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ANALYSIS ON ISOLATE GELATINOUS PROTEIN POLYMER MATERIAL FROM FENUGREEK SEED

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ABSTRACT

fenugreek seeds, a traditional plant with many uses in fields like medicine, cosmetics, and food nutrition, were used to isolate a gelatinous protein polymer material. Gelatinous protein polymer material will be employed for research in the therapeutic fields, such as antibacterial, antioxidant, antifungal, antidiabetic, anti-inflammatory, anti-cancer, immunomodulator, antitoxic, and antitarax. Fenugreek seeds are organic, safe, non-polluting, and eco-friendly. They are also readily and affordably available on the market. 25% of the nutritional content of fenugreek seeds is protein. This is why it was chosen for the extraction of the gelatinous protein polymer material. While some protein polymers are made using natural materials, the majority are created using chemicals. As a result, there are currently several natural and artificial polymers accessible for use as medicinal excipients. Many of the medications that are currently on the market have been either directly or indirectly produced from plants, which are a rich source of pharmaceuticals. Fenugreek seed gum (FSG) is a polysaccharide that is obtained from the seeds of Trigonella foenum guaiacum (Family Leguminosae). It contains galactomannans as its chemical ingredients. Polysaccharides typically serve important roles in pharmaceutical formulations as emulsifying, moisturizing, thickening, gelling, and suspending agents. This study's aim was to examine the film coating potential of FSG utilizing paracetamol as a model medication. The tablets were coated using the dip coating process with an aqueous coating solution made up of 2% FSG, 2% hydroxy propyl methyl cellulose (HPMC), and 1% sodium alginate. Lower friability, a longer disintegration time (14 min) compared to the core tablet's (3 min), improved hardness, and a better drug release profile were all displayed by the coated tablets. Drug release profiles for HPMC coated batches and FSG film coated batches were up to 8 hours and 12 hours, respectively. The FSG film's drug release rate values are quite similar to the HPMC release profile.

Keywords *- Gelatinous, Therapeutic, Natural, Protein polymer.*

INTRODUCTION:

The widespread use of plastic as a material for packaging has resulted in the production of numerous generations' worth of trash streams. The massive amount of waste made up primarily of plastic has contributed significantly to the existing environmental crisis. In an effort to protect the natural world, the majority of nations have begun to cut back on the amount of single-use plastics used in food packaging, which has resulted in a reduction in the cost of implementing pollution prevention measures. The photo-oxidation reaction is responsible for one of the most widely acknowledged occurrences that occurs when traditional plastic is used for packaging, and that phenomenon is the transfer of potentially poisonous and dangerous components from the packaging plastic matrix onto the food that is being wrapped. Methi is the common name for the seeds of the fenugreek plant, which belong to the Fabaceae family. Since ancient times, it has been put to use in the medical field to cure a variety of conditions, including wounds, abscesses, arthritis, bronchitis, and digestive

issues. According to Krystyna et al.'s research, fenugreek is an unusual spice crop whose qualities are being re-examined in light of the recent interest in alternative forms of treatment. In their capacity as particularly abundant providers of protein, lipids, fatty acids, and minerals. Due to the high nutritional content of this plant, it has a great number of potential applications in the manufacture of food, medicine, cosmetics, and pharmaceuticals in businesses that are favorable to the environment. There have been several reports of the medical benefits of fenugreek seeds, including their use as an anti-diabetic agent, an antioxidant, an anti-bacterial agent, an anti-cancer agent, an immunomodulator, an anti-cataract agent, and more. As a result of the significant potential value of fenugreek seeds, a great number of studies have published on occasion deterrent isolation approaches. The emergence of sustainable plastics that are biodegradable, eatable, safe, thermally resilient, and mechanically strong has been the focal point of the current trending global emphasis on the bio-economy as well as the growing awareness of the importance of health issues. Because the biological protection of the food product directly controls the product's shelf life, ensuring that it has adequate protection is highly important.

