

SYNTHESIS AND CHARACTERIZATION OF ALGINATE-CELLULOSE XANTHATE BEADS FROM CORN STALK WITH NA₂CO₃ AS POROGEN

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Abstract

In this study, the synthesis of porous beads from corn stalks was carried out. The cellulose extracted from corn stalks was converted into cellulose xanthate and combined with alginate to form porous alginate-cellulose xanthate beads by the ionic gelation method. This study attempted to use sodium chloride (NaCl) as a porogen and zinc acetate as a crosslinker. Beads were characterized to determine the porosity, swelling properties, and functional groups using Fourier Transform Infra-Red (FTIR). The geometry of beads was analyzed by optical microscopy, and its surface morphology was analyzed by Scanning Electron Microscopy-Energy Dispersive X-ray (SEM-EDX). The results showed that corn stalks as agricultural waste material could be used to synthesize porous beads material. The swelling and porosity of beads increased with increasing concentration of NaCl. The presence of porogen has increased beads formation. The results demonstrate the crosslinks between zinc acetate and alginate were successfully characterized using FTIR. NaCl concentration of 9.5% resulted in the highest swelling properties (52.80%) and porosity (81.4%) of the beads.

Keywords: synthesis of cellulose xanthate, swelling, corn stalk, porosity

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Introduction

Corn stalks are an agricultural waste that has potential as a biopolymer material. Boufi & Achraf (2016) explained that corn stalks contain 69% cellulose and 31% hemicellulose after a delignification process. The high cellulose content in corn stalks makes them suitable for producing beads. Cellulose is an abundant, biodegradable, and non-toxic material, which attracted researchers to use cellulose as a primary material for making beads. Research conducted by Sönmez *et al.* (2016) reported that the addition of cellulose in alginate/cellulose composite beads could increase the absorption of beads against Cu²⁺.

Cellulose is a long-chain polysaccharide consisting of β -D-glucopyranose units joined by β -1,4 glycosidic (Hokkanen *et al.*, 2016). The absence of branching in the cellulose chain causes cellulose to be semicrystalline, which has both an amorphous phase and a crystal phase (Harmsen *et al.*, 2010). Cellulose has a reactive group in the form of hydroxyl (-OH), which is

distributed evenly on the cellulose side, thus allowing the formation of hydrogen bonds between cellulose molecules (Harmsen *et al.*, 2010). Hydrogen bonds in cellulose are causing it to be insoluble in water and other organic solvents directly and, it needs to be converted into cellulose xanthate to reduce hydrogen bonds between cellulose. Cellulose xanthate is made by the addition of NaOH and CS₂ reagent. The addition of these compounds will replace the hydroxyl group on cellulose with the xanthate group. Alginate is a natural anionic polymer from brown seaweed. Besides having good biocompatibility, alginate can also improve mechanical strength, has low toxicity, and is inexpensive. In general, alginate is known as the overall part of the linear copolymers containing block residues (1.4) -d-mannuronate (M) and α -l-guluronate (G) (Lee and Mooney, 2012).

Alginate and cellulose can be used as the base for beads making. Beads from biopolymers can be used as drug carriers, planting media, and adsorbents. One of the commonly applied

methods for beads formation is the ionic gelation method. The ionic gelation method is based on the formation of hydrogels influenced by crosslinked agents (Patil *et al.*, 2010). Sönmez *et al.* (2016) explained the mechanism of zinc acetate as a crosslinker. Crosslinking is formed between the –COO group - from the residual guluronic alginate acid, which establishes an “egg-box” model with the negatively charged Zn²⁺ cation. The egg-box model causes the beads to have high mechanical properties (Lin *et al.*, 2012).

The adsorption capacity of beads can be increased through the use of a pore-forming agent or a porogen. The commonly used porogens are KCl, Na₂SO₄, NaHCO₃, K₂HPO₄, CaCO₃, and NaCl. The advantage of NaCl as a porogen is that the resulting surface area and the absorption capacity are higher compared to the product of solvating porogen (Mane, 2016). Additionally, the availability of NaCl is abundant and relatively inexpensive.

The purpose of this research is to make beads based on alginate and cellulose xanthate with the addition of NaCl as porogen. Cellulose xanthate used in this study came from the isolation of cellulose from corn stalks waste, which was synthesized into cellulose xanthate with the addition of NaOH and CS₂. Beads formed are then characterized for their swelling properties, changes in diameter, and morphology to determine the effect of the use of NaCl porogen on beads.

