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DEGRADATION OF BIOPLASTIC WASTE WITH AMMONIA-NITROGEN REDUCTION IN LANDFILL LEACHATE MEDIUM

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Abstract: Plastics made from fossil fuels lead to the deterioration of biodiversity and the environment. Small particles of plastic wastes, known as microplastics have been detected in the food chain of marine organisms and have contaminated deep-sea sediments. Research and development have been carried out to solve this problem through the invention of bioplastics as a replacement of petroleum-based plastics. Bioplastics can degrade faster than plastics made from fossil fuels, which require a long time to decompose. Bioplastics are mostly made from biomass and are safe for the environment. At the end of the life cycle of bioplastics, they are typically dumped at landfills as the ultimate disposal and they will go through the degradation process there. Thus, this study aims to investigate the degradation process of bioplastics, particularly corn starch-based bioplastics without fillers (BP) and corn starch-based bioplastics with chitosan (BPC) in landfills leachate, which mimics the on-site process. The characteristics of bioplastics before and after the experiments were evaluated. The small bioplastics (BP and BPC) particles were treated in landfill leachate in aerobic conditions. The experiment was conducted using batch culture with leachate medium collected from the Air Hitam Sanitary Landfill in Puchong, Selangor. The degradation of bioplastics (BP and BPC) was observed using Fourier transform infrared spectroscopy (FTIR) and scanning electron microscope for the morphology before and after the incubation period of 14 days. The FTIR spectra of both types of bioplastics before the batch culture experiment displayed the presence of O-H, C-H and C=O absorption peaks. However, after 14 days of incubation, the C-H group disappeared from both types of bioplastics towards the end of the experiment. It has been observed that the weight reduction of corn starch-based BP was 92.2% and corn starch-based BPC was 86.7% after 14 days. The ammonia-nitrogen reduction was 40.78% for BP and 38.03% for BPC. It can be concluded that BP degrades better than BPC.

Keywords: Bioplastics, biodegradation, batch culture, landfill leachate medium.

Introduction

The demand for plastic in recent years has continued to increase due to its various uses and availability, as well as it being light, durable and low cost. However, most of the plastics in the market come from synthetic polymer and non-bio-degradable materials that cause pollution to the earth. According to the Statistics of Plastic Usage, shoppers worldwide use approximately 500 billion plastic bags per year (Bejgarn *et al.,* 2015). These single-use plastics were predominantly made from non-renewable petroleum and natural gas (Kasmuri & Zait, 2018).

Improper plastic waste management and human behaviours are responsible for the pollution of the ocean, affecting marine organisms and human beings. Plastics can be broken down into different sizes, known as microplastics (MP, <5 mm) and macroplastics (MaP, 5–150 mm), by photochemical reaction (Yuan *et al.,* 2020; Zhu *et al.,* 2020). These persistent particles take a very long time to degrade and pose a potential threat to aquatic ecosystems (Gallo *et al.,* 2018). It was found that microplastics and macroplastics are taken up by different members of the food web, like lugworms, mussels and birds (Huerta Lwanga *et al.,* 2017). Large plastic items such as toothbrushes, plastic bottles, straw and bags have been transformed into microplastics and entered the food chain through marine organisms. Thus, human food consumption has been highly affected by toxic chemicals coming from the fragmentation of plastic waste (Gallo *et al.,* 2018). The mixture of polymers from plastic substances has been considered one of the vectors of contaminant and is lethal to the ecosystem.

Nowadays, most solid waste management facilities tend to handle waste through incineration. This incineration of waste, including plastic materials, normally releases poisonous gases, such as dioxins, furans, mercury and polychlorinated biphenyls, into the air (Chantara *et al.,* 2019; Cheng *et al.,* 2020). Moreover, these toxic substances are very harmful to the ecosystem and the surrounding environment (Chantara *et al.,* 2019).

Other than improper management of plastic waste, another issue is environmental problems. Non-recyclable food can be considered one of the main contributors to Malaysia's municipal solid waste (MSW) (Samad *et al.,* 2017). According to Yan and Chen (2015), food waste contributes a major part, about 64% of the total municipal solid waste produced in Malaysia. The high volume of solid waste is due to the country's expanding population, urbanisation and the huge amount of food consumption of Malaysians. Malaysians consumed approximately 61,000 tonnes of crustaceans in 2014, of which 60% became food waste (Yan & Chen, 2015). If collected and separated, this food waste could be an alternative source of bioplastics. Thus, it will reduce the amount of garbage in landfills.

Reducing food waste can be achieved by replacing conventional plastics with bioplastics made from organic wastes (such as food leftovers). The conversion of these waste materials can significantly lessen the amount of garbage in landfills and limit its impact on the environment. Therefore, it aligns with the United Nations' Sustainable Development Goals (SDGs). The target indicator for SDG 12 for sustainable consumption and production can be achieved by reducing the food losses in this supply chain (Esparza *et al.,* 2020), along with SDG 14 to conserve and sustainably use the oceans, seas and marine resources (Shruti & Kutralam-Muniasamy, 2019). At the same time, using organic wastes from marine resources as materials for bioplastics can significantly reduce the effect of pollution on the ecosystem and preserve the environment, particularly the marine ecology.

