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Research Article

# Banana Peel Powder as Release Retardant in Matrix Tablet Formulations: *In Vitro* Evaluations and *In-Vivo Toxicity* Studies in Zebra Fish Model

SRI DURGA DEVI NAGARAJAN [1], NANDHINI DEVI GANESAN [1], NANDAKUMAR SELVASUDHA[2]*, VIGNESH MURUGAN [1], SIDDHARTH A [1], PUGAZHENDHI JEYASEELAN [1], BAVANILATHA MUTHIAH [3]

[1] Department of biotechnology, Anna University,Chennai

[2] Associate Professor,School of Pharmacy,Sri Balaji Vidyapeeth Deemed to be University,Puducherry.

[3]Associate Professor, Sathyabama University, Chennai

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**ABSTRACT**

Frequent dosing is crucial for achieving therapeutic efficacy of many drugs. The approach of sustained release comes worthy here for attaining the desired therapeutic levels without the need of frequent dosing for the treatment of various disorders. Sustained delivery of the drug could reduce the dosing frequency, adverse effects such as gastrointestinal disorders and improve patient compliance. The present study aimed at formulating Celecoxib matrix tablets by direct compression and wet granulation techniques using banana peel powder as a release retardant. It is also hypothesized that the reported gastro-protective properties of banana peel powder aid a positive effect in reducing the gastro-intestinal side effects of Celecoxib in the formulation. The prepared peel powder was subjected to physicochemical and phytochemical analysis. The flow properties of granules and the uniformity of weight, crushing strength, friability, swelling index and drug content of compressed tablets were determined. *In-vitro* drug release studies of the matrix tablets were conducted and the release rate was found to decrease with increase in amount of peel powder. The kinetics of drug release was determined by fitting the release data to different kinetic models. Kinetic modeling of *in-vitro* dissolution profiles revealed the drug release mechanism ranges from Fickian transport to anomalous type and non-Fickian transport. The results suggested that wet granulation is a suitable method to formulate sustained release with banana peel powder and it can perform therapeutically better than conventional immediate release dosage form. *In-vivo* toxicity studies also revealed that the prepared formulations show no toxicity in zebra fish models.



## INTRODUCTION

The primary objective of the sustained release formulation which was introduced several years ago was to release the drug slowly over an extended period of time, ensuring safety and improving the efficacy of the medical treatment as well as patient compliance. Hydrophilic matrix tablets are most popular drug delivery systems for oral sustained release dosage forms because of their biopharmaceutical advantages over other types of dosage forms. The popularity of the matrix tablets included their ability of accurate drug release modulation by hydration as well as cost effectiveness, and the capacity to provide a prolonged and constant therapeutic effect [1].

***Author for Correspondence:***
*Email:* nkselvasudha@gmail.com

Although the fabrication of matrix tablets seems too simple, several variables are required to be analysed for a valid formulation with the desired release kinetics. Some of the variables included polymer type and level, drug dose and solubility, polymer-drug ratio, diluent type and level, polymer-diluent ratio and porosity of the matrix. There are many established binders for the formulation of tablets [2], however the quest for evaluation of newer ones are increasing day by day. One of such includes banana peel powder. Almost majority of the parts of the banana including its flower, fruit, leaf, stem, and weevil, have been used by mankind, however banana peel is often considered as a waste material of this plant, being disposed as an organic waste or used as an animal feed for goat and cow [3]. It is interesting to know that banana peel accounts for approximately one-third of the total weight of

banana fruit which is a rich in starch (3%), crude protein (6-9%), crude fat (3.8-11%), total dietary fibre (43.2-49.7%), and polyunsaturated fatty acids, pectin, essential amino acid, and micronutrients (K, P, Ca, Mg) [4]. Therefore, the objective of the present investigation was the isolation and evaluation of banana peel powder for fabrication of sustained release matrix tablets. In order to assess the *in-vivo* toxicity of the prepared tablets, embryonic zebra fish model was opted as it is both cost and time efficient. It acts as a powerful *in vivo* model system to assess biological interactions. They are also efficient platform by which detail mechanisms by which substances elicit specific biological responses could be investigated [5]. There is a significant resemblance in cellular structure, signalling processes, anatomy and physiology of the zebra fish and other high-order vertebrates, particularly early in development. It has been reported that over 90% of the human open reading frames are homologous to genes in fish [6]. Therefore the present prepared formulations can be preliminary screened for toxicity issues in this zebra fish model.

## MATERIALS AND METHODS

### Materials

Celecoxib as pure drug was received as gift from Kniss laboratories Pvt Ltd. Banana peels were obtained from local shops in Kotturpuram, Chennai. Microcrystalline Cellulose (MCC), Starch, Lactose, and Magnesium Stearate were of analytical grade and procured from reputed vendors. All other chemicals used in the experiment were of analytical reagent (AR) grade and used without further purification.

### Methods

### Banana Peel Collection, Preparation and Characterization

The banana peels of *Musa sapientum* used for the investigation were obtained from bananas bought from a local shop in Kotturpuram, Chennai. They were air-dried for two weeks and ground into powder with a mechanical blender and sieved with sieve #20. The powdered samples obtained were thereafter stored in clean bottles at room temperature (28 ± 2°C) until needed for use.

### Material Characterization

#### *Preparation of Aqueous, Alcoholic and Hydro-alcoholic Extract*

Ninety (90) grams of the powdered peels was dispensed in 900ml of distilled water in a 1L capacity conical flask. The mixture was stirred vigorously with a rotary shaker and then allowed to stand for 48h. It was stirred again and filtered through a Whatman filter paper lined funnel into a conical flask. The filtrate was evaporated at 40°C with a water bath to obtain the solid crude extract. The same procedure was carried out for ethanol extraction and water-ethanol mixture. The extracts of *Musa sapientum* (Banana)peels were analyzed for alkaloids, tannins, glycosides, steroids, favonoids, saponins, volatile oil and resins using standard procedures [7].

#### *Determination of pH Value and Particle Size*

The pH of 1% w/v aqueous solution of banana peel powder was determined by using pH meter (Systronics, Model no.361). Banana peel powder was dispersed in distilled water and a smear of the dispersion was made and examined under microscope. The size of the particles was measured using a calibrated eyepiece micrometer.

