International Journal of Environmental Bioremediation & Biodegradation, 2017, Vol. 5, No. 3, 77-85
Available online at http://pubs.sciepub.com/ijebb/5/3/1

DOI:10.12691/ijebb-5-3-1

Production of Efficient Microbial Complex for Organic Fraction of Municipal Organic Solid Waste Pretreatment Upstream Anaerobic Digestion

Mahamadi NIKIEMA[1,*], Marius K. SOMDA[1], Kifouli ADEOTI[2], Désiré TRAORE[1], Farid BABA-MOUSSA[2], Fatiou TOUKOUROU[2], Dayéri DIANOU[3], Alfred S. TRAORE[1]

[1]Centre de Recherche en Sciences Biologiques Alimentaires et Nutritionelles (CRSBAN), Universté Ouaga I, Pr Joseph KI-ZERBO, 03 BP 7131Ouagadougou 03, Burkina Faso
[2]Laboratoire de Microbiologie et Technologies Alimentaires (LAMITA), Université d'Abomey-Calavi, B.P. 526 Abomey-Calavi, Bénin
[3]Centre National de la Recherche Scientifique et Technologique (CNRST), 03 BP 7192 Ouagadougou 03, Burkina Faso, Ouagadougou, 03 BP 7192 Ouagadougou 03, Burkina Faso
*Corresponding author: mahamadinikiema87@gmail.com

Abstract The aim of this present study is to select high performance microbial strains for organic municipal solid waste biological pretreatment. Waste samples were collected at three municipal waste pre-collection centers in the city of Ouagadougou. Standard isolation and characterization methods have been used for strains selection in different biotopes. Waste biodegradation tests were carried out in bottle (300 mL) with 120 mL of useful volume composed of the buffer (K_2HPO_4 and NH_4Cl) and 2% of waste. Optimization tests of waste pretreatment were carried out in function of temperature and inoculum proportion (10 % and 25 %). The evolution of pH and total solid loss was monitored during fermentation. Sixteen (16) microbial strains were isolated from different matrices, including three (03) cellulolytic bacteria (CA1, CA2, CA3), three (03) *Streptomyces* sp (SS1, SS2, SS3), four (04) *Bacillus* sp (BS1, BS2, BDP, BAF), three (03) Yeasts (YBB, YDP, YEU) and three (03) molds (MS, MBB, MDP). The pH drops from 7 to 5.4 and persists until the 6th day was followed by a gradual increase in pH to 9. Temperature rise at 37 ℃ allowed a sudden acidification from the 3rd day (pH 5.6 at 30℃ and pH 4.72 at 37℃) and increases to pH 10. The CA3-SS3-BDP-YBB consortium has been identified as the best combination for a pre-fermentation of municipal waste. The TS reduction on day 25 ranged from 9.9 g/L or 49.5% of TS removal for TNS, 6.7 g/L or 33.5 % for MDP, 9.3 g/L or 46.5% for SS3, 6.3 g/L or 31.5% for YDP, 8.7 g/L or 43.5% for CA3, 7 g/L or 35% for MBB and 4.8 g/L or 24% for YBB. The optimization allowed a reduction of the pretreatment time to 4 days, obtaining a biomass adapted to anaerobic digestion.

Keywords: *Screening, microorganisms, biological pretreatment, organic waste*

Cite This Article: Mahamadi NIKIEMA, Marius K. SOMDA, Kifouli ADEOTI, Désiré TRAORE, Farid BABA-MOUSSA, Fatiou TOUKOUROU, Dayéri DIANOU, and Alfred S. TRAORE, "Production of Efficient Microbial Complex for Organic Fraction of Municipal Organic Solid Waste Pretreatment Upstream Anaerobic Digestion." *International Journal of Environmental Bioremediation & Biodegradation*, vol. 5, no. 3 (2017): 77-85. doi: 10.12691/ijebb-5-3-1.

1. Introduction

The world population will know a surprising increase from 6.7 to 9.2 billion [1]. At the same time, urban population is projected to increase from 3.3 billion in 2007 to 6.4 billion in 2050. This strong growth of urban population will be mainly due to the strong urbanization of the African and Asian countries. The corollary of this is an increasing production of municipal solid waste. According to Amoo and Fagbenle [2], from 2011, the world generates 2 billion tons of municipal solid waste (MSW) annually. These forms of municipal solid waste management are inadequate, the direct result being environmental pollution and degradation of population's living environment (microbial contamination, atmospheric air pollution by harmful odors, etc.). The organic fraction represents more than two-thirds [3,4] and could be treated under controlled conditions to reduce environmental impact and recover energy [5,6,7]. Among the systems for managing the organic fraction, treatment with aerobic digestion or anaerobic digestion is gaining renewed interest. It could be seen as an economic, decentralized and ecological solution to these problems through energy autonomy and sustainable agricultural development in rural areas [8]. Methanization becomes very delicate, in particular because of the complexity of the waste produced. According to

Eleazer et al. [9], Lacour [10] and Bayard et al. [11], the fermentable fraction of municipal waste consists mainly of leaves, branches, office paper, newspapers and food residues. It is made up of chemical compounds of different masses, sizes and chemical properties: hydrogen-carbon compounds including saccharides (soluble non-cellulosic compounds), cellulose, hemicellulose and lignin, lipid, protein and humic compounds. Pretreatment operations are then necessary to increase the anaerobic digestibility of the substrates and to improve process performance [10,12]. Indeed, mechanical, thermal, chemical and biological pretreatment forms have been developed by various authors [12,13,14]. These preparations upstream aim to accelerate the kinetics of methane production on the one hand, and to increase the rate of conversion of waste to methane on the other hand. Indeed, forms of pretreatment reported by Yadvika et al. [15] are more or less likely to be applied in developing countries. This is the case of aerobic pre-fermentation, thermochemical treatment, physico-chemical treatment. Thermo-chemical or physicochemical treatments of organic solid wastes are extremely expensive and can result in compounds such as hydroxfurfural and furfural which are highly toxic to microorganisms [12,16]. The use of microbial strains is one of the effective pretreatment techniques [16,17,18]. Many microorganisms, mainly bacteria, actinomycetes and fungi have the capacity to degrade organic matter [19,20,21,22,23]. Organic matter degradation is an extracellular process in which complex organic substances are decomposed into simple and soluble compounds. Several enzymes are involved in this process, hydrolytic enzymes which include cellulase, cellobiase, xylanase and amylase for degradation of polysaccharides to sugars, protease for protein degradation into amino acids, and lipase for degradation of lipids to glycerol and long chain fatty acids [24,25,26,28]. In this study, the ability of different microbial strains to degrade municipal organic waste was investigated to realize efficient microbial complex for biological pre-treatment the first time. Also, pretreated product was characterized.

