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Acidogenic and Methanogenic activities in Anaerobic Ponds

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Abstract— Anaerobic ponds are used rather often in hot climates. But the design of those ponds is still based on empiric rules. As for other types of extensive systems we should try to progress to the development of mathematical models describing in details the behaviour of such bioreactors.In the case of anaerobic ponds the two main processes of treatment (settling and biological activities) should be quantified separately. Moreover one should know more precisely where the biological activity mainly occurs: in the liquid phase or in the sediments accumulated on the bottom of the ponds. We focalised on the measurement and on the comparison of biological activities in the liquid layer and in the sediments. Acidogenic and methanogenic kinetics were quantified on biomasses sampled in a full scale anaerobic pond located in Tunisia. A study of the increase and decrease of acetic acid on the acidogenic and methanogenic phase respectively was carried out in closed glass bottles (500ml) maintained at mesophilic temperature (37°C).The aim of this study is to determine the kinetic of acidogenic and methanogenic phases. Liquid and sediment of anaerobic pond were centrifuged to collect biomass which was conserved in a physiological liquid in anaerobic conditions. We defined a methodology for kinetic measurement of acidogenic and methanogenic phases. From the calculated specific activities the respective contributions of the liquid layer and sediments can be calculated which provides precious indications for the modelisation of anaerobic ponds. Global values are also compared to values issued from literature.

Index Terms— acidogenic, anaerobic pond, biomass, kinetic, methanogenic, liquid and sediment layer.

1 INTRODUCTION

Anaerobic ponds are used for treatment of concentrated organic wastewater from domestic and industry especially in hot climates. Its design is based on empiric rules and consideration is given to the organic volumetric load. Anaerobic ponds are rather efficient on the elimination of organic matter based on sedimentation and anaerobic degradation.The anaerobic digestion is a complex process in which organic matter is converted into a mixture of methane and carbon dioxide. The overall conversion is carried out by a mixture of microorganisms through several biochemical reactions in series and in parallel [1]. Anaerobic reactions taking place in the sediment include solubilization of biodegradable particulate matter followed by acidogenesis, acetogenesis and methanogenesis. Generally the reactions occurring in the liquid layer are often neglected and are attributed to the convection movements and gas bubble (methane production) in the bottom related by the sediment layer.The anaerobic conversion of organic matter leads to the intermediate formation of volatile fatty acids (VFA), mainly butyrate, propionate and acetate, subsequently butyrate and propionate degradation due to syntrophic metabolism, results in the production of acetate. Acetate is further degraded by acetoclastic methanogens into CO_2 and CH_4, acidogenic activity is usually higher than methanogenic rate because it takes place first and its product (acetate) is used during methanogenesis.

The substrate used for acidogenic activity determination is usually glucose, which is considered as the main intermediate in the pathway of anaerobic digestion of carbohydrate complex organic [2]. About methanogenic activity determination VFA were used, especially acetic acid.

However few studies deal with the kinetics parameters of acidogenic and methanogenic activity in anaerobic ponds.

Accordingly, the work presented here assesses the biological activity both in liquid phase and in sediments accumulated at the bottom of an anaerobic pond. So we defined a methodology for kinetic measurement of acidogenic and methanogenic phases.

Acidogenic and methanogenic kinetics characterised by the increase (production of HAc) and decrease (consumption of HAc) of acetic acid respectively, were quantified on biomasses samples in a full scale anaerobic pond located in Tunisia.

2 MATERIALS AND METHOD

This study deals with the anaerobic waste stabilisation pond of the experimental plant of INRST (National Institute of Scientific and Technical Research) in Tunisia. We focalised on the measurement and on the comparison of biological activities in the liquid layer and in the sediments from anaerobic pond. This plant is in operation since 1992. Wastewater is settling in a primary settling tank and them distributed in the other ponds. The wastewater treatment system consists of 5 ponds connected in series. Out of these one is anaerobic pond, follows by facultative pond and three maturation ponds. This station received surfacic load in BOD_5 of 2930 kg/ha/d and a flow of 45 m^3/d for the anaerobic pond. Our study is only on the anaerobic pond and its main characteristics are: surface area (7.22*4.10) m^2; depth 3.25 m and 96 m^3 as volume. The removal efficiencies of the anaerobic pond only is 32%, 12%, 14% and 28% respectively for SS, soluble COD, raw COD and

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raw BOD, for a theoretical residence time of 27 days.

