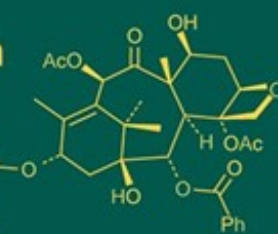
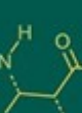
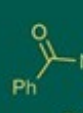
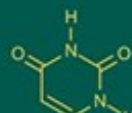


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**Dignati C Tandel**

College of Fisheries Science,  
Kamdhenu University, Veraval,  
Gujarat, India.

**Kailash H Vadher**

College of Fisheries Science,  
Kamdhenu University, Navsari,  
Gujarat, India.

**Rajesh V Chudasama**

College of Fisheries Science,  
Kamdhenu University, Veraval,  
Gujarat, India.

**Krina D Patel**

Center of Excellence in  
Aquaculture, Kamdhenu  
University, Ukai, Gujarat,  
India.

## Evaluation of proximate composition, qualitative phytochemical profile and antibacterial activity of orange, apple, papaya and mango fruit peels

Dignati C Tandel<sup>1</sup>, Kailash H Vadher<sup>2</sup>, Rajesh V Chudasama<sup>3</sup>, Krina D Patel<sup>4</sup>

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**ABSTRACT**

This study was conducted to evaluate the proximate composition, qualitative phytochemical screening as well as antibacterial activity of ethanolic extract of orange, apple, papaya and mango fruit peels. Proximate composition was determined using standard AOAC methods. Qualitative analysis of phytochemicals of the extract was carried out in accordance with standard methods, and antibacterial activity of the extract were tested in-vitro against *Vibrio parahaemolyticus* by the disc diffusion method with different concentrations (250, 500, 750, 1000 ppm). Phytochemical analysis of the extract indicates the presence of flavonoids, alkaloids, tannins, saponins, and phenolic compounds. The ethanolic extract of different fruit peels used at various concentrations showed varied antibacterial activity against the tested bacteria. Overall study concludes that orange, apple, papaya and mango peels contain phytochemicals, which may contribute to antibacterial property and ethanolic extract of peels shows antibacterial potential against *Vibrio parahaemolyticus*, varying according to extract concentration.

**Keywords:** Fruit peel, antibacterial activity, phytochemical, *Vibrio parahaemolyticus*

**1. Introduction**

The fruit processing industry generates large quantities of peels that are often discarded as low-value waste, despite containing lipids, proteins, minerals, vitamins and other bioactive compounds such as phenolics and flavonoids, carotenoids and essential oils (Habiba *et al.*, 2025) [1]. Growing interest therefore focused on valorizing these by-products as a source of functional ingredients, animal feed, nutraceuticals and natural preservatives, offering both waste reduction and sustainable resource recovery.

Proximate composition of fruit peels provides basic information about their nutritional characteristics and their applications. Parameters including moisture, crude protein, fat and total ash are commonly measured to characterize plant-derived materials and assess their nutritional qualities (Zahid *et al.*, 2021; Teshome *et al.*, 2023) [3, 2]. Previous studies have shown substantial variation in the proximate composition of fruit peels according to species, maturity stage and processing conditions. Comparative analysis of different fruit peels has revealed variation in lipids, proteins, carbohydrates, fibre and ash content which reveals the nutritional quality of peel material are depends on their source (Singh *et al.*, 2016; Zahid *et al.*, 2021) [4, 3]. Therefore, comparative evaluation of proximate composition is necessary to identify peels nutritional attributes and their suitability in food, feed and other functional applications.

In addition to their nutritional sources, fruit peels are an important source of secondary metabolites such as phenolic compounds, flavonoids, alkaloids, tannins and other phytochemicals. These metabolites have attracted considerable interest because of their antioxidant, antibacterial, and other biological properties (Wolfe *et al.*, 2008; Oikeh *et al.*, 2020) [6, 5]. Previous studies on phytochemical analysis have demonstrated that considerable variation in the concentration and composition of bioactive compounds is present among different fruit peels (Suleria *et al.*, 2020) [7]. Mango peel has been reported to contain high concentrations of phenolic and flavonoid compounds, including gallic acid, quercetin which contribute to its antioxidant activity (Kim *et al.*, 2010) [8]. Likewise, apple peel also possesses antioxidant activity associated with high concentrations of flavonoids and other phenolic compounds (Wolfe *et al.*, 2003) [9]. Phytochemical screening performed under a common experimental framework provides valuable information regarding the bioactive potential of different fruit peels.

