Two different strategies for baker's yeast fermentation process simulation

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Abstract – The simulation procedure represents an important tool to understand clearly the baker's yeast fermentation process. This work compares two different strategies to solve a mathematical model in order to predict the behaviour of the concentration of the state variables over a 20 h time period, in a well-mixed reactor. Mass balances written for all the components constitute a system of initial value problem. Considering the kinetics and the gas transfer rates relations, algebraic equations, as part of the differential system, a differential-algebraic system is defined. Two FORTRAN90-based simulators were employed for studies concerning the baker's yeast fed-batch fermentation. Both the simulation packages were seen to be an efficient tool for the simulation and tests of the non-linear process.

Keywords: baker's yeast fermentation, simulation, DAEs, ODEs.

1 Introduction

The building and use of mathematical models based on observed data is for long accepted as a basic scientific methodology. Models may be of a more or less formal character, but they have the basic feature that they attempt to link observations together into some pattern [1]. With the progress in digital technology, and thinking of bringing the theory into practice, computational modelling and model-based applications have emerged and are recognized as areas of great priority [2].

The conventional approach for process modelling is based on the balance equations for mass, energy, and, if necessary, momentum and population. This form of modelling requires further knowledge about reaction kinetics, thermodynamic, transport and physical properties.

Real processes in the chemical, biochemical and food industry are in their vast majority non-linear MIMO systems (Multiple Input Multiple Output). Their dynamics and control are difficult to study both for theoretical and practical reasons. In many instances experiments with real industrial processes are not carried out for reasons of economy and safety. Often on-line measurements are not available or simply they are too expensive. The simulation procedure represents an important tool to understand clearly the baker's yeast fermentation process.

The simulation consists on the integration of a set of non-linear differential equations, in order to the state variables, by considering or not as part of the system, algebraic equations, concerning mass transfer relations and kinetics laws.

The different methods of integration used are listed in the literature [3, 4]. In this article two different strategies are compared.
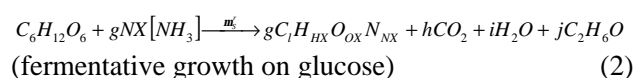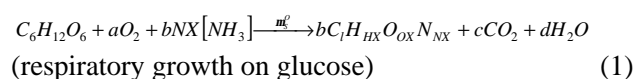
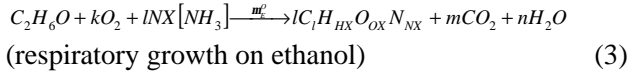## 2 Baker's yeast fermentation - Modelling approach

The simulation model consists of a set of algebraic equations, related to the kinetic models of baker's yeast growth and a set of five differential equations, in order to the state variables. The latter results from a mass balance of the fermenter. For more details, see [5].

Next sections describe the models implemented in the simulation programs.

2.1. Kinetic model

Yeast growth is characterized by three metabolic pathways

$$C_6H_{12}O_6 + aO_2 + bNX[NH_3] \xrightarrow{\ m_1^{O}\ } bC_lH_{HX}O_{OX}N_{NX} + cCO_2 + dH_2O$$

(respiratory growth on glucose)   (1)

$$C_6H_{12}O_6 + gNX[NH_3] \xrightarrow{\ m_s^{f}\ } gC_lH_{HX}O_{OX}N_{NX} + hCO_2 + iH_2O + jC_2H_6O$$

(fermentative growth on glucose)   (2)

$$C_2H_6O + kO_2 + lNX[NH_3] \xrightarrow{\ m_E^O\ } lC_lH_{HX}O_{OX}N_{NX} + mCO_2 + nH_2O$$

(respiratory growth on ethanol) $\qquad$ (3)

where S represents glucose; O oxygen; X biomass; E ethanol; C carbon dioxide and m_S^O, m_S^r, m_E^O: specific growth rates for the three pathways. In the sequel X, S, E, O, C mean concentrations.

