DOI: 10.36347/sjams.2017.v05i01.052

Scholars Journal of Applied Medical Sciences (SJAMS) **ISSN 2320-6691 (Online)**
Sch. J. App. Med. Sci., 2017; 5(1D):267-272 **ISSN 2347-954X (Print)**

(An International Publisher for Academic and Scientific Resources)
www.saspublishers.com

Original Research Article

Formulation and Design of Microparticulate Drug Delivery System of Lamivudine by Chitosan as Natural Polymer

Harekrishna Roy[1*], Bhabani Shankar Nayak[2]
[1]Research Scholar, Biju Patnaik University of Technology, Rourkela, Odisha, India
[2]Department of Pharmaceutics, Jeypore College of Pharmacy, Rondapalli, Koraput, Odisha, India

***Corresponding author**
Harekrishna Roy
Email: hareroy@gmail.com

Abstract: The objective of work is to formulate Lamivudine loaded microparticle by counterion elicited aggregation methodology. Natural polymer such as chitosan was chosen as polycation and smaller molecular electrolytes like sodium citrate, sodium sulphate and sodium orthophosphate were chosen as polyanions. The resulted aggregate microparticles were subjected to surface morphology, size distribution, in-vitro unharness and drug excipient interaction study. Sodium citrate (SC), sodium sulphate (SS) and sodium tripolyphosphate (TPP) were able to kind aggregates, as chitosan forms complexes and depends on pH and pKa of medium. The share of entrapped drug was additional in SC primarily based microparticle as compared to SS. The SS and SC microparticles had average particle size of three hundred metric linear unit and 1250 µm severally. Also, the SEM study unconcealed additional rough and ridges on surface of SC particle as compared to SS. the upper correlation coefficient (r^2) was found with Higuchi's equation for all formulations and formulation SC2 had bigger r^2 worth of 0.993 compared to any or all and obeyed fickian diffusion. There was no such major interaction were found throughout FTIR and DSC study. Additionally, stability study was performed and information showed no important amendment in assay worth for SC2. Furthermore change in crystalinity was observed by XRD study. The microparticles ready by above mentioned methodology had comfortable mechanical strength and were able to discharged drug for fifteen hours.
Keywords: microparticle, sodium citrate, sodium sulphate, Lamivudine, Chitosan

INTRODUCTION

A viral infection occurs when a virus enters the body through such processes as breathing air contaminated with a virus, eating contaminated food, or by having sexual contact with a person who is infected with a virus. A viral infection may also be caused by an insect bite. In a viral infection, the virus invades the inside of the body's cells in order to reproduce. A virus then spreads to other cells and repeats the process [1]. However, some people are at risk for developing serious complications of viral infection. In addition, certain types of viral infections, such as HIV/AIDS, are not self limiting and cause serious complications and are eventually fatal. In India, the number of HIV positive cases up to 2006 was 5.7 million. Between 5-7%adult are infected in at least in 10 urban areas, including Mumbai, Kolhapur, Pune, Hyderabad, Churachandrapura and Kohima. There are numerous dosage forms available as controlled drug delivery system.

Microparticulate drug delivery systems are most promising strategy based system for controlled drug delivery. They are the most reliable controlled drug delivery system and could be employed as oral drug delivery system or implantable devices.

Lamivudine belongs to a class (group) of HIV drugs called nucleoside reverse transcriptase inhibitors (NRTIs). NRTIs block an HIV enzyme called reverse transcriptase. (An enzyme is a protein that starts or increases the speed of a chemical reaction.) By blocking reverse transcriptase, NRTIs prevent HIV from multiplying and can reduce the amount of HIV in the body [2-4].

MATERIALS AND METHODS

Materials

Lamivudine was obtained as Gift sample from Cipla Ltd, Sikkim. Chitosan, HPMC (K4M, K15M), Sodium sulphate, Sodium citrate, Sodium

Tripolyphosphate were purchased from loba Chemicals, India. All other chemicals were of analytical grade.

