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ORGANIC MATTER CAPTURE BY HIGH-RATE INOCULUM-CHEMOSTAT  
AND MBBR SYSTEMS

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AND MBBR SYSTEMS

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## DEDICATION

*À Najmeh*

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## RÉSUMÉ

Le traitement des eaux usées biologique à charge élevée permet d'utiliser la matière organique pour la production d'énergie par un procédé méthanogène qui contribue au bilan d'énergie positif pour les stations de récupération des ressources de l'eau (StaRRE).

L'objectif principal de cette recherche était de maximiser la biotransformation de la matière organique soluble et colloïdale de l'affluent en matière particulaire par l'emploi d'un bioréacteur à lit mobile (MBBR) pour que cette matière particulaire soit captée et acheminée vers un procédé de digestion anaérobie.

L'hypothèse scientifique originale de ce projet est qu'un système innovateur de MBBR servant d'inoculum (MBBR-inoculum) suivi d'un chimostat maximise la biotransformation de la matière organique biodégradable soluble et colloïdale ( $CS_B : C_B + S_B$ ) en matière particulaire ( $X_B$ ) tout en minimisant l'hydrolyse des matières particulaires biodégradables provenant de l'affluent ( $X_B$ ) afin que tout ces deux sources de  $X_B$  soient captées pour maximiser la production de méthane.

Deux configurations ont été étudiées : 1) un système de MBBR-inoculum et chimostat et 2) un MBBR à charge élevée. Le MBBR-inoculum est en fait un MBBR à charge élevée (HR-MBBR).

Les essais ont été réalisés à échelle pilote dans une remorque du centre de recherche, développement et validation des technologies et procédés de traitement des eaux (CREDEAU) alimenté en eaux usées de la StaRRE de Repentigny pour une période continue de trois mois.

Les eaux usées étaient moyennement concentrées avait une concentration de la demande en oxygène chimique de 268 à 482 mg DCO/L, de la DCO soluble de 38 à 73 mg  $S_{DCO}$ /L et des matières en suspension de 344 à 477 mg MES/L. Les ratios  $f_{VT}$  and  $f_{CV}$  des eaux usées étaient de  $0,65 \pm 0,10 \frac{g \text{ MVES}}{g \text{ MES}}$  et de  $1,7 \pm 0,2 \frac{g \text{ X}_{DCO}}{g \text{ MVES}}$ , respectivement. La fraction  $CS_B$  représentait 20 à 30 % de la DCO totale (60 à 120 mg  $CS_B$ /L) tandis que la matière colloïdale et soluble non biodégradable,  $CS_U$ , représentait 5 % de la DCO totale (20 à 25 mg/L).

Dans le MBBR, le taux de charge organique a varié entre 1,5 et 20 g  $CS_B \text{ m}^{-2} \text{ d}^{-1}$  correspondant à un temps de rétention hydraulique de 25 à 54 min. Dans ce taux de charge, la surface représente la surface active protégée des médias où peut s'attacher le biofilm. L'effet du ratio de remplissage du

médias a varié entre 50 % et 35 % v/v correspondant à 1000 L et 700 L de médias « K5 » pour une surface utile de 800 et 560 m<sup>2</sup>, respectivement.

Dans l'inoculum du procédé inoculum-chémostat, le taux de charge organique a varié de 20 à 85 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup> (basé sur la superficie utile des supports mobiles), tandis que le temps de rétention (TRH<sub>Inoc</sub> + TRH<sub>chem</sub>) a varié entre 150 et 300 min. La température de l'eau a varié entre 17 et 21°C et le pH se gardait stable autour de 7,0 dans les deux études.

Dans le réacteur à lit mobile, la biotransformation maximale du CS<sub>B</sub> en X<sub>B</sub> d'environ 90 % a été atteinte à un taux de charge organique entre 1,5 et 5,5 g DCO m<sup>-2</sup> d<sup>-1</sup>, ce qui correspond à un temps de rétention hydraulique de 36 à 55 min.

Dans le procédé inoculum-chémostat, la biotransformation maximale du CS<sub>B</sub> en X<sub>B</sub> d'environ 80 % a été atteinte à un taux de charge organique entre 22 et 40 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>, ce qui correspond à un temps de rétention hydraulique de 3,7 h à 3,9 h.

Dans le réacteur à lit mobile, la concentration en oxygène dissous a montré un effet important sur l'efficacité opérationnelle et la biotransformation. De petites augmentations de la concentration de l'oxygène dissous de 1 à 2 mg O<sub>2</sub>/L à 2 à 3 mg O<sub>2</sub>/L a augmenté significativement l'efficacité de la biotransformation du CS<sub>B</sub> de 64 ± 13 % à 84 ± 6 %. L'augmentation de la concentration de l'oxygène dissous a contribué à l'oxydation de la matière organique particulaire par biofilm.

Le taux d'utilisation de l'oxygène dans le bioréacteur à lit mobile (53 mg O<sub>2</sub> L<sup>-1</sup> h<sup>-1</sup>) était trois fois plus élevé que dans le réacteur chémostat (16 mg O<sub>2</sub> L<sup>-1</sup> h<sup>-1</sup>).

Une conclusion qui a pu être établie du bioréacteur à lit mobile est que la biotransformation de CS<sub>B</sub> était sensible à la concentration de l'oxygène dissous tandis que c'était le temps de rétention hydraulique et le taux de charge organique qui était sensible dans le système inoculum-chémostat.

Ce projet a déterminé le potentiel du procédé innovateur d'inoculum-chémostat pour le traitement des eaux usées comme alternatif pour les StaRRE à énergie positive ou efficace.

Mots clés : chémostat, oxydation de la DCO, bioréacteur à lit mobile à taux élevé, captage de la matière organique

## ABSTRACT

High-rate biological treatment processes allow the recovery of organic matter from wastewater into energy via methanogenesis contributing to the energy positive development of water resource recovery facilities (WRRFs) with lower carbon footprints.

The main objective of this research was to maximize the bio-transformation of influent soluble and colloidal organic matter into particulate COD using a high-rate moving bed bioreactor (MBBR) for subsequent physico-chemical capture prior to transport to anaerobic digestion process.

The original scientific hypothesis of this project is that a high-rate innovative MBBR-inoculum followed by a chemostat system maximizes the bio-transformation of soluble and colloidal biodegradable organic matter ( $CS_B: C_B+S_B$ ) into particulate matter ( $X_B$ ) while minimizing the hydrolysis of particulate biodegradable organic matter from the influent ( $X_B$ ), so that both of these  $X_B$  sources are captured to maximize methane production.

Two configurations were studied: 1) an MBBR-inoculum and chemostat system and 2) a high-load MBBR. MBBR-inoculum is in fact a high-load MBBR (HR-MBBR).

The tests were carried out with the activated sludge pilot trailer of the center of research, development and validation of water treatment technologies and processes (CREDEAU) using real wastewater from the WRRF of Repentigny for a three month continuous operation.

The wastewater was moderately concentrated, based on the concentrations of chemical oxygen demand (COD), soluble COD and total suspended solids (TSS), ranging from 268 to 482 mg COD/L, 38 to 73 mg  $S_{COD}$ /L, and 344 to 477 mg TSS/L. The raw wastewater  $f_{VT}$  and  $f_{CV}$  indexes were  $0.65 \pm 0.10 \frac{g VSS}{g TSS}$  and  $1.7 \pm 0.2 \frac{g X_{COD}}{g VSS}$ , respectively. The  $CS_B$  fraction represented 20 to 30% total COD (60 to 120 mg  $CS_B$ /L) while the unbiodegradable colloidal and the soluble fraction ( $CS_U$ ) represented 5% of the total COD (20 to 25 mg/L).

In the MBBR system, the organic loading rates (OLRs) varied between 1.5 to 20 g  $CS_B m^{-2} d^{-1}$ , which corresponded to hydraulic retention times (HRTs) of 25 to 54 min. The effect of the media fill volume fraction was changed from 50% to 35% v/v, which corresponded to 1000 L and 700 L of K5 media and provided 800 and 560  $m^2$  of useful surface area, respectively.



In the inoculum of the inoculum-chemostat process, the OLR varied from 20 to 85 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup> (based on the useful surface area of the media), while the HRT (HRT<sub>Inoc</sub> + HRT<sub>chem</sub>) ranged from 150 to 300 min. The water temperature ranged from 17 to 21 °C and the pH was around 7.0 across both processes during the study.

In the MBBR system, the maximum biotransformation of CS<sub>B</sub> into X<sub>B</sub> of near 90% ± 3 was obtained at the OLR of 1.5 to 5.5 g COD m<sup>-2</sup> d<sup>-1</sup> that corresponded to HRT between 36 min to 55 min.

In the inoculum-chemostat process, the maximum biotransformation of CS<sub>B</sub> into X<sub>B</sub> of near 80 ± 3 %, was obtained at an OLR between 22 to 40 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup> at HRT between 3.7 h and 3.9 h.

In the MBBR system, dissolved oxygen concentration demonstrated a major effect on the operational efficiency and bio-transformation. Small increases in DO level ranged from 1-2 mg O<sub>2</sub>/L to 2-3 mg O<sub>2</sub>/ L led to a significant increase in CS<sub>B</sub> biotransformation efficiency from 64% ± 13 to 84 ± 6%. An increase in DO level contributed particulate organic matter oxidation by the attached biofilm.

The oxygen uptake rate (OUR) in the MBBR (53 mg O<sub>2</sub> L<sup>-1</sup> h<sup>-1</sup>) was about three times higher than in the chemostat reactor (16 mg O<sub>2</sub> L<sup>-1</sup> h<sup>-1</sup>).

It was concluded that in the MBBR system, the CS<sub>B</sub> biotransformation was sensitive to the DO concentration while in the inoculum-chemostat, it was more sensitive to the HRT and OLR.

This project determined the potential of the innovative inoculum-chemostat process for wastewater treatment as an alternative system towards energy positive/efficient WRRFs.

Key words: chemostat, COD oxidation, high rate MBBR, organic matter capture.

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## LIST OF SYMBOLS AND ABBREVIATIONS

AS	Activated sludge
BOD	Biochemical oxygen demand
C <sub>B</sub>	Biodegradable colloidal COD
Chemo	Chemostat process
COD	Chemical oxygen demand
CS <sub>B</sub>	Filtrable biodegradable organic matter
CS <sub>U</sub>	Filtrable unbiodegradable organic matter
C <sub>U</sub>	Unbiodegradable colloidal organic matter
COD	Chemical oxygen demand
DO	Dissolved oxygen
Eff	Effluent
f <sub>CV</sub>	Particulate matter COD over VSS ratio
HRT	Hydraulic retention time
IC	Inoculum-chemostat
Inf	Influent
Inoc	Inoculum process
f <sub>VT</sub>	Volatile suspended solids over total suspended solids ratio (VSS/TSS)
MBBR	Moving bed biofilm reactor
OLR	Organic loading rate
OUR	Oxygen uptake rate
P <sub>t</sub>	Total phosphorus
Q	Flowrate
RAS	Return activated sludge



$S_B$	Biodegradable soluble COD
$S$	Soluble matter ( $< 0.08 \mu\text{m}$ )
SRT	Sludge retention time
$S_U$	Unbiodegradable soluble organic matter
TS	Total solids
TSS	Total suspended solids
VDS	Volatile dissolved solids
VSS	Volatile suspended solids
WRRF	Water resource recovery facility
TKN	Total Kjeldahl nitrogen
WAS	Waste activated sludge
$X_{\text{COD}}$	Particulate matter
$X_B$	Biodegradable particulate matter
$X_{\text{OHO}}$	Ordinary heterotrophic organisms

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## CHAPTER 1 INTRODUCTION

### 1.1 Context

The world has been focusing on mitigating the problems resulting from population growth and economic development and among the issues to be addressed are the changes resulting from environmental impacts on wastewater quantity and quality. Wastewater needs to be processed to treat biological, chemical and physical contaminants due to the high demand for a clean and safe environment.

High land costs are the likely consequences of high population density and this has led to unconventional solutions for wastewater collection and handling, including pure oxygen systems, two-story settling tanks, deep aeration tanks and even underground plants. Whereas conventional wastewater treatment processes with large occupied surface areas are too technically sophisticated and costly and often require high energy inputs for operation, a conventional activated sludge (AS) process, comprised of an aeration tank with a secondary clarifier and a sludge recirculation line, as the aeration process takes longer than 6 hours across the treatment process (Saleh, 1994) resulting in 0.3-0.7 kWh per m<sup>3</sup> consumption of energy (Metcalf and Eddy-Aecom, 2014). However, wastewater may be considered as a source of energy and if properly extracted, it can provide significant portion of the energy requirement for treatment (Gude, 2015). The main source of this energy results from the large quantities of biomass from wastewater treatment processes (Rulkens, 2007).

Over the past few decades, increasing attention has been devoted to considering several options for making the treatment process energy-yielding rather than an energy-consuming, despite the fact that wastewater treatment is an energy intensive process. In this regard, researchers have conducted various studies to meet diversified social needs, such as the reduction of organic matter footprint and treatment costs for secondary or biological treatment. As a result of intensive studies, advanced wastewater treatment process technologies have been developed recently, i.e. advanced oxidation processes (AOPs), bio-filter processes, aerobic granular sludge (AGS), anammox, moving bed bioreactor (MBBR), etc.

The MBBR is a biological continuous flow process which combines the benefits of activated sludge and bio-filter processes without a need for sludge recirculation (Ødegaard et al., 1994). The

submerged carriers, on which the biofilm grows, are kept in a suspension by either a mixer or an aeration system.

Currently, due to a better understanding of the biological process and bacterial populations as well as having determined a way to apply them in the most energy efficient manner possible, the industry is moving away from the term "wastewater treatment plants" (WWTPs) to "water resource recovery facilities" (WRRFs) (U.S.EPA, 2016).

In the quest to enhance energy self-sufficiency (or autarcy), reduce the carbon footprint and achieve a sustainable operation, WRRFs typically employ an energy recovery process typically by operating an anaerobic digestion (AD) process for the production of biogas and energy (Jimenez et. al., 2015). However, in general, it is not possible in WRRFs to obtain energy neutral operations without concurrently minimizing energy usage (ex. from aeration) and maximizing energy (organic carbon) recovery. The AD system process produces biogas comprised of 50-70% methane ( $\text{CH}_4$ ) from biodegradable organic matter ( $\text{COD}_b$ ) (Mata-Alvarez et al., 2000).

The energy efficiency of WRRFs has been improved by reducing energy consumption by optimizing aeration systems and optimizing the capture of the readily biodegradable organic matter to produce more energy through anaerobic digestion systems (energy positive/efficient).

Biodegradable COD can be identified in two fractions that are either readily (RBCOD) or slowly biodegradable ( $S_B$ ) (Henze, 2000; Melcer et al., 2003). Also, biodegradable COD can be identified as filterable ( $CS_B$ ) and particulate ( $X_B$ ) forms which are quickly degradable/undegradable and decantable/undecantable. This filterable fraction may be divided into dissolved material (soluble;  $S_B$ ) or colloidal material ( $C_B$ ) (Orhon et al., 1997). They are rapidly oxidized (or stored) under aerobic conditions by heterotrophic bacteria to produce biomass, which through the process of solid-liquid separation can be recovered as biological sludge, a major substrate for biogas (Fang, 2010).

The slowly biodegradable, mostly particulate  $X_B$ , is not rapidly used up by bacteria due to its complex composition. For this reason, a conversion mechanism through the breakdown by extracellular enzymes into a readily biodegradable form (hydrolysis) is necessary prior to absorption and utilization, leading to delayed consumption of the organic matter (Henze, 2000). On the contrary, the readily biodegradable matter (mostly colloidal  $C_B$  and soluble  $S_B$ ) has relatively simple molecules that can be oxidized (or stored) and consumed directly by heterotrophic

bacteria under aerobic conditions and used for growth of the new heterotrophic biomass  $X_{OHO}$  (Fang 2010; Petersen et al., 2003) known as a bio-transformation process.

Continued research and experience resulting in the development of a high-rate (HR) wastewater treatment process, which is one of the most effective systems, with the possibility of short HRT of 30 to 90 minutes and a high portion of COD capture efficiency (80-85%).

High-rate biological treatment processes allow the recovery of organic matter from wastewater into energy via methanogenesis contributing to the development of energy positive WRRFs with lower carbon footprints (Tilley, 2011; Nogaj et al., 2015).

MBBRs can be operated at high loadings which enable near-exponential growth conditions for the biomass without increasing the reactor size and maximize the storage and bio-transformation of biodegradable organic matter. The transformation of the readily biodegradable material ( $CS_B$ ) to particulate biodegradable organics ( $X_B$ ) is performed by the biofilm developed in carriers which exist in the bio-reactor and consequently, there is no requirement for a return of mixed liquor (Husham et al., 2014; Guanglei et al., 2011).

## **1.2 Scientific hypothesis and objectives**

### **1.2.1 General objective**

The general objective of this project was to maximize the bio-transformation of influent soluble and colloidal organic matter into particulate COD using a high-rate moving bed bioreactor (MBBR) for subsequent physico-chemical capture prior to anaerobic digestion, improving the energy efficiency of wastewater treatment processes.

### **1.2.2 Specific objective**

The specific objective of this study was to develop an innovative high-rate process (inoculum-chemostat) to maximize the bio-transformation of soluble and colloidal biodegradable matter ( $CS_B$ :  $S_B + C_B$ ) into particulate matter ( $X_B$ ) and to capture this  $X_B$  to enhance methane production via anaerobic digestion. A constraint was to minimize the oxidation of biodegradable matter in this process. Results were compared with a typical high-rate MBBR process operated simultaneously.

Inoculum-chemostat can be easily integrated into an existing WRRFs as part of an upgrade process, or by the design of a new facility. In addition, it could improve the energy balance of the WRRFs, while reducing air consumption and the cost of the disposal of sewage sludge.

### **1.2.3 Original scientific hypothesis**

The original scientific hypothesis of this project is that a high-rate innovative MBBR-inoculum and chemostat system can maximize the bio-transformation of soluble and colloidal biodegradable matter ( $CS_B: C_B+S_B$ ) into particulate matter ( $X_B$ ) and its physico-chemical capture to maximize methane production, while minimizing hydrolysis of particulate biodegradable organic matter from the influent ( $X_B$ ).

### **1.2.4 Project phases**

The project was divided into three phases. The first phase, conducted in the laboratory, was performed using 1 L reactors with synthetic wastewater. The preliminary phase was completed from September 2013 to August 2014. The second phase, which is the subject of this thesis, was conducted in two biological treatment process configurations as a pilot scale demonstration. The pilot plant is comprised of two configurations as follows:

- 1) an inoculum-chemostat system combining a high-rate moving bed biofilm reactor (HR-MBBR) playing the role of an inoculum and a continuous flow stirred-tank reactor operated as a chemostat, and
- 2) a typical high-rate MBBR.

The third phase was the modelling of the treatment systems to integrate the results.

### **1.2.5 Organization of this dissertation**

This report is divided into 6 chapters. Chapter 1 presents the general and specific objectives, and the originality of this project. Chapter 2 presents a literature review and theoretical elements to improve the energy efficiency of WRRFs. Chapter 3 presents the methodology for the pilot plant operation conditions, process configurations, methodology, experimental design, measurements and analyses. Chapter 4 presents the results and discussion related to the bio-transformation efficiency of organic matter across the MBBR and inoculum-chemostat process at pilot scale, in

the format of a scientific article to be submitted to the journal of "Water Quality Research Journal". Chapter 5 presents additional results, chapter 6 provides a general discussion and chapter 7 presents conclusions and recommendations.

## CHAPTER 2 LITERATURE REVIEW

The body of the work is the main portion of the thesis or dissertation. This is where the theoretical or mathematical development is set out, along with the methodology and experiment design, measurements, results and analysis, as well as the necessary scientific discussion.

### 2.1 Wastewater characteristics

Anthropogenic waste is released daily as industrial and municipal wastewaters enter WRRFs. The characteristics of wastewater are mostly influenced by factors such as behaviour, lifestyle and living standards which can affect the design of the wastewater treatment systems (Henze and Comeau, 2008). A detailed characterization of wastewater and organic matter is provided for the purpose of wastewater reclamation/reuse to make it possible to perform appropriate and effective treatment methods to meet the discharge standards and levels of purification (Shon et al., 2007).

Chemical oxygen demand (COD) is the main parameter, representing the organic matter content of municipal wastewaters. Based on biodegradability, the total COD can be divided into biodegradable ( $COD_B$ ), unbiodegradable ( $COD_U$ ) and active biomass (heterotrophic biomass  $X_{OHO}$ ) fractions (Figure 2.1) (Melcer et al., 2003; Lee et al., 2006).

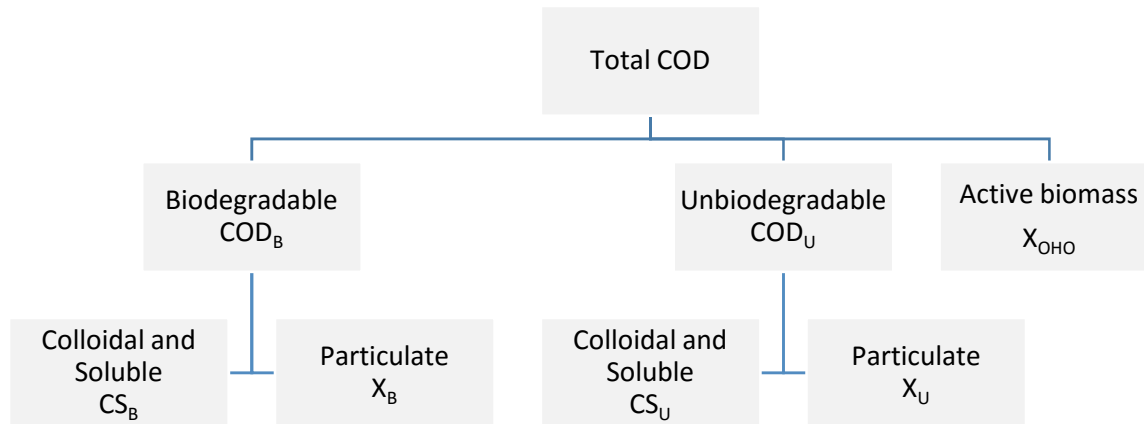


Figure 2.1: Municipal wastewater COD characterization (Melcer et al., 2003)

These fractions can be further subdivided based on their biodegradability into particulate biodegradable ( $X_B$ ), particulate unbiodegradable ( $X_U$ ), colloidal and soluble biodegradable ( $C_B$  and  $S_B$ , respectively) and soluble unbiodegradable ( $S_U$ ) (Melcer et al., 2003; Henze, 2000; Corominas et al. 2003).



Particles in wastewater can also be classified based on size fractions: 25% of COD as dissolved ( $< 0.08 \mu\text{m}$ ), 15% as colloidal ( $0.001\text{-}1 \mu\text{m}$ ), 25% as “supra” colloidal ( $1\text{-}100 \mu\text{m}$ ) and 35% as settling ( $> 100 \mu\text{m}$ ) (Dulekgurgen et al. 2006; Ødegaard, 2000).

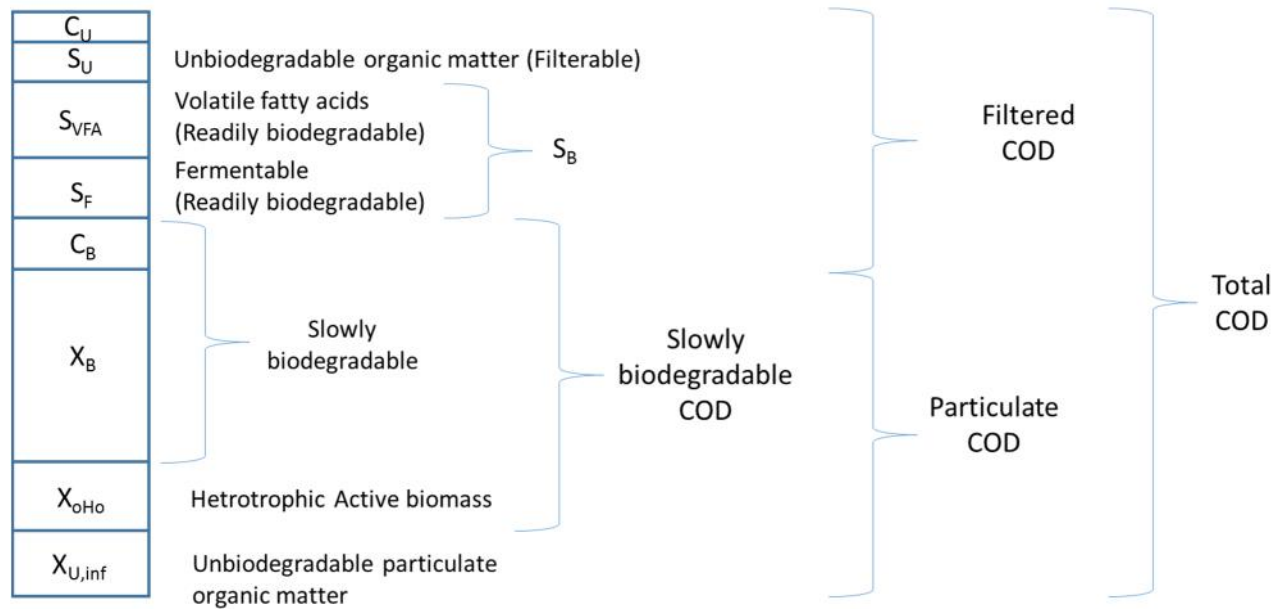


Figure 2.2: Schematic fractionation of COD components for municipal wastewater (adapted from Comeau, 2013)

The composition of typical municipal raw wastewater is presented in Table 2.1. High concentrations of wastewater represent low water consumption and/or infiltration, whereas diluted wastewater shows high water consumption/or infiltration. Storm water can further dilute wastewater (Henze and Comeau, 2008).

