Int. J. Chem. Sci.: 10(2), 2012, 783-792
ISSN 0972-768X
www.sadgurupublications.com

PREPARATION AND EVALUATION OF MUCOADHESIVE MICROCAPSULES EMPLOYING OLIBANUM RESIN FOR CONTROLLED RELEASE OF DICLOFENAC

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ABSTRACT

The objective of the study is to evaluate olibanum resin, a natural resin polymer as coat for mucoadhesive microcapsules and to design mucoadhesive microcapsules of diclofenac for controlled release. Olibanum resin coated microcapsules of diclofenac were prepared by an emulsification-solvent evaporation method employing different proportions of core and coat and the microcapsules were evaluated for size, drug content and microencapsulation efficiency, wall thickness, surface characters by SEM, drug release kinetics and mechanism and mucoadhesiveness. The olibanum resin coated microcapsules prepared were found to be discrete, spherical, free flowing and multinucleate monolithic type. Drug content was uniform (c.v.≤1.3 %) in each batch of microcapsules and the microencapsulation efficiency was in the range 99.3% - 102%. Diclofenac release from the olibanum resin coated microcapsules was slow and spread over a period of 24 h and depended on core: coat ratio, wall thickness and size of the microcapsules. Drug release from these microcapsules was majorly by non- Fickian diffusion in the case of larger microcapsules and by Fickian diffusion in the case of smaller microcapsules. Good linear relationships were observed between wall thickness of the microcapsules and release rate (K_0) and (K_1) values. In the *in vitro* wash-off test olibanum resin coated microcapsules exhibited good mucoadhesive property. Olibanum resin was found to be a new and an efficient microencapsulating agent for mucoadhesive microcapsules and the olibanum resin coated microcapsules were found suitable for oral controlled release of diclofenac over 24 h.

Key words: Olibanum Resin, Diclofenac, Mucoadhesive Microcapsules, Controlled Release.

INTRODUCTION

Controlled release drug delivery systems are aimed at controlling the rate of drug delivery, sustaining the duration of therapeutic activity and/or targeting the delivery of the

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drug to tissue. Drug release from these systems should be at a desired rate predictable and reproducible. Among various oral controlled release systems, microencapsulation and microcapsules are widely accepted[1,2]. A new novel promising technology for obtaining controlled release and enhancing the bioavailability is a combination of mucoadhesion principle and microencapsulation to result in mucoadhesive microcapsules. Mucoadhesive microcapsules consist of either entirely of a mucoadhesive polymer or having an outer coating enclosing the drug particles. They facilitate an intimate and prolonged contact with the absorption surface to provide controlled release and enhanced bioavailability of the contained drug over longer period of time to prolong its therapeutic action. The polymer used in mucoadhesive microcapsules plays a vital role in either controlling the drug delivery or enhancing bioavailability of the contained drug. Though a wide range of polymers are reported for preparing mucoadhesive microcapsules, there is a continued need to develop new, safe and effective polymers for mucoadhesive microcapsules. The objective of the present study is to evaluate olibanum resin as coat for mucoadhesive microcapsules for controlled release.

Olibanum is a gum resin obtained from *Boswellia serrata*, Roxburgh and other species of Boswellia. Olibanum consists[3] of chiefly of an acid resin (50 - 60%), gum (30 - 36%) and volatile oil (3 - 8%). The resin consists[4] mainly a resin acid (boswellic acid) and a resene (olibanoresene) in equal proportions. Ether soluble resin extracted from olibanum exhibited excellent release retarding and rate controlling properties in matrix tablets and microcapsules for controlled release[5-7]. Preliminary studies indicated that the resin also has good mucoadhesive property. In the present work, diclofenac was microencapsulated by olibanum resin and the microcapsules were evaluated for mucoadhesiveness and controlled release of diclofenac. Diclofenac has a short biological half-life of 1 – 2 h and is required to be administered repeatedly 3 or 4 times a day. It causes gastric disturbances such as nausea, ulceration with bleeding, vomiting, abdominal pain and constipation if present in large concentration in the gastrointestinal tract. Hence controlled release or sustained release formulations are needed for diclofenac to prolong its duration of action, reduce frequency of administration with better patient compliance and to reduce unwanted gastric disturbances. A few sustained release products of diclofenac are available commercially.

EXPERIMENTAL

Materials and methods

Materials

Diclofenac was a gift sample from M/s. Micro Labs Ltd., Pondicherry. Olibanum

gum was procured from M/s Grijan Co-operative Corporation Ltd., Govt. of Andhra Pradesh, Visakhapatnam. Sodium carboxy methyl cellulose (high viscosity grade 1500-3000 cps of a 1% w/v solution at 25°C) (Loba-chemie) and chloroform AR (Merck) were procured from commercial sources. All other materials used were of pharmacopoeial grade.

