MICROBIOLOGICAL ASPECTS OF THE AERATION TANKS OF AN ACTIVATED SLUDGE TREATMENT PLANT DURING DYSFUNCTION: CONSEQUENCES ON ITS TREATMENT PERFORMANCE.

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ABSTRACT

Benfreha, B.M., Belgharbi, A. A., Mokrani, S., Tir.Touil, M.A., Moussa-Boudjemâa, B. (2022). Microbiological aspects of the aeration tanks of an activated sludge treatment plant in dysfunction: Consequences on its treatment performance. *Lebanese Science Journal,* 23 (1): 17-32.

The present study was conducted in a sewage treatment plant, called "El Kouwaer", located in the department of Mascara, North-West of Algeria. Mainly, the objective of this investigation is to evaluate the relevant settling and pollution parameters and to assess the proliferation level of filamentous microbial communities during the dysfunction period to further the effectiveness of the operative treatment. Sludge parameter values were estimated such us Settling Test, Diluted Sludge Volume Index (DSVI), and Total Suspended Solids (TSS) content to set up the sludge classification, the performance analysis of treatment was conducted by computing pollution indicators and removal efficiencies of organic pollution, as well as the Principal Component Analysis (PCA) of dysfunction parameters. The results showed that in aeration tanks 65% of samples represented a maximum Filament Index (FI) of 6. Diluted Sludge Volume Index (DSVI) was greater than 300 mg/L in 23% of samples. A high level of filamentous bacteria biodiversity was also noted during the assessment of the (FI)

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including Microthrix parvicella, Bogiotae ,and Sphaerotilus natans. The effluent in the biological unit was characterized by BOD5/COD ratio of 0.17 and COD removal efficiency of 53.32%. Those results demonstrated that the sewage treatment plant is frequently affected by bulking problems. This study had confirmed the importance of continuous monitoring of different process parameters like Wastewater Biodegradability Index, Dissolved Oxygen, filamentous flora, and Dilute Sludge Volume Index (DSVI) to avoid irreversible damage at the level of a wastewater treatment plant.

Keywords: Sewage, activated sludge, microbiological dysfunction, treatment efficiency.

INTRODUCTION

Among the solutions likely used to increase the availability out of water, the recycling of sewages seems to be of great importance. In Algeria, wastewater discharged from the sewerage network is estimated at 92 million m^3permonth and the treated water volume is estimated at about 21 a million m^3 per month (ONA, 2019). El Kouwaer sewage treatment plant that is the subject of this study is located at Mascara in the northwest of Algeria. This semi-arid plant is in water deficit and the water availability is about 186 million m^3per year, while water requirements for agriculture are estimated at 58.7 million m^3per year (for an average of 7300 m^3/ha/year) (Benfetta et al., 2017). The sewage treatment plant of El Kouwaer is one of 75 activated sludge plants managed by the National Sanitation Office (ONA). It has a wastewater treatment capacity of 13000 m^3per day. This quantity could be used in the agricultural sector if the quality of treated water is improved.

Unfortunately, many types of dysfunctions can occur and constitute a permanent concern to the sewage treatment plant. The most frequent type of these dysfunctions is the excessive filamentous bacteria growth which may reduce the sludge settling and/or cause bulking phenomenon (Nierychlo et al., 2019; Faheem & Khan, 2009). Thus, pollution parameters such as BOD_5, COD and, TSS content may not be able to fulfill the lawful requirements relating to liquid discharges. According to several authors, there is no universal strategy to limit or prevent the presence of these filamentous organisms (Neisi et al., 2020; Henriet et al., 2017).

The reuse of treated wastewater effluent remains dependent on the improvement of its bacteriological quality (Crook & Lazarova, 2005). For instance, in Algeria, a list of crops can be irrigated with treated sewage required by regulation that gives us a clear recommendation of these crops that can be irrigated with treated water as cereals, fodder, and fruit trees.

The main objectives of this study are: (i) to characterize the activated sludge parameters in the aeration tanks during dysfunction period; (ii) demonstrating the relationship between sludge parameters and pollution parameters through comparing the plant's operational and dysfunction the periods; and (iii) highlighting the treatment performance through showing that once the sludge flora balance was adversely affected, the purification performances of the sewage treatment decrease and it will be complicated to correct them. Thus, parameter indicators must be sought to detect dysfunctions as earliest as possible and fix the problem in time.

This investigation was conducted to provide an idea on the relationship between the dysfunctional and sludge parameters including Filamentous Index (FI); Dilute Sludge Volume Index (DSVI) and other parameters like Dissolved Oxygen (O_2), COD and BOD5 demonstrated by the Principal Component Analysis (PCA).

Thus, representation of a model factor card for the power used to set early-stage dysfunctions that may occur in a similar activated sludge treatment plant. This approach is the first report in the field of studying the performances of an activated sludge treatment plant in Algeria.