2. Materials and Methods

2.1. Sampling and Sample Preparation

Sampling of municipal solid waste was carried out in three (3) pre-collects centers of Ouagadougou's town. For each tank, three (3) trash repetitions were performed. The first center is located in the district 2 (North latitude: 12° 22'; West longitude: 1° 32' and altitude: 335 m), the 2nd in the district 12 (North latitude: 12° 19', West longitude: 1° 31' and altitude: 349 m), and the third center in the district 3 (North latitude: 12° 23'; west longitude: 1° 32' and altitude: 326 m). Waste samples were mixed, sorted and dried under the sun for 7 days, then crushed and sieved (size ≤ 1 mm). Different matrices were used for the isolation of microbial strains. These include bovine dung, waste water collected at the national slaughterhouse in Ouagadougou, Burkina Faso (12°25'5.87"N, 1°28'29.23"O). Soil samples were collected from different locations in cultivated fields.

2.2. Determination of the Physicochemical Parameters of the Organic Fraction of Municipal Waste (OFMSW) and of the Final Product

The pH, salinity, dissolved sediment rate (TDS), electrical conductivity (CE) and resistivity (R) were measured by a multi-parameter analyzer of the 9420 WTW multi-parameter type. The dry matter (DM) and were determined according to the method described by Charnay [29]. Five grams (5 g) of sample were placed in the oven at 105 ± 2℃ to a constant weight, about 24 hours. Total ash (Ct) was determined using TAPPI 211Om-02 [30]. One gram (1 g) of waste powder was introduced into a crucible and placed in an oven at 525℃ for 3 hours. The organic matter content (MO) in the sample was obtained by the difference between dry matter and total ash. The method of Nicholson et al [30] was used to determine the lignin content (Klason lignin). The lignin content was achieved with 0.1 g of OFMSW in 1.5 ml of 72 % H_2SO_4 in a 50 ml polypropylene centrifuge tube and allowed to act for 2 h at 20℃. The mixture was diluted with 56.2 ml of distilled water and filtered under vacuum using the crucible previously weighed, before washing with distilled water and drying in a ventilated oven. Organic acids content were determined using the method described by Dilallo and Abertson [31]. An amount of 25 mL of the sample was titrated with H_2SO_4 (0.1 M) at pH 4.0. The sample was then boiled lightly for 3 minutes, cooled, titrated by assay with Na_2CO_3 (0.05 M), and the titration amount was noted from pH 4.0 to 7.0.

2.3. Isolation of Microbial Strains

The cellulolytic bacteria were isolated on a modified Nfb (N-free broth) base medium containing an antifungal, cycloheximide. Cellulose refined at 5 % (m/w) was carbon source according to method described by Chastrusse [32]. Casein agar starch medium (CAA) was used for the isolation of *Streptomyces* [33]. Trypticase Soja Agar (BioMérieux SA, France) was used for isolation of *Bacillus*. The chloramphenicol sabouraud agar (Biokar, France) was used for the isolation of yeasts and molds (ISO Standard, 2007).

2.4. Morphological and Biochemical Identification of Strains

Morphological characteristics such as colony morphology and cell morphology (shape, Gram reaction and arrangement) were studies for identification [36]. Also, bacterial strain was subjected to different biochemical tests including Gram 'staining, sporulation, catalase, oxidase, starch hydrolysis, citrate utilization, manitol mobility and nitrate reduction test [34,35,36,37]. The use of sugars by strains was performed. Medium composition was: KH_2PO_4 2.38 g/L, $(NH_4)_2SO_4$ 2.64 g/L, $MgSO_4$ 1 g/L, K_2HPO_4 5.65 g/L, 1 ml of saline solution ($CuSO_4$ ($5H_2O$) 6.4 g/L, $FeSO_4$ ($7H_2O$) 1.1 g/L, $MnCl_2$ ($4H_2O$) 7.9 g/L, $ZnSO_4$ ($7H_2O$) 1.5 g/L), Agar 15 g/L pH: 7.0. The carbohydrates tested were added: glucose, mannose, maltose, arabinose, saccharose and lactose [36].

The demonstration of cellulolytic activity of bacteria was carried out by the technique described by Chastrusse [32]. The bacterial colonies were subcultured on a 3% TSA rich medium (3 g of Trypcase Soja Broth and 15 g of Agar per 1 L of deionized water) and incubated for 5 days at 30℃. The strains were again transferred onto the refined cellulose Nfb medium, and placed at 30 ℃ for two weeks. Cellulolytic activity of strains were visualized with a solution of Red Congo 2.5 % (m/w) for 20 min. The excess of unbound dye was removed and replaced with a 5M NaCl solution which remained in contact with the medium for 30 min. Finally, the NaCl was removed and the dishes were rinsed gently with water. The lysis ranges then appear discolored, that is to say yellow-orange with respect to the rest of the medium which is red.

2.5. Waste Biodegradation Test

The protocol described by Dutta et al. [37] was modified to test the biodegradation of organic waste with isolated strains. The waste medium was prepared by mixing K_2HPO_4 (2g) and NH_4Cl (2g) in 1000 ml of distilled water. The media were prepared in 300 mL flasks with 120 mL of useful volume. In each flask, 2.4 g of waste powder was introduced for a 2% charge. After 24 hours of culture, the strains were transplanted into the 250 mL Erlenmeyer flakes containing nutrient broth. The cultures were incubated for 24 hours with stirring at 65 rpm at 30℃. The waste after sterilization at 121℃ for 15 min was inoculated with cultures (10%) (v/v).

2.6. Optimization of Waste Fermentation

A combination of the efficient strains was carried out; and strains were coded CA3, SS3, BDP and YB. The protocols described by Dutta et al. [37] and Poszytek et al. [38] were adapted for the optimization of the activity of these strains. The combination was cultured in broth (yeast extract: 2.5 g/L, peptone: 5 g/L, glucose: 1 g/L, NaCl: 5 g/L) with stirring at 75 rpm for 72 hours. The culture was supplemented in 24 hours with 25 mL of cellulose 5% (v/w). The degradation tests were carried out at different temperatures (30 and 37℃).

2.7. Statistical Analysis

XLSTAT software was used for statistical analysis of data. A one-way ANOVA and Fisher's test LSD was performed to compare the mean values of the different variables. *P* value of less than 0.05 was considered to indicate statistical significance.

3. Results and Discussion

3.1. Chemical and Physical Characteristics

The organic fraction of the municipal waste sampled was mainly composed of green waste (grass, dead leaves) and food waste (kitchen waste, condiment residues from markets). The major components of this fraction are shown in Table 1. High organic matter contents (85.14%) indicate a substrate of choice for microorganisms. The high lignin composition (29.21%) could be explained by the richness of the organic fraction of municipal waste in lignocellulosic biomass such as tree leaves, branches and grasses [9]. Indeed, the secondary cell walls of the wood and herb tissues are composed mainly of cellulose, hemicelluloses and lignin [39]. Lignin is extremely resistant to biodegradation [40,41]. The C/N ratio of the sample in the order of 31.65 is favorable for biodegradation because it is very close to the optimum between 25 and 30 [42,43]. The maximum tolerated value is 35 for urban waste. Beyond this value, several cycles of oxidation are necessary to reach the optimum value [29,44].

