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SYNTHESIS AND OPTIMIZATION OF CHITOSAN NANOPARTICLES OF SHRIMP SHELLS AS ADSORBENT OF PB^{2+} IONS

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Abstract

The objective of this study is synthesis chitosan nanoparticles from shrimp shells and optimization their ability as an adsorbent of Pb2+ ions. Synthesis of chitosan nanoparticles is done through several stages, namely deproteinase, demineralization, deacetylation, and resizing chitosan into nanoparticles. Deproteinase by using 2 N NaOH solution (a ratio of 1:6 w/v) while stirring at 90 °C for 1 hour. Demineralization by using 1 N HCl solution (a ratio of 1:12 w/v) while stirring at room temperature for 1 hour. Deacetylation by using 50% NaOH solution (a ratio of 1:10 w/v) at 120 °C for 3 hours. Deacetylation was performed by three times. Product of these process is called chitosan. Chitosan nanoparticles are obtained by adding a solution of 1% CH3COOH and a few drops of NH3 concentrated at 90 °C into chitosan powder to form a white gel is then washed to pH neutral and dried. Characterization of chitosan include analysis of deacetylation degree by using FTIR and analysis of particle size by using Particle Size Analyzer (PSA). Chitosan nanoparticles was then applied as an adsorbent of lead. Optimization of chitosan as an adsorbent include contact time and pH. Concentration of lead is determined using Atomic Absorption Spectroscopy (AAS). The results showed chitosan synthesis product has a size of ~600 nm, so that it can be expressed as nanoparticles with a degree of deacetylation of 62.80%. Chitosan nanoparticles as adsorbent optimum at pH 3 and a contact time of 2 hours with an adsorption capacity of 13,25 mg/g .

Keywords: chitosan, nanoparticles, shrimp shells, adsorbent, Pb^{2+} ions.

Introduction

The development of industry and mining has caused environmental pollution, especially waters. Large scale water pollution caused difficulties in the public water supply. As a result, people often use water that does not suit water quality standards for daily needs. For example, the case of pit water utilization in Bangka Belitung Islands for the production of clean water by the Regional Water Company. The opening of the subsoil in the mining process has made the mineral in the soil appears to the ground. The result is oxidation of sulfide minerals (pyrite-FeS_2) which carries the contents of heavy metals, such as lead (Pb), zinc (Zn), iron (Fe), aluminum (Al), and arsenic [1]. In addition, water pollution by heavy metals can also be caused by industrial activities, especially industry engaged in the processing of iron and steel, electroplating, mining, petroleum, tanneries, industrial batteries, automotive, and chemical industry [2].

Water contamination by metal can be reduced or eliminated in several techniques, such as adsorption, reduction, membrane processes, ion exchange, precipitation, and solvent extraction. Among these techniques, adsorption is often used for easy application, efficiently and economically. Separation by adsorption can use many adsorbents, such as zeolite, silica gel, activated alumina, activated carbon, bentonite, graphite, and chitosan [3,4]. However, in this study will be selected chitosan as an adsorbent material because it is relatively easy to obtain and has a high ability to bind metal ions.

The interaction between chitosan and metal ions occur due to ion exchange, absorption and the chelate during the process. Chitosan shows a high affinity to the metal of transition group 3, as well as metals other than alkali group with low concentrations [5]. Chitosan has a free amino group ($-NH_2$) and hydroxyl function as chelation sites with a metal ion. Chitosan is a chitin derivative obtained through deacetylation. Chitin is an organic constituents derived from crustacean, Orthopoda, annelids, Mollusk, and Nematodes. Chitin normally binds to the protein. In addition to skin and skeleton, chitin is also contained in the trachea, the gills, the intestinal wall, and the inside of the squid. However, most sources of chitin obtained from shrimp and crab

shells. Shrimp shells contains 24.3% of the dry weight [6].

Chitosan performance is strongly influenced by its characteristics, both chemical and physical. Chemical characteristics that are important for the interaction of chitosan-metal that is the degree of deacetylation (DD) [7], while the physical characteristics is chitosan particle size. The smaller the particle size, particle surface area will be greater thereby enhancing the ability of chitosan as adsorbent, especially chitosan in nano size [8]. This study aims to synthesize chitosan nanoparticles from shrimp shells and determine its adsorption capability to Pb^{2+} ions.

