Exploring the potential of N-acylated chitosan for the removal of toxic pollutants from wastewater

S Naidoo1,*, N Nomadolo1, W M R Matshe1, Z Cele1 and M Balogun1

1 Chemicals Cluster, Chemicals, Food, Agriculture and Health Division, Council for Scientific and Industrial Research, Meiring Naudé Road, Brummeria, Pretoria 0184, Gauteng, South Africa.

*Corresponding author E-mail: SNaidoo4@csir.ac.za

Abstract. Modified chitosan was prepared by chemical derivatisation using caproyl and palmitoyl fatty acid chains. The performance of the materials on the removal of phosphates in aqueous solutions was evaluated through a series of batch adsorption experiments. The phosphate adsorption studies showed enhanced adsorption efficiencies for caprroyl chitosan (63%) and for palmitoyl chitosan (71%) in aqueous solutions containing phosphate at a pH of 6.2. The pristine and modified chitosans showed increases in the removal efficiency at an optimal solution pH of 4. The adsorption kinetics studies performed on palmitoyl-chitosan indicated that a contact time of approximately 30 minutes is required to reach equilibrium at solution concentrations ranging from 10-20 mg/l. Co-existing anions such as nitrates and sulphates do affect the phosphate removal efficiency of modified chitosans in aqueous solutions. This study demonstrates that modified chitosans has the potential of being used as a rigid and degradable material for water treatment.

1. Introduction

Water scarcity, poor water quality and water-related disasters are among the major concerns related to current and future water resource management globally, and in particular South Africa [1]. An ongoing challenge persists in ensuring water security for the country, particularly in poor economically developed areas. It is critical to improve water resource management to ease the burden of water scarcity and availability [1]. Although conventional technologies do work, they are not as effective in treating complex and complicated polluted waters and their capability for the adequate removal of a wide spectrum of toxic chemicals and pathogens in wastewater is still limited [2-5]. This has resulted in a growing emphasis, and consequently opportunities, to develop new water treatment technologies and innovations.

Adsorption has been identified as the most suitable method for wastewater treatment in comparison to other physical and chemical methods owing to its high operation efficiency, limited power requirements, cost efficiency and robustness [5, 6, and 7]. While commercial activated carbon, a widely used adsorbent, has shown excellent adsorption capacity, it is very expensive and derived from non-renewable resources which places serious ecological strains [7].

Biopolymers have become highly attractive alternatives for adsorption owing to their availability, lower cost and lower impact on the environment [6, 7, and 8]. Chitosan is an abundant, hydrophilic biopolymer that has been identified as a promising material for water treatment applications. Its high
versatility include good adsorption, chelating and crosslinking properties [4, 9]. Owing to its multivalent backbone, chitosan can be readily chemically modified with hydrophobic moieties to yield N- and O-acylated materials. The hydrophobic modifications to chitosan have been shown to improve bacterial capture and increase its selectivity and reactivity to enhance its wastewater treatment capability [10]. This can render chitosan a good candidate for the treatment of wastewater over a wide pH range.

In this study, N-acylated chitosans were synthesized by chemical derivatisation with caproyl (C6) and palmitoyl (C16) fatty acid chains. The potential of the caproyl and palmitoyl chitosan for the removal of toxic pollutants such as phosphates was investigated by evaluating their performance in a series of batch adsorption experiments. The kinetics of the process, effects of initial concentration and pH, dosage and the effects of co-existing ions, were all used to evaluate the performance of the chemically derived chitosan.

2. Materials and Methods

2.1. Materials
Chitosan (CS) (low viscous, Fluke), caproyl chloride (d=0.976 g/ml, BioChemika Fluka), and palmitoyl chloride (d=0.906 g/ml, BioChemika Fluka) were all purchased from Sigma-Aldrich (South Africa). All other chemicals and reagents were of analytical grade, unless otherwise stated and purchased from Sigma and Merck. Deionized water (dH2O) was used in the experiments.

2.2. Synthesis of caproyl and palmitoyl chitosan
The caproyl (CC) and palmitoyl chitosan (PC) were prepared by a method adapted from Le Tien et al (2003) and Han et al (2008), with modifications as highlighted in Table 1. Briefly, chitosan solutions of varying concentrations were prepared by dissolution in 0.12 M acetic acid solution. The solutions were left to stir for 16 hours, to ensure complete dissolution.

