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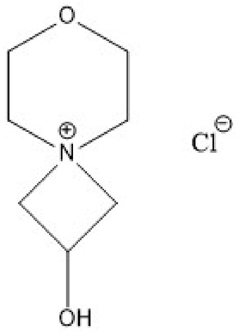
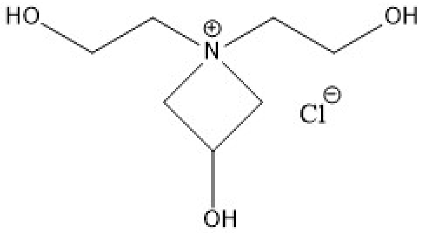
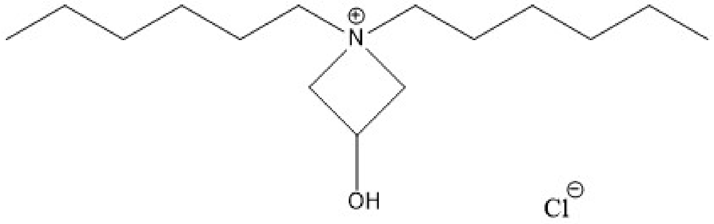
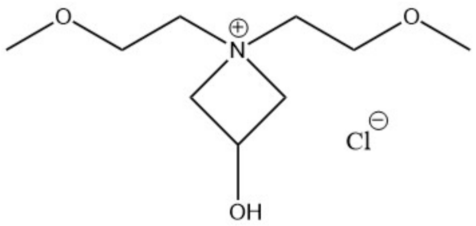

Conjugation of azetidinium salt to cellulose fibers

A study on how different azetidinium salts affects the water retention value of the cellulose

Degree project in the graduate engineer programme in chemical engineering

Emma Gustafsson

DEPARTMENT OF CHEMISTRY AND CHEMICAL ENGINEERING

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Cover: The molecular structure for the four different azetidinium salts used in this project

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Abstract

Cellulose is a material where the usage of it has increased over the years. In the future the hope is to be able to use it for even more applications, for example instead of plastics or as a part in textiles. To do this cellulose has to be modified in some way depending on what the aim of use is. Conjugation of azetidinium salt to the cellulose is one way to modify it. The hope is that when an azetidinium salt conjugates to the cellulose it will lower the water retention value of the cellulose.

The aim of this study was to see how the water retention value was changed after conjugation of azetidinium salts to cellulose. It was also studied how different methods for swelling of the cellulose in the pre-treatment step affected the results of oxidation and conjugation with azetidinium salts. So it was tried to swell the cellulose with $ZnCl_2$ instead of just water. The first step was to synthesize three different azetidinium salts and the determination of the structure was done with the help of H-NMR. The cellulose was oxidized with the help of TEMPO-oxidation before the azetidinium salt was conjugated to it. To analyze the result of the conjugation of azetidinium salts to cellulose FT-IR was used. For determination of the degree of oxidation a method with titration was used. The most important result for this report was the water retention value and the method for this is a universally used method called ISO 23714:2014.

Due to the fact that one of the synthesized salts failed two was borrowed from the storage at Chalmers. With water swelled cellulose all four salts conjugated to the cellulose. When using $ZnCl_2$ swelled cellulose the one salt tested did not conjugate and there was nothing that confirmed even if oxidation had taken place. In terms of water retention value, when using an azetidinium salt that has long coal chain that goes horizontal from the nitrogen atom the value got lower. So, it can be expected that for salts with even longer horizontal coal chains the water retention value should be even lower. Therefore, this would be something to study in the future if lower water retention values are desired.

Keywords: cellulose, azetidinium salt, water retention value, TEMPO-oxidation.

Acknowledgements

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Emma Gustafsson, Gothenburg, May 2023

List of Acronyms

Below is the list of acronyms that have been used throughout this thesis listed in alphabetical order:

Az-salt	Azetidinium salt
FT-IR	Fourier transform infrared spectroscopy
HCl	Hydrochloric acid
NaBr	Sodium bromide
NaOH	Sodium hydroxide
NaOCl	Sodium hypochlorite
NMR-spectroscopy	Nuclear magnetic resonance spectroscopy
TEMPO	2,2,6,6-tetramethyl-1-piperidinyloxy
WRV	Water retention value

Contents

1

Introduction

Cellulose is an important material for the future. The reason for this is the fact that the usage of fossil-based products need to be reduced. Hopefully products made from cellulose can take its place and be used instead in mainly products that earlier were made from plastic. Since cellulose comes from plants it is renewable and can be produced in abundance. Therefore, it would be much better to use cellulose instead of plastic in as many applications as possible. One negative thing with starting to use cellulose in such a widespread way is that arable land that earlier been used for food is converted to producing plants with more cellulose. This would be a factor if it is more profitable to help with cellulose production instead of food production. Another problem could be that deforestation is picking up rapidly and that would be catastrophic for the environment. But as long as these two problems are kept under control it would be beneficial to try to switch out as many plastic products as possible.

To be able to do a complete shift there is more research needed since cellulose does not have the same properties as plastic. This research is coming but there will still be some time before every plastic product has been switched out to one based on cellulose. It is not even fully established if all plastic products can be based on cellulose, or if some still have to be plastic. For the cellulose to be used instead it has to modified to get different material properties. Which properties that are needed takes different methods of modification. One method that is being researched now is how azetidinium salts affect material properties. Most of the research for this has been done on nanocrystalline cellulose. In this study the same thought has been used but instead of using nanocrystalline cellulose, cellulose fibers have been used to see how azetidinium salts affect the water retention value. The reason for why the water retention value has been studied is because for cellulose to be used in some plastic products the water retention value has to be as low as possible.

In this thesis the effect of azetidinium salt on cellulose fibers will be examined. If the different structure of the azetidinium salt affects the cellulose different will also be examined. This is done by using four different salts and conjugating them to the cellulose. Another factor to how and if the azetidinium salt is conjugated is to try two different swelling methods for the cellulose before oxidizing. The method of oxidizing will remain the same throughout all reactions. The degree of oxidation is important for the end result and therefore an extra two-week oxidation will be done to see how time affects the degree of oxidation.

1.1 Aim and limitations

The aim with this thesis is to examine how different structures of the azetidinium salt affect the water retention value of the cellulose. Another thing that is examine is if different swelling methods of the cellulose before oxidizing gives a different result. Limitations of this report is that only TEMPO-oxidation will be used for oxidation of the cellulose.

2

Theroy

2.1 Azetidinium salt

Azetidinium salt is a salt that is synthesized by a reaction between epichlorohydrin and a secondary amine. Often is the reaction done in two steps. The first reaction gives a not ring-closed salt. Most of the time a ring-closed salt is desired therefore a second step in the synthesis is needed. In the second step the not ring-closed salt is ring-cosed often with the help of heat.

There are two common types of azetidinium salt, these are the symmetrical ones or the asymmetrical ones. If an azetidinium salt is symmetrical or asymmetrical is decided if both end groups are of the same length. In figure 2.1 an example of how the structure of an azetidinium salt can be is shown. The R groups represent coal chains of various lengths and structures, they depend on which secondary amine that was used during synthesis. For this experiment symmetrical azetidinium salt has been used. The reason for why only symmetrical azetidinium salts has been used is because otherwise secondary amines would also need to be synthesized. As seen in figure 2.1 the end groups are attached to the nitrogen molecule.

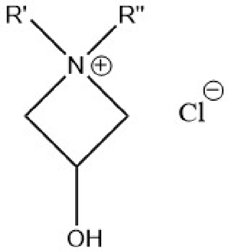

Figure 2.1: *General molecular structure of an azetidinium salt*

Something that is beneficial with the azetidinium salt is that it can be prepared in advance and then stored until it is supposed to be used. Earlier aziridinium ions were used in similar ways to azetidinium salts but they were more reactive and could not be stored for a longer amount of time. The reason for why these forms of molecules is used is because of the selective reactiveness it has. Azetidinium salts can be good to use for modification of other molecules because of that. In this study the azetidinium salt is used to modify cellulose. There is also a possibility that depending on the structure of the salt it will modify the cellulose in different ways [1].

