RESEARCH ARTICLE

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Preparation and evaluation of alginate microspheres of piroxicam for controlled release

Piroksikam aljinat mikrosferlerin kontrollü salınım için hazırlanması ve değerlendirilmesi

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ABSTRACT

Piroxicam is a Non-steroidal anti-inflammatory, analgesic and anti-pyretic drug which is widely used in muscle-skeletal disorder like osteoarthritis. Piroxicam had bad taste, half-life of 30 hrs and poor water solubility. Microspheres are multi-particulate drug delivery systems, spherical in shape and having a size range 50-100 microns. Methods such as Coaservation, Emulsification, Solvent evaporation and Ionic gelation methods are employed for preparing Microspheres.The present study is aimed at formulation of alginate microspheres with piroxicam by ionic gelation method. Obtained microspheres were characterized for particle size, drug content and invitro drug release. The prepared microspheres were found to be spherical, discrete and free flowing. The average size of microspheres prepared was found to be 950 m. Among 2 formulations, batch A microspheres releases piroxicam slowly and spread over a period of 22 hrs, release was by Non-Fickian diffusion mechanism as the 'n' value was 0.65.

Keywords: *Microspheres, piroxicam, ionic gelation method, sodium alginate, calcium chloride*

ÖZ

Piroksikam, osteoartrit gibi kas-iskelet sistemi hastalıklarında yaygın kullanılan steroid yapıda olmayan antiinflamatuvar analjezik ve antipiretik bir ilaçtır. Piroksikamın tadı ve suda çözünürlüğü kötü olmakla birlikte 30 saatlik yarılanma süresine sahiptir. Mikrosferler, küre şeklinde bir forma ve 500-100 mikronluk boyut aralığına sahip olan çok-partiküllü ilaç taşıyıcı sistemlerdir. Mikrosferlerin hazırlığında koaservasyon, emülsifikasyon, solvent buharlaşma ve iyonik jelasyon yöntemleri kullanılmaktadır. Bu çalışmada, iyonik jelasyon yöntemi kullanılarak piroksikam ile aljinat mikrosferlerin oluşturulması amaçlanmıştır. Elde edilen mikrosferler partikül boyutu, ilaç içeriği ve in vitro ilaç salınımı yönünden incelenmiştir. Hazırlanan mikrosferlerin küresel ve diskret olduğu ve serbest halde akabildiği tespit edilmiştir. Hazırlanan mikrosferlerin ortalama boyutu 950 m olarak bulunmuştur. İki formülasyonun içerisinde A grubu mikrosferlerin piroksikamı yavaş saldığı ve 22 saatlik bir sürede yayıldığı görülmüştür. "n" değeri 0.65 olup salınım difüzyon mekanizmasının non-Fickian olduğu bulunmuştur.

Anahtar Kelimeler: *Mikrosferler, piroksikam, iyonik jelasyon yöntemi, sodyum aljinat, kalsiyum klorür*

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INTRODUCTION

A well designed controlled drug delivery system can overcome some of the problems of conventional therapy and enhance the therapeutic efficacy of a given drug. In order to obtain maximum therapeutic efficacy, it becomes necessary to deliver the agent to the target tissue in the optimal amount in the right period of time there by causing little toxicity and minimal side effects. Various approaches are available in delivering a therapeutic substance to the target site in a sustainedcontrolled release fashion. One such approach is using microspheres as carriers for drugs. Microspheres are characteristically free flowing powders consisting of proteins or synthetic polymers which are biodegradable in nature and ideally having a particle size less than 200 µm (1,2).

Microspheres can also be defined as multi particulate drug delivery systems, spherical in shape and having a size in range 50-100 microns. These are spherical matrices composed of a polymer in which the drug is dissolved or dispersed. The Microspheres are used for controlled release of medicaments parenterally, that is, the Microspheres form one type of parenteral control release systems. The various techniques which are used for the preparation of Microspheres are: Air suspension technique (Wurster), Coacervation-Phase separation process, Spray drying & spray congealing, Pan coating, Solvent evaporation, Interfacial Polymerization,Single & double emulsion techniques and ion gelation method (3).

Polymeric microspheres as a parenteral drug delivery system are primarily developed for sustained release of drugs for prolonged systemic therapeutic effects after subcutaneous (SC) or intramuscular (IM) administration. Polymers used forformulation of microspheres are biodegradable and biocompatible. Polymers such as gelatin, albumin, carbapol and alginates are generally used in preparing the Microspheres. Methods such as coaservation, emulsification, solvent evaporation and ionic gelation methods are employed for preparing Microspheres. Polymers are substances of high molecular weight made up by repeating monomer units. These molecules may be linear or branched, and separate linear or branched chains may be joined by crosslinks. Polymers are used widely in pharmaceutical systems as adjuvants, coatingmaterials and, components of controlled and site-specific drug delivery systems (4).

