AAPS PharmSciTech, Vol. 11, No. 4, December 2010 (© 2010)
DOI: 10.1208/s12249-010-9543-4

Research Article

Formulation of Controlled-Release Capsules of Biopharmaceutical Classification System I Drugs Using Niacin as a Model

Monica C. Chuong,[1,3] Luca Palugan,[1,2] Tiffany M. Su,[1] Claudelle Busano,[1] Ronald Lee,[1] Giustino Di Pretoro,[2] and Anee Shah[1]

Received 31 December 2009; accepted 9 November 2010; published online 24 November 2010

Abstract. Vitamin B_3 is made up of niacin (nicotinic acid) and its amide, niacinamide. Both have equivalent vitamin activity, but only niacin (not niacinamide) is effective in lowering elevated low-density lipoprotein cholesterol and triglyceride levels in the blood. Administration of an extended-release (ER) oral tablet would frequently encounter food. If hydrogel is used to formulate the matrix of a biopharmaceutical classification system I drug (high solubility and high permeability), the dosage form absorbs water and swells. The softened outer layer may be slashed off by food present in the stomach, thus, exposing the core tablet more readily for water absorption and speeding up drug release from its original designed rate. This project aimed to formulate niacin CR pellets made of hydrophobic inert matrix. After niacin was melted with excipients and cooled, the mass was extruded and spheronized into pellets. Size distribution and flowability were determined before pellets were filled into hard gelatin capsule. The USP dissolution study revealed that a candidate formulation of 250 mg in strength released similar amount of niacin as its commercial reference, niacin controlled-release 500 mg tablet, in 6 h (223.9 ±23.8 mg, $n=4$ *versus* 259.4±2.6 mg, $n=3$). The differential scanning calorimetry study of the pellets in capsules stored in 40°C for 4 weeks, and the content assay of capsules in 40°C up to 6 months suggested that niacin was stable within the innovative formulation. *In vitro* release from this innovative ER capsules stored at 40°C up to 4 weeks were also investigated.

KEY WORDS: controlled-release; flowability; innovative formulation; *in vitro* release; niacin; particle size analysis; pellets; Raman image.

INTRODUCTION

Cardiovascular disease is responsible for nearly half the deaths in the USA (1). Hyperlipidemia (elevated serum cholesterol levels) has previously been shown to be a major predisposing factor to cardiovascular disease (2). It is diagnosed when serum triglyceride level is greater than 150 mg/dL and serum low-density lipoprotein level is more than 100 mg/dL (2). The therapeutic use of medicinal agents categorized under the antihyperlipidemic class is particularly effective in reducing the risk of mortality associated with cardiovascular disease (1–3). Niacin (also known as vitamin B_3 and nicotinic acid) is a Biopharmaceutical Classification System I drug based on its high solubility in water and good permeability. Its melting point is 236.6°C, pKa 2.2, and log P at 1.0 (4,5). Niacin reduces serum cholesterol levels by blocking hepatic production of triglycerides and also prevents the secretion of very low-density lipoproteins (6). However, clinical use of both prescription-required and over-the-counter niacin products formulated as immediate-release or controlled-release (CR) has declined due to the frequency of adverse reactions including flushing and gastrointestinal disturbances (6,7). The effective therapeutic dose of the commercial reference for treating hyperlipidemia in adults is between 1,200–4,000 mg per day (6). It was reported to release niacin more rapidly in hydrochloric acid medium than in phosphate buffer based on an *in vitro* dissolution study (8).

Therefore, the overall purposes of this project were threefold. The first aim was to develop an innovative extended-release (ER) niacin capsule with a near-zero order release profile in a two-stage *in vitro* dissolution study. In order to achieve so, a commercial CR tablet was chosen out of current marketing niacin products to reference the efficacy of our formulation. In addition, several pre-formulation works would be included to comprehend the physiochemical properties of the active pharmaceutical ingredient. The second aim was to recommend a dissolution study for ER niacin tablets because there are no such guidelines in the compendium yet (9). An appropriate assay, therefore, would also have to be developed to quantify *in vitro* amount of drug release, construct release *versus* time profile, and compare formulation candidates to the reference. The last was to determine the stability of the final dosage form employing

[1] Department of Pharmaceutical Sciences, Massachusetts College of Pharmacy and Health Sciences, 179 Longwood Ave, Boston, Massachusetts 02115, USA.
[2] Dipartimento di Scienze Farmaceutiche 'P. Pratesi', Università degli Studi di Milano, Milan, Italy.
[3] To whom correspondence should be addressed. (e-mail: monica.chuong@mcphs.edu)

accelerated storage test once the formulation candidate with the most optimal *in vitro* release was identified.

MATERIALS

Materials

Reagents and Standards

Niacin reference standard (Niacin RS, Supelco, MO) was purchased from Sigma (St Louis, MO). Hydrochloric acid (36.5–38%), sodium hydroxide pellets, and high-performance liquid chromatography (HPLC)-grade acetonitrile were purchased from VWR International (Bridgeport, NJ). Niacin USP, tribasic sodium phosphate was obtained from Professional Compounding Company of America (PCCA, Houston, TX). Salicylic Acid USP Dissolution Calibrators (Lot Q0D200) were purchased from USP (Rockville, MD).

Commercial Products

Two different batches of SloNiacin® Polygel® CR Niacin 500 mg tablets (Lot 235127, Expired October 2009, and Lot 246276, Expired November 2010, Upsher-Smith Laboratories, Inc. as the commercial reference) and 3% hydrogen peroxide were purchased over-the-counter from local pharmacies. SloNiacin® is an over-the-counter product, and the excipients listed in the product label were glycerol behenate, hydrogenated vegetable oil, hypromellose (hydroxypropyl methylcellulose), magnesium stearate, silicon dioxide, and Red 40 (6).

Materials Used in Formulation Works

Niacin USP (nicotinic acid), carbomer 940 (Carbopol® 940, molecular weights between adjacent crosslinks is 104,400 g/mol, pKa 6.0, glass transition temperature 100–105°C) (10), white wax (melting range, 62–65°C), and size 00 gelatin capsules were purchased from PCCA (Houston, TX). Avicel PH 101 (microcrystalline cellulose, particle size 50 μm) was a gift from FMC Co. (Newark, DE). Explotab (sodium starch glycolate, type A, pH 5.5–7.5) and Explotab low pH (type B, pH 3.0–5.0) were purchased from Penwest Pharmaceuticals Co. Kollidon 30 (Povidone, molecular weight 50,000), and Kollidon VA 64 (crospovidone, copolymer of 1-vinyl-2-pyrrolidone and vinyl acetate in 6:4 ratio by mass) were obtained from BASF Co. (Parsippany, NJ). Precirol ATO 5 (glyceryl palmitostearate), Gelucire 43/01 (C_{12}-C_{18} glyceride, melts above 43°C and required hydrophilic–lipophilic balance is 1), and Compritol 888 ATO (glyceryl behenate, melting point 70°C, RHLB 2) were gifts of Gattefosse SAS (Saint Priest, France), Eudragit NE-30 (ethyl acrylate methyl methacrylate copolymer 30% dispersion) was a gift from Evonik Degussa Co. (Piscataway, NJ). Surelease E-7-19019 (ethylcellulose dispersion), Methocel K4M CR (hypromellose 2208, 7–12% hydroxypropyl content, 19–24% methyl content, controlled-release grade), and Opadry (one-step film coating system in powder form, which combines polymer, plasticizer, and pigment) were gifts of Colorcon (West Point, PA).

