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Formulation and Evaluation of Immediate Release Pregabalin Tablets

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Dedication

I am deeply thankful to my family for the support and the encouragement they surrounded me with.

I am deeply grateful and thankful to my wife and children who inspired, supported and encouraged me to explore the best in me. I thank them for dedication and patience.

Declaration

I certify that this thesis submitted for the degree of master is the result of my own research, except where otherwise acknowledged, and this thesis has not been submitted for the higher degree to any other university or institute**.**

Signed: ……..

Mohammad Sameeh Shafiq Shana'a

Date:

Acknowledgment

I have no words to express my deepest gratitude to Almighty, compassionate, and supreme Allah, who enabled me to accomplish this work. I also invoke peace for the last prophet of Allah, Muhammad (SAAW), who is forever a torch of guidance for humanity as a whole.

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I would like to express my sincere thanks to Jerusalem Pharmaceuticals Company for their unlimited support in donating materials, providing equipment, laboratories and assistance. My special thanks to Hani Roumyh, Salah Shawar from the research and development department in Jerusalem Pharmaceutical Company for their assistance and help in the experimental work.

Finally, I take great privilege to express my heartfelt thanks to all the people who have been involved directly or indirectly with the successful completion of this work.

Abstract

Pregabalin is an anticonvulsant agent used for the peripheral and central neuropathic pain treatment, marketed as Capsule and Solution dosage forms. The purpose of this study is to develop an immediate release Pregabalin Tablet, as a new dosage form in the Palestinian market that is bioequivalent to the FDA approved Reference Pregabalin Capsules (Lyrica).

Pregabalin drug substance and the finished product during the time of development were not described in either the British or US pharmacopoeia.

The chemical and physical evaluation of bulk drug substance (Pregabalin) was accomplished following the manufacturer`s analytical method and specification. A new reversed-phase, isocratic LC method was developed and validated according to USP validation parameters, for the qualitative and quantitative determination of Pregabalin in pharmaceutical dosage forms using HPLC (LaChrome Elite) equipped with photodiode array UV detector. Mobile Phase is a mixture of Phosphate Buffer pH 6.9, and acetonitrile (94:6).

Chromatographic System is Column: C-18 ODS 5 to 10µm in diameter (25cm X 4.6 mm id), detector: UV set at wavelength 210 nm, Flowrate: 1.5 ml / min and Injection Volume: 20 µL.

Formula development was accomplished through a series of steps:

Selection of excipients through compatibility studies, selection of manufacturing process and in-vitro comparison studies versus the reference product.

Excipients compatibilities were studied by preparing a binary mixture of Pregabalin and excipient (1:1) then sealed in neutral glass vials and incubated at 40±2 °C/75 ±5% RH for 30 days. The mixtures were tested by means of FTIR for any possible interactions. The following excipients (Microcrystalline Cellulose, Pregelatinized Starch, Talc and Mg Stearate) were found to be compatible with Pregabalin.

A series of pre formulation trials were composed and processed by direct compression method. Selection of formula was based on:

Powder characteristics: (such as compressibility ratio, flowability, bulk density and tapped density), which would allow for a simplified method of manufacture, friability, hardness and disintegration of compressed tablets.

The selected formulation was applied to prepare Pregabalin Tablets in two strengths, i.e. tablets containing 75mg/tablet and 300mg/tablet. They are prepared as dose weight proportional. The compressed tablets were film-coated with PVA based polymer by using water as solvent.

The stability of film coated Pregabalin tablets were tested by incubating the finished products in their final package (PVC-Alum) at different storage conditions i.e. 25 ± 2 °C / 60 % ± 5% RH, 30 C ± 2 °C / 60% ± 5% RH and 40 C ± 2 °C / 75% ± 5% RH. Samples were tested biweekly for content of Pregabalin (assay), physical appearance, dissolution rate, and degradation products.

Pregabalin Tablets proved to be stable in all aspects for the period tested (1 month) at all storage conditions.

Biowavier study was performed between Pregabalin Tablets (75mg and 300mg) and the Reference Pregabalin Capsules (Lyrica 75 and 300mg Capsules) using paddle method rotated at 50 rpm in different dissolution media (0.06N Hydrochloric acid solution, Acetate buffer pH 4.5 and phosphate buffer pH 6.8. It was found that either Reference or Test products release more than 85% of their Pregabalin content in 15 minutes. As Pregabalin API, according to BCS is Class I drug and the dissolution profiles of Pregabalin Tablets is similar to that of Reference Pregabalin Capsules (Lyrica) under the same test conditions, and all excipients used are not suspect of having any relevant impact on bioavailability it is strongly believed that the developed Pregabalin Tablets are bioequivalent to the marketed Lyrica Capsules.

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Part I *Introduction*

Introduction

1.1 Background information on Pregabalin

Ref: {(1). (1) (2)}

1.1.1 Description

- Pregabalin is a white to off-white, crystalline solid powder and has Structural formula as shown in the next figure.

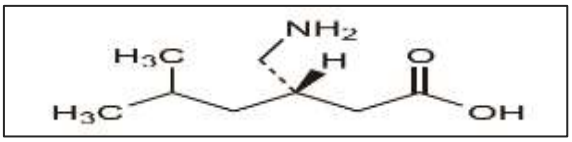

Figure 1: Structure formula of Pregabalin

- Chemical name: (*S*)-3-(aminomethyl)-5-methylhexanoic acid.
- The molecular formula is $C_8H_{17}NO_2$

Relative molecular mass is 159.23. CAS: 148553-50-8

1.1.2 General properties:

- **Solubility**: Sparingly soluble in water
- **Melting point (DSC)**: About 196.67°C
- **Optical rotation[α]** 23d (c = 1.06 in water): +10.0° to +12.0°
- **Isomerism:** Pregabalin contains one Chiral center in its structure and exist as S(+) and R(-) isomers.
- **Dissociation constants:** pKa1 of 4.2 for carboxylic acid and pKa2 of 10.6 for amine moiety
- **Experimental Log P**: 1.3, the log of the partition coefficient (n-octanol/0.05M phosphate buffer) at pH 7.4 is -1.35.

1.1.3 Impurities: **[Ref Alembic DMF]**

- **R-Isomer:** NMT 0.15%
- **Impurity-III:** ((R)-(-)-3-(Carbamoylmethyl)-5-methyl hexanoic acid (R-CMH): NMT 0.15% w/w.
- **Impurity-IV**: 2-Pyrrolidinone, 4-(2-methylpropyl): NMT 0.15% w/w).
- **Any other impurity:** NMT 0.10% w/w.
- **Total impurities:** NMT 0.50% w/w.

1.1.4 Pharmacokinetics

Pregabalin steady-state pharmacokinetics is similar in healthy volunteers, patients with epilepsy receiving anti-epileptic drugs and patients with chronic pain.

1.1.5 Absorption

Pregabalin is rapidly absorbed when administered in the fasted state, with peak plasma concentrations occurring within 1 hour following both single and multiple dose administration. Pregabalin oral bioavailability is estimated to be ≥ 90% and is independent of dose. Following repeated administration, steady state is achieved within 24 to 48 hours. The rate of pregabalin absorption is decreased when given with food resulting in a decrease in C_{max} by approximately 25-30% and a delay in T_{max} to approximately 2.5 hours. However, administration of pregabalin with food has no clinically significant effect on the extent of pregabalin bioavailability.

Pregabalin is well absorbed after oral administration, is eliminated largely by renal excretion, and has an elimination half-life of about 6 hours.

1.1.6 Distribution

In preclinical studies, Pregabalin has been shown to readily cross the blood brain barrier in mice, rats, and monkeys. Pregabalin has been shown to cross the placenta in rats and is present in the milk of lactating rats. In humans, the apparent volume of distribution of Pregabalin following oral administration is approximately 0.56 L/kg. Pregabalin is not bound to plasma proteins. At clinical doses of 150 to 600 mg/day, the average steady-state plasma Pregabalin concentrations were approximately 1.5 and 6.0 µg/mL, respectively.

1.1.7 Indications and Clinical uses

- Pregabalin is indicated for the treatment of neuropathic pain in adults.
- Pregabalin is indicated as adjunctive therapy in adults with partial seizures with or without secondary generalization.

Pregabalin is available in the market as immediate release Capsules and solution dosage forms. Pregabalin (LYRICA Capsules) are administered orally and are supplied as imprinted hard-shell Capsules containing 25, 50, 75, 100, 150, 200, 225, and 300 mg of Pregabalin and packaged in either HDPE bottles or PVC- Aluminum blisters, while Lyrica Oral solution contains 20mg/ml of Pregabalin.

1.2 Tablet: properties and manufacturing Techniques

1.2.1 Tablet properties

A tablet is a solid pharmaceutical dosage form that comprises a mixture of active substances and excipients, usually in powder form, pressed or compacted from a powder into a solid dose.

API has to be in pure form otherwise impurities can catalyze series of chemical reactions. The API should be stable against photolysis, oxidation, hydrolysis, etc. to keep the formulation a simple one. Sensitive particles require careful handling during manufacturing.

The excipients can include:

1. Diluents, binders or granulating agents, glidants (flow aids) and lubricants to ensure efficient tableting.
2. Disintegrants to promote tablet break-up in the digestive tract.
3. Sweeteners or flavors to enhance taste in chewable tablets.
4. Colors and pigments to make the tablets visually attractive.
5. A polymer coating is often applied to make the tablet smoother and easier to swallow, to control the release rate of the active ingredient, to make it more resistant to the environment (extending its shelf life), or to enhance the tablet's appearance.

The compressed tablet is the most popular dosage form in use today. About two-thirds of all prescriptions are dispensed as solid dosage forms, and half of these are compressed tablets. A tablet can be formulated to deliver an accurate dosage to a specific site; it is usually taken orally, but can be administered sublingually, buccally, rectally or intra-vaginally.

Tablet Requirements:

6. Should be an elegant product having its own identity while being free of defects such as chips, cracks, discoloration, contamination, and the like.
7. Should have the strength to withstand the rigors of mechanical socks encountered in its production, packaging, shipment, and dispensing
8. Should have the chemical and physical stability to maintain its physical attributes over time.
9. Must be able to release its active components in the body in a predictable and reproducible manner.

1.2.2 Tablet Manufacturing Techniques

The manufacture of tablet dosage forms is a complex multi-stage process under which the starting materials change their physical characteristics a number of times before the final dosage form is produced.

The main three manufacturing approaches are illustrated in the following flow charts (Figure 2):

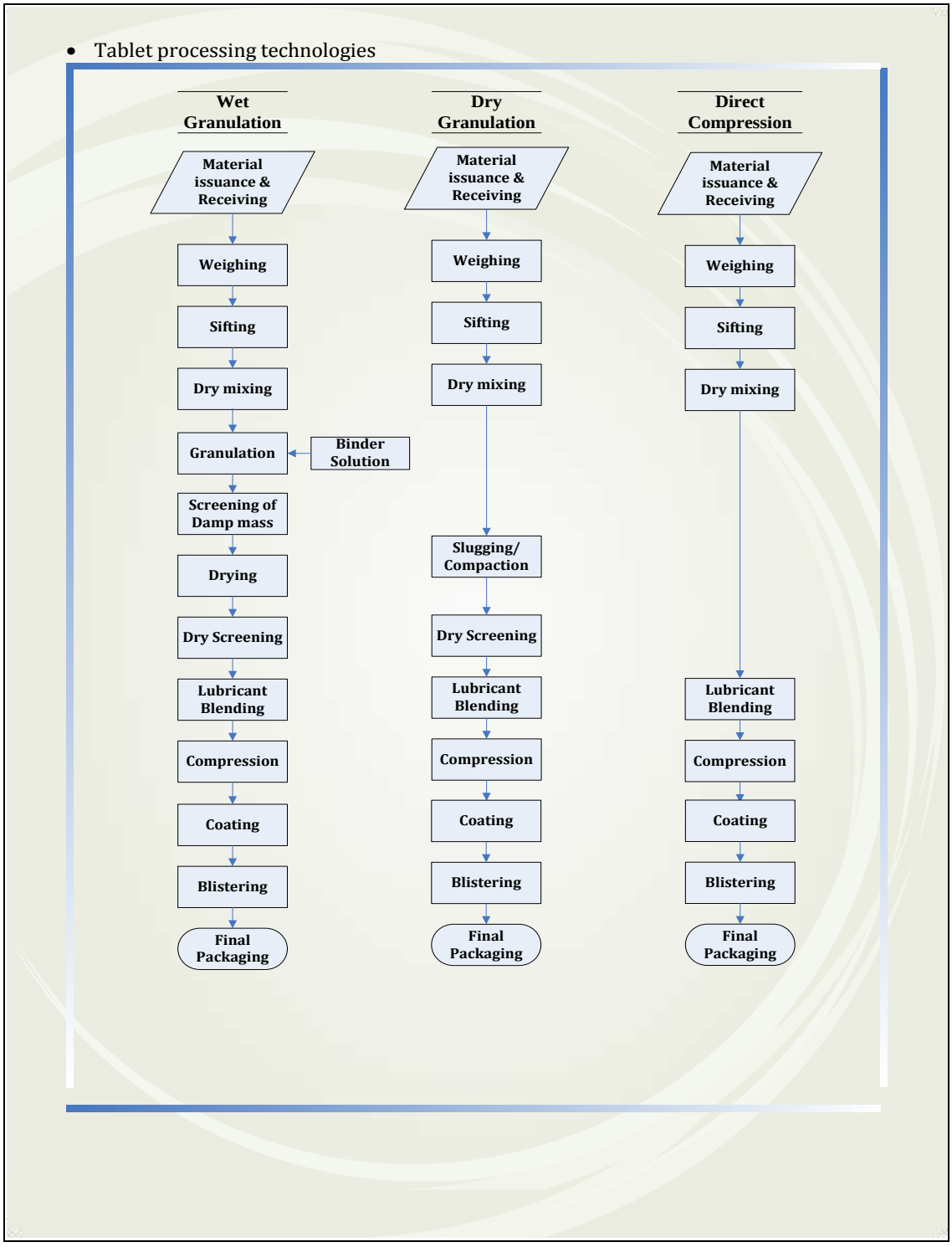

Figure 2: Manufacturing process flowchart

A. *Direct compression*

Direct compression consists of compressing tablets directly from powdered materials without modifying physical nature of materials. This method is applicable for crystalline chemicals having good compressible characteristic and flow properties.

If the bulk powder blend's properties do not suit direct compression Tableting, manufacturers will turn to granulation processes to create the desired flowability and low dust ability. These characteristics are required to minimize tablet weight variations and ensure high density for high tablet filling weight and high mold ability for hard tablet manufacture.

B. *Dry granulation*

Dry granulation method is defined as the formation of granules by slugging or compaction, if the Tableting ingredients are sensitive to moisture and/or unable to withstand elevated temperature during drying.

C. *Wet granulation*

Wet granulation forms the granules by binding the powders together with an adhesive, instead of by slugging or compaction.

1.3 Raw Materials (Excipients)

Excipients are the components of a finished drug product other than the active pharmaceutical ingredient (API) and are added during formulation for a specific purpose.

Criteria for Excipients selection

1. They should be nontoxic and acceptable to the regulatory agencies in all countries where the product is to be marketed.
2. They must be commercially available in an acceptable grade in all countries where the product is to be manufactured.
3. Their cost must be acceptably low.
4. They must not be contraindicated by themselves or because of a component (e.g., Sodium) in any segment of the population.
5. They must be physiologically inert.
6. They must be physically and chemically stable by themselves and in combination with the drug(s) and other tablet components.
7. They must be free of any unacceptable microbiologic “load”.
8. They must be color- compatible (not produce any off-color appearance) .
9. If the drug product is also classified as a food, the excipients must be approved direct food additives.
10. They must have no deleterious effect on the bioavailability of the drug(s) in the product.

Types of Excipients [Ref. (3)]

A. **Diluents (Or Fillers):** Diluents increase the volume to a formulation to prepare tablets of the desired size. Widely used fillers are lactose, microcrystalline cellulose (Avicel PH® from FMC Corp. and Emococel® from Mendell), starch and pregelatinized starch; the filler is selected based on various factors, such as the cost, and compatibility with other formulation ingredients.

B. **Binders (Or Adhesives):** Binders promote the adhesion of particles of the formulation. Such adhesion enables preparation of granules and maintains the integrity of the final tablet. Starch and pregelatinized starch are commonly used as binding agents.

C. **Lubricants:** Lubricant is a substance capable of reducing or preventing friction, heat, and wear when introduced as a film between solid surfaces. It works by coating on the surface of particles, and thus preventing adhesion of the tablet material to the dies and punches. Magnesium Stearate is one example of a lubricant.

   In addition lubricants play other roles in the preparation of tablets such as:

   - Improve the flow of granules in the hopper to the die cavity.
   - Reduce the friction between the tablet and the die wall during the tablet's ejection from the tablet machine.
   - Give sheen to the finished tablets.

D. **Glidant:** Substance that allows particles moving smoothly and continuously. Glidant works by removing moisture and as a result enhancing flow. Talc has both lubricant and glidant effects.

E. **Disintegrators (Or Disintegrating Agents):** To rupture or breakup of tablets, disintegrating agents must swell or expand on exposure to aqueous solution. Thus, the most effective disintegrating agents in most tablet systems are those with the highest water uptake property. In general, the more hydrophilic, the better disintegrating agents are therefore highly hydrophilic, e.g. Microcrystalline cellulose.

   Microcrystalline cellulose has various functions in direct compression. It can be used as a binder, disintegrant, and filler. Microcrystalline cellulose allows direct compression of tablets and wet granulation processes.

F. Coating Material:

[Ref. (4)]

The coating is being applied to a dosage form for a purpose ranging from the esthetic to a desire to control the Absorption of the drugs. The major components of film coating formulation consist of a polymer, plasticizer, colorant and solvent, Cellulose ether such as Hydroxypropyl methylcellulose (HPMC) and polyvinyl alcohol (PVA) are often the preferred polymers in film coating.

The major solvent used in film coating typically alcohols and purified water.

1.4 Packaging Material

[Ref. (5) (6) (7)]

Packaging process is defined as the collection of different components which surround the pharmaceutical product from the time of production until its use.

FDA definition "A container closure system refers to the sum of packaging components that together contain and protect the dosage form. This includes primary packaging components and secondary packaging components, if the latter are intended to provide additional protection to the drug product. A packaging system is equivalent to a container closure system".

A primary packaging component means a packaging component that is or may be in direct contact with the dosage form. A secondary packaging component means a packaging component that is not and will not be in direct contact with the dosage form.

Figure 3: Blister packaging for Pregabalin tablets after blistering stage

1.4.1 Selection of packaging material dependent on several parameters:

- Moisture barrier requirements.
- Light barrier requirements.
- Gas barrier requirements.
- Chemical properties.

1.4.2 Importance of packaging

Blister packaging is becoming more accepted for the manufacturers and consumers recognize its benefits. Blister packs can help patients follow drug regimens, protect drugs over a long shelf life, and are portable. Many aspects in blister packaging is better than conventional packaging.

a. Product integrity

Blister packaging helps retain product integrity because drugs that are prepackaged in blisters are shielded from adverse conditions. Furthermore, opportunities for product contamination are minimal, and each dose is identified by product name, lot number, and expiration date. Therefore, blister packaging ensures product integrity from the producer directly through distribution to the consumer.

b. Product protection

Blister packaging, however, keeps each tablet or capsule hermetically sealed in its own bubble. Drugs that are not taken remain in the original package and are fully protected against external conditions. A blister protects a moisture sensitive tablet right up to administration. In contrast, the moisture in the headspace of a multiple-unit bottle is replaced each time the bottle is opened.

Table 1 illustrates the different polymers used in packaging and their properties.

Table 1: Comparison of forming films

[Ref. (8)]

Type and Thickness of Forming Film	**WVTR** (g.mil/100in^2/day) @ 100°F and 90% RH	**OTR** (cc.mil/100in^2/day) @ 77°F
Cold form foil	0.00	0.00
Polycholorotrifluror ethylene	0.016	7.0
Polyvinyl dichloride (PVDC)	0.1-0.2*	0.15-0.9
High density polyethylene	0.3-0.4	139-150
Polypropylene (PP)	0.69-1.0	182
Low density polyethylene (LDPE)	1.2-2.0	3-5
Poly ethylene terephthalate (PET)	1.2-2.0	3-5
Poly vinyl chloride	0.9-5.1	5-20
Ethylene vinyl alcohol (EVOH)	1.4-5.4*	0.05-0.90
Polystyrene (PS)	7-10	350-400
Nylon	16-20	1.0
*** As measured on the unformed film at an ambient temperature of 104°F and 90% RH**		

c. Tamper evidence

Tamper evidence is strength of blister packaging. The dosage units are individually sealed in constructions of plastic, foil, and/or paper. The package must be designed so that one must tear the compartment to get at the product, and it must not be possible to

separate the backing materials from the blister without leaving evidence. Once a bottle has been opened, whatever tamper-evident mechanism it had is gone. With blister packaging, however, each tablet or capsule is individually protected from tampering until use, so any form of tampering with a blister package is immediately visible.

d. Other advantage as:

- Reduced possibility of accidental misuse as Child resistance.
- Patient compliance and Carry the correct information and identification of the product.

1.4.3 Blister packaging components

The four basic components of pharmaceutical blister packages as illustrated in Figure 4.

- The forming film.
- The lidding material.
- The heat-seal coating.
- The printing ink.

Forming films account for approximately 80–85% of the blister package, and lidding materials make up 15–20% of the total weight of the package. Because the forming film and the lidding material form an integrated package, they must match precisely.

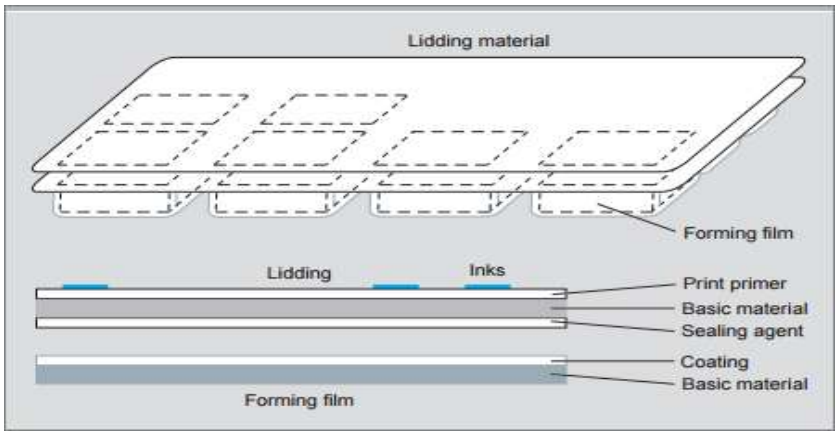

Figure 4: Basic components of blister packaging.

1.4.3.1 Forming film

The forming film is the packaging component that receives the product in deep drawn pockets. One key to package success is selecting the right plastic film for the blisters in terms of its property type, grade, and thickness. Consideration must be given to the height and weight of the product, sharp or pointed edges of the final package, and the impact resistance, aging, migration, and cost of the film. The plastic also must be compatible with the product. Factors influencing package production and speed of assembly must be taken into account, including heat sealing properties and the ease of cutting and trimming formed blisters.

Plastic forming films such as PVC, polypropylene (PP), and polyester (PET) can be thermoformed, but support materials containing aluminum are cold-formed. The forming film usually is colorless and transparent, but it can be obscured for use in child resistant packages or to protect light sensitive drugs. The forming web for blister packs nearly always is PVC, sometimes coated or laminated with additional components that enhance the oxygen and water-vapor barrier.

Types of forming films

PVC forming film is called rigid PVC because it is almost free of softening agents. Rigid PVC is a very clear, stiff material with a low WVTR. It exhibits excellent thermoform ability; a high flexural strength; good chemical resistance; low permeability to oils, fats, and flavoring ingredients; easy tintability; and low cost. These properties make rigid PVC the material of choice for blister packaging, and it essentially has 100% of the market for the plastic component. PVC films that are thermoformed have a thickness of about 10 mil.

The use of PVC has attracted much criticism because its combustion produces hydrochloride emissions, where many pharmaceutical companies now stipulate that any new blister machines must be capable of handling both PVC and PP.

a. **Polyvinylidene chloride (PVDC)–coated PVC**

   Although its volume in drug packaging is small, PVDC plays a critical role in blister packaging as laminations or coatings on PVC. PVDC is the most common coating in blister packaging because it can reduce the gas and moisture permeability of PVC blister packages by a factor of 5–10. Coated PVC films have a thickness of 8–10 mil; the thickness of the PVDC coat amounts to 1–2 mil. The coating is applied on one side and usually faces the product and the lidding material.

b. PVC/Chlorotrifluoroethylene (CTFE)

Films made from PVC and CTFE have the lowest water-vapor permeability of all films used for blister packaging. When compared with the water-vapor permeability of 10-mil PVC, the permeability of 8-mil PVC/0.76- mil CTFE is lower by a factor of 15. However, the environmental concerns regarding PVC also apply to PVC/CTFE films.

c. Polypropylene (PP)

There is an increasing trend toward using PP as a support material for blister packages. The water-vapor permeability of uncoated PP is lower than that of PVC and is comparable to that of PVDC-coated PVC. The thickness of PP films used in the thermoforming process ranges from 10 to 12 mil. Advantages of PP include easy recyclability, no release of toxins during incineration, and good moisture-barrier properties. PP is a possible replacement for PVC, especially in Europe. However, the use of PP has its drawbacks. One problem is thermoforming. The temperatures required for thermoforming PP and for the subsequent cooling process must be controlled precisely. Warping also can occur, Other difficulties associated with the use of PP include its thermal instability, higher rigidity than PVC, and susceptibility to post-processing shrinkage. In addition, PP is difficult to run on a standard blister machine and cannot be processed as fast as PVC. If a company runs PP and needs new equipment, it must go through a precise validation process, performing various tests on PP to satisfy FDA requirements.

d. Polyethylene terephthalate (PET)

PET is another material that may replace PVC, but its relatively high water-vapor permeability compared with that of PVC will prevent its universal use. PVDC coated PET could have the same water vapor barrier effect as PVC, but this does not appear to be promising in view of the larger goal to replace chlorous plastics with PET.

e. Polystyrene (PS)

Polystyrene (PS) is perfectly compatible with thermoforming, but its high water vapor permeability makes it unsuitable as a blister material for pharmaceutical purposes.

f. Oriented polyamide (OPA)/aluminum/PVC or nylon/aluminum/PVC

OPA/aluminum/PVC laminates are intriguing. With a laminate structure consisting of 1-mil OPA, 1.8- mil aluminum, and 2.4-mil PVC it is possible to eliminate water-vapor permeability almost entirely.

1.4.3.2 Lidding materials

The lidding material provides the base or main structural component upon which the final blister package is built. It must be selected according to the size, shape, and weight of the product as well as the style of the package to be produced. Lidding materials range in caliper or thickness from 0.36 to 0.76 mm, but 0.46–0.61 mm is the most popular range. The surface of the lidding material must be compatible with the heat-seal coating process. Clay coatings are added to the lidding material to enhance printing. Heat-sealing and printability are both important considerations in blister packaging, and the lidding material must offer the best workable compromise.

- **Characteristics**

  The lidding material can be clear plastic, but in pharmaceutical packaging it is either plain or printed 1- mil foil (for push-through blister types) or paper/foil or paper/PET/foil laminations (for child-resistant peel–push types). The lidding material must guarantee a WVTR that is at least as low as that of the forming films, and it must be suitable for the type of opening appropriate to the package (e.g., push-through or peel-off). Figure 5 shows a cross-section of a peel off–push through lidding material.

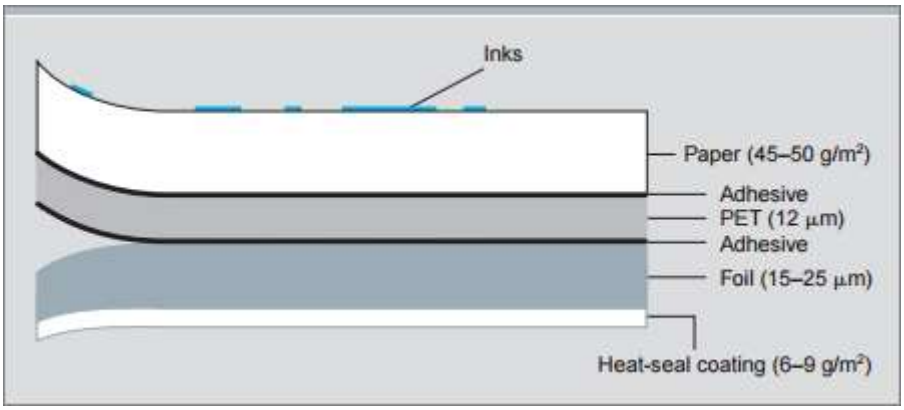

Figure 5: Cross section of a peel off–push through lidding material.

Table 2: Different types of lidding materials

Type and Thickness of Lidding material (mil)	Weight (g/m2)
0.8-mil Aluminum, hard, push-through	60
0.8-mil Aluminum, hard, heat seal–coated, side-printed, push-through	61
1-mil Aluminum, soft, child resistant	76
45 g(m22)/1-mil Paper/aluminum, peel-off	171
45 g(m22)/0.48-mil Paper/PET/aluminum, peel off–push through	142

- **Types of lidding materials**

  Hard aluminum is the most widely used push-through lidding material in Europe. The foil usually has a thickness of 0.8ml, other type of lidding material Soft aluminum (1 ml) frequently is used for child-resistant push-through foils. With the exception of the type of aluminum used, the structure of this lidding material corresponds to that of hard aluminum (0.8 ml). The softness and thickness of this type of aluminum help prevent children from pushing tablets through it.

  And there is Paper/aluminum In combinations of paper and aluminum, and there is Paper/PET/aluminum. Lidding material made of a paper/PET/aluminum laminate is often called peel off–push through foil.

1.4.3.3 Printing inks

Printing inks provide graphics and aesthetic appeal. They can be applied to the lidding material by letterpress, gravure, offset, flexographic, or silk-screen printing processes. Printing inks must resist heat sealing temperatures as high as 300°C without showing any discoloration or tackiness (blocking). In addition, they must sufficiently resist abrasion, bending, and fading and must be safe for use with the intended product. Printing inks should not contain excessive amounts of hydrocarbon lubricants, greases, oils, or release agents. Qualification tests should always precede production runs. Finally, printing inks must comply with FDA recommendations.

1.5 Tablets Quality

[Ref. (9)]

ICH Guidance Q6A and USP General Chapter 2, have a recommendation and specification to ensure that tablets are safe and effective at the time of release and over their shelf life.

Drug product quality test for oral drug product fall into two categories: universal tests that are applicable to all oral drug products and specific tests that should be considered for inclusion for specific types of oral products.

Tests that are universally applied to ensure safety, efficacy and quality include description, identification, assay-strength, and impurities (organic, inorganic and residual solvents).

Specific tests for tablets include disintegration, friability, breaking force-hardness, uniformity of dosage unit and dissolution.

1.5.1 Description

It is general in nature and not standard in itself. It communicates the general appearance (color, shape, diameter, thickness and surface) of the formulated tablets that are inspected and tested visually, while the dimensions were measured by using a suitable caliper.

1.5.2 Identification:

It is an aid to confirm that the drug product contains the labeled drug substance by providing a positive identification of the drug substances in the drug produced.

1.5.3 Assay

It is a specific and stability-indicating test to determine the content (potency) of the drug product. In most cases a prior acceptable of ±10% variation in limits of assay from the target (label claim 100%) is accepted Acceptance criteria of 95.0%-105.0% are used for drug product with narrow therapeutic index.

1.5.4 Impurities:

During product manufacture and over the shelf-life of the product, degradation product can form, as a result of degradation of the drug substance and excipients.

The procedure and acceptance criteria should specifically limit toxic material.

1.5.5 Hardness

It is the force required to cause the tablet break in a specific plane, tablet hardness or breaking force is measured by placing the tablet between two platens of the hardness tester, one of which moves to apply sufficient force to the tablet to cause fracture.

Figure 6: Hardness Tester

1.5.6 Friability [Ref. (10)]:

It is generally run once and applicable to most compressed non coated tablet. It is percentage weight loss after testing the tablet in a drum rotated 100 times.

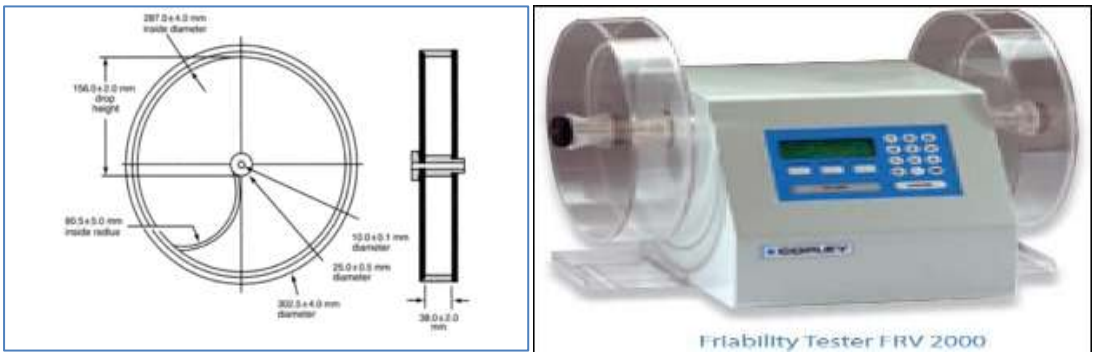

Figure 7: Friability Tester

Acceptance Criteria: maximum mean weight loss from the three samples of not more than1.0% is considered acceptable for most products.

1.5.7 Disintegration Test [ref. (11)]:

To test for disintegration time, one tablet is placed in each tube and the basket rack is positioned in a 1-L beaker of water at 37 ± 2.0 C such that the tablet remain 2.5 cm below the surface of liquid on their upward movement and not closer than 2.5 cm from the bottom of the beaker in their downward movement. Move the basket containing the tablets up and down through a distance of 5-6 cm at a frequency of 28 to 32 cycles per minute. According to the test the tablet must disintegrate and all particles must pass through the 10 mesh screen in the time specified. If any residue remains, it must have a soft mass.

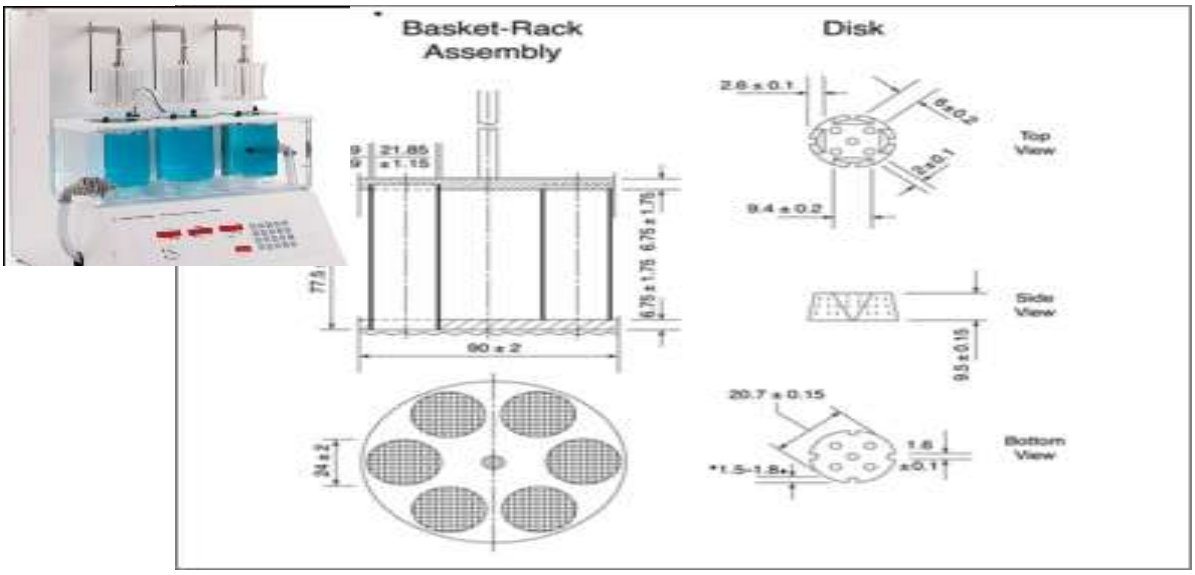

Figure 8: Disintegration apparatus. (All dimensions are expressed in mm.)