Table 2.1: Typical composition of municipal wastewater with minor contribution of industrial wastewater (Henze and Comeau, 2008)

Parameters	Unit	High	Medium	Low
Total COD	mg/L	1,200	750	500
Filtered COD	mg/L	480	300	200
Particulate COD	mg/L	720	450	300
BOD5	mg/L	560	350	230
TSS	mg/L	600	400	250
VSS	mg/L	480	320	200
VFAs	mg HAc/L	80	30	10
Total Kjeldahl nitrogen	mg N/L	100	60	30
Ammonia	mg N/L	75	45	20
Total P	mg P/L	25	15	6
Ortho-P	mg P/L	15	10	4

## 2.2 Wastewater treatment processes

Wastewater treatment is required due to environmental discharge requirements and to meet regional criteria and standards. The treatment processes can be divided into pre-treatment, primary, secondary, tertiary and advanced to reduce different parts of pollutants (Grady et al, 2011; Comeau, 2013; Metcalf and Eddy-Aecom, 2014). A typical municipal sewage treatment plant, including all proposed process configurations, is shown schematically in Figure 2.3.

Two major types of treatment processes can be incorporated in WRRFs, including the physico-chemical processes (coagulation, flocculation, sedimentation, filtration, disinfection) and biological treatment (Ballay et al., 1998). Treatment levels (pretreatment, primary, secondary, tertiary and advanced) are chosen according to the effluent discharge requirements.

The primary treatment is the preliminary level of wastewater treatment; it initiates the process by screening to trap floating solids, followed by primary sedimentation for gravitational removal of suspended solids. This level is sometimes defined as “mechanical treatment”, although chemical products may be used to accelerate the sedimentation process. The biological oxygen demand (BOD) can be reduced by 20-30% and the total suspended solids by some 50-60% during the primary treatment process (Metcalf and Eddy-Aecom, 2014).

Organic matter is consumed and removed as food by heterotrophic bacteria under aerobic conditions during the secondary (biological) treatment, and it is then converted to carbon dioxide, water, and energy for growth of new heterotrophic biomass  $X_{OHO}$  (Fang 2010; Petersen et al.,

2003). The biological process is followed by additional secondary sedimentation to reduce more of the suspended solids. About 85% suspended solids and BOD can be removed across the biological treatment process. Different forms of biological treatments can be incorporated for the removal of organic materials at this level of treatment, i.e. activated sludge, pond and constructed wetland systems, trickling filters (Qasim, 1985; Metcalf and Eddy-Aecom, 2014).

Tertiary treatment can remove over 99% of all pollutants from wastewater even supplying effluent of drinking water quality. The technologies performed at this level of treatment are very expensive, requiring a high level of technical and well trained operators. An example of a typical tertiary treatment process is the modification of a conventional secondary treatment plant to remove additional phosphorus and nitrogen (Qasim, 1985; Metcalf and Eddy-Aecom, 2014). Disinfection usually is built in as a final step before discharge of treated wastewater.

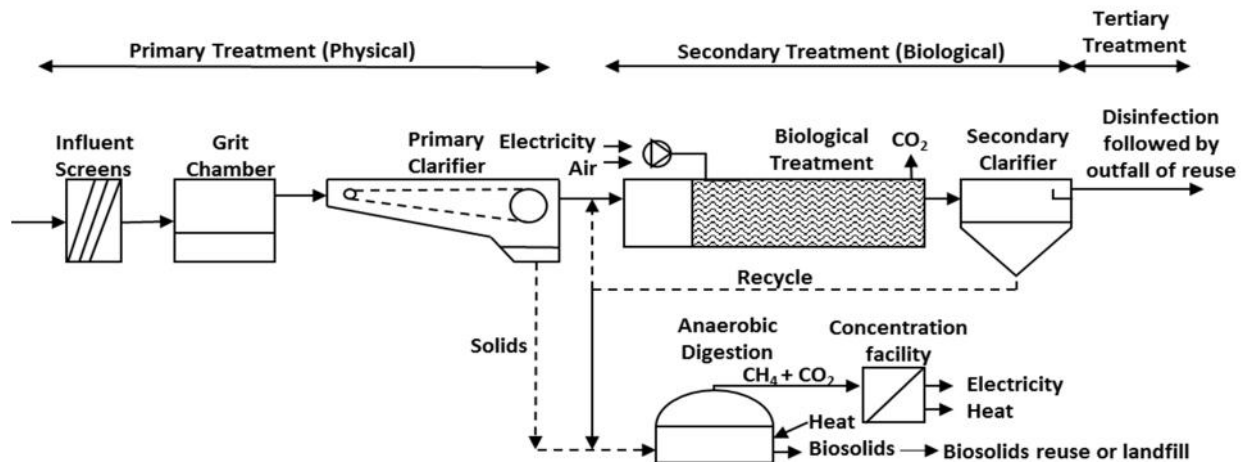


Figure 2.3: Identification of a typical wastewater treatment system (adopted Qasim, 1985; Metcalf and Eddy-Aecom, 2014; Nazaroff and Alvarez-Cohen, 2001)

## 2.3 Biological wastewater Treatment

Biological treatment includes (Grady et al., 2011; Comeau, 2013; Metcalf and Eddy-Aecom., 2014):

- Bio-transformation of particulate, colloidal and soluble biodegradable matter into new biomass and simple compounds, i.e.  $\text{CO}_2$ ,  $\text{H}_2\text{O}$ ,  $\text{N}_2$  or  $\text{HNO}_3$ , etc.
- Adsorption of non-decantable and unbiodegradable particulate and colloidal matter

And

- Conversion or removal of nutrients (N and P)

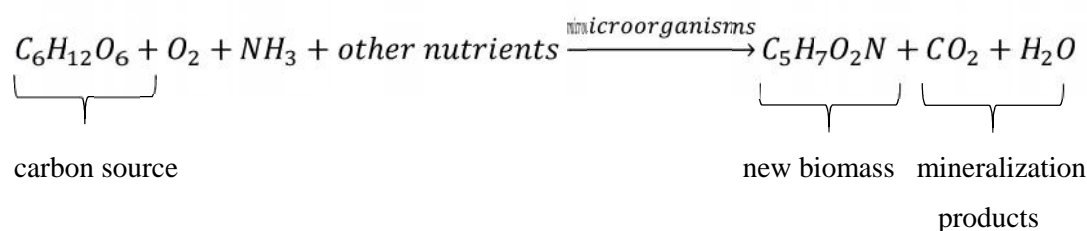
The bio-transformation of biodegradable matter into bacterial biomass is the result of purification of wastewater. Then, biomass can be removed from biologically treated wastewater by means of the secondary clarifier (Gray, 2005).

A biological process is a promising treatment technology to attain revenue from Certified Emission Reduction (CER) credits, as methane gas can be generated from anaerobic digestion and can be utilized as renewable energy. Biological treatments offer advantages such as operational flexibility to support a wide variety of effluent and wastewater characteristics. They also reduce the operating costs, including those of chemical reagents. However, the implantation of biological processes requires a certain area and microbial activity may be sensitive to operating conditions (Seabloom et al., 2005).

Biological processes require free or dissolved oxygen for microorganisms (ordinary heterotrophic organisms;  $X_{\text{OHO}}$ ) activity, converting organic matter to biomass and  $\text{CO}_2$ ; while in the latter process, complex organic matter are degraded into methane,  $\text{CO}_2$  and  $\text{H}_2\text{O}$  across three basic steps via anaerobic digestion (hydrolysis, acidogenesis including acetogenesis and methanogenesis) in the absence of oxygen (Chan et al., 2009; Comeau, 2013; Metcalf and Eddy-Aecom, 2014).

The microorganisms transform the organic matter through two biological oxidation and biosynthesis processes (Gray, 2005). The biosynthesis converts the colloidal and dissolved organic matter into new cells, forming biomass (Eq. 2-1).

(Eq. 2-1)



The biological oxidation end-products (i.e. mineral) remain in the wastewater and they are discharged with the effluent (no new biomass is produced). The biological process can be operated as 1) suspended growth versus attached growth systems, or both 2) continuous process system versus sequencing batch reactor, under aerobic (in the presence of oxygen with constant aeration), anoxic (in the absence of oxygen, but in the presence of nitrite or nitrate (NO<sub>x</sub>)) or anaerobic conditions (Wang et al., 2010).

The suitable method for treatment depends on the characteristics of the wastewater system effluent standards and regulations. The system performance also depends on the operating conditions such as the organic loading rate (OLR), the hydraulic retention time (HRT) and environmental conditions i.e. the pH and temperature. Temperature and pH directly affect the development of distinct species and the growth of microorganisms. Most bacteria cannot operate effectively at a pH higher than 9 or a pH less than 4. Typically, the optimum pH is between 6.5 and 7.5 (Metcalf & Eddy-Aecom, 2014).

Conventional aerobic treatments have been used frequently for industrial and municipal wastewater; however, high-rate bioreactors have been developed to reduce the capital costs of the process and to increase biogas production via anaerobic digestion. The advantage of high-rate biological treatment processes is the ability of organic carbon redirection into possible energy generation, by maximizing the bio-transformation of the substrate and minimizing the oxidation (no mineralization) of colloidal, particulate COD (Jimenez et al., 2015, Ødegaard et al., 2000). There has been a renewed interest in the HR wastewater process due to its high potential to recover energy positive/efficient in WRRFs (Tilley, 2011; Nogaj et al., 2015). The aerobic biological treatment process using high rate bioreactors can achieve a high COD removal (up to 70%) at short HRT (ranging from a few hours to a few days) (Chan, et al., 2009).

An overview of two major types of biological systems including suspended and attached growth processes are described in the next section.

## 2.4 Suspended growth process

Suspended growth (i.e., Activated sludge processes) is an effective process for the removal of organic carbon and nutrients in municipal wastewater plants; in this process, active microorganisms (heterotrophic biomass  $X_{\text{OHO}}$ ) are maintained in a liquid suspension by mixing and aeration methods. Additionally, the mixture of microorganisms and wastewater is transferred to a clarifier and sludge settles out of the treated wastewater, it is then returned to the main reactor to increase the concentration of microorganisms.

Activated sludge is the most commonly used suspended growth process where the  $X_{\text{OHO}}$  are fed by nutrients and organic matter to grow and form the biomass flocs (Chai and Lie, 2008; Spellman, 2008). Air can be introduced in both fine and coarse bubbles to provide respiration to suspend microorganisms and also to provide intimate contact between organic material in the water and oxygen. Following the bio-transformation of soluble and colloidal matter at operated hydraulic retention time (HRT), the mixture of  $X_{\text{OHO}}$  and wastewater is redirected to the secondary clarifier where the flocs are separated by gravitational settling and returned to the bioreactor to seed the process and increase the concentration of microorganisms. Once the microorganisms reach a desired concentration, surplus  $X_{\text{OHO}}$  are wasted from the system. The population ratio of biomass for providing proper food to microorganisms (F/M) is the most important factor affecting efficiency of an activated sludge (AS) system and the health of its biomass. The criterion for wasting sludge is defined based on constant sludge retention time (SRT), which leads to a constant F/M.

## 2.5 Attached growth process

In this process, the  $X_{\text{OHO}}$ , responsible for the conversion and removal of nutrient and organic matter is developed on inert packing material, such as rock, gravel, slag, sand, redwood and a wide range of plastics and synthetic materials.

Attached growth system (biofilm) is a reliable process for the removal of nutrients and organic carbon, since no return activated sludge stream is required (as a considerable advantage) in comparing with the suspended growth process; however, the surplus biomass has to be separated.

The most significant feature of this type of process is the development of biofilm on a carrier; they are mostly diffusion limited. The removal of biodegradable matter is affected by diffusion rates as well as the electron donor and electron acceptor concentration at different layers of the biofilm

(Tchobanoglous et al., 2003), whereas this factor illustrates the difference between attached and suspended growth processes. The liquid dissolved oxygen (DO) associated with diffusion limitation should be considered due to its effects on the biological reaction rate.

Different biofilm systems are already commonly used in WRRF's, such as trickling filters, rotating biological contactors (RBCs), fixed media submerged bio-filters, granular media bio-filters, fluidized bed reactors, moving bed biofilm reactor (MBBR), etc.

## **2.6 Moving bed biofilm reactor (MBBR)**

The development of the moving bed biofilm reactor (MBBR) originated in the 1970s (Loosdrecht et al., 2015). The MBBR provides a wide variety of attached growth systems where synthetic material is used as a carrier media. This process was first developed for the treatment of municipal wastewater for the removal of nitrogen (Ødegaard et. al., 1994). Afterwards, the Norwegian University of Science and Technology (NTNU) and the Norwegian company, Kaldnes Miljø teknologi (now Anox Kaldnes AS), developed a new attached growth system in 1988.

The MBBR, is a biological process in a complete mix with continuous flow across the process, combining the benefits of the activated sludge process and the bio-filter processes and there is no need for sludge recirculation (Ødegaard et al., 1994). The submerged carriers, on which the biofilm grows, are kept in a suspension by either a mixer, or an aeration system, to force an upward movement of the submerged carriers.





In this context, while the suspended growth aerobic process needs a DO concentration of 2-3 mg/L, this level of DO could be a limitation for the attached growth process, especially to achieve a high level of nitrification (Ødegaard, 2006).

The major disadvantage of the MBBR process is the operating costs associated with the aeration process. Fine bubble diffusers are not used in the MBBR process because coarse bubbles are more effective in having the media float to the water surface, which results in very poor oxygen transfer efficiencies. Furthermore, dissolved oxygen concentrations of 3 to 4 mg/L is the optimum level recommended by the manufacturer to maintain the aerobic conditions in the biofilm.

The biomass are fixed on carriers in the MBBR with the surface area provided by the carrier media. This carrier offers a number of advantages, i.e. non-cloggable, lower head loss, no need for back flushing and higher specific surface area. The high-density polyethylene carriers have a specific

gravity of  $0.95 \text{ g/cm}^3$  in the form of a wheel or cylinder reinforced on the inside with a cross to provide harborage for microorganisms (Ødegaard et al., 2006). There are different types of media with different sizes and shapes, provided by the Anox Kaldnes Company, such as K1, Kaldnes K2, Kaldnes K3, K5 and BiofilmChip M. Media size and surface area are usually used to evaluate different kinds of carriers (Table 2.2).

Table 2.2: Different type of Kaldnes MBBR carrier (adapted from McQuarrie and Boltz, 2011)

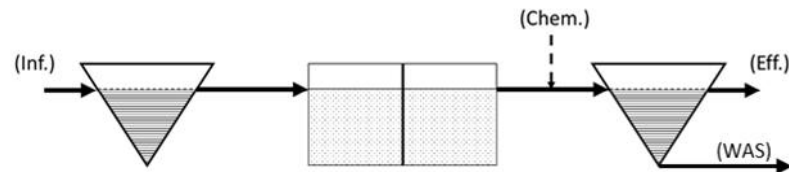
Type of carrier	Model of media	Dimension mm (diameter $\times$ depth)	Surface area ( $\text{m}^2/\text{m}^3$ )
	K1	$9.1 \times 7.2$	500
	K3	$25 \times 10$	500
	K5	$25 \times 3.5$	800
	BiofilmChip M	$48 \times 2.2$	1200

The biomass  $X_{\text{OHO}}$  is grown on the carrier elements (active surface) with a little lighter density than water (Loukidou and Zouboulis, 2001), while introducing air from the bottom of the reactor and a mechanical mixer performs uniform distribution of the plastic biofilm carriers and provides the required oxygen for processing in a complete mixed reactor (Ødegaard et al., 1994).

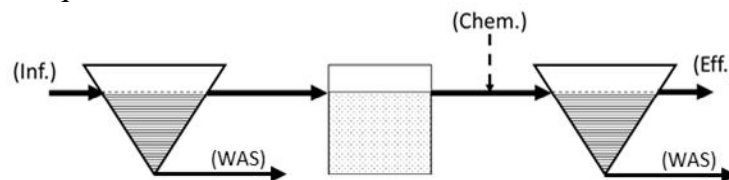
Several applications of different configurations in both industrial and municipal wastewater treatment can be operated by a MBBR process as a biological treatment process for BOD removal, nitrification and/or de-nitrification, or as a pre-treatment system ahead of an existing activated



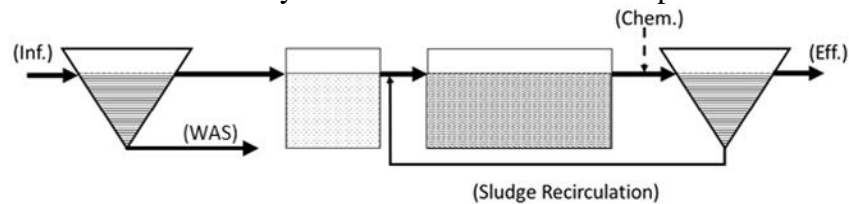
sludge system for increased organic matter removal. Different configurations and flow diagrams are presented in Figures 2.4 and 2.5.



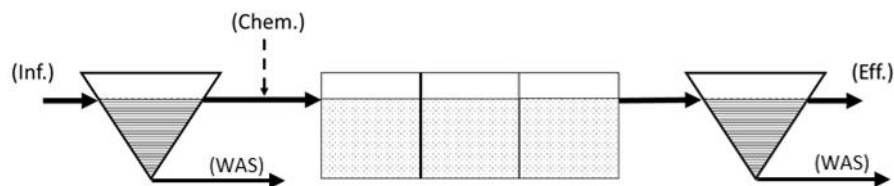
(a) MBBR followed by biomass separation, with chemical addition and flocculation when P removal is required



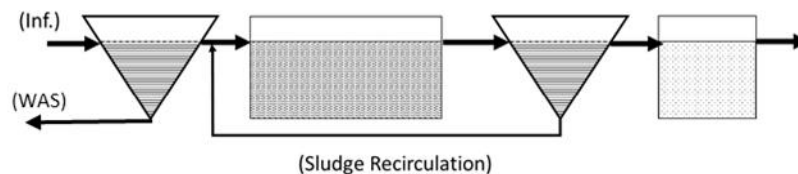
(b) High-rate MBBR followed by flocculation and biomass separation



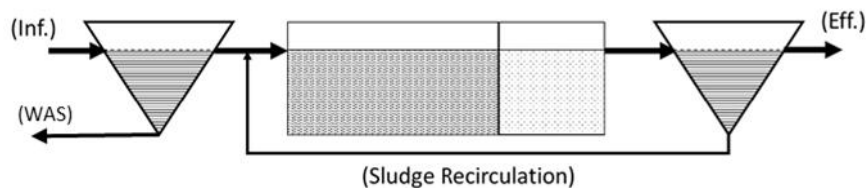
(c) MBBR pretreatment to AS. Used to upgrade existing AS process



(d) Number of MBBR's depending on pre-treatment and waste water characteristics



(e) Tertiary nitrification. MBBR placed after a conventional AS plant. In plants with stringent effluent standard, direct filtration may be used



(f) Combination of AS and MBBR where carriers are added to the last part of the AS reactor

Figure 2.4: Typical MBBR configuration for various application organic carbon and ammonia removal processes (adapted from Ødegaard, 2006)

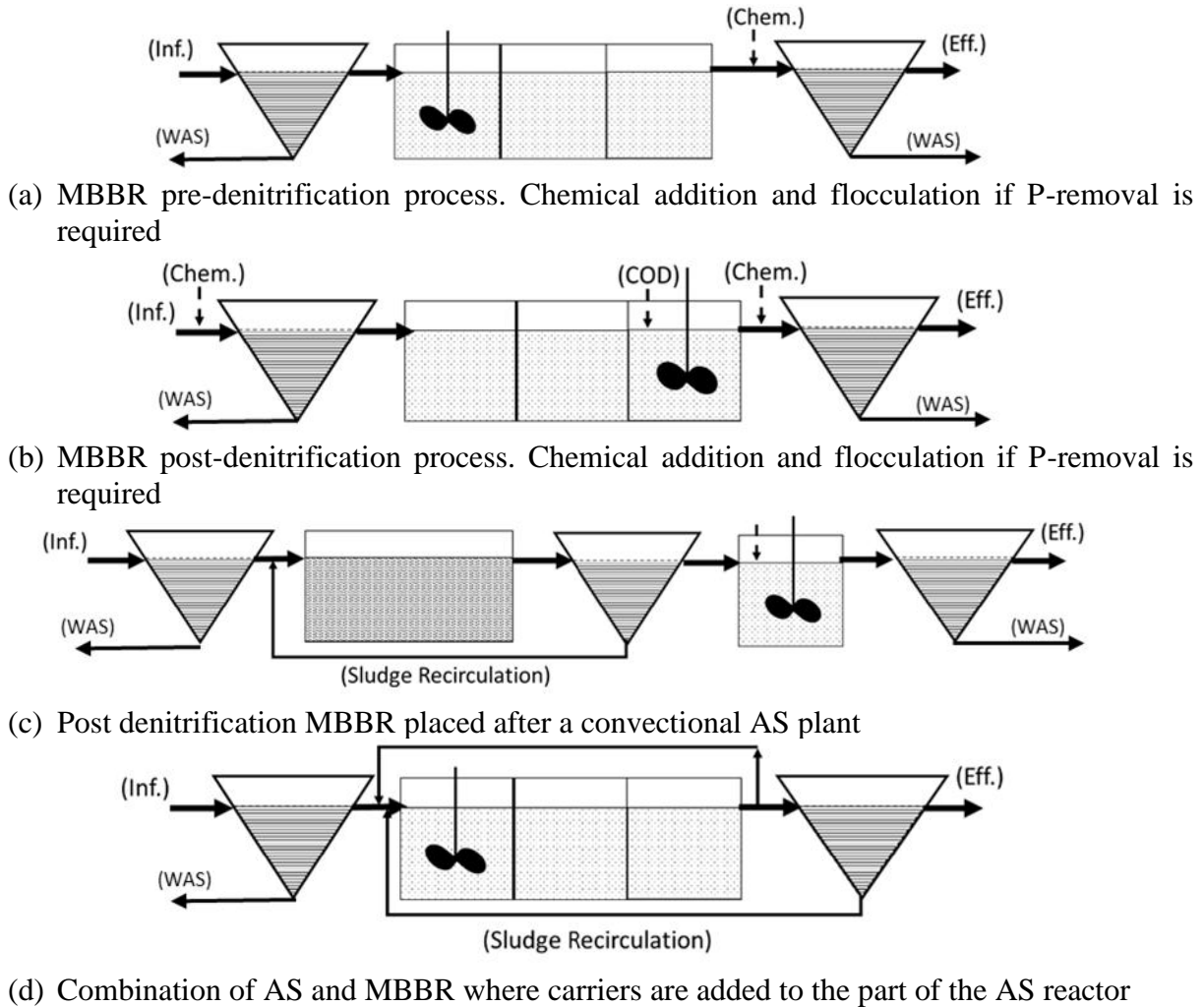


Figure 2.5: Typical MBBR configuration for various application nitrogen removal processes (adapted from Ødegaard, 2006)

The high-rate MBBR (Figure 2.4b) is used for the removal of readily biodegradable matter (mostly soluble); coagulation and floatation are used to separate suspended and colloidal matter. The process results in the maximized bio-transformation of substrate and minimizes the oxidation of colloidal, particulate COD (Jimenez et al., 2015, Ødegaard et al., 2000) to enhance maximum biogas production across the anaerobic digestion (AD) process.

### 2.6.1 Operating conditions

The moving bed biofilm reactor (MBBR) can be operated in aerobic, anoxic and anaerobic processes with system performance affected by various conditions, including hydraulic retention time (HRT), organic loading rate (OLR) and carrier filling rate (Li et al., 2011; Jianlong et al.,

2000). Although, it has been reported that by increasing HRT and OLR, there is an increase in organic matter and nutrient removal efficiency, higher costs and energy consumption requirements were also reported (Guo et al., 2010). That is the reason why low cost and efficient treatment is considered an operational optimum condition in the current research on MBBRs. Consequently, it was necessary to carry out a systematic study on the optimum biofilm carrier filling rate, OLR and HRT in MBBR to treat wastewater efficiently and cost effectively. In addition, the removal of 90% of soluble COD can be achieved in a pilot scale MBBR operation if an optimum media fill volume fraction, HRT, OLR and dissolved oxygen is applied (Chen et al., 2007; Schubert et al., 2013; Sima, 2013).

The dissolved oxygen concentration is also an important factor for biofilm growth: from 2 to 3 mg O<sub>2</sub>/L is required for Bio-transformation of organic matter (Ødegaard, 2006, McQuarrie and Boltz, 2011).

