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Title: Preparing Sustained Release Dosage Form of Nifedipine by Hot-melt Coating Method

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A nifedipine sustained release dosage form was prepared by using hot-melt batch coating. Substrates are sugar beads, mesh size 30-35. Stearic acid, Acid Triglyceride, and carnauba wax were coating agents used to make single layer coated beads and multiple layer coated beads containing nifedipine. Dissolution of nifedipine from the beads and from capsules containing the beads was evaluated for 24 hours. Goodness of fit, total drug released, and "acceptance value" according to the "Acceptance Table" in USP25-NF20 were criteria used to evaluate dissolution profile *in vitro*. Convolution process was applied to obtain predicted plasma concentrations for comparison to published data for two well known commercial products: Adalat® CC and Procardia® XL 30mg.

In vitro test results showed the new hot-melt batch coating process is feasible to produce zero-order kinetics for drug dissolution. Capsules containing dual coated beads with carnauba wax in the inner coating layer and stearic acid in the outmost layer yielded similar dissolution profiles and predicted plasma concentrations to Procardia XL. However, drug quantity deposited on the sugar bead was low, thus it is necessary in the future to alter formulation to improve drug loading capacity.

Preparing sustained release dosage forms of nifedipine by hot-melt batch coating method

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Hang Thi Le, Author

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CHAPTER 1: INTRODUCTION

Controlled and sustained drug release dosage forms

Controlled release drug dosage forms are "therapeutic systems" in which the rate of drug release is programmed and controlled depending on product formulation designed for specific treatment targets. This means the drug release kinetics are predictable and reproducible. Controlled drug release dosage forms have become popular modern therapeutic drug delivery systems.

"Sustained release" is a narrow concept in the overall area of controlled drug release, which provides many advantages. First, sustained drug release dosage forms increase therapeutic efficiency. It is well documented that most drugs have a relatively stable therapeutic window (1); the concentration range, in which the pharmacologic response is reasonably effective, and toxicity is at an acceptable low value. More clearly, within this therapeutic window, the toxicity index is at a reasonably low value and the drug produces convincing clinical effects. The range of safe concentrations for a model drug is illustrated in Figure 1.

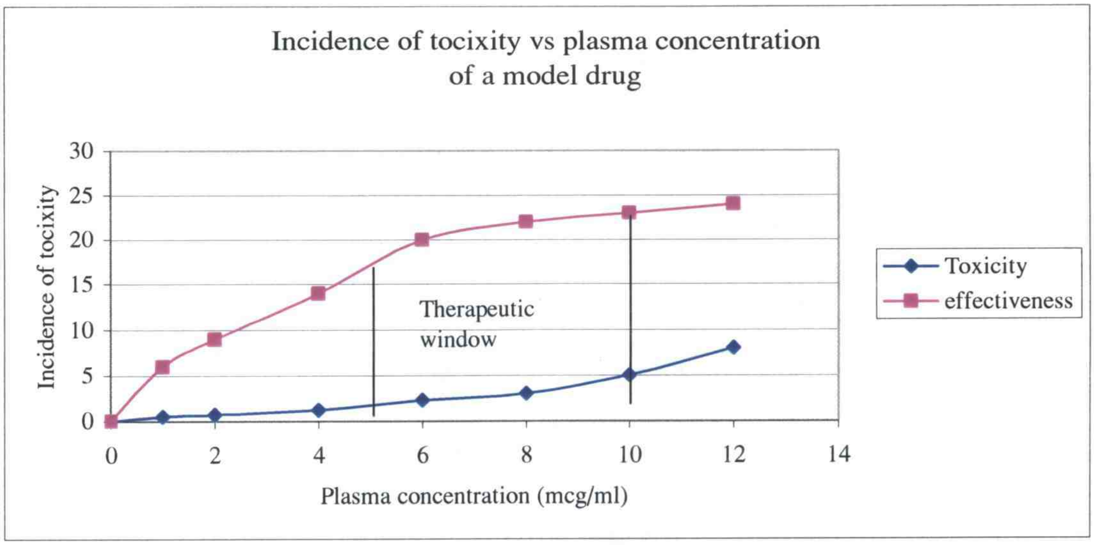


Figure 1: Relationship of Incidence of toxicity and plasma concentration

When drug is administered into the body plasma (and usually tissue) concentrations rise and then drop gradually with time. The dosing regimen chosen is a compromise between toxicity and effectiveness.

For oral drug administration, if a model drug is absorbed by a first-order kinetic process (immediate release product formulation) in the GI tract, and the one compartment model is fitted to the plasma concentration time curve, plasma concentration is calculated according to the formula:

$$C = \frac{k_{ab} F D}{(k_{el} - k_{ab}) V_d} (e^{-k_{ab} t} - e^{-k_{el} t}) \quad \text{(equation 1)}$$

where in F: absorption fraction
D: Dose
k_e is elimination rate constant,
k_{ab} is absorption rate constant
V_d: Volume distribution

The change in drug plasma concentration from equation 1 has the appearance indicated in Figure 2. If a sustained release dosage form of the same model drug is administered and the drug is released at a constant rate, for well absorbed drugs in the GI tract, the amount of drug released is equal to the amount of drug absorbed. Thus, the absorption profile is also constant and follows zero order kinetics. The expected concentration of drug in plasma can be described by equation 2 and is shown in Figure 3:

$$C = \frac{F k_0}{V_d k_{el}} (1 - e^{-k_{el} t}) \quad \text{(equation 2)}$$

Where in F: Absorption fraction
D: Dose
k_e: Elimination rate constant,
k_o: Absorption rate constant for zero order absoption
V_d: Volume distribution

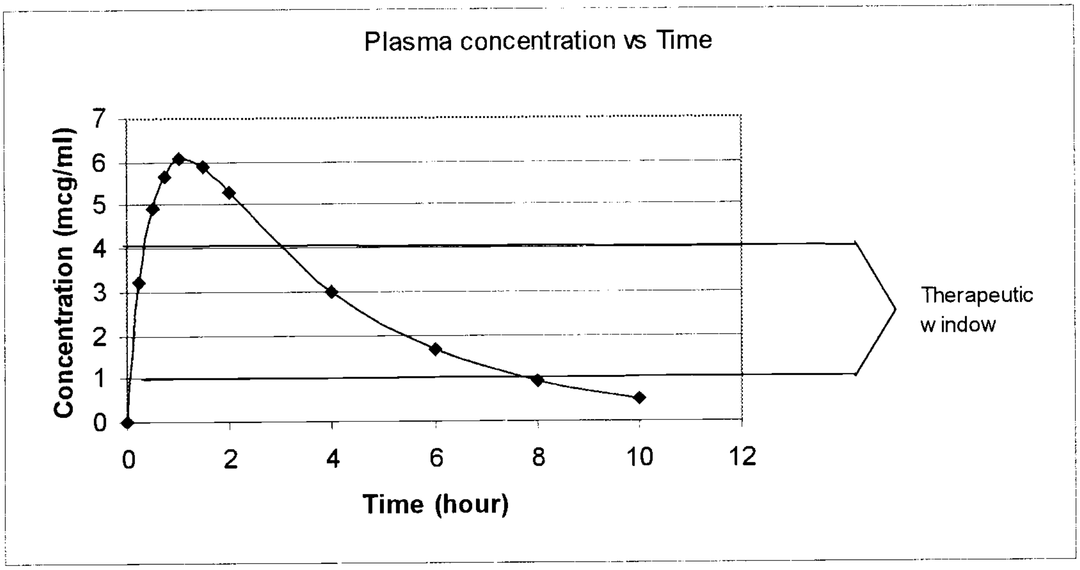


Figure 2: Plasma drug concentration of a model drug from an immediate release dosage form by oral administration, as predicted by equation 1.

Where:

$F = 1$ $K_{el} = 0.2$

$D=30mg$ $K_a = 2$

$V_d = 10$ L

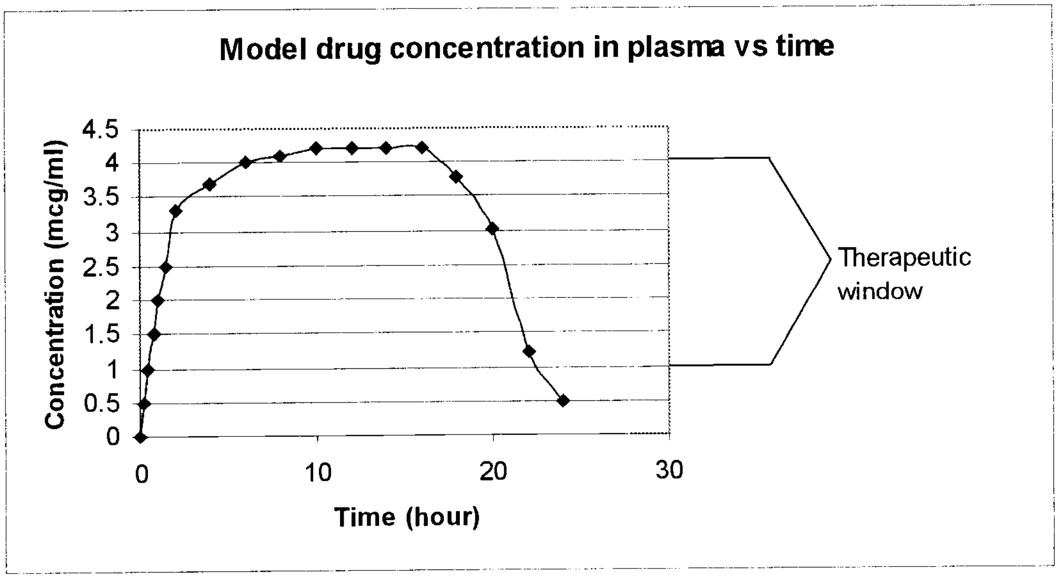


Figure3: Plasma drug concentration of a sustained release dosage form of a model drug

Where

$F = 1$ $V_d = 10$ L

$K_0 = 0.1$ $K_{el} = 0.1$

In Figure 3, the plasma concentration of the drug is within the therapeutic window for a longer time than from the immediate release drug dosage form (Figure 2). Thus, a sustained release dosage form maybe more beneficial in treatment. Second, sustained release dosage forms bring more convenience to patients as the interval of drug dosing can be lengthened, reducing the number of doses to be given. For some drugs, i.e. drugs treating hypertension, it is desirable to produce the maximum plasma concentration of drug in the morning. If a sustained release dosage form with an appropriate lag time before drug absorption begins is taken in the evening, the formulation may allow drug plasma concentrations to reach their maximum value in the morning (1).

Sustained release of nifedipine

Several sustained release dosage forms of nifedipine have been approved in the US and have been proven to be effective in treatting hypertension. This thesis research involves development of a nifedipine sustained release dosage form by hot-melt batch coating technique. An *in vitro* dissolution test was used as a preliminary test to evaluate rate of drug release. Furthermore, mathematical simulation was also used to predict the *in vivo* drug plasma concentrations expected from new and known formulations.

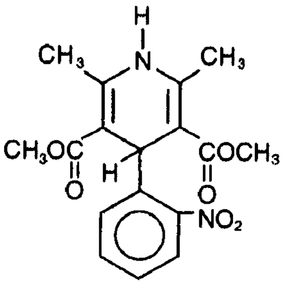
CHAPTER 2: LITERATURE REVIEW

Nifedipine

Chemical structure

Nifedipine is a derivative of dihydropyridine, a yellow crystalline substance. The chemical or scientific name of nifedipine is 3, 5-pyridinedicarboxylic acid, 1,4-dihydro-2,6-dimethyl-4-(2-nitrophenyl)-,dimethyl ester. Molecular formula: $C_{17}H_{18}N_2O_6$. Nifedipine molecular weight is 346.3 (2, 3).

The Chemical structure is as follows.



Chowdhury Hafizul (4) reviewed papers about the relationship between structure of nifedipine and activity. He indicated that the 1, 4 dihydropyridine ring was essential for calcium channel blocking activity. A phenyl group at the 4 position of 1, 4 dihydropyridine frame enhances potency of blocking activity. Ortho- and meta-substitution in the phenyl ring also increases inherent attributes of calcium blocking. Two ester groups in the 3 and 5 positions appeared to be optimal (6).

Nifedipine's chemical structure agrees with its physical characteristics. Nifedipine behaves as a medium-mass, non-polar molecule. Nifedipine is practically insoluble in water (more than 10,000 parts of solvent needed to dissolve one part of solute at 25^0C). In less polar solvents such as ethanol, methanol and other organic solvents, nifedipine is soluble (3). It is very sensitive to both ultraviolet and visual radiation. When it exposed to daylight, the photoproduct is a toxic substance, a nitrosophenylpyridine derivative. Under UV-light nifedipine converts to nitrophynylpyridine (2).

Pharmacology

Nifedipine is a calcium ion influx inhibitor, antianginal agent. Like other calcium blocking agents, nifedipine inhibits transmembrane influx of calcium ions into cardiac muscles and smooth muscles (3).

The contractile processes of cardiac muscle and vascular smooth muscles are dependent upon movement of extra cellular-calcium ions into these cells through specific ion channels. Nifedipine selectively inhibits calcium ion influx across the cell membrane of cardiac muscles and vascular smooth muscles without changing serum calcium concentrations (3, 5).

Mechanism of action

Although nifedipine has been used for a long time, the real mechanism of action has not been determined. It is well known that calcium flux plays an important role in muscle contraction. The hypothetical mechanism of action includes relaxation and prevention of Coronary Artery Spasm, reduction of Oxygen Utilization and hypertension (3, 5, 6).

Among calcium antagonists, the 1, 4 dihydropyridine derivatives are considered slow calcium blockers and are more powerful and more specific blockers of calcium entry than non dihydropyridine blockers (3, 5).

Indication

Nifedipine is indicated for vasospastic angina, chronic stable angina, hypertension, pulmonary hypertension, Raynauld disease and bronchial asthma (3, 5).

Pharmacokinetics

Absorption

The pharmacokinetics of immediate and extended release dosage forms of Nifedipine is well-studied. Nifedipine is absorbed well and complete after oral administration (5, 6, 7, 8, 9, 10). Although absorption is complete, nifedipine is suggested to undergo first-pass metabolism by cytochrome P450 3A4 (CYP 3A4) in the intestine. It was shown in rat's intestine that nifedipine undergoes first past metabolism in the intestine and in the liver by CYP 3A4 (11). Drugs like propanolol may alter hepatic blood flow, and the combination of administration of propanolol with nifedipine makes the bioavailability of nifedipine higher (12). Nifedipine's bioavailability is considered conventionally to be 50-70% because the first pass of metabolism, and the bioavailability does not seem to be affected by food (5, 8, 9). As regards to absorption in the intestine, a group of German scientists (7) found that concomitant food intake with nifedipine, especially high fat breakfasts, delays the drug emptying from the stomach. The enteric coated tablets of nifedipine have a longer lag-time of absorption, whereas the osmotic push pull oral dosage system was not affected by the presence of food in the gastrointestinal tract (8, 14). Within the dose range of 10mg - 30mg, bioavailability is proportional to dose. Two popular nifedipine dosage forms are Adalat® CC and Procardia® XL. Adalat® CC is sustained release matrix coated tablet formulation, while

Procardia® XL is an osmotic pump oral tablet dosage form. It is also observed that some well-known sustained released dosage forms including Adalat® CC and Procardia® XL have lower bioavailability than immediate release dosage forms. The bioavailability of Adalat® CC is 84-89 % of the value compared with immediate release capsules. It is estimated that the maximum plasma concentration of three capsules, 30mg of immediate release dosage form Adalat® Capsule (therefore equal dose) is about 29% higher than Adalat® CC (4). In the osmotic pump system, drug delivery does not change with dosing. Thus, the release profile of 30mg tablets plus 60mg tablets of nifedipine is not significantly different from a 90mg tablet (8). However, the drug in the push-pull osmotic tablet is not released totally. That is why drug loaded inside the osmotic system (Procardia® XL) exceeds the labeled dose by 10% (8, 9). Time before drug is detected in the plasma after oral administration is about 10 minutes for immediate-release dosage forms. The maximum plasma concentration occurs after 30 minutes for immediate release capsules (4) but ranges from 5 hours to 14 hours with sustained release dosage forms (15, 16). Immediate-release dosage forms can be swallowed whole, broken, and swallowed or broken and held sublingually. The nifedipine absorption rate from broken capsules is higher than from intact capsules (17). In some sustained release tablets with zero order release during 24 hours (Procardia® XL), plasma concentrations of drug is maintained at the plateau for a long time that is the absorption process limits elimination. The plasma concentration fluctuations of Gastro-Intestinal Therapeutic System (GITS) of nifedipine are four times less than the fluctuation of peak and trough values seen from immediate-release dosage forms; thereby reducing undesirable drug effects compared to the immediate-release formulation (8).

Nifedipine is absorbed well throughout intestinal mucosa as well as throughout nasal and oral membranes. In a crossover study carried out in healthy volunteers, the absorption rate constant of nifedipine from nasal administration and oral administration was compared. It was shown that if all subjects were administered the contents inside of immediate release nifedipine capsules, the absorption rate constant (k_a) for nasal application was significantly higher than that of oral administration (about 5 times greater). In addition, bioavailability of nifedipine through nasal cavities was quite high (Relative bioavailability was approximately 0.5). It was suggested that nasal administration of nifedipine was feasible for treatment of semi-emergency hypertension episodes (18). Sublingual absorption rate of nifedipine through the oral cavity was also higher than that of the swallowed oral immediate dosage form (17). Kubota et al. (20) applied moment analysis to calculate pharmacokinetics and bioavailability of intravenous, intraduodenal, buccal, rectal, and percutanously administered nifedipine. He discovered the intraduodenal route had bioavailability of only 52-57% due to first passage through the liver and intestine, whereas non-portal routes yielded 10-30% higher bioavailability than intraduodenal delivery. He concluded that percutaneous as well as buccal administration were both promising routes of administration for nifedipine.

Distribution

Nifedipine has a large volume of distribution (V_d) even though its V_d is relatively smaller than the two other main calcium channel blockers, verapamil and diltiazem. In healthy subjects, nifedipine has V_d value of 1.8L/kg (21). From other authors, the volume of distribution of nifedipine ranged in normal subjects from 1.4 liters to 2.2 liters/kg (8).

Percent of protein binding with nifedipine is high in healthy subjects: 92-98% nifedipine binding with plasma proteins (4). This ratio may reduce with impaired renal or hepatic function (4). The high binding ratio of nifedipine to plasma protein may also alter pharmacokinetic parameters of other drugs in plasma. In concomitant treatment of nifedipine, the volume of distribution of theophylline was changed. The theophylline plasma levels were lower than normal values. It is supposed that high affinity of nifedipine toward plasma proteins may cause displacement of theophylline from plasma proteins binding with theophylline (22). In patients who suffer hypertension associated with hyperlipidemia, hyperlipidemia reduces fraction unbound of nifedipine in plasma by approximately 31%. Consequently, total clearance of nifedipine is decreased. AUC, C_{max}, and half-life of nifedipine increase whereas bioavailability and volume of distribution at steady state remain unchanged in hyperlipidemia (23).

For Intravenous administration, nifedipine is best fitted with an open two-compartment model. If nifedipine (immediate dosage form) is given through oral and sublingual routes, the plasma concentration time curve revealed that either one or two-compartment models could be suitable for nifedipine (18). The distribution half-life (alpha phase) is about 0.09-0.17 hours after intravenous administration (8, 9).

In order to evaluate intraindividual variability of nifedipine, a crossover study was carried out in human beings. Nifedipine pharmacokinetic parameters in healthy subjects were estimated in twelve young healthy subjects. The study noted that variability in the AUC of nifedipine was quite small. Coefficient of intraindividual variability was 13% and that of between-subject variability was 54% when one week intervals were applied.

Long-term intraindividual variability (1.5 years) was relatively higher than short-term variability (24).

Metabolism

Elimination half life of nifedipine ranges from two hours to seven hours, and half-life does not change significantly with higher doses. In young healthy volunteers, half-life is observed to be from 1.5 to 3 hours with little variability (13). It is shown that, in impaired liver patients such as liver cirrhosis patients, half-life of nifedipine is prolonged. *In vitro* studies with human livers and experimental animals' livers also showed similar results with clinical observations (13). CYP 3A4 plays an important role in oxidation of nifedipine in the liver. Radio-labeled nifedipine was used to determine major metabolites of nifedipine, and two metabolites were observed, 2,6-dimethyl-4-(2-nitrophenyl)-5-methoxycarbonyl-pyridine-3-carboxylic acid and the corresponding 2-hydroxymethylpyridinecarboxylic acid. Quinidine, macrolide antibiotics, steroids and other dihydropyridine derivatives are substrates of CYP 3A4. It was noted that drugs which are substrates of CYP450 3A4 may undergo competition with nifedipine for metabolism. For example, Tacrolimus's clearance reduced when nifedipine is administered at the same time (30).

Drugs which are inhibitors of CYP 3A4 are likely to change pharmacokinetic parameters of nifedipine. All substances that inhibit CYP 3A4: cimetidine, ranitidine, some antibiotics such as erythromycin, fluocetine, anti-virus drugs such as indinavir, ritonavir, saquinavir, and imidazole derivatives, and the anti-fungal drugs such as ketoconazole, itraconazole, fluconazole, and valproic acid, increase nifedipine plasma concentrations. Consequently, the AUC of nifedipine increases. Thus, whenever drugs that inhibit CYP 3A4 are used at the same time with nifedipine, caution needs to be taken (5, 26). Several compounds in grapefruit juice and citrus juice also have inhibitory potency for human liver microsomes, CYP 3A4 (27, 28).

Also drugs which induce CYP 3A4 will stimulate the metabolism process of nifedipine. After pretreatment of nafcillin (the drug was administered at 500mg four times a day), immediate release nifedipine 10mg was dosed; statistical findings showed that nifedipine's plasma concentrations were reduced by more than 2.5 times following pretreatment with nafcillin. Metabolic induction by nafcillin increased clearance of nifedipine significantly (29). Rifampicin is well known for inducing CYP 3A4. Similar reduction in plasma concentrations were obtained for nifedipine after pretreatment of rifampicin in both human and experimental animals (30). Concomitant administration of nifedipine and carbamazepine, and phenobarbitol were also reported to reduce nifedipine's plasma concentrations due to increased metabolic induction capacity (3).

Genetically, Tateishi et al. evaluated nifedipine's oxydation activity and the CYP 3A4 level in both Caucasian and Japanese subjects through evaluation of microsomal samples. There was no remarkable ethnic difference between the two populations. Another study conducted on American subjects including samples of those with White, Black, Hispanic, Japanese and Chinese ethnic backgrounds also found that the promoter region, or polymorphism, did not appear to play a considerable role in constitution of CYP 3A4 expression. A study on six healthy Mexican volunteers concluded that no significant difference was observed in males and females. This implied that gender was not a factor in pharmacokinetic variability (31, 32).

Although nifedipine is sensitive to light, no changes of pharmacokinetic parameters were observed in human subjects when exposed to UV and normal day light. Nifedipine inside the body does not seem to be degraded by skin exposure to light (33).

Elimination

In normal renal function subjects, the elimination rate constant of nifedipine is about 0.1- 0.3 hour^{-1}. The clearance of nifedipine is about 0.5 L/hour/kg (8). Prolonged clearance of nifedipine in liver cirrhosis patients is confirmed by a clinical study in both healthy subjects and cirrhosis patients. Maximum plasma concentration, AUC, and relative bioavailability were much higher in hepatic failure patients. As a result, cutting down of nifedipine's dose in this type of patient was necessary depending upon the clinical situation (34). Besides liver failure, concomitant ethanol with nifedipine in rats showed that total clearance of nifedipine was reduced by 49% (35). In elderly patients, both clearance of nifedipine and volume of distribution decrease. By way of consequences, the bioavailability increased. The absorption and elimination constants in elderly subjects are similar to in young subjects. However the clearance of nifedipine was decreased by 36% (36) in Elderly subjects. Since nifedipine is beneficial for elderly patients, a reasonable calculation of dosing for these subjects is demanded (37).

Excretion

Metabolites of nifedipine are excreted by the renal route and only traces of unchanged nifedipine are found in the urine (<0.1%). Nifedipine's inactive metabolites are excreted 60-80% in the urine and the remains are excreted in feces through biliary excretion (4). Nifedipine can be used for patients suffering renal failure (38) even though there are not enough studies about the relationship between renal failure and excretion of nifedipine metabolism products. In one recent study (39), investigating the long-term effect of nifedipine on distribution of fluids in body compartments and on cardiac function in hypertensive patients who were maintained on kidney dialysis, two groups of patients were selected. One group was treated with nifedipine for one year with doses of 20-40mg/day. The other group consisted of subjects who had been treated by anti-hypertensive medicines in the past but not at that moment, and all subjects had stable blood pressures without drug for at least three months. The antropometric measurements were recorded. The paired-t test indicated that the group treated with nifedipine had a higher percentage of extra-cellular water at the beginning of dialysis; this difference was not significant after dialysis. No echocardiographic differences in findings were observed between the two groups. The higher extra-vascular water in hemodialysis patients is an unfavorable effect if patients suffer left ventricle heart failure. Therefore, it is suggested that nifedipine should be used with care in hemodialysis patients with heart failure.