Table 1. Main components of the organic fraction of municipal waste collected

Composition	Avarage (% of TS)
Total Solid (TS)	92.79 ± 0.09
Organic Matter (OM)	80.10 ± 0.32
Ash	17.11 ± 0.30
C/N	31.65 ± 0.05

3.2. Isolation of Microbial Strains

Table 2 shows the various isolated microorganisms and the matrices used. Five different microorganisms were isolated. Three types of cellulolytic bacteria and *Streptomyces* sp, two types of *Bacillus* sp were isolated. Cellulolytic bacteria and *Streptomyces* sp were isolated only from soil. Two strains of *Bacillus* were isolated from all matrices excepted from the cow dung. The maximum of microorganism was isolated from soil and putrefaction 'waste. Three strain of yeast were isolated from waste putrefaction, Bovine dung and wastewater. And three strains of mold were isolated from soil, waste putrefaction and cow dung.

3.3. Waste Biodegradation Test

3.3.1. PH Monitoring

Based on the pH-monitoring results for 20 days showed on Figure 1, initial pH of various assays is around 7, which is due to phosphate buffer used as reaction medium. The pH drops from 7 to 5.4 and persists until the 6th day. The pH gradually increases from the 6th day to 7-7.5 on the 15th day, which corresponds to the maturity of the tests. The decrease of pH is due to the fermentative activity of microorganisms [45,46]. Indeed, the decrease of pH could be due to the degradation of certain molecules, such as simple carbohydrates, starch, hemicelluloses, pectins and amino acids contained in waste releasing organic acids [28,45,46]. Cellulose, a more bulky polymer, is more resistant. Lignin and other highly aromatic aromatic polymers will be degraded later, slower and incompletely. According to Aissam [47], pH decrease could be attributed to release of H^+ ions by cells in reaction medium. This release occurs at the time of transporting antiport organic compounds from the culture medium to cells. pH increase from the 6th day could be explained by consumption of organic acids produced and hydrolytic action of bacteria on organic nitrogen forms such as proteins, urea [48,49,50]

but also as a result of reduced microorganisms activity [47,51]. Equally, reaction of denitrification during which the consumption of protons necessary to reduce nitrates or nitrites in molecular nitrogen and / or gaseous nitrogen oxide would also explains pH increase according to Berthe [52]. The results obtained show that all strains present capacity to degrade organic fraction of municipal solid waste. The strains CA3, SS3, BDP, YBB and MBB showed a high production of acids, that reflects their performance in degrading organic matter. The production of acids with TNS control was more increased; this could be explained by diversity of microorganisms in the waste. Flora action could be complementary and allow a better degradation of organic matter. Indeed, in waste there is a great diversity of microorganisms, mainly bacteria, *Actinomyces* and fungi, which are responsible for mineralization of organic matter [47,57].

3.3.2. Evolution of Total Solid Content

Figure 2 shows evolution of total solid (TS) during waste fermentation. Total solid control reveals amount of material converted to volatile matter following the biotransformation process [46,54]. The results obtained show capacity of strains to degrade organic matter contained in waste. The Figure 3 and Table 3 show respectively effect of different strain on TS % removal and reduction of TS after 25 days. The TS reduction on day 25 ranged from 9.9 g/L or 49.5 % of TS removal for TNS, 6.7 g/L or 33.5 % for MDP, 9.3 g/L or 46.5% for SS3, 6.3 g/L or 31.5% for YDP, 8.7 g/L or 43.5% for CA3, 7 g/L or 35 % for MBB and 4.8 g/L 24 % for YBB. The biodegradation experiments carried out in this study have assessed the efficiency of five different strain based exclusively on the percentage of TS removal and organic acids production. Strains CA3, SS3, BDP and YBB were retained. The TNS control showed TS reduction 9.9 g/L, which could be explained by a very wide diversity of pre-existing microorganisms in waste (Bacteria, *Actinomyces* and fungi) which consumes oxygen gas to mineralize organic matter in CO_2 and H_2O [55,56]. Few microorganisms are capable of completely mineralizing organic matter, organic by-products may be consumed directly by other microorganisms or they may subsist for a longer or shorter time in waste [57]. Bacteria and fungi had a broad spectrum of activity over a wide pH range, especially in high humidity [27,51]. They are responsible for start of degradation of organic matter. They degrade easily oxidizable products such as proteins, carbohydrates (sugars) and lipids. *Streptomyces* belong to the group of *Actinomyces* and intervene according to Crawford and Pometto [58] very late in degradation of organic matter. They attack organic substances not degraded by bacteria and fungi (chitin or tannins). Neutrophilic, they tolerate slightly basic pH and are not very competitive with other groups.

Table 2. Isolated microorganisms and indigenous sources

Microorganisms	Sources				
	Soil	Putrefaying Waste	Fermented food	Cow dung	Wastewater
Cellulolytic Bacteria	CA1, CA2 and CA3	NIM	NIM	NIM	NIM
Streptomyces ps	SS1, SS2 and SS3	NIM	NIM	NIM	NIM
Bacillus ps	BS1and BS2	BDP	BAF	NIM	NIM
Yeast	NIM	YDP	NIM	YBB	YEU
Mold	MS	MDP	NIM	MBB	NIM

NIM = Non isolated microorganism.

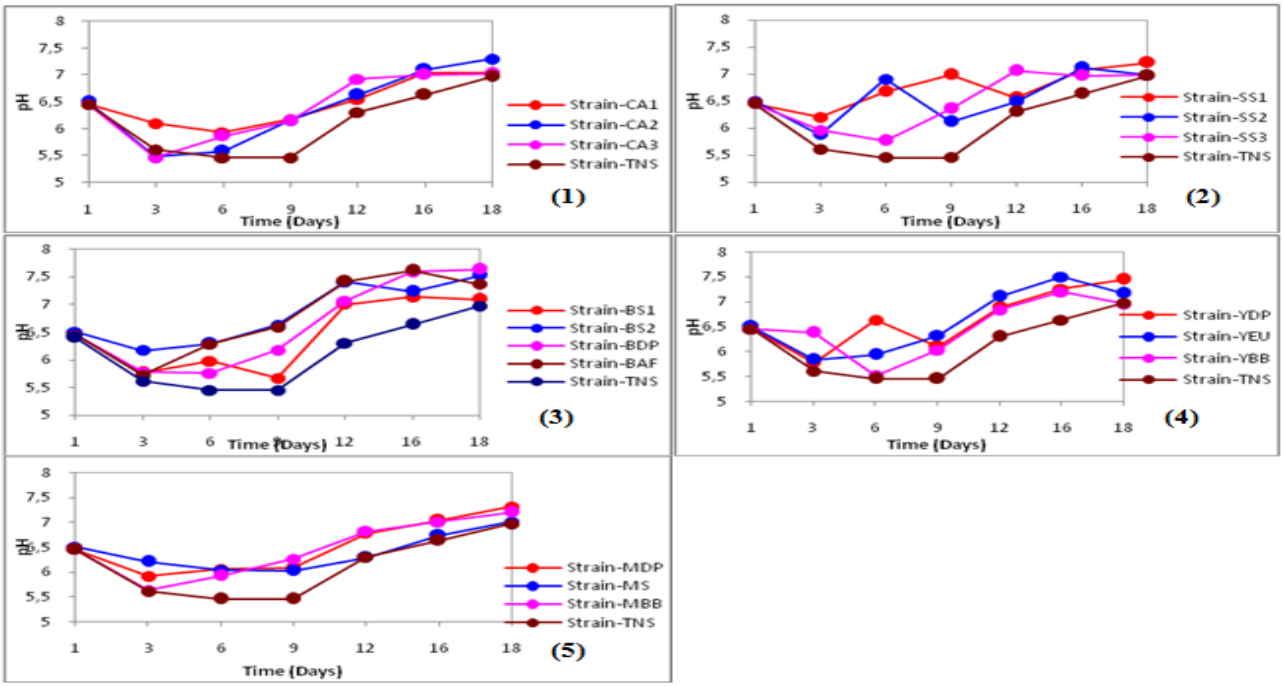

Figure 1. pH evolution according to the strains studied: (1) Cellulolytic bacteria, (2) *Streptomyces*, (3) *Bacillus* (4) yeasts and (5) Molds (TNS = Control)

