INTERNATIONAL RESEARCH JOURNAL OF PHARMACY

www.irjponline.com ISSN 2230 – 8407

Research Article

FORMULATION AND EVALUATION OF SOLID LIPID NANOPARTICLES OF A POORLY WATER SOLUBLE MODEL DRUG, IBUPROFEN

De Pintu Kumar[1]*, Dinda Subas C[2], Chakraborty Subrata[1], Rakshit Soumen[2]

[1]Department of Pharmaceutics, Dr. B.C.Roy College of Pharmacy and Allied Health Sciences, Dr. Meghnad Saha Sarani, Bidhannagar, Durgapur, 713206, W.B. India

[2]School of Pharmaceutical Education & Research, Berhampur University, Bhanja Bihar, Berhampur, Orissa, India

Article Received on: 06/10/12 Revised on: 17/11/12 Approved for publication: 09/12/12

*Email: depintu72@yahoo.co.in

ABSTRACT

Approximately 40% of lipophilic drug candidates fail to comply the commercial requirements of solubility and formulation stability, prompting significant research activity in advanced lipophile delivery technologies. Solid lipid nanoparticle (SLN) technology represents a promising new approach to lipophile drug delivery. Solid lipid nanoparticles typically are spherical with average diameters between 1 to 1000 nanometers. SLNs possess a solid lipid core matrix that can solubilize lipophilic molecules. The lipid core is stabilized by surfactants. Primary objective of this study is to enhance the solubility and dissolution rate of Ibuprofen by formulation into SLN using hot homogenization method. Further, this study also investigates the effect of various formulation parameters like stabilizer concentration, surfactant ratio, Lipid ratio and drug loading. SLNs were characterized for size distribution, entrapment efficiency, drug release and stability. SLN of Ibuprofen was prepared using stearic acid (lipid) Phospholipon 80 H (surfactant) and Tween-80 as stabilizer. The FTIR study shows no major interaction of Ibuprofen with other formulation ingredients, and the Differential Scanning Calorimetry (DSC) study revealed that the drug is molecularly dispersed into the lipid. The particle size determinations confirm the particle size distribution in the nanoparticular range (27% Volume to 56% volume). In-vitro drug release through the dialysis membrane from the prepared SLNs is much higher than the pure drug. The stability study indicates the stability of the formulations without changing its performance on storage. Hence formulation of Ibuprofen in SLN enhances the dissolution rate as well as it will enhance the bioavailability of the drug which could be stabilized during storage.

KEY WORDS: Solid Lipid Nanoparticle, Lipophilic drug, Phospholipon 80H, Ibuprofen

INTRODUCTION

Oral ingestion is the most convenient and commonly employed route of drug delivery due to its ease of administration, high patient compliance, cost-effectiveness, least sterility constraints and flexibility in the design of dosage form. As a result, many of the generic drug companies are inclined more to produce bioequivalent oral drug products. The high costs and time involved in new drug development, expiry of patents for a significant number of drug molecules, ease of manufacturing and ready availability of technology for the production of oral drug products are also driving the generic pharmaceutical companies towards the development of bioequivalent oral dosage forms. However, the major challenge with the design of oral dosage forms lies with their poor bioavailability. The oral bioavailability depends on several factors including aqueous solubility, drug permeability, dissolution rate, first-pass metabolism, pre-systemic metabolism and susceptibility to efflux mechanisms [1, 2, 3]. The most frequent causes of low oral bioavailability are attributed to poor solubility and low permeability [4]. In recent years it has become more and more evident that the development of new drugs alone was not sufficient to ensure progress in drug therapy. Exciting experimental data obtained *in vitro* were very often followed by disappointing results *in vivo* because of the insufficient drug concentration due to poor absorption, rapid metabolism and elimination, poor water solubility and high fluctuation of plasma levels due to unpredictable bioavailability after per oral administration. A promising strategy to overcome these problems involves the development of suitable drug carrier systems [5]. The size of the carrier depends on the desired route of administration and ranges from few nanometers (colloidal carriers) to the micrometer range (microparticles) and to several millimeters (implants) [6]. Colloidal particles ranging in size between 10-1000nm are known as nanoparticles. They are manufactured from synthetic/natural polymers and ideally suited to optimize drug delivery and reduce toxicity. The successful implementation of nanoparticles for drug delivery depends on their ability to penetrate through several anatomical barriers, sustained release of their contents and their stability in nanometer size[7]. Solid lipid nanoparticle are tiny colloidal carriers composed of biocompatible or biodegradable lipid matrix that is solid at body temperature and exhibit size range in between 100-1000nm. Lipid can either be highly purified triglycerids, complex glyceride mixtures or even waxes [8]. In addition to the lipid and drug, it contains surfactants as stabilizers. Hence SLNs are the effective lipid based colloidal carriers which were introduced as an alternative to the conventional carriers such as microemulsions, liposomes, microparticles and nanoparticles based on synthetic polymers or natural macromolecules. Typically they enhance the oral bioavailability of the low aqueous soluble drugs due to their potential to enhance gastrointestinal solubilization and absorption via selective lymphatic uptake. These properties can be harvested to improve the therapeutic efficacy of the drugs with low bioavailability, as well as to reduce their effective dose requirement [9].

