Innovare Academic Sciences

International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 7, Issue 1, 2015

Original Article

COMPATIBILITY STUDIES OF RASAGILINE MESYLATE WITH SELECTED EXCIPIENTS FOR AN EFFECTIVE SOLID LIPID NANOPARTICLES FORMULATION

VIVEKSARATHI KUNASEKARAN, KANNAN KRISHNAMOORTHY*

[1]Department of Pharmacy, Faculty of Engineering and Technology, Annamalai University, Annamalai Nagar 608002, Tamil Nadu, India, [2]Assistant professor, Department of Pharmacy, Faculty of Engineering and Technology, Annamalai University, Annamalai Nagar 608002, Tamil Nadu, India.
Email: egkkannan@yahoo.co.in

Received: 13 Jul 2014 Revised and Accepted: 25 Aug 2014

ABSTRACT

Objective: Compatibility study is an important element that should be performed at the early development stage of stable and effective solid dosage form.

Methods: The compatibility studies of drug with different polymers and surfactant were investigated by using different methods Fourier Transform Infrared Spectroscopy (FTIR), Differential Scanning Calorimetry (DSC) and Isothermal Stress Testing (IST). The compatibility study of Rasagiline mesylate was performed with Chitosan, Stearic acid and Poloxamer 407. Drug and polymer mixtures were prepared in 1:1 ratio and the compatibility study was performed at25°C±2/60%±5RHand 40°C±2/75%±5RHfor one month. The resulting compatible drug and excipients were used in the formulation of solid lipid nanoparticles by Microemulsion method.

Results: FTIR Spectroscopy results suggest that there is good compatibility between the drug and polymers. DSCresults showed that interaction between Rasagiline mesylate and Chitosan,the other excipients were compatible with RM and the IST study results clearly indicate the stable nature of the Rasagiline mesylate. The prepared RM-SLNs formulation shows a mean particle size within the nanometre range. SEM confirmed the size and shape of RM-SLNs. The results of zeta potential indicates a narrow size distribution. The RM-SLNs were found to be with acceptable morphometric properties, narrow size distribution, high entrapment efficiencyand good stability after 3 months of storage at different conditions (25°C±2/60%±5RH and 40°C±2/75%±5RH).

Conclusion: The FTIR, DSC and IST results shown that Stearic acid and Poloxamer 407were found to be compatible with Rasagilinemesylate. The Rasagiline mesylate loaded nanoscale solid lipid particles was successfully formulated with compatible excipients and it was found to be stable after accelerated stabilitystudies.

Keywords: Solid lipid nanoparticles, Rasagiline mesylate, Compatibility studies, Isothermal stress testing, Scanning electron microscopy.

INTRODUCTION

Compatibility study is an important element that should be performed at the early development stage of stable and effective solid dosage form. The design of successful formulation of nanoparticles depends on the selection of polymers and surfactant[1]. These polymers and surfactant can be selected on the basis of its compatibility and functionality with the drug[2]. Drug and polymers incompatibility can alter the physicochemical properties, stability and bioavailability of drugs, and subsequently affect its safety and therapeutic efficacy [3-6].

The compatibility testing is agreeable to computerization and high-throughput techniques that may contribute to the time consuming and reduction of cost [7,8]. Compatibility between drug and polymers can be thoroughly investigated and characterized by Fourier Transform Infrared Spectroscopy (FTIR), Differential Scanning Calorimetry (DSC), Isothermal Stress Testing (IST), Scanning Electron Microscopy (SEM),X-ray Powder Diffraction (XRD) and High Performance Liquid Chromatography (HPLC). Among these techniques FTIR and DSC are widely being used to evaluate the drug-polymer interactions. FTIR Spectroscopy is a simple technique, based on the vibrations of the atoms of a molecule. Disappearance of an absorption peak or reduction of the peak intensity combined with the appearance of new peak provides a clear indication for interaction between the drug and excipients[9]. DSC is a rapid analytical technique allows for the evaluation of possible interactions among the formulation components according to appearance, shift or disappearance of endothermic or exothermic peaks and/or variations in the relevant enthalpy values in thermal curves of drug-excipients mixtures [10-13]. IST is a commonly used method for evaluation of compatibility and involves storage of the drug-excipients mixtures with or without moisture at elevated temperature for a specific period of time and the drug content is determined by suitable method [9,14].

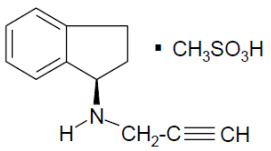

Fig. 1: Chemical structure of Rasagiline mesylate

The Anti-parkinsonism drug Rasagiline mesylate is a selective, second generation, irreversible monoamine oxidase B inhibitor, for the treatment ofidiopathic Parkinson's disease [15-18]. Rasagiline mesylate was approved by European drug regulatory authorities in 2005 and by the US FDA in 2006[19]. Rasagiline is a secondary cyclic benzylamine and indane derivatives with the chemical structure of N-propyl-1-(R)-aminoindan, the most potent propargylamine, which, although structurally related to selegiline [20]. The purpose of this work was to evaluate the compatibility between Rasagiline mesylate with various polymers and surfactant by FTIR, DSC and IST. The result of drug and compatible excipients use to prepare the solid lipid nanoparticle formulations and to evaluate the nanoparticles morphometric properties by photon correlation microscopy (PCS) and scanning electron microscopy (SEM).

MATERIALS AND METHODS

Materials

Rasagiline mesylate was kindly gifted from Orchid health care Pvt Ltd, Chennai, India. Chitosan was obtained commercially from

Central Marine Fisheries Research Institute, Cochin, India. Stearic acid and Tween 80 were purchased from SD Fine Chem Ltd, Mumbai, India. Lutrol (Poloxamer 407) was a gift sample from Signet Chemicals Lab, India. Acetonitrile (HPLC grade) were purchased from Sigma Aldrich, Mumbai, India. All other chemicals and reagents used were of analytical grade. Double distilled water was filtered through a 0.45 µm membrane (cellulose acetate) before use.

