

Effects of pipe materials on biofouling under controlled hydrodynamic conditions

S. Gamri, A. Soric, S. Tomas, B. Molle and N. Roche

ABSTRACT

Experiments were carried out to investigate pipe material impacts on biofouling, at high effluent concentration levels and under controlled hydrodynamic conditions. Two velocities (0.4 and 0.8 m s⁻¹) were used to monitor biofilm growth on polyethylene (PE) and polyvinylchloride (PVC) pipe walls, respectively. These conditions were established based on wastewater irrigation practices. A decrease in biomass is observed after 49 days of experiments for both velocities and may be related to biofilm detachment. Biofilm growth is greater at 0.8 m s⁻¹. For both velocities, PVC is less sensitive to biofilm growth than PE. Pipe straightness plays a primary role in biofilm growth control. This effect is more significant than pipe surface characteristics (roughness, hydrophobic/hydrophilic properties).

Key words | biofilm, irrigation, PE, PVC, shear stress, wastewater

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ABBREVIATIONS

COD chemical oxygen demand
 PE polyethylene
 PVC polyvinylchloride
 Re Reynolds number
 TOC total organic carbon
 τ shear stress [N m⁻²]

INTRODUCTION

Biofouling is encountered in most water systems and concerns microorganism accumulation and growth on wetted surfaces. It affects industrial equipment and leads to serious operational dysfunctions as well as significant treatment costs. Biofouling has been associated with performance losses (Flemming 2002), corrosion on steel surfaces (Videla & Characklis 1992), and potential pathogenic contamination (Lehtola *et al.* 2004). Physical and chemical cleaning is required to weaken and remove foulants (Nguyen *et al.* 2012). Within the context of wastewater reuse for industry (Mohsen & Jaber 2003) and agriculture (Qadir *et al.* 2010),

numerous issues are linked to biofilm development. Biofouling has been specifically identified as an important cause of drip-irrigation system clogging when distributing wastewater (Tarchitzky *et al.* 2013; Gamri *et al.* 2014). Biofilm grows on emitters (Carpa & Scicolone 2004) and pipes (Li *et al.* 2012) and is generally associated with physical and chemical deposits (Adin & Sacks 1991). Previous studies have proved biofilm resistance to disinfection (De Beer *et al.* 1994). Biocides are more efficient against suspended microorganisms than against biofilms. Therefore, alternative measures should be explored in order to control biofilm growth.

Biofilm development is a complex phenomenon affected by several environmental conditions (Characklis 1981) (e.g., nutrient availability, temperature, surface roughness, and hydrodynamic conditions). The understanding of these parameters is crucial to develop anti-fouling strategies. Various studies have been carried out to investigate the impact of pipe materials on biofilm development in drinking water distribution systems (Schwartz *et al.* 1998; Niquette *et al.* 2000; Hallam *et al.* 2001; Van der Kooij *et al.* 2005). Bacterial adhesion is influenced by chemical properties on surfaces

(Lehtola *et al.* 2004), roughness (Yu *et al.* 2010), and hydrophobicity (Hogt *et al.* 1983). Roughness is often amplified by metal surface corrosion (Beech & Sunner 2004), while non-corroded copper surfaces release substances which may inhibit bacteria attachment and slow down biofilm growth (Lehtola *et al.* 2004; Van der Kooij *et al.* 2005; Morvay *et al.* 2011).

Pipe material impacts on biofilm development have been analyzed by collecting samples from drinking water networks (Schwartz *et al.* 1998; Niquette *et al.* 2000) or by incubating pipe coupons in bacterial suspensions (Traczewska & Sitarska 2009; Yu *et al.* 2010). In both cases, hydrodynamic conditions have rarely been taken into account. In addition, water quality used in these studies generally meets drinking water standards. The aim of the present study is to explore the effects of common plastic pipe materials used in irrigation on biofouling within the context of wastewater reuse. Biofilm growth is studied at high effluent concentration levels and under controlled hydrodynamic conditions.

