Modelling VFA production kinetics from protein-rich industrial wastes

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Abstract

Organic industrial waste streams rich in proteins are good candidates for producing added-value compounds, such as volatile fatty acids, in mixed-culture fermentations processes. However, the lack of design tools such as kinetic models hinders the implementation of this technology as the outcome of these processes is greatly affected by operational and design factors. In this work a kinetic model for protein fermentation with mixed cultures is developed and calibrated using expressly generated experimental data. As a proof of concept, the degradation of cheese whey and mussel canning wastewater were simulated. Ethanol and acetate were in both cases the major products and different degradation rates were observed for carbohydrates and proteins. This served to underline the utility of the model developed in the design of processes providing value to volatile fatty acids.

Keywords: protein open fermentation; kinetic modelling; VFA production.

Introduction

Protein-rich wastes such as dairy and canning industry wastes are interesting substrates to produce volatile fatty acids (VFA) in mixed-culture fermentations (MCF). To boost process productivity kinetic-based models are a powerful tool to be used in the design phase. Moreover, it is very important to predict VFA yields as it highly affects the design of processes converting VFA into high added-value products (e.g. chain elongation or bioplastic production). However, there are no fermentation-oriented models available yet. The Anaerobic Digestion Model No.1 (ADM1) is a well-established model for the anaerobic digestion (AD) process and can be used as a starting point as fermenting the substrate to VFA is one of the first steps of AD. But the different kinetic and stoichiometric parameters provided are not suitable for MCF because the objective of ADM1 is to simulate methane production. On the contrary, an MCF kinetic model should allow to simulate VFA production and to optimise the process towards the yielding of a specific VFA or mixture of them. Moreover, in the specific case of protein fermentation little data is available in literature that can be used to determine kinetic parameters. The objective of this work is to develop a kinetic model for VFA production in mixed-culture fermentations specifically built for protein degradation. Its main parameters will be calibrated with new experimental data using two model proteinaceous substrates: casein and gelatine. As a proof of concept, the degradation of two protein-rich industrial waste streams will be simulated.

Materials and Methods

The model is built following the structure described in Batstone (2002) with some minor modifications. Methanogenesis is deleted from the model as we consider that it is effectively inhibited in our system. The experimental data was generated in batch tests with two different substrates (casein and gelatine hydrolysates) at pH 7 and at pH 9 for casein

while only at pH 7 for gelatine. Initial concentrations ranged from 5 to 10 g COD/L. More details on the experimental setup can be found elsewhere (Bevilacqua et al. 2018).

The following kinetic parameters were estimated: maximum specific growth rate of amino-acid degraders (μ_{max}), yield of amino-acid-degrading biomass (Y_{AA}), decay constant (k_{dec}) and the different VFA yields ($F_{AA,VFA}$). Since substrate consumption was not full in all the experiments, we included the extent of substrate consumed as an additional parameter to estimate (S_{AA}). The calibration procedure is done following the non-linear least squares method (Eq. 1) in MATLAB (lsqnonlin command).

$$\hat{\theta} = \arg\min\left(\sum_{k}\left(\sum_{j}\left(\sum_{i}\left(\frac{y_{j,i}(\theta) - y_{j,i,exp}}{\sigma_{j,i}}\right)^2\right)\right)\right) \quad (1)$$

where $\hat{\theta}$ is the set of parameters to estimate, y is the simulated concentration, y_{exp} is the experimentally measured concentration and σ is the experimental standard deviation. The subscript i refers to the different compounds, the subscript j refers to the different batch experiments and the subscript k refers to the different measurements over time.

The industrial wastes selected for the simulations are cheese whey (CW) and mussel canning processing wastewater (MCWW). CW is assumed to be composed of 4 g/L of lactose and 1 g/L of casein and MCWW to contain 10.6 g/L of carbohydrates and 4.3 g/L of gelatine. VFA yields of sugar degradation are adapted from Temudo et al. (2007).

Results and Conclusions

Calibration results (Figure 1 and Table 1) show differences depending both on substrate and pH. For example, the maximum specific growth rate of gelatine degraders is 44% lower than that of casein degraders. The extent of substrate consumption was affected by the substrate nature, and thus gelatine was the only substrate to be fully degraded. On the contrary, the pH value, had a less marked effect on parameter variability.

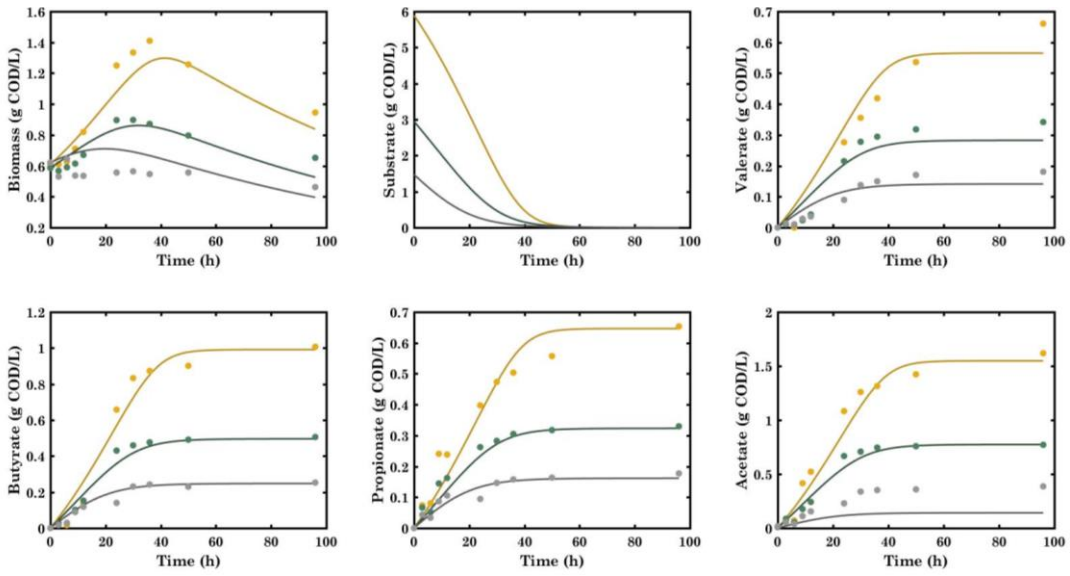

Figure 1. Calibration fit example (casein batch degradation at pH7).

