CONFIGURATION OF MOLECULAR IMPRINTED POLYMERS FOR SPECIFIC UPTAKE OF PHARMACEUTICAL IN AQUEOUS MEDIA THROUGH RADICAL POLYMERIZATION METHOD

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by

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LIST OF SYMBOLS

ΔE	Interaction energy	kJ/mol
Δm	Change in mass of permeate	kg
A_m	Effective filtration area	
C_f	Feed concentration of acetaminophen solution	mg/L
C_o	Initial concentration of acetaminophen	mg/L
C_p	Concentration of acetaminophen solution	mg/L
C_t	Residual concentration of acetaminophen	mg/L
J	Permeation flux	kg m^{-2} h^{-1}
J_1	First recorded permeate flux	kg m^{-2} h^{-1}
J_n	Subsequent permeate flux	kg m^{-2} h^{-1}
J_p	Permeate flux of tested solution	kg m^{-2} h^{-1}
J_{p2}	Second permeate flux of tested solution	kg m^{-2} h^{-1}
J_{w1}	Initial water flux	kg m^{-2} h^{-1}
M_w	Molecular weight	g/mol
Q	Adsorption or binding capacity	mg/g of polymer
Q_{MIP}	Adsorption capacity of MIP	mg g^{-1}
Q_{NIP}	Adsorption capacity of NIP	mg g^{-1}
R	Acetaminophen rejection	%
R_f	Resistance due to fouling	cm^{-1}
R_{ir}	Resistance due to irreversible fouling	cm^{-1}
R_m	Membrane intrinsic resistance	cm^{-1}
R_r	Resistance due to reversible fouling	cm^{-1}
R_t	Total resistance of membrane	cm^{-1}
t	Time	h

TMP	Transmembrane pressure	Pa
V	Volume of acetaminophen solution	L
vol%	Percentage by volume	
W	Mass of MIP and NIP	g
w/w%	Weight concentration of solution	
wt%	Percentage by weight	
μ	Viscosity of permeate	Pa.s

LIST OF ABBREVIATIONS

2-VP	2-vinylpyridine
4-VP	4-vinnylpyridine
AA	Acrylamide
ABCN	1,1'-Azobis(cyclohexanecarbonitrile)
AC	Acetaminophen
ACN	acetonitrile
Act-MIP	Acetaminophen imprinted polymers
AIBN	Azobis-(isobutyronitrile)
APS	Ammonium persulfate
AuNPs	Gold nanoparticles
AVIM-Cl	1-allyl-3-vinyllimidazole chloride
BAW	Bulk acoustic wave
BC	Binding capacity
BET	Brunauer-Emmett-Teyler
Bio-DLLME	Bio-dispersive liquid-liquid microextraction
BJH	Barrett-Joyner-Halenda
CAF	caffeiene
CO_2	Carbon dioxide
DCF	Diclofenac
DFZ	2,5-deoxyfructosazine
DMF	N,N-dimethylformamide
DVB	Divinylbenzene
EDGMA	ethylene glycol dimethacrylate
FLUMIP	Fluconazole molecularly imprinted polymers

fM:X	Ratio of functional monomer to cross-linker
FRR	Flux recovery ratio
FZ	Fructosazine
GE	Gold electrode
HEMA	2-hydroxyethyl methacrylate
HEMA	2-hydroxyethylmethacrylate
HMIP	Hybrid molecularly imprinted polymers
HPLC	High performance liquid chromatography
I:tM	Ratio of initiatir to total monomer
IA	Itaconic acid
IF	Imprinting factor
IPB	Induced promotion of binding
Itoh	Itopride hydrochloride
KPS	Potassium persulfate
MAA	Methacrylic acid
MBAA	N,N'-methylenediacrylamide
MBR	Membrane bioreactor
MIM	Molecularly imprinted membrane
MIPs	Molecularly imprinted polymers
MI-SPE	Molecular imprinting solid phase extraction
NIP	Non-imprinted polymer
NMP	1-methyl-2-pyrrolidone
NP	Nanoparticles
PANI	Polyaniline
PEDGA	Polyethylene glycol diacrylate
PES	Polyethersulfone
PHDFDMA	Poly(heptadecafluorodecyl methacrylate)

PMIP	Porous molecularly imprinted polymers
PP	Polypropylene
PPCP	Pharmaceutical and personal care products
PR	Paracetamol
PS	Polysulfone
PVDF	Polyvinylidene fluoride
RFR	Relative flux reduction
RT	Response time
SDG	Sustainable Development Goals
SDS	Sodium dodecylsulfate
SEM	Scanning electron microscopy
T:fM	Ratio of template to functional monomer
TFMAA	Trifluoromethyl acrylic acid
TGA	Thermogravimetric analysis
THF	Tetrahydrofuran
THP	Theophylline
TRIM	Trimethylolpropane trimethacrylate
UV-Vis	Ultraviolet-visible

KONFIGURASI POLIMER CETAK MOLEKUL UNTUK PENGAMBILAN ASETAMINOFEN SECARA SPESIFIK DALAM MEDIA BERAIR MELALUI KAEDAH PEMPOLIMERAN RADIKAL

ABSTRAK

Kaedah pempolimeran pemendakan digunakan untuk menyediakan polimer cetak molekul (MIP) untuk pengambilan asetaminofen dalam media berair. Asetaminofen, asid metakrilik (MAA), ethylene glycol dimethacrylate (EDGMA), 1,1'-Azobis(cyclohexanecarbonitrile) (ABCN) masing-masing digunakan sebagai templat, monomer berfungsi, cross-linker dan intiator. Molariti cross-linker dan monomer berfungsi divariasikan untuk kajian kesan mencetak oleh MIP. Kepekatan tinggi cross-linker menunjukkan keupayaan mengikat MIP yang lemah sementara molariti monomer yang tinggi menunjukkan prestasi yang lebih baik dalam kapasiti pengikatan. MIP optimum diperolehi dari nisbah molar templat: monomer: cross-linker pada 1:58:15 dengan kapasiti pengikatan 3.68 mg / g polimer. Seterusnya, PES kosong dan membran cetak molekul (MIM) dibuat menggunakan kaedah penyongsangan fasa. MIM yang ditambahkan dengan MIP optimum dalam larutan pemutus untuk mengkaji sifat antikotoran berbanding dengan membran tulen. Fluks relatif MIM telah menunjukkan tingkah laku antikotoran yang lemah pada sampel air sisa sebenar dan prestasi yang baik dalam larutan sintetik. Walau bagaimanapun, MIM dan membran kosong telah menunjukkan penolakan asetaminofen yang lebih baik dalam air sisa masing-masing pada tahap 71.31 dan 73.06%

CONFIGURATION OF MOLECULAR IMPRINTED POLYMERS FOR SPECIFIC UPTAKE OF PHARMACEUTICAL IN AQUEOUS MEDIA THROUGH RADICAL POLYMERIZATION METHOD

