

# Two-Phase Model of Hydrolysis Kinetics and Its Applications to Anaerobic Degradation of Particulate Organic Matter

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## ABSTRACT

The various equations of hydrolysis kinetics included into the generalized simulation model (METHANE) were tested on the anaerobic digestion of cellulose, sludge, and cattle manure. The good agreement between the model simulation results and experimental data was obtained. The Contois equation, taking into account a hydrolytic biomass, and the first-order equation with respect to the particulate substrate only, were shown to be the approximations of two-phase hydrolysis kinetics.

**Index Entries:** Anaerobic digestion; hydrolytic bacteria; simulation model; sludge; cattle manure; cellulose.

## INTRODUCTION

During anaerobic digestion the hydrolysis is normally rate-limiting if the substrate is in particulate form (1). The equations traditionally used for solids kinetics are the first-order equation in respect to the particulate substrate only, and the Contois equation that takes into account a hydrolytic biomass. According to Eastman and Ferguson (2), the first-order hydrolysis function is an empirical expression that reflects the cumulative effects of many processes. Large particles with a low surface-to-volume ratio would be hydrolyzed more slowly than small particles.

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The Contois equation has been more or less successfully tested on the anaerobic degradation of suspended solids. However, it was developed originally for dissolved substrates. For hydrolysis process a heterogeneous reaction system, in which a particulate substrate contacts with microbial cells and related enzymes, must be taken into consideration (3).

A structured approach to the modeling of anaerobic reactors has provided an important tool for the integral study of microbial ecology as well as the investigation of advanced control strategies of high-rate anaerobic reactors. There are few simulation models of anaerobic digestion that take into account hydrolysis of particulate organic matter (4,5). The simulation model (METHANE) (6,7) is among them.

In this article, the various versions of hydrolysis kinetics included into the model (METHANE) were tested on anaerobic digestion of sludge, cellulose, and cattle manure.

## SIMULATION MODEL OF ANAEROBIC DIGESTION

The following groups of model variables were included in the generalized simulation model of anaerobic digestion (METHANE).

1. suspended solids concentration ( $X_k$ ,  $k = 1, 2, 3$ );
2. active biomass concentration ( $B_i$ ,  $i = 1, 2, \dots, 10$ );
3. dissolved substrate concentration ( $S_j$ ,  $j = 1, 2, \dots, 15$ );
4. partial gas pressure ( $G_l$ ,  $l = 1, 2, \dots, 7$ ).

The processes of hydrolysis, acidogenesis, acetogenesis, and methanogenesis conducted by various groups of micro-organisms are considered (Fig. 1). It is assumed in the model that dissolved organic substrate  $S_1$  (DOS) is a mixture of carbohydrates (C), proteins (P), lipids (L), and an unknown organic substance (Z). The composition of the substrate  $S_1$  in an anaerobic reactor is formed under the effect of the following processes (Fig. 2):

1. The input of P, L, C and Z, contained in the influent of wastewater, in a dissolved form  $S_F$ ;
2. Hydrolysis of suspended organic matter  $X_F$ , coming into the reactor with the influent wastewater;
3. Lysis and hydrolysis of cell biomass  $B_{di}$ , formed in the reactor after the bacterial death.

A dissolved organic substrate  $S_1$  transforms into methane through the intermediate products  $S_j$ .

In Fig. 2 the rectangles denote substrate concentrations, lines outgoing from the rectangles denote the fractions constituting a given compound; value  $f$  shows the relative content of the C, P, L, and Z in the compound, and value  $p$  shows the relative content of biodegradable fraction of biopolymers. The particulate organic matter are divided into volatile VX, VB, and mineral

