



## Bioreactors for remediation of hydrocarbons in rivers and lagoons of San Luis Potosí

## Biorreactores para remediación de hidrocarburos en ríos y lagunas de San Luis Potosí

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### Abstract

In the present work the polluting hydrocarbons in water bodies of the state of San Luis Potosí were evaluated and determined in June 2019, to choose a representative carbon source for the airlift bioreactor. The sampling points were tourist areas: Chajir Lagoon, Puente de Dios pool, Tampaón River and Media Luna Lagoon, and aliphatic hydrocarbons with a carbon number between C8-C32 were found. We reported high concentrations of pollutants, such as eicosane (up to  $2,603.22 \pm 1,202.09 \text{ mg L}^{-1}$ ) and heneicosane (up to  $3,971.21 \pm 2,260.45 \text{ mg L}^{-1}$ ). The airlift bioreactor was designed based on hydrodynamic parameters: holdup gas Sauter mean diameter and Reynolds number that increased with the superficial gas velocity, both in the riser zone and downcomer zone and from  $U_g = 1.92 \text{ cm s}^{-1}$ , mixing and mass transfer are favoured in a 2 L airlift reactor. Also, the hydrodynamic parameters evaluated were higher in the riser zone. Furthermore, the consortium isolated by the Gutiérrez-Rojas working group composed of *Xanthomonas* sp., *Acinetobacter Bouvetii*, *Shewanella* Sp., and *Defluviobacter lusatiensis*, was able to consume 98.21% of the initial diesel concentration ( $13,000 \text{ mg L}^{-1}$ ). The airlift bioreactor evaluated in the investigation proved to be capable of remediation the studied water bodies.

**Keywords:** airlift bioreactor, aliphatic hydrocarbon, remediation, pollution, hydrodynamic.

### Resumen

En el presente trabajo se determinaron hidrocarburos contaminantes en cuerpos de agua del estado de San Luis Potosí, durante el mes de junio de 2019, con el fin de escoger una fuente de carbono representativa para el biorreactor airlift. Los puntos de muestreo fueron laguna Chajir, Puente de Dios, río Tampaón y laguna de Media Luna, se encontraron hidrocarburos alifáticos con un número de carbonos entre C8-C32. Reportamos altas concentraciones de contaminantes, como eicosano (hasta  $2603.22 \pm 1202.09 \text{ mg L}^{-1}$ ) y heneicosano (hasta  $3971.21 \pm 2260.45 \text{ mg L}^{-1}$ ). Se diseñó un biorreactor airlift con base a los parámetros hidrodinámicos: gas holdup, diámetro promedio de Sauter y número de Reynolds que incrementaron con la velocidad superficial del gas. Además, los parámetros hidrodinámicos evaluados fueron más altos en la zona de ascenso y a partir de un  $U_g = 1.92 \text{ cm s}^{-1}$ . Además, el consorcio aislado por el grupo de trabajo del doctor Mariano Gutiérrez Rojas compuesto por *Xanthomonas* sp., *Acinetobacter Bouvetii*, *Shewanella* Sp. y *Defluviobacter lusatiensis* cultivado en un biorreactor airlift, pudo consumir el 98.21% de la concentración inicial del diésel ( $13000 \text{ mg L}^{-1}$ ). El biorreactor airlift evaluado demostró ser capaz de remediar la remediación de los cuerpos de aguas muestreados.

**Palabras clave:** biorreactor airlift, hidrocarburos alifáticos, remediación, hidrodinámica, contaminación.

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## 1 Introduction

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Aliphatic (AHs) and polycyclic aromatic hydrocarbons (PAHs) are worrying pollutants because of their limited water solubility, high persistent, toxicity, bioaccumulation and biomagnification (Akhbarizadeh *et al.*, 2016). Additionally, prolonged exposure to hydrocarbons may have genotoxic, carcinogenic and teratogenic effects (Ghosal *et al.*, 2016). Furthermore, some studies have even reported harmful effects in the endocrine and reproductive systems of aquatic organisms (Wang *et al.*, 2019). Although most recent studies focus on PAHs, the removal of large amounts of AHs is important to remediation goals in polluted areas (Chaudhary, Bajagain, Jeong, & Kim, 2019).

San Luis Potosí is a state with a long mining history dating from 1561 (SGM, 2018). Furthermore, the exploitation and extraction of hydrocarbons in the Ebano field were recently approved in 2018 and began operations in the beginning of 2020 (CNH, 2019), emerging as an important source of oil pollution. Moreover, studies have been carried out using 1-hydroxypyrene (1-HPO) as a biomarker to pyrene in urine samples women and children from vulnerable rural populations, indicating presence of PAHs generated from the burning of biomass for heating and cooking in the homes of these communities (Martínez-Salinas *et al.*, 2010; Palacios-Ramírez *et al.*, 2018). Furthermore, in research published in 2016, *trans, trans*-muconic acid (t, t-MA) was used as biomarker to benzene and was detected in children from rural communities in San Luis Potosí (Flores-Jiménez *et al.*, 2016). Moreover, burning sugarcane is an important hydrocarbon source (pyrogenic) in the Huasteca Potosina (Flores-Jiménez *et al.*, 2016), and this could be more relevant than possible hydrocarbons petrogenic sources for adjacent communities (Maioli, Knoppers, & Azevedo, 2009).