#### *Flame Photometry Studies*

The prepared banana peel powder was analyzed using a flame photometer for the determination of potassium content. The potassium content was estimated using SYSTRONICS ® Model 128 µ Controller Based Flame photometer. (Centre for Environmental Studies, Anna University)

#### *Determination of Bulk density*

Weighed quantity of peel powder was carefully introduced into a 100ml graduated cylinder. The bulk density was calculated by dividing the weight of the sample by volume of the sample contained in the cylinder.

#### *Tapped Density*

Tapped density is the ratio of weight of the sample by volume of the sample contained in the cylinder after taps.

#### *Compressibility Index*

A simple test has been developed to evaluate the flowability of a powder by comparing the bulk density and tapped density of a powder and the rate at which it packed down. A useful empirical guide is given by Carr's compressibility index. The percent compressibility index (I) of banana peel powder was calculated using the following formula:

$$I = (1 - {V_f}/{V_0}) \times 100$$

*Where,*

$V_0$ = Initial volume,

$V_f$ = Final volume after tapping.

***Hausner Ratio***

Hausner ratio was calculated using the formula given below.

$$\textbf{Hausnerratio} = \left(\frac{\mathbf{V_0}}{\mathbf{V_f}}\right)$$

*Where,*

$V_0$ = initial volume,

$V_f$ = Final volume after tapping.

***Angle of Repose***

The angle of repose was calculated using the formula:

$$\boldsymbol{\theta = tan - 1\left(\frac{h}{r}\right)}$$

*Where,*

θ = angle of repose

h = height of pile,

r = radius of the base of the pile.

***Drug-Excipient Compatibility Studies by FTIR***

FTIR Studies are used for the detection of any possible chemical interaction between the drug and the polymer. The drug, polymer and the physical mixture of drug and polymer (each 10mg) were prepared. Pure drug Celecoxib, banana peel powder, prepared mixture formulation of pure drug and banana peel powder, were subjected to FT-IR spectral analysis. The samples were subjected to FTIR analysis using FT/IR-6300 type 'A' spectrophotometer (JASCO Inc, Easton, MD, USA). The samples were scanned from 4000$cm^{-1}$ to 400$cm^{-1}$ in the FTIR spectrophotometer. The IR spectra of formulation were compared with those of pure drug and polymers to detect any appearance or disappearance of peaks. The resulting spectra were sent for interpretation and results were obtained.

***Morphology by Scanning Electron Micrographs***

Morphology of the polymer was studied by scanning electron micrographs (SEM) taken with a scanning electron microscope (Hitachi S-300N, Germany). The samples were gold coated (About 100 A °) in KSE24M high vacuum evaporator and on metal stabs with the aid of double-sided adhesive tape. Scanned-in selected region depicting distinct morphological feature was photographed.

**Preparation of Matrix Tablets**

Matrix tablets (Celecoxib) were prepared by both dry granulation and wet granulation techniques. Three formulations each by both the techniques were prepared by varying the amount of MCC and banana polymer as per the formulation table (Table 1). In wet granulation 10% starch paste is used as binder. Lactose was used as diluent. Magnesium stearate was used as glidant (5%). The compositions of different formulations used in the study containing 100mg of Celecoxib in each case are shown in table 1. All the ingredients were passed through sieve #20. The powders except talc and magnesium stearate were blended with 10% starch paste. The wet mass was passed through sieve # 20 and the wet granules were dried at 50°C for 2 hours. The dried granules were lubricated with a mixture of talc and magnesium stearate (2:1). The lubricated granules were compressed with a maximum force of compression (4000-5000kg) using 12 mm round and slightly concave punches on a Cadmach CMD3 Rotary Tableting Machine. The prepared tablets were evaluated for tablet properties such as Hardness, thickness, weight variation, percent friability and drug content.

**Table 1:** Composition formula for the tablet formulations

| **Wet Granulation** | | | | |
|---|---|---|---|---|
| **Formulation Code** | **Drug (mg)** | **MCC(mg)** | **Peel powder(mg)** | **Lactose (mg)** |
| WG-1 | 100 | 150 | 100 | 150 |
| WG-2 | 100 | 100 | 200 | 100 |
| WG-3 | 100 | 100 | 150 | 150 |
| **Dry Granulation** | | | | |
| **Formulation Code** | **Drug (mg)** | **MCC(mg)** | **Peel powder(mg)** | - |
| DG-1 | 100 | 300 | 100 | - |
| DG-2 | 100 | 200 | 200 | - |
| DG-3 | 100 | 250 | 150 | - |

**Determination of Pre-Compression Characteristics**

Pre-compression characteristics were determined on the physical mixture of the formulations prepared by both dry and wet granulation techniques and evaluated the densities and flowability of the prepared powder.

**Evaluation of Tablets**

***Hardness, Friability, Weight Variation and Disintegration Time***

As hallmark parameters for tablet evaluation, hardness, friability and weight variation were measured and evaluated. The hardness of tablet of each formulation was measured by Monsanto hardness tester. Roche friabilator was used for testing the friability using the following procedure.10 tablets were weighed initially and placed in the friabilator, which was then

operated at 25 RPM for 4 mins or 100 revolutions, dropping the tablets a distance of 6inches in each revolution. The percentage friability was calculated using the following formula:

$$\% \text{ Friability} = \frac{\text{Initialweightoftablets} - \text{Finalweightoftablets}}{\text{Initialweightoftablets}} \times 100$$

Weight variation was evaluated and calculated by weighing 20 tablets by an electronic balance (least count-0.1mg). The average weight of 20tablets was determined and Individual weights of the tablets were compared with the average weight. Tablet disintegration tests are carried out in a two stationed digital disintegration apparatus in HCl buffer (pH 1.2) and disintegration time was noted for each formulation.