Methods

Preparation of samples

The mixture of Rasagiline mesylate with selected polymers (Chitosan and Stearic acid) and surfactant (Poloxamer 407) was prepared in 1:1 w/w ratio by the simple blending of the components in a mortar with pestle for 10 min at room temperature. The drug and polymers individually weighed in glass vials, each vial was sealed with Teflon-lined screw cap and the mixture of drug and polymers were stored in two different conditions at 25°C±2/60%±5 RH and 40°C±2/75%±5 RH for one month.

FTIR spectrophotometric analysis

Generally, the sample concentration in potassium bromide should be in the range between 0.2% and 1%. The dried potassium bromide was placed into a mortar 1% w/w of the drug sample was accurately weighed and mixed with the KBr powder, subsequently ground the mixture for 3-5 minutes. The powder mixtures were pressed through a mechanical press to form translucent pellets. These pellets were scanned by FTIR (Perkin-Elmer model 1600) in the 4000-400 cm^{-1} spectral range [21].

Differential scanning calorimetric analysis

Compatibility studies of the mixtures of the drug and excipients were performed by using differential scanning calorimetry (DSC, Shimadzu, Japan). 2-10mg of the drug and excipients mixture as individual samples and mixtures of drug and excipients were accurately directly weighed directly into standard aluminum pans and placed in the equipment under room temperature and it was scanned in temperature from 50-300°C. The heating rate was 20°C per min in nitrogen atmosphere (Flow rate: 20 ml/min) and the interactions were observed from obtained thermograms[22].

Isothermal stress testing

In Isothermal stress testing (IST) studies, drug with selected polymers and surfactant weighed in glass vials (n=3) and mixed with mortar for 3 mins, with 10% distilled water. Each glass vial sealed by Teflon lined screw cap and stored at 50°C in hot air oven (Technico, India). Samples without water were used as control and stored in refrigerator. These samples were regularly examined for any unusual colour change. After 4 weeks these samples were analyzed quantitatively at 272 nm by using UV spectrophotometer (1601, Shimadzu, Japan) [23].

Preparation of Rasagiline mesylate loaded solid lipid nanoparticles

Rasagiline mesylate (RM) loaded solid lipid nanoparticles were prepared from O/W microemulsion technique [24-26] containing Stearic acid as lipid phase, poloxamer 407 as surfactant and Tween 80 as co-surfactant. Briefly, the lipid phase (stearic acid) was heated 5-10°C above its melting point (69-70°C), to which Rasagiline mesylate was added.

The aqueous phase containing surfactant (poloxamer 407) and co-surfactant (Tween 80) were heated at 50°C and added to the melted lipid phase under magnetic stirrer (Remi, India.) at 500rpm for 10-15 min, resulting in O/W emulsion. The resulted emulsion was immediately added drop wise into cold distilled water under probe sonicator for 20 min (Lark, India) to solidify the nanoparticles.

Characterization of nanoparticles

Determination of process yield

The process yield of Rasagiline mesylate solid lipid nanoparticles were determined using the following formula: [26, 27]

Process yield = (Practical yield/Theoretical yield) × 100.

Determination of particle size and poldispersity index (PdI)

The particle size and poldispersity index (PdI) for the Rasagiline mesylate loaded solid lipid nanoparticles was carried out by photon correlation spectroscopy (PCS) using Malvern Mastersizer 2000 (Malvern Instruments Inc, Worcestershire, UK.) and the samples were measured in the range from 0.02 µm to 2000 µm.

Polydispersity index, a parameter calculated from the width of the particle size of distribution by using the equation = D (0.9) –D (0.1) /D (0.5). Where, D(0.9), D(0.5) and D(0.1) are corresponding to particle size immediately above 90%, 50% and 10% of the sample. [26, 28].

Zeta potential measurements

Zeta potential of Rasagiline mesylate loaded SLNs formulation was determined by laser Doppler electrophoresis (Zetasizer Nano ZS, Malvern, UK). The samples were diluted with deionized distilled water with a conductivity adjusted to 50µS/cm with sodium chloride. Helmholtz–Smoluchowski equation was used to convert the measured electrophoretic mobility to zeta potential. [26,29]

Scanning electron microscopy (SEM)

The morphology of RM loaded SLNs characterized by scanning electron microscopy (Carl Zeiss EVO-MA25, Germany). The samples were frozen at -80°C and lyophilized (Lark, Penguin Classic Plus, India) by freeze-drying method. 10 mg of freeze-dried samples were suspended in 1 ml deionized distilled water and one droplet of the suspension was placed on a glass surface. After drying the latex particles, coated with gold using an Ion Sputter coater and scanned at an accelerating voltage of 20 kV. [30]

Determination of entrapment efficiency

The entrapment efficiency (EE) of the RM loaded SLNs was determined by measuring the concentration of free drug inthe supernatant obtained after centrifuging at 16,000rpm for 30 mins at 0°C (Remi C 24, Mumbai, India). The amount of free drug was detected by HPLC (Thermoscientific, spectra system P-4000, USA) and the incorporated drug was calculated by subtracting free drug from total amount of drug in the formulation. The EE was calculated by using the following equation,[26,31]

$$\text{Entrapment efficiency (\%)} = \frac{(\text{W1(assay)} - \text{W2}))}{\text{W1}} \times 100$$

Where, W1 -the total amount of drug in the formulation,

W2 -amount of drug in the supernatant.

Determination of drug content

Rasagiline mesylate loaded SLNs (2 ml) were diluted to 10 ml with acetonitrile: water (5:95, v/v) andfiltered through 0.45 µm nylon Millipore membranes (Millipore, USA). The drug content was determined by using HPLCsystem (Thermoscientific, spectra system P-4000, USA) with UV detector (Kromosil 100) and C18 column (particle size 5 µm, 250 mm×4 mm). The absorbance was examined at 265 nm [26,32].