ABSTRACT

Precipitation polymerization method was used to prepare molecularly imprinted polymers (MIP) for the uptake of acetaminophen in aqueous media. Acetaminophen, methacrylic acid (MAA), ethylene glycol dimethacrylate (EDGMA), 1,1'-Azobis(cyclohexanecarbonitrile) (ABCN) were used as template, functional monomer, cross-linker and intiator respectively. The molarity of cross-linker and functional monomer were varied for the study of imprinting effect of MIPs. High concentration of cross-linker exhibit poor binding ability of MIPs while high molarity of monomer demonstrate better performance in binding capacity. The optimum MIP was observed from template:monomer:cross-linker molar ratio at 1:58:15 with binding capacity of 3.68 mg/g polymer. Next, pristine PES and molecularly imprinted membrane (MIM) were fabricated using phase inversion method. MIM was prepared by adding optimum MIPs in casting solution for the study of antifouling properties as compared to pure membrane. The relative flux of MIM has showed a poor antifouling behaviour in real wastewater sample while a good performance in synthetic solution. However, MIM and pristine membrane have revealed better rejection of acetaminophen in wastewater at 71.31 and 73.06% respectively.

CHAPTER 1

INTRODUCTION

1.1 Background

Pharmaceuticals are best to defined as substance that are mainly used in the purpose of therapeutic, preventive and diagnostic. In last few decades, the consumption of pharmaceuticals has been increase significantly due to world population growth, more investment in the health-care sector, advanced in research and development and persistent global market availability (aus der Beek *et al.*, 2016). Thus, there is a concern regarding to the presence of some environmental pollutants in drinking water including pharmaceuticals and personal care products (PPCP) that may have risk on the environment and human health due to long term exposure (de Jesus Gaffney *et al.*, 2015). Traditionally, biological receptors are used in recognition of compounds including pharmaceuticals due to their affinity and selectivity. However, high cost and low stability of biological receptors have limit their application. Recently, molecular imprinted polymers (MIPs) have gained attention to replace biological receptors in various applications including solid-phase separation, antibody mimics, enzyme mimics, drug delivery and development, capillary electrochromatography, organic synthesis and biomedicine (Zhang, 2014).

MIPs are defined as natural synthetic antibody-antigen systems. These polymers are selectively binding the molecule in which they were templated via a “lock and key” mechanism during the production process. MIPs competent to offer selectivity and specificity of the natural receptors with numerous advantages such as low cost, robustness in environmental conditions, universal applicability, and can be stored without special environmental storage conditions. The application of MIPs in sensors

required important parameters such as binding capacity, imprinting factor, and response time. Binding capacity (BC) is formulated as the ratio of the target molecule concentration adsorbed from test solution divided by early concentration of previous solution which need to multiply by 100%. The imprinting factor (IF) is described as the ratio of the template binds in the imprinted polymer to the template binds in the nonimprinted polymer. The standard definition of response time (RT) is the time taken to reach 63.2% of the last signal from the starting time of the stimulus is applied (Belbruno, 2019).

Generally, MIPs are produced by a synthesis method with the assist of template which creates selective binding cavities within a complex polymeric structure and accomplishes the properties of receptor. As shown in Figure 1.1, the molecular imprinting process involves the polymerization of functional monomers and cross-linkers with the addition of a target molecule which function as template. The removal of the template from the complex polymer network produces the specific imprinting sites that are corresponding to the size, shape and functionality of the target molecule (Dar *et al.*, 2020).

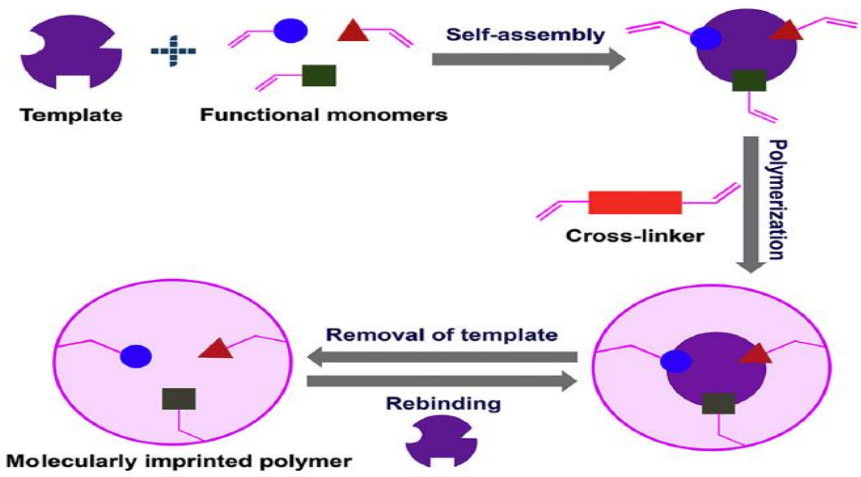


Figure 1.1: General process for the preparation of molecularly imprinted polymers (Dar et al., 2020)

Free radical polymerization of MIPs is the most demanding topic in the research history and it has unbreakable position in the imprinting science field. Preparation of MIPs via free radical polymerization often been conducted by using alkenyl-based raw materials through different type of polymerization method. The general methods used for free radical polymerization are bulk polymerization, precipitation polymerization, in-situ polymerization, emulsion polymerization and suspension polymerization (Zhou *et al.*, 2019). Precipitation polymerization method is a fast and straightforward method for synthesizing high purity of spherical imprinted polymers. The precipitation method was initially recognized in 1999 for obtain molecularly imprinted polymers (MIP) nanoparticles (NP) with an excellent characteristics in term of selectivity and load capacity (Wackerlig and Lieberzeit, 2015). This is method is carried out by homogenous nucleation of all component including monomer and cross-linkers where they were miscible at first. Then, the critical chain length of polymer chains was obtained and it is cannot dissolve any more in the continuous phase. Hence, spherical polymer particles was formed and precipitate was removed (Karnka, Chaiyasat and Chaiyasat, 2017).

Membrane is a thin soft layer material which function as selective barrier between the two adjacent phases and allows the transportation of particles, atoms or molecules to pass through it. The selective transport of membrane is the major advantages of membrane technology compared to other separation technologies. Besides, separation via membrane technology can be conducted isothermally at low temperatures and does not required additives compared to other thermal separation methods. The membrane technology also can be up-scale and downscale easily as well as to integrate the membrane into other separation or reaction processes. The passive transport of substances through membranes are governed by a driving force via pressure gradient, concentration gradient or electrical field gradient (Ulbricht, 2006).

