"DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE''

Dissertation Submitted to

THE TAMIL NADU Dr.M.G.R. MEDICAL UNIVERSITY

Chennai-32

In Partial fulfillment for the award of the degree of

MASTER OF PHARMACY

Submitted by

Reg.No-261510264

Under the guidance of

Dr.V.KAMALAKKANNAN, M.Pharm.,Ph.D.

Associate Professor, Department of Pharmaceutics.

J.K.K.NATTRAJA COLLEGE OF PHARMACY
KOMARAPALAYAM-638183, TAMILNADU.
OCTOBER-2017

EVALUATION CERTIFICATE

This is to certify that the dissertation work entitled "**DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE**" submitted by student bearing **Reg.No-261510264** to The TamilNadu Dr. M. G. R. Medical University, Chennai, for the partial fulfillment of the degree of MASTER OF PHARMACY was evaluated by us during the examination held on……

Internal Examiner

External Examiner

CERTIFICATE

This is to certify that the work embodied in the dissertation "**DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE"** submitted to The Tamilnadu Dr. M. G. R. Medical University, Chennai, was carried out by **Reg.No-261510264** for the partial fulfillment of the degree of Master of Pharmacy in under direct supervision of **Dr.V.KAMALAKKANAN**, **M.Pharm.,Ph.D** Associate Professor, Department of Pharmaceutics, J.K.K.Nattraja College of Pharmacy, Komarapalayam, during the academic year 2016-2017.

Place: Kumarapalayam.
Date:

Dr. R. Sambathkumar., M.Pharm., Ph. D.,
Principal,
J.K.K. Nattraja college of Pharmacy,
Komarapalayam - 638183
TamilNadu

CERTIFICATE

This is to certify that the work embodied in this dissertation entitled "**DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE**" submitted to The Tamil Nadu DR. M.G.R. Medical University, Chennai, in partial fulfillment to the requirement for the award of degree of MASTER OF PHARMACY is a bonafide work carried out by **Reg.No-261510264** during the academic year 2016-2017, under my guidance and direct supervision in the department of Pharmaceutics, J.K.K.Nattraja College of Pharmacy, Kumarapalayam.

Dr.V.Kamalakkannan., M.Pharm.,Ph.D
Associate Professor,
Department of Pharmaceutics,
J.K.K.Nattraja college of Pharmacy,
Kumarapalayam-638183.
Tamil Nadu.

Dr.R.Sambathkumar., M.Pharm., Ph.D.,
Principal,
J.K.K.Nattraja college of Pharmacy,
Kumarapalayam – 638183
TamilNadu

Place : Kumarapalayam.
Date

:

CERTIFICATE

This is to certify that the dissertation entitled "**DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE**" is a bonafide work done by **Reg.No-261510264** J.K.K.Nattraja College of Pharmacy, in part and fulfillment of the university rules and regulation for award of Master of Pharmacy under my guidance and supervision during the academic year 2016-2017.

Dr.V.Kamalakkannan., M.Pharm.,Ph.D.,
Associate.Professor,
Department of Pharmaceutics,
J.K.K.Nattraja college of Pharmacy,
Kumarapalayam-638183.
Tamil Nadu.

Dr.R.Sambathkumar., M.Pharm., Ph.D.,
Principal,
J.K.K.Nattraja college of Pharmacy,
Kumarapalayam – 638183
TamilNadu

Dr.S.Bhama, M.Pharm., Ph.D.,
Head of the Department,
J.K.K.Nattraja College of pharmacy,
Kumarapalayam-638183
Tamil Nadu

DECLARATION

The work presented in this dissertation entitled, **"DESIGN AND DEVELOP BILAYERED ORAL SUSTAINED MATRIX TABLETS OF PIOGLITAZONE HYDROCHLORIDE AND METFORMIN HYDROCHLORIDE**" was carried out by me, under the direct supervision of **Dr.V. KAMALAKKANNAN, M.Pharm.,Ph.D** Associate.Professor, Department of Pharmaceutics, J.K.K.Nattraja College of Pharmacy, Kumarapalayam.

I further declare that, the work is original and has not been submitted in part or full for the award of any other degree or diploma in any other university.

Place: Kumarapalayam.

Reg.No-261510264

Date:


DEDICATED TO
MY BELOVED
FAMILY,
STAFFS AND
FRIENDS

ACKNOWLEDGEMENT

I am proud to dedicate my deep sense of gratitude to the founder, (Late) Thiru J.K.K. Nattaraja Chettiar, providing us the historical institution to study.

My sincere thanks and respectful regards to our reverent Chairperson Smt. N. Sendamaraai, B.Com., Managing Director Mr. S. Omm Sharravana, B.Com., LLB., J.K.K. Nattraja Educational Institutions, Komarapalayam for their blessings, encouragement and support at all times.

It is most pleasant duty to thank our beloved Principal Dr. R.SAMBATHKUMAR, M.Pharm., Ph.D., J.K.K.Nattraja College of Pharmacy, Komarapalayam for ensuring all the facilities were made available to me for the smooth running of this project.

I express my whole hearted thanks to my guide Dr.V.KAMALAKKANNAN M.Pharm.,Ph.D Associate.Professor , Department of Pharmaceutics, for suggesting solution to problems faced by me and providing indispensable guidance, tremendous encouragement at each and every step of this dissertation work. Without his critical advice and deep-rooted knowledge, this work would not have been a reality.

My sincere thanks to Dr. R. Shanmuga Sundaram, M.Pharm., Ph.D., Vice Principal and Professor and Head of the Department, Department of Pharmacology, Dr. Kalaiarasi., M.Pharm.,Ph.D Asst.Professor, Department of Pharmacology, for their valuable suggestions during my project work.

It is my privilege to express deepest sense of gratitude toward Dr.M. Senthilraja, M.Pharm., Ph.D., Professor and Head, Department of Pharmacognosy and Mrs. P. MeenaPrabha, M.Pharm.,Asst.Professor, Department of Pharmacognosy for their valuable suggestions during my project work.

My sincere thanks to Dr. M. Vijayabaskaran, M.Pharm., Ph.D., Assistant Professor

and head Department of Pharmaceutical chemistryMrs. S. Gomathi, M.Pharm., Lecturer, Department of Pharmaceutical chemistry and for their valuable suggestions and inspiration.

My sincere thanks to Dr.N. Venkateswaramurthy, M.Pharm.,Ph.D Professor and Head, Department of Pharmacy Practice. Mrs. K. Krishna Veni, M.Pharm.,Asst.Professor, Department of Pharmacy Practice, for their help during my project.

My sincere thanks to Dr.V.Sekar, M.Pharm., Ph.D., Professor and Head of The Department of analysis, , and Dr.I.Caroline nimila,M.Pharm.,Ph.D., Assistant Professor, Department of Pharmaceutical Analysis for their valuable suggestions.

My sincere thanks to Dr. S. Bhama, M.Pharm.,Ph.D Associate Professor, Mr. R. Kanagasabai, B. Pharm. M.Tech., Assistant Professor, Mr. K. Jaganathan, M.Pharm., Asst.Professor, Department of Pharmaceutics, Mr. C. Kannan M.Pharm., Asst.Professor, Department of Pharmaceutics for their valuable help during my project.

I greatly acknowledge the help rendered by Mrs. K. Rani, Office Superintendent, Mrs. V. Gandhimathi, M.A., M.L.I.S., Librarian, and Mrs. S. Jayakala, B.A., B.L.I.S., Asst. Librarian for their co-operation.

My special thanks to all the Technical and Non Technical Staff Members of the institute for their precious assistance and help.

Last, but nevertheless, I am thankful to my lovable parents and all my friends for their co-operation, encouragement and help extended to me throughout my project work.

Reg.No: 261510264

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBRIVATIONS

API	-	Active pharmaceutical ingredient
° C	-	Degree centigrade
Conc	-	Concentration
CDER	-	Center for Drug Evaluation and Research
cm	-	Centimeter
DT	-	Disintegration time
DC	-	Drug content
FDTs	-	Fast dissolving tablets
FT-IR	-	Fourier Transform Infrared
g	-	Gram
GIT	-	Gastrointestinal tract
hr	-	Hour
IR	-	Infra Red
IgE	-	Immuno globulin E
IP	-	Indian Pharmacopoeia
IODs	-	Intraoral drugs
Kg	-	kilo gram
KBr	-	Potassium bromide

m. p.	-	Melting Point
MDTs	-	Mouth dissolving tablets
min	-	Minutes
mg	-	milli gram
ml	-	milli liter
mm	-	milli meter
MCC	-	Microcrystalline cellulose
NDDS	-	novel drug delivery systems
nm	-	nano meter
ODTs	-	Orodispersible tablets
PEG	-	Poly ethylene glycol
pH	-	Hydrogen ion concentration
QD	-	Quick dissolve
QoL	-	Quality of life
rpm	-	Revolution per minute
sec.	-	Second

CONTENTS

1. INTRODUCTION

As very few drugs are coming out of research and development and already existing drugs are suffering the problem of resistance due to their irrational use and complications involved in marketing new drug entities have increased, and moreover the desire to maintain a near-constant or uniform blood drug levels, as well as enhanced clinical efficacy of the drug for its intended use made some drugs more effective by slight alteration in the drug delivery. With concomitant recognition of the therapeutic advantages of Sustained drug delivery and Controlled drug delivery, greater attention has been focused on development of Sustained release drug delivery systems and Controlled drug delivery systems.

Controlled Release Drug Delivery systems are the dosage forms designed to deliver the drug at a predetermined rate, locally or systemically, for a specified period of time.

Sustained Release Drug Delivery systems are those that provide medication over an extended period of time.

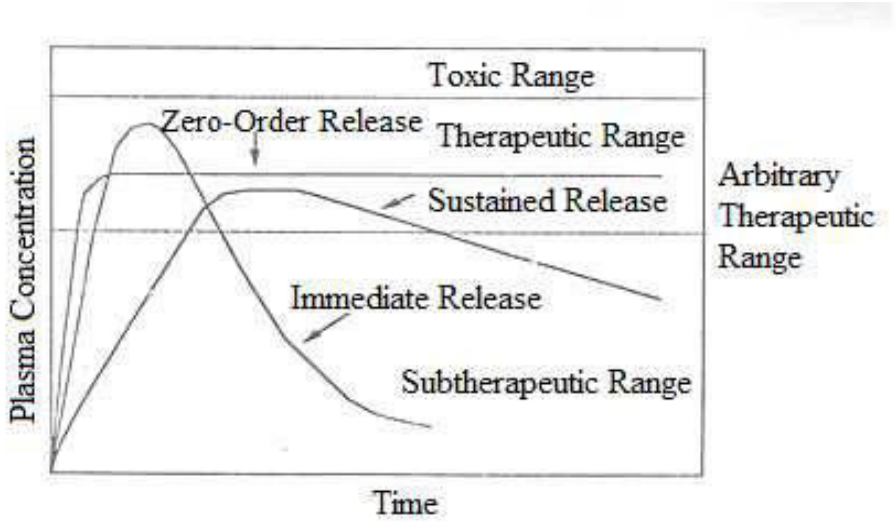


Figure :1 Comparative blood level profiles obtained from administration of conventional, controlled, and sustained release dosage forms.

The conventional tablet or capsule provides only a single and transient burst of drug. A pharmacological effect is seen as long as the amount of drug is within the therapeutic range. Problems occur when the peak concentration is above or below this range, especially for drugs with narrow therapeutic windows. The slow first order release obtained by a sustained release preparation is generally achieved by slowing the release of drug from a dosage form. In some cases this is accomplished by a continuous release process however systems that release small bursts of drug over a prolonged period can mimic the continuous release system.

1.1 Drawbacks of Conventional Dosage Forms

- Drugs with short half-life require frequent administration, which increases chances of missing dose of drug leading to poor patient compliance.
- A typical peak-valley plasma concentration-time profile is obtained which makes difficult to attainment of steady state condition.
- The unavoidable fluctuations in the drug concentration may lead to under medication or overmedication as the C_{SS} values fall or rise beyond the therapeutic range.
- The fluctuating drug levels may lead to precipitation of adverse effects especially of a drug with small therapeutic index, whenever overmedication occurs.

In order to overcome the drawbacks of conventional drug delivery systems, several technical advancements have led to the development of controlled and sustained drug delivery system. However despite of many advantages offered by oral sustained release dosage forms they have few disadvantages and Lag time is one of them. Sustained Release also provides promising way to decrease the side effects of the drug by

preventing the fluctuation of the therapeutic concentration of the drug in the body. There are several reasons for the attractiveness of these dosage forms (Ansel et al., 2003).

1.2 Sustained Release Drug Delivery Systems:

Sustained release drug delivery systems can be defined as any dosage form that prolongs the therapeutic activity of the drug by continuously releasing medication over an extended period of time. In absence of suitable clinical evidence of this therapeutic effect it can be defined as any dosage forms that give prolongation of the drug levels in the blood. The major advantage of this category is that, in addition to the convenience of reduced frequency administration, it provides levels that are devoid of the peak and valley effect (Altaf et al., 2003).

By providing smooth plasma level of drug over longer period of time, sustained - release drug delivery technology can minimize side effects, improve efficacy and by enabling once daily dosing- maximize patient compliance.

1.2.1 The goal in designing Sustained release delivery system is to:

- Reduce the frequency of dosing or to increase effectiveness of the drug by localization at the site of action, reducing the dose required, or providing uniform drug delivery.
- It would be a single dose for the duration of treatment whether it is for days or weeks, as with infection, or for the life time of the patient, as in hypertension or diabetes.
- It should deliver the active entity directly to the site of action, minimizing or eliminating side effects.

- This may necessitate delivery to specific receptors or to localization to cells or to specific areas of the body.

- The safety margin of high potency drug can be increase and the incidence of both local and systemic adverse side effects can be reduced in sensitive patient.

Introduction of matrix tablet as sustained release (SR) has given a new breakthrough for novel drug delivery system (NDDS) in the field of Pharmaceutical technology. It excludes complex production procedures such as coating and pelletization during manufacturing and drug release rate from the dosage form is controlled mainly by the type and proportion of polymer used in the preparations. Hydrophilic polymer matrix is widely used for formulating an SR dosage form. Matrix system is widely used for the purpose of sustained release. It is the release system which prolongs and controls the release of the drug that is dissolved or dispersed. In fact, a matrix is defined as a well-mixed composite of one or more drugs with gelling agent i.e. hydrophilic polymers. By the sustained release method therapeutically effective concentration can be achieved in the systemic circulation over an extended period of time, thus achieving better compliance of patients. Numerous SR oral dosage forms such as membrane controlled system, matrices with water soluble/insoluble polymers or waxes and osmotic systems have been developed, intense research has recently focused on the designation of SR systems for poorly water soluble drugs.

Various drug delivery techniques have been developed to sustain the release of drugs, including triple-layered tablets (Geomatrix® technology) and osmotic pumps with laser drilled holes (OROS® technology). These technologies are intricate and relatively expensive to manufacture. Thus, there remains an interest in developing novel formulations that allow for sustained release of drugs using readily available, inexpensive excipients.

Sustained Release systems generally don't attain zero order type release and usually try to mimic zero order release by providing drug in a slow first order. Repeat action tablet are an alternative method of sustained release in which multiple doses of drug are an alternative method of sustained release, in which, multiple doses are contained within a dosage form and each dose is released at a periodic interval. Delayed release system, in contrast, may not be sustaining, since often the function of these dosage forms is to maintain the drug in the dosage for some time before release, for example; Enteric coated tablet.

The ideal way of providing an exact amount of drug at the site of action for a precise time period is usually approximated by most systems. This approximation is achieved by creating a constant concentration in the body or an organ over an extended time; in other words, the amount of drug entering the system is equivalent to the amount of drug removed from the system. All forms of metabolism and excretion are included in the removal process; urinary excretion, enterohepatic recycling, sweat, fecal and so on. Since, for most of the drugs these elimination processes are first order, it can be said that a certain blood level, the drug will have a specific rate of elimination. The idea is to deliver drug at this exact rate for an extended period. This is represented mathematically as following,

$$\text{Rate in} = \text{Rate out} = k\text{elim} \times Cd \times Vd$$

Where *Cd* is the desired drug level, *Vd* is the volume of distribution, and *k* elim is the rate constant of drug elimination from the body. Often such exacting delivery rates prove to be difficult to achieve by administration routes other than intravenous infusion. Non-invasive routes, for example. Oral are obviously preferred.

1.2.2 Designing Sustained Release Drug Delivery Systems:

Most of the orally administered drugs, targeting is not a primary concern and it is usually intended for drugs to penetrate to the general circulation and perfuse to other body tissues. For this reason, most systems employed are of the sustained release variety. Sustained release dosage forms usually consists of two parts; an immediately available dose to establish blood level quickly and a sustained part that contains several times the therapeutic dose for predicted drug levels (Robinson JR et al., 2005).

Several are available to add the immediately available portion to the sustaining part. Simple addition of non-sustained portion to the tablet is the most direct method. Placement of initial dose in the tablet coat with the sustaining portion in the core represents an alternate approach. More common methods that are used to achieve sustained release of orally administered drugs are as follows:

1. Dissolution Controlled Systems
    - Encapsulation dissolution control
    - Matrix Dissolution control
2. Diffusion Controlled Systems
    - Reservoir devices
    - Matrix devices
3. Diffusion and Dissolution Controlled Systems
4. Ion exchange resins
5. pH independent formulations

It is assumed that increasing concentration at the absorption site will increase circulating blood levels, which in turn, promotes greater concentration of drug at the site of action. If toxicity is not an issue, therapeutic levels can thus be extended. In essence, drug delivery by these systems usually depends on release from some type of dosage

form, permeation through biological milieu and absorption through an epithelial membrane to the blood.

There are a variety of both physicochemical and biological factors that come into play in the design of such system.

Biological factors influencing oral sustained-release dosage form design:

- Biological half life.
- Absorption.
- Metabolism.
- Side effects

Biological half life:

The usual goal of an oral SR product is to maintain therapeutic blood levels over an extended period of time. To achieve this, drug must enter the circulation at approximately the same rate at which it is eliminated. The elimination rate is quantitatively described by the half-life (t1/2). Each drug has its own characteristic elimination rate, which is the sum of all elimination processes, including metabolism, urinary excretion and all over processes that permanently remove drug from the blood stream. Therapeutic compounds with short half-life are generally are excellent candidate for SR formulation, as this can reduce dosing frequency. In general, drugs with halflives shorter than 2 hours such as furosemide or levodopa are poor candidates for SR preparation. Compounds with long half-lives, more than 8 hours are also generally not used in sustaining form, since their effect is already sustained. Digoxin and phenytoin are the examples.

Absorption:

Since the purpose of forming a SR product is to place control on the delivery system, it is necessary that the rate of release is much slower than the rate of absorption. If we assume that the transit time of most drugs in the absorptive areas of the GI tract is about 8-12 hours, the maximum half-life for absorption should be approximately 3-4 hours; otherwise, the device will pass out of the potential absorptive regions before drug release is complete. Thus corresponds to a minimum apparent absorption rate constant of 0.17-0.23h^{-1} to give 80-95% over this time period. Hence, it assumes that the absorption of the drug should occur at a relatively uniform rate over the entire length of small intestine. For many compounds this is not true.

If a drug is absorbed by active transport or transport is limited to a specific region of intestine, SR preparation may be disadvantageous to absorption. One method to provide sustaining mechanisms of delivery for compounds tries to maintain them within the stomach. This allows slow release of the drug, which then travels to the absorptive site. These methods have been developed as a consequence of the observation that co-administration results in sustaining effect. One such attempt is to formulate low density pellet or capsule. Another approach is that of bioadhesive materials.

Metabolism:

Drugs those are significantly metabolized before absorption, either in the lumen or the tissue of the intestine, can show decreased bioavailability from slower-releasing dosage form. Hence criteria for the drug to be used for formulating Sustained-Release dosage form is,

- Drug should have low half-life(<5 hrs)
- Drug should be freely soluble in water.
- Drug should have larger therapeutic window.

- Drug should be absorbed throughout the GIT.

Even a drug that is poorly water soluble can be formulated in SR dosage form. For the same, the solubility of the drug should be increased by the suitable system and later on that is formulated in the SR dosage form. But during this the crystallization of the drug, that is taking place as the drug is entering in the systemic circulation, should be prevented and one should be cautious for the prevention of the same.

Side Effects:

It is believed that for some drugs, the incidence of side effects is a function of plasma concentrations. Theoretically, the incidence of side effects can be minimized by controlling the concentration at which the drug exists in plasma at any given time, and hence controlled release formulations appear to offer a solution to this problem.

Some other factors that influence include Duration of action, Margin of Safety, Role of Disease state and Role of Circadian Rhythm.

Physicochemical factors influencing oral sustained-release dosage form design

Dose size:

For orally administered systems, there is an upper limit to the bulk size of the dose to be administered. In general, a single dose of 0.5-1.0g is considered maximal for a conventional dosage form. This also holds for sustained release dosage form. Compounds that require large dosing size can sometimes be given in multiple amounts or formulated into liquid systems. Another consideration is the margin of safety involved in administration of large amount of a drug with a narrow therapeutic range.

Ionization, *pka* and aqueous solubility:

Most drugs are weak acids or bases. Since the unchanged form of a drug preferentially permeates across lipid membranes, it is important to note the relationship between the *pka* of the compound and the absorptive environment. Presenting the drug in an unchanged form is advantageous for drug permeation. Unfortunately, the situation is made more complex by the fact that the drug's aqueous solubility will generally be decreased by conversion to unchanged form. Delivery systems that are dependent on diffusion or dissolution will likewise be dependent on the solubility of the drug in aqueous media.

These dosage forms must function in an environment of changing pH, the stomach being acidic and the small intestine more neutral, the effect of pH on the release process must be defined. Compounds with very low solubility (<0.01mg/ml) are inherently sustained, since their release over the time course of a dosage form in the GI tract will be limited by dissolution of the drug. So it is obvious that the solubility of the compound will be poor choices for slightly soluble drugs, since the driving force for diffusion, which is the drug's concentration in solution, will be low.

Partition Coefficient

When a drug is administered to the GI tract, it must cross a variety of biological membranes to produce a therapeutic effect in another area of the body. It is common to consider that these membranes are lipidic; therefore the partition coefficient of oil-soluble drugs becomes important in determining the effectiveness of membrane barrier penetration.

