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MUCOADHESIVE MICROSPHERES- A PROMISING CARRIER IN DRUG DELIVERY OF ROSIGLITAZONE MALEATE FORMULATION

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Abstract: The aim of present study is development of mucoadhesive microspheres of Rosiglitazone maleate by combine the potential advantages of mucoadhesion with controlled drug delivery using various ratio of polymers.To achieve optimum therapeutic drug level over extended period of time, a controlled release system in the form of mucoadhesive microspheres will be developed using widely accepted and physiologically safe excipient and using simple techniques and reproducible methodologiesMucoadhesive microspheres of Rosiglitazone maleate are designed to increase its residence time in stomach by interacting with mucus membrane. Mucoadhesive microspheres of Rosiglitazone maleate will be prepared by using various ratios of carbopol-934, sodium carboxy methyl cellulose and sodium alginate by emulsification solvent evaporation techniques.

Key words: Formulation, Rosiglitazone maleate, Mucoadhesive microspheres

INTRODUCTION:

Drug action can be improved by developing new drugdelivery system, such as the mucoadhesive microspheredrug delivery system. These systems remain in close contact with the absorption tissue, the mucous membrane, releasing the drug at the action site leading to a bioavailability increase and both local and systemic effects[1].The oral route of drug administration constitutes the most convenient and preferred means ofdrug delivery to systemiccirculation of body. However oral administration of mostof the drugs in conventionaldosage forms has short-term limitations due to their inability torestrain and localize the system at gastro-intestinal tract.Microspheres are the carrier linked drugdelivery system in which particle size is ranges from 1-1000µm range in diameter having a core of drug and outer layers of polymer as coating material. The success of these microspheres is limited due to their short residence time at site of absorption. It would, therefore be advantageous to have means for providing an intimate contact of the drug delivery system withthe absorbing membrane. This can be achieved by coupling bio adhesion characteristics to microspheres and developing "mucoadhesive microspheres". Mucoadhesive microspheres have advantages like efficient absorption and enhanced bioavailability of the drugs due to a high surface to volume ratio, a much more intimate contact with the mucus layer and specific targeting of drugs to the absorption site[2]. Microspheres, as carrier for drug is one such approachwhich can be used in a sustained controlled release fashion [3]. Microspheres are small spherical particles, with diameters inthe micrometerrange (typically 1 µm to 1000 µm). Microspheres are sometimes referred to as microparticles [4]. Recent

advances in polymer science and drug carrier technologies have promulgated the development of novel drug carriers such as mucoadhesive microspheres that have boosted the use of bio adhesion in the drug delivery[5].Mucoadhesive microspheres include microparticles and microcapsules of 1 to 1000 μmin diameter consisting either entirely of mucoadhesive polymer or having an outer coating with adhesive property[6].Microspheres have the potential to be used for controlled as well as spatial drug delivery. Incorporating mucoadhesive tomicrospheres leads to efficient absorption and enhancedbioavailability of drug. Specific targeting of drug to the absorption site is achieved by using homing devices (ligand) like plant lectin, bacterial adhesion etc. on the surface of the microspheres. Mucoadhesive microspheres can be tailored to adhere to mucosal linings of GIT, thus offering the possibilities of localized as well as systemic absorption of drug in controlled manner[7, 8].

The aim of present study is development of mucoadhesive microspheres of Rosiglitazone maleate by combine the potential advantages of mucoadhesion with controlled drug delivery using various ratio of polymers which provide the drug for a prolonged gastrointestinal absorption in a sustained release manner. Rosiglitazone maleate is an antidiabetic drug belongs to thiazolidinediones class that can precisely control the blood glucose level in diabetic patient. It requires control release owing to its short biological half-life of 3-4 hours. It has many side effects like cardiovascular failure, fatigue, weight gain and mild anemia.

To overcome all above side effects and to reduce the dosing frequency of drug, it's necessary to develop a newer and safer formulation which release drug to the body for complete and prolonged duration. Thus, the development of controlled release dosage forms of Rosiglitazone maleate would be clearly advantageous that release the drug slowly for prolonged duration of action i.e. in sustained release manner.More ever the site of absorption of rosiglitazone maleate is in stomach. Dosage forms that are retained in the stomach would increase the absorption, improved drug efficiency and decrease dose requirements.

MATERIALS AND METHODOLOGY

MATERIALS

Rosiglitazone maleate is gift sample from Indswift Labs Pvt Ltd, Baddi (H.P.). Carbopol-934is procured fromColorcon Asia Pvt Ltd, Goa. Remaining all polymers and chemicals procured from Central Drug House, India.

METHODOLOGY

METHODS

Method of preformulation study:

Identification of drug-

The sample of Rosiglitazone maleate will be obtained as gift sample from Indswift Lab Pvt Ltd, Baddi (H.P.), identified and characterize as per requirement of official standards.

Infrared spectrum: The scanning will be done using KBr dispersion pellets. About 1 mg of powdered drug mixed with approximately 100 mg of KBr (spectroscopic grade) in a glass morter. The mixture will be compressed into transparentdrotes with special designed moisture free atmosphere and IR spectra will be obtained on IR spectrophotometer. The scanning will be done between 4000-400 cm-1. The spectra so obtained will be compared with reported in official compendia. The IR spectra of Rosiglitazone maleate. Characteristics peaks attributable to functional groups present in the molecule of drug assigned to establish the identity.

Ultraviolet spectrum:

10 mg of Rosiglitazone maleate will be accurately weighed and transferred in a 100 ml volumetric flask containing 20 ml of 0.1 N HCl. The drug will be dissolved by gentle shaking and volume made up to 100ml with 0.1 N HCl to make standard solution A. Then 10 ml of this solution will be transferred into a 100 ml volumetric flask and the volume will be made up to 100 ml with 0.1 N HCl to make standard stock solution B of 10 µg/ml concentration. In order to determine the absorption maxima (λmax) for Rosiglitazone maleate stock solution B will be scanned between 200 to 400 nm using by Shimadzu pharmspec UV-1700 spectrophotometer.