1.5.8 The Uniformity of dosage Unit [Ref. (12)]

The term "uniformity of dosage unit" is defined as the degree of uniformity in the amount of the drug substance among dosage units. Each unit in a batch should have a drug substance content within a narrow range around the label claim (consistency of dosage units).

The uniformity of dosage units can be demonstrated by unit-dose either of two methods, Content Uniformity or Weight Variation and that depend on the weight of active ingredient (Dose) and the percent of label claim (ratio of drug Substance) according to the table 3:

Table 3: Application of Content Uniformity (CU) and Weight Variation (WV) Tests for Dosage Forms

Dosage Form	Type	Subtype	Dose & Ratio of Drug Substance	
			≥25 mg and ≥25%	<25 mg or <25%
Tablets	Uncoated		WV	CU
	Coated	Film	WV	CU
		Others	CU	CU

a. In case of Weight Variation

The assay for the drug substance(s) on a representative sample of the batch using an appropriate analytical method is carried out. This value is expressed as percent of label claim. Assuming that the concentration (weight of drug substance per weight of dosage unit) is uniform, not fewer than 30 dosage units, For uncoated or Film Coated Tablets are selected, and 10 units are accurately weighted individually and the content, expressed as % of label claim, of each tablet from the weight of the individual table is calculated and the acceptance value from the result of the Assay is calculated According to USP, Chapter <905>.

b. In Case of Content Uniformity

Select not fewer than 30 units, and assay 10 units individually using an appropriate analytical method, calculate the acceptance value, the content uniformity procedures for all different dosage forms are found in the USP Chapter <905>.

1.5.9 Dissolution for routine testing:

[Ref (13)]

The pharmaceutical scientist would like to find a relationship between an in vitro characteristic of a dosage form, and it's in vivo performance.

Disintegration was originally thought to be this characteristic. The USP introduced its disintegration test in 1950.

With advances in methodology, the disintegration test was found to be too insensitive, and dissolution test methods were introduced in the USP in 1968.

Dissolution is principally useful as a QC test. It can be predictive of in vivo behavior.

1.6 Compatibility Study

[Ref (14)**]**

The drug-excipient compatibility studies are the first step for dosage form development in the pre-formulation stages of the development of a dosage form. The potential physical and chemical interactions between drugs and excipients can affect the chemical, physical and therapeutically properties and stability of the dosage form.

Physical interactions are common in dosage form and also difficult to detect. Physical interactions may or may not involve chemical changes, but may change dissolution, solubility, adsorption, solid dispersion complexation and other properties.

Chemical Interactions involve the reaction of API's and excipients to form unstable compounds that have a deleterious effect on the formulation.

Biopharmaceutical interaction are observed after administration of the medication, Some excipients may interact in physiological way when they are administration along with API's causing changes such as premature breakdown of enteric coat, increase in gastrointestinal motility.

Methods of estimation of drug excipient compatibility include thermal such as DSC, TEA, DTA and hot stage microscopy) and non-thermal analytical techniques as Vibrational spectroscopy, Powder X-ray diffraction and solid state NMR.

The samples for binary Mix Compatibility Testing are prepared by triturating API with the individual excipient as 1:1 powder mixes, stored under accelerated conditions as powder mixes or slurries in water; then analyzed by stability-indicating methodology, e.g. HPLC, FTIR.

Figure 9: FTIR Spectrophotometry used in analysis compatibility study samples

1.7 Bioequivalence & Biowaiver Study

[Ref. (15), (16) (17)]

1.7.1 Background

In 1995 the American Department of Health and Human Services, US Food and Drug Administration (HHS-FDA) instigated the Bio-pharmaceutics Classification System (BCS), with the aim of granting so-called biowaivers for scale-up and post-approval changes (SUPAC). (www.fda.gov/cder/ guidance/cmc5.pdf)

A Biowaiver means that in vivo bioavailability and/or bioequivalence studies may be waived (i.e. not considered necessary for product approval). Instead of conducting expensive and time-consuming in vivo studies, a dissolution test could be adopted as the surrogate basis for the decision as to whether two pharmaceutical products are equivalent. At that time the biowaiver was only considered for SUPAC to pharmaceutical products.

More recently, the application of the biowaiver concept has been extended to approval of certain orally administered generic products.

(www.fda.gov/cder/guidance/3618fnl.htm)

Only APIs with high solubility and high permeability and which are formulated in solid, immediate release (IR) oral formulations can be approved on the basis of the biowaiver procedure. A major advantage of the biowaiver procedure is the simplification of the product approval process and the reduction of the time required, thus reducing the cost of bringing new products to market.

1.7.2 Biowaiver Study and Biopharmaceutics Classification System (BCS)

The BCS is a scientific framework for classifying drug substances based on their aqueous solubility and intestinal permeability. When combined with the dissolution of the drug product, the BCS takes into account three major factors that govern the rate and extent of drug absorption from IR solid oral dosage forms: (1) dissolution, (2) solubility, and (3) intestinal permeability.

According to the BCS, drug substances are classified as follows:

Class 1: High Solubility – High Permeability

Class 2: Low Solubility – High Permeability

Class 3: High Solubility – Low Permeability

Class 4: Low Solubility – Low Permeability

Depending on the classification, the oral availability of the API may be expected to range from being heavily dependent on the formulation and manufacturing method (e.g. Class II

APIs: poorly soluble yet highly permeable) to being mostly dependent on the API permeability properties (e.g. Class III APIs: highly soluble yet poorly permeable).

The next diagrams depicting the products eligible for the biowaiver procedure under the HHS-FDA guidance and those eligible according to the WHO are presented in Fig. 10 and Fig. 11, respectively.

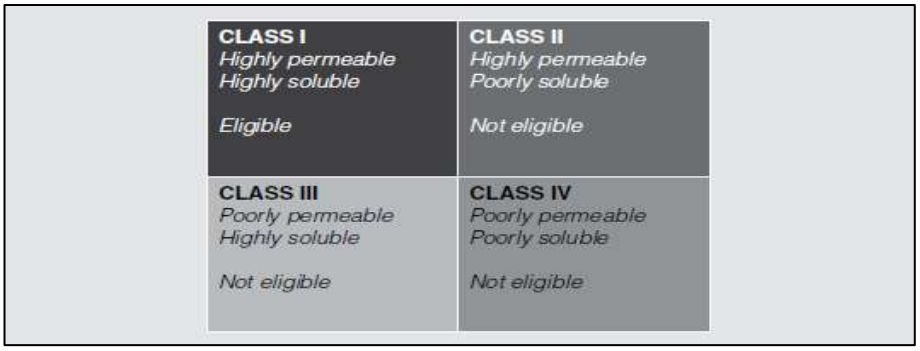

Figure 10: products eligible for the biowaiver procedure under the HHS-FDA guidance

Figure 11: products eligible for the biowaiver procedure under the WHO guidance

1.7.3 Solubility Definitions

A. Solubility FDA definitions

The aqueous solubility of a drug substance is considered as high according to the HHS-FDA BCS criteria when:

• the ratio of the highest orally administered dose (in mg) to the solubility (mg/ml) is 250 ml or lower over the pH range 1–7.5 at 37 °C.

According to HHS-FDA guidance, the determination of the equilibrium solubility is carried out with the shake-flask method (other methods such as acid or base titration are permitted when their ability to predict the equilibrium solubility is justified). The experiments should be carried out in triplicates at a temperature of 37± 1°C. at a number of pH conditions chosen to cover the pH range of 1–7.5. The buffer solutions given in the United States Pharmacopeia (USP) are

appropriate for the tests, but other buffers are also allowed for these experiments. The pH value of each buffer solution should be checked before and after each experiment. Degradation of the API due to pH or buffer composition should be reported together with other stability data.

The reason for the 250-ml cut-off criterion for the dose: solubility ratio is that in pharmacokinetic bioequivalence studies, the API formulation is to be ingested with a large glass of water (8 ounces corresponds to about 250 ml). If the highest orally administered dose can be completely dissolved in this amount of water, independent of the physiological pH value (hence the determination over the pH range 1–7.5), solubility problems are not expected to hinder the uptake of the API in the small intestine.

B. Solubility WHO definition

When an API shows a dose: solubility ratio of 250 ml or lower at 37°C over a pH range of 1.2–6.8, it can be classified as "highly soluble". The decrease in pH from 7.5 in the FDA guidance to 6.8 reflects the need to dissolve the drug before it reaches the mid-jejunum to ensure absorption from the gastrointestinal tract.

• Furthermore, the dose that is to be used for the calculation is the highest dose indicated in the Model List of Essential Medicines (EML).

C. Solubility EMA definition

The pH-solubility profile of the drug substance should be determined and discussed. The drug substance is considered highly soluble if the highest single dose administered as immediate release formulation(s) is completely dissolved in 250 ml of buffers within the range of pH 1 – 6.8 at 37±1 °C. This demonstration requires the investigation in at least three buffers within this range (preferably at pH 1.2, 4.5 and 6.8) and in addition at the pKa, if it is within the specified pH range. Replicate determinations at each pH condition may be necessary to achieve an unequivocal solubility classification (e.g. shake-flask method or other justified method). Solution pH should be verified prior and after addition of the drug substance to a buffer.

1.7.4 Permeability definition

A. Permeability FDA definition

The other important parameter for the BCS is the intestinal permeability of the API.

According to HHS-FDA a drug is considered highly permeable, when 90 % or more of the orally administered dose is absorbed in the small intestine.

Permeability can be assessed by pharmacokinetic studies (for example, mass balance studies), or intestinal permeability methods, e.g. intestinal perfusion in humans, animal models, Caco 2 cell lines or other suitable, validated cell lines. In vivo or in situ animal models or in vitro models (cell lines) are only considered appropriate by HHS-FDA for passively transported drugs. It should be noted that all of these measurements assess the fraction absorbed (as opposed to the bioavailability, which can be reduced substantially by first-pass metabolism).

HHS-FDA suggests use of two different methods for determining the permeability classification if results with one method are inconclusive.

B. Permeability WHO definition

When an API is absorbed to an extent of 85% or more, it is considered to be “highly permeable”. The permeability criterion was relaxed from 90% in the FDA guidance to 85% in the WHO “Multisource document”.

C. Permeability EMA definition

The demonstration of complete absorption in humans is preferred for BCS-based biowaiver applications. For this purpose, complete absorption is considered to be established where measured extent of absorption is ≥ 85 %. Complete absorption is generally related to high permeability.

1.7.5 Biowaiver Study Requirements

A. Biowaiver Study / FDA Requirements

To be considered bioequivalent according to the HHS-FDA biowaiver procedure, a pharmaceutical product:

a) Should contain a Class I API.
b) Should be rapidly dissolving, meaning it should release at least 85% of its content in 30 minutes in three different media (pH 1.2, pH 4.5 and pH 6.8, in a paddle (50 rpm) or basket (100 rpm) apparatus at 37 °C and a volume of 900 ml.
c) Should not contain excipients which could influence the absorption of the API.
d) Should not contain an API with a narrow therapeutic index.
e) Should not be designed to be absorbed from the oral cavity.

The reasoning for the above-mentioned dissolution restrictions is that when a highly soluble, highly permeable API dissolves rapidly, it behaves like a solution in the gastrointestinal tract. If this is the case, the pharmaceutical composition of the product is insignificant, provided that excipients which influence the uptake across the gut wall are excluded from the formulation.

The API is not prone to precipitation after its dissolution due to its good solubility under all pH conditions likely to be found in the upper gastrointestinal tract. The high permeability ensures the complete uptake (> 90%) of the API during its passage through the small intestine. The rapid dissolution of the product guarantees that the API is available long enough for the uptake in the small intestine (the passage time in the small intestine is approximately four hours) and negates any slight differences between the formulations.

Pharmaceutical products containing an API with a narrow therapeutic index should always be tested with in vivo methods, because the risk to the patient resulting from a possible incorrect bioequivalence decision using the biowaiver procedure is considered too high with these kinds of APIs.

As the BCS is only applicable to APIs which are absorbed from the small intestine; drugs absorbed from other sites (e.g. from the oral cavity) are not eligible for a biowaiver.

B. Biowaiver Study / WHO Requirements

In the "Multisource document",1 the WHO has broadened the scope of application of the biowaiver in three directions:

(1) The criteria for classification as a Class I API have been relaxed with respect to both the dose: solubility ratio and permeability requirements.

(2) The new requirements allow pharmaceutical products containing Class III APIs to be considered for a biowaiver, under application of more stringent dissolution criteria.

(3) The document further allows pharmaceutical products containing BCS Class II APIs that are weak acids which have a dose: solubility ratio of 250 ml or less at pH 6.8 to be eligible for the biowaiver procedure, provided that they dissolve rapidly at pH 6.8 and similarly to the comparator product at pH 1.2 and 4.5.

C. Biowaiver Study Requirements According to EMA

BCS-based biowaiver are applicable for an immediate release drug product if

- the drug substance has been proven to exhibit high solubility and complete absorption (BCS class I; for details see section III) and either very rapid (> 85 % within 15 min) or similarly rapid (85 % within 30 min) in vitro dissolution characteristics of the test and reference product has been demonstrated considering specific requirements.
- Excipients that might affect bioavailability are qualitatively and quantitatively the same. In general, the use of the same excipients in similar amounts is preferred.

Generally the risks of an inappropriate biowaiver decision should be more critically reviewed (e.g. site-specific absorption, risk for transport protein interactions at the absorption site, excipient composition and therapeutic risks) for products containing BCS class III than for BCS class I drug substances.

1.7.6 Determining Drug Product Dissolution Characteristics and Dissolution Profile Similarity

1.7.6.1 According to FDA Regulation

Dissolution testing should be carried out in USP Apparatus I at 100 rpm or Apparatus II at 50 rpm (or at 75 rpm when appropriately justified) using 500 mL of the following dissolution media: (1) 0.1 N HCl or Simulated Gastric Fluid USP without enzymes; (2) a pH 4.5 buffer; and (3) a pH 6.8 buffer or Simulated Intestinal Fluid USP without enzymes.

The dissolution testing apparatus used in this evaluation should conform to the requirements in USP (Dissolution). Selection of the dissolution testing apparatus (USP Apparatus I or II) during drug development should be based on a comparison of in vitro dissolution and in vivo Pharmacokinetic data available for the product. The USP Apparatus I (basket method) is generally preferred for capsules and products that tend to float, and USP Apparatus II (paddle method) is generally preferred for tablets. If the testing conditions need to be modified to better reflect rapid in vivo dissolution (e.g., use of a different rotating speed), such modifications can be justified by comparing in vitro dissolution with in vivo absorption data (e.g., a relative BA study using a simple aqueous solution as the reference product).

A minimum of 12 dosage units of a drug product should be evaluated to support a biowaiver request. Samples should be collected at a sufficient number of intervals to characterize the dissolution profile of the drug product (e.g., 5, 10, 15, 20, and 30 minutes). When comparing the test and reference products, dissolution profiles should be compared using a similarity factor (f2).

The similarity factor is a logarithmic reciprocal square root transformation of the sum of squared error and is a measurement of the similarity in the percent (%) of dissolution between the two curves.

FDA Acceptance Criteria: Two dissolution profiles are considered similar when the f2 value is ≥50. To allow the use of mean data, the coefficient of variation should not be more than 20 percent at the earlier time points (e.g., 10 minutes), and should not be more than 10 percent at other time points. Note that when both test and reference products dissolve 85 percent or more of the label amount of the drug in 15 minutes using all three dissolution media recommended above, the profile comparison with an f2 test is unnecessary.

1.7.6.2 According to WHO Regulation

Depending on the BCS classification of the API, based on solubility and permeability characteristics, the testing procedures are:

A. For pharmaceutical products containing Biopharmaceutics Classification System Class I (highly soluble, highly permeable) APIs

For rapidly dissolving pharmaceutical products containing BCS Class I APIs, more than 85% dissolution of the labelled amount is required within 30 minutes in standard media at pH 1.2, 4.5 and 6.8 using the paddle apparatus at 75 rpm or the basket apparatus at 100 rpm. The dissolution profiles of the comparator and the multisource products should be compared by an similarity factor (f2) > 50 or an equivalent statistical criterion.

If within 15 minutes more than 85% of the API are released from the comparator and the multisource formulation under the above-mentioned conditions the products will be considered very rapidly dissolving. In this case the products are deemed to be equivalent and a profile comparison is not required.

B. For pharmaceutical products containing Biopharmaceutics Classification System Class III (highly soluble, low permeability) APIs

A biowaiver can be considered only if both the multisource and the comparator product are *very rapidly dissolving*. Eighty-five percent or more dissolution of the labeled amount of the API should be achieved within 15 minutes in standard media at pH 1.2, 4.5 and 6.8 using the paddle apparatus at 75 rpm or the basket apparatus at 100 rpm. Generally, the risks of an inappropriate biowaiver decision should be more critically reviewed (e.g. site-specific absorption, induction/competition at the absorption site, excipient composition and therapeutic risks) for products containing BCS Class III APIs than for BCS Class I drugs.

C. For pharmaceutical products containing APIs with high solubility at pH 6.8 but not at pH 1.2 or 4.5 and with high permeability (by definition, BCS Class II compounds with weak acidic properties)

These are eligible for a biowaiver provided that the multisource product:

• is *rapidly dissolving*, i.e. 85% or more dissolution of the labeled amount of the API should be achieved within 30 minutes in standard media at pH 6.8 using the paddle apparatus at 75 rpm or the basket apparatus at 100 rpm.

The multisource product exhibits similar dissolution profiles, as determined with the f2 value or equivalent statistical evaluation, to those of the comparator product in buffers at all three pH values (pH 1.2, 4.5 and 6.8).

For multisource products containing BCS Class II API with dose: solubility ratios of 250 ml or less, at pH 6.8, the excipients should also be critically evaluated in terms of type and amounts of surfactants in the formulation.

1.7.6.3 According to EMA Regulations

The results of in vitro dissolution tests at three different buffers (Normally pH 1.2, 4.5 and 6.8) and the media intended for drug product release (QC media), obtained with the batches of test and reference products that were used in the bioequivalence study should be reported.

The results should be reported as profiles of percent of labeled amount dissolved versus time displaying mean values and summary statistics. Unless otherwise justified, the specifications for the in vitro dissolution to be used for quality control of the product should be derived from the dissolution profile of the test product batch that was found to be bioequivalent to the reference product.

1.8 Finished Product Stability Studies

[Ref (18), (19) (20)]

Stability is defined as the extent to which the product retains, within specified limits, and throughout its period of storage and use (i.e. its shelf-life), the same properties and characteristics that it possessed at the time of its manufacture.

1.8.1 Selection of Batches

Data from stability studies should be provided on at least three primary batches of the pharmaceutical product. The primary batches should be of the same formulation and packaged in the same container closure system as proposed for marketing. The manufacturing process used for primary batches should simulate that to be applied to production batches and should provide product of the same quality and meeting the same specification as that intended for marketing. Two of the three batches should be at least pilot scale batches and the third one can be smaller, if justified. Where possible, batches of the pharmaceutical product should be manufactured by using different batches of the active substance. Stability studies should be performed on each individual strength and container size of the pharmaceutical product unless bracketing or matrixing is applied.

1.8.2 Climatic Zones

In order to be able to reduce the amount of stability testing, the number of different long term testing conditions must be reduced to a sufficient extent. This has been proposed by Paul Schumacher in 1972 and Wolfgang Grimm in 1986 and in 1998 when they defined four different long term test conditions, which match with the climatic conditions of the target markets categorized in just four different climatic zones (CZ).

This concept is described in regulatory guidelines and pharmacopoeias and became an established standard in developing pharmaceutical products. At the fortieth meeting of the WHO Expert Committee on Specifications for Pharmaceutical Preparations, Geneva, October 2005, it was recommended to split the current Climatic Zone IV (hot and humid) into CZ IVA – for which 30°C/65% RH will remain the standard long term testing condition – and CZ IVB, for which, if justified, 30°C/75% RH will become the long term testing condition. The following criteria and long term testing conditions are, therefore, proposed:

Figure 12: Climatic zone definition according to WHO

[Ref. (19)]

CZ	Definition	Criteria Mean annual temperature measured in the open air / Mean annual partial water vapor pressure	Long term testing conditions
I	Temperate climate	≤ 15°C / ≤ 11 hPa	21°C / 45% RH
II	Subtropical and Mediterranean Climate	> 15 to 22°C / > 11 to 18 hPa	25°C / 60% RH
III	Hot and dry climate	> 22°C / ≤ 15 hPa	30°C / 35% RH
IVA	Hot and humid climate	> 22°C / > 15 to 27 hPa	30°C / 65% RH
IVB	Hot and very humid climate	> 22°C / > 27 hPa	30°C / 75% RH

Additional testing conditions i.e. accelerated and – if applicable – intermediate conditions have to be used as described in this guideline. The detailed analysis of meteorological measurements as described above, and the evaluation of the climatic conditions in each EMR member state resulted in the following classification and recommended testing condition for long term stability studies (Figure 13):

Figure 13: long term testing condition in the Eastern Mediterranean Region according to WHO

Country	CZ II	CZ III	CZ IVA	Recommended long-term testing condition*
Afghanistan	+	+		30°C/65% RH
Bahrain			+	30°C/65% RH
Djibouti			+	30°C/65% RH
Egypt	+	+		30°C/65% RH**
Iran (Islamic Republic of)	+	+	+	30°C/65% RH**
Iraq		+		30°C/35% RH
Jordan	+	(+)		30°C/65% RH**
Kuwait			+	30°C/65% RH
Lebanon	+	(+)		25°C/60% RH
Libyan Arab Jamihiriya	+	(+)		25°C/60% RH
Morocco	+			25°C/60% RH
Oman		(+)	+	30°C/65% RH
Pakistan	+	+	+	30°C/65% RH
Palestine	+			25°C/60% RH
Qatar			+	30°C/65% RH
Saudi Arabia		+	+	30°C/65% RH**
Somalia			+	30°C/65% RH
Sudan		+	+	30°C/65% RH**
Syrian Arab Republic	+	(+)		25°C/60% RH
Tunisia	+	(+)		25°C/60% RH
United Arab Emirates		+	+	30°C/65% RH
Yemen	+		+	30°C/65% RH

* The hottest and most humid climatic zone has been selected to establish the adequate stability testing condition for a particular country.
** Aqueous-based solutions in semi-permeable packaging, and dosage forms sensitive to low humidity, e.g., hard-gelatin capsules, may require testing at low humidity according to the procedure described in this guideline.

1.8.3 Storage Conditions / General Case

In general, a pharmaceutical product should be evaluated under storage conditions (with appropriate tolerances) that test its thermal stability and, if applicable, its sensitivity to moisture or potential for solvent loss. The storage conditions and the lengths of studies chosen should be sufficient to cover storage, shipment, and subsequent use with due regard to the climatic zone(s) in which the product is intended to be marketed.

In addition as part of the development phase, stability studies conducted on one batch of the pharmaceutical product for up to three months at 50°C/ambient humidity may be useful to identify the formulation and packaging material adequate for extremely hot and dry conditions. Photo-stability testing should be conducted on at least one primary batch of the pharmaceutical product if appropriate.

Long-term, accelerated, and, where appropriate, intermediate storage conditions for drug products are detailed in Table 4 below.

Table 4 : Recommended Storage condition in the stability study / General Case

Study	Storage condition	Minimum time period covered by data at submission
Long-term*	25°C ± 2°C/60% RH ± 5% RH 30°C ± 2°C/65% RH ± 5% RH	12 months
Intermediate**	30°C ± 2°C/65% RH ± 5% RH	6 months
Accelerated	40°C ± 2°C/75% RH ± 5% RH	6 months

***** It is up to the applicant to decide whether long-term stability sturdies are performed at 25°C ± 2°C/60% RH ± 5% RH or 30°C ± 2°C/65% RH ± 5% RH.
***** According to the WHO Eastern Mediterranean Region our region has a long-term study under 25°C ± 2°C/60% RH ± 5% RH
****** If 30°C ± 2°C/65% RH ± 5% RH is the long-term condition, there is no intermediate condition.

1.8.4 Storage Conditions In Palestine

According to the WHO in eastern Mediterranean region (EMR), the storage conditions in Palestine are summarized in table 5:

Table 5: Stability study storage condition in Palestine

Study	Storage condition	Minimum time period covered by data at submission
Long-term	25°C ± 2°C/60% RH ± 5% RH	12 months
Intermediate	30°C ± 2°C/65% RH ± 5% RH	6 months
Accelerated	40°C ± 2°C/75% RH ± 5% RH	6 months

Significant Change and Failure [Ref (20)]

In general, "significant change" for a finished product is defined as:

1. A 5% change in assay from its initial value; or failure to meet the acceptance criteria for potency when using biological or immunological procedures;
2. Any degradation product exceeding its acceptance criterion;
3. Failure to meet the acceptance criteria for appearance, physical attributes, and functionality test (e.g., colour, phase separation, resuspendibility, caking, hardness, dose delivery per actuation); however, some changes in physical attributes (e.g., softening of suppositories, melting of creams, partial loss of adhesion for transdermal products) may be expected under accelerated conditions.

Additional Stability study:

[Ref (20)]

Once the pharmaceutical product has been registered, additional stability studies are required whenever major variations are made like the following:

1. Change in the manufacturing process;
2. Change in the composition of the pharmaceutical product;
3. Change of the immediate packaging.

The stability parameters of drug dosage form can be influenced by environmental conditions of storage (temperature, light, air and humidity), as well as primary package components.

1.9 Test method Validation

[Ref. (21) (22)**]**

The quality of drug product starts from formulation of raw material up to evaluation of finished product at the end of shelf life, and for valid evaluation the analytical procedure must be developed and validated according to the regulations of FDA, USP, etc.

With refer to Food and drug administration guideline " Analytical Procedures and Methods Validation for Drugs and Biologics" several Parameters must be studied and evaluated during method development are specificity, linearity, robustness, ruggedness, range, accuracy, and precision. During early stages of method development, the robustness of methods should be evaluated because this characteristic can help you decide which method you will submit for approval.

The parameters of test method validation for determination of Pregabalin in product using HPLC System are:

1.9.1 Linearity and Range:

Linearity is the ability of the method to elicit test results that are directly proportional to analyte concentration within a given range. Linearity is generally reported as the variance of the slope of the regression line. Range is the interval between the upper and lower levels of analyte concentrations (inclusive) that have been demonstrated to be determined with precision, accuracy and linearity using the method as written. The range is normally expressed in the same units as the test results obtained by the method. A minimum of five concentration levels, along with certain minimum specified ranges are done. For assay, the minimum specified range is from 80-120% of the target concentration. For content uniformity testing, the minimum range is from 70-130% of the test or target concentration. For Related Substances/impurities, linearity demonstrated from 50% of the ICH reporting level to 150% of the proposed shelf life specifications of the related substance. For dissolution testing, linearity should be demonstrated ±20% over the range of the dissolution test (Q-Factor), the minimum range of dissolution test is from Q - 20% to Q + 20%.

1.9.2 Precision:

Precision is the measure of the degree of repeatability of an analytical method under normal operation and is normally expressed as the Relative Standard Deviation (RSD) for a statistically significant number of samples.

Precision is performed at one level, i.e. repeatability. Repeatability is the results of the method operating over a short time interval under the same conditions (injection precision or instrument precision). It is determined from a minimum of nine determinations covering the specified range of the procedure (for example, three levels, three repetitions each), or from a minimum of six determinations, at 100% of the test or target concentration.

1.9.3 Accuracy:

To verify that Pregabalin in Pregabalin Tablet is close to the true value, the accuracy of an analytical procedure measures the closeness of agreement between the value, which is accepted either as a conventional true value or an accepted reference value and value found (i.e. accuracy is a measure of exactness of an analytical method).

It is measured as the percent of analyte recovered by assay, by spiking samples in a blind study. Accuracy is evaluated by analyzing synthetic mixtures (Placebo) spiked with known quantities of Pregabalin.

To document accuracy a minimum of nine determinations over a minimum of three concentration levels covering the specified range (for example, three concentrations, three replicates for each) were collected. It is performed at 80%, 100% and 120% levels of label claim.

At each level studied, replicate samples are evaluated. The RSD of the replicates will provide the analysis variation or how the precision of the test method is. The mean of the replicates, expressed as % of label claim, indicates how the accuracy of the test method is.

1.9.4 Specificity (Stability Indicating Characteristics):

To verify that the assay method unequivocally measure accurately and specifically the Pregabalin in Pregabalin Tablets in the presence of other components that may be expected to be present in the sample matrix.

It is a measure of the degree of interference from such things as other active ingredients, excipients, impurities, and degradation products, ensuring that a peak response is due to a single component only. Specificity is measured and documented in a separation by the resolution factor between the analyte and neighbor peaks.

1.9.5 Robustness:

The robustness of an analytical procedure is a measure of its capacity to remain unaffected by small, but deliberate variations in method parameters and provides an indication of its reliability during normal usage.

1.9.6 Ruggedness (Intermediate Precision):

Ruggedness also known as Intermediate Precision and is the degree of reproducibility of test results by the analysis of the same samples under a variety of conditions, such as different analysts, instruments or days.

1.9.7 Stability of Standard and Sample Solutions (Dissolution Test):

The stability of the standard and sample solutions verified by preparation of a standard and sample solutions and stored under suitable conditions. The standard and sample solutions are analyzed over a specified period of time, using a freshly prepared standard and sample solutions at each time interval for comparison. The acceptable range for standard/sample solution stability is typically between 98% and 102% compared with the initial analysis of the standard / sample solutions.

The procedure may state that the standards and samples need to be analyzed within a time period demonstrating acceptable standard and sample solution stability.

Part II *Literature Review*

2.0 Literature review

2.1 Formulation and Analysis of Pregabalin

[Re. (23)]

Noorana Tehseen, Vinay Rao and Mohamad Abdul Hadi had designed twice daily mini-tablets formulation of Pregabalin under a patent "*Design and characterization of a twice daily mini-tablets formulation of Pregabalin"**; the*** system comprises of 15 matrix mini-tablets weighing 25 mg encapsulated in HPMC capsule. For achieving the sustain release profile, various viscosity grades of Hydroxypropyl methylcellulose polymer (HPMC K4M, K15M, K100M) were used. The mini-tablets were prepared by direct-compression method. The compatibility of drug with other ingredients was checked by FTIR studies.

The values of pre-compression parameters evaluated were within prescribed limits and indicated good free flowing property. The in-vitro performance of mini-tablets formulation showed the desired behavior; nearly 99.57 % of drug was sustained for a period of 12 hrs. FTIR results revealed that there was no interaction between dug and other excipients. The stability study revealed that the formulations were found to be stable.

2.2 Test method validation literature review

[Ref. (24) (25) (26) (27)**]**

There is no Compendial test method for the analysis of the Pregabalin tablets for that there are many published research for many of the researchers on development and validation the Pregabalin test method as:

2.2.1 "Analytical RP-HPLC Method for Development and Validation of Pregabalin in Bulk and the Determination of Pregabalin in Tablet Dosage Form"

In this article "The proposed gradient method was performed using a liquid chromatography of model Waters alliance, 2695 separation model. The chromatographic separation was achieved on a Waters X-Bridge C18, 3.5µm (150 mm X 4.6 mm) column. The gradient LC method employs solution A and B as mobile phase. The solution A contains 0.01 M Ammonium acetate in water (pH – 6.8). The solution B contains a mixture of acetonitrile and methanol in the ratio of 80: 20. The flow rate was 0.8 ml /min and the detection wavelength was 210 nm. For LCMS the above conditions HPLC connected to mass

instrument (Micro mass, Quattro micro TMAPI, ESCI Multi mode Ionization, ESI source , triple quad analyser) and detect the mass". **[ref.** (28)**]**

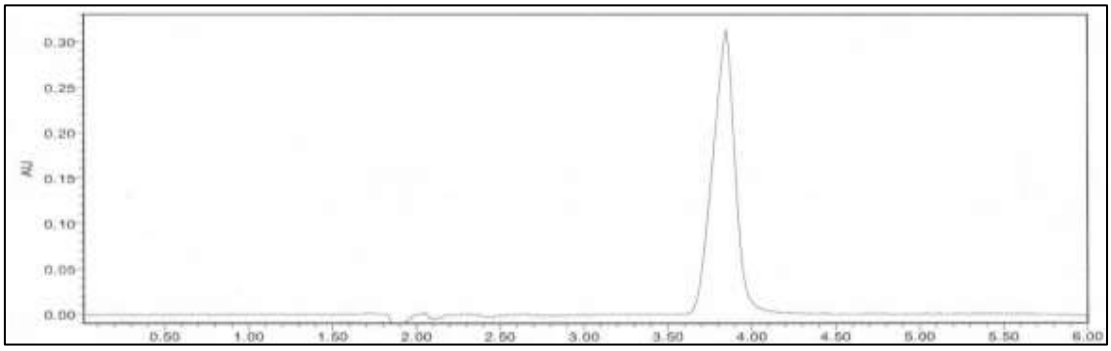

Figure 14: Chromatogram of Pregabalin assay test

2.2.2 "Monitoring of Pregabalin in Pharmaceutical Formulations and Human Serum Using UV and RP-HPLC Techniques: Application to Dissolution Test Method"

In this article "Chromatographic separation was performed on a KROMASIL® 100-5 C-18 column (250×4.6 i.d. mm) (5 µm particle size) as stationary phase with a UV detection at 210 nm using isocratic elution when buffer pH 7 and acetonitrile (96:4, v/v) were used as the mobile phase and the flow rate was 1 ml min-1 at ambient temperature, the retention time was 4.6 minutes". (29)

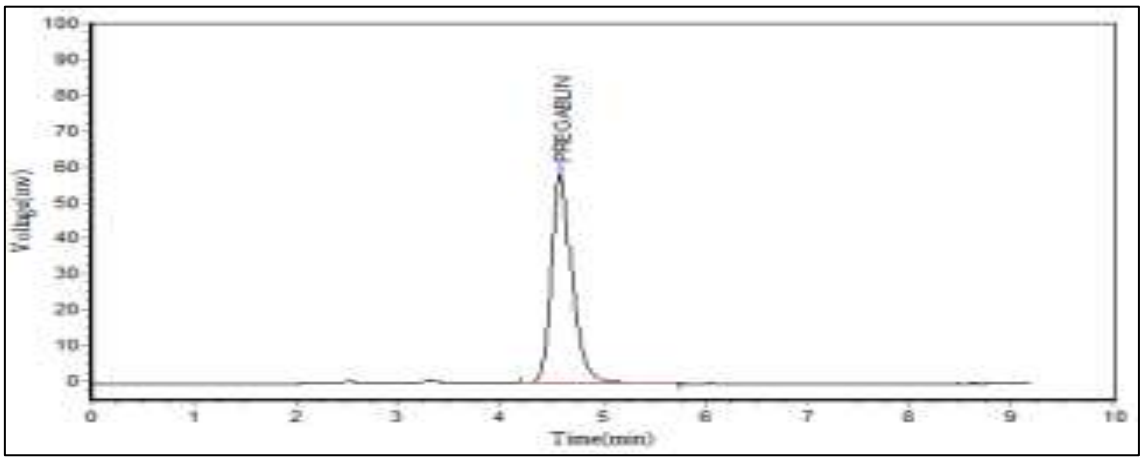

Figure 15: Chromatogram of Pregabalin Dissolution test

2.2.3 Development and Validation of HPLC Method for the Determination of Pregabalin in Capsules

In this article "A simple, precise, specific, and accurate reverse phase HPLC method has been developed for the determination of Pregabalin in Tablet dosage form. The chromatography was set on Hypersil BDS, C8, 150×4.6 mm, 5 µm column using photodiode

array detector. The mobile phase consisting of phosphate buffer pH 6.9 and acetonitrile in the ratio of 95:05 with flow rate of 1 ml/min. The method was validated according to ICH guidelines with respect to specificity, linearity, accuracy, precision and robustness. Lower limit of quantification is 0.6 mg/l. The pregabalin sample solution was found to be stable at room temperature for about 26 h". (30)

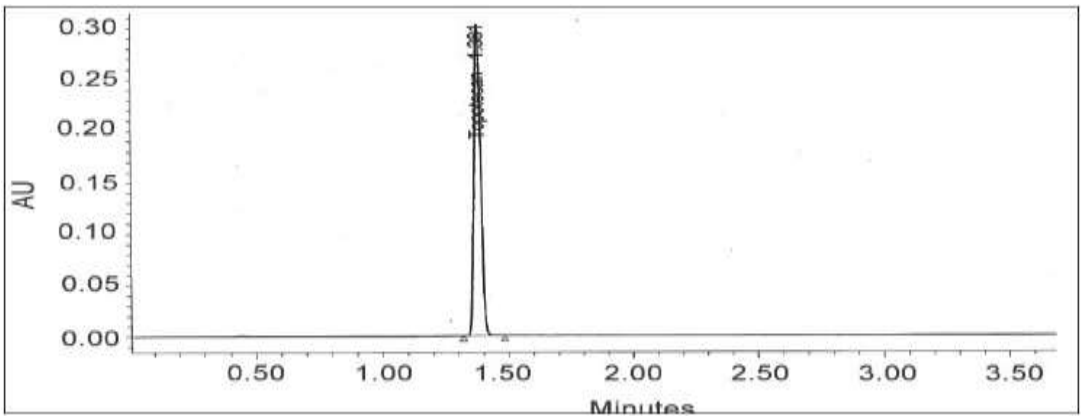

Figure 16: Chromatogram of Pregabalin Dissolution Test - Literature review

2.2.4 A novel method for the determination of Pregabalin in bulk pharmaceutical formulations and human urine samples

In this article "The proposed method was performed using a liquid chromatography of model LC - 2010 CHT (Shimadzu, Kyoto, Japan). Separation was operated on C18 5 µm ODS hypersil column (250 mm × 4.6 mm) using methanol - acetonitrile - 0.02 M di – potassium hydrogen orthophosphate (K2HPO4) (pH - 7.00) (3: 1: 16, v/v/v) mobile phase at a flow rate of 1.0 ml/min. di – potassium hydrogen orthophosphate solution was prepared by dissolving 3.5 g K2HPO4 in 1000 ml double distilled water. Final pH of the mobile phase was adjusted to 7.00 with 0.01 M orthophosphoric acid, prepared daily and degassed by passing through a 0.45 µm Ultipor filter and ultrasonication for 10 min. All separations were performed at room temperature with detection at 210 nm". (31)

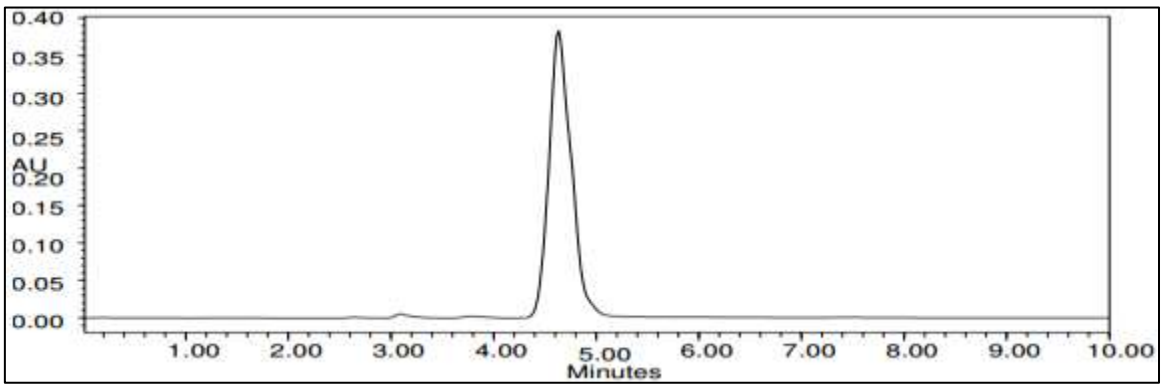

Figure 17: Chromatogram of Pregabalin Dissolution Test - Literature review

Part III *Problems and Objectives*

3.0 Problems and Objectives

3.1 The Research Problem

Pregabalin reference drug product (Lyrica / Pfizer) was developed as opaque hard gelatin shell capsules in dosage strengths of 25, 50, 75, 100, 150, 200, 225, and 300mg. To avoid any possible patients or pharmacist confusion, the capsules are colored, and imprinted with edible ink to indicate the strength and product code, as follows:

Table 6: Strengths and capsule sizes of marketed Lyrica capsules.

