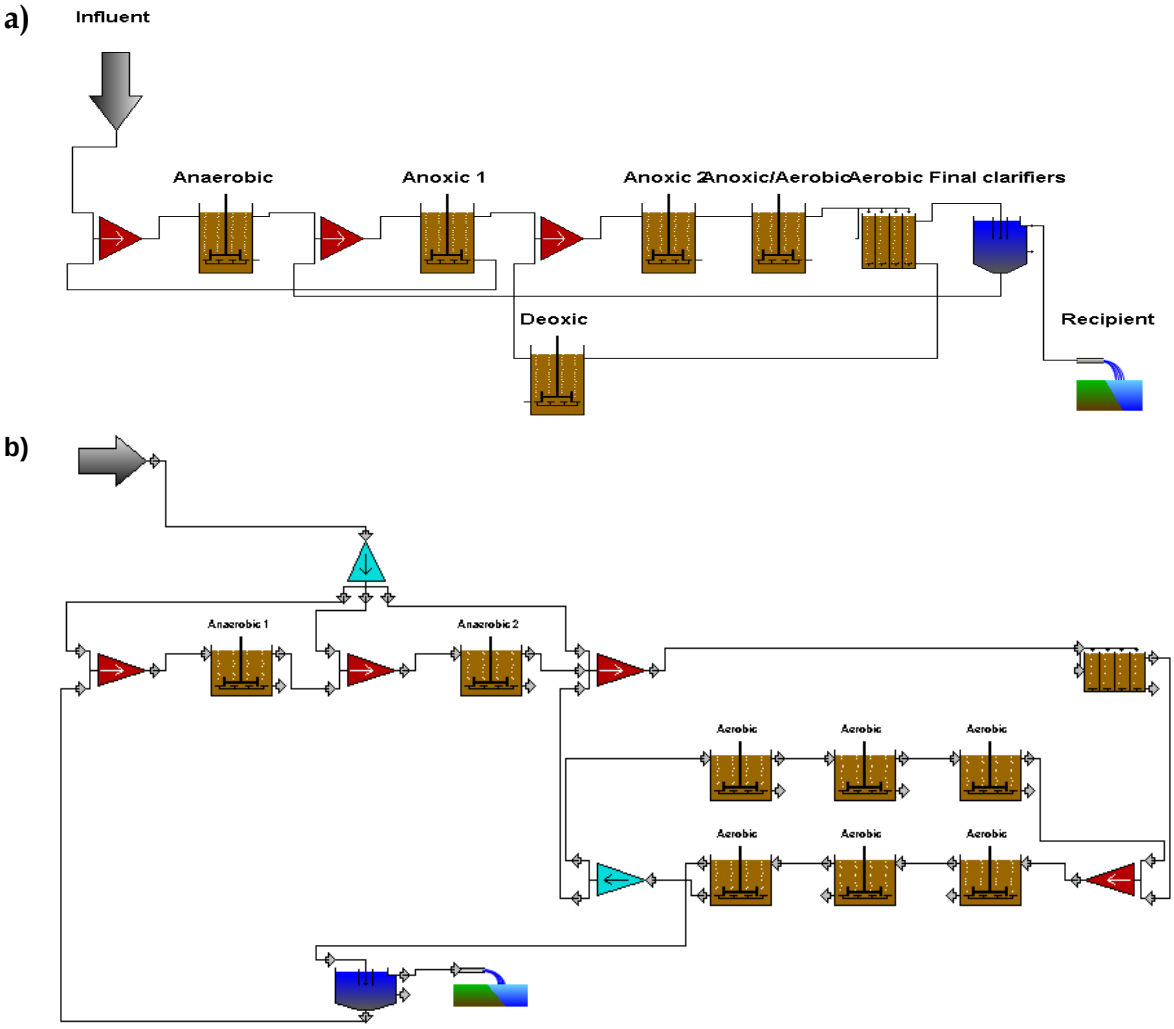


Figure 3.11. A schematic layout of the (a) MUCT process at the "Wschód" WWTP and (b) JHB process at the "Dębogórze" WWTP in the GPS-X simulator.

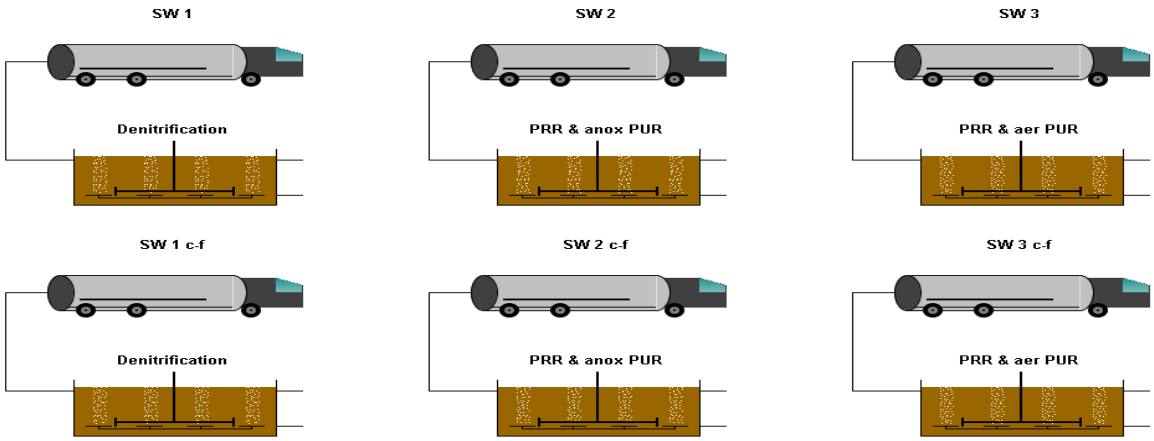


Figure 3.12. A schematic layout of the batch reactors in the GPS-X simulator.

3.3.3. Organization of the simulation study at Wschód and Dębogórze WWTP

The modeling study followed the steps presented in Figure 3.13.

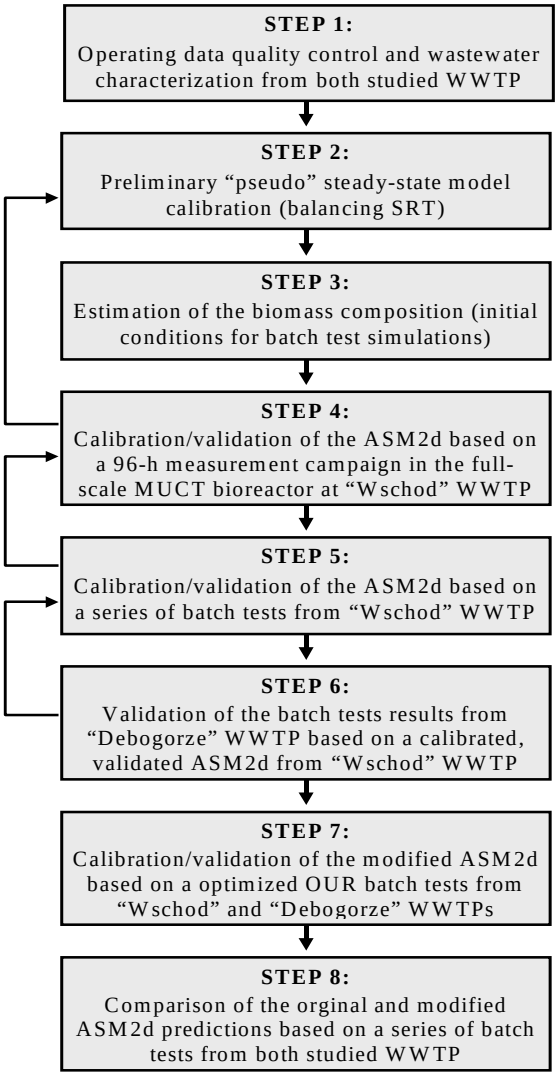


Figure 3.13. Pathway of the development and evaluation of the ASM2d and its modification for predicting the effects of X_S in combined N-P activated sludge systems.

Table 3.4. Procedure for the fractionation of organic matter in wastewater and evaluation of conversion factors based on the literature data and measurements carried out at "Wschód"and"Dębogórze" WWTPs during studied periods (Mąkinia, 2006)

Measured parameters				
Definition	**Symbol**	**Unit**	**Monthly average value Wschód/Dębogórze**	**Source of data**
Influent COD	COD_{in}	gCOD/m³	669/856	Laboratory analyses at study WWTP
Influent COD in filtered sample	$COD_{f,in}$	gCOD/m³	194/211*	
Volatile fatty acids	VFA	g/m³	-/167	
Influent BOD_5	$BOD_{5,in}$	gBOD_5/m³	298/319	
Influent biodegradable COD	$BCOD_{in}$	gCOD/m³	509/545	Calculation (Grady et al, 1999): $BCOD_{in} = \frac{BOD_{5,in}}{f_{BOD} \cdot (1 - Y_H \cdot f_P)}$
Effluent COD	COD_{out}	gCOD/m³	47.6/25.4	Laboratory analyses at study WWTP
Effluent COD in filtered sample	$COD_{f,out}$	gCOD/m³	38.5/20.5**	
BOD_5/BOD_U ratio	f_{BOD}	–	0.67	(Grady et al, 1999)
Heterotrophic biomass yield coefficient	Y_H	gCOD/ gCOD	0.63	
Unbiodegradable fraction from the biomass decay	f_P	–	0.2	

Model components – influent					
Fraction name	**Symbol**	**Unit**	**Equation**	**Average % of COD**	
				This procedure	**Calibration of SRT**
Inert soluble	S_I	gCOD/m³	$0.95 \cdot COD_{f,out}$	5.5/2.8	
Readily biodegradable	S_S	gCOD/m³	$COD_{f,in} - S_I$	23.5/21.8	
Slowly biodegradable	X_S	gCOD/m³	$BCOD_{in} - S_S$	52.6/41.8	47.2/38.8
Inert particulate	X_I	gCOD/m³	$COD_{in} - COD_{f,in} - X_S$	18.4/33.6	23.8/36.6

Conversion factors (i_N, i_P) – influent
$TKN - S_{NH} = i_{N,SS} \cdot S_S + i_{N,SI} \cdot S_I + i_{N,XS} \cdot X_S + i_{N,XI} \cdot X_I$
$i_{N,SI} = \frac{(TKN_{f,aer} - S_{NH,aer})}{COD_{f,out}}$ and $TKN_{f,aer}$ and $S_{NH,aer}$ were measured in the aerobic zone
$P_{tot.} - S_{PO4} = i_{P,SS} \cdot S_S + i_{P,SI} \cdot S_I + i_{P,XS} \cdot X_S + i_{P,XI} \cdot X_I$

Conversion factors (i_{VSS}) – bioreactor
$X_{COD} = X_H + X_A + X_{PAO} + X_{STO} + X_S + X_I$
$X_V = i_{VSS,BM} \cdot (X_H + X_A + X_{PAO}) + 0.6 \cdot X_{STO} + i_{VSS,XS} \cdot X_S + i_{VSS,XI} \cdot X_I$
$X = X_{inorg} + X_{VSS}$

Note: ** Mesured by coagulation-flocculation method of Mamais et al. (1993) during batch tests*
*** Correlated with daily measurements of $COD_{f,out}/COD_{out}$ ratio at "Wschód" WWTP*

In step 1, the influent wastewater and routine operating data of the MUCT and JHB bioreactor at the "Wschód" and "Dębogórze" WWTP, respectively, were collected and evaluated. The fractionation of organic matter in the settled wastewater was performed according to a similar procedure as applied by Mąkinia (2006). This procedure was developed based on the standard Dutch STOWA guidelines for wastewater and sludge characterization, presented in Table 3.4. However, some minor modifications and assumptions need to be justified. The standard laboratory analyses at "Dębogórze" WWTP did not include the measurement of soluble COD The additional measurements to evaluate the contribution of S_S to total COD were needed. During each batch experimental tests, the coagulation-flocculation method of Mamais et al. (1993) was used to determine the concentration of soluble COD in grab samples of the settled wastewater (see Section 2.1.3). Moreover it was nessecery for ASM2d to differentiate between two fractions of S_S, fermentable organic matter (S_F) and fermentation products (S_A). This was done using the actual laboratory measurements of VFAs (assumed to be equal to S_A) in the settled wastewater at both plants. The X_S/X_I ratio was estimated by calibrating the SRT (step 3) independently for each study period.

The nutrient (N and P) content of specific organic fractions was estimated by subtracting measured concentrations of NH_4-N from TKN and PO_4-P from TP, respectively (Table 3.4). The additional information with respect to the content of inert, soluble fraction (S_I) was also provided by the measurements of TKN and TP in the filtered grab samples withdrawn from the aerobic zone effluent. The conversion factors for X_S and X_I ($i_{N,SI}$, $i_{N,XS}$, $i_{N,XI}$ and $i_{P,XS}$) were adjusted to obtain the best fits between the measured and calculated values of the following parameters:

- differences between the concentrations of TKN - NH_4-N and TP - PO_4-P in the primary effluent ($i_{N,XS}$, $i_{N,XI}$ and $i_{P,XS}$, $i_{N,XI}$),
- particulate N and P concentrations in the aerobic zone of the bioreactor ($i_{N,XS}$, $i_{N,XI}$ and $i_{P,XS}$, $i_{N,XI}$),
- soluble N concentrations in the aerobic zone of the bioreactor ($i_{N,SI}$).

No changes were made to the values of conversion factors for S_S ($i_{N,SS}$ and $i_{P,SS}$,) S_F ($i_{N,SF}$ and $i_{P,SF}$) S_I ($i_{P,SI}$) and X_I ($i_{P,XI}$) in ASM2d.

The i_{VC} (MLVSS/COD) ratio was assumed based on the literature data (Grady et al., 1999). Hence, the default value of conversion factors in ASM2d was modified from 0.75 g VSS/g COD to 0.7 g VSS/g COD, which is equivalent to 1.43 g COD/g VSS and remains within the typical range of 1.42-1.48 g VSS/g COD as reported by Grady et al. (1999). The i_{VT} (MLVSS/MLSS) ratio was a target calibration parameter for the steady-state model calibration.

In step 2, the SRT were preliminary calibrated along with the effluent concentrations of the most important qualitative parameters, i.e. NH_4-N and NO_3-N, PO_4-P, based on a steady-state simulation for the summer study period. Due to the calibration of SRT, the initially estimated X_S/X_I ratio (step 1) was modified for each study period. The new value of the ratio was kept constant for the organic fractionation of settled wastewater samples, either used in the batch tests, or collected during the measurement campaign in the bioreactor.

In step 3, the result of a steady-state model calibration, also the initial biomass composition was determined for dynamic simulations of the batch tests. A special Excel spreadsheet was developed to facilitate the preparation of input files with the initial biomass composition. The GPS-X input files were automatically generated for all of the batch tests by inserting a single output GPS-X file from the previous step (step 2). The files contained steady-state concentrations of the model components in the RAS line (a sampling point of the process biomass used in the laboratory experiments).

In step 4, prediction capabilities of the original ASM2d was calibrated/validated under dynamic conditions, based on the comparison of measured and predicted concentrations of NH_4-N, NO_3-N and PO_4-P in the full-scale MUCT bioreactor. The "continuous" 96-hour measurement campaign from the summer study period provided data for this purpose (see Section 3.3.3.1). During the simulations of full-scale bioreactor performance, the DO and MLSS concentrations in the bioreactor were PI-controlled on the measured values.

In step 5, the batch tests (such as NUR, OUR, PRR and anoxic/aerobic PUR) carried out with settled wastewater without pretretment and after coagulation-flocculation from the summer study period at the "Wschód" WWTP were simulated with the same set of kinetic and stoichiometric parameters as used in step 4. The selected parameters were adjusted sequentially to fit the measured and predicted rates of the following batch tests biochemical processes (denitrification, oxygen uptake, anaerobic phosphate release, anoxic/aerobic phosphate uptake and nitrification) in order calibrated/validated ASM2d.

In step 6, the stoichiometric and kinetic coefficients adjusted for "Wschód" WWTP, were used to validate the results of the similar batch tests from the fall study period at "Dębogórze" WWTP. Then the examined model was further validated against the measured data from batch tests, originating at both studied plants from the other (spring and winter) study periods. The set of model parameters remained unchanged

in comparison with the calibration stage (steps 2-4). The initial biomass composition was obtained by a steady state simulation of the full-scale bioreactor performance using the average values of operating data from these periods. The procedure of generating the input files for dynamic simulations was the same as in step 3.

In step 7, Model Developer was used in order to modify ASM2d and then evaluate this new model, with two-step hydrolysis model, described in detail in the Section 3.3.1. For this purpose, the results of OUR batch tests from both studied plants carried out with the WWTP mixed liquor (taken from returned activated sludge RAS) and settled wastewater were used to adjust selected kinetic and stoichiometric coefficients in the modified ASM2d. Predictions of the original and modified ASM2d were fitted to the experimental data of the batch tests by automatic fine-tuning by means of the Optimizer module in the GPS-X ver. 5.0.2 simulation platform (Hydromantis, 2007). The filtered COD and OUR were set as the target variables for each test with the settled wastewater without pretretment or after coagulation-flocculation from the summer study period. The iteration loops were repeated until one set of model parameters could be used in all series of OUR batch tests.

Finally in step 8, the predictions with the original and modified ASM2d of all types of batch tests from both studied plants were compared, in terms of the process rates describing hydrolysis and/or COD, NO_3-N, PO_4-P, NH_4-N, OUR profiles. The average relative deviation (ARD) was introduced as a measure tool between the both models prediction accuracy:

$$ARD = \frac{1}{n_j} \cdot \sum_{j=1}^{m} \frac{\left| \left(y_{j,obs} - y_j \right) \right|}{y_{j,obs}} \cdot 100\% \qquad \textbf{(3.1)}$$

where:

ARD – *average relative deviation, %*

n_j – *number of experimental data points for the output variable j*

$y_{j,obs}$ – *measured (observed) value of the output variable j*

3.3.3.1. Full-scale measurement campaign at Wschód WWTP

In September, 2008, a 96-hour measurement campaign was carried out in the full-scale MUCT bioreactor. Wastewater samples were taken every two hours at the following locations: reactor inlet and effluent from anaerobic, anoxic, aerobic zone (Figure 3.2). The samples from inlet was filtered through a 0.45 µm filter membrane in order to analyze COD the filtered samples (COD_f). In the other sampling points, the samples were filtered under the gravity pressure using paper filters. The scope of laboratory analyses in each sampling point is presented in Table 3.5. The similar investigations have been described elsewhere (Swinarski, 2011).

Table 3.5. Scope of the laboratory analyses in the sampling points during continuous 96-h measurement campaigns in the full-scale bioreactors

Parameter	Sampling point in the activated sludge reactor			
	INF	**ANAEROBIC**	**ANOXIC**	**AEROBIC**
COD/COD_f	+	-	-	-
TP	+	-	-	-
PO_4-P	+	+	+	+
TN	+	-	-	-
NH_4-N	+	+	+	+
NO_3-N	-	-	+	+

Note: INF – *reactor inlet, ANAEROBIC – effluent from the anaerobic zone, ANOXIC – effluent from the anoxic zone, AEROBIC – effluent from the aerobic zone*

In addition, the on-line recordings of all flow rates (influent, two mixed liquor recycles, RAS and WAS), process temperature, and DO concentrations in the aerobic compartments were also collected for the modeling study. Variations in the single activated sludge reactor of influent flow rate (Q_{inf}), concentrations of TN, NO_3-N and PO_4-P during the measurement 96-h campaign are illustrated in Figure 4.12.

3.3.3.2. Collection of a database for the modeling study

The full-scale routine operating data were collected from the period of 2007-2009 at the „Wschód" WWTP and 2009 – 2010 at the „Dębogórze" WWTP. During that period, laboratory analyses of conventional parameters in the primary effluent (including sludge liquors) were performed approximately 10 times per month at the "Wschód" WWTP and 5 times per month at the "Dębogórze" WWTP. On the same days, composite samples of wastewater in the secondary effluent were analysed for the same range of parameters. Operating parameters measured in the bioreactors included the SRT, MLSS and MLVSS concentrations, process temperature and DO concentrations in all the aerobic compartments at studied WWTP. The last two parameters were measured on-line, similar to the flow rates (influent flow rate, internal recirculations and RAS flow rate), and recorded based on one-hour intervals.

The lab-scale data for modeling study were collected from batch tests results at both studied plants in order to evaluate primarily the effect of X_S on the denitrification capability and EBPR in the combined N-P activated sludge systems (see Section 3.2.3). The average values of laboratory analyses, operating parameters and on-line measurements were used further as input data for steady-state simulations, which generated the initial biomass composition for dynamic simulations of the batch experiments and performance of the full-scale bioreactors.

3.3.3.3. Data quality evaluation

The routine operating and performance measurements, averaged over the three and two study periods, respectively at the "Wschód" and "Dębogórze" WWTP, were evaluated (according to a similar procedures as applied by Mąkinia, 2006) using a continuity check for flow rates and mass balance calculations for oxygen demand (OD), solids, nitrogen and phosphorus. To begin with the continuity check for flow rates and mass balances for suspended solids (over the clarifier) and phosphorus (over the entire activated sludge system) were used to verify the conventional calculations of SRT (Figure 3.14). The appropriate set of equations can be written in the following form:

– continuity check: $$Q_{in} = Q_{out} + Q_{was} \quad \textbf{(3.2)}$$

– solids balance over the clarifier:

$$(Q_{out} + Q_{ras} + Q_{was}) \cdot X_{asr} = (Q_{ras} + Q_{was}) \cdot X_{ras} + Q_{out} \cdot X_{out} \quad \textbf{(3.3)}$$

– P balance over the entire system: $$Q_{in} \cdot P_{in} = Q_{out} \cdot P_{out} + Q_{was} \cdot P_{was} \quad \textbf{(3.4)}$$

where:

P_{in} – *total P concentration in (primary) influent, M(P)L^{-3}*
P_{out} – *total P concentration in secondary effluent, M(P)L^{-3}*
P_{was} – *total P concentration in WAS, M(P)L^{-3}*
Q_{out} – *effluent flow rate from secondary clarifier, L^{3}T^{-1}*
Q_{was} – *waste activated sludge (WAS) flow rate, L^{3}T^{-1}*
X_{asr} – *solids (MLSS) concentration in the bioreactor, ML^{-3}*
X_{out} – *solids concentration in secondary effluent, ML^{-3}*
X_{ras} – *solids concentration in RAS, ML^{-3}*

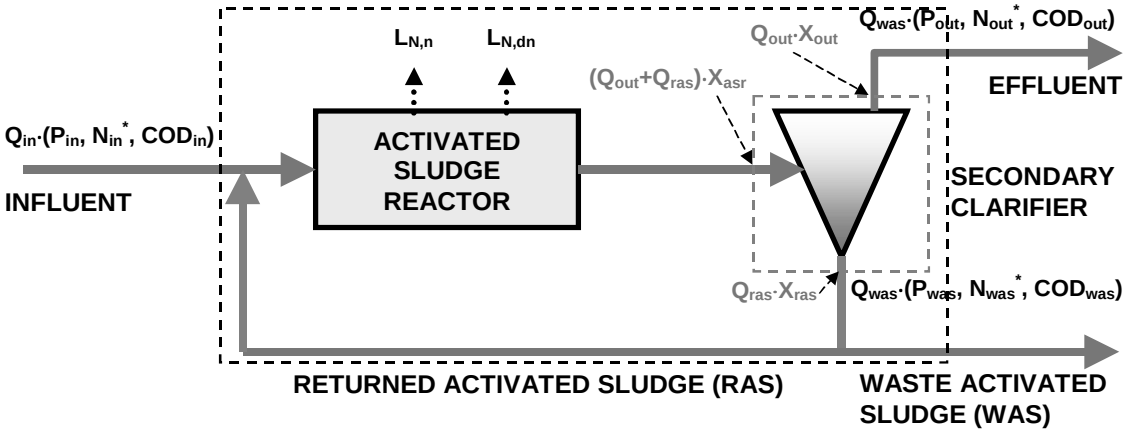


Figure 3.14. Schematic overview of the continuity and mass balances described in Equations 3.2-3.7 (Mąkinia, 2006)

Equation 3.3 can be balanced by adjusting X_{ras} and used further for the calculation of P_{was} in Equation 3.4, which leads to the estimation of Q_{was} and revised calculation of the SRT. Then, the OD mass balance over the entire system, including the total loads

of denitrified N ($L_{N,dn}$) and nitrified N ($L_{N,n}$), was calculated from the following equation:

$$OU_{tot} = Q_{in} \cdot COD_{in} - Q_{out} \cdot COD_{out} - 2.86 \cdot L_{N,dn} + 4.57 \cdot L_{N,n} - Q_{was} \cdot X_{was} \cdot i_{VT} \cdot i_{CV} \qquad \textbf{(3.5)}$$

where:

COD_{in} – *influent COD, M(COD)L*$^{-3}$
COD_{out} – *COD in secondary effluent, M(COD)L*$^{-3}$
i_{VT} – *MLVSS/MLSS ratio, MM*$^{-1}$
$L_{N,dn}$ – *total load of denitrified N, M(N)T*$^{-1}$
$L_{N,n}$ – *total load of nitrified N, M(N)T*$^{-1}$
OU_{tot} – *total oxygen uptake in bioreactor, M(O*$_2$*)T*$^{-1}$

The $L_{N,dn}$ and $L_{N,n}$ are part of the overall OD balance and can be calculated from the following mass balance equations:

– total denitrified N load $L_{N,dn} = Q_{in} \cdot N_{tot.,in} - Q_{out} \cdot N_{tot.,out} - Q_{was} \cdot N_{tot.,was}$ **(3.6)**

– total nitrified N load $L_{N,n} = Q_{in} \cdot TKN_{in} - Q_{out} \cdot TKN_{out} - Q_{was} \cdot TKN_{was}$ **(3.7)**

where:

$N_{tot.,in}$ – *influent total N concentration, M(N)L*$^{-3}$
$N_{tot.,out}$ – *effluent total N concentration, M(N)L*$^{-3}$
$N_{tot.,was}$ – *total N concentration in WAS, M(N)L*$^{-3}$
TKN_{in} – *total Kjeldahl N concentration in (primary) influent, M(N)L*$^{-3}$
TKN_{out} – *total Kjeldahl N concentration in secondary effluent, M(N)L*$^{-3}$
TKN_{was} – *total Kjeldahl N concentration in WAS, M(N)L*$^{-3}$

The calculated value of total oxygen uptake (OU_{tot}) in the bioreactor could not be verified with direct measurements, but it could be compared with predictions of the calibrated model (ASM2d) which were examined in the simulation study.

4

Results and Discussion

4.1. Characteristics of the wastewater samples from the studied plants

At the "Wschód" WWTP, the soluble fraction accounted for 19-39% of total COD in 24 wastewater samples were used in the experiments in reactor 2 (Figure 4.1 a). The average value of total COD was 627 (±81) g COD/m^3 including soluble COD = 194 (±38) g COD/m^3 vs. non-soluble (colloidal and particulate) COD = 433 (±73) g COD/m^3. For comparison, the average values of total and soluble COD determined (using the same method of Mamais et al., 1993) from the annual routine operating data were slightly deviating from the above concentrations, i.e. 594 and 172 g COD/m^3 (2007), and 715 and 192 g COD/m^3 (2008). At the "Dębogórze" WWTP, the soluble fraction accounted for 23-46% of total COD in 16 samples of the settled wastewater (Figure 4.1 b). The average value of total COD was 533 (±86) g COD/m^3 including soluble COD = 211 (±32) g COD/m^3 vs. non-soluble COD = 322 (±95) g COD/m^3.

In the earlier modelling study at these two plants (Mąkinia, 2006), similar measurements were carried out at both plants and those data are also shown in Figure 4.1 (a-b). The estimated ratio of biodegradable to non-biodegradable particulate (and colloidal) organic fractions varied in the range of 1.8-2.5 ("Wschód") and 1.4-1.5 ("Dębogórze") to fit the waste activated sludge (WAS) production. As a consequence, the ratios of (S_S/(S_S+X_S)) at the "Wschód" WWTP (0.32-0.40) fitted well into a typical range of 0.3-0.5 (Sahlstedt et al., 2003), whereas the corresponding values at the "Dębogórze" WWTP (0.50-0.54) slightly exceeded that range. Furthermore, Pagilla et al. (2008) reported results of the COD distribution in the primary effluents of both plants based on the filtration on different pore size filters. The dominant fraction originated from particulate organic compounds (>1.2 µm), which constituted 67 and 75 % of COD, respectively, at the "Wschód" and "Dębogórze" WWTPs (the latter plant was before the upgrade).

