

### Research Article

# Storage Stability Assessment of Indigenous Guava Fruits (*Psidium guajava* L.) cv. "Gola" in Response to $\gamma$ -Irradiation

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Postharvest losses in fruits and vegetables pose a key challenge to the global horticulture industry. Improved storage stability of horticultural commodities through irradiation is a technological intervention which has greater compliance over more than 60 foods and food products. The present study was intended to explore the effect of  $\gamma$ -irradiation doses on the postharvest quality of fresh guava fruits stored under ambient (temperature  $20 \pm 2^{\circ}$ C, RH 85%) conditions. Various storage specific quality attributes including physiological weight loss, respiration rate (CO<sub>2</sub>), ethylene estimation, ripening index, fruit firmness, ascorbic acid, nutraceutical potential, and enzyme kinetics were evaluated during postharvest storage at four days of interval. The results indicated that the fruits irradiated at doses (0.8 kGy and 1.0 kGy) exhibited significant reduction in physiological parameters in contrast with untreated fruit samples. Firmness, total phenolic compounds, flavonoids, and radical scavenging activity were successfully retained in irradiated guava fruits compared to control. Moreover, it has also been discovered that the fruits exposed to 1.0 kGy had considerably decreased the polyphenol oxidase (PPO) activity compared to nonirradiated fruits, thus resulting in lowering the enzymatic browning. Aside from that, irradiation guava fruits had considerably enhanced antioxidant enzyme activity ( $P \le 0.05$ ) in terms of catalase, peroxidase, and superoxide dismutase. In conclusion, the study revealed that  $\gamma$ -rays irradiation doses up to 1.0 kGy might be considered effective to improve the postharvest storage of native guava fruits variety with intact nutritional attributes.

#### 1. Introduction

Guava (*Psidium guajava L.*) is a vital commercial fruit crop of tropical to subtropical areas of the globe. It is renowned for its distinctive flavor, aroma, and higher vitamin C contents. It is a highly nutritious and functional fruit that the nutritionist frequently referred guavas as "super fruits" due to the presence of numerous health-promoting compounds [1]. It may carry approximately 84% moisture, 8-10% total carbohydrates, 1.9-2.2% proteins, 2.9-3.4% fiber, 0.6-0.8% crude fat, and ash contents 0.6-0.7% [1]. Guava fruit is also a generous source of micronutrients especially calcium, phosphorous, magnesium, potassium, iron, manganese, zinc, and vitamins A, B complex, vitamin C, etc. [2]. Additionally, guava fruit contains ample amounts of nutraceutical substances such as flavonoids (81-154 mg QE  $100 \text{ g}^{-1}$  dry weight) and total phenolic compounds (94-190 mg GAE  $100 \text{ g}^{-1}$  dry weight) [1, 3].

According to Food and Agriculture Organization's database, the annual production of guava fruit is approximately 55 million tons [4] referring to India and Pakistan collectively contributing about half of the total produce [5]. With 0.57 million tons of yearly production, guava subjugates the 4<sup>th</sup> place among Pakistan's major fruit crops, just behind

citrus, mango, and dates; however, Pakistan merely exported 0.2% of guava fruits in the fiscal year of 2021-2022, due to the lack of proper postharvest management practices [6]. Guava fruit has a high respiration rate just like other climacteric fruits; therefore, it ripens quickly and achieves early senescence if stored under ambient temperatures [7]. Higher respiration rates have been triggered by various physiological mechanisms and are governed by a natural plant hormone "ethylene" which has been generated from Lmethionine by the action of 1-aminocyclopropane-1-carboxylic acid (ACC) synthase enzyme in a complex signal transduction rout [8]. As a result, guava fruit attains its climacteric peak within a few days of harvest at normal room temperatures [7, 9]. If the appropriate measures are not implemented, significant changes that occur during postharvest storage include weight loss, abating of nutritional value, turgidity loss, and eventually decreased marketability [10]. Owing to these losses, it is estimated that approximately 30-50% of guava fruit is annually wasted [11]. Moreover, the potential for commercialization has been significantly restricted due to the guava fruit's short shelf life [12].

Different strategies are being instigated for perishable commodities to increase postharvest storage life, including controlled atmosphere (CA) storage [13], hypobaric storage [14], edible coatings [15–17], packaging [18], storage at low temperatures [19], and irradiation [20]. However, the use of low-dose gamma irradiation is one of the most promising interventions.

