Manufacturing of Biopolymers

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2022/2023

The title page picture shows successfully synthesized galalith pieces in flower shape. All originated from the second method (**5.5.2**).

Darke green – Procedure 1

Light orange – Procedure 2

Light green – Procedure 3

Yellow – Procedure 5, 6

White – Procedure 7

Table of Contents

1 Preface

From the beginning I knew that I wanted to write my Matura paper in chemistry because I enjoyed going to the laboratory and doing research and experiments. When my family bought new plates which claimed to be fully biodegradable but were still dishwasher proof my interest was risen. I wanted to figure out whether I would be able to manufacture a biopolymer on my own.

Prior to starting with the paper, I would like to thank everyone who helped me write my Matura paper. I would particularly like to thank Martina Zürcher for mentoring me patiently and passionately throughout the whole process of experimenting and writing my Matura paper. Her information, inputs and ideas were very helpful and highly appreciated for establishing and carrying out the experiments as well as writing the paper. Furthermore, I would like to thank Stefan Küffer for reliably providing the necessary material on time. Moreover, I would like to thank family and friends for supporting me throughout this process. Special thanks go to my mother Sonja Odolo for providing additional helpful inputs as well as proofreading my Matura paper.

2 Abstract

The goal of this paper was to manufacture biopolymers in a school laboratory. Hence, three main procedures were developed.

In the first procedure a biopolymer is manufactured from corn-starch. This is achieved by mixing starch with acid whereby hydrolysis occurs, and the formerly branched polymer is transformed into linear polysaccharide chains.

In the second procedure casein was produced. Casein is commonly known as milk protein and forms when milk is mixed with acid. Two methods to obtain casein were compared. Furthermore, differences in the final product originating from raw milk and pasteurized milk were analysed.

In the third procedure galalith was manufactured. Galalith experienced great success in the early 20th century where it was used as a common substitute for ivory or to produce accessories. It is formed by crosslinking casein with formaldehyde. To obtain galalith two different experiments were carried out. Here the self-made casein was processed into galalith. The first attempt did not yield the expected results however pleasant results were obtained from the second experiment. Further experiments examined how the resulting galalith changes depending on the amounts of formaldehyde used for synthesis.

3 Introduction

Polymers or plastics are nowadays part of our daily habits and a life without them would be unimaginable. Due to their versatility and different nature, they can be used for almost anything. Ranging from takeaway containers made from polypropylene to polyester sport shirts or credit cards made from PVC. What all these elements have in common are that they are almost exclusively made from fossil fuels which are not renewable. Furthermore, their recycling process is limited since polymers are usually not biodegradable. This changes with biopolymers which are of mostly natural origin and therefore more sustainable than regular polymers.

The aim of this project was to learn about polymers especially biopolymers. This should be achieved by carrying out experiments regarding biopolymers. The main experimental goal was to figure out, whether it was possible to manufacture a biopolymer that could be compared to a regular biopolymer. This should be done in a school laboratory. During this process the properties of a polymer should be discovered and whether a polymers characteristics change with altered starting conditions.

4 Theory

4.1 Definitions and Classification

What is commonly referred to as plastic is scientifically called polymer. This term is derived from the Greek word ‘poly’ which means many and the term ‘mer’ which means repeating unit. These repeating units are called monomers derived from the Greek term ‘mono’ which means one [1]. Polymers are a subset of macromolecules which are very long molecules consisting of numerous monomers connected by covalent bonds. Other examples for macromolecules are DNA and RNA as well as proteins, which are both found in the biological system [2]. Each monomer consists of the same atoms and is structured in the same way. Since polymers can become very long and therefore have an extremely high molecular weight their molecular structure is usually described by a so-called repeating unit (**Figure 1**). [3] [4]

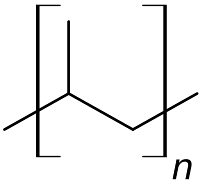
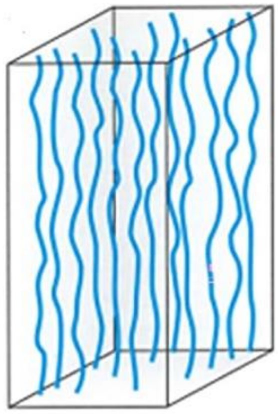


Figure 1 repeating unit of polypropylene

There are two possibilities to categorize polymers, either by their construction, which defines the typical properties such as reaction towards heat and mechanical stress, and by the way they are produced. Polymers can be classified by their properties into thermoplastics, thermosets and elastomers. Thermoplastics become soft and liquid when heated and solidify again when cooled. They are shaped in liquid state. Their structure is linear, and they are held together by intermolecular forces such as hydrogen bonds or van-der-Waals forces (**Figure 2**). A frequently used thermoplastic is polyethene which is often used for packaging and ‘plastic’ bags. [3] [5]



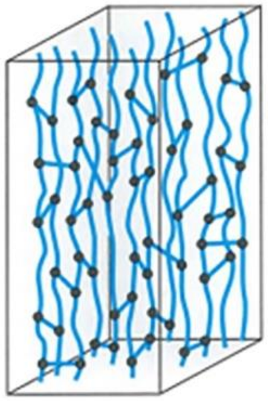


Figure 2 structure of a thermoplastic [5] (left)

Figure 3 structure of a thermosetting polymer [5] (right)

Thermosetting polymers can only be heated once and afterwards the structure is preserved even under the influence of heat. Thermosets are therefore often used when high temperatures are involved. The characteristic properties of thermosets are due to the cross-linked polymer chains which are held together by permanent covalent bonds (**Figure 3**). [6] [5]

Elastomers are the third group of polymers. They can be deformed by exterior pressure or pull but will eventually return into their original form when the stress is removed. They are lightly cross-linked polymers, and their structure can rearrange by rotation of the covalent bonds in the polymer chain. When the stress or heat factor is too high the structure is ruined and the polymer breaks (**Figure 4**). [3] [7]

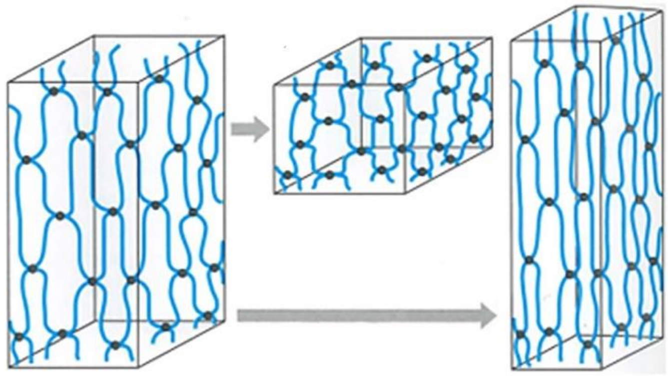


Figure 4 structure of an elastomer in relaxed, condensed and stretched state [5]

The second option of classification is in the way the polymers are produced and consists of the following three major production mechanisms.

The first method is called polymerisation. In this process the same type of unsaturated molecules form a chain. This happens through the opening of double- or triple bonds within the molecule to form a new covalent bond with a neighbouring monomer molecule. [6] The second option to form a polymer is through polycondensation. Thereby two molecules are connected to one another by the elimination of a small molecule, usually water. For this process to take place the molecules must own suitable functional groups which can be connected to one another by the condensation process. When the eliminated molecule is added to the polycondensate hydrolysis occurs and the polymer is split up into its reactants. Polyaddition works in a similar way but there are no molecules eliminated when forming the polymer. The atoms within the connecting molecules are just moved. Similar to the polycondensation, two functional groups are required, for the addition reaction to take place. [3] [4] [6]

4.2 Biopolymers

Polymers are currently used every day to fulfil various needs. Most of these polymers are from fossil origin and therefore not renewable since the supply of fossil fuels is limited. It is important to manufacture alternatives since otherwise it is impossible to continue producing enough synthetic material to keep up with the demands of a modern world. Biopolymers are therefore an ideal alternative. Biopolymers consist of exclusively or mainly biobased ingredients which are renewable and therefore not limited. Renewable primary products are organic raw materials, usually plant based. In the case of natural polymers, they are synthesized by a living organism. [8] [9]

