

# **Performance Evaluation of Industrial Brewery Wastewater Biologic Treatment in an UASB Reactor Using Activated Sludge in Republic of Congo**

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## **Authors' contributions**

*This work was carried out in collaboration with all the authors. Author KMM defined the subject and wrote the first draft of the manuscript. Author HMB performed the analysis. Author ACK wrote the protocol of the study. Author JMO supervised all the study. All authors read and approved the final manuscript.*

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## **ABSTRACT**

The brewing industry generates large amounts of wastewater which are released into surface water after treatment. The aim of this study is to evaluate the performance of the anaerobic treatment of brewery wastewaters in a UASB bioreactor containing activated sludge. After six-weeks operation, 30 samples were taken. Physicochemical analyzes were carried out on activated sludge (T, pH, VFA) and raw and treated waters (T, pH, AT, CAT, TSS, and COD). These analyzes showed that the conditions of the environment were favorable to an optimal growth of the bacteria:

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temperatures and pH were mostly mesophilic. the ionization of VFA was continuous and their concentration increased at the exit of the bioreactor thus revealing a significant conversion of organic materials by bacteria. The average values of the physicochemical parameters of the raw and treated wastewaters respectively increased from 31.5°C to 35°C for the temperature, from 8.9 to 7.5 for the pH, to 5.54 mg/l at 0 for AT, from 12.35 mg/l to 3.45 mg/l for TAC, from 234.08 mg/l to 129.61 mg/l for TSS and from 1637 mg/l to 282, 46 mg/l for COD. The effectiveness of the treatment allowed a COD reduction ranging from 70 to 94%.

*Keywords: Brewery wastewaters; anaerobic treatment; sludge; bacteria.*

## 1. INTRODUCTION

Most human activities, whether domestic, agricultural or industrial, produce wastewater. The amount of wastewater produced is steadily increasing worldwide as a result of population explosion and economic development. In developing countries, the vast majority of wastewater is directly discharged into the environment without prior treatment. This has consequences for human health, the quality of freshwater resources and ecosystems [1]. Industry is the human activity that generates the largest quantities of wastewater. Also, the beverage production industries are big consumers of water [2]. These wastewaters are highly biodegradable [3] and consist mainly of sugar, starch, ethanol, fatty acid, artificial sweeteners, fruit juice concentrates, flavoring agents, carbon dioxide / dissolved carbonic acid, bicarbonates, dyes, preservatives, cleaning agents (soda) and mineral salts [4]. These brewery wastewaters are also characterized by a high level of organic contaminants that require treatment before to be released in the nature or be reused [5]. For many experts, the future of the global beer Industry is now in Africa. This rush of global beer giants to Africa is due to the strong prospects for increasing production capacity due to economic growth, the rapid population boom, the emergence of a middle class and the urbanization. The volume of beer sold in Africa is expected to grow by 5%, ahead of Asia (3%) and Europe (1%) [6]. In Africa, the average annual intake of beverages per person is 9 liters. In Congo, the city of Brazzaville is the largest market in terms of population and beer consumption, followed by Pointe-Noire, the economic capital. A Congolese consumes on average 53 liters of beer a year. The development of the brewing industry in the Congo daily generates large volumes of wastewater, also called polluted water [7]. Brewery wastewaters are generally characterized by an increase in their physicochemical and microbiological parameters: the pH of the

effluents are often between 3 and 12, the temperature oscillate between 18 and 40°C, the concentrations of total suspended solids are between 200 and 1000 mg/l, the values of the COD and the BOD<sub>5</sub> are respectively in a range of 2000 to 6000 mg and 1200 to 3600 mg/l [8,9]. There are various methods of treating brewery wastewaters. Conventional treatment methods such as the aerobic system and the anaerobic system are effective because of their purifying efficiency. Biologic treatments play an important role in the brewery wastewaters management. It is based on microorganisms activity used to containing both chemical and microbial contaminants are generally treated by biological methods [10]. Wastewaters biological treatments can be aerobics or anaerobics. Aerobic treatments are often applied for the treatment of brewery wastewaters. But, recently anaerobic systems are becoming an attractive option [11]. One of the most used anaerobic treatments is the Upflow Anaerobic Sludge Blanket (USAB). In the USAB process, the brewery wastewaters pass through a bed of anaerobic sludge containing microorganisms [12]. The organic pollutants in the solution are degraded by microorganisms. At the top of the USAB reactor, a three-phase solid-liquid-gas separation, consisting of biomass, treated wastewater and biogas, is formed [13]. In order to reduce the impact of brewery wastewaters on the environment, the development of compact and efficient treatment systems such as the membrane bioreactor seems to be an appropriate solution [14,15]. It combines a biological treatment in anoxic, anaerobic and / or aerobic environment, allowing almost total elimination of organic pollutants, with the possibility of producing energy (methane), physical treatment by retention on a membrane ensuring separation total of solid (biomass) and liquid (treated) phases. In addition, membrane bioreactors make it possible to simultaneously clarify and disinfect water without the risk of formation of halogenated organic compounds, thus allowing the reuse of treated effluents [4].