In recent times, gamma irradiation has been emerged as a successful tool for the management of postharvest losses by controlling microbial contamination and shelf-life extension of horticultural produce by delaying the ripening process [21, 22]. Additionally, gamma irradiation has also been employed to improve the bioactive compounds in foods with the anticipated benefits of raising the overall phenolic content, enhancing color, and increasing antioxidant activity [23–25]. However, the use of irradiation for shelf life escalation relies on cultivar type, maturity stage, respiration pattern, and the level of irradiation dose [26, 27]. Therefore, standardization of the optimum dose of irradiation is crucial for a certain fruit and cultivar type as well because each fruit crop responds differently to similar doses of irradiation [28, 29]. Even though several postharvest studies were conducted to determine the effects of gamma irradiation upon the shelf life of fruits, the scientific data is still scarce, especially for locally grown guava cultivars. With all of these considerations in mind, the current research was designed to explore the impact of varying levels of gamma irradiation on the postharvest quality of guava fruit cv. "Gola."

#### 2. Materials and Methods

2.1. Collection of Fruit Samples. Indigenous guava variety "Gola" was selected based upon the results of our previous investigation [1]. Commercially mature, disease-free fruits were collected from a guava orchard of Adaptive Research Farm Sheikhupura, Government of Punjab, Pakistan. The fruit samples were immediately precooled and then graded to uniform size and shape before starting the experimental procedures.

2.2. Experimental Procedure. After visual screening and surface drying with a muslin cloth, guava fruits were subdivided in triplicate and stored under HDPE bags as per the following treatment plan; T<sub>0</sub> (Control/untreated), T<sub>1</sub> (0.2 kGy), T<sub>2</sub> (0.4 kGy), T<sub>3</sub> (0.6 kGy), T<sub>4</sub> (0.8 kGy), and T<sub>5</sub> (1.0 kGy). Each treatment including replication consisted of 12 fruits. Packed guava fruit samples were then exposed to irradiation as per the mentioned treatment design by means of Cobalt<sup>60</sup> isotope discharging gamma irradiation at a rate of 2.117 kGy hr<sup>-1</sup> and placed at the Nuclear Institute for Food & Agriculture (NIFA), Peshawar, Pakistan. Thus, each treatment received gamma irradiation for different time periods, i.e., 0.2 kGy for 5.6 minutes, 0.4 kGy for 11.3 minutes, 0.6 kGy for 17 minutes, 0.8 kGy for 22.6 minutes, and 1.0 kGy for 28.33 minutes. Irradiated fruit samples were then stored under ambient conditions.

2.3. Fruit Analysis for Multiple Quality Parameters during *Storage*. Transitions in the following storage-related quality parameters were estimated at a fixed interval of four (04) days.

2.3.1. Fruit Firmness. Fruit penetrometer FT10 (Wagner, Italy) fitted with an 8 mm plunger was used to measure the fruit's firmness as cited by Abbasi et al. [30].

2.3.2. Determination of Physicochemical Properties of Fruits. Physicochemical characteristics including total soluble solids (TSS), pH, titratable acidity, and ripening index of guava fruits under storage were determined by adopting standard protocols. A digital refractometer (ATAGO, Japan) was used for the measurement of TSS [31]. Similarly, pH values were recorded through a portable pH meter (Model: HANNA HI 2211, USA) which was precalibrated with standard buffers as explained by Shetgar et al. [32]. Titratable acidity (TA) was calculated by following AOAC method no. 942.15 [33], and the ratio of total soluble solids to titratable acidity was used to compute the ripening index (RI).

2.3.3. Nutraceutical Attributes. Nutraceutical attributes including ascorbic acid (vitamin C) contents, radical scavenging activity (RSA), total phenolic compounds (TPC), and total flavonoids contents (TFC) were measured by following the procedures as mentioned in our previous work [1].

*2.3.4. Physiological Characteristics.* Physiological loss in weight of stored guava fruits was calculated on every 4<sup>th</sup> day of the sampling interval and measured in percentage as follows:

Physiological Loss in Weight(%) = 
$$\frac{(A-B)}{A} \times 100$$
, (1)

where *A* and *B* are the weights of fruits before and after a fixed storage period, respectively [34]. Ethylene and  $CO_2$  exchange rate (respiration rate) were measured rendering to the method described by Nair et al. [7] with some modifications. Preweighed three (03) guava fruits from each of the replication were taken in an airtight container (3.0 L volume) enclosed with a rubber septum on the lid for one hour. For reading, a syringe of a handheld three gas analyzers (Felix, USA; model F-950) was injected into the container through a

rubber septum installed at the lid side. After recording the readings on gas analyzer, the respiration rate was computed as mL CO<sub>2</sub> kg<sup>-1</sup> h<sup>-1</sup>, whereas the Ethylene was measured in  $\mu$ L C<sub>2</sub>H<sub>4</sub> kg<sup>-1</sup> h<sup>-1</sup>, rendering to the below-cited equation.

