POLYETHERSULFONE MIXED MATRIX MEMBRANE CONTAINING IMPRINTED ZEOLITE FOR CRESOL REMOVAL IN HEMODIALYSIS APPLICATION

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A thesis submitted in fulfilment of the
requirements for the award of the degree of
Doctor of Philosophy

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JUNE 2020

DEDICATION

This thesis is dedicated to my father, Mr Budi Raharjo; my mother, Mrs Sri Rahayuningsih; my father in law, Mr Haryoto; my mother in law, Sri Utanti; Special for my wife, Herdina Mariyanti; my Son, Muhammad Farhan Raharjo; and my Daughter, Nindya Prameswari Raharjo

For their advices, their patience, their prays, their motivation and their faith.

Thank you for always understood.

ACKNOWLEDGEMENT

In preparing this thesis, I was in contact with many people, researchers, academicians, and practitioners. They have contributed towards my understanding and thoughts. In particular, I wish to express my sincere appreciation to my main thesis supervisor, Professor Datuk Dr. Ts. Ahmad Fauzi Ismail, for encouragement, guidance, critics and friendship. I am also very thankful to my co-supervisor Associate Professor Dr. Ts. Mohd. Hafiz Dzarfan Othman for their guidance, advices and motivation. Without their continued support and interest, this thesis would not have been the same as presented here.

I am also indebted to Universitas Airlangga (UNAIR) Surabaya Indonesia for funding my Ph.D study and Advanced Membrane Technology Research Centre (AMTEC), Universiti Teknologi Malaysia (grant no: Q.J130000.2409.01G46) for research funding and facilities. Technician at AMTEC UTM, also deserve special thanks for their assistance in supplying the relevant experiment in laboratory.

My fellow postgraduate student should also be recognised for their support. My sincere appreciation also extends to all my colleagues and others who have provided assistance at various occasions. Their views and tips are useful indeed. Unfortunately, it is not possible to list all of them in this limited space. I am grateful to my family members.

ABSTRACT

The adequacy of uremic toxins removal on hemodialysis treatment is essential to be achieved for kidney failure disease patient, as poor removal leads to heart failure, hypertension, and stroke. The combination of adsorption and diffusion process has become very advantageous for hemodialysis membrane. By this mechanism, the urea as water soluble uremic toxins (WSUT) and *p*-cresol as protein-bounded uremic toxins (PBUT) could be removed at one time. Therefore, this study aimed to develop the novel imprinted zeolite by *p*-cresol (IZC) then incorporated into polyethersulfone (PES) and poly(vinyl pyrollidone) (PVP) to produce hollow fiber mixed matrix membrane (HF-MMM). IZC was synthesized from sodium aluminate, NaOH, H_2O and SiO_2 through aging and hydrothermal process with an initial composition of $10SiO_2:Al_2O_3:4Na_2O:180H_2O$ by using imprinting technology and *p*-cresol as a template. Based on the properties and performance achieved, IZC could increase the selectivity to adsorb *p*-cresol 4.30 times greater compared to synthesized Zeolite Y (ZeoY-S). Adsorption study proved that IZC could adsorb *p*-cresol 2.5 and 3.5 times higher than ZeoY-S and commercialized zeolite Y (ZeoY-C), respectively. This is because the pore size of IZC had been successfully printed to *p*-cresol. The Brunauer-Emmet-Teller and transmission electron microscopy characterization proved that imprinting process was successfully applied. The investigation by isotherm and kinetics models showed that IZC was sensitive to attract the adsorbate, classifying it as having a strong adsorption behavior. Accordingly, the IZC is very promising to be applied as adsorbent in hemodialysis treatment. In the second phase, IZC as *p*-cresol's adsorbent was incorporated into PES-based polymeric membrane with small addition of PVP to produce HF-MMM by using dry/wet spinning process. The effect of air gap distance between spinneret and coagulant bath and percentage loading for PES, PVP, and IZC were studied and optimized to obtain the best performance of HF-MMM. The 40 cm of air gap distance, 16 wt.% of PES, 2 wt.% of PVP, and 1 wt.% of IZC loading were able to produce superior hemodialysis membrane. These optimized parameters showed sufficient uremic toxins removal i.e. 60.74% of urea, 52.35% of *p*-cresol in phosphate buffer saline solution and 66.29% of *p*-cresol in bovine serum albumin solution for 4 hours permeation by using dialysis system. These HF-MMMs also achieved pure water flux of 67.57 $Lm^{-2}h^{-1}bar^{-1}$ and bovine serum albumin rejection of 95.05%. Therefore, this membrane has been proven to be able to clean up WSUT and PBUT through an one-step process. Moreover, as compared neat PES membrane, MMM was able to remove *p*-cresol 186.22 times higher. Then, capability of IZC to adsorb *p*-cresol decreased to around 69% by changing the form of adsorbent from powder to composite in the membrane. By leaching study, it was obtained that percentage of zeolite leaching was less than 1 mgL^{-1} and categorized safe. In the final phase of the study, the HF-MMM developed was evaluated in terms of biocompatibility test, that is hemocompatiblity by using protein adsorption, platetels adhesion, blood clotting time test, activated partial thromboplastin time, prothrombin time, and cytotoxicity evaluation by using 3-(4,5-dimethylthiazol-2yl)-5(3-carboxymethoxy phenyl)-2-(4-sulfophenyl)-2H-tetrazolium (MTT assays). From the biocompatibility evaluation HF-MMM was observed to possess less protein adsorption, less activated state of the adhered platelets, non-toxic quality for red blood cells, and can prolong the clotting time and percentage of viability for more than 60%. These results proved that HF-MMM developed is safe for hemodialysis application.

