Chapter II

IMPORTANT PARAMETERS FOR THE ANAEROBIC DIGESTION PROCESS

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Introduction

The anaerobic digestion process is biologically mediated and involves different types of microorganisms. Thus, specific conditions must be respected and followed for the correct functioning of biodigesters. In this chapter, we will discuss the main parameters and how to calculate them.

Alkalinity

Alkalinity can be understood as a measure of the buffering capacity of a system, that is, the ability to avoid sudden changes in pH. It is often attributed to the balance between CO_2 dissolution and carbonic acid (H_2CO_3) formation in anaerobic environments.

The total alkalinity is given by the sum of the concentration of ions hydroxyl (OH^-), carbonate (CO_3^{2-}), and bicarbonate (HCO_3^-), being expressed as the concentration of calcium carbonate ($CaCO_3$).

It can be determined by titrating the sample with a sulfuric acid solution (e.g., 0.1 mol.L^{-1}) up to pH 4.5 and applying the equation:

$$\boldsymbol{Alk = M \times E \times \frac{100,000}{D}}$$

Equation 1

Where:

Alk = Alkalinity (mg $CaCO_3.L^{-1}$)

M = Sulfuric acid solution concentration (mol.L)

D = Sample volume (mL)

E = Sulfuric acid titred volume (mL)

100,000 = Coefficient to adjust the unit of measure

Determination of the VFA/TA ratio

One of the monitored parameters is the relationship between the accumulation of volatile fatty acid and total alkalinity ratio, known as the VFA/TA ratio. The result is a simple value dependent on the relationship of these two parameters, relative to short-chain organic acids (VFA) and alkalinity (TA).

They can be calculated by titrating the sample with sulfuric acid, following the equation:

$$\boldsymbol{VFA/TA = \frac{\left((V_{pH4.4} - V_{pH5.0}) \cdot \frac{20}{V_{sample}} \cdot \frac{M_{acid}}{0.1} \cdot 1.66 - 0.5\right) \cdot 500 \cdot V_{sample}}{0.5 \cdot M_{acid} \cdot V_{pH5.0} \cdot M_{CaCO_3} \cdot 1,000}}$$

Equation 2

Where:

VFA/TA = Intermediate alkalinity to partial alkalinity ratio

$V_{pH4.4}$ = Titrated acid volume to pH = 4.40 (mL)

$V_{pH5.0}$ = Titrated acid volume to pH = 5.00 (mL)
V_{sample} = Centrifuged sample volume (mL)
M_{acid}= Acid molarity (molar concentration of the hydrogen ion (dissociated) in the acid) ($mol.L^{-1}$)
M_{CaCO3} = calcium carbonate molar mass in 100 $g.mol^{-1}$

Hydraulic retention time

Hydraulic retention time (HRT) is the mean time that the substrate remains inside the biodigester, that is, the ratio between the biodigester volume and the feeding flow rate, determined using Equation 3.

$$HRT = \frac{V}{Q} \qquad \textit{Equation 3}$$

Where:

HRT = Hydraulic retention time (d)
V = Biodigester volume (m^3)
Q = Feeding flow rate ($m^3.d^{-1}$)

Organic loading rate

Organic loading rate (OLR) represents the amount of substrate added to the biodigester in a given period of time. It is obtained using Equations 4 or 5.

$$VOL = \frac{(Q \times S_V)}{V} \qquad \textit{Equation 4}$$

$$VOL = \frac{S_V}{HRT} \qquad \textit{Equation 5}$$

Where:

OLR = Organic loading rate ($kg_{VS}.m^{-3}d^{-1}$)
Q = Flow rate ($m^3.d^{-1}$)

Sv = Concentration of volatile solids in the substrate ($kg.m^{-3}$)
V = Reator volume (m^3)
HRT = Hydraulic retention time (d)

OLR influences the entire dynamics of the anaerobic digestion process. An optimal organic loading rate provides adequate conditions for microorganism growth and, consequently, higher process stability. Low OLRs may represent a low food/microorganism ratio, which results in low biological activity. High OLRs may present a high food/microorganism ratio, which can lead to the accumulation of volatile fatty acis and process failure. The ideal OLR is related to the biodigester model, applied technology, and substrate type.

OLR x temperature relationship

Safley and Westerman (1990) presented Equation 6 using different versions of the van't Hoff-Arrhenius relationship to estimate the limit of the organic loading rate relative to changes in the biodigestion temperature:

$$\frac{OLR_2}{OLR_1} = e^{p(T_2 - T_1)}$$

Equation 6

Where:

OLR_1 and OLR_2 = Organic loading rate ($kg_{VS}.m^{-3}.d^{-1}$)
T_1 and **T_2** = Temperature (^oC)
p = Constante 0.1 ($^oC^{-1}$)

Example 1

There is an UASB biodigester treating swine manure. It is operated with a organic loading rate of 0.3 $kg_{VS}.m^{-3}.d^{-1}$ and a mean temperature of 18 °C. What OLR can this reactor be subjected to if a heating system is installed, increasing the temperature of the reaction medium to 35 °C?