2.1 Centrifugation of biomass

Two liters of water from the liquid layer and 25 ml of sediment of anaerobic pond were sampled and centrifuged at 3000 rpm during 15 minutes separately. The collected biomass were rinsed out twice two times with physiological liquid and centrifuged again. Then biomass from liquid layer and sediment layer were kept separately in 500 ml glass bottles filled with physiological liquid. We injected nitrogen gas in each glass bottle to maintain anaerobic condition. 25 ml of each glass bottle were analysed for SS and VSS to evaluate the kinetic per gram of dry biomass.

2.2 Operating procedure

- Introduction into a new glass bottle (500 ml) of 150 ml of glucose (1.5g/l) and 150 ml of acetic acid (0.1g/l) solutions as substrate for acidogenic and methanogenic phases respectively;
- Addition of 25 ml of the biomass suspension (from liquid layer or from sediment) for the measurement of acidogenic or methanogenic activity in liquid or sediment layer, and 125 ml of physiological liquid (NaCl at 9g/l);
- Addition of 200 ml of deaerated distilled water;
- After homogenisation by shaking the glass bottle, nitrogen gas was bubbled to maintain anaerobic condition;
- Then the glass bottle was closed down with septum and conserved at 37° C.

2.3 Monitoring

Production (acidogenic phase) or consumption (methanogenic phase) in liquid or sediment layer from anaerobic pond of acetic acid was monitoring just after the operating procedure. Samples were collected every hour, centrifuged at 3000 rpm during 15 minutes and 0.5 ml of the supernatant was used for measurement of acetic acid (HAc).

2.4 Analyticals procedures

Parameters including SS, VSS, T°C, O_2 were analysed according to Standard Methods [3].

The kinetics of acidogenic and methanogenic were assessed and acetic acid was measured by the colorimetric method described by CEBEDEAU [4]. The principle of this spectrophotometric method is based on the variation of the solution coloration which is proportional to the AGV concentration. The AGV react with ethylene glycol and concentrated sulfuric acid to form esters in condition well define of temperature (100° C) and time (3mn); the esters formed give the hydroxymic acids and in presence of ferric chloride ($FeCl_3$) colour the solution which is read at 500 nm of optical density. The measurement is done tree times for each sample and the average represents the exact value of HAc express in equivalent HAc.

3 Results and Discussion

The methodology and procedures described above were applied to determine the kinetic parameters. In this section we give the results of acidogenic and methanogenic activities in liquid and sediment layer of anaerobic pond. Examples of measurements are provided on Fig. 1 and Fig. 2.

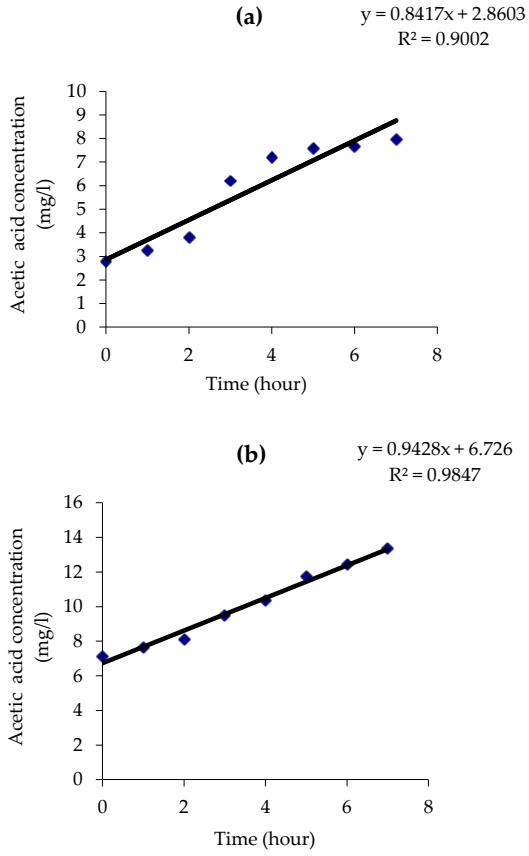

Fig. 1: Graphs of acetic acid production in liquid biomass (a) and sediment biomass (b) (Acidogenic Activity/ substrat: glucose (1.5 g/l))

