



Carboxylic acids production using residual glycerol as a substrate in anaerobic fermentation: A kinetic modeling study

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ABSTRACT

This work aimed to analyze the carboxylic acids (CA) production using residual glycerol (RG) from biodiesel as a substrate in the anaerobic fermentation and perform a kinetic study of this bioprocess. RG appeared to be a readily biodegradable carbon source to the acidogenic microorganisms. A yield of 0.62 mg acids mg COD_A⁻¹ was achieved, corresponding to 0.82 mg COD_{CA} mgCOD_A⁻¹. Caproic acid, a medium-chain CA, was produced even without external supplementation of electron donors. There was variation between the best fit models to describe the CA kinetics production. Most of the acids produced were better explained by exponential models, emphasizing the Cone and Fitzhugh models. Therefore, RG is a potential agro-industrial waste for CA production from a resource recovery perspective.

1. Introduction

In the last decades, anaerobic digestion (AD) has traditionally been adopted for commercial biogas production from organic waste [1]. Despite this, carboxylic acids (CA) – such as acetic (HAc), propionic (HPr), and butyric (HBu) acids – which are produced as intermediates in AD, are products of increasing market demand (more than 15,000 kton per year). CA can be applied as a carbon source for further processing or nutrient removal in wastewater treatment plants [2,3]. They are also widely used as building block chemicals to synthesize chemicals and base-biochemicals, pharmaceuticals, cosmetics, materials, bioplastics, and biofuels [4,5].

Bio-based CA production from DA anaerobic fermentation stage using agro-industrial wastewaters as a substrate is a viable alternative to assist in replacing dependence on fossil raw materials in the biorefinery concept [6]. Among the residues that can be used, the residual glycerol (RG) from the biodiesel production process seems to be a promising substrate for the synthesis of bio-CAs [7].

By 2027, about 4,000,000 tons of glycerol will be produced as a residue from the transesterification of vegetable oils and animal fats for biodiesel production [8]. If discarded as waste, RG requires 1.47 g O₂ g of glycerol⁻¹ for its complete oxidation, resulting in an additional

operating cost for the biodiesel industry with its treatment. Therefore, RG use in biotechnological processes, such as CA production in the anaerobic fermentation, might be a more viable alternative since it is an abundant and cheap waste and has a higher reduction degree than sugars (e.g., sucrose and glucose), which allows bioproducts production with higher yields [9,10].

In the resource recovery context, mathematical modeling is an attractive strategy to generate data for reactor design and processes simulation, which can support technical-economic analyses in recovery units, coming to a more efficient process and less need for highly qualified operators [11,12]. The use of different types of models is already well-established to describe the kinetics of biogas production from AD [13–15]. However, the technical literature still lacks knowledge about anaerobic fermentation kinetics strategies that can predict the dynamic and stationary behavior of the process, especially in conditions not tested empirically [16,17].

As far as we know, research that studies the acidogenic process of agro-industrial wastes and its kinetics is scarce [18,19]. Therefore, this work's objective was to analyze the CA potential production using RG as a substrate in the anaerobic fermentation and perform a kinetic study of this bioprocess.

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2. Materials and methods

2.1. Substrate and inoculum

Residual glycerol (RG) was collected from a biodiesel production unit (Petrobrás) located in Quixadá, Ceará, Brazil. The RG used in the experiments was generated from the transesterification of soybean oil (56%) and bovine tallow (44%) [7].

RG was characterized in terms of its physical-chemical composition: chemical oxygen demand total (COD_T), particulate (COD_P) and soluble (COD_S) of 1972.7, 1200.9, 771.8, respectively, alkalinity of 425,1 mgCaCO₃ L⁻¹, pH of 6.40, total nitrogen (TN) of 5.0 mg-N L⁻¹, total phosphorus (TP) 94.5 mg-P L⁻¹, sulfate (SO₄²⁻) of 118.2 mg L⁻¹, chloride (Cl⁻) of 26.5 g L⁻¹ and oils and greases (OG) of 195.8 mg L⁻¹. The anaerobic biomass used as inoculum (33.8 gTVS L⁻¹) was obtained from an anaerobic sludge blanket (UASB) reactor treating brewery wastewater situated in the city of Fortaleza, Ceará, Brazil. Physical-chemical analyses were performed according to APHA [20].

