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Formulation and evaluation of *Psidium guajava* leaf extract microparticles

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Abstract
Guava (*Psidium guajava*) is an evergreen bush or little tree of the family Myrtaceae, commonly grown in common tropical and subtropical regions. Tannins, flavonoids: quercetin, a pentacyclic triterpenoid, saponins, carotenoids, lectins, leucocyanidin, ellagic acid, oleanolic acid, triterpenes, and ursolic acid are some of its essential phytoconstituents.
Various parts of the plant are reported to have pharmacological properties and are used to cure ailments like malaria, gastroenteritis, spewing, and looseness of the bowels, diarrhea, wounds, ulcers, toothache, sore throat, and swollen gums. The plant has also found use in treating illnesses such as diabetes, hypertension, and obesity. The present study aims at formulation of the extract-loaded microparticles from *Psidium guajava* leaf extract and cellulose polymers, and their evaluation. The continuous hot extraction was done using a soxhlet extractor. The encapsulation was by using cellulose, coconut oil, and castor oil. The micromeritic studies showed good flow properties of the microparticles. The SEM, DSC, and XRD analyses depicted that the surface of microparticles was smooth, showing endothermic peaks revealing that the microparticles are amorphous in nature.

Keywords: Microparticles, *Psidium guajava*, guava, ethyl cellulose, solvent evaporation, bioactive compounds

Introduction
Herbs as medicines have been the bulwark of healthcare since time immemorial [1]. Herbal medicine is one of the principal modalities in traditional, complementary, and alternative medicine. The extensive use of herbal remedies in developing and developed countries has brought enormous acceptance to herbal medicine [2]. It would be apt to say that globally, an herbal renaissance is underway as the herbs are staging a comeback. Herbal medicine has played an important role in catering to healthcare, especially the primary health, of people residing in developing countries, with its usage being more widespread in rural areas. The absence of modern healthcare coupled with the accessibility, easy availability of local herbs, and cultural preferences have been the prime reasons for their wider acceptance and use in rural areas. Traditional ethno-medicine has served as a treatment for the plethora of ailments plaguing the rural populace: contagious, infectious, communicable, and non-communicable [3].

Guava, *Psidium guajava* (Linn.), a tropical plant with a long history of traditional use, is a member of the Myrtaceae family. Guava leaves find use in treating diarrhea, gastroenteritis, and other digestive complaints, while the Guava fruit finds its use in increasing platelets in patients with dengue fever [4]. Quercetin is one of the most abundant flavonoids found in guava leaves. Flavonoids attribute much of the therapeutic properties of Guava. The drugs of natural origin suffer from poor bioavailability because of their poor physicochemical properties. Their therapeutic potency, therefore, stands diminished despite their extraordinary potential. Thus, to overcome these limitations, developing a novel herbal drug delivery system (NHDDS) with an enhanced absorption profile is of prime importance. These novel formulations are advantageous over conventional formulations concerning enhanced solubility, stability, membrane permeability, bioavailability, improved pharmacological activity through sustained release profile, reduced toxicity [5]. These novel carriers deliver the drug at a predetermined rate throughout the treatment and channel the active entity to the site of action.

While the conventional dosage forms fail to achieve these, micro/nano-sized NHDDS have future potential in enhancing the activity and overcoming problems associated with plant medicines. The advent of advanced scientific techniques has opened new doors for developing novel herbal formulations [6]. Today, one of the most attractive researches on drug delivery is the design of polymeric microparticles that can deliver the drug at the target site, at the right time, at the appropriate doses [7].

Materials and Methods

Materials: Ethyl cellulose was supplied and hydroxy propyl methyl cellulose was supplied by Vikash Drugs, Princess Street, Mumbai. Acetone, coconut oil, castor oil, Tween80 were all of analytical grade

Collection and Authentication of Plant material

The leaves of *Psidium guajava* were collected from Jule Solapur, Solapur district Maharashtra, India. The authentication of *Psidium guajava* was done from Walchand College of Arts and Science, Solapur.

Development of Formulation

Psidium guajava leaf extract microparticles were fabricated by emulsion solvent diffusion evaporation technique. Accurately weighed quantities of *Psidium guajava* leaf extract, Ethylcellulose, HPMC were taken and acetone (15 ml) was added with stirring to avoid aggregation. The organic phase was transferred slowly into the oil phase containing coconut oil (8 ml), castor oil (2 ml), and Span 80 (0.1 ml) while continuous magnetic stirring at 1000 rpm for 60 min to remove the organic solvent. Subsequently, samples were filtered and microparticles were dried. The 5 batches of *Psidium guajava* leaf extract microparticles were prepared for assessing the influence of drug: polymer ratio.