Methods

Preparation of olibanum resin

Olibanum resin used as coat material was extracted from olibanum gum in the laboratory as follows: Powdered olibanum (10 g) was extracted repeatedly with 4 × 50 mL quantities of solvent ether. The ether extracts were collected in a porcelain dish and concentrated to dryness at 40°C. The dried mass obtained was powdered and passed through mesh No. 120.

Preparation of microcapsules

An emulsification-solvent evaporation method was tried to prepare olibanum resin coated microcapsules. Olibanum resin (2 g) was dissolved in chloroform (100 mL) to form a homogeneous solution. Core material i.e. medicament (diclofenac) (0.8 g) was added to 10 mL polymer (resin) solution which provides 0.2 g of polymer and mixed thoroughly. The resulting mixture was then added in a thin stream to 200 mL of an aqueous mucilage of sodium CMC (0.5% w/v) contained in a 450 mL beaker while stirring at 1000 rpm to emulsify the added dispersion as fine droplets. A Remi medium duty stirrer with speed meter (Model RQT 124) was used for stirring. The solvent, chloroform was then removed by continuous stirring at room temperature (28°C) for 3 h to produce spherical microcapsules. The microcapsules were collected by vacuum filtration and washed repeatedly with water. The product was then air dried to obtain discrete microcapsules. Different proportions of core: coat viz. 19:1 (MC1), 9:1 (MC2), 8:2 (MC3) and 7:3 (MC4) were used to prepare microcapsules with varying coat thickness.

Estimation of diclofenac

A UV-spectrophotometric method based on the measurement of absorbance at 276 nm in phosphate buffer of pH 7.4 was used to estimate the diclofenac content of the microcapsules. The method was validated for linearity, accuracy and precision. The method obeyed Beer's law in the concentration range of 0-10 µg/mL. When a standard drug solution was assayed repeatedly (n = 6) low RSD (< 0.5%) values ensured reproducibility of the method. No interference from the excipients was observed.

Characterization of microcapsules

Size analysis

For size distribution analysis, different sizes in a batch were separated by sieving using a range of standard sieves. The amounts retained on different sieves were weighed.

Microencapsulation efficiency

Microencapsulation efficiency was calculated using the equation,

$$\text{Microencapsulation Efficiency} = \frac{\text{Estimated percent drug content in microcapsules}}{\text{Theoretical percent drug content in microcapsules}} \times 100$$

Scanning electron microscopy

The microcapsules prepared were observed under a scanning electron microscope (Jeol JXA 8100 LTD, Tokyo, Japan). The samples were fixed on a brass stub using double sided sticking tape and then gold coated in vacuum by a sputter coater. The pictures were the taken at an excitation voltage of 15 KV.

Wall thickness

Assuming the microcapsules to be uniform and spherical, wall thickness of the microcapsules in the present study was determined by the method described by Luu et al.[8] using the equation:

$$h = \frac{\bar{r}\,(1 - p)d_1}{3[pd_2 + (1 - p)\,d_1)}$$

where 'h' is the wall thickness, '$\bar{r}$' is the arithmetic mean radius of the microcapsule, 'd_1' is the density of the core material, 'd_2' is the density of the coat material and 'p' is the proportion of the medicament in the microcapsule. Densities were determined using petroleum ether as displacement fluid for drug and water as displacement fluid for resin at room temperature. Mean radius of the microcapsules was determined by sieving.

Drug release study

Drug release from the olibanum resin coated microcapsules of size 20/35 and 35/50 was studied using an eight station dissolution rate test apparatus (Model Disso-2000, M/s LABINDIA) with a paddle stirrer at 50 rpm and 37 ± 0.5 °C. Phosphate buffer of pH 7.4

(900 mL) was used as dissolution fluid. A sample of microcapsules equivalent to 100 mg of drug was used in each test. Samples of dissolution fluid (5 mL) were withdrawn through a filter (0.45 μm) at different time intervals over a period of 24 h and were assayed for diclofenac content at 276 nm. The sample (5 mL) taken at each sampling time was replaced with fresh dissolution medium (5 mL). The drug release experiments were conducted in triplicate.