The pH was then adjusted accordingly by the dropwise addition of 7.5 M sodium hydroxide (NaOH). Caproyl chloride and palmitoyl chloride were added in separate reaction flasks to the different chitosan solutions and were left to react at ambient temperature, 40 °C and 50 °C (Table 1). On completion of each reaction, the volume of the slurry was then adjusted to 800ml using dH2O.

| Sample ID | Acyl chloride | Chitosan concentration (%) | pH of CS solution | Acyl Chloride:CS (w/w) | Temperature (degC) | Time (Hr) | Reaction Mix:Acetone (v/v) |
|-----------|---------------|-----------------------------|-------------------|------------------------|-------------------|----------|-------------------------|
| CC1       | Caproyl       | 0.83                        | 6.3               | 1:2                    | Ambient           | 1        | 2:1                     |
| CC2       | Caproyl       | 0.83                        | 7.0               | 2:5                    | 40                | 2        | 1:1                     |
| CC3       | Caproyl       | 0.83                        | 7.5               | 1:4                    | 50                | 4        | 1:2                     |
| CC4       | Caproyl       | 1.00                        | 6.3               | 1:2                    | 40                | 2        | 1:2                     |
| CC5       | Caproyl       | 1.00                        | 7.0               | 2:5                    | 50                | 4        | 2:1                     |
| CC6       | Caproyl       | 1.00                        | 7.5               | 1:4                    | Ambient           | 1        | 1:1                     |
| CC7       | Caproyl       | 1.20                        | 6.3               | 2:5                    | Ambient           | 4        | 1:1                     |
| CC8       | Caproyl       | 1.20                        | 7.0               | 1:4                    | 40                | 1        | 1:2                     |
| CC9       | Caproyl       | 1.20                        | 7.5               | 1:2                    | 50                | 2        | 2:1                     |
| PC1       | Palmitoyl     | 0.83                        | 6.3               | 1:4                    | 50                | 2        | 1:1                     |
| PC2       | Palmitoyl     | 0.83                        | 7.0               | 1:2                    | Ambient           | 4        | 1:2                     |
| PC3       | Palmitoyl     | 0.83                        | 7.5               | 2:5                    | 40                | 1        | 1:2                     |
| PC4       | Palmitoyl     | 1.00                        | 6.3               | 2:5                    | 50                | 1        | 1:2                     |
| PC5       | Palmitoyl     | 1.00                        | 7.0               | 1:4                    | Ambient           | 2        | 2:1                     |
| PC6       | Palmitoyl     | 1.00                        | 7.5               | 1:2                    | 40                | 4        | 1:1                     |
| PC7       | Palmitoyl     | 1.20                        | 6.3               | 1:4                    | 40                | 4        | 2:1                     |
| PC8       | Palmitoyl     | 1.20                        | 7.0               | 1:2                    | 50                | 1        | 1:1                     |
| PC9       | Palmitoyl     | 1.20                        | 7.5               | 2:5                    | Ambient           | 2        | 1:2                     |

1C1-CC9 represent the caproyl chitosan samples and PC1-PC9 represent the palmitoyl chitosan samples. The pH was then adjusted accordingly by the dropwise addition of 7.5 M sodium hydroxide (NaOH). Caproyl chloride and palmitoyl chloride were added in separate reaction flasks to the different chitosan solutions and were left to react at ambient temperature, 40 °C and 50 °C (Table 1). On completion of each reaction, the volume of the slurry was then adjusted to 800ml using dH2O.
The reaction media was neutralised to a pH range of 6.8-7.0, before being precipitated using acetone. The precipitate was then collected by filtration. The precipitate was then washed three times using excess methanol at 50°C. The product was then further purified using acetone and dried in a vacuum oven at 60°C.

2.3. Fourier Transform Infrared (FTIR) Spectroscopy

The infrared absorption spectra were measured using the Perkin Elmer Spectrum 100 FTIR spectrophotometer. The pristine, caproyl- and palmitoyl-chitosan were characterised to identify the newly formed functional groups of the materials using attenuated total reflectance fourier transform infrared (ATR-FTIR). The spectra were obtained using the Spectrum version 6.2.0 Software. The spectral range was between 650 cm\(^{-1}\) and 4000 cm\(^{-1}\) with 4 scans per run and analysed at ambient temperature.

2.4. Contact Angle Measurements

Contact angles were measured using the Krüss Drop Shape Analyser (DSA) 100. The contact angle measurements were performed using the sessile drop methodology to determine the hydrophilic/hydrophobic nature of pristine and modified chitosan. De-ionised water (dH\(_2\)O) was used as the wetting medium and the analyses were performed at ambient temperature.