Table 1: Batches of Microparticles

Batch Code	Drug: Polymer Ratio	Polymer Ratio (EC: HPMC)
F1	1:1	3:1
F2	1:2	3:1
F3	1:3	2:1
F4	2:1	2:2
F5	2:2	3:2

Evaluation

Micromeritic studies

Angle of repose

To measure the frictional forces, the angle of repose was determined by fixed funnel method.

$\theta = \tan^{-1}(h/r)$

Where θ is the angle of repose, *r* is the radius, and *h* is the height.

Bulk density

A measured quantity of microparticles (5gm) was added to a measuring cylinder, the volume was noted and bulk density was calculated. The cylinder was then tapped mechanically 100 times to get the tapped volume for computing the tapped density.

Bulk density = bulk mass /bulk volume

Tapped density = mass / tapped volume

Carr's index

It is an indication of compressibility calculated by

Carr's index = tapped density – bulk density/ tapped density X 100

Hausner ratio

A number correlated to flowability of powder or granular material calculated by

Hausner ratio = Tapped density/ bulk density

Scanning Electron Microscopy

The morphological properties and shape of *Psidium guajava* leaf extract microparticles were recorded using ZEISS. The sample was examined at 3 Kv. The SEM analysis was done at Diya labs, Mumbai.

Differential Scanning Calorimetry

The physical and chemical changes within a material in response to temperature were analyzed using DSC Q20 V24.11 Build 124 instrument. The samples were heated from 25 °C to 200 °C at a heating rate of 10 °C/min. The DSC analysis was done at Diya labs Mumbai.

XRD analysis

XRD patterns of extract-loaded microparticles were obtained using x-ray diffractometer to find out any change in the crystallinity of drug during microencapsulation. The sample of F1 was scanned at 2θ angle over the range of 0-80°. The XRD analysis was carried out at Diya labs Mumbai.

Result

Table 2: Flow properties of extract loaded microparticles

Batch code	Angle of repose	Bulk density (g/cm^3)	Tapped density (g/cm^3)	Carr's index	Hausner's ratio
F1	27.33	0.33	0.416	20.67	1.26
F2	32.7	0.277	0.33	17.07	1.19
F3	41.47	0.33	0.4	17.5	1.212
F4	42.27	0.428	0.6	28.66	1.40
F5	41.47	0.33	0.4	17.5	1.212

