

## Theoretical development of biofilm in hybrid growth sequencing batch reactor (HG-SBR) for degradation of phenol

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### ABSTRACT

A kinetic modeling based study was carried out to evaluate the development of biofilm in hybrid growth sequencing batch reactor (HG-SBR) for the treatment of phenol synthetic wastewater. The microorganisms from activated sludge were allowed to grow onto the surface of packing materials to improve the treatment performances. The overall evaluation of HG-SBR with different initial COD loading recorded above 90%, 66%, 59% for each reactor with GAC, bio-ring and rod-shaped AC, respectively. Rapid degradation (98% removal) recorded for GAC through the profile study with 200 mg/L initial concentration. The development of biofilm on GAC, bio-ring and rod-shaped AC were studied through the kinetic models. As a result of the calculations, First-order model, Stover-Kincannon model and Grau second-order model were applicable (correlation coefficient  $R^2 > 0.9$ ) to describe the development of biofilm. The biofilm develop on the packed fixed bed materials was homogeneous and the thickness was varied for each materials. High regression coefficient for the comparison between the experimental COD and predicted COD values were obtained. This result indicated the kinetic models applied are capable of describing role of biofilm in HG-SBR by simplified the laborious experimental data to convenient mathematical expression.

*Keywords:* Biofilm; Kinetic models; GAC; Bio-ring; Rod-shaped AC; Hybrid growth sequencing batch reactor

### 1. Introduction

Phenol substances are toxic and difficult for biologically degraded. Industrial effluent containing phenols have a carcinogenic, mutagenic and teratogeny effect [1]. Industrial effluents containing phenols were originated from various main industries such as petrochemical industries, petroleum refineries, resin manufacturing industries, dye synthesizes units, pulp and paper mills and pharmaceuticals industries. Phenol have been listed as 129 priority pollutants by United States Environmental Protection Agency (USEPA) and the untreated industrial effluents containing phenols have threatened both humans and animals [2]. The

toxicity level for human within the range of 10–24 mg/L and for animals such as fish is 9–25 mg/L. The lethal blood concentrations were 150-mg/100 mL [3].

Various biological methods have been attempted for removal of phenols such as biological and enzymatic treatments, isolation of phenol-degrading bacteria, packed bed reactor and immersed membrane bioreactor [4–7]. The utilization of packing materials in biological treatment system has gained a lot of attention recently. The indigenous bacteria from activated sludge were allowed to grow onto the surface of packing material by retained in the same container for certain period of time. The formations of biofilm in activated sludge improve the treatment performances by reducing the growth of filamentous bacteria [8]. The suspended bacteria formed biofilm when being contacted with the packing material.

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The hybrid growth SBR is a combination of the two types of microorganism's growth types which suspended and attached growth. The hybrid growth allows the existence of both bio-sludge and biofilm within a single reactor. This will serve as advantage for the removal of the toxic organic pollutant such as phenol due to the high biomass concentration. The biofilm formed by the attached growth will improve activated sludge settling characteristic. While, the suspended growth activated sludge treatment has the capability to produce high quality effluent. Apart from that, the hybrid growth SBR was selected for the removal of toxic substance such as phenol as it able to provide both stability and resistance to high phenol concentrations.

The overall developments consist of five main stages which is initial attachment, irreversible attachment, growth, maturation and dispersion [8–10]. The solid surface of the growth material was modified by adsorption of various solutes and the properties have slightly altered. The suspended bacteria approached the solid surface through motility and overcome the van der Waals forces between the cell and the surface [8]. This will create a reversible attachment of bacteria on the solid surface. The second stages continued by the production of extracellular polymeric substances (EPS) by the bacteria [8]. The condition considered as the transition from reversible to irreversible attachment. In irreversible attachment, the initial colonies of bacteria act as a matrix for adhesion the suspended flocs. After irreversible attachment, the biofilm begins to grow. Then, the micro colonies develop into a matrix of biofilm [8]. During the stage, EPS traps nutrients from the surrounding environment in order to continue to be produced. Finally, dispersion or de attachment stages occurs when biofilm reaches to stable and causes the biofilm formation on other surfaces and new colonies before space and nutrient become limited. Bacteria then return to planktonic state [11].

Several studies report a higher removal rates for the bioreactor with biofilm. For instances, Pishgar et al. [12] compared between the free and immobilized bacteria cell behavior for phenol biodegradation in anaerobic reactor. The immobilized cells were able to remove phenol at concentration of 100–700 mg/L in shorter time period. Dey

and Mukherjee, [13] utilized glass beads as the surface for growth of biofilm and the removal rate of 89% was achieved. A packed bed reactor has higher resistance to high phenol concentrations and is able to lead to higher elimination rates of phenol [14].

The biofilm formation on different materials depends on the condition of the solid surface. High surface area to volume ratio, surface condition and hydrophobic surfaces contributed to high biofilm formation [8]. Higher surface volume provided more attached surface for the bacteria. The formation of the biofilm is different on the smooth surfaces. The hydrophobic surfaces would enable the cell to overcome the repulsive forces that are active within the certain distances from the surface bed. This would assist the interaction occurs between the cell surface and the bed surface [8].

