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A simplified method to assess structurally identifiable parameters in Monod-based activated sludge models

Britta Petersen[a], Krist Gernaey[a,b,*], Martijn Devisscher[a], Denis Dochain[c], Peter A. Vanrolleghem[a]

[a] *Biomath, Ghent University, Coupure Links 653, B-9000 Gent, Belgium*
[b] *CAPEC, Chemical Engineering Department, Technical University of Denmark, Building 229, DK-2800 Lyngby, Denmark*
[c] *CESAME, Université Catholique de Louvain, Bâtiment Euler, 4 Av. Georges Lemaître, B-1348 Louvain-la-Neuve, Belgium*

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Abstract

The first step in the estimation of parameters of models applied for data interpretation should always be an investigation of the identifiability of the model parameters. In this study the structural identifiability of the model parameters of Monod-based activated sludge models (ASM) was studied. In an illustrative example it was assumed that respirometric (dissolved oxygen or oxygen uptake rates) and titrimetric (cumulative proton production) measurements were available for the characterisation of nitrification. Two model structures, including the presence and absence of significant growth for description of long- and short-term experiments, respectively, were considered. The structural identifiability was studied via the series expansion methods. It was proven that the autotrophic yield becomes uniquely identifiable when combined respirometric and titrimetric data are assumed for the characterisation of nitrification. The most remarkable result of the study was, however, that the identifiability results could be generalised by applying a set of ASM1 matrix based generalisation rules. It appeared that the identifiable parameter combinations could be predicted directly based on the knowledge of the process model under study (in ASM1-like matrix representation), the measured variables and the biodegradable substrate considered. This generalisation reduces the time-consuming task of deriving the structurally identifiable model parameters significantly and helps the user to obtain these directly without the necessity to go too deeply into the mathematical background of structural identifiability.

Keywords: Respirometry; Titrimetry; Structural identifiability; Activated sludge model; Parameter estimation

1. Introduction

Monod-type growth kinetics is most often used to describe experimental observations for biological substrate degradation processes. The activated sludge model no. 1 (ASM1) [1], a model that describes organic carbon source degradation and biological nitrogen removal, is a suitable illustration of the use of Monod growth kinetics to describe degradation processes. In this study, the focus will be on the structural identifiability of the parameters in Monod-based models.

A study of the structural identifiability of model parameters prior to practical model application, e.g. in the frame of parameter estimation or model calibration, is very important in order to obtain reliable parameter estimates. The key question of the structural and practical identifiability analysis can be formulated as follows [2]: "Assume that a certain number of state variables are available for measurements; on the basis of the model structure (structural identifiability) or on the

*Corresponding author. CAPEC, Chemical Engineering Department, Technical University of Denmark, Building 229, DK-2800 Lyngby, Denmark. Tel.: +45-45-25-28-00; fax: +45-45-93-29-06.

E-mail address: kvg@kt.dtu.dk (K. Gernaey).

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Nomenclature

$\underline{a}_k(\underline{p})$	Taylor series derivative
ASM	activated sludge model
ASM1	activated sludge model no. 1
f_{BA}	fraction of autotrophic biomass
f_{BH}	fraction of heterotrophic biomass
H_p	cumulative proton production (meq/l)
i	measured component (generalisation method)
i_{XB}	fraction of N in biomass (g N/g COD biomass)
j	the process that is considered (generalisation method)
k	the substrate under study (generalisation method)
K_S	heterotrophic half-saturation substrate concentration (mg COD/l)
K_{SA}	autotrophic half-saturation substrate concentration (mg N/l)
NH_4^+	ammonium
NO_2^-	nitrite
NO_3^-	nitrate
$\underline{p}$	parameter vector
r_{Hp}	proton production rate (meq/l min)
r_O	Oxygen uptake rate (mg O_2/l min)
$r_{O,ex}$	exogenous (=substrate degradation related) oxygen uptake rate (mg O_2/l min)
S_{ALK}	alkalinity (meq/l)
S_{NH}	ammonium nitrogen concentration (mg N/l)
$S_{NH}(0)$	ammonium nitrogen concentration at time $t=0$ (mg N/l)
S_{NO_3}	nitrate nitrogen concentration (mg N/l)
S_S	readily biodegradable substrate concentration (mg COD/l)
$S_S(0)$	readily biodegradable substrate concentration at time $t=0$ (mg COD/l)
S_O	dissolved oxygen concentration (mg O_2/l)
v_i	derivative
X	biomass concentration (mg COD/l)
$X(0)$	biomass concentration at time $t=0$ (mg COD/l)
X_{BA}	autotrophic biomass concentration
X_{BH}	heterotrophic biomass concentration
$\underline{y}$	output vector
Y_A	autotrophic biomass yield
Y_H	heterotrophic biomass yield
α_1	parameter combination
β_i	parameter combination
μ_{maxA}	maximum specific growth rate for autotrophic biomass (1/min)
μ_{maxH}	maximum specific growth rate for heterotrophic biomass (1/min)
v	stoichiometric coefficient (generalisation method)

basis of the type and quality of available data (practical identifiability), can we expect to obtain unique values for the model parameters?" It is important to notice the clear distinction between structural and practical identifiability in this statement. In the study of structural identifiability perfect noise-free data are assumed whereas in practice the data may be noise corrupted. As a result, parameters may be practically unidentifiable although they are structurally identifiable [3,4].

For linear systems the structural identifiability is well understood and there are several methods available for testing the identifiability (see e.g. [5], and for an overview of different methods [6]). On the contrary, the structural identifiability of nonlinear models (e.g. Monod models) is more complex to assess, and only a few methods are currently available [7]. In the case of nonlinear models the approach has typically been to work out different necessary and/or sufficient conditions that may allow to draw conclusions on structural identifiability [8]. The following list summarises available methods for nonlinear models, including both structural and application oriented references that have focused on Monod kinetic models.

