The effect of Microcrystalline cellulose as cushioning excipient during controlled release

AstraZeneca

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In the pharmaceutical industry, it is always important to have reproducible processes and raw materials of high quality to ensure good quality products. AstraZeneca, that is a leading manufacturer of different pharmaceuticals, works according to GMP to make sure that their processes deliver products of the same quality every time. A problem that has occurred at AstraZeneca is when a raw material is not properly understood and variations in the raw material affects the final product. Variations in drug release in one of AstraZeneca’s products, Product X, has been connected to the cushioning excipient Microcrystalline cellulose (MCC). Drug release variations has been noticed during change from one batch of MCC to another. The aim of this study was to investigate which material attributes of MCC that contributes to variation in the final product. Particle size and moisture content were identified as critical material attributes (CMA’s) and were therefore chosen to be investigated more thoroughly. By variating particle size and moisture content during manufacturing of Product X, the influence of these attributes could be investigated using Design of Experiment (DoE). An additional experiment that compared two MCC batches from different suppliers was also performed during this study. The results from these studies showed that particle size and moisture content of MCC did affect the drug release. Larger particles and high moisture content gave rise to a faster drug release compared to small particles and low moisture content that gave rise to a slower drug release. It is however hard to draw conclusions regarding how small differences in particle size and moisture content could affect the drug release.
Abstract
In the pharmaceutical industry, it is always important to have reproducible processes and raw materials of high quality to ensure good quality products. AstraZeneca, that is a leading manufacturer of different pharmaceuticals, works according to GMP to make sure that their processes deliver products of the same quality every time. A problem that has occurred at AstraZeneca is when a raw material is not properly understood and variations in the raw material affects the final product. Variations in drug release in one of AstraZeneca’s products, Product X, has been linked to the cushioning excipient Microcrystalline cellulose (MCC). Variations in drug release has been noticed during change from one batch of MCC to another. The aim of this study was to investigate which material attributes of MCC that contributes to variations in the final product. Particle size and moisture content were identified as critical material attributes (CMA’s) and were therefore chosen to be investigated more thoroughly. By variating particle size and moisture content during manufacturing of Product X, the influence of these attributes could be investigated using Design of Experiment (DoE). An additional experiment that compared two MCC batches from different suppliers was also performed during this study. The results from these experiments showed that the particle size and moisture content of MCC does affect the drug release. Large particles and high moisture content gave rise to a faster drug release compared to small particles and low moisture content that gave rise to a slower drug release. It is however hard to draw conclusions regarding how small differences in particle size and moisture content could affect the drug release.
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Abbreviations

- CMA – Critical Material Attributes
- DoE – Design of Experiment
- GMP – Good Manufacturing Practice
- LOD – Loss on drying
- MCC – Microcrystalline cellulose
- MUPS – Multiple-unit Pellet System
- RH – Relative Humidity
1. Introduction

1.1 Background
In the manufacturing of pharmaceuticals it is important that the process is robust and reproducible to continuously insure good quality. This is achieved by well monitored processes along with raw materials of the right quality.

All raw materials at AstraZeneca are analyzed before they are used in the production, to make sure the material meets the existing specification requirements. The production at AstraZeneca is based on manufacturing methods and validated processes that ensures reproducible processes. This is achieved by working according to Good Manufacturing Practice (GMP). Using this approach during manufacturing ensures good quality products every time.

When a raw material is not properly understood, the existing specification requirements may not be enough to ensure that the material is of the right quality. This in turn can give rise to problems during manufacturing and variations in the final product can occur, despite working according to GMP. Therefore, it is important for AstraZeneca to understand how raw materials affect manufacturing and the final product.

1.2 Purpose of the project
The purpose of this project was to investigate which material attributes of microcrystalline cellulose (MCC) that affect the drug release of Product X.

Product X is a controlled-release drug that is manufactured by AstraZeneca at one of their plants at Gärtuna, Södertälje. The product is a tablet that consists of active substance together with excipients, where MCC works as a cushioning agent. Variations in drug release of Product X has been noticed during change from one batch of MCC to another. Variations in drug release can also occur when using a new fraction of the same vendor batch or different sections of material in the same container.

The variations in drug release of Product X that can occur, sometimes results in the product not meeting the existing specification requirements. This makes it very important for AstraZeneca to investigate how MCC affects the drug release and why these variations occurs. This was achieved by examining MCC in order to get a better understanding in which material attributes that contributed to these variations. By identifying the critical material attributes (CMA), AstraZeneca could provide a more robust process that will provide a more capable manufacturing process.
1.3 Objectives

The main objective of this project was to identify material attributes of MCC that affect the drug release of Product X. To achieve this, enough information about MCC had to be collected using theory to make sure that the investigation focused on CMA’s. When the CMA’s were identified, a plan for how the project was going to proceed was formed. The practical experiment had to be designed and get started early in the project to make sure there was enough time to analyze all the material that was produced during the experiment. To achieve the main objective there were some intermediate objectives that had to be reached during the project:

1. Get fractions of MCC with different particle sizes
2. Get fractions of MCC with different moisture content
3. Decide the solid phase properties of the different fractions
4. Manufacture tablets from the different MCC fractions
5. Decide the drug release of the tablets
6. Evaluate the drug release results using statistical analysis

To achieve fractions of MCC with different particle sizes the MCC had to be sieved. The sieving of MCC was the first activity performed in the project. This resulted in three fractions of MCC that were sent on analysis to test the solid phase properties. The first attribute that was analyzed was particle size to make sure that the first intermediate objective was reached. When this was accomplished the remaining solid phase analysis could be performed subsequently to achieve the third intermediate objective.

When sieving had been performed, the MCC was kept at different humidities to gain different moisture content before tableting. The moisture content was measured immediately before tableting to get accurate values and make sure that the second intermediate objective was reached.

To make sure that the final results was dependent on either particle size or moisture content of MCC, the difference in these properties had to differ enough to eliminate the influence of other parameters. It was also important to keep all material- and process parameters, except MCC, constant during compression. The tableting was performed using Design of Experiment where three central points were manufactured. This was to eliminate the influence of variances in both the process and analytical method, which reduced the risk of faulty results. When the tablets were compressed the fourth intermediate objective was reached and the tablets were sent on drug release analysis. When the results from the drug release analysis could be collected the fifth intermediate objective was reached.

The last intermediate objective was reached by doing a statistical evaluation using regression analysis. The evaluation was based on the results from the different analyzes performed during the project. The regression analysis was performed to demonstrate which of the attributes that affected the drug release, and if there was an interplay present. Based on the result from the regression model, conclusions regarding how particle size and moisture content affects the drug release were drawn. The results from these conclusions were then used to reach the main objective.
1.4 Expected impact of the study
This study aimed to investigate the material MCC and how variations in particle size and moisture content affect drug release of Product X. The outcome of this study is expected to give AstraZeneca indications of whether, and how, these attributes influence variations in drug release. It has been noticed that the variations in drug release can be linked to the material MCC, that is an excipient in Product X. However, AstraZeneca needs to get a greater understanding in how MCC affects the drug release. By identifying the attributes that contribute to these variations, AstraZeneca could overcome the problem of drug release variations in Product X. This would provide a more robust and reproducible manufacturing, which is very beneficial, since it would lead to both economical saving and savings in time.

1.5 Boundary conditions
The project was performed at AstraZeneca during a period of 20 weeks at one of the plants of Sweden Operations in Gärtuna, Södertälje. It was limited to only investigate the material MCC and how it affects the drug release for Product X. During the project two experiments were performed. The first experiment investigated how particle size and moisture content affects Product X by using Design of Experiment (DoE). The second experiment compared the drug release for Product X containing MCC from two different suppliers as excipients. The solid phase analyzes and drug release analyzes were performed by qualified staff at laboratories located at the plant. The initial plan was to investigate both low dosage tablets and full dosage tablets, but due to time pressure at the laboratories, the project was limited to only investigating the low dosage tablets.
2. Theory and methodology

2.1 Scientific background

Product X that was investigated during this project is an oral solid dosage drug with controlled release. To achieve controlled release Product X is formulated as a Multiple-Unit Pellet System (MUPS) tablet. The tablets contain pellets of active substance coated with a film that controls the release of the drug. Tablets are formed by compressing pellets together with excipients, where MCC works as a cushioning agent.

2.1.1 Oral solid dosage forms

Oral solid dosage formulations are the most common drug delivery systems. The two main types are tablets and capsules where tablets are beneficial for both patients and manufacturers. This is due to many factors but two main reasons are good patient compliance and cost-effectiveness in large-scale manufacturing. Easy handling, plenty of manufacturing methods, consistent quality and dosing precision etc. are other attributes that makes tablets advantageous against other drug delivery systems. (Gad, 2008) (Lachman, et al., 1987)

A flow chart explaining the path of a tablet from active substance until eliminated from the patient is shown in Figure 1. The active substance is compressed into tablets together with inert substances that are used as excipients. As the patient consumes the tablet, the bioavailability plays an important role in how well the drug is absorbed in the patient. This later determines how well the drug is distributed to the right compartments. Depending on the patient’s metabolism and the pharmacological action the drug will eventually be eliminated and the patient needs to take a new tablet during long term treatment. (Vergnaud, 1993)

![Figure 1: Flow chart showing the path of tablets from active substance to elimination from the patient.](image-url)
2.1.2 Immediate release
In conventional dosage forms, the drug is released in the patient immediately as it reaches the gastrointestinal tract. As soon as the tablet is dissolved, the drug is released and distributed in the patient’s body. This gives an immediate high concentration of drug that decreases fast both in the gastrointestinal tract and in the blood and tissue as seen in Figure 2. During long-term treatment, continuous intake of the drug is necessary in order to keep the drug level at a therapeutic level. Continuous intake of conventional dosage forms leads to fluctuation in the drug level as seen in Figure 2, this contributes to excessive use of the drug during long-term treatment. During the intake and a short period after the intake, the patient suffers from over dosage, and after a while as the drug concentration decreases the patient suffers from under dosage. Due to this behavior, patients often suffer from side effects caused by the recurring over dosage. (Vergnaud, 1993)

![Conventional dosage form](image)

*Figure 2: A graph showing the concentration of drug in the gastrointestinal tract and blood and tissue when consuming conventional immediate-release drugs.*