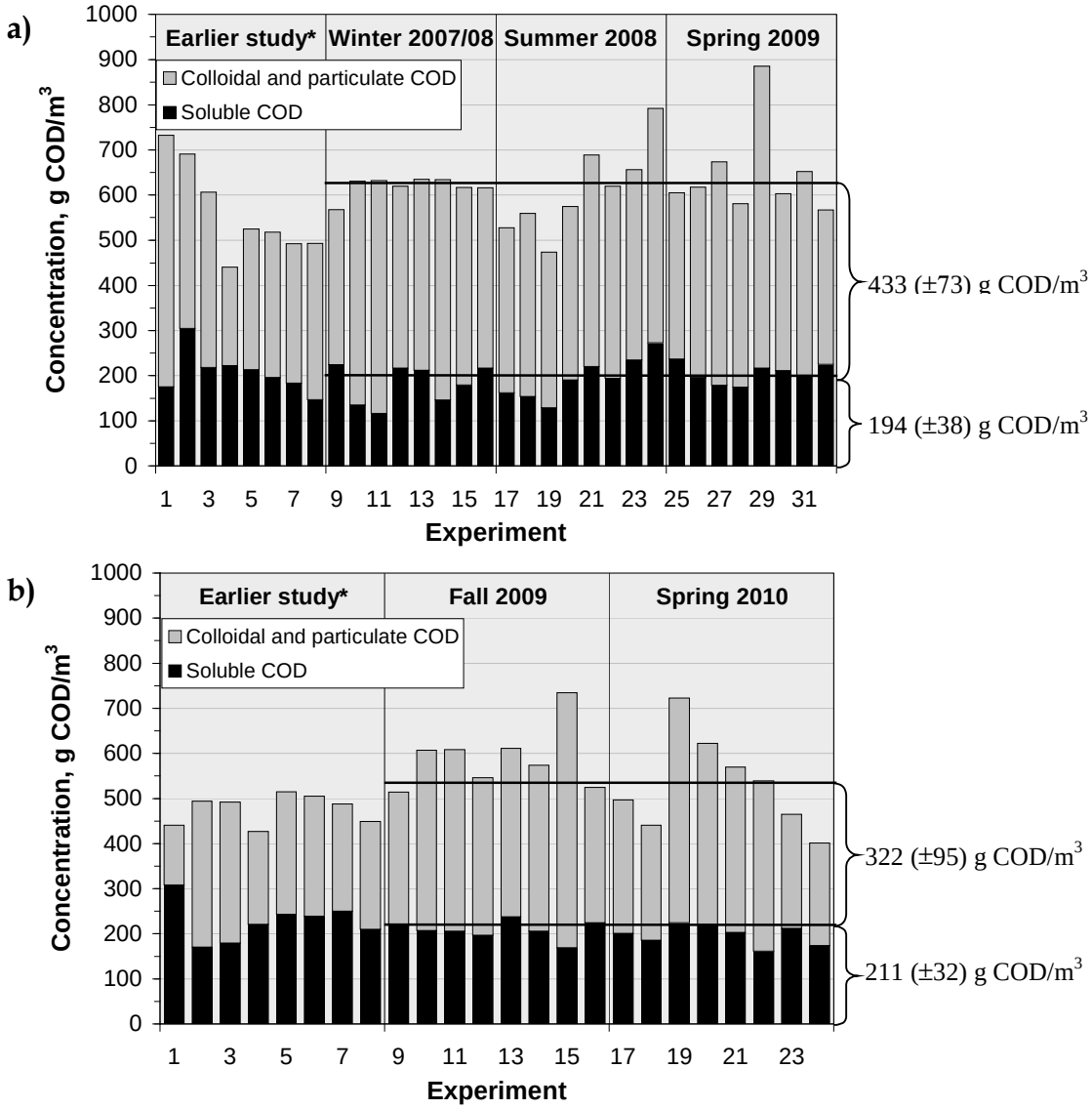


Figure 4.1. Measured fractions of COD (soluble vs. colloidal and particulate) in the 24-h settled wastewater samples from (a) the "Wschód" and (b) the "Dębogórze" WWTP (earlier study* – adapted from Mąkinia, 2006)

It should be noted that the effects of soluble, non-readily biodegradable organic compounds has better been described in the literature. Mamais et al. (1993) justified the rationale for their coagulation-flocculation method by the fact that readily biodegradable organic matter consists of simple molecules such as VFAs and low molecular weight carbohydrates that pass through the cell membrane and are metabolized immediately (see Section 2.1.3.1). Hu et al. (2002a) indeed confirmed that the COD quantified with this method corresponded closely with the low (<1 kDa) molecular weight fraction. Grady et al. (1999) noted that, for domestic wastewater, the coagulation-flocculation method gives results that correlate well with the conventional OUR tests. However, this statement remains in contradiction to the observations from other studies. Discrepancies between the physical-chemical method and respirometric measurements may especially be observed for samples

with a high content of industrial wastewater (Carrette et al., 2001). Even though colloidal particles, which normally pass the filter, are removed before filtration, the filtered COD is equal to the truly soluble COD rather than only to the readily biodegradable fraction (Xu and Hultman, 1996). The truly soluble COD is a sum of three fractions: inert, readily biodegradable and rapidly hydrolysable (Orhon and Cokgor, 1997). Due to the presence of rapidly hydrolysable COD in the membrane filtrate different respiration rates may occur for the same concentration of soluble COD (Sollfrank and Gujer, 1991). Ginestet et al. (2002) characterized samples of raw wastewater originating from seven French WWTPs. Respirometric measurements were carried out with samples of the raw, settled and "coagulated" (i.e. settled and precipitated with $FeCl_3$) wastewater. The latter group predominantly consisted of the readily hydrolysable fraction (37-90%), whereas the readily biodegradable and inert fractions accounted for 2-27% and 2-47% of soluble COD, respectively. Naidoo et al. (1998) examined the raw wastewater entering eight WWTPs in different European countries (see Section 2.1.3). The following four parameters were determined: total COD (COD_{tot}), COD after centrifugation (COD_{cent}), COD after filtration through a 0.45 µm (COD_f) and COD after coagulation and centrifugation ($COD_{coag+cent}$). In addition, the concentration of readily biodegradable substrate, $RBCOD_{NUR}$, was determined based on the specific NUR kinetics. The estimated contributions of $RBCOD_{NUR}$ ranged between 7 and 19% of total COD and these values were significantly lower than the soluble COD determined by the other methods, i.e. physical (filtration) or physical-chemical (coagulation-centrifugation). Koch et al. (2000) also found a poor correlation between soluble COD (after filtration on a 0.45 µm pore size filter) and S_S (estimated based on aerobic respiration tests) under Swiss conditions where the biodegradable fraction of wastewater primarily consists of X_S (due to short hydraulic retention times in the sewer systems).

4.2. Evaluation of the biochemical process rates based on the results of batch tests with biomass and wastewater from the studied plants

Table 4.1 and 4.2 contain a list of the ranges of all the specific rates, i.e. NURs, PRRs, anoxic/aerobic PURs and OURs, observed in the three kinds of batch experiments with the settled wastewater without pretreatment and after coagulation-flocculation. The obtained results in all the study sessions (winter, spring and summer-fall) from both studied plants are discussed in the following Sections. The full experimental data can be found in Appendix 2 and 3, respectively, for the "Wschód" and "Dębogórze" WWTP.

Table 4.1. Average values (± standard deviations) of the specific process rates observed during the batch experiments with the settled wastewater without pretreatment and pretreated with coagulation-flocculation method at the "Wschód" WWTP

Process rates		Winter session (Temp. range 11.8-14.2°C)		Spring session (Temp. range 14.8-16.7°C)		Summer session (Temp. range 18.9- 20.5°C)	
Type	**Unit**	**Settled wastewater**	**Pretreated settled wastewater**	**Settled wastewater**	**Pretreated settled wastewater**	**Settled wastewater**	**Pretreated settled wastewater**
				Conventional NUR test			
NUR1	**mg N/ gVSS·h**	4.4 (±0.92)	3.4 (±0.92)	5.3 (±0.14)	3.3 (±0.21)	3.2 (±0.35)	2.4 (±0.49)
NUR2		1.5 (±0.07)	1.1 (±0.07)	1.8 (±1.13)	1.2 (±0.28)	1.7 (±0.07)	1.0 (±0.01)
				PRR and anoxic/aerobic PUR test			
PRR	**mg P/ gVSS·h**	10.5 (±1.07)	8.1 (±2.28)	11.2 (±0.67)	13.1 (±0.91)	11.0 (±1.56)	10.2 (±2.56)
PUR_{Anoxic}	**mg P/ gVSS·h**	4.7 (± 0.35)	1.2 (±0.07)	6.8 (±0.78)	6.7 (±1.20)	5.7 (±1.48)	3.4 (±1.27)
NUR	**mg N/ gVSS·h**	1.8 (±0.21)	1.0 (±0.35)	3.0 (±0.71)	2.2 (±0.35)	2.3 (±0.07)	1.8 (±0.01)
$PUR_{Aerobic}$	**mg P/ gVSS·h**	8.6 (±1.41)	4.0 (±2.33)	11.4 (±0.07)	11.0 (±0.57)	6.1 (±0.42)	3.8 (±1.41)
OUR_{Max}	**mg O_2/ gVSS·h**	22.1 (±0.07)	18.3 (±0.07)	26.8 (±1.41)	25.9 (±0.42)	32.6 (±1.13)	29.7 (±1.89)
AUR	**mg N/ gVSS·h**	2.5 (±0.57)	2.3 (±0.71)	3.5 (±0.21)	3.5 (±0.01)	3.9 (±0.35)	3.8 (±0.78)
				Conventional OUR test			
OUR_{Max}	**mg O_2/ gVSS·h**	22.8 (±7.35)	16.9 (±7.99)	28.0 (±4.95)	20.8 (±6.51)	39.5 (±10.32)	27.8 (±7.99)
Y_H	**g COD/ g COD**	0.65 (±0.06)	0.66 (±0.04)	0.66 (±0.01)	0.66 (±0.05)	0.67 (±0.05)	0.62 (±0.06)

Table 4.2. Average values (± standard deviations) of the specific process rates observed during the batch experiments with the settled wastewater without pretreatment and pretreated with coagulation-flocculation method at the "Dębogórze" WWTP

Process rates		Fall session (Temp. range 16.0-17.8 °C)		Spring session (Temp. range 15.4-16.2 °C)	
Type	**Unit**	**Settled wastewater**	**Pretreated settled wastewater**	**Settled wastewater**	**Pretreated settled wastewater**
Conventional NUR test					
NUR1	**mg N/ gVSS·h**	3.6 (±0.64)	2.7 (±0.07)	3.2 (±0.57)	2.9 (±0.35)
NUR2		1.7 (±0.07)	1.1 (±0.21)	2.0 (±0.35)	1.2 (±0.07)
PRR and anoxic/aerobic PUR test					
PRR	**mg P/ gVSS·h**	9.6 (±0.44)	11.2 (±0.76)	7.9 (±0.85)	8.7 (±1.52)
PUR_{Anoxic}	**mg P/ gVSS·h**	2.0 (±0.07)	1.2 (±0.49)	1.8 (±0.14)	1.7 (±0.01)
NUR	**mg N/ gVSS·h**	1.8 (±0.42)	1.3 (±0.14)	2.3 (±0.49)	1.6 (±0.28)
$PUR_{Aerobic}$	**mg P/ gVSS·h**	5.9 (±1.06)	4.9 (±0.71)	5.6 (±0.35)	5.0 (±1.06)
OUR_{Max}	**mg O_2/ gVSS·h**	24.9 (±0.99)	21.8 (±1.34)	24.5 (±3.75)	19.1 (±1.70)
AUR	**mg N/ gVSS·h**	2.9 (±0.21)	2.9 (±0.14)	2.8 (±0.07)	3.1 (±0.07)
Conventional OUR test					
OUR_{Max}	**mg O_2/ gVSS·h**	20.4 (±1.98)	16.9 (±1.30)	21.8 (±3.46)	17.1 (±0.99)
Y_H	**g COD/ g COD**	0.69 (±0.01)	0.71 (±0.05)	0.68 (±0.02)	0.69 (±0.07)

4.2.1. Conventional NUR tests

Figure 4.2 (a-d) and 4.3 (a-d) illustrates sample results of the conventional NUR experiments carried out at the "Wschód" and "Dębogórze" WWTP. The average values of NURs from both studied plants are listed in Table 4.1 and 4.2, whereas Table 4.3 contains a review of the specific NURs reported in the literature for various types of activated sludge systems. In the experiments with the settled wastewater without the pretreatment, the observed double rates (NUR1 and NUR2) were associated with utilization of readily biodegradable (soluble S_S) organic compounds (only NUR1), and slowly biodegradable (soluble, colloidal and particulate X_S) organic compounds (both NUR1 and NUR2). The NUR1 values varied within the range 3.7-5.0, 5.2-5.4 and 2.9-3.4 g N/(kg VSS·h), respectively, during the winter, spring and summer sessions at the "Wschód" WWTP. The observed NURs were higher in the winter and spring study session (T = 11.8-14.2°C and T = 14.8-16.7°C) compared to the summer study session (T = 18.9-20.5°C). This higher rates at lower temperatures can be attributed to a different composition of the activated sludge

biomass (e.g. a higher content of denitrifying heterotrophs) and/or influent wastewater characteristics (e.g. a higher content of readily biodegradable compounds). In comparison, the NUR1 at the "Dębogórze" WWTP were varied only slightly during the fall and spring sessions within the range 3.1-4.0 and 2.9-3.4 g N/(kg VSS·h), respectively. The NUR2 values in the corresponding experiments varied within the range 1.4-1.5 (winter), 1.0-2.6 (spring) and 1.6-1.7 g N/(kg VSS·h) (summer) at the "Wschód" WWTP, whereas NUR2 values at the "Dębogórze" WWTP varied within the range 1.6-1.7 (fall) and 1.7-2.2 g N/(kg VSS·h) (spring).

When the pretreated samples of wastewater were used in the experiments, the observed NURs were associated with the utilization of the remaining soluble fraction. Consequently, the values of NUR1 and NUR2 were lower in comparison with the parallel tests with the settled wastewater. The NUR1 values varied within the range 2.7-4.0 (winter), 3.1-3.4 (spring) and 2.0-2.7 g N/(kg VSS·h) (summer), whereas the NUR2 values in the corresponding experiments were 1.0-1.1 (winter), 1.0-1.4 (spring) and 1.0 g N/(kg VSS·h) (summer) at the "Wschód" WWTP. In comparison, the NUR1 at the "Dębogórze" WWTP were varied only slightly during the sessions within the range 2.6-2.7 (fall) and 2.6-3.1 g N/(kg VSS·h) (spring), whereas the NUR2 values in the corresponding experiments were 0.9-1.2 (fall) and 1.1-1.2 g N/(kg VSS·h) (spring).

In addition to nitrate and COD, the behaviour of phosphate was also investigated during the conventional denitrification tests with the settled wastewater without the pretreatment and after coagulation-flocculation. An interesting behaviour of phosphate, similar at "Wschód" and "Dębogórze" WWTP, was observed in both reactors (see Figure 4.2 a-d and 4.3 a-d). At the beginning of the experiments, phosphate was released despite high concentrations of nitrate (in the range of approximately 10-20 g N/m^3) which, according the traditional opinion, inhibit that process (P release). The release continued until the readily biodegradable substrate was present in the solution, approximately 0.5-1.0 h and 0.5-1.5 h, respectively, during the summer-fall and spring study sessions at both plants. This behaviour was also observed in the winter study session at "Wschód" WWTP (data not shown).

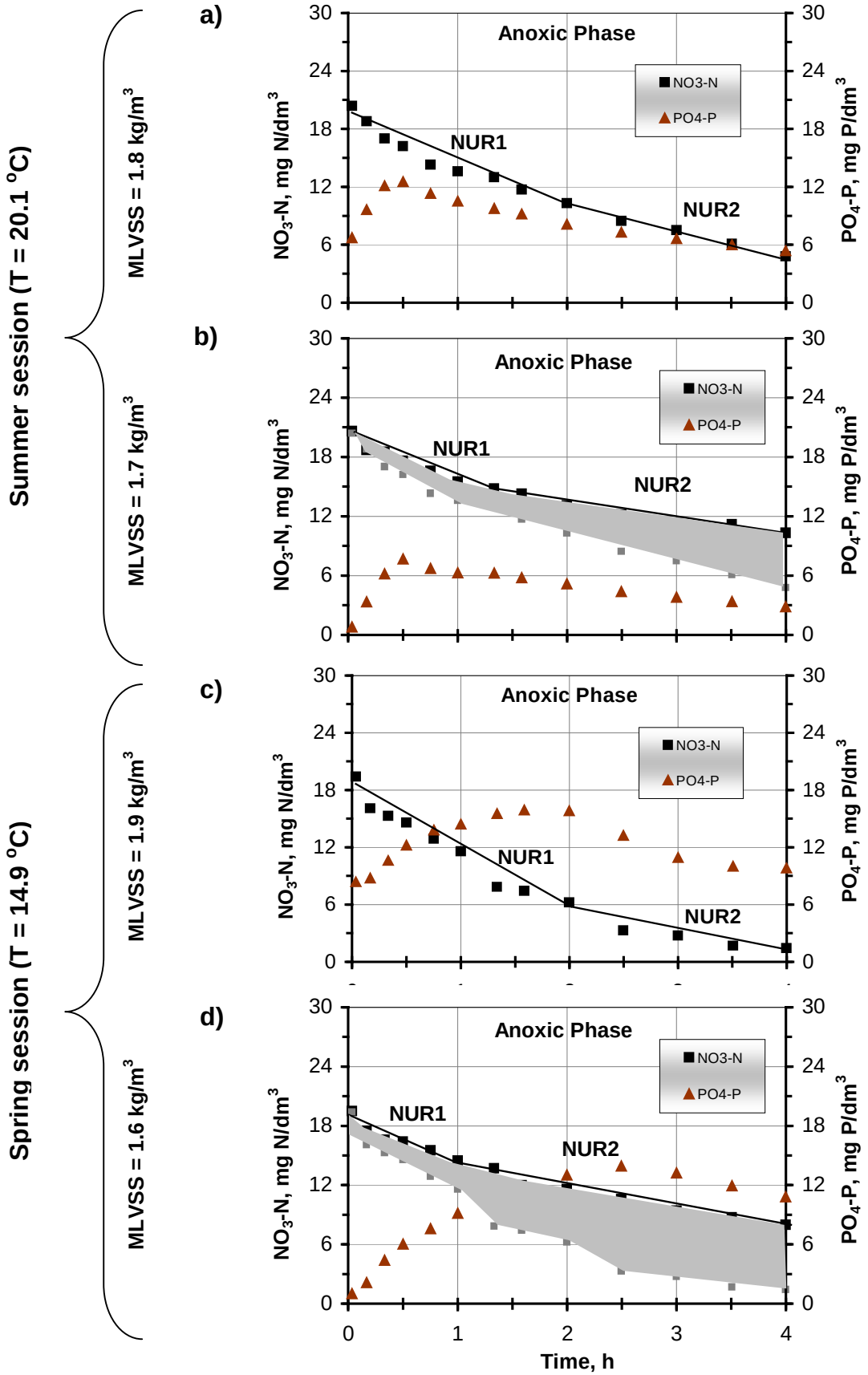


Figure 4.2. Sample results of the conventional NUR tests in two parallel reactors with mixed liquor from the "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

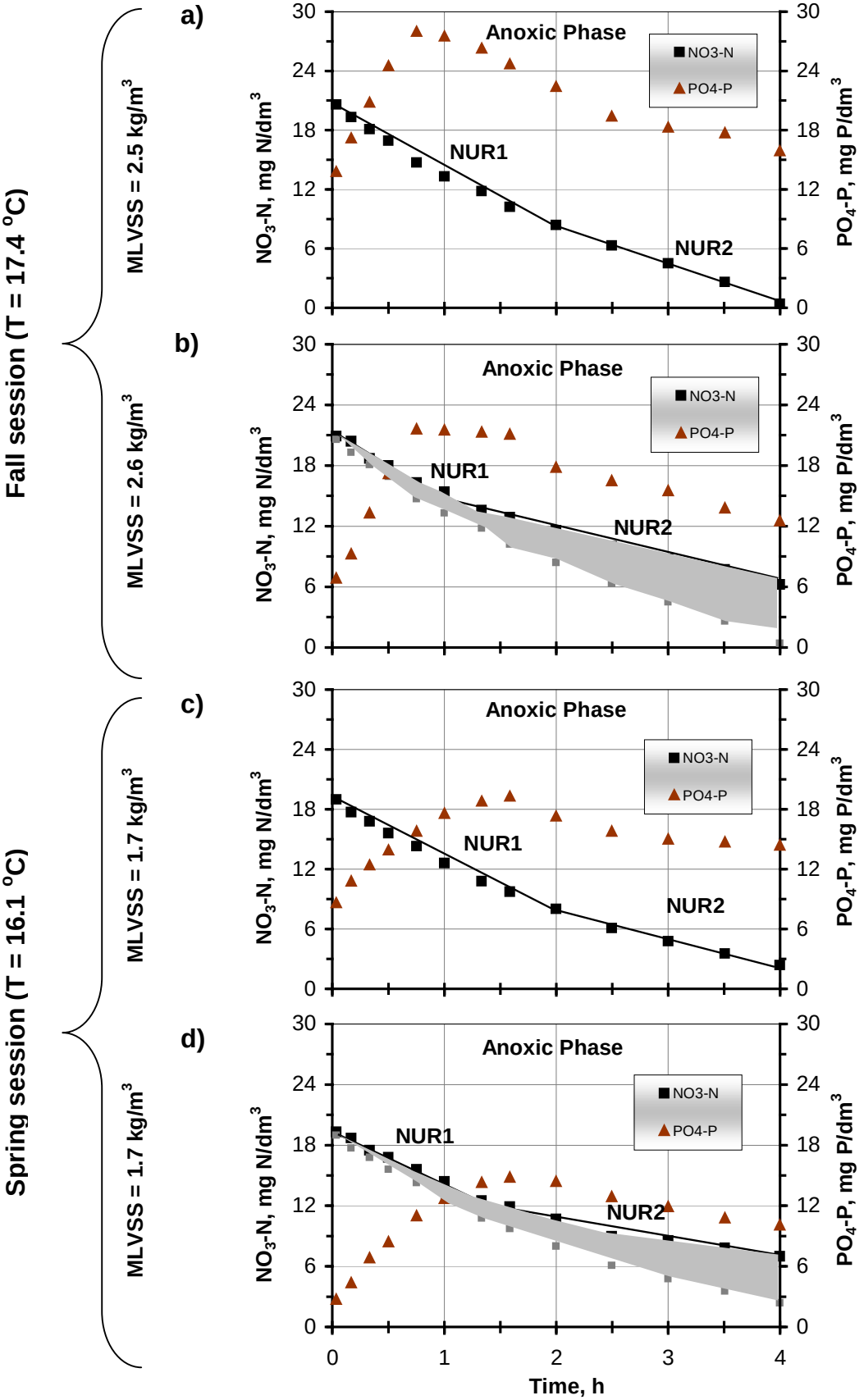


Figure 4.3. Sample results of the conventional NUR tests in two parallel reactors with mixed liquor from the "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

Summarizing, the calculations for estimating the denitrification efficiency with two different sample of the settled wastewater without pretreatment and after coagulation-flocculation, were performed in order to evaluate the impact of X_S. The experimental data revealed that two different rates, NUR1 and NUR2, were observed in terms of the available substrates in parallel batch reactors at the "Wschód" and "Dębogórze" WWTPs. The NUR1, accompanied by the decrease in COD concentrations, was associated with utilization of the readily biodegradable substrate in both batch reactors with settled wastewater without pretreatment and after coagulation-flocculation, whereas the NUR2 was associated with utilization of the slowly biodegradable substrate (without the decrease in measured COD concentrations). However, when the pretreated samples of wastewater were used in the experiments, the observed NURs were associated with the utilization of S_S and the remaining colloidal organic fraction (part of X_S). Consequently the removal of colloidal and particulate fractions by coagulation-flocculation, at both studied plants resulted in the reduced process rates of NUR1 and NUR2 in comparison with the parallel tests with the settled wastewater.

At the beginning of the experiments, NUR1 was similar at both reactors until the readily biodegradable substrate was present in the solution. The differences of denitrification efficiency were noted when the S_S was low and the remaining colloidal organic fraction couldn't keep process rates at the same level as in parallel reactor with the settled wastewater. The average NUR1 values varied in the range of 3.2-5.3 and 3.2-3.6 mg N/(g VSS·h), respectively, at the „Wschód" and „Dębogórze" WWTPs during all the study periods. The calculated ΔCOD:ΔN ratios associated with the NUR1 ranged from 5.1 to 10.4 („Wschód") and 6.4 to 8.1 („Dębogórze"). The average NUR2 values in the corresponding experiments varied in similar ranges, i.e 1.5-1.8 mg N/(g VSS·h) („Wschód") and 1.7-2.0 mg N/(g VSS·h) („Dębogórze"). The NUR1 and NUR2 values observed at both studied plants fit into the ranges reported by Naidoo et al. (1998) for similar experiments at eight municipal WWTPs in Europe. In that study, the NURs associated with utilization of the readily biodegradable and slowly biodegradable substrates remained in the range of 3.3-5.7 and 1.6-3.6 mg N/(g VSS·h), respectively.

When the pretreated samples of wastewater were used in the experiments, the observed lower NURs were associated with utilization of the remaining soluble fraction. The NUR1 values for the pretreated wastewater varied in the range 2.4-3.4 g N/(kg VSS·h) („Wschód") and 2.7-2.9 g N/(kg VSS·h) ("Dębogórze"), whereas the NUR2 values in the corresponding experiments were 1.0-1.2 g N/(kg VSS·h) ("Wschód"), and 1.1-1.2 g N/(kg VSS·h) ("Dębogórze"). The calculated ΔCOD:ΔN

ratios associated with the NUR1 ranged from 5.5 to 8.5 („Wschód") and 7.3 to 8.6 („Dębogórze"). Moreover, the removal of colloidal and particulate organic compounds resulted in decreasing the overall efficiency of NO_3-N removal during the 4-h tests by 21-37% and 24-28%, respectively, at the "Wschód" and "Dębogórze" WWTPs.

In comparison to the NURs based directly on the mass balance calculations over the anoxic compartment of the full-scale bioreactors estimated by Mąkinia et al., 2004 (i.e. 1.20-1.50 g N/(kg VSS·h) at the "Wschód" WWTP and 1.60-2.53 g N/(kg VSS·h) at the "Dębogórze" WWTP) were lower than the rates determined from batch tests. This exception was explained by the fact that the measurement was conducted at the end of the stabilization period when the denitrification process was reaching its optimal capacity at this plants after the winter nitrification loss (Mąkinia et al., 2004). However, all the rates measured at both studied plants are comparable with the NURs reported in the literature for various full-scale and laboratory scale activated sludge systems (Table 4.3). Henze et al. (1995) obtained denitrification rate between 1.0-5.0 g N/(kg VSS·h) at T=20°C with the municipal wastewater. For the same sample of wastewater Rodriguez et al. (2007) reported 4.3 g N/(kg VSS·h). In comparison the rates obtained for several agro-food by-products added to reactor with the municipal wastewater were considerably or only slightly lower, ranging from 2.0 g N/(kg VSS·h) (winery) to 4.1 g N/(kg VSS·h) (potato processing) at T = 20 °C. The authors explained this difference by of the lack of adaptation of the sludge to these substrates. Slightly higher rates, i.e. NUR1 = 4.9–6.3 g N/(kg VSS·h) and NUR2 = 2.2–2.8 g N/(kg VSS·h) at T=18.8-21.9°C were observed in the batch experiments with only the settled wastewater at the "Wschód" WWTP (Swinarski et al. 2007). These ranges are comparable, i.e. NUR1 = 2.8-6.1 g N/(kg VSS·h) and NUR2 = 1.4-3.1 g N/(kg VSS·h) at T=18.8-21.0°C, with the earlier studies conducted at the "Dębogórze" WWTP (Mąkinia et al., 2004), but significantly lower compared to the results obtained during the later study (Mąkinia et al., 2009), i.e. 1.4-2.0 g N/(kg VSS·h) at T=13.1-16.6°C. Slightly higher rates, i.e. NUR1 = 4.9-6.8 g N/(kg VSS·h) and NUR2 = 2.2-2.9 g N/(kg VSS·h) at T= 18.8–21.9°C, were observed during earlier studies conducted at the "Wschód" WWTP (Mąkinia et al., 2004). Considerably higher NURs were also measured in the anaerobic-anoxic system DEPHANOX (Bortone et al., 1996; Sorm et al., 1998) and two full-scale systems in the Czech Republic (Sorm et al., 1998).

Table 4.3. Review of the NURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g N/(kg VSS·h)	°C	
Kristensen et al. (1992)	Mixed liquor of tapwater and RAS with carbon in excess*, acetate** or hydrolysate***	Pilot-scale plant	1.3 – 1.7 * 3.4 – 4.8 ** 4.0 – 5.6 ***	20	All values converted to temp. 20 °C
	Mixed liquor of tapwater and RAS with carbon in excess*, acetate* or hydrolysate***	Full-scale plant	0.4 – 2.2 * 1.1 – 7.4 ** 1.4 – 7.9 ***	20	All values converted to temp. 20 °C
Bortone et al. (1996)	DEPHANOX fed with real wastewater	Lab-scale	5.3 – 14.4	20	Acetate
Chang and Hao (1996)	Anaerobic-aerobic SBR fed with a mixture of real wastewater and acetate	Lab-scale	0.4	20±2	End of the aerobic phase
Nyberg et al. (1996)	Single activated sludge fed with a wastewater	Lab-scale	3 10	n.d.	Methanol Ethanol
van Loosdrecht et al. (1997b)	Batch test fed with MUCT biomass and municipal wastewater without* and with primery clarification**	Full-scale	6.2 – 7.0 * 6.2 **	25	
Artan et al. (1998)	Anaerobic-aerobic SBR fed with synthetic wastewater with acetate and glucose	Lab-scale	1.5 2.0 – 3.7 8.0	n.d.	End of the aerobic phase Glucose Acetate
Sorm et al. (1998)	DEPHANOX fed with real wastewater	Lab-scale	23.7	20	
	A^2/O	Full-scale	25.3 – 28.6	20	
	A/O	Full-scale	8.3 – 9.8	20	
Aravinthan et al. (2000)	Autoclaved, alkaline, acid and combined solubilization methods with the excess sludge from WWTP	Lab-scale	7.7 – 10.4	15	Sludge hydrolysate
Kim et al. (2001)	Anaerobic-aerobic SBR fed with a mixture of real wastewater and acetate	Lab-scale	0.7 – 1.4	n.d.	End of the aerobic phase
Louzeiro et al. (2002)	SBR fed with real wastewater	Full-scale	0.8	n.d.	Methanol
Carrera et al. (2003)	The batch experiments with fresh biomass from the denitrifying reactor of the pilot plant (two-sludge system)	Lab-scale	0.48 ±0.22 1.06 ±0.12 1.7 ±0.24 2.9 ±0.72 4.3 ±1.44 6.7 ±0.72	6 ±0.5 8 ±0.5 10 ±0.5 15 ±0.5 20 ±0.5 25 ±0.5	Mixture of 60% methanol, 10% acetone, 10% isopropilic alcohol and 20% water
Andreottola et al. (2003)	Intermittent aeration	Full-scale	3.9 ±1.00	n.d.	
	Pre-denitrification	Full-scale	3.3 ±1.04	n.d.	

Table 4.3 (continued). Review of the NURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g N/(kg VSS·h)	°C	
Foglar and Briski (2003)	Stirred reactor fed with synthetic wastewater, continuous-flow	Lab-scale	4.35	n.d.	Methanol
Freitas et al. (2004)	Anaerobic-aerobic SBR fed with synthetic wastewater	Lab-scale	3.4 – 6.6	20-30	Acetate
Cappai et al. (2004)	Denitrification rate tests fed with mixing municipal wastewater with the industrial effluents	Lab-scale	1.1 2.7 3.3	n.d.	Not added Beet-sugar processing Cream prod. effluent
Kosińska (2005)	NUR batch tests fed with municipal wastewater	Lab-scale	1.45 * 2.62 **	14.7-18.5	Without* or with potato processing by-products**
Mąkinia (2006)	Conventional NUR batch tests fed with settled wastewater	Lab-scale	2.9-8.7	15-20	
Peng et al. (2007)	NUR batch tests fed with wastewater, pre-denitrification system	Lab-scale	0.74 3.2 9.6 12	n.d.	Starch waste Methanol Ethanol Acetate
Cherchi et al. (2008)	Anoxic-aerobic SBR fed with a acclimated sludge & synthetic wastewater	Lab-scale	6.37 ±3.6 6.07 ±0.7 13.6 ±1.86	20-23	MicroC™ Methanol Acetate
	WWTPs acclimated* and not-acclimated** sludge	Full-scale	4.72 ±0.48 * 4.34 ±0.52 **	20	MicroC™ added to post-denitrification stage
Swinarski et al. (2009b)	Conventional NUR batch tests fed with settled wastewater and food industry by-products	Full -scale	4.8 – 5.1 2.4 – 6.0 4.0 – 6.0	23.1-24 19.7-22.7 21.4-23	Distillery wastewater Brewery wastewater Fish-pickling process
	PRR and anoxic PUR batch tests fed with settled wastewater with or without food industry by-products	Full -scale	2.0 – 3.4 3.7 – 4.3 1.1 – 1.3 3.7 – 4.8	21.8-24.3 21.7-21.8 20.3-20.6 26.1-27.2	Not added Distillery wastewater Brewery wastewater Fish-pickling process

As it was mentioned, the behaviour of phosphate was also investigated during the experiments. The P release continued, despite high concentrations of nitrate (10-20 g N/m^3) at the beginning of the experiments, until the S_S was present in the solution (approximately 0.5-2.5 h) depending on the study period (temperature), biomass and initial COD concentrations in the batch reactors. A very similar behaviour of phosphate was observed by Brdjanovic et al. (2000) in the conventional NUR test with a full-scale plant mixed liquor and acetate as a carbon source. In that study, a significant amount of phosphate was released (from approximately 2 g P/m^3 to almost 10 g P/m^3) during the first hour of the test until a complete consumption of acetate. Simultaneously, the NO_3-N concentration decreased from approximately 25 g N/m^3 to 10 g N/m^3. These observations are, in accordance with the conclusion of Yuan and Oleszkiewicz (2008), that phosphate release continues as long as the substrate is present regardless of NO_3-N concentrations in the "anaerobic" phase.

