Biosorption of Cr(III), Fe(II), Cu(II), Zn(II) lons from Liquid Laboratory Chemical Waste by *Pleurotus ostreatus*

M. Arbanah*, M.R. Miradatul Najwa and K.H. Ku Halim

Faculty of Chemical Engineering, Universiti Teknologi MARA, 40 450 Shah Alam, Selangor, Malaysia

Abstract: Heavy metals which present in the liquid laboratory chemical waste should be removed due to their nonbiodegradability and toxicity characteristics. However, the conventional treatments in the removal of heavy metals have several limitations. This encourages researchers to introduce alternative treatment methods through biosorption. The process of biosorption is depending on pH, temperature and agitation speed. In the first batch treatment study, synthetic heavy metal solution was introduced prior to the treatment of liquid laboratory chemical waste. The highest biosorption efficiency for Fe(II) and Cu(II) was found to be at pH 6 while Cr(III) at pH 5 while Zn(II) at pH 4. About 80.52% of Fe(II) and 45.20% of Cu(II) was removed at pH 6. The biosorption efficiency of Cr(III) at pH 5 is 12.47% while only 5.04% for Zn(II). The results show that agitation speed of 150 rpm and temperature of 25°C is the best condition for biosorption of heavy metals. Throughout the research, the percentange 'heavy metal' removal was found to increase with the increasing contact time between *Pleurotus ostreatus (P.ostreatus)* and liquid laboratory chemical waste. The contact time can rise up to 10 hours. Almost 17.02% of Cr(III), 55.35% of Fe(II), 36.80% of Pb(II), 15.34% of Cu(II) and 13.34% of Zn(II) were removed from chemical waste under suitable treatment conditions. This validates that *P.ostreatus* is a good biosorbent agent for laboratory chemical waste treatment.

Keywords: Biosorption, laboratory waste, non-biodegradability P. ostreatus, heavy metals.

1. INTRODUCTION

In recent years, the increasing of research activities in the laboratories have promoted the production of effluent with high concentration of toxic chemical wastes. This lead to a rise in the disposal amount of chemical waste into the environment. The Department of Environment (DOE) Malaysia notified that the schedule waste generated was steeply increased from 1.1 million tonnes in 2006 to 1.7 million tonnes in 2009 [1]. Chemical waste laboratory is a waste that is generated during research activities, teaching and testing [2]. Substances existed in those waste are varies from organic chemicals, inorganic chemicals, acid solutions, toxic heavy metals, hazardous chemicals and highly complex mixtures [3]. Since the production of chemical waste usually occurs in small quantity and come from various sources, generally it will be stored for longer period of time. This is due to the complex and varied composition of chemical waste. Benatti et al. (2009) claims that, the complexity of chemical waste was greater than industrial wastewater and their composition is difficult to anticipate [2]. Therefore, the complexity of chemical waste cause additional problems during disposal as compared to industrial wastewater [4]. The difficulty exists due to wide variation of chemicals involved. Normally, the difference in chemical waste generation and operations always change depending on research objectives [2].

However, the major concern of chemical waste laboratory composition is its toxicity of heavy metals content. In fact, researches acknowledge that heavy metals cannot be eliminated by physical and chemical treatment, but they can only be stabilized [3].

The strict regulation on maximum acceptable concentrations of heavy metals in wastewater discharged into water and drinking waters, has triggered researchers to seek for suitable treatment methods to replace the conventional methods. Previously, removal of heavy metal from liquid laboratory chemical waste usually being achieved by physical-chemical process, landfill or incineration before discharging into natural body water system [3, 5]. In particular, the physical and chemical process involve chemical precipitation [6, 7], electrochemical treatment, reverse osmosis, ion exchange and adsorption on adsorbents [6]. Some of biomaterials, coconut coir has been recognized as highly potential adsorber in chemical waste treatment [8]. However, the conventional methods have drawbacks including generating toxic sludge product from treatment and this requires another special treatment with great difficulty [9]. Other than that, the high cost of adsorbent and the effectiveness of treatment are questioned when involving heavy metals in low concentration in the range of 1-100 mg/L [10, 11]. The problems have led to the restrictions of this conventional treatment methods in the treatment of chemical waste.