1.2 Problem Statement

There are several methods studied by researchers for the removal of pharmaceutical and personal care products (PPCPs) from water including advanced oxidation and photocatalytic degradation. However, these methods have few drawbacks including required high energy, low efficiency and formation of by-products. Pharmaceutical compounds also can be treated using membrane filtration but only via nanofiltration or reverse osmosis because most PPCPs can pass through membranes with large size of pore (Ravi, Choi and Choe, 2020). Therefore, molecular imprinted polymers (MIPs) are currently used to remove pharmaceuticals such as acetaminophen from water because of its attractive properties. Nevertheless, most of the synthesized MIPs for targeting small organic compounds are commonly formed in organic solvent and they are mostly unable to demonstrate template bindings in aqueous media (Zhang, 2014). The main reasons of having difficulty in producing MIPs with acceptable recognition in aqueous media include the destruction of hydrogen bonding between the template and monomer which lead to the decrease in recognition ability and imprinting efficiency. Next, the imprinted sites also normally destroyed when MIPs are synthesized in a non-polar or low-polar solvent. Besides, MIPs with hydrophobicity properties and poor dispersibility in water will be produced when using alkenyl monomer and crosslinker as raw materials during preparation process. The main factor which determine the binding capacity of MIP is the ratio of templates molecules to functional monomer because it will affects formation of the imprinting sites. Adequate amount of functional monomer will give sufficient binding sites for substances which been discussed in previous study (Li *et al.*, 2019). Therefore, the imprinting factor of MIP can been improved by varying the ratio of template:monomer:cross-linker during the preparation. Furthermore, a hybrid molecularly imprinted polymers (HMIP) membrane

can present as an alternative to conventional membranes. This new type of membrane materials has gained much attention in selective uptake of drug and pharmaceutical compound (Son, Katagawa and Kobayashi, 2011). The fouling of membrane also need to be considered due to the present of interaction between substance and membranes. This interaction can lead to the decrease of flux through the membrane with time and therefore reduce the performance of membrane (Ahmad *et al.*, 2019).

Therefore, this study will focused on the configuration of MIPs for the selective uptake the acetaminophen in aqueous media via precipitation polymerization method. The effect of ratio between monomer, cross-linker, and template on the binding property were studied. Besides, the preparation of hybrid molecularly imprinted polymers membrane for the specific targeted binding of acetaminophen and the fouling the membranes were studied.

1.3 Objective

1. To synthesize and configure molecularly imprinted polymers for specific uptake of acetaminophen in aqueous media through precipitation polymerization method.
2. To study the effect of the ratio between monomer, cross-linkers and template on the binding property
3. To prepare a hybrid molecularly imprinted polymers polyethersulfone (PES) membrane by embedding molecular imprinted polymer (MIP) for selective adsorption of acetaminophen.
4. To investigate the antifouling properties of pristine PES membrane and hybrid molecularly imprinted polymers PES membranes.

CHAPTER 2

LITERATURE REVIEW

2.1 Pharmaceuticals

Acetaminophen (AC) also known as paracetamol is an acylated aromatic amide which is widely used drugs for the treatment of pain and mild fever. It is one of the most popular drugs and considered as first-line therapy due to lack of side effects and low cost. This drugs also categorized as organic contaminants that are continuously released from hospital waste, domestic waste and manufacturing industries into the aquatic environment. Although only low concentration of acetaminophen present in the aquatic environment, there is still potential of harmful effect to the aquatic and human (Montaseri and Forbes, 2018). Acetaminophen or paracetamol with the correct amount of dosage is considered as safe drug. The lowest dose of acetaminophen that possible to cause toxicity is at 7.5 g for an adult or 150 mg/kg of body weight for a child. The toxicity of acetaminophen is mainly involving human organ such as liver and also can cause hepatic failure in kidney (Bertolini *et al.*, 2006). The chemical structure of acetaminophen was given in Figure 2.1.

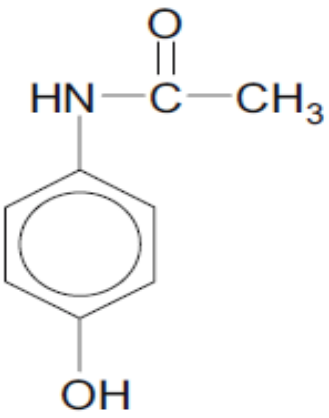


Figure 2.1: Chemical structure of acetaminophen (Bertolini et al., 2006)

2.2 Water-compatible MIP

In the previous decades, many studies have been conducted by researchers to develop MIPs that is compatible in aqueous media because of their potential in different applications including food safety, clinical diagnostics and environmental protection (Zhang, 2014). Addition of hydrophobic, ionic or metal coordination interactions are demonstrating to be able to improve the association of template and functional monomer in water (Vasapollo *et al.*, 2011). Recently, Mohiuddin *et al.* (2020) synthesized porous molecularly imprinted polymers (PMIP) for the extraction of diclofenac (DCF) from the environmental samples. PMIP is used as adsorbent for solid-phase extraction of DCF and it is been analysed by using high performance liquid chromatography. The 4-vinnylpyridine (4-VP), ethylene glycol dimethacrylate (EGDMA), acetonitrile (ACN) were used as functional monomer, cross-linker and porogen respectively. It showed high recovery of DCF (range: 97.02-98.69%) in sewage sample. The water-compatible MIPs by *in-situ* approach of fructosazine (FZ) and 2,5-deoxyfructosazine (DFZ) as templates was conducted for the detection in the food sample (Henry *et al.*, 2012). Acrylamide (AA), polyethylene glycol diacrylate (PEDGA) and water were use as functional monomer, cross-linker and porogen respectively. The extraction of FZ and DFZ from soy sauce sample was performed via molecularly imprinted solid-phase extraction. A green synthesis of novel water-compatible molecular imprinted conductive polyaniline (PANI) nanoparticles (NPs) for the paracetamol detection has been developed in the study of Luo *et al* (2016). The preparation of MIP was conducted in aqueous media has improve the ability of PANI MIP nanoparticles in water and also this process considered as environmental-friendly. Besides, another green and non-toxic molecular imprinted polymer was synthesized in water for the adsorption of ciprofloxacin (Zhu *et al.*, 2019). The preparation of MIP was

developed using 1-allyl-3-vinyllimidazole chloride (AVIM-Cl) and 2-hydroxyethyl methacrylate (HEMA) as bifunctional monomer with maximum rebinding capacities of 19.96 mg g^{-1} at 25 °C. Table 2.1 describes the water compatible MIPs which conducted by different research studies.