Stability studies

The purpose of the stability testing is to illustrate how the quality of a drug substance varies with time under the influence of a variety of environmental factors like temperature, humidity and light. The Rasagiline mesylate formulation was stored up to 3 months for stability studies at two different storage condition like (25°C±2/60°±5RH) and an accelerated condition (40°C±2/75%±5RH)[33,34]. The stored formulation was evaluated at 0, 1, 2 and 3 months for their particle size, polydispersity index, zeta potential, drug contentand drug entrapment.

RESULTS AND DISCUSSION

FTIR spectra of pure drug and a mixture of drug and selected excipients studies were performed in two different storage

conditions like 25°C±2/60%±5RH and 40°C±2/75%±5RH for one month.

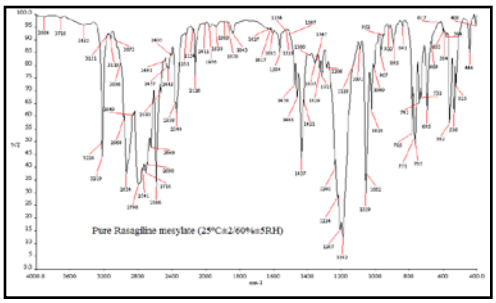

Fig. 2: FTIR spectra of pure Rasagiline mesylate (25°C±2/60%±5RH) FTIR spectra of pure Rasagiline mesylate are presented in each fig. together with single excipients spectrum is shown in the Fig. 3-5.

Data acquired from FTIR Spectrophotometric studies of drug-excipients mixtures stored at 25°C±2/60%±5RH, indicates no significant changes in the spectra. The pure Rasagiline mesylate characteristic spectra were shown, a band of 2934 $cm^{-1}$and 2849 cm^{-1}owing to Aromatic C-H stretching and Aliphatic C-H stretching groups. The other bands Peaks at 3219 cm^{-1} (Secondary amine N-H) and 1192 cm^{-1}(Aliphatic amino C-N) respectively. The pure Rasagiline mesylate spectrum is shown in Fig. 2.

FTIR spectra of Rasagiline mesylate, Chitosan and mixture of Rasagiline mesylate with Chitosan are shown in Fig. 3. Chitosan exhibits a band at 3421 cm^{-1} corresponds to the combine peaks of the stretching vibration of NH_2 and OH group. A characteristic broadband at 1157 cm-1 associated with the stretching of C-O. The band at 2920 cm^{-1} is attributed to the Aromatic C-H stretching (Table 1). The binary mixture of Rasagiline mesylate and Chitosan shows no significant changes in the peaks of Primary and Secondary amine, Aromatic C-H stretching, Aliphatic C-H stretching and aliphatic amine C-N, in reference to the observed value of Rasagiline mesylate and Chitosan.

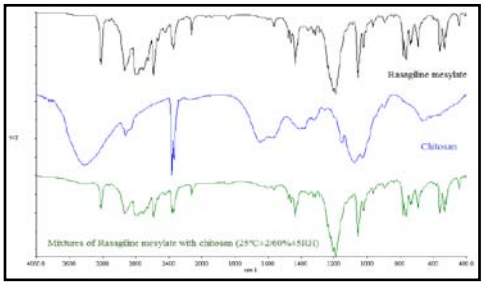

Fig. 3: FITR spectra of (A) pure Rasagiline mesylate (B) chitosan (C) mixtures of Rasagiline mesylate with chitosan (25°C±2/60%±5RH).

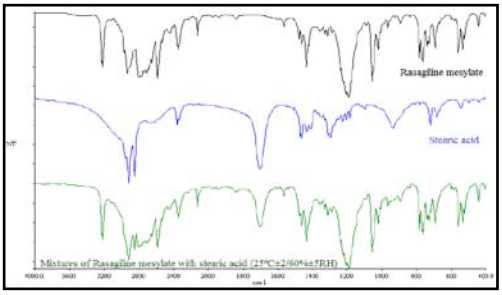

Fig. 4: FITR spectra of (A) pure Rasagiline mesylate (B) stearic acid (C) mixtures of Rasagiline mesylate with stearic acid (25°C±2/60%±5RH)

Table 1: FTIR spectroscopy data of RM and chitosan in (25°C±2/60%±5RH) storage condition

S. No.	F. G	Rasagiline mesylate		F. G	chitosan		F. G	RM+ chitosan cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm^{-1}	chitosan cm^{-1}		
1	Ar C-H	2850-3000	2934	Ar C-H	2850-3000	2920	1° NH /Sec NH	3421/3219
2	Ali C-H	2800-2900	2798	1° NH	3400-3500	3421	Ar C-H	2933
3	Sec N-H	3300-3400	3219	O-H	3200-3600	3421	Ali C-H	2800
4	Ali C-N	1000-1350	1192	C-O	1100	1157	Ali C-N	1191

F. G: Functional groups / RM: Rasagiline mesylate

Table 2: FTIR spectroscopy data of RM and stearic acid in (25°C±2/60%±5RH) storage condition

S. no	F. G	Rasagiline mesylate		F. G	stearic acid		F. G	RM + stearic acid cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm-1	stearic acid cm^{-1}		
1	Ar C-H	2850-3000	2934	Ali C-H	2800-2900	2849	Imide	1701
2	Ali C-H	2800-2900	2798	C=O	1720-1740	1702	Ar C-H	2917
3	Sec N-H	3300-3400	3219	O-H	2500-3300	2674	Ali C-H	2805
4	Ali C-N	1000-1350	1192	CH_2 bend	1100	1463	Ali C-N	1191

F. G: Functional groups / RM: Rasagiline mesylate

Table 3: FTIR spectroscopy data of RM and poloxamer 407 in (25°C±2/60%±5RH) storage condition

S. No.	F. G	Rasagiline mesylate		F. G	poloxamer 407		F. G	RM + poloxamer 407 cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm^{-1}	poloxamer 407 cm^{-1}		
1	Ar C-H	2850-3000	2934	NH OH	3400-3500	3447	Sec NH/NH OH	3119/3447
2	Ali C-H	2800-2900	2798	Ali C-H	2800-2900	2896	Ar C-H	2917
3	Sec N-H	3300-3400	3219	C-O	1100	1109	Ali C-H	2804
4	Ali C-N	1000-1350	1192	-	-	-	Ali C-N	1190

F. G: Functional groups / RM: Rasagiline mesylate

Data acquired from FTIR spectra of Rasagiline mesylate-excipients stored at 40°C±2/75%±5RH condition is shown in the Fig. 6-9.