2.5. Batch Adsorption Experiments

A 1000 mg/l phosphate ion (PO\(_4^{3-}\)) stock solution was prepared by dissolving potassium dihydrogen phosphate in dH\(_2\)O. The solution was then further diluted according to the relevant experimental concentrations. The binary solutions were prepared by diluting the stock solutions of PO\(_4^{3-}\) ions with nitrates, sulphates or Rhodamine B dye solutions separately.

The performance of the pristine chitosan and the modified chitosans were assessed for their application in the removal of phosphate ions from wastewater. A batch adsorption screening test was conducted by treating 20 ppm phosphate ions water samples at 50 ml volumes with pristine and modified chitosan. The experiments were conducted at 25 °C, at 200 rpm for 24 hrs. 5 ml of the phosphates solutions were aliquoted and analysed using the Mettler Toledo UltraViolet5Bio (UV5Bio) to determine the amount of phosphate ions adsorbed. The effect of varying parameters including dosage, pH, initial concentration and co-existing ions were evaluated under similar conditions.

The batch absorption kinetic studies were carried out in a beaker on a magnetic stirrer using 100 ml of phosphate solution at initial concentrations varying from 10 mg/l – 20 mg/l. 0.2g of palmitoyl chitosan was added to each flask, and left to stir at 200 rpm, at room temperature for 6 hr. The samples containing the supernatants were taken at predetermined time intervals and filtered to prevent any adsorbent interfering with the subsequent UV analysis.

The PO\(_4^{3-}\) removal percentage was determined by using Eq. (1):

\[
\text{% Removal} = \left( \frac{C_o - C_e}{C_o} \right) \times 100
\]

where \(C_o\) and \(C_e\) are the initial and equilibrium concentration (mg/l) of PO\(_4^{3-}\), respectively.

The equilibrium absorption capacity was determined using Eq. (2):

\[
q_e = \left( \frac{C_o - C_e}{m} \right) V
\]

where \(q_e\) is the equilibrium amount of PO\(_4^{3-}\)adsorbed per unit mass (m) of adsorbent (mg/g) and V is the sample volume (L).

The adsorption capacity at any time (t) was calculated by Eq. (3):
\[ q_t = \left( \frac{C_o - C_t}{m} \right) V \]  

(3)

where \( q_t \) is the amount of PO\(_4^{3-}\) adsorbed per unit mass (m) of adsorbent (mg/g) at any time (t), \( C_t \) is the concentration of (mg/L) of PO\(_4^{3-}\) at time (t) and V is the sample volume (L).

3. Results and Discussion

3.1. Synthesis of caproyl and palmitoyl derivatised chitosan

In this study, synthetic N-acylated chitosan was derivatised using caproyl (C6) and palmitoyl (C16) fatty acids. A series of modified chitosan were produced by varying the processing parameters to obtain the optimal processing conditions. Both the caproyl- and palmitoyl-chitosan were successfully synthesized, with yields of up to 71% and 66% for caproyl-chitosan and palmitoyl-chitosan, respectively.

3.2. FTIR Spectroscopy

The structural composition of the pristine and modified chitosan networks were confirmed using the FTIR spectra (Figure 1). The spectra were normalized for comparison. The characteristic peaks of chitosan were observed between 3400-3000 cm\(^{-1}\) (-OH stretch) and 2871 cm\(^{-1}\) (-CH\(_2\)- stretching bond). For the pristine chitosan, the carbonyl group from amide I and amide II (C-N stretching) are represented by the bands at 1650 cm\(^{-1}\) and 1587 cm\(^{-1}\), respectively. The evaluation of caproyl- and palmitoyl-chitosan, saw shifts in the peaks of the amide I and amide II bands to lower wavenumbers as observed with respect to the pristine chitosan. The band at 1587 cm\(^{-1}\) disappear while peaks around 1646 cm\(^{-1}\) (carbonyl stretching of the amide band I) and 1548 cm\(^{-1}\) (N-H bending vibrations of the amide band II) become more prominent.

Absorbance peaks at 2850-2950 cm\(^{-1}\), ascribed to -CH\(_2\), were also observed for both the caproyl- and palmitoyl-chitosan, suggesting successful modification of chitosan with the acyl functional groups. The intensity of the peaks at 2850-2950 cm\(^{-1}\) were much higher in the case of palmitoyl-chitosan which is ascribed to its longer chain length (C16) in comparison to the caproyl-chitosan (C6 chain).