ABSTRAK

Proses pembuangan toksin uremik pada rawatan hemodialisis yang mencukupi sangat penting bagi pesakit yang mengalami kegagalan fungsi buah pinggang, kerana ianya akan menyebabkan kegagalan fungsi jantung, tekanan darah tinggi, dan strok. Gabungan proses penjerapan dan peresapan telah memberi banyak faedah pada proses hemodialisis. Urea sebagai toksin uremik larut air (WSUT) dan *p*-cresol sebagai toksin uremik yang terikat dengan protein (PBUT) boleh dibuang sekaligus dengan mekanisme ini. Oleh itu, kajian ini bertujuan untuk membangunkan zeolit dengan teknik cetakan khas bagi *p*-cresol (IZC) yang kemudian digabungkan dengan polietersulfon (PES) dan poli(vinilpirolidone) (PVP) untuk menghasilkan membran matriks campuran (HF-MMM). IZC disintesis daripada natrium aluminat, NaOH, H_2O dan SiO_2 melalui proses penuaan dan hidrotermal, dengan komposisi awal $10SiO_2$: Al_2O_3: $4Na_2O$: $180H_2O$ menggunakan teknologi pencetakan dengan *p*-cresol sebagai templat. Berdasarkan ciri-ciri dan prestasi yang dicapai, IZC dapat meningkatkan kadar jerapan *p*-cresol sehingga 4.30 kali lebih tinggi daripada zeolite yang disintesis (ZeoY-S). Manakala, melalui proses penjerapan, IZC terbukti dapat menjerap *p*-cresol masing-masing pada 2.5 dan 3.5 kali ganda lebih tinggi daripada ZeoY-S dan zeolit yang dikomersialkan (ZeoY-C). Hal ini adalah disebabkan oleh saiz liang IZC yang telah berjaya dicetak kepada *p*-cresol. Pencirian Brunauer-Emmet-Teller dan mikroskop penghantaran elektron telah membuktikan bahawa proses pencetakan telah berjaya dilaksanakan. Kajian berdasarkan model isoterma dan kinetik menunjukkan bahawa IZC adalah penjerap yang sensitif dan boleh diklasifikasikan sebagai mempunyai sifat penjerapan yang kuat. Oleh yang demikian, IZC adalah sesuai untuk digunakan sebagai penjerap di dalam rawatan hemodialisis. Dalam fasa kedua, IZC sebagai penjerap *p*-cresol telah dimasukkan ke dalam membran polimer berasaskan PES dengan penambahan kecil PVP bagi menghasilkan HF-MMM dengan menggunakan proses pemutar kering/basah. Kesan jarak jurang udara antara spinneret dan tempat pembekuan serta peratusan muatan untuk PES, PVP, dan IZC telah dikaji dan dioptimumkan bagi memperoleh prestasi terbaik. Jarak 40 cm jarak udara, 16% PES, 2% PVP, dan 1% IZC mampu menghasilkan membran hemodialisis yang unggul. Parameter yang telah dikaji menunjukkan pembuangan toksin uremik yang mencukupi iaitu 60.74% urea, 52.35% *p*-cresol dalam larutan garam fosfat dan 66.29% *p*-cresol dalam larutan albumin serum bovine selama 4 jam dengan menggunakan sistem dialisis. HF-MMM ini juga boleh mencapai kelajuan fluks air pada takat 67.57 $Lm^{-2}h^{-1}bar^{-1}$ dan penolakan albumin serum bovine sebanyak 95.05%. Oleh itu, membran ini terbukti dapat membuang WSUT dan PBUT sekaligus. Selain itu, perbandingan di antara membran PES dan MMM, MMM mampu membuang *p*-cresol 186.22 kali lebih tinggi. Keupayaan IZC untuk menjerap *p*-cresol berkurang sekitar 69% apabila dicampurkan dalam membran komposit jika dibandingkan dengan dalam bentuk serbuk. Dapatan daripada kajian pelunturan menunjukkan pelepasan zeolit kurang daripada 1 mgL^{-1} dan dikategorikan selamat. Pada fasa akhir kajian, HF-MMM telah dinilai dari segi ujian keserasian-bio, iaitu keserasian hemo dengan menggunakan ujian penjerapan protein, lekatan platelet, ujian masa pembekuan darah, masa tromboplastin separa teraktif, masa protrombin, dan penilaian sitotoksisitas dengan menggunakan 3-(4,5-dimetilthiazol-2yl)-5(3-karboksimetoksifenil)-2-(4-sulfofenil)-2H-tetrazolium (Ujian MTT). Berdasarkan penilaian keserasian-bio, HF-MMM hanya menjerap sedikit protein, kurang aktif untuk lekatan platelet, bukan toksik untuk sel darah merah, memanjangkan masa pembekuan darah dan masa hidup sel limfosit lebih dari 60%. Keputusan ini membuktikan bahawa membran yang dibangunkan adalah selamat untuk aplikasi membran hemodialisis.

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LIST OF ABBREVIATIONS

Å	-	Angstrom
AC	-	Activated carbon
AFM	-	Atomic force microscopy
AGE	-	Advanced glycatin end products
AKI	-	Acute kidney injury
AMTEC	-	Advance membrane technology research center
APTT	-	Activated partial thromboplastin time
ATR	-	Attenuated total reflection
BET	-	Brunauer-Emmet-Teller analysis
BSA	-	Bovine serum albumin
BUN	-	Blood urea nitrogen
CC_{50}	-	Cytotoxicity concentration 50%
CI	-	Crystallinity index
CKD	-	Chronic Kidney Disease
CMPF	-	3-carboxyl-4-methyl-5-propyl-2-furanpropionic acid
CPFA	-	Couple plasma filtration adsorption
Da	-	Dalton
DAC	-	Diacetate cellulose
DLHF	-	Dual layer hollow fiber
DMAB	-	p-Dimethylaminobenzaldehyde
DMAc	-	Dimethylacetamide
DMF	-	Dimethylformamide
DMSO	-	Dimethyl sulfoxide
D-PBS	-	Dulbecco phosphate buffer saline
EDX	-	Enery dispersive X-Ray
EVAL	-	Ethylene-vinyl alcohol copolymer
FDA	-	Food and drugs administration
FFT	-	Fourier transform
FESEM	-	Field emission scanning electron microscopy
FTIR	-	Fourier transform infrared

GFR	-	Glomerular filtration rate
GNPs	-	Graphene nanoplatelets
HAP	-	Hydroxyapatite
HD	-	Hemodialysis
HFM	-	Hollow fiber membrane
HF-MMM	-	Hollow fiber-mixed matrix membrane
HFR	-	Hemodiafiltration
HPHD	-	Hemoperfusion-hemodialysis
HP	-	Hemoperfusion
HPLC	-	High performance liquid chromatography
HR-TEM	-	High resolution-transmission electron microscopy
ICP-MS	-	Inductively coupled plasma mass spectrometry
ID	-	Internal diameter
IFFT	-	Inverse fourier transform
IPD	-	Intra-particle diffusion
IR	-	Infrared
ISO	-	International organization of standardization
IUPAC	-	International union of pure applied chemistry
IZ	-	Imprinted-zeolite
IZC	-	Imprinted-zeolite Y/*P*-cresol
IZA	-	International zeolite database
MAS-NMR	-	Magic angle spinning-nuclear macgenic resonance
MEM	-	Minimum essential media
MFI	-	Microporous zeolite silicate
MIP	-	Molecularly imprinting polymer
MMM	-	Mixed matrix membrane
MTT	-	3-(4,5-dimethylthiazol-2yl)-5(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium
MW	-	Molecular weight
MWUT	-	Middle-molecular weight uremic toxin
NaY	-	Faujasite Na-zeolite Y
n.d.	-	Not detected
NIZ	-	Non-imprinted zeolite

Nm	-	Nanometer
NMP	-	N-methylpyrrolidone
NMR	-	Nuclear magnetic resonance
OD	-	Outer diameter
PA	-	Polyamide
PAN	-	Polyacrylonitrile
PBS	-	Phosphate buffer saline
PBUT	-	Protein bounded uremic toxin
PEG	-	Polyethylene glycol
PES	-	Polyethersulfone
PFD	-	Paired filtration dialysis
PLA	-	Poly (lactic acid)
PLA-PHEMA	-	Poly (lactic acid)-block-poly (2-hydroxyethyl methacrylate)
PLCL-SC	-	Poly (L-lactic-co-ε-caprolactone)-sericin
PMMA	-	Polymethymethacrylate
PP	-	Polypropylene
PPP	-	Platelet poor plasma
PPG	-	Polypropylene glycol
PRP	-	Platelet-rich plasma
PSf	-	Polysulfone
PSf-g-PLA	-	Polysulfone-graft-poly(lactic acid)
PT	-	Prothrombin time
PTFE	-	Polytetrafluoroethylene
PVC	-	Polyvinylchloride
PVDF	-	Polyvinylidene fluoride
PVP	-	Polyvinylpyrrolidone
RP-HPLC	-	Reverse-phase high performance liquid chromatography
SAED	-	Selected area electron diffraction
SDS	-	Sodium dodecyl sulfate
SEM	-	Scanning electron microscopy
TAC	-	Triacetate cellulose
TBOT	-	Tetrabutyl orthotitanate
TEM	-	Transmission electron microscopy