4.2.2. PRR and anoxic/aerobic PUR tests

Sample results of the combined PRR and anoxic/aerobic PUR experiments carried out at the "Wschód" and "Dębogórze" WWTP are shown in Figure 4.4-4.5 (a-d) and Figure 4.6-4.7 (a-d). The additional sample results of the AUR measurment during the PRR and aerobic PUR tests are illustrated in Figure 4.8-4.9 (a-d). The average values of PRR and anoxic/aerobic PUR processes from both studied plants are listed in Table 4.1 and 4.2, whereas Table 4.3 and Table 4.4-4.5 contain a review of the specific PRR and anoxic/aerobic PUR reported in the literature for various types of activated sludge systems. Moreover Table 4.6 presents a review of the specific AUR reported in the literature.

In the experiments with the settled wastewater from the "Wschód" WWTP (Table 4.1), the lowest average PRR were obtained in the winter study session (10.5 g P/(kg VSS·h)) at T= 11.8-14.2 °C, while the rates in the spring and summer study sessions were highly comparable (approximately 11.0 g P/(kg VSS·h)) at temperature 14.8-16.7 ˚C and 18.9-20.5 ˚C, respectively. In comparison, during the similar experiments from the "Dębogórze" WWTP (Table 4.2), the lowest average PRR were obtained in the spring study session (7.9 g P/(kg VSS·h) at T= 15.4-16.2 ˚C), while the rates in the fall study sessions were higher (9.6 g P/(kg VSS·h) at T = 16.0-17.8 ˚C). The effect of temperature had no or only minor significant on the kinetics of phosphate release in case of the "Wschód" and "Dębogórze" WWTP, respectively. The coagulation-flocculation had also no explicit impact on the PRRs as the observed rates in the parallel reactors varied in the range of maximum ±20%. The observation during the

PRR tests with the sample of pretreated wastewater showed lower values by approximately 10-20% for the summer and winter study session at "Wschód" WWTP. The opposit behaviour of P released (higher values ±20% in the pretreated sample) was observed during spring and fall study sessions at both studied plants. However, the average amounts of P released per MLVSS were similar in both reactors, i.e. 13.7 vs. 14.0 g P/g MLVSS ("Wschód") and 10.8 vs. 11.3 g P/g MLVSS ("Dębogórze").

The PRR results are comparable with the literature data (6-12.3 P/(kg VSS·h)) reported by Mąkinia (2006) for a number of different full-scale, pilot-scale and lab-scale BNR systems, but significantly lower compared to the results obtained during the previous study (Mąkinia et al., 2004), i.e. 15.1-21.4 g P/(kg VSS·h). The stored P was not completely released at the end of the anaerobic phase as in the earlier study (Mąkinia, 2006) the amounts of P released in the presence of acetate were higher by 40-70% compared to the experiments with the settled wastewater. It thus appears that anaerobic hydrolysis of X_S generates insignificant amounts of the substrate for maintenance of PAOs under anaerobic conditions. However, he PRRs observed at both plants remain in the range of literature data (4.4-18.8 mg P/(g VSS·h)) reported in similar experiments carried out with the mixed liquor from full-scale BNR systems (Kuba et al., 1997; Sorm et al., 1998; Tuncal et al., 2009; Puig et al., 2010) and full-scale pilot plants performing EBPR (Petersen et al., 1998; Tykesson et al., 2002). In the study of Tuncal et al. (2009), the PRRs were 10.8 and 16.2 mg P/(g VSS·h), respectively, in the line with and without primary clarification. Puig et al. (2010) found the opposite effect of by-passing primary clarifiers. The observed PRR decreased from 18.8 mg P/(g VSS·h) during the standard operation to 13.6 mg P/(g VSS·h) during by-passing the primary clarifiers. The rates measured at both plants were also comparable with the data from a full-scale UCT system (Kuba et al., 1997) and several laboratory scale anaerobic-aerobic SBRs treating either real wastewater enriched with acetate (Chang and Hao, 1996) or synthetic wastewater containing VFAs (Kim et al., 2001; Serafim et al., 2002; Freitas et al., 2004).

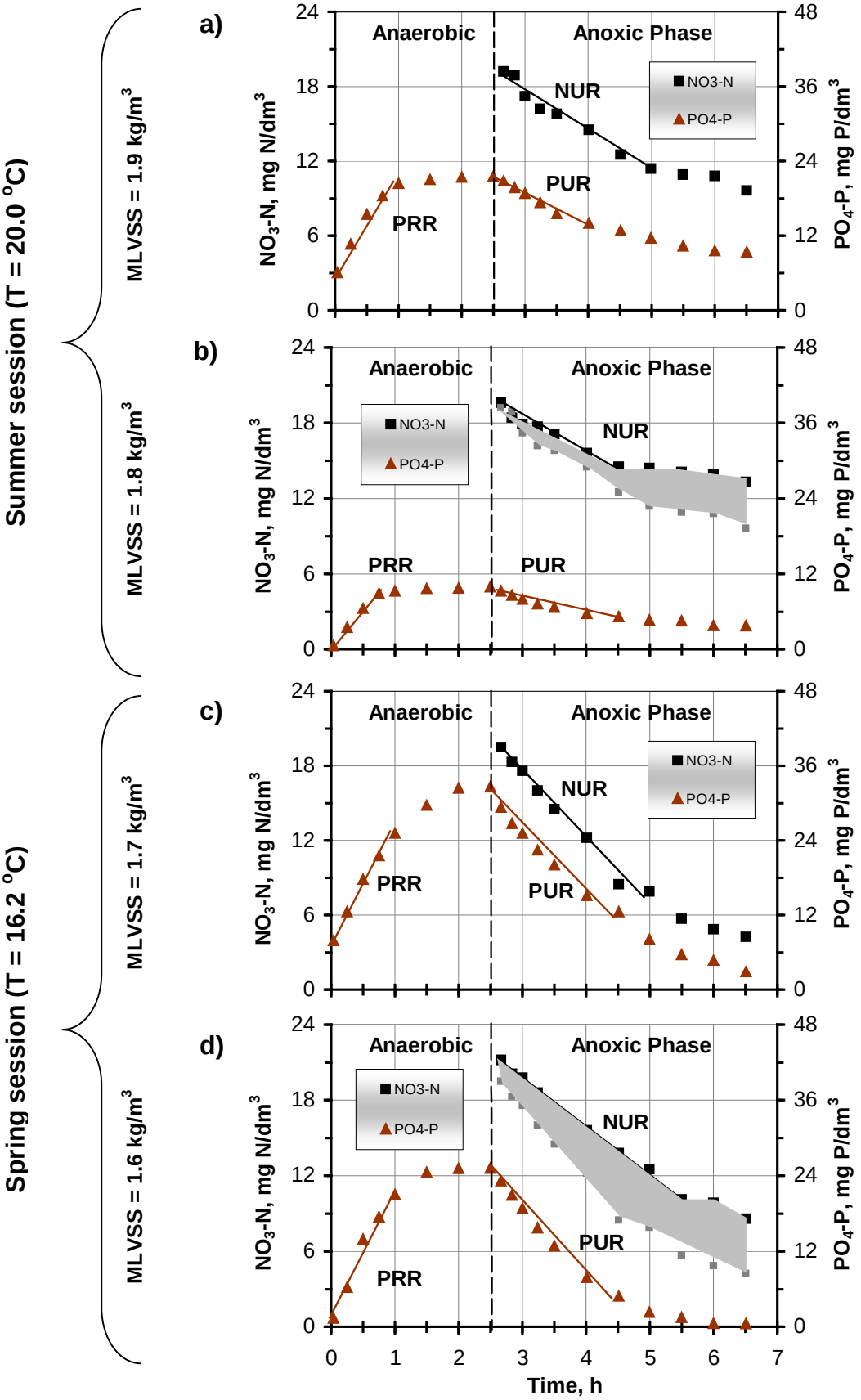


Figure 4.4. Sample results of the PRR and anoxic PUR tests in two parallel reactors with mixed liquor from the "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

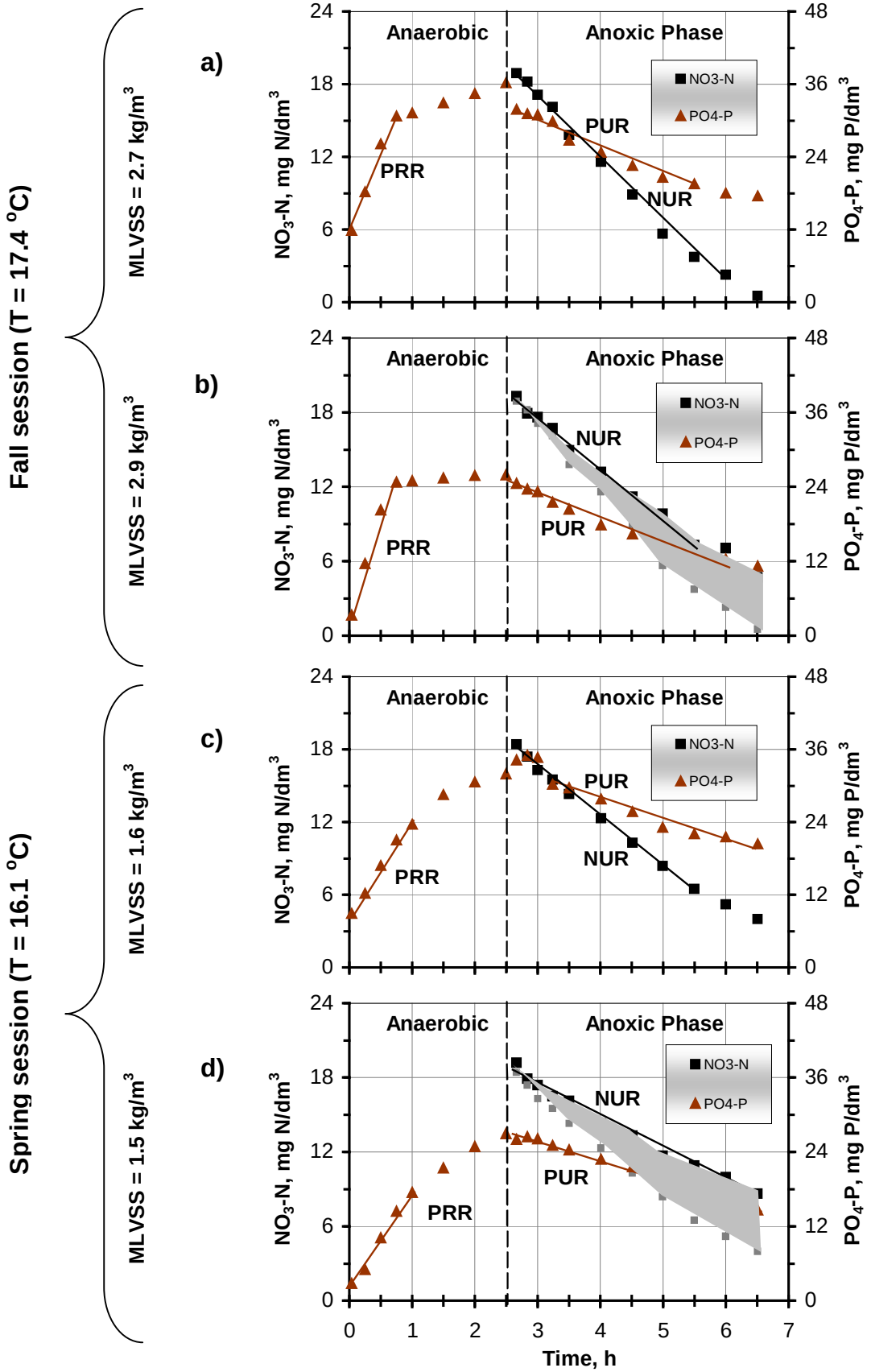


Figure 4.5. Sample results of the PRR and anoxic PUR tests in two parallel reactors with mixed liquor from the "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

Table 4.4. Review of PRRs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g P/(kg VSS·h)	°C	
Carucci et al. (1995)	Anaerobic-aerobic SBR fed with glucose	Lab-scale	6	20 ±1	
Chang and Hao (1996)	Anaerobic-aerobic SBR fed with a mixture of wastewater and acetate	Lab-scale	8 – 18	20 ±2	
Kuba et al. (1997)	UCT	Full-scale	18	20	
van Loosdrecht et al. (1997b)	Batch test fed with MUCT biomass and municipal wastewater	Full-scale	9 * – 19 **	25	Without* or with primery clarification**
Wachtmeister et al. (1997)	SBR operated under aanaerobic-aerobic (A/O) or anaerobic-anoxic (A2) conditions	Full-scale	10 – 70	25	Synthetic wastewater containing acetic acid
Artan et al. (1998)	Anaerobic-aerobic SBR fed with synthetic wastewater containing acetate and glucose	Lab-scale	15 – 37	n.d.	Lower values in the presence of nitrates
Petersen et al. (1998)	BIO-DENIPHO	Pilot-scale	8.6	21 ±1	
Sorm et al. (1998)	DEPHANOX fed with real wastewater	Lab-scale	12.3	20	
	A^2/O	Full-scale	9.9 – 12.0	20	
	A/O	Full-scale	4.4 – 7.0	20	
Kim et al. (2001)	Anaerobic-aerobic SBR fed with a mixture of real wastewater and acetate	Lab-scale	16.1 – 20.6	n.d.	
Levantesi et al. (2002)	Anaerobic-anoxic SBR fed with a mixture of VFAs	Lab-scale	43.4 ±2.73	n.d.	
Tykesson et al. (2002)	A/O	Pilot-scale	6.9	n.d.	
Serafim et al. (2002)	Anaerobic-aerobic SBR fed with a mixture of VFAs	Lab-scale	15.6 – 30.0	22	
Freitas et al. (2004)	Anaerobic-aerobic SBR fed with synthetic wastewater and acetate	Lab-scale	14.6 – 24.3	20-30	
Kosińska (2005)	PRR batch tests fed with municipal wastewater	Lab-scale	2.1 – 4.54 * 4.82 – 6.12 **	14.8-17.5	Without* or with potato by-products** addition
Puig et al. (2010)	Batch tests with MUCT biomass (pre-settling or raw influent by-passed directly into the anaerobic reactor)	Full-scale	13.6 – 18.8	20 ±0.5	Lower values with pre-settling

The average ratio of PO_4-P released to COD utilized (Y_{PO4}), which was measured during the initial 60 min of the anaerobic phase, ranged from 0.29 to 0.33 g P/g COD at the "Wschód" WWTP and from 0.34 to 0.59 g P/g COD at the "Dębogórze" WWTP for the experiments with the settled wastewater. For comparison, similar values were obtained for the experiments with the sample after coagulation-flocculation, i.e. 0.30-0.35 and 0.21-0.56 g P/g COD at the "Wschód" and "Dębogórze" WWTP, respectively. These values are only slightly different from a typical range (0.35-0.5) reported for activated sludge systems. Lower Y_{PO4} can be justified by either long SRTs or the presence of glycogen accumulating organisms (GAOs) (Brdjanovic et al., 2000). Furthermore, the type of substrate is another factor influencing Y_{PO4}. In the earlier study (Mąkinia, 2006), the ratios for acetate were significantly higher (by approximately 35-40%) in comparison with the settled wastewater. Ubukata (2005) also found that the organic compounds present in real wastewater contribute to a different phosphate release than acetate. The discrepancy observed at both studied plants for the Y_{PO4} coefficients in settled wastewater without pretreatment and after coagulation-flocculation with different sources of substrates may imply that X_S is a representative substrate for PAOs (see Section 2.2.4).

The results from PRR and anoxic/aerobic PUR (Test 2 and Test 3), carried out with the settled wastewater without pretreatment and after coagulation-flocculation, showed different processes rates between parallel batch reactors at both studied plants. The maximum PURs, observed at both plants under aerobic conditions (during the initial 60 min of the test), varied in the ranges of 6.1-11.4 and 5.6-5.9 mg P/(g VSS·h) in the settled wastewater without pretreatment vs. 3.8-11.0 and 4.9-5.0 mg P/(g VSS·h) in the wastewater after coagulation-flocculation, respectively, at the "Wschód" WWTP and "Dębogórze" WWTP. The anoxic PURs were significantly lower, i.e. 4.7-6.8 and 1.8-2.0 mg P/(g VSS·h) in the settled wastewater without pretreatment vs. 1.2-6.7 and 1.2-1.7 mg P/(g VSS·h) in the wastewater after coagulation-flocculation (Table 4.1-4.2). The anoxic PURs correlated strongly (r^2 = 0.96-0.99) with the NURs measured during the parallel experiments with the settled wastewater. The calculated ratios of utilized nitrate and phosphate varied in the range of 0.51-0.78 g N/g P and 0.74-1.41 g N/g P in the settled wastewater without pretreatment vs. 0.47-1.74 g N/g P and 0.83-1.01 g N/g P in the wastewater after coagulation-flocculation, respectively, at the "Wschód" WWTP and the "Dębogórze" WWTP. For comparison, Tuncal et al. (2009) found lower N/P ratios which were 0.4 and 0.5 g N/g P in the line without and with primary clarification, respectively.

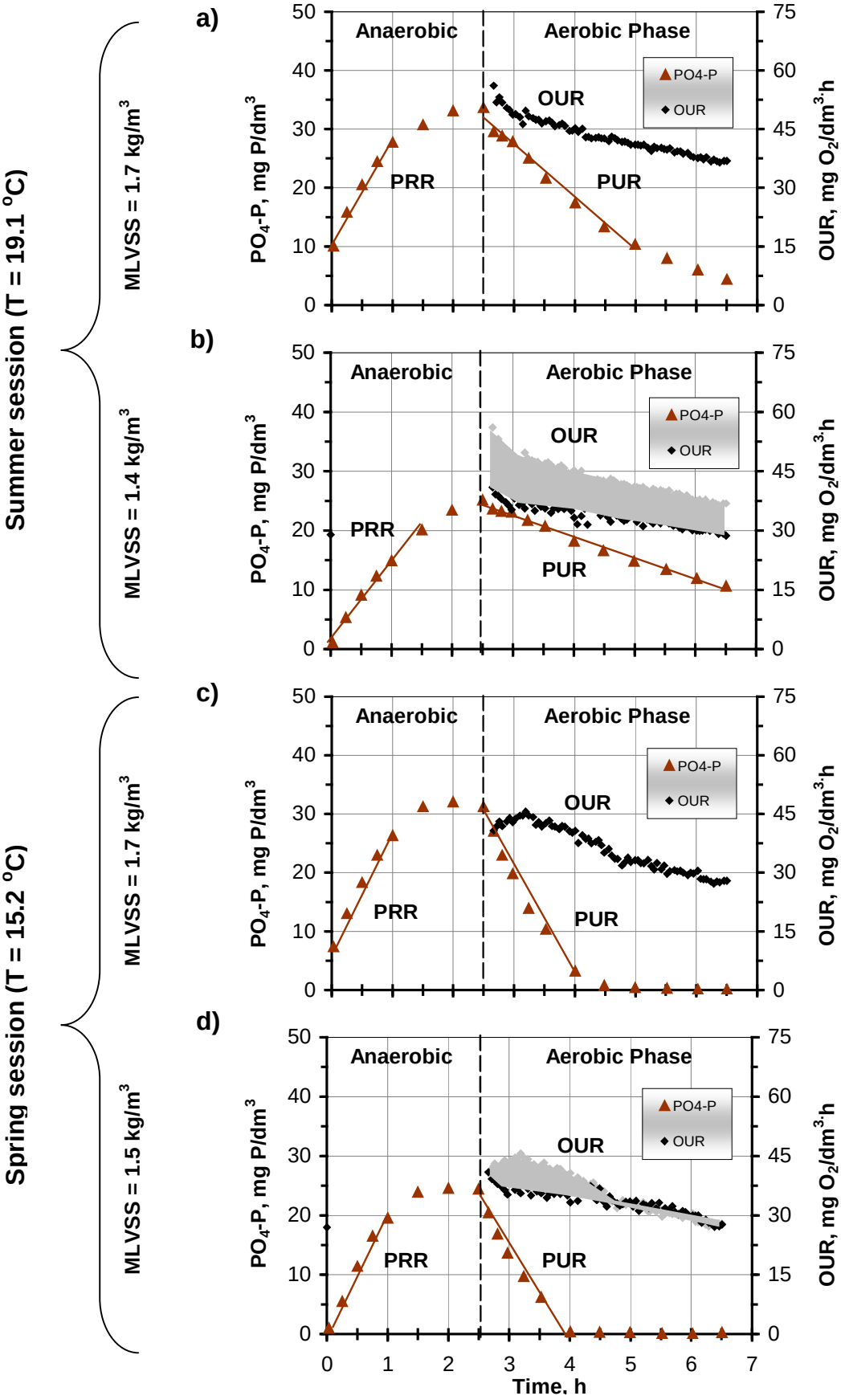


Figure 4.6. Sample results of the PRR and aerobic PUR tests in two parallel reactors with mixed liquor from the "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

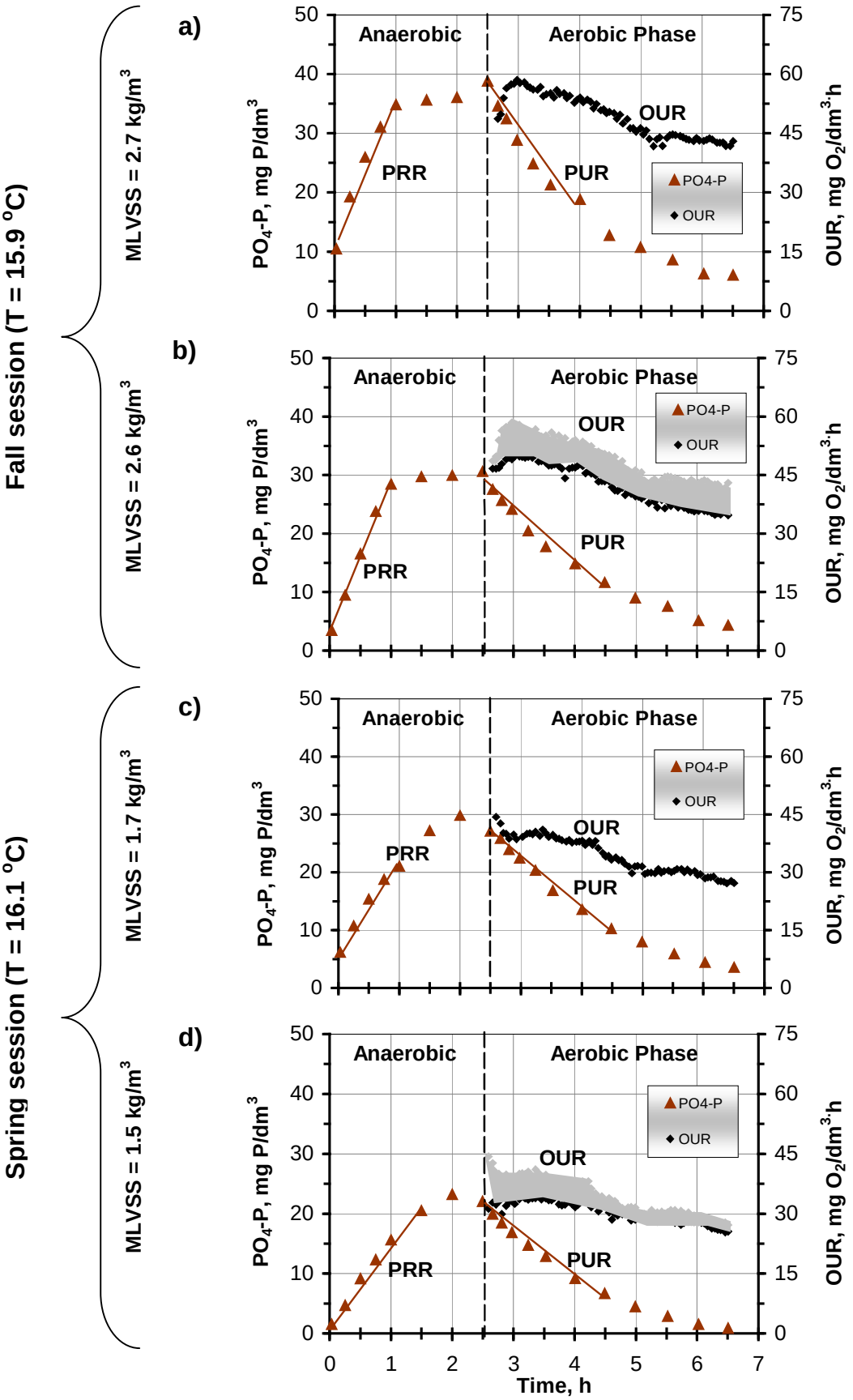


Figure 4.7. Sample results of the PRR and aerobic PUR tests in two parallel reactors with mixed liquor from the "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

The capabilities of denitrifying PAOs cannot be significant at both studied plants in terms of contribution to the overall utilization of nitrate. Mąkinia (2006) found at both plants that the denitrification rates associated with the anoxic storage of polyphosphate and the anoxic growth of PAOs constituted 16-21% of the denitrification rates associated with the anoxic activity of "ordinary" heterotrophs. This range was very close to the earlier findings of Hu et al. (2002b) that the specific denitrification rate of PAOs on internally stored PHB was only about 20% of the rate of the "ordinary" heterotrophs on Xs.

The literature data (anoxic/aerobic PURs) for various laboratory scale, pilot-scale and full-scale systems are summarized in Table 4.5 and 4.6. The rates measured at both studied plants are comparable with the data originating from a number of laboratory and full scale anoxic PURs experiments, i.e., DEPHANOX systems (Bortone et al., 1996), UCT systems (Kuba et al., 1997) and Hybrid process (You et al., 2001). Similar the aerobic PURs are comparable with a number of laboratory anaerobic-aerobic SBRs (Chang and Hao, 1996; Kim et al., 2001; Shoji et al., 2003; Freitas et al., 2004) and DEPHANOX systems (Bortone et al., 1996; Sorm et al., 1998).

Table 4.5. Review of anoxic PURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g P/(kg VSS·h)	°C	
Bortone et al. (1996)	DEPHANOX fed with real wastewater	Lab-scale	2.2 – 6.0	20	
Kuba et al. (1997)	UCT	Full-scale	6.0	20	
van Loosdrecht et al. (1997b)	Batch test fed with MUCT biomass and municipal wastewater	Full-scale	2.3 * – 5.9 **	25	Without* or with primery clarification**
Sorm et al. (1998)	DEPHANOX fed with real wastewater	Lab-scale	11.7	20	
	A/O	Full-scale	1.9 – 2.8	20	
	A^2/O	Full-scale	12.4 – 13.0	20	
Brdjanovic et al. (2000)	Phostrip	Full-scale	1.7	20	
You et al. (2001)	Hybrid process (RBC added to the aerobic zone of A_2O)	Lab-scale	0.32 – 0.39	20	Low PHA content, no acetate added
	Hybrid process (RBC added to the aerobic zone of A_2O)	Lab-scale	1.4 – 3.8	20	High PHA content, no acetate added
	Hybrid process (RBC added to the aerobic zone of A_2O)	Lab-scale	0.41 – 0.86	20	High PHA content, acetate added
Shoji et al. (2003)	A_2N	Lab-scale	5.4 – 13.3	20	
Freitas et al. (2004)	Ana-aerobic SBR fed with synthetic wastewater	Lab-scale	2.7 – 12.0	20-30	Acetate added

Table 4.6. Review of aerobic PURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g P/(kg VSS·h)	°C	
Carucci et al. (1995)	Anaerobic-aerobic SBR	Lab-scale	4.1	20 ±1	Glucose added
Bortone et al. (1996)	DEPHANOX fed with real wastewater	Lab-scale	6.0 – 14.4	20	
Chang and Hao (1996)	Anaerobic-aerobic SBR fed with a mixture of real wastewater	Lab-scale	7 – 11	20 ±2	Acetate added
Kuba et al. (1997)	UCT	Full-scale	13.0	20	
van Loosdrecht et al. (1997b)	Batch test fed with MUCT biomass and municipal wastewater	Full-scale	5.4 * – 13.2 **	25	Without* or with primery clarification**
Sorm et al. (1998)	DEPHANOX fed with real wastewater	Lab-scale	16.9	20	
	A/O	Full-scale	15.1 – 30.0	20	
	A^2/O	Full-scale	16.1 – 17.9	20	
Petersen et al. (1998)	BIO-DENIPHO	Pilot-scale	1.2 – 9.4	21 ±1	Function of the PHB concentr.
Helness and Odegaard (1998)	Bed biofilm reactor operated as a SBR	Lab-scale	8.4	n.d.	
Brdjanovic et al. (2000)	Phostrip	Full-scale	2.2	20	
Kim et al. (2001)	Anaerobic-aerobic SBR fed with a mixture of real wastewater and acetate	Lab-scale	10.9 – 12.1	n.d.	
Levantesi et al. (2002)	Anaerobic-anoxic SBR fed with mixture of VFAs	Lab-scale	43.0 ±3.43	n.d.	
Serafim et al. (2002)	Anaerobic-aerobic SBR fed with mixture of VFAs	Lab-scale	25.8 – 48.0	22	
Shoji et al. (2003)	A_2N	Lab-scale	8.2 – 22.4	20	
Freitas et al. (2004)	Anaerobic-aerobic SBR fed with synthetic wastewater containing acetate	Lab-scale	12.2 – 21.6 2.4 – 9.2	20-30	Ana-aer Ana-anox-aer
Zima et al. (2009)	Batch tests fed with settled wastewater	Full-scale	13.5	20	
Puig et al. (2010)	Batch tests with MUCT biomass (pre-settling or raw influent by-passed directly into the anaerobic reactor)	Full-scale	7.5-8.8	20 ±0.5	Lower values with pre-settling

For comparison, the anoxic and aerobic PURs reported for full-scale BNR activated sludge systems varied in the range of 1.9-13 and 3.6-30 mg P/(g VSS·h), respectively (Kuba et al., 1997; Sorm et al., 1998; Tuncal et al., 2009; Puig et al., 2010). In the studies of Tuncal et al. (2009) and Puig et al. (2010), the observed anoxic and aerobic PURs were approximately equal, however, the effects of primary clarification were opposite in those studies. In the former study, a significant increase of the PURs (by 70%) was observed in the line without primary clarification compared to the line with primary clarification, whereas by-passing raw wastewater to the biological stage in the latter study resulted in a small decrease of the PURs (by 15%), likely due to the shorter SRT and higher inert fraction in the sludge.

The calculations for estimating the denitrification efficiency of BNR systems performing EBPR different from those for purely denitrifying activated sludge plants for two reasons: virtually all the readily biodegradable influent COD is used in the anaerobic reactor and a substantial fraction of PAOs can denitrify on their stored substrate (Koch et al., 2001; Ekama and Wentzel, 1999a). In order to evaluate the impact of the anaerobic zone on denitrification, the measurements of NURs were also carried out during the PRR and anoxic PUR tests with both samples (settled wastewater without pretreatment and after coagulation-flocculation). In the experiments with the pretreated wastewater, lower values of the NURs (on average 30% vs. 24%) at "Wschód" and "Dębogórze" WWTP were observed under the anoxic conditions. For comparison, Tuncal et al. (2009) reported a similar (25%) reduction for the NUR measured in the line with primary clarification compared to the line without primary clarification (1.8 vs. 2.4 mg N/(g VSS·h)). The calculated specific NURs originating from a number of laboratory and full scale experiments are listed in Table 4.3. The anoxic PURs correlated strongly with the NURs measured in parallel during the experiments with the settled wastewater. The calculated ratios of the utilized nitrate and phosphate in the settled wastewater without pretreatment and after coagulation-flocculation varied within the range of 0.56-0.84 and 0.74-1.76 g N/g P at the Wschód WWTP vs. 0.95-1.99 and 0.93-1.09 g N/g P and the "Dębogórze" WWTP, respectively. For comparison, the same ratios of the utilized nitrate and phosphate obtained by Mąkinia (2006) with the settled wastewater varied within the range of 1.03-1.17 g N/g P (r^2 = 0.97-0.98) and 0.99-1.04 g N/g P (r^2 = 0.84-0.87) at the "Wschód" WWTP and "Dębogórze" WWTP, respectively.