**Corresponding Author:****Dignati C Tandel**

College of Fisheries  
Science, Kamdhenu  
University, Veraval,  
Gujarat, India.

## 2. Materials and Methods

### Fruit peel processing

Fruits were bought from the local market and washed and their peels were then collected. Peels were dried at 50 °C in a dryer for three to four days. These dried peels were ground into fine powder and stored in airtight containers.

### Proximate analysis

The proximate composition analysis was carried out by adopting standard methods outlined in the Association of Official Analytical Chemists (AOAC, 2023) [10]. The analysis was carried out in triplicate and results are presented as mean values.

### Preparation of extract

Ethanol was used as the solvent for extraction. For extraction, 10 gram of fruit peel powder were soaked in 80% ethanol for 24 hrs. Each preparation was filtered with Whatman filter paper and the extract was dried in a rotary evaporator. The dried extract was stored in an airtight, well-labelled container and stored at 4 °C in a refrigerator until further analysis (Shetty *et al.*, 2016) [11].

### 2.1 Qualitative analysis of phytochemical constituents

#### Test for tannins

The test was performed by adding a few drops of 0.1% ferric chloride solution to 1 ml of the sample extract and was observed for a brownish-green or a blue-black coloration (Thakur *et al.*, 2020) [12].

#### Test of saponin (frothing test)

In the test tube, 0.5 grams of extract was added in to 5 ml of distilled water and shaken vigorously. The solution was observed for stable froth. After adding a few drops of olive oil to the foam, it was mixed vigorously and observed for emulsion formation (Ayoola *et al.*, 2009) [13].

#### Test of flavonoids

The test was performed by adding 5 ml of dilute ammonium solution along with a few drops of concentrated sulfuric acid in 50 mg of peel extract in 100 ml of distilled water. The formation of yellow color indicates the presence of flavonoids (Thakur *et al.*, 2020) [12].

#### Test of terpenoids

0.5 gram of each extract was dissolved in 2 ml of chloroform. Subsequently, 3 ml of concentrated sulfuric acid was added to form distinct layers. The development of reddish-brown coloration at the interface between the two layers indicates the presence of terpenoids (Ayoola *et al.*, 2009) [13].

#### Test of alkaloids

For alkaloids, 50 mg of extract was dissolved in 2 ml of dilute hydrochloric acid and mixed. Filtered the solution and 1 ml of filtrate clear taken in clean test tube and added 1 to 2 drops of Mayer's

reagent and the precipitate was observed, which indicates the presence of alkaloids (Evans, 1997) [14].

### Test of steroids

To test steroids, 1 ml of peel extract was taken and 2 ml of chloroform was added. To this mixture, 1 ml of sulfuric acid was added and formation of a reddish-brown ring indicates the presence of steroids (Thakur *et al.*, 2020) [12].

### Test of phenols

1 ml of peel extract was taken in 2 ml of distilled water. Subsequently few drops of a ferric chloride were added and the appearance of blue/green colors confirmed the presence of phenols (Thakur *et al.*, 2020) [12].

### In vitro assay of extract for antibacterial activity

The antibacterial activity of the extract was evaluated using the disc diffusion method. Four sterile paper discs with one control were immersed in medium containing extract at concentrations of 250, 500, 750, and 1000 ppm, along with DMSO as a control. The discs were subsequently placed on the surface of petri dishes previously inoculated with *Vibrio parahaemolyticus* (ATCC17802<sup>TM</sup>) and incubated at 37°C for 24 h. After incubation, the diameter of zones of inhibition was measured. A sample producing a larger zone of inhibition than the control was considered to exhibit antibacterial activity (Haroen *et al.*, 2018) [15].