2.1.1 Epichlorohydrin

Epichlorohydrin is a chemical with a big area of use. At room temperature epichlorohydrin is clear, colorless and in liquid form. Due to the structure, seen in figure 2.2, of the molecule it is a rather reactive chemical and therefore is used in many different areas. For example, it is used within the manufacturing industry for paper, paint and pesticides. In laboratories epichlorohydrin is used as the main reactant to synthesis azetidinium salts as mentioned earlier.

Figure 2.2: *Molecular structure of epichlorohydrin*

In its pure form there are signs that epichlorohydrin is toxic for nature and life. In humans and animals there are studies that show an increased risk for cancer but also that it can affect the central nervous system. If epichlorohydrin is leaked into nature it can also affect and destroy plant life. What amount of epichlorohydrin that is needed for these effects is not fully established so it is best to be careful when handling epichlorohydrin. After reaction with other molecules the risk of harmful effects are shown to be significantly decreased. That is the reason why epichlorohydrin usage is so widespread in the manufacturing industry [2]

2.1.2 Secondary amine

For synthesis of azetidinium salt a secondary amine is used to react with the epichlorohydrin. A secondary amine is an amine where the nitrogen atom is bound to two coal atoms. To these two coal atoms carbon chains with different lengths can be attached depending on which secondary amine is used. In figure 2.3 a general example of a secondary amine is shown. The R- groups that are attached represent the coal chains with different lengths and structure [3].

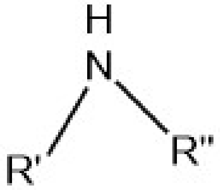

Figure 2.3: *Molecular structure of a secondary amine*

2.2 Cellulose

Cellulose is a renewable material since it origins from plants. Cotton consists of around 90 % cellulose while a regular tree consists of about 40-50 %. Since it is considered a renewable material, it is interesting for the manufacturing industry and there is ongoing research on how to make it more usable. In the future it might be possible to use cellulose instead of, for example, plastics. One problem for this is that cellulose is a hydrophilic material while plastics are a hydrophobic material. Cellulose has a relative high water retention value since it can absorb water and hold it. This is not possible for plastics since they are very hydrophobic. So, in order for it to be possible to use cellulose instead of plastic the water retention value has to decrease and be as low as possible. There are other applications for cellulose, for example using cellulose in textiles. In that case the need for modifications also exist but not necessarily the water retention value [4] [5].

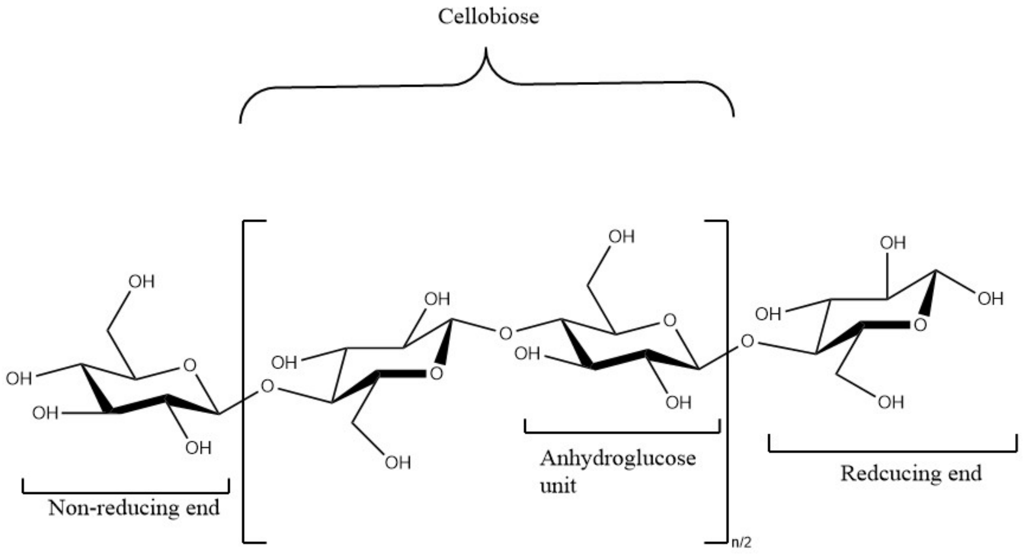

Figure 2.4: *Molecular structure for cellulose*

Cellulose is a material that is rather easier to modify due to its structure seen in figure 2.4. Cellulose consists of glucose units which make it into a polysaccharide. It also has two end groups that are important for its structure, these two are the non-reducing end-group and the reducing end-group see figure 2.4 to see how they are structured. For modification the anhydroglucose unit is the most important and these are the units there are most of in cellulose molecules. The reason for why this is important for modification is because the hydroxyl groups on the ring can react with other molecules. There is also just one reducing end and one non-reducing end but a lot of anhydroglucose units. Since one glucose unit also counts as a monomer and the cellulose molecule consists of more than one it has a degree of polymerization. This is how many times the monomer is repeated in the structure for cellulose it can be all from 100 - 20000 depending on where the cellulose comes from and how it has been treated [6] [7].

2.3 TEMPO oxidation

In TEMPO oxidation the molecule called TEMPO is used as the main catalyst in the oxidation reaction. The real name for the TEMPO molecule is 2,2,6,6-Tetramethyl-1-piperidinyloxy and has the structure as seen in figure 2.5. It is due to the structure of the molecule that it works great as a catalyst. The fact that is a stable free radical is also why it is used in many oxidation forms. It works best in oxidation reactions that occur in either aqueous media or organic media [8][9].

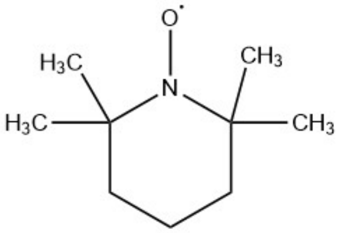

Figure 2.5: *Molecular structure of TEMPO*

In a TEMPO oxidation a co-oxidant is needed because the TEMPO molecule is mainly working as a catalyst. What molecule that is used as co-oxidant depends on the molecule that is supposed to be oxidized. For cellulose it has been determined that sodium hypochlorite (NaOCl) is one of the best co-oxidant. For cellulose another catalyst is also used in the oxidation step, the second catalyst is sodium bromide (NaBr). The reason for why these three are used in combination is because they will give a rather selective oxidation of the cellulose and not react in unfavorable ways with each other [10]. When oxidizing cellulose with TEMPO oxidation the aim is to form carboxyl groups on the anhydroglucose units in the cellulose see figure 2.6.

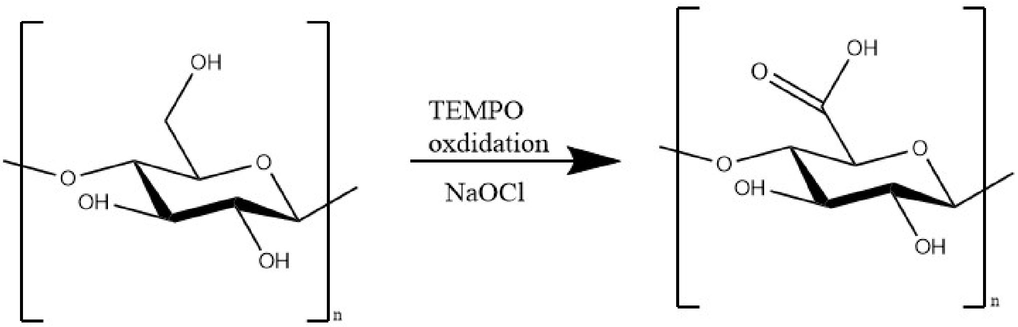

Figure 2.6: *Reaction of anhydroglucose unit when TEMPO oxidized*

To keep track of the reaction and see if it is oxidizing, the pH is kept at pH 10 with the help of sodium hydroxide (NaOH). When the pH decreases it means that a reaction is occurring and that NaOH is consumed. The oxidization process is said to be done when the pH is not decreasing anymore. In terms of yield, the TEMPO oxidation of cellulose is not efficient since it yields a low degree of oxidation. After about 24 hours the degree of oxidation is around 20 % which is rather low. The largest amount of oxidation will occur in the first hour and then it will go rather slowly for the rest of the time the reaction is ongoing. Exactly how long a TEMPO oxidation of cellulose can stand and still generate oxidation is not determined [10] [11].