The additional AUR and OUR_{max} measurements under the aerobic condition during the PRR and aerobic PUR test were carried out. The average difference of AUR and OUR_{max} for the pretreated wastewater was up to 10 and 30%, respectively, at both studied plants (Table 4.1-4.2). However, during a AUR experiments most of the time

hardly any differences in case of the process kinetics of ammonia uptake were observed in the parallel reactor with the pretreated sample. This suggests that chemical precipitation and removal of colloidal and particulate fractions had no or only minor effect on nitrification (Figure 4.8 and 4.9 a-d). It should be noted that a complete ammonia oxidation was not achieved in the both batch reactors. The nitrification rates listed in Table 4.1-4.2 are consistent with the results of an earlier study (Mąkinia, 2006) when the nitrification rates were measured at similar or higher temperatures. In that study, the specific AURs at the "Wschód" WWTP with the settled wastewater ranged from 3.3 g N/(kgVSS h) at T= 15.9°C to 5.4 g N/(kgVSS h) at T= 19.3 °C. In comparison, the lowest specific AUR = 2.0 g N/(kgVSS h) at T=20.4 °C with the settled wastewater from "Dębogórze" WWTP was measured during the experimental series in the period May-June when the nitrification capacity was not fully recovered after the winter nitrification loss. The other six AUR measurements varied within the range 3.0–4.9 gN/(kgVSS h) at T = 17.1–21.6 °C. These results are also compared to later study (Mąkinia et al., 2009) conducted at the same plants where during three phase batch test (anaerobic-anoxic-aerobic) the nitrification rates ranged from 2.0-2.8 g N/(kgVSS h) with the settled wastewater without pretreatment and 1.8-3.2 g N/(kgVSS h) with the settled wastewater after coagulation-flocculation at temperatures ranged from 12.8 to 16.6 °C. The lower reductions in the aerobic phase resulted from a similar effect of nitrification in the parallel reactors (that process was not affected by the removal of particulates and colloids).

Table 4.7 contains the AURs reported in the literature for a full-scale WWTP and a few laboratory scale anaerobic-aerobic SBRs fed with different substrates. The AURs measured with the settled wastewater without pretreatment and after coagulation-flocculation from "Wschód" and "Dębogórze" WWTPs are comparable with the rates from two parallel lines of the Media-Pusteria WWTP in Italy (Andreottola et al., 2003) and the laboratory scale SBRs fed with mixtures of real wastewater and acetate (Chang and Hao, 1996; Kim et al., 2001). Moreover, the OURs measured during aerobic PUR batch tests with the parallel samples of wastewater and biomass from both studied plants are also comparable with the rates (up to 50 mg O_2/gVSS·h) obtained by Ali et al. (1985) in the experiments with sludge from a laboratory scale plant. For comparison, similar results Jorgensen et al. (1992) estimated from a three full-scale plants in Denmark (city of Frederikssund, Kirkeskov and Odense) during tests with mixed liquor of real wastewater and activated sludge from areation tank (Table 4.8). The results of OUR measurements are further discussed in Section 4.2.3.

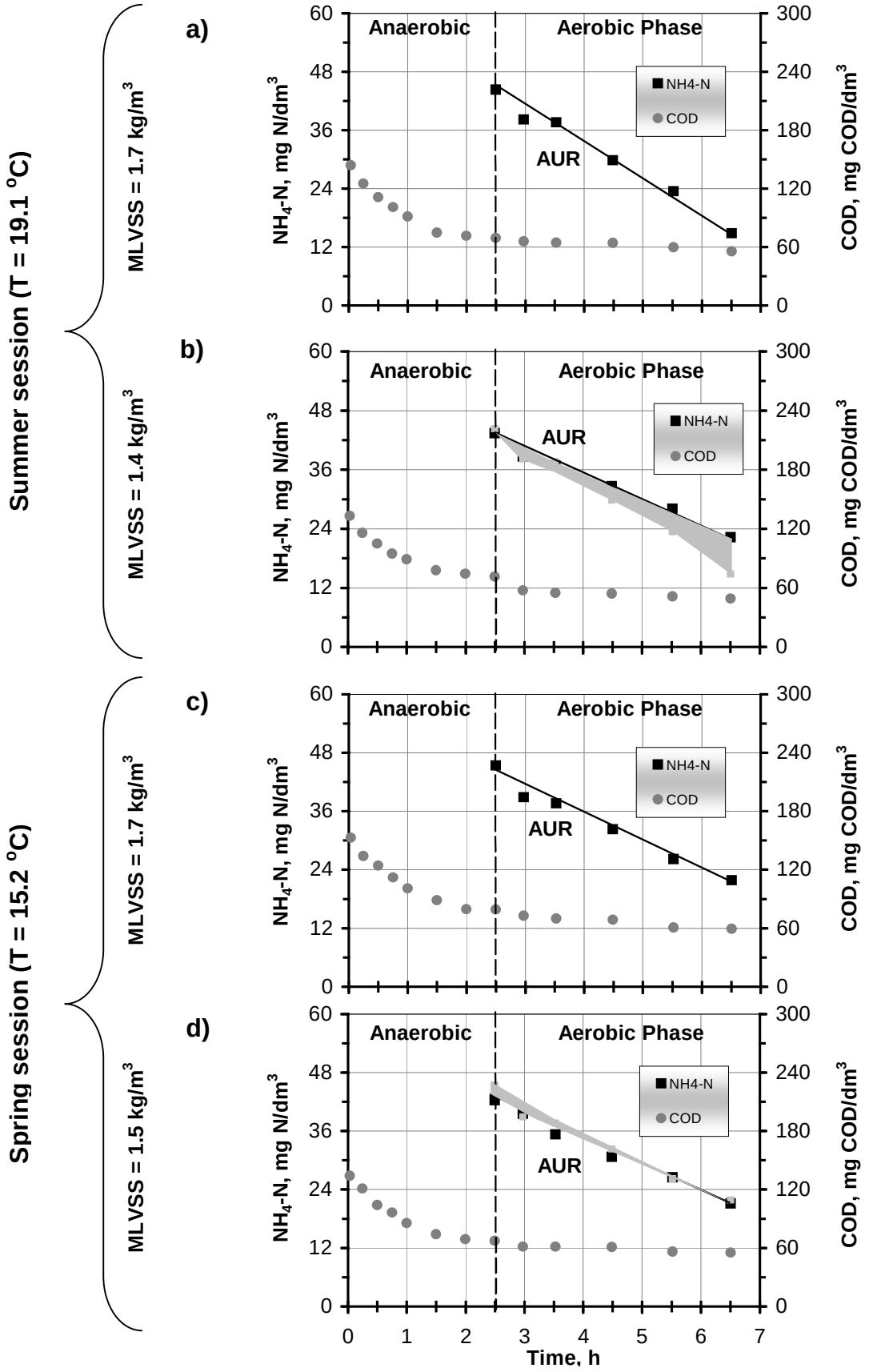


Figure 4.8. Sample results of the PRR and aerobic PUR tests with additional AUR measurment in two parallel reactors with mixed liquor from the "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

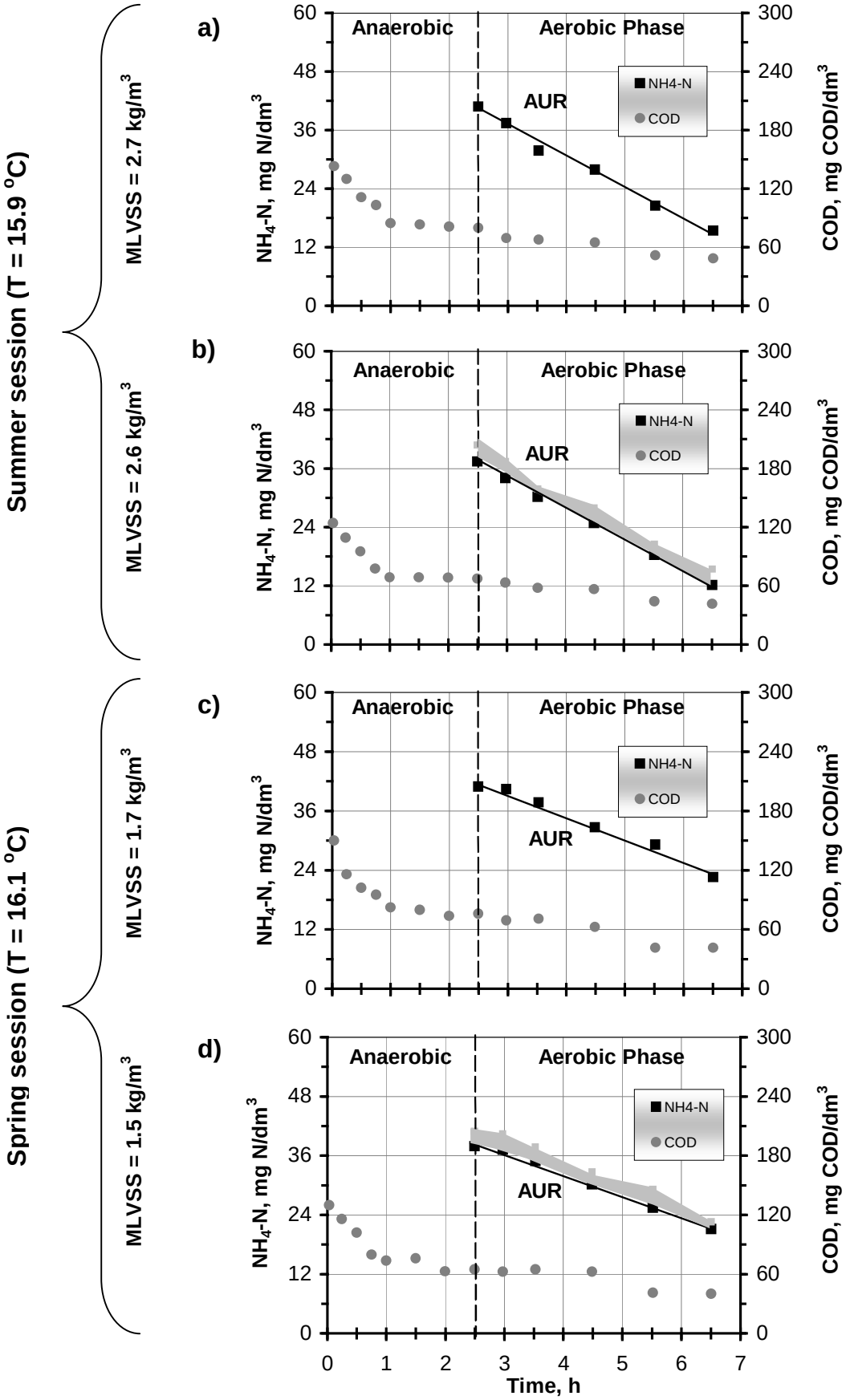


Figure 4.9. Sample results of the PRR and aerobic PUR tests with additional AUR measurment in two parallel reactors with mixed liquor from the "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

Table 4.7. Review of the AURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			g N/(kg VSS·h)	°C	
Kristensen et al. (1992)	Mixed liquor of tapwater and RAS	Pilot-scale plant	1.1 – 7.6	20	All values converted to temp. 20 °C
	Mixed liquor of tapwater and RAS	Full-scale plant	1.4 – 9.0	20	All values converted to temp. 20 °C
Chang and Hao (1996)	Anaerobic-aerobic SBR fed with a mixture of real wastewater	Lab-scale	2 – 3	20 ±2	Acetate added
van Loosdrecht et al. (1997b)	Batch test fed with MUCT biomass and municipal wastewater	Full-scale	3 – 4	25	Without primery clarification
Kim et al. (2001)	Anaerobic-aerobic SBR fed with a mixture of real wastewater	Lab-scale	4.2 – 5.6	n.d.	Acetate added
Andreottola et al. (2003)	Intermittent aeration	Full-scale	3.2 ±0.63	n.d.	
	Pre-denitrification	Full-scale	3.3 ±0.63	n.d.	
Freitas et al. (2004)	Anaerobic-aerobic SBR fed with synthetic wastewater	Lab-scale	1.0 – 1.7	20-30	Acetate added
Kosińska (2005)	AUR batch tests fed with municipal wastewater	Lab-scale	2.52 * – 2.58 **	14.7-18.5	Without* or with potato by-products** addition
Zima et al. (2009)	Batch tests fed with settled wastewater	Full-scale	4.6	20	
Puig et al. (2010)	Batch tests with MUCT biomass (pre-settling or raw influent by-passed directly into the anaerobic reactor)	Full-scale	5.0 – 5.3	20 ±0.5	Lower values with pre-settling

4.2.3. Conventional OUR tests

The average values of OUR determined in all series of batch respirometric experiments at both studied plants, are summarized in Table 4.1 and 4.2. In the OUR curves (Figure 4.6 a-d), the first phase reflects the primary metabolism of the added substrate (S_S and X_S), whereas the second phase implicitly originates from the utilization of the remaining X_S and/or stored polymers. The OUR measurements were performed with a 3 minutes intensity, until a stable profile at the lowest level (related to the hydrolysis and endogenous respiration) was achieved. The OUR tests showed a difference between both diagrams with the settled wastewater without pretreatment and after coagulation-flocculation. In the experiments with the settled wastewater, the observed OURs were associated with the utilization of S_S as well as colloidal and particulate organic compounds. The maximum OUR values (OUR_{max})

with the settled wastewater varied within the range 17.6–28.0, 24.5–31.5 and 32.2–46.8 g O_2/(kg VSS·h), respectively, during the winter, spring and summer sessions at "Wschód" WWTP. In comparison, similar experiments with the settled wastewater carried out at "Dębogórze" WWTP, showed lower values of OUR_{Max} varied within the range of 19.0–21.8 and 19.3–24.2 g O_2/(kg VSS·h), respectively, during the fall and spring sessions. The differences of OUR results were approximately from 20% to 80% between both studied plants, which may be explained lower concentration of biodegradable substrates at the "Dębogórze" compared to "Wschód" WWTP. When the pretreated samples of wastewater were used in the experiments, the observed OURs were associated with the utilization of S_S and the remaining colloidal organic fraction (part of X_S). Consequently, the values of OUR_{max} were lower in comparison with the parallel tests with the settled wastewater. The OUR_{max} values varied within the range 11.2 – 22.5 (winter), 16.2 – 25.4 (spring) and 22.1 – 33.4 g O_2/(kg VSS·h) (summer) at "Wschód" WWTP, whereas the values at "Dębogórze" WWTP (similar to sample with the settled wastewater without pretreatment) were lower and varied within the range 11.8 – 15.6 (fall) and 16.4 – 17.8 g O_2/(kg VSS·h) (spring).

Summarizing, the calculations for estimating the OUR efficiency with two different samples of the settled wastewater without pretreatment and after coagulation-flocculation, were performed in order to evaluate the impact of X_S. The experimental data revealed that two different rates, were observed in terms of the available substrates in parallel batch reactors at both WWTP. The average values of OUR_{max} in the sample with settled wastewater without pretreatment varied in the range of 22.8-39.5 and 20.4-21.8 g O_2/(kg VSS·h), respectively, at the "Wschód" and "Dębogórze" WWTP (Table 4.8). The average difference of OUR_{max} profiles observed between the parallel reactors with the settled wastewater without pretreatment were reaching from 20% to nearly 40% at both studied plants. This may be explained that in the initial period comparable high rates of OUR are resulted from a substantial amount of S_S in both samples of wastewater. However, over the time, when this substrate was ending, the difference between the values of OUR are deepened, in the reactor with the sample after coagulation-flocculation, caused by non-availability of X_S. The area under the OUR profile is an accurate indication of the level of biodegradable substrate utilized during the course of the experimental observation. According to activated sludge model definition (eg. ASM1), the first plateau of OUR mesurment corresponded to the degradation of S_S. The second OUR level corresponded to the degradation of S_H, which has much lower degradation rate than that of S_S. Finally, last plateau corresponds to the degradation of X_S. The diagrams of OUR values provided a good indication of the biological activity of X_H during utilization of pollutants in both samples of settled wastewater without and with pretreatment.

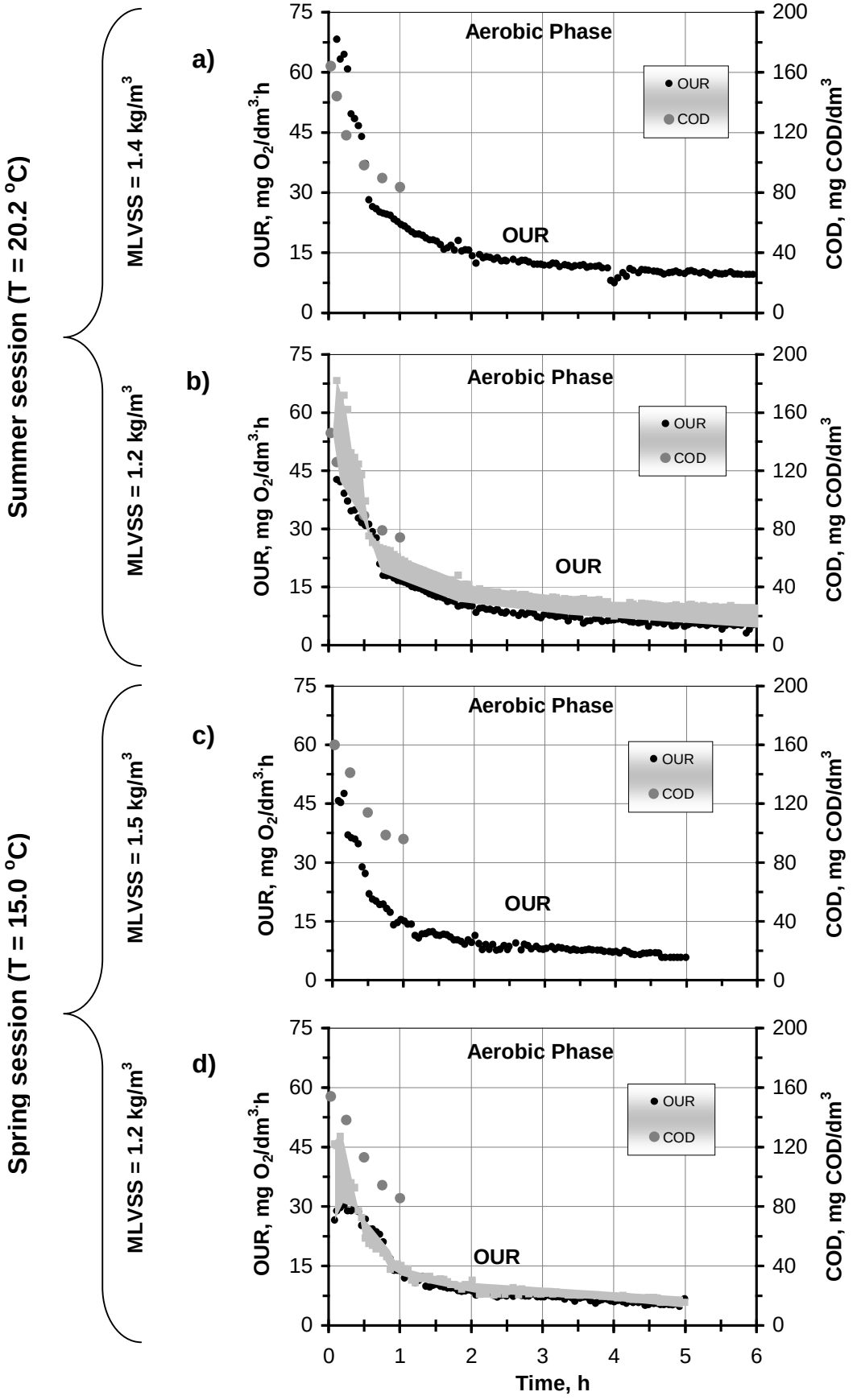


Figure 4.10. Sample results of the conventional OUR tests in two parallel reactors with mixed liquor from the "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

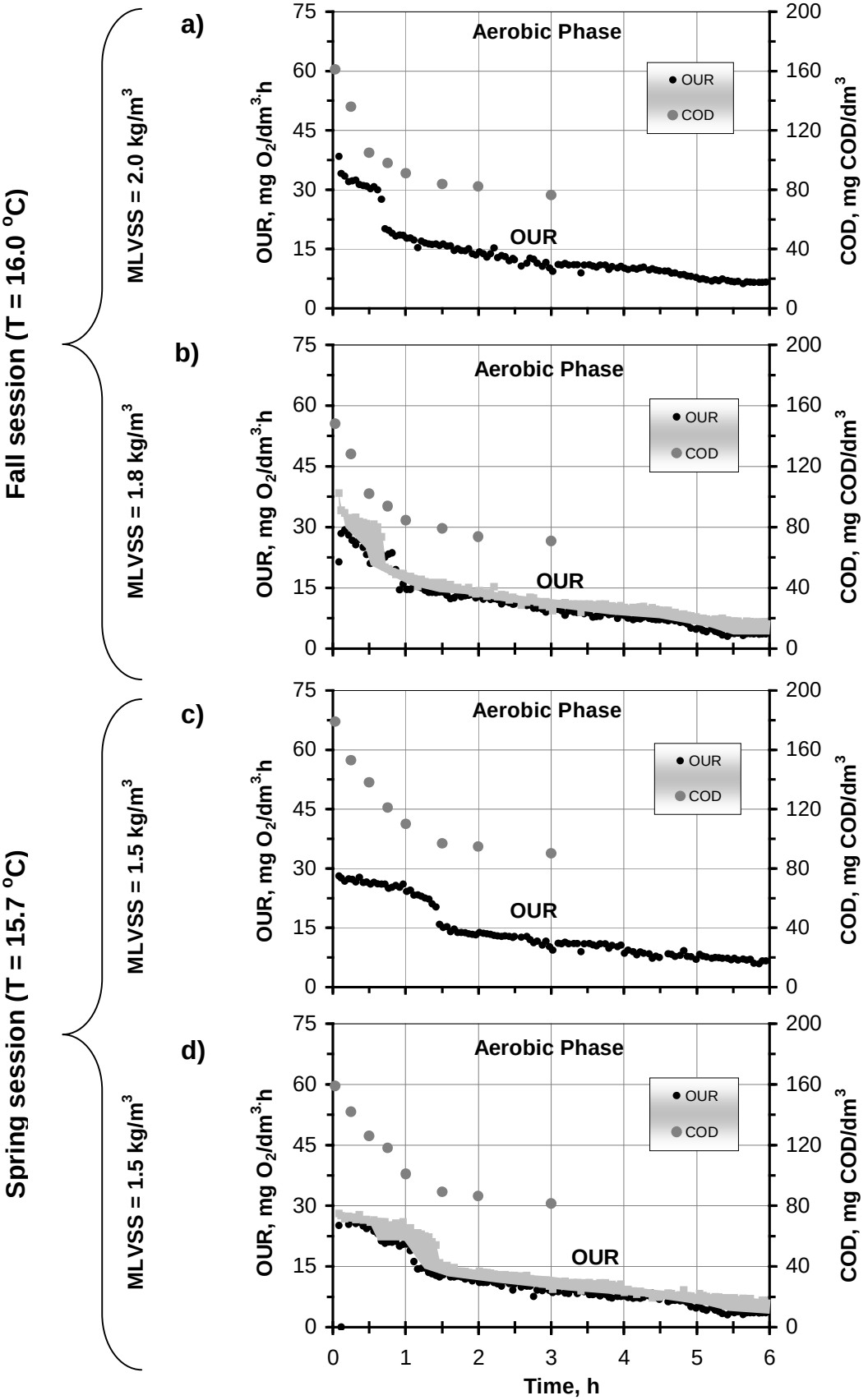


Figure 4.11. Sample results of the conventional OUR tests in two parallel reactors with mixed liquor from the "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

The values reported by Sozen (1995) and Ubay Cokgor (1997) for the OUR experiments with domestic wastewater associated with utilization of the readily biodegradable substrate varied within the range of 23-86 g O_2/m^3. For comparison, the values reported by Jorgensen et al. (1992) for the OUR experiments with real wastewater and activated sludge from a pilot-scale and full scale plant varied around the level of 20 and 22-53 mg O_2/gVSS·h, respectively (Table 4.8). In another study, Kristensen et al. (1992) demonstrated that hydrolysis of primary sludge can produce a broad range of readily biodegradable carbon sources. The authors concluded this based on the observation that OURs with hydrolysate (a liquid phase of the anaerobically hydrolysed primary sludge) as a carbon source were typically 10-20% higher compared to the OURs with acetate. In addition, Choi and Daehwan (2001) found that particulate COD contributed to the increase of total OUR (including nitrification) by approximately 5.5% when the particulate COD constituted 65% of total COD (50% of the particulate fraction was estimated to be biodegradable). The soluble solution was made of filtered nightsoil, while the particulate solution was made of settled wastewater and nightsoil.

Table 4.8. Review of the OURs reported in the literature for various types of activated sludge systems

Reference	System	Scale	Rate	Temp.	Remarks
			$gO_2/(kgVSS·h)$	°C	
Ali et al. (1985)	Mixed liquor of wastewater and biomass	Lab-scale plant	50	n.d.	
Kristensen et al. (1992)	Mixed liquor of tapwater and RAS	Pilot-scale plant	3 – 5 * 20 – 3 ** 22 – 36 ***	20	Carbon in excess*, acetate** or hydrolysate***
		Full-scale plant	8 – 10 * 21 – 29 ** 22 – 35 ***		
Jorgensen et al. (1992)	Mixed liquor of wastewater and activated sludge from areation tank	Pilot-scale	20	n.d.	
		Full-scale	22 – 53	n.d.	
Ubay Cokgor (1997)	Constantly aerated batch reactor with synthetic wastewater	Lab-scale	1.0 – 1.7	20	
Helness and Odegaard (1998)	Bed biofilm reactor operated as a SBR	Lab-scale	4.8	n.d.	
Kim et al. (2001)	Anaerobic-aerobic SBR fed with a mixture of real wastewater	Lab-scale	4.2 – 5.6	n.d.	Acetate addition
Kosińska (2005)	OUR batch tests fed with municipal wastewater	Lab-scale	10.1* 15.9**	14.7-18.5	Without* or with potato by-products** addition
Krzanowski and Wałęga (2007)	OUR tests fed with raw wastewater	Lab-scale	7.77	13.3-18.1	Sugar industry waste addition

Simultaneous measurement of COD and oxygen consumptions during OUR tests allowed to estimate of heterotrophic growth yield coefficients Y_H according to the Equation 2.6. The Y_H determined in all series of batch respirometric experiments at both studied plants, are summarized in Table 4.1 and 4.2. The values of Y_H estimated for the settled wastewater without pretreatment and after coagulation-flocculation at the "Wschód" WWTP remained in a relatively narrow range, i.e. 0.65-0.67 g cell COD/g COD and 0.62-0.66 g cell COD/g COD. For comparison, the yield coefficients obtained at "Dębogórze" WWTP were higher , i.e. 0.68 and 0.69 g cell COD/g COD and 0.69-0.71 g cell COD/g COD.

The heterotrophic growth yield coefficients results from the present study agree well with reported values in the literature (Table 4.9). The typical values of Y_H for municipal wastewater, reported by Henze et al. (1987), were within the range of 0.46-0.69 g COD /g COD. Comparatively, measured yields at "Wschód" WWTP, are similar to the classical default value Y_H = 0.67 g cell COD/g COD used in the ASM1 by (Henze et al., 1987) and typical range varying from 0.62 to 0.67 presented by Sperandio et al. (1999). In addition, estimated by the method of Sollfank and Gujer (1991) and further by Orhon et al. (1996), the value of Y_H = 0.63-0.64 for domestic wastewater was confirmed. Hence, the default value of Y_H = 0.63 g COD/g COD, used in ASM2 and ASM2d, was agreed within a typical range from earlier study. The same value was assumed as the ASM3 default for heterotrophic growth on cell internal storage products, X_{STO}, but Rieger et al. (2001) increased the yield to 0.8 g cell COD/g COD in ASM3P. A higher range of Y_H values (0.72-0.78 g COD/g COD) was found at the "Wschód" WWTP with a few external carbon sources such as acetate, ethanol, fusel oil and distilled raw alcohol (Swinarski, 2011). Dirks et al. (1999) proved that the difference in the Y_H coefficient could indeed result from the type of substrate. The reported Y_H values for the activated sludge from two studied Danish WWTPs were 0.71-0.72 and 0.66-0.67 g COD/g COD for acetate and ethanol, respectively.

Table 4.9. Review of the heterotrophic growth yield coefficients reported in the literature for various types of activated sludge systems

Reference	System	Scale	Y_H	Temp.	Remarks
			gCOD /gCOD	°C	
Ramanathan and Gaudy (1971)	Conventional activated sludge process	Full-scale	0.48 – 0.72	n.d.	
Henze et al. (1987)	Mixed liquor of municipal wastewater and activated sludge	Lab-scale	0.46 – 0.69	n.d.	
Kappeler and Gujer (1992)	OUR batch test fed with a mixture of domestic wastewater and activated sludge	Lab-scale	0.67	n.d.	
Strotmann et al. (1999)	Conventional activated sludge fed with a mixture of municipal wastewater and acetate or glucose	Lab-scale	0.61 – 0.87	n.d.	
Dirks et al. (1999)	Conventional activated sludge fed with a mixture of municipal wastewater and ethanol* or acetate**	Full-scale	0.66 – 0.67 * 0.71 – 0.72 **	n.d.	
Ng and Hermanowicz (2005)	Mixed liquor of synthetic wastewater and activated sludge	Lab-scale	0.50	n.d.	
Zhang and Hall (2006)	Simplified UCT conventional* or membrane** EBPR process	Pilot-scale plant	0.59 ±0.08 * 0.50 ±0.05 **	13.0-20.5	Mixed liquor of municipal wastewater and activated sludge
Swinarski (2011)	OUR batch tests fed with filtered biological wastewater	Full-scale	0.72 – 0.78	13.5-20	Acetate, ethanol, fusel oil or distilled raw alcohol

4.3. Simulation study based on the results of batch tests

4.3.1. Data quality evaluation

The quality of routine operating data was evaluated according to the procedure outlined in Section 3.3.3.3 (Figure 3.14 and Equations 3.2-3.7). Using the data listed in Table 4.10, the continuity check, overall phosphorus balance and clarifier solids balance were calculated. Based on these results, three parameters including Xras, Qwas and SRT were balanced. The analysis revealed that significant discrepancies existed between the conventionally calculated and balanced SRT as the balanced values only constituted 71-89% and 85-99% of the calculated ones for the "Wschód" WWTP and "Dębogórze" WWTP, respectively. The consequences of these discrepancies for the influent wastewater characterization are discussed in Section 4.3.2.

Table 4.11 contains the data for calculating the mass balance for nitrogen and oxygen demand. The ARD values for ASM2d predictions of the total oxygen uptake, OU_{tot}, in the bioreactors varied within the range of 5.0-7.3% ("Wschód" WWTP) and 1.0-6.1% ("Dębogórze" WWTP). Except for one case (summer study period) the predicted total oxygen uptakes ($OU_{tot,pre}$) at the "Wschód" WWTP were higher then the values calculated from the oxygen demand mass balance, $OU_{tot,cal}$. The model predictions at "Dębogórze" WWTP were lower in both study periods then the balanced values.