Materials and Methods

Material

Corn stalks were obtained from the district of Malang, East Java, Indonesia. All chemicals used in this study were of analytical grade, including the sodium hydroxide, chloride acid, acetic acid, zinc acetate, sulfuric acid, sodium chloride, carbon disulfide, ethanol were purchased from Merck (Germany), and sodium alginate was from PhytoTechnology Laboratories (USA).

Extraction of Cellulose

The process of bead production is shown in Figure 1. Firstly, corn stalks were cleaned and sun-dried. Dry samples were crushed into powder with a size of 100 mesh, then put into the oven for 24 h at 90 °C. Fifty grams of corn stalks powder were immersed in 1,000 mL NaOH 10% (w/v) at 80 °C for 90 mins, then washed several times with distilled water and press-filtered. The resulting pulp was treated with 200 mL NaClO₂ 1% (v/v), then CH₃COOH 10% (v/v) was added until pH 5 at 75 °C for 1 h. After that, it was washed with distilled water until the pH became neutral, then it was press-filtered. Cellulose obtained was hydrolyzed with 5% (v/v) HCl (1:20) at 95 °C for 1 h to obtain dispersed microfibril.

Synthesis of Cellulose Xanthate

Five grams of cellulose were immersed in 40 mL NaOH 20% (b/v) for 3 h. It was subsequently press-filtered and kept for 60 h at room temperature for aging. The obtained alkaline cellulose was reacted with 2.5 mL CS₂, and the sample was homogenized using a shaking incubator (shaking rate 150 rpm) at 25 °C for 3 h to acquire cellulose xanthate. After that, the suspension was dissolved in 30 mL NaOH 6% (w/w) (Wang *et al.*, 2013).

Beads Shaping

Sodium alginate (0.5 gr) was dissolved in 12.5 mL aquademin and added with a few drops of CH₃COOH until thoroughly dissolved, followed by the addition of cellulose xanthate with the ratio of alginate to cellulose xanthate of 1:3. Then porogen NaCl was added in varying concentrations of 3.3, 6.5, and 9.3% (designated as ACX-1, ACX-2, and ACX-3), whereas the beads without porogen are designated as ACX-0. Then each mixture was homogenized. As the solution formed, it was dripped using an 18 G syringe needle into a solution of 5% (w/v) of zinc acetate up to 100 mL and left for 24 h. The beads formed were filtered (Suvachittanont and Pookingdao, 2013).

Filtered beads were washed in distilled water and put in the shaker at 150 rpm for 48 h, followed by soaking for 24 h. The obtained beads were dried at 37 °C for 5 h, and the swelling properties were determined (Tas, 2008).

