Abatement of methane diffuse emissions by biotrickling filtration using polyurethane foam as carrier.

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ABSTRACT

Methane anthropogenic diffuse emissions (below 1 % vv^{-1}) are important, with a significant contribution of the waste management sector (estimatedaround 21%). Since biological alternatives are especially promising for the treatment of a wide number of gaseous pollutants, a biotrickling filter packed with polyurethane foam was employed to study the abatement of methane (0.2 – 1 % vv^{-1}) in air emissions. A long-term operation (180 days) under stable conditions in terms of process performance, pH and pressure drop was maintained. Short term assays were also performed to assess the effect of waterings, empty bed residence times (EBRT) and inlet loadsapplied. In spite of its hydrophobicity, maximum removal rates of 14.7 g $CH_4 m^{-3} h^{-1}$were achieved, with removal efficiencies up to 30 %.Water addition is an important operational parameter to

optimizeto avoid water limitations as well as the increase of mass transfer resistances.Applied inlet loads and EBRTs indicated that the best removal efficiencies were obtained when higher methane inlet load/inlet concentration werefed to the system with the best results achieved when EBRT is longer than 6 minutes.

Keywords

Methane, diffuse emissions, biotrickling filter, methanotrophs, EBRT.

1. INTRODUCTION

During the last decades it has been evidenced that variations in the atmosphere composition can be related with climate change as well as with anthropogenic activities. The burdens of greenhouse gases (GHG) have increased from 2005 to 2011, being methane a relevant compoundwhich has been under study in last decade. In fact, after being nearly stable in the period 1999 – 2006, an increase was clearly detected after 2007 (Hartmann, Klein Tank, et al., 2013).Moreover, due to its high global warming potential (20 – 25 times superior to CO_2) methane contributes significantly to the climate change even although the changing rate is lower than the carbon dioxide.(Karakurt, Aydin, et al., 2012)

Among all kind of industrial activitiesreleasing methane into the atmosphere,the contribution of the waste management sectoris around 20.6 % of the total of the anthropogenic methane emissions, with landfills and wastewater treatment plantsreported as the main sources(Karakurt, Aydin, et al., 2012).Emissions from these industries emit large flows with low concentrations of methane (usually under 1.5 % vv^{-1})which makes more difficult their treatment through conventional treatments. As representative examples,Girard et al. (2011)measured a range of 0.025 to 0.4 % $vv^{-1}$of methane emissions from a piggery industry;Melse and Van der Werf

(2005)measuredmethane concentrations in the range of0.01 – 2.8 % $vv^{-1}$from liquid manure storage; andYoshida et al. (2014) reported a range between 2 – 32.7 % of methane released to the atmosphere during measurement campaigns in a municipal WWTP in Copenhagen.

Conventional treatments of methane diffuse emissions, mostly based in physic-chemical operations such as incineration, have been demonstrated to be difficult to apply due to the extra costs derived from the use of additional fuel. On the contrary, according to recent works, biological technologies appear to be especially suitable for a wide range of gaseous pollutants at low concentrations, although the treatment of hydrophobic substances (such as methane) represents an additional challenge since the process is limited by their low solubility(Park, Moon, et al., 2005)

Different studies have been carried out with biological system to abate methane in a range of diffuse emissions. Thus, Menard et al., (2011) employed an inorganic packed biofilter to treat 80 g_{CH4} $m^{-3}h^{-1}$ (0.6 % vv^{-1}). Results indicated removal efficiencies around 37 % and removal rates of 30 g_{CH4} $m^{-3}h^{-1}$. Melse and Van der Werf, (2005) worked on a biofilter packed with expanded perlite and compost achieving maximum removal efficiencies between of 25 -75 % working in a range of 0.6 – 6 % vv^{1}. Girard et al.,(2011) treated methane concentration in the range 0.02 – 0.4 % vv^{-1} in a biotrickling filter packed with gravel reaching a maximum removal efficiencies of 43%.

The aim of this study was to assess the performance and limitations of the biofiltration of low methane concentrations (< 1 % vv^{-1}) at long-term operation using a biotrickling filter (BTF) packed with polyurethane foams as carrier due to their excellent properties in terms of available surface, void space and stability. Different operational parameters were tested in terms of methane elimination capacity (EC) and removal efficiency (RE) such as the watering influence, the empty bed residence times (EBRT) and inlet loads (IL) provided.

2. MATERIALS AND METHODS

2.1. Experimental setup

A fully instrumented biotrickling filter for the methane treatment (BTF)is made of glass and has a total volume of 13.4 L, an inner diameter of 0.1 m and a length of 1.7 m with sampling points at different heights (Figure 1). The influent gas fed to the reactor was obtained mixing a pure methane stream (supplied by CarburosMetalicos, Spain) with an air stream, being the mixture controlled by means of mass flow controllers (Bronkhorst High-Tech BBV, The Netherlands). Nutrient solution was provided by a peristaltic pump BlackStone Model BL710.