Because it acts as a barrier between the food and its surrounding environment, packaging plays a significant part in both the reduction of food waste and the accomplishment of predetermined food safety objectives. These edible plastics have the potential to be utilized in the intelligent and dynamic packaging materials for food products. The evolving requirements of the market for functional foods have been aided by recent improvements in the packaging of food products. The rapidly developing idea of intelligent and active packaging technology offers a wide variety of creative new ways to extend the shelf life of food goods, improve their quality, and ensure that they may be consumed safely. The reduction of waste and the growing demand for environmentally friendly or sustainable packaging that is made from plant extract and is either edible or biodegradable are both essential for maintaining a clean environment and good health throughout time. Edible films made from natural polymers are not only non-toxic and biodegradable, but they also have the added benefit of being easy to store and transport safely. When it comes to biomaterials made of protein, edible films made from a variety of sources have a significant impact on the expansion of environmentally friendly films due to the relative abundance of these films and their improved ability to make films. Protein-based films are the best option for hydrophilic surfaces because they create a barrier against oxygen and carbon dioxide. The structural characteristics of protein-embedded edible films confer a wide variety of capabilities, including intermolecular bonding. It is envisaged that films made from proteins will have a high oxygen barrier. Peanut protein, casein, whey protein, gelatin, soy protein, gluten protein, maize zein, and mung-bean protein are some examples of recently researched proteins that have been shown to have an edible film of biodegradable properties.

In recent years, fenugreek seeds have emerged as an attractive option for use as a source of protein within the context of concerns around food enrichment. Around 25 to 38% of fenugreek is comprised of seed protein, which is broken down into globulin (27.2%), prolamine (7.4%), glutelins (17.2%), and albumin protein (43.8%). The seeds of fenugreek, which are found among leguminous plants and can be consumed, are high in the percentage of protein and dietary fibers but low in the proportion of fat. The amount of protein that is found in fenugreek seeds has a quality that is comparable to that of soybean protein. In addition, the amount of lysine that can be found in these seeds is almost identical to that of soybean protein.

Additionally, the seeds of the fenugreek plant have a high concentration of bioactive substances such as polyphenols and saponins. Anti-diabetic qualities can be found in the polyphenolic chemicals found in

fenugreek seeds. They have potent antioxidant capabilities that help reduce excessive blood cholesterol, increases reduction in cell death and aging, and enhance the immune system. Fenugreek has a number of compounds that, when isolated, have the potential to engage in a variety of beneficial biological activities. These activities include providing protection against cancer, allergies, malaria, bacteria, and viruses. Because of this, there is a pressing need to increase the consumption and processing of resources with value addition.

Therefore, the exploitation of fenugreek protein in the preparation of edible film presents a sustainable way out for accelerating protein use, resulting in the creation of unique environmentally friendly and bio-based packaging. Due to the lack of published material on fenugreek protein concentrate that was utilized in the construction of edible film as a way of packaging food, research was conducted to study the possible applications of fenugreek protein-based edible film on food packaging. A leguminous plant known as fenugreek (Trigonella foenum-graecum), fenugreek is currently cultivated in Canada. It is native to the regions of northern Africa, the Mediterranean, western Asia, and northern India. Fenugreek seeds, which can be found in pods at the very end of the plant, have a long history of application in both the pharmaceutical and culinary fields. The study of extractable seed components, such as storage carbohydrates and saponins (Sauvaire et al., 1991, Taylor et al., 1997), has become increasingly popular during the past few years.