MATERIALS AND METHODS

Experimental set-up

The experimental study was performed using a pipe network under controlled flow conditions. The experimental set-up (Figure 1) was made up of independent systems in order to simultaneously study different pipe materials in different flow conditions. Each system was composed of a 0.1 m³ reservoir with a pump which supplied the feeding solution detailed in the section ‘Water quality analysis’ to the system and operated for 8 hours a day in a closed circuit. The tanks were covered to prevent solution exposure to light so as to limit algae development within the system.

Two plastic materials, polyethylene (PE) and polyvinylchloride (PVC), were selected because of their common use in irrigation systems. A single external pipe diameter (16 mm) was also used in order to maintain similar flow conditions for both materials. This pipe size is commonly used in irrigation. Five meter length pipes were placed at 1.5 meters above the ground at a 10% slope to prevent air entrapment in pipes and to facilitate draining. Each line was divided

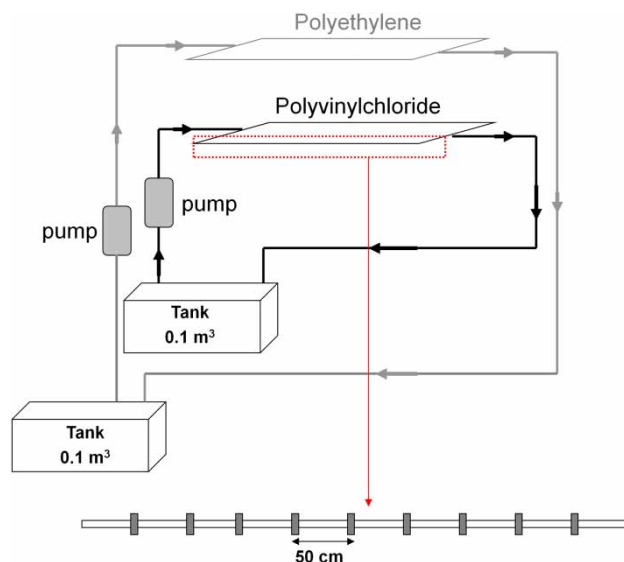


Figure 1 | Scheme of the experimental set-up.

into ten 50 cm sections. Connectors were designed to not disturb the water flow profile in pipes. Four lines were set up in order to monitor biofilm growth in PE and PVC pipes at 0.4 m s⁻¹ (Re = 5,700; $\tau = 0.7 \text{ N m}^{-2}$) and 0.8 m s⁻¹ (Re = 11,500; $\tau = 2.4 \text{ N m}^{-2}$) based on irrigation practices.

Experimental conditions are summarized in Table 1. Two experiments were carried out to investigate the impact of PE pipe configuration (experiment E1) and hydrodynamic conditions (experiment E2) on biofilm development. For experiment E1, all the sections were removed and analyzed at the end of the experiment to compare biofilm growth on straight and distorted pipes. For experiment E2, biofilm

Table 1 | Summary of the experiments performed

Experiment	Configuration	PE velocity [m s ⁻¹]		PVC velocity [m s ⁻¹]		Duration
		0.4	0.8	0.4	0.8	
E1	5 m length lines with 10 sections; straight and distorted PE pipes Biofilm sampling and analysis performed at the end of the experiment to analyze biofilm growth on PE (straight and distorted) and PVC pipes	0.4	0.8	0.4	0.8	4 weeks (8 h d ⁻¹)
E2	5 m length lines with 10 sections Weekly biofilm sampling and analysis to monitor biofilm growth under hydrodynamic conditions	-	-	0.4	0.8	10 weeks (8 h d ⁻¹)

development was monitored weekly by removing a 50 cm pipe section and by sampling the biofilm. Pipe sections were removed from upstream to downstream at the same location for each line of the experimental set-up. The removed sections were replaced by new ones.

In this study, we investigated the impact of configuration on biofilm growth in 16 mm PE pipe diameter. In field conditions, PE pipes are installed without any fasteners which causes local distortions (Figure 2(a)). Configuration effects are amplified by field topography. For experiment E1, five PE pipe sections were fastened to reduce distortion effects (Figure 2(b)). The five others were placed without any fasteners which causes a number of distortions. Such a configuration facilitates biofilm monitoring in conditions that are similar to those occurring in the field where the pipes are rarely straight.