Table 2.1: Water compatible MIPs from different studies

MIP	Template	Functional monomer	Cross-linker	Porogen	Reference
Porous molecularly imprinted polymers (PMIP)	Diclofenac	4-vinnylpyridine (4-VP)	Ethylene glycol dimethacrylate (EDGMA)	Acetonitrile (ACN)	(Mohiuddin *et al.*, 2020)
Water-compatible molecularly imprinted polymers (MIP) by in-situ approach	Fructosazine (FZ) and 2,5-deoxyfructosazine (DFZ)	Acrylamide (AA)	Polyethylene glycol diacrylate (PEDGA)	Water	(Henry *et al.*, 2012)
Water-dispersible molecular imprinted conductive polyaniline (PANI) nanoparticles (NPs)	Paracetamol	Aniline	Ammonium persulfate (APS)	N,N-dimethylformamide (DMF)	(Luo *et al.*, 2016)
Green and hydrophilic molecularly imprinted polymer	Ciproflaxin	1-allyl-3-vinylimidazole chloride (AVIM-Cl) and 2-hydroxethyl methacrylate (HEMA)	N,N'-methylenediacrylamide (MBAA)	Pure water	(Zhu *et al.*, 2019)

2.3 Type of polymerization method

Molecular imprinted polymers can be prepared through a different type of polymerization methods such as bulk polymerization, precipitation polymerization, dispersion polymerization, emulsion polymerization and electropolymerization. MIPs are commonly been synthesized via bulk polymerization in which polymers are

grounded into a desired particle size (Yoon and Byun, 2013a). A non-covalent molecular imprinted polymers for the bulk acoustic wave (BAW) sensor was synthesized for the determination of paracetamol (Tan *et al.*, 2001). Paracetamol, methacryclic acid (MAA) or 4-vinylpyridine (4-VP), ethylene glycol dimethacrylate (EDGMA), and azobis-(isobutyronitrile) (AIBN) were used as template, functional monomers, cross-linker and initiator respectively. MIPs and NIP were synthesized via bulk polymerization method to obtain particle size less than 25 μm. The recovery of paracetamol from human serum and urine by MIPs was obtained at 93.3 to 106.7%. Dispersion polymerization method assisted with supercritical carbon dioxide (CO_2) for the preparation of MIP in the adsorption of acetaminophen and aspirin was observed to be more efficient than bulk and emulsion polymerization methods (Yoon and Byun, 2013a). Acetaminophen and aspirin, methacryclic acid (MAA) and 4-vinylpyridine (4-VP), ethylene glycol dimethacrylate (EDGMA), azobis-(isobutyronitrile) (AIBN), tetrahydrofuran (THF), and poly(heptadecafluorodecyl methacrylate) (PHDFDMA) were used as templates, functional monomers, cross-linker, initiator, porogen and dispersion agent respectively. Apart from that, molecularly imprinted polymers via precipitation polymerization process was carried out for the specific extraction of paracetamol in human urine samples. Two analytical methods were used for the determination of paracetamol in which molecular imprinting solid phase extraction (MI-SPE) is combined with bio-dispersive liquid-liquid microextraction (Bio-DLLME) (Abbasi, Haeri and Sajjadifar, 2019). The recovery of paracetamol from human urine samples was obtained from 96 to 103.8%. The synthesis of MIP was conducted using paracetamol as template, methacrylic acid as functional monomer, ethylene glycol dimethacrylate (EDGMA) as cross-linker, and azobis-(isobutyronitrile) (AIBN) as initiator. In the previous study, molecularly imprinted polymers based voltammetric

sensor has been prepared for the selective uptake of acetaminophen (Menon, Jesny and Girish Kumar, 2018). The MIP was synthesized via electropolymerization method of o-aminophenol using acetaminophen as template and it is conducted on the surface of gold nanoparticles (AuNPs) modified gold electrode (GE) (AuNPS/GE) surface. The sensor has been successful to selectively uptake acetaminophen in synthetic solution and urine sample. Yoon and Byun, (2013a) also presented work in synthesize molecularly imprinted polymer via emulsion polymerization with acetaminophen and aspirin as templates. Sodium dodecylsulfate (SDS) is used as emulsifier with distilled deionized water as solvent and potassium persulfate (KPS) as initiator. The different polymerization method used for the selective recognition of acetaminophen which been described are tabulated in Table 2.2.

Table 2.2: Different polymerization method for the specific uptake of acetaminophen

MIP	Polymerization method	Target molecule	Application	Reference
MIP-BAW sensor for determination of paracetamol	Bulk polymerization	Acetaminophen	BAW sensor	(Tan *et al.*, 2001)
MIP with assisted supercritical CO_2	Dispersion polymerization	Acetaminophen and aspirin	HPLC	(Yoon and Byun, 2013a)
MIP as adsorbent for solid phase extraction	Precipitation polymerization	Acetaminophen	MI-SPE and Bio-DLLME	(Abbasi, Haeri and Sajjadifar, 2019)
MIP based voltammetric sensor	Electropolymerization	Acetaminophen	Electrochemical sensor	(Menon, Jesny and Girish Kumar, 2018)
MIP sensor for determination of acetaminophen and aspirin	Emulsion polymerization	Acetaminophen and aspirin	HPLC	(Yoon and Byun, 2013b)

2.4 Precipitation polymerization

Precipitation polymerization is one the effective method to prepare MIP compared to other method such as emulsion polymerization. The use of emulsifier in emulsion polymerization would interrupt the effectiveness of recognition and binding of the selected molecules (Karnka, Chaiyasat and Chaiyasat, 2017). The precipitation polymerization method is normally conducted using only dilute monomer solution at 2-5 w/w% monomer concentration to prevent the occurrence of macrogelation (Renkecz, László and Horváth, 2014). The work of Dai *et al.* (2011) designed a water-compatible MIPs via precipitation polymerization method. Diclofenac (DFC), 2-vinylpyridine (2-VP), ethylene glycol dimethacrylate (EGDMA) and toluene were used as template, functional monomer, cross-linker and porogen respectively. The MIP demonstrated a great attraction to DFC in aqueous media with a binding site capacity of 324.8 mg/g and it been used as adsorbent in solid-phase extraction for the removal of DFC from environmental water samples. A high recovery of DFC from tap water, river water and wastewater samples were obtained at 96.1%. Synthesize of molecular imprinted polymers particles by precipitation polymerization method without post treatment had been reported by (Karnka, Chaiyasat and Chaiyasat, 2017). Melamine, methacrylic acid (MAA), divinylbenzene (DVB) or ethylene glycol dimethacrylate (EDGMA), and acetonitrile (ACN) were used as template, functional monomer, cross-linker and porogen respectively. Approximately 20 mg/g MIP of binding efficiency was obtained using both cross-linkers (DVB and EDGMA) with melamine. Similar polymerization method was described by (Manzoor, Buffon and Rossi, 2015). In this study, MIPs for the recognition of fluconazole from commercial pharmaceutical samples was successfully synthesized via precipitation polymerization method. Fluconazole, methacrylic acid (MAA), ethylene glycol dimethacrylate (EDGMA), 2,2'-azo-bis-

isobutyronitrile (AIBN) were used as template, monomer, cross-linker and initiator respectively. A recovery of 91% of fluconazole was obtained using fluconazole molecularly imprinted polymers (FLUMIP) with solid phase extraction. The precipitation polymerization has showed a great attention among researcher to be used as the method for the preparation of MIPs. The benefits and drawbacks of precipitation polymerization method are tabulated in Table 2.3.