Based on the above mentioned problems, biosorption was proved to be an alternative technique

^{*}Address corresponding to this author at the Faculty of Chemical Engineering, Universiti Teknologi MARA, 40 450 Shah Alam, Selangor, Malaysia; Tel: +603-55444858; Fax: +603-55436300; E-mail: arbanahm@yahoo.com

for heavy metal biosorption in waste water treatment [12]. Biosorption is known to be more environmentally friendly because it leads to innocuous end product [13]. Currently, microbial potential has been utilized extensively in wastewater treatment, especially in the removal of metal contaminated effluent [14]. Based on previous study, microorganisms such as bacteria, algae and fungi are able to remove heavy metals effectively [15]. Among them, a microbial type of fungus is chosen. It could significantly reduce the concentration of heavy metals from ppm to ppb level which emerged to be potential treatment methods [10]. Both of living and dead fungi has been utilize in metal uptake [16]. The attention is given to fungi due to their good performance, low cost and eco- friendly option [17] and their availability in larger quantities [18, 19]. Fungi have chitin in their cell walls which can tolerate higher concentration pollutants and are able to grow on medium at low pH with low nitrogen content and low temperature.

Currently, white-rot fungi named P. ostreatus has been studied as a potential biomass in the removal of heavy metals from synthetics solutions as well as wastewater effluent from electroplating industry and tannery effluent [20, 21]. In a study done by Kapoor et al. (1999), P. ostreatus was chosen based on its simple growth requirements using common fermentation techniques and inexpensive medium [18]. However, P. ostreatus is not yet being applied in the treatment of liquid laboratory chemical waste. The uptake of heavy metal ions from chemical waste by P. ostreatus may offer another alternative method with great efficiency. Therefore, the aims of this research are to evaluate the potential of P. ostreatus as a heavy metal biosorbent in the liquid laboratory chemical waste treatment and also to determine the the biosorption influencing factors' such as pH, temperature and agitation speed on synthetic metal ion solutions before introducing to the real laboratory chemical waste.

2. MATERIALS AND METHODS

2.1. Samples and Microorganism Preparation

Samples of liquid laboratory chemical waste were taken from the chemistry and environmental research laboratory at Faculty of Chemical Engineering Laboratory, Universiti Teknologi MARA, Malaysia. The samples were taken since July 2010 and kept at 4°C prior to characterization and biotreatment [22]. Microorganism *P. ostreatus* was supplied by C&C

Mushroom Cultivation Farm located in southern state of Johor, Malaysia. It was subcultured every 10 days using Potato Dextrose Agar (PDA) medium (Merck).

2.2. Chemical Waste Characterization

Chemical Oxygen Demand (COD) was measured according to closed reflux method using Hach spectrophotometer DR/2800 in unit mg/L. Measurement was done by adding samples into vial containing COD reagents and heated at 150°C for 2 hours. The color forms in the vial was measured calometrically using Hach spectrophotometer DR/2800. Heavy metal concentration (mg/L) in chemical waste using atomic absorption was determined by spectroscopy (AAS) (PerkinElmer Analyst 700, USA) with background corrector. deuterium All measurements were carried out in an air/acetylene flame. A 10 cm long slot-burner head, a lamp and an air-acetylene flame were used throughout the analysis.