The overall objective of this work is to evaluate the performance of the anaerobic biological treatment of brewery wastewaters in an up flow anaerobic sludge blanket (USAB) in the Republic of Congo.

## 2. MATERIALS AND METHODS

### 2.1 Activated Sludge

The activated sludge was chosen to seed the UASB reactor. The mass mud is composed of various plant, animal, mineral and bacterial colonies particles. This form of formation is very complex and coexists with a microflora consisting of metazoans and protozoa and various bacterial strains predominantly flocculated. Activated sludge is composed of granulated bacterial flocs because it contains a considerable amount of methanogenic bacteria. These bacteria are small ranging from 1  $\mu\text{m}$  to 1 mm. They represent a population ranging from  $10^4$  to  $10^6$  bacteria per liter of controlled sludge. This sludge is used because of its ability to digest organic matter [16].

### 2.2 Operational Process of Brewery Wastewater Treatment

Wastewater treatment is carried out in five (5) phases:

**Mechanical pre-treatment:** this step consists of discharging the water from their solid particles and large sizes. The wastewaters first pass through a screen that allows the removal of coarse and fine particles through a sieve. The mesh size of the sieve is 1 mm. This screen is equipped with a worm which allows to remove the particles and dehydrate them before throwing them into a bucket. This riddled water then passes into a pumping sump constructed of reinforced concrete and has a capacity of 48  $\text{m}^3$  a day. It collects the unshelled water and sends it, by using its three submerged pumps, to a large equalization basin where the water is mixed by a mechanical stirrer of 3 kw.

**Equalization of wastewater:** in the equalization basin, complex organic materials (proteins, sugar polymers) are hydrolysed into sugar, amino acids and fatty acids. The mechanical stirrer mixes all the particles in solution. The wastewaters thus mixed are then conveyed to the neutralization basin by means of two submerged pumps.

**Neutralization of wastewater:** The water from the equalization basin is driven by the

submersible pumps to the neutralization basin in which the correction of the pH is carried out automatically to have a pH whose values are between 6.8 and 7.8. The neutralization basin is constructed of reinforced concrete and contains an immersed agitator.

### Wastewater treatment in the UASB bioreactor:

After pH correction in the neutralization basin, the wastewaters are then transferred to the bioreactor. It is built of reinforced concrete and aims to remove organic matter and suspended solids by the action of anaerobic bacteria. It is connected to the neutralization basin by a manifold equipped with four pipes pierced with pores that feed the bioreactor. Each pipe forms an open loop and carries two manual valves. This bioreactor consists of a three-phase separator that separates the treated water on the one hand (supernatant), biomass (activated sludge) and biogas on the other hand. The activated sludge used consists of a consortium of bacteria agglutinated into granules. In the bioreactor, dissolved organic matter and biodegradable suspended solids are converted into methane. Anaerobic digestion of organic pollutants has four main stages that can be summarized by the following physicochemical reactions, which take place in the different basins [5]:

- Hydrolysis: it takes place in the equalization and neutralization basins. High molecular weight organic polymers (polysaccharides and proteins) are hydrolysed into simple soluble low molecular weight molecules (amino acids, long chain fatty acids and sugars).
- Acidogenesis (fermentation): The products of hydrolysis are catabolised by acidogenic bacteria in volatile fatty acids (propionic, butyric and acetic acids), in neutral compounds (aldehydes, alcohols), in gaseous products ( $\text{CO}_2$ ,  $\text{H}_2$ ,  $\text{H}_2\text{S}$ ) and ammonium. This phase of the process has the effect of acidification of the reaction medium.
- Acetogenesis of fatty acids: the products of the hydrolysis and acidogenesis step are converted into acetate, carbon dioxide ( $\text{CO}_2$ ) and dihydrogen ( $\text{H}_2$ ) by acetogenic bacteria.
- Methanogenesis: this last phase will lead to a gasification by the production of biogas in the form of carbon dioxide ( $\text{CO}_2$ ) and methane ( $\text{CH}_4$ ).