2.1.3 Controlled release
The development of modified release drugs arose from the problems that occurred during long-term treatment with conventional drugs. The aim with the modified release drugs is to control the release profile and are often referred to as controlled-release systems. The release is modified in different ways by either delaying, sustaining or repeating the drug release for a drug. In comparison to conventional immediate-release drugs, controlled-release drugs offers plenty of benefits regarding patient safety and compliance. An advantages is for example that controlled-release system provides the ability to maintain the therapeutic levels of drug on a rather constant level over a long period. (Gad, 2008)
Kapil et al. and Karlson et al. both performed studies where controlled-release dosage forms are compared to immediate-release dosage forms during long-term treatment (Figure 3). The results from the studies show that the drug concentration is kept on a constant level during steady state when consuming the controlled-release drug. The results also show that immediate-release drug causes large fluctuations during long-term treatment. The fluctuations as mentioned in previous paragraph often lead to negative side effects due to the recurring over dosage. The controlled-release dosage forms are therefore beneficial since it prevents fluctuations of the drug levels and increases the duration of the therapeutic effect. This leads to higher efficiency with less amount of drug at the same time as it reduces the frequency of drug administration, which is convenient for the patient. (Gad, 2008) (Wen & Park, 2010) (Karlson, et al., 2014)

![Figure 3: A graph showing the difference in drug concentration when comparing controlled-release drugs and immediate-release drugs.](image)

Controlled-release systems are delivered either by single-unit doses or by multiple-unit doses. Single-unit doses delivers the drug in one depot that disintegrates over a period of time while multiple-unit doses delivers the drug in plenty of mini-depots that disintegrates separately over a period of time. The multiple-unit system has some advantages compared to single-unit systems, for example the risk of local irritation and dose dumping minimizes drastically. Dose dumping and local irritation occurs if a single-unit system raptures or is trapped somewhere in the gastric system. This is eliminated when using multiple-unit systems since the mini-depots is dispersed over a larger surface and if one mini-depot raptures the effect of this is too small to affect the patient in any great extent. (Bechegaard & Nielsen, 1978)
The multiple-unit system is also less dependent of the patient’s individual digestion pattern. The mini-depots are able to reach the small intestine independent of gastric emptying which leads to improved therapeutic effect and bioavailability. The results from Cnota et al. shows that less variation in bioavailability occurs when using multiple-units doses compared to single-unit doses. Multiple-unit doses also manage to maintain a systematic drug availability that contributes to a more stable drug concentration. (Bechgaard & Nielsen, 1978) (Cnota, et al., 2005)

Single-unit doses and multiple-unit doses are delivered through oral dosage forms either as membrane systems or matrix systems. Membrane systems implies that the drug is surrounded with a membrane that controls the rate of the drug getting released. Matrix systems on the other hand implies that the drug is embedded in a matrix and the drug is released as the matrix dissolves. By combining these a hybrid system can be achieved which gives an even better system to control the release rate of the drug. For example modified-release coated pellets can be imbedded in a tablet that works as a matrix system or filled in a capsule that works as a membrane system. (Wen & Park, 2010) This phenomena is called Multiple Unit Pellet Systems (MUPS) and is often referred to as MUPS in the form of tablets. (Bhad, et al., 2010)

2.1.4 Multiple Unit Pellet System (MUPS) tablets

MUPS are either compressed into tablets together with different excipients or filled into capsules as solid dosage forms. MUPS compressed into tablets shows a lot of advantages compared to MUPS filled into capsules, where low cost and easy manufacturing in large scale are two big factors to why tablets are more beneficial. Another advantage that makes MUPS tablets more beneficial compared to MUPS capsules is the difficulty to replicate MUPS tablets. This makes it possible for the manufacturer to maintain monopoly on the product even as the patent expires. (Bhad, et al., 2010) (Choudhary & Avari, 2013)

![Coated Pellet and Matrix Pellet](image)

*Figure 4: A picture showing how a coated- and matrix pellet are constructed.*

The pellets that are used in the manufacturing of MUPS tablets are either coated pellets or matrix pellets as shown in Figure 4 (Ozarde, et al., 2012). Where the coated pellet works as a membrane system where the drug is released by diffusion through the film of the pellet. The matrix pellet works as a matrix system, the drug is released as the pellet disintegrates. The pellets used in Product X are coated pellets which has a core that is coated with the drug and a film that is designed to control the release of the drug. When compressing coated pellets into MUPS tablets, it is important that the formulation of pellets and excipients prevent changes in the film that controls the release, to maintain the wanted release profile. (Beckert, et al., 1996) (Torrado & Augsburger, 1993) (Tunón, et al., 2003)
2.1.5 Challenges during manufacturing of MUPS-tablets

During manufacturing of MUPS tablets there are many factors that have to be taken into consideration to success. The greatest challenge in the making of MUPS tablets is the ability to maintain the drug release of the pellets after compression. When using coated pellets, which is the case in Product X, it is important that the film of the pellet that controls the release does not change during compression. The controlled-release coating can be damaged by deforming or densifying the pellets. If this occurs the release profile of the drug can change, which is not desirable. (Beckert, et al., 1996) (Torrado & Augsburger, 1993) (Tunón, et al., 2003)

A study performed by Tunón et al showed that the change in release profile could depend on two different mechanisms. If the pellets are deformed the film could either rupture or get stretched out which makes it easier for the drug to pass through the film. If this happens during compression the drug is released faster compared to the pellets that are not compressed into MUPS tablets. The other scenario is that the pellets are densified during compression, which makes the film thicker and makes it harder for the drug to pass through the film. This leads to a prolonged release profile compared to the pellets that are not compressed into MUPS tablet. (Tunón, et al., 2003) To avoid rupture of the film due to deformation of the pellet it is important to apply a coating that is able to follow deformation without rupturing. This is maintained by choosing the right substance and the right thickness of the film. (Beckert, et al., 1996)

To avoid further damage of the film it is important to add an excipient that works as a cushioning agent to the pellets before compression.

2.1.6 Cushioning excipients

The excipients that are used during manufacturing of MUPS tablets need to have a cushioning effect since coated pellets are pressure sensitive. The cushioning effect is important since the excipient need to be able to absorb the force that is formed during compression of the tablets. Damage of the pellets is either caused by the pressure of the punch or by pellets that are pressed to each other inside the tablet due to lack of excipient. The purpose of the excipient is to prevent the pellets from deforming, densifying and sticking together during compression since these scenarios affect the drug release. To prevent the pellets from sticking together the excipient should be forming a layer around every pellet during compression. (Beckert, et al., 1996) (Bodmeier, 1996) (Torrado & Augsburger, 1993) To insure that the damaging of pellets is minimized it is important that the mixture contains the right ratio between pellet and excipient. The amount of drug has to be enough to get the right dosage and the amount of excipient should be enough to prevent damage of the pellet film when compressed into MUPS tablets. The pellets are less damaged as the proportion of excipient increases. (Beckert, et al., 1996) (Torrado & Augsburger, 1993) The cushioning excipient used in Product X is microcrystalline cellulose (MCC).
2.1.7 Microcrystalline cellulose

MCC is a widely used excipient and is produced by hydrolyzing purified wood pulps which separates the amorphous regions of the cellulose. This is fulfilled by using mineral acid solutions which cleaves the \( \beta\)-1,4 linkage between glucopyranose units in the amorphous regions of cellulose. This cleavage generates microcrystals that are spray-dried to create agglomerates, these agglomerates generates the material MCC. Although the process eliminates amorphous parts from the original cellulose, MCC still contains both crystalline and amorphous regions, but the crystallinity of MCC is higher than in original cellulose. (Sun, 2008)

The desired particle size and moisture content of MCC is achieved by controlling the spray-drying step. Since MCC is often produced in continuous processes where a batch represents a period of time in the production, it can be difficult to prevent and discover variations within a batch. By understanding the physiochemical properties of MCC and how these can vary it could be possible to optimize the performance of MCC when used in tablet manufacturing. (Thoorens, et al., 2015)

2.1.8 Microcrystalline cellulose as a cushioning excipient

Since MCC is the cushioning excipient used in Product X it is important to understand which material attributes that have an impact on the final product and thereby could influence the drug release. There are some critical material attributes (CMAs) of MCC that are identified by different studies. CMAs regarding MCC as an excipient are moisture content, particle size, particle morphology, bulk density, tapped density, specific surface area, degree of polymerization and crystallinity. Depending on these attributes MCC behaves differently which has an impact on the final product. Variations in final products containing MCC could therefore depend on variations in MCC. (Khan, et al., 1981) (Kushner, et al., 2011) (Thoorens, et al., 2014) (Thoorens, et al., 2015)

Although it is proven that MCC has an impact on drug release when using it as an excipient in tablets, MCC behaves differently depending on the tablet formulation. Since formulations vary a lot between different products on the pharmaceutical market, it is important that each manufacturer understands how MCC affects their product. (Thoorens, et al., 2014) (Khan, et al., 1981) This makes it interesting for AstraZeneca to find out which attributes of MCC that play an important role in the drug release of Product X.

2.1.9 How moisture content affect tableting of MCC

During tableting there are many factors that can affect how well the process proceeds and the quality of the finished product. One factor that has an impact on tableting is the moisture content in the materials that are being compressed. The amount of water that a material contains can affect the material in different ways, and thereby affect the tableting. The flow ability of the powder is influenced by the hygroscopicity of the material since an adsorption film can be formed with water as solvent. This leads to greater particle to particle interactions in the material since the water works as a bridge between particles through surface adsorption mechanisms. (Gad, 2008)
Since MCC is a material that is hygroscopic, it is very important to have good control over the environmental moisture content, the relative humidity (RH). The amount of water that MCC contains will affect different properties of the material. A property that is well known among cellulose materials is that the particles swells in contact with water (Sun, 2008). Depending on how much water and where the water is located MCC will behave differently. The RH has an impact on the flow properties of the MCC, which affects the tablet ability. At higher RH water molecules on the surface of MCC particles increase. This increases the strength in the particle to particle interactions due to hydrogen bonds. These particle to particle interactions have a negative impact on the flow ability since it decreases as RH gets higher. At lower RH on the other hand the water tends to locate in the amorphous parts of the MCC molecules. (Sun, 2016)

Microcrystalline cellulose is a complex molecule since the structural properties of cellulose can vary depending on the original source and manufacturing conditions. Since this is the case, it is complicated to understand how moisture affects the material. Depending on surface area, pore volume and crystallinity moisture will be absorbed differently and it will have different impacts on how the material will behave. Awa et al. performed a study that investigated how MCC with different crystallinity affected the hydrophilic properties of tablets. The results in the study show that MCC that contains larger amount of amorphous parts and less crystalline parts tends to absorb more water. This is due to the hydrogen bonds that can occur between the amorphous parts and water molecules. (Awa, et al., 2015)

2.1.10 How particle size affects tableting of MCC

The particle size of the cushioning excipient is very important in order to manufacture MUPS tablets that complies with the requirements. There are a lot of factors that should be considered when choosing the excipient. Yao et al. shows that excipients of very small particles, around 5 µm, tend to protect the pellets very well during compression. This is due to the ability of distributing and creating a protective layer around the pellet, which protects the pellet from rupturing and sticking together during compression. However, studies show that the particle size of the excipients should not differ too much from the particle size of the pellets to prevent segregation. Segregation can lead to content uniformity and uniformity of the weight. These factors make particle size a very important aspect during compression. (Beckert, et al., 1998) (Wagner, et al., 1997) (Yao, et al., 1997) (Yao, et al., 1998) The particle size of the binder is also proven to have an influence on tablet hardness, smaller particles give rise to tablets with higher mechanical strength, and harder tablets (Nyström, et al., 1982).