Melting point

Melting point of the Rosiglitazone maleate will be determined by using thieles tube method. 300ml of heavy paraffin will be filled in thieles tube, the drug filled (small amount) in a capillary tube whose one end is sealed with the help of flame, will be tied with a thermometer and will be suspended in thieles tube filled with paraffin. The heating will be started and the point at which drug start melting will be noted, and the melting point of drug will be found to be 1220 C.

Partition coefficient:

The partition coefficient is defined as the ratio of unionized drug distributed between the organic phase and aqueous phase at equilibrium. For a drug delivery system, lipophilic/hydrophilic balance has shown to be a contributing factor for the rate and extent of drug absorption. Partition coefficient provides a means of characterizing lipophilic/hydrophilic nature of drug. The measurement of drug lipophilicity and indication of its ability to cross the lipoidal cell membrane is the oil/water partition coefficient in systems such as octanol/water, octanol/0.1 N HCl etc.

Procedure: The partition coefficient of drug (Rosiglitazone maleate) will be determined in solvent system Octanol/0.1 N HCl. Accurately weighed quantity of drug (10 mg) taken in one stoppered glass vial containing 5 ml of octanol, 5ml of 0.1 N HCl will be added to the vial. Then the glass vialwill be kept to equilibrate by shaking in vortex mechanical shaker for 24 hours and after shaking, the vial containing materials will be transferred into a separating funnel, kept overnight at room temperature for equilibrium. After appropriate dilutions, the aqueous phase will be analyzed for Rosiglitazone maleate against blank solution using shimadzu-1700 UV spectrophotometer at 242 λmax. The drug concentration in octanol phase will be determined by subtracting the amount in aqueous phase from the total quantity of drug added to the vials.

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Solubility determination of Rosiglitazone maleate:

The solubility analysis will be carried out according to the method of Higuchiand Connors. In brief, solubility studies of Rosiglitazone maleate will be performed in different solvent(e.g.alcohol, distilled water, methanol, DimethylSulphoxide, 0.1NHCletc).Saturated solutions of drug will be prepared in different solvent by adding the excess drug to the vehicles and shaking in screw capped tubes on the shaker for 48 hrs at 25^0C under constant vibration. After this period the solutions will be filtered, diluted and analyzed by U.V Spectrophotometer.

Compatibility studies:

The drug and polymer compatibility will be characterized by the means of FTIR spectroscopy. The compatibility will be checked by making physical mixture of drug and polymer(1:1ratio) and then the FTIR analysis of the mixture will be done to check the compatibility of drug with polymer and the peaks will not be changed in mixture FTIR, and it will shows similar peaks like pure drug FTIR.

Method of analysis of drug:

a) Construction of calibration curve of Rosiglitazonemaleate

Rosiglitazone maleate will be estimated by U.V. Sectrophotometer (ShimadzupharmspecUV-1700 UVdouble beam spectrophotometer) method. Pure Rosiglitazone maleate will be taken and the solutions will be prepared by using 0.1N HCl as solvent, viz.1 µg/ml, 2µg/ml, 3µg/ml, 4µg/ml, 5µg/ml, 6µg/ml, 7µg/ml, 8µg/ml, 9µg/ml and 10µg/ml. The absorbance will be measured by using U-Vspectrophotometer at 242nm(λ_{max}) using 0.1NHCl as blank. Straight line will be obtained for a drug concentration of 1µg/ml to10µg/ml. The drug obeys the Beers law between 1-14µg/ml concentrations. Same procedure will be used for the preparation of standard curve of Rosiglitazone maleate in 2.5 pH phosphate buffer.

b) Determination of interference of different polymers in estimation of Rosiglitazone maleate

Polymers used in the formulation of Rosiglitazone maleate mucoadhesive microspheres, may interfere in the estimation of drug. Hence, the interference due to these polymers will be checked using the maximum concentration used in the formulation. Polymers will be dissolved in the standard solution of Rosiglitazone maleate prepared in 0.1NHCl. This solution will be filtered and 5ml of filtrate will be suitably for different polymers solution with Rosiglitazone maleate.

Method of Preparation of Formulation:

Method of preparation of microspheres without drug:

The 20ml of aqueous polymer solution will be added drop wise to 200 ml of liquid paraffin containing 0.5% span20 as an emulsifying agent with constant stirring. The constant stirring will be carried out using magnetic stirrer. The beaker and its content will be heated at 80^0C with constant stirring for 4.5 hours until the aqueous phase will be completely removed by evaporation. The liquid paraffin will be decanted and collected microsphere will be will

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behed 3times with 100ml of n-hexane, filtered through whattman's filter paper, dried in an oven at 50^0C for 2hours and stored in a desiccater at room temperature.

For the purpose of assessing the effect of stirring speed and effect of polymer concentrations on particle size of microspheres, every batches will be prepared at two different stirring speeds(500 and1000 rpm) and at two different polymer concentrations(2% and1% conc.).

Table 1:Batch specifications of microspheres without drug for accessing effect of stirring speed on particle size of microspheres

Formulation code	**SodiumCMC**	**Carbopol-934**	**Sodium alginate**
MS1	1000mg	--	--
MS2	--	1000mg	--
MS3	--	--	1000mg
MS4	500mg	500mg	--
MS5	500mg	--	500mg
MS6	--	500mg	500mg

All batches will be prepared at 2% polymer concentration and every batch will be prepared at two different stirring speeds (500rpm and1000rpm)

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Table 2: Batch specifications of microspheres without drug for accessing the effect of polymer concentration on the particle size of microspheres

Formulation code	SodiumCMC	Carbopol-934	Sodium alginate
MC1	1000mg	--	--
MC2	--	1000mg	--
MC3	--	--	1000mg
MC4	500mg	500mg	--
MC5	500mg	--	500mg
MC6	--	500mg	500mg

All batches will be prepared at 500rpm stirring speed and every batch will be prepared at two different polymer concentrations (2%,1%).