Strength [mg]	Capsules Size	Capsules Color (Body/Cap)
25	4	White/White
50	3	White/White(with black ink band)
75	4	White/Orange
100	3	Orange/Orange
150	2	White/White
200	1	Light Orange / Light Orange
225	1	White/ Light Orange
300	0	White/Orange

The capsules are bulky materials in large capsule sizes, can be susceptible to moisture, ingredients can interact with capsules shell, more difficult to fill accurately, and costly, while the tablets are well accepted, elegant, may be presented in different sizes and shapes, have low manufacturing cost, may be coated, and possible for dose splitting.

In addition to above, the tablet dosage forms are generally more stable and high capacity of manufacturing, different manufacturing approaches and many other advantages.

The research regarding the development of Pregabalin immediate release tablets is rare, but the success in developing stable, reproducible, elegant Pregabalin Tablets, that is Biowaiver to reference capsules, will allow pharmaceutical companies that have no designated capsule line in the Production Department, to present their product in the market as tablet dosage form.

3.2 Objectives of the Thesis

Main objectives

Development, Formulation and evaluation of Immediate release Pregabalin Tablets

Specific Objective

1. To develop immediate release (IR) Pregabalin tablets with different strengths (75mg and 300mg per tablet).
2. To assess and evaluate the rate and extent of Pregabalin release from the tablets, using an appropriate dissolution method.
3. To develop and validate a suitable quantitative method of analysis for Pregabalin Tablet dosage form (dissolution, Assay and related Substances).
4. To study the stability of Pregabalin Tablets.
5. To carry out Biowaiver study (dissolution profiles of Pregabalin Tablets versus the reference Pregabalin Capsules).

Part IV *Methodology, Strategy of Research and Experiments*

4.0Methodology and Strategy of Research

4.1 Project Outline

Following are the proposed steps to fulfill the objectives outlined above

A. Pre-formulation

- Selection of API and excipients for formulation.
- Compatibility Study.
- Formulation Trials.
- Selection of Formula.

B. Formulation

- Implementation of Selected formula.
- Selection of Packaging Material.

C. Test Methods Development.

- Assay and Related test method validation.
- Dissolution Test method validation

D. Evaluation of Pregabalin Tablets

- Finished product evaluation.
- Biowaiver Study. (Comparison of Dissolution profile of selected formula versus Reference Product).
- Stability Study.

E. Data Analysis and Discussion.

F. Report results in thesis format.

4.2 Materials and Reagents:

All material used in the formulation of Pregabalin tablets are of pharmaceutical grade, the materials, packaging materials and reagents used are summarized in table (6).
The following list includes the materials required for formulation, packaging and analysis of Pregabalin tablets:

Table 7: Required Materials and Reagents used in the study

Purpose	No.	Item	Description	Function
Formulation	1	Pregabalin	Crystalline powder, Pharmaceutical grade	API
	2	Avicel PH102	Microcrystalline Cellulose Pharmaceutical grade	Filler/Binder
	3	Avicel PH200	Microcrystalline Cellulose , Pharmaceutical grade	Filler/Binder
	4	Starch Maize	Powder, Pharmaceutical grade	Disintegrant/ Glidant
	5	Starch (1500)	Pregelatinized starch Powder, Pharmaceutical grade	Binder/ Disintegrant
	6	Magnesium Stearate	Powder, Pharmaceutical grade	Lubricant
	7	Talc	Powder, Pharmaceutical grade	Glidant / Lubricant
	8	Opadry II White	Powder, Pharmaceutical grade	Coating
Packaging	1	PVC Brown Roll	250 µm thickness	Primary Packaging
	2	PVDC/PVC Brown Roll	250 µm/40 µm thickness	
	3	Aluminum Foil	20 µm thickness	
Analytical	1	Potassium dihydrogen phosphate.	Anhydrous powder, analytical grade	Analytical Reagent
	2	Potassium hydroxide.	Flakes, analytical grade	
	3	Acetonitrile	HPLC grade	
	4	Water	HPLC grade	
	5	Water	WFI grade	Solvent for dissolution
	6	Lyrica Capsules	25mg, 75mg, 150mg, 300mg Strengths	Reference Product

All mentioned material and reagents were donated by Jerusalem pharmaceuticals company, Al-Bireh – Ramallah – Palestine.

4.3 Tools and Equipment

Syringes, Vials, pipettes, glassware, stands, tubes and other equipment were supplied by Jerusalem Pharmaceuticals Company, all equipment and tools are tabulated in the next table.

Table 8: Tools and Equipment used in the study of Pregabalin Tablets

Purpose	No.	Item	Source/Model
Analytical & Evaluation Instrument	1	HPLC Equipped with Photodiode Array UV Detector	Lachrom Elite
	2	UV/VIS spectrophotometer	JASCO V-630
	3	FTIR Spectrophotometer	Nicolet
	4	Dissolution Tester	Electrolab
	5	Bulk/Tap density meter	NA
	6	Hardness Tester	HM LHT-2
	7	Friability Tester	Vankel
	8	Moisture Analyzer	Sartorius
	9	Digital Caliber	TESA Sr. 6N 1948
	10	Analytical balances	Radwag
	11	Magnetic Bars	Freed electric
	12	Hotplate with stirrer	Freed electric
	13	Computer	HP
	14	Funnels, different sizes	Glass A grade
	15	PH Meter	Metrohm 691 pH Meter
	16	Nylon Membrane 0.45 microns	Mercck Millipore Millex-HN
	17	Refrigerator	Kirsch Co.
	18	Sonicator	Elmasonic S450
	19	Water bath	Local manufacturer
	20	Spatulas	Stainless Steel Spatula
	21	Vortex	VLEP Scientifica
	22	Octadecylsilane chemically bonded to totally porous silica particles (4.6mm * 25cm) HPLC Column	Luna HPLC Column
	23	Three Climatic Chambers: (1) 25±2 C/60±5% RH (2) 30±2 C/65±5%RH (3) 40±2 C/75±5%RH	 Firlabo PGC-USA Firlabo
Production Equipment	24	Sifter & Meshes (Mesh#20, Mesh#30, Mesh#60, Mesh#120)	Sieve Master
	25	Tablet Compression Machine.	HM Pharmachine
	26	10 mmx 5mm oblong punches, 19mm x 9 mm oblong punches	Pearl Elizabeth
	27	Coating System	Local manufacturer
	28	Blistering Machine	
Tools	29	Beakers 100ml, 300ml, 1000 ml	Glass grade B
	31	Volumetric Pipettes	Glass grade A
	32	Graduated Cylinder (1000ml)	Glass grade B
	33	Volumetric Flasks (50, 100; 200;1000 ml)	Glass grade A
	34	Erlenmeyer flasks	Glass grade B

4.4 Pre-Formulation

4.4.1 Selection of API and excipients

Pregabalin is not found in any international pharmacopeia, it is a white to off-white, highly crystalline, non-hygroscopic powder.

Pregabalin used in this study is synthesized as the single enantiomer S, and has consistent polymorph Form I. (Ref. Patent US 2006/0270871 AI)

Particle Size is part of the specification of API, but is not expected to be critical parameter with regards to the bioavailability of the drug product, taking into the account the water solubility of Pregabalin.

According to "HandBook of Pharmaceutical Excipient 4th edition" there is a relation between the type, function and concentration of excipients.

The Specifications and COA of Pregabalin used are shown in Appendix 1.

All excipients used are of pharmaceutical grade and used in percentages according to FDA as illustrated in Table 8. They are tested and evaluated before use, and the COAs are shown in Appendix 1.

Table 9: List of some Excipients and their application

Excipient	Application	% Concentration
MCC PH102 "Avicel"	Adsorbant	20-90
	Anti-adherant	5-20
	Tablet disintegration	5-15
	Tablet binder/ diluent	20-90
	Capsule binder/ diluent	20-90
Starch 1500	Tablet binder (direct compression)	5-20
	Tablet disintegrant	5-10
Starch Maize	Tablet Glidant	3-10%
	Disintegrant	3-25
Talc	Glidant / lubricant for tablet	1- 10
Magnesium Stearate	Lubricant in Tablet and capsule	0.25-5

4.4.2 Compatibility Study:

[Ref (32) (33) (34) (35) (36)]

The purpose of this stage is to find compatible excipients with the active ingredient Pregabalin. The study is done by obtaining the FTIR spectra for each excipient alone and for its mixture with Pregabalin, then analyzed for any possible drug- excipient interactions.

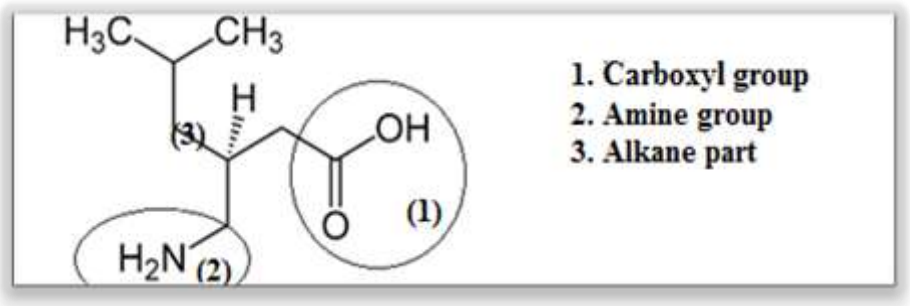

Figure 18: Pregabalin Structure

Sample preparation

The physical mixtures were prepared in 1:1 ratio for each excipient with Pregabalin and then passed through intended mesh as manufacturing procedure. Samples of Pregabalin and excipients were placed in glass vial type 1, closed and labeled. Then the vials were stored under stressed conditions at 40˚C for 30 days. The compatibility of drug with excipients was studied by FT-IR.

The Physical mixture were prepared according to Table 10

Table 10: Samples preparation for compatibility study

API	Excipient	Ratio [API: Excipient]	Mesh# for Excipient *
Pregabalin	MCC PH102	1:1	60
Pregabalin	Talc	1:1	60
Pregabalin	Magnesium Stearate	1:1	100
Pregabalin	Starch Maize	1:1	60
* Pregabalin was sieved through mesh No. 35			

4.5 Selection the formula

All excipients used are widely involvement in oral pharmaceutical dosage without any toxic or irritant problem or excipient-excipient physically or chemically interaction.

The reference Pregabalin drug product (Lyrica capsules) contains lactose monohydrate, maize starch and talc as excipients in addition to the API Pregabalin. In the design of Pregabalin Tablet we used Microcrystalline cellulose in two different grades as binder/ diluent, talc powder as Glidant, Magnesium stearate as lubricant and Starch/Pregeltinized starch used as Glidant-disintegrant

The planned series of formulae are listed in Table 11.

Table 11: Formulae of Pregabalin Tablets

Function	Ingredient	Quantity * (mg)						
		F-1	F-2	F-3	F-4	F-5	F-6	F-7
API	Pregabalin API	300×2 (91.7%)	300 (52.1%)	300 (52.1%)	300 (49.5%)	300 (45.7%)	300 (45.3%)	300 (45.3%)
Filler/ Binder	MCC - Avicel pH102	---	200 (34.7%)	---	200 (33.0%)	300 (45.7%)	---	300 (45.3%)
	MCC - Avicel 200	---	---	200 (34.7%)	---	---	300 45.3%)	---
Disintegrant	Starch Maize	---	40 (6.9%)	40 (6.9%)	70 (11.6%)	---	---	20 (3.0%)
	Pregelatinized starch 1500	---	---	---	---	20 (3.0%)	20 (3.0%)	---
Lubricant	Magnesium Stearate	27×2 (8.3%)	6 (1.0%)	6 (1.0%)	6 (1.0%)	6 (0.9%)	12 (1.8%)	12 (1.8%)
Glidants	Talc	---	30 (5.2%)	30 (5.2%)	30 (5.0%)	30 (4.6%)	30 (4.5%)	30 (4.5%)
Weight of Tablet*		654	576	576	606	656	662	662
*** Quantities are calculated for one unit.** *** Batch Size is 1000 tablet for each Trial formula.**								

The powder blends were tested before compression for their flowability, and the compressed tablets were tested for their mechanical properties (hardness, friability, average weight, weight uniformity, thickness, and disintegration time and tablet appearance).

The analytical methods are specified in the following sections of this chapter, and the results are summarized in Chapter 5, The Flowability of powder blends can be described according to Table 12.

Formula selection will depend on ease of production, flowability of powder blend and performance of compressed tablets.

Table 12: Carr Index Classification and Powder Flowability

Carr Index (%)	Flow	Hausner ratio
5 – 12	Free flowing	The Hausner ratio varies from about 1.2 for a free - flowing powder to 1.6 for cohesive powders.
12 – 16	Good	
18 – 21	Fair to passable*	
23 – 35	Poor*	
33 – 38	Very poor	
≥40	Extremely poor	
Calculation method		
Carr index (compressibility) = $\frac{\text{(Tapped density- Untapped density)}}{\text{Tapped density}} \times 100$		**Hausner ratio =** $\frac{\text{Tapped bulk density}}{\text{Poured bulk density}}$
* May be improved by the addition of glidant.		

All results are recorded, tabulated and discussed in Chapter 5.

4.6 Manufacturing Procedure

The general manufacturing procedure of tablet formulations shown in table 11 is:

1.0 All components are accurately weighted according to the intended formula.

2.0 All components are passed manually through the corresponding US Sieve No.
Pregabalin: Sieve No.35
Magnesium Stearate: Sieve No.100
All other components: Sieve No. 60

3.0 All components except lubricant and glidant are combined and mixed together manually in standard size of PE bag for about 10 minutes.

4.0 Lubricant and glidant are added to blend and mixed for 30 seconds.

5.0 Blends are compressed using pilot rotary tablet press, equipped with eight oblong punches

6.0 In-process control of weight variation is carried out during compression at least twice. Weight values reported in milligram. Mean and SD are calculated at weight variation ±5%. In addition to weight, compression difficulties e.g. sticking, capping and tablet appearance are noted.

7.0 Repeat the previous prescribed method of tablet manufacturing, and compressed about 1000 tablets from each tablet formulation.

4.7 Implementation of Selected Formula

Pregabalin tablets, with two strengths 300mg tablet and 75mg tablet were prepared and coated using the component ratio of selected formula, and the mentioned general manufacturing process

The composition of two strengths are dose weight proportional i.e. the percentage of each excipient relative to the API is the same or not exceeding Level 1 change, as illustrated in table 13.

Table 13: Selected formula F5 and the Size of Batch for further study

Ingredient	**Pregabalin Tablet 300mg**			**Pregabalin Tablet 75mg**		
	Quantity / Unit (mg)	**Relative to API**	**Relative to weight of core tablet**	**Quantity / Unit (mg)**	**Relative to API**	**Relative to weight of core tablet**
Pregabalin API	300.0	100%	45.7%	75.0	100%	45.7%
Avicel PH 102	300.0	100%	45.7%	75.0 mg	100%	45.7%
Pregelatinized starch 1500	20.0	6.6%	3.1%	5.0 mg	6.6%	3.1%
Magnesium Stearate	6.0	2.0%	0.9%	1.5 mg	2.0%	0.9%
Talc	30.0	10.0%	4.6%	7.5 mg	10.0%	4.6%
Weight of core Tablet	**656**	**-**	**-**	**164**	**--**	**-**
Opadry II white	14	2.1%*	--	3.5 mg	2.1%*	--
Water purified**	86	--	--	21.5 mg	--	--
Weight of coated Tablet	**670.0 mg**			**167.5 mg**		
Batch Size	**2000 Tab**			**8000 Tab**		
***Relative to weight of core tablet** ****Evaporated during drying in coating stage**						

The prepared tablet cores were coated, by using ready mixed coating powder (Opadry II white). The coating process was carried out in conventional coating pan rotated at 18rpm, inlet air temperature was 45-50C, and spraying rate was 10 ml/min

The Pregabalin tablets manufacturing procedure is summarized in the next flowchart:

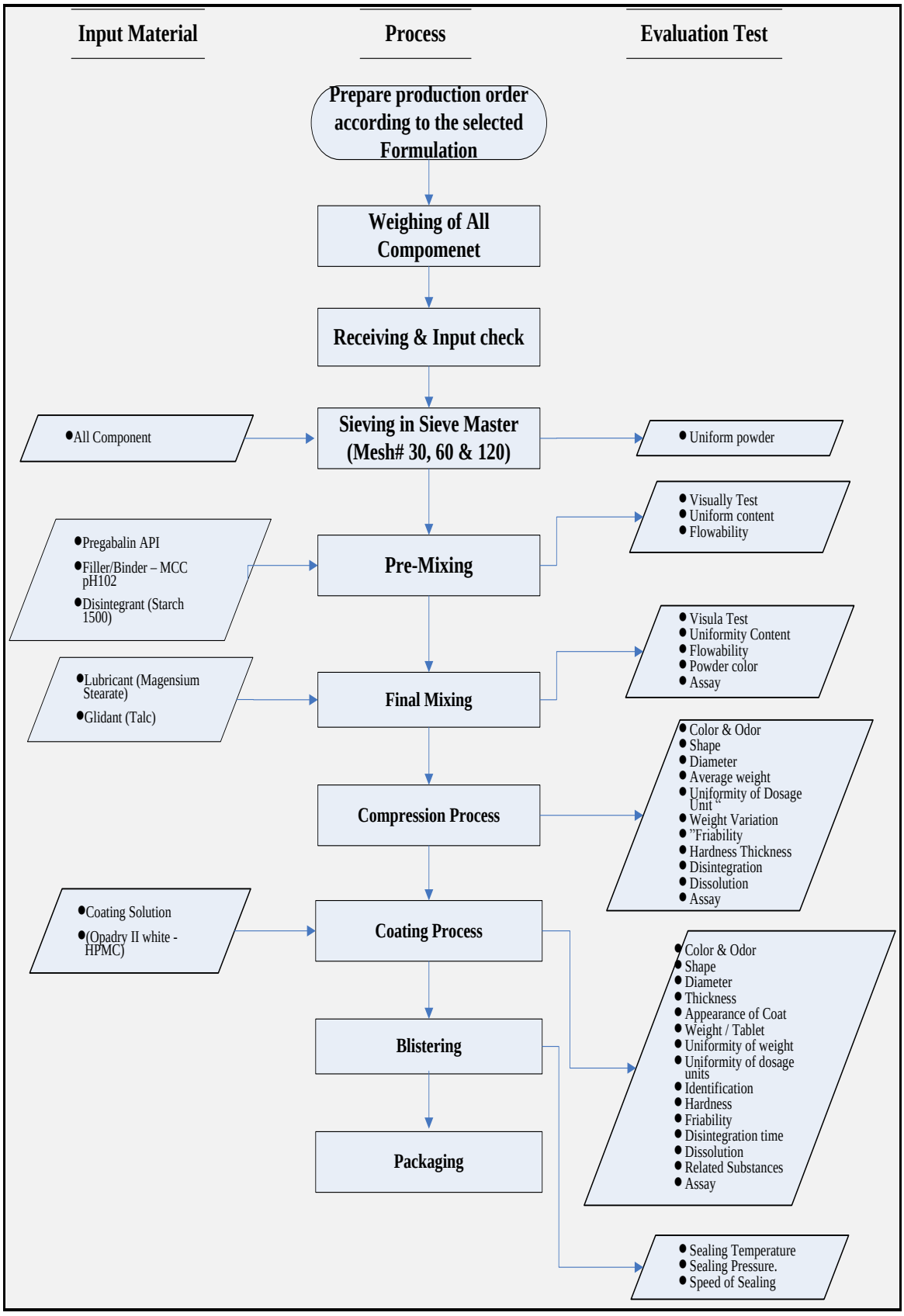

Figure 19: Process flow chart for manufacturing Pregabalin tablet by direct compression

4.8 Test Methods

4.8.1 Pregabalin blends Characterization

Hausner ratio is calculated for each formulation as follows:

Weigh an empty 100-ml glass cylinder (W_0). Pass the powder through sieve mesh # 16, and fill sufficient quantity of powder to an apparent volume of about 70 ml, and reweigh (W_1). Record the volume of powder as (V_0). The cylinder is allowed to jolt for 1000 taps using Erweka SVM Tapped density Tester, and the tapped volume is recorded (V_1). The bulk, tapped densities and Hausner ratio are calculated as follows:

- Bulk Density $= \frac{W1-W0}{V0}$
- Tapped Density $= \frac{W1-W0}{V1}$
- Hausner ratio $= \frac{\text{tapped density}}{\text{bulk density}}$

Evaluation criteria: Values of Hausner Ratio less than 1.25 indicate good flow, while values higher than 1.5 indicate poor powder flow.

4.8.2 General Appearance

The general appearance (color, shape, thickness and surface) of the formulated tablets was inspected and tested visually, while the dimensions were measured by using a suitable caliper.

Thickness: The thickness of the tablets was determined using a thickness TESA dial caliper 150mm/6 in. Ten tablets from each batch are used.

Thickness values are reported in millimeters. Mean and SD are calculated.

4.8.3 Hardness

Tablet hardness or breaking force is measured by placing the tablet between two platens of the hardness tester, one of which moves to apply sufficient force to the tablet to cause fracture. The force required to cause fracture is recorded. It is repeated for 10 tablets and the hardness values are reported in kilograms force (Kgf). Mean and SD are calculated.

Acceptance Criteria: NLT 5 Kgf

4.8.4 Friability

Friability is carried out as per USP 38 <1216>: For Pregabalin 75mg tablets, take a sample of whole tablets corresponding as near as possible to 6.5 g. For Pregabalin 300mg tablets, take a sample of 10 whole tablets. The tablets is carefully dedusted prior to testing..

Accurately weigh the tablet sample, and place the tablets in the drum of friabilator (Copley FR1000 Friabilator). Rotate the drum 100 times, and remove the tablets. Remove any loose dust from the tablets as before, and accurately weigh.

Acceptance Criteria: maximum mean weight loss from the three samples of not more than 1.0% is considered acceptable for most products.

4.8.5 Assay and related tests

Assay and related substances test method are described under "Test Methods Validation (Methodology)" section.

Acceptance limit: 90.0%-110.0%

4.8.6 The Uniformity of dosage Units:

The uniformity of dosage units in Pregabalin Tablets can be demonstrated by Weight Variation and that depend on the weight of active ingredient (≥25mg) and the percent of label claim (≥25%).

Uniformity of dosage units test as per USP Method <905> is carried out by means of weight variation. i.e. 10 tablets of each formulation are accurately weighed individually using an electronic balance (Precisa XT 2208). The Pregabalin content of each unit is calculated expressed as percent of label claim from the weight of the individual tablet and the result of the assay. Acceptance value (AV) is calculated according to USP chapter <905>.

Acceptance value: AV should not be more than 15.0. Each tablet should contain between 85.0% and 115.0% of label claim.

4.8.7 Disintegration Test

Carry out the test in Purified water using USP Disintegration tester. Record the time when the tablet disintegrate and all particles pass through mesh # 10 screen. If any residue remains, it must have a soft mass.

Acceptance Criteria: Disintegration time not more than 15min.

4.8.8 Dissolution Testing:

[Ref (13), (37)]

The dissolution rate studies are carried out by using dissolution tester apparatus USP Type II (paddle type).

Sample size (n): 6 tablets.

Detection UV Wavelength: 280nm

Table 14: Dissolution Parameters

Parameters	Types and Value
Dissolution apparatus	**USP Apparatus II (paddle)**
Dissolution medium	**0.06 N HCl solution**
Temperature	**37 ±0.5°C**
Initial volume	**900 ml**
Speed	**50 rpm**
Filter size	**0.45µm**
Volume withdrawn	**5 ml**
Volume replaced	**5 ml**
Sampling times:	**10, 15, 30, 45 minutes**

The dissolution test is performed as per method detailed in section "Validation of Test Methods".

The dissolution rates of tablets are monitored using Erweka EDT. Six Tablets are tested. The amount dissolved of Pregabalin is determined by Lachrom HPLC System at wavelength 280 nm.

Filtered sample solution is compared to standard solution of known concentration of Pregabalin WS. The percent drug dissolved at each sampling time is calculated after correction for the cumulative amount removed in previous samples.

Acceptance Criteria: Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes

4.9 Biowaiver Study

[Ref. (38)]

The comparison dissolution study is performed between the reference drug product (Lyrica 300mg and 75mg Capsules) and Samples from Pregabalin 300mg and 75mg Tablets processed by the selected formulation. Sample size for each run is six tablets.

Table 15: In-Vitro comparison dissolution parameters

Parameters	Types and Value
Dissolution apparatus	**USP Apparatus II (paddle)**
Dissolution medium	**1. 0.06N HCl solution** **2. Acetate buffer pH 4.5** **3. Phosphate buffer pH 6.8**
Temperature	**37 ±0.5°C**
Initial volume	**900 ml**
Speed	**50 rpm**
Filter pore size	**0.45µm**
Volume withdrawn	**5 ml**
Volume replaced	**5 ml**
Sampling times:	**10, 15, 30, 45 minutes**
Detection UV Wavelength	280nm

Similarity factor Calculation

The similarity factor (f2) given by SUPAC guidelines for Immediate release dosage form was used as a basis to compare dissolution profile. The dissolution profiles of products are compared using a similarity factor (f2). This similarity factor is calculated by following formula:

$$F2 = 50 * Log \left[\frac{100}{\sqrt{1 + \frac{\sum_{t=1}^{t=n} [\bar{R}(t) - \bar{T}(t)]^2}{n}}} \right]$$

$\mathbf{R_{(t)}}$ = mean % API dissolved of reference product at time point x

$\mathbf{T_{(t)}}$ = mean % API dissolved of test product at time point x

Two dissolution profiles are considered similar when the f2 value is ≥50. To allow the use of mean data, the coefficient of variation should not be more than 20% at the earlier time points (e.g., 10 minutes), and should not be more than 10% at other time points.

4.10 Finished Product Stability Studies

[Ref (18), (19)]: (19)]

The stability of the finished product is carried out in its final marketed package, in compliance with storage conditions illustrated below

Stability Study is evidence of how the quality of an Active Pharmaceutical Ingredient (API) or Finished Pharmaceutical Product (FPP) varies with time under the influence of a variety of environmental factors such as temperature, humidity and light.

Environmental Conditions

Samples from the selected formula will be kept at different environmental conditions as given below for a month and will be analyzed at initial, 14 days and 30 days.

Table 16: Stability Environmental Conditions

Study	Storage Condition	Testing time interval
Long Term	25°C ± 2°C/ 60% RH± 5% RH	initial, 14 days and 30 days
Intermediate	30°C ± 2°C/ 65% RH± 5% RH	initial, 14 days and 30 days
Accelerated	40°C ± 2°C/ 75RH± 5% RH	initial, 14 days and 30 days

1.

Stability parameters

The tablets Stability will be evaluated for the following parameters: Physical appearance, Dissolution rate, Disintegration time, Hardness, Friability, Drug content assay and Degradation.

4.11 Test Methods Validation (Methodology)

4.11.1 Assay and Related substances

Preparations and HPLC Conditions

Buffer Preparation

Accurately weigh and transfer about 1.36 g Potassium Dihydrogen phosphate in 1000ml of water, sonicate to dissolve .Adjust the pH of the Solution to 6.9 ± 0.1 with Potassium hydroxide. Filter using a 0.45 µm membrane filter.

Mobile Phase:

Prepare a filtered and degassed mixture of Buffer, and acetonitrile (94:6).

Stock Standard Solution (10.0 mg/ml):
Transfer an accurately weighed 2000 mg of Pregabalin to a 200-ml volumetric flask, dissolve and dilute with Mobile Phase to volume; and mix, Filter through 0.2µm membrane filter.

Nominal Standard Solution (1.0 mg/ml):
Transfer an accurately measured 5-ml of stock standard solution to a 50-ml volumetric flask and dilute with Mobile Phase to volume, and mix, to obtain a solution having a known concentration (1.0 mg/ml of Pregabalin).

System Suitability Solution (5.0 mg/ml):
Transfer an accurately weighed 250 mg of Pregabalin and 5.0 mg of RS Impurity IV (Lactam) to a 50-ml volumetric flask, Add about 20-ml o Mobile phase, sonicate for about 15 minutes, dilute with Mobile Phase to volume; and mix, Filter through 0.2µm membrane filter.

Chromatographic Conditions:
Detection Wavelength: 210 nm.
Column: Octadecylsilane chemically bonded to totally porous silica particles, 5 to 10µm in diameter (25cm X 4.6 mm id).
Flow Rate: 1.5 ml/min.
Column Temperature: Ambient Temperature.
Injection Volume: 20 µL.

Procedure:
General points to consider:

1. The stock standard and sample solutions are dissolved and diluted to final volume with Mobile Phase.
2. The test and standard concentrations are close if not the same.
3. The test samples are bracketed by standards during the analytical procedure.
4. Test samples are filtered before injection or before further dilution.

The criteria of test method validation for assay determination of Pregabalin in Pregabalin Tablets using HPLC System are:

4.11.1.1 Linearity and Range:

Preparation of Standards: Using the Stock Standard Solution (10.0 mg/ml of Pregabalin) to prepare separate standards covering the range between (40%-160%) of the nominal concentration according to the following table:

Table 17: preparation of linearity solution

Conc. (%)	Conc. of Pregabalin (mg/ml)	Volume Pipetted from Stock St Solution (ml)	Volumetric Flask Final Volume (ml)
40	0.4	2	50
60	0.6	3	50
80	0.8	4	50
100	1.0	5	50
120	1.2	3	25
140	1.4	7	50
160	1.6	4	25

Data Analysis:

Plot Intensity versus standard concentrations prepared for linearity over the range of standard solutions and calculate:

- The least squares linear regression analysis of the linearity data.
- The RSD for replicates of each concentration over the range.
- Determine slope and Y-intercept.

Acceptance Criteria:

The correlation coefficient (R^2) is not less than 0.990 for the least squares method of analysis of the line. The RSD of the standards will not be greater than 5.0% at all standard concentrations.

4.11.1.2 Intermediate Precision (Ruggedness):

Procedure:

This test is performed by analysis of the nominal standard solution (1.0 mg/ml of Pregabalin) by different analyst and on different days (matrix design). Inject the working standard solution at nominal concentration ***(1.0 mg/ml of Pregabalin)*** into the liquid chromatograph six times.

Calculate the relative standard deviations (RSD) of the replicate injections.

Acceptance Criteria: Verify that, the Relative Standard Deviation for the replicate injections is not more than 2.0%

4.11.1.3 Precision:

Inject the nominal standard solution six times into the liquid chromatograph.

Calculate the RSD of the replicate injections for the nominal standard concentration (100% Concentration, 1.0 mg/ml Pregabalin) and verify that the RSD is not greater than 2.0%

4.11.1.4 Accuracy:

Procedure:

Pregabalin Tablets Placebo Preparation:

Prepare Pregabalin Tablets placebo according to the formulation procedure of Pregabalin Tablets. The concentration of placebo in the analyzed sample (Nominal Concentration 1.0 mg/ml of ***Pregabalin***) is expected to be about 1.0 mg/ml.

Stock Placebo Preparation (10.0 mg/ml):

Transfer an accurately measured 1000mg of ***Pregabalin Tablets Placebo*** to a 100-ml Volumetric Flask. Dissolve and dilute with Mobile Phase to volume, mix and Filter through 0.2µm membrane filter, to obtain a solution having a concentration (10.0mg/ml of Placebo).

Preparation of Spiked Samples:

Place the required placebo volume into the analysis flask and add known amount of Pregabalin by volume from the stock standard solution to the analysis flask according to the following table:

Table 18: Preparation Solution of Accuracy test

Conc. (%)	Placebo Conc. (mg/ml)	*Pregabalin Conc.* (mg/ml)	Pipetted Volume of Stock Placebo (2)	Pipetted Volume of Stock St. (1)	Flask Volume (ml) (3)
80	0.8	0.8	4-ml	4-ml	50
100	1.0	1.0	5-ml	5-ml	50
120	1.2	1.2	3-ml	3-ml	25

1: Stock Standard Solution.
2: Stock Placebo Solution.
3: Volumetric flask (ml) diluted to final volume with Mobile Phase (as sample preparation in the Pregabalin TM)

- Analyze the prepared Solutions according to the test method of analysis for Pregabalin Tablets.
- Calculate the recovery data for each determination; calculate the average of recovery data and the RSD for each level.
- Verify that the mean recovery of the assay should be within 100±2.0% at each concentration over the range of 80% – 120% of the nominal concentration.

Acceptance Criteria: The mean recovery of the assay should be within 100±2.0% at each concentration over the range of 80% – 120% of nominal concentration.