Observation And Analyzation

Materials: shrimp shells (*Litopenaeus vannamei*), glacial acetic acid, $PbNO_3$, NH_3, HNO_3, distilled water.

Instruments: Fourier Transform Infrared Spectroscopy (FTIR) Nicolet Ovator 360 IR, Particle Size Analyser (PSA), Atomic Absorption Spectroscopy (AAS) Pinaacle 900T Perkin Elmer, Blenders National, magnetic stirrer, hot plate, oven, analytical balance, desiccator, Buchner, Whatman 41, sieves 150 mesh, and glass equipment.

Sample Preparation: Shrimp shells (*Litopenaeus vannamei*) peeled, cleaned, and dried at a temperature of 80 °C. Furthermore, shrimp shells mashed in a blender, then sieved with a 150 mesh sieve.

Synthesis of Chitosan Nanoparticles
Deproteinase: Shrimp shells that have been refined added 2 N NaOH solution with a ratio of 1:6 (w/v) with stirred and heated at a temperature of 90 °C for 1 hour. Once separated from the solution, shrimp shells washed with distilled water until the pH is neutral and then dried at 80 °C for 8 hours.

Demineralization: Dry solids deproteinase results subsequently demineralized by using a solution of HCl 1 N (ratio of 1:12 w/v) and stirred at room temperature for 1 hour. Once filtered, the solids are washed with distilled water until the pH is neutral and then dried at 80 ° C for 8 hours.
Deacetylation: Boiled chitin in 50% NaOH solution (ratio of 1:10 (w/v)) at a temperature of 120 °C for 3 hours. The solid is then separated by liquid, then washed with distilled water until the pH is neutral. Deacetylation was performed by three times. Later, the solids are dried at 80 °C for 8 hours. This result is called chitosan.

Chitosan nanoparticles: Chitosan 1 g added 15 mL of 1% acetic acid and few drops of NH_3(c) to form a white gel with stirred. Furthermore, the gel formed is washed with distilled water to pH neutral. Then, the product is dried at a temperature of 80 °C for 24 hours.

Determination of Deacetylation Degree (DD)
Deacetylation degree was calculated by the method of FTIR spectrum base line. Absorbance value is calculated using the formula:

$$A = \log \frac{P_o}{P}$$

Information: P_o = % transmittance at baseline; P = % transmittance at minimum peak

Then, the deacetylation degree is calculated using the formula:

$$\%\text{N-Deacetylation} = \left[1 - \left(\frac{A_{1655}}{A_{3450}} \times \frac{1}{1,33}\right)\right] \times 100\%$$

Determination of Adsorption Capacity (D)
The metal concentration is determined by plotting the absorbance value obtained using AAS on the standard curve. Then, the value of adsorption capacity (D) is determined using the formula:

$$D = \frac{(C_0 - C_1) \times V}{w \times 1000}$$

Information: C_0 = initial concentration of metal (before adsorption process); C_1 = final concentration of metal (after adsorption process); V = volume of solution; w = weight of chitosan nanoparticles

Study on Adsorption Efficiency Chitosan Nanoparticles in Various Mass as Adsorbent of Pb^{2+} Ions
A solution of lead (40 ppm, 10 mL) put in a glass beaker and 0.05 g chitosan nanoparticles added while stirred for 3 hours. Then, the filtrate was filtered and analyzed by AAS. The procedure is repeated for various mass of chitosan nanoparticles (0.10 g, 0.13 g, 0.15 g).
Study on Adsorption Efficiency Chitosan Nanoparticles in Various Contact Time as Adsorbent of Pb^{2+} Ions

A solution of lead (66 ppm, 10 mL) put in a glass beaker and 0.05 g chitosan nanoparticles added while stirred for 15 minutes. Then, the filtrate was filtered and analyzed by AAS. The procedure is repeated for various contact time (30 minutes, 1 hour, 2 hours, 3 hours, 4 hours, 5 hours).

