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RESEARCH ARTICLE

Preparation and In-Vitro, Ex-Vivo Evaluation of Oral Dosage Form Containing Mirtazapine Nano suspension

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Abstract

The aim of this study is to formulate and evaluate mirtazapine nanoparticles using solvent anti solvent technology. Mirtazapine is practically insoluble in water which acts as antidepressant. It is prepared as nano particles in order to improve its solubility and dissolution rate. Mirtazapine nanoparticles were prepared using anti-solvent precipitation method using different stabilizers (poloxamer 188, poloxamer 407, TPGS and SLS) at drug: stabilizer ratio 1:1 and 1:2, alone and in combination. Mirtazapine nanoparticles were characterized by particle size analysis, percentage of drug entrapment efficiency, in vitro dissolution study, ex vivo study, Fourier transform infrared spectroscopy, differential scanning calorimetry and atomic force microscope. Freeze dried nanoparticles were compressed into tablets by direct compression method using different excipients which are microcrystalline cellulose PH 102, starch, and Mg stearate as diluent, binder, and lubricant; The results showed that the particle sizes were ranged from 272 to 691 nm at drug: stabilizer ratio 1:1 and from 146 to 572 nm at drug : stabilizer ratio 1:2. Stabilizers combination has good surface affinity and could form a substantial mechanical and thermodynamic barrier at the interface of dug molecule. As the concentration of stabilizer increases the particle size decreases at fixed drug concentration. % EE was ranged from 75.5% to 95.9%. On the other hand dissolution rate increasing as the particle surface area is increase due to reduction of particle size to the nano range.

Keywords: *Mirtazapine, Nanoparticles, Particle Size, Poloxamer.*

Introduction

Nano suspensions are colloidal dispersions of nanosized drug particles stabilized by surfactants. They can also be defined as a biphasic system consisting of pure drug particles dispersed in an aqueous vehicle in which the diameter of the suspended particle is less than 1µm in size [1]. Poor aqueous solubility of drugs is major limiting factor with many new drugs in their successful launch in market in spite of their potential pharmacokinetic activity [2].

Several formulation techniques exist for the manufacturing of nanosuspension, precipitation has been applied to prepare submicron particles, especially for the poorly soluble drugs. Rapid addition of a drug solution to the anti-solvent leads to sudden super saturation of drug and formation of ultrafine crystalline or amorphous drug solids [3].Mirtazapine is an antidepressant drug used for the treatment of moderate to severe depression, molecular formula: C17H19N3 [4].

The drug has bioavailability of 50 % due to first-pass metabolism, high protein binding (80%) and very high half-life (20-40 h) [5]. The aim of this study is to formulate and evaluate mirtazapine nanoparticles using solvent anti solvent technology.

Materials and Method

Materials

Mirtazapine powder was purchased from (Hyper-chem, China), TPGS (Hangzhou, Hyperchem, China -9flower, China), SLS (Fine-Chem limited Mumbai, India), poloxamer 188 and poloxamer 407 (HIMEDIA (Mumbai, India), methanol (Scharlau Chemie, S.A. Spain). All other chemicals were of analytical grade.

Preparation of Mirtazapine Nanosuspension

Nanosuspensions of mirtazapine were prepared by the solvent evaporation technique, which is also termed as anti-solvent precipitation method. Mirtazapine powder was dissolved in methanol (4 ml) at room temperature. This was poured into 20 ml of water containing different types of stabilizer (alone and in combination) maintained at room temperature and subsequently stirred at agitation speed of 500 revolution per minute (rpm) on magnetic stirrer for 60 min.to allow the volatile solvent to evaporated [6]. The resultant organic solution of drug (organic phase) was added drop by drop by means of a plastic syringe positioned with the needle directly into aqueous solution of stabilizer. The ratios of drug to stabilizer used to prepare the nanosuspension were 1:1 and 1:2, as shown in Table (1, 2).

Table 1: Composition of Mirtazapine Nanosuspension Using Different Stabilizers at Drug: Stabilizer Ratio 1:1.

Formula	Mirtazapine (mg)	Poloxamer188 (mg)	Poloxamer407 (mg)	TPGS (mg)	SLS (mg)	Methanol (ml)	Water (ml)
F1	15	15				4	20
F2	15		15			4	20
F3	15			15		4	20
F4	15				15	4	20
F5	15	7.5	7.5			4	20
F6	15	7.5		7.5		4	20
F7	15	7.5			7.5	4	20
F8	15		7.5	7.5		4	20
F9	15		7.5		7.5	4	20
F10	15			7.5	7.5	4	20

Table 2: Composition of Mirtazapine Nanosuspension Using Different Stabilizers at Drug: Stabilizer Ratio 1:2

Formula	Mirtazapine (mg)	Poloxamer188 (mg)	Poloxamer407 (mg)	TPGS (mg)	SLS (mg)	Methanol (ml)	Water (ml)
F11	15	30				4	20
F12	15		30			4	20
F13	15			30		4	20
F14	15				30	4	20
F15	15	15	15			4	20
F16	15	15		15		4	20
F17	15	15			15	4	20
F18	15		15	15		4	20
F19	15		15		15	4	20
F20	15			15	15	4	20

Evaluation of the Prepared Nanosuspension

Particle Size and Size Distribution

ABT-9000 USA particle size analyzer which is a dynamic light scattering works by measuring the intensity of light scattered by the molecules in the sample as a function of time, at scattering angle 90° and a constant temperature of 25 °C.

