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Research Article

Formulation and Characterization of Eudragit RLPO based Sustained Release Nanoparticles

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ABSTRACT

Osteoporosis is the most frequent metabolic disease that affects bone. Bisphosphonates, especially, alendronate sodium, are indicated as first line regimen for this disease. Alendronate is highly efficient but presents low absorption after oral administration, due to high water solubility burst release occurs. The aim of the present study was to reduce burst release and to sustain drug release from Alendronate sodium Nanoparticles containing the polymer Eudragit RLPO. The nanoparticles were prepared by Emulsion solvent evaporation technique and were characterized for mean particle size, surface charge, size distribution, drug entrapment efficiency, drug loading capacity and *In vitro* drug release. Drug excipients compatibility studies performed using DSC instrument indicated that there were no interactions. The results revealed that double emulsion solvent evaporation method is suitable for preparing nanoparticle formulation. The drug loading capacity of the particles varied depending on the drug polymer ratio. The mean particle size of the selected batch was 238 nm.

INTRODUCTION

Oral route is the most popular for sustained delivery of drugs because of ease of administration. Controlled release delivery systems provide a uniform concentration after absorption, allow maintenance of plasma concentration within the therapeutic range, minimizes side effects and it is convenient route of drug delivery. [1] Too hydrophilic drugs would not be able to permeate through the gastrointestinal mucosa and too lipophilic drug will not dissolve in the aqueous gastrointestinal contents. For optimum absorption, the drug should have sufficient aqueous solubility to dissolve in the gastrointestinal contents and also adequate lipid solubility to facilitate its partitioning into the lipoid membrane and then into systemic circulation. Drugs having partition coefficient (log P) value in the range of 1 to 3 shows good passive absorption across lipid membranes, and those having log Ps greater than 3 or less than 1 have often poor transport characteristics.[2] Osteoporosis's characterized by low bone mass and structural deterioration of bone, both of which are associated with reduced bone strength and increased fracture risk. In the United States alone, 10 million people are estimated to already have osteoporosis, with more than one million osteoporotic fractures occurring every year.[3] Alendronate sodium belongs to BCS class III.[4] Bisphosphonates, especially, alendronate sodium, are indicated as first line regimen. Alendronate is highly efficient but presents low absorption after oral administration with bioavailability of less than 1%. Oral bisphosphonates must be taken with a full glass of water. A 30- to 60-minute wait is required before reclining or consuming other medications, beverages, or food to lower the risk of upper gastrointestinal adverse effects. The optimal length of oral bisphosphonate therapy is unknown. Drug could lead to many side effects, which are mainly related to the esophagus, the stomach and the intestine. Such effects are linked to a local contact of drug crystals with the mucosa. Encapsulation of active molecules allowed the obtaining of many advantages over conventional pharmaceutical forms such as, bioavailability and tolerance enhancement.

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Nanotechnology remains an attractive approach to overcome such limitations of conventional pharmaceutical forms. However, encapsulation of hydrophilic molecules like alendronate represents a real challenge. Preparation, characterization and drug release behavior of eudragit RLPO based nanoparticles loaded with alendronate sodium were investigated in this work. double emulsion evaporation method used to prepare these nanocarriers.[4,5]

Polymeric nanoparticles have versatile potential for efficient exploitation of different drug delivery formulations and routes because of the properties provided by their small size. There are reports that nanoparticular drug delivery systems possess various advantages over their corresponding conventional drug delivery systems. These possible benefits include controlled release, protection of the active pharmaceutical ingredient and drug targeting. Eudragit RLPO was selected as a polymer for preparation of alendronate sodium loaded nanoparticles due to its high permeability and application for sustained release drug delivery systems.[6] Techniques based on double emulsions are commonly used for the encapsulation of hydrophilic molecules, which suffer from low encapsulation efficiency because of rapid drug partitioning into the external aqueous phase when using single emulsions. The main issue when using double emulsions is their production in a well-controlled manner, with homogeneous droplet size by optimizing different process variables. [7]

In the present work an attempt was made to develop nanoparticles of alendronate sodium using eudragit RLPO by double emulsion solvent evaporation method. Alendronate sodium suffers from specific problems, of which the prominent being the low bioavailability. Drawbacks may be attributed to its low permeability. Therefore, there are continued efforts to improve the pharmaceutical formulation of alendronate sodium in order to achieve an optimal therapy and ensure the desired therapeutic response of prepared nanoparticulate formulations.

