MELiSSA

Memorandum of Understanding
ECT/FG/MMM/97.012

TECHNICAL NOTE 45.3

Modelling the liquefying compartment

Version 1
Issue 0

L. Poughon

September 2000

Document change log

Version	Issue	Date	Observations
0	0	25 July	Draft version
1	0	15 September	Final version 1

Content

Introduction

This technical note enlarges the studies started with TN 39.3 :"Liquefying compartment : analysis, stoichiometric and modelling approach". In this previous theoretical study, the basic equations of two models for the material balance analysis of the anaerobic digestion of organic matter were established. A first and simple dynamic model was also used and tested with some preliminary experimental results for the degradation of faeces in compartment I.

Since this work, the design and operating conditions of the liquefying compartment has changed (TN 41.x, 43.x), in order to improve its biodegradation efficiencies. A sequence of two fed-batch reactors (both for gas and liquid feed) is currently used.

The purpose of this technical note is to apply the mass balance analysis for the last past experiments carried out by EPAS (TN 43.2), using the material balance equations developed in previous TN 39.3.

In a second part a review of the models proposed in the literature for anaerobic digestion processes is presented. From this review, a model applicable to the liquefying compartment is proposed.

The methodology for the modelling of the first compartment is the same as the one which has been used for the other MELiSSA compartment.

I MELiSSA demonstration reactor (TN43.2) mass balance analysis

In the first part of this work, the material balance equations presented in TN 39.3, namely Hills Angelidaki and Hills Angelidaki Gottshalk (HAG) are used to analyse the experiments reported by Hermans and Demey (TN 43.2). These experiments are also used to check the consistency of the equations with the processes developed at EPAS.

I.1 Reactors design and operating conditions

The set-up of the anaerobic degradation process developed by EPAS for the "demonstration reactors" is summarised in table 1 and in figure 1.
The process itself is a series of 2 fed-batch bioreactors (figure 1) with:

- sequential feed and sampling;
- sequential flush of gas phase by N_2-gas;
- incubation with enzymes between reactor 1 and reactor 2.

The feed of reactor 1 is a faecal material the average composition of which is reported in Table 1.
The feed the reactor 2 is a diluted cake obtained from reactor 1. The cake is pre-treated by addition of enzyme and incubation during 2 days at 37°C and pH 5.1.

It must be outlined that the average compositions for the feed reported in table 1 vary in a quite large range. For faeces for example the dry matter content varies from 17.03 to 31.59 (i.e. 46%). This has a non negligible influence on the output of the reactors, which also have a large range of variation (Table 1). The most important variations at the output are for acetic acid and propionate, while the other VFA concentrations are more stable.

The composition of the feed for reactor 1 can be compared to the mean human faeces composition previously detailed (Table 2a - TN 39.3). A better modelling, a slightly modified faeces composition will be used in this study (Table2b); This composition is more consistent with the average feed composition reported for the experiments of TN 43.2. Except for the dilution factor of the faces feeding the reactor 1, the compositions, feed of reactor 1 (Table 1) and faeces compositions (Table 2b), are similar.

The mean feed composition used for reactor 2 is reported in Table 2c.

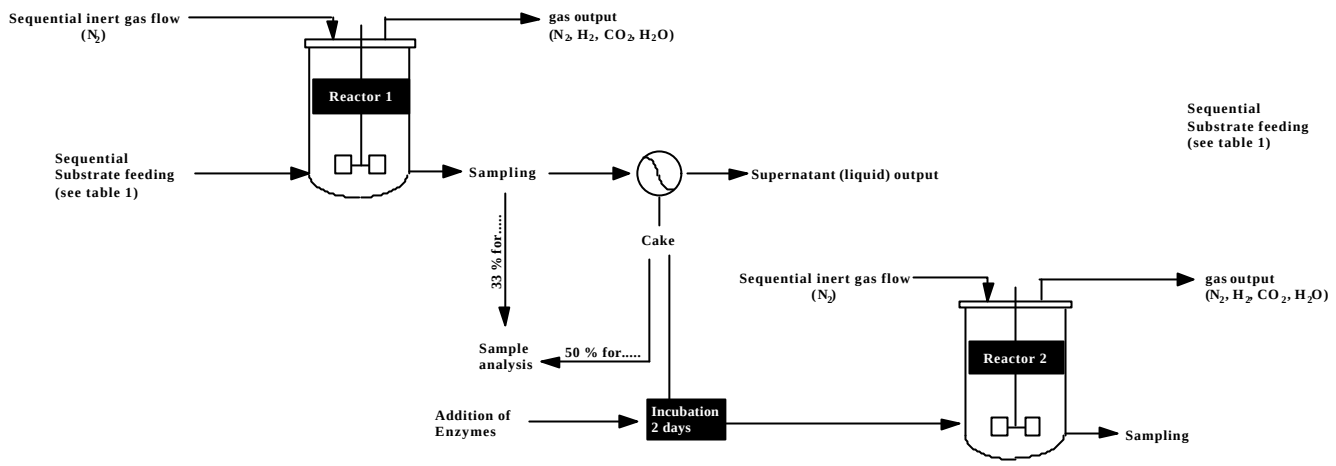

Figure 1 : Sequence of reactors for the demonstration of compartment 1

	Reactor 1		Reactor 2	
	Feed mean composition	**Output sampling composition**	**Feed mean composition**	**Output sampling composition**
pH	6.9	6.5 – 6.9	6.8	7.3 – 8.5
Dry matter (g/l)	23	**18** [7.54 – 26.49]	5.99	**4.9** [4.3 – 15.15]
Ash (g/l)	3.7	**3** [1.72 – 5.1]	0.76	**1.7** [1.5 – 3.2]
Total nitrogen (mg/l)	1241	**1250** [750 – 1970]	227	**300** [166 – 1735]
N-NH_3 (mg/l)	100	**700** [550 – 1400]	43	**90** [66 – 1075]
VFA (mg/l)	868	**2400** [588 – 3265]	210	**300** [48 –1400]
Acetic acid (mg/l)	354	**400** [0 – 1800]	47	**0 – 100** [0 – 630]
Propionic acid (mg/l)	218	**850** [312 – 1200]	66	**200** [47 – 1024]
Iso butyric acid (mg/l)	29	**240** [0 – 280]	24	**0** [0 – 55]
Butyric acid (mg/l)	167	**260** [0 – 346]	30	**0** [0 – 82]
Isovaleric acid (mg/l)	46	**510** [0 – 600]	43	**0** [0 –71]
Valeric acid (mg/l)	33	**30** [0 – 80]	-	**0** [0 – 17]
Caproic acid (mg/l)	20	**15** [0 – 32]	-	**0** [0 13.5]
Isocaproic acid (mg/l)	-	**0**	-	**0**
CO2 (mg)	0	**900** [500 – 1950] 562.8 mg/l (?)		?
CH4 (molar %)	0	**0***		**73** [51 – 79]
	Reactors set up			
PH	6.5		8	
Temperature	55°C		55°C	
Stirring	(Stirred)		(Stirred)	
Volume	1.6 litres		0.9	
Feeding rate	150 ml/2 days		150 ml/2 days	
Gas flushing rate	10 second / 6 hours		Each feed	
HRT	21 days		18 days	

Table 1 : Operating conditions for the demonstration reactors (From TN 43.2).
Output sampling on the reactor are given as an estimated mean value and in bracket the range of experimental values reported (TN 43.2).
* methanogenesis inhibited.