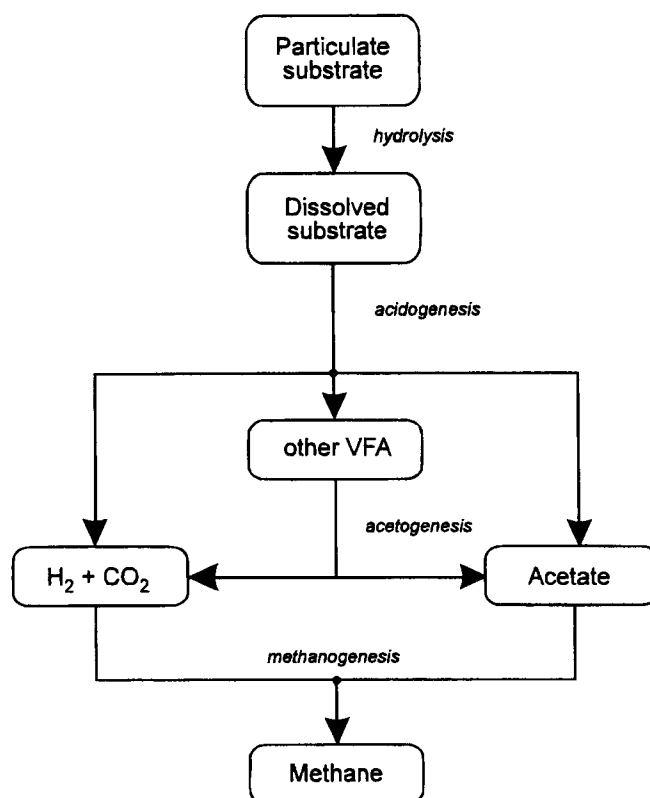


Fig. 1. Key stages of microbial transformation of complex organic matter.

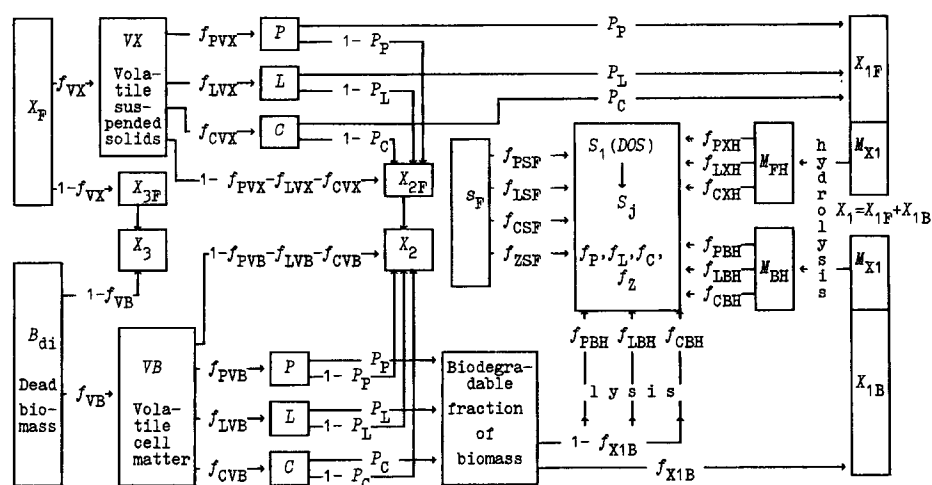


Fig. 2. Ways of formation of suspended and dissolved components in the anaerobic system.

$X_3$  fractions. Volatile organic matter  $VX$  and  $VB$  comprises  $P$ ,  $L$ ,  $C$ , and the nonbiodegradable substrates  $X_2$ . As result of hydrolysis of monomers of the degradable fraction of the influent suspended solids  $X_{1F}$  and the matter of the dead cells  $X_{1B}$ , the total mass of hydrolysis products will be equal to  $M_{FH}$  in the first case and  $M_{BH}$  in the second case (for simplicity below  $X_1$  will be written as  $X$  and the hydrolytic biomass will be written as  $B$ ).

Various versions of hydrolysis kinetics including the simplest first-order kinetics and the Contois equation are considered in the model <METHANE> for description of degradation of suspended solids:

$$v_H = k_H \cdot X \quad (\text{First-order}) \quad (1)$$

$$v_H = v_{mH} \cdot B \cdot \frac{X/B}{\tilde{K}_X + X/B} \quad (\text{Contois}) \quad (2)$$

where  $v_H$  is the rate of solids degradation,  $v_{mH}$  is the maximum specific hydrolysis rate,  $X$  and  $B$  are the concentrations of biodegradable suspended solids and hydrolytic biomass, respectively,  $k_H$  is the first-order kinetics constant,  $\tilde{K}_X$  is the half-saturation constant.

