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# ISOLATION AND EVALUATION OF STARCH FROM *MUSA PARADISIACA* LINN. AS A BINDER IN TABLET

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**Keywords:**

*Musa paradisiaca L.*, Starch, Binding agent, Characterization, Tablet

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**ABSTRACT:** A hydrophilic polysaccharide become isolated from green fruit of *Musa paradisiaca* L. by the usage of wet milling procedure and distilled water extraction the use of sodium sulphite solution, accompanied by way of maceration, centrifugation and sedimentation. The isolated polysaccharides became characterised for physicochemical properties, viscosity and flow behaviour, scanning electron microscopy (SEM), differential scanning calorimetry (DSC) and XRD study. Diverse concentrations of isolated polysaccharide had been studied for binding property towards Maize starch in traditional tablet formulation. In vivo toxicity study detected secure nature of the isolated polysaccharide. Standard, the outcomes of the present study indicated that isolated polysaccharide is resembles to starch and not useful for frozen products. Polysaccharide is shows Pseudoplastic nature with thixotropic behaviour and smooth surface and elongated oval shape granules seen on SEM Micrograph. Form experiment the optimized concentration banana starch as a binder is 5 to 6 %w/w for solid dosage forms. The outcomes of the study suggested that isolated polysaccharides is promising binding agent as a pharmaceutical excipient in drug delivery system.

**INTRODUCTION:** Binder is agent employed to impart cohesiveness to granules and remain the tablet intact after compression. Natural binders are used in pharmaceutical and food industry due to their low toxicity, low cost, biodegradable, and availability. Different type of natural polymer are used as binding agent like starch, gum etc. Starches mainly maize starch, rice starch, corn starch, potato starch wheat starch are widely used as binding and disintegrating agent.[1]

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*Musa paradisiaca L.* is belonging to family *Musaceae*. Banana is grown extensively in tropical and subtropical regions like India, Africa, Indo-malesian, and Islands of pacific. Although the composition of banana fruit has been defined, comparatively little work has been performed on the starch. Green banana is rich in starch as a main component that alters on ripening.

Commercially, banana may be another alternative source for starch and as new appliances in Pharmaceutical, Food and colour industry. Generally starch is deposited in the fruit in within the shape of granules, partially crystalline, whose morphology, chemical composition, and super molecular structure are characteristic of each particular plant species. Amylose and amylopectin are the main functional components of starch [2].

Green banana encompasses a large amount of starch throughout its unripe stage, which consists of around 20-25 % in the pulp of the fruits. Banana starch can be applied as substitute for potato, corn and wheat starch [3].

Banana fruit is either consumed in fully grown (due to its high sugar content) or unripe (in a number of native dishes requiring high starch content). Starch is conceivably the most important polymeric carbohydrate in phrase of its functionality that impart to products in diverse industries. Currently, in industries the new product development area is interesting in searching for starch with superior functional products such as viscosity, solubility, low retrogradation and syneresis tendency, etc. Since several long ago, the propensity is looking for alternative sources to obtain starch exhibiting better physicochemical and functional characteristics [4].

In the present study an effort was made to evaluate the efficacy of hydrophilic polysaccharide isolated from *Musa paradisiaca* fruit (obtained from *Musa paradisiaca* L., family *Musaceae*) as a tablet binder. It was investigated for isolation and characterization of starch, physicochemical properties, viscosity and flow characteristics, surface chemistry, thermal properties, and its suitability as Tablet binder in pharmaceutical field. The potential binding capability of hydrophilic polysaccharide was evaluated with standard starch paste as a tablet binder.

## MATERIALS AND METHODS:

**Material:** Green banana fruit was available from local market Nashik, Maharashtra. PVP as a disintegrant, magnesium stearate as lubricant, talc as a glidant, maize starch used as binder and Lactose as a diluent are obtained from Research lab Finechem industries, Mumbai, Maharashtra. Paracetamol dense powder Modern Industries, Nashik, Maharashtra was used as a model drug at a concentration of 250 mg per tablet. All other reagents and chemicals used were of analytical and pharmaceutical grade.

