Research Article

Preparation and Evaluation of Chitosan Microsphere of Metformin Hydrochloride and to Study the Effect of Drug to Polymer Ratio

A. Semalty

Department of Pharmaceutical Sciences, H.N.B Garhwal (Central) University, Srinagar (Garhwal)-Uttarakhand, India.

ABSTRACT

Metformin hydrochloride is biguanide used as oral hypoglycemic agent. But its shows short biological half life ($t_{1/2}$ 3-4) which leads to high dosage frequency. Therefore, the present study aims to prepare the controlled release formulation of metformin hydrochloride loaded in the chitosan microspheres. It also involves the study of the effect drug to polymer ratio on the performance of chitosan microspheres. Chitosan microspheres were prepared by Solvent Extrusion Method using citric acid as a crosslinking agent. Two formulations with different drug polymer ratios (M1 high drug-polymer ratio and M2 low drug to polymer ratio) were prepared for their effect on performance. Formulated microspheres were characterized for its drug content, compressibility index, swelling index, surface morphology and particle size (SEM) and *in- vitro* drug release study. The characterization of fabricated microsphere showed smooth cracked to smooth porous surface with narrow particle range (600-800µm) and high drug content (75.83 and 73 % for M1 and M2, respectively). Compressibility index was founded between 10.6 and 5.53 % for M1 and M2, respectively. Both the formulations showed good percent swelling index. SEM showed nonporous, smooth and cracky surface for both the formulations. The microspheres prepared with high drug to polymer ratio showed higher in vitro drug release at the end of 12 h of the study. It was concluded that the chitosan microspheres could be considered for controlled drug delivery of metformin hydrochloride. The microspheres prepared with high drug to polymer ratio showed good drug content, compressibility index, swelling index, surface morphology and greater *in- vitro* drug release.

Keywords: Metformin Hydrochloride, Chitosan, microspheres, Crosslinking Agent (Citric Acid).

INTRODUCTION

Novel Drug Delivery System (NDDS) are the advanced dosage form of APIs. NDDS are capable to improve the performance of APIs in term of drug utilization, absorption, bioavailability, targeting, safety, stability (in biological environment) and the patient compliance. It is advance drug delivery system which improves drug utilization, control drug release to give a sustained therapeutic effect, provide greater safety, and target a drug specifically to a desired tissue. Oral Drug Delivery is the most convenient route of administration. Most of the pharmaceutical preparations are given by this route. The design of oral sustained drug delivery system is primarily aims to achieve more predictable and increased bioavailability. Various methods are available for modifying release profiles with respect to site of action and time of release. Microspheres are one of the approaches widely used as drug carriers. Microspheres simply can be defined as solid, approximately spherical particles ranging from 1to 1000µm[1-4].

Chitosan is biodegradable modified natural carbohydrate polymer (polysaccharide) derived from chitin, which occurs predominantly in animals of arthropods and marine crustaceans. Chitosan has great pharmaceutical application because of its biocompatibility, high charge density, and nontoxic in nature. Chitosan is used because of its property to improve the solubility of poorly water soluble drugs as well as to control the release of drugs by slow erosion from hydrated matrix [5-7].

Among crosslinking agents Citric acid is one of the strong to moderate absorption enhancers that cause tissue disturbance with a slow recovery of functional groups[8].

Metformin Hydrochloride is a biguanide comes under the category of oral hypoglycemic agent. Metformin HCl suppress hepatic gluconeogenesis, enhance insulin mediated glucose disposal in muscle and fat, it enhances GLUT1 (glucose transporter) transport from intracellular site to plasma membrane. It shows short biological half life about 6 hrs and 50 to 60 % bioavailbility[9].

Therefore, the present study aims to develop and characterize the chitosan microspheres of metformin hydrochloride to improve the drug release profile so as to release the drug for extended period of time for reducing the dosage frequency. The study also evaluates the effect of drug to polymer ratio on the performance of prepared microspheres.

MATERIALS AND METHODS

Metformin hydrochloride was obtained as a gift sample from Panacia Biotech, Baddi. Chitosan was purchased from Sigma Aldrich Mumbai and Sodium Alginate was purchased from Himedia Laboratory Pvt. Ltd. Mumbai. All other chemicals used were of analytical grade.