Preparation of microparticles [5, 6]

Preparation of Drug Polymer Mixture

Accurately weighed quantity of chitosan was dissolved in 2% acetic acid solution. Then drug was added and dissolved in it. The mixture was stirred until homogeneous mixture was formed. *Preparation of 20% Salt Solution*: Accurately weighed quantity (20gm) of Salt (Sodium Citrate / Sodium Sulphate / Sodium Tripolyphosphate dissolved in 100ml of water by continuous stirring in order to get clear solution.

Preparation of Microparticles

The prepared Drug polymer mixture was injected into the salt solutions with the help of needle (24G). Microparticles were formed. The excess salt solution was removed and placed in the hot air oven at 35-40^{0}C for 72 hrs [7, 8].

Characterization of microparticles

Particle size

The particle size of all the batches of the formulated microparticles in a sample were measured with an optical micrometer fitted with a calibrated eye piece. Calibration of the microscope was done prior to particle size measurement of the microparticles. The mean of 100 particles was noted as particle size [9, 10].

Particle morphology characterization

Scanning electron microscope (SEM; S-3700N Hitachi high-technologies Europe) was used to characterize the external and internal morphology of microparticles.

Drug loading

Drug loading and demurrer potency of the ZDV loaded microparticles were determined by following methodology. Suspension of the assorted formulations was ready by suspending microparticles (equivalent to a hundred mg of pure Lamivudine) in solution. every suspension was sonicated for forty min to separate the free drug within the supernatant from the drug incorporated within the microparticles. The content of Lamivudine within the supernatant was analysed by UV photometer at 271 nm (Elico 4100) once appropriate dilution. the number of the drug incorporated in microparticles was calculated from the distinction in drug concentrations between the supernatant and therefore the original given concentrations. The demurrer potency was calculated in line with the subsequent equation.

Differential Scanning Calorimetry

Differential Scanning Calorimetry (DSC; DSC-60 Shimadzu Carporation, Japan) was performed after hermetically sealing the samples in flat bottom aluminium pans. Calibration was carried out using indium. The scanning was carried out at a temperature ranging from 40^{0}C to 300^{0}C at a rate of 20^{0}C/min under nitrogen purging.

Fourier transforms infrared spectroscopic analysis

The FTIR spectroscopic analysis (Shimadzu spectrometer, 8401S, Japan) of pure drug, individual polymers, their physical mixture and microparticles was carried out using KBr disk with hydrostatic press at a force of 5.2 cm^{-2}

In vitro drug release

The *in vitro* release of drug from the microparticles was carried out in basket type dissolution tester USP XXIII, Lab INDIA, with auto sampler containing 500 ml of 0.1N HCl for the first 2 hrs followed by next 15 hrs in phosphate buffer pH 6.8. The volume of the dissolution media was maintained at 900 ml with constant stirring (70 rpm) and temperature of bath was maintained at 37 ± 0.5°C. Aliquots (15 ml) of dissolution media were sampled at specified time intervals and replaced with fresh media immediately after sampling. Samples were analyzed for drug content by UV visible spectroscopy (Shimadzu UV 1601).The release data obtained were fitted into various mathematical models. Dissolution studies were carried out for all the batches of the prepared formulations [12-14].

RESULTS AND DISCUSSION

Lamivudine loaded microparticles were prepared by counter ion induced gelation/aggregation method using sodium citrate (SC), sodium sulphate (SS), Sodium tripolyphosphate. Physical appearance of SC based microparticles were brown colour particles with regular shape and rough surface, where as SS microparticles were light brown colour with flat surfaced particles STP based particles were of white coloured round particles. Since, chitosan has pka value of 6.5 and bears positive charge under low pH only, associates with opposite charge anion and forms ionic complexes as aggregates. The pH of the medium had a major effect on swelling of chitosan microparticles due to the ionization of both the poly anions (SC, SS and TPP) and chitosan. The more tightly cross-linked chitosan matrix does not swell (lower water uptake) as much as the loosely cross-linked chitosan matrix. In sodium citrate & sodium sulphate cross linking solutions swelling is very less due to poly anions tightly linked with chitosan whereas in sodium tri polyphosphate, the chitosan network is loose and has a high hydrodynamic free volume to accommodate more solvent molecules, thereby inducing chitosan–TPP matrix swelling [15,16].