Table 4.10. Accuracy evaluation of measurements and operating parameters based on the mass balance calculations for phosphorus and suspended solids at the two large "Wschód" and "Dębogórze" WWTPs

Plant	Study period	Measured parameters (average value)										Balanced parameters		
		Q_{in}	$P_{tot.,in}$	X_{asr}	$i_{P,X}$	X_{out}	$P_{tot.,out}$	$P_{f,out}$	Q_{ras}	X_{ras}	Q_{was}	SRT	X_{ras}	Q_{was}	SRT
		m^3/d	g P/m^3	g/m^3	g P/g	g/m^3	g P/m^3	g P/m^3	m^3/d	g/m^3	m^3/d	d	g/m^3	m^3/d	d
WS	Dec.,2007 – Mar., 2008	27333	17	6310	0.043	17.6	0.80	0.08	24302	10697	774	20.6	12964	793	15.4
	Jun. – Oct., 2008	26738	16.7	4850	0.054	10.3	0.60	0.26	21821	8547	921	17.5	10415	764	15.5
	Apr. – May, 2009	23534	12.6	5296	0.038	12.2	0.48	0.09	23739	7922	720	25.2	10218	733	18.0
DG	Sep. – Nov., 2009	5921	13.7	4650	0.039	5.6	0.48	0.19	3796	10772	169.7	32.0	11364	177	27.3
	May, 2010	5820	11.2	3890	0.042	5.0	0.24	0.06	4192	10267	155.9	30.5	8922	170	30.2

Note: *WS – "Wschód" WWTP, DG – "Dębogórze" WWTP*

Table 4.11. Accuracy evaluation of measurements and operating parameters based on the mass balance calculations for nitrogen and oxygen demand at the two large "Wschód" and "Dębogórze" WWTPs

Plant	Study period	Measured parameters (average value)									Calculated parameters	Predicted parameters
												ASM2d
		COD_{in}	COD_{out}	$N_{tot.,in}$	$N_{tot.,out}$	$N_{tot.,was}$	TKN_{in}	TKN_{out}	i_{VT}	i_{CV}	$OU_{tot,cal}$	$OU_{tot,pre}$
		g COD/m^3	g COD/m^3	g N/m^3	g N/m^3	g N/m^3	g N/m^3	g N/m^3	-	-	kg O_2/d	kg O_2/d
WS	Dec.,2007 – Mar., 2008	621	43.4	85.2	15.2	912	82.0	4.4	0.75	1.42	**7694**	**8175**
	Jun. – Oct., 2008	793	43.7	78.8	10.1	960	84.2	3.1	0.74	1.42	**15244**	**14136**
	Apr. – May, 2009	691	47.8	81.4	9.7	975	78.8	3.9	0.77	1.42	**9268**	**9731**
DG	Sep. – Nov., 2009	888	22.8	87.1	8.7	775	86.8	2.6	0.75	1.42	**3664**	**3628**
	May, 2010	893	30.2	84.2	8.0	737	83.9	2.3	0.72	1.42	**4175**	**3919**

Note: *WS – "Wschód" WWTP, DG – "Dębogórze" WWTP*

4.3.2. Influent wastewater characterization

The composition of organic matter in the settled wastewater at the two studied plants is presented in Table 4.12 ("Wschód" WWTP) and Table 4.13 ("Dębogórze" WWTP). The composition was characterized according to the modified version by Mąkinia (2006) of the standard Dutch STOWA guidelines (Roeleveld and van Loosdrecht, 2002) presented in Table 3.4.

The estimated S_S (sum of S_F and S_A) accounted for 19.0-23.9% and 19.3-20.9% of total COD at the "Wschód" and "Dębogórze" WWTP, respectively. The values from both plants correspond very well to the results reported by Ekama et al. (1986), Henze et al. (1987) and Lesouef et al. (1992) for settled wastewater from the six different WWTPs in South Africa, Denmark, Hungary, Switzerland and France (see Section 2.1.3). On the other hand, the results of Naidoo et al. (1998) and recently Sperandio et al. (2001) showed that Ss can be relatively low in the case of French wastewater, i.e. 7 -15% and 1-16% of total COD, respectively. Some of these values appeared relatively small compared to this study and to S_S fraction cited in previous works which are around 10–20% (Ekama et al., 1986; Solfrank and Gujer, 1991; Henze, 1992; Kappeler and Gujer, 1992; Wentzel et al., 1995). Furthermore, the S_S fraction in the study conducted by Mąkinia (2006) at the "Wschód" and "Dębogórze" WWTPs accounted even higher values 23.4-28.0% and 31.2-37.9% of total COD, respectively. Comparative studies performed on raw wastewater in 21 Dutch full-scale WWTPs, which were presented in Table 2.11, revealed that the ratio of S_S/total COD varied within the range of 9-42% with the average value of 26% (Roeleveld and van Loosdrecht, 2002). Ginestet et al. (2002) found in the studied plants that this ratio increased in settled wastewater compared to raw wastewater. More data with previous and recent study of wastewater fractionation can be also found in Table 2.3.

Another important issue dealing with the influent wastewater characterization refers to the X_S/X_I ratio which can be used to balancing the SRT in activated sludge systems (Koch et al., 2000; Meijer et al., 2001). In this study, the initially estimated ratio X_I/total COD varied within the range of 16.1-28.8% ("Wschód" WWTP) and 28.5-31.3% ("Dębogórze" WWTP). However, in order to fit accurately the balanced and predicted SRTs at both plants the X_I/total COD ratio was increased to 21.4−32.4% and 31.6−33.4%, respectively, Table 4.12 and 4.13. Apart from the summer study period at the "Wschód" WWTP the level of X_I fraction, accounted by Mąkinia (2006) for 20.5-24.1% of total COD, is comparable with the results of this study. For comparison, at the "Dębogórze" WWTP the X_I fractions were higher than the values obtained at the same plant (22.8−24.4%) by Mąkinia (2006). However, such high X_I

still fitted to the reported range in the other studies with settled wastewater. Comparative studies performed by Roeleveld and van Loosdrecht (2002) on the level of X_I fraction, reported 39% of total COD. In the literature even higher values can also be found. In the study of Petersen et al. (2002) the calibrated X_I fraction reached 50% of total COD. The authors attributed this exceptional situation to a flush effect in the sewers during a rain even which occurred at the beginning of a 7−day measurement campaign.

Table 4.12. Results of the COD fractionation in the settled wastewater at the "Wschód" WWTP

Wschód WWTP	Component	Calculated values during the study period					
		Dec., 2007 – Mar., 2008		Jun. – Oct., 2008		Apr. – May, 2009	
		Concen-tration	% of COD	Concen-tration	% of COD	Concen-tration	% of COD
		g COD/m³	%	g COD/m³	%	g COD/m³	%
Settled wastewater fractionation	S_I	38.9	6.2	36.1	4.5	37.1	5.4
	S_F	64.1	10.3	64.9	8.2	66.1	9.6
	S_A	85.0	13.6	86.0	10.8	87.7	12.7
	X_I	133.5	21.4	257.4	32.4	181.3	26.2
	X_S	302.5	48.5	349.6	44.1	318.8	46.1
Total COD		**624.0**	**100.0**	**794.0**	**100.0**	**691.0**	**100.0**

Table 4.13. Results of the COD fractionation in the settled wastewater at the "Dębogórze" WWTP

Dębogórze WWTP	Component	Calculated values during the study period			
		Sep. – Nov., 2009		May, 2010	
		Concentration	% of COD	Concentration	% of COD
		g COD/m³	%	g COD/m³	%
Settled wastewater fractionation	S_I	21.6	2.4	25.8	2.9
	S_F	36.2	4.1	39.0	4.4
	S_A	149.1	16.8	133.1	14.9
	X_I	296.4	33.4	282.2	31.6
	X_S	384.4	43.3	412.5	46.2
Total COD		**887.7**	**100.0**	**892.6**	**100.0**

The nutrient (N and P) content of the specific organic fractions was estimated by subtracting the measured concentrations of NH_4-N from TKN and PO_4-P from TP, respectively. The results of these calculations are presented in Table 4.14. Values of the conversion factors (i_N and i_P) were changed until the best fits were obtained between the measured and calculated TKN and TP concentrations for the entire period of the sampling program. Only one set of the conversion factors was used for all simulations at each studied plant (Table 4.15).

Table 4.14. Measured vs. calculated average influent concentrations of the nitrogen and phosphorus compounds at the studied WWTPs

WWTP	Study period	Parameter	Unit	Measured value*	Calculated value
WSCHÓD	Dec. 2007 – Mar. 2008	TKN – (NH_4-N)	g N/m^3	22.42	17.75
		TP – (PO_4-P)	g P/m^3	6.62	4.01
	June – Oct., 2008	TKN – (NH_4-N)	g N/m^3	20.62	22.03
		TP – (PO_4-P)	g P/m^3	5.29	5.58
	Apr. – May, 2009	TKN – (NH_4-N)	g N/m^3	20.09	19.77
		TP – (PO_4-P)	g P/m^3	4.55	4.54
DĘBOGÓRZE	Sep. – Nov., 2009	TKN – (NH_4-N)	g N/m^3	21.4	23.5
		TP – (PO_4-P)	g P/m^3	5.20	5.89
	May, 2010	TKN – (NH_4-N)	g N/m^3	21.4	23.6
		TP – (PO_4-P)	g P/m^3	4.87	6.09

Note: **Estimated based on the routine operating data*

Table 4.15. Conversion factors for the conservation equations in the examined ASM2d at the studied WWTPs

Definition	Symbol	Unit	ASM2d value	Calibrated value
				Wschód/Dębogórze
N content of S_I	$i_{N,SI}$	g N/g COD	0.01	0.05
N content of S_F	$i_{N,SF}$	g N/g COD	0.03	0.03
N content of X_I	$i_{N,XI}$	g N/g COD	0.02	0.05
N content of X_S	$i_{N,XS}$	g N/g COD	0.04	0.05
N content of X_H, X_{PAO}, X_A	$i_{N,BM}$	g N/g COD	0.07	0.07
P content of S_I	$i_{P,SI}$	g P/g COD	0.00	0.00
P content of S_F	$i_{P,SF}$	g P/g COD	0.01	0.01
P content of X_I	$i_{P,XI}$	g P/g COD	0.01	0.01
P content of X_S	$i_{P,XS}$	g P/g COD	0.01	0.007
P content of X_H, X_{PAO}, X_A	$i_{P,BM}$	g P/g COD	0.02	0.02
TSS to COD ratio for X_I	$I_{VSS,XI}$	g VSS/g COD	0.75	0.7
TSS to COD ratio for X_S	$I_{VSS,XS}$	g VSS/g COD	0.75	0.7
TSS to COD ratio for X_H, X_{PAO}, X_A	$I_{VSS,BM}$	g VSS/g COD	0.9	0.7

Note: *Shaded area marks calibrated value*

4.3.3. Calibration of the ASM2d using the results of batch experiments

The examined ASM2d was calibrated using the experimental data from a number of batch tests, described in Section 4.2, which were carried out in the summer and fall experimental series at the "Wschód" and "Dębogórze" WWTPs, respectively. During the study period at the "Wschód" WWTP, the process temperature in the full-scale bioreactor and the average temperature in the laboratory scale batch reactor varied within the range of 18.9-20.5 °C. The corresponding temperature ranges at the "Dębogórze" WWTP were 16.0-17.8 °C. Similar to the earlier study conducted by Mąkinia (2006), the models were calibrated from the process engineering perspective,

which means that parameters were selected for calibration and modified based on knowledge of the processes "mechanistical reasoning" rather than on their sensitivity (van Veldhuizen et al., 1999b).

The first step was to calibrate the examined ASM2d with the data from the "Wschód" WWTP. As a starting point, the default values of kinetic and stoichiometric coefficients were used for the ASM2d (Henze et al., 1999). The modeled processes are coupled with each other and calibration of a specific process inevitably affected previously fitted processes. Consequently, the final sets of the coefficients were obtained after several iteration loops in the calibration procedure. In the next step, the sets of adjusted parameters served as a basis for further calibration of the ASM2d with the data from the "Dębogórze" WWTP. The purpose of such an approach was to use the same sets of model parameters at both studied plants.

Table. 4.16. The average mixed liquor (biomass and wastewater) composition estimated based on steady state simulations of the full-scale bioreactor for parallel batch tests with the settled wastewater without pretreatment and after coagulation-flocculation performance during the three and two study periods at the "Wschód" and "Dębogórze" WWTPs

Model	Plant	Study period	Type of sample	Contribution of the component, %									Total
				X_{inorg}	X_{PP}	X_{PHA}	X_{STO}	X_{PAO}	X_A	X_H	X_S	X_I	%
Modified ASM2d	Wschód	Dec. 2007 - Mar. 2008	SW	12.0	3.4	0.7	0.0	12.9	1.3	19.6	6.9	43.2	**100.0**
			SW c-f	12.8	4.3	0.8	0.0	12.6	1.2	18.9	2.2	47.2	**100.0**
		June - Oct., 2008	SW	12.3	3.1	0.6	0.0	9.8	1.0	18.8	8.3	46.1	**100.0**
			SW c-f	13.1	3.5	0.6	0.0	10.4	1.1	17.5	3.4	50.4	**100.0**
		Apr. - May, 2009	SW	8.3	2.4	0.6	0.0	13.7	0.8	23.4	10.5	40.3	**100.0**
			SW c-f	10.2	3.2	0.7	0.0	14.3	0.8	21.4	3.2	46.2	**100.0**
	Dębogórze	Sep.- Nov., 2009	SW	10.0	3.3	4.9	0.0	9.4	0.9	19.1	8.5	43.9	**100.0**
			SW c-f	11.0	3.8	5.9	0.0	10.3	1.2	18.2	3.8	45.8	**100.0**
		May, 2010	SW	10.8	3.2	3.3	0.0	9.9	1.0	18.6	11.2	42.0	**100.0**
			SW c-f	11.5	3.5	3.7	0.0	10.2	1.1	19.7	3.7	46.6	**100.0**

In each iteration loop, the steady state simulation of the full-scale bioreactor performance provided the initial mixed liquor (biomass and wastewater) composition for dynamic simulations of the batch tests with the settled wastewater without pretreatment and after coagulation-flocculation. The composition obtained in the final loop is presented in Table 4.16. In addition, the ASM2d calibration based on a 96-h measurement campaign in the full-scale MUCT bioreactor at the "Wschód" WWTP has been described in several previous publications (eg. Mąkinia et al., 2011, Swinarski, 2011). The results of the 96-h measurement campaign vs. ASM2d predictions are presented in Figure 4.12.

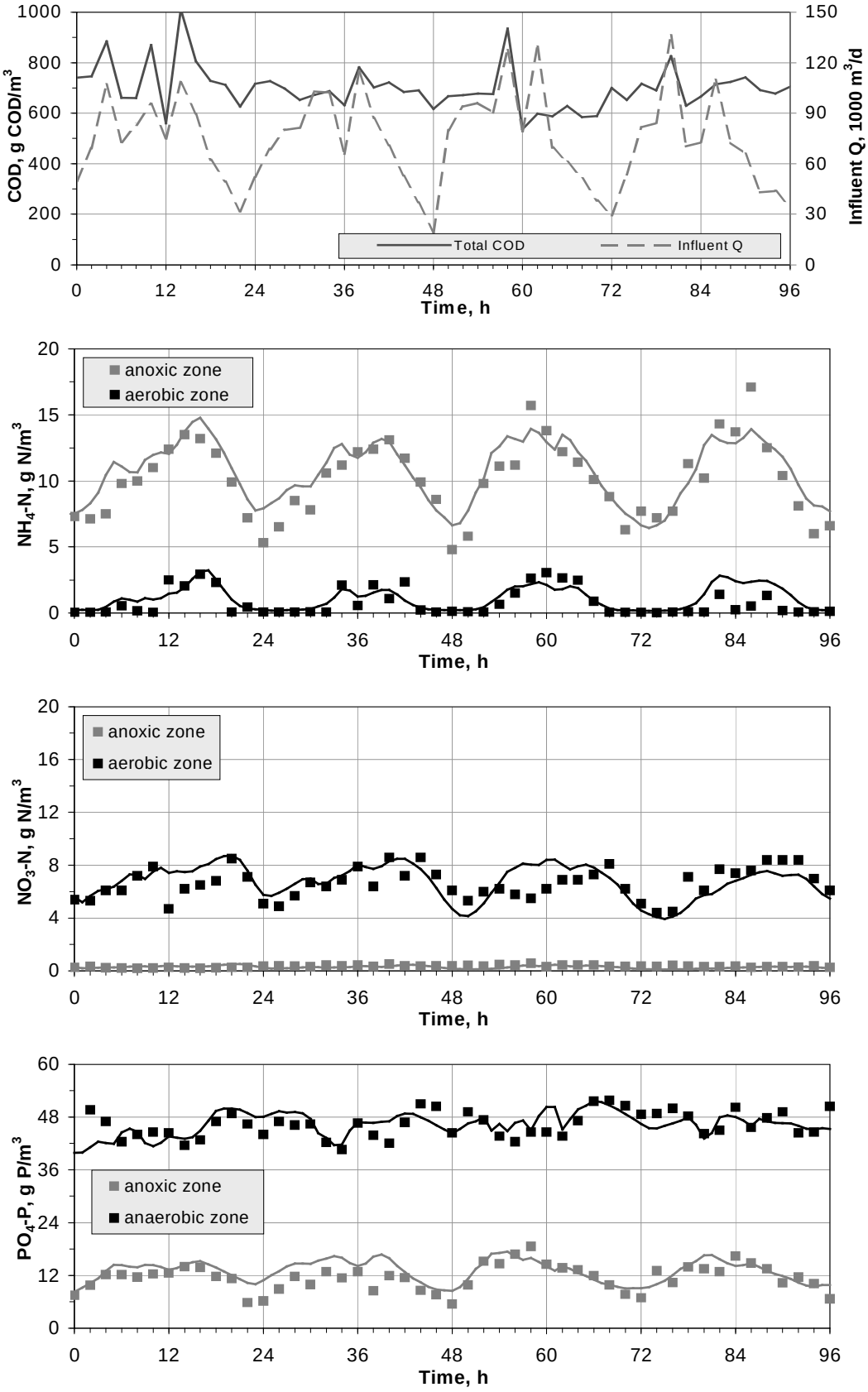


Figure 4.12. Measured data vs. ASM2d predictions for a 96-h measurement campaign in the full-scale MUCT bioreactor at the "Wschód" WWTP: (a) influent flow rate and total COD (b) NH_4-N concentrations in the anoxic and aerobic zone effluents, (c) NO_3-N concentrations in the anoxic and aerobic zone effluents, (d) PO_4-P concentrations in the anaerobic and anoxic zone effluents (Mąkinia et al., 2011).

The values of parameters adjusted at each calibration level of both studied plants with comparison to the default ASM2d values are presented in Table 4.17. With these calibrated values, the process rates (NUR, OUR, PRR and anoxic/aerobic PUR) during the batch tests with the settled wastewater without pretreatment and after coagulation-flocculation from the "Wschód" and "Dębogórze" WWTPs were simulated (Figure 4.13-4.20 a-d).

Table 4.17. List of the default values of parameters in ASM2d and the values adjusted during model calibration at both studied plants and parallel batch tests with the settled wastewater without pretreatment and after coagulation-flocculation.

Symbol	Unit	Default value (Henze et al., 1999)	Calibrated value at both studied plants
Hydrolysis:			
k_{hyd}	d^{-1}	3.0	2.5
η_{fe}	–	0.4	0.1
K_x	–	0.1	0.2
"Ordinary" heterotrophic organisms (X_H):			
μ_H	d^{-1}	6.0	3.0
Autotrophic (nitrifying) organisms (X_A):			
μ_A	d^{-1}	1.0	1.35
$K_{NH4,A}$	g N/m^3	1.0	1.3
$K_{PO4,A}$	g P/m^3	0.01	0.001
Phosphate accumulating organisms (X_{PAO}):			
q_{PHA}	d^{-1}	3.0	6.0
q_{PP}	d^{-1}	1.5	4.5
$\eta_{NO3,PAO}$	–	0.6	0.5
K_{PP}	g COD/g COD	0.01	0.02
$K_{SA,PAO}$	g COD/m^3	4.0	1.0
K_{IPP}	g P/g COD	0.02	0.1
K_{PHA}	g COD/g COD	0.01	0.2
K_{NH4}	g N/m^3	0.05	0.01
K_P	g P/m^3	0.01	0.001
Y_{PO4}	g P/g COD	0.4	0.32

The default Y_{PO4} values were assumed for stoichiometric coefficients except for the polyphosphate requirement for PHA storage (Y_{PO4}), which was experimentally determined based on anaerobic phosphate release measurements (Mąkinia et al., 2009; Swinarski et al. 2009a, 2009c). The value of Y_{PO4} = 0.32 mg P/mg COD was different from the ASM2d default of 0.40 mg P/mg COD, but remained within a typical range for simulation (0.30-0.43 mg P/mg COD), which was reported by Johansson et al. (1996).

Brdjanovic et al. (2000) used for simulation a higher value of Y_{PO4} (0.36 mg P/mg COD) compared to the values experimentally determined in batch tests with the WWTP mixed liquor and acetate (0.27-0.29 mg P/mg COD). The authors related lower values of Y_{PO4} at the studied plant (Haarlem Waarderpolder) to long SRTs or significant presence of glycogen accumulating organisms (GAOs) based on the results of anaerobic phosphate release tests in which acetate was still utilized even after depletion of polyphosphate in the PAO biomass. In the case of the "Wschód" and "Dębogórze" WWTPs, the results of similar experiments with surplus acetate (Mąkinia, 2006; Swinarski et al., 2009c) revealed that the acetate utilization hardly continued after polyphosphate depletion in the biomass. This suggests that GAOs did not play a significant role at the plant and there was no need to model their metabolism in this study (Mąkinia, 2006).

The calibration of denitrification process rates were carried out based on the results of two batch tests (conventional NUR and PRR/anoxic PUR) with the settled wastewater without pretreatment from "Wschód" WWTP during the summer study period according to previous study (Mąkinia, 2006). In the conventional NUR tests, model predictions were fitted to the measured NURs by adjusting two parameters: the maximum growth rate of heterotrophs (μ_H) and hydrolysis rate constant (k_{hyd}). In the PRR/anoxic PUR tests, the anoxic reduction factor for PAO growth ($\eta_{NO3,PAO}$) was adjusted to calibrate the NUR in the anoxic phase. The corresponding comparisons of adjusting above parameters were made for the parallel batch tests with the settled wastewater after coagulation-flocculation. In that case, no further modifications were needed to calibrate the results of NUR and PRR/anoxic PUR. Finally, the same sets of model parameters were used for both batch tests with the settled wastewater without pretreatment and after coagulation-flocculation from the "Dębogórze" WWTP. The calibrated value of this coefficient was lower than the default one (Table 4.17).

The nitrification process based on the measured data from PRR and aerobic PUR batch tests at both plants was calibrated with three kinetic parameters, including the maximum growth rate of autotrophs (μ_A), NH_4-N saturation coefficient ($K_{NH4,A}$) and PO_4-P (nutrient) saturation coefficient ($K_{PO4,A}$) (Table 4.23). The estimated values of μ_A and $K_{NH4,A}$ were higher compared to the ASM2d defaults. Such values of $K_{NH4,A}$ has been noted for some full-scale plants due to a higher diffusion limitation resulting from low turbulence and large floc sizes (Henze et al., 2000a). The $K_{PO4,A}$ coefficient for autotrophic organisms was reduced from 0.01 to 0.001 mg P/dm^3, according to modification, proposed in the literature. Meijer et al. (2001) presented that it was necessary to simulate high rates of the nitrification process without a

limitation caused by extremely low PO_4-P concentrations, which could be temporarily observed in the aerobic zone of the full-scale bioreactor.

Several other kinetic parameters, apart the previous modifications of $Y_{PO4,}$ were adjusted to calibrate the EBPR process. The PRR was calibrated with six parameters: the rate constant for storage of PHA (q_{PHA}), saturation coefficient for PAOs with respect to S_A ($K_{SA,PAO}$), saturation coefficient for PAOs with respect to polyphosphate (K_{PP}), anaerobic hydrolysis reduction factor (η_{fe}) and saturation coefficient for particulate COD (K_X). Except for the K_X, the coefficients were reduced in comparison with the ASM2d defaults (Table 4.17).

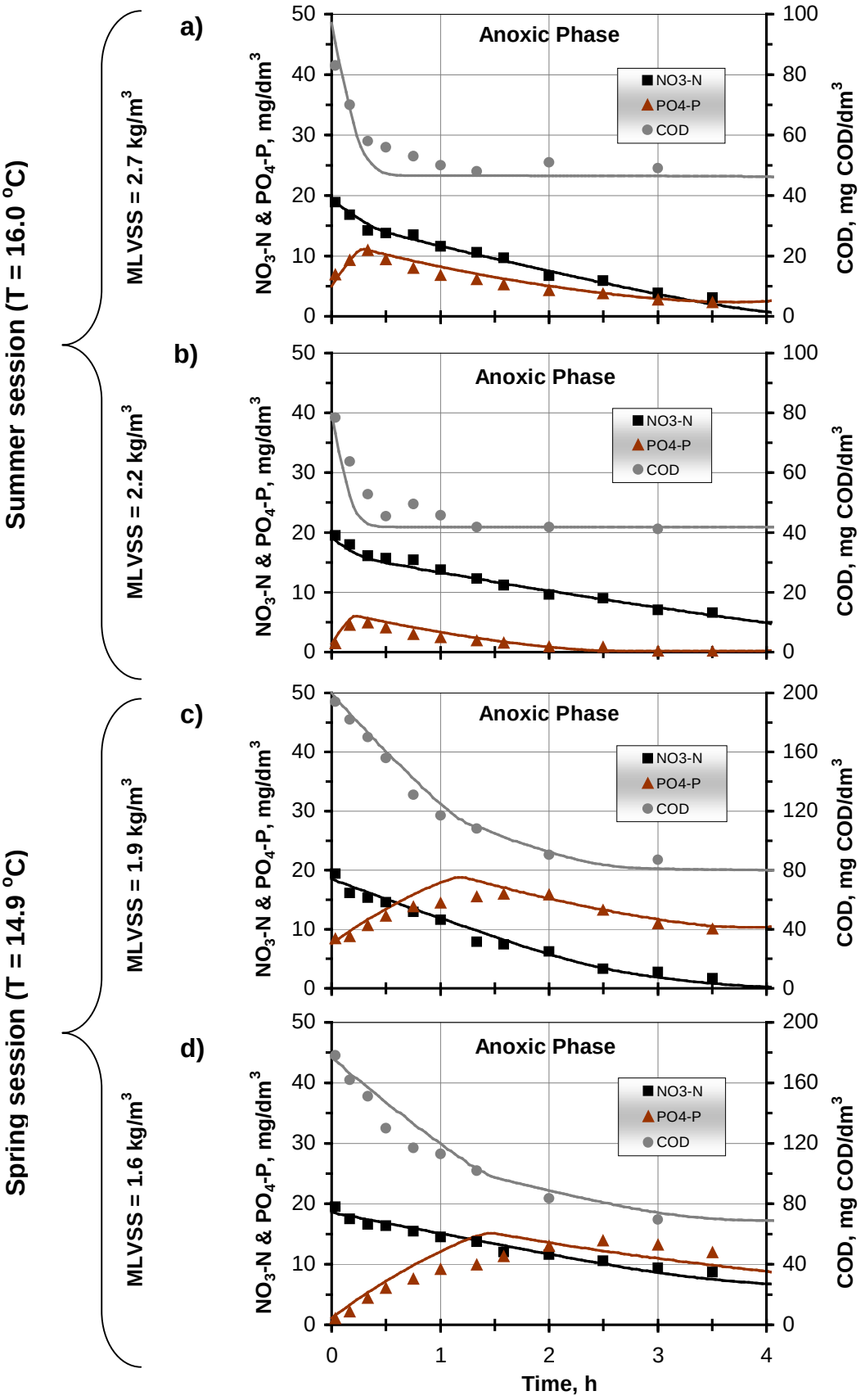


Figure 4.13. Measured data vs. ASM2d (— solid lines) predictions for conventional denitrification tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

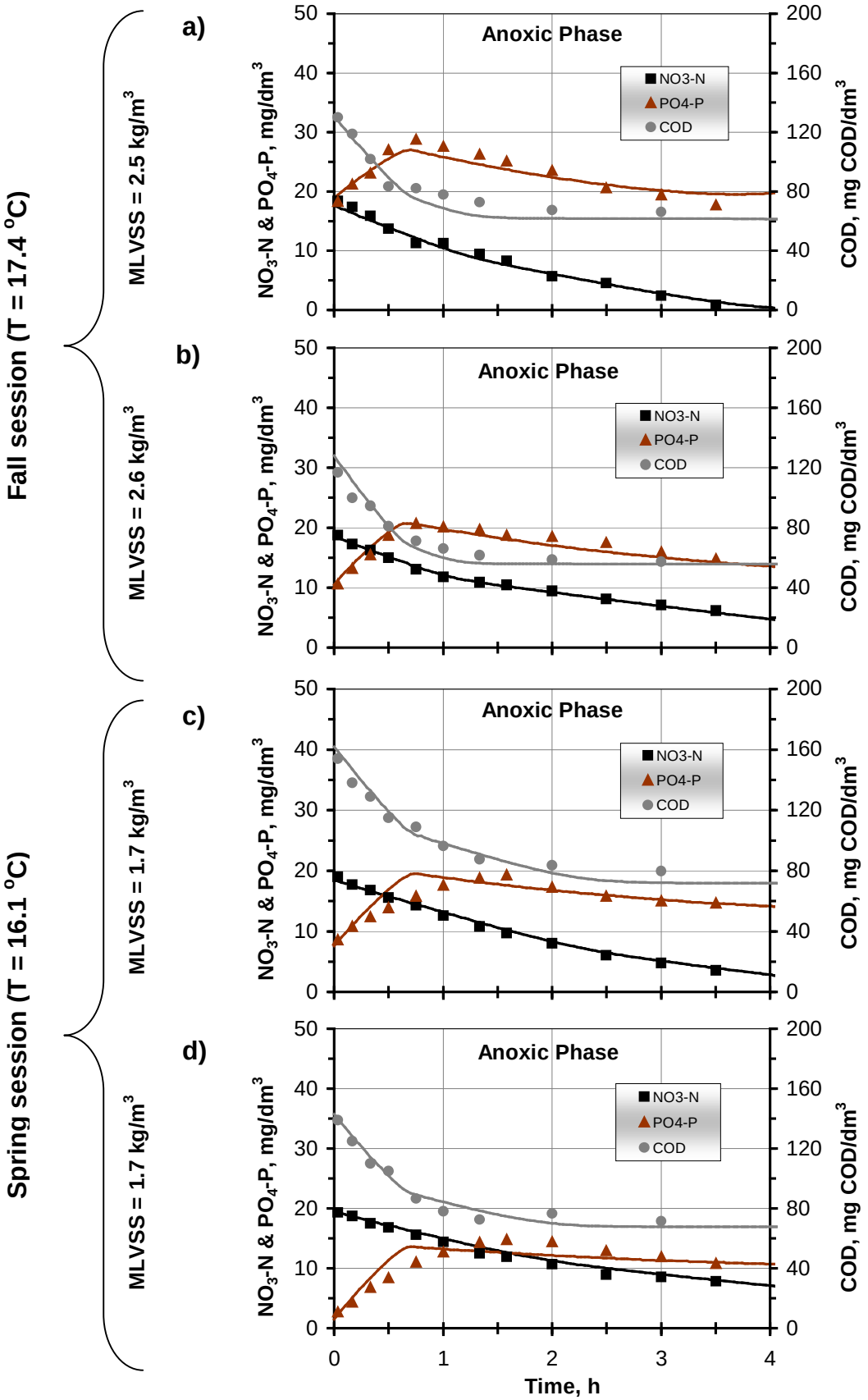


Figure 4.14. Measured data vs. ASM2d (— solid lines) predictions for conventional denitrification tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation- flocculation.