According to available literature, there is no report on the development of the biofilm theoretical in hybrid growth type of sequencing batch reactor for treatment of phenol. This study is crucial in order to understand the formation of biofilm for improvement of biodegradation in conventional SBR system. Besides, kinetic models are useful for optimization of particular treatment system by simplified mathematical expressions [15]. Thus, the main objective of this study was to investigate the biodegradation performances of HG-SBR by different types of packed bed material. In hybrid growth reactor, three types of packed bed were applied which is granular activated carbon, bio-ring and large rod-shaped activated carbon together with activated sludge. The theoretical development of biofilm on different packed bed material was assessed through kinetic models

## 2. Materials and methods

### 2.1. Experimental set-up

The laboratory scale SBR is shown in Fig. 1 three acrylic tanks (30 cm × 20 cm × 20 cm) with a total working volume of 7 L were used as a bioreactor. The SBR reactor was inoculated with 3 L activated sludge obtained from rubber waste industrial wastewater treatment plant Shorubber (Malaysia)

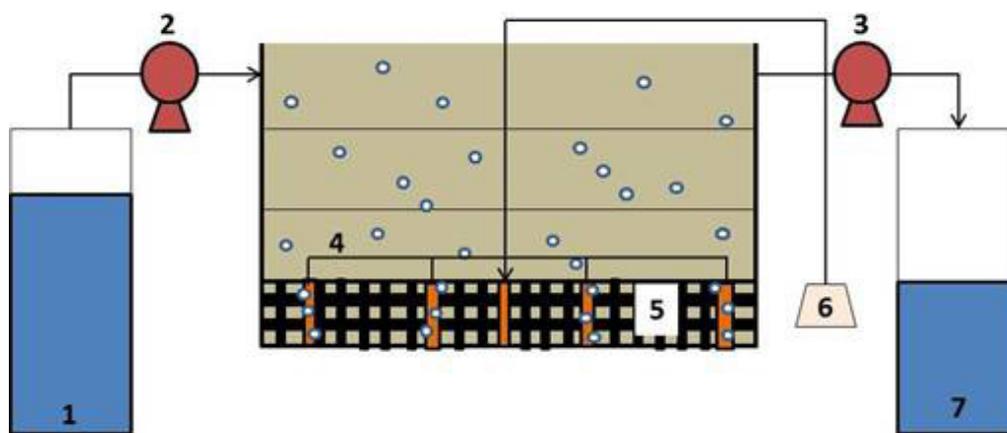


Fig. 1. Schematic diagram of HG-SBR (1) Influent tank, (2), (3) Dosing pump, (4) Suspended sludge and synthetic wastewater: 7 L, (5) Packed fixed bed materials (6) Aeration pump (7) Effluent tank.

Sdn. Bhd. The characteristics of activated sludge are given in Table 1. Three types of packed fixed bed material which were granular activated carbon (GAC), bio-ring and rod-shaped activated carbon were added to each bioreactor. Two types of biomass co-existed in these bioreactors which were suspended microbial biomass and microbial biomass in form of biofilm formed on the fixed bed. This condition created a hybrid growth sequencing batch reactor (HG-SBR). The operation modes for FILL: REACT: SETTLE: DRAW was in the time ratio of 1:20:2:2 for each reactor. Submerged air diffuser was applied to provide aeration during FILL and REACT modes with the aeration flow rate at 60 mL/min. The concentration of dissolved oxygen during FILL modes was within the range of 5.6–6.6 mg/L. However, the dissolved oxygen concentration during REACT was higher (6.6–7.6 mg/L). A set of two peristaltic pumps (Pro Minent: CONC 0308PP100A002) was used to feed the influent from the influent tank to both reactors and for the effluent discharged. The reactors were operated at room temperature.

## 2.2. Synthetic wastewater

The compositions of the synthetic wastewater utilized are given in Table 2. The mixture of synthetic wastewater contributed approximately 600–3000 ± 50 mg/L of chemical oxygen demand (COD) concentration. Sucrose (C<sub>12</sub>H<sub>22</sub>O<sub>11</sub>) and phenol (C<sub>6</sub>H<sub>6</sub>O) were the main substrate for the bacteria.

## 2.3. Biofilm attachment

Three types of packed fixed bed material granular activated carbon (GAC) (DARCO®), bio-ring (AQUADINE®)

Table 1  
Characteristics of inoculum activated sludge

Parameters	Value
Sludge volume index (SVI), mg/L	5.596
Mixed liquor suspended solids (MLSS), mg/L	24,780
Mixed liquor volatile suspended solids (MLVSS), mg/L	10,715
Chemical oxygen demand (COD), mg/L	7920
pH	6.8–7.2
Dissolved oxygen (DO), mg/L	6.2

Table 2  
Composition of synthetic wastewater

Compound	Concentration (mg/L)
C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>	563
C <sub>6</sub> H <sub>6</sub> O	100–1000
NH <sub>4</sub> Cl	172
MgSO <sub>4</sub>	49
FeCl <sub>3</sub>	12
K <sub>2</sub> HPO <sub>4</sub>	180
KH <sub>2</sub> PO <sub>4</sub>	35
NaHCO <sub>3</sub>	100

and rod-shaped activated carbon (AQUADINE®) used in this study were submerged into the activated sludge. The total of 1.5 kg of each packing materials was added to the reactors. The materials were inoculated under aerobic condition. The reactors were continuously feed with carbon source, nutrients and buffer solution.