Drug content

The first issue arises due to the existence of charge repulsion between cationic chitosan polymer (pKa = 6.5) and positively charged Lamivudine HCl (pKa = 8.6). In an attempt to enhance encapsulation, three different kinds of polyanions (cross linking agents) were employed. The encapsulation of drug was particularly dependent on the ionic interaction formed between chitosan and polyanions (Sodium citrate, Sodium sulphate & Sodium TPP), the amount of negatively charged groups of Sodium citrate, Sodium sulphate & Sodium TPP existed per mole was critical for Lamivudine association. Sodium citrate, Sodium sulphate and Sodium TPP formulations were showed drug content decreased with increase polymer concentration. In three polyanionic solutions more amount of drug entrapped in Sodium TPP then after Sodium citrate and finally Sodium sulphate [7, 8]. Drug Content decreased in the order of *Sodium TPP* > *Sodium Citrate* > *Sodium Sulphate*

***In vitro* drug release study**

Different types of microparticles were prepared by constant drug with varying chitosan concentration i.e. Drug: Chitosan. The release behaviours of chitosan microspheres produced by ionic gelation with 20 % (w/v) of different cross-linking agents such as Sodium citrate, sodium sulphate, sodium tripolyphosphate (poly anionic) salt solutions.

Sodium Citrate solution

First four formulations (SC1, SC2, SC3 and SC4) were ready by victimization sodium citrate cross-linking agent. Among 2hrs quite 45% (SC1-51.76, SC2-60.19, SC3-70.16 and SC4-81.25) of loaded NRTI complex discharged from all chitosan microparticles in zero.1N HCl. once 2hrs microparticles were transferred into phosphate buffer of pH scale 6.8. *Sodium sulfate solution*: Next four formulations (SS1, SS2, SS3 and SS4) were ready by victimization sodium sulfate cross-linking agent. However SS1 have low particle strength, they're simply broken and not forming microparticles. Formation of chitosan microparticles by crosslinking agents depends on pH scale of medium & charged cluster concentration of the fluid to be treated. Ionic strength doesn't have an effect on the advanced formation method. Remaining three formulations among 2hrs more than 55% (SS1- 48.12, SS2-53.19, SS3-59.46 and SS4-63.17) of loaded NRTI complex discharged from all chitosan microparticles in zero.1N HCl. Last four formulations (TPP1, TPP2, TPP3 and TPP4) were ready by victimization detergent builder cross-linking agent. Among 2hrs over hour (TPP1-70.25, TPP2-72.14, TPP3-78.68 and TPP4-82.37) of loaded NRTI coordination compound discharged from all chitosan microparticles in zero.1N HCl. Once 2hrs microparticles were transferred into phosphate buffer of pH scale 6.8. These unharness patterns continuing in phosphate buffer upto 15hrs.

