A TOTAL CONVERTING AND BIOSAFE LIQUEFACTION COMPARTMENT FOR MELISSA

TECHNICAL NOTE: 86.1.2

Optimisation of the Methanogenesis proces

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1. Introduction

This technical note presents the current state of the MAP project “A Total Converting and Biosafe Liquefaction Compartment for MELISSA” on behalf of the Laboratory for Microbial Ecology and Technology at the University of Ghent.

This note describes the composition of the feed, the current operation conditions of the mesophilic anaerobic digester, the overall C, N, P and S mass balances for the digester and in addition a short summarize about additional physico-chemical treatments of the digester effluent. On the one hand, some results about the electrolysis of the digester effluent are shown. On the other hand, preliminary results of a thermal microwave treatment for the liquefaction of the digester particulates are given. An energy cost estimation for the mesophilic digester, electrolysis cell and microwave digestion method has been calculated. Finally, the actions to be taken in the nearby future concerning the closed loop experiments are described.

2. Objectives and task description

Further research has been mainly focussed on the elucidation of the mass flows in the anaerobic digester as mentioned in WP 1.200. Additionally, electrochemical oxidation experiments were conducted with the supernatant of the digester effluent. Similarly to the subcritical gasification method, digester particulates (mainly algae cells and straw particles) were liquefied by means of a thermal microwave treatment. Results are summarized below including an estimation of the energy consumption of the electrolysis- and microwave treatment, relative to the treated fractions with respect to the overall mass balances of the MELiSSA loop.

3. Materials and Methods

3.1 Experimental set-up

A 10 Liter anaerobic PVC-reactor is used for the anaerobic digestion of the defined feed. As indicated in Figure 1, the digester is maintained at a constant temperature of 34°C by placing it in an incubator. The reactor is a CSTR-type (continuously stirred tank reactor) and is shaken two minutes/hour on a shaker platform (INNOVA shaker) at a constant 90 rpm.

The feeding of the reactor is fed-batch wise at regular time intervals. When the reactor is at the feed-mode, valve 2 (biogas outlet) is closed while valves 1 and 3 are opened. By means of a peristaltic pump, the feed is pumped in at a rate of 200ml/min. The feeding causes a replacement of a part of the liquid present in the reactor by the freshly added feed, resulting in a release of the effluent through the effluent pipe

(valve 3). Consequently, for each volume of the feed fed to the reactor, a same volume of effluent is withdrawn simultaneously.

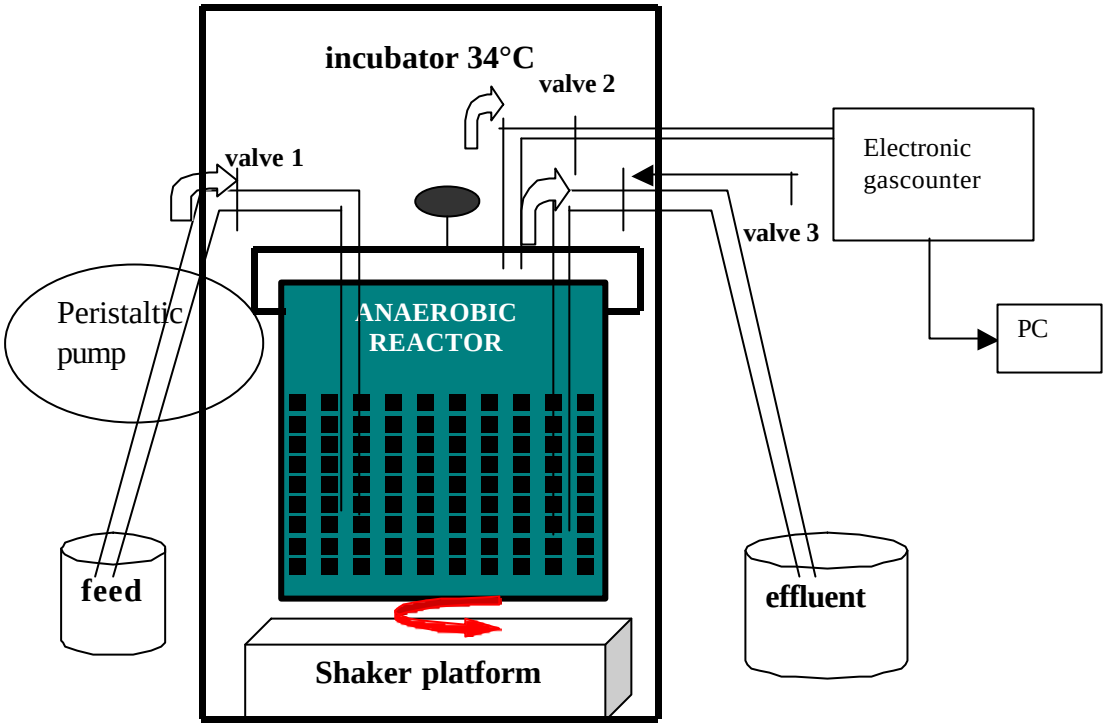


Figure 1: Anaerobic reactor and gas meter device

When the reactor is at the non-feed mode, valve 1 and valve 3 are closed. Valve 2 is opened permitting the biogas to escape from the reactor. Subsequently, the biogas passes by an electronic milligascounter device (Fachhochschule Bergedorf, Hamburg-Harburg, Germany) with a resolution of 1 ml and an accuracy of 3%. The biogas has been constantly measured and analysed over a period of 1 month (see results).

The volumetric loading rate of the mesophilic digester was gradually increased from 0.95 g COD/L.day to 2.17 g COD/L.day over a period of two weeks. The reactor has been run for 3 months on 2.17 g COD/L.day and has a stable performance.

In order to obtain the amount of 90 g of dried digester particulates as agreed on the 2nd progress meeting in Barcelona, the operating parameters of the digester were changed. The volumetric loading rate was further increased to 2.5 g COD/L.day over the month december and january. Moreover, the dry matter content of the synthetic feed was also strengthened up from 2.8% to 3.8% dry matter. The reactor was fed in quantities of 0.5 L feed/day. In order to maintain an hydraulic retention time of at least 15 days, the liquid reactor volume was increased from 5.5 L to 7.5 L.

