Formulation of a chitosan multi-unit dosage form for drug delivery to the colon

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To God

In difficult times when everybody else failed me and left me to carry on alone, He carried me and gave me His biggest present. His love and comfort. And Philip.

To the following persons He chose to cross my path and whose help made this study possible:

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Ek slaan my oë op na die berge: waar sal my hulp vandaan kom? My hulp is van die Here wat hemel en aarde gemaak het.
Psalm 121

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ABSTRACT

In some diseases it is preferable that the drugs used in their reatment are released in the colon. The colon is also suitable for systemic delivery of a variety of drugs. A variety of systems have been developed for the purpose of achieving colonic targeting. These approaches are either drug-specific (prodrugs) or formulation specific (coated or matrix preparations) and depends on the pH, transit time and pressure or bacteria in the colon. Different polymers, like chitosan, have been evaluated for their susceptibility to degradation by these bacterial enzymes. Chitosan is considered a good candidate for bacterial degradation and is widely available at low cost and has favourable biological properties.

To investigate the influence of formulation factors on the properties of chitosan minitablets, it was necessary to ensure that the chitosan had satisfactory powder flow characteristics to ensure uniform compression in the tablet press and to prevent unacceptable variation in the tablet properties such as weight, thickness, disintegration and strength. Moisture content of the powder, particle size and the inclusion of glidants had an effect on the flowability and it could be improved from a composite flow index value of 32.7 to a value of 58.8.

The compressibility of chitosan is very poor and different factors that might influence it, was investigated. Compression forces of between 15 and 20 bar resulted in tablets with acceptable physical characteristics. An increase in moisture content, using the powder fraction > 212 µm as well as a decrease in powder weight resulted in tablets with a higher tensile strength.

Lower compression forces resulted in tablets that are extremely porous. This suggests that the chitosan can only be compressed at high compression forces which are difficult to obtain using a standard tablet press. The standard tablet press was therefore modified to fill more powder in the die and generate higher compression forces.

Minitablets were compressed and the dissolution of isoniazide from these tablets was investigated. Varying the punch depth or the compaction of the powder did not result in the desired slower release of the drug as a result. The porosity of the tablets compressed at all the punch depth settings and compaction percentages was probably too high to have an effect on the wettablity of the tablets and as a result on the dissolution of the isoniazide from the tablets. The inclusion of excipients such as citric acid (an organic acid which would lower the pH in the tablet, allowing the chitosan to form a gel) and pectin (which would form an insoluble complex with the chitosan) into the formulation delayed the dissolution of the isoniazide from the minitablets.

Coating of the minitablets with an enteric coating (Eudragit S ®) initially delayed the dissolution of the isoniazide and would protect the tablets from the harsh environment of the stomach so that the tablets will reach the colon and release the drug.

Key words: Chitosan, minitablets, flowability, compressibility, dissolution, isoniazide

OPSOMMING

In sommige siektetoestande is dit verkieslik dat die geneesmiddel wat in die behandeling gebruik word, in die kolon vrygestel word. Die kolon is ook geskik vir geneesmiddel aflewering van 'n verskeidenheid sistemies werkende geneesmiddels. 'n Verskeidenheid sisteme is ontwikkel vir die hierdie doel. Hierdie benaderings is of geneesmiddelspesifiek (progeneesmiddels) of formuleringspesifiek (bedekte of matriks preparate) en is afhanklik van die pH, deurgangstyd deur die spysverteringskanaal, druk of bakterië wat in die kolon voorkom. Verskillende polimere, waaronder kitosaan, is ge-evalueer vir hulle vatbaarheid vir afbraak deur hierdie bakteriele ensieme. Kitosaan is geskik vir sulke afbraak en is maklik beskikbaar teen 'n lae koste en het ook gunstige biologiese eienskappe.

Om die invloed van formulerings faktore op die eienskappe van kitosaan minitablette te ondersoek, was dit nodig dat die kitosaan bevredigende vloei eienskappe vertoon om uniforme samepersing te verseker en sodoende onaanvaarbare variasie in die massa, dikte, disintegrasie en breeksterkte van die tablet te voorkom. Die voggehalte van die poeier, die deeltjiegrootte daarvan en die byvoeging van glymiddels het 'n effek gehad op die vloeibaarheid en kon die saamgestelde vloei indeks vehoog vanaf 'n waarde van 32.7 tot 58.8.

Die saampersbaarheid van kitosaan is baie swak en verskillende faktore wat dit kan beïnvloed is ondersoek. Drukke van tussen 15 en 20 bar het tablette met aanvaarbare tableteienskappe opgelewer. 'n Verhoging in die voggehalte, die gebruik van die deeltiegroottefraksie > 212 μm en die afname in die poeier massa het tablette met 'n hoër breeksterkte tot gevolg gehad. Laer drukke het baie poreuse tablette opgelewer. Dit is 'n aanduiding dat kitosaan slegs by hoë drukke, wat moeilik verkry kan word in standaard tabletperse, saamgepers kan word. Die standaard pers is daarom verander sodat meer poeier in die matrys gevul kon word en sodoende groter drukke deur die pers daarop uitgeoefen kon word.

Minitablette is saamgepers en die dissolusie van isoniazide uit die tablettte is ondersoek. 'n Verandering in die slaglengte sowel as die kompaksie van die poeier

het nie die vrystelling van die geneesmiddel vertraag nie. Die poreusheid van die tablette by al die verskillende slaglengtes en kompaksies was waarskynlik te hoog om 'n effek op die benatbaarheid van die tablette en die dissolusie eienskappe van die geneesmiddel te vertoon. Die insluiting van hulpstowwe soos sitroensuur ('n organiese suur wat die pH in die tablet verlaag en sodoende veroorsaak dat die kitosaan 'n gel vorm) en pektien (wat 'n onoplosbare kompleks met die kitosaan vorm) in die formulering, het stadiger vrystelling van die isoniasied tot gevolg gehad.

Die bedekking van die minitablette met 'n enteriese bedekking (Eudragit S®) het die aanvanklike vrystelling vertraag en beskerm die tablet teen die suuromgewing van die maag sodat die tablette die kolon kan bereik en die geneesmiddel daar vrystel.

Sleutelwoorde: Kitosaan, minitablette, vloeibaarheid, saampersbaarheid, dissolusie, isoniasied.

AIM AND OBJECTIVES OF THIS INVESTIGATION

AIM OF THE STUDY

The aim of the study is to investigate different physical, tableting and formulation factors that influence chitosan minitablets as a means to produce a colon-specific multi-unit dosage form.

BACKGROUND

Drug delivery to the colon

Colonic targeting has gained increasing interest for a number of years. A considerable number of publications dealing with colon targeting, colon-specific drug delivery and absorption from the cecum and other colon sections indicate a growing focus of research in this area (Bauer, 2001:31). Until recently the colon was considered a site for water reabsorption and carbohydrate fermentation. Recently, colonic drug delivery systems have attracted a great deal of interest, not only for the localized treatment of diseases but also for the systemic delivery of drugs (Sinha & Kumria, 2002:23).

The reason for the interest in the colon as a site for drug delivery can be ascribed to

1) a less hostile environment for drugs due to an almost neutral pH and a low diversity and intensity of digestive enzymes,
2) a transit time of up to 78 hours which increases the time for drug absorption,
3) better response to absorption enhancers and
4) lower doses, and therefore lower side effects associated with the treatment of localized diseases such as Crohn's disease and ulcerative colitis (Sinha & Kumria, 2002:24).

The interest has further been stimulated by the development of new therapeutic agents for the treatment of colonic diseases that has required colon-specific delivery systems to maximize the effectiveness of these drugs. Other factors such as the

desire to produce oral delivery systems for therapeutic peptides and proteins and the introduction of once-a-day sustained release formulations, that have required a better understanding of the transit of dosage forms through the colon, and of the colonic absorption of the drugs contained within them, also contributed to the interest (Watts & Illum, 1997:893).

To achieve drug delivery to the colon, it is necessary to develop a dosage form with the ability to deliver drugs in the colon after withstanding the hostile environment of the stomach and passing through the ileo-cecal junction. Different approaches have been used i.e. time-, pH- and bacteriological dependant systems (Watts & Illum, 1997:893).

Chitosan for colonic drug delivery

Chitin is the second most abundant polysaccharide in nature, after cellulose, and is found in the exoskeleton of crustaceans, insects and some fungi. Chitosan is obtained by the partial alkaline deacetylation of chitin from crustacean shells (Illum, 1998:1326). Because chitosan has favorable biological properties such as biodegradability, biocompatibility and low toxicity, it has attracted a lot of attention in the pharmaceutical field. The polycationic character of chitosan enables it to bind strongly to mammalian cells and mucus (Felt *et al.*, 1998:980).

Chitosan has been used in oral drug formulations in various ways such as to provide sustained release and to increase absorption of drugs. Recent studies showed that chitosan is readily degraded by the microflora in the colon. This property, together with its bioadhesive nature, makes chitosan a promising agent for colonic drug delivery. There are however problems associated with the use of chitosan as an excipient in the formulation of tablets such as the very poor flowability and poor compressibility of the chitosan powder.

Chitosan is soluble in dilute acid and precipitates at a pH above 7. Because of the solubility of chitosan at low pH ranges, its successful use in colon-specific delivery requires an enteric layer over the chitosan, which would protect it against the acidity in the stomach (Sinha & Kumria, 2002:23). As the formulation reaches the intestine

the pH increases and the enteric layer dissolves, releasing the chitosan. The micro flora in the colon will then degrade the chitosan and release the drug. Several researchers have used chitosan for the development of dosage forms for drug delivery to the colon (Hejazi & Amij, 2003:154, Munjeri *et al.*, 1997:273, Tozaki *et al.*, 1997:1016).

Multiparticulate systems

The administration of multiparticulate systems (such as pellets, beads or minitablets normally filled into hard capsules) offers several advantages over conventional single-unit matrix formulations. These include less risk of dose-dumping, less inter- and intra-subject variability and a higher degree of dispersion in the gastro-intestinal tract, thus minimizing irritation associated with high local drug concentrations (De Brabander *et al.*, 2000:82). These gelatin capsules or the small dosage forms themselves need to be protected from the environment of the stomach in an attempt to reach the intestines. Various enteric coatings can be applied for such a purpose, such as the acrylates, shellac and cellulose derivatives (Felton *et al*, 1995:17).

OBJECTIVES

To achieve the aim of the study, the following experiments will be undertaken:

- The investigation of the physical properties influencing the tableting of chitosan, such as the absorption of moisture, flowability and compression force to establish the ideal tableting conditions for chitosan.

- The preparation of different chitosan minitablets. These minitablets will be prepared using different formulations and methods and then evaluated for their use in a colon-specific drug delivery system.

- The preparation of enteric-coated minitablets and the *in-vitro* evaluation of these tablets to evaluate their effectiveness as colon-specific drug delivery systems.

CHAPTER 1

COLONIC DRUG DELIVERY

1.1 INTRODUCTION

Colonic targeting has gained interest over the past years. A considerable number of publications dealing with colon-specific drug delivery and absorption from the cecum and other colon sections indicate a growing focus in this area of research (Bauer, 2001:31).

Until recently the colon was considered as a site for water and electrolyte reabsorption, carbohydrate fermentation and the formation, storage and expulsion of faecal material. The colon is vulnerable to a number of disorders including ulcerative colitis, Crohn's disease, irritable bowel syndrome and carcinoma (Basit & Bloor, 2003:185). Colonic drug delivery systems have therefore lately attracted a great deal of interest not only for the local treatment of a variety of local diseases but also for the systemic delivery of drugs (Sinha & Kumria, 2002:23, Sinha *et al.*, 2004:101).

The reason for the interest in the colon as a site for drug delivery can be ascribed to the colon being a less hostile environment for drugs due to an almost neutral pH and low diversity and intensity of digestive enzymes. The transit time of up to 78 hours through the colon, increases the time for drug absorption. Other factors contributing to an increased interest of colon-specific drug delivery are:

i) a better response to absorption enhancers and lower doses and side effects associated with the treatment of localized diseases such as Crohn's disease and ulcerative colitis (Sinha & Kumria, 2002:24),
ii) the development of therapeutic agents for the treatment of colonic diseases that has required colon-specific delivery systems to maximize the effectiveness of these drugs,
iii) the desire to produce oral delivery systems for therapeutic peptides and proteins and

iv) the introduction of once-a-day sustained release formulations that has required a better understanding of the transit of dosage forms through the colon, and of the colonic absorption of the drugs contained within them (Watts & Illum, 1997:893).

Colonic delivery of drugs may also be useful in the treatment of diseases susceptible to the diurnal rhythm such as asthma, arthritis and inflammation (Lorenzo-Lamosa *et al.*, 1998:110). For example, the incidence of asthmatic attacks is the greatest during the early hours of the morning. Colon-specific formulations having a prolonged drug delivery might be an ideal delivery system in such cases.

A local means of drug delivery could allow topical treatment of inflammatory bowel disease, e.g. ulcerative colitis or Crohn's disease. Such inflammatory conditions are usually treated with glucocorticoids, sulphasalazine or 5-aminosalicylic acid and treatment might be more effective if these drug substances were targeted directly to the site of action in the colon. Lower doses might be adequate in such cases and systemic side will therefore be reduced.

1.2 STRUCTURE AND FUNCTION OF THE COLON

1.2.1 Morphology of the gastro intestinal tract

The colon forms the lower part of the gastro intestinal tract (GIT) and extends from the ileocecal valve to the anus as depicted in figure 1.1.

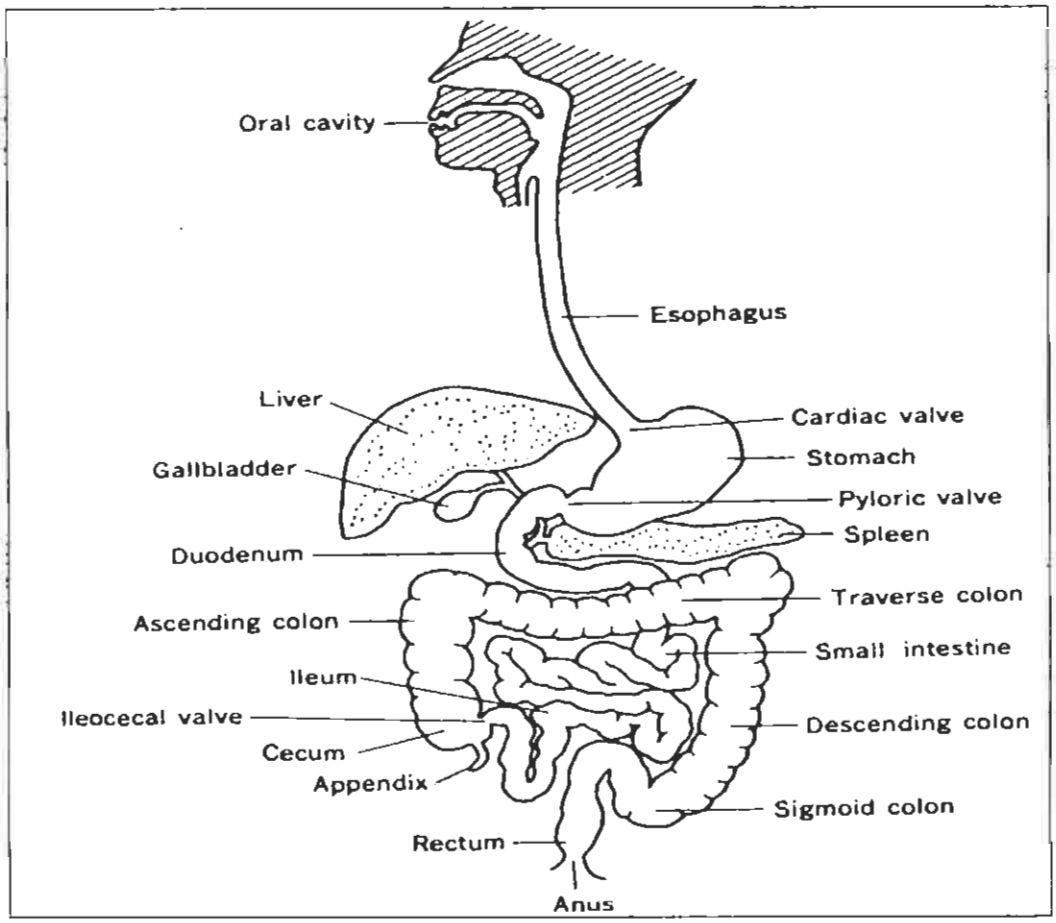

Figure 1.1: *A schematic drawing of the human gastro intestinal tract (Haeberlin & Friend, 1992:4).*

The large intestine is divided into the appendix, cecum, colon (ascending, transverse, descending and recto sigmoid), rectum and anal canal. The length of the large intestine is approximately 1.5 m in humans (Haeberlin & Friend, 1992:10).

Colonic movements produce motility patterns to maintain the following three functions: i) conservation of water, ii) the maintenance of an abundance of intraluminal bacterial population and iii) the capacity to control the delivery of faeces. The amount of material in the human colon is surprisingly small. On average the colon contains only about 220g of wet material, equivalent to 35g of dry matter, of which up to one third can be bacterial mass (Haeberlin & Friend, 1992:18).

1.2.2 Intestinal flora

The slow movement of material through the colon allows for a large microbial population to thrive there and more than 400 species of bacteria as well as a small number of fungi are found. In addition to food, saliva is one of the main sources of bacteria entering the intestine. About 10^7 colony forming units/ml (cfu/ml) is found in the saliva (Haeberlin & Friend, 1992:18).

Because of the pH of the empty human stomach of less than 3, only small numbers of bacteria inhibit the stomach. When food is present, the gastric pH is raised to above 4 and bacteria from the mouth and the ingested food proliferate to 10^4 - 10^8 cfu/ml. As soon as the pH of the stomach content drops due to mixing with gastric juice, the acid-sensitive bacteria die and the bacterial count decreases (Haeberlin & Friend, 1992:18).

The microflora of the proximal small intestine is similar to that of the stomach (10^3 - 10^4 cfu/ml) because of chemical and physical factors such as bile juice and peristalsis which tends to remove microflora at rates faster than they can reproduce. In the lower part of the small intestine, the number of bacteria increases to between 10^5 and 10^7 cfu/ml due to neutralization of gastric juice and lowered transit speed. Distal to the ileocecal sphincter, the bacterial concentration increases sharply to between 10^{11} - 10^{12} cfu/ml. The principle source of nutrition for the colonic micro-organisms is carbohydrates arriving in the proximal colon and the number of organisms are therefore the highest in that section of the colon (Haeberlin & Friend, 1992:18).

Drug delivery systems to the colon relying on enzymatic degradation is based on this sharp increase in the bacterial count in the colon compared to that of the stomach and small intestine.

1.2.3 Biotransformation by the intestinal microflora

There are two biotransformation reactions by the intestinal microflora namely hydrolytic and reductive reactions.

1.2.3.1 Hydrolytic reactions

The main hydrolytic enzymes produced by the intestinal bacteria are β-glucoronidase, β-glycosidase and β-galactosidase. The principle sources of nutrition for the bacteria are carbohydrates and dietary fibers that are not digested by secretions of the stomach and small intestines. These fibers are then degraded by the bacterial fermentation in the ascending colon (Haeberlin & Friend, 1992:22). These carbohydrates include starch, non-starch polysaccharides and oligosaccharides such as lactose, sorbitol and xylitol. They are degraded by the enzymes to produce short chain fatty acids, carbon dioxide, hydrogen, methane and hydrogen sulphide.

Protein digestion also occurs in the colon, although to a much lesser extent than in the ileum that contains peptidase. These proteins are comprised of dietary proteins as well as from pancreatic and small intestine enzymes. From within the colon there are sloughed colonic epithelial cells and proteins and peptides released from bacteria. Metabolic products include organic acids, hydrogen, carbon dioxide, ammonia, phenols and indoles. The success of colonic delivery of peptides and proteins will need to overcome this protease activity in the colon (Haeberlin & Friend, 1992:24).

1.2.3.2 Reductive reactions

Common reductive reactions by the intestinal flora include nitro group reduction (nitroreductase) azo group reduction (azoreductase) and azo bond cleavage. These reactions are important in the action of sulphasalazine and 5-aminosalicylic acid (5-ASA) used in inflammatory bowel disease (see 1.7.1.1).

1.3 pH OF THE COLON

Local pH within the lumen of the GIT can directly affect delivery systems that rely on enteric coatings, and indirectly influence them by altering local enzymatic activity. Since the pH gradient along the GIT forms the basis of several targeted lower

intestinal delivery systems, understanding the variation in this gradient is important in health and disease (Friend, 2005:248).

The highest pH levels in the gastro intestinal tract are found in the terminal part of the ileum (7.5 ± 0.5). On entry into the colon, carbohydrate fermentation predominates and results in a lower pH of 6.4 ± 0.6. This low pH inhibits the photolytic enzymes. In the distal regions of the colon there is little carbohydrate fermentation, resulting in higher pH levels (6.6 ± 0.8 in the transverse colon and 7.0 ± 0.7 in the descending colon) and therefore increased levels of protein digestion (Watts & Illum, 1997:895).

The relatively high value of the pH preceding and within the colon has led to the development and synthesis of polymers that dissolve at a pH around 7. These consist of copolymers of methacrylic acid, methylmetacrylate and ethylacrylate, such as Eudragit® (Vandamme *et al.*, 2002:220).

1.3.1 Effect of diet and drugs on the pH in the colon

The fall in the pH in the proximal colon is due to the presence of short chained fatty acids arising from the bacterial fermentation of polysaccharides. Consequently, polysaccharide drugs and diet can affect the colonic pH. For example, lactulose, a semi-synthetic disaccharide used as a laxative, is fermented by the colonic bacteria to produce large amounts of lactic acid. This results in a drop in the pH of the colon to approximately 5.0. Other pharmaceutical polysaccharides like ispaghula, guar gum and chitosan as well as a high fiber diet will have the same effect (Watts & Illum, 1997:895).

1.3.2 Effect of disease on the pH in the colon

The luminal pH of the distal intestine in patients with inflammatory bowel disease (IBD) can be lower than seen in healthy volunteers. In one study involving six patients with ulcerative colitis, the luminal pH was highly variable. Three patients had a colonic pH ranging from 5.0 to 7.0 while the other patients had a pH ranging from 2.3 to 3.4. In patients with Crohn's disease, relatively low pH values were also

measured (5.3 ± 0.3) in the right colon and were more acidic in the distal colon (Friend, 2005:249).

1.4 COLONIC TRANSIT OF MATERIALS

Intestinal transit time is important for nearly all orally targeting delivery systems. Gastric emptying of dosage forms is highly variable and depends primarily on the presence of food in the stomach. In various studies, gastric residence varied between 15 minutes and 12 hours. Small intestine transit is surprisingly constant at 3 - 4 hours and appears to be independent of food. Compared to other regions of the GIT, the movement of materials through the colon is slow and the total transit time is highly variable and influenced by a number of factors such as diet, mobility, stress, disease and drugs. Transit time varied between an average of 20,9 hours to 35 hours in some studies, while in one subject the tablet moved through the colon in just 2,5 hours (Watts & Illum, 1997:897).

1.4.1 The effect of physical factors of a dosage form on transit time

There have been a number of studies investigating the effect of the size of a dosage form on the rate it moves through the colon. The results of these studies would suggest that smaller units travel through the colon more slowly than larger ones. Hence, additional retention of a dosage form within the colon could perhaps be achieved by the use of a multiparticulate formulation, rather than a large single unit. Consequently, there may be advantages in formulating a dosage form as a multiparticulate rather than a single unit to ensure that it does not pass too rapidly through the colon and be excreted before all of the drug has been released (Watts & Illum, 1997:897).

Studies on the effect of density and capsule size of the drug delivery system on colonic transit have also been performed (Parker *et al.*, 1988:376). Density did not effect the transit time through the ascending colon and no significant change was detected with an increase in volume. In another study however, it was suggested that colonic transit of tablets was volume dependant (Adkin *et al.*, 1993:155) while Clark

et al. (1995:9) concluded that there is a critical density at which prolonged gastrointestinal residence time is achieved.

1.4.2 Effect of disease on colonic transit

Diseases affecting colonic transit have important implications for drug delivery; diarrhea will result in an increase in motility while constipation will result in a decrease in motility. Diseases such as Crohn's disease and ulcerative colitis are associated with symptoms such as abdominal pain, distention and altered transit (diarrhea or constipation). In one study, the residence time of individual tablets in the ascending colon of patients with ulcerative colitis varied from as little as 0,8 hours to more than 20 hours (Hardy *et al.*, 1988:82). Other studies showed little difference in overall transit times between healthy patients and patients with IBD, although the transit times were slower through the proximal colon but accelerated through the recto-sigmoid region of the colon (Friend, 2005:250).

1.4.3 Effect of diet on colonic transit

The principle dietary component which can affect colonic motility is dietary fiber. Dietary fiber supplementation increases fecal weight, by retention of water and by increasing bacterial mass, thereby reducing colonic transit times. The ingestion of food is known to stimulate colonic activity termed the "gastro colonic response". In a study by Price *et al.* (1993:1015), volunteers each received five 6 mm tablets. Upon reaching the ileocecal region, they also received a high-protein or a high-fat meal. Ingestion of the food was followed by an increased movement through the ileocecal junction but was not influenced by the type of meal.

1.5 DRUG DISSOLUTION IN THE COLON

As a rule, a drug must be in solution before it can be absorbed from the lumen of the GIT. In the more distal portions of the GIT, conditions are heterogeneous and drug dissolution is subject to the high viscosity of colonic contents. While not significantly

affecting the dissolution of water soluble drugs, viscous luminal contents in the colon can impede dissolution of drugs that are less water soluble (Friend, 2005:250).

1.6 ABSORPTION OF DRUGS FROM THE COLON

1.6.1 Routes and mechanism of absorption from the colon

The primary routes by which drugs are absorbed from the GIT are illustrated in figure 1.2.

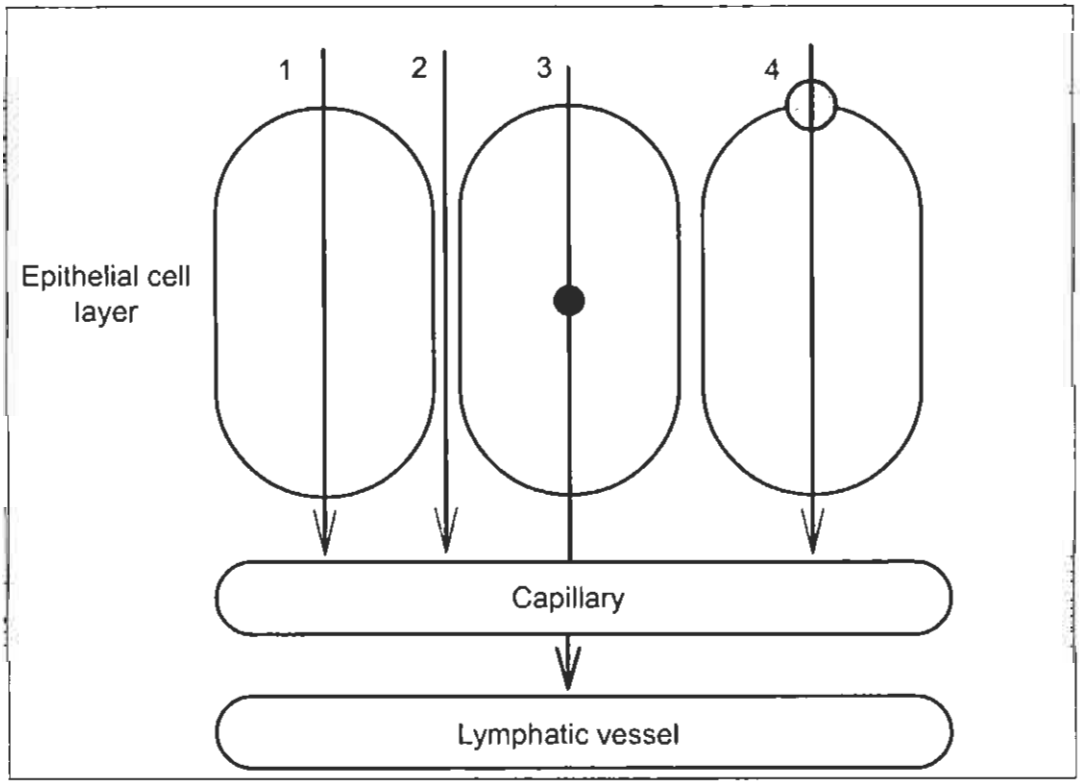

Figure 1.2: *Illustration of the main pathways of intestinal drug absorption: 1) transcellular absorption 2) paracellular absorption 3) transcellular absorption followed by incorporation into chylomicrons and transport into the lymphatic system and 4) active transport (Watts & Illum, 1997:898).*

The vast majority of drugs are absorbed from the colon by passive diffusion either through paracellular or transcellular routes. Transcellular absorption involves the passage of drugs through cells and this is the route most lipophyllic drugs will take, whereas paracellular absorption involves the transport of the drug through the tight

junctions between cells and is the route most hydrophilic drugs follow. There are, however, some exceptions. Certain drugs have chemical structures which will allow them to be carried across the intestinal wall by an active transport mechanism while some drugs with very high lipophylicity may be incorporated into chylomicrons inside the intestinal epithelial cells and absorbed into the systemic circulation via the lymphatic system (Watts & Illum, 1997:898).

1.6.2 Factors affecting colonic drug absorption

Studies have shown that paracellular absorption is constant throughout the small and large intestines, but transcellular absorption appears to be confined to the small intestine with negligible absorption by this route in the colon. The poor paracellular absorption of many drugs in the colon is due to the very tight epithelial cell junctions. For this reason, a variety of methods to enhance colonic permeability, mostly through the use of chemical enhancers, have been explored (Mrsny, 1992:23).

The colon also has a much smaller surface area compared to that of the small intestine. This is compensated for by a slower rate of transit through the colon allowing drug to stay in contact with the mucosa for a longer period.

Passive and active transport in the colon results in the net secretion of potassium and bicarbonate and the net absorption of sodium and chloride. Water passively follows the uptake of sodium and chloride causing a progressively more viscous colon content. This will theoretically reduce the dissolution rate of the drug and slow the diffusion of the dissolved drug to the mucosa (Watts & Illum, 1997:898).

The mucus barrier at the epithelial surface can also be a formidable physical barrier preventing uptake as a result of drug binding. The mucus is highly charged sieve like in nature and these factors can contribute in the absorption of large negatively charged molecules. The thickness of this mucus barrier will also have an effect as the transit through the barrier is diffusion limited. Certain drugs stimulate mucus secretion (for example carbachol) and will impede their own absorption. Mucus secretion is also elevated during intestinal infection while other pathological

conditions like inflammatory bowel disease can cause alteration in this layer (Mrsny, 1992:16).

The most important factor influencing the absorption in the colon is probably the inter-patient and intra-patient variability in the gastro-intestinal pH and transit times (as discussed above). Various other factors such as thickness of the water layer between the mucus and the epithelial cells, the presence of prostaglandins, increased muscular activity and osmolality of the lumen, all play a role in the absorption of drugs in the colon but the effects are probably less consequential (Mrsny, 1992:23).

1.7 SYSTEMS FOR DRUG DELIVERY TO THE COLON

The most direct route for delivery of drugs into the colon is by rectal administration. Since there is drawbacks such as patient acceptability and accessing the proximal colon using rectally administered dosage forms, orally administered colon-specific delivery systems have been developed. These include bacterial dependant delivery systems, enteric-coated systems (pH dependant systems) and time dependant formulations.

1.7.1 Bacterial dependant delivery systems

A microbial cleavage strategy utilizing the high enzymatic activity of microflora in the large intestine may be one of the most promising approaches in terms of site-specificity (Ishibashi *et al.*, 1998:32). Both prodrugs and dosage forms from which the release of drug is triggered by the action of colonic enzymes have been devised. The upper part of the GIT has a microflora count of less than $10^3 - 10^4$ cfu/ml. These are mainly gram-positive aerobic bacteria. The microflora count in the colon is in the region of $10^{11} - 10^{12}$ cfu/ml, consisting of mainly anaerobic bacteria. This huge amount of microflora ferments various types of substrates that have been left undigested in the small intestine, e.g., di-, tri- and polysaccharides. For this fermentation, the bacteria produce a large number of enzymes such as

azoreductases and polysaccharidases which include galactosidase, glycosidase, pectinase and dextranase (Zhang *et al.*, 2002:198).

1.7.1.1 Prodrugs

The realization that the enzymes of micro-organisms in the human colon may hydrolyze prodrugs and other molecules to active therapeutics has led to increased research activity in the area of microbially controlled drug delivery to the colon (Rubinstein & Sintov, 1992:235).

A successful prodrug-based delivery system is one in which the promoiety (the inactive portion of the prodrug) minimizes absorption of the drug until the active part is released (usually by enzymatic action) near the target site (Friend, 2005:253). Thus, the promoiety is used to increase the hydrophylicity of the parent drug, increase molecular size, or both, to minimize absorption of the drug prior to reaching the target site.

a) Azo-prodrugs

The colon is known to be a reductive medium in which azo groups can be cleaved with formation of the corresponding amines. This ability of microflora to reduce azo groups has been known for many years and was used in the food dye industry. This opportunity for reductive degradation of azo compounds by microflora has been exploited to prepare prodrugs of the anti-inflammatory agent 5-aminosalicylic acid (5-ASA). These prodrugs combine two drugs molecules linked by an azo-bond (-N=N-). An example is sulphasalazine, which is used in the treatment of IBS. A sulphonamide antibiotic, sulphapyridine, and 5-ASA, is combined and linked with an azo-bond. The prodrug is reduced to its compounds in the colon by a specific enzyme called azoreductase. New generation azo-prodrugs using 5-ASA have also been developed for example olsalazine (figure 1.3) which upon reduction of the azo-bond, generates two molecules of 5-ASA *(Rubinstein, 2005:34)*.

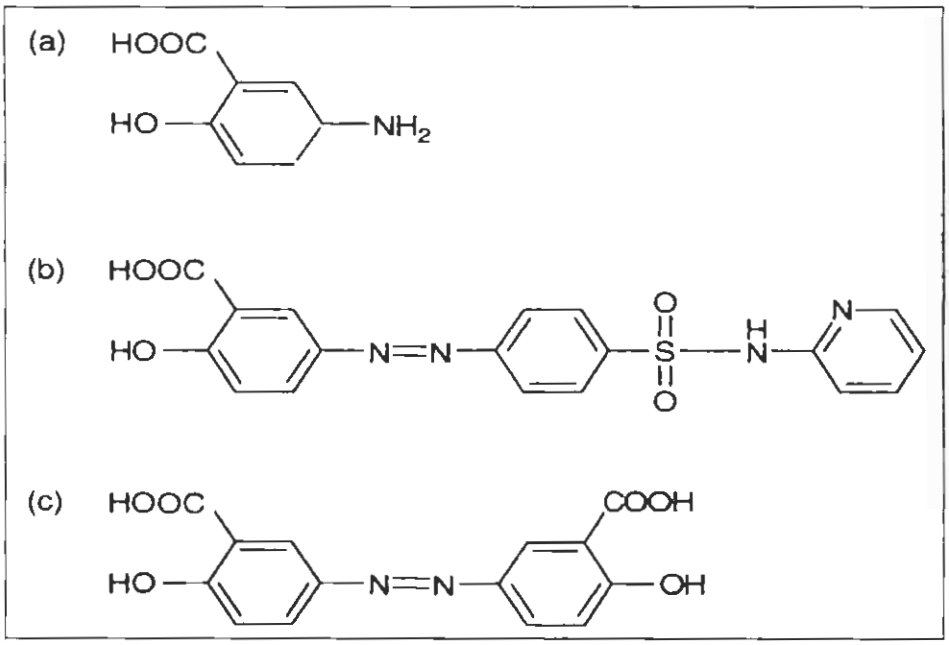

Figure 1.3: *Five amino salicylic acid (5-ASA) (a), sulphasalazine (b) and olsalazine (c) (Rubinstein, 2005:34).*

Polymers containing azo groups in the backbone, such as polyamides, have also been prepared for use as site selective degradable coatings (Schacht *et al.*,1996:327). The use of azo-polymers has been hindered as some azo-aromatic compounds are known to be potential carcinogens (Ibekwe *et al.*, 2004:28).

b) Glycosidic prodrugs

Corticosteroid prodrugs have been developed by the attachment of the active corticosteroid to glycosidic carriers. The glycoside bonds will be cleaved in the colon by the action of glycosidase enzymes to release the active drug (Vandamme *et al.*, 2002:223). This principle was demonstrated by Friend and Chang (1985:51) and Tozer *et al.* (1991:445) who linked galactose, glucose or cellobiose, known to serve as substrates for colonic bacteria, to selected steroid drugs commonly used in the treatment of inflammatory bowel disease.

1.7.1.2 Dosage forms using degradable polymers or polysaccharides

Degradable polymers can be used for colon-specific drug delivery systems because of the presence of the biodegradable enzymes found only in the colon, (Kakoulides *et al.*, 1998:95). These polymers shield the drug from the environments of the stomach and small intestine allowing delivery of the drug to the colon. In the colon the polymers undergo degradation and breakdown leading to a reduction in their molecular weight and loss of mechanical strength. As a result, they are then unable to bind to the drug any longer and the drug is released in the colon.

Polymers have been used to form prodrugs with the drug moiety, as a coating material or to embed the drug in their matrices or hydrogels. Another approach is simply to use the polymer such as chitosan as a capsule dosage form (Tozaki *et al.*, 2002:51, Tozaki *et al.*, 1997:1016). Examples of these polymers are the azo-polymers and disulphide polymers as well as the polysaccharides such as pectin, amylose, guar gum and chitosan (Krishnaiah *et al.*, 2001:235; Kakoulides *et al.*, 1998:95; Kopeček, 1990:279).

An extensive range of drug delivery systems based on polysaccharides have been investigated. Because many of the polysaccharides are already used as excipients in drug formulations and are constituents of the human diet, they are generally regarded as safe. Another advantage of these materials are that they are relative easy obtainable and inexpensive.

The disadvantage is that they are mostly hydrophilic and gel forming. Methods have therefore been devised to ensure that the drug does not prematurely diffuse from the dosage form before reaching the colon. To overcome this problem the natural polysaccharides are either chemically modified or mixed with hydrophobic, water insoluble polymers. This has the effect of limiting the swelling in the upper GIT, but still permitting a partial solubilisation of the matrix or coating in the colon due to bacterial degradation resulting in drug release (Ibekwe, 2004:29).

a) Pectin

Pectin is a polysaccharide found in the cell walls of plants. It is not digested in the upper GIT tract but is totally degraded by colonic bacteria (Ashford *et al.*, 1994:225; Wakerley *et al.*, 1996:73). One disadvantage of pectin is its solubility. This can however be adjusted by changing the degree of methoxylation or by preparing salts such as calcium pectinate which is insoluble (Rubinstein & Sintov, 1992:242). Several studies have been done on the coating properties of pectin alone and in combination with other polysaccharides such as chitosan and hydroxypropyl methylcellulose (Macleod *et al.*, 1999:251; Fernández-Hervás & Fell, 1998:115). These studies concluded that these coatings are capable of retarding the release of tablet core materials until it reaches the colon where the enzymes will degrade the coating, allowing drug release to occur.

b) Amylose

Amylose is one of the major components of starch, accounting for 15-25% of its total weight. It has good film forming properties and is resistant to pancreatic enzymes in the small intestine but will undergo degradation due to fermentation by a broad range of bacterial enzymes (Ibekwe, 2004:29). Amylose, in combination with the water-insoluble polymer ethyl cellulose, has been exploited as film coating for colonic drug delivery (COLAL™).

c) Chitosan

A detailed discussion of chitosan and its use in colonic drug delivery is given in chapter 2.

1.7.2 Enteric-coated systems

The highest pH levels in the GIT are found in the terminal part of the ileum (pH=7.5). On entry into the colon, the pH drops to 6.4. The pH then increases along the colon, reaching 6.6 in the transverse colon and 7.0 in the descending colon. The fall in the

pH on entry into the colon is due to the presence of short chain fatty acids arising from the bacterial fermentation of polysaccharides (Watts & Illum, 1997:895).

The enteric coated systems is coated with a polymer that effectively resist drug release under acidic conditions of the stomach but will dissolve at higher pH levels such as in the small intestine. As a consequence a considerable amount of drug may be released in the small intestine before it reaches the colon. Careful selection of enteric coat and thickness is therefore necessary to ensure that disintegration does not occur until the dosage form moves through the ileocecal junction into the colon.

1.7.3 Time dependant formulations

Another approach to colon targeting uses time as the release mechanism. Although gastric emptying time is highly variable, the small intestine transit time is fairly constant at between 3 - 4 hours. Time-controlled release systems may be swellable, soluble coatings or a matrix type system. These systems can resist the release of the majority of drug from the formulation for an additional 3 hours (i.e. the usual small intestine transit time) after gastric emptying and can deliver the drug primarily to the colon. Various polysaccharides and polymers are used in tablet formulations to retard drug release. These have been used either as matrices or as coating material.

A limitation associated to the time dependant formulations is the variability in the gastric empting of the dosage form depending i.e. on the amount and type of food present in the stomach.

1.8 SUMMARY

The colon is vulnerable to a number of disorders including ulcerative colitis, Crohn's disease, irritable bowel syndrome and carcinoma. Recommended treatment include the administration of anti-inflammatory drugs, chemotherapy drugs and antibiotics (Vandamme *et al.*, 2002:219). Some of these drugs need to be released in the colon to ensure direct treatment at the disease site. In addition to local therapy, the colon can also be utilized as a portal for the entry of drugs into the systemic circulation. For

example, molecules that are degraded or are poorly absorbed in the upper part of the GIT, such as proteins and peptides, may be better absorbed in the more benign environment of the colon. Systemic absorption from the colon can also be useful as a means of achieving chronotherapy for diseases that are sensitive to circadian rhythms such as asthma, angina and arthritis. The colon offers distinct advantages on account of a near neutral pH, a much longer transit time, reduced enzymatic activity and a much greater responsiveness to absorption enhancers. These advantages as well as the fact that colon specific drug delivery increases the bioavailability and results in a reduction in drug dose and side effects, makes the colon ideally suitable for delivery of a variety of drugs (Basit & Bloor, 2003:185).

Modified release formulations are usually based on either a single unit (tablets or capsules) or multi-unit (minitablets, pellets or granules) dosage forms. Multi-unit dosage forms tend to exhibit more uniform gastrointestinal transit and absorption characteristics due to their small size and divided nature. The slower rate of passage of multi-units through the colon can also be an advantage for colonic drug delivery.

A variety of approaches have been used and systems been developed for the purpose of achieving colonic targeting. These approaches are either drug-specific (prodrugs) or formulation specific (coated or matrix preparations) and mechanisms that depends on the pH, transit time and pressure or bacteria in the colon have been used. At present, the bacterial activated delivery system approach possibly has the greatest potential for colonic targeting as the levels of bacterial enzyme activity in the colon is the characteristic of this part of the gastrointestinal tract that is the most unique and exploitable (Ibekwe *et al.*, 2004:30).

CHAPTER 2

THE USE OF CHITOSAN IN COLONIC DRUG DELIVERY

2.1 INTRODUCTION

In 1881 Henry Braconnot discovered chitosan when he performed experiments with fungi. The discovery of chitin is essentially based on some reactions carried out on raw materials isolated from *Agaricus volvaceus, A. acris, A. cantarellus, A. piperatus Hydrium repandum, H. hybridum* and *Boletus viscidus*. Fungal material was partially purified by boiling in dilute potassium hydroxide which removed the proteins and pigments. Other impurities were removed by reactions with sulfuric acid. Braconnot actually produced chitosan but was unable to detect and described the chemical transformation (Muzzarelli, 2002:4).