**Post-aeration Wastewater:** This is a pool of 60 m<sup>3</sup> volume. This basin aims to aerate the effluent using a hydro-ejector pump before being discharged into the surface waters. It removes hydrogen sulphide and other volatile organic compounds.

### 2.3 Samples Collection and Preparation

All samples come from the BRASCO brewery located in Pointe-noire. The samples were taken over 6 weeks. 30 samples of raw wastewater to be treated were collected from the sump using an electronic sampler. At the level of the effluent channel, the 30 samples were collected using a ladle of 2 m long. Sampling and sample preparation for the physicochemical analysis were performed according to the parameters to be analyzed. The sludge samples were collected from the reactor.

**Temperature and pH:** The values of these two parameters are measured immediately after in situ sampling. Both of these parameters were measured on sludge or wastewater samples. These two measurements are carried out with stirring.

**Alkalimetric title (AT) and Complete alkalimetric title (CAT):** For AT, we put in two beakers 100 ml samples to study (influent and effluent), in which we added 3 to 4 drops of phenolphthalein. If the solution turns purple, it was titrated with 0.1N sulfuric acid solution until complete discoloration of the solution and the value was read on the burette. If there is no coloration, then the alkalimetric titer is zero. To determine the CAT, we added 3 to 4 drops of methyl orange in the solution to be studied. If the solution turns orange, it was titrated with sulfuric acid at 0.1N until complete discoloration of the solution. The TAC value was also read on the burette.

**Total suspended solids (TSS):** First, the filter papers were put in the oven for 1 hour at 105°C. After one hour, the filter papers were removed from the oven and put in the desiccator for 1 minute to reduce their moisture. These filter papers were weighed and their empty weight was noted as P1 in mg. Then, 100 ml of each well-homogenized sample of the brewery wastewaters were collected and filtered at the same time using the filter paper, the Buchner funnel and the vacuum pump. When the filtration was completed, the filter papers and filtrates were put in the oven for 1 hour at 105°C. When

the drying time was over, the filter papers were removed from the oven. These desiccated filter papers with total suspended solids were put in the desiccator for 1 minute then weighed. Weights found were noted P2 in mg. The weight difference between P2 and P1 represents the amount of total suspended solids in a 100 ml sample.

**Volatile Fatty Acids (VFA):** 50 ml of each sample was filtered with a filter paper. Using a pipette, 0.5 ml of the filtrate of each sample was taken and introduced into three 25 ml test tubes. Then, 1.5 ml of ethylene glycol and 0.2 ml of sulfuric acid were added to the test tubes. The test tubes were finally shaken to homogenize the mixture. These test tubes were heated in a boiling water bath for 3 minutes. Before letting them cool, 0.5 ml of hydroxylamine, 2 ml of sodium hydroxide solution, 10 ml of ferric chloride solution and 10 ml of distilled water were added respectively. Once cooled, the solutions were placed in the spectrophotometer for analysis.

**Chemical oxygen demand (COD):** in flasks already containing dichromate, 0.2 ml of each sample of brewery wastewater collected were added. These well-homogenized solutions were placed in the reactor with heating for 2 hours. The temperature gradually decreased to 40°C. Then, the flasks were removed from the reactor and left at room temperature for cooling. Once cooled, the solutions were placed in the spectrophotometer for analysis.

### 2.4 Analytical Methods

All analyzes were performed according to the current AFNOR standards and standard methods [17]. The temperature and the hydrogen potential (pH) were determined in situ by potentiometric methods according to standard NF T90-008 using the pH meter with integrated thermal sensor 3310WTW. The COD equivalent to the amount of K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub> consumed as well as the total suspended solids and volatile fatty acids are read directly with a Hach Lange DR3900 UV-Visible Spectrophotometer.