1. Transformation of the nonlinear model into a linear model: [6,8,9]. Applications: [2,10,11].
2. Series expansions:
   2.1. Taylor series expansion: [6,8,12–14]. Applications: [2–4,10,11,15].
   2.2. Generating series: [8,14,16]. Applications: [17].
3. Similarity transformation approach or local state isomorphism: [13,14,18,19]. Applications: [20,21].
4. Study of the observability properties of the nonlinear system: [22]. Applications: [10].

In general, for both linear and nonlinear models, it can be very difficult to predict which approach involves the least efforts for a particular example [13]. In the study of Petersen [17], the series expansion methods were chosen since these are relatively simple to apply. In the Taylor series approach the series is generated with respect to the time domain whereas the series is generated with respect to the input domain in the generating series approach. In fact the generating series approach can be considered as an extension of the Taylor series expansion method for the case where a class of inputs is considered [23]. Thus, it appears that in the specific case of a model with zero inputs, the generating series approach becomes equivalent to the Taylor series expansion [8,18], as also illustrated in [17].

As stressed by Dochain et al. [2], the structural identifiability will only depend on the model structure and on which variables are measured. In the study of heterotrophic substrate degradation via the Monod model carried out by Holmberg [3], measurements of both substrate and biomass were assumed to be available, and it was proven that all model parameters were structurally identifiable under such conditions. However, in a similar study assuming only biomass measurements it was not possible to identify all parameters structurally [19]. In both studies [3,19], it was assumed that significant growth took place (i.e. the biomass concentration was changing as a function of time). In the work of Dochain et al. [2] growth was neglected (the biomass concentration was assumed constant) in the heterotrophic substrate degradation model, and oxygen uptake rate data were considered as measurements. It appeared that in this situation only certain parameter combinations were structurally identifiable. If, however, growth is assumed to take place the structural parameter identifiability improves because the identifiable parameter combinations obtained assuming no growth [2] can be split up further [11]. In the work of Bourrel et al. [10], the structural identifiability of the Monod kinetics was studied for the denitrification process in a biofilm model assuming steady state with respect to growth, and it was shown that depending on the measured state variables (nitrate, nitrite, carbon substrate) different parameter combinations were identifiable. In another study the identifiability of a reduced order model, to be used to control nitrification and denitrification by applying measurements of dissolved oxygen and nitrate, was investigated [20,21]. Also in this study it appeared that a number of parameters were identifiable uniquely whereas others were only identifiable in combination with other parameters.

The objective of this study is to analyse the structural parameter identifiability of Monod-based activated sludge models. As an illustrative example a Monod model of the nitrification process is used. It is assumed that respirometric measurements, i.e. data of dissolved oxygen concentration (S_O) or oxygen uptake rate (r_O) and titrimetric measurements (cumulative proton production, H_p) are available. However, the technical methods by which such data are obtained are not considered. Two situations will be investigated: first a model structure that excludes biomass growth, which simplifies the study significantly, and secondly a model structure where biomass growth is included in the model. These two model structures allow the description of short- and long-term experiments respectively. Initially, the structural identifiability will be investigated by the Taylor series expansion method. Afterwards, it will be shown how it is possible to generalise the results of the identifiability study for the nitrification model based on an ASM1-like stoichiometric matrix notation. It will be proven with examples that the identifiable parameter combinations can be predicted by applying simple rules, based on only knowledge of the process under study, the measured variables and the substrate considered, thereby avoiding the more complicated mathematical approaches.

2. Theory

The Taylor series expansion approach to study the structural identifiability was originally developed by Pohjanpalo [12]. The basis of the Taylor series approach is that the output (measured variables) vector, $\underline{y}$, and its derivatives with respect to time, typically developed around initial time 0, can be assumed to be known and unique. The successive Taylor series derivatives, $\underline{a}_k(\underline{p})$, are defined as described in Eq. (1) with t^* as the arbitrary time instant where the derivatives are taken, $\underline{p}$ as the (unknown) parameter vector. The Taylor series approximation of the output $\underline{y}$ at time t^* is given in Eq. (2).

$$\underline{a}_k(\underline{p}) = \lim_{t \to t^*} \frac{\mathrm{d}^k}{\mathrm{d}t^k}(\underline{y}(t,\underline{p})) \quad k = 0, 1, \ldots \tag{1}$$

$$\underline{y}(t^* + \Delta t, \underline{p}) = \underline{y}(t^*, \underline{p}) + \Delta t \frac{\mathrm{d}}{\mathrm{d}t}(\underline{y}(t^*, \underline{p})) + \frac{\Delta t^2}{2!}\frac{\mathrm{d}^2}{\mathrm{d}t^2}(\underline{y}(t^*, \underline{p})) + \cdots + \frac{\Delta t^k}{k!} \times \frac{\mathrm{d}^k}{\mathrm{d}t^k}(\underline{y}(t^*, \underline{p})) \quad k = 0, 1 \ldots \tag{2}$$

Thus, the Taylor series derivatives, $\underline{a}_k(\underline{p})$, are functions of $\underline{p}$. A set of algebraic equations arise by equating the coefficients of the Taylor series expansion, and the method simply consists of solving this set of equations with respect to the parameters or combinations thereof. A sufficient condition for the model to be structurally identifiable is that there exists a unique solution for $\underline{p}$ from this set of (nonlinear) algebraic equations [8,12].

3. Practical application of the series expansion methods

The practical procedure for application of the series expansion methods can be described by the following steps:

1. Computation of the successive derivatives of the output (measured variable) function with respect to time.
2. Choice of the parameter set $\underline{p}$ to be identified.
3. Evaluation of the successive derivatives by inserting already known quantities and derivatives of lower orders.

4. Express the successive derivatives as function of the parameter combinations that were chosen in step 2.
5. Solve the set of algebraic equations resulting from step 4 with respect to $\underline{p}$.
6. If a unique solution can be found from step 5 the parameter set selected in step 2 is structurally identifiable.

