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Biosorption of Manganese Ions (II) by Immobilized Biomass on a Mixture of Silica Gel and Zeolite

Jasmidi[a], Moondra Zubir[b]*, Agus Kembaren[c], Rini Selly[d], Albinus Silalahi[e]

[a,b,c,d,e]Chemistry Department, Faculty of Science, State University of Medan, Jl. Willem Iskandar, Pasar V, Medan Estate, 20221 Medan, North Sumatera, Indonesia

[a]Email: moondrazubir@unimed.ac.id

Abstract

This study is the absorption capacity of immobilized S. cerevisiae biomass on silica gel mixture and zeolite in the manganese (II) ion in solution. with used was interaction using batch technique of immobilized biomass weight on silica gel and zeolite with solution of manganese ion (II) at concentration of manganese solution, pH and certain contact time. Manganese (II) concentrations before and after interaction were measured by a viscous spectic genome spectrometer using a formaldoxime complex at a maximum wavelength. The result showed that the weight of immobilized S. cerevisiae biomass on silica gel and zeolite influenced the absorption of Mn (II) in solution, the optimum absorption occurred on the use of 75 mg of immobilized biomass. The absorption of manganese (II) by immobilized biomass on silica gel and zeolite is not affected by contact time from the first 5 minutes to 75 minutes of contact time. The initial pH of the solution affects the absorption of manganese (II) by immobilized biomass, the higher the initial pH of the more absorbed manganese (II) solution, the Mn (II) absorption is relatively stable after pH 7. Initial concentration of manganese (II) II), optimum uptake occurs at concentrations of 50 mg / L, with a maximum absorption capacity of 10.989 mg or 2.0 x 10-4 mol of Mn (II) / g of immobilized S. cerevisiae biomass. The binding energy between S. cerevisiae biomass immobilized with manganese (II) is -30.69 kJ / mol.

Keywords: Biosorption; S. cerevisiae; silica gel; zeolite; manganese.

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* Corresponding author.

1. Introduction

Industrial development has been able to improve the quality of human life on one hand, on the other hand it is also give negatively impact to the environment from industrial waste which is containing hazardous chemical substances that contaminate the air, water and soil. One kind of the waste that can cause pollution are heavy metals, and the main target of this pollution are aquatic system such as rivers, lakes and others. Various industries which is using ingredients or compounds containing heavy metals in the production process, causing pollution. Heavy metals which is released into the environment caused by industrial activities are As, Cd, Cr, Cu, Hg, Mn, Pb, Sb, Ti and Zn.

Heavy metals in aquatic environments have different nature with contaminants of organic substances which this pollutant never tampered [1]. Various preventive and remediation have been made to overcome this heavy metals pollution. Traditional methods such as electrolytic, cation exchange, precipitation and adsorption using adsorbents conventionally been conducted, but the results obtained are often inadequate to the problem of pollution[2]. This general methods require high operational and technology costs because it is less suitable to be applied.

To minimize the content of heavy metals as the side product of industry, the waste management system should always strived to be able to reduce levels of heavy metals to the safe limit and adorption become one of this waste management system. Inorganic solids which is used in common are the one have the active site on the surface as silanol (-SiOH), siloxane (-Si-O-Si) and aluminol (-Al (OH)). Some of the qualified inorganic solids are: silica gel, zeolite, bentonite, clay, diatomaceous earth and marine life frameworks such as chitin and chitosan. Meanwhile, according to Gadd [3], microorganisms such as yeasts, fungi, bacteria and algae can absorb heavy metals and radionuclides from their external environment. The amount of metal that being absorbed is quite large and the various mechanisms of physics, chemistry and biology may involve in the uptake of these including adsorption, precipitation, complex formation and ion exchange. The uses of biomass as bio-sorbent has several weaknesses, thereas: small size, low density pose technical difficulties in use and easily damaged due to decomposition by other microorganisms. These weaknesses have been overcome by immobilization, so that the cell biomass turn into a hard material that can be packed in a column.