MATERIALS AND METHODS

2.1. Plant description and sampling methodology

2.1.1. Plant description

The wastewater treatment plant "El Kouwaer" is located 3.2 km from Mascara department (361 km from Algiers) in the so-called Kouwaer area. This plant was commissioned in August 1996. The domestic releases of El Kouwaer treatment plant comes mainly from Mascara department and is powered by a semi-separate network. The effluent is rather urban and the discharges of the industries do not pass through this station. It has a capacity of 768.301 inhabitant equivalents whose average quality was estimated at 9209 m^3 per day. When it was designed, it irrigates a huge agricultural area of 1095 ha (Figure 1).

2.1.2. Sampling methodology

During seventeen weeks, each week, one sample is taken upstream and downstream for measuring removal efficiency TSS, BOD5, and COD and at the level of the two aeration tanks for the evaluation of settling index: Filament Index (FI) and Diluted Sludge Volume Index (DSVI). Thus, seventeen samples were collected for each essay and preserved following the ISO Standard requirements. This method consisted of taking 200 ml of wastewater at regular intervals and transferring them into a container in which all samples of the same day were mixed to obtain a representative sample of 8 hours working period.

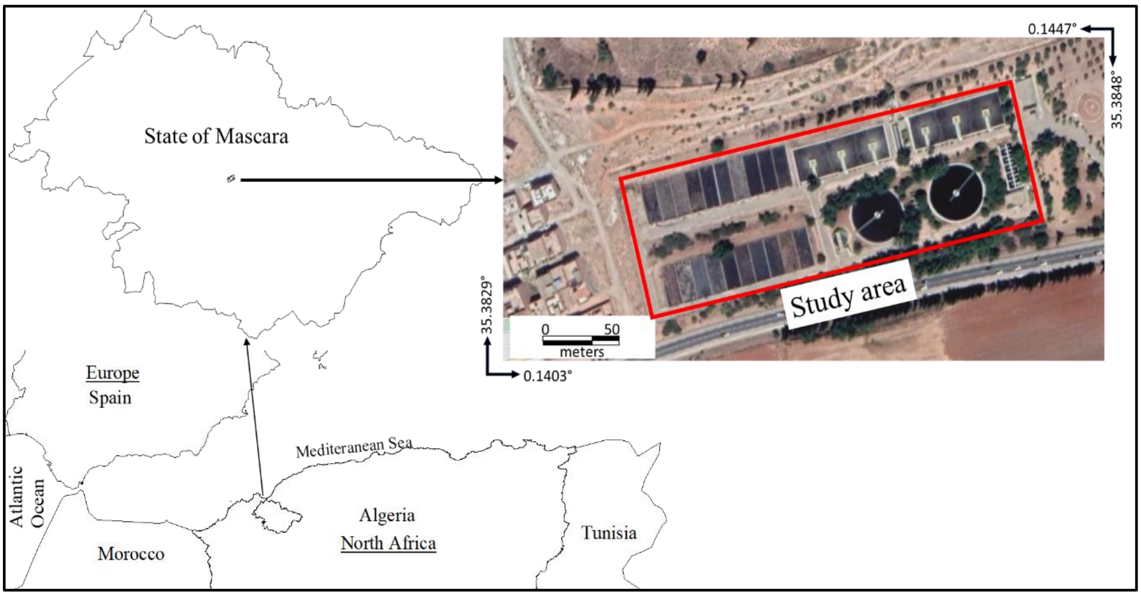

Figure 1. Localization of the study area

2.2. Microbiological analysis of wastewater before and after treatment

2.2.1. Sludge parameters

2.2.1.1. Settling test

For this purpose, 1 liter of activated sludge was placed in a test tube. If necessary, dilutions (1/4 or 1/2) were carried out with influent wastewater from the Clarifier. The height of the sludge was then evaluated for 30 min every 5 min (Prado et al., 2019).

2.2.1.2. Dilute Sludge Volume index (DSVI) and Total Suspended Solids (TSS)content

The DSVI was determined by using the lowest dilution at which the volume after 30 min of settling (V 30) was less than 300 ml. It was calculated using the formula (1) (Eikelboom, 2000; Van der Waarde et al., 2002; Jenkins et al., 2003).

$$DSVI = \frac{VD_{30}}{C_{ep}} \qquad (1)$$

Where:

DSVI: Dilute Sludge Volume Index (ml/g)
VD_{30}: Volume of sludge decanted into 30 min (ml).
C_{ep}: Concentration of TSS in the test tube (g/L).

Total Suspended Solids (TSS) content was performed according to the ISO standard. 100 ml of wastewater were passed through a filter of 0.7 µm porosity of a vacuum filtration system which one has determined the mass beforehand (denoted P_0).