Water quality analysis

A 200 mg L⁻¹ chemical oxygen demand (COD) synthetic effluent was used during the experiments to hasten microorganism growth. Its composition was the same used in Gamri *et al.* (2014). The components were diluted with 10 µm filtered non-chlorinated water. Filtration removes solid particles which may deposit in pipes; as the objective is to prevent the interaction between these particles and biofilm. The solution was renewed weekly after tank cleaning to ensure a continuous nutrient supply for bacteria. Water

quality characteristics are detailed in Table 2. The stationary water flow influences temperature variations. In experiment 1, for instance, we measured 28.4 ± 2.5 °C when the system is functioning and 23.5 ± 2.7 °C during shutdown phases. These variations are significantly less than temperature variations in fields where pipes are directly exposed to the sun. In such cases, water temperature inside pipes can reach 50 °C in summer. Nevertheless, for our experiments, we only focused on the impact of pipe materials and hydrodynamic conditions on biofouling. The temperature at which the experiments are performed is thus chosen to favor and then easily quantify biofilm growth.

Biofilm sampling

Biofilm was collected using a piston-shaped tool consisting of an elastomeric membrane attached to the end of a metal rod. This allows biofilm scarping to collect samples without abrading the inner pipe walls. Biofilm samples were dried at 105 °C for 24 hours. The samples were cooled in a desiccator and then weighed on a precision scale (±0.001 g).

RESULTS

Biofilm formation on plastic pipes

Biofilm formation on distorted and straight PE pipes

To analyze biofilm growth on distorted and straight pipes, some PE pipe sections were fastened to reduce distortion effects in experiment E1. Dried biomasses measured in straight and distorted PE pipes are detailed in Table 3. After PE pipe

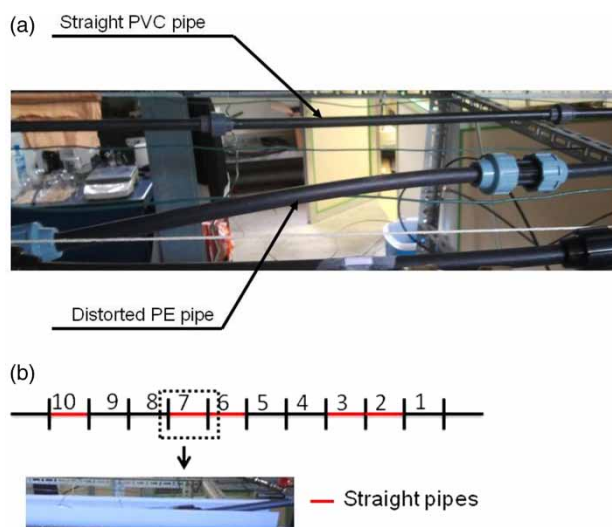


Figure 2 | PE and PVC configuration (a) and PE pipe fastening (b) in experiment E1.

Table 2 | Inlet water quality characteristics

Parameter	Unit	Experiment E1 mean value	Experiment E2 mean value
Total organic carbon	mg L ⁻¹	80.6 ± 17.9	89.3 ± 13.3
Temperature	°C	28.4 ± 2.5	27.8 ± 3.1
Conductivity	µS cm ⁻²	820 ± 75	790 ± 30
pH	-	7.5 ± 0.3	7.5 ± 0.3
Oxygen	mg L ⁻¹	6.8 ± 1.9	6.1 ± 2.3

fastening, we observed a 16-fold decrease in biofilm quantities at 0.4 m s^{-1} and a six-fold decrease at 0.8 m s^{-1} .