Compounds which are lipophilic in nature having high partition coefficient are poorly aqueous soluble and it retain in the lipophilic tissue for the longer time. In case of compounds with very low partition coefficient, it is very difficult for them to penetrate

the membrane, resulting in poor bioavailability. Furthermore, partitioning effects apply equally to diffusion through polymer membranes. The choice of diffusion-limiting membranes must largely depend on the partitioning characteristics of the drug.

Stability

Orally administered drugs can be subject to both acid-base hydrolysis and enzymatic degradation. Degradation will proceed at a reduced rate for drugs in solid state; therefore, this is the preferred composition of delivery for problem cases. For the dosage form that are unstable in stomach, systems that prolong delivery over entire course of transit in the GI tract are beneficial; this is also true for systems that delay release until the dosage form reaches the small intestine. Compounds that are unstable in small intestine may demonstrate decreased bioavailability when administered from a sustaining dosage form. This is because more drugs is delivered in the small intestine and, hence, is subject to degradation. Propentheline and Probanthine are representative example of such drug.

1.2.3 Advantages of Sustained Release Drug Delivery Systems:

Sustained release products offer many potential benefits over the conventional dosage formulations. They are;

Sustained blood levels

- For drugs with relatively short half lives, the use of sustained-release products may maintain therapeutic concentrations over prolonged periods.

Dosage frequency reduction

- Minimize or eliminate local side effects.
- Minimize or eliminate systemic side effects.

- Obtain less potentiation or reduction in drug activity with chronic use.
- Minimize drug accumulation with chronic dosing.

Improved patient compliance

- A reduction in the number of daily doses offered by sustained release products has the potential to improve compliance.

Improve efficiency in treatment

- Reduced peak to trough ratio of drug in systemic circulation.
- Improves control of condition i.e., reduced fluctuation in drug level.
- Improves bioavailability of some drugs.
- Reduced rate of rise of drug concentration in blood.
- Sustained & Consistent blood level with in the therapeutic window.
- Customized delivery profiles eg. Sustained release aspirin for morning relief of arthritis by dosing before bedtime.

Economy i.e. reduction in health care costs.

- The average cost of treatment over an extended time period may be less, with less frequency of dosing, enhanced therapeutic benefits and reduced side effects.
- The time required for health care personnel to dispense and administer the drug and monitor patient is also reduced.

Disadvantages

- Sustained release products contain a higher drug load and thus any loss of integrity of the release characteristics of the dosage form has potential problems.

- The larger size of sustained release products may cause difficulties in ingestion or transit through the gut.
- Sustained release products may cause decreased systemic availability in comparison to conventional dosage forms, which may be due to incomplete release, increased first-pass metabolism, increased instability, insufficient residence time for complete release, site specific absorption, pH dependent stability etc.
- Possibility of dose dumping due to food, physiologic or formulation variables or chewing or grinding of oral formulations by the patient and thus, increased risk of toxicity.

1.2 Bilayered Tablets:s

In order to achieve sustained therapeutic action oral SRDDS will release the drug at a slow rate and thus during the initial stages of medication, the plasma drug concentration generally stays below the minimum effective concentration and as a result the patient does not get any therapeutic benefit.

Bilayered SR tablets are a solution to above problem. These preparations provide an immediate dose required for the normal therapeutic response, followed by the gradual release of drug in amounts sufficient to maintain the therapeutic response for a specific period of time. The major advantage of this category is that, in addition to the convenience of reduced frequency administration, it provides levels that are devoid of the peak and valley effect.

They contain two layers formulated with the same drug or two different drugs. The first layer is a fast releasing layer consisting a loading dose of the drug while the second layer is a sustaining layer containing maintenance dose of the drug.

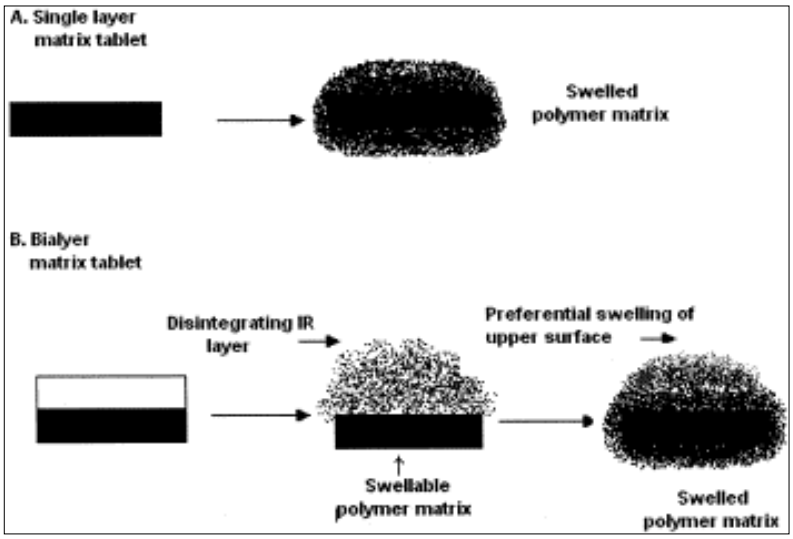


Fig 2:Drug release from Bilayered tablet:

- *Loading dose layer:* provides initial burst release that takes the drug concentration above MEC.
- *Maintenance dose layer:* provides slow sustained release that maintains the drug concentration above the MEC for the remaining period.

2. REVIEW OF LITERATURE

2.1 Past Studies on Bilayered tablets

- **Chitra karthikeyini.S *et.al.,* formulated** Bilayered tablets of Aceclofenac sodium. The Immediate release layer of bilayer tablet of aceclofenac sodium comprised of super disintegrant sodium starch glycolate for fast release layer and Eudragit RL 100 for sustaining layer. Bilayer tablets showed an initial burst effect to provide the loading dose of drug, followed by sustained release for 24 hrs. This modified release bilayer tablets also reduced dosing frequency, increase the bioavailability and provide better patient compliance.

- **Chowdary. K. P. R *et.al.,*** formulated Bilayered tablets of 10mg Glipizide by Wet granulation method with an immediate release layer consisted of Ac-Di-Sol and the sustained release layer consisted of Na CMC or HPMC with 5% ethyl cellulose as polymers. Both the bilayered tablets designed with Na CMC or HPMC with 5% ethyl cellulose gave glipizide release close to the theoretical Sustained Release needed for glipizide i.e; the polymers used sustained the release of the drug for a prolonged period of time.

- **Deelip derle *et.al*.,** has formulated and evaluated mucoadhesive bi-layer buccal tablets of propranolol hydrochloride tablets using the bioadhesive polymers such as sodium alginate and carbopol 971 P along with ethyl cellulose as an impermeable backing layer. The tablets were evaluated for weight variation, thickness, hardness, friability, surface pH, mucoadhesive strength, swelling index, *in vitro* drug release. Tablets containing sodium alginate and carbopol 971 P in the ratio of 5:1 showed the maximum percentage of *in vitro* drug release without disintegration in 12 hours. The swelling index was proportional to sodium alginate content and inversely proportional to carbopol 971 P

content. The surface pH of all tablets was found to be satisfactory, close to neutral pH; hence, no irritation would observe with these tablets. The mechanism of drug release was found to be zero-order kinetics.

- **Doddayya Hiremath *et.al.,*** formulated Bilayered tablets of Losartan Potassium by Direct Compression technique employing HPMC, xanthan gum and gum karaya as a polymers. The immediate release layer was formulated employing Sodium Starch Glycolate as the super disintegrant. Bilayer tablets showed an initial burst to provide the loadingdose of the drug, followed by the sustained release, indicating a promising potential of the losartan potassium bilayer tablet as an alternative to the conventional dosage form.

- **K.Shivanand *et.al.,*** Investigated on mucoadhesive bilayered buccal tablets of Tizanidine Hydrochloride (TZD HCl), using mucoadhesive polymers Carbopol 934(CP), HPMC K4M, HPMC K15M and Sodium carboxymethylcellulose along with ethyl cellulose as an impermeable backing layer. Preformulation studies of TZD HCl like compatibility studies with polymers, using FTIR and DSC were carried out. The bilayered buccal tablets were evaluated for weight variation, thickness, hardness, friability, surface pH, mucoadhesive strength, mucoadhesive time, swelling index, *in vitro* drug release and *ex vivo* permeation. FTIR and DSC found to be compatible with selected polymers. Bilayered buccal tablets containing CP and HPMC K4M in the ratio 1:1 (BT1) had the maximum percentage of *in vitro* drug release in 6 hours. The swelling index of the tablets increased with increasing amounts of CP. The optimized formulation (BT1) follows non-Fickian release mechanism.

- **Nagaraju. R *et.al.,*** formulated Sustained Release Bilayered tablets of Salbutamol and Theohylline by wet granulation method in which Immediate release granules

containing Salbutamol alone and the Sustained release layer consisting of both Salbutamol and Theophylline. The IR layer comprise of HPMCK4M, PVPK30 and starch whereas the SR layer comprise of various polymers, such as hydroxy propyl methylcellulose K4M (HPMC- K4M), hydroxy propyl methylcellulose K100M (HPMC-K100M), xanthan gum, ethyl cellulose and hydroxy propyl methylcellulose phthalate (HPMC-P) were studied.

- **Nakhat *et.al.,*** developed buccoadhesive bilayered tablets of terbutaline sulphate were prepared by direct compression method using bioadhesive polymers like Carbopol 934P, Methocel K4M, Methocel K15M and sodium carboxy methyl cellulose either alone or in combinations with backing layer of ethyl cellulose. The physical characteristics, swelling index, surface pH, *in vitro* bioadhesion strength, and *in vitro* release of formulated tablets were shown to be dependent on characteristics and composition of bioadhesive materials used. The modified in vitro assembly was used to measure and compare the bioadhesive strength of tablets with fresh porcine buccal mucosa as a model tissue. The maximum bioadhesive strength was observed in tablets formulated with Carbopol 934P alone and strength decreases with decrease in its content. All the formulations followed non-Fickian release mechanism. Carbopol 934P and Methocel K4M in the ratio of 1:1 could be used to design effective and stable buccoadhesive tablets of terbutaline sulphate.

- **Patra. C.N *et.al.,*** developed a bilayered tablet of propranolol hydrochloride using superdisintegrant sodium starch glycolate for the fast release layer and water immiscible polymers such as ethylcellulose, Eudragit RLPO and Eudragit RSPO for the sustaining layer. They concluded that the bilayered tablets showed an initial burst effect to provide the loading dose of the drug, followed by sustained release for 12 h, indicating a

promising potential of the propranolol hydrochloride bilayered tablet as an alternative to the conventional dosage form.

- **Prasanthi NL *et.al.,*** formulated Bilayered tablets of Propronolol Hydrochloride by wet granulation method. The Immediate release layer was formulated using sodium starch glycolate as the super disintegrant. The sustained release layer comprised of Xanthan gum, Locust bean gum and Guar gum as the polymers. From all the formulations over 35% of propanolol was released within the first hour of dissolution study. It was observed that bi-layered tablets prepared with xanthan gum were retarding the drug release more when compared with the tablets prepared with locust bean gum, guar gum.
- **Raghavendra Rao. N.G *et.al.,*** formulated Bilayered tablets of 5mg Glipizide by Wet granulation method using 1:2 ratio of Glipizide and Mannitol solid dispersion in the immediate release layer. Sustained release layer was designed by the use of synthetic matrix materials like various grades of HPMC, ethyl cellulose and the natural matrix forming materials like Xanthan gum, Guar gum, Gum Karaya. Finally they concluded that the ideal zero order release pattern was achieved by the blend of HPMC K100M, HPMC K15M and Ethyl cellulose, xanthan gum, guar gum, karaya gum can be used as rate controlling polymers by appropriate selection of the level of polymers in the matrix tablets.
- **Ramesh *et.al.,*** formulated Bilayered tablets of Metformin hydrochloride and Pioglitazone. Immediate release Pioglitazone layer was formulated by Dry granulation and Sustained release Metformin Hydrochloride was formulated by Wet granulation. Immediate release layer comprised of Cross caramellose sodium as super disintegrant and the Sustained release layer comprised of HPMCK4M, HPMC 15cps, and Na CMC as

polymers. Bilayered tablet showed an immediate release effect to provide the loading dose of the drug, followed by sustained release for eight hrs.

- **Sonia Pandey *et.al.,*** has developed buccoadhesive bilayered tablets of carvedilol were prepared by direct compression method using bioadhesive polymers like carbopol 934P and HPMC K4M.The formulations were tested for in vitro drug release, in vitro bioadhesion, moisture absorption and in vitro drug permeation through porcine buccal mucosa. The dissolution of Carvedilol from all the prepared tablets into phosphate buffer (pH 6.8) was controlled and followed by non-fickian release mechanism. Dissolution studies of the tablets of optimized batch containing 5% Carbopol 934P/65% HPMC K4M/30% lactose showed 82.7 % release of drug in 6 h. The mucoadhesive strength and residence time of the optimized batch are 17.93 g and 9.45 h respectively. The swelling index and microenvironment pH of the optimized batch after 6 h are 77.54 and 6.76 respectively. Procured sample of carvedilol was tested for its identification by taking FTIR of pure drug. Drug excipient compatibility was done at 30°C, 65% ± 5%RH and 40°C 75% ± 5% RH using open and closed vial for four weeks and observed for physical changes. Result does not show any physical changes to mixture after 4 weeks. The results indicate that suitable bioadhesive bilayered buccal tablets with desired permeability using 5-6% Carbopol 934P, 65-68%HPMC K4M and 30% Lactose.

- **Swamy P V *et.al.,*** has formulated and evaluated buccoadhesive Bilayer tablets of granisetron hydrochloride. Bilayer buccal tablets containing the drug were prepared by direct compression method using combination of polymers (such as hydroxypropyl methylcellulose 15 cps, sodium carboxymethyl cellulose and Carbopol 934p.) and ethyl cellulose as backing layer. The designed tablets were evaluated for various physical and biological parameters, drug content uniformity, *in-vitro* drug release, short-term stability,

drug- excipients interactions (FTIR).The formulation HF1 with the drug matrix layer composition- hydroxypropyl methylcellulose 15 cps (47% w/w), Carbopol 934p (3%w/w), and mannitol was found to be promising. This optimized formulation exhibited an *in vitro* drug release of 94% in 8 h along with satisfactory bioadhesion strength (4.3 gm). Short-term stability studies on the promising formulation indicated that there are no significant changes in drug content and *in vitro* dissolution characteristics ($p<0.05$). IR spectroscopic studies indicated that there are no drug-excipient interactions. The present study proves that buccoadhesive bilayer tablets of granisetron hydrochloride with controlled drug release properties can be successfully prepared by direct compression method using HPMC 15 cps and Carbopol 934p as mucoadhesive polymers and ethyl cellulose as backing layer.

- **Vijaya Kumar. B *et.al.,*** formulated Sustained release Bilayered tablets of Guaifenesin by wet granulation. The immediate release granules consists of Sodium starch glycolate as the super disintegrant and concluded that 20% of drug was released within first 30 minutes. The sustain release granules were formulated using Metalose 90 SH and Carbopol as polymers inorder to sustain the release. The bilayered tablets showed biphasic release, in the first phase the first fraction of the dose (immediate dose) was released as a burst effect because of fast releasing components of loading layer then second phase was released from matrix layer as a controlled non fickian diffusion release fashion.

- **Vinoth Kumar.G *et.al* ., formulated** Bilayered tablets of Cefixime trihydrate and Dicloxacillin sodium to avoid incompatibility. Bilayered tablets were formulated by wet granulation method in which one layer contains sustained release drug Dicloxacillin sodium and another layer contains immediate release drug Cefixime tri hydrate.

Dicloxacillin sodium layer was formulated using Povidone, HPMCK4M, HPMCK15M for sustained release and the immediate release layer of Cefixime trihydrate was formulated with Cross caramellose sodium.

- **Vishnu M. Patel *et.al.*,** has developed and characterized mucoadhesive bilayer buccal tablets of propranolol hydrochloride using the bioadhesive polymers sodium alginate (Na-alginate) and Carbopol 934P (CP) along with ethyl cellulose as an impermeable backing layer. The tablets were evaluated for weight variation, thickness, hardness, friability, surface pH, mucoadhesive strength, swelling index, in vitro drug release, ex vivo drug permeation, ex vivo mucoadhesion, and in vivo pharmacodynamics in rabbits. Tablets containing Na-alginate and CP in the ratio of 5:1 (F2) had the maximum percentage of in vitro drug release without disintegration in 12 hours. The swelling index was proportional to Na-alginate content and inversely proportional to CP content. The surface pH of all tablets was found to be satisfactory (7.0 ± 1.5), close to neutral pH; hence, buccal cavity irritation should not occur with these tablets. The mechanism of drug release was found to be non-Fickian diffusion and followed zero-order kinetics. The formulation F4 was optimized based on good bioadhesive strength (28.9 ± 0.99 g) and sustained in vitro drug permeation (68.65% ± 3.69% for 12 hours). The behavior of formulation F4 was examined in human saliva, and both the drug and the buccal tablet were found to be stable. The formulation F4 was applied to rabbit oral mucosa for in vivo studies. The formulation inhibited isoprenaline-induced tachycardia. The studies conducted in rabbits confirmed the sustained release as compared with intravenous administration.

2.2 Past Studies on Pioglitazone HCl:

Hallakou.S *et.al.,* aimed at improving insulin sensitivity would promote the recruitment of new adipocytes in vivo. To address this problem, they have studied the

in vivo effect of pioglitazone on glucose metabolism and gene expression in the adipose tissue of an animal model of obesity with insulin resistance, the obese Zucker (fa/fa) rat. Pioglitazone markedly improves insulin action in the obese Zucker (fa/fa) rat, but doubles its weight gain after 4 weeks of treatment.

- **Hofmann *et.al.,*** *studied* the Glucose Transport Deficiency in Diabetic Animals by Treatment with the Oral Antihyperglycemic Agent Pioglitazone HCl.
- **Kemnitz J W *et.al.,*** The antidiabetic effects of pioglitazone hydrochloride were evaluated in 6 spontaneously obese, insulin-resistant rhesus monkeys. The animals were studied during six successive 2-wk treatment phases separated by 2-wk rest periods.
- **Mehmood A. Khan, MD**, ***et.al.,*** Studied the metabolic effects of pioglitazone or rosiglitazone in patients with type 2 diabetes who were previously treated with troglitazone.
- **Mozhgan Dorkhan, Anders Frid,** studied the mechanism of action and clinical data behind the use of Pioglitazone HCl in management of type 2 diabetes in terms of glycemic and non-glycemic effects, tolerability and side effects, impact on vascular health.

2.3 Past Studies on Metformin HCl:

- **Kah Hay Yuen, Kok Khiang Peh and Boon Li Tan** studied bioavailability of two controlled-release metformin preparations (Diabetmin Retard and Glucophage Retard) and also to correlate the in vitro and in vivo data obtained with the two preparations.
- **Meltem Cetin, Alptug Atila, Selma Sahin, Imran Vural** developed the metformin HCl-loaded nanoparticle formulations. Nanoparticles were prepared by the nanoprecipitation method using both a single polymer (Eudragit®RSPO) and a polymer mixture (Eudragit/PLGA). The mean particle size ranged from 268.8 to 288 nm and the

nanoparticle surface was positively charged (9.72 to 10.1 mV). The highest encapsulation efficiency was observed when Eudragit®RSPO was used. All formulations showed highly reproducible drug release profiles and the *in vitro* drug release in phosphate buffer (pH = 6.8) ranged from 92 to 100% in 12 h.

- **Patil SA, Kuchekar BS, Chabukswar AR, and Jagdale SC** studied the formulation and characterization of solid dispersion (SD) of metformin hydrochloride using methocel K100M as the carrier by the solvent evaporation and cogrinding method. The influence of drug polymer ratio on drug release was studied by dissolution tests.
- **Subal Chandra Basak,Kesevan Senthil Kumar, Murugesan Ramalingam** formulated Metformin hydrochloride as a hydrophobic matrix sustained release tablet employing wax materials and the sustained release behavior of the fabricated tablet was investigated. Sustained release matrix tablets containing 500 mg metformin HCl were developed using different bees wax combinations. The tablets were prepared by wet granulation technique. The formulation was optimized on the basis of acceptable tablet properties and *in vitro* drug release.

2.4 Past Studies on Bilayered tablets of Pioglitazone HCl and Metformin HCl:

- **Aziz Karim *et.al.,*** studied the Bioavailability of pioglitazone and metformin, in 2 dose strengths, given either as a fixed-dose combination tablet or as coadministration of commercial tablets (coad), was studied in young healthy subjects in 2 separate studies. In study I (n = 63), single oral doses of 15-mg pioglitazone/500-mg metformin fixed-dose combination tablets or equivalent doses of commercial tablets were administered, in a fasting state, in an open-label, randomized, crossover study with a 7-day washout period between treatments. Study II (n = 61) was similar in design to study I, except the 15/850-mg fixed-dose combination tablet and coad treatments were evaluated.

3. AIM AND OBJECTIVES, PLAN OF WORK

The aim of this investigation is to Design and Develop Bilayered oral sustained matrix tablets of Pioglitazone hydrochloride and Metformin hydrochloride.

The concept of Bilayered tablet technology is utilized for stabilization of two incompatible drugs, taste masking of drugs, delivering two drugs having synergistic effects or to deliver a drug for biphasic drug release profile and for the purpose of extension of patents. A Bilayered tablet comprises of two layers among which the first layer is immediate release layer for sudden onset of action and the second layer is Sustained release layer to maintain the steady state concentrations of drug in the blood.

Pioglitazone HCl is thiazolidinedione (TZD) class of drug with hypoglycemic, antihyperglycemic and antidiabetic action. Chemically Pioglitazone is (RS)-5-(4-[2-(5-ethylpyridin-2-yl) ethoxy] benzyl) thiazolidine-2, 4-dione. Pioglitazone is used for the treatment of diabetes mellitus type 2 (previously known as non-insulin-dependent diabetes mellitus, NIDDM) in monotherapy and in combination with a sulfonylurea, Metformin. Pioglitazone has also been used to treat non-alcoholic fatty liver. Pioglitazone has also been found to reduce the risk of conversion from prediabetes to diabetes mellitus type 2 by 72%. It has short biological half life of 3-5 hrs (Ramesh et al).

