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Review Article

Development and Evaluation of Poly Herbal Orodispersible Films

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ABSTRACT

Diabetics was found to be most common chronic disease. The cause of diabetes is due to various reasons like genetics, lifestyle or many more. The present study focuses on the formulation and evaluation of Orodispersible films by incorporating medicinal plant extracts such as Annona squamosa, Boerhavia diffusa, Momordica charantia, Morinda citrifolia and along with pharmaceutical excipients including hydroxypropyl methylcellulose (HPMC), ascorbic acid and gelatine by using solvent casting method and prepared five formulations with different concentration of materials and evaluated for phytochemical tests and physicochemical parameters like colour, odour, texture, weight variation, pH, folding endurance, swelling index, disintegration test, dissolution test. The physicochemical evaluation of developed formulations had shown pale-green and pale-yellow colour, mild odour, soft texture, and the weight variation ranging from 0.19 ± 0.1 to 0.28 ± 0.2 , the pH of the films are around 6.0, the folding endurance is ≥ 300 , the swelling index ranges from 31.8 ± 2.5 to 3.59 ± 2.1 , the disintegration time ranges between 32 to 36 seconds. The maximum amount of % drug release of the formulations shown are F3-95.52, F4-93.72. So, our Oro dispersible films can be successfully used in future for the anti-diabetic purposes after the confirmation of clinical and toxicological studies for commercial production in the market.

INTRODUCTION

Diabetic mellitus is taken from the Greek word diabetes, meaning siphon- to pass through and the Latin word mellitus sweet. The term diabetes was first used by Apollonius of Memphis around 250 to 300 BC. Ancient Greek, Indian, and Egyptian

civilizations discovered the sweet nature of urine in the condition, and hence the propagation of the word diabetes mellitus came into being. Mering and Minkowski, in 1889 discovered the role of pancreas in the pathogenesis of diabetes. In 1992 Banting, Best, and James Bertram collip purified the hormone insulin from the pancreas of cows at

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the diabetes in 1992 ^[1]. Diabetic mellitus is a chronic metabolic disorder characterized by a high blood glucose concentration hyperglycemia (fasting plasma glucose more than 7.0 mmol/L, or plasma glucose more than 11.1 mmol/L 2 hours after the meal) caused by insulin deficiency, often combine with insulin resistance. Hyperglycemia occurs because of uncontrolled hepatic glucose output and reduced uptake of glucose by skeleton muscle with reduced glycogen synthesis. When the renal threshold for glucose reabsorption is exceeded, glucose spills over into urine (Glycosuria) and causes an osmotic diuresis (polyurea), which, in turn, results in dehydration, thirst and increased drinking (Polydipsia). Insulin deficiency causes wasting through increased breakdown and reduced synthesis of proteins. Diabetic ketoacidosis is acute emergency. Develops in the absence of insulin because of accelerated breakdown of fat to acetyl-CoA, which, absence of aerobic carbohydrate metabolism, is converted to acetoacetate and beta hydroxy butyrate (which causes acidosis) and acetone (a ketone).

Pancreatic Islet Hormones:

The islets of Langerhans contain 4 main cell types all of which secrete peptide hormones: B or β cells secrete insulin, α cells secrete glucagon, δ cells secrete somatostatins, and PP (gamma or F cells) cells secrete pancreatic polypeptide. The core of each islet contains mainly the predominant β cells surrounded by mantle of α cells interspersed with δ cells or PP cells. In addition to insulin, β cells secrete a peptide known as islets amyloid polypeptide or amylin which delays gastric emptying and opposes insulin by stimulating glycogen breakdown in striated muscle. Glucagon also opposes insulin, increasing blood glucose and stimulating protein breakdown in muscle. Somatostatin inhibits secretion of insulin and glucagon. It is widely distributed outside the pancreas and is also release from the hypothalamus, inhibiting the release of growth hormone from pituitary.

Insulin:

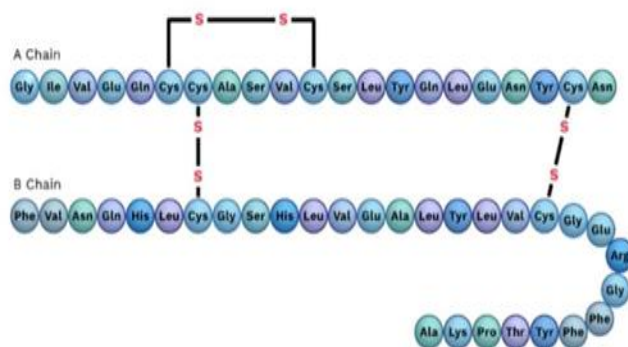


Figure 1: Structure of Insulin.

