Research Article

FORMULATION AND EVALUATION OF MUCOADHESIVE MICROCAPSULES OF METFORMIN HCL WITH GUM KARAYA

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ABSTRACT

The objective of this work was to develop mucoadhesive microcapsules of Metformin Hcl for controlled release. Metformin Hcl microcapsules were prepared with a coat consisting of alginate and Gum Karaya by employing Ionotropic Gelation process and Emulsification Ionotropic Gelation process. The microcapsules were evaluated for flow properties, Carr's index, hausner ratio, micro-encapsulation efficiency, drug release characteristics, surface characteristics; compatibility studies and mucoadhesive properties. As hausner ratio was less than 1.25 and Carr's index values were less than 25 from both the methods, hence they were found to be free flowing. Sharp endothermic peaks were noticed from the microcapsules formulated with two different techniques at 226ºC indicating the compatibility between the drug and the polymer Gum Karaya. Metformin Hcl release from the microcapsules was slow and followed zero order kinetics (r > 0.98) and followed non–fickian (n value 0.5 to 1) release and depended on the coat: core ratio and the method employed in the preparation of microcapsules. Among the two methods Emulsification Ionotropic Gelation method was found to be more suitable for Controlled release of Metformin Hcl over a long period of time. These microcapsules were subjected to in-vitro wash-off test and exhibited good mucoadhesive property.

Keywords: Ionotropic Gelation, Emulsification Gelation Technique, Metformin Hcl, Gum Karaya,

INTRODUCTION

Micro encapsulation has been accepted as a process to achieve controlled release and drug targeting. Micro encapsulation by various polymers and its applications are described in standard textbooks [1, 2]. Mucoadhesion has been a topic of interest in the design of drug delivery systems to prolong the residence time of the dosage form at the site of application or absorption and to facilitate intimate contact of the dosage form with the underlying absorption surface to improve the bioavailability of drugs[3]. Several studies reported drug delivery systems in the form of tablets, films, patches, and gels for oral, buccal, nasal, ocular, and topical routes. Amongst the polymers used for micro encapsulation alginate has gained much attention since it is nontoxic, biodegradable and can be prepared by a safe technique avoiding organic solvents [4].

Ionotropic-Gelation method is not practical because of the blockage of the spraying nozzle and the low yield of the product. Hence Ionotropic-Gelation of sodium alginate by the emulsification technique was developed as an alternative approach [5]. Gum Karaya is natural gum exudates obtained from stems and branches of "Sterculia Uren's" and family belongs to Sterculacia [6-7]. Gum Karaya is a negative charged colloid and a high-molecular weight complex acidic polysaccharide. The general utility of Gum Karaya is based on its viscosity. It was successfully evaluated for its suitability in the preparation of hydrophilic matrices, mini-matrices, and microcapsules of Metformin Hcl, an effective antidiabetic that requires controlled release owing to its short biological half-life of 3.4 ± 0.7 hours, was used as the core in micro-encapsulation [8-9]. The purpose of this research was to formulate and in-vitro evaluation of mucoadhesive microcapsules of Metformin Hcl with natural polymer Gum Karaya to achieve controlled release.

MATERIALS AND METHODS

Materials

Metformin Hcl U.S.P was kindly gifted by M/s Aurobindo Pharmaceuticals, Hyderabad. Girijan Co-operative Corporation Ltd (Visakhapatnam, India) supplied Gum Karaya (Grade1). Sodium alginate (having a viscosity of 5.5 cps in a 1% w/v aqueous solution at 25 ºC), calcium chloride and heavy liquid paraffin were procured from s. d. Fine Chemicals Pvt. Ltd., Mumbai, India. All chemicals used were of analytical grade.