MATERIALS AND METHODS

Alendronate sodium was a gift sample from Troikaa Pharmaceutical Pvt. Ltd, Ahmedabad, Eudragit RLPO, pluronic F 68 were obtained as gift sample from Madras Pharma, Chennai dialysis membrane purchased from Hi-media Ltd., India vasa scientific Bangalore. Ethanol and all other reagents and chemicals used were analytical grade.

Experimental methods

Preparation of alendronate sodium loaded Eudragit RLPO Nanoparticles

Nanoparticles were prepared by double emulsion–solvent evaporation technique. The weighed quantity of drug dissolved in aqueous phase was dispersed in ethanol containing Eudgrit RLPO and soya lecithin, under ultrasonication using an ultrasonicator to form w/o emulsion. The primary emulsion was added rapidly into an aqueous phase containing stabilizer under vigorous stirring and ultrasonicated to form w/o/w emulsion. The double emulsion was stirred for 4 h for the removal of organic phase. The nanoparticle dispersion was centrifuged at 15,000 rpm for 30 min, washed 3 to 4 times with distilled water and finally lyophilized to obtain free-flowing powder. The prepared nanoparticles were stored in tightly sealed containers under refrigeration.[8-10]

Physicochemical characterization of Nanoparticles

Compatibility study

Compatibility of drug and polymer were analyzed using FT-IR (Fourier transform infrared) spectroscopy, Shimadzu Corporation, Japan by the potassium bromide disc method. The samples (alendronate sodium, Eudragit RLPO and nanoparticles) were homogeneously mixed with potassium bromide and infrared spectrums were recorded in region of 4000-400 cm^{-1} by using FT-infrared spectrophotometer and DSC.[11]

Particle size and zeta potential

The mean diameter of alendronate sodium -loaded nanoparticles was measured by using a laser light scattering particle size analyzer (Malvern Mastersizer Hydro 2000 SM, Malvern Instruments Ltd.,) at 25 °C with a 90° scattering angle. Milli-Q water was used as a dispersant medium.the zeta potential, average particle size and polydispersibility index were recorded for all the formulations.[12]

Morphological Analysis

The morphology alendronate sodium -loaded nanoparticles was examined by scanning electron microscopes (Jeol JSM-840 A Tokyo, Japan.

Entrapment efficiency (EE)

The drug entrapped in the nanoparticles was estimated by dispersing the weighed amount of particles in ethanol by ultrasonication and followed by extraction of free drug into phosphate buffer pH 6.8. The extract was analyzed for drug spectrophotometrically at 565 nm after suitable dilution. The drug loading efficiency (LE) and drug entrapment efficiency (EE) was calculated using Eqs. 1 and 2 respectively.[13]

$$\text{Drug loading efficiency} = \frac{\text{Drug content in the product obtained in (mg)}}{\text{Total product weight in mg}} \times 100 \quad \ldots(1)$$

$$\text{Drug entrapment efficiency} = \frac{\text{Drug content in the product obtained in (mg)}}{\text{Total amount of drug added in mg}} \times 100 \quad \ldots(2)$$

***In vitro* dissolution study**

In vitro release was performed by using USP-I (basket type) dissolution apparatus at 37^{o}c ± 0.5^{o}c and at 50 rpm using dialysis bag (molecular weight cutoff 10-14kDa) was treated previous day prior to experiment. The dissolution media simulated gastric fluid (SGF), pH 1.2, for two h; simulated intestinal fluid (SIF), pH 6.8, for 48 h in 900 ml buffers. 10mg equivalent of drug was placed in the bag and dispersed in 1ml of medium, tied at both the ends and dipped in medium for the study and the samples were withdrawn and filtered through 0.22µ filter and diluted, at preset time intervals and replaced by an equal volume of fresh release medium. The procedure is continued till 48 h. The samples are analyzed by UV spectrophotometer at 565nm. A profile showing the cumulative drug release as a function of time was plotted.[14]

Results and Discussions

Five Formulations were developed keeping amount of drug at a fixed level and varying the polymer drug ratio1:2.5, 1:5, 1:10, 1:15, 1:20 and pluronic f-68 used in different concentration 0.5, 01, 1.5 % w/v to stabilize nanoparticles. (Table 2) by the double emulsion solvent evaporation method, several research workers have investigated utilization of Eudragit RLPO as a retardant polymer to encapsulate highly water soluble drugs using the double emulsion solvent evaporation method. This method used to modify the drug release of highly water soluble drugs.