Table 2.3: Advantages and disadvantages of precipitation polymerization method for the synthesis of MIP (Adumitrăchioaie et al., 2018)

Method	Advantages	Disadvantages
Precipitation polymerization	- Produced good yield of regular shape MIP beads - The individual growth of polymeric chains to microspheres - Do not necessary added porogen agents during the reaction - Easy operation and required shorter times	- Required a large enough polymer chains to be insoluble in the reaction mixture before the precipitation is taking place

2.5 Choice of monomer, cross-linker and solvent

The functional monomers are selected to bind with the target molecule as template for the creation of highly stable polymers network with capability of molecular recognition. Therefore, the decision on choosing monomer is very crucial for obtaining highly specific imprinting sites for the template molecule. The common monomers used for the preparation of MIPs are methacrylic acid (MAA), trifluoromethyl acrylic acid (TFMAA), acrylamide, 2-hydroxyethylmethacrylate (HEMA), 4-vinylpyridine (4-VPY), and 2-vinylpyridine (2-VPY) (Vasapollo *et al.*, 2011). For the selectively uptake

of acetaminophen using supercritical fluid polymerization, 4-VPY has showed a higher binding yield and affinity compared to MAA (Yoon and Byun, 2013b). From the study, 4-VPY has higher value of imprinting-induced promotion of binding (IPB) than MAA since it can have a strong interaction with electron-deficient aromatic rings, acid-base interactions and hydrogen bond acceptance. Therefore, 4-VPY has better affinity and selectivity compared to MAA due to easy interaction of monomer with the template via acid-base interactions and hydrogen bond interaction.

A study from Liu *et al.* (2010) has demonstrated the effect of monomer to the affinity for paracetamol (PR) and the imprinting factor (IF). Paracetamol, trimethylolpropane trimethacrylate (TRIM) and azo-bis(isobutyronitrile) (AIBN) were used as template, cross-linkers and initiator respectively, with itaconic acid (IA), methacryclic acid (MAA) and acrylamide (AA) as monomer. The adsorption of MIP with different monomer was conducted in a shaker operating at 150 rpm with 1 $g.1^{-1}$ of PR solution at 25 ℃ for 10 hours. It was observed that IA has the highest affinity with value of 47.95 mg/g compared to the synthesized using MAA (30.34 mg/g) and AA (39.70) as monomer. Similar result was observed for the IF in which IA has the larger value (1.49) compared to MAA (1.08) and AA (1.25). The affinity and IF of MIP was correlated to the interaction energy (ΔE) where it is assumed that the highest ΔE with the template will give the highest affinity of MIP. The relation between ΔE and affinity or IF was plotted in Figure 2.2.

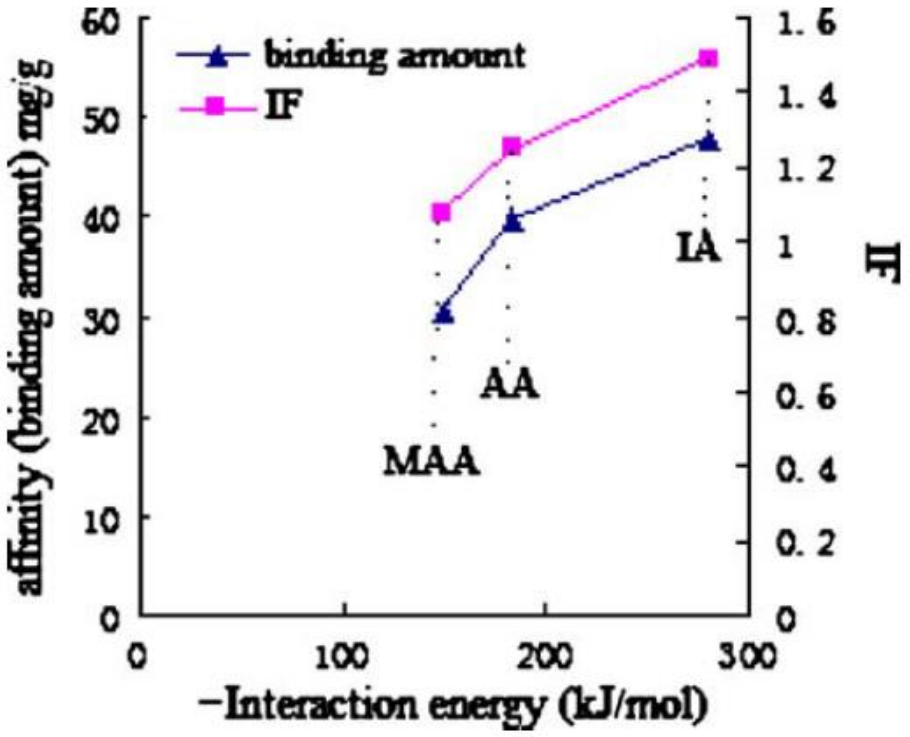


Figure 2.2: The relation of interaction energy with the affinity of IF (Liu et al., 2010)

The morphology of the polymers is changed when different monomer used in the production of MIPs. The characterization of polymers was evaluated by scanning electron microscopy (SEM). The effect of functional monomer on the morphology can be observed in the study reported by (Renkecz, László and Horváth, 2014). Two different characteristic of polymer morphologies were obtained through precipitation polymerization method including monodispere microspheres and segmented microparticles. The polymerization using 4-VPY as monomer with TRIM in the mixture of chloroform and paraffin oil produced a smooth monodisperse microsphere. Another polymerization method using MAA as monomer with EGDMA in the presence of toluene/paraffin oil resulted in a segmented microparticles. The morphology of MIPs for using both different monomer and cross-linker (MAA/EDGMA and 4-VPY/TRIM) are shown in Figure 2.3.