The metabolic pathways of fermentative growth on glucose and oxidative growth on ethanol are competitive. This competition is governed by the respiratory capacity of the cells. If the instantaneous oxygen uptake capacity exceeds the oxygen need for total respiratory glucose uptake, then, all sugar uptakes follows the respiratory pathway (1) with the remaining oxygen being spent on ethanol respiratory uptake (3). Otherwise, if the instantaneous oxygen uptake capacity is not enough, then, part of glucose uptake follows the respiratory pathway (1) while the remaining follows the fermentative pathway (2).

The kinetics equations for baker's yeast growth, considered as Monod equations, are determined as follows.

The total specific growth rate, m_t, is the sum of the growth rates for the three pathways

$$m_t = m_S^O + m_S^r + m_E^O . \qquad (4)$$

The specific growth rates, m_i, can be related to the corresponding substrate fluxes, q, and yield coefficients (Table 1), Y, by

$$m_t = Y_{X/S}^O q_S^O + Y_{X/S}^r q_S^r + Y_{X/E}^{OE} q_E^O \qquad (5)$$

where $Y_{X/S}^O$ and $Y_{X/S}^r$ represent the yield coefficients of biomass in glucose in the oxidative and fermentative phases, respectively; $Y_{X/E}^{OE}$ is the yield coefficient of biomass in ethanol in the oxidative phase in ethanol.

As ethanol uptake is influenced by the priority of glucose uptake, which functions as an inhibitor, the specific growth rate on ethanol can be described as

$$m_E^O = m_E^{\max} \frac{E}{E + K_E} \frac{K_i}{S + K_i} \qquad (6)$$

where $m_E^{\max}$ is the maximal specific growth rate, K_i is the inhibition parameter and K_E is the saturation parameter.

However, this equation holds true only if there is an available respiratory capacity of the cells.

Table 1. Yield coefficient values (proposed by [6])

Coefficient	Value
$Y_{X/S}^O$	0.49 g biomass/g glucose
$Y_{X/S}^r$	0.05 g biomass/g glucose
$Y_{X/E}^r$	0.10 g biomass/g ethanol
$Y_{X/E}^{OE}$	0.72 g biomass/g ethanol
$Y_{X/O}^O$	1.20 g biomass/g oxygen
$Y_{X/C}^{OE}$	0.64 g biomass/g oxygen
$Y_{X/C}^O$	0.81 g biomass/g carbon dioxide
$Y_{X/C}^r$	0.11 g biomass/g carbon dioxide
$Y_{X/C}^{OE}$	1.11 biomass/g carbon dioxide

The glucose uptake, q_S, is slightly different because it follows two metabolic pathways: oxidative and fermentative

$$q_S = q_S^O + q_S^r . \qquad (7)$$

The glucose, q_S, and oxygen, q_O, uptake follow a Monod kinetics, respectively

$$q_S = q_S^{\max} \frac{S}{S + K_S} \qquad (8)$$

$$q_O = q_O^{\max} \frac{O}{O + K_O} \qquad (9)$$

where $q_S^{\max}$ is the maximal specific glucose uptake rate, K_S and K_O are saturation parameters and $q_O^{\max}$ is the maximal specific oxygen uptake rate.

By (1), it can be seen that the oxidative glucose uptake depends on the availability of dissolved oxygen, and may be defined as

$$q_S^O = \frac{q_O^O}{a} \qquad (10)$$

where a is the stoichiometric coefficient of the oxygen in the respiratory pathway of glucose and q_O^O is the oxygen uptake on glucose.

Two situations may occur: excess of oxygen that

implies no fermentative growth of biomass; lack of oxygen and consequently excess of glucose that implies no respiratory growth on ethanol.

Table 2 resumes mathematically what has been said; only two of the three metabolic pathways coexist.