Typically, the series are developed around $t = 0$. However, systems that are singular (i.e. not solvable) around zero cannot be termed unidentifiable by the argument that a unique solution for $\underline{p}$ is lacking around $t = 0$, since later observations at $t > 0$ may provide additional information on $\underline{p}$ [8,12].

It should be stressed that it is the user that defines the parameters or parameter combinations $\underline{p}$ that are to be identified in step 2. Thus, the procedure becomes iterative, since so far there are no general rules for selecting the "right" combinations [2], although some developments occur to semi-automatically extract identifiable parameter combinations from the Taylor series expansion [24].

In the work of Margaria et al. [7] it was proven for rational function models (as also studied here) that an upper bound on the number of algebraic equations (step 5) required for the identifiability analysis does exist. However, in general it cannot be stated that this upper bound exists or is known. One therefore usually starts with a limited number of derivatives and adds more if necessary. There is no guarantee that new information could not have been obtained by including derivatives from an even higher order [6,13,18]. This lack of an upper bound means that this condition is often only sufficient, but not necessary, for identifiability [16,18]. The structure of the resulting (nonlinear) equations is most often far from simple, even for models of moderate complexity. Although symbolic manipulation software packages have proven very useful, this problem cannot always be resolved making it difficult to establish the identifiability properties [23].

For a model with zero input the generating series approach is equivalent to the Taylor series expansion method, as mentioned above. However, for models that include inputs, the generating series approach usually results in simpler equation structures than those of the Taylor series approach [14,23], although this was questioned by Godfrey and DiStefano [6]. In general, it can be difficult beforehand to judge which approach is the most suitable one for a particular model [13].

4. Model

Nitrification takes place in two steps (1) oxidation of ammonium (NH_4^+) to nitrite (NO_2^-) and (2) oxidation of nitrite (NO_2^-) to nitrate (NO_3^-). This process is illustrated in a simple form in Eq. (3). The amount of NH_4^+ incorporated into the biomass during growth is considered to be negligible in Eq. (3).

$$\begin{aligned} NH_4^+ + 1\tfrac{1}{2}O_2 &\rightleftharpoons NO_2^- + 2H^+ + H_2O, \\ NO_2^- + \tfrac{1}{2}O_2 &\rightleftharpoons NO_3^-. \end{aligned} \tag{3}$$

Note that both nitrification steps can be characterised by measurements of oxygen uptake whereas it is only during the first step that protons are produced. This makes it possible to characterise the first step by its proton production. In this study it is assumed that the rate of the second nitrification step is faster than the first step, similar to the ASM1 model [1], allowing the simplified approach illustrated in Eq. (4):

$$NH_4^+ + 2O_2 \rightleftharpoons NO_3^- + 2H^+ + H_2O. \tag{4}$$

Measurements of oxygen uptake (either by measurements of the oxygen uptake rate, r_O, or the oxygen concentration itself, S_O) can be carried out via respirometry [25]. The proton production (H_p) can be quantified via a titrimetric technique where the cumulative proton production is measured [26]. The considered model structure is based on ASM1 [1] with some modifications, and is summarised in Table 1. The ASM1 format is a compact way of presenting a model that

Table 1
Model used for interpretation of the respirometric and titrimetric data

Component (measurement, i) (substrate, k)→ Process (j)↓	1. X (mg COD/l)[a]	2. S_S (mg COD/l)[a]	3. S_O (mg O_2/l)[a]	4. S_{NH} (mg NH_4^+–N/l)[a]	5. S_{NO_3} (mg NO_3^-–N/l)[a]	6. H_p (meq/l)[a]	Process rate
1. Heterotrophic growth	1	$-\frac{1}{Y_H}$	$-\frac{1-Y_H}{Y_H}$	$-i_{XB}$		$\frac{i_{xB}}{14}$	$\mu_{maxH} X \frac{S_S}{K_S + S_S}$
2. Nitrification	1		$-\frac{4.57-Y_A}{Y_A}$	$-\frac{1}{Y_A} - i_{XB}$	$\frac{1}{Y_A}$	$\frac{i_{XB}}{14} + \frac{1}{7Y_A}$	$\mu_{maxA} X \frac{S_{NH}}{K_{SA} + S_{NH}}$

[a] In column headings: first entry, component (measurement, i), second entry (substrate, k) and last entry gives unit.

can readily be transformed into a state-space model presentation, i.e. a set of ODEs [1]. The model in Table 1 only considers substrate degradation related phenomena. This means that it is assumed that the effect of biomass decay related processes, aeration, etc. can be excluded from the available data. Modifications to ASM1 include:

- The active biomass was lumped into one fraction X, instead of subdividing it into a separate fraction of nitrifiers ($X_{BA} = f_{BA}X$) and heterotrophs ($X_{BH} = f_{BH}X$) as proposed by Henze et al. [1].
- In this study, a detailed model for the titrimetric data is only applied for the nitrification process (see Table 1), and not for the heterotrophic substrate degradation process. In the model protons (H_p) replace the ASM1 alkalinity (S_{ALK}) component, which means that the signs of the stoichiometric factors in the H_p column are the opposite of the signs that appear in the S_{ALK} column in the ASM1 matrix. In the model S_{NH} oxidation and uptake of S_{NH} for biomass growth during nitrification will produce H_p [26].
- For heterotrophic growth, the standard ASM1 conversion term for S_{ALK} is included in the H_p column of Table 1 (uptake of S_{NH} to be incorporated in new biomass produces H_p).

Note that the model in Table 1 assumes that biomass growth takes place. A simplified model, assuming that no biomass growth takes place, is often used as an approximation for interpretation of short-term biodegradation experiments [2,26]. When no net growth is considered, X is the equilibrium or steady state value for $X(t)$ and is therefore a constant. This model can readily be obtained by omitting the X column from Table 1, as well as the parameter i_{XB} (nitrogen incorporated into new biomass during growth). It should be noted that strictly speaking the mass balances are not correct in this approach, since it is only assumed that the substrate degradation processes induce a time-varying oxygen consumption and proton production or consumption. The assumptions are, however, reasonable, and no significant errors will be induced in the case of short-term experiments where significant growth can be assumed not to take place.