### *Swelling Index*

The tablets were weighed individually ($W_1$) and placed separately in Petri dishes containing 10 ml of phosphate buffer pH 7.4. At regular intervals (0.5,1,1.5,2,3,4,5,6,7,8,10,12 and 24 hours).The tablets were carefully removed from Petri dishes and excess water was removed using filter paper. The swollen tablets were re-weighed ($W_2$) and the % swelling index of each tablet was calculated using the following equation:

$$\% \text{ SwellingIndex} = \left(\frac{W_2 - W_1}{W_1}\right) \times 100$$

### *Content Uniformity*

Content uniformity tests were performed as per USP. Tablets are chosen randomly and drug content was determined in each individual part. The preparation complies with the test, if individual drug content is between 85% and 115% of the average content. The preparation fails to comply, if more than one individual content data is outside these limits or if one individual drug content is outside the limits of 75–125% of the average content.

### *In-Vitro* Release Study

*In vitro* drug release studies of the matrix tablets were carried out using a single-station dissolution test apparatus at 37 ± 0.5 °C and 50 rpm speed in 900 ml of HCl buffer (gastric simulated fluid, pH 1.2) as a dissolution medium for the first 2 h and in intestinal simulated phosphate buffer pH 7.4 (900 ml) for the next 4 h and pH 6.8 (900 ml) for the next 6 h. One tablet from each formulation was placed in a basket and the equipment was operated. Samples (1 ml) were withdrawn over a period of 0.5 h and the filtered the samples. The volume of dissolution medium was replenished with 1 ml of fresh dissolution medium. The absorbance of the samples was measured with a single-beam UV spectrophotometer at 255 nm and the amount of drug released was calculated.

### Release Kinetics

In order to study the drug release mechanism of the examined tablets, the dissolution profile was analyzed by various kinetic models including zero-order, first-order and Higuchi square root kinetic equations [8]. The best fit with higher correlation was found for all the formulations.

### *In-Vivo* Toxicity Studies

Preliminary safety evaluation is the prime concern for medicines intended for human use. Hence, the prepared tablet formulations were subjected to a preliminary toxicity screening. The suspension of the formulation with high amount of peel powder was prepared. Its toxicity was analyzed in zebra fish following OCED (FET) guidelines [9]. The embryos of zebra fish were taken in a 96 well-plate while each well containing 6 embryos in M4 marine broth which is taken as control well. Then the powdered formulation was prepared as solution of various concentrations (0.1, 0.2, 0.3, 0.4 and 0.5 ng/µL) and loaded into the well in triplicates around 2 to 4 hpf (hours post fertilization). Then the titer plate was visualized using ordinary stereoscopic microscope (Radical India Pvt. Ltd.) and various stages of development were characterized [10].

### Statistical Analysis

GraphPad Prism version 5.00 for Windows (GraphPad Software, San Diego California, USA, www.graphpad.com) [11] was utilized for the statistical treatment of the data. Differences between batches of tablets were considered significant when $p < 0.5$.

## RESULTS AND DISCUSSION

### Isolation and Characterization of Banana Peel Powder

The isolated banana peel powder as described earlier was subjected for preliminary phytochemical analysis along with its physicochemical characteristics. Phytochemical analysis carried out in aqueous, alcoholic and hydro-alcoholic solutions revealed presence of carbohydrates in all the three solutions

suggesting for starch like characteristics. While presence of terpenes by Salkowski test was negative in alcoholic solution and found to be positive in alcoholic and hydroalcohic solutions. Copper acetate test for terpenes was positive for all the three solutions, suggesting the presence of terpenes. Presence of phytosterols was observed in aqueous and hydro alcoholic solutions. Positive lead acetate test suggested presence of flavonoids. Phenols were found only in hydro-alcoholic extracts and saponin was absent in all of the extracts as shown in Table 2. Table 3 shows the physicochemical characteristics of the banana peel powder.

**Table 2:** Characterization of banana peels powder for phytochemical analysis.

| S.No | Name of the test | | Aqueous | Alcoholic | Hydro-alcoholic |
|---|---|---|---|---|---|
| 1 | Test for alkaloids | Wagner's test | - | - | - |
| | | Hager's test | - | - | - |
| 2 | Test for carbohydrates | Molisch test | + | + | + |
| | | Benedict test | + | + | + |
| | | Fehling test | + | + | + |
| 3 | Test for glycosides | M.Borntrager test | - | - | - |
| | | Legal test | - | - | - |
| 4 | Test for saponins | Froth test | - | - | - |
| | | Foam test | - | - | - |
| 5 | Test for terpenes | Salkowski test | - | + | + |
| | | Copper acetate test | + | + | + |
| 6 | Test for phytosterols | Libermann burchard test | + | - | + |
| 7 | Test for flavonoids | Alkaline reagent test | - | - | - |
| | | Lead acetate test | + | + | + |
| 8 | Test for phenols | Ferric chloride test | - | - | + |

**Table 3**: Physicochemical properties of banana peel powder

| Properties | Results |
|---|---|
| Colour | Pale brown |
| Odour | Characteristic |
| pH | 5.9 |
| Solubility | Soluble in warm water and Insoluble in organic solvents |
| Particle size | 74-92.5 μm (Very fine) |

### *Potassium Content by Flame Photometry*

Potassium content of banana peels was determined by Flame photometric analysis (SYSTRONICS ® Model 128 μ Controller Based Flame photometer). Potassium being water soluble was extracted from the peel powder by digestion with water in shaker incubator for 24 hours after which the sample was filtered and the filtrate was subjected to flame photometer. After calibration using known concentrations of potassium ions, the amount of potassium in the peel powder extract was determined. Concentrations of potassium ions were found to be 3.92 mg of $K^+$ per gram of peel powder.

### *Pre-Formulation Parameters of Banana Peel Powder*

The flow characteristics of the banana peel powder, as determined by their angle of repose, compressibility index and Hausner's ratio, are summarized in Table 4. It is obvious from these results that banana peel powder exhibited smaller angles of repose (32±1.03). Pharmaceutical powders having angle of repose in the range 31-35$^o$, shows 'good' flow property. Calculated compressibility index and Hausner's ratio of banana peel powder are tabulated in Table 4. The bulk densities for banana peel powder were relatively small. This lower density of banana peel powder potentially improves its dilution properties for low dose drug.