Metformin HCl is a biguanide oral anti hyperglycemic (anti diabetic) agent. It is used as an adjunct to diet and exercise for the management of type 2(non-insulin-dependent diabetes mellitus) diabetes mellitus in patients whose hyperglycemia cannot be controlled by diet alone. As Metformin HCl possess short biological half life (1.5-4.5 hrs), patient should go for frequent administration usually twice or thrice a day which

might be a risk to the patient. In order to overcome this Metformin HCl sustained release dosage forms are formulated (Ramesh et al).

PLAN OF WORK:

1. To formulate and evaluate the Bilayered tablets of Pioglitazone HCl and Metfofmin HCl.
2. To carry out the drug - excepient compatibility studies by IR spectral analysis.
3. To carry out the Precompressional parameters for the powder blend of IR layer of Bilayered tablets.
4. To carry out the Precompressional and Postcompressional parameters for Bilayered tablets.
5. To study the release kinetics and transport mechanism of drug from the formulations.
6. To study the comparative release profiles of tablets formulated with marketed formulation using the similarity factor.

4. DRUG & EXIPIENTS PROFILE

4.1 PIOGLITAZONE HCl

Pioglitazone HCl is thiazolidinedione (TZD) class of drug with hypoglycemic, antihyperglycemic and antidiabetic action. Pioglitazone is used for the treatment of diabetes mellitus type 2 (previously known as non-insulin-dependent diabetes mellitus, NIDDM) in monotherapy and in combination with a sulfonylurea, Metformin. Pioglitazone has also been used to treat non-alcoholic fatty liver. Pioglitazone has also been found to reduce the risk of conversion from prediabetes to diabetes mellitus type 2 by 72%.

Structure:

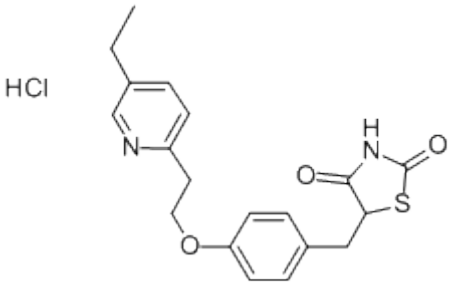


Figure 3: Structure of Pioglitazone HCl

Molecular Formula: $C_{19}H_{20}N_2O_3S$

Molecular Weight : 356.439

Chemical Name : (RS)-5-(4-[2-(5-ethylpyridin-2-yl) ethoxy] benzyl) thiazolidine-2, 4-dione.

Description : whitish odourless powder

Half-life : 3-5 hrs

Melting Point : 183-184^{o}C

pKa : 12.06

Mechanism of Action:

Pioglitazone acts as an agonist at peroxisome proliferator activated receptors (PPAR) in target tissues for insulin action such as adipose tissue, skeletal muscle, and liver. Activation of PPAR-gamma receptors increases the transcription of insulin-responsive genes involved in the control of glucose production, transport, and utilization. In this way, pioglitazone both enhances tissue sensitivity to insulin and reduces hepatic gluconeogenesis. Thus, insulin resistance associated with type 2 diabetes mellitus is improved without an increase in insulin secretion by pancreatic β cells.

Pharmacodynamics:

Pioglitazone, a member of the drug group known as the thiazolidinediones or "insulin sensitizers", is not chemically or functionally related to the alpha-glucosidase inhibitors, the biguanides, or the sulfonylureas. Pioglitazone targets insulin resistance and, hence, is used alone or in combination with insulin, metformin, or a sulfonylurea as an antidiabetic agent.

Pharmacokinetics:

Absorption: Following oral administration, in the fasting state, pioglitazone is first measurable in serum within 30 minutes, with peak concentrations observed within 2 hours. Food slightly delays the time to peak serum concentration to 3 to 4 hours, but does not alter the extent of absorption.

Distribution: The apparent volume of distribution at steady state was 0.63±0.41 liters/kilogram.

Metabolism: Metabolism Site is liver extensively. First pass metabolism is minimal.

Elimination: Following oral administration, approximately 15% to 30% of the pioglitazone dose is recovered in the urine. Renal elimination of pioglitazone is negligible, and the drug is excreted primarily as metabolites and their conjugates. It is presumed that most of the oral dose is excreted into the bile either unchanged or as metabolites and eliminated in the feces.

Contraindications: Take without regard to meals. Food slightly delays absorption rate but extent of absorption is not affected.

Dosage: initially 15 or 30 mg per day, maximum 45mg per day.

4.2 METFORMIN HCl

Metformin is a biguanide antihyperglycemic agent used for treating non-insulin-dependent diabetes mellitus (NIDDM). It improves glycemic control by decreasing hepatic glucose production, decreasing glucose absorption and increasing insulin-mediated glucose uptake. Metformin is the only oral antihyperglycemic agent that is not associated with weight gain. Metformin may induce weight loss and is the drug of choice for obese NIDDM patients. When used alone, metformin does not cause hypoglycemia; however, it may potentiate the hypoglycemic effects of sulfonylurea's and insulin. Its main side effects are dyspepsia, nausea and diarrhea. Dose titration and/or use of smaller divided doses may decrease side effects. Metformin should be avoided in those with severely compromised renal function (creatinine clearance < 30 ml/min), acute/decompensated heart failure, severe liver disease and for 48 hours after the use of iodinated contrast dyes due to the risk of lactic acidosis. Lower doses should be used in the elderly and those with decreased renal function. Metformin decreases fasting plasma glucose, postprandial blood glucose and glycosolated hemoglobin (HbA1c) levels, which

are reflective of the last 8-10 weeks of glucose control. Metformin may also have a positive effect on lipid levels.

Structure:

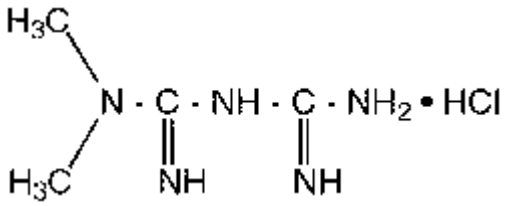


Figure 4: Structure of Metformin Hcl

Molecular Formula: $C_4H_{11}N_5$

Molecular Weight : 129.164

Chemical Name : 1-carbamimidamido-N, N-dimethylmethanimidamide.

Description : whitish odourless powder

Half-life : 1.5-4.5 hrs

Melting Point : 223-226^{o}C

pKa : 12.4

Mechanism of Action:

Metformin's mechanisms of action differ from other classes of oral antihyperglycemic agents. Metformin decreases blood glucose levels by decreasing hepatic glucose production, decreasing intestinal absorption of glucose, and improving insulin sensitivity by increasing peripheral glucose uptake and utilization. These effects are mediated by the initial activation by metformin of AMP-activated protein kinase (AMPK), a liver enzyme that plays an important role in insulin signaling, whole body energy balance, and the metabolism of glucose and fats. Activation of AMPK is required

for metformin's inhibitory effect on the production of glucose by liver cells. Increased peripheral utilization of glucose may be due to improved insulin binding to insulin receptors. Metformin administration also increases AMPK activity in skeletal muscle. AMPK is known to cause GLUT4 deployment to the plasma membrane, resulting in insulin-independent glucose uptake. The rare side effect, lactic acidosis, is thought to be caused by decreased liver uptake of serum lactate, one of the substrates of gluconeogenesis. In those with healthy renal function, the slight excess is simply cleared. However, those with severe renal impairment may accumulate clinically significant serum lactic acid levels. Other conditions that may precipitate lactic acidosis include severe hepatic disease and acute/decompensated heart failure.

Pharmacodynamics:

Metformin is an oral antihyperglycemic agent that improves glucose tolerance in patients with NIDDM, lowering both basal and postprandial plasma glucose. Metformin is not chemically or pharmacologically related to any other class of oral antihyperglycemic agents. Unlike sulfonylureas, metformin does not produce hypoglycemia in either patients with NIDDM or healthy subjects and does not cause hyperinsulinemia. Metformin does not affect insulin secretion.

Pharmacokinetics:

Absorption: Absorbed over 6 hours, bioavailability is 50 to 60% under fasting conditions. Administration with food decreases and delays absorption. Some evidence indicates that the level of absorption is not dose-related, suggesting that absorption occurs through a saturable process. Limited data from animal and human cell cultures indicate that absorption occurs through a passive, non-saturable process, possibly involving a paracellular route. Peak action occurs 3 hours after oral administration.

Distribution: The volume of distribution is 63-276 liters/kilogram, likely due to less binding in the GI tract and/or different methods used to determine volume of distribution.

Metabolism: Metformin is not metabolized.

Elimination: Intravenous single-dose studies in normal subjects demonstrate that metformin is excreted unchanged in the urine and does not undergo hepatic metabolism (no metabolites have been identified in humans) nor biliary excretion. Approximately 90% of the drug is eliminated in 24 hours in those with healthy renal function. Renal clearance of metformin is approximately 3.5 times that of creatinine clearance, indicating the tubular secretion is the primary mode of metformin elimination.

Contraindications: Avoid alcohol. Take with food to reduce gastric irritation.

Dosage: 500 or 750 mg per day

4.3 Poly Ethylene Oxide (PEO-303)

Synonyms: POLYOX.

Chemical Structure: $(-O-CH_2-CH_2-)_n OH$. They have same chemical structure as PEG but higher molecular weights. n = average number of oxyethylene groups

Category: Binding agent, Emollient, Cross linking agent, Film forming excipient.

Description: Polyethylene oxide is a biocompatible polymer, which is marketed as "POLYOX". It is a non-ionic, water soluble resin, with good lubricating, binding and film forming properties. POLYOX retards the release rate of drug/s and hence are widely used in pharmaceutical formulations like controlled release dosage forms, hot-melt technology and mucoadhesive dosage forms.

Solubility: POLYOX water-soluble resins are readily soluble in and will thicken wide variety of organic solvents at various temperatures. Organic solvents include most halogenated hydrocarbons, various ketones, alcohols, aromatic hydrocarbons and esters. POLYOX water-soluble resins are not generally soluble in aliphatic hydrocarbon solvents, glycols, diols and aliphatic ethers.

Stability: The chemical stability of PEO was found to be dependent on both the storage and processing temperature, and the molecular weight of the polymer. Storage of the polymer above its melting point significantly increased polymer degradation, and the degradation process was accelerated as the molecular weight was reduced. Vitamin E, Vitamin E Succinate and Vitamin E TPGS were found to be suitable stabilizers for PEO; however, ascorbic acid was shown to degrade the polymer in solution.

Applications:

- Wide range of molecular weights offers formulation flexibility.
- Versatile application in direct compression and granulation.
- Rapid hydration and swelling for use in osmotic pump technologies.
- Fast hydration and gel formation for use in hydrophilic matrices.
- Meets requirements of the United States Pharmacopoeia (USP) and compliance with US Food Chemicals Codex (Moroni A., Ghebresellassie I., 1995).

4.4 Carbopol 971P

Synonyms: Acrylic acid, 2-propenoic acid, Acroleic acid, Propenoic acid, Carbomer, Poly(acrylic acid).

Chemical Name: Prop-2-enoic acid

Molecular Formula: $C_3H_4O_2$

Molecular Weight: 72.06266 g/mol.

Category: Binding agent, Film forming excipient.

Description: Carbopol polymers are polymers of acrylic acid cross-linked with polyalkenyl ethers or divinyl glycol. They are produced from primary polymer particles of about 0.2 to 6 micron average diameter. The flocculated agglomerates cannot be broken down into the ultimate particle when produced. Each primary particle can be viewed as a network structure of polymer chains interconnected by cross-links. Without the cross-links, the primary particle would be a collection of linear polymer chains intertwined but not chemically bonded. They swell in water up to 1000 times their original volume (and 10 times their original diameter) to form a gel when exposed to a pH environment above 4.0 to 6.0. Because the pKa of these polymers is 6.0 to 0.5, the carboxylate groups on the polymer backbone ionize, resulting in repulsion between the negative charges, which adds to the swelling of the polymer.

Glass transition temperature: The glass transition temperature of Carbopol polymers is 105°C (221°F) in powder form. However, the glass transition temperature decreases significantly as the polymer comes into contact with water.

Applications:

- Controlled release in tablets. Carbopol polymers offer consistent performance over a wide range of desired parameters (from pH-derived semi-enteric release to near zero-order drug dissolution kinetics) at lower concentrations than competitive systems.

- Bioadhesion in buccal, ophthalmic, intestinal, nasal, vaginal, and rectal applications. Noveon AA-1 USP polycarbophil is the recognized industry standard for bioadhesion.
- Thickening at very low concentrations (less than 1%) to produce a wide range of viscosities and flow properties in topical lotions, creams and gels, oral suspensions, and in transdermal gel reservoirs.
- Permanent suspensions of insoluble ingredients in oral suspensions and topicals.
- Emulsifying topical oil-in-water systems permanently, even at elevated temperatures, with essentially no need for irritating surfactants (Perez-Marcos B et al., 1991).

4.5 Microcrystalline Cellulose

Synonyms: Avicel PH; Celex; cellulose gel; Celphere; Ceolus KG; crystalline cellulose; E460; Emcocel; Ethispheres; Fibrocel; Pharmacel; Tabulose; Vivapur.

Chemical name: Cellulose

Structure:

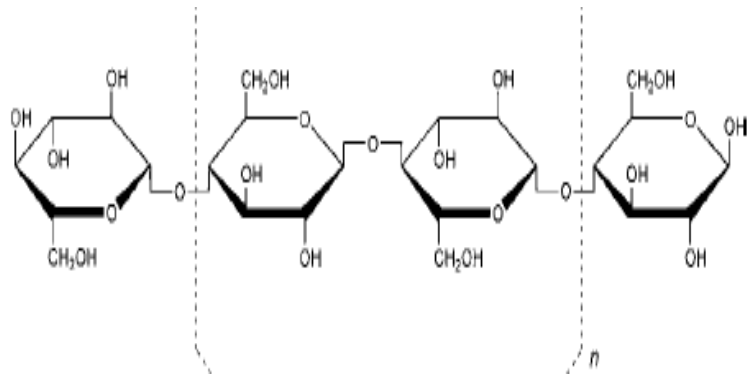


Figure 5: Structure of Microcrystalline Cellulose

Functional category: Adsorbent; suspending agent; tablet and capsule diluent; tablet disintegrant.

Description: Microcrystalline cellulose is a purified, partially depolymerized cellulose that occurs as a white, odourless, tasteless, crystalline powder composed of porous particles.

Applications: Microcrystalline cellulose is widely used in pharmaceuticals, primarily as a binder/diluent in oral tablet and capsule formulations where it is used in both wet-granulation and direct-compression processes. In addition to its use as a binder/diluent, microcrystalline cellulose also has some lubricant and disintegrant properties.

Stability and Storage Conditions: Microcrystalline cellulose is a stable though hygroscopic material. The bulk material should be stored in a well-closed container in a cool, dry place.

4.6 Spray Dried Lactose

Synonyms: FlowLac 100; Lactopress Spray-Dried; NF Lactose–316 Fast Flo; NF Lactose–315; Pharmatose DCL 11; Pharmatose DCL 14; Super-Tab Spray-Dried.

Chemical Name: Spray-dried lactose is a mixture of amorphous lactose, which is a 1:1 mixture of a-and-b-lactose, and O-b-D-galactopyranosyl-(1→4)-a-D-glucopyranose monohydrate.

Chemical Structure:

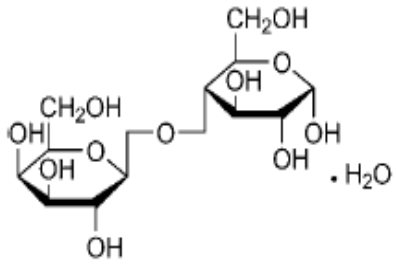


Figure 6: Structure of Spray Dried Lactose

Functional Category: Binding agent; directly compressible tablet excipient; tablet and capsule diluent; tablet and capsule filler.

Applications: Spray-dried lactose is widely used as a binder, filler-binder, and flow aid in direct compression tableting.

Description: Lactose occurs as white to off-white crystalline particles or powder. It is odourless and slightly sweet-tasting.

Storage: Spray-dried lactose should be stored in a well-closed container in a cool, dry place.

4.7 Cros Povidone

Synonyms: E1201; Kollidon; Plasdone; poly[1-(2-oxo-1-pyrrolidinyl)ethylene]; polyvidone; polyvinylpyrrolidone; PVP; 1-vinyl-2-pyrrolidinone polymer.

Chemical Name: 1-Ethenyl-2-pyrrolidinone homopolymer

Chemical Structure:

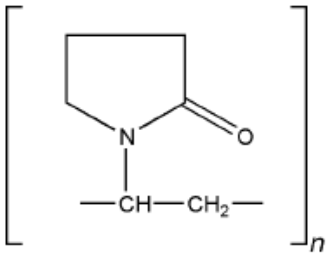


Figure 7: Structure of Cros Povidone

Molecular Formula: $(C_6H_9NO)_n$

Molecular Weight: 2.500 - 2.5000.000 g/mol.

Category: Disintegrant; dissolution aid; suspending agent; tablet binder.

Description: Povidone occurs as a fine, white to creamy-white colored, odourless or almost odourless, hygroscopic powder.

Solubility: Freely soluble in acids, chloroform, ethanol (95%), ketones, methanol, and water; practically insoluble in ether, hydrocarbons, and mineral oil.

Melting Point: Softens at 150℃.

Stability: Povidone darkens to some extent on heating at 150℃, with a reduction in aqueous solubility. It is stable to a short cycle of heat exposure around 110–130℃.

Storage: Povidone may be stored under ordinary conditions without undergoing decomposition or degradation. However, since the powder is hygroscopic, it should be stored in an airtight container in a cool, dry place.

4.8 Aerosil

Synonyms: Fumed silica, silicon dioxide, cristobalite.

Chemical Name: dioxosilane

Chemical Structure:

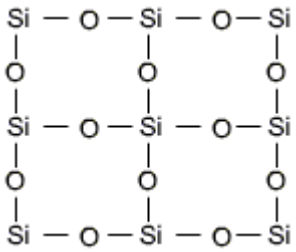


Figure 8: Structure of Aerosil

Molecular Formula: 60.0843 g/mol.

Molecular Weight: SiO_2

Category: It serves as a universal thickening agent, in milkshakes for example, and an anticaking agent (free-flow agent) in powders.

Description: Fumed silica is made from flame pyrolysis of silicon tetrachloride or from quartz sand vaporized in a 3000°C electric arc. Like silica gel, it serves as a desiccant. It is used in cosmetics for its light-diffusing properties. It is used as a light abrasive, in products like toothpaste. Other uses include filler in silicone elastomer and viscosity adjustment in paints, coatings, printing inks, adhesives and unsaturated polyester resins. Also used in the production of Kitty Litter.

Melting Point: Stable up to 1610^{o}C (2930^{o}F)

Solubility: Fumed silica will form dispersions in water, glycerine, butyl alcohol, mineral oil and a variety of other liquids, causing them to thicken or form gels. The dispersions often have thixotropic properties, i.e., viscosity that varies with rate of stirring.

Stability/Storage: These products are stable indefinitely at room temperature if kept dry. Their tendency to adsorb moisture suggests an effective shelf life of about two years, once opened.

4.9 Magnesium Stearate

Synonyms: Magnesium octadecanoate; octadecanoic acid, magnesium salt; stearic acid, magnesium salt.

Chemical Name: Octadecanoic acid magnesium salt.

Category: Tablet and capsule lubricant.

Description: Magnesium stearate is a very fine, light white, precipitated or milled, impalpable powder of low bulk density, having a faint odour of stearic acid and a characteristic taste. The powder is greasy to the touch and readily adheres to the skin.

Solubility: Practically insoluble in ethanol, ethanol (95%), ether and water; slightly soluble in warm benzene and warm ethanol (95%).

Stability & Storage: Magnesium stearate is stable and should be stored in a well closed container in a cool, dry place.

4.10 Talc

Synonyms: Altalc; E553b; hydrous magnesium calcium silicate; hydrous magnesium silicate; Luzenac Pharma; magnesium hydrogen metasilicate; Magsil Osmanthus; Magsil Star; powdered talc; purified French chalk; Purtalc; soapstone; steatite; Superiore.

Chemical Name: Talc

Category: Anticaking agent; glidant; tablet and capsule diluent; tablet and capsule lubricant.

Description: Talc is a very fine, white to grayish-white, odourless, impalpable, unctuous, crystalline powder. It adheres readily to the skin and is soft to the touch and free from grittiness.

Solubility: Practically insoluble in dilute acids and alkalis, organic solvents, and water.

Stability: Talc is a stable material and may be sterilized by heating at 160℃ for not less than 1 hour. It may also be sterilized by exposure to ethylene oxide or gamma irradiation.

Storage: Talc should be stored in a well-closed container in a cool, dry place.

5. MATERIALS AND METHODS

5.1. Materials Used: Table 1: Materials used in Formulations

S.No	Ingredients	Supplier
1	Pioglitazone HCl	Darwin pvt ltd. Vijayawada.
2	Metformin HCl	Darwin pvt ltd. Vijayawada.
3	Polyethyleneoxide (PEO-303)	Colorcon Asia pvt. Ltd
4	Carbopol 971P	HiMedia Pharmaceuticals Pvt Ltd, Mumbai
5	Microcrystalline Cellulose	SD Fine chemicals, Mumbai
6	Spray Dried Lactose	SD Fine chemicals, Mumbai
7	Crospovidone	SD Fine chemicals, Mumbai
8	Aerosil	SD Fine chemicals, Mumbai
9	Magnesium Stearate	SD Fine chemicals
10	Talc	SD Fine chemicals

5.2 Instruments Used: Table 2: Instruments used in Formulations

S.NO	Name of the Instrument	Model And Manufacturer
1	Digital balance	Essae –teraoku Ltd
2	Tablet dissolution test apparatus,USP	Labindia, Disso 2000
3	UV-Visible spectrophotometer	Elico Ltd., SL 150, Hyderabad
4	Compression machine	Cadmach Machinery , Kolkata
5	Roche Friabilator	Campbell Electronics
6	Monsanto Hardness Tester	Shreeji Chemicals , Mumbai
7	Disintegration apparatus	Thermonic Campbell electronics, Mumbai
8	pH meter	Elico Li 120

5.3. Methodology:

UV Spectrophotometric method was used in the present study for the estimation of Pioglitazone HCl.