Fig. 1 illustrates a model example of a short-term biodegradation experiment where substrate is added at $t = 0$ to a batch reactor and exogenous oxygen uptake rate data ($r_{O,ex}$) and H_p data related to the substrate degradation are collected.

5. Results

All symbolic manipulations were carried out with the MAPLE V software package (Waterloo Maple Software). The results of the different identifiability studies are summarised in Table 2. Below only the results for combined respirometric and titrimetric measurements are dealt with in detail. For the detailed mathematical study of the other examples the reader is referred to [17].

5.1. Structural identifiability assuming single data sets

The results of the structural identifiability study assuming the availability of single data sets derived from either respirometric or titrimetric measurements are given in columns 2 and 3 in Table 2 (see also [17]). From a practical point of view, it is an advantage to

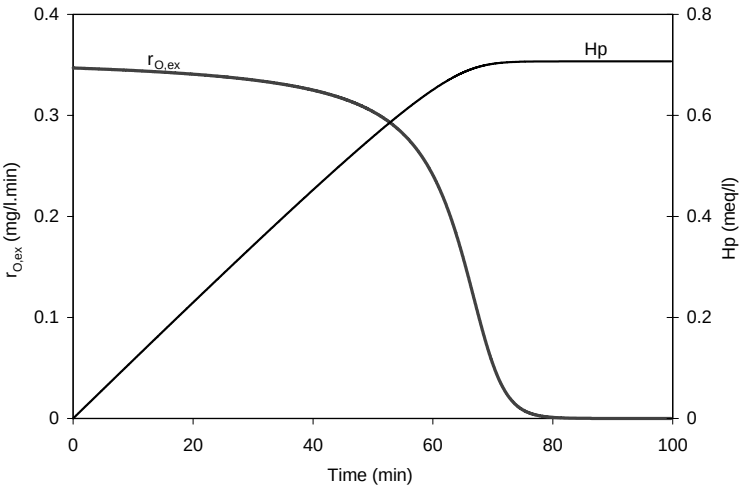

Fig. 1. Model example of a short-term biodegradation experiment with substrate added at $t = 0$; $r_{O,ex}$ and H_p data related to the substrate degradation process are shown.

Table 2
Schematic overview of the structurally identifiable parameter combinations for the nitrification process, depending on the available measurement(s) and the model structure

Process (j)	Nitrification		
Measurement (i) → Model structure ↓	S_O or r_O	H_p	$S_O + H_p$ or $r_O + H_p$
No growth	$\frac{4.57 - Y_A}{Y_A}\mu_{maxA} X$	$\frac{2}{14 Y_A}\mu_{maxA} X$	$\frac{4.57 - Y_A}{Y_A}\mu_{maxA} X$
	$(4.57 - Y_A)K_{SA}$	$\frac{2}{14}K_{SA}$	$(4.57 - Y_A)K_{SA}$
	$(4.57 - Y_{A1})S_{NH}(0)$	$\frac{2}{14}S_{NH}(0)$	$(4.57 - Y_A)S_{NH}(0)$
			$\frac{14}{2}(4.57 - Y_A)$
Growth	μ_{maxA}	μ_{maxA}	μ_{maxA}
	$\frac{4.57 - Y_A}{Y_A}X(0)$	$\frac{2 + i_{XB} Y_A}{14 Y_A} X(0)$	$\frac{4.57 - Y_A}{Y_A}X(0)$
	$\frac{4.57 - Y_A}{1 + i_{XB} Y_A}K_{SA}$	$\frac{2 + i_{XB} Y_A}{14(1 + i_{XB} Y_A)}K_{SA}$	$\frac{4.57 - Y_A}{1 + i_{XB} Y_A}K_{SA}$
	$\frac{4.57 - Y_A}{1 + i_{XB} Y_A}S_{NH}(0)$	$\frac{2 + i_{XB} Y_A}{14(1 + i_{XB} Y_A)}S_{NH}(0)$	$\frac{4.57 - Y_A}{1 + i_{XB} Y_A}S_{NH}(0)$
			$\frac{2 + i_{XB} Y_A}{14(4.57 - Y_A)}$

consider measurements of oxygen concentrations, S_O, which is the direct output of the dissolved oxygen electrode, for parameter estimation as an alternative to oxygen uptake rate measurements, r_O. The reason for this is that the differentiation step, which is needed to convert S_O data into r_O data, but at the same time also increases the noise on the data, can be avoided. The structural parameter identifiability will however not depend on whether one focuses on S_O or its derivative, r_O, as output, because it is a model property that holds independently of the noise level. The first row in Table 2 gives the results when no net biomass growth is assumed, whereas the second row shows the results when biomass growth is considered to take place in the study. As it can be seen from Table 1 the structure of the model describing respirometric and titrimetric measurements is rather similar, which also leads to similar identifiable parameter combinations with the stoichiometric factor as the only difference. For example, when no growth is assumed the identifiable parameter combination is $(4.57 - Y_A/Y_A)\mu_{maxA} X$ whereas the corresponding one for titrimetric data is $(2/14 Y_A)\mu_{maxA} X$.

5.2. Structural identifiability assuming two data sets

Now it will be assumed that respirometric and titrimetric data are obtained simultaneously in the same experiment. The measurements are assumed to be independent. Consequently, the information on identifiable parameter combinations based on r_O/S_O and Hp data separately, can be combined in the search for possibly new and improved parameter identification.