The mechanical mixer is also used to agitate the bulk of the liquid and distribute carriers uniformly in the MBBR reactor, and also to control the thickness of biofilm on the carrier's surface. However, Sheli and Moletta (2007) reported that by increasing OLR, it results in an augmentation of biomass in the MBBR system. About 70% of the total surface area can be represented as an effective surface area due to lesser attachment of biofilm on the outer surface of the media (Majeed et al., 2012). In addition, the size and shape of the media proved to be an effective factor in the system's removal efficiency, due to the biofilm thickness inside and outside of the carrier (Ødegaard, 2000). The thickness of the biofilm on the carrier's surface can be controlled by a mechanical mixer. However, as the organic loading rate (OLR) increased, attached biomass is augmented as well in the MBBR (Sheli and Moletta, 2007). It is recommended that the percentage of media should be below 70% of reactor volume to ensure the media can move freely (Rusten et al., 2006). However, the percentage of the media fill volume fraction can be determined based on the wastewater characteristics and specific treatment goals (Sima, 2013), whilst more than 90% of biomass is attached to the media rather than suspended in the liquid (Schmidt and schaechter, 2011).

However, carrier movement leads to attrition and collision of media in the reactor and causes biofilm detachment from the surface area; this may be mitigated by providing fins on the outside of the carrier media to protect against biofilm loss and to promote biofilm growth. Controlling adequate turbulence eliminates excess biomass and maintains sufficient thickness of biofilm in the

reactor (Ødegaard et al., 2000). Less than 100  $\mu\text{m}$  biofilm thickness is recommended for enough substrate diffusion in the biofilm (Ødegaard et al., 2006).

## 2.6.2 Applications of MBBR

There are more than 500 full scale wastewater treatment processes based on MBBR in 50 different countries which are operated in municipal and industrial wastewater conditions. The MBBR process offers a very compact treatment process, leading to low investment and annual costs. MBBR has been used in a variety of applications and has achieved acceptable results in the case of the removal of different contaminants removal (AnoxKaldnes, 2009).

In most applications, MBBR is used either alone, or combined with the other technologies, such as NEOSEP® membrane bioreactors, actiflo clarification, hydrotech discfilters, dissolved air flotation (DAF), activated sludge or conventional clarifiers. For example, a combination of hydrolysis/acidification with MBBR in which oxidation was used to upgrade centralized wastewater treatment plants in a pharmaceutical industrial park (PIP) in China (Lei et al., 2010). In this combination system, MBBR was used at DO level of above 3 mg/L with the aim of good fluidization of carriers at an HRT of 10.8 h, and HRT was gradually decreased to 5.4 h and then to 3.6 h by the enhancement of inflow. The COD and  $\text{NH}_4^+\text{-N}$  concentration in a good performance of the system were remained stable below 100 and 20 mg/L, respectively.

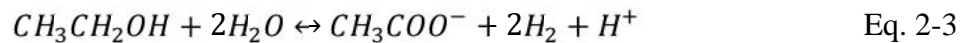
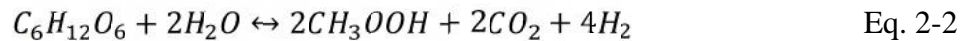
A combination process consists of one or more MBBRs reactor, followed by an activated sludge system patented by AnoxKaldnes™ Company. The high rate biofilm stage is designed to pre-treat the wastewater for the removal of readily bio-degradable organic matter prior to the activated sludge system (AnoxKaldnes™, 2009).

Norway acquired a wastewater treatment plant in Lillehammer WWTP in 2005, and the results indicated average effluent concentrations of 2.2 mg  $\text{BOD}_5\text{/L}$ , 2.9 mg total N/L and 0.12 mg total P/L. In addition, five WWTPs were used in Sweden for the removal of nitrogen and COD from municipal wastewater using the MBBR process

## 2.7 Anaerobic digestion

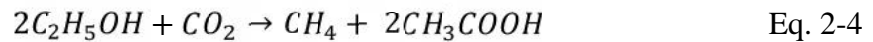
Anaerobic biodegradation of organic matter is performed in the absence, or presence, of oxygen and anaerobic microorganisms, respectively. Metabolic interaction between microorganism groups

resulted in an AD process comprising three stages, hydrolysis, acidogenesis and methanogenesis. The first group of microorganism secretes enzymes that hydrolyze polymeric materials (e.g. glucose and amino acids) to monomers, such as glucose and amino acids. They are subsequently converted to higher volatile fatty acids by acetogenic bacteria,  $H_2$  and acetic acids and into fatty acids in the next step. Finally, the third group of bacteria, methanogenic, converts  $H_2$ ,  $CO_2$  and acetate to  $CH_4$ . The AD is operated by mesophilic and thermophilic bacteria at temperatures ranging from  $30^\circ C$  - $65^\circ C$ . These are subsequently converted by a second group, i.e. acetogenic bacteria to higher volatile fatty acids,  $H_2$  and acetic acid. An acetogenesis reaction is shown below:



Finally, the third group of bacteria, methanogenic, converts  $H_2$ ,  $CO_2$ , and acetate, to  $CH_4$ . These stages are described in detail below (Shefali & Themelis 2002). The AD is carried out in large digesters (Figure 2.3) that are maintained either  $30-40^\circ C$  or  $50-60^\circ C$ , respectively.

The methanogenesis reactions can be expressed as follows:



(Acetic acid) (Methane) (Carbon dioxide)

Among the advantages promised by AD, it may be a source of renewable energy as well as its economic benefits offer a key operational advantage. Biogas generates power and heat leading to a reduction in the energy costs of facilities at plants. It has a considerable benefit and allows the digesters to be self-sufficient energy sources and self-paid to warm the digester (Stuart, 2006; Renou et al., 2008). If the energy (electricity or heat) produced by AD exceeds the internal demand, it can be sold off as generating revenue (Stuart, 2006).

## 2.8 Valorization of organic matter

Wastewater is a renewable resource for biogas production and sustainable water management must be ensured. The primary approach to sustainable water management is a degradation of organics to carbon dioxide ( $CO_2$ ).

Development of compatible treatment processes in WRRFs, which are compact, durable and capable of being operated at different operational conditions, is necessary to invest in wastewater infrastructure.

The main treatment configurations proposed in this project are composed of an innovative combined pilot scale MBBR operated under real wastewater conditions, followed by a chemostat process to biotransform the colloidal and soluble organic matter and recover the produced particulate organic matter. The latter is converted into biogas by anaerobic digestion and then upgraded to energy. This treatment chain is directly in line with the objective of maximizing the recovery of water resources and by using them in an energy efficient manner. In addition, it promotes the use of biosourced reagents, which are potentially biomethanizable, easily accessible and safe for health. (Beltrán-Heredia and Sánchez-Martín, 2009; Heubeck et al., 2011; Sutton et al., 2011; Metcalf and Eddy-Aecom, 2014).

## **2.9 MBBR E+ project**

This research was performed as a part of the MBBR E+ NSERC RDC project which started on January 1, 2013, and included 3 years of lab scale and pilot-scale studies. The pilot unit used (named BA+) had been previously funded by the Canada Foundation for Innovation (CFI). A view of the BA+ pilot unit was shown in Figure 2.6. The MBBR E+ process is proposed to improve the energy efficiency of WRRF, to reduce the carbon footprint and to promote better management of resources. Two liquid configurations were proposed, including the high rate of MBBR and inoculum-chemostat processes) in a separation step to maximize sludge recovery by anaerobic digestion (AD).

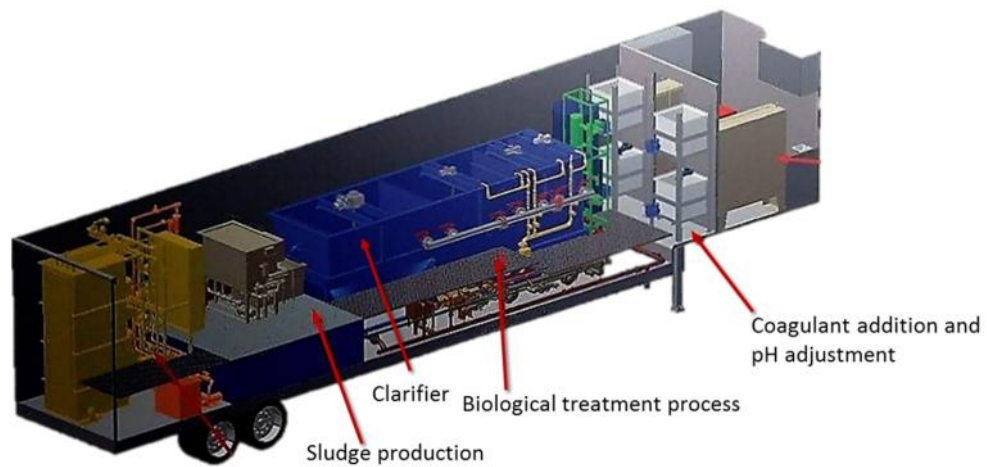


Figure 2.6: A view of interior and exterior BA+ pilot scale wastewater treatment plant

## CHAPTER 3 METHODOLOGY

The setup configuration for the pilot plant and the experimental design corresponding with the project objectives are presented in the first part of this chapter. This is followed by the experimental infrastructure, influent raw wastewater, operating protocols, sampling methodology, data validation process and evaluation of the process efficiency. The information which was provided in this section is complementary information on the methodology for this study that was not mentioned in the paper (chapter 4).

### 3.1 Pilot plant setup configurations

A pilot-scale wastewater treatment plant was installed at Repentigny WRRF in the city of Repentigny, Quebec (Figure 3.1).

Pilot scale treatment configurations were as follows:

- 1) An Inoculum-Chemostat (IC) system combining a high-rate moving bed biofilm reactor (HR-MBBR) playing the role of an inoculum and a continuous flow stirred-tank reactor operated as a chemostat,

and a

- 2) Typical A high-rate MBBR (HR-MBBR).

Both configurations were fed continually with Repentigny WRRF. The MBBR-E+ pilot scale process configurations are presented in Figure 3.1.

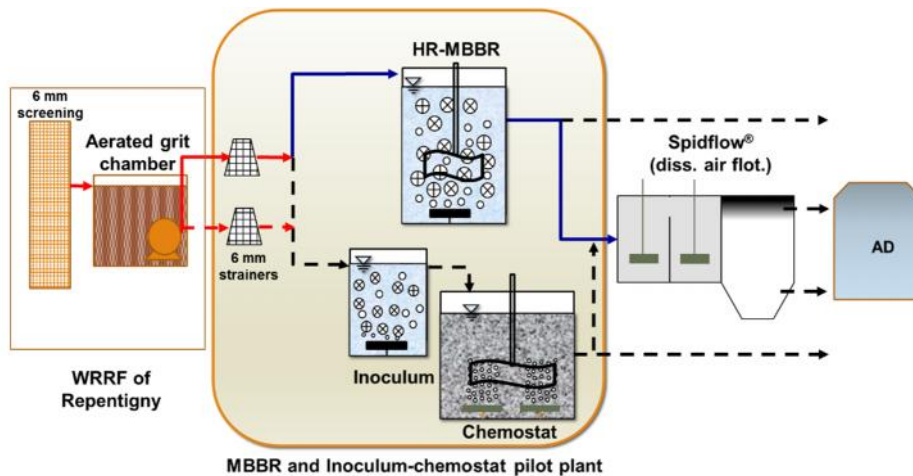


Figure 3.1: MBBR-E+ pilot system configuration



### 3.1.1 Pretreatment process

Two parallel Thompson cone strainers (produced by Miller-Leaman, USA) as a pretreatment process for the physical removal of escaped particles were installed in the influent diverted from Repentigny WRRF to the inlet of the pilot unit to protect the pilot plant's mechanical equipment. Strainers had a stagger customized size of 6 mm (1/4 in); the total surface area and open area are 1290 cm<sup>2</sup> (200 sq.in) and 51%, respectively.

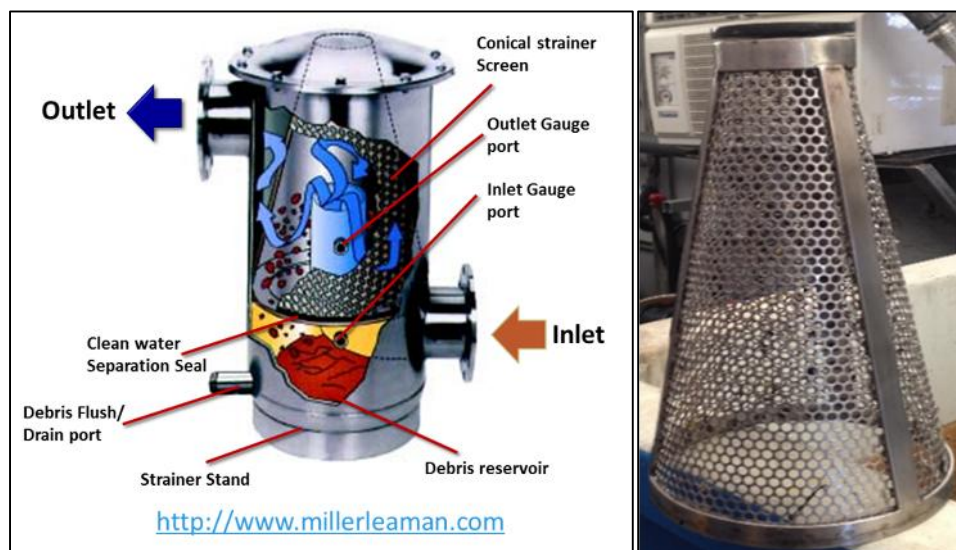


Figure 3.2: Miller-Leaman strainer installed on the influent line of pilot plant

### 3.1.2 MBBR configuration

The MBBR comprised a 2.1 m<sup>3</sup> with an external dimension 77.5 cm × 140 cm × 190 cm (L × W × H) (Figure 3.3). The useful height is 178 cm, which corresponds to a total liquid volume of 1.9 m<sup>3</sup>. It was partially filled with K5 AnoxKaldnes™ media type (d: 26 mm; h: 4 mm, Figure 2.4) that provides a specific surface area of 800 m<sup>2</sup>/m<sup>3</sup> if the reactor fill by 100% of total volume of the reactor. The media fill volume fraction was 37% and 53% of total liquid volume corresponding to 700 L and 1000 L of K5 media, respectively. The actual volume, occupied by the K5 media and biofilm, varies with changes in the thickness of the developed biofilm. This volume ranged from 11% to 16% total liquid volume for a fill volume fraction of 37% and 53%. Therefore, the actual volume of water in the MBBR reactor was changed by 89% and 84% (for 11% to 16% developed biofilm, respectively) of the liquid volume corresponding to 1.7 m<sup>3</sup> and 1.6 m<sup>3</sup>, respectively. HRTs have been recalculated according to changes in the volume of real water in the reactors. The MBBR

reactor is equipped with a mechanical mixer and a diffuser. The blower provided  $16 \pm 1 \text{ m}^3/\text{h}$  per volume of reactor coarse and fine bubbles via diffusers from the bottom of the reactor. Overflows were fitted with screens to prevent the loss of K5 media from the vessel.

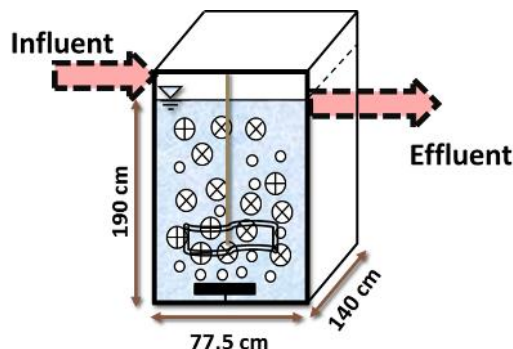


Figure 3.3: MBBR process configuration and dimensions

### 3.1.3 Inoculum-chemostat configuration

The IC process configuration is shown in Figure 3.4. The Chemostat reactor dimension ( $L \times W \times H$ ) was  $210 \text{ cm} \times 140 \text{ cm} \times 210 \text{ cm}$  for a total volume of  $5.6 \text{ m}^3$ . The useful height of the reactor was 60 cm, which corresponds to a working volume of  $4.7 \text{ m}^3$ . The reactor had a mechanical stirrer, blower and a fine bubble diffuser. A  $1.5 \pm 0.2 \text{ m}^3/\text{h}$  per volume of reactor air was introduced from the bottom of the reactor. The Chemostat was installed after a  $0.4 \pm 0.04 \text{ m}^3$  of MBBR (inoculum) to inoculate and enrich the microorganisms in the influent within a short HRT. The inoculum dimensions ( $L \times W \times H$ ) were  $70 \text{ cm} \times 70 \text{ cm} \times 100 \text{ cm}$  with a total volume of  $0.5 \text{ m}^3$ . The liquid depth ranges in the inoculum were changed as 90 cm, 75 cm and 50 cm, based on operated HRT, representing liquid volumes of  $0.44 \text{ m}^3$ ,  $0.37 \text{ m}^3$  and  $0.27 \text{ m}^3$ , respectively. The media fill volume fraction varied between 15% and 23% based on the total liquid volume. The actual volume of liquid considering the volume of media and the inoculum reactor varied between 80% and 95% total volume of  $0.25 \text{ m}^3$  to  $0.42 \text{ m}^3$ . Inoculum is completely mixed in the reactor by introducing fine air bubbles from the bottom of the reactor, without installing a mechanical mixer. Approximately  $2 \pm 1 \text{ m}^3/\text{h}$  air per volume of reactor was introduced to the inoculum to keep DO level up to  $6 \text{ mg O}_2/\text{L}$ .

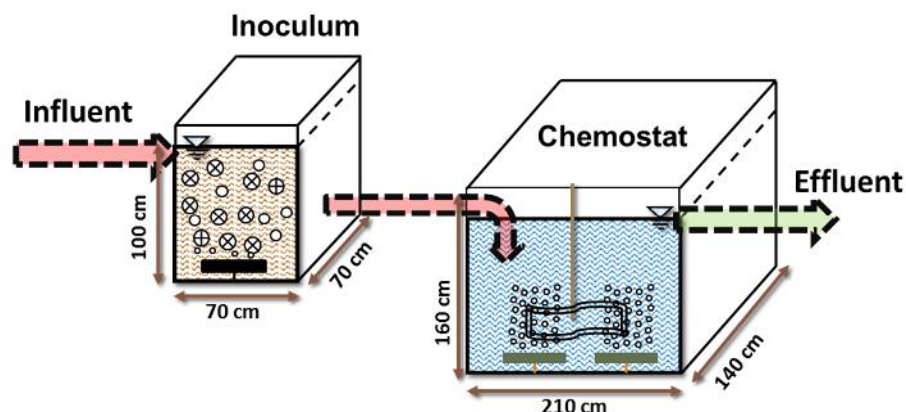


Figure 3.4: Inoculum-chemostat process configuration and dimensions

## 3.2 Raw wastewater characteristics

The influent raw wastewater was provided from Repentigny WRRF. The wastewater was composed of residential, institutional and backwashed water from the Repentigny drinking water treatment plant filters and a small proportion (10%) from industrial sectors. The raw wastewater was pumped from the aerated grit chamber to the pilot after screening (6 mm) and after fat and grease removal. The organic matter fractionation of raw influent is presented in Table 3.1.

Table 3.1: Total COD fractionation of pilot plant influent raw wastewater

total COD (300- 500 mg/L)	X (70-80%)		X <sub>B</sub>
			X <sub>U</sub>
	CS (20-30%)	C (4-15%)	C <sub>B</sub> (3-12%)
			C <sub>U</sub> (1-5%)
		S (15-20%)	S <sub>B</sub> (10-15%)
			S <sub>U</sub> (5%)
<div></div> Measured	<div></div> Calculated	<div></div> Not evaluated	

### 3.3 Operational conditions

The systems operational process is divided into two successive start-up and stabilized phases.

The start-up phase reached stability in one week. The start-up period allowed microorganisms to grow and develop as attached biofilm on the carriers across the MBBR and inoculum. The stable period was defined as the period when no significant change was observed in the characteristics of the effluent ( $CS_B$  concentration) based on the operated conditions. All sample analyses were performed over the stabilized period of each MBBR and IC process under specific operational conditions. The minimum duration for each specific condition was 2 weeks to acquire enough data. The variable conditions used in different experiments are summarized in chapter 4 (Table 1 and 2). The impact of hydraulic retention time (HRT), dissolved oxygen (DO) levels, organic loading rate (OLR) and media fill volume fraction were studied on the operation of the proposed configurations. The temperature was not controlled and changed over time during the pilot operation ( $20 \pm 3$  °C, Appendix A).

### 3.4 Sampling methods

Two series of composite and grab samples were taken three times per week from the influent and effluent of HR-MBBR and IC processes, respectively. Composite samples were scheduled over a period longer than the operated HRT. The schedule of preparing composite and grab samples from influent and effluent of HR-MBBR and IC processes based on the length of  $HRT < 3$  h and  $HRT > 3$  h were performed as shown in Figure 3.5. An additional composite sample from chemostat (Figure 3.5a), was performed if HRT was greater than 3 h ( $t_0$ ,  $t_0 + 1$  h,  $t_0 + 2$  h,  $t_0 + 3$  h,  $t_0 + 4$  h).

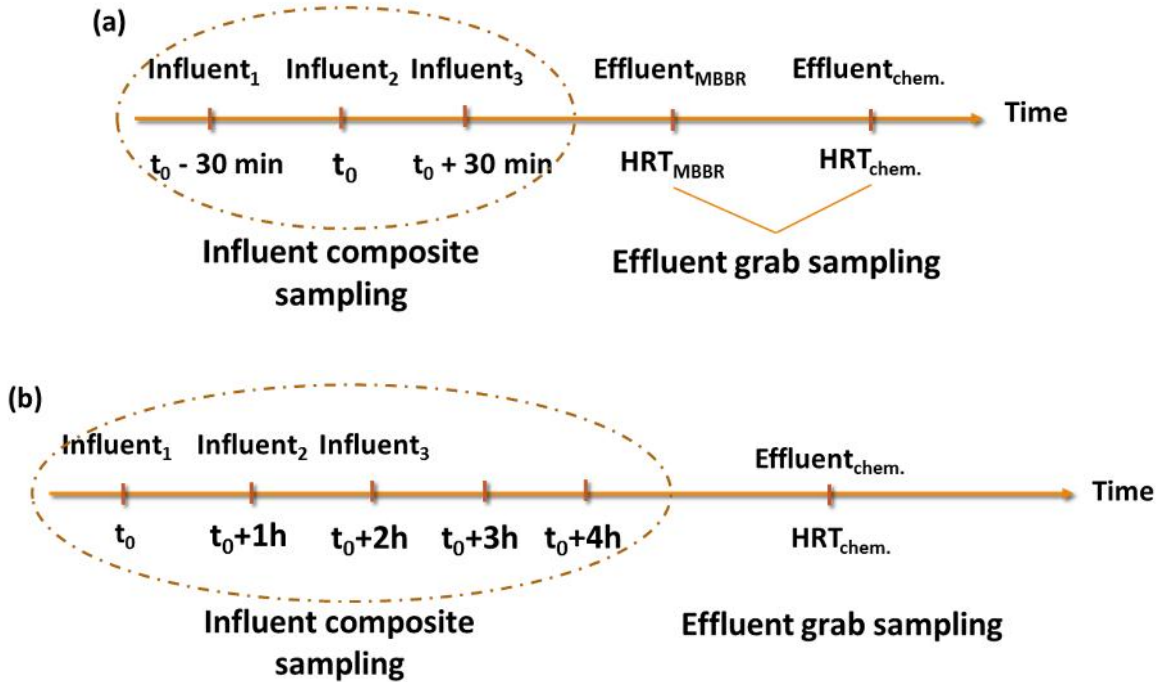


Figure 3.5: Scheduled sampling from influent and effluent of (a)  $HRT < 3 \text{ h}$  (b)  $HRT > 3 \text{ h}$

## 3.5 Analytical determinations

### 3.5.1 Total and filtered COD

Total COD and filtered COD of influent and effluent samples were measured based on the standard method 5220D, closed reflux colorimetric method (APHA et al., 2012), using the Hach Test-in-Tube (TNT) kits (Hach, Inc.). The COD tests were performed with high (0 to 1500 mg/L) and low (0 to 150 mg/L) range TNT tubes, while two standard samples were prepared for each test using dried potassium hydrogen phthalate (KHP). For each test 2 ml of sample were placed into an Hach vial, shaken, and digested on a heating block for 2 hours. The digested samples, after cooling to ambient temperature, were analyzed spectrophotometrically. The colloidal and filtered fraction (CS) represents the COD concentration filtered through a  $1.2 \mu\text{m}$  filter, while the soluble COD (S) portion measured after flocculation (by  $\text{ZnSO}_4$ ) and filtered through  $0.45 \mu\text{m}$  filter (soluble COD: S). The unbiodegradable soluble fraction ( $S_U$ ) was considered 5% total COD (EnviroSim, 2014).