Available Dosage forms

Since nifedipine is insoluble in water and sensitive to both UV and daylight, parenteral injection products are not common in the market. As mentioned earlier, nifedipine is absorbed rapidly and completely after oral administration (4, 8). For this reason, nifedipine's oral dosage forms are most popular. Immediate release dosage forms of nifedipine are available in 10mg and 20mg capsules with oral doses administered multiple times per day. Adalat® and Procardia® capsules are the most frequently prescribed inmmediate-release capsules. In these capsules, nifedipine is dispersed in liquid solvents which includes: glycerin, pepper mint oil, polyethylene glycol 400 and water. These ingredients inside the capsule delay the absorption of nifedipine slightly. After nifedipine's administration in capsule form, nifedipine can be detected in plasma in 10 minutes and the plasma concentration reaches its maximum after approximately 30minutes. Another excipient in the capsule is polyethylene glycol 400. Polyethylene glycol 400 at room temperature is a liquid. It is often used to enhance solubility of poorly soluble drugs. It is reported that (5) polyethylene glycol is used to increase bioavailability of nifedipine. The mixture of excipients inside the capsules guarantees that the bioavailability of nifedipine from capsules *in vivo* varies little among dosing units. Depending on clinical situations, immediate release nifedipine capsules can be swallowed whole, broken and swallowed, or broken and held sublingually. The sublingual route of administration is used for both adults and infants in semi-emergency cases. The sublingual dose of nifedipine in infants is 0.25-0.5 mg/kg (17). In adolescents, the commonly prescribed nifedipine dose is 30-90mg/day. Due to nifedipine's rather short elimination half-life (1.5-3 hours) (13), daily multiple dosing is necessary for immediate release dosage forms. This often leads to a preference of sustained release dosage forms of nifedipine.

Sustained release tablets containing 30mg, 60mg, and 90 mg nifedipine are available on the market. The sustained release dosage forms bring many benefits for patients. There are two popular sustained release tablets with brand names Adalat® CC and Procardia® XL. Adalat® CC is the film-coated tablet, so called coat core system. The formulation is composed of an external coat and an internal core (5, 8). The coating layer is constructed with hydroxypropyl methyl cellulose, polyethylene glycol, ferric oxide, and titanium dioxide. Hydroxylpropyl methyl cellulose is a hydrophilic polymer. Higher viscosity grades are used with organic solvents. The film absorbs water and disintegrates slowly in the water due to the high viscosity of the polymer in water. The film acts as a barrier to retard the release rate of drug into solution. Solid grades of polyethylene glycol used in combination with hydroxylpropyl methyl cellulose acts as a hydrophilic polishing material and as a plasticizer of the coating film. Ferric oxide and titanium dioxide are the color agents. The inert ingredients in the matrix are: hydroxylpropyl cellulose, lactose, cornstarch, crospovidone, microcrystalline cellulose, silicone dioxide and magnesium stearate. Hydroxylpropyl cellulose also slows the rate of drug dissolution. The high viscosity grades are often used as a binding substance in the matrix of tablets. Crospovidone and microcrystalline cellulose are disintegrants (40). The maximum drug concentration in plasma occurs at 2.5 – 5 hours after oral administration. Bioavailability of nifedipine from Adalat CC® is about 84-89% of the value seen with immediate release dosage forms (4).

The osmotic pump tablets (e.g. Procardia® XL) which exhibit zero order release kinetics are now favored. The largest advantage of these kinds of tablets is the ability to produce stable plasma concentrations. Inert ingredients in the formulations are cellulose acetate, hydroxylpropyl cellulose, hydroxypropyl methylcellulose, magnesium stearate, polyethylene glycol, polyethylene oxide, sodium chloride, and titanium dioxide. The tablet is called gastrointestinal therapeutic system (GITS). For drugs which are soluble in water, an elementary osmotic pump (EOP) is good enough. The delivery system, EOP, consists of only an osmosis core incorporating the drug and surrounded by a semi-permeable membrane. For water-insoluble drugs such as nifedipine, a more complicated system, GITS must be employed. EOP is replaced by a push-pull osmotic pump that allows a continuous delivery of nifedipine over a 24-hour period. Like Adalat® CC, Procardia® XL is dosed once-daily. The system consists of two separate layers: an underneath osmotic push pull layer and a drug layer. The drug layer contains nifedipine, high molecular weight polymers, polyethylene glycol, and standard tableting excipients. The osmotic layer is composed of a similar polymeric agent along with tableting excipients and the color agent ferric oxide. The entire system is surrounded by a semi-permeable membrane which is made of cellulosic material and serves as a rate-controlling barrier to osmosis. A small hole with a determined diameter is bored through the outer rate controlling barrier coat by a laser-driller. The drug is then released via this pore. The rate of drug release depends on the size of the orifice (8, 41, 42).

When the GITS tablet arrives in the gastrointestinal tract, the osmotic core imbibes water from surrounding medium and creates a flux of water into the tablet. Absorbed water plays two functions within the delivery system. First, water dilutes the drug and forms a suspension within the tablet. At the same time the fluid combines with the osmotic push layer, forming another suspension, too. The push pull layer expands-“pushing” the nifedipine suspension in the drug layer out the hole in the tablet. The release profile of drug is characterized as a zero-order kinetic process. More specifically, the maximum plasma concentration reaches a stable value in about 6 hours after oral administration of Procardia® XL. The GITS system has a special property. The rate of drug release is constant in spite of the fluctuation in environmental conditions. However, only approximately 90% of nifedipine is released from the system. Therefore, an excessive quantity of drug is necessary to be loaded in the GITS system (8). Although GITS has many advantages, the cost of manufacturing GITS is still rather high and manufacturing technology requires special advanced techniques.

Others researchers have also developed sustained release dosage forms of nifedipine. Zheng Wang et al. developed a double-layer nifedipine tablet containing 2-hydroxypropyl-β-cyclodextrin (HP-β-CyD) and 3% noninonic surfactant (HCO-60) as a fast release portion and hydroxylpropyl celluloses (HPCs) with different viscosity grades as a slow-release proportion. The spray dried product of nifedipine/HP-β-CyD (1:4 weight ratio) containing 3% HCO-60 as the fast-release portion was lightly compressed in the die. Nifedipine and HPCs (1:4 weight ratio) as the slow release portion was added onto the tablet. The two combined portions had a 1:3 weight ratio. After *in vitro* and *in vivo* tests the author found that the optimal formulation for 20mg nifedipine sustained release double-layer tablet is HP-β-CyD with 3% (HCO-60/HPC-low:HPC-medium) in a weight ratio 1/(1.5:1.5). This formulation exhibits a relatively good retarding effect on drug release and comparable bioavailability with commercially available slow-release nifedipine products (43).

In another study, Guang Yan et al. created a sustained release tablet by solid dispersion technique (44). The combination of hydroxypropylmethyl cellulose (HPMC) and PEG 6000 seems to be appropriate for insoluble drugs like nifedipine since PEG 6000 enhances the rate of nifedipine's dissolution from the HPMC matrix. First, a solid dispersion of nifedipine in PEG 6000 was prepared with the ratio 1:6. Then a three-layer tablet was made. The external layer consisted of high-viscosity grade HPMC (Methocel K15M), and nifedipine solid was dispersed into HPMC with the ratio (w/w) 1:2. The inner layer consisted of the low-viscosity grade HPMC (Methocel K100) and nifedipine was dispersed into this mixture with the ratio 1:2. The weight ratio of the outer layer to inner layers is 7:3. The mixture was compressed into flat-faced tablets. The obtained tablets were then compared with Adalat GITS 30 both *in vitro* and *in vivo*. The formulation had a similar *in vitro* dissolution profile as to the reference Adalat GITS 30 although the nifedipine profile release deviated slightly from the zero-order kinetics and the total drug released was a little less than the reference. Nevertheless, the *in vivo* results between the test formulation and Adalat GITS 30 were considerably different as compared with *in vitro* results. In fact, the plasma concentration of nifedipine of the test formulation was remarkably higher than that of the commercial product, Adalat GITS 30. No explanations were given for this discrepancy. However the plasma nifedipine concentrations of the test tablets were still lower than minimum toxic boundary.

The bioavailability of nifedipine using solid excipients and solubilizers also was measured in rabbits. Solid dispersions using PEG 6000, PEG monomethylether, β cyclodextrin, and sodium lauryl sulfate, showed *in vivo* results that the nifedipine in 95% PEG 6000 prepared by fusion method will give the highest bioavailability compared with nifedipine powder using other polymers or using PEG 6000 by solvent method. If β cyclodextrin is combined with nifedipine using a complexation process, an increase in both the solubility of nifedipine *in vitro* and bioavailability *in vivo* were observed.

There have not been any studies using hot-melt coating techniques to develop nifedipine formulations.

Development a nifedipine sustained release dosage form by direct blending hot-melt coating method

Introduction of hot-melt coating method

Coating techniques were invented hundreds of years ago. Then, it was applied in Pharmaceutical industry in the early part of the twenty century. The introduction of film coating in 1953 marked a new era of pharmaceutical coating. From that year to now, many different polymers have been invented and evaluated. Improved perforated pans and fluid bed coaters allow producers to produce precise, efficient, and elegant coated dosage forms. Although Hot-melt coating methods by spraying have been well known for a long time, this method has some major shortcomings compared to the organic solvent spray coating method. However, compliance with requirements of regulatory agencies such as the U.S. Food and Drug Administration (FDA), Environmental Protection Agency (EPA) and Occupational Safety and Health Administration (OSHA) make the hot-melt coating method now more attractive since this method does not harm the environment like spray coated organic solvent technique. In comparison with aqueous-based coating methods, hot-melt coating does not require control of microbial growth. In addition, the special equipment and coating agents for hot-melt coating process is commercially available. The hot-melt coating materials offered now for pharmaceutical formulations are not limited to only natural substances. Modern technology today allows pharmaceutical companies to produce various types of fatty substances with a number of aliphatic carbon molecules bonded to the main chain as well as in the branches. Various types of substitution groups can be also added into the molecular structure. In other words, the modifications in the molecular structures can create desirable physio-chemical properties of coating agents.

According to A.S Achata et al., Hot-melt coating by spraying is recommended for potential applications in pharmaceutical formulation mainly in three categories (45):

- Improvement of palatability of the dosage form.
- Prevention of environmental degradation of dosage forms such as physical degradation (light, humidity…), protection from drug incompatibilities, and protection from physiological degradation (enzymatic metabolization in GI tract, pH sensitivity).
- Slowing the rate of drug release from dosage forms. This application has drawn the most attention recently from formulation developers.

However, besides the advantages mentioned above, this method also has some disadvantages. First, coating agents normally used in hot- melt coating methods are lipids, waxes (fatty acids or their esters) that have melting points ranging usually from 40 - 200^0C (4). Temperature sensitive drugs may be degraded at a high temperature. Moreover, a limited amount of coating agent can be deposited on the surface of the coating cores. For high drug dosing, it is preferable to have the total or partial amount of drug dispersed into the substrate by other methods before coating. For example, in an article covered in Pharmazie 2000 June (45), a group of researchers formulated a sustained release dosage form of chloroquine. The tablets were compressed from cloroquine granules and coated by Compriltol 888 (Glyceryl behenat NF). Dissolution results indicated that the granules stayed intact in the tablets during the compression process (45).

Multiple-layer coating can increase coating weight gain. However, the multiple-layer coating process itself is limited. Coating agents in the outer coating layer must have a significantly lower melting point than inner coating agents. In terms of coating agents, there are also some considerations. The molecular weight, hydrophobicity, melting point, rigidity, flexibility and rheological behavior are physicochemical parameters that can provide helpful information to correlate the ability of excipients to prolong drug release. The thermal behavior such as for individual drug and excipients'stability, drug-excipient interaction at high temperature, and stability of the dosage form need to be investigated carefully. Another parameter of concern in addition to thermal behavior is the polymorphic characteristic of the coating material. The polymorphic behavior of the coating material may cause variability within batches of the dosage forms. Polymorphic forms have different physical properties such as transition temperature or melting point. It is reported that polymorphology can change intrinsic dissolution rates and other solid-state properties of materials. Therefore, a full investigation of polymorphology of excipients is necessary. Moisture absorbed within the coating layer is one factor that may affect the drug stability. In one study Achata et al. (46) investigated the interaction of coating excipients (Polyvinyl alcohol and glyceryl behenate) with moisture absorbed inside the coating layers and the influence of temperature and film thickness on the percent of water.

As to safety conditions during the hot-melt coating process, it should not be neglected. Safety for employees who are involved with the manufacturing must be considered seriously because working with a coating pan at high temperature is risky. Toxicology studies of commercial coating excipients are required with complete information of *in vivo* results since the coating agents are consumed along with the dosage forms (46).

Hot-melt coating in fluidized bed

Generally, any fluidized-bed coating equipment can be modified for hot-melt coating process (46). There are four well known hot-melt coating techniques which have been applied in the pharmaceutical industry: top spray, bottom spray, tangential spray and solid dispersion (46). Top spray coating or standardized fluidized bed is the most common for hot-melt coating and was an early application method for melt coating methods. Molten wax was sprayed from the top of the chamber. The core can be small pellets, granules, or particles. The top spray is restricted in application due to product fluidization and flow. Some authors reported that top spray fluidized-bed coater was the most suitable for hot-melt coating due to its ability to operate with product temperatures closest to the congealing temperatures of the molten excipient. In other words, top spray coating can reach minimum product temperature over the melting point (PT/MP ratio) (46, 47). The process is composed of three sequential steps. First, the molten material is sprayed onto substrate surface then it spreads around the substrates. Second, coated particles are congealed. In top spray coating, molten coating agent is maintained at 10-

60°C above melting point. Typically it is 40-60°C. The substrates are kept at 10-20°C below the melting point of the coating agents. An improvement over conventional top spray fluid bed is bottom spray (48). Bottom spray is useful with small substrates. It creates more ordered flow and it is appropriate for small coating levels. Large coating levels are also possible at the expense of PT/MP ratio (46).

Tangential spray in which the energy from disk aids spreading and smoothing of the coat is a modification of the equipment used for fluid bed method. Large coating levels are possible at expense of PT/MP ratio. One shortcoming of this device is its limited capacity (46, 49, 50).

An important part of spray coating is the spray delivery system. To deliver the coating agent onto the substrate without discontinuity, Achata et al. (46) emphasized the need for a hot air flow that leads directly into the nozzle. The spray gun is kept at a high temperature to prevent condensing of molten waxes in the gun, so that a heating device is necessary and the spray gun inside the expansion chamber needs to be well-insulated since the temperature in the chamber was much lower than the molten fluid material. "This is required to prevent the re-melting of coating material on the substrate when they come in contact with spray gun while falling back down into the bed" (46).

Kennedy et al. introduced a solid dispersion coating technique, which does not require the spraying process. This technique used a Wurster column in the fluid-bed coater. The substrate is combined with a coating agent in the fluid bed chamber due to the temperature inside the chamber by following four simple steps: Chamber warming up, preheating, melting, spreading, and cooling-congealing. The device for this technique is not as complicated as spray coating since the nozzle spray system is omitted. Nevertheless, compared with conventional spray methods, a series of weak-points can be found. The cores and the coating excipients are put into a chamber. Keeping the whole system at a high temperature is not very feasible. It was discovered that the porosity and density values of substrates affected the method's reproducibility. The nonpareil-sugar beads tend to agglomerate if the particle size is smaller than 40 mesh for coating agents PEG 1450-8000 and MPEG 20000 and 5000 (46). In an effort to make the spreading homogeneous, the optimal viscosity of the coating agent is less than 300 centipoises. This method also has a problem in that it allows only a low percentage of hot-melt coating 2.5-5%. In fact, in real cases a higher percentage of coating is required to be deposited. Kennedy et al. applied this improved technique for preparing chlopheniramine maleate coated beads and its dissolution profiles were also tested (50). A series of waxes were used in their study. The waxes were Atmul 84K (Mono-and diglycerides), Vybar 253 (hydrocarbone), Beewax 442, White (hydrocarbone), Synthetic spermaceti 4013 (hydrocarbone), Parafin 173 (hydrocarbone). Among all the prepared beads, beeswax coated beads of 2% met all specifications of Drug Release Test 1 for an "Extended-release Capsule of Chlorpheniramine maleate" according to USP25-NF20. Differences in melting points between Vybar (67^{0}C) and Synthetic Spermaceti 4013 (47.5) allowed them to consider multiple coatings. The dual coated beads (2% Vybar and 1.0% Synthetic Sparmaceti 4013) yielded a lower amount of drug release after 10 hours than the single coated beads of 2% Vybar. The author reported that the gap needed between melting points for two coating agents to be applied using the hot-melt fluid bed method for dual coating was at least 15^{0}C.

Hot-melt coating by direct blending

In fact, hot-melt coating by direct blending is the simplest way to make coated particles by the hot-melt coating method. Although the technique does not require complicated equipment, the obtained results are quite surprising and it can be applied for a wide range of different size substrates as well as multiple coated layers. The method consists of the following steps: Melting coating agent, drug dissolution or dispersion in the molten coating material, good mixing of the substrate and molten coating agent, cooling with continued stirring of the mixture, and congealing the coated particles. Like other coating techniques, the active ingredient can be deposited in the core by a granulating method, and then coated outside by a coating layer. The drug also can be dispersed into the coating agent and then the mixture is coated outside the coating core. Ready-made sugar beads of various sizes are commercially available. These sugar beads are homogenous in size and shape and easily adhere to waxes. Customers can select an appropriate size of sugar beads for a reasonable price.

The smaller the size the substrate is, the larger surface there is for coating agent to deposit onto. Weight gain during coating can reach a high value. However, extremely tiny particles are likely to agglomerate which increases the variability of the coated beads mixing and coating must be appropriately controlled to avoid variability. To obtain high weight gains with ready made substrates, the process is most simple if the core has a large enough surface area but is not too small in size (so as to avoid agglomeration). In other words, it is desirable that the coated beads contain a large amount of drug but the variability is reduced to a minimum value. For a laboratory scale research project it has been found that size range of sugar beads is 30-60 mesh work nicely as demonstrated herein. The coated beads then are loaded into hard gelatine capsules which are the final and complete dosage form. Coated beads may be used to compress into tablets, too. There are no documents that list waxes that should be applied in the coating process to obtain slow drug release. It is reasonable that waxes with high molecular weight and hydrophobicity are likely to reduce the drug dissolution rate in water. Conversely, substances which are hydrophilic or increase the wetting characteristics of the drug are likely to increase the rate of drug dissolution (e.g. PEG). Besides that, to create multiple coated layers, the melting point of coating agent of the inner layer is suggested to be higher than the melting point of the agent used in the outer layer by 10 to 20^{o}C. All the waxes need to be hard enough to congeal at room temperature. It is well-known that nifedipine is sensitive to light, yet, there are no reports on the behavior of nifedipine at high temperature. Thus, it is obligatory to investigate carefully the stability of the active substance to heat. Moreover, sugar beads are made of sucrose which is easily burned at high temperature. So the limiting temperature is 100^{0}C.

Fitting concentration-time curve

Zero-order kinetics absorption and zero order kinetic release

An absorption process is defined to be zero-order kinetics if the rate of absorption does not depend on the drug concentration in solution. The absorption rate is constant.

$$\frac{dA}{dt} = k_0$$

k_0 is absorption rate constant.
A: Amount of drug absorbed

The fitted equation of the drug concentration $C = a + k_0 t$

for the typical plot for zero-order kinetics, accumulative drug or drug concentration versus time, is a straight line. Indeed, the absorption process *in vivo* through the gastro-intestinal walls has been studied thoroughly. Only dissolved drug in the GI tract is absorbed into systemic circulation. Reasonably, *in vitro* studies are based on an assumption that amount of dissolved drug is equal to or reflects the absorbed amount of drug. This is true for well- absorbed drugs like nifedipine. So a nifedipine dosage form having a zero-order release kinetic dissolution profile *in vitro* tends to have zero-order absorption kinetics *in vivo.*

In an effort to make a zero-order absorption dosage form, the initial assessment is making a zero-order release dosage form for the drug. That means, the drug dissolution profile needs to follow zero-order kinetics compared with the usual first-order dissolution kinetics of pure drug. An ideal zero-order dosage form has a release concentration time curve that is a straight line with the linear coefficient (R^2) equal to one. Moreover, for oral products, drug should be totally dissolved within 24 hours.

Other types of drug release fitting

Besides zero-order kinetic release of drug, some other fitting equations are used to describe the drug release from a matrix tablet formuation. If the drug releases very fast it is likely to obey first-order kinetics. However, for many drugs that are not soluble or are captured in the matrix, the dissolution profile does not follow first-order. Several types fitting equations lying between first-order and zero-order kinetics are used to describe these drug release profiles.

E. Nicholas Griffin and Paul J. Niebergall (48) used three equations to fit one coated layer beads with coating agents: Beewax 422 P (Ester of various C15-C34 compounds), Compritol 888 ATO (C22 triglyceride), Syncrowax HGL-C (C16-C36 triglyceride), Syncrowax HR-C (C22 triglyceride), Lubritab (C16-C18 triglycerides). A first-order equation, a square root of time equation, and a dual equation that combines first-order and square root of time kinetic equation were used to describe the drug release profile.

First-order model: $C = C_n(1 - \exp(k_1 t))$

Square root of time model: $C = k_2 t^{0.5}$

Dual model: $C = C_n(1 - \exp(k_1 t)) + k_2 t^{0.5}$

Coated beads used in the study were prepared by "solid dispersion hot-melt coating method" (47). Substrates were sugar spheres coated with 3.5% chlorpheniramine maleate. Akaike criteria (AIC) was used to select the most appropriate model and R^2, r^2 values were to evaluate the goodness of fitness. The Nonlinear Regression Program Scientist software was used to fit the experimental dissolution data. It was found that the dual equation best described the drug dissolution concentration-time curve. Initially, release is governed by both processes: first-order and square root. Later on, release is governed by square root of time process. Tested beads were divided into two groups: Low-k_1 group and high-k_1 group. The authors defined the low-k_1 group as beads which have a first-order portion that lasts 8 hours and the high-k_1 group's first-order release portions lasted only approximately 1 hour. Because the coating quantity is was less than is applied by the new batch hot-melt coating process used in this current research (reference to U.S. patent application again), the "best" fitting model equations may or may not be the combination equation that worked best for the solid dispersion hot-melt coating method.

In a similar way - in another article - other different types of equations could be used to fit the dissolution profiles of sustained release dosage forms (44).

The Hixson-Crowell equation: $\frac{A_{release}}{A_{total}} = 1-(1-kt)^3$

Higuchi equation: $\frac{A_{release}}{A_{total}} = b + kt^{1/2}$

In fact, the Higuchi equation is the square root of time model. Suitable curve fitting can be evaluated by AIC, and R^2 values to see which model (equation) fits the dissolution data.

From reviewing coatings materials available and nifedipine, this study reposrted herein was performed to create a sustained release dosage form of nifedipine by direct blending batch hot-melt coating. Formulations prepared in this study have relatively more than 80% of drug released within 24 hours and the drug release was fitted using linear regression to a linear equation, an R^2 from fitting linear equation to the drug dissolution profile of more than 0.95 was considered acceptable.

CHAPTER 3: MATERIALS AND METHODS

Materials

Different types of waxes were used in the study.

Gelucire® 53/13:

Gelucire® 53/13 is a synthetic wax offered by Gattefosse'. Gelucire® 53/13 is an inert semi-solid waxy material which is amphiphilic in character. It is derived from the reaction of hydrogenated palm oil with PEG 1500 and the composition is approximately 20% mono-, di- and triglycerides. 72% mono- and di- fatty acid ester of PEG 1500 and 8% free PEG. Gelucire® 53/13 has a nominal melting point of 50^0C and a hydrophile-lipophile balance (HLB) value of 13. It has been tested for safety for pharmaceutical usage (48).

A series of synthetic waxes from Croda Inc. were evaluated including:

Emulsifying wax NF or Syncrowax AW1-C: This is an unusually long chain (C18-36) fatty acid. It can be used as a primary emulsifier. It is neutralized, and constitutes a long chain alternative to stearic acid. Melting point is 60-64°C (49).

C18-C36 Acide Triglyceride or Syncrowax HGL-C: This is the triglyceride ester of syncrowax AW1. The Acid Triglyceride or syncrowax HGL-C is the hardest of the synthetic syncrowax type. It has high degree of rigidity. Its properties are similar to carnauba wax. The melting point is 70-75^0C (49).

Synthetic beeswax, syncrowax BB4: This is made from a blend of other syncrowaxes and is similar in composition to Beeswax. The melting point is 60-65^0C (49).