Moreover, by considering in detail experiment E1 results, we observed heterogeneous biofilm growth between distorted and straight pipes. A wide discrepancy in biofilm growth between the three upstream sections was recorded (Figure 3(a)). Biofilm quantities collected in the second section are clearly more significant than for the two others. Dried biomass quadrupled between the first and the second section at 0.4 m s^{-1} and was seven times greater at 0.8 m s^{-1} . Pipe section configuration may explain the variations observed in biofilm quantities (Figure 3(b)). The first section upstream of the experimental set-up is curved which did not facilitate drainage of the system.

Biofilm growth on PE and PVC surfaces

Dried biomasses were measured after 4 weeks of experiment E1 in straight PE and PVC sections, and are shown in Figure 4. At 0.8 m s^{-1} , the difference between dried biomasses in straight PE and PVC is insignificant except for

Table 3 | Dried biomass measured on distorted and straight PE pipes

Velocity (m s^{-1})	Mean biofilm dry mass on distorted pipes (mg cm^{-2})	Mean biofilm dry mass on straight pipes (mg cm^{-2})
0.4	0.881 ± 0.392	0.055 ± 0.017
0.8	0.925 ± 0.528	0.161 ± 0.102

sample 7 where biofilm growth in straight PE is twice that observed in PVC (Figure 4(a)). Average dried biomass measured in straight PE was 24% higher than that measured in the same PVC sections ($0.119 \pm 0.044 \text{ mg cm}^{-2}$ in PE and $0.09 \pm 0.03 \text{ mg cm}^{-2}$ in PVC). The discrepancy between straight PE and PVC reaches 40% at 0.4 m s^{-1} ($0.055 \pm 0.017 \text{ mg cm}^{-2}$ in straight PE and $0.033 \pm 0.015 \text{ mg cm}^{-2}$ in PVC) (Figure 4(b)). For straight configuration, PE provides more favorable conditions for biofilm development than PVC.

Biofilm growth under different flow velocity conditions

The results of experiment E1 for straight PE sections (Figure 5) show that biofilm growth is more significant at 0.8 m s^{-1} when compared to 0.4 m s^{-1} . Average dried biomass is two times greater at 0.8 m s^{-1} (0.119 ± 0.044 at 0.8 m s^{-1} and 0.056 ± 0.017 at 0.4 m s^{-1}). Biofilm development is highly influenced by hydrodynamic conditions and is more significant in high velocity conditions. For PVC, dried biomass is also 50% greater at 0.8 m s^{-1} than at 0.4 m s^{-1} .

Figure 6 shows dried biomasses in PVC pipes over the ten weeks of experiment E2 at 0.4 and 0.8 m s^{-1} . Average biofilm mass in PVC pipes is six times higher at 0.8 m s^{-1} than at 0.4 m s^{-1} . The average dried biomass is about $0.005 \pm 0.003 \text{ mg cm}^{-2}$ at 0.4 m s^{-1} , while it is $0.033 \pm 0.031 \text{ mg cm}^{-2}$ at 0.8 m s^{-1} . Moreover, an increase in

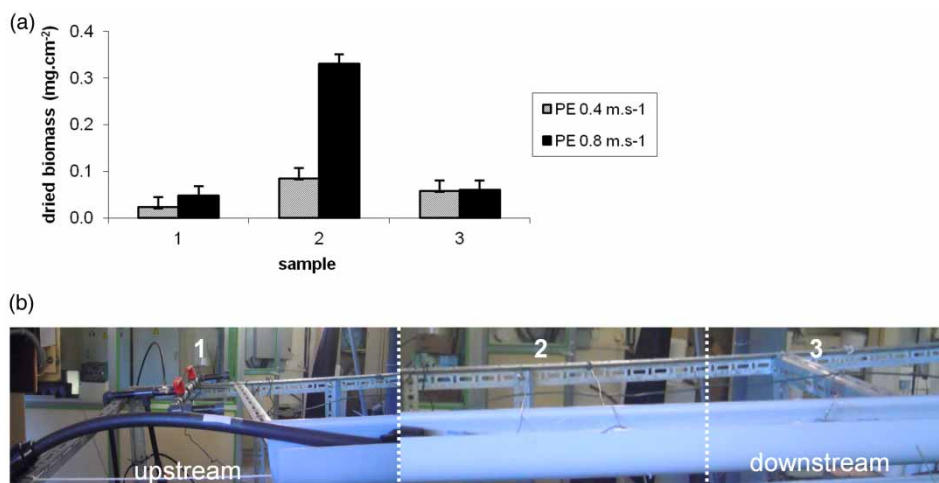


Figure 3 | Dried biomass collected in different PE sections (a) and its configuration (b) during experiment E1.