METHODS

Formulation

Several materials were selected to embed niacin in a lipophilic matrix in order to control its release profile. These included white wax and Gelucire 43/01. The other excipients were then added to the melted substances and stirred. Before complete cooling, the formulation was mixed into a cohesive doughy mass and removed from the beaker. The mass was then further blended on a tile plate to ensure homogeneous dispersion of all ingredients. Once the mass was thoroughly kneaded, it was extruded though sieve size 18 to form granules, then manually spheronized to create round pellets and allowed to cool to room temperature. Pellet flowability and size distribution were next characterized. Finally, the shaped pellets were once again sieved through size 16 (1.18 mm) and size 20 (0.85 mm) sieves to ensure a relatively homogeneous size distribution. The pellets were then stored in a container overnight. In the next morning, a calculated weight (based upon the amount of excipients present in the formulation candidate) that included 275 mg of niacin (110% of the 250 mg label claim) was filled into an appropriate size of hard gelatin capsule.

Characterization of Niacin USP Powder and the Innovative CR Niacin Pellets

Physical Characterization of Niacin USP Powder

Understanding the physical properties of Niacin USP was essential in succeeding a formulation. Niacin powder is known to be poor in flowability (9). The angle of repose for niacin was reported as 50.1±2.0 (n=4), Hausner ratio as 1.70 ±0.1, and Carr's Index as 41.0±2.3 (n=3) (11). All afore parameters indicated poor powder flowability, which presented a problem in formulation that needed to be overcome in order to scale-up later. The particle size distribution of niacin USP was also analyzed by Accusizer™ 780 A—PSS-NICOMP (Particle Sizing Systems, Santa Barbara, CA) which was linked via a single computer to the Nicomp 380 (12). This Windows-based software controller allows both instruments to work together so that the main peak of a sample could be analyzed by Nicomp 380. Experimental conditions applied in the niacin particle size measurement were: analysis time 60 s, resolution 256 channels (0.5–500 μl), flow rate 1 mL/s, and 60 mL of filtered saturated niacin solution prepared by bidistilled water as tested medium.

Chemical Properties of Niacin

Niacin Standard Curves in Water, 0.1 N hydrochloric acid (HCl), pH 2.2 hydrochloric acid medium, and pH 6.8 phosphate buffer

One hundred milligrams of niacin RS was dissolved in 100 mL of deionized water, 0.1 N HCl, pH 2.2 HCl medium and pH 6.8 phosphate buffer, respectively, to create stock solutions. These stock solutions were prepared under the guidelines of the USP32/NF27 (9), and the concentration was

further diluted into 0.5, 0.4, 0.3, 0.1, 0.04, 0.02, and 0.01 mg/mL for each solvent. The concentrations of niacin in each solution were quantified by two assays to construct standard curves. Using Vitamin B_6 (pyridoxine hydrochloride) as the internal standard, the area under the curve of niacin for each concentration in 0.1 N HCl was quantified by HPLC and output by ChromQuest. The Thermo Separation Product HPLC system was composed of a pump (P4000), ultraviolet detector (UV 1000), and autosampler (AS3000) with an X-Terra RP18–5 μM 4.6×250 mm column with a guard column (WAT 044480). Bother were products of Waters Co. (Midford, MA). *Mobile phase was* a mixture of water, methanol, and glacial acetic acid (73:27:1) containing 140 mg of sodium 1-hexanesulfonate per 100 mL. The column flow rate was set 0.45 mL/min with total run time at 30 min/cycle, wavelength 268 nm, and injection volume 50 μL. Absorbance wavelengths of niacin in the aforementioned media concentrations were also determined by an ultraviolet/visible spectrophotometer (UV/Vis, Hewlett Packard, model 8453) equipped with a sipper pump (Agilent, Model 89092AO). Standard curves were then plotted at concentrations of 0.01, 0.02, 0.04, 0.1, 0.3, 0.4, 0.5, and 1 mg/mL in each solution based upon UV absorbance and were used to quantify drug release from candidate formulations.

Drug Substance Solubility

The solubility of niacin as 1 g dissolved in 60 mL of water (4) was used to validate the purity of the niacin USP powder purchased for use in formulation and also to determine the solubility of niacin in the solvents of 0.1 N HCl, pH 2.2 HCl medium, and pH 6.8 phosphate buffer. An excessive amount of niacin USP was added in small incremental amounts to a fixed volume of the solvent (100 mL of water) with the system being vigorously stirred on a stir plate to ensure complete dissolution of the solute in the solvent. Once precipitated solute powder was seen in the system, the saturated solution was filtered though a 0.2-μm polytetrafluoroethylene (PTFE) syringe filter prior to being assayed by the aforementioned ultraviolet spectrophotometer. The separate solute samples were taken per solution and averaged to ensure consistent results. This process was repeated for the remaining three solutions (0.1 N HCl, pH 2.2 hydrochloric acid medium, and pH 6.8 phosphate buffer) to determine niacin solubility in each.

Drug Substance Stability in the Media of Various Physiological pH

To find the stability of Niacin USP powder in different pHs, the known amount was added into a fixed volume of 0.1 N HCl, pH 2.2 hydrochloric acid medium, pH 6.8 phosphate buffer, and 3% hydrogen peroxide to achieve the final concentrations at 0.4 mg/mL, with the exception of hydrogen peroxide stability testing performed at 0.1 mg/mL. The stability of niacin in the former three media was monitored up to 76 h, while niacin stability in 3% hydrogen peroxide was only monitored for 3 h due to the volatility and nature of rapid decomposition of hydrogen peroxide. Samples of all four solutions were assayed at each predetermined time point for all tested media.

Pellet Characterization

Pellet Flowability

The angle of repose of the pellets was measured by pouring a set amount of pellets through a funnel to form a cone, determining the height of the cone of pellets, and calculating the angle of repose, α, from the following equation (9):

$$\tan(\alpha) = \frac{height}{0.5 \cdot base} \tag{1}$$

The angle of repose was calculated in order to determine the flowability of the pellets, which was essential to the proper scale-up capsule filling.

Two other parameters, compressibility index and Hausner ratio, were also measured to determine the flowability of the pellets and to compare with the prediction made by angle of repose. The compressibility index has been used as an indirect measure of bulk density, size, shape, surface area, moisture content, and cohesiveness of pellets (9). Cited in the two equations described in the compendium (9), the compressibility index (also known as Carr's Index) and Hausner ratio were determined using the unsettled apparent volume (V_O) and the final tapped volume (V_f) of 6.78 g niacin powder in a 25-mL graduated cylinder after tapping the material 25 times.

$$\text{Compressibility Index} = 100 \times \left[\frac{V_o - V_f}{V_o}\right] \tag{2}$$

$$\text{Hausner Ratio} = \frac{V_o}{V_f} \tag{3}$$

Both these values were again used to assess the quality of pellet flowability.

Pellet Shape and Size

The pellet shape was determined through visual inspection of the final product as recommended by Randall (13) and the size distribution was determined by sieving analysis through ten different sieves ranging from mesh size 60 to 10 starting from smallest diameter to largest diameter. Conversion of pellet size into millimeters was done by using a table of sieve numbers and opening sizes listed in the literature and commercial catalog (9,14), and the results were reported as weight *versus* size for each group and cumulative percent of total weight *versus* mean particle size. After pellet size distribution was determined and before filling pellets into a hard gelatin capsule, a compressed size distribution of pellets was chosen to improve the consistency of particle surface area as the niacin release profile in simulated *in vitro* studies would be pellet surface area related.

In Vitro Dissolution Study

Both the commercial reference and the formulation candidates were tested by the following methods using a

USP dissolution apparatus I containing seven vessels (Distek Premiere 5100, North Brunswick, NJ).

Development of Assay Process

Selection of an appropriate assay process was essential to appropriately evaluate a candidate formulation. Since this was an ER capsule formulation project, dissolution studies needed to be conducted from simulated gastric medium to simulated small intestine medium *in vitro*. There are two methods of medium change described in USP32/NF27 (9) for extended-release dosage forms: method A (add directly) and method B (drain and add). But, Chuong *et al.* (8) suggested using method A for medium change during the dissolution study of a Polygel® formulation as the approach is more closely mimicking human gastrointestinal tract environment. Therefore, the protocol for method A was followed.