Previous research Jasmidi [4], using 50 mg of biomass free *S. cerevisiae* as biosorbent to 25 ml solution of Mn (II), 40 mg / L, pH 7 and a contact time of 45 minutes, the percentage of Mn (II) that being absorbed is around 32.67%. When using only a silica gel under the same conditions Mn (II) was absorbed as 17.55%. Whereas when using biomass *S.cereviseae* immobilized on silica gel (percentage of biomass in silica gel 1.12%), with the same conditions Mn (II) was adsorbed reached 41.71%. When using a solution of Cd (II), Jasmidi[5], with the same other conditions, Cd was absorbed reached 98.45% and when using a solution of Pb (II) at pH = 6.5 with the same other conditions was absorbed around 96,27%. It phenomena shows that by the immobilization, the weakness of the uses of biomass can be overcome and the absorptive capacity of the heavy metals become larger.

The uses of biomass immobilized on silica gel for treating industrial waste, still felt quite expensive therefore it is necessary to find other materials will be used as an absorben of heavy metals without decrease the advantages contained in biomass *S. cereviseae* immobilized on silica gel, such as by mixing natural zeolite that are relatively inexpensive and easy to obtain. Previously, the used of a mixture of silica gel and zeolite as the immobilizer biomass *S. cereviseae* to absorb Pb(II) was reported by Ginting[6] which using the composition of silica gel: zeolite 1: 1 Pb (II) absorbed reach 97.78 %.

To answer the problems above, *S. cereviseae* being immobilized on a mixture of silica gel and zeolite, then contacted with a solution of manganese (II) to vary the weight of biomass *S. cereviseae* immobilized, contact time, the initial pH of the solution and the concentration of manganese (II) solution. Then by using IR spectrometry, the functional groups in the absorbent material before and after absorbing manganese (II) was identified. By performing such treatment, it is expected to be obtained by the weight of biomass, the contact time, the pH of the initial solution, and the optimum concentration in absorbing ion Mn (II) also obtained as absorptive capacity ion Mn(II), as well as the known functional groups which play a role in the absorption of Mn (II) ion.

S. cerevisiae is a species of yeast, single cell creatures lacking of chlorophyll, do not have flagella, larger than bacteria, do not form mycelium, reproduce sexually and asexually, shapes are round to oval, widely used in beverage industry such as Wine, beer and distillate result, food industry such as bread, and also used for industrial alcohol, glycerol, enzymes and coenzyme.

Taxonomic position *of S. cerevisiae* is as mention below [7].

Phylum : *Eumycates*

Class : *Ascomycetes*

Order : *Saccharomycetales*

Family : *Saccharomycetaceae*

sub family : *Saccharomycetoideae*

Genus : *Saccharomyces*

Species : *Saccharomyces cerevisiae*

In this study, the biomass used is a type of yeast as *S. cerevisiae*, immobilized on Silica gel and zeolite, the metal is Mn(II) in solution. We will investigate the immobilized biomass weight, the effect of contact time, initial concentration of a solution of manganese (II), the influence of the initial pH of the solution and determination of functional groups contained in biomass *S. cerevisiae*, silica gel, zeolite and biomass *S. cerevisiae* immobilized on silica gel and zeolite before and after absorption of Mn (II).

2. Experimental

The materials used were: biomass culture, Yeast extract, Pepton, Dextrose, Manganese (II) sulfate monohydrate ($MnSO_4$ H_2O), hydroxylamine hydroxide, 35% formalin, 37% ammonia, sodium silicate, nitric acid, sulfuric acid, zeolite and aquades. For analysis and characterization were used: IR spectrometer, spectronic genesys, digital scale, pH meter, furnace, centrifugation, shaker, incubator and other glassware.