First it is assumed that no biomass growth takes place. Thus, the biomass concentration X is considered as a constant (column 1 in Table 1 is excluded), and i_{XB} is 0. These assumptions simplify the study significantly. For the nitrification process assuming no net biomass

growth (process 2 in Table 1) the expression for the oxygen uptake rate related to ammonium oxidation, i.e. the exogenous uptake rate $r_{O,ex}$, is given in Eq. (5). The proton production rate, r_{Hp}, due to ammonium oxidation, is given in Eq. (6).

$$r_{O,ex}(t) = \frac{4.57 - Y_A}{Y_A}\mu_{maxA}\, X \frac{S_{NH}(t)}{K_{SA} + S_{NH}(t)} \quad (5)$$

$$r_{H_p}(t) = \frac{2}{14 Y_A}\mu_{maxA}\, X \frac{S_{NH}(t)}{K_{SA} + S_{NH}(t)} \quad (6)$$

5.2.1. Step 1

The Taylor series expansion was applied according to the procedure lined out in the theory section and Eqs. (7)–(12) list the first three successive derivatives of $r_{O,ex}$ and r_{Hp} with respect to time at $t = 0$.

$$r_{O,ex}(0) = \frac{(4.57 - Y_A)}{Y_A}\mu_{maxA}\, X \frac{S_{NH}(0)}{K_{SA} + S_{NH}(0)} \quad (7)$$

$$\frac{dr_{O,ex}(0)}{dt} = -(4.57 - Y_A) \times \left(\frac{\mu_{maxA}\, X}{Y_A}\right)^2 \frac{K_{SA} S_{NH}(0)}{(K_{SA} + S_{NH}(0))^3} \quad (8)$$

$$\frac{d^2 r_{O,ex}(0)}{dt^2} = (4.57 - Y_A)\left(\frac{\mu_{maxA}\, X}{Y_A}\right)^3 \times \frac{K_{SA} S_{NH}(0)(K_{SA} - 2S_{NH}(0))}{(K_{SA} + S_{NH}(0))^5} \quad (9)$$

$$r_{H_p}(0) = \frac{2}{14 Y_A}\mu_{maxA}\, X \frac{S_{NH}(0)}{K_{SA} + S_{NH}(0)} \quad (10)$$

$$\frac{dr_{H_p}(0)}{dt} = -\frac{2}{14}\left(\frac{\mu_{maxA}\, X}{Y_A}\right)^2 \frac{K_{SA} S_{NH}(0)}{(K_{SA} + S_{NH}(0))^3} \quad (11)$$

$$\frac{d^2 r_{H_p}(0)}{dt^2} = -\frac{2}{14}\left(\frac{\mu_{maxA}\, X}{Y_A}\right)^3 \times \frac{K_{SA} S_{NH}(0)(2S_{NH}(0) - K_{SA})}{(K_{SA} + S_{NH}(0))^5} \quad (12)$$

5.2.2. Step 2

The parameter combinations that are considered identifiable in the study are given in Eqs. (13)–(16). The first three combinations are identical to the ones derived for S_O/r_O measurements (see Table 2, column 2). An additional parameter combination α_1 (Eq. (16)) is proposed and, if identifiable, would allow an identification of the autotrophic biomass yield Y_A. Inserting Y_A in Eqs. (13)–(15) would subsequently result in unique identification of $\mu_{maxA}\, X$, K_{SA} and $S_{NH}(0)$.

$$\beta_1 = \frac{4.57 - Y_A}{Y_A}\mu_{maxA}\, X \quad (13)$$

$$\beta_2 = (4.57 - Y_A) K_{SA} \quad (14)$$

$$\beta_3 = (4.57 - Y_A) S_{NH}(0) \quad (15)$$

$$\alpha_1 = \frac{14}{2}(4.57 - Y_A) \quad (16)$$

5.2.3. Steps 3 and 4

It is now sought to express the derivatives (Eqs. (7)–(12)) as a function of the parameter combinations (Eqs. (13)–(16)), as generally expressed in Eq. (17), where i is equal to the order of the derivative. The expressions for the actual case are given in Eqs. (18)–(21). In these equations v_0, v_1 and v_2 are the three first derivatives based on r_O measurements, whereas z_0 ($= r_{Hp}(0)$) is obtained by substituting the parameter combinations in the first derivative of H_p (Eq. (10)).

$$v_i = \frac{d^i r_{O,ex}}{dt^i}(0) \quad (17)$$

$$v_0 = \frac{\beta_1 \beta_3}{\beta_2 + \beta_3} \quad (18)$$

$$v_1 = -\frac{\beta_1^2 \beta_2 \beta_3}{(\beta_2 + \beta_3)^3} \quad (19)$$

$$v_2 = -\frac{\beta_1^3 \beta_2 \beta_3 (2\beta_3 - \beta_2)}{(\beta_2 + \beta_3)^5} \quad (20)$$

$$z_0 = \frac{\beta_1 \beta_2}{\alpha_1 (\beta_2 + \beta_3)} \quad (21)$$

5.2.4. Step 5

It was found that the equation set v_0, v_1, v_2 and z_0 (Eqs. (18)–(21)) could be solved with respect to the parameters β_1, β_2, β_3 and α_1, proving that the parameter combinations listed in Eqs. (13)–(16) are structurally identifiable. The solutions that were found for β_1, β_2, β_3 and α_1 are given in Eqs. (22)–(25).

$$\beta_1 = \frac{v_0(3v_1^2 - v_0 v_2)}{v_1^2 - v_0 v_2} \quad (22)$$

$$\beta_2 = \frac{-2v_0^2 v_1}{3v_1^2 - v_0 v_2} \quad (23)$$

$$\beta_3 = \frac{-4v_0^2 v_1^3}{(v_0 v_2 - 3v_1^2)(v_0 v_2 - v_1^2)} \quad (24)$$

$$\alpha_1 = \frac{v_0}{z_0} \quad (25)$$

It is noteworthy that it was not possible to identify the parameters based on a combination of the first two successive derivatives with respect to r_O (Eqs. (7) and (8)) and H_p (Eqs. (10) and (11)) data, respectively, indicating that information from higher

order derivatives (i.e. Eq. (9) or Eq. (12)) was needed. The analysis was also checked using the first three derivatives of the H_p measurements and the first derivative of r_O. This equation set could also be solved for the parameter combinations given in Eqs. (13)–(16).