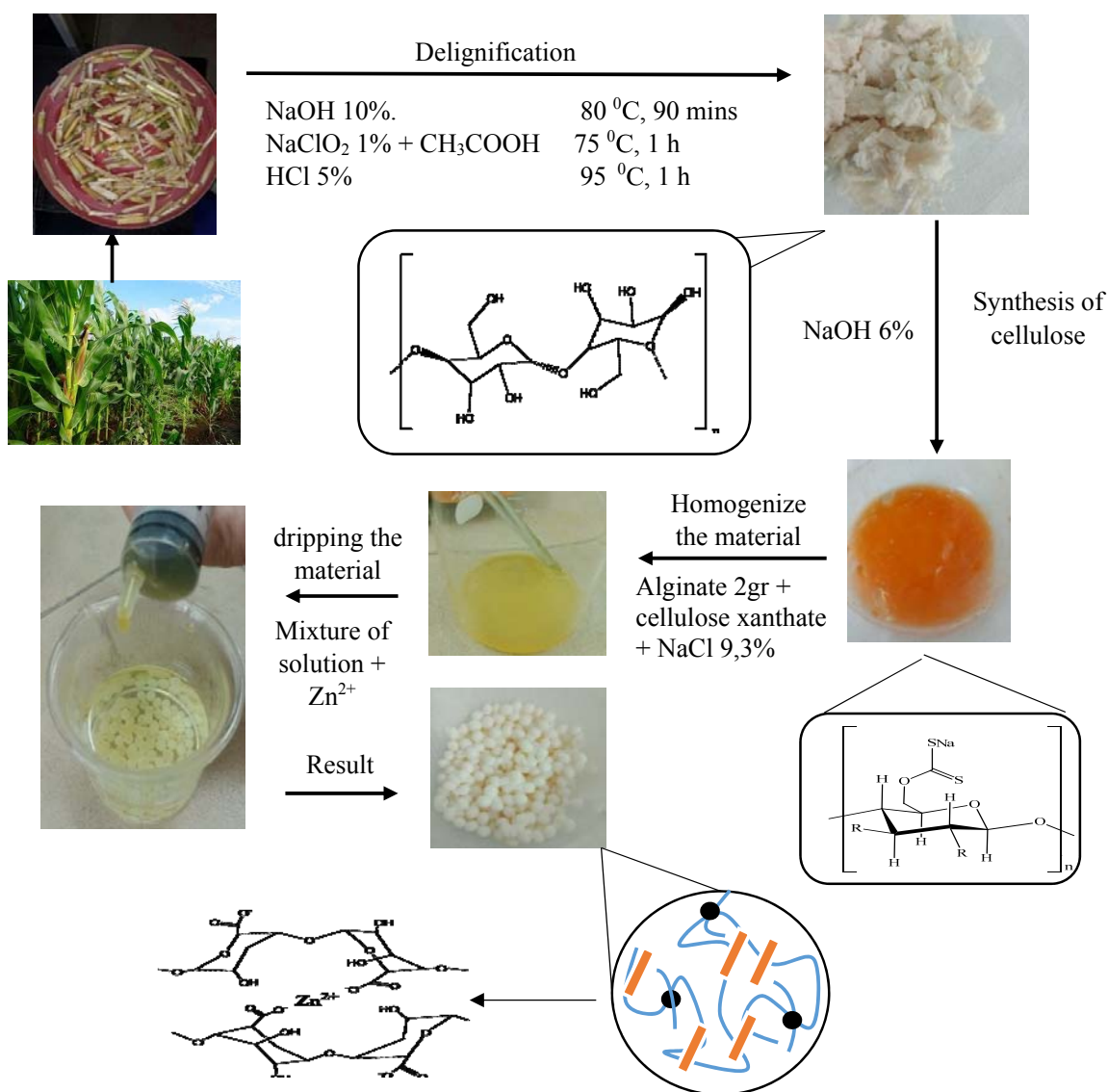


Figure 1. The schematic workflow for alginate-cellulose xanthate beads formation

Lignin Content Analysis

Analysis of lignin content in samples was carried out using Chesson's method (Datta, 1981). One gram of corn stalk powder (a) was added to 150 ml distilled water and heated in a water bath at 100 °C for 2 h. The mixture was filtered, and the residue was washed with hot water. After that, the sample was put in the oven, and weighed (b). The obtained residue was added into 150 ml 0.5 M H₂SO₄ and refluxed for 2 h at 100 °C. The sample was subsequently filtered and washed to neutral pH. Then the residue was put in the oven to achieve constant weight (c). The resulting residue was

soaked with 10 ml 72% H₂SO₄ at room temperature for 4 h. Afterwards, the mixture was added to 150 ml 0.5 M H₂SO₄ and refluxed for 2 h and filtered. Then the mixture was filtered, washed to neutral pH, and put in the oven to obtain its constant weight (d). Finally, the solid was heated until it turned to ash and weighed (e). The calculations using Chesson's method are as follows:

$$\% \text{ cellulose} = \frac{c-d}{a} \times 100\% \quad (1)$$

$$\% \text{ lignin} = \frac{d-e}{a} \times 100\% \quad (2)$$

Porosity and Swelling Analysis

The porosity of the beads was determined by placing the beads in a test tube. Filled with 5 mL of ethanol (V1). Beads were soaked for 24 h. The total volume following beads immersion was recorded (V2). The solvent trapped inside the beads' pores were removed, and the remaining amount of ethanol in the cylinder is indicated by (V3). Porosity (χ) was determined using the equation below (Eiselt *et al.*, 2000) :

$$\chi = \frac{(V1-V3)}{VT} \times 100 = \frac{V1-V3}{V2-V3} \times 100 \quad (3)$$

The swelling of beads was identified by soaking ± 50 mg of beads in 20 ml of aquademin. Beads were weighed after 24 h of immersion. The swelling test is determined based on equation (4).

$$Swelling = \frac{Wt-Wo}{Wo} \times 100\% \quad (4)$$

Where Wt is the weight of beads after soaking, and Wo is the weight before soaking.

Morphology Analysis

SEM-EDX was used to observe the morphology and elements present on the surface of alginate-cellulose xanthate beads.