Fig. 4 show the FTIR spectra of Rasagiline mesylate, Stearic acid and mixtures of Rasagiline mesylate and Stearic acid. The characteristic band Peaks at around 2849 cm^{-1} and 1702 cm^{-1} in Stearic acid are assigned to the Aromatic C-H stretching and C=O Asymmetric stretching, respectively.

The peaks present at 2674 cm^{-1} and 1463 cm^{-1} owing to Carboxylic O-H and CH_2 bending. Mixtures of Rasagiline mesylate and Stearic acid spectra showed no change in the positions of the bands at 1701 cm^{-1} (Imide), 2917 cm^{-1}(Aromatic C-H stretching), 2805 cm^{-1} (Aliphatic C-H stretching) and 1191 cm^{-1}(Aliphatic amine C-N) in Rasagiline mesylate (Table 2).

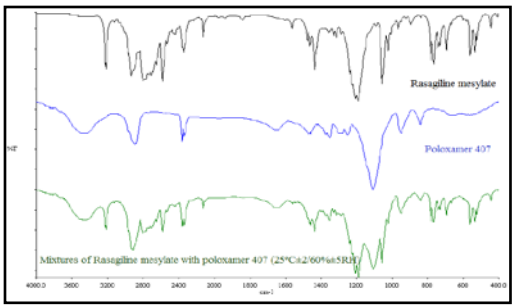

Fig. 5: FITR spectra of (A) pure Rasagiline mesylate (B) poloxamer 407 (C) mixtures of Rasagiline mesylate with poloxamer 407 (25°C±2/60%±5RH)

The Poloxamer 407 FTIR spectrum is characterized by principal absorption band peaks at 3447 cm^{-1} (NH OH), 2896 cm^{-1}(O-H bending) and 1109 cm^{-1}(C-O stretching) (Table 3). The Rasagiline mesylate-Poloxamer 407 mixtures showed the respective characteristic bands peaks at 3119 cm^{-1}, 3447 cm^{-1}, 2917 cm^{-1}, 2804 cm^{-1} and 1190 cm^{-1} shown in Fig. 5. The results confirm there is no change in the positions of the bands peaks in the mixtures Rasagiline mesylate and Poloxamer 407 spectra.

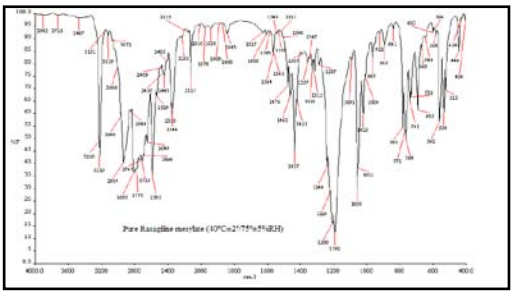

Fig. 6: FTIR spectra of pure Rasagiline mesylate (40°C±2˚/75°±5%RH)

FTIR spectra are usually analysis in a range between 4000 and 400 cm^{-1} with 4 cm^{-1} resolution. Spectra obtained from pure Rasagiline mesylate were found 2934 $cm^{-1}$and 2805 cm^{-1} for Aromatic C-H stretching and Aliphatic C-H stretching respectively. The other characteristic bands Peaks at 3219 cm^{-1} and 1192 cm^{-1}assigned for to Secondary amine N-H and Aliphatic amine C-N respectively. The pure Rasagiline mesylate spectrum is shown in Fig. 6.

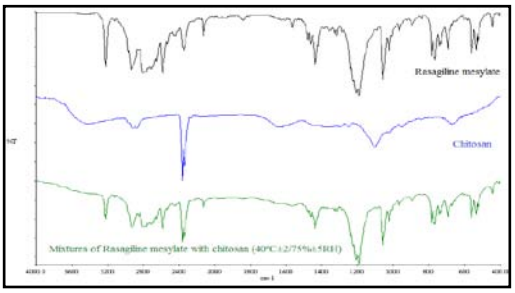

Fig. 7: FITR spectra of (A) pure Rasagiline mesylate (B) chitosan (C) mixtures of Rasagiline mesylate with chitosan (40°C±2/75%±5RH)

Table 4: FTIR spectroscopy data of RM and chitosan in (40°C±2/75%±5RH) storage condition

S. No.	F. G	Rasagiline mesylate		F. G	chitosan		F. G	RM + chitosan cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm^{-1}	chitosan cm^{-1}		
1	Ar C-H	2850-3000	2934	Ar C-H	2850-3000	2917	1° NH /Sec NH	3430/3219
2	Ali C-H	2800-2900	2805	1° NH	3400-3500	3430	Ar C-H	2917
3	Sec N-H	3300-3400	3219	O-H	3200-3600	3430	Ali C-H	2866
4	Ali C-N	1000-1350	1192	C-O	1100	1032	Ali C-N	1103

F. G: Functional groups / RM: Rasagiline mesylate

Table 5: FTIR spectroscopy data of RM and stearic acid in (40°C±2/75%±5RH) storage

S. No.	F. G	Rasagiline Mesylate		F. G	stearic acid		F. G	RM + stearic acid cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm^{-1}	stearic acid cm^{-1}		
1	Ar C-H	2850-3000	2934	Ali C-H	2800-2900	2849	Imide	1701
2	Ali C-H	2800-2900	2805	C=O	1720-1740	1702	Ar C-H	2917
3	Sec N-H	3300-3400	3219	O-H	2500-3300	2917	Ali C-H	2804
4	Ali C-N	1000-1350	1192	CH_2 bend	1100	1099	Ali C-N	1191