### Statistical analysis

All experiments were conducted in triplicate. Data were expressed as Mean  $\pm$  SE. Data were analysed using Microsoft Excel 2017 and Statistical Package for the Social Science (SPSS v.24.0). At each concentration, differences among fruit peel extracts were analysed using one-way ANOVA followed by Duncan's multiple range test at  $p < 0.05$ . (Snedecor *et al.*, 2014) [16].

## 3. Results and Discussion

### 3.1 Proximate composition of peel

Proximate composition of different fruit peels is presented in Table 1. Moisture content was relatively low across all samples ranging from  $5.89 \pm 0.09$  % in mango peel to  $6.50 \pm 0.08$  % in papaya peel. The low moisture content indicates effective drying and better storage stability. The crude protein content varied among all peels, with highest value recorded in papaya peel ( $10.20 \pm 0.07$ %), followed by orange peel ( $7.15 \pm 0.09$ %), mango peel ( $6.30 \pm 0.08$ %) and the lowest in apple peel ( $4.28 \pm 0.13$  %). Fat content was highest in apple peel ( $6.85 \pm 0.11$ %) and lowest in orange peel ( $1.35 \pm 0.07$ %). Ash content was highest in papaya peel ( $4.06 \pm 0.06$ %) followed by orange peel ( $3.66 \pm 0.04$ %), while mango and apple peels contained lower levels ( $2.62 \pm 0.08$  % and  $2.56 \pm 0.08$ % respectively).

**Table 1.** Proximate composition of different fruit peels

| Parameters   | Orange Peel     | Apple Peel      | Papaya Peel      | Mango Peel      |
|--------------|-----------------|-----------------|------------------|-----------------|
| Moisture (%) | $5.89 \pm 0.05$ | $5.82 \pm 0.10$ | $6.50 \pm 0.08$  | $5.79 \pm 0.09$ |
| Protein (%)  | $7.15 \pm 0.09$ | $4.28 \pm 0.13$ | $10.20 \pm 0.07$ | $6.30 \pm 0.08$ |
| Fat (%)      | $1.35 \pm 0.07$ | $6.85 \pm 0.11$ | $3.25 \pm 0.08$  | $2.55 \pm 0.12$ |
| Ash (%)      | $3.66 \pm 0.04$ | $2.56 \pm 0.03$ | $4.06 \pm 0.06$  | $2.62 \pm 0.08$ |

Value are mean  $\pm$  SE ( $n = 3$ )

### 3.2 Qualitative screening phytochemical

The results of qualitative phytochemical screening of ethanolic extract of orange, apple, papaya and mango peels revealed the presence of tannin, saponins, flavonoids, terpenoids, alkaloids, steroids and phenols (Table 2). Flavonoids and phenols were detected in all four fruit peel extracts, indicating that these phytochemicals are common constituents of orange, apple, papaya and mango peels. Tannins were

present in orange, apple and mango peel extracts but not detected in papaya peel extract. Saponins were observed in apple, papaya and mango peel extracts but not present in orange peels. Terpenoids were observed in orange and apple peel extracts, while alkaloids were present in orange and mango peel extracts. Steroids were found only in papaya peel extract. The variation in observation may due to difference in plant and extraction method.

**Table 2.** Qualitative phytochemical constituents of ethanolic extract of orange, apple, papaya and mango peel

|            | Orange peel | Apple peel | Papaya peel | Mango peel |
|------------|-------------|------------|-------------|------------|
| Tannin     | +           | +          | -           | +          |
| Saponin    | -           | +          | +           | +          |
| Flavonoids | +           | +          | +           | +          |
| Terpenoids | +           | +          | -           | -          |
| Alkaloids  | +           | -          | -           | +          |
| Steroids   | -           | -          | +           | -          |
| phenols    | +           | +          | +           | +          |

Key: + = Present, - = Absent

### 3.3 Antibacterial activity

Antibacterial activity of different fruit peel powder extract tested against *Vibrio parahaemolyticus* Bacteria by disc diffusion method and displayed in Table 3. All tested fruits peels showed antibacterial activity against *Vibrio parahaemolyticus*. The findings indicate that susceptibility depends on the extract concentration, higher concentra-

tions make bacteria more susceptible to the extract. Among the evaluated extracts, orange peel extract showed the highest inhibition, followed by papaya, mango and apple peel extracts. The maximum inhibition zone is observed for orange peel extract at 1000 ppm ( $10.13 \pm 0.22$  mm), whereas the minimum was observed for apple peel extract at 250 ppm ( $5.77 \pm 0.26$  mm).