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Method of preparation of microspheres with drug:

Drug loaded microsphere will be prepared by water in oil(w/o) emulsification solvent evaporation method. Forthis,100mg of drug dissolved in 5ml dimethylsulfoxide and then it will be dispersed into 45 ml of 2% aqueous polymer solution. A vortex homogenizer will be used for rapid mixing of the drug solution into the aqueous polymer solution for 3minutes. Then drug and polymer solution will be added dropwise to 400ml of the liquid paraffin containing 0.5% span20 as an emulsifying agent with constant stirring at 500rpm. The constant stirring will be carried out using magnetic stirrer. The beaker and its content will be heated at 80^0C with constant stirring for 4.5 hrs until the aqueous phase will be completely removed by evaporation. The liquid paraffin will be decanted and collected microsphere will be will behed5 times with 100ml of n-hexane, filtered through whattman's filter paper, dried in hot air oven at 50^0C for 2hrs and stored in a desicator at room temperature.

For each formulation, 3 batches of microspheres will be prepared for the purpose of assessing the reproducibility of drug loading, particlesize, % mucoadhesion and in-vitro drug release by this method.

Table 3: Batch specifications of microspheres with drug for accessing the effect of polymer concentration on the particle size of microspheres

Formulation code	Drug	SodiumCMC	Carbopol-934	Sodium alginate
F1	100mg	900mg	--	--
F2	100mg	--	900mg	--
F3	100mg	--	--	900mg
F4	100mg	450mg	450mg	--
F5	100mg	450mg	--	450mg

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F6	100mg	--	450mg	450mg

All formulations will be prepared at 2% polymer concentration and 500rpm stirring speed. Span20 (0.5%) used emulsifying agent..

RESULTS AND DISCUSSION

The present study will be an attempt to develop and evaluate mucoadhesive microsphere of Rosiglitazone maleate. Emulsification solvent evaporation method will be used for preparation of mucoadhesive microsphere of rosiglitazone maleate using various ratios of sodiumCMC, sodium alginate and carbopol-934.

In order to develop mucoadhesive microsphere of rosiglitazone maleate, it will be first identified by characteristics Infrared spectra and ultraviolet absorption maxima(λ_{max}) and then it will be subjected to melting point determination, partition coefficient determination and solubility analysis. After that the drug will be analysed in UV Spectrophotometer by constructing standard curve in 0.1 N HCl. It will be found that the drug will be confirming the pharmacopoeial standards with respect to melting point, partition coefficient, solubility, wavelength of maximum absorption(λ_{max}) and the characteristics of I R spectra.

IDENTIFICATION OF DRUGBY I R SPECTRA:

The purity of drug sample will be identified by scanning the drug sample on IR spectrophotometer. The peaks of the IR spectra of drug sample will be found to be similar with the standard IR spectra of pure Rosiglitazone maleate as reported.

The I.R.spectra of mixture of drug and polymer indicated no incompatibility between drug and polymers, hence, carbopol-934, sodium CMC and sodium alginate will be chosen as polymers for further investigations. The spectra of drug shows absorption bands at 3436.23cm^{-1} due to N-H stretching, 2931.44 cm^{-1} due to C-H stretching, 1745.53cm^{-1} due to C=O stretching of carbonyl group, 1704cm^{-1} due to C=O stretching of carboxylic group, 1641.32cm^{-1} due to C=O stretching of amide group, 1352cm^{-1} due to C-N stretching, 1245cm^{-1} due to C-O stretching 1064cm^{-1} due to C-O-C stretching and 864cm^{-1} due to characteristics of disubstituted benzene which is also seen in the spectra of mixture of drug and polymers that mean there is no interaction between drug and polymer.

The plain sodiumCMC absorption peaks found at 1751.43cm^{-1}, 1419cm^{-1}, 1319.35 cm^{-1}, 1085.13 cm^{-1} and 1033 cm^{-1}. The plain carbopol-934 absorption peaks found at 2954.16cm^{-1}, 1712.31cm^{-1}, 1452.34 cm^{-1}, 1409 cm^{-1} and 1244cm^{-1}. The plain sodium alginate absorption peaks found at 12921.43cm^{-1}, 2850cm^{-1}, 1419.35 cm^{-1}, 1085.13 cm^{-1} and 1029 cm^{-1}.

IDENTIFICATION OF DRUG BY ULTRAVIOLETSPECTROSCOPY:

The U.V.Spectrophotometer showed absorption maxima(λ_{max}) of Rosiglitazone maleate in 0.1NHCl as follows.

Λ_{max} in 0.1 HCl - 242 nm λ_{max} in methanol - 242 nm Reported λ_{max} - 242nm

DETERMINATION OF INTERFERENCE OF POLYMER IN ESTIMATION OF DRUG:

Polymer used in formulation of microspheres, may interfere in the estimation of drug. Hence the interference due to these polymers will be checked by using the maximum concentration of polymers with drug solution. This polymer and drug solution will be filtered and then suitably diluted. This diluted solution will be subjectedtoUVscanningbetween220-270nm.The λ_{max} will be observed for different polymer solutions with drug. The data tells that there is no significant interference of polymers observed in estimation of drug.

MELTING POINT DETERMINATION:

The melting point of Rosiglitazone maleate will be found to be 122^0C which is close to the reported melting point of the drug given in official compendia(122-123^0C).

PARTITION COEFFICIENT DETERMINATION:

The partition coefficient of the Rosiglitazone maleate in n-octanol/water system will be found to be 194 which is indication of lipophilic nature of the drug.

SOLUBILITY DETERMINATION:

The solubility determination of Rosiglitazone maleate will be performed in different aqueous and organic solvent. Rosiglitazone maleate freely soluble in ethanol, methanol, 0.1 N HCl and dimethyl sulphoxide and insoluble in water.