Method of preparation of microspheres of metformin hydrochloride

The microspheres were prepared by the solvent extrusion method[10-13]. In this method 1% w/v acetic acid was prepared by addition of 100 ml distilled water firstly. Then accurately weighed amount of chitosan (300mg) was dissolved in acetic acid solution resulting in 3% w/v solution of chitosan. Amount of drug weighed according to formulation ratio (150mg, 300 mg) and dissolved in chitosan solution with the help of magnetic stirrer. A 2 %w/v solution of sodium alginate was prepared by stirring it on a magnetic stirrer on 700 rpm for 4 hours and a 10 % w/v CaCl2 was prepared by dissolving it in distilled water. Alginate beads were prepared by the solvent extrusion method in CaCl2. The beads were washed by 30 ml chloroform three times and then washed by distilled water three times. Freshly prepared beads were soaked in the chitosan drug solution for 1 hr. Beads were taken out and soaked in freshly prepared citric acid cross linking agent solution (2%w/v) for 1 hr. All the microspheres were retained after 1 hr and spread over glass sheet dried in oven and then stored in a desiccator at room temperature.

Characterization of prepared microspheres

Drug Content

The drug content was calculated by taking the microspheres equivalent to 50 mg of drug. Microspheres were crushed fine and were dissolved in 0.1 N HCL diluted to 100ml. Then it was stirred on magnetic stirrer for 24 hours and filtered. At the end of 24 h sample was withdrawn, diluted suitably and measured spectrophotometrically (AU- 2701 Spectrophotometer) at 232.7 nm for the drug content.

Compressibility Index

The loose bulk density (ρb) and the tap (ρt) density of the microspheres were measured in a measuring cylinder. 1 g of prepared microcapsules was filled in 10 ml graduated cylinder. The initial volume was noted then cylinder was tapped on wooden surface. The density was measured by tapping the cylinder 100 times (from the height of one inch) at the rate of 240 taps/min. Each determination was carried out in triplicate and the densities were calculated from the mean of the three determinations. The density was calculated by using the formula: Density= Mass/Volume.
Then the Compressibility index was calculated by the formula:

$$\text{Compressibility index} = [(\rho t - \rho b)/\rho t] \times 100$$

Swelling Index

The swellability of microspheres in simulated physiological media (with respect to pH) was determined by swelling in the three different media which were (distilled water, pH1.2 acid buffer, pH6.8 phosphate buffer)[14-15].

Scanning Electron Microscopy

To detect the surface morphology and particle size of the microspheres, SEM of the microspheres was performed at Wadia Institute of Himalayan Geology, Dehradun by scanning electron microscope (Carl Zeiss SMT Evo Series).

In Vitro Drug Release Studies

In-vitro release profile of the microspheres was evaluated using Veego 8 stage USP dissolution test apparatus using 900 ml of pH 1.2 acid buffer (first dissolution media) maintained at 37±0.5°C at 50 rpm. Accurately weighed 100 mg of microspheres were placed and the dissolution was done for 2.5 hrs and at prefixed time of every 30 minute, 5 ml of solution were withdrawn. After suitably dilution, samples were assayed spectrophotometrically for the drug release at 232 nm using UV-visible Spectrophotometer. Then the dissolution media was replaced by 900 ml (pH 6.8 phosphate buffer) maintained at 37±0.5°C at 50 rpm. The study was continued till the end of 12 h. Samples were withdrawn, diluted suitably and then assayed spectrophotometrically for the drug release at 232 nm.

RESULTS AND DISCUSSION

In present work, the microspheres of Metformin hydrochloride were prepared by

solvent extrusion method by using chitosan and sodium alginate (polymers) and citric Acid (cross linking agent). The method adopted provides the advantage over conventional methods because of less or minimum use of[3] organic solvents. The prepared formulations of different drug to polymer ratio were evaluated for physical properties like particle size, bulk density, tap density, swelling index and percent drug loading.

The Percentage yield for Metformin HCl microspheres using Chitosan- sodium alginate was found to be 75 and 79.8 % for M1 and M2, respectively Drug content of both the formulations was estimated by UV spectrophotometic method. The microsphere formulations showed the percent drug content of 75.83% and 73.0 % for M1 and M2, respectively (Table 2). The physical parameters (Table 2) such as bulk density, tapping density, compressibility index were evaluated, which provided the basis for optimization of the flow property of microspheres. All the microspheres of various formulations showed good flow property. Bulk density is indicative of the package properties of the microspheres. All the microspheres of different formulations were less than 1.2g/ml. The size range of both formulations of microspheres was found to be 600 to 800µm. The order of swelling index (Table 3) for Metformin HCl microspheres formulations are below. In pH 1.2 after 2 hrs– M1>M2; In pH 6.8 after 4 hrs- M2>M1; In Distilled water after 2 hrs – M1>M2; In Distilled water after 4 hrs- M1>M2.

In the SEM study the surface of chitosan was irregular and rough while the surface of metformin was smooth slightly crystalline (Fig. 1). While the shape of M1 formulation was found slightly round with smooth cracked surface. M2 showed cracks on its surface. The nonporous nature of the surface might gave contributed in sustaining the drug release.