## 3. RESULTS AND DISCUSSION

### 3.1 Raw Brewery Wastewaters

The composition of raw brewery wastewaters is given in Table 1. Analyzes were performed on

individual samples and weekly mean samples. The average values presented were obtained from the values of the 30 individual samples. The values of the main parameters studied and WHO wastewater standards are summarized in Table 1. The main parameters of interest in the proper functioning of a biological treatment plant are: temperature, pH, TSS and COD. It appears that the average values of the temperature, the pH, the SS and the COD of the raw wastewater are respectively: 31.5°C, 8.9, 234.08 mg/l and 1637 mg/l. The pH alkalinity of these raw brewery wastewaters indicates the presence of carbonates and hydroxyls, which is confirmed by the values of AT and CAT. All average temperature, pH, MES and COD values are not in compliance with WHO wastewater discharge standards, which require a temperature <30°C, a pH between 6.5 and 8.5, MES <20 mg/l and a COD <90 mg/l. Also, temperature and pH influence the multiplication of bacteria. An excess of TSS can cause clogging of the pretreatment system [18] and raw brewery wastewaters with too much organic matter will be incompletely purified because it requires more intense microbial activity. So, these brewery wastewaters directly discharged into the receiving environment should affect the quality of surface water. These raw wastewaters from the brewery must therefore be treated before discharge.

### 3.2 Bacterial Activity in the USAB Reactor

The microorganisms composing the purifying mud require, like all living beings, special environmental conditions for the proper functioning of their metabolism. pH and temperature are two parameters that jointly

influence the life and biological activity of bacteria in activated sludge. The production of volatile fatty acids is a means of evaluation the effectiveness of bacterial activity. The weekly pH, temperatures and volatile fatty acids (VFA) ranges in the bioreactor, obtained over 6 weeks, are collated in Tables 2 and 3.

**Influence of pH:** It is observed that the pH in the UASB bioreactor varies between 6.5 and 11 depending on the weeks (Table 2). From week-4, the pH revolves around neutrality in the bioreactor. Methane-producing bacteria can function to the best of their ability within a very narrow pH range of 6.7 to 7.4 [19]. Indeed, the acidifying bacteria perform the acidification stage of the wastewater upstream of the anaerobic reactor. They therefore produce significant amounts of  $H_3O^+$  ions following the hydrolysis of the molecules thus causing a decrease in the pH of the effluents. Their activity decreases with the drop of pH to a total inhibition below a pH value of 4.5, toxic for most bacteria [19]. pH plays an important role in the bacterial growth of the species present in the anaerobic granule. The activity of the bacteria will therefore be optimal for weeks 3, 4, 5 and 6. For weeks 1 and 2, it is observed that the pH tends toward alkalinity. The pH variation causes effects on the pellet resistance of the scrubbing sludge in the UASB bioreactor. Under high pH conditions (8.5 -11.0), the granular structure of the sludge is weakened whereas in the pH range of 5.5 to 8.0 the granular structure of the sludge is relatively stable [20]. pH regulation is then necessary to promote the acidification reaction, particularly in the case of effluents heavily loaded with organic matter.

**Table 1. Brewery wastewater composition before treatment**

Parameters	Range values	Mean value	WHO wastewaters standards
Temperature (°C)	25-38	31,5	<30
pH	6,8-11	8,9	6,5-8,5
AT-Alkalimetric title (mg/l)	1-13	5,54	/
CAT-Complete alkalimetric title (mg/l)	5-17	12,35	/
TSS-Total suspended solids (mg/l)	80-347	234,08	<20
COD-Chemical oxygen demand (mg/l)	600-2510	1637	<90

**Table 2. pH and Temperature ranges values in USAB reactor**

Parameters	Week-1	Week-2	Week-3	Week-4	Week-5	Week-6
pH	6,85-11,0	7,12-10,26	6,6-7,62	6,5-6,8	6,6-7,0	6,67-6 ,8
Temperature (°C)	28,2-38,0	33,0-34,1	31,0-32	30,0-35,5	30,0-35,5	36,4-36,7

**Table 3. VFA influent and effluent range values**

Parameters	Week-1	Week-2	Week-3	Week-4	Week-5	Week-6
VFA-Volatil fatty acids (mg/l)-Effluent	17-18	18-33	30-50	17-18	18-30	28-55
VFA-Volatil fatty acids (mg/l)-Influent	88-190	24-49	120-155	114-150	110-135	119-158

**Table 4. Brewery wastewaters mean composition before and after treatment**

Parameters	Mean value before treatment	Mean value after treatment	WHO wastewater standards
Temperature (°C)	31,5	35	<30
pH	8,9	7,5	6,5-8,5
AT-Alkalimetric title (mg/l)	5,54	0	/
CAT-Complete alkalimetric title (mg/l)	12,35	3,45	/
TSS-Total suspended solids (mg/l)	234,08	129,61	<20
COD-Chemical oxygen demand (mg/l)	1637	282,46	<90