USP Dissolution Guideline for Niacin Tablet

The dissolution guidelines for a niacin containing formulations in the Niacin Monograph of USP32/NF27 (9) is for immediate-release tablets, which stated to use 900 mL 0.1 N HCl with the USP dissolution apparatus 1 (the basket method) at the stirring rate of 100 repetitions per min (rpm) for the duration of 60 min. However, there is no specific dissolution guideline for the extended-release niacin tablet. Therefore, the dissolution guidelines (9) were adopted for this project. This general chapter stated that "The study tablet is placed in 750 mL 0.1 N HCl with basket stirring rate at 100 rpm for 2 h and then add 250 mL of 0.20 M tribasic sodium phosphate in the vessel and if necessary, with 2 N hydrochloric acid or 2 N sodium hydroxide to a pH of 6.8± 0.05 (9). But, the USP32/NF27 does not specify the test duration in pH 6.8 phosphate buffer. Therefore, in this project, the dissolution time in pH 6.8 was chosen as 4 h with a stirring rate at 50 rpm, based upon the known retention of an oral dosage form in the small intestine and the decreased peristalsis experienced once niacin formulation leaves the stomach (15). Samples of 3 mL were collected at the schedule of 0.25, 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, and 6 h from each vessel and filtered through a 0.2-μm PTFE filter into a disposable test tube. The medium in each dissolution vessel was replenished after each withdrawal. Samples were then assayed with the UV spectrophotometer and converted into the cumulative amount of niacin release for the time point through utilization of the established standard curves in these two media, 0.1 N HCl and pH 6.8 phosphate buffer.

Apparatus Calibration

In order to validate the accuracy of results obtained from the dissolution apparatus utilized for the studies, it was necessary to obtain a correct calibration prior to conducting *in vitro* dissolution for formulation candidates. Based upon the USP Procedure (16), the USP dissolution calibrators were purchased. The 0.05 M phosphate buffer dissolution medium was prepared in accordance to the instruction sheet arrived with the USP dissolution calibrator with monobasic potassium phosphate, deionized water, sodium hydroxide, and appropriate amounts of either sodium hydroxide or phosphoric acid to achieve a pH of 7.4±0.05. A standard curve of the calibrators in the solvent was created in order to quantify the results of the calibration test. The calibration dissolution run was conducted at 37°C in 900 mL medium at 100 rpm for 30 min. After 30 min, 4 mL samples were obtained from each vessel and filtered through 0.02-μm PTFE filters. The samples were then assayed with ultraviolet spectrophotometer to determine percentage release of the calibrator and compared with the specific ranges listed in the instruction sheets inside the calibrator order package.

Pharmaceutical Stability and Thermal Analysis

In order to verify a successful formulation, the optimal candidate in the final dosage form was assessed for its pharmaceutical stability through an accelerated storage test. One batch of niacin ER capsules was placed in a 40°C, 60% relative humidity laboratory incubator up to 6 months while a second batch of capsules was placed at room temperature. At time 0, 2 weeks, 1, 3, and 6 months, three capsules at each predetermined sampling time were removed from the incubator. The contents of each batch of capsules were emptied from the hard gelatin capsules and ground up with a mortar and pestle. The ground up ingredients of one capsule were placed in a beaker of 0.1 N HCl and heated to a temperature of 65°C (monitored by a thermometer) while stirring on a stir plate to ensure complete melting of the lipophilic excipients used to embed the highly water soluble niacin. Once the lipophilic excipients fully melted, the embedded niacin was released and dissolved from the oily phase into the aqueous solvent phase. The beaker was then placed into an ice bath. This step was essential to remove low melting temperature lipophilic excipients from the solution so that they would not solidify in room temperature and block HPLC tubing. A four-milliliter sample was then removed from the beaker and filtered through a 0.2 μm PTFE filter before assaying with ultraviolet spectrophotometer.

Differential scanning calorimetry (DSC, Seiko Instruments model SSC/220 C) linked via a single computer to the EXSTAR software (Woodland, CA) were used to test the thermal behaviors of four samples in 5 to 6-mg weight range. They were niacin USP powder, the most optimal formulation pellets at time 0, pellets stored at 40°C for 2 weeks and 4 weeks.

***In Vitro* Release Study of ID 37 Niacin ER Capsules Stored at 40°C 2 Week and 4 Week**

Capsules filled with the most optimal formulation pellets were stored in 40°C 60% RH to conduct an accelerated storage test. After being in an incubator for 2 weeks and 4 weeks, respectively, three capsules each time were taken out and placed in 750 mL 0.1 N HCl to conduct dissolution study with the basket stirring at 100 rpm for 2 h and then add 250 mL of 0.20 M tribasic sodium phosphate into 1,000 mL of pH 6.8 phosphate buffer. The stirring rate of the apparatus was reduced to 50 rpm to continue dissolution study for another 4 h to examine the *in vitro* release pattern of the capsules.

Data Management

The release data of the innovative formulations were constructed into the release profiles of cumulative amount of drug release (in milligrams) *versus* time (in hours) and compared with that of the commercial reference, whose release profile was constructed concurrently for each batch. Excel 2000 (Microsoft) was used to manage raw data. Using SigmaStat 3.5 (statistical software), independent *t* tests were performed for two-group data analysis when normality and equal variance met for the total amount of drug release at the end of a dissolution study. Population differences are considered significant at $P<0.05$ (17).

RESULTS

Formulation

Overall, 38 total candidate formulations were developed and tested (Fig. 1a, b). Two novel formulations were considered as optimal candidates. The first step of the pellet-making process was to heat niacin together the lipophilic excipients in a glass beaker until softened and well blended (to approximately 65°C). The other excipients were then added to the softened substances and continuously stirred with a glass stir rod to ensure homogeneous dispersion of all ingredients. Before complete cooling, the formulation was blended into a cohesive doughy mass and removed from the beaker. The mass was then further blended on a tile plate. Once the mass was thoroughly kneaded, it was extruded through a sieve sized 16 (1.18 mm) to form granules. The granules were then manually spheronized to create round pellets and allowed to cool to room temperature. Once pellet size distribution analysis was conducted, the pellets in size between sieves 16 to 20 (0.85 mm) were harvested and package into capsules containing 110% of 250 mg niacin. Both formulations #36 and #37 considerably slowed the release profiles of a 275 mg (110% of our proposed strength, 250 mg) niacin containing capsule. The successful formulations were listed in Table I. Compared with formulation #36, Methocel K4M CR was removed from formulation #37, and the amount of carbomer present in the formulation increased. After further evaluation, formulation #37 was chosen as the optimal formulation due to its slightly higher amount of niacin release over a 6-h *in vitro* dissolution test.

Characterization of Niacin USP Powder and the Innovative CR Niacin Pellets

Physical Characterization of Niacin USP Powder

Based upon the Raman spectroscopic images, the physical shape of niacin USP powder was found to be an aggregated crystalline in structure (Fig. 2). The image also confirmed its cohesive property. This physical property of niacin observed under the microscope supports the poor flowability of the powder predicted by angle of repose, compressibility index, and Hausner ratio and presented a challenge in formulating a pellet with acceptable flowability characteristics.

Accusizer™ 780 A was used to examine niacin powder USP particle size distribution. Analysis was performed on 60 mL saturated filtered niacin solution and sample time was set at 60 s.

Assuming the tested Niacin USP was spherical in shape, the particle size distribution was reported in three different *Y*-axes: (a) number, (b) projection surface area, and (c) volume with respect to equivalent particle diameter as *X*-axis. The median particle size output by number particle size distribution of Accusizer™ was 7.13 μm (Fig. 3a).