MATERIALS AND METHODS

Materials: Phospholipon-80H was received as a gift sample from Lipoid GmbH, Frigenstrasse 4, D-67065, Ludwigshafen. Ibuprofen was purchased from local supplier, stearic acid (Central Drug House (P) Ltd. New Delhi); Tween-80 (Finar Chemicals Limited, Ahmedabad, India); Dichloromethane (Merck Specialities Private Limited, Worli, Mumbai, India); Potassium Dihydrogen Ortho Phosphate (Finer Chemicals Limited, Ahmedabad-380006, India) and Sodium Hydroxide (Central Drug House (P) Ltd. New Delhi-110002, India) and all other required chemicals were procured from the suppliers.

Compatibility Study between Drug And Excipients by Fourier Transform Infrared Spectroscopy (FT-IR) [10]

FT-IR spectra of pure drug (Ibuprofen), Stearic acid, Phospholipon 80H and their physical mixtures (1:1:1) were recorded by grinding and dispersing the samples with micronized IR grade KBr powder. The mixture was dissolved in chloroform and casted on a sodium chloride disk, and subjected to FT-IR measurement over the range of 4000–600 cm^{-1} (Perkin Elmer, Model - 78625).

Preparation of Ibuprofen SLN by Hot Homogenization technique: [11]

Solid Lipid Nanoparticles of Ibuprofen were produced using the lipid (Stearic acid); surfactant (Phospholipon-80H) and stabilizer (Tween-80) in different proportion by hot homozenization technique using the high speed homogenizer [Ultra Turrax T25 (Janke & Kunkel, IKA Labortechnik, Germany)] given in Table No.1. The organic phase was prepared by dissolving the drug and surfactant in dichloromethane and mixing it with the melted stearic acid, which was further, poured into aqueous Tween-80 solution (maintained at the same temperature as that of organic phase) of various concentrations which acts as a stabilizer and stirred with UltraTurrax T25 (UT) on a water bath at 24,000 rpm for 10 minutes. The formulation was then removed from water bath and the dispersion of SLN was mixed gently by slow magnetic stirring (1 h) at room temperature until cooling.

Particle size determination

Particle size of the solid lipid nanoparticles was analyzed by laser diffractometry using a Mastersizer 2000 instrument (Malvern) equipped with a Hydro 2000MU (A) dispersing unit.

In-vitro drug release study[10]

The release of Ibuprofen from the solid lipid nanoparticles was compared with the pure drug using a dialysis system comprising of a HiMedia Dialysis Membrane-70 mounted over a jacketed dialysis beaker. The dialysis membrane was filled with 2 ml of solid lipid nanoparticles, clipped and exposed to diffusion medium containing Phosphate buffer pH (7.2), stirred magnetically & maintained at a constant temperature of 37°C ± 0.5°C. Aliquots were drawn at specified time intervals and analyzed using a UV spectrophotometer (Shimadzu-1700).