Examples for natural polymers are polysaccharides as well as proteins. The most common polysaccharide is cellulose which is the principal component of plant’s cell membrane and consists of linear glucose chains. Starch is another polysaccharide that is commonly used to manufacture biopolymers. Casein is the most common protein used to manufacture biopolymers. An example for a natural elastomer is rubber. It consists of the latex of Hevea brasiliensis and is used for rubber products [10]. Even though there are synthetic alternatives 40 % of the rubber demands are covered by natural materials. [8] [9]

4.3 Starch

Corn-starch is a polymeric carbohydrate. It consists of polysaccharides which are long chains of the monosaccharide glucose linked together by covalent bonds. Polysaccharides are used by plants as an energy storage as well as structural material especially in the cell membrane. Corn-starch consists to 75 % of the polysaccharide amylopectin as well as to 25 % amylose (**Figure 5**). Amylopectin is a branched chain of glucose molecules while amylose is a linear chain of glucose molecules. Corn-starch is obtained by extracting the starch from the endosperm of the kernel. [9] [11] [12]

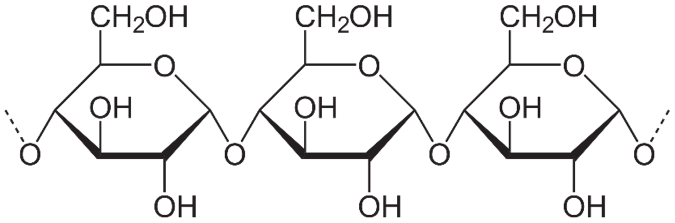


Figure 5 extract of an amylose polysaccharide [12]

Due to the cyclic structure, that is held together by stable hydrogen bonds, starch is very heat resistant and only soluble in hot water. The starch granules absorb the water which causes them to swell and burst open. As a result, this causes the semi-crystalline structures to break. The amylose and amylopectin gradually dissolve in the water and form a thick and viscous solution. This process is called gelatinization and makes starch a versatile ingredient in recipes since it thickens sauces with this procedure. For industrial usage starch polymers are often chemically modified with processes such as esterification and oxidation. Starch polymers are among the most abundant polymers due to their low-costs, renewability, and complete biodegradability. Furthermore, they are common in different plants such as corn, wheat, manioc or potatoes. This makes them easily accessible all over the world. [9] [13]

4.4 Casein

Casein is a protein obtained from milk. It makes up 80 % of the milk proteins, the remaining 20 % being in the whey. The amount of casein in milk depends on the animal’s age as well as its breed. Generally, it is between 24 – 29 $g \cdot L^{-1}$. Due to its high amount of existential amino acids casein is often used in protein supplements or as an additional ingredient in dairy products. In the industry it is commonly used as a binder for paint or as a glue. Casein consists of long protein chains which in turn consist of numerous amino acids. Due to the large size of the protein chains casein can form very stable intermolecular forces. [14]

Casein is gained by either adding acids to create acid casein mostly for industrial use or by adding rennet to the milk to form rennet casein. When heated, the milk flocculates and separates the casein and the whey. The casein can then be further processed into cheese, or it is washed and dried for industrial use. [14] [15]

4.5 Galalith

Through an accidental experiment the French chemist Auguste Trillat discovered that casein could be laid into a formaldehyde solution which turned it into a solid object. Through this reaction water is eliminated and the casein molecules become covalently bonded. Wilhelm Kirsche and Adolf Spitteler further developed the experiment and patented the new discovery as ‘Galalith’. [16] [17]

The discovery can be explained chemically as follows: When the casein is placed in a formaldehyde solution a polycondensation takes place. The amino groups of the casein protein react with formaldehyde. In this reaction the casein is covalently cross-linked, water is eliminated and galalith is formed (**Figure 6**). [16] [17] [18]

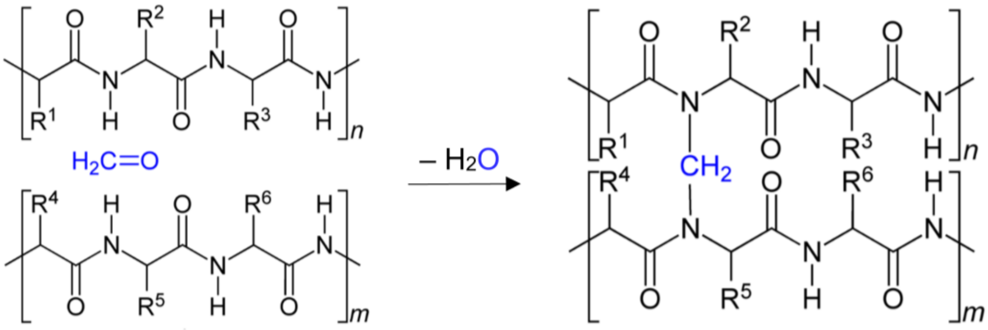


Figure 6 polycondensation of casein to form galalith, edited from [18]

Already in the late 19th century jewellery or accessories were made from galalith (**Figure 7**). It is a hard and shiny material that was used as a substitute for the expensive ivory. Due to its cost efficiency and easy production, it was widespread and very popular. Nevertheless, galalith is sensitive towards humidity and prone to breaking. After World War 2 it lost importance since new even more affordable synthetic polymers were developed without the negative side effects of galalith. Today galalith is only used very rarely and almost is completely substituted with synthetic polymers. [16] [19]

Figure 7 galalith colouring samples [17]

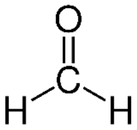
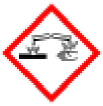
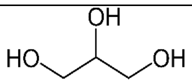
5 Experimental Procedure

5.1 General Information

All chemicals and laboratory equipment were provided by the chemistry department of the Kantonsschule Zug. The chemicals are from the manufacturers Fluka, Interwega and Carl Roth. Food colourings were obtained from Werner Schweizer AG and Dr. Oetker.
Safety goggles as well as a lab coat were always worn when experimenting in the laboratory. All experiments were performed in the fume hood and chemical residues were disposed of properly. If special safety measures had to be taken this is noted.

The H and P codes can be found on the website of the United Nations Economic Commission for Europe. [20]

Table 1

Name and Formula	**Structure**	**Dangers**	**Information**
Formaldehyde, Methanal, CH_2O	 ***Figure 8*** *formaldehyde molecule [21]*	Hazard statements: H301+H311+H331-H314-H317-H335-H341-H350-H370 Precautionary statements: P260+P280+P303+P361+P353+P304+P340+P305+P351+P338+P308+P311 [22]  [23]	Formaldehyde is in pure state a gaseous chemical. The utilized 37 % solution is a clear liquid. It is suspected of causing cancer which is why safety measures must always be followed. All the experiments were executed in the fume hood to minimize toxic damps. Goggles, a lab coat as well as latex gloves were always worn when experimenting. [22] Molar mass = 30.03 g · mol^{-1}
32% aqueous sodium hydroxide solution, NaOH (32 %) 10% aqueous sodium hydroxide solution, NaOH (10 %)		Hazard statements: H290-H314 Precautionary statements: P280+P301+P330+P331+P303+P361+P353+P305+P351+P338+P310+P406 [24]  [23]	Sodium hydroxide solution is a clear liquid. Due to the high concentration, it can cause chemical burn when coming into contact with skin or eyes. Use appropriate protective equipment. [24] Molar mass = 40.01 g · mol^{-1}
Glycerol, $C_3H_8O_3$ 1,2,3-Propantriol	 ***Figure 9*** *glycerol molecule [25]*	-	Glycerol is a clear and vicious liquid without any safety measurements. [26] Molar mass = 92.09 g · mol^{-1}

5.2 Experiment 1: Manufacturing of Casein – Method 1

5.2.1 Chemicals and Equipment

Pasteurised cow milk (3.9 % fat, Coop, Naturaplan)
Raw cow milk (Rüschenhof Zug)
White wine vinegar (MClassic, Migros, 4.5 %)
Food colourings “Rot” and “Violett” (liquid, Dr. Oetker)
1000 mL beaker
Hot plate and pot (*d* =18 cm)
Oven
Porcelain plate (*d* =20 cm)
Teaspoon (tsp.)
Plastic spoon
Funnel
Woven cloth
Clothespins

5.2.2 Experimental Procedure

Milk (500 mL) was poured into a 1000 mL beaker and reinfused into a pot (*d* = 18 cm). Ten drops of red food colouring were added, and the mixture was stirred well with a spoon. The pot was put on a hot plate, heated to just before boiling and removed from the hot plate. White wine vinegar (2 tsp.) was added and stirred, thereby the milk started to flocculate. The pot was put into a cold-water bath to let the casein cool down, while the mixture was stirred with the spoon. Two layers of woven cloth were fixed with clothespins to a funnel positioned on top of a 1000 mL beaker.