Preparations of microcapsules [10, 11]

In Ionotropic Gelation technique polymer Gum Karaya (1 g) and sodium alginate (1 g) were dissolved in 40 ml of water to form a homogeneous polymer solution. The active substance, Metformin Hcl (2.0 g), was added to the polymer solution and mixed thoroughly with a stirrer to form a viscous dispersion. The resulting dispersion was then added manually drop wise into calcium chloride (10% w/v) solution (40 mL) through a syringe with a needle of size no 18. The added droplets were retained in the calcium chloride solution for 20 minutes to complete the curing reaction and to produce spherical rigid microcapsules having core: coat ratio 1:1 (MC1). Similarly microcapsules with core:coat ratio 1:1.5(MC2) and 1:2 (MC 3) were also prepared. The microcapsules were collected and dried over night at room temperature.

In Emulsion Gelation technique polymer Gum Karaya (1 g) and sodium alginate (1 g) were dissolved in 40 ml of water. The drug (2 g) was added to the polymer solution and mixed thoroughly. The polymer dispersion was then added in a thin string to 50 ml of heavy liquid paraffin contained in a 250 ml beaker, which is kept under a Remi make medium duty stirrer with speedometer (RQ 121/D) at 500 rpm to emulsify the added dispersion as fine droplets. Then to this emulsion 40 ml of calcium chloride solution (15% w/v) was transferred while stirring at 500 rpm for 15 min to produce spherical microcapsules. The so formed microcapsules were collected and washed with petroleum ether repeatedly and finally the microcapsules were dried under room temperature. Different proportions of core: coat microcapsules viz., 1:1 (MC4), 1:1.5 (MC5) and 1:2 (MC6) were prepared.

EVALUATION OF MICROCAPSULES [12-14]

Size distribution and size analysis

For size distribution analysis, 250 mg of the microcapsules of different sizes in a batch were separated by sieving, using a range of standard sieves. The amounts retained on different sieves were weighed. The mean particle size of the microcapsules was calculated by the formula.

$$\text{Mean Particle Size} = \frac{\sum \left(\text{Mean Particle Size of the Fraction X Weight Fraction}\right)}{\sum \left(\text{Weight Fraction}\right)}$$

Flowability of microcapsules

The static angle of repose was measured according to the fixed funnel and free standing cone method. The bulk density of the mixed microcapsules, the Hausner's ratio and Carr's index, were calculated from the poured and tapped bulk densities of a know weight of sample using a measuring cylinder. The following formulas were used for calculating

$$\text{Hausner ratio} = \frac{D_p}{D_t}$$

$$\text{Carr's index} = \frac{\left(D_p - D_t\right)}{D_p} \times 100$$

Where D_p (Poured density) = Weight of the microcapsules ÷ V_p(Poured Volume), D_t (tapped density) =Weight of the microcapsules ÷ V_t (tapped Volume).

Drug content and entrapment efficiency [14-15]

Metformin Hcl content in the microcapsules was estimated by a UV spectrophotometric (UV-1700, Shimadzu, Japan) Actual drug content and encapsulation efficiency of the microcapsules was determined by following method 100 mg of microcapsules were crushed using mortar and pestle. The crushed microcapsules were placed in 100 ml of 0.1 N HCl (pH 1.2) and shaken for 1 h at 37±0.50 C in mechanical shaker. The samples were then filtered to obtained clear solution and analyzed for the drug content spectrophotometrically at 233 nm, it gives drug content for 100 mg of microcapsules from that calculate drug content for total quantity of micro capsules , from actual drug content, the value of encapsulation efficiency was determined using the formula given below. The method was validated for linearity, accuracy, and precision. The method obeyed Beer's law in the concentration range 1 to 10µg/ml. When a standard drug solution was assayed repeatedly (n = 6), the mean error (accuracy) and relative standard deviation (precision) were found to be 0.6% and 0.8%, respectively.

Microencapsulation efficiency

Microencapsulation efficiency was calculated using the following formula.

$$\text{Microencapsulation Efficiency} = \frac{\text{Estimated Percentage Drug Content}}{\text{Theoretical Percentage Drug Content}} \times 100$$

Surface accumulation study[16]

This study was conducted to estimate the amount of drug present on the surface of the formulations which may shows immediate release in the dissolution media.50mg of formulation were suspended in 50ml of 0.1 Hcl (pH1.2). The samples were shaken vigorously for 30 min by hand shaking. The amount of drug leached out from the surface was analyzed spectrophotometrically at 233 nm. Percentage of drug released with respect to entrapped drug in the sample recorded.