Drug Entrapment Efficiency[10]

The percentage of incorporated Ibuprofen (entrapment efficiency) was determined spectrophotometrically at 221 nm. After centrifugation of the aqueous suspension, amount of the free drug was detected in the supernatant and the amount of incorporated drug was determined as the result of the initial drug minus the free drug. The entrapment efficiency can be calculated using the following formula:

$$\text{Entrapment efficiency (EE\%)} = \frac{Wt._{\text{initial drug}} - Wt._{\text{free drug}}}{Wt._{\text{initial drug}}} \times 100$$

Differential Scanning Calorimetry (DSC)

DSC scan of about 5mg, accurately weighed ibuprofen and physical mixture (lipids and surfactant) and the SLN formulation were performed by using an automatic thermal analyzer system. (DSC60 Shimadzu Corporation, Japan) Sealed and perforated aluminium pans were used in the experiments for all the samples. Temperature calibrations were performed using indium as standard. An empty pan sealed in the same way as for the sample was used as a reference. The entire samples were run at a scanning rate of 10°C/min from 25-300°C.

Lyophilization

The SLN were lyophilized using an MAC Lyophilizer. The cryoprotectant was added to the SLN liquid. The SLN liquid was frozen at a temperature of –40°C for 4 h in cold trap and lyophilized for 24 h. The freeze-dried powder was re-suspended in water and characterized.

Stability Testing Studies

The intermediate stability testing studies for SLN-3 was performed for 6 months according to the ICH Guidelines. The SLN of Ibuprofen was kept at 30 °C ± 2 °C and 65% ± 5 % Relative humidity in stability chamber. Particle size measurement, Drug entrapment and Drug release were fixed as physical parameters for stability testing.

Table 1 Composition of different SLN formulations prepared by Hot Homogenization technique.

Batch No.	[Drug] Ibuprofen (mg)	[Lipid] Stearic acid (mg)	[Surfactant] Phospholipon-80H (mg)	Dichloro-methane (ml)	Water (ml)	[Stabilizer] Tween-80 (ml)
SLN-1	300	300	300	10	100	.5
SLN-2	300	300	300	10	100	1
SLN-3	300	300	300	10	100	1.5
SLN-4	300	300	300	10	100	2
SLN-5	300	300	300	10	100	2.5
SLN-6	300	300	600	10	100	1.5
SLN-7	300	600	300	10	100	1.5
SLN-8	600	300	300	10	100	1.5
SLN-9	300	300	150	10	100	1.5
SLN-10	300	150	300	10	100	1.5
SLN-11	150	300	300	10	100	1.5
SLN-12	-	300	300	10	100	1.5

Table: 2 Parameters for stability evaluation of Ibuprofen SLN

No of month	Temp.	Particle size in nano range	Drug entrapment	Drug release up to 6 hr.
0	30°C ± 2 °C	36 volume %	80.35%	75.58%
3	30°C ± 2 °C	37 volume %	80.02%	74.78%
6	30°C ± 2 °C	35 volume %	79.85%	74.31%