4.11.1.5 Specificity (Stability Indicating Characteristics):

Procedure:

Different reagents were added to the Nominal Standard Solution (1.0 mg/ml of Pregabalin) and added to the Pregabalin Tablets samples (placebo spiked in Pregabalin standard). Stressed standard and sample solutions prepared according to the following table:

Table 19: Stress condition and solution preparation – Specificity Test

Tablets No.	Pipetted Volume from Stock St.(1)	Pipetted Volume from Stock Placebo(2)	Reagent Added/ Stress Condition	Total Volume (ml)3
1	5-ml	5-ml	---	50
2	5-ml	5-ml	0.5M HCl	50
3	5-ml	5-ml	0.5M NaOH	50
4	5-ml	5-ml	6% H_2O_2 & Heat at 70$^oC$for 15 min.	50
5	5-ml	5-ml	Heat in water bath at 70^oC for 1-Hour	50

1: Stock standard solution prepared in section 5.3 (10.0 mg/ml of Pregabalin).

2: Stock Placebo Solution was prepared in section 6.3.3.3 (5.674 mg/ml of Pregabalin placebo).

3: Volumetric flask (ml) diluted to final volume with Mobile Phase solution.

Verify the Pregabalin peaks will have baseline of chromatographic resolution of at least 1.5 from all other sample compounds, or the unresolved components at their maximum expected levels.

4.11.1.6 Robustness:

Procedure:

Use the nominal standard solution (1.0 mg/ml Pregabalin).

Variation of Method Parameters:

- **Flow Rate:** Variation of the flow rate to 1.65 ml/min instead of 1.50 ml/min.
- **Detection Wavelength:** Variation of wavelength to 208nm instead of 210 nm.

Inject the nominal standard solution (1.0 mg/ml of Pregabalin) into the Liquid Chromatograph six times and analyze according to Pregabalin Tablets test method**.**

Data Analysis:

Verify that the influence of variation in method parameters is within the previously acceptance criteria, the parameter said to be within the methods robustness range.

Acceptance Criteria: Verify that, the Relative Standard Deviation for the replicate injections is not more than 2.0%.

4.11.2 Validation Methodology for Dissolution

Preparations and Chromatographic HPLC Conditions:

Phosphate buffer: Accurately weigh and transfer about 1.36 g Potassium dihydrogen phosphate in 1000ml of water, sonicate to dissolve. Adjust the pH of the Solution to 6.9 ± 0.1 with Potassium hydroxide. Filter using a 0.45 µm membrane filter.

Mobile Phase: Prepare a filtered and degassed mixture of Buffer, and acetonitrile (94:6).

Stock Standard Solution (3.333 mg/ml):

Transfer an accurately weighed 666.6mg of Pregabalin WS to a 200-ml volumetric flask. Dissolve and dilute with dissolution medium (distilled water) to volume, and mix, Filter through 0.2µm membrane filter, to obtain a solution having a known concentration of Pregabalin WS (3.333 mg/ml).

Nominal Standard Solution (0.3333 mg/ml):

Transfer an accurately measured 5-ml of stock standard solution to a 50-ml volumetric flask and dilute with dissolution medium to volume, and mix, to obtain a solution having a known concentration (0.3333 mg/ml of Pregabalin WS).

Chromatographic HPLC Conditions:

Detection Wavelength: 210 nm.

Column: Octadecylsilane chemically bonded to totally porous silica particles (4.6mm * 25cm).

Flowrate: 1.5 ml/min

Temperature: Ambient Temperature.

Dissolution Test Conditions:

Dissolution Medium: 0.06N HCl Solution.

Apparatus*:* 6 (Paddle).

R.P.M*:* 50

Time*:* 30 min.

Volume*:* 900 ml.

Procedure:

General points to consider:

5. The stock standard and sample solutions are dissolved and diluted to volume with dissolution medium.
6. The test and standard concentrations are close if not the same.
7. The test samples are bracketed by standards during the analytical procedure.
8. Test samples are filtered before measured or before further dilution.

The criteria of test method validation for Dissolution determination of Pregabalin in Pregabalin Tablets using Chromatographic HPLC are:

4.11.2.1 Linearity and Range:

Procedure:

Preparation of Standards: Using the Stock Standard Solution (3.333 mg/ml of Pregabalin WS) to prepare separate standards covering the range between (40%-150%) of the nominal concentration according to the following table:

Table 20: Solutions for Linearity Test

Conc. (%)	Conc. of Pregabalin WS (mg/ml)	Volume Pipetted from Stock St Solution (ml)	Volumetric Flask Final Volume (ml)
40	0.1333	2	50
60	0.1999	3	50
80	0.2666	4	50
100	0.3333	5	50
120	0.3999	3	25
150	0.4999	3	20

Data Analysis:

Plot Intensity versus standard concentrations prepared for linearity over the range of standard solutions and calculates:

- The least squares linear regression analysis of the linearity data.
- The RSD for replicates of each concentration over the range.
- Determine slope and Y-intercept.

Acceptance Criteria: The correlation coefficient (R^2) is not less than 0.98 for the least squares method of analysis of the line. Also, the RSD of the standards will not be greater than 5.0% at all standard concentrations.

4.11.2.2 Precision:

Inject the nominal standard solution six times into the liquid chromatograph.

Calculate the RSD for the replicate readings of the nominal standard concentration (100% Concentration, 0.3333 mg/ml Pregabalin WS) and verify that the RSD is not greater than 5.0%.

4.11.2.3 Accuracy:

It is measured as the **percent of analyte recovered** by assay, by spiking samples in a blind study. Accuracy is evaluated by analyzing synthetic mixtures (Placebo) spiked with known quantities of ***Pregabalin WS.***

To document accuracy a minimum of **nine** determinations over a minimum of three concentration levels covering the specified range (for example, three concentrations, three replicates for each) were collected. It is performed at 50, 75 and 100% levels of label claim.

At each level studied, replicate samples are evaluated. The RSD of the replicates will provide the analysis variation or how the precision of the test method is. The mean of the replicates, expressed as % of label claim, indicates how the accuracy of the test method is.

Procedure:

Pregabalin Tablets Placebo Preparation:

Prepare Pregabalin Tablets placebo according to the formulation procedure reported in the production file of Pregabalin 300mg Tablets. The concentration of placebo in the analyzed sample (Nominal Concentration 0.333 mg/ml of Pregabalin WS) was estimated according to the final formula.

Preparation of Spiked Dissolution Samples:

Place the required placebo weight into the dissolution vessels and add known amount of Pregabalin WS by volume from the stock standard solution to the dissolution vessels according to the following table:

Table 21: Sample preparation for accuracy test

Spiked Sample Preparation				
Conc. (%)	**Placebo wt. (mg)**	**Pregabalin weight (mg)**	***Pregabalin WS Vessel* Conc. (mg/ml)**	**Diluent Volume**
50	150.0	150.0	0.1666	900
75	225.0	225.0	0.2500	900
100	300.0	300.0	0.3333	900
Standard Preparation				
Conc. (%)		**Conc. of Pregabalin WS (mg/ml)**	**Volume Pipetted from Stock St Solution (ml)**	**Volumetric Flask Vol. (ml)**
50		0.1666	1	20
75		0.2500	15	200
100		0.3333	5	50
: Note: Dilute to volume of the Volumetric flask with dissolution medium.** *: Use Stock Standard Solution (3.333 mg/ml).**				

- Analysis of Spiked Samples
- After completion of Dissolution Test (after 30 minutes) filter a portion of dissolution solution from each vessel and take 10 ml of the filtrate to 25 ml with the dissolution medium and mix, then analyze collected samples according to the test method of analysis for Pregabalin Tablets.
- Calculate the recovery data for each determination; calculate the average of recovery data and the RSD for each level.
- Verify that the mean recovery of the assay should be within 100±5.0% at each concentration over the range of 50–100% of the nominal concentration.

4.11.2.4 Robustness / Dissolution Tester Conditions Variation:

Preparation of Spiked Dissolution Samples:

Transfer an accurately weighed 300mg of Pregabalin to the dissolution vessel and add weighed quantity (300mg) of Pregabalin Tablets placebo, complete to volume by add 900-ml of dissolution medium to the vessel.

Variation of Test Method Parameters:

- Detection Wavelength: Variation of Detection wavelength to 212 nm instead of 210 nm.
- Flow rate: Variation of Flow rate to 1.6 ml/min instead of 1.5 ml/min.

Variation of Dissolution Parameters:

- R.P.M: Variation of the RPM to 48rpm instead of 50rpm.
  After completion of the dissolution test filter a portion of solution from each vessel and analyze according to the test method of analysis for dissolution determination of dissolved ***Pregabalin WS*** in ***Pregabalin Tablets.***
- Print out the results.

Data Analysis & Acceptance Criteria:

Verify that the influence of variation in method parameters and dissolution conditions are within the previously acceptance criteria, the parameter said to be within the methods robustness range.

4.11.2.5 Stability of Standard and Sample Solutions:

Preparation of Sample Solutions:

- Transfer about 66.6 mg of placebo of Pregabalin Tablets and add 20-ml from Stock Standard solution (0.3333 mg/ml of Pregabalin WS) to a 200 ml volumetric flask. Dissolve with the aid of sonication in dissolution medium to volume, and mix.
- Analyze the prepared standard and sample solutions over three periods (Zero Time, after 3hr and after 24hr at a storage at room temperature) according to the test method of analysis for dissolution test of ***Pregabalin*** in ***Pregabalin Tablets.***
- Print out the Results.

Data Analysis and Acceptance Criteria: The acceptable range for standard and sample solutions stability is typically between 98%-102% compared with the initial analysis of the standard and /sample solutions.

4.11.2.6 Specificity/Placebo Interference

Purpose: The purpose of this test is to demonstrate that the results are not unduly affected by placebo constituents.

Procedure:

Placebo Preparation: Prepare placebo of Pregabalin Tablets according to the formulation procedure of Pregabalin Tablets without addition of active ingredient.

Standard Solution Preparation: Use the nominal standard solution for dissolution test.

Nominal Placebo Solution Preparation: Dissolve 250 mg of Placebo in 900 ml of Dissolution Medium at 37^{o}C. Filter portion of solution using 0.2 micron filter.

- Inject the standard and sample solutions in the HPLC and record the results.
- Determine the interference of placebo using the following formula:

$$\textbf{Interference \% } = 100*C*(A_P/A_{St})*V/L$$

Where,

C: is the concentration, in mg per ml, of the standard
AP: is the absorbance of the placebo
A_{St}: is the absorbance of the standard
V: is the volume, in ml, of the medium
L: is the label claim, in mg of the product

Acceptance Criteria: The interference of Placebo should not exceed 2%.

4.11.2.7 Ruggedness (Intermediate Precision):

Procedure:

Standard Solution: Use the nominal standard solution (0.3333 mg/ml of Pregabalin WS) .

This test was performed by analysis of the nominal standard solution by different analyst and on different days (matrix design). Inject the working standard solution at nominal concentration ***(0.3333 mg/ml of Pregabalin WS)*** using Chromatographic HPLC.

Calculate the relative standard deviations (RSD) of the replicate readings.

Acceptance Criteria: Verify that, the Relative Standard Deviation for the replicate readings is not more than 5.0%.

Part V Results and Discussion

5.0 Results and Discussion

Summaries of the analysis results, Validation, and discussions are presented in this chapter:

5.1 Selection of Pregabalin API

Pregabalin was tested and analyzed according to its manufacturer test methods. All results are found to be within limits and conform to specifications, as illustrated in table 20.

Table 22: Characteristics of Pregabalin

Test	Manufacturer Limits	Results
Assay	**98.0-102.0% (on anhydrous bases)**	**98.6%**
Characteristics	**White Crystalline powder**	**White Crystalline powder**
Particle size **D (90)** **D (50)** **D (10)**	**For Information**	**812µm** **72µm** **7µm**
Identification	**(IR) conform to standard**	**conform to standard**
Water Content	**NMT 0.50%**	**0.05%**
Residue on ignition	**NMT 0.10%**	**0.00%**
Specific Optical Rotation (ODB)	**+10.0 to +12.0**	**+11.8**
Bromide content	**Less than 50 ppm**	**< 50ppm**
Heavy Metals	**Less than 10ppm**	**< 10ppm**
Related Substances **Impurity- III** **Impurity- IV** **Any other impurity** **Total impurities**	 **NMT 0.15%** **NMT 0.15%** **NMT 0.10%** **NMT 0.50%**	 **BDL** **BDL** **BDL** **BDL**

The COAs of excipients are attached in Appendix 1.

5.2 Compatibility Study

Spectra obtained from pure Pregabalin were found C-H from Alkane part, C-N and N-H from Amine group and O-C and O-H from Carboxyl group.

As obtained in the next spectroscopy:

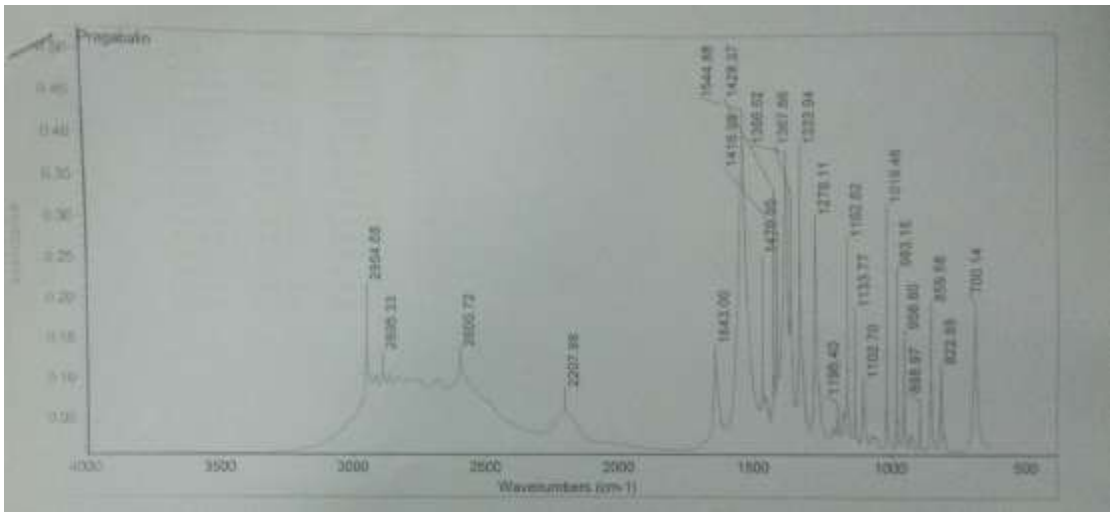

Figure 20: Pregabalin pure Spectroscopy

The results of previous spectroscopy is summarized and tabulated in Table 22.

Table 23: Pregabalin Function group using FTIR results.

F.G	FTIR Range	Pregabalin pure
Alkane (C-H)s	2850-3000 2or3 bands	2954.68
Alkane (C-H)s	1000-1250	2895.33
Amine (C-N)s	1550-1650	1162.62
Amine (N-H)b	1210-1320	1643
Carboxylic acid (O-C)s	2500-3300	1278.11
Carboxylic acid (O-H)s	2500-3300	2600.72

In Case of Mixture of Magnesium Stearate and Pregabalin

FT-IR spectroscopy of mixture shows very close spectra of the pure component as appears in the next spectra

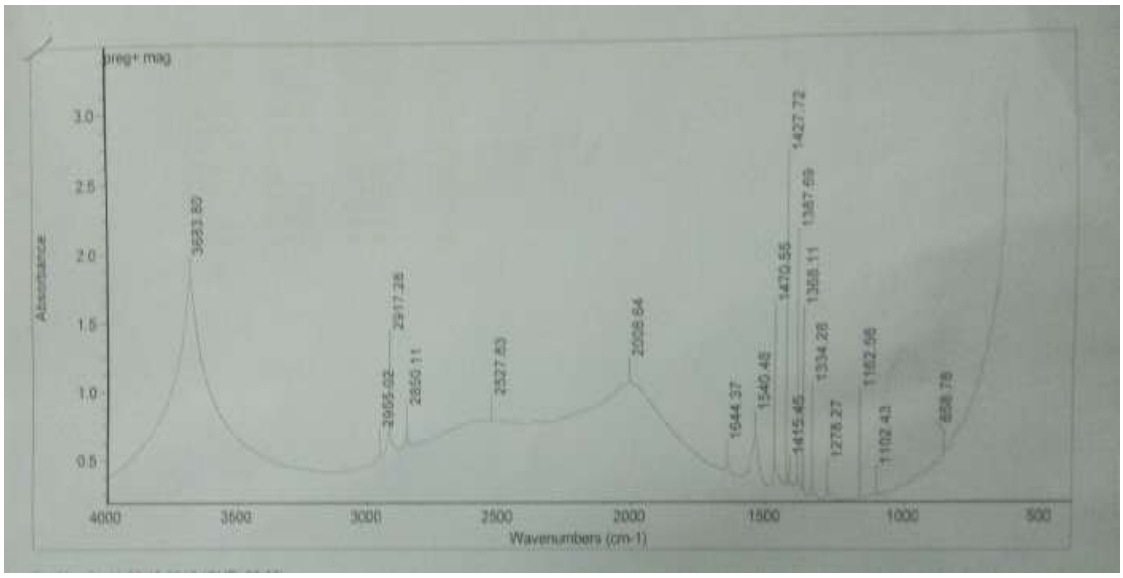

Figure 21: Spectroscopy of Mixture of Magnesium Stearate and Pregabalin

The previous spectra show that the main bond in Pregabalin and the Magnesium Stearate mixture is very similar as shown in the next table

Table 24: Pregabalin Function group using FTIR results

F.G	FTIR Range	Pregabalin pure	Pregabalin, Mg. Stearate (1:1)
Alkane (C-H)s	2850-3000 2or3 bands	2954.68	2955.02
Alkane (C-H)s	1000-1250	2895.33	2917.28
Amine (C-N)s	1550-1650	1162.62	1162.56
Amine (N-H)b	1210-1320	1643	1644.37
Carboxylic acid (O-C)s	2500-3300	1278.11	1278.2
Carboxylic acid (O-H)s	2500-3300	2600.72	2527.83

Previous results indicate that there is no interference between Pregabalin and magnesium stearate (Table-21). There was no major change in peaks of Alkane (C-H), Amine (C-N), Amine (N-H) or Carboxylic Acid (O-C) in reference to the observed value of pure Pregabalin. . Slightly shifting in Carboxylic Acid (O-H) may refer to the presence of water in Magnesium Stearate.

In Case of Mixture of Talc and Pregabalin

FT-IR spectroscopy of mixture shows very close spectra of the pure component as appears in the next spectra

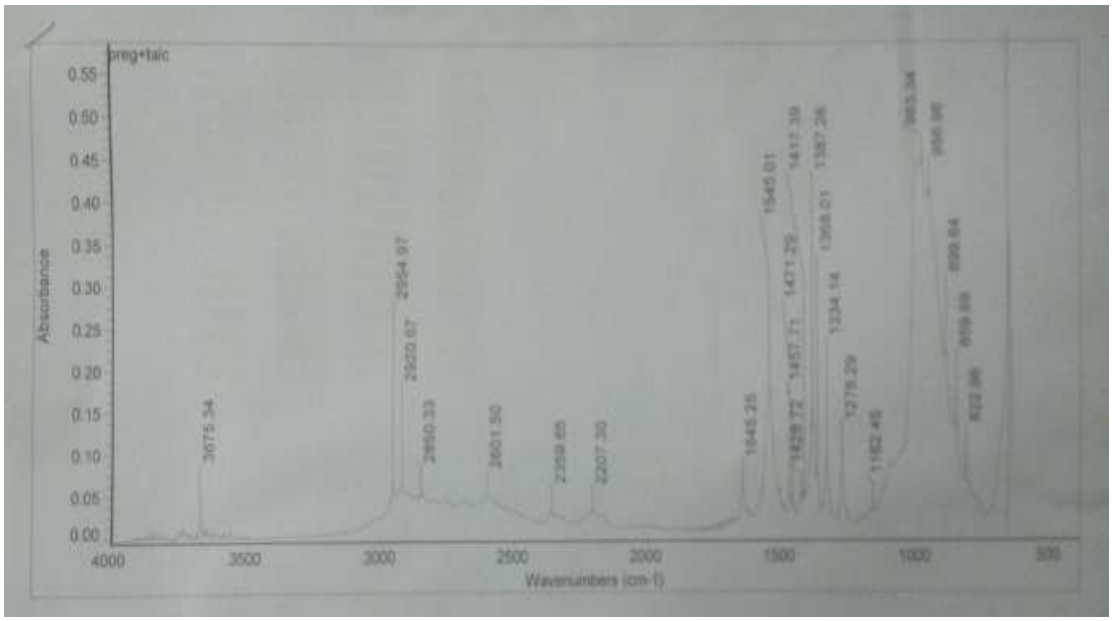

Figure 22: Spectroscopy of Mixture of Talc and Pregabalin

The previous spectra show that the main bond in pure Pregabalin and in the mixture is very similar as shown in the next table

Table 25: Pregabalin Function group using FTIR results

F.G	FTIR Range	Pregabalin pure	Pregabalin, Talc (1:1)
Alkane (C-H)s	2850-3000	2954.68	2954.97
Alkane (C-H)s	2850-3000	2895.33	2920.67
Amine (C-N)s	1550-1650	1162.62	11362.45
Amine (N-H)b	1210-1320	1643	1645.25
Carboxylic acid (O-C)s	2500-3300	1278.11	1278.29
Carboxylic acid (O-H)s	2500-3300	2600.72	2601.5

Previous results indicate that there is no interference between Pregabalin and Talc (Table-22). There was no major change in peaks of Alkane (C-H), Amine (C-N), Amine (N-H), Carboxylic Acid (O-C) and Carboxylic Acid (O-H) in reference to the observed value of pure Pregabalin.

In Case of Mixture of Microcrystalline Cellulose (Avicel) and Pregabalin

FT-IR spectroscopy of mixture shows very close spectra of the pure component as appears in the next spectra

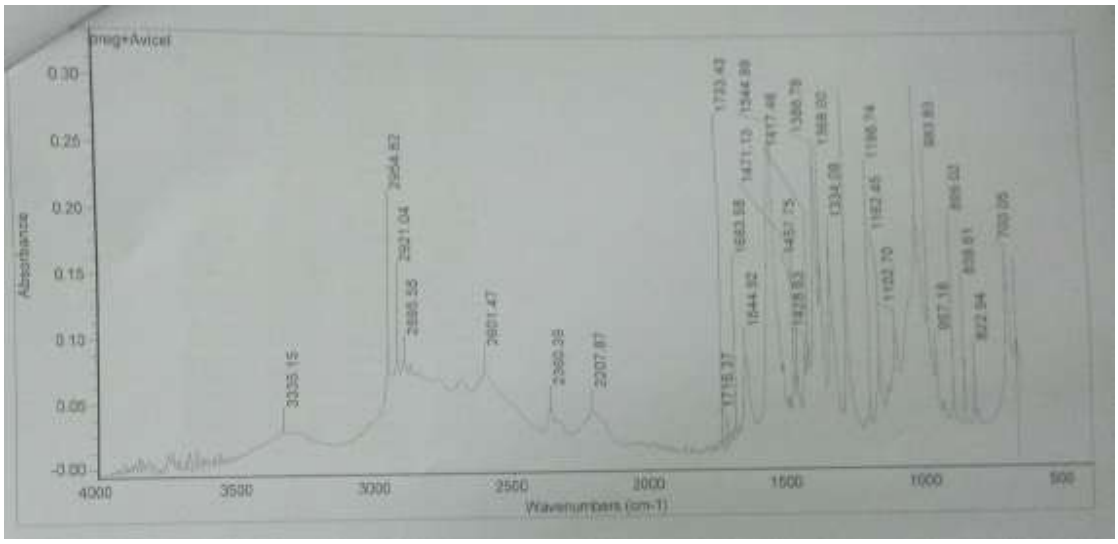

Figure 23: Spectroscopy of Mixture of Avicel and Pregabalin

The previous spectra shows that the main bond in pure Pregabalin and in the binary mixture is very similar as shown in the next table.

Table 26: Pregabalin Function group using FTIR results

F.G	FTIR Range	Pregabalin pure	Pregabalin, Avicel PH 102 (1:1)
Alkane (C-H)s	2850-3000 2or3 bands	2954.68	2954.82
Alkane (C-H)s	1000-1250	2895.33	2895.58
Amine (C-N)s	1550-1650	1162.62	1162.45
Amine (N-H)b	1210-1320	1643	1644.92
Carboxylic acid (O-C)s	2500-3300	1278.11	1278.2
Carboxylic acid (O-H)s	2500-3300	2600.72	2601.47

Previous results indicate that there is no interference between Pregabalin and Avicel (Table-23). There was no major change in peaks of Alkane (C-H), Amine (C-N), Amine (N-H), Carboxylic Acid (O-C) and Carboxylic Acid (O-H) in reference to the observed value of pure Pregabalin.

In Case of Mixture of Starch and Pregabalin

FT-IR spectroscopy of mixture shows very close spectra of the pure component as appears in the next spectra.

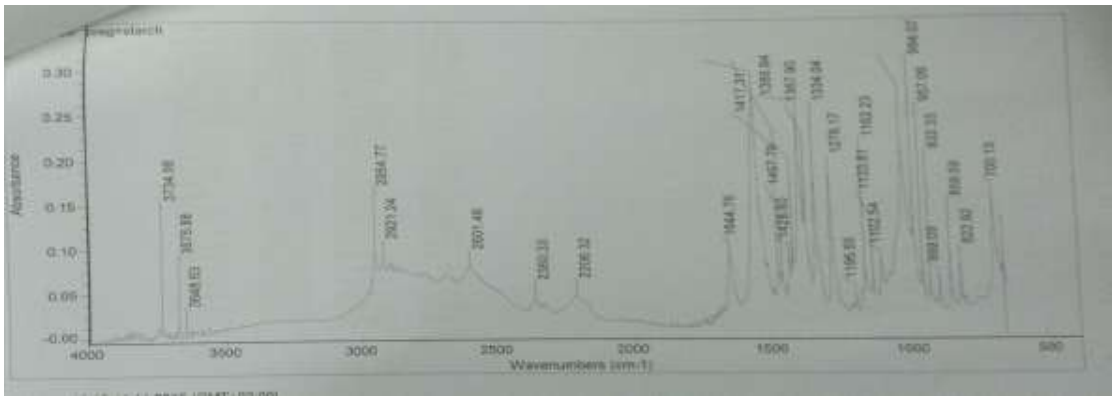

Figure 24: Spectroscopy of Mixture of Starch and Pregabalin

The previous spectra show that the main bond in pure Pregabalin and in the binary mixture is very similar as shown in the next table.

Table 27: Pregabalin Function group using FTIR results

F.G	FTIR Range	Pregabalin pure	Pregabalin, Starch 1500 (1:1)
Alkane (C-H)s	2850-3000 2or3 bands	2954.68	2954.77
Alkane (C-H)s	1000-1250	2895.33	2921.24
Amine (C-N)s	1550-1650	1162.62	1162.23
Amine (N-H)b	1210-1320	1643	1644.76
Carboxylic acid (O-C)s	2500-3300	1278.11	1278.17
Carboxylic acid (O-H)s	2500-3300	2600.72	2601.46

Previous results indicate that there is no interference between Pregabalin and Starch (Table-24). There was no major change in peaks of Alkane (C-H), Amine (C-N), Amine (N-H), Carboxylic Acid (O-C) and Carboxylic Acid (O-H) in reference to the observed value of pure Pregabalin.

As a summary the data obtained from FTIR Compatibility study clearly indicates insignificant changes in spectra obtained from physical mixture of Pregabalin and selected excipients. And all results of Compatibility study indicate that there is no interference between Pregabalin and other excipients as appeared in Figures (20-24) and Tables (18-22). There was no major change in peaks of Alkane (C-H), Amine (C-N), Amine (N-H), Carboxylic Acid (O-C) and Carboxylic Acid (O-H) in reference to the observed value of pure Pregabalin.

5.3 Analysis of Pre-formulation formulae

The results of physical analysis of Pregabalin blends are shown in Table 28; the tablets physical characteristics are summarized in table 29.

Table 28: Pre-formulation Powder Blends Physical Characteristics

Pre-formulation properties				
Formula	**Tapped Density**	**Untapped Density**	**Flowability "Hausner ratio"**	**Carr`s index (compressibility)**
F-1	-----	----	----	----
F-2	0.658	0.531	1.24	19.3%
F-3	0.663	0.529	1.30	20.2%
F-4	0.660	0.538	1.23	18.5%
F-5	**0.650**	**0.522**	1.25	**19.7%**
F-6	0.648	0.523	1.24	19.3%
F-7	0.641	0.519	1.24	19.0%
F-8*	0.650	0.522	1.25	19.7%
*** F-5 and F-8 are the same formula but compressed under different parameter and different punches.**				

Table 29: Pre-formulation Compressed Tablets Physical Characteristics

Compressed Tablet properties						
Formula	**Hardness (Kgf)**	**Friability (w/w) %**	**Average weight (mg)**	**Thickness (mm)**	**Dis. Time (seconds)**	**Tablet Appearance**
F-1	0.0	100%	---	---	---	No tablet was formed
F-2	5.5-7.9	0.9%	579	4.59	18 sec.	Capping
F-3	2.1-4.5	1.3%	540	4.60	15 sec.	Capping
F-4	3.5-5.0	0.9%	652	4.32	25 sec.	Uniform and Smooth
F-5	**9.0-12.5**	**0.3%**	**662**	**4.95**	**60 sec.**	**Uniform and Smooth**
F-6	3.5-4.8	0.8%	668	4.81	27 sec.	Uniform and Smooth
F-7	4.5-5.5	0.6%	663	6.89	22 sec.	Capping
F-8*	8.0-10.6	0.4%	660	4.88	54 sec.	Uniform and Smooth
*** F-5 and F-8 are the same formula but compressed under different parameter and different punches.**						

From the results shown in tables (28, 29), it is obvious that all powder blends have fair to passable flowability; Pregabalin with lubricant alone (F-1) has poor compaction characteristics. It is clear that the increase of filler/binder percentage from 33% to 45% (F-2 → F-5) improved the compaction characters of powder mixture. The use of starch maize as disintegrant caused tablets to cap (F-2, F-3, F-7), while use of pregelatinized starch instead of maize starch, lead get smooth and intact tablets (F-5, F-6); this may be due to the binding effect of pregelatinized starch in direct compression methods.

Furthermore, F3 and F4 formulae have low disintegration times (15 to 25 seconds) and high friability values (about or more than 1.0%).

Comparison between Formulae F-5 and F-6 as the candidate ones, we excluded F-6 from further study, because of the low hardness and disintegration time of compressed tablets that may cause to partially disintegration in the mouth during swallow.

Formula F-5 has the best physical and mechanical properties and promising candidate for further studies.

Formula (F-5) as optimized formula was selected according to different parameters from pre-formulation parameter until to obtain tablet properties as Flowability, hardness, disintegration, tablet appearance, for that we can transfer to the implementation and evaluation stages as shown in the next section.

5.4 Implementation of Selected Formula

Pregabalin tablets, with two strengths 300mg tablet and 75mg tablet were prepared using the same component ratio of selected formula (F5).

About 2000 tablets were compressed and coated from Pregabalin 300mg strength, and about 8000 tablets were compressed and produced from Pregabalin 75mg strength.

The same general manufacturing procedure was applied.

5.5 Selection of packaging raw material

The coated tablets were filled in PVC-Alum blisters, and all evaluation tests, such as sealing test and stability study were performed to evaluate the efficiency of PVC Blister as packaging material for Pregabalin tablet.

5.6 Pregabalin Tablets Evaluation

The two implemented batches of Pregabalin tablets (75mg and 300mg) were tested according to the developed and validated test methods. All results are summarized as the following:

General Appearance:

The general appearance (color, shape, dimension and appearance) of the formulated tablets was inspected and tested; all results were recorded and tabulated in the next tables.

Table 30: Evaluation results - Physical properties and general appearance

Tests	Acceptance Criteria	Results Pre. 300mg	Results Pre. 75mg
Color & Odor	White / off-white and Characteristic Odor	White / off-white	White / off-white
Shape	Oblong	Oblong	Oblong
Width	9.1 (+/- 0.2) mm (for 300mg strength) 5.1 (+/- 0.2) mm (for 75mg strength)	9.1	5.1
Thickness	4.7 (+/- 0.3) mm (for 300mg strength) 3.1 (+/- 0.2) mm (for 75mg strength)	4.7	3.1
Appearance of Coat	Smooth & Homogeneous	Smooth & Homogeneous	Smooth & Homogeneous

Table 31: Evaluation results – Tablet Thickness

Tab. No.	1	2	3	4	5	6	7	8	9	10	Avg.	SD
Per. 300mg	4.7	4.7	4.7	4.7	4.7	4.7	4.7	4.7	4.7	4.7	4.7	0.0
Pre. 75mg	3.1	3.1	3.1	3.1	3.1	3.1	3.1	3.1	3.1	3.1	3.1	0.0

Hardness:

The force required to cause fracture was recorded. It is repeated for 10 tablets and the measured values are averaged, all results are within limits as tabulated in table (32).

Table 32: Hardness Results

Tab. No.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**Avg.**	**SD**
300mg	9.0	9.6	10.2	9.9	10.5	11.0	11.2	9.6	10.2	9.1	10.0	0.7
75mg	8.3	9.6	8.8	9.5	9.9	8.4	10.2	7.9	9.8	9.5	9.0	0.7
Acceptance Criteria: NLT 5.0 Kgf												

Friability

The friability results are shown in Table 33, all results are within limits.

Table 33: Friability Results

Test	Acceptance Criteria	Results Pre. 300mg	Results Pre. 75mg
Friability	NMT 1.0%, 100 rounds	0.3%	0.1%

Uniformity of dosage units:

Results of uniformity of dosage units are shown in Tables 34, 35.

Table 34: Uniformity of dosage units result for Pregabalin 300 mg Tablets

Product	**Pregabalin 75 mg Tablets**
BN.	150915
Exp. Date	09/2017
Theoretical weight	167.5 mg

Parameter & Tablet No.	Weight (mg)	Content of Pregabalin in each unit expressed as a percent to label claim [%]
1	170.4	101.7
2	176.7	105.5
3	166.5	99.4
4	175.5	104.8
5	166.6	99.5
6	171.1	102.1
7	165.1	98.6
8	175.3	104.7
9	172.0	102.7
10	172.3	102.9
X (Average)	171.2	102.2
T (Target test sample amount at time of manufacture)		**100.0**
SD (Standard Deviation)		**2.3**
M		**101.5**
k		
AV (Acceptance Value) where (AV = | M-X+k.SD) NMT 15		**6.2**
Result: Pass According to USP, No need to other stage.		

Table 35: Uniformity of dosage units result for Pregabalin 75 mg Tablets

Product	**Pregabalin 300 mg Tablets**
BN.	150914
Exp. Date	09/2017
Theoretical weight	670.0 mg

Parameter & Tablet No.	Weight (mg)	Content of Pregabalin in each unit expressed as a percent to label claim [%]
1	677.9	101.2
2	684.7	102.2
3	678.3	101.2
4	703.2	105.0
5	686.0	102.4
6	682.7	101.9
7	685.9	102.4
8	632.9	94.5
9	675.4	100.8
10	687.9	102.7
$\overline{X}$ (Average)	679.5	101.4
T (Target test sample amount at time of manufacture)		**100.0**
SD (Standard Deviation)		**2.7**
M		**101.4**
k		**2.4**
AV (Acceptance Value) where (AV = | M-X+k.SD) NMT 15		**6.5**
Result: Pass According to USP, No need to other stage.		

All results of Acceptance Values (AV) are within the acceptable limits.

Disintegration Test:

All Pregabalin 300mg and Pregabalin 75mg tablets disintegrate within two minutes.

Acceptance Criteria: All tablets should disintegrate within 15 minutes.

Dissolution for routine testing:

Dissolution rate studies: The dissolution rate was carried out by using USP Type II (paddle type) dissolution apparatus.