2.2. Anaerobic batch assays

300 mL borosilicate flasks, with 250 mL working volume, were used as reactors for the anaerobic batch assays. The tests were performed in quadruplicate and followed Coelho et al. [18] and Morais et al. [19]. The reactors were inoculated with anaerobic sludge, and the food/microorganism (F/M) ratio adopted was 0.64 ± 0.03 gCOD gVSS⁻¹. The basal medium composition was described elsewhere [7], and the pH value was 7.0. 0.5% (v/v) chloroform insertion was the chemical strategy adopted to inhibit methanogenesis [8].

The bioreactors were sealed, and nitrogen gas (N₂) was used to purge the flasks for 1 min. During the experiment, the flasks remained at 35 ± 0.3 °C under orbital agitation at 150 rpm for 14 days [18].

1 mL samples were collected on days 0, 2, 4, 7, and 14 of the essays for COD analysis and CA quantification to monitor substrate consumption and acids production, respectively. On the 14th day, a 1 mL sample of biogas was analyzed in gas chromatography to confirm chloroform methanogenesis inhibition CA concentrations were determined by liquid chromatography. The applied analytical chromatographic methods were described in our previous works [18,19].

2.3. Calculations and kinetic modeling studies

Yields, productivity, and selectivity calculations were made from the equations shown in Table A1. The equations of the kinetic models used are shown in Table A2. Three and eight models were tested for substrate consumption and CA production, respectively. The fit evaluation of the mathematical models and kinetic parameters were calculated according to Coelho et al. [18]. The data obtained were useful for plotting the kinetic modeling graphs using the MATLAB® software R2016b version.

3. Results and discussion

3.1. Yields, maximum productivity, and selectivity

On the 4th day of the tests, it was detected the maximum CA productivity (μ_m) for HPr and HBU, and on the 7th day for HAc, isovaleric

acid (i-HVa), valeric acid (HVa), and caproic acid (HCA). The highest yields achieved were HPr, HAc, and HBU, as shown in Table 1. The average μ_m was 278.92 ± 62.46 mg L⁻¹ d⁻¹, and the total production CA yield (Y_{2CA}) was 0.62 mg acids mg COD_A⁻¹, corresponding to 0.82 mg COD_{CA} mgCOD_A⁻¹ (Y_{1CA}). Yin et al. [21] analyzed the anaerobic fermentation of glycerol PA in batch reactors (500 mL working volume) and used an F/M ratio of 3.3 gCOD gVSS⁻¹. In a condition similar to our study, the authors found a yield of 0.51 mg COD_{CA} mgCOD_A⁻¹, therefore very close to that found in the present experiment.

Similarly, there was a higher selectivity in the production and yield of HAc, HPr, and HBU. Literature reports that HAc is often the primary compound produced with the highest selectivity (30–80%) in organic waste fermentation, while HVa and other longer chain CA are synthesized with low selectivity [22]. Besides, many studies associate HPr high selectivity with the use of RG as a substrate in bioprocesses. Shen et al. [23] observed that the selectivity for HPr was 90.5%, using organic waste with 84.7% carbon in the form of glycerol.

The production of i-HVa and HVa is strongly related to the fermentation of protein-rich residues, according to studies conducted by Shen et al. [24]. They demonstrated that tofu and egg white fermentation promoted HVa synthesis with a selectivity of 18–25%, being the second acid produced in higher concentration after HAc, which helps to understand HVa low selectivity in the current research.

Biogas chromatographic analysis showed that chloroform inhibited methanogenesis since methane (CH₄) concentrations were not detected in the biogas, confirming the methanogenic inhibitor efficiency [8].