The TSS retained on the filter was dried at 105 °C for 2 h. The residue mass obtained after drying was measured (denote P1). TSS content is given by formula (2):

$$\boldsymbol{TSS} = \frac{[(\boldsymbol{b_1} - \boldsymbol{a_1}) - (\boldsymbol{b_0} - \boldsymbol{a_0})] * \mathbf{1000}}{\boldsymbol{Ve}} \quad \mathbf{(2)}$$

Where:

a_o: White filter weight before filtration (in mg)
b_o: White filter weight after filtration (in mg)
b_1: Sample filter weight after filtration (in mg)
a_1: Sample filter weight before filtration (in mg)
V_e: Sample volume (in ml).

2.2.1.3. Sludge classification scale based on filament

The filaments abundance was estimated as the Filament Index (noted FI) allowed to the sludge based on its filament density (Gnida et al., 2018; Paździor & Bilińska, 2020).

2.2.2. Performance analysis of treatment

2.2.2.1. Pollution indicators

The COD (Chemical Oxygen Demand) was measured by a COD VELP SCIENTIFICA reactor (ECO8 Usmate, Italy). The $BOD_5$was measured by a BOD meter VELP SCIENTIFICA (BMS6 Usmate, Italy). Furthermore, Dissolved Oxygen (O_2) was measured by an oximeter (Oxy 3205 WTW Germany) according to ISO standard.

2.2.2.2. Removal efficiencies of organic pollution

Monitoring of physicochemical parameters was performed using the techniques mentioned by Rodier et al. (1996) and according to the standards mentioned above for TSS, BOD5, and COD (Jóźwiakowska & Marzec, 2020).

Calculation of the purification efficiency of the parameters COD, BOD5, and the Total Suspended Solids (TSS) was done using the equation (3):

$$\boldsymbol{R}\,(\%) = \frac{\left(\boldsymbol{Cp_{effluent}} - \boldsymbol{Cp_{influent}}\right) * \mathbf{100}}{\boldsymbol{Cp_{effluent}}} \quad (\mathbf{3})$$

Where:

R (%): Treatment removal efficiency of the considered chemical parameter (in percent).

$Cp_{effluent}$: Considered parameter concentration of effluent water in the plant.

$Cp_{influent}$: Considered parameter concentration of influent water in the plant.

2.3. Statistical analysis

Statistical analysis was performed by Principal Component Analysis (PCA) and graphing of dysfunction parameters using XLSTAT version 7.5.2.

RESULTS

3.2. Sludge parameters during dysfunction period

3.2.1. Settling test

A rise in sludge was observed (Figure 2), and the Dilute Sludge Volume Index (DSVI) was higher than 300 ml/g for the seventh week of study. Thus, clearly showed a high density of filamentous microorganisms concordant with a filament index (FI=6) (Figure 3). A slight flaky flock of dark brown or black color with a large settling volume was also observed during the settling test.

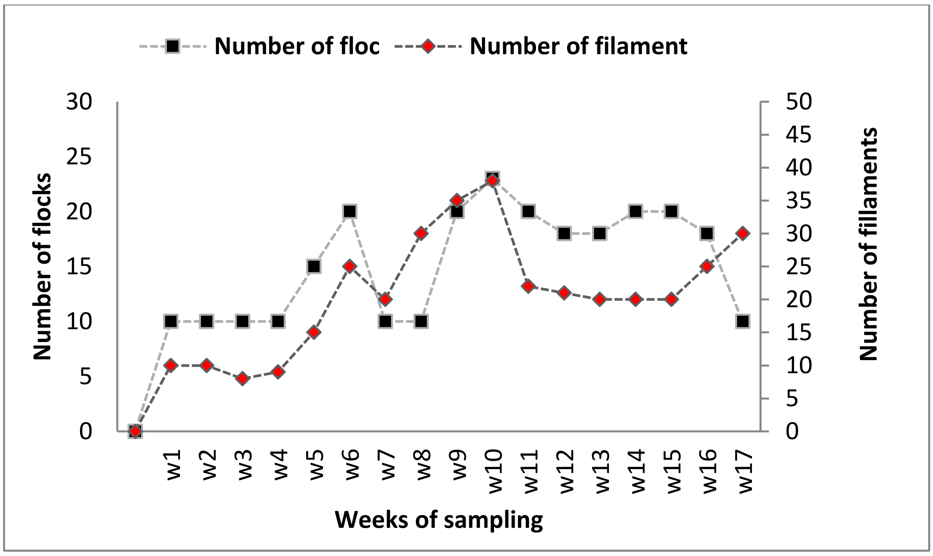

Figure 2. Numbers of filaments and flocks for the same observation during dysfunction period in the sludge of the plant's aeration tanks.