Stearic acid: Stearic acid was bought from J.Y. Baker Chemical Co., Phillipsburg, N.J. 08865. The BP 1993 and the USPNF XVII describe stearic acid as a mixture of stearic acid ($C_{18}H_{36}O_2$) and palmitic acid ($C_{16}H_{32}O_2$). The content of stearic acid is not less than 40.0% and the sum of the two acids is not less than 90%. The melting point is equal to or more than 54°C (40).

Carnauba wax: Carnauba wax consists primarily of a complex mixture of esters of acids and hydroacids. Also present are acids, oxypolyhydric alcohols, hydrocarbons, resinous matter and water. Carnauba is the hardest wax and has the highest melting of the waxes. The melting point is 81-86°C (40).

The range of coating agents used corresponding to the increase of rigidity and the increase of melting point is presented in Table 1.

Table 1: Waxes used in hot-melt batch coating process according to increase in melting point

Coating agent	Formula	Melting point	Rigidity
Gelucire 53/13	20% mono-, di- and triglycerides. 72% mono- and di- fatty acid ester of PEG 1500 and 8% free PEG	50°C	Lowest rigidity ↓ Highest rigidity
Stearic acid	Mixture in which Stearic acid ≥ 40% Stearic acid + Palmitic acid ≥ 90%	54°C	
Emulsifying wax	Unusually long chain (C18-36) fatty acid	60-64°C	
Synthetic beewax or syncrowax BB4	Blend of other syncrowaxes and it is similar in composition to Beeswax.	60-65°C	
Acide Triglyceride or Syncrowax HGL-C	The acide triglyceride, syncrowax HGL-C is the hardest of the synthetized syncrowax range. It has high degree of rigidity. It has properties similar to carnauba wax.	70-75°C	
Carnauba wax	A complex mixture of esters of acids and hydroacids	81-86°C	

Coating cores: Sugar sphaeres NF, 30-35 mash size were from the Paular Corporation. The sugar spheres met all the norms in USP25- NF20 (3) such as percent of sucrose, identification, specific rotation, microbial limits, loss of drying, particle size, and heavy mentals...

The active drug: Nifedipine, the active agent, was obtained from Sigma Chemicals. The product is a powder with minimum purity of nifedipine of 98%. Nifedipine also meets requirements of the nifedipine monograph in USP 25 NF (3). Beads are loaded in capsules size 0CS, natural transparent from Capsulgel, division of Warner-Lambert.

Methods

Description of hot-melt coating by direct blending batch method

Preparation of single layer coated beads

Stage 1: Coating material was melted and the temperature was kept 10°C – 20°C higher than its melting point but the temperature must be under 100^0 C when sugar beads present to avoid degradation of the sugar beads. The temperature is maintained until all waxes are melted and becomes a homogenous liquid. It is ideal if the coating process is carried out in a chamber or a container that has a consistent temperature throughout the whole container. A cylindrical mixing container fixed in a rotating axel with a temperature bath is recommended for blending and coating. Present facilities allowed hot-melt coating only with simple stirring.

Stage 2: Nifedipine is then dispersed in the molten coating material while stirring until the mixture was homogeneous. The temperature is still maintained 10°C – 20°C higher than the coating material agent's melting point.

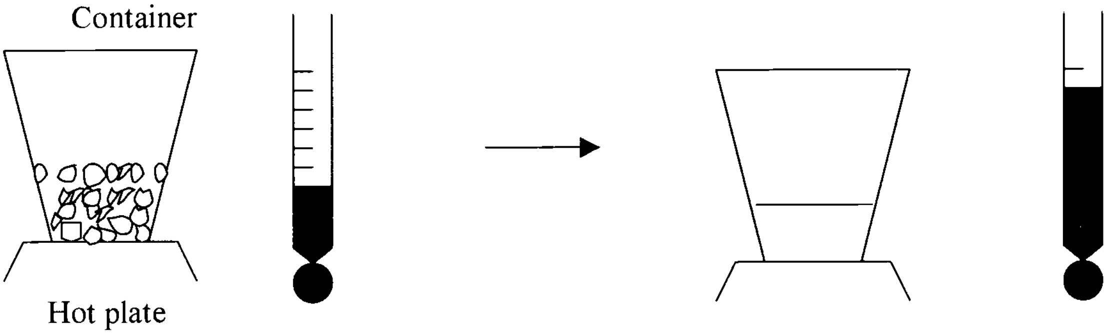
Stage 3: Sugar bead is maintained at the temperature equal to the mixture of the coating material and the drug. This avoids re-congealation of drug-wax mixture. Then the warmed beads are poured into this wax-drug mixture while blending continuously. The preheating process can be performed separately in another container as long as the same temperature is maintained.

Stage 4: This is the dispersion stage. By stirring for 5-10 minutes at a constant temperature inside the coating chamber (higher than melting point 10°C-20°C), the coating material then covers all the surface of the sugar beads. If the temperature is too high the viscosity of the molten mixture is too low, so the fluid can not stick on the surface of the sugar core. If the temperature is too close too the melting point, the mixture is likely to be heterogeneous.

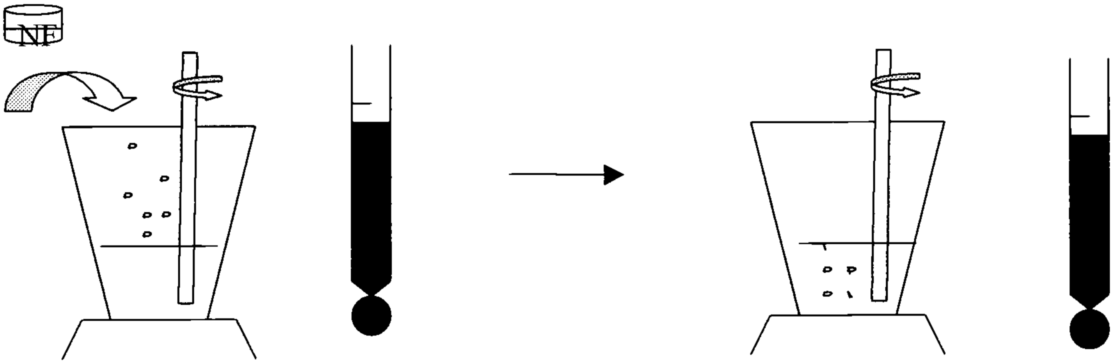
Stage 5: The container is removed from heating source and cooled at room temperature. To prevent agglomeration of beads, light stirring is needed. Finally, coated beads are sieved through mesh size 25 to eliminate any agglomerated beads and to assure uniformity of coated beads.

All the sequential steps are performed under a weak light or in the dark to avoid degradation of nifedipine.

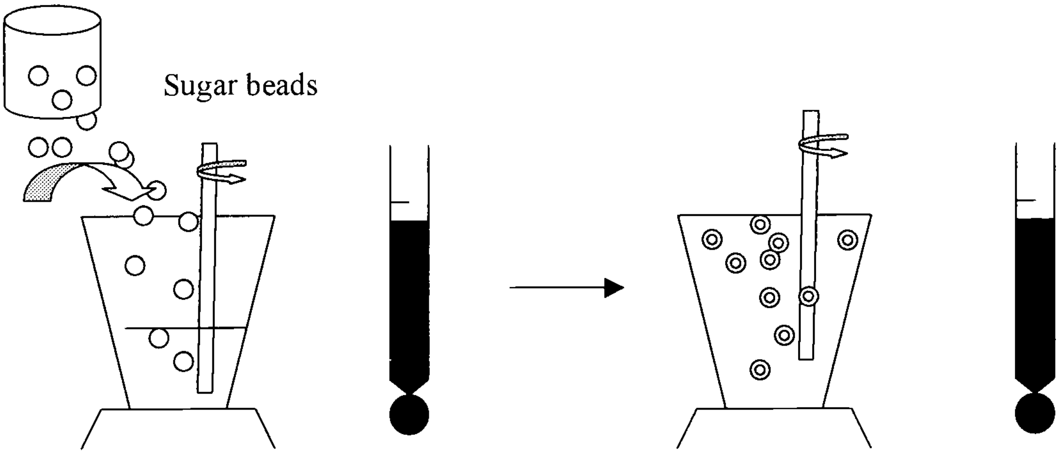
Figure 4: Stages of one layer coating process by hot-melt batch coating direct blending.



Stage 1: wax is melted 10-20°C over the melting point

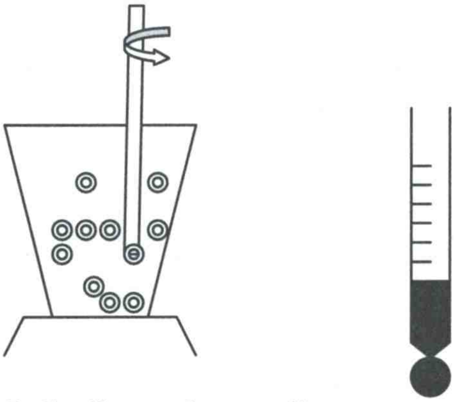


Stage 2: Nifedipine (NF) is dispersed into molten coating agent and then stirring homogenously



Stage 3: Pre-heated sugar beads are mixed with coating material

Stage 4: Coated beads are kept stirring until homogenous



Stage 5: Cooling and congealing at room temperature

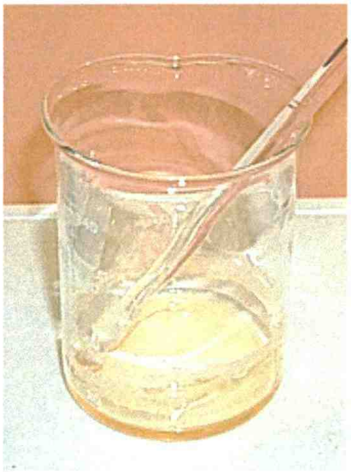
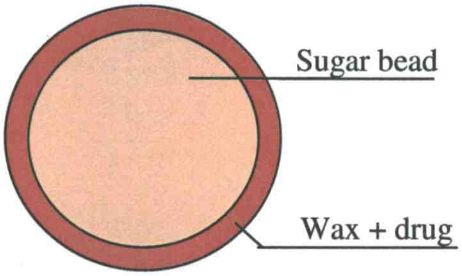


Figure 5: molten wax was being mixed with nifedipine

Figure 6: Sugar beads being coated by the mixture of wax and nifedipine

Drug and wax deposited as a single layer on sugar beads is illustrated in Figure 7.

Figure 7: Single layer beads



Preparation of multiple layer coated beads

The coated beads obtained from one layer coating process then can be used for a second layer coating process. The dual coating procedures are basically performed as two fully sequential single coating procedures. All the stages of the first coating process are repeated for the second coating layer with another wax. To prevent mingling of the new coating layer with the former one, the melting point of the second coating agent used for the second layer is preferably lower than for the first coating agent. Empirically, the gap of the melting point of two waxes is over $10^{o}C$ to insure that the inner layer is intact while undergoing the second hot-melt coating process. If their melting point is too close to each other, the problem is not only the difficulty in controlling the temperature, it is also the stability of the inner layer. If the melting point of the external coating agents approaches the melting point of the inner coating agent, the inner coat is likely to become soft and vulnerable. The inner coat becomes easy to be wiped off due to blending friction and it will be mixed with the wax of the out layer. With appropriate heating and cooling equipment it is anticipated that these problems can be minimized or avoided even when using waxes with very similar or identical melting points.

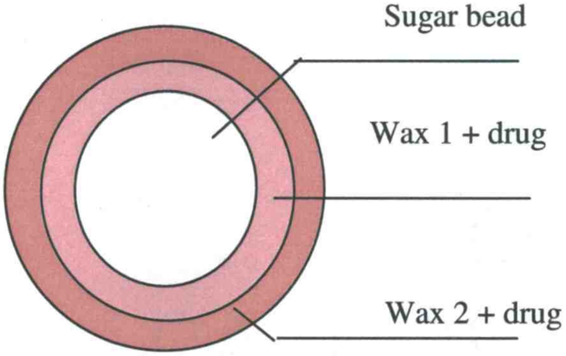
Multiple coatings can increase the weight gain. As mentioned above, the higher the wax melting point is, the more rigid the coating materials tend to be. The inner wax is often less hydrophilic and more rigid than the outer wax. Moreover, the outside layer is eroded first, then after time when water gets inside the beads both two layers are eroded and disintegrate. There are two mechanisms by which water penetrates into the matrix and causes drug release into the water medium. Water can diffuse through the coating layer into the matrix and dissolve the drug inside the beads. Another way water gets inside the beads is by penetrating through channels on the surface of the coating layer. The hydrophobic coating agent is less likely to swell and does not absorb large quantities of water. It is suggested that, with some formulations the coating layer is not totally integrated and its thickness is not the same over the whole surface of the beads. Thus channels distribute sporadically in the coating layer allowing water to penetrate through these channels due to the osmotic difference inside and outside of the beads. As drug and sugar dissolve, the pressure inside the beads pushes the drug out through these channels and parts of the layer are torn and finally the layer is broken. The whole process is slow with hydrophobic coating agents. This will result in a remarkable and desirable delay of drug release from the matrix or in the inner-coating layer.

The retardation of drug release appears to be promoted with multiple-layer coated beads. Theoretically, a third layer can also be put on the coated beads as long as the melting point of the coating agent is lower than the melting point of the second layer, or the heating/cooling/mixing process are properly controlled such as, for example, "chilled" beads being added to a thin stream of molten wax. However, the actual necessity of having more than two coating layers is not required for a dosage form. For the dosage form administered by oral route, drug should release totally in 24 hours. Dual-coating beads developed herein release drug over 24 hours.

Further, a combining of the hot-melt coating technique with other film coating techniques, for example organic spray coating, or aqueous spray coating is feasible depending on the specific purpose.

Dual coated beads obtained by hot-melt coating method are shown in Figure 8

Figure 8: Dual layer coated beads



Dissolution study

Dissolution tests were employed as indicated in USP25-NF20 (3). Vankel 7000 dissolution machine was used with apparatus two, the paddle. Since nifedipine is insoluble in water, the sustained release dosage form containing 30 mg is not totally soluble in 1000ml water. In USP25-NF20, a solution of sodium lauryl sulfate 1% in water is stipulated as the medium for dissolution testing of extended- release tablets because the organic surfactant will increase nifedipine's solubility.

The tolerance allowed for drug release in the USP25-NF20 is regulated and stipulates the cumulative percentage of nifedipine dissolved must conform to the "Acceptance Table":

Time (hours)	Amount dissolved
4	Between 5 and 17%
12	Between 43 and 80%
24	Not less than 80%

It is noted that sodium lauryl sulfate is a surfactant. Surfactants can increase the solubility for insoluable drugs like nifedipine. Polysorbate 80 (Tween 80) is a common surfactant also widely used in the pharmaceutical industry. A solution of 1% (w/w) polysorbate 80 (Tween 80) in water (1000ml) can also be used for the medium for the dissolution test (16), and was used in the current study. The dissolution medium was kept at a temperature of 37.0^0C ± 0.5 ^{0}C, and the paddle rotation rate set at 75 or 100 rpm. The amount of beads loaded with drug used in the dissolution study was equal to having 30mg nifedipine per capsule. The entire dissolution process was protected from light using an amber dark bowl to avoid nifedipine's degradation. The dissolution medium used (with polysorbate 80) seemed to be equivalent to the standard medium in USP25-NF20 since the commercial product Procardia® XL yielded dissolution results which satisfied the dissolution values after 24 hours. It was shown that the bioavailability of Procardia® XL is about 90%, and 94% of nifedipine was released in the dissolution test with 1% Tween 80 in water (1000ml). In addition, pure nifedipine powder yielded a high dissolution rate in this medium.

An additional test to show difference of using this dissolution medium is to place the formulated drug products in with simulated gastric fluid (0.1N HCl) and simulated intestinal fluid (USP phosphate buffer) which was also conducted in order to investigate the effect pH has on the dissolution profile.

Samples were taken at the following times: 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 7, 8, 10, 12, 14, 16, 18, 20, 22, and 24 hours. Four milliliters of dissolution media was withdrawn, filtered, and nifedipine concentrations were measured using a spectrophotometer at 334nm. The medium used in dissolution test was used as the blank sample and standard curves were used to determine the concentration of nifedipine dissolved from the beads. The standard curve was determined from solutions made up of dissolution media with nifedipine concentrations ranging from 10mcg to 45mcg/ml.

Analyzing the actual content of nifedipine in prepared beads was determined as following: An exact amount of coated beads was weighted, equal to approximately 3 mg nifedipine. The weighed beads were then put into a 100 ml amber volumetric flask, and dissolution medium was added to fill ¾ of the flask. Next, the flask was sonicated for 12 hours under protection from light in order to completely dissolve the nifedipine. Finally, the volume was increased up to the line on the volumetric flask and the fluid was filtered through the 0.45 μm membrane.

The solution was measured as soon as possible in the UV spectrometer at 334 nm with the blank sample being the dissolution medium. The actual measured content of nifedipine is used to calculate the percent of drug deposited on the beads and the percent of drug release.

The concentration of nifedipine is interpolated according to the standard curve

$$C_{nif} = \frac{Abs - \alpha}{k_1}$$

Abs: Absorbance of solution
α: Intercept of the linear equation of the standard curve
k_1: slope of the standard curve

The Actual content of nifedipine is calculated from the formula:

$$C_{actual} = \frac{C_{nif} * V}{m_s}$$

C_{inf}: Concentration of drug in medium
V: Volume of medium
M_s: weight of coated beads

Percent of drug deposited (D%) is calculated according to the formula:

$$D\% = \frac{C_{actual}}{C_{theoreticcal}} 100$$

The percent released is

$$R\% = \frac{C_t * V}{mC_{actual}} 100$$

C_t: the concentration of drug in the dissolution bowl after t hours
V: the volume dissolved medium: 1000ml
m: weight of beads in each bowl

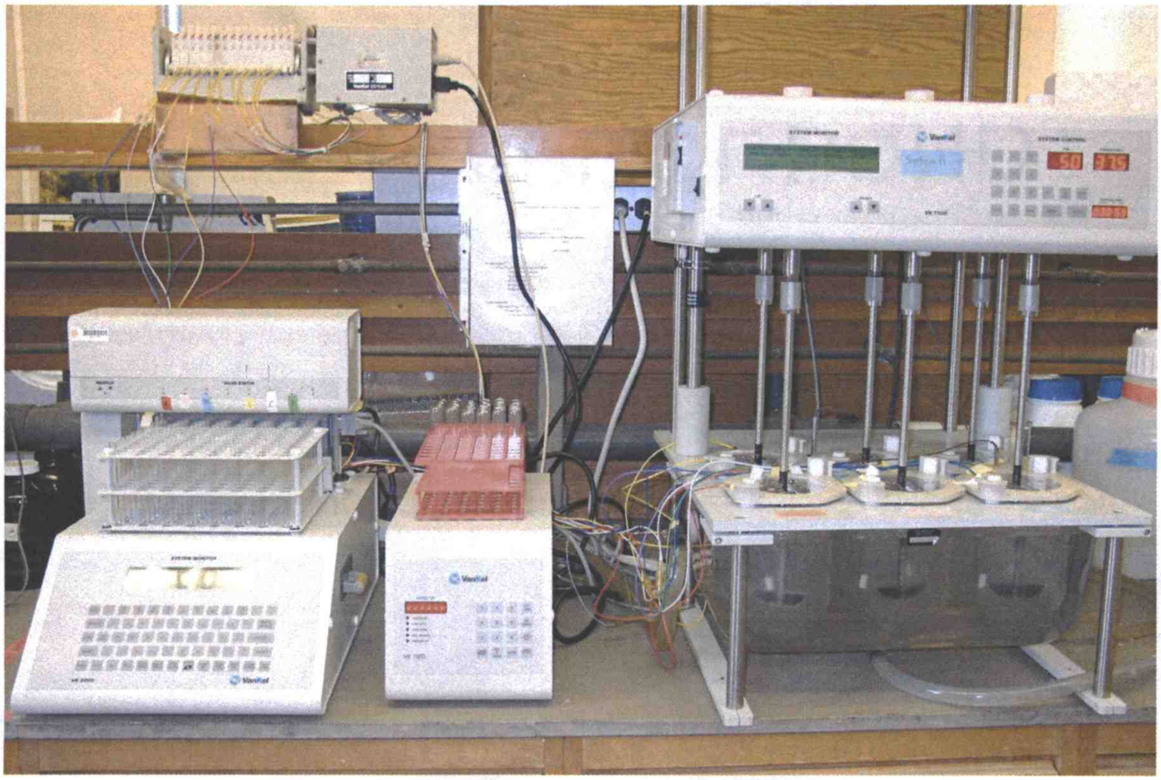


Figure 9: Dissolution machine, VanKel 7000 model

Goodness of fit

For the purpose of evaluating a dosage form with zero-order kinetics release, the dissolution profile is fitted by a linear regression equation:

$$A(\%) = \alpha + k_0 t$$

α: Intercept of the regression line
k_0: slope of the regression line

R^2 is one criteria of goodness of fit. R^2 is defined as the ratio Explained Sum of Squares divides by Total Sum of Squares. The Total Sum of Squares is equal to Explained Sum of Squares plus Sum Squared Residuals.

$$R^2 = \frac{ESS}{TSS}$$

$$ESS = \sum_{i=1}^{n} (\hat{Y}_i - \bar{Y})^2$$

$$SSR = \sum_{i=1}^{n} (Y_i - \hat{Y}_i)^2$$

$$TSS = \sum_{i=1}^{n} (Y_i - \bar{Y})^2 = ESS + SSR$$

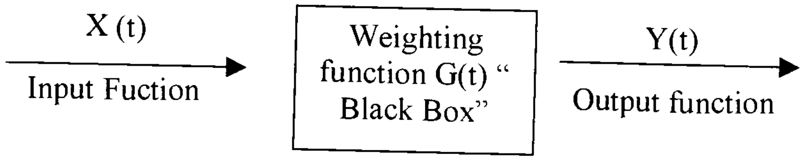
$$R^2 = \frac{ESS}{TSS} = 1 - \frac{SSR}{TSS}$$

The R^2 value is from zero to one. In the worst possible regression, the variation of the prediction of Y_i using X would capture none of the overall variation in the variation in Y_i. In this case, R^2 is equal zero. If $SSR = \sum_{i=1}^{n} (Y_i - \hat{Y}_i)^2 = 0$. If all values $Y_i = \hat{Y}_i$, the variation of the prediction of Y_i using X would capture all of the overall variation in Y_i and $R^2 = 1$. The closer R^2 is to 1, the better the goodness of fit .

All the prepared formulations were evaluated *in vitro* based on three criteria: the amount of drug released during a 24-hour period, the goodness of fit, and the "acceptance table" was also consulted to evaluate the dissolution profile.

Convolution and non-compartment analysis

Convolution is a simulation process. The schematic summary is as follows (53):



A black box is a physical system that transforms an input into an output. The input and the output are given by continuous or sectional continuous functions. If X(t) is the input function and Y(t) is the output function of a linear black box, then:

$$Y(t) = \int_0^t G(t-T)X(T)dT$$

It is said that Y(t) is given by the convolution between G(t) and X(t) where $0<T<t$.

If y(s), g(s), x(s) is the Laplace transformation of Y(t), G(t), X(t) and y(s), g(s), x(s) are called the transfer functions and its matrix is called the transfer matrix of the black box. The Laplace transformation equation is given by:

$$y(s) = g(s)x(s)$$

In pharmacokinetics, Y(t) is the function describing the plasma concentration-time curve following extravascular administration, X(t) is the function describing input (absorption function). During a time t, X(t) and Y(t) are continuous functions and the weighting function G(t) is normally a function for the concentration time curve describing bolus intravenous administration. In our case, linear pharmacokinetic compartments play as the linear black box.

The formulations with the most successful goodness of fit ($R \geq 0.95$) and total drug release over 80% were selected for the convolution step of the study in order to predict the plasma concentration-time curve for comparison with plasma concentration-time curves of two commercial products: Adalat® CC and Procardia® XL. Both dosage forms contain 30mg of nifedipine. The Kinetica software was used and programmed with the convolution functions. Dissolution profile (input function) and intravenous data (weighting function) are required, and predicted plasma concentrations versus time as the out put can be obtained by using Kinetic convolution.

“Non-compartmental analysis” is another function of Kinetica software. “Non-compartmental analysis” is used to calculate several pharmacokinetic parameters regardless of the number of compartments in the model that fits the drug concentration time curve. Required data for running non-compartmental analysis is a plasma concentration time profile. Therefore, the predicted plasma concentrations obtained in convolution process were used to calculate pharmacokinetic parameters using “non-compartmental analysis”. The parameters obtained were Half life ($t_{1/2}$), Mean Residence Time (MRT), Area Under the Curve (AUC), C_{max}, T_{max}, and Clearance (Cl).

