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EXPERIMENTAL ARTICLES

Anaerobic Degradation of Organic Waste: An Experience in Mathematical Modeling

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Abstract—Some key aspects of organic waste degradation were analyzed by means of mathematical models using data obtained in laboratory reactors. It was shown that an essential condition of effective methane production is the balance between sequential and parallel stages not resulting in accumulation of intermediate products that are potential inhibitors of the process. Decreased initial concentration of organic matter (dilution) and the introduction of seed culture (a methanogenic microbial community) favor the balancing of the process. Decomposition of easily degradable organic substances may lead to excessive accumulation of volatile fatty acids and acidification of the medium, which, in turn, blocks the degradation of difficult-to-degrade compounds. If the process is unbalanced, agitation eliminates the initiation centers for methanogenesis by averaging the reagent concentrations, which results in complete cessation of methane production. Such centers may be multicellular aggregates of *Methanosarcina* sp.

Key words: organic matter, anaerobic microbial community, balance of hydrolysis and methanogenesis rates, initiation centers for methanogenesis.

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Huge amounts of solid and liquid wastes accumulate as a result of human economic activities: farm animal manure, slaughterhouse wastes, landfill garbage, sludge from municipal water treatment facilities, etc. If the methane generated by such wastes is not utilized, its tremendous emissions into the atmosphere will contribute to global warming [1].

Anaerobic digestion of organic matter (OM) includes four sequential stages: polymer hydrolysis, acidogenesis, acetogenesis, and methanogenesis. If the rates of acidogenesis and methanogenesis are unbalanced [2], the system accumulates volatile fatty acids (VFAs). This results in the inhibition not only of methanogenesis (the terminal stage of the process), but also of hydrolysis (the primary stage) [3]. With balanced rates of separate stages, hydrolysis, being the first and slowest stage, limits the total rate of the whole process of OM conversion into methane.

Traditionally [4], calibration and verification of the mathematical models of OM degradation take into consideration only the dynamics of the chemical components. Modern analysis of behavior of anaerobic microbial communities includes the measurements of the concentrations of chemical components, molecular biological techniques, and mathematical modeling. In the present work, mathematical modeling was used to analyze some key aspects of OM degradation using data obtained in laboratory reactors.

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MATERIALS AND METHODS

The behavior of an anaerobic microbial community was analyzed using the individual components of the previously developed Methane generic simulation model [5]. In this model, the rate of transformation of the limiting substrate by the *i*th microbial group is represented as a product of several functions:

$$\rho_{SB_i} = \rho_{SB_{i\max}} FL_i FT_i FI_i B_i, \quad (1)$$

where B_i is the concentration of microorganisms; $\rho_{SB_{i\max}}$ is the maximum specific rate of substrate consumption by the *i*th microbial group under optimal conditions; and FT_i, FL_i, and FI_i are the functions describing the temperature dependence and the limiting and inhibition mechanisms, respectively. Methane is the first mathematical model of an anaerobic process in world practice that may be used by common researchers and engineers, rather than professional programmers (www.methane.da.ru). At the same time, individual processes could be described by simpler models, allowing more definite interpretation of existing or experimentally obtained data.

Assuming that conversion of the substrate (OM) into methane is accompanied by formation of only

Kinetic parameters of model (2)*

Livestock slurry	k, days^{-1}	ρ_m, days^{-1}	K_S, g l^{-1}	Y, g g^{-1}	K_h, g l^{-1}	K_m, g l^{-1}
Fattening swine	0.075	2.2	0.05	0.05	30	30
Ducks	0.07	2.2	″	″	12	12
Sows	0.04	2.0	″	″	11	11
Dairy cattle	0.03	2.0	″	″	11	11

* For all types of wastes, $n_h = n_m = 3$ in the inhibition function using parameters K_h and K_m.