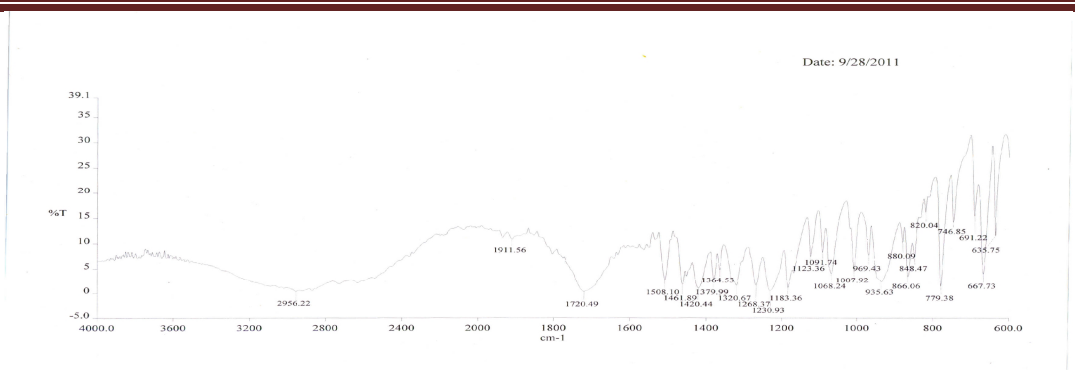

Figure 1: FTIR Spectra of pure Ibuprofen

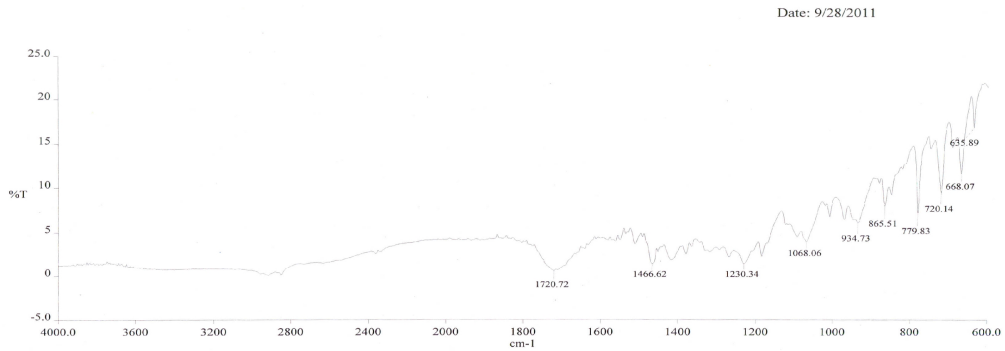

Figure 2: FT-IR Spectra of Physical Mixture of Ibuprofen with Stearic acid and Phospholipon 80H

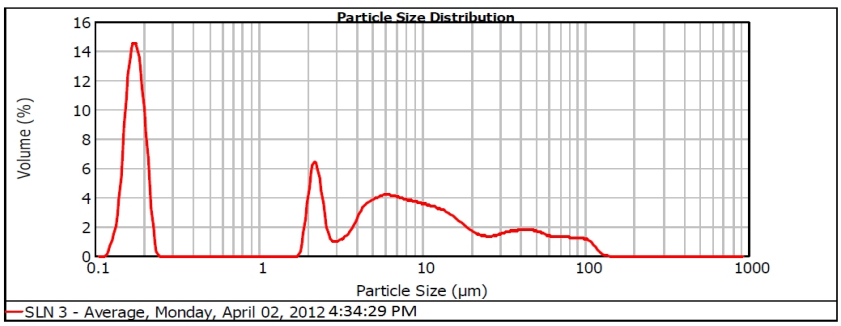

Figure 3: Particle size distribution of SLN-3 containing optimum stabilizer (Tween 1.5%)

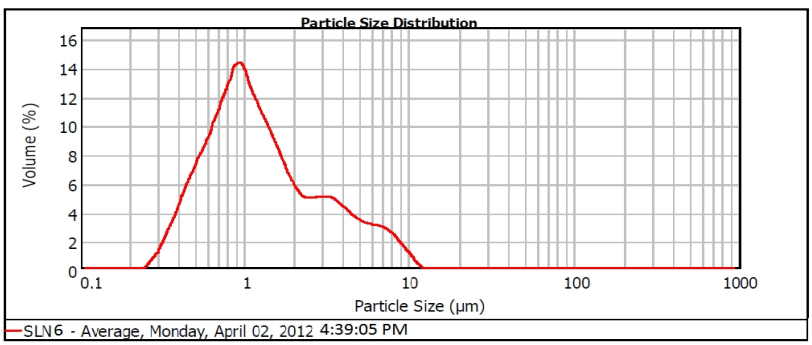

Figure 4: Particle size distribution of SLN-6 containing higher ratio of surfactant.