Unbiodegradable colloidal fraction was determined from equation 3.2, based on the assumption in Eq. 3.1.

$$\frac{CS_U}{S_U} = \frac{CS}{S} \quad \text{Eq.3.1}$$

$$C_U = CS_U - S_U \quad \text{Eq.3.2}$$

The colloidal COD (C) and particulate fractions (X), colloidal and soluble biodegradable ( $C_B$  and  $S_B$ ) are calculated from Equations 3.3, 3.4, 3.5 and 3.6, respectively.

$$C = CS - S \quad \text{Eq. 3-3}$$

$$X = COD - CS \quad \text{Eq. 3-4}$$

$$C_B = C + C_U \quad \text{Eq. 3-5}$$

$$S_B = S + S_U \quad \text{Eq. 3-6}$$

### 3.5.2 VSS and TSS

TSS and VSS analysis were measured based on standard method 2450D. The remaining solids on 1.2  $\mu\text{m}$  filter (MF-Millipore™, EMD Millipore, USA) were dried at 105 °C and 550 °C in ovens for measuring TSS and VSS, respectively (APHA et al., 2012). All filters were washed with distilled water prior to testing and then placed in aluminium dishes, and dried for 1 h and weighed before usage. Values were recorded and used in the following equations to determine VSS and TSS concentrations:

$$TSS = \frac{(B-A)(1000 \text{ mg/L})}{\text{Sample volume, L}} \quad \text{Eq. 3-7}$$

$$VSS = \frac{(B-C)(1000 \text{ mg/L})}{\text{Sample volume, L}} \quad \text{Eq. 3-8}$$

Where

A (g): weight of the filter (dried at 105 °C) + aluminum container

B (g): weight of the filter + aluminum container + residue (dried at 105 °C for 1 hour)

C (g): weight of the aluminum container + filter and residue (combusted at 550 °C for 20 min).

### 3.5.3 Biofilm mass

The mass of developed biofilm was measured in both HR-MBBR and inoculum during each of the operational conditions. For this purpose, 100 media carriers were collected from the reactors at

each sampling time, and divided into five replicates of 20 media carriers. For each replicate, this procedure was applied:

- All the carriers were dried at 105 °C for 24 h and then weighed to determine the total mass (media + biofilm);
- Carriers were placed in a container with water and stirred vigorously to detach the biomass. This step was repeated five times and the entire washing water was retained and the exact volume was measured using a graduated cylinder. Analyses of TSS and VSS were performed to obtain a ratio VSS/TSS;
- The media were washed in 6 M NaOH for 30 minutes, rinsed with warm water to remove any remaining biomass, dried again at 105 °C and all of the carrier was weighed in determining the mass of the media without the biofilm.

The difference between the total mass and the mass of the media at the final step represented the total amount of biofilm grown in all 20 carriers. Biofilm mass per carrier was calculated by dividing the total amount of biofilm by the number of balls (20).

### 3.5.4 Sludge volume index

SVI variation from effluent samples were determined to monitor settling characteristics of MBBR and chemostat suspensions. The sludge volume index (SVI) is the volume in milliliters occupied by 1 g of a suspension after 30 min settling (APHA et al., 2012). The suspended solids can be determined by dividing the settleable (after 30 min) sludge volume of one liter, well-mixed sample by total suspended solids of wastewater samples. The formula for SVI is written:

$$SVI = \frac{\text{settled sludge volume (mL/L)} \times 1000}{\text{Total suspended solids (mg/L)}} \quad \text{Eq. 3-9}$$

### 3.5.5 Other parameters (pH, DO, Nitrate)

The probes (Hach Company), connected to the automation system (PLC), were applied to continuous real-time monitoring and controlling of DO, pH and temperature. Oxygen utilization rate measurements (OUR) were measured every two hours and three hours in the MBBR and in the chemostat reactor, respectively.



### 3.6 Process efficiency

The process efficiency was evaluated according to the criteria of removal of soluble and colloidal biodegradable organic matter and observed bio-transformation of soluble and colloidal material into particulate matter. The removal efficiency was calculated by a correlation between soluble and colloidal biodegradable organic matter ( $CS_B$ ) from the influent and effluent:

$$CS_B \text{ Removal efficiency (\%)} = \frac{CS_{bAff}(\frac{mg}{L}) - CS_{bEff}(\frac{mg}{L})}{CS_{bAff}(\frac{mg}{L})} \times 100 \quad \text{Eq.3-10}$$

$$CS_{b_{inf}}(mg/L) = CS_{inf}(\frac{mg}{L}) - CS_{U_{inf}}(\frac{mg}{L}) \quad \text{Eq.3-11}$$

$$CS_{b_{Eff}}(mg/L) = CS_{Eff}(\frac{mg}{L}) - CS_{U_{Eff}}(\frac{mg}{L}) \quad \text{Eq.3-12}$$

The specific removal per used surface area ( $A_u$ ) of media was calculated as:

$$\text{Specific removal (g } CS_b m^{-2} d^{-1}) = Q(\frac{m^3}{d}) * \left( \frac{CS_{b_{inf}}(\frac{mg}{L}) - CS_{b_{Eff}}(\frac{mg}{L})}{A_u (m^2)} \right) \quad \text{Eq.3-13}$$

The yield of  $X_B$  and observed yield ( $Y_{obs}$ ) were calculated using these equations:

$$\text{Yield } X_B \left( \frac{g \text{ COD}_P}{g \text{ COD}_{CS\text{removed}}} \right) = \frac{[COD(\frac{mg}{L}) - CS_{Eff}(\frac{mg}{L})] - [COD_{inf}(\frac{mg}{L}) - CS_{inf}(\frac{mg}{L})]}{CS_{b_{inf}}(\frac{mg}{L}) - CS_{b_{Eff}}(\frac{mg}{L})} \quad \text{Eq. 3-14}$$

$$Y_{obs} \left( \frac{g \text{ TSS}}{g \text{ DCO}_{CS\text{removed}}} \right) = \frac{TSS_{Eff}(\frac{mg}{L}) - TSS_{inf}(\frac{mg}{L})}{CS_{b_{inf}}(\frac{mg}{L}) - CS_{b_{Eff}}(\frac{mg}{L})} \quad \text{Eq.3-15}$$

Otherwise, the retention time of the biofilm (SRT) and the maximum specific growth rate were measured as follows:

$$SRT(d) = \frac{\text{Total mass of biomass}}{\text{Total mass of purged biomass}} \quad \text{Eq.3-16}$$

$$SRT(d) = \frac{V(L) * VSS_{inf}(\frac{mg}{L}) + \text{number of media} * M_{VSS} (g)}{Q(\frac{m^3}{d}) * VSS_{eff}(\frac{mg}{L})} \quad \text{Eq.3-17}$$

$$\mu_m \left( \frac{g \text{ VSS}}{g \text{ VSS} \cdot d} \right) = \frac{\left( K_s + S_{b_{eff}}(\frac{mg}{L}) \right) * (1 + k_d * SRT(d))}{SRT(d) * S_{b_{eff}}(\frac{mg}{L})} \quad \text{Eq.3-18}$$

The effect of temperature was evaluated using the coefficient  $\theta$  using the modified Arrhenius equation:

$$k_T = k_{20} * \theta^{(T-20)} \rightarrow k_{20} = k_T * \theta^{(20-T)} \quad \text{Eq.3-19}$$



## CHAPTER 4      ARTICLE1: ORGANIC MATTER CAPTURE BY A HIGH RATE INOCULUM-CHEMOSTAT AND MBBR SYSTEM

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### Abstract

This main objective of this study was to develop an innovative process to maximize the bio-transformation of colloidal and soluble biodegradable matter ( $CS_B$ ) into particulate matter ( $X_B$ ) for energy recovery via methane production. Two configurations were studied, 1) a high-rate MBBR and 2) an inoculum-chemostat (IC) system consisting of a very high-rate moving bed biofilm reactor (HR-MBBR) inoculating a continuous flow stirred-tank reactor operated as a chemostat. The effect of process parameters such as hydraulic residence time (HRT), specific organic loading rate (SOLR) and dissolved oxygen (DO) level on the performance of the two high rate systems was determined using real wastewater at pilot scale. Results showed that in the HR-MBBR process, a very high  $CS_B$  bio-transformation efficiency ( $90 \pm 3\%$ ) was obtained in a wide range of SOLRs ( $2.0$  to  $5.5 \text{ g } CS_B \text{ m}^{-2} \text{ d}^{-1}$ ) corresponding to an optimum HRT of 36 minutes. The IC process reached

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\* Corresponding author

a maximum  $CS_B$  bio-transformation efficiency of  $77 \pm 3\%$ , at SOLRs ranging from 22 to 30 g  $CS_B$   $m^{-2} d^{-1}$  at an HRT of 3.7 hours. The DO concentration in the HR-MBBR influenced the  $CS_B$  bio-transformation ratio, while the HRT and the SOLR were the dominant factors influencing the  $CS_B$  bio-transformation ratio in the IC process. Based on these results, the IC process could be an interesting alternative to high rate systems towards obtaining energy positive/efficient from water resource recovery facilities.

Key words: chemostat, COD oxidation, high-rate MBBR, organic matter capture

## 4.1 Introduction

Environmental protection requirements and energy demand are major factors driving the energy-efficiency of water resource recovery facilities (WRRFs). Conventional processes, like activated sludge (AS), are widely used for wastewater treatment, but they require a significant amount of energy (Jimenez et al., 2015). Therefore, process optimization and innovative treatment strategies are required to improve the energy balance and obtain cost-effective WRRFs (Metcalf and Eddy-Aecom, 2014; Meerburg et al., 2015).

A central approach to obtain energy-positive WRRFs is to maximize the capture of organic matter for energy production via methanogenesis. The biodegradable organic matter consists of readily (RBCOD) and slowly biodegradable (SBCOD) fractions (Henze, 2000; Melcer et al., 2003). Readily biodegradable matter, is composed of soluble ( $S_B$ ) and colloidal ( $C_B$ ) matter that can be oxidized or stored directly by heterotrophic bacteria under aerobic conditions and used for the growth of new heterotrophic biomass  $X_{OHO}$  via bio-transformation processes. Slowly biodegradable matter (mostly particulate matter,  $X_B$  and ordinary heterotrophic organisms,  $X_{OHO}$ ) requires conversion into a readily biodegradable form by hydrolysis prior to absorption and utilization. Thus, optimizing aeration, minimizing hydrolysis, minimizing oxidation of particulate matter and capturing biodegradable organic matter to be sent to anaerobic digestion can improve the energy efficiency of WRRFs (Ødegaard et al., 2000; Jimenez et al., 2015).

High-rate biological treatment profits from the high bacterial activity under high food-to-microorganism ratios and low solid retention times (SRTs) with relatively short hydraulic retention times (HRTs) resulting in the maximization of bio-transformation and capture of organic matter from wastewater (Jimenez et al., 2015 and Grady et al., 2011). The high-rate moving bed bioreactor

(HR-MBBR) is a promising process which is successfully used for organic matter recovery at low HRTs (30-90 min) while still maintaining a high COD removal efficiency (80- 85%) (Ødegaard, 2000). Biomass is grown in such HR processes, transforms  $C_B$  and  $S_B$  ( $CS_B$ ) into  $X_{OHO}$ , minimizing the oxidation of  $X_B$  while increasing the production of  $X_{OHO}$  thus maximizing the energy generation potential (Jimenez et al., 2015; Brosseau et al., 2015).

The main objective of this study was to develop an innovative process combining an HR-MBBR and an AS process to maximize the bio-transformation of  $CS_B$  into  $X_B$  for energy recovery via methane production. For this purpose, two pilot-scale treatment configurations, including a high-rate MBBR (HR-MBBR) in parallel with a very high rate MBBR acting as an inoculum and an activated sludge chemostat (IC) system, were tested to address the following specific objectives:

- (a) Determine the operational parameters (HRT, specific organic loading rate, dissolved oxygen) to maximize the performance of each treatment process and
- (b) Maximize the bio-transformation of  $CS_B$  into  $X_B$  to allow the capture of  $X_B$  to maximize methane production.

## 4.2 Materials and methods

### 4.2.1 Pilot plant setup and configurations

The pilot plant, comprising of (a) an HR-MBBR ( $1.6 \text{ m}^3$ ) and (b) a very high rate MBBR inoculum ( $0.4 \text{ m}^3$ ) followed by a chemostat ( $4.7 \text{ m}^3$ ), (Figure 4.1), was installed at the Repentigny municipal WRRF, Quebec. The raw wastewater influent containing about 10% industrial loading was subjected to 6 mm screening, fat and grease removal and grit removal prior to being fed to the pilot plant trains. The wastewater characteristics and operating conditions of the HR-MBBR and IC are presented in Table 4.1.

Additional screening was provided by another 6 mm punched hole strainer which was connected at the inlet of both systems to remove trash and which was cleaned manually every two days. The HR-MBBR and inoculum were filled with the carrier type K5 from AnoxKaldnes<sup>TM</sup> with a specific surface area of  $800 \text{ m}^2/\text{m}^3$ . All reactors were completely mixed and were equipped with fine and coarse bubble aeration systems, and a mechanical mixer. Probes (Hach) connected to the automation system (PLC), were used for real-time monitoring of dissolved oxygen (DO), pH and temperature.

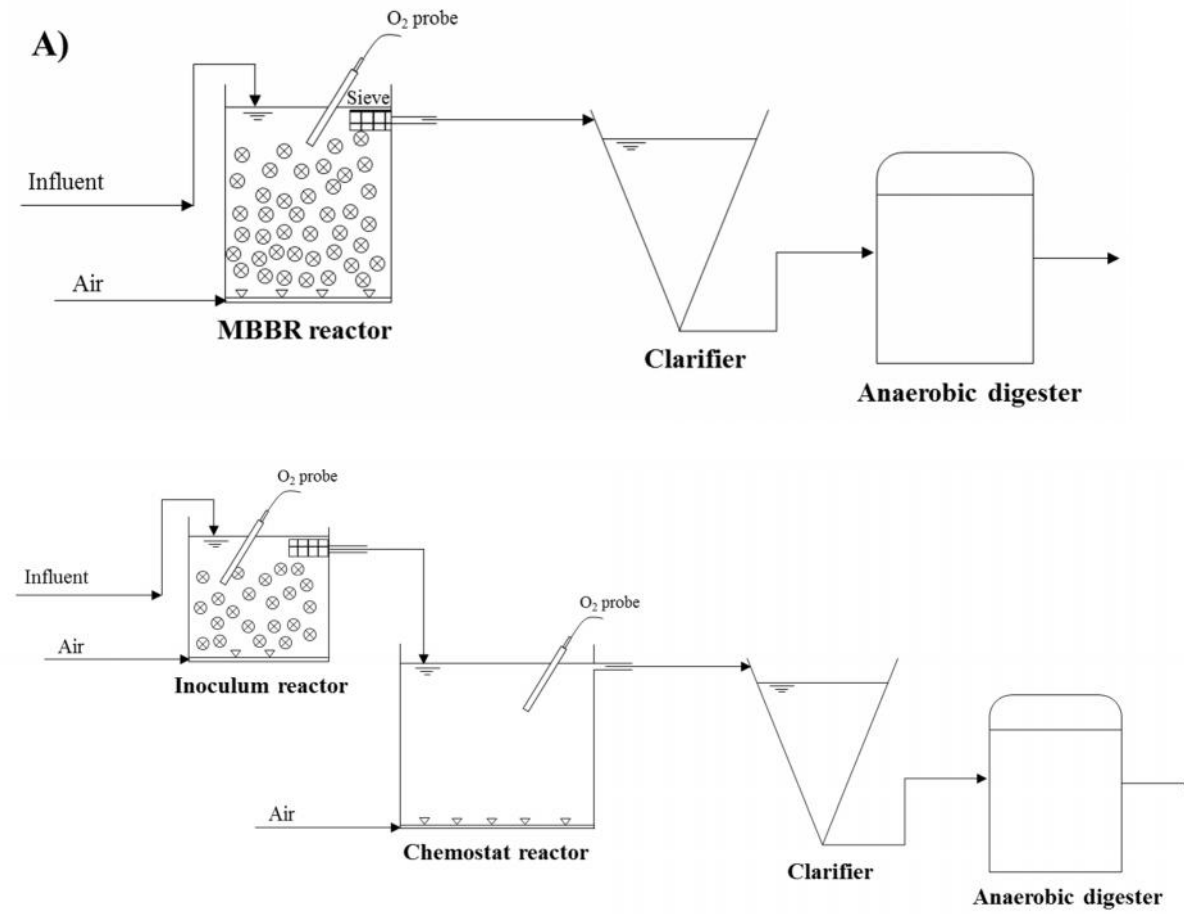


Figure 4.1: Configuration of the (A) HR-MBBR and (B) IC treatment systems

Table 4.1: Influent and process and operating characteristics for the pilot-scale reactors at different operating conditions (OC)

Parameter	Units	HR-MBBR					Inoculum					Chemostat				
		OC1	OC2	OC3	OC4	OC5	OC1	OC2	OC3	OC4	OC5	OC1	OC2	OC3	OC4	OC5
Influent																
Q	m³/h	1.8	2.6	3.8	1.8	3.8	2.0	1.35	1.0	2.0	2.0	2.0	1.4	1.0	2.0	2.0
Total COD	mg/L	403 ± 43	432 ± 77	409 ± 59	400 ± 111	390 ±100	440 ± 81	454 ± 22	455 ± 39	381 ± 36	370 ± 24	429 ± 48	440 ± 24	429 ± 41	380 ± 23	271 ± 10
Colloidal COD	mg/L	26 ± 13	26 ± 14	37 ± 8	31 ± 14	21 ± 15	27 ± 13	16 ± 9	52 ± 13	24 ± 14	29 ± 12	21 ± 13	10 ± 4	32 ± 13	8 ± 5	29 ± 5
Soluble COD	mg/L	61 ± 8	68 ± 15	69 ± 10	60 ± 9	61 ± 14	64 ± 14	68 ± 8	73 ± 7	61 ± 4	56 ± 11	55 ± 12	53 ± 6	50 ± 3	53 ± 7	42 ± 2
Process and operating characteristics																
Liquid volume	m³	1.6	1.6	1.6	1.6	1.7	0.42	0.36	0.36	0.43	0.43	4.7	4.7	4.7	4.7	4.7
HRT	min	54	36	25	54	25	13	16	22	13	13	141	209	282	141	141
COD loading	kg COD/d	17 ± 2	29 ± 5	39 ± 6	14 ± 4	38 ± 9	21 ± 4	14 ± 1	11 ± 1	17 ± 2	14 ± 1	21 ± 2	14 ± 1	10 ± 1	239 ± 23	182 ± 6
Fill volume fraction	m³/m³	50	50	35	50	50	13	16	16	25	45	-	-	-	-	-
SOLR*	g m <sup>-2</sup> d <sup>-1</sup>	26 ± 3	46 ± 9	85 ± 12	23 ± 6	59 ± 14	474 ± 87	319 ± 16	237 ± 20	238 ± 1	196 ± 1	-	-	-	-	-
Temp	°C	17-22	17-22	18-21	17-18	18-21	17-22	17-22	18-21	19-20	16-17	17-22	17-22	18-21	19-20	16-17
DO	mg/L	2-4	3-4	3-4	1.5-2	3-4	5-6	5-6	5-6	4-6	4-6	6-7	6-7	6-7	6-7	6-7

\* The specific organic loading rate (SOLR) was calculated based on total COD.

### 4.3 Aeration

The aeration system in the HR-MBBR process provided  $16.3 \pm 1.2 \text{ m}^3/\text{h}$  provided through coarse (1/3) and fine (2/3) bubbles to ensure proper aeration and media mixing. The aeration system in the IC system provided  $1.5 \pm 0.2$  and  $2.5 \pm 1.3 \text{ m}^3/\text{h}$  in the inoculum and chemostat processes, respectively, via fine bubble diffusers.

### 4.4 Sampling and analytical methods

The influent to each process was sampled 2 to 5 times per week. Multiple grab samples (taking into account the HRT) from the influent were mixed together to obtain a homogeneous composite sub-sample. Total and soluble COD, total and volatile suspended solids were analyzed at each sampling point according to Standard Methods 5220D (APHA et al., 2012). Filtered COD was determined using both  $1.2 \mu\text{m}$  glass microfiber filters (Whatman® 934-AH™, GE Healthcare Life Sciences, GBR) and  $0.45 \mu\text{m}$  cellulose membranes (MF-Millipore™, EMD Millipore). Flocculated-filtered COD (ffCOD) was measured using the method developed by Mamais et al. (1993). COD fractions characterized were thus particulate COD ( $X_{\text{COD}} > 1.2 \mu\text{m}$ ), colloidal and soluble COD ( $CS_{\text{COD}} < 1.2\mu\text{m}$ ) and soluble COD ( $ff\text{COD} = S_{\text{COD}} < 0.45 \mu\text{m}$ ). Colloidal COD ( $C_{\text{COD}}$ ) fraction was calculated from the difference between  $CS_{\text{COD}}$  and  $S_{\text{COD}}$ . The colloidal and soluble unbiodegradable fraction ( $S_U$ ) was considered to be the typical 5% of the total COD (EnviroSim, 2014). The following formula was used to calculate the  $C_U$ ,  $C_B$  and  $S_B$ , according to  $S$ ,  $C$ ,  $CS$  and  $S_U$  (given above) values:

$$C_U = S_U \times (C_S/S - 1) \quad \text{Eq.4-1}$$

$$C_B = C_{\text{COD}} - C_U \quad \text{Eq.4-2}$$

$$S_B = S_{\text{COD}} - S_U \quad \text{Eq.4-3}$$

The DO was measured with a portable DO-meter (HQ40d, Hach Company) and an LDO® probe (Hach Company).

The biofilm mass was measured every week by collecting carriers (20 carriers per sampling event) dried at  $105^\circ\text{C}$  overnight and weighed. The carriers were then soaked in 6% NaOH for 30 min to recover the biofilm from the carrier surface, after which the carriers were scraped clean and dried again at  $105^\circ\text{C}$  overnight. The difference between the dry weight of the carriers before and after

cleaning represented the mass of biofilm on the carriers. The amount of biofilm per square meter of protected surface area of carriers ( $\text{g TSS/m}^2$ ) was determined by dividing the obtained total solids (TS) of the detached biofilm over the protected surface area of the number of carriers sampled (Andreottola et al. 2000, 2003). Considering a protected surface area of  $23 \text{ cm}^2/\text{carrier}$  allowed to determine the specific biofilm concentration in  $\text{g/m}^2$ .

## 4.5 Statistical analysis

Statistical comparisons between the HR-MBBR and IC treatment efficiencies were conducted using the t-test function in Microsoft Excel 2013 with the least significant difference of  $P < 0.05$ .

## 4.6 Results and discussion

The effect of HRT, SOLR, media fill volume fraction and oxygen uptake rate (OUR) was considered in the following sections for maximizing the production of biodegradable sludge, based on the maximization of the removal efficiency of  $\text{CS}_B$  (bio-transformation of  $\text{CS}_B$  into  $\text{X}_{\text{COD}}$ ) as well as the minimization of biodegradable particulate matter ( $\text{X}_B$ ) hydrolysis.

A summary of the pilot-scale HR-MBBR and IC effluent characteristics of the five operating conditions is presented in Table 4.2.

No significant nitrification occurring as expected under such high-rate conditions as shown by the very low concentration of nitrate ( $0.1 \text{ mg N/L}$ ) in the effluent of the HR-MBBR and IC processes.

## 4.7 Effect of HRT, SOLR and DO on bio-transformation of $\text{CS}_B$ and hydrolysis $\text{X}_{\text{COD}}$

The effect of HRT on the bio-transformation of the  $\text{C}_B$  and  $\text{S}_B$ , and the hydrolysis of  $\text{X}_{\text{COD}}$  in the HR-MBBR at operating conditions OC1, OC2 and OC5 base on the HRT are shown in Figure 4.2a and Table 4.2.  $\text{C}_B$  and  $\text{S}_B$  bio-transformation, increased from  $75 \pm 5\%$  to  $83 \pm 6\%$  by increasing HRT from 25 min to 54 min. The bio-transformation of  $\text{C}_B$  and  $\text{S}_B$  into  $\text{X}_B$  showed no significant difference at HRTs longer than 36 min and reached a plateau at  $85 \pm 6\%$  (below an SOLR  $36 \pm 6 \text{ kg COD m}^{-2} \text{ d}^{-1}$ ).