3.2 Substrate composition

The composition of the 2.8% DM substrate was as follows:

90 g Spirulina (95%DM)	= 85.5 g DM	= 2.85g/L	= **10%**
210 g wheat straw (95%DM)	= 199.5 g DM	= 6.65g/L	= **24%**
2100 g fresh cabbage (9%DM)	= 189 g DM	= 6.3g/L	**= 22.5%**
210 g soya (90%DM)	= 189 g DM	= 6.3 g/L	= **22.5 %**
1800 g faeces (10%DM)	= 180 g DM	= 6g/L	**= 21.5 %**

Regarding the composition of the 3.8% DM substrate, the relative proportions of the different substrate components were kept similar.

3.3 Substrate preparation

First, Spirulina algae and soya were added to a small amount of tap water. 1 kg of chopped straw was received from Partner 3. This straw has been used for the digestion and preparation of 90 g of dried particulates which have been distributed at the end of january. The cabbage and faeces were originally grinded with a kitchen mixer in a separate amount of water and then added to the algae, straw and water.

3.4 Influent and effluent analysis

COD (dichromate method), TAN (destillation method), dry matter (106°C drying) and Kjeldahl nitrogen (destillation method) were determined according to the standard methods. The TOC/TC/TIC-analysis has been redetermined (spectrophotometric method, Dr.Lange) as agreed on the latest progress meeting.

The biogas composition was determined by means of gas chromatography. By including a septum between the digester and the gas meter device, gas sampling could be done on regular time intervals.

The phosphorous, sulfur and chloride concentrations present in the influent and effluent were computed by means of ion chromatography. Both influent- and effluent samples were first diluted 50 times, centrifuged and finally filtered over a 0.4 µm filter.

Peak identification and quantification of the components detected with this system were accomplished by internal standards, allowing to convert peak areas to concentration values.

4. Results

4.1 Substrate characterisation

The dry matter content, COD, TAN, Kj-N, VSS and ash-content of the 2.8% dry matter influent are displayed in Table 1.

Table 1: Substrate characteristics of the liquid phase

DM-content	COD	TAN	Kj-N	VSS	ash-content
2.8%	28 g/L	0.4 g/L	1.4 g/L	23 g/l	5 g/l

As agreed on the latest progress meeting, the total carbon (TC), total organic carbon (TOC) and the total inorganic carbon (TIC) were determined again. The values found for the influent are TC = 17.2 g/L, TOC = 9 g/L and TIC = 8.2 g/L.

4.2 Biogas production

The biogas production was measured on a two day basis for 1 month by means of an electronic gascounter. The average methane content of the biogas was determined by daily gas sampling. Since the volumetric loading rate was increased from 0.95 g COD/L.day over 2.17 g COD/L.day to 2.5 g COD/L.day, a theoretical biogas production of appr. 2.5 Liters/day, 6 Liters/day and 9.3 Liters/day respectively can be expected.

The biogas production was measured over two time periods: during two weeks at a B_v of 2.17 g COD/L.day (results not shown) and during a month at a B_v of 2.5 g COD/L.day (Figure 2).

Because the B_v was 2.5 g COD/L.day, the reactor volume was 7.5 Liter and assuming that 1 g of COD corresponds with 0.5 Liter of biogas, it could be calculated that approximately 9.3 Liters of biogas/day or 18.8 Liters per 2 days should be produced. The measured biogas production over the one month appeared to be 9.1 Liters/day on average. The average difference on a two day basis measurement between the experimental and theoretical gas production (Figure 2) was found to be 3 Liters.

After 1.5 months of feeding the reactor at 2.5 g COD/L.day, the biogas production however started to decrease. The theoretical biogas production of 9.3 Liters/day could not be reached anymore and decreased with 50%. Since hydrogen sulfide could clearly be smelled, the H_2S concentration in the biogas was measured and showed to be 2% of the biogas volume. The reactor was temporary fed (1week) at a lower

loading rate permitting the methanogenic bacteria to recover. Clearly, at a high rate the sulphate reducing bacteria become more prominent. The latter was probably related to the high B_v levels imposed to the digestor.

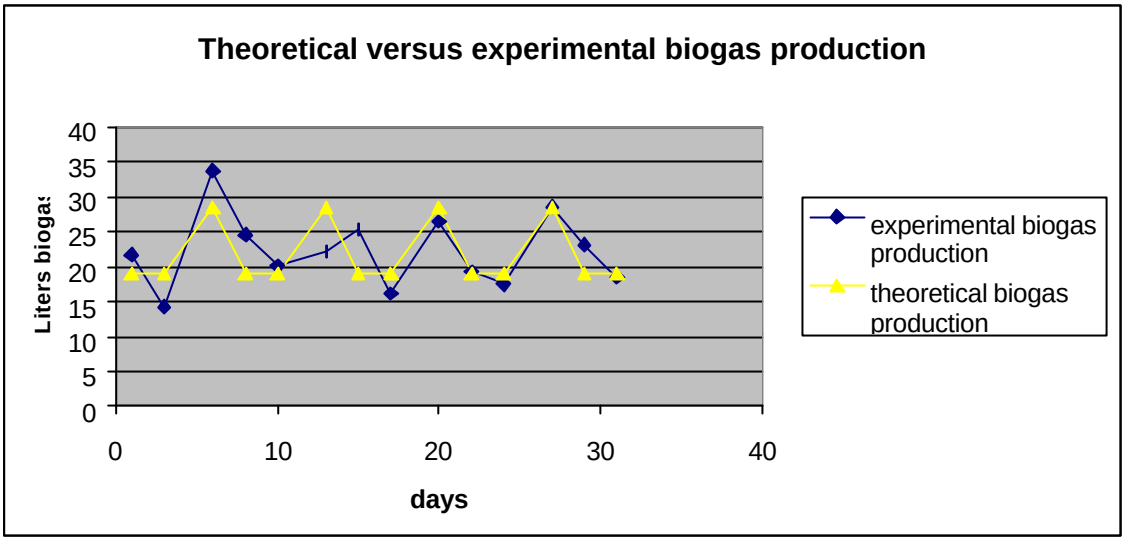


Figure 2: Comparison of theoretical and experimental biogas production

The average methane- and carbon dioxide content during this month was 62.2% and 37.4% respectively (1 measurement/week). Given the low standard deviation on the different measurements (< 2%) and the daily follow-up of the reactor pH, 1 measurement a week was considered satisfactory. The hydrogen sulfide (H_2S) content of the biogas was found to be relatively high and ranged from 1.5-2% on average .