Note : CO_2 is given in mg. Assuming this is the net production, a value is calculated considering the liquid volume of the reactor in order to normalise the value with the other

	Wet % mass	Dry Matter % mass	Organic Matter %mass	VFA %mass
Water	**71**			
Dry matter	**29**	100		
Organic Matter (O.M.)	24.94	86	100	
Ash	4.06	14		
NH4+	0.17	0.5		
N tot	1.72	5.1		
N orga	1.55	4.6	5.35	
Proteins		30	34.88	
Carbohydrates			50.00	
Lipids			10.47	
VFA	1.35	4	4.65	
Biomass				
Acetate		2	2.33	50.00
Propionate		0.64	0.74	16.00
Iso-butyrate		0.1	0.12	2.50
Butyrate		0.732	0.85	18.30
Isovalerate		0.092	0.11	2.30
Valerate		0.196	0.23	4.90
Isocaproate		0.028	0.03	0.70
Caproate		0.168	0.20	4.20

Table 2a : Mean human detailed composition (TN 39.3).
The dry matter is defined as : DM = organic matter + Ash
The organic matter contains biomass, proteins, lipids, carbohydrates and VFA
The Ash includes free NH_3.

	Wet % mass	Dry Matter % mass	Organic Matter %mass	VFA %mass
Water	**73**			
Dry matter	**27**	100		
Organic Matter (O.M.)	23.2	86	100	
Ash	3.8	14		
NH4+	0.135	0.5		
N tot	1.377	5.1		
N orga	1.242	4.6	5.35	
Proteins	8.1	30	34.88	
Carbohydrates			50.00	
Lipids			10.47	
VFA	0.864	3.2	3.72	
Biomass				
Acetate		1.30	1.51	40.78
Propionate		0.80	0.93	25.11
Iso-butyrate		0.10	0.12	3.341
Butyrate		0.61	0.71	19.23
Isovalerate		0.16	0.19	5.299
Valerate		0.12	0.14	3.801
Isocaproate		0	0	0
Caproate		0.07	0.08	2.304

Table 2b : Mean human detailed composition used to feed reactor 1.

	Wet % mass	Dry Matter % mass	Organic Matter %mass	VFA %mass
Water	**94.1**			
Dry matter	-	100		
Organic Matter (O.M.)	5.9	88.59	100	
Ash	0.76	11.41		
NH4+	0.043	0.64		
N tot	0.227	3.41		
N orga	0.184	2.76	3.71	
Proteins	1.1	16.59	22.34	
Carbohydrates	3.63	54.50	73.41	
Lipids			**0**	
VFA	0.21	3.15	4.24	
Biomass	0	0	0	
Acetate	0.047	0.70	0.80	*18.8%*
Propionate	0.066	0.99	1.12	*26.3%*
Iso-butyrate	0.024	0.36	0.40	*9.6%*
Butyrate	0.03	0.45	0.51	*12.0%*
Isovalerate	0.043	0.64	0.72	*17.2%*
Valerate		0	0	0
Isocaproate		0	0	0
Caproate		0	0	0

Table 2c : Mean feed composition used for reactor 2.

I.2 Mass balance models (TN 39.3)

In the previous technical note 39.3, the pathways of the anaerobic degradation of organic were reviewed and two mass balanced models were detailed for the anaerobic digestion of organic matter. These models will be used to analyse the results obtained for the demonstration reactors presented above.

I.2.1 Mass balance analysis with the Hills Angelidaki based mass balance model

The principle of the model of Hills Angelidaki is presented in Figure 2 and the set of the stoichiometric equations describing the anaerobic degradation of organic matter is reported in Table 3. The first step ("enzymatic hydrolysis") can be associated to the hydrolysis of proteins in the compartment, while the second step ("acidogenesis") can be associated to the "carbohydrate or fibre" degradation.

The compounds involved in these 6 equations structured model are insoluble organic matter, soluble organic matter, ammonia, biomass, acetic acid, propionic acid, butyric acid, molecular hydrogen, CO_2 and methane. The stoichiometric equations take into account both anabolic and catabolic reactions and then to fit experiments with this model its required only to fix the yield for a key substrate in each of the 6 equations.

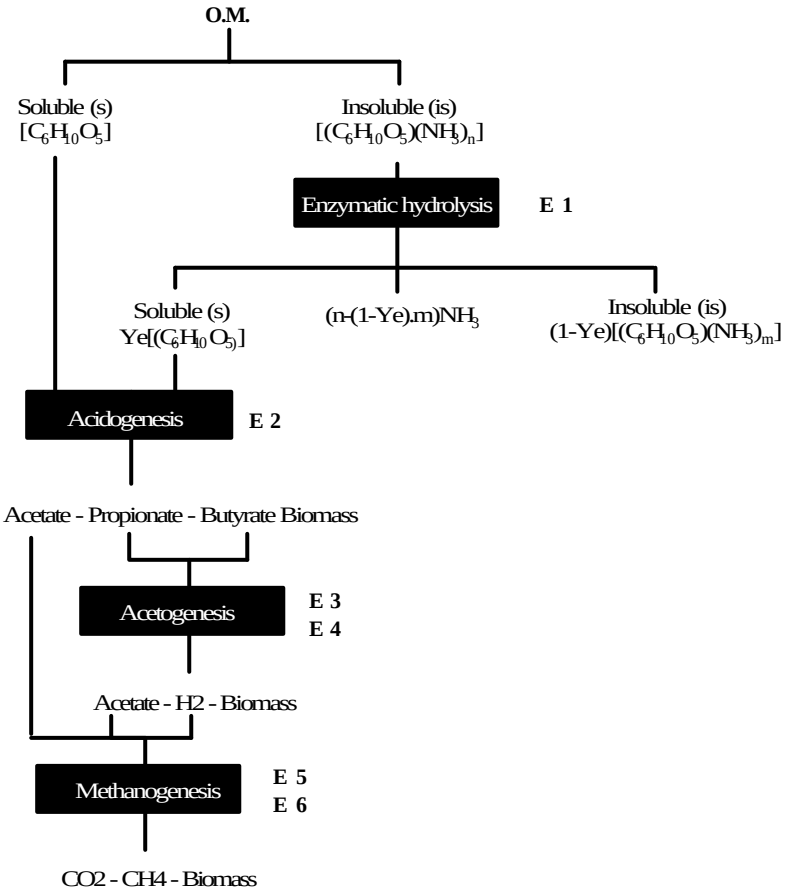

Figure 2 : Structure of the Hills Angelidaki model

The problem using this model is in the definition of the elemental composition of the organic matter degraded : $[C_6H_{10}O_6.nNH_3]_{is}$. This formula can not fit the elemental composition of the faeces (+ plant waste + biomass waste) feeding the compartment. The only solution is to consider a variant for the hydrolysis equation (equation E1b instead of equation E 1). But the CHONSP formula of the non hydrolysed part will not fit the composition of an existing biological compound.