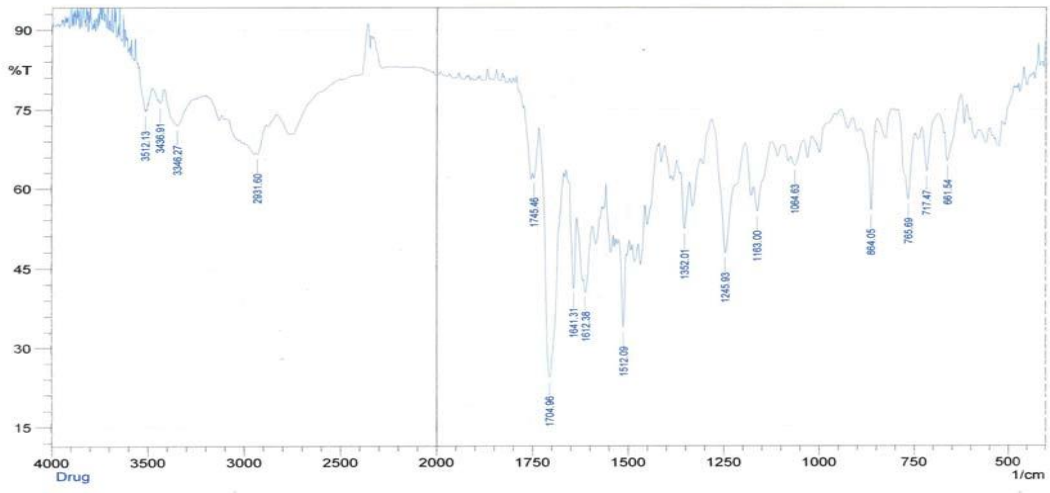

Figure 1:IR Spectrum of Rosiglitazone maleate

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Table 4:Interpretation of FTIR spectra of Rosiglitazone maleate

S.No.	Peak(cm^{-1})	Groups
1.	3436.23	N-H stretching
2.	2931.23	C-H stretching of benzene ring
3.	1745.33	C=O stretching in ketone
4.	1704.43	C=O stretching in carboxylic group
5.	1604.21	C=O stretching in amide group
6.	1352.45	C-N stretching
7.	1245.62	C-O stretching
8.	1064.11	C-O-C stretching
9.	864.18	di-substitution of benzene

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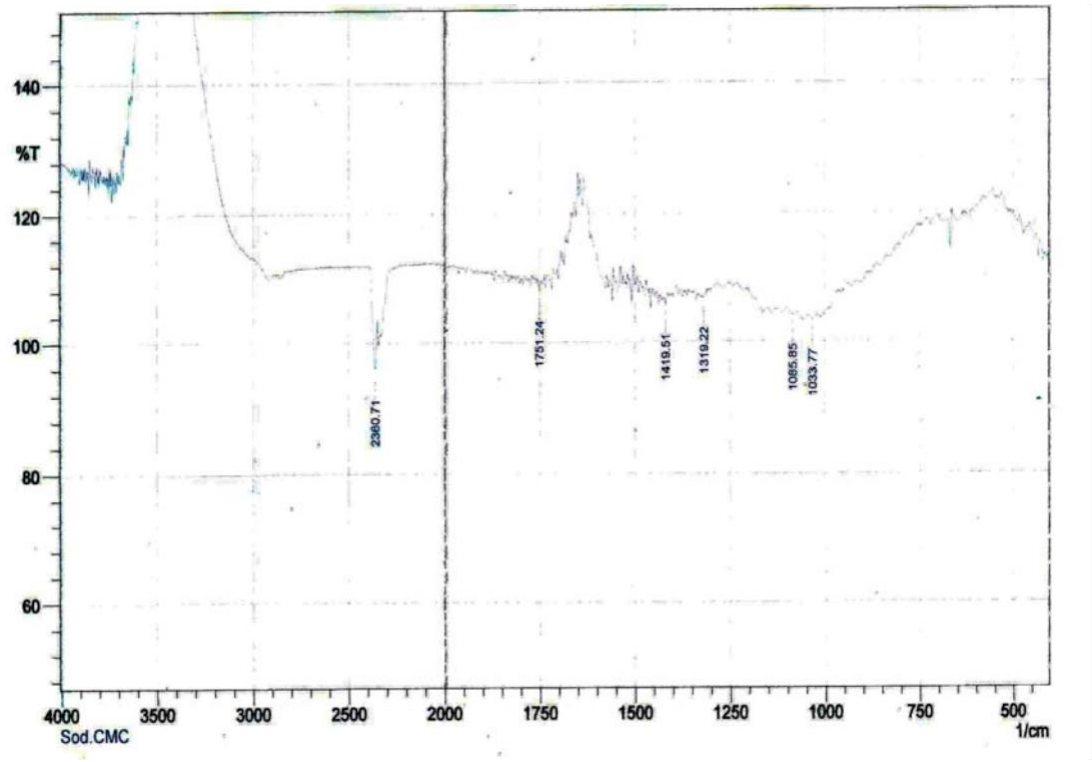

Figure 2:IR Spectrum of sodium carboxy methyl cellulose

Table 5:Interpretation of FTIR spectra of sodium carboxy methyl cellulose

S.No.	**Peak(cm^{-1})**	**Groups**
1.	3512.31	O-H stretching
2.	1751.12	C=O stretching
3.	1419.12	Carboxylate ion stretching
4.	1319.23	C-O stretching in sec.alcohol