4.3 Carbon and nitrogen massbalances

In Figure 3, a detailed overview of the C mass balances is given for the gas and liquid phase of the digester effluent.

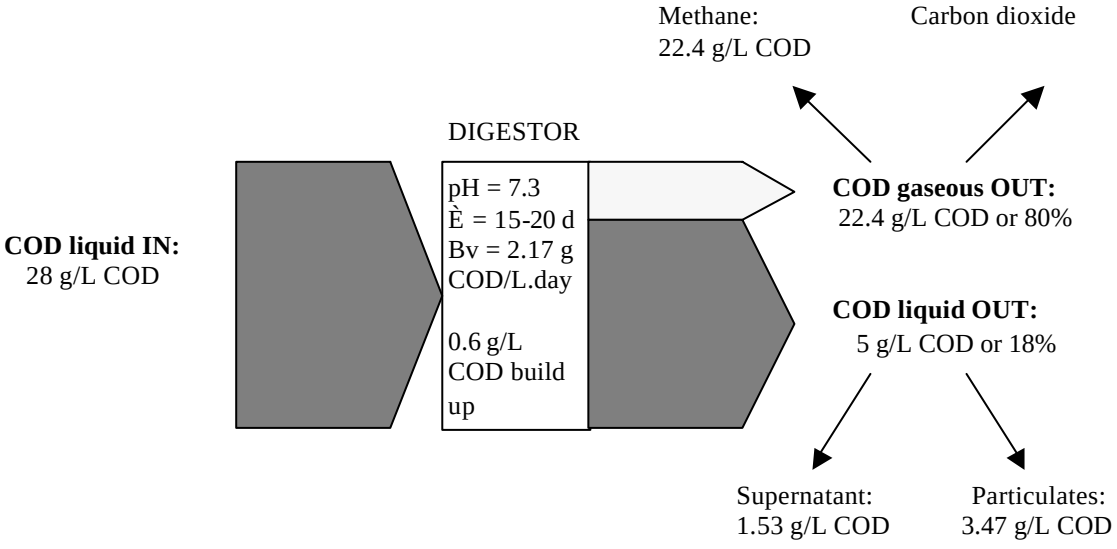


Figure 3: Carbon mass balances for the gaseous and liquid phase

Since the reactor is from the CSTR-type (no solid-liquid separation) but not completely mixed (only gently shaken), there is little accumulation of solid matter (varying from 1-3% of the influent COD).
Starting from the 2.8 % dry matter substrate, 12.3 g methane gas can be produced per liter substrate (28g/L COD). About 5 to 6 g COD per liter substrate remains in the effluent. According to literature and given the visual outlook of the digester residue (greenish with straw particles), the residue mainly consists of algae cells and straw particles.

In terms of nitrogen balances, the anaerobic digestor liquifies the organically bound nitrogen with high efficiency. As a matter of fact, 1g/L of organically bound nitrogen present in the 2.8% dry matter substrate can be completely liquefied into ammonia at a retention time of 20 days. The detailed nitrogen balances are shown in Figure 4.

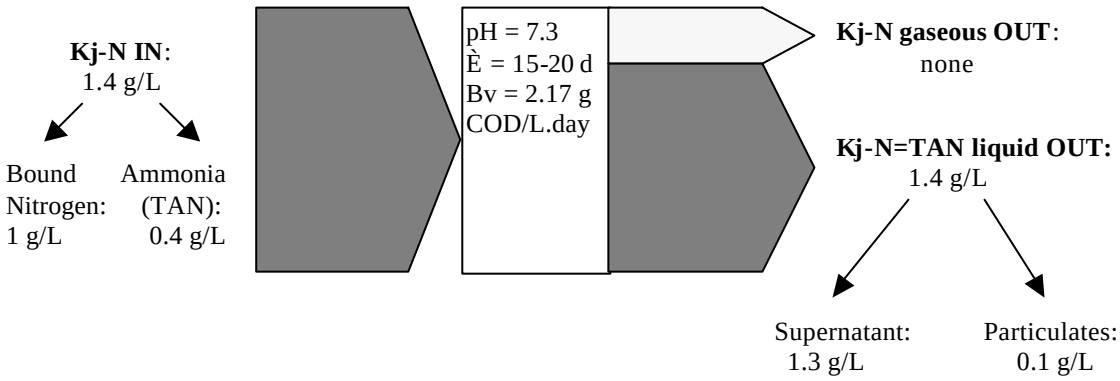


Figure 4: Nitrogen mass balances for the gaseous and liquid phase

The total ammonia (NH_3 and NH_4^+) constitutes together with the bicarbonate buffer a high buffer capacity in the digestor as already mentioned in the previous Technical Note. Since the average pH of the reactor is 7.3, part of the total ammonia will be present as ammonia and can partially be transferred to the gasphase. However, it is generally accepted that at a pH lower than 7 the total ammonia will be only present under the form of ammonium. Therefore, it can be assumed that the total ammonia in the digestor will be largely present in the form of ammonium.

4.4 Phosphorous and sulfur balances

As agreed on the latest Progress Meeting, the phosphorous and sulfur mass balances both for the liquid phase of the influent and effluent have been determined. The results are summarised in Table 2.

Table 2: Phosphorous, sulfur and chloride concentrations of the digester influent and effluent

	P-PO43- (mg/L)	S-SO4- (mg/L)	Cl- (mg/L)
Influent	520	198	162.6
Effluent	164.5	70	187.2