2.2 AVAILABILITY

Chitin is the second most abundant polysaccharide in nature, cellulose being the most abundant (Hejazi & Amiji, 2003:152). Chitin is found in the exoskeleton of crustaceans, insects and some fungi and several millions of tons are harvested annually. The shell wastes of shrimp, lobster, krill and crab are the main commercial sources of chitin. The principle sources of chitin are given in table 2.1 (Felt *et al.*, 1998:979).

Table 2.1: *Principle sources of chitin (Singla & Chawla, 2001:1048).*

Organism		Chitin content (%)
Crustaceans	Crab	72.10
	Shrimp	69.10
	Lobster	69.80
Insects	True fly	54.80
	Sulfur butterfly	64.00
Fungi	*Aspergillus niger*	42.00
	Mucor rouxii	44.50

The chitin content is given as the organic weight of the cuticle for the crustaceans and insects and as dry weight of the cell wall for the fungi.

2.3 CHEMISTRY

Chitin, (1-4)-linked 2-acetamido-2-deoxy-β-D-glucan, is a polymer consisting of n-acetylglucosamine units. *In vivo,* one out of ten units is deacetylated (Muzzarelli, 2002:5). The structure is given in figure 2.1.

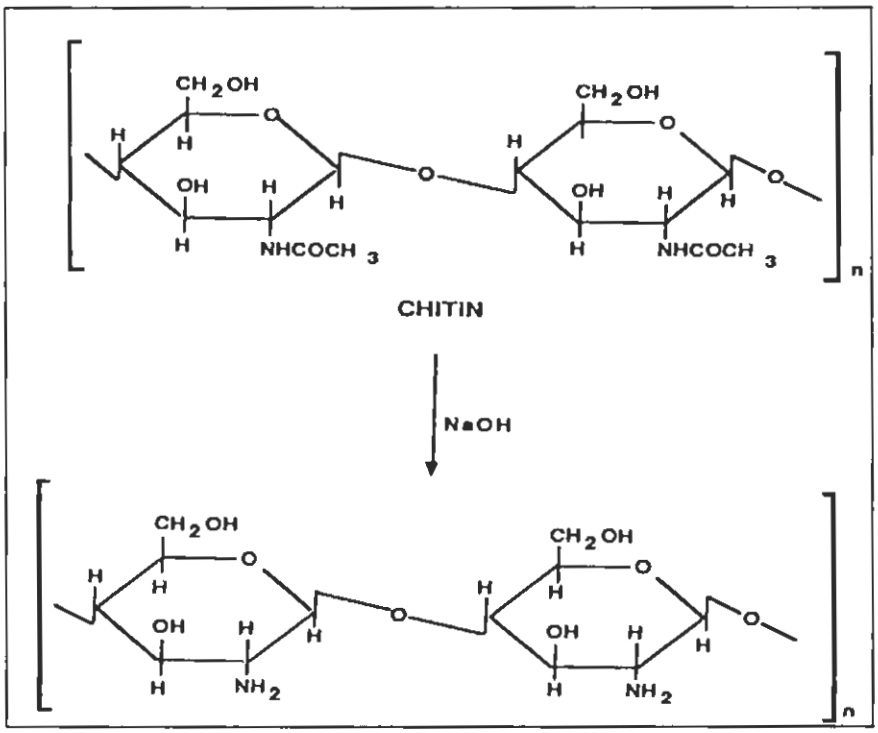

Figure 2.1: *The structure of chitin and chitosan.*

Partial deacetylation of chitin results in the production of chitosan which is a polymer consisting of copolymers of glucosamine and N-acetyl glucosamine.

Chitosan is commercially produced in different parts of the world like Japan, Poland, India, Russia and North America (Singla & Chawla, 2001:1048). The basic procedure for the manufacturing of chitosan is given in figure 2.2.

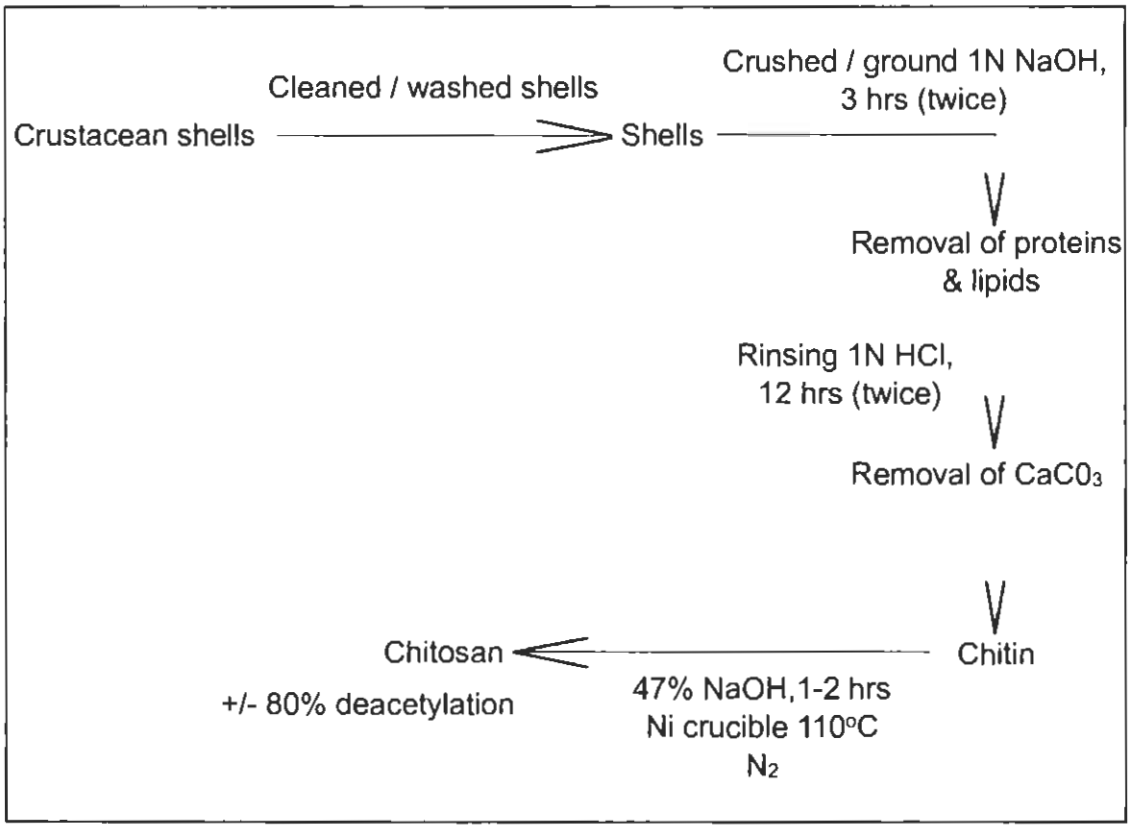

Figure 2.2: *The manufacturing of chitosan (Singla & Chawla, 2001:1048).*

2.4 PHYSICOCHEMICAL PROPERTIES

The term chitosan describes a series of chitosan polymers with different molecular weights, viscosities and degrees of deacetylation. Some authors use the term chitinosan for the spectrum of acetylated poly (N-glucosamines) ranging from chitin (0% deacetylated) to chitosan (100% deacetylated) (Rege *et al.*, 1999:49). The commercial range of chitosans available have a degree of deacetylation from 60 to 90 % and molecular weights in the range from 50 to 2000 kDa.

According to Rege *et al.* (1999:50), chitosan properties (solubility, ionizability, reactivity) and functionality (bioadhesiveness, transport enhancing properties, wound healing properties and complexing properties) depend on two fundamental parameters: degree of deacetylation and degree of polymerization (i.e. molecular weight).

2.4.1 Solubility

Chitosan is a weak base with a pKa value of the D-glucosamine residue of about 6.2 – 7.0 and is therefore insoluble at neutral and alkaline pH values (Hejazi & Amiji, 2003:153).

Chitosan forms salts with organic and inorganic acids although its solubility in inorganic acids are limited when compared with its solubility in common organic acids (Singla & Chawla, 2001:1049). In an acidic medium, the amine group of the polymer is protonated, resulting in a soluble, positively charged polysaccharide with a high charge density (Hejazi & Amiji, 2003:153). These salts are soluble in water and the solubility is dependant on the degree of deacetylation and the pH of the solution. Chitosan with a low degree of deacetylation ($\leq$40%) are soluble up to a pH of 9, while highly deacetylated chitosans ($\geq$85%) are soluble only up to a pH of 6.5. The solubility is also greatly influenced by the presence of salt in the solution.

Aiedeh and Taha (1999:104) changed the optimum pH range for the gel forming capacity of chitosan from acidic to alkaline values by covalently linking the chitosan backbone to carboxylic acid. They expected the carboxylic acid residues to interact with the neutral or slightly alkaline pH of the terminal ileum and ileocecal junction, thus yielding a suitable matrix for colon-specific drug delivery.

2.4.2 Viscosity

Increasing the degree of acetylation increases the viscosity of chitosan in solution because of the different conformations of high and low deacetylated chitosans in

aqueous solution. The viscosity is also dependant on the concentration of the chitosan as well as the temperature of the solution (Singla & Chawla, 2001:1049).

2.4.3 Toxicity

Chitosan has low oral toxicity with an LD50 in rats of 16g/kg. The toxicity might depend on the degree of deacetylation, molecular weight, purity and route of administration (Hejazi & Amiji, 2003:154).

2.4.4 Muco-adhesiveness

Chitosan has been shown to possess muco-adhesive properties due to molecular attractive forces formed between positively charged chitosan and negatively charged mucosal surfaces. These properties may be attributed to strong hydrogen bonding groups (-OH and –COOH), strong charges, high molecular weight and surface energy properties (Lehr *et al.*, 1992:45).

2.5 THE USE OF CHITOSAN IN THE PHARMACEUTICAL INDUSTRY

Properties such as biodegradability, low toxicity and good biocompatibility make chitosan suitable for use in biomedical and pharmaceutical formulations (Sinha *et al.*, 2004:2). Among the pharmaceutical applications it has been used as a vehicle for directly compressed tablets, as a binder, disintegrant, granulating agent as well as a carrier for sustained release preparations. Further applications include the use as a co-grinding diluent for the enhancement of dissolution rate and bioavailability of water-insoluble drugs and as penetration enhancer for peptide drugs (Sinha *et al.*, 2004:2). Nonetheless, chitosan have not been widely adopted as pharmaceutical excipient or formulation component. One area of concern involves the utilization in directly compressible formulations as virtually all the formulations necessitate the addition of other ingredients to facilitate compression. This reflects the fact that the commercially supplied chitosan lack good flow properties and compressibility (Rege *et al.*, 1999:50).

Other than the oral route, chitosan is also extensively used in routes such as the parenteral, ocular, nasal, buccal and transdermal routes (Weyenburg *et al.*, 2003; Dodane & Vilivalam, 1998; Felt *et al.*, 1998).

2.6 CHITOSAN IN COLONIC TARGETING

2.6.1 Introduction

Colon-specific drug delivery by the oral route avoids the inactivation of peptide drugs in the GIT, decreases the side effects due to gastric irritation after anti-inflammatory administration but allows local treatment of bowel diseases (Orienti *et al.*, 2002:51).

One of the most realistic possibilities of obtaining colon-specific release, involves the participation of the microflora in the large intestine. Different natural and synthetic polymers have been evaluated for their susceptibility to degradation by these bacterial enzymes. Promising polymers are natural polysaccharides whose glycosidic bonds are hydrolyzed in the colon. These polymers include chitosan, pectin, guar-gum, dextrans, amylose and chondroitin sulphate. Chitosan, in particular, is considered a good candidate as it is widely available at low cost and has favorable biological properties such as a low toxicity, biocompatibility and biodegradability.

2.6.2 Solubility

Chitosan is soluble in dilute acid and precipitates at a pH above 7 (Sinha & Kumria, 2001b:22) while it is insoluble at the alkaline pH of the colon. This insolubility may be a drawback in its use in colon-specific drug release. A possible solution may be the salification of chitosan to modify its solubility and other physical-chemical properties (Orienti *et al.*, 2002:52).

Because of the solubility of chitosan at low pH ranges, its use in colon-specific delivery requires an enteric layer over the chitosan which could protect it against the acidity of the stomach. As the formulation reaches the intestine, the pH increases and the enteric coating dissolves releasing the chitosan core.

Chitosan microspheres were coated with Eudragit L-100® and Eudragit S-100® to study the release of sodium diclofenac (Lorenzo-Lamosa *et al.*, 1998:111). The Eudragit® coating gave a pH-dependant release profile and a change in the molecular weight of the chitosan or the use of different chitosan salts could control the drug release.

Aiedeh and Taha (1999:103) prepared semi-synthetic derivatives of chitosan i.e. chitosan succinate and chitosan phthalate. Their study on the *in vitro* release of sodium diclofenac from matrices of these salts, showed that the formulations resisted dissolution under acidic conditions and improved dissolution under basic conditions, suggesting the suitability of such formulations for colon-specific drug delivery.

2.6.3 Microbial degradation

Chitosan is acted upon by enzymes produced by the microflora of the colon, specifically the azoreductases and the polysaccharidases, making it a candidate for colon-specific drug delivery systems. Recent research suggests that degradation by colonic bacterial enzymes might be one of the important properties of chitosan for the successful use in colonic targeting (Zhang & Neau, 2002:2761).

It was found that the susceptibility of chitosan to degradation by bacterial enzymes is dependant on both its molecular weight and degree of deacetylation. Chitosan with a lower molecular weight and a lower degree of deacetylation would be more susceptible to enzymatic degradation and would be a suitable candidate for the use in colon drug delivery.

2.6.4 Dosage forms

Different approaches to the design of colon targeted delivery systems containing chitosan have been used such as films and chitosan capsules.

2.6.4.1 Films / coating

Fernández-Hervás and Fell (1998:115) used pectin and chitosan mixtures as coatings for colon-specific drug delivery of indomethacin and paracetamol which were used as model drugs to represent poorly and highly soluble drugs, respectively. Pectin alone, when used at thick coats, was able to protect the cores from premature release. Mixtures of chitosan/pectin achieved better protection at a lower coat weight. The use of pectinolytic enzymes to simulate breakdown in the colon showed that the coating was susceptible to enzymatic breakdown and allowed drug release to occur.

2.6.4.2 Capsules

Chitosan capsules were coated with hydroxypropyl-methylcellulose (HPMC) and evaluated for colonic delivery of insulin (Tozaki *et al.*, 1997:1016). *In-vitro* studies showed that there is little release of the drug in simulated gastric juice or artificial intestinal juice. The presence of rat cecal contents increased the release of the drug, suggesting that the microflora present in the cecal content may have produced enzymes for the degradation of chitosan.

Chitosan capsules were also used for delivering 5- aminosalicylic acid (5-ASA) and a new thromboxane synthetase inhibitor, R-68070, successfully to the colon (Tozaki *et al.*, 2002:51; 1999:1155).

2.6.4.3 Beads

Hydrogel beads can be formed by polyelectrolyte complexation of chitosan with its counter ion, tripolyphosphate (TPP). A multiparticulate system consisting of such formed beads has been investigated by Zhang *et al.* (2002:197) for the colon-specific drug delivery of macromolecules using fluorescein isothiocyanate-labeled bovine serum albumin as the model compound. The study confirmed that the beads were degraded by colonic bacterial enzymes. The results also indicated that the colonic enzymes used, still attacked chitosan even after it has been cross-linked and its solubility reduced, resulting in higher protein release.

2.6.4.4 Granules / matrices

Nykänen *et al.* (1999:251) developed site-specific systems consisting of matrix granules of chitosan with enteric polymers for release of drugs in the lower part of the small intestine or in the colon. Colonic delivery could be achieved and the inclusion of organic acids retarded the *in vitro* release of the drugs.

2.7 SUMMARY

After cellulose, chitin is the second most abundant polysaccharide in nature (Hejazi & Amiji, 2003:152). Partial deacetylation of chitin results in the production of chitosan which is a polymer consisting of copolymers of glucosamine and N-acetyl glucosamine. Properties such as biodegradability, low toxicity and good biocompatibility make chitosan suitable for use in biomedical and pharmaceutical formulations (Sinha *et al.*, 2004:2). Among the pharmaceutical applications it has been used as a vehicle for directly compressed tablets, as a binder, disintegrant, granulating agent as well as a carrier for sustained release preparations. Other applications include the use as a co-grinding diluent for the enhancement of dissolution rate and bioavailability of water-insoluble drugs and as penetration enhancer for peptide drugs (Sinha *et al.*, 2004:2).

Colonic drug delivery is important as the colon is vulnerable to a number of disorders including ulcerative colitis, Crohn's disease, irritable bowel syndrome and carcinoma. The development of delivery systems to the colon can assist in the local treatment of these diseases as well as systemic delivery of drugs as the conditions in the colon is less harsh than in the upper part of the GIT.

One of the most realistic possibilities of obtaining colon-specific drug release, uses the ecosystem of the microflora in the large intestine. Different natural and synthetic polymers have been evaluated for their susceptibility to degradation by these bacterial enzymes. Promising polymers are natural polysaccharides whose glycosidic bonds are hydrolyzed in the colon such as chitosan, pectin, guar-gum, dextrans,

amylose and chondroitin sulphate. Chitosan, in particular, is considered a good candidate as it is widely available at low cost and has favorable biological properties.

In the study, chitosan minitablets was formulated that could be delivered in or near the colon where it would be degraded by bacteria and enzymes in the colon to release the contained drug.

In order to investigate the influence of formulation factors on the properties of the minitablets, it was necessary to ensure that the chitosan powder has satisfactory powder flow characteristics to ensure that compression of the tablets was possible and there were not unacceptable variation in the tablets (such as weight, thickness, disintegration and strength).

CHAPTER 3
FLOW CHARACTERISTICS OF CHITOSAN

3.1 INTRODUCTION

Good flow properties of powders are critical to the successful development of any pharmaceutical tablet or capsule formulation. It is essential that an accurate assessment of flow properties be made as early in the development process as possible so that an optimum formulation can be identified (Taylor *et al.*, 2000).

The initial characterization of the flow properties of solids was conducted by Jenike (1954) and Carr (1965). Jenike investigated adhesive/cohesive forces of particles as they relate to flow properties by examining their normal and shear stresses on powder beds. Carr evaluated interparticulate cohesive properties with angle of repose measurements and the effects of packing geometry with bulk and tap density measurements.

Augsberger and Shangraw (1966:418) addressed the need to determine powder flow for pharmaceutical formulations. They identified the need for indexes determining flowability after recognizing the importance of a uniform tablet weight. With that objective, they proposed the coefficient of variation of the average tablet weight as the flowability index.

3.2 PROPERTIES THAT INFLUENCE POWDER FLOW

3.2.1 Adhesion and cohesion

Adhesion and cohesion are molecular forces that produce a tendency for solid particles to stick to themselves (cohesion) and other surfaces (adhesion). Cohesion forces are composed mainly from van der Waal's forces which increase as particle size decreases and vary with changes in relative humidity. Other forces may be produced by surface tensional forces between absorbed liquid layers at the particle surfaces and electrostatic forces (Staniforth, 2000:601). These adhesion/cohesion

forces (and therefore the powder flow) can be characterized by different methods that investigate shear strength (using a shear cell such as a Janike shear cell), tensile strength and angle of repose.

3.2.2 Particle properties

3.2.2.1 Particle size

Since adhesion/cohesion occurs at the surfaces of the particles, particle size will influence the flow of powders. In general, fine particles with a high surface to mass ratio are more cohesive than coarser particles (Staniforth, 2000:604).

3.2.2.2 Particle shape

Particles with similar particle size but different shapes can have very different flow properties. For instance, particles that are spherical and has minimum interparticle contact, will generally have better flow properties than particles that are flakes or dendritic (Staniforth, 2000:604).

3.2.2.3 Packing density

Powder flow under the influence of gravity and therefore dense particles are generally less cohesive than less dense particles (Staniforth, 2000:604).

3.2.3 Packing geometry

A set of particles can be filled into a volume of space to produce a stable powder bed. Under influence of vibration, particles can be mobilized so that if the vibration is stopped, the bed will be stable again but will occupy a different volume than before. In general, such rearrangement result in a transition from loosely to more tightly packed particles. The more tightly packed particles will require a higher driving force to produce powder flow and will be more cohesive because of an increase in the number of interparticle contacts (Staniforth, 2000:604).

3.3 CHARACTERIZATION OF POWDER FLOW

When examining the flow properties of a powder, it is useful to be able to quantify the type of behavior. Many different methods have been described using direct methods (dynamic or kinetic) or indirect methods generally by measurements carried out on static beds.

3.3.1 Indirect methods

3.3.1.1 Angle of repose

Angles of repose have been used because of their relationship with interparticle cohesion. There are many different methods determining the angle of repose and they may produce different values (Staniforth, 2000:610). Angle of repose measurements are relatively simple and are used extensively (Kaerger *et al.*, 2004:173, Räsänen *et al.*, 2003:418, Lavoie *et al.*, 2002:887, Taylor *et al.*, 2000).

The disadvantages of angle of repose are that the different methods that are used as well as the handling of the samples prior to measurement may produce different values. For these reasons, angle of repose tend to be variable and are not always representative of flow under specific conditions (Staniforth, 2000:610).

The angle of repose (α) is determined from the height of the powder cone (h) and the diameter at the base (d) by the equation:

$$\tan \alpha = 2h/d$$

Powders with angles of repose higher than 50° have unsatisfactory flow properties while powders with angles of close to 25° have very good flow properties (Staniforth, 2000:610).

3.3.1.2 Shear cell determinations

Powder flow are commonly investigated under gravity loading conditions (angle of repose, tapped density, flow rate etc.). Although these methods are useful and have demonstrated the dependence of powder flow on various factors such as moisture content, they have been proved difficult to relate to features at particulate level (Guerin *et al.*, 1999:92). Thus, a more fundamental and physical measurement, such as the behavior of powders in a shear cell, can be used. These cells are designed to condition powders under a known load and to measure forces needed to shear powder beds.

This methodology is time and product consuming and correct and reproducible preparation of samples are difficult to achieve and results can be very operator and know-how dependant (Guerin *et al.*, 1999:92).

3.3.1.3 Bulk density measurements

The bulk density of a powder is dependant on the particle packing and changes as the powder consolidates. As explained above, a more consolidated powder is more resistant to powder flow and therefore the way with which a powder consolidates can be used as an indirect method of quantifying powder flow (Staniforth, 2000:612).

A powder (weight known) is placed in a measuring cylinder and the volume recorded before it is tapped by a mechanical device to consolidate the powder.

The initial bulk density (D_0) and the final bulk density (D_f) can be calculated. Hausner found that the ratio D_f/D_0 was related to interparticle friction and could be used to predict powder flow. Powders with low interparticle friction such as coarse spheres have ratios of approximately 1.2 while powders which are less free-flowing such as flakes have ratios of greater than 1.6.

Another method using these densities was developed by Carr:

$$\% \text{ compressibility} = (D_f - D_0 / D_f) \times 100$$

The relationship between powder flowability and % compressibility is given in table 3.1.

Table 3.1: *Relationship between powder flowability and % compressibility (Staniforth, 2000:613).*

% compressibility	Flow description
5 to 12	Excellent
12 to 18	Good
18 to 23	Fair
23 to 28	Poor (very fluid powders)
28 to 35	Poor (very cohesive powders)
35 to 40	Very poor
more than 40	Extremely poor

3.3.1.4 Critical orifice diameter

This measurement device uses a cylinder with a series of interchangeable base plate discs that have different diameter orifices which can be blocked by the presence of a simple shutter. The critical orifice diameter is the size of the smallest hole through which powder discharges when the shutter is removed.

In some cases, repetition of the experiment produces different critical orifice diameters, and minimum and maximum values are sometimes quoted (Staniforth, 2000:613). Several other devices that measures the critical orifice diameter is used and the values quoted for the diameter can differ accordingly. In this study an measurement apparatus was developed that minimizes the important problems that often occurs in some of the above mentioned devices such as so called dead corners and the angle of the powder bed (see 3.5.1.3).

3.3.2 Direct methods

3.3.2.1 Powder flow rate

The simplest method of determining the flow rate directly is to measure the rate at which powder discharges from a funnel/hopper. A simple shutter is placed over the outlet and the container filled with powder. The shutter is then removed and the time for the powder to discharge completely is measured. The flow rate can be calculated by dividing the powder mass discharged by the time. The potential problem with this method is that if the outlet opening is too large, the flow rate will not be able to discriminate between free-flowing powders and powders having poor flow characteristics.

Another method that is essentially similar to the above mentioned method is also used. The only difference is that the powder is discharged onto a balance (or the discharged powder weighted) and the mass flow rate can be determined.

3.3.2.2 Avalanche behavior

All the above mentioned methods give results that predict the flow behavior of the powder under working conditions. However, the prediction depends on the relationship between each partial answer given by the different analyses conducted on the powder, as the individual contributions of the powder's characteristics to movement are less clear. To circumvent this problem, the powders can be studied under dynamic conditions such as the study of avalanches (Lavoie *et al.*, 2002:887).

In this method the powder is rotated in a cylindrical drum which allows the powder to flow unconstrained. The frequency of avalanches can be used to characterize the powder flow. Lavoie *et al.* (2002:892) formulated a flowability index as well as a cohesion index, using the average avalanche time, which correlated well with Carr's index.

These avalanche behavior tests require significant amounts of powder and can be laborious.

3.3.2.3 Vibratory feeder

In this method, the powder is delivered through a funnel onto a vibrating feeder which in turn delivers it onto a pan on a balance. The mass of powder collected in the pan at regular intervals is used to calculate the powder flow index (Bhattachar, 2004:387).

3.4 COMPOSITE INDEX

Taylor *et al.* (2000) proposed a method to calculate a weighted composite index on completion of the individual flow tests. The method will designate a single score for each material tested as opposed to several test scores.

The method is based on the concept that each of the three tests conducted, add equally to the index. As the relative contribution of each individual test to the composite score was unknown, an arbitrary value of one third was assigned to each of the three tests and the performance of the model was tested against materials with so-called known flow properties. The composite index was therefore devised to yield a score of 100 for an optimal result for each of the tests as each test was transformed to a value between 0 and 33.33%.

The raw data were transformed using the mathematical equation of a straight line ($y = mx + c$). For the critical orifice data, the critical orifices of 34 mm and 4 mm were used as representing 0% and 33.33% respectively. (For the compression test the values were taken as 5% and 55% while the values for the angle of repose test were 25° and 75°.) When these values are plotted against each other, a straight line with a m-value of -1.111 and a c-value of 37.778 is produced (For the compression test the values were -0.666 and 36.666 and for the angle of repose test the values were -0.666 and 50). The raw test data can be mathematically transformed from this values to yield the score used to calculate the composite index.

3.5 EXPERIMENTAL DESIGN

In order to prepare minitablets with a tablet press, it is necessary for the powder formulation to have satisfactory powder flow characteristics. It is known that chitosan has an extremely poor flowability and this property restricts its use as an excipient in tablets. The problem is aggravated when small dies (in the case of minitablets the die opening is in the region of 3 - 4 mm) are used. It was therefore necessary in the study to do a thorough investigation into the flow properties of chitosan that would be used in the preparation of the minitablets.

3.5.1 Methods

3.5.1.1 Tap density

A volume of the powders were measured in a measuring cylinder and subsequently weighed. The measuring cylinder was placed on a vibrating apparatus and the powders were vibrated until the volume remained constant. The experiment was done in triplicate and Carr's index and the Hausner ratio was calculated for each powder.

3.5.1.2 Angle of repose

The powders were slowly poured through a funnel onto a surface on which the powder was free flowing. The funnel was kept in place 5 cm above the surface. The resulting powder cone was photographed and the maximum height and diameter was measured and the angle of repose was calculated for each powder.

3.5.1.3 Critical orifice diameter (COD)

3.5.1.3.1 Introduction

The measurement of the COD is done with an apparatus (figure 3.1) consisting of a cylinder with a series of interchangeable base plate discs that have different diameter

orifices which can be blocked with a shutter. Alternatively, the base plate is simply moved to such a position that a certain orifice is directly underneath the cylinder. The cylinder is filled with the powder of which the COD must be measured, a suitable orifice is selected and the shutter opened to allow the powder to flow through the orifice. If the powder flows unrestricted through the selected orifice, a smaller orifice is selected and the measurement repeated. The opposite is obviously true if the powder does not flow through the orifice. The COD is the size of the smallest orifice through which the powder discharges unaided three times in succession.

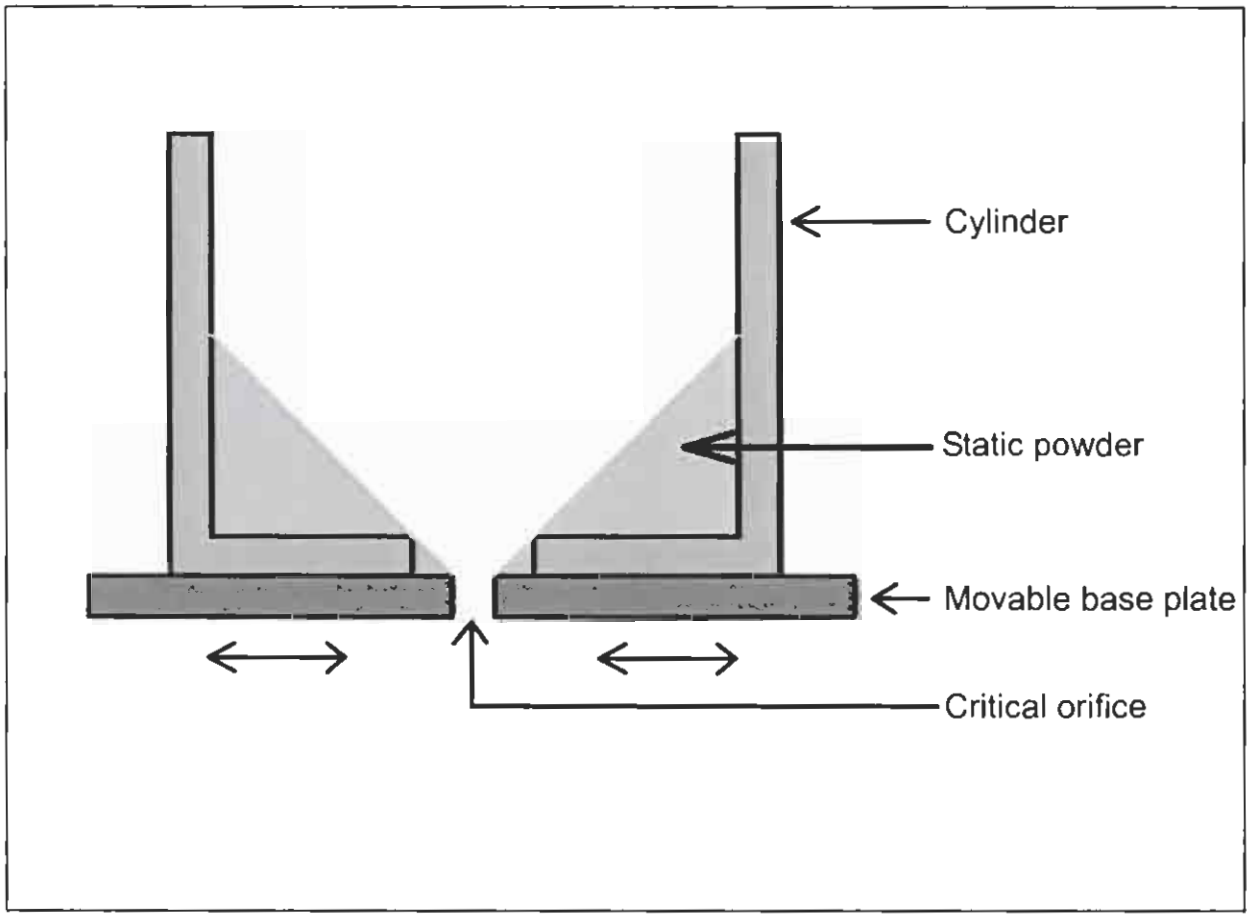

Figure 3.1: *Apparatus for the determination of the critical orifice diameter.*

3.5.1.3.2 Problems encountered with COD measuring

When the shutter is opened or the base plate moved to the "open" position, powder flows through the orifice until the cylinder is empty. Two possible problems may occur in such a measurement. Firstly the formation of static powder regions in the cylinder

and secondly the formation of holes in the powder bed through which the powder will flow.

Static powder

Regions of static powder are formed in the corners between the cylinder walls and the cylinder floor as well as between the floor and the shutter (figure 3.1). As the powder flows through the orifice, there is friction between the static particles and the moving particles. This friction will restrict the flow of the powder and eventually the flow of the powder will stop.

Holes in the powder bed

If the flow characteristic of the powder is poor, a hole can appear through the powder bed through which the powder will then flow. Static powder will help to create the problem which can be overcome to a great extent by changing the cylinder into a funnel shape to create an angle at which the powder can flow.

3.5.1.3.3 Development of an alternative COD apparatus

An apparatus consisting of a set of brass discs between 5 and 10 mm thick that can be stacked on top of each other to form a funnel was developed (figure 3.2 and figure 3.3). The angle between the opening and the orifice of each disc was machined to a set angle. There is also no static powder because of the absence of a cylinder floor and base plate. A cylinder could be fitted at the top of the funnel to create a holding chamber for the measured powder. Brass discs were chosen to minimize the effect of static electricity between the powder and the cylinder.

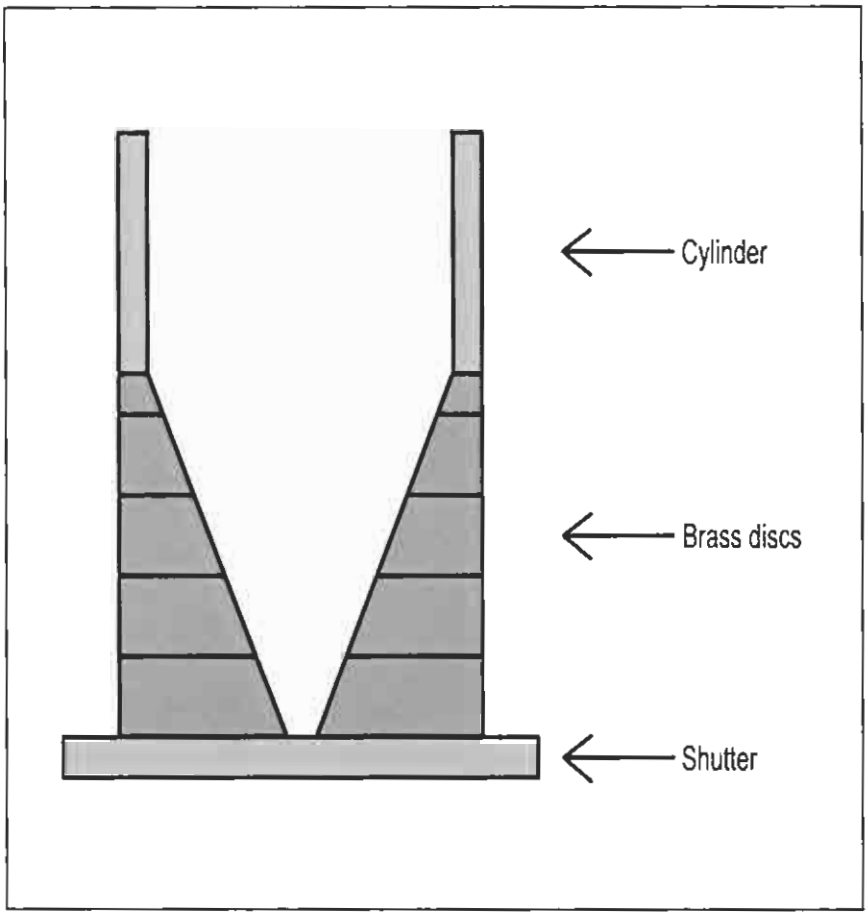

Figure 3.2: *Diagram of the new COD apparatus.*

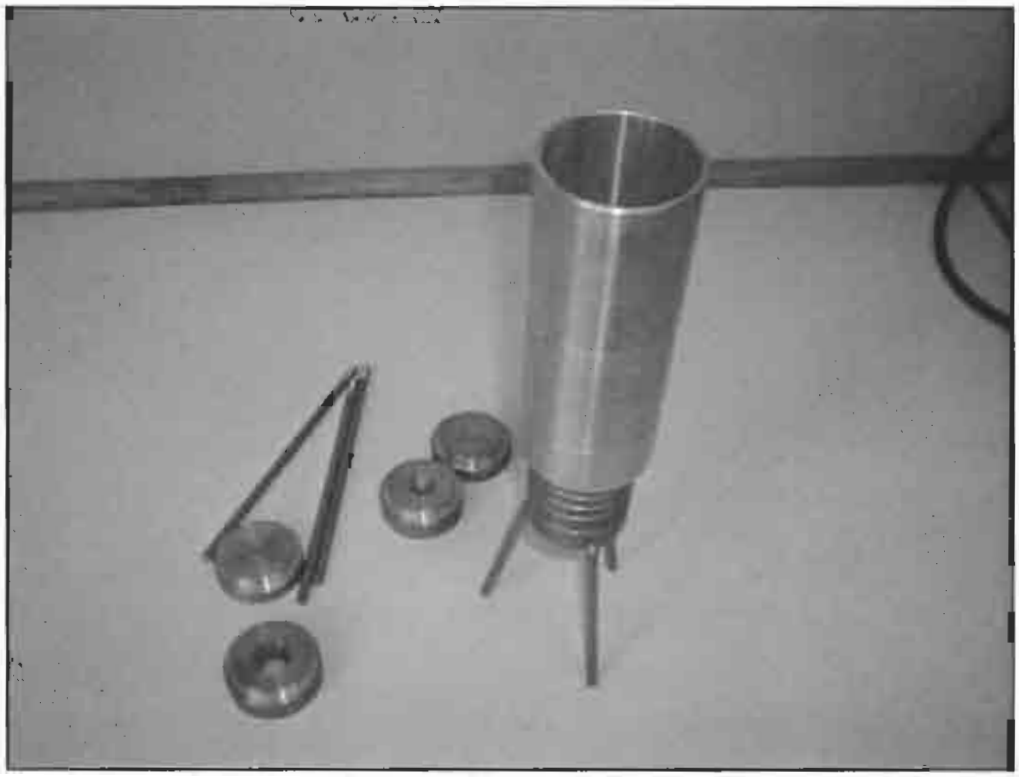

Figure 3.3: *The new COD apparatus showing the different components.*

3.5.1.3.4 Evaluation

To ensure that the amount of powder in the holding chamber does not influence the flow characteristics, an evaluation test was done.

Different amounts of powder was measured and placed into the holding chamber. The powder was allowed to flow through the orifice and the time to empty the chamber was noted. The experiment was repeated for several orifice diameters and different excipients. The mass of powder that flow through each orifice per second was calculated. The results showed that the amount of powder per time unit was constant for a specific excipient and orifice. It can therefore be concluded that the amount of powder in the holding chamber has no effect on the flow rate of the powders.

An application for a South African patent for this COD apparatus was registered in 2006 and was filed on 1 June 2006 in the patent office under the S.A. patent number 2006/04483. The application is attached in annexure B.

3.5.1.4 Composite index

The composite index was calculated according the method proposed by Taylor *et al.* (2000). The values of the critical orifice test used in the model was changed from between 4 mm and 34 mm (representing 33.333% and 0% respectively) to between 1.0 mm and 30 mm to accommodate the difference in test equipment and therefore the test values in the determination of the critical orifice test. The values used in the angle of repose test were between 30° and 50° while the values used in the tap density test were between 19% and 55%.

3.6 THE POWDER FLOW OF CHITOSAN IN COMPARISON TO THAT OF OTHER PHARMACEUTICAL EXCIPIENTS

3.6.1 Introduction

To evaluate the flowability of chitosan it was necessary to compare it with that of other frequently used pharmaceutical excipients using the methods as discussed above and apparatus that were available. The flowability of the excipients that were chosen are generally known and range from very poor (Emcocell 50M® and Prosolv 50®) to excellent (Emcompress® and Ludipress®).

3.6.2 Methods

The methods used to measure the tap density, the angle of repose and the critical orifice diameter were discussed in section 3.5.1. Carr's index, the Hausner ratio and the composite index were calculated. The materials used are given in annexure D.

3.6.3 Results

The results of the measurements are given in table 3.2. The measurement data are given in table A.1 to A.4 in annexure A.

Table 3.2: *Flowability of some pharmaceutical excipients.*

Excipient	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice (mm)	Composite index
Emcompress®	15.998	1.2	36.84	1.5	87.1
Emcocell 50M®	28.327	1.4	41.65	24	41.4
Emcocell 90M®	25.006	1.3	37.50	11	66.4
Chitosan	39.233	1.5	45.84	12	37.9
Avicel pH200®	30.338	1.4	38.35	18	51.9
Ludipress®	17.329	1.2	34.06	2	89.9
Prosolv MCC90®	25.339	1.3	37.98	9	67.7
Prosolv MCC50®	32.448	1.4	39.22	16	50.8

According to Carr's index, Emcompress® (15.998) and Ludipress® (17.329) have shown fair flowability while chitosan (39.223) has shown extremely poor flowability. The Hausner ratio confirms these results as chitosan has a ratio of 1.5 while Emcompress® and Ludipress® have ratios of 1.2.

Chitosan also has the worst angle of repose (45.84°) while that of the other excipients varies between 34.06° and 41.65°.

The critical orifice test also confirms the good flowability of Emcompress® (1.5 mm) and Ludipress® (2 mm) while chitosan could only flow through the orifice of 12 mm.

The composite index that takes all this data into consideration therefore shows that the flowability of chitosan (37.9) is very poor if compared with other pharmaceutical excipients. Excipients with known good flowability such as Emcompress® and Ludipress® have indexes of 87.1 and 89.9 respectively while even excipients that have poor flowability such as Emcocell 50M® (41.4) and Prosolv SMCC50® (50.8) have better indexes than chitosan.

It is therefore clear that the chitosan used in this study is not free flowing and problems with the filling of the small die in the tablet press can be expected. The addition of glidants as well as using a bigger particle size after sieving the chitosan might help alleviate this problem.

3.7 THE EFFECT OF RELATIVE HUMIDITY ON THE MOISTURE CONTENT OF CHITOSAN

3.7.1 Introduction

Chitosan is known to be hydrophilic and is muco-adhesive when wetted. This characteristic as well as the fact that the moisture content of powders normally influences the flowability of powders, could therefore play a role in the flowability of chitosan. Before the effect of moisture on the flowability was investigated, an investigation into the loss on drying as well as increase in moisture content over time

was done. These tests would establish the quantity of free water in the chitosan stored under laboratory conditions, the time needed to completely dry the chitosan as well as the moisture uptake over time under different moisture and temperature storage conditions.

3.7.2 Methods

3.7.2.1 Loss on drying

Ten grams of chitosan were weighed and dried under vacuum at 40 °C. It was weighed hourly until the weight remained constant.

3.7.2.2 Moisture increase

Chitosan was dried under vacuum at 40 °C for 12 hours. Ten grams of the dried chitosan were weighed and placed in a climate controlled room that was set at a temperature of 25 °C and a relative humidity (RH) of 60% or at 40 °C and 75% RH. The powder was weighed at regular intervals until the weight remained constant.

3.7.2.3 Powder flow

The methods used were discussed in section 3.5.1.

3.7.3 Results

3.7.3.1 Loss on drying

The result of the percentage weight remaining as a function of the drying time is given in figure 3.4 and the measurements values are given in table A.5 in annexure A.