Table 36: Dissolution results

Batch\ Vessel No.	**1**	**2**	**3**	**4**	**5**	**6**	**Average**	**SD**
F-5, 300	91.4	91.1	105.5	91.4	96.6	90.9	94.5%	5.8
F-5, 75	104.6	101.6	105.9	101.8	100.7	101.1	102.6%	2.1
Acceptance Criteria	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes							

Assay and Related Tests

Table 37: Results of chemical tests for Pregabalin Finished products

	Acceptance Criteria	Results F-5, 300	Results F-5, 75
Assay			
-Pregabalin	90.0-110.0 %	100.9%	99.0%
Related Substances:			
-Individual impurities. **-Total Impurities.**	NMT 0.2% NMT 1.0%	BDL BDL	BDL BDL

5.7 Biowaiver Study

5.7.1 Results of Dissolution Profiles

Dissolution profiles were performed for both implemented batches (Pregabalin 300mg and 75mg) versus reference product (Lyrica 300mg and 75mg Capsules) in different dissolution media: 0.06N HCl, Acetate Buffer pH 4.5 and Phosphate Buffer pH 6.8. The data are reported in Tables (36-41), and the summary of results is illustrated in tables (41-43) and figures (25-30).

5.7.1.1 Dissolution Profile data for Pregabalin 75mg tablets and Lyrica 75mg Capsules

Table 38: Dissolution Profile data for Pregabalin 75mg capsules and tablets in 0.06N HCl

Test conditions:							
Medium	*0.06N HCl*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 75mg *Dissolution Media 0.06N HCl*					
Pregabalin STD	***Dissolution time***	***Response Area (F5)***	***Dissolution results (F5)***	***Response Area (Lyrica)***	***Dissolution results (Lyrica)***
F5 STD	**10**	96126	**94.1%**	94580	**96.5%**
102480	**10**	97061	**95.0%**	99440	**101.5%**
102136	**10**	91602	**89.7%**	96215	**98.2%**
102184	**10**	103322	**101.2%**	94753	**96.7%**
101677	**10**	81011	**79.3%**	95658	**97.6%**
Avg. 102119	**10**	98858	**96.8%**	97207	**99.2%**
	15	102932	**100.8%**	96092	**98.1%**
	15	105060	**102.9%**	99526	**101.6%**
	15	104983	**102.8%**	98553	**100.6%**
Lyrica STD	**15**	102691	**100.6%**	99209	**101.3%**
97980	**15**	95776	**93.8%**	98887	**100.9%**
98198	**15**	102849	**100.7%**	97585	**99.6%**
97936	**30**	106771	**104.6%**	98323	**100.4%**
97774	**30**	103499	**101.4%**	98221	**100.3%**
Avg. 97972	**30**	108112	**105.9%**	98796	**100.8%**
	30	103958	**101.8%**	99096	**101.1%**
	30	102881	**100.7%**	98220	**100.3%**
	30	103255	**101.1%**	97923	**99.9%**
	45	106841	**104.6%**	98218	**100.3%**
	45	104793	**102.6%**	98335	**100.4%**
	45	108501	**106.2%**	98923	**101.0%**
	45	106891	**104.7%**	98191	**100.2%**
	45	102215	**100.1%**	98033	**100.1%**
	45	103753	**101.6%**	97824	**99.8%**

Table 39: Dissolution Profile data for Pregabalin 75mg capsules and tablets in Acetate Buffer pH 4.5

Test conditions:							
Medium	*Acetate Buffer pH4.5*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 75mg *Dissolution Media Acetate Buffer PH4.5*					
Pregabalin STD	***Dissolution time***	***Response Area (F5)***	***Dissolution results (F5)***	***Response Area (Lyrica)***	***Dissolution results (Lyrica)***
F5 STD	**10**	80314	**78.4%**	159638	**88.3%**
101297	**10**	77204	**75.4%**	156880	**86.8%**
104407	**10**	96166	**93.9%**	166399	**92.1%**
102310	**10**	79290	**77.4%**	172978	**95.7%**
101590	**10**	86953	**84.9%**	178595	**98.8%**
Avg. 102401	**10**	87156	**85.1%**	163046	**90.2%**
	15	87386	**85.3%**	171049	**94.6%**
	15	97833	**95.5%**	171955	**95.1%**
	15	100722	**98.4%**	174482	**96.5%**
Lyrica STD	**15**	86738	**84.7%**	179724	**99.4%**
180069	**15**	96124	**93.9%**	182784	**101.1%**
179052	**15**	98772	**96.5%**	179810	**99.5%**
179750	**30**	98469	**96.2%**	180649	**99.9%**
Avg. 180740	**30**	104090	**101.6%**	181270	**100.3%**
	30	102147	**99.8%**	183250	**101.4%**
	30	100316	**98.0%**	180589	**99.9%**
	30	105930	**103.4%**	183674	**101.6%**
	30	103672	**101.2%**	183262	**101.4%**
	45	103799	**101.4%**	183592	**101.6%**
	45	103978	**101.5%**	181436	**100.4%**
	45	102626	**100.2%**	183233	**101.4%**
	45	105254	**102.8%**	180719	**100.0%**
	45	104607	**102.2%**	181888	**100.6%**
	45	104557	**102.1%**	182620	**101.0%**

Table 40: Dissolution Profile data for Pregabalin 75mg capsules and tablets in Phosphate Buffer pH 6.8

Test conditions:							
Medium	*Phosphate Buffer pH 6.8*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 75mg *Dissolution Media Phosphate Buffer PH6.8*					
Pregabalin STD	***Dissolution time***	***Response Area(F5)***	***Dissolution results (F5)***	***Response Area (Lyrica)***	***Dissolution results (Lyrica)***
F5 STD	**10**	78050	**76.2%**	166031	**91.7%**
101436	**10**	79647	**77.8%**	166712	**92.0%**
100926	**10**	97237	**95.0%**	144813	**80.0%**
102348	**10**	78937	**77.1%**	154295	**85.2%**
101584	**10**	80140	**78.3%**	164757	**91.0%**
Avg. 101574	**10**	80171	**78.3%**	160033	**88.4%**
	15	92169	**90.0%**	173643	**95.9%**
	15	91980	**89.8%**	178367	**98.5%**
	15	105657	**103.2%**	169718	**93.7%**
Lyrica STD	**15**	93369	**91.2%**	176982	**97.7%**
180161	**15**	93763	**91.6%**	173317	**95.7%**
180583	**15**	90767	**88.6%**	138328	**76.4%**
181974	**30**	103938	**101.5%**	185828	**102.6%**
181785	**30**	96320	**94.1%**	186048	**102.7%**
Avg. 181126	**30**	105603	**103.1%**	181037	**100.0%**
	30	104241	**101.8%**	181324	**100.1%**
	30	99349	**97.0%**	178660	**98.6%**
	30	97191	**94.9%**	179581	**99.1%**
	45	108806	**106.3%**	185855	**102.6%**
	45	96647	**94.4%**	185632	**102.5%**
	45	105715	**103.2%**	179929	**99.3%**
	45	108858	**106.3%**	182223	**100.6%**
	45	99186	**96.9%**	180907	**99.9%**
	45	98221	**95.9%**	181737	**100.3%**

5.7.1.1 Dissolution Profile data for Pregabalin 300mg tablets and Lyrica 300mg Capsules

Table 41: Dissolution Profile data for Pregabalin 300mg capsules and tablets in 0.06N HCl

Test conditions:							
Medium	*0.06N HCl*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 300mg *Dissolution Media 0.06N HCL*					
Pregabalin STD	***Dissolution time***	***Response Area (F5)***	***Dissolution Area (F5)***	***Response Area (Lyrica)***	***Dissolution results (Lyrica)***
F5 STD	**10**	304397	**73.9%**	279689	**95.9%**
410344	**10**	296660	**72.1%**	280675	**96.2%**
413031	**10**	378433	**91.9%**	272800	**93.5%**
411849	**10**	316900	**77.0%**	270292	**92.7%**
Avg. 411741	**10**	314696	**76.4%**	265325	**91.0%**
	10	303098	**73.6%**	264638	**90.7%**
	15	388086	**94.3%**	283916	**97.3%**
Lyrica STD	**15**	347478	**84.4%**	289622	**99.3%**
294033	**15**	393724	**95.6%**	279436	**95.8%**
290970	**15**	394912	**95.9%**	280878	**96.3%**
291015	**15**	373659	**90.8%**	282238	**96.8%**
290773	**15**	356618	**86.6%**	278267	**95.4%**
Avg. 291698	**30**	397631	**96.6%**	294800	**101.1%**
	30	412645	**100.2%**	291630	**100.0%**
	30	402771	**97.8%**	289387	**99.2%**
	30	402478	**97.8%**	290575	**99.6%**
	30	402273	**97.7%**	289988	**99.4%**
	30	413730	**100.5%**	291956	**100.1%**
	45	404212	**98.2%**	293076	**100.5%**
	45	416548	**101.2%**	291202	**99.8%**
	45	400797	**97.3%**	290963	**99.7%**
	45	402912	**97.9%**	290352	**99.5%**
	45	401106	**97.4%**	292947	**100.4%**
	45	415559	**100.9%**	291207	**99.8%**

Table 42: Dissolution Profile data for Pregabalin 300mg capsules and tablets in Acetate Buffer pH 4.5

Test conditions:							
Medium	*Acetate Buffer pH 4.5*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 300mg *Dissolution Media Acetate Buffer 4.5*					
Pregabalin STD	***Dissolution time***	***Response Area (F5, 300 mg)***	***Response Area (F5, 300 mg)***	***Response Area (Lyrica, 300 mg)***	***Dissolution results (Lyrica, 300 mg)***
F5 STD	**10**	288565	**69.9%**	648252	**91.0%**
412833	**10**	312608	**75.7%**	638509	**89.7%**
412987	**10**	261059	**63.2%**	577614	**81.1%**
414233	**10**	289999	**70.2%**	622463	**87.4%**
412188	**10**	303518	**73.5%**	576747	**81.0%**
Avg. 413060	**10**	341769	**82.7%**	618778	**86.9%**
	15	325157	**78.7%**	660780	**92.8%**
	15	365802	**88.6%**	672888	**94.5%**
	15	361339	**87.5%**	610981	**85.8%**
Lyrica STD	**15**	330846	**80.1%**	649315	**91.2%**
735809	**15**	364754	**88.3%**	608804	**85.5%**
733348	**15**	364878	**88.3%**	659182	**92.6%**
731075	**30**	375392	**90.9%**	689192	**96.8%**
648252	**30**	396953	**96.1%**	697620	**98.0%**
Avg. 712121	**30**	408413	**98.9%**	665478	**93.5%**
	30	374296	**90.6%**	696077	**97.7%**
	30	412544	**99.9%**	669864	**94.1%**
	30	397018	**96.1%**	705375	**99.1%**
	45	397647	**96.3%**	704977	**99.0%**
	45	397886	**96.3%**	704834	**99.0%**
	45	410425	**99.4%**	686731	**96.4%**
	45	396555	**96.0%**	706567	**99.2%**
	45	411990	**99.7%**	682073	**95.8%**
	45	400770	**97.0%**	699422	**98.2%**

Table 43: Dissolution Profile data for Pregabalin 300mg capsules and tablets in Phosphate Buffer pH 6.8

Test conditions:							
Medium	*Phosphate Buffer pH 6.8*	**Volume**	900 mL	**Apparatus**	II (Paddle)	**Rotational Speed:**	50 RPM

Data of dissolution profile for Pregabalin 300mg *Dissolution Media Phosphate Buffer 6.8*					
Pregabalin STD	***Dissolution time***	***Response Area (F5, 300 mg)***	***Response Area (F5, 300 mg)***	***Response Area (Lyrica, 300 mg)***	***Dissolution results (Lyrica, 300 mg)***
F5 STD	**10**	285586	**69.1%**	612896	**83.2%**
412833	**10**	276586	**67.0%**	643429	**87.4%**
412987	**10**	302019	**73.1%**	645995	**87.7%**
414233	**10**	284134	**68.8%**	642968	**87.3%**
412188	**10**	256454	**62.1%**	622618	**84.6%**
Avg. 413060	**10**	254285	**61.6%**	659358	**89.6%**
	15	302760	**73.3%**	648352	**88.1%**
	15	335727	**81.3%**	690028	**93.7%**
Lyrica STD	**15**	372147	**90.1%**	662458	**90.0%**
739033	**15**	327984	**79.4%**	689209	**93.6%**
740162	**15**	338026	**81.8%**	667207	**90.6%**
734646	**15**	336058	**81.4%**	682886	**92.8%**
731153	**30**	377440	**91.4%**	682704	**92.7%**
Avg. 736248	**30**	376149	**91.1%**	708113	**96.2%**
	30	435745	**105.5%**	706277	**95.9%**
	30	377603	**91.4%**	704593	**95.7%**
	30	398975	**96.6%**	704914	**95.7%**
	30	375443	**90.9%**	713632	**96.9%**
	45	399793	**96.8%**	694198	**94.3%**
	45	402200	**97.4%**	709030	**96.3%**
	45	440199	**106.6%**	710550	**96.5%**
	45	401996	**97.3%**	703948	**95.6%**
	45	408589	**98.9%**	708811	**96.3%**
	45	400829	**97.0%**	715767	**97.2%**

5.7.2 Summaries of dissolution profiles

The dissolution results were calculated from the data obtained in the previous section 5.7.1. The averages and the relative standard deviations at each time interval were calculated and tabulated in the following tables.

5.7.2.1 For Pregabalin 75mg Tablets and Capsules

➢ ***Dissolution Profile in 0.06N HCl***

Table 44: Dissolution profile results of Pregabalin 75mg Tablets vs Lyrica 75mg Capsules in *0.06N HCl*

Time	Dissolution medium 0.06N HCl			
	Pregabalin 75mg Tablet (F5)		**Lyrica Capsules 75mg**	
	Batch No.: F5, 75 n=6		Batch No.: n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	**92.7%**	6.9 (7.4%)	**98.3%**	1.9 (1.9%)
15	**100.3%**	3.3 (3.3%)	**100.3%**	1.3 (1.3%)
30	**102.6%**	2.1 (2.0%)	**100.5%**	0.5 (0.5%)
45	**103.3%**	2.3 (2.2%)	**100.3%**	0.4 (0.4%)

The percent released for both Test and Reference products in 0.06N exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar.

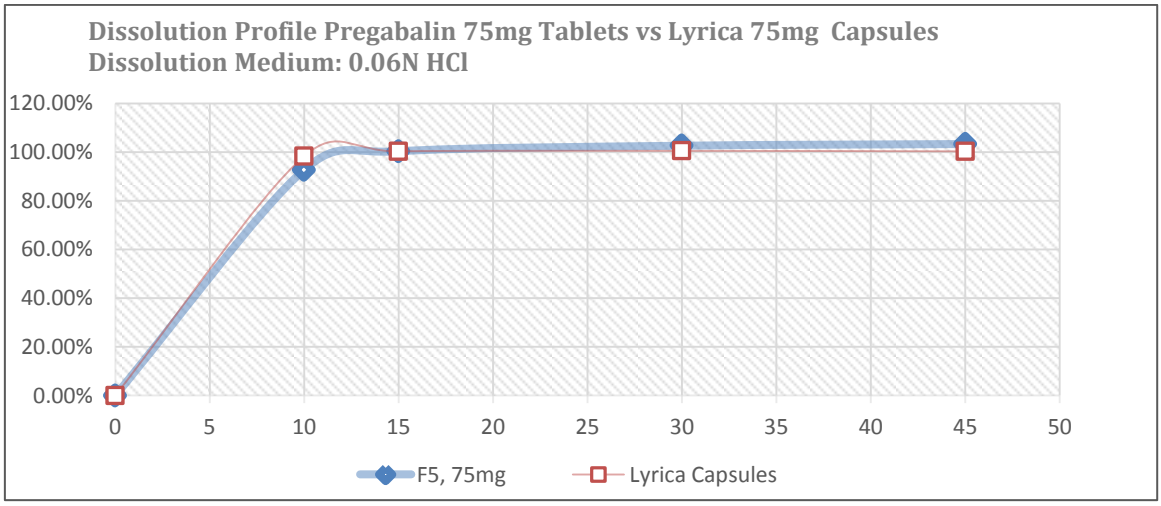

Figure 25: Dissolution profile of Pregabalin 75mg Tablets versus Lyrica 75 Capsules in 0.06N HCl

➢ ***Dissolution Profile in Acetate buffer pH 4.5***

Table 45: Dissolution profile results of Pregabalin 75mg Tablets vs Lyrica 75mg Capsules in Acetate buffer pH 4.5

Time	Dissolution medium *Acetate buffer pH 4.5*			
	Pregabalin 75mg Tablet (F5)		**Lyrica Capsules 75mg**	
	Batch No.: F5, 75 n=6		Batch No.: n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	**82.50%**	6.9 (8.4%)	**92.00%**	4.6 (5.0%)
15	**92.40%**	5.9 (6.4%)	**97.70%**	2.7 (2.8%)
30	**100.00%**	2.6 (2.6%)	**100.80%**	0.8 (0.8%)
45	**101.70%**	0.9 (0.9%)	**100.70%**	0.6 (0.6%)

The percent released for both Test and Reference products in Acetate buffer pH 4.5 exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar.

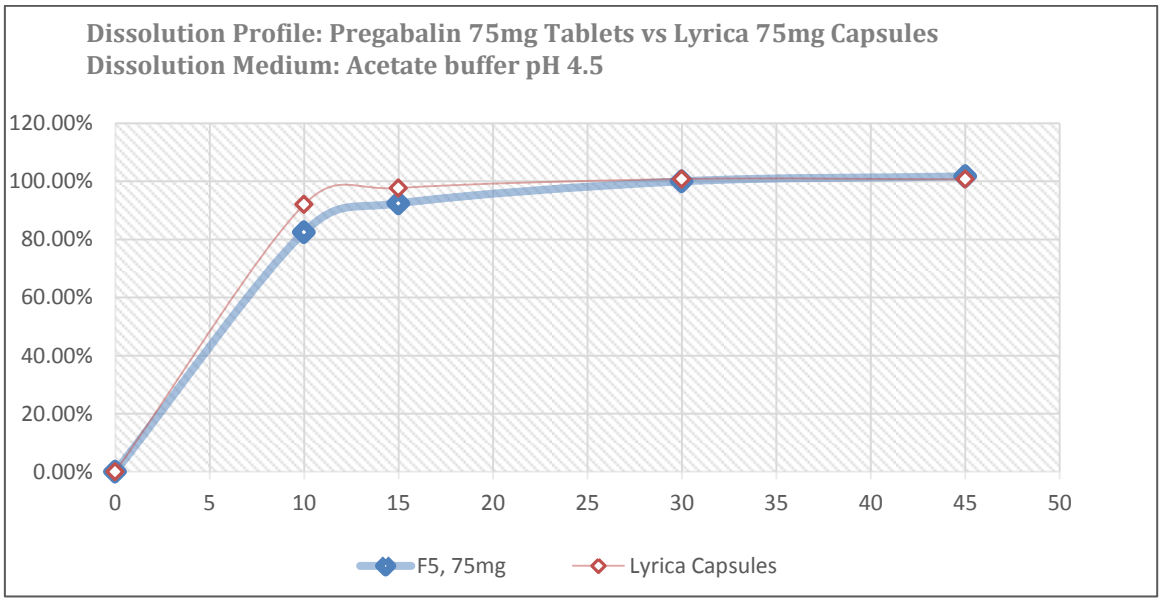

Figure 26: Dissolution profile of Pregabalin 75mg Tablets versus Lyrica 75 Capsules in Acetate buffer pH 4.5

➢ ***Dissolution profile in Phosphate buffer pH 6.8***

Table 46: Dissolution profile results (Pregabalin 75mg Tablets vs. Lyrica 75mg Capsules) in Phosphate buffer pH 6.8

Time	Dissolution medium Phosphate buffer pH 6.8			
	Pregabalin 75mg Tablet (F5)		**Lyrica Capsules 75mg**	
	Batch No.: F5, 75 n=6		Batch No.: n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	82.50%	7.2 (8.7%)	92.00%	4.7 (5.1%)
15	92.40%	5.4 (5.8%)	97.70%	8.3 (8.5%)
30	100.00%	3.9 (3.9%)	100.80%	1.7 (1.7%)
45	101.70%	5.4 (5.3%)	100.70%	1.4 (1.4%)

The percent released for both Test and Reference products in Phosphate buffer pH 6.8 exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar.

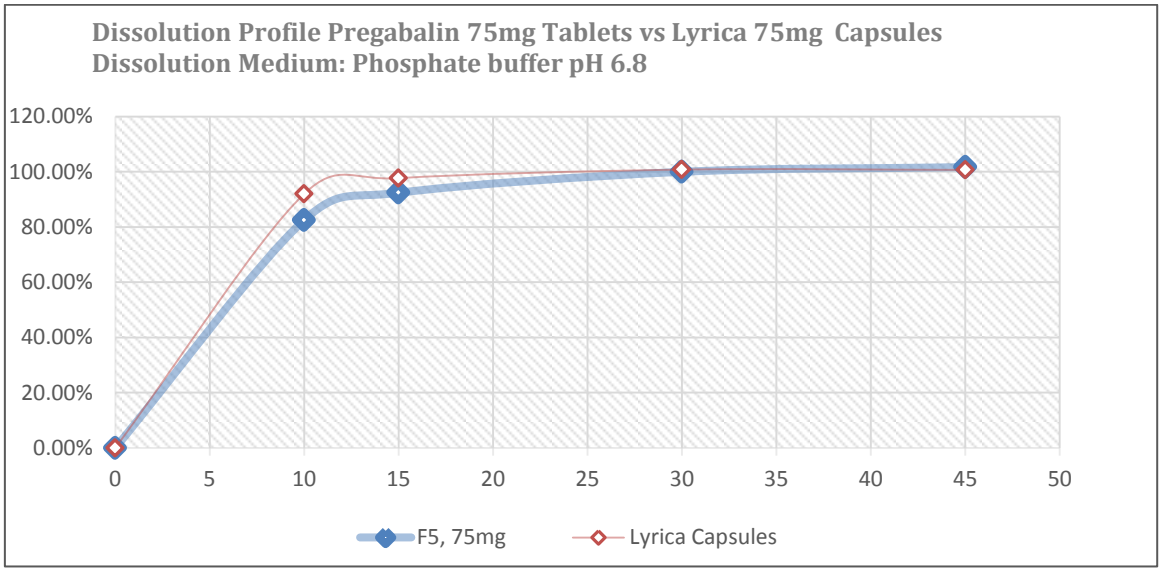

Figure 27: Dissolution profile of Pregabalin 75mg Tablets versus Lyrica 75 Capsules in Phosphate buffer pH 6.8

5.7.2.2 For Pregabalin 300mg Tablets and Capsules

- ***Dissolution Profile in 0.06N HCl***

Table 47: Dissolution profile results: Pregabalin 300mg Tablets vs Lyrica 300mg Capsules in 0.06N HCl)

Time	Dissolution medium 0.06 HCl			
	Pregabalin 300mg Tablet (F5)		**Lyrica Capsules 300mg**	
	Batch No.: F5 300 n=6		Batch No.: n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	77.50%	7.3 (9.4%)	93.30%	4.2 (4.5%)
15	91.30%	4.5 (4.9%)	96.80%	3.8 (3.9%)
30	98.40%	1.5 (1.5%)	99.90%	2.3 (2.3%)
45	98.80%	1.8 (1.8%)	100.00%	1.4 (1.4%)

The percent released for both Test and Reference products in 0.06N HCl exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar

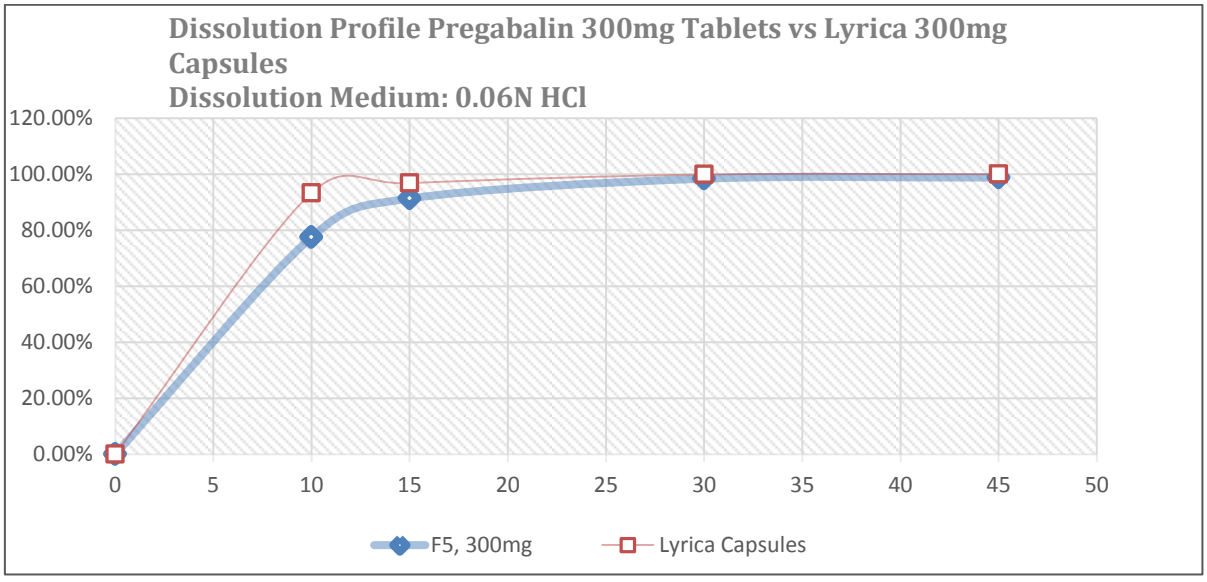

Figure 28: Dissolution profile of Pregabalin 300mg Tablets versus Lyrica 300 Capsules in 0.06N HCl

➢ ***Dissolution Profile in Acetate buffer pH 4.5***

Table 48: Dissolution profile results (Pregabalin 300mg Tablets versus Lyrica 300 Capsules) in Acetate *buffer pH 4.5*

Time	Dissolution medium Acetate buffer pH 4.5			
	Pregabalin 75mg Tablet (F5)		**Lyrica Capsules 75mg**	
	Batch No.: F5 300 n=6		Batch No.: n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	72.50%	6.6 (9.1%)	86.00%	2.3 (2.7%)
15	85.20%	4.6 (5.4%)	90.40%	2.3 (2.5%)
30	95.40%	3.9 (4.0%)	96.50%	1.4 (1.5%)
45	97.50%	1.7 (1.7%)	97.90%	1.0 (1.0%)

The percent released for both Test and Reference products in Acetate buffer pH 4.5 exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar.

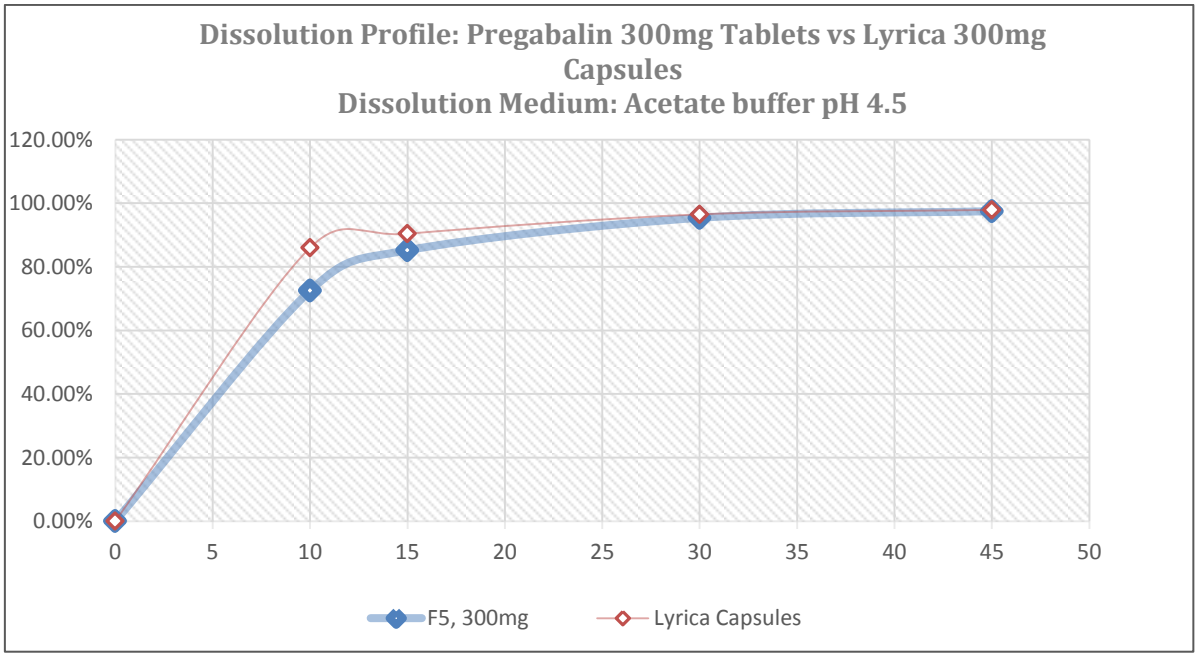

Figure 29: Dissolution profile of Pregabalin 300mg Tablets versus Lyrica 300 Capsules in Acetate buffer pH 4.5

➢ ***Dissolution Profile in Phosphate buffer pH 6.8***

Table 49: Dissolution profile results (F-5, 300mg/ Phosphate buffer pH 6.8)

Time	Dissolution medium Phosphate buffer pH 6.8			
	Pregabalin 300mg Tablet (F5)		**Lyrica Capsules 300mg**	
	Batch No.: n=6		Batch No.: F5 300 n=6	
	Release %	SD (RSD %)	Release %	SD (RSD %)
0	**0.0%**	0.0 (0.0%)	**0.0%**	0.0 (0.0%)
10	70.00%	4.4 (6.2%)	86.60%	2.3 (2.6%)
15	85.10%	6.8 (8.0%)	91.50%	2.3 (2.5%)
30	94.50%	5.8 (6.1%)	95.50%	1.4 (1.5%)
45	99.00%	3.8 (3.8%)	96.00%	1.0 (1.0%)

The percent released for both Test and Reference products in Phosphate buffer pH 6.8 exceeds 85% in 15 minutes, which means there is no need to calculate the similarity factor (f2), and the two products are considered similar.

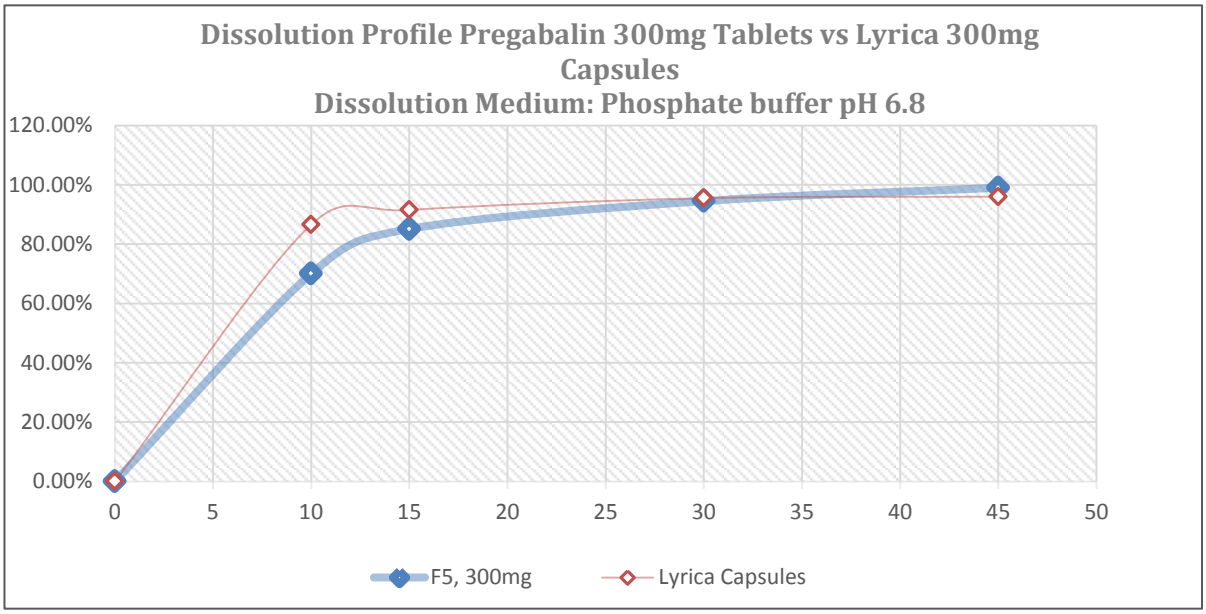

Figure 30: Dissolution profile of Pregabalin 300mg Tablets versus Lyrica 300 Capsules in Phosphate buffer pH 6.8

As a summary, the data obtained indicates that the developed Pregabalin Tablets and the reference Lyrica Capsules are similar in all aspects and considered bioequivalent.

5.8 Stability Study

The formulations F5, 300 and F5, 75 were stored at different storage conditions, at long term condition (25°C ± 2°C/ 60% RH± 5% RH), at intermediate conditions (30°C ±2°C/ 65%RH± 5%RH) and at accelerated conditions (40°C ± 2°C / 75RH± 5%RH), the stability samples were analyzed at zero time, 14 days and 30 days as schedule in the methodology stability section.

The stability results are tabulated in Tables (48-56).

5.8.1 Pregabalin Tablet 300mg (F5, 300) Stability Results

Table 50: Stability results of Pregabalin 300mg @ 25°C ± 2°C/ 60% RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conforms	Conforms	Conforms
Length	19.1 ± 0.2 mm	19.1	-	-
Width	9.1 ± 0.2 mm	9.1	-	-
Thickness	4.2 ± 0.2 mm	4.2	-	-
Hardness	6.0-12.0 Kgf	11.1	11.0	10.7
Friability	NMT 1.0%, 100 rounds	0.3%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	102.0%	99.0%	98.0%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	100.9%	102.5%	101.2%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	N.D N.D	0.0% 0.0%	0.0% 0.0%

Table 51: Stability results of Pregabalin 300mg @ 30°C ± 2°C/ 65% RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conform	Conform	Conform
Length Width	19.1 ± 0.2 mm 9.1 ± 0.2 mm	19.1 9.1	- -	- -
Thickness	4.2 (+/- 0.2) mm	4.2	-	-
Hardness	6.0-12.0 Kgf	11.1	10.5	10.5
Friability	NMT 1.0%, 100 rounds	0.3%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	102.0%	98.9%	99.0%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	100.9%	102.3%	102.0%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	N.D N.D	0.0% 0.0%	0.0% 0.0%

Table 52: Stability results of Pregabalin 300mg @ 40°C ± 2°C/ 75RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conform	Conform	Conform
Length Width	19.1 ± 0.2 mm 9.1 ± 0.2 mm	19.1 9.1	- -	- -
Thickness	4.2 (+/- 0.2) mm	4.2	-	-
Hardness	6.0-12.0 Kgf	11.1	11.3	11.1
Friability	NMT 1.0%, 100 rounds	0.3%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	102.0%	95.2%	96.0%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	100.9%	101.7%	101.2%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	N.D N.D	0.0% 0.0%	0.0% 0.0%

5.8.2 Pregabalin Tablet 75mg (F5, 75) Stability Results

Table 53: Stability results of Pregabalin 75mg @ 25°C ± 2°C/ 60% RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conform	Conform	Conform
Length Width	10.1 ± 0.2 mm 5.1 ± 0.2 mm	10.1 5.1	- -	- -
Thickness	4.2 (+/- 0.2) mm	3.2	-	-
Hardness	6.0-12.0 Kgf	10.1	9.5	10.1
Friability	NMT 1.0%, 100 rounds	0.2%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	100.0%	98.7%	99.8%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	99.0%	103.4%	101.2%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	0.0% 0.0%	0.0% 0.0%	0.0% 0.0%

Table 54: Stability results of Pregabalin 75mg @ 30°C ± 2°C/ 65% RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conform	Conform	Conform
Length Width	10.1 ± 0.2 mm 5.1 ± 0.2 mm	10.1 5.1	- -	- -
Thickness	4.2 (+/- 0.2) mm	3.2	3.2	3.2
Hardness	6.0-12.0 Kgf	10.1	9.1	9.5
Friability	NMT 1.0%, 100 rounds	0.2%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	100.0%	98.9%	100.7%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	99.0%	102.3%	101.0%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	0.0% 0.0%	0.0% 0.0%	0.0% 0.0%

Table 55: Stability results of Pregabalin 75mg @ 40°C ± 2°C/ 75RH± 5% RH

Characteristics	Acceptance Criteria	@ initial time	After 14 days	After 30 days
Appearance	Oblong white tablet, Characteristic Odor with smooth and homogeneous coating	Conform	Conform	Conform
Length	10.1 ± 0.2 mm	10.1	-	-
Width	5.1 ± 0.2 mm	5.1	-	-
Thickness	4.2 (+/- 0.2) mm	3.2	-	-
Hardness	6.0-12.0 Kgf	10.1	10.5	9.7
Friability	NMT 1.0%, 100 rounds	0.2%	-	-
Dissolution Test	Ref. (In-House)			
Pregabalin	Not Less than 75.0 % (Q) of the labeled amount of Pregabalin is dissolved within 30 minutes	100.0%	96.0%	96.0%
Assay	Ref. (In-House)			
Pregabalin	90.0-110.0 %	99.0%	101.7%	101.2%
Related Substances	Ref. (In-House)			
Individual impurities. Total Impurities.	NMT 0.2% NMT 1.0%	0.0% 0.0%	0.0% 0.0%	0.0% 0.0%

Discussion of Stability Results:

All stability study results indicate that Pregabalin tablets are stable in terms of Assay, Physical properties, dissolution and related substance at all storage conditions during the study period.