one intermediate product (VFA), we proposed the following system of differential equations:

$$\begin{cases} \dfrac{dW}{dt} = -kWf_h(S) \\ \dfrac{dS}{dt} = \chi kWf_h(S) - \rho_m f_m(S)\dfrac{SB}{K_S + S} \\ \dfrac{dB}{dt} = Y\rho_m f_m(S)\dfrac{SB}{K_S + S} - k_d B \\ \dfrac{dP}{dt} = (1 - Y)\rho_m f_m(S)\dfrac{SB}{K_S + S}, \end{cases} \quad (2)$$

where W, S, and B are the concentrations of OM, VFA, and methanogenic biomass, respectively; dP/dt is the methane production rate; t is time; k is the hydrolysis rate constant; ρ_m is the maximum specific rate of VFA utilization; k_d is the constant of methanogenic biomass decay rate; χ is the stoichiometric coefficient; K_S is the half-saturation constant for VFA utilization; and Y is the yield coefficient. In system (2), VFA utilization is described by the conventional Monod function and organic matter hydrolysis is described by the first-order function by the substrate. Dimensionless functions $f_h(S)$ and $f_m(S)$ describe the inhibition of methanogenesis and hydrolysis, respectively, by high VFA concentrations. Diffusion processes for VFA and methanogenic biomass were additionally introduced into model (2) to describe the effect of agitation. As a result, the model became distributed (in partial derivatives). The kinetic parameters of model (2) are presented in the table.

RESULTS AND DISCUSSION

The balance of hydrolysis/acidogenesis and methanogenesis rates. The ratio of initial concentrations of methanogenic biomass (inoculum) and substrate (OM) can obviously have a strong effect on the dynamics of a system. Under balanced degradation, polymer hydrolysis is the rate-limiting stage of the whole process and the curve of methane accumulation corresponds to the first-order kinetics by organic matter concentration (Fig. 1). Dilution of OM and introduction of microorganisms have a weak effect on the dynamics of methane accumulation, because fatty acids are not accumulated.

At high initial concentration of organic matter, when the quantity of methanogenic microorganisms becomes insufficient, intermediate substances accumulate, inhibiting methane production. In this case, methanogenesis becomes the rate-limiting stage of the process. The curve of methane accumulation now does not correspond to the first-order kinetics (Figs. 2–4). The decrease of initial concentration of organic matter (dilution) and addition of a seed culture of methanogenic microorganisms into the reactor (inoculation) contributes to the balance. The higher the initial OM concentration, the greater initial concentration of methanogenic microorganisms is required for the process to be balanced.

Conventionally, the kinetics of methane production is determined with allowance for the methane generated from residual amounts of organic matter present in the inoculum (control). Methane generation is sped up by adding an ample quantity of the inoculum into the reactor. If the volume of introduced microbial seed culture is comparable with the reactor volume, it is necessary to take into account that methane productions in the reactor and in the inoculum are not independent but can mutually influence each other [7].

The balance of destruction rates of easily degradable and difficult-to-degrade organic substances. Decomposition of easily degradable organic substances may lead to excessive accumulation of VFA and acidification of the medium (Fig. 5) which, in turn, blocks decomposition of difficult-to-degrade compounds. Introduction of the seed culture of methanogenic microorganisms provides efficient degradation of