3.3. Contact Angle

Disks of pristine chitosan and palmitoyl-chitosan were prepared for the contact angle measurements using the DSA100. A qualitative analysis of the hydrophilic/hydrophobic nature of the chitosan and modified chitosan was performed by evaluating the contact angle using dH\(_2\)O droplets. As depicted in Figure 2, contact angles of 52.5 ° and 92.8 ° were observed for the pristine and palmitoyl chitosan, respectively. The increase in the contact angle of the palmitoyl chitosan can be largely attributed to the presence of the hydrophobic groups.
3.4. Batch Adsorption Experiments

Screening for the best performing material. Samples of pristine chitosan and the synthesized caproyl- and palmitoyl-chitosan were assessed for their removal properties of PO$_4^{3-}$ ions from 20 mg/l synthetic phosphate solution. The tests were conducted at an adsorbent dosage of 150 mg at 25 °C. The phosphate removal efficiency of the adsorbents are highlighted in figure 3. There was a significant improvement in removal efficiency of PO$_4^{3-}$ ions from solution using the modified chitosan (40-71%) in comparison to the pristine chitosan (21%).

The best performing caproyl and palmitoyl chitosan (CC4 and PC3) were selected for the studies to evaluate the effects of dosage and pH on the adsorption efficiency of the pristine and modified chitosan.

Effect of solution pH. The adsorption of anions and cations at the solid-liquid interface is largely affected by the pH of the aqueous solution (9). Hence the effect of the solution pH on the adsorption efficiency was investigated. Figure 4 shows the phosphate removal efficiency of the pristine and N-acylated chitosan adsorbents over a solution pH range of 2-10. The study was conducted over 24 hours at 25°C, using a known solution concentration of 20mg/l and an adsorbent dosage of 150mg. An optimal pH of 4 was observed for the pristine and N-acylated chitosan. The pH of the solution directly affects both the speciation of phosphate ions in solution and the protonation of the chitosan.
To better understand the effects of pH on the material, the ionic properties of chitosan need to be considered. It is a cationic polyelectrolyte with a pKa value of approximately 6.3. At pH < pKa, the amine groups become readily protonated, and thus electrostatic interaction takes place between the protonated amine groups and the phosphate molecules. Zero valent $\text{H}_3\text{PO}_4$ which is a non-ideal state for adsorption onto active sites due to it being non-ionic, is dominant at pH < 4, and thus low adsorption efficiencies are observed at pH 2 [13]. Monovalent $\text{H}_2\text{PO}_4^-$ and divalent $\text{HPO}_4^{2-}$, which have high affinity for adsorption sites, are the dominant species between pH 3 and 7 [14]. Thus higher adsorption efficiencies are observed in this range. The increase in pH above 7, showed lowered adsorption affinity for $\text{PO}_4^{3-}$ which may be due to competing hydroxyl (OH) ions. In general, the N-acylated chitosan performed better than pristine chitosan at the various pH ranges.

Figure 3: The adsorption efficiency of pristine and modified chitosan for the removal of $\text{PO}_4^{3-}$ ions in an aqueous solution.

Figure 4: The effect of pH on the removal of $\text{PO}_4^{3-}$ ions in an aqueous solution using pristine and N-acylated chitosan.
Effect of adsorbent dosage. To determine the effect of dosage, adsorption tests were conducted using different adsorbent dosages ranging from 50 – 250 mg. The studies were carried out for a period of 24 hours using a solution concentration of 20 mg/l at its natural pH of approximately 6.2 was used for the evaluation, while the temperature was maintained at 25°C. The phosphate removal efficiency of the pristine and N-acylated chitosan as a function of dosage is represented in Figure 5.

An increase in the adsorption efficiency with increasing dosage for both the pristine (up to 150 mg) and modified chitosan (up to 200 mg) was observed (Figure 5). No significant adsorptive effects were observed thereafter. This observed increase and subsequent plateau in adsorption can be attributed to the availability of adsorption sites.