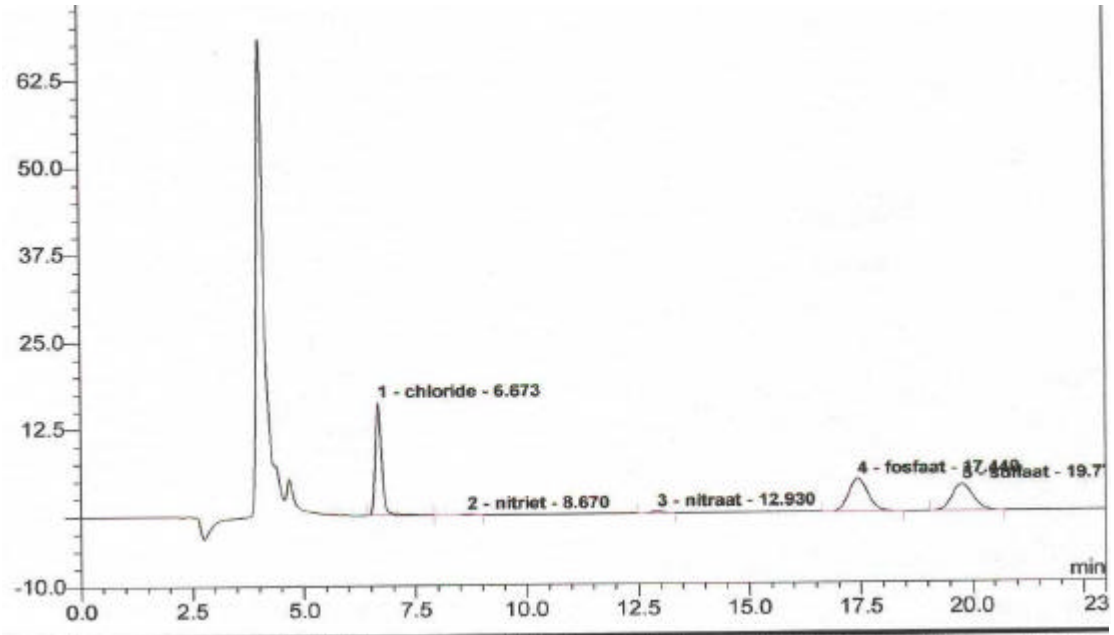
The measured sulfate levels in influent and effluent are rather high. The sulfate concentration in the effluent compared to the influent is considerably lower proving anaerobic sulfate reduction to hydrogen sulfide. Effectively, by means of gas chromatography, the measured biogas hydrogen sulfide concentrations attributed to circa 1.5-2% (Volume%) or 15-20 mL/L biogas. Since a 100% H_2S gas phase is in equilibrium with 3 g/L of S-sulfide in the liquid phase (*law of Henry*), 1.5-2% H_2S corresponds with 45-60 mg/L free H_2S in the liquid solution. From data in literature, it was found that from 50 mg/L free H_2S in the liquid phase on, the methanogens are inhibited. This could be an explanation for the breakdown of the methanogenesis at high loading rates (higher than 2.2 g COD/L.day). However, latest experiments show that the spontaneous acidification of the synthetic substrate during storage (from pH 6.5-7 to pH5-5.5) might be the cause for the breakdown of the digester at high loading rates. Further research will be carried to investigate this problem.

It must be noted that in case volatile fatty acids such as acetate rather than monomers (e.g. sugars) are the main components of the substrate COD, sulfate reduction will be less. In that case, less than 1/3 of the electron flow (normally 1/3) will be available for the sulfate reducing bacteria. This might be an explanation for the relative high amount of $S\text{-}SO_4^{2-}$ still present in the effluent.

As can be derived from Table 2, an uptake of phosphate in the digestor happens since the effluent phosphate concentration is only 1/3 of the original substrate phosphate concentration. Normally, about 75% of the phosphorus in fecal matter is under organic form. Phosphorus is generally much less mobile than nitrogen, being strongly adsorbed to organic matter and/or the anaerobic biomass. As a result, the low phosphate concentration in the effluent can be explained by the fact that the majority of the effluent phosphorous is adsorbed on the particulate matter and only a minority is still present in the liquid phase.

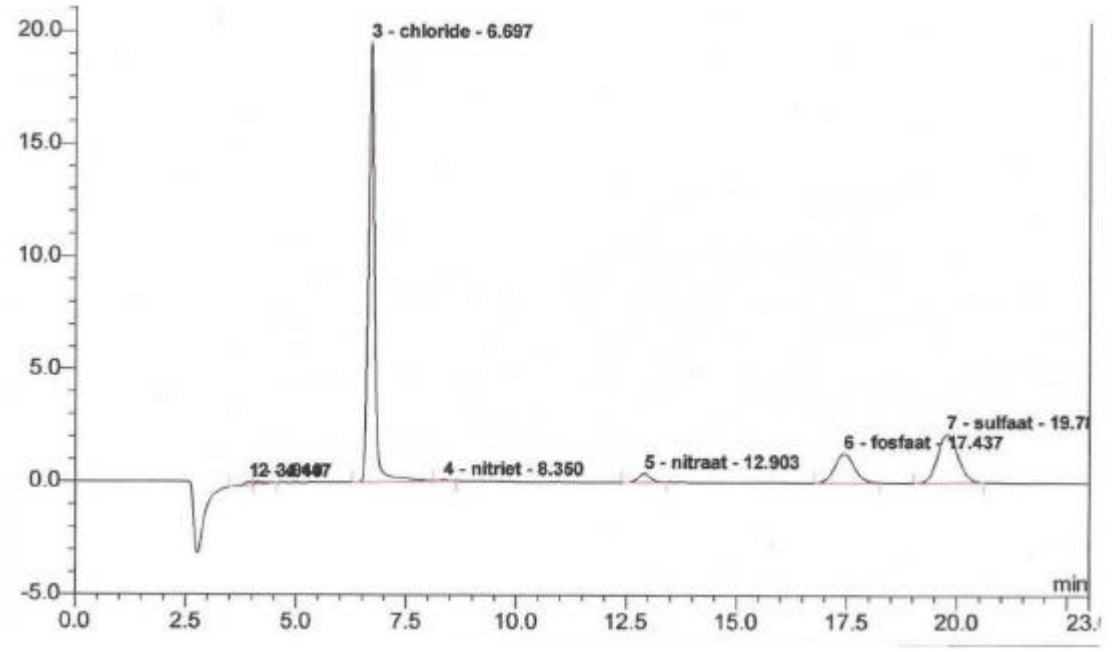
The chloride concentrations in the influent and effluent are similar and mainly derive from the fecal matter and non-edible parts of plants. The chloride concentrations in the effluent were found to be somewhat higher due to the hydrolysis of fecal matter during anaerobic digestion.

The chromatographs of the phosphate, sulfate and chloride determination by means of ion-chromatography are shown in Figure 5 and Figure 6.