Hydrolysis	$[C_6H_{10}O_6.nNH_3]_{is} \longrightarrow Ye[C_6H_{10}O_5]_s + (1-Ye)[C_6H_{10}O_5.mNH_3] + (n-(1-Ye).m)NH_3$	[E 1]
	$[C_aH_bO_b.nNH_3]_{is} \longrightarrow Ye[C_6H_{10}O_5]_s + (1-Ye)[C_{a'}H_{b'}O_{c'}.mNH_3] + (n-(1-Ye).m)NH_3$	[E 1b]
Acidogenesis	$[C_6H_{10}O5]s + 0.1115NH_3 \longrightarrow 0.1115C_5H_7O_2N + 0.744Acetate + 0.5Propionate + 0.4409Butyrate + 0.6909CO_2 + 0.0254H_2O$	[E 2]
Acetogenesis	$Propionate + 0.0458NH_3 + 1.764H_2O \longrightarrow 0.0458C_5H_7O_2N + 0.9345Acetate + 0.902CO_2 + 2.804H_2$	[E 3]
	$Butyrate + 0.0544NH_3 + 0.0544CO2 + 1.7818H_2O \longrightarrow 0.0544C_5H_7O_2N + 1.8909Acetate + 1.8909H_2$	[E 4]
Methanogenesis	$0.2644CO_2 + H_2 + 0.0058NH_3 \longrightarrow 0.0058C_5H_7O_2N + 0.5171H_2O + 0.2355CH_4$	[E 5]
	$Acetate + 0.022NH_3 \longrightarrow 0.022C_5H_7O_2N + 0.945CH_4 + 0.945CO_2 + 0.066H_2O$	[E 6]

Table 3 : Mass balanced equations of the Angelidaki model. It must be noticed that Angelidaki coupled directly H_2 producing and H_2 consuming reactions (Interspecies Hydrogen transfer).

I.2.1.1 Reactor 1

On the basis of the experimental observations reported in TN43.2:

Protein conversion efficiency : 60%
Fibre conversion efficiency : 29%
Total conversion efficiency : 40%

the yields reported in Table 4.1 were used to establish the mass balance analysis of reactor 1. The results obtained by coupling the 6 equations (Table 3) using these yields (Table 4.1) are reported in table 4.2.

The output concentrations calculated are close to the experimental ones (Table 1) for NH_3 and CO_2 (assuming the calculated value of 562 mg/l for CO_2). But the VFA production and the dry matter predicted by the Hills – Angelidaki model are higher than those reported experimentally. The acetate produced is higher than the average value reported experimentaly but is within the (large) range of the value reported (Table 1)

The dry matter calculated is 4 to 5g/l higher than the experimental one. This can result from our method used for the calculation of the dry matter :

Dry matter = Organic matter + ash
Organic Matter = Biomass + VFA + Proteins + Carbohydrates + Lipids

Moreover, it must be noticed that if equations E 3 to E 6 are inhibited (no H_2 production nor methane production), the ratio acetic acid/butyric acid/propionic acid is fixed by the stoichiometric equation E 2 at 1/**0.6**/0.67, while the experiments give a ratio of1/**2.1**/0.65. This suppose that the degradation is oriented toward propionate fermentation rather than to an acetate fermentation. This is perhaps a consequence of the inhibition of acetogenesis and methanogenesis.

E 1	E 2	E 3	E 4	E 5	E 6
Insoluble matter	Soluble matter	Propionic acid	Butyric acid	H_2	Acetate
Ye = 0.6 m=n	**0.25**	0	0	0	0

Table 4.1 : Yields for the coupling of equations reported in Table 3. Each yield characterises the assimilation or degradation of the key component available in the medium.

	Input	Output
Dry matter (g/l)	23	**23.3**
OM (biomass+VFA+protein +carbohydrates+lipids)	19.2	18.9
Ash (g/l)	3.8	4.4
Total nitrogen (mg/l)	1190	1190
$N\text{-}NH_3$ (mg/l)	78.7	721
VFA (mg/l)	710.7	**3048**
Acetic acid (mg/l)	290	**1156**
Propionic acid (mg/l)	178.7	896
Iso butyric acid (mg/l)	0	
Butyric acid (mg/l)	136.9	**890**
Isovaleric acid (mg/l)	0	0
Valeric acid (mg/l)	0	0
Caproic acid (mg/l)	0	0
Isocaproic acid (mg/l)	0	0
CO_2 (mg/l)	0	595
CH_4 (%)	0	0

Table 4.2 : Mass balance on reactor 1 calculated using equations of Table 3 and yields of Table 4.1.

I.2.1.2 Reactor 2

On the basis of the experimental observations reported in TN43.2:

Protein conversion efficiency : 40%
Fibre conversion efficiency : 60%
Total conversion efficiency : 29%

the yields reported in Table 5.1 were used to establish the mass balance analysis of the reactor 1. The results obtained by coupling the 6 equations (Table 3) using these yields (Table 5.1) are reported in table 5.2.

The concentrations calculated using the model are in agreement with the experimental values reported for N-NH3 and the VFA. But for gas composition, the methane molar ratio is lower than the experimental one. The problem of the dry matter at the output is the same as for reactor 1.

For the methane ratio in the gas, there is two possibilities :

- the first supposes that the model is inaccurate and that there is a higher production rate of CH4. The only mean to increase the CH_4 molar ratio is to produce methane through reaction E5 (methane from H_2). This supposes also that it exists H_2 is produced from VFA degradation or assimilation without, or with small CO_2 production.
- the second possibility is that a non negligible part of the CO_2 produced remains in the liquid phase under the ionic forms of carbonate and bicarbonate. This will then increases the part of methane into the gas. Such an assumption supposes that about 50% of the CO_2 formed remains in the liquid phase under ionic form, what is realistic as the reactor operates at pH 8. This option could be verify experimentally by measuring the total dissolved CO_2 and the gas CO_2.

E 1	E 2	E 3	E 4	E 5	E 6
Insoluble matter	Soluble matter	Propionic acid	Butyric acid	H_2	Acetate
Ye = 0.4 m=n	**0.4**	0.1	1	1	1

Table 5.1 : Yields for the coupling of equations reported in table 3. Each yield characterises the assimilation or degradation of the key component available in the medium.

	Input	Output
Dry matter (g/l)	5.99	5.52
OM (biomass+VFA+protein +carbohydrates)	5.23	4.7
Ash (g/l)	0.76	0.82
Total nitrogen (mg/l)	255.4	225.2
$N\text{-}NH_3$ (mg/l)	31.9	96.06
VFA (mg/l)	213.7	231.6
Acetic acid (mg/l)	47.83	0
Propionic acid (mg/l)	67.16	231.6
Iso-butyric acid (mg/l)	0	
Butyric acid (mg/l)	30.53	0
Isovaleric acid (mg/l)	0	
Valeric acid (mg/l)	0	
Caproic acid (mg/l)	0	
Isocaproic acid (mg/l)	0	0
CO_2 (mg/l)	0	511.29
CH_4 (molar %)	0	48.3%

Table 5.2 : Mass balance on reactor 2 calculated using equations of Table 3 and yields of Table 5.1.