5.9 Assay and Related Substances Test

5.9.1 Linearity and Range:

Different concentrations of Standard solution were prepared covering the range between (40%-160%) of the nominal concentration (1.0 mg/ml) required by the procedure for determination of *Pregabalin in Pregabalin Tablets*. Data and results are summarized in Table-56:

Table 56: Linearity and Range Data and Results

Conc. %	Conc. of Pregabalin (mg/ml)	Peak Area 1	Peak Area 2	Peak Area 3	Average	RSD
40%	0.4	397923	398777	405286	**400662**	**1.0%**
60%	0.6	606374	605375	600093	**603947**	**0.6%**
80%	0.8	800873	794467	786968	**794103**	**0.9%**
100%	1.0	995222	999234	1003545	**997228**	**0.3%**
120%	1.2	1194276	1204666	1192104	**1197015**	**0.6%**
140%	1.4	1379232	1379587	1375498	**1378106**	**0.2%**
160%	1.6	1592187	1584304	1578861	**1585117**	**0.4%**

Linearity regression analysis demonstrated acceptability of the method for quantitative analysis over the corresponding range. Data and results are reported in table-56 and figure-31 which demonstrate linearity as well over the specified range with a Correlation Coefficient (R^2 = 0.9998), Slope = ***982,963.5***, and y-Intercept = ***10,776.3***. Repetitions of standard solutions over the range of linearity are precise with RSD less than 1.1% for all levels, concentrations over the range are linear with correlation coefficient equal to "R^2 = 0.9998", and accurate for all recoveries over the range and are within the limit (100±2%).

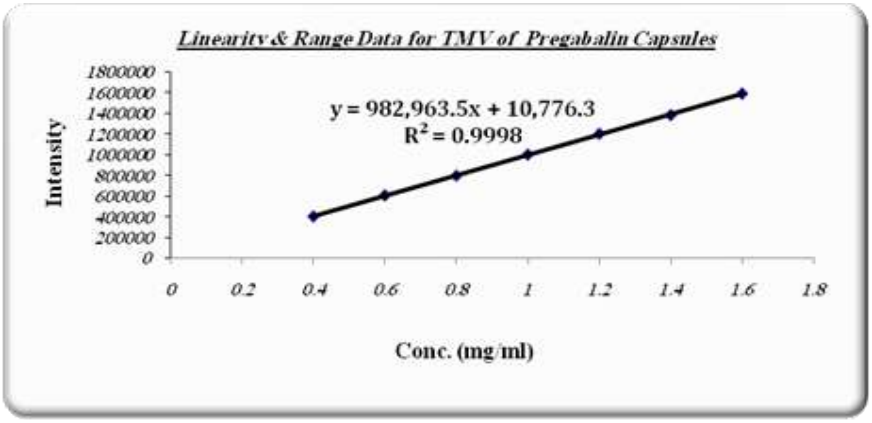

Figure 31: Linearity and Rang of Pregabalin

5.9.2 Precision (System Repeatability):

Six replicate injections of the working standard at nominal concentration (1.0 mg/ml) were made according to the test method of analysis for determination of ***Pregabalin in Pregabalin Tablets*** and the relative standard deviation (RSD) of the peak areas was calculated for the above replicate injections as illustrated in Table-57:

Table 57: Precision of Standard Injection Data and Results

Inj. No.	RT	Area
1	6.347	1006350
2	6.353	1009890
3	6.347	1008178
4	6.343	1016541
5	6.353	1004494
6	6.373	1007422
AV **RSD**	**6.353**	**1008813** **0.4%**

The relative standard deviation (RSD) of the peak areas for replicate injections of the working standard solution at nominal concentration was calculated, with referring to the data precision table-64, the RSD is about 0.4%, therefore the system is repeatable.

5.9.3 Ruggedness (Intermediate Precision):

Ruggedness was studied through analysis of nominal standard solution (1.0 mg/ml of Pregabalin) under a variation of analyst and analysis days (Matrix Design). Ruggedness data and results are illustrated in Table-58:

Table 58: Ruggedness Data & Results

Injection	Analysis by Different Analyst on Different Days (Matrix Design)	
	R.T	Area
I	5.880	1017260
II	5.870	996929
III	5.847	1003262
IV	5.887	1009491
V	5.847	1007657
VI	5.897	1002387
Average **RSD**	**5.871**	**1006164** **0.7%**
Limit	**RSD NMT 2.0%**	

Ruggedness was studied through analysis of nominal standard solution (1.0 mg/ml of Pregabalin) under a variation of analyst & analysis days (Matrix Design). The RSD result is about 0.7%, as illustrated in Table-67. All results are within limits (RSD is NMT 2.0%). This indicates that the method of analysis is precise within-laboratories variation.

5.9.4 Accuracy of the Drug Products:

Admixture the Synthetic mixture of the drug product components (Placebo: Placebo was prepared according to the formulation procedure reported in the production file of Pregabalin Tablets) with known amounts of Pregabalin, (80%, 100% & 120%) of the nominal concentration (1.0 mg/ml). Three preparations were made for each concentration and the recovery was calculated as illustrated in Table-59

Table 59: Pregabalin Recovery Data & Results for Accuracy of Pregabalin Tablets

Conc. %	Concentration (mg/ml)	Samples Area	% Accuracy (Recovery)	Average & RSD
Average Area for 80% of Nominal St. Conc.= 802332				
80%	0.8	799789	99.7%	99.7% & 0.1%
80%	0.8	799381	99.6%	
80%	0.8	800496	99.8%	
Average Area for 100% of Nominal St. Conc.= 1002818				
100%				
100%	1.0	1008526	100.6%	100.2% & 0.4%
100%	1.0	1001277	99.8%	
100%	1.0	1005557	100.3%	
Average Area for 120% of Nominal St. Conc.= 1204265				
120%				
120%	1.2	1206316	100.2%	100.5% & 1.2%
120%	1.2	1197834	99.5%	
120%	1.2	1203749	101.9%	

Three preparations for each recovery level (80%, 100% & 120%) of nominal concentration are prepared and injected into the liquid chromatograph, the recovery percent was calculated for Pregabalin as demonstrated in table-65. The recovery results are (99.7%, 100.2% & 100.5%, respectively) and are within limits (100±2%).

5.9.5 Robustness:

Robustness was studied through variation of method parameters, variation of flow rate to 1.65 ml/min. instead of 1.50 ml/min., and variation of detection wavelength to 208 nm instead of 210 nm. Data and results are illustrated in Tables-60

Table 60: Robustness Data and results

Injection No.	Variation in Flow Rate		Variation in Detection Wavelength	
	R.T	Area	R.T	Area
1	5.453	901066	5.883	1260983
2	5.470	898267	5.897	1266299
3	5.500	893574	5.870	1273643
4	5.490	897677	5.900	1255641
5	5.467	900116	5.873	1260652
6	5.513	905936	5.863	1259450
Average **SD** **RSD**	**5.482**	**899439** **4103.5** **0.5%**	**5.881**	**1262778** **6327.7** **0.5%**
Limit	**RSD NMT 2.0%**			

Robustness was studied through analysis of nominal standard solution (1.0 mg/ml of Pregabalin) under a variation of method parameters (Flow rate and Detection Wavelength). The RSD result for analysis under variation of flow rate is about 0.5% and under variation of detection wavelength is about 0.5% and are within acceptance criteria (RSD < 2.0%) as illustrated in table-66.This indicates that all parameters are within the methods robustness range.

5.9.6 Specificity:

Different reagents were added to the nominal standard solution of Pregabalin and Pregabalin spiked on the placebo of Pregabalin Tablets, the assay of the main peak (analyte) in each stressed solution is calculated according to test method of Pregabalin Tablets against the standard solution injected on the same day. The data and results are illustrated in Table-61

Table 61: Stability of Pregabalin standard under different stressing conditions
Av. Area for Pregabalin St.: 990317 **Initial %: 100%**

Stress Conditions	Pregabalin Standard		Pregabalin Std. Spiked on Placebo of Pregabalin Tablets	
	Area	%	Area	%
0.5M HCl	940561	95.0%	964391	97.4%
0.5M NaOH	985254	99.5%	955171	96.5%
6% H_2O_2	470910 508525*	47.6% 51.3%*	467481 437655*	47.2% 44.2%*
Heat in Water Bath at 70C for 1-Hr	986388 31541*	99.6% 3.2%*	983706 31636*	99.3% 3.2%*
***: Total of Degradants**				

With referring to the stability indicating table-6, it is clear that, Pregabalin is unstable and degraded in the presence of 6% Hydrogen peroxide, 0.5M Sodium Hydroxide, 0.5M Hydrochloric acid and at elevated temperatures. As illustrated in Table-6.

Finally, the peak of Pregabalin is clearly separated from other peaks (Degradation Peaks), in all cases studied, with a resolution greater than 1.5 as shown in Figure-34

As a conclusion, the test method is stability -indicating for determination of Pregabalin in Pregabalin Tablets.

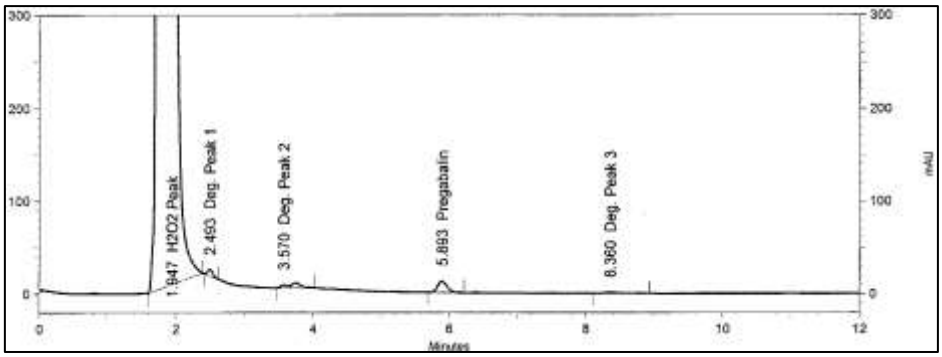

Figure 32: Stability Chromatogram for Pregabalin when Stressed with H_2O_2

5.9.7 System Suitability

Six replicate injections of the standard solution at nominal concentration (1.0 mg/ml of Pregabalin) and Six replicate injections of the Pregabalin and its impurity (IV) were made according to the test method of analysis for determination of ***Pregabalin*** in ***Pregabalin Tablets*** and the relative standard deviation (RSD) and Relative RT are calculated, Asymmetry and Theoretical Plates, Results are tabulated as illustrated in Table-69.

Table 62: System Suitability Data & Results

Inj. No.	Response Area (Pregabalin)	Tailing Factor	Column Efficiency	Impurity (IV) (Relative RT)	Impurity (IV) Resolution
1	1006350	1.056	9938	1.9	17.6
2	1009890	1.073	9917	1.9	18.3
3	1008178	1.069	9938	1.9	17.7
4	1016541	1.065	9840	1.9	18.0
5	1004494	1.058	9932	1.9	17.8
6	1007422	1.061	9951	1.9	17.4
AV RSD	**1008813 0.4%**	**1.064**	**9919**	**1.9**	**17.8**

The results of the System Suitability Parameters were calculated automatically by the software (EZChrom Elite) of the Lachrom Elite HPLC System.

Relative Standard Deviation (RSD):

The Relative Standard Deviation of peak area for replicate injections of Pregabalin standard is equal to 0.4%.

Column Efficiency

The column efficiency for Pregabalin peak is about 9919 theoretical plates.

Tailing Factor

The tailing factor for Pregabalin peak is about 1.1.

Relative retention time (Impurity IV)

The RRT for Impurity (IV) is about 1.9, as shown in the figure-3.

Resolution (Impurity IV)

The resolution between Pregabalin API and RS Impurity IV (Lactam) is about 17.8, as shown in the next figure.

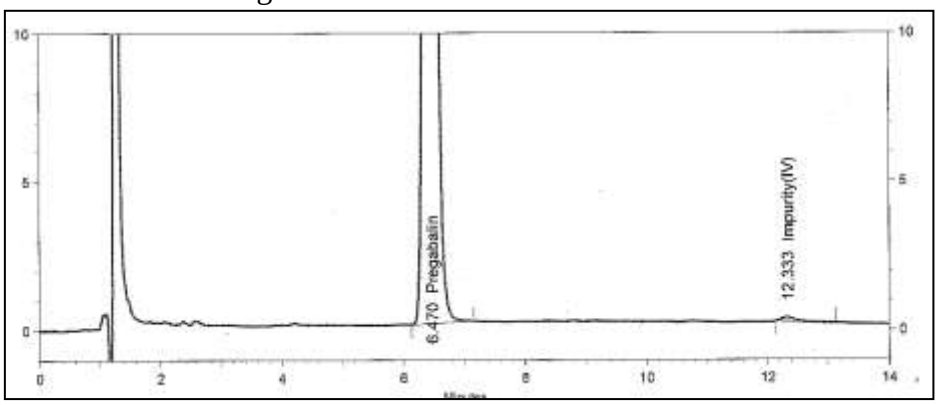

Figure 33: System Suitability Chromatogram (Pregabalin and RS Impurity IV)

5.10 Dissolution test method validation

5.10.1 Linearity and Range:

Different concentrations of Standard solution were prepared covering the range between (40%-120%) of the nominal concentration (0.3333 mg/ml) required by the procedure for determination of ***Pregabalin*** in ***Pregabalin Tablets***. Data and results are summarized in Table-63:

Table 63: Linearity and Range Data and Results

Conc. %	Conc. of Pregabalin (mg/ml)	1st Area	2nd Area	3rd Area	Av Area	RSD
40%	0.1333	0.714	0.696	0.71	**0.707**	**1.3%**
60%	0.1999	1.058	1.064	1.06	**1.061**	**0.3%**
80%	0.2666	1.423	1.386	1.396	**1.402**	**1.4%**
100%	0.3333	1.773	1.784	1.794	**1.779**	**0.4%**
120%	0.3999	2.135	2.143	2.142	**2.140**	**0.2%**
150%	0.4999	2.651	2.659	2.649	**2.653**	**0.2%**

Standard solutions were prepared over the range (40% to 150%) of the nominal concentration (0.3333 mg/ml of Pregabalin). Linearity regression analyses are acceptable of the method for quantitative analysis over the corresponding range. Data and results are reported in Table-63 and figure-28 which demonstrate linearity as well over the specified range with a Correlation Coefficient (R^2 = 0.999), Slope = ***2513.9***, and y-Intercept = **11.599**. Repetions of standard solutions over the range of linearity are precise with RSD less than 1.0% for all levels, concentrations over the range are linear with correlation coefficient equal to "R^2 = 0.999", and accurate for all recoveries over the range and are within the limit (100±5%)

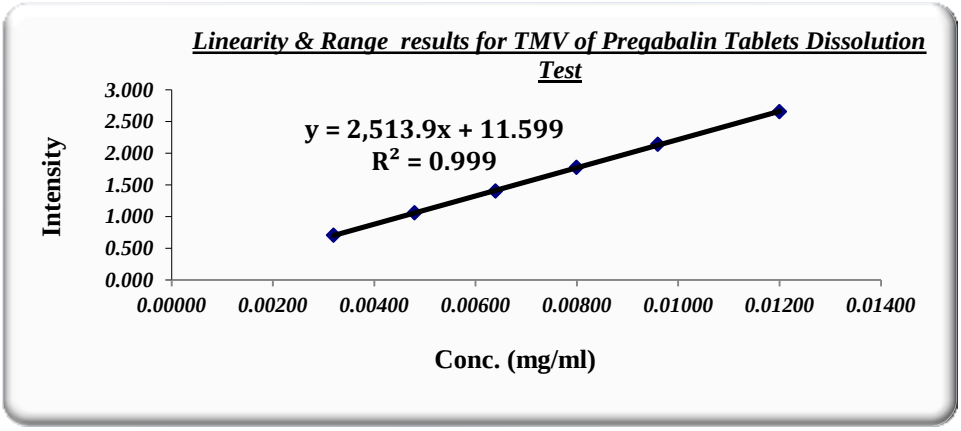

Figure 34: Linearity and Rang of Pregabalin

5.10.2 Precision (System Repeatability):

Six replicate readings of the working standard at nominal concentration (0.3333 mg/ml) were made according to the test method of analysis for determination of ***Pregabalin*** in ***Pregabalin Tablets*** and the relative standard deviation (RSD) was calculated for the above replicate readings as illustrated in Table-64

Table 64: Precision of solution, Data and Results

No.	RT	Response
1	6.120	1.791
2	6.120	1.798
3	6.150	1.804
4	6.120	1.78
5	6.120	1.776
6	6.120	1.78
AV **RSD**	**6.125** **0.2%**	**1.788** **0.6%**

The relative standard deviation (RSD) for replicate readings of the working standard solution at nominal concentration was calculated, with refering to the data precision table-71, the RSD is about 0.6%, therefore the method of analysis is repeatable.

5.10.3 Ruggedness (Intermediate Precision):

Ruggedness was studied through analysis of nominal standard solution (0.3333 mg/ml of Pregabalin) under a variation of analyst and analysis days (Matrix Design). Ruggedness data and results are illustrated in Table-65

Table 65: Ruggedness Data & Results

No.	Analysis by Different Analyst on Different Days (Matrix Design)
	Area
1	1.777
2	1.788
3	1.784
4	1.775
5	1.773
6	1.759
Average **SD** **RSD**	**1.776** **0.01** **0.6%**
Limit	**RSD NMT 5.0%**

Ruggedness was studied through analysis of nominal standard solution (0.3333 mg/ml of Pregabalin) under a variation of analyst & analysis days (Matrix Design). The RSD result is about 0.6%, as illustrated in Table-65. All results are within limits (RSD is NMT 5.0%). This indicates that the method of analysis is precise within-laboratories variation.

5.10.4 Accuracy of the Drug Products:

Admixture the Synthetic mixture of the drug product components (Placebo: Placebo was prepared according to the formulation procedure reported in the production file of Pregabalin Tablets) with known amounts of Pregabalin, (50%, 75% & 100%) of the nominal concentration (0.3333mg/ml). One Dissolution run (6-Vessels) was made for each concentration and the recovery was calculated as illustrated in Table-66.

Table 66: Pregabalin Recovery Data & Results for Accuracy of Pregabalin Tablets

Conc. %	Concentration (mg/ml)	Samples Area	% Accuracy (Recovery)	Average & RSD
Average Area for 100% of Nominal St. Conc.= 1.775				
100%				
Vessel-**I**	0.3333	1.778	100.2%	100.1% & 0.2%
Vessel- **II**	0.3333	1.772	99.8%	
Vessel- **III**	0.3333	1.775	100.0%	
Vessel- **IV**	0.3333	1.776	100.1%	
Vessel- **V**	0.3333	1.780	100.3%	
Vessel- **VI**	0.3333	1.777	100.1%	
Average Area for 75% of Nominal St. Conc.= 1.333				
75%				
Vessel-**I**	0.2500	1.342	100.7%	100.5% & 0.5%
Vessel- **II**	0.2500	1.348	101.2%	
Vessel- **III**	0.2500	1.331	99.9%	
Vessel- **IV**	0.2500	1.337	100.3%	
Vessel- **V**	0.2500	1.333	100.0%	
Vessel- **VI**	0.2500	1.347	101.1%	
Average Area for 50% of Nominal St. Conc.= 0.884				
50%				
Vessel-**I**	0.1666	0.880	99.6%	100.1% & 0.3%
Vessel- **II**	0.1666	0.887	100.4%	
Vessel- **III**	0.1666	0.886	100.3%	
Vessel- **IV**	0.1666	0.888	100.5%	
Vessel- **V**	0.1666	0.885	100.2%	
Vessel- **VI**	0.1666	0.882	99.8%	

One Dissolution run (6-Vessels) for each recovery level (50%, 75% & 100%) of nominal concentration is performed and analyzed using the Chromatographic HPLC, the recovery percent was calculated for each level of Pregabalin as demonstrated in table-66. The recovery results are (100.1%, 100.5% & 100.1%, respectively) and are within limits (100±5%).

5.10.5 Method Conditions Variation

Robustness was studied through variation of Chromatographic HPLC method parameters and dissolution conditions as variation in rotation speed to 48 rpm instead of 50 rpm, Detection wavelength to 212nm instead of 210nm and Flow rate 1.6ml/min instead of 1.5ml/min. Data and results are illustrated in Tables-67.

Table 67: Robustness Data and results for Pregabalin:

No.	Variation in Rotation Speed (RPM)	Variation in Flowrate	Variation in Detection Wavelength
	Response Area	**Response Area**	**Response Area**
1	1.782	1.670	1.448
2	1.774	1.673	1.445
3	1.767	1.671	1.449
4	1.773	1.676	1.446
5	1.775	1.672	1.441
6	1.770	1.673	1.445
Average	**1.774** **0.3%**	**1.673** **0.1%**	**1.446** **0.2%**
Limit	**RSD NMT 5.0%**		

Robustness was studied through analysis of nominal standard solution (0.3333 mg/ml of Pregabalin) under variation of HPLC method parameter (Detection Wavelength and Flow rate) and under variation of dissolution test condition RPM. The RSD result for analysis under variation of RPM is about 0.3%, under variation of detection wavelength is about 0.2% and under variation of flow rate is about 0.1%, and all results are within acceptance criteria (RSD < 5.0%) as illustrated in table-67.This indicates that all parameters are within the methods robustness range.

5.10.6 Specificity (Placebo Interference):

This test is to demonstrate that the results are not unduly affected by placebo constituents for that Placebo, Standard and Sample Solution was prepared at nominal concentration and the measurements was performed for both the nominal standard solution and for placebo solution at 210 nm wavelength for Pregabalin in order to calculate the interference percentage of placebo using the following formula:

$$\textbf{Interference \%} = 100*C*(A_P/A_{St})*V/L$$

Where,

C: is the concentration, in mg per ml, of the standard

A_P: is the absorbance of the placebo

A_{St}: is the absorbance of the standard

V: is the volume, in ml, of the medium

L: is the label claim, in mg of the product

Table 68: Interference Data and results

Pregabalin Conc. (mg/ml)	$A_{Placebo}$	$A_{St.}$	V	L	Interference %
0.3333	0.008	1.760	900	300	**0.45%**
Acceptance Criteria	**NMT 2%**				

Referring to Table-68, the result of interference is about 0.45% and it is within limits (the interference should be NMT 2%). This means that, the interference of placebo on results is too low and negligible in comparison with the interference limit.

5.10.7 Specificity / Stability of solution:

Sample solution was prepared by nominal standard solution spiked with placebo of Pregabalin Tablets; both of Sample and Standard solutions were studied at freshly prepared time, after 3hr and after 24hr, assay of the analyte in each solution are calculated according to test method of Pregabalin Tablets against the freshly prepared standard solution on the same time. The data and results are illustrated in Table-69:

Table 69: Stability of Pregabalin standard and Sample of Pregabalin Tablets under different test time.

Freshly STD	Stability STD	Stability STD Spiked with Placebo of Pregabalin Tablets
After 3 hr's		
1.781	1.807	1.787
		1.758
		1.766
Average Degree of Stability	**--- 101.4%**	**1.77 99.4%**
After 24 hr's		
1.786	1.813	1.790
		1.779
		1.783
Average Degree of Stability	**--- 101.5%**	**1.784 99.9%**

With referring to the stability indicating table-69, it is clear that, the Pregabalin standard and Pregabalin spiked with placebo of Pregabalin Tablets are stable after storage at room temperature for period of time starting from zero time (freshly prepared) and up to 24hr of preparation.

Part VI Conclusions and Recommendations

Conclusions and Recommendations

Final Conclusion

Successful attempts were made to formulate new line extension, Pregabalin 300mg and 75mg tablets by using excipients generally used in pharmaceutical industry and compatible with the active pharmaceutical ingredient. This new line extension is expected to be more stable, have high capacity of manufacturing and low manufacturing costs.

The developed Pregabalin tablets are dose weight proportional and proved to be biowaiver to the Reference Drug Product (Lyrica Capsules).

An HPLC assay and dissolution methods using a PDA UV detector were developed and validated for the determination of Pregabalin in pharmaceutical dosage forms, such as capsules and tablets. The method has been found to be precise, accurate and stability indicating, which is suitable for stability testing of Pregabalin capsules and tablets.

Pregabalin tablets proved to be stable for the period tested at accelerated and long-term storage conditions.

Recommendation and future work

The following are some suggestions concerning future work:

1. Complete the stability study of trials processed according to selected formulation up to 12 months at long term and intermediate storage conditions, and for 6 months at accelerated storage conditions.

2. Formula and particle size optimization to improve the flowability, compaction of powder mixture blend and performance of highest strength tablets.

3. Scale up the batch size of tablets to lab scale (25,000 units) and pilot scale (100,000 units) for stability testing according to ICH, FDA and WHO requirements.

4. Biowaiver study using Pilot batch sizes

5. Registration of product locally and in export countries.

Abbreviations

API: Active Pharmaceutical Ingredient

AR: Analytical Reagent.

BDL: Below Detection Limit.

BP: British Pharmacopoeia.

COA: Certificate of Analysis.

COA: Certificate of Analysis.

FDA: Food and Drug Administration.

FDA: Food and Drug Administration.

FPP: Finished Pharmaceutical Product.

FTIR: Foriour Transmitter infrared.

HCl: Hydrochloric Acid.

HPLC: High Pressure Liquid Chromatogram.

IPC: In-Process Control

IR: Immediate Release.

M: Molarity.

N: Normality.

NLT: Not Less Than.

NMT: Not More Than.

OTR: Oxygen Transition Rate.

R.T: Retention Time.

R^2: Linear Regression.

RP: Reversed Phase.

RPM: Round per minute.

RSD: Relative Standard Deviation.

SD: Standard Deviation.

STD: Standard

SUPAC: Scale up and Post Approval Changes.

USP: United State Pharmacopoeia.

VAL: Validation.

WHO: World Health Organization.

WS: Working Standard.

WVTR: Water Vapor Transition Rate.

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Appendix 1

COAs of Pregabalin and Excipients

Alembic
Touching Lives over 100 years

QUALITY CONTROL DEPARTMENT

CERTIFICATE OF ANALYSIS

Product	Pregabalin		
Batch No.	1402000112	A.R. No.	80000021629
Mfg. Date	JAN-2014	Retest Date	DEC-2016
Batch size	187.16 Kg	Date of Analysis	31/03/2014

SR. NO.	TESTS	RESULTS	SPECIFICATIONS
01	Description	White crystalline powder	White to off white crystalline powder.
02	Solubility	Complies	Sparingly soluble in water.
03	Identification by A) IR [KBr]	Comparable	Absorption spectrum of the test sample should be concordant with that of (S)-Pregabalin standard.
	B) HPLC	Complies	The retention time of main peak of test sample should match with the retention time of peak of (S)-Pregabalin standard in the test of related substances.
	C) XRD	Complies	The XRD pattern of test sample should match with the XRD pattern of (S)-Pregabalin standard and should match with 2θ values at 9.5, 12.2, 16.6, 18.2, 18.3, 19.0, 22.1, 23.1, and 23.4 (±0.2 θ)
04	Water by KF	0.03 % w/w	Not more than 0.50 % w/w
05	Residue on ignition	0.04 % w/w	Not more than 0.10 % w/w
06	Specific optical rotation(°) (on anhydrous basis) (c=1.06 in water, at 23°C)	+11.5°	+10.0 to +12.0
07	Bromide content	Less than 50 ppm	Not more than 50 ppm
08	Heavy Metals	Less than 10 ppm	Not more than 10 ppm

COA Date: 22/04/2014		Prepared By	Checked By	Approved By
	Sign.			
	Date	22/04/14	22/04/14	22/04/14
	Name	Bharat Bhoi	Kirti Ladani	Tumma Ilaiah
	Designation	Asst. Manager-QC	Dy. Manager-QC	Manager-QC

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CIN-L24230GJ2010PLC061123

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QUALITY CONTROL DEPARTMENT

CERTIFICATE OF ANALYSIS

Product	Pregabalin		
Batch No.	1402000112	A.R. No.	80000021629
Mfg. Date	JAN-2014	Retest Date	DEC-2016
Batch size	187.16 Kg	Date of Analysis	31/03/2014

SR. NO.	TESTS	RESULTS	SPECIFICATIONS
09	Assay (By chemical) (on anhydrous basis)	100.3 % w/w	98.0%w/w to 102.0%w/w
10	R-Isomer (By HPLC)	0.01 %	Not more than 0.15 %
11	Related substances by HPLC		
	Impurity-III	Not detected	Not more than 0.15 %w/w
	Impurity-IV	Not detected	Not more than 0.15 %w/w
	Any other impurity	0.06 % w/w	Not more than 0.10 %w/w
	Total impurities	0.11 % w/w	Not more than 0.50 %w/w
12	Residual solvents by GC		
	Cyclohexane	Not detected	Not more than 1000 ppm
	Toluene	6 ppm	Not more than 500 ppm
	Ethyl acetate@	Not detected	Not more than 5000 ppm
	Methanol	Not detected	Not more than 1800 ppm
	Chloroform	Not detected	Not more than 60 ppm
	Isopropyl alcohol@	Not detected	Not more than 5000 ppm
	Ethanol@	Not detected	Not more than 5000 ppm
	Total of class 3 solvents	Nil	Not more than 5000 ppm
Additional test :-			
13	Particle size (By Malvern) D(0.9)	364 µm	For information.

Batch complies with respect to above specification No. API/30000503/H-U-03. @: Class 3 solvents.

COA Date: 22/04/2014		Prepared By	Checked By	Approved By
	Sign.			
	Date	22/4/14	22/04/14	22/04/14
	Name	Bharat Bhoi	Kirti Ladani	Tumma Ilaiah
	Designation	Asst. Manager-QC	Dy. Manager-QC	Manager-QC

ALEMBIC PHARMACEUTICALS LIMITED

API UNIT – III, KARAKHADI

REGD. OFFICE: ALEMBIC ROAD, VADODARA – 390 003, Gujarat State, INDIA • TEL: (0265) 2280550 • FAX: (0265) 2284529
Website: www.alembic-india.com • E-mail : alembic@alembic.co.in

FACTORY : SURVEY NO. 862, 863 VILL. KARAKHADI, TAL. PADRA, DIST. VADODARA-391 450 • TEL. : 02662-280780, 280781 FAX : 02662-280782
E-mail : karakhadi.api@alembic.co.in

CIN-L24230GJ2010PLC061123

Colorcon

Certificate of Analysis

Product Name:	OPADRY® II COMPLETE FILM COATING SYSTEM 85F28751 WHITE
Product Number:	85F28751
Material Description:	White Powder
Lot No:	D9620542
Quantity Supplied:	25 KG
Ship To:	Jerusalem Pharmaceuticals PS
Bill To:	Jerusalem Pharmaceuticals PS
Sales Order No:	CCUK995079
Customer PO NO:	203/2014

Compliance Statement: This Product meets all agreed upon specifications

Specifications

Test	Method	Minimum	Maximum	Result	Analyst
ASH, %	QLO-QC-TM-0499	34.00	46.00	39.09 %	JH
APPEARANCE	QLO-QC-TM-0733	WHITE POWDER		WHITE POWDER	JH
COLOR DIFFERENCE DE, CIE	QLO-QC-TM-0576/05	0.0	1.5 Or Visual Match	1.6 CIE	JH
COLOR DIFFERENCE VISUAL	QLO-QC-TM-0576	COMPARES		COMPARES	JH
DISPERSION	QLO-QC-TM-0729	PASS		PASS	JH
IR SCAN	QLO-QC-TM-0744	COMPARES		COMPARES	JH
SPOCK CHECK	QLO-QC-TM-0730	PASS		PASS	JH

Ingredients	USFDA Pigment Certification
POLYVINYL ALCOHOL	N/A
TITANIUM DIOXIDE	N/A
POLYETHYLENE GLYCOL / MACROGOL	N/A
TALC	N/A

Regardless the solvents identified in the ICH Guideline on Residual Solvents Q3C and in the USP/NF General Chapter <467> Residual Solvents, only Class 2 and Class 3 solvents are likely to be present depending on the composition of the specific product formulation. If present, residual Class 2 solvents are below the Option 1 limit and residual Class 3 solvents are below 0.5%. See the Colorcon Product Regulatory Database for this product for detailed information on the solvents and levels that may be present in this particular product.

This Product was manufactured in a facility that is registered with the United States FDA under the provisions of the Bioterrorism Preparedness and Response Act.

Manufactured By: COLORCON
Manufacturing Site: Dartford, UK

Date Of Manufacture: 15-JAN-2014
Re-evaluation Date: 15-JAN-2016

The above analytical data has been reviewed and authenticated electronically by an authorized representative of the Quality Unit as evidence by the application of an electronic signature.

Approved By: Eve Newman
Electronic Signature ID: 1135808-1139096

Date: 17-JAN-2014

Flagship House Victory Way, Crossways Dartford Kent DA2 6QD England Tel: 44-1322-293000 Fax: 44-1322-627200 www.colorcon.com

IMERYS
Talc

DATE: 30/05/14

TO:

CERTIFICATE OF ANALYSIS N. 0921/13

PRODUCT: TALC LUZENAC PHARMA

REF./ORD.: 5929 / GL 1869

OUR REF.: 20554 SO

EXPIRY DATE : May 2017

		LOT	6412/14
		PRODUCTION DATE	28/05/14
1 -	WHITENESS Y (Minolta CR-300)		91.8
2 -	RESIDUE on 75 µm SIEVE (Alpine 200 LS) [%]		0.1
3 -	MICROBIOLOGY : aerobic bacteria [cfu/g]		< 20
	Fungi [cfu/g]		< 10
	Microbial limits : If intent for cutaneous administration : TAMC 100 cfu/g / If intent for oral administration : TAMC 1000 cfu/g and TYMC 100 cfu/g		
4 -	EUROPEAN PHARMACOPOEIA		
	04/2012:0438 talc monograph		
	Test and specifications		
	Identification (talc) test performed once a year		Positive
	Asbestos (test performed once a month)		Not detected
	Acid or alkalinity (compliant)		Compliant
	Water soluble substances (10 mg max.)	Mg	< 5.0
	Aluminium (2.0 % max.)	[%]	< 1.0
	Calcium (0.9 % max.)	[%]	0.3
	Acid soluble iron (0.25 % max.)	[%]	0.11
	Magnesium (17.0-19.5 %)	[%]	18.8
	Lead (10 ppm max.)-test performed once a year	[ppm]	< 10
	Loss on ignition 1050 °C (7.0 % max.)	[%]	5.4
5 -	UNITED STATES PHARMACOPOEIA (USP 37-NF32)		
	Test and specifications		
	Identification (positive) test performed once a year		Positive
	Asbestos (test performed once a month)		Not detected
	Acid or alkalinity (compliant)		Compliant
	Water soluble substances (5 mg max.)	Mg	< 5.0
	Aluminium (2.0 % max.)	[%]	< 1.0
	Calcium (0.9 % max.)	[%]	0.3
	Acid soluble iron (0.25 % max.)	[%]	0.11
	Magnesium (17.0-19.5 %)	[%]	18.8
	Lead (10 ppm max.) test performed once a year	[ppm]	< 10
	Loss on ignition 1050 °C (7.0 % max.)	[%]	5.4

Signed:

IMERYS
Talc
Imerys Talc Italy S.p.A.
QUALITY MANAGER
(Anna Maria BONORA)

Imerys Talc Italy S.p.A. – Sede Legale e Stabilimento: Via Nazionale 121 – 10060 Porte (TO) Italy
T +39 0121 304 911 - Fax +39 0121 503 403
Capitale € 4,865,155 int. Versato – REA TO n. 0748332 – Cod. Fisc. e Partita IVA IT 09330410017

11-4-2014

21-3-2015

...GAO CHEMICALS CO.,LTD

CERTIFICATE OF ANALYSIS

Product Name	MAGNESIUM STEARATE PO		Analysis basis	BP/USP	
Batch Number	20141005	Date of Mfg.	2014-10-05	Date of Expiry	2016-10-04
Quantity	100 kgs	Packages Size	25kg/bag		

Storage condition: Keep the container tightly closed in a dry and well-ventilated place away from light or heat or moisture

ITEM	SPECIFICATION	RESULTS
Appearance	White light no sand behaviour fine powder, no visible evidence of contamination by foreign matter	Conforms
Identification	Conform	Conforms
Loss on drying	Conform	Conforms
Acid number	195-210	200.4
Chloride	Not more than 0.25%	< 0.25%
Sulfate	Not more than 0.5%	< 0.5%
Free fatty acid	Not more than 6.0%	4.6%
Cadmium	Not more than 2ppm	<2ppm
Heavy metals (Lead)	Not more than 10ppm	< 10ppm
Nickel	Not more than 5ppm	<5ppm
Accumulate density	Not more than 0.30-0.35g/ml	0.32g/ml
Assay (Mg)	4.0% to 5.0%	4.7%
Pyrogen		
Total microbial count	Not more than 1000CFU/gram	Conforms
Yeast and mold	Not more than 100CFU/gram	Conforms
Escherichia coli	Negative	Not detect

Conclusion: The above results comply with current BP/USP.