Table 4.2: Summary of operating conditions, effluent characteristics and process performance for the pilot-scale HR-MBBR and IC

Parameter	Symbol	Units	HR-MBBR					IC				
			OC1	OC2	OC3	OC4	OC5	OC1	OC2	OC3	OC4	OC5
Operating conditions												
COD loading	-	kg COD/d	17 ± 2	29 ± 5	39 ± 6	14 ± 4	38 ± 9	21 ± 2	15 ± 1	11 ± 1	18 ± 2	18 ± 7
HRT	-	min	54	36	25	54	25	154	225	304	154	154
Solids retention time	SRT	d	1.4	1.4	1.6	-	1.6	0.6	0.8	0.7	-	0.6
Biofilm												
Total suspended solids	TSS	mg/L	-	641 ± 5	868 ± 122	500 ± 64	1168 ± 260	351 ± 68	357 ± 62	340 ± 60	119 ± 13	-
VSS/TSS ratio	f <sub>VT</sub>	g VSS/g TSS	-	0.63 ± 0.02	0.77 ± 0.02	0.64 ± 0.03	0.63 ± 0.02	0.74 ± 0.03	0.70 ± 0.00	0.70 ± 0.02	0.69 ± 0.02	-
Effluent												
Total COD	COD	mg COD/L	313 ± 39	357 ± 58	335 ± 71	338 ± 92	307 ± 89	414 ± 55	371 ± 12	406 ± 95	389 ± 31	341 ± 56
Colloidal COD	C <sub>COD</sub>	mg COD/L	12 ± 3	8 ± 4	17 ± 5	22 ± 6	6 ± 3	14 ± 8	8 ± 4	19 ± 11	10 ± 6	21 ± 7
Soluble COD	S <sub>COD</sub>	mg COD/L	24 ± 4	29 ± 8	38 ± 8	33 ± 5	29 ± 9	40 ± 10	40 ± 10	38 ± 3	41 ± 5	40 ± 4
TSS	X <sub>TSS</sub>	mg TSS/L	295 ± 67	327 ± 67	245 ± 80	316 ± 114	320 ± 141	364 ± 51	303 ± 63	256 ± 66	302 ± 54	319 ± 54
VSS	X <sub>VSS</sub>	mg VSS/L	166 ± 25	206 ± 35	182 ± 47	184 ± 48	170 ± 52	218 ± 31	171 ± 25	185 ± 29	185 ± 27	170 ± 31
VSS/TSS ratio	f <sub>VT</sub>	g VSS/g TSS	0.57 ± 0.07	0.63 ± 0.04	0.76 ± 0.07	0.60 ± 0.09	0.57 ± 0.13	0.60 ± 0.06	0.57 ± 0.07	0.74 ± 0.10	0.62 ± 0.04	0.59 ± 0.10
X <sub>COD</sub> /VSS ratio	f <sub>CV</sub>	g X <sub>COD</sub> /g VSS	1.7 ± 0.1	1.6 ± 0.1	1.9 ± 0.1	1.5 ± 0.1	1.8 ± 0.1	1.7 ± 0.2	1.7 ± 0.2	1.9 ± 0.4	1.8 ± 0.1	1.7 ± 0.2
Alkalinity	S <sub>Alk</sub>	mg CaCO <sub>3</sub> /L	-	-	156 ± 10	-	162 ± 11	184 ± 15	-	193 ± 18	-	-
pH	-	-	7.3 ± 0.2	7.5 ± 0.2	7.4 ± 0.2	6.7 ± 0.2	7.3 ± 0.2	7.9 ± 1.0	7.5 ± 0.1	7.7 ± 0.4	8.1 ± 0.4	6.7 ± 0.1
Process performance												
CS <sub>B</sub> biotransform. efficiency	R <sub>CSB</sub>	%	85 ± 6	86 ± 9	67 ± 4	64 ± 13	78 ± 13	56 ± 5	62 ± 5	74 ± 6	53 ± 6	40 ± 11
CS <sub>B</sub> specific removal rate	SR <sub>CSB</sub>	g CS <sub>COD</sub> m <sup>-2</sup> d <sup>-1</sup>	3 ± 1	6 ± 1	10 ± 2	3 ± 1	7 ± 2	39 ± 10	35 ± 4	28 ± 4	19 ± 7	13 ± 5
X <sub>COD,eff</sub> /(CS <sub>B</sub> +X <sub>OHO</sub> ) <sub>inf</sub> .	-	g X <sub>COD</sub> /g BCOD	0.83 ± 0.15	0.86 ± 0.06	-	-	0.88 ± 0.12	1.01 ± 0.18	0.91 ± 0.12	0.95 ± 0.33	-	-



The same tendency also has been observed between HRT and  $CS_B$  bio-transformation by Brosseau et al. (2016) and Aygun et al. (2008), however the bio-transformation efficiencies were systematically different due to influent COD concentration, available surface area for biofilm growth, and HRT in their experiments.

A lower value of  $X_{COD}$  and COD removal efficiency was observed at higher HRT (36 min and 54 min) probably due to the partial release of particulate matter from biofilms caused by abrasion at a long HRT (Hoang, 2013). Minimal hydrolysis of particulate organic matter can be achieved at a low HRT as can be achieved in a HR-MBBR process (Schubert et al., 2013).

Similarly, the effect of HRT on removal and bio-transformation of COD,  $X_{COD}$ ,  $C_B$  and  $S_B$  were evaluated through the IC process at operating conditions OC1, OC2 and OC3 in Figure 4.2b and Table 4.2.

A positive correlation was observed between  $SC_B$  bio-transformation and HRT in the IC system due to the prolonged contribution of inoculum by transferring and establishing active biomass in the chemostat at higher HRT.

The  $CS_B$  bio-transformation efficiencies were  $56\% \pm 5\%$ ,  $62\% \pm 5\%$  and  $74 \pm 6\%$  at HRTs of 154 min, 225 min and 304 min, respectively, across the IC process. The concentration of COD and  $X_{COD}$  did not effectively change in the IC process at HRTs 154 and 304 min based on removal efficiency compared to HR-MBBR, due to the minimum effect of hydrolysis on particulate matter.

This phenomenon supported Confer & Logan (1998) results which found that hydrolysis rate is much more on the biofilm surface than at the surface of sloughed biofilm.

The overall efficiency of biotransformation of influent biodegradable organic matter into particulate matter across each process was also characterized by the ratio of effluent particulate COD to influent total biodegradable COD. Results are presented in Table 4.2 as  $X_{COD,eff}/(CS_B + X_{OHO})_{inf.}$ . This fraction was lower across the IC process ( $0.96 \pm 0.22$  g  $X_{COD}$ /g BCOD) than the HR-MBBR process ( $0.86 \pm 0.11$  g  $X_{COD}$ /g BCOD) suggesting that less hydrolysis of particulate organic matter took place in the first one.

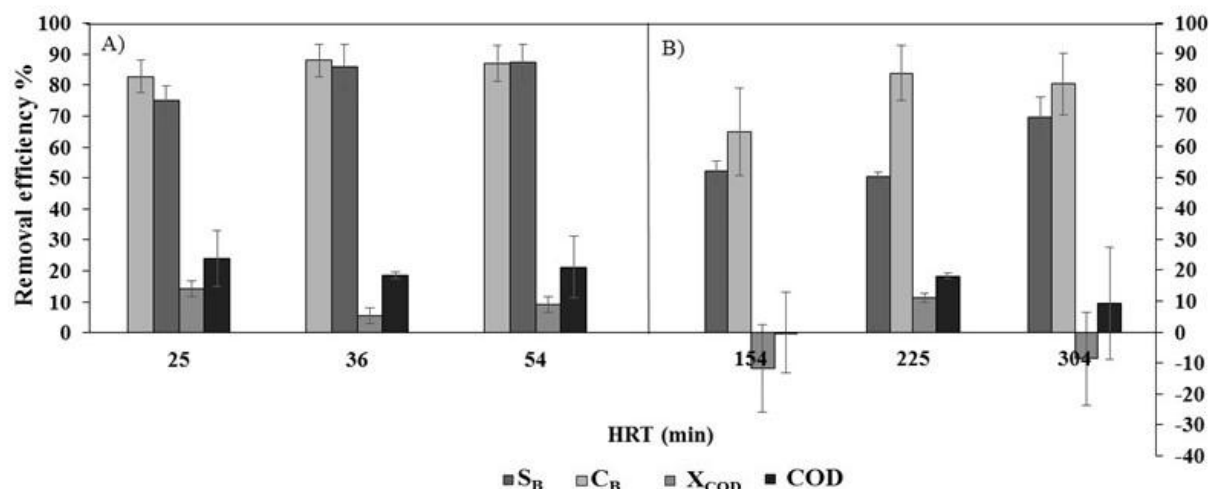


Figure 4.2: Effect of HRT on the removal efficiency of COD fractions for the A) HR-MBBR and B) IC processes

The effect of the SOLR on the removal of  $CS_B$  was also assessed in HR-MBBR and IC processes (Figure 4.3). The higher specific removal rates were attained as the SOLR was increased in both HR-MBBR and IC processes, whereas HRT and SOLR has been identified as an important constraint on the bio-transformation (especially for IC process).

A maximum  $CS_B$  bio-transformation rate ( $90 \pm 3 \%$ ) in HR-MBBR process was achieved at SOLR from 2.0 to 5.5 g  $CS_B \text{ m}^{-2} \text{ d}^{-1}$ , corresponding to an optimum HRT of 36 min. These values for IC process reached in maximum specific removal of  $80 \pm 3 \%$ , corresponding as SOLR ranged between 22 to 40 g  $CS_B \text{ COD m}^{-2} \text{ d}^{-1}$  at an optimum HRT of 225 min.

The observed linear pattern between SOLR and  $CS_B$  removal efficiency was observed with the study of Ødegaard et al. (2000), Brosseau et al. (2015) and Helness et al. (2005) in lab and pilot scale experiments with HR-MBBRs. Aygun et al. (2008) also demonstrated that by increasing the SOLR from 6 to 96 g COD  $\text{m}^2 \text{ d}^{-1}$ , the organic removal efficiency decreased from 95% to 45%.

In this context, Orantes and Gonzalez-Martinez (2003) established an asymptotic relationship between the mass of attached biofilm and SOLR, which no further biomass is attached at high SOLR. Hence, at high SOLR, less biofilm can be established through inoculum process and limited by short HRT, so less contribution of inoculum could reasonably be expected to transfer active biomass into the chemostat.

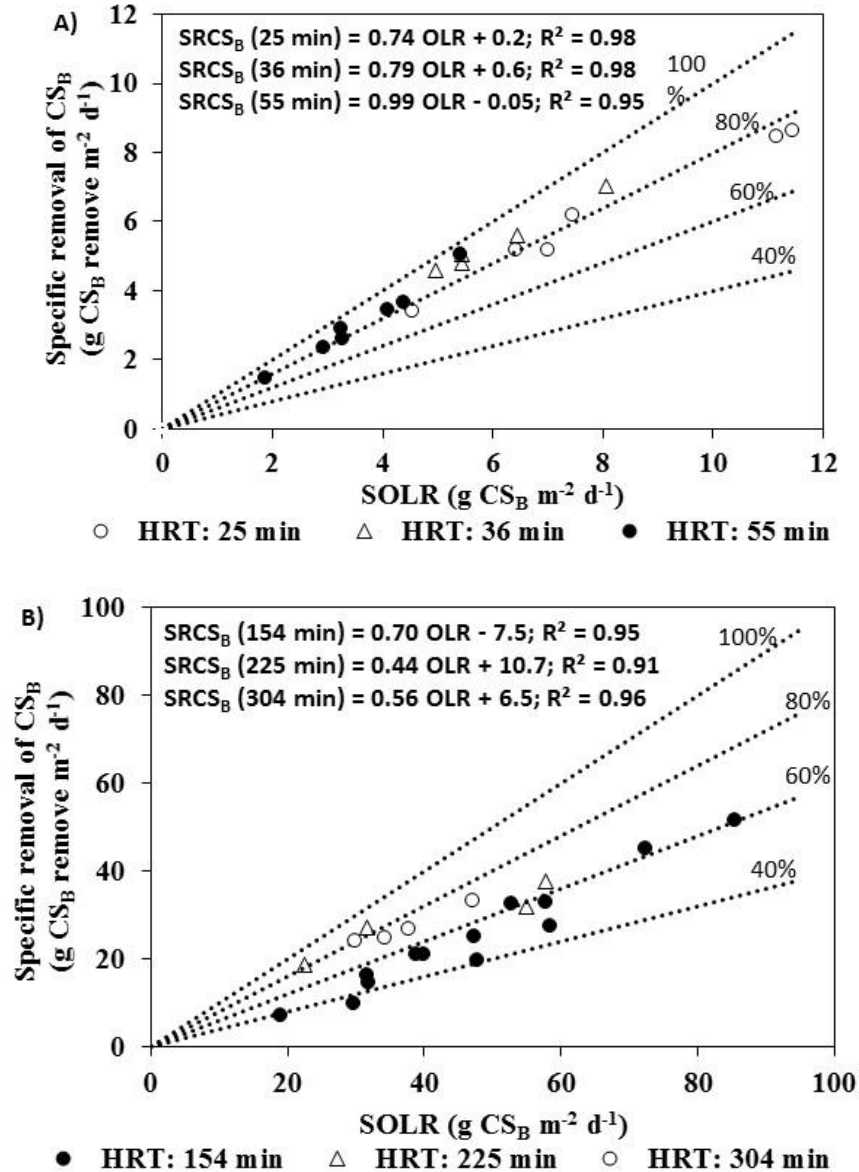


Figure 4.3: Bio-transformation of  $CS_B$  into  $X_{COD}$  as a function of  $CS_B$ -SOLR at different HRTs  
 A) HR-MBBR B) IC process

Variation of DO during aerated and non-aerated periods in the HR-MBBR and chemostat was monitored, based on the operating conditions of OC2 and OC1, respectively. The oxygen concentration dropped more rapidly in the HR-MBBR than in the chemostat when the aeration system was switched off for 3 minutes. Calculation of the oxygen uptake rate (OUR) in two reactors indicated over five-fold higher OUR in the MBBR ( $50 \pm 2\ mg\ O_2\ L^{-1}\ h^{-1}$ ) than in the chemostat ( $10 \pm 1.5\ mg\ O_2\ L^{-1}\ h^{-1}$ ). In this context, the SOUR value across HR-MBBR and chemostat process was  $55 \pm 1\ mg\ O_2\ g^{-1}\ VSS\ h^{-1}$  and  $53 \pm 6\ mg\ O_2\ g^{-1}\ VSS\ h^{-1}$ , respectively.

The highest OUR in the HR-MBBR can be correlated to the oxidation of more readily biodegradable matter produced by hydrolysis of biofilm surface (Confer & Logan, 1998) and the slowly biodegradable matter that results from lysis of decayed biomass in the HR-MBBR system, whereas the source of active biomass in the chemostat process was provided from inoculum continually with lower SRT ( $SRT_{IC-OC1}$ : 0.6 d and  $SRT_{HR-MBBR-OC2}$ : 1.4 d) and no further accumulation.

The removal efficiency of filterable biodegradable organic matter ( $CS_B$ ) by the IC and HR-MBBR processes was about 75% and 85%, respectively, which corresponded to HRT of 141 min and 36 min.

Further tests were conducted to assess the effect of oxygen concentration on the bio-transformation rate in the HR-MBBR process. For this purpose, the DO concentration was changed from 1-2 mg  $O_2/L$  to 2-4 mg  $O_2/L$  during OC4 and OC1, respectively, for a duration of one week each.

The role of the DO concentration as an effective and sensitive parameter controlling the removal of  $S_B$  and  $C_B$  fractions, but not that of particulate COD is illustrated in Figure 4.4 and Table 4.2. The maximum  $S_B$  and  $C_B$  removal efficiency,  $86 \pm 7\%$  and  $77 \pm 17\%$ , respectively, was obtained during OC1. The  $S_B$  and  $C_B$  removal efficiency was significantly decreased to  $67 \pm 20\%$  and  $53 \pm 12\%$ , respectively, as DO concentration was less than 2 mg  $O_2/L$  indicating that the DO concentration (below 2 mg  $O_2/L$ ) was a limiting factor in the HR-MBBR system. For an optimal COD removal, dissolved oxygen should be maintained higher than 2 mg  $O_2/L$  as indicated by a 13% decline in COD removal when the DO level was decreased from 2 to 1 mg/L while only a 6% increase in COD removal was observed with an increase in DO level from 2 to 6 mg/L (Wang et al., 2005).

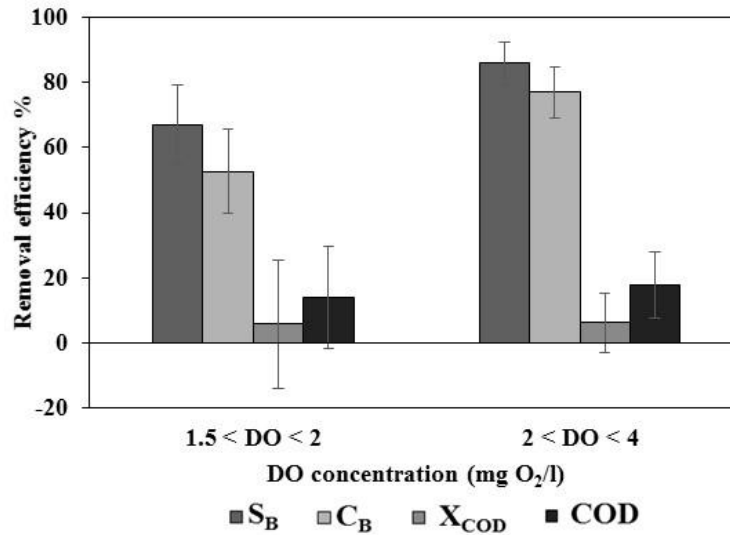


Figure 4.4: Effect of DO on the COD removal efficiency of the different COD fractions across the HR-MBBR process (OC4 and OC1)

#### 4.8 Effect of media fill volume fraction on bio-transformation of CSB

The effect of the two media fill volume fraction (OC3: 35% v/v, OC5: 50% v/v) on HR-MBBR treatment efficiency was assessed at HRT of 36 min. As the media fill volume fraction increased from 35% to 50% in the HR-MBBR, both  $S_B$  and  $C_B$  bio-transformation efficiency was increased from  $76 \pm 4$  and  $84 \pm 6$  to  $89 \pm 10$ ,  $90 \pm 8$ , respectively (Figure 4.5a and Table 4.2). Similarly Azizi et al. (2013) reported an effective treatment can be obtained by increasing media fill volume fraction up to 40% v/v, due to higher available surface area for biofilm growth. The removal of particulate and total COD decreased slightly by increasing the media fill volume fraction. Collision and attrition in the HR-MBBR reactor could lead to a biofilm detachment from the outer surface and increase the total and particulate COD in the effluent due to the high volume of media and shear forces (Ødegaard et al., 2000).

The effect of media fill volume fraction in the inoculum based on OC1, OC4 and OC5, aimed at transferring active biomass to the chemostat, demonstrated the opposite effect on bio-transformation of  $S_B$  and  $C_B$  in IC process. Increasing the media fill volume fraction from 15% v/v to 45% v/v in the inoculum decreased the bio-transformation of  $S_B$  and  $C_B$  from  $58 \pm 4\%$  to  $50 \pm 6\%$  and  $69 \pm 14\%$  to  $43 \pm 10\%$ , respectively, in the IC process (Figure 5b). The media fill volume fraction ranging from 15% to 22% in the inoculum did not significantly affect the removal of

particulate and total COD, while increasing the media fill volume fraction up to 45% significantly influenced the removal of total and particulate COD.

Higher media fill volume fraction (up to 45% v/v) may increase the development of active biomass in the inoculum (less biomass to be sloughed off the media) and may lead to less transferring of active biomass from inoculum to chemostat due to HRT constraints, therefore, less removal of  $S_B$  and  $C_B$  occurred in the chemostat.

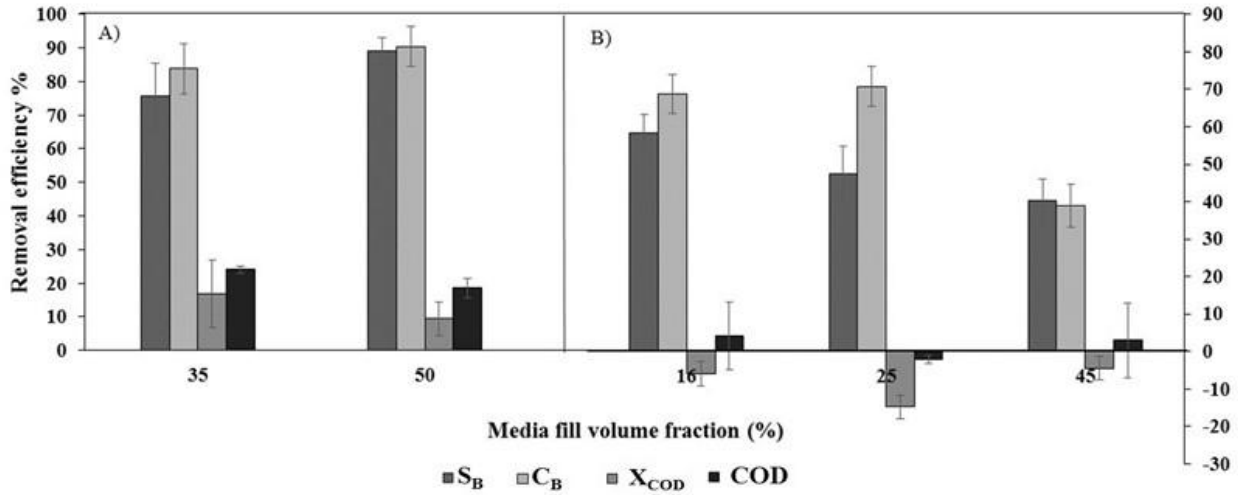


Figure 4.5: Effect of the media fill volume fraction on the COD removal efficiency of COD fractions in the A) HR-MBBR (OC3 and OC5) and B) IC (OC1, OC4 and OC5) processes

## 4.9 Effect of operating conditions on attached biofilm concentration

The attached biofilm growth concentration in the HR-MBBR was directly correlated to the SOLR (Figure 4.6).

Biofilm growth concentration in the HR-MBBR reactor reached a plateau of  $18.0 \pm 1.6$  g TSS/m<sup>2</sup> at SOLR more than  $8.0 \pm 2.7$  g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>. In this context, a 2-parameter exponential equation ( $R^2$ : 0.95) showed the best fit to the biofilm concentration data. The concentration of attached biofilm during OC1 to OC5 was increased from  $4.5 \pm 2.6$  g TSS/m<sup>2</sup> to  $18.5 \pm 1.2$  g TSS/m<sup>2</sup> by increasing the SOLR from  $3.1 \pm 0.9$  g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup> to  $15.6 \pm 2.8$  g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>, respectively.

The attached biofilm concentration in inoculum after an increase from  $6.0 \pm 0.5$  to  $13.1 \pm 0.8$  g TSS/m<sup>2</sup> reached a plateau with an average concentration of  $11.7 \pm 1.1$  g TSS/m<sup>2</sup>, while the SOLR ranged over  $39.7 \pm 13.8$  g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>. Moreover, under high SOLR in the inoculum, sloughing

phenomenon was observed frequently and elevated effluent (chemostat influent) suspended solids concentrations. Downing et al. (2013) and Bassin et al. (2016) also indicated that high SOLRs potentially enhanced biofilm detachment rates. In this context, Aygun et al. (2008) also reported a plateau occurred in biomass production level after SOLR reached  $50 \text{ g COD m}^{-2} \text{ d}^{-1}$ .

The trend of VSS/TSS ratio in attached biofilm across the HR-MBBR and inoculum ranged  $0.67 \pm 0.06$  and  $0.71 \pm 0.02 \text{ mg VSS/mg TSS}$ , respectively (Table 4.2). The VSS/TSS ratio obtained by Oliveira et al. (2014) based on pilot scale average values was  $0.69 \text{ mg VSS/mg TSS}$ , however, this value reported by Jahren et al., (2002) operated lab scale, equal to 0.91, was much higher. This may because of the fibrous materials with low VSS/TSS ratio (almost  $0.55 \text{ mg VSS/mg TSS}$ ) carried by raw wastewater and although biomass adhered to the carriers.

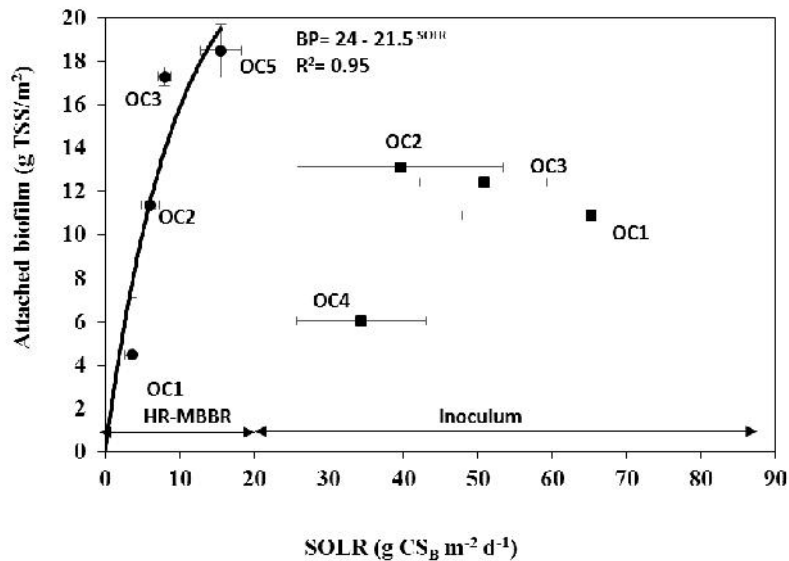


Figure 4.6: Effect of SOLR on the attached biofilm concentration for different operating conditions in the A) HR-MBBR and B) inoculum processes

#### 4.10 HR-MBBR and IC effluent

The  $f_{VT}$  (VSS/TSS) and  $f_{CV}$  ( $X_{COD}/VSS$ ) ratios in the effluent of HR-MBBR and IC processes are shown in Figure 4.7. The  $f_{VT}$  value in the HR-MBBR increased from 0.5 to  $0.8 \text{ g VSS/g TSS}$  with an increasing SOLR from 2 to  $16 \text{ g CS}_B \text{ m}^{-2} \text{ d}^{-1}$ . In the IC process, the  $f_{VT}$  value only increased from 0.60 to  $0.7 \text{ g VSS/g TSS}$  as the SOLR increased from 20 to  $90 \text{ g CS}_B \text{ m}^{-2} \text{ d}^{-1}$  despite some

fluctuations that may have resulted from detached biofilm. The values of  $f_{CV}$  in the effluent in both processes effluent was  $1.7 \pm 0.2$  g  $X_{COD}/g$  for all operating conditions.