Chemical Properties of Niacin USP

Niacin Standard Curves in Water, 0.1 N HCl, pH 2.2 HCl Medium, and pH 6.8 Phosphate Buffer

The quantification methodology using HPLC is not only more expensive and time-consuming than using UV/Vis but also has the disadvantage of hazardous organic chemical waste produced by the mobile phase. The niacin standard curve for 0.1 N HCl was measured through HPLC and UV spectrophotometer. After a good correlation between UV absorbance and HPLC AUC ratio of niacin and its internal standard, Pyridoxine HCl RS (vitamin B_6; Fig. 4a, b), this project continued the remaining quantifications by using UV. Data in pH 2.2 HCl medium, water and pH 6.8 phosphate

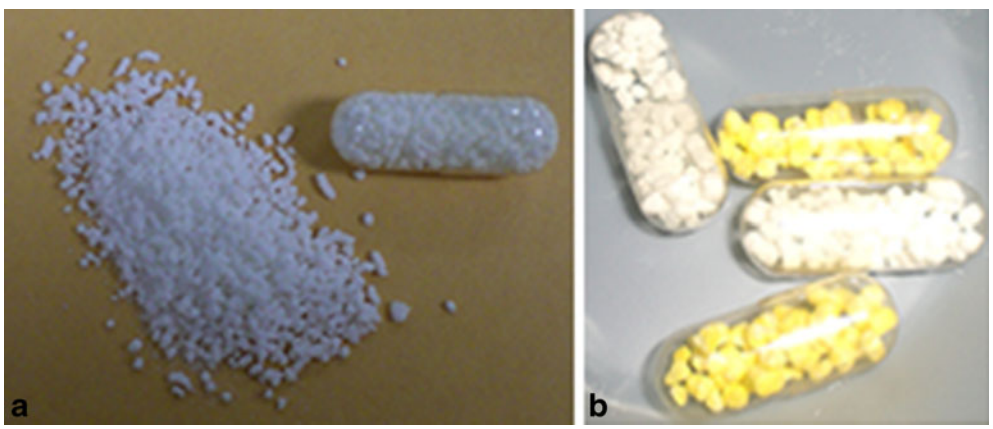


Fig. 1. Unencapsulated pellets and pellets filled into size 00 hard gelatin capsules in the final dosage form for ID #37 (**a**) and the coated pellets (**b**)

Table I. Composition and Function of Excipients in the Top Two Candidate Formulation Choices

Ingredient	Function	Melting Point (°C)	ID 36 (mg/capsule)	ID 37 (mg/capsule)
Carbomer	Suspending agent	260	7.5	25
Hypromellose	Binder	170–180	20	0
Gelucire 43/01	Lipophilic base	43	50	50
Docusate Sodium	Surfactant	153–157	4	4
White Wax	Lipophilic base	61–65	80	80

buffer are not shown. The most optimal absorbance wavelengths of niacin in the media of 0.1 N HCl and pH 2.2 HCl medium determined by UV/Vis were found at 261 nm, while the most optimal absorbance in deionized water and pH 6.8 phosphate buffer were determined to be 263 nm. The linearity of niacin standard curve in 0.1 N HCl ranged from 0.001 to 0.4 mg/mL; the linearity in pH 2.2 HCl medium ranged from 0.0016 to 0.5 mg/mL, while that in pH 6.8 phosphate buffer was from 0.0016 to 0.9 mg/mL; and in deionized water from 0.0016 to 0.4 mg/mL with correlation coefficients (R^2) greater than 0.999 for each solvent (data not shown). These curves were used to qualify the *in vitro* dissolution of candidate formulations.

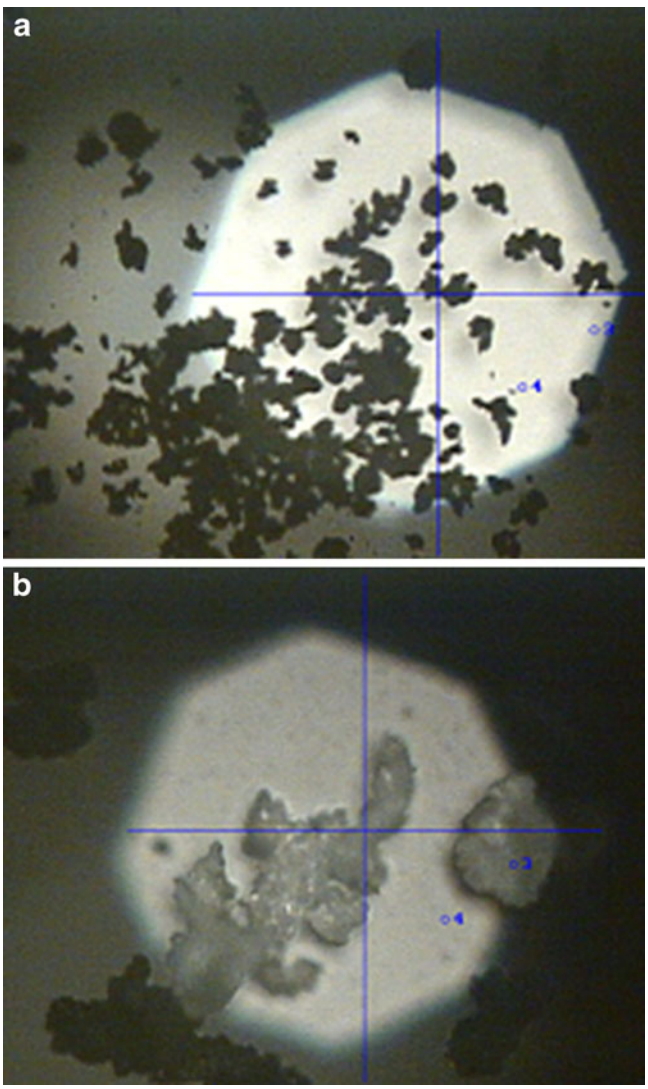


Fig. 2. Niacin powder under Raman Microscope 5× (**a**), and 20× (**b**) illustrating that niacin USP is irregularly shaped interlocking particles

Niacin Solubility in Simulated Gastrointestinal Media

Niacin USP purchased for formulation was found to have the solubility of 0.96 g in 60 mL of deionized water (Table II), which is similar to the value listed in Merck Index (4). This finding validated the purity of the niacin used for candidate formulations. The solubility of niacin, an ionizable species, in the four solvents used (from least to most) were water< pH 2.2 HCl medium<pH 6.8 phosphate buffer<0.1 N HCl (Table II). The relative standard deviation for the solubility of niacin in each solvent was determined to be less than 5%, indicating an accurate assessment.

Niacin Stability in Various Simulated Gastrointestinal Media

Through ultraviolet spectrophotometer assay of the samples, niacin USP powder was found to be stable in 0.1 N HCl, pH 2.2 HCl medium, and pH 6.8 phosphate buffer for a period up to 76 h (>3 days). Niacin was also determined to be stable in 3% hydrogen peroxide for a period of 3 h (Table III). The stability of niacin under these conditions was promising, indicating that niacin samples may be assayed together at the end of a 6-h dissolution experiment without a significant issue of solvent-based degradation.