TEOS	-	tetraethyl orthosilicate
TGA	-	Thermal gravimetry analysis
TMS	-	Tetramethylsilane
TPAH	-	Tetra-n-propylammonium hydroxide
TT	-	Thrombin time
UTM	-	Universiti Teknologi Malaysia
UV	-	Ultraviolet
Vis	-	Visible
WCA	-	water contact angle
WSUT	-	Water soluble uremic toxin
wt.	-	Weight
XRD	-	X-ray diffraction

LIST OF SYMBOLS

%	-	Percentage
Al_2O_3	-	Aluminium Oxide
$CaCl_2$	-	calcium chloride
cm	-	Centimeter
cm^{-1}	-	Per centimeter
CO_2	-	Carbon Dioxide
$FeCl_3$	-	ferric chloride
g/mol	-	Gram per mol
GO	-	Graphene Oxide
GS	-	Graphene Sheets
h	-	Hour
H_2O	-	Hydrogen Dioxide
HCl	-	Hidrogen Chloride
KBr	-	Sodium Bromide
KCl	-	Potassium Chloride
KH_2PO_4	-	Potassium dihydrogen phosphate
Kv	-	Kilo Volts
m/min	-	Meter per minute
mA	-	Mili Amphere
mg/g	-	Miligram per gram
mg/L	-	Milligram per liter
$MgCl_2$	-	Magensium Chloride
min	-	Minute
mL	-	Mililiter
mL/min	-	Mililiter per minute
n	-	Adsorption capacity
Na_2O	-	Sodium Oxide
$Na_3C_6H_5O_7$	-	Sodium citrate
$NaAlO_2$	-	Natrium Aluminate
NaCl	-	Sodium Chloride

NaH_2PO_4	-	Sodium dihydrogen phosphate
NaOH	-	Sodium Hydroxide
nm	-	Nanometer
q_{ref}	-	Final adsorption amoun
rpm	-	Rotation per minute
^oC	-	Degree celcius
R_i	-	Adsorption factor
SiO_2	-	Silicone dioxide
$t^{1/2}$	-	Square root of time
v/v	-	Volume per volume
wt.%	-	Weight percentage
θ	-	Teta
µL	-	Microliter
µm	-	Micrometer
δ	-	Chemical shift
Ra	-	mean roughness
Rq	-	root-mean-square roughness
Sa	-	mean of surface roughness
Sq	-	root mean square surface roughness

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CHAPTER 1

INTRODUCTION

1.1 Research Background

Membrane technology is widely applied to support the development of science and technology, both in terms of theoretical physics and chemistry. The application starts from secondary human needs, such as water filters and gas separation to primary human needs, such as an artificial kidney. Regarding its application, the membrane technology is used to treat kidney disease via hemodialysis (HD) treatment. Various membranes have been developed by researchers for the HD treatment. There are two types of membranes that are commercially available and widely consumed in hospitals and dialysis clinics to treat patient with a kidney failure.

The first one is low-flux dialysis membrane, in which the membrane can remove much of the water-soluble uremic toxins (WSUT), such as urea, creatinine, and uric acid, among others. However, it is difficult to remove the middle molecular-weight water-soluble of uremic toxins (MWUT), such as Cystatin C, β_2-Microglobulin, and β-Endorphin, as well as protein-bounded uremic toxins (PBUT), such as indoxyl sulfate, *p*-cresol, and phenol (Henrich, 2009). Meanwhile, the MWUT and PBUT are dangerous if accumulated into the blood, as it may cause endothelial or leukocyte dysfunction and exert proinflammatory and hepatotoxic effects that contribute to increased mortality (Ketteler, 2006). The second type of HD membrane is high-flux dialysis membrane. This membrane is able to remove some uremic toxins, which cannot be eliminated by low-flux dialysis membrane (Chelamcharla *et al*., 2005). This membrane use a higher pressure under larger pores compared to low-flux dialysis membrane. Though this, the removal of MWUT and PBUT are forced using high pressure and large pore size (Yamamoto *et al*., 2016). Regarding HD, the adequacy of dialysis fulfilled by a high-flux dialysis compared to the low-flux dialysis has been reported (Oshvandi *et al*., 2014).

Besides the high-flux membrane, another treatment that can be applied to remove MWUT and PBUT in blood purification is by using an adsorption mechanism called hemoperfusion (HP). Basically, the mechanism of HP is the hydrophobic properties of the sorbents or by chemical affinity (Botella *et al*., 2000). Carbon-based adsorbents, such as activated carbon (AC), have been used internally by oral or in extracorporeal devices (Mikhalovsky and Nikolaev, 2006). HP column is a simple device, in which a plastic column is filled by the adsorbent powder. The uncoated and coated charcoal were evaluated as adsorbent to eliminate the MWUT and PBUT. When the uncoated charcoal is used, the MWUT and PBUT can clearly be adsorbed better by the adsorbents than the coated charcoal can. Nonetheless, the main problem is the biocompatibility. The uncoated charcoal is highly incompatible with blood through direct contact, as it adsorbs not only the MWUT and PBUT, but also other proteins that are still needed by the body due to the hydrophobic properties of the sorbent. Based on the previous works, they stated that one hour of HP treatment was as effective as four hours using HD treatment (Yamamoto *et al*., 2016; Cheah *et al*., 2017). In the past, HP is rarely used for blood purification application primarily due to the biocompatibility issue of materials, particles release, and limitation to removing the WSUT despite having a very strong capability to remove MWUT and PBUT. By advanced manufacturing processes and improved biocompatibility, sorbent has an enormous potential to be developed (Ronco, 2006). Hence, the type of materials applied for HP application needs to be improved and innovated. The high selectivity of hydrophilic sorbents might also be very effective and beneficial for eliminate the MWUT and PBUT.

Combination of the strengthness of HD and HP can be very beneficial for blood purification. The module is a polymer membrane used to combine the diffusion and adsorption mechanism at one step called mixed-matrix membrane (MMM) (Tijink *et al*., 2012). MMM exhibits many advantages, such as flexible large-scale operation, simple, time efficiency, minimum membrane fouling and flux decline, and energy saving (Suen, 2015). The principal of MMM is the synergetic of different functions by different materials (Ulbricht, 2006). The purpose of developing this membrane is to harness its time efficiency during HD application, since it is able to remove WSUT, MWUT, and PBUT in a one-step dialysis. Besides that, MMM is also able to improve

the biocompatibility of a polymer. Apart from that, there can also be a combination of main polymer and inorganic material, such as multi-walled carbon nanotubes (Nidzhom *et al*., 2016); activated carbon (Tijink *et al*., 2012; Pavlenko *et al*., 2016); nano-hydroxyapatite (Sun and Wu, 2014a; Sun and Wu, 2014b); and silicalite or zeolite (Tantekin-Ersolmaz et al., 2000). However, there are requirements that need to be fulfilled for material used as additive, such as containing hydrophilic group, and is biocompatible and non-toxic. When a main polymer, such as polyethersulfone (PES), is in direct contact with blood during HD process, the proteins tend to be adsorbed onto the polymer surface. Then, this protein layer causes fouling on the inner surface of the HD membrane and decreases the function of pores in the inner membrane surface (Irfan and Idris, 2015). This membrane is a problem-solving for many cases and weaknesses occured during blood purification treatment. Therefore, there is a need to further study the MMM to provide the best solution in blood purification.