The polydispersity index (PDI) which is a measure of the width of the size distribution of each formula of LAF nanosuspension also determined, it is a measure of the distribution of particle size of nanoparticles obtained from a particle analyzer, PDI is an index of spread or variation or width within the particle size distribution. Also, the analyzer determines the specific surface area of each sample [7].

Determination of Drug Entrapment Efficiency of Nanosuspension

The freshly prepared nanosuspension was centrifuged at 6,000 rpm for 20 min. The supernatant solution was filtered and separated. 1 ml of this filtrate was diluted with water and the absorbance at maximum λ max was measured by UV spectrophotometer using water as blank. It was calculated by subtracting the amount of free drug in the supernatant from the initial amount of drug taken [8].

In Vitro Dissolution Profile of Nanosuspension

The dissolution was performed using dialysis membrane-60 (HIMEDIA) in 900 ml; it carried out by using USP type II apparatus. Samples of 5ml were withdrawn at predetermined intervals, and the samples were 5, 10, 15…120 min respectively), replaced with fresh dissolution medium. The samples were filtered through a 0.45μm membrane filter and diluted if necessary.

Absorbances of these solutions were measured at 315 nm using UV-visible spectrophotometer [9].

Ex- vivo Permeation Study

The study was approved by rate intestinal animals (duodenum) was isolated and washed with buffer. The non-everted tissue of 6 cm length was tied at one end and filled with 5 ml drug/formulation solution containing 15 mg drug and was tied at the other end, making the sac, injected with help of a tuberculin syringe and then tied. These segments were then tied onto the paddle of dissolution USP apparatus Type II and immersed in phosphate buffer (pH 6.8) medium.

The study was performed at 50 rpm. The transported drug from the absorption compartment was sampled (5 ml) with replacement phosphate buffer (pH 6.8) at 5, 10, 20, 30, 45, 60, 70, and 90 min and analyzed spectrophotometrically. The experiment was carried out in triplicate (n = 3) using fresh dissolution medium as well as fresh intestinal segment each time [10].

Freeze Drying of Nanosuspension

The principle of freeze drying method is that small amounts of a product will be frozen in and thereafter it will be placed under vacuum. The ice immediately changes into vapor, without to defrost first, also called sublimate. During process, the outside part will be the first part that dewaters.

After that, the water is removed closer and closer till the core of the product. Hereby the structure of the product stays intact. Due to the vacuum the ice will evaporate immediately without turning into water again [11].

Formation of Mirtazapine Nanoparticles Tablet

Mirtazapine formulated in to tablets by direct compression method containing drug equivalent to 15mg mirtazapine. All ingredients were properly mixed to gather then compressed in to tablets prepared by after freeze drying of formula (F15) that gave the best in vitro dissolution profile in minute in comparison with other nanoparticle formulas and pure drug, show as in Table (3) [12].

The amount of nanoparticle tablets were prepared using Avicel PH102 (MCC), starch and, magnesium stearate as a diluent, binder ,and lubricant at different concentration and tested to obtain the optimum formula that show the accepted hardness and the best in vitro dissolution profile [13].

Table 3: Composition of Mirtazapine Tablets

Materials	Quantity per tablet (mg)	
	F 15a	F 15b
Lyophilized Powder	45	45
Avicel PH 102 (MCC)	72	65
Starch	30	37
Magnesium Stearate	3	3
Tablet Weight (mg)	150	150

Precompression Studies of the Prepared Nanoparticle Powder

The flowability of a powder is of critical importance in the production of pharmaceutical dosage forms in order to get a uniform feed as well as reproducible filling of tablet dies otherwise high dose variations will occur. The powder flowability of prepared mirtazapine tablets were characterized by angle of repose, Hauser's ratio and carr's index [14].

Evaluation of Mirtazapine Nanoparticle Tablets

Tablets were evaluated for hardness test, friability test, content uniformity test and weight variation tests [15], and dissolution study.

Effect of Concentration of Avicel pH102 on the Dissolution of Tablet

F15a and F15b were prepared using Avicel pH102 as a diluent in different concentration to determine the effect of its concentration on the dissolution profile of the prepared tablets.

Effect of Addition Starch on the Dissolution of Tablet

F15a and F15b were prepared using starch as a binder and drug carrier in concentration at 20%, 25% respectively.

In Vitro Dissolution Profile of Mirtazapine Tablets

An in vitro dissolution test was conducted in a dissolution apparatus according to the USP paddle method. The temperature was maintained at 37 ± 0.5°C, and the stirring rate was at 50 rpm.

The commercial mirtazapine tablet accurately weighed bulk drug and nanosuspensions were dispersed in 900 ml of dissolution medium (0.1 N HCL). 5 ml samples were drawn, and the same volume of fresh dissolution medium was added at 5, 10, 15, 20, 30, .120 min, respectively. Then, the samples were filtered through a 0.1-µm syringe filter immediately before dilution, when necessary. Drug content was determined with a UV spectrophotometer at 315 nm for 0.1 N HCL (pH 1.2) [16].

Differential Scanning Calorimetry (DSC)

DSC investigations were performed using DSC apparatus model DSC-6. Samples of about 5 mg of pure drug powder and selected formula are placed in an aluminum pan and the experiment was carried out under nitrogen atmosphere at a flow rate of 40 mL/min and scanning rate of 10°C/min in the range of 15-300°C [17].