Overall, the performance of the modified chitosan was found to be superior to the pristine chitosan and the longer chain palmitoyl performs better compared to the caproyl-chitosan. Chitosan is mainly characterised by hydrogen bonds, and upon acylation of chitosan the number of hydrogen bonds are reduced, and the modified materials are characterised by hydrophobic interactions due to the acyl chains substituted onto the chitosan [11]. The hydrophobic interactions induce greater stability of the modified chitosan matrix, and therefore enhanced adsorption is observed for the modified chitosan materials. The shorter C6 acyl chain displays weak hydrophobic interactions while the stronger hydrophobic interactions of the longer C16 acyl chain offer greater network stability of the polymer. Thus higher adsorption rates observed for palmitoyl chitosan could be attributed to this. Based on the results obtained, the palmitoyl-chitosan was selected as the adsorbent of choice for the remaining studies.

Effect of contact time. The batch kinetics study for palmitoyl chitosan was carried out at solution concentrations of 10 – 20 mg/l at a pH of 6.2 using the optimum adsorbent dosage of 200 mg. The adsorption capacity as a function of time is represented in Figure 6. Fast adsorption rates are observed in the early stage of the study, and this can be ascribed to the increase in the driving force as a result of the concentration gradient. The adsorption capacity increases with increase in contact time. A contact time of approximately 30 minutes is required for the phosphate solutions at concentrations 10, 15 and 20 mg/l to reach equilibrium, after which the adsorption capacity increases at a minute interval.

Effect of initial concentration. The effect of solution concentration on the removal efficiency and adsorption capacity of palmitoyl-chitosan was studied by varying solution concentration from 15 – 50 mg/l. The test was conducted at the optimum adsorbent dosage of 200 mg at 25°C for 2 Hr.
Figure 7 illustrates the effect of the solution concentration on both the phosphate adsorption capacity and phosphate removal efficiency of palmitoyl chitosan.

The available adsorption sites become saturated as the initial concentration is increased and hence a decrease in the removal efficiency of the palmitoyl-chitosan with increasing concentration is observed. The adsorption capacity increases with the initial solution concentration. According to Leduc et al, this trend could be a result of the effective collision between the phosphate ions and the surface of the palmitoyl chitosan as well as the concentration gradient which provides a greater driving force to lower the mass transfer resistance.

Figure 6: The effect of contact time on the adsorption of \(\text{PO}_4^{3-}\) at various initial phosphate solution concentrations.

![Figure 6: The effect of contact time on the adsorption of \(\text{PO}_4^{3-}\) at various initial phosphate solution concentrations.](image)

Figure 7: The effect of solution concentration on adsorption capacity and adsorption efficiency of palmitoyl chitosan.

Effect of co-existing ions. Wastewater is often made up of various constituents that could inhibit the overall capability of an adsorbent to remove a particular pollutant, largely due to these constituents competing for the active sites on the modified chitosan. Hence, a study was carried out to investigate the effect co-existing ions have on the removal of phosphates by the palmitoyl-
chitosan (Figure 8). The competing ions to be evaluated included anions (nitrates and sulphates) and organics (Rhodamine B).

In general, the phosphate removal efficiency of the palmitoyl-chitosan was reduced in the presence of a competing ion, with a most notable reduction in performance when phosphates and nitrates were in solution. The reduction in the phosphate adsorption in the presence of both nitrates and sulphates can be attributed to the affinity of these anions to the active sites on the modified chitosan. The presence of the Rhodamine B dye did not affect the adsorption of phosphates owing to the electrostatic repulsion between the protonated amine groups on the palmitoyl chitosan and the positively charged Rhodamine B.

![Figure 8](image-url)

**Figure 8:** The effect of co-existing ions on the removal efficiency of palmitoyl chitosan.

4. **Conclusion**

In this study, N-acylated chitosan was synthesized via chemical derivatisation of chitosan with caproyl (C6) and palmitoyl (C16) chains. The functional groups and hydrophobicity of the N-acylated chitosan were confirmed by FTIR spectroscopy and contact angle measurements, respectively. The phosphate adsorption studies indicate an enhancement in the adsorption efficiencies from 20% using pristine chitosan to 63% for caproyl-chitosan and 71% for palmitoyl-chitosan in aqueous solutions containing PO$_4^{3-}$ ions at a pH of 6.2.

However, all the materials show improved performances at the optimal solution pH of 4. The adsorption kinetics studies performed on palmitoyl-chitosan indicated that a contact time of approximately 30 minutes is required to reach equilibrium at solution concentrations ranging from 10-20 mg/l. Co-existing anions such as nitrates and sulphates do have an effect the phosphate removal efficiency of modified chitosan in aqueous solutions. Low adsorption capacities were obtained in the various studies, and hence the material will need to be optimised to increase adsorption capability of the modified chitosan. This study demonstrates that the palmitoyl-chitosan has the potential of being used as a cheap, rigid and degradable material for water treatment. Selectivity of other pollutants will also be investigated in the future.