2.4 NMR spectroscopy

Nuclear magnetic resonance spectroscopy, also known as NMR spectroscopy, is used to analyze samples. To prepare a sample for NMR a small drop of the analyte is taken. This small drop is put in a small glass tube. To the glass tube an appropriate solvent is also added, examples are deuterated chloroform, dimethyl sulfoxide and methanol-D. For the solvent to not appear in the analyze is it important to use solvent consisting of deuterium which is an isotope of hydrogen. The reason for this is that one of the most common ways to do NMR analysis is to analyze the hydrogen atoms in the sample.

The two most common ways to do NMR analyse is to either analyze with regards to the nucleus of 13-coal or the nucleus of 1-hydrogen. For this study 1-hydrogen analyze has been performed. If a sample has a very low amount of hydrogen atoms or none at all but still has coal atoms a 13-coal analyze can be performed instead. In 1-hydrogen NMR the placement of the hydrogen atoms can be determined by how the peak looks in the spectrum. The peak depends on coupling with other hydrogen atoms and what the bond to the carbon atom looks like[12].

NMR analysis is done with a very strong magnet and the magnetic field this magnet creates. The hydrogen nucleus is positively charged which makes it possible to affect it with the magnetic field. When the magnetic field is applied to the sample the hydrogen nucleus magnetic fields will order themselves in two different ways regarding to the applied magnetic field. The nucleus magnetic field will either be parallel against the applied magnetic field or anti- parallel. Depending on how the nucleus magnetic field has oriented it will have different amounts of energy. If the field has oriented itself parallel with the applied magnetic field, it has a lower energy than anti-parallel. Therefor there will be slightly more nucleus that has oriented themselves parallel than anti-parallel.

When the nucleus magnetic field has oriented itself against the applied magnetic field radiofrequencies are emitted. These radio frequencies are adsorbed by the nu-cleus which increases the energy level inside the nucleus. When the impulses of radiofrequencies are turned off again the nucleus will emit the adsorbed energy to return to its original state. This emitted energy is the one that is detected and an-alyzed. Depending on where in the molecule the nucleus is and which surrounding atoms it has it will adsorb different frequencies. That is why the exact placement of a hydrogen atom in a molecule can be determined[13][14].

2.5 Fourier transform infrared spectroscopy

Fourier transform infrared spectroscopy also known as FT-IR uses the absorption of light. The absorption then gives an infrared spectrum for your samples. To create this spectrum the FT-IR machine shoots a beam of light consisting of lights at different wavelengths. This is done multiple times and every time the spectra of the wavelengths of the light are shifted. In the machine there is a detector that detects what wavelengths that are not absorbed and which are radiated from the sample if there are any. The data from the detector is then converted into a spectrum with the help of a Fourier transform. In the given spectrum there are peaks that correspond to different types of molecular structures that can be present in the samples. This can be determined since different molecular structures absorb or emit light at different wavelengths. For example, in this study esters should be formed when the azetidinium salt has conjugated to the cellulose and these are generally seen around 1700 in the spectrum. The biggest downside with FT-IR is that if that analyze is supposed to be on a solid material it has to be completely dry. If there is still water present this could interfere with the spectrum. Another thing that could give a spectrum that is hard to read is if the sample consists of too many different molecular structures. This would lead to a very complex spectrum and therefore it can be hard to exactly determine what type of molecular structures that are present [15][16].

Most of the times FT-IR is used to determine what the structure of the sample is. In this case FT-IR is used to verify if the reactions have gone as planned. This is possible because the desired types of molecular structures in the sample are known. As said before it is known that esters would form when the azetidinium salts conjugate to the cellulose. If no difference can be seen at the sample for oxidized cellulose and conjugated cellulose at around 1700 no conjugation has occurred [17].

2.6 Water retention value

Water retention value is a way to measure how much water something can hold, in this case cellulose. It is confirmed that the water retention value increases if the cellulose is allowed to swell since this leads to internal fibrillation since the cellulose is stirred while swelling. The internal fibrillation will increase the capacity to hold water since small pores will be created inside the cellulose fiber where more water can be stored. The water retention value can be modified by different reactions. For example, oxidation of cellulose will further increase the water retention value. WRV can be good to know while making products of cellulose since different products have different needs of holding water. For example, when making diapers it is important that the water retention value is high. For manufactures to be able to compare their WRV with other a standard method of how to measure the water retention value has been formed [18] [19] [20].

3

Method

3.1 Synthesis of azetidinium salt

To synthesis azetidinium salts epichlorohydrin is used as the main reactant. Depending on what the desired structure of the azetidinium salt is different secondary amines can be used. It is the secondary amine that decides the structure and size of the azetidinium salt. In this case morpholine, diethanolamine and dioctylmethylamine has been used to synthesis three different azetidnium salts. The common structure of a ring-closed azetidnium salt can be seen in figure 2.1. The two R-groups are dependent on which secondary amine that is used. For the synthesis an already established protocol has been followed.

To start the synthesis 10 *ml* isoproanol is added to a round bottom flask with a magnetic flea. Thereafter 11 *mmol* of epichlorohydrin is added to the same flask. Lastly 10 *mmol* of the secondary amine is added. This solution is then left stirring at room temperature overnight. This first reaction gives a not ring-closed salt. To get a ring-closed salt further reactions have to happen. For the further reactions only the dried not ring-closed salt is needed. The isopropanol is evaporated in a rotavapor at 50 °C, the reason for the low temperature is to not affect the salt.

When only the dry salt remains and possibly a tiny amount of the isopropanol is left 10 *ml* of a solvent containing water and isopropanol with the ratio 9:1 is added to the salt. This solution is left stirring at 80 °C for two hours. After those two hours the same procedure is done to evaporate the solvent with the help of a rotavapor. Due to the low temperature, it might be hard to get rid of all the water. To help the water evaporate a small amount of methanol can be added. This can be done multiple times during the time the solution is on the rotavapor. Even with the help of methanol it might not be possible to get rid of all the solvent. Depending on what the salt is supposed to be used for, that might be okay. In this case it was okay since later on it was going to be used in a reaction where water was one of the components. A sample of the dry ring-closed salt was taken for NMR analysis. From the NMR result it can be decided if the salt has ring-closed and if it has reacted as it supposed to and led to the desired structure of the azetidinium salt.

3.2 Oxidation of cellulose fibers

For oxidation of cellulose fibers a method called TEMPO-oxidation is used. For this specific oxidation sodium hypochlorite is used as the co-oxidant and sodium bromide as an extra catalyst. An already established protocol was used for the oxidation procedure [10].