Insulin was the first protein for which an amino acid sequence was determined. It consists of two polypeptide chains (A and B of 21 and 30 amino acid residues, respectively).

Synthesis and Secretion:

Like other peptide hormones insulin is synthesized as a precursor (pre proinsulin) in rough

endoplasmic reticulum. Pre-proinsulin is transported to the Golgi apparatus, where it undergoes proteolytic cleavage first two proinsulin and then to insulin plus a fragment of uncertain function called C-peptide^[2]. Insulin and C-peptide are stored in granules in β cells and normally co secreted by exocytosis in equimolar amounts together with smaller and variable amounts of proinsulin. The main factor controlling synthesis and secretion of insulin is the blood glucose concentration. β cells respond both to the absolute glucose concentration and to the rate of change of

blood glucose. Other stimuli to insulin release include amino acids and fatty acids the parasympathetic nervous system, peptide hormones for the gut and drugs that act on sulfonylurea receptors. There is a steady basal release of insulin and also a response to increase blood glucose. This response has two phases: an initial phase reflecting release of stored hormone, and slower, delayed phase reflecting both continued release of stored hormone and new synthesis. The response is abnormal in diabetes mellitus.

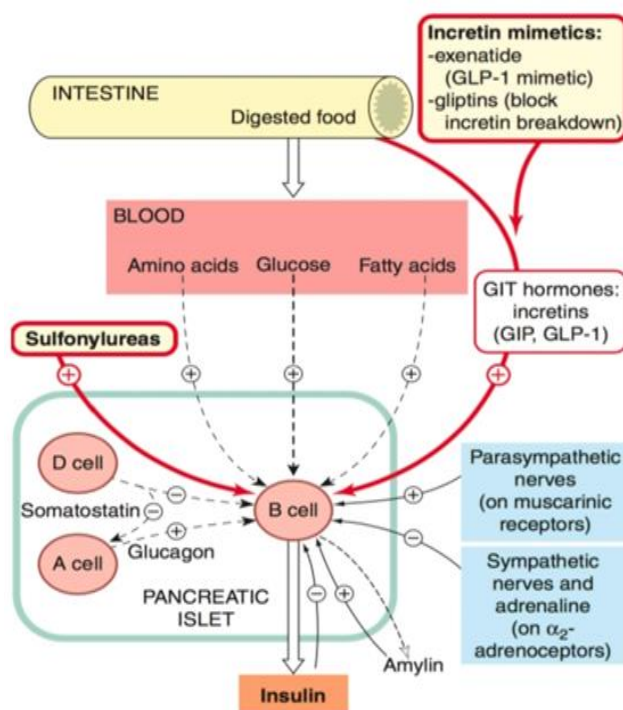


Figure 2: Factors regulating insulin secretion.

ATP sensitive potassium channel determines the resting membrane potential in β cells. Glucose enter β cells via a membrane transporter called Glut-2, and its subsequent metabolism via glucokinase and glycolysis increase intracellular ATP. This blocks potassium ATP channel, causing membrane depolarization and opening of voltage dependent calcium channels leading to calcium influx. The resulting increase in cytoplasmic

calcium triggers insulin secretion, but only in the presence of amplifying messengers including diacylglycerol, non-esterified arachidonic acid and 12-lipoxygenase products of arachidonic acid. Phospholipases are commonly activated by calcium, but free arachidonic acid is liberated in β cells by an ATP sensitive calcium in sensitive phospholipase. Consequently, in β cells calcium entry and arachidonic acid production are both

driven by ATP, linking cellular energy status to insulin secretion.

Effects of insulin on carbohydrates metabolism:

Insulin influences glucose metabolism in most of the tissues, especially the liver, where it inhibits glycogenolysis and gluconeogenesis while stimulating glycogen synthesis. It also increases glucose utilization, but overall effects are to increase hepatic glycogen stores. In muscles, unlike liver, uptake of glucose is slow and is the rate-limiting step in carbohydrates metabolism. The main effects of insulin are to increase facilitated transport of glucose via a transport called Glut-4, and to stimulate glycogen synthesis and glycolysis. Insulin increases glucose uptake by Glut-4 in adipose tissue as well as in muscle, enhancing glucose metabolism. One of the main end products of glucose metabolism in adipose tissue is glycerol, which is esterified with fatty acids to form triglycerides thereby affecting fat metabolism.

Effects of insulin on fat metabolism:

Insulin increases synthesis of fatty acids and triglycerides in adipose tissue and in liver. It inhibits lipolysis, partly via dephosphorylation (and hence inactivation) of lipases. It also inhibits the lipolytic actions of adrenaline, growth hormone and glucagon by opposing their actions on adenylate cyclase.