Study on Adsorption Efficiency Chitosan Nanoparticles in Various pH Solution as Adsorbent of Pb^{2+} Ions

A solution of lead (66 ppm, 10 mL) put in a glass beaker and its pH value is set to 2 using nitric acid. Then, 0.05 g chitosan nanoparticles was added to lead solution while stirred for 3 hours. Then, the filtrate was filtered and analyzed by AAS. The procedure is repeated for various pH (pH 2 and pH 3).

Results And Discussion

Synthesis process of chitosan from shrimp shells includes three stages, namely deproteinase, demineralization, and deacetylation. Deproteinase using 2 N NaOH solution aims to eliminate the proteins contained in the shells, such as glutamic acid. Demineralization using 1 N HCl solution (ratio of 1:12 w/v), aims to eliminate the minerals contained in the shells, such as Ca, P, Na, and Zn [9]. Deacetylation using 50% NaOH solution, intended to cut the bond between an acetyl group to the nitrogen atom (amino group, -NH). In the process of deacetylation used high concentrations of alkaline solution because the crystalline structure of chitin cells unit and the number of peptide bonds between the nitrogen atom to the carboxyl group. The next process is resizing chitosan to be nanoparticles by using a solution of acetic acid 1% added a few drop of ammonia. The resulting product was fine brownish-white powder. Rendement of the product obtained from shrimp shells by 62%. Changes of the structure of chitin into chitosan can be seen in Figure 1.

Characterization of Chitosan Nanoparticles of Synthesis Result

Characterization of chitosan nanoparticles using FTIR and Particle Size Analyzer (PSA). FTIR serves to determine the absorption of functional groups, while PSA serves to determine the particle size and distribution of size.

Based on the analysis of PSA (Figure 2) obtained chitosan particle size of ~600 nm that can be expressed chitosan products including nano particles. This is in accordance with Mohanraj-Chen (2006), nanoparticles are granular or solid particles with a size range of 10 to 1000 nm [10]. Based on the number distribution, chitosan nanoparticles are relatively homogeneous shown with a narrow number distribution.

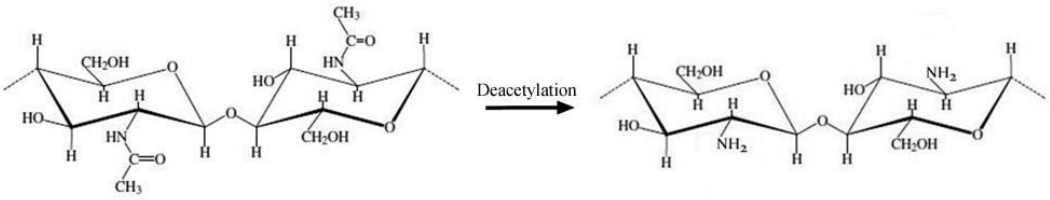

Figure 1. The chitin transformed into chitosan by deacetylation process.

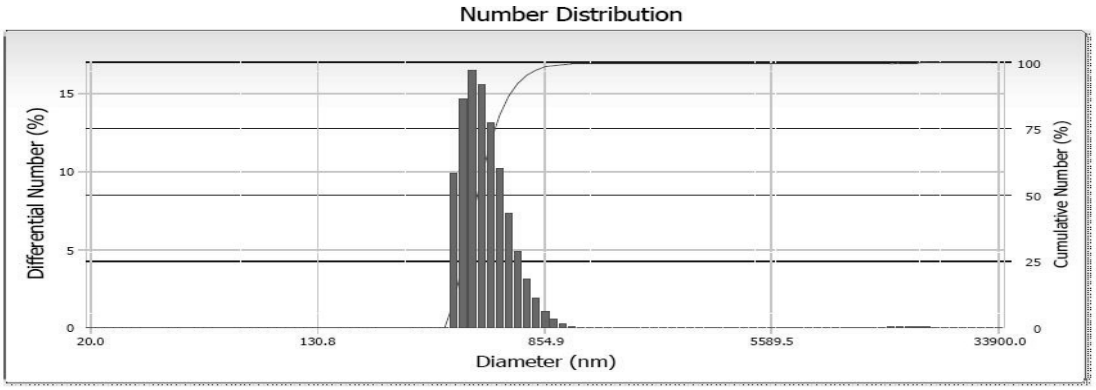

Figure 2. Particle size distribution chitosan nanoparticle using PSA. D (10%): 403.6 (nm), D (50%): 489.6 (nm), D (90%): 660.9 (nm).