Figure 1 here

2.2. Packing material and inoculation procedure

The packaging material employed was 4.5 L of open pore polyurethane foams (OPUF) cubes (75 – 90 % porosity, 250% absorption cube size 2.8 cm^3,)(Levapor GmbH, Germany). This was distributed into three independent sectionsof 30 cmof height each improving the homogeneous distribution of the nutrient solution as well as promotes the maintenance of a low pressure drop.

The inoculum used in the BTF was 4.5 L of sludge taken out from the aerobic chamber of a three stage Anaerobic Hybrid Membrane Biological Reactor (Buntner et al., 2011) with a final volatile suspended solids concentration (VSS) of 3 g L^{-1}. In order to promote the immobilization of the biomass inoculum onto the support, the packing material mixture was mixed with the inoculum and kept in contact during 24 hours prior to the filling of the bioreactor column.

A specific mineral medium for methanotrophic microorganisms was employed and was prepared in five main solutions as performed by (Nikiema, Brzezinski, et al., 2010). The composition in g L^{-1} was: Solution A: Na_2HPO_4, 86; KH_2PO_4, 53, Solution B: K_2SO_4, 17; $MgSO_4.7H_2O$, 3.7; $CaCl_2 \cdot H_2O$, 0.7, Solution C: $ZnSO_4.7H_2O$, 0.287; $MnSO_4.7H_2O$, 0.233; $CuSO_4.5H_2O$, 0.125; KI, 0.083; H_3BO_3, 0.062; $CoCl_2.6H_2O$, 0.048; $NaMoO_4.2H_2O$, 0.048, H_2SO_4 (1 mM), 1mL L^{-1}, Solution D_NH_4^+: NH_4Cl, 50 or D_NO_3^-: $NaNO_3$, 50, Solution E: $FeSO_4 \cdot 7H_2O$, 11.2; HCl (1 M), 1mL L^{-1} (adapted from(Cornish, Nicholls, et al., 1984).

2.3. Temperature, pH and pressure drop

Temperature, pH and conductivity in the liquid phase as well as temperature and relative humidity in the gas phase were measured by means of a Profilux 3 device (Profilux Iberia, Spain). Temperature was always maintained around 22-23ºC. The pressure drop along the packing bed was measured by means of a glass U-tube manometer. Leachate samples were periodically collected for analysis from the bottom of the bioreactor. A GLP-22 sensor was employed for measuring pH off-line (Crison Instruments S.A., Spain).

2.4. Analytical methods

Gas samples were periodically extracted from the inlet and outlet ports of the biofilter for CH_4 analysis. A calibrated GC 6850 Serie II (Agilent Tech. S.A., Spain) equipped with a GS-CarbonPlot and a flame ionization detector (FID) was used for CH_4 quantification. Injector, oven and detector temperatures were 290, 170 (isothermal) and 320ºC, respectively. Helium was employed as carrier gas, while a split ratio of 15:1 was applied. This method allowed measuring CH_4 concentrations in a range from 0 to 7000 ppm_v. CO_2 concentration was measured using a non-dispersed infrared sensor (GMP343, Vaisala, Finland).Ammonium, nitrite and nitrate were determined according to the Standard Methods (APHA-AWWA-WPCF-1998).

2.5. Microbiological tools

The abundance of different populations of microorganisms present in the biomass of the BTF was examined by fluorescent in-situ hybridization (FISH), according to Amann (1995). The specific probes used were ARC915 for Archaea, MG705 and MG84 for Type I methanotrophs, MA450 for Type II methanotrophs.Illumina HiSeq® technology was employed to sequence the libraries of the biomass employed by the inoculum. Data were analysed throughout bioinformatics which analyse transcriptome and its variability, identify non-standard RNA molecules and analyse small ncRNAs.

3. RESULTS AND DISCUSSION

3.1. Strategies for start-up and biomass colonisation (Period 1)

According to our previous experiences(Hernández, Gómez-Cuervo, et al.), the main operational parameters selected for treating these diffuse emissions (2000 ppmv) were 4.7 minutes of empty bed residence time (EBRT) and 0.5 g N L^{-1} of ammonia (NH_4^+) as nitrogen sourcein the nutrient solution. After 15 days of operation removal efficiencies (REs)achieved a range of 10 - 16.5 %, with corresponded to maximum elimination capacities (ECs) of 3.91g CH_4 $m^{-3}h^{-1}$ (Table 1). However, pH stability was difficult to maintain with some decreases observed (down to 5.8), which appeared to be especially problematic in this initial stage of biomass colonisation of the biofilm. Indeed, biomass development in BTF systemsis a crucial stage, especially when treating diffuse emissions (<1%) since an appropriate and previously acclimated biomass should be convenient to reach a stable operation according previous reports (Nikiema and Heitz, 2009; Avalos Ramirez, Jones, et al., 2012). These authors indicated that more than 3 weeks were necessary to reach a steady state employing as inoculum lixiviate from a biofilter which was treating methane for more than 6 months at inlet concentrations of 0.1 – 0.7 % vv^{-1} (IL around 55 – 61 g CH_4 $m^{-3}h^{-1}$).