I.2.2 Mass balance analysis with the Hills Angelidaki Gottschalk (HAG) based mass balance model

The second model proposed in TN 39.3, is a more structured model than the "classical" Hills Angelidaki model. It involves more stoichiometric equations (17 stoichiometric equations which can be coupled to 2 different equations for the growth of micro-organisms).

The principle of the chain of reactions is reported in Figure 3 and the equations are listed in Table 6.1.

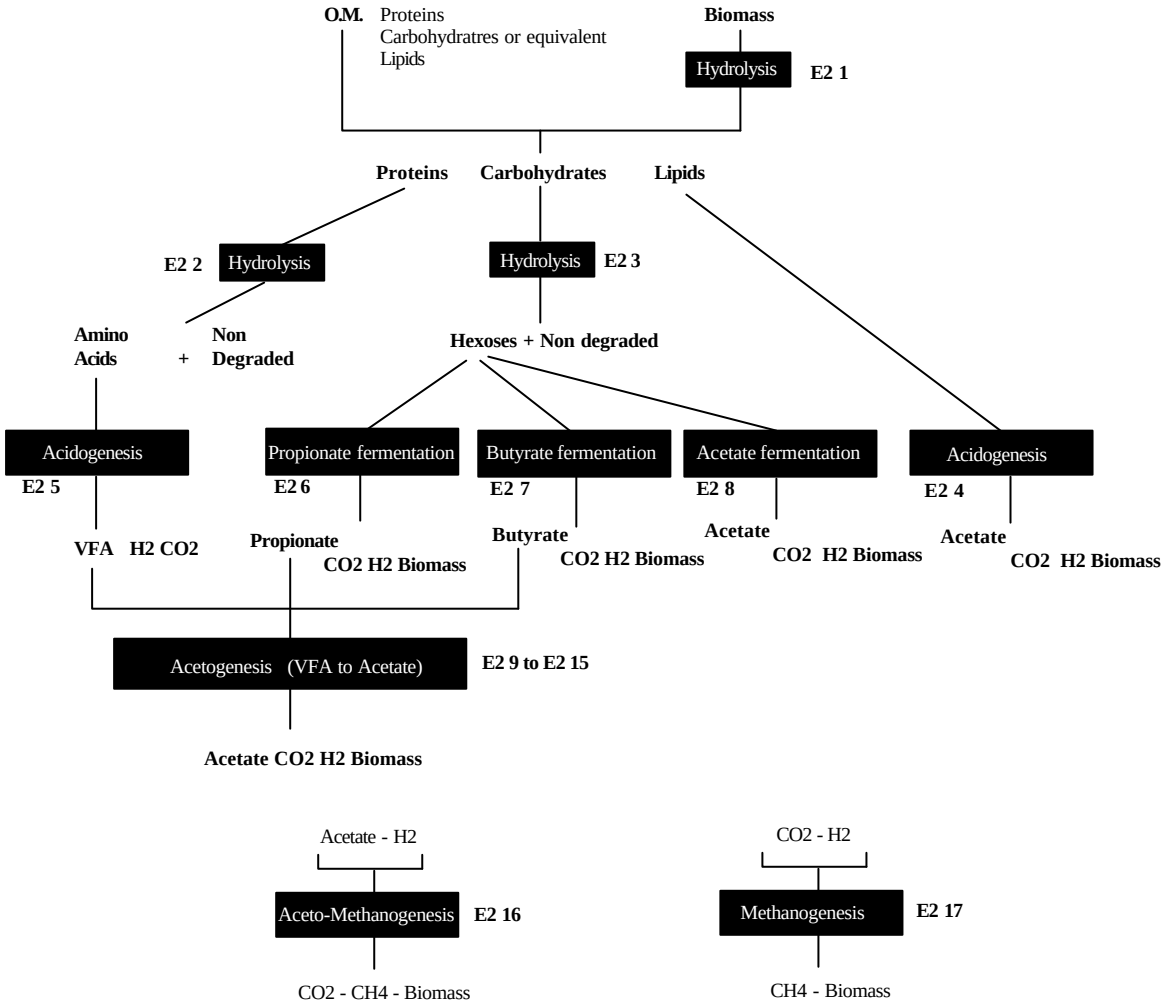

Figure 3 : Structure of the HAG model

Reaction	Equation number
Biomass hydrolysis into proteins, carbohydrates and lipids	E2 1
Proteins hydrolysis into amino-acids	E2 2
Carbohydrate hydrolysis into simple hexose	E2 3
Acidogenesis of lipids	E2 4
Acidogenesis of amino acids assuming biomass yield of 0.01 mole acetate/ mole biomass*	E2 5
Acidogenesis of hexose into propionate assuming biomass yield of 0. 1 mole hexose/ mole biomass*	E2 6
Acidogenesis of hexose into butyrate assuming biomass yield of 0. 1 mole hexose/ mole biomass*	E2 7
Acidogenesis of hexose into acetate assuming biomass yield of 0. 1 mole hexose/ mole biomass*	E2 8
Acetogenesis of VFA assuming biomass yield of 0. 05 mole acetate/ mole biomass*	E2 9 ; E2 10 ; E2 11 ; E2 12 ; E2 13 ; E2 14 E2 15
Aceto-methanogenesis	E2 16
Methanogenesis	E2 17

Table 6.1 : Set of equation of the HAG model.
*Biomass formula is $C_5H_7O_2N$ and hexose formula is $C_6H_{12}O_6$.
The stoichiometric equations are listed in Table 6.2.

	Step 1: hydrolysis of organic matter	**Kinetics**
	$\text{Urea} + H_2O \longrightarrow CO_2 + 2\,NH_3$	% degradation
E2 2	$[CHONS]_{proteins} + H_2O \longrightarrow [CHONP]_{poolAA}$	% degradation
E2 3	$[CHONS]_{carbohydrates} + H_2O \longrightarrow [CHONP]_{oses}$	% degradation
	$[CHONS]_{fibres} + H_2O \longrightarrow [CHONP]$	% degradation