Before the oxidation process was started the cellulose was swelled in water overnight. 1 *g* cellulose (dry weight) equal to 6.025 *g* of the wet BKP cellulose was added to 60 *ml* of water and stirred at room temperature overnight. When the cellulose had been swelling overnight a TEMPO-solution was added dropwise. The TEMPO-solution consisted of 20 *ml* deionized water where 0.017 *g* of TEMPO and 0.0189 *g* of NaBr had been dissolved. After the TEMPO-solution 3.7 *ml* of NaOCl, adjusted to pH 10 with the help of 0.1 *M* HCl, was added dropwise. When both solutions have been added to the cellulose the pH is checked, if needed the pH is adjusted to pH 10 with the help of 0.5 *M* NaOH. The oxidation solution is then left stirring at room temperature for 24 hours. During these 24 hours the pH is monitored, and the change is noted. If the pH-value goes below 10, more 0.5 *M* NaOH is added until the pH is 10 again. For the best result it is important that the pH is kept at 10 for as many of these 24 hours as possible. After 24 hours the cellulose solution is vacuum filtered through a nylon-filter to obtain only the oxidized cellulose.

A method using zinc chloride in the swelling step of the cellulose was tested. Instead of letting 1 *g*(dry weight) of cellulose swell in only water it was swelled in a solution consisting of 100 *ml* deionized water and 5 *g* zinc chloride. The cellulose was left stirring in this solution overnight just as before. After the swelling step the rest of the oxidation process was the same as for cellulose swelled in water.

To see if the oxidation process yields a higher degree of oxidation with more time a two-week reaction was set up. For this reaction 1 *g* (dry weight) of cellulose was swelled in water over night as before. Then the steps for TEMPO-oxidation as stated above were followed. The big difference is that the reaction are kept stirring for two weeks and then filtered. Over the two weeks the pH of the reaction is measured twice a day to be able to keep the pH at 10.

3.3 Conjugation azetidinium salt to cellulose

For the conjugation of the azetidinium salt to cellulose the cellulose has to be acidic. The first step is therfore to acidify the cellulose. This is done by in this case adding 0.700 *g* of the oxidized cellulose to a beaker. To the same beaker 18 *ml* of deionized water and 2 *ml* 0.1 *M* HCl is added. The beaker is then put on stirring at room temperature for about an hour or to the pH is around 3. During this time a sample of oxidized cellulose is taken for measurement of water content. This is needed to be able to calculate the dry weight of the used cellulose. When the pH is around 3 the solution is again vacuum filtered through a nylon filter to obtain the acidified oxidized cellulose. During the filtering process the cellulose is washed with water and ethanol.

When the acidified oxidized cellulose is obtained, it is added to 40 *ml* of deionized water. From the dry weight of the used cellulose the amount of azetidinium salt needed for the reaction is calculated. The wanted molar ratio between the dry weight cellulose and azetidinium salt is 1.7:1. Due to the low degree of oxidation for cellulose a surplus of cellulose is needed. Normally the degree of oxidation for cellulose is between 20-30 %.

The correct amount of azetidinium salt is dissolved in 10 *ml* of water. This is then added to the cellulose solution dropwise. The reaction solution is then left stirring at 90 °C for 6 hours. After these 6 hours the heat is turned off and the reaction is stirred over night at room temperature to cool down. When the solution has cooled down it is vacuum filtered through a nylon filter again. The cellulose is washed with water and ethanol during this filtration. This time it is extra important to wash with ethanol too so that salt that has not reacted is washed away. All salts are not soluble in water therefore ethanol is also used to wash them away.

A sample of cellulose can now be taken and analyzed with the help of FT-IR. From the FT-IR it can be determined if the salt has conjugated to the cellulose. The sample is compared to samples from non-treated cellulose and oxidized cellulose. Depending on how the spectrum looks it can be confirmed if the salt has conjugated to the salt but also to see if the oxidation been successful. That is why an oxidized sample and a reference non treated one also is analyzed.

The amount of cellulose used in this procedure is used for conjugation with one azetidinium salt. Since four different azetidinium salts were used in this study the procedure was repeated four times to make sure the same amount of cellulose was used for all four.

3.4 Determination of degree of oxidation

After oxidation the degree of oxidation can be determined. This is done with the help of titration to see how much carboxyl acid that has been produced. To do this, three samples from the oxidized and acidified cellulose is taken. Three samples of cellulose that just have been swelled overnight is also taken to act as a reference. The reason why the three samples are taken is to get a more accurate value, and sometimes things can go wrong when titrating, so it is also for extra safety. To get as accurate results as possible it is important that the cellulose for the different samples weigh approximately the same. In this case 0.020 *g* of cellulose where the fourth decimal was allowed to vary was used for the samples. The exact weight are presented in table A.1 in the appendix. Every sample was then prepared by adding 40 *µl* 0.5 *M* NaCl to the cellulose and thereafter 19.6 *ml* of distilled water. This mixture was then allowed to be stirred overnight so that the cellulose went into suspension.

The next day when the cellulose has gone into suspension the first sample is taken, and the pH is checked. A pH of 3 is wanted so if it is higher 1 *M* HCl is added. The amount of HCl added is noted. When the pH is right the titration starts. This is done in an automatic titration machine which adds 0.01 *M* NaOH at intervals. On the titration machine the sample is stirred and the NaOH is added until the pH is 10. Then the data from the titration can be downloaded and plots can be made to compare the three samples. The same procedure is then repeated for the three reference samples.

From the titration data the amount of carboxyl groups that have been formed can be calculated. This is done by finding the data connected to the highest ERC. ERC is a function that detects the first derivative of the titration curve. At the highest ERC the relation between the amount of carboxyl groups and hydroxy groups is equal also called the equilibrium point. With the help of the 1:1 molar relationship between NaOH and R-COOH the amount of moles R-COOH can be calculated. Since the volume of NaOH used at that point and the concentration is known, equation 3.1 can be used to calculate the amount of moles NaOH.

$$n = c * V \tag{3.1}$$

To be able to calculate the carboxyl group content per gram cellulose the weight of the cellulose needs to be calculated. To do this the mass of wet cellulose is multiplied with the dry content of the cellulose. For the carboxyl group content per gram cellulose equation 3.2 is used.

$$Carboxyl\ group\ content = \frac{n(NaOH)}{dry\ weight\ cellulose} \tag{3.2}$$

To calculate the degree of oxidation equation 3.3 is used:

$$Degree\ of\ oxidation = \frac{V(NaOH) * [NaOH] * M * V(0)}{2 * m(0) * V(1)} * 100 \tag{3.3}$$

V(NaOH) is the volume of NaOH added, [NaOH] is the concentration of the NaOH, M is the molar weight for cellulose, V(0) is the starting volume, m(o) is the dry weight of the cellulose and V(1) is the end volume including everything which has been added during the titration.

3.5 Determination of water retention value

To see if the cellulose has changed when reacted with azetidinium salt the water retention value was investigated. To do this, three samples from each of the four reactions between cellulose and azetidinium salts were prepared. Firstly, one watch glass per sample needs to be washed, dried and weighed with the weight noted. In this case 12 watch glasses were needed. The fibers are then weighed out to 0.086 *g* in dry weight. For the analysis to be accurate the weights had to be very similar, only the fourth decimal is allowed to vary. The fibers were weighed onto the filter part of the cylindrical tube. The filter is then put into a small plastic beaker with 6 *ml* deionized water. Then 20 *ml* water is added to the fibers and filter. This mixture is then allowed to soak for four hours. Do four samples at a time with an interval of 20 minutes.