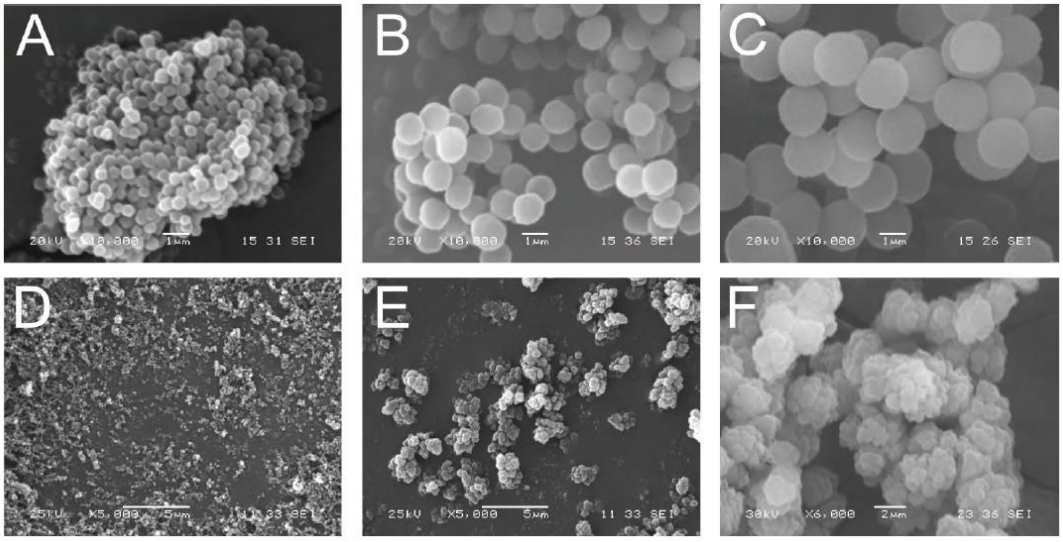


Figure 2.3: Polymer particles obtained in the 4-VPY/TRIM/choloroform:paraffin oil system after A) 10 minutes; B) 1 hour; C)24 hours and particles in the MAA/EDGMA/toluene:paraffin oil system after D) 1 hour; E) 3 hours; F) 24 hours, using SEM measurements. The bars in A-C, in D-E and in F denote 1, 5, 2 μm respectively (Renkecz, László and Horváth, 2014)

However, the use of MAA as functional monomer in the preparation of MIP was quite extensive due to its functionality which can acts as hydrogen bond donor and acceptor. In the study from (Abdel Ghani *et al.*, 2016), it was revealed that the use of MAA for the preparation of itopride hydrochloride (Itoh) MIP have a higher interaction between functional monomer and template compared to other monomers. The experiment was conducted by using three different functional monomers namely, methacrylic acid (MAA), 4-vinylpyridine (4-VP) and acrylamide (AM) for the selection of the most preferable monomer for the MIP synthesis. Similarly, MAA has been observed to be a suitable functional monomer compared to methyl methacrylate (MMA) (Golker *et al.*, 2014). The study had carried out a total of 16 MIPs synthesis using bupivacaine as template molecules, ethylene glycol dimethacrylate (EDGMA) as cross-linker, methacrylic acid (MAA) or methyl methacrylate (MMA) as monomer. From the result obtained, it exhibited that the hydrogen bond between bupivacaine and MAA is

higher compared to the bonding between bupivacaine and MMA. This is due to the acidic proton from MAA which provide an additional interaction to the hydrogen bond between bupivacaine and MAA. The study also revealed that the increasing of MAA molar fraction can lead to a higher binding capacities.

The choice of cross-linker is also important for the synthesis of MIP due to its capability to control the morphology of polymer matrix. Cross-linker also able to provide a stable imprinted binding sites and mechanical stability to the polymer matrix for retaining the molecular recognition capability of MIP. The common cross-linker used are ethylene glycol dimetahcrylate (EDGMA) and trimethylolpropane trimethacrylate (TRIM) (Vasapollo *et al.*, 2011). Some authors discovered that cross-linker has an influence to the size of MIP in which increasing concentration of EDGMA lead to more cloudy solution that result to a bigger size of MIP particles. This is due to a better binding of oligomers was expected with increased of cross-linker concentration that lead to development of larger polymer matrix (Shahar, Tal and Mandler, 2016). The study from (Shoravi *et al.*, 2010), it has been found that the most stable interactions was displayed between acidic proton of MAA and carbonyl oxygen atoms of EDGMA. The average hydrogen bond occupancies of the interaction between EDGMA and MAA were also produce the highest values compared to cross-linking agent TRIM.

Furthermore, the solvents plays also a vital role during the imprinting process of MIP. The solvent or normally termed as porogen aids in gather monomer, template, initiator and cross-linker into one phase in the polymerization process. It also has responsibility to form the pores in MIP and enhanced the formation of monomer-template complex. Common solvents are acetonitrile, chloroform, toluene and dichlorometane. Toluene or chloroform are the solvents with less polarity has capability

to increase the creation of hydrogen bonds while more polar solvent have tendency to distrupt the hydrogen bonds (Vasapollo *et al.*, 2011). Nevertheless, some authors has showed that the use of polar solvents such as acetonitrile able to form a strong monomer-template complex. For example, a non-covalent MIP were successful prepared using acetonitrile as solvents and been used as MIP-BAW sensor for the detection of paracetamol in human serum and urine samples (Tan *et al.*, 2001). The combination of polar solvent (acetonitrile) and apolar solvent (toluene) also revealed a good effect on MIP in term of thermal stability (Zhang *et al.*, 2019).

2.6 Ratio between template:monomer:cross-linker

The influence of feed formulation such as the ratio of template to functional monomer (T:fM) and the ratio of functional monomer to cross-linker (fM:X) was studied by (Fremielle Lim and Holdsworth, 2018). Methacryclic acid (MAA) and ethylene glycol dimethacrylate (EDGMA) were used as monomer and cross-linkers respectively in the preparation of caffeiene (CAF) and theophylline (THP) MIPs via precipitation polymerization. Various T:fM ratios ranging from 1:2 (TM2), 1:6 (TM6) to 1:8 (TM8) were used to study the effect on the binding efficiency and selectivity of MIPs. It was observed that both TM2 and TM8 produce comparable binding capacities although result different level of template concentration. The formation of template-monomer complex is highly favoured at high concentration of MAA (TM8) influence the increasing of template incorporation. For the functional monomer to cross-linker ratio, variation of fM:X ranging from 1:2 (MX2, 33% MAA), 1:5 (MX5, 17% MAA) to 1:10 (MX10, 9% MAA) were investigated. The result showed that MX10 give the most favourable binding result regardless MX2 produce the binding capacities and template incorporation for both CAF and THP MIPs. Thus, we can assured the

importance of polymer rigidity on the MIP's performance at higher cross-linker content. The study also the most favourable initiator to total monomer (I:tM) was 1:5 in which high initiator content result a faster polymerization reaction.

2.7 Comparison between MIP and NIP

The preparation of molecularly imprinted polymers (MIPs) is followed by the synthesis of control polymer which is non-imprinted polymers (NIP). The preparation of NIP is carried same as MIP but without the addition of template. The physical and chemical differences between MIPs and NIPs was discussed in previous study (Ndunda, 2020). It was showed that the specific surface are of MIP is higher compared to NIP. MIP also have larger pore size which make it more porous than NIP. Besides, it also been discovered that the pore volume and pore area of MIPs is higher compared to NIPs. Thus, more formation binding sites were observed in the polymer matrix due to its high pore volume. Furthermore, the binding properties of MIP and NIP also has significant difference between each other in term of binding affinity. The binding properties NIP has influence toward the binding properties of MIP in which MIP will displays a weak imprinting effect when NIP displays no binding properties toward target molecule. Whereas, MIP will have excellent binding properties if NIP displays binding properties toward target molecule.