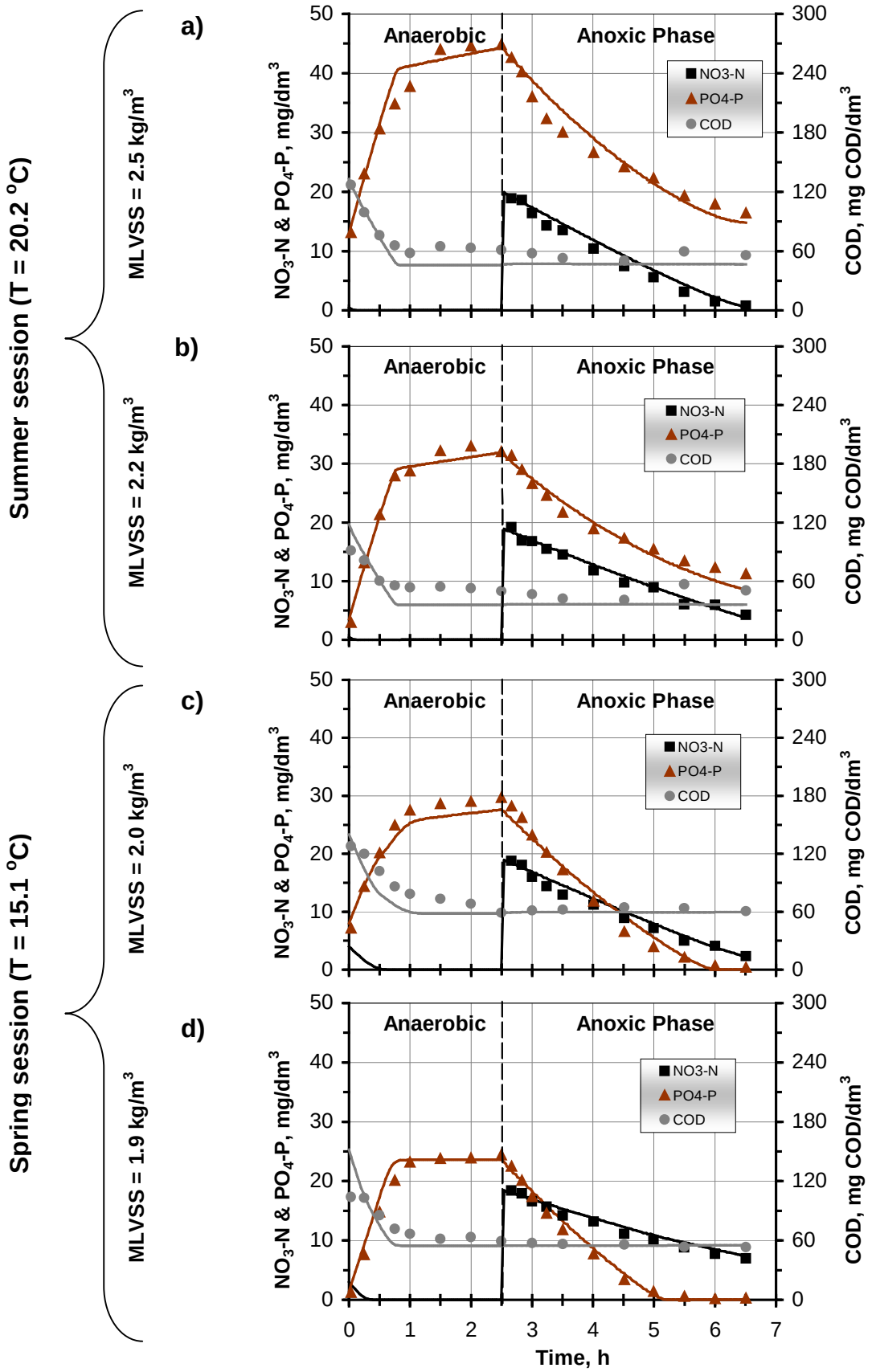


Figure 4.15. Measured data vs. ASM2d (— solid lines) predictions for for PRR and anoxic PUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

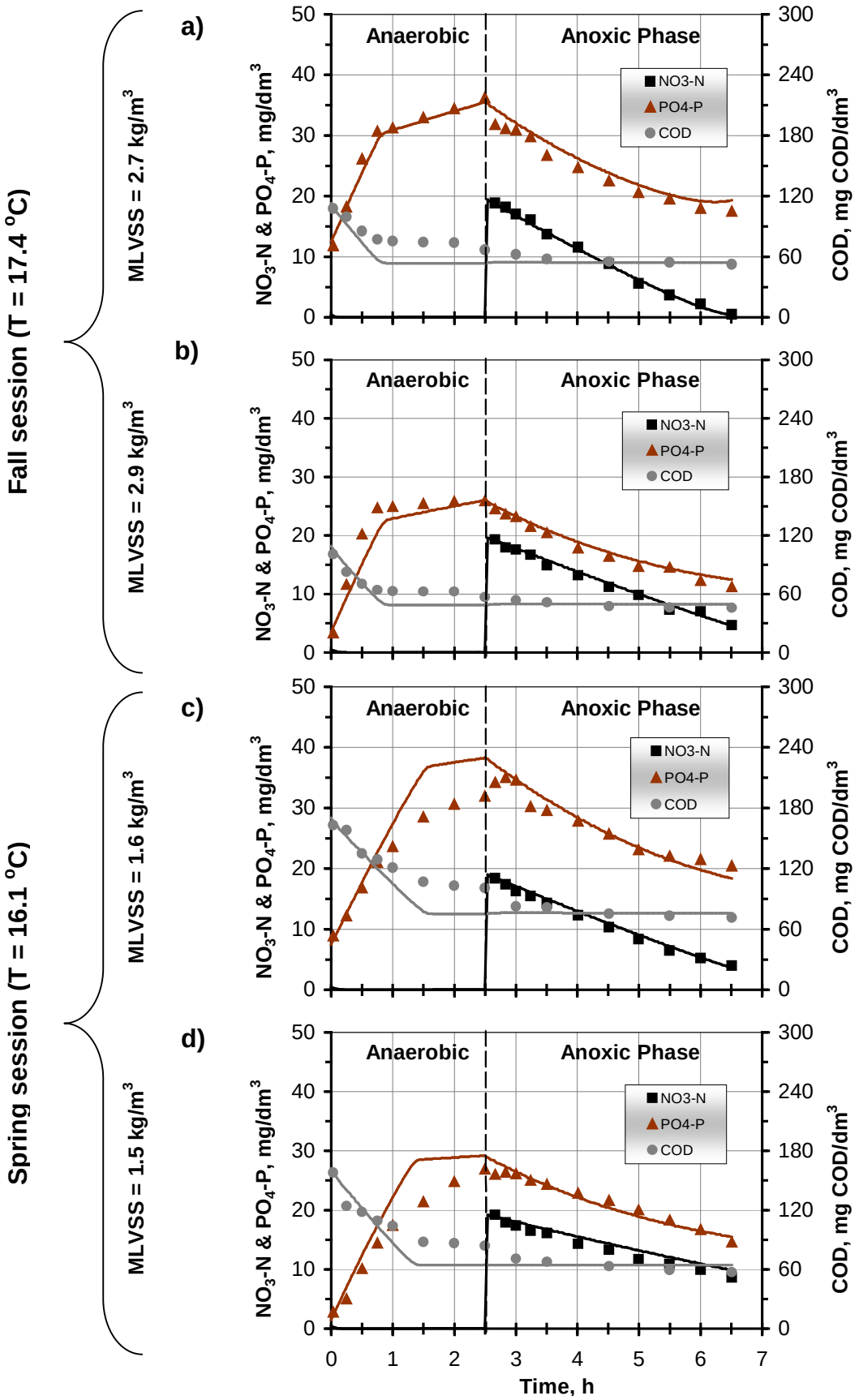


Figure 4.16. Measured data vs. ASM2d (— solid lines) predictions for for PRR and anoxic PUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

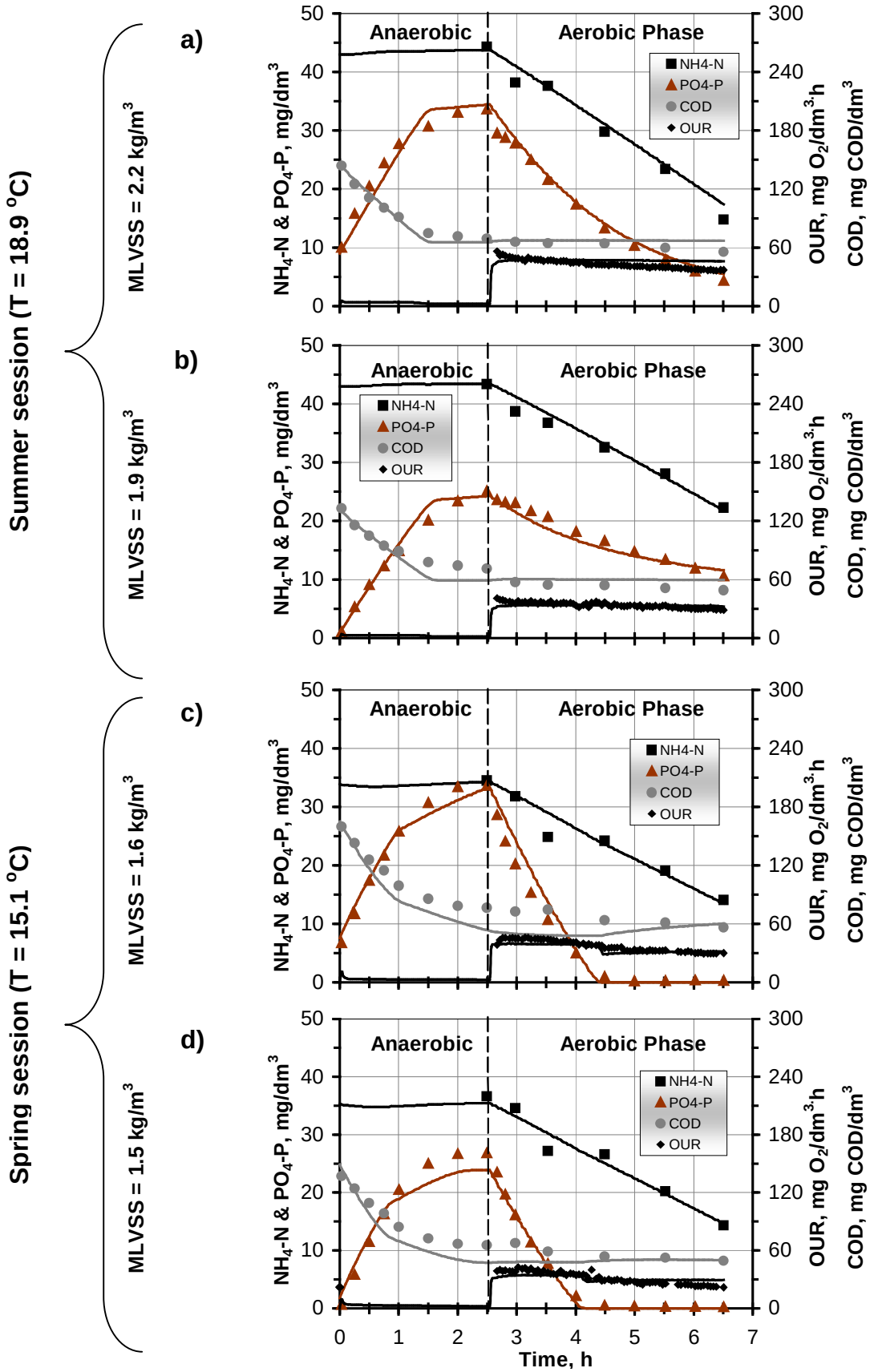


Figure 4.17. Measured data vs. ASM2d (— solid lines) predictions for PRR and aerobic PUR tests with additional AUR measurments in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

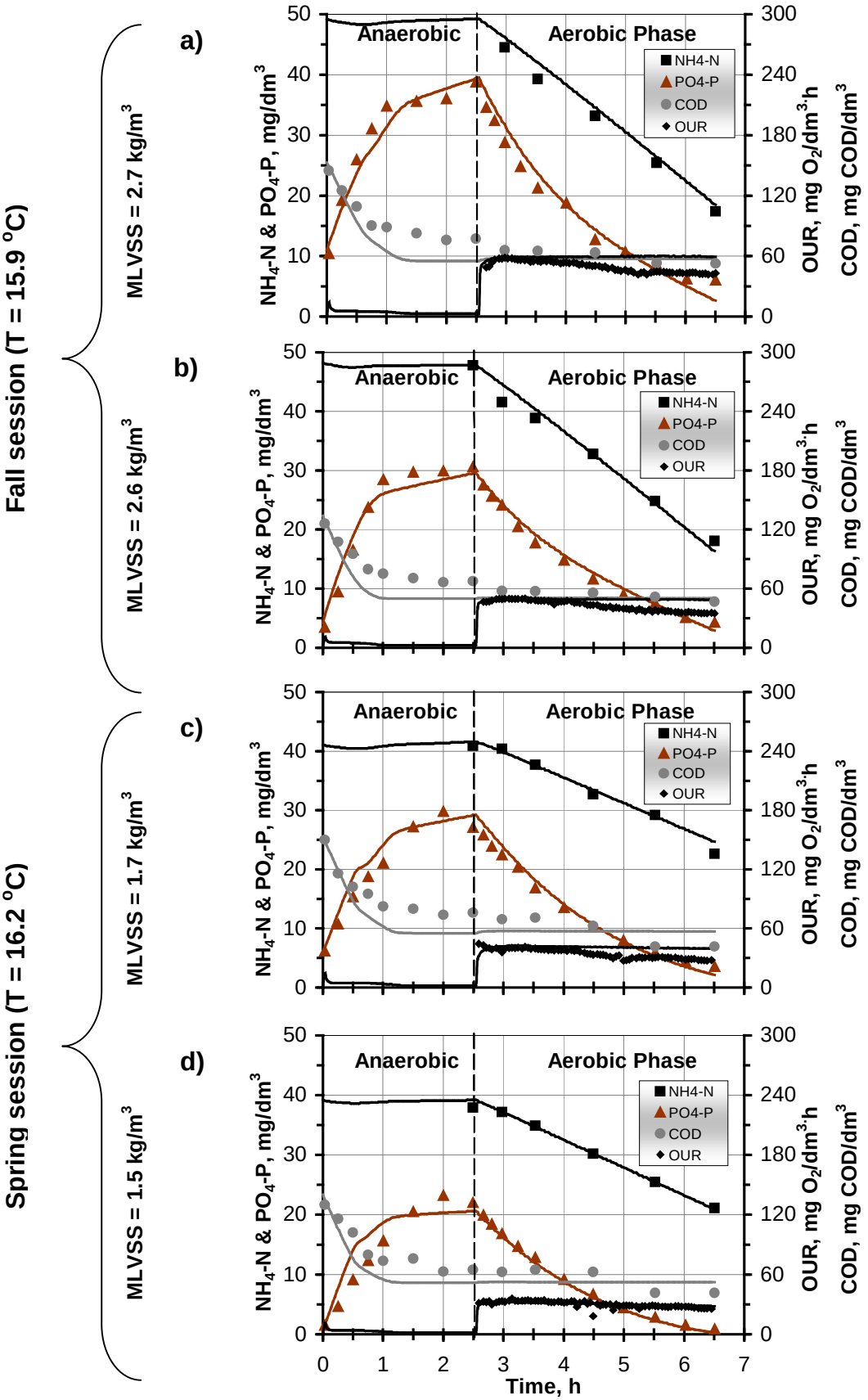


Figure 4.18. Measured data vs. ASM2d (— solid lines) predictions for PRR and aerobic PUR tests with additional AUR measurments in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

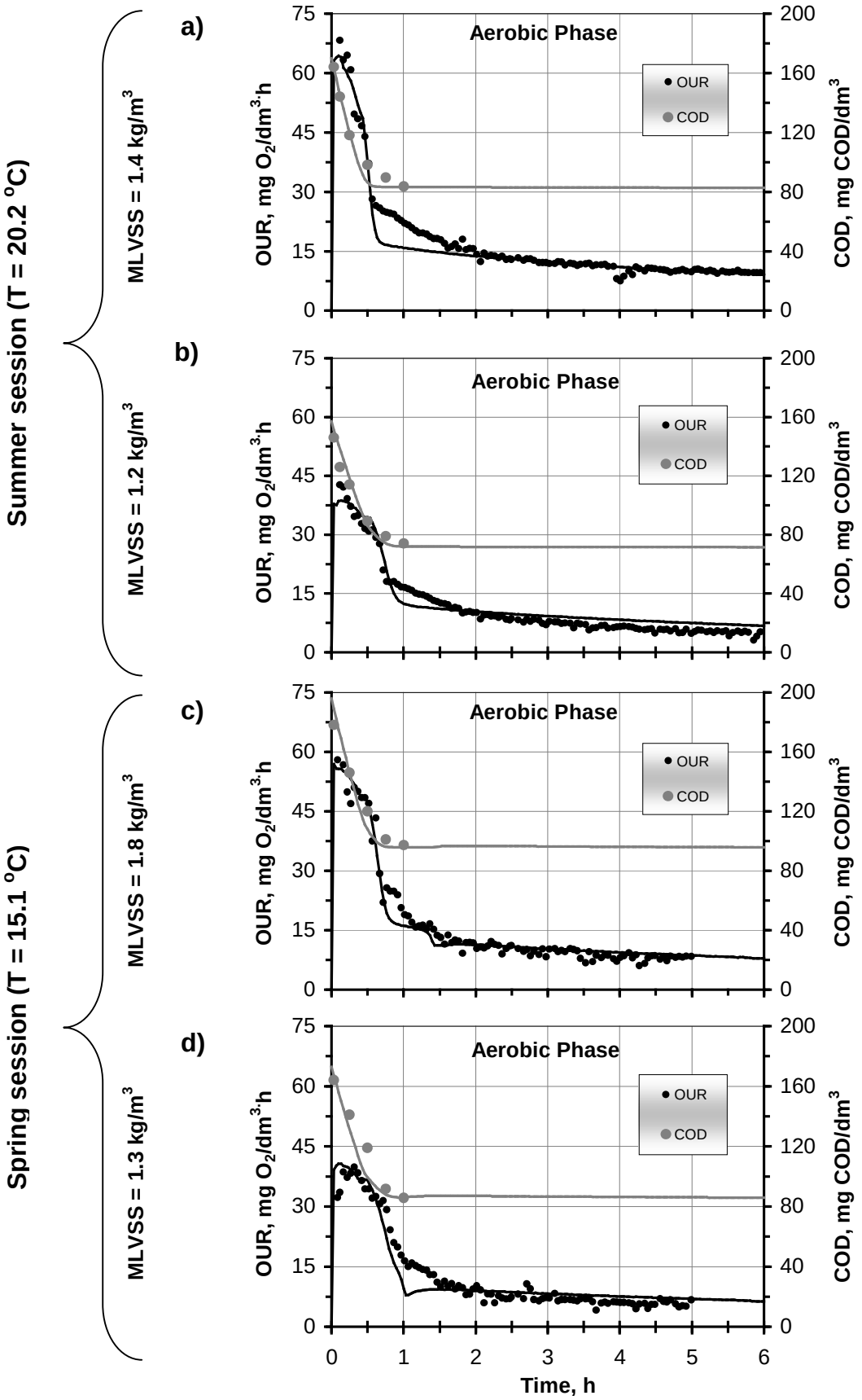


Figure 4.19. Measured data vs. ASM2d (— solid lines) predictions for OUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

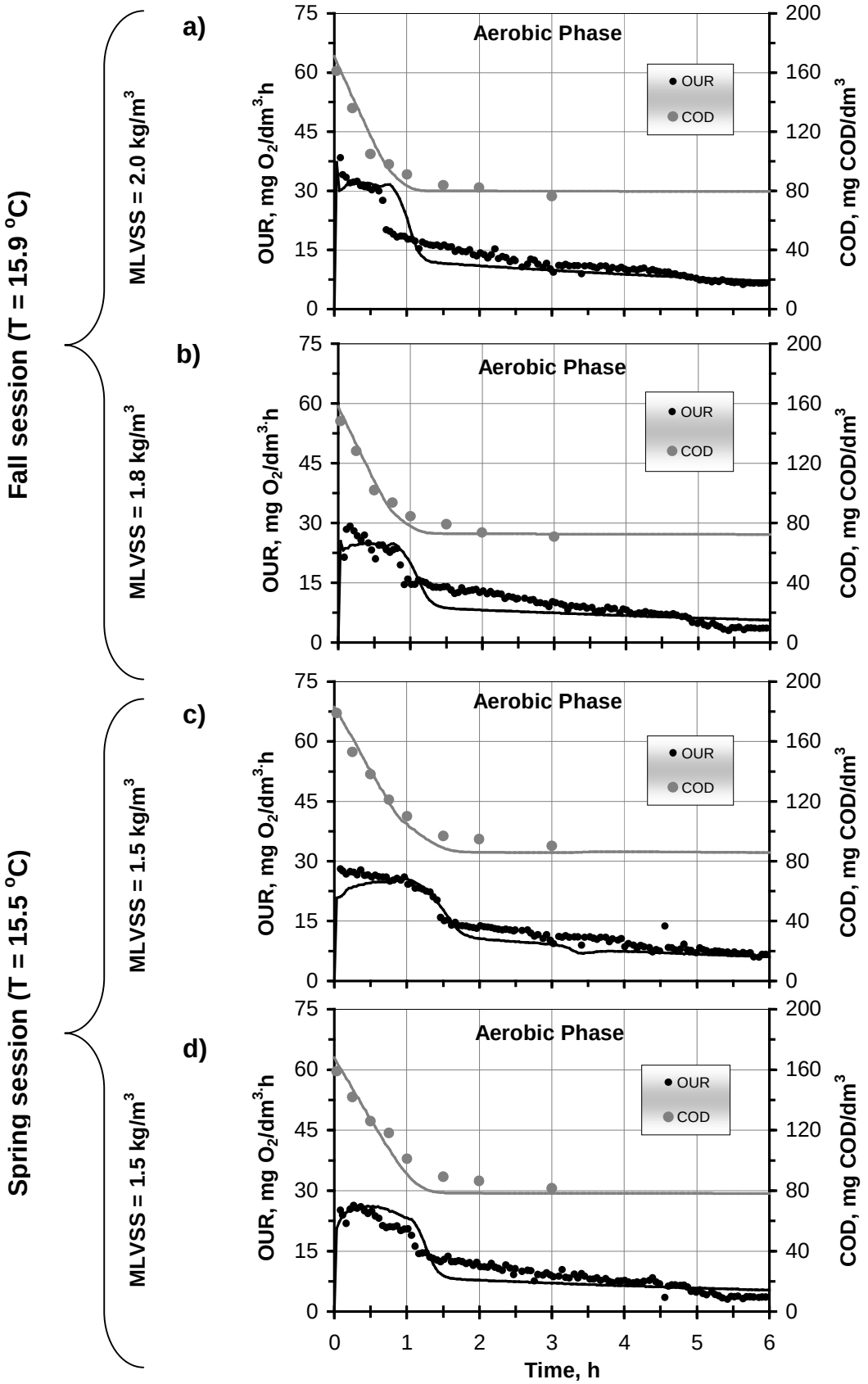


Figure 4.20. Measured data vs. ASM2d (— solid lines) predictions for OUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

4.3.4. Calibration of the modified ASM2d using the numerical optimalization method based on OUR batch tests results

The new model (modified ASM2d), was calibrated using results of the batch tests (conventional OUR) calibrated in original ASM2d from the "Wschód" and "Dębogórze" WWTPs (Figures 4.23 and 4.26 a-c). As mentioned in Section 3.3.1, the modified ASM2d incorporates one new component rapidly hydrolyzable substrate (X_{SH}) and three new processes; anaerobic, anoxic and aerobic hydrolysis of X_{SH}. The previous study of Mąkinia and Czerwionka (2009) were used to estimate the amount of new component X_{SH} (Table 4.18). According to the COD fractionation concept proposed by Melcer et al. (2003), in this study for batch tests with the settled wastewater without pretreatment, the average value of colloidal/soluble fraction ($X_{SH\ (C/S)}$ = 12% of X_S at ASM2d) was appropriated from the calculation of actual data from the "Wschód" and "Dębogórze" WWTPs. In the case of the sample with the settled wastewater after coagulation-flocculation, it was necessary to estimate the value of soluble fraction ($X_{SH\ (S)}$ = 6% of X_S at ASM2d) in order to fit all series of batch tests predictions to the kinetic parameters of the modified ASM2d proposed for settled wastewater without pretreatment (Figure 4.21). The average mixed liquor (biomass and wastewater) composition, including the new X_{SH} component was estimated for parallel the batch tests with the settled wastewater without pretreatment and after coagulation-flocculation during the three and two study periods at the "Wschód" and "Dębogórze" WWTPs (Table 4.19).

The adjusted in the new model, after optimalization step for OUR batch test, average value of stoichiometric and kinetic parameters from "Wschód" and "Dębogórze" WWTP are listed in Table 4.21. Both models were compared based on their predictions of the effect of X_S (particulate and colloidal organic compounds) in biological nutrient removal activated sludge systems (see Section 4.2). In comparison with the original ASM2d, the modified model had no or only minor effect in NUR, PRR and anoxic/aerobic PUR tests (see Section 4.3.5), but better predicted the aerobic behavior of OUR in batch experiments (Figure 4.23-4.26 a-c). In order to fit the kinetic parameters in modified ASM2d for OUR batch tests with the settled wastewater without pretreatment and after coagulation-flocculation from both studied plants was necessary the numerical optimalization using the Nelder-Mead simplex method. The stoichiometric Y_H coefficient value was directly determined from the OUR experiments from both studied plants (see Section 4.2.3).

Table 4.18. Estimation the amount of new component X_{SH} in modified ASM2d based on the results of the previous study of Mąkinia and Czerwionka (2009)

Serie	Wschód WWTP					Dębogórze WWTP				
	Settled wastewater			Colloidal fraction		Settled wastewater			Colloidal fraction	
	COD_{in}	$COD_{f(1.2),in}$	$COD_{f(0.1),in}$	$COD_{col,in}$	% of SBCOD	COD_{in}	$COD_{f(1.2),in}$	$COD_{f(0.1),in}$	$COD_{col,in}$	% of SBCOD
	gCOD/m³			gCOD/m³		g COD/m³			gCOD/m³	
1	650	237	213	24	7.5	817	259	227	32	8.7
2	630	188	152	36	11.8	923	260	223	37	8.9
3	590	182	143	39	13.6	753	200	175	25	7.4
4	548	164	131	33	12.3	799	241	186	55	15.3
5	451	138	106	32	14.5	685	209	159	50	16.2
6	821	191	154	37	9.2	641	212	175	37	12.8
7	620	176	132	44	14.5	597	209	161	48	17.9
8	1390	247	174	73	10.7	557	193	156	37	14.8
9	1001	236	172	64	13.0	855	179	158	21	5.5
10	797	165	112	53	13.6	517	187	145	42	18.1
Ave. value	**750**	**192**	**149**	**44**	**12**	**714**	**215**	**177**	**38**	**12**

Note: $COD_{f(1.2),in}$ – grab sample; soluble COD analysis after filtration (GF/C = 1.2 µm); $COD_{f(10.1),in}$ – grab sample; soluble COD analysis after filtration (GF/C = 0.1 µm); % of SBCOD – % of colloidal fraction (X_{SH}) used in modified ASM2d; calculated on the average value of X_S based on study on COD fractionation in the settled wastewater at both studied plants

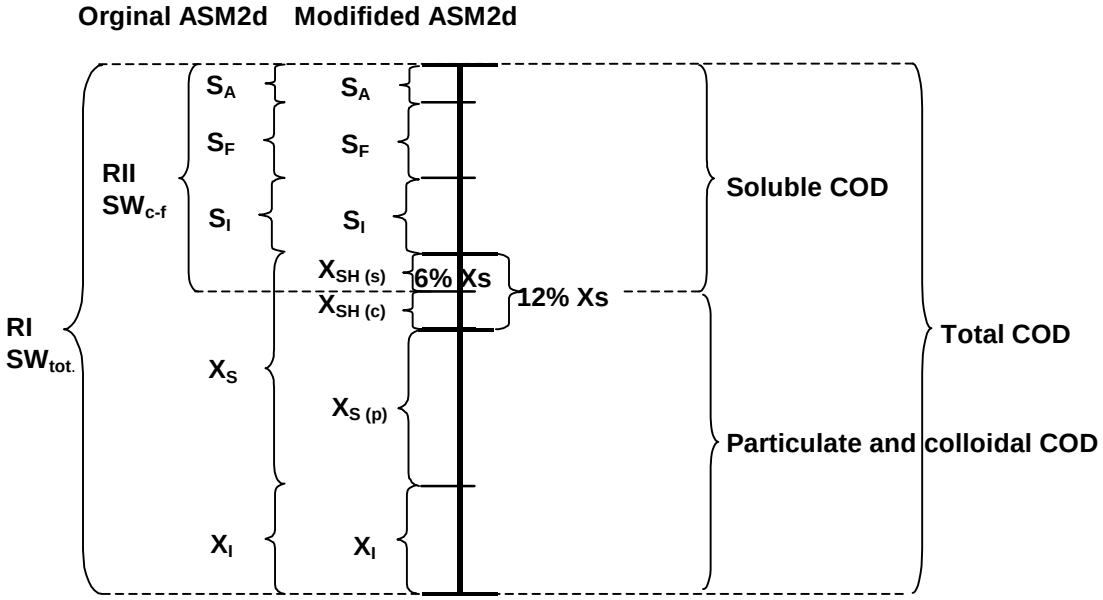


Figure 4.21. Comparasion of the COD fractionation for batch tests with the settled wastewater without pretreatment and after coagulation-flocculation in the original and modified ASM2d (based on a concept of Melcer et al., 2003).