2.1. Media Preparation

Media for *S. cerevisiae* growth, consists of 10 grams of yeast extract and 20 grams of peptone were dissolved in 1000 ml of aquades, and 40 grams of dextrose dissolved in 100 ml of aquades. This solution then sterilized at 121^o C for 15 minutes. Planting (inoculation) of *S. cerevisiae* culture in growth media of YEPD, yeast extract peptone media added with dextrose until reach 2% concentration at 30^o C for 24 hours. After the growth cell was unactive by heating in a water bath at 80^o C for 25 minutes, then separated it by centrifugation.

2.2. Natural Zeolite Activation

To activate the natural zeolite, 200 grams of natural zeolite crushed and then sieved at 200 mesh, then soaked with 200 ml of 0.1M HCl, stirred for 1 hour then washed with distilled water, the treatment was repeated again, then washed again until the pH zeolite was neutral. Then the zeolite dried in an oven at 110^o C for 2 hours. The dried zeolite was heated in a furnace at 350^o C for 3 hours, then the active zeolite will be obtained.

Furthermore, the biomass of *S. cerevisiae* immobilized on silica gel and zeolite. Prepared biomass of *S. cerevisiae*, for every 50 mg dry weight of biomass mixed with 5 ml of 6 N sulfuric acid subsequently stirred. While the suspension rapidly stirred with 10 ml of sodium silicate and 13 grams of active zeolite, the mixture stirred until gel formed for about 1 hour. The formed gel stayed for one night then cut into pieces and washed with distilled water until the water was neutral. The gel then dried in an oven at 80^o C to obtain a fixed dry weight. This dried gel then crushed and sieved with 200 mesh size. The biomass of immobilized S. *cereviseae* then ready for research.

2.3. Adsorption Procedure

Immobilized Biomass Weight Dependence

6 pieces of erlenmeyer was prepared then inserted a series of immobilized biomass of 12.5, 25, 50, 75, 100 and 125 mg mixed each with Mn (II) 40 ppm and pH adjusted 7 then shacked in a shaker for 45 minutes. The analytical suspension of the above treatment then centrifuged at a rate of 3000 rpm for 10 min. A separate solution was taken 4 ml and 2 ml of 1: 1 and 0.5 ml of the formaldoxym solution was subsequently diluted to 50 ml, the result solution was determined by absorbance at the maximum wavelength.

Contact Time Dependence

Prepared 6 pieces of erlenmeyer and then introduced a weight amount of immobilized biomass having optimum uptake at the first treatment mixed each with Mn (II) 40 ppm and pH adjusted 7 then shacked in shaker with variation of time 5, 15, 30 , 45, 60 and 75 minutes. Then treated like the above steps.

Initial Ph Solution Dependence

Prepared 6 erlenmeyer pieces and then introduced a weight amount of immobilized biomass having optimum uptake at the first treatment mixed with each Mn (II) solution which concentrated 40 ppm and the pH adjusted 2, 4, 5, 6, 7 and 8 with Using NH_3 and HNO_3 solution then shaked off in shaker with optimum time at second treatment. Then treated like the above steps. For initial concentration variation of Mn (II) solution. Prepared 7 pieces of erlenmeyer and then introduced a weight amount of immobilized biomass which having optimum uptake at the first treatment mixed with Mn (II) solution with varied concentration ie 10, 20, 30, 40, 50 , 60 and 70 ppm and the pH of the solution is set at the optimum pH of the third treatment then shaken in the shaker with optimum time at the second treatment. Then treated like the above steps.