A: The equation proposed by Safley and Westerman (1990) is applied.

Where:

OLR_1 = 0.3 $kg_{VS}.m^{-3}.d^{-1}$

OLR_2 = Unknown variable

T_1 = 18 °C

T_2 = 35 °C

$$\frac{OLR_2}{0,3} = e^{0,1(35-18)}$$

$$OLR_2 = 1.64\ kg_{VS}.m^{-3}.d^{-1}$$

Methods for evaluating anaerobic sludge and substrate quality

There are laboratory tests carried out on a small scale and under controlled or monitored conditions to evaluate the anaerobic digestion kinetics (microorganism activity and substrate degradability characteristics, among others). Characterizing the chemical and physical composition of residues is an essential step, but biokinetic tests are also extremely important to visualize the real interaction between microorganisms and substrates. These kinetic tests consist of anaerobic respirometric tests and usually involve the evaluation of biogas or methane production from the known mass of inoculum or substrate biomass (or organic matter defined as VS, COD, or TOC).

Several methods, standardized or not, are found in the literature for evaluating anaerobic kinetics. Some focus on the efficiency of microorganisms, others focus on substrate degradability under anaerobic conditions, and others aim to evaluate the biochemical potential of methane (or energy potential) of the substrate. Some evaluate the toxicity of inhibitory substances in the anaerobic process. However, all methods are based on the batch incubation of substrate(s) mixed with anaerobic inoculum under controlled conditions. Table 1 shows a summary of the usual and standard methods for studying anaerobic kinetics.

Table 1. Summary of anaerobic kinetic methods.

Method	Purpose and/or application
SMA	Measures the specific activity of methane production in anaerobic sludges. Used to make comparisons between inoculums or evaluate efficiencies in anaerobic reactors
ISO 13.641	Anaerobic toxicity test. It is based on the measure of inhibition of biogas production after three days of incubation. Used to evaluate the influence of different chemical compounds on aerobic digestion
ISO 11.734	Estimates the degradation of various organic substances by measuring biogas production
ASTM E2170-01	Estimates the degradation of chemical substances by measuring biogas production and by chemical analysis of the residual concentration. A recognized method in the USA
DIN 38.414-8	Measures the degradation of sludge and effluents by measuring biogas production
VDI 4.630	Method for measuring BBP and BMP. Applied to various types of substrates, including agricultural residues and crops. A recognized method in Germany and European countries

Specific methanogenic activity (SMA)

This test is primarily used to evaluate the performance of methanogenic microorganisms (or inoculum). According to Aquino et al., (2007), SMA can be used as a parameter for monitoring the "efficiency" of the methanogenic population in a biological reactor. The studies of Valcke and Verstraete (1983), Zeeuw (1984), and Dolfing and Bloemen (1985) were pioneers in the development and use of SMA tests as a tool to characterize and evaluate anaerobic reactors in sanitary effluents. All tests for determining SMA available in the literature are based on measuring the methane production rate as a function of inoculum concentration. However, there is no standardized methodology for this purpose, and, depending on the methodology, establishing a relationship between SMA results in the studies available in the literature is extremely difficult. Although SMA is a very important parameter, the available methods do not have standardization and can hardly be used to make comparisons between experiments.

Aquino et al., (2007) performed a literature review on the possible methodologies available on this subject. The methodological differences range from the use or not of the culture medium for inoculum conditioning to ways of measuring the gases produced in the digestion. The

first considerations about the test were made based on batch tests by Zeeuw (1984), who measured the methane production rate of sludges from a known organic load and VS concentration. The substrate applied in the study by Zeeuw (1984) varied from a mixture of volatile acids, usually acetic, propionic, and butyric acids, to the use of a single substrate, mainly acetate, besides adding metal solutions to the nutrient solution, ensuring that there were no limitations for methane production. This method measures gas production via liquid displacement, using a sodium hidroxide solution, in which carbon dioxide is dissolved in the medium, ensuring that the displaced liquid comes from the amount of methane released by the sludge.

Dolfing and Bloemen (1985) proposed a methodology based on the gas chromatography analysis of methane produced in the headspace of serum vials. In this method, the gas is sampled with a lock syringe to keep the gas at the same pressure as the vial. The mixture of acids or acids separately (e.g., only acetate or propionate) and an anaerobic buffer solution are added to the sludge. The use of volatile acids individually has some advantages, as the knowledge of their degradation allows estimating the maximum conversion rates for each substrate to obtain information about inhibition or limitation of the process by high or insufficient concentrations of some type of acid. Moreover, it can be useful in identifying the bacterial genera present in the sludge.