If large particle size differences occur within the formulation the risk of segregation during manufacturing increases according to Deng et al. Events that take place before compression in the manufacturing process are critical steps that can result in segregation of particles. For example as the blend is transferred from a container to the tablet press the risk of air-induced segregation occurs. The risk for segregation depends on the flowability of a material, materials with increased flowability has a larger risk of segregating since the particles are free-flowing and easily separates from each other. (Deng, et al., 2010)
When using an excipient like MCC that is in the form of powders it is important to consider the possibility of segregation within the material. Powders often consist of particles of different sizes and depending on how the material is handled both during transportation and during the manufacturing process, it is always a risk that the particles segregate. During manufacturing of MUPS tablets, it is important to understand how the particle size of the excipient together with pellets will affect the performance of the final product. (Beckert, et al., 1998) (Jaklic, et al., 2015)

2.2 Methodology

2.2.1 Pre-study
An initial pre-study was performed at AstraZeneca, the study investigated two different batches of MCC. These batches had previously shown variations in drug release when changing from one to another. The study was made to distinguish if any apparent differences between the two MCC batches could be seen. The material attributes that were investigated during this study were crystallinity, particle size and surface area. The result from the analyzes that were made on the MCC showed that the two batches were very similar regarding these attributes. This made it difficult to draw any clear conclusions. This could indicate that the difference in drug release was not influenced by these material attributes or that small differences in these material attributes could have a significant impact on the drug release. Further information had to be collected, through a literature study, to make a decision on which material attributes that was going to be investigated.

2.2.2 Literature study
The aim of the literature study was to receive information that was needed to understand which material attributes of MCC that could affect drug release. It was also important to get essential information about how MUPS tablet works and how drug release can be controlled by multiple-unit systems. The information that was collected during the literature study was necessary in order to make a good decision on which attributes that was going to be investigated during the project. It was also necessary to be able to draw conclusions from the results in this study.

The literature study was performed by searching for keywords like controlled-release, tablets, multiple-unit systems, MUPS, pellets, microcrystalline cellulose, cushioning excipients etc. The databases that has been used in order to find articles and books, are the one that was provided by the library at Linköpings University and the database that was offered by AstraZeneca. Some books that has been used during the project was located on the site at Gärtna.

The result from the literature study implied that particle size, particle shape, tapped density, moisture content and crystallinity were attributes that could affect the final product. (Thoorens, et al., 2014) It also showed that MCC has different impacts depending on the formulation which makes it relevant to investigate which effect it has on Product X. (Thoorens, et al., 2014) (Khan, et al., 1981)
2.2.3 Material attributes to investigate
The results from the pre-study together with the literature study gave some indications on which material attributes that could be of interest during this study. The final decision on which material attributes that were going to be investigated was decided together with the supervisor of the project and competent staff at AstraZeneca. These people had a lot of knowledge about the product and about the material MCC, and their inputs were of great value when designing the project.

The material attributes that were chosen to be in the focus of this study were particle size and moisture content. These were listed as CMA according to the literature and after some discussion with staff at AstraZeneca they were chosen due to the possibility to modify these attributes. Particle size was modified by sieving the material and thus obtaining fractions with different particle sizes. Moisture content was modified by storing the material at different humidities, which gave rise to different moisture contents. This made it possible to investigate the impact of variations in MCC regarding particle size and moisture content, and how this affects drug release. To be able to see if and how these attributes interact a Design of Experiment (DoE) was performed.

2.2.4 Design of experiment
During the project, two separate experiments were performed where one aimed to see the effect of particle size and moisture content and the other aimed to compare MCC from two different suppliers. The first trial was performed using a full factorial design. These results were later compared to the second trial to see if the same conclusions could be drawn from both trials.

Experiment 1
When performing a factorial design a number of versions is selected for a number of variables. The versions are called levels and the variables are called factors. The aim of a factorial design is to run experiments with all possible combinations of the different versions of the variables. The number of experiments are calculated by the formula \( l_1 \times l_2 \times \ldots \times l_k \), where \( l_1 \) is equal to the number of levels for the first factor, \( l_2 \) is equal to the number of levels for the second factor and \( l_k \) equal to the number of levels for the k:th factor. (Box, et al., 1978)

In experiments, the levels are often coded so that zero should represent the midrange of the levels of the factors. This is maintained by representing the highest and lowest levels of the factors with +1 and -1, which makes it possible to create a standard first-order design. The midrange of the levels of the factors are called “center points” while the different combinations obtained by the design of the experiment are called “factorial points”. In a standard first-order design, the design consists of \( n_f \) factorial points and \( n_0 \) center points. The center points are important to include in the experiment to be able to provide error degree of freedom and adequate power for a test for lack of fit. A standard-first design is always orthogonal which is fulfilled by two requirements. Each factor should have half of its levels at low respectively high levels, and the sum of cross products of the coded level should be zero. (Dean & Voss, 1999)
This experiment aimed to investigate how the drug release was affected when particle size and moisture content of MCC varied in the different combinations seen in Figure 5. All possible combinations were investigated which was fulfilled by using a full factorial design.

**Figure 5:** Shows all possible combinations that were investigated by using design of experiments.

**Table 1:** A table showing the factors investigated in the experiment, and the different levels of the factors.

<table>
<thead>
<tr>
<th>Factor</th>
<th>Low level (-1)</th>
<th>Central point (0)</th>
<th>High level (+1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(A) Particle size</td>
<td>Small</td>
<td>Medium</td>
<td>Large</td>
</tr>
<tr>
<td>(B) Moisture content</td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
</tr>
</tbody>
</table>

**Table 2:** A table showing the experimental settings for how the trials were performed.

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Order</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2</td>
<td>-1</td>
<td>-1</td>
</tr>
<tr>
<td>2</td>
<td>5</td>
<td>+1</td>
<td>-1</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>-1</td>
<td>+1</td>
</tr>
<tr>
<td>4</td>
<td>6</td>
<td>+1</td>
<td>+1</td>
</tr>
<tr>
<td>5</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>6</td>
<td>7</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>7</td>
<td>4</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
The experiment was designed as a standard first-order design, which included a $2^2$ full factorial design consisting of two factors that varied at two levels each. The two factors were represented by particle size (A), that could vary between small and large, and moisture content (B), that could vary between low and high (Table 1). The number of experiments needed in this design was calculated from the formula $l_1 \times l_2$ where $l_1 = 2$ and $l_2 = 2$ and gave rise to four experiments. The experiment also included three central points where both particle size and moisture content were kept at medium level. The experimental settings seen in Table 2 were generated from the software Modde 11 and a more graphic picture of the set-up is seen in Figure 6. By designing the experiment as a full factorial design all combinations within the square in Figure 6 should be represented in the final result.

![Figure 6: A picture that shows which factors and levels that will be analyzed in the experiment. The red dots corresponds to a trial where the factors are at either low- or high level, the middle point corresponds to the three central points that will be tested.](image)

**Experiment 2**

The aim of the experiment that compared MCC from the current supplier, Supplier 1, with another supplier, Supplier 2, was to investigate how these MCC batches differ and how it affects the drug release. It was also of interest to see if the result from this study behaved similar as in the full factorial design. The experiment was designed as in Figure 7, and each tableting was performed two times to eliminate variations in the tableting and analyze method.

![Figure 7: A figure showing how Experiment 2 was designed.](image)
2.2.5 Evaluation
Regression analysis was used in the statistical evaluation of Experiment 1. The regression analysis aimed to investigate how particle size and moisture content affected the drug release of Product X. By using regression analysis, information about which attributes that have a significant impact on the drug release could be obtained. It was also possible to reveal if any interplay between these attributes was present.

The raw data from the drug release analyzes were also an important input during the evaluation of the results. Together with the results from the regression analysis it was possible to draw conclusions regarding particle size and moisture content of MCC, and how it influences the drug release of Product X.

The results from Experiment 2 was evaluated by comparing the raw data from the drug release analysis together with solid phase characteristics. This was of interest to reveal how MCC from different suppliers differed from each other and if that affected the drug release of Product X. The results from Experiment 1 and Experiment 2 were then compared to see if similar conclusions could be drawn from the two experiments.
3. Materials and methods

3.1 Materials

3.1.1 Microcrystalline cellulose
In this study Microcrystalline cellulose with different particle sizes was investigated. Three fractions with different particle sizes were obtained by sieving the original MCC from the current supplier. Unsieved MCC was also used in the project from both the current supplier, Supplier 1, and from another supplier, Supplier 2. The particle size, surface area, crystallinity, true density and bulk density was determined from the solid phase analyzes of the different MCC powders.

3.1.2 Coated pellets
Coated pellets for low dosage was used for tableting. These are manufactured by AstraZeneca at the plant Gärtuna.

3.1.3 Others
Other materials that were used during the project was excipients included in the tablet, and solutions and substances needed during the different analyzes.