5.	1085.26	C-O-C stretching
6.	1033.13	C-O stretching

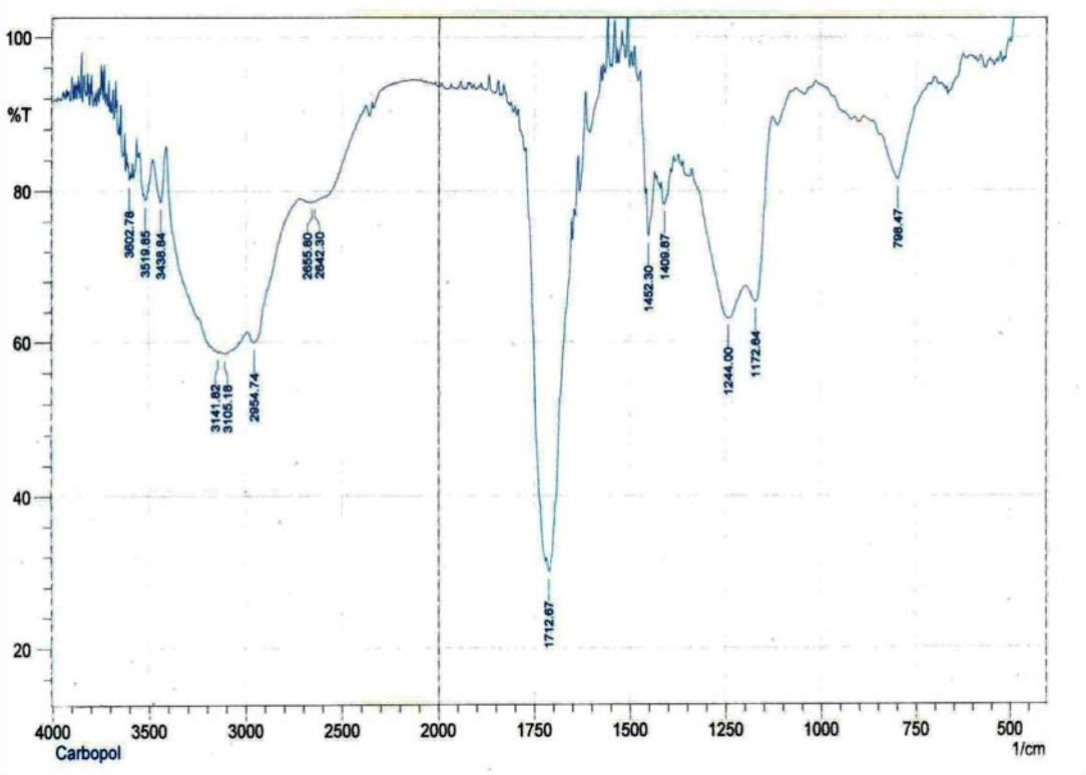

Figure 3:IR Spectrum of carbopol-934

Table 6:Interpretation of FTIR spectra of carbopol-934

S.No.	**Peak(cm^{-1})**	**Groups**
1.	3602.31	O-H stretching
2.	2954.12	C=H stretching

3.	1712.12	C=O stretching
4.	1452.23	C-H bending
5.	1409.26	C-OH bending
6.	1244.13	C-O stretching

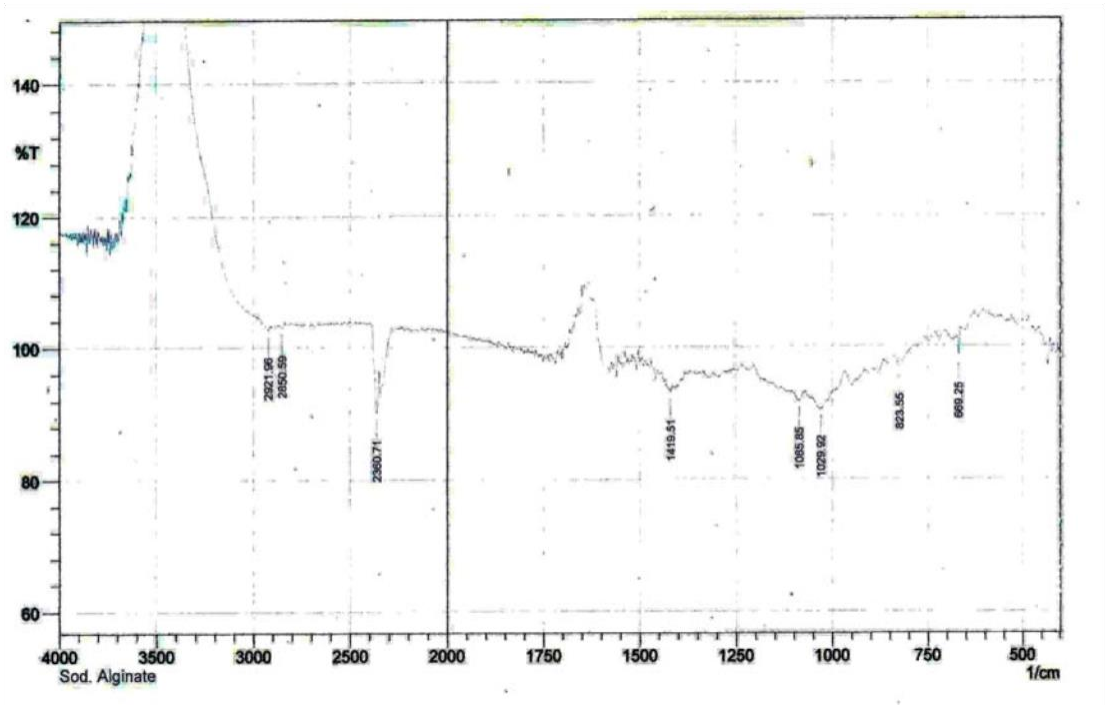

Figure 4:IR Spectrum of sodium alginate

Table 7:Interpretation of FTIR spectra of sodium alginate

S.No.	**Peak(cm^{-1})**	**Groups**

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1.	2921.31	O-H stretching
2.	2850.12	C=H stretching
3.	1419.12	Corboxylate ion stretching
4.	1085.26	C-O-C stretching
5.	1033.13	C-O stretching

