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Kamlesh Kumar Maurya
Centre of Food Science and
Technology, Institute of
Agricultural Sciences, Banaras
Hindu University, Varanasi,
Uttar Pradesh, India

Anil Kumar Chauhan
Centre of Food Science and
Technology, Institute of
Agricultural Sciences, Banaras
Hindu University, Varanasi,
Uttar Pradesh, India

Abhishek Dutt Tripathi
Centre of Food Science and
Technology, Institute of
Agricultural Sciences, Banaras
Hindu University, Varanasi,
Uttar Pradesh, India

Arpit Srivastava
Centre of Food Science and
Technology, Institute of
Agricultural Sciences, Banaras
Hindu University, Varanasi,
Uttar Pradesh, India

Suresh Kumar Srivastava
School of Biochemical
Engineering, Indian Institute of
Technology (BHU), Varanasi,
Uttar Pradesh, India

Rajendra Singh
Pharma Stats, 9 Tudor Drive
Somerset, New Jersey 08873,
USA

Correspondence

Kamlesh Kumar Maurya
Centre of Food Science and
Technology, Institute of
Agricultural Sciences, Banaras
Hindu University, Varanasi,
Uttar Pradesh, India

Effect of protein and low molecular weight surfactants supplementation on physicochemical properties of spray dried guava (*Psidium guajava*) powder and its characterization

Kamlesh Kumar Maurya, Anil Kumar Chauhan, Abhishek Dutt Tripathi, Arpit Srivastava, Suresh Kumar Srivastava and Rajendra Singh

Abstract

The present study investigated the effect of proteins and low molecular weight surfactants (LMS) supplementation on guava powder characteristic produced by spray drying technology. Guava (*Psidium guajava*) fruit was selected as sugar rich food and sodium caseinate (NaCas) was selected as a model protein. Sodium stearoyl lactylate (SSL) and Polysorbate-80 (Tween-80) were chosen as model ionic and non-ionic low molecular weight surfactants. The required ratio of NaCas: Tween-80 and NaCas: SSL were 0.5 and 0.05%, respectively. Surface protein layered guava powder in sugar-protein systems was very sensitive in the presence of low molecular weight (M_w) surfactants as it lies below the critical micelle concentration of NaCas. These phenomena are explained on the basis of surface-glass transition temperature (T_g), nature of surfactants and glass transition temperature of sugars used. X-ray diffraction (XRD) and scanning electron microscopy (SEM) results revealed that the powders of guava-NaCas, supplemented with 0.05% SSL and tween-80 showed amorphous behavior. The spray dried powders produced in this study were extremely stable at room temperature and could be easily reconstituted in aqueous condition.

Keywords: sugar-rich foods (guava fruits), sodium caseinate, low molecular weight surfactants

Introduction

In ancient time people preserved their food through drying method for long time. Various method used in drying in different food material such as solid, paste, slurries or solution. Food powder are instantly present in routine life and this powder generally obtained in agricultural raw material by different processing method such as freeze drying, sun drying, vacuum drying spry drying etc. Spray-drying is a well-established and widely used method for transforming a wide range of liquid food products into powder form. The process involves spraying finely atomized solution droplets into a chamber where hot and dry air rapidly evaporates the solvent and converts the droplets into dry particulates. Spray-dried powders can be stored at ambient temperature for prolonged periods without compromising the powder stability. They are also cheaper to transport and easier to handle in manufacturing plants. Spray-dried powders are economical to produce compared to other processes such as freeze-drying [1]. Spray-drying has many applications, particularly in the food, pharmaceutical and agro chemical industry. Guava (*Psidium guajava* L.) is a highly nutritious fruit and very popular as poor man's apple. It has assumed special significance because of its hardy nature and prolific bearing even on marginal soils where other fruit crops fail badly. Though guava is native to tropical America, after its introduction to India in 17th Century by Portuguese it widely adapted to Indian conditions. The fruit has a considerable nutritional importance since it is rich in vitamins C, A and riboflavin, as well as in proteins, fibers and mineral salts. The fruit is also rich in minerals like phosphorous (23-37 mg/100 g), calcium (14-30 mg/100 g), iron (0.6-1.4 mg/100g) as well as vitamins like niacin, pathogenic acid, thiamine, Omega-3 and 6 poly unsaturated fatty acids and large amount of dietary fiber [2]. During the last two decades its cultivation increased in India from 0.94 lakh hectares to 2.33 lakh hectares. Though there was no significant increase in its productivity, the production increased from 1.09 million tonnes in 1991 to 2.69 million tonnes in 2012 and 3.91 million tonnes in 2015-16 [3]. Nanda *et al* reported overall postharvest losses of 18.05 per cent in guava. About 13.92% of this loss was during the farm operations

and among that harvesting losses were 4.36% [4].

