Biofouling Reduction in Electro Coagulation Integrated Membrane Bioreactor using Quorum Quenching Technique

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ABSTRACT

Membrane fouling reduction is requisite for effective operation of membrane treatment technologies. During Phase 1, four electro bioreactors were operated at different electric exposure modes i.e., 5’ON-10’OFF, 5’ON-20’OFF, 5’ON-30’OFF, 5’ON-40’OFF besides control reactor and their effect on sludge characteristics was studied. 5’ON-30’OFF and 5’ON-40’OFF modes were optimized from stage 1 and further used with and without Quorum Quenching (QQ) Beads in stage 2. From stage 2, 5’ON-30’OFF (QQ) was selected for further study. During Phase 2, continuous operation of submerged membrane electro-bioreactor (SMEBR(QQ)) and submerged membrane bioreactor (SMBR(QQ)) was studied in which hollow cylindrical (HC) beads having *Rhodococcus sp.* (BH4) specie were used for membrane biofouling reduction. Porous aluminum anode and stainless steel cathode were used as electrodes in SMEBR(QQ). Various parameters including Mixed Liquor Suspended Solids (MLSS), Sludge Volume Index (SVI), Time to Filter (TTF), Particle Size Distribution (PSD), COD and nutrients removal and Transmembrane pressure (TMP) were examined to visualize the microbial and sludge activity. By introducing HC QQ beads TMP rise delayed from 44 to 46 days and with the addition of electrocoagulation as SMEBR(QQ) TMP rise further delayed to 30KPa within 58 to 60 days. This showed that, SMEBR(QQ) reduced the fouling rate more than SMBR(QQ) depicting long filtration time in membrane bioreactor (MBR).

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List of Abbreviations

Abbreviation	**Description**
ASP	Activated sludge process
CD	Current Density
CEBs	Cell entrapping beads
C-MBR	Conventional Membrane bioreactor
COD	Chemical oxygen demand
SVI	Sludge Volume Index
EPS	Extra polymeric substance
F/M	Food to microorganism ratio
HF	Hollow-fiber
HRT	Hydraulic retention time
J	Operational flux
MLSS	Mixed liquor suspended solids
MLVSS	Mixed liquor volatile suspended solids
NTU	Nephelometric turbidity unit
OLR	Organic loading rate
PSD	Particle size distribution

QQ-MBR	Quorum quenching membrane bioreactor
Rc	Cake resistance
Rp	Resistance due to pore blocking
Rm	Intrinsic membrane resistance
Rt	Total resistance
SMBR	Submerged membrane bioreactor
SMP	Soluble microbial product
SRT	Sludge retention time
TMP.	Trans-membrane pressure
TN	Total nitrogen

1. Introduction

1.1 Background

The most obvious important realities are often the ones that are hardest to see and talk about. Sustainable Water consumption is one of those realities. From the overall resources of water on earth 97% of which is in the oceans, 3% is freshwater, 66% of which is tied up as ice in icy masses and at the poles. This leaves roughly 1% as freshwater in streams, lakes, as groundwater and in the atmosphere, out of which only 0.5% is available for human use (World Bank, 2005). However, because of a growing worldwide populace with desires of higher expectations for everyday comforts and cultivation, that 0.5% is under serious threat. Water will become scarce in regions where it is currently abundant - such as Central Africa and East Asia - and scarcity will greatly worsen in regions where water is already in short supply - such as the Middle East and the Sahel in Africa. These regions could see their growth rates decline by as much as 6 percent of GDP by 2050 due to water related impacts on agriculture, health, and incomes. (World bank, 2016). The uneven condition of water protection has driven the Pakistan government to reexamine gathering, treating and reusing the gigantic measures of produced wastewater to make a reasonable wellspring of water for agricultural and landscaping purposes.

In the Sustainable Development Goals (SDGs) lies the solution to Pakistan's three central challenges: development, democracy and defense. The objectives of Sustainable Development Goals (SDGs) underpin good governance and integrate three dimensions of sustainable development – economic development, social inclusion, and environmental sustainability (Sheikh, 2016). Water sustainability can only be possible if treated wastewater is used for agricultural and landscaping purposes. Human exercises create enormous volume of sewage and wastewater that require treatment before release into conduits. Frequently this wastewater contains intemperate measures of nitrogen, phosphorus, and metal mixes, and in addition natural contaminations that would overpower conduits with an outlandish weight. In a time where there is developing concern of the worldwide effect of our recent environmental management strategies, and the need to diminish sanitation issues, infections, and diseases, there is an important need to create more environmentally responsible, suitable wastewater treatment technologies whose performance is balanced by environmental, societal sustainability and economy. For designing a WWTP, besides generating a high-quality effluent, other factors are also important i-e tendency to lower the consumed energy, costs, release of waste materials from different portions of operational units of WWTP and lastly footprints by saving the area needed for construction.

To reduce the impacts of wastewater on environment, it is necessary to remove excess amount of nutrients from wastewater. Different physicochemical and biological processes have been used for removing nutrients from wastewater. Biological treatment has capacity to transform nutrients and to remove the concentration of organic and inorganic compounds from the wastewater. There are two types of biological treatment methods, aerobic and anaerobic treatment. The most common and renown method previously was activated sludge process in which wastewater is treated aerobically and microorganisms that carries out the treatment are kept in liquid suspension with provision of mixing through aeration. Adequate amount of air is provided for both proper mixing of wastewater for microbe’s suspension and to provide minimum of 2mg/L of dissolved oxygen to microorganisms for their growth and food degradation. The activated sludge process has three basic components: 1) a reactor in which the microorganisms are kept in suspension, aerated, and in contact with the waste they are treating; 2) liquid-solid separation; and 3) a sludge recycling system for returning activated sludge back to the beginning of the process.

Large amounts of extracellular polymeric substances (EPS) can be found in mixed liquor of sludge through which bacteria are held together in flocs. Although this process gives 95% removal efficiency in wastewater treatment but the major disadvantage of ASP is the production of excess sludge by conversion of organics into bacterial cells. This dry weight excessive sludge is a secondary solid waste that must be disposed off in a cost effective and safer way. The handling of the excess sludge may account for 25% to 65% of the total plant operation cost. (Liu, 2003).

From the last two decades Membrane biotechnology was used for wastewater treatment. (Wei, 2012). MBR can replace the physical separation processes of conventional wastewater treatment technology by filtering the biomass through a membrane. Thus, the effluent water quality is considerably higher than by conventional treatment process, due to which there is no need of additional tertiary disinfection process.

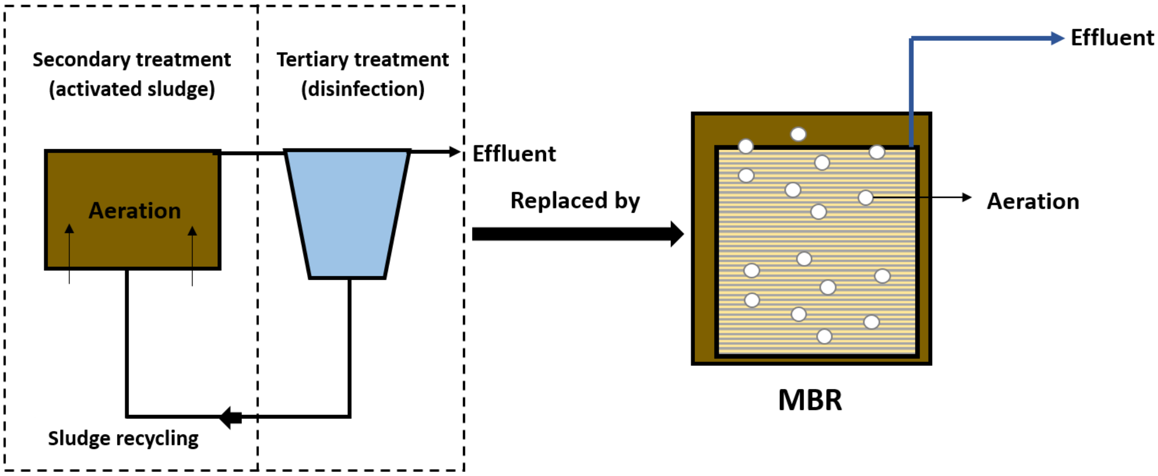

Figure 1.1 Schematic illustration of MBR replacing Conventional ASP

Some of the major advantages of MBR includes; (Judd, 2008)

- Production of high quality, largely disinfected and clarified effluent in a single stage as the effective pore size of usual membrane is <0.1 µm which is smaller than the pathogenic microbes in the wastewater.
- In ASP, an appropriately longer HRT for growth of microorganisms is required because separation of solids depends on growth of mixed liquor solid particles to an adequate size (> 50µm) so that they can be removed by settlement. In an MBR the particles should only be larger than the membrane pore size.
- Operation at greater MLSS concentrations to enhance ammonia removal by growth of specific nitrifying bacteria due to reduction in the required reactor size.
- Lesser sludge production due to longer SRTs.

Fouling is the blocking of membrane pores, which reduces the flow of permeate through membrane (Judd, 2008). Rapid fouling during operation depends upon many factors which includes sludge characteristics, membrane characteristics, operating parameters, and feed water characteristics. (Giwa, 2015). In MBR, the membrane requires both physical and chemical cleaning due to which life of membrane decreases and cost of treatment increases. (Shadi W. Hasan, 2014). Number of researches have already worked to control membrane fouling by physical and chemical techniques so that the loss of permeate could be stopped. Major drawback of these methods includes a very short time enhancement in the rate of filtration. Control on the formation of bio cake layer on membrane surface provides greater life span of membrane resulting into larger amount of permeate. Quorum sensing is the production of signal molecules by bacteria. When the concentration of these molecules arises, they combine with the receptor protein to form biofilm and EPS. EPS is the main factor of membrane fouling. To stop this EPS and biofilm production quorum quenching technique is used. Electro-technologies combine with the wastewater treatment processes provides effective solutions due to their high efficiency, ease of control and lesser dependence on external chemicals. The Submerged Membrane Electro-Bioreactor "SMEBR" proves to boost wastewater treatment performance which works both on membrane filtration and electro kinetic phenomena.

Therefore, it is important to develop new methods to deal with wastewater treatment.

1.2 Objectives

1. Establishment of automated lab scale submerged electro coagulation integrated membrane bioreactor in IESE NUST
2. Optimization of intermittent supply of the induced current
3. Investigate the effect of hollow cylindrical quorum quenching beads on performance efficiency of MBR and biofouling reduction
4. Effect of Quorum Quenching technique in combination with induced electric current on biofouling reduction rate of SMEBR

1.3 Scope of Study

1. Formulated Lab scale submerged electro coagulation MBR setup with PVDF hollow fiber membrane and working volume of 19 L to treat synthetic wastewater.
2. Examined performance efficiency of MBR between QQ operated and SMEBR with QQ.
3. Impact of applied current density and cell entrapped beads(CEBs) consisting of quorum quenching bacteria on bio fouling reduction rate in the treatment of synthetic municipal wastewater.

2 Literature Review

2.1 Membrane Configuration

2.1.1 Cross flow filtration

In this technique, high velocity liquid passes cross flow or parallel to the membrane medium surface to produce shear pressure to scour the surface. Additional energy is needed to produce crossflow, but thickness of cake layer can be controlled. This filtration technique is mainly effective when feed water has high quantity of macro molecules and suspended solids. Most of wastewater filtrations and MBR processes are using this technique. (Yoon, 2016)

2.1.2 Dead end filtration

In this basic form of filtration, feed moves to the filter medium. Particles settle on the filter surface. This filtration technique is mainly effective when feed water has low level of pollutants. As the medium is replaced or backwashing is performed occasionally. Many tertiary filtrations, preliminary treatment for seawater RO and surface water filtrations are using dead-end filtration technique. (Yoon, 2016). After cake layer removal, membrane may be used further.

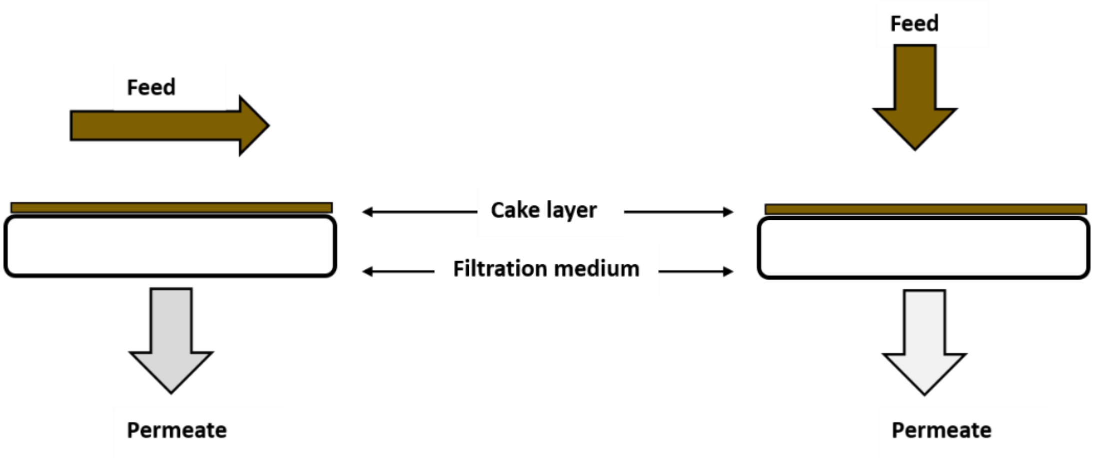

Figure 2.1 a) Cross flow Filtration b) Dead end Filtration

2.2 Membrane bioreactor systems

As the name proposes, Membrane bioreactor techniques are those that gives membrane separation with biological treatment. An MBR technology may

substitute the two physical separation techniques by straining the biomass through a membrane due to which the water quality is considerably greater as compared to the conventional treatment, avoiding the need for tertiary disinfection process. (Judd, 2008).

2.3 MBR process description

A membrane in bioreactor is responsible to stop impurities and biomass to achieve a significant amount of permeate. In MBRs activated sludge biological processes are used, however secondary clarifier is alternated by membrane module. By this method, complete solids-liquid partition may be done. Thus, with lesser necessities for processing MBR gives efficient treatment. Membrane comprises of a permeable layer which is actually a hindrance to bacteria, solids and other undesirable molecules.

Membrane process may be distributed into various categories per molecular weight, pore size and the pressure upon which they are regulated. Pressure on membrane for the separation of unwanted solids from water increases as the pore size of membrane decreases.

2.3.1 Microfiltration

Micro filtration (MF) is the treatment of eliminating particles or biological creatures in the 0.1 µm to 10.0 µm range. Although micro particles may be separated by use of depth materials such as in fibrous media, only a micro filter having an exactly described pore size may guarantee quantitative retaining. Mostly microfiltration removes many types of bacteria but viruses cannot be retained by this type of membrane. Micro filtration may be used before Nano-filtration or reverse osmosis. (Maqbool, 2014)

2.3.2 Ultrafiltration

Ultrafiltration (UF) may be used for eliminating dissolved molecules and very small particles from fluids. Materials from 0.01 to 1 µm range in size are removed by many ultrafiltration membranes, while water and salts will pass through. (Yoon, 2016)

2.3.3 Nano filtration

Nano filtration membranes, are becoming popular for removing very minute particles and viruses having pore size varying from 0.001 to 0.01 µm. (Taylor & Jacobs, 1996). It is mostly used in water purification for micro pollutant removal, decolorization and softening.

2.3.4 Reverse osmosis

Reverse osmosis RO membranes are proficient of retaining even minutest solute particles of as small as 0.0001 µm. (Taylor & Jacobs, 1996). In this process two solutions having different concentrations interacts together and then separated by a semi porous membrane. Water moves from higher concentration to lower concentration solution which we call osmosis. After applying pressure water will move from lower to higher concentration hence due to this reverse effect it is called as reverse osmosis. By this procedure one may remove approximately all salts.

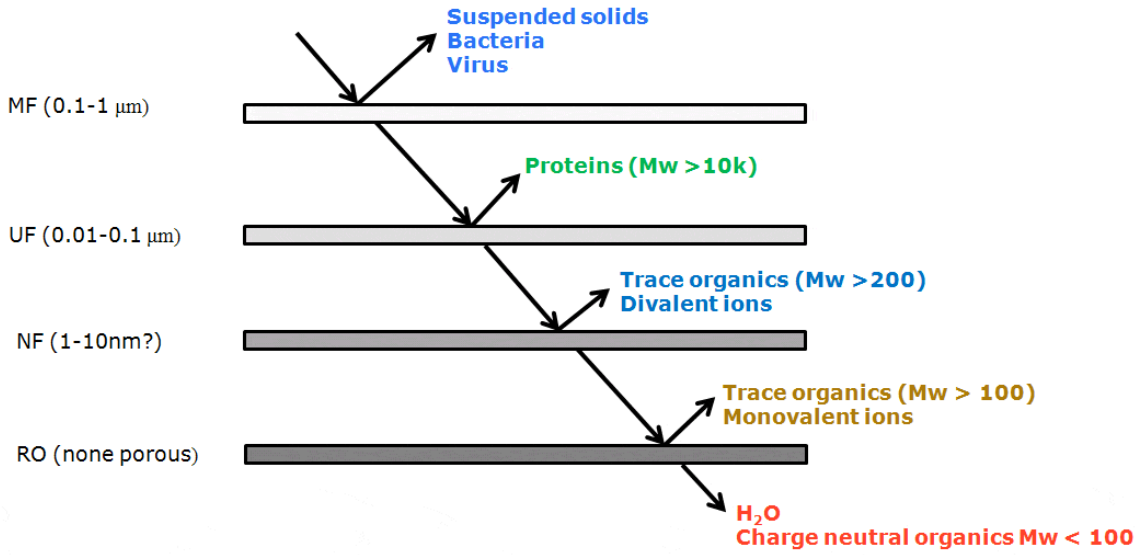

Figure 2.2 Particle separation as a function of pore size (Yoon, 2016)

2.4 Configuration of Membrane

2.4.1 Submerged MBR

In SMBR, membrane is directly immersed in the bioreactor where filtration happens under vacuum. This design is discovered to be more significant than external membrane. In Submerged MBR shear stress as compared to SS-MBR is high due to aeration but that can be controlled by varying aeration rate due to which there is low membrane fouling rate and high permeate flux. The advantages of submerged MBR includes, control of Oxygen demand in activated sludge, smaller footprints, lower operating costs in terms of lesser energy consumption (Günder, 2001). Disadvantages includes high aeration cost and susceptibility to membrane fouling. (Hasan, 2011)

2.4.2 Side stream (SS) MBR/ External MBR

In External MBR, the membrane is placed outside the bioreactor tank where sludge is recirculated at a higher flow rate. To end the deposition of suspended solids on membrane surface significant cross flow velocity is required which is the main reason for high energy consumption. (Clech et al., 2005). Advantages of external MBR includes complete solids removal from wastewater, solids and nutrients removal in one unit, small footprint, permeate disinfection, rapid start up and low sludge production. Whereas disadvantages include membrane fouling, aeration restrictions, high membrane and operating costs, process complications and high cleaning requirements. (Hasan, S. 2011)

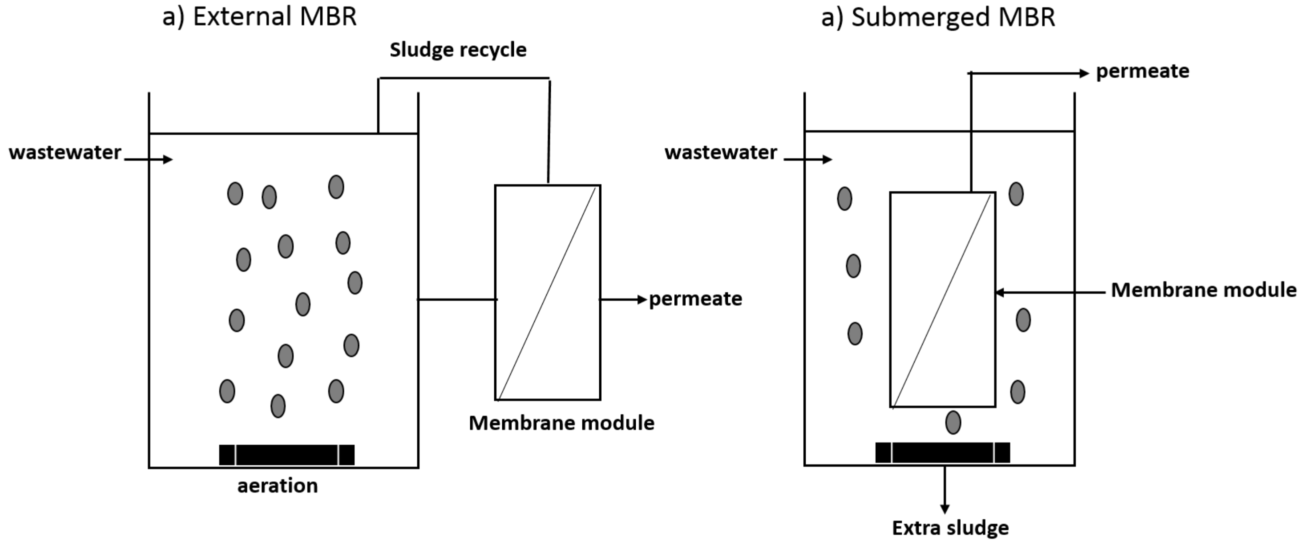

Figure 2.3 a) side stream MBR b) submerged MBR

2.5 Membrane Types and Materials

2.5.1 Hollow Fiber

Hollow fiber membranes are made up of fibers or long strands mostly composed of organic polymers. These strands are attached on a secondary assembly or structure which functions as a manifold for effluent passage, and thus enhances the life of membrane by preventing the cake formation (Frederickson, 2005). In hollow fiber membrane, there are two different approaches of filtration according to the direction of effluent flow that includes inside-out and outside-in (Hasan, 2011).

