International Journal of Drug Delivery 5 (2013) 167-176
http://www.arjournals.org/index.php/ijdd/index

Original Research Article

ISSN: 0975-0215

Comparative pharmacokinetic study of two lyophilized orally disintegrating tablets formulations of vinpocetine in human volunteers

Rasha M. Kharshoum[1], Rania A. Sanad [2] , Ahmed M. Abdelhaleem Ali[1]

*Corresponding author:

Rasha M. Kharshoum

[1]Department of Pharmaceutics, Faculty of Pharmacy, Beni-Suief University Egypt
[2]Department of Pharmaceutics, National Organization of Drug Control and Research (NODCAR), Cairo, Egypt

Abstract

Vinpocetine is a poorly water soluble drug, commonly used in treatment of various cerebral insufficiency conditions. The aim of this work was to formulate vinpocetine in the form of orally disintegrating tablets (ODTs) and enhance its solubility and dissolution rate. This objective was addressed using lyophilization technique of either solid dispersion using polyethylene glycol 4000 (PEG 4000) or inclusion complex with 2-hydroxypropyl β-cyclodextrin (2HP-β-CD). Differential scanning calorimetry (DSC) and fourier transform-infrared (FT-IR) spectroscopy were used to characterize the solid state of the prepared solid complex. Tablets were prepared by direct compression using 2^3 factorial design to evaluate the effect of formulation variables (Ac-di-sol concentration 5 or 10%, the ratio of soluble polymer 1:1 or 1:3 and binder type 6% w/w Avicel PH102 or 6% w/w carboxymethyl cellulose) on release characteristics. Results showed that lyophilized ODTs disintegrated within few seconds and had significantly faster dissolution rate (70-100 % in 5 minutes) compared to the commercial oral tablet (Cavinton®). This was achieved at high content of PEG 4000 or 2 HP-β-CD in presence of 10 % w/w Ac-Di-Sol and 6 % w/w Avicel PH102. The extent of per oral absorption of vinpocetine was determined in healthy human volunteers using randomized crossover design. The relative bioavailability of selected solid dispersion and inclusion complex formulations were found to be 171.98 % and 196.06 % respectively. The study indicated that complexation of vinpocetine with 2-HP-βCD or dispersion in PEG 4000 followed by lyophilization are two successful strategies for enhancing the bioavailability of the drug from ODTs.

Keywords: Bioavailability; Inclusion complex; Lyophilization; Orodispersible tablets; Solid dispersion; Vinpocetine

Introduction

Vinpocetine is a neurotropic agent [1]. which improves cerebral metabolism, increases ATP concentration, reduces blood clotting, and a powerful antioxidant [2]. It is superior to other preparations used to treat cerebral vascular disorders in that its effect is selective. It increases the cerebral blood flow to ischemic areas in patients with cerebrovascular disease [3,4]. The very poor aqueous solubility of vinpocetine gives rise to difficulties in preparing oral pharmaceutical formulation or injectable solution and results in variable oral bioavailability. In this study, ODTs containing vinpocetine were prepared by direct compression. Solid dispersion which is one of the methods that widely and successfully applied to improve the solubility, dissolution and consequently the bioavailability of poorly soluble drugs was applied. The solid dispersion is based on the concept that the drug is dispersed in an inert water-soluble carrier at solid state. Several water soluble carriers such as methyl cellulose, polyvinyl pyrrolidone and polyethylene glycols 4000 and 6000 are used as carriers for solid dispersion. Cyclodextrins and their derivatives have been frequently used to enhance the bioavailability by increasing the drug solubility, dissolution and/or permeability [5]. They also act as penetration enhancers by increasing the drug availability at the surface of biological barrier [6]. Vinpocetine is exposed to extensive first-pass metabolism to the inactive apovincaminic acid [7]. Therefore ODTs of vinpocetine are designed to increase drug absorption from oral mucosa and send the drug directly to systemic circulation bypassing the first pass metabolism of the liver. This will result in increased fraction of bioavailable drug and also result in a rapid onset of action via a more convenient and comfortable delivery route to the patients, especially elderly and those with swelling difficulties.

Materials and methods

Materials

Vinpocetine was generously gifted by Memphis pharmaceutical company, Egypt. 2-Hydroxy propyl- ß-cyclodextrin was purchased from the Chemical Co., Milwaukee, WI, USA. Polyethylene glycols 4000, Carboxymethylcellulose (CMC) and Pluronic F127 were

purchased from Fluka AG Buchs SG, Switzerland. Avicel PH 102 (microcrystalline cellulose) was obtained from FMC Corporation, Pennsylvania, USA. Magnesium stearate, aspartame, menthol and granular mannitol were purchased from El Nasr Chemical and Pharmaceutical Company, Egypt.

Preparation of solid dispersion using lyophilization technique

Freeze drying involved dispersion of appropriate quantities of vinpocetine and PEG4000 (water soluble polymer) at ratios of 1:1 or 1:3 w/w in 1.5 % w/v tartaric acid solution (used as cryoprotectant). The mixture was mixed by stirring to obtain clear solution. The clear monophase solution was frozen at -20 C then exposed to freeze drying for 24 hours using Novalyphe freeze drier (Novalyphe-NL 500, Savant instruments, Halbrook, NY, USA) The resultant freeze dried particles were kept over anhydrous calcium chloride in a dessicator for further investigations [8]. The inclusion complex was prepared by freeze-drying technique of the drug and 2-HP-β-CD at molar ratio (1:1) or (1:3) using the same method explained above.

Characterization of prepared solid dispersion

Differential scanning calorimetry (DSC)

DSC study was performed for vinpocetine powder, PEG 4000, vinpocetine-PEG4000 physical mixture (1:1) and the prepared solid dispersion. Samples (3-4 mg) were hermetically sealed in a flat bottomed aluminum pans and heated in the DSC instrument in an atmosphere of nitrogen to eliminate the oxidative and pyrrolytic effects. A temperature range of 25 to 300 C was used and the heating rate was 10 C/min. The DSC of the inclusion complex was also determined using DSC-50 instrument (Schimadzu, Japan).