Table 2. Baker's yeast kinetics

$aq_S \pounds q_O$ **Respirative Regime**	$\mathbf{m}_S^O = Y_{X/S}^O \cdot q_S$	(11)
	$\mathbf{m}_S^r = 0$	(12)
	$\mathbf{m}_E^O = min\left(\mathbf{m}_{E_1}^O, \mathbf{m}_{E_2}^O\right)$	(13)
$aq_S > q_O$ **Respiro-Fermentative Regime**	$\mathbf{m}_S^O = Y_{X/S}^O \cdot \dfrac{q_O}{a}$	(14)
	$\mathbf{m}_S^r = Y_{X/S}^r \cdot \left(q_S - \dfrac{q_O}{a}\right)$	(15)
	$\mathbf{m}_E^O = 0$	(16)

Two auxiliary equations, (17) and (18), must be added to the equations listed in Table 2, for the estimation of the specific growth rate on ethanol, defined as:

$$\mathbf{m}_{E_1}^O = \mathbf{m}_E^{max} \frac{E}{E + K_E} \frac{K_i}{S + K_i} \qquad (17)$$

and

$$\mathbf{m}_{E_2}^0 = \frac{Y_{X/O}^{OE}}{Y_{X/E}^{OE}} \left(q_O - aq_S\right). \qquad (18)$$

The relevant kinetic data were taken from Sonnleitner and Käppeli [7] (Table 3).

Table 3. Kinetic parameters

Parameter	Value
q_S^{max}	3.5 g gluc^{-1} g biom^{-1}h^{-1}
q_O^{max}	0.256 g O$_2$$^{-1}$ g biom^{-1}h^{-1}
$\mathbf{m}_E^{max}$	0.17 h^{-1}
K_E	0.1 gl^{-1}
K_i	0.1 gl^{-1}
K_S	0.2 gl^{-1}
K_O	0.1 mgl^{-1}

2.2. Mechanistic model

The mechanistic model for the fed-batch fermentation is obtained from mass balances for all components, considering that the reactor is well mixed.

Furthermore it is assumed that the yield coefficients (Y's) are constant and the dynamics of the gas phase can be neglected.

Then the set of differential model equations is:

·   mass balance for the biomass

$$\frac{dX}{dt} = \left(\mathbf{m}_S^O + \mathbf{m}_S^r + \mathbf{m}_E^O - D\right)X \qquad (19)$$

·   mass balance for the sugar

$$\frac{dS}{dt} = \left(-\frac{\mathbf{m}_S^O}{Y_{X/S}^O} - \frac{\mathbf{m}_S^r}{Y_{X/S}^r}\right)X + \left(S_f - S\right)D \qquad (20)$$

where S_f is the substrate concentration in the feed,

·   mass balance for the ethanol

$$\frac{dE}{dt} = \left(\frac{\mathbf{m}_S^r}{Y_{X/E}^r} - \frac{\mathbf{m}_E^O}{Y_{X/E}^{OE}}\right)X - DE \qquad (21)$$

·   mass balance for the oxygen

$$\frac{dO}{dt} = \left(-\frac{\mathbf{m}_S^O}{Y_{X/O}^O} - \frac{\mathbf{m}_E^O}{Y_{X/O}^{OE}}\right)X - DO + OTR \qquad (22)$$

·   mass balance for the carbon dioxide

$$\frac{dC}{dt} = \left(\frac{\mathbf{m}_S^O}{Y_{X/C}^O} + \frac{\mathbf{m}_S^r}{Y_{X/C}^r} + \frac{\mathbf{m}_E^O}{Y_{X/C}^{OE}}\right)X - DC - CTR \qquad (23)$$

The accumulation of the working volume during the fed-batch process is represented by

$$\frac{dV}{dt} = DV . \qquad (24)$$

The dilution rate (ratio feed rate/volume), D, is defined by

$$D = \frac{F}{V} . \qquad (25)$$

The gas transfer rates are given by -

$$OTR = K_L^O \, a\left(O^* - O\right) \qquad (26)$$

$$CTR = K_L^C \, a\left(C - C^*\right) \qquad (27)$$

where $K_L^i a$ are overall mass transfer coefficients for oxygen and carbon dioxide and $O*$ and $C*$ are the corresponding equilibrium concentrations.