2.5.2 Flat Sheet

Flat sheet membranes involves two flat sheets paper type backing material of an organic polymer, attached within a frame. The filtration is offered by inserting the space about ¼” apart between the sheets of membrane by vacuum (Frederickson, 2005). The permeate passes from the outside in through the membrane unit. Flat sheet membranes that are used in Membrane Bioreactor needs a biofilm so that they may be attached to the membrane unit.

2.5.3 Tubular

Tubular membrane units have one or more tubes of different diameters. These tubes are made up of microporous material which gives strength and membrane is attached as a defined porous surface sheet on the inside of the tube. Through these tubes Wastewater is pumped and treated effluent flows through pores. These types of membranes are more tough and robust that may be exposed to high pressures.

2.6 Membrane Bioreactor Operating Parameters

Trans-membrane Pressure (TMP) and permeate flux are the key elements of any membrane filtration process. TMP is basically the pressure difference between in and outside of membrane, i-e pressure at permeate side and pressure at sludge side. TMP is also used to calculate the membrane flux. Membrane fouling rate has a direct relation with flux. A clean membrane usually has a low TMP, on the other hand depending on the severity of fouling, a fouled membrane has a relatively high TMP. (Günder , 2001).

$$J = \frac{TMP}{\mu Rt}$$

J = Permeate flux, L/m^2. s

TMP= Trans Membrane pressure, kPa

μ = Permeate viscosity, Pa.s

R_t= Total Resistance = 1/m

$$R_t = R_C + R_F + R_M$$

Where,

R_C = Cake resistance from cake layer, 1/m

R_F = fouling resistance due to gel formation and solute adsorption into membrane pore, 1/m

R_M = inherent membrane resistance, 1/m

2.7 Limitations of MBR

2.7.1 Membrane Fouling

Membrane fouling states as the adsorption of feed water elements or deposition of other pollutants such as solutes, cell debris produced in the bioreactor, microbes, and colloids on the external and internal surface of membrane. There is an increase in the total resistance to filtration method due to collection of these materials and hence enlarging the energy requirement. It also creates an increase in TMP, decrease in the permeate flux, and hence worsening of membrane. Several factors relate fouling such as operating conditions, membrane characteristics, sludge properties and feed water characteristics. Among these factors operating conditions have secondary influence on membrane fouling by changing sludge conditions and sludge properties have direct impact on membrane fouling (Hasan, 2011). Fouling is basically the objectionable attachment of microorganisms to the surface and into pores of membrane. Effluent flux starts to decrease as fouling rate increases (Lee et al., 2001).

Main causes of membrane fouling include (Maqbool, 2014)

- Formation of thick cake layer
- Sludge adhesion on membrane medium
- Colloids deposition on membrane
- Temporal changes in foulants

2.8 Membrane Fouling Categories

2.8.1 Organic Fouling

Fouling connected with in both drinking water and wastewater is known as organic fouling. It is a most essential restriction to use of membranes in water and wastewater treatment. Organic fouling classification covers both biofouling and colloidal fouling. This type of fouling integrates both natural colloids and macromolecules (Amy, 2008)

2.8.2 Scaling

Adsorption of particles on membrane surface causing it to lump is stated as scaling. Scaling results in reducing a life period of membrane and use of high energy. It also causes the decline in permeate flux. Due to these reasons membrane water or wastewater treatment process becomes much more overpriced. Scaling control strategy includes addition of acids which can prevent the precipitation of salts on membrane surface. (“*Particles, scaling and biofouling Membrane technology,” 2015*)

2.8.3 Colloidal Fouling

Colloids are small fine particles having a size range of 1-1000 nm. In pressure-driven membrane techniques, these small particles have a strong and tough propensity to foul membranes, which results into significant loss in water motion and produces bad quality water.(Chuyang, 2011)

2.8.4 Biofouling

Membrane bioreactors (MBRs) have been in domestic and commercial use from above two decades, but membrane biofouling still remains a main limitation that restricts their broad application. Membrane biofouling is generally linked with cake layer formation due to undesirable accumulation of microorganisms over time on the surface of membrane. This happens because of deposition and growth of microbial cells on the membranes. Biofilm formation is based on following steps.

1. Conditioning film formation (proteins, macromolecules etc.)
2. Attachment of Planktonic cells on membrane surfaces
3. Microorganism colonies formation by primary biological bond
4. Development of Mature biofilm

2.9 Phases of Biofouling

2.9.1 Conditioning fouling

Conditioning fouling stage which is initial fouling caused by the interaction between EPS and SMP present in MLSS with membrane surface. Ognier et al. (2002) observed rapid irreversible fouling during initial stage, passive colloids and organics adsorption were also found even at zero flux operation. Ma et al. (2005) coupled vacuum pump instead of suction pump with air back flushing and could reduce the conditional fouling due to colloidal adsorption. Intensity of passive adsorption may affect the pore size distribution and surface chemistry of membrane. Cake layer start to develop on the membrane surface which not effect flux in the initial stage but over the time cake partially or completely block the membrane pores which result in TMP rise.

2.9.2 Steady Fouling

Even at below critical flux operation of membrane in biomass, temporally attached flocs on membrane surface may contribute to second stage of slow fouling. Most of the membrane surface already covered with EPS/SMP will promote more biomass flocs and colloidal attachment.

2.9.3 TMP jump

The abrupt rise of TMP or “Jump” is the result of filtration at constant flux and several mechanisms may be proposed for the TMP jump. Inhomogeneous fouling (area loss) model, Inhomogeneous fouling (pore loss) model, Critical suction pressure model, Percolation theory, Inhomogeneous fiber bundle model (Judd, 2008).

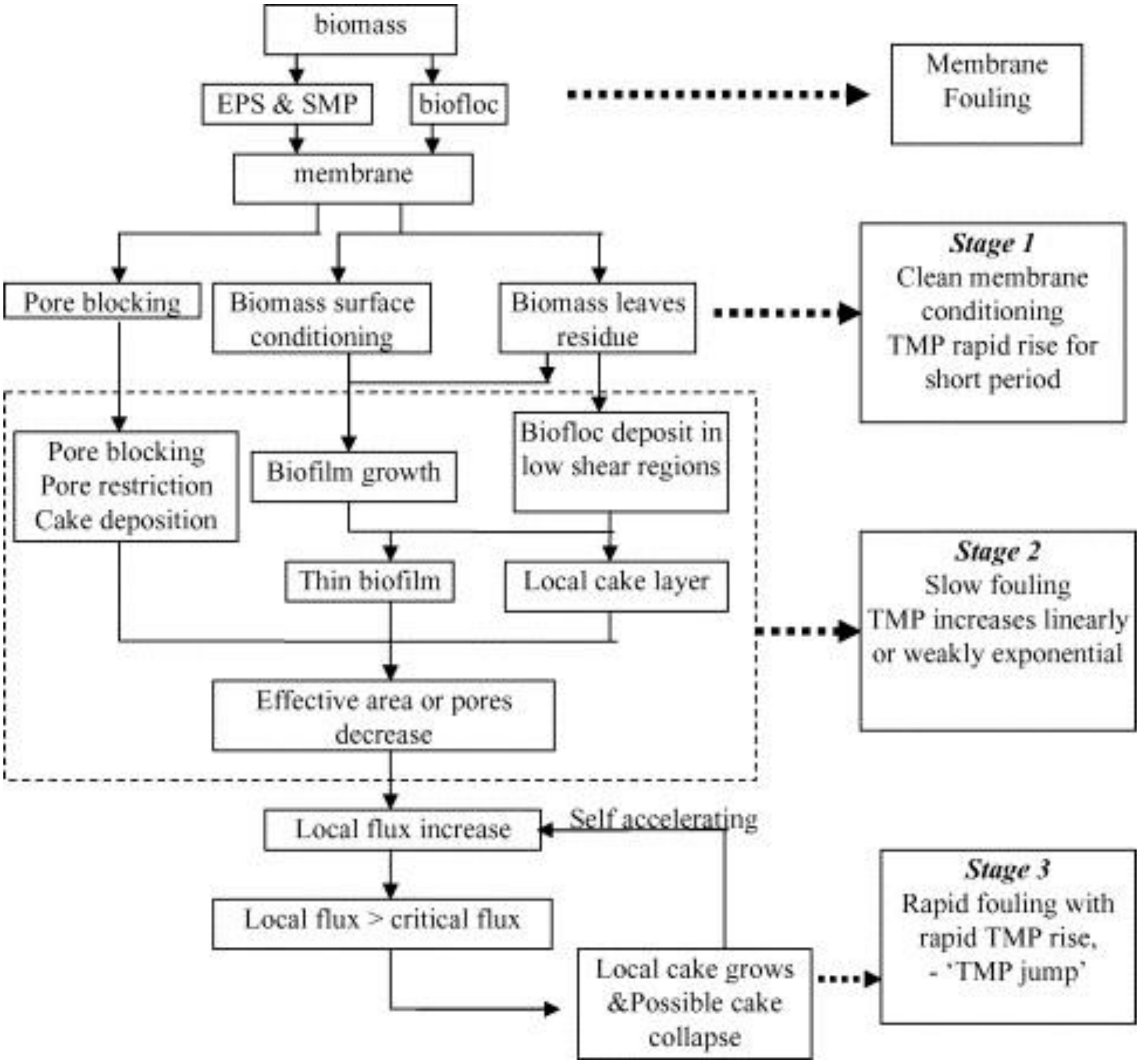

Figure 2.4 Mechanism of fouling during MBR operations (Zhang et al., 2006)

2.10 Factors Effecting Biofouling

The important factors affecting biofilm formation on membrane depends on carbon: nitrogen: phosphorus ratio, redox potential, pH and temperature. Extracellular polysaccharide (EPS) is the substance produced by microorganism is responsible for the slimy nature of biofilms (Baker, 1998).

Table 2.1 Variables influencing microbial adhesion to membrane surfaces

Microorganism	Surface	Feed water
Species	Roughness	Boundary layer
Composition of mixed population	Surface tension	Suspended matter
Population density	Surface charge	Shear forces
Growth phase	Porosity	Temperature
Nutrient status	Chemical composition	pH
Physiological responses	Hydrophobicity	Flux
Hydrophobicity		Viscosity
Charges		Dissolved organic substance

(Nguyen, 2012)

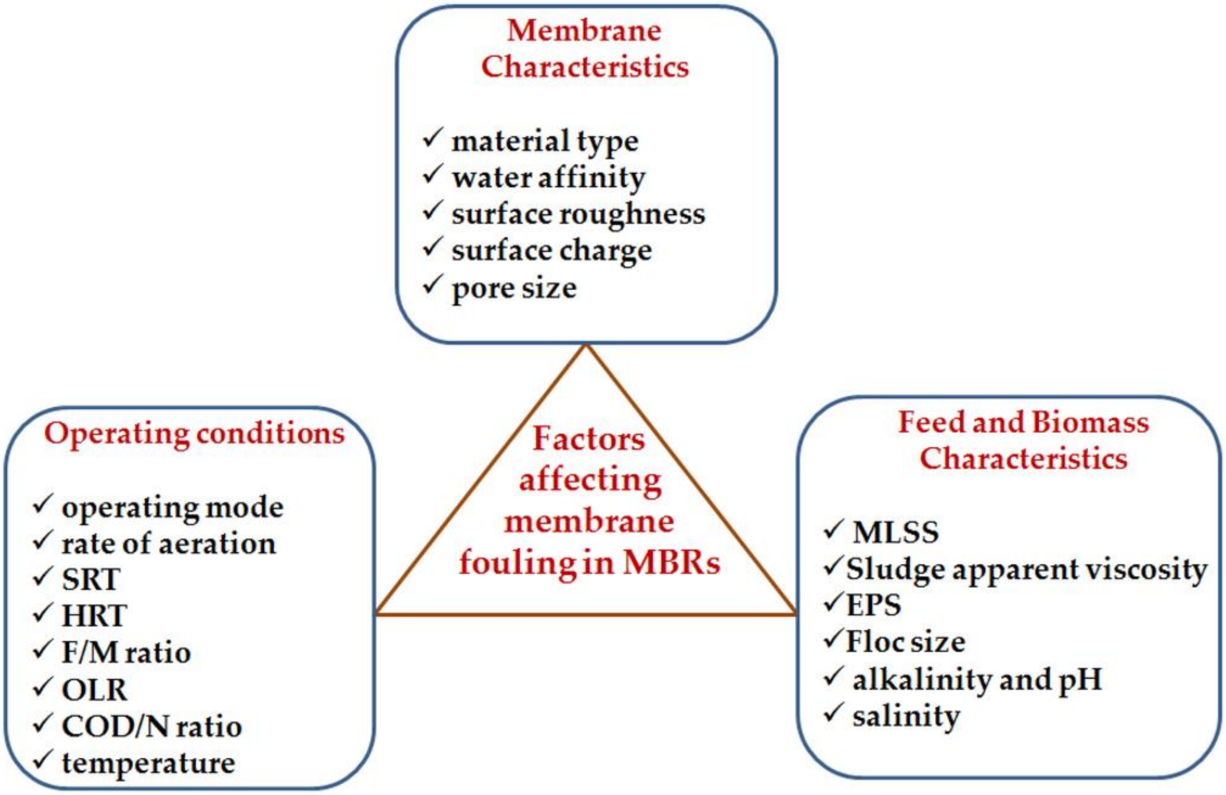

Figure 2.5 Factors affecting membrane fouling (Iorhemen et al., 2016)

2.11 Fouling Control Strategies and Membrane Cleaning

- Periodic relaxation modes
- Patterned membrane
- Backwashing with different flow rates
- Addition of moving media and adsorbent
- Air scouring / scrubbing
- Mechanical cleaning through biofilm carriers
- Novel approaches

The drawback of all these strategies is that they controlled the fouling up to short time and delayed TMP rise for few days than conventional MBR (Deng, et al., 2014; Wu, et al., 2011).

2.12 Quorum Sensing

Bacteria behave sometime in linked manner by cell to cell interaction called quorum communication/sensing and communicate or connect with one other by signal molecules called autoinducers. These signal molecules collect in the environment and when a critical concentration is achieved then associated manner is showed i.e virulence, secretion of polysaccharide and protein. Acyl homoserine lactones (AHLs) based quorum sensing is most common in gram negative bacteria in wastewater where more than 25 species are identified. More than half dozens of quorum sensing types have been described in bacteria. AHLs are the signaling molecules which are of twelve types.

2.12.1 Role of QS in biofouling

Shrout and Nerenberg (2012) described the phenomenon of quorum sensing and its main steps

- protein produces signal molecule by cell
- signal molecule concentrate in environment and
- regulatory protein accepts the signal molecule to complete the communication.

Mostly signal producing and receiving species are different, but some microbes show both the phenomenon simultaneously. Signal molecules receiving microbes play an important role in biofilm formation, those secreted EPS on receiving signal molecules. EPS helps in agglomeration of microbial flocs and biofilm formation. Researcher found a direct relation of AHLs concertation and EPS production. High AHLs concentration led to high membrane biofouling.

2.12.2 Quorum sensing control strategies

Three different point of attack on AHLs may control membrane biofouling:

1. Signal molecules generator cells
2. Generated signal molecules
3. Signal receptor cells

Recently, Quorum quenching method has been beforehand ended up being an objective for both quorum sensing signal synthase and sensors or response controller proteins. These systems may be connected to hinder AHLs-interceded quorum sensing in Gram-negative and AIPs-intervened quorum sensing in Gram-positive microscopic organisms (Harshad Lade, 2014).

2.13 Relevant studies carried out on quorum quenching (QQ)

Quorum quenching is a novel technique for biofouling control in MBR. It brought revolution in mitigating the most concerned problems of this technology. Yeon at al. (2009) deactivated the AHLs by hydrolyzing at lactone ring by lactonase and at acyl-amide linkage by acylase. They used porcine kidney enzymes I (EC 3.5.1.14) and found reduction in AHLs concentration, EPS production and delayed TMP rise in MBR having acylase. He prepared the magnetic enzymes carrier (MEC) on which enzyme (acylase) was immobilized to resolve the problem of stability of free enzymes and results showed that immobilized acylase perform better than the free moving acylase with same quantity.

Oh et al. (2011) worked on the isolation of quorum quenching bacteria (those produce QQ enzymes) and found four species out of which *Rhodococcus* and *Panibaccilus* stains found to be more effective, they encapsulated the *Rhodococcus* sp.BH4 in micro-porous membrane and submerged in MBR run parallel to the control MBR operated at similar filtration mode. TMP profile showed substantial difference between MBR with QQ bacteria encapsulated in membrane and control MBR.

Jahangir et al. (2012) found a suitable position of micro porous membrane having encapsulated QQ bacteria and they reported that biofouling was less in MBR having micro porous membrane in membrane tank as compared to the condition in which micro porous membrane was place in separated bio-tank with recirculated sludge and QQ activity was also dependent on the rate of recirculation.

Kim et al. (2012) worked on effect of QQ on microbial dynamics in MBR, QQ reduce the auto inducer producing microbial species which ultimately reduce the EPS production and results in less biofouling.

(SangRyoung Kim, 2012) In this study, quorum quenching microscopic organisms captured in free-moving globules were connected to the hindrance of biofouling in a MBR. Permeable microstructure cell ensnaring globules (CEBs) were readied by entangling quorum quenching microscopic organisms *(Rhodococcus* sp. BH4) into alginate dabs. The moderation of biofouling was credited to both physical (contact) and organic (quorum quenching) impacts of CEBs, the last being a great deal more vital. Results demonstrated that due to the CEBs with ensnared quorum quenching microorganisms, EPS generation from microbial cells in the biofilm was lower, and in this manner, empowered biofilm to quagmire off from the film surface more effectively.

Muhammad Faisal Siddiquia (2012) researched the counter quorum sensing movement of PBE from the Piper beetle was identified to relieve biofouling of layer in MBR. *Agrobacterium tumefaciens* strain NTL4 was utilized to decide the generation of AHLs in. The biocake displayed AIs movement, which demonstrated that QS was in pleasant association with film biofouling. PBE was affirmed to moderate layer biofouling through AIs generation restraint. It was likewise found that the expansion of PBE diminished the measure of EPS in biocake; while the expansion of HHL expanded the measure of EPS arrangement. In this manner discoveries of this study uncovered that PBE could be a novel operator to target AIs for film biofouling control.

Kim et al. (2013) prepared cell entrapping beads (CEBs) of sodium alginate having entrapped *Rhodococcus* sp.BH4, MBR setup installed for the analysis was of 1.6L batch type. This type of engineered quorum quenching mechanism was found to be most effective than others.