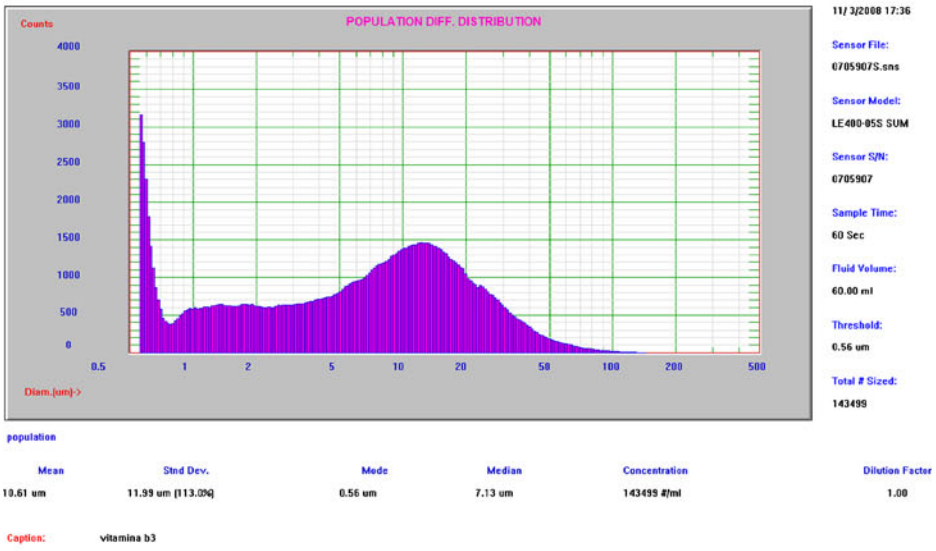
Pellet Characterization

Pellet Flowability

The optimal candidate formulation (6.78 g of pellets) was poured through a funnel to determine the angle of repose. This amount was obtained by sieving 10 g of pellets through a size 20 sieve (0.85 mm) to ensure relative homogeneity of size in order to decrease outliers. The funnel height was maintained approximately 2.8 cm from the top of the pellet pile. The angle of repose for formulation #37 was determined to be 35.13° (Table IVa). This angle was small enough to provide good flow properties to the pellets according to the scale of Flow Properties and Corresponding Angles of Repose given by USP32/NF27 (9). This proved that the pellet formulation

Fig. 3. Accusizer™ 780 A output the particle size distribution of ▶ niacin powder USP by: **a** number, **b** projection surface area, and **c** volume, with respect to equivalent particle diameter. Analysis was performed on 60 mL saturated filtered niacin solution; sample time was 60 s

a

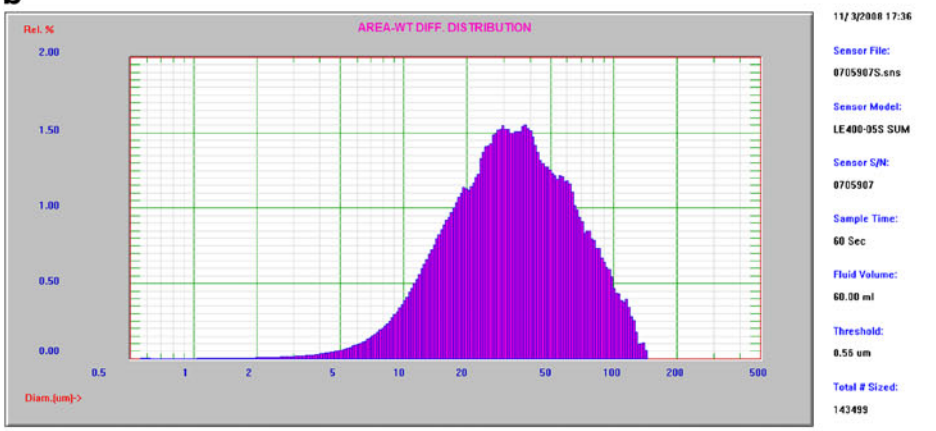


population

Mean	Std Dev.	Mode	Median	Concentration	Dilution Factor
10.61 um	11.99 um (113.0%)	0.56 um	7.13 um	143499 #/ml	1.00

Caption: vitamina b3

b

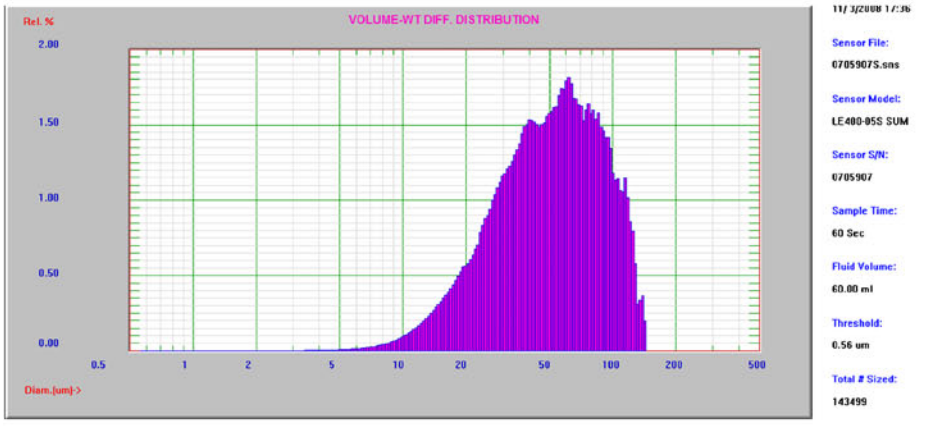


population

Mean	Std Dev.	Mode	Median	Concentration	Dilution Factor
39.61 um	26.13 um (66.0%)	38.00 um	32.32 um	143499 #/ml	1.00

Caption: vitamina b3

c



population

Mean	Std Dev.	Mode	Median	Concentration	Dilution Factor
56.84 um	30.27 um (53.3%)	61.77 um	51.14 um	143499 #/ml	1.00

Caption: vitamina b3

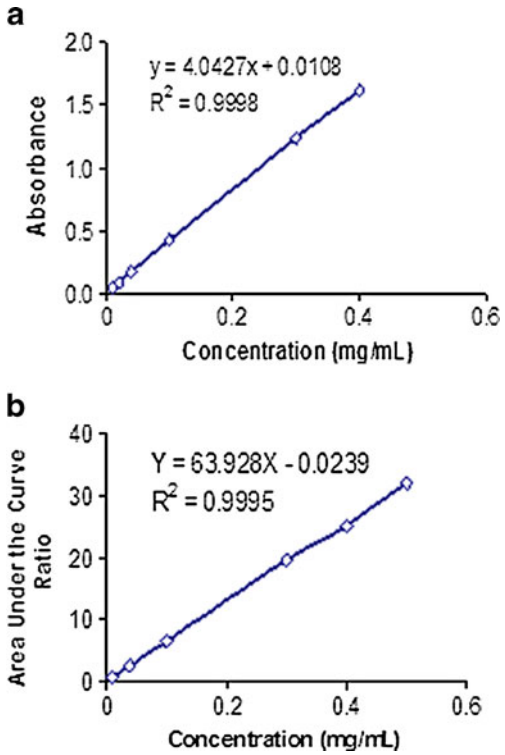


Fig. 4. Niacin standard curve in 0.1 N HCl constructed by ultraviolet spectrophotometer (**a**), and high-performance liquid chromatographic method (**b**). The niacin monograph in USP32/NF27 (9) indicates that niacin may be quantified by either method

was able to overcome the poor flow properties of niacin USP and increased ease of capsule filling at the industrial level. The compressibility index was determined to be 12.9 while the Hausner ratio was computed to be 1.15 (Table IVb). Combining these two values, the pellet flowability of formulation #37 was again determined to be good based on the table values given in USP32/NF27 (9). This further validated the acceptable flowability properties of the optimal candidate, ensuring a pharmaceutically elegant formulation.

Pellet Size and Shape

Determination of particle size was important to assess batch variability, and the ideal pellet shape for the formulation is a spherical form to facilitate homogeneous diffusion of an active ingredient and proper pellet coating. However, due to the limitations of manual spheronization, the pellet shape of formulation #36 was determined to be irregular granular. Assessment of the good flow properties of the pellets performed demonstrated that this slightly irregular shape was not a hindrance to formulation. Furthermore, it is expected that, at the industrial level, with the use of an extruder and spheronizer, the pellet shape will indeed be more spherical, further increasing the elegance and homogeneity of the product.

One batch of prepared formulation #37 pellets was sieved through ten different sieve sizes of increasing diameter to determine the distribution of pellet size. It was found that the largest amount of pellets were from sieve #20 and sieve #18 (Fig. 5a). The largest concentration of pellet size was thus determined to be between 0.850 and 1.0 mm. To quantify the pellet size, the cumulative percent of pellet particle size was graphed, demonstrating that the size of the total amount of pellets was concentrated above 1.0 mm (Fig. 5b). Furthermore, in order to decrease variability during dissolution studies, capsules for *in vitro* studies were filled only with pellets sieved through sieves #20 and #16 (0.85 mm to1.18 mm).