2.3. Cultivation and Inoculation of *Pleurotus* ostreatus

P. ostreatus was cultivated in a Petri dish contains potato dextrose agar PDA (Merck) consists of D(+) glucose 20 g/L, agar-agar 15 g/L and potato inlusion 4 g/L at 28±1°C for 10 days until sporulation occured. The inoculums was prepared by transferring 3 mycelium agar disk (8 mm diameter) taken from the edge of white colony P. ostreatus on PDA into 250 mL Erlenmeyer flask containing 100 ml of malt extract medium. Then, it was incubated in a rotary shaker at 150 rpm at 28±1°C for 15 days. Malt extract medium was prepared by adding 20 g of malt extract into 1000 mL of distilled water. P. ostreatus was separated by culture broth by filtarion and washed several times with distilled water to remove culture broth. The growth media pH was kept constant at pH 6 and was exposed under UV light for 15 minutes before used and all the transferring activities was done under the laminar flow. The sterilization was carried out in autoclave at 121°C for 120 minutes.

2.4. Preparation of Synthetic Heavy Metal lons Solution

The stock solutions of analytical grade metal salts were prepared by dissolving $Cu(NO_3)_2$, $FeN_3O_9.9H_2O$, $CuSO_4.5H_2O$, $ZnCl_2$, $N_2O_6.Pb$, CrO_3 in distilled water. The pH in metal ions solutions were adjusted to desired values with 1 M HCl or 1 M NaOH. Fresh diluted solutions were used throughout each of experiments.

2.5. Analysis of Heavy Metal Removal in Batch Process

First batch experiments were conducted using synthetic heavy metals solutions where the effect of pH, temperature and agitation speed were evaluated. The heavy metals removal uptake was performed in 250 mL Erlenmeyer flask containing 100 mL desired concentration of heavy metals liquid at 25°C in the incubator shaker at 150 rpm. The affect of pH was studied by varying pH between 2 to 6 by adding 1 M HCI and 1 M NaOH. The pH was adjusted at the beginning of experiment and not being controlled throughout the study. 6 g of living P. ostreatus (approximately 100 pellets) with average size between 4 to 5 mm were added into 100 mL synthetic metal ions solution. The effect of contact time between P. ostreatus and metal ion solution were studied by collecting about 5 mL of sample after 1 hour while other parameters were kept constant. The experiments were repeated at different agitation speeds (50, 100, 150, 200 rpm) and temperatures (20, 25, 30 and 35°C). Each experiment was followed by centrifugation and filtration through Whatman filter paper No.1. The residual filtrates were placed in incubator at 20°C before being analyzed using AAS. After suitable operating treatment conditions (pH, temperature and agitation speed) were identified from previous batch studies, the biosorption study was conducted using liquid laboratory chemical waste.

2.6. Analysis of Biosorption Efficiency

The amount Cr(III), Cu(II), Fe(II), Zn(II) and Pb(II) ions absorbed by each gram of *P. ostreatus* and the efficiency of biosorption (E) were calculated using following equation:

$$E = \left(\frac{Ci - Cf}{Ci}\right) \times 100$$
⁽¹⁾

Where Ci is the initial concentration (mg/L); Cf is the final concentration (mg/L).

3. Results and Discussions

3.1. Characteristics of Chemical Waste

Two types of samples obtained from chemical waste container at the university laboratory were characterized and compared with Standard B guideline set up by Department of Environment (DOE) Malaysia. The waste was produced from the experiment done by undergraduate students laboratory on COD analysis and an experiment entitled 'Determination of Chromium'. COD, pH, and turbidity before treatment were determined and presented in Table 1. Since the chemical waste of Sample 2 involved a higher concentration of acid sulfuric, thus it resulted in low pH which was less than one. Furthermore, Sample 2 contains high organic contents which contributed to higher COD value. The main heavy metal ion concentration involves in each experiments were also presented in Table 1.

3.2. Synthetic Heavy Metals Biosorption Studies

3.2.1. Effect of Contact Time

The effect of contact time was determined by taking samples at certain time interval within 10 hours (Figure 1). Figure 1 shows the removal of heavy metals ion at different contact time interval. It was identified that within the first 10 minutes, there was a rapid increase biosorption efficiency of Fe(II) to 89.84%, Cu(II) to 44.56% and Cr(III) to 78.65%. It was necessary to evaluate the suitable contact time for a rapid uptake so that small absorbent volume can be used [23]. A

Test Parameter	Unit	Sample 1 ^ª	Sample 2 ^b	Standard B*
Temperature	°C	28		40
рН		3.03	< 1	9
COD	mg/L	15, 000	30, 500	100
Cr(III)	mg/L	2.9	-	1.00
Fe(II)	mg/L		272.05	
Cu(II)	mg/L		0.33	
Zn(II)	mg/L		1.26	
Pb(II)	mg/L		0.49	

Table 1: Characteristics of Liquid Laboratory Chemical Waste

*Environmental Quality Act, 1974 Environmental Quality Regulations (Sewage and Industrial effluents), 1979.