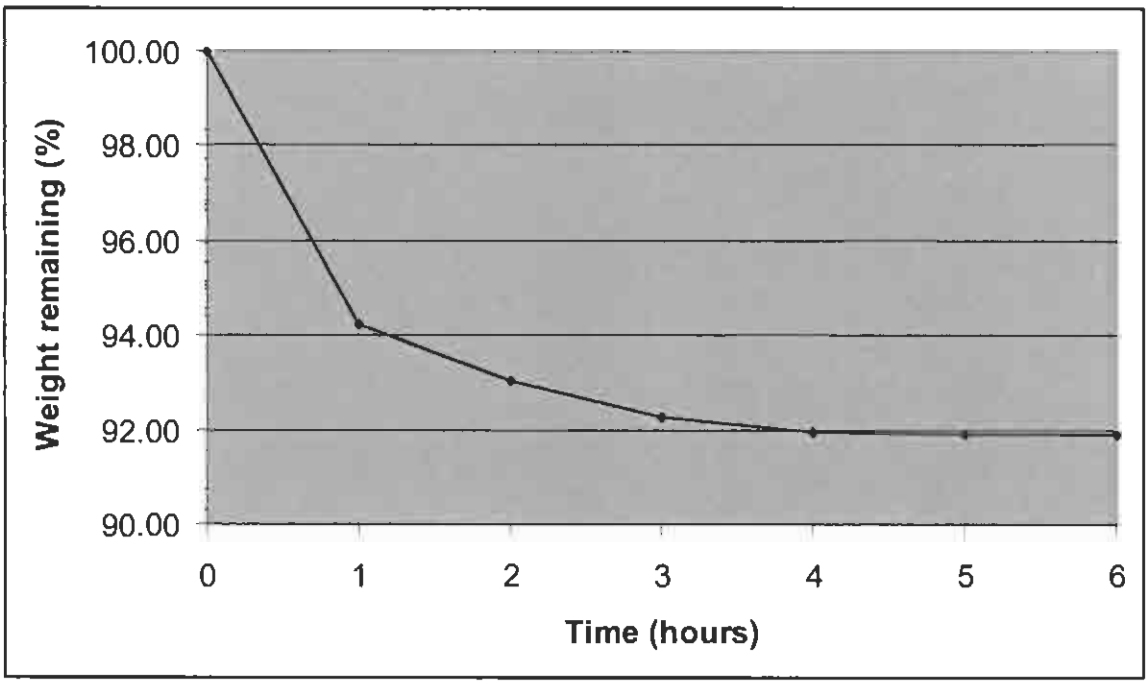

Figure 3.4: *Percentage weight of chitosan powder remaining as function of drying time.*

3.7.3.2 Moisture increase

The results of the moisture uptake are given in figure 3.5. The measurements values are given in table A.6 and A.7 in annexure A.

The chitosan used contained 8.10% of free moisture, of which most (5.7%) was lost in the first hour of drying. The chitosan could be dried completely in 6 hours. The moisture uptake was faster when the chitosan was stored at 40 °C and 75% RH and the percentage water uptake was also higher (8.93%) compared to storage at the lower temperature and RH (7.07%). Chitosan readily absorbs and looses moisture when stored under conditions where the RH varies and this characteristic is increased by an increase in temperature. This variation in the moisture content could be significant if it has an effect on the flowability of the chitosan powder and an experiment to establish this effect was therefore done.

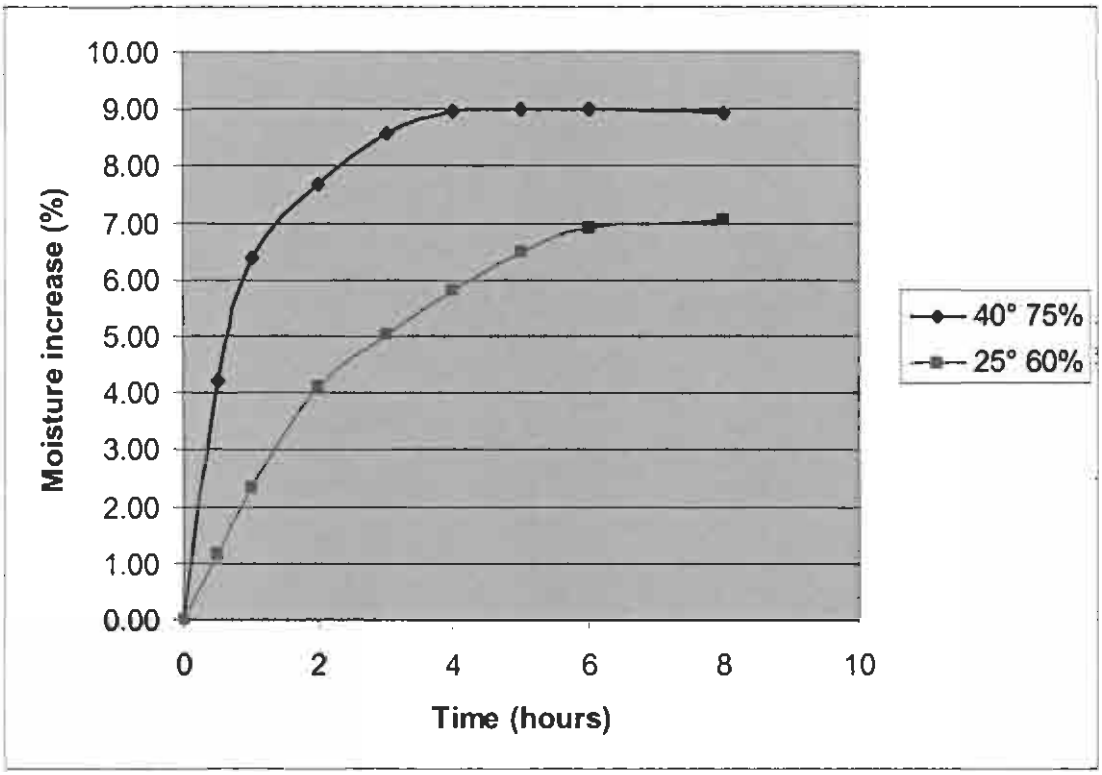

Figure 3.5: *Moisture uptake of chitosan at 25 °C 60% RH and 40 °C 75% RH*

3.8 THE EFFECT OF MOISTURE ON THE FLOWABILITY OF CHITOSAN

3.8.1 Introduction

The results of the previous experiment showed that storage of chitosan under different temperatures and relative humidities, changes the moisture content of the chitosan powder. To establish the extent of this change in moisture content on the flowability of chitosan, chitosan was stored under different relative humidities in closed containers and the flow characteristics were measured.

3.8.2 Methods

Chitosan was stored for 48 hours at 25 °C in desiccators containing saturated solutions of chemical salts. These saturated salt solutions will result in different relative humidities in the desiccators as given in table 3.3.

The tap density, angle of repose and critical orifice were measured and the composite index calculated for each fraction as described in section 3.5.1 and the materials used are given in annexure D.

Table 3.3: *Relative humidities of different chemical salts in a closed container.*

Chemical	Relative humidity (%)
Lithium chloride ($LiCl_2$)	11
Magnesium chloride ($MgCl_2$)	33
Magnesium nitrate ($MgNO_3$)	54
Sodium chloride (NaCl)	75

3.8.3 Results

The results of the measurements are given in table 3.4. The measurement data is given in table A.8 to A.11 in annexure A.

Table 3.4: *Results of powder flow of chitosan stored under different humidities.*

Chemical	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice (mm)	Composite index
None	30.917	1.4	44.23	10	50.8
LiCl	30.737	1.4	45.00	11	48.5
$MgCl_2$	32.879	1.5	44.62	12	46.0
$MgNO_3$	32.740	1.5	44.23	15	43.3
NaCl	36.062	1.6	45.38	16	37.1

All the flow indexes show that the flowability of the chitosan decreases with an increase in the moisture content under which it was stored. It is the most apparent in the critical orifice measurement and Carr's index. As a result, the composite index of the dry chitosan is 50.8 while that of the powder stored with the sodium chloride (75% RH) is 37.1. It is clear from this data that a high moisture content negatively effect the flow of chitosan and it would therefore be necessary to conduct experiments under conditions that are moisture and temperature regulated. All the

experiments in the study were therefore done at a laboratory temperature of between 25 – 30 °C and a relative humidity of approximately 35%.

3.9 DETERMINATION OF THE PARTICLE SIZE OF CHITOSAN

3.9.1 Introduction

It is known that particle size of powders play an important role in the flow characteristics of the powder. The particle size and particle size distribution of the chitosan used in the study were determined so that a particle size fraction, to be used in future experiments, could be identified.

3.9.2 Method

Several stainless steel sieves with a woven mesh were stacked on top of each other on a collector tray (The sieve with the smallest mesh were placed at the bottom followed by meshes which get progressively coarser towards the top of the series). Fifty (50) grams of chitosan were loaded onto the coarsest sieve of the assembled stack and placed on the mechanical vibration apparatus. The powder was sieved for 30 minutes and the amount of powder remaining on each sieve was weighed and the particle size fraction was calculated.

3.9.3 Results

The cumulative particle size distribution is given in figure 3.6.

From the particle size distribution it can be seen that a large percentage (44.70 %) of the particles falls between 63 -150 μm in size. Their is also a relatively large percentage of the particles smaller than 63 μm (8.52 %) which contributes to the poor flowability of the chitosan as well as a relatively large percentage above 212 μm (19.63 %).This makes it possible to separate the chitosan in different size fractions. As a result the effect of particle size on the flowability of the chitosan was determined.

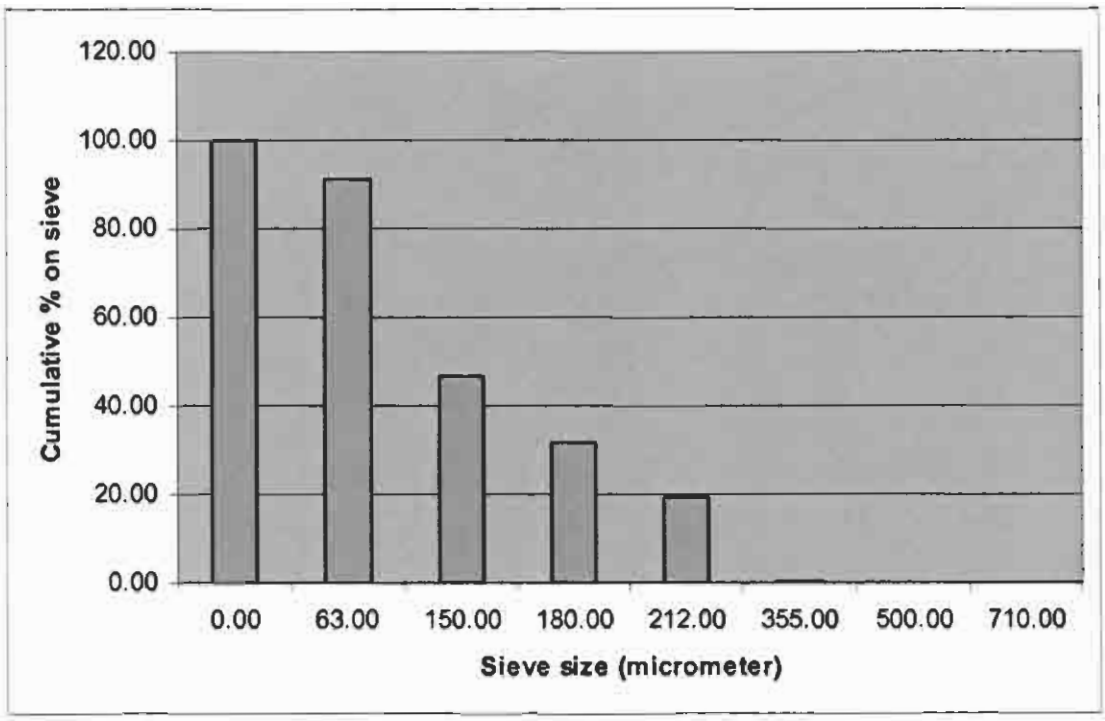

Figure 3.6: *Cumulative size distribution of chitosan powder.*

3.10 THE EFFECT OF PARTICLE SIZE ON THE FLOWABILITY OF CHITOSAN

3.10.1 Introduction

It is known that smaller particles will in general have a negative influence on the flowability of a powder because of the higher surface area per weight of the particles while the opposite is true for bigger particles. In this experiment, chitosan was sieved in four different particle size fractions and the effect on flowability measured.

3.10.2 Methods

Chitosan was sieved in four different size fractions namely < 90 μm; 90 – 150 μm; 150 – 212 μm and > 212 μm. The tap density, angle of repose and critical orifice were measured and the composite index calculated for each fraction as described in section 3.5.1.

Scanning electron microscope (SEM) pictures of the chitosan as received as well as that of the fraction > 212 μm were also taken and is given in figure 3.7 and figure 3.8 respectively.

3.10.3 Results

The results of the measurements are given in table 3.5. The measurement data is given in table A.12 to A.15 in annexure A.

Table 3.5: *Flowability of different size fractions of chitosan.*

Size (μm)	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice (mm)	Composite index
Unsieved	39.233	1.5	45.84	12	37.9
< 90	40.663	1.7	45.65	24	23.0
90 – 150	35.655	1.6	43.08	13	44.7
150 – 212	31.341	1.5	45.79	9	48.9
> 212	29.6650	1.4	43.15	9	54.9

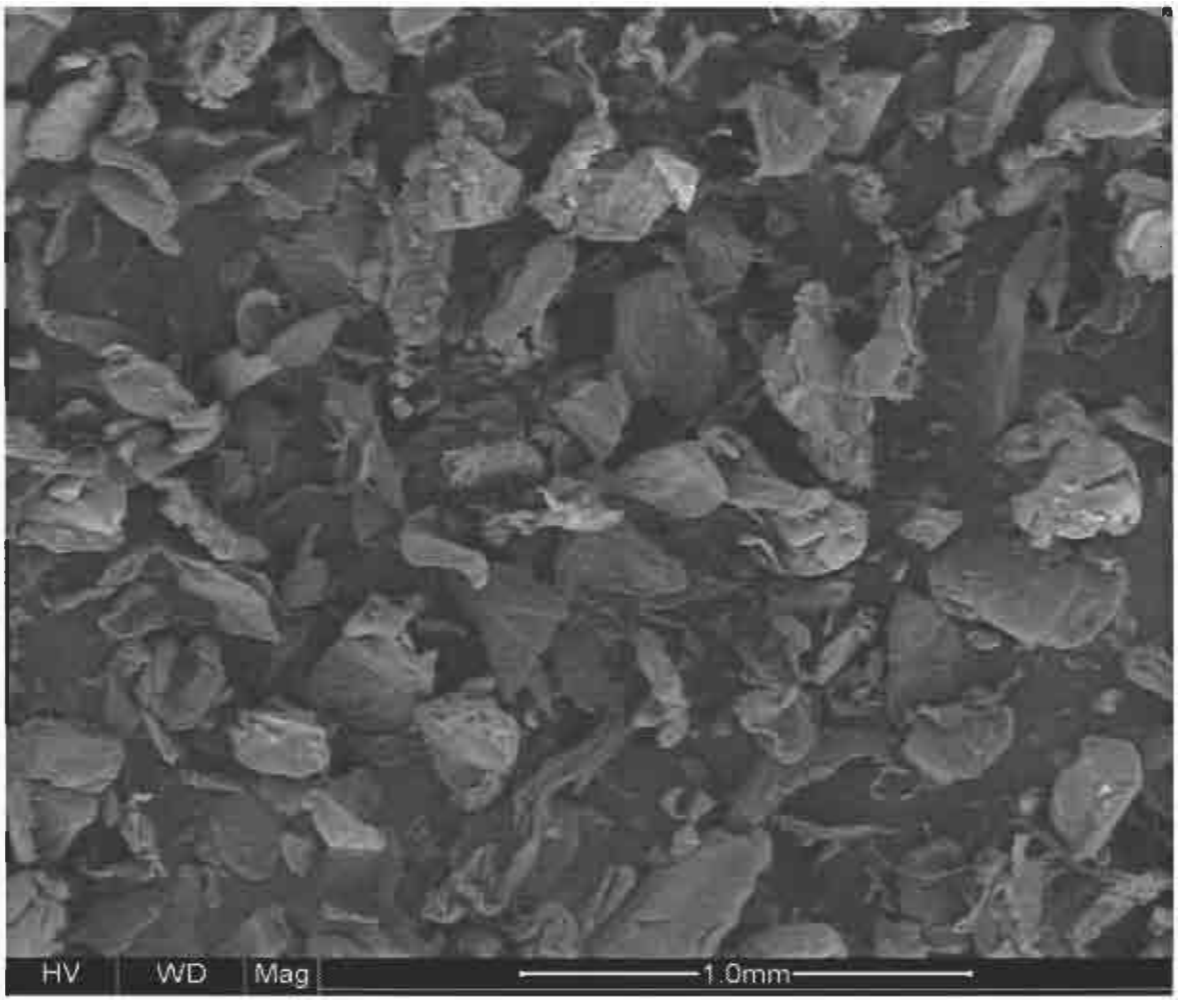

Figure 3.7: *SEM picture of chitosan used in the study (batch nr: 021010).*

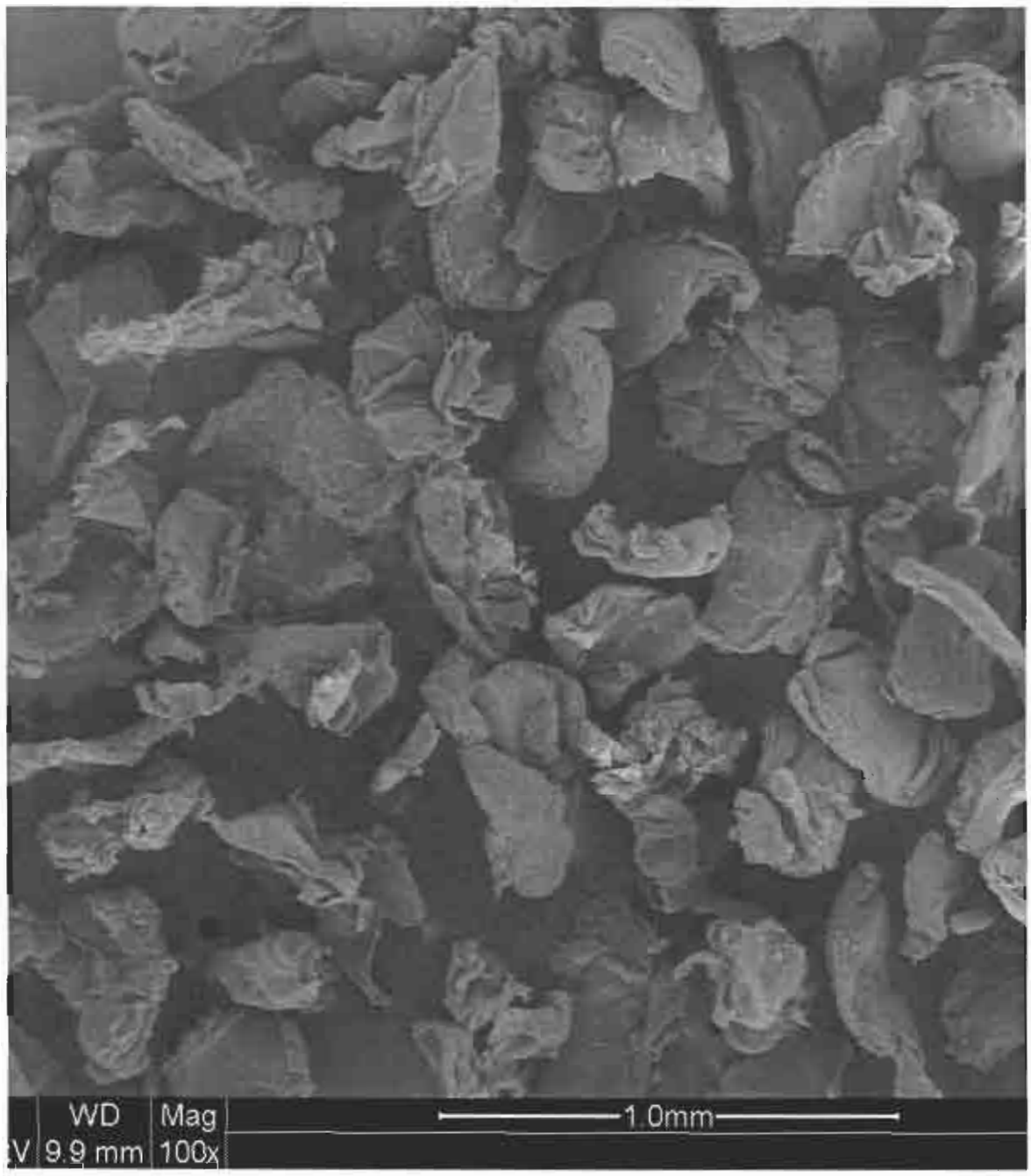

Figure 3.8: SEM picture of chitosan sieved: fraction > 212 μm *(batch nr: 021010).*

It can be seen from the data that the bigger the particle size, the better the flowability becomes. The smallest fraction (< 90 μm)) especially has a very poor flowability with a composite index of 23.0 while the fraction with a particle size of more than 212 μm has a composite index of 54.9 which is better than that of other excipients like Avicel pH200® (51.9) but not as good as those of free flowing excipients like Ludipress® (89.9) and Emcompress® (87.1).

3.11 THE EFFECT OF GLIDANTS ON THE FLOWABILITY OF CHITOSAN

3.11.1 Introduction

The flowability of chitosan can probably be increased with the addition of glidants. Two different glidants, Cab-O-Sil® and talc were chosen and the flowability measured at different concentrations of the glidants.

3.11.2 Method

Chitosan with a particle size bigger than 212 μm were used in the study. The chitosan were weighed and mixed with the glidant in a turbula mixer (Willy A. Bachhofen, Basel, Switzerland) for 5 minutes. The resulting powder mixtures contained either 1, 2 or 3% Cab-O-Sil® or 0.5, 0.75 or 1% talc.

The tap density, angle of repose and critical orifice were measured and the composite index calculated for each fraction as described in section 3.5.1.

3.11.3 Results

The results of the measurements are given in tables 3.6 and 3.7. The measurement data is given in table A.16 to A.23 in annexure A.

Table 3.6: *The effect of concentration Cab- O- Sil® on the flowability of chitosan with a particle size > 212 μm.*

Concentration (% w/w)	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice (mm)	Composite index
0	31.691	1.5	45.17	6	53.1
1	31.193	1.5	40.84	5	61.9
2	30.000	1.4	42.28	4	61.8
3	28.571	1.4	41.93	3	64.9

Table 3.7: *The effect of concentration talc on the flowability of chitosan with a particle size > 212 μm.*

Concentration (% w/w)	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice (mm)	Composite index
0	31.915	1.5	44.66	7	52.6
0.50	30.716	1.4	42.05	5	60.3
0.75	30.943	1.4	42.55	6	58.1
1.00	30.251	1.4	41.91	6	59.9

Both the glidants improve the flowability of the chitosan over the concentration range used. The improvement is apparent at small quantities of either glidants and the addition of higher concentrations did not show more improvement. It was therefore decide to use talc in a concentration of 0.5% as a glidant in future experiments.

3.12 THE EFFECT OF PARTICLE SIZE AND GLIDANT ON THE TABLET WEIGHT

3.12.1 Introduction

From the results of the previous experiment it was evident that an increase in the particle size (> 212 μm) and the addition of a glidant (talc 0.5%) improve the flow rate when measured with the apparatus and methods as described. To establish if the improvement in the flow rate has practical implications when the die of the tablet press is filled, the effect of the increase in particle size and the addition of talc on the filling of the die and hence on the mass of the tablets, were investigated.

3.12.2 Methods

Twenty tablets each of the raw material, chitosan with a particle size of > 212 μm and chitosan with a particle size of > 212 μm with 0.5 % talc, were compressed in a tablet press (Manesty F3, Manesty Machines, Liverpool, England). The diameter of the tablets were 4 mm and the powders were left in laboratory conditions (25 – 30 °C, ±

35% RH) for 24 hours before tableting. The tablets were compressed in a single run producing the 20 required tablets and were weighed immediately after compression.

3.12.3 Results

The results of the measurement are given in table A.24 in annexure A.

The percentage relative standard deviation (% RDA) for the tablets compressed with chitosan raw material was the highest (1.42) while the % RDA for the tablets compressed with chitosan with a particle size of > 212 μm were lower at 1.03. The tablets compressed with the chitosan containing 0.5% talc had the lowest % RDA (0.43).

These values confirm that the die was filled more evenly with the chitosan containing talc as a glidant because of the better flow properties of the powder.

The better flow properties of chitosan on the addition of a glidant is important in the filling of the die during the compression of tablets. This will lead to tablets with a better uniformity of content of the drugs in the tablets as well as more reproducible tablets for further investigations.

3.13 SUMMARY

In order to investigate the influence of formulation factors on the properties of the minitablets that are to be used as a multi-unit colonic delivery system, it is necessary to ensure that the chitosan used has satisfactory powder flow characteristics. If the flow of the chitosan is poor, it will lead to an unacceptable variation in the tablets (such as weight, thickness, disintegration and strength) causing the effect of formulation factors to be obscured by the inherent difference between the tablets.

When measured with the methods as described above and the composite flow index calculated (see table 3.8), it can be seen that the flow characteristics of the chitosan

can be improved from a value of 32.7 to a value of 58.8 depending on the stored conditions, particle size and the addition of glidants.

Table 3.8: *Flow characteristics of chitosan.*

	Carr's index (%)	Hausner ratio	Angle of repose (°)	Critical orifice diameter (mm)	Composite index
Stored under high RH	36.062	1.6	45.38	16	37.1
Chitosan as received	39.233	1.5	45.84	12	37.9
Store under low RH	30.917	1.4	44.23	10	50.8
Size > 212 μm	29.665	1.4	43.15	9	54.9
Size > 212 μm + talc 0.5%	30.716	1.4	42.05	5	60.3

The importance of this measured flow characteristics can be seen when the percentage standard deviation of the tablet weight of tablets produced with chitosan as received, and chitosan with a particle size of > 212 μm with or without talc as a glidant is compared.

The percentage standard deviation (% RDA) for the tablets pressed with chitosan as received was the highest (1.42) while the % RDA for the tablets made with chitosan with a particle size of > 212 μm decreased to 1.03 and that for the tablets made of the chitosan containing 0.5 % talc was 0.43.

Therefore in the further investigations, only chitosan with the bigger particle size with talc 0.5% as a glidant will be used while the powders will be stored under laboratory conditions (25 – 30 °C, ± 35% RH) which will be monitored.

CHAPTER 4

COMPRESSIBILITY OF CHITOSAN

4.1 INTRODUCTION

The compressibility of chitosan, together with the flowability of the powder, are the most important challenges or problems that must be solved before tablets can be compressed successfully. Since the compressibility characteristics of chitosan are poor, the tablets in the initial part of the study were compressed with the modified infrared (IR) press that allowed for more controlled and higher compression forces. Factors such as compression force, moisture content of the powder, powder weight and size fraction of the powder on the physical properties of the tablets were investigated.

4.2 APPARATUS AND METHODS

4.2.1 Tablet compression

For each experiment, flat faced tablets with a diameter of 8 mm were prepared at compression forces ranging from 15 - 20 bar. A modified IR press were used to press the tablets according to the following procedure (Marais, 2000:66; Louw, 2003:64).

A steel die with an opening of 8 mm was placed on a steel base plate and the bottom opening was sealed using a steel pellet. The amount of chitosan powder needed was weighed and transferred to the die. The top plunger was inserted through the top opening of the die and slightly pressed down into the die. The casing with the die and plungers were inserted into the press and the plunger screw adjusted to touch the top plunger. A compression cycle was initiated on the computer. The pressure increased to the set compression force and was held at around the required compression force for 45 seconds after which the compression was stopped. The forces exerted during the compression cycle were logged on the computer through an interface. The tablet were then manually removed from the die, weighed and the average compression force calculated. An illustration of the compression unit can be seen in figure 4.1.

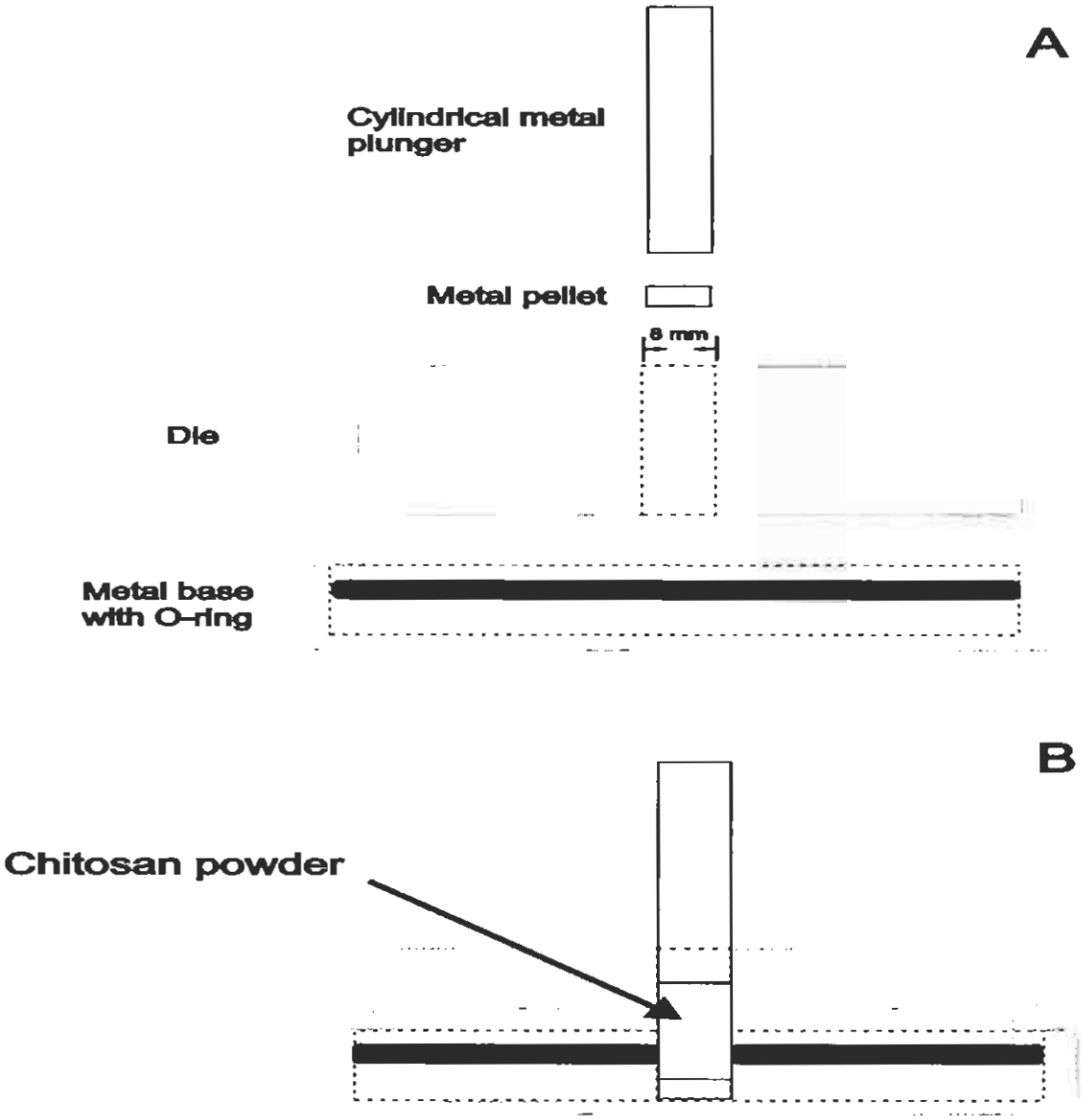

Figure 4.1: *An illustration of the compression unit showing a) the parts and b) the assembled unit.*

4.2.2 Tensile strength

The crushing strength, diameter and thickness of the tablets were determined using a Pharmatest® tablet test unit (model PTB-311, Hainburg, West-Germany). The tensile strength was then calculated (Fell & Newton, 1968:658) using the equation:

$$T = 2P / \pi Dt$$

where:

T is the tensile strength (N/mm^2), P is the crushing strength (N), D is the diameter (mm) and t is the thickness (mm).

4.2.3 Disintegration

4.2.3.1 Apparatus

Disintegration studies were done in a 3 station Erweka ZT503 disintegration apparatus (Erweka Apparatbau GmBH, Hausenstamm, Germany). The apparatus was fitted with a thermostat that kept the test medium at 37 °C.

4.2.3.2 Method

The test vessels (800 ml glass beaker) were fitted into the heated water bath, filled with the test medium to the correct height and allowed to equilibrate at the specified temperature. A tablet was placed in each of the test tubes in the test stations, the stations placed into the test medium and the test started at time (t) = 0. The time was noted at which all the particles of a tablet passed through the sieve at the bottom of the test tube.

The tablets however often did not disintegrate (or take an unpractical time to disintegrate) but rather only swell and/or break in smaller pieces at compression forces high enough to prepare tablets of acceptable crushing strength. In order to distinguish between the disintegration characteristics of the tablets, an arbitrary scale was established using the appearance of the tablets as an indicator of the disintegration behaviour and not the actual disintegration time.

Table 4.1: *Arbitrary scale for the interpretation of the disintegration of chitosan tablets.*

Scale	Description
1	Disintegrate totally
2	Disintegrate except for very fine particles remaining
3	Disintegrate but coarse particles or pieces remaining
4	Tablet split into 2 pieces
5	Tablet split but stays in one piece
6	Tablet stays intact

4.2.4 Wettability

A device (see figure 4.2 and 4.3) was developed to measure the amount of water absorbed by the chitosan tablets and the subsequent swelling in the tablets.

A chamber that could be filled with approximately 10 ml water was machined from a transparent plastic with two tubes leading into the chamber. The one tube (rubber/plastic) was connected to a glass reservoir containing water. This reservoir was used to prime the system with water. The other tube (glass) lead into a container with water that was placed on a balance. A sinter glass disc was placed on the chamber so that it could be wetted by the water and covered with a shutter. Another transparent holder was machined with a 8 mm shaft and a plunger fitting into the shaft. A tablet with a 8 mm diameter could be placed into the one end of the shaft while on the other end a gauge was fitted measuring the depth of the plunger. The tablet holding device was place on top of the water holding chamber in such a way that the tablet was in contact with the shutter and the plunger touching the top of the tablet.

A computer was fitted to the balance to log the weight of the water container every second using software obtained from Mettler Toledo (BalanceLink). The shutter was then removed and the plunger depth recorded every ten seconds.

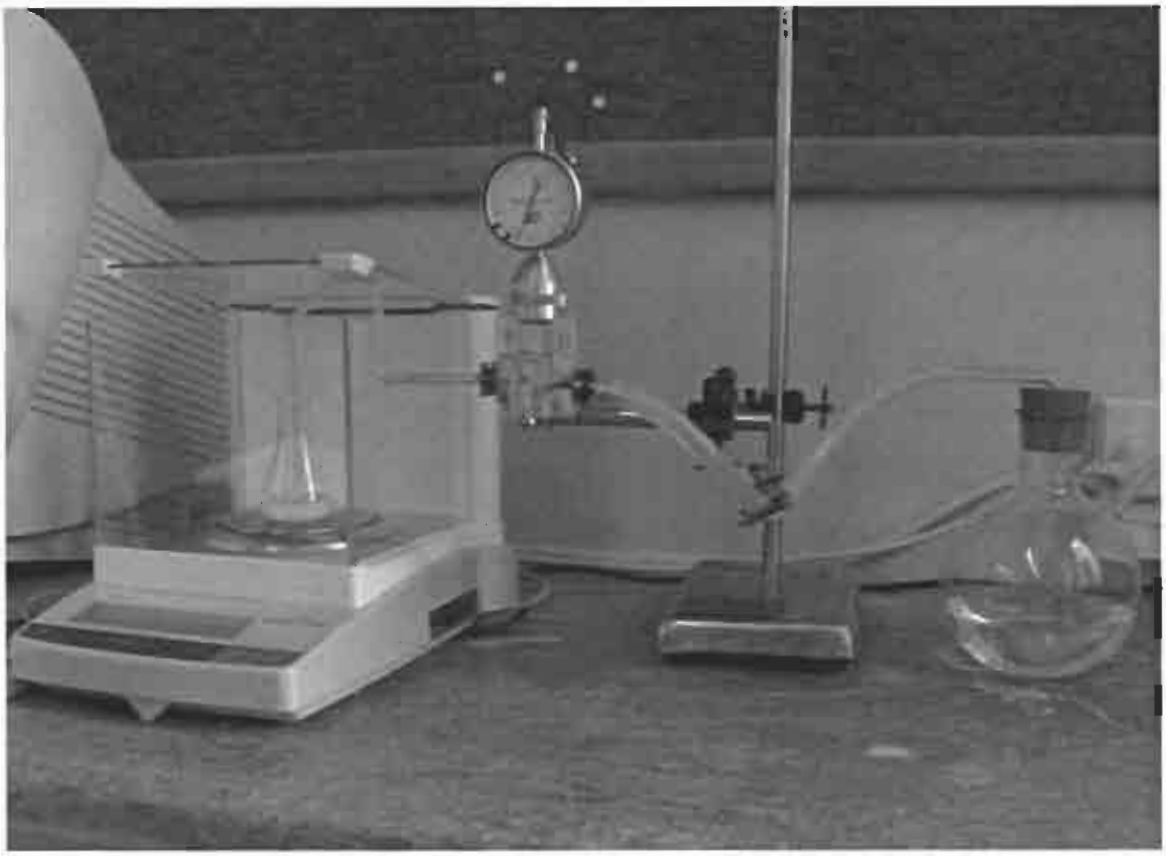

Figure 4.2: *The apparatus for measuring the wettability and swelling characteristics of chitosan tablets.*

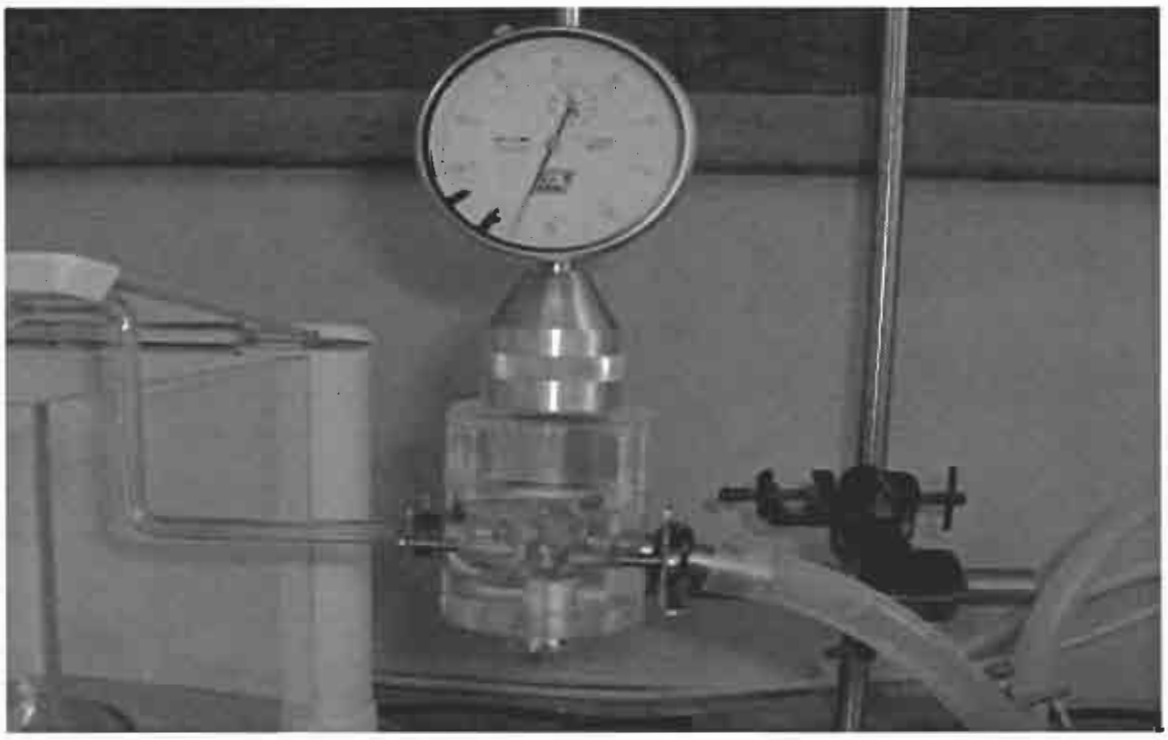

Figure 4.3: *The water and tablet holding chamber of the apparatus for measuring the wettability and swelling of chitosan tablets.*

4.3 THE INFLUENCE OF COMPRESSION FORCE AND MOISTURE CONTENT OF THE CHITOSAN POWDER ON THE TENSILE STRENGTH OF CHITOSAN TABLETS

4.3.1 Introduction

As indicated in chapter 3, the moisture content of the chitosan powder influenced the flow properties of the powder. An investigation was done to determine the effect of the moisture content of chitosan powder on the tensile strength of the tablets. Tablets were also compressed at different compression forces to establish its effect on the tensile strength of the tablets and also to establish the compression force that could be used to prepare tablets of acceptable physical properties.

4.3.2 Method

4.3.2.1 Sample preparation

a) Sample 1

Ten grams chitosan powder were dried at 60 °C in an oven for 24 hours at a relative humidity (RH) of 3.5%. The dried powder was placed in a dessicator for 2 hours to allow the powder to cool down to laboratory temperature. Tablets were then compressed at different compression forces.

b) Sample 2

A sample of chitosan was left in the laboratory for 24 hours at a RH of between 50% and 55% after which the tablets were compressed. To determine the moisture content of the powder, approximately 10 g of the sample were weighed accurately, dried for 24 hours at 60 °C and then weighed again. The percentage moisture that the powder contained was calculated.

c) Sample 3

A sample of chitosan powder was placed in a glass container with a RH that was regulated between 80% and 90%, for 4 hours. The tablets were then compressed

and the percentage moisture that the powder contained was calculated as described for sample 2.

4.3.3 Results

The results of the investigation are given in figure 4.4 and the measurement values in table A.25 – A.27 in annexure A.

The tensile strength of the tablets increased with increasing compression force irrespective of the moisture content. At lower compression forces the relation between the tensile strength and compression force was linear but linearity was lost at higher compression forces due to the tensile strength reaching a maximum value.

The tensile strength of the tablets compressed with the dried chitosan powder were less at all the compression forces used, compared to that of tablets compressed with the powder that was stored under laboratory conditions (containing 9.39% moisture) and that of the powder stored at a high RH (containing 11.86% moisture). A compression force of between 15 and 19 bar yielded tablets of acceptable crushing strength which were used in follow-up experiments.

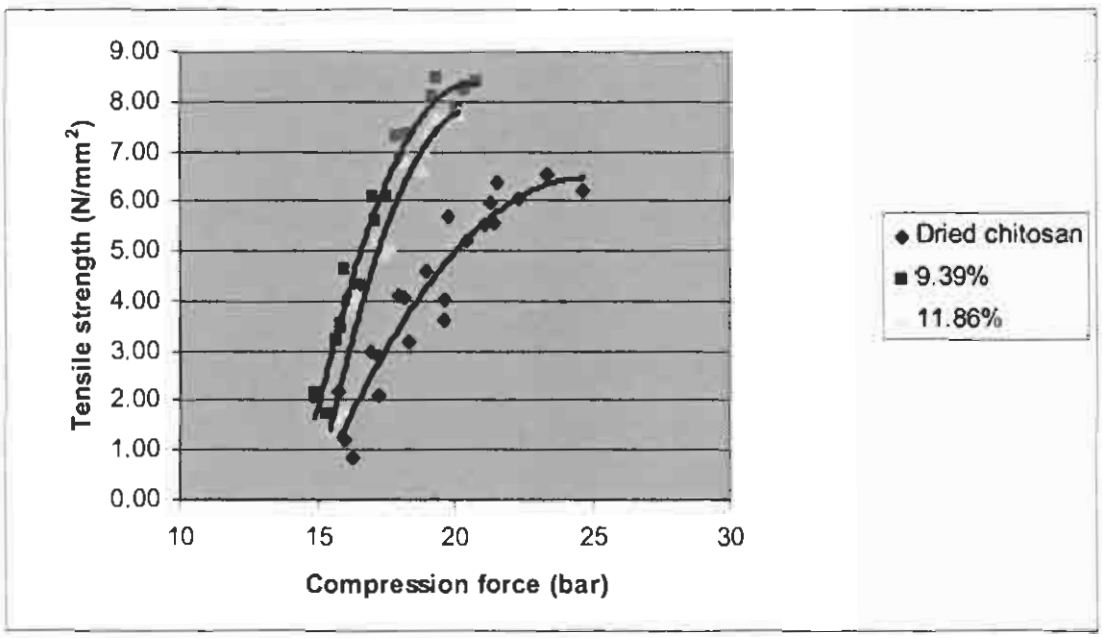

Figure 4.4: *The influence of moisture content of chitosan powder on the tensile strength of chitosan tablets.*

The moisture content of the chitosan powder did have an influence on the tensile strength of the tablets. In further investigations, the powders used to compress the tablets were stored under the same laboratory conditions (25 – 30 °C, ± 35% RH) and the tablets prepared at compression forces between 15 and 19 bar.