## 2.4. Analytical procedures

Analyses of chemical oxygen demand (COD), mixed liquor suspended solid (MLSS) were carried out based on the standard methods [16]. Phenol concentration was determined by UV-V spectrophotometer (Hitachi U-2800, Japan) from 200 nm to 800 nm. The maximum absorbance wavelength ( $\lambda_{max}$ ) of phenol was 270 nm. The thickness of biofilm was determined with the assumption of complete coverage [17]. The bed materials with biofilm were dried and weighted before and after drying. Biofilm comprises of 99% of water. The thickness of biofilm  $L_f$  was calculated as follows [8]:

$$L_f = \frac{W}{\rho S_b (0.99)} \quad (1)$$

where  $W$  represents the weight of evaporated water (g),  $\rho$  are the density of water (g/cm<sup>3</sup>), and  $S_b$  are the surface of bed (cm<sup>2</sup>).

## 3. Results and discussion

### 3.1. Bioreactor performance

In order to assess the overall performance of HG-SBR reactor, the COD removal efficiency under different substrate concentration was measured. COD were used as a major parameter to compare the performances of all reactors with different packed fixed bed materials. The results obtained from COD monitoring are illustrated in Table 3. The variation of initial COD concentration ranged between 608 mg/L to 3012 mg/L. In order to prevent shock loading, the bioreactor was allowed to recover by lowering the concentration to 857 mg/L. Based on the results obtained, the COD removal efficiencies were higher than 90% for HG-SBR with GAC for all different initial COD concentration. While, for HG-SBR with rod-shaped AC, the removal efficiencies were slightly lower than GAC which was within range of 66.6–96.3%. Bio-ring recorded the lowest removal efficiencies. The lowest percentage removals recorded for bio-ring were 59.8% and the highest was 95%.

Evaluation of the experimental result indicates that each three reactor show excellent removal efficiency of COD. Debik and Coskun, [18] recorded the high average COD removal of 94.5 ± 8.5 for static granular bed reactor compared to static anaerobic sludge bed reactors which are 94.2 ± 1.7. Jafarzadeh et al. [19] reported the average COD reduction for the anaerobic hybrid reactor for petrochemical waste treatment system ranging from 42.1 to 85.9%. In the anaerobic hybrid reactor, both sludge bed and fixed bed with corrugated plastic sheet as packing material.

Fig. 2 illustrates the phenol concentration decay curve in each HG-SBR reactor with different packed fixed bed materials. The initial phenol concentration chose for this

Table 3  
Overall performance of HG-SBR

Run no.	Initial COD loading (mg/L)			Final COD loading (mg/L)			Removal efficiencies (%)		
	GAC	Quartz Ring	Rod-shaped AC	GAC	Quartz Ring	Rod-shaped AC	GAC	Quartz Ring	Rod-shaped AC
1	608	608	608	170.3	244.5	203.5	72.0	59.8	66.6
2	857	857	857	83.2	151.0	104.5	90.3	82.4	87.8
3	1108	1108	1108	46.5	58.0	41.0	95.8	94.8	96.3
4	857	857	857	103.5	120.0	96.0	87.9	86	88.8
5	1584	1584	1584	57.2	105.2	67.9	96.4	93.4	95.7
6	857	857	857	53.5	120.4	91.3	93.7	86.0	89.3
7	2060	2060	2060	103.0	135.5	86.5	95.0	93.4	95.8
8	857	857	857	47.1	62.5	56	94.5	92.6	93.4
9	2536	2536	2536	328.9	836.5	107	87.0	67.0	95.7
10	857	857	857	30.0	62.5	13	96.5	92.7	98.5
11	3012	3012	3012	97.1	149.0	131	96.8	95.0	95.7

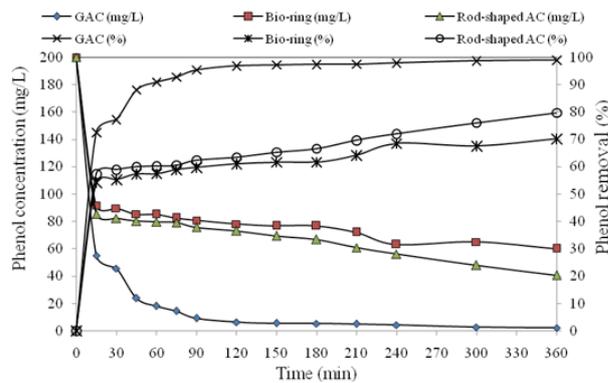


Fig. 2. Phenol removal profile in HG-SBR after 6 h reaction.

profile study was 200 mg/L. The removal of phenol was monitored for the first 6 h during the REACT phase. It was observed that the phenol removal rate in HG-SBR with GAC was higher compare to bio-ring and rod-shaped AC. Phenol rapidly decreased from 200 mg/L to 55 mg/L after 15 min REACT phase started and continuously decreased until 75 min. After 6 h the percentage removal recorded was 98%.