Evaluation of mucoadhesiveness

The mucoadhesive property of the olibanum resin coated microcapsules was evaluated by an *in vitro* adhesion testing method known as wash-off method[9]. The mucoadhesiveness of the microcapsules prepared was compared to that of non-bioadhesive material, ethylene vinyl acetate (EVA) microcapsules. Pieces of goat intestinal mucosa (2×2 cm) were mounted onto glass slides (3×1 inch) with cyanoacrylate glue. Two glass slides were connected with a suitable support. About 50 microcapsules were spread on to each wet rinsed tissue specimen and immediately thereafter the support was hung on to the arm of a USP tablet disintegration test machine. By operating the disintegration test machine the tissue specimen was given a slow regular up and down movement in 900 mL test fluid at 37ºC taken in a 1 L vessel of the disintegration test machine. At the end of 30 min, 1 h and later at hourly intervals up to 8 h, the machine was stopped and the number of microcapsules still adhering on to the tissue was counted. The test was performed in 0.1 N HCl and in phosphate buffer of pH 6.2.

RESULTS AND DISCUSSION

Olibanum resin coated microcapsules of diclofenac could be prepared by the emulsification-solvent evaporation method developed. The olibanum resin microcapsules prepared were found to be discrete, spherical, free flowing with an angle of repose in the range 15° - 20°. The nature of the method of preparation indicated that the microcapsules were of multinucleate monolithic type. SEM (Fig. 1) indicated that the microcapsules were spherical with smooth surface and completely covered with polymer (resin) coat. Size analysis showed that a large proportion of microcapsules in a batch were in the size range of -20 +35 (670 μm) and -35 +50 (398 μm). The emulsification-solvent evaporation method used to prepare the mucoadhesive microcapsules employing olibanum resin is reproducible with regard to size and size distribution of the microcapsules. Drug content was uniform (C.V.≤ 1.3%) in each batch of microcapsules. The microencapsulation efficiency was in the range 99.3% - 102%. Microcapsules prepared with various ratios of core: coat were found to have different wall thickness with both the drugs. Smaller microcapsules have thinner walls in each case. (Table 1)

Table 1: **Drug content, microencapsulation efficiency, wall thickness and release characteristics of olibanum resin coated microcapsules of diclofenac**

Micro-capsules (core: coat ratio)	Drug Content (%)	Micro-encapsulation Efficiency (%)	Wall Thicknes (µm)	T_{50} (h)	T_{90} (h)	K_0 (mg/ h)	K_1 (h^{-1})	'n' in Peppas Equat on
Size 20/35								
MC1(19:1)	94.4(0.39)*	99.3	9.5	3.8	7.1	11.1882	0.3431	0.7158
MC2(9:1)	90.(0.51)	100	17	3.9	10.8	6.0578	0.2261	0.5833
MC3 (8:2)	80.4(0.67)	100.5	20.1	5.6	17.2	4.0464	0.1833	0.5783
MC4 (7:3)	71.4(0.84)	102	21.5	8.2	>24	3.4826	0.0783	0.6195
Size 35/50								
MC1(19:1)	94.9(0.74)	99.9	7.2	1.1	4.1	14.2496	0.5762	0.4032
MC2(9:1)	90.4(0.71)	100.4	11	1.4	7.0	8.4605	0.3636	0.3839
MC3 (8:2)	81(0.52)	101.3	13	2.4	10.5	5.4151	0.2282	0.4184
MC4 (7:3)	70.7(1.3)	101	14.5	2.9	16	3.6551	0.1724	0.4631

Figures in parentheses are coefficient of variation values (C. V. %)