An important alternative to hydrocarbon remediation is bacterial bioremediation. For example, UAMI consortium constituted by *Acinetobacter bouvetii*, *Xanthomonas* sp., *Shewanella* sp. and *Deftuvibacter lusatiensis*, isolated from *Cyperus laxus*'s rhizosphere, in an oil-contaminated swamp in Mexico (Díaz-Ramírez *et al.*, 2003; Sandoval-Herazo *et al.*, 2020) or marine bacteria like *Arthrobacter bambusae* able to uptake phenol (Zakaria *et al.*, 2020). Subsequently, the Gutiérrez-Rojas working group showed the UAMI consortium ability to uptake Maya

crude oil (Medina-Moreno, *et al.*, 2005). Furthermore, the UAMI consortium has shown that it can uptake diesel, hexadecane, pyrene and phenanthrene using airlift bioreactors (Hernández-Martínez *et al.*, 2019; Medina-Moreno *et al.*, 2013; Nápoles-Álvarez *et al.*, 2017). Meanwhile, the efficiency of consortiums can be improved by using bioreactors, such as airlift bioreactors (Nápoles-Álvarez *et al.*, 2017; Suárez-García *et al.*, 2019). The airlift bioreactors (ALB) are a low-cost alternative used in remediation of hydrocarbons (Manowattana *et al.*, 2018; Nápoles-Álvarez *et al.*, 2017). Furthermore, ALB has advantages over other types of reactors such as bubble column and stirred tank: reduced cell damage, high aeration rate and more significant mass transfer (Gamboa-Suasnavart *et al.*, 2019; Sandoval-Herazo *et al.*, 2020). The hydrodynamics highly influences efficiency of ALB in the gas phase (evaluated with gas holdup and Sauter mean diameter) (Jamshidi & Mostoufi, 2017). Such turbulent flow ALB is studied because of favour mixing and mass transfer (Cunha *et al.*, 2016; Pawar, 2017).

Although there are studies that document the presence of hydrocarbons in indigenous communities of San Luis Potosí, hydrocarbon pollution in vulnerable environments which are indispensable a resource for the communities and ecosystems of the region, such as water bodies has not been studied. Likewise, one aggravating factor is the lack of remediation proposals because they are an important point for solving or mitigating this problem. The aim of this research was to design airlift bioreactors based on the evaluation of hydrodynamic parameters and results sampling hydrocarbons in rivers and lagoons in San Luis Potosí state.

## 2 Materials and methods

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### 2.1 Evaluation of hydrocarbon pollution in water bodies of San Luis Potosí

#### 2.1.1 Sampling points

In Huasteca Potosina, four natural water bodies with a large influx of tourist were sampled (Fig. 1): Chajir Lagoon, near Ebano (22°06'31.5"N, 98°30'55.7"W), Media Luna Lagoon, touristic and agricultural zone (21°51'39.5"N, 100°01'37.5"W), Puente de Dios waterfall, touristic zone (21°55'50.4"N, 99°24'59.5"W) and Tampaon River in Aquismon

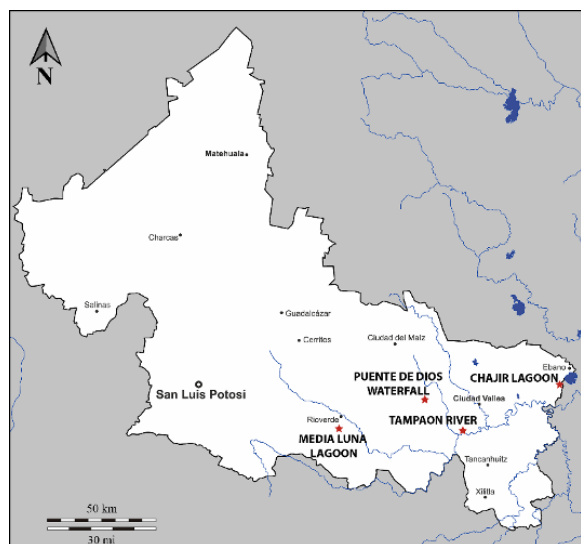


Fig. 1. Sampling points, state of San Luis Potosí.

touristic zone ( $21^{\circ}47'43.4''\text{N}$ ,  $99^{\circ}08'50.9''\text{W}$ ). Samples were taken from surface waters in wide-mouth amber glass containers (full and without bubbles) from waters at a depth of between 1 and 1.5 m and transported to the laboratory at a low temperature ( $4^{\circ}\text{C}$ ). The sampling was carried out according to the Mexican normative (NMX-AA-014-1980) and Mexican normative project (PROY-NMX-AA-121/1-SCI-FI-2008).