When the first four samples have been soaking for four hours the filter is put back into the centrifuge tube. The centrifuge tube is then weighed, and more water is added if needed to balance the weight of the four centrifuge tubes. A difference of 0.1 *g* is allowed. They are then centrifuged using a 12265 rotor in a Sigma 4-16ks centrifuge for 10 minutes at 4000 rpm, 1896 g and 23 °C. The wet fibers are taken out of the filters and placed on a watch glass after the centrifugation. The watch glass with the wet fibers is weighed and the weight of the cellulose is calculated. Repeat these steps for all 12 samples. When all the samples have been centrifuged and the wet fibers are weighed the watch glasses with the fibers on are put in an oven. Originally, they were supposed to be in the oven for 24 hours at 100 °C. Due to discoloration on some of the samples after one hour the temperature was dropped to 70 °C for a couple of hours. Overnight the temperature was dropped to 50 °C then in the morning the temperature was turned up to 100 °C and this time no more discoloration occurred. Due to the lower temperature the samples were in the oven for almost 48 hours instead of 24. The second night the temperature was again turned down to 50 °C and then turned up to a 100 °C in the morning again. During this time, it is important to weigh the sample at semi-regular intervals to see the mass change. When there has not been any change in mass for 5-6 hours at 100 °C the samples are deemed to be completely dry. The completely dry samples are then weighed and the mass of the dry fibers are noted.

After all these steps the water retention value is calculated with equation 3.4:

$$WRV = \frac{m(wet) - m(dry)}{m(wet)} \tag{3.4}$$

m(wet) is the weight of the wet sample right after centrifugation. m(dry) is the weight of the completely dry sample, in this case after 48 hours in the oven.

4

Results

In the process of making salts three different ones were made. Only two of them succeeded and gave a correct ring-closed form. Therefor two other salts that had been confirmed correct in early studies were used to give a total of four different azetidinium salts. The NMR results for the three azetidinium salts synthesized for this study will be shown. Unfortunately, there are no NMR results for the two borrowed salts to present in this paper.

For the oxidation of cellulose two different methods were used for the pre-treatment step. One where the cellulose swelled in water over night and one where the cellulose swelled in a mixture of water and zinc-chloride. When looking at the FT-IR data it shows that when it was swelled in zinc-chloride no oxidation occurred and therefore no visible reaction with the azetidinium salt. Due to the fact that the $ZnCl_2$ swelled cellulose did not react with the first salt it has not been used for further analyses. This means that when the FT-IR results for conjugation with the three other azetidnium salts $ZnCl_2$ swelled cellulose has not been used. There have not been any studies on the degree of oxidation or the water retention value for it either.

For the degree of oxidation one 24-hour oxidation was performed and one 2-week oxidation. It was compared how the longer time would affect the degree of oxidation and if it would be worth the extra time for future conjugations with azetidinium salts. A higher degree of oxidation would be beneficiary since it would help the azetidinium salts to conjugate to the cellulose.

In this chapter the result of the water retention value test is also presented. Here it can be seen how the different azetidinium salts affect the WRV for cellulose.

4.1 NMR

The first NMR spectrum in figure 4.1 shows the spectrum for the failed attempt to do ring-closed dioctylmethyl azetidinium salt. From the figure it can be seen that there are some peaks between 6.5 and 6 ppm in the spectrum that should not be there if the salt had been formed correctly. These peaks show that there probably are double bonds that have been formed which are not desired in our salt. There could also just be a contaminant that has come into the NMR tube but the probability of that is low. Since the structure was not correct this salt was excluded from further use in this study.

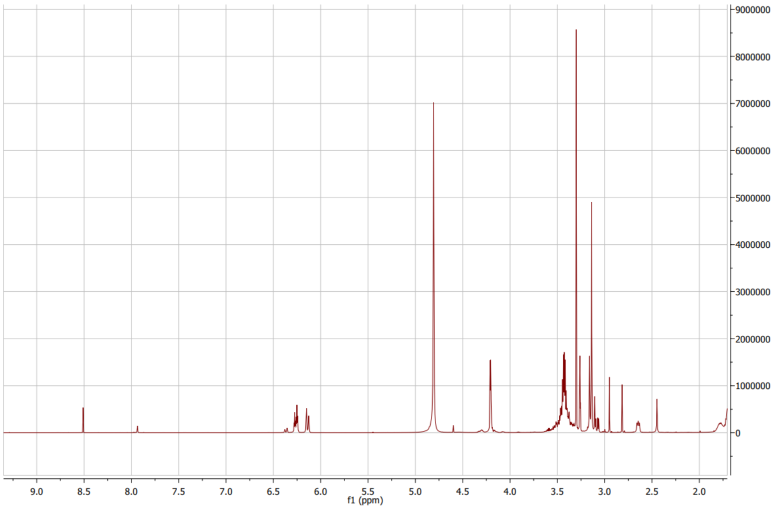

Figure 4.1: *NMR spectrum for the failed attempt of ring-closed dioctylmethyl azetidinium salt*

Figure 4.2 shows the molecular structure of the ring-closed morpholine az-salt and in figure 4.3 the corresponding NMR spectrum is presented.

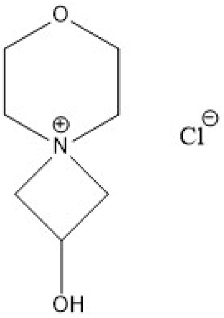

Figure 4.2: *Molecular structure for morpholine azetidinium salt*

The NMR spectrum is correct for the most part. One peak show that there is still some water present which means that not all the solvent was removed before finishing the evaporation. There is also one peak that shows there are two different positions of a specific hydrogen atom. The best would be if that hydrogen atom always had the same position, but it is okay to use it even if both positions are present. Beside the NMR spectrum is the molecular structure for the morpholine azetidinium salt.

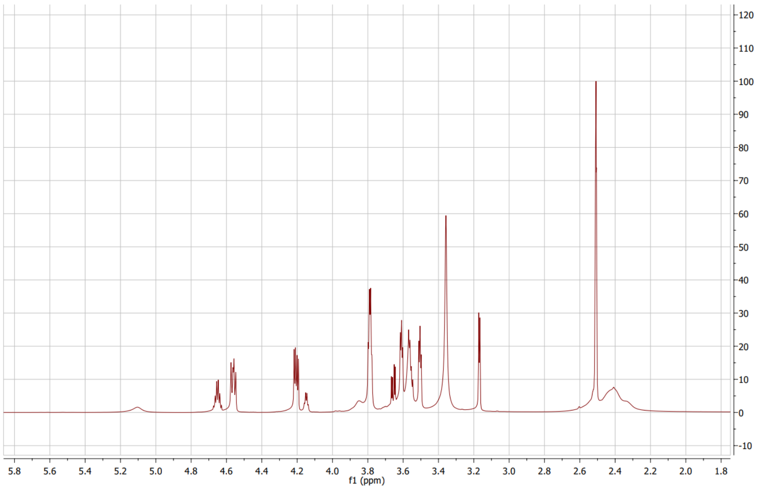

Figure 4.3: *NMR spectrum for ring-closed morpholine azetidinium salt*

The last salt that was synthesized is diethanol az-salt. In figure 4.5 the molecular structure is presented with the correpsonding NMR spectrum in figure 4.4.

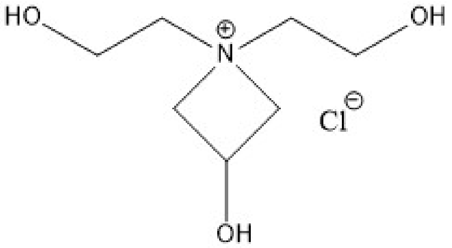

Figure 4.4: *Molecular structure for diethanol azetidinium salt*

Also, in this NMR spectrum there are peaks that show that there is some water present. Other than that, the spectrum is as expected. The problem with water presence is okay for the intended use of the salt but for other uses it might not.