Contact time

Figure 1: Effect of contact time on biosorption efficiency of Cu(II), Fe(II) and Cr(VI) by *P. ostreatus*. Biosorption conditions: Initial metal ions concentration, 20 mg/L; volume of metal ions concentration, 100 mL; wet weight of *P. ostreatus*, 6 g; agitation speed, 150 rpm.

constant biosorption efficiency was achieved at one hour on absorption process where the maximum removal was occurred with values of 91.98% for Fe(II), 56.56% for Cu(II) and 22.30% for Cr(VI). The constant value at equilibrium stage is due to the vacant space is fully filled by heavy metal ions and after some time, there is no available place for heavy metals ion binding to the cell wall surface. As a result, a repulsive force occurred. The results revealed that biosorption of heavy metals ions occurred in two stages of process where the initial rapid uptake due to surface absorption to the cell walls, and subsequent slow uptake due to membrane transport into the cytoplasm of the cells Therefore, the subsequent studies were [24]. conducted at maximum biosorption as early as one hour of contact time.

3.2.2. Effect of pH

Among the most important parameters to be monitored during the biosorption is the pH of the aqueous solution [25]. The study of pH is important because it influence the site dissociation of functional groups and solution chemistry of heavy metals. Solution chemistry includes hydrolysis, complexation by organic/inorganic ligands, redox reactions. precipitations, speciation and biosorption availability of heavy metals [26]. In this study, the effects of initial pH of aqueous solution, between 2 to 6, were investigated because of the fact that biosorption is meaningless at higher pH [21]. Figure 2 shows the comparison of percentage biosorption efficiency of heavy metals ions removal from aqueous solution by *P. ostreatus* at different operating pH.

It can be seen that the biosorption efficiency of P. ostreatus on Fe(II) and Cu(II) ions was gradually increased with solution pH up to 6. It was found that, the highest biosorption efficiency for Fe(II) and Cu(II) were 80.52% and 45.20% which occurred at pH 6. However, it is different with Zn(II) and Pb(II) where the highest biosorption efficiency was achieved at pH 4 (5.04%) and pH 5 (12.47%). Almost similar results with Javaid et al. [21] were obtained in this research, wherein the maximum biosorption of Cu(II) and Zn(II) were between pH value range of 4.5 to 5. Kapoor et. al. [18] also reported that the biosorption sharply increased when pH of the solution increased up to 4. As the pH increased, the negative charge density on the cell surface increases due to the deprotonation of the metal bindings sites which effect the increment of efficiency [18, 27]. The result for Pb(II) was similar with the study done by Vimala and Das [28]. It was found that the suitable pH using others oyster mushrooms Pleurotus platypus and Agaricus bisporus were at 5.

Dursun (2006) revealed that increasing pH which is higher than optimum pH can lower the biosorption efficiency. This can be seen from the results of biosorption efficiency for Pb(II) and Zn(II), which decreased from 12.47% to 11.58% (pH 5 to 6) and



Figure 2: Effect of pH on biosorption capacity of Fe(II), Cu(II), Zn(II), Pb(II) by *P. ostreatus*. Biosorption conditions: Initial metal ions concentration, 20 mg/L; volume of metal ions concentration, 100 mL; wet weight of *P. ostreatus*, 6 g; agitation speed, 150 rpm; contact time, 1 hour.