While, slightly different removal trends observed for bio-ring and rod-shaped AC. The degradation rates are slower for bio-ring as only 62% removal recorded after 3 h. For rod-shaped AC 66% phenol removed after 3 h. The total phenol removal for HG-SBR with bio-ring and rod-shaped AC after 6 h was 70% and 80% respectively. Fig. 3 depicts the changes in the UV-Vis spectra of 200 mg/L phenol for three HG-SBR reactor. The decreased of the peak at 269 nm after 6 h indicated the degradation of phenol molecules. The rapid degradation of phenol recorded for all reactors due to the adequate amount of biomass both attached on the materials and suspended. The bacteria in hybrid growth reactor are more rest toward the toxicity of the phenol. Lim et al. [20] reported that attached growth and suspended growth biomass in moving bed sequencing batch reactors enhanced the nitrogen removal. Phenol was then converted by bacteria to carbon dioxide. However, the biodegradation

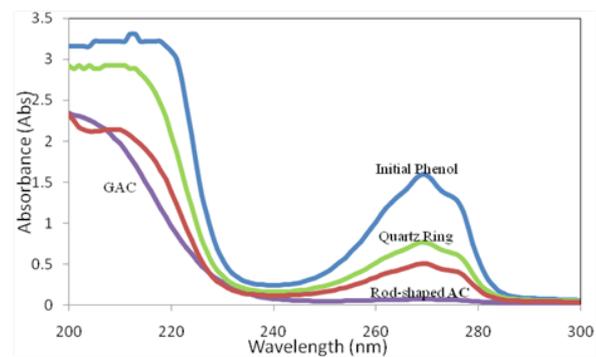


Fig. 3. Uv-vis spectra analysis of phenol at 200 mg/L ( $\lambda_{max}$ : 269 nm).

of phenol under aerobic conditions was initiated by the oxygenation of phenol hydroxylase enzyme to form catechol [21]. The degradation continues by catechol undergoes ring cleavage. The ring cleavage can occurred either at ortho or meta position which depends on types of microorganism strain [22]. This will lead to the formation of succinyl Co-A, acetyl Co-A, pyruvate and acetaldehyde. These intermediates will be utilized for metabolic activity of microorganism known as Kreb's cycle [23].

### 3.2. Development of Biofilm

The development of biofilm was crucial to enhance the performances of the bioreactor. In this study, the suspended sludge containing microbial co-existed with the microbial on the biofilm in the same reactor. The assumption can be made that the cycle of the biofilm development from initial attachment until dispersion are continuously occurred. The thickness of the biofilm attached onto the packed fixed bed materials used is shown in Table 4. Besides surface area and sizes of the materials used, the duration for the biofilm to develop effected the thickness of the biofilm [24]. Thicker biofilm indicated that larger number of bacteria accumulated on the surface. This would promote the removal of the sub-

Table 4  
Biofilm thickness and HG-SBR performances

Experimental data	GAC	Quartz ring	Rod-shaped AC
Inoculation period (day)	30	15	15
Biofilm thickness ( $\mu\text{m}$ )	21.32	3.93	4.98
COD removal (%)	98.81	86.69	95.02
Phenol removal (%)	98.88	70.07	79.70

strates in HG-SBR. The COD and phenol removal for GAC were highest with 98.81% and 98.88%, respectively. This can be contributed to the thickness of the biofilm on the GAC compared to bio-ring and rod-shaped AC. The movement of fluid over the biofilm caused a shear effects which later contribute to the detachment of biomass from the material surface [24]. This would increase the biomass concentration and sequentially increase the substrates consumption.

The formation of the biofilm onto packed fixed bed materials will lead to adsorption process. Continuous adsorption process eventually exhausts the materials. Thus, the immobilized materials requires to regenerate without changing the textural properties of the material [25] and able to function for further mineralization. For the HG-SBR system, offline bio-regeneration process occurred. This involves the removal of adsorbed phenol from contaminated activated carbon through desorption and biodegradation process [26]. Following this, phenol in bulk solution will be biologically degraded by microbial action. One of the factor affected the bio-regeneration was the reversibility of the adsorption process. The irreversible adsorption of phenol was caused by high energy bonding between the phenol molecule and packed fixed bed materials as adsorbent. Besides that, in aerobic condition, the bio-regeneration was also affected by the oxidative polymerization of phenolic compound adsorbed onto activated carbon [27]. The oxidative phenomenon occurred with the formation of the electron-donating phenol intermediates products such as cresol, dimethylphenol, and isopropylphenol [25]. This would explain the different thickness between the activated carbon based material (GAC and rod-shaped AC) with bio-ring. The larger thickness of biofilm form onto GAC and rod-shaped AC. The increase of biofilm thickness provides larger amount of biomass concentration on the surface of the materials for degradation of phenol.