A galactomannan is a storage polysaccharide that can be found in the seed endosperm. This galactomannan is comparable to locust bean gum, guar gum, and tara gum, but it is more highly replaced. The capacity of galactomannans like guar and locust bean gum (LBG) to thicken and stabilize a wide variety of food items has led to their widespread application in the food industry (Stephen & Churns, 1995). Recent studies have suggested that in addition to these qualities, fenugreek gum may also have a surface active quality. It was discovered by Garti, Madar, Aserin, and Sternheim (1997) that pure fenugreek gum may be used to create stable emulsions with a remarkably small droplet size of 3 micrometers. Because the gum that was employed in that study still had 0.8% of its original protein content, it was impossible to attribute the surface activity to the hydrophilic gum on its own. In addition, Huang, Kakuda, and Cui (2002) observed that fenugreek gum demonstrated the highest stabilizing capabilities among 11 commercial gums and five laboratory generated gums in an oil/water emulsion model system. This was the case when comparing fenugreek gum to the other gums. Given the limited amount of study that has been done on fenugreek gum as a food gum, gaining a more comprehensive understanding of the physical properties and structure of fenugreek gum will help throw light on the structural origin of the gum's capabilities and will also facilitate the eventual application of fenugreek gum in the food business. Extracting a fenugreek gum with low levels of protein contamination was one of the primary goals of the current study, along with analyzing the gum's chemical structure and physical characteristics.

The most important function of the film coating unit in the manufacturing of pharmaceuticals. Film coatings are applied to dosage forms for a variety of purposes, including the production of visually appealing properties, the masking of unpleasant taste or odor, the facilitation of digestion, the improvement of stability, and the modification of the release characteristics of the drug. The procedure of film coating can be utilized for many different pharmaceutical products, including tablets, beads, pellets, granules, capsules, and drug crystals. Both aqueous polymeric dispersion, also known as latex, and polymeric solution, which can be based on an organic solvent or water, can be used to create a film layer. It is the primary component in the vast majority of film-coated formulations, and it can be derived from a variety of sources (natural, synthetic, or semi-synthetic), including cellulosic, acrylics, vinyl, and mix polymers. It has been utilized for a variety of

soluble pharmaceuticals. A natural polymer called fenugreek natural gum was extracted from the seed of the plant Trigonella foenum graecum, which belongs to the family Leguminosae. Natural polymers have advantages over synthetic and semi-synthetic polymers in a number of ways, including the fact that they are inexpensive and readily available, nonirritant, biodegradable, biocompatible, and environmentally benign, and that traditionally, fenugreek seeds have been employed in the treatment of diabetic illnesses. Canada, northern Africa, the Mediterranean, western Asia, and northern India are among the places in Asia and India where it is farmed. The most significant benefits of this formulation are a decreased frequency of administration, sustained plasma drug level, absence of a dose-dumping effect, and reduced severity of adverse effects. It has been documented in the scientific literature that fenugreek seed gum can be utilized in a variety of ways, including as an oral medication release retardant, binder, mucoadhesive, emulsifiers, as a gelling agent, and in the creation of nanoparticles.

OBJECTIVE

1. There has not been any research work on the film coating that has been published as of yet.
2. The sustained-release formulation is typically favored over the conventional dosage form in order to maintain a continuous therapeutic effect for a greater amount of time.

RESEARCH METHDOLOGY

The removal of fenugreek gum from its seeds The seeds of fenugreek were rinsed in water before being ground into a coarse powder with a grinder. In addition, the coarse powder was let to soak in distilled water for ten hours, and then muslin fabric was used to separate the gum from the bulk of the material. In order to finish the extraction procedure, the filtrate was subjected to many rounds of precipitation with ethanol. The gum was air dried at a temperature of 60 degrees Celsius, powdered, and then put into a polythene container for subsequent usage.

EVALUATION OF THE PHYSICOCHEMICAL PROPERTIES OF THE FENUGREEK GUM POWDER

Organoleptic evaluation : Organoleptic qualities such as color, odor, taste, fracture, and texture, all of these properties were analyzed and determined.Determination of purity of Gum Tests for alkaloids, carbohydrates, flavonoids, steroids, amino acids, terpins, saponins, oils, lipids, tannins, and phenols were carried out on gum in order to ascertain its level of purity and characterize its chemical composition..