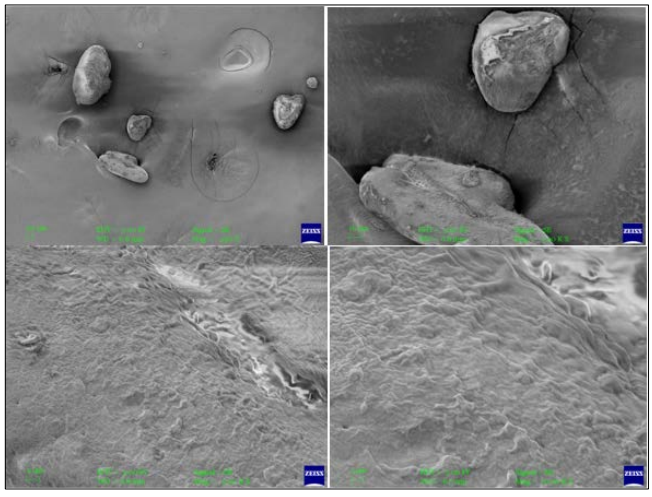

Fig 1: SEM photographs of microparticles F1

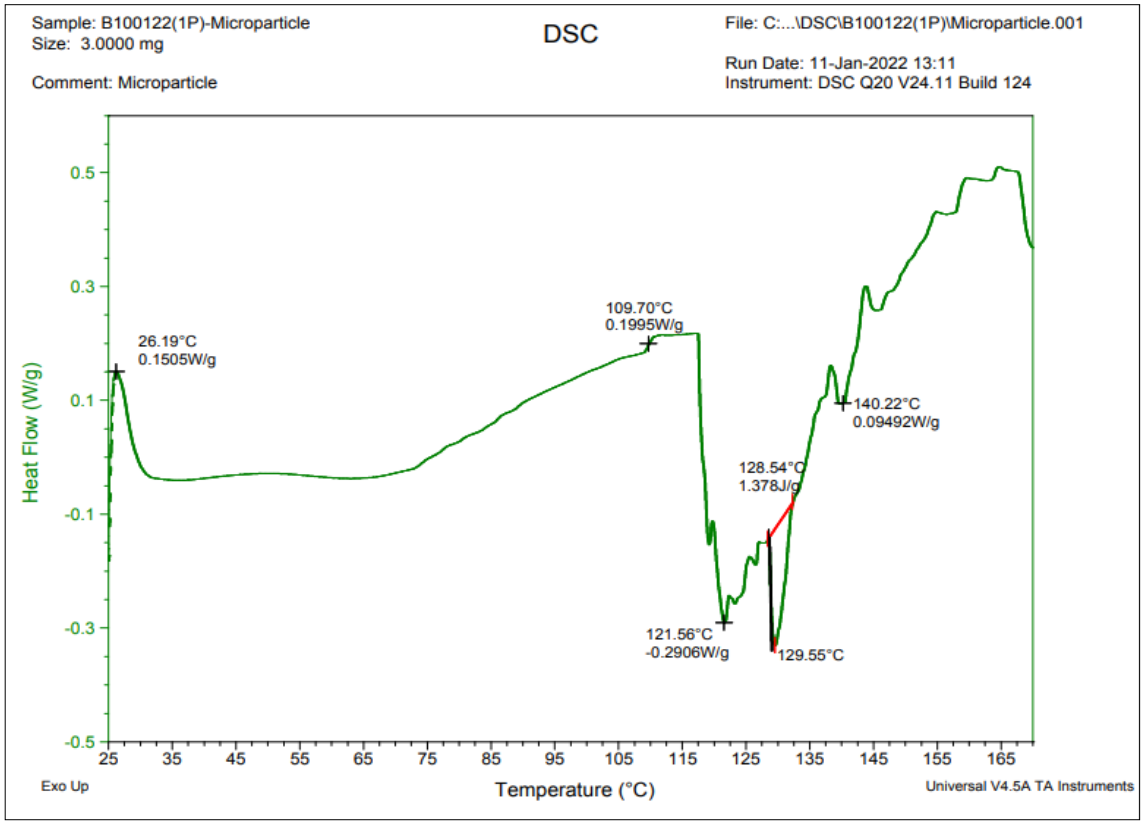

Fig 2: DSC thermogram of microparticles

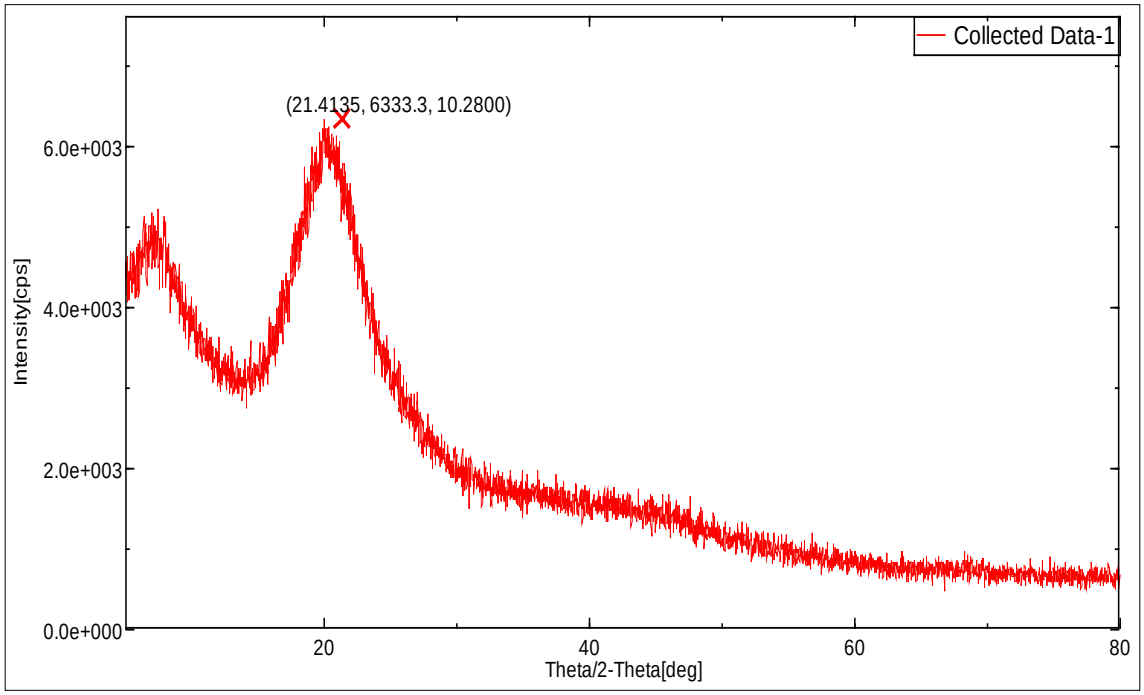

Fig 3: X-ray diffraction spectra of microparticles

Discussion

In the present study we have formulated *Psidium guajava* leaf extract microparticles and evaluated using various parameters.

Micromeritic studies are shown in table 2. The angle of repose for the batch F1was 27.33º, which shows excellent flow property, while Carr's Index 20.67%, fair flow and Hausner's ratio 1.26 depicts passable flow.

Scanning Electron Microscopy showed that the particles have smooth surface and are in 10µm range. The DSC analysis showed endothermic peak at 129.55 °C and energy associated with it is 1.378 J/g while XRD analysis showed that the material is amorphous as the peaks are diffused.

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