According to Biebl [25] and Viana et al. [26], glycerol can be metabolized anaerobically by either oxidative or reductive pathways. The oxidative pathway consists of the initial conversion of glycerol by the enzyme glycerol dehydrogenase into dihydroxyacetone, phosphorylated by dihydroxyacetone kinase. After phosphorylation, dihydroxyacetone is converted either to succinic acid, which is subsequently transformed into HPr, or pyruvate by the pyruvate-ferredoxin oxidoreductase enzyme (present in strict anaerobes). Subsequently, pyruvate acts as a precursor for simpler compounds synthesis, such as HAc, HBU, formic acid, lactic acid (HLA), ethanol (EtOH), hydrogen (H₂), and carbon dioxide (CO₂). Pyruvate proportions destined for each metabolic pathway depends on the environmental conditions, the enzymes that mediate the reactions, and the microorganisms present in the inoculum [27].

In contrast, the reductive pathway occurs through glycerol dehydration by glycerol dehydratase enzyme, which uses vitamin B12 as a cofactor and catalyzes the synthesis of 3-hydroxypropionaldehyde, which is reduced to 1,3-PDO by the NADH-H⁺ 1,3-propanediol dehydrogenase-dependent enzyme, regenerating NAD⁺. The total glycerol conversion never takes place entirely by the reductive route since it is necessary to produce an additional reducing equivalent. The main factor that seems to affect 1,3-PDO production is the microbial species involved in the bioprocess [26,28]. Since there was no 1,3-PDO production in this work, it is possible to infer that microorganisms mostly used the oxidative pathway. Other authors found a similar profile of CA production. Silva et al. [29] studied the potential for RG acidification in anaerobic batch reactors and showed that 80% of the acids produced corresponded to HAc, while HPr reached less than 20%.

Dams et al. [7] investigated the CA production from RG (5 g L⁻¹) in batch reactors (100 mL working volume) inoculated with anaerobic

Table 1
Maximum productivity, selectivity, and yields of carboxylic acids.

Parameter	HAc	HPr	HBU	i-HVa	HVa	HCA
^a Yield	0.36 ± 0.02	0.16 ± 0.01	0.09 ± 0.01	0.02 ± 0.01	3.7 ± 2.5	5.9 ± 4.0
^b Maximum productivity	131.4 ± 17.3	136.3 ± 5.2	104.3 ± 28.4	5.7 ± 1.1	1.7 ± 0.3	2.7 ± 0.3
^c Selectivity	57 ± 1.2	26 ± 1.3	13 ± 0.8	2 ± 1.3	1 ± 0.4	1 ± 0.6

a) Yield of CA about the available COD (mg acids mgCOD_A⁻¹); b) Maximum productivity of CA (mg CA L⁻¹ d⁻¹); c) Percentage of a specific acid about the total acids produced (%).