The instrument which determined this concentration is UV-VIS Spectrophotometer. Serial data of 50 ml solution of manganese (II) containing 1.0; 2.0; 3.0; 4.0 and 5.0 ppm previously added 2 ml solution of NH_3 (1: 1) and 0.5 ml formaldoxim. Then measured its absorbance, performed in the same manner. Determination the equation of calibration curve obtained from the data series absorbance of standard solution of manganese (II). Furthermore, based on these similarities can be calculated concentrations of manganese (II) before and after treatment. The concentration of manganese (II) which is absorbed by the immobilized biomass is the difference between the concentration before and after treatment. To determine the weight of the biomass *S. cerevisiae* immobilized, contact time, initial pH and initial concentration of the optimum solution concentration data manganese (II) adsorbed on treatment is processed in units of mg Mn (II) / gram of biomass immobilized, then make into chart. The optimum phase is at the highest absorption for each gram of biomass immobilized on silica gel and zeolite. To determine the absorptive capacity and energy ties between the biomass immobilized with manganese ions, the data uptake of manganese (II) then processed using the equation Langmuir between the concentration of metals remaining in solution (Ceq) with a concentration ratio of metals the residue of the adsorbed metal concentration (Ceq / Cb). Furthermore, data processing will produce a linear regression equation. Based on linear regression equation can be determined as absorptive capacity and equilibrium constant, then based on the value equilibrium constant (K) using the formula $\Delta G = - RT \ln K$, the binding energy for ion manganese (II) which is absorbed can be calculated. To determine the functional groups contained in biomass *S. cerevisiae* immobilized, silica gel, zeolite and biomass *S. cerevisiae* immobilized on silica gel and zeolite before and after interacting with the ion Mn (II) is using IR.

3. Results and Discussion

The effects of the weight of biomass S. *cerevisiae* immobilized on silica gel and zeolite on the absorption of Mn (II) in solution using 25 ml solution of Mn (II) 40 ppm, 45 minute contact time, pH 7.0, and the varied weight of biomass *S. cerevisiae* immobilized ie 12.5, 25, 50, 75, 100 and 125 mgam shows as figure 1. It shows that the increase in weight of the absorbent cause a rise in the percentage of manganese (II) absorbed. This increase appear because the increasing number of absorbent make the active center (active sites) in absorbent material that interacts with ion manganese (II) also increase. However, if the uptake of Mn (II) is converted in units of mg Mn (II) per gram of immobilized biomass dry weight, the number of the immobilized biomass relatively proportional with an increase in weight of Mn (II) absorbed per gram of biomass immobilize, the uptake of Mn (II) is relatively high on the weight of the absorbent as 75 mg, the addition of more biomass immobilized will decrease the absorbance of Mn (II).

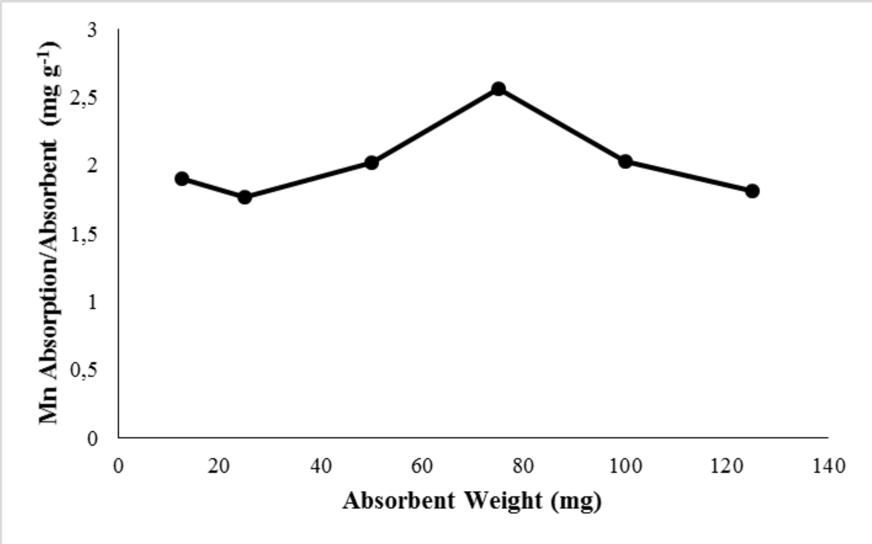

Figure 1: The influence of immobilized Mn (II) weight with immobilized biomass on silica gel and zeolite.