F. G: Functional groups / RM: Rasagiline mesylate

Table 6: FTIR spectroscopy data of RM and poloxamer 407 in (40°C±2/75%±5RH) storage condition

S. No.	F. G	Rasagiline Mesylate		F. G	poloxamer 407		F. G	RM + poloxamer 407 cm^{-1}
		standard cm^{-1}	RM cm^{-1}		standard cm^{-1}	poloxamer 407 cm^{-1}		
1	Ar C-H	2850-3000	2934	NH OH	3400-3500	3446	Sec NH/NH OH	3119/3448
2	Ali C-H	2800-2900	2798	Ali C-H	2800-2900	2891	Ar C-H	2934
3	Sec N-H	3300-3400	3219	C-O	1100	1113	Ali C-H	2800
4	Ali C-N	1000-1350	1192	-	-	-	Ali C-N	1191

F. G: Functional groups / RM: Rasagiline mesylate

The data obtained from the FTIR spectra of Rasagiline mesylate, Chitosan and mixture of Rasagiline mesylate-Chitosan are shown in Fig.7. Characteristic bands of Chitosan were observed at 3430 cm^{-1}represents to the combine peak of NH_2 and OH groups stretching vibration [35]. The peaks at 1032 cm^{-1} and 2917 cm^{-1} were assigned to the presence of stretching of C-O and Aromatic C-H stretching. Spectrograph of both the Rasagiline mesylate and Chitosan physical mixture shows at 3430 cm^{-1} (Primary amine), 3219 cm^{-1} (Secondary amine), 2917 cm^{-1}(Aromatic C-H stretching), 2866 cm^{-1} (Aliphatic C-H stretching) and 1103 cm^{-1} (Alkane C-N) respectively (Table 4). These results signify that the mixture of Rasagiline mesylate-Chitosan is compatible.

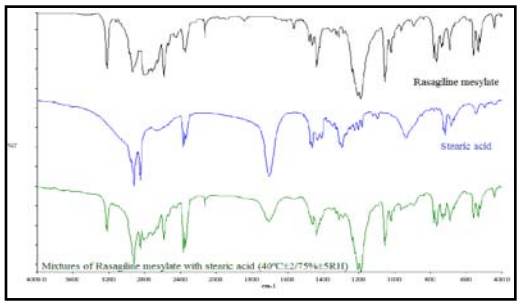

Fig. 8: FITR spectra of (A) pure Rasagiline mesylate (B) stearic acid (C) mixtures of Rasagiline mesylate with stearic acid (40°C±2/75%±5RH)

The Rasagiline mesylate-Stearic acid mixtures are subjected to FTIR spectra studies and its spectrum is compared with the Rasagiline mesylate and Stearic acid bands peaks are shown in Fig. 8. The characteristic bands of Stearic acid were observed at 2849 cm^{-1} (Aliphatic C-H stretching), 1702 cm^{-1} (C=O Asymmetric stretching), 2917 cm^{-1} (Carboxylic O-H) and 1099 cm^{-1}(CH_2 bending). The FTIR spectrum of the Rasagiline mesylate-Stearic acid mixture shows the band peaks at 1701 cm^{-1} (Imide), 2917 cm^{-1}(Aromatic C-H stretching), 2804 cm^{-1} (Aliphatic C-H stretching) and 1191 cm^{-1}(Aliphatic amine C-N) in (Table 5). These results showed, bands peaks were not altered in binary mixtures, it represents no interactions between Rasagiline mesylate and Stearic acid.

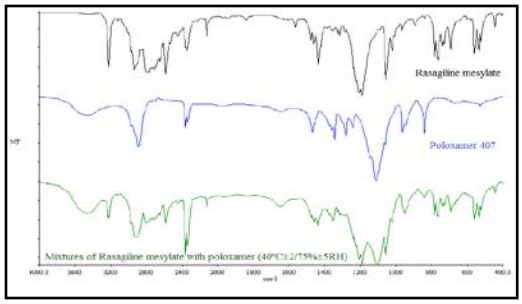

Fig. 9: FITR spectra of (A) Pure Rasagiline mesylate (B) poloxamer 407 (C) mixtures of Rasagiline mesylate with poloxamer (40°C±2/75%±5RH)

The FTIR spectra study of Poloxamer 407 reveals that characteristic band peaks appeared at 3446 cm^{-1} (NH OH), 2891 cm^{-1}(O-H bending) and 1113 cm^{-1} (C-O stretching) in Fig. 9. The binary mixture of Rasagiline mesylate and Poloxamer 407 shows the respective characteristic peaks at 3119 cm^{-1} (Secondary amine), 3447 cm^{-1} (NH OH), 2934 cm^{-1} (Aromatic C-H stretching), 2800 cm^{-1} (Aliphatic C-H stretching) and 1191 cm^{-1} (Aliphatic amine C-N) (Table 6), which is similar to that in individual spectrum. It represents that there were no changes in the characteristic band peaks.

DSCthermoanalytical curves of drug and drug-excipients mixtures are illustrated in Fig.10-13 and the drug and mixtures of drug-excipients thermal behaviour was compared.

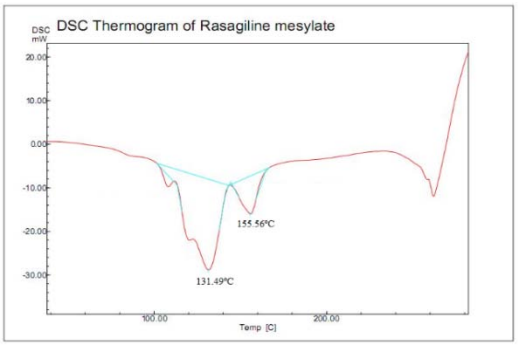

Fig. 10: DSC thermogram of Rasagiline mesylate

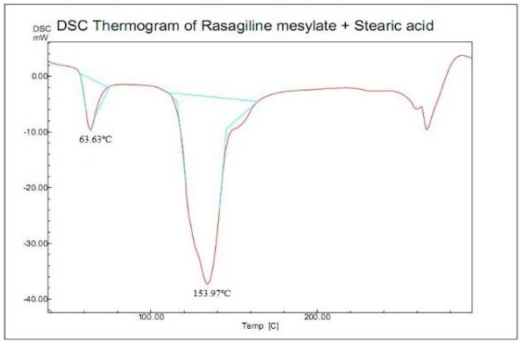

Fig. 11: DSC thermogram of Rasagiline mesylate and stearic acid

The DSC thermogram of Rasagiline mesylate showed an endothermic peak at 131.49°C (It represents the dehydration of bound water) and 155.56°C (melting point). Rasagiline mesylate-Stearic acid mixtures showed the endothermic peak at 63.63°C and 153.97°C (Fig.11). The melting endothermic peak of Rasagiline mesylate at 155.56°C in Rasagiline mesylate-Stearic acid mixture confirms there is no interaction between Stearic acid and Rasagiline mesylate.