CHAPTER 4: RESULTS AND DISCUSSIONS

Standard curve of nifedipine assay

Linearity of the UV absorbance of nifedipine with respect to its concentration has a direct impact on the accuracy of assay results. A prepared standard curve is a good tool to convert the magnitude of absorbance into real drug concentrations. The expected concentration of nifedipine is 30mg/1000ml in the dissolution flask, equal to 30 mcg/ml, so a range of eight solutions with well-known concentrations from 10 to 45 mcg/ml were prepared in 1% polysorbate 80 (Tween 80) solution. The absorbance of each solution is reported in Table 2. The standard curve and the regression equation of the line are presented in Figure 10. The coefficient of variation is calculated to be equal to 0.6686 and the linear coefficient, R^2 value is 0.9998. The linear relationship between the concentration of nifedipine and the absorbance is stable and the standard curve validates for nifedipine assay. Standard curve was regularly rebuilt every three months to account for fluctuation of energy in the spectrophotometer.

Table 2
Absorbance of nifedipine standard curve

concentration	Absorbance	Estimated concentration		%known concentration
10.1	0.1414	10.2027972		101.0178
15.15	0.2102	15.01398601		99.10222
20.2	0.2849	20.23776224		100.1869
25.25	0.3554	25.16783217		99.67458
30.3	0.4314	30.48251748		100.6024
35.35	0.4994	35.23776224		99.6825
40.4	0.5778	40.72027972		100.7928
45.45	0.6432	45.29370629		99.65612
			average	100.0894
			sd	0.668602
			%Cv	0.668005

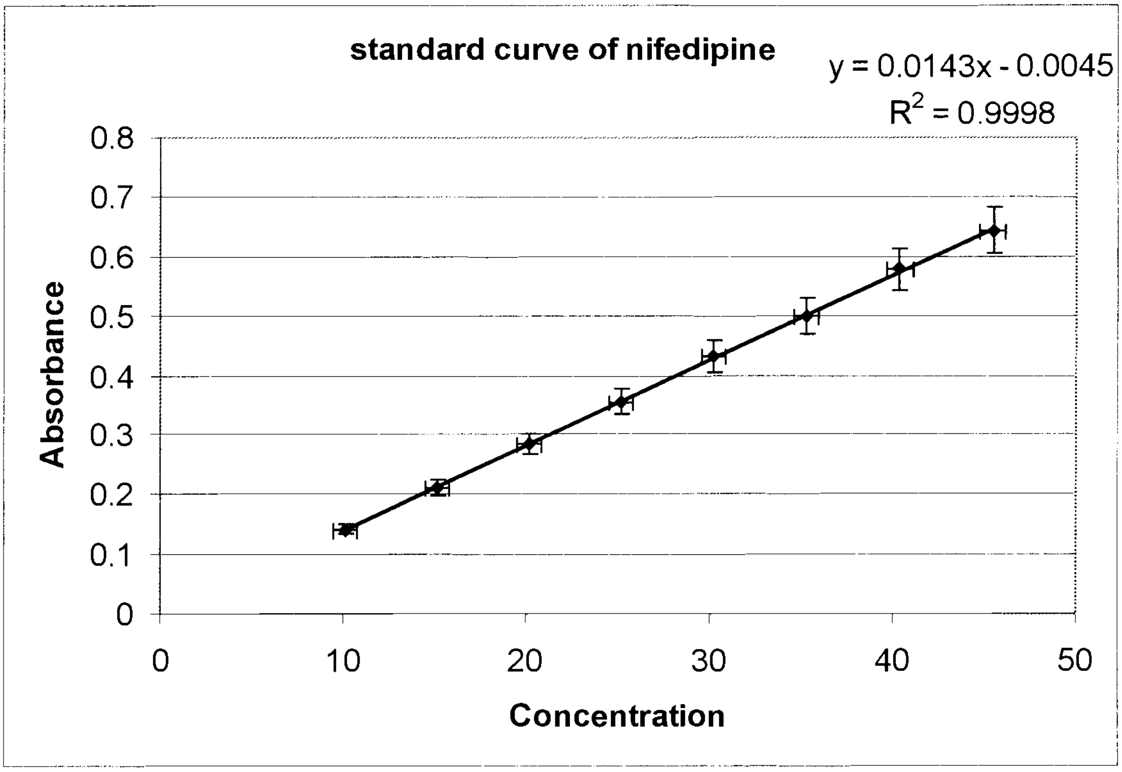


Figure 10: Standard curve and regression equation of nifedipine concentration vs absorbance

Dissolution profile of pure nifedipine

Since nifedipine dissolves very slowly in water and the solubility is extremely low, it is considered insoluble in water (2). By comparison, nifedipine is completely absorbed after oral administration (1). The profile of nifedipine dissolution may not reflect correctly the actual rate in which drug is absorbed *in vivo*. Therefore, a dissolution test of pure nifedipine in dissolution media was performed to confirm the appropriateness of dissolution as previously outlined. A rapid dissolution rate is plausible because it is likely to agree with *in vivo* absorption. Moreover, the dissolution profile of pure nifedipine is used as a useful reference to compare the rate of drug release from other dosage forms.

Table 3 reports the dissolution profile of 20mg pure nifedipine powder in 1000ml of 1% (w/w) polysorbate 80 water solution. Dissolution conditions were set up with the paddle rotating at 75 rpm. The graph depicting the dissolution profile is in Figure 11.

Table 3
Percentage of pure nifedipine dissolved during 24 hours at 75 rotations per minute

Time(hours)	Pure nifedipine
0	0
0.25	5.6987
0.5	12.659
1	25.6598
1.5	36.987
2	45.2365
3	50.36
4	53.6598
5	62.6598
6	64.6598
7	66.6541
8	68.6598
10	70.9874
12	72.3659
14	79.698
16	81.987
18	82.3694
20	84.6542
22	85.6598
24	86.9876

From the dissolution profile, it can be seen that the paddle speed of 75 rpm appears to be too slow because the drug does not dissolve totally within 24 hours and the dissolution rate is slow as well. If the dissolution paddle rotation rate is increased to 100 rpm, the stronger agitation should enhance the dissolution rate of nifedipine. Dissolution profile of pure nifedipine with the setting of 100 rpm for the dissolution paddle is shown in Table 4 and the dissolution profile is depicted in Figure 12.

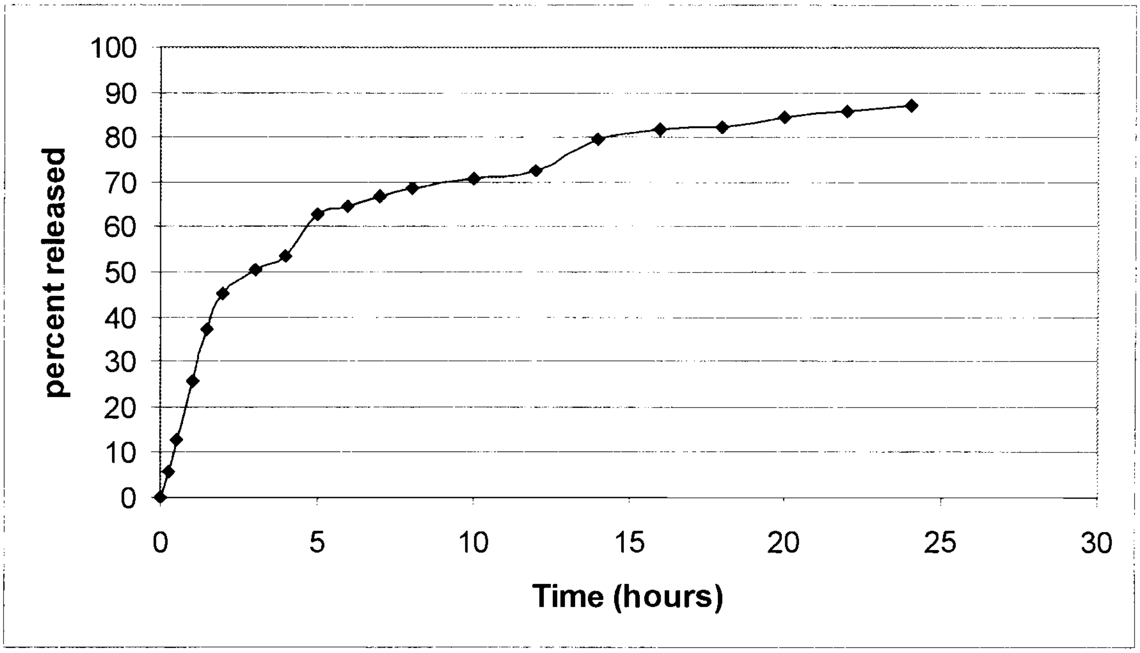


Figure 11: Dissolution profile of pure nifedipine with a paddle stir rate at 75 rpm during 24 hours

Table 4
Percentage of nifedipine released after 24 hours at 100 rotations per minute (rpm)

Time (hours)	Percentage released
0	0
0.25	30.41915
0.5	40.87435
0.75	49.02709
1	55.34133
1.5	64.33825
2	70.29048
3	83.76204
4	84.26229
5	87.13963
6	89.60019
7	91.84597
10	95.00843
22	96.77449
23	99.36995
24	100.7285

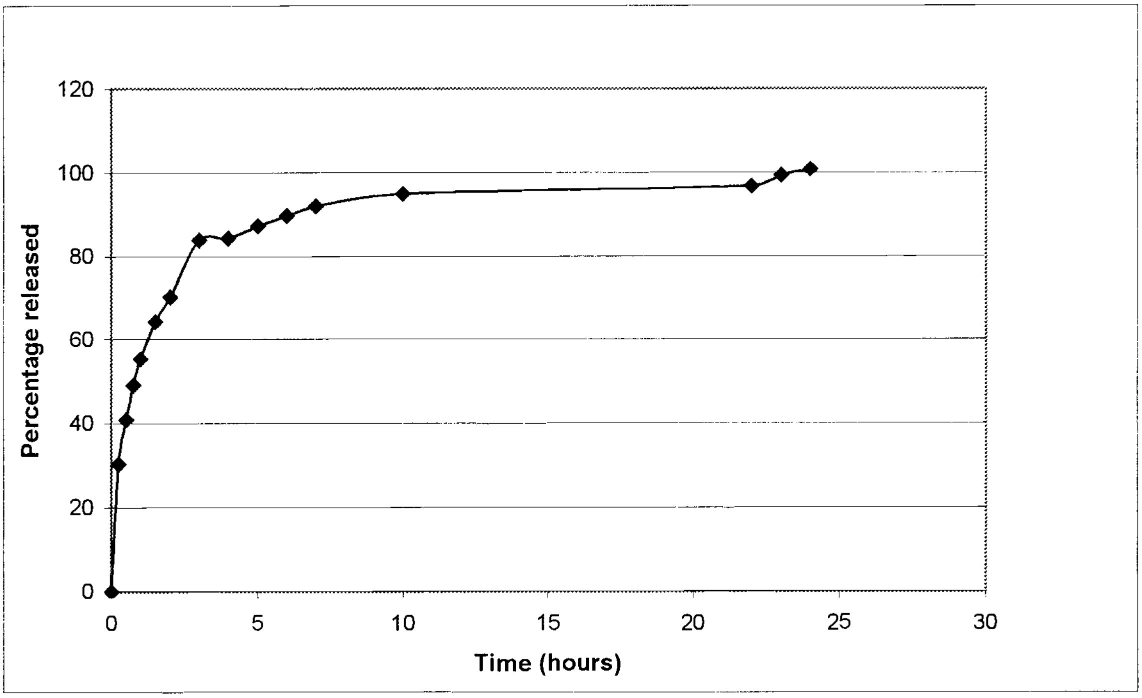


Figure 12: Dissolution profile of pure nifedipine at 100 rpm

Data above show that with the dissolution paddle set at 100 rpm, nifedipine dissolves more quickly compared with the setting of 75 rotations per minute. The 100 rotations per minute rate appears to be more appropriate for dissolution testing than the 75 rotations per minute setting and probably reflects better drug absorption *in vivo* because pure nifedipine dissolves more quickly and completely at the 100 rotations per minute rate. By itself, the dissolution profile of pure nifedipine is not only a good reference for testing dosage forms but it also makes sure that all the dissolution conditions are sufficient to protect nifedipine from degradation during the long duration of dissolution testing.

Dissolution profile of immediate release dosage form

The common immediate release dosage forms are soft elastic gelatin capsules wherein nifedipine was dissolved and was loaded inside the capsule.

The rate of nifeidpine release from an immediate release soft elastic gelatin capsule is predicted to be more rapid than the pure nifedipine powder.

The dissolution profile of Adalat® capsule is presented in Table 5 and Figure 13.

Table 5

Percentage of nifedipine released from immediate dosage forms Adalat® Capsule

Time (hours)	Percent released
0.05	22.41379
0.0833	24.82759
0.1667	91.67487
0.25	97.29064
0.333	99.26108
0.4167	100.00
0.5	99.95074
0.5833	100.5419
0.6667	99.75369
0.75	100.197

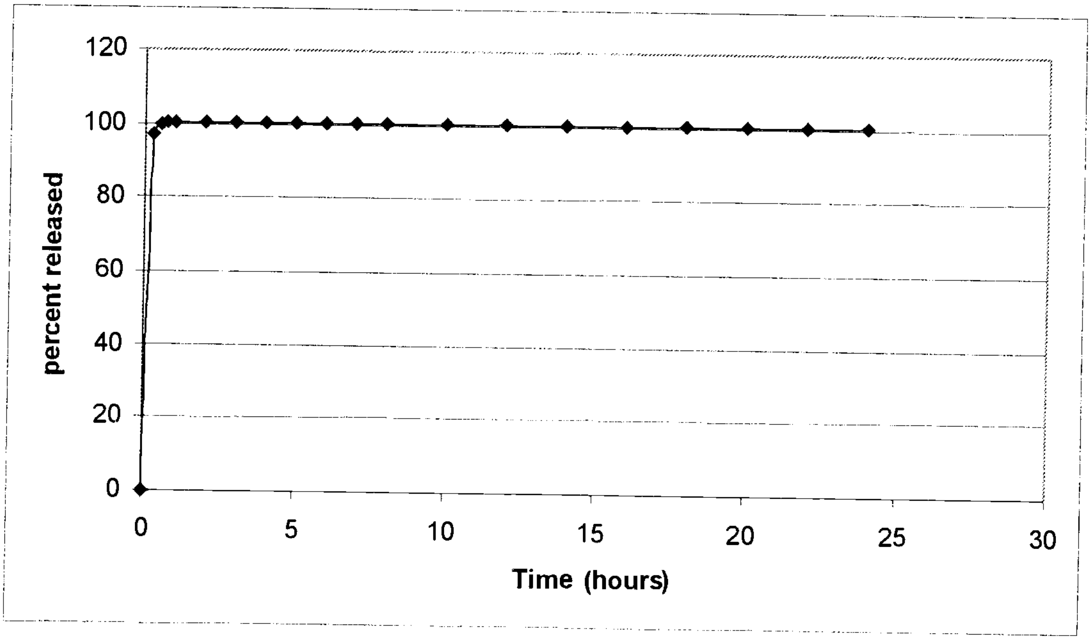


Figure 13: Dissolution profile of nifedipine from Adalat®CC Capsule

The dissolution profile of immediate release dosage form is also a reference to evaluate the rate of drug dissolved from the coated beads.

Investigation of the effect temperature that has on stability of nifedipine

One severe condition drugs have to be able to endure is the temperature of the hot-melt coating process. This method of formulation is negated if the drug is not resistant to heating. As mentioned above, the coating temperature in the direct blending method needs to be higher than the melting point of the wax by at least 10°C, with carnauba wax having the highest melting point at 83-86°C. For this reason, it is imperative to test the stability of nifedipine at the higher temperature range of about 90-95°C. In this investigation, a solution containing nifedipine within the normal detectable concentration range of light absorption at 334 nm (10-30 mcg/ml) was prepared. The actual solution of 20mcg/ml was used. An absorbance value of 0.2775 was measured at 334nm. This solution was kept at 95^{0}C away from light in a brown dark container for eight hours.

In an 8 hour time range, periodic absorbance values were measured for the 20mcg/ml nifedipine solution at the following intervals: 1, 2, 4, 6, 8 hours, during which nifedipine was kept away from light to protect it from deterioration.

The recorded absorbances are shown in Table 6

As a whole, the CV% is small between recorded UV absorbance readings. This shows that there is no significant difference in absorbance of drug from time zero to 8 hours. In conclusion, at temperatures as high as 95^{0}C, nifedipine does not show any significant degradation. So nifedipine could be used in the hot-melt coating process at high temperatures.

Table 6
The absorbance of nifedipine solution during 8 hours of storage at 95^{0}C

Time (hours)	Absorbance
0	0.2775
1	0.2778
2	0.2802
4	0.2790
6	0.2781
8	0.2791
Average	0.278617
Standard deviation	0.001007
CV%	0.36136

Investigating the effects of light on the degradation of nifedipine

It is well documented that nifedipine is not stable when exposed to ultraviolet or day light radiation. This characteristic of nifedipine was examined under normal laboratory light. A dissolution test on pure nifedipine was carried out in three amber colored dissolution flasks (protected from the light) and three samples of beads were put into three control transparent dissolution flasks, not protected from light. The experiment was conducted during normal day light hours and in the laboratory with overhead fluorescent light conditions. The purpose of control samples was to estimate the effectiveness of the light-protection system when performing the dissolution test.

The same dissolution testing procedures were carried out in the sample and control groups. Dissolution fluid samples were collected after 0.25, 0.5, 1, 2, 4, 6, and 8 hours. The samples were measured at the wavelength of 334 nm on UV- VIS spectrophotometer. Results are presented in Table 7.

The statistical treatment is paired "t" test. The test showed that there is a significant degradation of nifedipine observed in the transparent dissolution flasks (p=0.000001 and CI 95%: 0.060599 -0.1197458). This confirmed that nifedipine is sensitive to light. The amber colored dissolution flasks were able to protect nifedipine during dissolution tests without any additional external covers or protection procedures.

Table 7
Absorbance of dissolution samples from beads protected and not-protected from light

Time (hours)	protected beads' Absorbance	Non-protected beads' absorbance
0	0	0
0.25	0.0717	0.0235
0.5	0.0994	0.0356
1	0.1093	0.0416
1.5	0.1184	0.041
2	0.1447	0.0512
3	0.1628	0.0503
4	0.177	0.0645
5	0.1883	0.0532
6	0.1973	0.0588
7	0.2017	0.0590
8	0.1998	0.0503

Effect of coating agent nature on the weight gain

The maximum weight gain, and the percentage of coating materials that can be loaded onto nonpareil spherical sugar beads also needs to be considered. The affinity of coating material to adhere onto beads when used depends on the nature of coating agents themselves.

Nifedipine was mixed with waxes at a wax/drug ratio of 8/7 (7 grams of nifedipine, 8 grams of coating materials, 100 grams nonpareils-yields a 15% weight gain) and 15/7 (7 grams of nifedipine, 15 grams of coating materials, 100 grams nonpareils-yields a 22% weight gain). The actual percentage of drug loaded onto beads was determined according to the assay in the methods section. The actual percentage of drug loaded divided by the theoretical or expected percentage of drug that should have been loaded is equal to the effectiveness of hot-melt coating process or the relative percentage of coating mixture deposited. This is presented in Table 8 and Figure 14.

Table 8
Relative percent of coating mixture deposited on sugar bead substrates

Wax (15g)	**Nifedipine (7g) + wax (8g)**	**Nifedipine (7g) + wax (15g)**
Gelucire	80.1%	75.5%
Emulsifying wax	81.3%	76.5%
Stearic acid	76.3%	67.4.%
Acid Triglyceride syncrowax	78.1%	70.0%
Carnauba wax	75.2%	72.2%
Synthetic beewax	70.8%	69.3%
Natural beewax	66.4%	65.7%

By measured observation, the weight gain seen for single coating layer is up to 22%. Application of a coating of more than 22% caused agglomeration of coated beads in the current process which occurs remarkably quickly during cooling even with a stirring rod mixing. Applying a coating over 22 % also reduces the amount of wax deposited on the sugar beads.

For carnauba wax and Acid Triglyceride when the weight gain increases to 22%, the percentage of coating agent and drug deposited on the beads is over 70% of the theoretical value. For Gelucire 53/13, and Emulsifying wax, the weight gain is greater than 75% whereas stearic acid is barely over 65% of the theoretical value. It should also be noted that the ratio of nifedipine to the wax affects the actual weight gain as well.

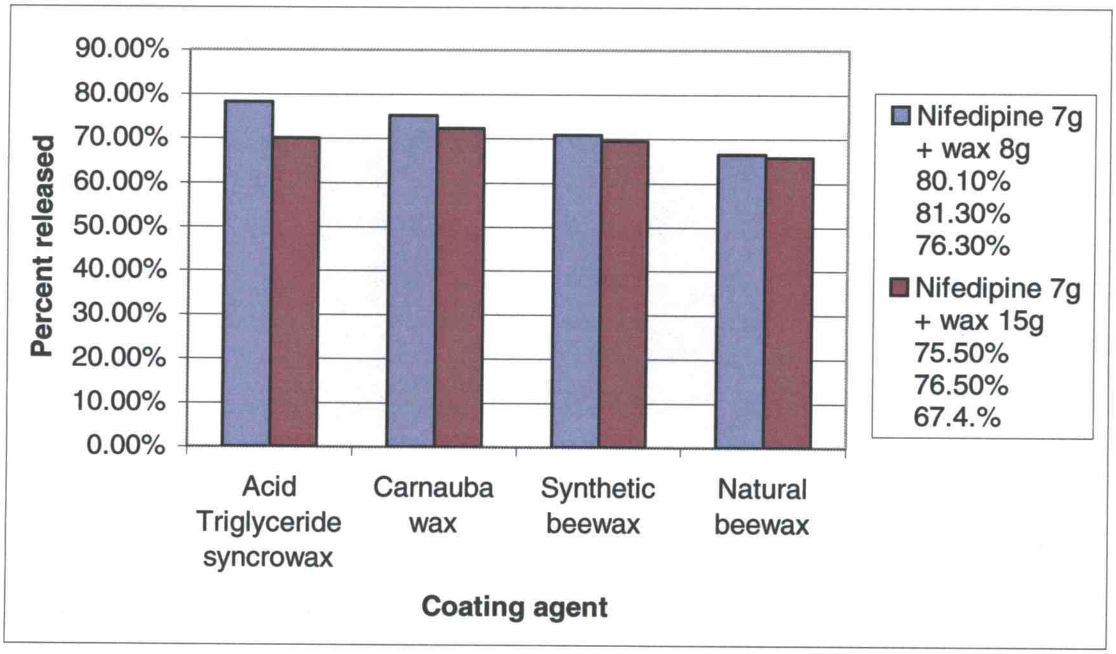


Figure 14: Relative drug deposited at two weight gain values: 15% and 22%

The more nifedipine added into the drug:wax mixture a greater reduction of actual weight gain onto sugar beads occurs since nifedipine itself reduces the stickiness of the mixture.

The effect of different waxes on the dissolution rate of nifedipine

The *in vivo* GI absorption of drugs has been well studied for decades. The process includes active and passive absorption. Both processes require the absorbed substances to be dissolved. For example, lipids are emulsified by biliary salts. It is documented that the *in vivo* absorption profile is closely related to the dissolution rate *in vitro* for many drugs and some dissolution systems. As mentioned earlier, waxes with high rigidity, hydrophobicity, and a higher melting point tend to delay the dissolution rate of loaded drug on beads longer. This is very useful in formulation since adjustment of the dissolution rate is related to a change of the ratio of coating agents/substrates or the ratio of coating agents/drug used in the formulation. To estimate the relationship of physicochemical characteristics of various coating materials and their effects on dissolution rate, seven single layer coated bead formulations were prepared according to the procedures mentioned in "materials and methods" section. All beads were prepared having the same ratio between wax and drug 2:1 (15/7), and the weight gain was 22%. The formulations are summarized in Table 9. The only difference in these formulations is the coating materials: Gelucire 53/12, emulsified wax, stearic acid, triglyceride, carnauba, synthetic beeswax and natural beeswax.

Table 9
Single layer nifedipine coated beads with different types of waxes

Formulation	**Sugar bead (100g)**	**Wax (15g)**	**Nifedipine (7g)**	**% weight gain**	**Theoretical percent of drug in the beads**	**Actual percent of drug in the bead**
1	x	Gelucire	x	22%	5.7377	75.5%
2	x	Emulsifying wax	x	22%	5.7377	76.5%
3	x	Stearic acid	x	22%	5.7377	67.4.%
4	x	Acid Triglyceride syncrowax	x	22%	5.7377	70.0%
5	x	Carnauba wax	x	22%	5.7377	72.2%
6	x	Synthetic beeswax	x	22%	5.7377	69.3%
7	x	Natural beeswax	x	22%	5.7377	65.7%

The coating process was carried out exactly as reported with the single coating layer process. Waxes were preheated and melted in a beaker, then nifedipine was mixed into the coating agents to form a homogenous mixture, then beads were poured in while the mixture was stirred by hand with a glass stirring rod. The beads were cooled to the room temperature with continued stirring and then sieved through a number 25 mesh screen to eliminate the few agglomerated beads that formed.

The prepared beads were evaluated by means of their dissolution profiles of nifedipine. The difference in dissolution profiles was much more observable with the slower dissolution test setting at 75 rpm. The percentage of nifedipine dissolved from each test formulation within 24 hours is shown in Table 10.