The  $f_{VT}$  value in the effluent reported by Brosseau et al. (2016) was 0.81 to 91 g VSS/g TSS, while  $f_{CV}$  varied between 1.24 to 1.6 g  $X_{COD}/g$  for all operating conditions based on HRT and SOLR, the same ratio also was reported by Karizmeh (2012).

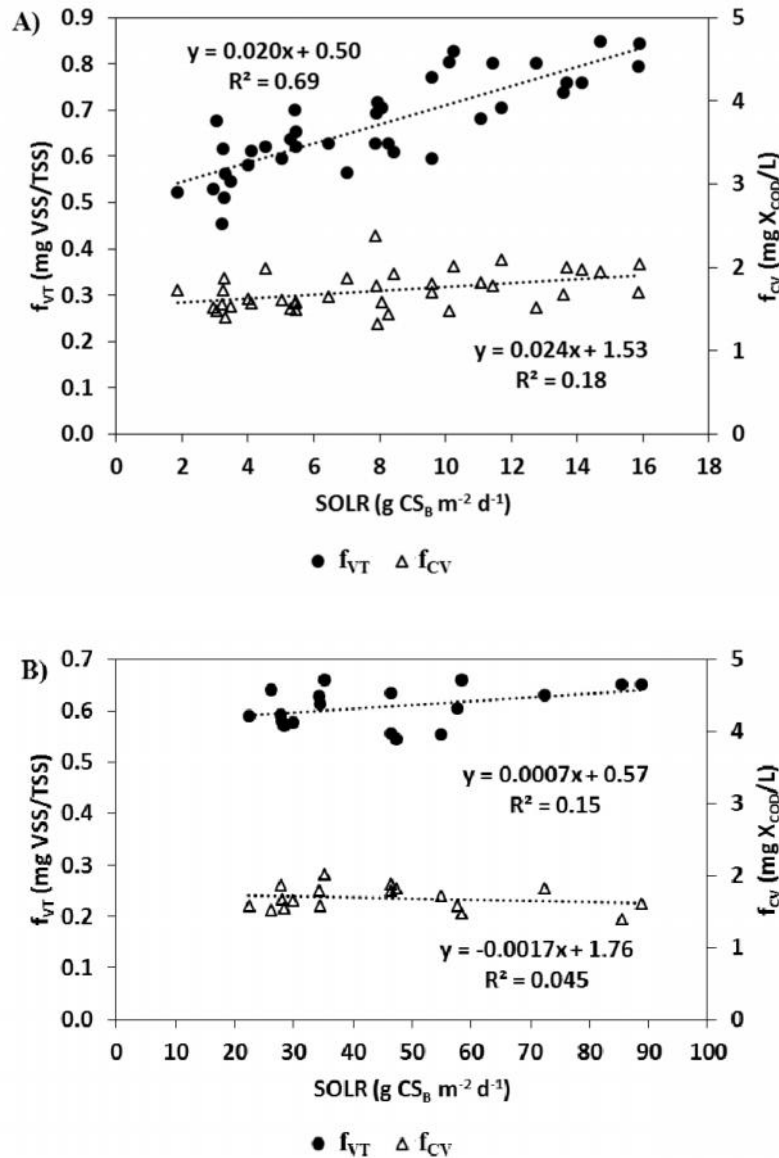


Figure 4.7: Effect of SOLR on  $f_{VT}$  and  $f_{CV}$  ratio for A) HR-MBBR and B) IC process effluents

The effect of the HRT on the effluent COD fractions was evaluated for the HR-MBBR and IC processes (Figure 4.8). The particulate matter fraction increased after the HR-MBBR or the IC



processes, with the largest proportion observed with an HRT of 36 minutes in the HR-MBBR and of 225 minutes in the IC process. The fractionation of COD showed that particulate and soluble COD were predominant in the influent and effluent of HR-MBBR and IC processes, whereas the COD contained a small portion of colloidal matter.

Particles agglomeration occurred with increasing HRT up to 36 and 225 min across the HR-MBBR and IC process, however, with increasing HRT from 36 min to 54 min in HR-MBBR, a movement from particulate toward smaller particle (colloidal) matter was observed. The same characteristics in the MBBR effluent were observed by Brosseau et al. (2016) between 37 and 40 min HRT.

Particle agglomeration also resulted from increasing the HRT from 0.75 to 4 hours (Melin et al., 2005; Åhl et al., 2006; Ødegaard et al., 2010; Karimzadeh, 2012), but Karimzadeh et al. (2014) later demonstrated that by independently decreasing HRT and SOLR, a shift toward smaller particle size was observed. Moreover, during degradation of particulate matter and formation of smaller particles more surface area of substrate is available for hydrolysis (Dimock & Morgenroth, 2006).

The average value of SVI on different operating condition was measured to evaluate the sludge settleability of each process.

Slightly better settling sludge was obtained in the IC process (SVI of  $70 \pm 11$  ml/g) than in the HR-MBBR ( $94 \pm 10$  ml/g). Better flocculating solids may have resulted from inoculum-chemostat process on which configuration most favors the proper maintenance of SVI in a higher SOLR even at lower SRT ( $SRT_{IC}: 0.6 \pm 0.1$  d and  $SRT_{HR-MBBR}: 1.5 \pm 0.1$  d). These results are supported by Y. Liu et al. (2006) which demonstrated that low organic loading rate resulted in irregular shape with poor settling characteristics and high SVI value, According to the theory, low substrate concentrations favor the growth of filamentous over floc-forming bacteria (J. Chudoba et al., 1973; Jan Chudoba, 1985).

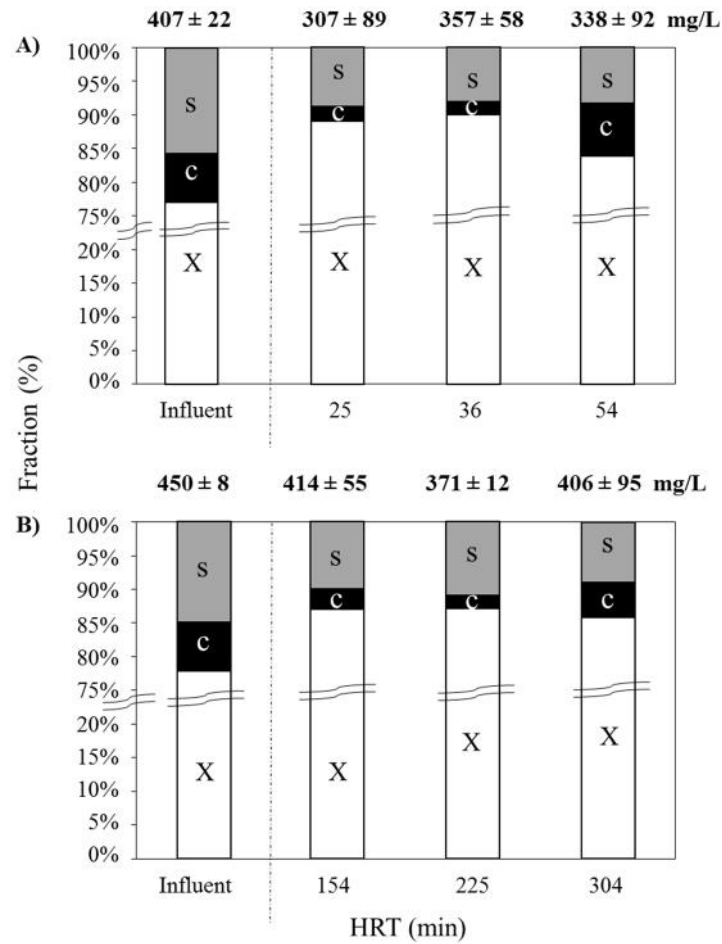


Figure 4.8: Influent and effluent mean COD fractions as a function of HRT for the A) HR-MBBR and B) IC processes

## 4.11 Aeration requirements

The maximum efficiency of HR-MBBR and IC processes bio-transformation were  $90 \pm 3 \%$  and  $77 \pm 3 \%$ , respectively, which corresponded to HRTs of 36 min and 3.7 hours, SRTs of  $1.5 \pm 0.1$  d and  $0.6 \pm 0.1$  d, and SOLR of 2.0 to 5.5 g  $CS_B$   $m^{-2} d^{-1}$  and 22 to 30 g COD  $m^{-2} d^{-1}$ .

The oxygen uptake rate (OUR) in the HR-MBBR ( $50 \text{ mg O}_2 \text{ L}^{-1} \text{ h}^{-1}$ ) was determined to be two and half times greater than in the IC process  $20 \text{ mg O}_2 \text{ L}^{-1} \text{ h}^{-1}$  (accounting for the inoculum process OUR of  $10 \text{ mg O}_2 \text{ L}^{-1} \text{ h}^{-1}$ ). The total oxygen demand was calculated to be  $0.54 \pm 0.03$  and  $0.81 \pm 0.07 \text{ kg O}_2/\text{kg } CS_B$  added for the HR-MBBR and IC processes, respectively. The blower provided  $16 \pm 1 \text{ m}^3/\text{h}$  and  $3.5 \pm 0.2 \text{ m}^3/\text{h}$  of air in the HR-MBBR and IC reactors to maintain DO level 3-4

mg O<sub>2</sub>/L and 6-7 mg O<sub>2</sub>/L, respectively. It should be noted that oxygen transfer rates and efficiency (OTE) at full scale may differ due to the shallow depth of the pilot reactors.

In a high-rate activated sludge process, Jimenez et al. (2015) reported that the maximum removal efficiency was obtained at an HRT ranging between 30 and 45 minutes, at an SRT of  $0.6 \pm 0.1$  d. They observed that the optimal removal of S<sub>B</sub> (80%) required an oxygen concentration of  $0.38 \pm 0.12$  kg O<sub>2</sub>/kg COD (based on the use of net oxygen consumption of bio-transformation).

Poor oxygen transfer efficiencies (rapidly rising bubbles) related to the coarse diffuser in the HR-MBBR can be also lead to less dissolved oxygen and excessive power requirement compared with the fine diffuser in the chemostat.

The dissolved oxygen concentration dropped below 2 mg/L in the HR-MBBR process within 5 minutes of the non-aeration period, while this value took almost 30 minutes in the chemostat. In this context, much of the energy can be saved across the IC process with DO control strategy by using programmable logic controllers (PLC) for multi loop controllers of aeration system (turn automatic switching range on/off dissolved oxygen transmitters), however, cost-effectiveness analysis need to conduct further results to fully compare the IC and HR-MBBR processes.

From an energy efficiency point of view, operating the IC process as an interesting alternative to high-rate system may lead to diminishing the consumption of energy through aeration system and also resulted in the efficient production of energy across the anaerobic digester by minimizing hydrolysis of X<sub>B</sub>.

## 4.12 Conclusion

The objective of this study was to determine the potential of an innovative high-rate inoculum-chemostat (IC) process compared with a typical HR-MBBR process for colloidal and soluble organic matter transformation into particulate matter for anaerobic digester methane production. The effect of SOLR, OUR and HRT on the removal and bio-transformation of C<sub>B</sub> and S<sub>B</sub> fractions were studied using real wastewater in a pilot scale system operated under five operating conditions. The SOLR in the HR-MBBR and IC processes were varied between 2 to 16 g m<sup>-2</sup> d<sup>-1</sup> and 20 to 90 g m<sup>-2</sup> d<sup>-1</sup>, respectively, using different HRTs.

The following conclusions were drawn:

- $CS_B$  bio-transformation into  $X_B$  in the HR-MBBR process increased with HRT (and SOLR) up to 36 min with a  $CS_B$  capture efficiency as high as  $90 \pm 3 \%$ , while in the IC process, an HRT of 3.7 hours was required for a  $CS_B$  capture of up to  $77 \pm 3 \%$  at SOLRs between 22 to  $30 \text{ g } CS_B \text{ m}^{-2} \text{ d}^{-1}$ .
- The SOUR value across the HR-MBBR and chemostat, to maintain a DO level above 2 mg  $O_2$ /), was similar in both systems ( $55 \pm 1$  and  $53 \pm 6 \text{ mg } O_2 \text{ g}^{-1} \text{ VSS h}^{-1}$ , respectively).
- A slightly better settleability of produced particulate matter, based on SVI values, was obtained in the IC process ( $70 \pm 11 \text{ mL/g}$ ) than in the HR-MBBR ( $94 \pm 10 \text{ mL/g}$ ), possibly due to better flocculating solids may have resulted from higher SOLR and lower SRT values in the IC than the HR-MBBR process

The innovative IC process can be a competitive alternative process to maximize the bio-transformation of  $CS_B$  to minimize  $X_B$  and  $X_{OHO}$  oxidation to improve the energy balance at WRRFs.

## **CHAPTER 5      ADDITIONAL RESULTS**

Additional results including results validation, hydraulic behavior, reactor stability, comparing the two proposed configurations, are presented in this chapter.

### **5.1 Results validation**

Validation of the results were performed based on several steps. As a prerequisite, the stability of the influent characteristics was evaluated by monitoring for sudden and significant changes in pH, color and temperature, according to fluctuations of influent concentrations in the WRRF. In the second validation step, the pattern of the operating conditions in the pilot system was monitored; flowrate, blower, mixer and power supply, mixed liquor DO and pH levels based on the operational conditions mentioned in section 3.3 and Table 3.1. As the mass balance determination was not possible for the COD, the preferred method for data validation was a stable condition for the removal of soluble organic matter and biodegradable colloidal across the reactors (Aygun et al., 2009; Helness et al., 2005; Schubert et al, 2013; Ødegaard and al., 2000; Karizmeh 2012).

### **5.2 Hydraulic behavior**

Good hydraulic behavior plays a crucial rule for the proper operation of a process. Tracer studies were performed to examine the hydraulic characteristics and select an appropriate hydraulic model to simulate the pilot scale HR-MBBR system (Appendix A).

A summary of the  $t_{10}$ ,  $t_{50}$ ,  $t_{90}$ , and Morrill index, calculated from the data is presented in Table 5.1. The first test showed that the HR-MBBR reactor was moderately mixed with some dead zones based on the Morrill index ( $t_a/T < 1$ ), but determination of the Morrill index in the second test indicated that a dead zone did not exist ( $t_a/T \geq 1$ ) (Argaman & Rebhun, 1964).

Table 5.1: Summary of results for the tracer tests in the HR-MBBR reactor

Parameters	Symbol	Unit	values		Formula
			Test 1	Test 2	
Theoretical retention time	T	min	40	40	$V/Q$
Time representing 10% of total tracer amount passage	$t_{10}$	h	0.07	0.12	-
Median time corresponding to 50% of tracer passage	$T_M$	min	0.72	0.75	-
Time representing 90% of total tracer amount passage	$T_{90}$	min	2.0	2.0	-
Time of tracer appearance in the effluent	$t_a$	min	20	45	-
Median retention time	$T_h$	min	22	30	-
Time for the initial observation of the draw at the outlet	$t_i$	min	0.3	0.42	-
Morril dispersion index	MDI	-	28.5	16.7	$t_{90}/t_{10}$
Volumetric efficiency	Ev	-	3.5	5.9	$100/MDI$
Displacement efficiency	DE	-	0.5	1.1	$t_a/T$
Efficiency factor	n	-	1.02	1.01	$t_a/(t_a - t_M)$
Index of model detention time			0.02	0.02	$T_M/T$
$M_{DI}^{dex} = 1$ Indicate plug flow ideal basin; MDI $\rightarrow$ complete mixed reactor. $t_a/T$ Less than 1, no dead zone. $n \rightarrow$ basin ideal. $n = 8$ , very good efficiency; good for $n = 3$ , bad for $n = 2$ very bad for $n = 1$ A high value of $t_M/T$ indicates plug flow.					

### 5.3 Reactor stability

The IC and HR-MBBR stability was determined by treatment efficiency and effluent characteristics of parameters including CS, C, S,  $X_{COD}$ , TSS and VSS in both the HR-MBBR and IC processes (Tables 5.2 & 5.3).

Each operating condition was conducted during 2 weeks including a growth and stabilization period of 1 week followed by a characterization period of 2 weeks.

Results indicated some stability for the processes except for the particulate matters, according to variation  $X_{COD}$  in the influent. The effluent from the IC process was relatively stable regardless of the operating conditions. All reactor characterization results are presented in detail in Appendix A.

Table 5.2: Summarized results for influent and effluent characterization of the HR-MBBR process

HRT (min)	DO	Media %	Date	#	Influent (mg/L)							Effluent (mg/L)						
					TSS	VSS	COD	CS	S	X	C	TSS	VSS	COD	X	S	C	CS
54	2-4	50	OC1	7	314 ± 68	183 ± 32	383 ± 51	81 ± 21	56 ± 13	302 ± 36	24 ± 12	294 ± 66	165 ± 25	313 ± 39	277 ± 37	24 ± 4	12 ± 3	36 ± 4
36	3-4	50	OC2	6	323 ± 68	204 ± 36	455 ± 90	94 ± 18	67 ± 15	360 ± 90	26 ± 14	326 ± 67	205 ± 35	356 ± 58	320 ± 54	28 ± 8	8 ± 4	36 ± 8
25	3-4	35	OC3	7	262 ± 68	187 ± 38	409 ± 59	106 ± 14	69 ± 10	302 ± 56	37 ± 9	244 ± 80	182 ± 47	334 ± 71	280 ± 67	38 ± 8	16 ± 5	54 ± 7
54	1.5-2	50	OC4	8	361 ± 90	189 ± 30	399 ± 81	90 ± 22	59 ± 8	308 ± 68	31 ± 15	316 ± 57	183 ± 30	338 ± 52	283 ± 84	32 ± 5	22 ± 4	55 ± 7
25	3-4	50	OC5	6	406 ± 58	221 ± 25	454 ± 22	84 ± 17	68 ± 8	370 ± 27	16 ± 9	405 ± 59	203 ± 14	362 ± 20	323 ± 17	33 ± 4	5 ± 3	39 ± 5

Table 5.3: Summarized results for Influent and effluent characterization of IC process

HRT (min)		DO	Media %	Date	#	Influent (mg/L)							Effluent (mg/L)						
						TSS	VSS	COD	CS	S	X	C	TSS	VSS	COD	X	S	C	S
13	141	6 ± 1	13	OC1	7	337 ± 69	204 ± 32	410 ± 37	92 ± 19	64 ± 14	348 ± 79	27 ± 13	364 ± 51	218 ± 31	414 ± 55	360 ± 55	40 ± 10	14 ± 8	54 ± 10
16	209		16	OC2	6	317 ± 65	177 ± 21	390 ± 29	82 ± 22	61 ± 14	308 ± 67	21 ± 15	303 ± 63	171 ± 25	333 ± 61	290 ± 55	35 ±112	8 ± 5	43 ± 13
22	282		16	OC3	6	260 ± 45	188 ± 17	455 ± 39	125 ± 15	73 ± 7	330 ± 42	52 ± 13	256 ± 66	185 ± 29	406 ± 95	349 ± 78	38 ± 3	19 ± 8	57 ± 12
13	141		25	OC4	5	285 ± 57	171 ± 29	366 ± 42	80 ± 24	56 ± 13	286 ± 27	24 ± 14	300 ± 47	180 ± 26	444 ± 59	395 ± 76	39 ± 7	11 ± 6	49 ± 7
13	141		45	OC5	8	293 ± 57	185 ± 36	369 ± 58	85 ± 19	56 ± 11	284 ± 57	29 ± 12	268 ± 58	170 ± 46	344 ± 77	281 ± 77	41 ± 4	22 ± 9	62 ± 11

## CHAPTER 6 GENERAL DISCUSSION

### 6.1 Influent characteristics

The pilot plant influent characteristics (Appendix A) showed that the wastewater was moderately concentrated with significant variations during the day. The  $f_{vt}$  (VSS/TSS) and  $f_{cv}$  ( $X_{COD}$ /TSS) ratios were  $0.65 \pm 0.10$  and  $1.7 \pm 0.2$ , respectively. The soluble fraction of the COD (40 to 70 mg/L) was between 15 and 20% of the total COD and the CS fraction (60 to 120 mg/L) represented 20 to 30% of the total COD, respectively.

Typical reference data for based on total COD are shown in Tables 6-1. In this context,  $CS_U$ , S and  $X_{COD}$  fraction represent about 5-12%, 9-30% and 57-75% of total COD (adapted from Ekama et al., 1986; Henze et al., 1987; Henze et al., 1992; Henze et al., 1987; Orhon et al., 1996; Dulekgurgen et al., 2006).

Moreover, the BioWin software (EnviroSim, 2014) also reported typical ratios of municipal raw wastewater S/COD, CS/COD,  $f_{VT}$  and  $f_{CV}$  of 0.21, 0.38, 0.81 and 1.6, respectively.

Table 6-1 : COD fractionation of domestic wastewaters

Location	$CS_U$ (%)	CS (%)	X (%)	Reference
South Africa	5	20	75	Ekama et al. (1986)
Hungary	9	29	62	Henze et al. (1987)
Denmark	2	20	78	Henze et al. (1992)
Switzerland	11	32	57	Henze et al. (1987)
Turkey I	4	9	87	Orhon et al. (1996)
Turkey II	35		65	Dulekgurgen et al. (2006)
Repentigny, QC	$8 \pm 2$	$20 \pm 5$	$74 \pm 7$	This study

The specific loading rate during pilot operation varied between 2 and 20 g  $CS_B m^{-2} d^{-1}$  (1000 to 9000 g  $CS_B/d$ ) and 19 to 87 g  $CS_B m^{-2} d^{-1}$  (1000 to 4000 g  $CS_B/d$ ) in the HR-MBBR and IC processes, respectively. Loading rates of 4 to 100 g  $CS_B m^{-2} d^{-1}$  (Helness et al., 2005) and 1 to 85 g  $CS_B m^{-2} d^{-1}$  (Ødegaard, 2000) have been reported in literature.



A summary of data obtained during different operating conditions in this study is presented and compared with the data from the literature are presented in Table 6.2.

Table 6-2 : Comparison of operating and performance results with literature data

Parameter	Units	This work	Literature	
		Value	Value	Reference
Dissolved oxygen (DO)	mg O <sub>2</sub> /L	2 - 5	2 - 3	Ødegaard, 2006
			> 4	Émile, 2014
Temperature coefficient ( )		1.01 – 1.04	1.07	M&EA, 2014
Media filling ratio	% v/v	35 and 50	60	M&EA, 2014
			50	Émile, 2014
Organic loading rate (OLR)	g CS <sub>B</sub> m <sup>-2</sup> d <sup>-1</sup>	2 - 16	10 - 80	Émile, 2014
			4 - 100	Helness et al., 2005
			1 - 85	Ødegaard, 2000
Minimum recommended HRT	min	28	30	M&EA, 2014
			25	Émile, 2014
CS <sub>B</sub> Removal efficiency at minimum recommended HRT	%	80	70	M&EA, 2014
			70	Émile, 2014
HRT <sub>optimum</sub>	min	40 - 60	40 - 60	M&EA, 2014
			40 – 60	Émile, 2014
Maximum filtered COD removal rate	g CS <sub>B</sub> m <sup>-2</sup> d <sup>-1</sup>	10	15	Helness et al., 2005
			12	Ødegaard, 2000
			27	Émile, 2014
Observed yield (Y <sub>obs</sub> )	g VSS/g COD	-	0,38	Émile, 2014
			0.3 – 0.45	Van Haandel et al., 2012
Biofilm density	g TSS/m <sup>2</sup>	6 - 20	28	M&EA, 2014
Mixed liquor concentration	mg TSS/L	3000 - 8000	3870 - 8400	M&EA, 2014
Note : M&EA (Metcalf & Eddy-Aecom, 2014)				

## 6.2 Biotransformation efficiency

CS<sub>B</sub> biotransformation efficiency generally increased with an increase in HRT up to a certain limit for both tested configurations (Figures 2), however, it decreased by increasing the loading rate (Figure 3). The observed trend was compatible with previously reported laboratory and pilot studies (Brosseau, 2015, Helness et al., 2005 & Aygun et al., 2009). However, the DO deficiency led to a significant decrease of bio-transformation in the HR-MBBR process. Indeed, providing an adequate DO concentration, especially in the HR-MBBR, was found to be critical to obtain an appropriate efficiency. An optimum DO concentration 2 to 3 mg O<sub>2</sub> L<sup>-1</sup> was recommended (Ødegaard, 2006) but it should be increased up to 4 to 6 mg O<sub>2</sub>/L if nitrification is also needed (Metcalf and Eddy-Aecom, 2014). Furthermore, a high oxygen uptake rate (OUR) in the HR-MBBR (50 mg O<sub>2</sub> L<sup>-1</sup> h<sup>-1</sup>) is indicative of particulate COD hydrolysis and of organic matter oxidation, as catalyzed by a high concentration of biomass in the reactor.