No.	Ret.Time min	Peak Name	Height µS	Area µS*min	Rel.Area %	Amount mg/L	Type
1	6.67	chloride	16.130	2.776	37.93	3.640	BM *
2	8.67	nitriet	0.105	0.031	0.42	0.753	MB*
3	12.93	nitraat	0.300	0.096	1.31	1.073	BMB
4	17.44	fosfaat	4.661	2.365	32.32	10.807	BMB
5	19.77	sulfaat	3.762	2.050	28.02	4.018	BMB
Total:			24.959	7.318	100.00	20.291	

Figure 5: Determination phosphorous, sulfur and chloride mass balances of a 50-fold diluted influent sample with ion-chromatography



No.	Ret.Time min	Peak Name	Height µS	Area µS*min	Rel.Area %	Amount mg/L	Type
1	3.94	n.a.	0.136	0.036	0.68	n.a.	BM
2	4.15	n.a.	0.121	0.028	0.53	n.a.	MB
3	6.70	chloride	19.545	3.268	62.00	4.094	BMB*
4	8.35	nitriet	0.034	0.008	0.16	0.713	Rd*
5	12.90	nitraat	0.382	0.123	2.33	1.125	BMB
6	17.44	fosfaat	1.281	0.662	12.57	3.496	BMB
7	19.78	sulfaat	2.161	1.146	21.74	2.580	BMB
Total:			23.658	5.271	100.00	12.008	

Figure 6: Determination phosphorous, sulfur and chloride mass balances of the influent with ion-chromatography

4.5 Energy consumption of the anaerobic digester

From the results of the laboratory study, the energy balance of the anaerobic digester at mesophilic (34°C) conditions can be estimated. Assuming that the incubator is well insulated and no heat is lost, the only energy input required is on the one hand the heat required for maintaining the digester liquid (5.5 Liters) at 34°C and on the other hand the energy required for shaking (2 minutes/hour).

The energy balance of an anaerobic digester is generally given by equation (1):

$$NEP = BEP - PE \quad \textbf{(1)}$$

with NEP the netto energy production, BEP the bruto energy product and PE the process energy.

The bruto energy product BEP can be defined as the energy content of the biogas as a result of the presence of methane, expressed in kJ/Liter biogas. The BEP can be calculated as follows (2):

$$BEP = COD_{conversion} . V_{biogas} . F_{methane} . E \quad \textbf{(2)}$$

with $COD_{conversion}$ = 23 g/L, V_{biogas} = 0.5 L/g COD, $F_{methane}$ (average methane content of the biogas) = 0.62 and E (energy of the biogas) = 35 kJ/Liter biogas. It can be calculated that BEP = 250 kJ/Liter of biogas or **1500 kJ/day** (assuming 6 Liters of biogas/day at a B_v of 2.19 g COD/L.day).

The process energy can be defined as (3):

$$PE = Q_{tot} + E_{el} \quad \textbf{(3)}$$

with Q_{tot} the required thermal process energy and E_{el} the required electrical process energy. The term Q_{tot} is generally defined as (4):

$$Q_{tot} = Q_0 + Q_w + Q_p \quad \textbf{(4)}$$

with Q_{tot} the energy required to heat the influent to 34°C, Q_w the heat losses through the walls and Q_p the heat losses through the pipes. As a matter of fact, the PE term will largely consist of thermal losses only (Q_0) through the reactor. Since the reactions in the digestor are exotherm, only a small energy input will be required. It can be estimated that at least a temperature of 20°C can be maintained without any energy input, assuming that the digestor is well insulated. If we then calculate the Q_0 term as given in equation (5) with density ρ = 1030 kg/m³, flow rate Q = 0.5L/day, heat capacity c_p = 4.18 kJ/kg.K and Δ T = 14°C, the Q_0 term will be equal to 30 kJ/day.

$$Q_0 = r.Q.c_p.\Delta T \quad \textbf{(5)}$$

Consequently, it can be concluded that for a well insulated mesophilic digester, the required process energy PE will only be a minor fraction (10% max.) of the bruto energy product BEP. The required electrical process energy E_{el} will even be a minor fraction of the PE since this will mainly be the energy needed to stirr the reactor. If we assume an energy requirement of 30 W per Liter reactor volume and a shaking frequency of 2 minutes/hour, it can be calculated that E_{el} will be equal to 0.2-0.5 kWh/day.

4.6 Electrochemical oxidation of the digester residue (additional)

The anaerobic digester effluent was centrifuged prior to electrochemical treatment at 7000 g for 15 min. Subsequently, the supernatant phase was withdrawn and used for the electrochemical treatments. By means of potentio-kinetic measurements, the current efficiencies of different types of electrode materials were tested (see Figure 8). In this study, four types of electrode materials were subjected to an increasing potential while measuring the current simultaneously (Figure 8). Different cathode materials were tested: stainless steel (316L), massive Ti and a Ti fibre (manufactured by Bekaert®). In the separated cell (Figure 7), a cation-selective Nafion® membrane (Bekaert®) was used. In this case, a 4g/L sodium chloride solution functioned as the catholyt.The massive titanium electrode clearly showed the lowest resistance (or highest current) at all tested potentials.

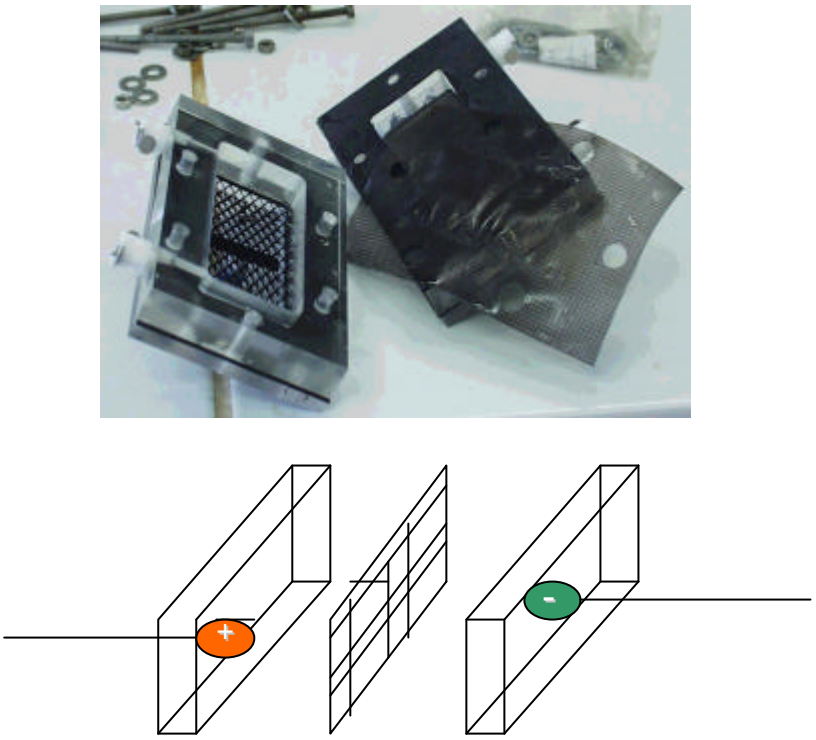


Figure 7: Picture of the separated electrolysis cell (above): left: anode compartment, right: cathode compartment with NAFION membrane on top
General scheme of experimental set-up (down): left: anode, right: cathode, middle: NAFION membrane