Fourier Transforms Infrared Spectroscopy

The Fourier transform infrared spectroscopy (FT-IR) spectra were recorded for pure drug and optimized formulation using KBr pellet technique. The pellets were prepared using KBr hydraulic press under hydraulic pressure of 150 kg/cm^2. The spectra were scanned over 3600-400 cm-1 at ambient temperature with a resolution of 4 cm-1; using FT-IR 2500 apparatus and spectra were recorded [18].

Atomic Force Microscopy (AFM)

The AFM is capable of scanning the surfaces in controlled environmental conditions and is complementary to SEM imaging. The size and surface morphology of mirtazapine nanoparticle were confirmed by atomic force microscopy after drying of the formula. Samples were determined in tapping mode, exerting pyramidal cantilevers with Pt probes. All results were recorded under ambient laboratory condition and scanning frequency of 2Hz. Resonance frequency was 79.491 kHz and a constant force in the range 2.5-10Nm^{-1}, driving amplitude 334.6mv. Silicon chip was newly operated by peeling off its upper layer to Form the sample. Particle size, 3D-dimension graph and histogram of particle size distribution were obtained [19].

Statistical Analysis

The results of the experiments are given as a mean samples ± standard deviation (SD) and were analyzed according to t-test and one way analysis of variance (ANOVA) using Sigma Plot 11 software at which the results would be significant if $p<0.05$, highly significant if $p<0.01$ and the results would be non - significant if $p>0.05$.

Results and Discussion

Particle Size Analysis

The particle size of F1-F4 at drug: stabilizer ratio 1:1 was ranged from 429 - 691 nm measured by particle size analyzer (as shown in Table 4) while for F11-F14 at drug : stabilizer ratio 1:2 the particle size ranged from 379-572 nm as in (Table 4) using poloxamer 188 , poloxamer 407 as primary stabilizers. poloxamer stabilizer gave larger particle size in both ratios 1:1 and 1:2 drug : stabilizer in F1, F2, F11 and F12 while SLS gave smaller particle size in both ratios 1:1and 1:2 drug :stabilizer in F4 and F 14.

Polydispersity index is a parameter used to define the particle size distribution obtained from the particle size analyzer. Polydispersity index gives degree of particle size distribution at range from 0.032 to 0.341 at ratio 1:1 and 0.021 to 0.211 at ratio 1:2 depending on formulation variables. The formula F10 showed lowest PDI (0.029) at drug: stabilizer ratio 1:1 and 0.114 at drug: stabilizer ratio 1:2, as seen in (**Table 4**); that indicate good uniformity of nanoparticle size.

Uniformity of particle size is determined by polydispersity index values in which the low value means the best uniformity. The range of PDI values (0-0.05) means (monodisperse system), 0.05-0.08 (nearly monodisperse), 0.08-0.7 (mid-range polydispersity), and >0.7 (very polydisperse) [19].From the obtained results, one can conclude that the poloxamer 188 and poloxamer 407 are suitable as a primary stabilizer for nanoparticles because of poor adsorption and affinity of poloxamer to the drug molecules [20].

Effect of Combination of Two Polymers on the Size of Mirtazapine Nanoparticles

The particle size of (F5-F10) of drug: stabilizer ratio 1:1 was ranged from 320-647 nm (**Table 4)**, (F15- F20) of drug: stabilizer ratio 1:2 was ranged from 146-449 nm (**Table 5**). At ratio 1:1 drug : stabilizer large particle size gave in combination of TPGS and poloxamer 407 in F8 (647 nm), in formulation that has TPGS as a primary stabilizer as in F3 that has particle size 481 nm, the addition of stabilizer has a paradoxical effect on nanoparticle stabilization and in F2 that contain poloxamer 407 as stabilizer has particle size 444nm, while in combination of two stabilizer in F8 gave large particle size indicating poor stabilization, and their combination was not appropriate for mirtazapine nanosuspensions.

The addition of stabilizer is not always beneficial for particle size reductions, which makes interaction between the polymer and drugs and increase the particle size for some drugs [21]. Nanoparticles formulation generally requires addition of appropriate stabilizers to lower the free surface energy of the nanoparticles and prevent particle aggregation and/or particle growth. The high surface free energy of nanoparticles is readily lowered by lowering the solid-liquid interfacial tension upon addition of surfactants [22].

The formula F15 showed lowest PDI (0.021), as seen in (**Table** 2) at drug: stabilizer ratio 1:2, that indicate good uniformity of nanoparticle size. Uniformity of particle size is determined by polydispersity index values in which the low value means the best uniformity when used two stabilizers (poloxamer 188+ poloxamer 407).

Effect of the Type of Solvent on the Particle Size

F1 show effect of solvent on particle size when used of ethanol instead of methanol get particle size 431nm when used ethanol while when used methanol particle size 429 nm therefore ethanol cannot effect the formula particle size, Ethanol as solvent produces very uniform-sized nanoparticles on precipitation [23].

Effect of the Temperature on the Particle Size

F2 show effect of temperature on particle size of formula when increase temperature from 250C to 500C led to increase size of particle from 444nm to 635nm, an increase in temperature can alter the dynamic viscosity and the diffusion coefficient observed the effects of temperature on the stability of mirtazapine nano-suspensions. They found that, nano-suspensions stored at 40°C were unstable, compared to those stored at 25°C and 4°C [24].