Fig. 4 depicts the SEM images of raw GAC, bio-ring and rod-shaped AC and the biofilm formed on each type of packing materials. Based on the SEM images, there was a slightly difference between the surface of packing materials. The surface of GAC and rod-shaped AC are more porous compared to bio-ring. This will affect the formation of the biofilm. It was observed that the cocci-shaped microbes' cells were more dominant on the surface of GAC. The sizes of the attached microorganism were range from 0.2 to 5  $\mu\text{m}$ . There were also a few larger round shaped microbes' cells identified with the size of 10  $\mu\text{m}$ . This probably the micro-colonies that formed into mature biofilm [10]. Similar larger flocs were also identified attached on the bio-ring surface. Besides that, small granular shapes were also formed. The formation of biofilm in larger flocs was identified more on the surface of bio-ring due to the ring shaped

of the material itself. The inner side of the ring reduced the repulsive force between the biofilm and the surface as less fluid stream pass through compared to the outer side of the bio-ring. The biofilm attached to the rod-shaped was more homogeneous. Some larger size of microbes' cells was also identified on the surface of the rod-shaped AC.

### 3.3. Kinetic modeling

The comparison between prediction and the actual biodegradation of HG-SBR can be described through kinetic analysis. There are numerous types of kinetic models introduced for biological system. Overall, the kinetic models can be divided into two types of classes which are structured and unstructured models [28]. Structured models are generally complicated as it takes metabolic pathway into consideration. On the other hand, the unstructured kinetic models are mostly employed by various researchers [18,19, 28–30] due to simplicity and suitable for technical purpose. In unstructured kinetic models, microorganisms are considered as part of the component or reactant in the biological system. Three types of model are used in this study which was modified Stover-Kincannon, first order kinetic models and Grau second order.

#### 3.3.1. First order substrate removal model

The first-order kinetics was assumed to occur in the reactor. Thus, the rate of change of the substrate concentration in HG-SBR system could be expressed as [30]:

$$-\frac{dS}{dt} = \frac{QS_0}{V} - \frac{QS}{V} - k_1S \quad (2)$$

Under steady-state conditions, the rate of change in substrate concentration ( $-dS/dt$ ) negligible and the equation modified as:

$$\frac{S_0 - S}{t} = k_1S \quad (3)$$

where  $S_0$  and  $S$  are the influent and effluent substrate (mg/L);  $k_1$  is the first-order substrate removal rate constant (1/min);  $Q$  is the flow rate of wastewater (L/min) and the clean-bed volume of the filter was represent by  $V$  (L). Fig. 5 depicts the plotted graph  $(S_0 - S)/t$  against  $S$ . The value of  $k_1$  was obtained from the slope of the straight line. The value of  $k_1$  was 2.945  $\text{min}^{-1}$  for GAC, 1.1212  $\text{min}^{-1}$  for bio-ring and 1.0080  $\text{min}^{-1}$  for rod-shaped AC. Experimental data obtained a relatively good correlation which was 0.9094 and 0.9097 for both bio-ring and rod-shaped AC respectively. HG-SBR reactor with GAC as packing material has higher correlation compared to the other two ( $R^2 = 0.9782$ ). The first-order substrate removal rate constant  $k_1$  can be used to describe the growth-linked biodegradation kinetics [31]. The biodegradation was influenced by both phenol concentration and growth of biofilm. GAC recorded higher yield coefficient constant which could be attributed to higher proportion of biodegradable organic waste that synthesized into new cell.

#### 3.3.2. Stover-Kincannon model

This kinetic model initially introduced by Stover and Kincannon in the early 1970. Stover-Kincannon kinetic models are