Fourier transform-infrared (FT-IR) spectroscopy

The FTIR spectra of the pure drug, drug- PEG4000 physical mixture (1:1) and the prepared solid mixture as well as the inclusion complex were recorded using a Bruker FTIR spectrometer (Thermoscientific, Germany) according to the KBr disc technique. The FTIR measurements were performed in the scanning range of 4000 - 400 cm^{-1} at ambient temperature.

Preparation of ODTs

ODTs were prepared by direct compression technique using a single punch tablet press (Model TDP, Shanghai Tianhe China) [9]. A full factorial design (2^3) was applied for the screening study in which three factors were used at two levels. These factors were the concentration of super disintegrant either 5% or 10% w/w, the ratio of water soluble carrier for solid dispersion (PEG 4000) to vinpocetine either 1:1 or 1: 3 (w/w) and the type of binder (Avicel PH102 or CMC at a concentration of 6 %w/w). A combination of pluronic127 (2%w/w) as surfactant to increase the wettability of the particles, magnesium stearate (0.5% % w/w) as lubricant, aspartame (1% w/w) as sweetening agent, and menthol as flavoring agent (0.5% w/w) were used for the preparation of ODTs as shown in the Table 1. All the ingredients were weighed and mixed in a mortar and pestle then magnesium stearate was added. The blended material was compressed using flat–faced punch (8 mm). Vinpocetine ODTs which contain vinpocetine in the form of inclusion complex with 2-HP-β-CD were prepared as mentioned above, only replacing the solid dispersion carrier of vinpocetine with vinpocetine/ 2-HP-β-CD inclusion complex in a molar ratio of 1:1 or 1:3 as shown in Table 2. All other additives were added with the same concentration.

Pharmaceutical evaluation of the prepared fast disintegrating sublingual tables

Weight variation was evaluated using twenty tablets from each formulation selected at random and weighed individually. Average weight was calculated and the individual weights were compared with the average weight. The weight of not more than two tablets must not deviate from the average weight by more than 5% [10]. Percentage friability was performed according to British Pharmacopoeia by accurately weighing ten tablets from each formulation. Then, the tablets were placed in the friabilator drum which rotates at 25 rpm for a period of 4 minutes. Subsequently, the tablets were brushed and reweighed. The percentage loss in weight was calculated and taken as a measure of friability. Tablet hardness was evaluated using tablet hardness tester (Dr Schleunger Model 6D, Germany). Ten tablets from each formulation were examined and the mean hardness value was calculated and expressed in Kilograms [11]. The uniformity of vinpocetine content in different tablets was determined by crushing ten tablets from each formulation and determining the content of each tablet individually. The weight of each tablet was dissolved in 100 ml of 0.1N HCl. The solution was then filtered, properly diluted and the absorbance was spectrophotometrically measured at 314.8 nm and then vinpocetine content of each tablet was calculated (see Table 2).

In-vitro disintegration time

This test was carried out using six tablets each inserted in each of the six cells of the disintegrator, USP Disintegration tester (Hanson research, USA). Simulated saliva fluid (SSF) pH= 6.75 kept at 37 ± 1 C was used as the disintegration medium and the basket was raised and lowered at a constant frequency of 30 ± 2 cycles/min. The test results were presented as the average of six determinations. For each formulation the total time of disintegration was measured [12,13].

In-vivo disintegration time

The in vivo disintegration time of the prepared ODTs was evaluated in six human volunteers according to code of ethics used to treat human volunteers adopted by Beni Suief University. Each of the six subjects was given a coded tablet. Tablets were placed on the tongue and immediately the time for disintegration was

recorded. The subjects were asked to spit out the content of the oral cavity after tablet disintegration and rinse their mouth with distilled water. The swallowing of saliva was prohibited during the test, and saliva was rinsed from the mouth after each measurement. The test results are presented as mean value ± standard deviation figure.

Table 1: Composition of vinpocetine ODTs prepared by solid dispersion

Ingredients	F1	F2	F3	F4	F5	F6	F7	F8
Solid dispersion*	1:1	1:1	1:1	1:1	1:3	1:3	1:3	1:3
Ac-di-sol	5%	5%	10%	10%	5%	5%	10%	10%
CMC	6%	-	6%	-	6%	-	6%	-
Avicel PH102	-	6%	-	6%	-	6%	-	6%
Mint flavor	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%
Aspartame	1%	1%	1%	1%	1%	1%	1%	1%
Magnesium stearate	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%
Pluronic 127	2%	2%	2%	2%	2%	2%	2%	2%
Granular Mannitol	Quantity sufficient to make final weight of 200 mg							

Solid dispersion*: contain 5 mg vinpocetine and PEG 4000 (1:1) and (1:3) w/w.

Table 2: Composition of vinpocetine ODTs prepared by inclusion complexation

Ingredients	F9	F10	F11	F12	F13	F14	F15	F16
Inclusion complex*	1:1	1:1	1:1	1:1	1:3	1:3	1:3	1:3
Ac-di-sol	5%	5%	10%	10%	5%	5%	10%	10%
CMC	6%	-	6%	-	6%	-	6%	-
Avicel PH102	-	6%	-	6%	-	6%	-	6%
Mint flavor	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%
Aspartame	1%	1%	1%	1%	1%	1%	1%	1%
Magnesium stearate	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%	0.5%
Pluronic 127	2%	2%	2%	2%	2%	2%	2%	2%
Granular Mannitol	Quantity sufficient to make final weight of 200 mg							

Inclusion complex *: contain 5 mg vinpocetine and 2HP-B-CD of molar ratio (1:1) or (1:3).