After this start-up of 15 days (Period P1a), during the following 2 months the BTF was operated using nitrate as N-source to guarantee stable conditions. In order to promote a

faster growth and colonisation of the carrier, the inlet load was doubled from 23.5 to 50 g CH_4 m^{-3} $h^{-1}$by increasing the inlet concentration up to 0.99 % vv^{-1} (9900 ppmv) and operating at an EBRT of 8 minutes. Along this period (P1b)a stable performance of the system was obtained, with maximum ECs around 11-14g CH_4 m^{-3} $h^{-1}$and REs up to 23-27% (Table 1). It is interesting to mention that there is a slight correlation between the nitrogen concentration in the nutrient solution and the REs and ECs obtained, with the minimum values obtained at the lowest N concentration (0.2 g $N\text{-}NO_3^-$ L^{-1}) and the maximum values when 0.7 g $N\text{-}NO_3^-$ L^{-1} wasapplied (Period P1e). pHalways ranged in neutral values (6.9-7.5) without the addition of any external buffer, and the pressure drop remained at low values.

3.2. Short-term optimisation tests (Period 2)

During the following weeks, different short-term assays were performed in order to characterise the BTF in terms of the following operational parameters: i) influence of water addition (nutrient liquid flow); ii) EBRT either by changing the inlet flow or by changing the inlet methane concentration; and iii) inlet load applied.

Water addition (Period P2a)

The need of moisture by the microorganisms, as well as nutrients, must be balanced with the adverse effect that water causes on methane mass transfer, diminishing its bioavailability by microorganisms. In order to study the effect of this operational

parameter, four different flows of the nutrient solution were supplied: 0.05 $L_{nutrient}$ L^{-1}_{bed} d^{-1}(224 mLd^{-1}),0.02$L_{nutrient}$ L^{-1}_{bed} d^{-1}(85 mLd^{-1}) and 0.01 $L_{nutrient}$ L^{-1}_{bed} d^{-1}(43 mLd^{-1})and finally 0.03$L_{nutrient}$ L^{-1}_{bed} d^{-1}(129 mLd^{-1}), maintaining always the methane inlet load in 49 g CH_4 m^{-3} h^{-1}(inlet concentration around 1%) and the EBRT at 8 minutes. Although the better results were obtained with 0.02$L_{nutrient}$ L^{-1}_{bed} d^{-1} with REs up to 24.9% and ECs of 12 g CH_4 m^{-3} h^{-1}, the most important finding was the strong limitation of the system at the lowest liquid flow supplied(0.01$L_{nutrient}$ L^{-1}_{bed} d^{-1}), with the lowest RE obtained (19.5 % RE).The values used in this work compatible with a stable operation (0.02-0.05 $L_{nutrient}$ L^{-1}_{bed} d^{-1}) arein the lower range compared withothers reported previously, among 0.03- 1 $L_{liquid\ phase}$ L_{bed}^{-1} d^{-1}(Avalos Ramirez et al., 2012a; Nikiema and Heitz, 2010; Veillette et al., 2011), operating at higher inlet loadings(between 68 – 100 g CH_4 m^{-3} h^{-1}), with lower methane inlet concentrations(0.3 - 0.7 % vv^{-1}) andsimilar EBRTs (4-6 minutes). The trickling liquid velocity applied in these watering assays varied from 0.02 mh^{-1} (0.01 $L_{nutrient}$ L^{-1}_{bed} d^{-1}) to 0.12 mh^{-1} (0.05 $L_{nutrient}$ L^{-1}_{bed} d^{-1}), whereas the values used by the other authors ranged 0.35 -0.57 m h^{-1}. The application of lower trickling liquid velocities might be especially convenient if the water distribution along the packaging is homogeneous. The use of co-current air-water pattern flow in which both water and gas are fed from the top of the bioreactor, as employed in this BTF, is especially suitable for this purpose, although counter-current pattern flows are more commonly used due to its better simplicity for control gas flow direction.

Influence of the EBRT (Period 2b)

Taking into account previous references (Gómez-Cuervo et al, n.d), the influence of the EBRT was checked in this system in the operational range from 2 to 8 minutes, more specifically 2, 4.7, 6 and 8 minutes. Two different approaches were followed.Firstly, maintaining the methane inlet concentration at the low value considered as an objective in this paper, i.e., 0.2 % vv^{-1}(2000 ppm_v) while diminishing the inlet flow which led to a variation of the IL from 45 to 12 g CH_4 m^{-3} h^{-1} (Figure 2a). Secondly, maintaining constant the inlet load at 50 g CH_4 m^{-3} h^{-1} while diminishing the inlet flow which led to an increase inthe inlet concentration from 0.2 and 0.98 % vv^{-1}(Figure 2b).