**Isolation of Starch:** Unripe banana of the widespread variety *Musa paradisiaca* was collected and authenticated from botanical survey of India, WRC, Pune. Banana starch was isolated from fruit by modification method of Kim *et al.*, [5]. The fruits were peeled and cut into 5-6 cm cubes (500 g total weight) and immediately rinsed in sodium sulfite solution (1.22 g/L) and then macerated at low speed in a grinder for 2 min. The homogenate was consecutively shifted through sieve No. 60 # & 100 # respectively and washed it with distilled water until the clear filtrate came out. The collected filtrate was then centrifuged at 7000 rpm for 20 min and upper off-white sediment scrapped out. The centrifuged mass was dispersed with distilled water and again centrifuged for 20 min at 7000 rpm, followed by the repetition of the cycle for 2 to 3 times to obtain clear white starch. The white starch sediments were dried in a convection oven at 40 ± 5 °C for 24 h. The dried mass was ground with a mortar and pestle to pass a No. 100 # sieve and stored at room temperature in a sealed container.

**Physicochemical Characterization of Banana starch:**

**Determination of Chemical composition:** The moisture content of banana (*Musa paradisiaca*) starch was calculated from the weight loss upon 2 hr. heating at 130 ± 5°C in an oven [6]. Protein content was determined by obtaining the total nitrogen content by Kjeldahl method [7]. The factor N x 5.7 was used for the estimation of the total protein content of the sample.

**Total Ash:** The dried banana starch (2g) was weighed in a tare silica crucible and incinerated at a temperature 400 ºC until free from carbon, cool and weighed. The percentage of ash was calculated on the dried polymer basis [8].

**Aqueous solubility:** The aqueous solubility of starch was determined gravimetrically by accurately weighing starch (1g) into 100 ml of distilled water and hydrated at room temperature for 24 hr. thereupon; the dispersion was filtered through a pre-weighed filter paper of medium porosity. The residue on the filter paper was weighed after drying in oven at 50 ºC for 24 hr. Solubility of starch was determined by taking difference between weights [9].

**Viscosity and flow behavior:** The flow behavior of starch paste was determined by measuring viscosity. Viscosity of 5% starch paste was

measured by Brookfield viscometer LV (DV-E viscometer) using spindle number S-62. The shear rate was increased from 0.3, 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, to 6 rpm and maintain the temperature at 25 ºC [10].

**Freeze-Thaw Stability:** 5 mL of 5 % banana starch paste was subjected to a one cycle freeze-thaw process with 20$^{o}$C for 18 hr storage in a freezer, followed by 6 hr storage at room temperature at 27 ºC. These samples were then centrifuged at 3000 rpm for 10 min. The percentage of water separated after the freeze-thaw cycle was measured [11].

**Scanning electron microscopy:** For SEM studies, the samples were fixed to a conductive double glued tape of copper; which was covered with a 20 nm of thick coal layer. It was deposited under a vacuum using an evaporator in a JEOL JSM-6390 (Japan) electron microscope. Later on, samples were covered in the ionizer metals JEOL with a 50 nm thickness gold layer. A film piece was mounted on aluminum stubs using a double- sided tape and then coated with a gold layer (40–50 nm), allowing surface and cross-section visualization. All samples were examined using an accelerating voltage of 20 kV at 500X and 1000X magnification.

**FT-IR Spectroscopy:** IR spectrum of pure banana starch was recorded using Fourier Transform Infra-Red spectrophotometer (Shimadzu IR Affinity 1). The samples were previously triturated and mixed thoroughly with potassium bromide in 1:99 (sample: potassium bromide) ratio. Then, sample was scanned by DRS method. The scans were obtained at a resolution from 4000 to 400 cm$^{-1}$

**X-ray diffraction:** X-ray diffraction pattern were obtained by A Shimadzu model XRD 7000 X-ray diffractometer, adjusted to a scanning speed of 1°/min with copper radiation (k = 1.5406A) at 40 kV and 30A, was used to determine the crystallinity of starch and its composites with banana fibers.

**Differential Scanning Calorimetry:** Starch gelatinization was determined with DSC (Lab: METTLER) instrument using STAR$^{e}$ SW 8.10 software. DSC equipment was calibrated with indium. 0.85 mg of starch was weighed into aluminium pan & 70 % moisture content adjusted by adding de-ionised water. The pan was hermetically sealed & left to equilibrate for 1hr. at room temperature. An empty sealed in the same way was used as reference. Sample was scanned from temperature 30 $^{o}$C to 300 $^{o}$C at rate of 10$^{o}$C/min under a nitrogen flow of 10mL /min. Gelatinization temperature was determined by automatically computing initial temperature (Ti), maximum peak temperature (Tp), final temperature (Tf) & gelatinization enthalpy (ΔH) from the resulting Thermogram.