Table 10
Percentage of nifedipine released from seven formulations at 75 rpm paddle rotation speed

Time (hours)	Pure material	Gelucire	Emulsified wax	Stearic acid	Acid Triglyceride	Carnauba	Synthetic beeswax	Natural beewax	IR capsule
0	0	0	0	0	0	0	0	0	0
0.25	5.6987	6	4.462903	4.476469	5.362647	6.809552	6.322202	3.968059	97.29064
0.5	12.659	21.3895	11.67953	6.986502	6.466803	8.288515	12.49318	5.562801	99.95074
1	25.6598	27.3256	22.79506	11.34171	7.248909	9.404441	15.43188	9.057143	100.197
1.5	36.987	31.2365	32.64803	15.57594	8.679785	10.57514	20.39326	11.56072	100.197
2	45.2365	38.2156	39.02819	19.77104	10.28052	11.46484	23.82099	13.38493	100.197
3	50.36	48.2351	45.68564	26.92718	12.91064	15.1943	27.1695	17.46263	100.197
4	53.6598	51.67048	53.36083	31.30091	15.23132	15.88234	28.75619	20.65602	100.197
5	62.6598	52.85914	58.00632	35.27667	16.86394	17.23819	31.21426	24.24231	100.197
6	64.6598	54.43749	59.68583	38.94771	18.6431	19.18374	32.56064	26.00785	100.197
7	66.6541	56.81592	61.07675	39.59649	20.02315	20.24477	34.19748	28.51773	100.197
8	68.6598	62.9568	66.598	45.04001	24.27565	22.09271	36.4568	32.65987	100.197
10	70.9874	65.12365	69.659	49.25931	26.88315	24.44905	37.3215	32.6598	100.197
12	72.3659	66.6259	70.21465	50.36342	29.88446	25.95681	38.26548	34.6598	100.197
14	79.698	71.2654	77.6585	54.20285	33.04218	28.19167	39.12658	37.1268	100.197
16	81.987	74.3625	78.25997	54.43626	35.78913	29.67009	39.32654	40.36548	100.197
18	82.3694	77.6549	80.23156	57.6105	38.56482	32.39343	40.65489	42.3265	100.197
20	84.6542	81.6254	83.1256	60.96267	41.63734	33.97906	42.3659	44.6987	100.197
22	85.6598	84.6254	84.6598	62.50208	44.81126	36.74603	44.65987	45.1478	100.197
24	86.9876	84.6598	85.6598	64.81108	48.17387	38.6597	45.65849	47.86541	100.197

The dissolution profile of seven formulations is presented in Figure 15

Drug deposited on the sugar beads was over 70% generally, but nifedipine deposited with Natural beeswax and synthetic beeswax was less than 70%. In regards to the dissolution profile, carnauba wax-coated beads yielded the slowest release rate of the drug. Nifedipine dissolution rate from emulsifying wax was similar to the dissolution profile of pure nifedipine at 75 prm. Coated beads containing Emulsifying wax had the highest total amount of drug release, approximately 86% which is equal to that of pure nifedipine.

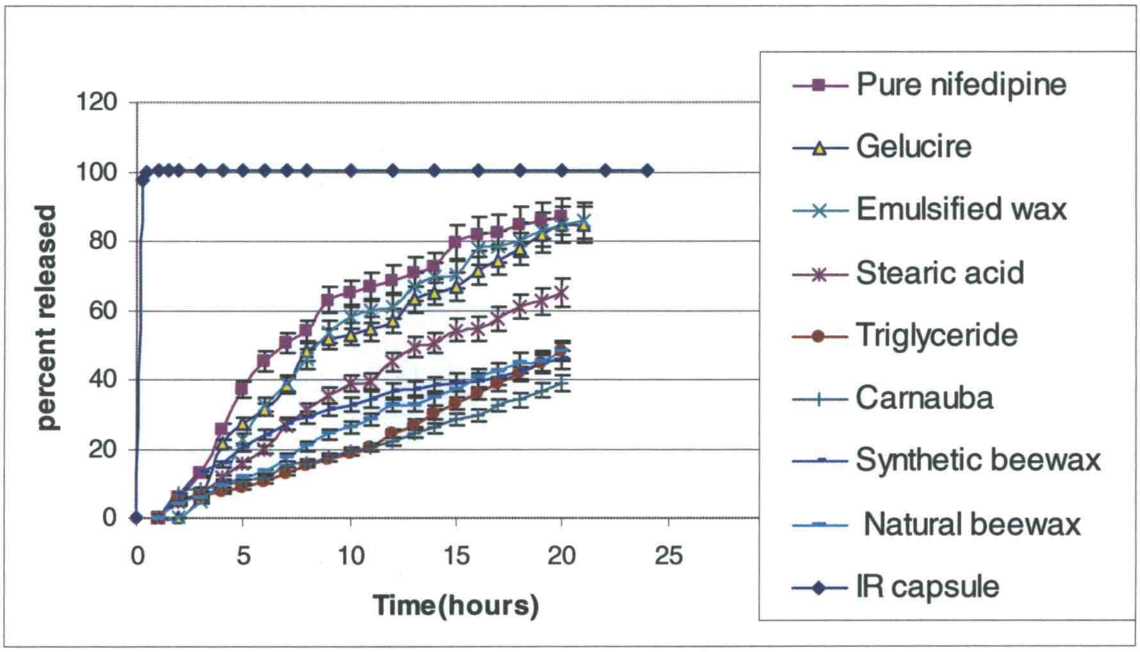


Figure 15: Dissolution profile of nifedipine from seven formulas with different coating materials

Similar to Emulsifying wax, Gelucire 53/13 did not reduce the dissolution rate of nifedipine much compared to pure drug, as the percentage of nifedipine released was 85%. Stearic acid had a moderate effect in retarding nifedipine release (64%). Carnauba wax coated beads gave the lowest rate of drug release, and the lowest percentage of dissolved drug, only 38% in 24 hours. For synthetic beeswax and natural beeswax, 47% of the drug was released. The obtained dissolution profiles of most formulations coincide with what was expected based on early pilot experiments and wax characteristics.

with what was expected based on early pilot experiments and wax characteristics. Among the selected waxes, carnauba wax has highest melting point (83-86°C), the highest rigidity, and hydrophobicity. Consequently, its coating layer was not easy to erode in water and the beads retain the drug for the longest time compared to other coating agents. Ranking right after carnauba is Acid Triglyceride with the next highest melting point (70-75°C) and its rigidity is rather high. An exception in the study is natural beeswax. Its melting point is higher than (80°C) Acid Triglyceride but it did not retard nifedipine release as well as Acid Triglyceride. Emulsifying wax is the most hydrophilic material among trial waxes, so it did not suppress drug release effectively. From the data, in order to develop a prolonged release dosage form, it is recommended that Acid Triglyceride, stearic acid, carnauba, natural beeswax, synthetic beeswax, or mixtures of these waxes can be used.

Investigation of the effect of pH on the drug dissolution profile

There have not been any published papers investigating the effect of pH on the dissolution profile of hot-melt coated beads containing nifedipine. The release of drug from the beads may vary with pH since nifedipine is an organic base and is ionized in gastric fluid but not in intestinal fluid. Therefore, an investigation of nifedipine release from coated beads was conducted in three 1% Tween 80 solutions: Simulated gastric fluid (pH = 1.5), simulated intestinal fluid (pH = 7.4) and water (slightly acid, pH = 6.5). The stearic acid coated beads were made by direct hot-melt coating method. The formulation consisted of substrate: sugar beads 100grams, coating ingredients: stearic acid 15 grams, nifedipine 7grams. The beads were prepared following the single coating layer procedure in "Materials and methods" section. Stearic acid was melted and homogenously mixed in with nifedipine. The mixture was coated onto the sugar nonpareil beads. Finally, the cooled coated beads were sieved through number 25 mesh screen. Dissolution profiles of nifedipine in the three media are reported in Table 11 and the dissolution profile presented in Figure 16.

Table 11
Percentage of nifedipine released over 24 hours in three media of varying pH

Time (hours)	Water	Simulated Gastric Fluid	Simulated intestinal Fluid
0	0	0	0
0.25	5.312517	3.321456	4.36587
0.5	7.688188	6.32654	7.321456
1	10.59207	8.214563	11.32654
1.5	13.75205	12.698741	14.365987
2	21.12076	20.32456	22.36541
3	28.97586	26.321415	28.845216
4	35.31427	33.321456	36.95746
5	43.67114	40.698547	42.654789
6	50.02058	50.62453	48.365984
7	53.33195	52.321487	53.21456
8	58.55656	56.857412	57.654974
10	61.03917	62.36541	60.324568
12	63.99588	64.321478	63.698741
14	66.90241	68.587463	65.321456
16	70.11542	72.654789	69.654475
18	74.29991	75.321456	74.326549
20	77.006	78.451632	77.958462
22	78.09912	79.354789	79.654389
24	79.9219	81.369577	82.365479

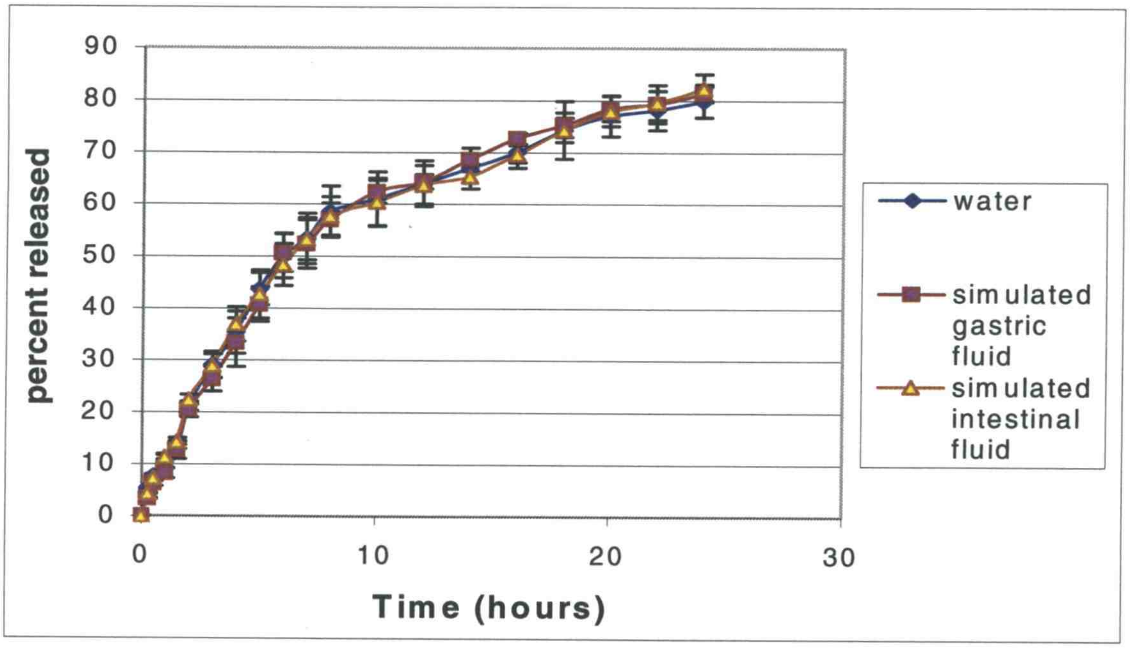


Figure 16: Nifedipine dissolution profile from stearic acid coated beads in three different media of varying pH

The total percentage of nifedipine released in the three media is presented in Figure 17

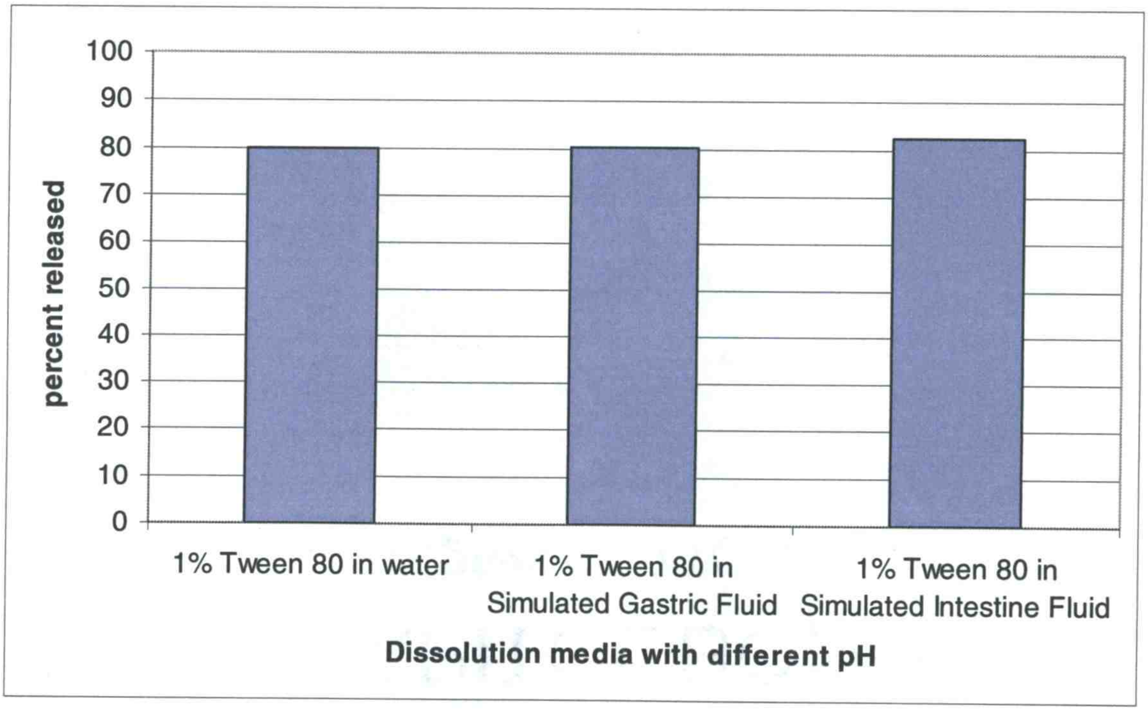


Figure 17: Total amount of nifedipine released from stearic acid coated beads in three different pH media

The nifedipine dissolution profiles from stearic acid coated beads in three pH values did not reveal any significant differences. The total percentage of drug released was 79.92%, 81.37% and 82.37% in water with 1% Tween 80, acidic simulated gastric fluid with1% Tween 80 and simulated intestinal fluid with 1% Tween 80, respectively. The variability among all specific values at specific time points was small (all the standard deviations are smaller than 2%). It appears that the pH does not have an effect on the nifedipine dissolution process from wax coated beads.

Investigating the change in the ratio of waxes to drug release

From the results presented in previous sections for hot-melt coating, the characteristics of the waxes are useful information for developers. The interaction between water and the coated beads strongly depends on the polarity of the coating agents' surface. Rigidity and hydrophilic properties of the coating materials influence the disintegration and the dissolution of drug from the beads. Hence, a binary mixture of waxes should have some characteristics of both coating materials. If no chemical reactions occur, the expected rigidity and hydrophobicity will lie between the two coating substances and an adjustment of the ratio of the two waxes may create a desirable drug dissolution rate. Stearic acid and Gelucire 50/13 were chosen to evaluate this prediction. Four different single layer coated beads were prepared. These beads were prepared with the same weight gain (22%) and ratio of total amount waxes/nifedipine/coating material of 15/7/100 (7gr of nifedipine, 15gr of mixture coating materials, 100gr of sugar beads). The only modification is the ratio of stearic acid to Gelucire 50/13 among these formulations. Specifically, the sugar beads were coated by the mixture of nifedipine and pure coating substances: stearic acid or Gelucire 53/13. In two other formulations, the mixture of stearic acid/ Gelucire 50/13 with the ratio 5/1 and 4/2 were the coating agents (Table 12). The preparation of these beads follows all the steps of single layer coated beads as described in the "Materials and Methods" section. Previously, it was shown that stearic acid delayed the rate of drug release longer than Gelucire 50/13. The binary mixtures of these waxes when used in hot-melt coating should have dissolution profiles faster than that of stearic acid and longer than that of Gelucire 53/13. Once again, dissolution tests were carried out at 75 rpm and the percentage of nifedipine dissolved measured. The values are presented in Table 13.

Table 12
Coated beads with different ratios of stearic acid and Gelucire 53/13

Formu-lation	Sugar bead (100g)	Wax (15g)	Nifedipine (7g)	% Gain weight	Theoretical percentage of drug in the bead	Actual percentage of drug deposited on the bead
1	x	Gelucire	x	22%	5.7377	80.3
2	x	Stearic acid	x	22%	5.7377	64%
3	x	Stearic acid : Gelucire 50/13 (5:1)	x	22%	5.7377	79%
4	x	Stearic acid : Gelucire 50/13 (4:2)	x	22%	5.7377	74%

Table 13
Release profile of nifedipine from coated beads with different ratios of stearic acid to Gelucire 50/13

Time (hours)	Gelucire	Stearic acid	Stearic acid : Gelucire (5:1)	Stearic acid : Gelucire (4:2)
0	0	0	0	0
0.25	6	4.476469	4.863435	9.182658
0.5	21.3895	6.986502	6.522333	11.49321
1	27.3256	11.34171	12.32498	18.20525
1.5	31.2365	15.57594	16.6693	23.93523
2	38.2156	19.77104	19.47145	26.28898
3	48.2351	26.92718	23.49703	36.11723
4	51.67048	31.30091	28.62088	43.73516
5	52.85914	35.27667	32.0789	48.24491
6	54.43749	38.94771	37.01887	53.0168
7	56.81592	39.59649	39.24971	55.83549
8	62.9568	45.04001	46.6587	56.2659
10	65.12365	49.25931	49.9875	58.36249
12	66.6259	50.36342	51.6597	61.6594
14	71.2654	54.20285	55.6987	63.45699
16	74.3625	54.43626	56.9874	67.6549
18	77.6549	57.6105	59.8745	72.36549
20	81.6254	60.96267	61.9874	74.2659
22	84.6254	62.50208	64.9587	76.6548
24	84.6598	64.81108	67.9874	78.6548

The dissolution profiles within 24 hours of four formulations are shown in Figure 18.

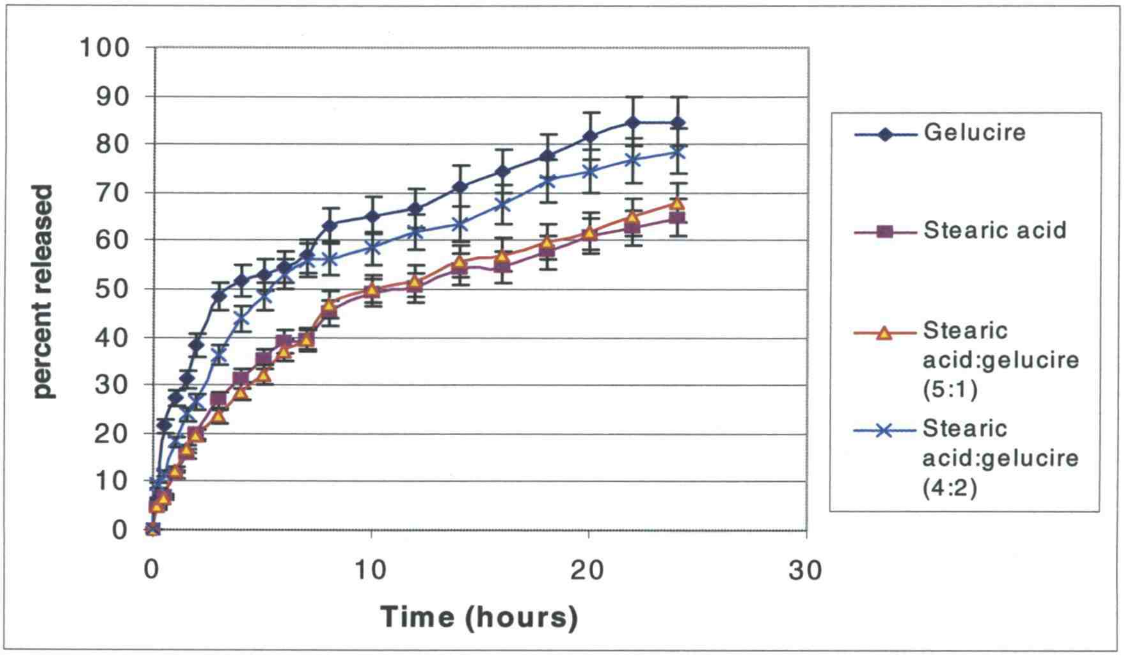


Figure 18: Nifedipine dissolution profiles from beads with different ratios of coating agents, stearic acid and Gelucire 50/13

The obtained results were similar to what was expected. The percentage of dissolved nifedipine within 24 hours was highest with the coating agent solely of Gelucire 50/13 (85%) and lowest with solely stearic acid (64%). As the ratio of stearic acid to Gelucire 50/13 increased, the dissolution rate decreased (79% with ratio 4/2 and 68% with the ratio 5/1). This confirms the deduction that coating the beads with a mixture of two different waxes containing drug will have a dissolution rate between the upper and the lower dissolution rates observed using each as a pure single wax coating material.

It is also noted that the small amount of Gelucire 53/13 mixed with stearic acid (the formulation stearic acid: Gelucire 50/13 of 5/1) did not change significantly the nifedipine dissolution rate compared with pure stearic acid coated beads.

Preparation of coated beads in order to obtain zero-order sustained release of nifedipine

Single layer coated dosage form

To develop a sustained release dosage form of nifedipine using sugar beads as substrates, the amount of drug loading and the upper weight limit of coating that can be added to the dosage form beads is an important consideration. The minimum amount of drug in a sustained release dose of nifedipine is 30mg. The largest weight of a capsule is about one gram, so the minimum amount of drug to deposit on the sugar beads is 30mg/1000mg = 3 %. The drug percentage in the mixture of wax and drug is estimated to be smaller than 50%. So the percentage of coating agents (mixture of nifedipine and waxes) is minimum at 2 * 3% = 6%. The capacity of coating materials adhering to the sugar beads during the laboratory coating process is about 70%. Thus, the minimum total theoretical weight gain is 6%/0.7 = 9%. If 7 grams of waxes are used, the quantity of nifedipine used in coating is 7 grams for 100grams of sugar beads. This will yield the theoretical percentage of nifedipine to be loaded onto sugar beads of about 6% and the actual percentage of nifedipine that should be on 100g of sugar beads will be about 4-5%.

Using single layer coated beads, with the criteria to select the dissolution profile and potential coating agents from the previous section “The effect of different waxes on the dissolution of nifedipine”, a series of single coated beads were evaluated. Gelucire 50/13 and Emulsifying wax had only slight effect on delaying the release of nifedipine from the sugar beads. The retardation of nifedipine release increases with an increasing amount of wax in the coating layer. However, when the weight gain reaches 22% or higher (in Table 8) agglomeration of beads can occur. Hence, more rigid coating agents are preferred to delay drug release. Stearic acid was the first coating material chosen for this experiment. Nifedipine was mixed with molten stearic acid. Then, sugar beads were coated with this mixture by direct blending method. The ratios, wax/drug, in the coating layer were 7/7 (100g sugar beads, 7g stearic acid, 7g nifedipine) and 15/7 (100g sugar beads, 15g stearic acid, 7g nifedipine). The dissolution criteria were as outlined in the " Materials and Methods" section. Because, at the 100 rpm dissolution paddle speed, pure nifedipine dissolves relatively rapidly, it is likely to reflect better the absorption that occurs *in vivo* than the 75 rpm rate, so the 100rpm rate was selected. Table 14 and Figure 19 present the percentage of drug release. The pure material is used as the reference.

Table 14
Percentage of nifedipine released from stearic acid coated beads

Time (hours)	nifedipine: stearic acid (7:7)	nifedipine: stearic acid (7:15)	pure nifedipine
0	0	0	0
0.25	11.97469	5.312517	30.41915
0.5	21.28794	7.688188	40.3265
1	34.79209	10.59207	49.02709
1.5	37.92421	13.75205	55.34133
2	54.54496	21.12076	64.33825
3	64.79101	28.97586	70.29048
4	71.48617	35.31427	83.76204
5	78.32221	43.67114	84.26229
6	80.93965	50.02058	87.13963
7	82.29152	53.33195	89.60019
8	85.5724	58.55656	91.84597
10	90.78644	61.03917	95.00843
12	92.23214	63.99588	95.0021
14	93.4636	66.90241	95.3214
16	94.27527	70.11542	95.6543
18	95.02657	74.29991	96.2365
20	95.67526	77.006	96.4532
22	97.30536	78.09912	96.77449
24	99.1294	79.9219	100.7285

The ratio of stearic acid/drug equal to 7/7 (1:1) was not as successful as desired in slowing nifedipine release from the beads and did little to restrain nifedipine from dissolving and diffusing into dissolution media. The ratio 15/7, (2:1) of coating material to drug in the formulation slowed the dissolution of nifedipine significantly. However, at that ratio, (2:1) coating material to drug, the dissolution profile substantially deviated from desired zero-order release kinetics ($R^2 = 0.8559$) and the release pattern did not compare favorably with the criteria for nifedipine extended-release tablets in USP25-NF20. The total percentage of dissolved nifedipine was not complete even after 24 hours (<80%) and the rate of drug release is high in the first four hours (35%), compared to the value in the "acceptance table" (17%). The low R^2 demonstrates that this formulation did not satisfy zero-order kinetics of drug release.