The porous structure of zeolites makes them true shape-selectivity molecular sieves with wide-ranging applications in catalysis, ion exchange, and adsorption processes (Cejka *et al*., 2010; Zaarour *et al*., 2014). Other than that, zeolites can be modified through its selectivity pore size by molecularly imprinting polymer (MIP) concept. Khasanah *et al*. (2013) has managed to produce the imprinted zeolite for the improvement of the selectivity of a voltammetric sensor in uric acid analysis. This imprinting zeolite-modified glass carbon has showed a good performance and high sensitivity, precision, accuracy, and low detection limit. Zeolite be synthesized with a three-dimensionally ordered mesoporous-imprinted structure using a carbon template to improve the catalytic and separation performance (Chen *et al*., 2011). Furthermore, combination of zeolite with PES as HD membrane is prabably solution for efficient removal of WSUT, MWUT, and PBUT during treatment in order to provide a better quality of life for a patient with kidney failure.

1.2 Problem Statement

On the discussion of HD, reported that adequacy of dialysis be achieved by using high-flux compared to the low-flux dialysis (Oshvandi *et al*., 2014). However, the high-flux dialysis has several weaknesses, such as higher cost compared to that of a low-flux dialysis, and there are many requirements that have to be fulfilled by patients prior to receiving the treatment, such as body weight, hemoglobin level, phosphate concentration, and blood pressure (Yilmaz *et al*., 2014). It causes the blood and dialysate flow under a high-pressure pass to the HD membrane, thus resulting in a dangerous drop in blood pressure, causing fewer symptoms to the patients (Oshvandi *et al*., 2014). Furthermore, not every patient can be treated by a high-flux dialysis. Based on these problems, there needs to be an enhanced alternative membrane on low-flux dialysis that is capable of removing the WSUT, MWUT, and PBUT simultaneously.

Based on the selectivity problem for the sorbents used for PBUT removal applied into HP treatment as described previously, this study aims to develop zeolite Y to be more selective on *p*-cresol uremic toxin by using the MIP concept prior to fabricating it to the HF-MMM with polyethersulfone (PES) and polyvinylpyrrolidone (PVP) by using dry/wet inversion spinning system (Luo *et al*., 2015; Trotta *et al*., 2012). This study aims to develop a combination of HD and HP in one-step dialysis by combining PES/PVP with modified zeolite Y to produce excellent membrane to remove *p*-cresol as PBUT. The adsorbent used is an innovation of zeolite Y through the MIP concept to produce a more selective zeolite Y. This is caused by the pores size of zeolite that have been imprinted and fitted onto the *p*-cresol. The zeolite Y was chosen because of its suitable pore size (0.6 to 0.8 nm) for *p*-cresol, which had a size of 0.66 nm in large; 0.76 nm in length; and 0.39 nm in thickness (Wernert *et al*., 2005). The zeolite Y was imprinted by using *p*-cresol as a representative of PBUT called the imprinted zeolite Y/*p*-cresol (IZC). The *p*-cresol was chosen as a target uremic toxin because of its higher toxicity compared to other types of PBUT. Additionally, the modification of zeolite through IZC could significantly improve the *p*-cresol removal as a powder form for it to be embedded into the HF-MMM. So far there have been no reported regarding MMM containing highly selective adsorbents for special toxins.

The MMM that has been developed contains less selective. Thus, it were adsorb many potential compound that are still needed and less able to adsorb toxins especially PBUT. The HF-MMM developed could offer the adequacy removal of WSUT and PBUT during the performance. Since they have a dual function in the one-step treatment, the WSUT could be excellently removed by the diffusion process, while the PBUT was removed by the adsorption mechanism in the one-time step. Additionally, the comprehensive study of the low-flux membrane fabrication, including the effect of air gap distance between the spinneret and coagulant bath, percentage loading of main polymer (PES), additive (PVP), and adsorbent filler (IZC) could eventually produced the low flux superior membrane to remove urea and *p*-cresol uremic toxins.

1.3 Research Objectives

The work aims to synthesize the high-capability of IZC adsorbent for *p*-cresol removal then fabricated to PES to produce an excellent hybrid MMM. The specific objectives of this study are:

1. To investigate the adsorption properties of IZC towards *p*-cresol by varying adsorption time and *p*-cresol concentration and selectivity to phenol.

2. To examine the effects of air gap and loadings of PES, PVP, and IZC on the characteristics and separation performance of PES/PVP/IZC HF-MMM.

3. To investigate the hemocompatibility and cytotoxicity properties of PES/PVP/IZC HF-MMM compare to that of neat PES membrane.

1.4 Scope of the Study

To realize the above-mentioned objectives, the subsequent scopes of studies have been finalized as follows:

(1) Synthesizing zeolite Y (ZeoY-S) and IZC by using $NaAlO_2$, NaOH, and SiO_2 under aging at room temperature for 24h and hydrothermal crystallization treatment at 100°C for 24h by molar ratio $4Na_2O:10SiO_2:Al_2O_3:180H_2O$ then adding *p*-cresol for IZC (*to accomplish objective 1*).

(2) Characterizing synthesized zeolite Y (ZeoY-S), non-imprinted zeolite (NIZ), IZC and commercialized zeolite Y-CBV100 (ZeoY-C) by using X-ray Diffraction (XRD), Fourier Transform Infrared (FTIR), and Thermal gravimetry analysis (TGA) (*to accomplish objective 1*).

(3) Characterizing the morphology, pore structure and size of ZeoY-S, IZC and ZeoY-C by using scanning electron microscopy (SEM), transmission electron microscopy (TEM), and Brunauer-Emmet-Teller (BET) (*to accomplish objective 1*).

(4) Studying of adsorption time on ZeoY-C, ZeoY-S, and IZC for 5 to 240 min using synthetic *p-cresol* solution, by ranging the initial concentration of *p*-cresol from 10 to 50 ppm at fixed adsorbent weight of 50 mg (*to accomplish objective 1*).

(5) Analysing the adsorption studies by Nuclear magnetic resonance for ^{1}H and ^{29}Si (NMR) by comparison of IZC and NIZ (*to accomplish objective 1*).

(6) Analysing the adsorption isotherm of Langmuir and Freundlich parameter and adsorption kinetics for Pseudo first-order, second-order and intra-particle diffusion (IPD) models for ZeoY-C, ZeoY-S and IZC (*to accomplish objective 1*).

(7) Measuring the selectivity of the ZeoY-C, ZeoY-S, and IZC by using phenol as influence analyte (*to accomplish objective 1*).

(8) Preparing several spinning dope solutions for the fabrication of HF-MMM at different PES, PVP, and IZC loading (*to accomplish objective 2*).

(9) Fabricating of HF-MMM using dry/wet phase inversion technique between PES/PVP and IZC with a study of air gap distance between spinneret and coagulation bath during spinning process varying from 10 to 50 cm; PES loading ranging from 14, 16, and 18 wt.%; PVP loading ranging from 1.6; 2; and 3 wt.%, and different loading of IZC ranging from 1; 2.5; 5; and 7.5 wt.% (*to accomplish objective 2*).

(10) Characterizing the physico-chemical composition of PES/PVP/IZC HF-MMM by using SEM, Energy-dispersive X-ray spectroscopy (EDX), Atomic force microscopy (AFM), FTIR, and TGA (*to accomplish objective 2*).

(11) Determining water flux, BSA retention, WCA, porosity, and pore size in order to consideration the optimum parameter (*to accomplish objective 2*).

(12) Determining membrane separation properties using urea and *p*-cresol removal by using PBS and BSA solution as well as by a single fiber permeation system and modulation system (*to accomplish objective 2*).

(13) Determining concentration of IZC leached out during permeation by using Inductively Coupled Plasma Mass Spectrometry (ICP-MS) (*to accomplish objective 2*).