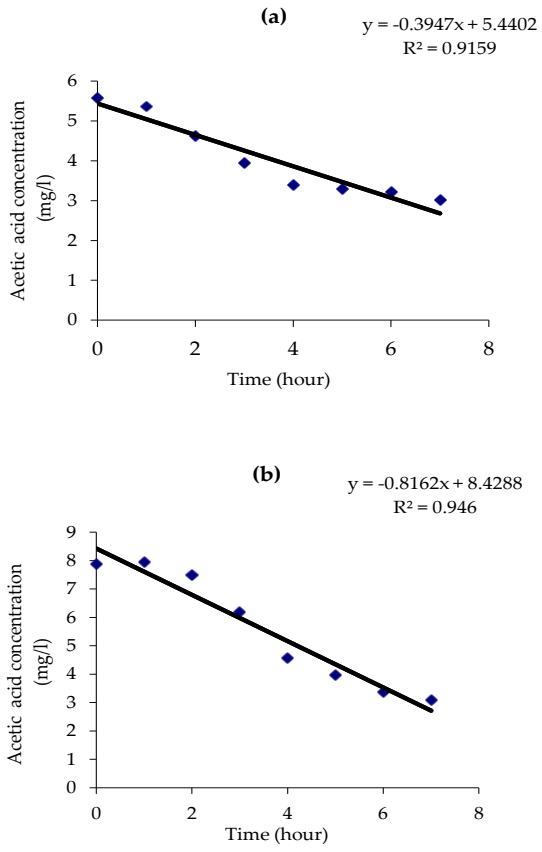

Fig. 2: Graphs of acetic acid consumption in liquid biomass (a) and sediment biomass (b) (Methanogenic Activity/ substrat: acetic acid: (0.1 g/l))

TABLE 1
PARAMETERS MEASUREMENT IN LIQUID SEDIMENT LAYYERS OF ANAEROBIC POND

Step	Substrate	Concentration (g/l)	Activities (mg HAc./l/h)	(gCOD/gSS/d)	(gCOD/gCOD/d)
Acidogenic (liquid biomass)	glucose	1.5	0.84	0.093	0.065
Acidogenic (sediment biomass)	glucose	1.5	0.94	0.048	0.034
Methanogenic (liquid biomass)	acetic acid	0.1	0.39	0.043	0.030
Methanogenic (sediment biomass)	acetic acid	0.1	0.81	0.041	0.029

TABLE 2
COMPARISON OF KINETIC PARAMETERS OF ACIDOGENIC AND METHANOGENIC ACTIVITIES (THIS STUDY AND LITERATURE)

Step	Substrate	Concentration (g/l)	Activity (g COD/g VSS/d)	References
Acidogenic (sludge)	glucose	1.5	0.174	[1]
Methanogenic (sludge)	acetic acid	0.125	0.195	[1]
Methanogenic (sludge)	acetic acid	0.25	0.394	[1]
Acidogenic (sludge)	Soluble matter	-	0.426-0.994	[2]
Methanogenic (sludge)	acetic acid	-	0.994-2.13	[2]
Acidogenic (pure cultures)	glucose	-	13 (max)	[3]
Acidogenic (liquid biomass)	glucose	1.5	0.138	(this study)
Methanogenic (liquid biomass)	acetic acid	0.1	0.064	(this study)
Acidogenic (sediment biomass)	glucose	1.5	0.086	(this study)
Methanogenic (sediment biomass)	acetic acid	0.1	0.074	(this study)

It should be considered that the methanogenic step rate is usually lower than the acidogenic one, especially when soluble substrates are considered [2], our data correspond to this case.

Our kinetics coefficients are smaller than those mentioned by [5], who obtained kinetics values in the range 0.426-0.994 from acidogenic activity and 0.994-2.13 gCOD/gVSS/d from methanogenic activity when organic matter in wastewater is dominated by dissolved easily degradable matter or by acetic acid respectively. Or [2] who used gas chromatography for measurement obtained results similar to our study.

This difference could be related to the physiological status of biomass in the liquid phase where the conditions (ORP for example) are probably not optimal for anaerobic activity. In our case there was also a delay between sampling of biomass and kinetic measurements.

5 Conclusions

We provide here a methodology to measure acidogenic and methanogenic activities in liquid and sediment biomass of anaerobic pond. The obtained values confirm that part of the activity is located in the liquid phase. In a further step those kinetics will be used to modelise and quantify anaerobic degradation combined with settling suspended solids in anaerobic ponds.

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