ISO 13641 standards

ISO 13641 standards – Water quality – Determination of inhibition of gas production: This procedure establishes protocols to determine the toxicity of possible substances in anaerobic systems. It is divided into two standards: part 1, which refers to the general test procedure; and part 2, which refers to adaptations of methodology for low concentrations of microbial biomass. Basically, the protocols define methodological guidelines for estimating the 50% inhibitory concentration (IC50) of biogas production. It is based on the incubation of an anaerobic inoculum together with a standard substrate and mixed to different concentrations of the inhibitory agent to be evaluated. The produced gas volume is measured after incubation for three days at 35 °C and compa-

red with the test gas production without inhibitor addition. This procedure is dedicated to the evaluation of acute toxicity to the anaerobic process. There are no standardized procedures for chronic toxicity testing (long-term testing).

ISO 11734:1995

ISO 11734:1995 - Evaluation of "ultimate" anaerobic biodegradability of organic compounds in digested sludge - Method by measurement of the biogas production: This standard presents the description of a standardized kinetic test to evaluate organic chemical compounds against anaerobic microorganisms. The test consists of exposing the anaerobic inoculum to the chemical compound of interest for a period more than 60 days. The evaluation of biogas production is carried out through manometric measurements.

ASTM E2170-01 (2008)

ASTM E2170-01 (2008) - Determining anaerobic biodegradation potential of organic chemicals under methanogenic conditions: Similar to the ISO standard, this standard, elaborated by the metrology agency of the United States, also presents the description of a kinetic test of degradation of organic chemical compounds under anaerobic conditions. The test consists of exposing the anaerobic inoculum to the chemical compound of interest for a period between 25 and 30 days. The evaluation of biogas production is carried out through manometric measurements. Both ISO and ASTM standards are mainly applied in the evaluation of substances used in the medical field.

DIN 38414-8

DIN 38414-8 - Determination of the amenability to anaerobic digestion: This German standard establishes basic conditions for the execution of kinetic tests in batches to evaluate the anaerobic degradation of organic substrates using volumetric tests of biogas production.

VDI 4630

VDI 4630 - Fermentation of organic materials – Characterisation of the substrate, sampling, collection of material data and fermentation tests: This German standard is recognized in the European Union and establishes conditions for carrying out batch, semi-continuous, and continuous kinetic tests to evaluate the anaerobic degradation of organic substrates. It is an improvement of the DIN 38414-8 standard and is widely used by the European community to evaluate the biochemical methane potential (BMP) of different substrates. It is also used as a reference for bench-scale process simulation to assist the operation of large-scale biogas production plants.

The VDI 4630 (2006) standard establishes rules and the need for equipment to carry out fermentation tests on organic materials. Batch tests can provide information on a) the possibility of biogas production and the anaerobic biological degradability of a given material or mixture of materials; b) the qualitative evaluation of the degradation rate of the material under study; and c) the evaluation of the inhibitory effect of the investigated material in a given time interval.

Batch tests do not generate information on a) the process stability with reactors continuously fed with the investigated material; b) the biogas production under practical conditions different from those under which the test was carried out due to possible positive or negative synergistic effects; c) the mono-fermentation of the substrate under process conditions; and d) the organic loading rate limits.

The results of fermentation tests depend primarily on the used sludge (anaerobic inoculum) activity. The inoculum is usually collected in a biogas plant to provide the highest diversity of anaerobic microorganisms possible. The inoculum must contain a concentration of dry organic matter (volatile solids) higher than 50% of the total solids.

Some restrictions must be considered to determine the amount of substrate and inoculum used in the test: a) the amount of substrate must not exceed the amount of inoculum ($VS_{substrate}/VS_{inoculum} \leq 0.5$) to predict inhibitions in the batch test; b) the biogas production from the substrate must be at least 80% higher than the inoculum contribution; and c) the

solids concentration in the batch test must not exceed 10%, ensuring adequate mass transfer during the test.

Materials with known biogas production capacity are used to ensure the activity of the anaerobic inoculum. Possible reference material is crystalline cellulose, which produces between 740 $L_N.kg_{VSadd}^{-1}$ to 750 $L_N.kg_{VSadd}^{-1}$. These values must be recovered by at least 80% in a control test. This recovery value ensures that the inoculum has satisfactory biological activity and is suitable for carrying out BMP tests.