Moreover, bioplastics are easily biodegradable, which is sustainable and safer for the ecosystem. However, the best management practice in handling bioplastics needs to be studied extensively. The life-cycle assessment of these packaging applications and the end-of-life option of bioplastic materials have been reviewed recently (Thakur *et al.,* 2018; Dilkes-Hoffman *et al.,* 2019). It can be observed that most of these bioplastics will end up in landfills after usage (approximately 93% in landfills and oceans) (Mehdi Emadian *et al.,* 2017). In landfills, the organic components of these bioplastics will break down due to the biodegradation process, with studies showing that in composted soil, they have a biodegradability rate of nearly 90% (Mehdi Emadian *et al.*, 2017).

In addition, the organic materials from the bioplastics will be dissolved in the landfill's leachate. This nitrogen-containing nutrient is an oxygen-consuming compound and can cause the depletion of dissolved oxygen in water. It can be presumed that there is a large variety of organic and inorganic substances with a high concentration of contaminants, such as ammonia-nitrogen and heavy metals, in landfill leachate (Chang *et al.,* 2018; Kasmuri & Tarmizi, 2018). This high number of pollutants, particularly ammonia-nitrogen in water bodies, leads to the declination of fish and many other aquatic organisms in the ecosystem (Sprovieri *et al.,* 2020).

Hence, this study focuses on the consumption rate of a group of bacteria in treating ammonia-nitrogen and the simultaneous degradation of bioplastics in a landfill leachate medium. The organic substance of corn starch-based bioplastics without filler (BP) and corn starch-based bioplastic with chitosan (BPC) have been used as substrate or the source of food, for the bacteria in the batch culture experiment.

Materials and Methods

Preparation of Bioplastics

The bioplastic materials included sodium hypochlorite, glycerol and acetic acid (Merck KGaA, Darmstadt, Germany); chitosan from shrimp shells as the filler (Sigma Aldrich, Darmstadt, Germany) and corn starch (Shah Alam, Selangor, Malaysia). Deionised water (Merck Millipore) was obtained from the Environmental Laboratory, School of Civil Engineering, College of Engineering, Universiti Teknologi MARA, Shah Alam, Selangor, Malaysia. The proportion of materials used to develop corn starch-based bioplastics without filler (BP) and corn starch-based bioplastics with chitosan (BPC) is shown in Table 1.

An amount of 60 mL of deionised water was measured and poured into the beaker. Approximately 9.5 g of corn starch was measured and placed in the beaker with the deionised water. The mixture was stirred and mixed using a glass rod. After that, 5 mL of glycerol and 5 ml of acetic acid were measured and poured into the same beaker. The mixture was placed on a hot plate (50˚C) and stirred until it became sticky and almost transparent. Finally, the mixture was spread on an aluminium foil and was left for a week to cool at room temperature (Kasmuri & Zait, 2018).

Preparation of the Nitrification Medium

A synthetic nitrification medium was prepared for the batch culture experiment (Kasmuri & Lovitt, 2018). This synthetic medium shown in Table 2 was dissolved in 1 L of deionised water together with the ammonium sulphate $(NH_4)_2SO_4$ stock solution, which was prepared by mixing 5 g of $(NH_4)_2SO_4$ with 1 L of deionised water to make the final concentration of 0.5 g/L, separately. For experimental purposes, this stock solution was later diluted to make the 85 mg/L of ammonium sulphate concentration. Both synthetic medium and the stock solution of $(NH_4)_2SO_4$ were autoclaved separately at 121°C for 15 minutes (Kasmuri & Lovitt, 2018). This combination of synthetic medium and ammonium sulphate solution was done for the preparation of the nitrification medium.

Table 1: List of materials and proportions used to form bioplastics (Kasmuri & Zait, 2018)

Materials	Proportion	
	Corn Starch-based Bioplastics without Filler (BP)	**Corn Starch-based Bioplastics with Chitosan (BPC)**
Deionized water	60 mL	60 mL
Corn-starch	9.5 g	9.5 g
Glycerol	5 mL	5 mL
Acetic acid	5 mL	5 mL
Chitosan	0 g	2.5 g
Length	130 mm	150 mm
Width	110 mm	100 mm

Table 2: Synthetic Medium
(Kasmuri & Lovitt, 2018)

Formulation	Weight (g)
Na_2HPO_4	13.5
KH_2P_O4	0.7
$NaHCO_3$	0.5
$MgSO_4.7H_2O$	0.1
$FeCl_3.6H_2O$	0.014
$CaCl_2.2H_2O$	0.18

Batch Experiment Procedures

Three sets of flask were prepared, of which each had a total of 100 mL of synthetic medium (refer to Table 2). Another 100 mL of ammonia-nitrogen containing approximately 85 mg/L of the diluted stock solution of ammonium sulphate $(NH_4)_2SO_4$ was added into the three sets of flasks. Each set of flask consists of two types of bioplastics, BP and BPC, by which two flasks each contained 0.9 g of BP and another two contained the same amount of 0.9 g of BPC, separately. The bioplastic specimens were cut into a microplastic size (<5 mm). Then, a pre-weighed amount of 0.9 g bioplastic of each BP and BPC was placed in the three sets of flasks, respectively.