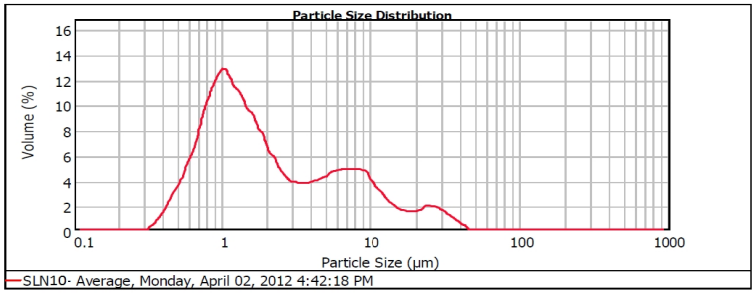

Figure 5: Particle size distribution of SLN-10 containing lower ratio of Lipid

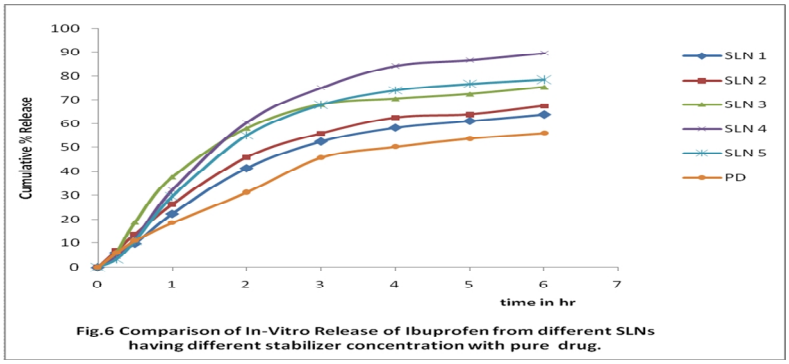

Fig.6 Comparison of In-Vitro Release of Ibuprofen from different SLNs having different stabilizer concentration with pure drug.

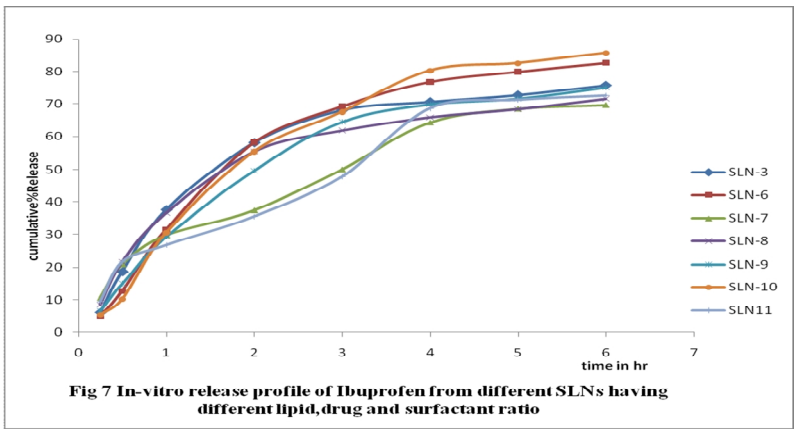

Fig 7 In-vitro release profile of Ibuprofen from different SLNs having different lipid, drug and surfactant ratio