(14) Performing hemocompatibility test for PES/PVP/IZC HF-MMM using protein adsorption, platelet adhesion, hemolysis assay, and blood coagulation test by Activated Partial Thromboplastin Time (APTT) and Prothrombin Time (PT) (*to accomplish objective 3*).

(15) Conducting toxicity test for PES/PVP/IZC HF MMM using MTT assay by using *Lymphocytes* cell (*to accomplish objective 3*).

1.5 Significance of the Study

An excellent membrane had been successfully developed, where it was applied into the HD treatment by increasing its selectivity. The imprinted zeolite as a selective adsorbent onto the *p*-cresol as a representative of PBUT was synthesized. The results proved that the imprinting process was successfully printed on the zeolite framework. The HF-MMM composite of PES, PVP, and IZC was fabricated by dry/wet phase inversion technique had successfully removed the WSUT and PBUT in a one-time process. The developed membrane has a high novelty, as there had been no report of HF membrane combined with a selective adsorbent such as IZC. This success became a basis for the development of selective adsorbents for other uremic toxins to provide a better quality of life for kidney disease patients. This hybrid membrane will be the most promising alternative for more adequate uremic toxin removal for life-sustaining of CKD patients. Thus, the results will be able to be continuously developed by the Malaysian government to produce own Malaysia's dialysis membrane, together with the domestic industry collaboration. As known, Malaysia is still importing dialysis membranes to meet the needs of dialysis membrane for hospital and clinical use. Therefore, it will have a positive impact on the country's economy and give better live sustainability for kidney failure patients in Malaysia.

1.6 Organization of the Thesis

This thesis is divided into seven chapters. In the first chapter (Chapter 1), the introduction on the application of membrane technology to the hemodialysis treatment include a problem statement, objectives, scopes, and the significance of study are provided. Chapter 2 consists of literature reviews of the topics in line with the interest of this thesis. The details of kidney, kinds of uremic toxins, hemodialysis mechanism, including the membranes used, were described. This chapter also deliberates the zeolite, imprinted zeolite, and its biocompatibility. Meanwhile, the materials and methods used in this study were described in Chapter 3. This chapter also explains for the characterization approaches and performance testing for the materials and membrane developed. Chapter 4 describes the synthesis of ZeoY-S and IZC by using

molecularly imprinting polymer concept. Then, those zeolites were characterized by using XRD, FTIR, TGA, SEM, TEM, NMR, and BET, as well as ZeoY-C as comparison. Furthermore, the adsorption and selectivity study showed that IZC was proven to improve 20% for adsorption capability and 4.3 times the selectivity, which is higher than that for ZeoY-C and ZeoY-S. The adsorption behavior under isotherm and kinetics models was also studied and discussed in depth. On the other hand, Chapter 5 describes the fabrication of IZC incorporated with PES/PVP to produce HF-MMM by using dry/wet phase inversion technique. The characterization of PES/PVP/IZC HF-MMM by using SEM, EDX, AFM, and TGA were also described. In this section, the spinning parameter, such as the air gap distance between the spinneret and the coagulation bath and varied loading of PES, PVP, and IZC were studied. Then, the performance result was obtained via water flux, BSA retention, WCA, porosity, size, and the membrane separation performance for urea and *p*-cresol removal. Those performances were measure under a cross-flow single fiber permeation and modulation system developed. Finally, Chapter 6 describes the biocompatibility test, including the hemocompatibility evaluation (protein adsorption, platelet adhesion, hemolysis assay, APTT, and PT), and cytocompatibility evaluation were tested by using MTT assay for fabricated HF-MMM. This evaluation is very crucial for the application of the materials applied into a human. Finally, the general conclusion achieved in this study, together with suggestions and recommendations for further study, are described in Chapter 7.

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APPENDIX A

Analysis Data for Crystallinity Index (%CI)

Crystallinity index for ZeoY-C

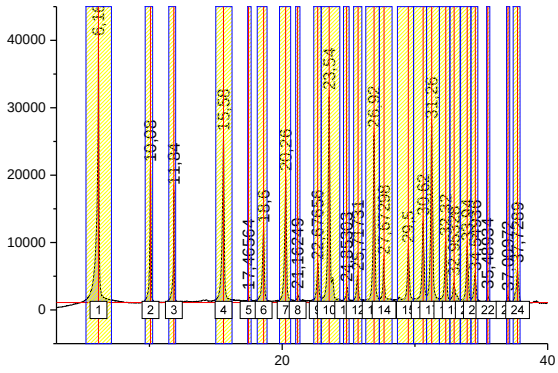

Figure A.1.1 Peak analyzer preview for total crystalline peaks for ZeoY-C

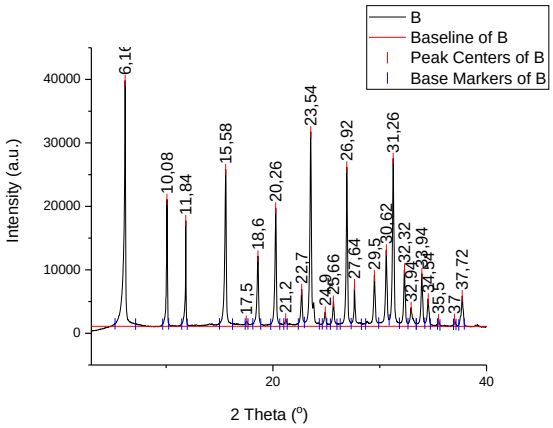

Figure A.1.2 Peak of total crystalline peaks for ZeoY-C

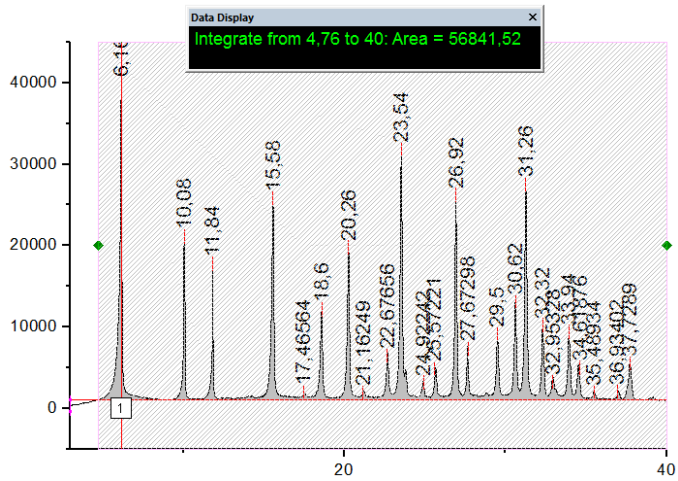

Figure A.1.3 Peak of total all peaks (crystalline and amorphous) for ZeoY-C

$$\%\,CI = \frac{Area\ of\ crystalline\ peaks}{Area\ of\ all\ peaks\ (crystalline + amorphous)}\ x\ 100$$