Particle size is often used to characterize Nanoparticles and affects biological handling of nanoparticles. The smaller particle sizes will ensure lowered level of reticulo endothelial system (RES) uptake, improve utilization ratio of drug and diminish drug side effects. Table 1 and Fig. 3 depict particle size and zeta potential of eudragit RLPO. The mean size of the particle size of Eudragit RLPO nanoparticles was 238 nm, the value of zeta potential less than−30 mV or higher than +30 mV can be used to assure the stabilityof nanoparticles suspensions. The values obtained for the prepared nanoparticles was found to be -20.1 (Table 1) which indicates high degree of stability due to inner particle repulsion but found to agglomerate after few days.

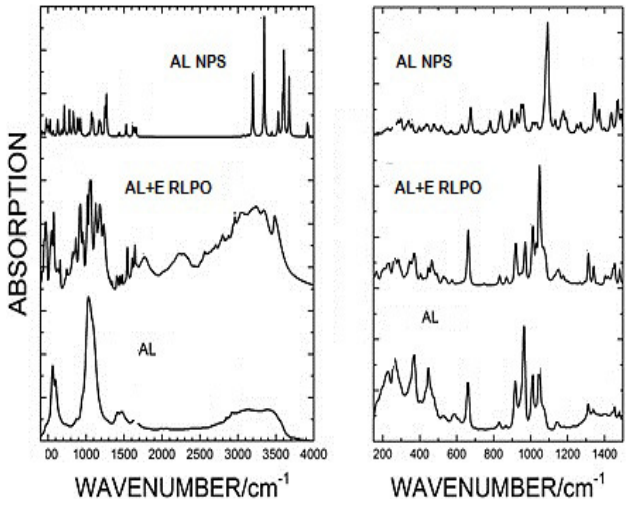

Figure 1: FTIR spectra of pure ALS, physical mixture of ALS and Eudragit RLPO, ALS NPs

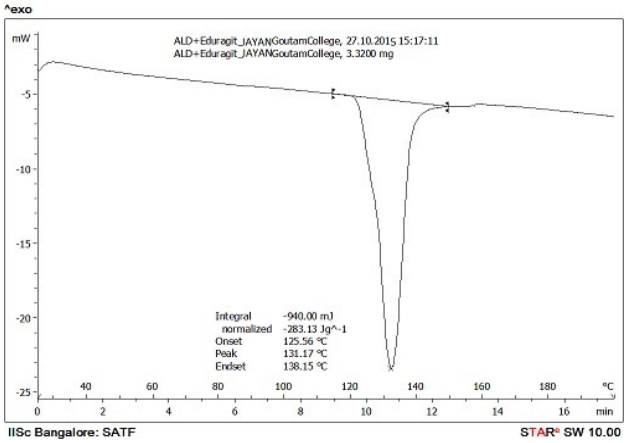

Figure 2: DSC of physical mixture of Eudragit RLPO and Alendronate Sodium

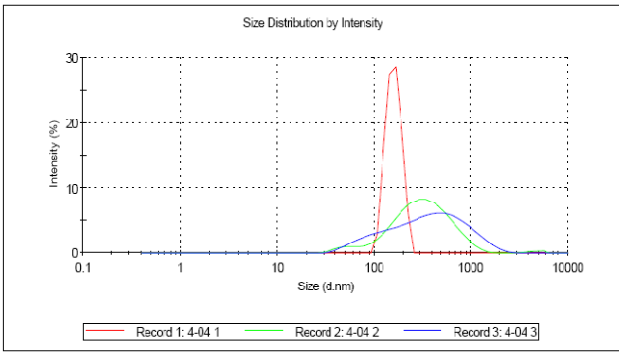

Figure 3: Particle size distribution of best formulation of ALS-loaded nanoparticles

Table 1: Physicochemical characterization of Nanoparticles

Formulation code	Particle size	PDI	Zeta potential
E1	361	0.619	−14.61
E2	423	0.561	−15.61
E3	238	0.478	−20.1
E4	245	0.471	−17.13
E5	388	0.494	−21.2