	Step 2: acidogenesis (assimilation)	
E2 5	$[CHONS]_{poolAA} + 0.3484\,H_2O$ $\Downarrow$ $0.0241[CHONSP]_{Bio\_1} + 0.16373\,CH_3COOH + 0.00612\,C_2H_5COOH$ $+ 0.0039\,C_3H_7COOH + 0.01514\,C_3H_7COOH_{[isobutyrate]}$ $+ 0.0075\,C_4H_9COOH + 0.01016\,C_4H_9COOH_{[isovalerate]}$ $+ 0.01734\,C_5H_{11}COOH_{[isocaproate]}$ $+ 0.11816\,CO_2 + 0.1074\,H_2 + 0.2489NH_3 + 0.00635\,H_2SO_4$	Kinetic law
E2 6	$1.0833\,[CHO]_{oses} + 0.1\,NH_3 \longrightarrow 0.1\,[CHONSP]_{Bio\_2} + 0.6667CH3COOH + 0.6667\,CO_2 + 0.9\,H_2O$	Kinetic law
E2 7	$1.0833\,[CHO]_{oses} + 0.1\,NH_3 \longrightarrow 0.1\,[CHONSP]_{Bio\_3} + C3H7COOH + 2\,CO_2 + 2\,H_2 + 0.3\,H_2O$	Kinetic law
E2 8	$1.0833\,[CHO]_{oses} + 0.1\,NH_3 \longrightarrow 0.1\,[CHONSP]_{Bio\_4} + 3\,CH3COOH + 0.3\,H_2O$	Kinetic law
E2 4	$[CHON]_{lipids} + 0.1\,NH_3 + 0.575\,H_2O \longrightarrow 0.1\,[CHONSP]_{Bio\_5} + 0.875\,H_2 + 0.5\,CH_3COOH$	Kinetic law

	Step 3: acetogenesis	
E2 9	$C_2H_5COOH + 1.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_6} + CO_2 + 3\,H_2 + 0.875\,CH_3COOH$	Kinetic law
E2 10	$C_3H_7COOH + 1.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_7} + 2\,H_2 + 1.875\,CH_3COOH$	Kinetic law
E2 11	$C_3H_7COOH_{[Isoburate]} + 3.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_8} + 2\,CO_2 + 6\,H_2 + 0.875\,CH_3COOH$	Kinetic law
E2 12	$C_4H_9COOH + 3.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_9} + CO_2 + 5\,H_2 + 1.875\,CH_3COOH$	Kinetic law
E2 13	$C_4H_9COOH_{[Isovalerate]} + 3.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_10} + CO_2 + 5\,H_2 + 1.875\,CH_3COOH$	Kinetic law
E2 14	$C_5H_{11}COOH + 3.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_11} + 4\,H_2 + 2.875\,CH_3COOH$	Kinetic law
E2 15	$C_5H_{11}COOH_{[Isocaproate]} + 5.85\,H_2O + 0.05\,NH_3 \longrightarrow 0.05\,[CHONSP]_{Bio\_12} + 2\,CO_2 + 8\,H_2 + 1.875\,CH_3COOH$	Kinetic law

	Step 4: methanogenesis	
E2 16	$CH_3COOH + 0.022\,NH_3 \longrightarrow 0.022\,[CHONSP]_{Bio\_13} + 0.945\,CO_2 + 0.945\,CH_4 + 0.066\,H_2O$	Kinetic law
E2 17	$0.5\,CO_2 + 1.8909\,H_2 + 0.0109\,NH_3 \longrightarrow 0.0109\,[CHONSP]_{Bio\_14} + 0.4452\,CH_4 + 0.978\,H_2O$	Kinetic law

Table 6.2: Equations for the HAG model.

$[CHONSP]_{proteins}=CH_{1.56828}O_{0.3063}N_{0.2693}S_{0.00635}$ $[CHONSP]_{poolAA}=CH_{1.9800}O_{0.5122}N_{0.2693}S_{0.00635}$
$[CHONSP]_{Carbohydrates}=CH_{1.6667}O_{0.8333}$ $[CHONSP]_{Lipids}=CH_2O_{0.125}$
$[CHONSP]_{oses}=C_6H_{12}O_6$ $[CHONSP]_{Bio}=C_5H_7O_2N$

I.2.2.1 Reactor 1

The yields reported in Table 7.1 were used to establish the mass balance analysis of the reactor 1 with the HAG model. The results obtained by coupling the 17 equations (Table 6) using these yields (Table 7.1) are reported in table 7.2.
The biomass of the feed is assumed to be completely hydrolysed (E2 1 =1), then the OM in the reactor is composed of proteins, carbohydrates and lipids only.

It can be outlined that the degradation of proteins and amino acids using this model is sufficient to represent the VFA composition , except acetic acid, measured at the output of the reactor (Table 7.2). The model is based on 60% of degradation for proteins and only 20% for carbohydrates and 0% for lipids, what is lower than the values used in the previous model and lower than the values calculated by Demey et al. (TN 43.2). As a consequence, the remaining carbohydrate+lipids are equals to 51g for an initial dry matter of 100g, what can be compared to the 46g calculated by Demey et al. (TN43.2).

E2 1	E2 2	E2 3	E2 4	E2 5	E2 6	E2 7	E2 8	E2 9
1	0.6	0.2	0	1	0	0	1	0
E2 10	E2 11	E2 12	E 13	E2 14	E2 15	E2 16	E2 17	
0	0	0	0	0	0	0	0	

Table 7.1 : Yields for the coupling of equations reported in Table 6.

	Input	Output
Dry matter (g/l)	23	**23.5**
OM (biomass+VFA+protein+carbohydrates+lipids)	19.2	**18.8**
Ash (g/l)	3.8	4.4
Biomass g for 100g feed (before hydrolysis)	*33*	*5.4*
Proteins g for 100g feed [after biomass hydrolysis]	*10 [33]*	*13*
Carbohydrates +lipids [after biomass hydrolysis]	*53 [61]*	*51*
Total nitrogen (mg/l)	1189.8	1189.8
$N\text{-}NH_3$ (mg/l)	78.7	692.0
VFA (mg/l)	710.7	**6002.0**
Acetic acid (mg/l)	290.2	**3867.**
Propionic acid (mg/l)	178.7	954.3
Isobutyric acid (mg/l)	23.8	251.7
Butyric acid (mg/l)	136.9	195.1
Isovaleric acid (mg/l)	37.7	215.0
Valeric acid (mg/l)	27.1	158.3
Caproic acid (mg/l)	16.4	16.4
Isocaproic acid (mg/l)	0	344.2
CO_2 (mg/l)	0	**889**
CH_4 (%)	0	0

Table 7.2 : Mass balance on reactor 1 calculated using equations of Table 6 and yields of Table 7.1.