As soon as the mixture had cooled to RT, it was filtered through the funnel and the cloths. After most of the whey had been strained through the cloths, the casein-cake was further pressed out by hand. Thereby, both the whey and the casein were pressed through the loose cloths. Not wanting to lose all the casein by pressing it through the cloth, the casein was scraped from the cloth with a spoon and put on a plate. The casein was formed to a rectangular shape and spread out to a hight of 4 mm (**Figure 10**).

Figure 10 *casein from pasteurised milk before drying*

The consistency of the resulting casein was liquid and jellylike, but it was a compact mass with no loose crumbs or difficulty to press the mass together. The casein was oven-dried for 55 min at 50 °C and 55 min at 70 °C. Removing the dried product from the plate was very hard. It had to be scratched away with a knife, however, doing so holes in the plate formed. On the downside not the whole plate had completely dried, so the casein was further air-dried over night at RT.

The experimental procedure was repeated with raw cow milk and purple food colouring. Even though the same amounts of ingredients were used in this experiment, a lot less product resulted. With raw milk 5.9 g product were obtained whereas with pasteurized milk 8 g were obtained. Before baking, the mixture was a lot drier than that in the first experiment. Small holes and creases remained after pressing the casein by hand (**Figure 11**). The consistency was a lot less liquid but felt crumblier; the casein pieces did not stick well to each other. The casein was oven-dried for 1 h at 50 °C and 1.5 h at 50 °C with circulating air. Afterwards it was air-dried over night at RT.

Figure 11 *casein from raw milk before drying*

5.2.3 Results

Pasteurized milk:
After baking the casein for 55 min at 50 °C the casein had hardened but was not solid yet. The surface could not be touched without leaving a deformation on the casein. The outer corners of the product had darkened in colour (**Figure 12**). After oven-drying for a second time for 75 min at 50 °C the surface of the casein had hardened so it could be touched. The complete surface of the plate was smooth and had darkened in colour. On the downside which had not completely hardened yet light patches were still visible.

__Figure 12__ casein after drying in the oven

When the casein had air-dried overnight it was a solid, hard object. It could be broken into pieces, but force had to be applied. On the edges smaller pieces crumbled and there were cracks on the sides. The three holes in the middle of the plate had increased in size and on one side the plate curled in. Small holes from air bubbles were visible on the surface. The casein plate had now completely darkened in colour except for the rims around the holes (**Figure 13**). The thickness of the plate had drastically decreased to less than 1.5 mm varying throughout the piece. When touching the plate, it felt oily and it smelled like spoilt milk.

__Figure 13__ casein after air-drying

Raw Milk:

After 1 h in the oven there was a light crust on the surface but when pressing on the crust it cracked. The colour had darkened to a darker purple.

When removing it from the oven the surface had completely hardened. There were no large cracks visible but for the one that had already existed before baking. The colour had darkened a lot, especially the sides had a dark rim. On the underside the plate had almost not dried and was still pale purple except for the dark rim. When touching the underside, it was soft and wet and there was no crust.

After letting the casein air-dry overnight there was a crust on both sides. The object felt solid and did not break nor did any small parts crumble. The small holes and creases from the casein were visible but besides that it was a smooth surface. The surface was oily and smelled like spoilt milk, too. The thickness had decreased to 2–3 mm. The rim of the plate had a darker colour (**Figure 14**).

__Figure 14__ casein after air-drying

5.3 Experiment 2: Manufacturing of Casein – Method 2

5.3.1 Chemicals and Equipment

Cow milk (3.9 % fat, Coop, Naturaplan)
Raw cow milk (Rüschenhof Zug)
White wine vinegar (MClassic, Migros, 4.5 %)
Food colourings “Zitronengelb” (Werner Schweizer AG) and “Blau” (liquid, Dr.Oetker)
500 mL and 25 mL graduated cylinder
Hot plate and pot (d =18 cm)
Strainer (d =16 cm)
Plastic spoon
Latex gloves
Baking tray
Aluminium foil
Paper towel
Oven

5.3.2 Experimental Procedure

Milk (250 mL) was measured with a 500 mL graduated cylinder and poured into a pot (*d* = 18 cm). White wine vinegar (25 mL) was measured in a 25 mL graduated cylinder but not added to the milk yet. The pot was put on a hot plate on medium heat and the milk was continuously mixed with a spoon. As soon as the milk started to fume the temperature was reduced, the vinegar was added, and the mixture was stirred. As soon as the mixture started to flocculate the pot was removed from the hot plate. The casein flocculate was compressed, and the whey was removed through a strainer. The casein was washed with dem. H_2O until the filtrate was clear. The filtration residue was kneaded until all the liquid was squeezed out. A tiny amount of food colouring (“Zitronengelb”) was added and kneaded into the casein. A baking tray was laid out with aluminium foil, the casein was placed on it and spread out to a height of 4 mm. The casein was oven-dried for 1 h at 70 °C, then removed and air-dried in the fume hood for 6 d.

The experiment was repeated with the doubled amount and the omission of food colouring so that the product could be used for following experiments. After letting the product air-dry for 6 d, the resulting casein plate was broken into smaller pieces by hand.

The same experimental procedure was applied with raw cow milk. Blue food colouring (5 drops) was added directly to the milk. After the flocculated casein separated from the whey it felt a lot more fibrous. It was neither very stretchy, nor did it break directly when force was applied. Pressing out the whey was easiest compared to the previous experiments. The mass was compact and did not have to be forcefully pressed together. The more whey was pressed out the less stretchy and plastic it became. As a result, the casein could not be formed into a rectangular shape for drying. After the casein was oven-dried for 40 min at 50 °C, it was still very soft, and whey had pressed out. It was patted dry with a paper towel. The casein was oven-dried for additional 55 min at 50 °C with circulating air. Afterwards it was air-dried overnight.

5.3.3 Results

Pasteurised milk:
After drying, the casein was a slightly oily solid plate (**Figure 15**). Removing it from the aluminium foil was easy, it did not stick to the foil at all. The structure felt crumbly, but it did not break and stayed compact. On the surface, cracks as well as some holes were visible on the underside of the casein plate. The colour stayed the same and did not change during the baking process.

Figure 15 casein after air-drying

Raw Milk:
After baking the casein for the first time there were not a lot of changes (**Figure 16**). On the surface the fibrous texture of the casein was well visible whereas on the underside holes from air enclosures were visible. These were filled with the emitted whey. There was no change in colour or shape and the casein still felt soft.

Figure 16 casein after baking for 40 min

After baking it for the second time and after air-drying, it had hardened (**Figure 17**). It was a solid but still flexible object. Where there were air enclosures before, there were now holes. It did not crumble nor break but it was flexible to a certain point. There were no cracks, but the fibrous texture was still visible. The colour did not darken but it was unevenly transparent. The plate felt oily but the smell was not as unpalatable as in the preceding experiments.