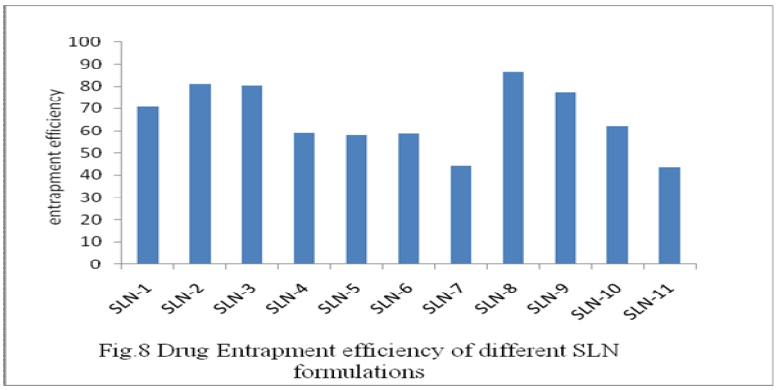

Fig.8 Drug Entrapment efficiency of different SLN formulations

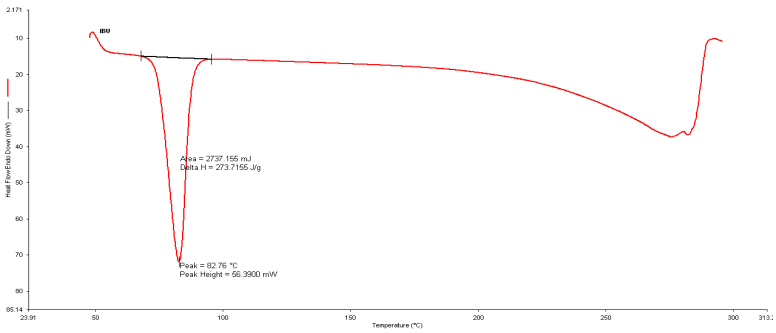

Figure 9: DSC thermogram of the pure drug Ibuprofen

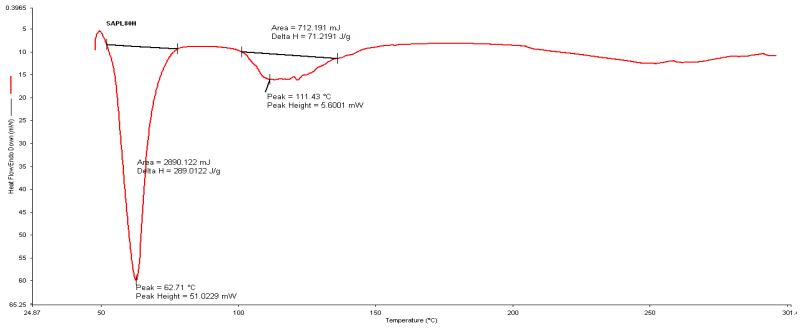

Figure 10: DSC thermogram of the physical mixture of Stearic acid and Phospholipon 80H

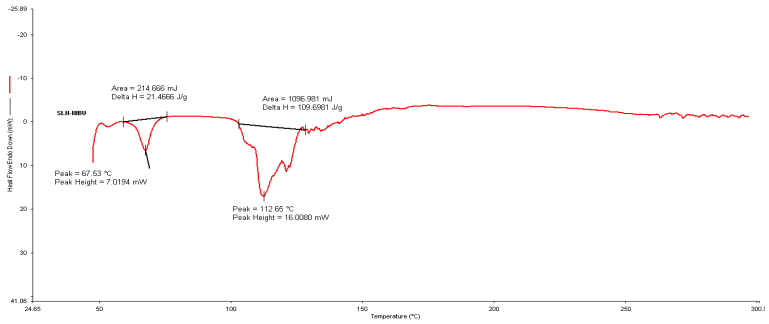

Figure 11: DSC thermogram of the Ibuprofen SLN

RESULTS AND DISCUSSION

Compatibility study by FT-IR

The FTIR Spectra of pure Ibuprofen and the physical mixture (1:1:1) of drug with Stearic acid and Phospholipon80H given in the Fig.1 and Fig.2 respectively .The IR spectra of pure drug shows principle peaks at 1721 cm^{-1} (C=O stretching vibration of –COOH group), 870, 779 cm^{-1} (Aromatic stretching bending vibration). The physical mixture on the other hand shows peaks at 1720.72, 865.51 and 779.83 cm^{-1}. Thus, it is concluded that the physical mixture of the drug, Ibuprofen does not show any major interactions with the formulation components like lipid (Stearic acid) and surfactant Phospholipon80H.

Particle Size determination:

Particle size of the solid lipid nanoparticles was analyzed by laser diffractometry using a Mastersizer 2000 instrument (Malvern) equipped with a Hydro 2000MU (A) dispersing unit. The particle size distribution of SLN-3 showed [Fig. 3] that about 36% volume of the particles are in nano range (below 1μ) and the rest of the particles are in between 1 to 100μ . Further modification in the formulation SLN-6 (increase in surfactant ratio) cause higher volume % of particle fall in nano size (56 volume %) as shown in Fig. 4. but when the lipid ratio is increased in SLN-10 the particle size distribution becomes less in nano range only 27% volume in Fig. 5