In vitro dissolution of vinpocetine ODTs tablets

The dissolution of vinpocetine from its tablets was performed in 200 ml phosphate buffer (pH 6.8), maintained at a temperature of 37 ± 0.5 C using the USP Dissolution Tester, Apparatus I (Hansson research, USA) at rotation speed of 50 rpm. The dissolution test was done by placing a tablet from each formulation in the dissolution stainless steel basket. Aliquots from the

dissolution medium were withdrawn after certain time intervals. The withdrawn samples were filtered through Millipore filter membrane of 0.45 μm pore size and analyzed for vinpocetine content by measuring their absorbance at 314.8 nm using a UV-spectrophotometer (Jasco-V 530, Japan).

In-vivo studies

The studies were carried out to compare the pharmacokinetics of vinpocetine from formulation (F8 treatment A) and formulation (F16 treatment B) in comparison to the conventional Cavinton ® tablets (ACAPI , Egypt) and labeled as treatment C. A single dose of vinpocetine (10mg) was given to the volunteers using randomized- single dose three-way (open-label study) randomized crossover design (Table 3). Six healthy man volunteers aged between 20 to 40 years (median weight: 75 kg and median height: 183 cm) were chosen. Health status of the volunteers was confirmed by complete medical history, physical examination and laboratory analysis for complete hematological and biochemical examination, all these were carried out at baseline. None of the volunteers had any history of drug or alcohol abuse, nor did they have any acute or chronic gastrointestinal, cardiac, vascular, hepatic or renal disease. The protocol of the study was conducted according to Helsinky agreement protocol and according to the requirements of the ethical committee of the faculty of medicine, Cairo University, Egypt. The drug was administered orally after fasting overnight and washout period of 1 week. Venous blood samples (5 mL) were collected into heparinized tubes at the following time intervals: 0, 0.5, 1, 1.5, 2, 3, 4, 6, 8, and 10 hours after administration of a treatment. Plasma was obtained by centrifugation at 2000 rpm (using centrifuge R32 , Bombay, India) for 10 minutes and stored at -20ºC until the time of analysis.

Table 3: *In-vivo* randomization plan for two vinpocetine formulations F8 and F16 ODTs compared to the conventional Cavinton® (10 mg) tablets.

Volunteer Number	Phase I	Phase II	Phase III
1	F8	F16	Cavinton
2	F16	F8	Cavinton
3	Cavinton	F8	F16
4	F8	Cavinton	F16
5	F16	Cavinton	F8
6	Cavinton	F16	F8

Chromatographic conditions

A modified HPLC method for determination of vinpocetine in plasma was adopted [14]. The HPLC apparatus consisted of isocratic pump LC-10 AS and a UV/VIS detector SPD-10A connected to a C-R6A integrator (Shimadzu, Koyoto, Japan). The analytical column was Ponapak C18 HPLC column (4.6 250 I.D mm), particle size 125 ºA (Waters Associates, Ireland). The mobile phase composed of methanol: water in ratio of 80:20 (v/v), containing 0.1% w/w triethylamine, and pH 7 adjusted with glacial acetic acid. The flow rate was 2 ml/min. The detection wavelength was set at 274 nm. All assays were performed at ambient conditions.

Plasma analysis

A calibration curve of vinpocetine in plasma was constructed in the concentration range (5-20 ng/ml). Then one mL sample of the plasma was mixed with acetonitrile (1 mL) and stock solution of the internal standard (ketotifen fumarate) (1 mL). The mixture was vortexed for 1 minute and then centrifuged for 10 minutes at 3000 rpm. The upper layer was separated and transferred to another tube then filtered through 0.45 μm Millipore® filter for analysis with HPLC. Twenty microliters (μL) of the samples were injected to the column for analysis. Spectrophotometric detection at 274 nm was interpreted in the form of the reported peak areas. The recovery (20-600 ng/mL) varied between 97.58 and 100.23 %.

Pharmacokinetic analysis

Pharmacokinetic parameters were obtained from plasma data following administration of two treatments and control to each subject using WinNonlin® (version 1.5, Scientific consulting, Inc., Cary, NC, USA). Non-compartmental analysis was adopted for calculation of C_{max} (ng/mL) and t_{max} (h); the observed maximal drug concentration and the time needed to reach this concentration respectively. The relative bioavailability was calculated as (AUC_{test} / $AUC_{standard}$) x100.

Statistical analysis

An analysis of variance (ANOVA) test was performed for untransformed data for the pharmacokinetic parameters C_{max}, t_{max}, AUC_{0-t}, and $t_{1/2}$ using the software SPSS 11.0 (SPSS Inc., Chicago, USA) at p-value (p 0.05).

Results and discussion

Differential scanning calorimetry (DSC)

The possible interaction between the drug and the soluble carrier (PEG) was studied by DSC (Figure. 1). Pure vinpocetine powder showed a melting endotherm at 152.8 C and the scan of PEG 4000 showed a broad endotherm at 62.5 C. It was clear that the sharp endothermic peak of the drug, became shorter in the thermograms of the physical mixtures which may be attributed to the reduced purity of samples after mixing[15]. The DSC scan of the solid dispersion shows the absence of a drug peak as shown in

Figure.1d, suggesting that vinpocetine was molecularly dispersed and in an amorphous form or is present as a solid solution inside PEG 4000 matrix[16]. DSC thermograms of pure vinpocetine, (vinpocetine/ HP-ßCD), 1:1 physical mixtures and the inclusion complex are shown in Figure.2. The thermogram for HP-ßCD, demonstrated an endothermic peak around 60 ºC corresponding to water loss. The characteristic peak of the drug totally disappeared in the complex thermogram. This result confirmed complete inclusion between drug and the cyclodextrin. Similar effect was reported by Liu and Zhu [17] for inclusion complexes of prazocin hydrochloride with HP-β-CD.