$$\%\,CI = \frac{56398.11}{56841.52}\ x\ 100$$

$$\%\,CI = 99.22\ \%$$

Crystallinity index for ZeoY-S

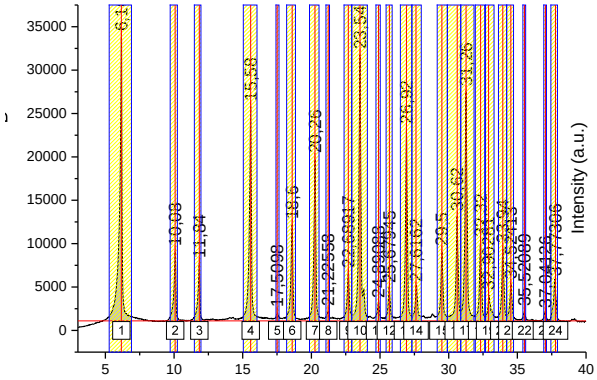

Figure A.2.1 Peak analyzer preview for total crystalline peaks for ZeoY-S

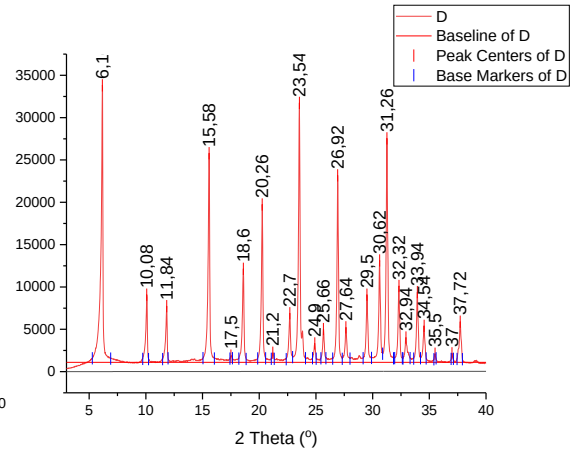

Figure A.2.2 Peak of total crystalline peaks for ZeoY-S

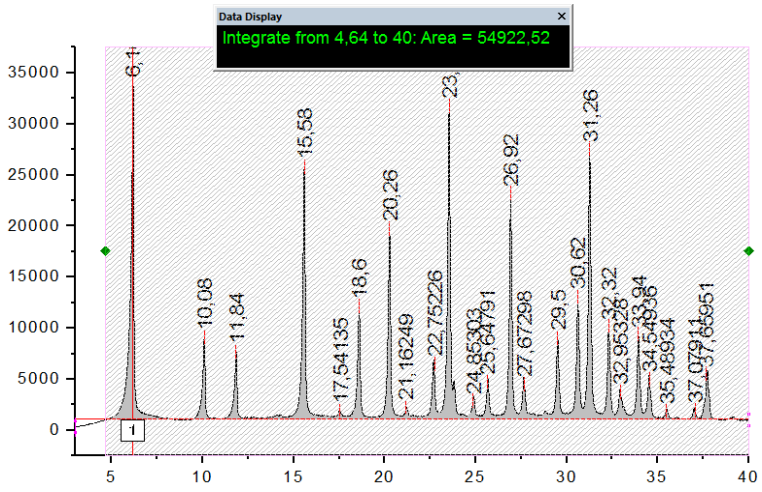

Figure A.2.3 Peak of all peaks (crystalline and amorphous) for ZeoY-S

$$\%\,CI = \frac{Area\ of\ crystalline\ peaks}{Area\ of\ all\ peaks\ (crystalline + amorphous)}\ x\ 100$$

$$\%\,CI = \frac{54115.24}{54922.52}\ x\ 100$$

$$\%\,CI = 98.53\ \%$$

Crystallinity index for IZ

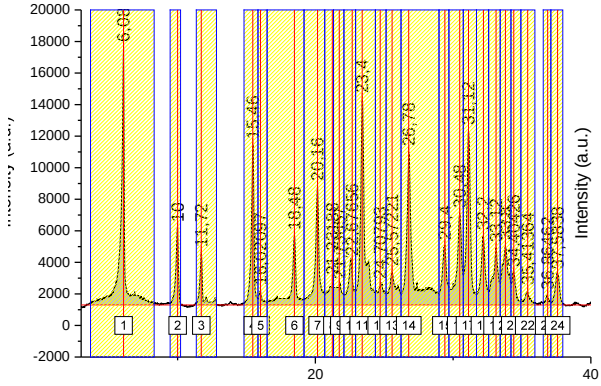

Figure A.3.1 Peak analyzer preview for total crystalline peaks for IZ

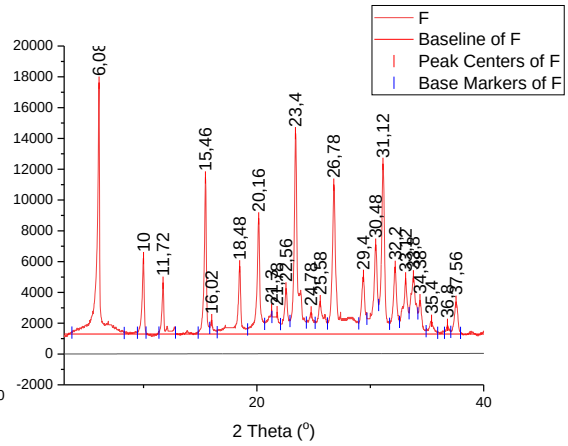

Figure A.3.2 Peak of total crystalline peaks for ZeoY-C

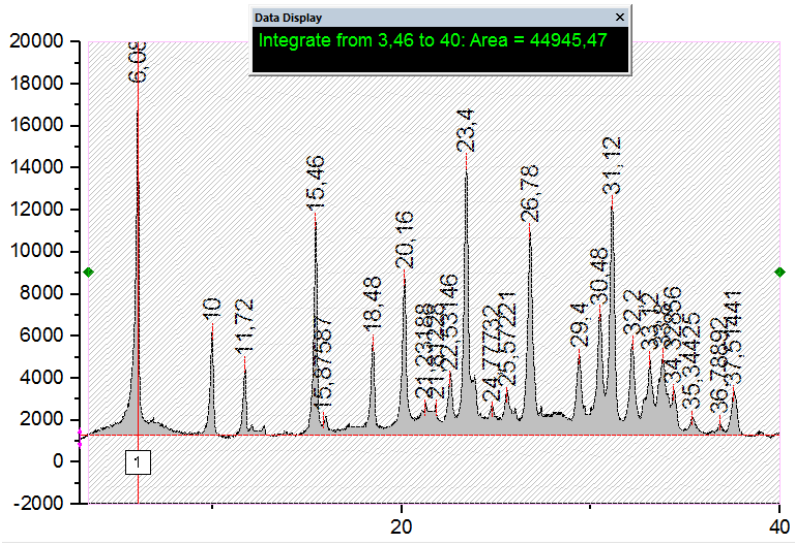

Figure A.3.3 Peak of total all peaks (crystalline and amorphous) for IZ

$$\%\,CI = \frac{Area\ of\ crystalline\ peaks}{Area\ of\ all\ peaks\ (crystalline + amorphous)}\ x\ 100$$

$$\%\,CI = \frac{44257.35}{44945.47}\ x\ 100$$

$$\%\,CI = 98.47\ \%$$

APPENDIX B

Determination of Unit Cell

Unit cell of ZeoY-C

Table B.1 Analysis data for determination of unit cell of ZeoY-C

Peak	2θ	θ	d	h	k	l	$h^2+k^2+l^2$	a (Å) $\sqrt{d^2\ x\ (h^2+k^2+l^2)}$
1	6.172	3.086	14.306	1	1	1	3	24.78
2	15.584	7.792	5.6815	3	3	1	19	24.76
3	23.510	11.755	3.7710	6	2	2	44	25.01
Average unit cell (Å)								**24.85**

Unit cell of ZeoY-S

Table B.2 Analysis data for determination of unit cell of ZeoY-S

Peak	2θ	θ	d	h	k	l	$h^2+k^2+l^2$	a (Å) $\sqrt{d^2\ x\ (h^2+k^2+l^2)}$
1	6.167	3.0884	14.321	1	1	1	3	24.81
2	15.574	7.787	5.6854	3	3	1	19	24.76
3	23.524	11.762	3.7788	5	3	3	43	24.78
Average unit cell (Å)								**24.78**