$$\begin{aligned} \text{Respiration Rate} \left(\text{mLkg}^{-1}\text{hr}^{-1}\right) \\ &= \frac{\text{CO}_2 \text{ Reading}(\text{ppm}) \times \text{Container Volume}(\text{mL}) \times 100}{\text{Sample Weight}(\text{kg}) \times \text{Sealed time}(h)}, \end{aligned}$$

$$(2)$$

while the released ethylene gas was measured as follows:

Ethylene 
$$(\mu L.kg^{-1}hr^{-1})$$
  
=  $\frac{\text{Ethylene Reading(ppm)} \times \text{Container Volume(mL)} \times 100}{\text{Sample Weight(kg)} \times \text{Sealed Time(h)}}$ .  
(3)

2.3.5. Antioxidant Enzymes Kinetics. Kinetics of antioxidant enzymes (polyphenol oxidase, peroxidase, catalases, and superoxide dismutase) were measured according to the scientific protocols adopted by Ali et al. [35] with slight modifications. For this purpose, frozen fruit pulp (1g) was taken and homogenized by using 2 mL of phosphate buffer having a pH of 7.2. After homogenization, the samples were centrifuged for 10 minutes by using refrigerated centrifugation at  $10,000 \times g$  to collect the supernatant, which was further used to compute enzymatic activities. Catalase (CAT) enzyme activity was estimated, referring to the method as reported by Liu et al. [36]. For this purpose,  $100 \,\mu$ L of enzyme extract was mixed with a freshly prepared 5.9 mM solution of  $H_2O_2$  $(100 \,\mu\text{L})$  to start the enzymatic reaction. UV-visible spectrophotometer was used to note the absorbance at 240 nm wavelength, and catalase activity was calculated in terms of units per mg of protein, where one unit (U) was defined as 0.1 unit per minute change in the absorbance. Similarly, peroxidase (POD) enzyme activities were quantified according to the procedures as mentioned by Xiao et al. [37] with few modifications. A reaction mixture was first prepared in 50 mL of phosphate buffer having  $28 \,\mu\text{L}$  of guaiacol and 19  $\mu$ L of H<sub>2</sub>O<sub>2</sub>. Then, 200  $\mu$ L of enzyme extract was combined with 3.4 mL of the abovementioned reaction mixture. The change in the absorbance was observed within 2 minutes at 470 nm wavelength, and POD activity was expressed as U mg<sup>-1</sup> protein in terms of 0.1 unit change in absorbance caused by one unit of enzyme (U) per minute.

The assay of superoxide dismutase enzyme (SOD) was done in terms of its capability to inhibit the photochemical reduction of nitroblue tetrazolium (NBT) as described by Štajner et al. [38]. A multicarrier solution was made in a falcon tube by adding together 800  $\mu$ L of double distilled water, 500  $\mu$ L phosphate buffer (pH 5.0), 100  $\mu$ L NBT, 200  $\mu$ L methionine, 200  $\mu$ L Triton-X, and 100  $\mu$ L of riboflavin. After that, the enzyme extract (100  $\mu$ L) was added in the abovementioned multicarrier solution. Then, the tubes containing the solutions were wrapped in aluminum foil and exposed to UV light for 15 min. Finally, the absorbance was recorded at 560 nm by using a spectrophotometer. One unit of SOD enzyme activity (U) was defined as the amount of enzyme that inhibited 50% NBT photochemical reduction and expressed as U mg<sup>-1</sup> protein. The kinetics of the polyphenol oxidase (PPO) enzyme were determined according to the method reported by Lo'ay and Taher [39]. For this purpose, one gram of fruit pulp was mixed with 20 mM Tris-HCl buffer (pH 7.0) and was then centrifuged at 10,000 rpm for 5 min. The supernatant was collected at -20°C to which 200  $\mu$ L of fruit extract was instantly mixed along with 3 mL of methyl catechol. The change in activity was noted in terms of absorbance change on the UV-visible spectrophotometer at the wavelength of 400 nm for 4 minutes. One unit (U) of enzyme activity was defined as the amount of enzyme that causes a 0.01 unit increase in the absorbance per minute.

Protein contents were calculated by means of bovine serum albumin (BSA) as a standard [40].

2.4. Statistical Analyses. All the experiments were executed in triplicate, rendering a completely randomized design (CRD) with a factorial layout. The factors were different irradiation doses and storage time. Analysis of variance (ANOVA) was employed to analyze the data, whereas the LSD test was used for the comparison of means (P < 0.05), using MINITAB 18.1 software.