5.4. Confirmation of Pioglitazone HCl through UV spectral analysis:

The λmax of Pioglitazone HCl was observed by scanning the standard drug solution. Accurately weighed amount of Pioglitazone HCl (25mg) was dissolved in methanol to form a clear solution and it was made up to volume in 25ml volumetric flask with methanol. 1 ml of this solution was transferred in to 100 ml of volumetric flask and it was made up to volume with pH 1.2 buffer. The resulting solution (10 µg/ml of Pioglitazone HCl) was scanned over the range 200-400 nm against pH 1.2 buffer as blank using UV spectrophotometer. The wavelength at which maximum absorbance detected was recorded as the λmax. The absorption spectrum was noted at 269nm.

5.5 Construction of Calibration Curve:

- ***Preparation of pH 1.2 (0.1N HCl) Buffer***: The 8.5ml of hydrochloric acid (0.1N) is added 1000ml of distilled water.
- ***Preparation of Stock solution***: 25mg of Pioglitazone HCl was dissolved in methanol in 25ml volumetric flask to form a clear solution and later it was made up to volume with methanol.
- ***Preparation of Required concentrations***: 1ml of stock solution was withdrawn and made up to volume with pH 1.2 buffer in 10ml volumetric flask to obtain 100µg/ml concentration. 0.5ml, 1.0ml, 1.5ml, 2.0ml, and 2.5ml of solution were withdrawn from the 100µg/ml concentration and made up to volume with pH 1.2

buffer in 10ml volumetric flak to obtain 5µg/ml, 10µg/ml, 15µg/ml, 20µg/ml and 25µg/ml respectively, then analyzed Spectrophotometrically at 269nm

5.6. Methods of Estimation of Metformin HCl:

UV Spectrophotometric method was used in the present study for the estimation of Metformin HCl.

5.7. Confirmation of Metformin HCl through UV spectral analysis:

The λmax of Metformin HCl was observed by scanning the standard drug solution. Accurately weighed amount of Metformin HCl (25mg) was dissolved in distilled water to form a clear solution and it was made up to volume in 25ml volumetric flask with distilled water.1 ml of this solution was transferred in to 100 ml of volumetric flask and it was made up to volume with pH 1.2 buffer, PH 6.8 phosphate buffer and Distilled water. The resulting solution (10 µg/ml of Metformin HCl) was scanned over the range 200-400 nm against pH 1.2 buffer and pH 6.8 phosphate buffer and Distilled water as blank using UV spectrophotometer. The wavelength at which maximum absorbance detected was recorded as the λmax. The absorption spectrum was noted at 232nm.

5.8 Construction of Calibration Curve:

- ***Preparation of pH 1.2 (0.1N Hal) Buffer***: About 8.5ml of hydrochloric acid (0.1N) is added 1000ml of distilled water.
- ***Preparation of pH 6.8 Phosphate Buffer***: About 6.8 grams of Potassium dehydrogenate phosphate and suitable amount of Sodium hydroxide were dissolved in water until a clear solution was formed. Later it was made up to volume with water in 1000ml volumetric flask.
- ***Preparation of Stock solution***: 25mg of Metformin HCl was dissolved separately in Distilled water, pH 1.2 buffer and pH 6.8 phosphate buffer in 25ml volumetric

flask to form a clear solution and later it was made up to volume with respective buffers.

- ***Preparation of Required concentrations***: 1ml of stock solution was withdrawn and made up to volume with respective buffers in 10ml volumetric flask to obtain 100µg/ml concentration. 0.2ml, 0.4ml, 0.6ml, 0.8ml, and 1ml of solution were withdrawn from the 100µg/ml concentration and made up to volume with respective buffers in 10ml volumetric flak to obtain 2µg/ml, 4µg/ml, 6µg/ml, 8µg/ml and 10µg/ml respectively, then analyzed Spectrophotometrically at 232nm.

5.9 Calculation of the Theoretical Release Profile of Metformin from Bilayer Tablets:

The total dose of Metformin for a once-daily SR formulation was calculated by the following equation.

$$Dt = Dose\ (1+ (0.693 \times t)/ t_{1/2})$$

Where,

Dt = total dose of drug,

Dose = dose of the IR part,

t = time (hr) during which the SR is desired (12 hr),

$t_{1/2}$ = half-life of the drug (3 hr).

$Dt = 132.5\ (1+ (0.693 \times 12)/ 3) = 500$

Hence, the formulation should release 132.5 mg in first hour like conventional tablets and 33.4 mg per hour up to 12 hours thereafter.

5.10 Preparation of Bilayer Tablets:

In this present investigation Bilayered tablets of Pioglitazone HCl and Metformin HCl were formulated by Direct Compression Technique and Wet Granulation Technique.

5.10.1 Formulation of Immediate Release layer

Immediate release layer of Pioglitazone HCl was prepared by dry granulation technique. Pioglitazone, cross Povidone and Micro crystalline cellulose (MCC) were passed through sieve no # 40. The Sunset yellow lake was passed through sieve no #200. All the above were mixed in geometric proportion in a poly bag for 15 minutes. Aerosil and magnesium stearate were passed through sieve no # 60. Sifting was performed and the lubricated material was passed through the poly bag and mixed for 2 minutes. Compositions of different trial formulations for the IR layer were given in table 6.5.1. The final weight of the IR layer was fixed to 150 mg.

5.10.2 Formulation of Sustained Release layer

5.10.2. 1 Direct Compression Technique:

Sustained release layer of Metformin HCl was prepared by dry granulation technique. Metformin HCl, Poly ethylene oxide (PEO-303) and MCC were passed through sieve no # 40. All the above were mixed in geometric proportion in a poly bag for 15 minutes. Talc and magnesium stearate were passed through sieve no # 60. Sifting was performed and the lubricated material was passed through the poly bag and mixed for 2 minutes. Compositions of different trial formulations for the IR layer were given in table 6.5.2. The final weight of the SR layer was fixed to 800 mg.

5.10.2. 2 Wet Granulation Technique:

Granules of Sustained release layer was formulated by uniformly mixing required amount of Metformin HCl with measured quantities of polymer and diluent as specified in the formulation table 6.5.3 using 1:1 ratio of ethanol and water as diluting fluid. Now the wet damp mass was passed through sieve no #20 and the granules were dried in hot air oven at 50^{o}C. Talc and magnesium stearate were added and mixed thoroughly before compression of granules. The final weight of the SR layer was fixed to 800 mg.

5.10.3 Compression of Bilayer Tablets:

The extended release blend of Metformin HCl (800 mg) was compressed lightly using single punch tablet machine (Cad mach machinery Co Pvt.Ltd, India) equipped with 12mm circular, flat and plain punches. Over this compressed layer, the immediate release layer of Pioglitazone HCl (150mg) was placed and compressed to obtain hardness in the range of 6-7kg/cm^2 to form a bilayered matrix tablet.

5.11 Preformulation Studies:

5.11.1 Bulk Density:

The powder blend of all formulations was evaluated separately inorder to determine their bulk densities. Powder blend was weighed (M) and later the weighed powder blend was transferred in to the measuring cylinder and volume occupied was noted (V_b).

$$D_b = \frac{\text{Mass of the powder blend (M)}}{\text{Vol. occupied by powder blend } (V_b)}$$

V_b is known as the Bulk volume and Bulk density is expressed in terms of g/ml.

5.11.2 Tapped Density:

Powder blend was transferred into the measuring cylinder and subjected for 100 tappings. The obtained volume was noted as the tapped volume. Tapped density is expressed as g/ml and tapped density is given by the formula;

$$D_t = \frac{\text{Mass of the powder blend (M)}}{\text{Tapped volume } (V_t)} .$$

5.11.3 Angle of Repose:

Angle of repose is the maximum angle possible between the surface of the pile of granules and the horizontal plane. This is one of the measures for flow properties. Powder blend was allowed to flow through the funnel attached to a stand and later height and radius of the heap of the powder blend formed was noted. Based on the height and radius obtained Angle of repose was calculated using the formula;

$$\tan(\theta) = \frac{\text{Height of the heap (h)}}{\text{Radius of the heap(r)}}$$

Table 3: Specifications of Angle of Repose:

Angle of repose	Flow property
<25	Excellent
25-30	Good
30-40	Passable
>40	Poor

5.11.4 Carr's Index (or) %Compressibility:

Carr's Index is one more measure to know the flow properties. It is indicated by the letter (I) and expressed in terms of percentage

$$I = \frac{\text{Tapped density - bulk density}}{\text{Tapped density}} \times 100$$

Table 4: Specifications of %Compressibility:

Compressibility index (%)	Flow properties
<10	Excellent
11-15	Good
16-20	Fair
21-25	Passable
26-31	Poor
32-37	Very poor
>38	Very very poor

5.11.5 Hausner's Ratio:

The Hausner ratio is a number that is correlated to the flowability of a powder or granular material.

Hausner's ratio was calculated by using the formula;

$$\text{Hausner's ratio} = \frac{\text{Tapped density}}{\text{Bulk density}}$$

Table 5: Specifications of Hausners ratio:

Hausners Ratio	Type of flow
Less than 1.25	Good Flow
1.25 – 1.5	Moderate
Greater than.5	Poor flow

5.12 Post Compressional Parameters:

5.12.1. Weight Variation:

Twenty tablets were weighed collectively and individually. Average weight was calculated and based on the obtained weights % weight variation was calculated using the formula,

$$\%\ \text{Weight Variation} = \frac{\text{Average weight} - \text{Individual weight}}{\text{Average weight}} \times 100$$

Table 6: Specifications of Weight Variation:

Average weight of tablet	% deviation
80 mg or less	10
More than 60mg but less than 250 mg	7.5
250 mg or more	5

5.12.2. Hardness:

Hardness of the tablet was tested by placing the tablet longitudinally in between the two plungers of the Monsanto tablet hardness tester and the obtained hardness was mentioned in terms of kg/sq.cm.

Limits for Hardness are 4-6kg/sq.cm.

5.12.3. Friability:

The friability of the tablets was determined by Roche Friabilator in which the tablets were subjected to the combined effect of abrasions and shock in a plastic chamber revolving at 25rpm and dropping the tablets at a height of 6 inches in each revolution.

Pre weighed sample of tablets were placed in the friabilator and allowed to rotate for 100 revolutions. Later the tablets were dedusted and the tablets were reweighed. Percent friability is given by the formula;

$$\%F = (1-W/W_0) \times 100$$

Where, W_0 is the weight of the tablets before the test

W is the weight of the tablets after the test

Limits for friability are %friability should not be more than 1%.

5.13 Estimation of Drug Content:

Equivalent to 10mg each of Pioglitazone HCl and Metformin HCl was accurately weighed from powdered bilayered tablets and it was dissolved in methanol and distilled water respectively to form a clear solution. Later it was made up to volume with methanol and distilled water respectively. One ml of the sample was withdrawn, suitably diluted

with pH 1.2 buffer and pH 6.8 phosphate buffer respectively and analysed spectrophotometrically at 269nm and 232nm respectively.

5.14 *In vitro* Dissolution Studies:

An *in vitro* drug release study from the prepared bilayered tablets, in triplicate , was determined using the USP eight station Dissolution Rate Test Apparatus(model QAE 016 and NRE 002, M/S Campbell Electronics) employing a paddle stirrer . With 900 ml of pH of 1.2 and followed by phosphate buffer pH 6.8 was used as dissolution media and maintained at 37±0.5^{o}C at a rotational speed of 100 rpm, for 2 hrs and 10 hrs respectively. Then the dissolution samples were analysed in UV-VIS double beam spectrophotometer, while keeping the dissolution media as a blank at 232nm.

5.15 Drug Release Kinetics:

To study the mechanism of drug release from the SR layer of the matrix tablets, the dissolution data were fitted into the following equations:

5.15.1 *Zero order equation:*

$$Q_t = Q_0 + k_{0t} \qquad \text{---------- (1)}$$

Where, Q_t is the amount of drug released at time t, Q_0 is the initial amount of drug in the solution (more times, Q_0 =0) and k_0 is the zero-order release rate (Kenneth A. Connors, 1991).

5.15.2 *First order equation:*

$$\ln Q_t = \ln Q_0 = k_1 t \qquad \text{---------- (2)}$$

Where, Q_t is the amount of drug released at time t, Q_0 is the initial amount of drug in the solution and k_1 is the first-order release rate constant (Kenneth A. Connors, 1991).

5.15.3 ***Higuchi's equation:***

$$Q = k_H t^{1/2} \quad \text{---------- (3)}$$

Where, Q is the amount of drug released at time t, k_H is the Higuchi diffusion rate constant (Higuchi W I, 1962).

5.15.4 ***Koresmeyer's equation*****:**

$$M_t / M_\infty = Kt^n \quad \text{---------- (4)}$$

Where, M_t is the amount of drug released at time t, M_∞ is the amount of drug released after infinite time, k is a kinetic constant incorporating structural and geometric characteristics of the tablet, and n is the diffusional exponent of the drug release mechanism (Koresmeyer et al., 1977).

5.16 Similarity factor:

The similarity factor (f_2) is used to compare the dissolution profile of each formulation with that of marketed formulation. In this approach, recommended by the FDA guidance for the industry, a value between 50 and 100, the two profiles are nearly identical (Shah V.P et al 1998).

6. Results and Discussion

6.1 Estimation of Pioglitazone HCl:

Table 7: Calibration Curve Data

S.No.	Concentration(µg/mL)	Absorbance
1	5	0.093±0.001
2	10	0.192±0.003
3	15	0.291±0.004
4	20	0.373±0.003
5	25	0.449±0.002

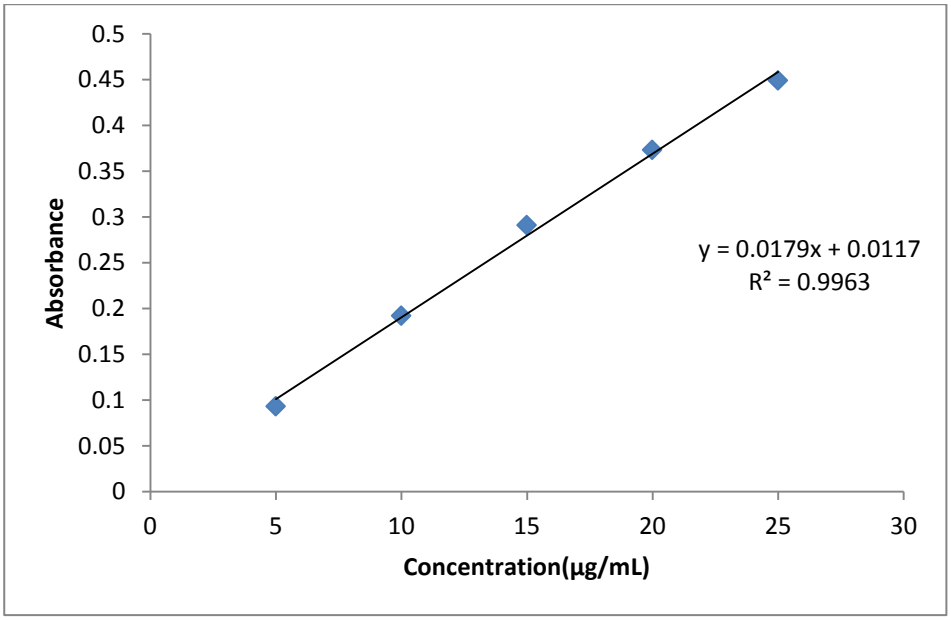


Figure 9: Calibration Curve for Pioglitazone HCl

6.2 Estimation of Metformin HCl:

Table 8 :Calibration Curve Data

(In pH 1.2 buffer)

S.No.	Concentration(µg/mL)	Absorbance
1	2	0.025±0.003
2	4	0.042±0.002
3	6	0.060±0.003
4	8	0.074±0.001
5	10	0.089±0.002

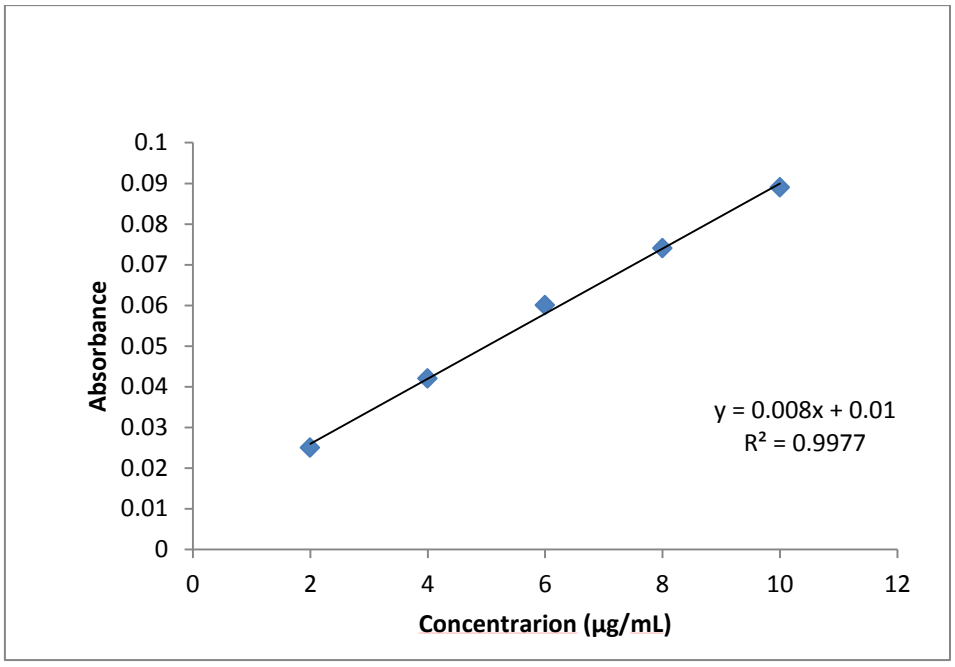


Figure 10: Calibration Curve for Metformin HCl

(In pH 1.2 buffer)

Table 9: Calibration Curve Data

(In pH 6.8 phosphate buffer)

S.No.	Concentration(µg/mL)	Absorbance
1	2	0.156±0.001
2	4	0.292±0.001
3	6	0.442±0.002
4	8	0.585±0.001
5	10	0.728±0.003

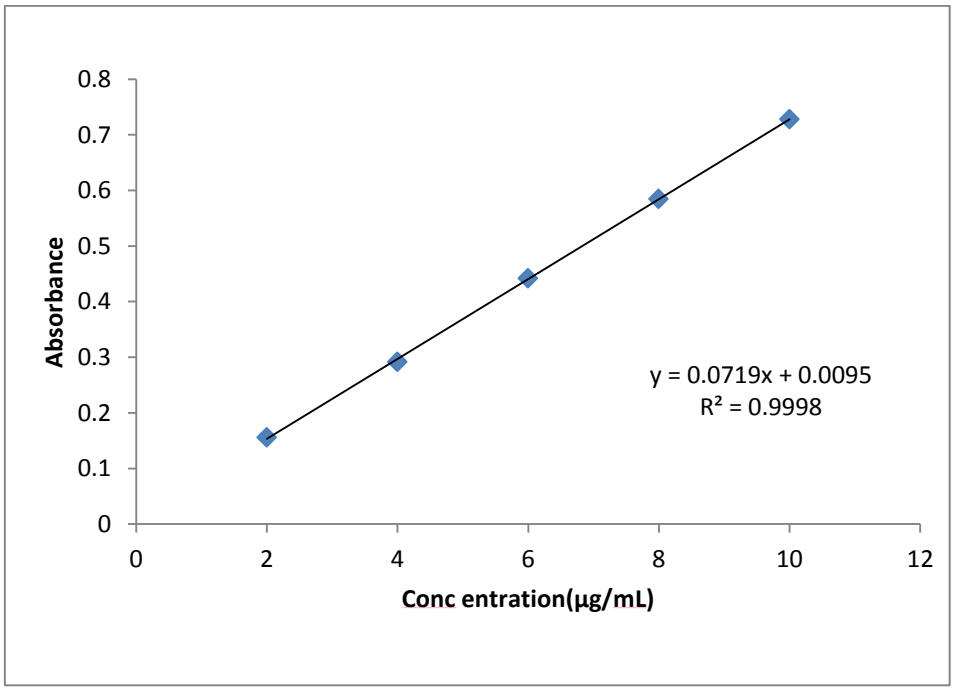


Figure 11: Calibration Curve for Metformin HCl

(In pH 6.8 phosphate buffer)

Table 10 : Calibration Curve Data

(In Distilled water)

S.No.	Concentration(µg/mL)	Absorbance
1	2	0.142±0.002
2	4	0.283±0.002
3	6	0.422±0.001
4	8	0.560±0.003
5	10	0.684±0.002

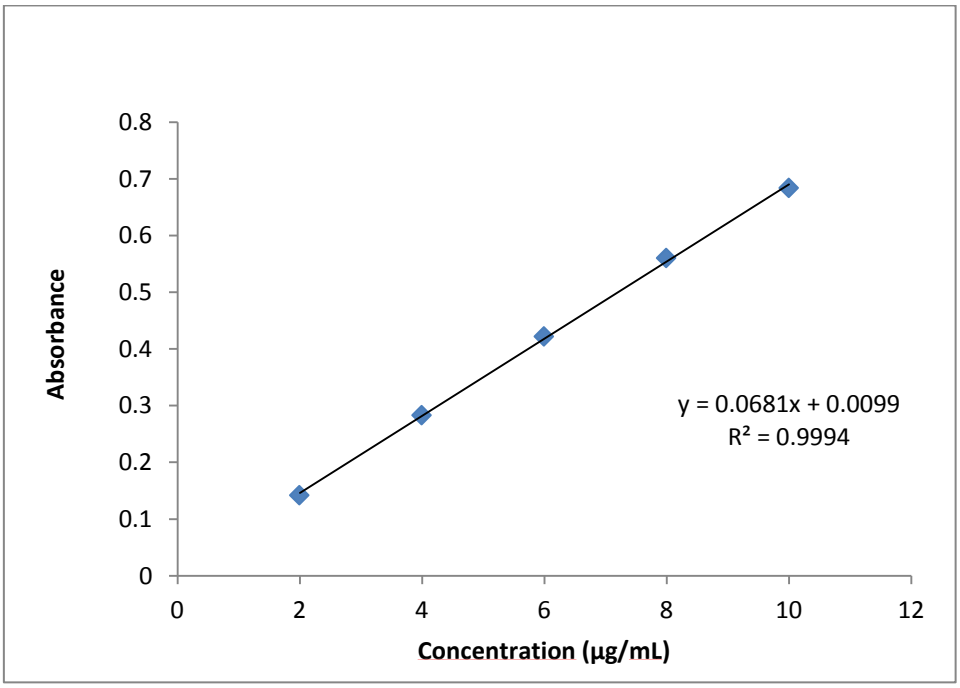


Figure 12: Calibration Curve for Metformin HCl

(In Distilled water)

6.3 Preformulation Studies for IR layer:

Preformulation studies were carried out through Drug-Excepient compatibility studies using FTIR.