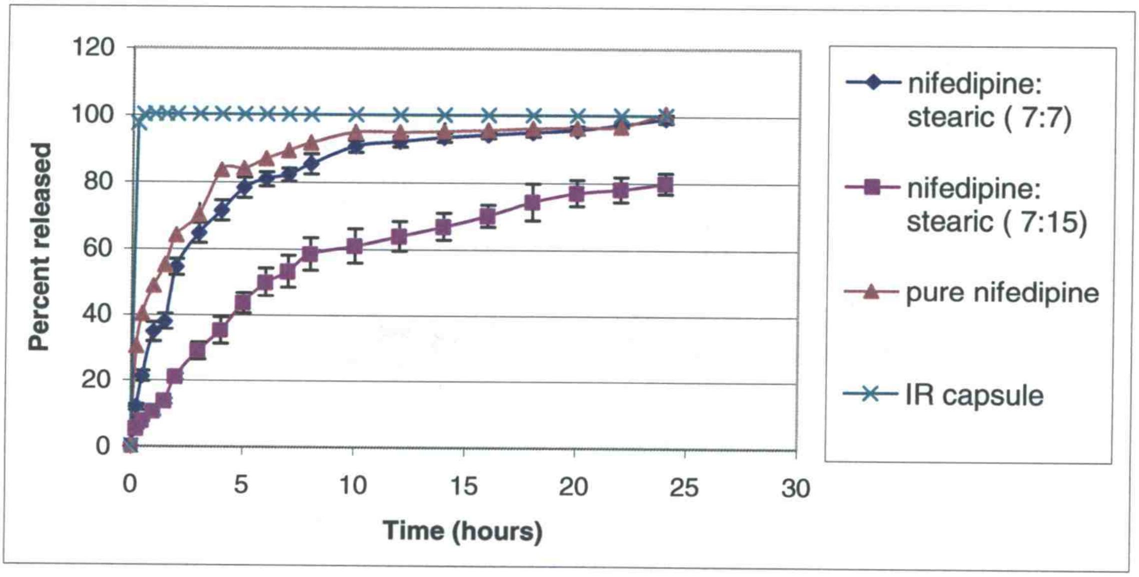


Figure 19: Nifedipine dissolution profiles from stearic acid coated beads

The coefficient (R^2) used to evaluate linearity is shown in Table 15.

Table15
R^2 value of stearic acid coated beads

	Nifedipine: stearic (7:7)	Nifedipine: stearic (7:15)	Pure nifedipine
R^2	0.6505	0.8559	0.535

Another synthetic wax, Acid Triglyceride was chosen as a coating agent for single layer coating of nifedipine onto sugar beads. Acid Triglyceride is more hydrophobic than stearic acid with a higher melting point. The desire is to keep nifedipine in the coated layer longer than that observed when stearic acid was used as the coating agent. Five formulations of single layer coated beads using Acid Triglyceride as a coating agent were prepared. In these formulations, the nifedipine and sugar beads quantity was fixed (7grams and 100 grams). The amount of Acid Triglyceride was modified from 3 to 15 grams. These formulations are reported in Table 16.

Table 16
Five formulations with different ratios of nifedipine and Acid Triglyceride

Formu -lation	Sugar bead (100gr)	Wax - triglyceride	Nifedi- pine (gr)	% Weight gain	Theoretical percent of drug in the beads	Actual percent of drug in the bead
1	x	15 g	7	22%	5.7377	70.0%
2	x	7g	7	14%	6.140	78.5%
3	x	5.5g	7	12.5	6.222	65.57%
4	x	4	7	11	6.306	50.62%
5	x	3	7	10	6.364	51.02%

The actual contents (relative percentage of drug deposited) of nifedipine in the five single layer acid triglyceride formulations were also analyzed. These values ranged from 50% to 70% of theoretical loading. Percentage of nifedipine released from the five formulations is shown in Table 17.

Table 17
Percentage of nifedipine released from Acid Triglyceride coated beads

Time (hours)	Formulation 1 (Triglyceride-nifedipine 15:7)	Formulation 2 (Triglyceride-nifedipine 7:7)	Formulation 3 (Triglyceride-nifedipine 5.5:7)	Formulation 4 (Triglyceride nifedipine 4:7)	Formulation 5 (Triglyceride-nifedipine 3:7)
0	0	0	0	0	0
0.25	2.876478	3.312665	7.084636	4.746405	9.20905
0.5	3.745363	5.672745	8.185	5.556063	16.86955
0.75	4.786177	6.292555	9.246152	6.36231	21.45759
1	6.882705	7.78342	10.16442	12.17847	28.35744
2	10.83146	10.86126	13.84694	16.43524	39.33822
3	12.67532	14.7278	17.03645	19.92445	44.19078
4	15.50577	17.54675	20.23702	24.4708	50.4964
5	19.00069	20.7552	23.11766	28.43947	55.61991
6	21.19172	24.06457	26.32595	32.62721	59.30279
7	24.56451	27.71218	29.36638	37.49588	64.02353
8	26.16501	30.09796	31.89405	41.54646	66.85281
10	31.60745	35.61113	37.30503	46.42731	70.87132
12	35.25171	40.27065	42.9028	51.88228	73.75016
14	40.48879	44.68646	48.00788	54.30814	75.56958
16	43.43194	49.21164	53.01791	57.80894	77.29854
18	47.22814	53.15085	58.17097	61.92428	77.38925
20	52.16663	57.3399	62.89123	64.56775	78.58562
22	54.79144	62.54878	66.50082	70.13837	80.36034
24	59.29954	65.52178	70.56985	72.29664	83.56126

The dissolution profile of these formulations is presented in Figure 20.

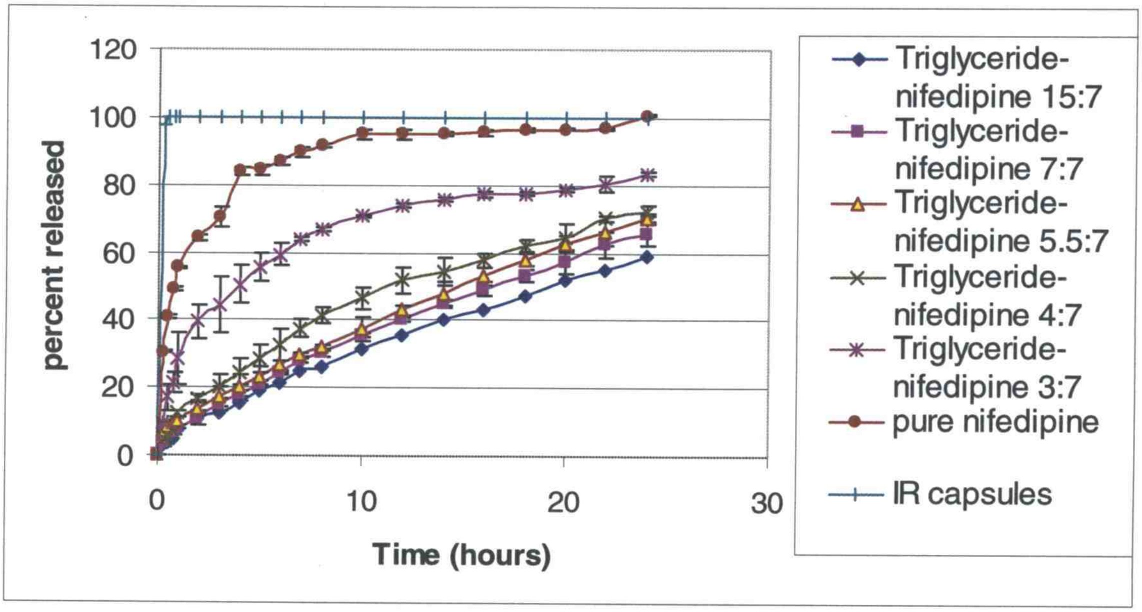


Figure 20: Dissolution profile of five formulations with different ratios of nifedipine to Acid Triglyceride

The R^2 value again was used to evaluate the linearity of the drug dissolution profile. R^2 of all formulations and pure nifedipine dissolution profiles are presented in Table 18.

Table 18
R^2 value of dissolution linearity of four formulations and pure nifedipine

	Formulation 1	formulation 2	formulation 3	formulation 4	formulation 5	pure nifedipine
R2	0.9858	0.9856	0.988	0.9459	0.7527	0.535

Figure 21 presents the total percentage of drug released within 24 hours from the prepared formulations.

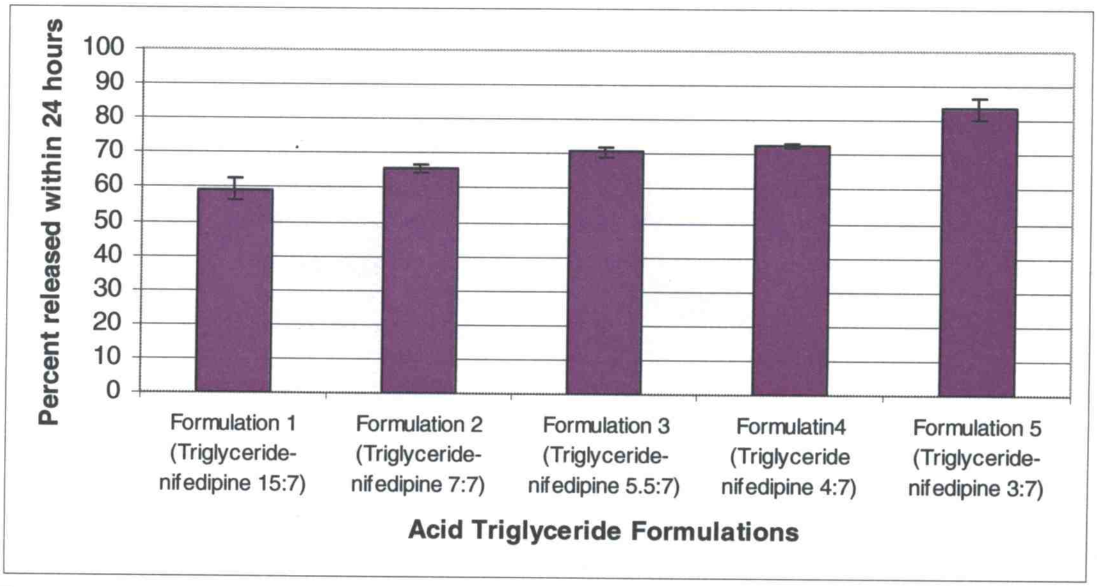


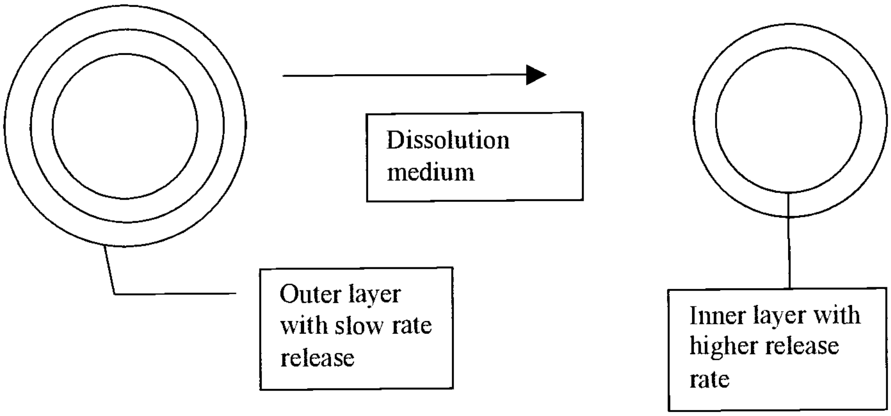
Figure 21: Total percentage released of nifedipine over 24 hours of dissolution for formulations with different ratios of Acid Triglyceride/nifedipine

The ratio of Acid Triglyceride/nifedipine ranged from 3/7 to 15/7 for 100 grams sugar beads. If this ratio of coating material to nifedipine was 4/7, 5.5/7, 7/7 and 15/7, the release rate approximates zero-order kinetics ($R^2 \geq 0.8614$). However, the larger the amount of Acid Triglyceride present on the sugar beads the more prolonged nifedipine's release rate. Actually, with ratios of 4/7, 5.5/7, 7/7, and 15/7, nifedipine was not completely released from the beads. The percentage of drug released from the beads with the ratio of coating material to drug of 4/7, 5.5/7, 7/7, 15/7 was not over 80%. These formulations also do not satisfy the requirement for the total drug release of 80% as indicated in the "acceptance table", USP25-NF20 (3). To enhance drug release, it is necessary to decrease this ratio. When the ratio of coating material to drug was 3/7 and 4/7, the release rates of nifedipine deviate dramatically from a straight line. Formulation 5 yielded a total drug release of over 80%, but percentage of drug released was much higher than 17% at the four-hour time period and the R^2 value was only 0.7527. One thing to be emphasized here is the amount of drug deposited on the sugar beads. It can be seen that the amount of drug deposited on Formulations 4 and 5 was just above 50%. This phenomenon can be explained by the low ratio of wax/nifedipine (4/7 and 3/7). At these low ratios, the glutinous characteristic of the mixture was reduced remarkably. Consequently, the molten coating mixture could not cling to the surface of sugar beads and was easily wiped away during blending. It was also observed that the coating mixture 4/7, 3/7 wax/nifedipine precipitated in the bottom of coating pan in large masses, contributing to the low percent drug loading onto beads.

Preparing the dual coated dosage forms

As mentioned above, the criteria objective for the sustained release dosage forms in this project is that the drug product follows zero-order release kinetics and the amount of drug released from the wax/drug matrix should be complete. The "Acceptance Table" in USP25-NF20 is a reference, too. For single coated layer beads, to obtain a reasonable total amount of drug release, the release rate during the first hours is too fast and when the drug inside the matrix is released during the last hours it is too slow from the beads. In other words, the release rate from single layer beads appears to slow down over time and the fitted curve of the dissolution profile deviates from zero-order kinetics. In contrast to this, if the fitted curve of the drug dissolution profile follows zero-order release kinetic profile which may occur when using more than one coating agent with the drug, at 24 hours too much drug is still retained inside the beads. One remedy to the problem could be dual coating of the sugar beads.

To obtain zero-order release in dual coated beads, the drug deposited on the outer layer dissolves first. Then the drug in the inner layer dissolves later. To reduce the rate of drug release during the first hours, the ratio of drug and coating agent in the outside layer should have less drug and more coating agent than in the inner coat. When the outer layer erodes then channels leading to the second layer are revealed.



The outer layer must not be very durable since after time, the inner layer must be revealed, and drug from the inner layer must be released. To enhance the rate of drug release during the last hours, the drug loaded in the inner layer must release faster than the drug loaded in outer layer. For this reason, the amount of drug in the outer layer, and should contain less than the amount of drug in the inner layer. If a lag time is desired before drug is released, the outer layer should not contain any drug at all. The proper selection of waxes for the outer layer should be waxes with medium hydrophobicity and medium rigidity. Since the wax used for the inner coating layer needs to have a higher melting temperature than the wax used in the outer layer, and the drug in the inner layer needs to be released totally after 24 hours, a relatively high ratio of drug to coating agent is needed for the inner coating mixture to assure that the drug is nearly completely released after 24 hours.

Specifically, stearic acid is a coating agent with medium rigidity. Single layer coating experiment has shown that if the ratio of stearic acid/nifedipine equals 2, up to 80% nifedipine is released and if this ratio equals 1, 100% of drug is released. Therefore, stearic acid was selected as a coating agent for outermost layer. Carnauba wax has a melting point 20^0C higher than stearic acid. It was selected as the coating agent for the inner-coating layer.

Five different formulations of dual coated beads were prepared in which stearic acid was the outer layer and the carnauba wax was the inner layer. Procedures for preparation as indicated in the "Materials and Methods" section were followed. The ratio of wax/drug ranged from 2 (15/7 and 10/5) to 5 (10/2) and stearic acid only (22/0 and 15/0). The ratio of carnauba wax and drug also changed from 7/10 and 5/10. Nifedipine was thoroughly mixed with carnauba wax. Blending this mixture onto the nonpareil sugar beads to make the first coating layer was performed. The second coating process was carried out with molten stearic acid with or without nifedipine. Details of the formulations are listed in Table 19.

Table19
Dual coating formulations with stearic acid on the outer-coating layer

Formu-lation	**Sugar bead (100g)**	**Carnauba /nifedipine (g/g) (inner layer)**	**Stearic acid /nifedipine (g/g) (outer layer)**	**weight gain (%)**	**Theoretical percentage of drug in the beads**	**Actual percentage of drug in the bead**
1	x	0	15/7 g	22%	5.7377	86.4%
2	x	7/10	10/5	32%	12.8	65.8%
3	x	5/10	10/2	27%	9.4	58.0%
4	x	2.5/5	15/5	35%	11.1	66.8%
5	x	5/10	22/0	37%	7.29	54.02%
6	x	5/10	15/0	30%	8	59.11%

Figure 22: Sugar beads mesh size screen 30-35

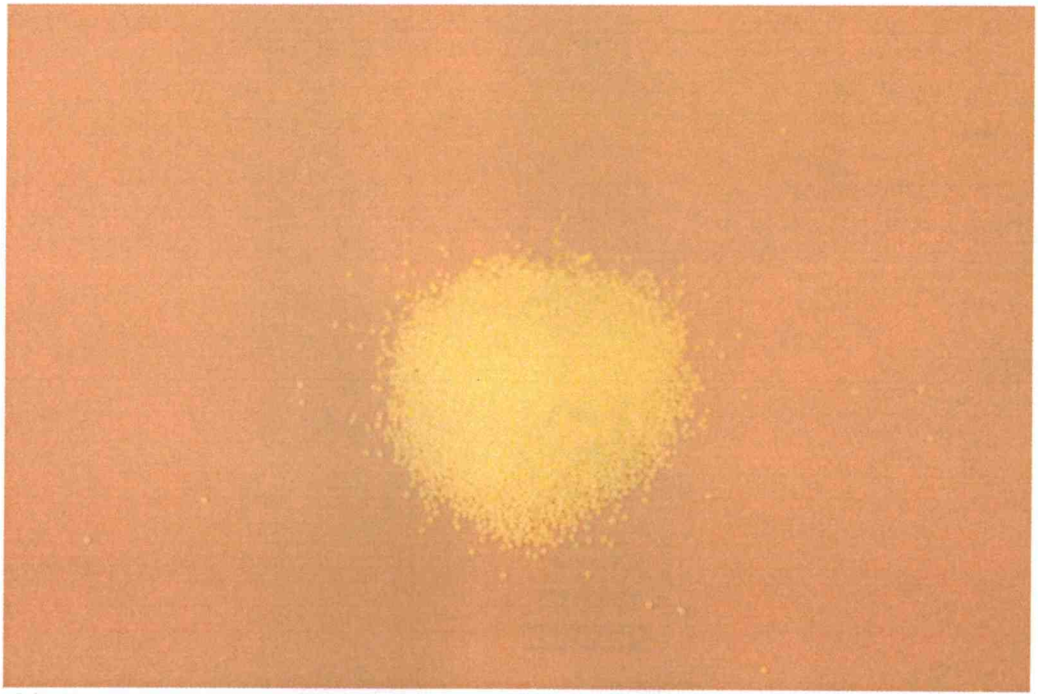


Figure 23: Dual coated beads, Formulation 6 (Inner layer: Carnauba wax 5gr and nifedipine10gr, Outer layer: Stearic acid 15gr and sugar beads 100gr)

Figure 24: Capsules containing dual coated beads of Formulation 6

Dissolution tests with the dissolution paddle set 100 rpm were conducted. The percentage of nifedipine released from the beads is presented in Table 20.

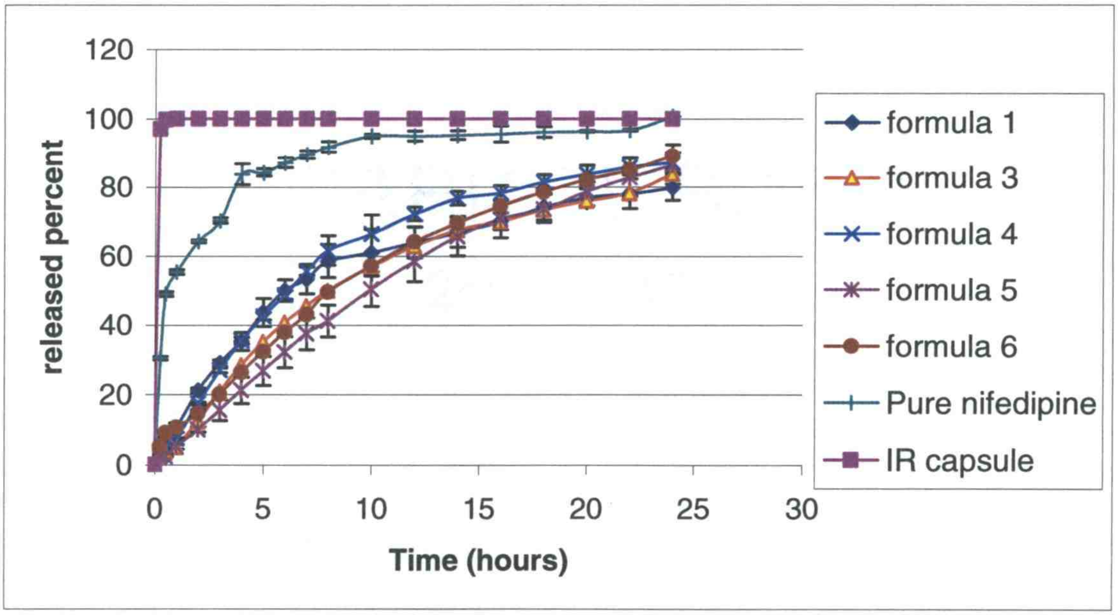


Figure 25: Dissolution profile of nifedipine from dual coating beads compared with that of pure nifedipine

Table 20
Percentage of nifedipine released from dual coated beads compared with single layer coated beads

Time (hours)	Formulation 1	Formulation 2	Formulation 3	Formulation 4	Formulation 5	Formulation 6	Pure nifedipine
0	0	0	0	0	0	0	0
0.25	5.312517	2.082489	2.266667	2.659168	1.959697	5.193252	30.41915
0.5	7.688188	3.9987	3.554335	5.078201	2.358635	9.103679	49.02709
1	10.59207	6.76483	5.036701	7.475163	5.14594	10.58643	55.34133
2	21.12076	10.3256	12.36059	17.44274	10.09012	14.58676	64.33825
3	28.97586	14.65443	20.69973	26.92955	15.52751	20.03013	70.29048
4	35.31427	20.32654	28.33877	35.53408	21.18945	26.35561	83.76204
5	43.67114	25.81068	35.00016	42.60898	26.7635	32.26366	84.26229
6	50.02058	29.6584	40.6224	48.75902	32.19905	37.87827	87.13963
7	53.33195	37.79925	45.45762	55.61317	37.49706	43.02176	89.60019
8	58.55656	41.3265	49.90841	61.67062	41.18393	49.55627	91.84597
10	61.03917	47.59377	56.94777	66.47655	50.36995	57.12204	95.00843
12	63.99588	54.9158	62.92865	72.21405	58.52836	64.21914	95.0021
14	66.90241	60.15038	67.65225	76.7584	65.81219	69.61763	95.3214
16	70.11542	64.6598	70.0039	78.36429	70.81323	74.57444	95.6543
18	74.29991	69.9874	73.51982	81.52637	73.79208	78.64319	96.2365
20	77.006	76.6598	76.11212	83.86402	78.71075	82.31921	96.4532
22	78.09912	80.13943	78.31753	85.98042	82.8949	85.17163	96.77449
24	79.9219	82.61757	83.94085	87.20256	86.64685	89.17801	100.7285

Dissolution profile of these formulations is shown in Figure 25. Total percentages of nifedipine dissolved within 24 hours from the various formulations are presented in Figure 26.

As expected, for low and medium rigidity waxes, a higher ratio of wax/drug in the outer coating layer will have lower amount of drug dissolved during the first hours than outer coatings with a lower wax/drug ratio. Formulation 3 and Formulation 6 had the same inner-coating layer (carnauba/drug 5/10). The stearic acid/nifedipine in the outer coating layer of Formulation 3 was 3/10 and for Formulation 6 it is 0/15. The percentage of drug released during the first 8 hours from Formulation 3 was higher than that of Formulation 6. Formulation 2 had the ratio of carnauba/drug of 7/10, and the total drug dissolution after 24 hours was 82.61%, lower than that of Formulation 6 having a carnauba/drug ratio of 5/10. In conclusion, the single layer coating of stearic acid alone had the greatest effect retarding nifedipine release during the first eight hours of the dissolution period. All linear regression coefficients (R^2) are listed in Table 21.