4.4 THE INFLUENCE OF THE POWDER WEIGHT AND SIZE FRACTION ON THE TENSILE STRENGTH OF CHITOSAN TABLETS

4.4.1 Introduction

An experiment was done to establish the effect of different powder weights and size fractions of the chitosan powder on the tensile strength of the tablets.

4.4.2 Methods

Chitosan tablets (8 mm in diameter) of two different size fractions of the powder (< 90 µm and > 212 µm) and different weights of the powder (150 mg, 175 mg and 200 mg) were compressed in the modified IR press at different compression forces. The crushing strength, diameter, thickness and weight of the tablets were determined and the tensile strength calculated.

4.4.3 Results

The results of the investigation are given in figure 4.5 – 4.9 and the measurement values in table A.28 – A.33 in annexure A.

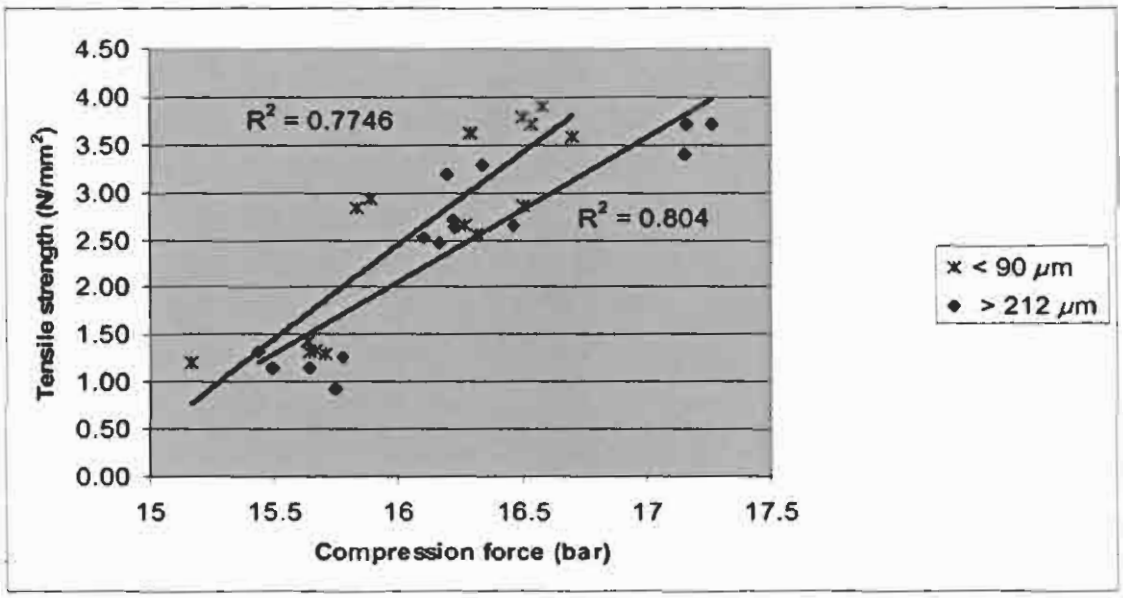

Figure 4.5: *The influence of particle size on the tensile strength of 150 mg chitosan tablets.*

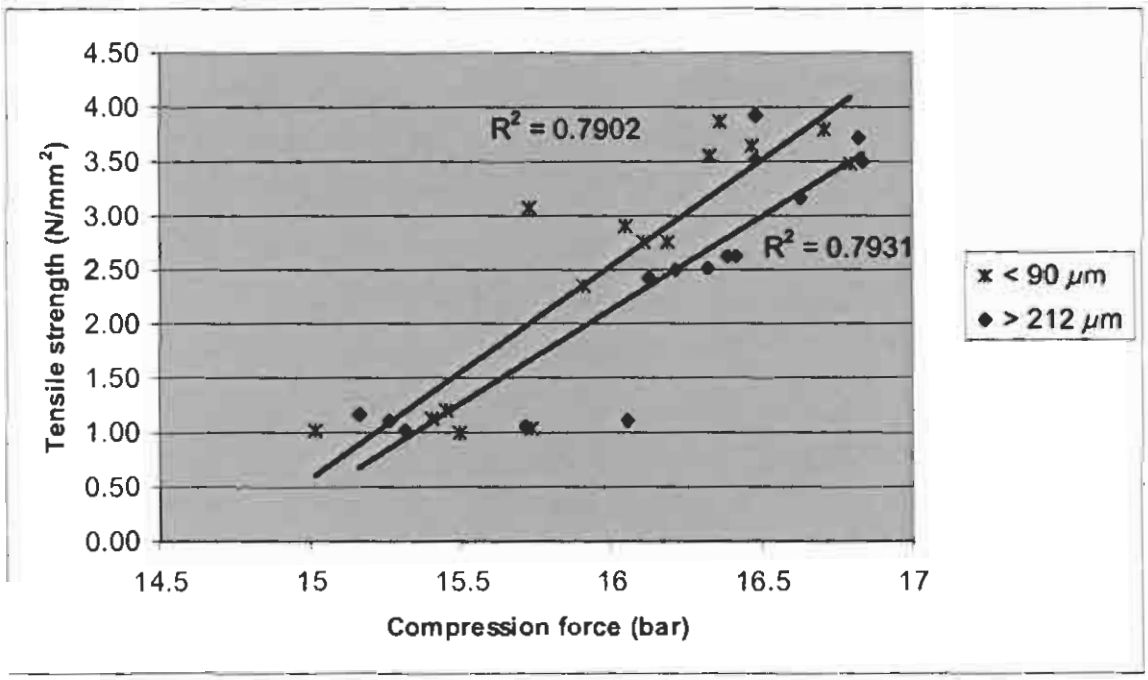

Figure 4.6: *The influence of particle size on the tensile strength of 175 mg chitosan tablets.*

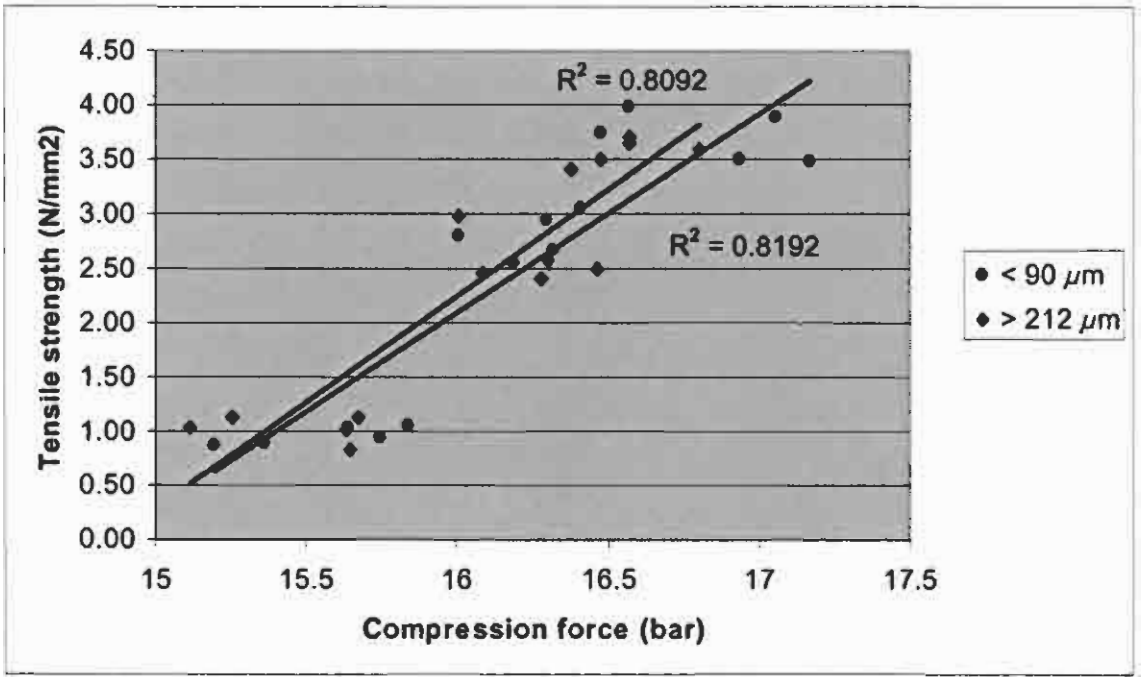

Figure 4.7: *The influence of particle size on the tensile strength of 200 mg chitosan tablets.*

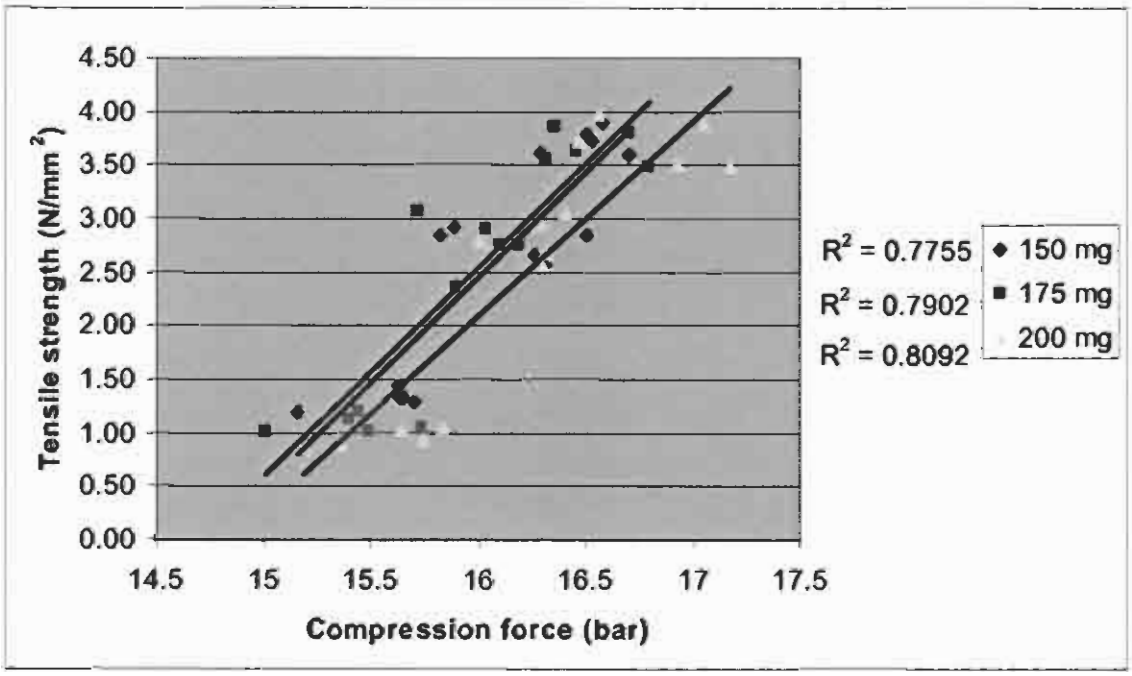

Figure 4.8: *The influence of powder weight of chitosan with a particle size < 90 µm on the tensile strength of chitosan tablets.*

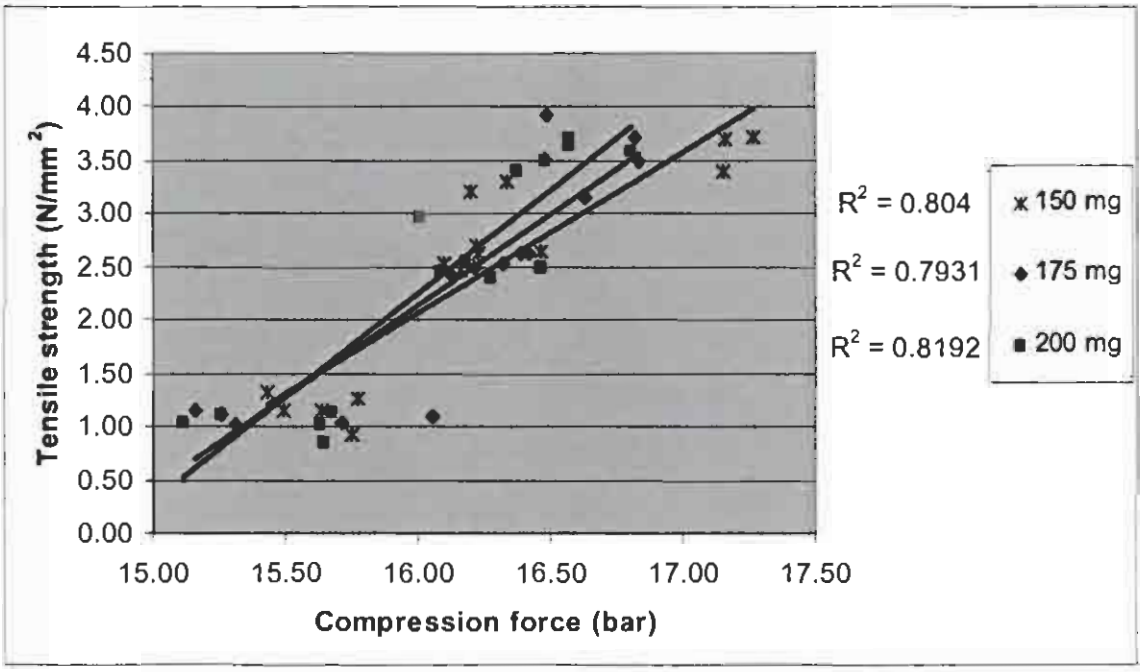

Figure 4.9: *The influence of powder weight of chitosan with a particle size > 212 µm on the tensile strength of chitosan tablets.*

a) Influence of particle size

In both the 150 and 175 mg tablets, the tensile strength of the tablets prepared from the < 90 µm sieve fraction was higher than that of the > 212 µm fraction, although the difference were less pronounced in the heavier tablets. In the 200 mg tablets, however, the rank order changed resulting in tablets prepared from the > 212 µm fraction exhibiting higher tensile strength compared to the < 90 µm sieve fraction.

It is clear from this results that the particle size does have an influence on the compaction of the powder and therefore on the tensile strength of the tablets. As a result, and because of the better powder flow characteristics, it was decided to only use the powder fraction > 212 µm in further investigations.

b) Influence of tablet weight

Tablets that were prepared from the fraction with the smaller particle size (< 90 µm) and weighing 150 and 175 mg showed no significant difference in the tensile

strength. The tablets of 200 mg however, showed a lower tensile strength at all the compression forces. When the fraction > 212 μm was used, the tensile strength of the tablets increased as the tablet weight increases.

The amount of the chitosan powder used in the compression of the tablets (tablet weight) also has an effect on the tablet characteristics. It is clear from this investigation that the tablet weight need to be considered when tablet characteristics are compared.

4.5 THE INFLUENCE OF TALC ON THE TENSILE STRENGTH OF CHITOSAN TABLETS

4.5.1 Introduction

To produce minitablets of chitosan, it was necessary to improve the flowability of the chitosan powder to ensure that the powder consistently fills the die of the tablet press with every compression cycle. In order to achieve this flowability, talc was added to the chitosan at concentrations ranging from 0.5% to 1.0%. The effect of the talc concentration was measured in terms of its effect on the tensile strength of the tablets.

4.5.2 Method

Powder mixtures were prepared with chitosan (particle size > 212 µm) and talc at different concentrations (0.5%; 0.75% and 1.0% w/w). Five tablets of each mixture were compressed with the IR press at each of three different compression forces ranging between 16 and 18.5 bar.

4.5.3 Results

The results are presented in figure 4.10 and the measurement values in table A.34 – A.37 in annexure A.

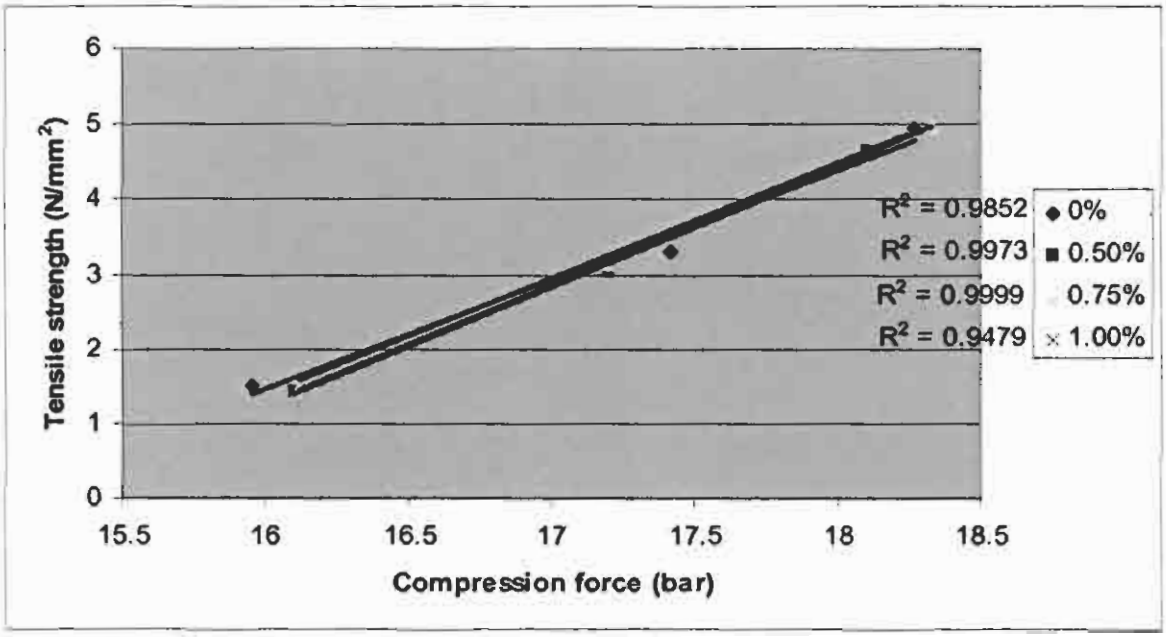

Figure 4.10: *Tensile strength of chitosan tablets as a function of the percentage talc.*

In section 3.11 the influence of talc on the flow characteristics of chitosan was investigated and it was found that talc improved the flowability of the chitosan over the concentration range used (0.5% to 1.0%) and that the improvement is apparent at the smallest quantities used. The addition of talc at concentrations of 0.75% and 1.0% did not show an improvement over a concentration of 0.5% talc.

It is clear from the above results that the talc, irrespective of the concentration, did not influence the tensile strength of the tablets at any compression force. Due to these results and also because of the improvement in flowability, 0.5% talc was included in all the mixtures prepared in further investigations.

4.6 THE INFLUENCE OF COMPRESSION FORCE ON THE DISINTEGRATION OF CHITOSAN TABLETS

4.6.1 Introduction

Tablets that are formulated for local drug delivery in the colon must have a slow disintegration time to prevent the dumping of the total amount of drug as soon as it reaches the colon. The disintegration time and the wettability of the tablets are

therefore important in the successful formulation of tablets for drug delivery in the colon. The most important factor that influences the disintegration and wettability of tablets are the compression force at which the tablets are compressed.

In this experiment the disintegration of chitosan tablets as a function of compression force was investigated.

4.6.2 Method

Six chitosan tablets, each weighing 150 mg, were compressed in the modified IR press at each of three different compression forces ranging between 16 and 18 bar. The disintegration times were measured using an Erweka disintegration apparatus. The tests were done in distilled water at 37 °C.

4.6.3 Results

The results are given in figure 4.11 and the measurement values in table A.38 in annexure A.

In the experiment the actual disintegration time was measured and not the arbitrary scale. An increase in the compression force resulted in an increase in the disintegration time of the tablets from 3.1 minutes to 566.2 minutes. The tablets did however absorb water and swelled within a few minutes (between 2 minutes for the tablets compressed at the lower compression force and 5 minutes for the tablets compressed at the higher compression force).

The comparative fast swelling of the tablets as compared to the disintegration time suggested that the tablet might be very porous. In the light of the above data, it was decided to investigate the wettability of the tablets as a means to describe the influence of compression force on the characteristics of the tablets.

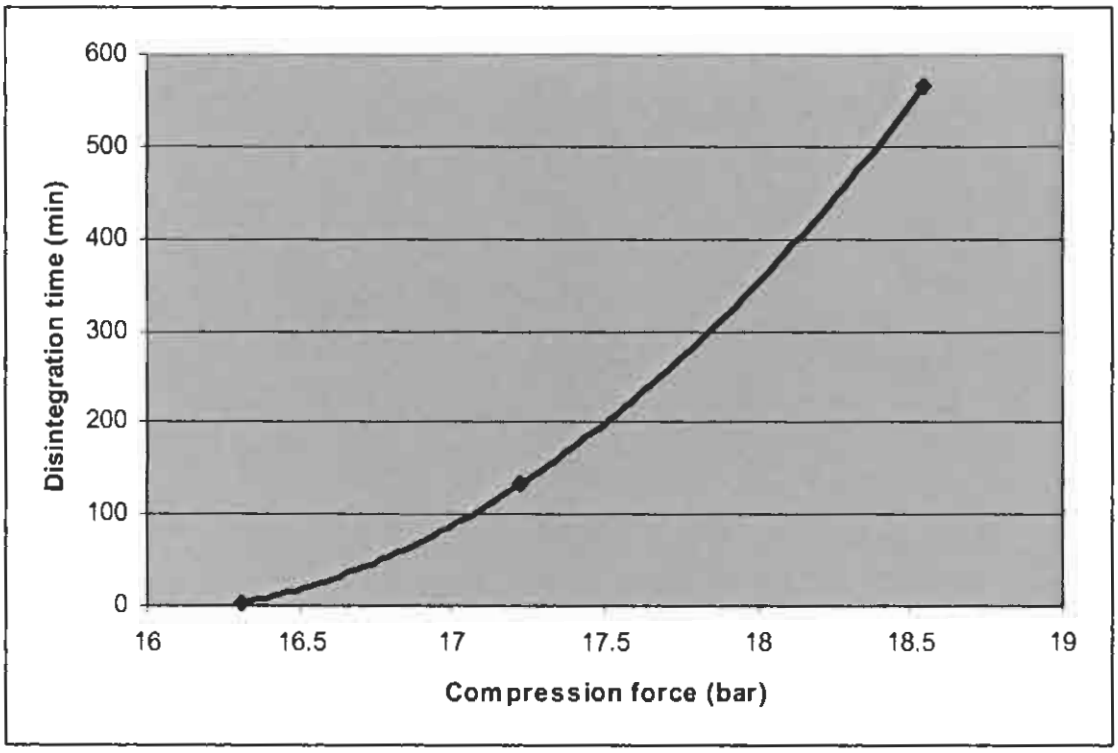

Figure 4.11: *The influence of compression force on the disintegration time of chitosan tablets.*

4.7 THE INFLUENCE OF COMPRESSION FORCE ON THE WETTABILITY OF CHITOSAN TABLETS

4.7.1 Introduction

The wettability of the tablets will be a valuable parameter to describe the porosity of the tablets. The wettability could therefore be an indication as to whether the chitosan particles were just loosely compacted or whether a tablet was formed. It would also give an indication of the compression force needed to compress tablets with acceptable dissolution characteristics should a drug be incorporated into the tablet.

The wettability can be measured by the amount of water the tablet absorbs per time unit and by the swelling of the tablet that occurs because of the absorption of the water.

4.7.2 Method

Seven tablets, each weighing approximately 150 mg and with a diameter of 8 mm were compressed at four different compression forces between 15,51 and 19,65 bar. The amount of water absorbed as well as the thickness of each tablet were recorded with the apparatus that was developed (see section 4.2.4).

4.7.3 Results

The average amount of water absorbed against time for the tablets compressed at each compression force is shown in figure 4.12 and the average increase in the thickness of the tablets prepared at each compression force is presented in figure 4.13. Figure 4.14 shows the total amount of water absorbed by the tablets as a function of the compression force.

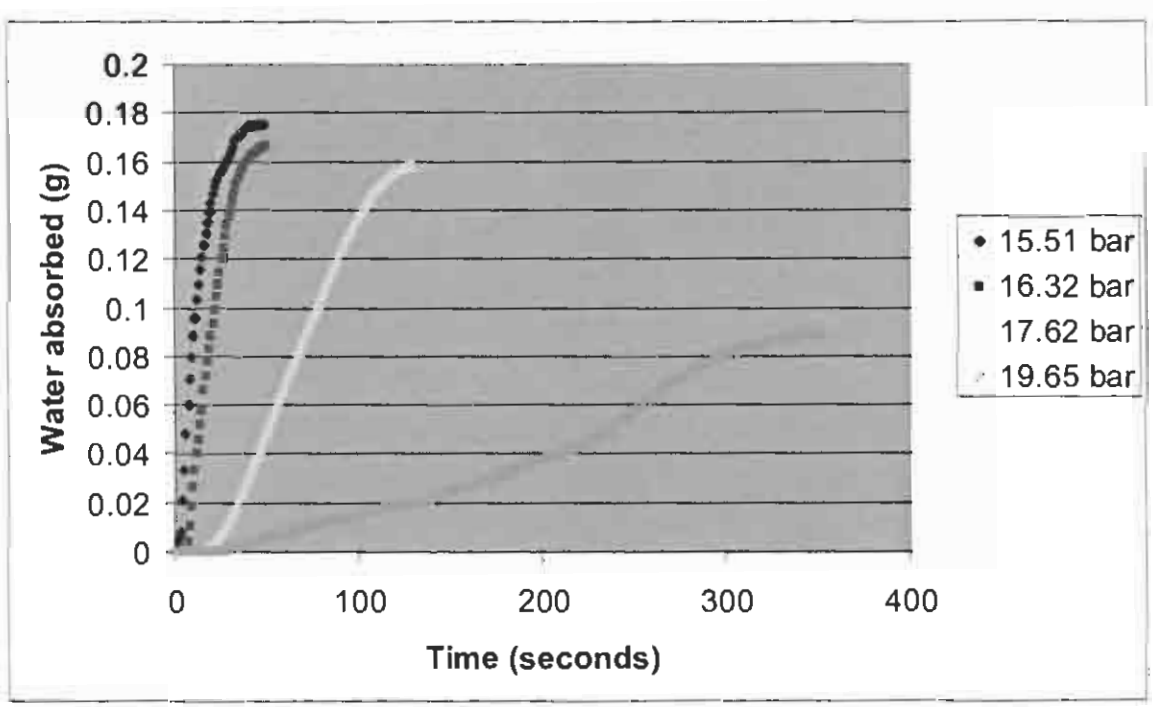

Figure 4.12: *Amount of water absorbed against time of tablets compressed at different compression forces.*

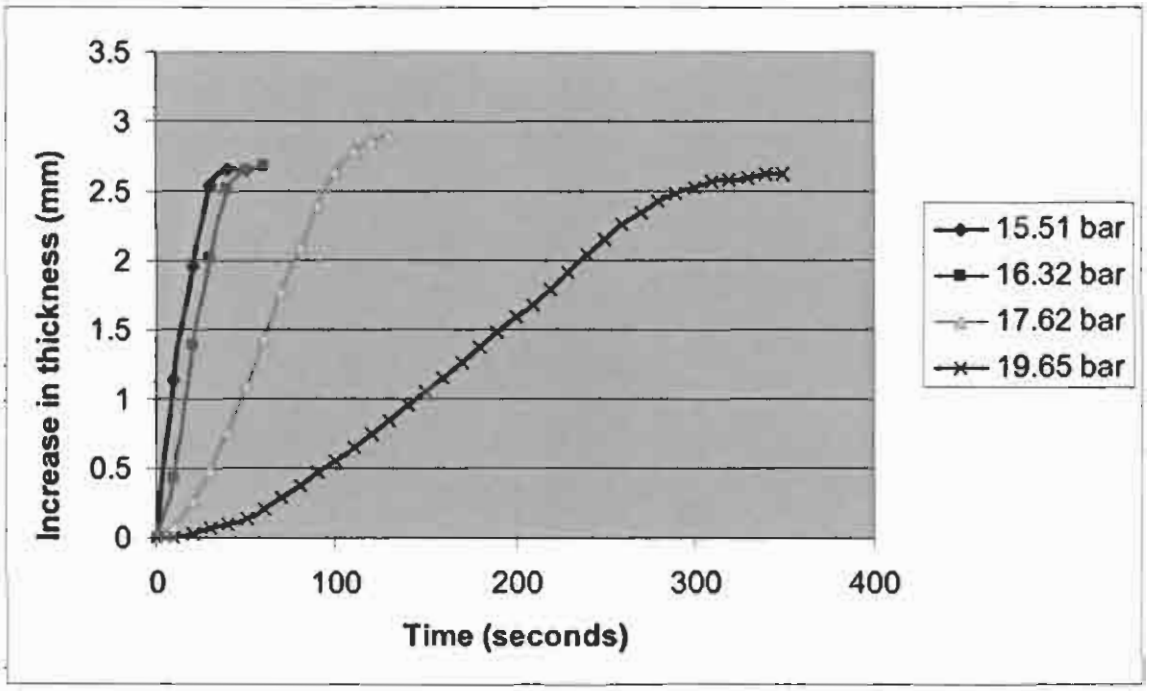

Figure 4.13: *Increase in thickness of tablets compressed at different compression forces.*

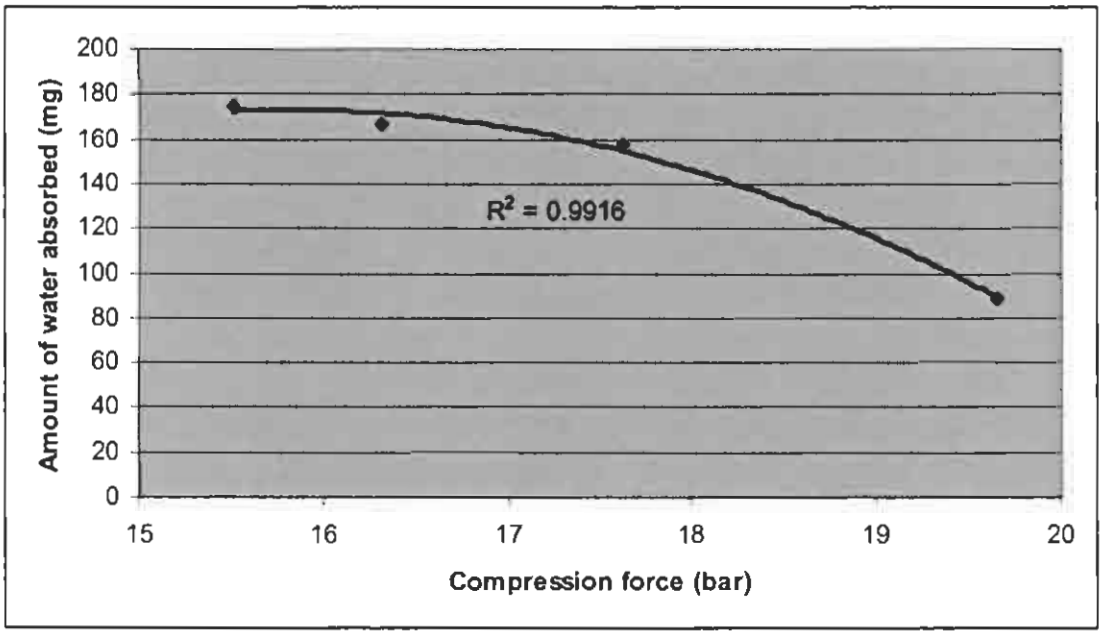

Figure 4.14: *Amount of water absorbed at different compression forces.*

The results (figure **4**.12) showed that an increase in the compression force resulted in a decrease in the amount of water that the tablets could absorb (from 175 mg at the lower compression force of 15.51 bar to 89 mg for the tablets compressed at 19.65 bar). This was probably because of the decrease in the openings (void spaces) between the particles as the compression force increased.

The relationship between the water absorption and the compression force was not linear and at higher compression forces, the decrease in the amount of water absorbed was much more markedly with a small change in the compression force (see figure 4.14). It could be seen that the amount of water absorbed at compression forces of between 15.5 and 17.5 bar, decreased from approximately 175 mg to 160 mg (8.6%) while at 19.5 bar the absorption decreased to 89 mg (44.4%).

This data suggested that the chitosan can only be compressed at high compression forces and that the tablets compressed at lower compression forces are not properly compacted.

The increase in the thickness of the tablets confirmed this compaction of chitosan as all the tablets increased in thickness by between 2.63 to 2.90 mm for all the compression forces used. There was a difference of only 0.02 mm in the thickness of the tablets compressed at 15.5 bar and those compressed at 19.5 bar, while the water absorbed at these compression forces differed by 86 mg (see figure 4.13). This can be due to the fact that the tablets are very porous and that the water was absorbed into the openings between the particles.

The absorption rate of the water showed a decrease with an increase in the compression force (figure 4.12). At the lower compression forces, the maximum amount of water absorbed was reached at approximately 50 seconds while at the higher force used (19.65 bar) it was reached only after approximately 300 seconds. This suggested that the porosity decreased as the compression force increases.

4.8 MODIFICATION OF THE ECCENTRIC TABLET PRESS

Tablets that are formulated for local drug delivery in the colon (such as in the case of some of the anti-inflammatory drugs used in inflammatory bowel disease) must have a slow disintegration time to prevent the dumping of the total amount of drug as soon as it reaches the colon. The relative long transit time through the colon adds to the fact that these tablets must have a slow disintegrating matrix to allow drug diffusion out of the tablet as it moves through the colon. For this reason the poor compression

characteristics of chitosan is a challenge that must be solved in order to directly compress chitosan tablets.

The poor compression of chitosan may be due to inherent poor binding between the chitosan particles and external factors such as the moisture content of the powders. The problem is aggravated by the fact that chitosan has a very low packing density and therefore the die can only be filled with a small amount per weight of the powder. If the die is filled with the powder and then compressed (especially at high compression forces), the compressed volume of this powder is obviously very small and the resulting tablets (if tablets can be compressed at all) have a low crushing strength because of their porous nature. Figure 4.15 and 4.16 show the porosity of chitosan tablets compressed at different compression forces on the eccentric tablet press.

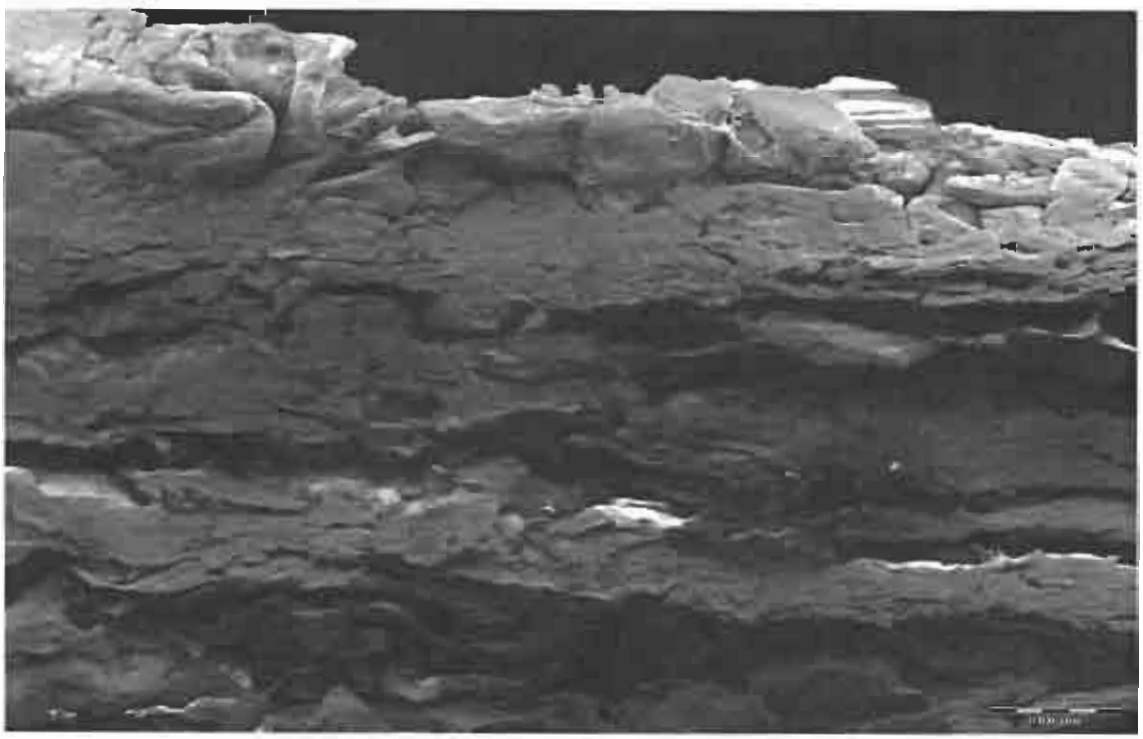

Figure 4.15: *SEM picture of a chitosan tablet compressed at a setting of 36.*

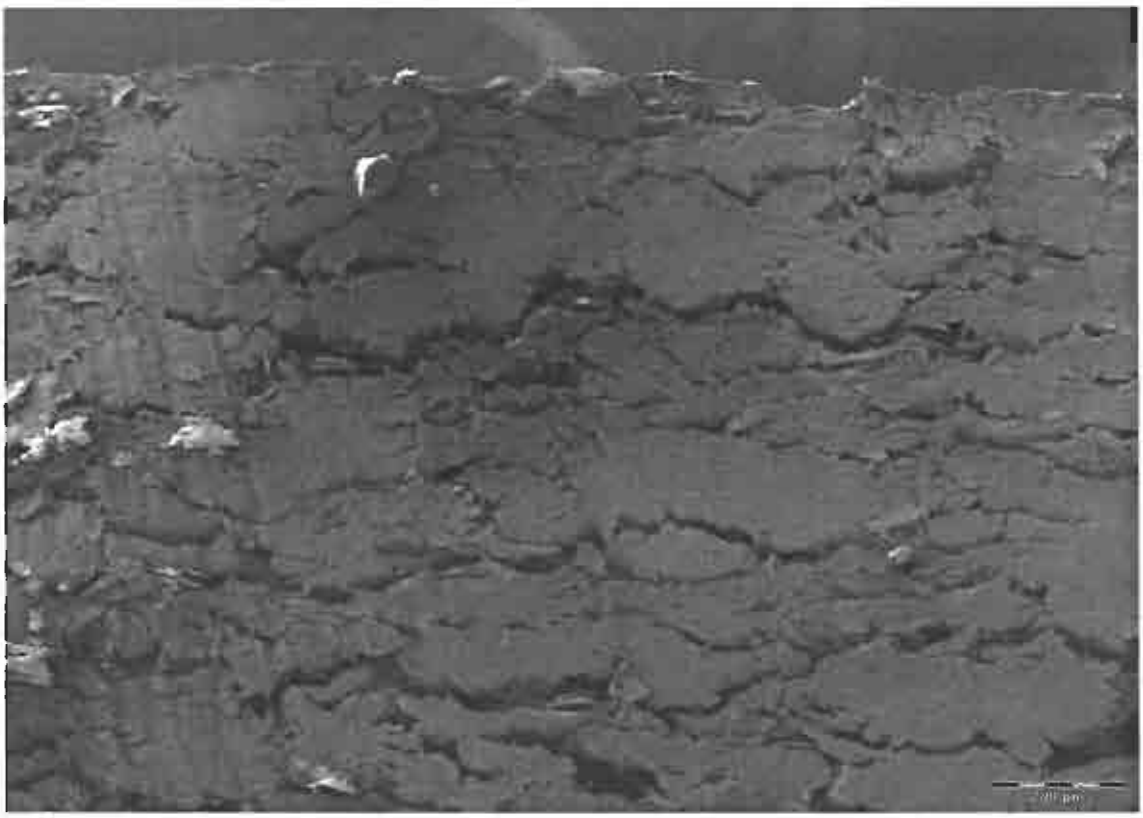

Figure 4.16: *SEM picture of a chitosan tablet compressed at a setting of 44.*

Results of the above experiments show that an increase in the compression force exerted by the IR press resulted in tablets with a much longer disintegration time as well as an increase in the tensile strengths of the tablets. From the wettability study however it was clear that lower compression forces resulted in tablets that are extremely porous and will absorb more water at a faster rate, without swelling more, than the tablets compressed at higher compression forces. This data suggested that the chitosan tablets can only be compressed at high compression forces and that the tablets compressed at lower compression forces were still compacted powder. These high compression forces needed to compress the powder are difficult if not impossible to obtain using a standard eccentric tablet press in which the force exerted on the powder is achieved by adjusting the distance between the upper and lower punches and not hydraulically as in the IR press.

The aim of the study was the formulation of chitosan minitablets since the use of multiparticulate systems such as minitablets offers several advantages over conventional single-unit matrix formulations. These include less risk of dose-dumping, less inter- and intra-subject variability and a higher degree of dispersion in

the gastro-intestinal tract thus minimizing irritation associated with high local drug concentrations. These minitablets can, for practical reasons, only be compressed using the eccentric tablet press in which the compression forces necessary to compress the minitablets, is difficult to obtain as explained above.

The problem of obtaining these higher compression forces might be solved by filling more chitosan powder into the die before compression of the powder. This would mean that a greater force can be exerted on the powder since this greater amount of powder will be compressed into the same volume as the smaller amount of powder. As a result, the eccentric tablet press will need to be modified to fill more chitosan powder into the die and therefore generate higher compression forces if acceptable minitablets were to be compressed.

4.8.1 Eccentric press

In the IR press the compression force is exerted hydraulically and controlled by computer software while on an eccentric tablet press, the compression force is exerted manually and controlled by the distance between the upper and lower punches during the compression cycle. Another crucial difference between the two machines is the fact that the amount of powder that can be used to compress the tablets can be varied easily using the IR press. Since each tablet is compressed individually and the powder weighed before compressing, the tablet weight can simply be changed by weighing more or less of the powder before placing it in the die. The die can also be replaced with another in which the diameter of the opening or height of the die itself is different.

In an eccentric press however the amount of powder can only be varied by using different dies in which the diameter varies. The height can not be changed as only dies of a set height will fit into the press. The only means therefore to manipulate the compression force is to change the depth of movement of the upper punch into the die during compression (using the scale provided on the machine, ranging between 0 and 50).

One possible solution to the problem is to fill more powder into the die before compression, by compacting the chitosan powder. To accomplish that, it was decided to modify the standard eccentric tablet press (Manesty Machines, Liverpool, England) that was available. In the proposed modification to the press, the electric motor used to drive the press would be substituted with a stepper motor which could stop and reverse the cycle at any stage. The idea was that the motor must be programmed in such a way that the cycle could be stopped after the upper punch moved into the die for a predetermined distance. This distance should be calculated as a percentage of the total depth of the die (percentage compaction). The cycle must then be reversed so that the die could again be filled with the powder, stopped again and then reversed to the original cycle until the tablet was compressed and ejected.

4.8.2 Modifications to the press

The standard electric motor on the eccentric press was replaced by a powerful stepper motor. The stepper motor differs from the standard motor in the fact that it could be controlled with a computer. This control include stopping the motor at a very precise distance that the motor motioned (step) and also reversing the direction of the motor. Both these controls were essential if the compression cycle was to be reversed at a certain stage. To increase the torque of the motor, a 10:1 gearbox was installed between the stepper motor and the press. The newly installed stepper motor and gearbox is shown in figure 4.17.