Effects of insulin on protein metabolism:

Insulin stimulates uptake of amino acids into muscle and increases protein synthesis; it so decreases protein catabolism and inhibits oxidation of amino acids in the liver^[3].

Prediabetics:

Prediabetes is defined as a state of abnormal glucose homeostasis in which blood glucose levels are elevated above those considered normal, but not high enough to meet the criteria required for a diagnosis of diabetes^[4,5]. It is characterized by impaired fasting glucose or impaired glucose tolerance. Evidence increasingly demonstrates that prediabetes is a toxic state, in addition to being a risk factor for diabetes^[6]. Emerging evidence suggests that prediabetes is associated with pathophysiological changes in several tissues and organs, which would support its recognition as a distinct pathological entity. Prediabetes level ranges from 70 mg/dL to 99 mg/dL in patients with prediabetes, except to see blood glucose levels elevated between 110 mg/dL to 125 mg/dL. In addition to type 2 diabetes, prediabetes is a risk factor for the development of cardiovascular disease, and stroke. Once diagnosed with prediabetes, patients should be checked for progression to type 2 diabetes every one to two years. If screening is negative for prediabetes, repeated screening should be carried out every 3 years as per the United States Prevention Services



Figure 3: Prediabetic chart

Task Force (USPSTF). Lifestyle changes through improved nutrition and physical activity are the first line treatment for preventing the transition from prediabetes to diabetes which can be as high as 70%.

Polyherbal treatment for diabetes mellitus:

It is well known fact that type 2 diabetic mellitus patients suffer from higher blood glucose concentration with a greater possibility of abnormal metabolism of carbohydrates, fat, and protein than healthy individuals. Therefore, urea and creatine are considered useful and simple biomarkers as for predicting or assessing renal function in the diabetic patient. However, the fasting blood glucose is ≥ 126 mg/dL is an easy option although it must be evaluated with multiple tests including repeated fasting blood glucose on different occasion to confirm type 2 diabetic mellitus. WHO collaborates assists health ministries in establishing mechanisms for the introduction of traditional plant medicines into primary healthcare programs, in assessing safety and efficacy, in ensuring adequate supplies, and in the quality control of raw materials. Herbal formulations in general can be standardized schematically a to formulate the medicament using raw materials is collected from different localities

and conditions and a comparative chemical efficacy of different batches of formulation is to be observed ^[7]. A preparation with better clinical efficacy has to be selected. The routine physical, phytochemical and pharmacological, organoleptic, physiochemical parameters are to finished product and to validate the whole manufacturing process ^[8]. *Allium sativum*, *Boerhavia diffusa*, *Anacardium occidentale*, *Momordica charantia*, *Coriandrum sativum*, *Ocimum tenuiflorum*, *Murraya koenigii*, *Allium fistulosum*, are well known herbs available throughout India including neighboring Countries and they are commonly used for the treatment of various diseases including diabetic mellitus.

Oral films:

The conventional delivery system shows various problems like gastro intestinal irritation, very low concentration of drug in blood and incomplete absorption of drug from gastro intestinal tract and mainly poor patient compliance ^[9,10]. To avoid such problems and to achieve maximum therapeutic efficacy ^[11,12], preparation of mouth dissolving film of water-soluble drug is better alternative which gives improved patient compliance and rapid onset of action due to disintegration of film in saliva and pre gastric absorption of drug.

Types of oral film preparations:

Oral films are prepared by different methods which includes

1. Solvent casting method
2. Hot melt extrusion
3. Electrostatic spinning
4. Printing
5. Electrostatic spray deposition

Solvent casting method:

It is one of the most commonly used methods for the formulation of film. It is prepared using water soluble polymers, excipients and drug. Due to the application of high shear force a homogenous mixture is formed. The solution obtained is poured into foil spread with coating knife to obtain uniform thickness^[13].

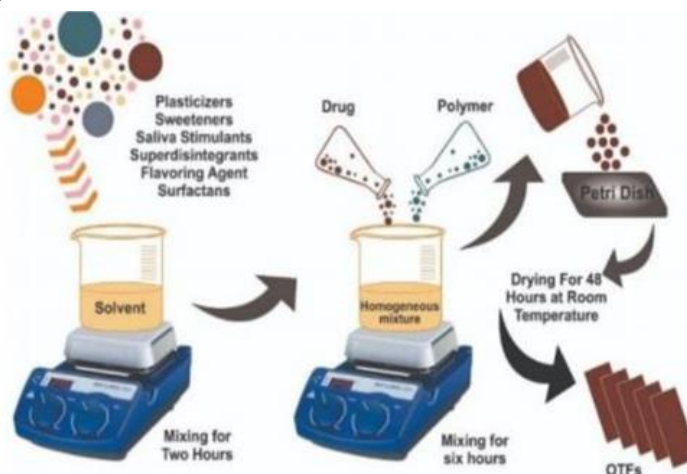


Figure 4: Process of Solvent casting method.