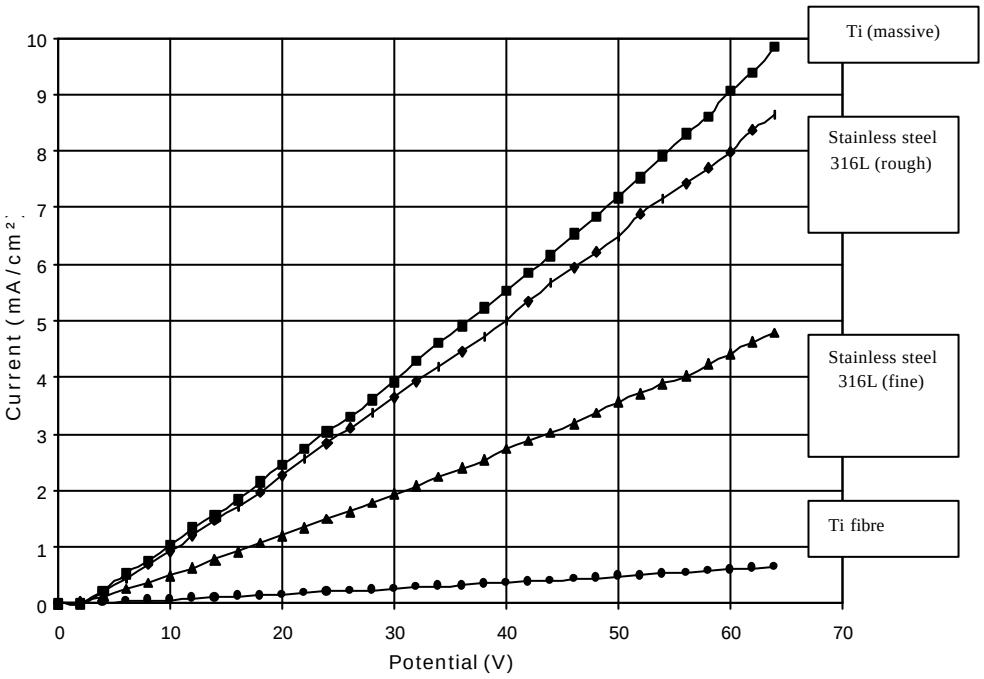


Figure 8: Potentio kinetic measurements of titanium –and stainless steel electrode materials in a 0.4% NaCl solution with graphite as anode material

In terms of nitrogen removal (Figure 9), all present nitrogen (1.4 g/L) in the digester effluent was under the form of total ammonia (TAN was similar to Kj-N). With the separated cell (NAFION membrane), up to 96% of the initial nitrogen content could be removed from the liquid phase and converted into nitrogen gas. On the contrary, only 24% of the total ammonia could be removed with the mixed cell (no membrane) after 2 hours of electrolysis. Nitrogen removal happened most rapidly during the first hour of electrolysis, both with and without membrane.

The difference in TOC-removal was less pronounced compared to the electrochemical ammonia oxidation. Without a membrane, up to 28% TOC removal could be reached while with a membrane up to 46% of the TOC was oxidised (Figure 10).

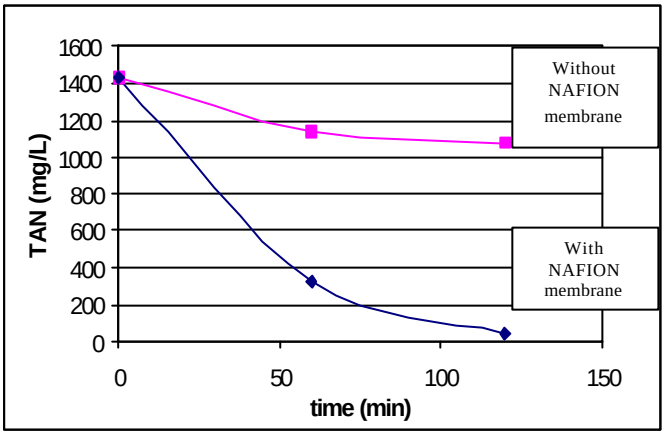


Figure 9: TAN removal by means of an electrochemical oxidation method

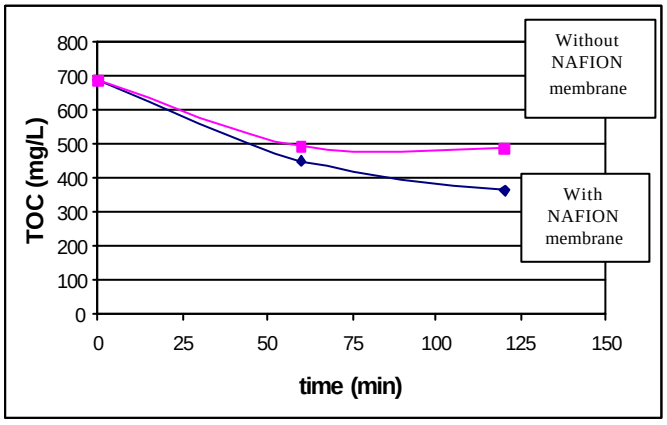


Figure 10: TOC removal by means of an electrochemical oxidation method

It could clearly be seen that the removal of organic carbon was promoted by more acidic conditions. Effectively, mainly during the last hour of electrolysis the difference in TOC removal with and without a membrane became significant. It is in this time period that the effluent gradually acidified from pH 8 to pH 2-3. Control of

pH was not considered as a viable option due to the need of chemicals. It is however possible to neutralize the acidified anolyte by adding the alkaline catholyte (pH rises at the cathode to pH 11-12).
Current efficiencies were the highest during the first half of the electrolysis run during which up to 35% of the initial TOC (0.68 g/L) and 77% of the initial TAN (1.4g/L) could be removed.
Only the energy requirements of the electrolysis runs with a NAFION membrane have been calculated since the presence of a membrane appeared to be positive both for COD and TAN removal.
The power requirement of the electrolysis cell is generally given by equation (1):