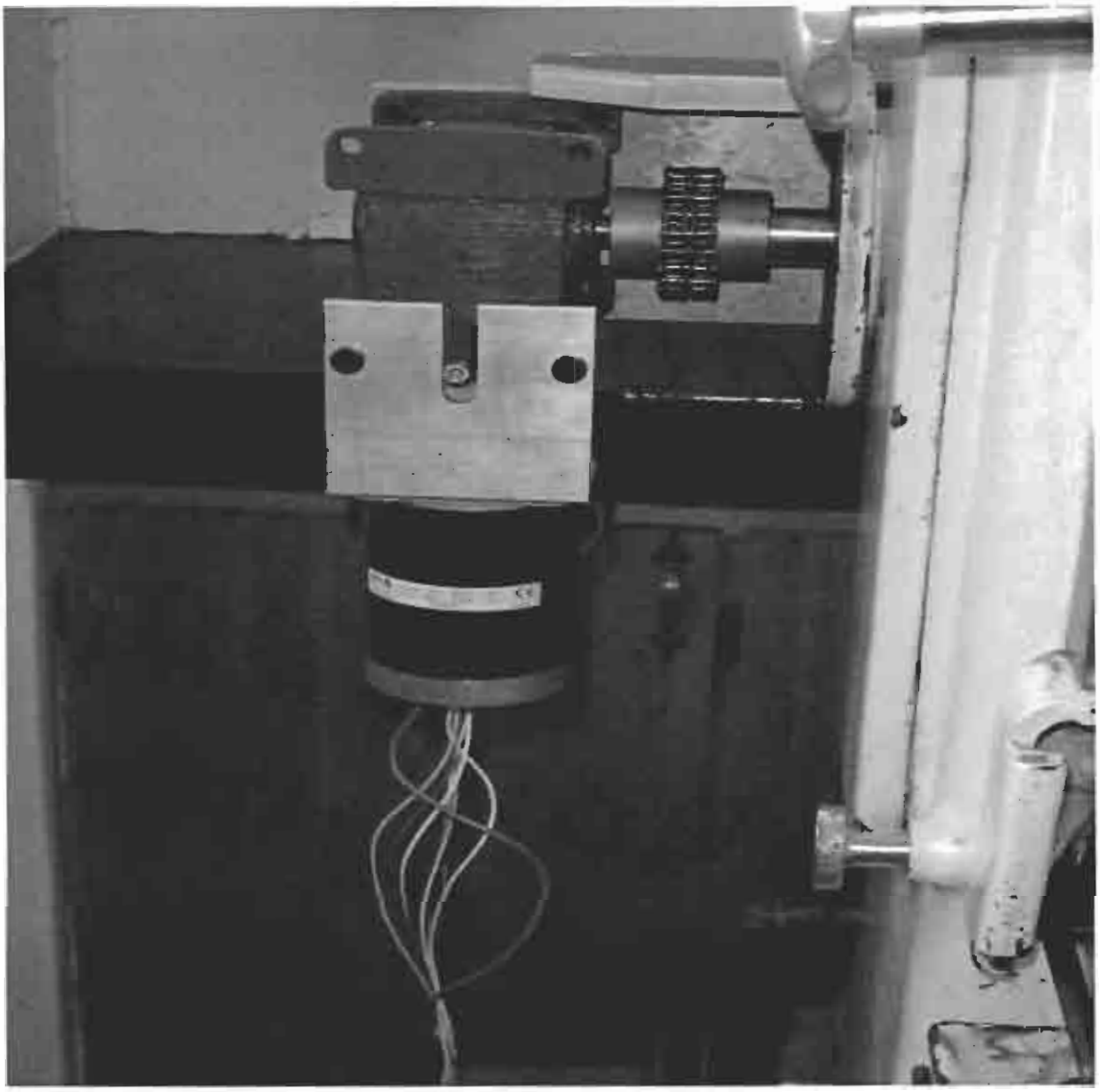

Figure 4.17: *New stepper motor fitted to the eccentric tablet press.*

The newly installed stepper motor is controlled by an embedded controller with a human machine interface for easy operation. This controller was programmed to prompt the user and accept input which was used to set the parameters of the machine. The complete system is shown in figure 4.18.

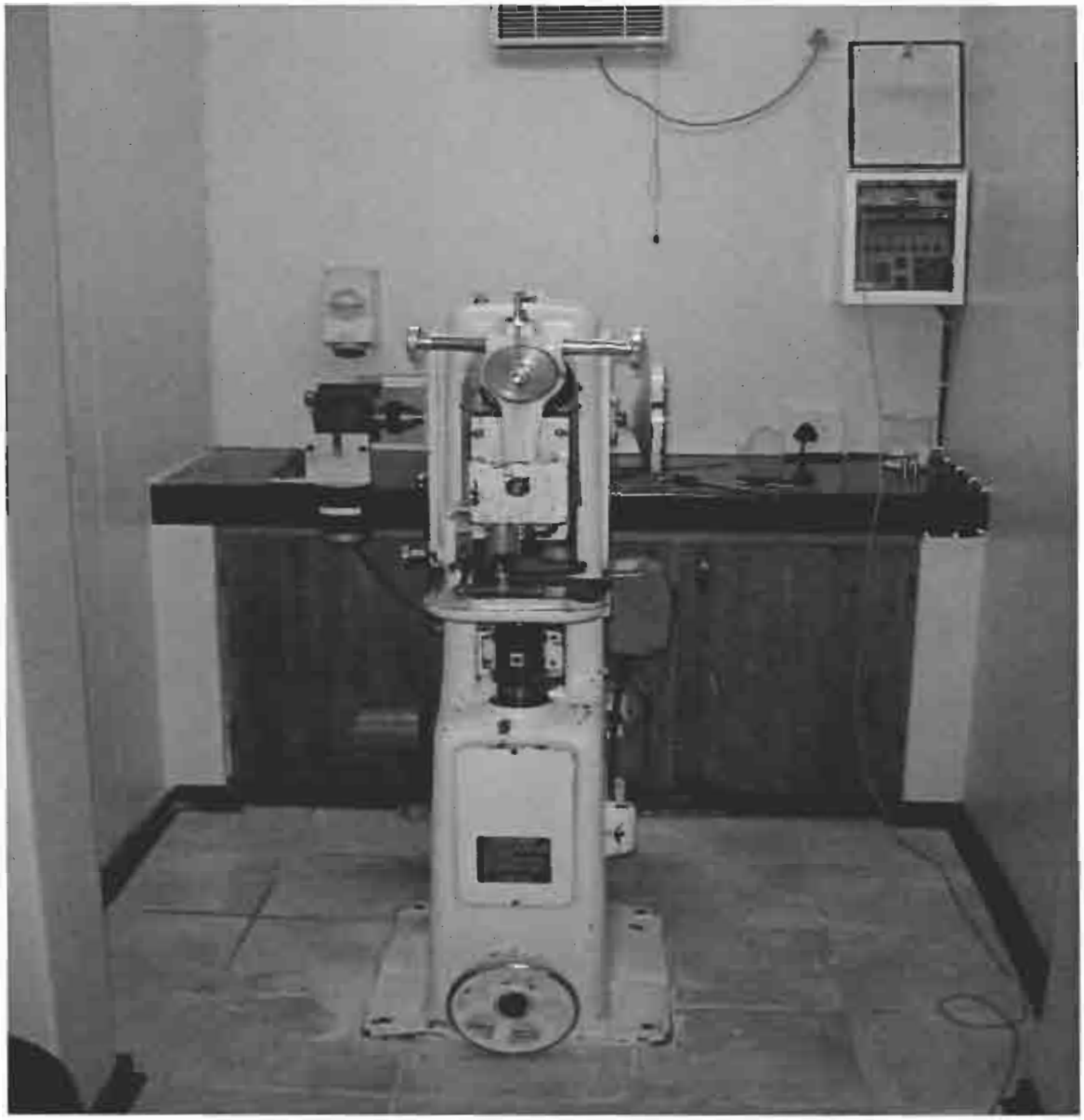

Figure 4.18: *Complete eccentric press and controller.*

The vertical movement of the punch is determined by the internal mechanical design of the press. This relation is not linear and dependent on the crank profile of the press. In order to computerize the press, the first step is to determine the relation between motor revolutions and punch movement. To automate this press, a mathematical model relating the revolutions to the vertical displacement of the punch needed to be derived. This was done by gathering data relating the revolutions of the motor to the vertical displacement of the punch.

4.8.3 Obtaining data

Relating the revolutions of the stepper motor to the vertical displacement of the punch required experimental data. The data was obtained by stepping the stepper motor and measuring the resulting displacement of the punch by using a vernier calliper. This process was repeated for different stroke lengths which affects the offset position of the punch. Although not perfect, this method yielded sufficiently accurate results. The next step was to process the measured data and fit a suitable mathematical model to the data.

4.8.4 Processing of data

The measured data for various stroke lengths (SL) is shown in figure 4.19.

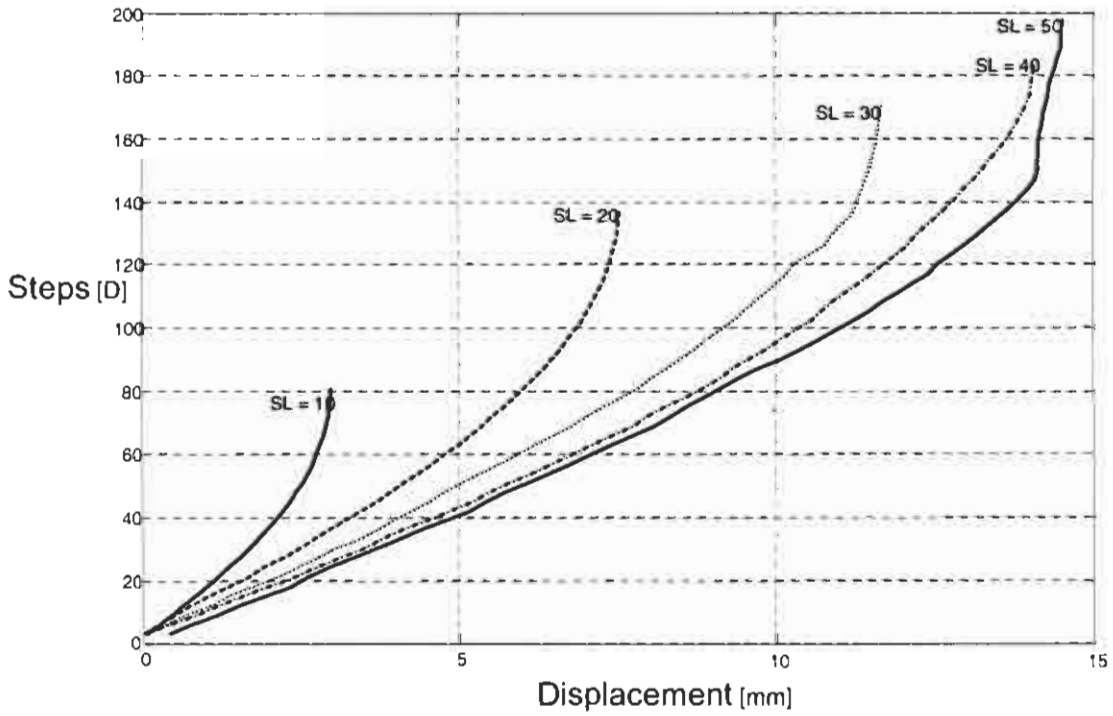

Figure 4.19: *Steps vs. displacement.*

By converting the independent variable to percentage compression instead of displacement, a direct relationship could be found between the required compression and the amount of steps the motor should make. Figure 4.20 shows the graph for the processed data.

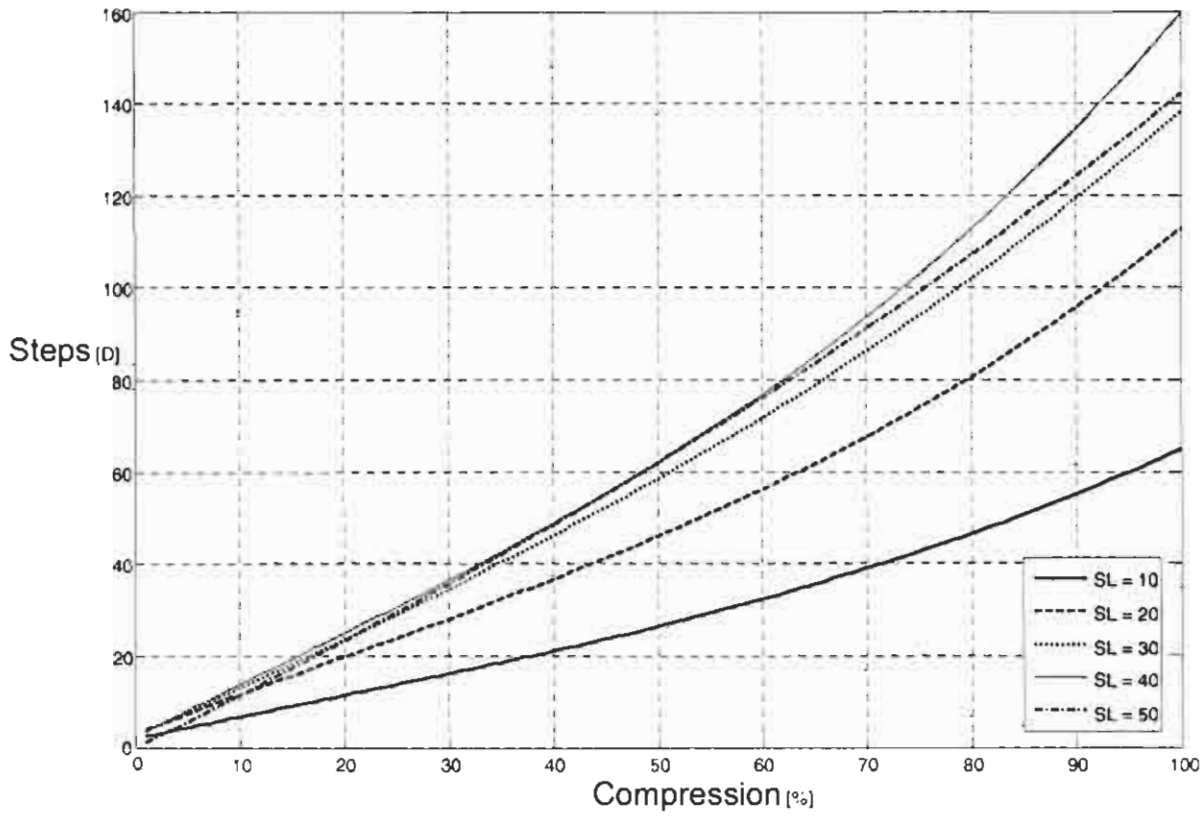

Figure 4.20: *Stepper motor steps vs. compression.*

4.8.5 Fitting of the model

The curves as plotted in figure 4.20 should be used by the controller to calculate the amount of steps to be taken by the stepper motor. This was done by fitting polynomial expressions to the data and programming these expressions into the controller. Figure 4.21 shows the measured data, fitted model, derived polynomial and discrepancy between data and model for a stroke length with a setting of 10. The models fit the data very good and could be used with confidence.

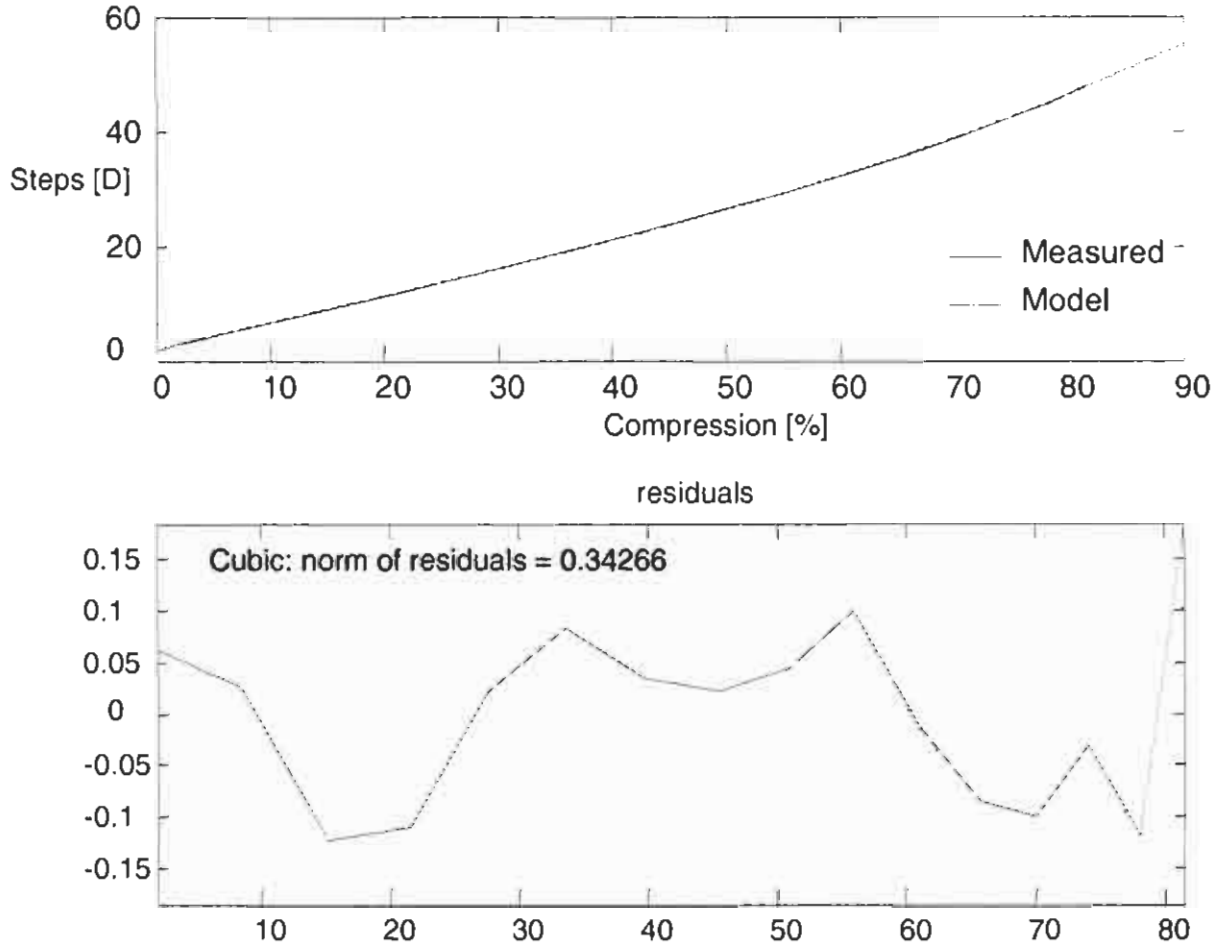

Figure 4.21: *Fitted model for stroke length = 10.*

4.8.6 Models obtained

The third order mathematical equations presented are the result of the data gathering and models the mechanical movement within the system. The equations relate the percentage compression (C) to the amount of steps (D) to be taken for various stroke lengths. Stroke length values between these are obtained by interpolation. Any mechanical changes to the system should be accompanied by the described procedure to derive new mathematical models of the press.

$$D_{SL-10} = 0.000027C^3 - 0.0012C^2 + 0.49C + 2$$

$$D_{SL-20} = 0.000052C^3 - 0.0003C^2 + 0.88C + 2$$

$$D_{SL-30} = 0.000032C^3 - 0.000047C^2 + C + 2.4$$

$$D_{SL\ 40} = 0.00007C^3 - 0.00029C^2 + 1.2C + 2.3$$

$$D_{SL\ 50} = 0.000017C^3 - 0.001C^2 + 1.1C - 0.17$$

4.8.7 Summary

The computerised system has several advantages when compared to the original system. Firstly, the press can now be controlled via a human-machine interface which makes the setup procedure a lot easier. After start-up, a password must be entered to use the press. This eliminates the possibility of unauthorised access and subsequent damage to the press. A correct password grants access and the user is prompted to enter certain parameters required for setup (such as motor speed, percentage compaction and the quantity of tablets that must be compressed). All inputs are validated to ensure safe operation and to prevent damage to the equipment. Should the user enter parameters which could damage the punch, the controller will give an appropriate warning message.

Another advantage is that the system has more flexibility. The old eccentric press was only capable of a single fill whereas this system can perform multiple fills. This is ideal for experimentation with different compressions and fills as required.

Software can be modified to set almost any parameter of the system. Future requirements can be met by appropriate modification of the existing software. Most importantly, this whole conversion was done at a fraction of the price of a new commercial tablet press with the same capabilities.

Figures 4.22 and 4.23 show a cross section of the minitablets compressed at a setting of 36 with compaction percentages of 20% and 40% respectively using the double fill modification. It can be seen that the tablets are much less porous than those compressed without any compaction of the powder.

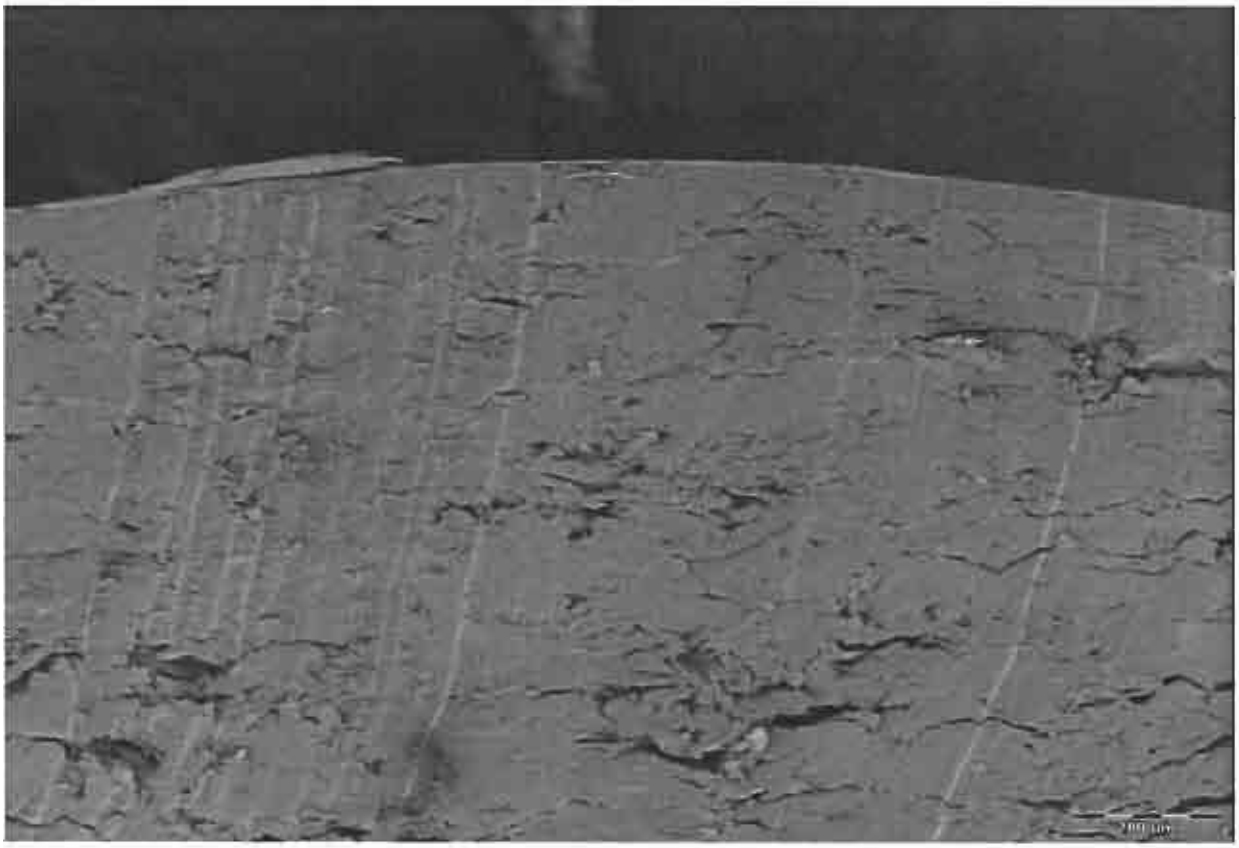

Figure 4.22: SEM pictures of *chitosan tablets compressed at a setting of 36 with a compaction percentage of 20%.*

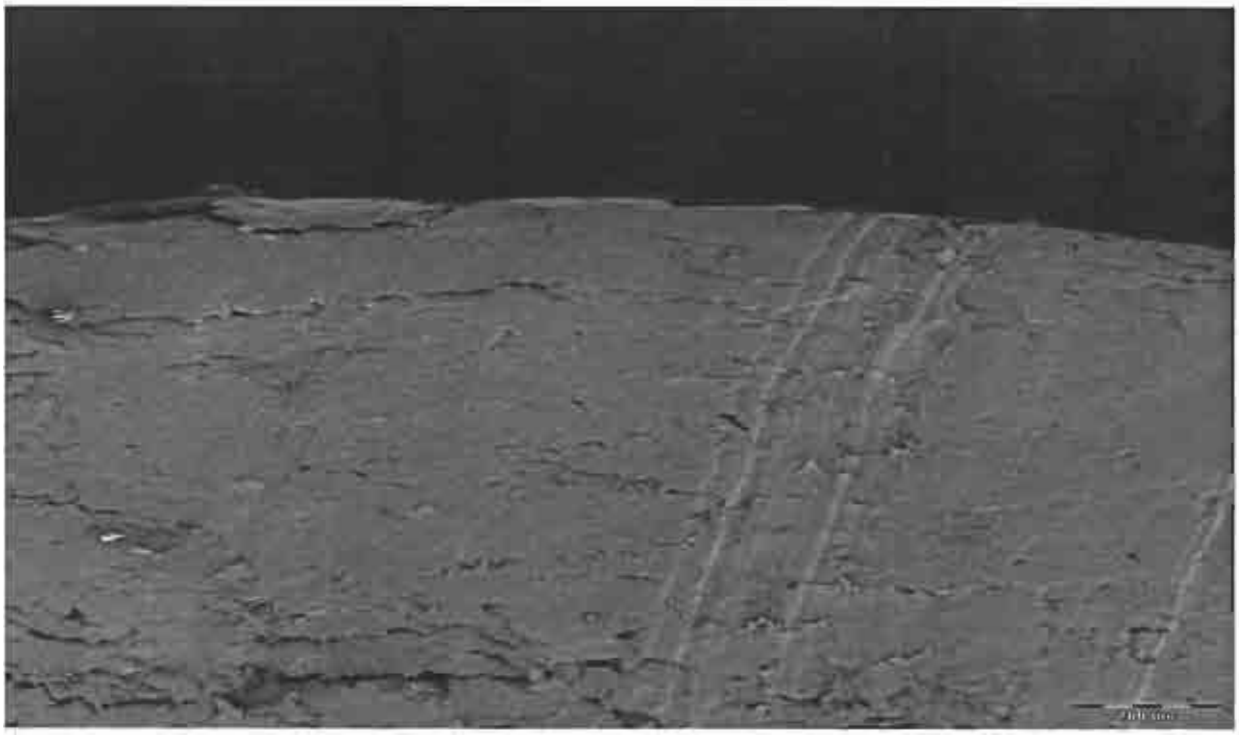

Figure 4.23: SEM pictures of *chitosan tablets compressed at a setting of 36 with a compaction percentage of 40%.*

4.9 THE EFFECT OF PUNCH DEPTH ON THE TABLET PROPERTIES

4.9.1 Introduction

As was seen from figures 4.15 and 4.16, the minitablets compressed during the normal compression cycle was more porous than those subjected to double compression (figures 4.22 and 4.23). To establish if there is a difference in the tablet characteristics between these minitablets, an investigation was done to determine the effect of the punch depth on the tablet properties.

4.9.2 Method

Tablets were compressed at different punch depths (machine settings 30, 32, 34, 36 and 38) using chitosan powder and talc (0.5% w/w). The weight, diameter, thickness and crushing strength were determined and the tensile strength calculated. The disintegration of the minitablets in water were also investigated.

4.9.3 Results

The results are given in table 4.2 and the measurement values given in table A.39 – A.43 in annexure A.

Table 4.2: *Tensile strength and thickness of chitosan minitablets as a function of the punch depth.*

Punch depth given as the machine setting	Average tensile strength (N/mm^2)	Average crushing strength (N)	Average thickness (mm)
30	2.98 ± 0.32	33.72 ± 3.93	1.83 ± 0.02
32	5.40 ± 0.25	54.68 ± 2.49	1.63 ± 0.04
34	5.65 ± 0.10	56.03 ± 1.75	1.60 ± 0.03
36	5.67 ± 0.27	57.73 ± 3.56	1.63 ± 0.03
38	5.79 ± 0.47	57.95 ± 5.56	1.60 ± 0.04

The tensile strength increased with an increase in the punch depth. The increase was the biggest between the lower settings of 30 to 32 while the increase was much less at the higher machine settings. At a machine setting of 30, the low tensile strength (2.98 N/mm^2) suggested that minitablets were compressed that was very porous and binding between the chitosan particles did not take place or was weak. At the higher machine settings (greater punch depth) the tensile strength varied between 5.40 and 5.79 N/mm^2 at settings of 32 and 38 respectively. The ANOVA test has shown that there was a significant difference in the tensile strengths of the minitablets compressed at a setting of 30 compared to those compressed at the higher settings. There were, however, no significant differences between the tensile strengths of the minitablets compressed at the higher settings. It should be noted that the tablet thickness decreased if the punch depth increased from a setting of 30 to a setting of 32. An increase to settings higher than 32 did not lead to a difference in the tablet thickness. This is probably because the compression force is not enough to compress the powder in such a way that strong binding takes place between the chitosan particles and as a result the tablets relaxed to the measured thickness.

The disintegration test showed that the tablets compressed at a setting of 30 disintegrate within 1 – 2 minutes (scale 1 on the arbitrary scale) and it can be concluded that the compression force used to compress the tablets was too low for effective binding between the particles to take place. The low crushing strength of 33.72 N of the tablets confirms this result. The tablets compressed at the other settings however were wetted within 2 – 5 minutes but failed to disintegrate (scale 4 and 5 on the arbitrary scale) and there appeared to be no difference in the disintegration behaviour. These results, as well as the fact that there was not a big difference between the crushing strength of these tablets (of 54.68 to 57.95 N) nor in the thickness (1.60mm to 1.63 mm), suggests that varying the punch depth did not have an influence on the tablet properties probably because of poor binding between the chitosan particles.

4.10 THE EFFECT OF COMPACTION ON THE TABLET PROPERTIES

4.10.1 Introduction

The compaction of the powder before compressing the tablet, resulted in tablets that are less porous than the tablets where the powder were not compacted before compressing the tablets (see figures 4.22 and 4.23 as compared to 4.15 and 4.16). To establish the effect of the percentage compaction on the tablet characteristics, minitablets were compressed at different compaction percentages and then evaluated.

4.10.2 Method

Tablets were compressed at different compaction percentages at a punch depth setting of 36, using the chitosan powder containing isoniazide (4% w/w) and talc (0.5% w/w). The weight, diameter, thickness and crushing strength were determined and the tensile strength calculated. The disintegration was also done in water as the disintegration medium.

4.10.3 Results

The results of the tensile strengths are given in table 4.3 and those of the tablet thickness in figure 4.24. The measurement values are given in table A.45 – A.48 in annexure A.

Table 4.3: *Tensile strength of chitosan minitablets as a function of the compaction percentage.*

Compaction (%)	Average tensile strength (N/mm^2)	Average crushing strength (N)	Average tablet thickness (mm)
0	5.79 ± 0.13	57.45 ± 1.80	1.59 ± 0.02
20	6.00 ± 0.34	68.47 ± 3.82	1.83 ± 0.05
40	6.35 ± 0.48	82.95 ± 6.40	2.09 ± 0.02
60	6.89 ± 0.32	104.73 ± 4.97	2.43 ± 2.43

It was found that the tensile strength increased with an increase in the percentage compaction. The ANOVA test shows a significant difference between the tensile strengths except between 0% and 20% compaction and between 20% and 40% compaction. The effect can be seen more clearly in the increase in the crushing strength from 57.45 N to 104.73 N. This increase in the tensile strength (and crushing strength) is probably due to the fact that there is a bigger volume of powder present as a result from the compaction and refill of the die, which resulted in better compaction of the powder.

This volume increase also lead to an increase in the tablet thickness as can be seen in figure 4.24 although the tablets were compressed at the same punch depth setting of 36. This can only be due to the tablets that relaxed after compaction.

The disintegration showed that the tablets did not disintegrate but rather broke in 2 pieces or just split (4 – 5 on the arbitrary scale). One of the six tablets compressed at a compaction percentage of 40 and two compressed at a 60% compaction stayed intact (6 on the arbitrary scale). The tablets were still wetted easily within minutes which lead to the conclusion that the tablets were still porous but that binding at the higher compaction percentages be appears to be stronger.

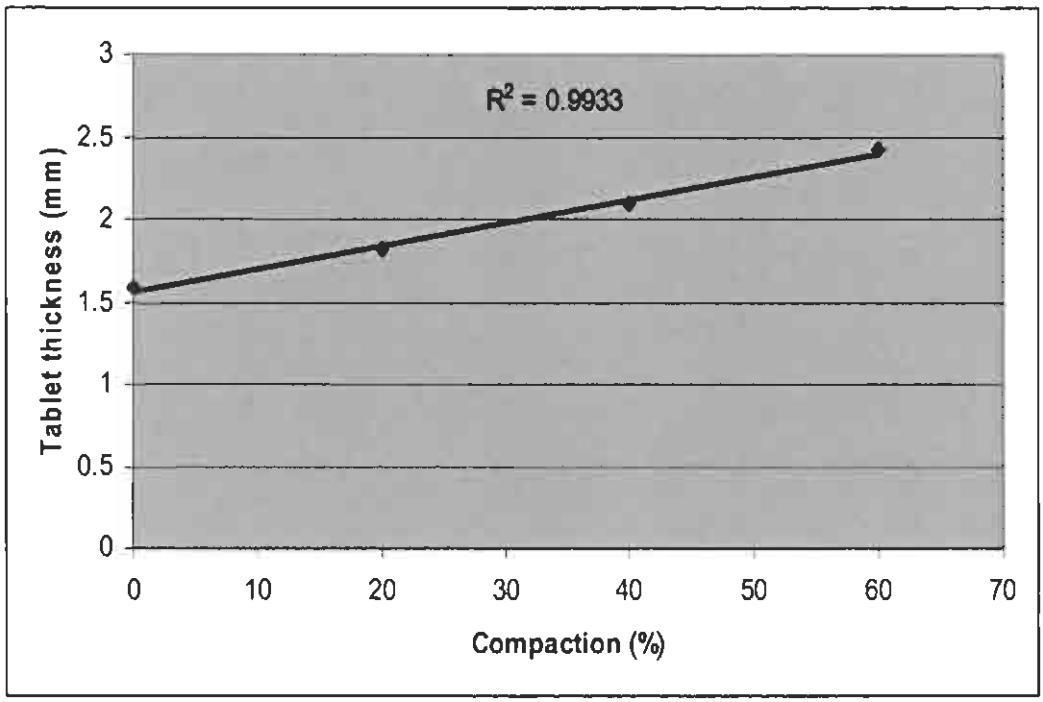

Figure 4.24: *Tablet thickness of chitosan minitablets compressed at different percentages compaction.*

4.11 SUMMARY

In the study, different factors that might influence the compressibility of chitosan were investigated. Compression forces of between 15 and 20 bar resulted in tablets with acceptable physical characteristics. Tablets showed an increase in tensile strength as well as in disintegration time with an increase in compression force. The moisture content of the powder not only influences the flow properties but also the tensile strength. An increase in moisture content resulted in tablets with higher tensile strength. The particle size of the chitosan powder also influences the tensile strength. Tablets compressed with the particle size fraction > 212 µm resulted in tablets with a higher tensile strength. This was however dependant on the weight of the tablets. A decrease in weight of the powder used also resulted in tablets with a higher tensile strength. The inclusion of talc in the formulation did not influence the tensile strength. In view of this findings, it was clear that these factors will need to be kept constant if differences in the drug delivery from the minitablets is to be investigated.

The punch depth did not have an influence on the tablet properties except at a punch depth of 30 were binding between the particles were very weak resulting in tablets with a crushing strength of 33.72 N. Increasing the punch depth up to a setting of 38 resulted in tablets with comparable thickness (1.60 –1.63 mm), crushing strengths (between 54.68 and 57.95), wettability (between 2 and 5 minutes) and disintegration (arbitrary scale 4 -5).

Varying the percentage compaction, however lead to tablets with much higher crushing strengths (104.73 N at a compaction of 60%) and an increase in tablet thickness from 1.59 mm to 2.43 mm because of the higher compression force generated as a result of the bigger volume of powder that was compressed.

The disintegration however was similar to those of the tablets compressed at different punch depths. This suggests that although the tensile strength and crushing strength of the tablets were higher, the porosity was also still high.

Further experiments will investigate the effect of the process variables, as discussed above, as well as the influence of formulation variables on drug release from the minitablets.

CHAPTER 5
DRUG RELEASE FROM CHITOSAN MINITABLETS

5.1 INTRODUCTION

It is important that the drug release from matrix systems intended for colonic drug delivery can be controlled and manipulated. Because of the distal location of the colon in the GIT, a colon-specific drug delivery system should prevent drug release in the stomach and small intestine, and effect an abrupt onset of drug release upon entry into the colon (Yang *et al.*, 2002:1). Several physiological changes occur along the GIT but only the presence of specific bacterial populations in the colon and the change in pH have been extensively explored as triggering mechanisms for drug release from colon-specific dosage forms. The ideal colon-specific drug delivery system would therefore prevent drug release in the stomach and small intestine, and start releasing the drug at controlled rates upon entry into the colon (Zambito & Di Colo, 2003:274).

The tablet characteristics of the chitosan minitablets that were compressed in previous experiments could be changed by process variables such as the punch depth and compaction of the powder which could alter the compression force exerted on the chitosan powder during compression. These tablet characteristics included the tensile strength as well as the wettability and disintegration of the tablets. The minitablets did not disintegrate but rather only swelled in the disintegration medium to form a matrix. It is important that the drug release from these matrices can be controlled to ensure that there is no dose dumping and that the drug is delivered to the colon at a suitable rate.

As a result, investigations was done to see if the drug release from the minitablets could be manipulated by process- and formulation variables. Initially the release of isoniazide, as an example of a water-soluble drug, from minitablets that were compressed at different process variables that effected the compression force (punch depth and compaction of the powder) were investigated. Secondly the formulation of the minitablets were changed to include either citric acid or pectin and the release of

the isoniazide from the tablets determined. The minitablets were then coated to establish if the release of the isoniazide could be delayed and the tablet protected from the acidic conditions of the stomach by coating the tablet with an enteric polymer (Eudragit S®).

5.2 METHODS

5.2.1 Preparation of the minitablets

Minitablets with a diameter of 4 mm were compressed using the modified eccentric press at different process variables. The minitablets contained isoniazide (4 % w/w) as an example of a water soluble drug and talc (0.5% w/w) as a glidant. The powders were weighed and mixed in a Turbula mixer for 5 minutes before compressing the tablets. The tablets were then left for 24 hours in a sealed container before measuring the drug release.

5.2.2 Dissolution studies

5.2.2.1 Apparatus

The dissolution studies were performed in a six-station Erweka DT6R dissolution apparatus (Erweka Apparatbau GmBH, Hausenstamm, Germany) fitted with a thermostat that kept the dissolution medium at 37 °C. The apparatus was adapted to hold dissolution beakers with a volume of 200 ml and using paddles designed to fit into the smaller beakers.

5.2.2.2 Method

The dissolution vessels were filled with the required dissolution medium and left to equilibrate to the set temperature. The paddles were adjusted to a depth of 25 mm from the bottom of the beakers and the lids were secured on each beaker. The paddles were rotated at 50 r.p.m with a variable speed synchronous motor. The minitablets were then introduced into the beakers and the time set at t = 0. Between

6 and 8 minitablets were used in each vessel resulting in a maximum isoniazide concentration that was within the range of the standard curve concentrations. Samples were taken at the same height through a Sartorius membrane filter fitted with 25 mm prefilters at t = 2, 5, 10, 15, 20, 30, 45, 60 and 120 minutes and transferred into 10 ml glass tubes. The volume of the samples were 5 ml and replaced with fresh dissolution medium at the same temperature. After the final sample had been taken, the remaining dissolution medium was placed in a soniccator for 30 minutes to allow the remaining drug to dissolve. A sample was taken from the soniccated dissolution medium as described above. The samples were subsequently analyzed. The dissolution tests were done in triplicate and the absorbances also measured in triplicate.

5.2.3 Analysis

A standard curve was drawn up in each of the dissolution media and the validity checked at each analysis with two samples of known concentration. A mother solution with a concentration of 500 μg/ml were prepared by dissolving 50 mg of isoniazide in the dissolution medium and then made up to 100 ml with the solution. Standard solutions were then prepared by diluting between 1 ml and 8 ml of the mother solution to 100 ml with the dissolution medium yielding solutions with concentrations ranging between 5 μg/ml and 40 μg/ml. The UV absorbances of the standard solutions were then measured in triplicate at 263 nm against the dissolution medium as a blank, using a Unicam-spectrophotometer (Helios α, Unicam Ltd, Cambridge, UK). The absorbances were plotted against the concentration and linear regression were then used to fit the best straight line through the data points. The standard curves exhibited a linear relationship in the concentration range used with correlation coefficients (r^2) > 0.999.

5.2.4 Statistical comparison of dissolution profiles

5.2.4.1 Mean dissolution time

The mean dissolution time (MDT) is defined as the mean time for the drug to dissolve under *in vitro* dissolution conditions (Ritger & Peppas, 1987:31). The MDT can be calculated from the profile of the cumulative mass of drug dissolved using the equation:

$$MDT = \frac{\sum_{i=1}^{n} t_{mid} \Delta X_d}{\sum_{i=1}^{n} \Delta X_d}$$

Where i is the sample number, n is the total number of sample times, t_{mid} is the time at the midpoint between sample times and ΔX_d is the additional mass of drug dissolved between i and $i - 1$.

The mean dissolution time (MDT) for each dissolution profile were calculated and compared using a 1 – way ANOVA test (Analyse-it, Microsoft). A contrast test was also done between all the combinations of data from all the dissolution profiles to establish if there was a significant difference between the profiles.

5.2.4.2 Similarity factor

The similarity factor (f_2) is used to compare the dissolution profile of a formulation against that of a reference formulation (Moore & Flanner, 1996:64). The f_2 value is 100 when the test and reference profiles are identical and approaches zero as the dissimilarity increases. An f_2 value of between 50 and 100 indicates similarity between two profiles. The similarity of each dissolution profile was calculated against all the other dissolution profiles from the specific experiment and the f_2 values were quoted.

5.3 EXPERIMENTAL

5.3.1 The influence of process variables on drug release

5.3.1.1 Introduction

The minitablets did not disintegrate but only swelled in the disintegration medium to form a matrix. The characteristics of the minitablets could be changed by changing the process variables. It is important that the drug release from these matrices can be controlled to ensure that there is no dose dumping and that the drug is delivered to the colon at a suitable rate. As a result, investigations was done to see if the drug release from the minitablets could be manipulated by process variables.

5.3.1.2 Method

Minitablets were compressed as described in section 5.2.1 at different punch depths (machine settings 30, 32, 34, 36 and 38) and percentages compaction (20%, 40% and 60%) at the same punch depth setting of 34, using the chitosan powder containing isoniazide (4% w/w) and talc (0.5% w/w). The dissolution of the isoniazide was determined as described in section 5.2.2 using 200 ml water as a dissolution medium.