As Figure 2a depicts, the increase of the EBRT caused an increase in the removal efficiency (from 12 to 27%), since both the curves corresponding to the IL and the EC become closer. However, in this experiment the operation at longer EBRT at a constant inlet concentration (0.2% vv^{-1}) meant a lower IL applied, which implied a lower EC obtained (3.4g CH_4 m^{-3} h^{-1}).Figure 2b shows the dynamics of the system when the IL was maintained constant (around 50g CH_4 m^{-3} h^{-1}). Although it is again observed the increase of RE at longer EBRTs (from 13.5 to 22%), in this case the system was able to remove a significantly higher load (EC up to 10.7g CH_4 m^{-3} h^{-1}), which indicates that the increase of concentration is a key factor to enhance the removal capacity of the system.

It is clear from both experiments the key effect of EBRT on the performance of the system, with lower REs observed below 5 minutes(between 13.5 and 16.2%) with a slightly higher effect at lower ILs (12 vs. 45 g CH_4 m^{-3} h^{-1}). When the EBRT was lengthened up to 8 minutes the removal efficiencies achieved the maximum values of 27.8%. On the other hand, concerning removal rates (ECs) the trends are different in both assays. When the inlet concentration was maintained constant (Figure 2b) a longer EBRT implied a lower IL applied, which implies a lower EC obtained in spite of the higher RE. Thus, at 2 minutes the EC was 6.1 g CH_4 m^{-3} h^{-1} but later on it trended to reduce it up to 3.4 g CH_4 m^{-3} h^{-1}. On the opposite, working at stable IL both removal rates and removal capacities tend to increase with EBRT. Although the concentration is also highly increased in this experiment, it seems not to have a relevant role.

Effect of the inlet load (Period 2c)

Since EBRT is crucial in terms of optimising removal efficiencies, the objective of this assay was to test the effect of the increase of the ILwhen the EBRT was maintained in an optimal range. Thus, the IL was raised from 7.6 to 73.4 g CH_4 m^{-3} $h^{-1}$by increasing the inlet concentration (IC) of methane (0.15 – 1.47 % vv^{-1}) whereas maintaining steady the EBRT at 8 minutes (Figure 2c). The most clear evidence was the continuous increase of the EC following the trend of the IC, with a limitation beyond IL of 58 g CH_4 m^{-3} h^{-1} (at 1% $vv^{-1}$of inlet concentration), when the maximum EC was obtained

(10.5 g CH_4 m^{-3} h^{-1}).

Results demonstrated a direct relation between the methane provided (in terms of both IC and IL) and the EC of the BTF. Removal rates above 10.5g CH_4 m^{-3} h^{-1} were obtained when IC was over 1 % vv^{-1} and IL higher to 58 g CH_4 m^{-3} h^{-1}. In contrast, the minimum EC took place when the IC was below 0.2 % vv^{-1} and IL lower than 15 g CH_4 m^{-3} h^{-1}. Nevertheless, maximum RE (23.6%) was observed when the lower amount of methane was provided (0.15% vv^{-1}). Higher IC led a fluctuation of the performance between 14.1 and 19.0 %.

Figure 2 here

3.3. Final steady-state operation (P3)

Period 3 correspond to two periods in which steady-state was maintained at two different methane inlet concentrations, i.e., 0.5% during almost 2 months (Period P3a), and finally 0.2% again during the last 10 days of operation (Period P3b), maintaining in both stages the minimum EBRT compatible with significant performances according to the previous experiments, i.e., 4 minutes.

The results obtained during P3a, operating at an inlet loading of 50g CH_4 m^{-3} h^{-1} led to the achievement of maximum REs of 25.6%, corresponding to ECs of 14.7g $CH_4 m^{-3}$ h^{-1}. On the other hand, according to the results obtained during the short-term

tests, the reduction of the IL and inlet concentration on P3b led to a reduction in the EC and a significant increase in the RE of the system (up to 29%).

3.4. Microbial ecology identification

Illumina technology was performedto characterize the inoculum used in the BTF. Results indicated a high diversity of microorganisms since around 1200 genera and more than 800 specieswere identified. Most of them (95 %) werebacteria being Protobacteria the phylum more abundant with more than 34 %. Concerning methanotrophic microorganisms, the main families of methanotrophs belong to *Protobacteria*phylum being *Gammaproteobacteria* and *Alphaprotobacteria* the dominant classes.Another phylum includingmethanotrophs is Verrucomicrobia, although there is bare information about their role in biofiltration. Apart from these other kind of microorganisms Wendlandt et al. (2010) described that another way of microbial methane oxidation could be carried out by a consortium of methanotrophic archaea, although archaea were not addressed by this Illumina analysis.