5.04% to 2.83% (pH 4 to 6), respectively [25]. Some authors proved that the situation of lower biosorption efficiency at higher pH was due to metal hydrolysis process caused by the formation of hydroxylated complexes, wherein at higher pH, metal ions such as Zn(II) and Pb(II) will formed Zn(OH)₂ and Pb(OH)₂ [29, 30]. However, the lowest biosorption was obtained at pH 2 for Fe(II) (30.02%), Cu(II) (6.55%), Pb(II) (8.57%), and Zn(II) (1.83%). At pH 2, the solution was found to be filled with hydrogen ion (H^{+}) which is more affinity at binding sites of *P. ostreatus* than heavy metal cations. This reason explained further by Bayramoglu and Arica [31]. The result on low biosorption efficiency occurred at lower pH which is similar to the study done by Kapoor et. al. [18]. It was found that biosorption of metal ions inhibited at pH 3.

The biosorption efficiency for synthetic solution is in the order of Fe(II)> Cu(II)> Pb(II)> Zn(II). The results proved that the difference in the biosorption efficiency for each metal depends on the type of metal cations. These results shows that there was a relationship to the mechanism of biosorption-cation-exchange process [27]. Apparently, pH is one of the important parameter need to be considered during biosorption process of heavy metal from liquid laboratory chemical waste and the selection of pH must be suitable. However, it must not be toxic to *P. ostreatus*. Chojnacka [27] found that suitable the pH to perform metal-binding experiments at ambient temperature was pH 5. This is similar to the conditions of present experiments. Therefore, it was clearly shows that pH between 4 to 6 was suitable for the biosorption of Fe(II), Cu(II), Zn(II) and Pb(II).

3.2.3. Effect of Temperature

The effect of temperature during biosorption of Cu(II), Fe(II), Zn(II) and Pb(II) by P. ostreatus was studied at temperature between 20 to 35°C. Based on the results obtained, the biosorption efficiency increased at 25°C but decreased at 35°C (Figure 3). As temperature increased from 20°C to 25°C, biosorption efficiency increased due to higher affinity of active sites for heavy metals ion attraction. It was found that the highest biosorption efficiency obtained for Cu(II), Fe(II), Zn(II) and Pb(II) were 84.85%, 44.95%, 6.49% and 8.5%, respectively. This indicates that 25°C was the best temperature for biosorption process. The temperature is suitable for the cell wall components of P. ostreatus mycelium to reorient and allowed more binding. Since the biosorption efficiency of heavy metals ion is an exothermic process, the increasing temperature from 25 to 35°C decreased the biosorption efficiency. The results also show that Fe(II) biosorption decreased from 84.85 % to 69.73 %, Cu(II) from 44.95 % to 13.84 %, Zn(II) from 6.49 % to 3.86 % and Pb(II) from 8.4 % to 7.85 %. It was believed that at 35°C, some of the active sites responsible for biosorption on



Figure 3: Biosorption efficiency (%) of *P. ostreatus* at different operating temperature. Biosorption conditions: initial metal ions concentration, 20 mg/L; pH 4(Fe), pH 5(Pb(II), pH6(Fe(II), Cu(II)); wet weight of *P. ostreatus*, 6 g; contact time, 1 hour; agitation speed, 150 rpm.

the biosorbent surface were distorted due to high temperature [26]. Furthermore, too high temperature is unfavorable for practical application because it can increase operating cost.

Figures 4, 5 and 6 show the reduction of biosorption efficiency of Cr(III) ions with increasing temperature from 25 to 35° C. The contact time was up to 72 hours. It was determined that the maximum removal of Cr(III)

ions was at pH 5 (25°C) with biosorption efficiency of 17.02%. It was the highest biosorption efficiency compared to pH 7 (15.10%) and pH 9 (12.59%) at the same temperature. It was contributed by the lower proton competition between Cr(III) and H⁺ ions. Thus, it increased the potential of Cr(VI) to bind to *P. ostreatus* active sites. It was found that during 2 hours of contact time, the affinity of *P. ostreatus* surface for Cr(III) biosorption increased to 15.07% compared to 7.75% at



Figure 4: Biosorption efficiency of Cr(III) on effect of temperature, 25 and 35°C at different contact time (hours). Biosorption condition: agitation speed 150 rpm; pH 5.