#### 3. Results

3.1. Physicochemical Attributes. The effect of irradiation upon physicochemical attributes like TSS, pH, titratable acidity, and ripening index of guava fruits stored under ambient conditions was explained in Figure 1. There was an increasing trend of TSS values (Figure 1(a)) in relation to the storage period; however, the rate of increasing TSS values was significantly different with the irradiation dose. The data showed that the maximum TSS value (13.45%) was recorded in the control (T<sub>0</sub>, nonirradiated) samples on the 20<sup>th</sup> day of storage. The fruit samples that were treated with  $1.0 \text{ kGy} (T_5)$  showed the retaining behavior in terms of TSS values and showed an increase of 51.4% in comparison to control samples (65%). In contrast to TSS values, the percentages of titratable acidity (TA) decreased with each storage interval; however, the irradiation doses significantly affected the TA values (Figure 1(b)). It was also revealed that all the treatments were at par till the 4<sup>th</sup> day of storage, but a significant difference (P < 0.05) can be seen on the last day of the storage period especially for the treatments  $(T_5)$  and  $(T_0)$ . Figure 1(c) shows the pH values as affected by the irradiation doses, and it was depicted that there was an increasing trend in terms of pH values; however, in T<sub>5</sub>, the rate of increase was significantly lower (14.78%) than that of control samples (30.37%) at the end of storage period. The ratio of TSS to TA was expressed as a ripening index (RI) which resulted in similar increasing behavior (Figure 1(d)). The significantly lowest values of RI were recorded in T<sub>5</sub>, while the highest value of RI (61.22) was noted in control samples on the 20<sup>th</sup> day of storage.

3.2. Nutraceutical Attributes. The results pertaining to the effect of irradiation doses upon the nutraceutical attributes of guava fruit samples were shown in Figure 2. It was clearly

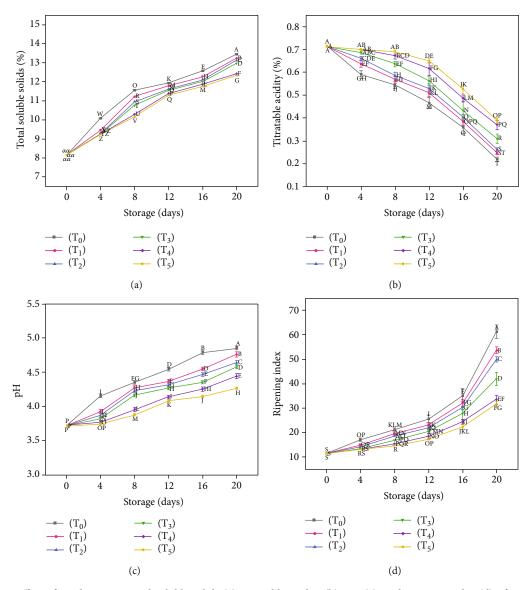


FIGURE 1: Effect of irradiation on total soluble solids (a), titratable acidity (b), pH (c), and ripening index (d) of guava fruit.

demonstrated that all the treatments varied significantly with each other in terms of radical scavenging activity (RSA), ascorbic acid, total phenolic contents (TPC), and total flavonoid contents (TFC).

It was observed that all nutraceutical parameters were decreased with the passing interval of the storage period; nevertheless, the irradiation doses considerably (P < 0.05) preserved the tested nutraceutical attributes to the end of storage time. The maximum decrease in RSA (85.75% to 35.49%) was recorded in untreated fruit samples, while T<sub>5</sub> showed a minimum decrease (85.75% to 48.70%) in antioxidant activity. The ascorbic acid (vitamin C) values dropped in all samples as well, but they did so at a significantly slower rate (P < 0.05) as the irradiation dose was increased (Figure 2(b)). In unirradiated samples (T<sub>0</sub>), the vitamin C contents significantly decreased at a considerably higher rate (64.42%); however, the rate of decreasing the ascorbic acid value was found to be much slower than that of control samples (41.97% and 34.67% in T<sub>4</sub> and T<sub>5</sub>, respectively) during

the whole span of storage. Likewise, total phenolic contents and total flavonoids also illustrated the same trend analogous to RSA and ascorbic acid values (Figures 2(c) and 2(d)). The maximum retention in both TPC (188.30 to 117.53 mg GAE/100 g) and TFC (143.76 to 109.10 mg QE/100 g) was recorded in  $T_5$  (1.0 kGy).

3.3. Fruit Firmness (N). The impact of  $\gamma$  -irradiation on the firmness of guava fruit is presented in Figure 3(a). Data analysis proved an overall significant difference (P < 0.05) in the mean values of treatment during storage. The maximum retention in firmness (49.34 N to 5.85 N) was observed in T<sub>5</sub>; however, the firmness retention rate was reduced with the decreasing irradiation dose.