Percentage yield

The seed from 10 grams of fenugreek was removed and separated. After being separated from the rest of the material, the gum was thoroughly dried, and the % yield was computed using the following formula.

$$\% \text{ Yield} = \text{Practical Yield} \times 100 / \text{Theoretical Yield}$$

Solubility

In order to assess the solubility, one part of dry gum powder was mixed with a variety of solvents and then agitated.

After adding 25 milliliters of water and a precisely measured quantity of fine powdered FSG gum to a measuring cylinder with a glass stopper of 25 milliliters capacity, the mixture was vigorously shaken continuously for one hour at a rate of once every ten minutes. After that, it was allowed to relax for three hours at room temperature. After then, the amount of space that the gum took up was measured. The identical process was carried out three times, and then the mean value was determined by applying the following formula to the results.

Swelling index = (W2 - W1) × 100/W2,

Where W1 is the initial weight of tablet and W2 is the weight of hydrated tablet.

Bulk density, **Tapped density** :

$$\textbf{Eq 1 Bulk density } (\rho) = \frac{\text{Weight of sample}}{\text{Tapped volume}}$$

$$\textbf{Eq 2 Tapped density } (\rho_b) = \frac{\text{Weight of sample}}{\text{Bulk volume}}$$

In order to estimate the density of FSG, 10 grams of FSG powder were carefully weighed into a measuring cylinder with a capacity of 100 milliliters, and then the volume of the powder was read off so that the bulk volume could be calculated. After then, a reading was taken of the volume of the powder after at least every 50 taps, and this continued until a steady volume of powder was acquired. This is an example of a tapped resource. proportion of powder to the total volume. Both the bulk density and the tapped density were determined through the use of equation 1 and equation 2 respectively.

Angle of repose: A measurement of the flow properties was made using the angle of repose. The frictional forces between the particles in the powder cause the powder to flow improperly. Angle of repose quantifies these frictional forces. It can be calculated by following formula:

Tan θ=h/r or θ = tan–1 h/r.

Where,

- h= height of pile;
- r= radius of the pile base
- θ = angle of repose.

On a burette stand at the height of two to three centimeters, a dry and clean funnel was fastened in place. After that, a piece of graph paper was selected, after which it was positioned on a surface that was both level and well-flattened, and a sufficient quantity of powder, ten grams, was selected. It was allowed to flow gently through the funnel until the mass was almost touching the tip of the funnel. The pencil was used to sketch the circle of the pile, and then the midpoint of the pile was located so that its radius could be determined. The experiment was repeated three times, after which the average height and radius were determined. This formula was utilized in the determination of the angle of repose.

Compressibility of powder, measured by Carr's consolidation index Powder compressibility was measured by utilizing five grams of gum powder, which was then deposited into a ten milliliter measuring cylinder with the assistance of a funnel before the measuring cylinder was placed on the bulk density device. The volume that the powder occupied when it was first measured (known as the V0 volume) was recorded. After that, the volume measuring cylinder was tapped repeatedly until a stable reading was achieved. After the tapping was finished, the final volume was recorded (the tapped volume, Vt), and the compressibility was determined by applying the following formula:

Consolidation Index = [(Tapped density – Fluff density)/Tapped density] × 100

Where Wf is the final weight sample and Wi initial weight of sample.

pH of gum Sample

After weighing and collecting 5 grams of the substance in triplicate in a separate beaker, it was combined with 20 milliliters of distilled water, and the resulting suspension was agitated for five minutes before the pH of the solution was determined using a calibrated digital pH meter.

Ash content %

A sample of gum weighing three grams was placed in a crucible made of silica that had been heated, measured, and weighted in advance. At the very bottom of the crucible was a layer of powder that was very finely and evenly distributed. The heating medium was gradually applied to the crucible as the temperature rose in order to bring it to a red hot state and remove the carbon. After the sample had been cleaned of all traces of carbon, the crucible was allowed to cool before its weight was recorded. The process was carried out a further two times in order to maintain the same weight. The air-dried medication was used as a reference point in order to calculate the percentage of total ash.