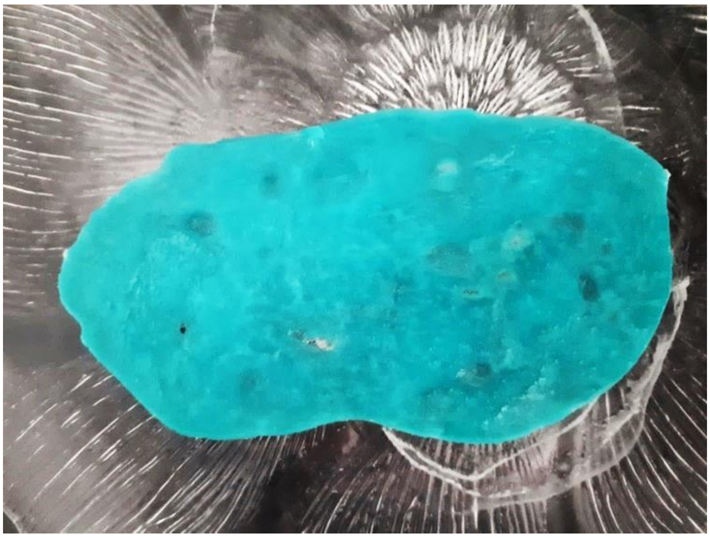


***Figure 17** casein after air-drying*

5.4 Experiment 3: Manufacturing of Galalith – Method 1

5.4.1 Chemicals and Equipment

Casein from bovine milk (Fluka)
Casein (self-made, Experiment 2: Manufacturing of Casein – Method 2)
10 % aqueous sodium hydroxide solution, NaOH (10 %)
Formaldehyde solution 37 %, CH_2O
600 mL beaker
Hot plate
Glass rod
Test tube
Test tube stand
Test tube holder
3 mL pipettes
Watch glass (*d* = 12 cm)
Aluminium foil

5.4.2 Experimental Procedure

Casein (3 g) was weighted into a test tube. NaOH (3 mL, 10 %) was pipetted and added to the casein. The mixture was stirred with a glass rod and the test tube was clamped to a stand. The 600 mL beaker was filled with H_2O, set on a hot plate on high heat and when the H_2O started to boil, the test tube was lowered about 5 cm into the H_2O (**Figure 18**).

Figure 18 *setup of the galalith synthesis*

The casein mixture was continuously stirred with a glass rod, but no changes occurred. After 5 min the lower part of the casein in the test tube started turning into a darker orange colour. After additional 5 min the H_2O in the beaker started turning orange. After a total of 15 min the test tube was removed from the boiling H_2O (**Figure 19**).

CH_2O (5 mL) was pipetted into the test tube and mixed well. No reaction occurred. Aluminium foil was fixed over a watch glass and the casein was scraped out of the test tube. On the watch glass the casein was still very crumbly. Individual pieces sticked to each other but they could not be shaped by hand. The raw product was air-dried for 7 d.

The experiment was executed a 2rd time with self-made casein that was broken into small pieces ($d \approx 3$ mm). These casein pieces never dissolved while heated.

Figure 19 casein before adding formaldehyde

5.4.3 Results

Fluka casein:

After air-drying for 7 d in the fume hood, the galalith had hardened and still had a crumbly texture (**Figure 20**). The individual crumbs were hard and very small. They did not break when force was applied by hand. They did not stick together and could not be moulded. The larger pieces ($d \approx 3$ mm) had taken on a daker colour. The remaining crumbs were still pale-yellow in colour.

Figure 20 dried galalith crumbs on watch glass

Own casein:

After the test tube was heated in the water bath a colour change of the casein occurred. Originally it was an eggshell colour which turned to a bright yellow followed by a light orange and finally brown. Additionally, an increase of liquid in the test tube was noticeable. When setting the test tube in the water there was almost no liquid in the test tube, whereas after 15 min there should have been enough liquid for the casein to dissolve. On the watch glass the casein pieces were well visible (**Figure 21**). A thin layer of brown liquid had formed around them. The colour of the mixture was brown and dark orange, and the liquid was non-transparent. The casein pieces were a lighter yellowish colour. Even though there were two different structures on the watch glass it seemed as if they were connected and some of the casein had dissolved.

__Figure 21__ galalith with self-made casein on watch glass (left)

__Figure 22__ galalith after drying for 7 d (right)

After air-drying for 7 d in the fume hood, the galalith had hardened. The casein pieces were still visible but the liquid around them had dried out. The colour had faded but was still orange. Darker circles were noticeable around the casein pieces (**Figure 22**). The part with the casein crumbs could easily be removed from the aluminium foil. The rest was harder because the galalith layer was too thin and broke or could not be removed at all. The larger pieces felt solid and did not break easily whereas the thinner galalith layer broke more easily.

5.5 Experiment 4: Manufacturing of Galalith – Method 2

5.5.1 General Experimental Setup

5.5.1.1 Chemicals and Equipment

Formaldehyde solution 37 %, CH_2O
32 % aqueous sodium hydroxide solution, NaOH
Casein from bovine milk (Fluka)
Casein (self-made, Experiment 2: Manufacturing of Casein – Method 2)
Food colourings “Blaugrün”, “Zitronengelb”, “Blattgrün” and “violet rötlich” (Werner Schweizer AG)
100 mL, 250 mL and 400 mL beaker
10 mL and 25 mL graduated cylinder
Glass rod
Pipettes
Electric cutter
Silicone moulds (d = 25 mm)
Spoon spatula
Latex gloves
Tea leaf strainer

5.5.1.2 Experimental Procedure

Procedure 1

Due to the hazardous properties of formaldehyde, it was decided to first analyse whether the experiment works with an aqueous 3 % CH_2O solution. Therefore, CH_2O (16 g, 37 %) was measured in a 250 mL beaker and H_2O was added until a total mass of 200 g was reached.

Casein (10 g, Fluka) was put in a 100 mL beaker. Dem. H_2O (3 mL) and NaOH (1 mL, 32 %) were added. The mixture was stirred with a glass rod and set aside in the fume hood for 2 h. The mixture looked a lot drier afterwards and felt very crumbly (**Figure 23**).

Figure 23 *casein before additional liquid*

Additional H_2O (1 mL) was added and mixed into the casein. The mixture still felt crumbly and not compact enough to form a solid object therefore additional H_2O (1 mL) was added. A portion of the casein was put in a pyramidal silicone mould. The casein was pressed in as firmly as possible by hand, but when removing, parts crumbled, and the piece did not remain compact. The formed casein was moved to a 400 mL beaker. NaOH (4 mL, 32 %) was added to the remaining casein (10 g) and mixed with a glass rod. The resulting mixture felt more compact and less crumbly and was moulded in a pyramidal silicone mould. The casein was pressed in firmly by hand and removed from the mould. There was almost no crumbling, but when putting it into the 400 mL beaker to the other casein piece it parted in two and crumbled (**Figure 24**).

__Figure 24__ casein pieces seen from top

The previously prepared CH_2O solution (3 %) was carefully poured into the 400 mL beaker to fully cover all casein pieces (about 150 mL). About 2/3 of the moulded casein immediately crumbled away even though the CH_2O solution was not poured directly on the casein pieces. After additional 5 min all the casein crumbled away (**Figure 25**). The beaker was covered with aluminium foil.

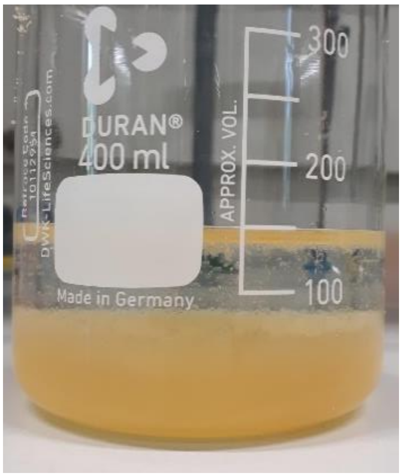


__Figure 25__ casein pieces after adding formaldehyde.

After 4 weeks the galalith had not changed a lot. The formaldehyde solution had hardly evaporated as a result it was decided to strain the liquid from the jellylike casein. The solution was separated through a tea leaf strainer on a 200 mL beaker. The wet galalith was then put into a cubic silicone mould. It was not possible to press the casein by hand because it had soaked up too much liquid. It was set aside in the fume hood for 3 weeks.

Procedure 2

Because of the unsatisfying results before straining the galalith the experiment was repeated. The initial steps in the procedure were the same as in the first experiment. However, the mixture was not put aside for 2 h before moulding. The resulting casein was moister than in the first experiment but was still too crumbly to form a compact figure. Therefore, NaOH (2 mL, 32 %) was added and mixed with a glass rod. Surprisingly, the casein was now crumblier than before. The remaining CH_2O solution (~ 75 mL, 3 %) was added to the casein mixture (16 g) and stirred with a glass rod. The consistency was very jellylike, and it seemed as if some of the casein had absorbed the liquid. There were still casein particles visible but now they were not crumbly. The mixture was put into a square silicone mould and set aside in the fume hood for 7 d.

Mixing the casein directly with the formaldehyde produced the most promising results which is why it was followed further in a series of experiments (**procedures 1–7, 5.5.2**).