Unit cell of IZ

Table B.3 Analysis data for determination of unit cell of IZ

Peak	2θ	θ	d	h	k	l	$h^2+k^2+l^2$	a (Å) $\sqrt{d^2\ x\ (h^2+k^2+l^2)}$
1	6.157	3.0785	14.343	1	1	1	3	24.84
2	15.575	7.7875	5.6859	3	3	1	19	24.78
3	23.586	11.793	3.7690	5	3	3	43	24.71
Average unit cell (Å)								**24.78**

APPENDIX C

Determination of Si/Al Ratio

Determination of Si/Al ratio based on XRD pattern

The equation for determination of Si/Al ratio of zeolites studies is:

$$\frac{Si}{Al} = \frac{1.66656}{a - 24.191} - 1$$

Table C.1 Analysis data for Si/Al ratio obtained from XRD pattern

Zeolite	Average *a* (Å)	Si/Al
ZeoY-C	24.85	1.53
ZeoY-S	24.78	1.83
IZ	24.78	1.83

Determination of Si/Al ratio based on FTIR spectra

The equation for determination of Si/Al ratio of zeolites studies is:

a. $N_{Al} = [(4.425 - 4.054\ x\ 10^{-3})\ x\ \sigma][N_{Al} + N_{Si}]$

b. $\frac{Si}{Al} = \frac{192 - N_{Al}}{N_{Al}}$

Table C.2 Analysis data for Si/Al ratio obtained from FTIR spectra

Zeolite	σ	$N_{Al} + N_{Si}$	N_{Al}	Si/Al
ZeoY-C	1022	192	72.768	1.64
ZeoY-S	1014	192	72.192	1.66
IZ	1016	192	72.384	1.66

Determination of Si/Al ratio obtained from EDX analyzer

(a) Table C.3 Analysis data for Si/Al ratio obtained from EDX analyzer

Zeolite	Silicon (wt.%)	Aluminium (wt.%)	Si/Al
ZeoY-S	73.16	26.84	2.73
IZ	73.94	26.06	2.84

Determination of SiO_2/Al_2O_3 ratio obtained from FTIR spectra

The equation for determination of Si/Al ratio of zeolites studies is:

a. $N_{Al} = [(4.425 - 4.054\ x\ 10^{-3})\ x\ \sigma][N_{Al} + N_{Si}]$

b. $\frac{SiO_2}{Al_2O_3} = \frac{192 - N_{Al}}{N_{Al}/_2}$

Table C.4 Analysis data for SiO_2/Al_2O_3 ratio obtained from FTIR spectra

Zeolite	σ	$N_{Al} + N_{Si}$	N_{Al}	SiO_2/Al_2O_3
ZeoY-C	1022	192	72.768	3.30
ZeoY-S	1014	192	72.192	3.32
IZ	1016	192	72.384	3.31

APPENDIX D

Adsorption Test for Zeolites

Standard calibration curve for *p*-cresol by using UV-Vis Spectrophotometer

Table D.1: Standard calibration curve for *p*-cresol by UV-Vis spectrophotometer

Concentration (mg/L)	Absorbance
10	0.37
20	0.872
30	1.237
40	1.761
50	2.122

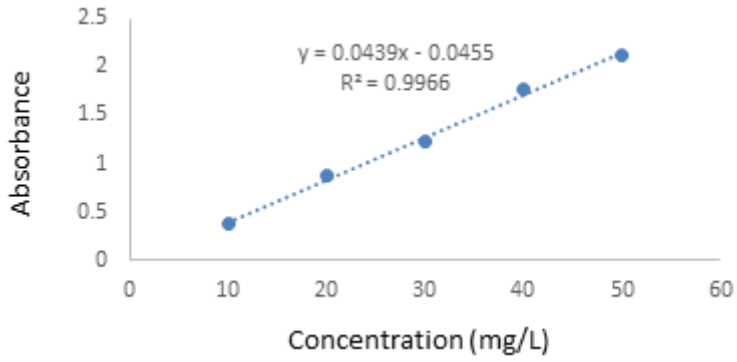

Figure D.1 Calibration curve for *p*-cresol

Adsorption of *p*-cresol at varying time

Table D.2 Analysis data for zeolites powder for *p*-cresol removal at varying time of adsorption

Time (min)	Average Qads (mg/g) ZeoY-C	SD (%)	Average Qads (mg/g) ZeoY-S	SD (%)	Average Qads (mg/g) IZ	SD (%)
5	2.99	0.08	7.69	0.11	13.90	0.15
10	4.87	1.37	9.09	0.20	16.10	0.21
15	5.61	0.05	11.04	0.20	21.10	0.06
30	6.37	0.33	14.38	0.22	21.12	0.09
45	7.33	0.31	14.52	0.02	21.15	0.06
60	9.84	0.08	14.53	0.21	21.10	0.05
90	10.05	0.15	14.64	0.11	21.09	0.15
120	10.11	0.13	14.69	0.03	21.04	0.09
150	10.03	0.18	14.72	0.07	21.19	0.07
180	10.12	0.11	14.86	0.05	21.14	0.03
210	10.03	0.13	14.85	0.03	21.07	0.18
240	10.16	0.27	14.79	0.07	21.19	0.19

Adsorption of *p*-cresol at varying concentration

Table D.3 Analysis data for zeolites powder for *p*-cresol removal at varying concentration

Concentration (mg/g)	Average Qads (mg/g) ZeoY-C	SD (%)	Average Qads (mg/g) ZeoY-S	SD (%)	Average Qads (mg/g) IZ	SD (%)
10	2.74	0.13	3.79	0.17	4.76	0.09
20	5.03	0.09	7.00	0.12	9.28	0.14
30	7.41	0.27	9.88	0.16	13.56	0.07
40	9.12	0.28	12.15	0.08	17.45	0.07
50	9.84	0.08	14.45	0.30	21.10	0.05

APPENDIX E

Selectivity Measurement

Chromatogram of phenol and *p*-cresol for HPLC

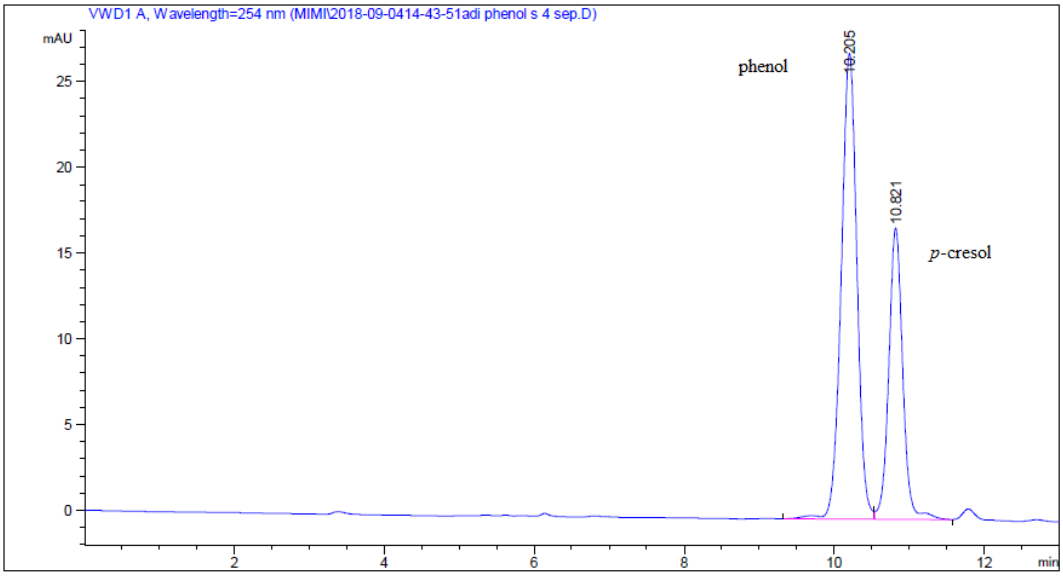

Figure E. HPLC Chromatogram of phenol and *p*-cresol

Analysis data for average area and concentration of phenol and *p*-cresol obtained after adsorption test for 50 mg adsorbent, 50 mg/L adsorbate, 1 hour adsorption time, and 210 rpm for agitation.