**Table 4:** Powder Characteristics and Flow Properties (pre-formulation characteristics) of banana peel powder

| Properties | Results |
|---|---|
| Bulk density (g/cc) | 0.525±0.02 |
| Tapped density (g/cc) | 0.588±0.06 |
| Angle of repose | 32.05°±1.03 (Good) |
| Hausner's ratio | 1.117±0.091 |
| Carr's index | 10.544±1.12 % |

***Drug Excipient Compatibility Studies by FTIR Analysis***

In order to investigate the compatibility of the drug and the isolated banana peel powder, the infrared spectra of Celecoxib-Banana peel powder physical mixture and Celecoxib sample were analyzed and interpreted. The characteristic bands of Celecoxib were well preserved in the physical mixture indication no interaction between the drug and the banana peel polymer. Addition of banana peel powder did not affect the native bands of drug sample confirming the compatibility.

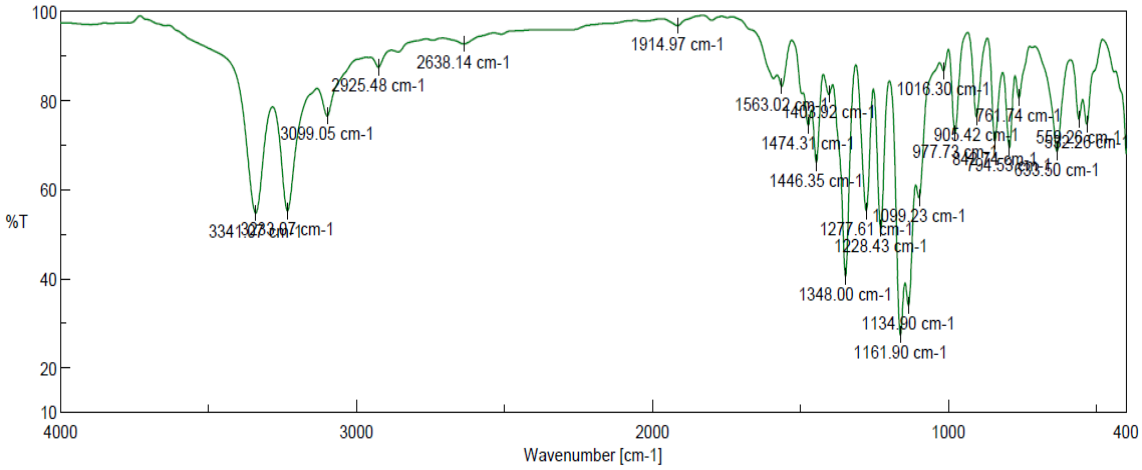


**Figure 1:** FTIR spectra of celecoxib sample. Detector - TGS; Resolution - 4 $cm^{-1}$

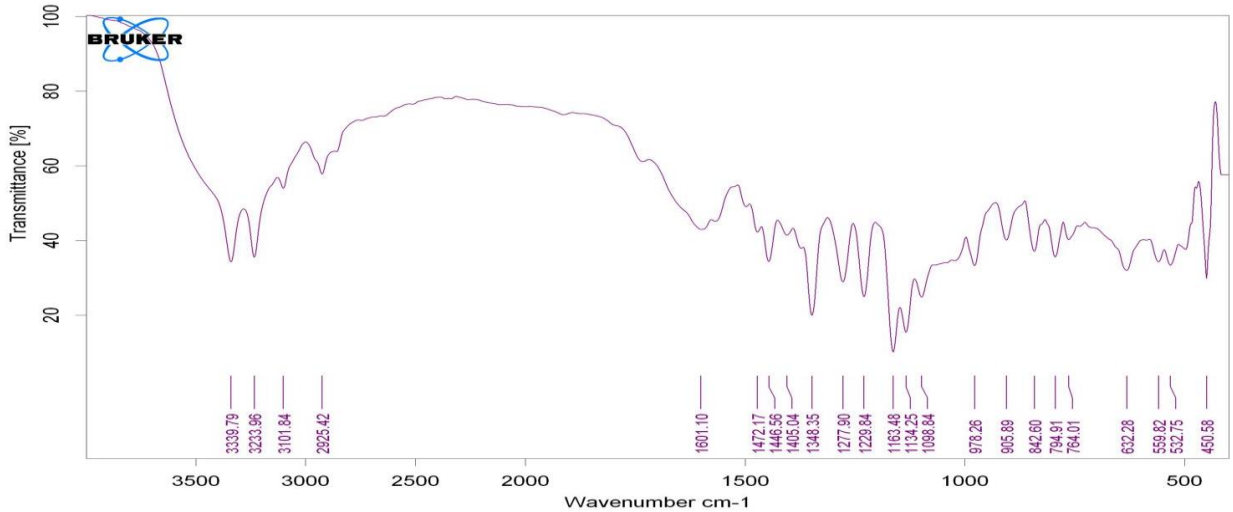


**Figure 2:** FTIR spectra of physical mixture of drug and banana peel powder in the ratio of 1:2

**Table 5.**FITR spectra analysis results ofCelecoxib and physical mixture

| Functional group | Celecoxib ($cm^{-1}$) | Physical mixture ($cm^{-1}$) |
|---|---|---|
| -$NH_2$ stretching | 3341.07 | 3340 |
| S=O | 1348,1161.9 | 1348.35,1163 |
| -NH Stretching | 1563.02 | 1563.02 |
| C-F | 1228,1277 | 1229.8,1277.9 |
| CH aromatic | 3099 | 3101 |
| CH alkyl | 2925 | 2925.42 |

***Morphology Studies by Scanning Electron Micrographs***

Morphology of the polymer was studied by scanning electron micrographs (SEM) taken with a scanning electron microscope (Hitachi S-300N, Germany). The results as scanned-in selected region depicting distinct morphological feature were photographed and presented in Fig 3.