### 2.1.2 Evaluation of aliphatic hydrocarbons

The evaluation of aliphatic hydrocarbons was carried out by gas chromatography (Trace 1310- TRIPLUS, Thermo Scientific USA) using the next operative conditions. Table 1 shows the operating parameters for chromatographic analysis (Tec-Caamal et al., 2018). The aliphatic hydrocarbon reference (ASTM D2887-12, RESTEK) was used.

## 2.2 Airlift bioreactor design and operation for water remediation

Four different superficial gas velocities ( $U_g$ ) (0.76, 0.88, 1.92 and  $3.18 \text{ cm s}^{-1}$ ) were used evaluating

Reynolds number, gas holdup, Sauter mean diameter and superficial aqueous phase velocity of bioreactor airlift. A 2 L internal loop airlift bioreactor of cylindrical glass was used. The draft tube is 5.98 cm in diameter ( $D_2$ ) and 17.27 cm ( $L_2$ ) in height, while the external diameter is 9.2 cm ( $D_1$ ) and the external height is 22.56 cm ( $L_1$ ). With a geometric relation of  $D_2/D_1 = 0.65$  and  $L_2/L_1 = 0.77$  (Lizardi-Jiménez & Gutiérrez-Rojas, 2011) geometric parameters whose efficiency has been previously evaluated (Sandoval-Herazo et al., 2020).

### 2.2.1 Hydrocarbonoclastic bacterial consortium

A hydrocarbonoclastic consortium was used which was composed of *Acinetobacter bouvetii*, *Shewanella* sp., *Deftuvibacter lusatiensis* and *Xanthomonas* sp. The consortium was isolated from the rhizosphere of *Cyperus laxus*, a hydrocarbon pollution tolerant plant native to the swamps of southern Mexico. The culture grew in mineral medium, consisting of ( $\text{g L}^{-1}$ ): 6.75  $\text{NaNO}_3$ , 2.15  $\text{K}_2\text{HPO}_4$ , 1.13  $\text{KCl}$  and 1.10  $\text{MgSO}_4 \cdot 5\text{H}_2\text{O}$ . pH was adjusted to 6.5 (Tec-Caamal et al., 2018), 13000  $\text{mg L}^{-1}$  of diesel was used as a carbon source.

### 2.2.2 Hydrodynamic parameters and operational conditions

Aqueous phase velocity ( $V_{aq}$ ) and diesel phase velocity ( $V_{diesel}$ ) were experimentally evaluated to study the flow patterns in the model medium, sodium polyacrylate hydro-gel ( $\rho = 1.0 \text{ g cm}^{-3}$ ) and oligosyloxane stained spheres ( $\rho = 0.77 \text{ g cm}^{-3}$ ) were used to simulate water and oil, respectively. A digital stopwatch and video camera were used evaluate velocities of each individual sphere. (Lizardi-Jiménez & Gutiérrez-Rojas, 2011). The relationship between height and elapsed time was quantified in both zones: downcomer and riser. The measuring was recording in video, and a digital chronometer was used to measure time.  $V_{aq}$  y  $V_{diesel}$  ( $\text{cm s}^{-1}$ ) were used to calculation the Reynolds numbers (Re) for each case, using the following equations:

Table 1. Operating parameters for chromatographic analysis.

Descriptions	Conditions
Column	TR-5 column (30 cm x 0.32mm)
Carrier gas	Helium
Flame ionization detector temperature	300 °C
Injector temperature	290 °C
Oven temperature program	45 °C, hold 1 min; 45 to 110 °C at $5 \text{ }^{\circ}\text{C min}^{-1}$ ; 110 °C to 330 °C at $15 \text{ }^{\circ}\text{C min}^{-1}$ .

$$Re_{aq} = \frac{DV_{aq}\rho_{aq}}{\mu} \quad (1)$$

$$Re_{diesel} = \frac{DV_{diesel}\rho_{diesel}}{\mu} \quad (2)$$

where  $Re_{aq}$  and  $Re_{diesel}$  are aqueous and diesel phase Reynolds numbers, respectively.  $D = D2$  for the riser zone, and  $D = (D1-D2)$  for the downcomer zone.  $D1$  is the ALB external diameter, cm;  $D2$  is draft tube diameter, cm;  $\rho_{aq}$  (aqueous phase density,  $\text{g cm}^{-3}$ );  $\rho_{diesel}$  is diesel phase density,  $\text{g cm}^{-3}$ , and  $\mu$  bulk viscosity is oil in water emulsion,  $\text{g cm s}^{-1}$ .

The gas holdup ( $\varepsilon_G$ ) was estimated by measuring the expanded bed. This method involved measuring the surface height when air flowed through the column and comparing it to the repose height (Besagni & Inzoli, 2016). The gas holdup was calculated based on the following equation:

$$\varepsilon_G = \frac{H_t - H_s}{H_t} \quad (3)$$

where  $H_s$  is the liquid height without aeration and  $H_t$  is the liquid height with aeration.  $\varepsilon_G$  was calculated by its relationship with several hydrodynamic parameters (density, viscosity of mixture and surface tension).