Pre-treated leachate from the Air Hitam Sanitary Landfill, Puchong was used as the inoculum in the batch culture experiment. Consequently, the measuring cylinder was used to inoculate 5 mL of pre-treated leachate (as the inoculum for the source of bacteria) in two flasks of BP and BPC. Meanwhile, another two flasks of BP and BPC were not inoculated with pre-treated leachate and were used as the controls for the experiment. The control flasks were employed to determine if there would be any ammonia-nitrogen concentration reduction without bacteria inoculation from the pre-treated leachate sample. The schematic diagram of the batch experiment for one set of flasks is shown in Figure 1.

Then, the conical flasks were capped with glass wool to prevent spillage of the medium. The flasks were then placed in a mechanical shaker at 180 rpm and 28°C under aeration with a dissolved oxygen rate of above 2 mg/L (Kasmuri & Lovitt, 2018). The samples were measured daily to determine the concentrations of ammonia-nitrogen, nitrite-nitrogen and nitrate-nitrogen based on APHA (2005). The BP and BPC degradations were observed before and after the incubation period of the batch culture experiments. The incubation period

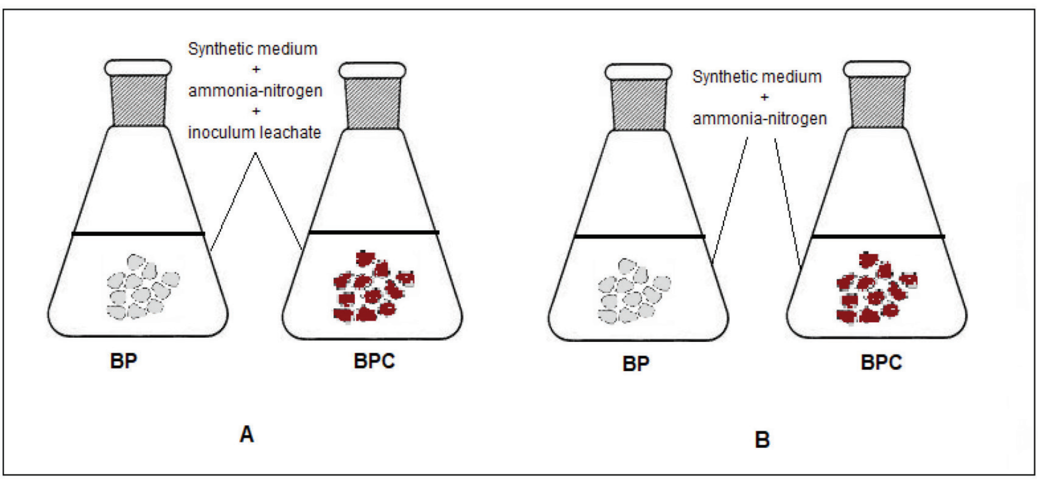

Figure 1: Schematic diagram of the bioplastics in the batch culture experiment, (A) Flasks with the inoculum of leachate, (B) Flasks without the inoculum of leachate (control)

was observed for 14 days. After 14 days in the incubator, the mechanical stirrer was used to take the samples out. The bioplastics particles were separated from the solution using the centrifuge machine, which was set to 4000 rpm for 15 minutes (Johnsson & Steuer, 2018). After the centrifugation process, the wet weight of the BP and BPC was determined. After that, the test tubes with the samples were dried in a hot-air oven at 50°C overnight to obtain the dry weight of the samples (APHA, 2015). Later, the residuals of the BP and BPC weight were determined by measuring the percentage reduction using the Sartorius ENTRIS 224-1S analytical balance with a readability of 0.0001 g (Mohan *et al.*, 2016). The weight loss of the bioplastics in percentage was determined using Equation 1 shown below:

$$\%\ \text{weight loss} = \frac{(Wo - W)}{Wo} x\ 100 \quad \textbf{(Equation 1)}$$

where Wo is the initial weight of the bioplastics (g) and W is the residual weight of the bioplastics (g).

Observation from Scanning Electron Microscope (SEM)

The surface morphology of the BP and BPC was observed using a scanning electron microscope (SEM). The SEM was integrated with a personal computer inside the Polymer Composite Research Laboratory (PoCRe), Institute of Science, Universiti Teknologi MARA. Prior to and after the experiment, small BP and BPC bioplastics samples were taken and coated with Au for 60 seconds to enhance the sample conductivity before the SEM observation (Li *et al.,* 2020).

Observation from Fourier Transform Infrared Spectroscopy (FTIR)

Four samples of the BP and BPC were analysed in this study. The functional groups were analysed at the Instrument Lab II of the School of Chemical Engineering (UiTM, Shah Alam). It was done using PerkinElmer® Fourier transform infrared spectroscopy (FTIR). FTIR was used to record the changes in the functional groups of the BP and BPC before and after 14 days of the batch experiment. The samples of BP and BPC were dried, thoroughly grounded and pressed into tablets for FTIR observation (Li *et al.,* 2020).