**Acute oral Toxicity study of banana starch:** Acute oral toxicity of isolated banana starch on mice was carried out in compliance with the protocol of Institutional animal ethical committee as per animal ethical committee of Bhujbal Knowledge City, MET's institute of pharmacy, Adgaon, Nasik, Maharashtra, India Registration number: 1344/ac/10/2012-2013/CPCSEA under CPCSEA, India). Acute toxicity studies had performed according to organization for economic cooperation and development (OECD) guidelines. Mice weighing between 20-25 gm in groups of five was used (n=5). The animals were fasted for 4 h with free access to water only. The isolated mucilage was administered orally in doses of 2000 mg/Kg in 0.5 ml of distilled water to different groups of mice and was observed over 14 days for mortality and physical/behavioural changes.

**Formulation of conventional Tablet [12-15]:** Various Tablet formulations were prepared by conventional wet granulation technique using different concentrations of Banana starch given in **Table 1** and Maize starch in **Table 2**. Briefly, slug of powder was prepared by mixing of model drug (paracetamol), PVP and lactose in mortar, starch paste was added as required. Granules were obtained by passing the dump mass through No. 16 # and dry in hot air oven at 45 °C for 30min and again granules were sieved through No. 22 #. Magnesium stearate [2% w/w] and Talc was used as lubricant and glidant. The Tablets were compressed on a single punch [Royal Artist, Mumbai, India] Tablet compression machine for each batch.

**TABLE 1: FORMULATION OF TABLET BY USING BANANA STARCH AS A BINDER**

| Ingredient (Batch) | B1 | B2 | B3 | B4 | B5 | B6 |
|---|---|---|---|---|---|---|
| Model drug [mg] | 250 | 250 | 250 | 250 | 250 | 250 |
| Banana starch[%] | 2 | 3 | 4 | 5 | 6 | 7 |
| PVP [mg] | 12 | 12 | 12 | 12 | 12 | 12 |
| Lactose [mg] | 26 | 26 | 26 | 26 | 26 | 26 |
| Mg. Stearate [mg] | 6 | 6 | 6 | 6 | 6 | 6 |
| Talc [mg] | 6 | 6 | 6 | 6 | 6 | 6 |
| Total | 300 | 300 | 300 | 300 | 300 | 300 |

**TABLE 2: FORMULATION OF CONVENTIONAL TABLET BY USING MAIZE STARCH AS A BINDER**

| Ingredient (Batch) | M1 | M2 | M3 | M4 | M5 | M6 |
|---|---|---|---|---|---|---|
| Model drug [mg] | 250 | 250 | 250 | 250 | 250 | 250 |
| Maize starch [%] | 2 | 3 | 4 | 5 | 6 | 7 |
| PVP [mg] | 12 | 12 | 12 | 12 | 12 | 12 |
| Lactose [mg] | 26 | 26 | 26 | 26 | 26 | 26 |
| Mg. stearate [mg] | 6 | 6 | 6 | 6 | 6 | 6 |
| Talc [mg] | 6 | 6 | 6 | 6 | 6 | 6 |
| Total | 300 | 300 | 300 | 300 | 300 | 300 |

Tablets were evaluated for the hardness, friability, thickness, and weight variation and disintegration time.

**Evaluation of Tablet:**

**Hardness:** To withstand mechanical shocks of handling in manufacture, packaging and shipping, Tablets require some amount of strength, or hardness and resistance to friability. For each formulation, the hardness of 6 Tablets was determined using the Monsanto hardness tester. The Tablet was held along its oblong axis in between the two jaws of the tester and a zero reading was taken. Then constant force was applied by rotating the knob until the Tablet fractured. The value at this point was noted [14, 16].