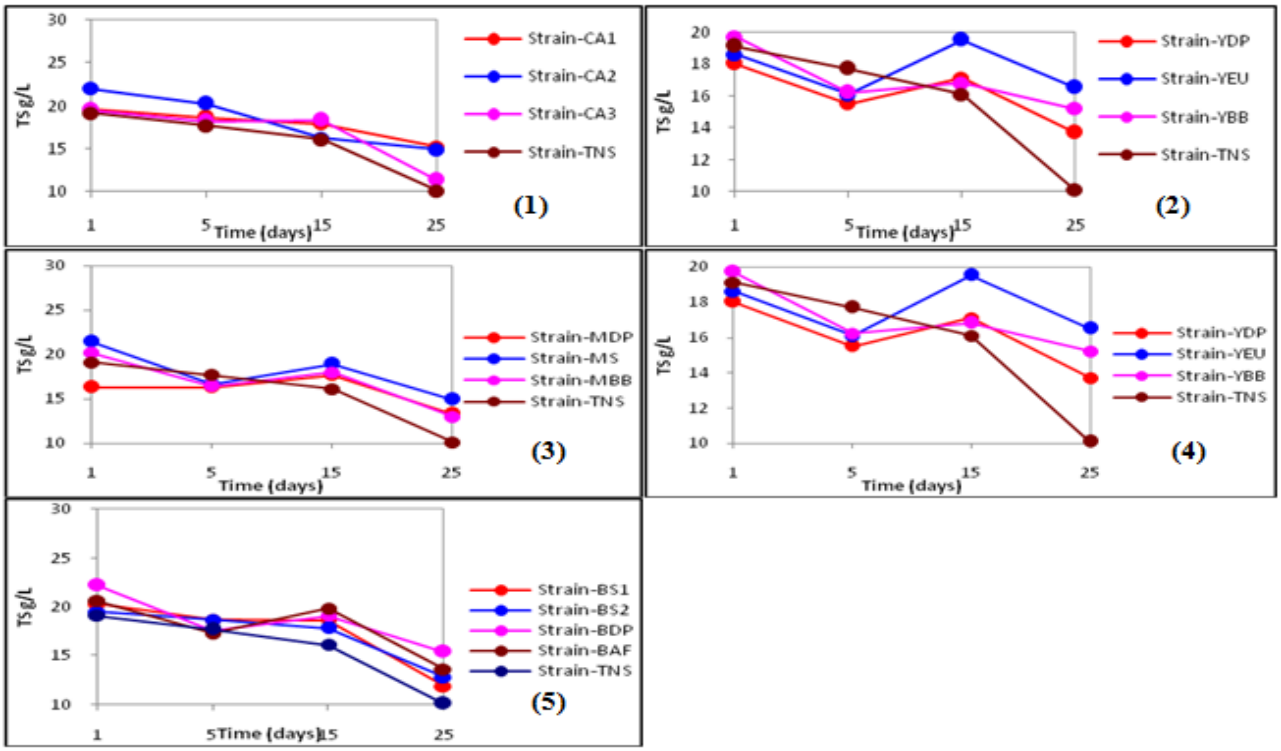

Figure 2. Total solid reduction (MS) during treatment: (1) Cellulolytic bacteria, (2) *Streptomyces*, (3) *Bacillus* (4) yeasts and (5) Molds (TNS = Control)

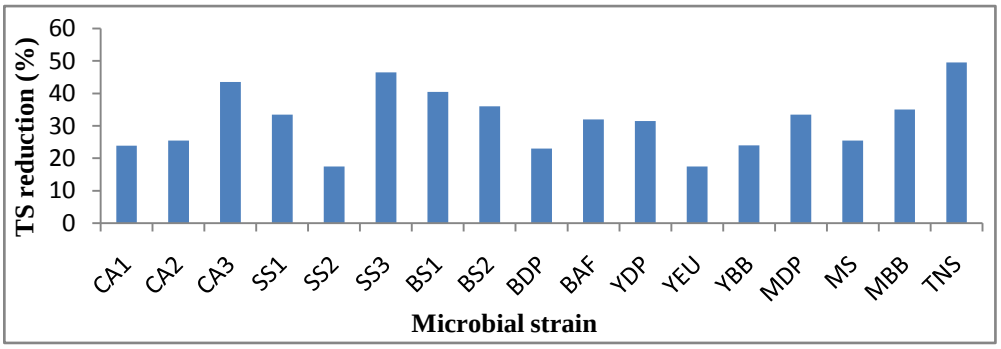

Figure 3. Effect of different strain on total TS % removal

Table 3. Total solid reduction of different strain after 25 days of treatment

Strain	TS reduction g/L
CA1	4.78
CA2	5.1
CA3	8.7
SS1	6.7
SS2	3.5
SS3	9.3
BS1	8.1
BS2	7.2
BDP	4.6
BAF	6.4
YDP	6.3
YEU	3.5
YBB	4.8
MDP	6.7
MS	5.1
MBB	7
TNS	9.9

3.3.3. Identification of Effective Strain

The characteristics of different strains are given in Table 4. CA3 was stick-shaped, filamentous, Gram positive, oxidase positive and produced spore. This strain utilized various sugars including glucose, maltose, lactose, arabinose and saccharose. It showed negative growth on acetate, starch, urea and nitrates. CA3 showed cellulolytic activity visualized using Congo red, which is in accord with Chastrusse [34]. The SS3 strain was of filamentous shell shape, Gram positive, oxidase positive and produced spore. It was able to use glucose, maltose, lactose, arabinose and saccharose. SS3 utilized starch, urea and nitrates. SS3 registered negative growth acetate. The strain BDP was found to be Gram positive, oxidase positive and produced spore. It was able to use glucose, maltose, arabinose, lactose, saccharose except mannose. BDP was capable to use other substrates such as nitrates and acetate except starch. YBB yeast strain used glucose, maltose, lactose, arabinose, mannose, manitol and saccharose. It utilized nitrates and acetate except starch. The MDP strain gave white colonies, the texture was cottony. MDP had very rapid and extensive growth. Microscopic observation showed an aspergillum head. CA3 and BDP could belong to *Bacillus* genus, SS3 to *Streptomyces* genus, YBB could belong to *Candida* genus and MDP strain could belong to *Aspergillus* genus.