3.2 Methods

3.2.1 Sieving
Sieving was performed by using an analytical sieve shaker (Retschen siever AS 200 ‘g’) with three different sieve sizes to yield three different fractions. The sieve shaker was set to the amplitude 1,50 during 5 minutes with an interval of 10 seconds. During the sieving 20 grams of MCC from the current supplier, Supplier 1, was added to the sieves at a time and the sieve sizes were 90 µm, 180 µm and 300 µm. This yielded three fractions with a small, medium and large particle size. The small with a theoretical size of 0-90 µm, the medium with a theoretical size of 90-180 µm and the large with a theoretical size of 180-300 µm. The sieving was performed until enough material for all the trials in Experiment 1 could be collected for all fractions. When the sieving was finished samples from all fractions were sent on solid phase analysis to investigate the actual particle size and other material attributes.

3.2.2 True density
True density of the MCC fractions were measured using a helium gas pycnometer (AccuPyc 1330, Micromeritics). This was performed by filling the tube in the pycnometer to about 70-80% and inserting the weight of the containing MCC. The tube was then placed in a chamber of known volume where helium flows in and works as a displacement medium. The volume of the helium that leaves the chamber was then measured and the true volume of the MCC powder could be obtained. The instrument then divided the volume with the weight of the material to maintain the true density. (Micromeritics, u.d.)
3.2.3 Particle size and Surface area

Particle size was measured with two different techniques, the first one was performed with a QicPic (Sympatech) on the sieved fractions to ensure that sieving was successful. Since the QicPic (Sympatech) was not validated for MCC powders, another measurement with a Mastersizer 3000 (Malvern) was performed. The Mastersizer 3000 (Malvern) was validated for MCC powders and ensured that the measurement showed accurate results. The Mastersizer 3000 (Malvern) could also provide the surface area for the particles.

QicPic Measurement

The particle size distribution of the different MCC fractions were measured by a dynamic image analysis (DIA) (QicPic, Sympatec) together with the software Winfox 5.0. Approximately 5-10 ml of MCC was added to the feeder for each measurement, two measurements per fraction were made to ensure that the obtained values were accurate. Each fraction of MCC were analyzed with different instrumental parameters that were evaluated by preliminary experiments. These parameters were for example feed rate that could vary between 10-30% and dispersing time that varied between 20-80 seconds. The parameters were fitted to fulfill the specification that the measurement should include more than $10^5$ particles to minimize statistical and sampling errors that can occur if the sample is too small (Yu & Hancock, 2008) (Masuda & Inooya, 1971).

Mastersizer 3000

The validated particle size distribution measurement of MCC powders were performed with the Mastersizer 3000 (Malvern) together with the feeder Aero S (Malvern). The parameters that were used during the measurements were predetermined from the validation of the instrument. These parameters were for example feed-rate that was set to 50%, measurement time was set to 18 seconds and the dispersion air pressure was set to 1 bar. Between 8-15 ml of the MCC was added to the feeder for each measurement. Each fraction of MCC was analyzed two or three times to ensure that the values were accurate.

3.2.4 Bulk density (T.A.P.)

A GeoPyc 1360 (Micromeretics) envelope density analyzer was used to analyze the bulk density of the different MCC Powders. Analysis was performed with the T.A.P. Density option which measures packing volume and calculates bulk density. The sample was placed in a sample cell that is a precision cylinder, which rotated while a specific force was applied to the sample. The difference in volume before and after the force had been applied was measured, and the bulk density could be calculated. (Micromeritics, u.d.)

3.2.5 Crystallinity

The crystallinity of the different MCC Powders were measured with an X-ray powder diffraction instrument (X’Pert Pro, PANalytical). The sample that was analyzed was spread out on a plate that was illuminated with X-ray from different angles (Figure 8). Depending on how the particles are structured, the phase of the scattered light will differ. The scattered light is sent to a detector that evaluates the crystallinity of the material. The result was then presented as graphs that showed the intensity curves of the scattered light. Depending on the curve appearance the crystallinity of the material could be determined (Figure 9).
3.2.6 Moisture content
To obtain different moisture content in Experiment 1, the different MCC powders had to be stored at different humidities. The design of the experiment that investigates the different particle sizes and moisture content included MCC with small and large particle sizes with both low and high moisture content. The three central point with medium particle size and medium moisture content were also included in the experiment. Due to this, the fraction with medium particle size was stored in a room with a theoretical humidity of 45%. The fractions with small and large particle sizes had to be divided and was either stored in a room with the theoretical humidity of 20% or an exicator containing saturated NaCl that gives rise to a theoretical humidity of 75% at equilibrium. (Alshawa, et al., 2009) The MCC from Supplier 1 and Supplier 2, which was used in Experiment 2, were both stored in the room with a theoretical humidity of 45%.
To facilitate the uptake of moisture during the storage in different humidities, the MCC powders were spread out on paper sheets in a thin layer. The powders that were stored in an exicator were spread out in a petri dish.

To control the moisture content of the different powders both water activity (aw) and loss on drying (LOD) was measured right before tableting. Two measurements were performed with both techniques right before mixing the MCC with pellets and on the MCC that was left after the mixing. This was to see if the moisture content changed during handling of the material since all handling was performed at normal humidity which corresponds to 45% RH.

Water activity is a measurement that provides the ratio of water vapor pressure of the material to the vapor pressure of pure water at the same temperature. If the water activity is expressed as percentage the equilibrium relative humidity (ERH) is obtained. The water activity should thereby show similar values as the relative humidity in the room. An Aqualab was used to measure the water activity. The Aqualab uses the technique of a cooled mirror sensor to obtain the water activity of the material at the same time as it measures the temperature of the material using an infrared thermometer. (Aqualab, 1998)

Loss on drying is a technique that measures the amount of volatile substances, primarily water, in a material. This was executed by using a moisture analyzer (Mettler Toledo) that dried 3 mg MCC at 110°C until all volatile substances left the material. By weighing the sample before and after drying the loss on drying can be calculated.

3.2.7 Tableting

When the different MCC fractions had been stored in the desired humidities each MCC fraction was mixed together with pellets and lubricant according to the recipe of the drug. The mixing was performed with a Turbula mixer (Willy A. Bachofen AG Maschinenfabrik) during a specific period of time obtained from the recipe of Product X. As the mixing was finished, the mixture was added to a single punch tablet press (Korsch EK0). The tablets were manufactured to a specific tablet weight, which was obtained by controlling the filling die. If the tablets did not reach the right weight adjustments in the filling depth had to be made on the tablet press (Korsch EK0). As the tablets reached the right weight samples for IPC and drug analysis could be collected. During tableting the press force was controlled and kept within a specific interval that was given in the recipe of Product X.

The manufacturing of low dosage tablets were performed using an 8 mm round punch. Due to human error the punch broke as it was two trials left of the low dosage tablets. To be able to finish the manufacturing of low dosage tablets the punch had to be replaced with a 7 mm punch. The tablet weight and press force were kept at the same level as with the 8 mm round punch to make sure that most parameters were kept constant. This will be taken into consideration as the results of the drug release will be evaluated.
3.2.8 In Process Controls (IPC)

IPC were performed to see how well the tablets fulfilled the given specifications regarding tablet weight and tablet hardness. It is important to control the weight and hardness of the tablets to make sure that the process is reproducible and that the amount of drug in every tablet is the same.

The IPC were performed on ten different tablets that were collected during the tableting. The first IPC control was to weigh the tablets to insure that they fulfilled the given specifications regarding weight. The weight was controlled by weighing the ten tablets together on a scale (Mettler Toledo PG 203) to maintain a mean value of the tablet weight.

The ten tablets were then tested for tablet hardness to see how the hardness differed between tablets containing different MCC fractions. The tablet hardness was measured individually on each tablet using a C50 tablet hardness tester (Holland). As the measurements were finished the mean value of the ten tablets was calculated.

3.2.9 Drug release

The drug release analysis was performed at the quality control (QC) laboratory at Gärtuna. Qualified staff performed the drug analyzes by a validated method to ensure that the results from the analysis were credible.

The drug release analysis was performed by placing six tablets from each trial in acid baths that mimics the environment in the stomach. The drug content in the acid bath was then measured with HPLC at different time intervals for 20 h.
4. Results

4.1 Solid phase analysis

Particle size was first measured by the QicPic (Sympatech) in conjunction with sieving to assure that the sieving was successful. As the sieving was finished all fractions including the unsieved MCC powders from Supplier 1 and Supplier 2 were sent on solid phase analysis.

4.1.1 Crystallinity

The results from the crystallinity measurements for all MCC powders are shown in Figure 10. All graphs show one distinguishing peak which indicates that the material consist of crystalline structure. However, the absence of multiple distinct peaks indicates that the material consist of amorphous structure as well.

Figure 10 shows that the MCC powder from Supplier 1 had similar crystallinity as the three sieved fractions, small, medium and large. A difference in crystallinity could be seen when comparing Supplier 1 and Supplier 2. The MCC powder from Supplier 2 had a higher crystallinity which is shown by a higher peak in Figure 10.

![Figure 10: A graph showing the result from the crystallinity measurements. Higher peaks corresponds to higher crystallinity. The peak that deviates from the other and shows a higher crystallinity belongs to the MCC from Supplier 2.](image-url)
4.1.2 MCC used in experiment 1

Table 3: Showing the results from the solid phase analyzes that has been performed on the sieved fractions. The particle size that is marked with bold text is the results from the Mastersizer 3000 (Malvern).

<table>
<thead>
<tr>
<th>Particle size</th>
<th>True density (g/cm³)</th>
<th>D_{10} (µm)</th>
<th>D_{50} (µm)</th>
<th>D_{90} (µm)</th>
<th>Surface area (m²/kg)</th>
<th>Bulk density (g/cm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Small</td>
<td>1.5676</td>
<td>42.89</td>
<td>77.13</td>
<td>111.95</td>
<td>127.10</td>
<td>0.419</td>
</tr>
<tr>
<td>Medium</td>
<td>1.5712</td>
<td>115.93</td>
<td>164.79</td>
<td>218.25</td>
<td>38.82</td>
<td>0.353</td>
</tr>
<tr>
<td>Large</td>
<td>1.5636</td>
<td>225.75</td>
<td>272.32</td>
<td>343.78</td>
<td>22.00</td>
<td>0.325</td>
</tr>
</tbody>
</table>

The results from the true density measurement in Table 3 shows that the three MCC fractions had very similar values in true density.