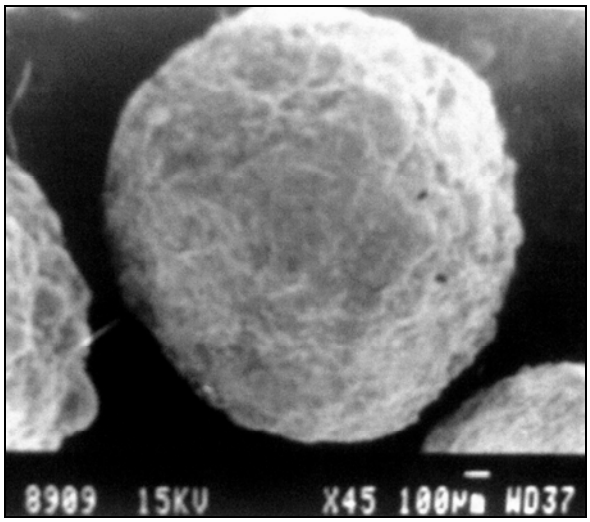

Fig. 1: SEM of Olibanum resin coated microcapsule (size 20/35) of diclofenac

Diclofenac release from the olibanum resin coated microcapsules was slow and spread over a period of 24 h and depended on core:coat ratio, wall thickness and size of the microcapsules. The release data were analyzed as per zero order, first order, Higuchi[10] and Peppas[11] equation models. Drug release from the olibanum resin coated microcapsules was diffusion controlled and followed first order kinetics indicated by the higher correlation coefficient values obtained (Table 2). When the release data were analyzed as per Peppas equation, the release exponent 'n' was in the range 0.5783 - 0.7158 in the case of microcapsules of size 20/35 indicating that the release mechanism from these microcapsules was by non - Fickian diffusion. Whereas in the case of smaller microcapsules (size 35/50) the release exponent 'n' was in the range 0.3839 – 0.4631 indicating Fickian diffusion as the release mechanism. When the release was relatively rapid as in the case of size 35/50, it followed Fickian diffusion. Good linear relationships were observed between wall thickness of the microcapsules and release rate (K_0) and (K_1) values (Figs. 2-3).

Table 2: Correlation coefficient (R^2) values in the analysis of release data of olibanum resin coated microcapsules of diclofenac as per various kinetic models

Microcapsules (core : coat ratio)	Correlation Coefficient 'R^2' values			
	Zero Order	First order	Higuchi	Peppas
Size 20/35				
MC1 (19:1)	0.9778	0.8195	0.945	0.9794
MC2 (9:1)	0.8995	0.9591	0.9883	0.9839
MC3 (8:2)	0.9105	0.9136	0.9929	0.9896
MC4 (7:3)	0.9062	0.9919	0.9881	0.9835
Size 35/50				
MC1 (19:1)	0.8318	0.9959	0.978	0.9694
MC2 (9:1)	0.7664	0.9784	0.9497	0.9275
MC3 (8:2)	0.7906	0.986	0.9636	0.9678
MC4 (7:3)	0.7768	0.9587	0.9499	0.9174

Olibanum resin coated microcapsules exhibited good mucoadhesive property when compared to non-mucoadhesive EVA microcapsules. The wash-off was slow in the case of olibanum resin coated microcapsules when compared to EVA microcapsules. In the case of olibanum resin coated microcapsules the wash-off was faster at intestinal pH (6.2) than at

gastric pH (1.2). The rapid wash-off observed at intestinal pH (6.2) is due to ionization of carboxyl group in the acid resin of olibanum at this pH, which increases its solubility and reduces adhesive strength. Thus, the results indicated that olibanum resin coated microcapsules have good mucoadhesive property (Table 3).

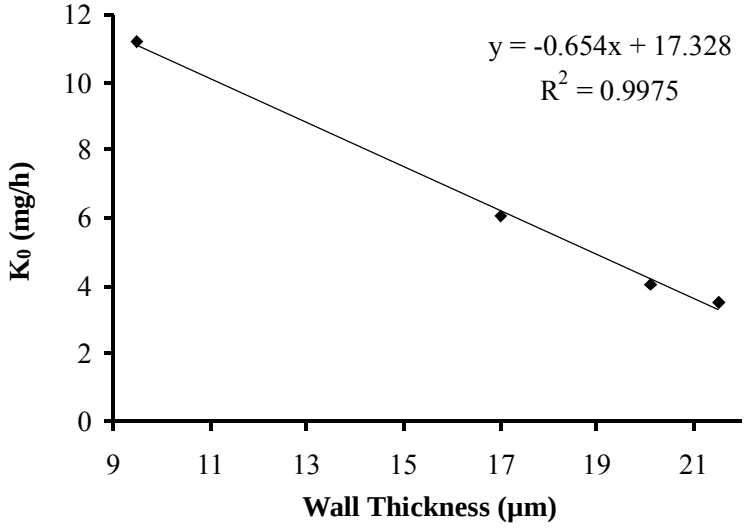

Fig. 2: Relationship between wall thickness and release rate (K_0) of olibanum resin coated microcapsules of diclofenac (Size 20/35)