Table 4. Biochemical characteristic of strains

Microbial strain	CA3	SS3	BDP	YBB	MBB
Glucose	+	+	+	+	+
Maltose	-	+	+	+	+
Lactose	+	+	+	-	-
Arabinose	+	+	+	+	+
Mannose	-	+	-	+	+
Manitol	-	+	+	+	+
Saccharose	+	+	+	+	+
Citrate	-	-	-	-	+
Urée	-	+	+	-	+
Starch	-	+	+	-	+
Nitrate	-	-	+	-	+
Acetate	-	-	-	-	+
Catalase	+	+	+	+	+
Motility	-	-	+	-	+
Spore formation	+	+	+	+	+
Gram's reaction	Gram+	Gram+	Gram+	……	…….
Cell shape	rod	Coccus	rod	Elongated	…….
Form	Filamentous	Filamentous	isolated	isolated	Aspergillum head
Presumptive	*Bacillus* sp.	*Streptomyces* sp.	*Bacillus* sp.	*Candida* sp.	*Aspergillus* sp.

Responses: + Positive; - Negative; Reference: API 20 E V4.1 and API 20 C AUX V4.0.

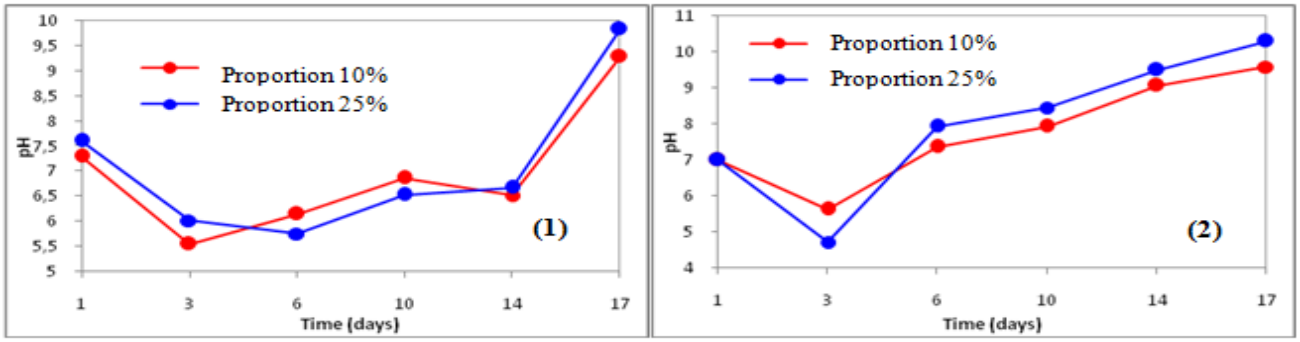

Figure 4. pH evolution in function of temperature with consortium CA3-SS3-BDP-YBB: (1) 30℃ and (2) 37℃

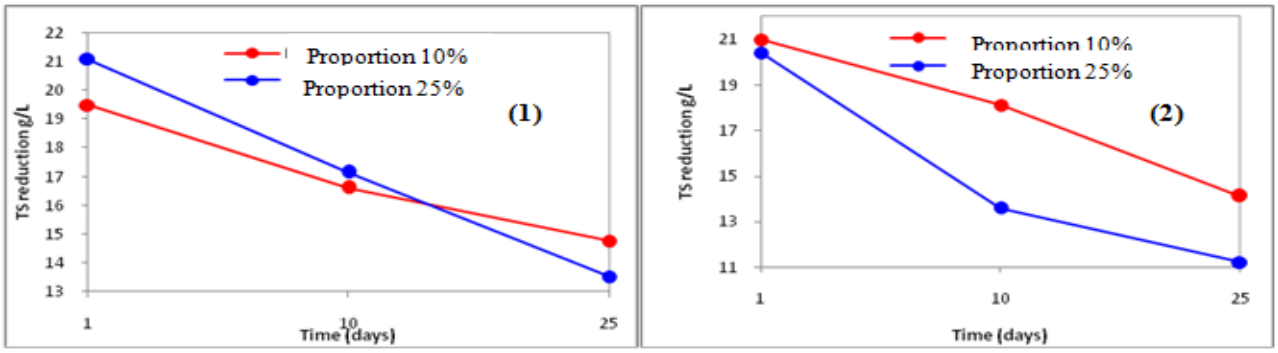

Figure 5. Evolution of TS reduction in function of temperature with consortium CA3-SS3-BDP-YBB: (1) at 30℃ and (2) 37℃

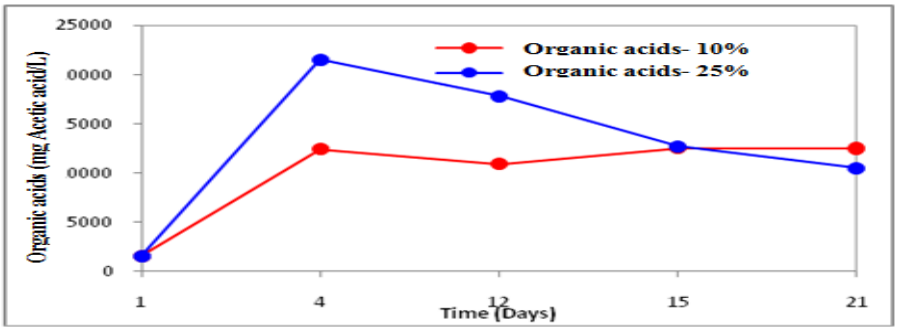

Figure 6. Evolution of organic acids during fermentation with 10% and 25% of inoculum

3.4. Optimization of Waste Biodegradation

pH evolution during fermentation with consortium CA3-SS3-BDP-YBB proportions 10 % and 25 % is shown in Figure 4. pH evolution with 10 % and 25 % of inoculum did not show a significant difference at 30℃ ($p > 0.05$). The pH drop from 7 to 5.75 until the 6th day was followed by its gradual increase about 9. Temperature increase at 37℃ resulted a high acidification after day 3 (pH 5.6 for 30℃ and 4.08 for 37℃). Analysis of pH variance during pretreatment as function of inoculums proportion showed significant differences at 10 % and 25% inoculum at 37℃ ($p < 0.05$). This difference is not perceptible at 30℃. This indicates that temperature has effect on organic matter biodegradation. Total solid reduction was 5.25 g/L (26.25 % of organic matter reduction) with 10 % of inoculums (Figure 5a), 6.5 g/L with 25 % at 30 ℃ (32.5 %). At 37 ℃, TS reduction was 5.9 g/L (29 %) for 10 % inoculums and 8.8 g/L for 25 % (44 %). Evolution of organic acids in function of inoculums proportion (10% and 25%) is given in Figure 6. Inoculums quantity has significant effect on organic acids production the 4th days ($P < 0.05$). Initially organic acids concentrations increased rapidly to maximum value of 12400 acetic acid/L for 10% and 21500 mg acetic acid/L for 25%. According to Ugwuanyi et al. [63] accumulation of organic acids in aerobic system is considered evidence of fermentative metabolism. Similar variations in total organic acids production patterns were observed by Parawira et al. [59] and Suchowska-Kisielewicz et al. [65] when they studied the production of organic acids from vegetable, tea, solid potato and municipal solid wastes. Mshandete et al. [60] was found initially the concentrations increased rapidly to maximum value of 12 g/l during 84 h for untreated waste during anaerobic digestion, in case the high organic acids production affects the performance of the subsequent methane bioreactor. Pretreatment have effect on hydrolases and VFAs profiles during acidogenic phase of anaerobic digestion and is suitable both during start-up and subsequent process stability [60].