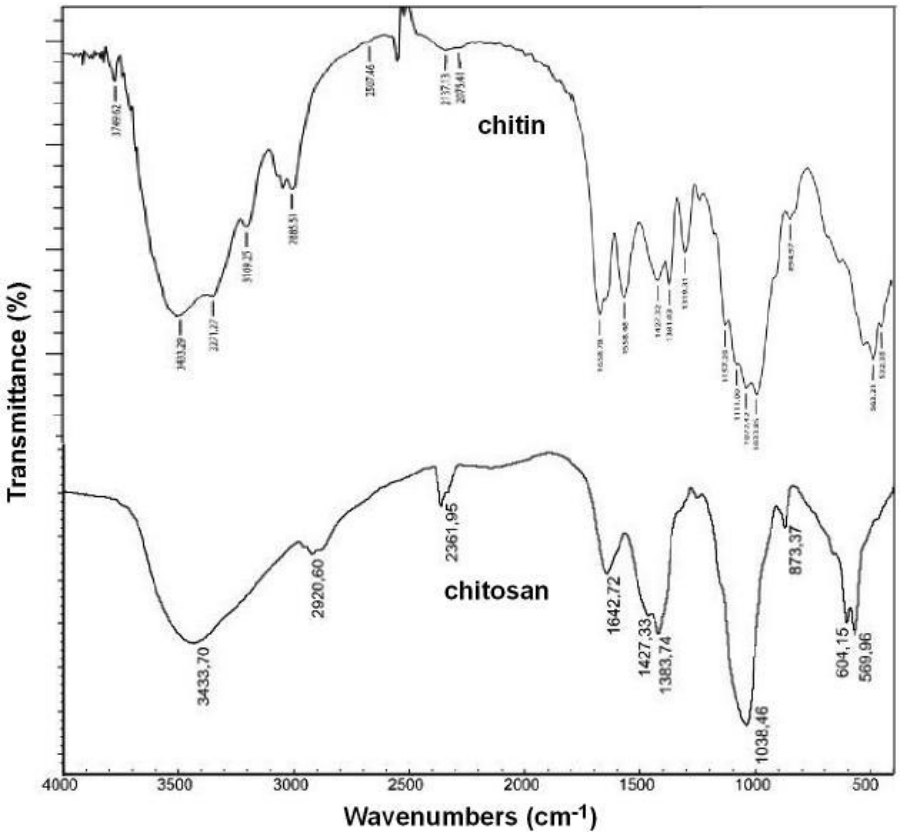

Figure 3. FTIR spectrum of chitin isolated from shrimp shells (top) and its conversion to chitosan (below).

Figure 3 shows chitin and chitosan FTIR spectra obtained in this study. Chitin FTIR spectrum show strong absorption in the area of wavenumber 3433.29 cm^{-1} which is a stretching vibration of the hydroxyl group (-OH). Medium absorption band at wavenumber 2885.5 cm^{-1} indicates -CH_2 symmetric stretching vibration. Sharp absorption at 1658.78 cm^{-1} due to stretching vibration of the carbonyl (-C=O). Absorption at 1157.29 cm^{-1} due to the -C-O stretching vibration. N-H stretching vibration occurs at wavenumber 3271.3 cm^{-1} and 3109.25 cm^{-1}. Absorption band at 1558.43 cm^{-1} is the absorption bending of secondary amine (-NH), while the absorption band at 1319.31 cm^{-1} is -CN bending absorption indicates N-acetyl group (-$NHCOCH_3$). Absorption band at 1072.42 cm^{-1} and 1033.85 cm^{-1} due to stretching vibration of C-O-C of the ring glucosamine. Based on the FTIR spectrum, isolated compounds have the ring of glucosamine and N-acetyl groups that can be expressed chitin was isolated from shrimp shells.