Size Analysis

Wet, dry, and swollen beads are characterized using an optical microscope. Differences in the size of the diameter of the wet, dry, swollen beads and their pore sizes were obtained by analyzing the captured images using Image-J software (Wu *et al.*, 2017).

Results

Cellulose Content

The extraction of cellulose in corn stalks have been successfully carried out using NaOH. The addition of NaOH to the sample can reduce lignin levels but could not entirely eliminate lignin (Table 1). Test results on microfiber cellulose samples showed that lignin was still present at 19.67%.

Based on FTIR spectra of cellulose extracted from corn stalks (Figure 2a) there were absorptions observed at 3471 cm⁻¹ (OH stretching group), 2929 cm⁻¹ (CH group), 1479 cm⁻¹ (CH₂) 1384 (antisymmetric CO), 1045

cm⁻¹ (COC groups), 870 cm⁻¹ (β -glycoside bonds), 664 cm⁻¹ (stretching CC), and 1641 cm⁻¹ (C=O group) (Zheng and Meng, 2016).

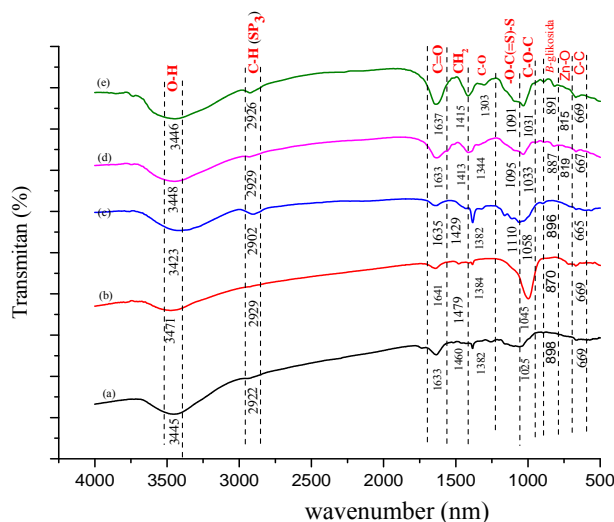


Figure 2. FTIR spectra of (a) corn stalk powder (b) microfiber cellulose (c) cellulose xanthate (d) ACX-0 beads (e) ACX-3 beads

Table 1. Lignin and cellulose content in the sample

Sample	Cellulose (%)	Lignin (%)
Cornstalk powder	8.31	27.72
Microfiber Cellulose	38.43	19.67

The FTIR of microfiber cellulose (Figure 2b) showed a reduction in absorption intensity at wave numbers 3471, 2929, and 1641 cm⁻¹, which showed that the absorption of lignin compounds in the sample was reduced. On the other hand, the absorption at wave number 1045 cm⁻¹ was shown to increase, demonstrating the absorption of C-O-C cellulose groups (Zheng and Meng, 2016). After the addition of NaOH and CS₂ to cellulose, new uptake appeared in 1161 cm⁻¹ and 1110 cm⁻¹ (Figure 2c), which showed the typical uptake of cellulose xanthate (-O-C(=S)-S). Additionally, the wide and high bending vibration absorption of CH₂ (1429 cm⁻¹) confirmed that a substitution reaction had occurred mainly in the C-6 hydroxyl cellulose group (Zheng & Meng, 2016). FTIR spectra of beads (Figure 2d and 2e) showed a new absorption at 819 cm⁻¹ for ACX-0 beads and 815 cm⁻¹ for ACX-3, which was assumed to be an absorption of Zn-O bonds.

Shape, Diameter, and Weight of Swollen Beads and Their Porosity

Diameter measurement of wet and dry beads was performed using ImageJ software can be seen in Figure 3, whereas the shapes of dry beads are shown in Figure 4. Figure 4 shows that ACX-0 beads are flat compared to the other beads, which tend to be more rounded. The average diameter of wet beads obtained was 3.32 mm. After the beads are dried, the diameter of beads decreased up to 159.21% (Table 2).