Cheong et al. (2014) inoculated the bacteria, *Pseudomonas* sp.1A1 in ceramic microbial vessel (CMV) and submerged these in MBR and compared the result with control MBR (C-MBR) and MBR with CMV having inactivated QQ bacteria. They found substantial reduction of AHLs concentration and EPS production in MBR having CMV with activated bacteria.

2.14 Electro Coagulation vs Chemical Coagulation

There are many negatively charged fine colloids (0.01 to 1µm) in Chemical coagulation in wastewater. Coagulation practice destabilizes this colloidal suspension. Coagulation reactions have various side reactions with other composites in wastewater which depend on the sewage characteristics. Chemical deterioration or destabilization is caused by coagulation of compounds chemically. These chemicals are added into the wastewater to form larger flocs through perikinetic flocculation (combination of fine particles in the size of 0.01 to 1 µm). Typical agglomirants include natural and synthetic organic polymers (anionic, cationic) or alum ($Al_2 (SO_4)_3$), ferric sulphate ($Fe_2 (SO_4)_3$), ferro sulphate ($FeSO_4$) and Ferric chloride. Ferric chloride and Alum are commonly used for wastewater treatment. Sludge production and nutrient removal in the coagulation-flocculation process was studied by Aguilar et al. (2002). Based on their research, approximately 100% of orthophosphate was removed and total phosphate was removed between 98.93% - 99.90%, while, the ammonia and nitrogen removal was very low (between 73.9 – 88.77 %). Aguilar and their team also showed that 41.6% of the sludge volume was reduced by using coagulants

Later, study on coagulant addition on nutrient removal and membrane fouling in the submerged membrane bioreactor (SMBR) was carried out by Song et al. (2008). They added two coagulants alum and ferric chloride into the aerobic tank of a pre-anoxic nutrient removal unit. As a result, they found that alum in a concentration of 200-500 (mg/l) is more effective in nutrients removal than ferric chloride. Through their experiments, 98% of phosphorus was removed.

2.15 Electrical double layer

Two main forces which are acting between colloids are the Van der Waals attraction forces and the electrical repulsion forces. Later forces are usually large enough to keep colloids apart. However, if the particles are given enough energy to overcome this repelling force, then the Van der Waals forces will dominate and bring the particles together. Therefore, if sufficient energy is supplied by heating or mixing, the colloids may begin to agglomerate. Van der Waals forces cannot be increased; if the electrical forces are reduced then the energy barrier can be decreased or totally removed. If colloidal particles are close enough together, Van der Waals forces will cause them to agglomerate, but the effect will be opposed by the electrostatic repulsion of the zeta potential. Most colloids are electrically charged, either positively or negatively. These charged colloids then attract ions in the solution of opposite charge to form a surrounding layer of counter-ions (opposite charge). This layer of counter-ions surrounding the colloids is called the ‘diffuse layer’. The surface charge of the colloid and the diffuse layer together form the ‘electrical double layer’, A large proportion of the counter-ions are situated at close distance to the colloid, called the ‘Stern layer’. The separating boundary between these two layers is referred to as the ‘Stern plane’ (Metcalf and Eddy, 2003).

As colloidal particles approach one another, no repulsive force acts between them, and the Van der Waals forces are allowed to bring colloids together. To reduce the electrical forces, zeta potential must be lowered. This may be achieved by adding a higher concentration of ions with a higher charge. These additional ions may replace to the existing counter-ions and they can also reduce the thickness of the diffuse layer. This in turn reduces the Stern or zeta potential and the repelling force, causing stabilization.

2.16 Description of Electrocoagulation

There are several physicochemical techniques for wastewater treatment like ion-exchange, filtration, chemical precipitation, etc. Additional chemical addition is a common aspect for all of them. Introducing the electrocoagulation (EC) method in activated sludge compartment is one method to increase the functioning of the wastewater treatment system. It is not necessary to add chemical compounds in electrochemical technology like electrofiltation (Li et al. 2010) and electrocoagulation (Holt 2005). Whereas, electrocoagulation is snatching the attention due to its lesser capital cost and better performance in comparison with electrochemical technology. In electrocoagulation process, colloidal particles can be removed by adsorption to the metallic hydroxide or by binding with the opposite charged ions and less sludge is produced. Electrocoagulation process starts with electrolysis reactions around the anode (aluminum/iron) area to produce flocs of hydroxide as coagulants once the current is provided.

Advantages of electrocoagulation as compared to the chemical coagulation includes:

- No addition of liquid chemical coagulant compounds
- Less consumption of Alkalinity
- The operation is easy to perform
- No coagulation agents needed in EC process, Hence less operating cost along with less sludge production
- EC with six iron plates electrodes is cost effective technology (Irdemez et al. 2006),
- Less generation of by-products in the effluent as well as wasted solids (BaniMelhem and Elektorowicz 2010)

Colloids and Microbial populations are usually negatively charged; therefore, their behavior can be changed in the presence of the electrical field whether by continuous or intermittent electrical field. They diverted away from the surface of the membrane hence reducing filtration flux and membrane fouling (Wei et al. 2012). The electrical field also have effect on activated sludge constituents. The sludge produced by electrocoagulation consists of large flocs with less bounds of water. As a significance, the volume of produced sludge becomes unexceptional (Mollah et al. 2001).

2.17 Factors affecting EC process

Chemical characteristics of the solute, conductivity, temperature, the pH value (should be in a range of 6 to 8) and formation of sludge plays an important role in electro coagulation process (Ilhan et al. 2008).

2.17.1 Current density

The current density an important factor in electrocoagulation procedure. It is calculated from surface area of anode (m^2) and current (A). Current density represents the number of anode ions (aluminum or iron). The number of these ions increases as current density increases. When high current density is applied in the unit the current efficiency (CE) also reduces. High current density can also reduce the removal efficiency of the treatment. Aluminum electrodes have current efficiencies around 130%, while, iron electrodes have 100% current efficiency. This shows that due to oxidation of chlorine anions, aluminum sheets oxidize over time. In order to have finest and optimum performance, the current density should be in the range of 10 to 150 A/m2; which is suitable for a long period of time, unless the periodical cleaning of the electrode's surface is needed. (Chen 2004).

2.17.2 Conductivity

Sodium chloride (NaCl) is sometimes added into the water or wastewater for increasing the conductivity. NaCl is used to increase the ionic influence in moving the electric charge. Sodium chloride also reduces the energy consumption because it increases conductivity (Chen 2004).

2.17.3 pH

The level of pH influences the EC process from two viewpoints: solubility and current efficiency (CE). Aluminum anodes have higher current efficiency (CE) in both acidic and basic environments than in neutral situations. CO^2 is oversaturated in acidic condition while, in alkaline condition more OH^- can release from cathode.

2.17.4 Temperature

Chen (2004) found that in temperatures higher than 60^O C, current efficiency might increase, and thus power consumption decreases. On the other hand, the aluminum oxide layer formed on the surface of anode is destroyed in higher temperatures due to high current efficiency. At high temperatures $Al\ (OH)_3$ shrinks and makes denser flocs, having more affinity to settle down on the electrode’s surface.

2.17.5 Power supply and exposure time

Other important parameters for the electrocoagulation process are power supply and exposure time. The power might be given as continuous mode (without any OFF/ON timer) or intermittent mode. The negative point for continuous mode is oxidation of anode and passivation of cathode. Therefore, the current should be in intermittent mode i-e first OFF, and then ON after some minutes. High exposure time might increase the amount of sludge production while low exposure time might decrease the efficiency of EC tasks, while. Therefore, exposure time is very essential for the performance of Electrocoagulation. Previously, many researchers studied the direct current exposure time (Ibeid (2012); Hasan et al. (2012); Wei (2012), Bani-Melhem and Elektorowicz (2010, 2011)).

2.18 Mechanism of EC

Electrocoagulation process depends on the current density. Lower current density is required when separation units (such as sedimentation tank, sand/coal filtration and membrane filtration) work with electrocoagulation processes. In contrast, higher current density is needed for separation of coagulants particles, particularly in a flotation process. The mechanisms of electrocoagulation can be precised as follows (Li et al. 2010):

- **Electrophoresis**: when current is provided in the reactor, charged particles move towards the oppositely charged electrodes to make flocs.
- **Electro-osmosis and Electro-migration**: movement of water in through membrane or other porous structure in a reactor due to the electrical gradient forces.
- Releasing the materials of cathode (hydroxyl ions) and anode (metallic cations) over time and reaction of these compounds with other elements which already existed in the solute.
- The metallic ions which are realized from anode reacts with OH- ions.
- Long chain of metallic hydroxides compounds can clutch undesirable elements such as colloids.
- Oxidation of unwanted organic compounds.
- **Electro-filtration**: removing the unwanted compounds by sticking to bubbles which are produced during the EC process.

2.19 Reactions occurred around Electrodes

When current is applied, some of the cationic monomeric forms such as aluminum ions releases from anode into the reactor. Due to which flocculation of colloidal particles occurs. (Bouamra et al. 2012). By oxidation of water, oxygen gas and hydrogen ions are produced around the anode zone. By reduction process hydroxide ions and hydrogen gas are produced around the cathode zone.

$$2H_2O + 2e^- \rightarrow H_2 + 2OH^- \quad (2.1)$$

$$Al(s) \rightarrow Al^{3+} + 3e^- \quad (2.2)$$

$$Al^{3+} + 3H_2O \rightarrow Al\,(OH)_3 + 3H^+ \quad (2.3)$$

$$nAl(OH)_3 \rightarrow Al_n(OH)_{3n} \quad (2.4)$$

Based on level of pH several reactions are carried out in solution (Eq. 2.5 to 2.8) (Hasan, 2011)

$$Al^{3+} + H_2O \rightarrow H^+ + Al(OH)^{2+} \quad (2.5)$$

$$Al(OH)^{2+} + H_2O \rightarrow H^+ + Al(OH)_2^+ \quad (2.6)$$

$$Al(OH)_2^+ + H_2O \rightarrow H^+ + Al(OH)_3 \quad (2.7)$$

In a reactor, aluminum ions react with free hydroxide ions to typically make forms like: $Al(OH)^{+2}$, $Al(OH)_2^{+1}$, and $Al\,(OH)^-_4$. These compounds can be transformed into the polymeric form like: $Al_8\,(OH)^{+1}_{20}$, $Al_{13}\,(OH)^{+5}_{31}$. These processes continue until long chain of aluminum hydroxides are produced (Eq. 2.8).

2.20 Membrane electro bioreactor

The electro-bioreactor is a modern technology for reducing mineral and organic contaminants, and nutrients in wastewater. In general, electro-bioreactor's performance depends on biological parameters, wastewater characteristics and operating conditions. SMEBR is a hybrid unit for membrane processes, biological activities and electro kinetic process

3 Methodology

The research methodology was divided into two phases, Batch scale setup and continuous scale setup. Furthermore, phase one includes two stages including four electrical exposure modes in stage one along with control reactor and two exposure modes selected from stage one with and without quorum quenching bacteria in stage two. Phase two of continuous reactors was operated on the selected electrical exposure mode from phase one and quorum quenching membrane bioreactor.

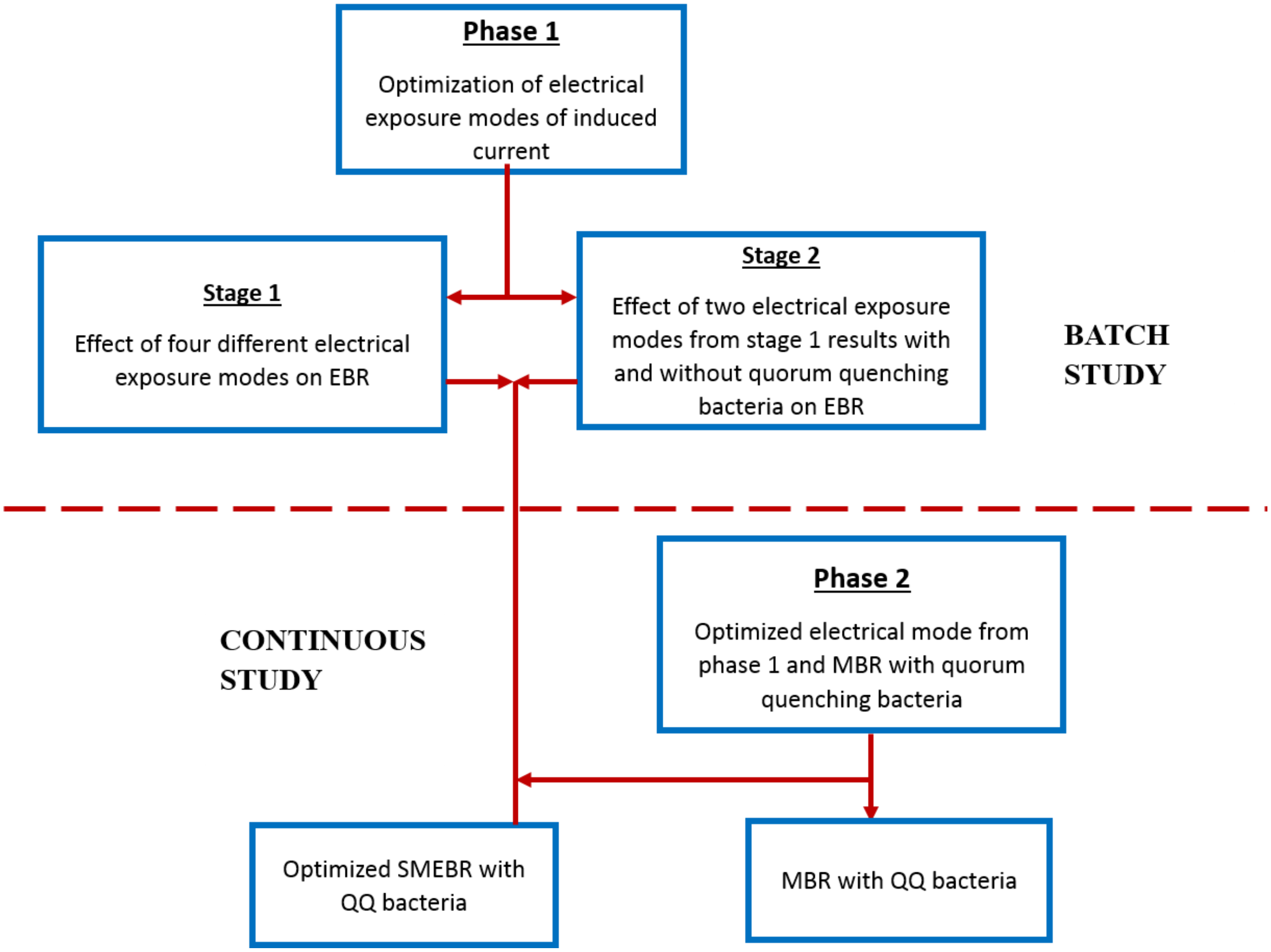

Figure 3.1 Flow sheet description of experimental study

3.1 Phase 1

Batch scale study consists of two stages to determine the effective intermittent electrical exposure mode for continuous scale study that improves sludge properties by improving microbial activity. Stage 1 consists of four electrical exposure modes and control reactor (Figure 3.1). Stage 2 of phase 1 includes the optimized electrical exposure modes with and without quorum quenching bacteria on sludge and microbial activity. Operating parameters for visualizing the effective microbial activity includes Mixed Liquor Suspended Solids (MLSS), Sludge Volume Index (SVI), Chemical Oxygen Demand (COD), pH, Particle size distribution (PSD), Time to Filter (TTF) and soluble Extracellular Polymeric Substances (EPS). Electrical operating parameters includes electrical exposure mode (ON, OFF) and current density. Operating period of both stages of phase 1 was 35 days. (Figure 3.2)

PHASE 1

Batch scale study

Objective: Optimization of the intermittent supply of the induced current

STAGE 1 (35 days)

Study the effect of electrical exposure modes on microbial activity.

Electrical current density = 10 A/m^2

Electrical exposure modes
Control
5 min ON – 10 min OFF
5 min ON – 20 min OFF
5 min ON – 30 min OFF
5 min ON – 40 min OFF

Observed parameters

1. Mixed Liquor Suspended Solids (MLSS)
2. Sludge Volume Index (SVI)
3. Chemical Oxygen Demand (COD) removal
4. pH
5. Particle size distribution (PSD)
6. Time to Filter (TTF)
7. Extracellular Polymeric Substances (EPS)

STAGE 2 (35 days)

Study the effect of optimized exposure modes along with QQ beads on microbial activity.

Electrical current density= 10 A/m^2

Electrical exposure modes
Control
5 min ON – 30 min OFF (QQ)
5 min ON – 40 min OFF (QQ)
5 min ON – 30 min OFF
5 min ON – 40 min OFF

Observed parameters

1. Mixed Liquor Suspended Solids (MLSS)
2. Sludge Volume Index (SVI)
3. Chemical Oxygen Demand (COD) removal
4. Particle size distribution (PSD)
5. Time to Filter (TTF)
6. Extracellular Polymeric Substances (EPS)

Figure 3.2 Methodological approach of PHASE 1

3.2 Experimental setup

3.2.1 Phase 1(stage 1)

Experimental structure of Phase 1 (stage 1 and 2) contains 1.3-L batch bioreactors. These bioreactors consist of porous stainless steel cathode and aluminum anode. Aerobic conditions and DO was maintained through diffusers placed at the bottom of reactors. Air was supply with the help of aerator at an air flow rate of 45 L/min. Electrodes were adjusted at a defined space of 5 to 10 cm for affective electro kinetics. (Figure 3.2). For proper mixing of activated sludge, electrodes were kept to be porous. Sludge was collected from NUST MBR wastewater treatment plant and immediately used to prevent any changed to sludge and microbial properties. Same mixed liquor suspended solids (MLSS) concentration of 5000 mg/L was used initially. Current density of 10-12 A/m^2 was used with four electrical exposure modes (5'ON-10'OFF, 5'ON-20'OFF, 5'ON-30'OFF and 5'ON-40'OFF) in this study. Phase 1 consists of one control reactor and four reactors of different electrical modes placed side by side (Figure 3.3).

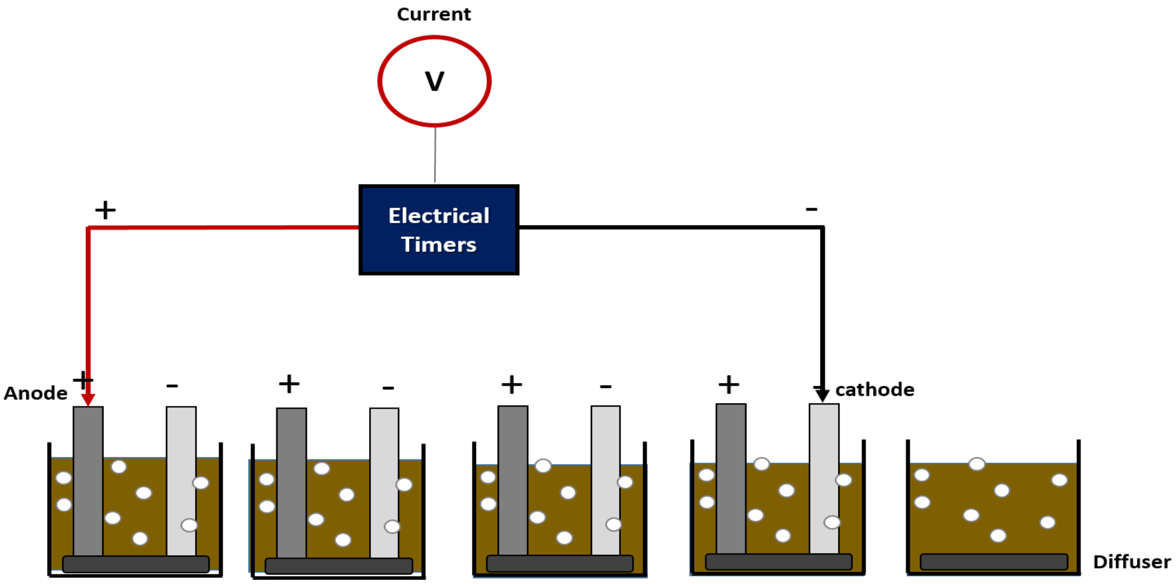

Figure 3.3 Batch Setup

Table 3.1 working conditions of batch setup phase 1 (stage 1)

Parameters	**Values**	**Current density**	**Exposure modes**
Working volume	1.3 L	**10-12 A/m**	Control
HRT	12 hr.		5 min ON – 10 min OFF
SRT	15 and 20 d		5 min ON – 20 min OFF
OLR	2 kg/m^3/d		5 min ON – 30 min OFF
			5 min ON – 40 min OFF

3.2.2 Phase 1 (stage 2)

The similar stage 1 electro-bioreactors were used in this step. Two electrical exposure modes were chosen for this experiment 5’ON-30’OFF and 5’ON-40’OFF. These modes were further divided into with and without quorum quenching electro-bioreactors. Same value of current density 10-12 A/m^2 was used as in stage 1. MLSS in stage 2 was initially 5000 to 8000 mg/L. Control reactor with no current was placed side by side for comparison. This run was activated for a period of 35 days. Same synthetic water was added in reactors for better microbial activity.