Analyst:	Liu Yan	Checker	Liu Jiayin	Supervisor	Qi Jin
Signature		Signature		Signature	

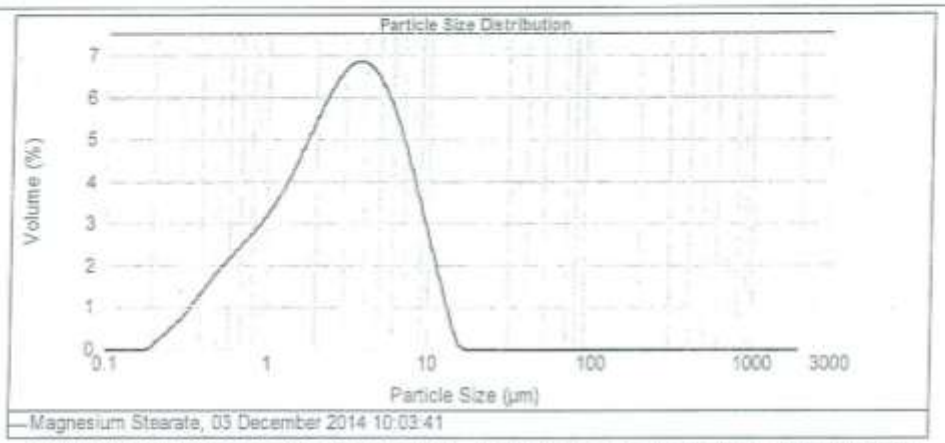
Hubei 9.12.2014

MASTERSIZER 2000

Result Analysis Report

Sample Name: Magnesium Stearate	**SOP Name:** Magnesium Stearate	**Measured:** 03 December 2014 10:03:41	
Sample Source & type: Supplier	**Measured by:** Administrator	**Analysed:** 03 December 2014 10:03:42	
Sample bulk lot ref: 201411295	**Result Source:** Measurement		
Particle Name: Fraunhofer	**Accessory Name:** Scirocco 2000 (A)	**Analysis model:** General purpose	**Sensitivity:** Normal
Particle RI: 0.000	**Absorption:** 0	**Size range:** 0.020 to 2000.000 um	**Obscuration:** 13.91 %
Dispersant Name:	**Dispersant RI:** 1.000	**Weighted Residual:** 2.115 %	**Result** Off
Concentration: 1.0009 %Vol	**Span :** 2.436	**Uniformity:** 0.75	**Result units:** Volume
Specific Surface Area: 3.65 m²/g	**Surface Weighted Mean D[3,2]:** 1.648 um	**Vol. Weighted Mean D[4,3]:** 3.642 um	

d(0.1): 0.702 um d(0.5): 2.891 um d(0.9): 7.744 um



Operator notes:

Jerusalem Pharmaceuticals Co. (Al-Quds)

Microbial Limits Test

Page No.

Mg Stearate

Batch No.	2014 11295				
Code	21131600	Dosage Form	TR.M	Reg. #	
Mfg Date	10/2014	Theoretical Batch Size		Superseded	
Exp Date	10/2016	Volume/Weight		Issue Date	

Test Date: 31/12/2014

1- Total Microbial Count:

- Transfer 10g or 10mL of sample to 90mL of TS B and 90 ml of lactose broth.
- Withdraw 1mL into each of 2 TSA Plates for total aerobic count.
- Withdraw 1mL into each of 2 SDA Plates for Total combined yeasts and molds count.
- Incubate the TSA, at 32.5 +/- 2.5 C for 72 hrs and the SDA at 22.5+/- 2.5 C for 5 days.

Result: less than 10cfu/g of total bacterial Count

less than 10cfu/g of total yeast and molds.

Limits: Not To cfu/g of Bacteria.

Not to cfu/g of yeast and molds.

2- Test for Staph. aureus, Pseudomonas aeruginosa, Salmonella, E.coli & other microorganism: according to QC013.

Result: Absence of S. aureus, Salmonella, E.coli and P. aeruginosa.

Limits: Absence of Salmonella and E.Coli

Conclusion: Accepted

Microbiologist: Date: 1/12/2014

Micro. supervisor: Date: 01/12/2014

Quality Unit Manager: Date: / /

Form #qp27-3 rev c

ZHONGRAO CHEMICALS CO.,LTD

E-mail: Julia@zhongraochemical.com

CERTIFICATE OF ANALYSIS

Product Name	PREGELATINIZED STARCH (STARCH 1500)		Analysis basis	BP2011	
Batch number	20141245	Date of Mfg.	2014-12-29	Date of Expiry	2017-12-28
Quantity	25 kgs	Packages Size	25kg/drum		
Storage condition	Keep the container tightly closed in a dry and well-ventilated indoor area, away from light or heat or moisture				

ITEM	STANDARD	RESULT
Characteristics	White to yellowish white powder	Conforms
Solubility	Swells in cold water	Conforms
Identification	Conforms	Conforms
Acidity 3.0% (w/v)	4.5-7.0	PH5.5
Loss on drying	Not more than 15.0%	6.0%
Sulphated ash	Not more than 0.6%	0.2%
Iron	Not more than 0.002%	<0.002%
Oxidizing substances	Conforms	Conforms
Microbial limits	Conforms	Conforms
Sulphur dioxide	Not more than 50ppm	20ppm
Foreign matter	Conforms	Conforms

Conclusion: The above results comply with BP2011.

Analysis:	Luyan	Checker	Lin Jiayin	Supervisor	Yujie
Signature		Signature		Signature	

A member of

[illegible]NGBAO CHEMICALS CO.,LTD
[illegible] Hangzhou, 310030 China
[illegible] E-mail: info@chongbaochemical.com
[illegible]chongbaochemical.com

PRODUCT INSTRUCTION

Product Name: PREGELATINIZED STARCH (STARCH 1500)

Specification:

ITEM	STANDARD
Acidity 3.0% (w/v)	4.5-7.0
Loss on drying	Not more than 15.0%
Sulphated ash	Not more than 0.6%
Iron	Not more than 0.002%
Oxidizing substances	Conforms
Microbial limits	Conforms
Sulphur dioxide	Not more than 50ppm
Foreign matter	Conforms

Physical properties

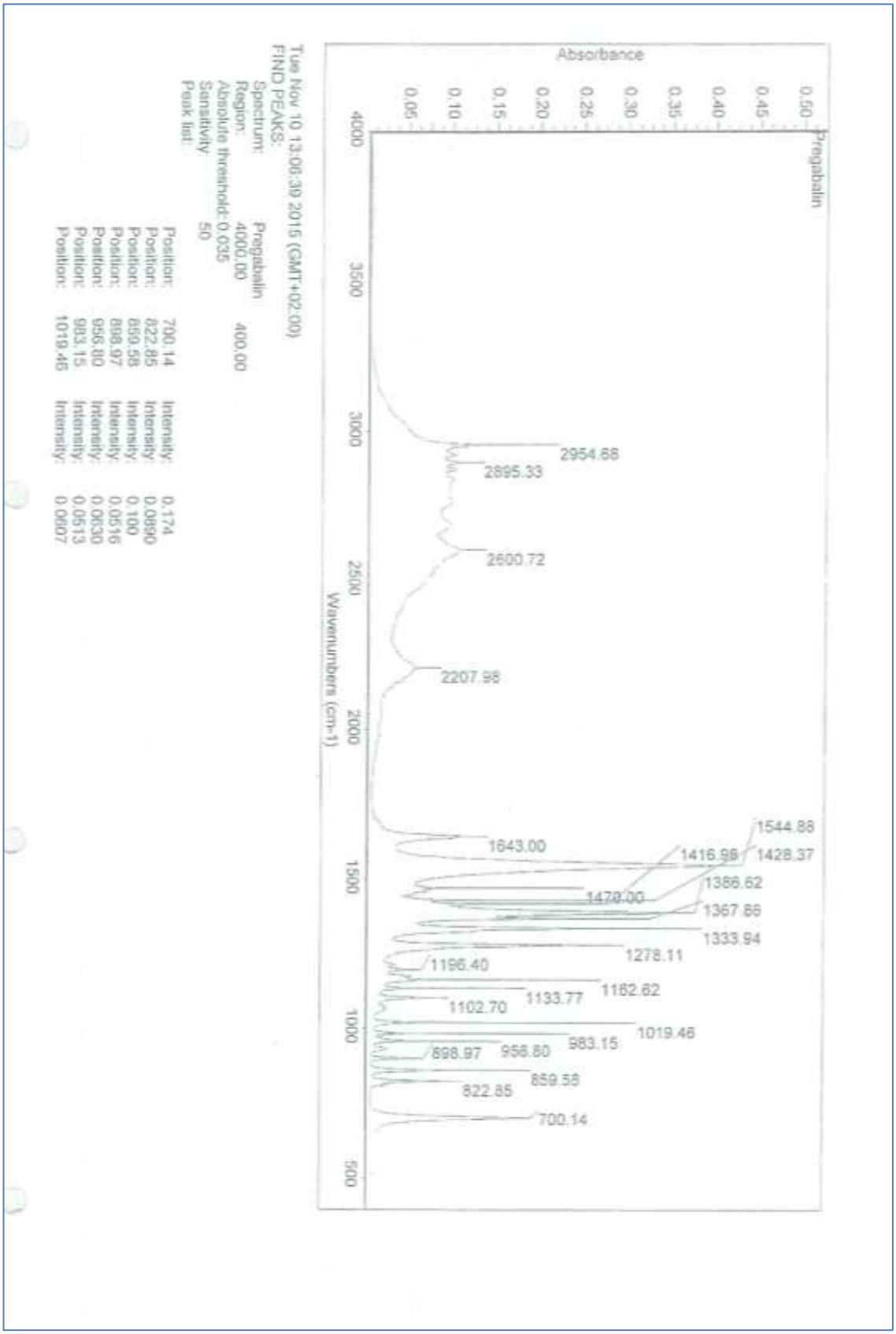
Character: White powders, no smell, tasteless; insoluble in Organic solvents; parts dissoluble in cold water.

Application:

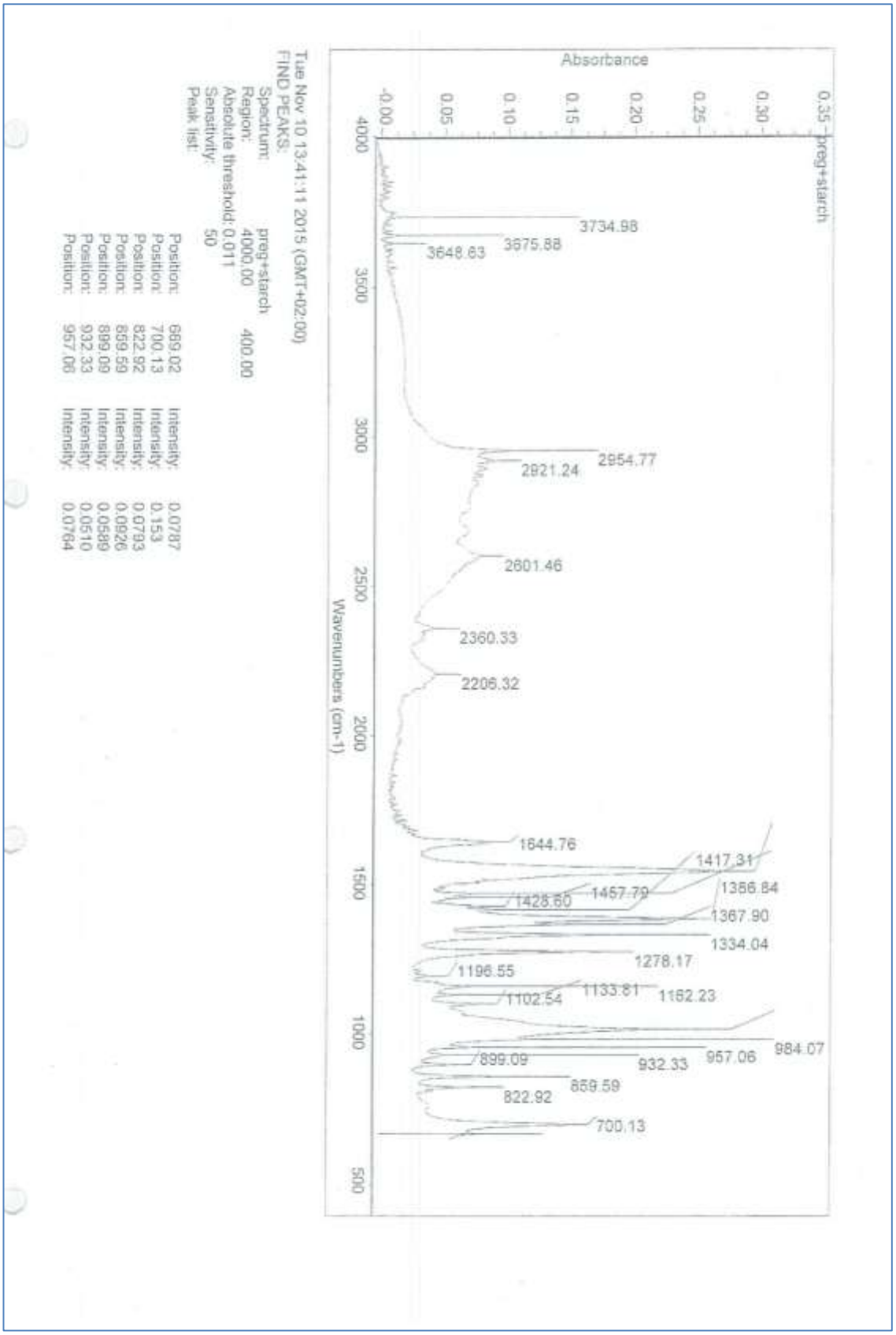
The product is mainly used as the adjuvant of solid medicine preparation.It has better fluidity and lubrication. It can be incused into flakes directly as powders, dry bonding agent of Troches, disintegrating agent, wet means manufacture grains bonging agent, packing agent. The use quantum is 5 to 80 percent.It also can be used capsule diluting agent, disintegrating agent.

Appendix 2

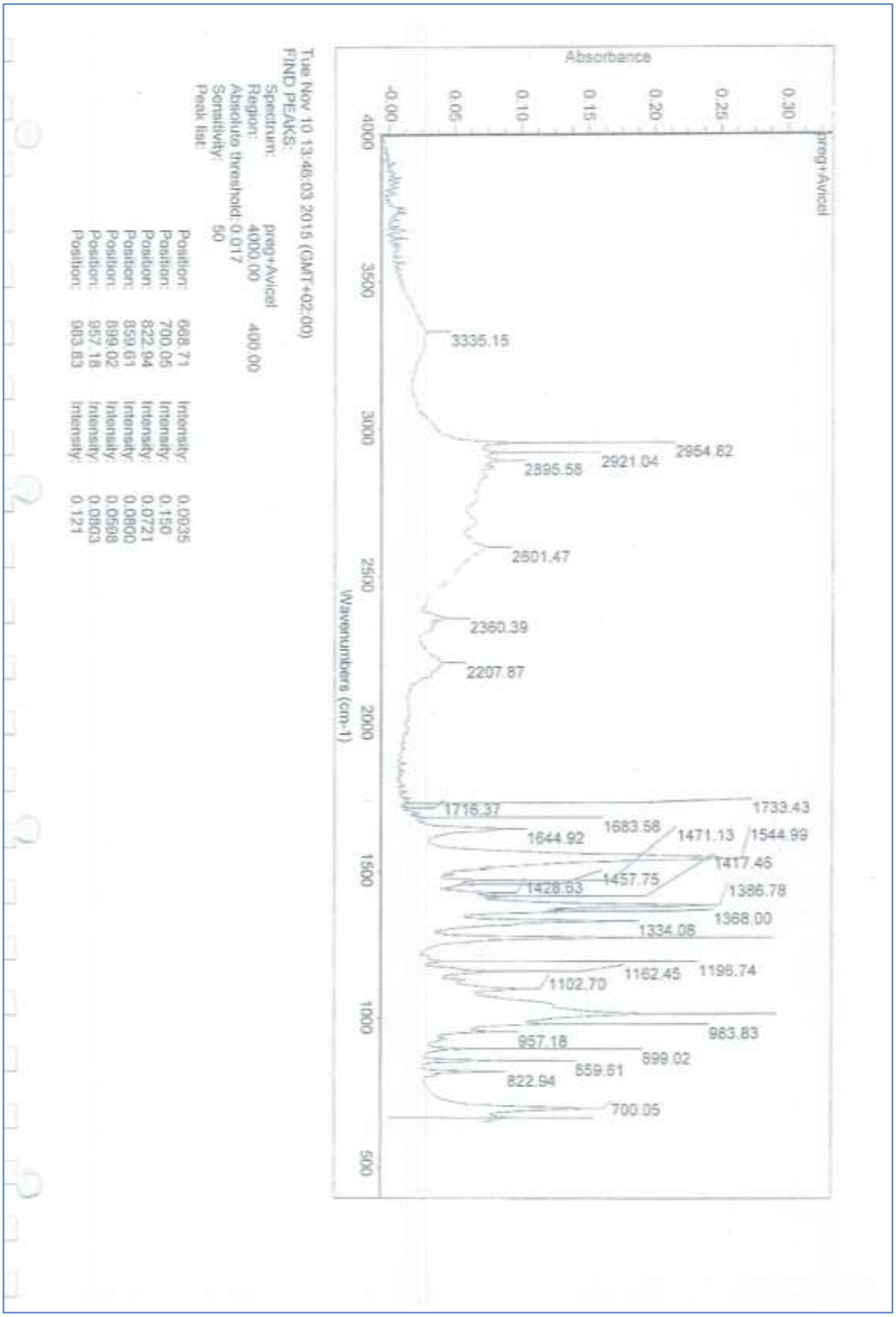
Compatibility Study FTIR Spectra


Tue Nov 10 13:06:39 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: Pregabalin
Region: 4000.00 400.00
Absolute threshold: 0.035
Sensitivity: 50
Peak list:
Position: 700.14 Intensity: 0.174
Position: 822.85 Intensity: 0.0890
Position: 659.58 Intensity: 0.100
Position: 898.97 Intensity: 0.0516
Position: 956.80 Intensity: 0.0630
Position: 983.15 Intensity: 0.0513
Position: 1019.46 Intensity: 0.0607
Absorbance
0.05
0.10
0.15
0.20
0.25
0.30
0.35
0.40
0.45
0.50
Pregabalin
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
2954.68
2895.33
2600.72
2207.98
1643.00
1544.88
1416.98
1428.37
1386.62
1478.00
1367.86
1333.94
1278.11
1196.40
1162.62
1133.77
1102.70
1019.46
983.15
956.80
898.97
859.58
822.85
700.14

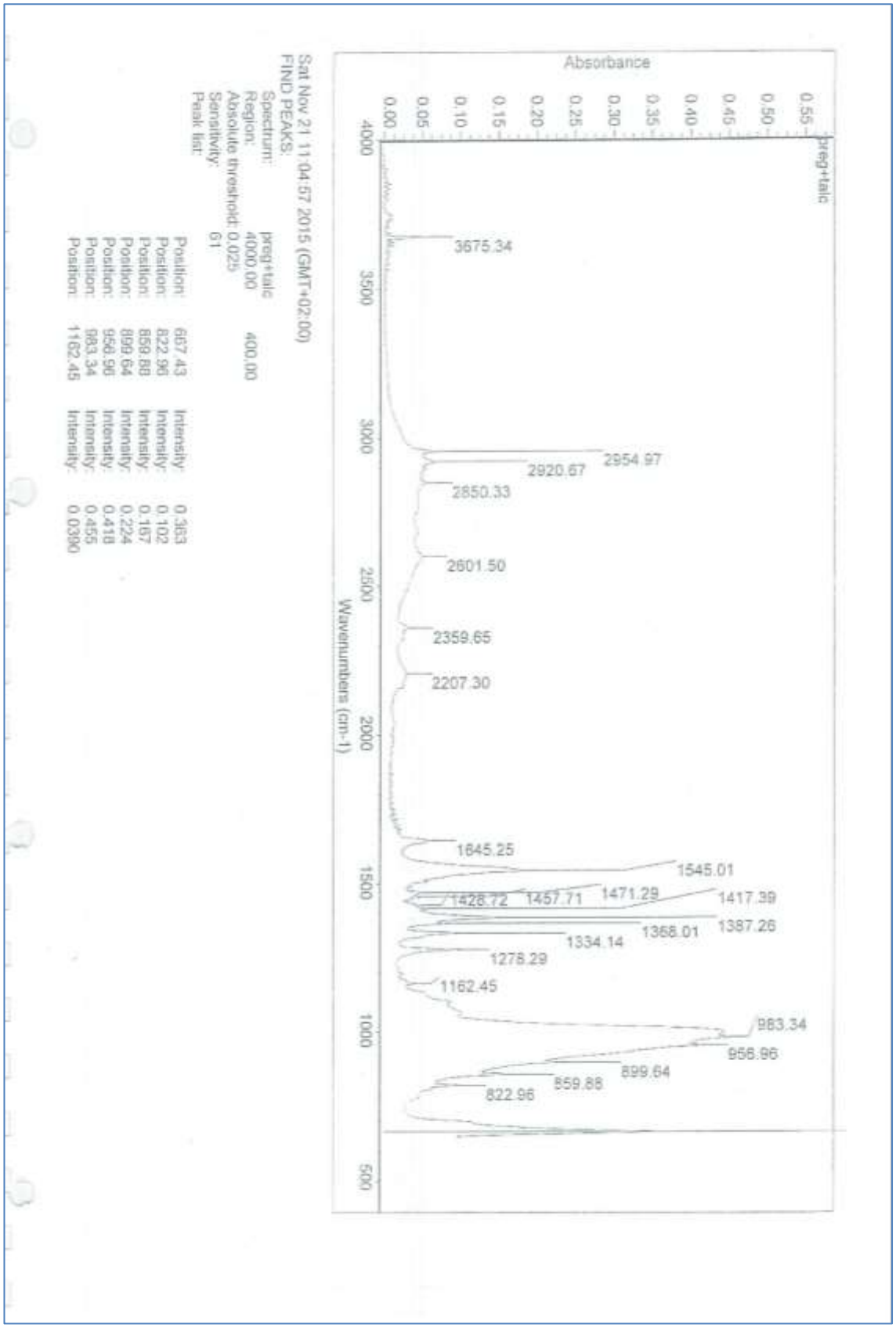
Position:	1102.70	Intensity:	0.0699
Position:	1133.77	Intensity:	0.0492
Position:	1162.62	Intensity:	0.0804
Position:	1196.40	Intensity:	0.0492
Position:	1278.11	Intensity:	0.274
Position:	1333.94	Intensity:	0.251
Position:	1367.86	Intensity:	0.213
Position:	1386.62	Intensity:	0.360
Position:	1416.98	Intensity:	0.123
Position:	1428.37	Intensity:	0.105
Position:	1470.00	Intensity:	0.0888
Position:	1544.68	Intensity:	0.402
Position:	1643.00	Intensity:	0.113
Position:	2207.98	Intensity:	0.0588
Position:	2600.72	Intensity:	0.110
Position:	2895.33	Intensity:	0.106
Position:	2954.68	Intensity:	0.130


Tue Nov 10 13:41:11 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: preg+starch
Region: 4000.00 400.00
Absolute threshold: 0.011
Sensitivity: 50
Peak list:
Position: 669.02 Intensity: 0.0787
Position: 700.13 Intensity: 0.153
Position: 822.92 Intensity: 0.0793
Position: 859.59 Intensity: 0.0926
Position: 899.09 Intensity: 0.0589
Position: 932.33 Intensity: 0.0510
Position: 957.06 Intensity: 0.0764
Absorbance
-0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
preg+starch
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
3734.98
3675.88
3648.63
2954.77
2921.24
2601.46
2360.33
2206.32
1644.76
1417.31
1366.84
1457.79
1428.60
1367.90
1334.04
1278.17
1196.55
1133.81
1102.54
1162.23
984.07
957.06
932.33
899.09
859.59
822.92
700.13

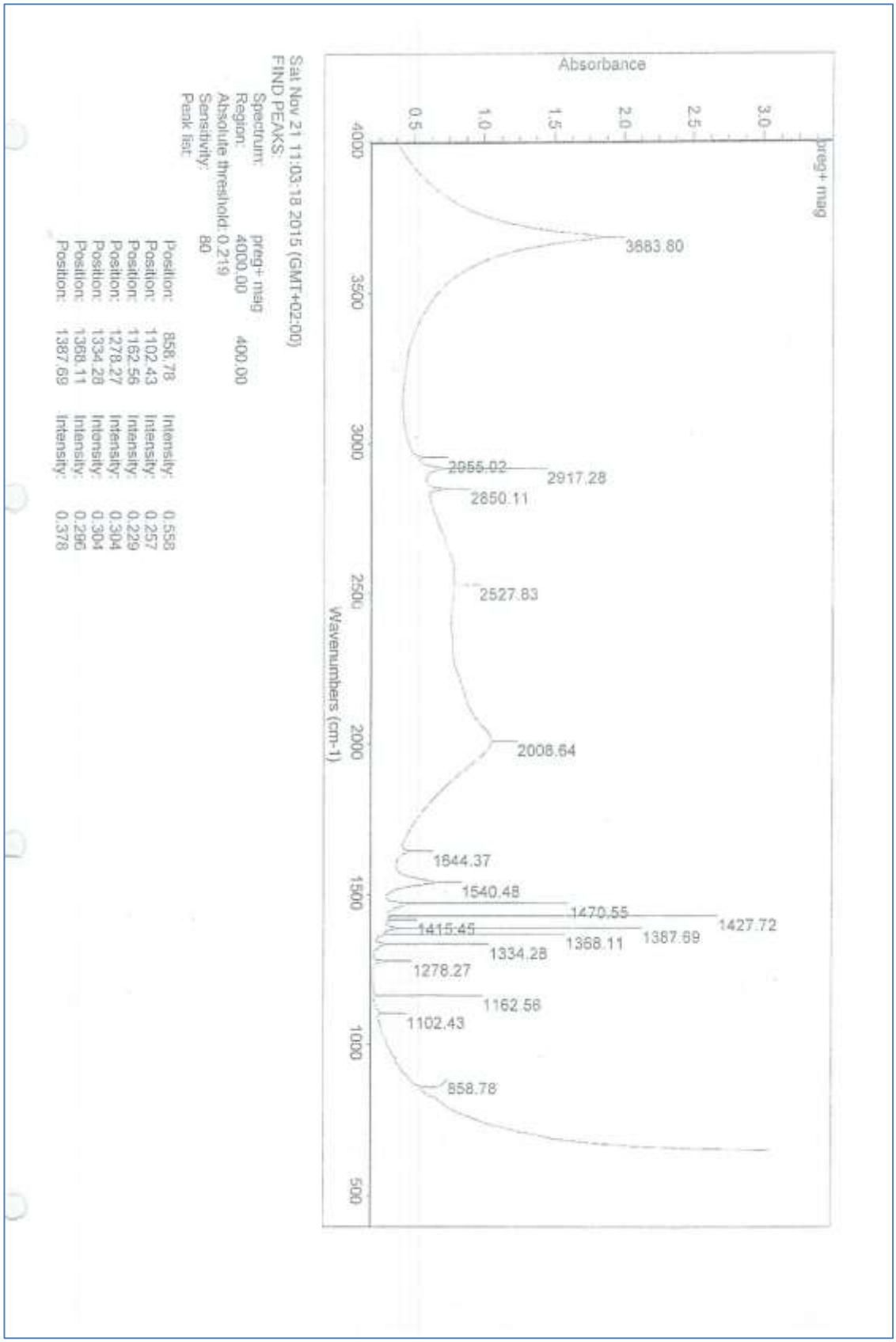
Position:	984.07	Intensity:	0.121
Position:	1018.26	Intensity:	0.212
Position:	1102.54	Intensity:	0.0798
Position:	1133.81	Intensity:	0.0592
Position:	1162.23	Intensity:	0.0719
Position:	1196.55	Intensity:	0.0409
Position:	1278.17	Intensity:	0.183
Position:	1334.04	Intensity:	0.174
Position:	1367.90	Intensity:	0.156
Position:	1386.84	Intensity:	0.250
Position:	1417.31	Intensity:	0.0958
Position:	1428.60	Intensity:	0.0861
Position:	1457.79	Intensity:	0.0705
Position:	1470.94	Intensity:	0.0733
Position:	1545.19	Intensity:	0.270
Position:	1644.76	Intensity:	0.0894
Position:	2206.32	Intensity:	0.0431
Position:	2360.33	Intensity:	0.0409
Position:	2601.46	Intensity:	0.0769
Position:	2921.24	Intensity:	0.0898
Position:	2954.77	Intensity:	0.0995
Position:	3648.63	Intensity:	0.0146
Position:	3675.88	Intensity:	0.0153
Position:	3734.98	Intensity:	0.0130


Tue Nov 10 13:46:03 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: preg+Avicel
Region: 4000.00 400.00
Absolute threshold: 0.017
Sensitivity: 50
Peak list:
Position: 668.71 Intensity: 0.0935
Position: 700.05 Intensity: 0.150
Position: 822.94 Intensity: 0.0721
Position: 859.61 Intensity: 0.0800
Position: 899.02 Intensity: 0.0598
Position: 957.18 Intensity: 0.0803
Position: 983.83 Intensity: 0.121
Absorbance
0.00
0.05
0.10
0.15
0.20
0.25
0.30
preg+Avicel
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
3335.15
2954.82
2921.04
2895.58
2601.47
2360.39
2207.87
1733.43
1716.37
1683.58
1644.92
1544.99
1471.13
1457.75
1417.46
1428.93
1386.78
1368.00
1334.08
1196.74
1162.45
1102.70
983.83
957.18
899.02
859.61
822.94
700.05

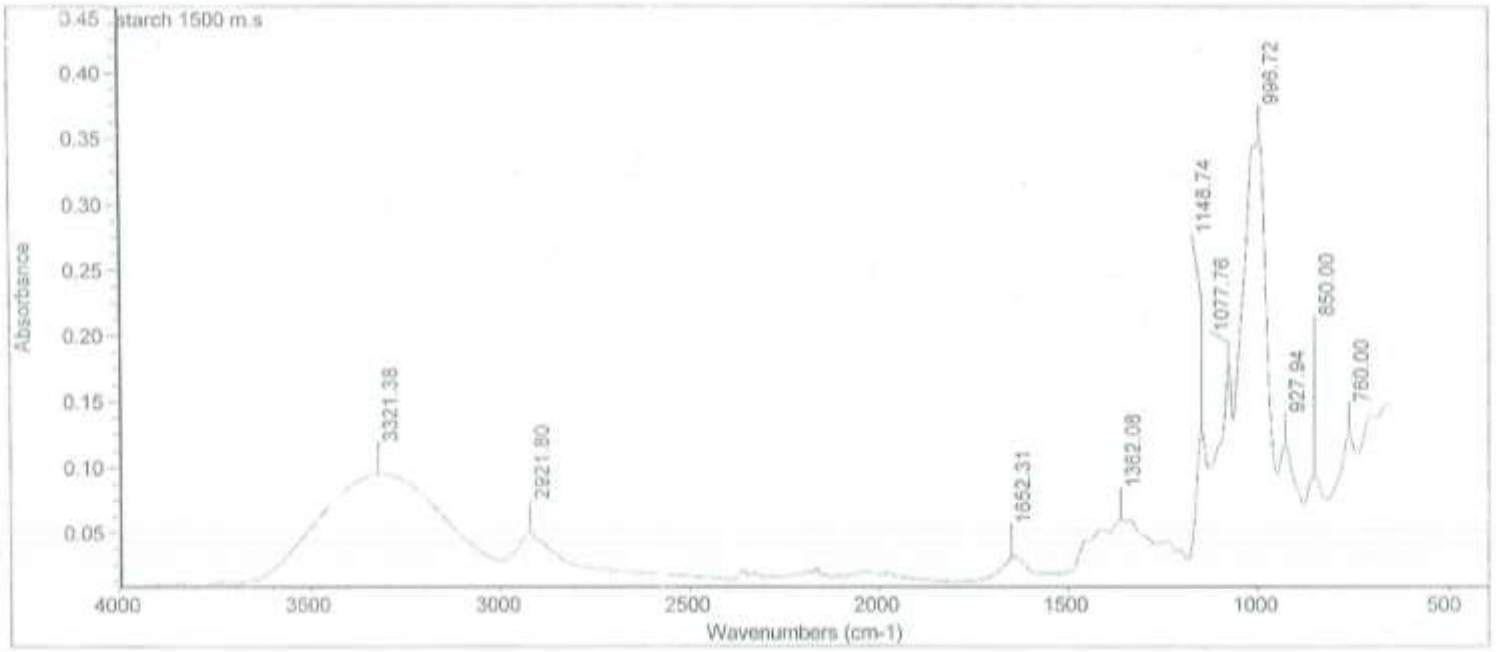
Position:	1019.01	Intensity:	0.191
Position:	1102.70	Intensity:	0.102
Position:	1162.45	Intensity:	0.0814
Position:	1196.74	Intensity:	0.0419
Position:	1278.20	Intensity:	0.176
Position:	1334.08	Intensity:	0.170
Position:	1369.00	Intensity:	0.152
Position:	1386.78	Intensity:	0.237
Position:	1417.46	Intensity:	0.0913
Position:	1428.63	Intensity:	0.0844
Position:	1457.75	Intensity:	0.0684
Position:	1471.13	Intensity:	0.0688
Position:	1544.99	Intensity:	0.253
Position:	1644.92	Intensity:	0.0947
Position:	1683.58	Intensity:	0.0299
Position:	1716.37	Intensity:	0.0221
Position:	1733.43	Intensity:	0.0183
Position:	2207.87	Intensity:	0.0400
Position:	2360.39	Intensity:	0.0419
Position:	2601.47	Intensity:	0.0735
Position:	2895.58	Intensity:	0.0820
Position:	2921.04	Intensity:	0.0824
Position:	2954.82	Intensity:	0.0941
Position:	3335.15	Intensity:	0.0278


Sat Nov 21 11:04:57 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: preg+talc
Region: 4000.00 400.00
Absolute threshold: 0.025
Sensitivity: 61
Peak list:
Position: 667.43 Intensity: 0.363
Position: 822.96 Intensity: 0.102
Position: 859.88 Intensity: 0.157
Position: 899.64 Intensity: 0.224
Position: 956.96 Intensity: 0.418
Position: 983.34 Intensity: 0.455
Position: 1162.45 Intensity: 0.0390
Absorbance
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
0.40
0.45
0.50
0.55
preg+talc
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
3675.34
2954.97
2920.67
2850.33
2601.50
2359.65
2207.30
1645.25
1545.01
1471.29
1457.71
1428.72
1417.39
1387.26
1368.01
1334.14
1278.29
1162.45
983.34
956.96
899.64
859.88
822.96