Results and Discussion

Results from Scanning Electron Microscope (SEM)

The photo-fragmentation of the surface of the bioplastics before and after the batch culture experiment was taken using SEM to investigate the changes of the bioplastics. The surface morphology of the two types of bioplastics, BC and BPC was observed. On Day 0, under the characterisation by SEM, the BP shows few irregular shapes surrounded by several dark spots. However, the overall surface of BP was relatively smooth, with flat elevation (refer to Figure 2 (a)).

In Figure 2 (b), after 14 days of the experiment, the photo-fragmentation from the SEM shows white flakes on the far right of the surface of the BP. The white flakes are also scattered in the middle of the specimen but are not too noticeable. These white spots may come from the pre-treated leachate. It was observed that after 14 days of the experiment, the BP was fully decomposed. Larger particles were broken down into smaller particles. Hence, the visible pores between the particles are clearly shown in the observation by SEM. The BP's morphology has a rough surface structure and irregular texture. More extensive cracks with fractures and ripple structures were deepened and can be seen in the photo-fragmentation in Figure 2 (b), as compared with Figure 2 (a). Thus, this surface morphology indicates the degradation process of the BP, which occurred after the 14 days of the experiment.

As shown in Figure 2 (c), under the characterisation by SEM, the BPC particles were seen to be sticking together. The substances from corn starch and chitosan can be described as being combined and homogenised in the BPC. Here, their shapes can be seen to be well-defined as separation lines between the particles are visible with uniform textures on the bioplastic surface. It can be seen that the overall surface texture of the BPC was less smooth compared with the BP. These microspheres exist on the overall surface of the BPC and were in contrast with Figure 2 (a) of the BP. Based on Figure 2 (c), a few pores can be observed in the BPC sample before the batch experiment. However, after 14 days of the batch experiment, the surface of the BPC has dramatically changed, which is shown in Figure 2 (d), with irregular forms of particles separated with non-uniform textures. The photo-fragmentation under the SEM of the components of BPC showed that it may have started to decompose and form clumps due to the degradation process after 14 days of the batch experiment. The hollow and unevenly distributed clumps formed were due to the decomposed particles not sticking together. There were more visible porous pores in the BPC sample after 14 days of the degradation process. There is a significant different surface texture that can be seen between Figure 2 (c) and Figure 2 (d) of the BPC before and after the batch experiment.

Bajer *et al.* (2020) in their study examined bio-packaging materials made from bioplastics derived from starch/chitosan/aloe vera using atomic force microscopy (AFM). The micrographs of the AFM on the effects of aloe vera on a chitosan/starch blend showed that the samples' morphology was very rough and had numerous recesses that resemble steep hills. In addition, from the study of the ultra-violet (UV) irradiation effect on the surface topography of the bioplastics with aloe vera, no visible changes were detected in the morphology of the samples (Bajer *et al.,* 2020). However, this finding is in contrast with the present study as significant changes were observed in both samples of BP and BPC in the batch culture experiment. However, the study by Bajer *et al.* (2020) evaluated the influence of

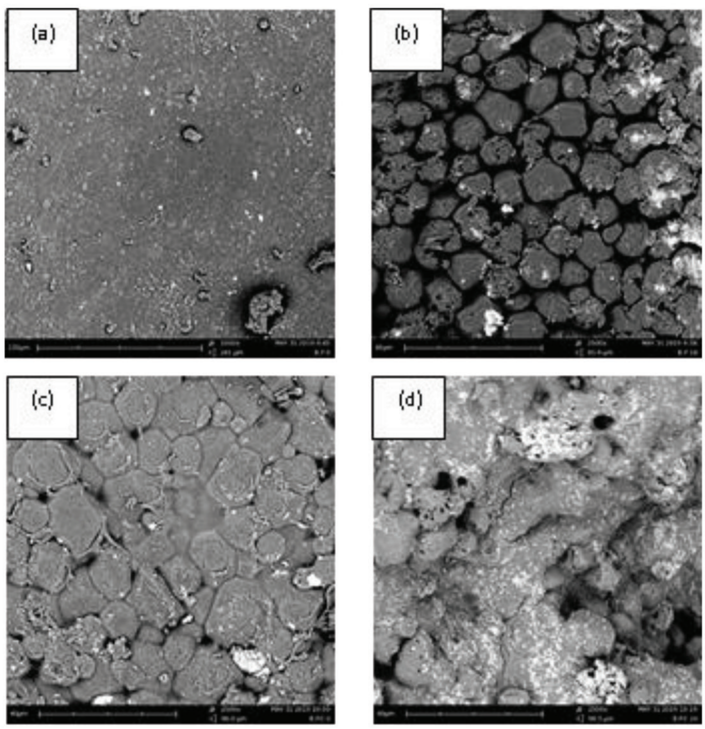

Figure 2: The SEM micrographs of the bioplastics, (a) BP day 0, (b) BP day 14, (c) BPC day 0 and (d) BPC day 14

UV irradiation on starch/chitosan/aloe vera bioplastics, which is different with this paper, which examines the effect of leachate inoculum on the biodegradation of BP and BPC through a batch culture experiment.