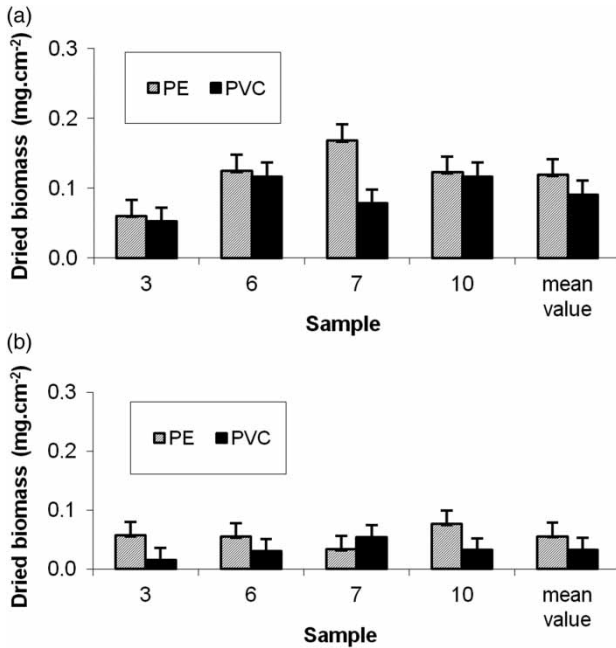


Figure 4 | Dried biomasses measured in PVC and straight PE pipes (a) at 0.8 m s⁻¹ and (b) 0.4 m s⁻¹ after 28 days of the experiment E1.

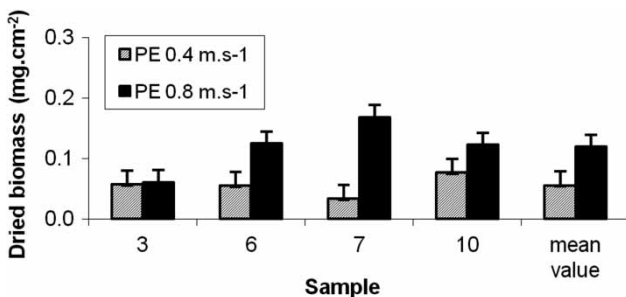


Figure 5 | Biofilm dry mass measured at 0.8 m s⁻¹ and 0.4 m s⁻¹ in straight PE pipes after 28 days of experiment E1.

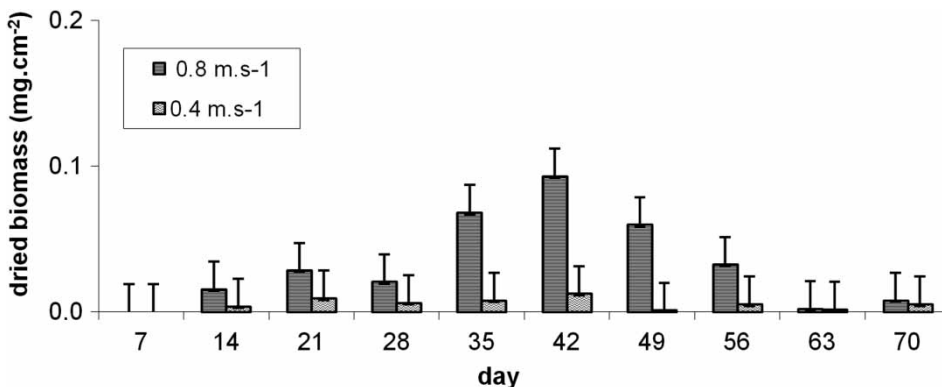


Figure 6 | Dried biomasses measured at 0.8 m s⁻¹ and 0.4 m s⁻¹ in PVC during experiment E2.

biomass is observed until day 49 when a decrease occurs. At the end of the experiment, dried biomass decreased by 90% and 60% at 0.8 m s⁻¹ and 0.4 m s⁻¹, respectively.