The production of acetate predicted by the mass balanced model is higher (3867 mg/l) than the measured one (400 mg/l). The acetate can be decreased if we assumed that it is metabolised into CO_2 or assimilated into biomass. In any case these solutions will lead to increase the CO_2 production and decrease free NH_3.
On the model point of view, the high production of acetate and CO_2 are directly the consequence of amino acids degradation, and then this production can not be reduced without reducing the degradation yield of proteins. It can then be wondered if the equation for the degradation of amino acids is correct. The response is yes as this equation is consistent with the equation proposed by Angelidaki et al. (1999) for the degradation of gelatin by acidogenic bacteria :

HAG model : amino acid degradation	$[CHONS]_{poolAA} + 0.3484\,H_2O \Downarrow 0.0241[CHONSP]_{Bio\_1} + 0.16373\,CH_3COOH + 0.00612\,C_2H_5COOH + 0.0039\,C_3H_7COOH + 0.01514\,C_3H_7COOH_{[isobutyrate]} + 0.0075\,C_4H_9COOH + 0.01016\,C_4H_9COOH_{[isovalerate]} + 0.01734\,C_5H_{11}COOH_{[isocaproate]} + 0.11816\,CO_2 + 0.1074\,H_2 + 0.2489\,NH_3 + 0.00635\,H_2SO_4$
Angelidaki et al. (1999) : amino acid degradation	$[CHONS]_{poolAA} + 0.3006\,H_2O \Downarrow 0.017013[CHONSP]_{Bi} + 0.29742\,CH_3COOH + 0.02904\,C_2H_5COOH + 0.022826\,C_3H_7COOH + 0.013202\,C_4H_9COOH + 0.07527\,CO_2 + 0.001\,H_2S + 0.28298\,NH_3$

This problem doesn't appear with the previous model as the degradation of substrate is only an hydrolysis to ammonia, to"soluble carbohydrate" and to insoluble parts. The "soluble carbohydrate" being further degraded into VFA.
We can not explain here the discrepancy for acetate between the mass balance model and the experiments. It can be due to :

- the degradation of amino acids, which produced lesser amount of acetate and that in fact the proteins hydrolysis observed in the reactor is a release of ammonia without a degradation of the carbon chain;
- a part of acetate was not measured or another compound is formed. As there isn't a C mass balance in the experiment, we can not be sure that there is no loss in measurement.

It can be noted in this model that the degradation of lipids is set to 0 (no lipids degradation), and only a part of carbohydrates (probably the easiest degradable part) and proteins are degraded, what seems consistent with the experimental observations (there is a visually observable lipidic phase inside the reactor).

I.2.2.2 Reactor 2

The yields reported in Table 8.1 were used to establish the mass balance analysis of the reactor 2 with the HAG model. The results obtained by coupling the 17 equations (Table 6) using these yields (Table 8.1) are reported in Table 8.2.
As for reactor 1, the biomass of the feed is assumed to be completely hydrolysed (E2 1 =1), then the OM in the reactor is only composed of proteins and carbohydrate (or fibres). Lipids are not considered (Table 2c)

The results obtained with this model are in agreement to those obtained experimentally.

The number of parameters in the model gives a higher degree of freedom for the set of equations. Then for the propionate for different couples for the yields of reaction E2 9 and E2 5, similar results for the propionate production can be obtained.

E2 1	E2 2	E2 3	E2 4	E2 5	E2 6	E2 7	E2 8	E2 9
1	0.4	0.3	0	1	1	0	0	0.7
E2 10	E2 11	E2 12	E 13	E2 14	E2 15	E2 16	E2 17	
1	1	1	1	1	1	1	1	

Table 8.1 : Yields for the coupling of equations reported in Table 6 for reactor 2.

	Input	Output
Dry matter (g/l)	5.99	4.7
OM (biomass+VFA+protein+carbohydrates+lipids)		3.9
Ash (g/l)	0.76	0.81
Proteins g for 100g feed	*22*	*13.3 [i.e 8g from 60g DM]*
Carbohydrates (hexose polymer)	*73*	*51.3 [i.e 30.6 from 60g DM]*
Total nitrogen (mg/l)	255.4	224.7
$N\text{-}NH_3$ (mg/l)	31.9	80.71
VFA (mg/l)	217.7	241.41
Acetic acid (mg/l)	47.8	0.0
Propionic acid (mg/l)	67.2	241.4
Isobutyric acid (mg/l)	24.4	0.0
Butyric acid (mg/l)	30.5	0.0
Isovaleric acid (mg/l)	43.8	0.0
Valeric acid (mg/l)	0	0.0
Caproic acid (mg/l)	0	0.0
Isocaproic acid (mg/l)	0	0.0
CO2 (mg/l)	0	1128.51
CH4 (%)	0	51%

Table 8.2 : Mass balance on reactor 2 calculated using equations of Table 6 and yields of Table 8.1.

I.3 Conclusions of mass balance analysis and about mass balance models

Models

The two models can be used and give quite different results.
The first one (Hills-Angelidaki), can be used for both reactors. Nevertheless, it lacks of flexibility as it considers the substrate to degrade as a whole, and as the 3 VFA concentrations ratio is stoichiometrically fixed for the reactor.

The second model (HAG) works fine for the reactor 2 (complete degradation up to methanogenesis), but presents a discrepancy in the mass balance analysis of experiments of the reactor 1 as it leads to produce a high amount of acetate. The HAG model is more complete but requires to fix more parameters (more yields for mass balance analysis and more kinetic parameters for the dynamic modelling).

It can be noted that the last material balance model published in the literature tends to a structured representation of the anaerobic digestion reaction, in a way comparable to the HAG model (Table 9).

Reactor 1

For the mass balance analysis of reactor 1, the problems encountered concern :

- the dry matter at the output of the reactor (for the two models);
- the VFA (acetate) production (for HAG model).

The two values calculated are too high compared to the experimental measurements. As the dry matter includes the VFA, the two problems are linked.

In the case of the HAG model, it is difficult to understand the origin of the problem. Even if a part of the VFA is in the gas phase, this part is too small to explain the discrepancy between calculated and measured values. It is also difficult to consider that acetate is degraded into CO_2 as this will drastically increase the gas production.
The model is based on elements conservation and then at least C and N balances on experiments would probably help to understand the process and then to know if there is a measurement problem (C and N unbalanced) or if the model (i.e. the chain of reactions and equations) is not adapted.

Reactor 2

For the mass balance analysis of the reactor 2, the problems encountered concern :

- the dry matter at the output of the reactor (with Hills-Angelidaki model only);
- the methane ratio in the gas.

For the methane ratio, it is probable that a “buffer effect” is responsible of the apparent discrepancy between experimental and calculated values. At pH 7-8, ionic forms of CO_2 (carbonate, bicarbonate) can represent 7 time the dissolved form. Then the theoretical ratio of CH_4 in the gas of 50% can appear higher in experiments as a lot of the CO_2 produced stays in the liquid phase. For

example, considering a total amount of 1g CO_2, a liquid phase of 1L with pH 8 and a gas phase of 1L composed of 30% of CO_2, this lead to a distribution at thermodynamic equilibria of 55% of the total CO_2 (gas+liquid)in the liquid phase.

The HAG model represents quite well the mass balance of reactor 2. It can be noted that the feed is defined without lipids or fats. It can be wondered if the discrepancies observed with reactor 1 are not related to this compound and to an hydrophobic lipidic phase.

As a conclusion, the HAG model seems a better option for the mass balance analysis and constitute the basis for the overall model of the anaerobic compartment I. In its structure and in its principle, this model is close to the model detailed by Angelidaki et al. in 1999 for the bioconversion of complex substrates to biogas.

II Dynamic model of the compartment

Our purpose in this section is not to simulate the reactors of the liquefying compartment. After a review of the existing models (theoretical model and validated model), we will develop the basic equations and the structure of the future model for the anaerobic fed-batch digestors.
The dynamic model presented can either be used with the HAG or the Hills-Angelidaki material balance models previously described.