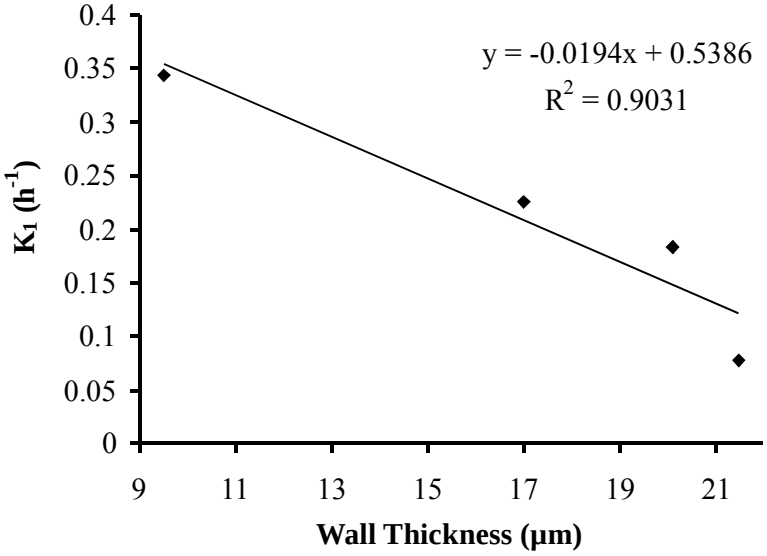

Fig. 3: Relationship between wall thickness and release rate (K_1) of olibanum resin coated microcapsules of diclofenac (Size 20/35)

Table 3: Results of *in vitro* wash-off test to assess mucoadhesive property of olibanum resin coated microcapsules

Microcapsules (Size 20/35)	Percent Microcapsules Adhering to Tissue After Time (h)									
	In 0.1 N HCl, pH 1.2					In Phosphate Buffer, pH 6.2				
	1	2	4	6	8	1	2	4	6	8
MC3	83 (2.1)*	67 (2.8)	52 (3.0)	40 (1.4)	28 (1.8)	64 (2.2)	42 (2.4)	28 (1.8)	7 (2.2)	6 (1.8)
EVA	56 (2.8)	40 (3.1)	10 (2.6)	--	--	51 (2.4)	39 (3.2)	9 (1.8)	--	--

*****Figures in parentheses are coefficient of variation (C.V.%) values

CONCLUSION

(i) Spherical olibanum resin coated microcapsules of diclofenac could be prepared by the emulsification-solvent evaporation method developed. The method is industrially feasible as it involves emulsification and removal of solvent, which can be controlled precisely. Microencapsulation efficiency was in the range 99.3% - 102 %.

(ii) Diclofenac release from the olibanum resin coated microcapsules was slow and spread over a period of 24 h and depended on core : coat ratio, wall thickness and size of the microcapsules. Drug release from these microcapsules was majorly by non-Fickian diffusion in the case of larger microcapsules and by Fickian diffusion in the case of smaller microcapsules.

(iii) Good linear relationships were observed between wall thickness of the microcapsules and release rate (K_0) and (K_1) values.

(iv) In the *in vitro* wash-off test olibanum resin coated microcapsules exhibited good mucoadhesive property.

(v) Olibanum resin was found to be a new and an efficient microencapsulating agent for mucoadhesive microcapsules for oral controlled release. Controlled release mucoadhesive microcapsules of diclofenac could be designed employing olibanum resin. Olibanum resin coated microcapsules of diclofenac exhibited good mucoadhesion and controlled release characteristics and were found suitable for oral controlled release for 24 h. Olibanum is reported as non-toxic[12] and since it is of natural origin it is biocompatible and cheaper.

REFERENCES

1. A. Kondo, In, "Microcapsule Processing and Technology", Marcel Dekker, Inc., New York (1979) p. 18.
2. M. H. Gutcho, In, "Microcapsules and Microencapsulation Techniques", Noyes Data Corporation, New Jersy (1976) p. 236.
3. S. K. Nigam and C. R. Mitra, Indian Drugs, **16**, 80 (1979).
4. R. S. Srinivas and B. Madhu, Indian J. Chem., **176**, 168, (1976).
5. K. P. R. Chowdary, P. Mohapatra, and M. N. Murali Krishna, Indian J. Pharm. Sci., **68**, 497 (2006).
6. K. P. R. Chowdary and P. Srinivas, Asian J. Chem.**, 20**, 5391 (2008).
7. K. P. R. Chowdary, P. Mohapatra, and M. N. Murali Krishna, Indian J. Pharm. Sci., **68,** 461(2006).
8. S. N. Luu, P. F. Carlies, P. Delort, K. Gazzola and D. Lanfont, J. Pharm. Sci., **62**, 452 (1973).
9. C. M. Lehr, J. A. Bowstra, J. J. Tukker and H. E. Junginu, J. Cont. Rel., **13,** 51 (1990).
10. T. Higuchi, J. Pharm. Sci.*,* **52**, 1145 (1963).
11. P. L. Ritger and N. A. Peppas, J. Cont. Rel., **5**, 37 (1987).
12. G. Joerg, B. Thomas and J. Christof, In, "Physicians Desk Reference for Herbal Medicines", Medical Economics Company, Montvale, New Jersey, Edn., **2**, (2000) p. 319.

Accepted : 03.10.2009