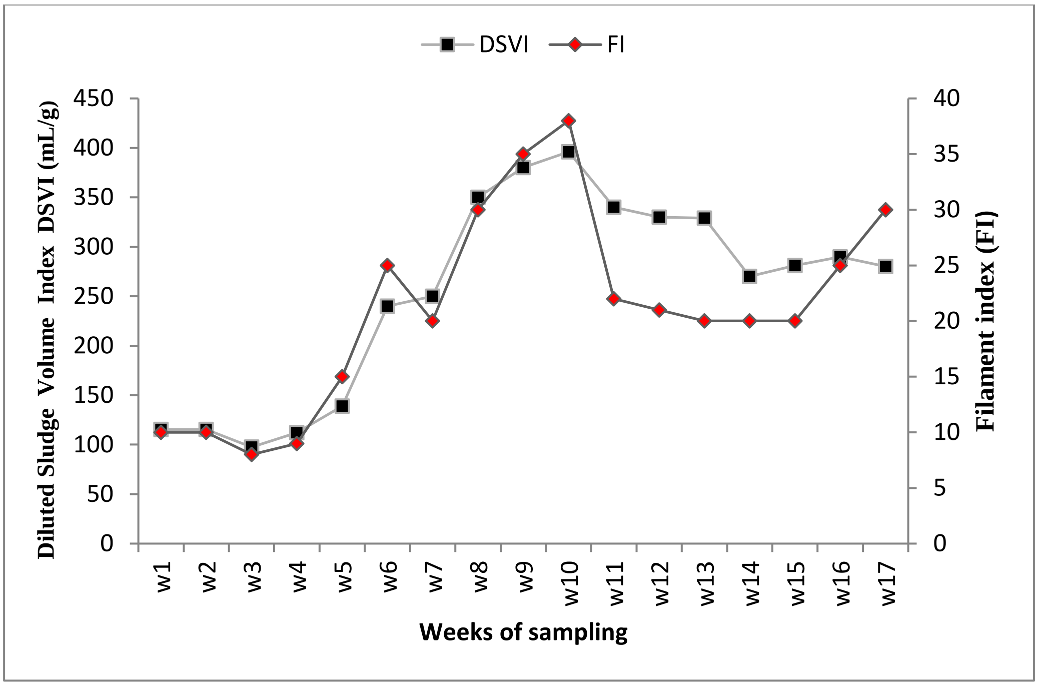

Figure 3. Variation of the Filamentous Index (FI) as a function of the Dilute Sludge Volume Index (DSVI) for the same sample during dysfunction period in the sludge of the plant's aeration tanks.

3.2.3. Abundance of filamentous bacteria and Filament index

The Filament Index (FI) enabled the assessment of the abundance of filaments present in the flock. Results showed that the filamentous bacteria were very abundant in most samples with an average density of 5 to 10 filaments per flock, and a Filament index (FI) of 4 to 12% of samples. Abundant density was characterized by more than 20 filaments per flock with FI from5 to 24% of samples. Excessive density, more than filaments per flock represented a FI ranging between 6 to 65% (Figure 4) and (Figure 5). This technique allowed getting an overall idea of the abundance of filaments in a sample. Indexes are assigned to the sludge according to their filament density according to Paździor and Bilińska (2020).