5.5.2 Formaldehyde Treatment Before Moulding

Due to the hazardous properties of formaldehyde, it was decided to first analyse whether the experiment works with an aqueous 3 % CH_2O solution. Therefore, CH_2O (16 g, 37 %) was measured in a 250 mL beaker and H_2O was added until a total mass of 200 g was reached.

Following the same procedure, another aqueous formaldehyde solution (CH_2O, 8.5 %) was created.

Procedure 1
Casein (10 g, Fluka) was put into a 100 mL beaker. H_2O (3 mL) was measured in a 10 mL graduated cylinder and added to the casein. A tiny amount of food colouring ("Blaugrün") was added, and the mixture was stirred with a glass rod. NaOH (1 mL, 32 %) was added to the mixture and the casein was stirred again. The mixture felt crumbly and not as if it could hold together. CH_2O (10 mL, 3 %) was measured in a 25 mL graduated cylinder and added to the casein. The mixture was stirred with a glass rod. It still felt crumbly as if the casein absorbed all the CH_2O solution. The casein was put into silicone moulds and pressed in as tightly as possible by hand.

Because there was a lot of mixture left and there was no need to create more galalith objects the amounts were halved for the following experiments.

Procedure 2
Following procedure 1 above, casein (5 g, Fluka) was put into a 100 mL beaker. H_2O (1 mL) and NaOH (1 mL, 32 %) as well as a tiny amount of food colouring ("Zitronengelb") were added. CH_2O solution (10 mL, 3 %) was poured into the beaker and mixed well. The mixture was cohesive and lightly jellylike. It still seemed as if the casein absorbed the liquid but now the mass was no longer crumbly. The casein was put into silicone moulds and pressed into it by hand.

Procedure 3
Casein (5 g, Fluka) was put into a 100 mL beaker and H_2O (1 mL) and NaOH (1 mL, 32 %) were added. A tiny amount of food colouring ("Blattgrün") was added, and the mixture was stirred with a glass rod. The mixture was more compact than the previous ones. CH_2O solution (10 mL, 8.5 %) was added to the casein. The mixture started to bubble but stopped when stirred with a glass rod. It was the most compact mixture so far and there was almost no crumbling. The casein was put into silicone moulds and was pressed in by hand.

Procedure 4
Casein (5 g, Fluka) was put into a 100 mL beaker. NaOH (2 mL, 32 %) and H_2O (5 mL) were added. A tiny amount of food colouring ("violet rötlich") was added to the mixture and it was stirred well with a glass rod. CH_2O (3 mL, 37 %) was added but no bubbly reaction occurred. The consistency was similar to the last experiment (**Procedure 3**), but it crumbled more. As in the previous experiments, the mixture was added to silicone moulds and pressed in as firmly as possible by hand.

Figure 26 casein in silicone mould, procedure 1, procedure 2, procedure 3, procedure 4

Because the results from procedure 4 were the most promising ones it was decided to repeat the experiment with self-made casein from experiment 3 (**Experimental Procedure 5.3.2**). Therefore, the casein (13.7 g) was powdered in an electric cutter.

Procedure 5

Casein (5 g) was put into a 100 mL beaker. A tiny amount of food colouring (“Zitronengelb”) as well as H_2O (5 mL) were added and mixed with a glass rod. The mixture was a lot more liquid and creamier than with the premanufactured casein. There were almost no casein crumbs visible. The mixture had a smooth texture and was not crumbly at all. NaOH (2 mL, 32 %) was added to the mixture and a change in colour was visible. The previously yellow mixture now had an orange colour. Additionally, a jellylike thickening was observed. The mixture was now even smoother, and it seemed as if the casein had dissolved. After about 1 min the mixture had clearly hardened but still felt jellylike. CH_2O (3 mL, 37 %) was pipetted and added to the casein mixture. It did not mix nicely and there were always undissolved casein pieces left. It seemed as if the casein and the NaOH had hardened so much that they could not be mixed with the CH_2O. The mixture was homogenized as well as possible, put into silicone moulds and pressed in as firmly as possible by hand. It was the first time that a liquid was pressed out whereas the previous times the casein was just pressed to a compact block. The moulds were put in a fume hood to air-dry for 12 d.

Because the handling and the results with the self-made casein were surprisingly different from those with the premanufactured casein it was decided to repeat the experiment.

Procedure 6

The experimental procedure is the same as for the previous procedure (**procedure 5**) with the exception that a different food colouring (“Blaugrün”) was used. After adding NaOH, the mixture turned form a turquoise colour to a dark-yellow and brownish colour. This time CH_2O (3 mL, 37 %) was previously prepared to avoid the hardening through addition of NaOH. By adding the CH_2O shortly after the NaOH, the formation of clumps was successfully avoided. The resulting mixture was homogenous and creamy. Some bigger casein particles, but no clumps were visible. The mixture was put into the silicone moulds and pressed in as firmly as possible by hand. No liquid could be pressed out because the mixture was too fluid. It was put in a fume hood to air-dry for 12 d.

Procedure 7
With the remaining self-made casein (3.7 g) the experimental procedure from the previous experiments (**procedures 5, 6**) was repeated. The casein was put in a 100 mL beaker and H_2O (4 mL) was added and stirred. The mixture was creamy, and the casein partly dissolved. NaOH solution (2 mL, 32 %) was added and mixed. There was no change in colour this time. CH_2O (2 mL, 37 %) was added and stirred. The mixture felt more liquid in the previous experiments. The mixture was added to silicone moulds and pressed in as tightly as possible by hand. It was set aside in a fume hood to air-dry for 12 d.

5.5.3 Results

5.5.3.1 General Experiment Setup

Procedure 1
The casein covered in formaldehyde solution had been put aside for 4 weeks. Because the casein shapes crumbled apart when the formaldehyde solution was added no figures could be obtained. The casein mixture had not changed a lot and there were still two layers. A lower one with the casein and an upper one with just the formaldehyde solution. There was no visible change in the amount of formaldehyde. It seemed as if the casein had enclosed some of the formaldehyde solution because it was more jelly like than 4 weeks ago.

After the galalith had dried in the fume hood for 3 weeks it had significantly decreased in size (**Figure 27**). Therefore, there were no problems removing it from the mould. It had also darkened in colour except for an area on the downside of the plate. It was assumed that the plate my not have dried completely which is why it was flipped over. After additional 5 d the stain did not go away. The plate felt evenly hard no matter of the colour. The plate could not be broken in two by hand. The galalith had slightly curled and was no longer completely flat.

Figure 27 dry galalith after 3 weeks

Procedure 2
After the galalith had dried in the fume hood for 7d it had hardened. Doing so it had broken into numerous pieces with cracks (**Figure 28**). The pieces had also decreased in size since they no longer covered the complete floor of the mould. The galalith had darkened in colour with the exception for the parts on the downside. These were of the same light-yellow colour as before the drying but equally hard as the darker parts. The size of the pieces varied with the longest sides being about 2 cm. The galalith felt hard but due to the numerous creases it was not very stable and could easily be broken in two. The structure felt crumblier compared to the product of procedure 1.

Figure 28 *dried galalith pieces*

5.5.3.2 Formaldehyde treatment before moulding

Table 2 summarizes all experiments from procedures 1-7 where the formaldehyde was added to the casein before moulding.

Procedure	**1**	**2**	**3**	**4**	**5**	**6**	**7**
Casein [g] Fluka	10	5	5	5	-	-	-
Casein [g] selfmade Exp. 3	-	-	-	-	5	5	3.7
H_2O [mL]	3	1	1	5	5	5	4
NaOH, 32 % [mL]	1	1	2	2	2	2	2
CH_2O, 3 % [mL]	10	10	-	-	-	-	-
CH_2O, 8.5 % [mL]	-	-	10	-	-	-	-
CH_2O, 37 % [mL]	-	-	-	3	3	3	2
Colouring	Blau grün	Zitr. gelb	Blatt grün	Violett	Zitr.gelb	Blau grün	-

Table 2

Procedure 1

The galalith had dried in the fume hood for 8 d. The galalith had hardened to a solid object. It had decreased in size and there were no problems removing it from the moulds. There were no large cracks on the surface, but it crumbled when touched especially at the edges. There was no change in colour, but the colour was not mixed homogenously and yellow crumbs were visible where the food colouring was not mixed well.