5.3.1.3 Results

The results of the dissolution tests are given in figures 5.1 and 5.2 and in table A.44 and A.49 in annexure A. The MDT values are given in table 5.1 and 5.3 while the similarity factors are given in tables 5.2 and 5.4. The MDT values for each dissolution profile are given in tables A.58 and A.59 in annexure A.

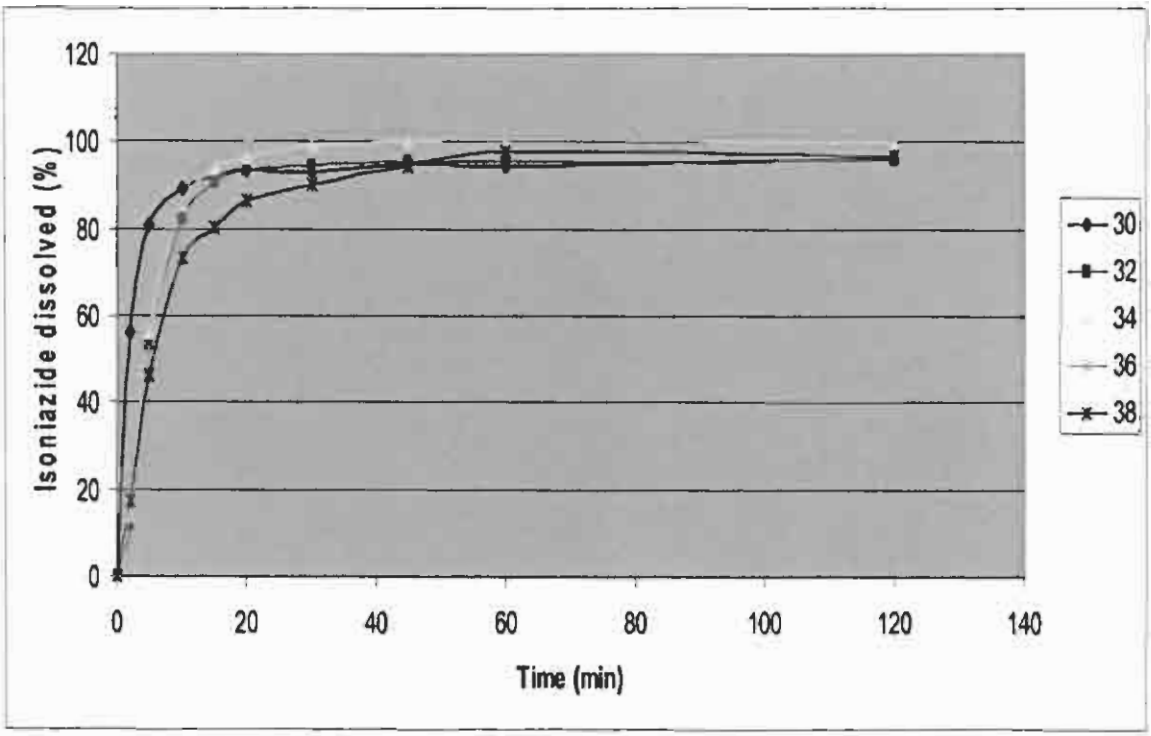

Figure 5.1: *Dissolution profile of isoniazide from chitosan minitablets compressed at different punch depths.*

Table 5.1: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the punch depth.*

Punch depth setting	*Mean dissolution time (MDT)*
30	*4.92 ± 1.11*
32	*6.46 ± 0.64*
34	*6.31 ± 1.40*
36	*7.24 ± 0.06*
38	*9.05 ± 0.37*

Table 5.2: *The similarity factor (f_2) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the punch depth.*

	Setting 30	Setting 32	Setting 34	Setting 36
Setting 32	42.23 ± 1.85	-	-	-
Setting 34	43.37 ± 1.20	72.76 ± 12.50	-	-
Setting 36	41.14 ± 1.72	64.18 ± 1.87	72.77 ± 7.48	-
Setting 38	40.93 ± 1.05	64.22 ± 5.06	57.25 ± 2.41	53.52 ± 2.41

The dissolution from the tablets compressed at a punch depth setting of 30 was faster at the initial stage with 56.07% isoniazide dissolved at 2 minutes and 80.87% at 5 minutes compared to between 11.12% and 17.44% at 2 minutes and between 46.19% and 56.05% at 5 minutes for the other tablets.

The MDT values increased from 4.92 to 9.05 with increasing punch depth. The ANOVA variance test is given in table 1 in annexure C and showed that there was only a significant difference between the profiles at settings 30, 32 and 34 compared to the profile at setting 38.

The similarity factors showed a bigger similarity between the dissolution profiles at the higher settings (between 53.52 and 72.76) while all the profiles showed a smaller similarity to the profile at a setting of 30 (40.93 to 43.37).

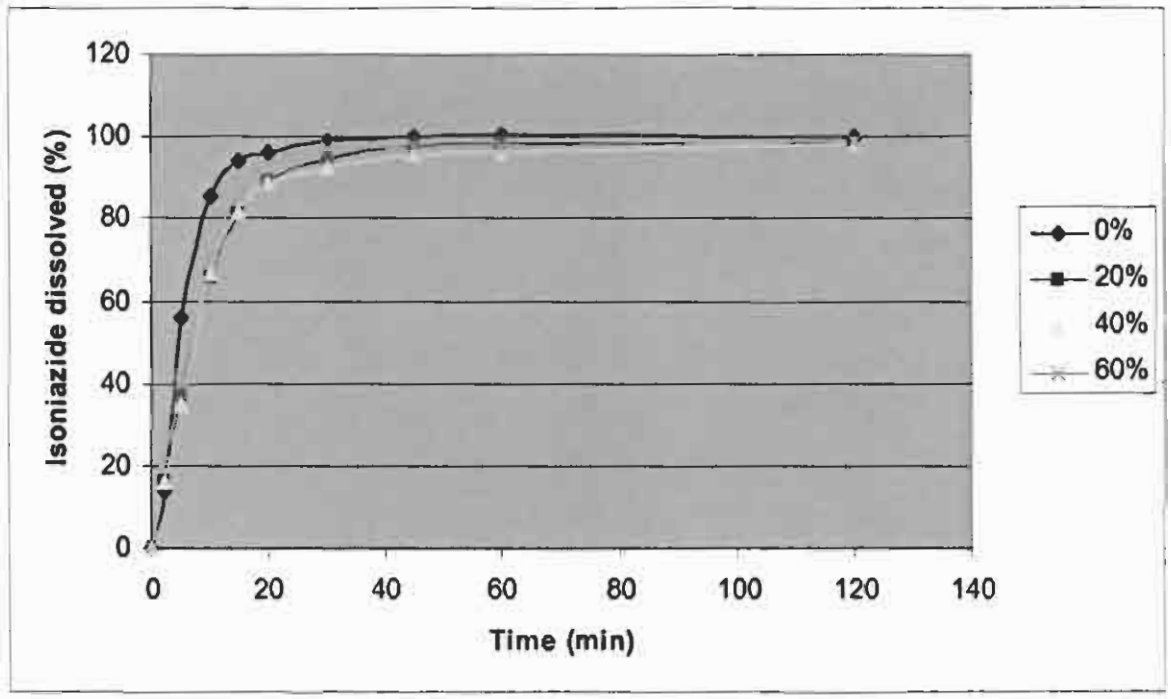

Figure 5.2: *Dissolution profile of isoniazide from chitosan minitablets compressed at different compaction percentages.*

Table 5.3: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the percentage compaction.*

Percentage compaction	*Mean dissolution time (MDT)*
0	*6.31 ± 1.40*
20	*9.47 ± 0.60*
40	*10.76 ± 8.84*
60	*8.84 ± 2.01*

Table 5.4: *The similarity factor (f_2) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the percentage compaction.*

	0 %	20 %	40 %
20 %	52.87 ± 1.43	-	-
40 %	51.95 ± 1.87	79.28 ± 9.26	-
60 %	58.29 ± 1.44	73.83 ± 4.33	74.87 ± 15.20

The MDT values varied between 6.31 to 10.76 and the ANOVA variance test (given in table 2 in annexure C) showed that there was no significant difference between the profiles.

The similarity factors showed a bigger similarity between the dissolution profiles at the 20%, 40% and 60% compactions (between 73.83 and 79.28) while these profiles showed a smaller similarity to the profile at 0% compaction (56.18 to 58.47).

The mean dissolution time (MDT) of the profiles could be increased from 4.92 to 9.05 by increasing the punch depth setting. At a punch depth setting of 34 the MDT of the profiles could be increased from 6.31 to 10.76 by an increase in the compaction. The profiles at the higher punch depth settings and percentage compaction were very similar but showed a smaller similarity only to the profile of the tablets compressed at a punch depth setting of 30 and 0% compaction.

Table 5.5 shows the effect of the process variables on the dissolution rate of isoniazide from the minitablets. The percentage of isoniazide dissolved after 5 minutes are given at different punch depth settings and percentage compaction. It can be seen that the percentage isoniazide dissolved decrease with an increase in the punch depth and percentage compaction.

Table 5.5: *Percentage isoniazide dissolved at t = 5 minutes at different punch depth settings and percentage compaction.*

	Percentage compaction			
Punch depth setting	0%	20%	40%	60%
30	80.87			
32	53.47			
34	56.05	37.30	35.22	40.45
36	52.46			
38	46.20			

After approximately 20 minutes however, the percentage isoniazide dissolved increased to over 90% at all the different punch depth settings and percentages compaction.

This suggested that although the tablets becomes less porous with an increase in the punch depth and percentage compaction, as seen figure 4.22 and 4.23, the porosity was such that water still penetrated the tablets easily and as a result there was a rapid dissolution of the isoniazide. When investigations were done into the disintegration of the minitablets, it was found that the tablets were wetted (and swelled to an extent that the tablets actually split in two or more separate parts) by the water in approximately 2 – 4 minutes regardless of change in the process variables.

This also suggested that to compress chitosan into a matrix from which the drug would dissolve over a longer period of time, a very high compression force would be needed to ensure that the water penetrates into the matrix at a slower rate. Other options to achieve this goal might be the inclusion of excipients such as an organic

acid (which would lower the pH in the tablet, allowing the chitosan to form a gel) or pectin (which would form an insoluble complex with the chitosan) into the formulation.

5.3.2 The influence of formulation variables on drug release

5.3.2.1 Introduction

Following the investigation of the dissolution of isoniazide from the chitosan minitablets, is was concluded that the dissolution may not be sufficiently delayed by the process variables. It was therefore decided to investigate the influence of the addition of an organic acid to the formulation. A soluble organic acid could regulate the pH in the matrix to a pH where the chitosan will be soluble and form a gel. The gellation of chitosan in the matrix may prolong the release of the isoniazide out of the tablet, as it will need to diffuse through the gel layer that is formed. Organic acids have been used in pharmaceutical dosage forms to manipulate the dissolution rate of drugs. Narisawa *et al.* (1997:85) used organic acids in Eudragit ® coated tablets to increase the osmotic pressure in the formulations. It was concluded that rapid drug release after lengthy lag times could be achieved with such formulations. Ishibashi *et al.* (1998:31) used organic acids and a acid soluble polymer in a enteric coated capsule to create a dosage form for colonic delivery which is pH dependant (by means of the enteric coating) and time dependant (because of the decrease in the pH as the organic acid dissolves). Nykänen *et al.* (1999:251; 2001:155) produced minitablets for colonic drug delivery containing an enteric polymer that is insoluble at a low pH and then added organic acids (citric acid, succinic acid, tartaric acid) to lower the pH as it dissolved. The dissolution rate of the drug (ibuprofen) decreased and was dependant on the organic acid used as well as the concentration of the organic acid incorporated.

Pectin is a polygalacturonic acid and the chain molecule is negatively charged. The gellation of pectin makes it possible to reduce the penetration of water into the dosage form and hence the dissolution of the drug incorporated into it. Furthermore, the carboxylic groups of the pectin and the amino groups of the chitosan interact and reduce the release of the drug contained in the dosage form (Vandamme *et al.*,

2002:223). The potential of pectin as a carrier for colonic drug delivery has been demonstrated by Ashford *et al.* (1994:225). The use of high-methoxy pectin or cross-linking with calcium have been investigated. Interpolymer complexes with chitosan has also been investigated by Meshali and Gabr (1993:177). The complexes of chitosan and pectin is mainly used as coating or films which are less soluble in the upper GIT and is degraded by the colonic microflora (Fernández-Hervás & Fell, 1998:115).

An investigation was done to establish if the inclusion of citric acid or pectin (2%, 4% and 8%) into the chitosan minitablets have any effect on the dissolution rate of isoniazide from the tablets.

5.3.2.2 Method

Minitablets were compressed as described in section 5.2.1 at a constant punch depths (machine settings 34) and a percentage compaction of 40% using the formulations in table 5.6. The dissolution of the isoniazide was determined as described in section 5.2.2 using 200 ml phosphate buffer (pH 7.2) as a dissolution medium.

Table 5.6: *Tablet formulations containing citric acid or pectin.*

	Percentage w/w of excipients			
Chitosan	95.5	93.5	91.5	89.5
Isoniazide	4.0	4.0	4.0	4.0
Talc	0.5	0.5	0.5	0.5
Citric acid or pectin	0	2.0	4.0	6.0

5.3.2.3 Results

The results of the dissolution tests is given in figure 5.3 and figure 5.4. The percentages isoniazide dissolved are given in table A.55 and A.56 in annexure A while the MDT values and the similarity factors for the profiles are given in tables 5.7

to 5.10. The MDT values for each dissolution profile are given in tables A.60 and A.61 in annexure A.

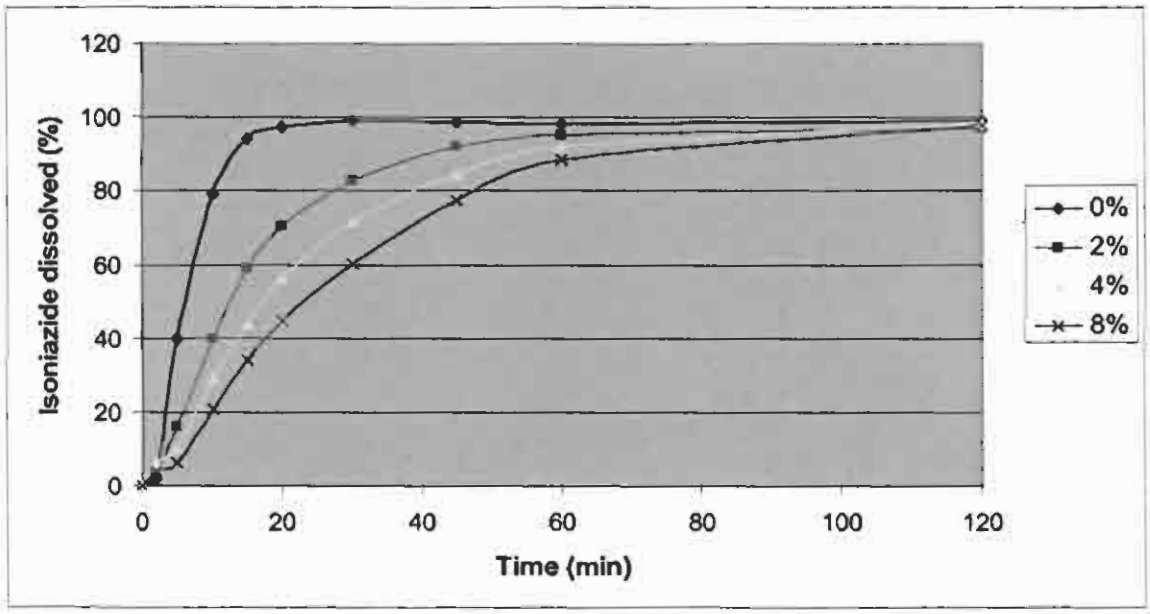

Figure 5.3: *Dissolution profile of isoniazide from chitosan minitablets containing different percentages of citric acid.*

Table 5.7: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of citric acid.*

Percentage citric acid	*Mean dissolution time (MDT)*
0	*7.65 ± 1.18*
2	*16.96 ± 1.61*
4	*24.22 ± 1.78*
8	*29.94 ± 0.86*

Table 5.8: *The similarity factor (f_2) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of citric acid.*

	0%	2%	4%
2%	37.24 ± 0.72	-	-
4%	30.23 ± 1.05	56.02 ± 3.54	-
8%	25.78 ± 0.47	43.48 ± 2.24	60.67 ± 5.79

Because of this lower pH and gellation, the dissolution rate of isoniazide from the tablets were also slower with an increase in the citric acid content. The MDT values increased from 7.65 to 29.94 and the ANOVA variance test (given in table 3 in annexure C) has shown that there was a significant difference between all the profiles. The similarity factors show a big dissimilarity between the dissolution profiles at the 0% citric acid and the other profiles (similarity factor between 25.78 and 37.24).

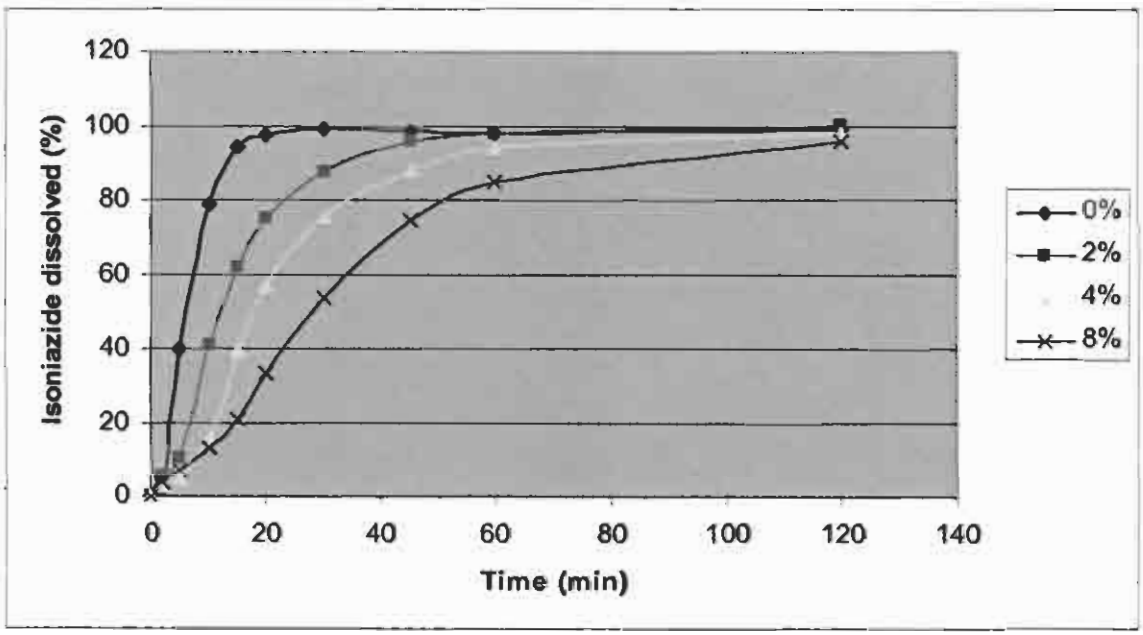

Figure 5.4: *Dissolution profile of isoniazide from chitosan minitablets containing different amounts of pectin.*

Table 5.9: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of pectin.*

Percentage pectin	*Mean dissolution time (MDT)*
0	*7.65 ± 1.18*
2	*16.62 ± 0.86*
4	*23.88 ± 1.20*
8	*33.24 ± 1.71*

Table 5.10: *The similarity factor (f_2) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of pectin.*

	0%	2%	4%
2%	38.39 ± 3.75	-	-
4%	28.04 ± 1.56	47.75 ± 5.01	-
8%	22.41 ± 0.71	34.66 ± 3.75	48.13 ± 4.44

It is clear from the results of the dissolution test that the inclusion of pectin into the minitablets lead to a decrease in the dissolution rate of isoniazide from the tablets.

The MDT values increased from 7.65 to 33.24 and the ANOVA variance test (given in table 4 in annexure C) has shown that there was a significant difference between all the profiles while the similarity factors showed a big dissimilarity between all the dissolution profiles.

Table 5.11 shows the effect of the formulation variables on the dissolution rate of isoniazide from the minitablets. The percentage of isoniazide dissolved after 5 minutes from tablets contain the different percentages of citric acid or pectin are given. It can be seen that the percentage isoniazide dissolved decrease with an increase in the percentage citric acid or pectin.

Table 5.11: *Percentage isoniazide dissolved at t = 5 minutes from tablets containing the different percentages of citric acid or pectin .*

Percentage citric acid or pectin	Formulation variables	
	Citric acid	Pectin
0	39.77	39.77
2	16.16	10.62
4	10.01	4.92
8	6.36	6.91

When the process variables were changed, the percentage isoniazide dissolved at t = 5 minutes decreased from 80.87% (punch depth 30) to 40.45% (percentage

compaction 60%). When the citric acid or pectin was added to the formulation, the percentage isoniazide that was dissolved decreased to 6.36% and 6.91% respectively.

After approximately 20 minutes the percentage isoniazide dissolved increased to 44.64% (citric acid 8%) and 33.42% (pectin 8%) as compared to the more than 90% at all the different punch depth settings and percentages compaction. The percentage isoniazide dissolved reached 90% only after approximately 70 minutes for the citric acid 8% formulation and after approximately 90 minutes for the pectin 8% formulation as compared to 20 minutes when the process variables were changed.

It can therefore be concluded that the inclusion of an organic acid like citric acid will decrease the pH and as a result increase the solubility of the chitosan in the tablet. Because of the gel forming characteristic of chitosan, the tablet formed a gel matrix from which the dissolution rate of the isoniazide was slower than from the minitablets without citric acid. The gel forming characteristics of the pectin and the possible formation of a complex between chitosan and pectin, also delayed the dissolution of the isoniazide from the minitablets.

The inclusion of citric acid or pectin in the tablet formulation could thus be used to ensure a slower and more controlled release of the drug as it passes through the colon making it more useful as a colon-specific drug delivery system.

5.3.3 The influence of an enteric coating (Eudragit S®) on drug release

5.3.3.1 Introduction

Chitosan is a weak base with a pKa value of about 6.2 – 7.0 and therefore insoluble at neutral and alkaline pH values. In an acidic medium, the amine group of the polymer is protonated, resulting in a soluble, positively charged polysaccharide with a high charge density (Hejazi & Amiji, 2003:153). For drug delivery to the colon, it is therefore necessary to protect the tablets from the acid environment of the stomach

to ensure that the chitosan does not dissolve at the low pH and release the drug in the stomach.

Different mechanisms can be used to protect dosage forms from the stomach contents and lots of research has been done on the subject. The mechanism that attracts the most interest is to coat the dosage form with an enteric coating such as the methacrylate polymers which will only dissolve at the required pH. Other approaches include the formation of biodegradable complexes which will be degraded by the bacteria in the small intestine or colon or placing the tablets into capsules that could be coated or are manufactured from insoluble polymers.

To establish if the minitablets used in this study could be coated to ensure drug delivery to the colon, it was decided to apply coatings of Eudragit S® of different thickness to the tablets and investigate the dissolution of isoniazide from the tablets in 0.1 M HCl as the dissolution medium.

5.3.3.2 Method

Minitablets were compressed at a constant punch depth (setting 34) and a percentage compaction of 40% using the same powder mixture as described above (see section 5.2.1). The tablets were coated with a varying amount of Eudragit S® by submerging the tablet in a 12.5% solution of the polymer in 2-propanol. The coating was allowed to dry before the next coat was applied. Figures 5.5, 5.6 and 5.7 show the surface of the uncoated tablets, the surface of the coated tablet and a cross section of a coated tablet respectively. The tablets were weighed at the start of the experiment and after each coating to establish the amount of coating on the tablets and are expressed as the percentage increase in the initial weight. The dissolution of isoniazide from the tablets were determined using 0.1 M HCl as the dissolution medium as described in section 5.2.2.

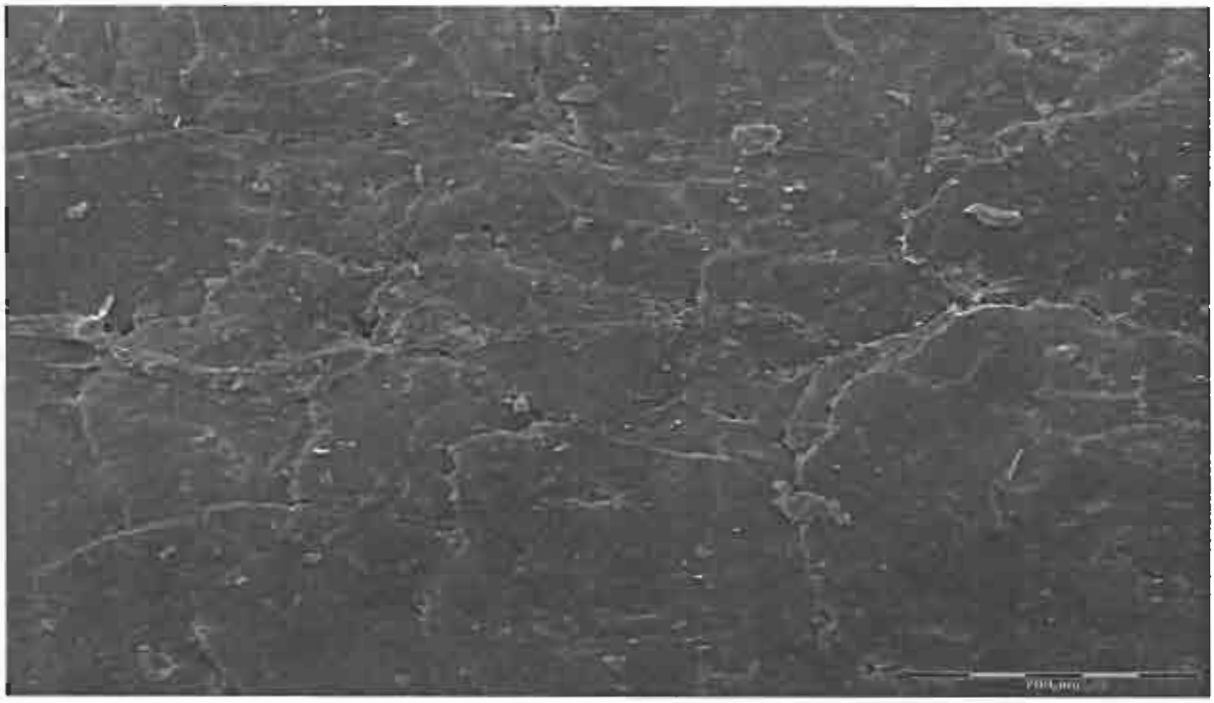

Figure 5.5: *Surface of the uncoated chitosan minitablets.*

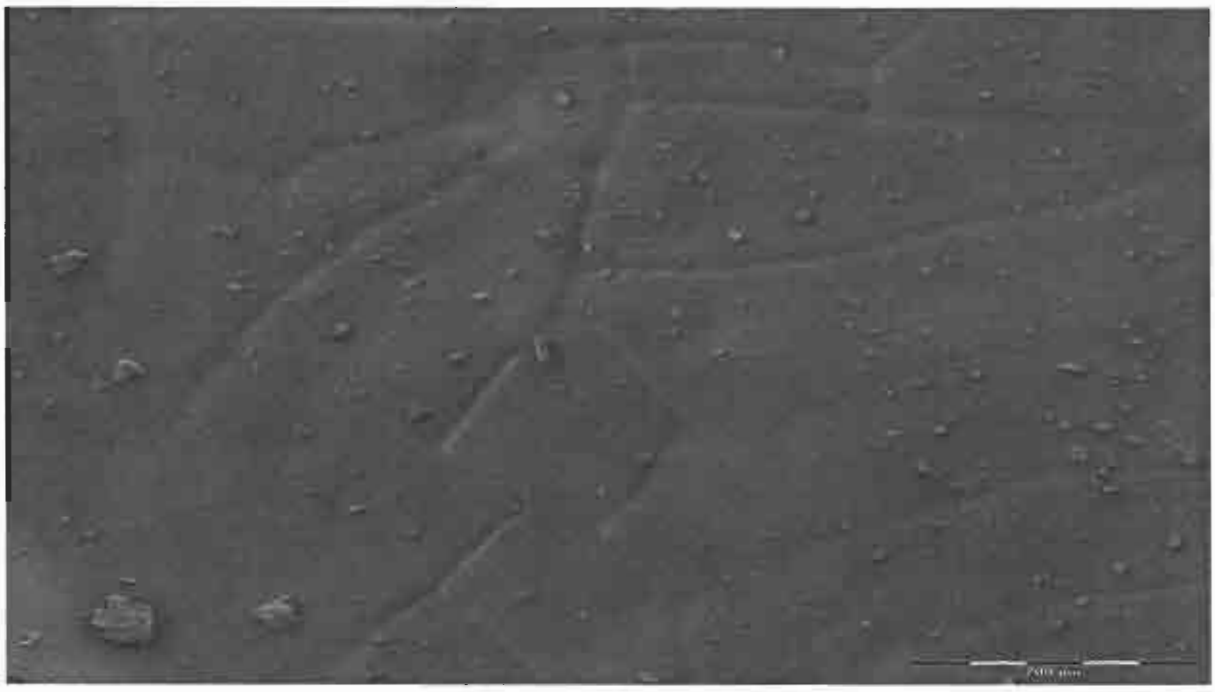

Figure 5.6: *Surface of the coated chitosan minitablets.*

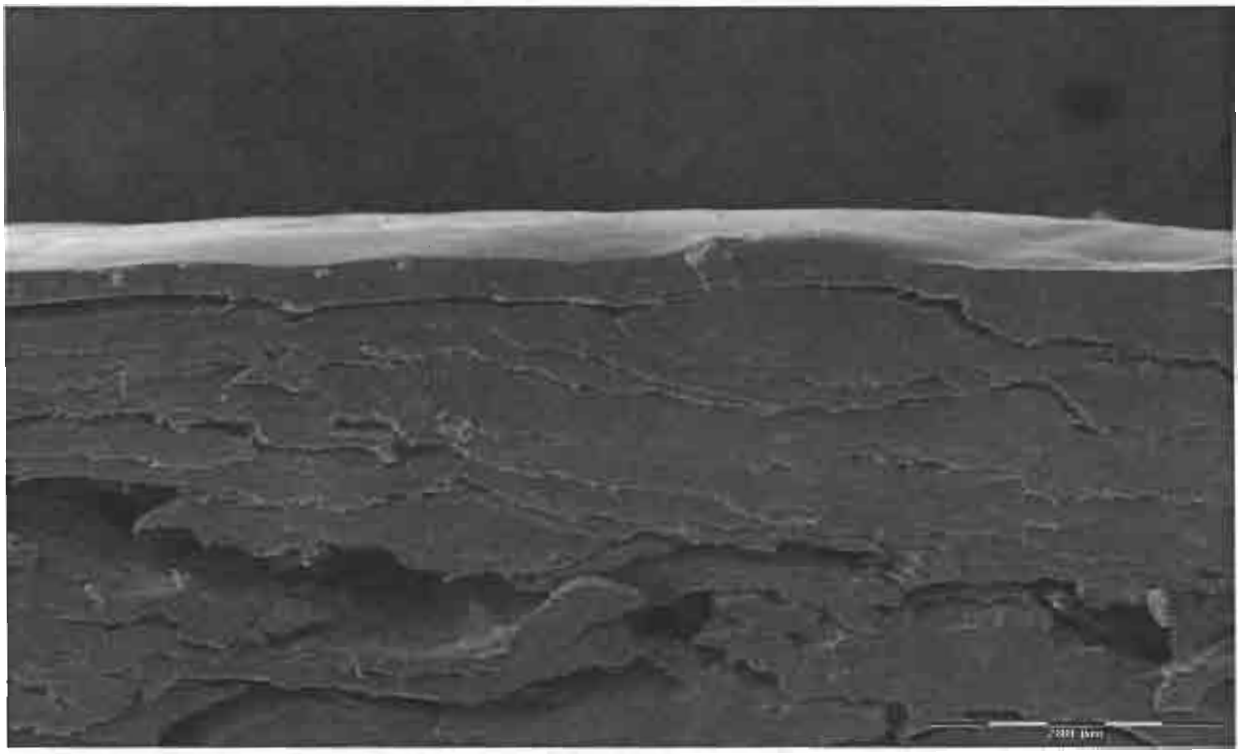

Figure 5.7: *Cross section of the coated minitablets showing the coating layer.*

5.3.3.3 Results

The results of the dissolution test is given in figure 5.8 and in table A.57 in annexure A while the MDT values and the similarity factors for the profiles are given in table 5.12 and 5.13 respectively. The MDT values for each dissolution profile are given in tables A.62 in annexure A.

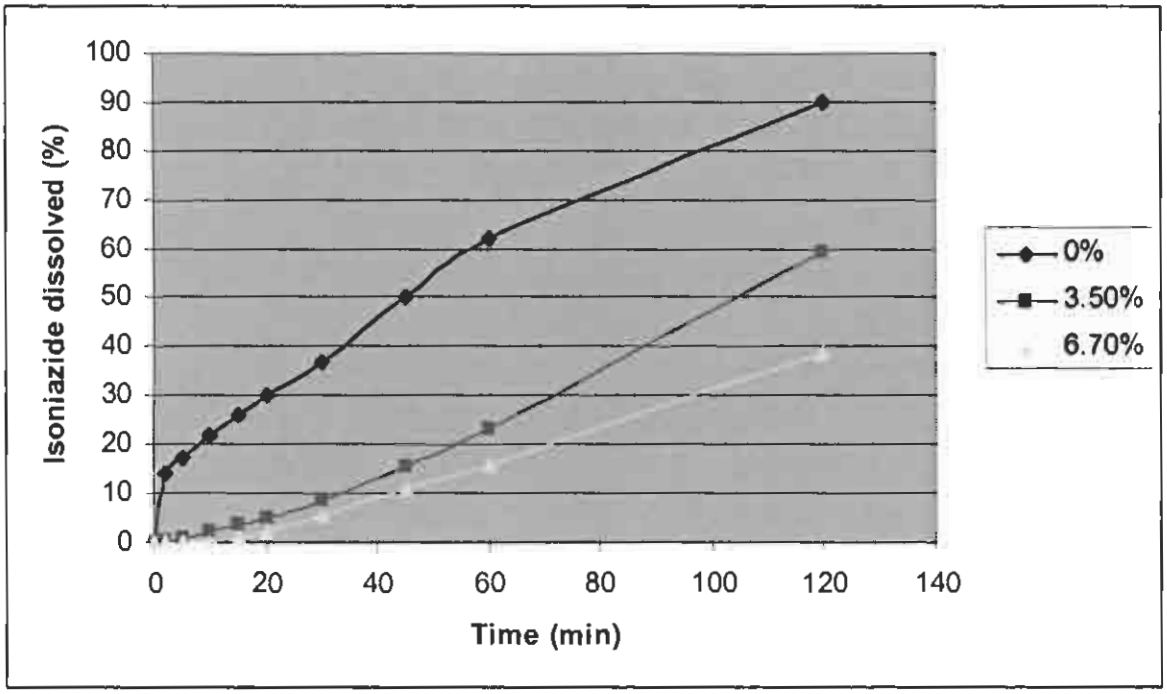

Figure 5.8: *Dissolution profile of isoniazide from chitosan minitablets coated with different amounts of Eudragit S®.*

Table 5.12: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of Eudragit S®.*

Percentage Eudragit S®	*Mean dissolution time (MDT)*
0	*44.24 ± 1.48*
3.5	*68.40 ± 2.75*
6.7	*68.31 ± 4.67*

Table 5.13: *The similarity factor (f_2) of the dissolution profiles of isoniazide from chitosan minitablets coated with different amounts of Eudragit S®.*

	0%	3.5%
3.5 %	33.40 ± 1.10	-
6.7 %	29.03 ± 0.76	60.38 ± 4.99

The MDT values differ from 44.24 for the uncoated tablets to 68 .40 and 68.31 for the tablets coated with a 3.5% and a 6.7% coating respectively. The ANOVA variance test shows a significant difference in the MDT values of the uncoated tablets when compared to the coated tablets. There were no significant differences between the tablets that were coated with a 3.5% coating and those that were coated with the 6.7% coating.

The similarity factor also shows that there is a bigger similarity between the coated tablets (f_2 = 60.38 ± 4.99) than between the uncoated tablets and those coated with the Eudragit S® (f_2 = 33.40 and 29.03).

It is clear from the results that the Eudragit S® coatings that were applied protected the tablets from the low pH and resulted in a much slower onset of dissolution of the isoniazide from the tablets. It can be concluded that the coating of the minitablets with an enteric coating will protect the tablets from the harsh environment of the stomach and will result in the tablets reaching the intestines were bacteria in the colon can degrade the matrix of the tablets.

5.4 SUMMARY

The ideal colon-specific drug delivery system would prevent drug release in the stomach and small intestine and start releasing the drug at controlled rates upon entry into the colon (Zambito & Di Colo, 2003:274). As a result, investigations was done to see if the drug release from the minitablets could be manipulated by process- and formulation variables.

The process variables (punch depth and compaction) did have some effect on the dissolution of isoniazide from the minitablets. The dissolution rate was still high with 90% isoniazide dissolved in 20 minutes in all the formulations. This suggests that although the tablets became less porous with an increase in the punch depth and percentage compaction, the porosity was such that water still penetrated the tablets easily and as a result there was a rapid dissolution of the isoniazide.

Following the investigation of the dissolution of isoniazide out of the chitosan minitablets that were compressed under different process variables, is was concluded that the dissolution may not be sufficiently delayed by these process variables. It was therefore decided to investigate the influence of the addition of citric acid or pectin to the formulation. The inclusion of these excipients lead to a substantial decrease in the dissolution rate of isoniazide from the minitablets. After approximately 20 minutes the percentage isoniazide dissolved increased to 44.64% (citric acid 8%) and 33.42% (pectin 8%) as compared to the more than 90% at all the different punch depth settings and percentages compaction.

Dosage forms to be used for drug delivery to the colon need to be protected from the harsh environment of the stomach to prevent the dosage form of releasing the drug in the stomach or the small intestine. Different mechanisms can be used to protect dosage forms from the stomach contents and lots of research have been done on the subject. The mechanism that attracts the most interest is to coat the dosage form with an enteric coating such as the methacrylate polymers which will only dissolve at the required pH. The minitablets that were compressed in the experiments were coated with Eudragit S®. It is clear from the results that the Eudragit S® coatings that were applied protected the tablets from the low pH and resulted in a much slower onset of dissolution of the isoniazide from the tablets and could as such be used as a colonic drug delivery system.

SUMMARY AND FUTURE PROSPECTS

The aim of the study was to develop a multi-unit dosage form for colonic drug delivery consisting of chitosan minitablets. The different factors that influence the compression of the tablets and the manipulation of the drug release from the tablets were investigated.

The first reason for delivering drugs to the colon is the fact that the colon is a much less harsh environment for drugs than other parts of the GIT because of the almost neutral pH and the low intensity of enzymes. This is beneficial for certain drugs such as peptides which is very vulnerable to degradation in the low pH of the stomach and to enzymatic breakdown. The second reason for delivering drugs to the colon is the need for local treatment of certain diseases.

To deliver the drug to the colon, it must be protected from the environments of the stomach and small intestine. For that reason, different systems have been developed and include bacterial dependant delivery systems, enteric-coated systems and time dependant formulations. Enteric formulations are currently the most widely used systems and consists of dosage forms coated with polymers that would dissolve in the neutral pH of the distal part of the small intestine and the colon. Bacterial dependant systems are very promising for colonic delivery of drugs and consist of degradable polymers that undergo degradation in the colon because of the presence of biodegradable enzymes found only in the colon. Chitosan is an ideal polymers for such systems and is widely available at low cost and has a low toxicity.

There are however two problems associated with the use of chitosan in producing tablet dosage forms namely the poor flowability and compressibility of the chitosan powder.

The first part of the study, the flowability and means to improve the flow characteristics of the chitosan were investigated. The flow of the chitosan powder were determined using measurements of the angle of repose, tap density and critical orifice diameter (COD). It was found that the available apparatus for the determination of the COD had several disadvantages and an novel COD flow

apparatus was developed. The apparatus was patented (SA Patent 2006/004483) and details about the apparatus was presented at the 66th International Congress of FIP held in Salvador, Brazil. This apparatus made it possible to obtain COD values that could be used to characterize the flow properties of the chitosan. A composite flow index (CI), using the values obtained from the above mentioned methods, was calculated. Powders exhibiting good flow would have a composite index value of 100 and when exhibiting poor flow (or no flow at all) the value would be 0.

Compared to other tablet excipients, chitosan showed poor flowability having a CI value of 37.9 while powders with better flow properties such as Ludipress® had a CI value of 89.9. High moisture content lead to an decrease in the CI of the chitosan while a bigger size fraction and the addition of a glidant (talc) increased the CI. A CI value of 60.3 was calculated for the powder fraction > 212 μm containing 0.5% talc as a glidant and this powder mixture showed good flowability that could be compared to that of Emcocell 90M® (66.4) and Prosolv MCC90® (67.7). This powder mixture was used in further experiments during the study. The chitosan rapidly absorbed moisture from the atmosphere (up to 8.93%) and this absorption lead to a decrease in the flowability. This absorption was influenced by the relative humidity (RH) and temperature and as a result, care had to be taken to regulate these factors in the laboratory.

In the second part of the study the compressibility of chitosan was investigated. Initially tablets were compressed with a modified IR press because of better control and higher compression forces that could be used in the compression of the tablets. An apparatus was also developed to measure the wettability of the tablets. This apparatus could measure the amount of water per second that was absorbed by the tablets. The results showed that tablets could be compressed and that the tensile strength of the tablets could be increased by a increase in the compression force. The moisture content, the tablet weight and particle size fraction also influences the tablet characteristics. The wettability experiments showed that the tablets were wetted easily by water, even at the higher compression forces. The tablets absorbed more water at lower compression forces but the increase in the thickness as a result of swelling in the water, were not significantly different. The disintegration studies showed a much longer disintegration time as the compression force increased.

These results lead to the conclusion that the tablets that were compressed were very porous and that high compression forces were necessary to compress tablets that could be used as a dosage form intended for colonic drug delivery.

These high compression forces could not be obtained using the eccentric tablet press with which the minitablets were to be compressed. As a result it was decided to modify the eccentric press so that the chitosan could be compacted during the compression cycle and the die the filled again. The resulting increase in the amount of powder in the die would increase the compression force exerted on the powder. The motor on the eccentric press was replaced with a stepper motor that was controlled with a computer to stop and reverse the compression cycle after the chitosan was compacted. The die would the be filled again with the chitosan after which the cycle was stopped again and reversed to the original compression cycle. The tablets that was compressed using the modified press were less porous and had acceptable tablet characteristics such as a high crushing strength. The percentage compaction could also be varied and a higher percentage compaction lead to tablets with a much higher crushing strength (104.73 N at a compaction percentage of 60%) as compared to a crushing strength of approximately 50 N if not compacted. The disintegration study however showed that the tablets were still wetted and swelled within 2- 5 minutes.

It is important that the drug release from these minitablets can be controlled and manipulated. In the last part of the study, the release of isoniazide (as an example of a water soluble drug) from the minitablets was investigated.

Changing the process variables (percentage compaction and punch depth) still lead to porous tablets from which the dissolution of isoniazide was rapid. An increase in the percentage compaction an well as an increase in the punch depth retarded the dissolution rate but a longer dissolution time was desirable. Other option to achieve this goal was the inclusion of excipients such as an organic acid (that would lower the pH in the tablet, allowing the chitosan to gel) or pectin (which would form an insoluble complex with the chitosan). Results showed that the inclusion of these excipients prolonged the dissolution of the isoniazide from the tablets and that the tablets could be used for drug delivery to the colon.

The coating of the minitablets with an enteric coated polymer (Eudragit S®) delayed the dissolution and would therefore be able to protect the tablets from the low pH of the stomach and would dissolve in the neutral pH of the distal small intestine and colon resulting in colonic drug delivery.