An optimum HRT value of 36 min was determined to achieve the highest removal and biotransformation of CS<sub>B</sub> in the HR-MBBR process, which corresponded to a removal efficiency of near  $90 \pm 3 \%$ , when the OLR was between 2.0 and 5.5 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>. The maximum removal ( $80 \pm 3 \%$ ) was achieved in the IC process with an OLR of 22 to 40 g CS<sub>B</sub> m<sup>-2</sup> d<sup>-1</sup>, which corresponded to HRT of 3.7 h. The observed pattern agreed with related studies (Helness et al., 2005 & Aygun et al., 2009). Aygun et al., 2009 and was also demonstrated that by increasing the OLR from 6 to 96 g COD m<sup>2</sup>.d<sup>-1</sup>, the Organic removal efficiency decreased from 95.1% to 45.2%.

## CHAPTER 7 CONCLUSIONS AND RECOMMENDATIONS

### 7.1 Conclusions

The main objective of this project was to maximize the biotransformation of influent soluble and colloidal biodegradable matter into particulate matter to be recovered by a physico-chemical process and sent to anaerobic digestion for maximum energy production. Thus, the biotransformation process had to minimize the oxidation of biodegradable organic matter. For this purpose, two parallel treatment processes were compared during a pilot test with real wastewater from the Repentigny WRRF.

The first process configuration consisted of an aerated high-rate MBBR (HR-MBBR) reactor and the second one was an inoculum (a very high rate MBBR) followed by an activated sludge chemostat reactor (IC).

Various operating conditions were tested for HRT, media fill volume fraction, DO level and OLR.

The following conclusions were.

1. The HR-MBBR reactor achieved 90% capture efficiency of the colloidal and soluble biodegradable organic matter ( $CS_B$ ) at a filling ratio of 50% v/v and an HRT of 36 min. At a fill volume fraction of 35 % v/v the removal efficiency decreased to about 80% at the same HRT.
2. The IC process achieved  $77 \pm 3$  %  $CS_B$  capture at an  $HRT_{inoc}$  of 12 min and a fill volume fraction of 15% v/v in the inoculum followed by an  $HRT_{Chemo}$  of 140 min in the chemostat.
3. A DO concentration above 3 mg  $O_2$ /L, corresponding to  $16 \pm 1$  m<sup>3</sup>/h air per volume of reactors, was required to reach the maximum  $CS_B$  bio-transformation in the HR-MBBR reactor, due to the high amount of active biomass. The air flowrate in the chemostat reactor was  $1.5 \pm 0.2$  m<sup>3</sup>/h to achieve a DO level of 5 mg  $O_2$ /L.
4. In the IC system, the maximum capture of  $CS_B$  and biotransformation efficiency reactor was obtained at an inoculum reactor media filling ratio of 15% v/v with 20-26 min HRT and  $1.5 \pm 0.2$  m<sup>3</sup>/h of air, with a chemostat HRT of 300 min.

## 7.2 Recommendations

To improve the study results and achievements, the following recommendations are proposed:

1. Obtain further detailed investigation, such as a respirometry test to determine the effect of the inoculum process on transferring active biomass from inoculum into chemostat.
2. Operate settler at the end of process and prepare composite samples over 24 hours from influent and effluent of the process for characterization of observed yield.
3. Evaluate the biofilm mass, at least 3 times a week, or at each sampling time.
4. Perform additional tests to evaluate the effect of DO levels and aeration control (On/Off) on  $CS_B$  removal efficiency across the IC process.
5. Perform additional tests to optimize the aeration in the HR-MBBR and the effect of biofilm thickness on oxygen consumption.
6. Characterize mixer performance through chemical tracer tests throughout the reactors to obtain optimum mixing values for more energy saving.
7. Improve the energy efficiency through optimization of HRT, media fill volume fraction, carrier type (i.e. Biofilm Chip M) and DO to enhance energy recovery with proposed innovative approach of IC process.
8. Conduct a detailed energy audit on both inoculum- chemostat and HR-MBBR processes.
9. Analyse the  $CS_B$  bio-transformation efficiency data from HR-MBBR and IC processes during low temperature (temperature  $\geq 10^\circ \text{C}$ ).
10. Determine the current WRRF's energy usage and benchmark this to the proposed process in terms of energy usage and cost for the entire facility and for each of the major power demands in the WRRF.

Evaluate the potential approaches for the installation of the inoculum-chemostat processes in a full-scale system for performing different case studies.

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## APPENDIX A – RAW WASTEWATER CHARACTERISTICS

Table A1: Repentigny WRRF raw wastewater characteristics

Parameter	Symbole	Units	Average	Number of samples
Total COD	COD	mg COD/L	$346 \pm 22$	44
Particulate COD	X <sub>COD</sub>	mg COD/L	$254 \pm 50$	44
Colloidal DCO	C <sub>COD</sub>	mg COD/L	$16 \pm 10$	44
Soluble DCO	S <sub>COD</sub>	mg COD/L	$80 \pm 31$	44
TSS	TSS	mg TSS/L	$255 \pm 104$	44
VSS	VSS	mg VSS/L	$154 \pm 49$	44
Azote total Kjeldahl	TKN	mg N/L	$37 \pm \text{n.d.}^*$	5
Total ammonium	S <sub>NH4</sub>	mg N/L	$30 \pm \text{n.d.}$	5
Oxidized nitrogen	S <sub>NOx</sub>	mg N/L	$<0.05 \pm \text{n.d.}$	5
Total phosphorus	TP	mg P/L	$4.5 \pm \text{n.d.}$	5
Orthophosphates	S <sub>PO4</sub>	mg P/L	$1.7 \pm \text{n.d.}$	5
Turbidity	-	NTU	$69 \pm 7$	10
Alkalinity	S <sub>Alk</sub>	mg CaCO <sub>3</sub> /L	$230 \pm 20$	5
pH	-	-	$7.5 \pm 0,5$	44

\*n.d: not detected

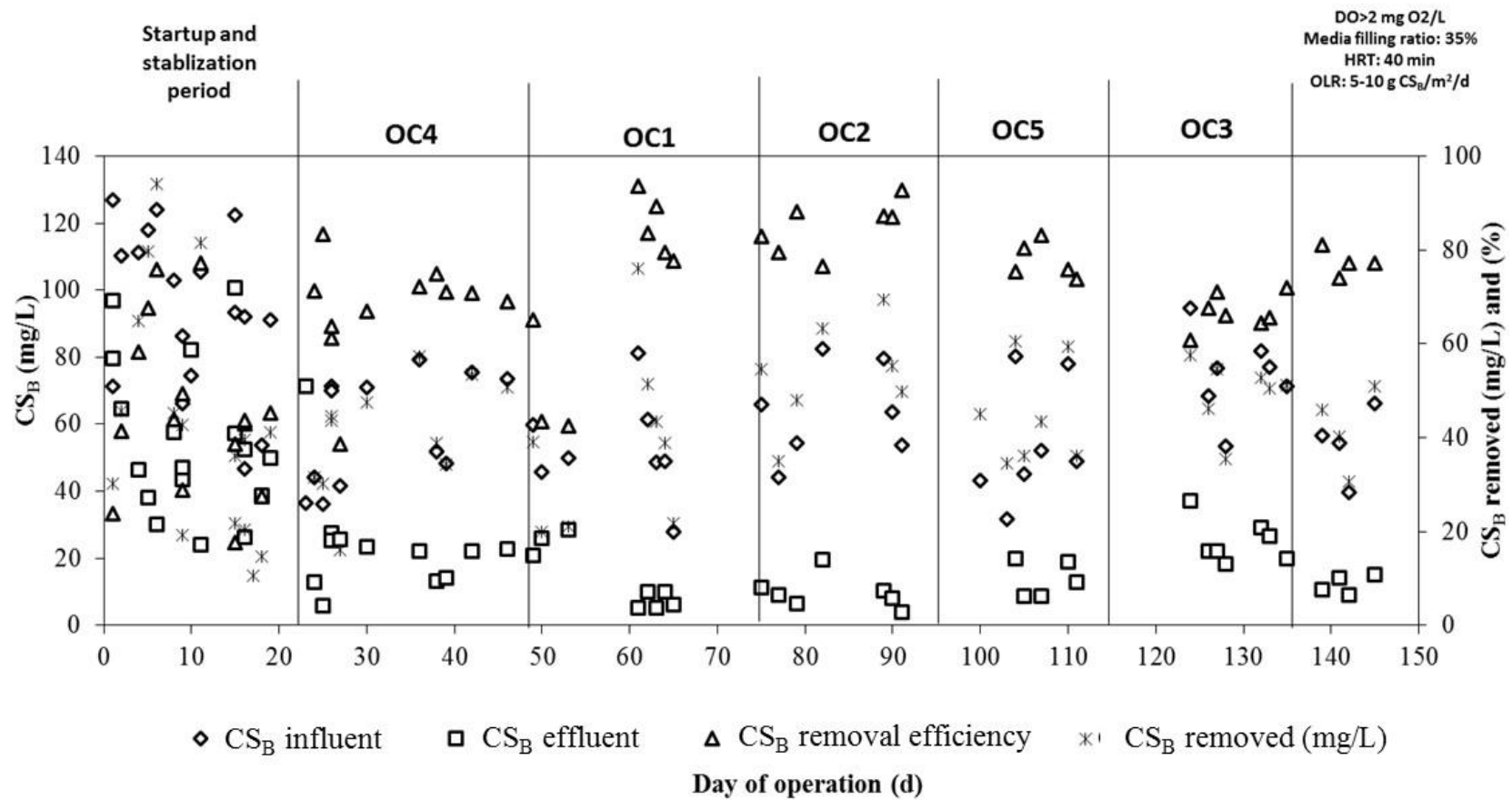


Figure A1: Variation of the influent CS<sub>B</sub> concentration and its removal efficiency at different operational condition across the HR-MBBR process



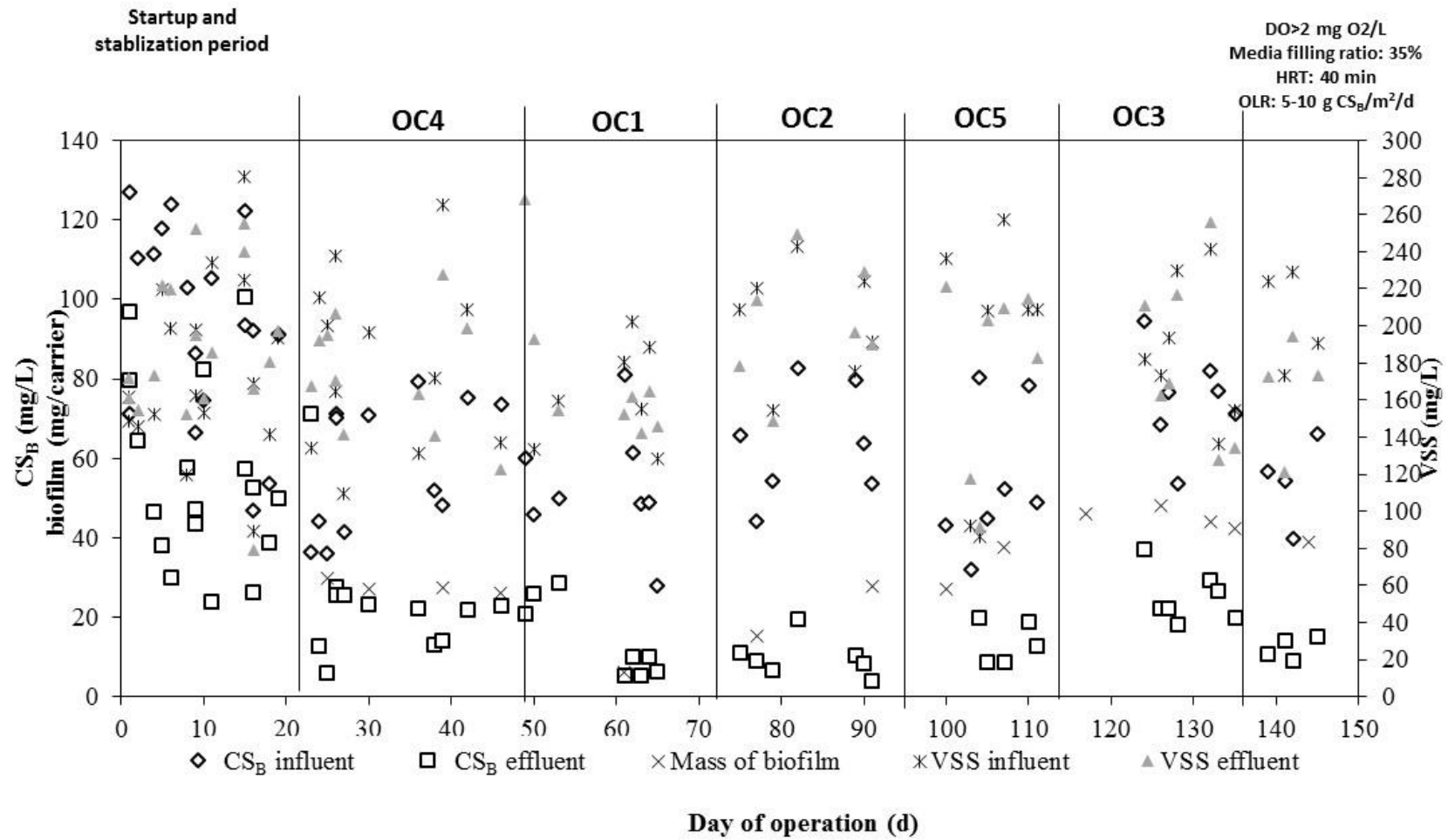


Figure A2: The variation of the influent CS<sub>B</sub> concentration, developed biomass, influent and effluent VSS concentration at different operational condition across the HR-MBBR process

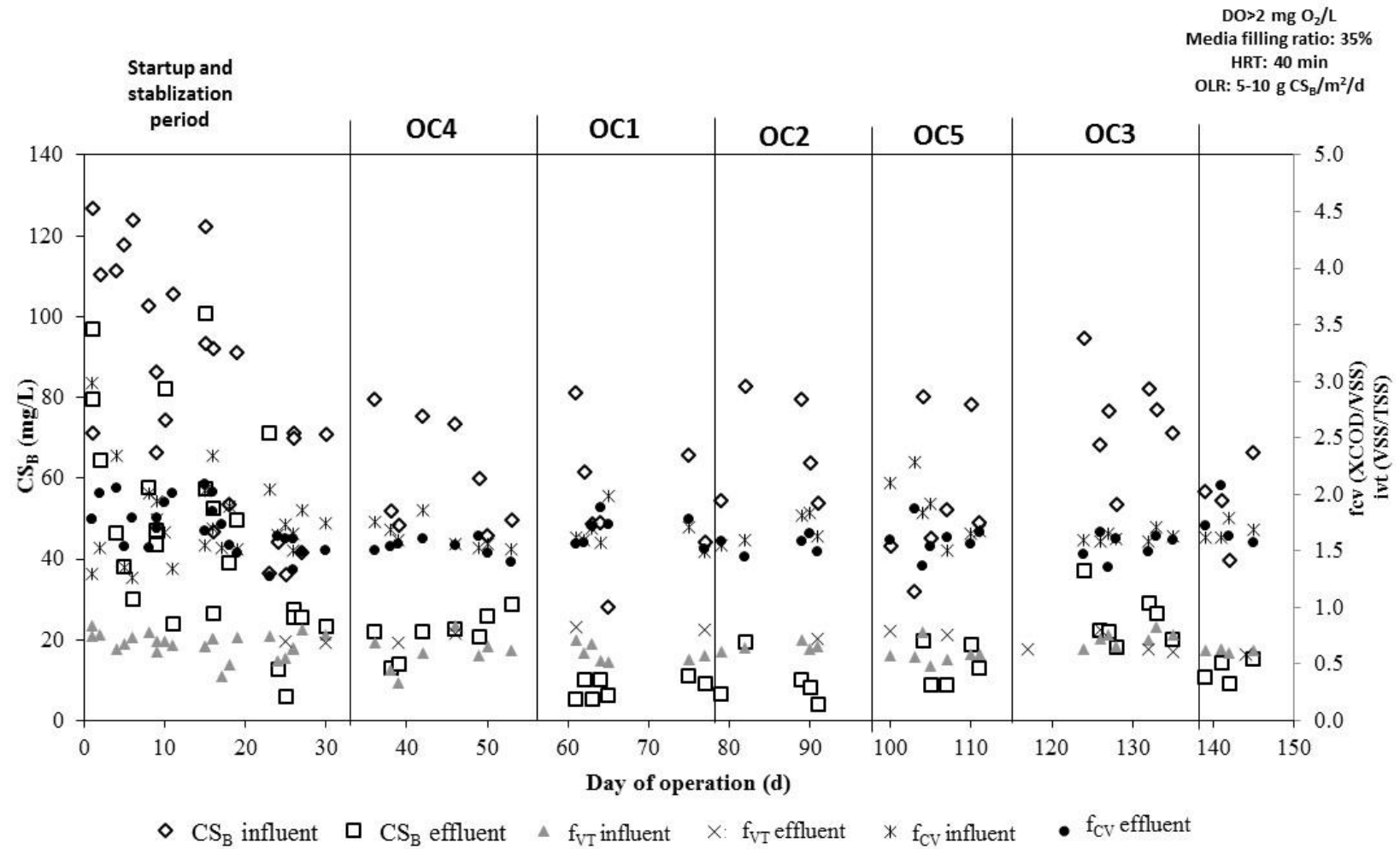


Figure A3: The variation of the influent CS<sub>B</sub> concentration, influent and effluent f<sub>VT</sub> and f<sub>CV</sub> at different operational condition across

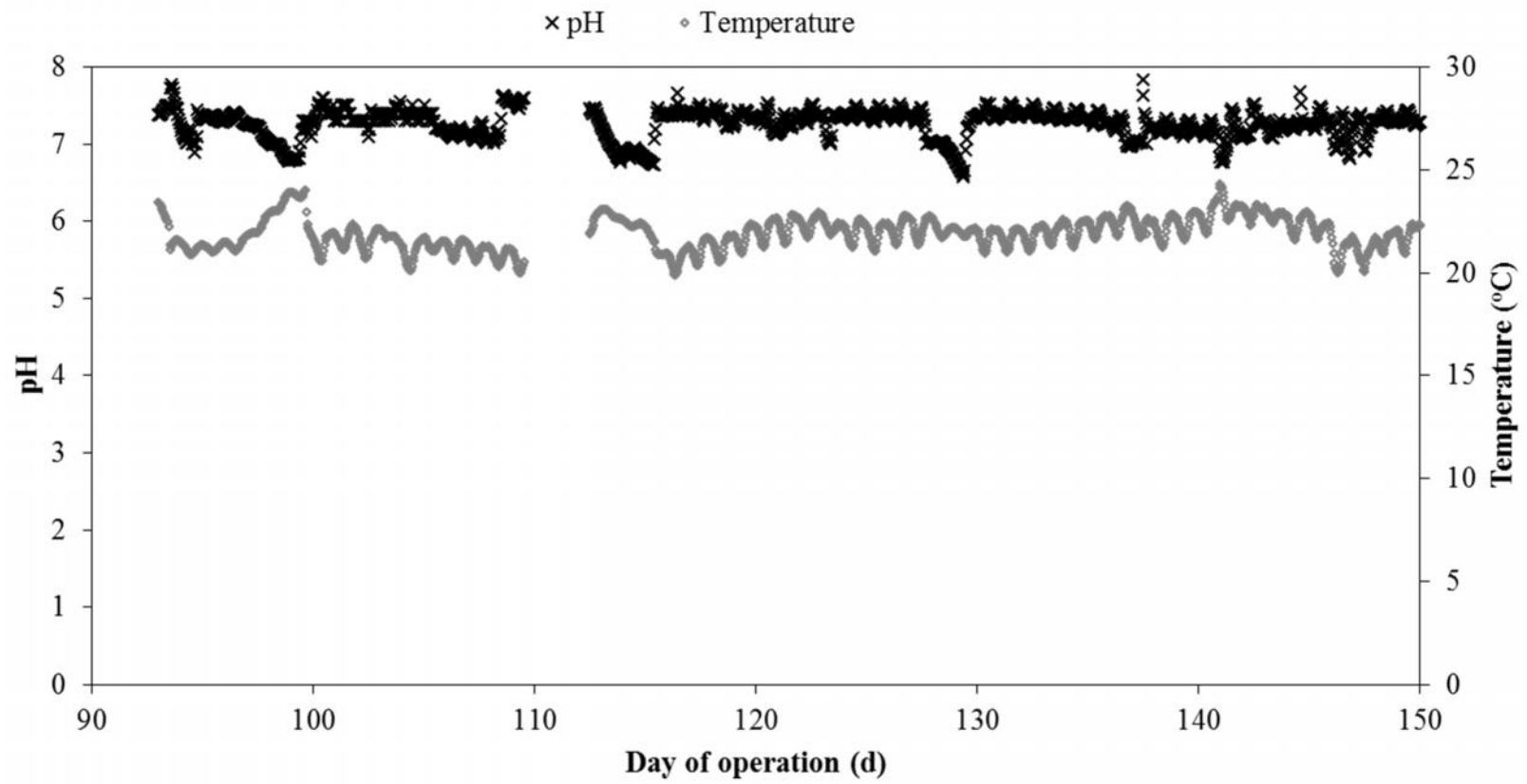


Figure A4 : Variation of pH and the temperature during the pilot period operating conditions

## **APPENDIX B – LAB PROTOCOLS FOR CHEMICAL ANALYSES**

### **1. Flocculated, filtered COD of sludge and wastewater**

#### **Objectives**

This method of flocculation and filtration removes colloidal and particulate fractions of a sample of wastewater or mixed liquor to keep only the soluble fractions. It is made typically to determine rapidly biodegradable COD (RBCOD; Mamais et al., 1992) and the soluble inert COD following a respirometric test at high load (Method zinc sulphate; Wentzel et al., 1995; 1999) or to prepare the dilution water to measure the active biomass in a sample of mixed liquor (method using aluminum sulphate; Cronje et al., 2002).

#### **Zinc sulphate method**

##### **Equipment:**

- Zinc sulphate solution ( $\text{ZnSO}_4$ ) to 100 g/L,
- 150 ml beaker (100 ml sample)
- pH meter calibrated with pH 7 and pH 10,
- 6 M NaOH,
- Syringe filter 0.45  $\mu\text{m}$ ,
- Syringe,
- Stirrer plate and a magnetic bar.

##### **Preparation of the solution of $\text{ZnSO}_4$ :**

- The  $\text{ZnSO}_4$  is sold commercially as heptahydrate.
- Molecular weight  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ : 287.53 g/mol
- $\text{ZnSO}_4$  Molar mass: 161.47 g/mol

For 1000 ml:

- Dissolve 178 g of zinc sulphate heptahydrate in a 1000 mL flask filled half of Milli-Q water.
- Make up to gauge with Milli-Q water.

**Procedure:**

- Add 1 ml of the solution to 100 g/L of  $\text{ZnSO}_4$  per 100 ml sample,
- Shake vigorously on the plate for about 1 minute,
- Adjust the pH to 10.5-11 with 6 M NaOH,
- Allow to settle for 5 minutes,
- Collect the supernatant with a syringe,
- Filter the supernatant sample, discarding the first 5 ml.

**2. Chemical oxygen demand (COD) and total soluble**

1. Identify the tubes for each sample on the lab sheet. DO NOT USE IF TUBE scratched. Analyses are done in triplicate, and should not vary by over 5%. Turn the digestion furnaces.
2. Turn the digestion oven by pressing the button on the back of the oven and wait to see the temperatures displayed on the screen. Press "START" to preheated to 150 ° C.
3. Make the sample dilutions as indicated on the lab sheet.
4. If an automatic pipette was used, check it using the method before using it to collect your samples.
5. Put your sample into a beaker with a magnetic stirrer and placed on the stir plate. Take the pipette, halfway between the vortex, 2.0 ml of sample (diluted if necessary) thoroughly and add the COD tubes (0-150 or 0-1500 as shown in the lab sheet). For inhomogeneous samples you must pass the blender before removing it.
6. In each furnace, there must be two white. Whites contain 2.0 ml preferably milli-Q water and the digestion solution. In addition, you must insert into your sequence 3 standard of 500 mg/L or 50 mg/L depending on the range of work.
7. Tighten the cap and vortex 10 seconds and put the tubes in the preheated oven digestion. Press the "Start" button to start the digestion of 2 hours at 150 ° C. SCREW THE VORTEX WELL BEFORE CAP.
8. After 2 hours at 150 ° C, turn off the oven and vortex tubes 10 seconds before putting them in test tubes to support (wear gloves during this step). Allow to cool and decant the tube before reading spectrophotometer for 30 minutes.