Hot melt extrusion:

Hot melt extrusion is essential method for shaping thin films from molten mixture of raw materials and excipients. During the preparation process, the active ingredients film-forming material and other excipients are mixed and heated to their

melting point to create a homogenous molten mixture. This mixture is forced through a perforated die with external pressure to form desired film. Finally the film is chopped, chilled, and packed to produce the final product. Finally, the film is chopped, chilled and packed to produce final product^[14].

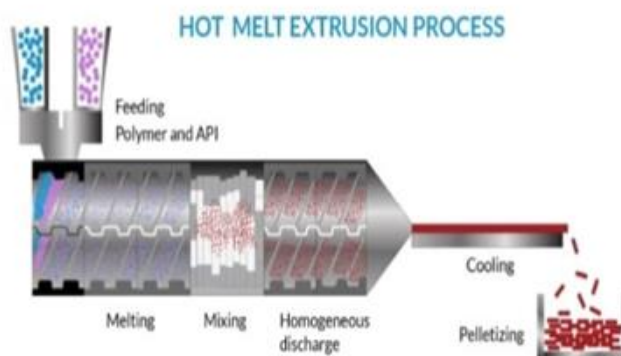


Figure 5: Hot Melt Extrusion Process

Electrostatic spinning:

Electrostatic spinning is technique to generate fibrous structures through elongation of electrified

jets with an external field. Its versatility produces fibers that are being utilized in the diverse field, namely tissue engineering, wound dressing, and controlled drug delivery^[15].

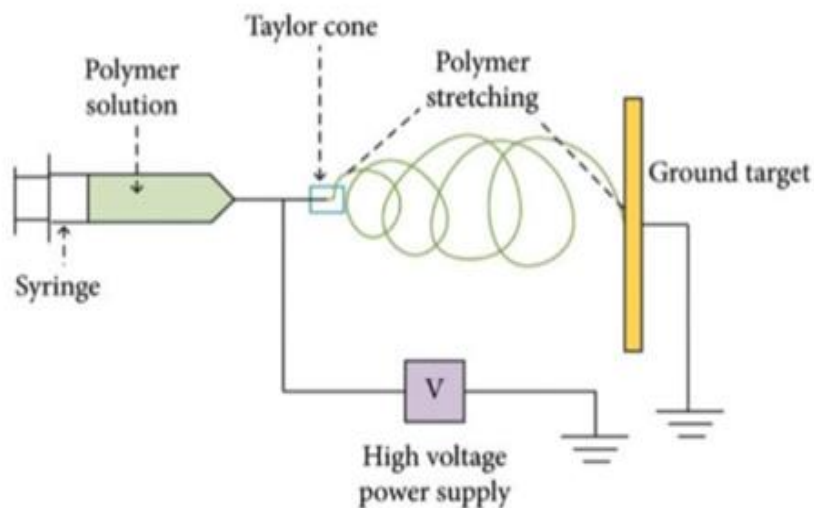


Figure 6: Electrostatic spinning method

MATERIALS AND METHODS:

Collection of materials:

Materials used in this film preparations are as follow:

Annona squamosa, *Boerhavia diffusa*, *Momordica charantia*, *Moorinda Citrifolia*, HPMC, Gelatine

Scientific name: *Annona squamosa*



Figure 7: *Annona Squamosa*

Uses^[16]:

1. Regulation of hyperthyroidism and lipid peroxidation.

2. It is used as anti-inflammatory, anti-oxidant, anti-diabetic and anti-lipidemic activity
3. It has diuretic properties which can help to flush out and reduce the risk of kidney stones and also shows anti-ulcer activity.
4. It also shows hepatoprotective activity

Scientific Name ^[17]: *Boerhavia diffusa*



Figure 8: *Boerhavia diffusa*

Uses:

1. It is used in treating obesity.
2. It is used effectively in treating the diseases called dropsy, a condition where excess of water fluid get accumulated in the tissue and body cavities.
3. Other benefits include treatment of anaemia, nerve weakness, paralysis, constipation and cough.
4. *Boerhavia diffusa* roots are used for anticonvulsant, analgesic, expectorant, laxative.
5. It promotes mucous removal from bronchial tubes and hence beneficial in treating of asthma.

Scientific name: *Momordica Charantia*



Figure 9: *Momordica charantia*

Uses:

1. It is used in Diabetes management.
2. It is used in gastrointestinal issues.
3. It is used in treating infections like malaria, cholera, and other infections.
4. It is used and anti-inflammatory and antioxidant.
5. It is used as anthelmintic to treat intestinal worms.