Table 3.2 Working conditions of batch setup phase 1 (stage 2)

Parameters	**Values**	**Current density**	**Exposure modes**
Working volume	1.3 L	**10-12 A/m**	Control
HRT	12 hr.		5 min ON – 30 min OFF (QQ)
SRT	20 d		5 min ON – 40 min OFF (QQ)
OLR	2 kg/m^3/d		5 min ON – 30 min OFF
			5 min ON – 40 min OFF

3.3 Synthetic wastewater composition

Synthetic wastewater structure includes glucose 1000 mg/L, ammonium chloride 382 mg/L, Potassium di-Hydrogen phosphate 47.7 mg/L, Calcium Chloride 9.73 mg/L, Magnesium Sulfate 9.73mg/L, Ferric Chloride 1 mg/L and sodium bicarbonate (NaHCO3) 120 mg/L for optimum function of activated sludge.

Table 3.3 Synthetic Wastewater Composition

Chemicals	**Formula**	**Quantity (mg/L)**
Glucose	$C_6H_{12}O_6.H_2O$	1000 mg/L
Ammonium Chloride	NH_4Cl TH	382
Potassium di-Hydrogen Phosphate	$KH_2\ PO_4$	47.7
Calcium Chloride	$CaCl_2$	9.73
Magnesium	$MgSO_4\ .7H_2O$	9.73
Ferric Chloride	$FeCl_3$	1
Sodium bicarbonate	$NaHCO_3$	120

3.4 Phase 2

Two lab-scale MBR setup with a 19 L of working volume were installed in Water and Wastewater Laboratory, IESE-NUST as shown in Figures 3.3 and 3.4. Functional conditions are presented in Table3.1. Sludge was taken from NUST MBR wastewater treatment plant and used immediately. MBR was operated on 13 hr. HRT and 20 days SRT and worked on optimized 8-minute filtration and 2-minute relaxation mode. Air was supplied with the help of aerator at an air flow rate of

8L/min with the help of air compressor having 6 diffusers in each tank. To maintain the MLSS excessive sludge was wasted daily at an SRT f 20 days. Relay units and water controller level was used to maintain the water level in bio reactors. Direct current supply linked with electrical timer was used to facilitate the needed exposure mode (ON-OFF) and current density. Peristaltic pumps were used to extract the liquid out of membrane at a constant flow rate. Operational conditions are shown in Table. 3.4

Table 3.4 Working Parameters for lab scale MBR

Parameter	**Value**
Working volume	19 L
HRT	13.5 hr.
SRT	20 d
Flux	20 L/m2/hr.
Current density	10-12 A/m
OLR	1.8 kg/m^3/d

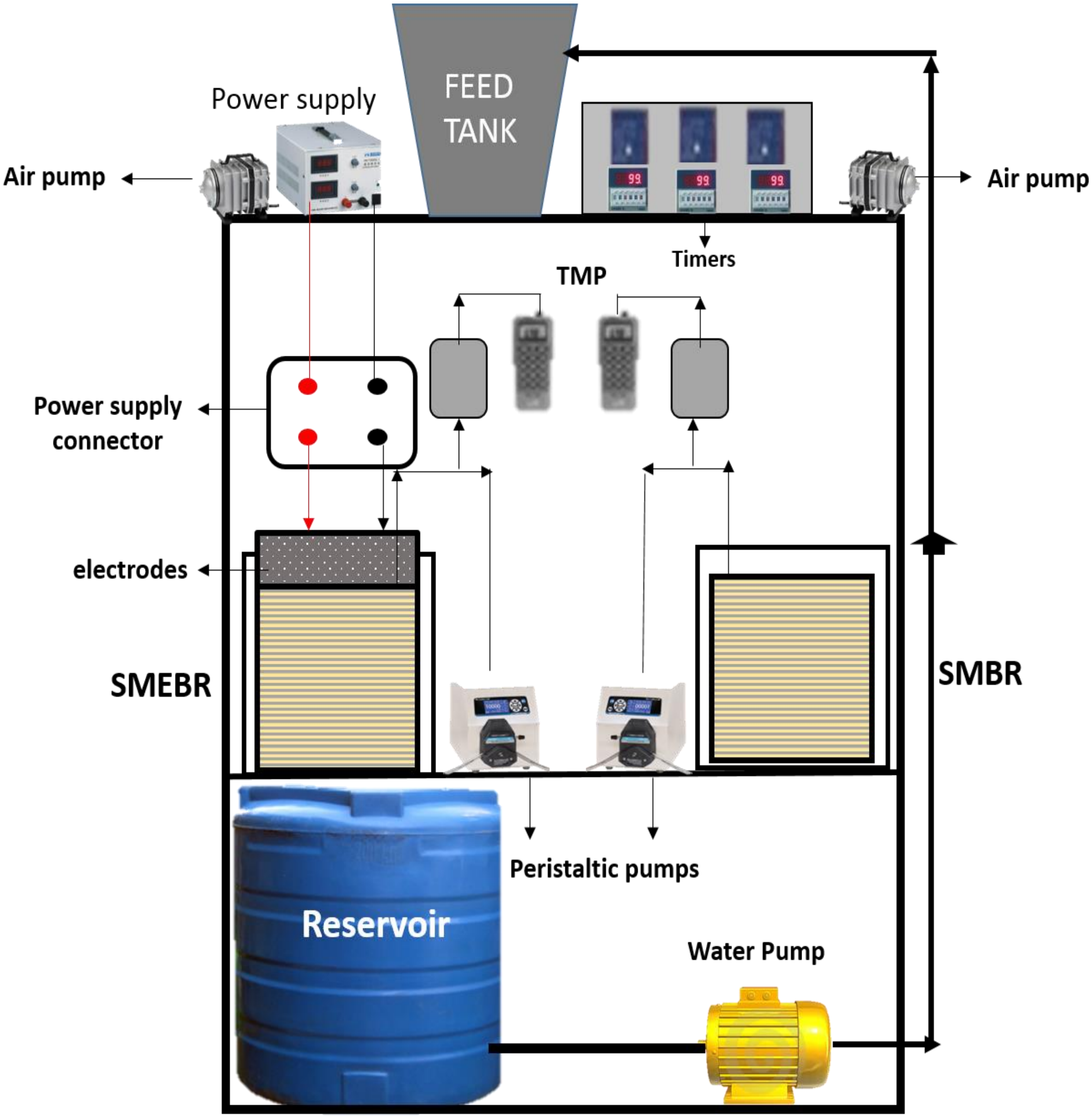

Figure 3.4 Schematic diagram of lab scale MBR & SMEBR

3.5 Membrane characteristics

Hollow Fiber Poly Vinyl di Fluoride membrane module was developed by Mitsubishi Rayon Japan. Fibers were connected horizontally to the module. If membrane module is outside-in that means water flows from outside of the membrane to the inside.

Table 3.5 Membrane characteristics

Manufacturer	Mitsubishi Rayon Engineering Co. Ltd., Japan
Material of membrane	PVDF
Pore size	0.05 µm
Filtration area	0.07 m^2
Suction Pressure	10-30 KPa
Temperature	15-35 ^{O}C

3.6 Analytical Methods

3.6.1 Resistance analysis

To evaluate the fouling stamina of Membrane, resistance analysis in series (RIS) model was used

$$Rt = \frac{\Delta P}{\mu . J . ft}$$

$$Rt = Rc + Rp + Rm$$

Where,

Rt = total hydraulic resistance (1/m)

ΔP = TMP (Pa)

μ = (Pa.s) permeate dynamic viscosity

J = operational flux of permeate ($m^3/m^2/s$)

ft = temperature correction factor corresponds to 20◦C, $ft = e^{-0.0239(T-20)}$

Rp = pore blockage resistance (1/m)

Rc = cake layer resistance (1/m)

Rm= intrinsic membrane resistance (1/m)

Rc developed by the development of cake on the membrane surface, Rp was produced due to minute microbial flocs which trapped between the membrane pores. Rm was determined after chemical cleaning of membrane. For $Rm + Rp$, cake layer on membrane surface was first detached and then membrane was placed in deionized (DI) water after which TMP and flux was measured. Rc was calculated by subtracting Rm+Rp from Rt, (Wang et al., 2009).

Table 3.6 Methodology for different Parameters

Sr #	**Components**	**Analytical method**
1	Mixed Liquor Suspended Solids	APHA et al., 2012
2	Mixed Liquor Volatile Suspended Solids	APHA et al., 2012
3	Sludge Volume Index	APHA et al, 2012
4	Chemical Oxygen Demand (COD)	Close reflux titrimetric method (APHA et al., 2012)
5	Particle Size Distribution (PSD)	Particle size distribution Analyzer
6	Time to filter (TTF)	Method 2710H
7	Extracellular Polymeric Substances (Protein) EPSp	folin method (Lowry, 1951)

8	Extracellular Polymeric Substances (Carbohydrates) EPSc	phenol/sulfuric-acid method (Dubois, 1956)
9	Ammonia-nitrogen	Colorimetry method
10	Phosphate	UV-Spectrophotometer

3.6.2 Membrane fouling

Membrane fouling is measured through the changes of transmembrane pressure over time. Initially the TMP of MBR and SMEBR were from 2 to 3 kPa. The increase in TMP causes reduction in membrane flux. TMP increases over time because of gathering of sludge particles and microbial flocs on the surface of membrane. Once the membrane fouls due to maximum increase in TMP, the membrane was cleaned physically, chemically and acidically.

$$\text{Fouling Rate} = \frac{TMP\ (KPa)}{Time\ (d)}$$

4 Results and Discussion

4.1 Phase 1

Results of this stage was based on categorizing the different exposure modes with constant current density of 10-12 A/m^2 that can cause the effective removal of biofouling of membranes in wastewater treatment by maintaining the microbial activity.

4.1.1 Current density vs Mixed Liquor Suspended Solids

Figure 4.1 showed the variations in Mixed Liquor Suspended Solids (MLSS) over time period at different exposure modes and same current density of 10-12 A/m^2. It could be clearly seen that MLSS of 5’ON-10’OFF and 5’ON-30’OFF increased with this current density. In case of 5’ON-10’OFF, it increased from 5000 to 11000 mg/L and from 5000 to 9000 mg/L in case of 5’ON-30’OFF respectively. No significant increase in MLSS was observed under normal conditions i-e with no current provision.

At lesser time OFF exposure mode, more aluminum ions produced and dissolved in solution due to which an overall increase in MLSS was observed in 5’ON-10’OFF current exposure mode. This high increase in MLSS was because of inorganics produced due to electro dissolution of aluminum anode and chemical sludge. Similar trend was observed during RUN 2 (Control, 5’ON-30’OFF (QQ), 5’ON-40’ OFF (QQ), 5’ON-30’OFF, 5’ON-40’OFF) where no significant increase in MLSS was observed in control reactor, where as there is an important increase in MLSS of all other current exposure modes among which the MLSS of 5’ON-30’OFF and 5’ON-30’OFF (QQ) was more significant.

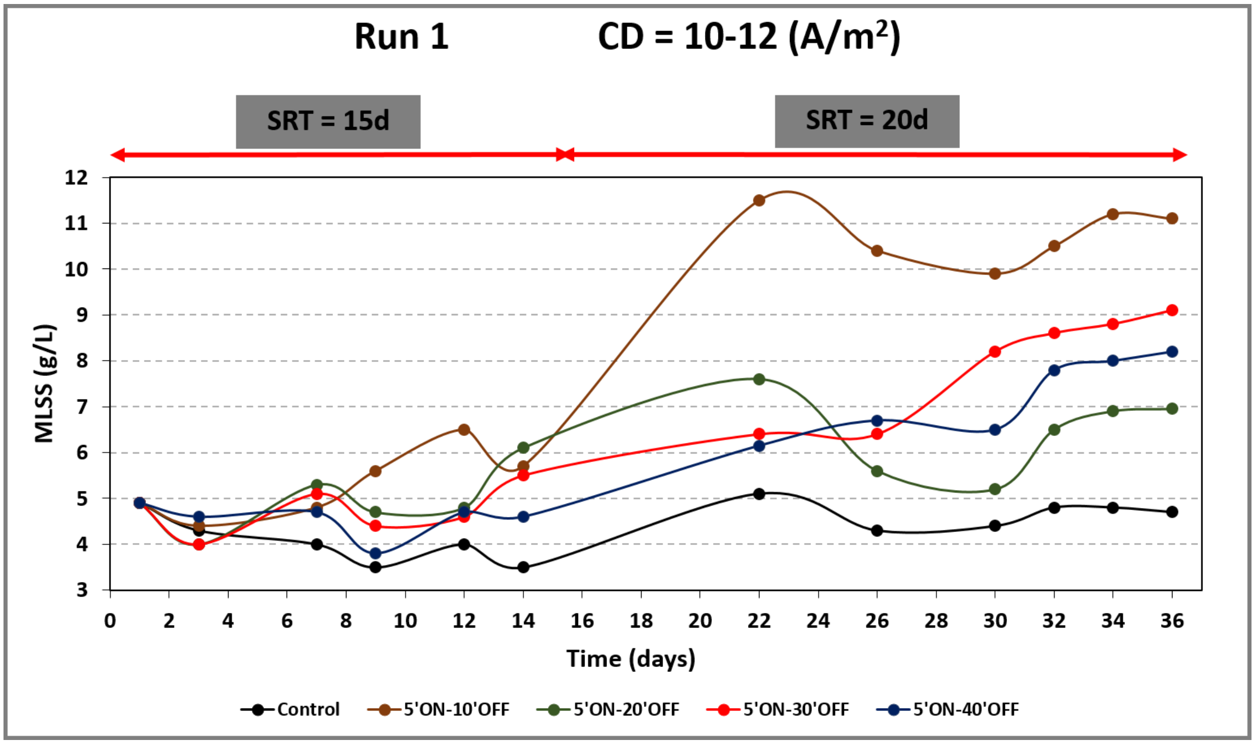

Figure 4.1 MLSS at different Electrical Exposure Modes

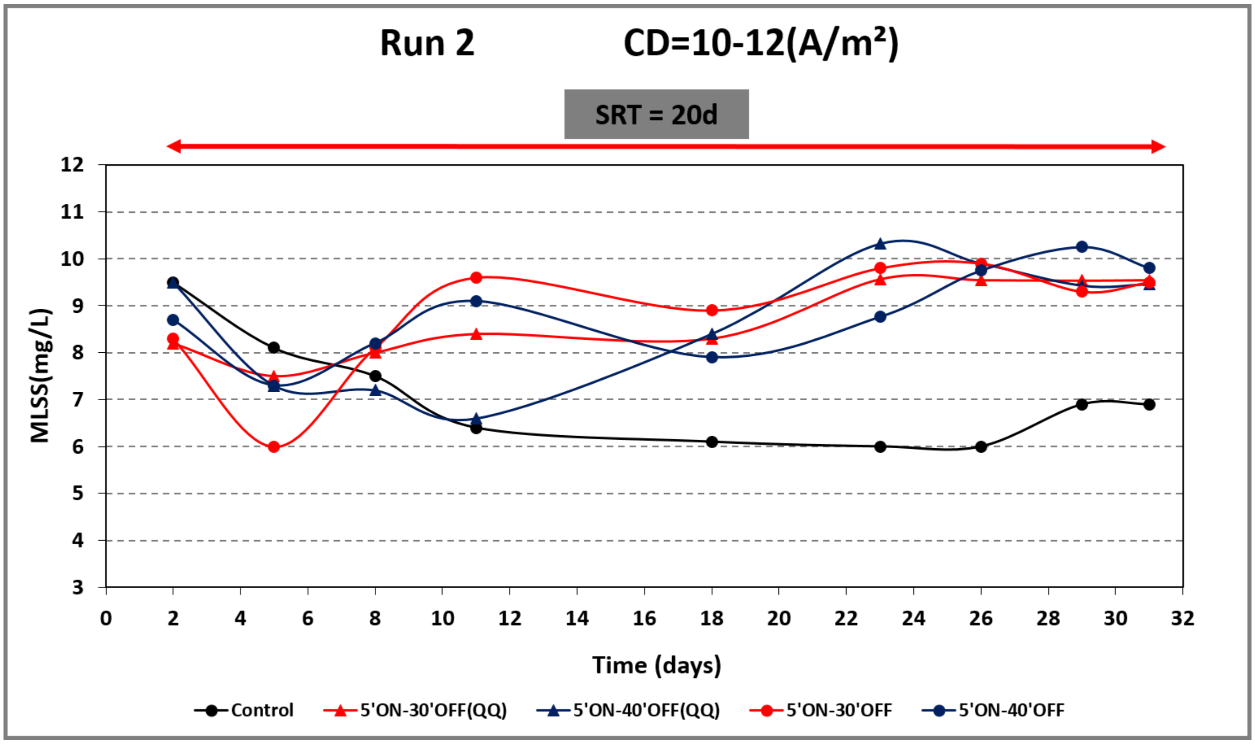

Figure 4.2 MLSS at different Electrical Exposure Modes

4.1.2 Current density vs COD

Changes in COD values of different exposure modes with same current density is shown in figure. COD removal of all electrical modes 5’ON-10’OFF, 5’ON-20’OFF, 5’ON-30’OFF AND 5’ON-40’OFF and control was above 85%; among which removal efficiency of 5’ON-30’OFF was further noticed to be above 95% at the end of 36 days operational period under current density 10-12 A/m^2. In RUN 2 (Control, 5’ON-30’OFF (QQ), 5’ON-40’ OFF (QQ), 5’ON-30’OFF, 5’ON-40’OFF), COD removal of 5’ON-30’OFF (QQ), 5’ON-40’ OFF (QQ), 5’ON-30’OFF was observed above 90% whereas, COD removal percentage of control and 5’ON-40’OFF was below 90%.