Procedure 2

The galalith had dried in the fume hood for 8 d. There was a shrinkage in size, but the flower shape of the mould was still visible on the galalith object. The galalith did not stick to the mould but when the mould was flipped upside down to remove the galalith pieces one fell apart. On the surface of the other two pieces cracks were visible but, on the downside, they had a smooth surface. The galalith did not crumble when touched unless it was taped against a hard object. There was no change in colour visible and the colour was homogenous.

Procedure 3

The galalith had dried in the fume hood for 8 d and there was a decrease in size. It was easy to remove the galalith from the mould and it did not stick at all. It felt like a solid object and even though it had large cracks on the surface it did not break when touched gently. If wanted, it would be easy to break the pieces apart due to the cracks but there was almost no crumbling not even on the sides. On the downside it felt solid, and the colour was a lighter green compared to rest of the object.

Procedure 4

The galalith had dried in the fume hood for 8 d. Similar to the other experiments there was a decrease in size but no problems removing the galalith form the mould. There were no cracks on the surface and the galalith felt like a hard and solid object. There was no crumbling when held in one’s hand and very little when tapped against a hard object. The galalith could not be broken in two by hand unless a lot of force was applied. Due to the dark colour, there were no colour changes noticeable.

__Figure 29__ finished galalith samples, procedures 1 – 4

Procedure 5

After the galalith had dried in the fume hood for 10 d it had hardened. There were no difficulties in removing the casein from the mould but there was less shrinkage in size compared to the experiments with bought casein. On the surface casein crumbs were visible and they formed a 2 mm thin layer on top of the galalith, but no cracks were formed. On the bottom side bubbles and holes were noticeable as well as larger more uneven pieces. As a result, the casein was not as nicely shaped as the results from previous experiments. The galalith felt solid and did neither crumble nor did it break. The colour was even throughout the piece.

Procedure 6

The galalith had dried in the fume hood for 10 d. Removing the casein from the mould did not cause any problems. It had decreased in size as much as the results from procedure 5. On the surface more casein crumbs were visible but still no cracks. This layer felt crumblier and if wanted the casein crumbs could be scraped off with a metal spatula. In general, the object felt solid and did neither break nor crumbled the casein layer unless wanted. On the bottom side, the galalith was more even with just very few bubbles from air enclosures. As a result, the bottom layer of the galalith felt more compact than the upper part with casein crumbs. The colour was homogenous and there were no different structures visible.

Procedure 7

The galalith had hardened in the fume hood for 10 d and felt solid afterwards. On the top was a layer of casein crumbs. The bottom part felt solid and hard but when poked with a pointy metal stick it left a mark. The texture was homogenous as well as the colour. There was no noticeable change in colour remarkable. On the bottom side there were no holes visible, and the surfaces was smooth and completely shaped like the flower mould.

Figure 30 *finished galalith samples, procedures 5-7*

5.6 Experiment 5: Manufacturing of a Biodegradable Polymer from Starch

5.6.1 Chemicals and Equipment

Corn starch (Patissier, Migros)
White wine vinegar (MClassic, Migros, 4.5 %)
Glycerol (Interwega)
Food colouring “violet rötlich” (Werner Schweizer AG)
250 mL, 50 mL and 10 mL graduated cylinder
3 mL plastic pipettes
Hot plate and pot (*d* = 18 cm)
Plastic spoon
Whisk
Spatula
Baking tray lined with parchment paper

5.6.2 Experimental Procedure

Corn starch (30 mL) was measured in a 250 mL graduated cylinder and poured into a pot (*d* = 18 cm). H_2O (120 mL) was measured in the same graduated cylinder and added to the corn starch. White wine vinegar (10 mL) was measured into a 50 mL graduated cylinder and added to the mixture. Glycerol (10 mL) was pipetted in a 10 mL graduated cylinder and poured to the pot. The resulting suspension was stirred well with a whisk and put on a hot plate at medium heat. After 5 min the mixture became viscous, and the heat was reduced to the lowest setting. A tiny amount of food colouring (“violet rötlich”) was added. The heat was increased to medium heat, and the mixture was stirred with a spatula. After 7 min the mixture became viscous, and the sticky mass was poured onto a baking tray lined with parchment paper (**Figure 31**). The mixture was spread out to an even height of 3–4 mm and air died in the fume hood for 7 d.

Figure 31 *corn-starch polymer before air-drying*

5.6.3 Results

After 1 d the surface of the polymer had dried. It was turned around and folds and creases were noticeable on the underside. They lead to no further problems because they were not severe enough to break the polymer or to form cracks. The colour was still bright red.

After letting the polymer air-dry in the fume hood for additional 6 d it had rolled up (**Figure 32**). The sides of the polymer curled and even when cutting it into smaller pieces they individually rolled up. The colour was still bright red, but the surface was slightly oily.

The polymer was stored in an open glass bowl in a cupboard. When looking at it after 4 months the structure was still the same. When touching the polymer, it felt a lot oilier. Additionally, it stained when touched.

Figure 32 *dried starch polymer*

6 Discussion

6.1 Experiment 1: Manufacturing of Casein – Method 1

The obtained casein still contained a lot of water after it had flocculated which made its filtration tedious. Considerable amounts of the casein passed through the woven cloth due to the thick consistency of the mixture. To prevent further loss of casein the wet cake was put into the oven. The very humid cake took 2 h to settle instead of the expected 15 min. Only after overnight airdrying the cake appeared completely dry.

Overnight the casein plate curled in. This was probably due to uneven drying. This could have been overcome by flipping the plate regularly in the oven. Additionally, the holes in the casein plate had increased and the plate became crumblier. The reason for these rather drastic changes is the humidity which was still included in the raw material even after drying in the oven. The humidity in turn resulted from the troubles pressing out the casein.

The plate felt oily when it was dry. This is no surprise since milk with 3.9 % fat was used. When the milk flocculates the protein and fats in the milk mostly separate from the liquid whey and form casein. The observed oiliness of the cake proves that a major amount of the milk fat remained in the filter cake. This was further confirmed by the second casein plate where the experiment was executed with raw milk. The results from these experiments felt even oiler. This is due to the raw milk which usually contains 3.9 - 4.2 % fat which is even more than the whole milk that was used for the first experiment [27]. Additionally, the fat in pasteurized milk is homogenized this means the fat units are smaller and wider spread. This can lead to a less oily feeling compared to raw milk [28].

Strikingly, it was a lot easier to strain the casein gained from raw milk compared to the whole milk casein. When pressing the whey through the cloth almost all the casein stayed in the strainer. There were no issues pressing out the casein since the casein was thick enough so that the whey could properly be pressed out. However, the amount of casein after straining was considerably smaller. There was just about half of the product compared to the pasteurized milk even though the same amounts of milk and vinegar were used.

This unexpected observation could be due to various reasons: After pressing out the whey from the casein resulting from the raw milk there was a lot less water in the casein mixture. Whereas through the difficulty in pressing out the water from the casein obtained from pasteurized milk a lot more water remained in the casein cake. This led to an increase in volume as well as in mass of the casein product resulting from pasteurized milk. Comparing the masses of the dried products shows that the casein resulting from pasteurized milk weights about 2 g more. This accounts to +33 % compared to the casein from raw milk that weights 5.9 g.

Another assumption was that raw milk requires more acid to flocculate completely. This would suggest that the milk had not flocculated completely when the whey was filtered from the casein. This assumption could only be further investigated by another experiment adding an excess of vinegar to the milk to make sure that all the casein separated from the whey.

The third explanation for the smaller amount of obtained casein would be that raw milk generally contains less proteins. This is hardly unlikely since pasteurized milk is formed from raw milk and the protein molecules cannot form from nothing. However, when pasteurizing milk, the proteins can get partially denaturised. This means the proteins that were previously coiled up do now untie and form long chains of proteins. By doing so the structure and properties of the proteins change. Additionally, the homogenization results in smaller agglomerates of fats which makes them spread wider. Therefore, it could be followed that proteins in partly denaturised state are more likely to react with acid that in coiled up state. This is due to the increase in surface when the protein strings are not coiled up within themselves [28].

6.2 Experiment 2: Manufacturing of Casein – Method 2

The casein plate had been pressed out as firmly as possible by hand. This was a lot easier than in the previous experiment since the cake was not as thick. Due to that it was rather surprising that the drying period was still noticeably longer than the expected 30 min. Since the mass was a lot drier and more liquid was pressed out it was expected that the drying time would not be as elongated as in the previous experiment (**5.2**). This however indicates that the mass was still not dry enough. Whereupon it is wondered how the instructions [29] had pressed out the casein since theirs must have been a lot drier for a short drying period.