3.4. Physiological Characteristics (Weight Loss, Respiration Rate, and Ethylene Rate). The graph cited in Figure 3(b) depicted the impact of irradiation on the physiological weight loss in fruit samples during the storage period. It

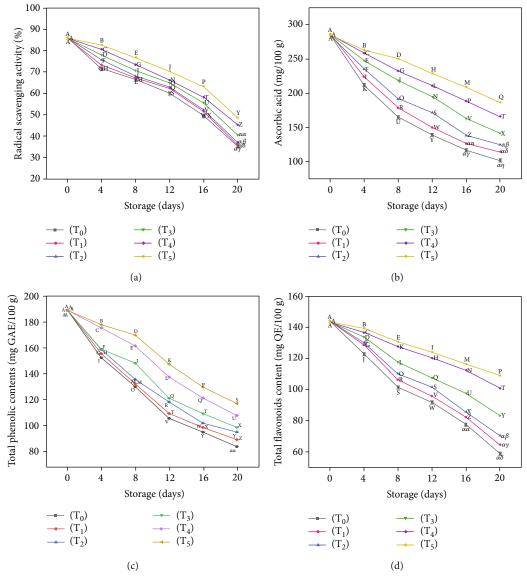


FIGURE 2: Effect of irradiation on nutraceutical attributes of guava fruit. Radical scavenging activity (a), ascorbic acid (b), total phenolic contents (c), and total flavonoid contents (d).

was noted that unirradiated fruit samples lost the maximum weight (23.49%), while significantly, the lowest loss in weight (10.88%) was recorded in fruit samples irradiated with 1.0 kGy (T<sub>5</sub>) at the end of the storage period (20<sup>th</sup> day).

The influence of irradiation dose upon the rate of respiration (CO<sub>2</sub>) and ethylene rate is shown in Figures 3(c) and 3(d), respectively. The data indicated that the irradiation dose at 1.0 kGy (T<sub>5</sub>) significantly reduced the respiration and ethylene production rates, thus resulting in delayed ripening. The respiration rate (CO<sub>2</sub>) and ethylene gas production were first increased until the attainment of the climacteric peak, and then these rates significantly decreased with each storage intervals. Data also indicated that the climacteric peak was attained on the 4<sup>th</sup> day of storage in all treatments except in T<sub>5</sub>, where the climacteric peak was significantly delayed to the 8<sup>th</sup> day of storage period.

3.5. Antioxidant Enzymes Kinetics. The impact of irradiation dose on the kinetics of the antioxidant enzymes in guava fruits during storage is shown in Figure 4. According to the revealed data, 1.0 kGy gamma irradiation dose (T<sub>5</sub>) had a highly significant (P < 0.05) impact on polyphenol oxidase (PPO), peroxidase (POD), superoxide dismutase (SOD), and catalase (CAT) activities as compared to nonirradiated fruit samples. It was observed that gamma irradiation significantly (P < 0.05) inhibited the activity of PPO  $(0.2144 \text{ Umg}^{-1} \text{ protein})$  compared to untreated samples  $(0.2965 \text{ Umg}^{-1} \text{ protein})$ . In contrast, the activities of POD enzymes were significantly (P < 0.05) enhanced under the influence of irradiation (Figure 4(b)) till the 12<sup>th</sup> day of storage and then considerably decreased. It was also found that the irradiation doses  $0.2 \text{ kGy} (T_1)$  and  $0.4 \text{ kGy} (T_2)$  had a nonsignificant impact on POD activity compared to control samples  $(T_0)$ . Similarly, Figure 4(c) shows SOD activity as

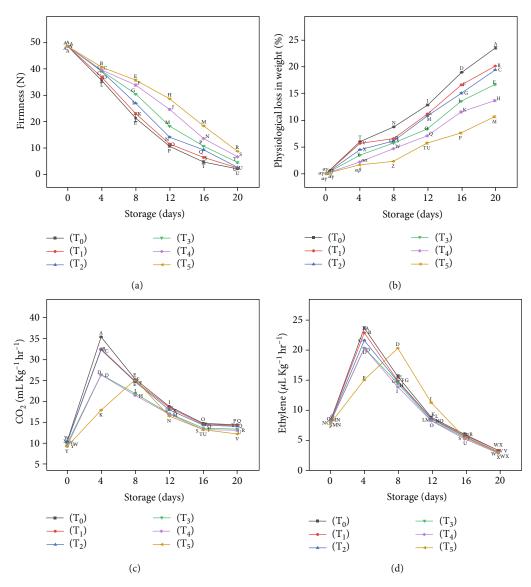


FIGURE 3: Effect of irradiation on firmness (a), physiological loss in weight (b), respiration (c), and ethylene (d) parameters of guava fruit.