Viscosity

The viscosity of gum was measured by making different concentrations of gum suspension. These concentrations, which were initially 0.4%, 0.8%, and 1% w/v, were made at a temperature of 25 degrees Celsius. Utilizing a Brookfield Rheometer, we determined the viscosity of the prepared suspension on both the first day and the next day..

FTIR

Look into it. 100 milligrams of the gum powder were mixed with four hundred milligrams of potassium bromide, and then the mixture was compressed using a hydraulic press.

Moisture content (MC) %

An evaporated dish that included 10 grams of FSG was cooked in a hot air oven to 105 degrees Celsius until a consistent weight was achieved. It was possible to determine the average of the three readings. MC (percent) = Wf - Wi x 100.

tablet is formed by pressing under a pressure of 15 tons. In a spectrophotometer made by Perkin Elmer, the tablet was examined using a wavelength range that went from 4000 to 400 cm-1.

X-ray diffraction analysis (XRD)

Using an X-ray diffraction spectrometer (Bruker, AXS/8, Berlin, Germany), we were able to record a spectrum of X-ray diffraction. Tablets were formed by pressing the powdered dry gum. The X-ray diffraction spectra were obtained by recording them with Cu-ka radiation at 40 kV and 60 mA. The X-ray diffraction grams were run at a scanning speed of two degrees per minute and a chart speed of two degrees divided by two centimeters for every two.

Procedures for the preparation of the coating suspension

2% weight-per-volume of fenugreek gum and 1% sodium alginates were dispersed in distill water at 40-50 degrees Celsius with constant stirring using a magnetic stirrer. The mixture was allowed to mix for up to two hours. In a similar fashion, HPMC coating suspension was also made using 2% weight-per-volume (w/v) HPMC.

Viscosity of coating suspension

The Brookfield LVDV-IV+ digital rheometer was used at 100 RPM with the spindle in order to determine the viscosity of the optimum coated suspensions of gum.

PREPARATION CORE TABLET OF PROPRANOLOL HYDROCHLORIDE FOR FILM COATED TABLET FORMULATION

Core tablets of propranolol hydrochloride were made by employing a variety of polymers in varying proportions and then compressing the mixture directly, as detailed in the formulation table. After being thoroughly combined for ten minutes and having quantities of the medication and polymer combination accurately weighed, the mixture was sent through sieve no. 80. In order to achieve a total bulk weight of 400 mg for each tablet, satisfactory quantities of the diluents, specifically lactose, were utilized. The resulting powder mixture was then compressed to a hardness of 6-8 kg/cm2 using a single punch tablet press with 8 mm round punches..

Preparation of coated tablets :

Tablets of paracetamol that had been acquired were first coated by being immersed for five minutes in a prepared coating suspension of FSG. After that, the coated tablets were dispersed in a 5% solution of CaCl2 for the same amount of time. Tablets that had a film coating were dried in an oven with hot air. Along the same lines, HPMC-coated tablets were also manufactured.

EVALUATION OF CORE AND FILM COATED TABLETS

The same amount of weight throughout The evaluation of the tablets' weight uniformity was found by selecting 4 tablets at random and measuring their individual weights on an analytical balance. This led to the discovery that the tablets' weights were not consistent with one another.

Friability

The Roche Friabilator was used to make an assessment of the tablets' level of friability. This device rotates the tablets in a plastic hollow at a speed of 25 revolutions per minute while dropping them from a height of 6 inches in per revolution, which combines the effects of abrasion and shock on the tablets. The pre-weighed sample of tablets was then placed in the friabilator cavity and put through 100 revolutions over the course of four minutes. After that, the tablets were reweighed after being e-dusted with a muslin cloth and reweighing.