FTIR spectrum of chitosan (Figure 3) compared with chitin FTIR spectra observed some differences, including reduced absorption band at 2920.60 cm^{-1} indicating reduced compound having a methylene group. The loose compounds may be a carotenoid compound. It is supported by a color change of chitin whiter. Other absorption band were also disappeared i.e. stretching absorption of -NH (3271.3 cm^{-1}, 3109.25 cm^{-1}), bending absorption of secondary amine (1558.43 cm^{-1}), and C-N vibration (1319.31 cm^{-1}), indicates the loss of an acetyl group. The hydroxyl (-OH) stretching vibration of chitosan look wider than chitin allowing overlap with the stretching absorption of amine (-NH). Consequently, stretching vibration of –NH chitosan is not visible. Additionally, vibration of chitosan spectrum appear at 1642.72 cm^{-1} which is N-H bending vibration of primary amine (R-NH_2) indicates degree of deacetylation increased. The degree of deacetylation (DD) expressed the some number of acetyl groups are converted to amine groups. From these results obtained DD value of 62.80% (Figure 4). Identification of FTIR spectrum of functional groups of chitin and chitosan can be observed in Table 1.

Table 1. Identification of Functional Groups of Chitin and Chitosan

Functional Groups	Wavenumber (cm^{-1})	
	Chitin	Chitosan
-C-O-C- (glucosamine ring)	1033.85; 1072.42	1038.46
-C-O- (stretch)	1157.29; 1111	-
-C-N- (bend)	1319.31	-
N-H (bend, primary amine)	-	1642.72
N-H (bend, secondary amine)	1558.48	-
$-CH_3$ and $-CH_2$ (bend)	1427.32; 1381.03	1427.33; 1383.74
-C=O (amide)	1658.78	1642.72
-C-H (stretch)	2885.51	2920.60
-N-H (stretch)	3271.27 ; 3109.25	-
-OH (stretch)	3433.29	3433.70

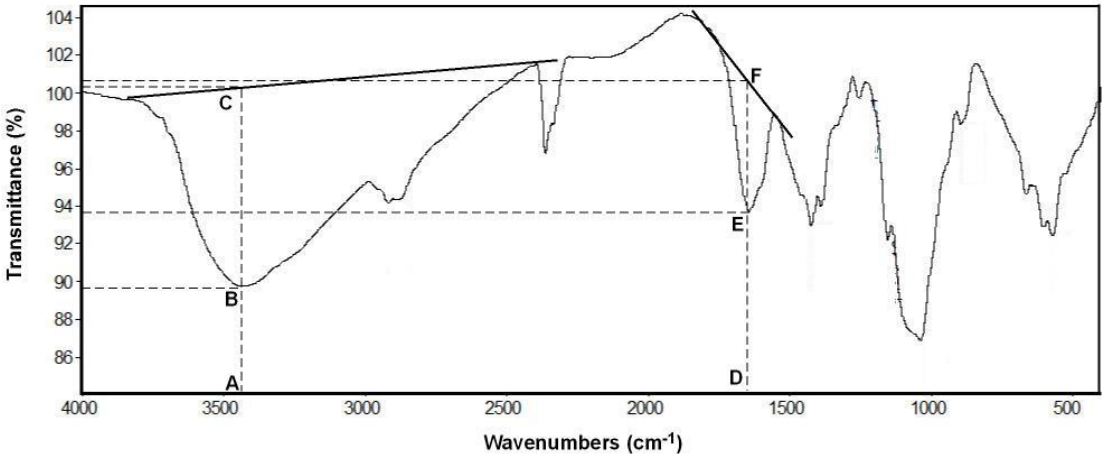

Figure 4. Determination of deacetylation degree of chitosan from shrimp shells.