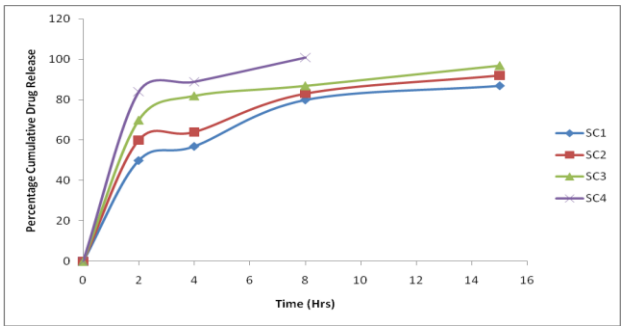

Fig-1: Invitro Drug release study of formulation SC1 to SC4

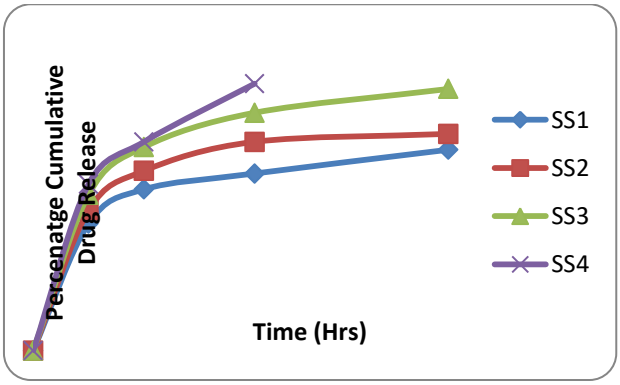

Fig-2: Invitro Drug release study of formulation SS1 to SS4

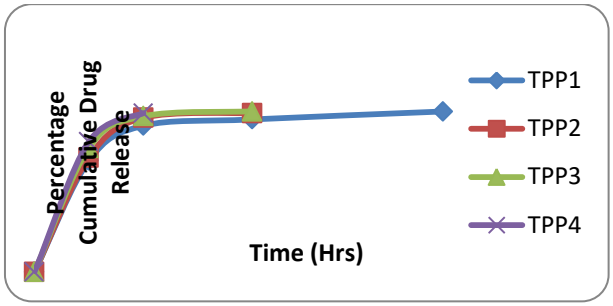

Fig-3: Invitro Drug release study of formulation TPP1 to TPP4

Scanning electron microscopy

Microparticles by Sodium citrate were regular (not spherical) in shape and had a rough surface in all formulations. Whereas, microparticles by Sodium sulphate solution have larger particles, flat and rigid structures in all formulations. The average particle size of microparticles found to be lies between 1100-1300 µm. Fine and regular along with porous and rough surface structure was observed in case of Sodium TPP formulations. The change in surface morphology and fluffy physical state of formulations may also contribute to the enhanced solubility and dissolution rate of drug from the formulations.

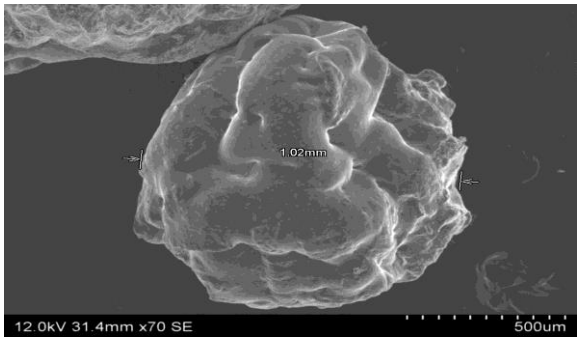

Fig-4: SEM of Selected microparticle

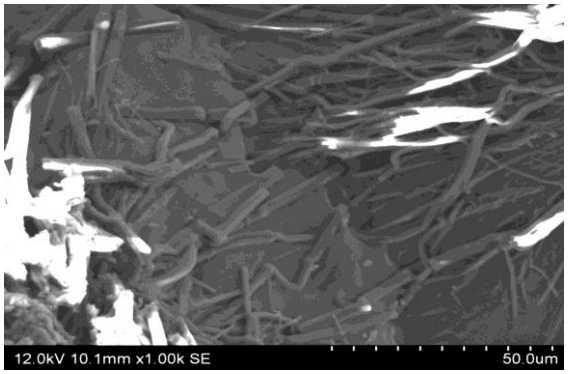

Fig-5: Internal Observation of TPP based formulation

FT-IR Study

Peaks were recorded during FTIR study, there was no such interaction observed between drug and excipients.

Differential scanning calorimetry (DSC):

Differential scanning colorimetry studies were performed to assess the compatibility between drug and excipients by endothermal peak. Pure drug showed endothermal peak at 178.62 _C and onset of peak was at 146.26 _C. DSC study showed no change in endothermal peak in SS2.