The results in Table 3 shows that the sieving was successful since the three fractions contain particles of different sizes. This was first indicated when measuring the particle size distribution with the QicPic, which gave the results in Table 3 with narrow text. The results that is marked with bold text is the results that were obtained from the validated instrument Mastersizer 3000 (Malvern). The results from the Mastersizer 3000 (Malvern) show that the three fractions contained different particle size distributions. The small fraction contained very small particles (D_{50}=65 µm) while the large fraction contained very large particles (D_{50}=281 µm). The medium fraction contains particles with sizes in between the small and large particles, (D_{50}=162 µm), which was the aim with the sieving. Figure 11 shows a graph with the particle size distribution curves from the Mastersizer (Malvern) for the sieved fractions. The graph shows that all fractions showed normal distribution except for the small fraction that had a left-skew.

![Particle Size Distribution](image)

Figure 11: A graph showing the particle size distribution curves obtained from the Mastersizer 3000 for the different sieved fractions. The blue curve corresponds to the small fraction, the red curve corresponds to the medium fraction and the green curve corresponds to the large fraction.
The surface area that is presented in Table 3 shows the mean surface area with the unit m\(^2\)/kg. The results show that smaller particles had a larger surface area, 127.10 m\(^2\)/kg, and larger particles had a smaller surface area, 22.00 m\(^2\)/kg. Table 3 also presents the bulk density that also showed larger values for smaller particles, 0.419 g/cm\(^3\), and lower values for larger particles, 0.325 g/cm\(^3\).

4.1.3 MCC used in experiment 2

Table 4: Showing the results from the solid phase analyzes that has been performed on the MCC powders from different suppliers.

<table>
<thead>
<tr>
<th>Supplier</th>
<th>True density (g/cm(^3))</th>
<th>D(_{10}) (µm)</th>
<th>D(_{50}) (µm)</th>
<th>D(_{90}) (µm)</th>
<th>Surface area (m(^2)/kg)</th>
<th>Bulk density</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.5656</td>
<td>55.15</td>
<td>166.00</td>
<td>386.00</td>
<td>60.94</td>
<td>0.394</td>
</tr>
<tr>
<td>2</td>
<td>1.5669</td>
<td>42.55</td>
<td>180.00</td>
<td>419.00</td>
<td>57.55</td>
<td>0.452</td>
</tr>
</tbody>
</table>

The results in Table 4 shows the solid phase results for the MCC from Supplier 1 and Supplier 2. When comparing the results the particle size distribution and bulk density differed between the two suppliers, while true density and surface area showed very similar results. The curves in Figure 12 show that the particle size distribution had a left-skew for both suppliers, which implies that the mean particle size was less than the median for both suppliers.

![Figure 12: A graph showing the particle size distribution curves obtained from the Mastersizer 3000 for the MCC from different suppliers. The red curve corresponds to Supplier 1 and the green curve corresponds to Supplier 2.](image-url)
4.2 Moisture content and IPC controls

During the tableting process the initial step was to measure water activity and LOD on the MCC powders used in the manufacturing of tablets. Both LOD and water activity was measured twice on the MCC to see how it varied during handling of the material that was performed in normal humidity, RH around 45%. The first measurement was performed right before mixing the MCC with pellets and the second was performed on the MCC that was left over after the mixing. The results that are presented below shows that the moisture content was very hard to control since the material acclimatized to the new environment very fast. When moving the material from low humidity to normal humidity the LOD and water activity increases. When moving material from high humidity to normal humidity the LOD and water activity decreases.

As the tablets had been manufactured, IPC were performed to control the tablet weight and tablet hardness. The manufacturing process aimed to keep the tablet weight and the settings on the tablet press (Korsch EK0) as constant as possible, while the tablet hardness was allowed to vary when using different particle sizes and moisture content.

4.2.1 Experiment 1

The first experiment was performed using the MCC with varying particle size and moisture content.

Table 5: Shows the results from water activity and LOD that was measured on the MCC fractions before tableting, and the results from the IPC as the low dosage tablets had been manufactured. The tablet weight is presented as the difference from the set point in mg.

<table>
<thead>
<tr>
<th>Particle size (d50)</th>
<th>Humidity (RH%)</th>
<th>Water activity (a_w)</th>
<th>LOD (%)</th>
<th>Difference from weight set point (mg)</th>
<th>Tablet hardness (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>162</td>
<td>45</td>
<td>0.358</td>
<td>0.354</td>
<td>4.38 4.43 -0.2</td>
<td>81.7</td>
</tr>
<tr>
<td>65</td>
<td>20</td>
<td>0.285</td>
<td>0.324</td>
<td>3.66 4.10 +0.3</td>
<td>106.0</td>
</tr>
<tr>
<td>65*</td>
<td>75*</td>
<td>0.523</td>
<td>-</td>
<td>6.11 - -1.9</td>
<td>151.6</td>
</tr>
<tr>
<td>162</td>
<td>45</td>
<td>0.382</td>
<td>0.368</td>
<td>4.69 4.68 +1.3</td>
<td>90.3</td>
</tr>
<tr>
<td>281</td>
<td>20</td>
<td>0.281</td>
<td>0.306</td>
<td>3.89 4.33 +0.4</td>
<td>78.2</td>
</tr>
<tr>
<td>281</td>
<td>75</td>
<td>0.675</td>
<td>0.617</td>
<td>7.97 7.34 0.0</td>
<td>81.4</td>
</tr>
<tr>
<td>162*</td>
<td>45*</td>
<td>0.389</td>
<td>0.394</td>
<td>5.06 5.17 +0.1</td>
<td>106.6</td>
</tr>
</tbody>
</table>
The results from the water activity and LOD measurements are presented in Table 5. Both water activity and LOD was measured at two occasions, the first measurement was performed right before mixing it with pellets. The second measurement was performed on the MCC that was left after the mixing. The results show that higher humidity gave rise to higher water activity and LOD. The greatest difference occurred between the MCC that was stored at low humidity and the MCC that was stored at high humidity. The difference between the MCC that was stored in low humidity and normal humidity was however not that big, especially when comparing the 2nd measurements. The results shows a difference between the 1st and 2nd measurement for both water activity and LOD. The results show a clear increase in both water activity and LOD during handling of the MCC that had been stored at low humidity. A decrease is noticeable for the MCC with large particles that was kept at high humidity. Since there was not enough material with high moisture content and small particles after mixing it with pellets, it was not possible to perform a second measurement during that trial.

The tablet weight for low dosage was kept at a constant level around a given set point to make sure that the tablets contained the same amount of material and drug substance (Table 5). The weight was allowed to vary within a specific limitation, which was ±5mg from the set point. The tablet hardness that is presented in Table 5 shows that as the particles got smaller the tablet hardness increased.

The trials marked with a * in Table 5 was performed with a 7mm punch, while the rest of the trials were performed with a 8mm punch. The results show that tablet hardness seems to increase for these tablets, which was taken into consideration during the evaluation.

4.2.2 Experiment 2
The second experiment was performed with MCC from Supplier 1 and MCC from Supplier 2, which were all kept at medium humidity. The aim with this experiment was to see how the drug release differs when using different suppliers.

Table 6: Shows the results from water activity and LOD that was measured on the MCC powders before tableting and the results from the IPC as the low dosage tablets had been manufactured. The tablet weight is presented as the difference from the set point in mg.

<table>
<thead>
<tr>
<th>Supplier</th>
<th>Water activity (aw)</th>
<th>LOD (%)</th>
<th>Difference from weight set point (mg)</th>
<th>Tablet hardness (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1st</td>
<td>2nd</td>
<td>1st</td>
<td>2nd</td>
</tr>
<tr>
<td>1</td>
<td>0.374</td>
<td>0.381</td>
<td>4.80</td>
<td>4.71</td>
</tr>
<tr>
<td>1</td>
<td>0.403</td>
<td>0.390</td>
<td>4.96</td>
<td>4.84</td>
</tr>
<tr>
<td>2</td>
<td>0.381</td>
<td>0.372</td>
<td>4.60</td>
<td>4.48</td>
</tr>
<tr>
<td>2</td>
<td>0.401</td>
<td>0.382</td>
<td>4.48</td>
<td>4.31</td>
</tr>
</tbody>
</table>

The results in Table 7 show that the water activity and LOD did not change much during handling of the material, since the first and second values were very similar. The water activity was very similar between the two different MCC powders although the LOD for the MCC from Supplier 2 was a bit lower than the LOD for the MCC Supplier 1. The tablets containing MCC from Supplier 2 also obtained lower tablet hardness than the tablets containing the MCC from Supplier 1.
4.3 Drug release analysis

4.3.1 Experiment 1

The drug release analysis was performed at six different tablets for each trial. During the analysis the drug concentration was measured at 5 different time points and a percentage of the total drug concentration could be calculated. The mean values from these results are presented in Table 7.

Table 7: Shows the results from the drug release analysis after 1-, 4-, 8-, 12- and 20 hours as a percentage of the total drug amount. The presented results are mean values of six different tablets that was analyzed for each trial. The trials marked with * was compressed with a different punch compared to the other trials.

<table>
<thead>
<tr>
<th>Trial Nr:</th>
<th>Particle size (d50)</th>
<th>Moisture content (RH%)</th>
<th>Drug release at 1 h (%)</th>
<th>Drug release at 4 h (%)</th>
<th>Drug release at 8 h (%)</th>
<th>Drug release at 12 h (%)</th>
<th>Drug release at 20 h (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>162</td>
<td>45</td>
<td>10,54</td>
<td>26,58</td>
<td>47,94</td>
<td>68,22</td>
<td>88,62</td>
</tr>
<tr>
<td>2</td>
<td>65</td>
<td>20</td>
<td>6,73</td>
<td>20,97</td>
<td>41,94</td>
<td>63,34</td>
<td>86,1</td>
</tr>
<tr>
<td>3*</td>
<td>65</td>
<td>75</td>
<td>7,16</td>
<td>24,06</td>
<td>46,07</td>
<td>66,15</td>
<td>85,83</td>
</tr>
<tr>
<td>4</td>
<td>162</td>
<td>45</td>
<td>10,21</td>
<td>26,79</td>
<td>47,54</td>
<td>67,36</td>
<td>87,05</td>
</tr>
<tr>
<td>5</td>
<td>281</td>
<td>20</td>
<td>9,62</td>
<td>25,63</td>
<td>46,33</td>
<td>66,16</td>
<td>88,76</td>
</tr>
<tr>
<td>6</td>
<td>281</td>
<td>75</td>
<td>11,43</td>
<td>29,12</td>
<td>50,43</td>
<td>69,72</td>
<td>88,52</td>
</tr>
<tr>
<td>7*</td>
<td>162</td>
<td>45</td>
<td>12,18</td>
<td>31,84</td>
<td>54,93</td>
<td>76,18</td>
<td>96,81</td>
</tr>
</tbody>
</table>