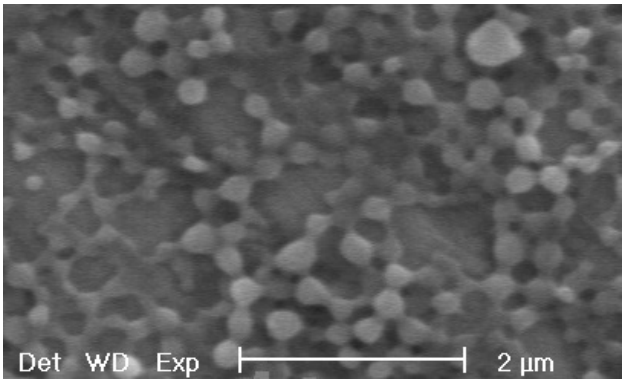

Figure 4: SEM photography of optimized formulation of ALS NPs

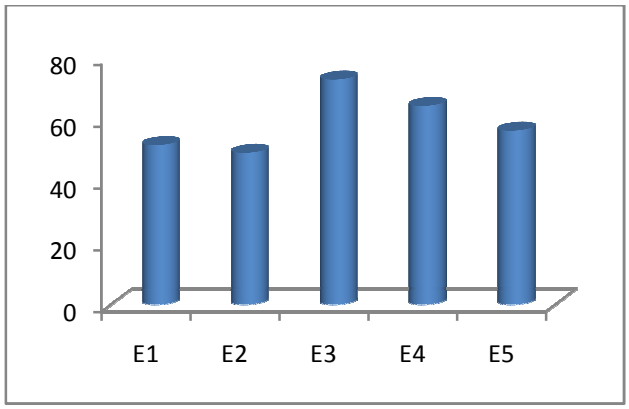

Figure 5: Percentage Drug Entrapment efficiency

Table 2: Composition and % Drug entrapment efficiency of ALS loaded nanoparticles

Formulation code	D:P Surfactant	Pluronic F-68 % w/v	% EE *
E1	1:2.5	-	51.65 ± 0.39
E2	1:5	0.5	49.16 ± 0.58
E3	1:10	1	72.87 ± 0.68
E4	1:15	1.5	64.35 ± 0.19
E5	1:20	0.5	56.35 ± 0.33

*All the tests were carried out in triplicate n=3 mean ± SD

It was observed that the entrapment efficiency increased with the increase in concentration of polymer in the formulations. The maximum entrapment was found in E-3 of 72.87 and lowest entrapment in E2 of 49.16.

All the five formulations of prepared Nanoparticles of alendronate sodium and pure drug alendronate sodium (E) were subjected to *in vitro* release studies.

The *in vitro* dissolutions study shows the release profile of the formulation is good. Highest release was shown by pure drug. The nanoparticle formulations produced the release profiles with an initial burst effect in which drug release ranged between 38% and 67% within 4 h and the lowest by E3 in a time period of 48 hrs. These results indicated that the prepared nanoparticles are useful controlled delivery system for osteoporosis treatment. This indicates that burst release of water soluble drug can be prevented and sustain the release of the drug. Encapsulated colloidal formulation reduces the adverse effects of alendronate sodium when administered orally. All formulations showed an interesting bi-phasic release with an initial burst release of alendronate sodium. The prepared nanocarrier composites are good releasing rate in both pH medium than other reported systems.

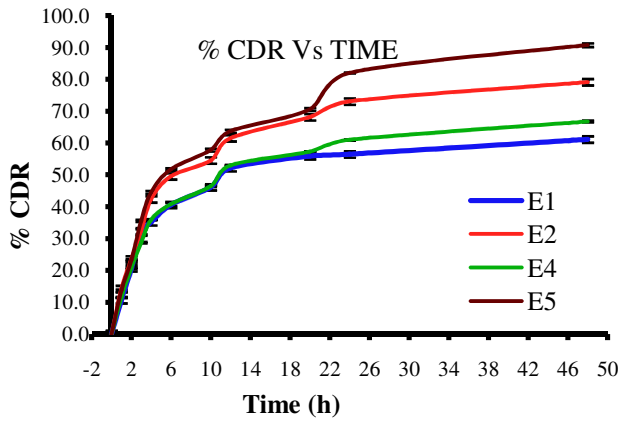

Figure 6: *In vitro* release profile of ALS nanoparticles

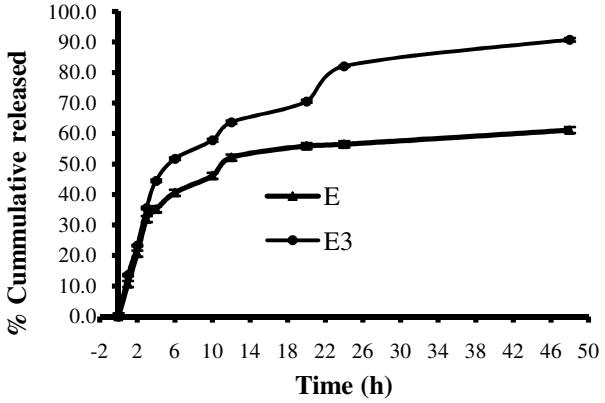

Figure 7: *In vitro* release profile of pure drug (E) and best formula of ALS nanoparticles

CONCLUSION

Release kinetics studies showed that alendronate sodium release from the Nanoparticles were First order. It can be concluded that the formulated nanoparticulate delivery system of highly water soluble drug alendronate sodium using widely accepted and physiologically safe polymer (Eudragit RLPO) was capable of exhibiting sustained release for prolonged period of time.