Figure 5: Biosorption efficiency of Cr(III) on effect of temperature, 25 and 35°C at different contact time (hours). Biosorption condition: agitation speed 150 rpm; pH 7.

 35° C (Figure 4). It also was found that at pH 7 for 2 hours contact time, the difference was 2.72% (Figure 5) and at pH 9, the difference was 9.43% (Figure 6).

A big difference in biosorption efficiency (9.43%) was found at temperature between 25 and 35°C at pH 9 with (Figure 6). The decreased in biosorption

efficiency was due to the damage of active binding sites at higher temperature [26]. At pH 9 and temperature of 35° C, the lowest biosorption efficiency of Cr(III) was determined after 72 hours of contact time with a value of 7.81%. It was mentioned that higher temperature will decrease the biosorption efficiency,



Figure 6: Biosorption efficiency of Cr(III) on effect of temperature, 25 and 35°C at different contact time (hours). Biosorption condition: agitation speed 150 rpm; pH 9.



Agitation speed (rpm)

Figure 7: Biosorption efficiency (%) of *P. ostreatus* at different agitation speed. Biosorption conditions: initial metal ions concentration, 20 mg/L; wet weight of *P. ostreatus*, 6 g; pH 4, Zn(II); pH 5, Pb(II); pH6, Fe(II) and Cu(II); temperature, 25°C; contact time, 1 hour.

wherein during the biosorption process of Cr(III), there was distortion of some sites of the cell surface occurred [32]. At pH 9, the percentage removal of Cr(VI) was low. The maximum uptake was at 48 and 72 hours were only 12.59% and 8.90% of biosorption efficiency was obtained (Figure **6**). The lower removal at pH 7 was caused by the metal hydrolysis process where there was a formation of hydroxylated complexes of Cr(III) [28, 30]. Overall, Cr(III) were effectively absorbed by *P. ostreatus* at the temperature 25°C with the best pH of 5.

3.2.4. Effect of Agitation Speed

The boundary layer which surrounds the biosorbent contains important active site for biosorption process. This boundary layer became an external mass transfer between two phases of biosorbent and heavy metals ion solution. Since the biosorption rate was dependent on the external film diffusion, appropriate agitation speed is important to minimize mass transfer resistance [33]. Figure 7. shows that the biosorption increased with increasing agitation speed from 50 to 150 rpm but it decreased at 200 rpm. Absorption was maximum at 150 rpm with efficiency removal of 84.85%, 44.95%, 9.05%, 1.05% for Fe(II), Cu(II), Pb(II), Zn(II). However, lowest biosorption was determined at 50 rpm for Fe(II), Cu(II), Pb(II) and Zn(II), with 64.7%, 35.15%, 8.25% and 1.05% were obtained, respectively. It was believed that at 150 rpm, all active binding sites on the biosorbent surface were sufficient enough for metal ions biosorption, thus, it made more binding to occur. At the agitation speed of 200 rpm, a lower efficiency in biosorption was identified with values of 69.40%, 35.92%, 7.55%, 1.2%, 14.36% and 35.92% for

Fe(II), Cu(II), Pb(II) and Zn(II), respectively. This might be due to the vortex phenomena occurred at these conditions. Therefore, there is no chance for heavy metals ion to bind on the surface of P. ostreatus [34]. The lowest removal efficiency was observed at 50 rpm for Fe(II), 64.71%; Cu(II), 35.15%; Pb(II), 8.25% and Zn(II), 1.05%. At lower speed, the lower dispersion of biosorbent in aqueous solution reduces the capability between surface and heavy metal ions to contact with each other. During this condition, the biosorbent is not fully dispersed in the chemical waste which led towards agglomeration of particles and it takes more time to reach equilibrium. Javaid and Bajwa [20] also reported the same results. Hence, it is important to have a sufficient agitation speed for a maximum biosorption. Table 2 shows the best operating conditions from single solution studies.