High-generation plastics would ultimately increase the volume of leachate produced in landfills and release greenhouse gases into the environment (Mehdi Emadian *et al.,* 2017). Therefore, it has been observed that the leachate inoculum can be considered as a novelty for this study. According to Mehdi Emadian *et al.* (2017), almost all plastic waste is disposed in landfills. However, the biodegradation of bioplastics in landfills has not been extensively studied. Thus, the biodegradation of bioplastics in compost, soil and aquatic environments need to be thoroughly examined and discussed (Mehdi Emadian *et al.,* 2017). Furthermore, best management practices on handling solid waste made up of plastics need to be considered to reduce the impact of plastic disposal in landfills.

Results of the Ammonia-nitrogen Concentration

This study focused on the simple nitrification process using inoculum from leachate samples to reduce ammonia-nitrogen in the BP and BPC batch culture experiment. Subsequently, high ammonia-nitrogen concentrations in landfill leachate and other pollutants can be found. Thus, effective treatment is needed to reduce ammonia-nitrogen from landfill leachate (Sprovieri *et al.,* 2020).

Figure 3 shows the reduction of ammonia-nitrogen in the batch culture experiment with an initial input concentration of 85 mg/L. It can be observed that the ammonia-nitrogen concentrations in both BP and BPC gradually decreased in 14 days of the batch experiment. As time increased, the ammonia-nitrogen concentration showed a decreasing pattern due to the nitrification process that helped remove ammonia-nitrogen (Kasmuri & Lovitt, 2018). The consumption of nitrifying bacteria from the leachate inoculum can explain the depletion of ammonia-nitrogen as their substrate. This is supported by Kasmuri and Lovitt (2018), who showed that the ammonia-nitrogen oxidation process occurred in batch cultures from soil and fishpond.

Consequently, it can be seen that the concentration of ammonia-nitrogen in the flask with the BP had a higher reduction level than the concentration of ammonia-nitrogen in the flask with the BPC on Day 1 as shown in Figure 3. From day 1 until day 7, the BPC experienced a few drops in ammonia-nitrogen concentration in stages in the batch experiment. The flask with the BP, meanwhile, saw the ammonia-nitrogen concentration experiencing a smooth decline for

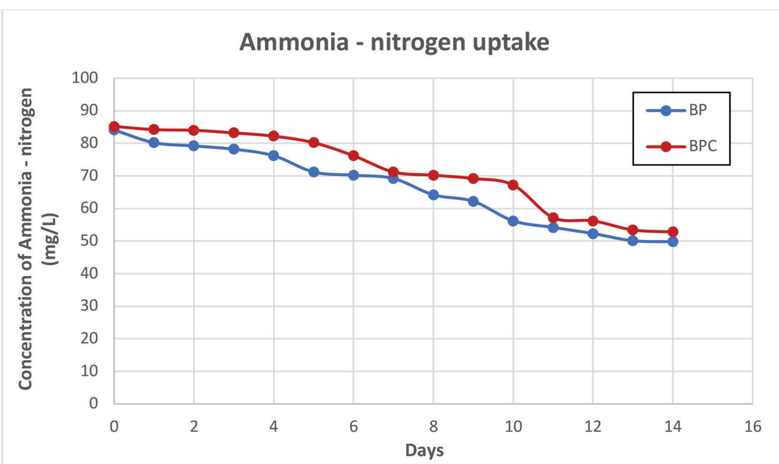

Figure 3: Ammonia-nitrogen uptake in batch culture experiment

the same period of the batch culture experiment. The next 7 days showed that the ammonia-nitrogen concentration in the BP had the same pattern of decrease throughout the experiment. The same goes for the flasks with the BPC, in which the ammonia-nitrogen concentration reduced concurrently throughout the experiment for the final 7 days.

In the 14 days of batch culture experiment, the percentages of ammonia-nitrogen concentration reduction for flasks with the BP and BPC were 40.78% and 38.03%, respectively. The higher percentage of ammonia-nitrogen removal in the flask with the BP showed that the ammonia-nitrogen oxidation process by the nitrifying bacteria accelerated faster than the flask with the BPC. This percentage difference of 2.76% may show that these bacteria can consume the substrate in the flask with the BP more quickly than the BPC bioplastic. The reduction of ammonia-nitrogen and the co-metabolism of the bioplastics in the batch culture experiment confirms the findings of Fernandez-Fontaina *et al.* (2014) and Martínez-Jardines *et al.* (2018). This biotransformation of organic micropollutants in the batch culture experiment was regarded as a co-metabolism process. Here, the bacteria would degrade the non-growth substrates (in this case, the BP and BPC) in the presence of the primary substrate (ammonia-nitrogen).

Hwang *et al.* (2020) stated that food wastes involving starch contain plenty of sugars and carbohydrates. Here, in this batch culture experiment, the BP contains only corn starch, while the BPC contains corn starch and chitosan. The addition of chitosan as the solid filler might have hindered the microbial activity in the BPC. This is supported by Vernaez *et al.* (2019) in their study on the poly (lactic acid) (PLA)-chitosan bioplastic. They stated that the additive of chitosan leads to hydrophilic behaviour due to its hydroxyl and amino groups from chitin (Vernaez *et al.,* 2019). From the results, it can be hypothesised that the bonds between the corn starch and chitosan have proportionately slowed down the consumption of ammonia-nitrogen oxidation and simultaneously reduced the process of co-metabolising the second substrate, which is the BPC.