Microspheres inthe finished product are in a dry powder form which is produced either as the endproduct of the manufacturing process or by the removal of the solvent/dispersion liquid medium after formation of microspheres by lyophilization or filtration with finaldrying. Prior to administration, a microsphere product is reconstituted in a liquiddiluent which can be supplied in a separate container or in the liquid compartmentof a dual-chamber prefilled syringe. While the size and size distribution of a microsphere product are two key factors that control the drug release rate and the resultant duration of sustained release, these two factors also affect the syringeability and injectabilityof the product with respect to a specific gauge size of the syringe used. The easeof reconstitution and injectability via SC and IM routes of administration of microspheresare the main advantages over implants and viscous injectable polymeric gelsas a parenteral drug delivery system. The small particle size and the dispersibility of microspheres upon injection have also made microspheres more acceptable by patientsas compared with implants which can cause discomfort during and after implantation (5).

Piroxicam is a weakly acidic and highly lipophilic anti-inflammatory drug (Figure 1) available for oral,

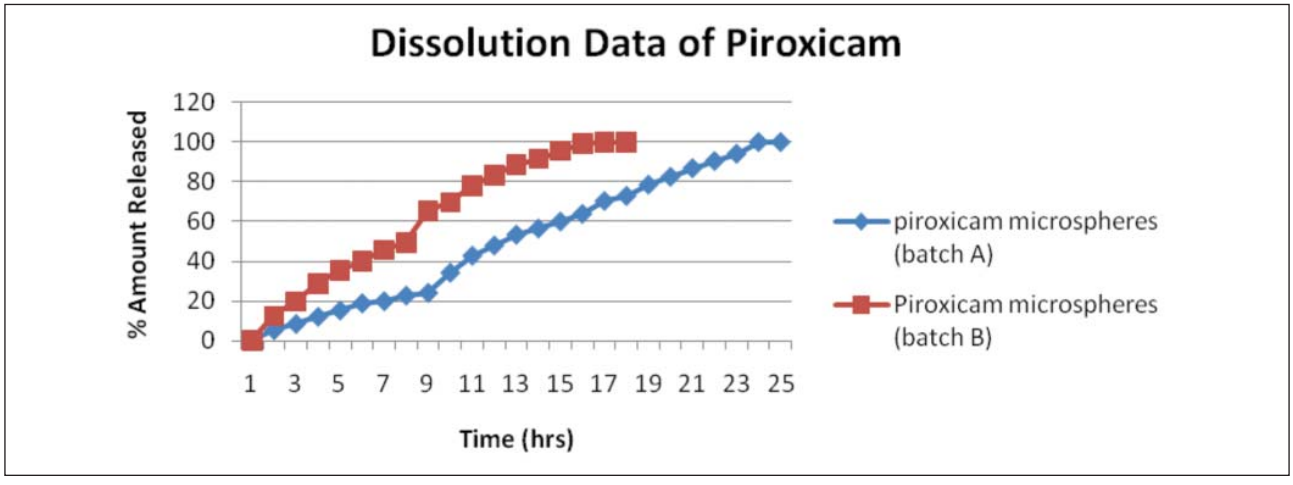

Figure 1. *Dissolution data of piroxicam microspheres.*

parenteral and topical administration. The drug inhibits the synthesis of prostaglandins in inflammation. The pharmacokinetics of piroxicam is characterized by a high oral absorption, and a long biological half-life (50-60 h), which makes possible single daily dose administration of the drug. Most of the orally administered dosage forms have several physiological limitations such asvariability in gastrointestinal time (GI) time, incomplete drug release from devices and shortresidence time of pharmaceutical dosage forms in absorption region of GI tract. This leads to low bioavailability of sustained release dosage forms and even if slow release of drug is attained, the drug release passes the absorption site and therefore lowering the efficacy of drugs. To overcome these problems several attempts have been made to develop oral dosage forms, capable of having prolonged retention time in stomach to extend the duration of drug delivery (6).