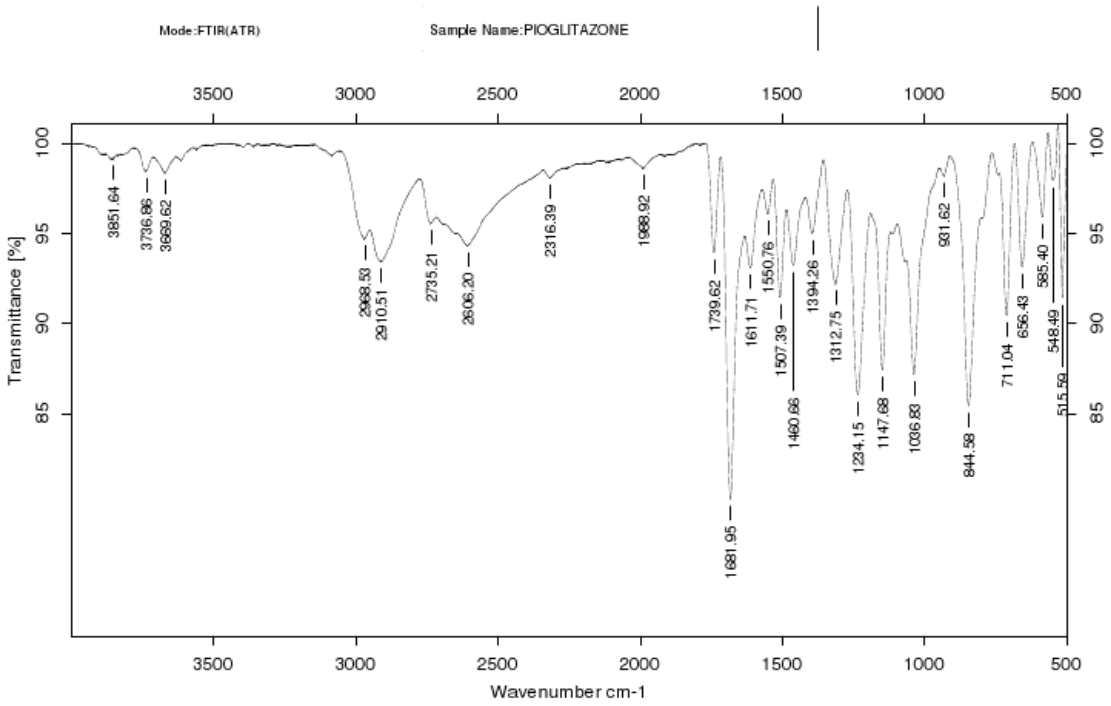


Figure 13: FT-IR Spectrum of pure Pioglitazone HCl

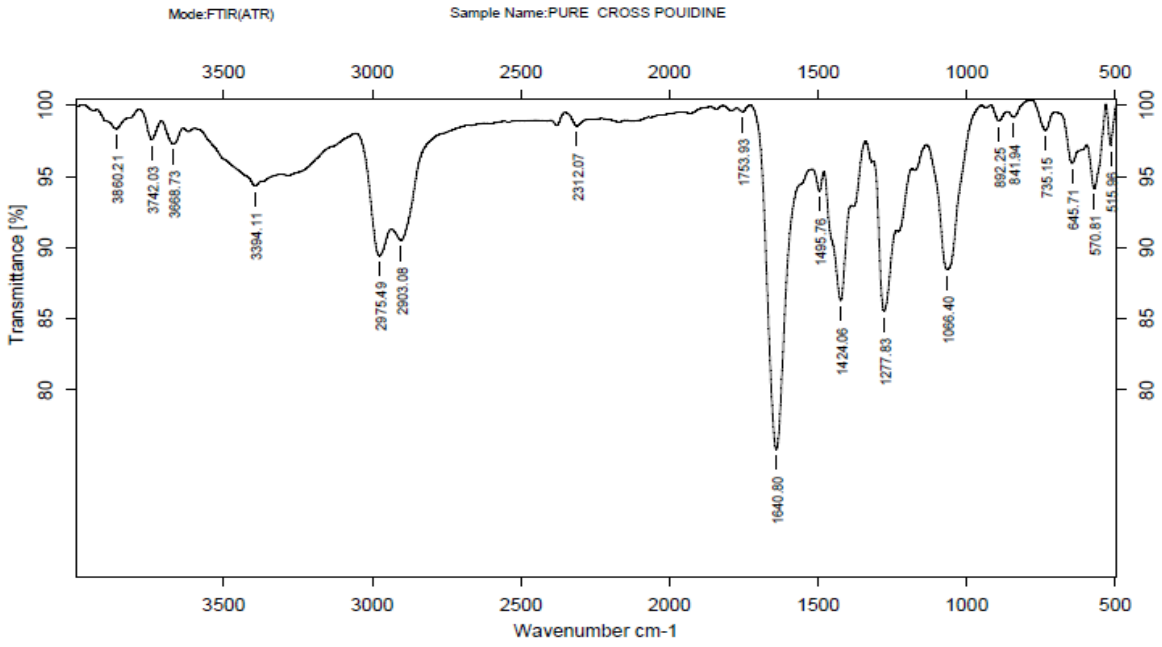


Figure 14: FT-IR Spectrum of pure Crospovidone

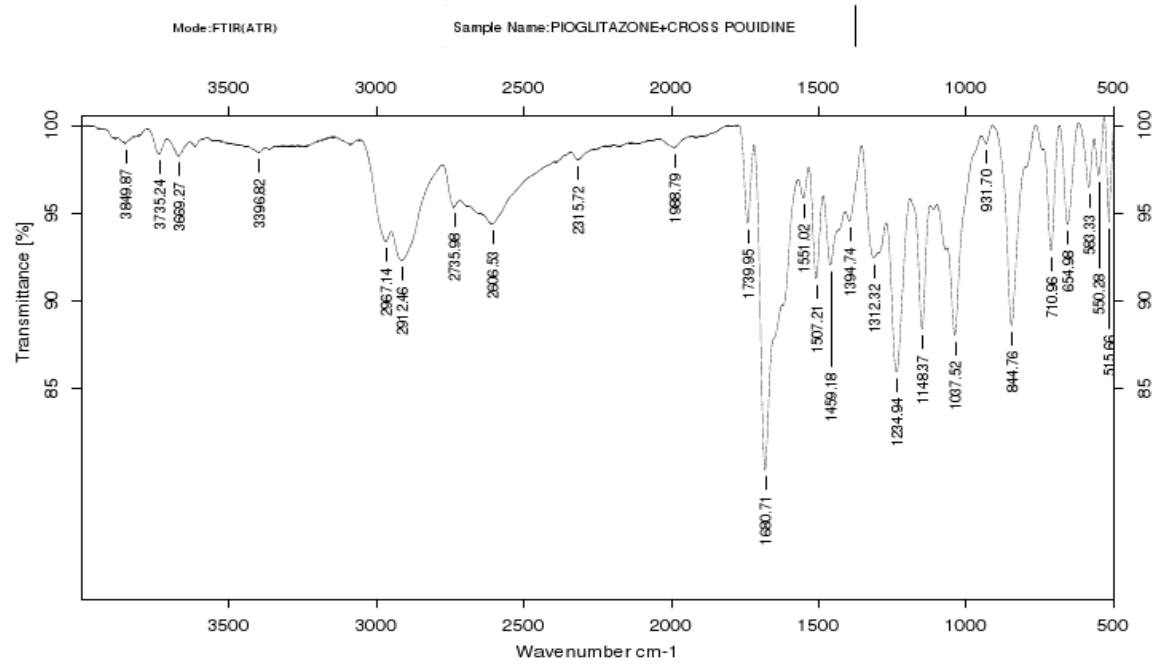


Figure 15: FT-IR Spectrum of prepared formulation

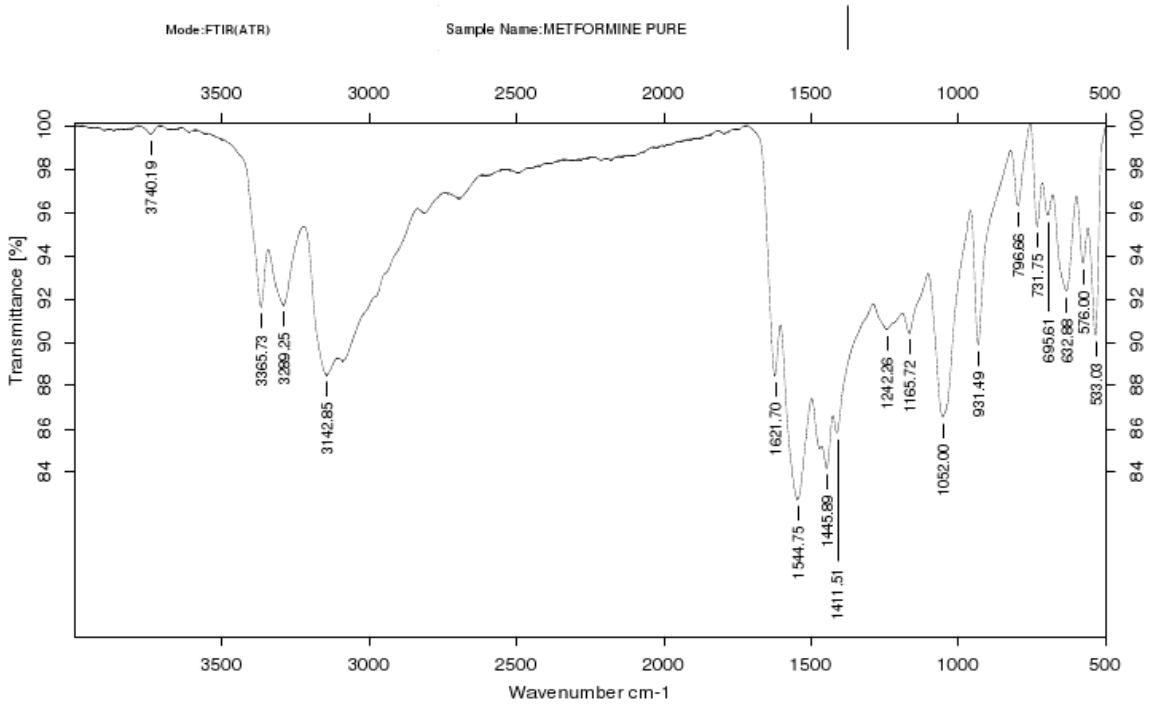


Figure 16: FT-IR Spectrum of pure Metformin HCl

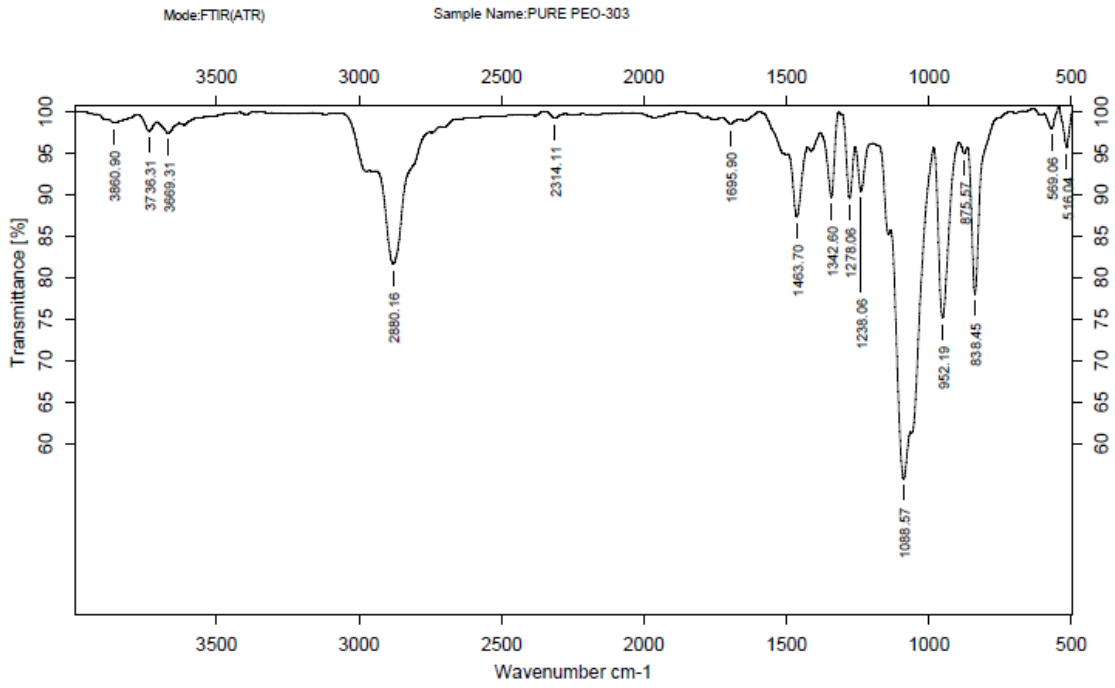


Figure 17: FT-IR Spectrum of pure PEO-303

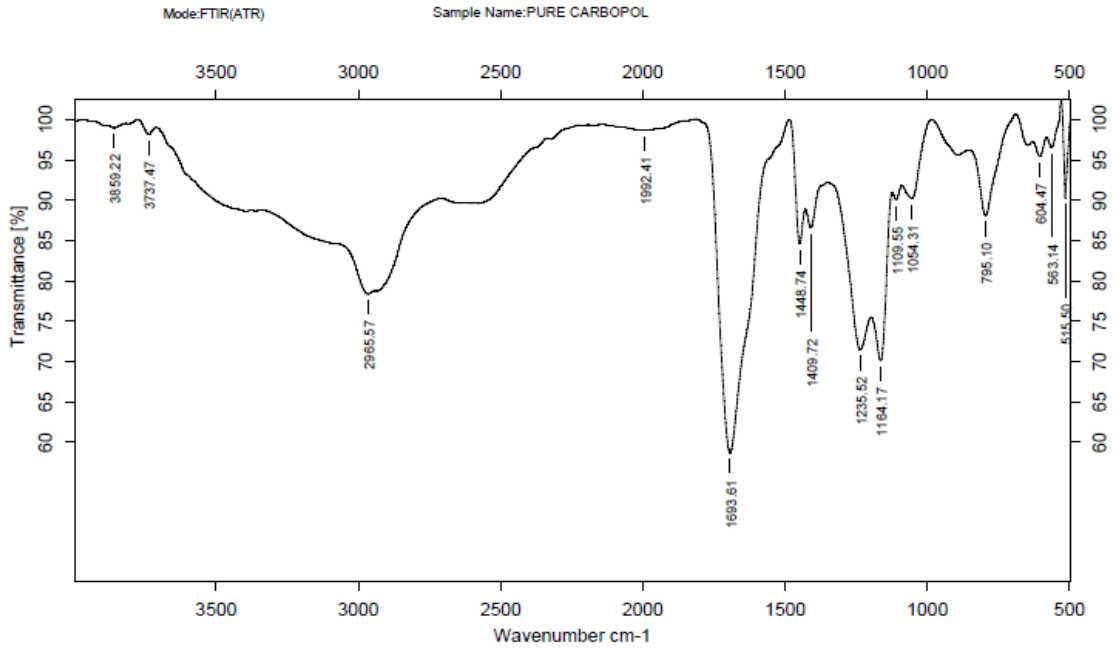


Figure 18:FT-IR Spectrum of pure CARBOPOL 971P

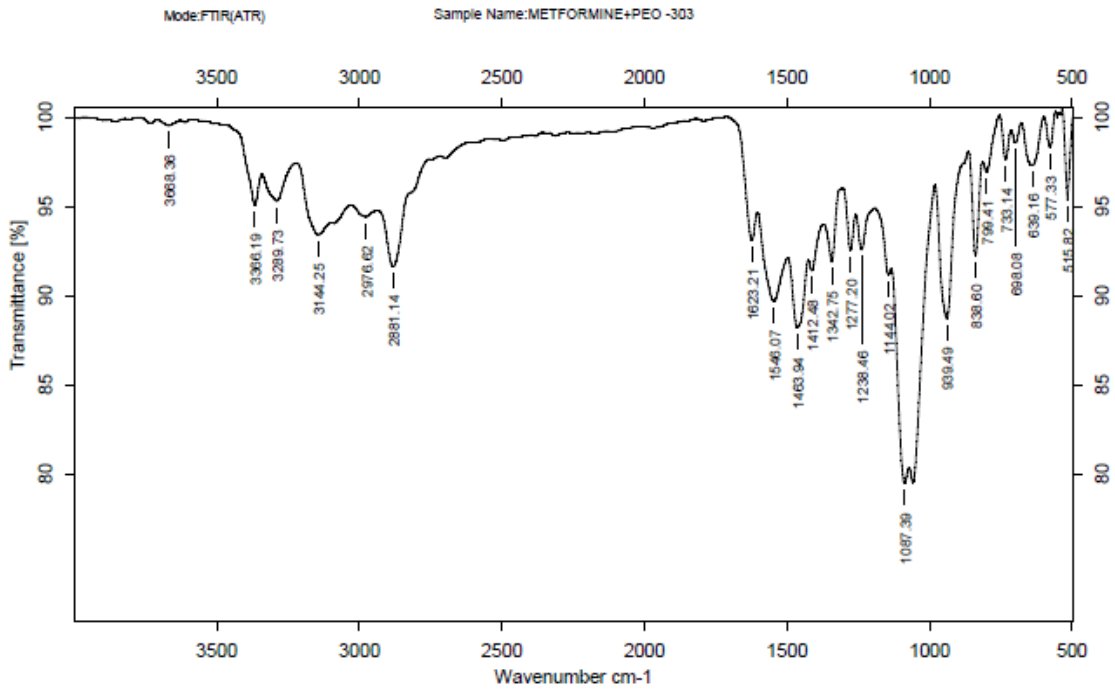


Figure 19: FT-IR Spectrum of formulation prepared with PEO-303

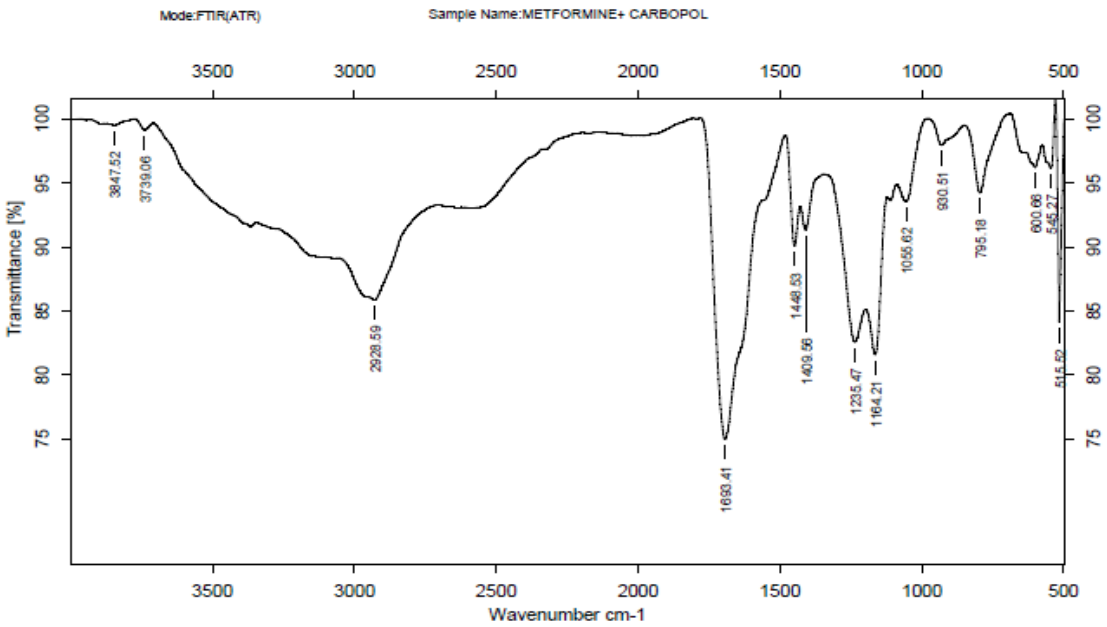


Figure 20: FT-IR Spectrum of formulation prepared with CARBOPOL 971P

6.4 Micromeretic properties:

Table 11: Micromeretic parameters for IR layer formulations

Formulation code	Micromeretic properties				
	Bulk density (gm/ml)	Tapped density (gm/ml)	Angle of repose (^{o})	Compressibility index (%)	Hausner's ratio
F1	0.598	0.643	21.5	16.36	1.16
F2	0.659	0.732	21	15.97	1.23
F3	0.688	0.768	22.5	16.08	1.19

Table 12: Micromeretic Parameters for SR layer Formulated with PEO-303

Formulation code	Micromeretic properties				
	Bulk density (gm/ml)	Tapped density (gm/ml)	Angle of repose (^{o})	Compressibility index (%)	Hausner's ratio
F4	0.576	0.654	21.5	16.07	1.18
F5	0.532	0.612	23	15.90	1.17
F6	0.569	0.636	21	15.50	1.19
F7	0.592	0.664	22.5	16.67	1.21
F8	0.657	0.743	22	16.07	1.16
F9	0.689	0.776	24	17.63	1.14

Table 13: Micromeretic Parameters for SR layer Formulated with CARBOPOL 971P

Formulation code	Micromeretic properties				
	Bulk density (gm/ml)	Tapped density (gm/ml)	Angle of repose (^{o})	Compressibility index (%)	Hausner's ratio
F10	0.546	0.624	22.5	16.09	1.19
F11	0.572	0.642	22	16.76	1.16
F12	0.587	0.676	21.5	16.50	1.21
F13	0.643	0.714	21	15.97	1.18
F14	0.623	0.703	21.5	16.50	1.17

6.5 Formulation tables:

Table 14: Composition for IR layer of Bilayered tablet

S.No	Ingredients (mg/tab)	Formulations		
		F1	**F2**	**F3**
1	Pioglitazone HCl	15	15	15
2	Cross povidone	2.5	5	7.5
3	Lactose	74.5	72	69.5
4	MCC	50	50	50
5	Aerosil	3	3	3
6	Mg stearate	3	3	3
7	Sunset yellow	2	2	2
Total Weight		150	150	150