The separation from the whey and the casein was again easiest with raw cow milk. With little effort most of the whey could be separated from the casein by hand. Noticeable was the fibrous structure of the casein. The more whey was pressed out the less stretchy and stringier the casein became. This is different compared to the first procedure with pasteurized milk (**5.2.2**). In the first procedure the casein became crumblier and easier to mould when removing more of the whey. Whereas in the second procedure the casein became remarkably harder with practically no stretchiness the more whey was pressed out.

After the casein was oven dried it had given off liquid which was patted dry with a paper towel. This had never happened in the previous experiments. It seemed as if the casein did not “want” the extra liquid and ejected it. The casein was still considerably wet and therefore went back into the oven.

Furthermore, it is to mention that all casein plates started to form mould fungus. The first ones started just a few weeks after their synthesis but by 3 months after their manufacturing all the plates were covered in mould. This exemplifies that casein on its own is not a durable polymer. If it is intended for more durable products, it must be further processed. This is done by transforming it into galalith. If the casein is needed for industrial use or commercial products mould cannot be tolerated. Therefore, companies pay attention to the sterility of their products, so no spores are present when producing casein. The casein is stored in a dry environment since it is harder for fungus to reproduce there.

It is no surprise that the manufactured casein started to mould. All the used equipment was clean but nevertheless it is impossible not to get in touch with spores that could form on casein since they are present in air. To prevent this as good as possible the casein plates were dried in the fume hood where most of the humidity should have left the plates. Afterwards the casein plates were all stored in the same plastic container. Whenever one casein plate started to mould it was removed from the others put it was inevitable that the mould would spread within the box.

6.3 Experiment 3: Manufacturing of Galalith – Method 1

The experimental instructions [30] suggested that the casein should dissolve in aqueous sodium hydroxide solution. While carrying out the experiment it was therefore expected that this would happen. It did not: After the casein and the sodium hydroxide solution were both added to the test tube the solution immediately disappeared. The casein had soaked up the liquid. This could be expected since the casein outweighs the sodium hydroxide solution in volume. After the casein was added to the test tube it was full to about one third. When the 3 mL of aqueous sodium hydroxide solution were added, this did not contribute to additional height rather it disappeared in the cracks between the casein crumbs. The formed mixture showed a thick texture similar to wet sand. Therefore, it was difficult to reach the bottom of the test tube to stir the mixture. Due to the lack of liquid, it was impossible for the casein to dissolve when the test tube was put into the water bath because there was not enough liquid to dissolve it.

After about 5 min the lower part of the casein in the test tube gradually changed colour and turned a darker orange. At first it was believed that this was a good sign because this indicated that a reaction started to occur. Unfortunately, nothing further happened, and the mixture remained in its crumbly texture. Since there was never enough liquid for the casein to dissolve it was impossible for the reaction to fully take place.

Surprisingly the water in the water bath started turning orange which indicated that the casein somehow moved from the test tube to the water bath. Since the casein was the only orange coloured substance used in this experiment it first was believed that the casein produced the colour change in the water bath. This is impossible since casein has a melting temperature of 280 °C [31]. Furthermore, it is just very slightly soluble in water. Therefore, the orange colouring in the water bath had to originate from somewhere else. An extraction of an orange substance form the casein cake could be assumed.

The crumbly texture of the final product can be ascribed to the lack of liquid. The casein never had the chance to dissolve nor was it able to react with the formaldehyde. Therefore, the crumbly texture of the casein remained, and just small crumbs synthesized where enough sodium hydroxide and formaldehyde were available.

Since it was expected that the casein would dissolve in the sodium hydroxide solution it was not further processed and just broken into pieces so it would fit into the test tube. In retrospect it would have been smarter to grind it into powder. Analogous to the first experiment the casein did not dissolve. However, a change of colour occurred. After about 5 min the mixture turned into a light yellow. Over the next ten minutes the mixture gradually changed from yellow over orange to brown. This was unexpected since no additional colour was added to the mixture. Since the casein was the only orange coloured substance used in this experiment it first was believed that the casein produced the colour change in the water bath. This is impossible since casein has a melting temperature of 280 °C [31]. Furthermore, it is just very slightly soluble in water. Therefore, the orange colouring in the water bath had to originate from somewhere else and a reaction has taken place. This reaction is caused by heating the casein with the sodium hydroxide solution as seen when comparing to the experiment where the mixture is not heated (**5.5.2, Procedure 7**). When the temperature is increased by 10 °C a reaction is said to run at twice the initial rate. Applying this to the experiment where the casein is heated to 100 °C compared to room temperature the reaction is said to take place about 2^8 as fast compared to when not heated.

This is a significant number since it took about 15 min for the casein to turn brown. At room temperature this would have taken 64 hours which is an unreasonable amount of time. Furthermore, it is important that certain reaction require a minimal temperature to occur. This means that the reaction that caused the casein mixture to turn brown might not even have taken place at room temperature.

6.4 Experiment 4: Manufacturing of Galalith – Method 2

6.4.1 General Experimental Setup

The experimental instructions [32] suggested to first mix the casein with the sodium hydroxide solution and mould it. Followingly, the casein pieces were covered in a formaldehyde solution to transform the casein into galalith.

This procedure was not promising since a lot more liquid than suggested had to be added to the casein. With the given amount of water and sodium hydroxide solution the mixture was too dry and could not be formed into a mould without crumbling and falling apart. Originally 3 mL of water and 1 mL of sodium hydroxide were added to the casein. To make it a mouldable mixture additional 2 mL of water were added: this is +50 % of the originally added liquids. After the first shape barely held together additional 4 mL of sodium hydroxide solution were added this is +100 % of the originally added liquid and +400 % of the originally added sodium hydroxide solution. The shaping was then more successful. Disappointingly, both figures fell apart and crumbled when the formaldehyde solution was added.

At first, it was expected that the crumbling took place because the casein was not pressed into the mould hard enough and therefore was not properly moulded. However, a lot of effort was put into moulding and pressing the casein by hand. Furthermore, the shapes were removed from the mould with great caution to prevent as much crumbling as possible.

The second assumption was that letting the casein rest in the fume hood for 2 h had extracted too much liquid out of the mixture. Therefore, the experiment was repeated without letting the casein rest for 2 h in the fume hood. Unfortunately, the mixture was not wet enough either to be moulded without crumbling. Thus, 2 mL sodium hydroxide solution were added. Surprisingly the mixture was even crumblier afterwards. Therefore, it can be concluded that letting the casein rest for 2 h was not the cause for the unsatisfying result. Additional liquid had to be added to the second experiment too.

A possibility for the unsuccessful approach might be the casein itself. Looking at the casein packaging the handwritten date 16.8.06 is visible. This usually indicates the purchase or opening date. Furthermore, the packaging states that the product should be kept under argon and that it is sensitive to humidity. Looking at the product specification of casein it is stated that it should be retested after 3 y [33]. Thus, it is very well possible that in the meantime the casein had degraded and is no longer the same as it was when purchased. When executing the experiments, no reference was available and thence no suspicion about the quality of the casein came up. In retrospect it would have been a good idea to order a new casein sample and repeat the experiment for reference and examine possible differences.

Due to the unsatisfying results and the crumbling of the casein when the formaldehyde solution was added it was decided not to repeat the experiment with a higher concentrated formaldehyde solution since the experiment had already failed earlier. However, the casein was mixed with the formaldehyde solutions in procedure 2. It was expected that the same results would be obtained as in the first procedure if the casein were pressed first, and the formaldehyde added afterwards. Therefore, it seemed more promising to put the casein and formaldehyde directly into a mould.

The large amount of crumbling can be ascribed to the use of too much liquid. Thus, the mixture could not be pressed into the mould and the galalith dried without forcefully pressing it together.

Nevertheless, the results were more promising than with the casein that crumbled apart as soon as the formaldehyde solution was added. In retrospect the first procedure was moulded after the formaldehyde was added and therefore followed the same procedure. Differences being the time it was exclusively mixed with the formaldehyde solution without the intention of drying.