## RESULTS AND DISCUSSION

### N-Order Solids Degradation Kinetics

During the hydrolysis the particulate substrates contact with hydrolytic microbial cells and the released enzymes (8). Two main phases must be taken into account for a description of the hydrolysis kinetics. The first phase is bacterial colonization during which the hydrolytic bacteria cover a surface of the solids. The bacteria located on or near the particle surface release the enzymes and produce monomers, which can be utilized by the hydrolytic bacteria themselves, as well as by other bacteria. The daughter cells that fall off into the liquid phase are trying to attach to some new place of a particle surface. According to Hobson and Wheatley (3), when an available surface is covered with bacteria, the surface area will be changed at a constant depth per unit of time (second phase).

Following a two-phase approach let us write the equations for the concentration of hydrolytic biomass  $B$  and the concentration of suspended solids  $X$  in the continuous-flow stirred-tank digester, respectively:

$$\begin{aligned} \frac{dB}{dt} &= Y \cdot v_H(B, X) - k_d \cdot B - \frac{1}{\theta} \cdot B \\ \frac{dX}{dt} &= -v_H(B, X) + \frac{X_F - X}{\theta} \end{aligned} \quad (3)$$

where  $k_d$  is the rate coefficient of biomass decay,  $Y$  is the yield coefficient,  $X_F$  is the influent suspended solids concentration,  $\theta$  is the solids retention time, and the  $v_H(B, X)$  is the hydrolysis rate function that equals to

$$v_H(B, X) = \begin{cases} v_{mH} \cdot B, & \text{if } B < \tilde{B} \\ v_{mH} \cdot \tilde{B}, & \text{if } B \geq \tilde{B} \end{cases} \quad \begin{matrix} \text{(A)} \\ \text{(B)} \end{matrix} \quad (4)$$

where  $\tilde{B}$  is the saturated biomass concentration covering the contact surface of the hydrolyzed particles, and  $v_{mH}$  is the maximum specific hydrolysis rate.

Vavilin et al. (9) obtained the following generalized-type function that describes the rate of degradation of solids surface limited by a contact area between particles and bacterial mass

$$v_H = v_{mH} \cdot K \cdot X_F^{1-n} \cdot X^n \quad (5)$$

where  $K$  is the dimensionless constant and  $n$  is the degree coefficient. For the spherically symmetrical particles, it was obtained:

$$\tilde{B} = K \cdot X_F^{1/3} \cdot X^{2/3} \quad (6)$$

$$K = 6 \cdot \frac{\rho_B}{\rho_X} \cdot \frac{\delta}{d_{XF}} \quad (7)$$

$$v_H = v_{mH} \cdot K \cdot X_F^{1/3} \cdot X^{2/3} \quad (8)$$

where  $\rho_B$ ,  $\rho_X$  are the bacterial and solids density, respectively,  $\delta$  is the depth of bacterial layer;  $d_{XF}$  is the average input diameter of the particles hydrolyzed. It was assumed that the hydrolysis rate is limited by a contact area between particles and bacterial mass, and the size of the hydrolyzed particle is much greater than the depth of bacterial layer.