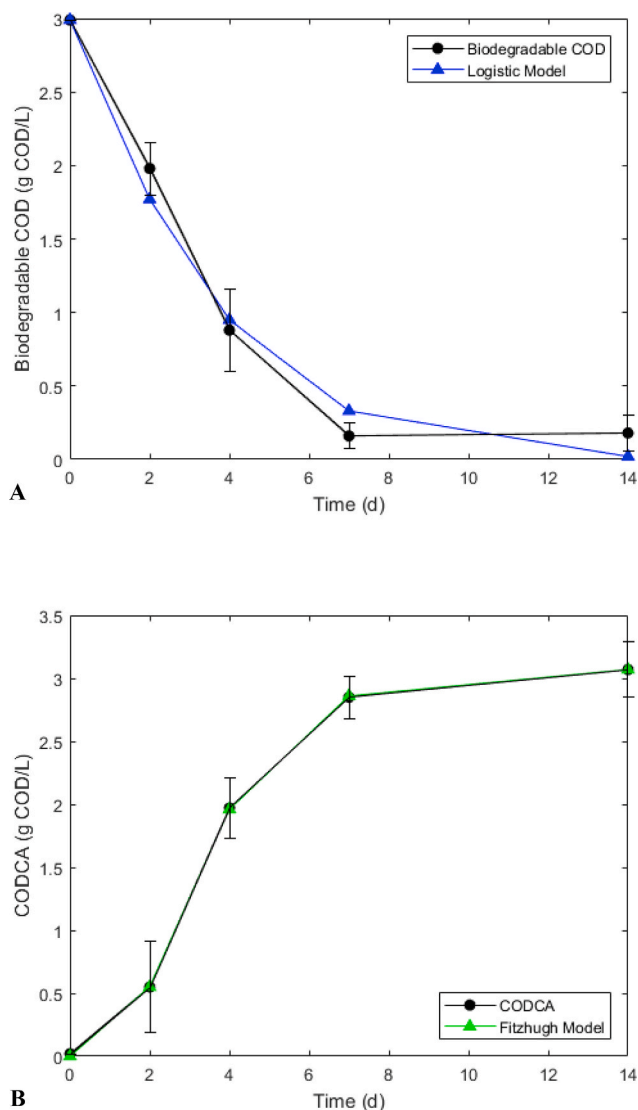


Fig. 1. Kinetic curves of (A) substrate consumption (Logistic model) and (B) total carboxylic acids production in its COD form (Fitzhugh model).

sludge (10 g) at room temperature (37 °C), pH 6.5, and stirring at 120 rpm for 14 days. Chloroform 0.05% (v/v) was also the adopted strategy to inhibit methanogenesis. The authors observed that there was a 100% conversion of the RG applied and reached higher concentrations of HAC (0.30 g L⁻¹), HPr (0.52 g L⁻¹), HBU (0.62 g L⁻¹), and 1,3-PDO (0.92 g L⁻¹). HCA concentrations were not detected.

In our essays, 95% of the applied RG was converted, and the following concentrations were found (in g L⁻¹): 0.31 (HAC), 0.14 (HPr), and 0.07 (HBU). Besides, i-HVa, HVa, and HCa were produced in low concentrations (0.01 g L⁻¹). In contrast to the results obtained by Dams et al. [7], 1,3-PDO was not detected, as previously mentioned. Despite the similar CA production profile, some factors may have contributed to the differences observed, such as the RG concentration, pH, temperature, type and treatment of the inoculum, and F/M ratio [27].

There was also HCA production, a medium-chain CA with a higher value-added than shorter CA, even with low yields (Table 1). An HBU peak was observed on the 4th day of the experiment, reaching 423.4 ± 113.48 mg L⁻¹. However, the following consumption was observed, reaching 318.5 ± 31.62 mg L⁻¹ on the experiment's last day. This decrease in HBU concentration may indicate that this acid was consumed for chain elongation. HCa could have been elongated in the presence of EtOH, HLa, or H₂ produced by different metabolic pathways of RG

conversion since there was no external insertion of electron donors [30]. Thus, the use of RG as raw material for anaerobic fermentation and medium-chain CA production seems to be propitious.

3.2. Kinetic study of substrate consumption and total carboxylic acids production in COD form

The biodegradable substrate present at the beginning of the batch (2.99 g COD L⁻¹) was gradually consumed over the fourteen days of the experiment, as shown in Fig. 1A. The soluble fraction corresponded to 85% of the organic matter present in the bioreactors on the first day of the test, indicating that the substrate was readily available to acidogenic microorganisms and showing RG biodegradability [31]. Glycerol is a hydrolyzate of the lipids present in complex organic matter. Since most of the residue was composed of glycerol in its free form, the hydrolysis of

Table 2
Kinetic parameters estimated from the modeling of substrate consumption and total carboxylic acids production in its COD form.