**Pre-compression Characteristics of Physical Mixture**

Pre-compression studies were carried out on the physical mixture of the formulations prepared by both dry and wet granulation to evaluate the

densities and flowability of the prepared powders. In formulations prepared by dry granulation, based on the angle of repose, Carr's index and Hausner's ratio, formulation DG-2 had excellent flow properties while formulation DG-1 and formulation DG-3 had good flow properties. In formulations prepared by wet granulation, based on the evaluation parameters, all the three formulations showed excellent flow properties without the addition of any glidant. The excellent flow property in formulations is attributed to the wet granulation process. Thus formulations prepared by wet granulation had enhanced flow properties when compared to formulation prepared using dry granulation.

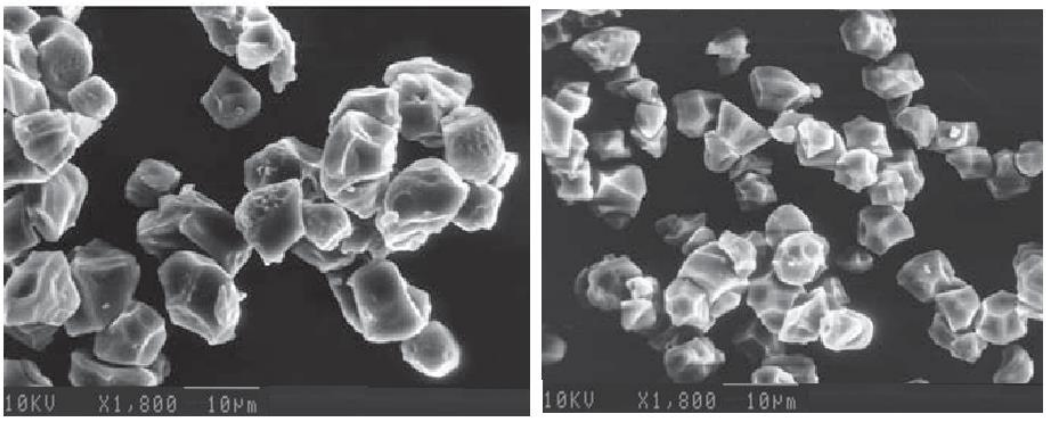


**Figure 3:** Scanning electron photomicrographs of banana peel polymer

**Table 6:** Pre-compression Properties of Granules prepared by both dry and wet granulation

| Formulation | Angle of Repose (θ) | Compressibility Index (%) | Hausner's Ratio | Bulk Density (g/cc) | Taped Density (g/cc) | Total Porosity (%) |
|---|---|---|---|---|---|---|
| **WG-1** | 26.00°±0.71 | 8.754±0.28 | 1.096±0.96 | 0.542±0.04 | 0.594±0.05 | 63.47±1.01 |
| **WG-2** | 27.14°±0.86 | 8.837±0.31 | 1.097±0.99 | 0.557±0.02 | 0.611±0.09 | 64.26±1.21 |
| **WG-3** | 25.46°±0.89 | 10.784±0.94 | 1.121±0.56 | 0.546±0.08 | 0.612±0.10 | 64.14±1.71 |
| **DG 1** | 32.8°±0.91 | 13.52±0.96 | 1.165±0.73 | 0.533±0.09 | 0.621±0.05 | 64.17±1.46 |
| **DG 2** | 29.05°±0.79 | 10.147±0.89 | 1.101±0.76 | 0.542±0.12 | 0.597±0.06 | 63.85±1.16 |
| **DG 3** | 34.59°±0.87 | 8.967±0.87 | 1.098±0.87 | 0.591±0.07 | 0.591±0.03 | 63.96±1.25 |

*WG = Wet Granulation and DG = Dry Granulation*

**Table 7:** Properties of the Compressed Tablet prepared by both Dry Granulation and Wet Granulation techniques

| Formulation | Diameter (mm) | Thickness (mm) | % Weight Variation | Hardness (Kg/cm2) | % Friability | Drug Content (%) |
|---|---|---|---|---|---|---|
| **DG-1** | 8.10±0.86 | 3.12±0.12 | 502.966±6.41 | 2.5±0.11 | 0.8±0.01 | 102±1.23 |
| **DG-2** | 8.13±0.82 | 2.89±0.20 | 498.87±4.36 | 2.5±0.19 | 0.63±0.02 | 95±1.10 |
| **DG-3** | 8.22±0.71 | 2.97±0.13 | 506.33±6.87 | 3.5±0.51 | 0.75±0.01 | 88±1.12 |
| **WG-1** | 7.99±0.91 | 3.04±0.16 | 493±2.64 | 8±0.74 | 0.47±0.02 | 108±1.61 |
| **WG-2** | 8.14±0.39 | 3.10±0.10 | 491.33±7.23 | 8.2±0.69 | 0.35±0.03 | 103±1.54 |
| **WG-3** | 7.96±0.47 | 2.98±0.11 | 499.7±0.61 | 13.5±0.81 | 0.62±0.06 | 98±1.08 |

Post compression parameters of the tablets prepared by dry and wet granulation methods

## Evaluation of Prepared Tablets

The in-process quality control tests on the different formulations included Angle of Repose, Compressibility Index, Hausner's ratio, Bulk Density, Tapped Density and Drug Content (Table 6).The powder mixtures of tablet formulation were evaluated for Angle of Repose, Compressibility Index, Bulk Density, Total Porosity and Content Uniformity. The results of Angle of Repose (<30) indicate good flow properties of the powder mixture (except DG-1 & DG-3), this was further supported by lower Compressibility Index values. Generally, Compressibility Index values up to 15% results

in excellent flow properties. The total porosity of the powder mixture is also good, indicating that the packing of powder mixture is almost closet type and also further confirming that the particles are not of greatly different size. The drug content in the weighted amount of the powder mixture was found to be almost uniform. All these results indicated that the powder mixture of the tablet possessed satisfactory flow properties, compressibility and drug content. The tablets were subjected to various evaluation tests such as thickness, uniformity of weight, drug content, hardness and friability. All formulation showed uniform thickness. The average percentage deviation of all the tablet formulation was found to be within the limit, and hence all formulation passes the test for uniformity of weight, as per official requirement. The hardness and friability test is the indirect measure of tablet strength. The Formulations prepared by wet granulation technique showed a comparatively high hardness value, this may be due to difference in compression force. In the present study the percentage friability for the tablet was below 1%, indicating that the friability is within the prescribed limit.