The Sauter mean diameter ( $d_{32}$ ) for water and diesel was calculated using a method of digital image analysis (Tec-Caamal *et al.*, 2018; Yang *et al.*, 2018). There were ten photos taken for each condition to a height of 12 cm from the top of the downcomer with a digital camera, and they were analysed using software (ImageJ v1.8.0, USA). A reference solid with known dimensions was used to calculate  $d_{32}$  in the ALB inner part. Using a correlation factor, the measurement error was avoided. The Sauter mean diameter was calculated for each  $U_g$  using equation 4:

$$d_{32} = \frac{\sum_{i=1}^k n_i x_i^3}{\sum_{i=1}^k n_i x_i^2} \quad (4)$$

where  $n_i$  is the droplet number for each specific diameter ( $x_i$ ). To calculate  $d_{32}$ , an abiotic medium was designed, simulating a three-phase medium (gas, aqueous and diesel). The aqueous phase and diesel phase have a viscosity ( $\mu$ ) of  $0.00871 \text{ g cm}^{-1} \text{ s}^{-1}$  and  $0.0105 \text{ g cm}^{-1} \text{ s}^{-1}$ , respectively. The  $d_{32}$  was used to describe the preferment droplet size and to calculate the bubble rise velocity ( $U_b$ ) using equation 5.

$$U_b = 1.3 \sqrt{gd_{32}} \quad (5)$$

where  $g$  is the constant gravity ( $980 \text{ cm s}^{-2}$ ).

### 2.2.3 Residual diesel and suspended solids

10 mL samples of ALB were taken and were centrifuged at  $4,000 \times g$  for 30 min at  $4^\circ\text{C}$ . There were three phases formed: diesel, aqueous phase and solid phase. Suspended solids that include the microbial consortium were measured in the solid phase after being heated in a low-pressure oven at  $60^\circ\text{C}$  for 48 hours. The diesel phase and the aqueous phase were used to determine the residual diesel, whereby they were subjected to an extraction 1:1 with hexane and diesel concentrations in samples were analysed by gas chromatography (Medina-Moreno, *et al.*, 2014).

## 3 Results and discussion

### 3.1 Evaluation of hydrocarbon pollution in water bodies of San Luis Potosí

In the month of June 2019, in Chajir Lagoon, Media Luna, Tampaón River and Puente de Dios, AHs harmful to environmental and human health with carbon number between  $C_8$  and  $C_{32}$  were found (Table 2), such as eicosane ( $44.41 \pm 15.85$ - $2,603.22 \pm 1,202.09 \text{ mg L}^{-1}$ ), pentadecane ( $0.38 \pm 0.08$ - $36.83 \pm 8.02 \text{ mg L}^{-1}$ ) and octacosane ( $15.69 \pm 0.32$ - $25.34 \pm 0.18 \text{ mg L}^{-1}$ ). In Chajir lagoon, aliphatic hydrocarbons were found between  $C_8$ - $C_{32}$ , except for dodecane ( $C_{12}$ ). The hydrocarbon with a higher concentration was eicosane ( $C_{20}$ ) ( $622.30 \pm 159.28 \text{ mg L}^{-1}$ ); furthermore, heptacosane ( $15.24 \pm 3.99 \text{ mg L}^{-1}$ ) and octacosane ( $25.34 \pm 0.18 \text{ mg L}^{-1}$ ) concentrations were higher than  $15 \text{ mg L}^{-1}$ , which, according to the Mexican official standard, is the maximum permissible limit of wastewater pollutant discharges into freshwater (NOM-143-SEMARNAT-2003). Moreover, in Tampaón River, AHs were found between  $C_8$ - $C_{32}$ ; pentadecane ( $36.83 \pm 8.02 \text{ mg L}^{-1}$ ) nonadecane ( $18.16 \pm 3.45 \text{ mg L}^{-1}$ ) eicosane ( $2603.22 \pm 1202.09 \text{ mg L}^{-1}$ ), heneicosane ( $3971.21 \pm 2260.45 \text{ mg L}^{-1}$ ) and heptacosane ( $17.85 \pm 0.09 \text{ mg L}^{-1}$ ) concentrations were higher than the maximum permissible limit of the Mexican official standard (LMOS). Meanwhile, in Puente de Dios, all AHs ( $C_8$ - $C_{32}$ ) except dodecane ( $C_{12}$ ) were found, and only eicosane ( $44.41 \pm 15.85 \text{ mg L}^{-1}$ ) and octacosane ( $15.69 \pm 0.32 \text{ mg L}^{-1}$ ) exceeded LMOS. In Media Luna lagoon, only two AHs exceeded LMOS: eicosane ( $333.40 \pm 116.11 \text{ mg L}^{-1}$ ) and octacosane ( $17.57 \pm 0.46 \text{ mg L}^{-1}$ ).

Table 2. Hydrocarbons in rivers and lagoons of San Luis Potosí. Sampling points: Chajir lagoon, Puente de Dios, Tampaon River and Media Luna lagoon. Sampling date (June 2019).