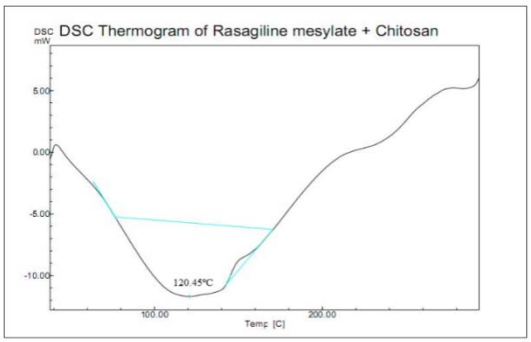

Fig. 12: DSC thermogram of Rasagiline mesylate and chitosan

DSC thermogram of Rasagiline mesylate and Chitosan can be observed in Fig.12 and it showed the endothermic peak at 120.45°C. There was no peak appears in the range of Rasagiline mesylate melting peak. The thermogram of Rasagiline mesylate and Chitosan mixture confirm that Chitosan was incompatible with Rasagiline mesylate.

The DSC curve of Rasagiline mesylate and Poloxamer 407 showed endothermic peaks at 55.91°C and 155.53°C in fig. 13. In this Rasagiline mesylate melting peak appears at the same value of temperature (155.55 °C). The binary mixtures of thermogram indicate no interaction between these substances.

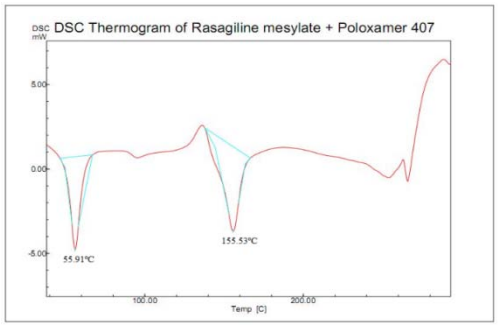

Fig. 13: DSC thermogram of Rasagiline mesylate and poloxamer 407

In the Isothermal stress testing, drug and mixtures of drug-excipients results were confirmed there is no change in physical appearance at ambient temperature. The mixtures also predominantly examined for physical stable, liquefaction or gas formation and drug degradation with any type of excipients. The percentage of drug remaining at the end of the study at 50°C was shown in the Table-7. The Rasagiline mesylate loaded solid lipid nanoparticles were prepared by Microemulsion technique. The process yield, mean particle size, polydispersity index, zeta potential, drug content and entrapment efficiency of Rasagiline mesylate loaded SLNs results were shown in Fig. 14, 15 and table 8. The prepared RM loaded solid lipid nanoparticles process yield was 88.6 % and the mean particle size range of 165 to 210 nm Fig.14. The polydispersity index (PdI) ratio provides the information about the homogenicity of particle size distribution and it should be <0.3 (Pathak et al., 2009), the PdI values of RM-loaded SLNs was 0.226, which represents that the SLNs had a narrow size distribution and signifying SLNs monodispersity. The Zeta potential (surface charge) is a factor for evaluation of the stability in colloidal dispersion through the electrostatic repulsion between particles. The particles are electrochemically stable, when the zeta potential value of the nanoparticles is more than ±30mV. The zeta potential value of RM-SLNs was -36.2 mV Fig.15.

Table 7: Results of UV analysis of the samples, under Isothermal stress testing after 4 weeks.

Sample	Ratio (Drug-excipient)	% Drug remaining[a]		changes in physical appearance
		control sample[b]	stress sample[c]	
Rasagiline mesylate (RM)	-	100.32±1.11	99.03±1.46	No
RM+Chitosan	1:1	97.77 ±0.54	100.64±0.55	No
RM+Stearic acid	1:1	99.35 ±1.46	99.35±1.66	No
RM+Poloxamer 407	1:1	101.61±0.56	100.32±0.55	No

[a]Mean ± standard deviation (n=3)
[b]Drug-excipients blendswithout added water and stored in refrigerator (2° to 8°C)
[c]Drug-excipients blends with 10% added water and stored at 50° C for 4 weeks

Table 8: Process yield, particle size polydispersity index and zeta potential for RM-loaded solid lipid nanoparticles

Evaluation parameters	Formulations code
	RMSLN
Process yield (%)	86.1
Mean particle size (nm)	165-210
Polydispersity Index	0.226
Zeta potential (mV)	-36.2
Drug content* (%)	97.85±0.08
Drug entrapment (%)	82.92

*(n=3±S. D.)

Table 9: Stability studies of Rasagiline loaded solid lipid nanoparticles formulation

Temperature	Evaluation parameter	Observation (month)			
		0	1	2	3
25°C±2/60%±5RH	Mean particle size (nm)	165-210	165-215	170-210	165-215
	Polydispersity index (PdI)	0.226	0.222	0.219	0.224
	Zeta potential (mV)	-36.2	-35.9	-36.0	-36.3
	Drug content*(%)	97.85 ±0.08	97.91 ±0.28	97.41 ±0.22	97.40 ±0.13
	Drug entrapment (%)	82.92	82.80	82.59	82.61
40°C±2/75%±5RH	Mean particle size (nm)	165-210	167-215	165-210	170-215
	Polydispersity index (PdI)	0.226	0.223	0.226	0.220
	Zeta potential (mV)	-36.2	-36.4	-36.0	-36.1
	Drug content* (%)	97.85 ±0.08	97.89 ±0.18	97.83 ±0.07	97.24 ±0.41
	Drug entrapment (%)	82.92	82.81	82.52	82.45

*(n=3±S. D.)