3   Numerical methods for integration

The set of model equations that describes the mechanistic for the fed-batch fermentation, differential equations (19-24) plus the algebraic equations (11-16, 25-27), forms a system of differential-algebraic equations. Differential-algebraic (DAE) systems are different from ordinary differential (ODE) systems in that, while they include ODE systems as a special case, they also include problems that are quite different from ODEs, [3]. Some of these systems can cause severe difficulties for numerical methods. Consequently, the numerical solution of these systems is a very active area of research.

The index of a DAE system is a measure of its degree of singularity. Roughly speaking, ODE systems, $y' = f(t, y)$, have index zero. Differential equations coupled with algebraic equations, $y' = f(y, z)$, $0 = g(y, z)$, have index one if $g = 0$, can be solved for z given y (that is, if $\partial g / \partial z$ is non-singular) and otherwise have an index higher than one. The system in study is of index one so, the higher order index system would not be here discussed. The initial conditions for the DAE system in contrast to the ODE system, must be consistent, in the sense that they must satisfy the constraints of the system and possibly also some of the derivatives of the constraints. DASSL is a package freely available on the Internet via NetLib. DASSL uses a fixed-leading-coefficient BDF methods for index-one DAEs, and treats the linear systems as full or banded, but in various details it addresses the issues of DAE problems directly. More details on the algorithm are described in [3]. The error tolerances, ATOL and RTOL (absolute and relative error tolerances, respectively), were taken to be 10^{-4}. This value was obtained after several simulation runs where no significant differences were found in the final results.

When considering baker's yeast growth model as an ODE system, the algebraic equations are calculated prior to the set of differential equations. The method employed is a variable step algorithm, based on embedded Sarafayan method, involving the Butcher´s formulas of 4th and 5th order [4], SINTEG routine. The truncation error is forced to be less than a limit defined by the user [8].

All computations reported were performed in double precision on a Pentium 500 MHz computer.

4   Simulation procedure

The simulation programs of baker's yeast production were developed in FORTRAN90 workspace.

Figure 1 describes, in a schematic form, the MAIN program developed for the fed-batch fermentation. The procedures inside the block dashed line, depend on the strategy used: DAE/ODE systems.

The only difference in the ODE based integration method is that, as already referred, the algebraic equations are calculated before the definition of the five differential equations routine, FN. FUN routine defines the differential-algebraic system.
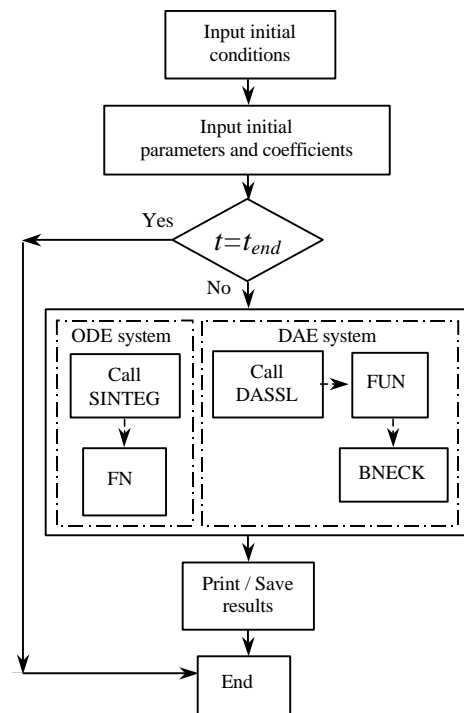

Figure 1. Flowchart of the MAIN program routine.

Figures 2 and 3 present, respectively, the flowcharts for the routine where the differential and algebraic system equations are defined, FUN, and the routine where the switch between the respirative and the respiro-fermentative models is verified, BNECK. DASSL integrator automatically calls these two routines, defined by the user.