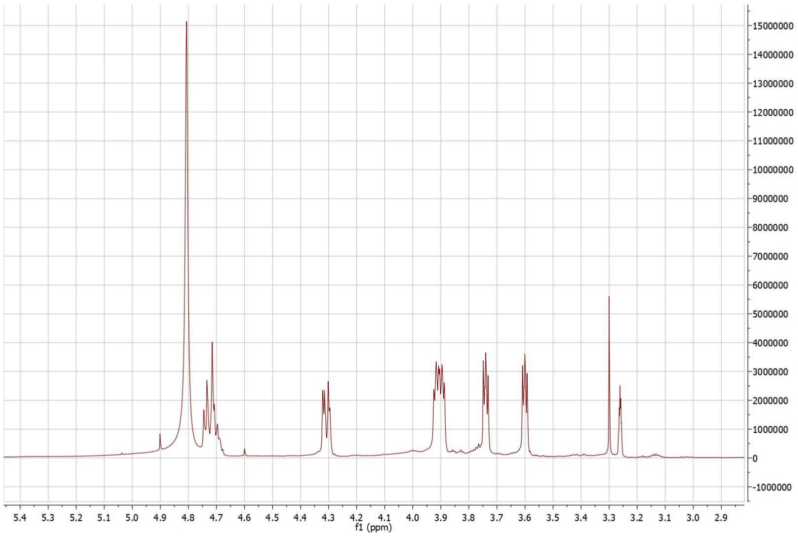

Figure 4.5: *NMR spectrum for ring-closed diethanol azetidinium salt*

As mentioned before there are is NMR spectrum for the two other salts that have been used therefore it is not presented here. Instead, it is assumed that the two salts have ring-closed in the right way and has the structure as seen in figure 4.6

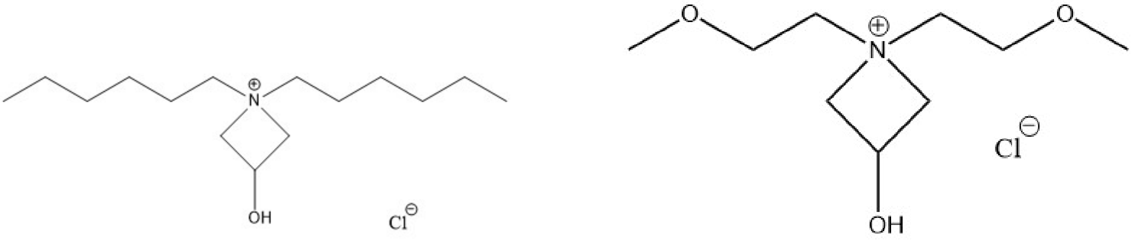

Figure 4.6: *Molecular structure for 1,1-dihexyl-3-hydroxy azetidinium salt (left) and* COC_2*-N-*C_2OC *azetidinium salt (right)*

In the appendix the predicted NMR spectrum for the four salts that was used is presented and which hydrogen atom most likely corresponds to which peak. The computer predicted NMR spectrum might deviate a bit from the real spectrum. This can be due to the fact that different solvents has been accounted for and used. It can also deviate since it is a prediction and might not account for everything that can interfere with the sample in reality. See figure A.1, A.2, A.3 and A.4 for the predicted NMR spectrums.

4.2 FT-IR

For FT-IR samples has been taken both on just the oxidized cellulose but also when azetidinium salt has conjugated on to it. The sample of oxidized cellulose has been compared with a sample of non-treated cellulose to see whether or not the oxidization has occurred. Samples from the conjugation with different salt have been compared to the oxidized cellulose. In figure 4.7 the whole spectrum for the oxidized cellulose and the reference sample is presented.

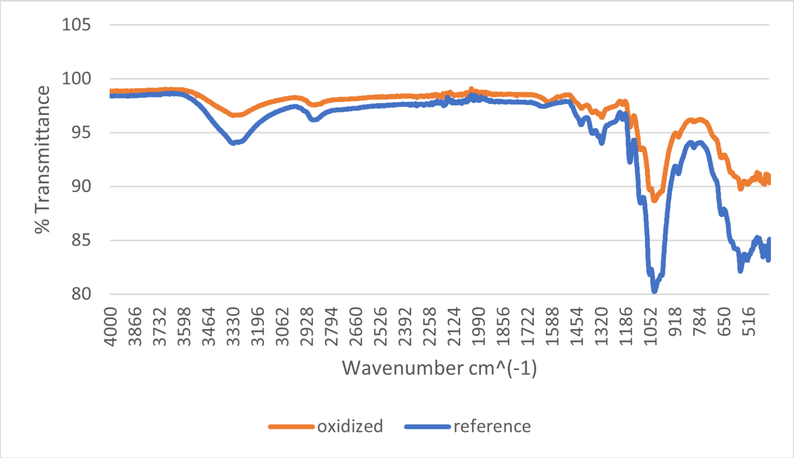

Figure 4.7: *Full FT-IR spectrum for the reference and oxidized cellulose*

Here it can be seen that oxidation probably has happened. When doing TEMPO-oxidization a carboxyl group is formed as seen in figure 2.6. The peak for that is around 1738 cm^{-1} [17]. In this case there is no peak there but in some instances the carboxyl peak merges with a water peak and instead appears around 1600 cm^{-1} and that is most likely what has happened here. A small peak can be seen around 1600 which indicates that oxidization has occurred.

In figure 4.8 a comparison between samples from conjugation with the different salts to oxidized cellulose is shown. The probable number of esters formed for the different salts can also be compared. For this comparison only a bit of the spectrum is presented. This is due to the fact that the esters bonds that supposed to be formed usually has a peak around 1700 cm^{-1} [17].

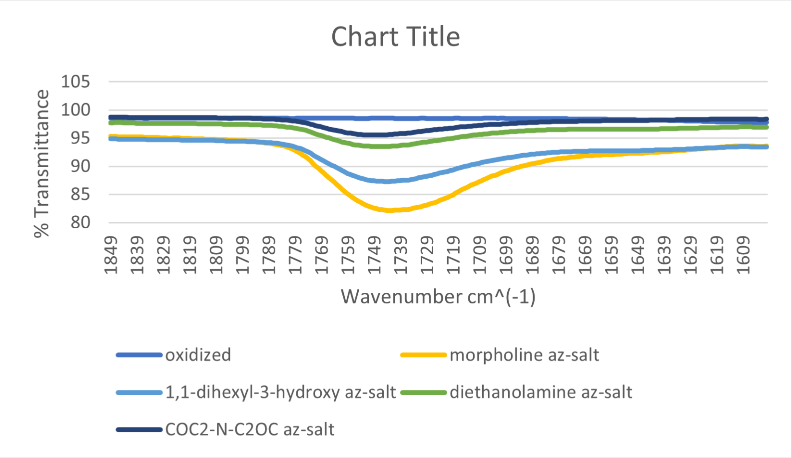

Figure 4.8: *Part of FT-IR spectrum for azetidinium salts conjugated to cellulose*

From figure 4.8 it can be determined that esters probably have been formed for conjugation with all four salts. The number of esters varies notably from the different salts. It can be seen that conjugation with morpholine az-salt generated the most chemical bonds at around 1700 cm^{-1}. From this spectrum it cannot be fully determined that it is only ester bonds that have been formed since other bonds also occur around the same wavenumber. To be absolutely sure that there are ester bonds and that the azetidinium salts have conjugated correctly further test would be needed.