Table II. Solubility of Niacin USP Powder in Various Simulated Gastrointestinal Media

Solvent	Solubility in 60 mL (Mean±1 SD)
Water	0.97 g
0.1 N HCl	1.80±0.01 g
pH 2.2 HCl buffer	1.05±0.03 g
pH 6.8 phosphate buffer	1.20±0.01 g

Table III. Degradation of Niacin USP Powder in Various Simulated Gastrointestinal Media

Media	Concentration (mg/mL)	Time (h)	Degradation
0.1 N HCl	0.4	76	Not detected
pH 2.2 HCl Buffer	0.4	76	Not detected
pH 6.8 Phosphate Buffer	0.8	76	Not detected
Hydrogen Peroxide	0.1	3	Not detected

In Vitro Dissolution Study

Apparatus Calibration

A non-disintegrating calibrator was utilized to validate the dissolution apparatus, since both the commercial reference tablet and the hydrophobic matrix candidate formulations were non-disintegrating systems. Therefore,

Table IV. Pellet Flowability of ID #37: Angle of Repose and Compressibility Index and Hausner Ratio

A	
Flowability parameters	Tested pellets 6.78 g
Height of pellet cone	1.9 cm
Average of eight measured radius	2.7 cm
Tan α	0.704
Angle of Repose	35.13°
Flow property	Good
B	
Total weight of niacin pellets tested	6.78 g
Poured volume	15.5 mL
Tapped volume	13.5 mL
Poured density	0.438 g/cm^3
Tapped density	0.502 g/cm^3
Compressibility (Carr) Index	12.9
Hausner ratio	1.15
Flow character	Good

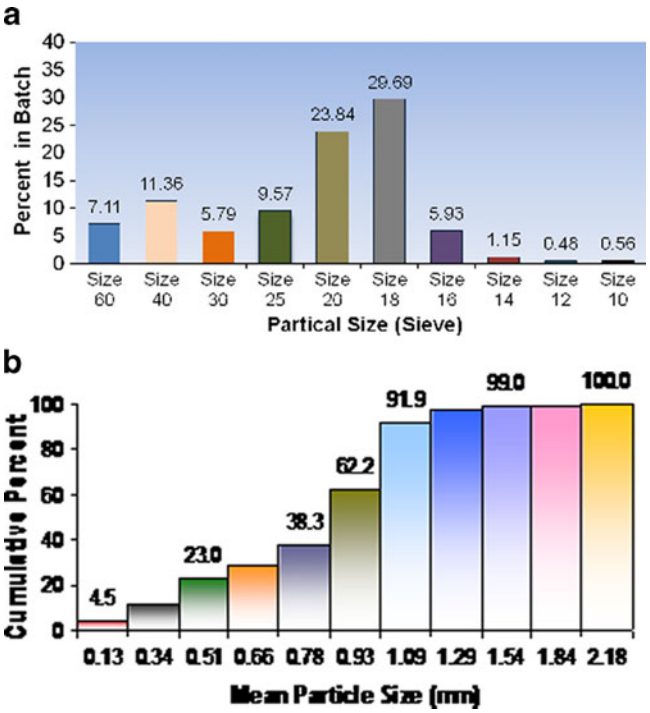


Fig. 5. Formulation pellet size distribution: **a** weight distribution (in grams) in each tested sieve size; **b** cumulative percent *versus* mean particle size (in millimeters) plot

USP salicylic acid non-disintegrating calibrators (300 mg Lot Q0D200) were used to calibrate the seven-vessel dissolution station utilized in the study. The maximal absorption wavelength for salicylic acid was measured at 296 nm. The average amount of salicylic acid released from a USP non-disintegrating calibrator was determined at 24.7± 0.4% with the relative standard deviation at 1.62%. This result suggested that the dissolution apparatus would be used in this study met the required percentage range of release from 23—30% mandated in the calibrator instruction sheet sent by USP (16).

In Vitro Dissolution

As mentioned in the METHODS section, the methods listed in the USP32/NF 27 (9) were used to compare niacin ER pellet-filled capsules with a commercial reference. To simulate a fasting state *in vivo* study, the first 2 h of the *in vitro* dissolution run were conducted in 750 mL 0.1 N HCl (9). At the completion of acidic stage, 250 mL of 0.20 M tribasic sodium phosphate were added, and the pH was adjusted to 6.8±0.5 with 2 N sodium hydroxide and/or 2 N HCl. The last 4 h of the *in vitro* study were conducted at pH 6.8 ± 0.5, simulating the *in vivo* path of the formulation through to the more basic small intestine (15). The goal of the optimal formulation was to create a release profile that was both slower than the commercial reference and also more linear. Both formulations #36 and #37 were able to achieve a release profile slower than the commercial reference. At the end of 6 h, formulation #36 released an average of 182.5±4.5 mg of niacin (n=3), while formulation #37 released an average 223.9±23.8 mg (n=4). This was compared was the incomplete release of the commercial reference after 6 h averaging 259.4 ±0.6 mg of release (n=3, Table V, Fig. 6). The commercial reference which loaded with 500 mg of niacin only had a 51.9% release. Our formulation candidates loaded with 250 mg of niacin. Formulation #36 demonstrated a 73% release, while #37, an 89.6% release. Both of the most optimal formulations of the project were thus superior to the commercial reference based upon the total release. Formulation #37 had the most efficient release of all (Table V).

However, a formulation candidate cannot be judged alone by the total amount of release at the end of a dissolution study. Achieving a linear release is also a goal of the optimal formulation candidate, allowing for a zero-order release factor that would allow dissolution of the dosage form to be consistent in possible future *in vivo* studies. Therefore, the linearity of the release profiles of formulations #36 and #37 were further compared with that of the commercial reference (Fig. 6). Both formulations #36 and #37 had slower release profiles than the commercial reference. However, based upon the linear regression coefficient (R^2 value) the linearity of each release profile from least to most, formulation #36 (0.84, n=3)< commercial reference (0.86, n=3)<formulation #37 (0.93, n=4). When comparing #37 to #36, it was found that although the rate of release under *in vitro* gastric conditions (during the first 2 h in 0.1 N HCl) was approximately the same, formulation #37 actually sped up its release in *in vitro* small intestine pH medium (Fig. 6).

Content, *in Vitro* Release and Thermal Analysis of Capsules Retrieved from Accelerated Stability Study

After formulation, candidate #37 showed near-zero-order *in vitro* release pattern for 6 h and most efficient in drug release (89.6%); more capsules were fabricated by filling pellets equivalent to 275 mg of niacin into a size 00 hard gelatin capsule. They were stored in a 40°C, 60% RH laboratory incubator to conduct accelerated stability study. Heat-stressed capsules were removed from the incubator according to a preset schedule to examine the content, *in vitro* release, and heat-flow behaviors. The obtained results were then compared with the data obtained from analyzing the capsules stored at room temperature for the same time period. DSC was conducted with four samples: niacin USP powder and formulation #37 pellets stored at 40°C for Time 0, 2 weeks, and 4 weeks. The melting point of niacin, 236.6°C, was consistent among all four DSC curves (Fig. 7a–d). The melting event at 43°C, 67°C, and 180°C in the reported DSC temperature heat-flow curves (Fig. 7b to d) matched the melting points of Gelucire 43/01, white wax, and Methocel

Table V. Comparative Values of *In Vitro* Release Efficiency in Two Competing Candidate Formulations *versus* the Commercial Tablet Reference

Formulation ID (mg niacin/dosage form)	Niacin released in 6 h (mg)
Commercial reference (500 mg/tablet)	259.4±0.6
ID 36 (250 mg/capsule)	182.5±4.5
ID 37 (250 mg/capsule)	223.9±23.8