Table 15: Composition for SR layer of Bilayered Tablets prepared by Direct Compression technique

S.No.	Ingredients (mg/tab)	Formulations					
		F4	**F5**	**F6**	**F7**	**F8**	**F9**
1	Metformin HCl	500	500	500	500	500	500
2	PEO-303	50	100	150	200	250	296
3	MCC	246	196	146	96	46	-
4	Mg stearate	2	2	2	2	2	2
5	Talc	2	2	2	2	2	2
Total Weight		800	800	800	800	800	800

Table 16: Composition for SR layer of Bilayered Tablets prepared by Wet Granulation technique

S.No.	Ingredients (mg/tab)	Formulations				
		F10	**F11**	**F12**	**F13**	**F14**
1	Metformin HCl	500	500	500	500	500
3	Carbopol 971P	50	75	100	150	200
4	MCC	246	221	196	146	96
5	Mg stearate	2	2	2	2	2
6	Talc	2	2	2	2	2
7	Diluting Fluid (ethanol and water 1:1)	qs	qs	qs	qs	qs
Total Weight		800	800	800	800	800

6.6 Post Compressional Parameters:

Table 17: Post Compressional Parameters Bilayered Tablets of Pioglitazone HCl and Metformin HCl by Direct Compression technique

Formulation code	Average Weight (±SD)	Thickness (mm)	Hardness kg/cm^2	Friability (%)	Drug Content (%)	
					Pioglitazone HCl	Metformin HCl
F4	950.4±0.55	8.47±0.01	7.30±0.04	0.23±0.05	98.75±0.73	99.40±0.77
F5	948.6±1.34	8.48±0.03	7.34±0.42	0.21±0.06	98.26±1.08	98.89±1.73
F6	947.6±0.89	8.52±0.03	7.21±0.23	0.2±0.05	98.50±1.05	100.20±0.45
F7	948.8±0.84	8.52±0.02	7.11±0.17	0.17±0.04	97.09±0.73	99.39±0.73
F8	950.4±0.52	8.51±0.02	7.17±0.30	0.19±0.03	99.03±0.89	99.62±0.86
F9	949.6±0.48	8.50±0.03	7.28±0.24	0.20±0.08	99.24±0.56	99.64±0.52

Table 18: Post Compressional Parameters of Pioglitazone HCl and Metformin HCl by Wet Granulation technique

Formulation code	Average Weight (±SD)	Thickness (mm)	Hardness kg/cm^2	Friability (%)	Drug content (%)	
					Pioglitazone HCl	Metformin HCl
F10	950.4±0.35	8.14±0.05	7.28±0.11	0.22±0.04	99.75±0.43	99.10±0.37
F11	949.6±0.98	8.09±0.02	7.30±0.41	0.21±0.02	99.26±1.18	99.37±1.03
F12	950.6±0.46	8.19±0.02	7.21±0.43	0.23±0.04	98.90±1.01	99.28±0.75
F13	951.8±0.78	8.19±0.03	7.19±0.15	0.19±0.03	99.09±0.23	99.09±0.33
F14	950.6±0.41	8.10±0.05	7.14±0.20	0.17±0.04	98.03±0.59	98.72±0.46

6.7 Cumulative Percent Drug Release:

Table 19: Cumulative Percent Drug Release data for IR layer formulations:

Time in minutes	Cumulative % drug release		
	F1	F2	F3
0	0	0	0
1	29.06±1.62	39.38±1.65	68.13±0.31
2	37.50±0.31	51.15±1.10	74.06±0.36
3	47.71±1.26	62.19±0.31	77.50±0.18
4	53.96±0.95	71.77±0.48	82.19±0.36
5	61.35±0.95	76.25±0.83	88.75±0.18
6	64.79±0.79	80.63±0.54	91.56±0.18
7	68.65±0.36	84.48±0.36	96.56±2.17
8	72.50±1.08	89.06±0.31	97.19±1.98
9	75.94±1.43	91.56±0.83	97.81±0.72
10	78.65±0.79	92.40±0.36	99.69±0.18

Table 20: Cumulative Percent Drug Release for Bilayered Tablets Formulated by Direct Compression technique

Time (hrs)	Cumulative %drug released					
	F4	**F5**	**F6**	**F7**	**F8**	**F9**
0	0	0	0	0	0	0
1	94.62±0.37	90.67±0.37	87.3±0.37	62.64±0.78	32.64±1.16	20.76±1.64
2	95.04±0.00	93.43±0.19	90.57±0.10	80.94±0.10	46.26±0.94	31.64±1.39
3	-	-	-	94.79±0.59	61.25±1.48	39.05±1.42
4	-	-	-	96.11±0.16	67.38±1.86	47.5±1.06
6	-	-	-	-	76.31±1.80	54.17±0.97
8	-	-	-	-	84.3±1.92	60.31±1.29
10	-	-	-	-	91.3±1.32	69.47±1.02
12	-	-	-	-	99.06±0.09	79.62±0.11

Table 21: Cumulative Percent Drug Release for Bilayered Tablets Formulated by Wet Granulation technique:

Time in hrs	Cumulative %drug released				
	F10	**F11**	**F12**	**F13**	**F14**
0	0	0	0	0	0
1	61.38±0.37	48.84±0.63	42.3±0.62	36.6±0.58	32.88±0.99
2	82.68±1.40	63.9±0.65	59.7±0.37	50.1±0.68	44.1±1.00
3	-	78.56±0.75	81.69±0.61	67.53±0.35	61.29±1.36
4	-	83.04±0.93	85.60±0.86	70.14±1.00	64.38±1.73
6	-	-	89.51±0.54	79.26±0.46	68.45±1.23
8	-	-	-	82.68±0.59	70.73±1.03
10	-	-	-	92.78±0.52	73.42±1.11
12	-	-	-	99.21±0.05	74.72±1.00

6.8 Cumulative Percent Drug Release Profiles:

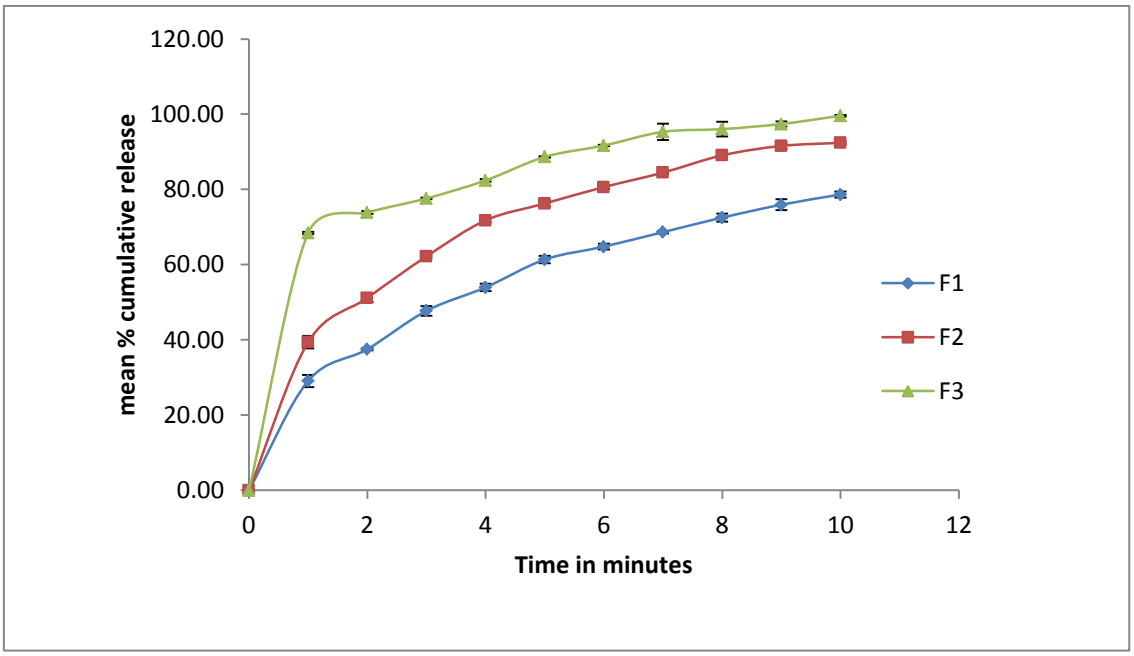


Figure 21: Cumulative Percent Drug Release profiles for IR layer formulations

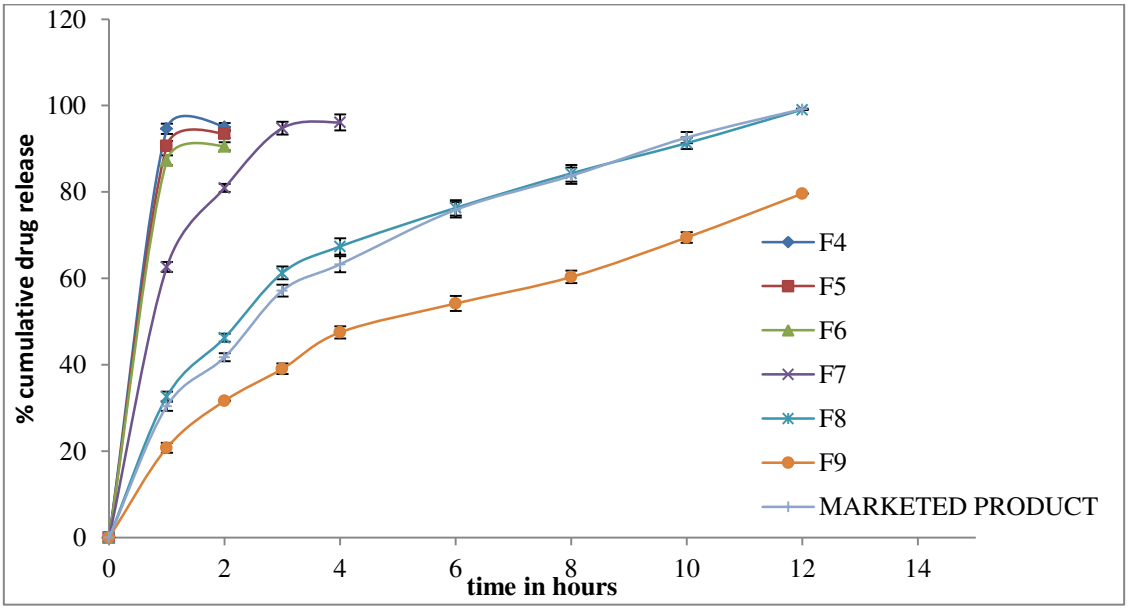


Figure 22:Cumulative Percent Drug Release profiles for Bilayered Tablets Formulated by Direct Compression technique

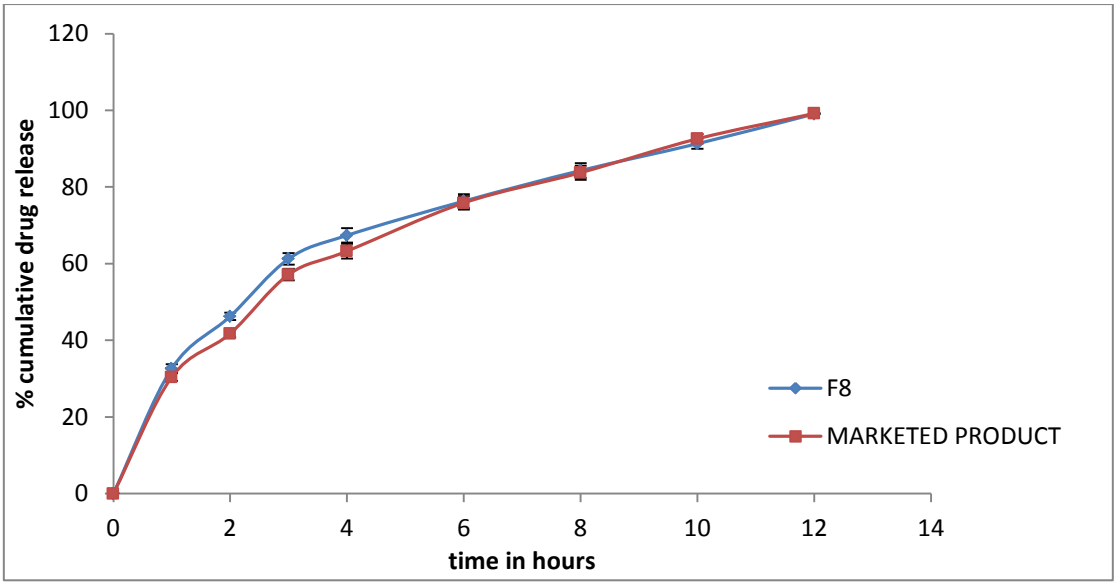


Figure 23: Comparision of Percent Drug Release profiles for F8 and Marketed formulation

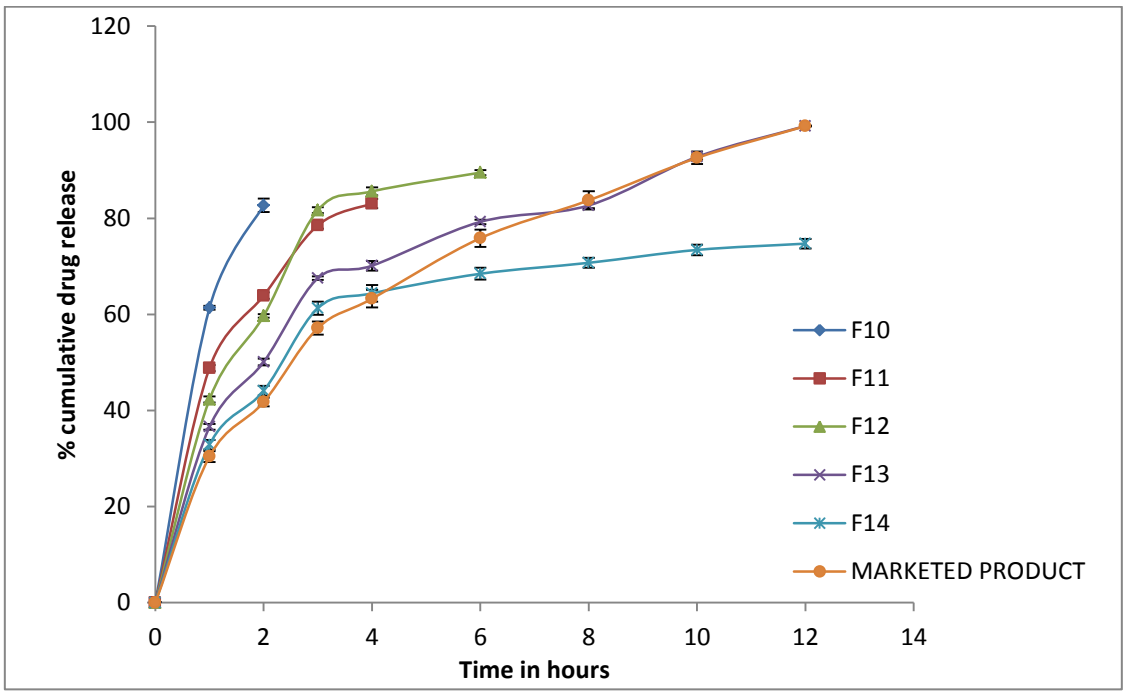


Figure 24: Cumulative Percent Drug Release profiles for Bilayered Tablets Formulated by Wet Granulation technique

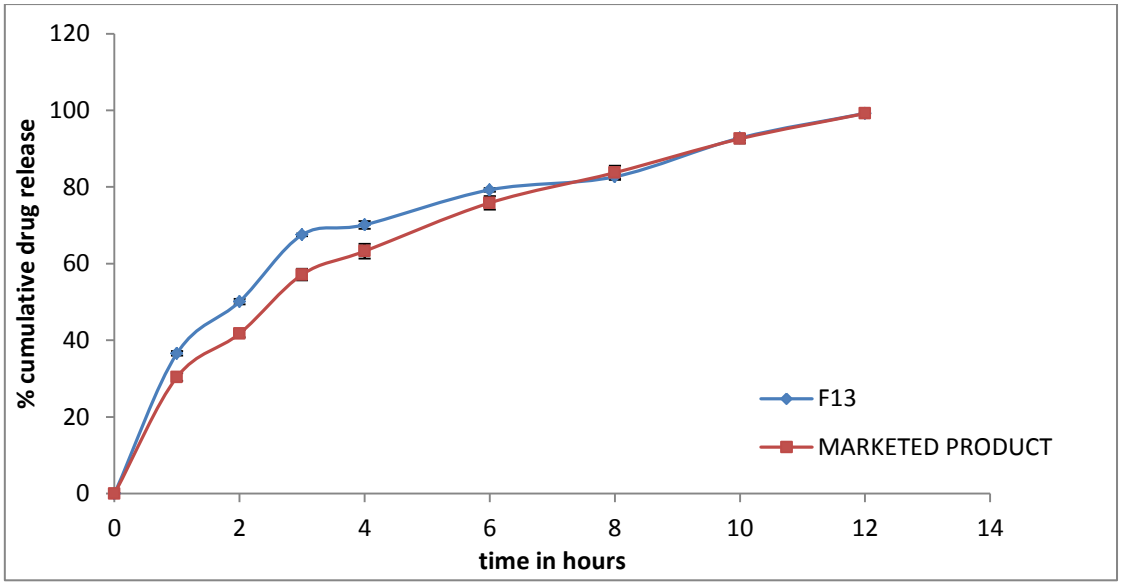
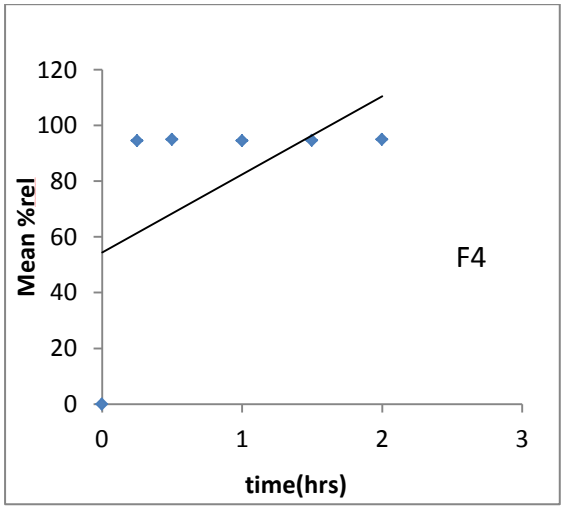
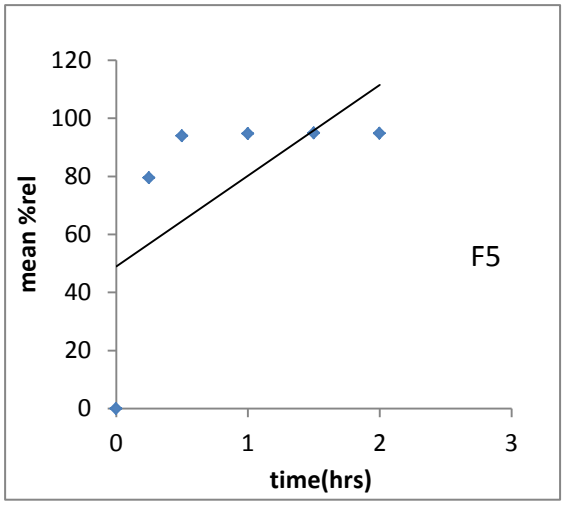
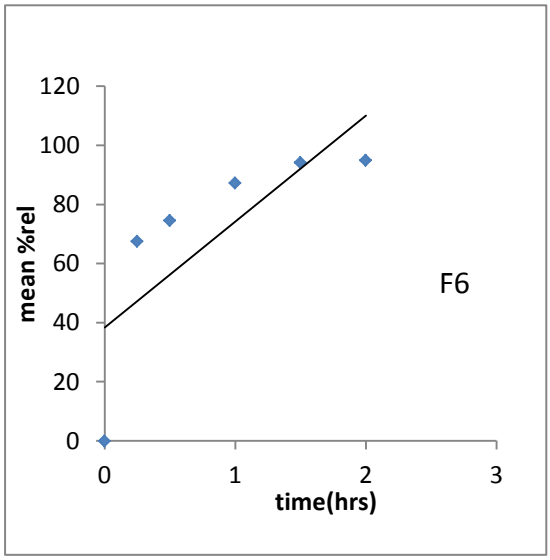


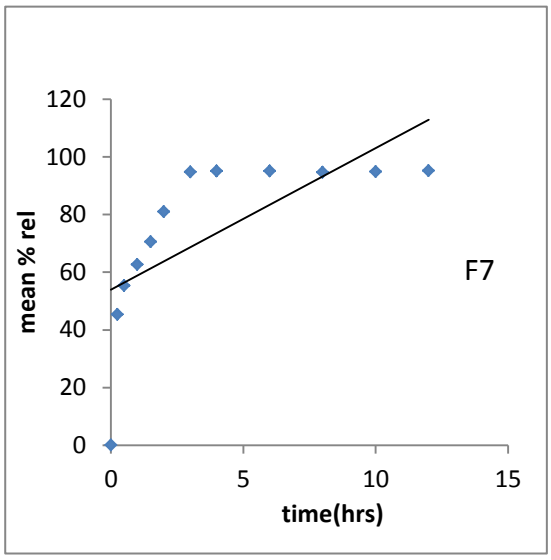
Figure 25: Comparision of Percent Drug Release profiles for F13 and Marketed formulation

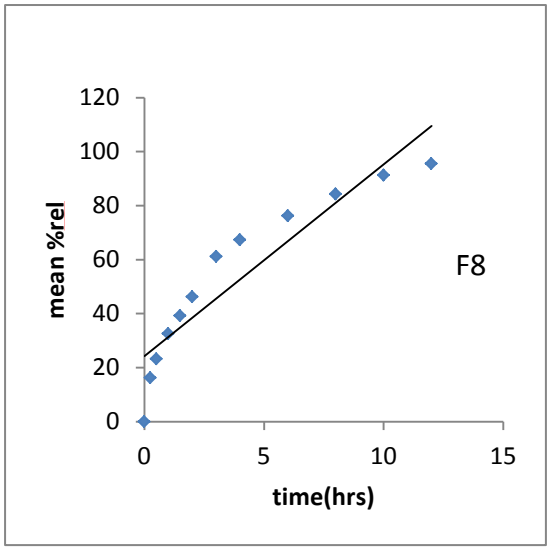
6.9 Zero Order Release Plots:











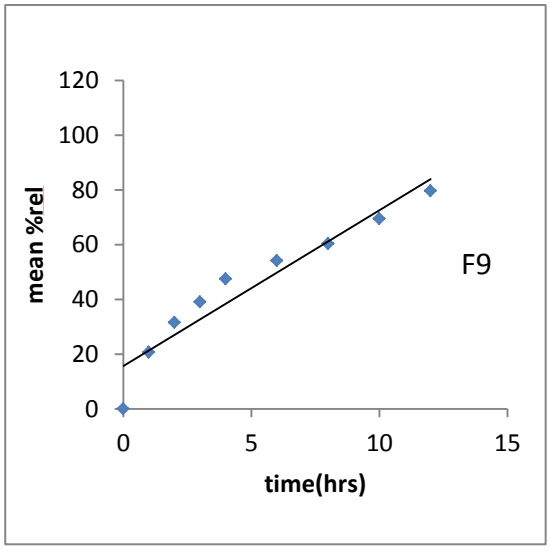


Figure 26: Zero Order Release Plots for Bilayered Tablets Formulated by Direct Compression technique

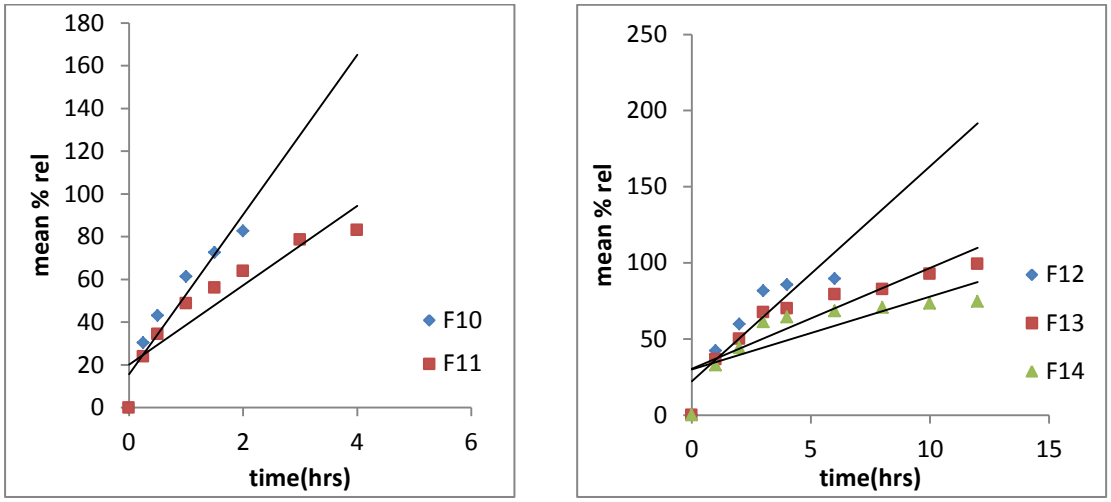


Figure 27: Zero Order Release Plots for Bilayered Tablets Formulated by Wet Granulation technique

6.10 First Order Release Plots:

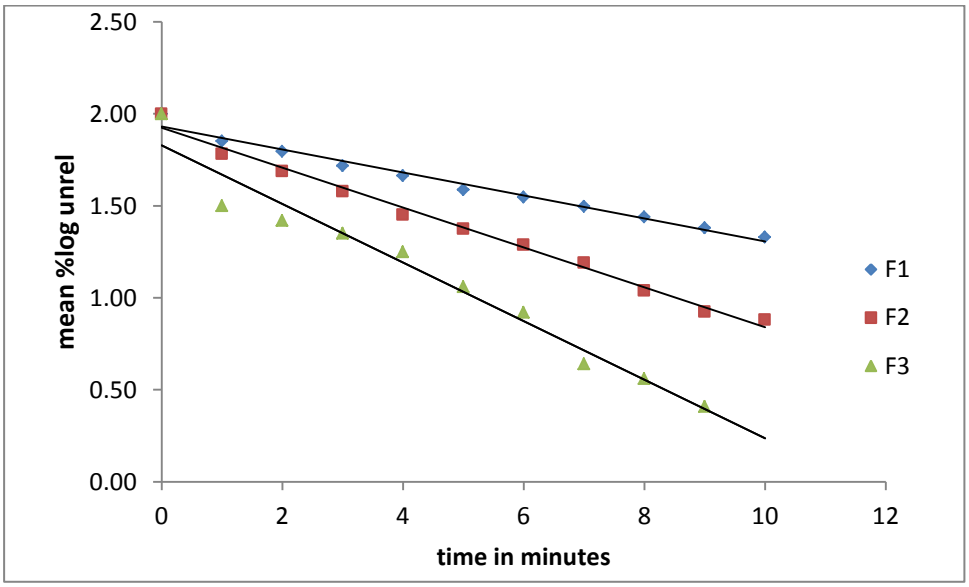


Figure 28: First Order Release Plots for IR layer formulations

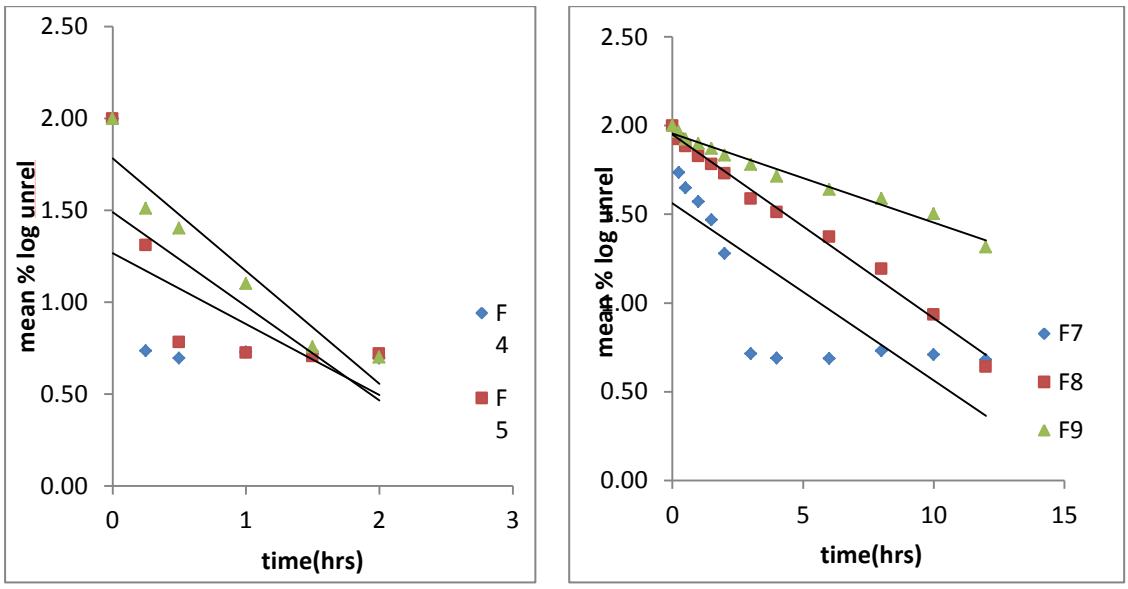


Figure 29: First Order Release Plots for Bilayered Tablets Formulated by Direct Compression technique

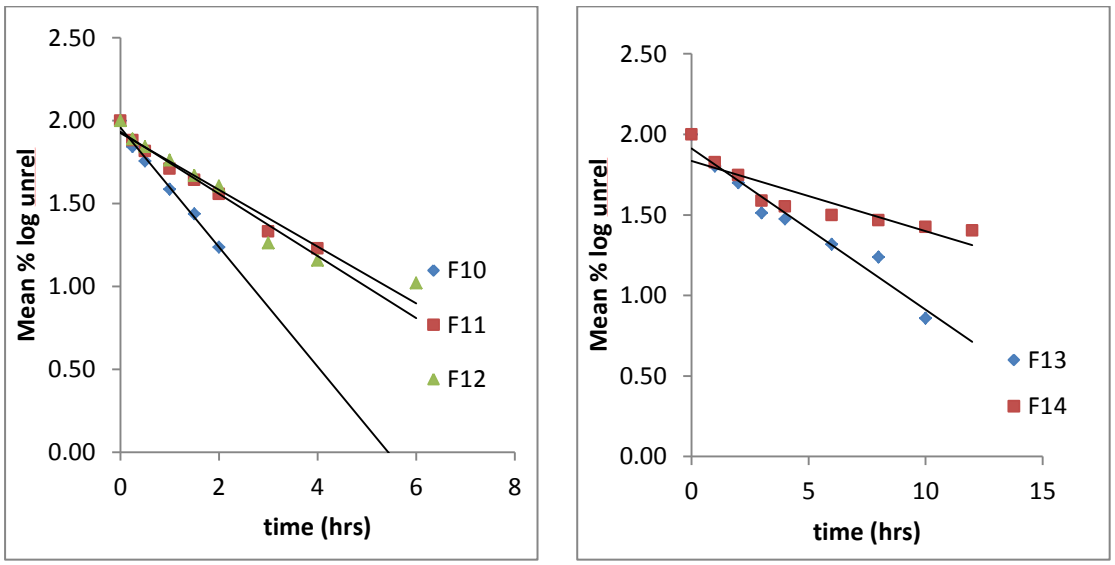


Figure 30: First Order Release Plots for Bilayered Tablets Formulated by Wet Granulation technique

6.11 Higuchi's Plots:

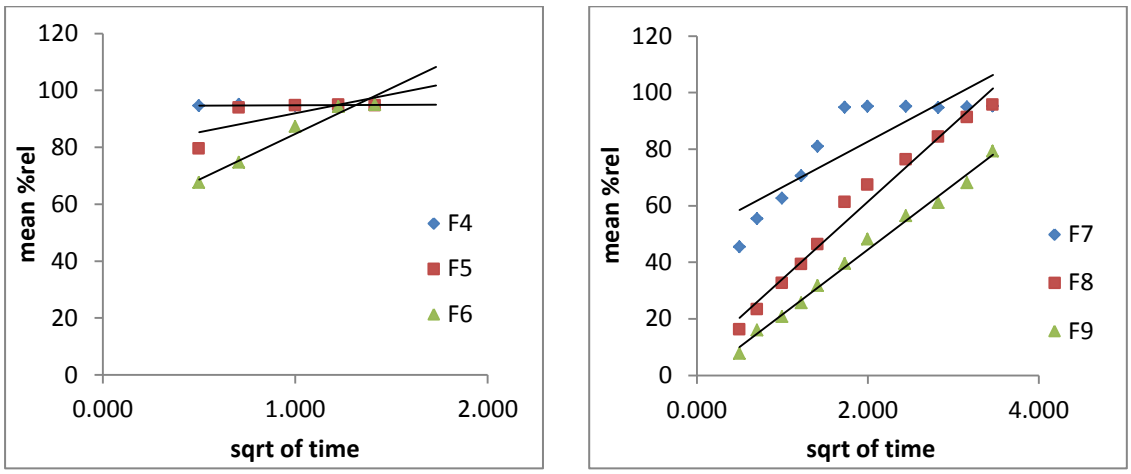


Figure 31: Higuchi's Plots for Bilayered Tablets Formulated by Direct Compression technique

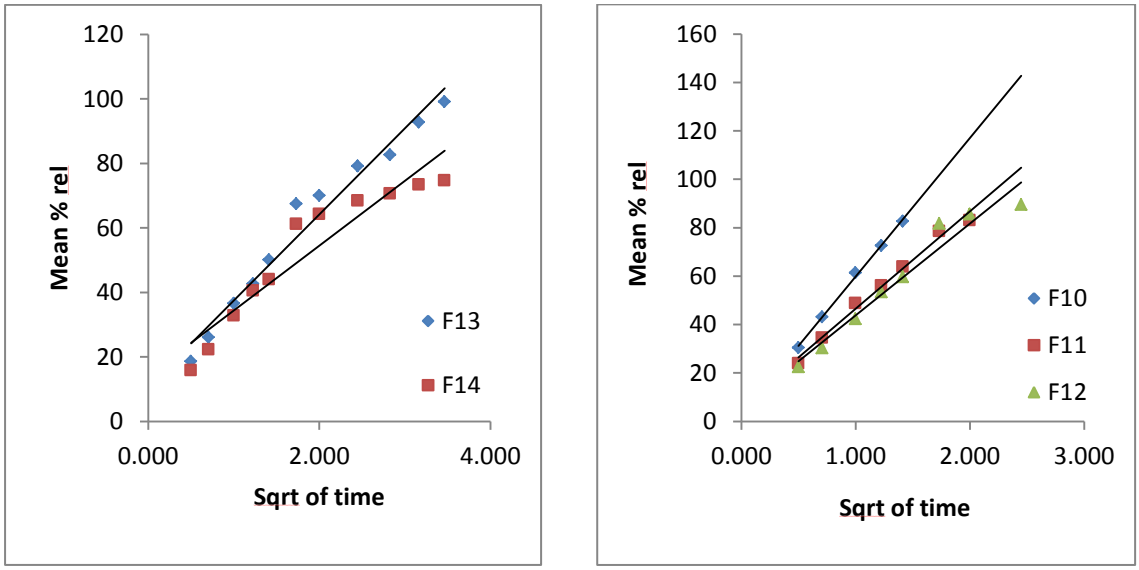


Figure 32: Higuchi's Plots for Bilayered Tablets Formulated by Wet Granulation technique

6.12 Peppas Plots:

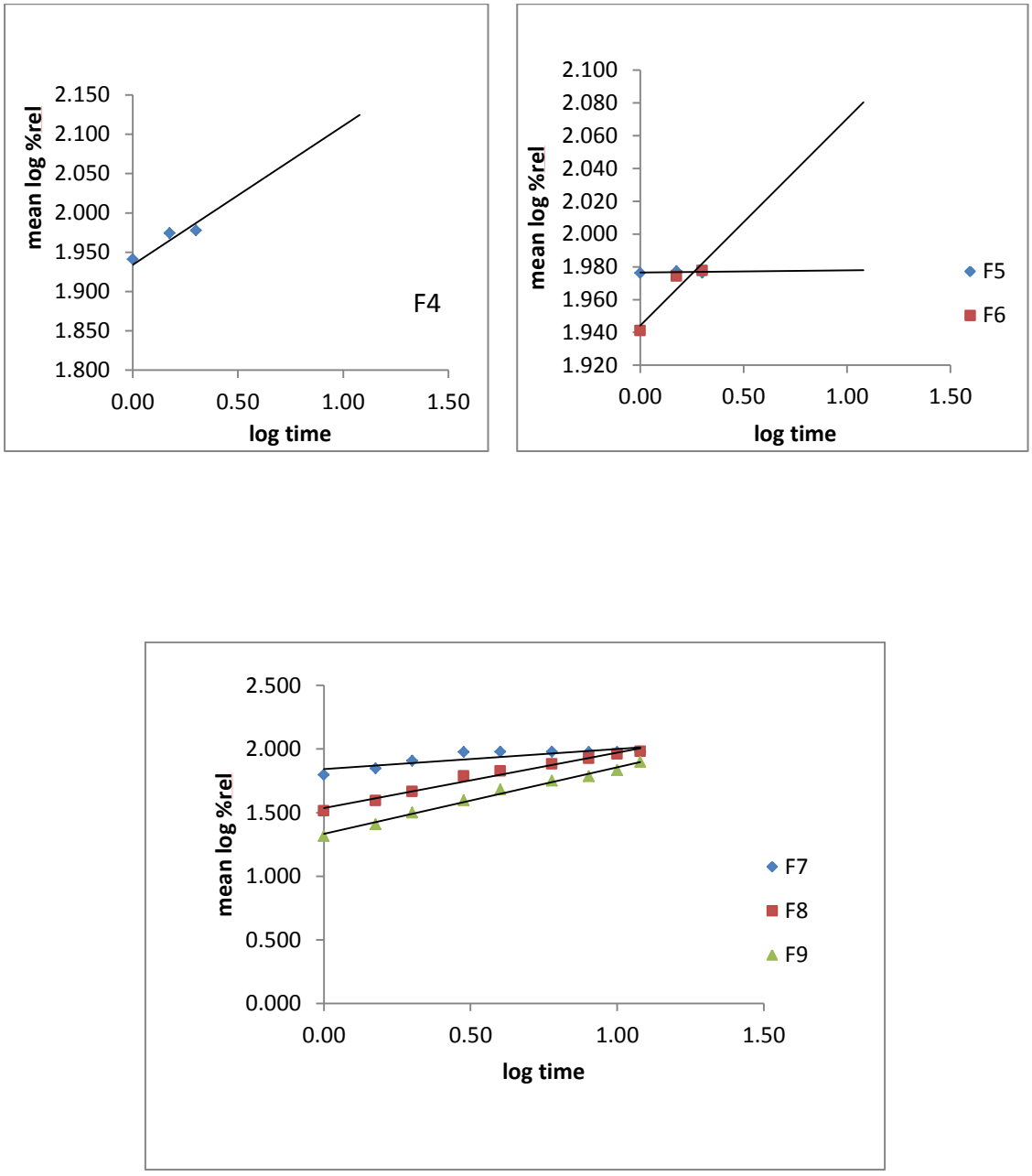


Figure 33: Peppas Plots for Bilayered Tablets Formulated by Direct Compression technique

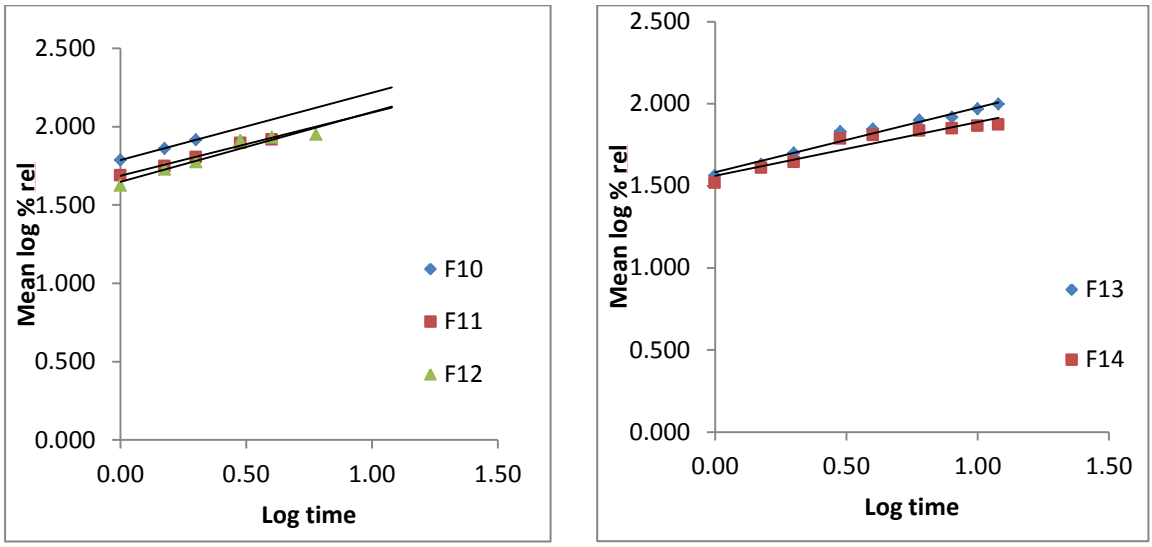


Figure 34: Peppas Plots for Bilayered Tablets Formulated by Wet Granulation technique

6.13 *In-vitro* Drug Release Kinetics:

Table 22: *In-vitro* Drug Release Kinetics for IR layer formulations:

Formulation code	Correlation coefficient		$K(hr^{-1})$
	Zero order ($mol.L^{-1}.min^{-1}$)	First order (min^{-1})	
F1	0.875	0.981	0.142
F2	0.812	0.990	0.248
F3	0.600	0.916	0.432

Table 23: *In-vitro* Drug Release Kinetics for Bilayered Tablets Formulated by Direct Compression technique:

Formulation Code	Zero order		First order		Higuchi		Peppas	
	K Value ($mol.L^{-1}.h^{-1}$)	r^2 value	K value (h^{-1})	r^2 value	K value	r^2 value	n value	r^2 value
F4	28.03	0.311	0.888	0.321	0.245	0.159	0.001	0.159
F5	31.27	0.405	1.179	0.567	13.40	0.546	0.075	0.665
F6	35.86	0.592	1.440	0.915	32.13	0.961	0.176	0.983
F7	4.90	0.464	0.227	0.646	16.12	0.743	0.199	0.896
F8	7.10	0.855	0.237	0.990	27.36	0.979	0.466	0.990
F9	5.69	0.911	0.115	0.917	22.92	0.983	0.589	0.991

Table 24: *In-vitro* Drug Release Kinetics for Bilayered Tablets Formulated by Wet Granulation technique:

Formulation Code	Zero order		First order		Higuchi		Peppas	
	K value ($mol.L^{-1}.h^{-1}$)	r^2 value	K value (h^{-1})	r^2 value	K value	r^2 value	n value	r^2 value
F10	37.35	0.896	0.829	0.991	57.25	0.997	0.482	0.998
F11	18.57	0.859	0.430	0.979	40.20	0.988	0.454	0.994
F12	14.07	0.820	0.393	0.948	37.95	0.952	0.469	0.983
F13	7.010	0.830	0.313	0.891	26.71	0.968	0.430	0.985
F14	5.251	0.716	0.108	0.836	20.09	0.892	0.408	0.952

6.14 Discussion:

The present work was carried out on the Design and Development of Bilayered tablets of Pioglitazone HCl and Metformin HCl comprising of immediate release layer for sudden onset of action followed by Sustained release layer to maintain the steady state concentrations of the drug. Micro crystalline cellulose used as diluents and release retarding polymers Polyethylene oxides (PEO-303) and Carbopol 971P were used in this investigation.

Calibration plots for Pioglitazone HCl and Metformin HCl shows good linearity indicating that selection of UV-spectrophotometry method for estimation of above named drugs is correct.

6.14.1 Preformulation studies:

Drug and Excepient compatibility studies of IR layer:

The individual IR spectra of Pioglitazone HCl, Crospovidone and of optimized formulation **(F3)** were shown in the figures 6.3.1, 6.3.2, and 6.3.3**.** The following principle peaks were observed from the IR spectral analysis.