Gammaprotobacteria and *Alphaprotobacteria* classes include species which consume methane under aerobic conditions. *Gammaproteobacteria* (Methanotrophs Type I) are present in environments with limiting methane concentrations and high amount of nitrogen available whereas *Alphaproteobacteria* classes (which include methanotrophs Type II) are predominant in high methane concentration environments with low levels

of dissolved oxygen, as well as limitations in nitrogen and cupper(Amaral and Knowles, 1995; Hanson and Hanson, 1996). Therefore, it would be expected that *Gammaprotobacteria* would be the predominant class to be developed in the BTF used in this work.Results obtained from the application of Illumina technology to the inoculum sample showed that 2.1 % of the microorganisms belong to the genera identified as methanotrophs (Semrau, DiSpirito, et al., 2010): *Methylocaldum.*, *Methylomicrobium* and *Methylomonas*, which belong to the family *Methylococcaceae*, corresponding to the class *Gammaproteobacteria*.On the other hand, it was identified a slight presence (0.6%) of bacteria belonging to the family *Methylocystaceae* and *Beijerinckiaceae*, both belonging to the class *Alphaproteobacteria*and also identified as methanotrophs (Semrau, DiSpirito, et al., 2010). Thus, this inoculum could be used as a source of methanotrophs although its limited number may limit the development of the process in the BTF.

Once characterised the inoculum with the Illumina technology, samples from the BTF were taken to be characterised byFluorescence in situ hybridization (FISH) in order to identify the presence of selected methanotrophs. The samples were taken after the first two months of operation, during the stable period P1e, in which an inlet loading of 48 $g_{CH4}m^{-3}h^{-1}$ was applied and an average elimination capacity of 10.9 $g_{CH4}m^{-3}h^{-1}$ was obtained.Figure 3 shows the identification of Type I methanotrophs and archaea, as well as the limited presence of Type II methanotrophs.

Therefore, the presence in the inoculum of methanotrophs able to grow in the environment imposed in the BTF (very low methane concentration) such as those belonging to the family *Methylococcaceae*, resulted in their further development as observed during steady state operation (Period 1e). This kind of microorganismsare able to carry out the methane oxidation and their growth must be stimulated to achieve higher removal rates in the BTF.

Figure 3 here

3.5. Pressure drop

Pressure drop control along the operation of the BTF is essential in terms of operational energy costs. Along this work, the pressure drop was always maintained below 0.36 cm of H_2O m_{bed}^{-1}, as the result of the characteristics of the carrier selected, the limited growth of methanotrophs and also the configuration of the packaging inside the BTF, distributed in three independent sections of 30 cm height each. In fact, inlet load variations, changes in the water addition and modifications in EBRT applied did not cause a significant increase of this parameter, which is interesting in terms of its suitability at long-term. These values of pressure drop are significantly lower comparing with other works previously reported for BTF systems. Avalos Ramirez et al. (2012a) compared the effect of the porous and non-porous media and the pressure drop obtained during in a biofilter packed with inert material. Pressure drop ranged between 0.05 – 0.8

cm of H_2O m_{bed}^{-1} after two weeks of operation. Delhoménie et al.(2002) observed a pressure drop of 3.5 cm of H_2O m_{bed}^{-1} in an organic packed biofilter with particles of 5 mm.

3.6. Carbon balance

A carbon balance was carried out during the whole experiment in order to check the development of the biomass. A monitoring of the carbon dioxide produced throughout the whole assay (data not show) has demonstrated the correlation between the methane removal efficiency and the CO_2 production. The methane oxidation also could be employed by a biomass growth, since bacteria obtain their carbon source from these process. Thus, periods as P2 (Table 1) in which the methane concentration provided was around 0.9 % vv^{-1} and EBRT at 8 minutes,the carbon balance indicated a tendency of the bioreactor to consume carbon to the bacteria growth. Furthermore, the nitrogen concentration is a crucial factor to the development of the biomass which increase from 0.1 to 0.5 mmol of C for biomass growth when the nitrogen concentration is increased from 0.5 to 0.7 g $N\text{-}NO_3$ L^{-1}. Conversely, periods as P1.2 and P1.3, in which inlet concentration and EBRT were lower (IC around 0.2-0.5 % vv^{-1} and EBRT at 4 minutes) the carbon balance indicated a higher carbon in the outlet than the provided. This may probably due to the collapse of the system and the lysis of the bacteria which release carbon. In the P3, in which optimization tests were performed, the carbon balance indicated a biomass growth only when the inlet concentration provided was higher than

0.8 % vv^{-1}.

Table 1 here

4. CONCLUSIONS

A biotrickling filter packed with polyurethane foam was used to treat diffuse methane emissions (0.2-1% vv^{-1}) at long term operation (approximately 6 months) under stable conditions (pH was neutral and pressure drop was always low) using an inoculum taken from a wastewater treatment plant in which methanotrophs were detected. The process was full monitored in terms of flows, concentrations and carbon mass balance. The main conclusion indicates the limitations of the treatment of this highly hydrophobic substance, since removal efficiencies were always limited below 30% and maximum removal rates achieved values up to 14.7g CH_4 m^{-3} h^{-1}. Although the influence of water addition may represent an additional mass transfer resistance to the methane diffusion process, this work evidences that this addition has to be optimised in order to avoid problems caused by water limitation, which is especially significant in the case of inorganic packagings with limited water retention as the PU foam used. The short term studies carried out indicate that the EBRT is the key factor to maximize methane abatement, with better results in the range of 6-8 minutes, being also the increase in concentration and inlet loadings beneficial to achieve higher removal rates but leading to lower removal efficiencies, so a balance between both parameters should be defined.