According to Kasmuri and Lovitt (2018), a long incubation time is needed for the nitrifying bacteria to consume a high concentration of ammonia-nitrogen. This long incubation time is required since the nitrifying bacteria is a slow-growing bacterium (Thakur & Medhi, 2019). It can be estimated that the ammonia-nitrogen will be removed entirely if a longer incubation time is provided for the batch experiment.

Results of Nitrite-nitrogen and Nitrate-nitrogen Concentration

Figure 4 shows the increased concentration of nitrite-nitrogen (NO_2-N) and nitrate-nitrogen (NO_3-N) in the two sets of flasks with the BP and BPC throughout the batch culture experiment. All these flasks had an initial concentration of 85 mg/L of ammonia-nitrogen. The left axis on Figure 4 represents the concentration of NO_3-N, while the right represents the concentration of NO_2-N. Figure 4 illustrates the trends of nitrite-nitrogen and nitrate-nitrogen concentrations in 14 days. At the beginning of the experiment, a limited amount of nitrite-nitrogen and nitrate-nitrogen existed caused by trace concentrations (NO_2-N and NO_3-N) in the inoculum sample from the pre-treated leachate. Then, after some time, more concentrations of nitrite-nitrogen were accumulated in flasks with both the BP and BPC. This increased nitrite-nitrogen concentration could happen as the nitrifying bacteria has utilised the ammonia-nitrogen in the first step of the nitrification process to nitrite-nitrogen. Then, in the second stage, the bacteria consumed the nitrite-nitrogen and the production of nitrate-nitrogen was established (Thakur & Medhi, 2019). This complete cycle of the nitrification process is noticeable from the results shown in Figure 4.

In Figure 4, the initial concentration of nitrite-nitrogen for the BPC is slightly higher than the BP. From day 0 to day 3, it is clearly shown that the nitrite-nitrogen in both the BP and BPC had a minor increase in concentration.

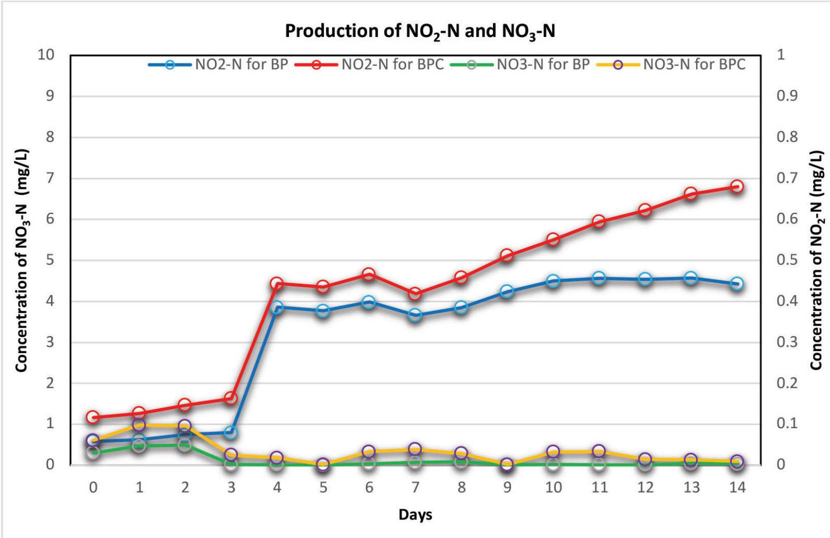

Figure 4: The production of nitrite-nitrogen and nitrate-nitrogen in batch culture experiment

However, on day 3 up till day 4, in both the BP and BPC flasks, the nitrite-nitrogen concentration saw a rapid climb and fluctuated until day 7. This graph from Figure 4 showed a similar trend in the development of nitrite-nitrogen (Kasmuri & Lovitt, 2018). A study conducted by Kasmuri and Lovitt (2018) also showed a rapid climb of nitrite-nitrogen after a certain period.

The nitrite-nitrogen in the BPC flask continued to increase throughout the experiment. The nitrite-nitrogen concentration in both flasks with the BP and BPC increased until day 14. As Kasmuri and Lovitt (2018) stated, it is expected that the concentration of nitrite-nitrogen to increase slowly and start to decrease as the nitrate-nitrogen increases. However, only the BP reached the maximum amount of nitrite-nitrogen. Hence, it decreased after day 13, while the BPC did not show the same pattern. The nitrifying bacteria accumulated nitrite-nitrogen in the experiment as nitrification occurs in the experiment.

Nitrite-nitrogen turning into nitrate-nitrogen only takes place when there is enough nitrite-nitrogen concentration in the sample. There was an insufficient concentration of nitrite-nitrogen in the both BP and BPC flasks for nitrate-nitrogen to be reflected in the graph significantly. Overall, the nitrate-nitrogen concentration experienced a moderate fluctuating trend throughout the 14 days and had a peak value at 0.05 mg/L during day 2. It shows that 14 days were not enough for nitrite-nitrogen to be oxidised into the nitrate-nitrogen form. If we extend the experiment period, the value of nitrate-nitrogen will start to increase, while the value of nitrite-nitrogen will decrease.