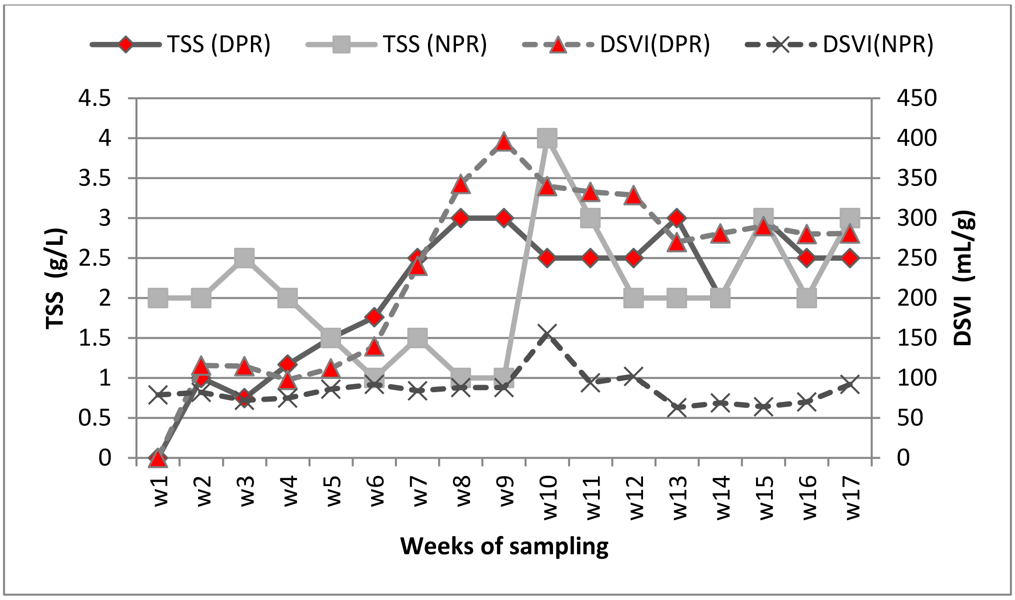

Figure 4. Values of Dilute Sludge Volume Index (DSVI) and Total Suspended Solids (TSS) at the level of the plant's aeration tanks during normal operation (NPR) and dysfunction period (DPR).

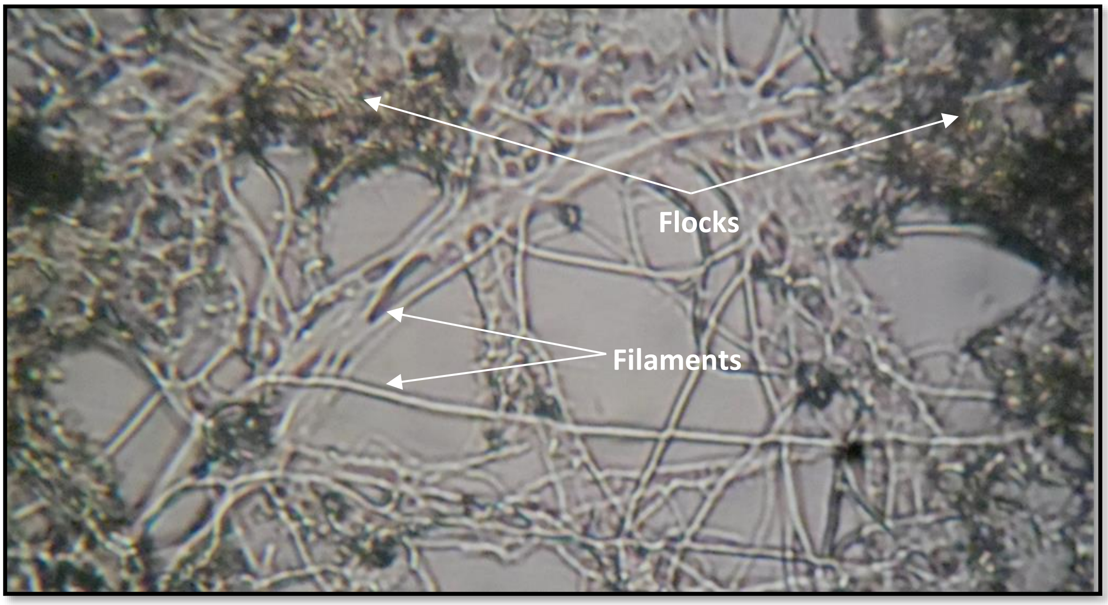

Figure 5. Appearance of the excessive proliferation of filamentous bacteria at the level of the sludge of the plant's aeration tanks (Filament Index (FI=6).

3.3. Analysis of treatment performance

3.3.1. Pollution indicators

In order to characterize the nature of the effluent, the ratio of BOD_5/COD was used. According to Abdouni et al. (2021), this ratio depends on the nature and origin of sewage that can be domestic or industrial. During the dysfunction period, results obtained showed that the biodegradability index was characterized by an average equal to 0.17 and varied between 0.1 in April to 0.26 in June. During the normal period, the biodegradability index expressed an average of 0.6 and varied between 0.15 recorded in March and 0.8 unregistered in June (Figure 6).