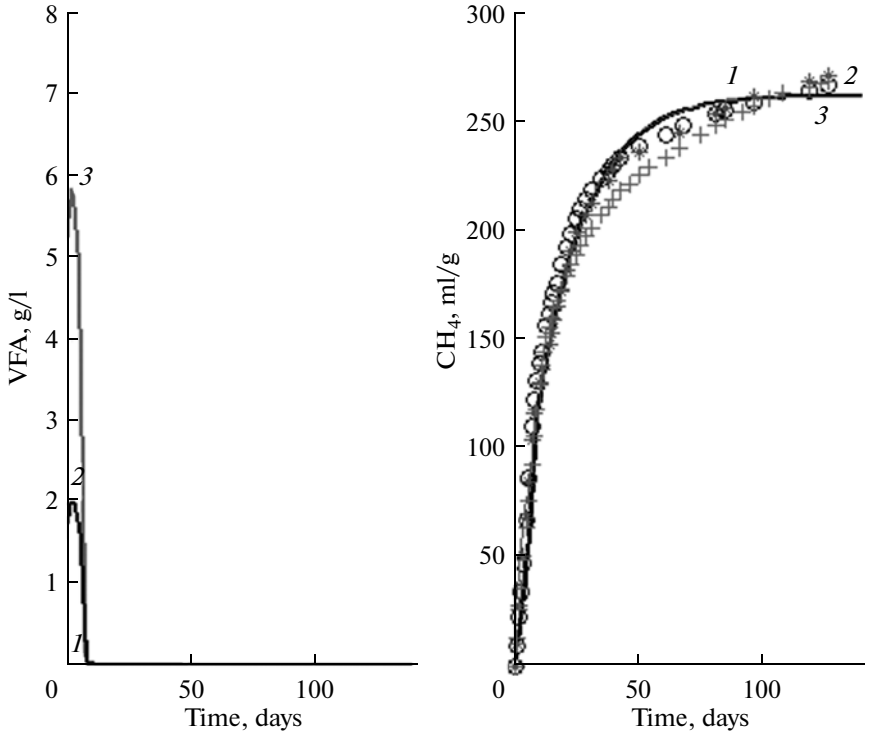

Fig. 1. Experimental data and results of the modeling of the behavior of an anaerobic system degrading fattening swine slurries: diluted and inoculated waste (*1*), diluted waste (*2*), and undiluted waste (*3*). The symbols correspond to the experimental data [6]; the curves correspond to the solutions of dynamic model (2). Methane volume is normalized to the initial quantity of the substrate.

poorly degradable OM. This process is also facilitated by a decrease of the total OM quantity, when decomposition of easily degradable OM does not lead to acidification. Aeration, which results in forced oxidation of redundant VFA, after its termination provides rapid OM transformation into methane [9].

Contribution of aceticlastic and hydrogenotrophic methanogenesis. The major substrates for methanogenic microorganisms are acetate and hydrogen/carbon dioxide [10]:

$$CH_3COOH + H_2O \longrightarrow CH_4 + H_2CO_3, \quad (3)$$

$$4H_2 + H_2CO_3 \longrightarrow CH_4 + 3H_2O. \quad (4)$$

Two types of methanogenic microorganisms transforming acetate into methane are known [11]. *Methanosaeta* sp., characterized by low values of the half-saturation constant K_S, dominates at low acetate concentrations (below 1 mM), while *Methanosarcina* sp. predominates at high acetate concentrations.

Two-stage methane production during mesophilic fermentation of glucose has been described in [12], when hydrogenotrophic methanogenesis prevailed at the first stage with low pH values and aceticlastic methanogenesis dominated at the second stage with neutral pH.

At lower temperatures, the predominant mechanism of methane production is aceticlastic methanogenesis [13], because H_2/H_2CO_3 is preliminarily transformed into acetate by homoacetogenesis:

$$4H_2 + 2H_2CO_3 \longrightarrow CH_3COOH + 4H_2O. \quad (5)$$

The kinetics of methane generation from hydrogen and carbon dioxide with the formation of acetate as an intermediate product has been modeled in [12]. It has been shown that relatively low acetate concentrations (about 25 mM) at low temperatures may terminate aceticlastic methanogenesis.

Under thermophilic conditions and in the presence of inhibitors such as ammonium and VFA, the predominant mechanism of methane production is a process in which acetate is preliminarily oxidized to H_2 and H_2CO_3 [14]:

$$CH_3COOH + 4H_2O \longrightarrow 4H_2 + 2H_2CO_3. \quad (6)$$

This reaction is performed by acetate-oxidizing bacteria and occurs in syntrophic association with hydrogenotrophic methanogens transforming H_2 and H_2CO_3 into CH_4. The results of modeling presented in Figure 6 make it possible to estimate the biomass doubling time by the formula $T = 0.693/\mu_m$ [5], where μ_m is the maximum specific rate of biomass growth. From the dynamics of acetate oxidation and methane generation, we obtain generation times of 46 and 1.7 h for acetate-oxidizing bacteria and hydrogenotrophic methanogens, respectively. Thus, hydrogenotrophic