Scientific Name: *Morinda citrifolia*



Figure 10: *Morinda citrifolia*

Uses:

1. Noni is believed to have analgesic properties, potentially reducing joint pain, and other inflammatory conditions.

2. Noni has traditionally been used to support digestion and promote a healthy gut.

3. Noni is known as detoxifying agent, potentially cleansing the body from the inside out.

4. Noni has been linked to potential benefits like improved joint health, increased physical endurance, and support for maintaining normal blood pressure.

5. Noni helps to reduce inflammation and pain associated with various conditions.

HPMC:



Figure 11: HPMC

Uses:

1. For medications and supplement HPMC is used to coat pills and capsules forming a protective film that can help with drug release and improve the products appearance.
2. It acts as a binding agent in the production of tablets ensuring the granules stick together.
3. HPMC is an ingredient in eye drops and other ophthalmic solutions providing lubrication and moisture.

4. HPMC can be used to control the release of medication over time, allowing for more efficient and effective drug delivery.

Gelatine



Figure 12: Gelatine

Uses:

1. Gelatine is used as gelling agent in food.
2. Gelatine is used in beverages.
3. Gelatine is used in medications, drug or vitamin capsules.

Extraction of Herbals:

Maceration is a common solid crude drug extraction technique that involves selecting the polarity of the solvent and using heat and or agitation to increase the solubility of the chemicals of interest from the sample. Compare to other conventional and novel extraction procedures, it can be done with low cost and simple to use.

Steps involved in maceration:

1. Preparation

2. Soaking
3. Maceration
4. Separation
5. Concentration

Preparation

Select the plant part that to be used, clean and dry the plant part by shade drying and grind it to a fine powder.

Soaking

Select suitable solvent based on polarity, soak it for 3-7 days with regular agitation. The stoppered container is used to avoid evaporation of the solvent.

Maceration

The container is closed tightly and left undisturbed for a period, typically three days or longer, with occasional shaking or stirring to enhance extraction.

Separation

After the maceration period, the solvent is carefully separated from the solid material (marc) through filtration or decantation.

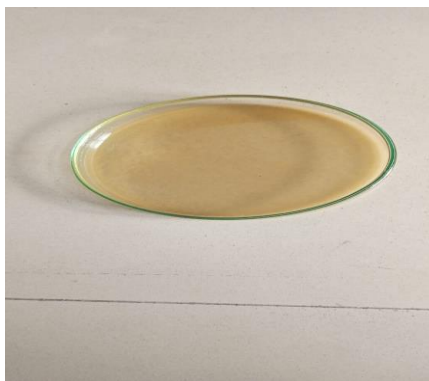
Concentration

The extracted liquid (macerate) can be further concentration using techniques like evaporation.

Preparation of Oral dispersion films:

Solvent casting method

Take the polymer and other excipients like saliva stimulating agents, plasticizer, surfactant dissolve in solvent (de ionised water) and mix for 15 minutes (part-1) and in another beaker take the active ingredients of suitable amounts and dissolve in solvent(part-2) and now add the part 2 to the part 1 and mix for 10



to 15 minutes by using magnetic stirrer, after acquiring the homogenous mixture pour the content into the petri plate and allow to dry in the hot air oven at 50-60°C of temperature for about 2 hours, after evaporation of solvent they are removed from oven and kept aside until it cooldown then the film in the petri plates is scrapped carefully with a scraper and are made into 2x2 cm² sized str

Figure 13:

Preparation of film

Evaluation of Oral dispersion films^[18]:

Physical evaluation: Physical characteristics were examined and noted. Films were evaluated for Physical evaluation colour, state, appearance, odour.

Thickness: The thickness is measured by using vernier callipers. The thickness of the film is measured at 5 points from the centre and for all the

Figure 15:
Digital pH meter

Surface pH test: Surface pH of the film



four corners. This is essential to ascertain uniformity in the thickness of the film as this directly related to the accuracy of dose in the strip.

can be determined by placing the film on the surface of 1.5% W/V agar gel followed by placing pH paper on the film. The change in the colour of pH paper is observed and reported.

Figure

14: Vernier calliper

Folding endurance: The flexibility of the film can be measured quantitatively in terms of folding endurance and is determined by repeatedly folding the film at 180° angle of the plane at the same plane until it breaks or folded at 300 times without breaking is computed as folding endurance value

Weight variation: Individual film should be weighed and the average weights are calculated. Then the average weight of the film. A large variation in weight indicates the inefficiency of the method employed and is likely to have non uniform drug content.

Measurement of pH: The pH of the film was tested using the Digital pH meter. One strip of film is mixed in 50 ml of distilled water and keep it for 1 hour and the pH was measured.