In Figure 13 a graph showing the raw data results from all trials are presented, this graph gives a great overview of the results. What is seen both in Table 7 and in Figure 13 is that the central point in trial nr.7 shows a faster drug release compared to the other central points, trial nr.1 & 4. This is explained by the fact that trial nr.7 was compressed with a different punch compared to the other central points. Because of this, it was decided that trial nr.7 was ignored during the evaluation of the results. Trial nr.3 was compressed with the same punch as trial nr.7, but since it is impossible to draw any conclusions on how the drug release was affected in this case, trial nr.3 is still used in the evaluation.
Figure 13: A graph showing the release profile for all the tablets that were analyzed, six for each trial. The colors correspond to the particle size while the shape corresponds to the moisture content. The green line that shows the fastest drug release corresponds to trial nr. 7 that has been removed in the statistical evaluation.
Because of the decision of removing trial nr.7 from the evaluation, an additional graph of all the raw data, except trial nr.7, was created and is shown in Figure 14. A graph showing the mean values in Table 7 was also created and is shown in Figure 15. Both Figure 14 and Figure 15 shows that larger particles (red color) and higher moisture content (diamond shape) gave rise to a faster drug release. Smaller particles (blue color) and lower moisture content (square shape) gave rise to slower drug release. The largest differences occurs at 4-, 8- and 12- hours, where the drug release could differ up to 15%, which is seen in Figure 14. The mean values in Figure 15 also show the largest differences at 4-, 8- and 12 hours, but between the mean values, the largest difference in drug release was around 10%.

**Figure 14**: A graph showing the release profile for all the tablets except for run nr. 7. The colors correspond to the particle size while the shape corresponds to the moisture content. The graph shows that larger particle size and higher moisture content gives rise to faster drug release. Smaller particles and lower moisture content give rise to slower drug release.
Figure 15: A graph showing the mean drug release for each trial except trial nr. 7. The colors correspond to the particle size while the shape corresponds to the moisture content. The graph shows that larger particle size and higher moisture content gives rise to faster drug release. Smaller particles and lower moisture content give rise to slower drug release.

4.3.2 Experiment 2

Table 8 shows the results from the drug release analysis, the values are a mean of six tablets that were analyzed for each trial. The values in Table 8 indicate that the drug release is very similar between the two suppliers.

Table 8: Shows the results from the drug release analysis after 1-, 4-, 8-, 12- and 20 hours as a percentage of the total drug amount. The presented results are mean values of six different tablets that was analyzed for each trial.

<table>
<thead>
<tr>
<th>Supplier</th>
<th>Drug release at 1 h (%)</th>
<th>Drug release at 4 h (%)</th>
<th>Drug release at 8 h (%)</th>
<th>Drug release at 12 h (%)</th>
<th>Drug release at 20 h (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>9,06</td>
<td>24,25</td>
<td>44,22</td>
<td>63,91</td>
<td>87,93</td>
</tr>
<tr>
<td>1</td>
<td>9,75</td>
<td>25,65</td>
<td>46,52</td>
<td>67,14</td>
<td>90,21</td>
</tr>
<tr>
<td>2</td>
<td>7,59</td>
<td>22,84</td>
<td>44,64</td>
<td>66,16</td>
<td>88,01</td>
</tr>
<tr>
<td>2</td>
<td>7,58</td>
<td>22,83</td>
<td>44,13</td>
<td>65,42</td>
<td>87,43</td>
</tr>
</tbody>
</table>
The graphs in Figure 16 and Figure 17 were created to get a greater overview of the raw data and the mean values. Figure 16 represents all the raw data that was obtained from the drug release analysis, while Figure 17 represents the mean values in Table 8. The red lines represent the tablets containing MCC from Supplier 1 and the blue lines represents the tablets containing MCC from Supplier 2. Both Figure 16 and Figure 17 shows that the drug release for these tablets are very similar, looking at Figure 16 with all raw data, the largest variation in drug release seems to lie around 5%. The mean values in Figure 17 shows less variation in drug release.

![Raw data comparison of different suppliers](image)

*Figure 16: A graph showing the release profile for all the tablets that were analyzed, six for each trial. Red lines corresponds to Supplier 1 and blue lines corresponds to Supplier 2. The release profiles between the two suppliers were very similar.*
4.4 Statistical evaluation

The results from the drug release analysis for Experiment 1, that is presented in Table 7, were used in the statistical evaluation that was performed using regression analysis. Trial nr. 7 has not been used in the statistical evaluation since it showed deviating results. The statistical analysis was performed using Modde 11.

Table 9: The $R^2$ and $Q^2$ values from the statistical analysis. The regression model is very good for the time points 4-, 8- and 12 hours, while the model gets inferior during 1 hour and 20 hours.

<table>
<thead>
<tr>
<th>Time</th>
<th>$R^2$</th>
<th>$Q^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1h</td>
<td>0.902</td>
<td>0.607</td>
</tr>
<tr>
<td>4h</td>
<td>0.998</td>
<td>0.981</td>
</tr>
<tr>
<td>8h</td>
<td>0.998</td>
<td>0.992</td>
</tr>
<tr>
<td>12h</td>
<td>0.978</td>
<td>0.842</td>
</tr>
<tr>
<td>20h</td>
<td>0.787</td>
<td>0.649</td>
</tr>
</tbody>
</table>

Figure 17: A graph showing the mean drug release for the tablets containing MCC from different suppliers. The release profiles were very similar for the two suppliers.
To investigate how useful a regression analysis is, the R² and Q² values that are presented in Table 9, are very important to take into consideration. The R² represents the goodness of fit while the Q² value represents the goodness of prediction. The goodness of fit measures how well the regression model fits the raw data and can vary between 0 and 1, a perfect model has the R² value 1. The goodness of prediction estimates the predictive power of the model, which indicates how well the model predicts results of new experiments. This parameter gives a more realistic and useful indication since the goal of modelling is to estimate how the result will behave during new circumstances. The Q² value varies between 0 and 1, and a model with Q2>0.5 is considered a good model. It is important that the R² value and Q² value does not differ with more than 0.2-0.3 units since that indicates an inappropriate model. (Eriksson, et al., 2009)

As seen in Table 9 the R² and Q² values are close to 1 for the times 4-, 8- and 12 hours, which indicates that the model has a good fit. The times at the edges, 1 hour and 20 hours, shows inferior models since their R² and Q² values are further from the desired value 1. It also shows at 1 hour since the R² and Q² values differ more than 0.3 units.

In Figure 18 the coefficient plots for the drug release at different times are presented. The coefficient plots show which factors that are significant at the specific time. The factors that do not affect the drug release at a specific time have been excluded in the plot since the factor is not significant. During the statistical evaluation a nonlinear relationship was discovered which had to be modeled by adding a square term, PS*PS. The square term represents the nonlinear relationship and was added on the largest factor, which was particle size. It is important to know that the square term could have been added on moisture content as well. To predict exactly which factor that contributes to the nonlinear relationship, further experiments would have to be performed.

The plots in Figure 18 shows which factors that significantly affected the drug release at the different time points. At 4-, 8- and 12 hours the results in Figure 18 show that both particle size and moisture content affected the drug release. There was also a nonlinear relationship present at these times that could either depend on particle size as in Figure 18, but it could equally depend on moisture content. At 1 hour particle size had a significant impact on the drug release and the nonlinear relationship was present at this time as well. At 20 hours, only particle size had a significant impact on the drug release, this indicate that there was a linear relationship between particle size and drug release at this time.
Figure 18: Coefficient plots for the times 1 hours, 4 hours, 8 hours, 12 hours and 20 hours. Showing that particle size affects the drug release at all times, while moisture content affects the drug release at 4-, 8- and 12 hours. A nonlinear relationship is present at all times except at 20 hours where a linear relationship is shown between particle size and drug release.
5. Discussion

5.1 Experiment 1

5.1.1 Solid phase results
The MCC that was sieved and used in Experiment 1 had a distinct difference in particle size distribution (Table 3 & Figure 11), which was desirable to be able to draw conclusions regarding its impact on drug release. Since the particles differed in particle size, this also gave rise to differences in surface area. Larger particles had a smaller surface area while smaller particles had a larger surface area (Table 3). This is explained by the fact that the surface area occupies a larger segment of a small particle compared to a large particle. The result from the bulk density analysis that also is presented in Table 3 shows that MCC that consisted of small particles had a higher bulk density compared to MCC that consisted of large particles. The bulk density was measured by pressing together the material with a specific force, and since smaller particles more easily are packed together this gives rise to a higher bulk density.

5.1.2 Raw data of the drug release
The results in Table 7 show the mean values that were obtained from the drug release analysis. The analysis was performed for six different tablets for each trial to make sure that large variations between tablets within a trial did not occur. The drug release was then measured at 5 different times to obtain a release profile for all trials, which are presented in Figure 13. A deviation that is seen in both Table 7 and Figure 13 is that trial nr. 7, that corresponds to a central point, shows a significant faster drug release compared to the other central points, trial 1 & 4. This was explained by the usage of a different punch during this trial, and because of the difference in drug release, this trial was ignored during the evaluation of the results.

The results from the drug release analysis that are presented in Figure 14 and Figure 19 show that the smaller particles give rise to a slower drug release while larger particles give rise to a faster drug release. This could be explained by the smaller particles’ ability to distribute around the pellets, which protects the pellets from rupturing or sticking together during compression. This theory was also presented in the study by Yao et al. that showed that small particles had a great way of protecting the pellets from damage during compression (Yao, et al., 1997). Larger particles on the other hand are inferior at protecting the pellets, which leads to more damage of the pellets during compression. Beckert, et al., Torrado & Augsburger and Tunón et al. show in their studies that it is important to choose a formulation that protects the pellets well during compression (Beckert, et al., 1996) (Torrado & Augsburger, 1993) (Tunón, et al., 2003). This is essential to be able to keep the wanted drug release profile. Protected pellets will release the drug in the rate of the controlled-release film, which is the wanted profile, while damaged pellets will release the drug faster since the film is no longer intact. Hence, the drug release is slower in the tablets with small particles and faster in tablets with large particles.
5.1.3 Segregation
Deng et al. highlights the risk of segregation within the blend when using particles with large size differences (Kushner, et al., 2011). In the case of Product X the pellets are larger than all fractions of MCC, but the biggest difference is obtained in the mix of small particles together with pellets. The risk that can occur during segregation is content uniformity and uniformity of the weight which can contribute to wrong dosage (Beckert, et al., 1998) (Wagner, et al., 1997) (Yao, et al., 1997) (Yao, et al., 1998). However, this was not noticed during this study, since the weight and drug release were rather stable for the tablets within a trial (Table 5 & Table 7). These results could indicate that segregation in the formulation during compression did not occur, although the particle size of MCC differed from the size of the pellets.