Table 2. Wet and dry beads diameter

Beads	Wet AVE (mm) ± S.D	Dry AVE (mm) ± S.D
ACX-0	3.36 ± 0.26	1.67 ± 0.04
ACX-1	3.25 ± 0.02	1.28 ± 0.04
ACX-2	3.42 ± 0.02	1.14 ± 0.03
ACX-3	3.24 ± 0.17	1.15 ± 0.01

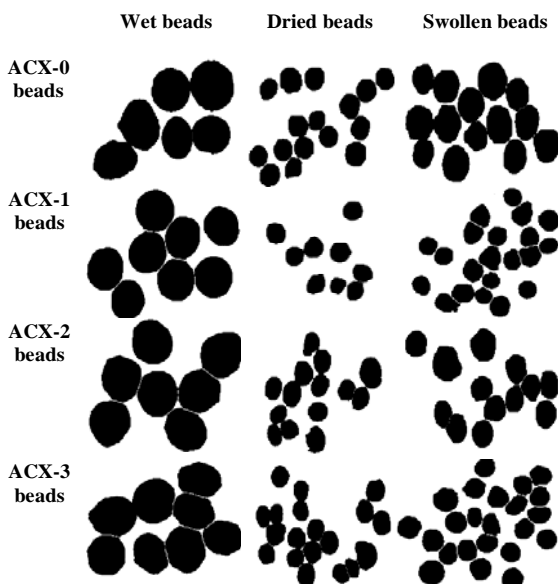


Figure 3. Image analysis of wet, dried, and swollen beads (in water for 24 h) beads using ImageJ

The addition of a porogen to the beads affected the swelling properties and diameter of the beads. Figure 5 shows a graph of changes in the diameter and weight of swollen beads after soaking with distilled water for 24 h. Swelling properties of beads ranged from 35.66 - 75.73%, while the changes in diameter of the beads ranged from 6.2 - 32.5%. Swelling properties and diameter increased with increasing porogen

concentration. The addition of porogen to the beads also affected the porosity value. The porosity test results obtained (Figure 6) showed that beads porosity increased with increased concentrations of NaCl. ACX-2 and ACX-3 beads had a higher value than ACX-0 beads. In comparison, thereby ACX-1 beads had the lowest porosity of 55.56%.

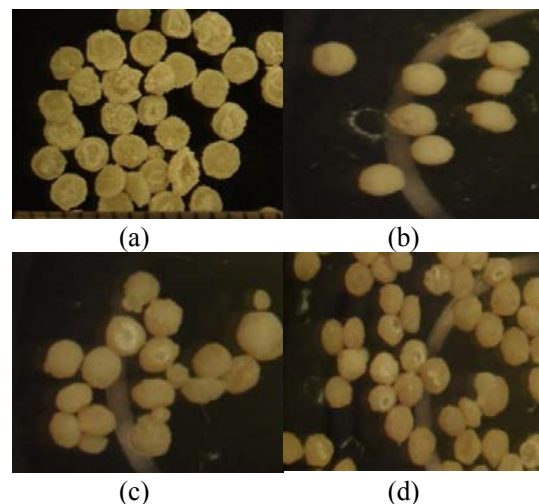


Figure 4. Characterization of optical microscope dry beads (a) ACX-0 (b) ACX-1 (c) ACX-2 (d) ACX-3

Surface Morphology

Figure 7 shows the SEM images of ACX-0 beads (Figure 7a and 7b) and ACX-3 (Figure 7c and 7d). ACX-0 beads had a rough and porous surface with an average diameter of $6.7 \times 10^{-2} \pm 0.01 \mu\text{m}$, while the ACX-3 beads had a surface that forms rough folds and several gaps with a diameter of $20.79 \times 10^{-2} \pm 0.03 \mu\text{m}$.

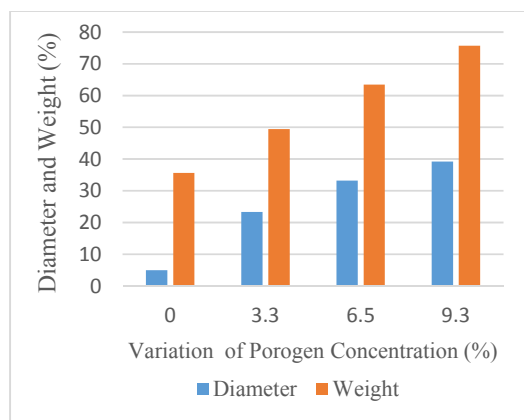


Figure 5. Diameter and weight of swollen beads with different concentrations of porogen