The friability (f) is given by the formula.

$$\% \text{ Friability} = \frac{(w1-w2)}{w1} \times 100$$

The weight of the tablets before the test is denoted by W1, and the weight of the tablets after the test is denoted by W2. The limit should not be more than one percent.

EVALUATION OF COATED TABLETS

The evaluation of the physicochemical attributes of coated tablets, such as weight uniformity, hardness, friability, and disintegration time, was carried out in the same manner as was carried out for uncoated tablets.

Dissolution studies

The in vitro release of FSG and HPMC film coated tablets was investigated using an eight station (USP) Type II dissolving apparatus at a temperature of 37 0.5 degrees Celsius with a speed of 50 revolutions per minute in 0.1 percent hydrogen chloride as the dissolution medium for a period of two hours. In order to maintain a constant media volume, a 5 mL sample was taken from the dissolution medium at defined time intervals and then replaced with an equivalent volume of fresh medium (also 5 mL). Following the filtration process, each sample was put through an analysis with a double beam UV visible spectrophotometer set to a maximum of 249 nm. After two hours, the dissolution media in each batch were switched out for phosphate buffer with a pH of 7.2. This investigation was carried out in triplicate for each batch.

DATA ANALYSIS.

Table 1: Phytochemical properties of FSG gum powder

Test	Results
Test for mucilage (Ruthenium red test)	+
Monosaccharide Test	-
Test for Tannins (Ferric chloride test)	-
Test for proteins (Ninhydrin test)	-

Test for alkaloids (Wagner's test)	-
Test for glycosides (Keller – Killaini test)	-
Test for Carbohydrates (Molisch's test)	+
Test for flavonoid (Shinoda test)	-
Test for reducing sugar (Felhing's test)	-

Table 2: Organoleptic properties of isolated FSG powder

Gums	**Colour**	**Odour**	**Taste**	**Fracture**
FSG	Yellow	Characteristics	Bitter	Irregular

Table 3: Solubility profile of isolated FSG powder

Solvents	**Results**
Cold water	Slightly soluble
Hot water	Viscous colloidal dispersion
Ethanol	Insoluble
Benzene	Insoluble
Acetone	Insoluble

Table 4: Some physicochemical properties of FSG powder

Parameters	**Results**
Percentage yield	25%
Solubility	Slightly soluble
Swelling Index	10.2ml
Bulk Density	0.667

Tapped Density	0.809
Angle of repose	27.85
Carr's index	17.55%
H.R	1.246
Moisture Content%	21.40
pH of mucilage	6.7
Ash content%	5.98
Melting point	248-256^{0}C

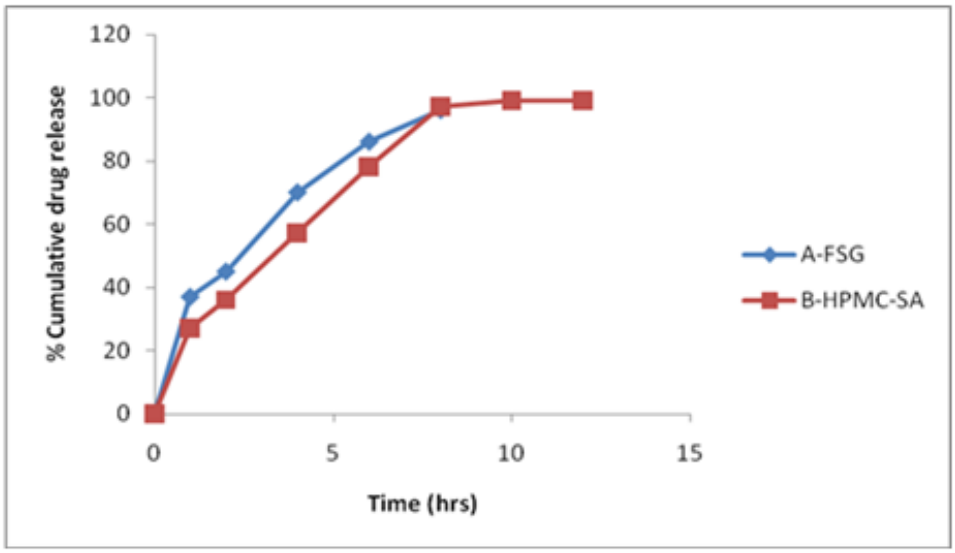

FIG .1 Dissolution profile of paracetamol coated tablets

RESULTS AND DISCUSSIONS :