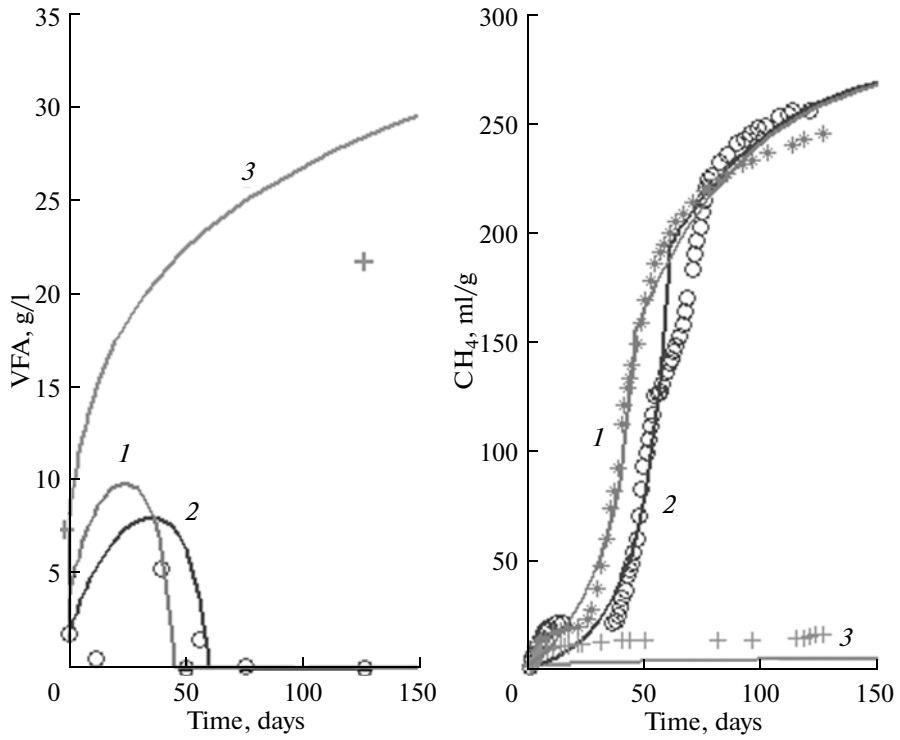

Fig. 2. Experimental data and results of the modeling of the behavior of an anaerobic system degrading dairy cattle slurries: diluted and inoculated waste (*1*), diluted waste (*2*), and undiluted waste (*3*). The symbols correspond to the experimental data [6]; the curves correspond to the solutions of dynamic model (2).