The only significant differences are found in substrate consumption extent: 80% of casein was consumed at pH9 while at pH7 only an average of 57% was degraded (three

experiments at pH7 were performed). Comparing the parameters values obtained in our calibration using experimental data (Table 1) with literature values (Table 3) underlies the importance of using appropriate kinetic parameter values. Moreover, there is a significant disparity between the values found in literature that would be translated in completely different results if used in a kinetic model. Additional kinetic parameters at different conditions are required to build-up a parameter library that permits choosing the best parameters set depending on the protein to be degraded and the operational conditions.

Table 1. Estimated and literature kinetic parameter values for the pH values tested. Confidence intervals of the estimated parameters are determined following t distribution at 95% confidence level. 1: Batstone (2002), 2: Flotats et al. (2006), 3: Angelidaki et al. (1999).

	CASEIN			**GELATINE**	
	pH7	**pH9**	**Literature**	**pH7**	**Literature**
μ_{max} (d^{-1})	0.89 ± 0.23	0.87 ± 0.22	2.36-3.06[1]	0.50 ± 0.19	15.987 ± 54.27[2]
Y_{AA} (g COD BM/g COD AA)	0.17 ± 0.03	0.18 ± 0.03	0.085[1]	0.16 ± 0.04	0.086[3]
K_{dec} (d^{-1})	0.14 ± 0.04	0.15 ± 0.04	0.02[1]	0.00 ± 0.02	
S_{AA} (%)	56.8 ± 5.7	79.0 ± 5.9		100 ± 9.51	
$F_{AA,Val}$	0.16 ± 0.02	0.19 ± 0.02		0.07 ± 0.01	
$F_{AA,But}$	0.20 ± 0.02	0.16± 0.02		0.13 ± 0.02	
$F_{AA,Pro}$	0.13 ± 0.02	0.10 ± 0.01		0.15 ± 0.02	
$F_{AA,Ac}$	0.30 ± 0.03	0.35 ± 0.02		0.45 ± 0.04	

As a proof of concept, the degradation of two industrial wastes (CW and MCWW) in batch-mode at pH7 was simulated with the MCF model developed in this work. In both cases the VFA end concentrations show that acetate and ethanol are the main products due to the high carbohydrates initial concentration as they are the only products of its degradation at pH7 (Fig 2.). Different kinetic trends are evident in Fig. 2 with fast and slow conversion rates related to the degradation of carbohydrates and proteins respectively, e.g. ethanol reaches much sooner the end concentration value as sugars are degraded much faster than amino acids. It can be noted as well that ethanol concentration is higher than acetate concentration in CW degradation and that in MCWW degradation the proportion of uneven carboxylates is higher than in CW degradation. These facts will have implications in the further processes using VFA as substrates as, for example, chain elongation or polyhydroxyalkanoates (bioplastic) production.

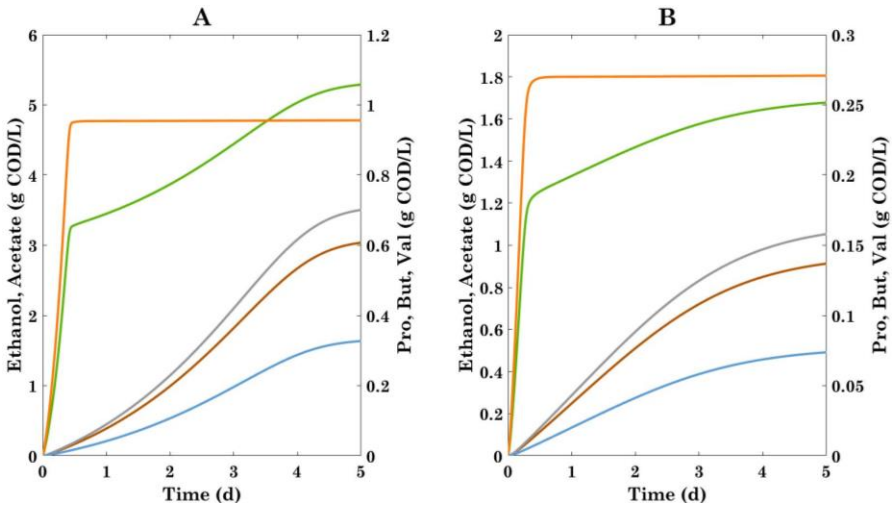

Figure 2. VFA and ethanol concentration profiles in the industrial streams degradation simulation. A: MCWW, B: CW. (—): ethanol, (—): acetate, (—): propionate, (—): butyrate, (—): valerate.

In this work we have developed a model that simulates the kinetics of volatile fatty acids production in the degradation of protein-rich wastes in mixed-culture fermentations. We can now use this model to design a process that targets a specific volatile fatty acid or group of them. This is a key issue as the outcome of the processes transforming them into high-added value products (e.g. bioplastic or biofuels) is heavily affected by the concentration of the different acids. The application scope of this model must be broadened with the inclusion of parameters from other proteins to increase the number of protein-rich industrial wastes that can be modelled.

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