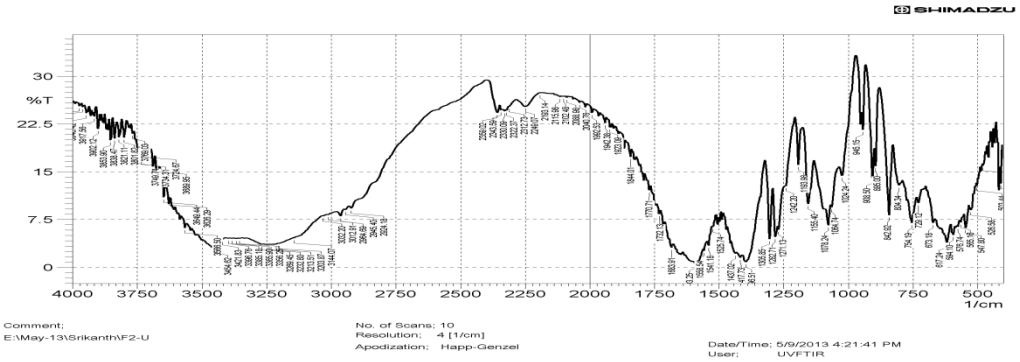

Fig-6: FTIR of Selected formulation mixture

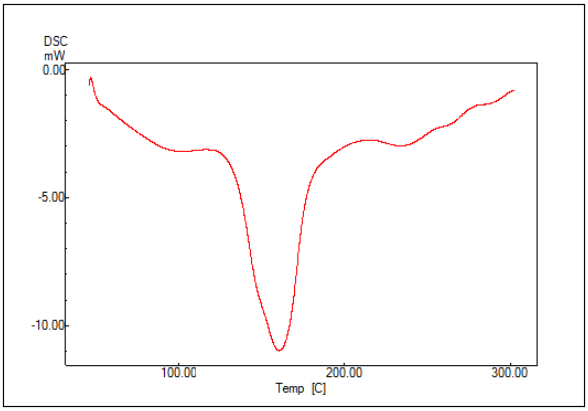

Fig-7: DSC Result of Drug and Mixture

X-Ray Diffraction Study

Change in crystallinity of drug its individual components were analysed by the help of XRD 7000, Shimadzu. Below figure illustrated the comparative x-ray powder diffraction pattern of mixture of drug and polymer. Pure drug showed the classical diffractogram of the crystalline substance. The XRD pattern of the drug loaded chitosan microparticles indicates the presence of drug in the amorphous state. The reason may be the interaction of drug and solvents used during processing.

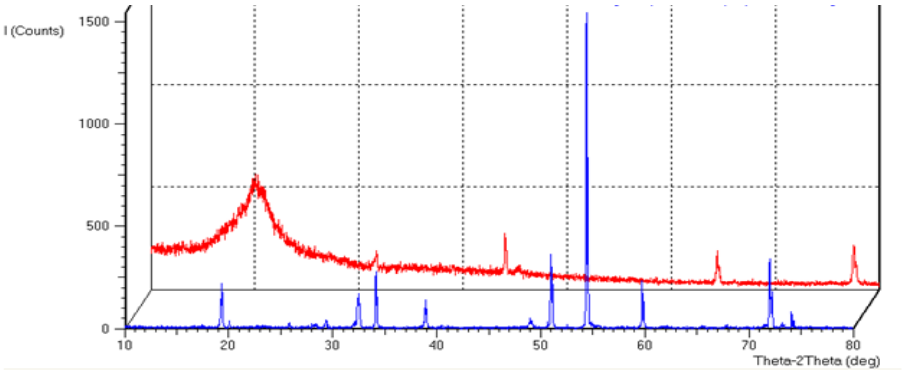

Fig-8: XRD of Drug and polymer mixture.

CONCLUSION

During the study an attempt has been made for microparticles drug delivery system of Lamivudine by counterion induced aggregation method. During this study chitosan as natural polymer was choosen and formed complexes with opposite charged ions provided by SC, SS and TPP. TPP formulations showed

promising result in drug content as well as release manner. Furthermore *in-vivo* study also has to carry out.