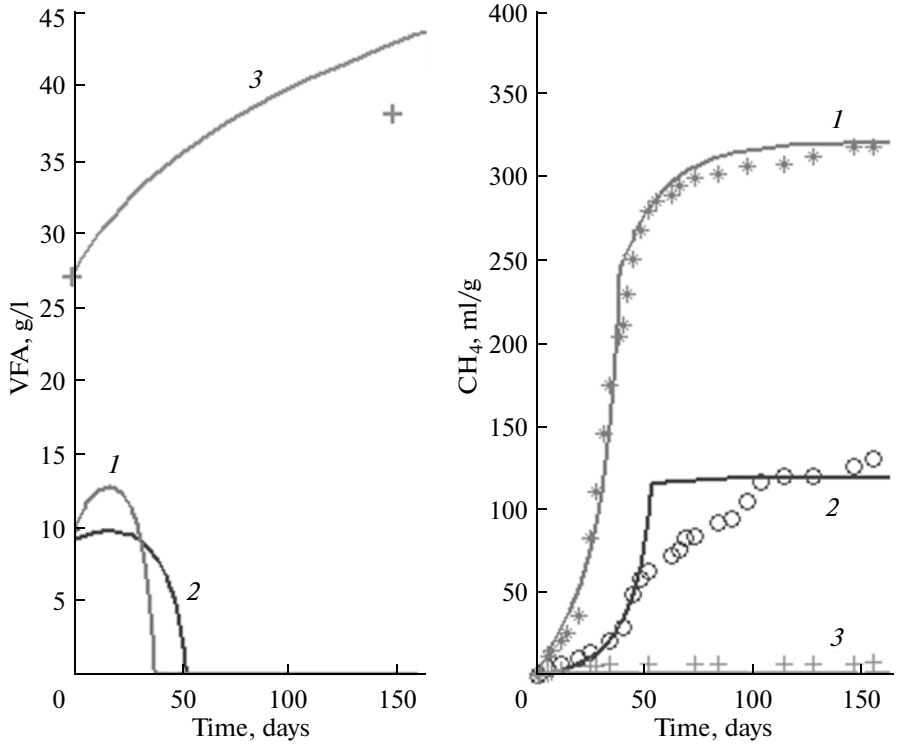

Fig. 3. Experimental data and results of the modeling of behavior of an anaerobic system degrading duck slurries. The designations are as in Figure 2.

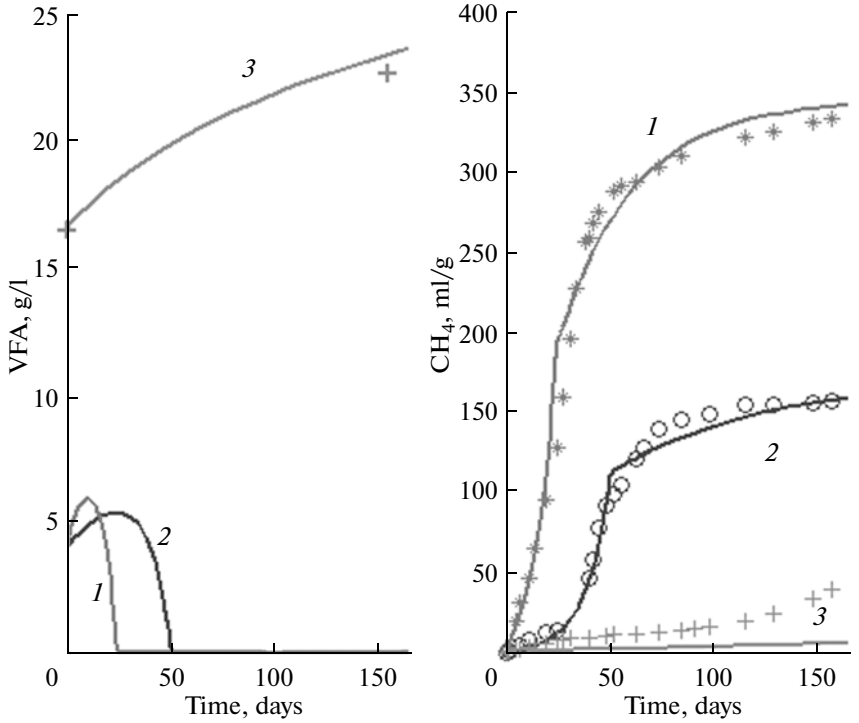

Fig. 4. Experimental data and results of the modeling of the behavior of an anaerobic system degrading sow slurries. The designations are as in Figure 2.