At this weight, indicate absorbent material arranged in such a way so as to produce a significant interaction between the active centre absorbent with a manganese ion (II). Meanwhile by increasing the absorbent weight at the comparison may not fulfilled and contributes to the effectiveness of ion absorption of manganese (II) by practitioners ie biomass *S. cerevisiae* immobilized on silica gel and zeolite. It suggest in accordance with the statement Gadd [8], about biosorption by biomass, that bio-sorption of heavy metals on the cell surface of microorganisms depends on the concentration or amount of biomass and also congestion of biomass.

The effects of contact time on the absorption of manganese (II) by biomass immobilized on silica gel and zeolite using 25 ml solution of Mn (II) 40 ppm, 70 mg immobilized biomass, pH 7.0, and the contact time was varied at 5, 1 5 , 30, 45, 60 and 75 minutes describe in figure 2. The uptake of Mn (II) by the immobilized biomass happen relatively quick, which at the contact time of 5 minutes the amount of manganese is absorbed relatively equivalent to the amount of manganese with longer contact time.

The rapid absorption of ion Mn (II) by biomass immobilized on silica gel and zeolite due to the interaction between the active groups with ion Mn (II) through chemical and physical mechanisms such as adsorption, ion exchange and complex formation. Metal absorption by biomass according to Volesky and Philips [9],the absorption of uranium by *S. cerevisiae* is a rapid process, with 60% of the amount of uranium was absorbed occurs in the first 15 minutes of contact time, Amaria[10] obtaining results that the absorption of zinc (II) by biomass *S. cerevisiae* immobilized on silica gel reaches 75.55% from the maximum amount of zinc absorbed after 5 minutes of contact time, Jasmidi[11] obtained the result that the contact time of 5 minutes amount of manganese (II) that are absorbed by biomass *S. cerevisiae* reach 72.47%.

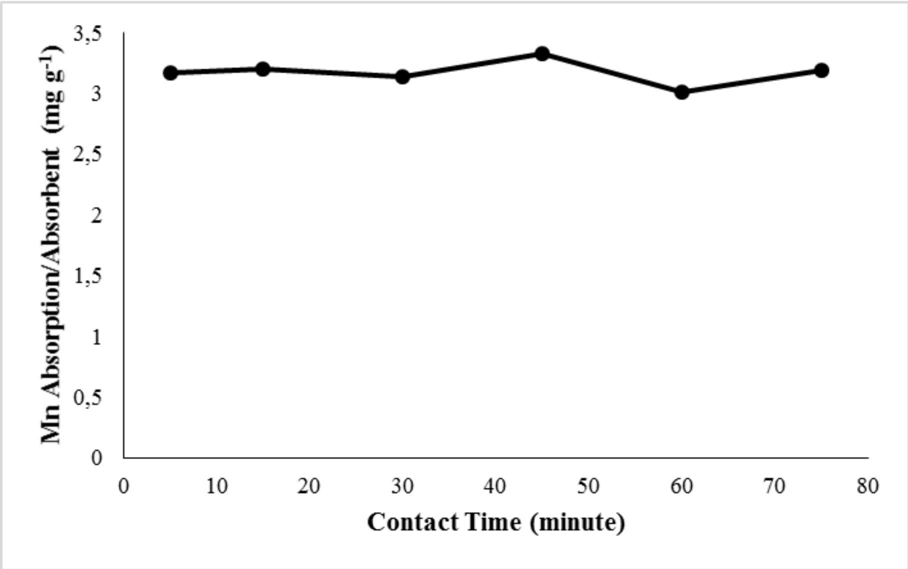

Figure 2: Effect of contact time on the uptake of Mn (II) by biomass S. cerevisiae immobilized on silica gel.

The effects of initial pH of the solution to the absorption of manganese (II) by biomass immobilized using 25 ml solution of Mn (II) 40 ppm, 75 mg of biomass immobilized, contact time of 15 minutes and the pH varied ie 2, 4, 5, 6, 7 and 8. Figure 3 shows the uptake of manganese (II) by biomass immobilized in silica gel is influenced by the initial pH of the solution. The absorption of manganese rises with increasing initial pH of the solution to pH 7.