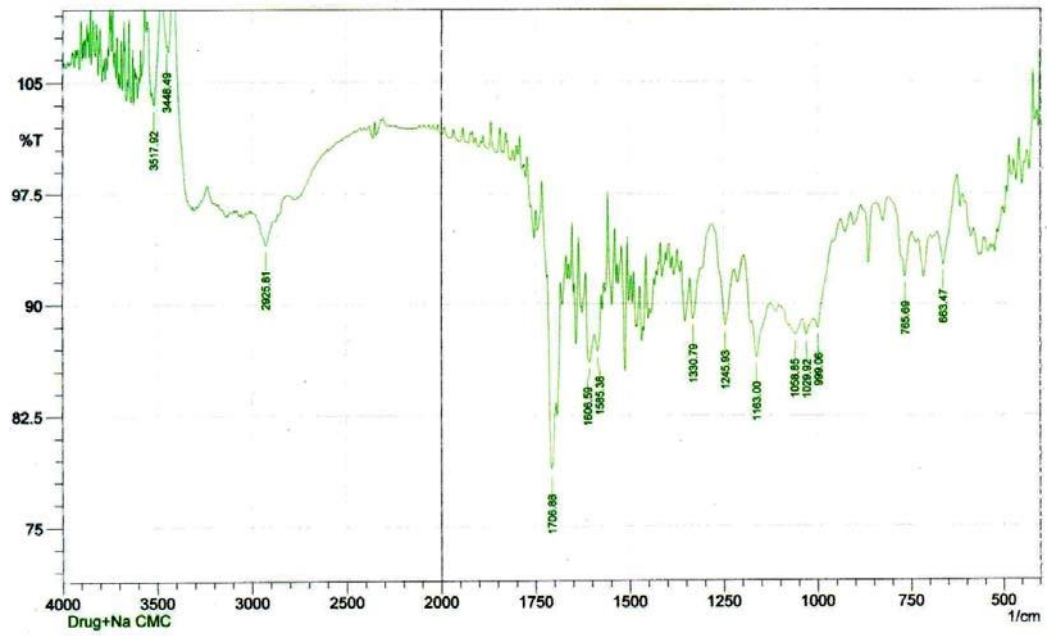

Figure 5:IR Spectrum of mixture of Rosiglitazone maleate and sodium carboxy methyl cellulose

Table 8:Interpretation of FTIR spectra of mixture of Rosiglitazone maleate and sodium carboxy methyl cellulose

S.No.	**Peak(cm^{-1})**	**Groups**

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1.	3436.23	N-H stretching
2.	2931.23	C-H stretching of benzene ring
3.	1745.33	C=O stretching in ketone
4.	1704.43	C=O stretching in carboxylic group
5.	1352.45	C-N stretching
6.	1245.62	C-O stretching
7.	1064.26	C-O-C stretching

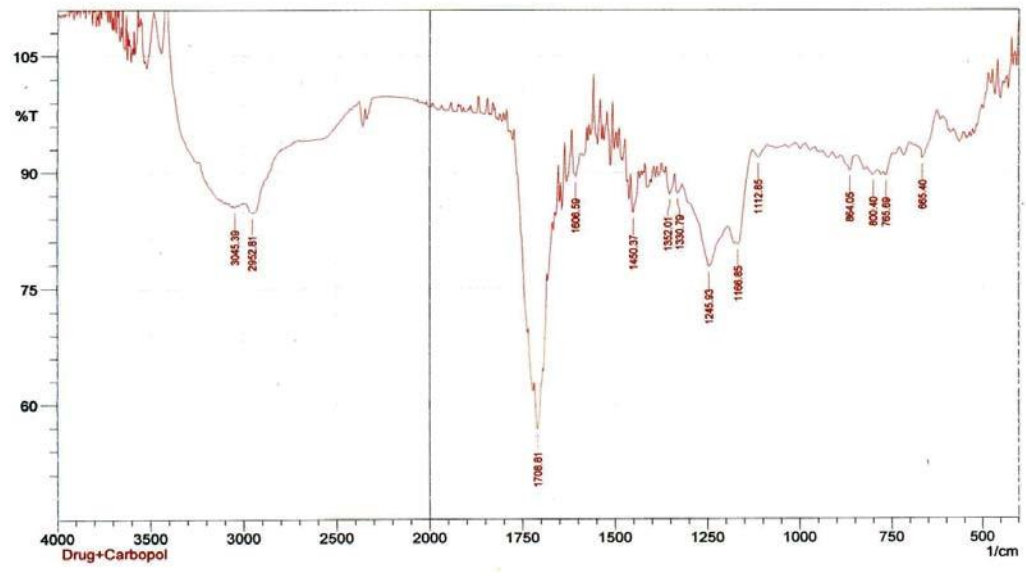

Figure 6:IR Spectrum of mixture of Rosiglitazone maleateand carbopol-934

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Table9: Interpretation of FTIR spectra mixture of Rosiglitazone maleate and carbopol-934

S.No.	Peak(cm^{-1})	Groups
1.	3436.23	N-H stretching
2.	2931.23	C-H stretching of benzene ring
3.	1745.33	C=O stretching in ketone
4.	1704.43	C=O stretching in carboxylic group
5.	1352.45	C-N stretching
6.	1245.62	C-O stretching
7.	1064.26	C-O-C stretching

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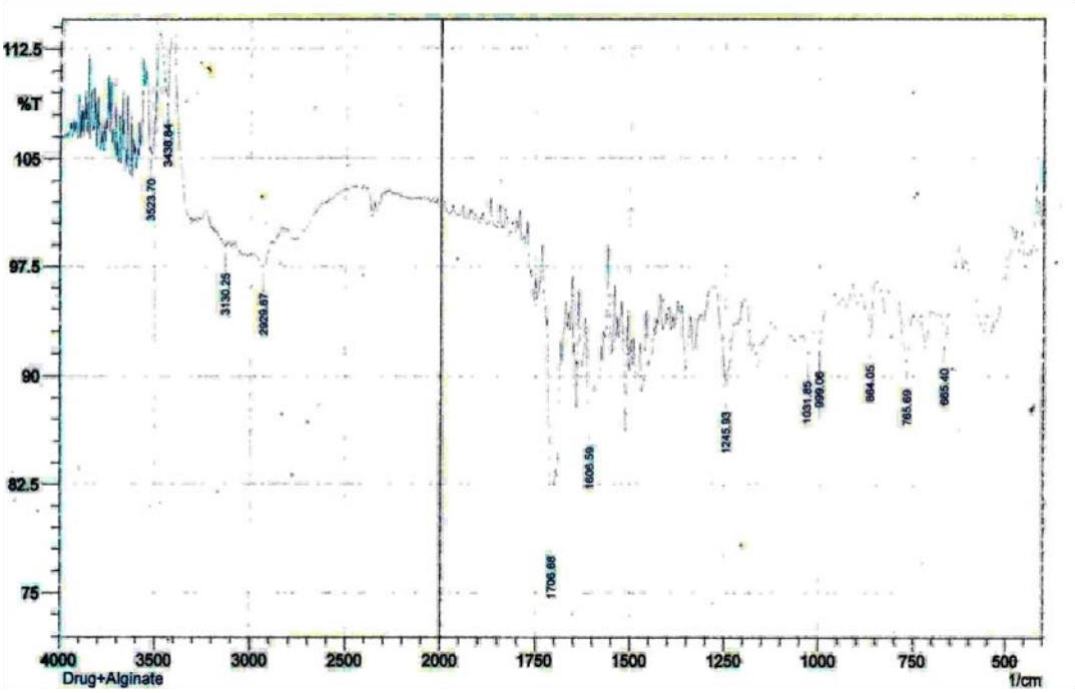