For the cylindrical symmetrical particles it was obtained:

$$\tilde{B} = K \cdot X_F^{1/2} \cdot X^{1/2} \quad (9)$$

$$K = 4 \cdot \frac{\rho_B}{\rho_X} \cdot \frac{\delta}{d_{XF}} \quad (10)$$

$$v_H = v_{mH} \cdot K \cdot X_F^{1/2} \cdot X^{1/2} \quad (11)$$

The case with  $n = 0$  corresponds to a disk-like particle. The first-order kinetics following from equation 5 at  $n = 1$  can be considered as an approximation of equation 8 or equation 11. Therefore, the hydrolysis constant  $k_H = v_{mH} \cdot K$  is the function of the ratio between the characteristic sizes of bacterial layer and solids hydrolyzed. Shimuzu et al. (10) have reported that after ultrasonic lysis of the biopolymers of waste sludge, the hydrolysis constant of the first order kinetics  $k_H$  increases from  $0.16 \text{ day}^{-1}$  to  $1.2 \text{ day}^{-1}$ . It is evident that the characteristic size of suspended solids decreases, but the contact area between particulate organic material and bacteria increases substantially in this case.

Obviously, the two-phase model (3) that takes into account a colonization phase describes a biomass washing phenomenon for a continuous-flow system if

$$\theta < \theta_{cr} = 1/(Y \cdot v_{mH} - k_d) \quad (12)$$

The simplest equation, 1 is unable to describe the biomass washing. It gives lower concentration of effluent suspended solids at low  $\theta$ . The  $n$ -order kinetics fits well experimental data of sludge and cellulose mesophylic digestion over wide range of SRT (Fig. 3).

It is easy to see that at  $n = 1$  a piece-wise (discrete) function (4) has the same limiting cases as the continuous Contois function (2). The ratio between the solids and biomass concentrations  $\xi = X/B$  regulates a degradation rate, which is proportional to the biomass concentration  $B$  at high  $\xi$  and to the biodegradable solids concentration  $X$  at low  $\xi$ .

A generalized continuous function for the model (3) may be written in the form

$$v_H = v_{max} \cdot \sigma(B) \cdot A(X_F, X) \quad (13)$$

where  $\sigma(B)$  is the fraction of the surface occupied by bacteria or released enzymes as a function of the concentration of hydrolytic bacteria,  $A(X_F, X)$  is the relative surface area subjected to degradation as a function of the influent and effluent solids concentrations.

Assuming the Langmuir function for an adsorption kinetics of hydrolytic biomass and the Monod function for a surface degradation kinetics, the function (13) can be written as

$$v_H = v_{max} \cdot \frac{\lambda B}{1 + \lambda B} \cdot \frac{X}{K_X + X} \quad (14)$$

where  $\lambda$  is the equilibrium constant equal to the ratio between the adsorption and desorption rates constants.

### Balance Between Hydrolysis and Methanogenesis

If the hydrolysis step is rate limiting, using the first-order equation (1) for a batch process, the degradable suspended solids concentration  $X$  and the methane volume released  $Q$  are given as

$$X = X_0 \cdot e^{-k_H t} \quad (15)$$

$$Q = M_{Ym} \cdot X_0 \cdot (1 - e^{-k_H t}) \quad (16)$$

where  $M_{Ym}$  is the methane to substrate conversion factor, which takes into account substrate converted to biogas;  $X_0$  is the initial concentration of degradable organic matter. The values of  $M_{Ym}$  and  $k_H$  for different wastes can be found in the literature (14).