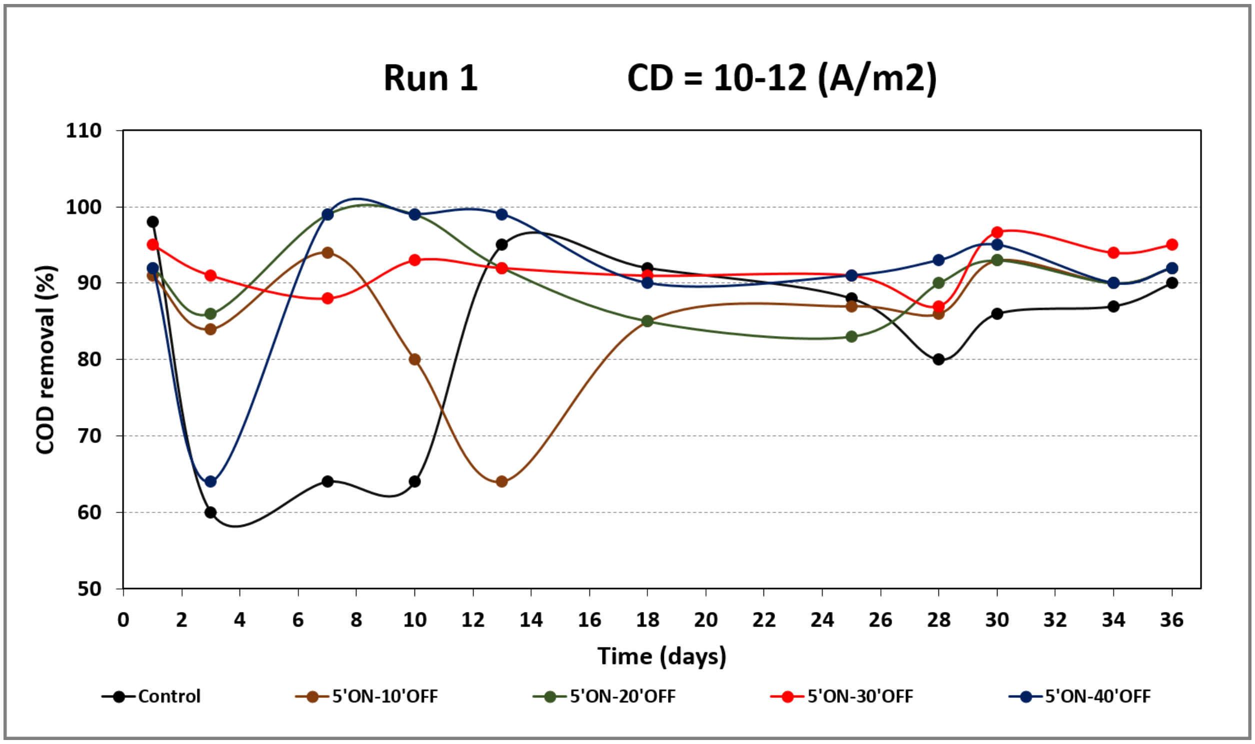

Figure 4.3 COD at different Electrical Exposure Modes

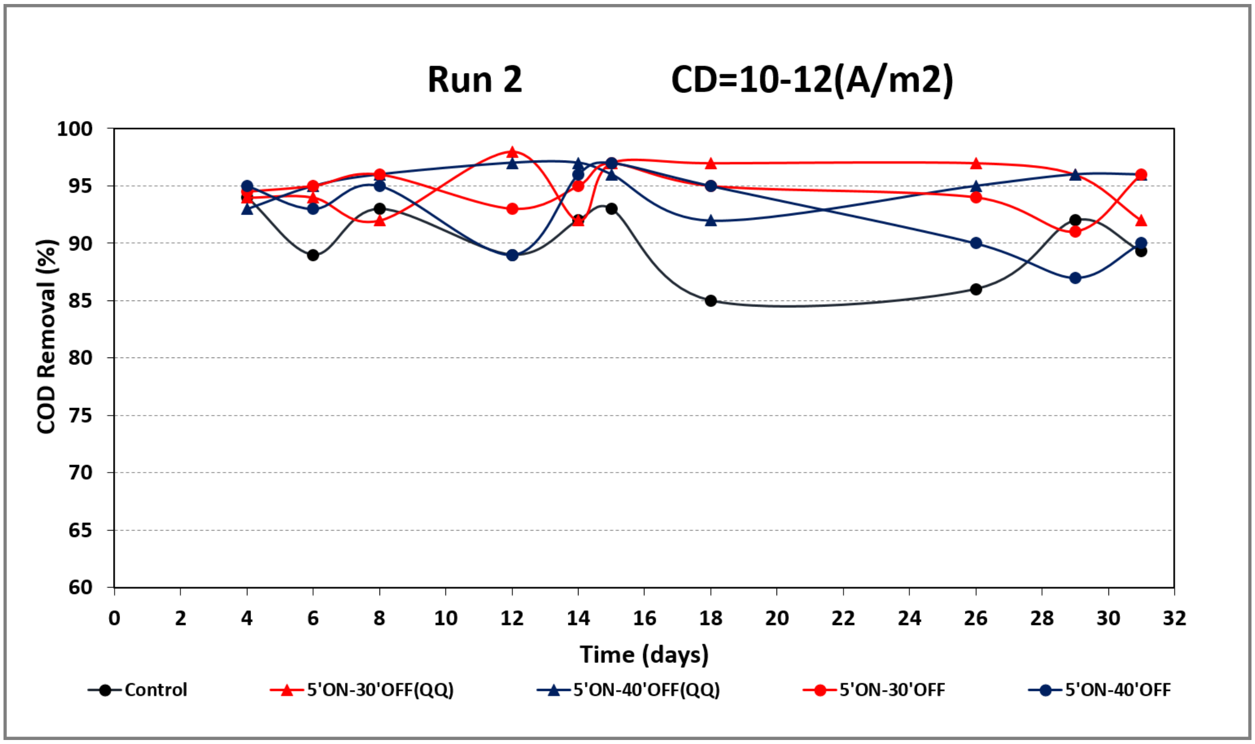

Figure 4.4 COD at different Electrical Exposure Modes

4.1.3 Current density vs sludge volume index

Between the SVI range of 50-150 ml/g, sludge usually settles more slowly due to which it traps more particulate material and forms a uniform cover before it settles down. Through the study it has been noticed that SVI of all electrical modes are in the range of 50-150 mL/g at the end of 36 days' operational period under current density 10-12 A/m^2. The continuously dropping SVI until 36th day of operation means that the sludge could be better dewatered as it become more settleable. SVI results of first run (5'ON-10'OFF, 5'ON-20'OFF, 5'ON-30'OFF AND 5'ON-40'OFF and control) are shown in figure. It is observed that SVI of 5'ON-10'OFF, 5'ON-30'OFF, 5'ON-40'OFF and control was below 100 ml/g whereas SVI of 5'ON-20'OFF was observed above 100 ml/g. On the other hand, SVI of all the current exposure modes of run 2 was observed below 100 ml/g.

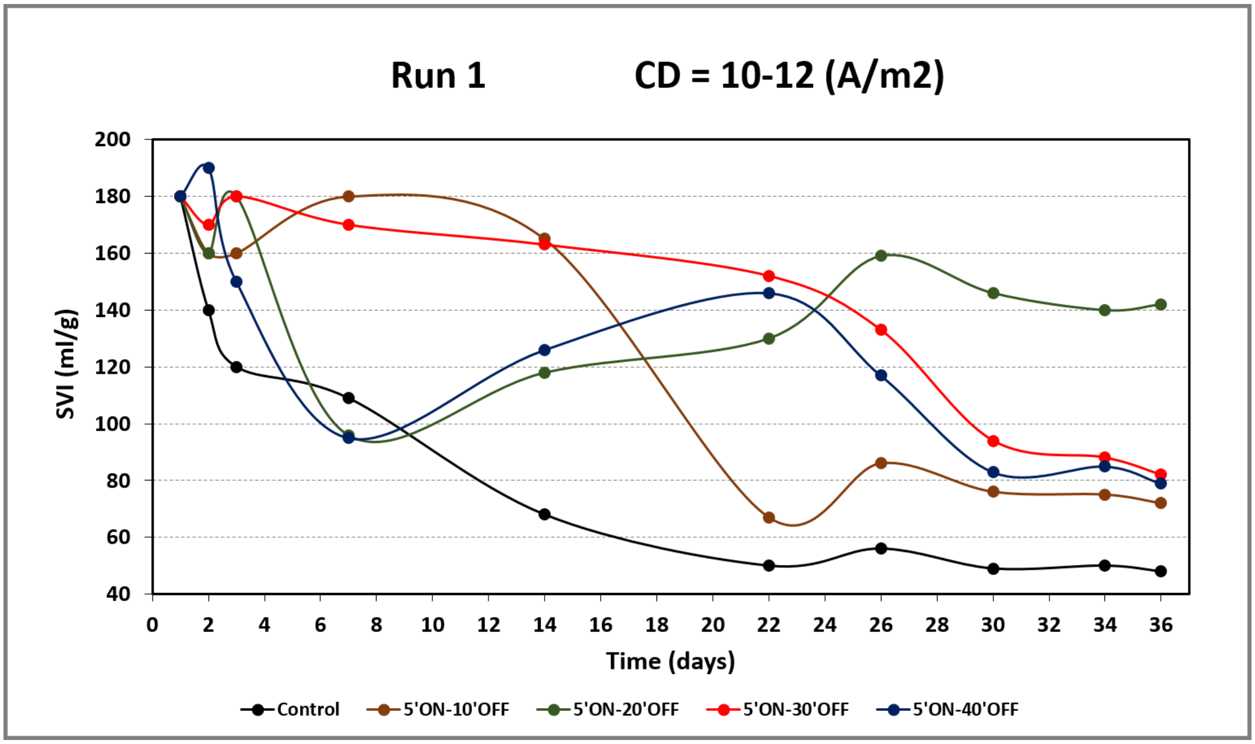

Figure 4.5 Run 1 SVI at different Electrical Exposure Modes

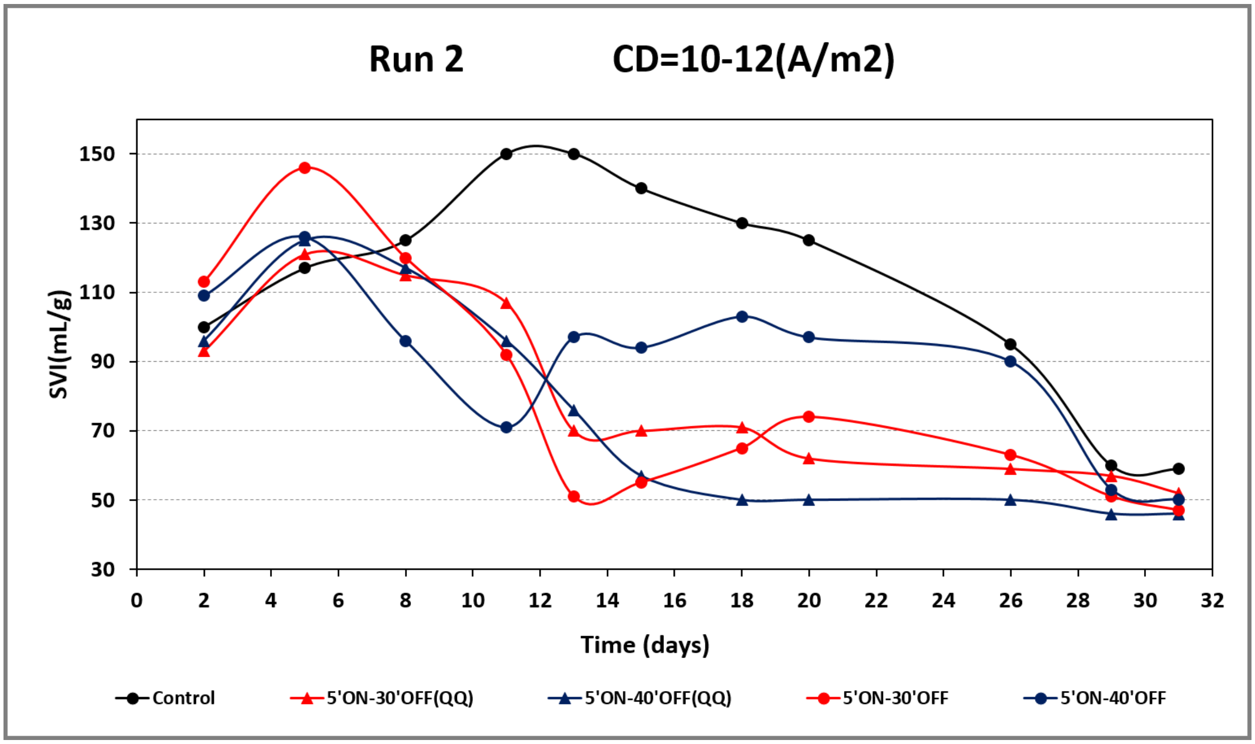

Figure 4.6 Run 2 SVI at different Electrical Exposure Modes

4.1.4 Current density vs pH

In electrocoagulation, number of released H^+ ions around anode zone increases while hydroxide ion production increases around cathode, due to which pH remains acceptable throughout the study. Literature shows a slight increase in pH at higher current densities around 40-60 A/m^2 (Ibeid, 2011). In this study pH was found to be between 6.5 to 7.5 throughout the study. While sodium bicarbonate was added in small amounts to further equalize pH.

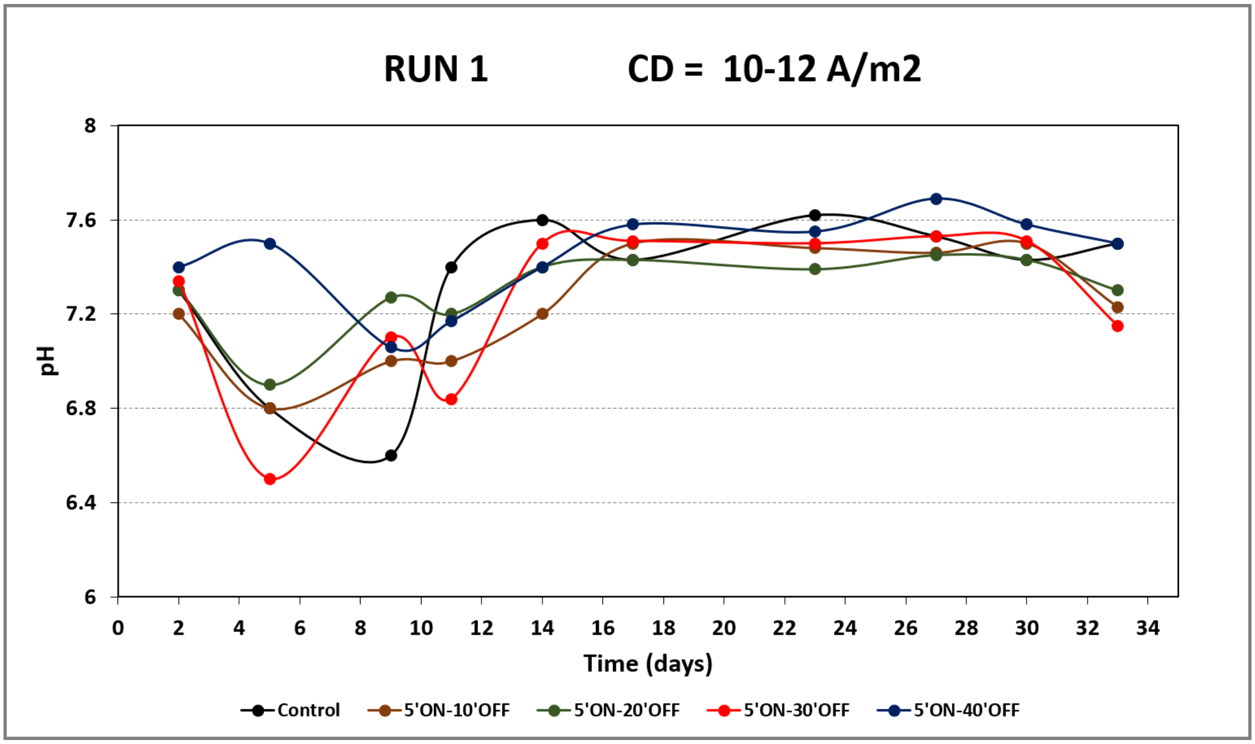

Figure 4.7 Run 1 pH at different Electrical Exposure Modes

4.1.5 Current density vs Particle Size Distribution

Figure 4.8 presents difference of particle size diameter at different current exposure modes over time. It could be concluded that the decrease in floc size was 50, 37.4, 37, and 29 at 5’ON-10’OFF, 5’ON-20’OFF, 5’ON-30’OFF AND 5’ON-40’OFF current exposure modes respectively. Among these the major percentage reductions were observed in 5’ON-10’OFF and 5’ON-30’OFF. This is because of electro osmosis phenomena in Electro Bioreactor in which negative charged particles of sludge were enclosed by positive charged particles. When current was applied in the solution these positive ions moves towards cathode by repelling water molecules out of the sludge (Hasan, S (2011). This would result in overall decrease in floc size particle. For lesser time off current exposure mode (5’ON-10’OFF) at 10-12 A/m^2, there was significant decrease in particle size while for 5’ON-20’OFF and 5’ON-30’OFF, the overall reduction in PSD was almost equal and for greater time OFF the reduction in PSD decreased. Whereas different results were observed in Run 2 where there was a slight increase in mean PSD of current exposure modes than control reactor. These results followed the literature in which there was a significant increase in PSD up to 19th day of operation and then major reduction was observed due to electro osmosis phenomena (Giwa et al., 2015).

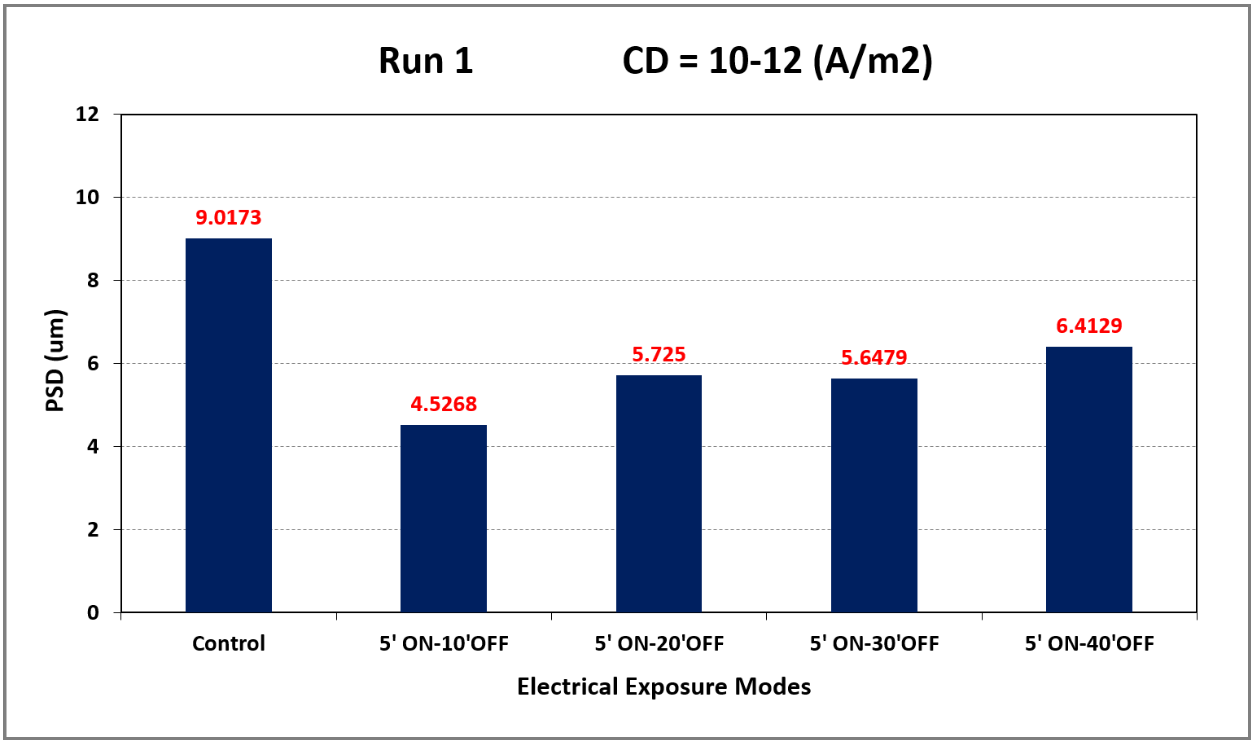

Figure 4.8 Run 1 PSD at different Electrical Exposure Modes

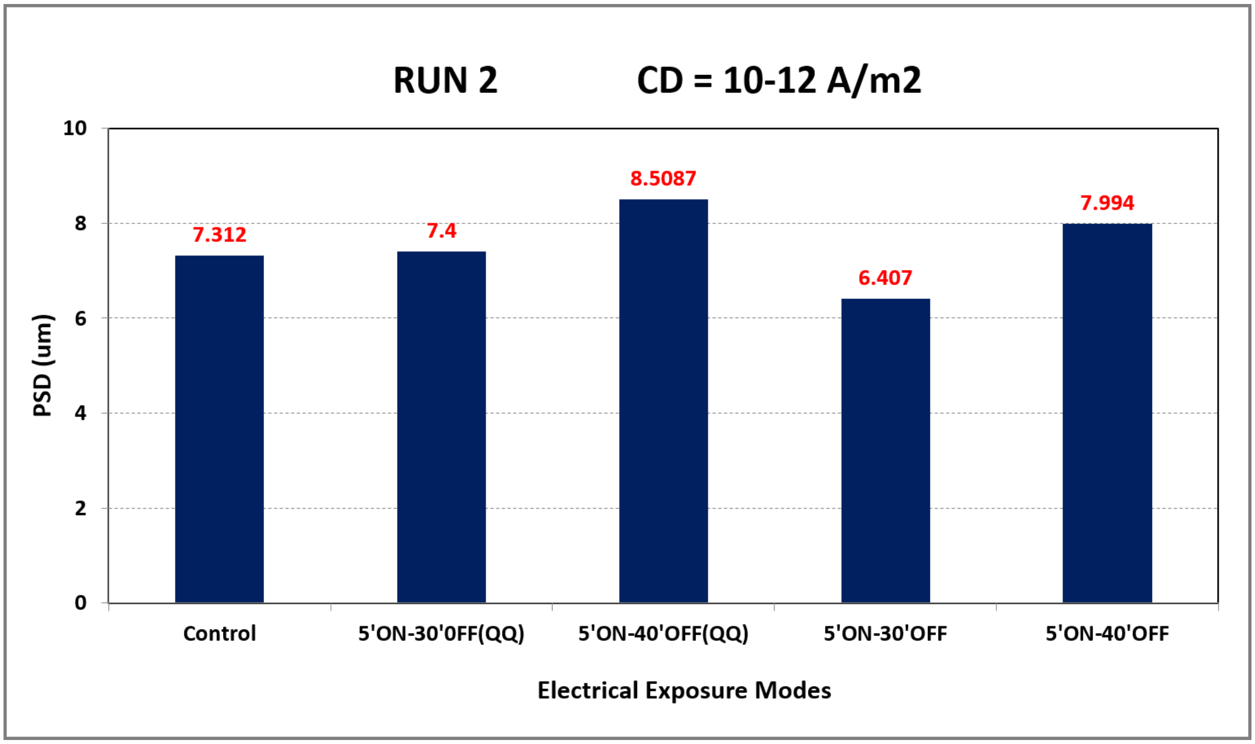

Figure 4.9 Run 2 PSD at different Electrical Exposure Modes

4.1.6 Current density vs Time to Filter

TTF is basically used to monitor the quality of sludge. The major reduction in TTF observed in run 1 was 22% of 5’ON-30’OFF compared to startup value. Whereas there was 1%, 31%, 16% and 15% decrease in TTF of 5’ON-10’OFF, 5’ON-20’OFF, 5’ON-30’OFF AND 5’ON-40’OFF respectively. These results depict the improvement in sludge filterability of 5’ON-20’OFF and 5’ON-30’OFF.