Spray dried powder can be classified into two broad groups, non-sticky and sticky respectively. The stickiness problem causes considerable economic loss and limits the application of drying techniques, such as spray-drying for food and as well as pharmaceutical material [5, 6, 7]. It was found that the preferential migration of proteins combined with their film-forming property upon drying, is responsible for overcoming the stickiness of sugar-protein solutions. Inlet air temperature negatively influenced the bulk density due to the increase of powders porosity. The lower the bulk density, the higher the solubility of powder [8]. It has also been deduced that significantly small quantity of proteins and low molecular surfactants (LMS) is required to convert sugar-rich foods into powder form, in comparison to commonly used additives such as maltodextrins. Previous finding suggests that 0.13% of NaCas and whey proteins are required to convert sucrose (a model sugar-rich food) into fruit powder while more than 40% of maltodextrin (DE 6) is required to achieve the same product yield [9]. However, addition of large amounts of these carriers alters the resultant powder quality and risks consumer disapproval. In the present study, LMS (0.05%) was used which is small amount i.e. 800 times less than maltodextrin, so it is more economically.

Materials and Methods

Materials

Sodium caseinate (NaCas) (Sigma-Aldrich, USA) with a protein content of 92.9% was used as a model protein. Sodium stearoyllactylate (SSL) and Polysorbate-80 (Tween-80) were used as model surfactants. SSL is an ionic surfactant which has a lower molecular weight (451.6g/mol) and a comparatively higher hydrophile lipophile balance (HLB) value, while Tween-80 is a non-ionic surfactant with comparatively larger molecular weight (1310g/mol) and a lower HLB value. Both the surfactants are water soluble and are suitable for formation of oil-in-water emulsions (Mc Clements, 2005) [10].

Methods

25 kg guavas fruit were crushed in the fruit mill (PSF121, India) and then transferred to the pulper (RI-05, India) for obtaining the pulp. The extraction was done for the 4-5 times to obtain maximum pulp quantity. Fruit pulp was homogenized in a previously sterilized mechanical blender (GT56, company name India). The pulp was stored in a refrigerator in a previously sterilized stainless steel container at 4 °C with lid for further use.

Sample preparation

Before being dehydrated, the pulp (quantity of juice should be mentioned) was diluted in distilled water until reaching a total soluble solid content of 12° Brix. Protein solutions were prepared by heating the solution at 45±0°C and gently agitating on a magnetic stirrer. Solutions of guava pulp-protein-SSL and guava pulp-protein-Tween-80 were prepared by adding 0.05% of each surfactant to the solutions. The solutions were heated to 45±5°C to ensure that all solids were completely dissolved. Solutions of 0.5%, (w/w) NaCas were also prepared to determine the critical micelle concentration of NaCas. All prepared solutions were well mixed with guava pulp with help of beater. Two different sample were prepared having combination of (1) NaCas (0.5) +SSL0.05% (guava pulp) and (2) NaCas(0.5)+Tween-80 (0.05% (guava pulp)

resulting in two type of guava powders.

Powder production

Fruit pulp powder was prepared by using spray dryer NERO FSD 4 with 25 kg/h water evaporation rate, designed to combine spray drying. The inlet and outlet temperatures were maintained at 180 and 80°C, respectively. The air flow rate was maintained at 36 m³/h, while the aspiration was 100%. The powders were collected from cyclone and the cylindrical part of the dryer chamber by slightly sweeping the chamber wall [11]. The yield was calculated as the ratio of the mass of solids collected after spray-drying to the amount in the feed solutions. 5.2 kg guava powder was obtained using of 25 kg guava fruit with 20.8% recovery of guava powder in spray drying (Fig. 1).