**Friability:** Friability test was used to determine Tablet strength. Roche friability tester was used for testing friability. The present test subjected a number of Tablets to the combined effect of shock abrasion by utilizing a plastic chamber which revolves at a speed of 25 rpm, dropping the Tablets to a distance of 6 inches in each revolution. A sample of preweighed 6 Tablets was placed in Roche friability tester which was then operated for 100 revolutions *i.e.* 4 minutes. The Tablets were then dusted and reweighed. Tablets lose less than 1.0% of their weight is generally considered as acceptable. Percent friability [% F] was calculated as follows [17].

$$\% F = \frac{\text{Initial weight - Final weight}}{\text{Initial weight}} \times 100 \quad \text{..... [1]}$$

**Weight variation test:** Individually 20 Tablets were weighed and average weight was calculated. Not more than two of the individual weights deviated from average weight by more than the percentage shown in the **Table 3** and none deviates by more than twice that percentage [8].

**TABLE 3: SPECIFICATIONS FOR TABLETS AS PER INDIAN PHARMACOPOEIA**

| Dosage Form | Average Weight of Tablet | % Deviation |
|---|---|---|
| Uncoated and film-coated tablets | 80 mg or less | 10 |
| | More than 80 mg but less than 250 mg | 7.5 |
| | 250 mg or more | 5 |

**Disintegration time:** Disintegration time was performed by using USP Tablet disintegration testing apparatus (Meta lab) at 37 ± 0.5 °C as per USP guidelines [17].

**RESULT AND DISCUSSION:**

**Physicochemical properties of banana starch:** The yield of banana starch from green banana fruit on wet basis was found to be 10.70 %w/w. The physicochemical properties of banana starch are summerised in **Table 4**. The outcome proved that wet-milling process is a good procedure for banana starch isolation with low levels of other components from the fruits. The protein content was found to be 1.09 % which is less. Luis A. Bello reported the protein content in "macho" and "criollo" banana starch are 2.03 % and 1.95 % respectively. The total ash content in banana starch is 3.75%w/w [2]. Freeze thaw stability study for banana starch predeicted that 30 % of water was drained from the incorporated in starch paste whereas higher freeze thaw stability of 50% was detected as compared to stability obtained by Luis A. Bello. The outcome predicted that banana starch is not useful for frozen products.

**TABLE 4: PHYSICOCHEMICAL PROPERTIES OF BANANA STARCH**

| Sr. no. | Parameter | Quantity |
|---|---|---|
| 1 | Moisture content [% w/w] | 0.688 ± 0.0085 |
| 2 | Protein [% w/w] | 1.09 ± 0.01527 |
| 3 | Total ash [%] | 3.75 ± 0.22 |
| 4 | Aqueous solubility[mg/ml] | 0.873 ± 0.0075 |
| 5 | Freeze-Thaw stability [ml] | 1.56 ± 0.057 |

**Viscosity and flow behavior:** The rheogram showed pseudoplastic flow of starch paste, which interpreted that viscosity of banana starch paste can't be expressed by any single value. Viscosity of banana starch paste decreases with increasing rate of shear **Fig. 1**.