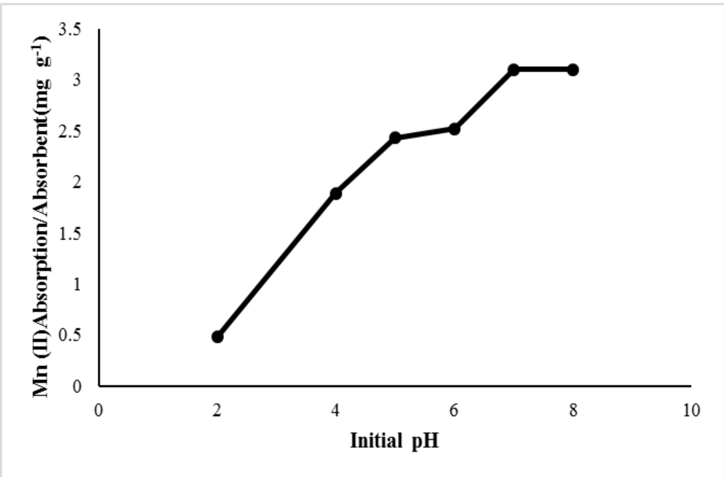

Figure 3: Influence of initial pH to uptake manganese (II) by biomass S. cerevisiae immobilized on silica gel and zeolite

It indicate the acidic (low pH) functional groups which contained in the absorbent protonated resulting of an increase in hydrogen ions and hydronium ions. Meanwhile metal ions in solution before absorbed, initially by undergo hydrolysis, produces protons and complex metal hydronium which is more absorbed than the free metal cation [12] according to the equation:

$$M^{2+} + nH_2O \rightarrow [M\,(OH)_n^{2-n}]^{+} + nH^{+}$$

In acidic conditions above, the equation will shift to the left side, the amount of metal hydroxo complexes will reduce and the amount of free metal cations increase. In acidic conditions the absorbent surface is positively charged and there will be repulsion between the active sites of the adsorbent with a metal ion uptake occurs consequently become smaller. According to Stum and Morgan [13], the degree of acidity (pH) greatly affects the surface charge which is owned by the suspended solids. The charge of surface depends on the degree of ionization and the pH of the medium. On silica gel, silica gel surface electric charge in the water can be explained by the acid-base properties of the surface of silica gel as hydrated as the equation:

$$Si\text{-}OH_2^{\ +} \xrightarrow{k_1} Si\text{-}OH \xrightarrow{k_2} Si\text{-}O^{-}$$

Acid **Alkali**

At low pH, the silica surface charged positive, while at high pH silica surface is charged negative, due to the release of a proton from the hydroxyl group. Likewise for a zeolite, at a low pH functional groups tend to be in a state of protonated by the acid, partially positively charged. It will reduce the ability to adsorb the metal ion Mn^{2+} as possible an interaction of the electrostatic repulsion between the active sites of the adsorbent and adsorbate (metal ions) are both charged positive. In addition it is also possible competition between H^{+} and Mn^{2+} ions to bind to the active group adsorbent. As a result, the low pH less than the amount of metal ions adsorbed and the increase in the pH will turn the adsorption into maximum. It suggest due to the increasing of pH will reduce the level of protonation of functional groups on the zeolite so partial positive charge also fell, as a result at this level of pH the adsorption of metal ions at the most optimum conditions. The similar case in biomass *S. cerevisiae*. This is according to a statement Tebo [14], which the bio-sorption process occurs as a consequence of the interaction between charged sites on the surface of the cell walls of microorganisms with other components exopolimer with positively charged metal ions or through the extracellular complexing agent with metal ions. Meanwhile Harris and Ramellow[15] estimated, the zero point charge or isoelectric point of the protein functional groups making up the cell walls of microorganisms occurs at pH 3. At pH less than 3, the active sites have a net positive charge, while at a pH greater than 3, the active site has a negative net charge. This leads to electrostatic attraction between ions of manganese (II) which is charged positive with active sites on the biomass immobilized on silica gel and zeolites are charged negative. The result in increased absorption of manganese (II) at pH greater than 3.Previously, studies indicate that the performance of zeolite changed when pH varied from 2 to 10 was reported. The removal efficiency is Zn (II) using a zeolite A of pH 2 to 6 tend to increase and capacity the optimum adsorption occurs at pH 6 and minimum adsorption occurred at pH 2 [16].