Hardness of the developed formulations varied from 2.5 kg/cm$^2$ to 3.5 kg/cm$^2$ in dry granulation and from 8 kg/cm$^2$ to 13.5 kg/cm$^2$ in wet granulation. The formulations prepared by dry granulation did not pass the United States Pharmacopoeia (USP) limits of 4 kg/cm$^2$ to 6 kg/cm$^2$. Formulations WG-1 and WG-2 prepared by wet granulation passed the U.S.P hardness limits. Friability of the formulations prepared by dry granulation varied from 0.75% to 0.8% and in wet granulation from 0.35% to 0.47% which were less than 1% as per the official requirements of USP. In both the formulations prepared by dry and wet granulation, friability values decreased with increase in concentration of peel powder. Tablets were weighed individually and weight variation was calculated. The requirements of USP are met if the weights of not more than 2 of the tablets differ from the average weight by more than the 5% percentage (for tablets whose average weights are more than 324mg). Formulation DG-2 under dry granulation and formulations WG-1 and WG-3 under wet granulation meets the USP requirements for weight variation. Tablets were chosen randomly from each formulation, crushed and the powders were evaluated for drug content using UV spectrophotometer. The results complies the tests specified by USP. All limits were within 85% - 115%.


**Figure 4:** Tablets prepared by dry granulation (A) and wet granulation (B)

In tablets formulated by wet granulation, formulation WG-1 showed 8.33±0.96% of drug release in 6.5 hours and formulations WG-2 and WG-3 showed 8.76±0.86% and 8.66±0.89% of drug release respectively. In tablets formulated by dry granulation, formulation DG-1 showed 12.76±0.99% of drug release in 6.5 hours and formulations DG-2 and DG-3 showed 7.95±0.87% and 10.1±0.95% of drug release respectively. Release percentage of marketed formulation of sustained release formulation was evaluated as 4.5±0.67% at the end of 60 minutes [12]. The percentage release of prepared optimum formulation using banana peel powder was determined as 0.67±0.73%. Thus the prepared formulation using banana peel has better retardant activity when compared to synthetic polymers used in marketed preparations.

**Release Kinetics**

In order to explore the mechanism of drug release, drug release data were analyzed by four mathematical models as shown in Table 7, through various types of regression model parameters and comparing, zero order was the best fitting kinetic model for DG-3 and WG-3, Higuchi was the best fitting model for WG-2 and Hixson Crowell model was the best regression fitting degree for WG-1. Additionally, the data of all the formulations showed values of "n" in Korsemeyer –peppas model between 0.6716 and 1.4752, which could be attributed to Anamalous transport for DG-2 and DG-3, Supercase II transport for DG-1, WG-1 &WG-2 and non-Fick Diffusion for WG-3 (Table 8).

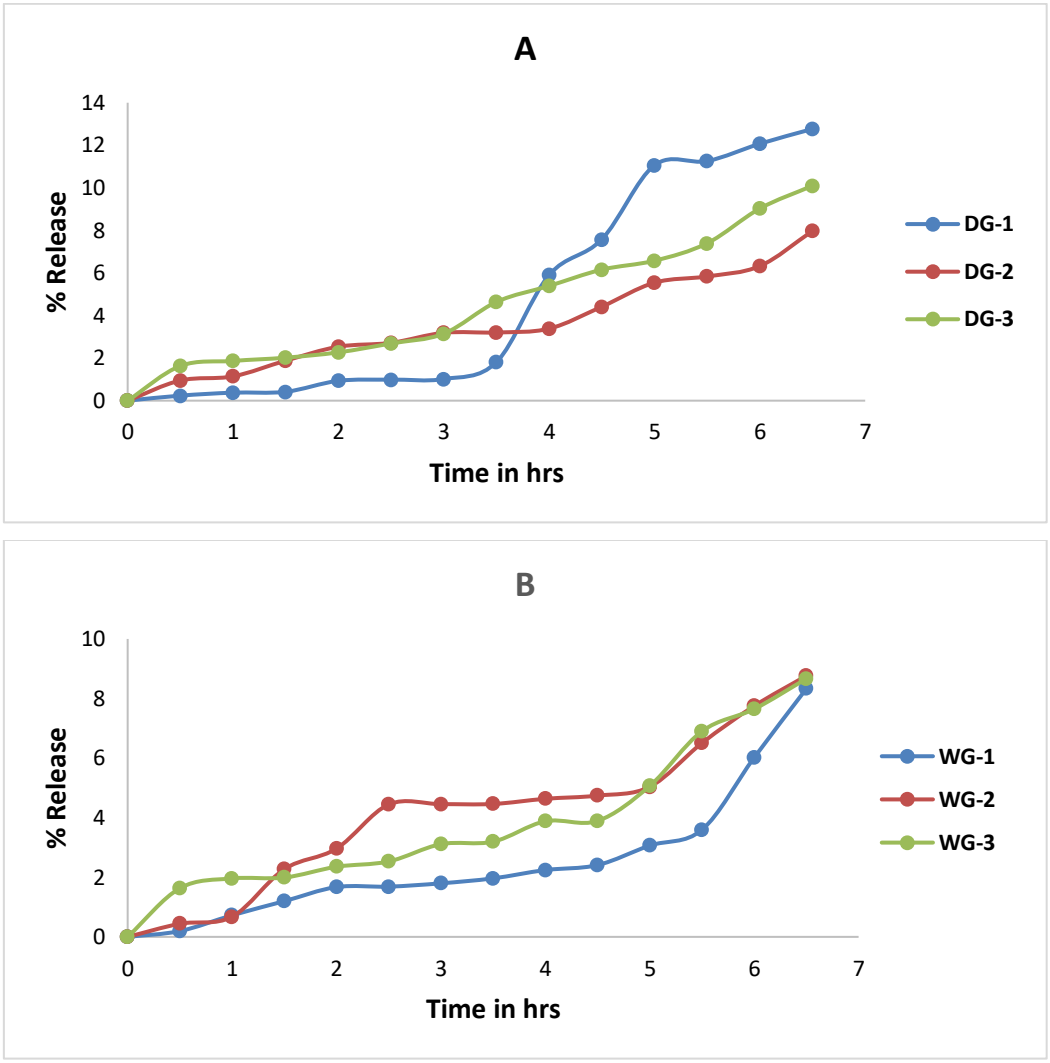


**Figure 5:** *In-vitro* Dissolution studies of physical mixture of dry and wet granulation method.A. % Release of Dry granulation and B. % Release of Wet granulation