The following principle peaks were observed from IR spectra of Pioglitazone HCl:

- N-H Stretching - 2967.14cm^{-1}
- C-H Stretching - 2912.46cm^{-1}
- C-H Stretching - 2735.98cm^{-1}
- C = O Stretching - 1739.95cm^{-1}
- C-O-Ar Stretching - 1242.94cm^{-1}

The observed principle peaks were identical in the IR spectra of drug and in the IR spectra of the optimized formulation **(F3)**. Hence there was no chemical or physical interaction between the drug and the excipient used in this investigation.

Drug and Excepient compatibility studies of SR layer:

The individual IR spectra of Metformin HCl, PEO-303, Carbopol 971P and of optimized formulations **(F8** and **F13)** were shown in the figures 6.3.4, 6.3.5, 6.3.6, 6.3.7 and 6.3.8. The following principle peaks were observed from the IR spectral analysis.

The following principle peaks were observed from IR spectra of Metformin HCl:

- N-H (Primary amine) Stretching - 3366.19cm^{-1}
- N-H (Primary amine) Stretching - 3289.73cm^{-1}
- N-H (Secondary amine) Stretching - 3144.25cm^{-1}
- C -N Stretching - 1623.21cm^{-1}
- C -N Stretching - 1546.07cm^{-1}

The observed principle peaks were identical in the IR spectra of drug and in the IR spectra of the optimized formulation **(F8** and **F13)**. Hence there was no chemical or physical interaction between the drug and the excipients used in this investigation.

6.14.2 Studies on Immediate release layer of Pioglitazone HCl:

Precompressional parameters such as angle of repose, bulk density, tapped density, compressibility index, Hausner's ratio for physical mixtures of immediate release

layer formulations (F1, F2 and F3) were evaluated and results were reported in Table – 6.4.1.

Bulk density and tapped density for the formulations were in the range of 0.589-0.892gm/ml and 0.650-0.946 gm/ml. The angle of repose for the formulations was found to be in the range of 19.5^{o}- 25^{o}. Compressibility index and Hauser's ratio were in the range of 15.00-25.00% and 1.04-1.19. The results obtained confirm that all the formulations exhibited the good flow properties and good packing characteristic.

Studies on *In-vitro* dissolution profile of Immediate release layer formulations:

Based on the results, the cumulative % drug released for F1, F2 and F3 formulations were found to be 78.65±0.71, 92.40±0.36, and 99.69±0.18 respectively. The formulations followed first order kinetics with a regression value of 0.981, 0.990, and 0.916 respectively.

The results depict that, the maximum amount of drug was released from the formulation F3 when compared to other formulations F1and F2. This is due to the more concentration of superdisintegrant in the formulation. Hence, formulation F3 was confirmed as an optimized immediate release layer.

6.14.3 Studies on SR layer of Metformin HCl

Studies on SR layer formulated by Direct Compression technique:

The precompressional parameters ie; Bulk density and tapped density for the formulations were in the range of 0.589-0.892gm/ml and 0.650-0.946 gm/ml. The angle of repose for the formulations was found to be in the range of 19.5^{o}- 25^{o}. Compressibility index and Hausner's ratio were in the range of 15.00-25.00% and 1.04-1.19. Similarly the

post compressional parameters like, weight variation, hardness, friability, and percent drug content were found to be within the limits.

All the formulations (F4 to F7) showed the sudden release of Metformin HCl in the initial hours, which is due to faster dissolution of the highly water soluble drug from the superficial layers of matrix and its diffusion out of the matrix which leads to the entry of dissolution media through the pores. F8 showed sustained drug release up to 12 hours which is nearly identical to the release profile of marketed formulation. F9 showed a release over 12 hours, due to increase in the concentration of PEO-303 in the formulation, resulted in a decreased drug release rate.

A sufficient polymer concentration in the hydrophilic matrix system is required to form a uniform gel barrier around the tablet upon hydration. This barrier is expected to prevent the drug from immediate release into the dissolution medium. If the polymer concentration is low, a complete gel layer may not form resulting in a significant amount of drug being released too quickly in the worst case, tablet disintegration. F4 to F7 failed to sustain the drug release due to in sufficient polymer concentration in the matrix system. It can be seen that polymer concentration of less than 30% are insufficient to produce adequate extended release of metformin HCl. Similar results were reported by Dow for HPMC ER matrices. The *in vitro* dissolution study also shows that an increased PEO level in the formulation resulted in a decreased drug release rate (F9).

A further increase in PEO concentration from 30% to 60% resulted in a slower drug release profile. This effect of slower Metformin HCl release for higher polymer level is due to the longer period of time required to reach the polymer chain disentanglement concentration at the tablet surface, which in turn equates to greater resistance of the matrix to surface erosion.

To ascertain the mechanism of drug release, *in-vitro* release data were fitted into various release kinetic models such as First order, zero order, Higuchi, and Peppas. The first order plots obtained were linear when compared with the zero order plots. Hence the order of release for formulations followed first order kinetics. From the above results F8 formulation was best among other formulations and the release was governed by Fickian diffusion.

Studies on SR layer formulated by Wet Granulation technique:

The Cumulative % drug release for F10, F11, F12, F13 and F14 was 82.68±1.40, 83.04±0.93, 89.51±0.54, 99.21±0.05 and 74.72±1.00. All the formulations (F10 to F12) showed the sudden release of Metformin HCl in the initial hours, which is due to faster dissolution of the highly water soluble drug from the superficial layers of matrix and its diffusion out of the matrix which leads to the entry of dissolution media through the pores. F13 showed sustained drug release up to 12 hours which is nearly identical to the release profile of marketed formulation. F14 showed a release over 12 hours, due to increase in the concentration of Carbopol 971P in the formulation, resulted in a decreased drug release rate.

A sufficient polymer concentration in the hydrophilic matrix system is required to form a uniform gel barrier around the tablet upon hydration. This barrier is expected to prevent the drug from immediate release into the dissolution medium. If the polymer concentration is low, a complete gel layer may not form resulting in a significant amount of drug being released too quickly in the worst case, tablet disintegration. F10 to F12 failed to sustain the drug release due to insufficient polymer concentration in the matrix system. It can be seen that polymer concentration of less than 18% are insufficient to produce adequate extended release of Metformin HCl. The *in vitro* dissolution study also

shows that an increased Carbopol level in the formulation resulted in a decreased drug release rate (F14).

A further increase in Carbopol concentration above 18% resulted in a slower drug release profile. This effect of slower Metformin HCl release for higher polymer level is due to the longer period of time required to reach the polymer chain disentanglement concentration at the tablet surface, which in turn equates to greater resistance of the matrix to surface erosion.

To ascertain the mechanism of drug release, *in-vitro* release data were fitted into various kinetic models such as First order, zero order, Higuchi, and Peppas. The first order plots obtained were linear when compared with the zero order plots. Hence the order of release for formulations followed first order kinetics. From the above results F13 formulation was best among other formulations and the release was governed by Fickian diffusion.

The similarity factor (f_2) was also calculated in order to compare optimized formulation (F8 and F13) with that of the reference formulations. Comparission of the profiles indicated that the formulations (F8and F13) had a profile similar to the reference formulation (f_2 = 51.41and 51.21) respectively. So these two formulations were comparable with the marketed formulation.

7. SUMMARY AND CONCLUSION

In the present investigation, Sustained release Bilayered tablets of Pioglitazone HCl and Metformin HCl were formulated by Direct Compression technique and Wet Granulation technique. Bilayered tablets comprise of IR for sudden onset of action formulated with Crospovidone and SR layer formulated with Polyethylene oxide (PEO-303) and Carbopol 971 P inorder to sustain the drug release.

Drug-excepient compatibility were studied by FT-IR spectral analysis, the results revealed that there were no interactions between drug and excepients in this investigation for the development of the Bilayered tablet formulation.

The Precompressional parameters for IR, SR layer formulations ie; Angle of repose, Bulk density, Tapped density, Compressibility index, Hausner's ratio were studied and found to be in satisfactory limits indicating that the Physical mixtures of the formulations are suitable to formulate the Bilayered tablets. Postcompressional parameters for Bilayered tablets ie; Weight variation, Hardness, Friability, Drug content, were evaluated and the results obtained were satisfactory.

The *in-vitro* drug dissolution studies were carried out for the formulations in pH 1.2 and pH 6.8 phosphate buffer for 2hrs and 10hrs respectively and based on the *in-vitro* drug release profile IR layer formulation (F3) was optimized for the further development of Bilayered tablets. The formulation F8 comprising of PEO-303 and the formulation F13 comprising of CARBOPOL 971P sustained the drug release for a period of 12 hrs. Dissolution profile of formulations F8 and F13 were compared with the dissolution profile of marketed formulation and Similarity factor for the formulations F8 and F13 was found to be 51.41 and 51.21 respectively.

The similarity factor (f_2) was also calculated in order to compare optimized formulation (F8 and F13) with that of the reference formulations. Comparission of the profiles indicated that the formulations (F8and F13) had a profile similar to the reference formulation (f_2 = 51.41and 51.21) respectively. So these two formulations were comparable with the marketed formulation.

The conclusions drawn from the results include:

- Pioglitazone HCl and Metformin HCL and the excepients selected for this investigation were compatible and it was confirmed by FT-IR studies.
- Precompressional and Postcompressional parameters were found to be within the satisfactory limits and hence suitable to formulate Bilayered tablets.
- The order of cumulative % drug release from IR layer formulations was found to be F3>F2>F1.
- The IR layer formulation ie; F3 was optimized because it released the maximum amount of the drug.
- The results of *in-vitro* drug release profile of Bilayered tablets depicts that increase in polymer concentration, increases the retardation of drug release from the SR layer of a Bilayered tablet.
- The desired drug release rate obtained for F8 and F13 was found to be near to that of the theoretical desired drug release rate.
- The desired drug release rate obtained for F8 and F13 was found to be near to that of the drug release rate of Marketed formulation.
- The formulations F8 and F13 were suitable to sustain the drug release for a period of 12hrs, followed first order kinetics exhibited Higuchi's model and Krosmeyer-

Peppas exponential coefficient 'n' < 0.5 indicates that the release was governed by Fickian diffusion.

- Hence can conclude that formulated Bilayered tablets of Pioglitazone HCl and Metformin HCl were developed successfully with IR layer comprising of Crospovidone and SR layer comprising of PEO-303 and CARBOPOL 971P as polymers by Direct Compression technique and Wet Granulation technique.
- From the above results it can be concluded that by using PEO-303 and CARBOPOL 971P we can successfully formulate Bilayer tablets of Pioglitazone HCl and Metformin HCl which showed sustained drug release up to 12hours.

FUTURE WORK TO BE DONE

Based on the above studies the sustained release bilayer matrix tablets of Pioglitazone HCl and Metformin HCl with polymers such as PEO-303 and CARBOPOL 971P could be suitable for sustaining the drug release over a prolonged period. The formulations prepared were found to be linear in releasing the drug for a prolonged period of time i.e. 12 hours. Then these formulations can be further subjected to pharmacodynamic and pharmacokinetic studies in a suitable animal model. Hence the above found formulations may be suitable for once a day administration.

8. BIBLIOGRAPHY

1. Almer LO, Johansson E, Melander A. Influence of sulfonylurea on the secretion, disposal and effect of insulin. Eur J Clin Pharmacol., 1982; 27-32.
2. Altaf AS, Friend DR, MASRx and COSRx. Sustained-Release Technology, Rathbone MJ, Hadgraft J, Robert MS, Modified Release Drug Delivery Technology, Marcell Dekker Inc., New York, 2003.
3. Ansel H.C, Allen L.V, Popovich N.G. Pharmaceutical Dosage Forms and Drug Delivery Systems. Philadelphia, Lippincott Williams and Wilkins., 2003; 230-231.
4. Becker D, Rigassi T, Bauer-Brandl A. Effectiveness of binders in wet granulation: comparison using model formulations of different tablet ability. Drug Dev Ind Pharm., 1997; 791–808.
5. Brogden RN, Heel RC, Pakes GE. Glipizide: A review of its pharmacological properties and therapeutic use of Drugs. 1979; 329-353.
6. Chinam niranjan patra, Arethi bharani kumar, Hemant kumar pandit, Satya prakash singh Meduri vimala devi, “Design and evaluation of sustained release bilayer tablets of propranolol hydrochloride”, Acta Pharm., 2007; 479–489.
7. Chitra karthikeyini.S, Jayaprakash.S, Abirami.A, Mohamed Halith.S, “Formulation and Evaluation of Aceclofenac sodium Bilayer sustained release tablets”, International Journal of ChemTech Research, Oct-Dec 2009; vol-1, No.4, 1381-1385.
8. Choy Fun Wong, Kah Hay Yuen, Kok Khiang Peh, Formulation and evaluation of controlled release Eudragit buccal patches. International Journal of Pharmaceutics. 1999; vol-178, 11–22.

9. Dahl TC, Calderwood T, Bormeth A, et al. Influence of physicochemical properties of hydroxyl propyl methylcellulose on naproxen release from sustained release matrix tablets. J Control Release 1990; 1–10.
10. Dawood bhai S, Rhodes CT. Pharmaceutical and cosmetic uses of talc. Drug Dev Ind Pharm 1990; 2409–2429.
11. Delonca H, Joachim J, Mattha AG. Binding activity of hydroxypropyl cellulose and its effect on the physical characteristics of granules and tablets. Farmaco (Prat) 1977; 157–171.
12. Doddayya Hiremath, Prakash Goudanavar, Mohd Azharuddin, Rajagopal. Udupi.H and Sarfaraz.MD, "Design and Characterization of Bilayer Controlled Release Matrix Tablets of Losartan Potassium", International Journal of Pharmaceutical Research, October-December 2010, vol-2, Issue 4, 34-39.
13. Ene'zian GM. Direct compression of tablets using microcrystalline cellulose, Pharm Acta Helv 1972; 321–363.
14. Ertel KD, Carstensen JT. Chemical, physical, and lubricant properties of magnesium stearate. J Pharm Sci 1988; 625–629.
15. Fonner D.E, Buck J.R., Banker GS. Mathematical optimization techniques in drug product design and process analysis. J. Pharm. Sci. 1970; vol-59, 1587-1596.
16. Gennaro, Remington A.R., the Science and Practice of Pharmacy, 20th Edition. Vol-1. Philadelphia, Lippincott Williams and Wilkins, 2000; 906.
17. Goodman and Gilmans – the pharmacological basis of therapeutics. 11th ed., McGraw Hill medical publishing division. New York; 2006; 971-972.
18. Gwen M. Jantzan, Joseph R. Robinson. Sustained and Controlled release drug delivery systems In: Gilbert S. Banker. Editors. Modern pharmaceutics. 4th

Edition Revised and expanded. Marcel Dekker Inc; New York. USA.2008; 503-530.

19. Heller.J, Washington.D.C, Penhale.D.W.H, Use of bioerodible polymers in self-regulated drug delivery systems, in: P.I. Lee, W.R. Good (Eds.), Controlled Release Technology. Pharmaceutical Applications, Washington DC, ACS Symposium Series. 1997; vol-76, 281–282.
20. Hogan JE. Hydroxy propyl methyl cellulose sustained release technology Drug Dev Ind Pharm. 1989; 975–999.
21. Huikari A, Kristoffersson E. Rheological properties of methylcellulose solutions: general flow properties and effects of added substances. Acta Pharm Fenn 1985; 143–154.
22. Indian Pharmacopeia 2007, Government of India Ministry of Health & Family Welfare, vol-1, 182
23. Jain K.K., Drug Delivery Systems. 1st Edition,Humana Press, 2008, 217.
24. Kibbe AH, ed. Handbook of Pharmaceutical Excipients, 3rd edn. London andWashington, DC: Pharmaceutical Press and American Pharmaceutical Association, 2000; 642–643.
25. Klug ED. Some properties of water-soluble hydroxyalkyl celluloses and their derivatives. J Polym Sci 1971; 36(Part C), 491–508.
26. Laurence L.Brunton, Louis Sanford Goodman and Donald Blumenthal. Goodman and Gilman's Manual of Pharmacology an Theraeutics, McGraw-Hill Professional,2007; 590-594
27. Leon Shargel et; al, "Applied Biopharmaceutics & Pharmacokinetics", Fifth edition, 1941; 813-825.

28. Madhusudan Rao Yamsani et; al, "Factors Influencing the Design and Performance of Oral Sustained/Controlled Release Dosage Forms", International Journal of Pharmaceutical Sciences and Nanotechnology, vol-2, Issue-3, October-December 2009; 583-593.
29. Mitchell K, Ford JL, Armstrong DJ, et al. Influence of drugs on the properties of gels and swelling characteristics of matrices containing methylcellulose or hydroxyl propyl methyl cellulose. Int J Pharm 1993; 165–173.
30. Nagaraju.R, and Rajesh Kaza, "Formulation and Evaluation of Bilayer Sustained Release Tablets of Salbutamol and Theophylline", International Journal of Pharmaceutical Sciences and Nanotechnology, vol-2, Issue 3, October – December 2009; 639 -646.
31. Nicholas G. Lordi. Sustained release dosage forms In: Herbert A. Lieberman. Editors. The theory and practice of industrial pharmacy. 3rd edition (Indian), Varghese publishing house; Mumbai. 430 – 456.
32. Paroni R, Comuzzi B, Arcelloni C and Genovese S, Comparison of Capillary electrophoresis with HPLC for diagnosis of factitious hypoglycemia, Clinical Chemistry, 46, vol-11, 2000; 1773 – 1780.
33. Raghavendra rao.N.G, Ashok Yadav, Upendra Kulkarni, "Formulation and Evaluation of Zero Order Release Glipizide Bilayer Matrix Tablets Using Natural and Synthetic Polymers", International Journal of Current Pharmaceutical Research, vol-2, Issue 1, 2010.
34. Rajan K. Verma, Sanjay Garg, "Development and evaluation of osmotically controlled oral drug delivery system of glipizide" ,European Journal of Pharmaceutics and Biopharmaceutics vol-57, 2004; 513–525

35. Ramesh, D.Sathis kumar, Guruviah and Harani," Formulation and Evaluation of Bilayered sustained release matrix tablets of Metformin HCl and Pioglitazone", American-Eurasian Journal of Scientific Research, 176 – 182.
36. Raymond C Rowe, Paul J Sheskey, Sian C Owen, Handbook of Pharmaceutical Excipients Fifth edition 2006.
37. Raymond C Rowe, Paul J Sheskey, Sian C Owen, Handbook of Pharmaceutical Excipients. 5th ed., Pharmaceutical Press: London; 2006; 111-115, 346-349, 611-616.
38. Robinson JR, Eriksen SP, **"**Theoretical formulation of sustained-release dosage forms. " J Pharm Sci vol-55; 1966; 1254
39. Robinson JR., Lee VHL. Fundamentals of controlled drug delivery: Influence of drug properties and routes of drug administration on the design of sustained and controlled release systems In: Robinson JR. Editors. Controlled drug delivery: fundamentals and applications, 2nd ed. Marcel Dekker Inc; New York: 2005; 38 - 94.
40. Sakr AM, Elsabbagh HM. Evaluation of guar gum as a tablet additive: a preliminary report. Pharm Ind 1977; 399–408.
41. Shah NH, Lazarus JH, Sheth PR, Jarowski CI. Carboxymethylcellulose: effect of degree of polymerization and substitution on tablet disintegration and dissolution. J Pharm Sci 1981; 611–613.
42. Shah V.P, Tsong.Y, Sathe.P, Liu.J, *in vitro* dissolution profile comparison statistics and analysis of the similarity factor, f_2, pharma res.15, 1998; 889-896.
43. Shalin A. Modi**,** P. D. Gaikwad, V. H. Bankar, S. P. Pawar, Sustained Release Drug Delivery System : A Review, International Journal of Pharma Research and Development, vol-2, Issue-12, 2011 February, 147 - 160

44. Shammi Goyal, Jitendra Kumar Rai, R.K.Narang, Rajesh K.S, Sulfonyl Ureas for Antidiabetic therapy, an overview for Glipizide; International Journal of Pharmacy and Pharmaceutical Sciences, vol-2, Suppl 2, 2010; 1 – 6.
45. SkinnerGW, HarcumWW, Barnum PE, Guo JH. The evaluation of fine-particle hydroxyl propyl cellulose as a roller compaction binder in pharmaceutical applications. Drug Dev In Pharm 1999; 1121–1128.
46. The merck index- an encyclopedia of chemicals, drugs and biological. 14th ed., The Merck publishing group: USA 2006; 671.
47. The United States Pharmacopeia. The National Formulary 22, United States Pharmacopeial Convention, Asian Edition, 2004; 867.
48. Vijaya Kumar.B, Prasad.G, Ganesh.B, Swathi.C, Rashmi.A, Amarender Reddy.G, "Development and Evaluation of Guaifenesin Bilayer Tablet", International Journal of Pharmaceutical Sciences and Nanotechnology, vol-3, Issue 3, October - December 2010; 1122-1128.
49. Vinoth Kumar.G, Anand babu.K, Ramasamy.C, "Formulation and Evaluation of Bilayered Tablets of Cefixime trihydrate and Dicloxacillin sodium", International Journal of PharmTech Research, April-June 2011, Vol-3, No.2, 613-618.
50. Virag Szente, Karoly Suvegh, Tamas Marek, Prediction of the stability of polymeric matrix tablets containing Famotidine from the positron annihilation lifetime distributions of their physical mixtures. Journal of Pharmaceutical and Biomedical Analysis 2009; vol-49, 711–714.
51. Vyas.SP and Roop. K. Khar, Controlled Drug Delivery Concepts and Advances, First edition 2002, New Delhi, 196-217.
52. Wahlin-Boll E, Almer LO, Melander A. Bioavailability, pharmacokinetics and effects of glipizide in type II diabetics. Clin Pharmacokinetics 1982; 363.

53. Weiner M, Bernstein IL. Adverse Reactions to Drug Formulation Agents: A Handbook of Excipients. New York: Marcel Dekker. 1989; 93–94.
54. Welling P.G, Dobrinska M.R., Design considerations and bioavailability assessment of controlled drug delivery systems. In, Robinson JR, Lee VHL, Controlled Drug Delivery- Fundamentals and Applications, 2nd Edition, New York, Marcel Dekker Inc, 1987; 5-7.
55. Yie W. Chien. Oral Drug Delivery and Delivery Systems In: Yie W. Chien. Editors. Novel Drug Delivery Systems, 2nd ed. Marcel Dekker Inc; New York. USA.2007; 139 – 196.