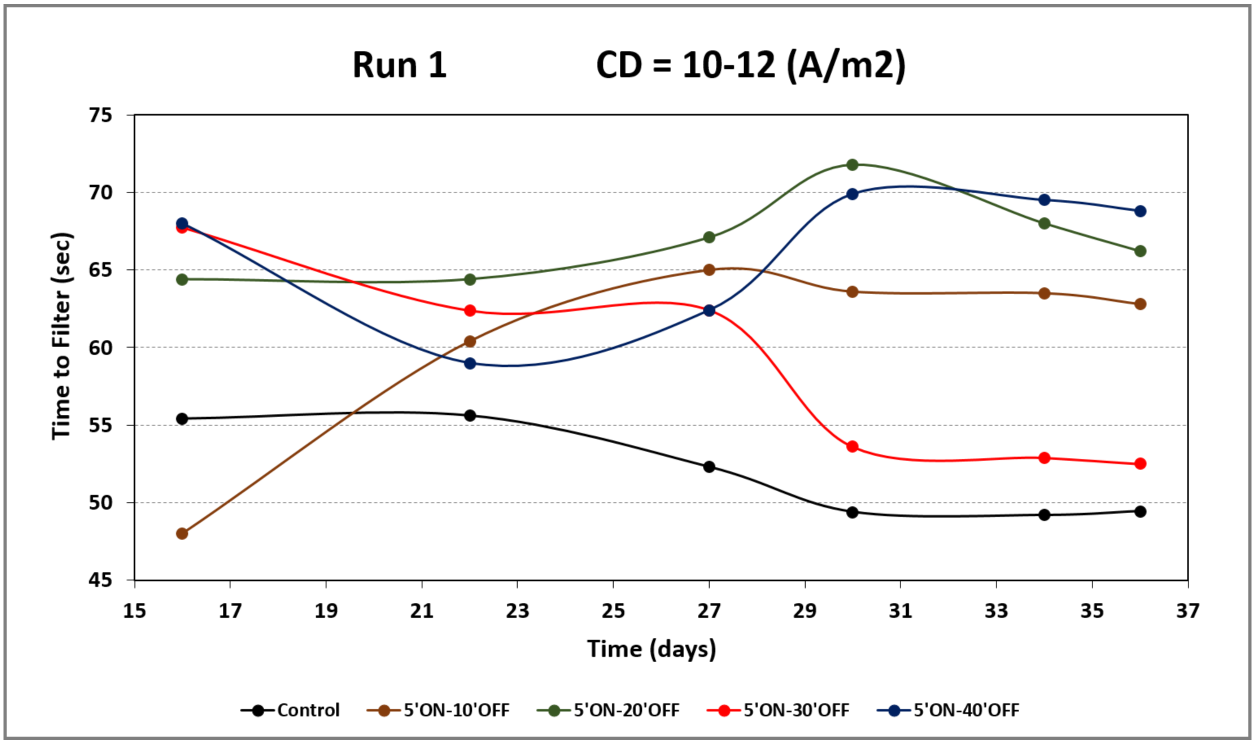

Figure 4.10 Run 1 TTF at different electrical exposure modes

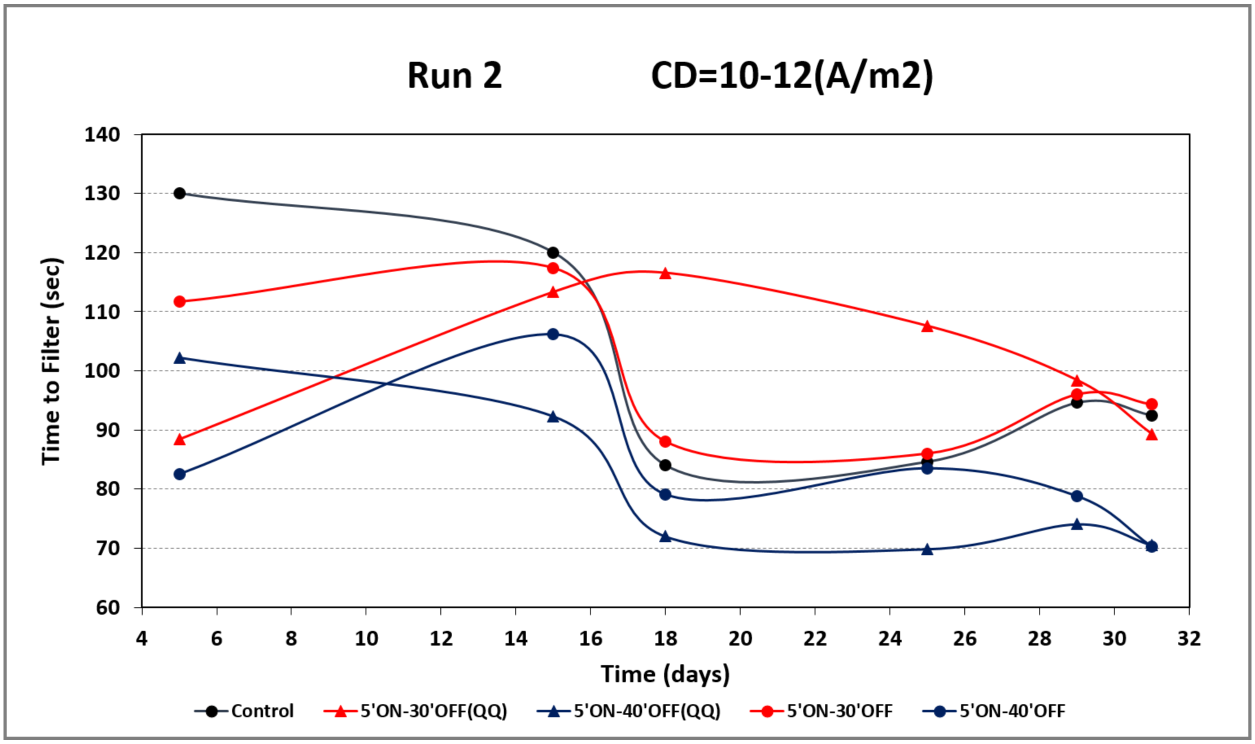

Figure 4.11Run 2 TTF at different electrical exposure modes

4.1.7 Current density vs soluble Extracellular polymeric substances

EPS is considered to be the most important parameter in membrane fouling. It creates a hydrated gel type layer on membrane surface (Reid et al., 2008). Proteins and carbohydrates are known as the major components of EPS that causes membrane fouling. (Xiong & Liu, 2013). There are three types of EPS (i) soluble EPS (ii) loosely bound EPS and (iii) tightly bound EPS. In both stages of Phase 1, only soluble EPS was investigated. At greater time off the EPS production in case of both protein and carbohydrates were less. Figure 1 shows the lesser EPS production in 5’ON-30’OFF and 5’ON-40’OFF at current density 10-12 A/m^2 and MLSS around 7000-8000 mg/L. While higher EPS production was observed at 5’ON-10’OFF. This might be because microorganisms were unable to tolerate the current at lesser time OFF with current density 10-12 A/m^2. So, larger time OFF helped them to regain their activity. Similarly, at stage 2 there was a significant decrease in EPS production of hybrid EBR 5’ON-30’OFF with quorum quenching beads as compared to non-hybrid bioreactor 5’ON-30’OFF.

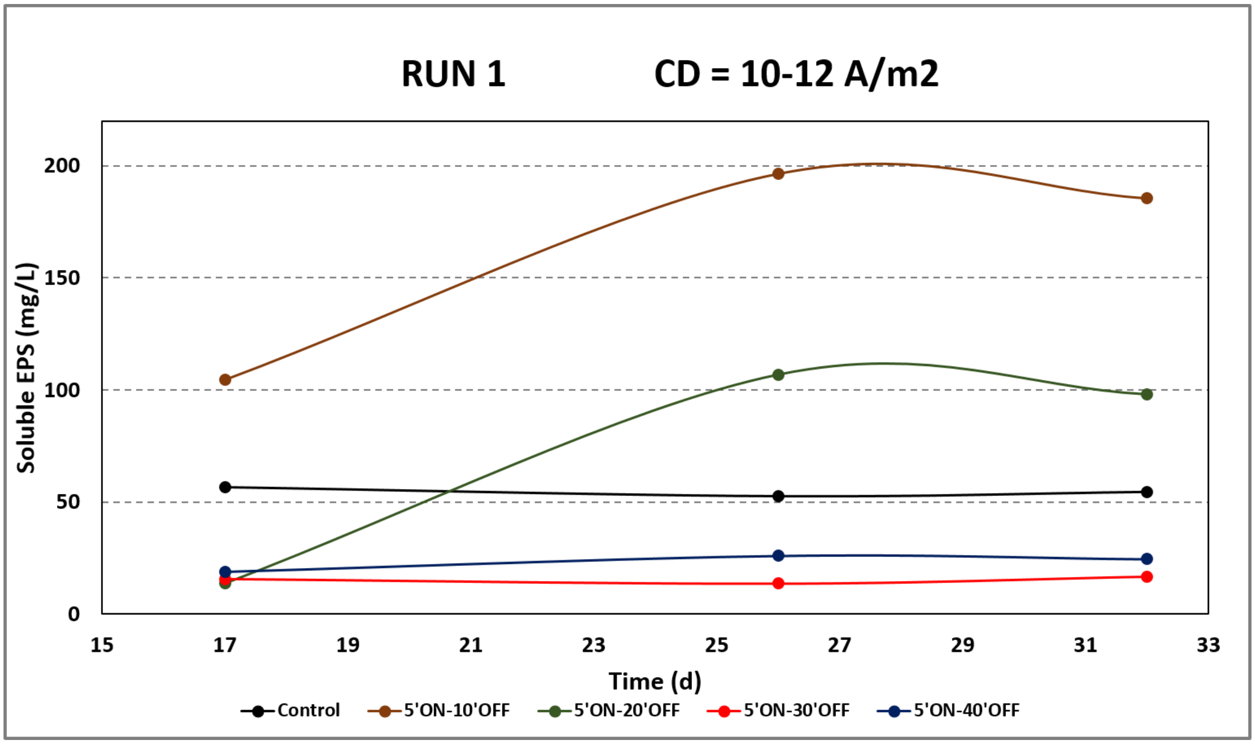

Figure 4.12 Run 1 EPS at different Electrical Exposure Modes

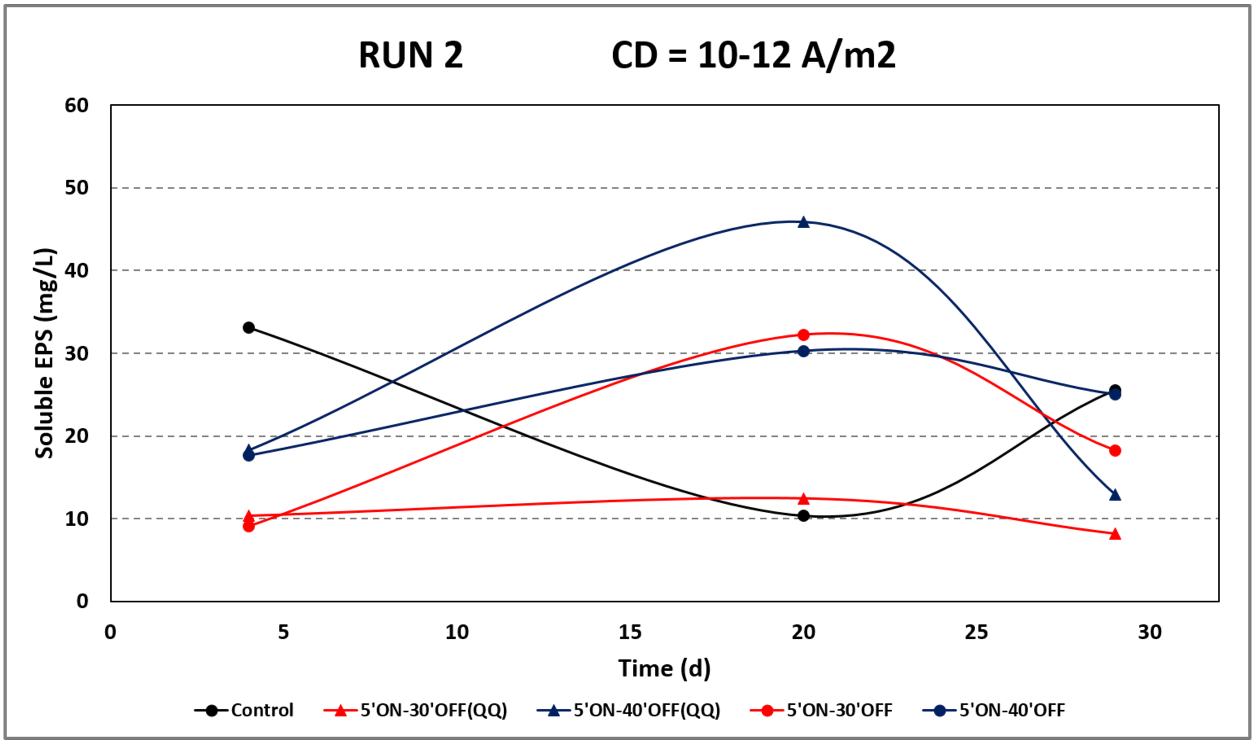

Figure 4.13 Run 2 EPS at different Electrical Exposure Modes

4.2 PHASE 2

4.2.1 Mixed Liquor Suspended Solids

Figure 4.14 shows the dissimilarity in MLSS of both MBRs. Provision of current into solution results into generation of aluminum ions that eventually dissolves into solution, which results into increase in overall suspended solids Hasan, S. (2011). It could be observed that MLSS increased from 3700 to 8000 mg/L in case of hybrid SMEBR at SRT of 20 days This can be explained by the phenomena called electro dissolution in which the dissolving of a substance from electrode into electrolysis occurs. By the 26th day of operation MLSS of SMEBR(QQ) become gradually stable because of the limited substrate for microbial growth due to the consumption by electrocoagulation. Whereas in simple MBR with QQ beads an increase in MLSS was 3200 to 5000 mg/L was observed.

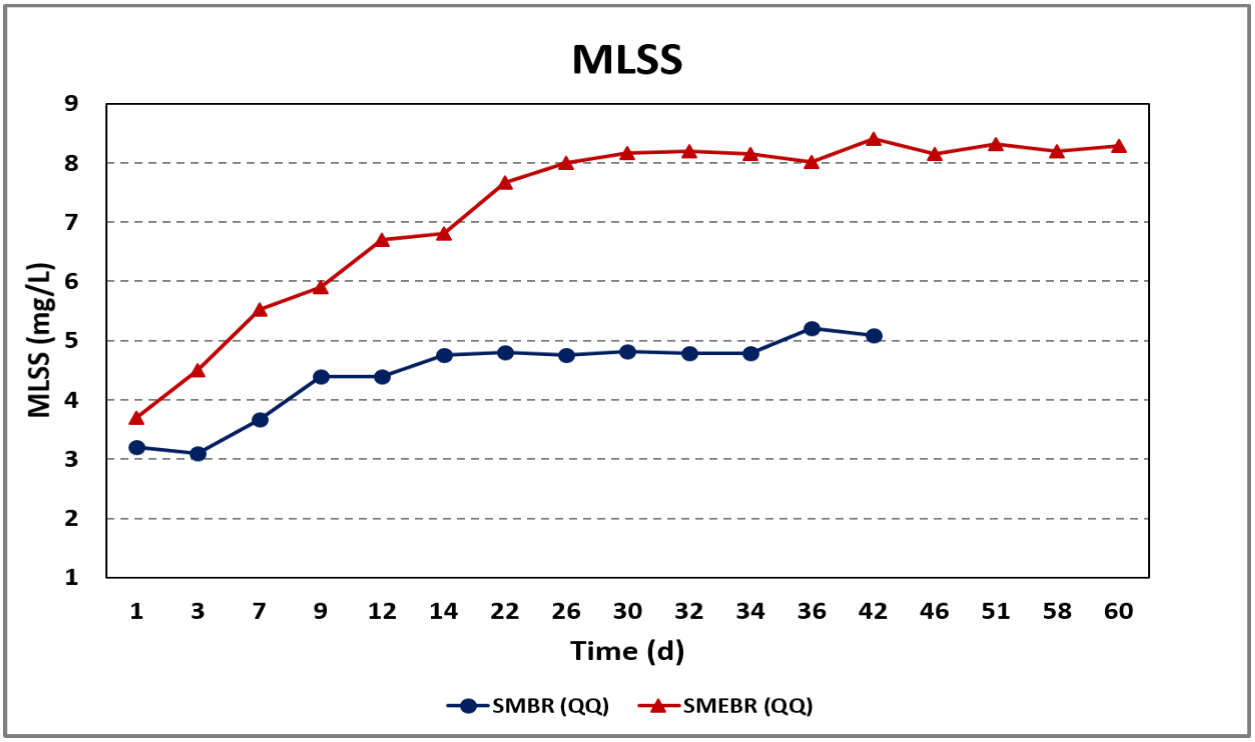

Figure 4.14 MLSS concentration of SMEBR(QQ) & SMBR(QQ)

4.2.2 Chemical Oxygen Demand (COD)

Figure 4.15 shows the COD removal percentage and effluent concentration in both SMEBR(QQ) and MBR(QQ). COD removal % in MBR with QQ beads remained in between 90-94% throughout the operation. It can be clearly observed that initially COD removal was 90 to 93% because of acclimatization period of electric current by microorganisms. As the operation continued high MLSS offers more solid surfaces to intermingle electrically with organic colloids hence COD removal percentage increased to 97% in SMEBR(QQ).