This technology has been used widely in the design of controlled release and sustained release dosage forms. It has a myriad of applications. It includes, to mask the bitter taste of drugs like paracetamol, nitrofurantoin etc. To reduce gastric and other gastro intestinal (G.I) tract irritations, for example, sustained release Aspirin preparations have been reported to cause significantly less G.I. bleeding than conventional preparations (7). There is a growing proportion of elderly patients suffering from diseases like osteoarthritis or rheumatoid arthritis. These patients require non-steroidal anti-inflammatory drugs (NSAIDS) therapy for treatment. But NSAIDS are well known for their gastrotoxic and duodenotoxic effects. Piroxicam, a non-steroidal anti-inflammatory drug exhibit better tolerance than aspirin, indomethacin and naproxen. Poly lactic acid microspheres of piroxicam have been prepared by solvent evaporation method and by spray-drying method the other application is in a scenario where a liquid can be converted to a pseudo-solid for easy handling and storage, e.g., Eprazinone (8).

The objective of the present experiment was to prepare and evaluate alginate microspheres containing piroxicam. Alginate microspheres are prepared by Ionic gelation method using sodium alginate and calcium chloride. In Ionic gelation method, the mucilage of sodium alginate in water is initially prepared, the drug is dispersed in mucilage and the dispersion containing drug is pressed through a needle into calcium chloride solution. Sodium alginate mucilage containing the drug is pressed through a needle; it forms a succession of droplets which undergo a curing reaction in calcium chloride solution. During this curing reaction, sodium alginate is converted into calcium alginate, an insoluble polymer and forms spherical microspheres. As the mucilage is passed through a needle to produce spherical shaped microspheres. This method is also known as an Orifice method (9).

MATERIALS and METHODS

Piroxicam is obtained as a gift sample from Dr. Reddy's Lab, Hyderabad. All other chemicals used,such as, Sodium alginate and Calcium chloride were of analytical grade.

Preparation of Microspheres

Sodium alginate mucilage (2% w/v) was prepared. Twenty ml of the mucilage was taken in a small beaker. Piroxicam was added and dispersed with a spatula. The mucilage containing piroxicam was taken in a syringe fitted with a needle no: 22 and the mucilage is pressed slowly to get a succession of droplets. The droplets were collected into calcium chloride solution (10% w/v) taken in a beaker. The microspheres formed were allowed for curing reaction for about 20 minutes. The hardened microspheres formed were collected by decantation, washed with water and dried at 850c for 4 hours. The dried microspheres were collected and subjected to evaluation (10).

Evaluation of Microspheres

Estimation of drug content: A UV-Spectrophotometric method based at wavelength of 333 nm in 0.1 N HCl was used for the estimation of piroxicam.

Microspheres (50 mg) were weighed and taken into a 25 mL volumetric flask, 20 mL of methanol was added and mixed thoroughly while heating in a hot water bath to dissolve the drug contained in the microspheres. The solution was made up to the volume with methanol. The solution was then suitably diluted with 0.1 N HCl and the absorbance of resulting solution is measured at 333 nm (11).

Evaluation of size: The size of microspheres prepared was evaluated by Sieving method using standard sieves.

Drug release study: Piroxicam release from the microspheres prepared was studied in 900 mL of 0.1 N HCl using Electrolab 8 station dissolution rate test apparatus employing paddle stirrer. A temperature of 370c was maintained at each time microspheres, 100 mg of piroxicam microspheres was used in each test (12). Samples of 5 mL dissolution medium were withdrawn at different time intervals and assayed for piroxicam content at a wavelength of 333 nm. The dissolution medium withdrawn at each time was replaced with fresh quantity of dissolution medium (13).

RESULTS

Piroxicam release from microspheres prepared was studied in 0.1 N HCl. Piroxicam (Batch A) release from the microspheres prepared was slow and spread over a period of 22 hours, release follows first order kinetics, and it was diffusion controlled (Table 1).

Alginate microspheres (Batch B) prepared employing 90% sodium alginate and 10% piroxicam provided slow release of piroxicam over a period of 10 hrs.

DISCUSSION

Alginate microspheres containing piroxicam were prepared by Ionic gelation method. The prepared microspheres were found to be spherical, discrete and free-flowing. The microspheres prepared were evaluated for size, drug content and drug release characteristics. The microspheres prepared were passed through mesh no: 16 and retained on mesh no: 25. The average size of the microspheres prepared was found to 950 µ. The percent drug content was found to be 5.7 mg of piroxicam per 100 mg of microspheres (Table 2).

For the first batch, Batch A, analysis of release data as per Korse-Meyer Peppas equation indicated that the release was by Non-Fickian diffusion as the 'n' value was 0.65. These results are illustrated in Figure 1 and Table 3. Whereas for the second batch, Batch B, Piroxicam release from these microspheres was by Fickian diffusion mechanism. According to the second Fick's law, the basic equation of mass uptake by polymer film can be given by the Equation (Masaro, 1999).

$$\frac{Mt\ n}{M\alpha} = Kt^n$$

Where the exponent n is called the type of diffusion mechanism, and k is constant which depends on diffusion coefficient and thickness of film. Fickian diffusion (Case I) is often observed in polymer system when the temperature is well above the glass transition temperature of the polymer *(Tg)* (13).