Table E.1: Analysis data for average area and concentration

Compound	Standard		ZeoY-S		IZ	
	Average area (mAU*s)	Conct. (mg/L)	Average area (mAU*s)	Conct. (mg/L)	Average area (mAU*s)	Conct. (mg/L)
Phenol	474.21	50	394.84	41.63	423.33	44.64
p-Cresol	266.48	50	229.09	42.98	187.69	35.22

Analysis data for Qads and percentage of removal

Table E.2: Analysis data for Qads and percentage removal

Compound	ZeoY-S		IZ	
	Qads (mg/g)	Percentage removal (%)	Qads (mg/g)	Percentage removal (%)
Phenol	8.37	16.74	5.36	10.73
p-Cresol	7.02	14.03	14.78	29.57

APPENDIX F

Zeolite leached out during permeation (1% IZ loading)

Concentration of zeolite leached out during permeation for varying conditions

Condition	Pressure (bar)	Time (hour)	ppm (mg/L)				
			Rep 1.	Rep 2.	Rep 3.	Average	SD
Unwashed	1	1	0.1057	0.1027	0.0824	0.0969	0.0127
Washed	0.5	1	0.0433	0.0515	0.0571	0.0506	0.0069
Washed	0.3	1	0.0195	0.0235	0.0184	0.0204	0.0027

Percentage leaching of zeolite during permeation for varying conditions

Condition	Pressure (bar)	Time (hour)	Percentage Leaching (%)				
			Rep 1.	Rep 2.	Rep 3.	Average	SD
Unwashed	1	1	0.2114	0.2054	0.1648	0.1939	0.0254
Washed	0.5	1	0.0867	0.1029	0.1143	0.1013	0.0139
Washed	0.3	1	0.0390	0.0469	0.0367	0.0409	0.0054

APPENDIX G

Protein Adsorption Test

BSA and Fibrinogen standard calibration curve

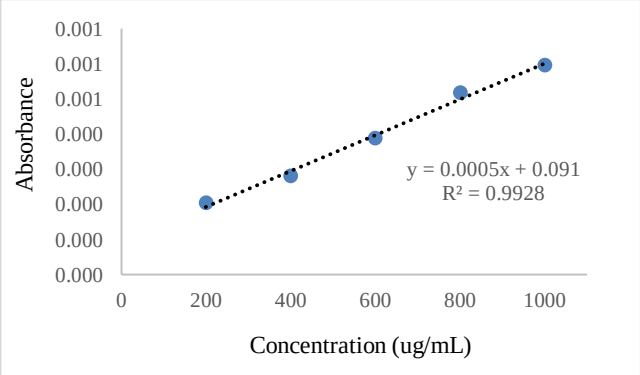

Figure G.1 Calibration curve of BSA

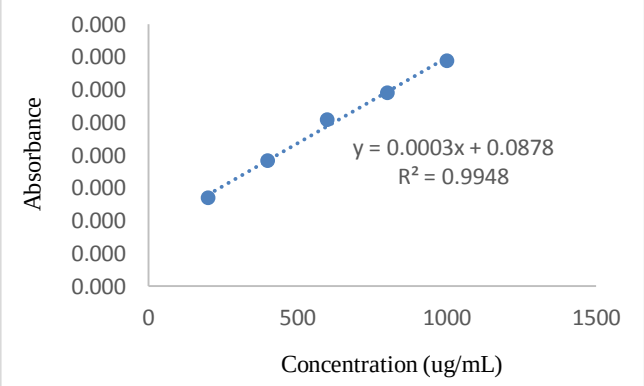

Figure G.2 Calibration curve of Fibrinogen

LIST OF PUBLICATIONS

Journal with Impact Factor

1. **Yanuardi Raharjo**, Ahmad Fauzi Ismail*, Mohd Hafiz Dzarfan Othman, Nik Ahmad Nizam Nik Malek, Djoko Santoso, Preparation and characterization of imprinted zeolite-Y for *p*-cresol removal in haemodialysis, Materials Science & Engineering C, 103(2019)109722, DOI: https//doi.org/10.1016/ j.msec.2019.05.007 (**Q1, IF: 4.959**).

2. **Yanuardi Raharjo**, Muhammad Nidzhom Zainol Abidin, Ahmad Fauzi Ismail*, Mohd Hafiz Dzarfan Othman, Pei Sean Goh, Djoko Santoso, Selection of dialysis membranes for acute kidney injury patients: A Review, Journal of King Saud University-Science (**Q2, IF: 2.835**) (*In Review*).

3. **Yanuardi Raharjo**, Ahmad Fauzi Ismail*, Mohd Hafiz Dzarfan Othman, Djoko Santoso, New imprinted zeolite-mixed matrix membranes for *p*-cresol removal in hemodialysis application (Journal of Separation and Purification) (**Q1, IF: 5.107**) (*Under progress on Writing*).

Indexed Journal

1. **Yanuardi Raharjo**, Mochamad Zakki Fahmi, Siti Wafiroh, Alfa Akustia Widati, Eviomitta Rizki Amanda, Ahmad Fauzi Ismail*, Mohd Hafiz Dzarfan Othman, Djoko Santoso, Incorporation of Imprinted-zeolite to polyethersulfone/cellulose acetate memrbane for creatinine removal in hemodialysis treatment, Jurnal Teknologi, 81:3(2019)137-144, DOI: https//doi.org/10.11113/jt.v81.13075 (SJR 2018 = 0.18, Q2 **Indexed by Scopus**).

2. **Yanuardi Raharjo**, Ahmad Fauzi Ismail*, Mohd Hafiz Dzarfan Othman, Sarina Mat Rosid, Mohd Ariff Azali, Djoko Santoso, Effect of polymer loading on membrane properties and uremic toxins removal for haemodialysis application (Journal of Membrane Science and Research (SI:ISET2019)) (SJR 2018 = 0.34, Q2 **Indexed by Scopus**) (In Press).

Book Chapter:

Ahmad Fauzi Ismail*, Muhammad Nidzhom Zainol Abidin, Sumarni Mansur, Muhamad Zulhilmi Zailani, Noresah Said, **Yanuardi Raharjo**, Sarina Mat Rosid, Mohd Hafiz Dzarfan Othman, Pei Sean Goh, Hasrinah Hasbullah, Hemodialysis Membrane for Blood Purification Process, In Membrane Separation Principles and Applications from Material Selection to Mechanism and Industrial Uses, (Eds: Ahmad Fauzi Ismail, Mukhlis A. Rahman, Mohd Hafiz Dzarfan Othman,Takeshi Matsuura, Colin F. Poole), 2019, Elsevier Publishing, Oxford, UK, ISBN: 978-0-12-812815-2.