Hydrocarbons	Sampling points (concentration mg L <sup>-1</sup> )			
	Chajir lagoon	Puente de Dios	Tampaon River	Media Luna lagoon
Octane (C <sub>8</sub> )	0.13 ± 0.02	0.10 ± 0.10	0.08 ± 0.01	0.12 ± 0.01
Nonane (C <sub>9</sub> )	0.55 ± 0.05	0.45 ± 0.45	0.40 ± 0.02	0.51 ± 0.02
Decane (C <sub>10</sub> )	1.11 ± 0.06	1.33 ± 0.26	0.86 ± 0.04	0.98 ± 0.03
Undecane (C <sub>11</sub> )	0.94 ± 0.06	0.75 ± 0.02	0.52 ± 0.01	0.85 ± 0.02
Dodecane (C <sub>12</sub> )	-	-	0.67 ± 0.12	-
Tridecane (C <sub>13</sub> )	0.53 ± 0.01	0.40 ± 0.01	0.45 ± 0.00	0.45 ± 0.01
Tetradecane (C <sub>14</sub> )	0.84 ± 0.04	0.54 ± 0.01	0.71 ± 0.02	0.60 ± 0.01
Pentadecane (C <sub>15</sub> )	3.53 ± 0.99	0.38 ± 0.08	36.83 ± 8.02	1.71 ± 0.62
Hexadecane (C <sub>16</sub> )	0.61 ± 0.17	0.23 ± 0.07	3.83 ± 0.62	0.30 ± 0.06
Heptadecane (C <sub>17</sub> )	2.01 ± 0.54	-	3.60 ± 1.77	0.87 ± 0.27
Octadecane (C <sub>18</sub> )	3.01 ± 0.83	0.22 ± 0.11	5.62 ± 0.28	1.33 ± 0.44
Nonadecane (C <sub>19</sub> )	4.06 ± 1.20	0.34 ± 0.09	18.16 ± 3.45	1.89 ± 0.64
Eicosane (C <sub>20</sub> )	622.30 ± 159.28	44.41 ± 15.85	2603.22 ± 1202.09	333.40 ± 116.11
Heneicosane (C <sub>21</sub> )	1.20 ± 0.26	-	3971.21 ± 2260.45	-
Docosane (C <sub>22</sub> )	1.86 ± 0.41	0.17 ± 0.07	2.97 ± 0.60	0.35 ± 0.07
Tricosane (C <sub>23</sub> )	11.20 ± 2.69	0.56 ± 0.28	1.86 ± 0.69	-
Tetracosane (C <sub>24</sub> )	2.84 ± 0.73	0.38 ± 0.09	13.49 ± 6.88	1.32 ± 0.40
Pentacosane (C <sub>25</sub> )	1.75 ± 0.37	0.18 ± 0.07	0.37 ± 0.13	-
Hexacosane (C <sub>26</sub> )	3.96 ± 0.22	3.00 ± 0.07	3.26 ± 0.06	3.35 ± 0.11
Heptacosane (C <sub>27</sub> )	15.24 ± 3.99	1.01 ± 0.39	7.82 ± 1.62	0.23 ± 0.14
Octacosane (C <sub>28</sub> )	25.34 ± 0.18	15.69 ± 0.32	17.85 ± 0.09	17.57 ± 0.46
Nonacosane (C <sub>29</sub> )	2.22 ± 0.19	1.01 ± 0.02	1.40 ± 0.16	1.07 ± 0.03
Triacosane (C <sub>30</sub> )	0.24 ± 0.13	0.33 ± 0.01	0.71 ± 0.10	0.20 ± 0.09
Hentriacosane (C <sub>31</sub> )	0.26 ± 0.15	-	0.70 ± 0.03	-
Dotriacosane (C <sub>32</sub> )	5.16 ± 0.37	0.28 ± 0.18	1.05 ± 0.06	0.10 ± 0.00

The sample points were near Ciudad Valles and Río Verde, both with gravel mining (SGM, 2018). A study conducted on the Tisza river, Romania, related gravel mining activity to hydrocarbon pollution of petrogenic origin and burning of wood and coal used in home heating as a source pyrogenic hydrocarbons (Beldean-Galea *et al.*, 2016). Burning of wood and coal was also identified as a source of hydrocarbon pollution in the studies of Martínez-Salinas *et al.*, 2010 and Palacios-Ramírez *et al.*, 2018 in San Luis Potosí. Furthermore, the production of sugarcane in Huasteca Potosina is an important source of hydrocarbons that are produced by burning sugarcane, mainly for the sampling point of Tampaon River that is within the sugarcane zone (Flores-Jiménez *et al.*, 2016).