Determination of deacetylation degree:

A_{1650} =

$$\log\frac{DF}{DE} = \log\frac{100.3-84}{93.9-84} = \log\frac{16.3}{9.9} = \log 1.646 = 0.216$$

A_{3450}

$$= \log\frac{AC}{AB} = \log\frac{100.1-84}{88.9-84} = \log\frac{16.1}{5.9} = \log 2.729 = 0.436$$

$$DD = \left[1 - \left(\frac{0.216}{0.436} \times \frac{1}{1.33}\right)\right] \times 100\% = 62.8\%$$

Optimization of Chitosan Nanoparticles of Shrimp shells as Adsorbent of Pb^{2+} Ions

Adsorption mechanism is the collection of pollutant particles from the solution to the surface of the adsorbent. Adsorption is affected by several factors including: the nature of the adsorbent and adsorbate, adsorbent mass, contact time, and pH. Chitosan is a cationic polymer that can bind to the metal, because the amino group of chitosan can form covalent bonds with metal ions. Moreover, the interaction between chitosan and metals is also possible due to other forces such as van der Waals forces, electrostatic forces, and hydrogen bonding.

Analysis of adsorption efficiency of chitosan nanoparticles to Pb^{2+} ions include various of mass chitosan nanoparticles, various of contact time between chitosan nanoparticles with Pb^{2+} solution, and various of pH solution. The analysis results of adsorption efficiency of chitosan nanoparticles to Pb^{2+} ions in various mass chitosan nanoparticles is shown in Figure 5. In all mass chitosan nanoparticles showed efficiencies above 99.5%. The data showed that chitosan nanoparticles has a large adsorption capacity of Pb^{2+} ions. When used as much as 0.05 g chitosan nanoparticles obtained adsorption efficiency of 99.72%. However, when mass chitosan nanoparticles be multiplied up to 3 times proved incapable Pb^{2+} ions adsorbed up to 100%. This is likely due to chitosan nanoparticles not perfect contact with the Pb^{2+} ions in solution. Generally, the greater the mass of adsorbent used, adsorption

efficiency of chitosan nanoparticles to Pb^{2+} ions increased.

The results of the analysis of the adsorption efficiency chitosan nanoparticles to Pb^{2+} ions in variety of contact time can be seen in Figure 6. Optimum contact time of chitosan nanoparticles to adsorp Pb^{2+} ions is 2 hours. At the contact time above 2 hours adsorption efficiency of the adsorbent showed improvement, but it is relatively small (<0.1%).

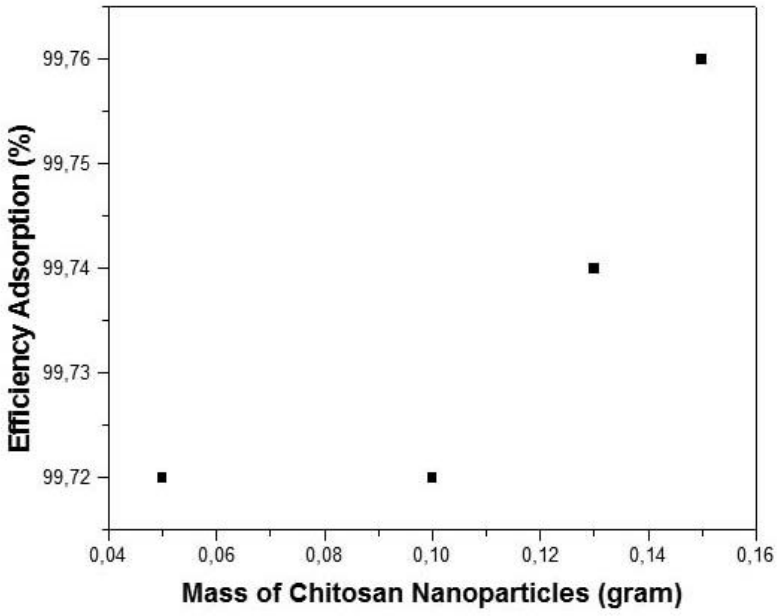

Figure 5. Adsorption efficiency of chitosan nanoparticles to Pb^{2+} ions in variety of adsorbent mass in concentrations of Pb^{2+} ions 40 ppm as much as 10 mL.