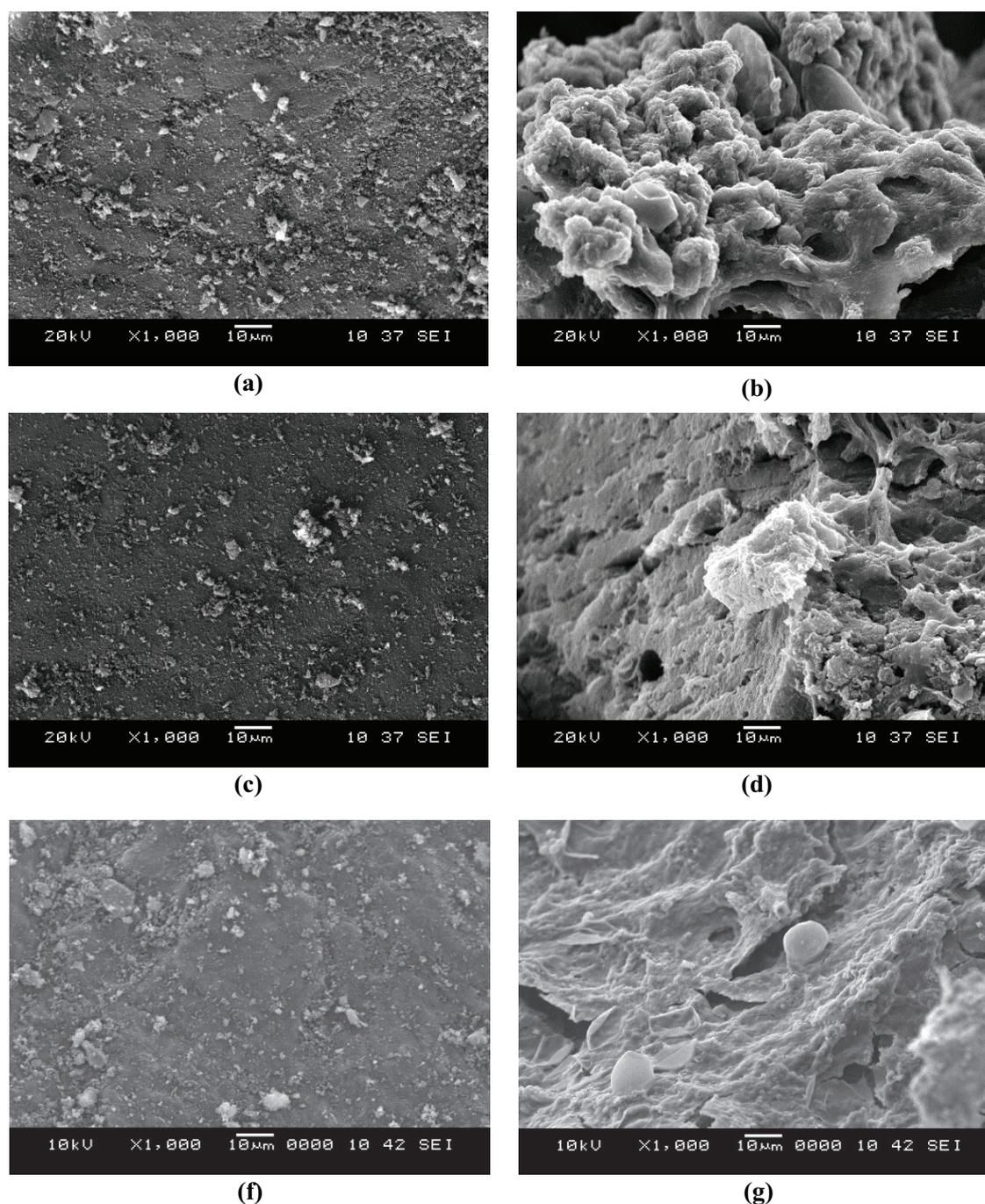


Fig. 4. SEM images of surface of packing materials (a) GAC (c) Bio-ring (f) Rod-shaped AC and attached biofilm onto packing materials (b) GAC (d) Bio-ring (g) Rod-shaped AC.

widely used mathematical models for determining the kinetic constant in biofilm reactor [30,32–35]. Stover-Kincannon kinetic model suggested that substrates utilization rate can be expressed through organic loading rate using mono-molecular kinetics for reactor with biofilm. However, it was difficult to measure the active surface area that supports the biofilm growth [18]. Thus, the original kinetic model has been modified. The effective volume of the reactor was taken into account. Modified Stover-Kincannon special feature include

the utilization of the concept of total organic loading rate as the major parameter to describe the kinetic of HG-SBR in term of organic matter removal [29]. For this study, the major parameter was the COD. The removal of COD in HG-SBR can be determined on the basis of the COD removal rate as a function of the COD concentration. This can be proved through Table 5 where the lowest COD removal rate of each fixed packed material was 250.1 mg/L-d, 207.7 mg/L-d and 231.1 mg/L-d for GAC, bio-ring and rod-shaped AC respectively. The high-

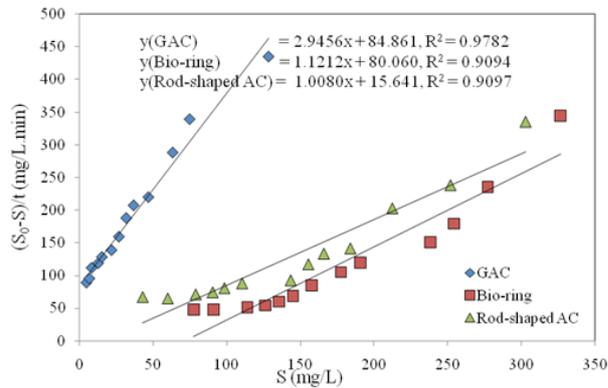


Fig. 5. First-order kinetics model.

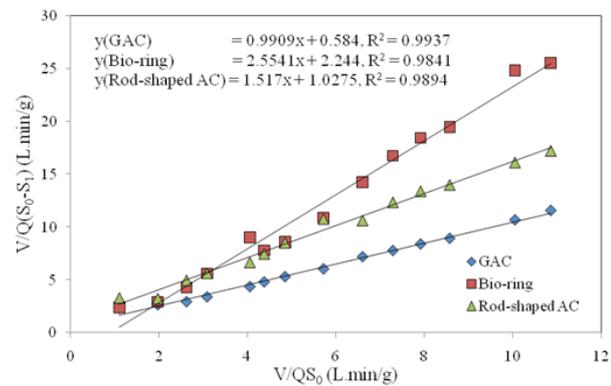


Fig. 6. Stover-Kincannon kinetic model.