### 3.2 Airlift bioreactor design and operation for water bodies remediation

#### 3.2.1 Hydrodynamic parameters and operational conditions

Gas holdup or gas phase retention coefficient ( $\epsilon_G$ ) is an important criterion for understanding hydrodynamic behaviour and design of ALB (Bertollo *et al.*, 2017). In Fig. 2 and 3, the variations for gas-liquid flow and gas-liquid-diesel flow ALB, respectively, are shown as a function of the next superficial gas velocities ( $U_g$ ) 0.76, 0.88, 1.92 and 3.18 cm s<sup>-1</sup>. The  $\epsilon_G$  in gas-liquid flow ALB (Fig. 2) for a downcomer zone varied from 0.22 to 0.33 cm and varied with the increase of  $U_g$ . Nevertheless, in the riser zone, an increase of  $\epsilon_G$  higher than in the downcomer zone was observed (from 0.25 to 0.43 cm). In Fig. 3, corresponding to gas-liquid-diesel flow ALB, in a downcomer zone,  $\epsilon_G$  between 0.19 and 0.25 cm was

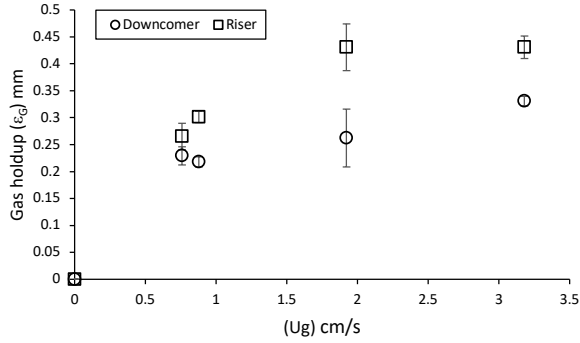


Fig. 2. Gas holdup ( $\epsilon_G$ ) as a function of superficial gas velocity ( $U_g$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid flow (two-phase).

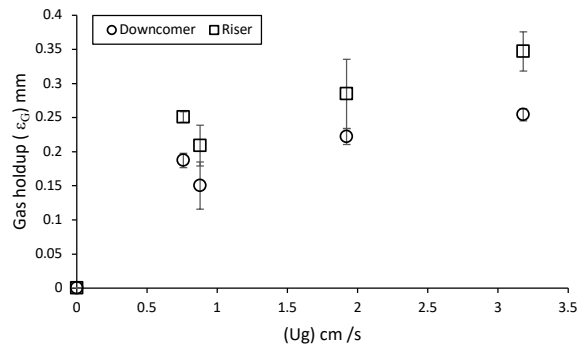


Fig. 3. Gas holdup ( $\epsilon_G$ ) as a function of superficial gas velocity ( $U_g$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid-diesel flow (three-phase).

observed. Furthermore, in the riser zone,  $\epsilon_G$  were higher than in the downcomer zone (from 0.21 and 0.35 cm). To summarise,  $\epsilon_G$  in both riser zones (gas-liquid flow ALB and gas-liquid-diesel flow ALB) were higher than in downcomer zones. Likewise, this indicates that in the downcomer zone, the coalescent rate was lower than in the riser zone, because  $\epsilon_G$  in downcomer zones were lower than in riser zones (Jamshidi & Mostoufi, 2017). The difference between  $\epsilon_G$  in riser versus downcomer zones is related to the difference in hydrostatic pressure at the ALB bottom (Lizardi-Jiménez & Gutiérrez-Rojas, 2011). Moreover,  $\epsilon_G$  increases in the riser and downcomer zones were directly proportional to the  $U_g$  increases, as previously reported (Mendes & Badino, 2016). Also,  $\epsilon_G$  in diesel were lesser than  $\epsilon_G$  in gas-liquid flow ALB, in both the riser and downcomer zones.

Fig. 3 shows changes in the Sauter mean diameter ( $d_{32}$ ) as a function of  $U_g$  in gas-liquid flow ALB.  $d_{32}$  values in the riser zone varied between 3.40-6.26 mm. The values increased as  $U_g$  increased; moreover, in the downcomer zone (from 1.38 to 2.49 mm), the

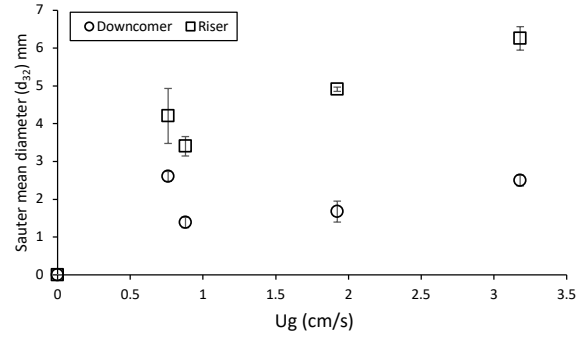


Fig. 4. Sauter mean diameter ( $d_{32}$ ) as a function of superficial gas velocity ( $U_g$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid flow (two-phase).