3.5. Characteristics of Product Pretreated

After 25 days of fermentation of the organic solid waste, the pH profile and the loss in total solid made it possible to determine an optimal time of biodegradation which was four (04) days at 37 ℃. The pH was 6.55 and 4.08 respectively for 10 % and 25 % of inoculums. The total solid reduction on day 4 was 1.825 g/L for 10 % of inoculums and 1.725 g/L for 25 % of inoculums.

The total solid (TS) of the product after fermentation was 1.82 % to 10 % inoculums and 1.72 % to 25 % of inoculums. Better biogas production has been found with total solid ratios of between 2 and 4 % on bovine dung, food waste and jatropha oil residues (68). The mean values of some physicochemical parameters of the final product in terms of salinity, total dissolved sediment (TDS), electrical conductivity (EC) and resistivity were respectively 3.91 g/L, 7.03 g/L, 6, 45 mS/cm, 105.5 Ω.cm to 10 % of inoculums and 3.55 g/L, 6.42 g/L, 6.45 mS/cm 116.2 Ω.cm to 25 %). Indeed, the authors have expressed influence of these parameters on anaerobic digestion [62,63,64,65]. The results obtained was in agreement with those obtained by Manonmani et al. [61], which found 3.88 - 11.9 mS/cm of conductivity, 2.5 - 8.92 g/L of salinity, 2.89 - 6.98 of TDS and 6.3 -8.1 of pH. Also Zeng et al. [66] found similar TDS values of 6.80 g/L but a high conductivity of 13.61 mS/cm in the animals dung, the preferred substrate for anaerobic digestion. In a study about industrial wastewater anaerobic digestion, Graterol [67] found similar pH, conductivity and TDS values between 6.05 - 6.97, 2.58 - 4.28 and 1613 - 2675 mg/L at the stability point of the process.

Table 5. Physico-chemical parameters of the product after 4 day treatment

Parameter	Inoculum proportion	Avarage
pH	10%	6.55
	25%	4.08
TS g/L	10%	1.825
	25%	1.725
Salinity (g/L)	10%	3.91 ± 0.51
	25%	3.55 ± 0.65
TSD (g/L)	10%	7.03 ± 0.89
	25%	6.42 ± 1.12
EC (mS/cm)	10%	7.02 ± 0.87
	25%	6.45 ± 1.15
R (Ω*cm)	10%	105.5 ± 14.16
	25%	116.2 ± 23.65
Organic acids (mg acetic acid /L)	12400	21500

TS= Total Solid; TSD = Total Solid Dissolved; EC= Electrical Conductivity; e= Resistivity.

4. Conclusion

This work allowed realizing a microbial consortium presenting a very wide spectrum of organic waste degradation that will be used in biological pre-treatment upstream of anaerobic digestion. This is complex consisting in a combination of cellulolytic bacteria (CA3) of *Bacillus* genus, *Streptomyces* sp (SS3), *Bacillus* sp (BDP) and *Candida* sp (YBB). MBB molds of the genus *Aspergillus* also exhibit a broad spectrum of biodegradation. The optimization of biodegradation by varying the temperature and the proportion of inoculums allowed a reduction of the pre-fermentation time at four (04) days and a partially digested stable product. The finished product is a preferred substrate for launch of anaerobic digestion.

Acknowledgements

We thank the West African Research Association (WARA) and UEMOA through the CRSBAN / University Ouaga I Prof. Joseph KI-ZERBO for their financial support.

Conflict of Interest Statement

We declare that we have no conflict of interest.

List of Abbreviations

MSW= Municipal Solid Waste
TS= Total Solid;
TSD= Total Solid Dissolved;
EC= Electrical Conductivity
R= Resistivity
VFAs = Volatile Fatty Acids

References

[1] United Nations, World Urbanization Prospects The 2007 Revision Highlights. Vol. ESA/P/WP/2, Department of Economic and Social Affairs, 2007. http://www.un.org/esa /pop ulation/unpop.htm.

[2] Amoo OM, Fagbenle R. Renewable municipal solid waste pathways for energy generation and sustainable development in the Nigerian context, *International Journal of Energy and Environmental Engineerin*, 4(1): 42. July 2013.

[3] Sérémé, A, Mey, P. Valorisation agricole des ordures ménagères en zone soudano-sahélienne: cas de la ville de Bobo Dioulasso, *Revue du CAMES Science et Médecine*, Série A vol 4, 2006.

[4] Mbouaka, E. M., Étude de l'éfficacité agronomique des composts des ordures ménagères au Burkina Faso: cas de la ville de ouagadougou, p79, Juin 2000.

[5] Menikpura, S. N. M, Basnayake, B.F.A, New applications of 'Hess Law' and comparisons with models for determining calorific values of municipal solid wastes in the Sri Lankan context, *Renewable Energy*, 34:1587-1594, November 2009.

[6] Mao, C., Feng, Y., Wang, X., Ren, G. Review on research achievements of biogas from anaerobic digestion. *Renewable Sustain Energy Revew*. 45: 540-455, 2015.

[7] Cooper, C. D., Kim, B., Macdonald, J., Estimating the Lower Heating Values of Hazardous and Solid Wastes Estimating the Lower Heating Values of Hazardous and Solid Wastes, *Journal of the Air & Waste Management Association*, 49: 4, 471-476, June 2017.

[8] Tou I, Touzi SIA, Production de Biométhane à Partir des Déjections Animales, 103-108, 2001.

[9] Eleazer, W.E., Odle, W.S., Wang, Y., Barlaz, M.A. Biodegradability of Municipal Solid Waste Components in Laboratory-Scale Landfills, *Environmental Science & Technology*, 31(3): 911-917, 1997.

[10] Lacour, J. Valorisation de résidus agricoles et autres déchets organiques par digestion anaérobie en Haïti, Mémoire de thèse, p217, 2012. https://tel.archives-ouvertes.fr/tel-00825479/document

[11] Lacour, J., Bayard, R., Emmanuel, E., Gourdon, R. Evaluation du potentiel de valorisation par digestion anaérobie des gisements de déchets organiques d ' origine agricole et assimilés en Haïti. *Déchet -Revue Francophone d'Ecologie Industrielle,* 60: 31-41, 2011.

[12] Boulanger, A. Préparation d' un déchet ménager pour l ' optimisation du potentiel et de la cinétique méthanogène, Mémoire de thèse, Institut des Sciences et Industries du Vivant et de l'Environnement AgroParisTech, p316, 2011.