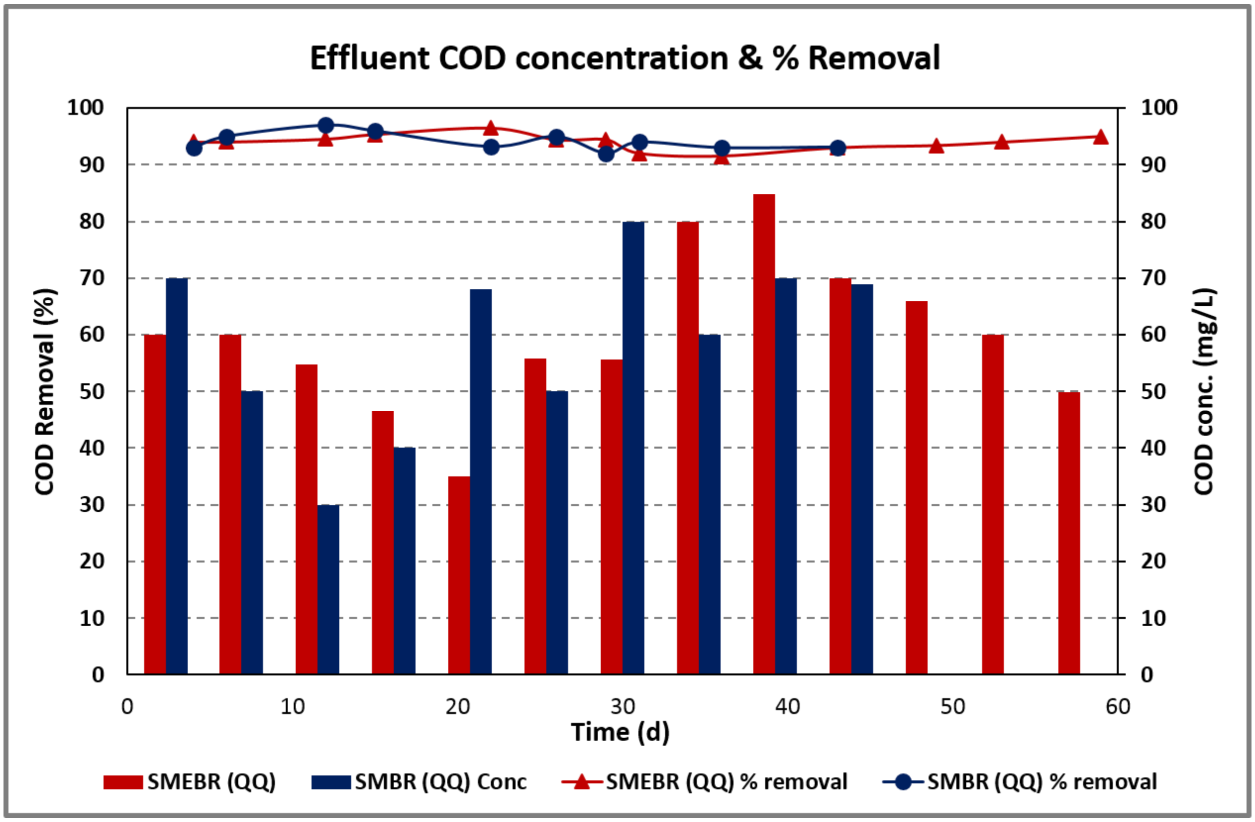

Figure 4.15 COD removal percentage and concentration of SMEBR(QQ) & SMBR(QQ)

4.2.3 Sludge Volume Index

SVI is determined to find out the quality and quantity of sludge that must be wasted. Figure 4.16 shows the SVI over time of both SMEBR with QQ beads and MBR with QQ beads. The continuous decrease in SVI of hybrid SMEBR declared that sludge could be better dewatered and was more settleable. Whereas, the sludge of MBR with QQ beads took slightly greater time for settling as compared to SMEBR with QQ beads. Although the difference in SVI values was not significant because of greater MLSS of hybrid SMEBR. SVI of SMEBR(QQ) reduced from 145 to 53 mL/g at the end of operation while SVI of MBR(QQ) was 145 to 95 mL/g.

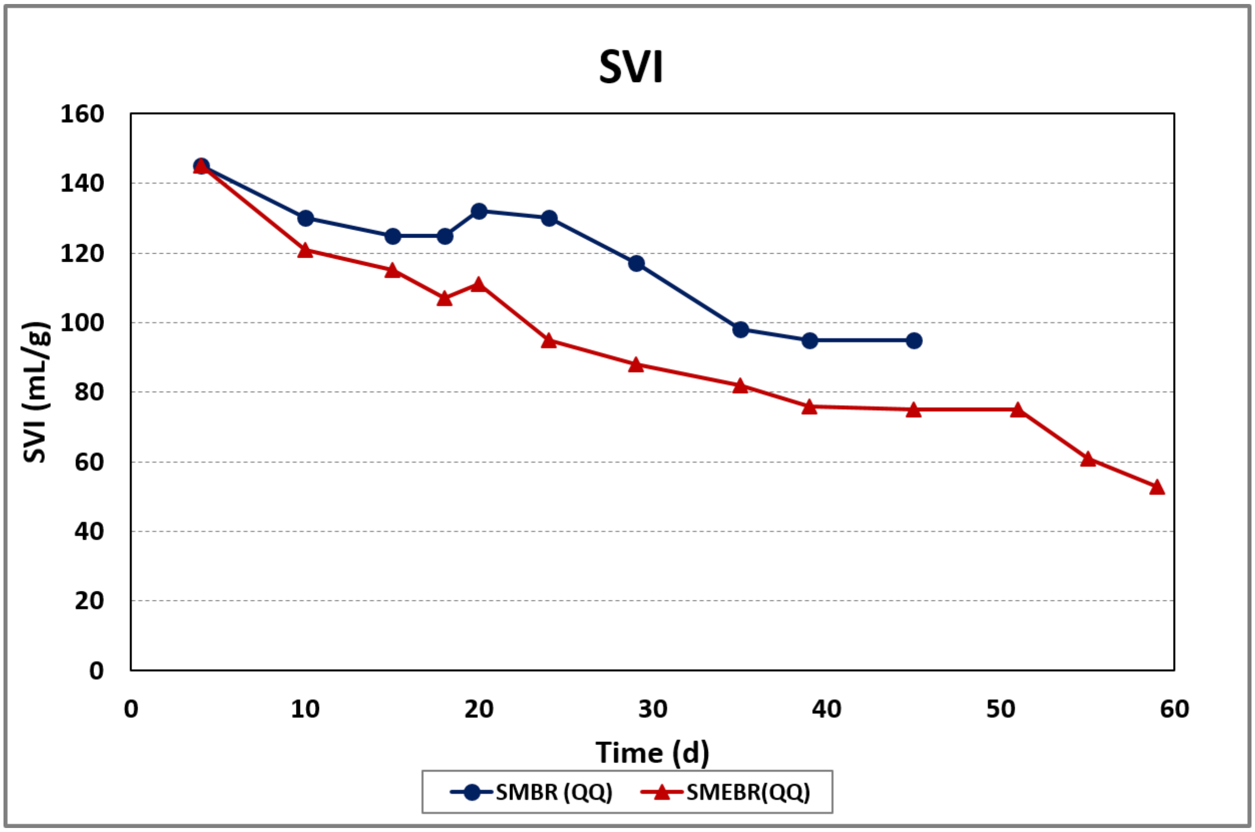

Figure 4.16 SVI concentration of SMEBR(QQ) & SMBR(QQ)

4.2.4 Ammonium removal

The ammonium removal is illustrated in Figure 4.17. Nitrogen was removed by transformation of ammonium into nitrite and nitrate through nitrification processes.

In the conventional activated sludge process, organic matter is converted to carbon dioxide by microorganisms which grow in the flocs. This process needs lots of electrical energy for providing oxygen. In anaerobic systems, most of the organic compounds converts to biogas whereas Anammox process does not need organic compounds for removing ammonia or nitrogen (Kartal et al., 2010). So annamox baxteria are seemly to be present in electro bioreactor system due to possibility of alternative aerobic/ anaerobic conditions in a reactor which converts ammonium and nitrite into nitrogen gas by consuming inorganic carbon source. In Figure 4.17 Ammonium removal in SMEBR(QQ) was above 90% while in SMBR(QQ) it was in between 80-85%. A small difference maybe due to annamox process by floc formation through electrocoagulation.

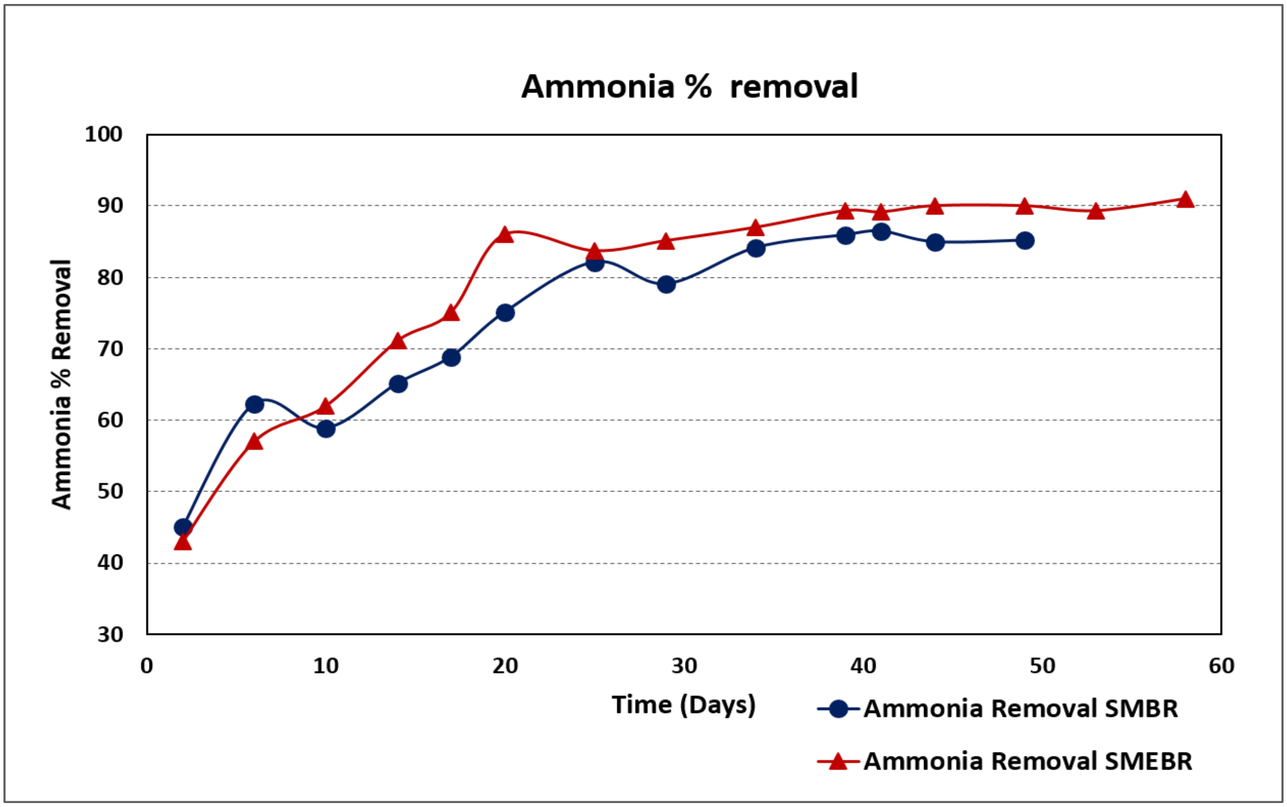

Figure 4.17 Ammonia removal percentage of SMEBR(QQ) & SMBR(QQ)

4.2.5 Phosphorus removal

The electrochemical dosing of aluminium ion at these electrical parameters in the system formed complexes with phosphorus (PO_4^{-3}) and remove it from the liquid part of the activated sludge. The complete elimination of phosphorus (PO_4^{-3}) in SMEBR, in addition to biological degradation by the microorganisms, could be accredited to electro kinetics. Hasan, (2011) found significant amount of phosphorus deposits on the electrodes, majorly on cathode surface. He observed that phosphorus made various different chemical complexes at cathode surface. In short, removal mechanism of phosphorus or other metals could be attributed to the absorption to sludge flocs, precipitation of the metal hydroxides to form complexes with or droped on the electrode surface, mainly on cathode surface (Hasan, 2011). Removal of phosphorus due to electrocoagulation demonstrates the method of the chemical reaction in which the organic pollutants react with the metal ions released through electrooxidation in the presence of DC voltage. Hence maximum removal of phosphate from wastewater by SMEBR(QQ) was observed. Whereas, MBR(QQ) showed very less phosphate removal as compared to SMEBR(QQ).

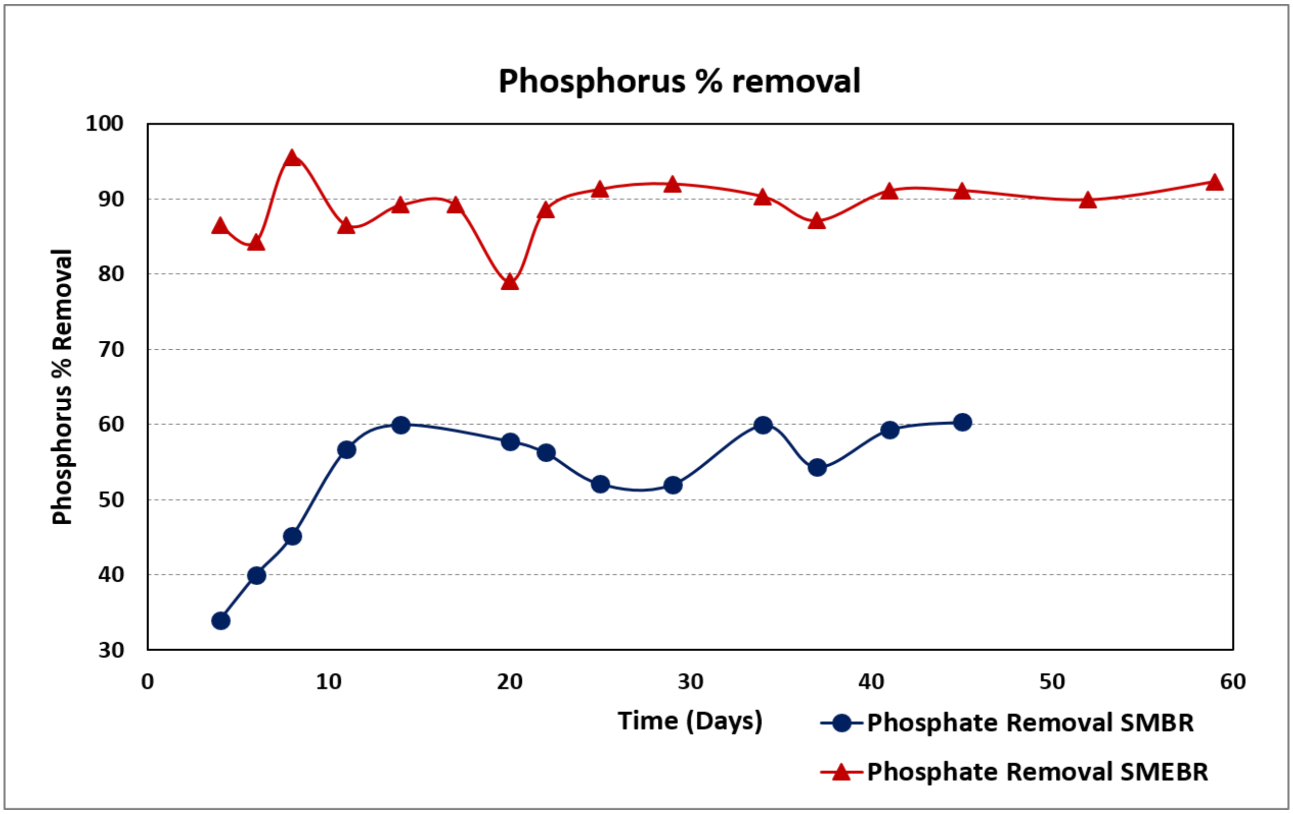

Figure 4.18 Phosphorus removal percentage of SMEBR(QQ) & SMBR(QQ)

4.2.6 Particle size distribution

Figure 4.19 shows the particle sizes of both SMEBR(QQ) and MBR(QQ). It can be clearly observed that at first the Particle size increased in SMEBR(QQ) due to electrocoagulation process but then decreased because positive counterions of sludge were attracted by the cathode thus they repelled water molecules out of the sludge particles. This trend was also observed in Giwa, Ahmed & Hasan (2015), particle size first increased up to 19th day and then decreased when discharge of bound water became predominant over accretion of particles. Particle size of MBR(QQ) showed constant decreasing trend until 35 days of operation and then became constant.

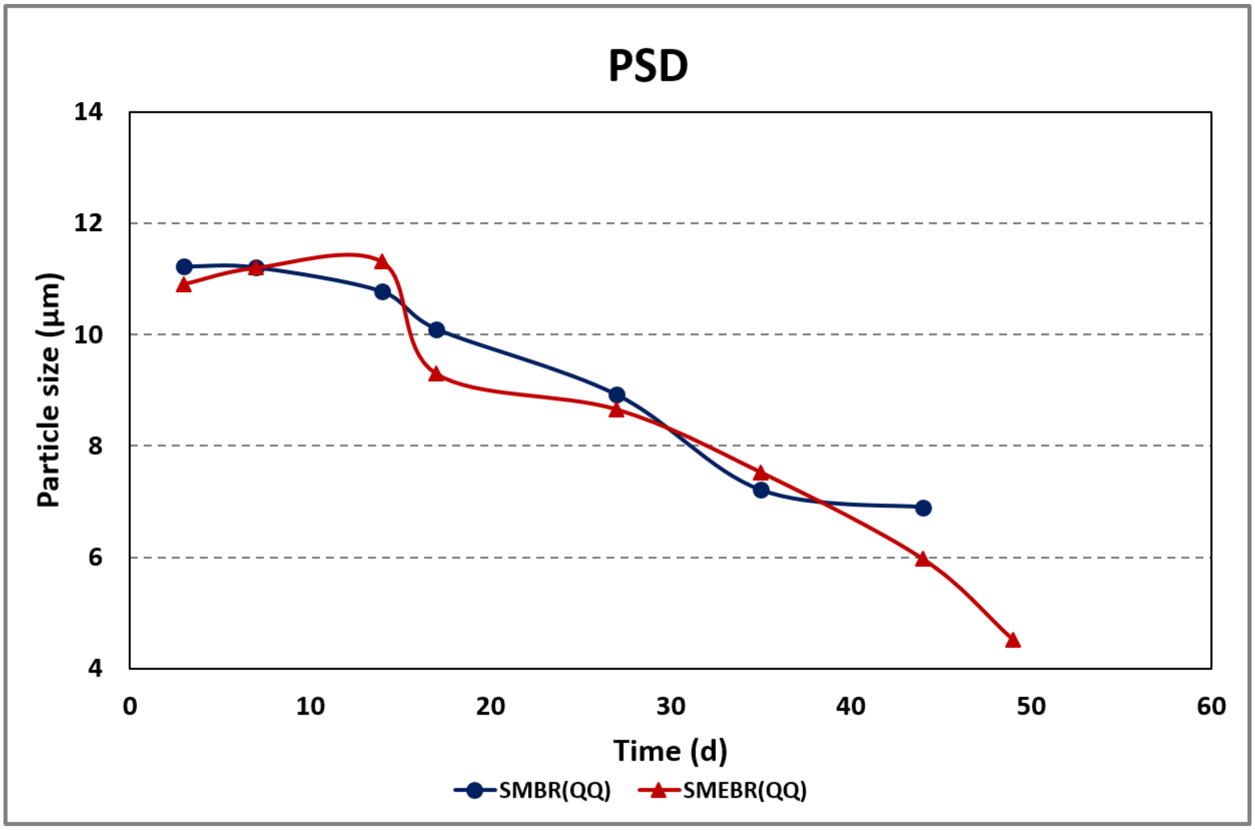

Figure 4.19 Particle Size of SMEBR(QQ) & SMBR(QQ)

4.2.7 Time to Filter

TTF is basically the time required to filter half of 100 mL sludge through vacuum filtration assembly. It is used to monitor the quality of sludge. TTF of SMEBR(QQ) was found to be slightly better than SMBR (QQ). In SMEBR(QQ) by filtration process water moves easily out of the sludge due to electro osmosis phenomena according to which the charged colloidal particles moved towards the opposite charged electrodes in the presence of electric field and thus water molecules migrate out of the mass hence leading to good separation. Dewaterability of sludge in SMEBR(QQ) improved more proficiently than SMBR(QQ) throughout the operation period. TTF has reduced from 108 to 80 secs compared to the initial stage of treatment whereas TTF of SMBR(QQ) decreased from 130 to 87 secs until end of operation. Therefore, sludge handling could be easier in case of SMEBR(QQ).