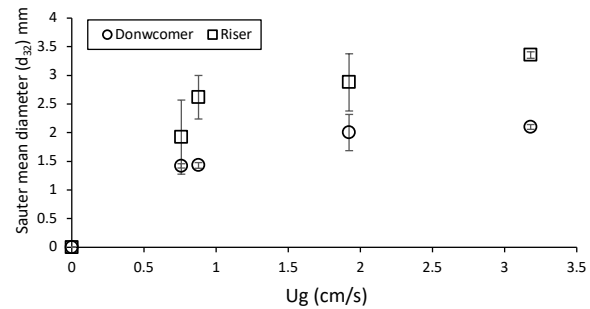


Fig. 5. Sauter mean diameter ( $d_{32}$ ) as a function of superficial gas velocity ( $U_g$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid-diesel flow (three-phase).

same behaviour was shown, but the values were lower than register values in riser zone. In the same way, in Fig. 4, increased  $d_{32}$  in the riser zone as a function of  $U_g$  of gas-liquid-diesel flow ALB was shown. The  $d_{32}$  in the riser zone varied from 1.92 to 3.35 mm and varied from 1.42 to 2.10 mm in the downcomer zone, whereby, in the downcomer zone, the values of  $d_{32}$  were lower than in the riser zone of gas-liquid-diesel flow ALB, but both had the same behaviour. Similarly,  $d_{32}$  increased when  $U_g$  increased, causing the viscosity in the liquid-phase to increase; this behaviour was repeated in other studies carried out by Li *et al.*, 2009 and Dejaloud *et al.*, 2018.  $d_{32}$  for ALB with gas-liquid flow or with gas-liquid-diesel were higher in the riser zone and increased when the  $U_g$  increased. Likewise, this behaviour is consistent with what was observed in other studies about mass transfer and shear stress carried out by Bannari *et al.*, 2011 and Han *et al.*, 2017 on behaviour of a non-Newtonian fluid.  $d_{32}$  in gas-liquid flow ALB was higher than in gas-liquid-diesel ALB, and in other research, when  $d_{32}$

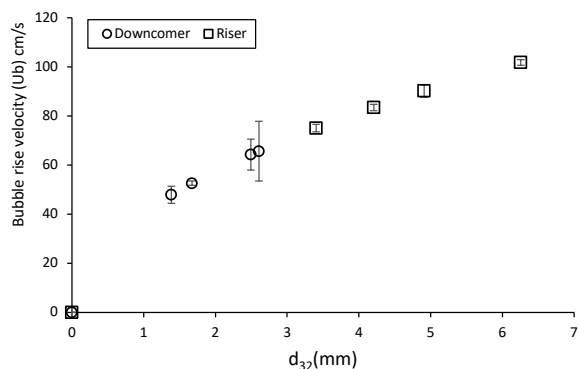


Fig. 6. Bubble rise velocity ( $U_b$ ) as a function of Sauter mean diameter ( $d_{32}$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid flow (two-phase).

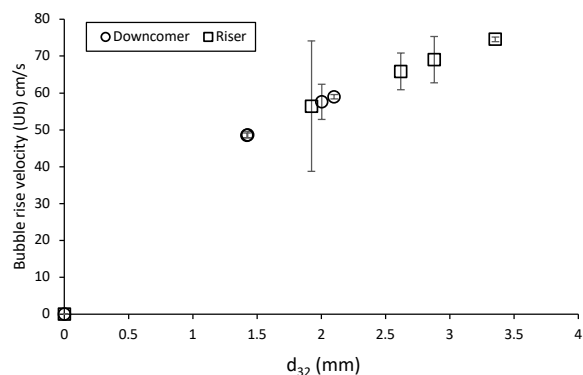


Fig. 7. Bubble rise velocity ( $U_b$ ) as a function of Sauter mean diameter ( $d_{32}$ ): ( $\square$ ) riser zone and ( $\circ$ ) downcomer zone, evaluated in a gas-liquid-diesel flow (three-phase).

was evaluated, as well as in ALB, the same behaviour was observed, (Dejaloud *et al.*, 2018).

In Fig. 6, variations of bubble rise velocity ( $U_b$ ) as a function of  $d_{32}$  in the gas-liquid flow ALB are shown.  $U_b$  and  $d_{32}$  are necessary for the comprehension of hydrodynamic fundamentals for scaling and efficient reactor control (Jamshidi & Mostoufi, 2017). The results showed that  $U_b$  increased from 47.91 to 65.69  $\text{cm s}^{-1}$  as bubble size increased.  $U_b$  data were consistent with the results observed in  $d_{32}$ , so it also increased with increasing  $U_g$ , which is behaviour that was also observed in the research of Prakash *et al.*, 2001 but with a three-phase bubble column reactor of water, air and yeast cells.

Fig. 7 shows the variation of  $U_b$  as a function of  $d_{32}$  in gas-liquid-diesel ALB. The values of  $U_b$  in gas-liquid-diesel ALB were lower than  $U_b$  in gas-liquid ALB. Besides that, the riser zone had higher  $U_b$  values

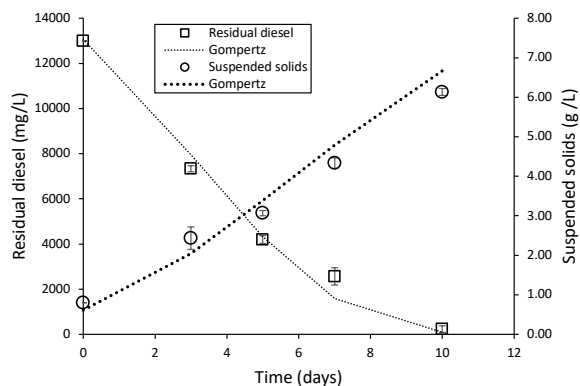


Fig. 8. Comparison between ( $\circ$ ) residual diesel and ( $\square$ ) suspended solids as a function of time (days) in a mineral medium. Lines in residual diesel and suspended solids (SS) represents Gompertz model with  $R^2 = 0.9897$  and  $R^2 = 0.9898$ , respectively.

because in the riser zone, where air-flow circulation in ALB begins, air bubbles have a larger diameter than air bubbles of the downcomer zone where only arrive small bubble size, causing a difference of flow between the riser zone and the downcomer zone, generating a lower volumetric mass transfer coefficient ( $k_La$ ) in the downcomer zone with respect to the riser zone (Zheng *et al.*, 2018).