[13] Angelidaki I, Mogensen AS, Ahring BK. Degradation of organic contaminants found in organic waste, *Biodegradation*, 11(6): 377-383, 2000.

[14] Hansen, T.L., Jansen, J.C., Davidsson, A., Christensen, T.H. Effects of pre-treatment technologies on quantity and quality of source-sorted municipal organic waste for biogas recovery. *Waste Management*, 27:398-405, May 2007.

[15] Yadvika, Santosh, Sreekrishnan, T.R., Kohli, S., Rana, V. Enhancement of biogas production from solid substrates using different techniques — a review, 95: 1-10, March 2004.

[16] Bisaria, V.S., Ghose, T.K. Sorption characteristics of cellulases on cellulosic substances, in: Bioconversion of Cellulosic Substances into Energy, Chemicals and Microbial Protein. *In: symposium proceedings, (Editor: Ghose, TK), IIT, New Delhi*, 155-164, 1978.

[17] Hamdi, M., Gareia, J.L., Ellouz, R. Integrated biological process for olive mill wastewaters treatment. *Bioprocess Ingineering*, 8:79-84, 1992.

[18] Hamdi, M., Garciab, J.L. Anaerobic Digestion of Olive Mill Wastewaters after Detoxlfication by Prior Culture of Aspergillus, *Process Bichemistry*, 28: 155-159, 1993.

[19] Schober, G., Trosch, W. Degradation of digestion residues by lignolytic fungi. *Water Ressource*, 34(13): 3424-3430, 2000.

[20] Fourti, O., Jedidi, N., Hassen, A. Comparison of methods for evaluating stability and maturity of co-composting of municipal solid wastes and sewage sludge in semi-arid pedo-climatic condition, *Natural Science*,3(2):124-135, 2011.

[21] Kumar A, Bisht BS, Joshi V., Dhewa T. Review on Bioremediation of Polluted Environment: A mTool. *International Journal of Environmental Sciences*, 1(6): 1079-1093, March 2011.

[22] Gautam SP, Bundela PS, Pandey AK, Awasthi MK, Sarsaiya S. Diversity of Cellulolytic Microbes and the Biodegradation of Municipal Solid Waste by a Potential Strain. International Journal of Microbiology, p12, Octobre 2012.

[23] Neumann, D., Heuer, A., Hemkemeyer, M., Martens, R., Tebbe CC. Importance of soil organic matter for the diversity of microorganisms involved in the degradation of organic pollutants, *International Society for Microbial Ecology*, 8(6): 1289-1300, January 2014.

[24] Pathak, V.M., Navneet. Review on the current status of polymer degradation: a microbial approach, *Bioresour Bioprocess*, 4:15, 2017.

[25] Kirk, T.K., Farrell, R.L. Enzymatic Combustion: The microbial degradation of lignin, 465-505, 1987.

[26] Bayer, E.A., Shimon, L.J.W., Shoham, Y., Lamed, R. Cellulosomes — Structure and Ultrastructure, *Journal of Structural Biology*, 124: 221-234, November 1998.

[27] Sinsabaugh, R., Antibus, R.K., Linkins, A.E. An enzymic approach to the analysis of microbial activity during plant litter decomposition An enzymic approach to the analysis of microbial activity during plant litter decomposition, *Agriculture, Ecosystems and Environmental*, 34:43-54, July 1991.

[28] Tuomela, M., Vikman, M., Hatakka, A., Itavaara, M. Biodegradation of Lignin in a Compost Environment: A Review. *Bioresource Technology*, 72: 169-183, 2000.

[29] Batstone, D.J., Keller, J., Angelidaki, I., Kalyuzhnyi, S. V., Pavlostathis, S.G., Rozzi, A. The IWA Anaerobic Digestion Model No 1 (ADM1). *Water Science Technology*, 45(10):65-73, 2002.

[30] Charnay, F. Compostage des déchets urbains dans les Pays en Développement : élaboration d'une démarche méthodologique pour une production pérenne de compost, Mémoire de thèse, Universite de limoges ecole doctorale, p228, 2005.

[31] TAPPI (Technical Association of the Pulp and Paper Industry), Ash in Wood and Pulp, T. CM-86, 2002.

[32] Nicholson, D.J., Leavitt, A.T., Francis RC. Cellulose chemistry and technology a three-stage klason method for more accurate determinations of hardwood lignin content. *Cellul. Chem. Technol.*, 48(12): 53-59, 2014.

[33] DiLallo, R., Albertson, O.E. Volatile acids by direct titration. Water Environ, 33(4):356-365, 2014.

[34] Chastrusse C. Localisation et diversité de bactéries celllulolytiques lors d'un cycle de dessiccafion/réhumectation, ORSTOM Dakar, 1998.

[35] Sharma D, Kaur T, Chadha BS, Manhas RK. Antimicrobial Activity of Actinomycetes Against Multidrug Resistant Staphylococcus aureus, E. coli and Various Other Pathogens. *Tropical Journal of Pharmaceutical Research*,10 (6):801-808, December 2011.

[36] Holt, J.G., Krieg, N.R., Sneath, P.H.A., Staley, J.T. W.S. Genus Acetobacter and Gluconobacter. Bergey's Manual of Determinative Bacteriology. *In: 19th edn Williams and Wilkens, MD, USA*, 71-84, 1994.

[37] Carr, F.J., Chill, D., Maida, N. The Lactic Acid Bacteria: A Literature Survey. *Critical Revew Microbilogy*, 28(4): 281-370, July 2002.

[38] Cisse, H., Savadogo, A., Taale, E., Tapsoba, F., Guira, F., Traore. Y. Influence des substrats carbonés et minéraux sur l ' activité des substances BLIS (Bacteriocin-Like Inhibitory Substances) produites par des souches de Bacillus isolées à partir d ' aliments fermentés au Burkina Faso, *Journal of Applied Biosciences*, 106: 10236-10248, October 2016.

[39] Rehman, H.U., Siddique, N.N., Aman, A., Nawaz, M.A., Baloch, A.H., Qader, S.A.U. Morphological and molecular based identification of pectinase producing Bacillus licheniformis from

rotten vegetable, *Academy of Scientific Research and Technology*, 13(2): 139-144, 2015.

[40] Mandimbisolofo, V.N., Marson, R., Daniel, R. Contribution à l ' étude des propriétés biologiques des extraits de feuilles de senecio faujasioïdes (Asteraceae) Contribution to the survey of the biologic properties of the excerpts of leaves of senecio faujasioïdes (Asteraceae), 33-40, 2013.

[41] Pridham, T.G., Gottlieb, D. The utilization of carbon compounds by some determination. *University of Illinois, Urbana, Illinois*, 56:107-114, 1948.

[42] Dutta, M., Chamendra, N., Pai, S.G., Pramod, T. Original Research Article Isolation and screening of agro-waste substrates for protease production through solid state fermentation. *International Journal of Current Microbiology and Applied Sciences*, 3(3): 774-781, 2014.

[43] Poszytek, K., Ciezkowska, M., Sklodowska, A. Microbial Consortium with High Cellulolytic Activity (MCHCA) for Enhanced Biogas Production. *Frontiers in Microbilogy*, 7:1-11, March 2016.