5.1.4 Moisture content
To achieve MCC fractions with different moisture content they were stored at different humidities. To control the moisture content before manufacturing both water activity and LOD was measured on MCC. The measurements were performed at two occasions, once right before mixing it with the pellets, and the other was performed on the MCC that was left after the mixing. This was to be able to see how the moisture content in MCC varied as it was moved to a room with normal humidity that corresponds to 45% RH, since the mixing and manufacturing was performed at 45% RH. The results for both water activity and LOD is presented in Table 5.

The results from the water activity and LOD measurements are presented in Table 5. The water activity shows significant differences in the 1st measurement between the fractions that had been stored in different humidities. The fractions that was stored in 20% RH shows a water activity around 0.3 a_w, the fractions that were stored at 45% RH show a water activity around 0.4 a_w. The two fractions that were kept at 75% RH show higher values compared to the other fractions, but they differ from each other. This could be explained by the usage of different excicators that may have functioned differently. The LOD shows a similar trend during the 1st measurement, higher humidity gives rise to higher LOD values. What is of interest, is what happens in the 2nd measurement for both water activity and LOD. The results that are presented in Table 5 clearly show that differences between the 1st and 2nd measurements occur for both water activity and LOD. Since the time interval between the 1st and 2nd measurements did not differ more than 10-15 minutes, these results indicate that the moisture content in MCC changes very fast as the humidity changes. MCC that was stored in 20% RH showed an increase in water activity and LOD as it was moved to 45% RH during manufacturing. MCC that was stored in 75% RH showed a decrease in water activity and LOD as it was moved to 45% RH during manufacturing. These results demonstrate the difficulty in controlling the moisture content in MCC, since it acclimatized very fast to the environment. The manufacturing of tablets went on for 30-40 minutes, and considering these results it is a great uncertainty in how the moisture content changed during that time.
5.1.5 Tablet hardness
In the study performed by Kushner et al. it was shown that tablet hardness is influenced by the particle size of the excipients. This was also noticed in the IPC results in Table 5 that clearly shows that the tablet hardness increases as the particles got smaller. This is the same conclusion as Nyström, et al. draw in their study, where they found that smaller particles give rise to tablets with higher mechanical strength (Nyström, et al., 1982). It was also noticeable that the tablets that were compressed with a different punch, marked with a * in Table 5, had a significant higher tablet hardness compared to the other tablets. The IPC results in Table 5 also show that tablet hardness increases slightly as the moisture content in MCC gets higher. The largest increase was however noticed for the tablets compressed with another punch. How much of the increase that is due to the moisture content between the trials with small particles and different humidities is hard to say, since they were compressed with different punches. A study by Khan et al. shows that higher moisture content in MCC gives rise to tablets with increased tablet hardness (Khan, et al., 1981). An explanation to this could be that water molecules locate differently in MCC depending on the relative humidity. At higher humidity the water molecules locate on the surface of the MCC particles. This leads to a greater particle to particle interaction due to the ability of forming hydrogen bonds between the particles (Sun, 2016). This gives rise to tablets with increased hardness and mechanical strength.

5.1.6 Disintegration time
Khan et al. show that tablets containing MCC as an excipient obtained differences in disintegration time as the moisture content varied. Higher moisture content gives tablets with longer disintegration time. The results in the study do however show that the disintegration time only differs with a few minutes as the moisture content is varied from around 2 to 8% (Khan, et al., 1981). If changes in disintegration time occur in Product X, differences in a few minutes should not affect the overall drug release that continues for over 20 hours in any great extent.

5.1.7 Drug release and statistical evaluation
The plots that are presented in Figure 14 and Figure 15 indicate that moisture content has an influence on the drug release. When plotting all the raw data it is visible that higher moisture content gave rise to a faster drug release. And vice versa lower moisture content gave rise to slower drug release. It is hard to draw any clear conclusions to why this behavior occurs, but one hypothesis could be that it actually depends on particle size. Since MCC particles swells as the moisture content increases, the particles obtain a larger particle size (Sun, 2008). The results in Figure 14 and Figure 15 show a clear difference in drug release, where larger particles give rise to a faster drug release. Hence, the drug release takes place faster as the moisture content increases, due to the increase in particle size.
To obtain more information on how particle size and moisture content affected the drug release a statistical evaluation using regression analysis was performed. The analysis gave rise to a model that was very good at 4-, 8- and 12 hours which is seen by the high $R^2$ and $Q^2$ values (Table 9). The values in Table 9 shows that the model was inferior at 1 hour and 20 hours due to lower $R^2$ and $Q^2$ values, which was expected. This is because the drug release had just started at 1 hour, which makes it hard to distinguish any variations between the trials. At 20 hours the drug release was almost finished, which makes the release profiles to level off. This analysis gave rise to coefficient plots that are presented in Figure 18. The coefficient plots contain information regarding which material attributes that affect the drug release at the times 1-, 4-, 8-, 12- and 20 hours.

The regression analysis showed that there was no interplay between particle size and moisture content present during the experiments. However, a nonlinear relationship could be detected during the analysis. This was taken into consideration by modelling a square term on the largest factor, which was particle size. It is important to remember that the square term could equally have been modeled on moisture content because there is no information on which of these attributes that actually contributes to the nonlinear relationship. To obtain information about this additional experiments would have to be performed. The coefficient plots in Figure 18 show that particle size had a significant effect on drug release at all times, moisture content did however only affect the drug release significantly at 4-, 8- and 12 hours. The nonlinear relationship seems to be present at all times except at 20 hours. This gave rise to a linear relationship between particle size and moisture content at 20 hours.

During the statistical evaluation attributes like crystallinity and true density have been ignored. This is partly due to the results that are shown in Figure 10, which imply that the sieved MCC fractions contain the same amount of crystalline regions in the structure. This is because the MCC originates from the same batch, and crystallinity rarely variates within a batch. The true density that is presented in Table 3 also shows similar values for the three sieved MCC fractions. Since both these attributes are almost identical for the three different MCC fractions, it is safe to say that variations that occur in the drug release in Experiment 1 do not arise from differences in crystallinity or true density.

5.1.8 Summary Experiment 1

The results that were obtained from this experiment show that both particle size and moisture content of MCC did effect the drug release of Product X (Figure 14, 15 & 18). The largest differences in drug release occurred at 4-, 8- and 12 hours where the drug release could vary up to 15% according to the raw data in Figure 14. Since AstraZeneca has very narrow specification requirements regarding drug release for Product X, these results show a significant variation that would cause problems in the production. This was however expected since this experiment examined MCC fractions that showed clear differences in particle size and some difference in moisture content (Table 3 and Table 5). But since this study shows that particle size was the largest factor according to the regression analysis it could indicate that variations in particle size distribution in the production contributes to the variation in drug release. A hypothesis is that segregation in the MCC material occurs since differences in drug release has been observed during change from one batch to another, and sometimes when using different sections in the same container.
Currently, the particle size distribution is analyzed on around five samples for each batch, which gives a narrow picture of the actual particle size distribution since the batches are delivered in very large volumes. Moisture content was also shown to have an impact on the drug release, during the experiment. It was however noticed how fast the MCC acclimatized to the environment (Table 5). This makes it very hard to control the moisture content in MCC when dealing with large volumes in the manufacturing.

It is important to have in consideration that the results that have been used in the statistical evaluation have some errors due to the usage of different punches. The results for the tablets that were used with another punch are marked with * in Table 5 and Table 7. For example the center point, run nr. 7, has been removed from the statistical evaluation due to deviating values. Trial nr. 3 was also compressed with another punch but is still used in the evaluations since it is impossible to tell how it is affected by the punch. This contributes to some uncertainty in the results that are presented in this study.

5.2 Experiment 2

5.2.1 Solid phase results
The MCC batches that originate from different suppliers show a clear difference in crystallinity that is seen in Figure 10. The MCC from Supplier 2 contained larger regions of crystalline structure compared to the MCC from Supplier 1. The true density was almost identical between the two suppliers and the particle size distribution shows that Supplier 1 had lower d50 and d90 values compared to Supplier 2 (Table 4). Figure 12 shows graphs of the particle size distribution for the two MCC batches. The graphs show that MCC from Supplier 1 had a larger amount of medium particles while MCC from Supplier 2 had a larger amount of small particles. The bulk density was also shown to differ between the two suppliers, where Supplier 1 showed a lower bulk density compared to Supplier 2. The results from this experiment were expected to show if differences in particle size could affect the drug release. The impact of crystallinity was also investigated, since the crystallinity differed significantly between the two batches.

5.2.2 Moisture content
In Table 6, the IPC results are presented together with the results from the water activity and LOD measurements. The water activity was very similar between the two different batches, which was expected since they were stored at the same humidity. The LOD did however show slightly lower values for Supplier 2. This is explained by the difference in crystallinity between the two batches. The MCC from Supplier 2 contained a larger amount of crystalline structure and a smaller amount of amorphous regions. The study performed by Awa et al. shows that MCC binds water in the amorphous regions through hydrogen bonds (Awa, et al., 2015). Because MCC from Supplier 2 has less amount of amorphous parts, a smaller uptake of water occurred. The MCC from Supplier 1 had less crystalline parts and larger amount of amorphous parts, which allowed a greater uptake of water. It is however important to remember that the LOD values only differed around 0.3-0.4% between the two batches, which was not a great difference.
5.2.3 Tablet hardness
The tablet hardness that is presented in Table 6 shows that the tablets that was compressed with MCC from Supplier 1 forms significantly harder tablets. The tablet hardness for these tablets was around 90 N compared to the tablets compressed with MCC from Supplier 2 that was around 65 N. This could partly depend on the fact that the tablets with MCC from Supplier 1 had a higher moisture content (Table 6). Khan et al. explains that MCC with higher moisture content gives rise to tablets with increased tablet hardness compared to tablets containing MCC with less moisture content. Their results do however not show as great differences as in this experiment (Khan, et al., 1981). The large difference must therefore be explained by some other factor as well. Another attribute that is proven to affect the tablet hardness is particle size. Tablets that are compressed with small particles result in harder tablets compared to tablets containing larger particles. (Kushner, et al., 2011) (Nyström, et al., 1982) The MCC from Supplier 1 showed lower $d_{50}$ and $d_{90}$ values, while the $d_{10}$ was higher compared to MCC from Supplier 2 (Table 4). That the $d_{50}$ and $d_{90}$ values were lower for Supplier 1, indicates that Supplier 1 contained smaller particles compared to Supplier 2, which could explain the high tablet hardness.