Scanning electron microscopy (SEM)

The samples for the SEM analysis were prepared by sprinkling the microcapsules on one side of the double adhesive stub. The stub was then coated with fine gold dust. The microcapsules were then observed with the scanning electron microscope (Leica Electron Optics, Cambridge, USA) at15kv.

Differential scanning calorimetry

Differential scanning calorimetry (DSC) thermograms were obtained by a differential scanning calorimeter (DSC 220C, Seiko, Tokyo, Japan) at a heating rate of 10ºC/min from 30 to 300ºC in a nitrogen atmosphere 20ml/min with a sample weight of 3mg.

Infrared spectroscopic studies

Fourier–transformed infrared (FT–IR) spectra were obtained on a Perkin Elmer 2000 FT–IR system (Perkin Elmer, Norwalk, CT) using the KBr disk method (2 mg sample in200 mg KBr). The scanning range was 400 to 4000 cm-1 and the resolution was 1 cm-1.

In vitro release studies [17-19]

Microcapsules containing equivalent to 100mg of Metformin Hcl were packed in hard gelatin capsule and subjected to in-vitro drug release studies. Release of Metformin Hcl from the capsule was studied in pH 1.2 Hcl (900 mL) using XXIV 8-station dissolution rate test apparatus (Model TDT - 08L, M/s Electro lab, Mumbai, India) with a rotating paddle stirrer at 50 rpm and 37ºC ± 1ºC. A sample of microcapsules equivalent to 100 mg of Metformin Hcl was used in each test. Samples of dissolution fluid were withdrawn through a filter (0.45 µm) at different time intervals and were assayed at 233 nm for Metformin Hcl content using a Shimadzu UV-1700 double beam spectrophotometer (Shimadzu Corporation, Japan). The drug release experiments were conducted in triplicate (n = 3). The release data obtained were fitted to zero order, first order, Higuchi and Korsmeyer peppas equations to determine the corresponding release rate and mechanism of drug release from the mucoadhesive microcapsules.

In vitro mucoadhesion study [20-22]

The sheep stomach mucosa was used for in vitro mucoadhesion evaluation. The mucosa was removed and cut into pieces 2 cm long and 2 cm wide and were rinsed with 2 ml of 0.1 N HCl (pH 1.2). Fifty microcapsules of each were scattered uniformly on the surface of the stomach mucosa. After 20 minutes, the tissue were taken out and fixed on a polyethylene support at an angle 45⁰. About 50 microcapsules were spread onto each wet rinsed tissue specimen, and immediately thereafter the support was hung onto the arm of a USP tablet disintegrating test machine. When the disintegrating test machine was operated, the tissue specimen was given a slow, regular up-and-down movement in the test fluid at 37ºC contained in a 1 L vessel of the machine. At the end of 30 minutes, at the end of 1 hour, and at hourly intervals up to 12 hours, the machine was stopped and the number of microcapsules still adhering to the tissue was counted.

RESULTS AND DISCUSSION

Microcapsules of Metformin Hcl could be prepared by ionotropic gelation process and emulsification gelation process employing Gum Karaya as the polymer. The microcapsules were found to be discrete spherical and free flowing. The size analysis of different batches of microcapsules showed that about 80% of the prepared microcapsules were in the size range of 939-1204 µm (16/20 sieve). The size distribution of microcapsule was found to be normal in all the batches. The microcapsules imparted good flow ability as indicated by angle of repose (13.44 - 24.160), the Carr's index (10.55 g cm-3 – 23.64 g cm-3) and the Hausner Ratio (0.93 – 1.29). The Microencapsulation efficiency was in the range of 73% to 86%, with various formulations. The surface morphology were also studied and the results were tabulated in table 1. The SEM photographs indicated that the microcapsules were spherical and completely covered with the coat polymer (Fig 1 and 2).