Swelling index: Buccal films were weighed individually (designed as W_1) and placed separately in 1% agar plates, incubated at $37^\circ\text{C} \pm 1^\circ\text{C}$ and examined for the physical changes. At regular 1 hour time intervals until 3 hours, patches were removed from the gel plates and excess surface water was removed carefully using filter paper. The swollen patches were then reweighed (W_2) and the swelling index (SI) was calculated using the following formula. The experiments were performed in triplicate, and the average values were reported.

$$\text{SI} = (W_2 - W_1) / W_1 \times 100$$

Disintegration test: Disintegration apparatus mentioned in official pharmacopoeias is used for determining the disintegration time of a film. Normally, the disintegration time is the function of composition of film as it varies with the formulation and generally ranges from 5 to 30s. Mostly, the USP disintegration apparatus is used for this test. There are no official guidelines

available for determining disintegration time orally fast disintegration films. There are two methods for determining disintegration of film:

Slide frame method: A drop of distilled water is poured on to the film clamped into slide frames placed on the petri dish. Time taken by the film to dissolve is noted.

Petri dish method: A film placed onto 2ml distilled water taken in the petri dish. Time taken

by the film to dissolve completely is considered as the disintegration time.

Dissolution test: The dissolution test for film is done through the paddle method. The dissolution medium will essentially be selected as per the sink conditions and highest dose of the API. Many time the dissolution test can be difficult due to the tendency of the strip to float onto the dissolution medium when the paddle apparatus is employed so, to avoid such conditions we use sinkers to make the film to float.

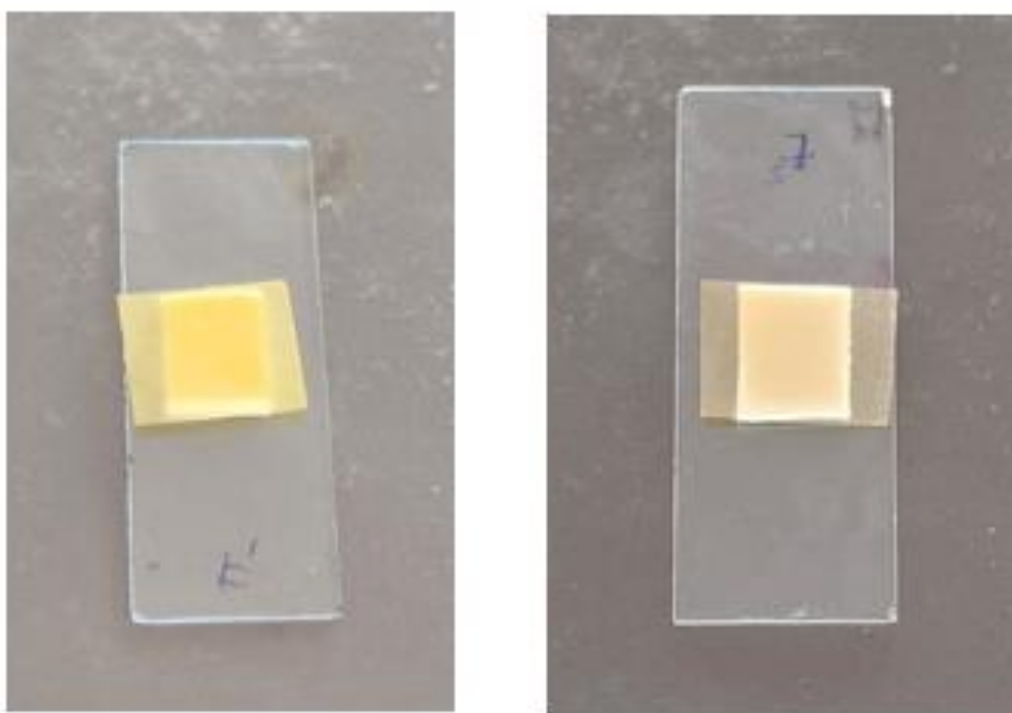


Figure16: Films Attached to Slides to Maintain Sink Condition.



Figure 17: Dissolution Apparatus.

Phytochemical Tests:

Table 1: Phytochemical Tests.

Test	Results
Alkaloids	
Mayer's test	+
Wagner's test	+
Hagner's test	+
Dragondroff's test	+
Test for carbohydrates Molisch's test	+
Test for reducing sugars Fehling's test	+
Test for Tannins	+
Lead acetate test	+
Ferric chloride test	



Figure 18: Phytochemical tests 1



Figure 19: Phytochemical tests 2

Formulation of Orodispersible films:

Table 2: Formulations for Orodispersible films.