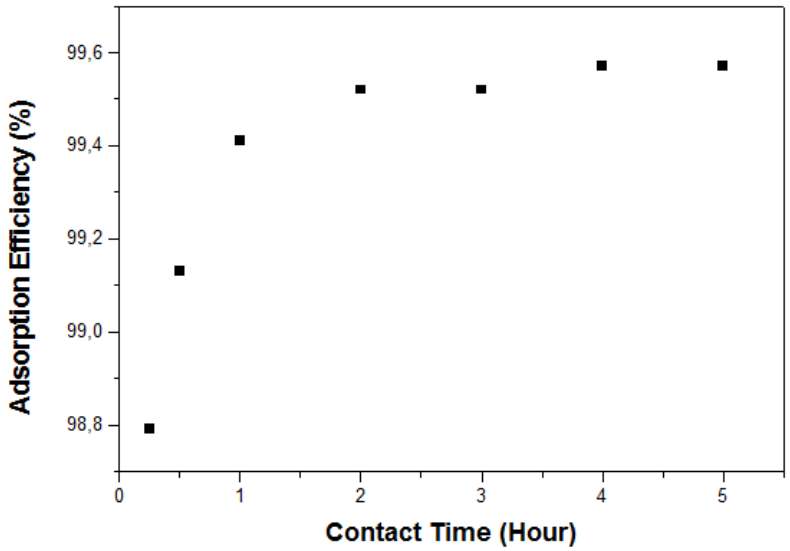

Figure 6. Adsorption efficiency of chitosan nanoparticles to Pb^{2+} ions in variety of contact time in concentrations of Pb^{2+} ions 66 ppm as much as 10 mL.

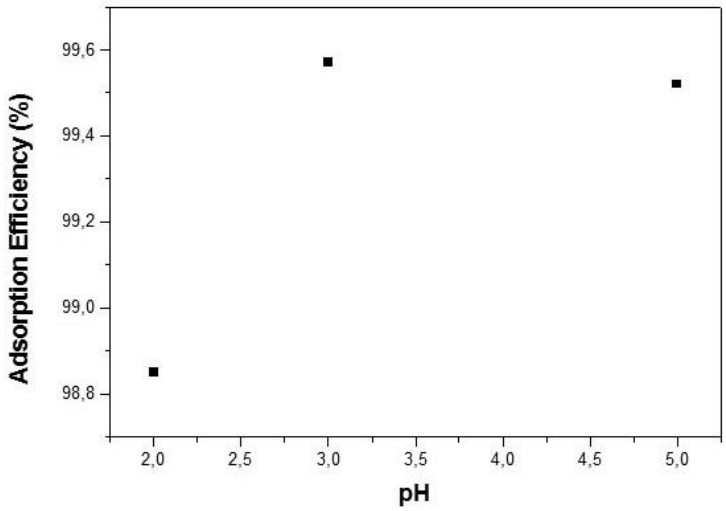

Figure 7. Adsorption efficiency of chitosan nanoparticles to Pb^{2+} ions in various of pH value in concentrations of Pb^{2+} ions 66 ppm as much as 10 mL.

Analysis of pH effect on the adsorption efficiency of chitosan nanoparticles can be seen at Figure 7. Among the three pH values, chitosan nanoparticle shows optimum adsorption efficiency at pH 3. On the pH near neutral to alkaline, adsorption efficiency of chitosan nanoparticles will be lower due to Pb^{2+} ions will precipitate. Further, the optimum adsorption capacity (D) chitosan nanoparticles to Pb^{2+} ions can be determined, i.e. 13.25 mg/g.

$$D = \frac{(66.5363 - 0.2837)\ g/mL \times 10\ mL}{0.05\ g \times 1000} = 13.25\ mg/g$$

Conclusion

Based on identification group by FTIR spectrum and Particle Size Analyser (PSA) can be stated that the study was able to isolate chitin from shrimp shells and synthesized into chitosan nanoparticles with a rendement of 62% and deacetylation degree of 62.80%. Application of chitosan nanoparticles as an adsorbent of Pb^{2+} ions indicates the optimum condition at the contact time of 2 hours and pH 3 with an adsorption capacity of 13.25 mg/g. The degree of deacetylation of the chitosan is still possible to be improved to produce a greater adsorption capacity.

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