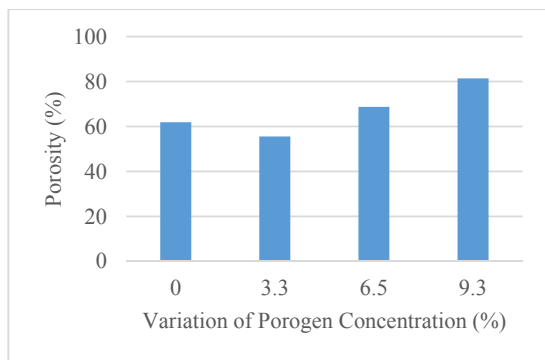


Figure 6. Beads porosity with different concentrations of porogen

Surface Compound

Based on elemental analysis using EDX as depicted in Figure 8, the following elements were detected on the surface of the beads ACX-0: C, O, H, S, Zn (Figure 8a) while ACX-3 beads had a Na content of 7.05% and Cl of 0.05% (Figure 8b).

Discussion

There are two principles of this research. First, the basic ingredients of alginate and cellulose xanthate from cellulose derivatives extracted from corn stalks are the basic ingredients for making beads. The manufacturing technique used was the dropping method. Extraction results showed that lignin was still present in cellulose microfiber because NaOH used to remove lignin only reacted with ester bonds connecting lignin with hemicellulose in the lignin-carbohydrate (Modenbach, 2013).

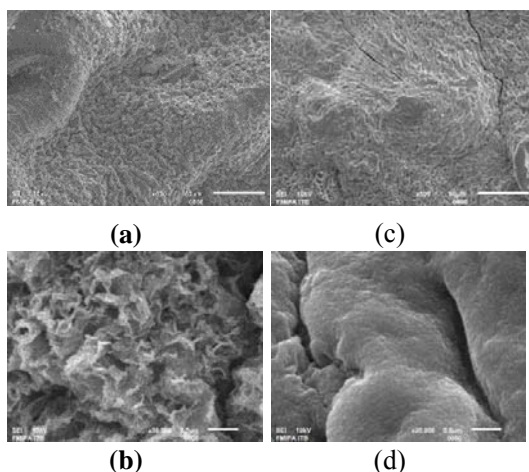


Figure 7. SEM images of ACX-3 beads (a) 500x magnification, (b) 30000x, and ACX-0 beads (c) 500x magnification, (d) 30000x

Making beads using dropping techniques resulted in a variety of bead sizes (Table 2). The advantage of dropping is that it is easy and inexpensive. However, to produce more uniform beads, it is necessary to consider the injection speed range and the drop distance (Gericke *et al.*, 2012).

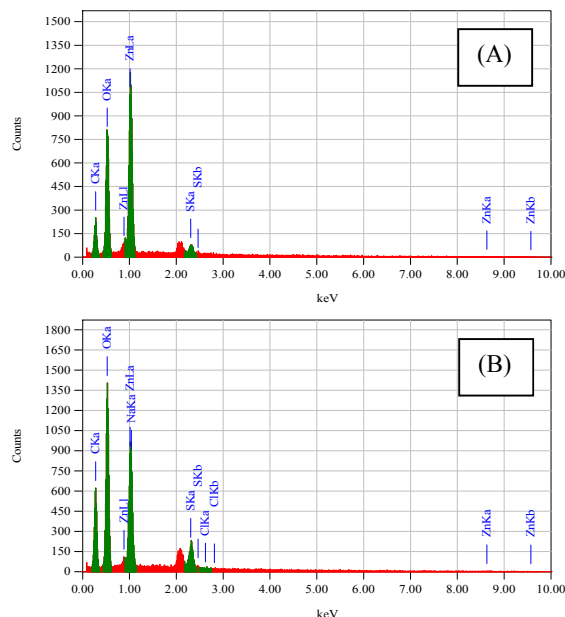


Figure 8. EDX spectra of (A) ACX-0 and (B) ACX-3 beads

Secondly, the feasibility of using NaCl as a pore-forming agent was assessed in this study. Compared to previous research (Sönmez *et al.*, 2016), with the same material alginate and cellulose as a material for making beads, the addition of NaCl as porogen could create a rougher and more hollow surface. Increased levels of NaCl in turn increased the ability of cellulose beads to bind water, which is marked by an increase in swelling and diameter of the beads. This is supported as well by porosity data, which demonstrated increased porosity with increasing levels of the porogen. The presence of porogen caused the space inside the beads to increase thereby increasing the absorption ability of the beads. Beads ACX-1 had a lower porosity than ACX-0. According to Yu *et al.* (2008), this could be due to the low concentration of NaCl porogen, resulting in beads with closed pores and the tendency to be randomly shaped.

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