Table. 4.19. The average mixed liquor (biomass and wastewater) composition, consisted of new X_{SH} component used at modified ASM2d, estimated for parallel batch tests with the settled wastewater without pretreatment and after coagulation-flocculation during the three and two study periods at the "Wschód" and "Dębogórze" WWTPs

Model	Plant	Study period	Type of sample	Contribution of the component, %									Total
				χ_{inorg}	X_{PP}	X_{PHA}	X_{STO}	X_{PAO}	X_A	X_H	X_S	X_{SH}	X_I	%
Modified ASM2d	Wschód	Dec. 2007 - Mar. 2008	SW	12.0	3.4	0.7	0.0	12.9	1.3	19.6	6.1	0.8	43.2	**100.0**
			SW c-f	12.8	4.3	0.8	0.0	12.6	1.2	18.9	1.7	0.5	47.2	**100.0**
		June - Oct., 2008	SW	12.3	3.1	0.6	0.0	9.8	1.0	18.8	7.3	1.0	46.1	**100.0**
			SW c-f	13.1	3.5	0.6	0.0	10.4	1.1	17.5	2.9	0.5	50.3	**100.0**
		Apr.- May, 2009	SW	8.3	2.4	0.6	0.0	13.7	0.8	23.4	9.2	1.3	40.3	**100.0**
			SW c-f	10.2	3.2	0.7	0.0	14.3	0.8	21.4	2.5	0.7	46.2	**100.0**
	Dębogórze	Sep.- Nov., 2009	SW	10.0	3.3	4.9	0.0	9.4	0.9	19.1	7.5	1.0	43.9	**100.0**
			SW c-f	11.0	3.8	5.9	0.0	10.3	1.2	18.2	3.2	0.6	45.8	**100.0**
		May, 2010	SW	10.8	3.2	3.3	0.0	9.9	1.0	18.6	9.8	1.4	42.0	**100.0**
			SW c-f	11.5	3.5	3.7	0.0	10.2	1.1	19.7	2.9	0.7	46.7	**100.0**

Table 4.20. Results of the modified ASM2d optimization using different combination of stoichiometric and kinetic coefficients in various simulation scenario for the OUR batch tests with settled wastewater during the calibrated summer/fall study periods at the "Wschód" and "Dębogórze" WWTPs

Scenario	Wschód WWTP (summer study)					Dębogórze WWTP (fall study)				
	Stoichiometric coefficient	Kinetic coefficients				Stoichiometric coefficient	Kinetic coefficients			
	Y_H	k_{hyd}	$k_{hyd,r}$	K_x	K_{xr}	Y_H	k_{hyd}	$k_{hyd,r}$	K_x	K_{xr}
	gCOD/gCOD	1/d	1/d	-	-	gCOD/gCOD	1/d	1/d	-	-
1	X	1.85	10	0.1	X	X	1.88	10	0.1	X
2	0.625	0.697	1.58	X	X	0.625	0.85	2.43	X	X
3	0.625	X	10	0.2	X	0.625	X	25	0.2	X
4	0.625	0.7	X	0.2	X	0.625	0.85	X	0.2	X
5	0.625	0.7	X	X	0.03	0.625	0.85	X	X	0.03
6	0.625	X	19	X	0.03	0.625	X	25	X	0.03
7	X	3	X	0.2	0.03	X	2.96	X	0.2	0.03
8	X	X	0.198	0.2	0.03	X	X	0.2	0.2	0.03
9	X	1.82	10.9	X	0.03	X	1.94	10.0	X	0.03
10	0.625	X	X	0.2	0.03	0.625	X	X	0.2	0.03
Average value	**0.625**	**1.5**	**8.6**	**0.18**	**0.03**	**0.625**	**1.6**	**12.1**	**0.18**	**0.03**

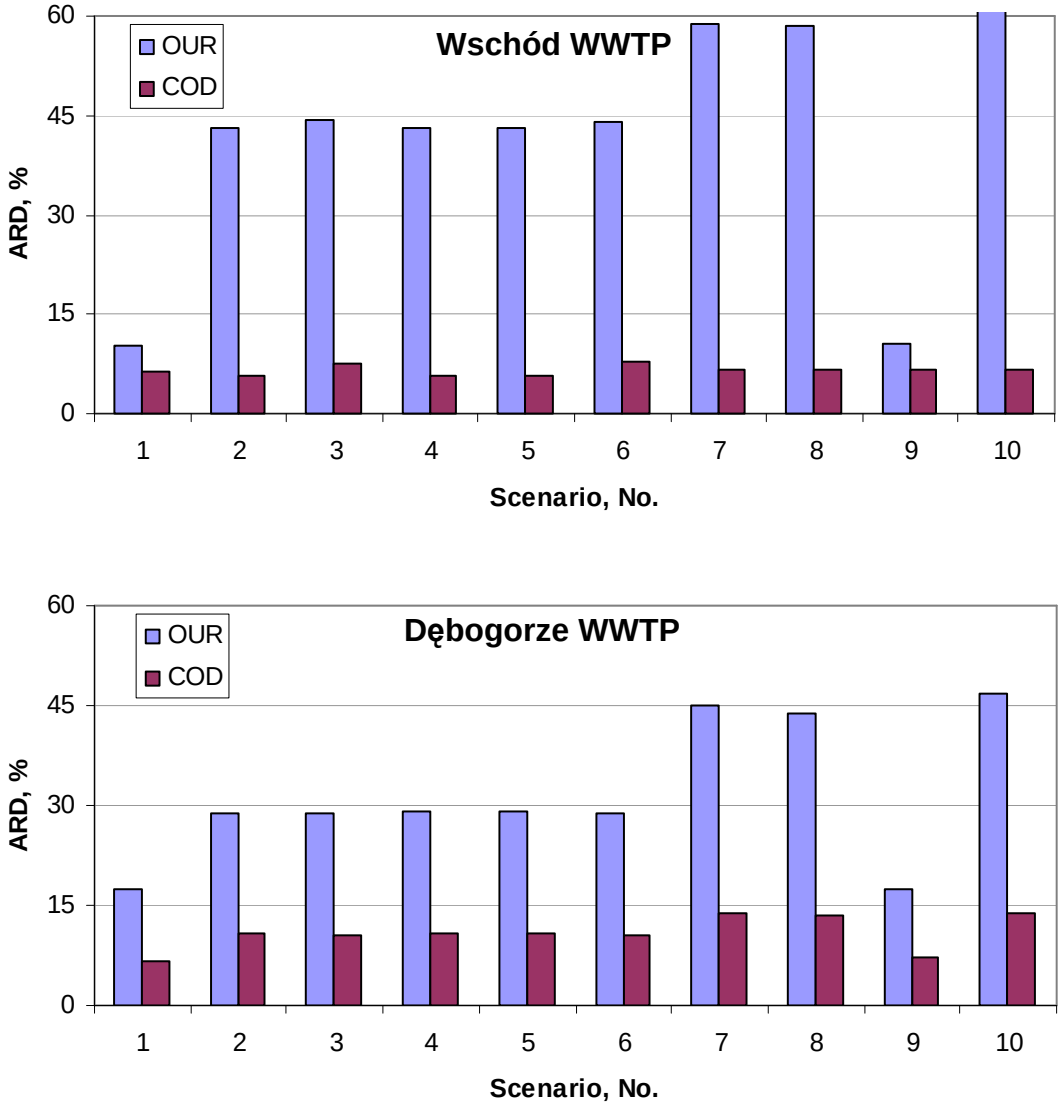


Figure 4.22. Results of the modified ASM2d optimization using different combination of stoichiometric and kinetic coefficients in various simulation scenario for the OUR batch tests with settled wastewater during the calibrated summer/fall study periods at the "Wschód" and "Dębogórze" WWTPs.

Table 4.21. Results of the modified ASM2d optimization using the best combination of stoichiometric and kinetic coefficients in various simulation scenario for the OUR batch tests with settled wastewater during all study periods at the "Wschód" and "Dębogórze" WWTPs

Scenario	WWTP					
	Wschód (Y_H = 0.65 from OUR test)			Dębogórze (Y_H = 0.68 from OUR test)		
	k_{hyd}	$k_{hyd,r}$	K_x	k_{hyd}	$k_{hyd,r}$	K_x
	1/d	1/d	–	1/d	1/d	-
Winter 1	0.8*	10	0.1	X	X	X
Winter 2	2.3	10	0.1	X	X	X
Summer/Fall 1	2	10	0.1	2.2	10	0.1
Summer/Fall 2	1.6	10	0.1	2.1	10	0.1
Spring 1	1.2	10	0.1	2.0	10	0.1
Spring 2	1.8	10	0.1	1.9	10	0.1
Average value	**1.8**	**10**	**0.1**	**2.1**	**10**	**0.1**

Note: * k_{hyd} not taken into concideration in calculation of average value

Table 4.22. Comparasion of the stoichiometric and kinetic coefficients in the calibrated models of original and modified ASM2d

Symbol	Unit	Calibrated value at ASM2d	Calibrated value at modified ASM2d	
			Wschód	Dębogórze
Stoichiometric coefficients				
Y_H	gCOD/gCOD	0.625	0.65	0.68
Kinetic coefficients of hydrolysis				
K_{hyd}	d^{-1}	2.5	2	
$K_{hyd,r}$	d^{-1}	-	10	
η_{fe}	–	0.1	0.1	
η_{fer}	–	-	0.4	
K_x	–	0.2	0.1	
K_{xr}	–	-	0.03	
$\eta_{NO3, Hyd}$	–	0.6	0.6	
$\eta_{NO3, Hydr}$	–	-	0.4	
K_{O2}	g O_2 / m^3	0.2	0.2	
K_{NO3}	g N/m^3	0.5	0.5	

Note: *Shaded area marks calibrated value*

The results of the modified ASM2d optimalization using different combination of stoichiometric and kinetic coefficients in various simulation scenario for the OUR batch tests with settled wastewater during the calibrated summer/fall study periods at the "Wschód" and "Dębogórze" WWTPs are presented in Figure 4.22 and listed in Table 4.20. The best combination of stoichiometric and kinetic coefficients for the OUR batch tests with settled wastewater were estimated from all studied sessions at both plants. The k_{hyd} from one of winter study period at "Wschód" WWTPs had not been taken into concideration in calculation of average value, because was too low in comparison to other kinetic hydrolysis rate constant coefficients from both plants (Table 4.21). The final result of optimalization step for OUR batch tests in the modified ASM2d was adjusted accordingly average value of kinetic parameters for both "Wschód" and "Dębogórze" WWTPs simultaneously. The results of parameter estimation based on conventional OUR tests have revealed that hydrolysis rate constant (k_{hyd} and $k_{hyd,r}$) in a two step-hydrolysis model, were 2.0 and 10 d^{-1}, respectively, whereas the estimated single hydroliysis rate constant in ASM2d was 2.5 d^{-1}. The stoichiometric and kinetic coefficients in original and modified version of ASM2d are listed in Table 4.22. The comparison of both examined models predictions vs. measured OURs during the two parallel types of batch tests are presented in Figure 4.23-4.24 (a-c) and Figure 4.25-4.26 (a-c) for the "Wschód" and "Dębogórze" WWTP, respectively.

The predictive capabilities of the original and modified ASM2d have been confirmed by average relative deviation (ARD). The results of average ARD differences between both models from OUR batch tests were further discussed in next Section 4.3.5. Table 4.23 and 4.24 contains the average values of ARD for the models predictions and measured data from OUR batch tests with the settled wastewater without pretreatment and after coagulation-flocculation. In general, the new model better predicts the COD concentrations and OUR behaviors in conventional OUR batch tests with the settled wastewater without pretreatment and coagulation-flocculation at both studied plants (see Figure 4.23-4.26 a-c).

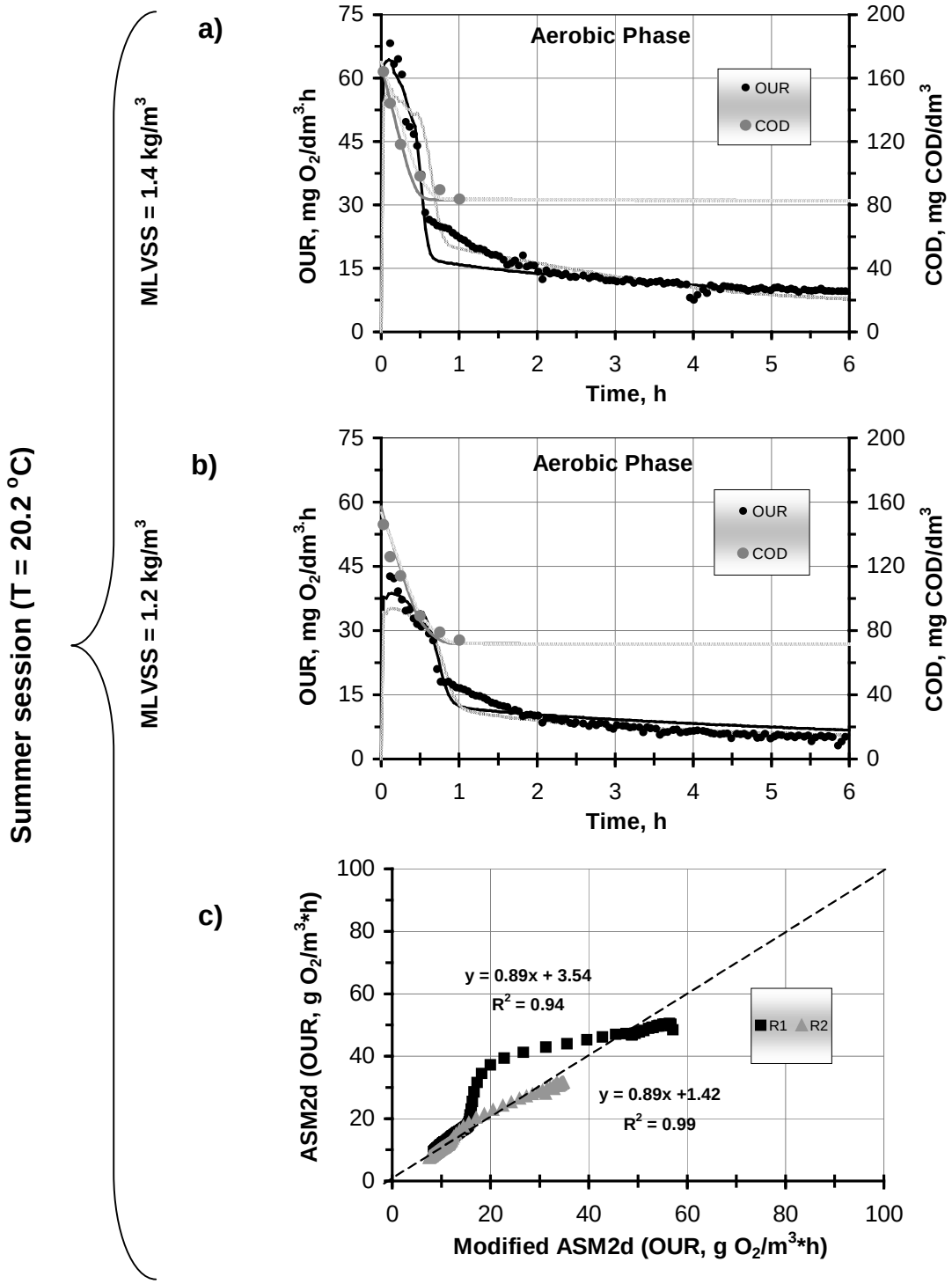


Figure 4.23. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d after optimalization step for OUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a) the settled wastewater without pretreatment, (b) the settled wastewater after coagulation-flocculation and (c) deviations between the original and modified ASM2d with respect to the OUR predictions of settled wastewater without pretreatment (R1) or after coagulation-flocculation (R2).

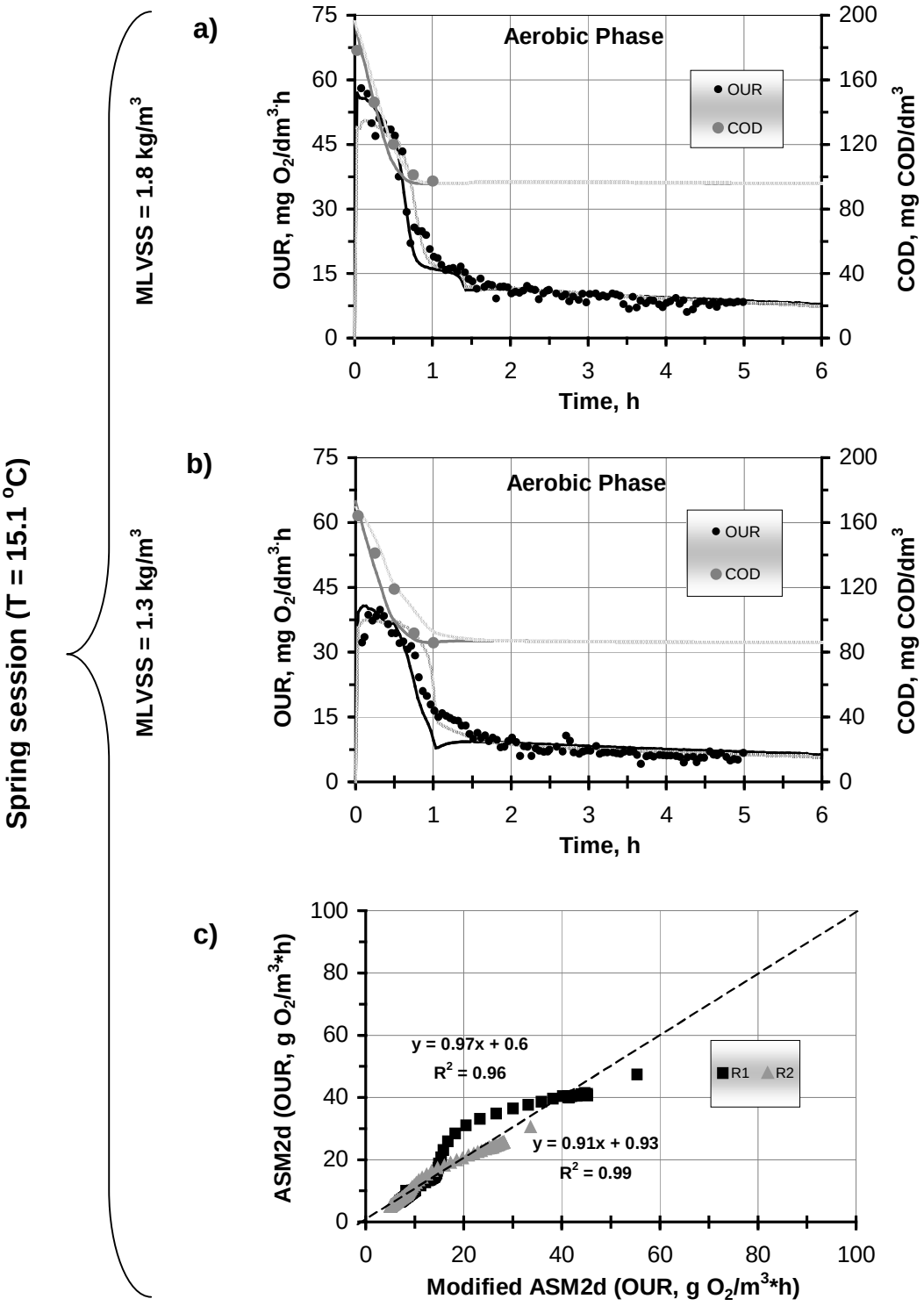


Figure 4.24. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d after optimalization step for OUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a) the settled wastewater without pretreatment, (b) the settled wastewater after coagulation-flocculation and (c) deviations between the original and modified ASM2d with respect to the OUR predictions of settled wastewater without pretreatment (R1) or after coagulation-flocculation (R2).

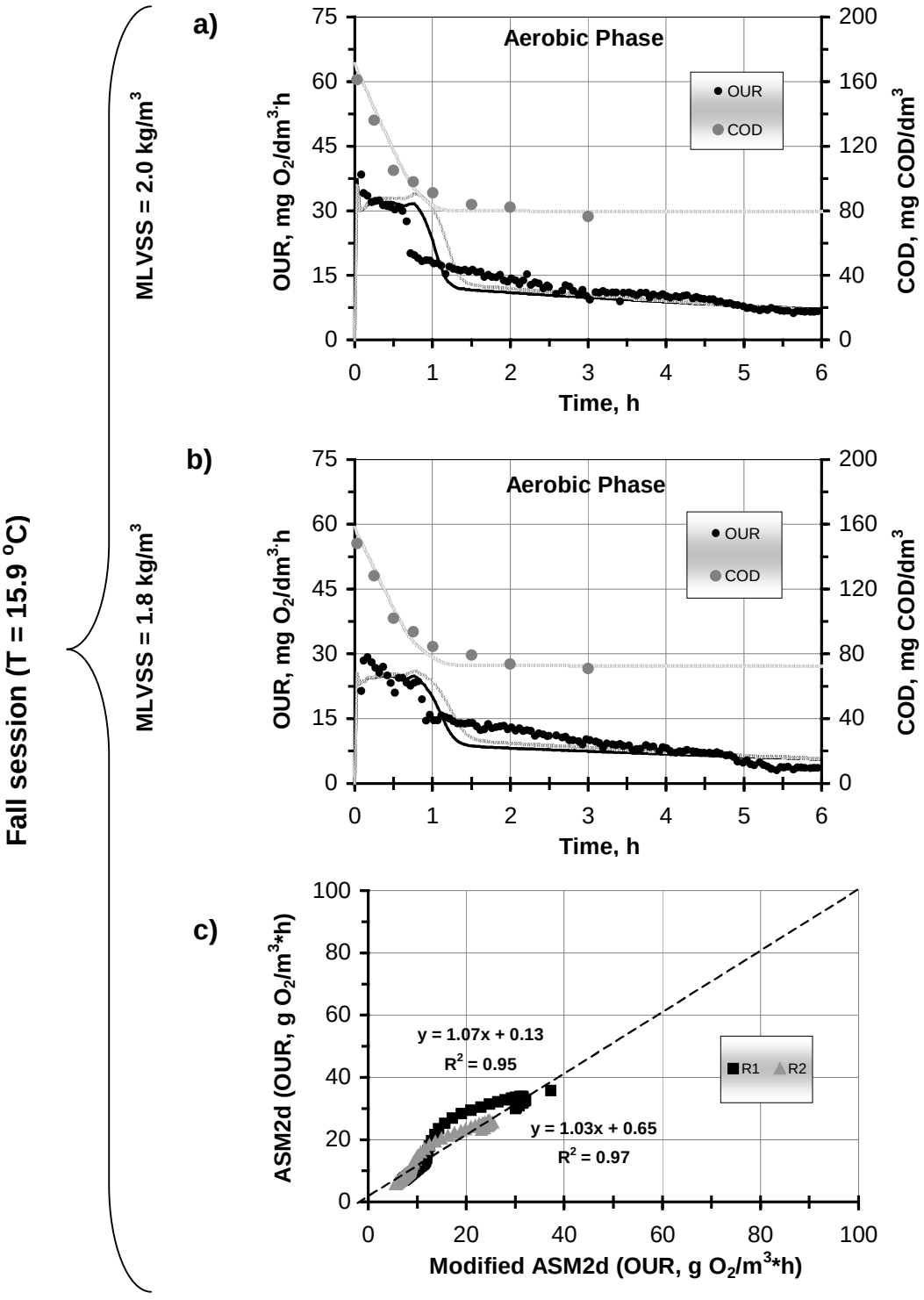


Figure 4.25. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d after optimalization step for OUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a) the settled wastewater without pretreatment, (b) the settled wastewater after coagulation-flocculation and (c) deviations between the original and modified ASM2d with respect to the OUR predictions of settled wastewater without pretreatment (R1) or after coagulation-flocculation (R2).

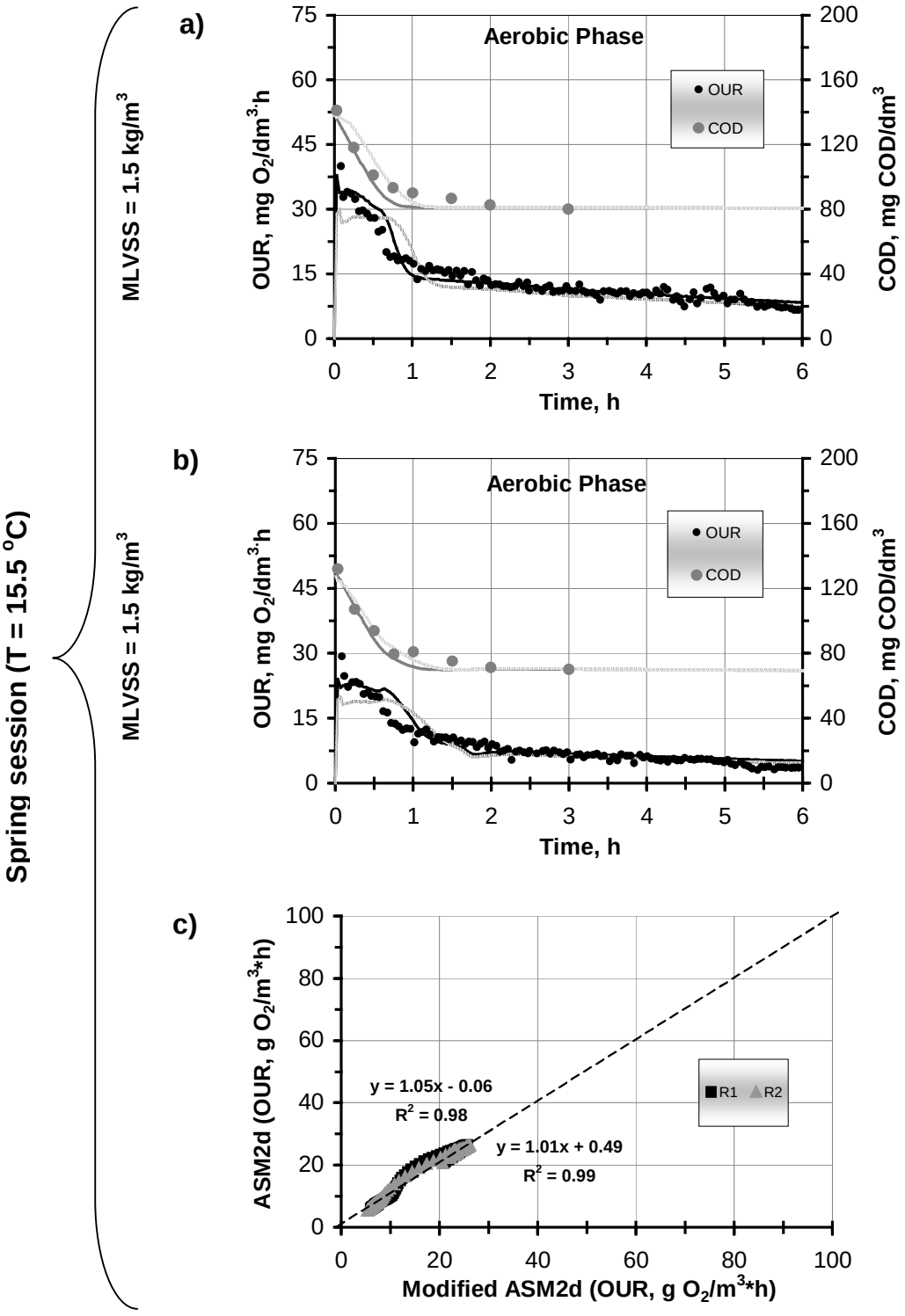


Figure 4.26. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d after optimalization step for OUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a) the settled wastewater without pretreatment, (b) the settled wastewater after coagulation-flocculation and (c) deviations between the original and modified ASM2d with respect to the OUR predictions of settled wastewater without pretreatment (R1) or after coagulation-flocculation (R2).

4.3.5. Comparison of the original and modified ASM2d predictions

The predictive capabilities of the original and modified ASM2d have been confirmed by average relative deviation (ARD). Table 4.23 and 4.24 contains the average values of ARD for the models predictions and measured data from the NUR, PRR, anoxic/aerobic PUR and OUR batch tests with the settled wastewater without pretreatment and after coagulation-flocculation at both studied plants.

At "Wschód" WWTP accurate predictions of OURs in the modified ASM2d were confirmed by low ARDs, which were in the range of 9.7-15.8% and 11.8-30.3% in the samples with the settled wastewater without pretreatment and after coagulation-flocculation, respectively. For comparison, the corresponding errors obtained with the original ASM2d varied in the range of 11.3-29.5% and 18.9-45.8%. The measured process rates of OURs, better fitted by the modified ASM2d than its original version during all study period at the "Wschód" WWTP (Table 4.23). The accurate predictions in modified ASM2d resulted in the lowest ARD values (9.7-11.8%) during summer (calibration) study period of the OUR tests with the settled wastewater without pretreatment and after coagulation-flocculation, respectively. Much higher ARDs (14.6-30.3%) were calculated for the results of similar experiments carried out during winter study period. In the case of the SCOD utilization during the OUR tests at "Wschód" WWTP, the ARD values showed accurate predictions in both models. The errors for the original vs. modified ASM2d amounted to 4.3-4.8% vs. 2.7-5.5% and 4.4-5.2% vs. 3.1-5.4% for the settled wastewater without pretreatment and after coagulation-flocculation, respectively.

At "Dębogórze" WWTP, discrepancies between the modified ASM2d predictions and measured data also occurred at low ARDs during OUR tests. The errors with the original ASM2d for the OUR simulation with the settled wastewater without pretreatment and after coagulation-flocculation varied in the range of 12.4-16.6% and 20.8-29.1% respectively, whereas lower values e.g. 13.1-15.8% and 16.4-23.4% (except one case) were calculated for the results of the same experiments carried out with the modified ASM2d. In the case of the SCOD utilization during the OUR tests at "Dębogórze" WWTP, the ARD values showed no or only minor difference between the measured and simulated results in both models. The errors for the original vs. modified ASM2d amounted to 4.3-4.8% vs. 2.7-5.5% and 4.4-5.2% vs. 3.1-5.4% for the settled wastewater without pretreatment and after coagulation-flocculation, respectively. The comparison of both examined models predictions vs. measured OURs during the two parallel types of batch tests is presented in Figure 4.23-4.26 (a-c) for the "Wschód" and "Dębogórze" WWTP.