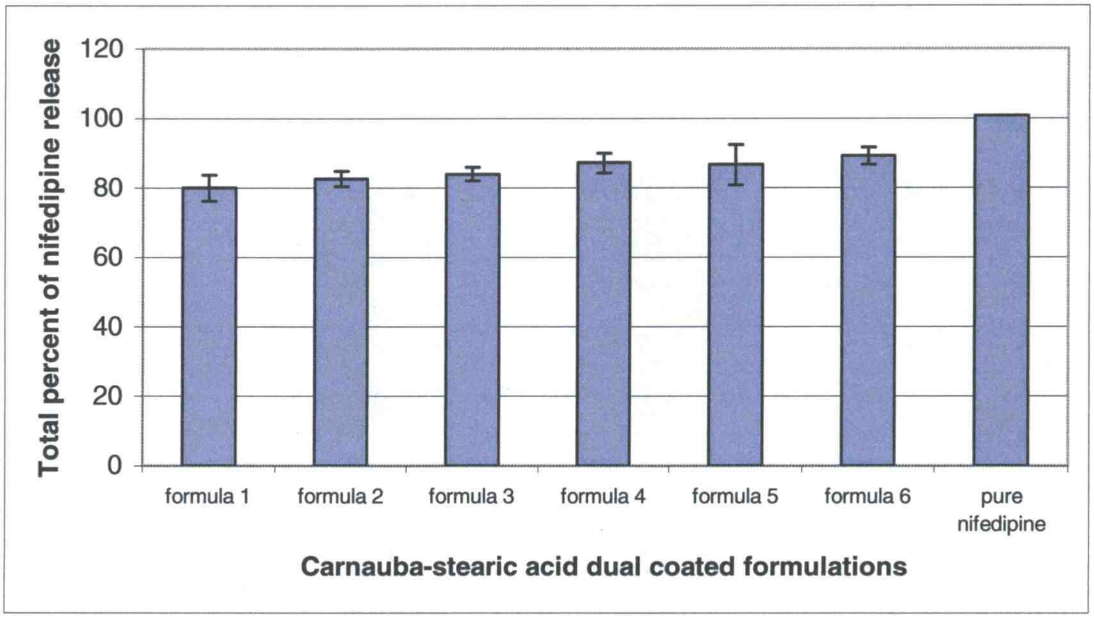


Figure 26: Total percentage of nifedipine release from carnauba-stearic acid coated beads within 24 hours

Table 21
R^2 value of nifedipine dissolution profiles for linearity of six dual coated beads compared with pure nifedipine

	Formulation 1	Formulation 2	Formulation 3	Formulation 4	Formulation 5	Formulation 6	pure nifedipine
R^2	0.8499	0.9734	0.9148	0.8678	0.9696	0.9632	0.535

Considering criteria for the percentage of drug released and linearity of the drug release profile, it is suggested that a dual coating with the outermost layer of single wax is optimal. It should be noted that the lag time for drug release did not occur even if the outermost layer did not contain any nifedipine. This can be explained by the heterogeneity of the coating crust. The outermost layer has numerous channels, and these channels play a dominant role to let the fluids reach the inner drug layer. Even though the drug was totally deposited on the inner layer, some part of this layer gets exposed to the water medium. As a result, a small amount of drug was still released during the first hours. The optimal coating material/drug ratio for the inner layer (carnauba/nifedipine) is 5/10. This ratio assures that the drug is not trapped tightly in the coating matrix. Although the fact that drug release after 4 hours is over 17% (26.36%), the "Acceptance value" in Acceptance Table " (USP25-NF20) for nifedipine extended- release tablets, Formula 6 had the best R^2 value ($R^2 = 0.9632$) and the total amount of drug released during 24 hours was highest (89.178%). One remaining concern is that the drug deposited on the sugar beads of Formulation 6 during preparation is rather low, approximately 60% because the low ratio of carnauba wax/nifedipine reduces the adherence of the coating mixture. Increasing the ratio carnauba/nifedipine (5/10) cannot be done because this will lead to a decrease of total amount of drug released. Some improvements may be made in the formulation, these include replacing carnauba wax with another less hydrophobic wax or use smaller nonpareil beads sizes in hopes that drug deposition on the beads will increase. It is also suggested that another approach to prepare zero-order kinetics sustained release dosage form is coating solid pellets of nifedipine. Combining nifedipine with other excipients to form pellets to be coated could give prolonged release, but the solid inner pallet matrix should not be so durable that nifedipine is unable to release relatively quickly when the coating layer erodes during the 24 hours of dissolution testing.

Further, to test the change in the dissolution profile when the coating agent in the outer layer is changed, stearic acid could be replaced by Acid Triglyceride as the outer coating agent, and carnauba wax remain as the inner coating agent. Nifedipine also can be loaded only on the inner coating layer or on both inner and outer coating layers. Since

Acid Triglyceride is more lipophilic than stearic acid, the nifedipine dissolution would be much more prolonged. To make sure the drug is released within 24 hours, the amount of drug loading and the ratio of Acid Triglyceride/nifedipine must be smaller than when using stearic acid.

Table 22
Dual coated nifedipine formulations with Acid Triglyceride as the outer layer

Formul-ation	Sugar bead (100g)	Carnauba /nifedipine (g/g) (inner layer)	Triglyceride /nifedipine (g/g) (outer layer)	weight gain (%)	Theoretical percentage of drug in the beads (%)	Actual percentage of drug in the bead
1	x	2.5/5	2.5/2.5	12.5%	6.67%	65.06%
2	x	2.5/5	5/4	16.5%	8.5%	63.16%
3	x	2.5/5	0.5/0	4.5%	2.5%	66.04%
4	x	5/10	2.5/0	17.5%	4.4%	60.54%

With these conditions, four formulations were prepared similarly as the carnauba wax-stearic acid dual coated beads were. All the formulations prepared are listed in Table 22. The ratio of carnauba wax and nifedipine was similar as in dual coated beads with stearic acid as shown above (5/10 and 2.5/5). The ratio of Acid Triglyceride and nifedipine ranged from 2.5/2.5 to 5/4 and Acid Triglyceride alone (0.5g, 2.5g). The quantity of sugar beads used was fixed at 100 grams for all formulations.

Percentage nifedipine released into the Tween 80 1% dissolution media is presented Table 23. Dissolution profiles of these formulations are presented in Figure 27. Total percentages of nifedipine released after 24 hours from these formulations are shown in Figure 28.

Table 23
Dissolution profile of four different types of Carnauba- Acid Triglyceride dual coated beads

Time (hours)	Formula 1	Formula 2	Formula 3	Formula 4
0	0	0	0	0
0.25	2.001917	1.691425	4.24039	1.43146
0.5	4.276405	2.03083	3.126469	1.22082
1	8.360115	2.67356	6.196427	1.371664
1.5	11.81883	3.319984	8.693154	1.639164
2	16.43675	5.720236	12.18369	3.103425
3	21.87237	8.111238	16.2251	4.343003
4	27.04555	10.54741	20.02067	5.930645
5	30.19992	12.95556	22.39777	7.656657
6	34.57943	15.35384	25.65501	9.490128
7	35.21292	16.77463	26.1265	10.94024
8	38.4502	18.88639	28.53196	13.35285
10	43.64615	23.60068	32.38882	17.34714
12	48.32337	28.31932	35.8534	21.39696
14	51.61159	32.80855	38.29553	25.26969
16	54.77811	37.2756	40.58237	28.31327
18	56.71833	41.21796	42.07476	31.96604
20	59.06147	44.96421	43.8123	35.19418
22	61.9965	48.63597	45.96355	38.2649
24	64.57264	51.53266	47.87203	41.26834

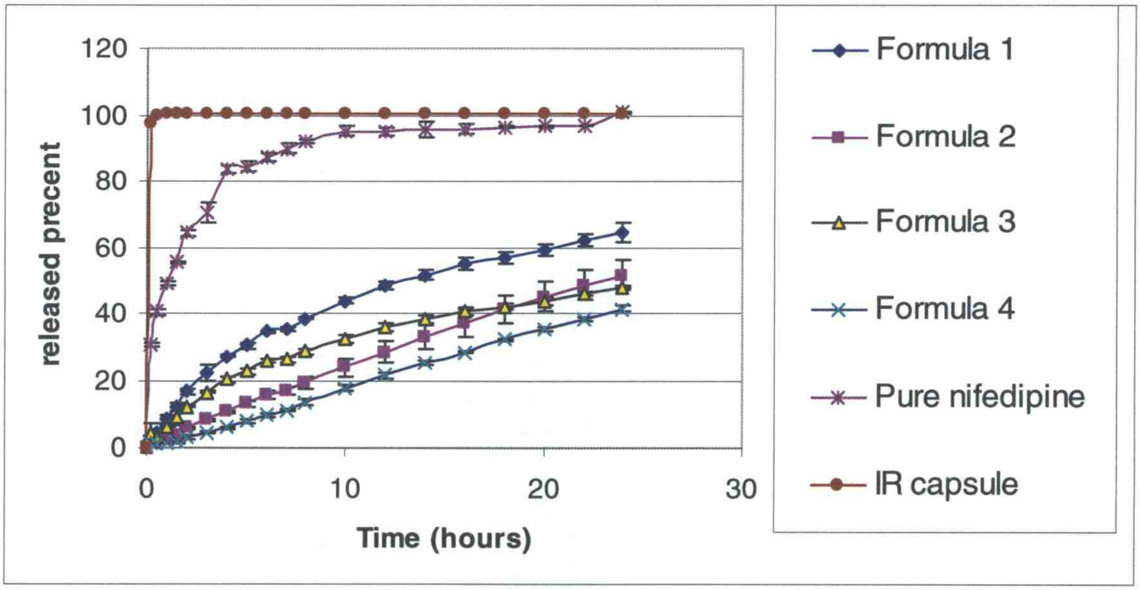


Figure 27: Nifedipine dissolution profiles from dual coated beads when Acid Triglyceride is the outer layer and carnauba wax is the inner layer

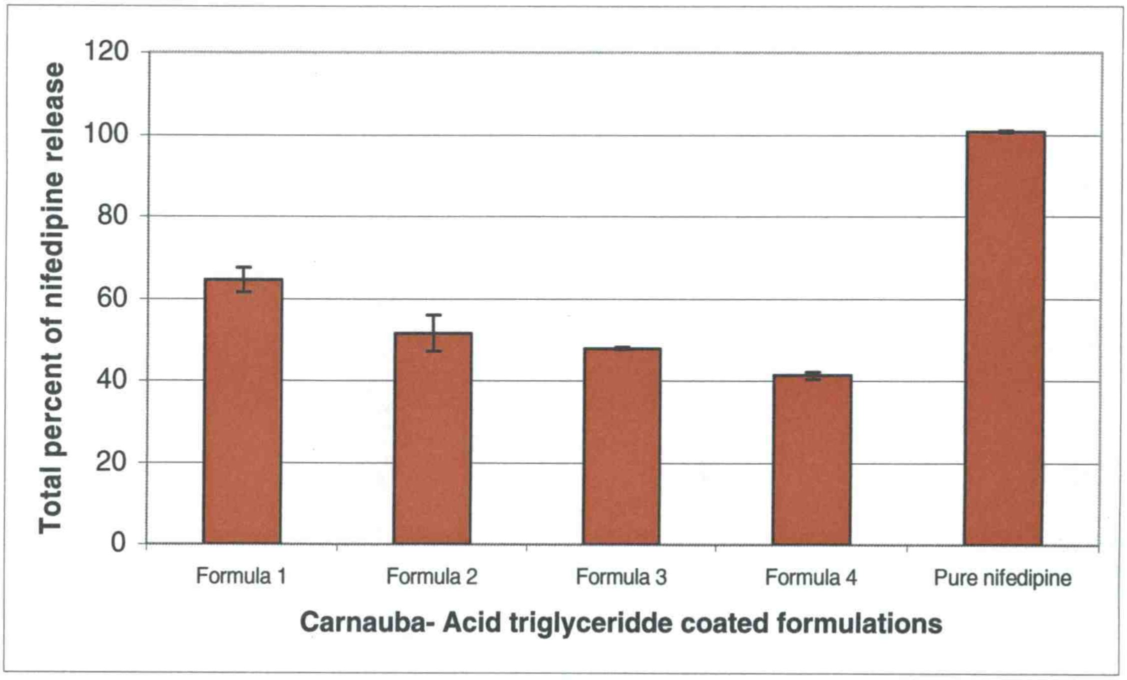


Figure 28: Total percentage of nifedipine release from Carnauba wax- Acid Triglyceride dual coated formulations

R^2 value of the Regression equation for each formulation is given in Table 24.

Table 24
R^2 value of dissolution profile of four carnauba wax-acid triglyceride dual coated formulations

	Formulation 1	Formulation 2	Formulation 3	Formulation 4	Pure nifedipine
R^2	0.913	0.9974	0.9146	0.9977	0.535

Compared with dual coated beads using stearic acid as a coating agent, dual carnauba wax-Acid Triglyceride coated beads had a lower percentage of drug released during 24 hours, but the fit to linearity was good. The percentage of drug dissolved from Formulation 1 in the first four hours was 27% and only 64.57% in 24 hours. The linearity of drug release was relatively good (R^2 = 0.913). The amount of drug deposited on the sugar beads was around 60 % of theoretical for all formulations. If the wax/drug ratio was reduced to promote an increase in the total amount of drug released, the linear fitting of the drug release profile deviates significantly from straight line, as seen with decreasing R^2, and the percentage of dissolved drug during the initial four hours increased. Dual coated beads using Acid Triglyceride and carnauba wax are not superior for drug release in comparison with the dual coated beads using carnauba wax with stearic acid.

Investigating the dissolution profile of capsules containing beads

After preparation, the coated beads need to be loaded into suitable capsules. Hard gelatin capsules are reported to disintegrate in about 15 minutes. It is hoped that the dissolution profile of the beads alone and beads loaded into hard gelatin capsules is not different. To test this, a dissolution test comparing beads alone to beads inside a capsule was carried out. Dual coated beads, Formulations 5 and 6 of Table 18 exhibited the best drug release profile, and were selected to compare differences in dissolution of nifedipine from capsules containing beads to beads not placed inside gelatin capsules. Coated beads equal to 30mg nifedipine were put into size 0CS capsule. The dissolution test was performed with dissolution paddle set at 100 rotations per minute. Time for drawing samples was as before to 24 hours. The dissolution profile was computed from the mean of the concentrations from six dissolution flasks. Percentage of drug released from capsules containing beads and beads alone are presented in Table 25 for comparison.

The nifedipine dissolution profile from the beads alone and from the beads inside capsules is presented in Figure 29 for comparison.

The dissolution data shows that the gelatin capsule shells slightly delays nifedipine release during 24 hours. The total percentage of drug dissolved after 24 hours was also a little lower than that of bare beads. Total percentage of drug released from

Formulation 6 inside gelatin capsules was 83.66%, and the value from bare beads without capsules was 86.65%. In Formulation 5, the difference in total percentage of drug released was less between beads in capsules and bare beads, only 1.5% difference. The paired t test indicated that there is no significant difference in nifedipine drug dissolution profiles from the beads in capsules and that of the beads alone. The small difference in drug release can be explained by the gelatin crust characteristics. The time for water to enter the hard gelatin capsules and to dissolve the gelatin in water is about 15 minutes. Coated beads placed inside the capsules have a small delay before encountering water. Nevertheless, this delay is not a significant effect on the overall release profile of drug from the beads.

Table 25
Percentage of nifedipine released from capsules containing stearic acid coated beads and stearic acid coated beads not inside capsules

Time (hours)	formulation 5	Formulation 5-capsule	formulation 6	Formulation-capsule
0	0	0	0	0
0.25	1.959697	1.216878	5.193252	1.902924
0.5	2.358635	1.396051	9.103679	2.151798
1	3.15332	2.01321	10.58643	5.327392
2	10.09012	5.958703	14.58676	11.23624
3	15.52751	9.140892	20.03013	16.35257
4	21.18945	16.01681	26.35561	21.50072
5	26.7635	21.41599	32.26366	27.24812
6	32.19905	26.95993	37.87827	32.75239
7	37.49706	32.1631	43.02176	38.3526
8	41.18393	36.3431	49.55627	41.52852
10	50.36995	46.97629	57.12204	49.58292
12	58.52836	55.09509	64.21914	57.70946
14	65.81219	62.05958	69.61763	63.8991
16	70.81323	68.6548	74.57444	69.81843
18	73.79208	72.1235	78.64319	75.02884
20	78.71075	76.32654	82.31921	79.59153
22	82.8949	80.26548	85.17163	83.16745
24	86.64685	83.6548	89.17801	87.69786

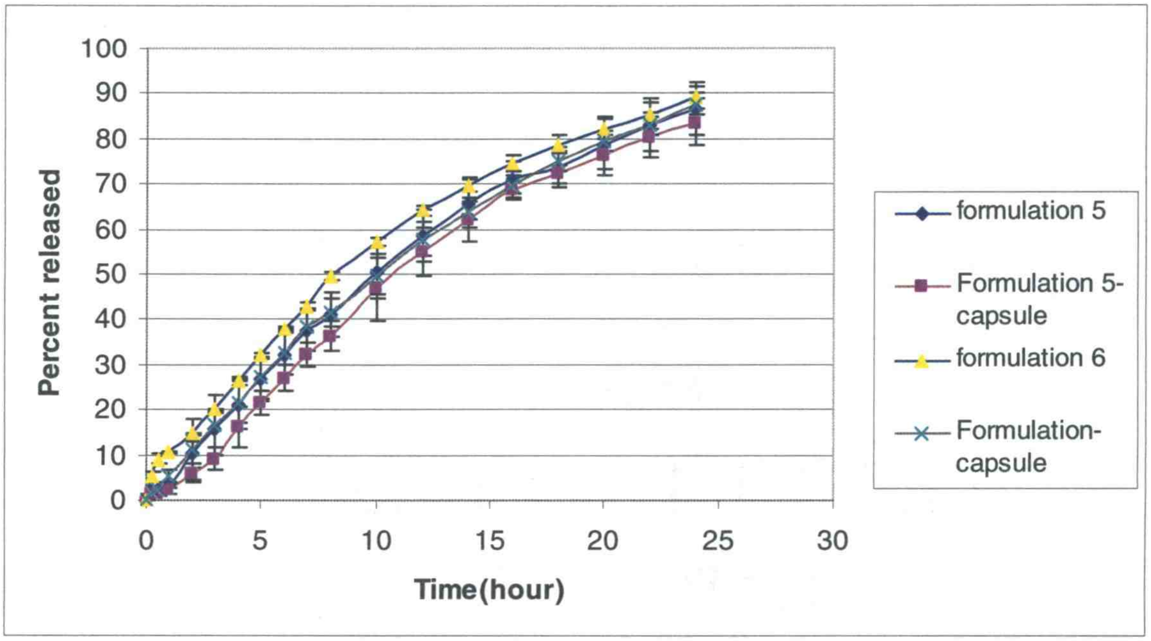


Figure 29: Nifedipine dissolution profile from stearic acid coated beads and the same beads placed in capsules

Drug content uniformity

In Formulation 6, Table 19, dual coated beads of carnauba wax and stearic acid yielded the best dissolution profile among the prepared beads. As indicated in the USP25-NF20, Uniformity of Dosage units (3) states the following " The requirement for dosage form uniformity are met if the amount of the active ingredient in each of the 10 dosage units as determined from the relative standard deviation is less than 6%". Nifedipine content uniformity is defined as following: First, determination of the actual content of nifedipine in beads should be done exactly like the method in dissolution study (Chapter 3, "Materials and Methods" section). Second, 10 capsules filled with beads, Formulation 6, containing 30 mg of nifedipine was prepared on the basis of the actual percentage of nifedipine that was determined in the previous steps. Nifedipine was then dissolved in 100ml of a mixture of methanol and acetonitrile (1:1). This solution was further diluted from 1 ml to 100 ml. The absorbance was read at 334 nm on the spectrophotometer as soon as possible. The blank sample is a mixture of methanol and acetonitrile (1:1). The content is interpolated from the standard curve. The results obtained are presented in Table 26.

The relative standard deviation is 5.718 % meeting the stipulation of USP25-NF20. Therefore, the variability within a batch lies in the acceptance range (6%).

Table 26
Content of nifedipine inside carnauba wax-stearic acid coated beads loaded in each capsule

Number of capsules	Absorbance	Amount in one capsule
1	0.3812	26.972028
2	0.4235	29.93007
3	0.4244	29.993007
4	0.4565	32.237762
5	0.3998	28.272727
6	0.4135	29.230769
7	0.4065	28.741259
8	0.4368	30.86014
9	0.4499	31.776224
10	0.4503	31.804196
	Average	29.98182
	STDEV	1.7155297
	CV %	5.7184322

Convolution

Convolution is a simulation process. Herein it is used to simulate the plasma nifedipine concentrations that would be observed if the test formulation were given to a human volunteer. Convolution performed with Kinetica is used to compare the expected plasma concentrations of the prepared formulations and the "standard dosage forms", usually the commercially available dosage forms.

Two well-known dosage forms of sustained release nifedipine in circulation are Adalat®CC, and Procardia® XL. The optimal formulation in this study is number 6 (based on the percentage of drug released and the R^2) in Table 19 (The inner layer is carnauba and nifedipine with the ratio 5gram/10gram, 100 g sugar bead and the out layer is stearic acid, 15 gram).

Convolution was performed with the following assumptions (52, 53):

First, GI absorption of nifedipine is equal or faster than *in vitro* dissolution of the drug. This would cause *in vivo* absorption to occur at the same rate as *in vitro* dissolution of nifedipine. As mentioned above, nifedipine is absorbed quickly after oral administration. The nifedipine dissolution profile is used for the *in vivo* absorption profile and this assumption is used for well-absorbed drugs like nifedipine. Second, as indicated in the "Materials and methods" section, the drug must follow linear pharmacokinetics. It has been indicated earlier that the total dose 30 mg must be absorbed during the 24-hour period after administration, and pharmacokinetic parameters are linear and obey superposition. Additionally, nifedipine undergoes a first pass effect which reduces the plasma concentrations. It has been shown that, CYP 3A4 metabolizes nifedipine. As a result of the first pass effect, bioavailability of nifedipine is decreased to 50-70% for immediate release dosage forms, and 45-55% for sustained release dosage forms. No exact adjustment can be made for the first pass effect. An approximate adjustment in convolution is made where the nifedipine drug dissolution concentration were multiplied by 0.5 to account for drug lost during first pass metabolism.

Another problem to deal with in convolution is the "linear box". Linear box should be the same for both the weighting function and input function. However, this ideal condition can not be satisfied because the absorption profile is replaced by the nifedipine dissolution profile. In addition, intravenous data can vary among subjects. Thus, if a population average IV data set is not available, published data used for the output function on a group of healthy subjects will be selected and considered to be representative of the whole population. Although, this lessens the reliability of convolution results, some pharmacokinetic studies show that intraindividual variability on healthy subjects is not significant especially during short term administration. On the whole, the convolution is just a rough foretelling of plasma concentrations. Many confounding factors may make the actual plasma concentrations *in vivo* differ from the predicted values.

Intravenous data:

Intravenous data were obtained from 12 healthy subjects (54).

The plasma concentration of bolus intravenous injection is given in Table 27.

Table 27
Intravenous data for nifedipine from twelve healthy subjects

Time (hours)	Plasma concentration (mcg/l)
0	18.44
0.1	18.89
0.2	13.58
0.3	10.84
0.5	8.69
0.75	6.25
1	4.68
2	3.76
4	2.40
6	2.75
8	1.95

Dissolution data:

Dissolution profiles with the dissolution paddle set at 100 rpm for Adalat® CC, Procardia® XL, and Formulations 5, 6 (Table 18) in capsules is presented in Table 28 and Figure 30.

It is noted that the dissolution profile of Procardia® XL followed zero-order release kinetics well.