Fourier-Transform infrared spectroscopy

Analysis by FTIR spectroscopy was carried out to assess any possible interaction between drug and PEG 4000. FTIR spectra of the drug, PEG 4000, physical mixture and the solid dispersion are shown in Figure. 3. The spectrum of pure vinpocetine (Figure. 3a) shows its characteristic peaks at 1716.34 cm^{-1} which has the characteristic shoulder of carbonyl stretching band (C=O) present in vinpocetine and 3000-2840 cm^{-1} assigned to aromatic stretching. FTIR spectra of PEG 4000 (Figure. 3b) shows the characteristic peaks at 2950–2750 cm^{-1}(C–H stretch), and 1468.84 cm^{-1} for (C–O stretching). FTIR spectra of physical mixture and solid dispersion (Figures. 3c and 3d) show no substantial shifting of the position of the functional groups. The peaks are only broadened, indicating no major interaction between vinpocetine and hydrophilic carriers[18]. FTIR spectroscopy of inclusion complex was used to confirm the interaction between vinpocetine and HP-ß-CD. The changes or shifts in the drug absorption spectrum occur upon complexation [19]. The IR spectra of vinpocetine, (vinpocetine/2HP-ßCD), 1:1 physical mixtures and the inclusion complex, are illustrated in Figure. 4. The spectra of HP-ß-CD illustrated an intense broad absorption bands at 3500-3300 cm^{-1} corresponding to the free –OH stretching vibration [20], the vibration of –CH and –CH2 groups (C-H aliphatic stretching) appeared in the region 2950-2600 cm^{-1}. A shorter band appeared in the region 1650-1640 cm^{-1}, that could be ascribed to the hydrated bonds within cyclodextrin molecules. The spectra of the physical mixtures were the superposition of pure components spectra indicating the absence of interaction between vinpocetine and 2HP-ßCD in the physical mixture. This result was in good agreement with the work of Fernandes [21]. On the other hand, the IR spectra of (vinpocetine/ 2HP-ßCD) inclusion complex showed considerable differences when compared with those of the physical mixtures as shown in Figure. 4d. This can be explained by the dissociation of the intermolecular hydrogen bonds associated with crystalline drug molecules. The broadening and decrease in the intensity of the drug aromatic stretching band observed in these systems, might be due to its restriction within the cyclodextrin cavity [22].

The weight uniformity test showed that none of the tablets deviated by more than 5 %, indicating that all formulations fulfills the pharmacopoeial limits for weight variation.

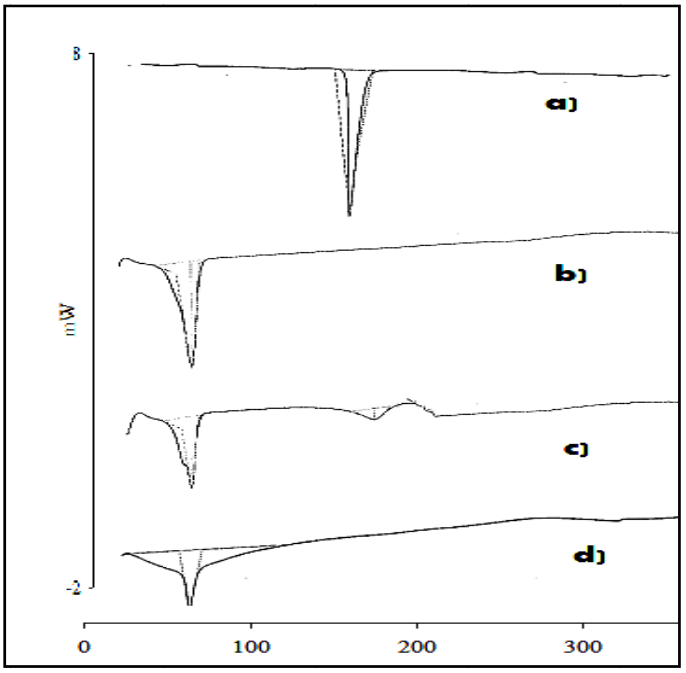

Figure. 1: DSC of solid dispersion systems; (a) Pure vinpocetine ; b) Pure PEG4000 c) Physical mixture and d) Solid dispersion

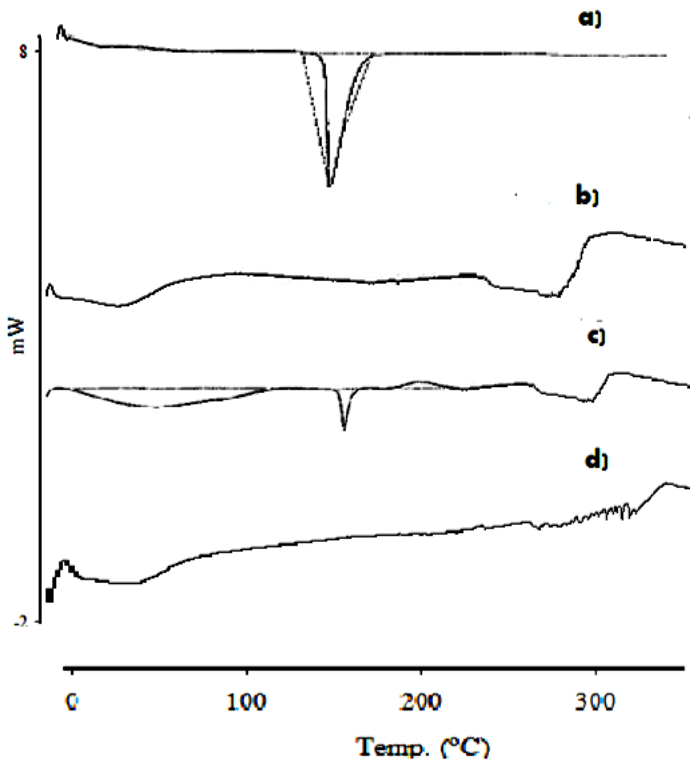

Figure.2: DSC thermograms of vinpocetine/2-Hydroxyypropyl-β-cyclodextrin solid systems. a) Pure vinpocetine; b) Pure HP-β-CD; c) Physical mixture and d) Inclusion complex