Selected DSC thermogram of the drug and microcapsule were shown in Fig 3 respectively. The DSC analysis (Fig.3) of pure metformin HCl showed a characteristic, sharp endotherm peak at 226°C corresponding to its melting point and indicates the crystalline nature of the drug. The DSC analysis of drug and polymer revealed negligible change in the melting point of metformin HCl indicating no modification or interaction between the drug and polymer.

Table 1: Characterization of Metformin Hcl microcapsules formulated with gum karaya by different techniques

| Method	Core: coat ratio	Angle of repose	Carr's index	Hausner's ratio	Surface accumulation study (%)	Drug content	Entrapment efficiency (%)	Average particle size (μ)
MC1	1:1	24.16	23.64	1.29	3.68	43.24	86.48	1073.1
MC2	1:1.5	19.74	19.30	1.20	2.66	41.23	82.47	1127.8
MC3	1:2	15.13	14.62	1.09	1.55	38.2	76.40	1204.4
MC4	1:1	22.49	19.04	1.23	2.18	41.66	83.32	939.75
MC5	1:1.5	17.78	16.55	1.17	1.88	39.56	79.12	1129.6
MC6	1:2	13.44	10.55	0.93	1.01	36.56	73.12	1141.5
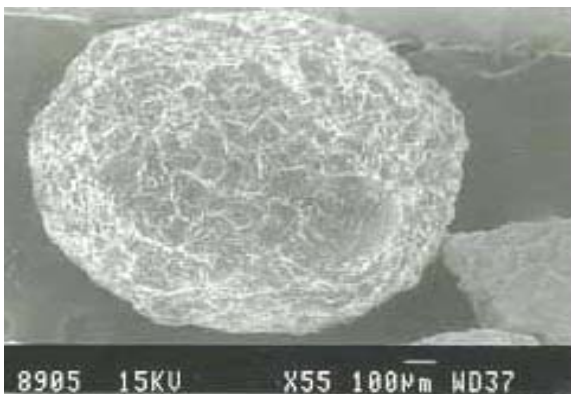
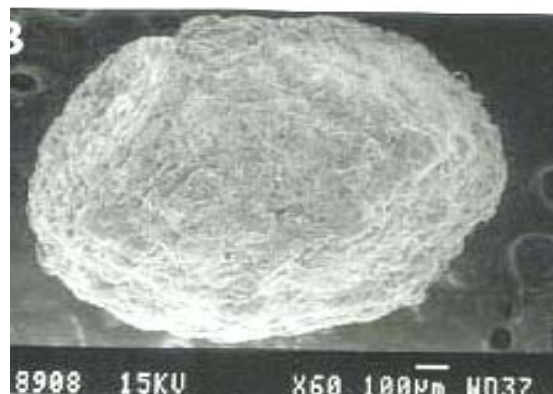**Fig. 1: Microcapsule of Metformin Hcl formulated by ionotropic gelation**

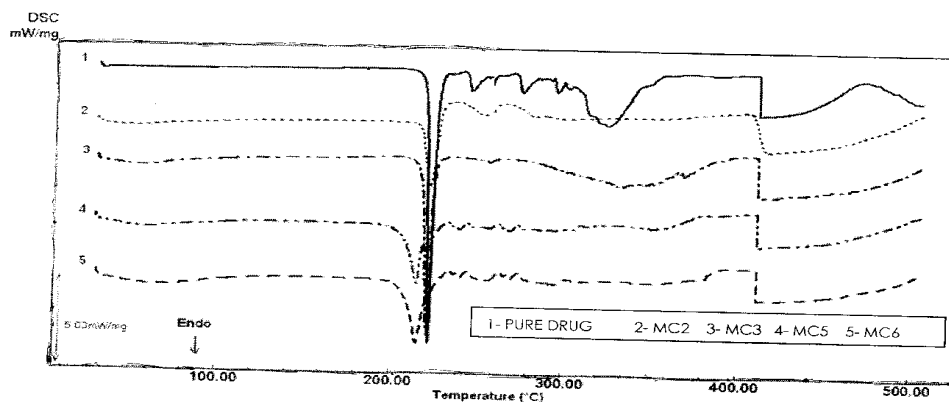**Fig. 2: Microcapsule of Metformin Hcl formulated by emulsification gelation**