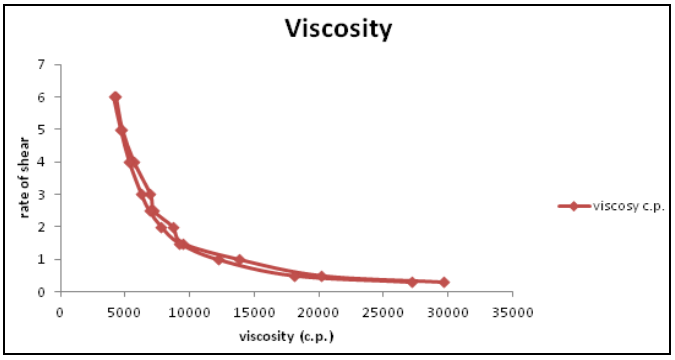


**FIG. 1: VISCOSITY OF BANANA STARCH PASTE**

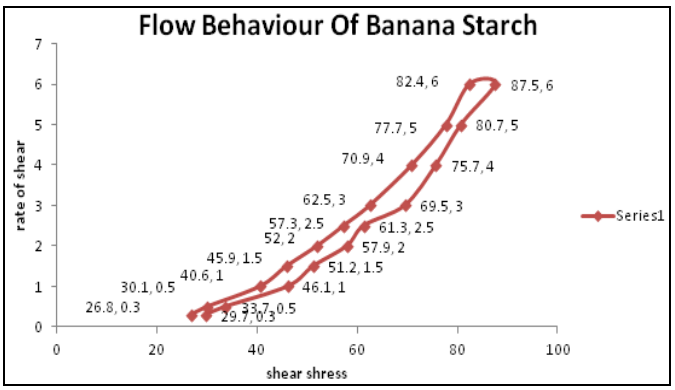


**FIG. 2: RHEOLOGICAL BEHAVIOUR OF BANANA STARCH**

The flow behavior of banana starch paste was determined by plotting rheogram shear stress against rate of shear shown in **Fig. 2**. Rheogram showed Non-Newtonian (shear thinning characteristic) flow of starch paste. The shear stress increases with escalation of shear rate. The down curve is frequently displaced to the left of up curve. This specified that break down of structure does not reform immediately when stress is reduced. Hence, starch paste showed thixotropic behaviour.

**Morphological characteristics:** SEM image of banana starch is shown in Fig. 3. The banana starch is smooth surface texture and size of the banana starch granule is in range of 25.95 µm to 27.09 µm × 9.19 µm to 15.28 µm with a higher heterogeneity showing round, oval and polygonal shapes. Sizes and shapes of starch granules have impact in a number of physicochemical, functional and nutritional characteristics as larger granules were developed with high paste viscosity and small granules had higher digestibility [11]. These differences of banana starch granules could affect to the other properties such as swelling power, viscosity or gelatinization temperature [3].

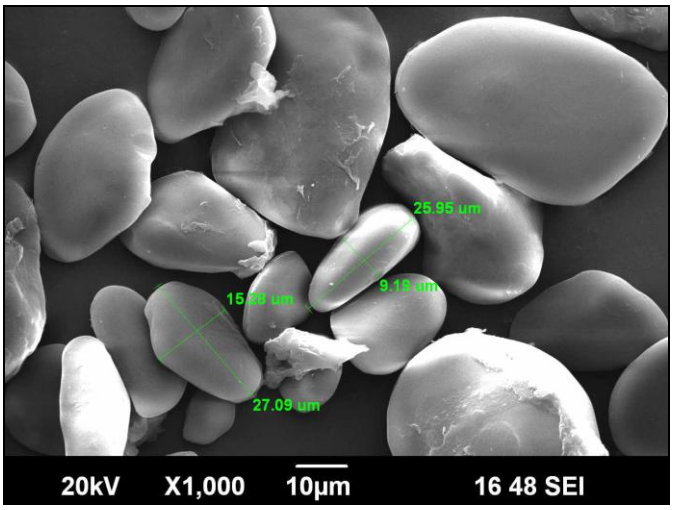


**FIG (a): MAGNIFICATION 1000X WITH 10 µM SCALE**

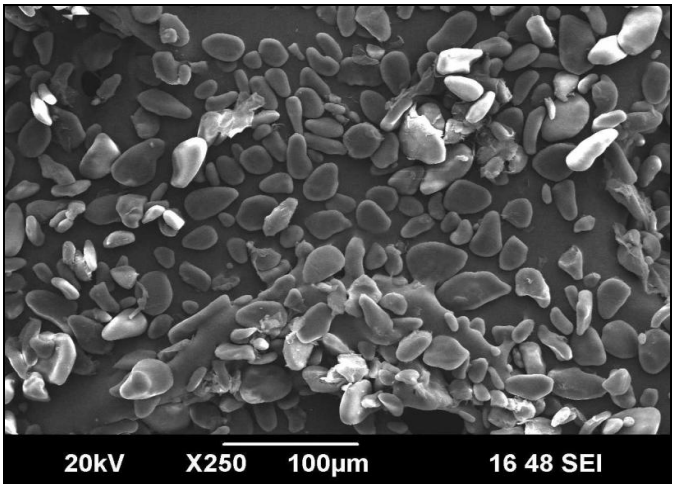


**FIG (B): MAGNIFICATION 250X WITH 100 µM SCALE**

**FIG. 3: SEM MICROGRAPH OF BANANA STARCH GRANULE**

**FT-IR Spectroscopy:** The FT-IR spectroscopic spectrum of banana starch is shown in **Fig. 4.** The broad band occurring at 3576-3103 $cm^{-1}$ was detected due to presence of hydroxyl [-OH] groups. The peak was obtained at 2982 $cm^{-1}$ with stretching modes of the C-H bonds of methylene groups [-CH2]. Absorption bands around 1254-1193 $cm^{-1}$ indicated the presence of C-O stretching of ether group present in starch.