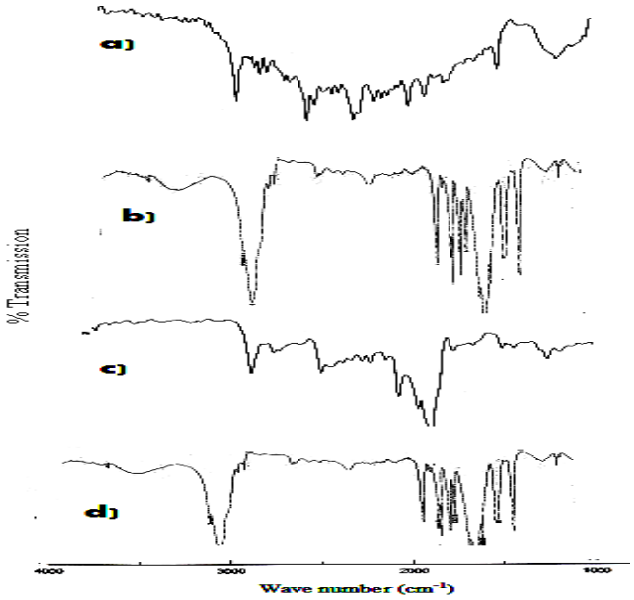

Figure.3: FTIR patterns of solid dispersion system; a) Pure vinpocetine, b) PEG4000;
c) Physical mixture and d) Solid dispersion

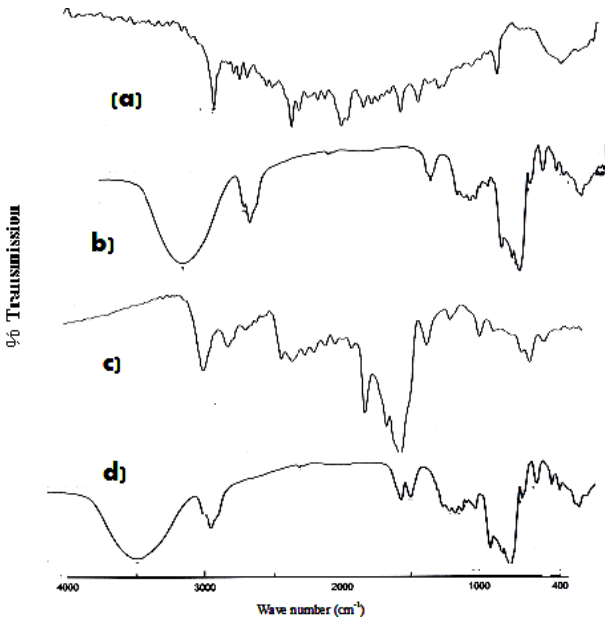

Figure.4: FTIR patterns of vinpocetine-2HP β-cyclodextrin solid systems. a) Pure vinpocetine; b) Pure HP-β-CD; c) Physical mixture and d) Inclusion complex

Pharmaceutical characterization of vinpocetine ODTs tablets

All formulations were evaluated for various physical parameters which were reported in Table 3 and 4. All the tablets maintained hardness in the range of 5.11 – 8.55 kg/cm^2. The percentage friability was in the range of 0.31-0.89 %. Drug content calculated for different formulations was highly uniform and was found to be more than 95 % from all formulations.

In-vitro and in-vivo disintegration time

The in-vitro disintegration study showed that ODTs containing 6% w/w CMC with 5 % w/w Ac-di-sol (F1 and F9) showed longer disintegration time (more than 3 minutes) compared to ODTs containing 6% w/w Avicel PH102 with 5 % w/w (F2 and F9), which may be attributed to disintegration properties of Avicel PH102. The formulations F4, F8, F12 and F16 possessed the lowest disintegration time due to the presence of the high concentration of Ac-di-sol (10 % w/w) with Avicel PH102 (6 % w/w). It was also noticed that in-vivo disintegration time values were shorter than the corresponding in-vitro disintegration times for all formulation.

In-vitro dissolution studies

The pH of the mouth saliva ranges between 5.6 and 7.6 units. Therefore, in the dissolution studies, simulated saliva fluid (SSF) that has a pH of 6.8 ± 0.5 was adopted as a dissolution medium [3, 23]. The cumulative vinpocetine dissolved as a function of time from ODTs compared to the marketed product (Cavinton®) are illustrated in Figures. 5 and 6. Remarkable differences in the dissolution profiles of the prepared ODTs and those of the commercial drugs are observed. Figure 5 illustrates the dissolution profile of vinpocetine from the ODTs prepared by solid dispersion technique. It is obvious that the tablets prepared according to 1:3 w/w drug to polymer exhibit higher dissolution rate than 1:1 drug to polymer. F8 exhibited the highest extent of drug dissolution compared with the other tested tablets of different formulations. The dissolution of the marketed vinpocetine formulation was only 20 % in 30 minutes. This could be due to the high hydrophobic nature of the drug and poor wettability. The improvement in the dissolution rate of the solid dispersion system may be attributed to the decrease in degree of crystallinity of the drug due to lyophilized solid dispersion and the surface acting properties of the carrier which together contributed to the increased solubility and wetability of the drug [24]. The dissolution profile of the binary system formulation of the drug with HP-β-CD illustrated in Figure. 6 showed that the rate and extent of drug release was increased as the 2HP-β-CD ratio increased. This finding agrees with that mentioned by Ribeiro et al. [17] who attributed this improvement in drug dissolution to the solubilization action of cyclodextrin operating locally in the aqueous hydrodynamic layer surrounding the drug particles [25]. Formulations F8 and F16 demonstrated the highest dissolution rates after 5 minutes so they were selected for the in-vivo study.