[44] Ghaffar, S.H., Fan, M. Structural analysis for lignin characteristics in biomass straw Structural analysis for lignin characteristics in biomass straw. *Biomass and Bioenergy*, 1-16, August 2013.

[45] Ruiz-dueñas, F.J., Martínez, Á.T. Review Microbial degradation of lignin: how a bulky recalcitrant polymer is efficiently recycled in nature and how we can take advantage of this. *Microbial Biotechnology*, 2(2):164-177, 2009.

[46] Skyba, O., Douglas, C.J., Mansfield, D. Syringyl-Rich Lignin Renders Poplars More Resistant to Degradation by Wood Decay Fungi. *Applied and Environnmental Microbiology*, 79(8):2560-2571, February 2013.

[47] Ostrem, K. Greening waste: anaerobic digestion for treating the organic fraction of municipal solid waste. Dep Earth Environ Eng Fu Found Sch Eng Appl Sci Columbia Univ.;(The Earth Engineering Center and the Henry Krumb School of Mines): 1-59, 2004.

[48] Culot M. Filières de valorisation agricole des matières organiques. *Faculté universitaire des sciences agronomiques*, p73, 2005.

[49] Lacour, J. Valorisation de résidus agricoles et autres déchets organiques par digestion anaérobie en Haïti, *Université Quisqueya (Haïti)*, 2012. http://theses.insa-lyon.fr/publication/2012ISAL0026/these.pdf.

[50] Akin, H. Evolution du pH pendant la fermentation alcoolique de moûts de raisins: modélisation et interprétation métabolique, Mémoire de thèse, *Institut National Polytecnique de Toulouse*, 2008.

[51] Taiek, T., Boutaleb, N., Bahlaouan, B., El jaafari, A., Khrouz, H., Safi, A. et El antri, S. Valorisation de déchets de poisson alliés à des rejets brassicoles en vue d'obtenir un biofertilisant, *Déchets Sciences et Techniques*, 64:24-30, 2014.

[52] Aissam, H. Etude de la biodégradation des effluents des huileries (margines) et leur valorisation par production de l'enzyme tannase, *Université Sidi Mohamed Ben Abdellah*, p155, 2003.

[53] Fadil, K., Chahlaoui, A., Ouahbi, A., Zaid, A., Borja, R. Aerobic biodegradation and detoxification of wastewaters from the olive oil industry, *International Biodeterioration & Biodegradation*, 51:37-41, 2003.

[54] Fountoulakis, M., Dierssen, M., Lubec, G. Proteomic analysis of the fetal brain, *Proteomics*, 2:1547-76, 2002.

[55] Lornage R el. Comparaison de trois filieres de stockage d ' ordures menageres: Etude du comportement des déchets en pilotes semi industriels et caractérisation des émissions liquides et gazeuses associées, *Université Claude Bernard - Lyon I*; 2007. https://tel.archives-ouvertes.fr/tel-00133769.

[56] Albrecht, R. Co-compostage de boues de station d'épuration et de déchets verts: nouvelle méthodologie du suivi des transformations de la matière organique, *Université Paul Cezanne Aix-Marseille III*, p189, 2007.

[57] Berthe C. Etude de la Matière Organique contenue dans des lixiviats issus de différentes filières de traitement des déchets ménagers et assimilés. Université de Limoges; 2006.

[58] Douma, N.T. Valorisation par compostage des résidus solides urbains de la commune de Chlef, Algérie, *Université de limoge/université de blida*, 2013.

[59] Lourhzal, W., Tahri, E.H., Faïd M. Ensilage des déchets de poisson et essai d ' alimentation sur les rats. *Actes Inst Agron Vet.*, 23(1):15-20, Janvier 2003.

[60] Yulipriyanto, H. Emission d'effluents gazeux lors du compostage de substrats organiques en relation avec l'activité microbiologique (nitrification/dénitrification), *Université de Renne I*, 2001. https://tel.ar chives-ouvertes.fr/tel-00654701.

[61] De Araujo Morais, J.J. Influence des pré-traitements mécaniques et biologiques des Ordures Ménagères Résiduelles (OMR) sur leur comportement bio-physico-chimique en Installation de Stockage de Déchets (ISD), *Institut National des Sciences Appliquées de Lyon*, 2006.

[62] Bardos, P. Review Composting of Mechanically Segregated Fractions of Municipal Solid Waste – A Review. SITA Environ Trust, p143, November 2014.

[63] Crawford, D.L., Pometto, L. Lignin Degradation by Streptomyces viridosporus: Isolation and Characterization of a New Polymeric Lignin Degradation Intermediatet., *Applied and Environmental Microbiology*, 45(3):898-904, March 1983.

[64] Ugwuanyi, J.O., Harvey, L. M. M. B. Effect of aeration rate and waste load on evolution of volatile fatty acids and waste stabilization during thermophilic aerobic digestion of a model high strength agricultural waste. *Bioressource Technology*, 96:721-30.

[65] Parawira W, Murto M, Zvauya R, Mattiasson B. Comparative performance of a UASB reactor and an anaerobic packed-bed reactor when treating potato waste leachate, *Renewable Energy*, 31:893-903, 2006.

[66] Jedrczak, M.S.A. Effect of aerobic pretreatment of waste on the rate of anaerobic treatment processes. *Journal Mater Cycles Waste Management*, 15:138-45, 2013.

[67] Mshandete, A, Murto M, Kivaisi AK, Rubindamayugi MST, Mattiasson B. Influence of recirculation flow rate on the performance of anaerobic packed-bed bioreactors treating potato-waste leachate. *Environmental Technology*, 25(8):929-36, 2004.

[68] Manonmani, P., Goel, M., Muazu, L. Biogas production potential of different substrate in various combinations, *International Journal of Advanced Research in Biology Engineering Science and Technology*, 2(6):76-94, June 2016.

[69] Panswad, T., Anan, C. Impact of high chloride wastewater on an anaerobic / anoxic / aerobic process with and without inoculation of chloride acclimated, *Water Research*, 33(5):1165-72, 1999.

[70] Lefebvre, O., Moletta, R. Treatment of organic pollution in industrial saline wastewater: A literature review, *Water Research*, 40: 3671-82, October 2006.

[71] Fang, C., O-thong, S., Boe, K., Angelidaki, I. Comparison of UASB and EGSB reactors performance, for treatment of raw and deoiled palm oil mill effluent (POME), *Journal of Hazardous Materials*, 189(1-2) :229-34, 2011.

[72] De Vrieze, J., Christiaens, M.E.R., Walraedt, D., Devooght, A., Zeeshan, U., Boon, N. Microbial community redundancy in anaerobic digestion drives process recovery after salinity exposure, *Water Research*, 111:109-17, 2017.

[73] Zeng, L., Mangan, C., Li, X. Ammonia recovery from anaerobically digested cattle manure by steam stripping, *Water Science & Technology*, 54(8): 137-45, 2006.

[74] Graterol, E.M.G. Biological treatment of industrial wastewater for biogas production, *Master's thesis, University of Stavanger*; p55, 2011.