Normalization of biogas volume

Biogas production must always be expressed in a normalized way to standard temperature and pressure conditions (273 K and 1,013 hPa). Equation 7 is used for normalization:

$$V_N = \frac{V \cdot (p - p_w) \cdot T_0}{p_0 \cdot T} \qquad \textit{Equation 7}$$

Where:

V_N = **Biogas volume normalized to normal temperature and pressure conditions** (mL, L or m^3)

V = Produced biogas volume (mL, L or m^3)

p = Biogas pressure at the reading time (hPa)

p_w = Water vapor pressure as a function of room temperature (hPa)

T_0 = Temperature under normalized conditions (273 K)

p_0 = Pressure under normalized conditions (1,013 hPa)

T = Biogas temperature (K)

Expression of results

Table 2 shows important parameters for monitoring bioreactors and their measurement units. The monitoring of these variables contributes to better process control and knowledge of the biodigester operating conditions.

Table 2. Definitions of important variables and measurement units for the biodigester control.

Parameter	Expression	Unit
Temperature	T	^{o}C, K
Substrate concentration	S_o	% (e.g., $g_{TS}.100\ g_{FM}^{-1}$) % (e.g., $g_{VS}.100\ g_{FM}^{-1}$) $g_{VS}.L^{-1}$ ou $kg_{VS}.m^{-3}$ $g_{VS}.kg_{FM}^{-1}$
Volatile fatty acis	VFA	$mg_{HAc}.L^{-1}$
Intermediate alkalinity to partial alkalinity ratio	VFA/TA	mg_{HAc}/mg_{CaCO3}
Particle size	ps	mm
Added organic loading rate	OLR_{add}	$kg_{VSadd}.m^{-3}_{reactor}.d^{-1}$
Removed organic loading rate	OLR_{rem}	$kg_{VSrem}.m^{-3}_{reactor}.d^{-1}$
Hydraulic retention time	HRT	h or d
Redox potential	E_H	mV
Biochemical methane potential	BMP	$L_{N\ CH4}.kg_{VSadd}^{-1}$, $L_{N\ CH4}.kg_{FMadd}^{-1}$
Biogas productivity	PB	$Nm^3_{biogas}.m^{-3}_{reactor}.d^{-1}$ $L_{N\ biogas}.L_{reactor}^{-1}.d^{-1}$
Methane productivity	PM	$Nm^3_{CH4}.m^{-3}_{reactor}.d^{-1}$ $L_{N\ CH4}.L_{reactor}^{-1}.d^{-1}$
Biogas production	PBd	$L_N.d^{-1}$
Biogas yield	BY	$L_{N\ biogas}.kg_{VSadd}^{-1}$, $L_{N\ biogas}.kg_{FMadd}^{-1}$
Methane yield	MY	$L_{N\ CH4}.kg_{VSadd}^{-1}$, $L_{N\ CH4}.kg_{FMadd}^{-1}$
Biogas composition ($v\ v^{-1}$)	Bc	% CH_4($L_{N\ CH4}.100\ L_{N\ biogas}^{-1}$), % CO_2($L_{N\ CO2}.100\ L_{N\ biogas}^{-1}$), ppm$V_N$ H_2S ($mL_N.m^{-3}$)
Digestate composition	Dc	% (e.g., $g_{TS}.100\ g_{FM}^{-1}$), % (e.g., $g_{VS}.100\ g_{FM}^{-1}$), $g_{VS}.L^{-1}$, $kg_{VS}.m^{-3}$

HAc = acetic acid; MF = fresh matter; TS = total solids; VS = volatile solids; add = added; rem = removed; N = normal.

Source: Kunz et al., 2016.

Problems, causes, and solutions in the biodigester operation

Table 3 shows corrective actions for the most frequent operational problems that can occur in a biodigester.

Table 3. Possible problems found in the biodigester operation, causes, and measures to solve them.

Parameter	Expression	Unit
Redox potential close to zero	Anoxic or oxic condition	Measure DO Measure NO_X Check the inoculum activity
Excess scum and foam	System overload	Decrease the organic loading rate
Solid dragging	High flow rate	Decrease the system flow rate
Light gray colored sludge	Redox potential out of the anaerobic condition	Measure DO Measure the redox potential Check the inoculum activity
Biogas does not burn	Low methane concentration (less than 15%)	Check the inoculum activity Decrease the feeding flow rate
Accumulation of volatile fatty acis	Inhibition of methanogenesis	Decrease the feeding flow rate Check for changes in the substrate for the possible presence of inhibitors agents
Lack of alkalinity	Substrate quality	Supplement the alkalinity
Low pH	Accumulation of VFA	Decrease the flow rate Adjust the alkalinity
Temperature below the recommended operating condition	Heating system failure	Inspect the heating system
Sudden reduction in biogas production	System overload or underload Presence of inhibitory agents	Check the flow rate Check the S concentration in the substrate Evaluate whether there was a change in the substrate characteristic
High concentration of fixed solids in the sludge (> 50%)	Substrate characteristic	Controlled sludge disposal Substrate pre-treatment for FS removal

DO= Dissolved oxygen; $NO_x = NO_2^- + NO_3^-$; VFA = volatile fatty acis.

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