Future investigations into the use of chitosan minitablets for colonic drug delivery could include

- the use of chitosan salts that are soluble at lower pH values,
- the use of different tablet excipients in combination with the process variables in the compression of the minitablets and
- the effect of enzymes or colonic bacteria on the dissolution of drugs from the minitablets.

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Publication

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A NEW FLOW METER FOR THE MEASUREMENT OF POWDER FLOW BY MEANS OF THE CRITICAL ORIFICE DIAMETER.

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BACKGROUND

The critical orifice diameter (COD) is one of the well known parameters used in quantifying powder flow. This parameter can be defined as "*the smallest orifice diameter through which a powder can flow freely under the influence of gravity.*" Taylor *et al.* (2000) included this parameter in their composite index (CI) for a variety of pharmaceutical powders with different flow properties.

Many flow apparatuses used to determine the COD show inherent problems including (i) static powder regions in the corners between the cylinder wall and the cylinder floor, (ii) static regions between the floor and the shutter, (iii) formation of "rat holes" or "pipes" in the powder bed through which powders fall rather than flow and (iv) adhesion phenomena due to the material from which these apparatuses are manufactured (Staniforth, 2002:202-205). These factors affect spontaneous powder flow and result in inaccurate results, especially in powders exhibiting poor to extremely poor flow.

The results obtained with the new flow meter indicate:

- a more accurate determination of actual powder flow ;
- a higher scale of scrutiny for distinguishing between the flow of commonly used pharmaceutical powders (especially those with good and poor flow) and
- the value of the COD as indicator of powder flow (compared to the angle of repose [AOR] and Carr's index or the % compressibility (%C).

Other advantages of this apparatus fall in the field of education. It is relatively cheap, easy for students to set-up and use, strong and rigid with low maintenance and "student proof".

MATERIALS

Excipients used in this study were obtained from commercial suppliers and included Avicel® PH-200, Chitosan, Emcocell® 50M & 90M, Emcompress®, Ludipress®, Prosolv® SMCC50 & SMCC90, and Tablettose®.

APPARATUS AND METHODS

Flow meter for measuring critical orifice diameter

The new flow meter comprises of a number of brass cylinders (8 to 15 mm thick) which can be stacked on top of each other to form a funnel (figures 1 and 2 and sketches under "Technical drawings"). Each cylinder has a circular bore which pass through the centre of the cylinder with an inlet and an outlet (orifice diameter), the latter being smaller than the inlet. The passage is thus in the form of a cone. The arrangement is such that the bore of each cylinder tapers from the inlet to the outlet at an angle of approximately 30° (corresponding with the angle in industrial tablet press hoppers). The bores of the cylinders are aligned with one another with the outlet of the bore of any cylinder the same as the inlet of the cylinder directly below it. The orifice diameters range from 1 mm tot 24 mm. The apparatus further includes:

- a hopper for containing the powder and an additional cylindrically shaped container for containing more powder, which fits onto the hopper. The diameter of the hopper and the container are the same;

- a stand defining an opening with a shutter for opening and closing the opening. Supports are provided at the bottom of the stand to support the apparatus in an upright position and away from the surface on which it is positioned.

The apparatus was set up as shown in figure 3, and filled with a sample of the powder, whilst the shutter was closed. The shutter was opened and the powder allowed to flow through the passage towards the outlet. If the passage outlet was too small, the powder would not be able to flow through the said passage outlet. The

lower most cylinder was then removed so that the passage had a larger diameter. The bottom cylinder was removed from the lower end of the apparatus until the smallest bore through which the powder could flow freely was determined. This bore opening was taken as the critical orifice diameter of the powder. A smaller critical orifice diameter value indicated better flow. The critical orifice diameter of each excipient was determined as the average (of 3 successive runs) of the smallest orifice through which the powder flowed freely on.

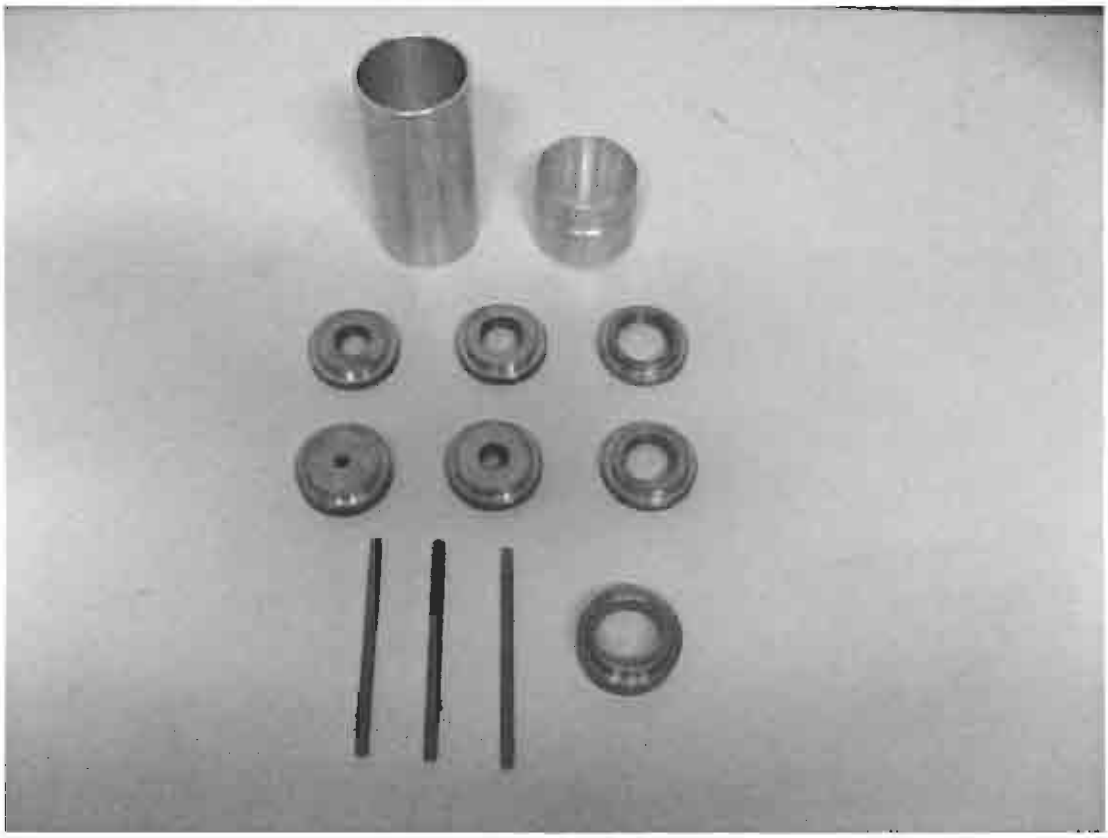

Figure 1: The various components of the flow meter.

Figure 2: Set-up showing the funnel formed by the cylinders when stacked correctly.

Figure 3: *A picture showing the set-up of the flow meter.*

Angle of repose

Approximately 200 ml of powder was poured through a funnel from a height of 8 cm onto a leveled glass plate. The angle of repose (θ) was determined from the angle that the side of the conical powder heap made with the horizontal plane. Lower angles of repose represent better flow.

Carr's Index (Percent compressibility index)

Approximately 100 ml of powder was gently poured into a tared graduated cylinder and the initial volume and weight of the material was noted. The cylinder was vibrated until the volume remained constant and the final volume was noted. Lower percent compressibility values represent better flow (see equation below)

Composite Index

The composite index (CI) of each powder was calculated using the integrated equations supplied by Taylor *et al.* (2000).

RESULTS AND DISCUSSION

Results

The results of the tests are presented in table 1, whilst figure 4 provides a graphical view of the data.

Table 1: ***The COD, AOR, %C and composite index (CI) of the various excipients.***

Excipient	COD (mm)	AOR (degrees)	% C	CI
Avicel® PH-200	1.5	32.5	18.0	89.1
Chitosan	12.0	45.8	34.7	57.4
Emcocell® 50M	24.0	41.7	28.3	51.1
Emcocell® 90M	11.0	37.5	25.0	70.6
Emcompress®	1.5	36.8	16.0	87.6
Ludipress®	2.0	34.1	17.3	88.0
Tablettose®	6.0	34.1	17.3	77.3
Prosolv ®SMCC50	16.0	39.2	30.3	60.3
Prosolv ®SMCC90	9.0	38.0	25.3	72.2

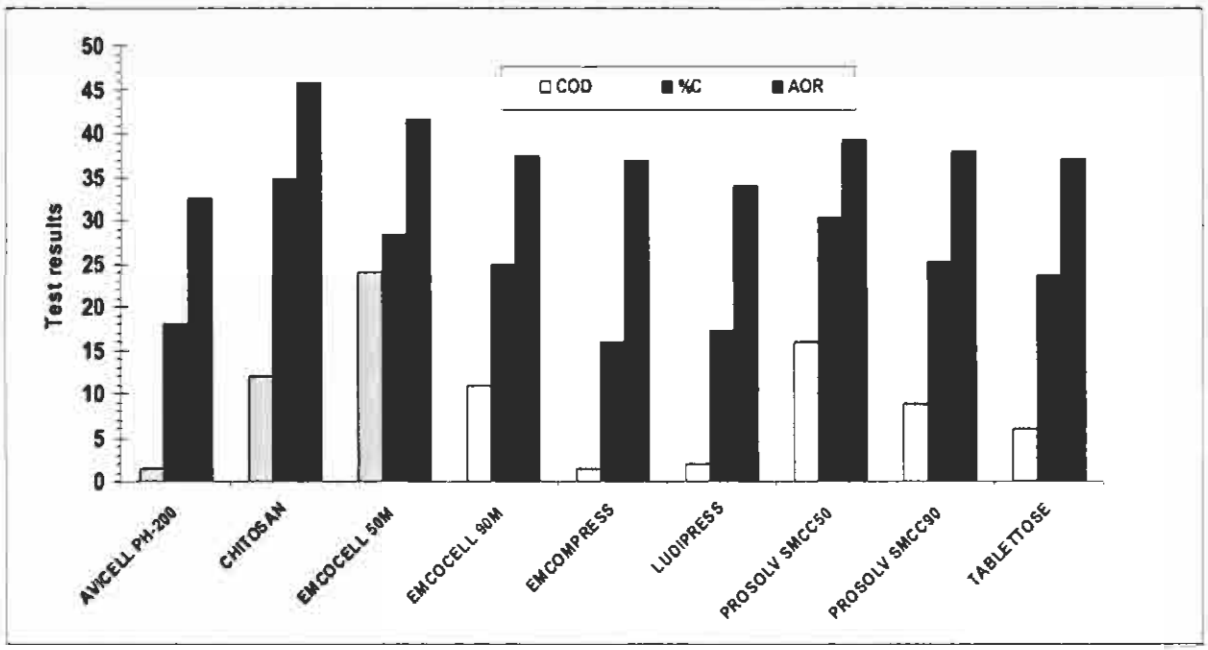

Figure 4: ***Graphical presentation of the results of different flow tests on various pharmaceutical excipients.***

Figure 5 shows the relationship between the test results (COD, %C and AOR) of the excipients and the composite index for that particular excipient.

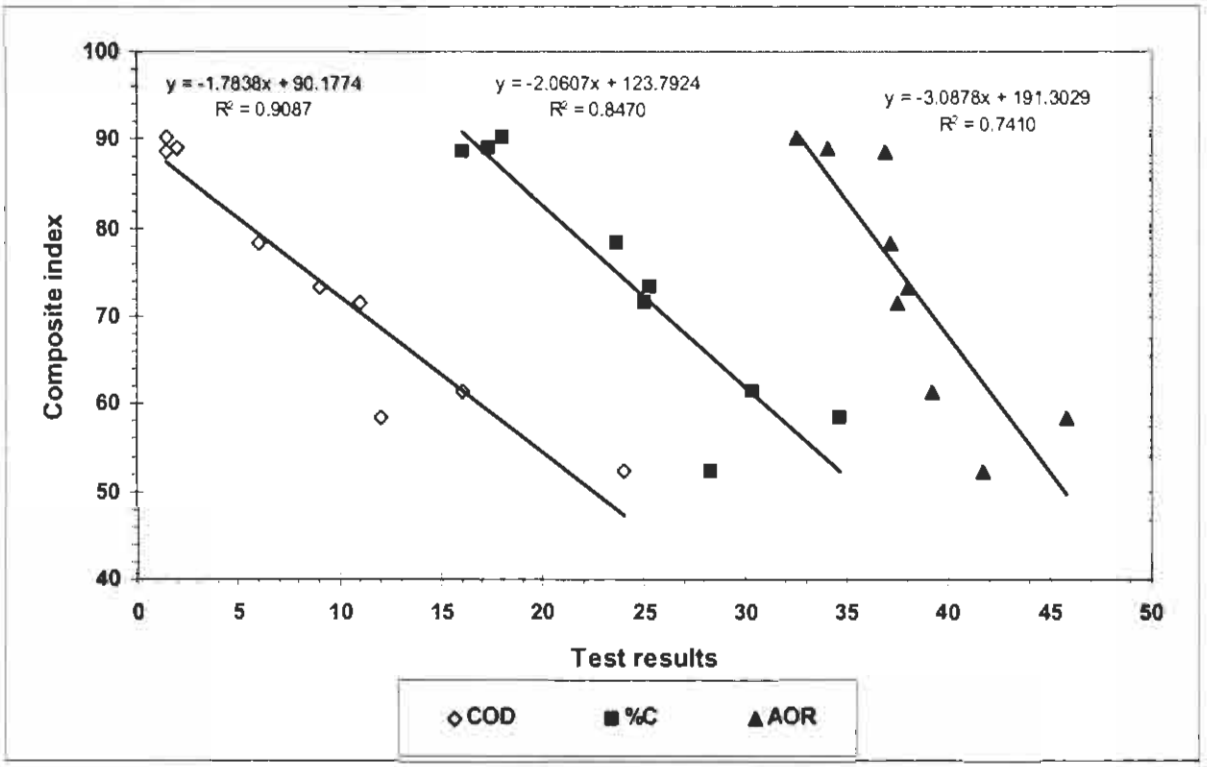

Figure 5: ***Relationship between the composite index and the COD, %C and AOR of the various pharmaceutical excipients.***

Discussion

All three tests could distinguish between the flow properties of the excipients that were tested. The difference between the results for a particular test however varied from 16x for the COD, 2.2x for the %C and 1.4x for the AOR, indicating a much higher degree of sensitivity for differences in powder flow obtained with the test for COD than for the other two tests (table 1 and figure 4). These results could possible be contributed to the specific design of the flow meter used to determine the COD, especially the material used, the angle of the tapered funnel and the absence of static areas in the apparatus.

The low COD's (1.5 to 2.0 mm) measured for Avicel® PH200, Ludipress® and Emcompress® clearly demonstrated the ability of the flow meter to identify (recognize) excellent flow properties and its ability to distinguish / discriminate between powders with subtle differences in flow.

The relationship between the COD and the composite index showed a much higher correlation over the entire range of excipients tested ($r^2 > 0.91$), compared to values

of $r^2 > 0.84$ for the %C and $r^2 > 0.74$ for the AOR, which emphasized the accuracy of the COD (and the apparatus used) as an indicator of powder flow (figure 5).

Interestingly, although maybe not significant, there was a higher correlation between the results from the tests for %C and the COD ($r^2 \approx 0.598$) than between the AOR and the COD ($r^2 \approx 0.484$), whilst the highest correlation was obtained between the results from the tests for AOR and %C ($r^2 \approx 0.774$).

CONCLUSIONS

- All three tests could separate between the various powders according to their inherent flowability. From figure 5 it is clear that the selected excipients could be divided into three categories, namely those with CI values:
  - between 80 and 90, namely Avicel® pH200; Emcompress® and Ludipress® (indicating excellent flow);
  - between 70 and 80, namely Tablettose®, Emcocell® 90M and Prosolv® SMCC90 (good to fair flow) and
  - below 70, namely chitosan, Emcocell® 50M; Prosolv® SMCC50 (poor to extremely poor flow)..
- Whilst both the test for COD and %C could accurately distinguish between powders with fair/good to good/excellent flow (concluded form the deviation of the individual scores from the straight lines drawn through the scores), all three tests somewhat fall short in accurately measuring the flow properties of the powders with poor to extremely poor flow (indicated by a much larger deviation from the respective lines).
- The composite index, as constructed by Taylor *et al.* (2000), provides an extremely accurate estimate of the flowability of powders. However, due to the apparent higher accuracy or sensitivity of the COD test (compared to the other two tests used in determining the CI), this model can perhaps be refined by increasing the weight assigned to this parameter in the determination of the composite index. Furthermore, the inclusion of the flow rate of powders (which is

an important factor in high-speed tablet production) could also be introduced into this model.

- The flow meter can also be used to accurately determine the flow rate of pharmaceutical powders through the critical orifice diameter for each powder.

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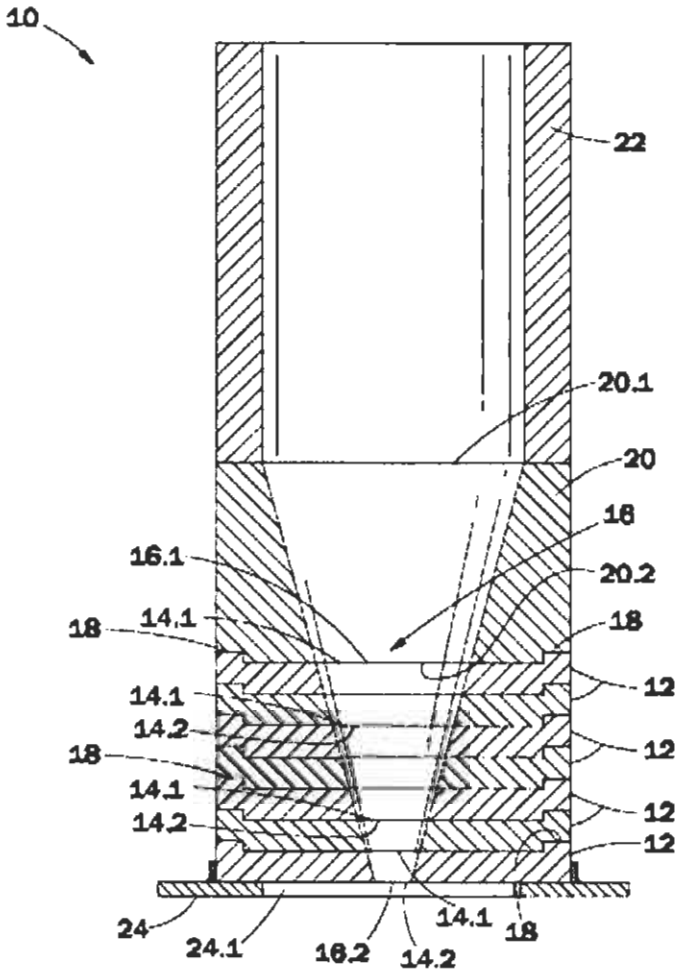
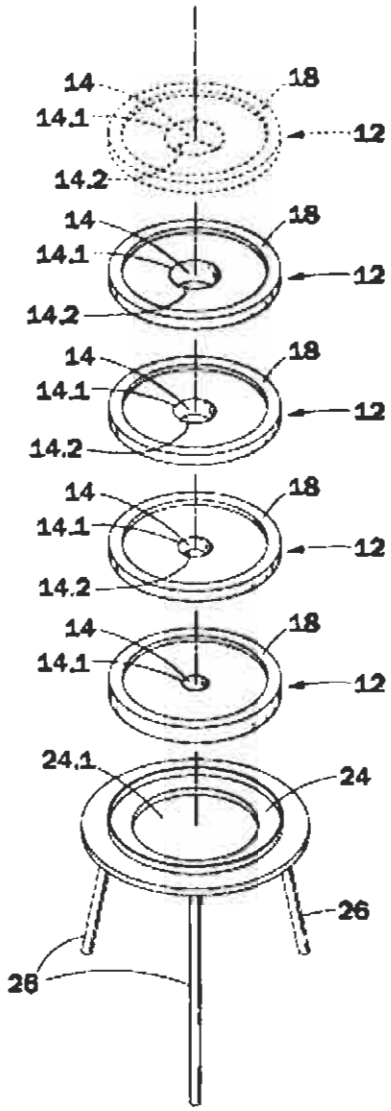
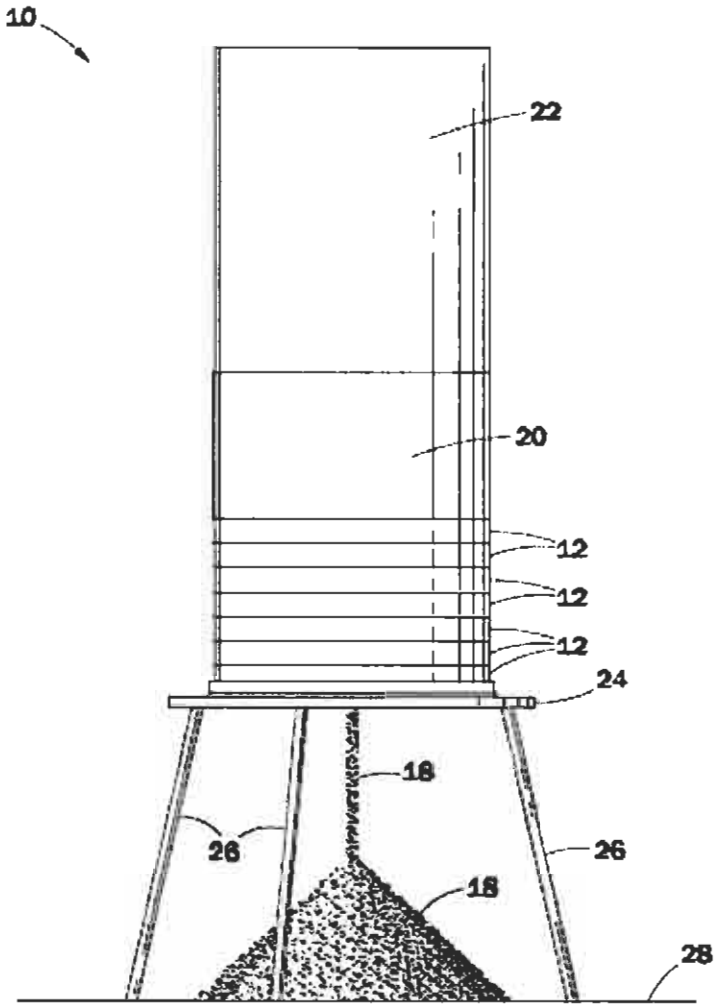
TECHNICAL DRAWINGS



Drawing 1: *Longitudinal sectional side view*



Drawing 2: *Exploded perspective view*



Drawing 3: *Side view of the apparatus*

ANNEXURE A

Table A.1: *The measurement of the tap density of some pharmaceutical excipients.*

Excipient	Powder mass (g)	Sample 1 (ml)	Sample 2 (ml)	Sample 3 (ml)	Average (ml)
Emcompress®	73.25	84	84	84	84.00
Emcocell 50M®	30.54	73	71	71	71.67
Emcocell 90M®	32.36	75	75	75	75.00
Chitosan	20.10	65	65	66	65.33
Avicell PH-200®	30.08	69	70	70	69.67
Ludipress®	52.43	84	80	84	82.67
Prosolv SMCC90®	34.65	75	74	75	74.67
Prosolv SMCC50®	33.04	70	69	70	69.67

Table A.2: *The determination of Carr's index and Hausner ratio of some pharmaceutical excipients.*

Excipient	Powder mass (g)	Initial bulk density (g/ml)	Final bulk density (g/ml)	Carr's index (%)	Hausner ratio
Emcompress®	73.25	0.7325	0.8720	15.998	1.2
Emcocell 50M®	30.54	0.3054	0.4261	23.327	1.4
Emcocell 90M®	32.36	0.3236	0.4315	25.006	1.3
Chitosan	20.10	0.2010	0.3077	39.233	1.5
Avicell PH-200®	30.08	0.3008	0.4318	30.338	1.4
Ludipress®	52.43	0.5243	0.6342	17.329	1.2
Prosolv SMCC90®	34.65	0.3465	0.4641	25.339	1.3
Prosolv SMCC50®	33.04	0.3304	0.4743	32.448	1.4

Table A.3: *The determination of the angle of repose of pharmaceutical excipients.*

Excipient	Height (mm)	Diameter (mm)	Tan α	Angle of repose (°)	Average angle of repose (°)
Emcompress®	56	144	0.77778	37.87	
	30	84	0.71429	35.54	
	56	148	0.75676	37.12	36.84
Emcocell 50M®	65	136	0.95588	43.71	
	46	103	0.89320	41.77	
	63	153	0.82353	39.47	41.65
Emcocell 90M®	57	141	0.80851	38.96	
	55	147	0.74830	36.81	
	50	134	0.74627	36.73	37.50
Chitosan	81	137	1.18248	49.78	
	48	101	0.95050	43.55	
	70	144	0.97222	44.19	45.84
Avicell PH-200	56	153	0.73203	36.21	
	67	156	0.85897	40.66	
	59	150	0.78667	38.19	38.35
Ludipress®	52	158	0.65823	33.35	
	45	143	0.62937	32.19	
	61	164	0.74390.	36.65	34.06
Prosolv SMCC90®	51	140	0.72857	36.08	
	57	146	0.78082	37.98	
	61	146	0.83562	39.88	37.98
Prosolv SMCC50®	57	150	0.76000	37.23	
	64	148	0.86486	40.86	
	62	150	0.82667	39.58	39.22

Table A.4: *Determination of the composite index of some pharmaceutical excipients.*

Excipient	Critical orifice (mm)	Tap density	Angle of repose (°)	Transformed value critical orifice	Transformed value tap density	Transformed value angle of repose	Composite index
Chitosan	12	39.233	45.84	20.69	10.26	6.93	37.9
Emcocell 50M®	24	28.327	41.65	6.90	20.64	13.92	41.4
Prosolv SMCC50®	16	32.448	39.22	16.09	16.71	17.97	50.8
Avicell PH-200®	18	30.338	38.35	13.79	18.72	19.42	51.9
Emcocell 90M®	11	25.006	37.50	21.84	23.75	20.83	66.4
Prosolv SMCC90®	9	25.339	37.98	24.14	23.49	20.03	67.7
Emcompress®	1.5	15.998	36.84	32.76	32.38	21.93	87.1
Ludipress®	2	17.329	34.06	32.18	31.11	26.56	89.9

Table A.5: *Loss on drying of chitosan at 40 °C under vacuum.*

Container 1				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture lost (g)**	**Loss on drying (%)**
0	58.49	10.00	0.00	0.00
1	57.92	9.43	0.57	5.70
2	57.78	9.29	0.71	7.10
3	57.72	8.23	0.77	7.70
4	57.69	9.20	0.80	8.00
5	57.68	9.19	0.81	8.12

Container 2				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture lost (g)**	**Loss on drying (%)**
0	57.12	10.00	0.00	0.00
1	56.54	9.42	0.58	5.80
2	56.42	9.30	0.70	7.00
3	56.35	9.23	0.77	7.70
4	56.32	9.20	0.80	8.00
5	56.30	9.18	0.82	8.16

Container 3				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture lost (g)**	**Loss on drying (%)**
0	56.70	10.00	0.00	0.00
1	56.12	9.42	0.58	5.80
2	56.02	9.32	0.68	6.80
3	55.92	9.22	0.78	7.80
4	55.89	9.19	0.81	8.10
5	55.89	9.19	0.81	8.10

Table A.6: *Moisture increase at 25 °C and 60% RH.*

Container 1				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	61.80	10.00	0.00	0.00
0.5	61.90	10.10	0.10	1.00
1	62.01	10.21	0.21	2.10
2	62.15	10.35	0.35	3.50
3	62.25	10.45	0.45	4.50
4	62.34	10.54	0.54	5.41
5	62.40	10.60	0.60	6.00
6	62.44	10.64	0.64	6.40
8	62.45	10.65	0.65	6.50

Container 2				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	48.15	10.00	0.00	0.00
0.5	48.29	10.14	0.14	1.40
1	48.41	10.26	0.26	2.60
2	48.59	10.44	0.44	4.40
3	48.71	10.56	0.56	5.60
4	48.79	10.64	0.64	6.40
5	48.85	10.70	0.70	7.00
6	48.89	10.74	0.74	7.40
8	48.90	10.75	0.75	7.50

Container 3				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	60.46	10.00	0.00	0.00
0.5	60.57	10.11	0.11	1.10
1	60.69	10.23	0.23	2.30
2	60.91	10.45	0.45	4.50
3	60.96	10.50	0.50	5.00
4	61.04	10.58	0.58	5.80
5	61.11	10.65	0.65	6.50
6	61.16	10.70	0.70	7.00
8	61.18	10.72	0.72	7.20

Table A.7: *Moisture increase at 40 °C and 75% RH.*

Container 1				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	61.81	10.00	0.00	0.00
0.5	62.26	10.45	0.45	4.50
1	62.52	10.71	0.71	7.10
2	62.62	10.81	0.81	8.10
3	62.71	10.90	0.90	9.00
4	62.75	10.94	0.94	9.40
5	62.75	10.94	0.94	9.40
6	62.74	10.93	0.93	9.30
8	62.74	10.93	0.93	9.30

Container 2				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	58.50	10.00	0.00	0.00
0.5	58.91	10.41	0.41	4.10
1	59.01	10.51	0.51	5.10
2	59.20	10.70	0.70	7.00
3	59.30	10.80	0.80	8.00
4	59.34	10.84	0.84	8.40
5	59.35	10.85	0.85	8.50
6	59.35	10.85	0.85	8.50
8	59.34	10.84	0.84	8.40

Container 3				
Time (h)	**Total mass (g)**	**Mass chitosan (g)**	**Moisture increase (g)**	**Moisture increase (%)**
0	57.12	10.00	0.00	0.00
0.5	57.53	10.41	0.41	4.10
1	57.81	10.69	0.69	6.90
2	57.92	10.80	0.80	8.00
3	57.99	10.87	0.87	8.70
4	58.03	10.91	0.91	9.10
5	58.03	10.91	0.91	9.10
6	58.04	10.92	0.92	9.20
8	58.03	10.91	0.91	9.10

Table A.8: *The measurement of the tap density of chitosan stored under different humidities.*

Chemical	Powder mass (g)	Sample 1 (ml)	Sample 2 (ml)	Sample 3 (ml)	Average (ml)
None	3.96	15.3	15.2	15.1	15.20
LiCl	4.05	15.3	15.2	15.2	15.23
$MgCl_2$	4.12	14.8	14.5	15.0	14.77
$MgNO_3$	4.57	15.0	124.9	14.5	14.80
NaCl	4.33	14.0	14.0	14.2	14.07

Table A.9: *The determination of Carr's index and Hausner ratio of chitosan stored under different humidities.*

Chemical	Powder mass (g)	Initial bulk density (g/ml)	Final bulk density (g/ml)	Carr's index (%)	Hausner ratio
None	3.96	0.1801	0.2607	30.917	1.4
LiCl	4.05	0.1841	0.2658	30.737	1.4
$MgCl_2$	4.12	0.1872	0.2789	32.879	1.5
$MgNO_3$	4.57	0.2077	0.3088	32.740	1.5
NaCl	4.33	0.1968	0.3078	36.062	1.6

Table A.10: *The determination of the angle of repose of chitosan stored under different humidities.*

Chemical	Average height (mm)	Diameter (mm)	Tan α	Average angle of repose (°)
None	24.3	50	0.97333	44.23
LiCl	25.0	50	1.0000	45.00
$MgCl_2$	24.7	50	0.98667	44.62
$MgNO_3$	24.3	50	0.97333	44.23
NaCl	25.3	50	1.01333	45.38

Table A.11: *Determination of the composite index of chitosan stored under different humidities.*

Chemical	Critical orifice (mm)	Tap density	Angle of repose (°)	Transformed value critical orifice	Transformed value tap density	Transformed value angle of repose	Composite index
None	10	30.917	44.23	22.99	18.17	9.62	50.8
LiCl	11	30.737	45.00	21.84	18.354	8.33	48.5
$MgCl_2$	12	32.878	44.62	20.69	16.31	8.97	46.0
$MgNO_3$	15	32.740	44.23	17.24	16.44	9.62	43.3
NaCl	16	36.062	45.38	16.09	13.27	7.70	37.1

Table A.12: *The measurement of the tap density of different size fractions of chitosan.*

Particle size (μm)	Powder mass (g)	Sample 1 (ml)	Sample 2 (ml)	Sample 3 (ml)	Average (ml)
< 90	17.00	60	59	59	59.33
90 – 150	17.00	64	65	64	64.33
150 – 212	17.00	68	69	69	68.67
> 212	17.00	71	70	70	70.33

Table A.13: *The determination of Carr's index and Hausner ratio of different size fractions of chitosan.*

Particle size (μm)	Powder mass (g)	Initial bulk density (g/ml)	Final bulk density (g/ml)	Carr index (%)	Hausner ratio
< 90	17.00	0.1700	0.2865	40.663	1.74
90 – 150	17.00	0.1700	0.2642	35.655	1.6
150 – 212	17.00	0.1700	0.2476	31.341	1.5
> 212	17.00	0.1700	0.2417	29.665	1.4

Table A.14: *The determination of the angle of repose of different size fractions of chitosan.*

Particle size (μm)	Height (mm)	Diameter (mm)	Tan α	Angle of repose (°)	Average angle of repose (°)
< 90	89.0	166.0	1.07229	47.00	
	95.0	182.0	1.04396	46.23	
	88.0	184.0	0.95652	43.73	45.65
90 - 150	84.0	188.0	0.89362	41.78	
	89.0	184.0	0.96739	44.05	
	88.0	186.0	0.94624	43.42	43.08
150 - 212	82.0	166.0	0.98795	44.65	
	89.0	172.0	1.03488	45.98	
	85.0	160.0	1.06250	46.74	45.79
> 212	78.0	190.0	0.82105	39.39	
	90.0	172.0	1.04651	46.30	
	90.0	188.0	0.95745	43.75	43.15

Table A.15: *Determination of the composite index of different size fractions of chitosan.*

Particle size (μm)	Critical orifice (mm)	Tap density	Angle of repose (°)	Transformed value critical orifice	Transformed value tap density	Transformed value angle of repose	Composite index
< 90	24	40.663	45.65	6.90	8.89	7.25	23.0
90 – 150	13	35.655	43.08	19.54	13.66	11.53	44.7
150 – 212	9	31.341	45.79	24.14	17.77	7.02	48.9
> 212	9	29.665	43.15	24.14	19.37	11.42	54.9

Table A.16: *The measurement of the tap density of chitosan with different percentages of Cab-O-Sil®.*

Cab-O-Sil® %	Powder mass (g)	Sample 1 (ml)	Sample 2 (ml)	Sample 3 (ml)	Average (ml)
0	11.65	47.5	48.0	48.0	47.83
1	12.60	48.0	48.5	48.0	48.17
2	12.20	48.5	49.5	49.0	49.00
3	12.25	50.0	50.0	50.0	50.00

Table A.17: *The determination of Carr's index and Hausner ratio of chitosan with different percentages of Cab-O-Sil®.*

Cab-O-Sil® %	Powder mass (g)	Initial bulk density (g/ml)	Final bulk density (g/ml)	Carr index (%)	Hausner ratio
0	11.65	0.1664	0.2436	31.691	1.5
1	12.60	0.1800	0.2616	31.193	1.5
2	12.20	0.1743	0.2490	30.000	1.4
3	12.25	0.1750	0.2450	28.571	1.4

Table A.18: *The determination of the angle of repose of chitosan with different percentages of Cab-O-Sil®.*

Cab-O-Sil® %	Height (mm)	Diameter (mm)	Tan α	Angle of repose (°)	Average angle of repose (°)
0	90	180.0	1.00000	45.00	
	88	174.0	1.01149	45.33	
	84	167.0	1.00599	45.17	45.17
1.0	78.0	183.0	0.85246	40.45	
	75.0	175.0	0.85714	40.60	
	76.0	172.0	0.88372	41.47	40.84
2.0	84.0	178.0	0.94382	43.34	
	78.0	178.0	0.87640	41.23	
	75.0	165.0	0.90909	42.27	42.28
3.0	82.0	173.0	0.94798	43.47	
	79.0	186.0	0.84946	40.35	
	72.0	160.0	0.90000	41.99	41.93

Table A.19: *Determination of the composite index of chitosan with different percentages of Cab-O- Sil®.*

Cab-O-Sil %	Critical orifice diameter (mm)	Tap density	Angle of repose (°)	Trans-formed value critical orifice	Trans-formed value tap density	Trans-formed value angle of repose	Composite index
0	6	31.691	45.17	27.58	17.44	8.05	53.1
1	5	31.193	40.84	28.73	17.91	15.27	61.9
2	4	30.000	42.28	29.88	19.05	12.87	61.8
3	3	28.571	41.93	31.03	20.41	13.45	64.9

Table A.20: *The measurement of the tap density of chitosan containing different percentages of talc.*

Talc %	Powder mass (g)	Sample 1 (ml)	Sample 2 (ml)	Sample 3 (ml)	Average (ml)
0	11.65	48.0	47.0	48.0	47.67
0.5	12.60	49.0	47.5	49.0	48.50
0.75	12.20	48.5	47.5	49.0	48.33
1.0	12.25	48.5	49.0	49.0	48.83

Table A.21: *The determination of Carr's index and Hausner ratio of chitosan containing different percentages of talc.*

Talc %	Powder mass (g)	Initial bulk density (g/ml)	Final bulk density (g/ml)	Carr's index (%)	Hausner ratio
0	11.65	0.1664	0.2444	31.915	1.5
0.5	12.60	0.1800	0.2598	30.716	1.4
0.75	12.20	0.1743	0.2524	30.943	1.4
1.0	12.25	0.1750	0.2509	30.251	1.4

Table A.22: *The determination of the angle of repose of chitosan containing different percentages of talc.*

Talc %	Height (mm)	Diameter (mm)	Tan α	Angle of repose (°)	Average angle of repose (°)
0	80.0	157.0	1.01911	45.54	
	83.0	165.0	1.00606	45.17	
	80.0	170.0	0.94118	43.26	44.66
0.50	86.0	192.0	0.89583	41.86	
	83.0	175.0	0.94857	43.49	
	85.0	197.0	0.86294	40.79	42.05
0.75	95.0	199.0	0.95477	43.67	
	91.0	210.0	0.86667	40.91	
	86.0	184.0	0.93478	43.07	42.55
1.0	85.0	200.0	0.85000	40.36	
	81.0	186.0	0.87097	41.05	
	82.0	168.0	0.97619	44.31	41.91

Table A.23: *Determination of the composite index of chitosan containing different percentages of talc.*

Talc %	Critical orifice diameter (mm)	Tap density	Angle of repose (°)	Trans-formed value critical orifice	Trans-formed value tap density	Trans-formed value angle of repose	Compo site index
0	7	31.915	44.66	26.43	17.22	8.90	52.6
0.5	5	30.716	42.05	28.73	18.37	13.25	60.3
0.75	6	30.943	42.55	27.58	18.15	12.42	58.1
1.0	6	30.251	41.91	27.58	18.81	13.48	59.9

Table A.24: *The effect of particle size and the addition of talc on the tablet weight of chitosan tablets.*

Tablet nr	Chitosan as received (mg)	Chitosan with particle size > 212 μm (mg)	Chitosan with particle size > 212 μm + talc 0.5 % (mg)
1	28.0	27.6	26.7
2	27.8	24.6	26.5
3	28.5	25.7	26.8
4	29.4	28.1	26.3
5	30.9	27.7	25.7
6	28.7	26.0	26.1
7	28.7	27.9	26.7
8	29.7	28.1	26.0
9	25.5	28.4	26.7
10	26.8	28.5	26.4
11	31.0	27.8	25.9
12	28.6	28.4	26.8
13	25.5	27.7	26.3
14	28.6	27.7	27.0
15	28.5	27.5	26.1
16	29.8	26.5	27.0
17	28.9	26.9	26.9
18	28.6	26.6	27.2
19	29.1	28.3	25.8
20	28.8	27.0	26.4
Average weight	28.59	27.35	26.47
Standard deviation	1.42	1.03	0.43
% standard deviation	4.97	3.77	1.64

Table A.25: *The effect of moisture of chitosan powder on the tensile strength of tablets: dried powder.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.79157	200.5	4.26	8.09	55.6	0.79
15.97944	198.9	4.10	8.06	63.7	1.23
16.25999	201.6	4.41	8.11	47.0	0.84
16.97002	200.5	3.49	8.04	131.2	2.98
17.22488	199.0	3.68	8.08	97.2	2.08
17.24987	198.2	3.52	8.05	129.9	2.92
17.98482	198.7	3.33	8.03	172.6	4.11
18.15814	197.6	3.34	8.02	171.2	4.07
18.34092	200.2	3.46	8.05	140.2	3.20
19.00564	202.7	3.42	8.04	199.0	4.61
19.63885	199.8	3.32	8.04	169.1	4.03
19.64098	201.8	3.38	8.06	156.1	3.65
19.76408	198.1	3.14	8.02	225.6	5.70
20.41594	200.3	3.21	8.03	210.4	5.19
21.12465	195.6	3.10	8.02	215.7	5.52
21.34668	200.1	3.17	8.03	238.6	5.96
21.43082	201.2	3.24	8.01	226.8	5.56
21.50191	202.6	3.20	8.03	258.2	6.39
22.34802	198.2	3.08	8.03	235.9	6.07
23.32914	201.8	3.24	8.05	267.6	6.53
24.69593	202.1	3.20	8.01	250.5	6.22

Table A.26: *The effect of moisture of chitosan powder on the tensile strength of tablets: 9.39% moisture.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
14.91444	197.5	3.99	8.06	108.7	2.15
14.93505	200.8	4.07	8.05	105.8	2.05
15.35566	198.8	3.79	8.07	84.2	1.75
15.69325	194.2	3.44	8.01	139.7	3.23
15.80605	195.0	3.44	8.01	149.8	3.46
15.81956	197.4	3.43	8.00	154.0	3.57
15.98923	199.1	3.34	8.00	195.7	4.66
16.16021	194.7	3.29	8.00	165.1	3.99
16.55865	192.7	3.27	8.00	178.2	4.33
17.02451	196.1	3.14	8.00	240.7	6.10
17.09144	196.9	3.20	8.00	225.6	5.61
17.50035	193.3	3.11	7.99	238.2	6.10
17.90908	192.2	2.97	7.98	272.5	7.32
18.01508	196.0	3.06	7.98	262.7	6.85
18.25186	192.5	3.01	7.99	277.4	7.34
19.20426	199.4	3.05	7.99	310.1	8.10
19.31658	195.3	2.96	7.99	315.4	8.49
20.07455	195.0	3.02	8.02	300.3	7.89
20.37200	197.8	3.05	7.99	315.0	8.23
20.40690	197.2	3.02	7.99	315.9	8.33
20.80310	193.7	2.95	7.99	311.0	8.40

Table A.27: *The effect of moisture of chitosan powder on the tensile strength of tablets: 11.86% moisture.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.46253	200.6	3.98	8.08	74.4	1..47
15.68746	196.7	3.81	8.03	80.5	1.67
15.98299	196.8	3.79	8.04	87.0	1.82
16.31689	196.0	3.19	7.99	158.7	3.96
16.40350	194.2	3.29	8.02	165.1	3.98
16.40787	201.2	3.33	8.00	172.8	4.13
17.24193	199.1	3.17	7.99	208.4	5.24
17.47993	195.1	3.16	8.00	196.5	4.95
17.63690	196.5	3.19	8.00	204.3	5.09
18.23085	192.3	3.02	8.00	258.7	6.81
18.85121	195.2	3.04	7.99	252.5	6.62
18.89502	195.7	3.03	7.99	268.8	7.01
19.06126	194.0	3.00	7.99	278.7	7.40
19.33474	199.3	3.11	7.99	299.1	7.66
19.65520	197.1	3.07	7.99	294.2	7.63
20.13680	195.0	3.04	7.99	297.9	7.80
20.16114	193.0	2.98	8.01	290.7	7.75