Table 4: Pharmaceutical evaluation data of the prepared vinpocetine ODTs prepared by solid dispersion

Formula Number	Average Weight (mg) ± S.D*	% Friability	Hardness (Kg ± S.D.*)	*In-vivo* Disintegration Time ±S.D.* (sec.)	*In-vitro* Disintegration Time ±S.D.* (*Sec.*)	% Content Uniformity
F1	103.36 ±0.12	0.316	7.81 ± 0.720	> 3min	> 3min	102.5±0.93
F2	99.13 ±0.09	0.471	7.52 ± 0.811	8711±6.92	116.17±9.91	94.31 ±2.8
F3	100.33 ±0.07	0.708	7.07 ± 0.613	44.6±5.36	55.15±6.29	112.01±0.99
F4	101.69 ±0.19	0.625	8.55± 0.441	37.15±6.20	42.39±6.47	99.03 ±1.95
F5	104.77 ±0.049	0.870	6.99± 0.860	98.6±4.82	106.50±10.73	105.81±2.0
F6	102.29 ±0.39	0.711	6.37 ± 0.815	61.71±4.76	74.15±6.52	92.94 ±1.24
F7	100.75±0.24	0.835	5.73 ± 0.449	34.79±3.76	40.6±4.25	90.48 ±1.49
F8	103.44±0.39	0.791	5.51 ± 0.742	27.68 ±1.55	31.71±5.75	103.05±2.75

Table 5: Evaluation data of the prepared vinpocetine ODTs prepared by inclusion complexation

Formula Number	Average Weight (mg) ± S.D*	% Friability	Hardness (Kg ± S.D.*	*In-vivo* D*isintegration Time* ± S.D.* (sec.)	*In-Vitro* Disintegration Time ± S.D.* (*Sec.*)	% Content Uniformity
F9	100.10±0.53	0.795	7.11 ± 0.130	> 3min	> 3min	104.6 ±0.81
F10	102.54±1.15	0.405	6.01 ± 0.328	83.11±3.02	96.17±3.11	98.61±1.472
F11	99.18±0.99	0.779	6.87 ± 0.453	64.6±5.36	71.15±6.20	102.71±2.01
F12	101.58±1.37	0.921	6.11± 0.157	29.19±2.89	33.27±6.38	96.25 ±0.92
F13	99.91±1.15	0.779	7.93± 0.536	89.27±4.14	112.49±6.59	103.62±2.57
F14	98.18±2165	0.891	7.89 ± 0.079	70.48±8.50	84.558±4.12	101.15±0.98
F15	102.47±0.93	0.55	4.87 ± 0.421	37.25±4.82	44.95±5.28	98.34±1.49
F16	99.33±1.97	0.611	5.11 ± 0.130	18.37±5.43	23.20±6.615	101.55±3.11

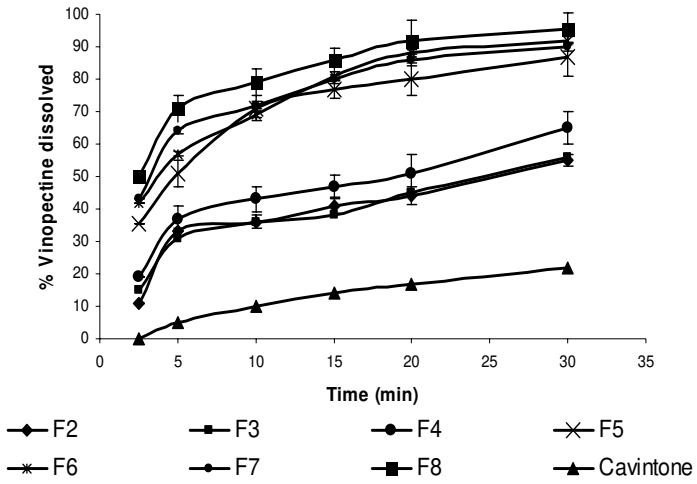

Figure.5: In-vitro dissolution profiles of vinpocetine from ODTs prepared by solid dispersion in phosphate buffer pH 6.8 at 37 ± 0.5ºC

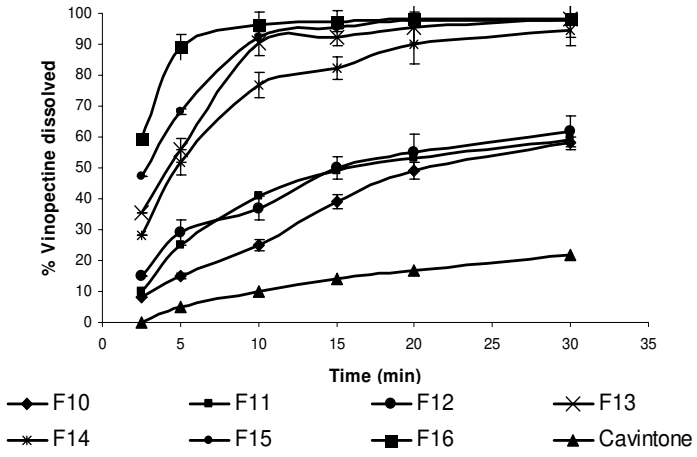

Figure.6: In-vitro dissolution profiles of vinpocetine from ODTs prepared by inclusion complex in phosphate buffer pH 6.8 at 37 ± 0.5ºC

In-vivo absorption studies

The mean plasma concentration–time courses from vinpocetine following per oral administration of ODT F8, F16 and Cavinton are illustrated in Figure. 7. All the pharmacokinetic parameters were evaluated using WinNonLin® Professional. Vinpocetine was detected in plasma immediately after 15 minutes from administration of the ODT in six subjects. This rapid onset indicated superiority of absorption from per oral route using ODTs. Conversely, vinpocetine appeared in plasma after 30 minutes for subjects received the regular oral marketed Cavinton® tablets. The mean C_{max} estimated from F8 and F16 were 34.41 ± 8.24 ng/mL and 39.18 ± 8.7130 ng/mL respectivel,while it was 25.20 ± 8.09 ng/mL for Cavinton® tablets. The differences between the three treatments for C_{max} appeared to be statistically significant ($p < 0.05$). The mean AUC_{0-t} estimated from ODTs which reflects the total amount of drug absorbed over the 10 hrs time period, t_{max} and $t_{1/2}$ were determined (see Table 6). The relative bioavailability of F16 was 196.04 compared to 171.37% for F8 when Cavinton® tablets were taken as the reference standard.