$$x = V.I.t \quad \textbf{(1)}$$

with *V* the applied voltage (Volt), *I* the applied electric current (Amperes) through the cell and *t* the time (hours). The *law of Faraday* shows the relationship between the amount of oxidised COD and the applied current following equation (2.1):

$$m(COD, kg) = \frac{M}{n}.\frac{1}{96500}.i.3600.t.10^{-3} \quad \textbf{(2.1)}$$

where M stands for the molecular mass of the oxidised compound(s), n the number of electrons released during the oxidation reaction, *i* the theoretical desired current to oxidise the compound(s) and *t* the electrolysis time. The current efficiency can than be calculated as the ratio of the theoretical current *i* over the applied current *I* .
Another way to calculate the current efficiency is the use of the instantaneous current efficiency (ICE), calculated according to the definition of Panizza et al [13]:

$$ICE = \frac{(\text{COD decrease}).(\text{Volume of solution})}{(\text{Mass of oxygen equivalent to electricity})} = \frac{F.V.(COD_t - COD_{t+})}{8.I.\Delta t} \quad \textbf{(2.2)}$$

where COD_t and $COD_{t+\Delta t}$ are the CODs at times t and $t+\Delta t$ (g/dm^3), respectively, I is the current (A), F is the Faraday constant (96 487 C/mol) and V is the volume of electrolyte. The average current efficiency η is then calculated as the average of the ICE.
The current efficiencies could only be calculated on COD basis due to the heterogenous composition of the digester leachate (concentration and identification of the residual recalcitrant compounds was not known).
Combining equation (1) and (2.1) results in the expression of the power consumption (3):

$$x_{(kWh/kgCODremoved)} = \frac{V.26,8}{\frac{M}{n}.0,01.r_{k(\%)}} \quad \textbf{(3)}$$

with ρ the current efficiency of the process. According to the experiments conducted so far, the supernatant largely exists of large organic coloured molecules with a molecular weight ranging from 500 to 50000 Dalton units such as tannic acid and other lignin-derivatives. Therefore, if we assume an average molecular weight of 20000 and a required number of electrons of 500, the M/n ratio would be 40. From equation (2), it can be calculated that the required theoretical current *i* will be in the order of 0.1 A, with t = 2 hours and m = 320 mg COD (see Figure 9). Consequently, the current efficiency of the process will be approximately 10-20%. Finally, following equation (3), it can be estimated that the average energy consumption of the electrochemical process with NAFION membrane will be in the order of 50-100 kWh/kg COD removed. Assuming a substrate with 28 g/L COD (2.8% dry matter), 1.4-2.8 kWh/Liter substrate is required. This figure can be regarded as high since the cost for a biological treatment is a factor 5-20 lower. However, the method could only be applied to remove recalcitrant organics, in this case organics which are still left in the liquid phase of the digester effluent. These organics correspond to 5.6% of the total influent COD or 1.56 g/L COD. Consequently, for each liter of waste to be fermented, only **0.078-0.15 kWh/Liter substrate** is required in order to oxidise 50% of the residual liquid organics of the digester effluent.

For the ammonia removal, the following oxidation reaction will be of applicance:

$$2NH_3 \rightarrow N_2 + 6e^- + 6H^+$$

Taking into account the molecular weight of ammonia (M = 17), it can be calculated by means of equation 2.2 that the average current efficiency for ammonia oxidation is in the order of 50%. By means of equation (3), the estimated energy consumption for ammonia oxidation solely will be in the order of 150-200 kWh/kg ammonia removed. This higher energy demand can be explained by the low M/n ratio due to the high electron requirements (6 electrons per molecule of ammonia). As a result, the overall energy requirement for electrochemical ammonia removal will be a factor 2 higher compared to COD oxidation.

The electrochemical oxidation of ammonia gas makes it possible to convert the highly reactive form of nitrogen (ammonia) into the harmless non-reactive form nitrogen gas in a controlled way. In case of a nitrogen excess under the form of ammonia within the MELiSSA loop, nitrogen can be removed electrochemically in a selective way . Eventually, nitrogen gas can be converted again by nitrogen fixating plants into the nutritive form.

4.7 Microwave digestion of the digester residue (additional)

Regarding the liquefaction results of the thermal microwave treatment, the digester particulates were first removed from the 500 mL samples of the liquid phase by centrifugation at 7000g for 15 min. Since 12.6% or 3.52 g/L of the influent COD is under the form of particulates, 1.76 g particulates could be recovered per 500 mL of digester effluent. To these samples, 100 mL of distilled water was added. As a result, the total initial COD of the samples was approximately 17.6 g/L corresponding to a similar dry weight figure. From this mixture, 20-40 mL samples were prepared and subjected to thermal microwave treatment. The other Microwave procedures were:
CEM Mars 5 Microwave oven of 1200 Watt with 14 individual 100 ml teflon pressure vessels, runs of 20 min at T = 60-210°C, pressure = 1-12 bar.
As can be seen in Figure 11, a significant amount of organically bound nitrogen is transferred from the particulates to the supernatant phase during microwave digestion (see also Table 3). In the contrary, the TAN-levels both in the mixed liquour and the supernatant remain constant during the thermal treatment. Since the organically bound nitrogen is mainly contained into the particulates, the increase of Kj-N in the supernatant phase after thermal treatment clearly proves liquefaction of the particulates such as straw. Moreover, a significant increase of the soluble COD and TS (total solids) could be observed.