Table 4: Particle Size, PDI and EE% of Formulas at Drug: Stabilizer Ratio 1:1

Formula	Stabilizers	Particles size	PDI	EE%
F1	Poloxamer 188	429	0.171	83.9
F2	Poloxamer407	444	0.214	83.3
F3	TPGS	481	0.228	86.2
F4	SLS	272	0.082	89.1
F5	Poloxamer188+Poloxamer407	342	0.178	89.3
F6	Poloxamer188+TPGS	429	0.242	77.6
F7	Poloxamer188+SLS	320	0.139	89.1
F8	Poloxamer407+TPGS	647	0.341	76.5
F9	Poloxamer407+SLS	376	0.115	85.1
F10	TPGS+SLS	386	0.032	86.7

Table 5: Particle Size, PDI and EE% of Formulas at Drug: Stabilizer Ratio 1:2.

Formula	Stabilizers	Particles size	PDI	EE%
F11	Poloxamer188	383	0.079	88.2
F12	Poloxamer407	401	0.192	93.7
F13	TPGS	394	0.025	79.6
F14	SLS	203	0.035	89.9
F15	Poloxamer188+Poloxamer407	146	0.021	95.9
F16	Poloxamer188+TPGS	402	0.211	87.1
F17	Poloxamer188+SLS	241	0.102	87.9
F18	Poloxamer407+TPGS	449	0.153	93.6
F19	Poloxamer407+SLS	273	0.133	89.8
F20	TPGS+SLS	205	0.096	86.3

Determination of Drug Entrapment Efficiency of Nanosuspension

The EE% of the formulations from 76.5%-95.9 % (Tables 4, 5) The drug entrapment efficiency of F15was high when compared to other formulations. Drug - entrapment efficiency addition of non-ionic stabilizers (combination of poloxamer 188 and poloxamer 407)with ratio of drug: stabilizer(1:2) with higher EE% (95.9) in F15 which could be due to decreased partitioning of mirtazapine into the outer aqueous phase and better dispersion obtained by adding a hydrophilic stabilizers [25].

Dissolution Study

The dissolution profile of the formulas that in nanosize were studied in 0.1N HCl, the results showed that the formula F15 that contain poloxamer 188 and poloxamer 407 stabilizers gave the best release in 20 min in comparison with other formulas and the formula shows a maximum cumulative percentage drug release of 100 % within 20 min. From the above result depending on the particle size and PDI will be the formula no. F15 which has the mean particle size (146 nm) and E E% (95.9).

The release of F15 was compared with the pure drug in media of 0.1N HCl and in phosphate buffer pH6.8 (Figure 3) the maximum cumulative percentage drug release reach to 99.9 % within 20 minutes. In solution (pH 6.8) release of F15 reach 94.8 % in 40 minutes. The obtained results are in good accordance with Noyes-Whitney equation which states that the increase in saturation solubility and the decrease in particle size lead to an increased dissolution rate [26].

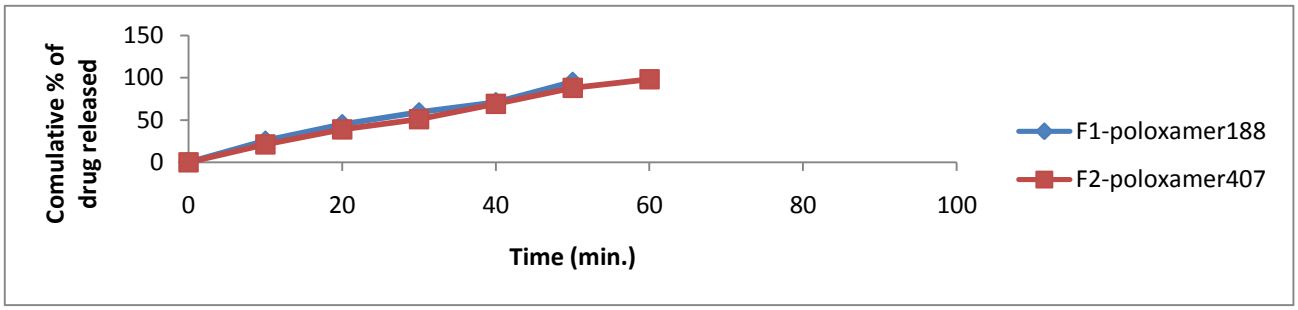

Figure 1: In vitro drug release profile of mirtazapine formulation nanosuspension in 0.1N HCl at 37°C (F1, F2)

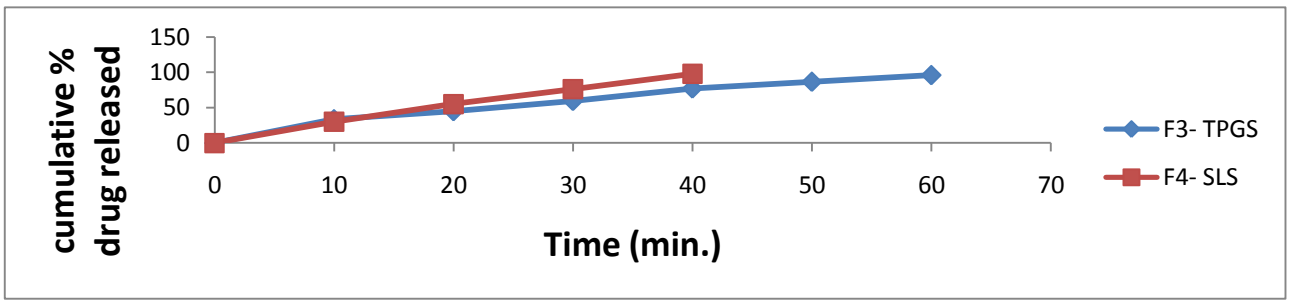

Figure 2: In vitro drug release profile of mirtazapine formulation nanosuspension in 0.1N HCl at 37°C (F3, F4)