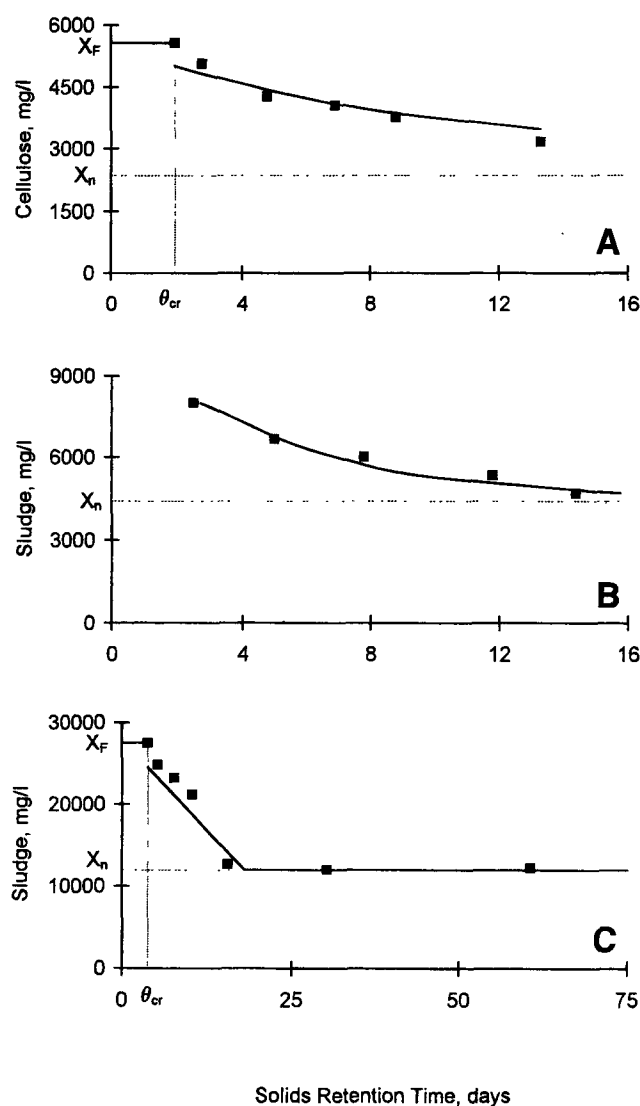


Fig. 3. Steady-state concentrations of cellulose (A) and sludge (B, C) with the  $n$ -order hydrolysis kinetics as a function of solids retention time:

- A)  $n = 0.5$ ,  $v_{mH} \cdot K = 0.19 \text{ day}^{-1}$ ,  $X_n = 2360 \text{ mg/L}$ ;
- B)  $n = 0.3$ ,  $v_{mH} \cdot K = 0.15 \text{ day}^{-1}$ ,  $X_n = 4400 \text{ mg/L}$ ;
- C)  $n = 0$ ,  $v_{mH} \cdot K = 0.06 \text{ day}^{-1}$ ,  $X_n = 12000 \text{ mg/L}$ ;

where  $X_n$  = nondegradable suspended solids concentration,  $\theta_{cr}$  = critical (biomass washing) value of solids retention time.

Symbols: experimental values; lines: model prediction. Experimental data are taken from refs. 11–13.

Table 1  
The Characteristic Values of Maximum Growth Rate Constants of Biomass  
and Hydrolysis Rate Constants Used in (Methane) Model

Process	Feed	Temperature	Figure	Constants <sup>1</sup>	Source of experimental data
Hydrolysis	Sludge	35°C	4	$k_H=0.25 \text{ day}^{-1}$ (Eq. 1)	5
	Cellulose	28°C	5	$k_H=0.15 \text{ day}^{-1}$ (Eq. 1)	15
		15°C	5	$v_{mH}=0.45 \text{ g/g} \cdot \text{day}$ , $\tilde{K}_X=1.5 \text{ g/g}$ (Eq. 2)	15
		6°C	5	$v_{mH}=0.11 \text{ g/g} \cdot \text{day}$ , $\tilde{K}_X=1.8 \text{ g/g}$ (Eq. 2)	15
	Cattle manure	6°C	6	$k_H=0.13 \text{ day}^{-1}$ (Eq. 1)	17
Acidogenesis	Sludge	35°C	4	$5.0 \text{ day}^{-1}$	5
	Cellulose	28°C	5	$10.0 \text{ day}^{-1}$	15
		15°C	5	$4.0 \text{ day}^{-1}$	15
		6°C	5	$2.0 \text{ day}^{-1}$	15
	Cattle manure	6°C	6	$2.0 \text{ day}^{-1}$	17
Acetogenesis	Sludge	35°C	4	$0.8 \text{ day}^{-1}$	5
	Cellulose	28°C	5	$0.28 \text{ day}^{-1}$	15
		15°C	5	$0.12 \text{ day}^{-1}$	15
		6°C	5	$0.07 \text{ day}^{-1}$	15
	Cattle manure	6°C	6	$0.11 \text{ day}^{-1}$	17
Methanogenesis	Sludge	35°C	4	$0.5 \text{ day}^{-1}$ (A) $2.0 \text{ day}^{-1}$ (H)	5
	Cellulose	28°C	5	$0.2 \text{ day}^{-1}$ (A) $0.45 \text{ day}^{-1}$ (H)	15
		15°C	5	$0.096 \text{ day}^{-1}$ (A) $0.18 \text{ day}^{-1}$ (H)	15
		6°C	5	$0.048 \text{ day}^{-1}$ (A) $0.13 \text{ day}^{-1}$ (H)	15
	Cattle manure	6°C	6	$0.07 \text{ day}^{-1}$ (A) $0.28 \text{ day}^{-1}$ (H)	17