Ingredients	F1%	F2%	F3%	F4%	F5%
<i>Momordica charantia</i>	-	-	0.25	0.2	-
<i>Annona squamosa</i>	0.25	0.15	0.15	-	0.25
<i>Morinda citrifolia</i>	0.25	0.15	-	0.15	-
<i>Boerhaavia diffusa</i>	-	0.30	0.10	0.15	0.25
Gelatine	0.3	-	-	-	-
HPMC	0.7	0.8	0.6	0.7	0.75
Ascorbic acid	0.2	0.2	0.2	0.2	0.2
Water	10	10	10	10	10

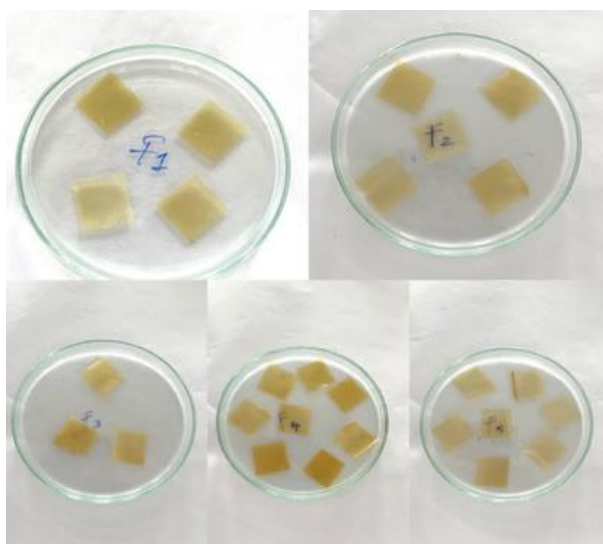


Figure 20: Formulations of orodispersible films.

RESULTS:**Evaluation tests for Orodispersible films:****Table No: 3. Evaluation tests for Orodispersible films.**

Tests	F1	F2	F3	F4	F5
Colour	Pale green	Pale yellow	Pale yellow	Pale yellow	Pale yellow
Odour	Mild	Mild	Mild	Mild	Mild
Texture	Soft	Soft	Soft	Soft	Soft
Folding Endurance	≥ 300	≥ 300	≥ 300	≥ 300	≥ 300
Weight Variation	0.19 $\pm$ 0.1	0.28 $\pm$ 0.2	0.21 $\pm$ 0.1	0.28 $\pm$ 0.2	0.23 $\pm$ 0.05
pH	6.0	6.0	5.4	5.6	6.0
Swelling Index	33.5 $\pm$ 1.5	35.9 $\pm$ 2.1	32.7 $\pm$ 2.6	34.5 $\pm$ 1.7	31.8 $\pm$ 2.5
Disintegration test (sec)	36	33	35	32	34

%.Drug release test:**Table No: 4. %. Drug release test of all the formulations.**

Time(mins)	F1 (%)	F2 (%)	F3 (%)	F4 (%)	F5 (%)
0 mins	0%	0	0	0	0
5 mins	9.12	6.60	10.92	8.76	7.32
10 mins	17.34	15.96	17.76	15.96	17.13
15 mins	32.70	27.30	23.70	25.50	29.64
20 mins	43.50	41.70	43.50	34.50	35.76
25 mins	56.10	57.90	65.10	45.30	48.36
30 mins	75.90	70.50	74.10	65.10	70.32
35 mins	83.10	84.90	83.10	86.30	84.36
40 mins	84.10	92.10	94.80	92.82	86.34
45 mins	85.44	92.82	95.52	93.72	88.32

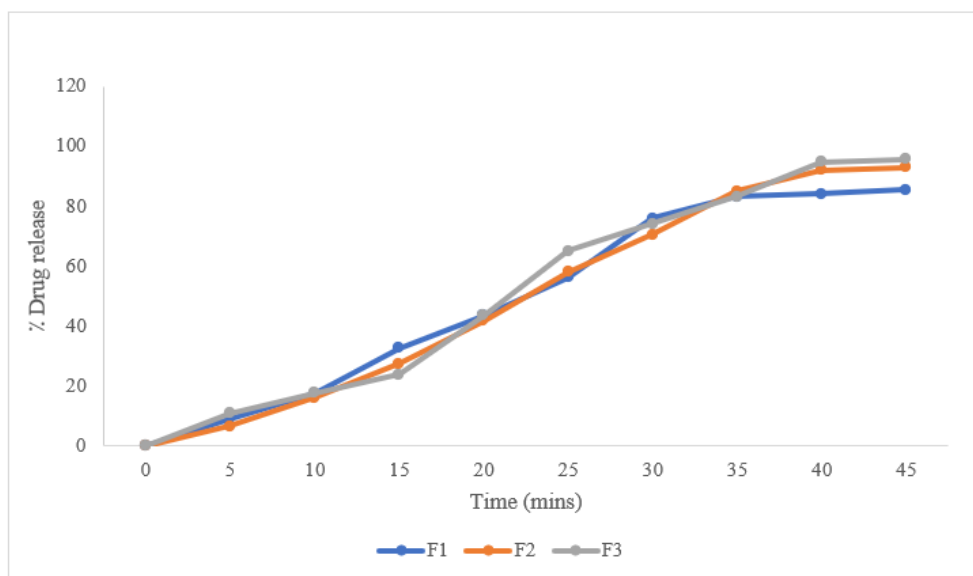


Fig No: 21. % Drug release of F1, F2, F3.