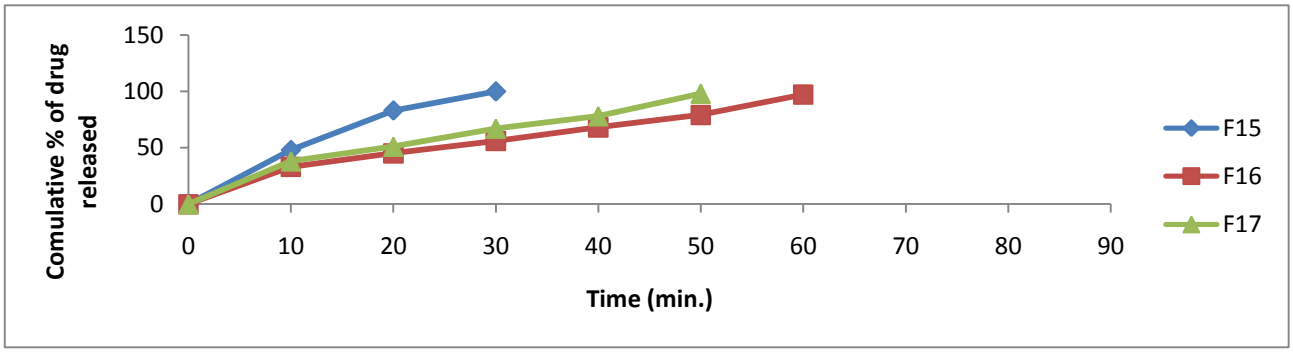

Figure 3: In vitro drug release profile of mirtazapine formulation nanosuspension in 0.1N HCl at 37°C (F15, F16, F17)

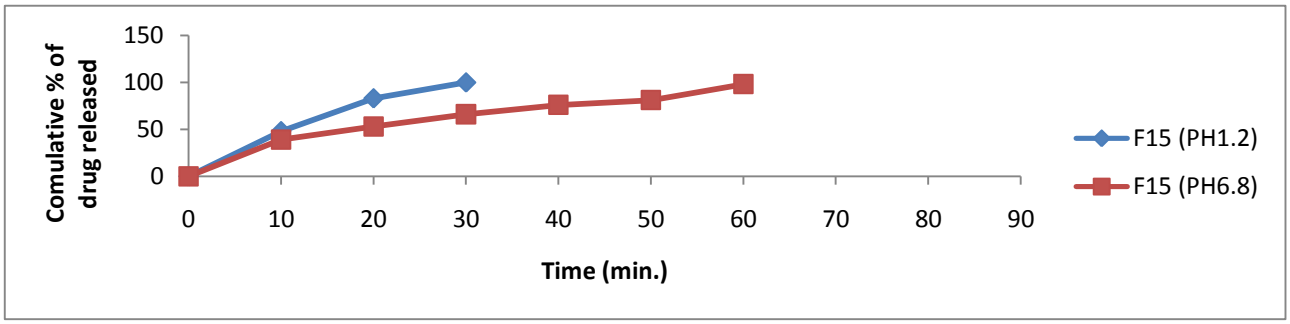

Figure 4: Dissolution profile of mirtazapine (F15) nanosuspension in 0.1N HCl (pH 1.2) and in phosphate buffer (pH6.8) at 37C°

Ex- Vivo Permeation Study

The absorption was estimated through the duodenum (proximal part) of the intestine, as mirtazapine mostly gets absorbed through the duodenum. The drug permeated across the intestinal wall and its concentration was measured in phosphate buffer solution.

The absorption of drug in nanosuspension (F15) was observed to be significantly increased as show in Figure (5). The observations suggest that the reduction in particle size leads to increase in the permeation, concentration of the drug particles and eventually improves the absorption of the drug [27].

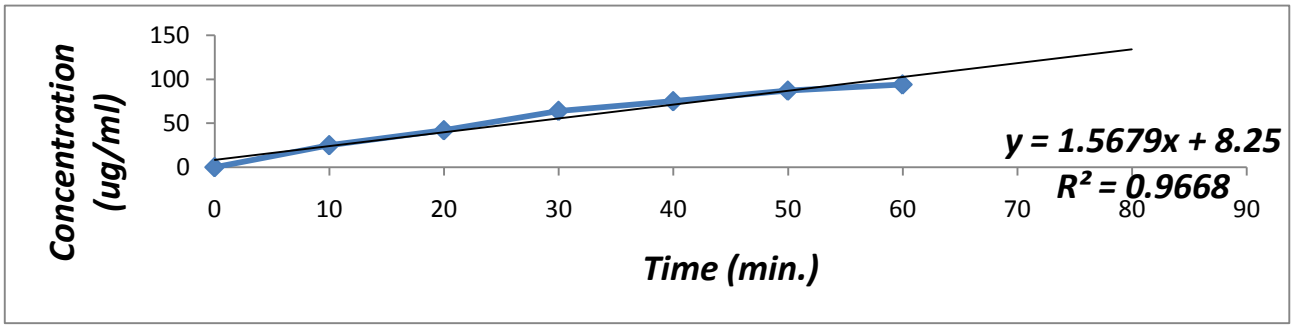

Figure 5: Ex – Vivo Absorption of Mirtazapine Nanosuspension through Rat Intestine

Drug Content in Lyophilized Powder

The drug content result of lyophilized powder of the selected formula (F 15) 97.64% of mirtazapine when determined by UV-visible spectrophotometer at λmax 315 nm.