<sup>1</sup>A = acetate utilizing methanogens, H = hydrogens utilizing methanogens. For cattle manure the constants of the single pathway are presented.

The (METHANE) model was used to analyze the dynamics of sludge degradation after the sewage sludge feed of continuously fed labor digester was stopped (5). Some constants of the model for the case-studies simulated are summarized in Table 1. From Fig. 4 it can be concluded that



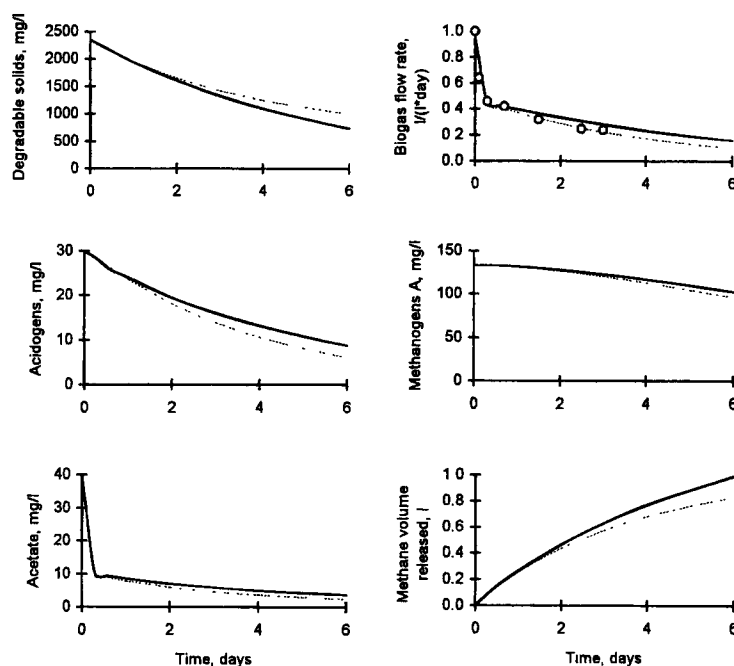


Fig. 4. Simulation of the sludge mesophilic digestion dynamics after stopping of sewage sludge feed of a continuously fed digester.

Symbols: experimental values; lines: model prediction. Experimental data are taken from ref. 5. — First order equation; - - - Contois equation; ○ Experimental data.

a high initial biomass concentration does not limit the process that can be described by the simple equations (15), (16).

During the colonization phase the hydrolytic bacteria cover the surface of the solids. The rate of this process is proportional to the concentration of hydrolytic biomass. Figure 5 shows one-year graphs of cellulose degradation at the different temperature with micro-organism inoculum taken from the sediments of highly polluted treatment pond (15). In this case the dynamics of released methane volume can not be described by the simple equation (16) because the low initial microorganism concentrations. The first-order equation (1) of hydrolysis kinetics was applied in the (METHANE) model for 28°C only and the Contois equation (2) was applied for 15°C and 6°C.

## Two Parallel Pathways of Degradation of Cattle Manure

It was mentioned above that the particle surface decreases at a constant depth per unit of time. However, availability of the particle surface for enzyme attack may change during the hydrolysis. Hobson (16) described the particle degradation of pig and cattle wastes by two growth rates in Monod equations for hydrolytic bacteria.