The higher C_{max}, faster t_{max} and the improved bioavailability observed for F8 and F16 than the commercially available conventional tablet (Cavinton®) may be attributed to rapid disintegration and dissolution of the drug in saliva even with the absence of water. Moreover, in case of ODTs, the drug partially absorbed through the mucosal membrane and consequently the bioavailability was increased [26]. Furthermore, the rapid absorption of vinpocetine from the buccal mucosa, pharynx and esophagus as the saliva passes down into the stomach (pregastric absorption) led to a decreased pre-systemic biotransformation [27]. The increased relative bioavailability of F16 compared F8 might be due to the effect of 2-HP-βCD incorporated product which have the ability to interact with macromolecules of mucosal membrane more efficiently causing marked improvement in drug absorption [28, 29].

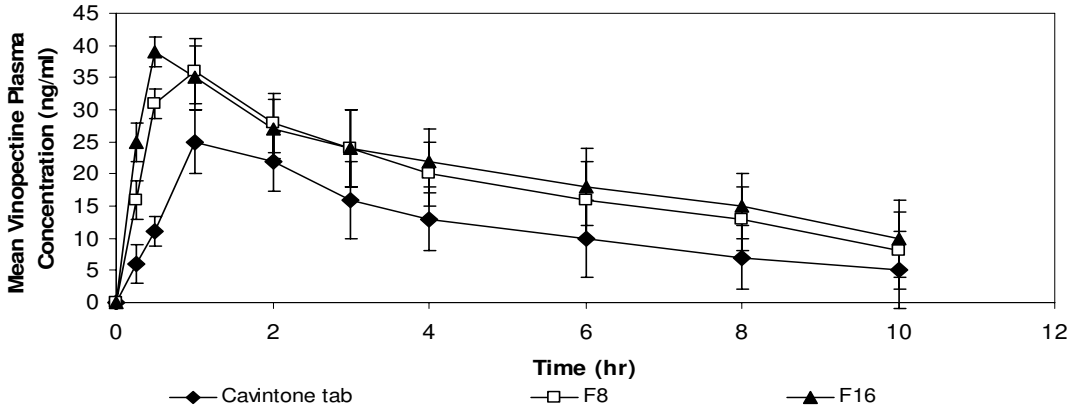

Figure.7: Plasma concentration of vinpocetine following oral administration of Cavinton ® tablets and per oral administration of F8 and F16 ODTs.

The ability of 2-HP-β CD to augment the extent and stability of supersaturated solutions of various poorly water soluble drug candidates with consequent improvement of their oral bioavailability has been assessed by Vandecruys et al. [30]. An additional factor that could contribute to the improved oral absorption and enhanced bioavailability of drugs by hydrophilic cyclodextrins is the increased permeation of drug molecules across the membrane lipid bilayers, as a result of their enhanced availability at the biological barrier and the alteration of the membrane fluidity [31-33].

Table 6: Pharmacokinetic parameters of vinpocetine following oral administration of a single dose (10 mg) of marketed formulation (Cavinton® tablets) and selected ODTs formulation (F8) and (F16)

Pharmacokinetic parameter	Cavinton® tablets	Orodispersible tablets (F8)	Orodispersible tablets (F16)
C_{max} (ng/ ml) ± SD	25.20 ± 8.09	34.41 ± 8.24	39.18 ± 8.71
t_{max} (hour) ± SD	1.47 ± 0.49	1.05 ± 0.38	0.83 ± 0.29
$AUC_{(0-t)}$ (ng.hr/ ml) ± SD	249.43 ± 14.67	428.71 ± 9.35	488.72 ± 12.82
$t_{1/2}$ (hour) ± SD	1.54 ± 0.68	1.85 ± 0.73	1.67 ± 0.64
%Relative bioavailability	---	171.98 %	196.04 %

Conclusion

From the present study it can be easily demonstrated that, orally rapid disintegrating tablets of vinpocetine prepared by freeze-drying technique is considered a successful formulation methodology. This method resulted in improved water solubility characteristics of vinpocetine and maximized its bioavailability compared to the marketed product. Vinpocetine ODTs prepared from inclusion complex with 2-HPβCD showed better results compared with those prepared by solid dispersion technique. Therefore; the study suggests that the developed ODT formulation F16 may be a better alternative to conventional oral formulation of vinpocetine with possibility of reduction of the dose.

References

[1]. Bereczki D, Fekete I. A systematic review of vinpocetine therapy in acute ischaemic stroke. Pharmacol. Eur. J. Clin., 1999(55): p. 349-352.

[2]. Feigin VL, Doronin BM, Popova TF, Gribatcheva EV, Tchernov DV. Vinpocetine treatment in acute ischaemic stoke: a pilot single-blind randomized. clinical trial. Eur. J., 2001(8): p. 81-85.

[3]. Chen JCJ, Tao W, Mei N, Cao S, Jiang X. Determination of apovincaminic acid in human plasma by high-performance liquid chromatography using solid-phase extraction and ultraviolet detection. J Chromatogr B., 2006. 830: p. 201–206.

[4]. Kiss B, Karpati E. Mechanism of action of vinpocetine. Acta. Pharm. Hung., 1996(66): p. 213-214.

[5]. Del Valle, E.M.M., Cyclodextrins and their uses. a review. Process Biochemistry, 2004. 39: p. 1033-1046.

[6]. Challa R, Ahuja A, Ali J, Khar RK. Cyclodextrins in drug delivery: an updated review. AAPS PharmSciTech , Article 34, 2005. 6(2): p. E329-E357.

[7]. Polgar M, Vereczkey, Nyary I. Pharmacokinetics of vinpocetine and its metabolite, apovincaminic acid, in plasma and cerebrospinal fluid after intravenous infusion. J. Pharm. Biomed. Anal., 1985. 3: p. 131-139.