Table 4.23. The ARD values for model predictions of the principal process rates investigated in the batch experiments with the settled wastewater without pretreatment and after coagulation-flocculation (c-f) from "Wschód" WWTP

Experiment	Study period	Process rate	ARD, %			
			Wschód WWTP			
			Settled wastewater		Settled wastewater (c-f)	
			ASM2d	Modified ASM2d	ASM2d	Modified ASM2d
Conventional NUR	Winter	Nitrate utilization	27.3	33.6	12.8	25.7
		SCOD utilization	15.2	16.0	18.3	16.4
	Summer	Nitrate utilization	7.4	12.1	4.0	4.4
		SCOD utilization	12.2	11.6	12.2	11.6
	Spring	Nitrate utilization	20.0	27.1	7.2	7.0
		SCOD utilization	8.7	6.0	6.4	6.8
PRR/anoxic PUR	Winter	Phosphate release	4.7	5.4	27.6	28.7
		Phosphate uptake	15.2	27.6	6.7	8.9
		Nitrate utilization	18.6	31.2	8.1	12.2
	Summer	Phosphate release	4.6	4.9	14.7	15.3
		Phosphate uptake	6.5	14.1	8.4	17.3
		Nitrate utilization	11.9	25.4	5.1	9.7
	Spring	Phosphate release	8.7	10.4	22.6	22.1
		Phosphate uptake	27.2	28.4	36.7	31.9
		Nitrate utilization	9.0	20.0	5.7	8.5
PRR/aerobic PUR	Winter	Phosphate release	5.5	4.1	45.2	45.4
		Phosphate uptake	29.5	44.3	10.5	24.0
		Ammonia utilization	3.4	2.9	3.3	4.2
		Oxygen uptake	7.2	8.3	12.0	6.6
	Summer	Phosphate release	8.8	8.2	11.5	11.8
		Phosphate uptake	12.8	16.1	6.2	11.1
		Ammonia utilization	4.0	3.6	3.2	3.2
		Oxygen uptake	17.7	13.3	12.6	12.5
	Spring	Phosphate release	7.1	7.3	36.5	37.4
		Phosphate uptake	55.7	59.3	49.5	59.5
		Ammonia utilization	4.0	4.3	3.7	3.5
		Oxygen uptake	9.4	9.5	10.3	10.6
Conventional OUR	Winter	Oxygen uptake	29.5	14.6	45.8	30.3
		SCOD utilization	4.3	3.8	5.0	3.1
	Summer	Oxygen uptake	11.3	9.7	18.9	11.8
		SCOD utilization	4.8	5.5	5.2	5.4
	Spring	Oxygen uptake	16.2	15.8	23.3	19.2
		SCOD utilization	4.3	2.7	4.4	4.5

Table 4.24. The ARD values for model predictions of the principal process rates investigated in the batch experiments with the settled wastewater without pretreatment and after coagulation-flocculation (c-f) from "Dębogórze" WWTP

Experiment	Study period	Process rate	ARD, %			
			Dębogórze WWTP			
			Settled wastewater		Settled wastewater (c-f)	
			ASM2d	Modified ASM2d	ASM2d	Modified ASM2d
Conventional NUR	Fall	Nitrate utilization	28.9	27.3	4.4	6.0
		SCOD utilization	8.2	8.5	6.8	6.6
	Spring	Nitrate utilization	4.6	21.6	3.6	8.6
		SCOD utilization	5.7	5.3	4.9	4.9
PRR/anoxic PUR	Fall	Phosphate release	6.4	6.4	12.6	12.9
		Phosphate uptake	6.2	8.4	5.3	8.6
		Nitrate utilization	5.5	15.1	4.7	11.5
	Spring	Phosphate release	10.8	10.3	18.2	18.8
		Phosphate uptake	5.8	6.4	8.9	10.0
		Nitrate utilization	6.4	15.4	8.5	7.5
PRR/aerobic PUR	Fall	Phosphate release	7.4	7.9	11.0	11.8
		Phosphate uptake	11.7	17.6	8.6	19.9
		Ammonia utilization	4.8	4.6	4.2	3.8
		Oxygen uptake	23.3	17.9	19.4	17.9
	Spring	Phosphate release	12.0	12.1	34.5	34.4
		Phosphate uptake	7.8	9.7	12.1	16.5
		Ammonia utilization	2.6	2.3	4.1	3.9
		Oxygen uptake	20.2	14.6	14.7	13.5
Conventional OUR	Fall	Oxygen uptake	16.6	15.8	29.1	23.4
		SCOD utilization	5.5	5.2	5.0	5.2
	Spring	Oxygen uptake	12.4	13.1	20.8	16.4
		SCOD utilization	4.5	4.9	5.4	4.3

The model predictions of the principal process rates investigated in the batch experiments, including NUR, PRR, anoxic/aerobic PUR, OUR, AUR and SCOD utilization, have showed variable effects. The ARDs with the OUR simulation obtained during PRR and aerobic PUR tests better predicted in the modified ASM2d than its original version, whereas PRR, AUR and SCOD utilization showed no or only minor difference between the measured and simulated results in both samples of settled wastewater (Table 4.23 and 4.24). In the case of NUR simulation obtained in both experiments (the conventional NUR and PRR, anoxic PUR tests) with the settled wastewater without pretreatment, the ARDs in modified ASM2d were slightly higher, 12.1-36.6% and 20.0-31.2% ("Wschód" WWTP) vs. 21.6-27.3% and 15.1-15.4% ("Dębogórze" WWTP), than experiments in original ASM2d, 7.4-27.3% and 9.0-18.6% ("Wschód" WWTP) vs. 4.6-29.0% and 5.5-6.4% ("Dębogórze" WWTP). For comparison, simulations of the corresponding NURs with the settled wastewater after coagulation-flocculation in the original and modified ASM2d, showed (in general) only minor difference of the ARD values between the measured and simulated results in both models (Table 4.23 and 4.24). Similar discrepancies of ARD values for the examined models predictions of the anoxic/aerobic PURs were estimated at both plants. The average ARDs between the measured data and original vs. modified ASM2d predictions for the sample of settled wastewater without pretreatment varied within the range of 6.5-27.2% vs. 14.1-28.4% (anoxic PUR) and 12.8-55.7% vs. 16.1-59.3% (aerobic PUR) at the "Wschód" WWTP. The corresponding ARD values for the sample of settled wastewater after coagulation-flocculation were 6.7-36.7% vs. 8.9-31.9% (anoxic PUR) and 6.2-49.5% vs. 11.1-59.5% (aerobic PUR), respectively. At the "Dębogórze" WWTP for the anoxic PUR tests, no significant differences were observed in the prediction capabilities of ASM2d in comparision to modified model, whereas for the aerobic PUR discrepancies were noted. In this respect the ARDs between the measured data and original vs. modified ASM2d simulations for the aerobic PUR tests varied within the range of 7.8-11.7% vs. 9.7-17.6% (settled wastewater) and 8.6-12.1% vs. 16.5-19.9% (sample after coagulation-flocculation). The comparison of both examined models predictions vs. measured NURs, PRRs, anoxic/aerobic PURs, AURs and SCOD utilizations in two parallel types of batch tests are presented in following Figures 4.27-4.32, at both studied plants. The full experimental data together with mathematical modeling in original and modified ASM2d obtained from the "Wschód" and "Debogórze" WWTP can be found in Appendix 4.

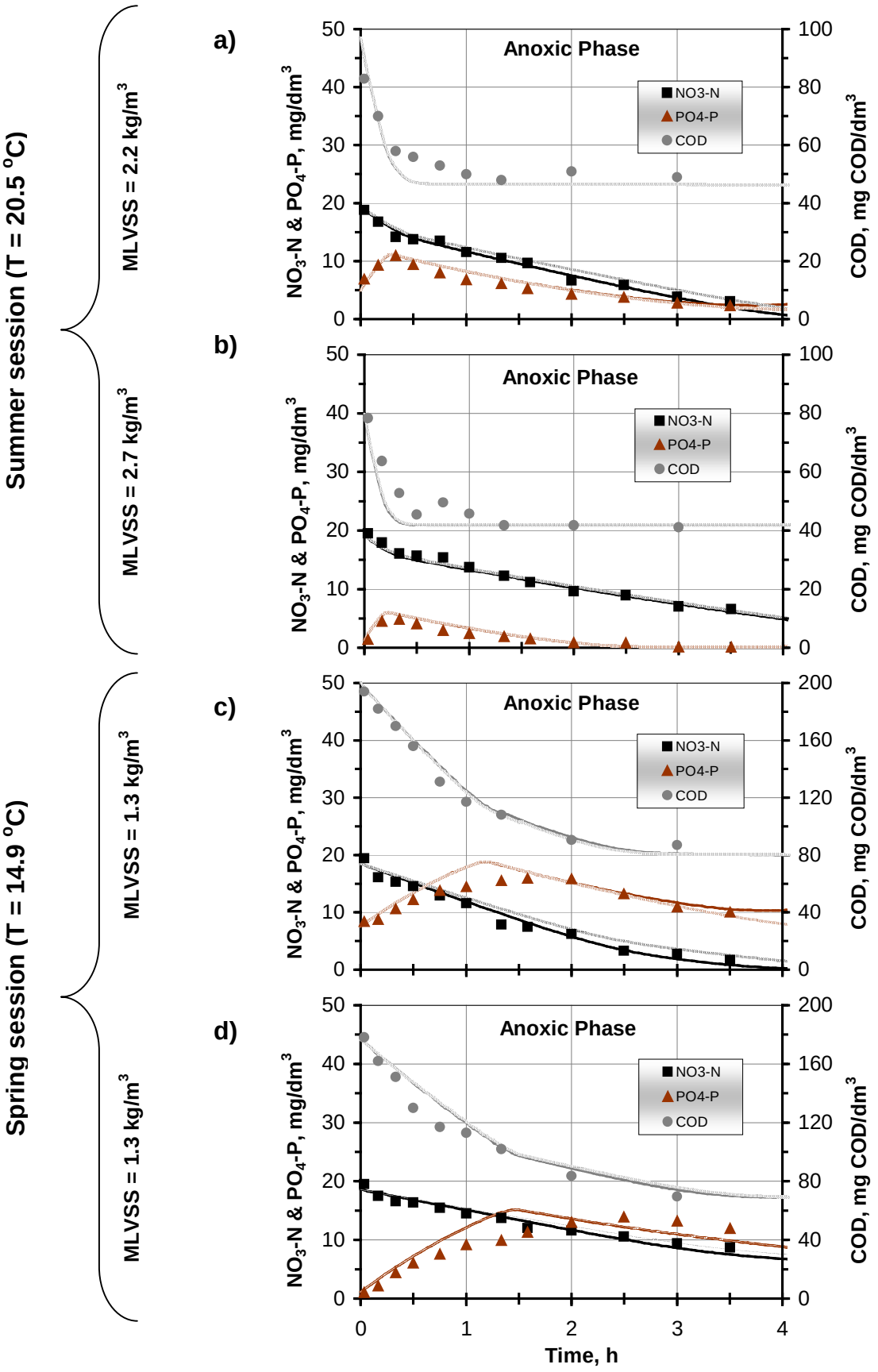


Figure 4.27. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for denitrification tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

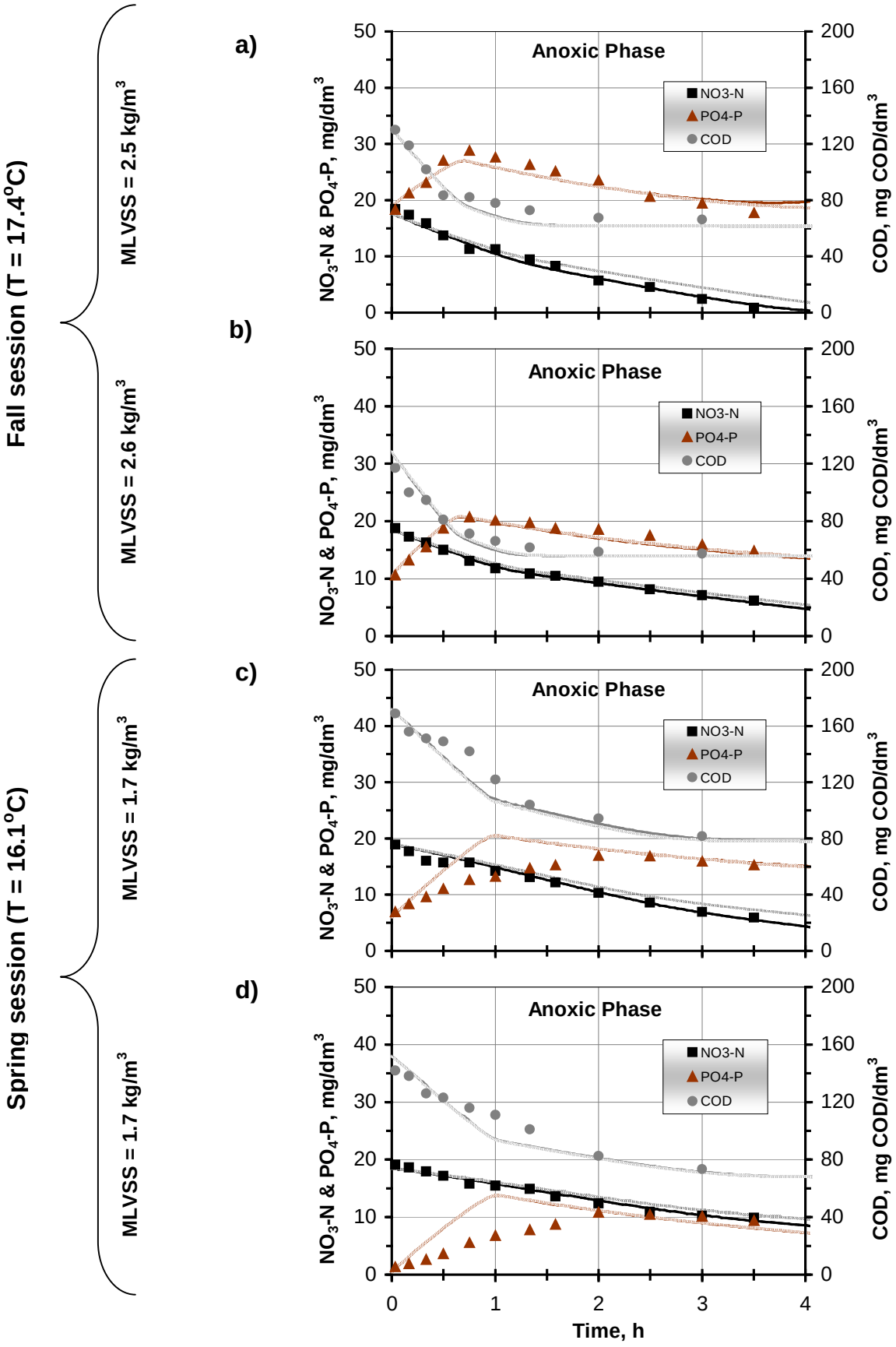


Figure 4.28. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for denitrification tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

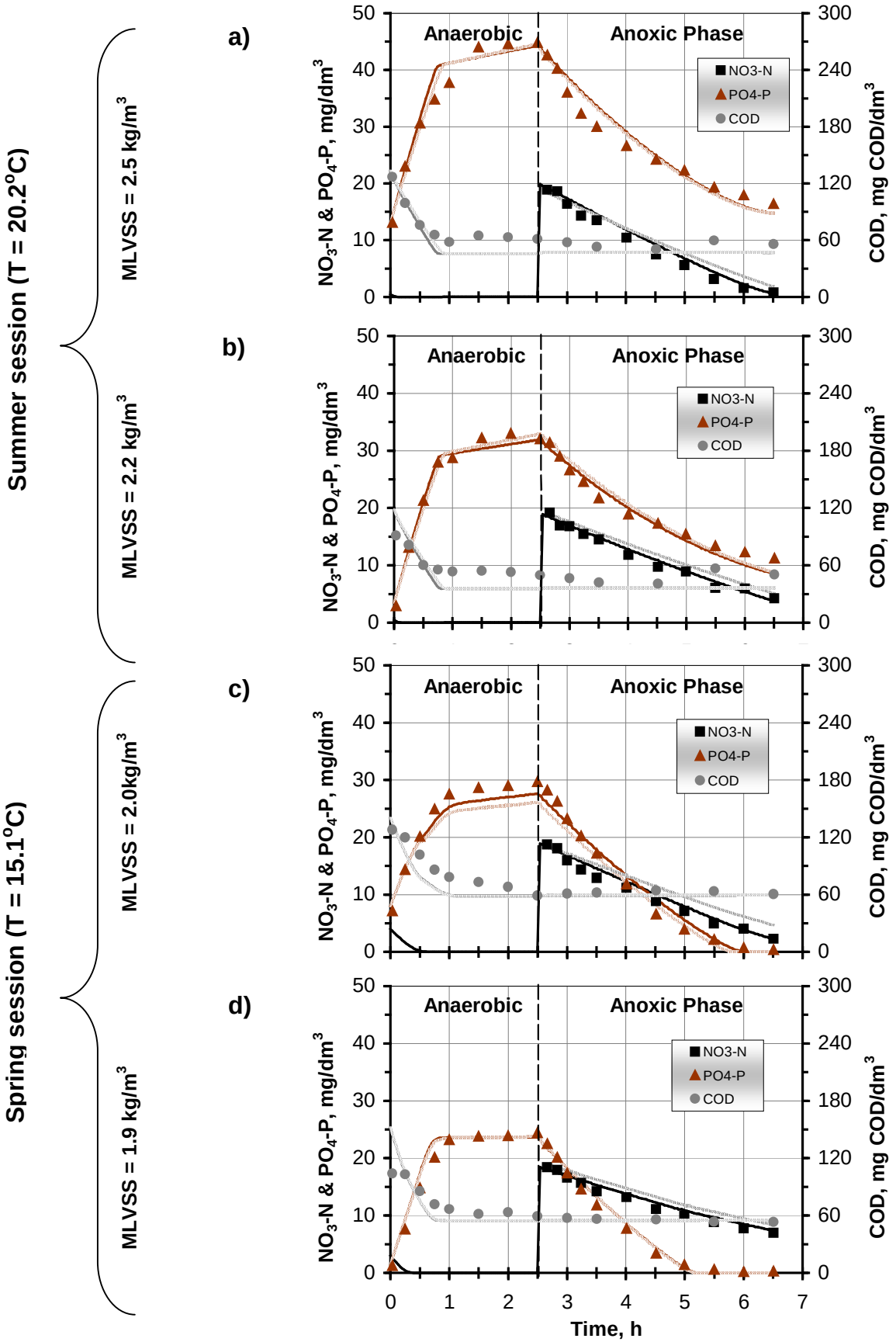


Figure 4.29. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for PRR and anoxic PUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

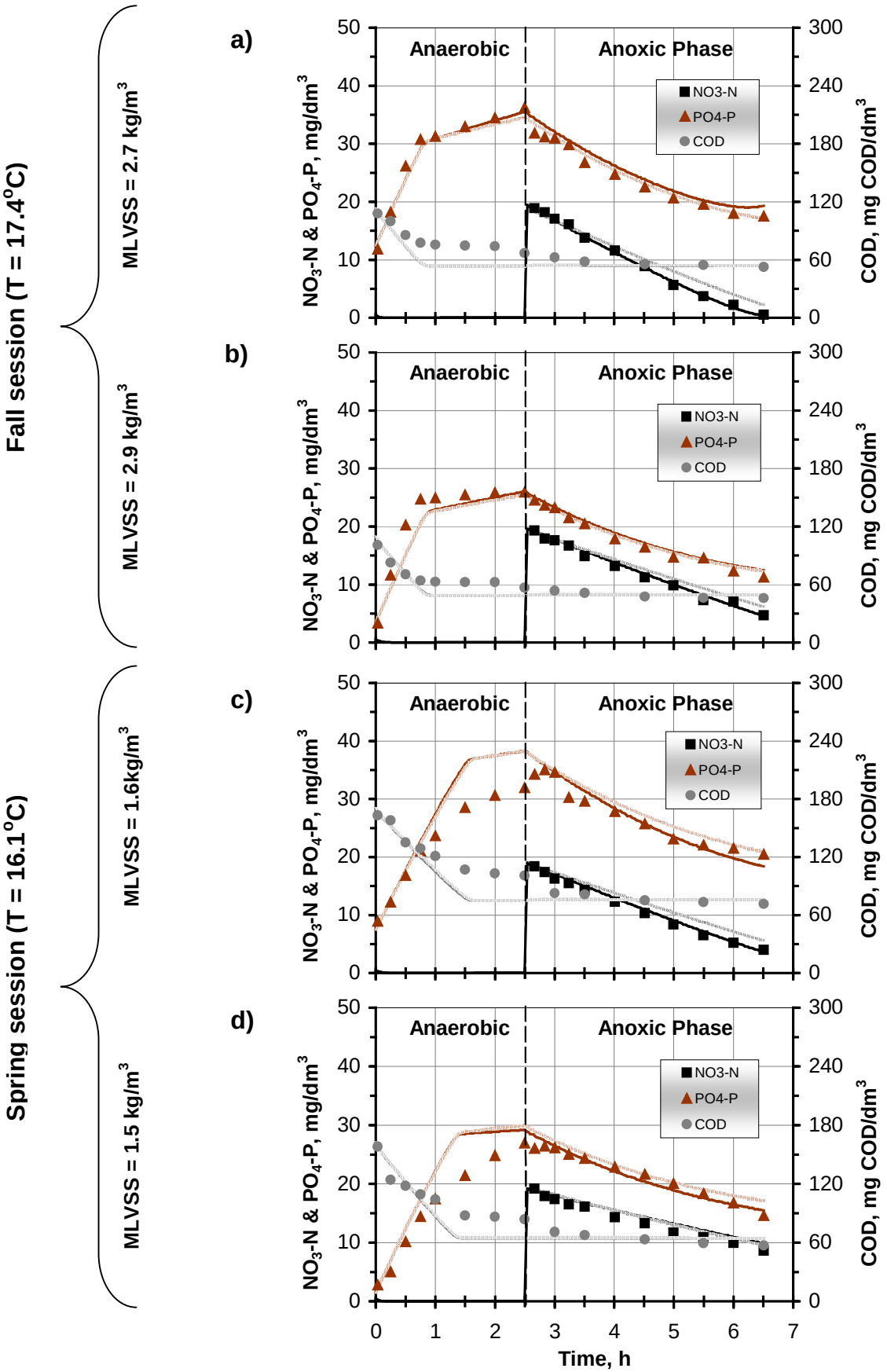


Figure 4.30. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for PRR and anoxic PUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

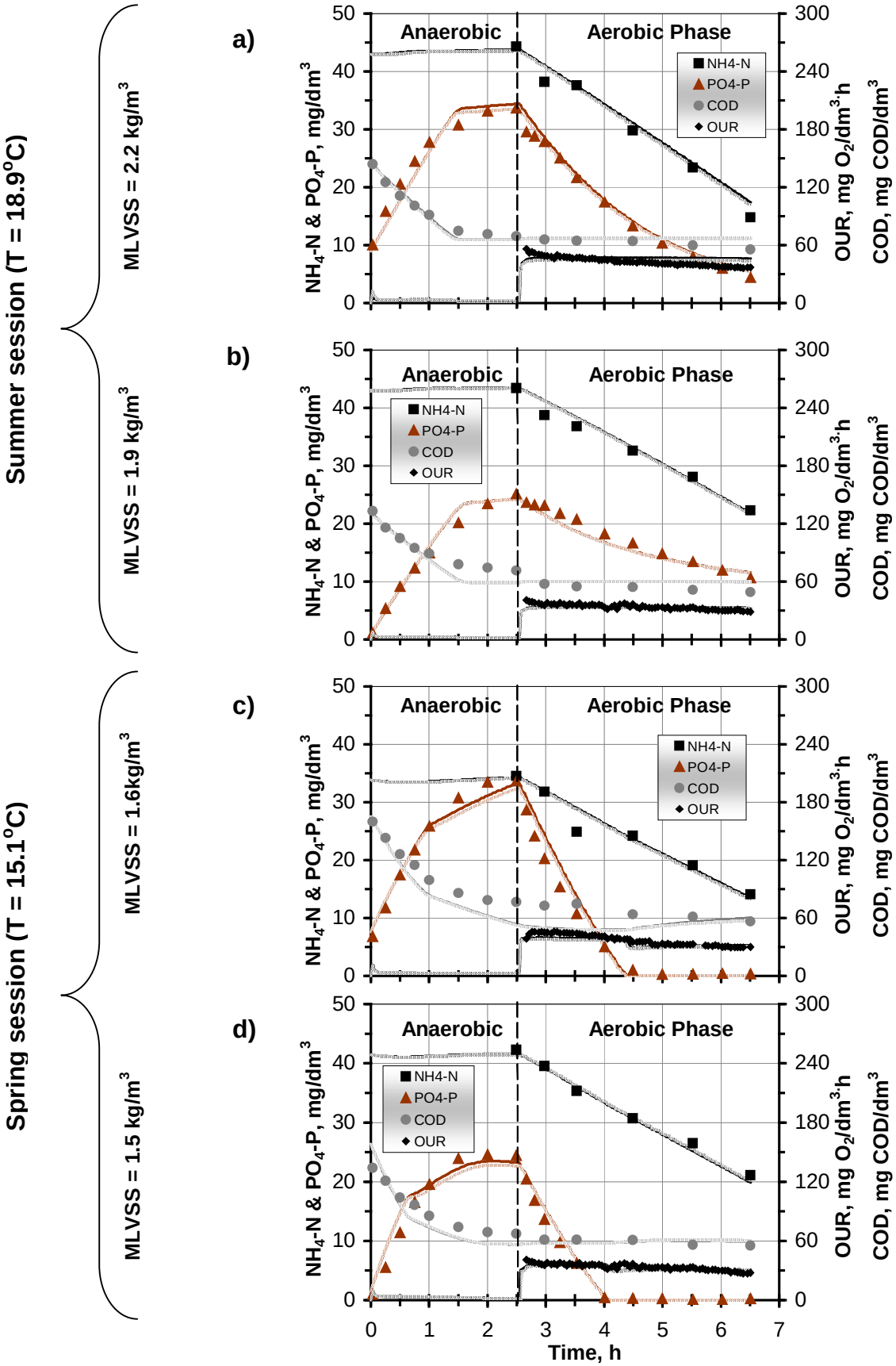


Figure 4.31. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for PRR and aerobic PUR tests in two parallel reactors with mixed liquor from "Wschód" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

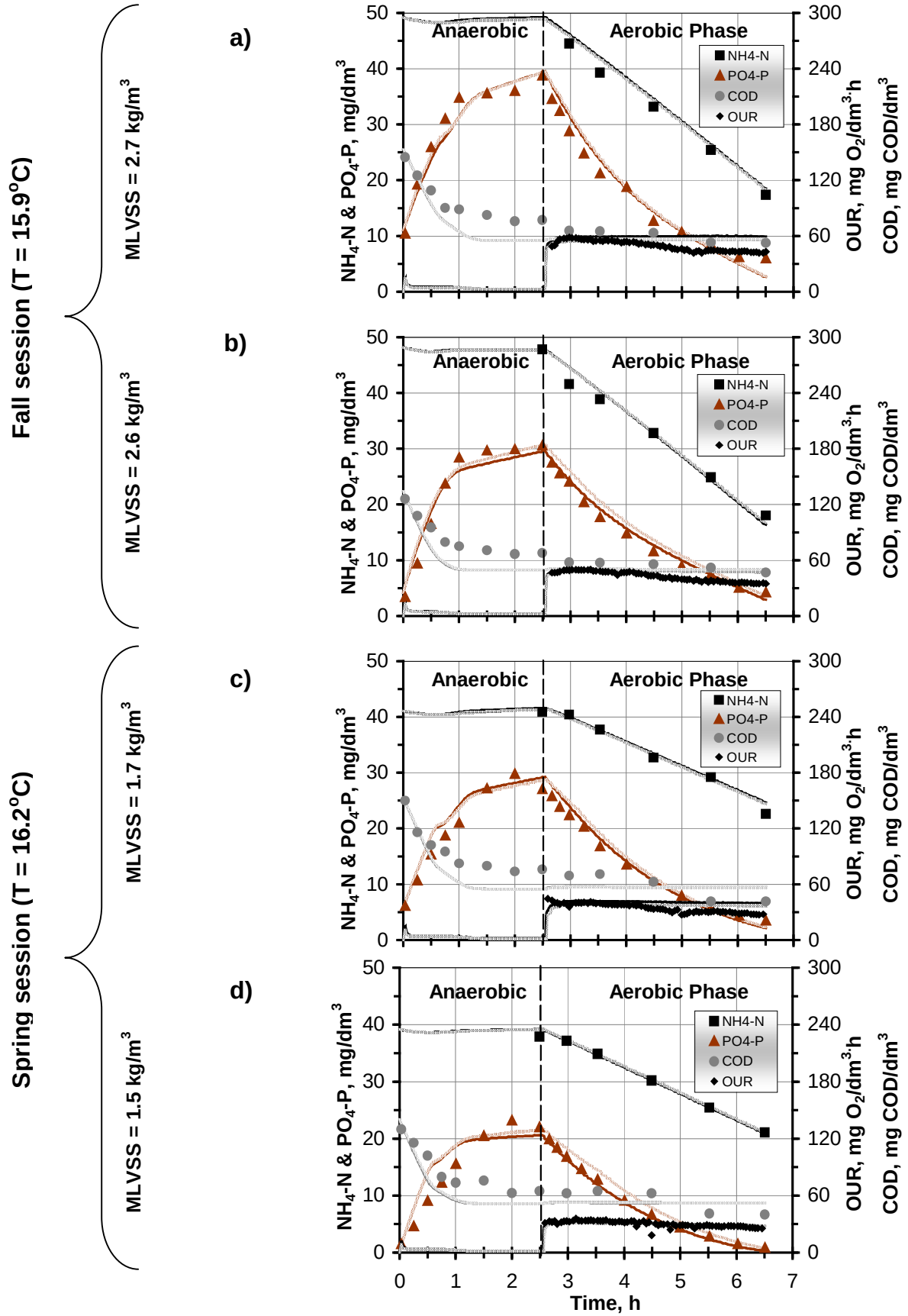


Figure 4.32. Measured data vs. original (— solid lines) and modified (— shaded lines) ASM2d predictions for PRR and aerobic PUR tests in two parallel reactors with mixed liquor from "Dębogórze" WWTP: (a,c) the settled wastewater without pretreatment, (b,d) the settled wastewater after coagulation-flocculation.

5

Conclusions

Conclusions

From this study, the following conclusions can be derived:

1. A novel procedure, based on the standard batch experiments and pretreatment of wastewater sample with coagulation-flocculation method, has been developed to evaluate the effects of colloidal and particulate organic compounds on denitrification and EBPR in activated sludge systems.
2. The colloidal and particulate organic fractions play an important role in enhancing these two biochemical processes. Except for one process (phosphate release), the removal of these fractions resulted in the reduced rates of biochemical processes. The average reductions were as follows:
   – 24% and 35%, respectively, for the NUR1 and NUR2 during the conventional NUR tests, and 30% for the NUR during the anoxic PUR test;
   – 32% and 25%, respectively, for the anoxic and aerobic PURs;
   – 13% for the average OUR during the aerobic PUR tests, and 24% during the conventional OUR test (maximum measured values).
3. The results of the experimental part of this study provided a comprehensive database for calibration and evaluation of dynamic models of the hydrolysis process. A new mathematical model considering two-step hydrolysis process has been developed as an extension of ASM2d. The new model incorporates an additional variable (rapidly hydrolyzable substrate, X_{SH}) and three processes – hydrolysis of X_{SH} under aerobic, anoxic and anaerobic conditions. The results of parameter estimation based on conventional OUR tests have revealed that hydrolysis rate constant (k_{hyd} and $k_{hyd,r}$) in a two step-hydrolysis model, were 2.0 and 10 d^{-1}, respectively, whereas the estimated single hydroliysis rate constant in ASM2d was 2.5 d^{-1}.
4. In comparison with ASM2d, the new model has better predicted the dissolved oxygen behaviour in the conventional OUR tests at both studied plants. The average ARDs were 17.2% and 27.6% (original ASM2d) vs. 13.8% and 20.2% (modified ASM2d), respectively, for the settled wastewater without pretreatment and after coagulation-flocculation. In contrast, the ARDs for COD concentrations measured during the conventional OUR tests were very similar (4.4-5.0%) for both models.
5. Predictions of both models have also been examined based on the nitrification, denitrification and EBPR measurments in the batch tests. The new model has showed variable effects on the improvements in predicted behaviour of NH_4-N, NO_3-N and PO_4-P. The smallest differences between the average ARDs for both models (0.3 and 0.6%) were found for the AUR and PRR, whereas the largest differences (5.9 and 6.6%) were found for the PUR and NUR.

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List of appendices

The four appendices, summarized below, has been placed on a CD included at the end of the Ph.D. Thesis.

Appendix 1 – contains the detailed description of ASM2d. In 15 pages of Appendix 1 can be found 6 Tables, 1 Figure and 2 Equation. In Table 1.1-1.2 are presented the list of the ASM2d soluble and particulate components with their definitions/descriptions. Stoichiometric and composition matrixes for the model components and kinetic rate expressions contain Tables 1.3-1.4. Kinetic and stoichiometric description in the ASM2d are presented in Table 1.5 and 1.6. The Figure 1.1 shows transformations of pollutants in activated sludge reactors according to earlier ASM2. The practical meaning of reactions defined in ASM2d as well as the most important model limitations and proposed modification are presented in Section 1.1 and 1.2, respectively.

Appendix 2 – contains the experimental data obtained in winter, summer and spring study sessions from "Wschód" WWTP in Gdańsk during the period 2007 - 2009. In 85 pages of Appendix 2 can be found Tables and Figures together with detailed experimental data description from each batch test. In Tables and Figures are presented the results of all the specific rates, i.e. NURs, PRRs, anoxic/aerobic PURs and OURs with additional on-line mesurments (e.g. T, DO, ORP, pH), observed in the three kinds of batch experiments with the settled wastewater without pretreatment and after coagulation-flocculation.

Appendix 3 – contains the experimental data obtained in fall and spring study sessions from "Dębogórze" WWTP in Gdynia during the period 2009 - 2010. In 60 pages of Appendix 3 can be found Tables and Figures together with detailed experimental data description from each batch test. In Tables and Figures are presented the results of all the specific rates, i.e. NURs, PRRs, anoxic/aerobic PURs and OURs with additional on-line mesurments (e.g. T, DO, ORP, pH), observed in the three kinds of batch experiments with the settled wastewater without pretreatment and after coagulation-flocculation.

Appendix 4 – contains the experimental data together with mathematical modeling in original and modified ASM2d obtained from the "Wschód" and "Debogórze" WWTP during the study period 2007 – 2010. In 21 pages of Appendix 4 can be found Figures with experimental data and mathematical modeling in both models of ASM2d from each batch test. In Figures are presented the results of the experimental data v.s. mathematical modeling (original/modified ASM2d) observed in all the batch experiments i.e. NURs, PRRs, anoxic/aerobic PURs and OURs, with the settled wastewater without pretreatment and after coagulation-flocculation.