Model	Parameters	Value
Substrate consumption		
First-order	k_B (d ⁻¹)	0.28 ± 0.04
	R^2	0.972
	NRMSE	6.539
Monod with Growth	K_S (g COD L ⁻¹)	8.53 ± 0.14
	X (g VSS L ⁻¹)	4.69 ± 0.03
	μ_{max} (d ⁻¹)	0.33 ± 0.03
	k_L (L g COD ⁻¹ d ⁻¹)	0.04 ± 0.00
	k_B (d ⁻¹)	0.18 ± 0.02
	R^2	0.974
Logistic	NRMSE	6.357
	k_L (L g COD ⁻¹ d ⁻¹)	0.23 ± 0.03
	k_B (d ⁻¹)	0.05 ± 0.01
Total carboxylic acids COD production	R^2	0.983
	NRMSE	5.138
	First-order	
First-order	k_{CA} (d ⁻¹)	0.23 ± 0.05
	R^2	0.930
	NRMSE	10.594
Second-order	k_{CA}'' (L g COD ⁻¹ d ⁻¹)	0.13 ± 0.05
	R^2	0.834
	NRMSE	16.248
Fitzhugh	k_{CA} (d ⁻¹)	0.61 ± 0.05
	n	4.87 ± 2.13
	R^2	0.999
Cone	NRMSE	0.382
	k_{CA} (d ⁻¹)	0.30 ± 0.06
	n	3.16 ± 0.57
BPK	R^2	0.999
	NRMSE	0.853
	m	0.80 ± 0.03
Monomolecular	t_0 (d)	1.05 ± 0.02
	μ_m (g COD L ⁻¹ d ⁻¹)	0.59 ± 0.08
	k_{CA} (d ⁻¹)	0.19 ± 0.03
	R^2	0.930
	NRMSE	10.594
	k_{CA} (d ⁻¹)	0.25 ± 0.04
Modified Gompertz	λ (d)	0.26 ± 0.13
	R^2	0.936
	NRMSE	10.100
Logistic	μ_m (g COD L ⁻¹ d ⁻¹)	0.75 ± 0.08
	λ (d)	1.29 ± 0.59
	R^2	0.999
Logistic	NRMSE	0.677
	μ_m (g COD L ⁻¹ d ⁻¹)	0.78 ± 0.15
	λ (d)	1.52 ± 0.67
	R^2	0.996
	NRMSE	2.390

k_B : substrate degradation rate constant; K_S : saturation constant/Monod constant; X : final biomass concentration; μ_{max} : maximum microbial growth rate; k_L : Logistic model constant; k_{CA} : first-order CA production rate constant; n : shape constant; λ : lag phase time; m : BPK constant model; R^2 : coefficient of determination; NRMSE: normalized mean square error.

lipids and other biomolecules possibly present, such as long-chain fatty acids (LCFA), was not a limiting stage of anaerobic fermentation [21, 32].

Substrate consumption followed first-order kinetics and could be described by the three tested models, especially by the Logistic model (Fig. 1A), which provided the highest R^2 and the lowest values of NRMSE (Table 2). Although the Logistic model was the best to describe substrate consumption, the Monod with growth model was also satisfactory to describe this process. K_S was estimated from the modeling, resulting in a value of 8.53 ± 0.14 gCOD L⁻¹ (Table 2). Thus, this model application becomes useful in CA production studies because it allows the affinity analysis of acidogenic microorganisms to the applied substrates, enabling to compare the biomass preference by a particular type of substrate. According to the Monod with growth model, COD consumption for cell growth was practically negligible, as evidenced by the final concentration of 4.69 ± 0.03 gVSS L⁻¹, being slightly higher than the average initial biomass concentration (4.62 ± 0.51 gVSS L⁻¹).

Among the eight models tested for CA production in terms of organic matter, the Fitzhugh model was the one that best described the cumulative CA curve (Table 2). The Modified Gompertz model also showed an excellent fit but returned higher NRMSE values.

The cumulative CA curve (Fig. 1B) resembles an elongated S (elongated S shape), indicating a delay phase followed by an increase after this period, revealing a sigmoidal shape. Such a kinetic profile is related to the degradation of complex substrates than simple carbohydrates [33]. From this profile, the fit of sigmoidal models, such as Logistic and Modified Gompertz, is confirmed to describe this bioprocess's kinetics (Table 2). However, as CA production had a short delay time, exponential models, such as the Cone and Fitzhugh model, also describe also describe CA production kinetics satisfactorily.

The experimental data allow inferring that the kinetics of CA production from RG is similar to the anaerobic kinetic CH₄ production, whose cumulative curves generally may have a discrete lag phase and

can be described by both sigmoidal and exponential functions. The existence of a lag phase in the RG anaerobic fermentation is in agreement with the experiments performed by Silva et al. [29].