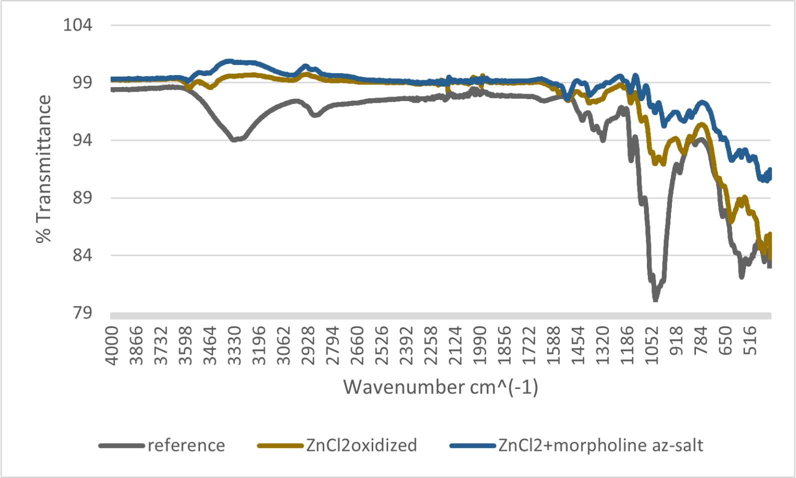

Figure 4.9: *FT-IR spectrum for* $ZnCl_2$ *swelled cellulose*

An analysis of the $ZnCl_2$ swelled cellulose was also done. Since this was done at an early stage only morpholine azetidinium salt was conjugated onto the oxidized cellulose for this. So, in figure 4.9 the full spectrum for a reference sample, a sample of oxidized cellulose and a sample from the attempt to conjugate salt onto it is shown. Here it can be seen that something has happened with the cellulose both in the oxidation step and in the conjugation step. But when looking at the part of the spectrum where the ester peaks are when cellulose has been conjugated onto water swelled oxidized cellulose there are no peaks for the conjugation with $ZnCl_2$ swelled cellulose. This part of the spectrum can be seen in figure 4.10. From the IR spectrum it cannot be determined exactly what happened when the azetidinium salt was conjugated to the cellulose, but the desired reaction has not occurred.

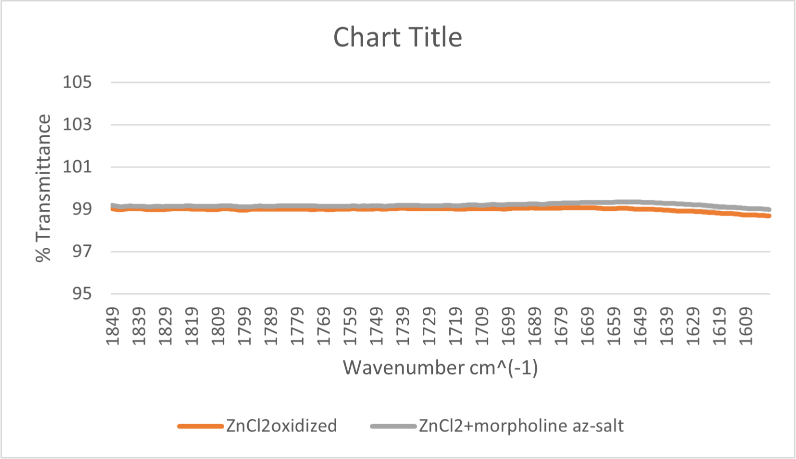

Figure 4.10: *Part of FT-IR spectrum for* $ZnCL_2$ *swelled cellulose*

4.3 Degree of oxidation

In this section the result from the determination of degree of oxidation is presented. For all the data used in the calculations and the results of these calculations see table A.1 and A.2 in the appendix. In table 4.1 the results for the mean value of the carboxyl group content and the mean degree of oxidation are presented for the two different oxidation times and the reference.

Table 4.1: Mean value for carboxyl group content and degree of oxidation

Sample	Carboxyl group content (mmol/g)	Degree of oxidation (%)
24 h	3.183	23.33
2 week	4.394	32.25
Reference	1.205	8.88

4.4 Water retention value

In table 4.2 m(wet), m(dry) and the water retention value for all the samples is presented. Samples 1-3 used morpholine AZ-salt, 4-6 used 1,1-dihexyl-3-hydroxy AZ-salt, 7-9 used diethanol amine AZ-salt and 10-12 used COC2-N-C2OC AZ-salt.

Table 4.2: Water retention value data

Sample	m(wet) (g)	m(dry) (g)	WRV (g/g)
1	0.4441	0.0232	18.142
2	0.4270	0.0084	49.833
3	0.4327	0.0460	8.407
4	0.4574	0.0385	10.881
5	0.4101	0.0431	8.515
6	0.3774	0.0421	7.964
7	0.3364	0.0281	10.972
8	0.3375	0.0126	25.786
9	0.3234	0.0189	16.11
10	0.3673	0.0257	13.292
11	0.3996	0.0296	12.5
12	0.3467	0.0252	12.758

As seen the water retention value differs depending on which type of salt was used. This means that the salts affect the cellulose differently depending on the structure of the salt. Due to the big fluctuation for some of the salts no mean value was calculated since that could give a misleading picture. So the conjugation with azetidinium salt do change the water retention value depending on the structure and therefore different salt should be used for different desired outcomes.

5

Conclusion and discussion

As seen in figure 4.8 all four different azetidinium salt has conjugated to the cellulose. Even if it has happened in different amounts, it has still happened. Therefore, the method for conjugating azetidinium salts to cellulose is successful for most azetidinium salts. This is just a part of what was studied in this thesis, but it was not the main goal. To be absloutely sure that there is esters that have formed another another molecular form that also has a peak around 1700 cm^{-1} further testing could have been done. Now it is assumed that the reaction has gone as planned.

However the main goal was to see how different structure of azetidinium salts affects cellulose. For that the biggest focus was on how the water retention value changed with different salts. For morpholine cellulose the water retention value varied quite a lot, and this can be because of the structure. Since the structure is quite small and does not have any long side chains it might be that the effect of the salt only affects a small area of the cellulose directly where it has conjugated. Also, for diethanolamine azetidinium salt the water retention value varies. This can also be because of the structure. It has longer horizontal coal chains from the nitrogen molecule than the morponline azetidinium salt, but it is shorter than what the other two salts have.

The lowest water retention value was obtained by conjugating 1,1-dihexyl-3-hydroxy azetidinium salt to the cellulose. This was also the salt where the horizontal coal chains from the nitrogen molecule were the longest. So, from that one can assume that with longer coal chains on the salt comes lower water retention value. To see if this assumption is correct can conjugation with even larger azetidinium salts be tried to see how that affects the water retention value.

When it comes to how the salts conjugated it depends on the oxidation of the cellulose. Since the degree of oxidation is rather low it might be that the azetidinium salts only conjugate to small parts of the cellulose or with a great distance between each other. Therefore, an attempt to see how time affects the degree of oxidation was implemented. As seen from the degree of oxidation, 2 weeks gave a slightly higher degree of oxidation but probably not enough to make any huge difference for the conjugation. So the conclusion is that the cellulose has oxidized after 24 hours but to a rather low degree. To be able to modify the cellulose better with azetidinium salts a higher degree of oxidation is desired. After two weeks the carboxyl group content is higher than after 24 hours, but the difference is not so high that the extra time is worth the result. Since the degree of oxidation only increased by 10 % over the 13 extra days it can be determined that most of the oxidation takes place over the first 24 hours. The reference sample has not gone through an oxidation reaction, but it still has some degree of oxidation. This can be due to how it was treated by the manufacturer before coming to Chalmers. It has been treated in some way since it has had to be extracted from plant matter. Since BKP has been used the cellulose has also been bleached so something in that treatment could also be behind the carboxyl group content. The reason cannot be confirmed here instead the whole process of the cellulose from the plant or tree until it arrives at Chalmers would have to be monitored and tested after the different treatments.

An attempt to try a different method of swelling was also performed. In that case $ZnCl_2$ was used in the swelling step instead of just deionized water. The result after the swelling was a white slurry instead of a more see through slurry when water was used. After oxidation there was cellulose that was more rubbery in consistency. Though after an attempt to conjugate azetidinium salt to the cellulose nothing appears to have happened, at least not in the same way as for just water swelled cellulose. There had been no ester bond formed that should have shown up at 1700 cm^{-1}. In future work it should be tried to wash the cellulose after the swelling step when $ZnCl_2$ is used. Cause it might be that the $ZnCl_2$ interfered with both the oxidation and the conjugation and lead to different reactions than the intended ones. So, a washing step might be a good idea to get rid of the excess $ZnCl_2$ molecules. Since it seems like the cellulose had swelled more according to looks it might lead to a better degree of oxidation when non reacted $ZnCl_2$ molecules do not interfere. To be absolutely sure that there are ester bonds and that the azetidinium salts have conjugated correctly further test would be needed.