Evaluation of Mirtazapine Nanoparticles Powder Flow ability

Angle of repose and compressibility index of the powder of the formulas (F15a and F15b) was reported in Table 6.

Table 6: Flow Properties of Prepared Blends of Mirtazapine Tablets Incorporating Drug Nanoparticles

Formula	*Angle of repose*	*Carr·s index*	*Hausner ratio*	*Physical*	*property*
				Angle of repose	Carr·s index
F15a	13.6	11.4	1.16	Excellent	good
F15b	21	27.5	1.34	good	poor

Evaluation of Mirtazapine Tablets

The mechanical properties of pharmaceutical tablets are quantifiable by the friability, hardness or crushing strength. The hardness of all the formulas as shown in Table (7) had an acceptable value 7, 5.5 kg/cm^2. The hardness of F15a containing MCC (Avicel) ® 72 mg was 7kg /cm2 larger than F15b. During compression, MCC (Avicel)® PH 102 is believed to undergo stress relief deformation by several mechanisms, this might be attributed to the hydrogen bonds formed among the hydroxyl groups of the adjacent cellulose particles of (Avicel)®, which are brought closely together by plastic deformation during compression, so that it produces hard tablets at low compression forces [28]. The loss in total weight of the tablets due to friability was found in all formulation, which indicated to be less than 1% which was in accordance to the IP specifications for friability and which confirms the mechanical stability of tablets [29]. Physical properties of the prepared tablets, weight variation and drug content, demonstrated in Table (7). The weight

variation of F15a, F15b was within the pharmacopoeia limit which is ± 7.5% of the average weight. Weight variation of the prepared tablets was within the limit (199.2 mg, 197.9 mg) and this indicates that there is no deviation from the limit of 7.5% of USP pharmacopoeia limits [30]. The content uniformity of the prepared formulas was within the accepted pharmacopeia limits (85%-115%) and this mean that all the formulations revealed good uniformity and had yielded results from 101%, 98.7 respectively of the theoretical claim.

Table 7: Mechanical Strength and Physical Properties of the Prepared Mirtazapine Incorporating Drug Nanoparticles

Formula	Hardness (kg/cm2)	Friability %	Weight variation (mg)	Drug content (%)
F15a	7	0.25	199.2	101
F15b	5.5	0.67	197.9	98.7

Effect of Concentration of MCC (Avicel) ® on the Dissolution of Tablet

The dissolution profiles of the prepared tablets were significantly ($p<0.05$) affected by the concentration of MCC (Avicel) ®. F15 a, which contain higher amount of MCC, maintains its ranking first in the dissolution profile in spite of larger hardness, but when the concentration of MCC decreased by addition of other excipient the release also decreased. MCC enhances drug dissolution by speeding tablet disintegration, and utilizes dual disintegration mechanisms of wicking and swelling for more rapid disintegration so that MCC act as dissolution enhancer [31].

Effect of Addition of Starch on the Dissolution of Tablet

Starch was added to the prepared formula F15a and F15b at concentrations 20% w/w and 25% w/w as a binder and drug carrier; respectively. However, the addition of starch at low concentration (20% w/) was significantly ($p<0.05$) affect on the release of mirtazapine nanoparticles from F15a, while at high concentration (25% w/w) the release was ($p<0.01$) very slow. This may due to the effect of binder films that can form viscous gels on the granule surface and will retard dissolution [32].

In Vitro Dissolution Study of Tablet

The release profiles of the prepared mirtazapine tablets incorporating drug nanoparticles (F15a, F15b) and tablet marketing of mirtazapine as a reference were tested in 0.1N HCl (pH 1.2) and phosphate buffer solution (pH 6.8) as shown in Figures (6) and (7); respectively. In both media of dissolution velocity of mirtazapine from the nanoparticles tablets (F15a) was faster compared with F15b and the marketed tablet of mirtazapine.

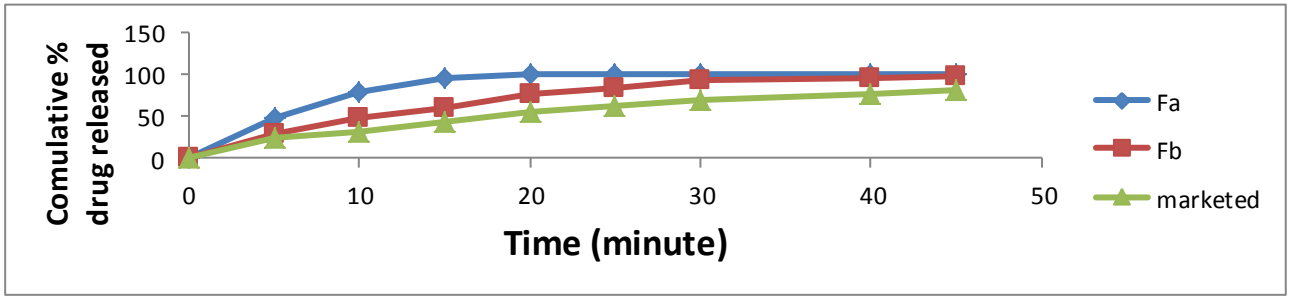

Figure 6: Dissolution profile of prepared tablets and mirtazapine marketed in buffer (pH 1.2) at 50 r.p.m and 37°C