The morphology of RM loaded SLNs was examined using scanning electron microscopy. The photographic result shows a fine spherical shape, smooth surface, and particle size was within the nanometric range Fig.16.

The entrapment efficacy and drug content of the prepared Rasagiline mesylate loaded solid lipid nanoparticles was 82.92% and. 97.85±0.08%.

The results of Rasagiline mesylate loaded nanoparticles (RMSLN) formulation stability studies indicate there is no significance change in particle size, polydispersity index, zeta potential, drug content and drug entrapment before and after 1, 2 and 3 months and these values are shown Table 9.

CONFLICT OF INTERESTS

The authors declare that they have no competing interests.

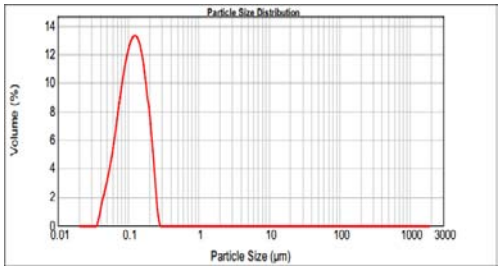

Fig. 14: Particles size of Rasagiline loaded solid lipid nanoparticles

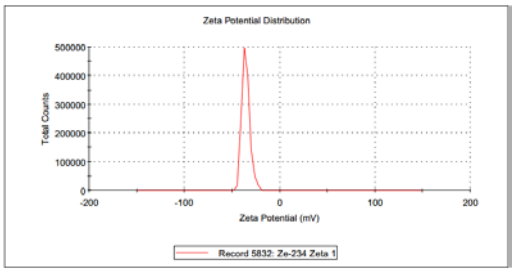

Fig. 15: Zeta potential value of Rasagiline loaded solid lipid nanoparticles

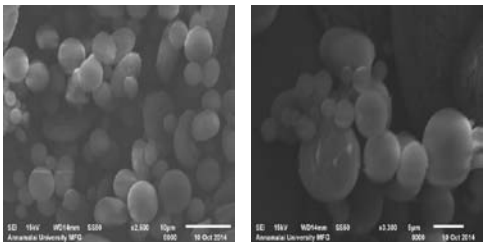

Fig. 16: SEM image of Rasagiline mesylate loaded solid lipid nanoparticles

CONCLUSION

The compatibility studies of Rasagiline mesylate with various polymers and surfactant were investigated using different analytical methods. FTIR and DSC were used as rapid methods to evaluate possible incompatibilities of drug and excipients. FTIR spectra results confirmed the absence of physical or chemical interactions between Rasagiline mesylate and the corresponding excipients. The results of thermoanalytical studies (DSC) have shown an incompatibility between Rasagiline mesylate and Chitosan. The presence of Chitosan is responsible for degradation of the binary mixtures at different w/w ratio. There is no significant change in the thermogram of Rasagiline mesylate with other excipients.

The results of Isothermal stress testing showed that there is no change in colour and drug content after 4 weeks of storage under stressed conditions. It clearly indicates the stable nature of Rasagiline mesylate with other excipients used in the present study. The compatibility studies conclude that Rasagiline mesylate is compatible with the excipients like stearic acid and poloxamer 407. The RM loaded SLNs formulations developed using these compatible excipients was found to be acceptable morphotmetric properties, narrow size distribution, high entrapment efficiency and good stability after 3 months of storage at different conditions (25°C±2/60%±5RH and 40°C±2/75%±5RH).