Fig. 3: DSC thermo gram of pure drug and polymer with different techniques

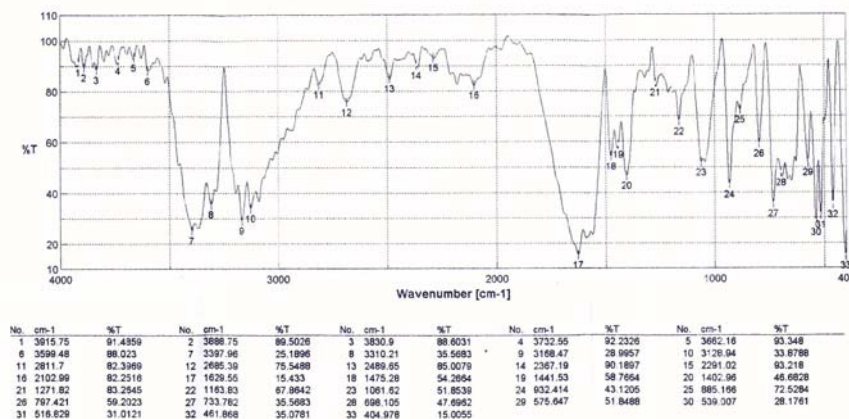| No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T
1	3915.75	91.4959	2	3888.75	89.5026	3	3830.9	88.6031	4	3732.55	92.2326	5	3662.16	93.348
6	3599.48	86.023	7	3397.96	25.1896	8	3310.21	35.5683	9	3168.47	90.1897	10	3126.94	33.8788
11	2811.7	82.3969	12	2685.39	75.5488	13	2489.65	85.0079	14	2367.19	90.1897	15	2291.02	93.218
16	2102.99	82.2516	17	1629.55	15.433	18	1475.28	54.2864	19	1441.53	58.7664	20	1402.96	46.6828
21	1271.82	80.2545	22	1163.83	67.8642	23	1061.62	51.8539	24	932.414	43.1205	25	885.166	72.5284
26	797.421	59.2023	27	733.782	35.5683	28	698.105	47.6962	29	575.647	51.8488	30	539.007	28.1761
31	516.829	31.0121	32	461.868	35.0781	33	404.978	15.0055						

Fig. 4: IR Spectra of Metformin Hcl

Compatibility study of drug and polymer were conducted by employing I.R. Spectral studies. The IR spectrum of Metformin Hydrochloride, Gum Karaya & its formulation is shown in figure: 4-6. The following characteristic peaks were observed with Metformin Hydrochloride as well as the formulations containing Metformin

Hydrochloride. C=N - (stretching) 1629.55 ,1655.59, 1669 cm^{-1}, C-N - (stretching) 1061.62, 1029.48, 1030.77 cm^{-1}, N-H - (stretching) 3397.96, 3378.67, 3394.1 cm^{-1} . As the identical principle peaks were observed in all the cases, hence it shall be confirmed that interactions do not exist between the drug and polymer.
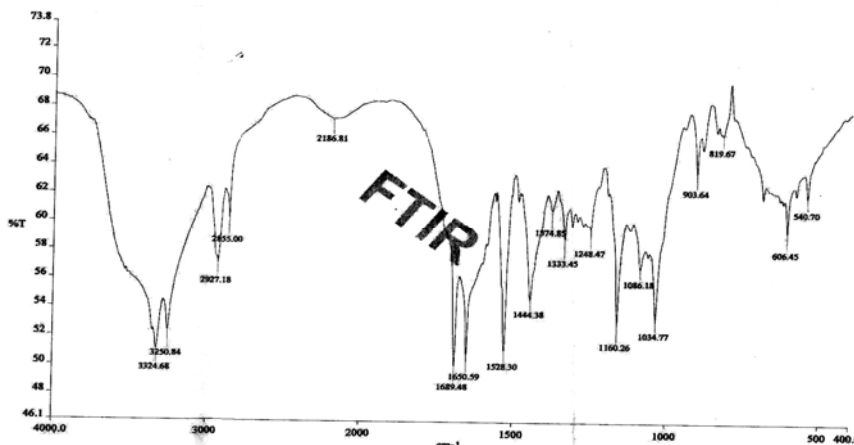