Table 5  
Substrate removal rate

$S_0$ (mg/L)	$Q(S_0 - S_1)/V$ (mg/L·d)		
	GAC	Quartz Ring	Rod-shaped AC
608	250.1	207.7	231.1
857	442.2	403.4	430
1108	606.6	600.0	609.7
1584	872.5	845.0	866.3
2060	1118.3	1099.7	1127.7
2536	1261.2	1371.1	1388
3012	1665.7	1636.0	1646.3

est removal rate recorded by GAC with 1665.66 mg/L·d, and then followed by rod-shaped AC 1646 mg/L·d and Bio-ring 1636 mg/L·d. Only a small difference of removal rate recorded for each fixed bed material and the trend was similar. Based on the experimental results, the COD removal rate increased with increase in COD concentration with high correlation coefficient ( $R^2 > 0.9800$ ).

Equations of the modified Stover–Kincannon model are as follows [29]:

$$\frac{dS}{dt} = \frac{Q}{V}(S_0 - S) \quad (4)$$

$ds/dt$  defined in two ways as follows:

$$\frac{dS}{dt} = \frac{U_{\max} \left( \frac{QS_0}{V} \right)}{K_B + \left( \frac{QS}{V} \right)} \quad (5)$$

$$\frac{dS}{dt} = \frac{kXS}{K_s + S} \quad (6)$$

$$\left( \frac{dS}{dt} \right)^{-1} = \frac{V}{Q(S_0 - S)} = \frac{K_B V}{U_{\max} QS_0} + \frac{1}{U_{\max}} \quad (7)$$

where  $dS/dt$  is substrate removal rate (g/L·min);  $U_{\max}$ , maximum utilization rate constant (g/L·min);  $K_B$ , saturation value constant (g/L·min);  $k$ , maximum rate of substrate

removal (1/day);  $X$ , microorganisms concentration (VSS) in HG-SBR (g/L);  $K_s$ , half-velocity constant (g/L),  $V$ , clean bed volume of HG-SBR (L).

By plotting the inverse of the loading removal rate  $V/[Q(S_0 - S)]$  against the inverse of the total loading rate  $V/QS_0$ , a straight line graph obtained and that  $1/U_{\max}$  and  $K_B/U_{\max}$  are the intercept and slope of the straight line respectively. Fig. 6 depicted the modified Stover–Kincannon for HG-SBR with GAC, bio-ring and rod-shaped AC. The maximum removal rate constant ( $U_{\max}$ ) was 1.712 g L/min, 0.4456 g L/min, 0.9732 g L/min for GAC, bio-ring and rod-shaped AC, respectively. While, the saturation value constant ( $K_B$ ) for GAC 1.696 g L/min, bio-ring 1.138 g L/min and rod-shaped AC 1.476 g L/min. Correlation coefficient for each of the materials high which more than 0.9800. Apparently the  $U_{\max}$  value obtained for GAC was three times larger than the  $U_{\max}$  for bio-ring. This would indicate that the utilization rates of substrates by biofilm formed onto GAC was more effective compared to the other fixed packed bed material. Generally, as the  $U_{\max}$  increased the reactor efficiency were also increased. This can be suggested that the phenol removal rate were affected by organic loading rate of the influent and the availability of biofilm to carry out the biodegradation process.

### 3.3.3. Grau second order kinetic model

The general equation of a second-order kinetic model was expressed as below [18,36–37]:

$$-\frac{dS}{dt} = k_s X \left( \frac{S}{S_0} \right)^2 \quad (8)$$

Eq. (8) was integrated and linearized, the equation obtained:

$$\frac{S_0 t}{S_0 - S} = t + \frac{S_0}{k_s X} \quad (9)$$

The second terms on the right part of Eq. (9) were assumed as a constant, the equation will become:

$$\frac{S_0 t}{S_0 - S} = bt + a \quad (10)$$

The dimensionless Grau second order constant  $a$  and  $b$  were calculated from the intercept and slope of the straight

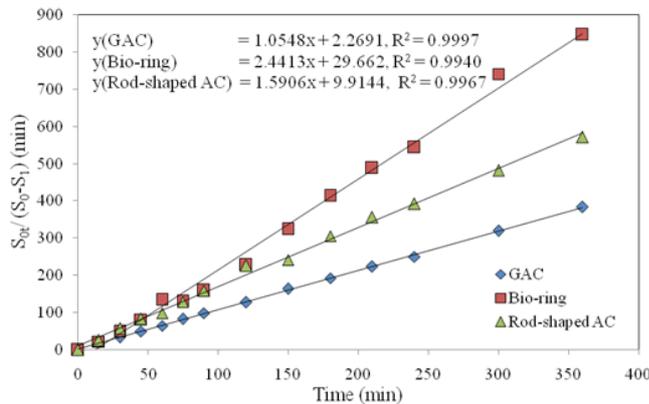


Fig. 7. Grau second-order kinetic model.