affected by the gamma irradiation dosages; which unveiled a significant increase especially in the case of  $T_4$  and  $T_5$  compared to control ( $T_0$ ). The highest SOD activity (1672 U mg<sup>-1</sup> protein) was recorded for  $T_5$  on the 8<sup>th</sup> day of storage, and then it decreased gradually; however, at the completion of the storage period, all treatments disclosed significantly higher values of SOD enzyme activity compared to unirradiated samples. Likewise, the irradiation also significantly affected the catalase activity as compared to untreated samples. The activity of catalase enzyme (CAT) dropped down in response to the storage time, while the degree of decreasing CAT activity was significantly reduced under the impact of gamma irradiation treatments (Figure 4(d)).

#### 4. Discussion

The nonresidual feature of ionizing radiations is one of the most potential advantages of this intervention because it mitigates the deleterious residual effects of unsafe chemical substances that are used to extend the postharvest storage life of agricultural goods. Internationally, food irradiation has been recommended as an effective and safe way of management to escalate the postharvest of perishable commodities. In the present investigation, gamma irradiation significantly improved the majority of the storage-related quality parameters of guava fruits. Among the physicochemical attributes, total soluble solids were increased with the storage time; however, gamma irradiations inclined to reduce the rate of TSS increase (Figure 1).

The TSS levels rise as a result of the enzymatic breakdown of complex carbohydrates into simple sugars during maturation [41]. Gamma radiations at doses 0.8 to 1.0 kGy effectively reduce this conversion, thus delaying the ripening process. Similarly, the pH values were also increased with the attainment of fruit maturity, while the titratable acidity (TA) values had a tendency to decrease with the passage of the storage time. When pH shifts from acidic to basic, the activities of PPO and POD enzymes rise significantly,

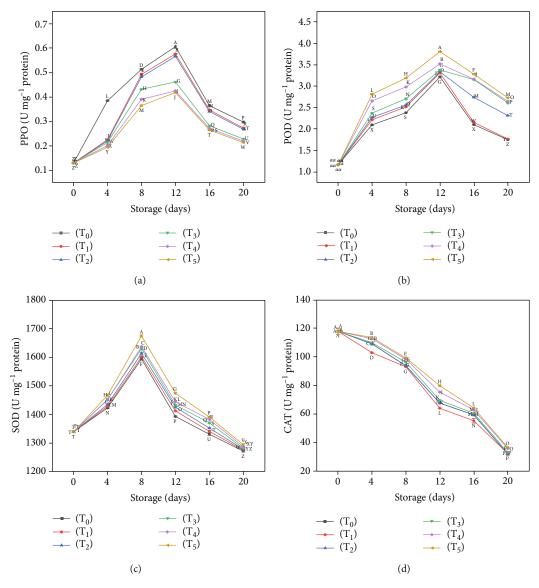


FIGURE 4: Effect of irradiation on enzyme kinetics of guava fruit. Polyphenol oxidase (a), peroxidase (b), superoxide dismutase (c), and catalase (d).

affecting the flavylium cation-to-carbonyl ratio of anthocyanin and rapidly change into colorless carbonyl compounds [42]. The climacteric fruits continue to mature even after harvest, and as a consequence, the starch is hydrolyzed to produce sugars and organic acids. While these organic acids are used as the main substrate for respiration, the titratable acidity of stored fruits decreases as a result [43]. The findings of the present investigation are closely in line with Majeed et al. [44], who noted that gamma irradiation doses up to 1.5 kGy may be successfully used to minimize postharvest decay of strawberries without significantly altering the fruit's pH and TA values. Among the physicochemical characteristics of fruits, the ripening index (RI), which is the ratio of TSS-to-TA, is a vital quality parameter to be studied during postharvest storage. Ripening index (RI) provides a better understanding for horticulturists and even for progressive growers regarding ideal picking time and the actual maturity stage of fruits as well. Furthermore, instead of measuring soluble solids and titratable acidity separately, the ratio provides a more accurate indication of the fruit's flavor [45]. In the present study, RI of guava fruits gradually increased in all the treatments with each passing storage interval; however, gamma irradiation imposed a positive effect on reducing the rate of RI augmentation (Figure 1). This is because the total soluble solids were nearly retained in the treated guava fruit samples, while the titratable acidity values were significantly dropped down till the final attainment of storage span in the current investigation. Rodriguez-Nunez et al. [46] also pointed out that the ripening index steadily amplified with the passage of storage time. The observations noted from the current study, regarding the physicochemical parameters, are also in line with the findings of Rosario et al. [47] and Barkaoui et al. [48].