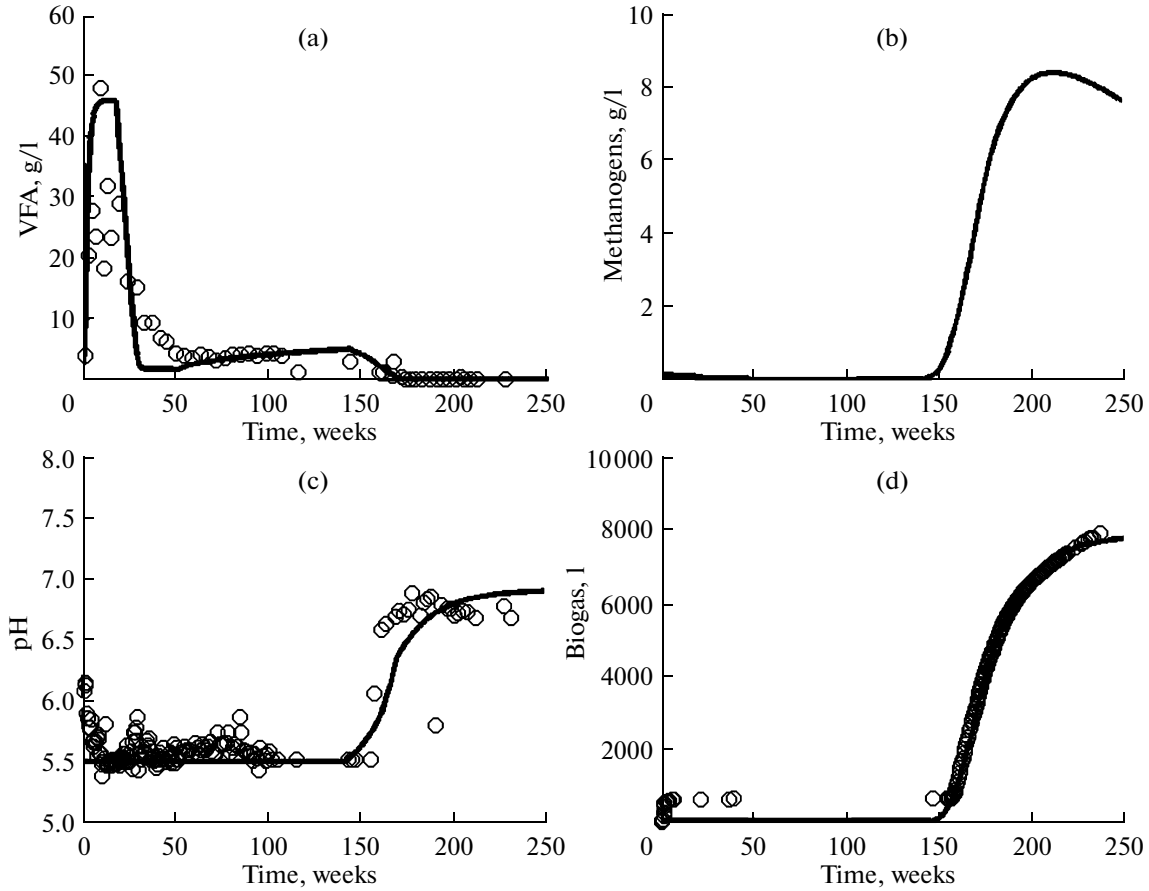

Fig. 5. Dynamics of VFA and methanogenic biomass concentrations in the leachate in the course of municipal solid waste degradation, with leachate averaged over the reactor volume pH, and total biogas volume in the cylinder reactor. The symbols correspond to the experimental data [8]; the curves correspond to the solutions of the dynamic model. Reactor volume: 100 l.