### 3.2.2 Residual diesel and suspended solids

Diesel was used as a carbon source since it is principally composed of AHs (> 90%) (Chaudhary, Bajagain, Jeong, & Kim, 2019), so it can be used as a composite model for the hydrocarbons found. Residual diesel and suspended solids (SS) in the mineral medium are presented in Fig. 8. The SS began with an initial concentration of 0.80  $\text{g L}^{-1}$  until reaching a maximum concentration of  $6.13 \pm 0.09 \text{ g L}^{-1}$  after ten days of culture. Furthermore, the initial concentration of diesel was reduced from 13,000  $\text{mg L}^{-1}$  to  $230 \pm 150 \text{ mg L}^{-1}$ , whereby 98.23% of initial diesel concentration was uptake, and the consortium could uptake 96.58% of concentration hydrocarbon found in Tampaon River, which had the highest concentration of hydrocarbons found at the sampling points. In comparison, the bacterial *Gordonia alkanivorans* reduced diesel concentration from 500  $\text{mg L}^{-1}$  to 30  $\text{mg L}^{-1}$ , absorbing 94% of diesel, in a shaking flask bioreactor (SFB) in 11 days of incubation (Chen *et al.*, 2017), degrading less than in our results in ALB, even though his concentration was much lower.

Table 3. Maximum specific growth rate ( $\mu_m$ ), maximum specific uptake rate ( $Q_m$ ), inflection time (ti) and lag phase ( $\lambda$ ) for the suspended solids and residual diesel.

Data	$\mu_m/Q_m$ (d <sup>-1</sup> )	$\lambda$ (d)	ti (d)
Suspended solids	0.59	4.36	5.63
Residual diesel	1871.77	3.19	3.25

Also, in other research, was reduced diesel concentration from 10,000 mg L<sup>-1</sup> to 1,800 mg L<sup>-1</sup>, absorbing 82% of diesel after four days of the culture, using the *Pseudomonas luteola* bacteria stimulated by aminoglycosides in a SFB (Atanasković et al., 2016); the degradation was more than that obtained in our research in a similar timeframe of five days, where the initial concentration was reduced to 4,020 ± 170 mg L<sup>-1</sup>.

The next parameters were estimated using the Gompertz model: maximum specific growth rate ( $\mu_m$ ) 0.59 d<sup>-1</sup>, maximum specific uptake rate ( $Q_m$ ) 1,871.77 d<sup>-1</sup>. Furthermore, inflection time (ti) is the maximum velocity reached and lag phase ( $\lambda$ ), which is the start time (Table 3). The diesel uptake, started first at 3.19 days, shortly reached its maximum velocity (3.25 days); then growth started at 4.36 days and finally reached its maximum velocity at 5.63 days. The consortium yield at ten days of culture was 0.42 g<sub>SS</sub> g<sub>diesel</sub><sup>-1</sup>, which is similar to other research with hexadecane used as carbon source at 0.41 g<sub>SS</sub> g<sub>diesel</sub><sup>-1</sup> (Tec-Caamal et al., 2018). Furthermore, productivity after ten days of culture was 1.28 g<sub>diesel</sub> L<sup>-1</sup>d<sup>-1</sup>.

## Conclusions

In all water bodies sampled in the state of San Luis Potosí, polluting aliphatic hydrocarbons were found, such as eicosane, heneicosane, octacosane or heptacosane that had concentrations above the LMOS. The AHs found could have multiple sources, but pyrogenic sources emerged as the main source of hydrocarbon pollution, such as that generated by agricultural activity in the area, mainly for the burning of sugar cane.

Hydrodynamic parameters  $\varepsilon_G$ ,  $d_{32}$ ,  $U_b$  and  $Re$  increased as the superficial gas velocity ( $U_g$ ) increased. The best results were obtained with a  $U_g$  of 1.92 cm s<sup>-1</sup>, this  $U_g$  showed a turbulent flow in the riser zone and a transitional flow in the downcomer zone in an ALB with gas-liquid-diesel

flow. In addition, mixing and mass transfer improved as the  $U_g$  increased. Downcomer zone was relevant hydrodynamic limitation for mass transfer in the ALB.

ALB was able to degrade 98.21% of the diesel used as a carbon source after ten days of incubation. Based on productivity and degradation observed, the ALB used is an alternative for the remediation of hydrocarbon pollution in the sampled rivers and lagoons.

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