ACKNOWLEDGEMENTS

This work was supported by the Spanish Government (projects NOVEDAR_Consolider, CSD2007-00055 and INNOTRAZA, CTQ2010-20240) and the UE project LIVEWASTE (LIFE12 ENV/CY/000544).The authors belong to the Galician Competitive Research Group GRC2010/37.

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Table1: Summary of all the conditions establish in the BTF throughout the whole experiment.

	P1					**P2**			**P3**	
	P1a	**P1b**	**P1c**	**P1d**	**P1e**	**P2a**	**P2b**	**P2c**	**P3a**	**P3b**
						Watering assay	**EBRT assay**	**IL assay**		
Operational days	*15*	*8*	*8*	*12*	*28*	*28*	*20*	*16*	*59*	*10*
$\Delta P(mm_{H2O}\ m_{bed}^{-1})$	5.4	5.4	3.6	3.6	3.6	3.6	3.6	3.6	3.6	3.6
EBRT (min)	4	8	8	8	8	8	2.0 - 8.0	8	4	4
T (ºC)	22	21.3	21.3	21.3	21.3	23.23	23.23	23.23	23.2	23.2
Inlet Concentration (%)	0.2	1	1	1	1	1.5	0.2 - 1	0.15 - 1.46	0.5	0.2
Inlet Load (g m-3 h-1)	23.5	50.0	49.4	49.4	48.2	49.3	23.5 - 50	7.6 - 73.7	50.1	20.0
RE range (%)	0 - 16.5	10 - 27	10 - 25	18 - 25	20 - 23	19 - 25	13 - 28	14 - 24	12- 25.6	7.1 - 29
RE average (%)	9±4.0	19,3 ± 3,1	18.0± 8	21± 4.0	23± 3.0	20.0± 3.0	18.7± 5.5	18± 3.0	16.2± 3.8	22.2± 8
EC range (g m-3 h-1)	0 - 3.9	5.0 - 14.0	3.0 – 12.	9.0-11	10.0-11.0	8.9 - 12.6	3.4 - 10.7	1.8–10.5	7.4 - 14.7	1.0 - 7.0
EC average (g m-3 h-1)	2.0± 1.3	9.2 ± 3.1	8.4± 3.0	9.9± 1.6	10.9± 3.9	9.5± 1.7		8.05± 3.7	8.9± 2.1	4.7± 2.4