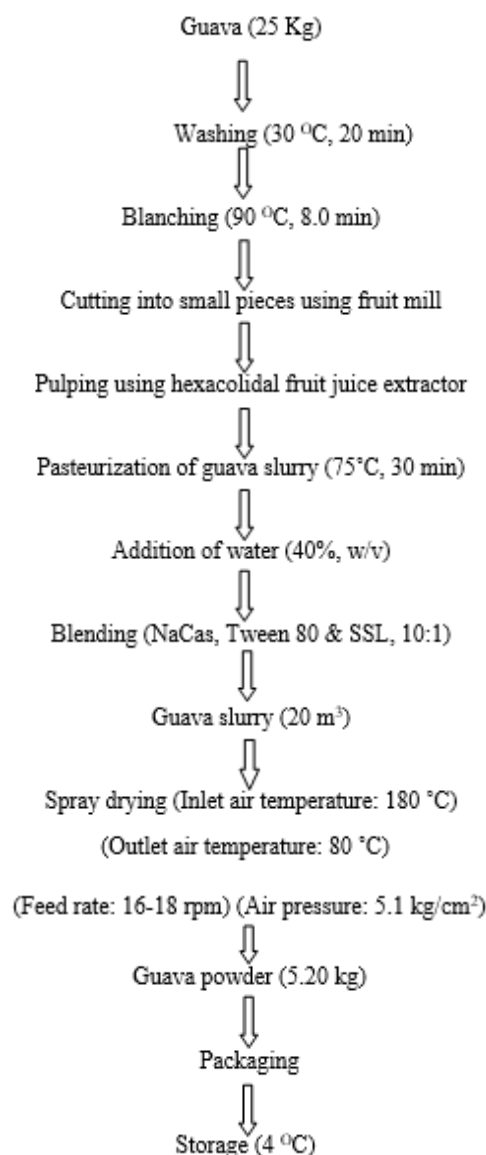


Fig 1: Process flow chart for development of guava powder.

Moisture

The moisture content was determined by drying the powder samples in a vacuum oven (Thermoline Scientific, Australia) at 70 °C for 24h by AOAC method, 927.05 (AOAC, 2000). The samples were removed from the oven, cooled in desiccators and weighed. The drying and weighing processes were repeated until constant weights were obtained. All the trials were performed in triplicate.

Water activity

Water activity or a_w is the partial vapor pressure of water in a substance divided by the standard state partial vapor pressure of water. In the field of food science, the standard state is most often defined as the partial vapor pressure of pure water at the same temperature. Using this particular definition, pure distilled water has a water activity of exactly one.

Glass transition temperature (T_g)

T_g was measured using Differential Scanning Calorimetry (DSC) (ASTM E1356, USA). DSC was used in representative samples. Differential scanning calorimetry (DSC) analysis was carried out over the temperature range of 20 to 180°C using Mettler DSC-028 (Mettler Toledo, USA) with heating rate of 10 °C/min. Sample weights ranged between 10 and 15mg and were conditioned at room temperature prior to analysis as per the previous report [12].

X-ray diffraction (XRD)

X-ray diffraction has been in use in two main areas, for the fingerprint characterization of crystalline materials and the determination of their structure. Each crystalline solid has its unique characteristic X-ray powder pattern which may be used as a "fingerprint" for its identification. Once the material has been identified, X-ray crystallography may be used to determine its structure, i.e. how the atoms pack together in the crystalline state and what the interatomic distance and angle are etc. XRD was studied by PANalytical-X-ray diffractometer designed for obtaining the ultimate quality diffraction data, combined with ease of use and flexibility to quickly switch to different application. Start Position [°2 θ] and their value was 10.0100, End Position [°2 θ] and value was 79.9900, Step Size [°2 θ] and value was 0.0200, Scan Step Time[S] was 0.3000, Divergence Slit Type [°] was fixed, Specimen Length [mm] was 10.00, Receiving Slit Size [mm] was 1000, Measurement Temperature was 25 °C, Cu used as anode material. Power supply was 25kv power supply with 30mA current and Goniometer Radius [mm] was used 240.00. This diffraction parameter was used in experiment.