Figure 7: IR Spectrum of mixture of Rosiglitazone maleate and sodium alginate

Table 10:Interpretation of FTIR spectra mixture of Rosiglitazone maleate and sodium alginate

S.No.	Peak(cm^{-1})	Groups
1.	3436.23	N-H stretching
2.	2931.23	C-H stretching of benzene ring
3.	1745.33	C=O stretching in ketone
4.	1704.43	C=O stretching in carboxylic group

5.	1352.45	C-N stretching
6.	1245.62	C-O stretching
7.	1064.26	C-O-C stretching

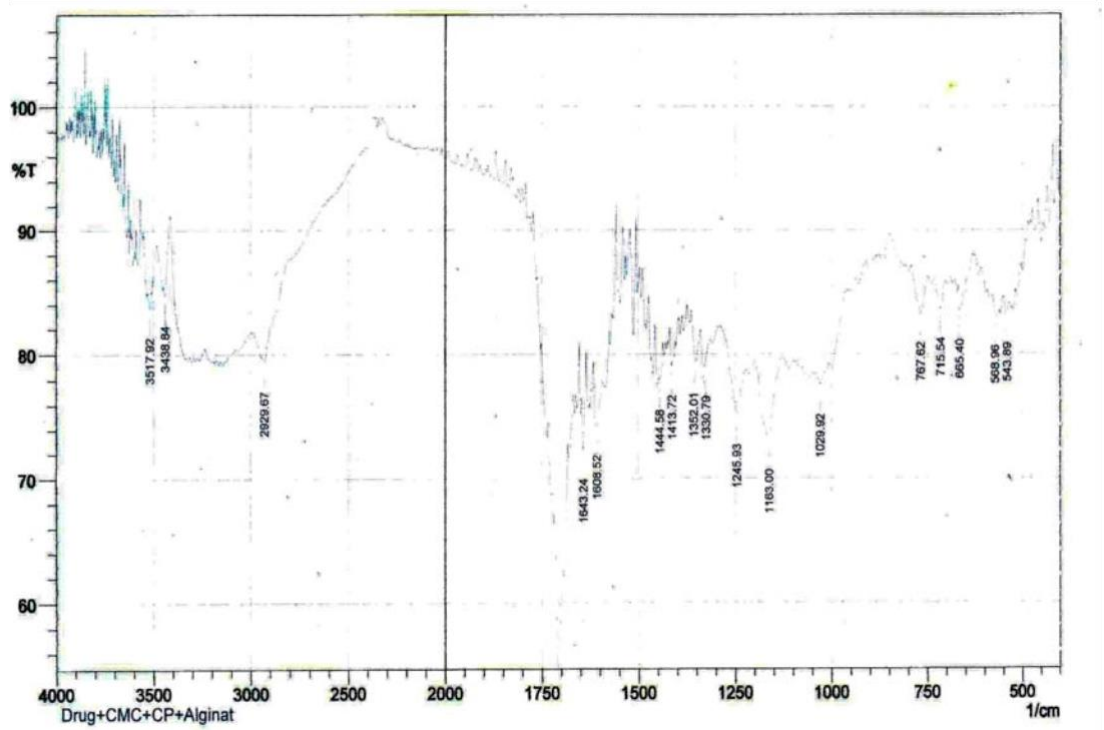

Figure 8:IR Spectrum of mixture of Rosiglitazone maleate, sodium carboxy methyl cellulose, carbopol-934 and sodium alginate

Table 11:IR Spectrum of mixture of Rosiglitazone maleate, sodium carboxy methyl cellulose, carbopol- 934 and sodiumalginate

S.No.	**Peak(cm^{-1})**	**Groups**
1.	3436.23	N-H stretching

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2.	2931.23	C-H stretching of benzene ring
3.	1745.33	C=O stretching in ketone
4.	1704.43	C=O stretching in carboxylic group
5.	1352.45	C-N stretching
6.	1245.62	C-O stretching
7.	1064.26	C-O-C stretching

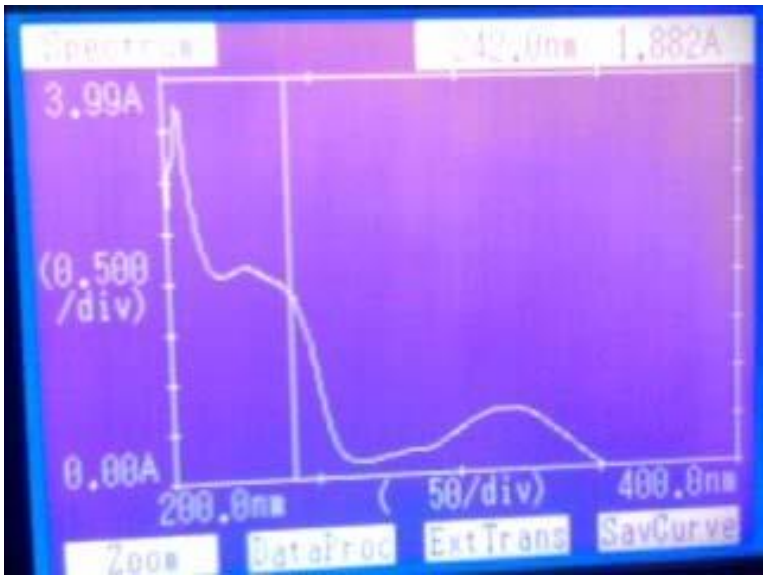

Figure 9 U V Spectrophotometer shows absorption maxima (λmax) of Rosiglitazone maleate at 242 nm