Three models (Monomolecular, Modified Gompertz, and Logistic) estimated the lag phase between approximately zero (0.26 ± 0.13 d, Monomolecular model) and two days (1.52 ± 0.67 d, Logistic model). The form factor n of the Fitzhugh model ($n = 4.89 \pm 2.13$) and Cone ($n = 3.16 \pm 0.57$) also indicates a delay period.

Despite being a readily available substrate, highly reduced glycerol is more difficult to break than carbohydrates under fermentative conditions (e.g., in the absence of external electron acceptors), explaining the mildly delay phase that was observed. Besides, RG impurities – which usually make up 20% of the residue – such as sodium chloride (NaCl), LCFA, and sulfates can make glycerol metabolization more difficult [21, 31]. From the waste physical-chemical characterization (topic 2.1), it was possible to infer that glycerol had some of the impurities commonly reported in the literature. Despite this, the abundance of microorganisms in the inoculum allowed the consortium to adapt to the environment after the first 48 h of testing.

The maximum production rate (μ_m) was calculated using the Modified Gompertz, Logistics, and BPK. The Modified Gompertz and Logistic models estimated values close to μ_m , while the lowest value was found with the BPK model (Table 2). The discrepancies between the models' delay times compared with the experimental data may be related to the parameters taken into account by each mathematical formula.

3.3. Kinetic study of individual carboxylic acids production

In general, exponential models were more suitable to simulate individual CA production from RG anaerobic fermentation. Only HAC had its production better described by a sigmoidal model, e.g., the Logistic model (Table 3). Among the evaluated CA, HPr was the one with the best-described kinetics, presenting an exponential curve modeled

Table 3
Kinetic parameters estimated from the modeling of individual carboxylic acids production.

Model	Parameter	HAc	HPr	HBu	i-HVa	HVa	HCa
First-order	k_{CA} (d ⁻¹)	0.125	0.396	0.509	0.140	0.135	0.122
	R^2	0.857	0.955	0.626	0.935	0.915	0.862
	NRMSE	14.852	8.348	36.653	9.276	10.874	14.554
Second-order	k_{CA}'' (L g ⁻¹ d ⁻¹)	0.133	1.380	4.416	3.394	10.771	5.849
	R^2	0.748	0.886	0.518	0.833	0.810	0.753
	NRMSE	19.737	13.337	41.619	14.897	16.286	19.470
Fitzhugh	k_{CA} (d ⁻¹)	0.456	1.135	4.346	0.289	0.320	0.398
	n	8.805	8.061	10223.70	2.739	3.442	6.422
	R^2	0.999	0.999	0.824	0.995	0.996	0.998
	NRMSE	1.376	0.924	25.152	2.491	2.236	1.626
Cone	k_{AC} (d ⁻¹)	0.177	0.458	0.462	0.197	0.191	0.176
	n	3.902	3.94	19.181	2.513	2.761	3.498
	R^2	0.998	0.999	0.824	0.989	0.992	0.996
	NRMSE	1.579	0.625	25.144	3.868	3.351	2.488
BPK	m	0.806	0.574	0.508	0.788	0.859	0.809
	t_0 (d)	1.928	1.872	1.902	1.928	1.215	1.934
	μ_m (g L ⁻¹ d ⁻¹)	0.137	0.154	0.099	0.007	0.002	0.003
	k_{CA} (d ⁻¹)	0.103	0.259	0.311	0.113	0.117	0.101
	R^2	0.857	0.955	0.626	0.935	0.915	0.862
	NRMSE	14.852	8.348	36.653	9.276	10.874	14.554
Monomolecular	k_{CA} (d ⁻¹)	0.144	0.144	0.522	0.153	0.151	0.142
	λ (d)	0.808	0.808	0.126	0.491	0.601	0.858
	R^2	0.886	0.278	0.629	0.948	0.933	0.894
	NRMSE	13.29	33.571	36.510	8.320	9.642	12.764
	μ_m (g L ⁻¹ d ⁻¹)	0.233	0.254	0.846	0.195	0.093	0.005
Modified Gompertz	λ (d)	2.812	1.03	1.938	3.884	3.932	2.594
	R^2	0.999	0.999	0.824	0.799	0.823	0.998
	NRMSE	1.072	0.931	25.144	16.370	15.741	1.872
	μ_m (g L ⁻¹ d ⁻¹)	0.247	0.224	0.689	0.218	0.073	0.099
Logistic	λ (d)	3.198	0.918	1.946	3.897	3.914	3.935
	R^2	0.999	0.998	0.824	0.799	0.823	0.844
	NRMSE	1.013	1.765	25.144	16.370	15.741	15.476