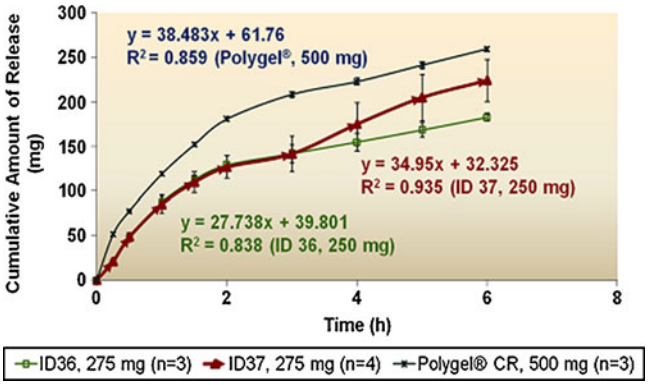


Fig. 6. Comparative dissolution of innovative formulations ID #36 and #37 and reference table product (see text)

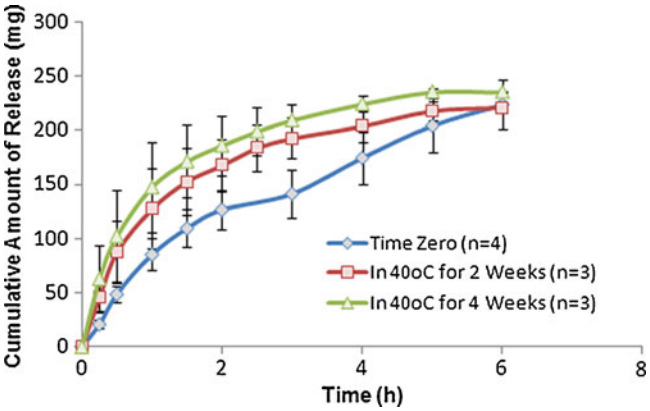


Fig. 8. Comparative dissolution profiles of niacin ID #37 ER capsules stored at 40°C for two and 4 weeks with that of time 0 samples (see text)

K4M CR listed in the literature (10) and Table I. Three formulation #37 capsules were also randomly taken out from the container stored at 40°C to conduct *in vitro* dissolution study. The *in vitro* release of niacin from formulation #37 capsules which had been exposed to 40°C for 2 and 4 weeks, respectively, were less linear and more hyperbolic in shape than those stored in room temperature (Fig. 8). The observation suggested implicitly that the porosity of the pellet matrix might have been markedly affected by environmental humidity and temperature, especially that the melting point of Gelucire 43/01 is around 43°C. The storage temperature of 40 ±1°C might melt Gelucine 43/01. And the interaction of niacin which is a highly water soluble active ingredient with water molecules contributed by the environmental humidity might lead to channel formations and releases its drug faster than the original design.

In order to verify stability of our proposed formulation strength, 250 mg, a content assay was expected to be over 90% of niacin present in a tested capsule. The capsule batch stored at room temperature up to 6 months were determined to be 107.4±0.2% (n=3). Its relative standard deviation (RSD) was 0.21%. Average niacin retrieved from the capsule batch stored at 40°C for 6 months was determined to be 107.1 ±0.7% (n=3) and the RSD was 0.69%.

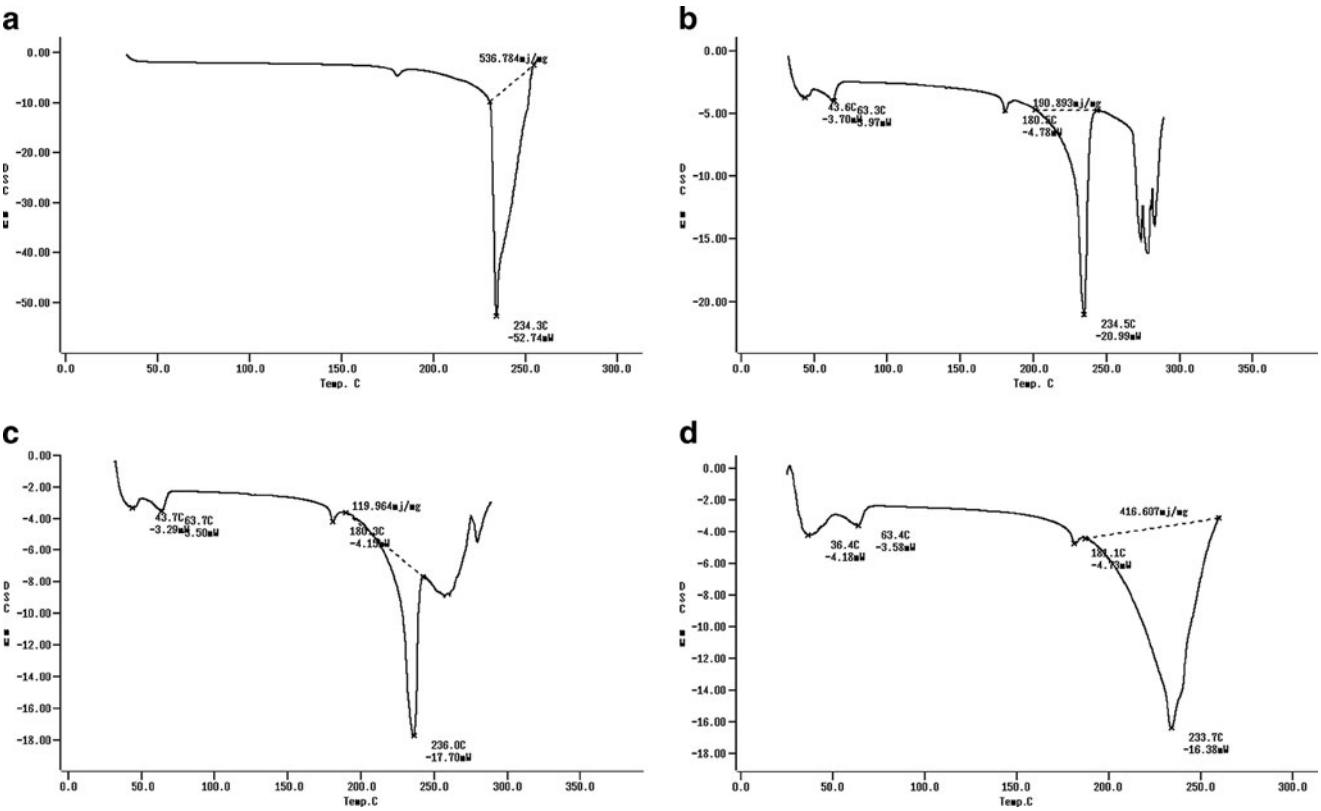


Fig. 7. Differential scanning calorimetric curves of Niacin USP powder (5.5 mg, **a**), formulation ID #37 time 0 pellets (5.2 mg, **b**), and 40°C 2-week (5.5 mg, **c**) and 4-week pellets (5.4 mg, **d**). The melting point of niacin, 236.6°C, was consistent among all DSC curves. The melting at 43°C, 67°C, and 180°C in **b**–**d** matched the melting points of Gelucire 43/01, white wax, and hypromellose (refer to Table I)

DISCUSSION

Niacin powder under Raman Microscope 5× (figure a) and 20× (figure b) illustrating that niacin USP powder is irregularly shaped interlocking particles. This was corresponded to niacin being characterized as poor flowability based on the measurements of angle of repose (50.12±1.093, n=4), compressibility index (41.02±2.29, n=3) and Hausner ratio (1.70±0.07, n=3) (11).