Table A.28: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 150 mg chitosan < 90 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.56321	149.5	3.15	8.03	47.8	1.20
15.66209	147.6	3.06	8.03	51.5	1.33
15.63906	152.5	3.17	8.03	53.1	1.33
15.63459	147.9	3.08	8.01	50.7	1.44
15.70226	149.0	3.10	8.02	50.7	1.30
16.31262	147.9	2.67	8.02	86.2	2.56
15.83073	151.4	2.70	8.02	96.8	2.84
16.26390	148.0	2.68	8.01	89.5	2.65
16.50657	150.4	2.73	8.01	98.1	2.85
15.88811	146.1	2.66	8.00	94.4	2.93
16.29036	144.4	2.48	7.99	112.8	3.62
16.70159	147.3	2.57	7.99	116.0	3.59
16.58302	149.5	2.59	7.99	126.7	3.90
16.50577	149.0	2.56	7.99	121.8	3.79
16.53593	147.6	2.57	7.99	120.1	3.72

Table A.29: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 175 mg chitosan < 90 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.73426	173.1	3.72	8..05	49.0	1.04
15.99061	172.0	3.90	8.04	39.6	1.01
15.88080	172.9	3.78	8.04	39.6	1.01
15.39685	172.5	3.70	8.02	52.7	1.13
15.44424	173.5	3.73	8.04	56.4	1.20
15.90645	166.9	3.14	8.01	93.2	2.36
16.10509	174.7	3.20	8.01	114.8	2.75
16.18730	173.5	3.15	8.01	109.5	2.76
15.72481	174.0	3.17	8.00	122.2	3.07
16.04658	170.0	3.02	8.01	110.3	2.90
16.32487	173.9	2.98	8.00	133.2	3.86
16.70989	173.5	3.02	8.01	144.6	3.80
16.46620	172.8	2.97	7.99	135.7	3.64
16.79609	175.5	3.05	7.99	133.2	3.48
16.36072	174.3	3.04	8.00	147.9	3.87

Table A.30: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 200 mg chitosan < 90 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.19513	196.6	4.42	8.04	48.2	0.86
15.74762	197.8	4.39	8.04	52.3	0.84
15.36194	196.9	4.41	8.04	49.4	0.89
15.83545	196.7	4.34	8.04	57.6	1.05
15.63835	198.7	4.38	8.02	57.2	1.04
16.30489	197.8	3.62	8.03	117.3	2.57
16.00161	197.7	3.63	8.01	127.5	2.79
16.31678	199.0	3.70	8.02	124.6	2.67
16.29846	192.8	3.46	8.01	128.3	2.95
16.40845	198.7	3.57	8.00	137.3	3.06
16.95276	199.4	3.48	8.03	154.0	3.51
16.47579	199.5	3.47	8.02	163.4	3.74
17.16922	201.1	3.52	8.03	154.5	3.48
17.05534	200.8	3.42	8.00	166.9	3.88
16.56907	199.9	3.42	8.00	170.8	3.97

Table A.31: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 150 mg chitosan > 212 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.74653	150.7	3.26	8.03	38.4	0.93
15.48916	151.1	3.27	8.04	47.4	1.15
15.63823	150.8	3.23	8.03	47.0	1.15
15.77285	151.7	3.22	8.03	51.1	1.26
15.43161	150.6	3.12	8.03	52.3	1.33
16.22508	150.6	2.70	8.01	89.5	2.63
16.09585	151.0	2.82	8.01	89.9	2.53
16.45999	152.5	2.80	8.02	93.6	2.65
16.16095	152.6	2.78	8.00	86.6	2.48
16.22059	152.2	2.77	8.01	94.4	2.71
17.26252	154.2	2.65	8.00	123.8	3.72
16.33406	149.3	2.57	8.00	113.6	3.30
17.16025	149.3	2.59	7.98	120.5	3.71
17.15249	151.2	2.58	7.99	110.3	3.40
16.19683	149.9	2.57	8.00	113.6	3.21

Table A.32: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 175 mg chitosan > 212 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
16.05221	175.8	3.94	8.04	54.9	1.10
15.25829	176.5	3.82	8.03	53.5	1.11
15.31302	176.3	3.88	8.07	50.3	1.02
15.71220	175.5	3.77	8.04	49.9	1.05
15.16130	175.4	3.87	8.05	56.8	1.16
16.38524	173.6	3.23	8.03	107.5	2.64
16.32143	174.4	3.21	8.02	102.2	2.53
16.12774	174.0	3.20	8.01	97.7	2.43
16.21472	175.7	3.19	8.01	100.1	2.49
16.41417	174.3	3.24	8.01	107.1	2.63
16.62777	176.6	3.04	8.03	121.4	3.16
16.82086	175.6	3.02	8.01	141.4	3.72
16.83010	177.1	3.05	8.00	133.2	3.50
16.48027	174.6	2.97	8.00	131.2	3.51
16.48131	176.5	3.05	8.00	150.8	3.93

Table A.33: *The effect of powder weight of chitosan powder on the tensile strength of tablets: 200 mg chitosan > 212 µm.*

Compression force (bar)	Weight (mg)	Thickness (mm)	Diameter (mm)	Hardness (N)	Tensile strength (N/mm^2)
15.64513	199.7	4.59	8.07	49.0	0.84
15.62967	200.7	4.50	8.05	58.4	1.03
15.11246	198.4	4.46	8.05	58.8	1.04
15.67065	199.7	4.35	8.05	62.1	1.13
15.25390	198.5	4.38	8.04	62.1	1.12
16.18253	200.1	3.71	8.02	119.7	2.56
16.27448	199.1	3.71	8.03	112.4	2.40
16.46416	201.5	3.71	8.01	116.5	2.49
16.08239	199.5	3.74	8.02	116.0	2.46
16.00423	200.0	3.66	8.02	137.3	2.98
16.80484	201.5	3.53	8.02	159.8	3.59
16.57151	200.5	3.50	8.01	163.4	3.71
16.47764	201.5	3.48	8.03	153.6	3.50
16.37490	196.3	3.38	8.01	145.1	3.41
16.56882	200.6	3.45	8.01	158.1	3.66

Table A.34: *The effect of talc (0%) on the tensile strength of chitosan tablets.*

Compression force (bar)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
15.96151	2.98	8.03	58.0	1.54
15.92333	2.97	8.02	53.5	1.43
15.99811	3.02	8.03	59.7	1.57
15.59077	2.92	8.03	58.4	1.58
16.30464	3.07	8.03	53.9	1.39
17.76108	2.57	8.01	120.5	3.73
17.25157	2.42	8.00	96.0	3.16
17.49757	2.56	8.02	105.8	3.28
17.14985	2.60	8.02	103.4	3.16
17.43215	2.31	8.01	94.0	3.23
18.45168	2.42	8.02	158.5	5.20
18.86093	2.17	8.02	136.9	5.01
17.99206	2.44	8.03	150.8	4.90
18.15536	2.42	8.02	152.0	4.98
18.67692	2.45	8.02	143.0	4.63

Table A.35: *The effect of talc (0.5%) on the tensile strength of chitosan tablets.*

Compression force (bar)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
16.03890	2.95	8.03	49.4	1.33
15.92326	3.04	8.02	62.5	1.63
15.94901	3.06	8.04	60.1	1.55
16.32875	3.07	8.05	51.1	1.32
16.25244	3.07	8.03	51.1	1.32
17.01199	2.63	8.01	103.8	3.14
17.25157	2.60	8.04	101.3	3.08
17.49757	2.61	8.02	96.8	2.94
17.14985	2.60	8.03	98.9	3.01
17.43215	2.64	8.03	102.2	3.07
18.77476	2.49	8.04	149.6	4.76
17.82656	2.47	8.03	142.6	4.58
18.51889	2.52	8.04	151.6	4.76
17.71528	2.54	8.04	147.5	4.60
17.68946	2.48	8.03	142.2	4.54

Table A.36: *The effect of talc (0.75%) on the tensile strength of chitosan tablets.*

Compression force (bar)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
16.02712	3.04	8.02	51.5	1.34
16.14354	3.05	8.04	62.4	1.62
16.03218	3.03	8.04	57.6	1.50
16.42897	3.02	8.03	58.4	1.53
15.96072	3.05	8.02	48.6	1.26
17.24774	2.59	8.05	100.1	3.06
17.23752	2.59	8.04	103.8	3.17
17.35005	2.61	8.04	107.5	3.26
17.04007	2.47	8.03	97.2	3.12
17.10186	2.62	8.04	103.8	3.14
18.24299	2.45	8.02	158.9	5.15
18.14633	2.44	8.03	149.1	4.84
18.59067	2.44	8.03	147.1	4.78
18.18329	2.44	8.01	160.7	5.23
18.17580	2.44	8.00	151.6	4.94

Table A.37: *The effect of talc (1.0%) on the tensile strength of chitosan tablets.*

Compression force (bar)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
16.17970	3.07	8.03	44.9	1.16
16.12265	3.07	8.04	53.5	1.38
16.14488	3.00	8.02	47.0	1.24
16.32685	3.06	8.01	47.8	1.24
15.82018	3.06	8.01	56.2	1.46
16.84974	2.62	8.03	104.2	3.15
17.00567	2.61	8.02	111.6	3.39
16.70114	2.52	8.03	97.7	3.07
16.94377	2.49	8.03	102.6	3.27
16.77442	2.55	8.04	95.2	2.95
18.55916	2.41	7.99	145.1	4.80
18.44156	2.39	8.00	146.7	4.88
17.88948	2.41	8.02	141.0	4.64
18.41422	2.47	8.03	141.4	4.54
17.88847	2.45	7.99	144.6	4.70

Table A.38: *The effect of compression force on the disintegration time of chitosan tablets.*

Compression force (bar)	Tablet weight (mg)	Disintegration time (min)
16.53287	146.6	5.03
16.19140	149.4	2.17
16.41147	149.6	4.33
16.27154	148.3	3.83
16.02618	149.4	1.50
16.43819	149.1	1.83
Avg = 16.31194		Avg = 3.12
17.88503	147.2	241.47
17.20121	147.3	194.00
16.89011	150.4	150.25
16.94229	150.1	175.00
17.08261	150.0	29.58
17.35134	148.5	118.33
Avg = 17.22542		Avg = 133.43
18.41270	153.5	447.00
18.76015	154.2	571.47
18.50980	150.5	340.00
19.14630	150.3	600.75
18.29721	151.1	640.08
18.15234	149.9	598.00
Avg = 18.54642		Avg = 566.22

Table A.39: *The effect of punch depth on the tensile strength of chitosan minitablets. Setting 30.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
23.2	1.82	3.93	32.3	2.87
23.7	1.83	3.94	36.4	3.21
23.1	1.82	3.95	30.6	2.71
23.9	1.83	3.95	36.8	3.24
22.3	1.80	3.94	28.2	2.53
24.1	1.85	3.95	38.0	3.31

Table A.40: *The effect of punch depth on the tensile strength of chitosan minitablets. Setting 32.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
23.5	1.63	3.94	57.2	5.67
24.0	1.67	3.95	57.6	5.56
23.4	1.67	3.95	51.1	4.93
22.3	1.61	3.95	53.9	5.40
22.0	1.58	3.94	53.1	5.43
23.0	1.64	3.95	55.2	5.42

Table A.41: *The effect of punch depth on the tensile strength of chitosan minitablets. Setting 34.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
22.9	1.63	3.94	56.8	5.63
22.2	1.58	3.94	53.9	5.51
23.0	1.63	3.95	58.4	5.77
21.7	1.55	3.94	54.3	5.66
22.7	1.61	3.95	55.6	5.57
22.5	1.60	3.95	57.2	5.76

Table A.42: *The effect of punch depth on the tensile strength of chitosan minitablets. Setting 36.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
22.3	1.63	3.98	59.2	5.81
22.4	1.61	3.96	52.3	5.22
22.2	1.61	3.98	58.0	5.76
22.9	1.68	3.98	62.9	5.99
22.4	1.63	3.96	55.6	5.48
22.0	1.63	3.97	58.4	5.75

Table A.43: *The effect of punch depth on the tensile strength of chitosan minitablets. Setting 38.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
20.4	1.53	3.97	47.4	4.97
22.4	1.65	3.99	59.7	5.77
21.1	1.60	3.99	60.5	6.03
22.0	1.60	3.97	63.7	6.38
22.3	1.63	3.99	58.8	5.76
21.3	1.58	3.97	57.6	5.85

Table A.44: *Dissolution of isoniazide from chitosan minitablets pressed at different punch depths.*

Time (min)	Dissolved Setting 30 (%)	Dissolved Setting 32 (%)	Dissolved Setting 34 (%)	Dissolved Setting 36 (%)	Dissolved Setting 38 (%)
0	0	0	0	0	0
2	56.07	11.98	14.25	13.03	17.44
5	80.87	53.47	56.05	52.46	46.20
10	89.16	82.13	85.41	85.96	73.42
15	89.21	90.86	94.20	97.11	80.30
20	93.37	93.52	96.50	100.20	86.26
30	93.30	94.46	99.18	101.97	90.29
45	94.90	95.43	100.09	102.38	94.34
60	94.33	95.35	100.51	101.32	97.80
120	96.37	95.74	100.30	101.02	97.04

Table A.45: *The effect of 0% compaction on the tensile strength of chitosan minitablets.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
22.0	1.62	3.97	59.2	5.86
21.8	1.58	3.97	58.4	5.93
21.7	1.57	3.96	54.3	5.56
22.0	1.59	3.98	58.4	5.88
22.1	1.61	3.97	58.0	5.78
21.8	1.58	3.96	56.4	5.74

Table A.46: *The effect of 20% compaction on the tensile strength of chitosan minitablets.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
26.8	1.88	3.98	65.4	5.56
23.9	1.72	3.98	65.4	6.09
25.9	1.83	3.98	67.4	5.89
26.5	1.84	3.98	75.6	6.57
26.0	1.85	3.98	67.4	5.83
26.2	1.83	3.99	69.5	6.06

Table A.47: *The effect of 40% compaction on the tensile strength of chitosan minitablets.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
30.1	2.11	3.98	76.8	5.82
30.0	2.08	3.98	78.0	6.00
29.7	2.05	3.97	84.6	6.62
30.1	2.09	3.97	80.1	6.15
31.0	2.11	3.99	94.4	7.14
30.7	2.11	3.98	83.8	6.35

Table A.48: *The effect of 60% compaction on the tensile strength of chitosan minitablets.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
36.1	2.43	4.00	106.6	6.98
36.4	2.43	3.98	101.3	6.67
36.1	2.42	3.98	100.9	6.67
36.4	2.42	4.00	114.0	7.50
36.2	2.44	3.99	102.6	6.71
35.7	2.41	4.00	103.0	6.80

Table A.49: *Dissolution of isoniazide from chitosan minitablets compressed at different compaction percentages.*

Time (min)	Isoniazide dissolved (%) No compaction	Isoniazide dissolved (%) 20% compaction	Isoniazide dissolved (%) 40% compaction	Isoniazide dissolved (%) 60% compaction
0	0	0	0	0
2	14.25	16.13	16.30	20.93
5	56.05	37.30	35.22	40.45
10	85.41	65.68	66.48	70.70
15	94.20	81.02	82.01	85.75
20	96.50	89.37	88.93	93.65
30	99.18	94.76	92.70	97.30
45	100.09	97.83	95.92	97.16
60	100.51	98.58	96.06	97.56
120	100.30	98.62	98.32	98.94

Table A.50: T*he tensile strength of chitosan minitablets containing no citric acid.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
153.3	2.61	8.02	114.0	3.47
145.8	2.44	8.02	107.5	3.50
150.5	2.56	8.02	101.7	3.15
151.2	2.56	8.01	109.1	3.39
153.2	2.60	8.01	108.3	3.31
150.8	2.54	8.00	105.6	3.31

Table A.51: *The tensile strength of chitosan minitablets containing 2% citric acid.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
152.4	2.67	8.06	92.8	2.75
152.9	2.65	8.04	92.8	2.77
151.3	2.57	8.01	106.9	3.31
152.0	2.56	8.01	108.9	3.38
153.3	2.65	8.04	98.9	2.96
151.8	2.59	8.01	105.0	3.22

Table A.52: *The tensile strength of chitosan minitablets containing 4% citric acid.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
153.7	2.64	8.04	97.7	2.93
152.8	2.64	8.04	90.7	2.72
152.0	2.64	8.03	89.5	2.69
150.8	2.61	8.04	87.9	2.67
151.5	2.61	8.03	94.4	2.87
151.4	2.64	8.05	96.2	2.88

Table A.53: *The tensile strength of chitosan minitablets containing 6% citric acid.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
152.5	2.61	8.02	100.5	3.06
153.8	2.60	8.01	93.6	2.86
150.7	2.55	8.02	89.2	2.78
149.6	2.56	8.02	95.4	2.96
153.6	2.62	8.02	98.1	2.97
154.4	2.62	8.03	90.6	2.74

Table A.54: *The tensile strength of chitosan minitablets containing 8% citric acid.*

Tablet weight (mg)	Thickness (mm)	Diameter (mm)	Crushing strength (N)	Tensile strength (N/mm^2)
151.0	2.62	8.03	92.3	2.79
151.8	2.60	8.03	89.9	2.74
150.7	2.60	8.03	88.3	2.69
150.4	2.58	8.01	95.2	2.93
154.2	2.68	8.03	93.6	2.77
150.4	2.60	8.01	89.1	2.72

Table A.55: *Dissolution of isoniazide from chitosan minitablets containing different amounts of citric acid.*

Time (min)	Isoniazide dissolved (%) No citric acid	Isoniazide dissolved (%) 2% citric acid	Isoniazide dissolved (%) 4% citric acid	Isoniazide dissolved (%) 8% citric acid
0	0	0	0	0
2	1.89	2.99	6.09	4.06
5	39.77	16.16	10.01	6.36
10	79.10	39.99	29.27	20.71
15	94.18	58.85	43.79	33.94
20	97.28	70.28	56.43	44.67
30	99.05	82.96	71.72	60.38
45	98.84	91.88	84.45	77.41
60	98.32	95.30	92.09	88.35
120	99.16	97.01	98.22	97.79

Table A.56: *Dissolution of isoniazide from chitosan minitablets containing different amounts of pectin.*

Time (min)	Isoniazide dissolved (%) No pectin	Isoniazide dissolved (%) 2% pectin	Isoniazide dissolved (%) 4% pectin	Isoniazide dissolved (%) 8% pectin
0	0	0	0	0
2	1.89	5.47	2..71	3.99
5	39.77	10.62	4.92	6.91
10	79.10	41.09	15.89	13.19
15	94.18	61.95	39.40	20.67
20	97.28	74.82	57.21	33.42
30	99.05	87.76	75.55	53.78
45	98.84	95.66	88.06	74.48
60	98.32	98.29	94.27	84.85
120	99.16	100.16	97.88	95.64

Table A.57: *Dissolution of isoniazide from chitosan minitablets coated with different amounts of Eudragit S®.*

Time (min)	Isoniazide dissolved (%) No Eudragit S®	Isoniazide dissolved (%) 3.5% Eudragit S®	Isoniazide dissolved (%) 6.7% Eudragit S®
0	0	0	0
2	14.34	0.46	0.03
5	17.19	0.95	0.03
10	21.87	2.14	0.14
15	26.20	3.57	1.13
20	30.08	5.18	2.41
30	36.95	8.63	5.50
45	50.13	15.37	10.80
60	62.02	23.11	15.82
120	89.82	59.22	38.95

Table A.58: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the punch depth.*

	Setting 30	Setting 32	Setting 34	Setting 36	Setting 38
MDT1	**3.66**	**6.15**	**7.82**	7.18	9.39
MDT2	5.40	6.04	6.06	7.30	8.65
MDT3	5.70	7.20	5.05	7.23	9.11
Avg MDT	4.92	6.46	6.31	7.24	9.05
SD	1.11	0.64	1.40	0.06	0.37

Table A.59: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets as a function of the percentage compaction.*

	0 %	20%	40%	60%
MDT 1	7.82	10.13	12.88	11.16
MDT 2	6.06	9.32	8.11	7.73
MDT3	5.05	8.96	11.28	7.62
Avg MDT	6.31	9.47	10.76	8.84
SD	1.40	0.60	2.43	2.01

Table A.60: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of citric acid.*

	0%	2%	4%	8%
MDT 1	7.00	15.21	22.32	30.15
MDT 2	6.94	18.35	24.49	29.00
MDT3	9.01	17.33	25.86	30.68
Avg MDT	7.65	16.96	24.22	29.94
SD	1.18	1.61	1.78	0.86

Table A.61: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from chitosan minitablets containing different amounts of pectin.*

	0%	2%	4%	8%
MDT 1	7.00	16.56	24.57	31.55
MDT 2	6.94	17.51	24.57	33.21
MDT3	9.01	15.79	22.49	34.96
Avg MDT	7.65	16.62	23.88	33.24
SD	1.18	0.86	1.20	1.71

Table A.62: *The mean dissolution time (MDT) of the dissolution profiles of isoniazide from minitablets coated with different amounts of Eudragit S®.*

	0%	3.5%	6.7%
MDT 1	45.86	70.92	69.31
MDT 2	42.97	68.83	63.07
MDT3	43.88	65.43	72.04
Avg MDT	44.24	68.40	68.31
SD	1.48	2.75	4.67

ANNEXURE B

S.A. PATENT NO: 2006/04483

APPARATUS FOR ASSESSING FLOWABILITY OF A NON-GASEOUS FLUID

INTRODUCTION AND BACKGROUND TO THE INVENTION

This invention relates to apparatus for assessing flowability of a non-gaseous fluid and a method of assessing flowability of a non-gaseous fluid by using such apparatus.

Known methods of assessing the flowability of a powder include angle of response; critical orifice diameter; Carr's percent compressibility index; and Hausner's ratio.

According to the angle of response method, a body of powder is contained in a container and allowed to pass through a hole defined in the bottom of the container onto a horizontal surface. A heap of powder is formed below the hole of the container on the surface. The angle between the side of the heap and the surface is known as the angle of response. A smaller angle represents a better flow of powder.

In the critical orifice diameter method, a body of powder is again contained in a container and allowed to pass through a hole defined in the bottom of the container. However, the diameter of the hole is varied until the hole with the smallest diameter through which the powder can still pass is found. The diameter of this hole is the critical orifice diameter. Flowability of the powder is therefore represented by the orifice diameter of the smallest opening through which the powder falls freely. Smaller values indicate better flow.

The Hausner ratio and the Carr index, which are measures of inter-particle friction and potential powder arch or bridge strength and stability, respectively, have been widely used to estimate the flow properties of powders. The Carr's percent compressibility and the Hausner ratio methods are calculated using the equations $([\rho_{tap}-\rho_{bulk}]/\rho_{tap})x100$ and ρ_{tap}/ρ_{bulk}, respectively, wherein p is density. The "bulk" and "tap" densities are determined by pouring a known mass of powder through a funnel into a graduated cylinder. The volume of powder is then read directly from the cylinder and used to calculate the "bulk" density. The "tap" density is determined by using a tap density tester.

A Hausner ratio value of less than 1.20 is indicative of good flowability of the powder, whereas a value of 1.5 or higher suggests a poor flowability of the powder. The Carr index is also called "percent compressibility" and a value between 5 and 15, 12 and 16, 18 and 21, and 23 and 28 indicates excellent, good, fair, and poor flow properties of the material, respectively. Lower values therefore represent better flow.

US patent number 4,274,286 discloses an apparatus for measuring the flowability of powder. The apparatus comprises a container in the form of a cylinder; a disc defining a central hole and which is located at the bottom end of the container; and a removable plate which closes the hole. The disc is replaceable with discs having holes of different diameters. The cylinder has an inside diameter which is larger than the diameter of the largest hole. The apparatus is used to measure the critical orifice diameter of a powder, as described above.

In the case of the prior art apparatus, including the apparatus of US 4,274,286, the diameter of the cylinder shaped container is larger than that of the largest hole. A first

disadvantage of these apparatus is therefore that in regions between the inner walls of the cylinder and the hole in the disc, powder does not flow. Static regions are thus formed where the powder is stationary. Friction is created between the stationary powder and the flowing powder in these regions. This friction restricts the flow of the powder through the hole and measurements are therefore not accurate.

A second disadvantage of prior art apparatus is that air pockets are formed in the body of powder contained in the container. These air pockets affect the flowability of the powder and produce inaccurate measurements.

OBJECT OF THE INVENTION

It is therefore an object of the present invention to provide apparatus for assessing flowability of a non-gaseous fluid and a method of assessing flowability of a non-gaseous fluid by using such apparatus with which the aforesaid disadvantages can be overcome or at least minimised.

SUMMARY OF THE INVENTION

According to a first aspect of the invention there is provided apparatus for assessing the flowability of a non-gaseous fluid comprising:

- a plurality of bodies being stackable one on top of the other, each having a bore with an inlet and an outlet, which is relatively smaller than the inlet, such that each bore tapers from the inlet to the outlet, the bores further being aligned with each other when the bodies are stacked and the outlet of the bore of one body being the same size as the inlet of the bore of the body directly below it; and

- a passage provided by the bores of the bodies, for passing the fluid from a passage inlet at an upper end of the apparatus to a passage outlet at a lower end of the apparatus,

the arrangement being such that the size of the passage outlet is variable by successively removing bodies from the passage outlet of the apparatus.

Each bore may pass through the centre of each body.

Each body may be an annular disc.

Each bore may be circular, the arrangement being such that the passage is in the form of a cone.

The bodies may each define a retaining formation, the arrangement being such that the retaining formations of abutting bodies mate with one another to retain the bores of the bodies in alignment.

The retaining formation of abutting bodies may be complementary.

The apparatus may include a first container for containing the non-gaseous fluid and which may be connected to the passage inlet.

The first container may be in the form of a hopper.

The hopper may have a hopper inlet and a hopper outlet.

The diameter of the hopper outlet may be the same as the diameter of the passage inlet.

The apparatus may further include a container for containing fluid.

The container may be cylindrically shaped and may be provided towards the inlet of the hopper.

The diameter of the container may be the same as the diameter of the inlet of the hopper.

The bodies may be made from a material which does not provide electrostatic charges, such as glass, metal and metal alloys, more specifically brass.

Each body may be from 1 mm to 20 mm thick, preferably from 8 mm to 15 mm.

The diameter of the bores may range from 0.01 mm to 300 mm.

The apparatus may further include a stand on which the bodies are provided.

The stand may define an opening, the arrangement being such that the opening is aligned with the passage outlet.

The stand may further include a shutter for opening and closing the opening.

According to a second aspect of the invention there is provided a method of assessing the flowability of a non-gaseous fluid by using an apparatus according to the first aspect of the invention.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention will now be described further by way of a non-limiting example with reference to the accompanying drawings wherein:

figure 1 is a longitudinal sectional side view of an apparatus for assessing the flowability of a non-gaseous fluid according to a preferred embodiment of the invention;

figure 2 is an exploded perspective view of a plurality of bodies of the apparatus of figure 1; and

figure 3 is a side view of the apparatus of figure 1, in use.

DESCRIPTION OF A PREFERRED EMBODIMENT OF THE INVENTION

Referring to figures 1 and 3, an apparatus for assessing the flowability of a non-gaseous fluid 19 according to a preferred embodiment of the invention is generally designated by reference numeral 10.

The apparatus 10 comprises a plurality of bodies 12 being stacked one on top of the other. Each body 12 has a bore 14 with an inlet 14.1 and an outlet 14.2, which is relatively smaller than the inlet 14.1. The arrangement is such that each bore 14 tapers from the inlet 14.1 to the outlet 14.2. The bores 14 of the bodies 12 are aligned with one another and the outlet 14.2 of the bore 14 of one body 12 is the same size as the inlet 14.1 of the bore 14 of the body 12 directly below it. A passage 16 for passing the fluid

19 from a passage inlet 16.1, towards an upper end of the apparatus 10, to a passage outlet 16.2, towards a lower end of the apparatus 10, is provided by the bores 14 of the bodies 12. The arrangement is such that the size of the passage outlet 16.2 is variable by successively removing bodies 12 from the passage outlet 16.2 of the apparatus 10.

The bodies 12 are in the form of annular discs and the bores 14 are circular and pass through the centre of each body 12. The passage 16 is thus in the form of a cone, as shown in figure 1. Each body 12 is provided with a retaining formation 18 around its periphery, so that abutting bodies 12 mate with one another. These formations 18 prevent the bores 14 of the bodies 12 from moving out of alignment with each other. The mating formations 18 of abutting bodies 12 are complementary.

The bodies 12 are made from brass and each body is from 8 mm to 15 mm thick. The diameter of each bore 14 ranges from 0.01 mm to 300 mm. The thickness of the body 12 depends on the desired diameter of the inlet 14.1 and outlet 14.2 of the bore 14. The desired size of the bore 14, in turn, depends on the type of non-gaseous fluid 18, of which the flowability is to be assessed by the apparatus 10.

A hopper 20 for containing the non-gaseous fluid 19, and having a hopper inlet 20.1 and a hopper outlet 20.2, is connected to the passage inlet 16.1 of the apparatus 10. The diameter of the hopper outlet 20.2 is the same as the diameter of the passage inlet 16.1. A cylindrically shaped container 22 for containing even more fluid 19 is provided towards the upper end of the hopper 20. The diameter of the container 20 is the same as the diameter of the hopper inlet 20.1.

The apparatus 10 further includes a stand 24 defining an opening 24.1 with a shutter (not shown) for opening and closing the opening 24.1 and thus the passage outlet 16.2. The bodies 12 rest on the stand 24 with the opening 24.1 aligned with the passage 16. Supports 26 are provided towards the lower end of the stand 24 to support the apparatus 10 in an upright position and away from a surface 28 on which it is positioned.

In use, the apparatus 10 is provided with the body having the bore 14 with the smallest outlet 14.2 diameter defining the passage outlet 16.2. The shutter is closed and a body of non-gaseous fluid 19, such as a powder, is provided in the container 22, hopper 20, and passage 16 of the apparatus 10. The shutter is then opened and the fluid 19 allowed to flow through the passage 16 towards the passage outlet 16.2 and onto the surface 28. If the passage outlet 16.2 is too small, the fluid 19 would not be able to flow through said passage outlet 16.2. The lower most body 12 is then removed so that the passage outlet 16.2 has a larger diameter. Bodies 12 are removed from the lower end of the apparatus 10 until the smallest bore 14 through which the fluid 19 can freely flow is determined. The diameter of the outlet 14.2 of this bore 14 is the critical orifice diameter, which represents the flowability of the fluid 19. A smaller critical orifice diameter value indicates better flow of the fluid.

It is foreseen that the apparatus could be used to determine the flowability of different types of non-gaseous fluid. A first advantage of the apparatus 10 is that no static regions are formed, where the fluid is stationary. A second advantage of the apparatus 10 is that air pockets are not formed in the body of fluid. Measurements taken by using the apparatus 10 are therefore relatively more accurate.

It will be appreciated that variations in detail are possible with apparatus for assessing powder flow and a method of assessing powder flow by using such apparatus according to the invention without departing from the scope of this disclosure.

ABSTRACT

This invention discloses an apparatus 10 for assessing the flowability of a non-gaseous fluid 19. The apparatus 10 comprises a plurality of bodies 12 being stacked one on top of the other and each having a bore 14 with an inlet 14.1 and an outlet 14.2. The outlet 14.2 is relatively smaller than the inlet 14.1 and each bore 14 tapers from the inlet 14.1 to the outlet 14.2. The outlet 14.2 of the bore 14 of one body 12 is the same size as the inlet 14.1 of the bore 14 of the body 12 directly below it. A passage 16 for passing the fluid 19 from a passage inlet 16.1 to a passage outlet 16.2 is provided by the bores 14 of the bodies 12. The arrangement is such that the size of the passage outlet 16.2 is variable by successively removing bodies 12 from the passage outlet 16.2 of the apparatus 10.

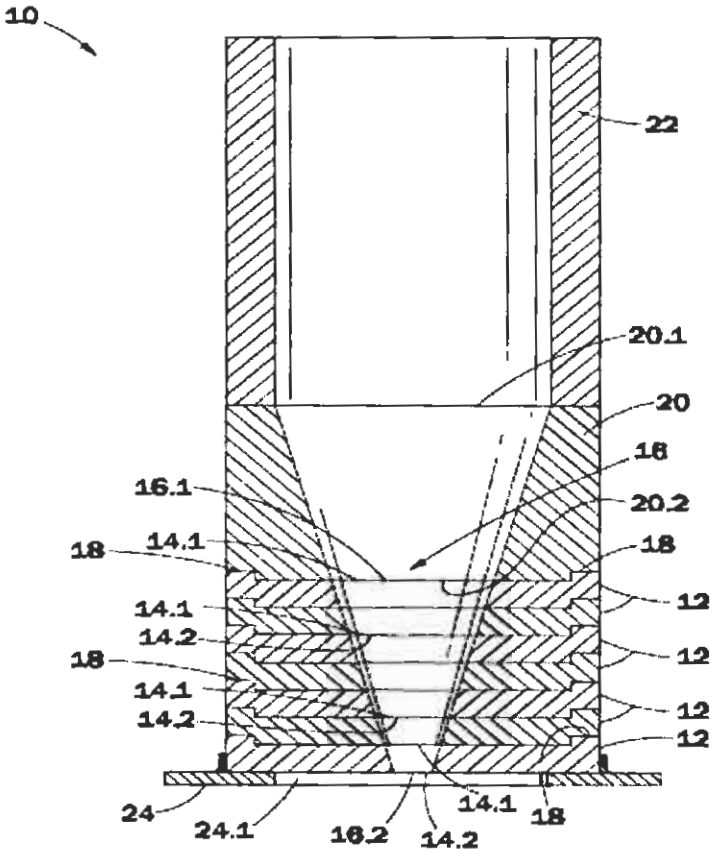


Figure 1

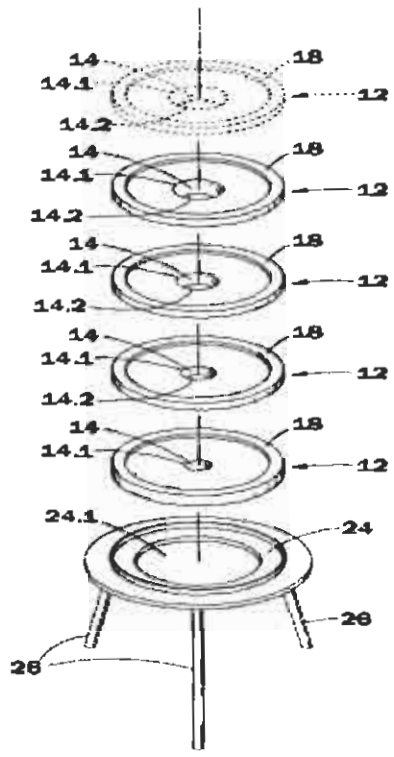


Figure 2

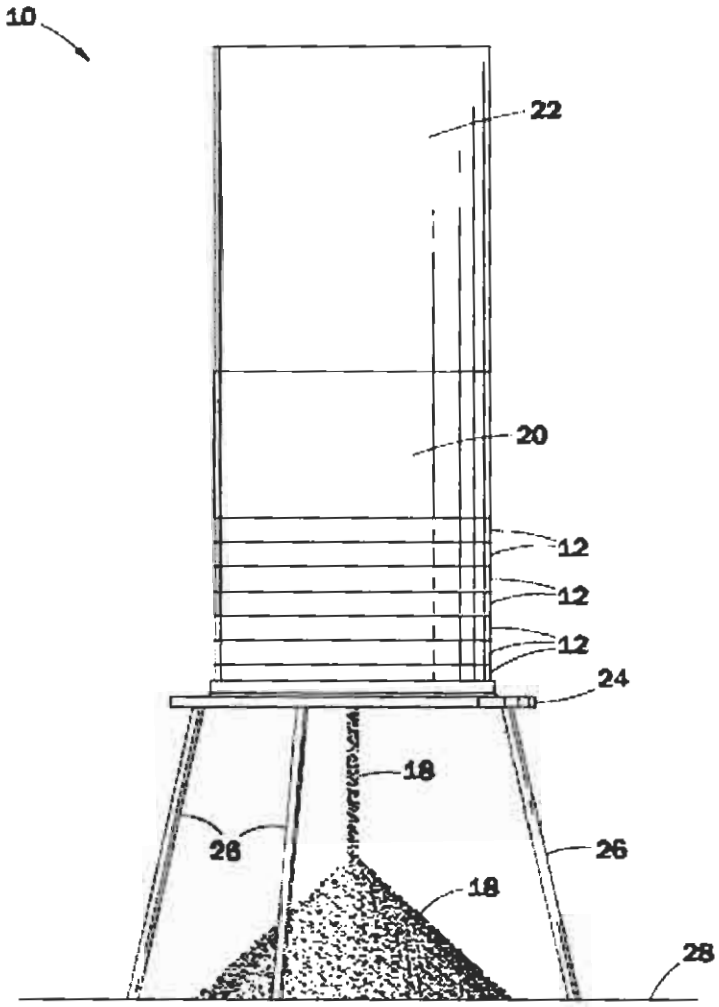


Figure 3

ANNEXURE C

Statistical tests on the dissolution profiles

Table 1: *ANOVA test on the dissolution profiles of isoniazide from the minitablets pressed at different punch depths.*

n	15

Machine setting	N	Mean	SD	SE
30	3	4.921	1.106	0.6385
32	3	6.465	0.643	0.3711
34	3	6.308	1.400	0.8083
36	3	7.235	0.058	0.0332
38	3	9.049	0.371	0.2141

Source of variation	SSq	DF	MSq	F	p
Machine setting	27.405	4	6.851	9.17	0.0022
Within cells	7.474	10	0.747		
Total	34.879	14			

		Tukey 95% CI		
Contrast	Difference			
30 v 32	-1.544	-3.867	to 0.779	
30 v 34	-1.387	-3.710	to 0.936	
30 v 36	-2.314	-4.637	to 0.009	
30 v 38	-4.129	-6.452	to -1.805	(significant)
32 v 34	0.157	-2.166	to 2.480	
32 v 36	-0.770	-3.093	to 1.553	
32 v 38	-2.584	-4.908	to -0.261	(significant)
34 v 36	-0.927	-3.250	to 1.396	
34 v 38	-2.742	-5.065	to -0.419	(significant)
36 v 38	-1.814	-4.137	to 0.509	

Table 2: *ANOVA test on the dissolution profiles of isoniazide from the minitablets pressed at different percentages compaction.*

n	12

Percentage compaction	n	Mean	SD	SE
0	3	6.310	1.402	0.8093
20	3	9.470	0.599	0.3460
40	3	10.757	2.428	1.4016
60	3	8.837	2.013	1.1621

Source of variation	SSq	DF	MSq	F	p
Percentage compaction	31.414	3	10.471	3.41	0.0732
Within cells	24.539	8	3.067		
Total	55.953	11			

		Tukey 95% CI	
Contrast	Difference		
0 v 20	-3.160	-7.739	to 1.419
0 v 40	-4.447	-9.026	to 0.133
0 v 60	-2.527	-7.106	to 2.053
20 v 40	-1.287	-5.866	to 3.293
20 v 60	0.633	-3.946	to 5.213
40 v 60	1.920	-2.659	to 6.499

Table 3: *ANOVA test on the dissolution profiles of isoniazide from the minitablets containing different percentages citric acid.*

n	12

Percentage citric acid	n	Mean	SD	SE
0	3	7.650	1.178	0.6802
2	3	16.963	1.602	0.9248
4	3	24.223	1.785	1.0306
8	3	29.943	0.859	0.4959

Source of variation	SSq	DF	MSq	F	p
Percentage citric acid	834.235	3	278.078	141.20	<0.0001
Within cells	15.755	8	1.969		
Total	849.990	11			

		Tukey 95% CI		
Contrast	Difference			
0 v 2	-9.313	-12.983	to -5.644	(significant)
0 v 4	-16.573	-20.243	to -12.904	(significant)
0 v 8	-22.293	-25.963	to -18.624	(significant)
2 v 4	-7.260	-10.929	to -3.591	(significant)
2 v 8	-12.980	-16.649	to -9.311	(significant)
4 v 8	-5.720	-9.389	to -2.051	(significant)

Table 4: *ANOVA test on the dissolution profiles of isoniazide from the minitablets containing different percentages pectin.*

n	12

Percentage pectin	n	Mean	SD	SE
0	3	7.650	1.178	0.6802
2	3	16.620	0.862	0.4974
4	3	23.877	1.201	0.6933
8	3	33.240	1.705	0.9845

Source of variation	SSq	DF	MSq	F	p
Percentage pectin	1061.377	3	353.792	218.38	<0.0001
Within cells	12.960	8	1.620		
Total	1074.337	11			

		Tukey 95% CI		
Contrast	Difference			
0 v 2	-8.970	-12.298	to -5.642	(significant)
0 v 4	-16.227	-19.555	to -12.899	(significant)
0 v 8	-25.590	-28.918	to -22.262	(significant)
2 v 4	-7.257	-10.585	to -3.929	(significant)
2 v 8	-16.620	-19.948	to -13.292	(significant)
4 v 8	-9.363	-12.691	to -6.035	(significant)

Table 5: *ANOVA test on the dissolution profiles of isoniazide from the minitablets coated with different amounts of Eudragit S®.*

n	9

Percentage Eudragit S	n	Mean	SD	SE
0	3	44.237	1.478	0.8531
3.5	3	68.403	2.755	1.5905
6.7	3	68.140	4.598	2.6547

Source of variation	SSq	DF	MSq	F	p
Percentage Eudragit S	1155.466	2	577.733	56.06	0.0001
Within cells	61.830	6	10.305		
Total	1217.296	8			

		Tukey 95% CI		
Contrast	Difference			
0 v 3.5	-24.167	-32.209	to -16.125	(significant)
0 v 6.7	-23.903	-31.945	to -15.861	(significant)
3.5 v 6.7	0.263	-7.779	to 8.305	

ANNEXURE D

Materials used in the study

Table D.1: *Materials used in the study.*

Material	Batch number	Manufacturer
Avicell PH200	M939C	FMC International, Cork, Ireland
Chitosan	021010	Warren Chem, Cape Town, RSA
Citric acid	1019688	Saarchem, Wadeville, RSA
Emcocell 50M	559553	Penwest, Patterson, USA
Emcocell 90M	959136	Penwest, Patterson, USA
Emcompress	8070	Penwest, Patterson, USA
Eudragit S 100	B030505062	Röhm GmbH, Darmstadt, Germany
Lithium chloride	1023975	Merck Chemicals, Saarchem, Wadeville, R.S.A.
Ludipress	25 - 0194	BASF, Ludwigshafen, Germany
Magnesium chloride	1019979	Merck Chemicals, Saarchem, Wadeville, R.S.A.
Magnesium nitrate	0133799	Merck Chemicals, Saarchem, Wadeville, R.S.A.
Pectin	67H16351	Sigma, St. Louis, USA
Prosolv SMCC50	P550010	Penwest, Patterson, USA
Prosolv SMCC90	P951027	Penwest, Patterson, USA
Sodium chloride	50345	Merck, Darmstadt, Germany
Talk	1036932	Saarchem, Wadeville, RSA

Figure D.1: *Certificate of analysis for the chitosan used in the study.*

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CERTIFICATE OF ANALYSIS FOR CHITOSAN

ORIGINAL

Report No.: 021010

Batch No.: 021010	Quantity: 1005kgs	Report Date : NOV. 05, 2002
Expiry date		Before NOV. 04, 2004.
ITEMS TESTED		INSPECTION RESULTS
Appearance		Off white
Moisture		5.93%
Ash		0.89%
Deacetylation		91.46%
Viscosity (0.5%)		21cps
Mesh		80mesh

厦门江都经济发展公司进出口事业部

AFRICA BANK LIMITED IB CENTRE CAPE TOWN
LETTER OF CREDIT NUMBER 827-01-0074500-G
"Drum" s No.: 68—134

WCI 7109