In the process of isolating FSG gum by the use of hot water extraction and ethanol treatment, fenugreek seeds produced 25% by weight of gum. The gum that had been extracted was put through a series of tests for identification using ruthenium red and by dissolving it in hot distilled water. After being exposed to ruthenium red, the particles became pink and a gelatinous mass was produced. The results of the remaining tests all pointed to the gum being of a polysaccharide composition. The presence of carbohydrates was revealed in the findings of tests conducted on the purity of FSG. Other phytoconstituents were not present in the powder that was separated. This demonstrates the isolated gum's high level of purity. Table 1 displayed the results of the study. The organoleptic qualities test revealed further characteristics of the separated gum, such as its color,

odor, taste, fracture, and texture. The color of FSG was discovered to be yellowish, and its flavor was determined to be characteristically bitter. The crack was jagged, and the gum's texture was all over the place when it was isolated. Table 2 presented the findings of the study. To find out how soluble FSG is, warm water was used in the experiment. Some organic solvents include ethanol, benzene, butanol, chloroform, and ether.

With warm water, the FSG could form viscous colloidal dispersions, but it was insoluble in organic solvents such ethanol, benzene, butanol, chloroform, and ether. Table 3 displayed the results of the analysis. In table 4, the results of evaluating the physicochemical properties of fenugreek gum are shown. All of these values were found to fall within the acceptable range, as shown by the reference values for natural gum. Isolated FSG has ash values of 5.98, pH 6.7, moisture content 21.40%, melting point 248-2560C, bulk density 0.667, tapped density 0.809, vehicles index 17.55, H.R 1.246, and was angle of repose 27.85. The pH values of a 2% solution of the FSG were found to be slightly acidic or close to neutral, which showed that the FSG is non-irritating to the mucous membrane of the buccal cavity and the gastrointestinal tract, and that it can be employed for the creation of buccal and oral drug delivery systems. The FSG was discovered to have a swelling index of 10.2 milliliters, which is an indicator of good water absorption; as a result, it produces a hydrated three-dimensional network from which medicine can be released effectively through diffusion.

The hygroscopic nature of any substance can be determined by its capacity to take up moisture on its own. If the excipient is hygroscopic, it has the potential to change many of the features of the dosage forms. As a result, it is essential to ascertain the hygroscopic nature of the excipient as well as the amount of moisture that is capable of being absorbed by the excipient. According to the findings of this study, the FSG are hygroscopic, which means they have to be kept in airtight containers while they are not in use. The flow through the FSP ranged from unpassable to acceptable. Therefore, in order to increase the flow, it is necessary to add glidants. On the first day, the viscosity of the isolated FSG was found to be 33cp, 34cp, and 43cp when the concentration was 0.4, 0.8, and 1% respectively. On the second day, the viscosity was found to be 33cp, 35cp, and 45cp when the concentration was 0.4, 0.8, and 1% respectively. It seemed to indicate that when the concentration grew, the medication released rate would decrease due to the swelling or gelling property of the FSG.

CONCLUSION

With paracetamol pills serving as a model, researchers looked at the feasibility of using fenugreek gum either as a film-coating agent or polymer. A variety of criteria, including uniformity of weight, friability, disintegration time, and dissolving profiles, were examined in relation to the tablets. It is possible to make use of fenugreek seed gum for the creation of visual features of dosage forms, hiding an unpleasant taste or odor, making digestion easier, boosting stability, and changing the drug release properties of the drug.

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