REFERENCES

[1] Mohammed S Khan,.Vishakante Gowda D, Shivakumar HG. Porous nanoparticles of metoprolol tartrate produced by spray-drying: Development, characterization and *in vitro* evaluation. Acta Pharm2012; 62: 383–394.

[2] Thakkar Hetal, Patel Bindesh, Thakkar Sneha. A Review on techniques for oral bioavailability enhancement of drugs. International Journal of Pharmaceutical Sciences Review and Research, 2010*;* 4: 33.

[3] Stepnick LS. The frequency of bone disease. In: MacGowan JALGR, Noonan AS, Elderkin AL (eds). Bone Health and Osteoporosis: A Report of Surgeon General Washington, DC, USA, 2004; 68–87.

[4] Arik Dahan, Jonathan M. Miller and Gordon L. Amidon. Prediction of Solubility and Permeability Class Membership: Provisional BCS Classification of the World's Top Oral Drugs, The AAPS Journal 2009; 11:740-46.

[5] Xiaodong Li, Michael S Ominsky, Kelly S Warmington, Sean Morony, Jianhua Gong, Jin Cao, Yongming Gao, Sclerostin Antibody Treatment Increases Bone Formation, Bone Mass, and Bone Strength in a Rat Model of Postmenopausal Osteoporosisjournal of bone and mineral research 2009; 24:578-588.

[6] Brown JB, Pedula K, Barzilay J, Herson MK, Latare P: Lactic Acidosis Rates in Type 2 Diabetes.Diabetes Care 1998; 21:1659–1663.

[7] BhosaleUV andKusum Devi V Preparation and *In Vitro* Evaluation of Acyclovir Loaded Eudragit RLPO Nanoparticles as Sustained Release Carriers RJPS, 2011; 1(1)

[8] Muhammad Iqbal, Nadiah Zafar, Hatem Fessi, Abdelhamid Elaissari. Double emulsion solvent evaporation techniques used for drug encapsulation. International Journal of Pharmaceutics 2015; 496 :173–190.

[9] Jong SuepBaek, HaeHyun Kwon, JiSook Hwang, Ha-Chang Sung, Jeong Min Lee, Sang Chul Shin., e*t al.,* Alendronate-loaded microparticles for improvement of intestinal cellular absorption, Journal of Drug Targeting. 2011; 19 (: 37–48.

[10] Mansi Shah P, Pooja Patel K, Lin S, Madan PL. Formulation variables affecting release of diclofenac sodium from eudragit-loaded microparticles. Asian Journal of Pharmaceutical Sciences2011; 6: 241-250.

[11] Tahereh Sadat Jafarzadeh Kashi, SolmazEskandarion, Mehdi Esfandyari-Manesh, Seyyed Mahmoud Amin Marashi, NasrinSamadi, Seyyed Mostafa Fatemi, et al., Improved drug loading and antibacterial activity of minocycline-loaded PLGA nanoparticles prepared by solid/oil/water ion pairing method. International Journal of Nanomedicine 2012; 7:221–234.

[12] Amolkumar B. Lokhande, Satyendra Mishra, Ravindra D. Kulkarni, Jitendra B. Naik. Influence of different viscosity grade ethylcellulose polymers on encapsulation and *in vitro* release study of drug loaded nanoparticles. Journal of Pharmacy Research . 2013;7:414-420.

[13] Cong Zhang , ConghuiGu, Fan Peng, Wei Liu, Jiangling Wan, Huibi Xu et al., Preparation and Optimization of Triptolide-Loaded Solid Lipid Nanoparticles for Oral Delivery with Reduced Gastric Irritation. Molecules2013; 18*:* 13340-13356.

[14] Lacramioara Ochiuz, Cristian Grigoras, Marcel Popa, Iulian Stoleriu, Corneliu Munteanu, Daniel Timofte *et al.,* Alendronate-Loaded Modified Drug Delivery Lipid Particles Intended for Improved Oral and Topical Administration. Molecules 2016; 21: 858.