Fig.5: IR Spectra of Gum Karaya

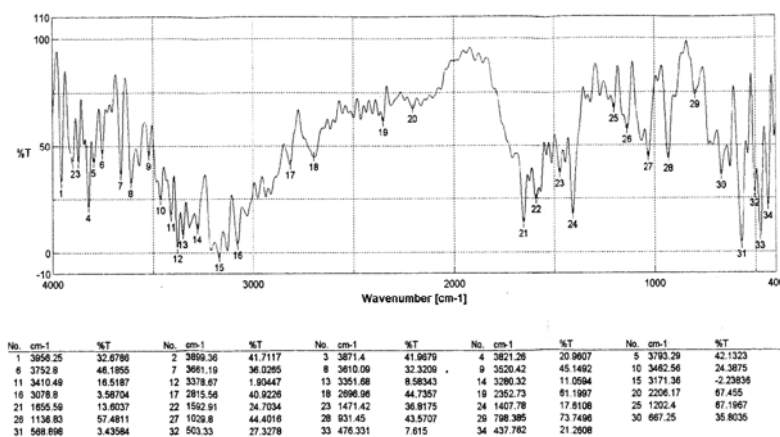| No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T	No.	cm-1	%T
1	3958.25	32.6786	2	3899.36	41.7117	3	3871.4	41.9679	4	3821.26	20.9607	5	3793.29	42.1323
6	3752.8	46.1855	7	3661.19	36.0265	8	3610.09	32.3209	9	3520.42	45.1492	10	3462.56	24.3875
11	3410.49	16.5187	12	3378.67	1.90447	13	3351.68	8.58343	14	3280.32	11.0594	15	3171.36	-2.23836
16	3078.8	3.58704	17	2815.56	40.9226	18	2696.96	44.7357	19	2352.73	61.1997	20	2206.17	67.455
21	1655.59	13.6037	22	1592.91	24.7034	23	1471.42	36.9175	24	1407.78	17.6108	25	1202.4	67.1967
26	1136.83	57.4811	27	1029.8	44.4016	28	931.45	43.5707	29	798.385	73.7496	30	667.25	35.8035
31	568.896	3.43584	32	503.33	27.3278	33	476.331	7.615	34	437.762	21.2608			

Fig.6: I.R Spectra of Metformin Hcl with Gum Karaya

Metformin Hcl release from the microcapsules was studied in 0.1 Hcl (pH1.2) for 12 hours as prescribed for Metformin Hcl tablets in USP XXIV. Metformin Hcl release from the microcapsules was slow, spread over extended period of time and depended on the composition of the coat and method employed for the preparation of microcapsules (Fig 7).
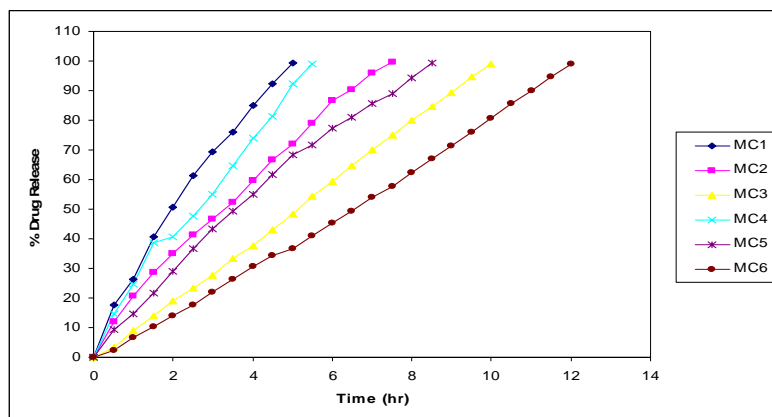