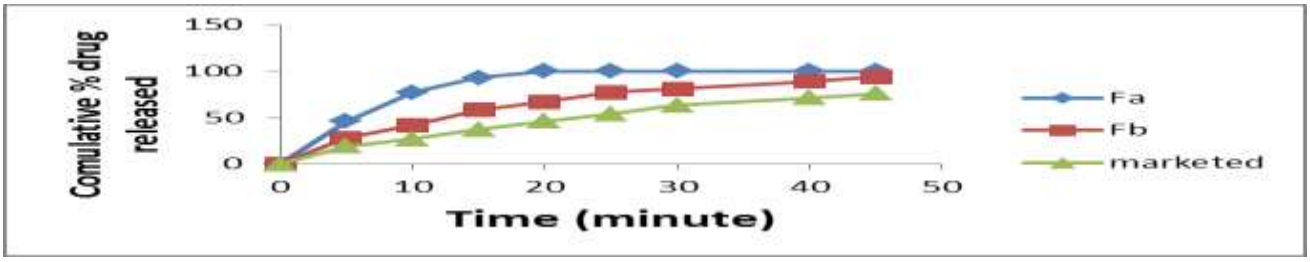

Figure 7: Dissolution profile of prepared tablets and mirtazapine marketed in buffer (pH 6.8) at 50 r.p.m and 37°C

Fourier Transform Infrared Spectroscopy

FTIR is one of the most widely reported spectroscopic techniques for solid-state characterization. IR spectroscopy of mirtazapine (Figure 8), N-H stretching 3439 cm^{-1}, Methyl group attached to a N2 atom gives rise to a band at 2932 cm^{-1} , Bands for C-C stretching of the phenyl group appeared at 1585 cm^{-1} and 1491 cm^{-1}. The primary aromatic amines with N directly on the ring give bands at 1336-1200 cm^{-1}. The benzene ring C-H appears in the range of 1359-1074 cm^{-1} and 788-636 cm^{-1} for the in plane and out of plane bending vibrations respectively [33, 34]. The characteristic bands of mirtazapine as lyophilized powder show N-H stretching 3466 cm^{-1}. The benzene ring C-H appears 1109cm^{-1}, C-H stretching vibrations band of methyl group at 3101.94 cm^{-1}, Bands for C-C stretching of the phenyl group appeared at 1642 cm^{-1}, as show in Figure 9. The FTIR study demonstrate that no physical or chemical interactions of Mirtazapine with other excipients [35].

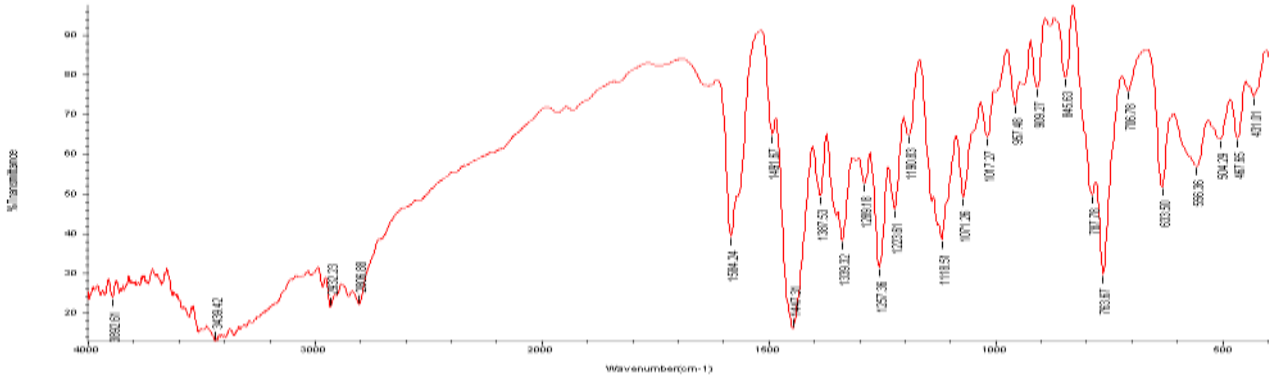

Figure 8: FT-IR Spectra of Mirtazapine Pure Powder

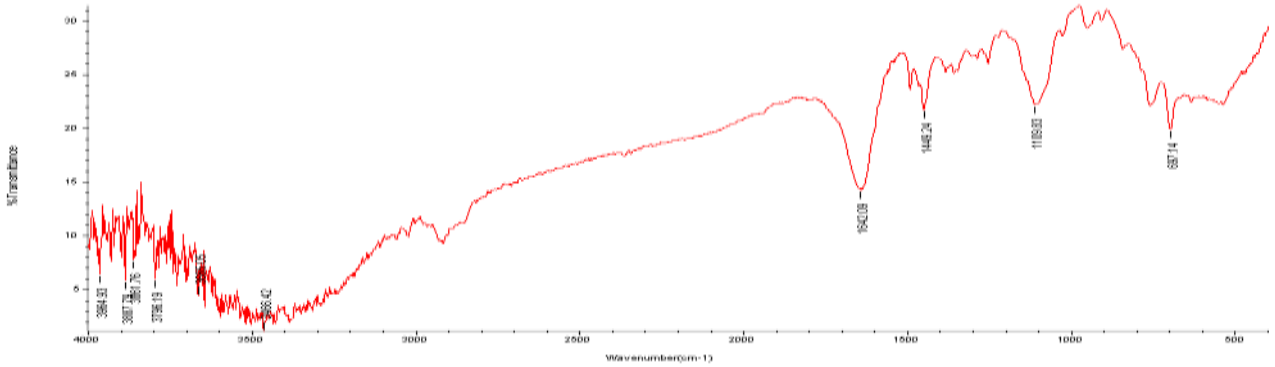

Figure 9: FTIR spectrum of lyophilized powder

Differential Scanning Calorimetry

Figure (10) demonstrate DSC of mirtazapine showed sharp characteristic endothermic peak at 117°C and this agrees with published results. This gives an indication that the drug has crystalline nature with high purity. For lyophilized powder, the melting point of mirtazapine disappeared giving a strong indication that the drug lost the crystallinity state and converted to an amorphous form [36].