REFERENCES

1. Aulton ME. Pharmaceutics: the scientific principles of dosage form design. 2nd edition. Churchill Livingstone; 2002. p. 136.
2. Akhilesh Vikarm Singh, Lila Kanta Nath. Evaluation of compatibility of lamivudine with tablet excipients and a novel synthesized polymer. J Mater Environ Sci 2011;2(3):243-50.
3. Bogdan Tit, Adriana Fulias, Geza Bandur, Eleonora Marian, Dumitru Tit. Compatibility study between ketoprofen and pharmaceutical excipients used in solid dosage forms. J Pharm Biomed Anal 2011;56:221-7.
4. Picciochi R, Diogo HP, Da Piedade MEM. Thermochemistry of paracetamol. J Therm Anal Cal 2010;9:391-401.
5. Tita B, Fulias A, Szabadai Z, Rusu G, Bandur G, Tit D. Compatibility study between ibuprofen and excipients in their physical mixtures. J Therm Anal Cal 2011;105:517-27.
6. Neto HS, Noak CS, Matos JR. Thermal analysis and compatibility studies of prednicarbate with excipients used in semi solid pharmaceutical form. J Therm Anal Cal 2009;97:367-74.
7. Tatsuyoshiwakasawa, Kyoko sano, Yutaka hirakura, Toshimasatoyooka, Satoahikitamura. Solid-state compatibility studies using a high throughput and automated forced degradation system. Int J Pharm 2008;355:164-73.
8. Carlson E, Chandler W, Galdo I, kudla T, Ta C. Automated integrated forced degradation and drug-excipients compatibility studies. J Assoc Lab Autom 2005;10:374-80.
9. Karin Liltorp, Trine Gorm Larsen, Birgitte Willumsen, Rene Holm. Solid state compatibility studies with tablet excipients using non thermal Methods. J Pharm Biomed Anal 2011;55:424-8.
10. Corvi Mora P, Cirri M, Mura P. Differential scanning calorimetry as a screening technique in compatibility studies of DHEA extended release formulations. J Pharm Biomed Anal 2006;42:3-10.
11. Harding L, Qi S, Hill G, Reading M, Craig DQM. The development of microthermal analysis and photo thermal microspectroscopy as novel approaches to drug-excipient compatibility studies. Int J Pharm 2008;354:149-57.
12. Ceschel GC, Badiello R, Ronchi C, Maffei P. Degradation of components in drug formulations: a comparison between HPLC and DSC methods. J Pharm Biomed Anal 2003;32:1067-72.
13. Lira AM, Araujo ASS, Bas lio IDJ, Santos BLL, Santana DP, Macedo RO. Compatibility studies of lapachol with pharmaceutical excipients for the development of topical formulations. Thermochim Acta 2007;457:1-6.
14. Serajuddin ATM, Thakur AB, Ghoshal RN, Fakes MG, Ranadive SA, Morris KR, Varia SA. Selection of solid dosage form composition through drug excipient compatibility testing. J Pharm Sci 1998;88:696-704.
15. Rascol O, Brooks DJ, Melamed E, Oertel W, Poewe W, Stocchi F, *et al*. Rasagiline as an adjunct to levodopa in patients with Parkinson's disease and motor fluctuations (LARGO, lasting effect in adjunct therapy with rasagiline given once daily, study): a randomised, double-blind, parallel-group. Lancet 2005;365:947-54.
16. Orly Weinreb, Silvia Mandel, Orit Bar-Am, Merav Yogev-Falach, Yael Avramovich-Tirosh, Tamar Amit, *et al*. Multifunctional neuroprotective derivatives of rasagiline as anti-alzheimer's disease drugs. Neurother: J Am Soc Exper Neuro Ther 2009;6:163-74.
17. Youdim MB, Maruyama W, Naoi M. Neuropharmacological, neuroprotective and amyloid precursor processing properties of selective MAO-B inhibitor anti-Parkinsonian drug, rasagiline. Drugs Today 2005;41:369-91.
18. Blandini, Armentero MT, Fancellu R, Blaugrund E, Nappi G. Neuroprotective effect of rasagiline in a rodent model of Parkinson's disease. Exper Neuro 2004;187:455-9.
19. David R, P Guay. Rasagiline (TVP-1012): a new selective monoamine oxidase inhibitor for parkinson's disease. Am J Geri Pharmacother 2006;4(4):331-46.
20. Irene A Malaty, Hubert H Fernandez. Role of rasagiline in treating Parkinson's disease: effect on disease progression. Ther Clin Risk Manage 2009;5:413-9.
21. Lifeng Qi, Zirong Xu, Xia Jiang, Caihong Hu, Xiangfei Zou. Preparation and antibacterial activity of chitosan nanoparticles. Carbohydr Res 2004;339:2693-700.
22. Santanu Mallik, Mahendra D. Kshirasagar, Vipin S Ani. Studies on physical/chemical compatibility between drugs with

various pharmaceutical excipients. Der Pharm Let 2011;3(5):173-8.

23. Cotton ML, Wu DW, Vadas EB. Drug-excipient interaction study of enalaprilmaleate using thermal analysis and scanning electron microscopy. Int J Pharm 1987;40:129-42.
24. Doijad RC, Manvi FV, Godhwani DM, Joseph R, Deshmukh NV. Formulation and targeting efficiency of cisplatin engineered solid lipid nanoparticles. Indian J Pharm Sci 2008;70(2):203-7.
25. Mayur K, Padhi BK, Chougule M, Mishra A. Methotrexate loaded solid lipid nanoparticles for topical treatment of psoriasis: formulation and clinical implication. Drug Deliv Tech 2002;5:1-13.
26. Viveksarathi K, Kannan K. Multi criteria decision making to select the best method for the preparation of solid lipid nanoparticles of rasagiline mesylate using analytic hierarchy process. J Adv Pharm Technol Res 2014;5(3):1-7.
27. Barnabas Wilson, Malay Kumar Samanta, Kumaraswamy Santhi, Kokilampal Perumal, Sampath Kumar, Nallupillai Paramakrishnan, *et al*. Poly (n-butylcyanoacrylate) nanoparticles coated with polysorbate 80 for the targeted delivery of rivastigmine into the brain to treat Alzheimer's disease. Brain Res 2008;1200:159-68.
28. Kashanian S, Azandaryani AH, Derakhshandeh K. New surface-modified solid lipid nanoparticles using N-glutarylphosphatidylethanolamine as the outer shell. Int J Nanomed 2011;6:2393-401.
29. Zhinan Mei, Huabing Chen, Ting Weng, Yajiang Yang, Xiangliang Yang. Solid lipid nanoparticle and microemulsion for topical delivery of triptolide. Eur J Pharm Biopharm 2009;56:189-96.
30. Shuyu Xie, Luyan Zhu, Zhao Dong, Yan Wang, Xiaofang Wang, Wen Zhong Zhou. Preparation and evaluation of ofloxacin-loaded palmitic acid solid lipid nanoparticles. Int J Nanomed 2011;6:547-55.
31. Sanjay singh, Ajay Kumar dobhal, Achintjain, Jayant Kumar pandit, Subhashis Chakraborty. Formulation and evaluation of solid lipid nanoparticles of a water soluble drug: zidovudine. Chem Pharm Bull 2010;58(5):650-5.
32. Marcos Fernandez, Sofia Negroa, Karla Slowing, Ana Fernandez-Carballido, Emilia Barcia. An effective novel delivery strategy of rasagiline for Parkinson's disease. Int J Pharm 2011;419:271-80.
33. Viveksarathi K, Rajarajan R, Kannan K, Manavalan R. Dosage form design and evaluation of Eperisone hydrochloride matrix film coated extended release tablets. Int J Pharm Pharm Sci 2012;4(2):575-81.
34. Laith Hamza Samein. Preparation and evaluation of nystatin loaded-solid-lipid nanoparticles for topical delivery. Int J Pharm Pharm Sci 2014;6(2):592-7.
35. Rajan K Verma, Sanjay Garg. Selection of excipients for extended release formulations of glipizide through drug-excipient compatibility testing. J Pharm Biomed Anal 2005;38:633-44.