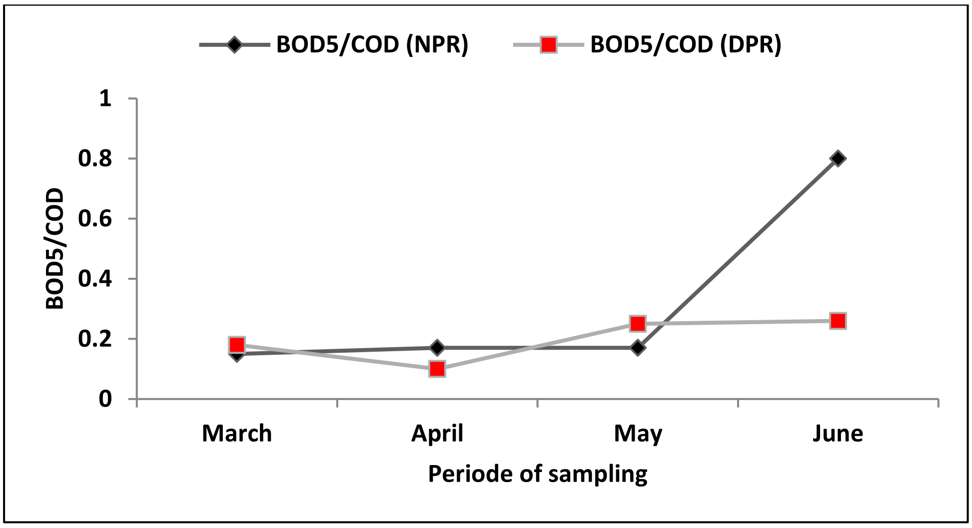

Figure 6. Comparison of the BOD5/COD ratio of the plant's effluent during normal operation (NPR) and dysfunction period (DPR).

3.3.2. Removal efficiency of organic pollution (Organic pollution abatement)

Results showed that during the normal period (NPR) operation removal efficiency of TSS, COD, and BOD_5 was 93.42%, 92.14% and 96.11%, respectively (Figure 7). Whereas, during the time of dysfunction period (DPR), the reduction of these parameters was in the following order 90.47%, 53.32%, and 85.51%, respectively. During the normal period (NPR), pollution abatement at El Kouwaer plant was conforming as mentioned by the directive of European Union. During the dysfunction period, the pollution was insufficient especially in terms of BOD_5.

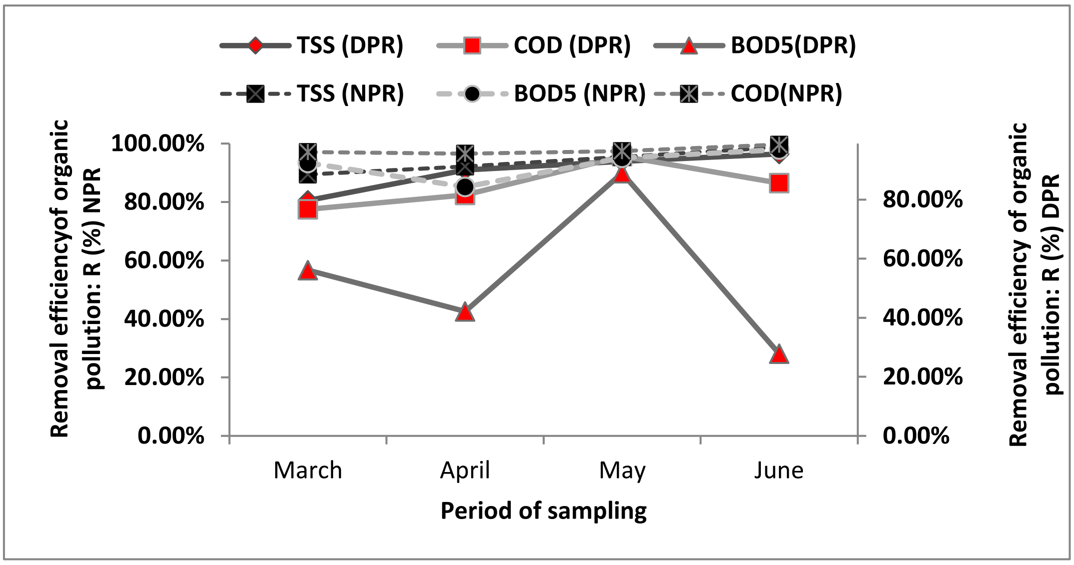

Figure 7. Comparison of the organic pollution parameters (COD, BOD5, and TSS) of the effluent during the normal operation (NPR) and dysfunction period (DPR).

3.4. Principal Component Analysis (PCA) of dysfunction parameters

This analysis aimed to identify the existence of any relationship between dysfunction parameters (BOD_5/COD ratio, dissolved oxygen, DSVI, and FI). Results showed that F1 and F2 projections accounted 86.64% of the information. Thus, it was quite sufficient and allowed to ignore other axes the F1 axis provided 50.81%, while the F2 axis provided 35.83% (Figure 8).