Scanning electron microscopy (SEM)

The scanning electron microscope (SEM) uses a focused beam of high-energy electrons to generate a variety of signals at the surface of solid specimens. The signals that derive from electron-sample interactions reveal information about the sample including external morphology (texture), chemical composition, and crystalline structure and orientation of materials making up the sample (Quanta 200 model, company FEI used this experiment). All spray-dried powders were observed under a scanning electron microscope. Samples were mounted on aluminum stubbed with the help of silver glue. After that we applied 20 KV power supply and observed the sample in SEM.

Results and Discussion

Protein-SSL and protein-Tween-80 combination at concentration of 0.5 and 0.05%, respectively were added to the concentrate (guava pulp). It formed a film around the solids in the feed that facilitated production of a non-hygroscopic, free flowing, powder.

All the powders produced by spray drying were good appearance in irrespective of the color of the feed material (Fig. 2). The spray dried powders produced in this study were extremely stable at room temperature and could be easily

solubilized in water at room temperature (Cuq *et al.*, 2011) [13]. Surfactant percentage amount used in guava pulp, powder production was very low than 40% in comparison to maltodextrine (DE 6) [9].

Controlled addition of low molecular surfactants in the sugar-protein matrix will allow quantification of surface behavior of sugar-protein-LMS systems in powder formation process in spray-drying. By adding protein-LMS good quality of powder obtained without stickiness (Fig.2). Both protein and LMS compete for the air-water interface of a droplet. Since low molecular surfactants are smaller in size compared to proteins, they are kinetically advantaged to occupy the surface of a droplet. Since they have low glass transition temperature, the low molecular surfactants cannot be used as drying aids. They can effectively use to control the amount of protein at the surface of a droplet or a particle.

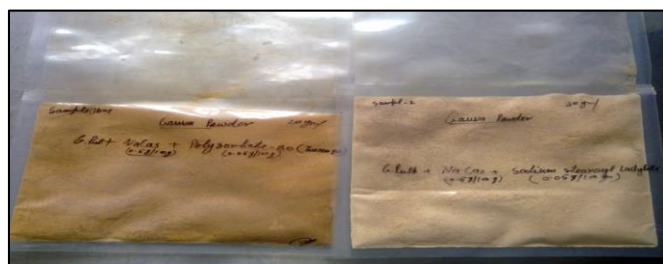


Fig 2: NaCas + Polysorbate Guava Powder NaCas+ Sodium stearoyl lactylate Guava Powder

It is interesting to note that in the case of guava sugar-NaCas solution, none of these two low molecular weight surfactants were capable of displacing the proteins from the droplet surface probably due to a very high concentration of protein. As suggested above, it can be attributed that the excess protein molecules forced to make tight multiple layers on the surface and the sub-surface of the droplets. The surface elemental analysis would later indicate that the surface of the droplet is supersaturated with proteins. This suggests that the ability of the low molecular surfactants to displace the proteins depends on the amount of protein in the system.

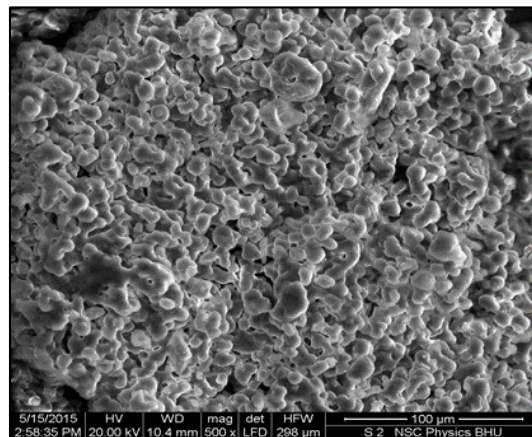
The a_w values of all powders of NaCas-SSL and NaCas-Tween80 (0.05%) are within the range of a_w values (≈ 0.85) of commercially water activity is low spray-dried powders. Water activity values of NaCas-SSL (a_w 0.85) mixed fruit pulp powder were high than NaCas-Tween-80 water activity (a_w 0.20). So NaCas-Tween-80 guava powder is more stable and their self-life is higher comparison of NaCas-SSL powder and more economical. The high a_w values are due to the fact that these two powders were semi crystalline and the moisture was mostly free water (a_w 0.85) in this sample thus, they can be considered biochemically or microbiologically quite stable. Crude fiber content in NaCas-SSL and NaCas-Tween-80 combination were 3.62 and 3.61% respectively.