In-vitro drug release study:-

In-vitro drug release study of ibuprofen from various SLN formulations was done by using dialysis membrane. Cumulative % drug release when plotted against time for pure drug (PD) and formulation SLN- 1 to SLN-5 with different stabilizer concentration (0.5-2.5%) in the [Fig. 6] it has been found that from the SLN formulations the drug release is much higher than the pure drug and SLN-3 with the stabilizer concentration 1.5% gives 75.58% drug release up to 6 hr period with higher dissolution rate in the initial period than other four formulations. Hence 1.5% stabilizer concentration (Tween 80) is considered as an optimum concentration for this type of formulations. Keeping the stabilizer concentration (1.5%) fixed in the formulations SLN-6 to SLN-11 drug lipid and surfactant ratio is changed. The formulation SLN-12 is prepared without the drug which is also studied for diffusion and the samples were taken as blank. Cumulative % drug release from SLN-6 to SLN-11 is compared against the optimum formulation SLN-3 and plotted against time [Fig. No 7]. The effect of surfactant concentration was studied by changing the ratio of surfactant (Drug:Lipid:Surfactant=1:1:1 in SLN-3) in the SLN-6 (Drug:Lipid:Surfactant=1:1:2) and SLN-9 (Drug:Lipid:Surfactant=1:1:0.5). There is slight enhancement in the release (82.56) with increasing the surfactant (SLN-6) and also the release is decreased (74.87%) with decrease in concentration of surfactant (SLN-9). The effect of drug loading is found in SLN-8 (Drug:Lipid:Surfactant=2:1:1) and in SLN-11 (Drug:Lipid:Surfactant=0.5:1:1). Change in drug loading cause no further increase in release of drug. Lipid ratio is changed in SLN-7 (Drug:Lipid:Surfactant=1:2:1) and SLN-10 (Drug:Lipid:Surfactant=1:0.5:1). Increase in the lipid ratio in SLN-7 cause decrease (69.58%) in the release of drug.

Drug Entrapment Efficiency: The percentage of entrapped Ibuprofen in different SLN formulations with different drug, lipid, surfactant and stabilizer ratio was found spectrophotometrically. The results are given in the following Fig.8

Highest entrapment 86.32% is found in SLN-8 (Drug: Lipid: Surfactant=2:1:1) and the lowest 43.3% is found in SLN-11 (Drug: Lipid: Surfactant=0.5:1:1). Hence increase in drug loading increase the percentage of drug entrapped and decrease in drug loading cause decrease in entrapment efficiency.

Differential Scanning Calorimetry (DSC)

DSC thermogram of pure drug, Ibuprofen (Fig.9) exhibits a sharp endothermic peak at 82.76°C. The DSC curve for physical mixture of lipid (Stearic acid) and the surfactant (phospholipion 80 H) showed the presence of endothermic peaks at about 62.71°C and 111.43°C (Fig.10). On the other

hand, the SLN thermogram of Ibuprofen (Fig.11) displayed complete disappearance of characteristic peak Ibuprofen, but it shows both the characteristic peaks of lipid (Stearic acid) and the surfactant (phospholipion 80 H) at 67.53°C and 112.65°C with minimum shifting. The disappearance of the characteristic peak of Ibuprofen is due to the fact that the drug is molecularly dispersed within the lipid matrix as found for rizatriptan by Rahul Nair et.al. [10].

Lyophilization

In recent years, lyophilization has been widely used to improve the chemical and physical stability of SLN over a long period of time. However, it may destroy the surfactant film around the nanoparticles due to a "freezeout" effect, and lead to particle aggregation during the resolubilization or redispersion process. The polymers could not provide a sufficient protective effect. The mean particle size increased significantly after lyophilization without any cryoprotectant, so effective cryoprotectants should be chosen to avoid these problems associated with lyophilization. There are many kinds of cryoprotective agents that can be used for lyophilization, such as sorbitol, glucose, fructose, mannose, mannitol, maltose, dextran, and trehalose. The most effective cryoprotectant in all is mannitol. It could prevent the nanoparticles from aggregating effectively during the lyophilization process. Mannitol could form a film around the surface of the nanoparticles, which prevents nanoparticles from aggregating, so there was no significant change in particle size before and after lyophilization. SLN was re-dispersed in aqueous media.

Stability Testing Studies

The intermediate stability testing studies for SLN-3 was performed for 6 months according to the ICH Guidelines. The SLN-3 of Ibuprofen was kept at 30 °C ± 2 °C and 65% ± 5 % Relative Humidity in stability chamber. Particle size measurement, Drug entrapment and Drug release were fixed as physical parameters for stability testing. The results are given in Table 2.