2.8 Polyethersulfone (PES) membrane

Membrane can categorized into three type of membrane which is polymeric, ceramic and biological membrane. Although there are various type of membrane in many applications, polymeric membrane has become the common membrane used in the majority of research membrane area. Besides, polymeric membranes also used extensively in wastewater treatment application due to its low cost, easy preparation

and able to remove waste at high efficiency. (Ladewig and Al-Shaeli, 2017). Polymeric membranes is commonly comprised of hydrophobic polymers such polysulfone (PS), polyethersulfone (PES), polypropylene (PP) and polyvinylidene fluoride (PVDF). Among other polymeric membranes, PES is the most preferable choice by researchers for the ultrafiltration due to excellent mechanical strength, outstanding thermal and chemical stability (Huang *et al.*, 2012). In the previous study, PES membrane was prepared for the wastewater filtration which focused on the filtration of activated sludge (Maximous *et al.*, 2009). Moreover, PES membranes also been utilized in the membrane bioreactor (MBR) treatment for the removal of pharmaceutical compounds including acetaminophen, ketoprofen, naproxen, and trimethoprim (Schröder *et al.*, 2012).

2.9 Fabrication of membrane via phase inversion method

Numerous method available for the preparation of synthetic membranes includes phase inversion, sol-gel process, stretching films, track-etching and sintering of powders (Ladewig and Al-Shaeli, 2017). Phase inversion is the most common method used for the synthesis of polymeric membranes in all sort of morphology including asymmetric and symmetric. In this method, there are different process involved such as thermal precipitation, solvent evaporation, dry phase inversion and precipitation from the vapour phase. The phase inversion process is the process where the liquid phase is changed to solid phase. A thermodynamically stable polymer solution was produced during the phase inversion process. This solution is referred to a liquid-liquid demixing in which the cast polymer films separates to polymer-rich phase (membrane matrix) and polymer-lean phase (membrane pores). During the phase inversion process, the polymer will be soluble in at suitable temperature in the solvent and produced a solid state (membrane) via precipitation. Among other phase inversion

techniques, immersion-precipitation is the common process in which its mechanism can be represented in polymer/solvent/nonsolvent phase diagram as shown in Figure 2.4 (Buonomenna *et al.*, 2011).

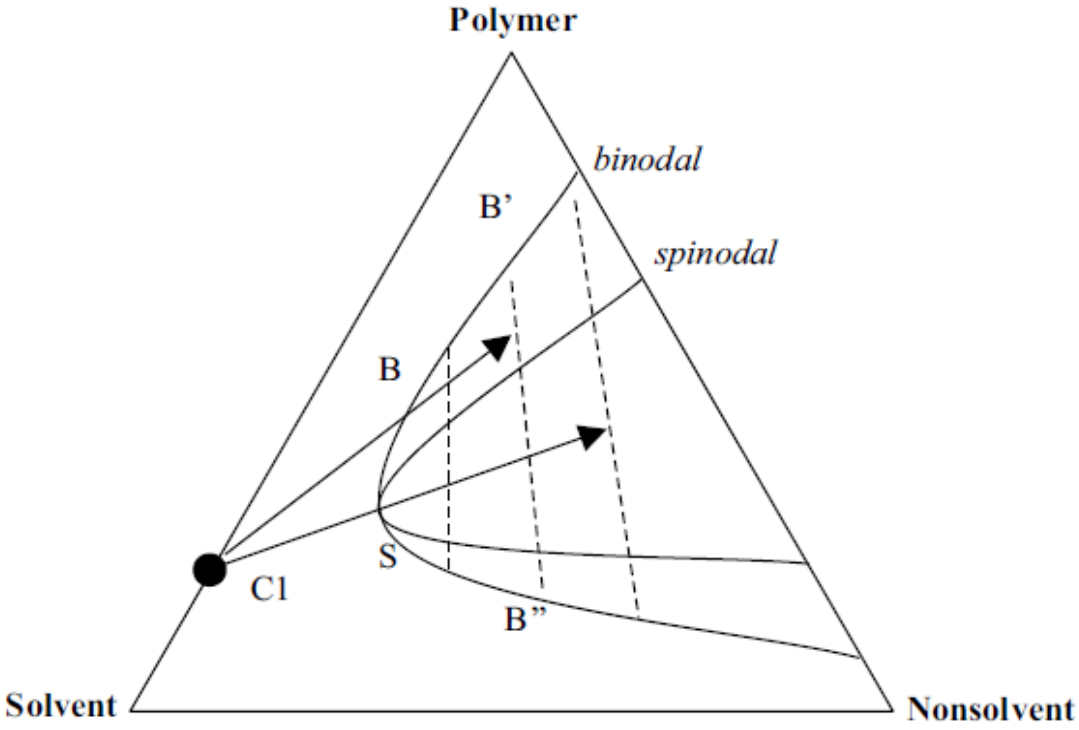


Figure 2.4: Ternary phase diagram (solvent/polymer/nonsolvent) for membrane formation by immersion-precipitation (Buonomenna *et al.*, 2011)

There are two types of phase inversion used for the membrane fabrication which is dry phase inversion and wet phase inversion. Dry phase inversion is the process to create porous membrane via evaporation of solvent from the mixture of polymer, solvent and nonsolvent. This process did not required coagulation bath to make a solid membrane. Hydrophilic PES membrane was been prepared via dry phase inversion method (Arahman, 2014). On the other hand, the wet phase inversion is the process to fabricate membrane by immersion-precipitation techniques using non-solvent as a coagulation media. Three different polymers such as polysulfone (PSf),

polyethersulfone (PES) and polyvinylidene fluoride (PVDF) were used to prepared flat sheet ultrafiltration membranes via wet phase inversion method (Keskin *et al.*, 2020). Maximous *et al.* (2009) reported the wet phase inversion method is used to prepare pure PES membrane and Al_2O_3/PES membrane for the wastewater application.