 Table 2: Operating Conditions for Highest Biosorption from Single Metal Ions Solutions

Operating conditions	Unit		
рН		pH 6 - Fe(II); Cu(II) pH 5 - Pb(II); Cr(III) pH 4 - Zn(II)	
Temperature	°C	25	
Agitation speed	rpm	150	

3.3. Laboratory Chemical Waste Biosorption Studies

The best operating conditions evaluated from the synthetic metal ions biosorption studies was being conducted in this experiment. Figure **8** shows that the



Time (hours)

Figure 8: Biosorption efficiency of Cr(III) from liquid laboratory chemical waste sample. Biosorption conditions: wet weight of *P. ostreatus*, 6 g; pH, 5; agitation speed, 150 rpm; temperature, 25°C.

highest biosorption efficiency was obtained at 48 hours of contact time with 17.02% removal. Nevertheless, the rapid biosorption was identified at 2 hours of contact time and remained nearly constant later. Figure **9** shows the removal of Fe(II), Cu(II), Pb(II), and Zn(II) from liquid laboratory chemical waste of Sample 2. The graph shows that the highest biosorption efficiency was identified for Fe(II) with 55.35% of removal and the lowest biosorption efficiency was identified for Zn(II) with 13.35% of removal.

It was found that the biosorption of heavy metals in chemical waste is in the order of Fe(II) > Pb(II) > Cu(II)> Zn(II) with 55.35% > 36.80% > 15.34% > 13.34%. Based on the results obtained, it can be concluded that the biosorption efficiency for the laboratory chemical waste was lower than the synthetic heavy metal solution. The degree of heavy metals removal from single, multiple system and wastewaters usually is different and it is influenced by several factors. This was expected due to the complex composition of chemical waste which influenced the biosorption efficiency. This was also observed by Ahluwalia and Goya [35] in their study.

4. CONCLUSION

Based on the data presented above, *P. ostreatus* employed in this study has potential to be applied for removal of Cr(III), Cu(II), Fe(II), Zn(II), Pb(II). The



Figure 9: Biosorption efficiency (%) on heavy metals uptake from liquid laboratory chemical waste sample 2. Biosorption conditions: wet weight of *P. ostreatus*, 6 g; pH 4(Zn), pH 5(Pb), pH6(Fe(II), Cu(II)); agitation speed, 150 rpm; temperature, 25°C.

results obtained shows that pH plays an important role in the biosorption by P. ostreatus. The highest biosorption of Fe(II) and Cu(II) was found to be at pH 6 with 80.52% and 45.20% removal. However, it is different for Zn(II) and Pb(II), where the highest biosorption efficiency was achieved at pH 4 (5.04%) and pH 5 (12.47%). Increasing the pH can lower the biosorption efficiency. Higher removal of Cr(III) from chemical waste was found at pH 5 (17.02%). The best operating condition for biosorption was at 25°C with 150 rpm. The highest biosorption efficiency of Cu(II),Fe(II), Zn(II) and Pb(II) were determined at 25°C with 84.85%, 44.95%, 6.49% and 8.5% of removal, respectively. Absorption was maximum at 150 rpm with efficiency removal of 84.85%, 44.95%, 9.05%, 1.05% for Fe(II), Cu(II), Pb(II), and Zn(II), respectively.

In conclusion, the pH 5, temperature 25°C and agitation speed 150 rpm were the most effective conditions for the treatment of chemical waste. These operating conditions show significant effect towards treatment of liquid laboratory chemical waste, particularly to heavy metals like Cr(III), Cu(II), Fe(II), Zn(II), and Pb(II). Therefore, it shows that *P. ostreatus* also can be used as a low cost with abundant sources and efficient biosorbent in the removal of heavy metals in wastewater. This research is benefit in helping to reduce heavy metals content in liquid laboratory chemical waste.

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