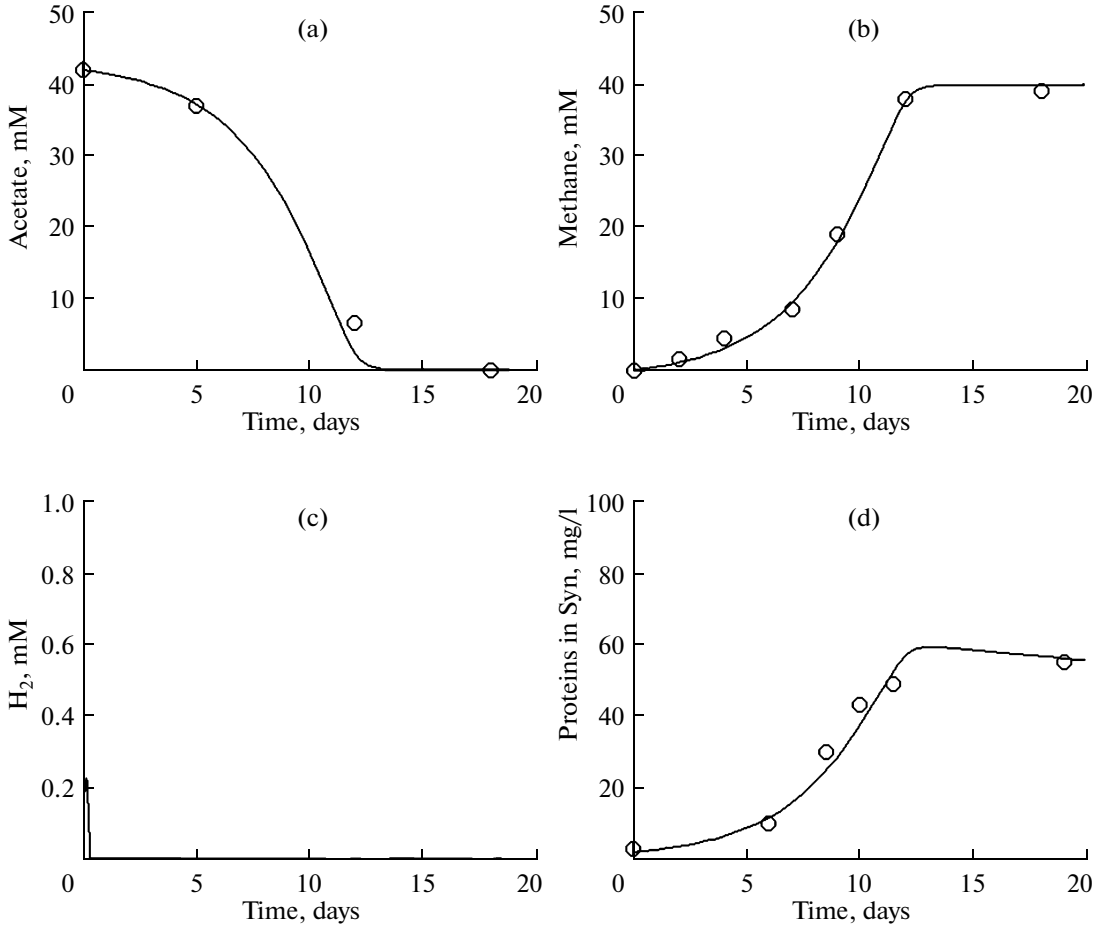

Fig. 6. Dynamics of the total methane volume during syntrophic acetate oxidation and the concentrations of acetate, hydrogen, and biomass measured by total protein content. The symbols correspond to the experimental data [14]; the curves correspond to the solutions of the dynamic model based on chemical equations (4, 6).

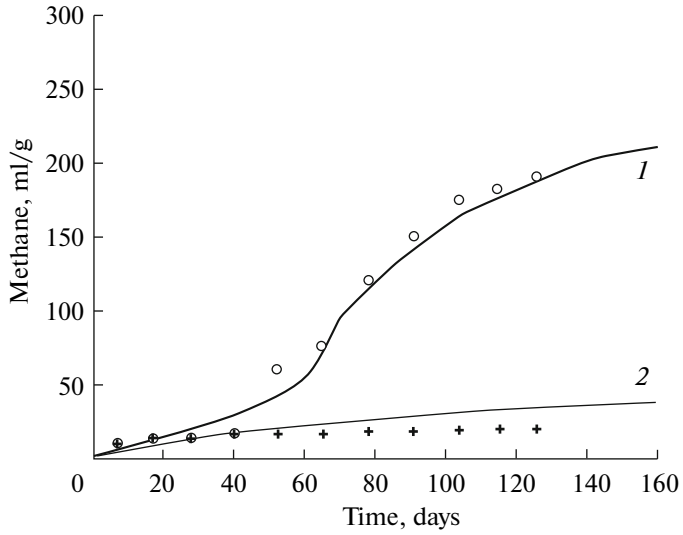

Fig. 7. The negative effect of agitation during anaerobic digestion of dairy cattle slurries. The symbols correspond to the experimental data [6]; the curves correspond to the solutions of the dynamic model with six peaks of initial concentrations of methanogenic biomass in space. Weak agitation (*1*) and intense agitation (*2*).