Important to notice is that α_1 (Eq. (16)), the parameter combination that will allow to identify Y_A, is in fact nothing else but the ratio between the stoichiometric factors relating $r_{O,ex}$ and r_{Hp} to the ammonium degradation rate (see Tables 1 and 2). The consequence is that from Eqs. (14) and (15) K_{SA} and $S_{NH}(0)$ become identifiable separately. Only $\mu_{maxA} \cdot X$ remains as a parameter combination with combined measurements.

For the identifiability study explicitly taking net biomass growth into account the study followed the same pattern as described above. However, the difference here was that X was a function of time with a known initial biomass concentration. This complicated the expressions of the successive derivatives significantly and below only the first two derivatives of $r_{O,ex}$ are illustrated (Eqs. (26) and (27)) since the third derivative already got too complicated to show here.

$$r_{O,ex}(0) = \frac{(4.57 - Y_A)}{Y_A} \mu_{maxA}\, X(0) \frac{S_{NH}(0)}{K_{SA} + S_{NH}(0)} \tag{26}$$

$$\frac{dr_{O,ex}(0)}{dt} = -\frac{\mu_{maxA}^2\, X(0) S_{NH}(0)(4.57 - Y_A)(X(0)K_{SA}(1 + i_{XB}\, Y_A) - S_{NH}(0)\, Y_A(S_{NH}(0) + K_{SA}))}{Y_A^2 (K_{SA} + S_{NH}(0))^3} \tag{27}$$

The results of the study including net-growth is given in Table 2, where it can be seen that it appeared possible to separate μ_{maxA} and $X(0)$ in the identifiable parameter combinations when growth is explicitly considered. Further, an extra term including the parameter i_{XB} appears in the parameter combinations for $S_{NH}(0)$ and K_{SA}. Again, the parameter combination $(2 + i_{XB}\, Y_A)/(14(4.57 - Y_A))$ that contains the information on Y_A is defined by the ratio between the stoichiometric factors relating $r_{O,ex}$ and r_{Hp} to the ammonium degradation rate (see Tables 1 and 2).

Summarising, by considering growth and combined respirometric-titrimetric measurements the parameters μ_{maxA}, $X(0)$, K_{SA}, $S_{NH}(0)$ and Y_A become structurally identifiable under the assumption that i_{XB} is known.

6. Generalisation of structurally identifiable parameter combinations

A summary of the structurally identifiable parameter combinations resulting from this study is listed in Table 2. The investigation of the structural identifiability via the series expansion methods is an iterative procedure, as mentioned in the introduction and further illustrated in the examples above (see further examples in [17]). Essentially the parameter combinations arise from generation of the Taylor series coefficients. Indeed, one may find inspiration in the Taylor series coefficients to propose parameter combinations that can then be proven to be identifiable (see procedure in previous sections). However, there can be many parameter combinations that may not all be easily interpretable. Moreover the mathematical manipulations are often very complicated and time-consuming.

In the evaluation of the results obtained in this study however, it appeared possible to generalise the parameter identifiability results listed in Table 2 based on the ASM1-like stoichiometric matrix (Table 1). Indeed, the identifiable parameter combinations can be predicted based on the knowledge of the process under study, the measured component and the substrate component that is degraded. The rules of this generalisation are illustrated in Table 3. With reference to Table 1, ν denotes the stoichiometric coefficient, j the process, i the measured component, while the substrate under study is denoted k. Considering that some components are consumed (e.g. S_O, S_S) whereas others are produced (e.g. X, H_p), the absolute values of the stoichiometric coefficients ν should be taken. In the case two components involved in the same process are measured, the parameter combinations for a single measurement listed in Table 3 still hold, but with the additional identifiable parameter combination $\nu_{i(1),j}/\nu_{i(2),j}$, where (1) and (2) indicate the two measured components, respectively. The generalisation of Table 3 was confirmed with the identifiable parameter combinations listed in Table 2, but also with structural identifiability studies found in the literature, as will be illustrated below.

6.1. Example 1

Nitrification is considered (see Table 1), S_{NH} is added, H_p is measured and biomass growth is assumed to take place. Thus, in this case we have, $i = 6$, $j = 2$ and $k = 4$ (see Table 1). According to Table 3 the identifiable parameter combinations are as follows:

1. $\mu_{max,j}$, i.e. the maximum specific growth rate related to process 2 which is μ_{maxA}.

Table 3
Parameter combinations for Monod degradation kinetics based on ASM1-like matrix notation

S_O/r_O or H_p measurements		S_O/r_O or H_p measurements	
Model structures			
No growth	Growth	No growth	Growth
$\lvert v_{i,j}\rvert\mu_{\max,j}X$	$\mu_{\max,j}$	$\lvert v_{i,j}\rvert\mu_{\max,j}X$	$\mu_{\max,j}$
$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}K_j$	$\lvert v_{i,j}\rvert X(0)$	$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}K_j$	$\lvert v_{i,j}\rvert X(0)$
$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}S_k(0)$	$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}K_j$	$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}S_k(0)$	$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}K_j$
	$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}S_k(0)$		$\frac{\lvert v_{i,j}\rvert}{\lvert v_{k,j}\rvert}S_k(0)$
		$\frac{\lvert v_{i(1),j}\rvert}{\lvert v_{i(2),j}\rvert}$	$\frac{\lvert v_{i(1),j}\rvert}{\lvert v_{i(2),j}\rvert}$

See text for a detailed explanation of the generalisation rules (in the subscripts of the stoichiometric factors below, i indicates the measured component, j indicates the process that is considered, and k indicates the substrate under study).