Table 28
Nifedipine dissolution profiles from Adalat® CC, Procardia® XL, Formulation 5, and Formulation 6 in capsules

Time (hour)	Adalat CC(%)	Procardia XL (%)	Formulation 5-capsules (%)	Formulation 6-capsules (%)
0	0	0	0	0
0.25	0	0	1.216878	1.902924
0.5	2.537338	0.299219	1.396051	2.151798
0.75	4.421212	0.299219	2.01321	3.173818
1	9.523771	0.299219	3.6869	5.327392
2	19.12344	0.298919	5.958703	11.23624
3	25.59389	5.847255	9.140892	16.35257
4	44.08357	11.75056	16.01681	21.50072
5	61.47698	18.921	21.41599	27.24812
6	79.17245	24.3494	26.95993	32.75239
7	88.54713	29.75962	32.1631	38.3526
8	92.87991	31.38114	36.3431	41.52852
10	99.44311	42.28583	46.97629	49.58292
12	101.1172	53.13048	55.09509	57.70946
14	101.1416	62.24538	62.05958	63.8991
16	101.4825	70.36515	68.6548	69.81843
18	102.1176	78.34675	72.1235	75.02884
20	102.4599	84.92345	76.32654	79.59153
22	102.9572	90.04244	80.26548	83.16745
24	102.3569	94.26357	83.6548	87.69786

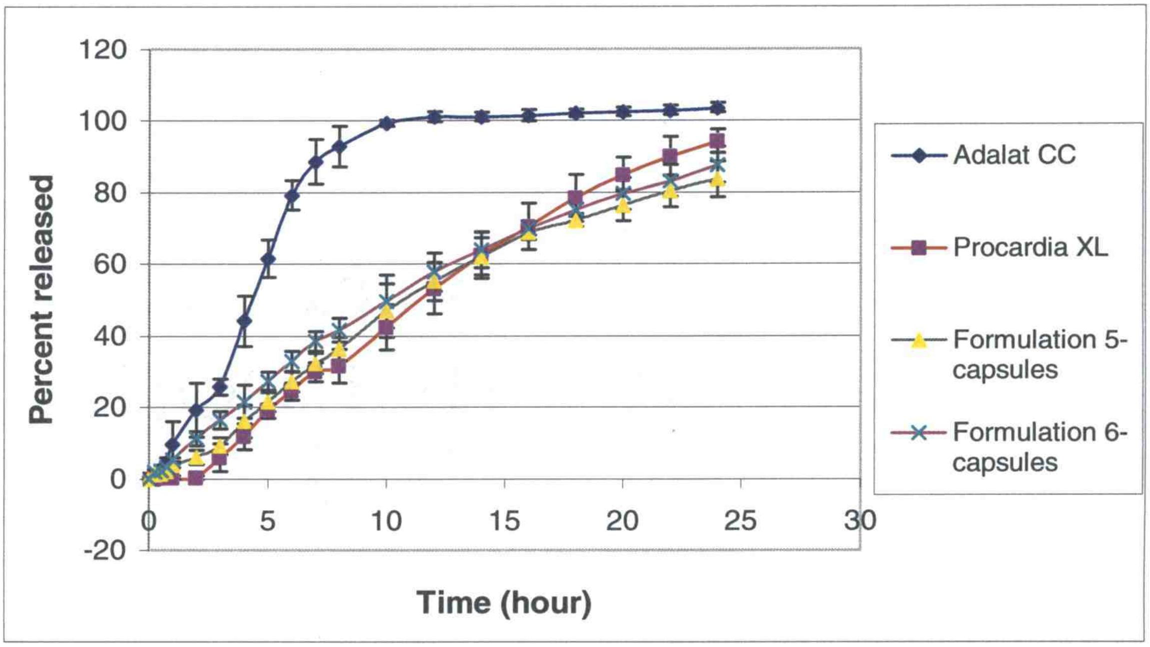


Figure 30: Nifedipine dissolution profiles for Adalat® CC, Procardia® XL, Formulation 5, and Formulation 6 (Table 19)

The R^2 value is equal to 0.975 and the dissolution profile satisfied the "acceptance table" in USP25-NF20. Within 24 hours 94% of nifedipine dissolved from the osmotic system. This result absolutely coincided with the published papers about Procardia® XL. However Adalat® CC capsules did not yield zero-order release kinetics. Drug dissolved quickly during the first ten hours and the dissolution profile did not satisfy the "Acceptance Table".

The adjusted amount of drug released from the dosage form wherein an accounting for first pass metabolism is given in Table 28. The amount is calculated according to the formula:

$$A = C\%*30mg/100*0.5$$

A: amount of drug released

C%: Relative percent of total drug concentration in the dissolution medium

Table 29
Amount of drug release (mg) after accounting for first pass metabolism

Time (hours)	Adalat® CC	Procardia XL	Formulation 5-capsules	Formulation 6-capsules	Pure nifedipine	IR capsule
0	0	0	0	0	0	0
0.25	0	0	0.182532	0.285439	4.562873	14.5936
0.5	0.380601	0.044883	0.209408	0.32277	6.048975	14.99261
0.75	0.663182	0.044883	0.301981	0.476073	7.354064	15.02956
1	1.428566	0.044883	0.553035	0.799109	8.301199	15.02956
2	2.868515	0.044838	0.893805	1.685437	9.650737	15.02956
3	3.839083	0.877088	1.371134	2.452885	10.54357	15.02956
4	6.612535	1.762584	2.402521	3.225108	12.56431	15.02956
5	9.221546	2.83815	3.212398	4.087218	12.63934	15.02956
6	11.87587	3.652411	4.04399	4.912858	13.07094	15.02956
7	13.28207	4.463943	4.824465	5.752891	13.44003	15.02956
8	13.93199	4.707172	5.451465	6.229278	13.77689	15.02956
10	14.91647	6.342874	7.046443	7.437438	14.25126	15.02956
12	15.16757	7.969572	8.264263	8.656419	14.25032	15.02956
14	15.17125	9.336807	9.308936	9.584865	14.29821	15.02956
16	15.22237	10.55477	10.29822	10.47276	14.34815	15.02956
18	15.31763	11.75201	10.81853	11.25433	14.43548	15.02956
20	15.36899	12.73852	11.44898	11.93873	14.46798	15.02956
22	15.44357	13.50637	12.03982	12.47512	14.51617	15.02956
24	15.54821	14.13954	12.54822	13.15468	15.10928	15.02955

The adjusted dissolution data (Table 29), and intravenous data (Table 27) were put into Kinetica with the convolution menu. Kinetica interpolated the absent time points in the IV data to infer the output allowing prediction of plasma nifedipine concentrations. Nifedipine plasma concentration values versus time curve of the four formulations are presented in Table 30 and the plasma nifedipine concentrations vs. time curve are also presented in Figure 31.

Table 30
Predicted plasma concentration (μg/l) by convolution

Time (hours)	Adalat CC	Procardia XL	Formulation 5-capsules	Formulation 6-capsules	pure nifedipine	IR capsule
0	0	0	0	0	0	0
0.25	0	0	0.399125	0.624142	9.97721	31.9105
0.5	0.839839	0.099039	0.334327	0.512448	10.1542	22.8688
0.75	1.197	0.067625	0.433066	0.688982	9.82595	15.7373
1	2.5073	0.046301	0.856315	1.19362	8.96734	11.2711
2	2.78595	0.023356	0.771498	1.64976	6.13682	7.31174
3	2.73336	1.1554	1.07289	1.86837	5.31647	5.81891
4	5.4233	1.67619	2.00046	2.11565	6.50578	4.65244
5	6.36383	2.31164	2.13268	2.435	4.18057	3.717
6	7.27279	2.35997	2.3985	2.61549	4.22866	3.82647
7	6.38613	2.53698	2.56495	2.85684	4.25794	4.09599
8	5.38708	1.93319	2.52971	2.58026	4.0125	3.52138
10	4.63346	2.87625	3.05289	2.82374	3.53669	2.891
12	3.78826	3.2915	3.02376	3.00267	2.66004	2.30668
14	3.02285	3.30219	2.95742	2.83994	2.19471	1.83337
16	2.51652	3.29369	2.94139	2.77388	1.80401	1.45708
18	2.09486	3.33445	2.48931	2.64699	1.5004	1.15802
20	1.68061	3.16951	2.40637	2.48048	1.19265	0.920341
22	1.38711	2.90196	2.27557	2.24522	0.980654	0.731445
24	1.17701	2.65014	2.07766	2.24834	1.33079	0.581313

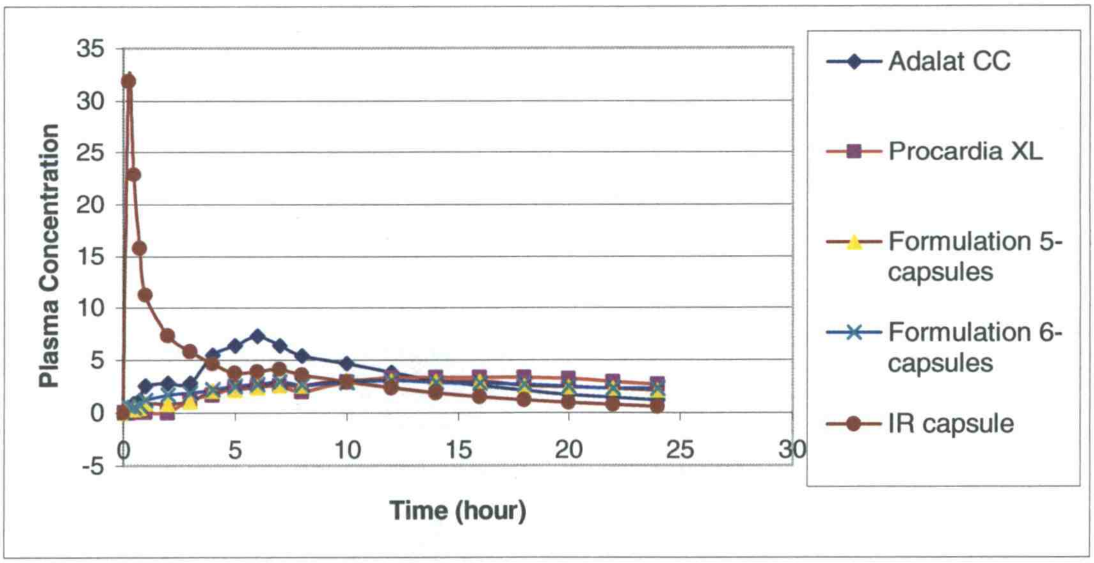


Figure 31: Predicted plasma concentrations of nifedipine by convolution

After obtaining the predicted plasma concentrations for the four formulations, the concentration versus time values were also put into non-compartmental analysis in Kinetica. The pharmacokinetic parameters obtained from non-compartmental analysis are listed in Table 31.

Table 31
Pharmacokinetic parameters obtained by non-compartment analysis

Dosage form	C max (mcg/L)	T max (hour)	AUC (0-24 hour) (hour*mcg/l)	$t_{1/2}$ (hour)	MRT (hour)	Clearance (L/hour)
Adalat CC	7.272	6.000	92.394	7.322	13.160	266.526
Procardia® XL	3.334	18.000	119.272	15.869	30.7017	205.143
Formulation 5	3.053	10.000	112.696	18.885	32.328	266.204
Formulation 6	3.002	12.000	167.085	18.245	33.258	225.235
IR capsule	31.911	0.25	187.141	6.035	7.812	244.268

The two commercial products, Adalat® CC and Procardia® XL, are dissimilar in their dissolution profiles. Nifedipine in Adalat® CC was released faster than Procardia® XL and Adalat® CC's release kinetics did not follow zero-order kinetics. Consequently, the predicted plasma concentration time curves are not the same. The predicted C_{max} of Adalat® CC occurs at 6 hours whereas Procardia® XL is 18 hours. Formulations 5 and 6 have values for C_{max}, T_{max}, Half-life ($t_{1/2}$), Mean Residence Time (MRT), and Clearance (Cl) which are closer to Procardia® XL. The predicted plasma concentrations of the two Formulations, 5 and 6, are more comparable to Procardia® XL, too. Generally, the fluctuations in drug concentrations of Formulations 5 and 6 were small, ranging only 2-4 µg/l different from Procardia® XL. By comparison, Adalat® CC and immediate release capsule (Adalate® capsule), the plasma concentrations were quite different from

Procardia® XL and the two Formulations 5 and 6. The deviation of drug concentrations in plasma was as high as 31 and 7 μg/l for Adalat® capsule (an immediate release dosage form of nifedipine) and Adalat® CC, respectively to the other formulations.

CHAPTER 5: CONCLUSIONS

The hot-melt coating method by direct blending was applied to develop a sustained release dosage form of nifedipine that follow zero-order release kinetics. It was shown to be feasible to prepare nifedipine coated beads and the technique used to prepare the coated beads could be adjusted to produce a dosage form that gave dissolution profiles of drug which conformed to zero-order release kinetics. Other new formulations also gave good sustained release patterns suitable for many drugs but emphasis was placed on zero-order release in one aspect of this research. A capsule containing coated beads equal to 30 mg of nifedipine proved to be a suitable sustained release dosage form of nifedipine. *In vitro* test results of dissolution profiles, and convolution to predict plasma concentrations versus time profiles showed that almost all results of the test formulation lie in the acceptable range. However there are some concerns that need further investigation and improvements. The amount of drug deposited on the sugar beads is relatively low using the laboratory process. Therefore, changing to small sugar bead sizes, and changing the inner coating material may be a remedy to improve loading of the dosage form. Polymorphology of the coating materials may cause variability of drug release, so a future full investigation of thermal behavior would be required, too. The possibility of variations in drug release between batches requires validation of the process to make sure the method is practical.

In a positive vein the hot-melt coating method can be expanded and adapted to a broader scale of applications as long as the drug is confirmed stable at the temperatures involved. To increase the amount of drug loading on the beads for higher dosing, smaller sugar beads could be used or replaced by a drug matrix. Drug substrates could be prepared first by various granulation methods for instance, i.e. the extrusion method. In addition, combining the hot–melt coating method with other popular aqueous organic coating processes and agents may be a good direction for making sustained release dosage forms.

REFERENCES

1/ Malcolm Rowland, Thomas N. Tozer, Randy Rowland, *Clinical pharmacokinetic: Concepts and applications*, Lippincott, Williams & Wilkins; 3rd edition, January 15, 1995.

2/ United State Pharmacopeia 25, National Formulary 20 (USP25-NF20).

3/ Pharmacy net work group, http://www.pharmacynetworkgroup.com/c/procardia-pharmacology.htm

4/ Chowdhury Hafizul Ahsan, *Doctor of philosophy*, Falculty of medicine, Clinical Pharmacology group, University of Southamton, United Kingdom,1991.

5/ *The Physicians' Desk Reference*, 2002.

6/ RX list, The internet drug index, available at: http://www.rxlist.com/cgi/generic2/nifedip.htm

7/ Foster, Hamann, Richards, et al., *Nifedipine in normal subjects*, The Journal of Clinical Pharmacology, April, 1983.

8/ Tricia Agger, Tracy Arrossa, Kevin Cleveland, Gloria Gest, Ed Richardson, *Nifedipine: Procardia®, Procardia XL®, Adalat®, Adalat CC.*, Pfizer lab documents 2001.

9/ *Procardia® (nifedipine) capsules for oral* use, Pfizer Labs, 2000.

10/ Chabanian AV, *Calcium channel blockers*. Lessons learned from MIDAS and other clinical trials. JAMA 276:829-830, 1996.

11/ Takahiro Iwao, Katsuhisa Inoue, Yayoi Hayashi, Hiroaki Yuasa and Jun Watanabe, *Metabolic extraction of nifedipine during absorption from rat small intestine*, Drug metabolism Pharmacokinetics, 17 (6): 546-553, 2002.

12/ Zylber-Katz, E.; Turetz-Abramovitch, M.; Koren, G.; Levy, M. Dep. Med., Hadassah Univ. Hosp., Jerusalem, Israel., *Pharmacokinetic interaction between nifedipine and propranolol*. Fundamental & Clinical Pharmacology, 2(1), 29-35, 1988.

13/ Raemsch, Klaus D.; Sommer, Johannes. Inst. Pharmacokinet., Bayer A.-G., Wuppertal, *Pharmacokinetics and metabolism of nifedipine*. Fed. Rep. Ger. Hypertension, 5 (4, Pt. 2), 18-24, 1983.

14/ Schug, B.S.; Brendel, E; Chantraine, E; Wolf, D; Martin, W; Achall, R; Blume, H.H., *The effect of food on the pharmacokinetics of nifedipine in two slow release formulations: pronounced lag-time after a high fat breakfast*, British Journal of Clinical Pharmacology, 53(6), 582-588, 2002.

15/ Xing Liu, Dawei Chen, and Ruhua Zhang, *Evaluation of monolithic Osmotic Tablet System for Nifedipine Delivery in vitro and in vivo*, Drug Development and Industrial Pharmacy, Vol. 29, No 7, pp. 813-819, 2003.

16/ Guang Yan, Henan Li, Ruhua Zhang, and Derong Ding, *Preparation and Evaluation of a Sustained- released Formulation of Nifedipine HPMC Tablets*, Drug development and industrial Pharmacy, 26(6), 681-686, 2000.

17/ Grossman E, Messerli FH, Grodzicki T, Kowey P. *Should a moratorium be placed on sublingual nifedipine capsules given for hypertensive emergencies and pseudoemergencies?* JAMA 276:1328-1331, 1996.

18/ Palma-Aguirre, Jose Antonio; Rosas-Alcazar, Guillermo; Rodriguez, Juan Manuel; Leon-Urrea, Francisco; Montoya-Cabrera, Miguel Angel, *Bioavailability and pharmacokinetics of nifedipine administered by different routes in healthy volunteers*, Pharmaceutical Research, 16(2), 309-313, 1988.

20/ Rie Kubota, Takako Komiyama, and Hideyo Shimada, *Evaluation of the Method for Nifedipine Administration for a rapid onset of Clinical effect*, Yakigaku zasshi, 121(5) 355-364, 2001.

21/ Daniel J. Cobaugh, PharmD, *Electronic continuing education for specialists in poision information.* availabale at: http://www.urmc.rochester.edu/urmc/aapcc/

22/ Adebayo, G. I.; Mabadeje, A. F. B. Dep. Pharmacol., Coll. Med., Lagos, Nigeria., *Effect of nifedipine on antipyrine and theophylline disposition*, Biopharmaceutics & Drug Disposition, 11(2), 157-64, 1990.

23/ Eliot, Lise A, Foster, Robert T, Jamali Fakhreddin, *Effects of hyperlipidemia on the pharmacokinetics of nifedipine in the rat.* Pharmaceutical research, 16(2), 309-313, 1999.

24/ Soons P A;Schoemaker H C; Cohen A F; Breimer D D, Intraindividual variability in nifedipine pharmacokinetics and effects in healthy subjects, Journal of Clinical Pharmacology, 32(4),324-31, 1992.

25/ Shimada T, Guengerich FP., Evidence for cytochrome P-450NF, the nifedipine oxidase, being the principal enzyme involved in the bioactivation of aflatoxins in human liver, Proc Natl Acad Sci USA. 1989 Jan; 86 (2):462-5, 1989.

26/ *Adalat Retard*, Bayer Corp., Patient information. 939, 2003.

27/ A Miniscalco, J Lundahl, CG Regardh, B Edgar and UG Eriksson, *Inhibition of dihydropyridine metabolism in rat and human liver microsomes by flavonoids found in grapefruit juice*, Bristish journal of Clinical Pharmacology, 53(6), 2002.

28/ Bailey DG, Spencer JD, Munoz C, Arnold JM., 1992, *Interaction of citrus juice with felodipine and nifedipine,* Lancet Feb 2;337(8736):268-9,1991.

29/ C. C. Lang, S. K. Jamal, Z. Mohamed, M, R. Mustafa, A. M. Mustafa & T. C. Lee, 2003, *Evidence of an interaction between nifedipine and nafcillin in humans*, British Journal of Clinical Pharmacology, Volume 55 Issue 6 Page 588 - June 2003.

30/ Hoyo-Vadillo, C.; Castaneda-Hernandez, G.; Herrera, J. E.; Vidal-Garate, J.; Salazar, L. A.; Moreno-Ramos, A.; Chavez, F.; Tena, I.; Hong, E. Cent. Invest. Estud. Avanzados, IPN, Mexico City, Mex., *Pharmacokinetics of oral nifedipine: Relevance of the distribution phase*, Journal of Clinical Pharmacology, 29(3), 251-6., 1989.

31/ Tateishi, Tomonori, watanable, Minoru, Moriya, Hitonobu, Yamaguchi, Susumu, , *No ethnic difference between Caucasian and Japanese hepatic in the expression frequency of CYP3A5 and CYP3A7 proteins*, Biochemical Pharmacology, 57(8), 935-937,1999.

32/ Ball, Simon E; Scatina, Jo Ann; Kao, John; Geraldine M et al., *Population distribution and effects on drug metabolism of genetic variant the 5' promoter region of CYP3A4*, Clinical Pharmacology and therapeutics, 66(3), 288-194,1999.

33/ Al-Ajmi, H, S: Dawe, R.S; Renwick, A.G; Maclin, B.S.; Ferguson, J'Gibbs, N.K., *The effects of whole-body sun-bed ultraviolet a exposure on the pharmacokinetics of the photolabile drug nifedipine*, Photodermatology, photoimmuology and phptomedicine, 16(3), 2000.

34/ Leucuta S E; Grigorescu M; Dumitrascu D Facultatea de Farmacie, Institutul de Medicina si Farmacie, Cluj-Napoca, *Nifedipine pharmacokinetics in liver cirrhosis patients after the administration of a single oral dose*, REVISTA DE MEDICINA INTERNA, NEUROLOGIE, PSIHIATRIE, NEUROCHIRURGIE, DERMATO-VENEROLOGIE. MEDICINA INTERNA), 42(2), 185-90, 1990 Mar-Apr.

35/ Boje, Kathleen M.; Fung, Ho Leung., Sch. Pharm., State Univ. New York, Buffalo, Amherst, NY, USA., *Characterization of the pharmacokinetic interaction between nifedipine and ethanol in the rat,* Journal of Pharmacology and Experimental Therapeutics 249(2), 567-71, 1989.

36/ Robertson, D.R.C; Waller, D.G; Renwick, A.G.;George,C.F., *Age-related changes in the pharmacokinetics and pharnacodymamics of nifedipine*, British Journal of Clinical Pharmacology, 25(3), 197-305, 1998.

37/ Gong L, Zhang W, Zhu Y, Zhu J, Kong D, Page V, Ghadirian P, LeLorier J, Hamet P., *Shanghai trial of nifedipine in the elderly,* National Library of Medicine 14(10):1237-45,1996.

38/ Cipla corporation, 1998, *Antianginals & Antihypertensives*

39/ Gulielmo M. Trovato, Elio Iannetti, Giuseppe Carpinteri, Spanti D., *Effects of nifedipine on fluid compartments assessed by multifrequency bioelectric impedance in hemodialysis patients*, Delta nephrology and dialysis unit, Vittorio Emanuele II Hospital, Catania. 2003.

40/ *Hand book of pharmaceutical excipients*, American Pharmaceutical association, 2000

41/ Xing Liu, Dawei Chen, and Ruhua Zhang, *Evaluation of Monlithic Osmotic Tablet System for Nifedipine Delivery In Vitro and In Vivo,* Drug Developmant and Industial Pharmacy, Vol.29, No.7,pp.813-819, 2003.

42/ Longxiao Liu, Gilson Khang, John M. Rhee, Hai Bang Lee, *Monolithic osmotic tablet system for nifedipine delivery*, Journal of control release, 2000.

43/ Zheng Wang, Fumitoshi Hirayama, *In vivo and in-vitro evaluation of a modified-release oral dosage form of nifedipine by hybridization of hydroxypropyl-ß-cycoldextrin and hydroxypropylcellulose in dogs,* Journal of pharmaceutics and pharmacology, 1993.

44/ Guang Yan, Henan Li, Ruhua Zhang, and Derong Ding, *Preparation and Evaluation of a Sustained- released Formulation of Nifedipine HPMC Tablets*, Drug development and industrial Pharmacy, 26(6), 681-686, 2000.

45/ Faham A, Prinderre P, Piccerelle P, Farah N, Joachim J, *Hot-melt coating technology: Influence of Compritol 888 ATO and granule size on chloroquine release*, Pharmazie June, 2000.

46/ A.S.Achata,P.S.Adusumilli, K.W. James, and C.T. Rhodes, *Development of hot-melt coating methods*, Drug development and Industrial Pharmacy, Drug Development and Industial Pharmacy, Vol.23 (5), pp.441-449, 1997.

47/ John P. Kennedy, *Hot-melt coatings in a Fluidized Bed,* International Processing Corp., 2000.

48/ E. Nicolas Griffin and Paul J. Niebergall, *Release Kinetics of a Controlled-Release Multiparticulate Dosage form Prepared Using a Hot-Melt Fluid Bed Coating Method,* Pharmaceutical Development and Technology, 4(1), 117-124, 1999.

49/ John P, Kennedy, and Paul J. Niebergall, *Development and optimization of a solid dispersion Hot-melt fluid bed coating method*, Pharmaceutical Development and Technology, 1(1), 51-62, 1996.

50/ John P, Kennedy, and Paul J. Niebergall, *Evaluation of Extended - Release Applications for solid Dispersion Hot-melt Fluid bed Coating Agents*, Pharmaceutical Development and Technology, 3(1), 95-101, 1998.

51/ Gatefosse' factory and laboratories, C*ontrolled release and increased bioavailability*, April, 1999.

52/ Croda document, Croda Inc. 7 Century Drive, Parsippany, NJ 07054

53/ *Convolution and Deconvolution in Kinetica*, Clinical Information Engineering (Innaphase), 2000.

54/ Foster, Hamann, Richards et al., *Nifedipine in normal subjects*, The Journal of Clinical Pharmacology, April 1983.