LIST OF FIGURES

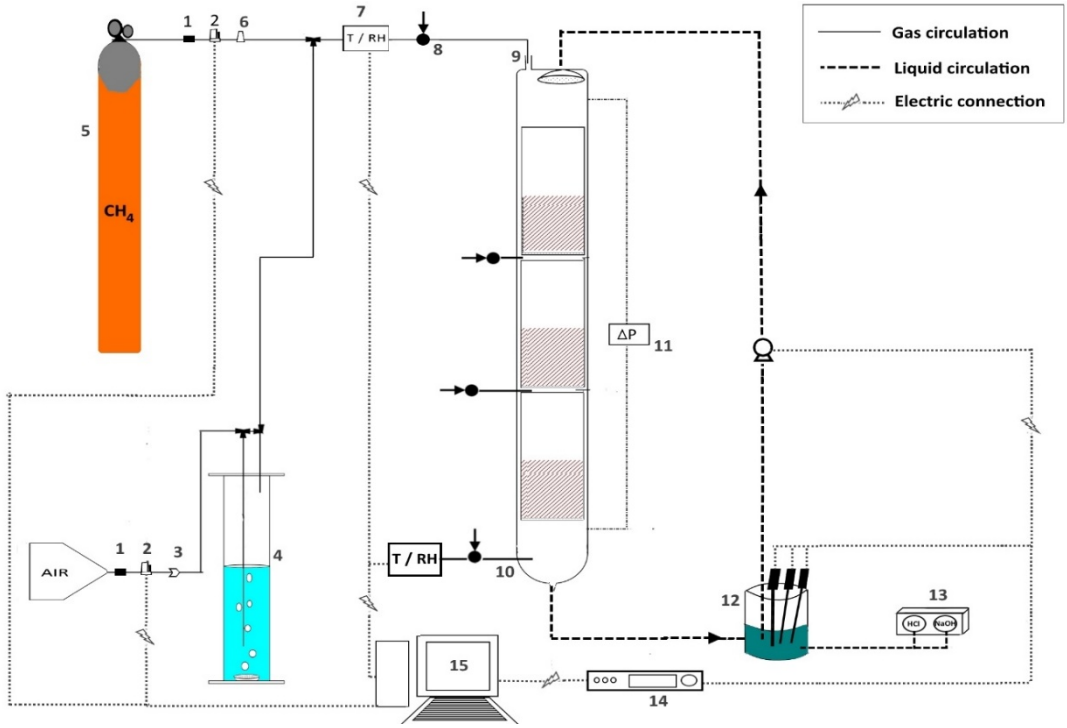
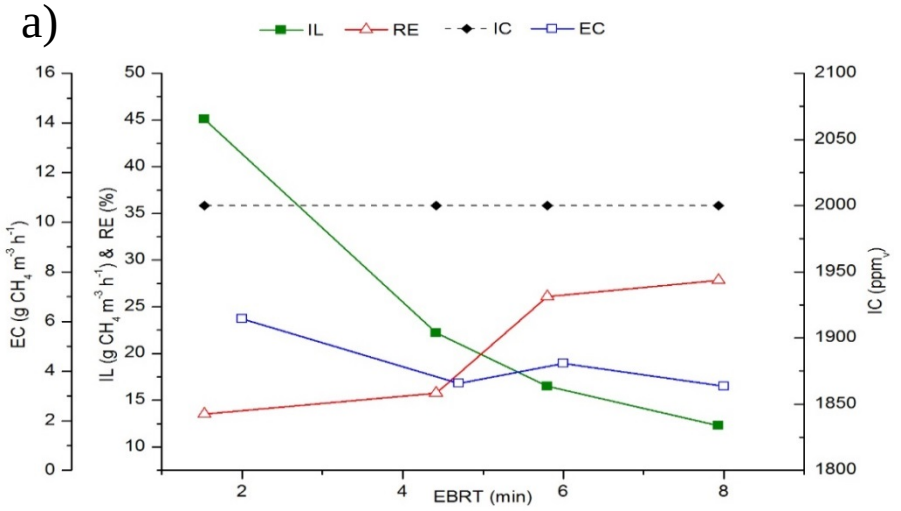
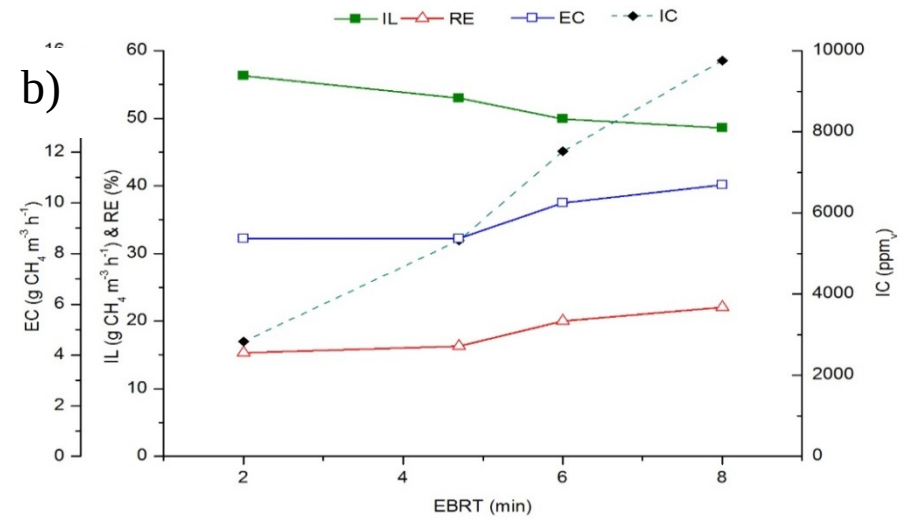


Figure 1: Schematic representation of the biofiltration setup (1): Particle filter; (2): air mass flow meter; (3): backflow valve (4): humidification tower; (5):methane gas cylinder; (6): coalescent filter; (7):relative humidity and temperature gas phase sensors; (8): sample points; (9): biofilter influent; (10): biofilter effluent; (11): pressure drop U-tube device; (12): mix and control tank with temperature, conductivity and pH sensors; (13): pH control system; (14): Profilux device for parameters monitoring; (15): data acquisition system / PC.

a)


IL
RE
IC
EC
EC (g CH4 m-3 h-1)
IL (g CH4 m-3 h-1) & RE (%)
IC (ppmv)
EBRT (min)

b)


IL
RE
EC
IC
EC (g CH4 m-3 h-1)
IL (g CH4 m-3 h-1) & RE (%)
IC (ppmv)
EBRT (min)