Fig.7: Dissolution Profiles of the drug release from the mucoadhesive microcapsules of Metformin Hcl

Table 2: Correlation coefficient (R) values in various kinetic models tested to describe drug release from the mucoadhesive microcapsules of Metformin Hcl

Formulation	Correlation coefficient values				n
	Zero order	First order	Higuchi model	Korsmeyer-Peppas model	
MC1	0.9573	0.8389	0.9826	0.9927	0.5958
MC2	0.9816	0.8670	0.9608	0.9832	0.6470
MC3	0.9853	0.8179	0.9571	0.9856	0.6701
MC4	0.9519	0.8727	0.9831	0.9926	0.6074
MC5	0.9547	0.8906	0.9823	0.9960	0.6169
MC6	0.9666	0.8448	0.9738	0.9967	0.7143

n: Diffusional exponent derived from Peppas equation

The model that best fits the release data was evaluated by correlation coefficient (r). The correlation coefficient (r) value was used as criteria to choose the best model to describe the drug release from the microcapsules. The r value in various models is given in Table 2.

In most of the formulated microcapsules the r values were higher in zero order models than that of first order model indicating the drug release from the most of the microcapsules was according to zero order kinetics. To analyze the mechanism of release of drug from the microcapsules the equation,$Q = Kt^n$ was used, where Q is the percentage of drug released; t is the release time; K is a constant incorporating structural and geometric characteristics of the release device, n is the release exponent indicative of mechanism of release.

When n approximates to 0.5, a fickin/diffusion control release is implied, where $0.5 < n < 1$ non fickin transport and $n = 1$ for zero order release [17]. The drug release mechanism from the microcapsules was non fickin transport as n value is in between 0.59 to 0.72 The drug release from the selected formulation followed zero order kinetics and controlled by Korsmeyer-Peppas mechanism. The wash-off test was conducted upto 8hrs at pH 1.2. The wash-off was slow in the case of microcapsules containing alginate-Gum Karaya as coat increases when compared to that of ethylene vinyl acetate microcapsules (Table 3). Hence the results indicate that the microcapsules with a coat consisting of alginate and Gum Karaya exhibited good mucoadhesive properties in the in vitro wash-off test when compared to non-mucoadhesive material, ethylene vinyl acetate microcapsules.

Table 3: In vitro Wash-Off test data of Metformin Hcl microcapsules formulated with Gum Karaya by employing different techniques

Formulation	0.1 Hcl (pH 1.2) and time in hours				
	1 hr	2 hr	4 hr	6 hr	8 hr
MC1	76	64	52	26	18
	(1.8)*	(1.2)	(1.0)	(1.6)	(1.5)
MC2	80	72	56	40	25
	(0.7)	(1.2)	(1.8)	(1.4)	(2.0)
MC3	94	80	75	53	32
	(1.0)	(1.8)	(1.1)	(1.5)	(1.0)
MC4	94	80	56	38	22
	(1.5)	(1.8)	(1.5)	(1.2)	(1.8)
MC5	96	84	64	48	35
	(1.8)	(1.2)	(1.8)	(2.0)	(1.8)
MC6	98	88	80	68	49
	(1.5)	(1.8)	(1.2)	(1.8)	(2.0)
EVA	95	60	20	95	60
	(1.8)	(2.0)	(1.0)	(1.8)	(2.0)

*Figures in parentheses are coefficient of variation (CV) values

CONCLUSION

The micro capsules prepared with a coat consisting of alginate and gum karaya by emulsification gelation process exhibits spherical , free flowing , good entrapment and exhibit good mucoadhesive property hence these microcapsules was slow and extended release over prolonged periods of time and depended on composition of the coat. Drug release was diffusion controlled and followed zero-order kinetics. These mucoadhesive microcapsules are, thus, suitable for oral controlled release of Metformin Hcl.

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