**Influence of temperature:** The temperature range in the UASB bioreactor varies between 28.2°C and 38°C depending on the week (Table 2). All enzymatic activity of bacteria depends on the temperature. The biological limits of microorganisms should be respected according to their group membership: The psychrophilic, mesophilic or thermophilic. In the majority of cases, industrial plants operate in mesophilic conditions between 20 and 40°C. This is the case of our experiments, which are well within the mesophilic temperatures range. Thus, methanogenic bacteria develop in 3 days at 35°C while the growth time is 50 days at 10°C [19]. When the reactor temperature is under 30°C, the activity of the methanogenic bacteria is greatly reduced. Therefore, to ensure proper operation, the mesophilic UASB reactor must operate at a temperature between 30 and 35°C: This temperature range is observed for weeks 2, 3, 4 and 5. In UASB bioreactor a change of mesophilic conditions (temperature from 20 to 40°C) to thermophilic conditions (temperature from 50 to 60°C) can cause important structural modifications [21] such as malfunctions of the anaerobic purification by creating a thermal shock. In the case of a sudden drop in temperature, the activity of the bacteria drops with the consequence of a slowing down of the purification capacities of the mud. In this case, only an inhibition and a rise of the temperature will make it possible to find an optimal purification [22].

**Influence of volatile fatty acids (VFA):** The concentration of VFA oscillates between 17 and 55 mg/l at the entry of the bioreactor and between 24 and 190 mg/l at the exit (Table 3).

VFAs represent an important part of the substrates converted by methanogenic bacteria. The effectiveness of the bacterial activity can therefore be interpreted by the elimination of volatile fatty acids which is an index of the presence of organic matter at the exit of the bioreactor. VFAs are less present in influent brewery wastewaters (Table 3). But after hydrolysis the organic polymers are broken down into fatty acid chains. This increases the amount of fatty acids that are then digested by the bacteria and converted to CO<sub>2</sub> and CH<sub>4</sub> in the bioreactor. Weeks 3, 4, 5, and 6 have better VFA production ranges certainly due to optimal mesophilic pH and temperature conditions. The VFA amount increase in the effluents denotes a very favorable balance to the ionized form of the fatty acids rather than to the molecular form. Indeed, VFA are weak acids that are not completely dissociated in water unlike strong acids. Depending on the pH, the molecular form remains in the medium in greater or lesser amounts. Or assimilated in their molecular form by bacteria, they can become toxic if their concentration in the medium becomes too high, which would result in the decline of activity or death of bacteria. The ionized form is not toxic because of their impossibility to enter the cell [23]. It is therefore necessary to maintain a very favorable balance to the ionized form in the environment so as not to poison the bacteria.

### 3.3 Treated Brewery Wastewaters

It appears that all the average values of the physicochemical parameters of the treated wastewater compared to raw influents are respectively passed from 31.5°C to 35°C for the

temperature, from 8.9 to 7.5 for the pH, from 5.54 mg/l to 0 for AT, from 12.35 mg/l to 3.45 mg/l for TAC, from 234.08 mg/l to 129.61 mg/l for TSS and from 1637 mg/l to 282.46 mg/l for DOC. These results show the effectiveness of the treatment. Indeed, the average temperature of the treated wastewaters at the reactor outlet is that recommended for optimal growth of mesophilic bacteria [21]. The pH neutrality of the brewery's treated wastewater indicates the absence of carbonates and hydroxyls, which is confirmed by the drop in AT and CAT values. There is also a decline in TSS and a drop in DOC. These results indicate that continuous organic matters in the brewery's raw wastewater were largely degraded. However, the levels of treated wastewaters temperature, TSS and COD do not meet the WHO standard effluent. These waters therefore require additional treatment before considering their return to the natural environment.

### 3.4 Performance of the Activated Sludge in the USAB Reactor

The purification performance of activated sludge used will be analyzed through the monitoring of

TSS and COD over time. The average of TSS and COD values of the treated wastewaters can be used to assess the performance of activated sludge treatment. As shown in Fig. 1 and Fig. 2, the parameters varied considerably in terms of the TSS and COD.

**Total suspended solids (TSS):** Fig. 1 shows the continuous reduction of TSS throughout treatment. The average allowance rate in TSS varies between 14% and 77%. This low yield may be related to the nature of the TSSs carried with the effluent. In fact, the effluents are composed of cross-linked particles which sediment easily and low density particles formed of granules, the majority of the cross-linked particles are stopped at the decanter while the low density particles are entrained with the effluent [24]. To prevent these particles from affecting the effectiveness of the treatment, a sand / gravel filter should be placed downstream of the treatment. This will improve the removal of TSS while providing a reduction in COD.