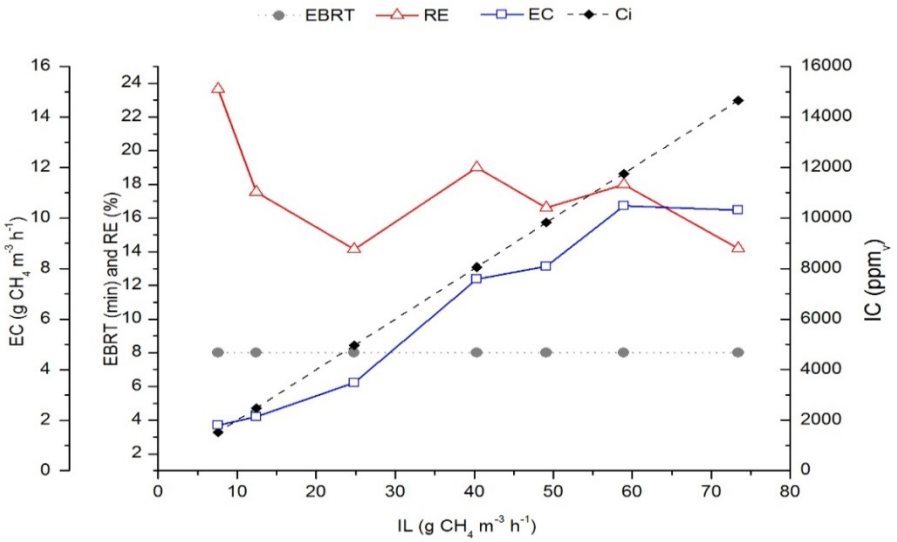
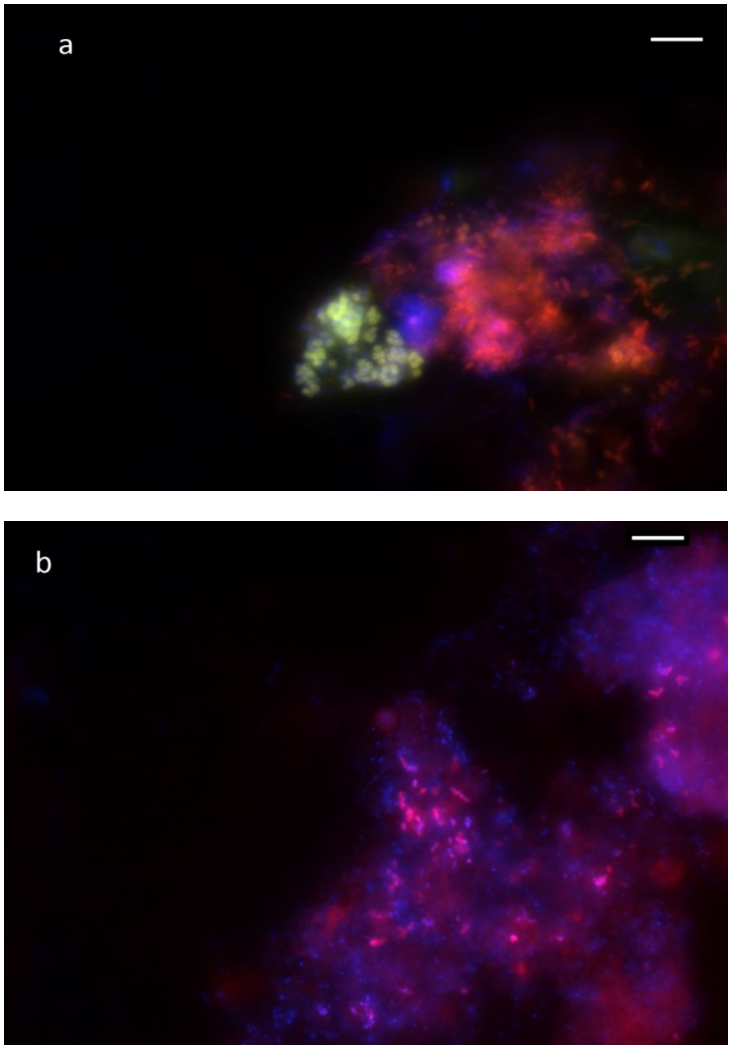


Figure 2: Optimization tests results: a)EBRT assay keeping constant inlet concentration varying IL; b)EBRT assay holding steady inlet load and changing inlet concentration; c) Inlet load variations assay.

c)


a
b

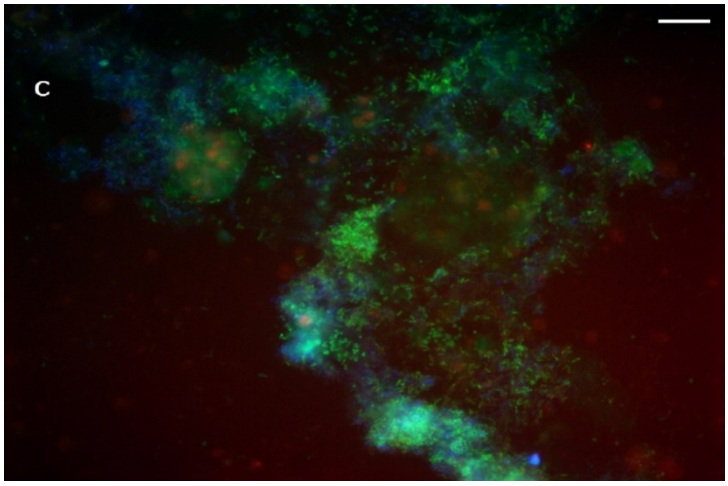


Figure 3: Fluorescence in-situ hybridation (FSH) of BTF biomass during the experiment: a) Aerobic methanotrophsType I with specific probes MG705 +MG84 (green) over general probe EUB330 (red) and Dapi (blue); 100x. b) Archaea with probes ARC915 (red) over Dapi (blue); 100x. c) Aerobic methanotrophs Type II barely detected with specific probe MA450 (red) over general probe EUB330 (green) and Dapi (blue); 100x.