Effect of initial concentration of a solution of manganese (II) on the absorption of manganese (II) by the immobilized biomass.

The effects of the initial concentration of a solution of manganese (II) on the absorption of manganese (II) by biomass immobilized using immobilized biomass 75 mg, 15-minute contact time, initial pH 7.0, 25 ml volume of solution with a concentration of Mn (II) varied ie 10, 20, 30, 40, 50, 60 and 70 ppm. The influence of the initial concentration of Mn (II) against the uptake of Mn (II) by biomass *S. cerevisiae* immobilized on silica gel and zeolite, described form the relationship between the initial concentration of Mn (II) and Mn uptake (II) by biomass *S. cerevisiae* immobilize as in figure 4.

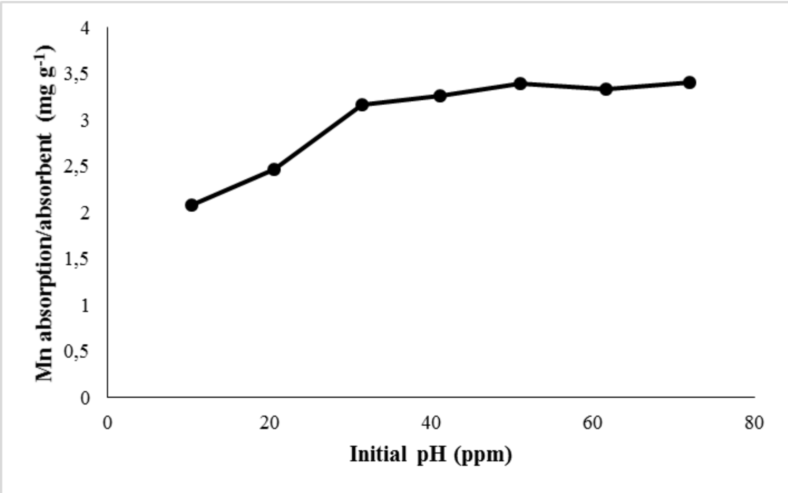

Figure 4: Dependence of the initial concentration of a solution of manganese (II) with the uptake of manganese (II) by biomass S. cerevisiae immobilized on silica gel and zeolite

It indicate the amount of manganese (II) adsorbed increases with the concentration of Mn (II) in the solution is mixed with immobilized biomass. A relatively sharp increased uptake occurs on the initial concentration of up to 50 mg / L. The addition of the initial concentration of a solution of manganese (II) relatively did not increase uptake value. This phenomenon is demonstrated by the relatively horizontal chart patterns started with the initial concentration of the solution of manganese 50 mg / L. This circumstances is in accordance with Langmuir adsorption theory about the absorbent surface that in this case is immobilized biomass *S. cerevisiae* that there are numbers of active sites is proportional to the surface area. In the active site only one molecule could absorbed. The bond between the absorbent and the absorbed substances can occur in physics or chemistry. But the bond must be strong enough to keep the transfer of molecules which have been absorbed along the surface of the absorbent [17]. Langmuir theory assumes that the absorption sites are equivalent and the bond does not depend on bond of formed on the active sites located nearby. Chemical absorption occurs due to the formation of the bond between the active site with a substance that is absorbed. This bond form a single absorption layer on the surface of the absorbent *(monolayer adsorption).* When the active sites on the surface of the absorbent is not saturated with the absorbed substances by increasing the concentration of metal ions, the amount of metal ion that is absorbed by the biomass will increase linearly. Furthermore, if the active sites on the surface of the biomass cell has been saturated with metal ions, the increased concentration of metal ions does not increase the metal ions. To gain absorption capacity for manganese (II) by biomass *S. cerevisiae* immobilized on silica gel and zeolite the data obtained linearity Langmuir absorption of manganese (II) are presented in Figure 5. By using the linear regression equation correlation coefficient (r), maximum absorption capacity and affinity or the equilibrium constant (K) for manganese uptake by biomass *S.cerevisiae* immobilized on silica gel and zeolite was obtained: 0.996; 10.989 mg/g or $2.0x10^{-4}$ mol/g; 4.0 22 L/mg or 220.960,64 L/mol, The results showed uptake of manganese (II), the largest occurred in the initial concentration of 50 mg/L, with a maximum absorption capacity of manganese (II) amounted to 10.989 mg / g or 2.0 x 10^{-4} mol/g biomass *S. cerevisiae* immobilized on silica gel and zeolite. When the adsorption energy is calculated based on the value of K using the formula $\Delta G = - RT \ln K$ at 27 °C and R = 8.314 JK^{-1} mol^{-1}, obtained adsorption energy price = 30.69 kJ/ mol.