In vitro drug release studies of both the formulations were performed initially in Acid buffer (pH 1.2) for 2.5 h followed by phosphate buffer (pH 6.8). It was found that drug release from the formulations was significantly different for both the formulations. At the end of 12 h the drug release was found to be 86.6, 80.38 % for formulations M1 and M2, respectively. The different kinetic models for Metformin HCl Microspheres release were also studied the release versus time curves were plotted for determining the order of release. It was found that Formulation M1, M2 followed first order kinetics up to the end of 6 h and then started following the zero order kinetics.

As the polymer to drug ratio was higher (in M1) the extent of drug release increased. A significant decrease in the rate and extent of drug release is attributed to the increase in the density of polymer matrix (as in case of M2)[16-17]. Additionally, the larger particle size at higher polymer concentration also restricted the total surface area resulting in slower release.

CONCLUSION

The microspheres of metformin hydrochloride were prepared using chitosan, sodium alginate and citric acid. It was concluded that formulation M1 showed higher drug content, good flow properties (evidenced by density and compressibility index data), good surface morphology and promising drug release. Therefore it can be concluded that these microspheres can be used to sustain the drug release effectively and minimizing the dosage frequency of Metformin hydrochloride.

ACKNOWLEDGEMENT

Authors are thankful to Wadia Institute of Himalayan Geology, Dehradun, Uttarakhand for providing SEM facility.

Table 1: Formulation design for microspheres of metformin hydrochloride

Formulation	Drug (mg)	Sodium Alginate (%w/v)	Chitosan (%w/v)	Crosslinking Agent(%w/v) (citric Acid)
M_1	300	2	3	2
M_2	150	2	3	2

Table 2: Characterization of metformin hydrochloride microspheres

Formulation	Bulk Density in g/ml (ρb)	Tapped Density in g/ml (ρt)	Compressibility Index (%)	Drug Content (%)
M_1	0.210	0.235	10.6	75.83
M_2	0.222	0.235	5.53	73.0

Table 3: Percent swelling Index of metformin hydrochloride microspheres

Time (hrs)	Distilled Water		pH 1.2		pH 6.8	
	M1	M2	M1	M2	M1	M2
0.5	5.6	4.7	3.3	4.9	6.7	6.3
1	22.3	19.6	8.6	9.2	11.8	13.8
2	41.8	41.3	38.5	35.3	37.4	46.4
4	68.4	63.8	58.2	69.4	54.6	57.6

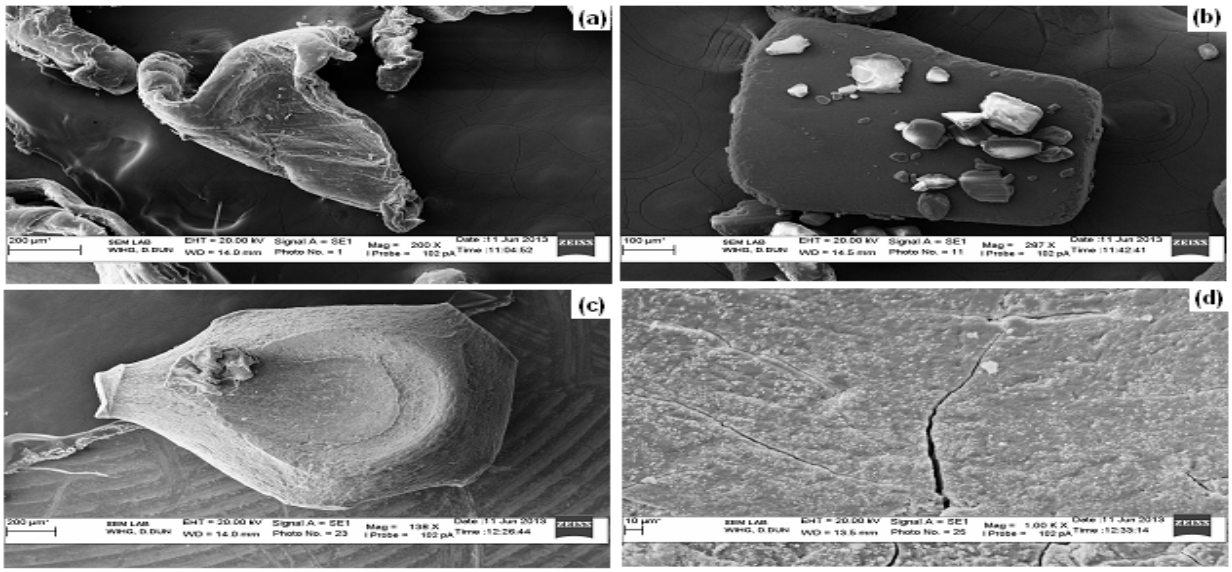

Fig. 1: SEM Photograph of (a) Chitosan; (b) Metformin HCl; (c) Formulation M_1; (d) Formulation M_2