[8]. Gupta, S, Srivastav S, Vajpai M. Solid dispersion technique for improving solubility of some poorly soluble drugs. Journal of Pharmacy Research, 2012. 3(4): p. 574-1586.

[9]. Ali AA, Ali AMAH. Optimization of propranolol HCl release kinetics from press coated sustained release tablets. Pharmaceutical Development and Technology, 2012(00): p. 1-9.

[10]. Rudnic E, Schwartz JB , Oral Solid Dosage forms In: Remington's Pharmaceutical Sciences. 18th edition,1990, Mack publishing Company. Easton, Pennsylvania, USA. p. 633-1665.

[11]. The Biritich Pharmacopoia. 2001: Vol. I and II .London :The statiory office,. p.705-706,1810-1812

[12]. Kottke MK, Rudnic EM. Tablet Dosage Forms, in Modern Pharmaceutics, G.S. Banker, and Rhodes, C.T., Editor. 2002, Marcel Dekker Inc.: New York. p. 437-511.

[13]. Alderborn G. Tablets and compaction, in Pharmaceutics The Science of Dosage Form Design, M.E. Aulton, Editor. 2002,

Churchill Livingstone: Edinburgh. p. 397-439.

[14]. Abd Elbary A, Foda N, El-Gazayerly,O, El Khatib M. Reversed phase liquid chromatographic determination of vinpocetine in human plasma and its pharmacokinetic application. 2002. Anal. Lett. (35): p. 1041-1054.

[15]. Botha SA, Lötter AP. Compatibility studies between isosorbide mononitrate and selected excipients used in the development of extended release formulations, . Drug.Dev.Ind.Pharm, 1990. 16(4): p. 673.

[16]. Murrell W. Nitroglycerin as a remedy for angina pectoris. Vol.151. 1879: Lancet.

[17]. Liu L, Zhu S. Preparation and characterization of inclusion complexes of prazosin hydrochloride with beta-cyclodextrin and hydroxypropyl-beta-cyclodextrin. J Pharm Biomed Anal, 2006. 40(1): p. 122-7.

[18]. Ahuja N, Katare O P, Singh B. Studies on dissolution enhancement and mathematical modeling of drug release of a poorly water-soluble drug using watersoluble carriers. Eur. J. Pharm. Biopharm, 2007. 65(1): p. 26-38.

[19]. Ribeiro, LSS. Ferreira DC, Veiga FJB. Physicochemical investigation of the effects of water-soluble polymers on vinpocetine complexation with β-cyclodextirn and its sulfobytyl ether derivative in solution and solid state. . Eur. J. Pharm. Sci. , 2003. 20(3): p. 253-266.

[20]. Jun SW, Kim MS, Kim JS, Park HJ, Lee S, Woo JS, Hwang SJ. Preparation and characterization of simvastatin/hydroxypropyl-β-cyclodextrin inclusion complex using supercritical antisolvent (SAS) process. . Eur. J. Pharm. Biopharm, 2007. 66(3): p. 413-421.

[21]. Fernandes CM, Teresa VM, Veiga FJ. Physicochemical characterization and in vitro dissolution behavior of nicardipine-cyclodextrins inclusion compounds. Eur. J. Pharm. Sci. , 2002. 15(1): p. 79-88.

[22]. Ficarra R. et al. Study of the inclusion complex of atenolol with beta-cyclodextrins. J Pharm Biomed Anal, 2000. 23(1): p. 231-6.

[23]. Conine JW, Pikal MJ. Pharmacutical Dosage FormsTablets, ed. H.A. Liberman, Lachman, L.,Schwartz J.B. Vol. 1. 1989: New York :Marcel Dekker. p.329-340.

[24]. Farzana SB, Ibrahim S. Khattab , Lyophilized gliclazidepoloxamer solid dispersions for enhancement of invitrodissolution and invivo bioavailability. International Journal of Pharmacy and Pharmaceutical Sciences, 2011. 3(2).

[25]. Ribeiroa L, Ferreirab DC, Veiga FJ. In vitro controlled release of vinpocetine-cyclodextrin-tartaric acid multicomponent complexes from HPMC swellable tablets. J Control Rel, 2005. 103: p. 325-339.

[26]. Jacobsen J, Christrup L, Jensen N. Medicated chewing gum: pros and cons Am J Drug Delivery, 2004. 2(2): p. 75-88.

[27]. Seager H. Drug-delivery products and the Zydis fast-dissolving dosage form. J Pharm Pharmacol,, 1998. 50(4): p. 375-382.

[28]. Loftsson T, Jarho P, Masson M, Jarvinen T. Cyclodextrins in drug delivery. Exp. Opinion Drug Del, 2005. 2: p. 335-351.

[29]. Senel S, Hincal AA. Drug permeation enhancement via. buccal route: Possibilities and limitations. J. Control. Rel., 2001. 72: p. 133-144.

[30]. Vandecruys R, Peeters J, Verreck G, Brewster ME. , Use of a screening method to determine excipients which optimize the extent and stability of supersaturated drug solutions and application of this system to solid formulation design. Int. J. Pharm., 2007. 342(1-2): p. 168-175.

[31]. Yoo SD, Yoon BM, Lee HS, Lee KC. Increased bioavailability of clomipramine after sublingual administration in rats. J. Pharm. Sci. , 1999. 88(11): p. 1119-1121.

[32]. Udata C, Patel J, Pal D, Hejchman E, Cushman M, Mitra AK. Enhanced transport of a novel anti-HIV agent--cosalane and its congeners across human intestinal epithelial (Caco-2) cell monolayers. Int. J. Pharm. , 2003. 250(1): p. 157-168.

[33]. Martins PS, Ochoa R, Pimenta AM, Ferreira LA, Melo AL, da Silva JB, Sinisterra RD, Demicheli C, Frézard F. Mode of action of beta-cyclodextrin as an absorption enhancer of the water-soluble drug meglumine antimoniate. Int. J. Pharm., 2006. 325((1-2)): p. 39-47.