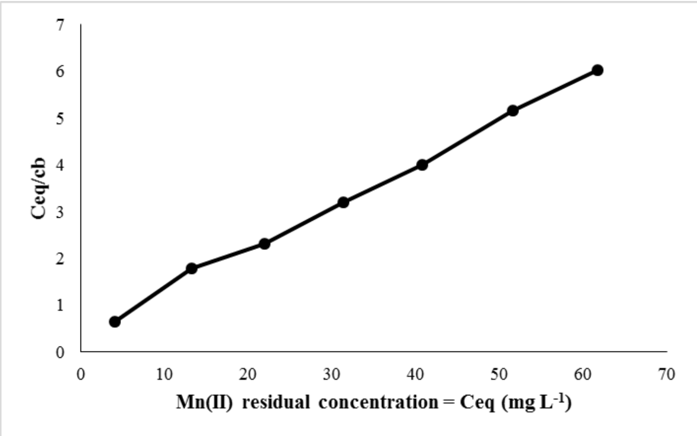

Figure 5: Linearity curve of Langmuir absorption of manganese (II) with biomass S. cerevisiae immobilized on silica gel and zeolite

Based on the adsorption energy value can be stated that the adsorption occurs is chemical adsorption. It is related to the opinion of Adamson [18], the minimum limit of the chemical adsorption energy is 20.92 kJ / mol. Results of other studies have shown that the uptake of cadmium (II), the largest occurred on the initial concentration of 60 mg / L, with a capacity of maximum absorption of cadmium (II) amounted to 53.476 mg/g or 4.758 x 10^{-4} mol / g biomass *S. cerevisiae* immobilized on silica gel and adsorption energy value = 27.09 kJ/ mol, while the uptake of lead (II), the largest on the initial concentration of 60 mg/L, with maximum capacity of absorption of lead (II) amounted to 49.505 mg / g or 2.389 x 10^{-4} mol / g biomass *S. cerevisiae* immobilized on silica gel and the obtained energy value adsorption = 28.086 kJ/ mol [5].

4. Conclusion

Severe biomass of S. cerevisiae immobilized on silica gel and zeolite affects the absorption of Mn (II) in solution which the optimum absorption occurs in the use of 75 mg of immobilized biomass. Manganese (II) absorption by immobilized biomass on silica gel and zeolite is not affected by contact time from the first 5 minutes to 75 minutes of contact time. The initial pH of the solution affects the absorption of manganese (II) by the immobilized biomass, the higher the initial pH of the solution the more manganese (II) is absorbed, the Mn (II) uptake is relatively stable after pH 7. Initial concentration of manganese (II) has an effect on the absorption of manganese (II), optimum absorption occurs at concentrations of 50 mg / L, with a maximum absorption capacity of 10.989 mg or 2.0 x 10-4 mol of Mn (II) / g biomass S. Cerevisiae immobilized. The energy price of the bond between the immobilized S. cerevisiae biomass with manganese (II) is 30.69 kJoule / mol.

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