II.1 Review of models

There are 3 tendencies in anaerobic digestion modelling :

- unstructured and non segregated models (Hashimoto et al., 1981). These are the simplest models, but their simplicity often reduce their accuracy.
- unstructured segregated models (Hill, 1983), which involved several equations and more than one type of biomass (i.e. more than one species).
- structured segregated model, which involves more or less the metabolism of the organisms involved in the process (Angelidaki et al., 1999).

The models detailed in the first part of this technical note, and used in the mass balance analysis of reactors 1 and 2, can be classified as partially structured segregated models.

The biological models used for the description of the growth and of the degradation/assimilation/production of compounds are based on first order models (Garcia-Ochoa et al., 1999), Contois models or Monod models (Angelidaki et al., 1999).
Beside the large variety of models that can be found, there is also a large variety of reactors : continuous stirred tank reactor (CSTR), fixed bed reactors (FBR) or continuously expending reactors (CER) (Thomas and Nordstedt, 1999).

Selected models for anaerobic degradation are given in Table 9. They are a good overview of the kind of models that can be found in the literature. Usually, when used for the modelling of experiments, the models are consistent, as the parameters are often identified to fit the experiments.
The model presented by Angelidaki et al. (Table 9) is interesting for our purpose of modelling the first compartment, as its basis is close to the so-called "HAG" model, and as the biological kinetic used by Angelidaki are based on a Monod model, what is also the basis of the model, we want to establish for the anaerobic degradation in the first compartment. Then the values used by Angelidaki et al. (1999) can also be used for our model.

Reference	Vasiliev V.B, Vavilin V.A.,Rytow S.V. and Ponomarev A.V. "Simulation model of anaerobic digestion of organic matter by a micro-organism consortium : basic equations. Water Resources **20**. 6: 633-643 1993	Jeyaseelan S. "A simple mathematical model for anaerobic digestion process". Wat. Sci. Tech. 35. 8: 185-191. 1997.	Angelidaki I., Ellegaard L., Ahring B.K. "A comprehensive model of anaerobic bioconversion of complex substrates to biogas". Biotechnol. Bioeng. **63**: 363-372. 1999	Garcia-Ochoa F., Santos V.E., Naval L., Guardiola E., Lopez B. "Kinetic model for anaerobic digestion of livestock manure". Enzyme and Microbial Technology. **25**: 55-60. 1999
Number of stoichiometric equations	13 equations Complex chain of reaction and inter-relation between micro-organisms	No material balance equations	10 equations	6 Reactions – Not balanced
Number of different biomass type	7 species involved	2 groups of bacteria	8 groups of bacteria + 2 enzymatic steps	2 groups of bacteria
Number of parameters (biological yields and kinetics)	Not specified, but huge	16 parameters – Values estimated from literature review	89 parameters – values from previous studies and from literature	10 parameters – Identified from experiments
Model validation	Theoretical model (basic equations)	Theoretical model	Model validated on experiments (Lab-scale manure)	Model validated on experiments
Compounds considered (except biomass)	12 compounds Proteins ; Carbohydrates ; lipids Unknown compound Acetate ; Propionate NH_3 ; H_2O ; H_2SO_4 H_2, CH_4, H_2CO_3 (i.e. CO_2) 3 kind of suspended solid	5 compounds Proteins ; Carbohydrates ; lipids Organic acid CH_4	18 compounds Proteins and insoluble proteins ; aminoacids Lipids ; Glycerol trioleate ; Oleate Carbohydrates soluble and insoluble ; hexose Acetate ; Butyrate ; Propionate ; Valerate. CO_2 ; CH_4 ; NH_3 ; H_2 ;H_2S	5 compounds Unknown substrate Unknown degradable substrate Volatile acids CO_2 CH_4
Process	Continuous Stirred Tank Reactor Gas/liquid transfer and pH used	Continuous Stirred Tank Reactor	Continuous Stirred Tank Reactor Gas/liquid equilibria	Batch – fed-batch Reactor
Biological/Enzymatic kinetic	Function of temperature, pH, inhibition and limitation by substrates Limitation is based on a Moser equation	Monod equation with a biomass decay term	Monod equation with a biomass decay term and inhibition (Haldane and Non competitive models)	First order reactions and changes in kinetic rates expression (different growth phases)

Table 9 : Overview of the variety of models found in the literature

II.2 Basic equation of the dynamic model for the anaerobic reactors of compartment I

The set of equation presented here were first established in TN 39.3 and TN 39.1.
The biological kinetic equations were slightly changed here taking into account the parameters reported in some models described in the literature.
The hydrodynamic model was adapted to the operating conditions in which experiments are performed.

II.2.1 Enzymatic kinetic model

The models are usually first order reactions. Angelidaki et al. (1993) assumed that the enzymatic hydrolytic reaction of the organic substrate (first of the anaerobic reactions) is inhibited by VFA.

$$r_{Sk} = \lambda . \frac{Ki_{VFA}}{1 + Ki_{VFA}} . C_k$$

II.2.2 Biological kinetic model

The biological model used is based on Monod equations. The inhibition can be non competitive or can be describe by an Haldane equation.
The inhibitory compounds reported in models are ammonia, VFA and acetate.

Evolved models (Angelidaki et al., 1999 ; Vasiliev et al., 1993) take also into account the influence of temperature and pH on the maximum growth rate of the organisms.

The maximum growth rate for a micro-organism

$$\mu_{max}(T, pH) = \mu_{max}(T).F(pH)$$

For Angelidaki et al (1993)

$$\mu_{max}(T) = \mu_{max}(Topt) - \alpha(Topt - T) \qquad \text{if } T < Topt$$

$$\mu_{max}(T) = \mu_{max}(Topt).\frac{(T\,max - T)}{(T\,max - Topt)} \qquad \text{if } T > Topt$$

$$F(pH) = \frac{1 + 2.10^{0.5'pK_l - pK_h)}}{1 + 10^{(pH - pK_h)} + 10^{(pK_i - pH)}}$$

$\mu_{max}(Topt)$, Tmax , Topt , pKl and pKh being characteristic of a bacteria group

Biomass decay rate

Note : In the model of Vasiliev et al. (1993), the lysis of biomass participates to the definition of the substrate degraded

$$r_D = k_D . X$$

Growth rate for a microorganism

$$r_X = \mu.X - k_D.X$$

with non-competitive inhibition : $$\mu = \mu_{max}(T, pH).\prod_k \frac{C_k}{(Ks_k + C_k)}.\frac{1}{\left(1 + \frac{C_k}{Ki_k}\right)}$$

with Haldane equation : $$\mu = \mu_{max}(T, pH).\prod_k \frac{C_k}{\left(Ks_k + C_k + \frac{C_k^2}{Ki_k}\right)}$$

Substrate consumption/production rate

$$r_{Sk} = \frac{-1}{Y_{X/Sk}}\mu.X$$

The yields $Y_{X/Si}$ are calculated from the stoichiometric equations.