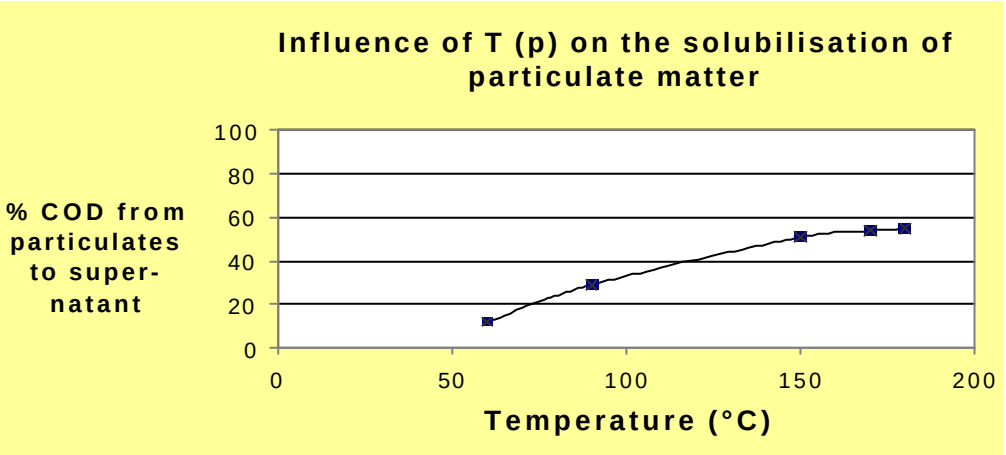


Figure 11: Liquefaction of digester particulates as a function of temperature

In Figure 11, it can be seen that a maximum solubilisation degree of 55%±5% was reached at 180°C at 9 bar for 20 min. In one run at 1200 Watt, 14 vessels containing each 20-40 ml of sample (17.6 g/L COD) or 0.35-0.7 g of particulates can be subjected to thermal treatment. This corresponds to 4.9-9.8 g of particulates per run (20 minutes). From these figures, it can be calculated (1 kWh = 3600 kJ) that 40-80 kWh/kg particulates are required to liquify 50% of the digester particulates. This

corresponds with 1.14-2.28 kWh/Liter substrate if the method would be applied to the undigested substrate. However, since only 12.6% or 3.5 g COD of the influent COD is under the form of recalcitrant digester particulates, **0.14-0.28 kWh/Liter** substrate is the estimated energy requirement for the thermal microwave treatment.

Table 3: Overview of the nitrogen balance after microwave treatment (20 min, 180°C, 9 bar)

mixed liquour (25-30 g/L COD)	Kj-N (mg/L)	TAN (mg/L)	Kj-N minus TAN = organically bound N (mg/L)
BEFORE TREATMENT	1550	550	1000
AFTER " "	1500	639	861
supernatant (1 g/L COD initially)	Kj-N (mg/L)	TAN (mg/L)	Kj-N minus TAN = organically bound N (mg/L)
BEFORE TREATMENT	614	520	94
AFTER " "	1314	453	861

5. Discussion and Conclusion

The computing of the mass balances revealed relatively high free H_2S concentrations in the gas- and liquid phase of the mesophilic digester.

Sulfide in anaerobic reactors normally originates from the biological reduction of sulfate with hydrogen. Consequently, due to the rather high sulfate concentrations, it can be assumed that the sulfate reducing organisms present in the digestor severely compete with the hydrogen utilising organisms. The sulfate brought into the reactor mainly originates from non-edible parts of plants and fecal matter. Sulfate itself is not really inhibitive. Even up to concentrations of 20 g SO_4^{2-}, quasi normal methano-genesis can be possible. During digestion of normal organics, circa 1/3 of the electronflux proceeds via hydrogen gas as an intermediate. Hence, the amount of COD scavenged by the sulfate reduction process will normally not surpass 25-30% of the total amount converted. It is generally agreed that for each gram of COD-H_2 removed during digestion, about 0.5 g of S-sulphate can be reduced into S-sulphide. Consequently, per g COD present in the feed, 0.5/3 g S-SO_4^{2-} can be produced. In this case, the latter means that theoretically 4.66 g S-SO_4^{2-} can be reduced. As a matter of fact, free H_2S concentrations in the liquid phase of 50 mg/L or more can inhibit acetotrophic methanogens by about 50% while complete inhibition generally occurs at a free H_2S level of circa 200 mg/L, even at high COD/S-sulfate ratios. Other

parameters determining whether inhibition will occur or not are operating process parameters such as temperature, the shaking regime and the ratio between the gas- and liquid volume in the digestor. Due to the mesophilic temperature, the hydrogen sulfide solubility is rather high. As a result, liquid sulfide levels inside the digestor at high volumetric loading rates can be expected to be close to inhibitory levels for methanogenesis. This might be an explanation for the decrease in biogas production at higher loading rates (> 2.2 g COD/L.day).

The mass balances for the mesophilic digester have been computed in detail regarding the C, N, P and S compounds. The reactor showed a reliable performance as long as the volumetric loading rate was equal or below 2.2 g COD/L.day. At a loading rate of 2.5 g COD/L.day for one month, the biogas production decreased. This phenomenon could partially be related to the relatively high (close to inhibition) free H_2S levels in the digester. Probably, a relatively high proportion of the electrontransfer (more than 2/3) passes by the consumption of the volatile fatty acids such as acetate. The energy consumption of the mesophilic digester showed to be only a minor fraction of the potential energy content of the methane.
In addition, electrolysis and thermal treatment of the recalcitrant compounds present in the digester effluent showed a breakdown of 50% or more. The energy consumption turned out to be high for both methods if it would be applied to the raw digester effluent. However, taking into account the overall mass balances, the energy consumption of both methods is significantly lower.
An overview of the C and N mass balances is given in Figure 12 (end of document).

6. Outlook

A long term feeding of the mesophilic digester (by means of batch fermentation tests) with the digestion products of partner 2, 3 and 4 are envisaged. The digestion residu of the mesophilic digester has been dried by Partner 1 and has been sent in quantities of 30 grams to partner 2, 3 and 4 as a first closed loop experiment agreed on the 2nd progress meeting.
A second closed loop experiment is envisaged during the month april. Currently several analysis methods are evaluated for the determination of lignine, cellulose and hemi-cellulose to determine the fiber breakdown during digestion. The Van Soest method seems to be the most appropriate one so far.
A second anaerobic digester will be set up in february in order to provide sufficient particulates to distribute among the different partners for a second closed loop experiment.

7. References

Kroiss, H. & Wabnegg, F.P. (1983). Sulfide toxicity with anaerobic wastewater treatment. In: W.J. Van der Brink (ed.), Proceedings European Symposium on Anaerobic Wastewater treatment (AWWT), Netherlands.

Panizza, M., P. Michaud, G. Cerisola, C. Comminellis: "Electrochemical treatment of wastewaters containing organic polutants on boron-doped diamond electrodes" Electrochemistry Communications 3 (2001), 336-339.

Parkin, G.F., Lynch, N.A., Kuo, W.C., Van Keuren, E.L., Bhattacharya, S.K. (1990). Interaction between sulfate reducers and methanogens fed acetate and propionate. Res. J. WPCF Vol 62: 780-788.

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Figure 12: C and N massbalances of the mesophilic digester