5.2.4 Drug release
The results from the drug release analyzes that are presented in Table 8, Figure 16 and Figure 17 show that the drug release is very similar between the different suppliers. Supplier 1 shows a slightly faster drug release during the first hours, but after 8 hours the drug release even out and the tablets release the drug at similar speed. These results show that although that there are differences in crystallinity, particle size and bulk density occurred between the two suppliers, the tablets that show very similar release profiles (Figure 16 and Figure 17). This could indicate that the differences in these attributes do not affect the drug release. However, it could also be that they affect the release in different ways, which in the end results in similar release profiles. To obtain more information regarding this, additional experiments would have to be performed.

5.3 Comparison of Experiment 1 and Experiment 2
The results in Experiment 1 show that particle size and moisture content had a significant effect on the drug release of Product X. The largest effect was obtained by varying the particle size (Figure 14 and Figure 15). The results in Experiment 2 show that the drug release between the two suppliers were very similar. It was however hard to draw any clear conclusions regarding how differences in material attribute between the two suppliers affected the drug release of Product X.

Since particle size distribution between the MCC batches from different suppliers does not show as huge differences as in Experiment 1 (Table 3 & 4 and Figure 11 & 12), it would be interesting to know exactly how much the particle size seems to influence the drug release in Experiment 2. This information could however not be obtained by these experiments. But it does seem like small differences in particle size distribution do not give rise to any large differences in drug release. The MCC that is used during manufacturing of Product X is controlled before usage, and the particle size distribution is not allowed to vary in any great extent. The results from the two experiments in this study indicate that large variations in particle size have a significant effect on the drug release, but it is uncertain how small variations in particle size distribution would affect the drug release. Hence, there could be other factors that also contribute to the large variations that can occur during production.
6. Conclusion

The aim of this study was to investigate which material attributes of MCC that affect the drug release of Product X. This was obtained by performing two separate experiments, where Experiment 1 investigated the impact of particle size and moisture content using DoE. Experiment 2 aimed to compare the drug release between tablets that had been compressed with MCC from two different suppliers. The result from Experiment 1 showed that both particle size and moisture content had a significant effect on the drug release of Product X. Larger particles and higher moisture content gave rise to a faster drug release. Smaller particles and lower moisture content gave rise to a slower drug release. There was however, no interplay present between particle size and moisture content, but a nonlinear relationship was observed during the statistical analysis. To obtain information about which of the attributes that contributed to the nonlinear relationship further experiments would have to be performed. The results from Experiment 2 showed small variations in drug release, even though the MCC from the different suppliers varied in crystallinity, particle size distribution and bulk density. It was however hard to draw any clear conclusions regarding how these attributes affect the drug release of Product X.

Altogether, the study shows that large variations in particle size and moisture content in the material MCC give rise to significant variations in drug release of Product X. It is however hard to tell how small variations in particle size distribution of MCC affect the drug release of Product X.

7. Further recommendations

The next step in the process of investigating how MCC affects the drug release of Product X is to analyze the results from the full dosage study that was excluded in this report. This was because the results could not be obtained in time, since the laboratory that performed the analysis had too much work and could not prioritize the full dosage samples. The results from these samples will however be interesting to analyze since the same punch was used throughout the manufacturing. This will lead to less uncertainty in the results, compared to the results in this study that have some uncertainties due to the usage of different punches.

AstraZeneca should also consider to analyze particle size distribution and moisture content of MCC more often in connection to manufacturing, to see if any correlation occur between these attributes and drug release are visible. This is of interest to see if the variations that occur in MCC today, regarding particle size distribution and moisture content, seems to contribute to the variations that occur in the production.

Analysis that could be of interest is to measure particle size distribution at different sections of the same container of MCC. This could give indications in how large variations that occur within a batch regarding particle size distribution. The analysis could also provide information regarding if MCC segregates within the container.
8. Acknowledgement

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References


Appendix 1

**AstraZeneca**

AstraZeneca is a global pharmaceutical company that conducts both research and manufacturing of pharmaceuticals. AstraZeneca has chosen to put their main focus on three disease areas which are cardiovascular and metabolic diseases, oncology and respiratory, inflammation and autoimmunity. AstraZeneca also carries out research regarding the disease areas infection and neuroscience.

The company operates in more than hundred countries worldwide and has around 61 500 employees. There are manufacturing sites in seventeen countries and the largest site is placed in Södertälje. The tablet manufacturing in Södertälje produces around 10 billion tablets every year to markets all over the world. The research centers are placed in Sweden, USA and the United Kingdoms.

In Sweden there are 6 600 employees where 4 100 are stationed at the manufacturing sites in Södertälje and 2 500 are stationed at the research center in Göteborg. (AztraZeneca, u.d.)

**Good Manufacturing Practice (GMP)**

Good Manufacturing Practice (GMP) is a way of working during manufacturing of pharmaceuticals to assure good quality products. The Current Good Manufacturing Practice (CGMP) refers to a standard that is used by the US Food and Drug Administration (FDA) to ensure good pharmaceutical quality. CGMPs regulations assure that the processes of manufacturing pharmaceuticals reach certain requirements. These requirements include that the processes should have proper design, monitoring and controls, of both the production process and facilities. Pharmaceuticals that are manufactured according to CGMP assures products with identity, strength, quality and purity. This is obtained by using strong quality management systems, using raw materials of appropriate quality and robust operating procedures, detecting and investigating deviations that affect product quality and assuring reliable testing laboratories. By implementing systems that control this, contaminations, mix-ups, deviations, failures and errors can be prevented. The “C” in GMP stands for Current and implies that technologies and systems that are up-to-date should be used to comply with the regulations. (ISPE, u.d.)
Appendix II

Theory for some methods that were used during this project

**QicPic – Particle size**
The QicPic measures the particle size distribution for different materials. The instrument uses the technique of a high-speed camera that can take up to 450 pictures per second together with a powerful disperser that separates the particles. This makes it possible to capture each particle separately without having to deal with overlapping particles. To obtain good resolution a specially developed pulse light source is used to reduce the motion blur. A special imaging objective that only transmits light rays that are parallel to the optical axis generates black dots that correspond to particles. The high-speed camera then captures the black dots and the results are stored in the database of the PC. The software Windox 5.0 then uses algorithms to evaluate the results from the measurement. (Sympatec, u.d.)

**Mastersizer 3000 – Particle size**
The Mastersizer 3000 (Malvern) uses the technique of laser diffraction to measure particle size of powders. The feeder Aero S (Malvern) that uses compressed air to accelerate the particles, dispersed the powder into the instrument. By vacuum the particles flows through the instrument into the measurement cell of glass (Malvern, 2016). As the sample reaches the measurement cell, laser illuminates the sample and depending on the angle of the scattered light the particle size can be calculated. Larger particles give rise to a smaller angle between the scattered light while smaller particles give rise to a larger angle between the scattered light as seen in Figure 8. The samples of MCC contained particles of different sizes that gave rise to a scatter pattern that was used to calculate the particle size distribution of the samples. (Malvern, 2016)

![Figure 19: How the Mastersizer 3000 analyzes particle size with laser beams. Larger particles give rise to a smaller angle between the scattered light while smaller particles give rise to a larger angle between the scattered light.](image-url)

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**X`Pert Pro - Crystallinity**

The instrument (X`Pert Pro, PANalytical) uses the technique of X-rays and the phase shifts that can occur between X-ray waves. X-rays are electromagnetic waves that has a shorter wavelength than visible light. The wave can be described as a sine wave that repeats periodically every $2\pi$ radians. The wavelength of electromagnetic waves are described as the spatial length of each period. Identical waves that are not moving equally are said to have a phase shift (Figure 20). The phase shift can be measured as a linear shift, $\Delta$ with the unit of the wavelength.

![Phase shift, $\Delta$](image)

*Figure 20: A graph that illustrates a phase shift that is used in the evaluation of the scattered light.*

The principle of X-ray diffraction uses phase shifts to determine if the material has a crystalline structure. It is explained by using Bragg’s equation (1) that describes the X-ray diffraction as a set of lattice planes that reflects the X-ray. When equation (1) is fulfilled, the reflected X-ray waves moves in phase (Figure 22).

\[
\Delta = n\lambda = 2d \sin \theta \quad (1)
\]

The $d$ corresponds to the distance between the parallel planes and $2\Theta$ corresponds to the diffraction angle (Figure 21). The $n$ in this equation corresponds to an integer, in the cases where $n$ corresponds to a decimal equation (2) holds. Equation (2) describes the reflected X-ray waves that do not move in phase, which leads to destructive interference (Figure 22).

\[
n\lambda = p\Delta \quad (2)
\]
The p corresponds to a deeper plane, which the X-ray can reach. (Dinnebier & Billinge, 2008)

Figure 21: A figure showing the angles and distances that is used in Bragg’s equation (1). The d corresponds to the distance between the parallel planes and \(2\Theta\) corresponds to the diffraction angle.

Crystalline materials get an intensity maximum exactly in the direction where equation (1) applies (Figure 23). All other directions suffer from destructive interference described by equation (2) which generates no intensity at all. Amorphous materials on the other hand generate a diffuse scattering, which shows diffracted intensity in all directions (Figure 23). Since crystalline and amorphous materials give rise to different intensity curves it is possible to determine if the material has a crystalline or amorphous structure. (Dinnebier & Billinge, 2008)

Figure 22: An illustration of X-rays that are in phase to the left, and X-rays that are not in phase to the right.

Figure 23: An illustration of how the intensity graphs will look like depending on if the material is crystalline or not. Crystalline materials give rise to distinct intensity tops that are seen in the graph on the left hand, while amorphous materials show intensity in all directions, which give rise to the graph on the right hand.