DISCUSSION

Configuration impacts

The impact of pipe configuration has been investigated by comparing biofilm growth in distorted and straight pipe sections, some of which were fastened (experiment E1). PE fastening has greatly reduced biofilm development (up to 16 times at 0.4 m s⁻¹). Thus, pipe fastening helps to reduce biofilm growth. Pipe distortions may induce water accumulation during shutdown phases which increase microorganism growth potential.

Furthermore, biomass measured in the second section is obviously more significant when compared to the first and third ones. The first section upstream of the experimental set-up is curved (Figure 3(b)). This favors water accumulation in the second section of the experimental set-up when the pump shuts down. Owing to pipe slope, water accumulation was probably reduced from the third section to the experimental set-up downstream which helped pipe drainage. In the first pipe section, water accumulation is impossible because of its configuration. Therefore, low biofilm levels were measured.

It appears that pipe configuration plays an important role in biofilm development. Water accumulation during shutdown phases helps maintain a wet environment which

fosters biofilm growth. However, few studies have investigated this effect on biofilm growth. [Momba & Kaleni \(2002\)](#) observed high microorganism concentration during biofilm sampling in PE tanks used to store drinking water. In this experiment, some residual water was observed in distorted sections where high biomasses were measured. This attests water accumulation in PE pipes after pump shutdown which is conducive to bacterial growth.

Pipe surface impacts

The average biomasses obtained in straight PE are 24% higher at 0.8 m s^{-1} and 40% higher at 0.4 m s^{-1} than for PVC. This confirms the results obtained by [Yu *et al.* \(2010\)](#), where the biofilm quantities observed on PE coupons were 30–40% higher than those measured on the PVC coupons after 90 days of incubation in drinking water. In drinking water networks, biofilm development is more significant for PE than in PVC ([Van der Kooij *et al.* 1995](#); [Traczewska & Sitarska 2009](#); [Yu *et al.* 2010](#)). The same trend is observed when higher COD concentration effluents are used.

In plastic materials, the discrepancy in biofilm growth has been linked to the combination of two parameters. The first one concerns component release. PE releases biodegradable organic compounds and phosphorus which promote biofilm development on plastic surfaces. [Lehtola *et al.* \(2004\)](#) observed a significant rate of phosphorus which was released from PE pipes into water for a period of 2–3 weeks. A previous study performed by [Lehtola *et al.* \(2002\)](#) showed that biofilm development is influenced by phosphorus availability in water systems. However, in these experiments, pipes have been previously used with tap water for 3 weeks to check hydraulic operation. If a release of phosphorus was observed, it should have been during tap water operations. Thus, this parameter cannot explain the difference observed between biofilm quantities in PE and PVC pipes.

The second parameter is related to surface characteristics. PE and PVC are considered as low roughness materials ([Niquette *et al.* 2000](#)) in comparison with others such as steel, iron, or cement. Therefore, they are less likely to foster biofilm development ([Traczewska & Sitarska 2009](#)). The observations made by scanning electron microscopy showed a small difference between PE and

PVC surface structure ([Yu *et al.* 2010](#)). PVC surfaces are relatively smoother than those of PE and therefore reduce microorganism adhesion. [Traczewska & Sitarska \(2009\)](#) also found that PVC surfaces are more regular than those of PE. These observations, at high magnification ($\times 6,000$), showed visible cracks at PE surfaces and were associated with its greater ability to foster biofilm development. Surface roughness may partly explain the significant biofilm development on PE surfaces.

In addition, hydrophobic materials are generally more conducive to bacterial adhesion than hydrophilic surfaces ([Hogt *et al.* 1983](#)). [Verrier *et al.* \(1987\)](#) showed, through experiments conducted on four species of methane bacteria, that PE is more hydrophobic than PVC. The results obtained, which highlight the potential of the PE to develop more biofilm than PVC, can thus be explained by surface characteristics. The roughness of PE surface, even close to the PVC one, and its hydrophobic character offer more favorable conditions for microorganism adhesion than PVC.