k_{CA} : first-order CA production rate constant, k_{CA}'' : second-order CA production rate constant; n : form constant; λ : lag phase time; μ_m : maximum CA productivity; m : constant of the BPK model; R^2 : coefficient of determination; NRMSE: normalized root mean square error.

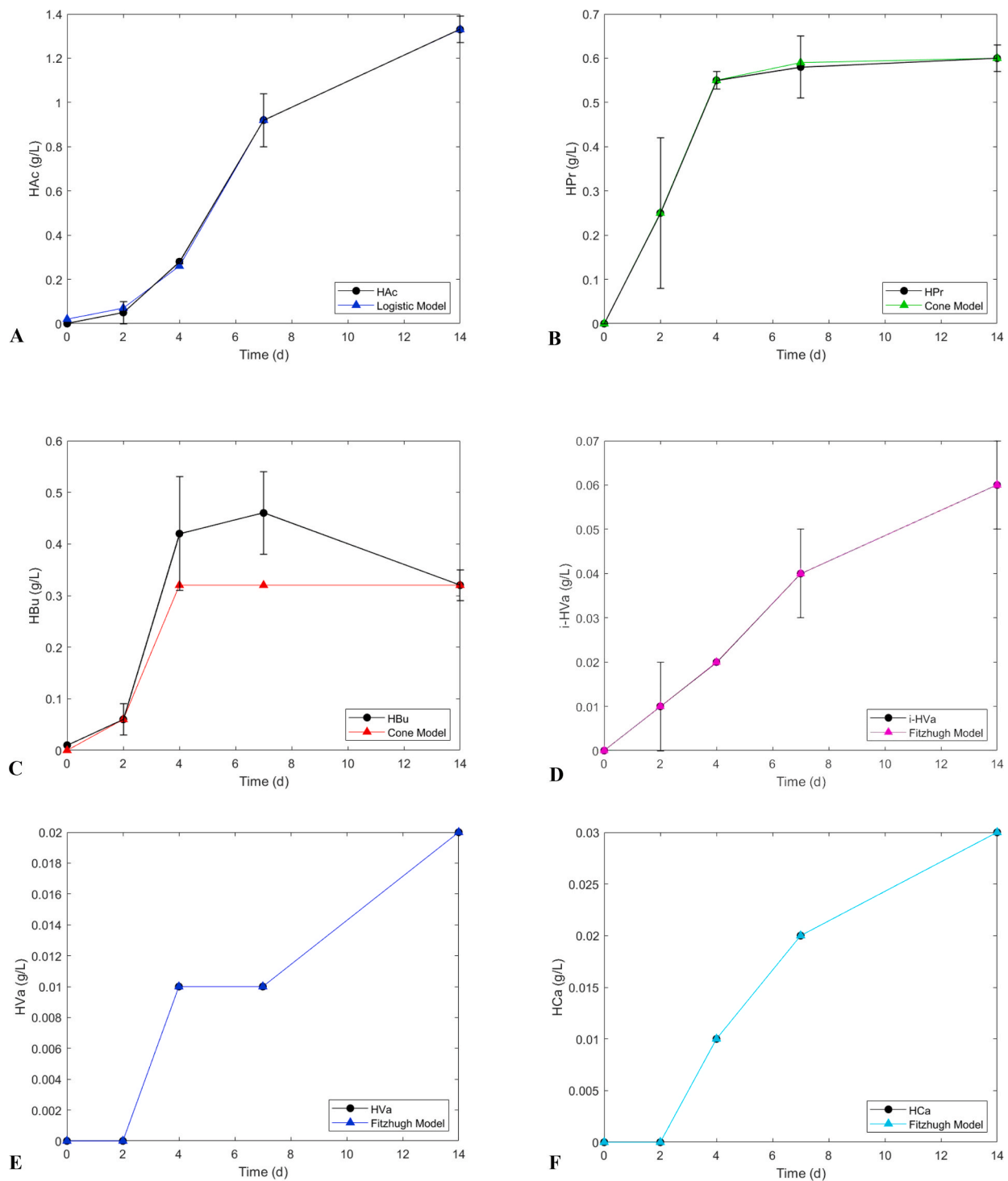


Fig. 2. Carboxylic acids production kinetic curves. (A) HAC. (B) HPr. (C) HBu. (D) i-HVa. (E) HVa. (F) HCa.

optimally by the Cone model (Fig. 2B).

The exception was the adjustment to the HBu progression curve (Fig. 2C), likely caused by the consumption of HBu by microorganisms for oxidative processes, such as chain elongation via reverse β -oxidation [30]. The models selected to describe CA production from RG do not consider the possibility of a decrease in product concentration, reason by which they were not suitable to describe HBu experimental data satisfactorily.

Evaluating by the mathematical models that best described the cumulative CA curve, it is noted that the first-order CA production rate

constant (k_{CA}) was higher for HBu ($k_{CA} = 0.408 \text{ d}^{-1}$, Cone model) and lower for i-HVa ($k_{CA} = 0.289 \text{ d}^{-1}$, Fitzhugh model). HPr, HVa, and HCa presented intermediate values, with k_{CA} equal to 0.458, 0.320, and 0.398 d^{-1} , respectively. HAC progression was best described by the Logistic model, which does not estimate the k_{CA} value. However, the Fitzhugh model, which also adjusted satisfactorily, estimated k_{CA} in 0.456 d^{-1} .

The BPK model was the most reliable in predicting maximum acid productivity (μ_m), providing the values of ($\text{mg L}^{-1} \text{ d}^{-1}$): 137 (HAC), 154 (HPr), 99 (HBu), 7 (i-HVa), 2 (HVa) and 3 (HCa). The models that