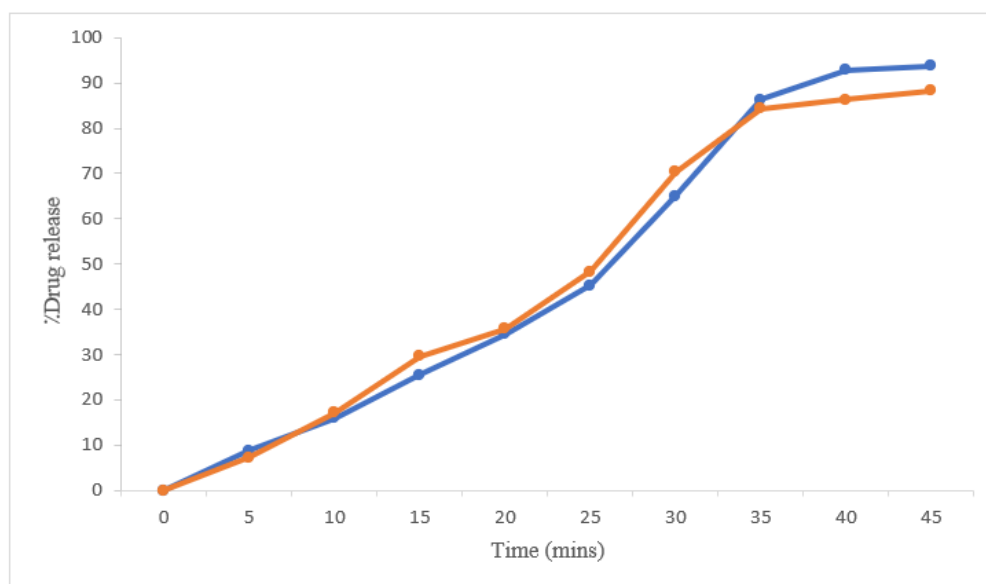


Fig No: 22. % Drug release of F4, F5.

DISCUSSION:

F1: The colour is pale green with the mild odour and the texture is soft. The folding endurance of the film is ≥ 300 and the weight variation of the film is 0.19 ± 0.1 . The pH of the film is 6.0 and swelling index of the film is 33.5 ± 1.5 . The disintegration time of the film is in 36 sec and % drug release was found to be 85.44 at 45mins.

F2: The colour is pale yellow with the mild odour and the texture is soft. The folding endurance of the film is ≥ 300 and the weight variation of the film is 0.28 ± 0.2 . The pH of the film is 6.0 and swelling index of the film is 33.5 ± 1.5 . The disintegration time of the film is in 33 sec and the % drug release was found to be 92.82 at 45mins.

F3: The colour is pale yellow with the mild odour and the texture is soft. The folding endurance of the film is ≥ 300 and the weight variation of the

film is 0.21 ± 0.1 . The pH of the film is 5.4 and swelling index of the film is 32.7 ± 2.6 . The disintegration time of the film is in 35 sec and the % drug release was found to be 95.52 at 45mins.

F4: The colour is pale yellow with the mild odour and the texture is soft. The folding endurance of the film is ≥ 300 and the weight variation of the film is 0.28 ± 0.2 . The pH of the film is 5.6 and swelling index of the film is 34.5 ± 1.7 . The disintegration time of the film is in 32 sec and the % drug release was found to be 93.72 at 45mins.

F5: The colour is pale yellow with the mild odour and the texture is soft. The folding endurance of the film is ≥ 300 and the weight variation of the film is 0.23 ± 0.03 . The pH of the film is 6.0 and swelling index of the film is 31.8 ± 2.5 . The disintegration times of the film is in 34 sec and the % drug release was found to be 88.32 at 45mins. From the above discussion, all the five formulations are formulated and evaluated for physicochemical parameters are found to be pale yellow in colour with the mild odour, soft texture. The F1, F2, F5 are shown the pH of 6.0 which is near to the pH of saliva. The maximum % drug release of the formulated film shown are F3- 95.52%, F4- 93.72, F2- 92.82, F5- 88.32, F1- 85.44.

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