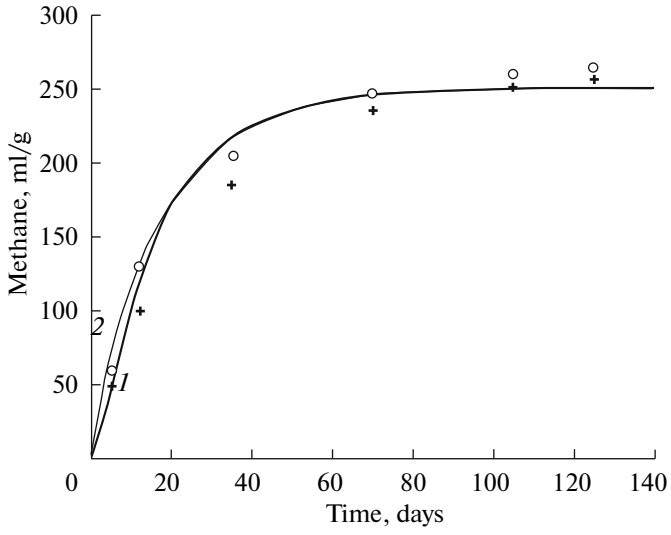

Fig. 8. Weak effect of agitation on methane production during the degradation of fattening swine slurries. Designations are as in Figure 7.

methanogens, unlike acetate-oxidizing bacteria, are extremely fast-growing microorganisms.

The kinetics of methane generation from municipal solid waste under mesophilic and thermophilic conditions was modeled in the work [15]. The thermophilic process was shown to involve acetate-oxidizing bacteria.

Initiation centers for methanogenesis. The literature conventionally emphasizes the necessity of adequate agitation in an anaerobic reactor so that the respective hydrolytic enzymes degrading solid and suspended OM will be evenly distributed over the reaction space. However, some works experimentally demonstrated [16] that stability and efficiency of the reactor improved at a lower intensity of agitation. We found that intense agitation in batch reactors terminated methanogenesis [17]. When the overall rate of OM transformation into methane is limited not by hydrolysis, but by methanogenesis, initial segregation of the zones of active methanogenesis and hydrolysis/acidogenesis (microniches) facilitates OM transformation into methane, whereas intense agitation blocks methane production. This effect is described by a distributed model developed on the basis of (2) (Fig. 7).

OM transformation into methane needs a sufficient quantity of methanogenic biomass in the initiation centers (seed culture). If the process is unbalanced, agitation eliminates the initiation centers for methanogenesis by averaging the reagent concentrations, which leads to complete cessation of methane production. In the case of a balanced process, when intermediate VFA concentration does not reach the inhibiting values and methane accumulation corresponds to the first-order curve, agitation has no effect on methane production (Fig. 8). Application of the distributed model for description of a continuous-flow reactor demonstrated that a plug-flow reactor, where the substances shift along the reactor axis, has higher stability of functioning compared to a complete-mixing reactor with the forced averaging of reagent concentrations [18].

In the work [19], fluorescent in situ hybridization (FISH) revealed that *Methanosarcina* sp., forming multicellular aggregates, is a dominant population in the course of municipal solid waste degradation. VFA concentrations in a multicellular aggregate may be much lower than in the solution, and methane generation is not inhibited. The multicellular aggregates of *Methanosarcina* sp. are potential initiation centers of methanogenesis. The metabolic diversity of *Methanosarcina* sp. allows these methanogens to exist on different substrates [20].

Dannenberg et al. [21] demonstrated that methane is produced from acetate by *Methanosarcina barkeri* inoculum only in the absence of agitation. The same authors found that intense agitation during the fermentation of OM from a soil suspension influenced the hydrogenotrophic methanogens much less than the acetioclastic methanogens.

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