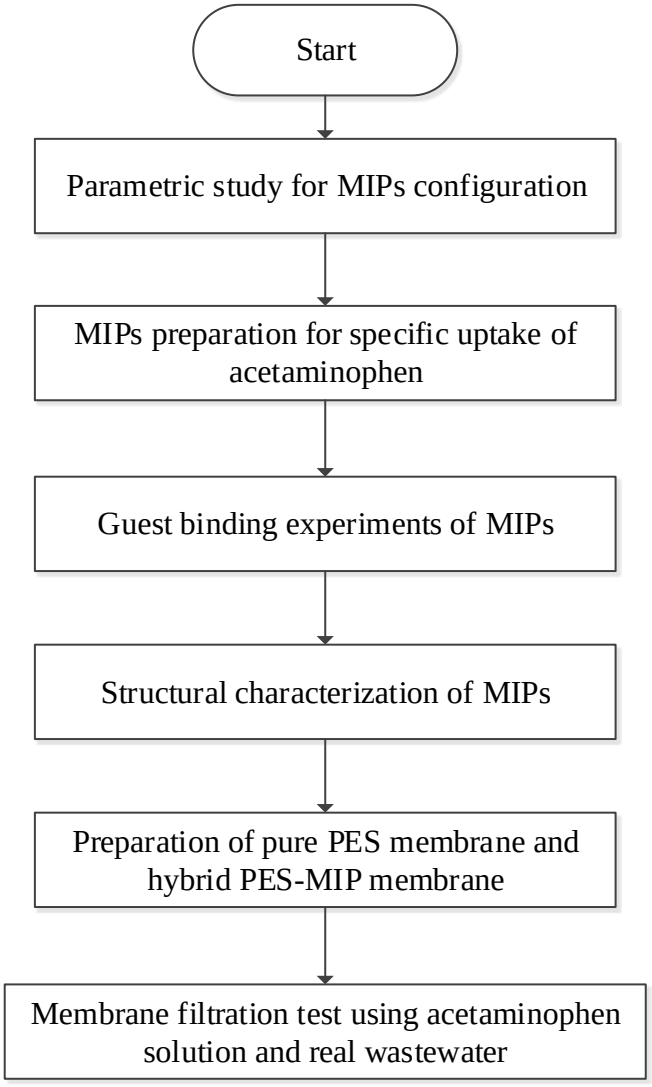
2.10 Antifouling properties of membrane

The major problem arise during the performance of membrane will be membrane fouling. Membrane can be susceptible toward fouling due to deposition of particulate, precipitation or chemical contamination. Particulate matter may cause the membrane fouling through the contamination or adsorption in the pores which lead to the formation of cake layer on the membrane surface (Galloway, Gottberg and Systems, 2006). PES membrane which is comprised of hydrophobic polymer can also easily prone to fouling and have low membrane flux although it has many numerous tremendous properties such as good thermal resistance and easy preparation (Maximous *et al.*, 2009). Membrane fouling will give great impact on application and also increase the operating cost of ultrafiltration membranes. There are two types of membrane fouling which is reversible and irreversible fouling. Backwashing and cross-flushing can be the solutions for the reversible fouling. For the irreversible fouling, it can be removed by adding chemical reagents and the repetition of chemical cleaning may decrease the membrane performance. The membrane fouling could lead to the reduction of membrane flux, high energy consumption, additional cost due to chemical cleaning processes and reduce the membrane life (Huang *et al.*, 2012). Numerous studies have conducted to study the antifouling properties of membrane. Evaluation of fouling properties of pure PES membrane and Al_2O_3/PES membrane was conducted in the previous study (Maximous *et al.*, 2009).

CHAPTER 3

METHODOLOGY

The overview of research activity are illustrated in Figure 3.1. Preparation of molecularly imprinted polymers (MIPs) via precipitation polymerization method was carried out in the laboratory. Besides, the fabrication of polyethersulfone (PES) membrane by wet phase inversion method was conducted in the laboratory. The characterization for both MIPs and PES membrane were performed and discussed in this chapter.



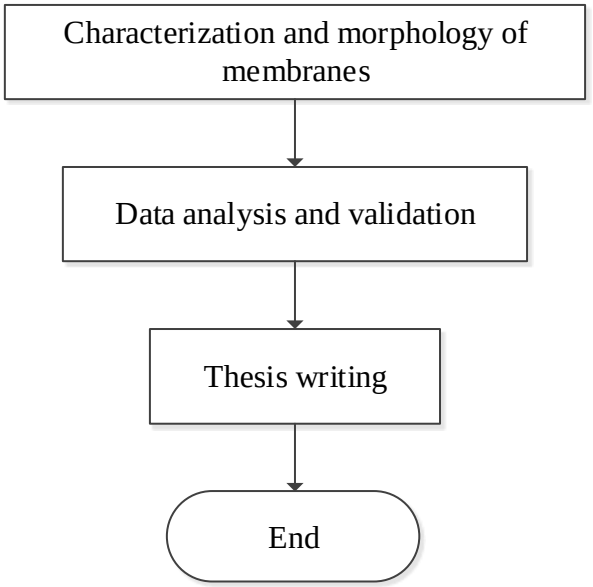


Figure 3.1: Flow diagram of research activity on MIPs and PES membrane

3.1 Materials

Acetaminophen (analytical standard), methacrylic acid (MAA) (purity: 99%, contains 250 ppm monomethyl ether hydroquinone as inhibitor), ethylene glycol dimethacrylate (EDGMA) (purity: 98%, contains 90-110 ppm monomethyl ether hydroquinone as inhibitor), 1,1'-Azobis(cyclohexanecarbonitrile) (ABCN) (purity: 98%), methanol (anhydrous, 99.8%) were purchase from Sigma Aldrich. Toluene (analytical grade), acetonitrile (ACN), 1-methyl-2-pyrrolidone (NMP) were purchased from Merck. Polyethersulfone (PES Ultrason E6020 P with M_w = 58,000 g/mol) was purchased from BASF. Pure nitrogen gas was used during the preparation of MIP. Distilled water was used for washing process of MIP and during the phase inversion process of membrane. Liquid nitrogen was used for the membrane cracking process.

3.2 Preparation of acetaminophen imprinted polymer (Act-MIP)

First and foremost, we had conducted the study on the parameters for the configuration of MIP for the uptake of acetaminophen. The parameters include the ratio between template:monomer:cross-linker and type of monomer, cross-linker and solvent used. The preparation of acetaminophen MIP are performed through precipitation polymerization method based on the method by Ahmad et al (2018).

The acetaminophen imprinted polymers (Act-MIP) was prepared by dissolving template (acetaminophen) in the porogen in a 50 ml glass bottle. 5ml of toluene and 10 ml of acetonitrile were used as the porogen. Functional monomer (MAA), crosslinker (EDGMA) and initiator (ABCN) were then added to the mixture and purged with pure nitrogen for 5 min. The glass bottle was sealed with parafilm and placed in water bath at 65 °C (stirred at level 5) for 20 h. As a control polymer, non-imprinted polymer (NIP) was simultaneously prepared in the same procedure but without the addition of template.

The mole ratio of template:monomer was firstly maintained at 1:58. Three acetaminophen imprinted polymer with different mole ratio of cross-linker were prepared as represented in Table 3.1. The weight of template (acetaminophen) and initiator (ABCN) used for the preparation of MIPs were maintained at 0.0183 g and 0.181 g respectively.

Table 3.1: Different cross-linker mole ratio of acetaminophen-MIPs

Polymer	Cross-linker ratio (mol)	Volume of cross-linker (μL)
MIP 1	3	94.2
MIP 2	15	471
MIP 3	30	942