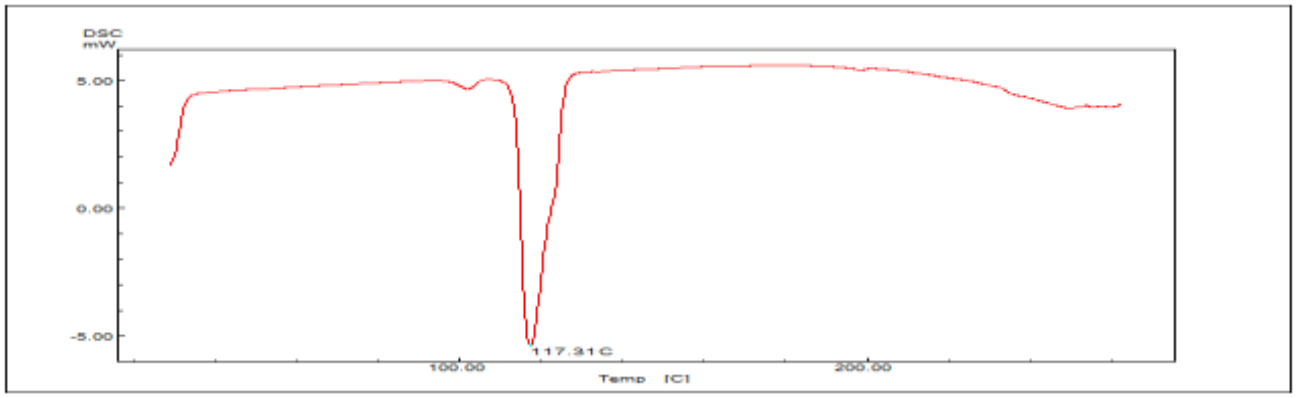

Figure 10: DSC Thermogram of Mirtazapine Pure Powder

Atomic Force Microscopy Study (AFM)

AFM is akind of scanning probe microscopes (SPM). It is an instrument that measures the properties of surfaces. AFM is capable of scanning the surfaces in controlled environmental conditions and is complementary to SEM imaging. With the high precision of the AFM, in principle it is possible to determine the dimensions of nanoparticles with high accuracy. AFM allows the visualization of samples with resolution in three dimensions x-, y- and z-directions in atmospheric or submerged conditions. The morphological analysis of mirtazapine pure powder performed by AFM showing spherical shaped nanoparticles. It was found to be stable and no aggregation of particles could be observed [37, 38]. The morphological analysis and particle size performed by AFM showing irregular to spherical shaped nanoparticles with a size of 189 nm as seen in Figure (11).The formulation was found to be stable and no aggregation of particles could be observed. The particle size of F15 obtained by AFM was comparable to or equal to that measured by ABT-9000 nano laser and this agreement in particle size measurements provide the good size distribution and the stability of mirtazapine nanparticles [39].

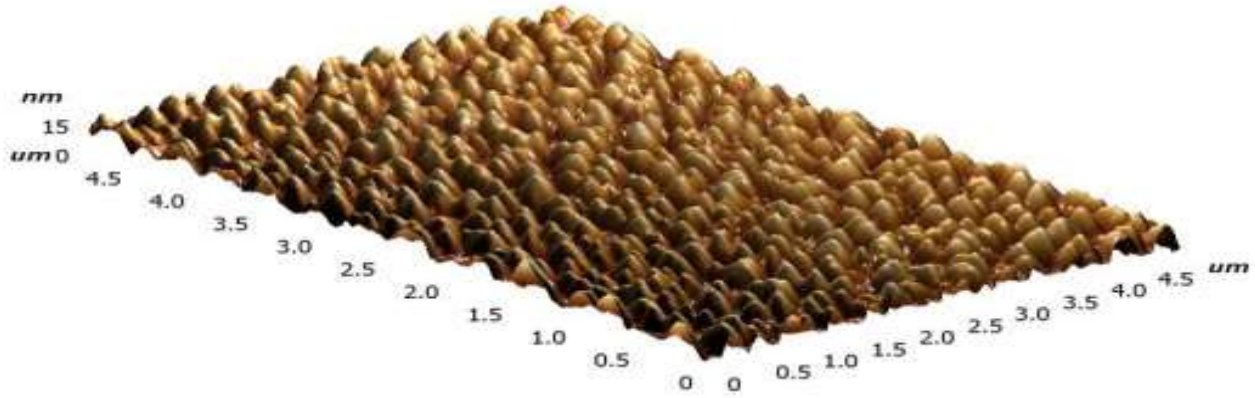

Figure 11: AFM of F15

Conclusion

Nano particulate systems such as anti-solvent precipitation have a great potential method, being able to convert poorly soluble mirtazapine. Poorly absorbed and labile biologically active substance in to promising deliverable drugs.

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