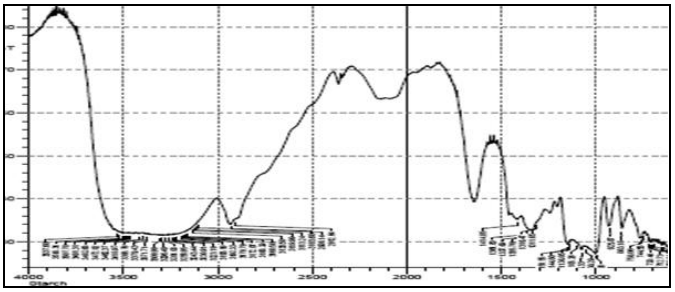

**FIG. 4: IR SPECTRUM OF BANANA STARCH**

**X-ray diffraction [powder]:** X-ray diffraction pattern of banana starch is shown in **Fig. 5.** The main diffractions peaks, centred at 6.21, 10.62, 11.74, 15.58, 17.68, 24.175 and 26.96º of 2θ (interplanar distances ‘‘d” of 14.21, 8.32, 7.52, 5.68, 5.012, 3.67and 3.303 Å, respectively).

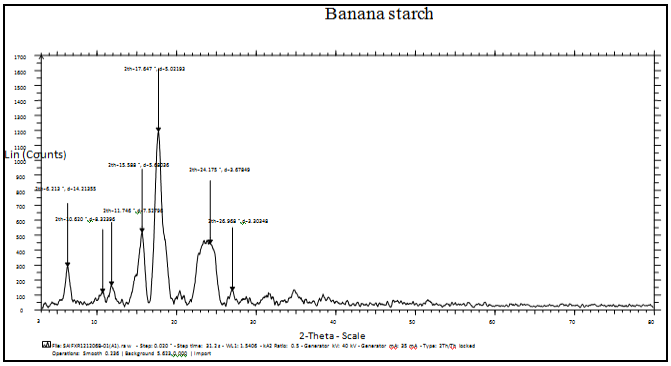


**FIG. 5: X-RAY DIFFRACTION PATTERN OF BANANA STARCH**

Amylose and amylopectin are two main components present in starch and detected. Pattern of crystallization was dictated by the ratio of amylose & amylopectin component during composite making. Hence, the two peaks at 17.68 and at 24.175º of 2θ (d = 5.012 and 3.67 Å) may be confirmed the occurrence of amylose and amylopectin.

**Differential Scanning Calorimetry:** The gelatinization temperature obtained from DSC curves was defined as the peak temperature (Tp); the amount of energy required for gelatinization to occur was considered the gelatinization enthalpy (DH). Differential Scanning Calorimetry [DSC] of green banana starch is presented in **Fig. 6**.

Gelatinization properties were defined by the three following temperatures: To (the onset temperature at which the gelatinization starts), Tp (the peak temperature which represents the endothermic peak on the DSC Thermogram) and Tc (the conclusion temperature at which the sample is fully gelatinized). As per DSC thermogram To, Tp and Tc were found to be 39.16 ºC, 71.46 ºC and 122.66 ºC These temperature were different than that of reported gelatinisation temperature ‘‘macho” variety banana (69.6 ºC, 74.5 ºC and 81.6 ºC) and ‘‘criollo” variety banana starch (71.4 ºC, 75.0 ºC and 80.4 ºC) (La´zaro *et al*., 2008). Tp is near to the reported value but much variation found in to and Tc. The deviation in To & Tc may be due to differ in variety of Banana.

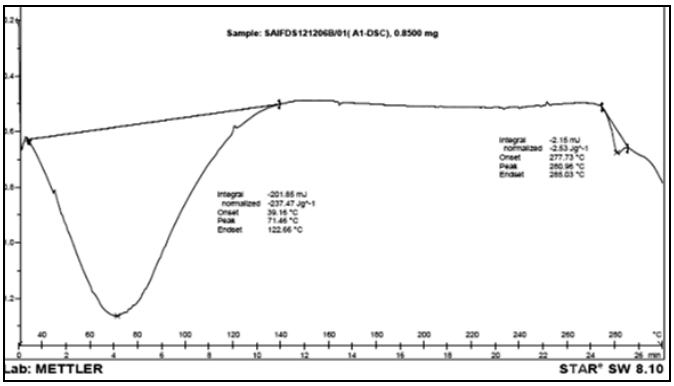


**FIG. 6: DSC CURVE OF BANANA STARCH**

**Acute oral toxicity study of Banana starch:** Animals were subjected for both in short term (24 h, with a dose of 2000 mg/kg body weight) and one month toxicity studies, which detected no death or abnormal behaviors in animal. Furthermore, this fruit has been traditionally used by the local people without reporting any toxic manifestations. Thus it can be claimed that the starch is safe for use and in particular, the amount used here is safe for further study.