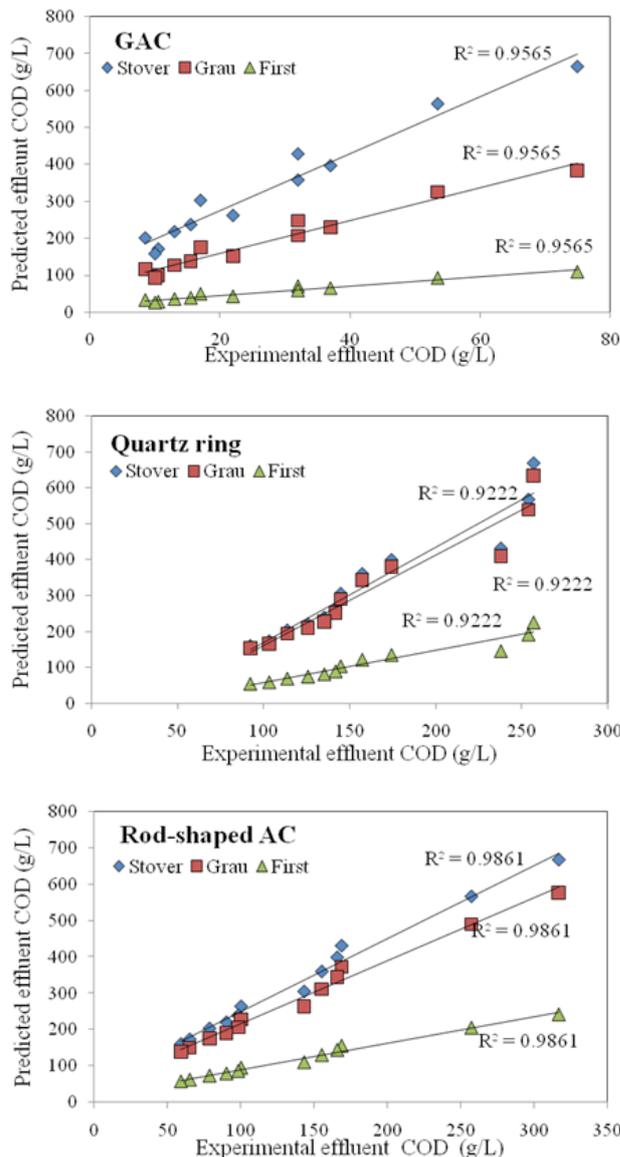


Fig. 8. Comparison of predicted and experimental COD values.

line of the graph in Fig. 7. The constant value of  $b$  for GAC, bio-ring and Rod-shaped AC are 1.0548, 2.4413 and 1.5906 respectively. While for a constant, the value obtained are 2.2691, 29.662 and 9.9144 for GAC, bio-ring and rod-shaped AC respectively. Based on the result obtained, obvious differences of Grau second order constant were observed. This might be due to variation of packed fixed bed material used.

The multicomponent Grau second-order substrate kinetic removal rate constant ( $k_s$  ( $\text{min}^{-1}$ )) was calculated from the equation  $a = S_0/(k_s X)$  [28]. The calculated removal rates constant are  $0.069 \text{ min}^{-1}$  for GAC,  $0.021 \text{ min}^{-1}$  for bio-ring and  $0.087 \text{ min}^{-1}$  for rod-shaped AC.

### 3.4. Prediction and validation

The prediction of the bioreactor performances could be made through all the constant value from three separate models studied. Besides that, the validity of the model could be tested. Each of the model obtained high statistically quality of the modeling ( $R^2 > 0.900$ ) for First-order, Stover-Kincannon and Grau second-order model. Fig. 8 shows the comparison between the predicted and experimental COD values for HG-SBR with GAC, bio-ring and rod-shaped AC packed fixed bed materials.

Based on the result, the predicted results were in a good agreement with experimental data which indicated by high regression value. The best fitted were recorded by rod-shaped AC. This might be due to the properties of the rod-shaped AC which larger in size compared to GAC. Smaller sizes of GAC might give the interference to the experimental data for biomass concentration as it can be include with together with the biomass collected.

## 4. Conclusion

This work investigates the biodegradation performances of hybrid growth SBR with GAC, quartz ring and rod-shaped AC as packing materials for the development of biofilm. It has been observed that the COD removal efficiencies ranging from 66 to 95% were achieved during the experimental studies with a variation of initial COD loading was range between 608 mg/L until 3012 mg/L. The rapid degradation of phenol recorded for all reactors due to the adequate amount of biomass both attached on the materials and suspended. The bacteria in hybrid growth reactor were more resist toward the toxicity of the phenol. The biofilm develop on the fixed bed packed materials was homogeneous and the thickness varied for each materials. Thicker biofilm indicated that larger number of bacteria accumulated on the surface. This would promote the removal of the substrates in HG-SBR. Bio-kinetics models such as first order, Stover-Kincannon and Grau second order kinetic model were applied for HG-SBR in degrading phenol. Stover-Kincannon and Grau second order model gave higher correlation coefficients range from 98 to 99%. Therefore, both models can be applied for the design of the HG-SBR.

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