The runs were taken at the same conditions, namely-Initial values:

$X(0)=0.3$ g/l, $S(0)=1.3$ g/l, $E(0)=0.8$ g/l
$O(0)=.0066$ g/l, $C(0)=0.002$ g/l, $V(0)=2$ g/l,

S_f=30 g/l

Final volume: 5 l

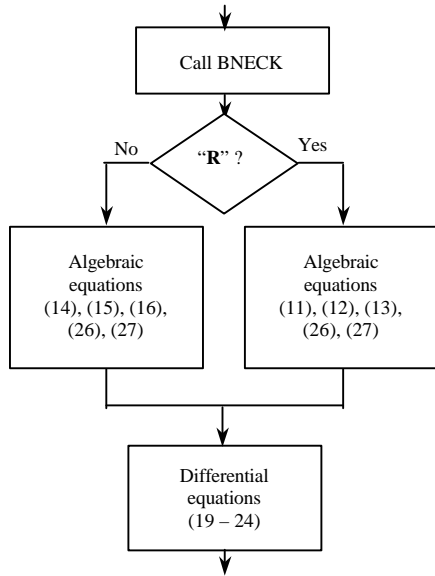Manipulated variable: Constant Glucose feed-rate, F=0.12 l/h.

Figure 2. Flowchart of the FUN routine where the differential algebraic system is defined. "**R**" refers to the respirative regime.
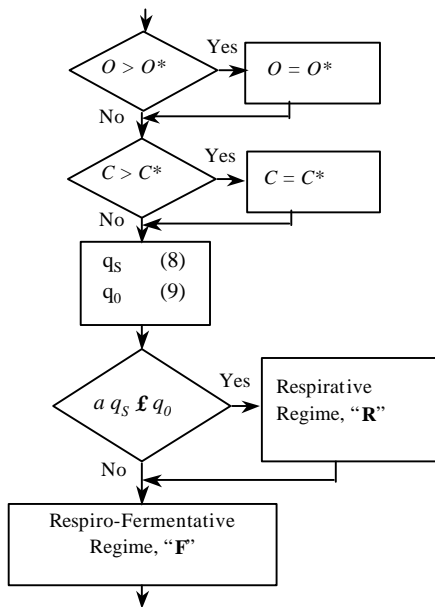

Figure 3. Flowchart of the BNECK routine

5 Results

The state variables, biomass, X, sugar, S, ethanol, E, oxygen, O, and carbon dioxide, C, concentration profiles, during a 20 hours run, do not differ significantly for both different strategies. Figures 4 and 5 show, respectively, the X, S and E, and O concentration profiles. These results regard to the DAE-based system. The carbon dioxide was kept constant during all runs.

Nevertheless the ODE-based method shows some small oscillations, particularly in biomass, X, and oxygen, O, profiles, possibly due to numerical problems. These oscillations are reflected on the relative error behaviour.
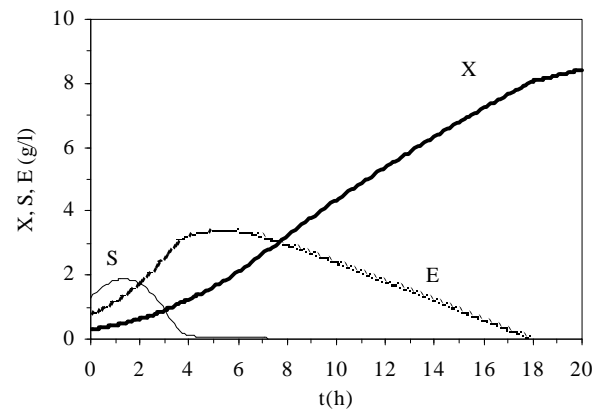