estimated the lag phase time (Monomolecular, Modified Gompertz, and Logistic) showed the occurrence of a delay for the individual production of all CA (Table 3). This fact is also easily observed from the individual CA progression curves (Fig. 2), except for the HPr curve (Fig. 2B), which showed an apparent exponential profile. HAC, for instance, shows a sigmoidal profile.

The lag time for HPr production was the shortest among the estimated values ($\lambda = 1.03 \pm 0.40$ d, Modified Gompertz model). The models estimated the lag phase time between 0.1 (HPr production, Modified Gompertz model) and 3.9 (HCA, Logistic model). A mildly lag phase may be attributed to the highly reducing environment in the absence of external electron acceptors and the impurities present in the RG.

The parameters estimated from the kinetic modeling in this work can be useful for new bioreactors design and strategies to control and simulate in bioprocessing software for CA production using RG as substrate and anaerobic sludge as inoculum [34].

4. Conclusions

Residual glycerol (RG) from biodiesel production appeared to be a readily biodegradable carbon source to the acidogenic microorganisms. A yield of 0.62 mg acids mg COD_A⁻¹ was achieved, corresponding to 0.82 mg COD_{CA} mg COD_A⁻¹. The production of a medium-chain CA – caproic acid (HCA) – was detected even without electron donors' supplementation. Propionic (HPr), butyric (HBu), iso-valeric (i-HVa), valeric (HVa), and caproic (HCA) acids production kinetics are well described by exponential models (Fitzhugh and Cone models), while a sigmoidal model (Logistic model) is more suitable to describe acetic acid (HAC) kinetics production. Therefore, RG seems to be a promising substrate for CA production in the anaerobic fermentation process.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.biombioe.2020.105874>.

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