There was constant decrease in the TTF of SMEBR(QQ) until 60 days of operation whereas, TTF of SMBR(QQ) first observed decrease until 20th day and then increase and then decrease.

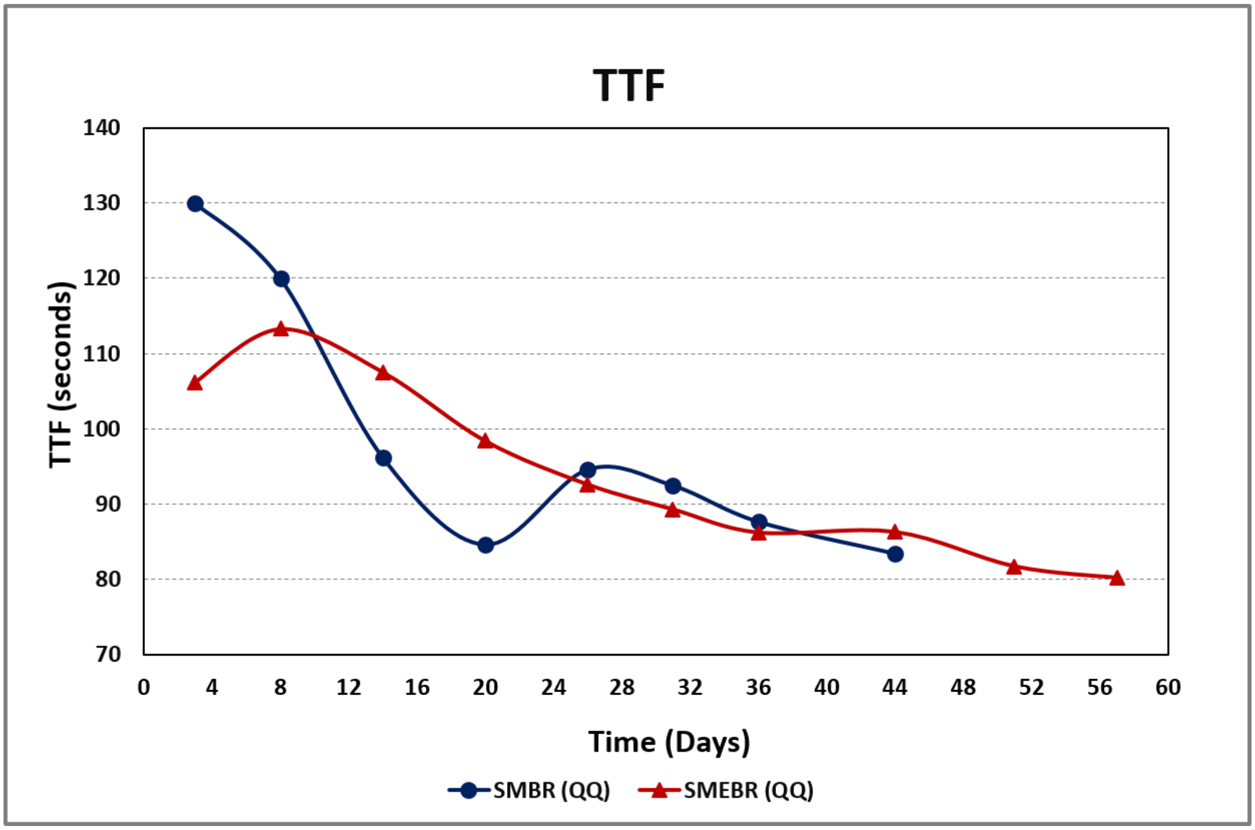

Figure 4.20 TTF of SMEBR(QQ) & SMBR(QQ)

4.2.8 Extra Cellular Polymeric Substances

EPS act as a platform and provide an environment for microorganism to agglomerate by polymer tangle on surface of membrane. According to (Shin et al., 2001) EPS have negative charge on it. Proteins are pondered as major donors to increase in the negative charges in the mixture (Wilén et al., 2003). If Protein Concentration becomes high, then it increases the hydrophobicity of Mixed Liquor causing fouling of membrane abruptly. (Deng, et al., 2014). The positively charged Al^{+3} ions neutralizes these negative charged EPS particles.

(Maqbool, 2014) studied the reduction in EPS production through spherical QQ beads having rhodococcus bacteria. He found very less PN concentration in QQ-MBR than C-MBR which indicated less hydrophobicity of activated sludge flocs and inhibited the biofilm formation on the membrane surface. Initial sludge EPS concentrations were similar as both reactors were fed with same sludge concentration. As shown in Figure. SMEBR(QQ) showed a verified decrease in EPS. Loosely bound (LB) and tightly bound (TB) EPS became stable after 40 days of operation. Electrocoagulation reduced the production of both PN and PS than SMBR(QQ). It may be due to electro osmosis phenomena because of applied current. The working principle of this phenomena is that colloidal particles are charged and when electric field is applied, it will cause the particles to transfer and attracted towards oppositely charged electrodes. Thus, water molecules move out of the mass hence provides good separation of loosely bound and tightly bound water from sludge. So, it could be decided from these results that electric field and QQ bacteria had significant impact on EPSp and EPSc.

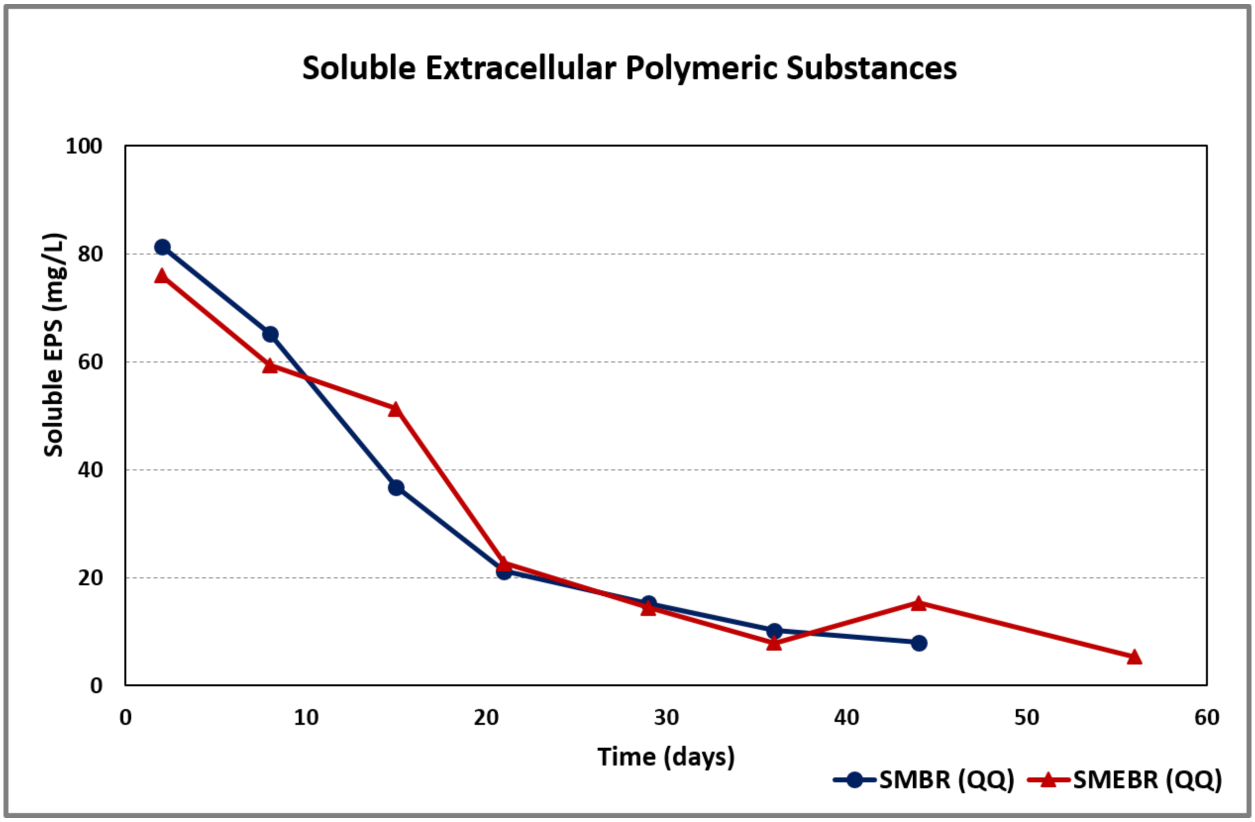

Figure 4.21 Soluble EPS of SMEBR(QQ) & SMBR(QQ)

4.2.9 Loosely & Tightly Bound EPS

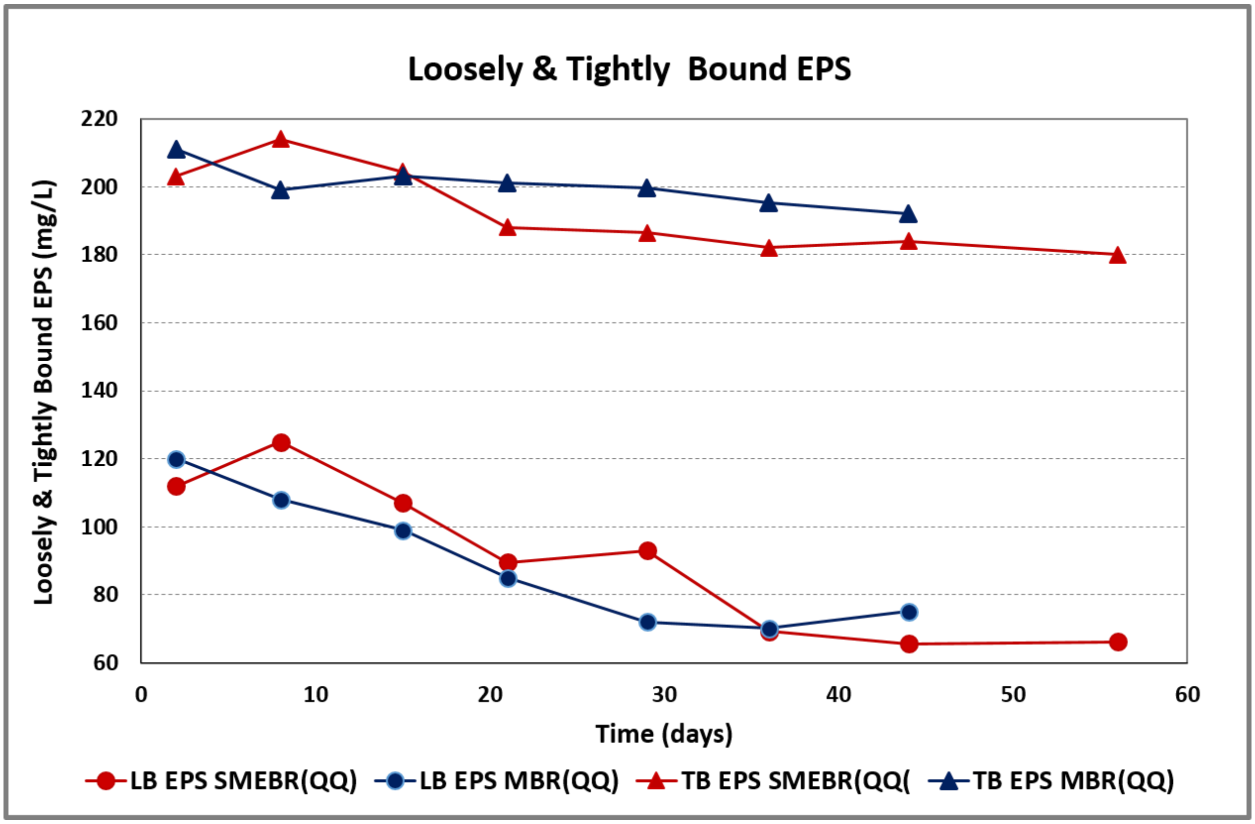

Figure 4.22 Loosely and Tightly Bound EPS of SMEBR(QQ) & SMBR(QQ)

4.2.10 Transmembrane Pressure (TMP)

TMP Profiles are used to determine the fouling behavior of membranes. Hollow cylindrical Quorum Quenching beads having entrapped Rhodococcus bacteria were used about 0.5% of the total effective reactor volume in both reactors. TMP profiles of both SMBR(QQ) and SMEBR(QQ) were compared in figure and considerable difference in fouling behavior of both membranes was observed. (Pervez, 2016) found the rapid TMP rise to 30 KPa within 10-12 days of Conventional MBR. By introducing HC QQ beads TMP rise delayed to 44-46 days and with the addition of electrocoagulation as SMEBR(QQ) TMP rise further delayed to 30KPa within 58-60 days. This clearly shows the effectiveness of SMEBR(QQ) innovation. Average membrane fouling rate ($\Delta P/\Delta t$) in SMEBR(QQ) was found to be 0.5KPa/day and while greater in SMBR(QQ), 0.68 KPa/day. This shows that electrocoagulation further enhanced the reduction in membrane biofouling in SMBR(QQ). it cannot be depicted from these results that whether electric current activate QQ microbes further or electrocoagulation was the only process to reduce membrane biofouling, but the synergic effect of both techniques showed encouraging results and opened the new doors for further researches.

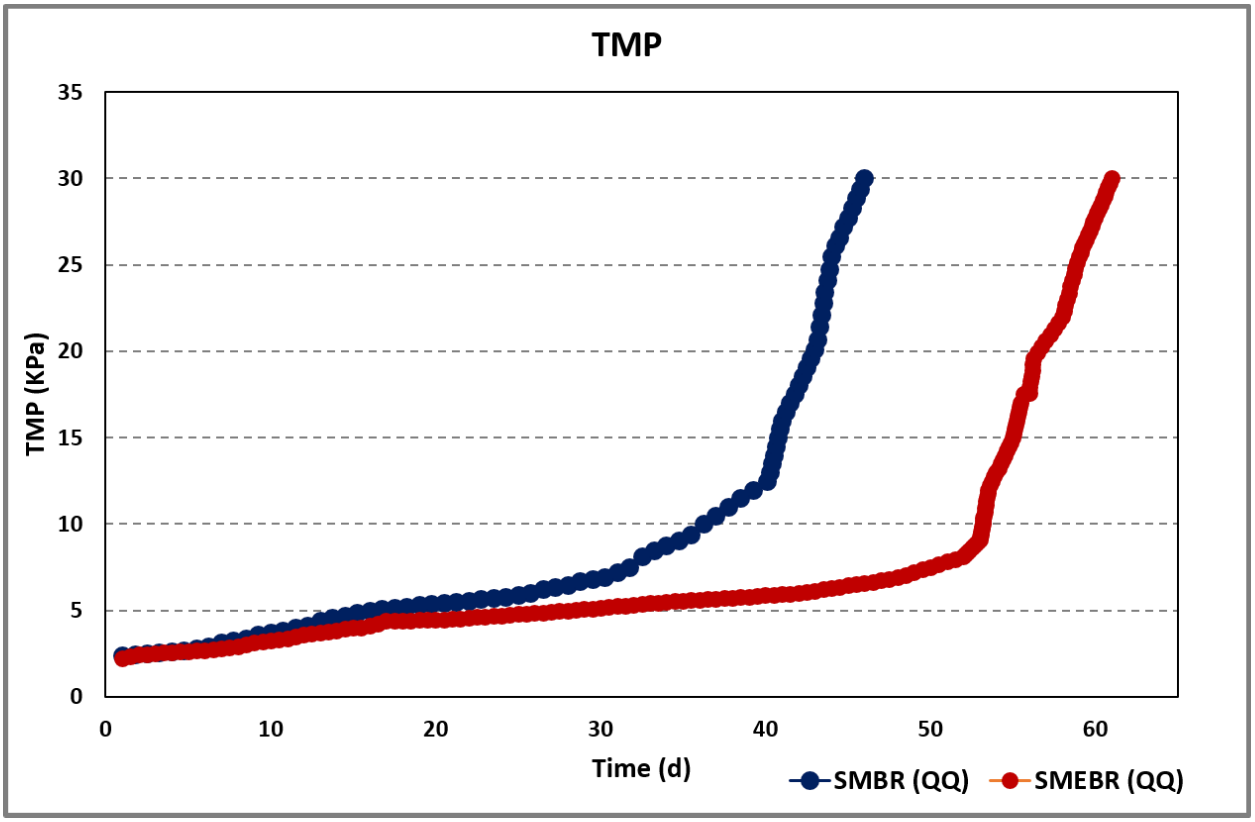

Figure 4.23Transmembrane Pressure (TMP) of SMEBR(QQ) & SMBR(QQ)

5 Conclusions and Recommendations

5.1 Conclusions

Phase 1

This study was done in two phases. Phase 1 consists of batch study with four electro bioreactors and one control reactor. The electrical operation exposure mode in batch study were different in each of the two stages. Stage one was operated to optimize the two affected electrical exposure modes that were then further studied in stage two with and without hollow cylindrical Quorum Quenching beads. 5’ON-30’OFF and 5’ON-40’OFF were selected through stage one results and then operated in stage two with and without QQ HC beads. Through phase 1 it was concluded that Current density with 10-12 A/m^2 with exposure mode of 5’ON-30’OFF(QQ) enhanced microbial activity and improved sludge characteristics

Phase 2

In this phase two parallel MBRs were operated, Submerged membrane electro bioreactor (SMEBR) with Hollow Cylindrical Quorum beads (HC QQ) and MBR with HC QQ beads.

SMEBR with QQ beads delayed TMP rise, reduced soluble, loosely bound and tightly bound EPS, improved sludge dewaterability in terms of TTF and reduced particle size for effective sludge activity. No adverse effect of electric current on HC QQ beads was observed as the synergic effect of both control strategies on membrane fouling was more than a single technique. The fouling rate was more in MBR(QQ) than SMEBR(QQ) which results in rapid membrane biofouling whereas addition of electric current prolonged the filtration duration in SMEBR(QQ). No adverse effect of electric current on HC QQ beads was observed in term of treatment performance. Introduction of electric current for electrocoagulation in MBR(QQ) led to further inhibition of biofilm formation which improved the filtration and permeability of membrane in SMEBR(QQ)

The major conclusions from this research were;

- Significant delay in TMP profile was observed as addition of DC voltage to SMEBR(QQ) reduced membrane fouling rate 1.36 times lesser than SMBR(QQ).
- COD removal in SMEBR(QQ) was 95-97% whereas in SMBR(QQ) COD removal was 93-94%. A relatively higher difference of COD removal in SMEBR(QQ) was mainly due to the exclusion of colloidal organics along with biological oxidation.
- Phosphorus removal was significantly affected by the current density. Average phosphorus removal in MBR(QQ) was 62-64% respectively. In contrast, average phosphorus removal in SMEBR(QQ) was above 90%.
- Average Ammonium removal in SMEBR(QQ) was above 90% which was greater than SMBR(QQ) (80-85%).
- Time to filter (TTF) and Particle size (PSD) was less in SMEBR(QQ) than SMBR(QQ) as compared to initial values. Particle size in SMEBR(QQ) was first increased than decreased when bound water released form sludge particles.
- EPS in both SMEBR(QQ) and SMBR(QQ) became stable after 40 days of operation and net decrease in SMEBR(QQ) EPS was found to be greater than SMBR(QQ).

5.2 Recommendations

- To study the effect of different current densities on Hollow cylindrical quorum quenching beads and membrane fouling
- Real wastewater replacing synthetic one and then compare TMP rise-up between conventional and SMEBR(QQ).
- Investigate the fate of different metals in SMEBR(QQ).
- Cost and power requirement analysis of SMEBR(QQ).
- Convert SMEBR(QQ) to pilot scale and full scale applications

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