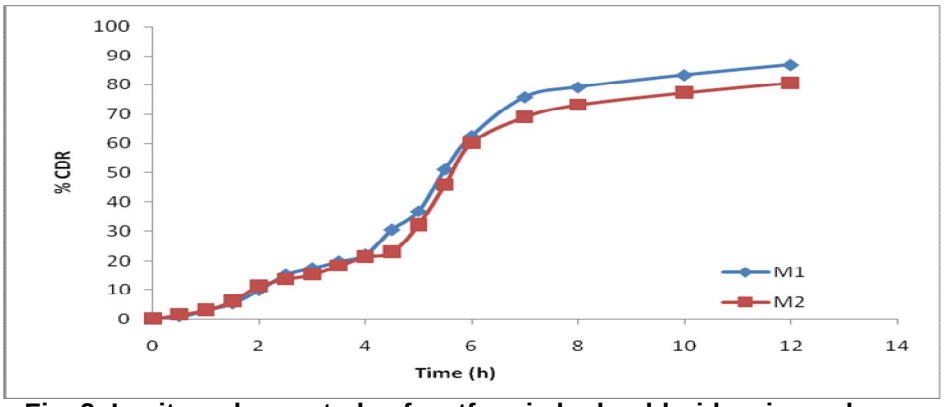

Fig. 2: In-vitro release study of metformin hydrochloride microspheres

REFERENCES

1. Lachman L, Lieberman HA and Kanig JL. The Theory and practice of industrial pharmacy. 3rd edn. Varghese Publishing House, Bombay. 1987;172-173.
2. Semalty M, Yadav S and Semalty A. Preparation and characterization of gastroretentive floating microspheres of ofloxacin hydrochloride, Int J Pharm Sci Nanotech. 2010;3:819-23.
3. Robinson J and Lee V. Controlled Drug Delivery: Fundamentals and Applications, Marcel Dekker: New York. 1978; 335-410.
4. Arul B and Kothai R. Sangmeswaran B. Jayakar B. Formulation and evaluation of Chitosan microspheres containing Isoniazid. Indian J Pharm. Sci. 2003;65:640-642.
5. Illum L. Chitosan and its use as a pharmaceutical excipient. Pharm Res. 1998;15:1326-1331.
6. Hejazi, Amiji Chitosan based gastrointestinal delivery systems. J. Control Release, 2003;89:151-165.
7. Roy S, Panpalia SG, Nandy BC, Rai VK and Tyagi LK. Effect of method of preparation on Chitosan microspheres of Mefenamic acid. Int *J Pharm Sci* Drug Res. *2009*;1:36-42.
8. Varshosaz J and Alinagri R. Effect of Citric acid as crosslinking agent on insulin loaded Chitosan microspheres. Iranian Polymer Journal. 2005;14(7):647-656.
9. Tripathi KD. Essentials of medical pharmacology. 6th edition. Jaypee Brothers Medical Publishers. 2008:267-269.
10. Builders P, Olobayo O, Kunle, Larry C, Builders I and Anthony A. Preparation and evaluation of mucinated sodium alginate microspheres for oral delivery of Insulin. European J Pharm Biopharm. 2008;70(3):777-783.
11. Mahmoud M, Saleh R, Auda S and Ibrahim M. Emulsification/internal gelation as a method for preparation of diclofenac sodium alginate microspheres. Saudi Pharma J. 2013; 61-69.
12. Anal A and Stevens W. Chitosan alginate beads for controlled release of Ampicillin. Int J Pharm. 2005; 290(1-2):45-54.
13. Shabaraya A and Narayanachrayulu R. Design and evaluation of chitosan microspheres of metoprolol tartrate for sustained release. Indian J Pharm Sci. 2003;65:250-252.
14. Bahera L, Sahoo K and Patil V. Preparation and In Vitro characterization of oral sustained release Chitosan coated Cefepime hydrochloride microspheres. Int J Pharm Tech Res. 2010;2:798-803.
15. Zhang Y and Wei W. Preparation and Evaluation of alginate Chitosan microspheres for oral delivery of insulin. European J Pharm Biopharm. 2011;77:11-19.
16. Srinatha A, Pandit J and Singh S. Ionic cross linked chitosan beads for extended release of Ciprofloxacin. Indian J Pharm Sci. 2008;70:16-21.
17. Semalty M, Verma D and Semalty A. Development and evaluation of microcapsules of Verapamil hydrochloride, Indian Drugs. 2011;48(8):33-38.