**Binding properties of Banana starch:** Binding properties of banana starch was compared with maize starch. Banana starch and Maize starch was used in concentration as 2% w/v, 3% w/v, 4% w/v, 5% w/v, 6% w/v, & 7% w/v. The outcomes of the studies are mentioned in **Table 5** and **6**. Hardness and disintegration time of Tablets was increased as the concentration of banana starch and maize starch was increased. Friability of the Tablet was decreased as concentration of banana starch and maize starch was increased. Batches B4 & B5 showed the better result of hardness, friability and disintegration time of the Tablet. Hence, 5 – 6% banana starch paste can be applied for the further study.

**TABLE 5: EVALUATION PARAMETER OF CONVENTIONAL TABLET PREPARED BY BANANA STARCH BINDER**

| Parameter (Batches) | B1 | B2 | B3 | B4 | B5 | B6 |
|---|---|---|---|---|---|---|
| Hardness [Kg/cm$^3$] | 1.3±0.28 | 2.5±0.5 | 3.6±0.28 | 4.3±0.28 | 4.8±0.28 | 5.6±0.28 |
| Friability [%] | 7.15 | 5.21 | 1.64 | 0.93 | 0.65 | 0.27 |
| Weight variation | 302.8±1.3 | 302.6±1.7 | 302.8±1.3 | 303.3±1.7 | 303.6±1.49 | 303.8±1.34 |
| Thickness [mm] | 4.29±0.01 | 4.3±0.02 | 4.29±0.05 | 4.28±0.01 | 4.29±0.01 | 4.28±0.01 |
| Disintegration time [min.sec.] | 2.26±0.51 | 5.05±0.43 | 10.09±0.46 | 15.00±0.39 | 21.01±0.47 | 32.17±0.54 |

**TABLE 6: EVALUATION PARAMETER OF CONVENTIONAL TABLET PREPARED BY MAIZE STARCH BINDER**

| Parameter (Batches) | M1 | M2 | M3 | M4 | M5 | M6 |
|---|---|---|---|---|---|---|
| Hardness | 1.6±0.28 | 2.6±0.28 | 3.3±0.28 | 4.16±0.28 | 4.8±0.28 | 5.3±0.28 |
| Friability | 7.27 | 4.77 | 1.53 | 0.93 | 0.49 | 0.32 |
| Weight variation | 303.3±1.10 | 304.3±1.10 | 304±1.29 | 303.6±1.49 | 303.16±1.21 | 303.8±1.34 |
| Thickness | 4.29±0.01 | 4.29±0.01 | 4.29±0.01 | 4.3±0.01 | 4.29±0.01 | 4.3±0.01 |
| Disintegration time [min.sec.] | 2.1±0.17 | 5.36±0.57 | 10.24±0.11 | 15.39±0.38 | 21.05±0.43 | 31.30±0.86 |

**CONCLUSION:** Banana starch has been isolated from the unripe fruit of the plant *Musa paradisiaca* (Fam. *Musaceae*). The yield of banana starch obtained from green banana fruit on wet basis was 10.70% w/w. Aqueous dispersions of the banana starch was swelled to form highly viscous dispersions which exhibited pseudoplastic flow (shear-thinning) behaviour. As per the outcome of study, banana starch will be used as binder for solid dosage forms in a 5 to 6% concentration. The relative abundance and easy availability of banana starch may reduce cost where it is found growing abundantly wild or cultivated. Thus, banana starch can be further applied as a viscosity modifier, thickening agent in food industry as well as in the pharmaceutical industry. Materials with such properties have been used as stabilizers and suspending agents in foods, cosmetics and in liquid or solid dosage forms. As per the outcomes of freeze-thaw stability study it was concluded that banana starch is not suitable for frozen products.

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**CONFLICT OF INTEREST:** The authors report no conflict of interest.

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