One of the characteristics of spray-dried products is the low moisture content which is less than 5% (Masters, 1991). NaCas-SSL guava powder has low moisture content ($3.81 \pm 0.01\%$). The moisture content of all the powder samples is well within the range, NaCas-Tween-80 (0.05%) have high moisture content ($6.57 \pm 0.02\%$). The moisture content of all protein containing powders was high due to higher protein content in the former as proteins have greater water holding capacity in their amorphous state than in their crystalline state.

Scanning electron microscopy (SEM) analysis

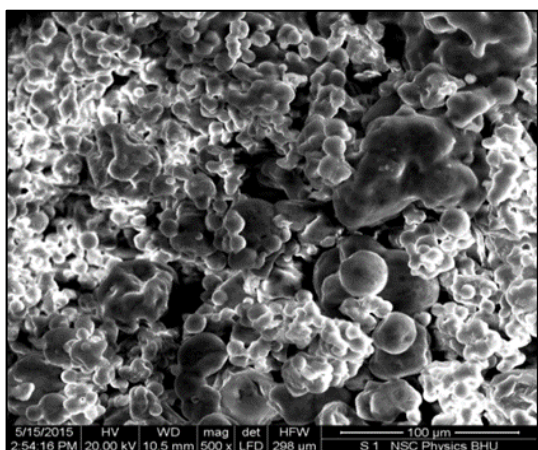
The scanning electron micrographs showed that the (NaCas 0.5%+Tween-80 0.05%) and (NaCas0.5%+SSL0.05%) formation of particle are mostly spherical (Fig.3a and b) which show amorphous behavior of hexagonal it may be crystalline and spherical form are amorphous [14]. The morphology of these particles apparently gives a good indication of the amorphous nature of the powders. More regular and spherical particles were seen at higher inlet temperatures. Similar behaviors were verified for acai [15], Products produced by spray drying. At higher drying temperatures, particles tend to inflate and form a crust, which is related to rapid water evaporation and the high pressure generated inside the particles [16, 17]. Stated that the differences in morphology and particle size of spray-dried powders occur as a result of the drying temperatures and atomization pressure. Low molecular weight surfactants such as SSL and Tween-80 supplementation provides amorphous powder which enhances its solubility and it can be used in food and beverage industry.

Fig 3a: NaCas (0.5%) + (0.05%) Tween-80 Guava powder (SEM)



HV 20.00kv, WD 10.4 mm, Mag 500x, Hfw 298, Size 100 μm

Fig 3b: (NaCas (0.5%) +SSL 0.05% Guava powder (SEM)



HV 20.00kv, WD 10.5 mm, Mag 500x, Hfw 298, Size 100 μm

X-ray diffraction (XRD) analysis

Guava powder of NaCas-SSL 0.5% and NaCas-Tween-800.05% can be seen from X-ray diffractograms that all powders containing sugar i.e. sucrose or fructose. NaCas–SSL (0.05%) and sucrose or fructose–NaCas–Tween-80 (0.05%) powder are amorphous (Fig. 4). The sharp peaks obtained through XRD reveals crystalline nature of fruit powder. Both guava powder (NaCas + SSL and NaCas+Tween80) X-ray diffractograms shows peaks are not sharp, which amorphous implicit behavior of powder. The amorphous nature of the powders may be due to very rapid evaporation and particle formation process as indicated by the low a_w values of the resultant powders.