Position:	1278.29	Intensity:	0.105
Position:	1334.14	Intensity:	0.0978
Position:	1368.01	Intensity:	0.0874
Position:	1387.26	Intensity:	0.154
Position:	1417.39	Intensity:	0.0604
Position:	1428.72	Intensity:	0.0530
Position:	1457.71	Intensity:	0.0488
Position:	1471.29	Intensity:	0.0514
Position:	1545.01	Intensity:	0.181
Position:	1845.25	Intensity:	0.0602
Position:	2207.30	Intensity:	0.0282
Position:	2359.65	Intensity:	0.0305
Position:	2601.50	Intensity:	0.0492
Position:	2850.33	Intensity:	0.0551
Position:	2920.67	Intensity:	0.0665
Position:	2954.97	Intensity:	0.0662
Position:	3675.34	Intensity:	0.0582


Absorbance
0.5
1.0
1.5
2.0
2.5
3.0
preg+ mag
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
3683.80
2955.02
2917.28
2850.11
2527.83
2008.64
1644.37
1540.48
1470.55
1427.72
1415.45
1387.69
1368.11
1334.28
1278.27
1162.56
1102.43
858.78
Sat Nov 21 11:03:18 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: preg+ mag
Region: 4000.00 400.00
Absolute threshold: 0.219
Sensitivity: 80
Peak list:
Position: 858.78 Intensity: 0.558
Position: 1102.43 Intensity: 0.257
Position: 1162.56 Intensity: 0.229
Position: 1278.27 Intensity: 0.304
Position: 1334.28 Intensity: 0.304
Position: 1368.11 Intensity: 0.296
Position: 1387.69 Intensity: 0.378

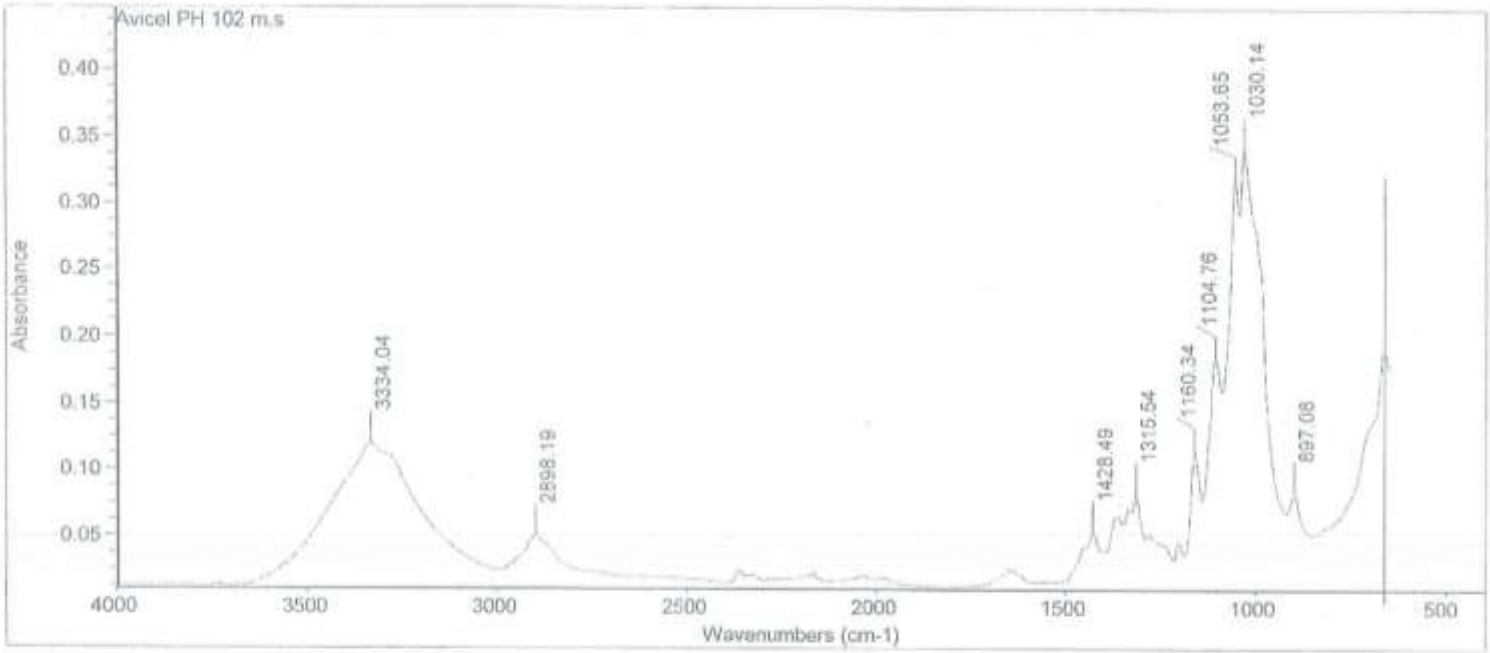
Position:	1415.45	Intensity:	0.333
Position:	1427.72	Intensity:	0.344
Position:	1470.55	Intensity:	0.440
Position:	1540.48	Intensity:	0.654
Position:	1644.37	Intensity:	0.444
Position:	2068.64	Intensity:	1.056
Position:	2527.83	Intensity:	0.780
Position:	2850.11	Intensity:	0.716
Position:	2917.28	Intensity:	0.700
Position:	2955.02	Intensity:	0.550
Position:	3683.80	Intensity:	1.823



Mon Nov 09 12:57:03 2015 (GMT+02:00)
FIND PEAKS:

Spectrum:	starch 1500 m.s		
Region:	4000.00	400.00	
Absolute threshold:	0.021		
Sensitivity:	50		
Peak list:			
Position:	760.00	Intensity:	0.125
Position:	850.00	Intensity:	0.0930
Position:	927.94	Intensity:	0.117
Position:	996.72	Intensity:	0.352
Position:	1077.76	Intensity:	0.180
Position:	1148.74	Intensity:	0.128
Position:	1362.08	Intensity:	0.0600

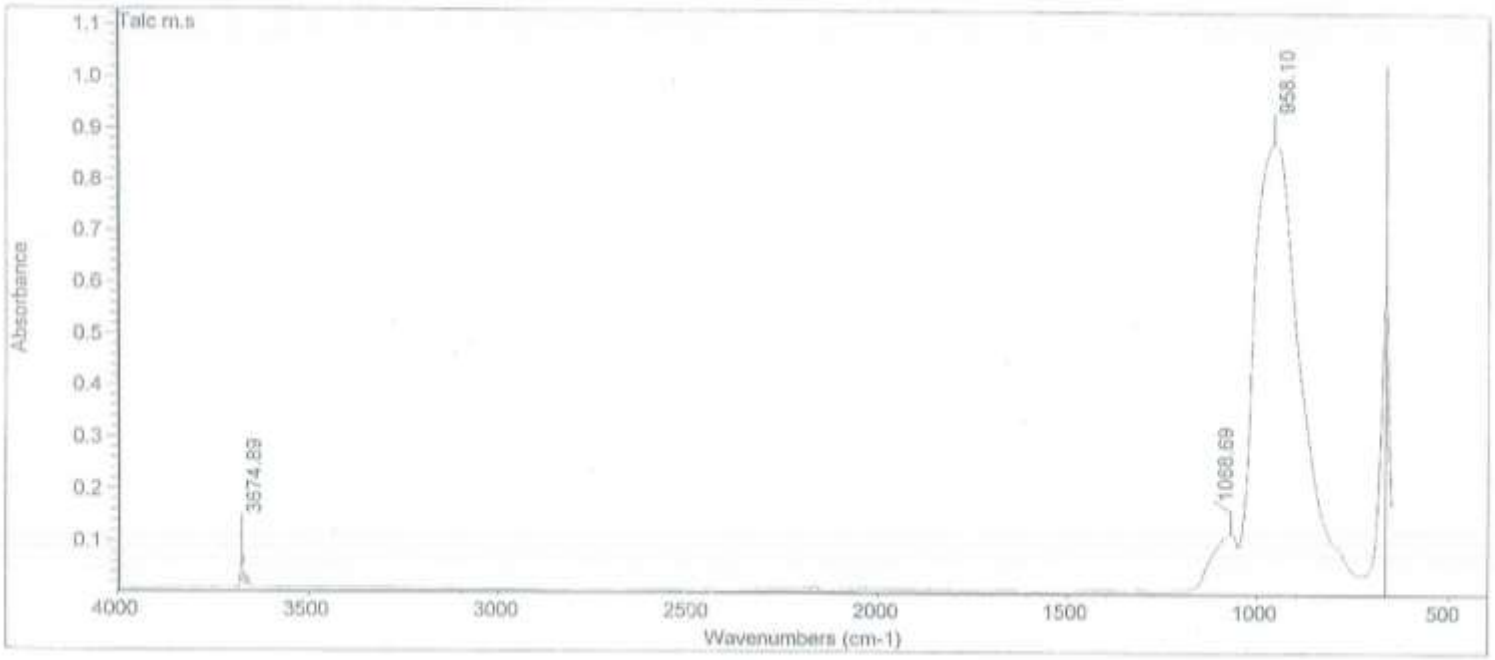
Position: 1652.31 Intensity: 0.0314
Position: 2921.80 Intensity: 0.0502
Position: 3321.38 Intensity: 0.0044



Mon Nov 09 12:59:35 2015 (GMT+02:00)
FIND PEAKS:

Spectrum:	Avicel PH 102 m.s		
Region:	4000.00	400.00	
Absolute threshold:	0.024		
Sensitivity:	50		
Peak list:			
Position:	663.06	Intensity:	0.194
Position:	897.08	Intensity:	0.0832
Position:	1030.14	Intensity:	0.343
Position:	1053.65	Intensity:	0.322
Position:	1104.76	Intensity:	0.186
Position:	1160.34	Intensity:	0.117
Position:	1315.54	Intensity:	0.0823

Position: 1428.49 Intensity: 0.0530
Position: 2898.19 Intensity: 0.0494
Position: 3331.04 Intensity: 0.120

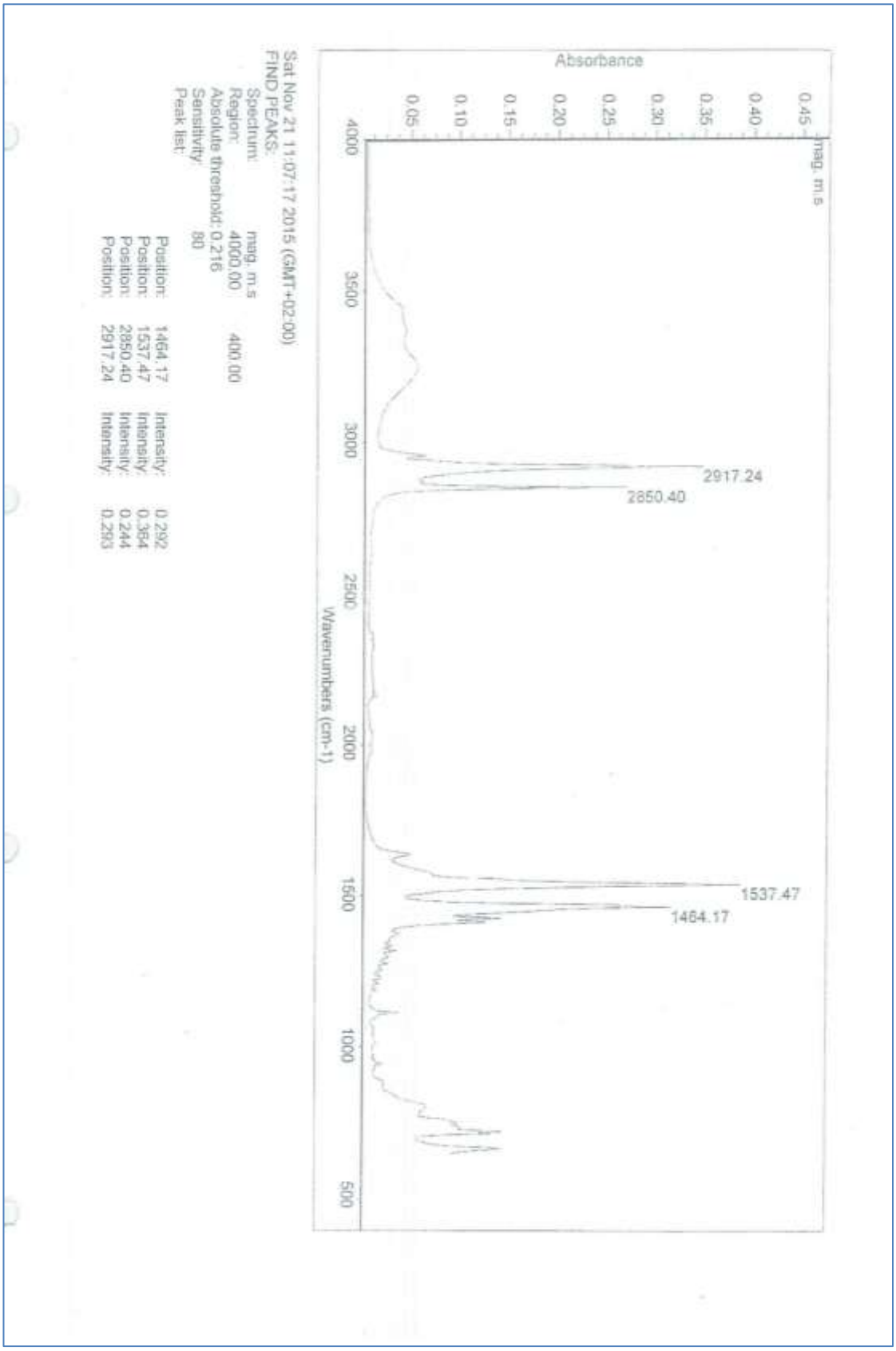


Sat Nov 21 11:08:11 2015 (GMT+02:00)
FIND PEAKS:

Spectrum:	Talc m.s	
Region:	4000.00	400.00
Absolute threshold:	0.028	
Sensitivity:	61	

Peak list:

Position:	666.14	Intensity:	0.823
Position:	958.10	Intensity:	0.879
Position:	1068.69	Intensity:	0.121
Position:	3674.89	Intensity:	0.0909


Absorbance
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
mag. m.s
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
2917.24
2850.40
1537.47
1464.17
Sat Nov 21 11:07:17 2015 (GMT+02:00)
FIND PEAKS:
Spectrum: mag. m.s
Region: 4000.00 400.00
Absolute threshold: 0.216
Sensitivity: 80
Peak list:
Position: 1464.17 Intensity: 0.292
Position: 1537.47 Intensity: 0.364
Position: 2850.40 Intensity: 0.244
Position: 2917.24 Intensity: 0.293

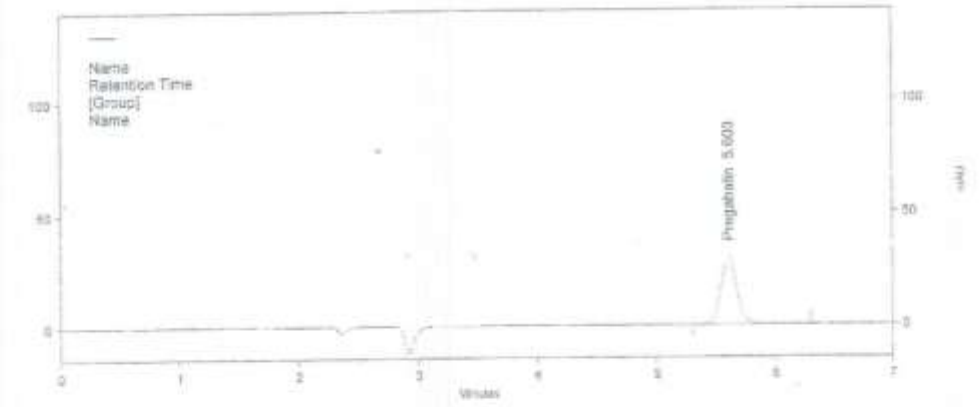
Appendix 3

Typical Chromatograms of Stability Study

File Nam: C:\EZChrom Elite\Enterprise\Projects\General\Data\Pregabalin Tab\191115\007-Rep3.dat
Sample I.D 007
Run Time: 19/11/2015 10:13:30
Analysis Time 19/11/2015 10:20:37
Method Name: C:\EZChrom Elite\Enterprise\Projects\General\Method\Pregabalin.met
Injection Volume: 20
Vial Number: 107
Data Description: STD ASSAY
Print Time: 10/12/2015 09:21:25

Pregabalin 75 mg Tab B.No.150915
Pregabalin 300 mg Tab B.No.150914
Stability"After 14 Days"

Mobile Phase:Buffer PH6.9:CH3CN(94:6)
Column:C18,250*4.6 mm i.d
Detection Wavelength:210nm
Flow Rate:1.5ml/min



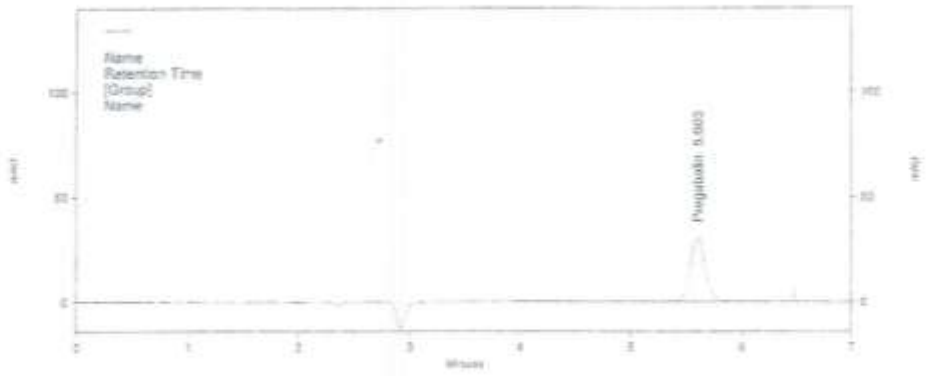
UV Results Pk #	Retention Time	Name	Area	Theoretical plates (USP)	Asymmetry	Area Percent
1	5.603	Pregabalin	1101837	9486	1.06784	100.000
Totals			1101837			100.000

Analyzed By:

File Name: C:\EZChrom Elite\Enterprise\Projects\General\Data\Pregabalin Tab\191115\009.dat
Sample I.D 009
Run Time: 19/11/2015 10:21:37
Analysis Time 19/11/2015 10:28:49
Method Name: C:\EZChrom Elite\Enterprise\Projects\General\Method\Pregabalin.met
Injection Volume: 20
Vial Number: 108
Data Description: ASSAY B.No.150915 At 25C+60%RH
Print Time: 10/12/2015 09:21:43

Pregabalin 75 mg Tab B.No.150915
Pregabalin 300 mg Tab B.No.150914
Stability"After 14 Days"

Mobile Phase:Buffer PH6.9:CH3CN(94:6)
Column:C18,250*4.6 mm i.d
Detection Wavelength:210nm
Flow Rate:1.5ml/min



UV Results

Pk #	Retention Time	Name	Area	Theoretical plates (USP)	Asymmetry	Area Percent
1	5.603	Pregabalin	1139181	8439	1.06548	100.000
Totals			1139181			100.000

Analysed By:

Appendix 4

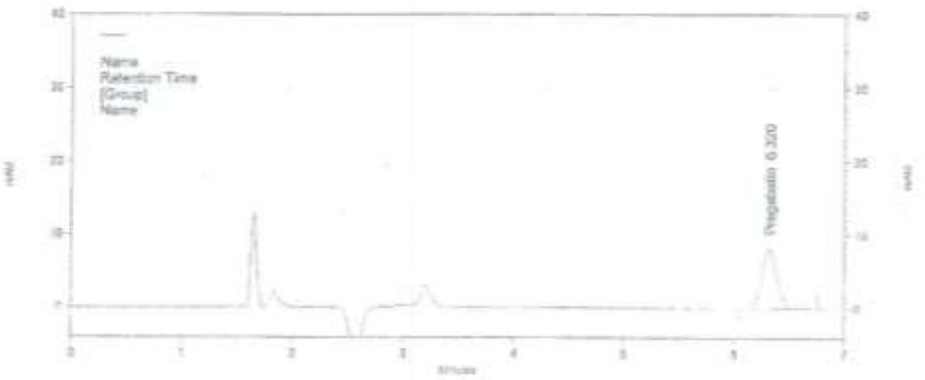
Typical Chromatograms for

Biowaiver Study

File Nam: C:\EZChrom Elite\Enterprise\Projects\General\Data\Pregabalin Tab\091115\031.dat
Sample I.D 031
Run Time: 09/12/2015 20:05:08
Analysis Time 09/12/2015 20:12:41
Method Name: C:\EZChrom Elite\Enterprise\Projects\General\Method\Pregabalin.met
Injection Volume: 20
Vial Number: 129
Data Description: DISS 3 B.No.150914 After 10min In Phosph.Buffer PH6.8
Print Time: 10/12/2015 10:49:41

Pregabalin 75 mg Tab B.No.150915
Pregabalin 300 mg Tab B.No.150914
Dissolution Profile"Phosphate Buffer PH6.8"

Mobile Phase:Buffer PH6.9:CH3CN(94:6)
Column:C18,250*4.6 mm i.d
Detection Wavelength:210nm
Flow Rate:1.5ml/min



UV Results

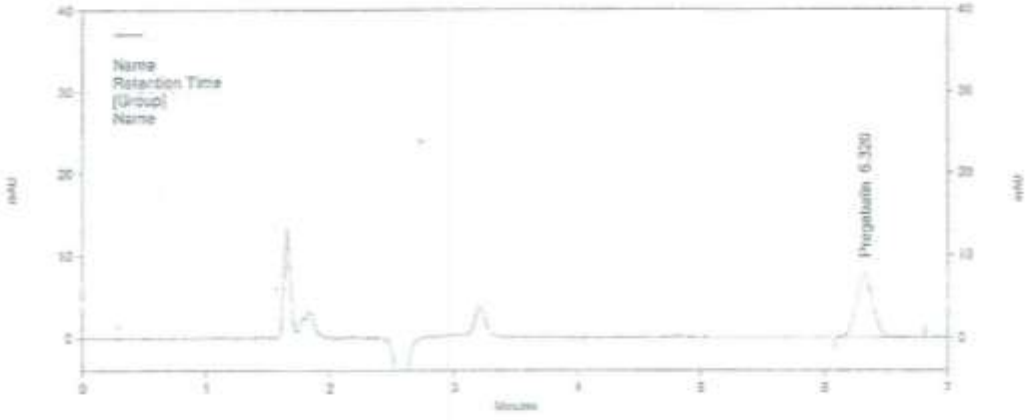
Pk #	Retention Time	Name	Area	Theoretical plates (USP)	Asymmetry	Area Percent
1	6.320	Pregabalin	302019	10460	1.04783	100.000
Totals			302019			100.000

Analyzed By:

File Nam: C:\EZChrom Elite\Enterprise\Projects\General\Data\Pregabalin Tab\091215\029.dat
Sample I.D 029
Run Time: 09/12/2015 19:47:48
Analysis Time 10/12/2015 10:49:23
Method Name: C:\EZChrom Elite\Enterprise\Projects\General\Method\Pregabalin.met
Injection Volume: 20
Vial Number: 127
Data Description: DISS 1 B.No.150914 After 10min In Phosph.Buffer PH6.8
Print Time: 10/12/2015 10:49:24

Pregabalin 75 mg Tab B.No.150915
Pregabalin 300 mg Tab B.No.150914
Dissolution Profile"Phosphate Buffer PH6.8"

Mobile Phase:Buffer PH6.9:CH3CN(94:6)
Column:C18,250*4.6 mm i.d
Detection Wavelength:210nm
Flow Rate:1.5ml/min



UV Results

Pk #	Retention Time	Name	Area	Theoretical plates (USP)	Asymmetry	Area Percent
1	6.320	Pregabalin	285586	10500	1.04963	100.000
Totals			285586			100.000

Analyzed By:

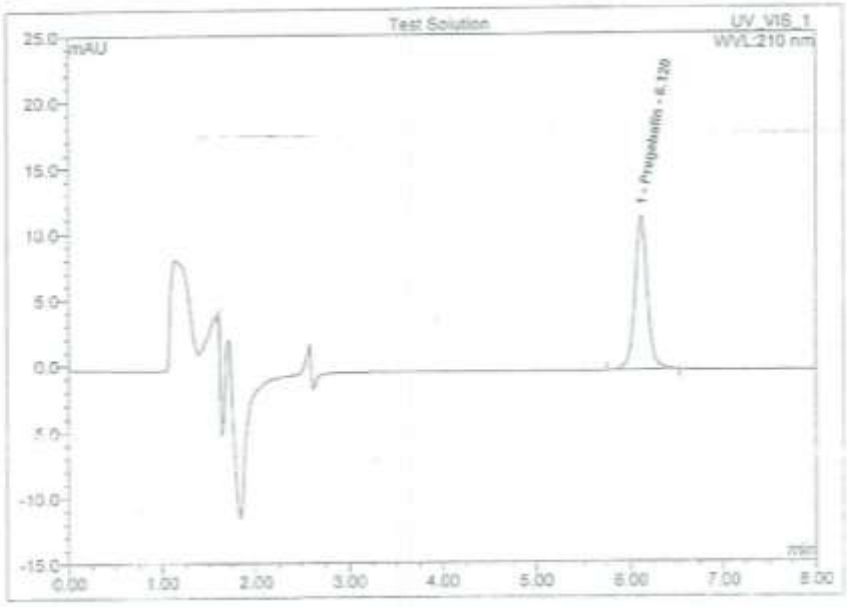
Appendix 5

Typical Chromatograms for Test methods Validation of Pregabalin Tablet

2 Test Solution

Precision Test — Nominal standard solution of Pregabalin – Rep.1

Sample Name:	Test Solution	Injection Volume:	20.0
Vial Number:	BA1	Channel:	UV_VIS_1
Sample Type:	unknown	Wavelength:	210
Control Program:	Pregabalin Tablet	Bandwidth:	n.a.
Quantif. Method:	Pregabalin Tablet	Dilution Factor:	1.0000
Recording Time:	11-7-15 14:20	Sample Weight:	1.0000
Run Time (min):	8.00	Sample Amount:	1.0000



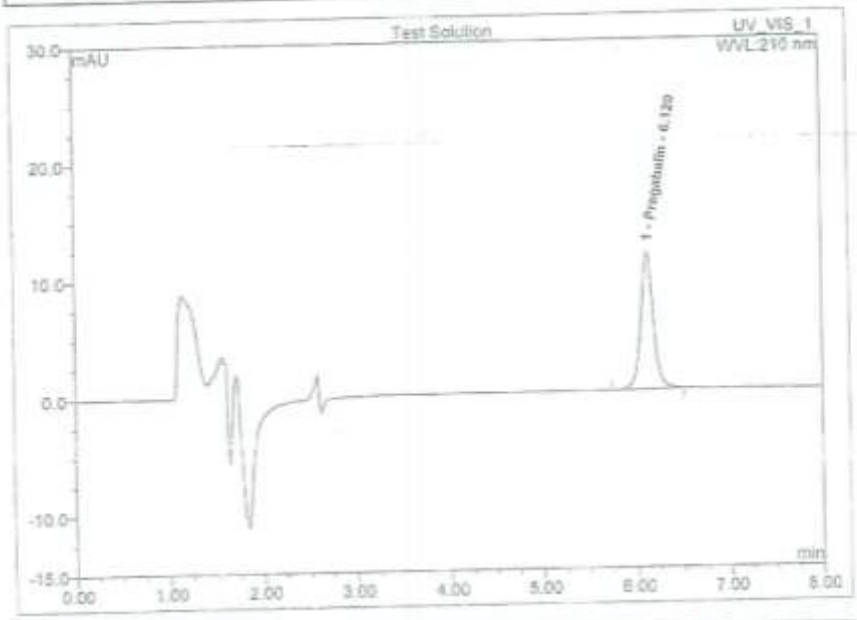
No.	Ret.Time min	Peak Name	Height mAU	Area mAU*min	Rel.Area %	Peak Asy.	Type
1	6.12	Pregabalin	11.651	1.791	100.00	1.01	BMB*
Total:			11.651	1.791	100.00	1.007	

default.lntegration

3 Test Solution

Precesion Test — Nominal standard solution of Pregabalin – Rep.2

Sample Name:	Test Solution	*Injection Volume:*	20.0
Vial Number:	BA1	*Channel:*	UV_VIS_1
Sample Type:	unknown	*Wavelength:*	210
Control Program:	Pregabalin Tablet	*Bandwidth:*	n.a.
Quantif. Method:	Pregabalin Tablet	*Dilution Factor:*	1.0000
Recording Time:	11-7-15 14:29	*Sample Weight:*	1.0000
Run Time (min):	8.00	*Sample Amount:*	1.0000



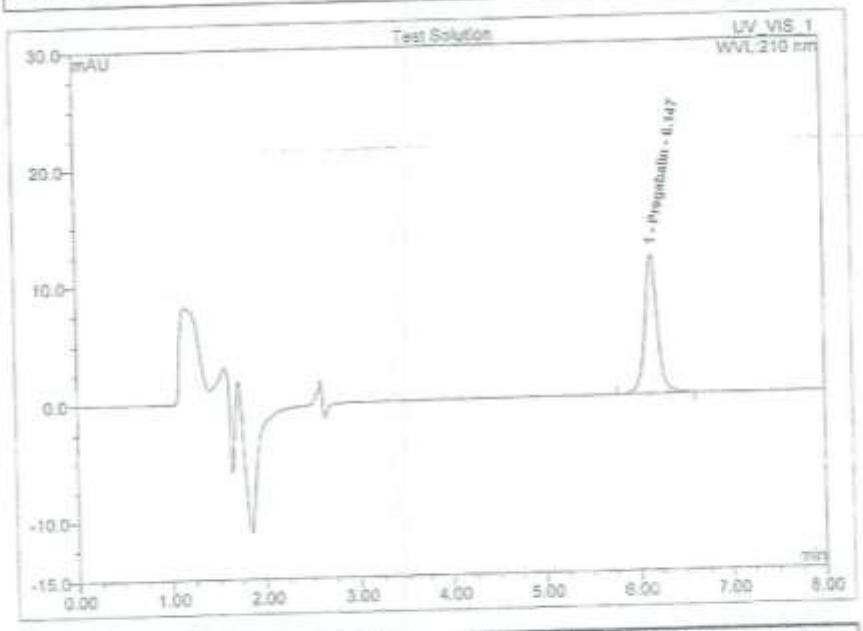
No.	Ret.Time min	Peak Name	Height mAU	Area mAU*min	Rel.Area %	Peak Asy.	Type
1	6.12	Pregabalin	11.649	1.798	100.00	1.02	BM9*
Total:			11.649	1.798	100.00	1.020	

default/Integration

4 Test Solution

Precision Test --- Nominal standard solution of Pregabalin -- Rep.3

Sample Name:	Test Solution	*Injection Volume:*	20.0
Vial Number:	BA1	*Channel:*	UV_VIS_1
Sample Type:	unknown	*Wavelength:*	210
Control Program:	Pregabalin Tablet	*Bandwidth:*	n.a.
Quantif. Method:	Pregabalin Tablet	*Dilution Factor:*	1.0000
Recording Time:	11-7-15 14:38	*Sample Weight:*	1.0000
Run Time (min):	8.00	*Sample Amount:*	1.0000



No.	Ret.Time min	Peak Name	Height mAU	Area mAU*min	Rel.Area %	Peak Asy.	Type
1	6.15	Pregabalin	11.674	1.804	100.00	1.01	BMB*
Total:			11.674	1.804	100.00	1.009	

default/Integration

تطوير وتقييم شكل صيدلاني جديد بخاصية التحرر الفوري (أقراص بريغابالين)

إعداد: محمد سميح شفيق شناعة

إشراف: د. نعمان مالكية

الملخص

مادة بريغبالين هي مادة دوائية تستخدم لعلاج الالم من مصدر(neuropathy) لدى الكبار ولمعالجة الالم العضلي الليفي وتتوفر في الأسواق بعدة تراكيز على شكل كبسولات و محاليل فموية.

الغرض من هذه الدراسة هو تطوير وتقييم أقراص صيدلانية لهذه المادة بخاصية التحرر الفوري , على شكل صيدلاني جديد يحقق شروط التكافئ الحيوي مع المستحضر المعتمد عالميا على شكل كبسولات (Lyrica).

تم تحليل وتقييم المادة الفعالة (Pregabalin) بناء على مواصفة وطريقة تحليل الشركة المصنعة للمادة الفعالة, أما تحليل الشكل الصيدلاني الجديد (الأقراص) فقد تم تطوير طريق تحليل كروماتوجرافي, بما يشمل التثبت من طرق الفحص (نسبة التحرر, المحتوى, الشوائب).

تم إجراء كافة التحاليل والتجارب المتعلقة بالتحليل الكروماتوجرافي باستعمال جهاز كاشف بالاشعة فوق البنفسجية على طول موجي 210 نانوميتر و عمود فصل كروماتوجرافي رقم (C18, 25cm*4.6mm, 5-102 µm) والمحلول الناقل (phosphate buffer pH6.9:Acetonitrile) بنسبة (94:6) بمعدل تدفق 1.5 ملم لكل دقيقة...

تم تطوير الصيغة النهائية للمستحضر مرورا بسلسة من الخطوات تشمل: إختيار مواد غير فعالة بناء على دراسات التوافق (Compatibility Study) بين المادة الفعالة والمواد غير الفعالة وتعريضها الى ظروف بيئية صارمة لتحفز اي تفاعلات كيميائية محتملة او تغييرات فيزيائية, ثم الى مرحلة التقييم باستخدام جهاز التحليل بالاشعة تحت الحمراء (FTIR), حيث تبين ان المواد المستخدمة (سيليلوز, نشا, تالك, مغنسيوم ستيرات) تتوافق مع المادة الفعالة.

بالانتقال الى المرحلة الثانية من تجارب التصنيع المخبري تبين ان طريقة الكبس المباشر فعالة للوصول الى الاقراص المستهدفة , وذلك لعدة صيغ وتراكيز مختلفة من المواد سابقة الذكر, ومن ثم اختيار الصيغة الاكثر توافقا مع معايير التقييم الاولي, والتي تبحث في خصائص الانضغاط, السيولة, الكثافة, القساوة والتفكك وذلك لكل صيغة على حدة.

لقد تبين ان الصيغة المختارة (F5) لديها الخصائص الافضل, حيث طبقت لانتاج تركيزين مختلفين من اقراص بريغبالين وهما 300ملغم و 75 ملغم, بحيث تتناسب وزنة القرص مع التركيز المستهدف, ومن ثم تغليف المستحضر ببوليمر بولي فينيل الكحول في وسط مائي لتغلب على خصائص بريغبالين الحسية ولتحسين ثباتية المستحضر.

تمت مرحلة التغليف الاساسي باستخدام غلاف (PVC-Aluminum) ثم وضع المستحضر تحت دراسة الثياتية تحت ظروف اختبار (45-25) درجة مئوية وعلى رطوبة (75-60)% لفترات مختلفة لتقييم ثباتية المستحضر تحت ظروف الدراسة ظويلة الامد والدراسة المتسارعة حسب متطلبات FDA وقد اثبتت هذه الدراسة استقرار الشكل الصيدلاني المطور وعدم حصول اي انحرافات حادة أو جوهرية خلال فترة الدراسة.

اجري التكافئ الحيوي الاحلالي (Biowaiver Study) بين اقراص بريغبالين (75 & 300) ملغم المطورة والمستحضر المرجع المعتمد عالميا (Lyrica capsules 300mg & 75mg) حسب متطلبات مؤسسة الغذاء والدواء الامريكية على جهاز التحرر باستخدام Paddle على سرعة 50 دورة في الدقيقة في عدة محاليل على درجة حموضة (1.2, 4.5, 6.8). وقد تبين ان نسبة التحرر في الشكلين الصيدلانيين –كبسولات وأقراص– في جميع التجارب والمحاليل قد تجاوز 85% خلال 15 دقيقة من الفحص, وبناء عليه فان اقراص بريغبالين تكافئ المسحضر الاصيل بيولوجيا بالاضافة الى خصائص الاستقرار المصاحبة لهذا الشكل الدوائي, مما يدعم العمل على تسجيل وتسويق المستحضر على شكل اقراص فورية التحرر.