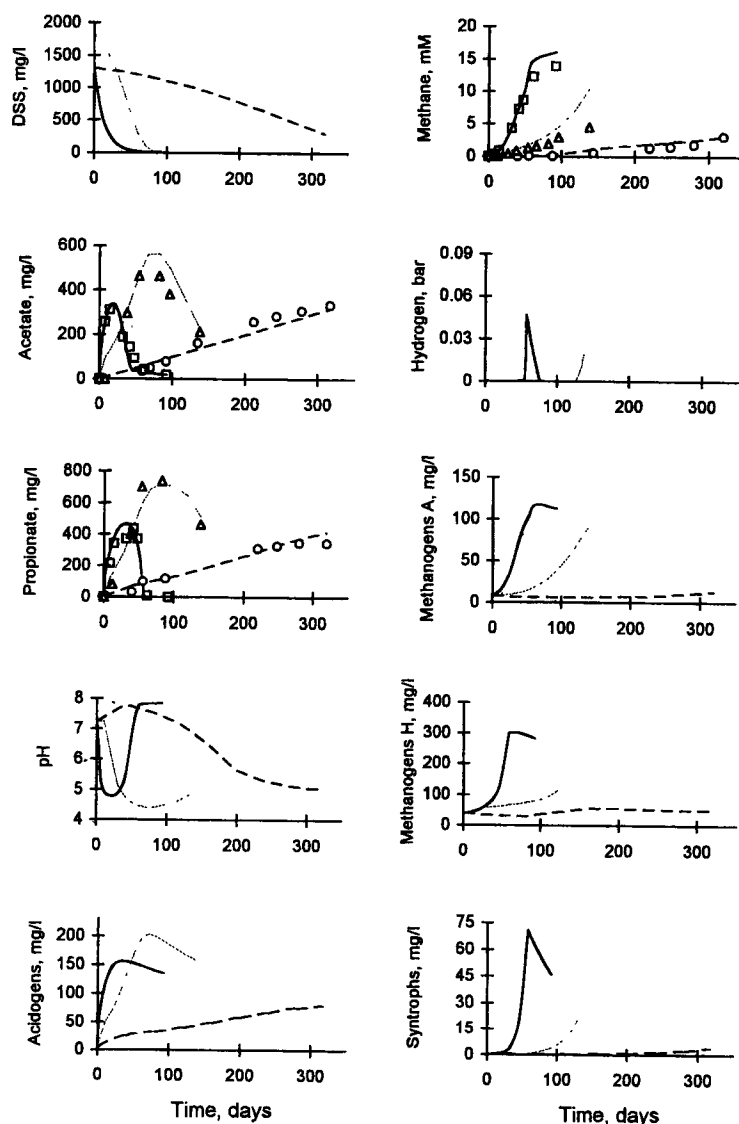


Fig. 5. Dynamics of anaerobic conversion of cellulose at different temperatures. Symbols: experimental values; lines: model prediction. Experimental data are taken from ref. 15. --- 6°C ○; ---- 15°C △; — 28°C □.

As result of a long-time (2.5 yr) selection, Kotsyurbenko et al. (17) have obtained an acclimated microbial consortium fermenting cattle manure at 6°C with methane production. Using this consortium as a seed material, a cattle manure degradation was studied at 6°C. No methane production was obtained during manure degradation without addition of inoculum and the final products were volatile fatty acids.

Figure 6 shows the results of simulations of the system with inoculum. It can be concluded that there are two parallel pathways of cattle

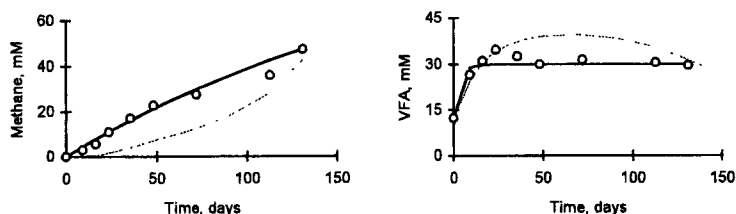


Fig. 6. Dynamics of anaerobic conversion of cattle manure at 6°C described by a single and two parallel pathways.