II.2.3 pH calculation

The prediction of the pH (or H^+ concentration) is difficult because of the presence of compounds for which the ionised form and the equilibria constant are unknown (proteins, carbohydrates, amino-acids pool, lipids). Nevertheless Angelidaki et al. (1993) have proposed an expression for the pH simulation:

$$Ch(pH) = [HCO_3^-] + 2.[CO_2^{2-}] + [Acetate^-] + [Propionate^-] + [Butyrate^-] + [H_3PO_4^-] + 2.[HPO_4^{2-}] - [NH_4^+] + [Anion^-] - [Cation^+]$$

$$[H^+] = \frac{Ch(pH) + \sqrt{Ch(pH)^2 + 4.Kw}}{2} \quad \text{for } Ch(pH) > 0$$

$$[H^+] = \frac{2.Kw}{-Ch(pH) + \sqrt{Ch(pH)^2 + 4.Kw}} \quad \text{for } Ch(pH) < 0$$

This expression takes into account the buffering effect of the medium. This calculation is performed using an iterative algorithm, as the concentration of each ionic compound depends on pH, the quantity of the compound (ionic and non ionic forms) and the acid base equilibria constant.The acid/base equilibria constants for all the compounds involved in the first compartment have been detailed in TN 17.1 and TN 23.1

II.2.4 Dynamic of the reactor

Design and operation of the anaerobic reactors in the first compartment

The following design (Figure 4) for the operation of the reactors of compartment I is based on the operating conditions described in TN 43.2 (Table 1).
The reactor is a fed-batch reactor for both gas and liquid.

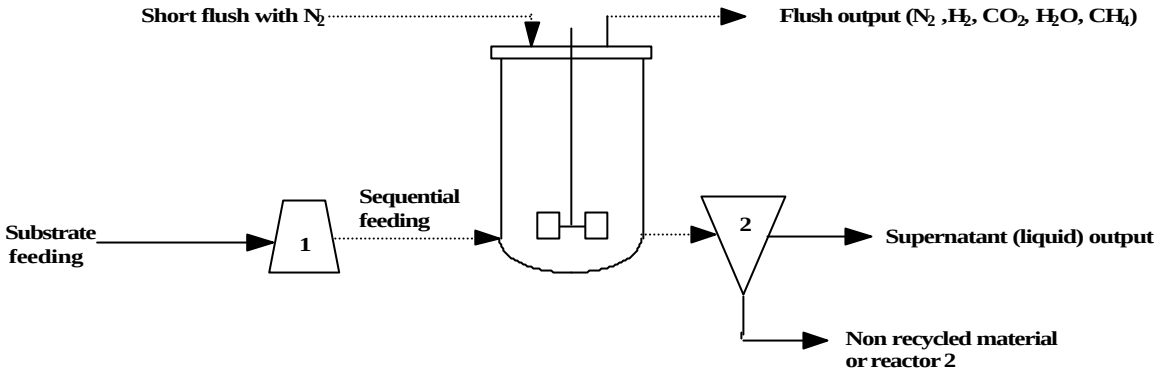

Figure 4: Liquefying fed-batch reactor.
[1] is a pre-treatment process (or post-treatment if there is recycling).
[2] is the separation process for recycling and/or liquid output of the reactor. It is evident that at least a value of the efficiency of these processes must be known for their integration in a dynamic model.

The gas flush is very rapid. If the flow rate of N_2 for the flush is sufficiency high, it can be assumed that the gas phase inside the reactor is completely replaced in the reactor by a N_2 gas phase.
Assuming that there is no variation in the liquid volume during the different phases of feeding/sampling, the gas volume can be considered as constant, then gaseous compounds produced by the anaerobic degradation (H_2O, CO_2, H_2, CH_4) increase the gas pressure inside the reactor. A normal gas pressure is restored at each flush.

Hydrodynamic equations for the fed-batch reactor (not including pre and post treatment)

Batch phase

Reaction rate for compound Si : $\phi_{Si}\big|_{Reac} = \sum r_{Si}$

Gas/Liquid transfer (Liquid → Gas) : $.\phi_{Si}\big|_{GL} = K_L a(C^*_{Si}\big|_L - C_{Si}\big|_L)$

Liquid phase $V_L . \dfrac{dC_{Si}\big|_L}{dt} = V_L .\phi_{Si}\big|_{GL} + V_L .\phi_{Si}\big|_{Reac}$

Gas phase $V_G . \dfrac{dC_{Si}\big|_G}{dt} = -V_L .\phi_{Si}\big|_{GL}$

Pressure variation (assuming perfect gases and gas volume constant): $\dfrac{dP}{dt} = 10^{-3} \sum_{Si} \dfrac{dC_{Si}\big|_G}{dt} .RT$

Liquid feeding phase

$$V_L.C_{Si}\Big|_{L\,AFTER\,SAMPLING} = V_{Feed}.C_{Si}^{in}\Big|_L - V_{Sampling}.C_{Si}\Big|_{L\,BEFORE\ SAMPLING}$$

In order to maintain the liquid volume V_L, the volume for feeding and sampling must be the same

In the case of differences between the volumes of feeding and sampling, the liquid and the gas volumes change : $V_{L\ AFTER\ SAMPLING} = V_{L\ BEFORE\ SAMPLING} - (F_{sampling} - V_{feed})$

Gas flush

Restoration of normal pressure : P=1 atm

All compounds (except N_2) set to 0.

The quantity (moles) of compound flushed is : $n_{Si} = C_{si}\Big|_{G\,BEFORE\ FLUSH}.V_G$.

Conclusion

The two material balance models were used to check the experiments reported for the reactors used in the liquefying compartment and described in TN 43.2.

For reactor 1, whatever model is used, a discrepancy concerning the VFA production, mainly acetate, is observed between the values predicted by the models and the experimental measurements. The problem is less important for the Hills-Angelidaki model than for the HAG model. As there is no experimental C,N balance available, it is difficult to determine the origin of this discrepancy. It can be either :

- an inadapted model (i.e. the degradation can not be represented by the equations established) ;
- some compounds are not measured and then not considered in the model
- all the acetate produced is not measured (perhaps it is partly dissolved in the lipidic phase)

For reactor 2, the two material balance models are quite equivalent. The HAG model gives a better prediction for the residual dry matter than the Hills-Angelidaki model. The problem of the ratio of methane in the gas phase is probably related to acid/base equilibria for carbon dioxide. This problem will be easily solved in dynamic models, where these equilibria are taken into account.

The basic equations for the dynamic model of an anaerobic digestor were established. These equations can be used for both the HAG and the Hills-Angelidaki model. Biological parameters for HAG model can be found in the literature (Angelidaki et al., 1999), where a similar model can be found.
Before dynamic simulations, it is necessary to solve the discrepancy observed when considering reactor 1, and then to known if the material balance model can be validated.
The dynamic model will be probably developed under Matlab instead of the building of a specific program in Fortran (as for NitriSim and PhotoSim), as this could be a better format for the exchange between partners (ADERSA, UAB).

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