The batch culture experiment maintained a dissolved oxygen concentration of above 2 mg/L to provide an aerobic condition for the nitrifying bacteria to oxidise ammonia-nitrogen to nitrite-nitrogen. The nitrifying bacteria are obligate aerobic autotrophs of ammonia-oxidising bacteria, such as *Nitrosomonas, Nitrosococcus* and *Nitrosopira*, which are involved in converting NH_3-N to NO_2-N. Several important parameters need to be considered for a complete nitrification process (Thakur & Medhi, 2019). The nitrite-oxidising bacteria (NOB), for example, *Nitrobacter* would reduce NO_2-N to NO_3-N (Thakur & Medhi, 2019). Other parameters such as carbon source, the C/N ratio, temperature and pH are also important in achieving the full nitrification process (Thakur & Medhi, 2019).

Results for Percentage Reduction of Bioplastics

Table 3 shows the percentage reduction after 14 days for the BP and BPC. The BP has a higher percentage of weight reduction compared with the BPC. The mass percentage reductions for the BP and BPC were 92.2% and 86.7%, respectively (refer to Table 3). It can be concluded that BP degrades faster than BPC in the span of 14 days. However, it can be estimated that both types of bioplastics can be degraded entirely within one month.

It can be observed that the composition and structure of the bioplastics play an important role in the biodegradation process (Mehdi Emadian *et al.,* 2017). Based on this study, the addition of chitosan has affected the biodegradation process of the BPC in the batch experiment. Thus, a high soluble sugar content in a bioplastic's composition as a carbon source increased the bioplastics biodegradability (Mehdi Emadian *et al.,* 2017; Hwang *et al.,* 2020).

Results from Fourier Transform Infrared Spectroscopy (FTIR)

The characteristics of the chemical structure and molecular interaction in the blends of the BP and BPC were determined using FTIR. The spectroscopy of the FTIR spectrum was done to investigate the changes induced by the biodegradation process in the batch culture experiment. The main characteristics of the absorption bands appear in the range that is adequate for all the mixture components of the BP and BPC.

Figure 5 shows the graph combination before and after FTIR for the BP while Figure 6 shows before and after FTIR for the BPC. The red lines on the graph represent the FTIR results before the experiment and the blue lines are the results of FTIR after the experiment. The results of the FTIR absorption peak for both types of bioplastics and their functional groups were summarised in Table 4. The FTIR readings indicate the difference in the chemical

Table 3: Percentage reduction of bioplastic after 14 days

Corn Starch-based Bioplastics (BP)				**Corn Starch-based Bioplastics with Chitosan (BPC)**			
Test Tube	**Test Tube Weight (g)**	**Oven-Dried Sample + Test Tube (g)**	**Sample Weight (g)**	**Test Tube**	**Test Tube Weight (g)**	**Oven-Dried Sample + Test Tube (g)**	**Sample Weight (g)**
BP 1	13.61	13.68	0.08	**BPC 1**	13.59	13.64	0.05
BP 2	13.53	13.60	0.07	**BPC 2**	13.64	13.72	0.08
BP 3	13.40	13.46	0.07	**BPC 3**	13.50	13.74	0.24
Total			0.22	**Total**			0.37
Average			0.07	**Average**			0.12
Initial BP in each flask			0.9	**Initial BPC in each flask**			0.9
Percentage removal (%)			92.2				86.7