After performing the stability study it was observed that at intermediate stability testing conditions there was no change in the particle size distribution, drug entrapment and drug release of the optimized formulation. At zero days the cumulative % drug release was found to be 75.58% and after three months it was 74.78% and after six months the release comes to 74.31%. This change in drug release is very negligible. The particle size which generally increases with the age of formulation is also observed with very little change. So from this study we can conclude that the prepared SLN of ibuprofen will be stable during storage.

CONCLUSION

Solid Lipid Nanoparticles of Ibuprofen, a poorly water soluble model drug is prepared by hot homogenization technique using stearic acid (lipid) Phospholipon 80 H (surfactant) and Tween-80 as stabilizer. The preformulation study shows the compatibility (FTIR study) of Ibuprofen with the other formulation ingredients and the drug is molecularly dispersed (DSC study) into the lipid. The particle size determinations confirm the particle size distribution in the nanoparticular range. In-vitro drug release through the dialysis membrane from the prepared SLNs is much higher than the pure drug. The stability study indicates the stability of the formulations without changing its performance on storage. Hence formulation of Ibuprofen in SLN enhances the dissolution rate as well as it will enhance the bioavailability of the drug which could be stabilized during storage.

ACKNOWLEDGMENT:

The authors are thankful to the management of Dr. B.C. Roy College of Pharmacy and AHS for providing the necessary facilities for carrying out the research work and Lipoid (Ludwigshafen, Germany) for providing Phospholipon 80 H.

REFERENCES

1. Sakaeda T, Okamura N, Nagata S, Yagami T, Horinouchi M, et al. Molecular and pharmacokinetic properties of 222 commercially available oral drugs in humans. Biol Pharm Bull 2001;24: 935-940.
2. Vieth M, Siegel MG, Higgs RE, Watson IA, Robertson DH, et al. Characteristic physical properties and structural fragments of marketed oral drugs. J Med Chem 2004;47: 224-232.
3. Wenlock MC, Austin RP, Barton P, Davis AM, Leeson PD. A comparison of physiochemical property profiles of development and marketed oral drugs. J Med Chem 2003;46: 1250-1256
4. Vieth M, Siegel MG, Higgs RE, Watson IA, Robertson DH, et al. Characteristic physical properties and structural fragments of marketed oral drugs. J Med Chem 2004;47: 224-232
5. Nagi A, Abdullah R, Ibrahim S, Bustamam A. Tamoxifen Drug Loading Solid Lipid Nanoparticles Prepared by Hot High Pressure Homogenization Techniques. Journal of Pharmacology and Toxicology 2008;3 (3): 219-224
6. MacGregor KJ, Embl eton JK, Lacy JE, et al. Influence of lipolysis on drug absorption from the gastro-intestinal tract. Adv Drug Deliver Rev. 1997; 25: 33-46.
7. Mukherjee S, Ray S , Thakur R.S ; Solid Lipid Nanoparticles: A Modern Formulation Approach in Drug Delivery System ; Indian Journal of Pharmaceutical Sciences, July-August 2009;349-357
8. Muller RH, Lucks JS, Arzneistofftra"geravs festen Lipidteilchen, Feste Lipidnanospha"ren (SLN), European Patent No. EP 0605497(1996)
9. Kamboj S, Bala S, Nair AB Solid Lipid Nanoparticles: An effective lipid based technology for poorly water soluble drugs International Journal of Pharmaceutical Sciences Review and Research. 2010;5(2)
10. Rahul Nair, K.S.Arun Kumar, K.Vishnu priya, T.Md.Badivaddin, Sevukarajan M Preparation and Characterization of Rizatriptan Loaded Solid Lipid Nanoparticles J Biomed Sci and Res.,2011;3(2):392-396
11. Nagi A. ALHaj, Rasedee Abdullah, Siddig Ibrahim and Ahmad Bustamam, Tamoxifen Drug Loading Solid Lipid Nanoparticles Prepared by Hot High Pressure Homogenization Techniques, American Journal of Pharmacology and Toxicology 2008;3 (3): 219-224

Source of support: Nil, Conflict of interest: None Declared