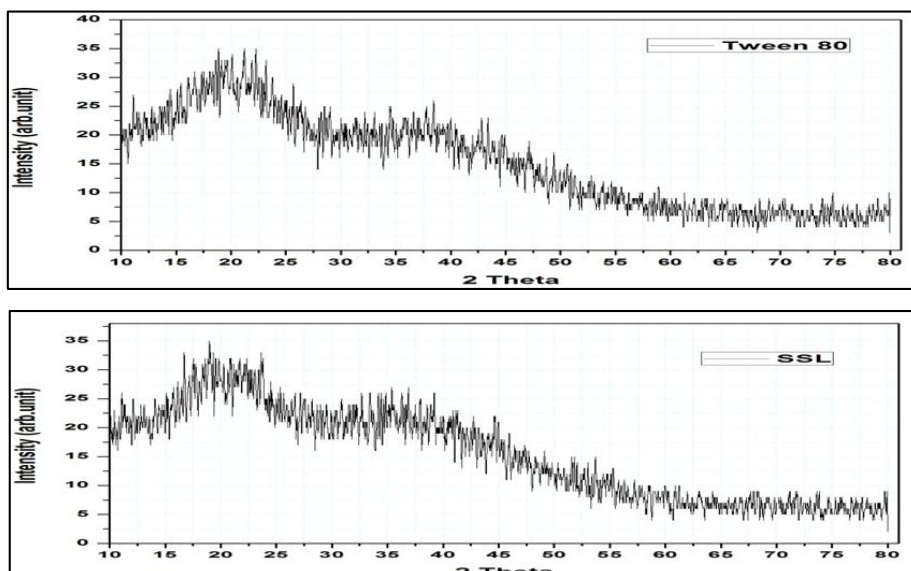


Fig 4: Diffractograms of sample (Tween 80 and SSL containing guava powder)

Differential scanning calorimetry (DSC) and TGA analysis

The T_g of multi-component mixtures was predicted using a mass weighted mean rule. The multi-component mixture is assumed to be composed of ‘n’ individual binary solid–water

mixtures, where ‘n’ is the number of solid components. T_g for each binary solid–water mixture is determined using Gordon–Taylor Equation [18].

$$T_{g, \text{ (solid-water)}} = \frac{X_s T_{g,s} + X_w K_s + w T_{g,w}}{X_s + X_w K_s + w}$$

Where,

$T_{g, \text{ solid-water}}$ is the glass transition temperature of a solid–water binary mixture

$T_{g, s}$ is glass transition temperatures

X_s and X_w are the mass fractions of solid and water in solution respectively.

$T_{g, s}$ is the $T_{g, s}$ of anhydrous solid,

$T_{g, w}$ is the $T_{g, w}$ of pure water

$K_s; w$ is a dimensionless proportionality constant that provides the moisture dependence of $T_{g, s}$.

The T_g values of spray-dried guava powder of tween is 80 °C and guava powder of SSL is 78°C (Fig.5a and b) with an increase in inlet temperature, glass transition temperatures decreased. Similar results were reported by Akkaya *et al* [19]. At glass transition temperature, an amorphous material undergoes a change from a very viscous glassy to rubbery nature due to an increase in molecular mobility and a decrease in viscosity at glass transition temperature, which may result in structural changes such as stickiness and collapse of the product [20, 21]. The glass transition temperature (T_g) of a spray-dried guava powder can be used as an indicator of stability during long periods of storage [22]. The melting temperature (T_m) obtained NaCas with SSL guava powder is 205 °C (Fig.5 c). The melting temperature (T_m) obtained NaCas with tween-80 guava powder is 210 °C (fig.5b). The amorphous form of guava powder was estimated from the enthalpy of fusion. Above the melting point of sample, sample composition was degraded and finally decomposes the sample characteristic. Fig.5c and d. represents the thermo gravimetric analysis of the guava powder sample rapid thermal degradation between 80 to 250 °C (NaCas with tween-80 guava powder) and 78 °C to 250 °C (NaCas with SSL guava powder). Maximum thermal degradation in the temperature range 205 to 250 °C (NaCas with SSL guava powder) and 210 °C to 250 °C (NaCas with tween-80 guava powder) with maximum peak at 250 °C. It is well known that a slight increase in moisture content of encapsulated powders containing sugar results in a decrease in glass transition temperature of the product below room temperature and the product will become sticky, hence microencapsulated spray-dried products should be kept below the glass transition temperature to obtain higher stability. Surface T_g of almost all guava powders have increased immensely indicating excess surface coverage of proteins in sucrose or fructose–NaCas powders. This reflects the super saturation of proteins at the droplet–air interface of the fructose or sucrose–NaCas droplets.