**Table 3.** Antibacterial activity of different fruit peel powder extracts against *Vibrio parahaemolyticus* Bacteria on Mueller-Hinton agar using disc diffusion method

| Compound | Concentrations (ppm)   |                      |                   |                    |
|----------|------------------------|----------------------|-------------------|--------------------|
|          | 250                    | 500                  | 750               | 1000               |
|          | Zone of Inhibition(mm) |                      |                   |                    |
| OPE      | $8.07 \pm 0.35^a$      | $8.60 \pm 0.37^a$    | $9.27 \pm 0.19^a$ | $10.13 \pm 0.22^a$ |
| APE      | $5.77 \pm 0.26^c$      | $6.20 \pm 0.20^c$    | $7.00 \pm 0.23^c$ | $8.07 \pm 0.09^b$  |
| PPE      | $7.10 \pm 0.23^b$      | $7.57 \pm 0.33^b$    | $8.40 \pm 0.23^b$ | $9.53 \pm 0.26^a$  |
| MPE      | $6.23 \pm 0.24^{bc}$   | $6.73 \pm 0.28^{bc}$ | $7.77 \pm 0.14^b$ | $8.60 \pm 0.35^b$  |

Values are means  $\pm$  SE,  $n = 3$ , different superscripts indicate significant difference ( $p < 0.05$ ) by one-way analysis of Variance (ANOVA), OPE, Orange peel extract; APE, Apple peel extract; PPE Papaya peel extract; MPE, Mango peel extract.

## 4. Discussion

Phytochemical analysis of fruit peels revealed the presence of tannins, saponins, flavonoids, terpenoids, alkaloids, steroids and phenols (Abalaka *et al.*, 2016 [17]; El-Desoukey *et al.*, 2020 [18]; Jesumirhew *et al.*, 2022 [19]; Vasundra and Dutta, 2024) [20]. Phytochemicals such as saponins tannins, flavonoids and phenols are attributed to having antibacterial effects (Rahman *et al.*, 2011) [21]. Results of the study agree with the finding of Oikeh *et al.*, 2020 [5] and Abalaka and Bello, 2016 [17], who investigated the antibacterial activities of orange peel extract against different Gram-negative bacteria. Studies demonstrated that ethanolic extract of papaya peel also has an antibacterial effect against different bacteria (Akujobi *et al.*, 2010; Egbuonu *et al.*, 2016) [22, 23]. Mohamad Asri *et al.* (2022) [24] reported a ethanolic extract of papaya peel is capable of inhibiting a 14 different bacteria including *Vibrio parahaemolyticus*. The mango peel showed moderate inhibition, the zone of inhibition increased with higher concentration. Sampani *et al.* (2026) [25] observed antibacterial activity of mango peel extracts against several Gram-negative bacteria, and Kućuk *et al.* (2024) [26] reported strong antibacterial activity of mango peel extract against *E. coli*, *S. aureus*. El-Desoukey *et al.* (2020) [18] reported antibacterial activity of apple peel extract against *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aerogenes*.

## 5. Conclusion

The study indicates that orange, apple, papaya and mango peels contain phytochemicals, particularly phenolics and flavonoids, which may contribute to their antibacterial properties. Ethanolic extract of peels shows antibacterial potential against *Vibrio parahaemolyticus*, varying according to extract concentration. Although this study fo-

cused on only one extraction method, further investigation using various extraction methods should be conducted to obtain best method to extract antibacterial compounds.

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