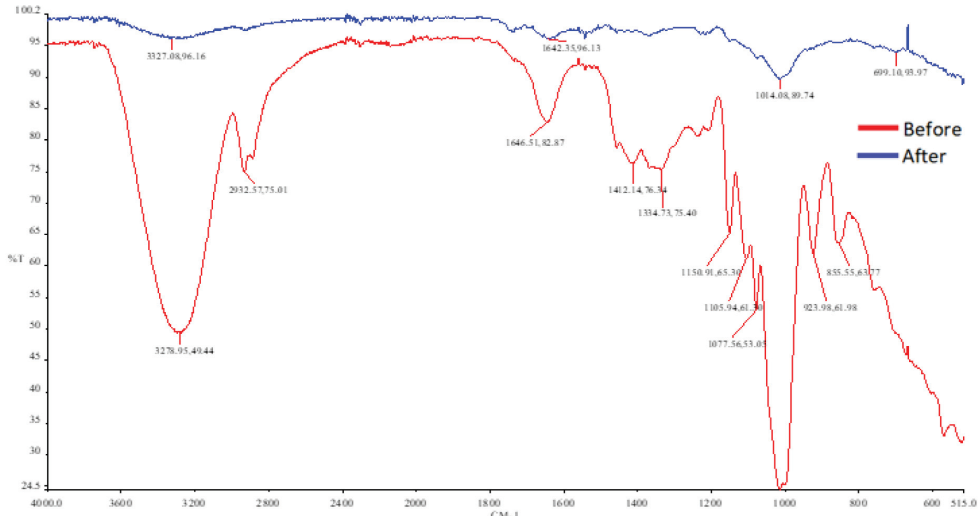

Figure 5: The FTIR of the BP bioplastic before and after the experiment

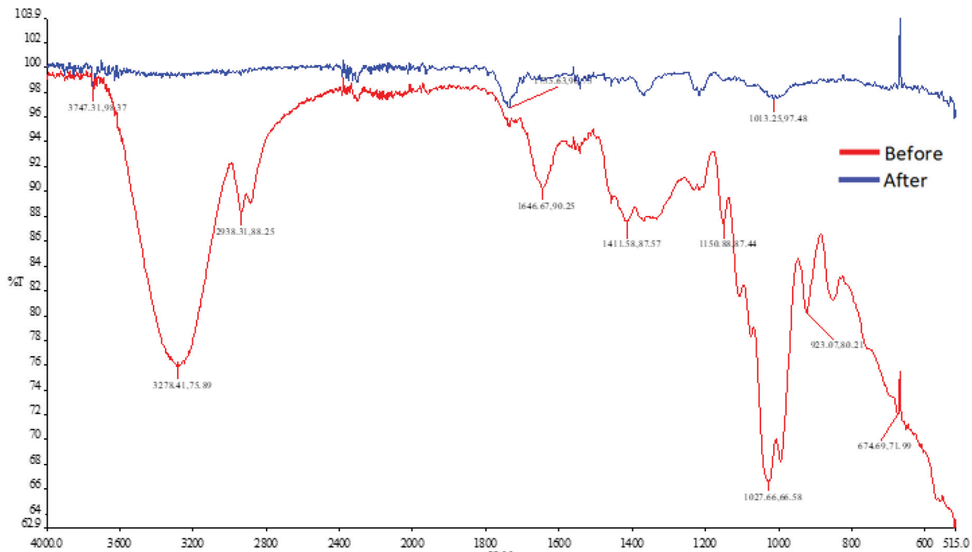

Figure 6: The FTIR of the BPC bioplastic before and after the experiment

Table 4: The summary of the main FTIR absorption peak for both type of bioplastic and their functional group

Functional Group	Wavenumber [cm^{-1}]			
	Corn-starch Based (BP)		Corn-starch Based with Chitosan (BPC)	
	Before	After	Before	After
O-H	3278.95	3327.08	3278.41	3747.31
C-H	2932.57	—	2938.31	—
C=O	1646.51	1642.35	1646.67	1735.63

composition of the bioplastics before and after the batch culture experiment.

It can be observed that the red lines in Figures 5 and 6 on the BP and BPC exhibit similar patterns of the FTIR spectrum. The O-H stretching band's hydroxyl bonds for the BP were 3278.95 cm^{-1} and 3278.41 cm^{-1} for the BPC. This reduction peak is in contrast with the findings of Bajer *et al.* (2020), as the mixture of the starch/chitosan/aloe vera has two hydroxyl peaks that characterise free water in the samples. It has been related to the aloe vera gel, classified as a highly hydrated organic material (Bajer *et al.,* 2020). However, in both the BP and BPC samples, no free water existed before the batch culture experiment as compared with Bajer *et al.* (2020).

It can be seen in Figures 5 and 6, in which the blue lines indicate the FTIR spectrum after the experiment for both BP and BPC bioplastics, the reduction curve of hydroxyl bonds O-H has flattened. The BP had a value of 3327.08 cm^{-1} and the BPC had exhibited a higher range of 3747.31 cm^{-1}. The value changes after the 14 days must be due to both bioplastics absorbing water during the experiment phase. It can be seen that the BPC have a high value of FTIR spectrum compared with the BP due to the addition of chitosan. The chitosan additive displays a hydrophilic behaviour due to it being in the hydroxyl group (Vernaez *et al.,* 2019).

Another significant result found from each graph (Figures 5 and 6) is that the alkanes with the C-H bond have disappeared for both types of bioplastics after the 14 days of the incubation period. It can be noted that before the batch culture experiment, the red lines for both graphs indicated an FTIR spectrum of 2932.57 cm^{-1} for the BP and 2938.31 cm^{-1} for the BPC. At the end of the incubation period, no C-H bond existed in both samples of bioplastics due to the biodegradation process and the breakdown of starch by microorganisms (in this case, the inoculum of leachate) (Tai *et al.,* 2019). High carbon contents in the form of polymeric biomass of starch in both samples of the BP and BPC enhanced the decomposition process (Hwang *et al.,* 2020).

Conclusion

This study finds that corn starch-based bioplastics with chitosan (BPC) has a more complex and compact structure than corn starch-based bioplastics (BP). After the degradation process in the batch culture experiment, both samples had an irregular shape and rough texture with the presence of pores across the samples. The FTIR spectra displayed the presence of O-H, C-H and C=O absorption peaks and the functional group of C-H disappeared after the experiment. It can be concluded that BP can degrade faster than BPC at a rate of 5.5%. Throughout the 14 days, the percentage removals of ammonia-nitrogen for BP and BPC were 40.78% and 38.03%, respectively. The percentage weight reductions for BP and BPC were 92.2% and 86.7%, respectively. The ammonia-nitrogen removal and the weight reduction percentage were better in BP. Therefore, it can be denoted that bioplastics can be successfully degraded in the leachate medium with BP performing better than BPC.

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