Fruits are becoming a more regular part of daily diets due to the recent advancement and widespread dissemination of nutritional information among the people, as well

as the unique health benefits and secret nutraceutical prospects of fruits that go beyond the basic nutrition. Generally, during the storage studies, the nutraceutical attributes of fruits decrease with the passage of time; therefore, while selecting any postharvest intervention, the preservation of this dynamic feature of fruits should be taken into account [35]. In the current investigation, those fruits which were exposed to gamma irradiation, significantly retained their nutraceutical attributes. Vitamin C is considered as one of the most delicate vitamins which can be lost due to any undesirable action. That is why in postharvest storage studies, the estimation of vitamin C (ascorbic acid) has a significant impact in selecting the possible postharvest intervention. In the current investigation, as a result of gamma irradiation treatment to guava fruits, it was noted that the ascorbic acid showed the tendency to retain its values till the completion of storage span. The retaining behavior of ascorbic acid may be due to the conversion of L-ascorbic acid into dehydroascorbic acid by the effect of gamma irradiation [49]. That observation may lead to prolonging the postharvest storage life of guava fruits and is supported by the results of Rosario et al. [47]. Similarly, the retention of TPC and TFC by irradiation treatment would be the result of increased activity of a regulatory enzyme known as phenylalanine ammonia lyase (PAL) in the biosynthesis of phenolic compounds [50]. The retention trend of phenolic compounds may also be related to the release of smaller phenolic compounds (e.g., tannic acid and gallic acid) from the glycosidic linkage with the breakdown of complexes having higher molecular weights (e.g. tannins) as a result of gamma irradiation [51, 52]. It is well documented that the guava fruits especially the "Gola" cultivar has remarkable antioxidant activity [1]. However, during the postharvest storage of climacteric fruits like guava, the antioxidant activity (also known as radical scavenging activity/RSA) significantly decreases with the passage of time, and the same behavior was observed in the present study. Nonetheless, it is noteworthy that the RSA values had been significantly (P < 0.05) retained by gamma irradiation at dosage levels of 0.8 kGy to 1.0 kGy in comparison to the control. The reason behind that phenomenon may be hypothesized in such a way that with the result of gamma irradiation, CAT enzyme activity is enhanced due to which free radicals may be consumed and resultantly antioxidant activities retained [53]. The findings of our study are also in accordance with Rodriguez-Nunez et al. [46]; Pandey et al. [49]; Rosario et al. [47]; Razali et al. [41]; and Hassanein et al. [43]. It was also discovered in the present study that the firmness of those guava fruits tends to maintain itself when exposed to gamma irradiation. Usually, the firmness of under-storage fruits decreases due to the loosening of the cell wall and other enzymatic reactions; however, the gamma irradiation significantly (P < 0.05) slows down the loss of firmness. This could be because gamma irradiation modifies the ripening-induced enzymes including lyase, polygalacturonase, pectin methyl esterase, and rhamnogalacturonase, which are responsible for the synthesis of cell wall and thus possess the firmness of irradiated fruit [54]. The current research's findings regarding the firmness of guava fruits

stored under irradiation treatment were also in accordance with the investigations of Sau et al. [55] and Mendes et al. [56], who produced the similar results while studying the effect of low doses of gamma irradiation upon off-season guavas and cherry tomatoes, respectively. Data analysis for the current research work showed that all the fruit samples significantly lost weight during storage at ambient conditions till the 20<sup>th</sup> day. The maximum weight was lost in the nonirradiated fruit samples, while the minimum weight loss was observed in those guava fruit samples which were exposed to an irradiation dosage of 1.0 kGy. The reduced weight loss could be linked to the inhibitory effects of gamma irradiation on physiological processes like respiration and transpiration. Therefore, the reduction in weight loss would also aid in impeding the postharvest quality decline in guava fruits. An effective decrease in weight loss during storage was also found by different researchers while studying the impact of gamma irradiation upon a variety of fruits including; peaches [57], off-season guavas [55], strawberries [58], apple [27], and raspberries [59].

In the present study, the respiration and ethylene production rates were enhanced in all treatments until the fourth day of storage and then gradually declined. Guava is a climacteric fruit; therefore, its respiration and ethylene exchange rates increase during ripening and even after harvest [13]. It is worth to mention here that the irradiation dose at the rate of 1.0 kGy significantly delayed the climacteric peak (8<sup>th</sup> day instead of 4<sup>th</sup> day for other treatments) and recorded as the lowest rates of CO<sub>2</sub> and ethylene till the 20<sup>th</sup> day of storage. The suppression in respiration rate by irradiation treatment may be due to the consumption of oxygen during storage. Thus, we can assume that gamma irradiation may delay the ripening process and resultantly the senescence. Additionally, the slower rates of ethylene hormone may be attributed to the inactivation of ethylene biosynthetic enzymes like acetyl-CoA carboxylase (ACC) synthase and ACC oxidase [60]. The findings of the current study are in close agreement with Gunes et al. [61] and Jat et al. [54], who studied the influence of ionizing radiations upon strawberries and fresh-cut apple slices, respectively.