REFERENCES

1. NL Prashanti, Harekrishna Roy, N. Jyothi, V. Sri Vajrapriya. A Brief Review on Chitosan and Application in Biomedical Field. American Journal of Pharmatech Research. 2016; 6(4): 41-51.
2. https://aidsinfo.nih.gov/drugs/126/lamivudine/0/patient. Accessed on 16 Sept 2016.
3. https://en.wikipedia.org/wiki/Lamivudine. Accessed on 22 July 2016.
4. V. Sri Vajrapriya, Harekrishna Roy, NL Prashanti, N. Jyothi. Polymers In Drug Delivery Technology, Types Of Polymers And Applications. Scholars Academy Journal of Pharmacy. 2016; 5(7): 305-308.
5. Harekrishna Roy, P. Venkateswar Rao, Sanjay Kumar Panda, Asim Kumar Biswal, Kirti Ranjan Parida, Jharana Dash. Composite alginate hydrogel microparticulate delivery system of zidovudine hydrochloride based on counter ion induced aggregation. Int J Applied Basic Med Res. 2014; 4(Sup 1): S31-36.
6. Pradip Kumar Dutta, JoydeepDutta and V S Tripathi. Chitin and chitosan: Chemistry, properties and applications. Journal of Scientific & Industrial Research. 2004; 63: 20-31.
7. Harekrishna Roy, Chandan Kumar Brahma, Ravi Kumar, Sisir Nandi. Formulation of saquinavir mesylate loaded microparticle by counterion induced aggregation method: Approach by hyperosmotic technique. Drug Invention Today. 2013; 5:259-266.
8. GavhaneYogeshkumar N, GuravAtul S, YadavAdhikrao V. Chitosan and Its Applications: A Review of Literature. International Journal of Research in Pharmaceutical and Biomedical Sciences. 2013; 4 (1): 312-331.
9. N. Jyothi, Harekrishna Roy, N. Lakshmi Prasanhti and V. Sri Vajrapriya. A Brief Review of Microparticle Drug Delivery System. World Journal of Pharmacy and Pharmaceutical Sciences. 2016; 5(7): 701-712.
10. Vaghari et al. Recent advances in application of chitosan in fuel cells. Sustainable Chemical Processes. 2013; 1(16): 1-12.
11. Harekrishna Roy. Formulation of Sustained Release Matrix Tablets of Metformin hydrochloride by Polyacrylate Polymer. . Int J Pharma Res Health Sci. 2015; 3(6): 900-906.
12. Roy H, Brahma CK, Nandi S, Parida K. Formulation and design of sustained release matrix tablets of metformin hydrochloride: Influence of hypromellose and polyacrylate polymers. Int J Appl Basic Med Res. 2013; 3: 55-63.
13. Harekrishna Roy, Sanjay Kumar Panda, KirtiRanjanParida, Asim Kumar Biswal. Formulation and In-vitro Evaluation of Matrix Controlled Lamivudine Tablets**.** Int J Pharma Res Health Sci. 2013; 1(1): 1-7.
14. Harekrishna Roy ,Anup K Chakraborty ,Bhabani Shankar Nayak, Satyabrata Bhanja, Sruti Ranjan Mishra, P. Ellaiah. Design and in vitro evaluation of sustained release matrix tablets of complexed Nicardipine Hydrochloride; International Journal of Pharmacy and Pharmaceutical Sciences. 2010, 2, 182-132.
15. https://en.wikipedia.org/wiki/Citrate. Accessed on 13 Sept 2016.
16. Denkbas EB, Ottenbrite RM. Perspectives on: chitosan drug delivery systems based on their geometries. J Bioact Compat Polym. 2006; 21:351-368.
17. Genta I, Perugini P, Conti B, Pabanetto F. A multiple emulsion method to entrap a lipophilic compound into chitosan microspheres. Int J Pharm. 1997; 152:237-246.
18. Shen EC, Wang C, Fu E, Chiang CY, Chen TT, Nieh S. Tetracycline release from tripolyphosphate-chitosan crosslinked sponge: a preliminary in vitro study. J Periodontal Res. 2008; 43:642-648.