**Chemical oxygen demand (COD):** Fig. 2 demonstrates the continued reduction of COD throughout treatment. This allowance

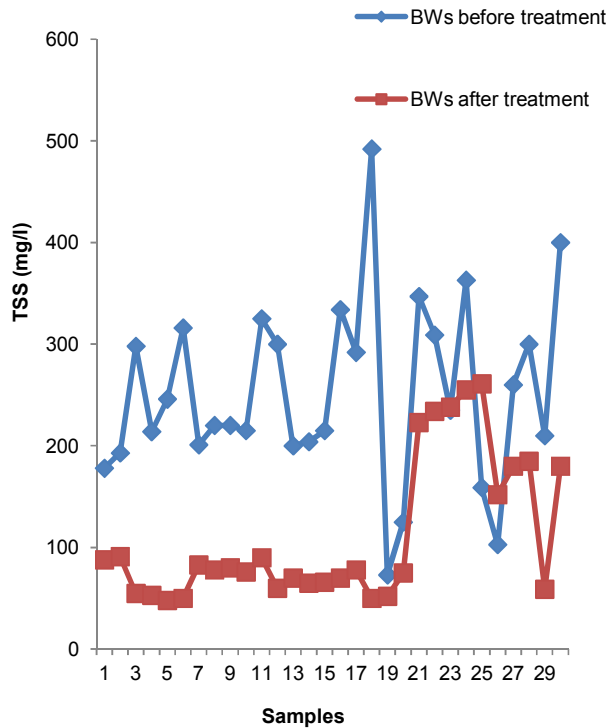
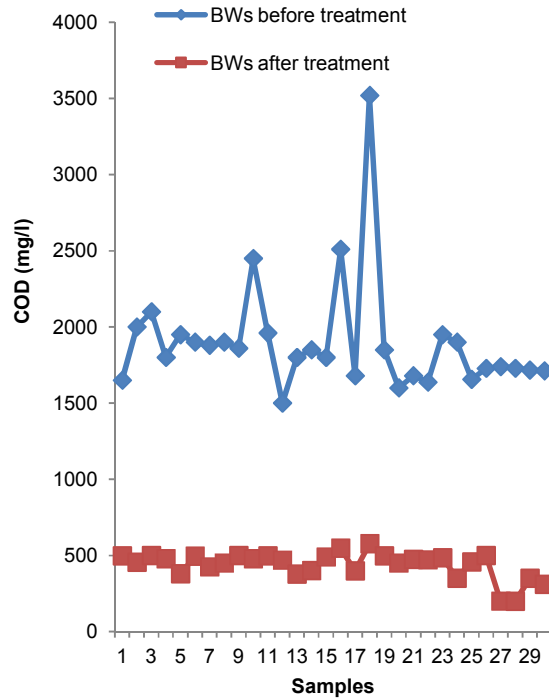


Fig. 1. Variation of TSS concentration in brewery wastewaters (BW) before and after biologic treatment



**Fig. 2. Variation of COD concentration in brewery wastewaters (BWs) before and after biologic treatment**

corresponds to an average percentage of COD removal between 70% and 94%. The results obtained are consistent with those reported Stadlbauer et al. [25] who found COD removal efficiencies of 85 to 90% from anaerobic purification of beer brewery wastewaters with biofilm reactors. Austermann-Haun and Seyfried [26] also reported 80% COD removal efficiency from a pilot-scale UASB reactor treating clear beer brewery wastewater. Another study using a laboratory-scale upflow sludge blanket reactor at ambient temperatures gave a COD removal of 89% [27]. In addition, higher abatement rates were obtained by Torres et al. [28] that achieved a 99% reduction in COD and 100% organic matter in the anaerobic treatment of brewery wastewater using a ceramic membrane. In other words, the performance of the UASB currently being reviewed could be improved.

#### 4. CONCLUSIONS

The wastewater management of breweries is of major interest. Indeed, this study has shown that raw wastewaters from breweries are polluted and require treatment before being released into the

natural environment. For this purpose, biological treatments are an interesting alternative because they succeed to purify liquid effluents with a high content of organic matter. In this study, we have demonstrated that organic pollution of wastewaters from breweries suffers considerable degradation by bacteria over time with a reduction of 70 to 94% of COD. However, the reduction in TSS remains low, which indicates that the suspended solids are slightly removed from the effluents. Finally, the average values of treated effluents in TSS and COD remain above the standard recommended by WHO for wastewaters.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.



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