Impact of hydrodynamic conditions on biofilm

Biofilm development in both PE and PVC at 0.8 m s^{-1} is greater than 0.4 m s^{-1} . An increase in the number of attached bacteria and substrate consumption was observed when velocity increases ([Lehtola *et al.* 2006](#)). Substrate transfer in biofilms is then favored by high velocities. Furthermore, in the experiments performed by [Wäsche *et al.* \(2002\)](#) using a range of Reynolds numbers between 650 and 6,000 for pipes with 26 mm diameter, an increase of glucose gradients was measured with high Reynolds number. Hydrodynamic conditions promote substrate supply into biofilms, and therefore favor their development. However, high shear forces may induce biofilm detachment ([Characklis 1981](#)). Low flow velocities induce high resistance to substrate transfer (nutrients, oxygen, etc.) from bulk fluid to the microorganisms embedded in biofilms. High flow velocities cause high turbulence of fluid bulk which enhances substrate transfer and biofilm growth. Nevertheless, higher flow velocities induce high shear forces which may lead to biofilm surface erosion. In this study, the impact of hydrodynamic conditions on biofilm development was investigated using only two velocities. Our results confirm those obtained on previous studies performed in similar

experimental conditions. Nevertheless, further experimentation using additional hydrodynamic conditions is required to validate this hypothesis and to determine the shear force threshold at which biofilm detaches.

Furthermore, biofilm development was monitored for 70 days in PVC pipes (experiment E2). The results obtained confirm that biofilm growth is greater at 0.8 m s^{-1} than at 0.4 m s^{-1} over the whole experimental period. Moreover, biofilm development decreases from day 49. This decrease may be related to biofilm detachment. This phenomenon has been observed by Belkhadir *et al.* (1988) after the biofilm growth phase, when its thickness reaches a maximum value. As biofilms grow thicker, fluid shear stress at biofilm interface increases and substrate limitations are observed particularly in the part of biofilm layer closest to the pipe surface. This may weaken biofilm and lead to its detachment.

CONCLUSIONS

This study has investigated biofilm growth on PE and PVC pipes under controlled hydrodynamic conditions using a synthetic effluent containing 200 mg L^{-1} COD. Two velocities (0.4 and 0.8 m s^{-1}) were used based on irrigation practices.

Hydrodynamic conditions play a primary role in biofilm growth. Biofilm growth is favored by high velocities. Biofilm development is more pronounced at 0.8 m s^{-1} than at 0.4 m s^{-1} . High flow velocities cause high turbulence of fluid bulk which enhances substrate transfer and biofilm growth, as suggested by numerous authors working on drinking water distribution systems. In irrigation networks used for wastewater reuse, increasing pipe velocities may not be a solution to reduce biofilm development. Moreover, a decrease in biomass occurred after 49 days at both velocities and may be related to biofilm detachment. Competition is observed between biofilm growth and destruction.

For both velocities, PVC demonstrates a lower susceptibility to biofilm growth when compared to PE. The discrepancy between PE and PVC is due to pipe surface characteristics (roughness and hydrophobic/hydrophilic properties) and configuration (straight or distorted pipes).

Pipe configuration should be taken into account in biofouling studies. At 0.4 m s^{-1} , biomass levels are 40% higher

in straight pipes than PVC pipes and are up to 16 times in distorted PE pipes when compared to straight ones. Water accumulation in pipes was seen to favor higher biofilm growth. In water and wastewater networks, water accumulation can be observed under low flow conditions. Irrigation systems are characterized by discontinuous operation which results in water accumulation in distorted pipes during the system shutdown.

Our work confirms that future research should consider pipe configuration and hydrodynamic conditions in more detail in order to investigate biofouling in PE pipes. In further studies, we will focus on biofilm kinetics under different hydrodynamic conditions by monitoring biomass growth and detachment.

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