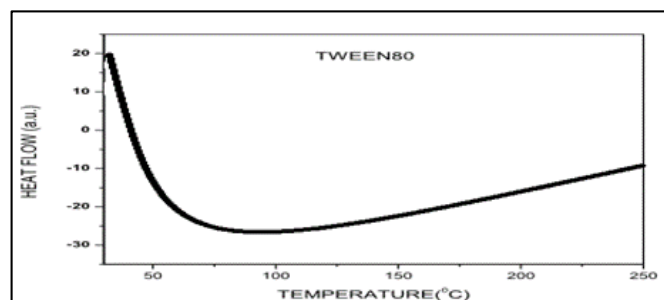


Fig 5a: DSC graph of NaCas + tween80 guava powder

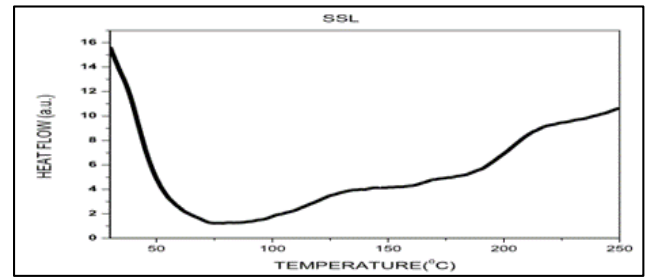


Fig 5b: DSC graph of NaCas + SSL guava powder

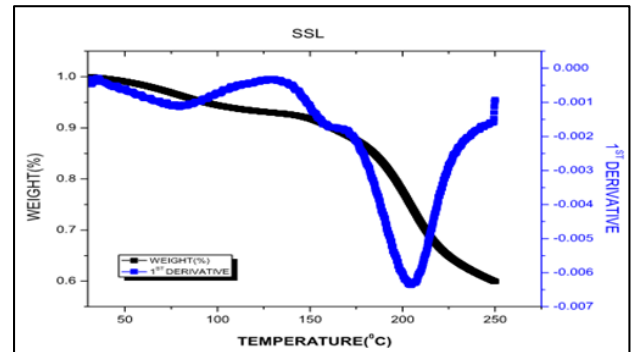


Fig 5c: TGA and DTGA of sample

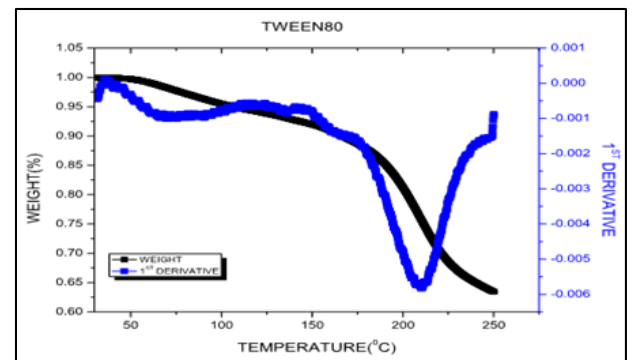


Fig 5d: TGA and DTGA of sample

Conclusions

Spray drying is potential measure to minimize postharvest. In the present investigation, amorphous guava powders were produced through spray drying with the incorporation of 0.5% of sodium caseinate-SSL (initial bulk concentration), respectively. This is an indication that NaCas can act as a very effective drying and bulking agent. The a_w values of NaCas-SSL supplemented guava powder showed a_w values (≈ 0.85) resembling to commercially produced spray-dried powders as previously reported [23]. NaCas–SSL guava powder showed low moisture content enhancing product shelf life. Crude fiber content of NaCas-SSL guava powder was lesser, showing water solubility. The glass transition temperature (T_g) of NaCas with SSL guava powder was found to be 78 °C which is above the T_g . Thermal stability of powder decreased above T_g and it can be vigorously decomposed. However, T_g of NaCas with tween-80 guava powder was found to be 80 °C which is higher than that of SSL guava powder and shows decomposition at higher temperature.

T_m of guava powder supplemented with NaCas and SSL and NaCas with tween-80 was found to be 205 and 210 °C. The enthalpy of fusion data revealed the amorphous nature of powder. TGA analysis of the guava powder sample showed rapid thermal degradation between 80 to 250 °C (NaCas with Tween80 guava powder) and 78°C to 250°C (NaCas with SSL

guava powder). This study deduce that small quantity of proteins and surfactants are required to successfully convert sugar-rich foods into powder form than other additive like maltodextrins. Addition of these carriers does not alter the resultant powder quality and poses to be economical with more consumer acceptability.

Conflict of interest statement

All the authors mutually agree to submit the manuscript in the Journal Powder Technology. The authors declare no conflicts of interest.

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