The formulation of a novel dosage form proved to be a challenge. Since the goal of the project was to slow, the release profile of niacin, hydrophilic excipients commonly used for capsule formulations, including lactose and starch, were immediately excluded from formulation testing. Initial formulation experiments also ruled out other excipients including Kollidon® 30 or Kollidon® VA 64, and Avicel® PH 101 and PH 200. Experimentation with other recommended excipients for sustained-release formulations included Compritol® 888 ATO and Precirol® ATO 5, which also failed to modify the release of niacin. These two chemicals are novel materials with lubricant, glidant, and putative binding action (18). Eventually, carbomer (Carbopol® 940) and Methocel® K4M CR was chosen as a controlled-release polymer to help linearize the release profile of the candidate formulations. Both Gelucire® 43/01 and white wax (beeswax) were chosen for their highly lipophilic characteristics, an essential embedding substance to control the release of niacin from the formulation. A surfactant in tiny amount was added to help with homogeneous blending of the hydrophilic and lipophilic ingredients together to form a workable doughy mass. After being extruded through sieve size #20 (0.85 mm), the needed pellets to contain 110% of 250 mg niacin was determined. In order to decrease variability in dissolution results, capsules were filled only with pellets sieved through sieves #20 and #18 (1 mm) in order to ensure homogeneity of particle size because the release performance in an *in vitro* dissolution study is pellet specific surface dependent.

The gastric release profile was the most important aspect due to the acidity of niacin determining the more acidic stomach to be its primary site of absorption and its impeded absorption due to ionization in the basic pH of the small intestine. The differences between formulations #36 and #37 were achieved by altering the amounts of the excipients methocel and carbomer. Methocel® K4M CR is a tablet binder and viscosity increasing agent. This agent was used in formulation #36, but not in formulation #37, which might be explained why niacin release in pH 6.8 phosphate buffer from formulation #36 was hindered (Fig. 6). The amount of Carbomer® in formulation #37 was almost three times that of formulation #36 (20 mg *versus* 7.5 mg, Table I). Carbomer (Carbopol® 940) is an aqueous gel of acidic polymers, which become ionized and expel each other at approximately pH 7.0 into the largest volume. This character allowed niacin to diffuse through open pore channels in the dosage form and release into the dissolution medium. Since both the gastric release profiles of formulations #36 and #37 were similar, ultimately formulation #37 was chosen over #36 because its more efficient release profile in phosphate buffer stage dissolution, the simulated small intestine condition. Formulation #37 was thus the most optimal formulation candidate of all for the reasons of being efficient in niacin release from the lipid-based pellets, higher in release linearity. Additional merits added to the preference of this formulation, ID #37, are its shorter list of required excipients and outer coating over the pellets is not required. All of these therapeutic and economical strengths endorsed the selection of formulation #37.

Differential scanning calorimetric study of niacin USP (5.5 mg), formulation #37 pellets at time 0 (5.2 mg), at 40°C for 2 weeks (5.5 mg) and 4 weeks (5.4 mg; Fig. 7a–d). The melting point of niacin was consistent among all curves, and no significant unidentified peak was observed before the melting point. Accelerated storage study has been used to detect the content loss. If a product has 90% or more of its label claim at the end of 3 months at 65°C, 75% RH or 6 months at 40°C, 60%, it may be referred as 2-year room temperature shelf life (19). But, it is questionable whether an *in vitro* release study of ER capsules underwent an accelerated storage condition may be used to reflect the outcome of *in vivo* release, because a drug product is generally stored in room temperature. Accelerated storage study at 40°C and 65°C have been used to establish the shelf life of a pharmaceutical product, but are seldom used to predict *in vivo* release. The reason is that hydrophobic inert matrix for a water soluble active ingredient to make into ER pellets are frequently made from extruable drug dough mass containing some lower melting point excipients. Thus, future study to compare *in vitro* drug release between capsules stored in room temperature up to 2 years and those undergoes accelerated storage at 40°C up to 6 months will have to be conducted.

CONCLUSIONS

The formulation of a novel dosage form for niacin was achieved. After pre-formulation characterizations and purity validations of the niacin USP powder, 38 different candidate formulations were subjected to *in vitro* dissolution studies. The choice was made to forego coating of the pellets due to coating inconsistencies expressed through *in vitro* studies. The optimal formulation, ID #37 (250 mg in strength), surpassed the commercial reference product (500 mg in strength), which known adverse effects include skin flushing and gastrointestinal distress, in percent of release, linearity of release, and slower release profile. The *in vitro* release study for the innovative niacin ER 250 mg capsules stored in room temperature up to 2 years in comparison to those stored in 40°C up to 6 months would have to be investigated in the future.

ACKNOWLEDGEMENTS

Authors would like to express their gratitude to the Summer Undergraduate Research Fellowship Committee and the Faculty Development Committee of Massachusetts College of Pharmacy and Health Sciences for their financial supports toward this project.

REFERENCES

1. Sanborn KD. Physician Desk Reference, 62th ed. SloNiacin Tablet, Niaspan ER Tablet, Advicor Tablet. Montvale New Jersey: Thomson, 2008. p. 318, 1759, 1768, 3279

2. Bamba R, Scanu AM. "Niacin and Lipoprotein(a): Facts, Uncertainties, and Clinical Considerations.". Am J Cardiol. 2008;101(8A):44B–7B.
3. Dale MM, Moore PK, Rang HP, Ritter JM. Pharmacology. Scotland: Elsevier Science Limited; 2003. p. 309–13.
4. Budavari S. The Merck index: An encyclopedia of chemicals, drugs and biologicals. New Jersey: Whitehouse Station: Merck & Co., Inc; 2001. p. 6552.
5. Lemke TL, Williams DA, Roche VF, Zito SW. Foye's principles of medicinal chemistry. Philadelphia: Lippincott Williams & Wilkins; 2008. p. 1349.
6. MicroMedex. "DRUGDEX® Evaluations—Niacin". New Hersey: Thomson Healthcare, 2008. <http://www.thomsonhc.com/hcs/librarian/ND_T/HCS/ND_CPR/SearchByCategory/ND_PR/Drugs/CS/532650/DUPLICATIONSHIELDSYNC/7BBE60/ND_PG/PRIH/ND_B/HCS/SBK/1/ND_P/Drugs/PFPUI/b8Y1pS2rM9KA5/PFActionId/hcs.common.RetrieveDocumentCommon/DocId/1278/ContentSetId/31/SearchTerm/niacin>.
7. Davidson M. Niacin use and cutaneous flushing: mechanisms and strategies for prevention. Am J Cardiol. 2008;101(8A):14B–9B.
8. Chuong MC, Lee R, Saxena V, Palugan L. Examining whether USP disintegration apparatus with a disk is superior to USP dissolution apparatus in predicting drug release during simulated fed state. Dissolution Technologies. 2009;16(1):25–32.
9. US P32/NF27. Chapter 711: Dissolution, Chapter 776: Optical Microscopy, Chapter 786: Particle Size Distribution, Chapter 1174: Powder Flow, and Niacin in USF Monograph. Rockville MD: Convention, Inc.; 2009. p. 263, 302, 307, 688 and 3077.
10. Rowe RC, Sheskey PJ, Quinn ME . Handbook of Pharmaceutical Excipients. Washington DC: American Pharmaceutical Association and Pharmaceutical Press; 2009. p. 111, 327, 517, 772 and 779.
11. Chuong MC and Palugan L. Innovative Sustained and Modified Release Niacin Tablets: Formulation and Drug Release Patterns. Massachusetts College of Pharmacy and Health Sciences, Faculty Development Grant Report; 2008
12. http://www.labrecyclers.com/product.html?InventoryID=1156
13. Randall CS. Chapter 6: Particle Size Distribution. In: Physical Characterization of Pharmaceutical Solids. New York: Marcel Dekker, Inc.; 1995. p. 158 and 170.
14. vwr catalog; 2008. p. 1748.
15. Dressman JJ, Kramer J. Pharmaceutical dissolution testing. New York: Informa Healthcare USA Inc.; 2005. p. 335–6.
16. www.usp.org
17. Zar JH. Biostatistical analysis. Upper Saddle River: Prentice Hall; 1999. p. 80–1.
18. www.gattefossecanada.ca/fr/products/pharmaceutical/sustained_release.shtml#compritol
19. Cha J, Ranweiler JS, Lane PA. Stability Study. In: Ahuja S, Scypinski S, editors. Handbook of modern pharmaceutical analysis. San Diego: Academic; 2001. p. 445–83.