Table 12:Melting point of Rosiglitazone maleate

S.No.	Reported	Observed
1.	122-123^{0}C	122^{0}C

Table13:Partition coefficient of Rosiglitazone maleate in different oil in water system

S.No.	o/w system	Partition coefficient
1	Octanol/Water	194
2	Octanol/0.1nHCl	0.23
3	Cyclohexane/Water	0.32

Table14:Solubility o fRosiglitazone maleate in various solvent

S.No.	Solvent	Solubility
1	Ethanol	Freely soluble
2	Dimethylsulphoxide	Freely soluble

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3	Methanol	Freely soluble
4	0.1NHCl(pH1.2)	Freely soluble
5	Phosphate buffer(pH5.8)	Soluble
6	Water	Insoluble

Table 15: Standard curve of Rosiglitazone maleate in 2.5 pH phosphate buffer

S.No.	**Concentration(µg/ml)**	**Absorbance(nm)**
1	1	0.045
2	2	0.089
3	3	0.125
4	4	0.174
5	5	0.204

6	6	0.252
7	7	0.293
8	8	0.329
9	9	0.364
10	10	0.402

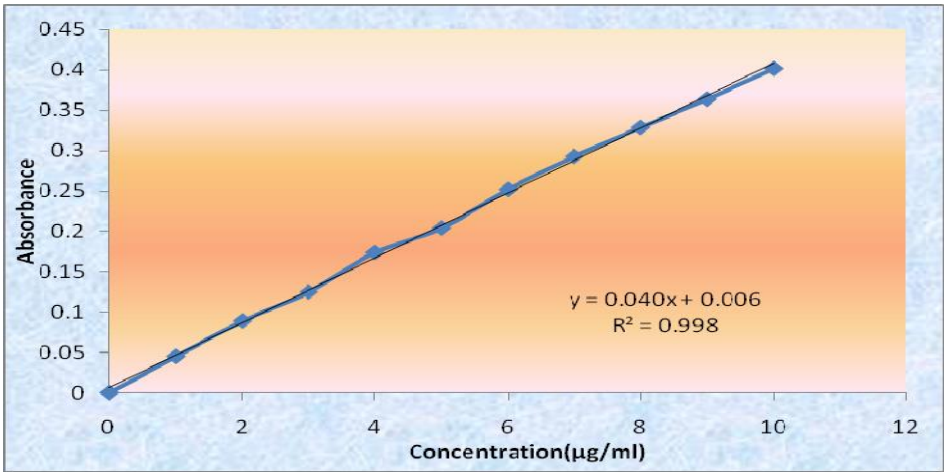

Figure 10:Standard curve of Rosiglitazone maleate in 2.5pHphosphate buffer

Table 16:Standard curve of Rosiglitazone maleate in 0.1NHCl

S.No.	**Concentration(µg/ml)**	**Absorbance(nm)**
1	1	0.034
2	2	0.063
3	3	0.099

4	4	0.135
5	5	0.18
6	6	0.213
7	7	0.249
8	8	0.285
9	9	0.324
10	10	0.363

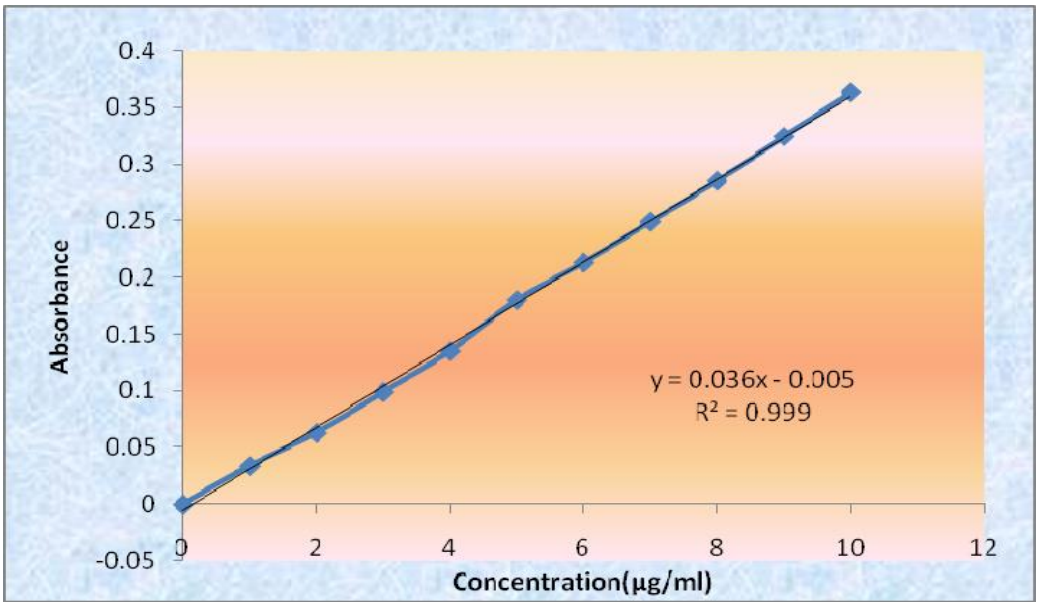

Figure 11:Standard curve of Rosiglitazone maleate in 0.1NHCl

CONCLUSION

Rosiglitazone maleate is an oral antidiabetic drug belongs to thiazolidinediones class that has been widely used in management of diabetes type-2. Being an antidiabetic, it is safe and efficacious candidate for treating diabetes type- 2. To achieve optimum therapeutic drug level over extended period of time, a controlled release system in the form of mucoadhesive microspheres will be developed using widely accepted and physiologically safe excipient and using simple techniques and reproducible methodologies Mucoadhesive microspheres of Rosiglitazone maleate are designed to increase its residence time in stomach by interacting with mucus membrane. Mucoadhesive microspheres of Rosiglitazone maleate will be

prepared by using various ratios of carbopol-934, sodium carboxy methyl cellulose and sodium alginate by emulsification solvent evaporation techniques.

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