Figure 4. Biomass, X, sugar, S, and ethanol, E, concentration profiles on a 20h simulation run, under the defined initial conditions
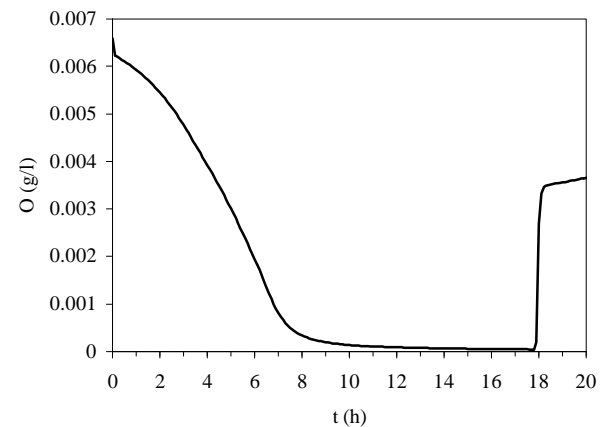

Figure 5. Oxygen, O, concentration profile on a 20h simulation run, under the defined initial conditions

Figures 6 and 7 present the relative errors (defined, in percentage, as the difference between the values

obtained in DAE-based method and the ODE-based procedure, divided by the first) for the four state variables.

Both oxygen, O, and sugar, S, have small concentration values. However, the oxygen has higher relative error, namely after 8 h run; small variations in small concentration values increase the relative errors. This is also the reason for the behaviour for the ethanol, E, relative error after 18 h run. This behaviour indicates that one of the integrator is more sensitive to describes the decay of the concentration during the time period.
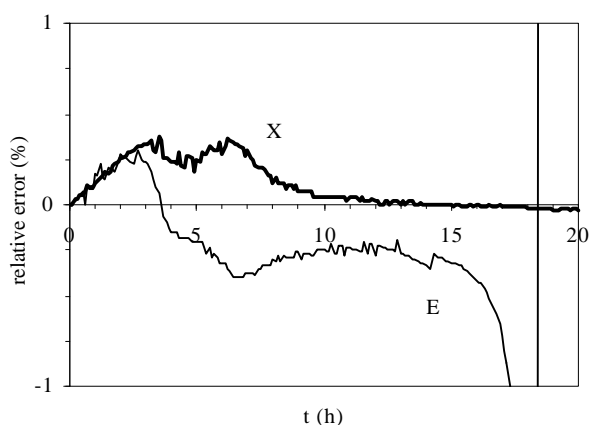

Figure 6. Relative error (%) between the two model strategies for the biomass and ethanol concentrations
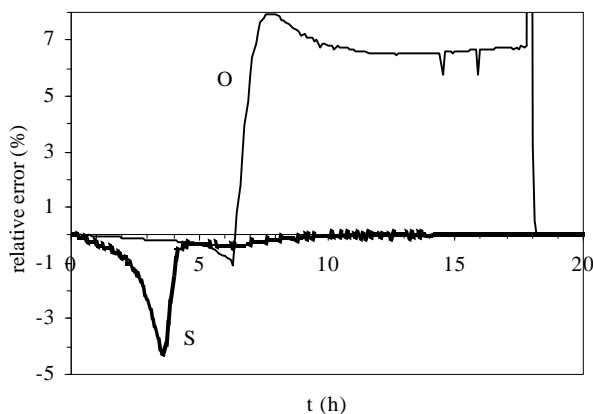

Figure 7. Relative error (%) between the two model strategies for the sugar and oxygen concentrations

6 Conclusions

Two simulators were developed and compared for studies of fed-batch baker's yeast fermentation.

The process was implemented in open-loop mode at

constant feed rate of glucose substrate (F=0.12 l/h).

For the same initial conditions, both strategies seem to characterize the case study.

In the DAE-based method, the initial state should be consistent and well defined.

The differences in the final results obtained by using the two methods were measured in terms of concentrations relative error (%). Some differences could be found, namely for some values of run time period where small concentrations of the state variables were obtained. The DAE based method seems to be more robust and less sensitive to numerical problems.

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