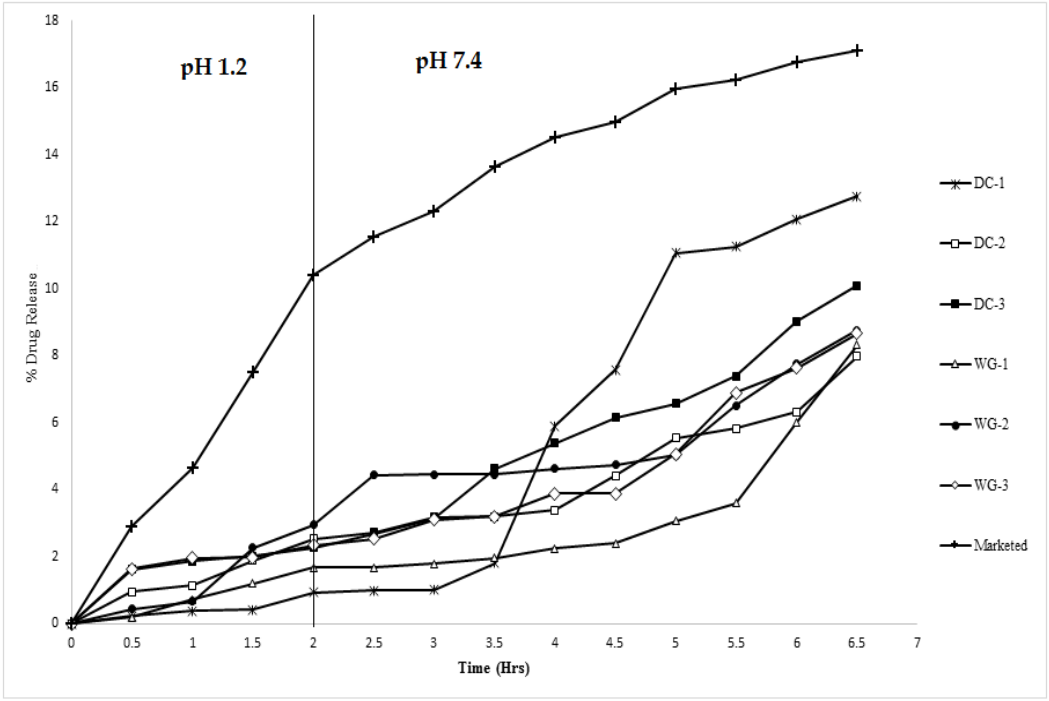


**Figure 6:** Percentage drug release from tablets formulated by dry and wet granulation

**Table 8:** Determination of release mechanism of drug from the tablets prepared from banana peel.

| Formulation code | Zero Order | First Order | Higuchi model | Hixon crowell | Korsemeyer | |
|---|---|---|---|---|---|---|
| | $R^2$ | $R^2$ | $R^2$ | $R^2$ | $R^2$ | n |
| DG-1 | 0.7586 | 0.753 | 0.6374 | 0.7549 | 0.774 | 1.4752 |
| DG-2 | 0.9362 | 0.9335 | 0.8816 | 0.9344 | 0.9315 | 0.6716 |
| DG-3 | 0.9506 | 0.9467 | 0.8687 | 0.948 | 0.866 | 0.7712 |
| WG-1 | 0.7597 | 0.7504 | 0.761 | 0.7535 | 0.9171 | 1.1753 |
| WG-2 | 0.8732 | 0.8757 | 0.9202 | 0.8749 | 0.9104 | 1.1242 |
| WG-3 | 0.9386 | 0.9363 | 0.8733 | 0.9371 | 0.8767 | 0.4604 |

**Table 9:** Mechanism of transport of drugs predicted based on the 'n' value obtained from Korsemeyer Peppas model.

| Formulation Code | Type of transport |
|---|---|
| DG-1 | Supercase II transport |
| DG-2 | Anamalous transport |
| DG-3 | Anamalous transport |
| WG-1 | Supercase II transport |
| WG-2 | Supercase II transport |
| WG-3 | Non- Fickian transport |

### *In-Vivo* Toxicity Assessment

In the present experiment, exposure of Zebra fish to test samples concentrations as high as 50 mg/100mL for 48 hpf had no effect on survival. In the experiment, exposure of Zebra fish up to 20 mg/100 mL sample did not cause pericardial and yolk sac edema, craniofacial malformations, or immobility, swim bladder non-inflation and axial malformations. Hallmark stages and physiological formations viz. Ring stage, Appearance of yolk syncytial layer (YSL) nuclei, visible embryonic shield, well extended tail and visible lateral stripes were noted normal when studied up to 48 hpf. However post 30 mg/100 mL concentration, there were observed no appearance of yolk syncytial layer (YSL) nuclei, no visible embryonic shield, no well extended tail formation and no visible lateral stripes formation. The same findings were true up to 50 mg/100mL concentration. The multinucleate layer of non-yolky cytoplasm immediately beneath the cellular blastoderm is the yolk syncytial layer (YSL). It is the first lineage-restricted extra-embryonic structure to form in the Zebra fish embryo [13]. Although it does not contribute cells or nuclei to the embryo, it is nevertheless important for embryonic development [14]. It is crucial for induction of the embryonic organizer [15], early patterning of mesoderm and endoderm [15, 16], epiboly [17, 18] and cardiac morphogenesis [18]. No sub-lethal effects induced by the test formulations on Zebra fish embryos were noted, including growth inhibition, abnormal spontaneous movement, slower heart rate, complete hatching failure, and morphological deformities.