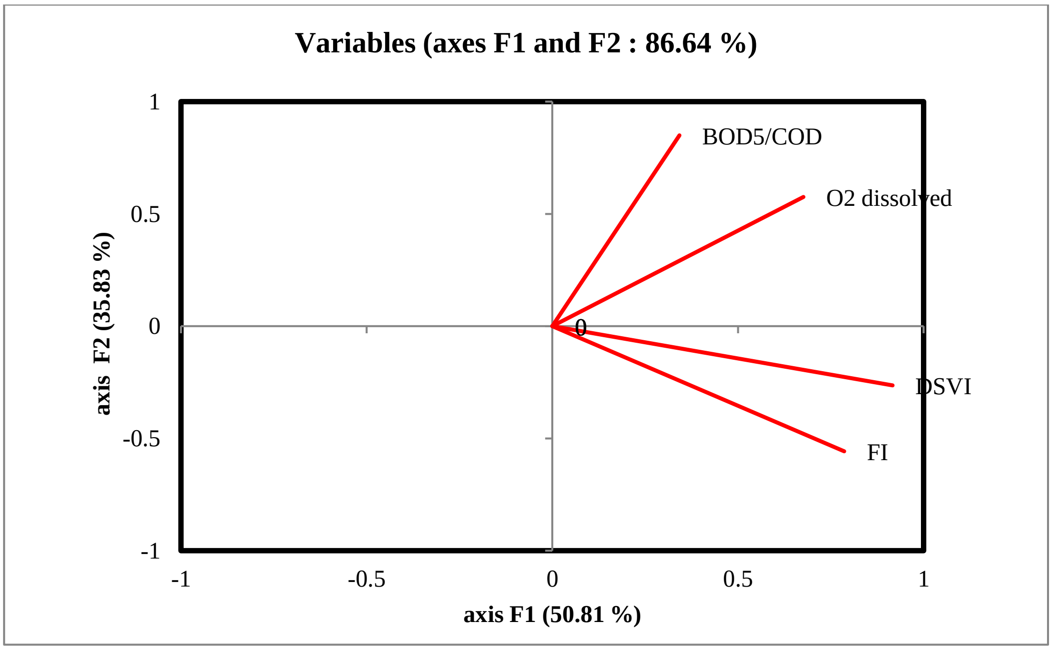

Figure 8. ACP graphing showing Cartesian diagram of the correlation between dysfunction parameters (BOD5/COD, O2, DSVI, and FI) at El Kouwaer wastewater treatment plant during dysfunction period.

Pearson coefficient showed a significant positive correlation between Filamentous Index (FI) and the Dilute Sludge Volume Index (DSVI) with r=0.807 and between biodegradability index (BOD_5/COD) and dissolved oxygen in the aeration tanks with r =0.541. This allowed concluding that there was a strong correlation between these parameters.

DISCUSSION

Overgrowth of filamentous bacteria is associated with deteriorated sludge settling, leading sometimes to serious operational problems known as the sludge bulking phenomenon **(**Liao et al., 2004; Vervaeren et al., 2005**).** This study revealed the extent of this problem at the level of an important wastewater treatment plant, El Kouwaer, located in the North-west of Algeria.

In this current investigation, the evaluation of sludge parameters during the dysfunction period showed that DSVI average was higher than 300 ml/g, which was in perfect agreement with Pandolfi (2006) who has described the filament density mechanism compared to the flocks establishing junctions between them and content of the sludge, in such cases the sludge index is still ≥ 200 ml/g.

On another side, the results of this current study demonstrated that filamentous bacteria showed great biodiversity (Van Der Waarde et al., 2002; Wagner & Loy, 2002; Wágner et al., 2015) and were abundant in most samples (FI=6 in 65% of the samples). Comparing to Mielczarek et al. (2012) who recorded that the filamentous index fluctuated of FI between (2-3). Whereas, Paździor and Bilińska (2020), noted FI variation among (2-4). Both authors concluded that these results weakened the plant's processing properties. The comprehensive baseline survey conducted as part of this study revealed also that during this period the plant was well in a dysfunctional state. Thus, gives a general idea that the proliferation of filamentous microorganisms responsible for the "bulking" phenomenon promoted the continuing disruption and fluctuation which affect directly the purifying performances. Importantly, it was evident that total filament abundance alone cannot always explain dysfunction related to the bulking phenomenon. For this reason, an analysis of Dilute Sludge Volume Index (DSVI) seems to be essential to any conclusion.

In this current work, based on DSVI analysis and Filamentous Index (FI), the current plant studied can be identified as a WWTP with severe sludge settlement problems. In order to visualize the relationship between filament abundance and the settling properties, a time-series study of the filamentous organism abundance present and the measured DSVI values were analyzed during 17 weeks. The abundance of filamentous bacteria observed as well as the drastic increase of DSVI values (>200 mL/g) was accompanied by the proliferation of *Microthrixparvicella, Nostocoïdalimicola* I, II and III, *Bogiotae sp*, and *Sphaerotilus natans* observed during FI elaboration. The cumulative abundance of these filamentous organisms seems to explain the DSVI pattern observed. While the study performed by Nierychlo et al. (2020) showed lower values of DSVI ranging from 80 to 150 mL/g.