9. Turn on the HACH spectrophotometer and allow to heat 15 minutes

***BEFORE YOU READ A SAMPLE, MAKE SURE IT IS AT ROOM TEMPERATURE AND THAT IS PRECIPITATE decanted AND THAT THE TUBE IS CLEAN (Kimwipe)***

10. Select the appropriate program in the spectro.
11. Wipe the first white to play with a Kimwipe taking care not to re-suspend the particles, insert it into the spectro with the HACH logo facing you.
12. Press "zero."
13. Measure the second white, if it has a negative value, press "zero" (becomes zero) and read the first white (and note its value on the lab sheet). If the second white has a positive value, the note on the lab sheet.
14. Read COD samples. 9 and 10 but by pressing the "Sample" button. Note the COD values on the lab sheet.

### 3. Determination of the wastewater soluble unbiodegradable COD (SU)

#### Background

Organic matter in municipal wastewaters (WW) is quantified by measuring the chemical oxygen demand (COD). Total COD can be divided based on biodegradability into biodegradable ( $COD_B$ ), unbiodegradable ( $COD_U$ ) and Active biomass ( $X_{BH}$ ,  $X_{BA}$ ) fractions (Figure 1). These three fractions as shown in Figure 2 can be further subdivided based on their size into particulate biodegradable ( $X_B$ ), particulate unbiodegradable ( $X_U$ ), colloidal ( $C_B$ ), soluble biodegradable ( $S_B$ ) and soluble unbiodegradable ( $S_U$ ) COD (Henze, 2000; Corominas et al. 2003; Melcer et al., 2003; Lee et al., 2006). The unbiodegradable colloidal COD is not considered here as it would represent a small fraction for typical municipal wastewaters.

This protocol describes a methodology to determine the soluble unbiodegradable COD ( $S_U$ ) in a pretreated (after the trash and grit removal) municipal *wastewater*.

#### Method

$S_U$  can be determined by following these 3 steps:

##### Step 1: Flocculation/Filtration COD Method

*Material required:*

- Zinc sulfate ( $ZnSO_4$ ) solution as 100 gr/L
- 150 ml beaker
- pH meter (calibrate in high pH 7 and 10)
- Sodium hydroxide solution as 6M
- Syringe filter 0.45  $\mu m$
- Magnetic stirrer

*Steps to measuring ffCOD with zinc sulfate solution ( $ZnSO_4$ )*

1. Prepare 1000 ml of 100 gr/L  $ZnSO_4$  solution ( $ZnSO_4 \cdot 7H_2O$ : 287.5 g/Mol;  $ZnSO_4$ : 161.5 g/Mol):
  - 1.1 Dissolve 178,0 g of zinc sulfate powder in a 1000 ml of flask filled with 500 ml of demineralized water (DM);
  - 1.2 Fill up to 1000 ml with DM

2. Add 1 ml of 100 g/L zinc sulfate solution (as 100 g/L concentrations) in 100 ml of wastewater sample;
3. Mix vigorously during 1 minute;
4. Adjust the pH to 10.5-11 by using a 2 M sodium hydroxide;
5. Let settle for a 20 minutes period;
6. Carefully withdraw the supernatant using a syringe;
7. Filter the supernatant using a 0.45  $\mu\text{m}$  syringe filter;
8. Analyze the filtrate COD as flocculated filtered COD ( $ffCOD$ ).

### Step 2: Aerobic Batch test

The batch reactor test under aerobic condition is conducted for degradation of the biodegradable fractions (soluble) of filtrate solution from previous step based on operating at long SRT (i.e. 20 days) to provide complete degradation of soluble biodegradable portion. The change in COD concentration with time is monitored during batch test operation while decreasing until COD reach in a steady state value ( $ffCOD$ ).

By considering these two steps  $S_U$  is determined:

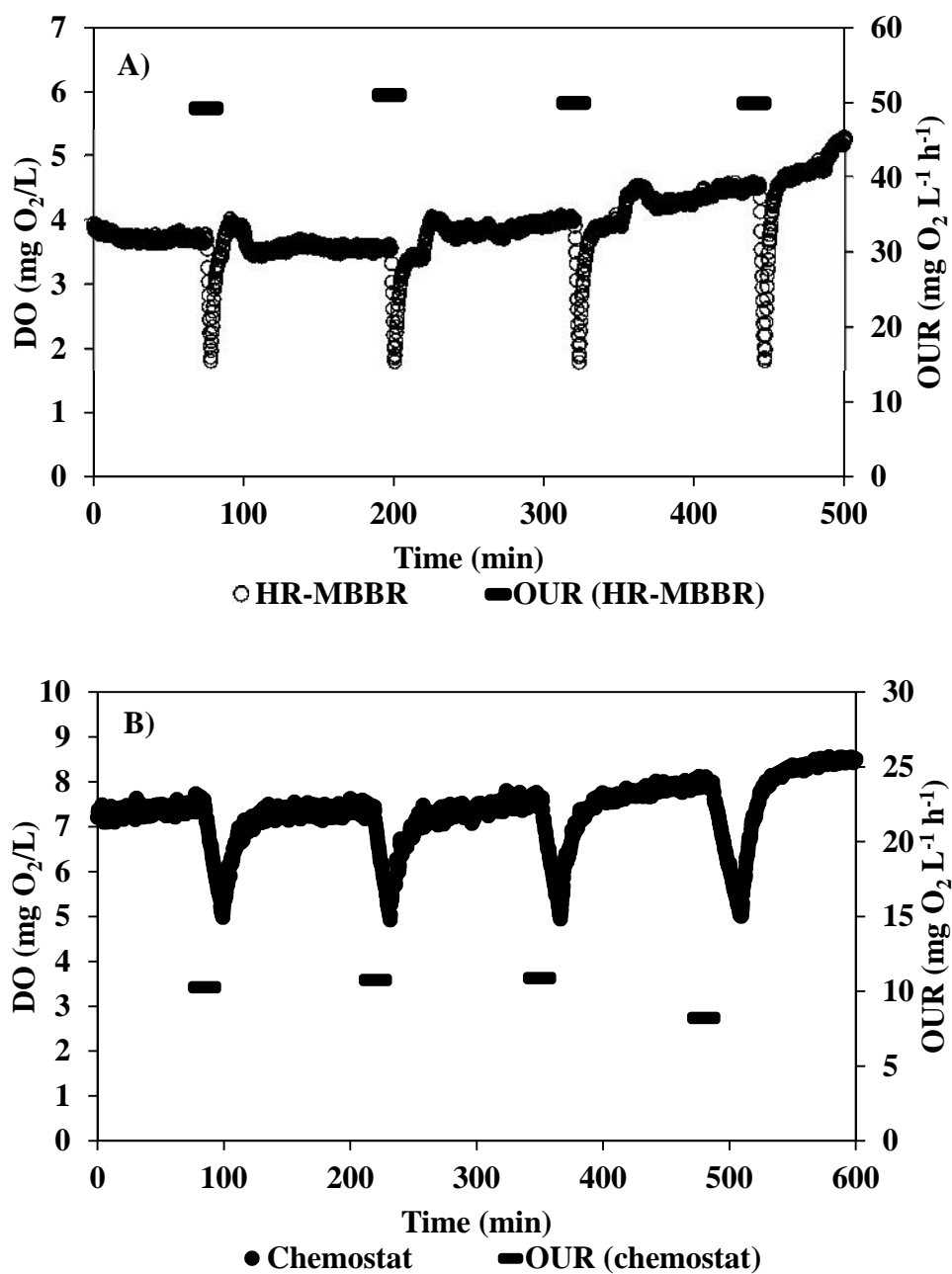
The parallel physicochemical and biological method between flocculation filtration method and batch test under aerobic condition is conducted to measure soluble unbiodegradable COD.

It is proposed that all colloidal, particulate biodegradable and unbiodegradable will remove by flocculation filtration ( $ffCOD$ ) method (Mamais et al. 1993) and the supernatant contains only soluble biodegradable and Unbiodegradable COD ( $ffCOD_{Inf}$ ). Therefore, a batch test method is used for estimating the mass of soluble Unbiodegradable which remain under aerobic condition ( $ffCOD_{EFF}$ ) at sludge ages greater than about 20 days.



## APPENDIX C – DO PROFILES

Figure C5: DO profile under aerated and non-aerated periods and OUR for the A) MBBR (OC2) and B) chemostat (OC1) processes



## APPENDIX D – HYDRAULIC TRACER TESTS

Table D6: Tracer results for the first experiment

Time	Time <sup>2</sup>	t	Abs.	Abs. BDF	Conc. RWT	Q		Mass RWT	Cum. mass	Cum.	t/T	C/C <sub>0</sub>	C. t	t.C. t	t <sup>2</sup> .C. t
min	h <sup>2</sup>	h			mg/L	m <sup>3</sup> /h	L/min	mg	mg	%	min		mg/L.h	mg/L.h	mg/L.h <sup>3</sup>
0	0.00E+00	0.00E+00	0.03	0.000	0.00	2.91	48.5	0	0	0%	0.00	0.0	0.00E+00	0.00E+00	0.00E+00
0.2	7.72E-06	2.78E-03	0.032	0.002	-0.03	2.91	48.4	0	0	0%	0.00	0.0	-7.92E-05	-2.20E-07	-6.11E-10
0.3	3.09E-05	2.78E-03	0.982	0.952	7.03	2.91	48.4	28	28	1%	0.01	3.9	1.95E-02	1.08E-04	6.03E-07
0.5	6.94E-05	2.78E-03	1.444	1.414	10.46	2.91	48.4	71	99	3%	0.01	5.7	2.91E-02	2.42E-04	2.02E-06
0.7	1.23E-04	2.78E-03	0.435	0.405	2.97	2.91	48.4	54	153	5%	0.02	1.6	8.24E-03	9.15E-05	1.02E-06
0.8	1.93E-04	2.78E-03	0.306	0.276	2.01	2.91	48.4	20	173	6%	0.02	1.1	5.57E-03	7.74E-05	1.08E-06
1.0	2.78E-04	2.78E-03	0.220	0.190	1.37	3.02	50.3	14	187	6%	0.03	0.8	3.80E-03	6.33E-05	1.06E-06
1.2	3.78E-04	2.78E-03	0.226	0.196	1.41	3.02	50.3	12	199	6%	0.03	0.8	3.92E-03	7.63E-05	1.48E-06
1.3	4.94E-04	2.78E-03	0.214	0.184	1.32	3.02	50.3	11	210	7%	0.04	0.7	3.68E-03	8.17E-05	1.82E-06
1.5	6.25E-04	2.78E-03	0.213	0.183	1.32	3.02	50.3	11	221	7%	0.04	0.7	3.66E-03	9.14E-05	2.28E-06
1.7	7.72E-04	2.78E-03	0.230	0.200	1.44	3.02	50.3	12	233	8%	0.05	0.8	4.01E-03	1.11E-04	3.09E-06
2.0	1.11E-03	5.56E-03	0.210	0.180	1.29	3.07	51.1	23	256	8%	0.06	0.7	7.19E-03	2.40E-04	7.99E-06
2.5	1.74E-03	8.33E-03	0.218	0.188	1.35	3.07	51.1	34	290	9%	0.07	0.7	1.13E-02	4.70E-04	1.96E-05
3.0	2.50E-03	8.33E-03	0.217	0.187	1.35	3.02	50.3	34	324	10%	0.09	0.7	1.12E-02	5.61E-04	2.80E-05
4.0	4.44E-03	1.67E-02	0.201	0.171	1.23	2.53	42.2	54	378	12%	0.12	0.7	2.04E-02	1.36E-03	9.09E-05

Table D7: Tracer results for the first experiment (continued)

Time	Time <sup>2</sup>	t	Abs.	Abs. BDF	Conc. RWT	Q		Mass RWT	Cum. mass	Cum.	t/T	C/C <sub>0</sub>	C. t	t.C. t	t <sup>2</sup> .C. t
min	h <sup>2</sup>	h			mg/L	m <sup>3</sup> /h	L/min	mg	mg	%	min				
5.0	6.94E-03	1.67E-02	0.193	0.163	1.17	3.16	52.6	63	441	14%	0.15	0.6	1.95E-02	1.62E-03	1.35E-04
7.0	1.36E-02	3.33E-02	0.183	0.153	1.09	3.08	51.3	116	557	18%	0.21	0.6	3.64E-02	4.25E-03	4.96E-04
10	2.78E-02	5.00E-02	0.166	0.136	0.97	2.93	48.8	151	708	23%	0.29	0.5	4.83E-02	8.06E-03	1.34E-03
14	5.44E-02	6.67E-02	0.161	0.131	0.93	3.02	50.3	191	899	29%	0.41	0.5	6.20E-02	1.45E-02	3.37E-03
20	1.11E-01	1.00E-01	0.180	0.150	1.07	2.99	49.8	299	1197	39%	0.59	0.6	1.07E-01	3.57E-02	1.19E-02
30	2.50E-01	1.67E-01	0.132	0.102	0.71	3.33	55.5	495	1693	55%	0.88	0.39	1.19E-01	5.95E-02	2.98E-02
41	4.67E-01	1.83E-01	0.113	0.083	0.57	3.02	50.3	356	2049	66%	1.21	0.31	1.05E-01	7.18E-02	4.91E-02
62	1.07E+00	3.50E-01	0.079	0.049	0.32	3.05	50.8	477	2526	82%	1.82	0.18	1.12E-01	1.16E-01	1.20E-01
80	1.78E+00	3.00E-01	0.064	0.034	0.21	2.98	49.7	237	2763	89%	2.35	0.11	6.28E-02	8.37E-02	1.12E-01
105	3.06E+00	4.17E-01	0.049	0.019	0.00	2.98	49.7	130	2893	93%	3.09	0.00	0.00E+00	0.00E+00	0.00E+00
155	6.67E+00	8.33E-01	0.047	0.017	0.00	3.01	50.2	0	2893	93.4%	4.56	0.00	0.00E+00	0.00E+00	0.00E+00

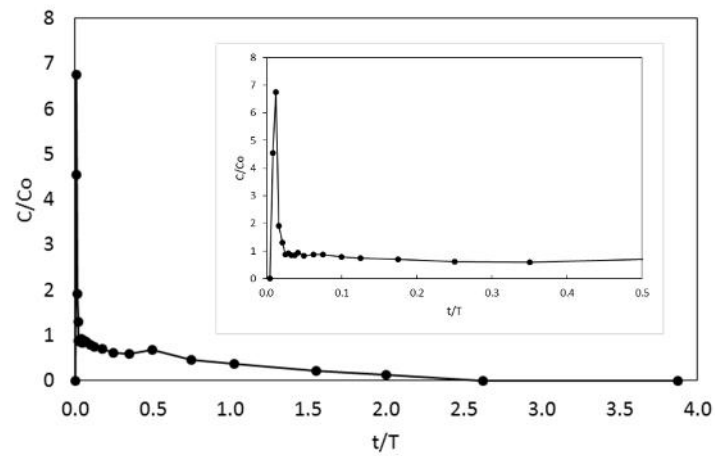


Figure D1: Normalization curve of measured tracer concentration in the effluent of HR-MBBR (1<sup>st</sup> test)

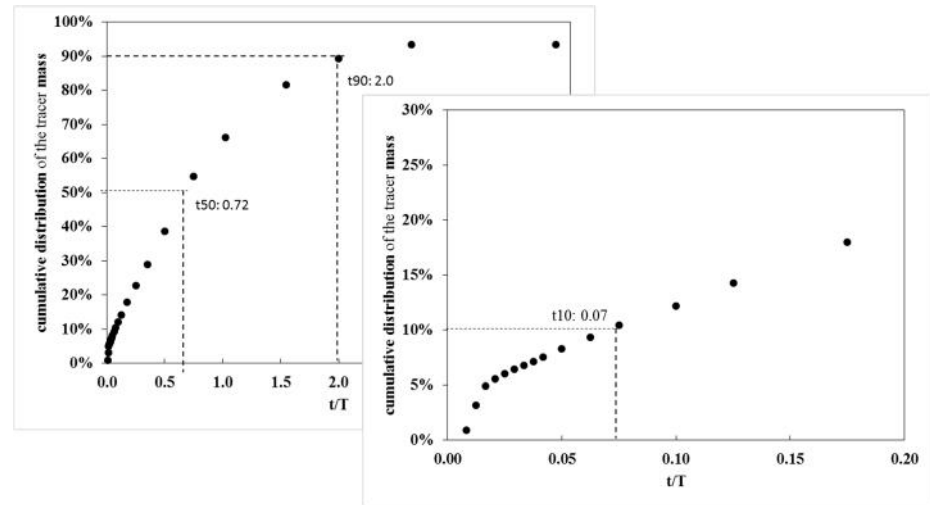


Figure D1 : Cumulative curve of measured tracer from the effluent of HR-MBBR (1<sup>st</sup> test)

Table D2: Tracer results for second test

Time	Time <sub>2</sub>	t	Q		t/HRT <sub>theo</sub>	Abs.	Conc. RWT	Conc. blank	C/C <sub>0</sub>	Recovered mass	cumulative mass	Cumulative mass	C* t	t * C * t	t <sup>2</sup> * C * t
min	Hour	H	m3/h	L/min	-	-	ppm	ppm	-	mg	mg	%	mg/L*h	mg/L*h	mg/L*h <sup>3</sup>
0.00	0.00	0.00	3.00	50.00	0.00	0.006	0.03	-0.13	-0.08	0.0	0	0%	0.00E+00	0.00E+00	0.00E+00
0.42	0.00	0.01	3.00	50.00	0.01	0.091	0.66	0.51	0.34	4.0	4	0%	3.54E-03	2.46E-05	1.71E-07
0.75	0.00	0.01	3.00	50.00	0.02	0.206	1.53	1.37	0.90	15.7	20	1%	7.62E-03	9.52E-05	1.19E-06
1.00	0.00	0.00	3.00	50.00	0.03	0.193	1.43	1.27	0.84	16.5	36	1%	5.31E-03	8.85E-05	1.47E-06
1.25	0.00	0.00	3.00	50.00	0.03	0.199	1.47	1.32	0.87	16.2	52	2%	5.50E-03	1.14E-04	2.39E-06
1.50	0.00	0.00	3.00	50.00	0.04	0.202	1.50	1.34	0.88	16.6	69	2%	5.59E-03	1.40E-04	3.49E-06
1.75	0.00	0.00	3.00	50.00	0.04	0.193	1.43	1.27	0.84	16.3	85	3%	5.31E-03	1.55E-04	4.52E-06
2.17	0.00	0.01	3.00	50.00	0.05	0.192	1.42	1.27	0.83	26.5	112	4%	8.80E-03	3.18E-04	1.15E-05
2.50	0.00	0.01	3.00	50.00	0.06	0.193	1.43	1.27	0.84	21.2	133	4%	7.08E-03	2.95E-04	1.23E-05
3.00	0.00	0.01	3.00	50.00	0.08	0.19	1.41	1.25	0.82	31.6	165	5%	1.04E-02	5.21E-04	2.61E-05
4.00	0.00	0.02	3.00	50.00	0.10	0.187	1.38	1.23	0.81	62.0	227	7%	2.05E-02	1.37E-03	9.10E-05
5.00	0.01	0.02	3.00	50.00	0.13	0.184	1.36	1.21	0.79	60.9	287	9%	2.01E-02	1.68E-03	1.40E-04
6.00	0.01	0.02	3.00	50.00	0.15	0.181	1.34	1.18	0.78	59.8	347	11%	1.97E-02	1.97E-03	1.97E-04
7.00	0.01	0.02	3.00	50.00	0.18	0.183	1.35	1.20	0.79	59.6	407	13%	2.00E-02	2.33E-03	2.72E-04
8.00	0.02	0.02	3.00	50.00	0.20	0.178	1.32	1.16	0.76	59.0	466	15%	1.94E-02	2.58E-03	3.44E-04

Table D2: Tracer results for second test (continued)

Time	Time <sub>2</sub>	$\Delta t$			t/HRT theo	Absorb	Conc. RWT	Conc. blank	C/C <sub>0</sub>	Recovered mass	cumulative mass	Cumulative mass	C* $\Delta t$	t * C * $\Delta t$	t <sup>2</sup> * C * $\Delta t$
min	Hour	H	m <sup>3</sup> /h	L/min	-	-	ppm	ppm	-	mg	mg	%	mg/L*h	mg/L*h	mg/L*h <sup>3</sup>
9.50	0.03	0.03	3.00	50.00	0.24	0.17	1.26	1.10	0.72	84.9	551	18%	2.75E-02	4.36E-03	6.90E-04
11.00	0.03	0.03	3.00	50.00	0.28	0.171	1.26	1.11	0.73	82.9	634	21%	2.77E-02	5.08E-03	9.32E-04
12.50	0.04	0.03	3.00	50.00	0.31	0.163	1.20	1.05	0.69	80.9	715	24%	2.62E-02	5.46E-03	1.14E-03
15.00	0.06	0.04	3.00	50.00	0.38	0.159	1.17	1.02	0.67	129.3	844	28%	4.25E-02	1.06E-02	2.65E-03
20.00	0.11	0.08	3.00	50.00	0.50	0.141	1.04	0.88	0.58	237.9	1082	36%	7.37E-02	2.46E-02	8.19E-03
30.00	0.25	0.17	3.00	50.00	0.75	0.117	0.86	0.70	0.46	397.2	1479	49%	1.17E-01	5.87E-02	2.94E-02
40.00	0.44	0.17	3.00	50.00	1.00	0.104	0.76	0.61	0.40	327.9	1807	59%	1.01E-01	6.74E-02	4.50E-02
50.00	0.69	0.17	3.00	50.00	1.25	0.096	0.70	0.55	0.36	288.5	2095	69%	9.12E-02	7.60E-02	6.33E-02
60.00	1.00	0.17	3.00	50.00	1.50	0.092	0.67	0.52	0.34	266.0	2361	78%	8.62E-02	8.62E-02	8.62E-02
75.75	1.59	0.26	3.00	50.00	1.89	0.059	0.42	0.27	0.18	309.8	2671	88%	7.08E-02	8.94E-02	1.13E-01
95.00	2.51	0.32	3.00	50.00	2.38	0.056	0.40	0.25	0.16	248.9	2920	96%	7.93E-02	1.26E-01	1.99E-01
126.00	4.41	0.52	3.00	50.00	3.15	0.036	0.25	0.10	0.06	267.2	3187	105%	5.03E-02	1.06E-01	2.22E-01
151.00	6.33	0.42	3.00	50.00	3.78	0.032	0.22	0.07	0.04	103.0	3290	108%	2.81E-02	7.07E-02	1.78E-01
161.00	7.20	0.17	3.00	50.00	4.03	0.026	0.18	0.02	0.01	22.5	3313	109%	3.75E-03	1.01E-02	2.70E-02
181.00	9.10	0.33	3.00	50.00	4.53	0.022	0.15	-0.01	0.00	7.5	3320	109%	-2.50E-03	-7.54E-03	-2.27E-02
208.00	12.02	0.45	3.00	50.00	5.20	0.034	0.24	0.08	0.05	50.6	3371	111%	3.71E-02	1.29E-01	4.46E-01

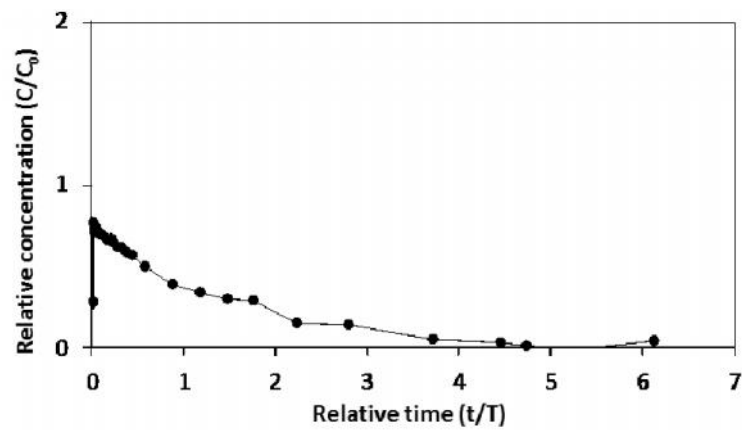


Figure D2 : Normalization curve of measured tracer concentration in the effluent of HR-MBBR (2<sup>nd</sup> test)

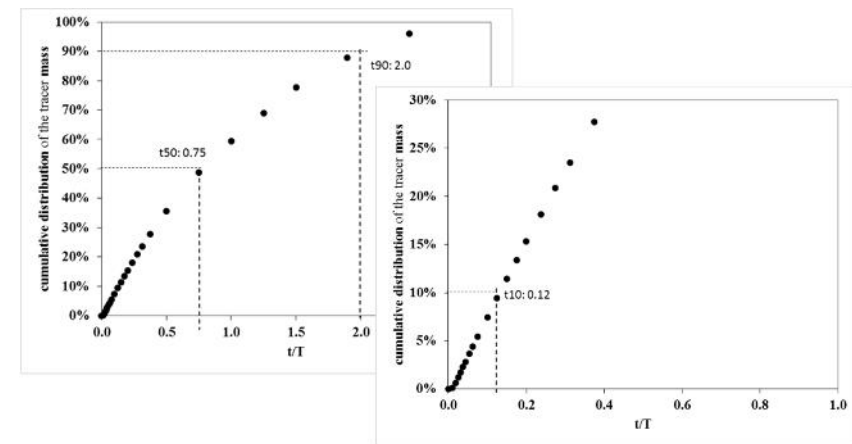


Figure D3 : Cumulative curve of measured tracer from the effluent of HR-MBBR (2<sup>nd</sup> test)