Therefore, it is expected that the Mt /Mαis proportional to the square-root of time i.e. n= 0.5 .Other mechanisms has been established for diffusion phenomenon and categorized based on the exponent n, as follow

n > , Supercase II
n = 1, Case II
1 > n > 0.5, Anomalous
0.5 > n, pseudo-Fickian
n > , Supercase II

The Case II diffusion is the second most important mechanism of diffusion for the polymer. This is a process of moving boundaries and a linear sorption kinetics, which is opposed to Fickian. A sharp penetration front is observed by which it advances with a constant rate. An exponent between 1 and 0.5 signifies anomalous diffusion. Case II and Anomalous diffusion are usually observed for polymer whose glass transition temperature is higher than the experimental temperature. The main difference between these two diffusion modes concerns the solvent diffusion rate (13).

Furthermore as the figure illustrates, there seems to be a proportional linear increase of an amount released as the time span increases. The graph shows there is a more or less strong correlation of the data sets. However, at 60% for batch B and at 20% for batch A the dissolution pattern seems to waiver slightly. This might be attributable to power interruptions, clogging, instrument disruption

Table 1. Formula of alginate microspheres of piroxicam prepared by ionic gelation

Ingredients	Quantity
Sodium alginate (2% w/v)	2 gms
Calcium chloride (10% w/v)	10 gms
Piroxicam batch A	80 mg
Piroxicam batch B	40 mg

Table 2. Drug content data

Formulation	Drug content (mg/100 mg)
Batch A	5.696
Batch B	0.671

Table 3. Release parameters

Formulation	T50 (hr.)	T90 (hr.)	K0 (mg/hr.)	K1 (hr.^-1)	n' in Peppas
Microspheres (batch A)	5.4	18	4.48	0.128	0.65
Microspheres (batch B)	1.6	7	8.24	0.183	0.45

Table 4. Dissolution data of piroxicam microspheres

Time (hrs)	Piroxicam microspheres (batch A)	Piroxicam microspheres (batch A)
0	0	0
0.25	5.41	12.352
0.5	8.53	19.846
0.75	12.14	28.695
1	15.27	35.32
1.25	18.88	40.18
1.5	19.97	45.67
1.75	22.82	49.2
2	24.17	65.34
3	34.29	69.74
4	42.84	77.92
5	48.01	83.42
6	53.42	88.51
7	56.55	91.61
8	60.047	95.57
9	63.89	99.1
10	70.39	100
11	72.91	100
12	78.57	-
14	82.54	-
16	86.87	-
18	90.49	-
20	94.102	-
22	100	-
24	100	-

or etc. Mostly, nonetheless, the data sets are indicative of the release profile for the two data sets with respect to time.

For the release parameter study, UV spectroscopic method had to be used. There are different wavelengths that can be used to measure absorbance. Normally, the maximum absorbance wavelength has to be used because it is assumed to be unique to a compound. It is also the point where there is the highest absorbance of the ultraviolet rays. For piroxicam, the λmax used was 333nm. This is similar with a study done by M. A. Dhageet al, wherethe λmax for UV-Spectrophotometric determination of piroxicam was 334.35 nm (14).

According to these tabulated data (Table 4), the contents of batch A seems to have taken longer, by thirteen hours, to dissolve. On the contrary, for batch B, the contents, the 100 mg preparation, have been exhausted at the eleventh hour. At the 0.25 hour mark, the dissolved concentration of batch A was 5.41 mg, which is almost one third of the dissolved content in batch B at the same time mark. Beginning with the third hour mark, however, the percentage of dissolved batch A components seems to increase to 50% as compared to batch B components at the same point of time (15).

CONCLUSION

Alginate microspheres containing piroxicam could be prepared by Ionic gelation method employing an orifice. The microspheres were spherical, discrete and having a size of 950µ. Piroxicam release from the microspheres(batch A) was slow and spread over a period of 22 hrs, release by Non-Fickian diffusion mechanism. Hence, these alginate microspheres can be used for controlled drug delivery preferably for oral route as the size of the microspheres is large. In order to have an advanced and comprehensive picture of such dosage forms, it is recommended that further studies be performed vis-à-vis piroxicam. It would be a beneficial endeavor if one investigates further studies such as, in-vivo, clinical, Bioavailability and Bioequivalence studies.

CONFLICT of INTEREST

The authors would like to declare that they have no competing/ conflict of interest.

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