The first-order kinetics equation 16 was used for description of two parallel pathways of manure degradation:

$$\begin{aligned}
 k_H &= 0.005 \text{ day}^{-1}, M_{Ym} = 2.2 \text{ mM/mM}, X_0 = 45 \text{ mg/L} & (\text{CH}_4) \\
 k_H &= 0.03 \text{ day}^{-1}, M_{Ym} = 2.5 \text{ mM/mM}, X_0 = 7 \text{ mg/L} & (\text{VFA})
 \end{aligned}$$

where  $M_{Ym}$  = conversion factor of methane or VFA to particulate substrate. Some constants of (METHANE) model used for description of a single pathway are presented in Table 1.

Symbols: experimental values; lines: model prediction. Experimental data are taken from ref. 17. — Two pathways, - - - Single pathway, ○ Experimental data.

manure degradation with the final products of volatile fatty acids and a predominance of acetate (i) and methane (ii), produced without acetate consumption. The first pathway is conducted by micro-organisms presented in raw cattle manure and the second pathway is conducted by micro-organisms presented in inoculum. A much worse fitness of the model to experimental data was obtained assuming as usually the single pathway of solids degradation and methane production through acetotrophic and hydrogenotrophic methanogenesis. Some model constants are presented in Table 1.

## CONCLUSIONS

N-order kinetics with the rate constant dependable on a surface-to-volume ratio should be applied for description of solids degradation.

A delay of methane production during batch test happens usually because of the low initial biomass concentrations.

At low temperature the cattle manure is transformed into methane without acetate consumption.

## ACKNOWLEDGMENTS

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**NOMENCLATURE**

$B_i$	concentration of biomass of $i$ -th group of microorganisms
$B$	concentration of biomass of hydrolytic microorganisms
$\tilde{B}$	saturated hydrolytic biomass concentration covering the particle surface
$B_{di}$	concentration of dead biomass of $i$ -th group
$C$	carbohydrate fraction of organic matter
$d_X$	current diameter of particles
$d_{XF}$	input diameter of particles
$f$	relative contents of carbohydrates, proteins, lipids and undefined organic matter
$G_i$	partial pressure of gas components
$M_{Ym}$	methane to substrate conversion factor
$M_{BH}$	monomers of the biodegradable fraction of dead biomass
$M_{FH}$	monomers of the biodegradable fraction of suspended solids
$K$	dimensionless constant
$k_H$	kinetic rate constant
$k_d$	biomass decay-rate coefficient
$K_X$	half-saturation coefficient for the generalized function (14)
$\tilde{K}_X$	half-saturation coefficient for the Contois function (2)
$L$	lipid fraction of organic matter
$n$	degree coefficient
$P$	protein fraction of organic matter
$p$	relative content of biodegradable fraction of biopolymers
$Q$	methane volume released
$S_j$	concentration of dissolved substrate components
$S_1$	$DOS$ = concentration of dissolved carbohydrates, proteins and lipids
$t$	current time
$VB$	volatile cell matter
$VFA$	volatile fatty acids
$VX$	volatile suspended solids
$X$	$X_1 = DSS$ = concentration of biodegradable suspended solids
$X_2$	concentration of nonbiodegradable fraction of suspended solids and biomass
$X_3$	concentration of mineral fraction of suspended solids and biomass
$X_F$	concentration of influent biodegradable suspended solids
$X_0$	concentration of initial biodegradable suspended solids
$Y$	yield coefficient
$\delta$	depth of bacteria layer on particles

$\theta$	solids retention time
$\theta_{cr}$	critical (biomass washing) value of solids retention time
$\lambda$	equilibrium constant equal to the ratio between the adsorption and desorption rate constants
$\xi$	parameter equal to the ratio between biomass and solids concentration
$\rho_B$	bacterial density
$\rho_X$	solids density
$\sigma$	the fraction of the surface occupied by bacteria
$v_H$	rate of solids degradation
$v_{max}$	maximal rate of solids degradation
$v_{mH}$	maximal specific hydrolysis rate

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