The study of the performance of any wastewater treatment plant requires a diagnosis of the pollution parameters which gives better visibility of the dysfunctions. In that context, the biodegradability of wastewater can be estimated by the mean of BOD5/COD ratio. The entry in the biological unit was characterized by BOD5/COD ratio of 0.17 during the dysfunction period. This report indicates clearly that a part of the organic material is not biodegradable. Whereas, during the normal period a ratio equal to 0.6 was recorded. It was indicated in the literature that wastewater characterized by BOD5/COD ratio rangingamong0.2-0.5 is biodegradable. In this case, chemical pretreatment is recommended (Gottschalk et al., 2010; Wágner et al., 2015), which means that the organic loads of the plant's treated wastewater is hardly biodegraded. This was confirmed by the COD removal efficiency value that was 53.32% during the malfunctioning period compared to 92.14% during the normal period. In Ouardigha-Morocco, Boutayeb et al. (2012) reported an index of WWTP approximating 0.3. An increasing number of studies have observed higher index values varying between 0.5-0.6 (Tahri et al., 2015; Hamaidi-Chergui et al., 2016; Sa et al., 2021**)**. In this situation, it is recommended to consider physic-chemical treatment**.**

During the dysfunction period, analysis of the evolution rate of removal efficiency including three parameters (COD, BOD_5, and TSS) showed lower performance, especially for COD. Servais et al. (1999) explain this decreased performance by the effluent nature that is not easily biodegradable and by the significant reduction in the role of microorganisms in the degradation and transformation of organic matter. Thus, elimination of organic pollution and especially excessive growth of filamentous bacteria, observed during the dysfunction period of the current plant, strongly limited the proper operation of the clarifier. The function of this latter is to ensure efficient settling that favors the elimination of sludge responsible for the majority of BOD_5 (Al-Sulaiman & Khudair, 2018). Consequently, filamentous bacteria in sewage treatment plants are also in relation to nutrients removal (Milobedzka & Muszyński, 2014).

An increasing number of studies had reported satisfactory performance concerning various analyzed parameters like Hamaidi-Chergui et al. (2016) in a WWTP of Medea-Algeria; Diman et al. (2016) in Al Hoceima-Morocco and Paździor & Bilińska (2020) in Poland. The comparison of these results with those of the normal treatment period showed an attractive return for all parameters studied.

A significant correlation was observed between dysfunction parameters. This funding's was also mentioned by other authors (Karefa et al., 2017; Rekrak & Fellah, 2020; Salama et al., 2020)**.** They demonstrated the relationship between the Dilute Sludge Volume Index (DSVI) and the density of filamentous bacteria (FI). Firstly, according to the same authors, the higher DSVI of 200 ml/g (bulking) represents an important development of filaments in the sludge making this index an evaluable tool for preventive dysfunction. Secondly, a relationship is established between the index of biodegradability and dissolved oxygen the tank's level required to prevent the swelling. This observation is visible in the current analysis since there was a high correlation between the biodegradability index and dissolved oxygen (r = 0.541).

CONCLUSION

Our understanding of the dysfunction caused by the bulking phenomenon of the wastewater treatment plants at issue contributes to the field of environmental microbiology and sewage engineering.

The oversizing of the present plant is visible by a very low organic matter biodegradability ratio of 0.17. This gives an anchoring of events ranging from excessive filamentous bacteria proliferation (maximum of FI=6 and DSVI higher than standard of 300 mL/g). As well as, a low elimination efficiency, especially, for COD which leads to carrying out for each activated sludge treatment plant regular monitoring considering the relationship between pollution and sludge parameters of the aeration tanks.

It can be concluded that the development of filamentous bacteria was related to the malfunction indicators represented by the Filament Index (FI), biodegradability index (BOD5/COD) ratio, dissolved oxygen (O_2) in the aeration basins, and Dilute Sludge Volume Index (DSVI), which are early tools that can help to identify potential malfunction problems at an early stage and reduce high operation costs.

Studies of the last few years are dedicated to exploring more techniques (Martienssen & Adonadaga, 2015; Sheik et al., 2015; Nguyen et al., 2016; Deepnarain et al., 2020; Jiang et al., 2021) confirming that if the in-situ identification and growth rate of filamentous bacteria in the activated sludge can be determined researches can more accurately assess the effect of operating conditions on filament growth and improve the mathematical modeling of bulking. For this purpose, quantification methods of the in-situ growth rate of Sphaerotilus natans (as a model of filamentous bacteria) in activated sludge using the specific 16S rRNA/ rDNA ratio of the species are being explored.

Probably, there are few works during the dysfunction period in the presence of excessive proliferation of filamentous bacteria causing a "bulking phenomenon "at other plants based on activated sludge located in Algeria. Several factors, as well as, sewage composition, dissolved oxygen concentration, aeration system, Dilute Sludge Volume Index (DSVI) and Total Suspended Solids (TSS) concentration must be monitored on an ongoing basis. Moreover, regular observations made in the biomass of activated sludge (dry, fresh, and Gram staining) can be used to the determination of the flock structure and filaments abundance.

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