6.4.2 Formaldehyde Treatment Before Moulding

In each procedure three figures were created with the exception for procedure 7 where not enough casein was left to create a third. This should make sure that enough material for comparisons is available. Since all attempts were pressed by hand unequal pressing likely possible to occur. Furthermore, the number of broken pieces can provide additional information about how likely the polymer is to crumble.

Table 3 *broken pieces per procedure*

Procedure	**1**	**2**	**3**	**4**	**5**	**6**	**7**
Broken	1	2	1	0	0	0	0 (2 pieces)

Table 3 illustrates how many out of three galalith samples broke apart or crumbled completely. From these results it can be concluded that the amount of formaldehyde and sodium hydroxide have a clear influence on the stability of the polymers.

Furthermore, it is noticeable that in procedure 2 more pieces broke than in procedure 1. This even though more formaldehyde was added compared to the amount of casein. The second piece broke while taking pictures for the title page even though great attention was paid not to break a piece. The pieces from procedure 1 however did not break completely just individual casein crumbs came off. Concludingly it can be stated that both pieces are not very resistant which can be ascribed to the lower formaldehyde concentration than stated in the experimental instructions [32]. It is very well possible that the casein pieces were not pressed equally as hard into the mould since they were just pressed by hand. Therefore, the breaking in procedure 2 can be ascribed to external factors.

Moreover, it is noticeable that in procedure 3 one of the pieces broke as well. Although, the formaldehyde concentration was 8.5 % and thereby 0.5 % higher than suggested by the experimental instructions [32]. The 0.5 % are not significantly more and are easily compensated with external factors such as less firm pressing. This must have been the cause for breaking. Because the mass was already very compact before pressed into the mould not as much pressure was applied as in the previous procedures. There it deemed necessary because the casein still crumbled whereas in the third procedure the crumbling was almost non-existent when moulding. Due to that large cracks are visible on the surface of the galalith which cause one piece to break when put into a glass jar.

When the sodium hydroxide solution was added to the casein in procedure 6 a reaction occurred. The previously turquoise colour turned into a dark yellow. The same reaction was observed in procedure 5 where light orange turned into yellow. Comparing this to the results of procedure 7 where no food colouring was used it is noticeable that this is due to the food colouring. To verify this “blaugrün” food colouring was added to a test tube with a few drops of water. The liquid was a bright turquoise colour. When 3 mL of sodium hydroxide were added to the liquid it turned into the same dark yellow colour. Therefore, it can be concluded that the used food colouring reacts with basic solutions and turns yellow.

In procedure 1 where the same food colouring “blaugrün” was used surprisingly no change in colour occurred. The change of colour is therefore caused or prohibited by the casein since this is the only different ingredient. A possible explanation it that the premanufactured casein had soaked up the sodium hydroxide solution as it previously did with water. Therefore, the sodium hydroxide did not have the chance to react with the food colouring and change the colour of the mixture.

In Procedure 5 a reaction had already occurred before the formaldehyde was added. The jellylike consistency that was present when the sodium hydroxide was added had hardened. Therefore, it was difficult to stir the formaldehyde into the mixture since clumps of the casein and sodium hydroxide solution did not want to disappear. Neither a reaction nor a thickening had occurred when using the premanufactured casein which is why this was completely unexpected. Due to the surprising results the experiment was repeated in approach 6. This time special attention was paid to work quickly and therefore avoid the thickening process. With success the reaction was prohibited.

Furthermore, it can be stated from the table that the best results were obtained when the 37 % formaldehyde solution was used. If enough formaldehyde if not an access was added. Therefore, the reaction was able to fully take place. Whereas when not enough formaldehyde is added not all the casein becomes covalently crosslinked resulting in crumbling because the casein does not hold together. Furthermore, when adding a solution with high concentration less water is added to the mixture. This simplifies drying since all added water must evaporate additionally to the water formed in the condensation.

The casein becomes covalently crosslinked through hydrolysis. The amino groups in the casein and the formaldehyde form a hydrolysis and become crosslinked through methylene. During the synthesis of galalith the casein and the formaldehyde perform a condensation (**Figure 33**). This occurs by the amino groups of the casein and the formaldehyde by eliminating water. The water then evaporates and prevents the polymer to perform a hydrolysis and turn back into its reactants. Due to the cross-linked methylene chains that are formed galalith is a thermosetting polymer.

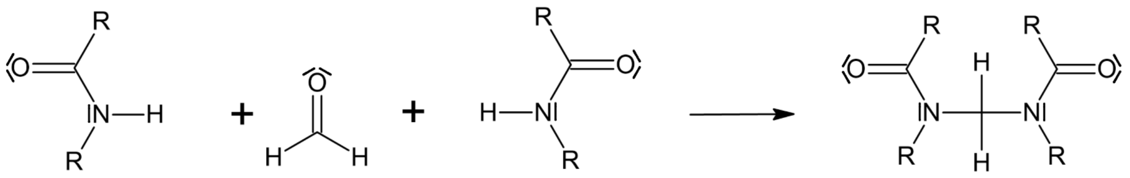


Figure 33 *condensation of galalith*

When properly synthesised formaldehyde loses its hazardous properties. Water is eliminated when the methylene bridge is formed. Nevertheless, galalith is not completely harmless. Usually an excess in product is added to ensure that the condensation fully takes place. Therefore, not all the casein is synthesized, and remnants remain in the product. Therefore, galalith is unsuitable as tableware. Even if the formaldehyde is just present in very small amounts, they can have terrible consequences.

6.5 Experiment 5: Manufacturing of a Biopolymer from Starch

Corn-starch primarily consists of amylopectin which is a branched polysaccharide. Due to the branching the long chains are caught in each other. When corn-starch is heated with acid a hydrolysis takes place. Finally, linear glucose molecules remain. The added glycerol serves as lubricant. Thus, the individual polysaccharide chains can glide past each other. Without glycerol the polymer would break and splinter. [6]

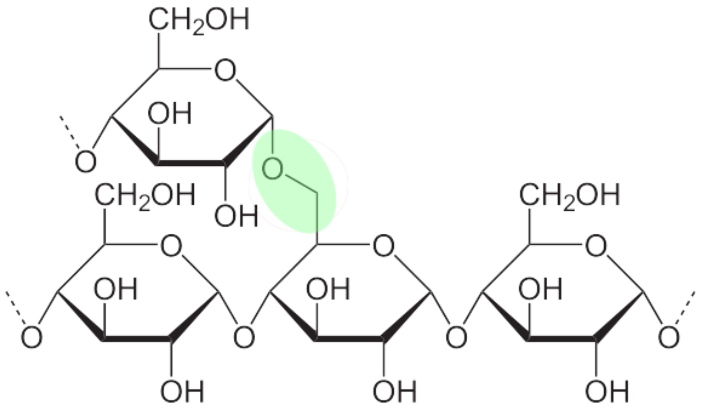


Figure 34 amylopectin molecule and highlighted bond where hydrolysis occurs, edited from [34]

The experiment was executed repeatedly with 15 mL instead of 10 mL glycerol. There were no major differences when stretching the polymer by hand. Therefore, it was decided not to include this as an additional experiment. Nevertheless, further research could be executed on how the properties change when a much larger amount of glycerol is added to corn-starch.

Apart from the ingredients which are obviously different galalith and starch polymers further wanted to be compared. The first mayor difference being the type of polymer and how they are constructed. Galalith is a thermosetting polymer and therefore heat resistant and durable this is due to the cross linkage of the polymer chains. However, a starch polymer is a thermoplastic and therefore not cross linked and held together by covalent bonds. Furthermore, galalith is gained through condensation whereas a starch polymer is gained through hydrolysis. Therefore, it can be followed that the synthesis of galalith eliminates water whereas the hydrolysis of a starch polymer requires acid. Moreover, the starch polymer is a thin and flexible plate. Up to a certain point it is elastic through the glycerol. The galalith on the other hand is a small compact product which is neither elastic nor flexible. The different properties originate from different methods to synthesize them. Furthermore, the usage of the two polymers is different. While galalith was used for buttons and accessories starch polymers are used to produce foils. [6] [16]

7 List of Abbreviations

d	day(s)
dem.	demineralized
h	hour(s)
min	minute(s)
RT	room temperature (19–23 °C)
tsp.	teaspoon
y	year(s)

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9 Table of Figures

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