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A

Appendix

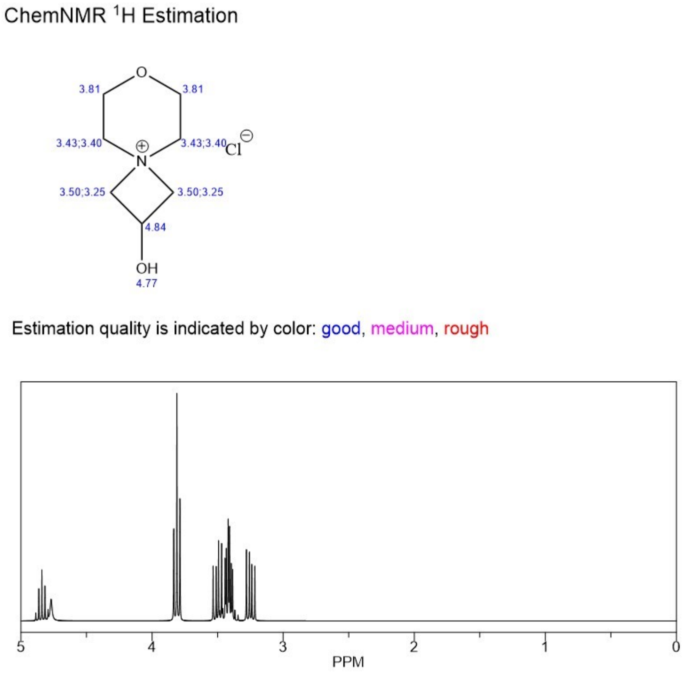

Figure A.1: *Predicted NMR spectrum for morpholine az-salt*

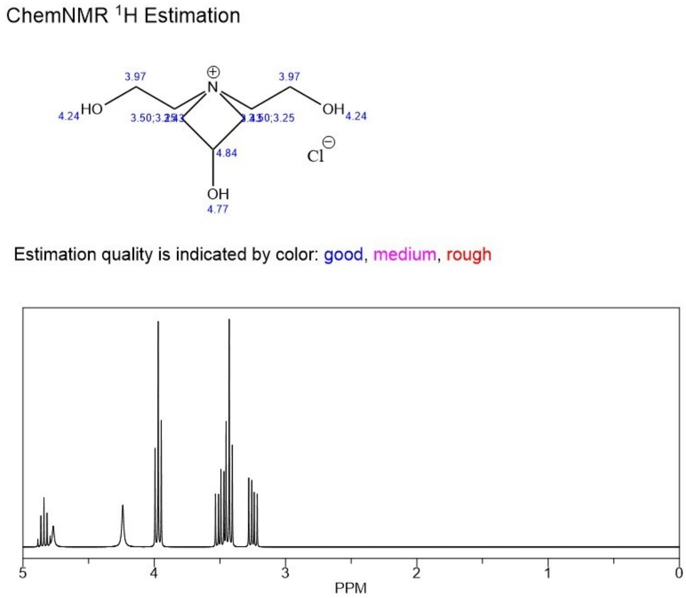

Figure A.2: *Predicted NMR spectrum for diethanolamine az-salt*

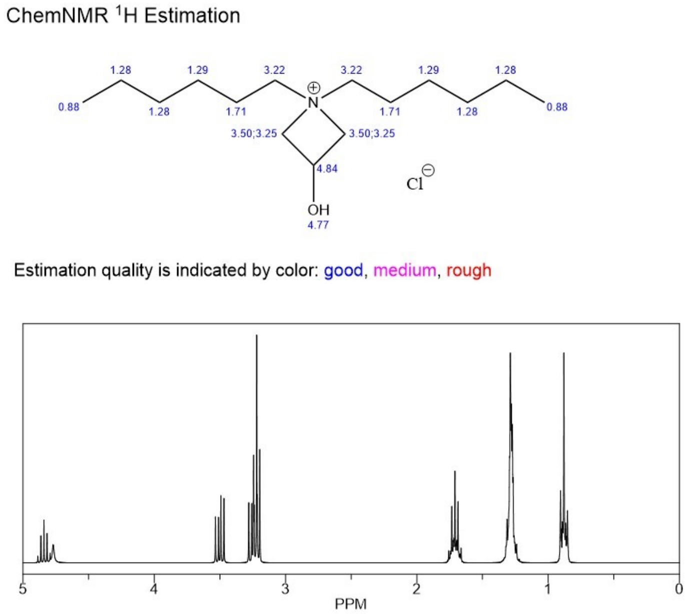

Figure A.3: *Predicted NMR spectrum for 1,1-dihexyl-3-hydroxy az-salt*

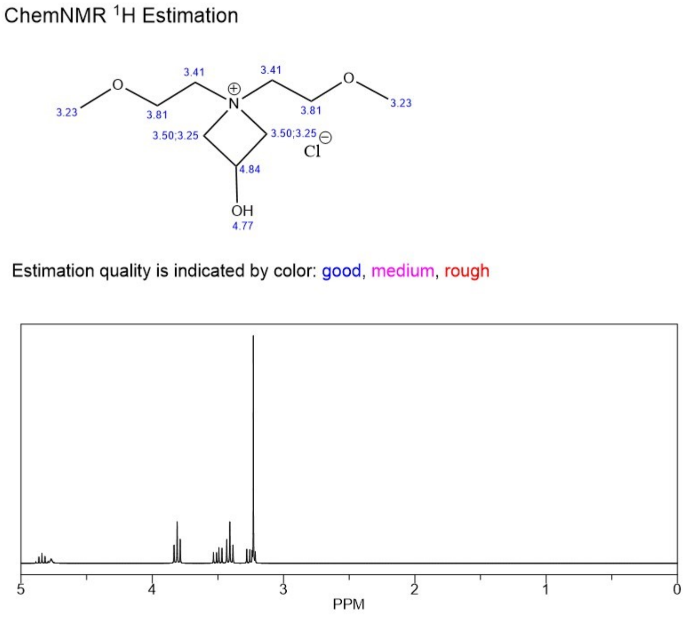

Figure A.4: *Predicted NMR spectrum for* COC_2*-N-*C_2OC *az-salt*

Table A.1: All data for degree of oxidation and carboxyl group determination

Sample	volume NaOH (ml)	pH	ERC	wet weight cellulose (g)	dry weight cellulose (g)	moles NaOH	HCl added (ml)
24h1	2.1805	7.256	69.8	0.0209	0.00666919	0.021805	0.02
24h2	2.1205	7.573	66.4	0.0208	0.00663728	0.21205	0.02
24h3	2.0080	6.486	69.5	0.0204	0.00650964	0.020080	0.02
2week1	2.1550	7.804	60.1	0.0200	0.004616	0.021550	0.02
2week2	2.0520	7.935	66.2	0.0207	0.00477756	0.020520	0.02
2week3	2.0120	7.915	65.4	0.0204	0.00470832	0.020120	0.02
Reference1	2.3875	7.869	68.6	0.0201	0.01791312	0.023875	0.025
Reference2	2.0685	7.976	66.8	0.0201	0.01791312	0.020685	0.022
Reference3	2.0275	7.705	63.6	0.0202	0.01800224	0.020275	0.018

Table A.2: Degree of oxidation and carboxyl group content for all samples

Sample	Carboxyl group content (mmol/g)	Degree of oxidation (%)
24h1	3.269	23.88
24h2	3.195	23.40
24h3	3.085	22.71
2week1	4.669	34.13
2week2	4.239	31.17
2week3	4.273	31.45
Reference1	1.333	9.64
Reference2	1.155	8.48
Reference3	1.126	8.53

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