2.

$$|v_{i,j}|X(0) = |v_{6,2}|X(0) = \left(\frac{i_{XB}}{14} + \frac{1}{7Y_A}\right)X(0) = \frac{2 + i_{XB}Y_A}{14Y_A}X(0)$$

3.

$$\frac{|v_{i,j}|}{|v_{k,j}|}K_j = \frac{|v_{6,2}|}{|v_{4,2}|}K_2 = \frac{2 + i_{XB}Y_A}{14Y_A}\frac{Y_A}{1 + i_{XB}Y_A}K_{SA} = \frac{2 + i_{XB}Y_A}{14(1 + i_{XB}Y_A)}K_{SA}$$

4.

$$\frac{|v_{i,j}|}{|v_{k,j}|}S_k(0) = \frac{|v_{6,2}|}{|v_{4,2}|}S_4(0) = \frac{2 + i_{XB}Y_A}{14(1 + i_{XB}Y_A)}S_{NH}(0)$$

These parameter combinations are completely in accordance with the ones derived in this study for the case where H_p is measured and biomass growth is considered in the model (see Table 2).

6.2. Example 2

Nitrification is again considered (see Table 1), S_{NH} is added, H_p and S_O are measured and growth is not considered (i.e. $i_{XB} = 0$). Thus, $i(1) = 3$, $i(2) = 6$, $j = 2$ and $k = 4$. The parameter combinations become:

1. $|v_{i,j}|\mu_{\max,j}X$. Now both $i(1)$ and $i(2)$ can be considered to write up a series of structurally identifiable parameter combinations. In this example only $i(1) = 3$ will be written as $|v_{3,2}|\mu_{\max,2}X = ((4.57 - Y_A)/Y_A)\mu_{\max A}X$

2.

$$\frac{|v_{i,j}|}{|v_{k,j}|}K_j = \frac{|v_{3,2}|}{|v_{4,2}|}K_2 = \left(\frac{4.57 - Y_A}{Y_A}Y_A\right)K_{SA} = (4.57 - Y_A)K_{SA}$$

3.

$$\frac{|v_{i,j}|}{|v_{k,j}|}S_k(0) = \frac{|v_{3,2}|}{|v_{4,2}|}S_4(0) = \left(\frac{4.57 - Y_A}{Y_A}Y_A\right) \times S_{NH}(0) = (4.57 - Y_A)S_{NH}(0)$$

4.

$$\frac{|v_{i(1),j}|}{|v_{i(2),j}|} = \frac{|v_{3,2}|}{|v_{6,2}|} = \frac{4.57 - Y_A}{Y_A}\frac{14Y_A}{2} = \frac{14}{2}(4.57 - Y_A)$$

Again, these results are in accordance with the results of Table 2 obtained via Taylor series expansions. Identical results would have been obtained in case $i(2) = 6$ was chosen for the derivation.

6.3. Example 3

If we now look beyond the identifiability studies carried out in this study, then an obvious example to check is the situation where both $r_{O,ex}$ and S_S are measured. The equation

$$\int_0^t r_{O,ex}\,dt = \frac{S_S(0)}{(1 - Y_H)}$$

results from the oxygen balance in the batch reactor, and from this one would expect that the biomass yield Y_H becomes identifiable if both $r_{O,ex}$ and S_S are measured. The integral of $r_{O,ex}$ indicates how much oxygen is consumed and can be regarded as an oxygen measurement. Furthermore, heterotrophic growth is considered and S_S is the substrate, i.e. $i(1) = 2$, $i(2) = 3$, $j = 1$, $k = 2$. The yield should now appear from the following combination:

1.

$$\frac{|v_{i(1),j}|}{|v_{i(2),j}|} = \frac{|v_{2,1}|}{|v_{3,1}|} = \frac{1}{Y_H}\frac{Y_H}{1 - Y_H} = \frac{1}{1 - Y_H}$$

This is proving, as expected, that the yield becomes identifiable if both S_O and S_S are measured.

6.4. Example 4

If we return to the literature, Holmberg [3] proved that all parameters μ_{maxH}, K_S, $S_S(0)$, $X(0)$ and Y_H were identifiable in the case where S_S and X measurements were available and biomass growth was considered. Hence, heterotrophic growth is considered (Table 1), S_S is substrate and S_S and X are measured. In this example $i(1) = 1$, $i(2) = 2$, $j = 1$ and $k = 2$ and, according to the generalisation, the identifiable parameter set should be:

1. $\mu_{max,j} = \mu_{maxH}$
2. $|v_{i,j}|X(0)$. As in example 2 both $i(1)$ and $i(2)$ can be considered, here $i(1) = 1$ will be chosen $= |v_{1,1}|X(0) = X(0)$
3. $$\frac{|v_{i,j}|}{|v_{k,j}|}K_j = \frac{|v_{1,1}|}{|v_{2,1}|}K_1 = \left(1\frac{1}{Y_H}\right)K_S = \frac{1}{Y_H}K_S$$
4. $$\frac{|v_{i,j}|}{|v_{k,j}|}S_k(0) = \frac{|v_{1,1}|}{|v_{2,1}|}S_2(0) = \left(1\frac{1}{Y_H}\right)S_S(0) = \frac{1}{Y_H}S_S(0)$$
5. $$\frac{|v_{i(1),j}|}{|v_{i(2),j}|} = \frac{|v_{1,1}|}{|v_{2,1}|} = 1\frac{Y_H}{1} = Y_H$$

Thus, since the biomass yield Y_H becomes identifiable from step 5 all the parameters μ_{maxH}, K_S, $S_S(0)$, $X(0)$ and Y_H become identifiable by applying the generalisation rules, similar to the results obtained by Holmberg [3].