A life cycle assessment of the N-acylated chitosan will be performed at a later stage. The study will assist in identifying ways to minimize waste generated during the treatment process and determine the environmental load of a single batch of the N-acylated chitosan. The potential use of the phosphate-loaded N-acylated chitosan as fertilizer will be investigated.
5. Acknowledgement
The authors are grateful to the Council for Scientific and Industrial Research for their financial support. Assistance from Dr. Lindani Mdlalose regarding the interpretation of the adsorption data is gratefully acknowledged.

6. References
[1] WWAP (UNESCO World Water Assessment Programme). (2019). *The united nations world water development report 2019: Leaving no one behind*. Paris: UNESCO.
[2] Nechita, P. (2017). Applications of chitosan in wastewater treatment. doi:10.5772/65289
[3] Gehrke, I., Geiser, A., & Somborn-Schulz, A. (2015). Innovations in nanotechnology for water treatment. *Nanotechnology, Science and Applications, 8*, 1-17. doi:10.2147/NSA.S43773
[4] Huang, P., Zhao, X., & Ye, L. (2016). Synthesis of hydrophobic cationic chitosan flocculant and its sludge dewatering property. *Journal of Macromolecular Science, Part B, 55*(3), 299-309. doi:10.1080/00222348.2016.1146975
[5] Rajeswari, A., Amalraj, A., & Pius, A. (2015). *Removal of phosphate using chitosan-polymer composites* doi:10.1016/j.jece.2015.08.022
[6] Leduc, J., Leduc, R., & Cabana, H. (2014). Phosphate adsorption onto chitosan-based hydrogel microspheres. *Adsorption Science and Technology, 32*, 557-569. doi:10.1260/0263-6174.32.7.557
[7] Tan, I. A. W., Ahmad, A. L., & Hameed, B. H. (2008). *Adsorption of basic dye using activated carbon prepared from oil palm shell: Batch and fixed bed studies* doi:https://doi.org/10.1016/j.desal.2007.07.005
[8] Schmuhl, R., Krieg, H., & Keizer, K. (2001). *Adsorption of cu(II) and cr(VI) ions by chitosan: Kinetics and equilibrium studies* doi:10.4314/wsa.v27i1.5002
[9] Pandey, N., Shukla, S. K., & Singh, N. B. (2017). Water purification by polymer nanocomposites: An overview. *Nanocomposites, 3*(2), 47-66. doi:10.1080/20550324.2017.1329983
[10] Muñoz-Bonilla, A., & Fernández-Garcia, M. (2012). Polymeric materials with antimicrobial activity. *Progress in Polymer Science - PROG POLYM SCI, 37*, 281-339. doi:10.1016/j.progpolymsci.2011.08.005
[11] Le Tien, C., Lacroix, M., Ispas-Szabo, P., & Mateescu, M. (2003). N-acylated chitosan: Hydrophobic matrices for controlled drug release. *Journal of Controlled Release, 93*(1), 1-13. doi:DOI: 10.1016/S0168-3659(03)00327-4
[12] Han, J., Guenier, A., Salmieri, S., & Lacroix, M. (2008). Alginate and chitosan functionalization for micronutrient encapsulation. *Journal of Agricultural and Food Chemistry, 56*(7), 2528-2535. Retrieved from SCOPUS database.
[13] Yue, Q., Wang, W., Gao, B., Xu, X., Zhang, J., & Li, Q. (2010). Phosphate removal from aqueous solution by adsorption on modified giant reed doi:10.2307/25679789
[14] Mdlalose, L., Balogun, M., Setshed, K., Chimuka, L., & Chetty, A. (2018). Adsorption of phosphates using transition metals-modified bentonite clay. *Separation Science and Technology, 1-12*. doi:10.1080/01496395.2018.1547315
[15] Liu, Q., Hu, P., Wang, J., Zhang, L., & Huang, R. (2016). Phosphate adsorption from aqueous solutions by zirconium (IV) loaded cross-linked chitosan particles doi:https://doi.org/10.1016/j.itice.2015.08.012
[16] Liu, X., & Zhang, L. (2015). Removal of phosphate anions using the modified chitosan beads: Adsorption kinetic, isotherm and mechanism studies doi:https://doi.org/10.1016/j.powtec.2015.02.05