According to the findings of the current study, it was exposed that gamma irradiation had a sizable impact on the kinetics of antioxidant enzymes. The impact of gamma irradiation on the activities of polyphenol oxidase enzyme (PPO) in guava fruits unveiled that irradiation inhibited PPO activity significantly (P < 0.05). At doses above 0.6 kGy, gamma irradiation had a significant impact upon PPO activity inhibition. The irradiation-induced suppression of PPO activities has also been found to be substantially advantageous in retaining the elevated values of total phenols in guava fruits, predominantly for those irradiated at 1.0 kGy [53]. A possible cause for lower PPO enzyme activity could be the reduced membrane reassortment which frequently results in a complex of phenolic compounds with antioxidant enzymes [35]. The inhibition of PPO also resulted in nonbrowning pigmentation of guava fruits exposed to gamma irradiation [53]. In the current study, irradiated guava fruits exhibited higher activities of POD, SOD, and CAT enzymes. High levels of superoxide radical

and H<sub>2</sub>O<sub>2</sub> would be generated as a result of reserve stress brought on by irradiation to fruits, boosting their selfimmune mechanisms and thus enhancing POD enzyme activities [62]. Due to this mechanism, the treated guava fruit samples were less prone to postharvest stress. Superoxide dismutase and catalase are also known as antioxidant detoxification enzymes that play critical roles in the scavenging of reactive oxygen species (ROS). As the name indicates, SOD enzyme is accountable for the dismutation (oxidation and reduction simultaneously) of superoxide (O<sup>-2</sup>) free radicals to O2 and H2O2, hence protecting the cells from superoxide-induced radical damage [63]. Increased SOD activity in irradiated guava fruits may offer additional resistance against oxidative destruction prompted by O<sup>-2</sup> radicals. On the other hand, enhanced CAT enzyme activity plays a key role in the defense mechanism against hydrogen peroxide  $(H_2O_2)$  damage, as the CAT enzyme is responsible for the accelerated breakdown of H2O2 into oxygen and water molecules [63]. In the current research experiment, CAT activity showed a declining trend due to the general ripening process after harvest. However, irradiation doses had a significant (P < 0.05) effect upon maintaining the higher values of CAT enzymes, particularly in the case of irradiation dosage at 1.0 kGy as compared to nonirradiated fruit samples in which CAT activity declined rapidly. Higher CAT activity in irradiated fruit samples indicates that the cells have better capacity to scavenge  $H_2O_2$ . These outcomes are in line with earlier research conducted on blueberry fruit, which disclosed elevated SOD, POD, and CAT activities after 2.5 kGy gamma radiation treatment [64]. The results of our study are also consistent with those of Hussain et al. [65] and Zarbakhsh and Rastegar [66].

#### 5. Conclusion

Without a doubt, the guava fruit is incredibly healthy and rich in nutraceutical components. Therefore, it is imperative to reduce postharvest losses (accounted 30-50% for guava fruits annually) in order to retain this type of nutrient-dense fruit. Since gamma irradiation has shown favorable results for the majority of the storage-related quality indicators, the findings of the current study justify its use for postharvest storage of guava fruit. Most importantly, the use of gamma irradiation (1.0 kGy for 28.33 minutes) substantially ( $P \le 0.05$ ) postponed the climacteric peak to the 8<sup>th</sup> day as opposed to the 4<sup>th</sup> day for other treatments and tended to preserve the nutraceutical potential of guava fruits up to the storage of 20 days. The current investigation additionally exhibited that the PPO enzymes' activity was considerably  $(P \le 0.05)$  decreased (0.2965 Umg-1 protein to)0.2144 U mg-1 protein), which subsequently reduced fruit browning. I may be, therefore, concluded that gamma irradiation can be effectively applied as a better postharvest intervention to maintain the storage stability of guava fruit and to improve its shelf life up to 20 days. We proposed using ionizing radiation in conjunction with other ecologically friendly techniques to increase the shelf life of guava and other tropical fruits after harvest.

#### **Data Availability**

The data used to support the findings of this study will be available upon request.

#### **Conflicts of Interest**

The authors have no conflict of interests regarding the current research work.

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