**Table 10:** *In-vivo* toxicity assessment in zebra fish model

| Time of observation (hpf) | 2 hours | 3 hours | 6 hours | 20 hours | 48 hours |
|---|---|---|---|---|---|
| **Visible Observation** | Ring stage | Appearance of yolk syncytial layer (YSL) nuclei | Visible embryonic shield | Well extended tail | Visible lateral stripes |
| **Control sample** | + | + | + | + | + |
| **Test sample (mg/100 ml)** | | | | | |
| **10** | + | + | + | + | + |
| **20** | + | + | + | + | + |
| **30** | + | - | - | - | - |
| **40** | + | - | - | - | - |
| **50** | + | - | - | - | - |

- Presence of visible Observation
- Death of the embryo/No visual observation

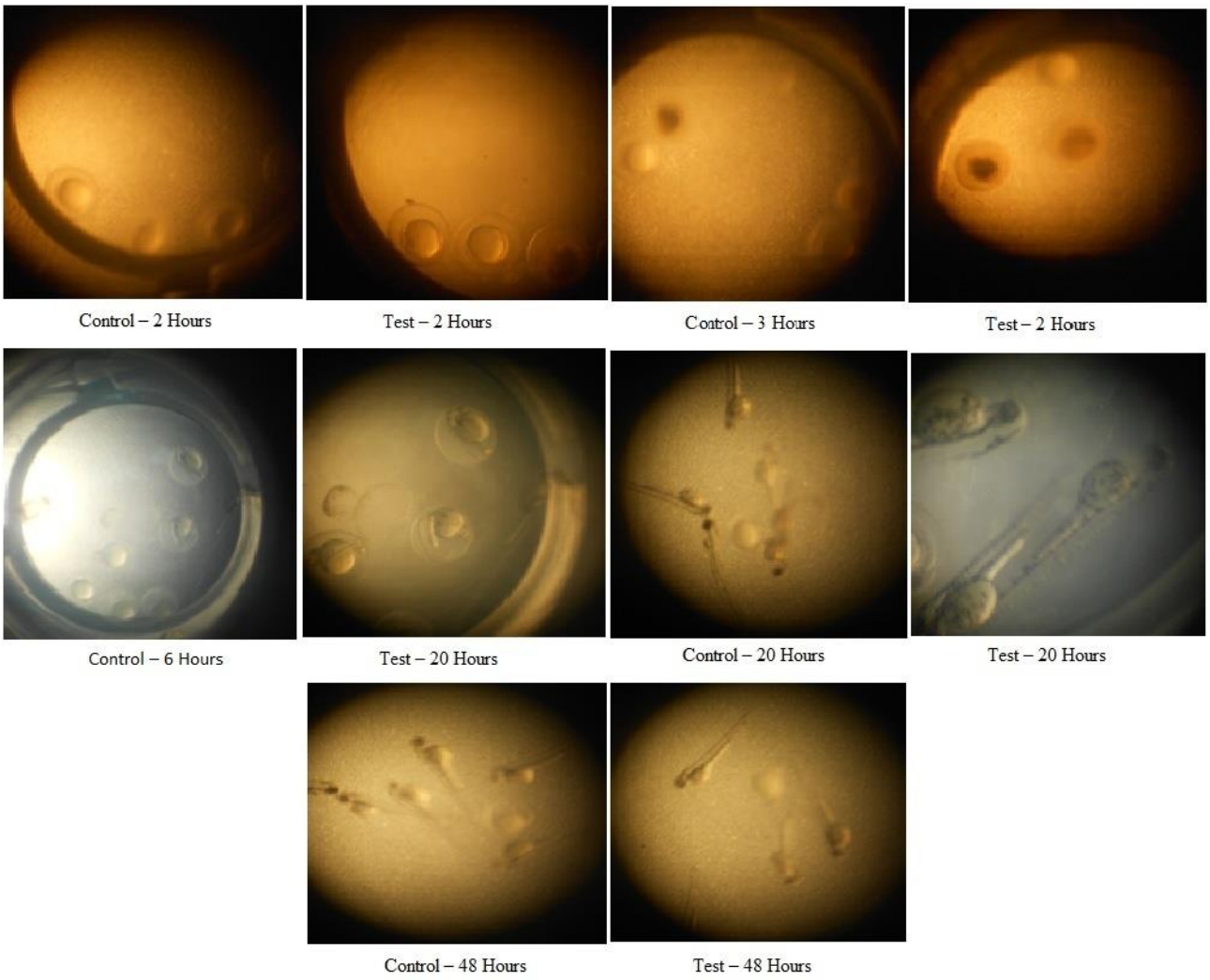


**Figure7:** Various stages of embryonic development of Zebra fish compared with control sample and 20 mg/100 mL test sample.

In our study, the highest observed effect concentration (HOEC) for the formulations that caused yolk sac and pericardial edema, craniofacial malformations including a reduction in eye size, swim bladder non-inflation and immobility in Zebra fish exposed for 48 hpf was 30 mg/100mL. The HOEC that caused non-appearance of yolk syncytial layer (YSL) nuclei, absence of visible embryonic shield, absence of well extended tail and absence of visible lateral stripes was 30mg/100mL upto 50 mg/100mL at 3 hpf upto 48 hpf. In conclusion, the studied formulations should be regarded as safe in terms of developmental toxicity in Zebra fish.

## CONCLUSION

The present study successfully formulated Celecoxib matrix tablets by direct compression and wet granulation techniques using banana peel powder as a release retardant. In the present research work tableting properties of Banana peel powder were evaluated with an aim of finding the possibility whether Banana peel powder is an excellent pharmaceutical excipient for tablet formulation. Following conclusions have been drawn from the present study.

1) Banana peel powder showed irregular granule morphology.

2) It exhibits excellent flow characteristics.

3) It showed excellent functional properties as release retardant.

4) Its compatibility and compressibility are comparable.

5) Result also shows that in tablet formulation, Banana peel powder would be more useful in formulating sustained release tablets by wet granulation technique. Thus, Banana peel powder could be useful in the formulation to produce tablet with desirable release control and mechanical properties for particular purposes.

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**DECLARATION OF INTEREST**
The authors declare that there is no conflict of interest in the present manuscript.

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