6.5. Example 5

In the work of Spérandio and Paul [11], the structural identifiability was also studied for the heterotrophic growth process, but here assuming only measurements of $r_{O,ex}$. The identified parameters were μ_{maxH} $((1 - Y_H)/Y_H)X(0)$, $(1 - Y_H)K_S$ and $(1 - Y_H)S_S(0)$, which is in fact equivalent to the ones obtained in this study for growth during the nitrification process. The parameter combinations derived by Spérandio and Paul [11] could also be obtained directly based on the generalisation rules outlined above with $i = 3$, $j = 1$ and $k = 2$.

1. $\mu_{max,j} = \mu_{maxH}$.
2. $$|v_{i,j}|X(0) = |v_{3,1}|X(0) = \frac{1 - Y_H}{Y_H}X(0)$$
$$\frac{|v_{i,j}|}{|v_{k,j}|}K_j = \frac{|v_{3,1}|}{|v_{2,1}|}K_1 = \frac{1 - Y_H}{Y_H}Y_H K_S = (1 - Y_H)K_S$$
$$\frac{|v_{i,j}|}{|v_{k,j}|}S_k(0) = \frac{|v_{3,1}|}{|v_{2,1}|}S_1(0) = \frac{1 - Y_H}{Y_H}Y_H S_S(0) = (1 - Y_H)S_S(0)$$

7. Discussion

The structural identifiability of a nitrification model was studied assuming that respirometric and titrimetric measurements were available. Initially, the series expansion method was applied to assess the structural identifiability of the model parameters. The study was carried out for (i) a model structure that did not include net biomass growth and (ii) a model structure where biomass growth was explicitly taken into account. With respect to parameter identifiability, the difference between the two model structures was that the no-growth parameter combination including $X(0)$, Y and μ_{max} could be split up further into μ_{max} on the one hand, and a parameter combination including $X(0)$ and Y on the other hand when biomass growth was considered.

In the study of Brouwer et al. [27], the nitrification kinetic parameters for a two-step nitrification model were estimated. In that study the assumed structurally identifiable parameter combinations were defined to be the ones related to no growth although growth was considered in the model applied for parameter estimation. The same goes for the study of a one-step nitrification model by Vanrolleghem and Verstraete [28], and also for the parameter estimations presented by Spanjers and Vanrolleghem [29]. From a structural point of view, a wrong approach was taken in these studies by including growth in the model (e.g. i_{XB} appears in the model) whereas the assumed structurally identifiable parameter combinations were based on a no-growth model structure (i_{XB} did not appear in the identifiable combinations although it should have been). However, the experiments considered in these studies were all of short-term character where significant growth is unlikely to take place. Thus, the practical identifiability, based on a model incorporating growth, of the structurally identified parameter combinations resulting from a no-growth model structure would not have suffered much. The possible error in these studies is in fact related to the factor $(1 + i_{XB} \cdot Y_{A1})$.

Indeed, to be able to practically identify the structurally identifiable parameters assuming growth, the available data must show a significant increase of the amount of biomass, e.g. visible in a gradual increase of $r_{O,ex}$ during a long-term experiment. If the data do not reflect such significant biomass growth, the separation of the parameters μ_{max} and $X(0)$ will not work in practice,

causing high correlation between these two estimates. A good example for a respirometric experiment that does show significant biomass growth is presented in the work of Kappeler and Gujer [30] and is also applied elsewhere [11].

An important result of this study is that the autotrophic yield, Y_A, becomes uniquely identifiable by combining respirometric and titrimetric data. This is an important finding since the yield is an essential parameter in substrate degradation models. Indeed, the yield determines the distribution of consumed substrate between biomass growth and energy production. It is in fact not surprising that a unique identification of the biomass yield requires two kinds of measurements, since the yield coefficient relates two measures, that can link how much biomass is produced per unit of substrate degraded. Holmberg [3], who identified the yield coefficient uniquely by assuming combined measurements of biomass and substrate already illustrated this. In the present case with combined respirometric and titrimetric measurements, both measurements reflect how much biomass is produced per unit of substrate degraded. The biomass yield becomes identifiable for the combined measurements because an additional parameter combination becomes identifiable compared to a situation where only a single measurement is available. This additional parameter combination appears to be nothing else than the ratio of the two stoichiometric factors that relate the respective measured variables to substrate degradation.

Finally and most substantially, it was proven and illustrated that it is possible to generalise the structural parameter identifiability analysis based on an ASM1-like stoichiometric matrix. As stressed above one of the bottlenecks of the application of the series expansion methods is that the user initially has to propose the parameter combinations that may be identifiable from the Taylor series coefficients. Afterwards algebraic manipulations are required (each time) to prove that the selected parameter combinations are indeed structurally identifiable. If the problem is not solvable with the proposed combinations, other parameter combinations have to be proposed and evaluated, resulting in an iterative procedure. Thus, the proposed generalisation rules are a powerful tool to assess the structurally identifiable parameter combinations directly, only based on knowledge of the process under study, the measured component(s) and the substrate component(s) that is degraded. Thereby the rather time-consuming task of assessing the structural identifiability of parameters of models, described by the Monod growth kinetics in ASM1-like matrix presentations, has been reduced significantly.

In this paper the focus has been on activated sludge models. However it should be clear that the results are generally applicable for Monod-based kinetic models.

8. Conclusions

An essential first step in parameter estimation of models that are applied for data description is the assessment of the structural identifiability of the model parameters. In this study the structural identifiability of the nitrification process was studied via the series expansion methods, considering respirometric and titrimetric data. It appeared that the parameter identifiability improves when combined respirometric and titrimetric data are available, since the autotrophic yield becomes uniquely identifiable in this situation.

Secondly, an important outcome of this study was that the results of the structural identifiability study could be generalised. Based on simple generalisation rules the structurally identifiable parameter combinations can be assessed directly from an ASM1-like matrix representing the Monod model under study, thereby reducing the time needed for a structural identifiability study significantly. Thus, the structurally identifiable parameters can be obtained directly without considering the mathematical aspects of structural identifiability.

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