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Effect of Gamma Radiation and Storage Time on the Microbial and Physicochemical Properties of Dried Byadgi Chili (*Capsicum annuum*)

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Abstract: The effects of gamma radiation up to 10 kGy and storage time (0 to 16 weeks) on microbial and physicochemical properties of dried Byadgi chili were investigated. After 2.5, 5.0, 7.5, and 10.0 kGy of radiation, the samples were kept at room temperature with the control sample (0 kGy) and analyzed at 0, 4, 8, 12, and 16 weeks. Microbial aspects including total aerobic count (TAC), *E. Coli*, coliform, yeast, and mold were tested, as were physicochemical characteristics such as moisture, water activity, surface color (L*, a*, b*, chroma, and hue), aflatoxin, capsaicin, and dihydrocapsaicin. As the radiation dose and storage period were increased, water activity rose in lockstep with moisture. Gamma radiation had no effect on the color of the surface up to 10 kGy, although it did not aid with the retention of the initial color value during storage. Mold growth was observed in the control, 2.5, 5.0, and 7.5 kGy radiated samples, but no aflatoxin was found. The increase in radiation dose and storage time had a negative effect on capsaicin and dihydrocapsaicin. TAC was also observed at the end of the storage time, despite radiation at 10.0 kGy successfully sterilizing dried Byadgi chili. The principal component analysis (PCA) biplot revealed two distinct groupings based on duration of storage times. Radiation at 10.0 kGy had an excellent response to the microbial profile, with only minimal changes in quality attributes after eight weeks of storage.

Keywords: capsaicin; dihydrocapsaicin; principal component analysis (PCA); high performance liquid chromatography (HLPC)

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Citation: Balakrishnan, N.; Yusop, S.M.; Rahman, I.A.; Dauqan, E.; Abdullah, A. Effect of Gamma Radiation and Storage Time on the Microbial and Physicochemical Properties of Dried Byadgi Chili (*Capsicum annuum*). *Agriculture* **2022**, *12*, 639. https://doi.org/10.3390/agriculture12050639

Academic Editor: Quan-Sheng Chen

Received: 16 March 2022
Accepted: 25 April 2022
Published: 28 April 2022

1. Introduction

Dry chili (*Capsicum annuum* L.) is an essential ingredient in Malaysian cuisine for making true spicy meals, such as sambal, curry, and other local foods such as '*satay*' sauce, '*rendang*', and '*asam pedas*'. Aside from spiciness, dried chili adds value to a dish by adding a brilliant red natural color and flavor [1]. The top listed countries that produce dried chilies around the globe are India, China, Spain, Mexico, Pakistan, Morocco, and Turkey [2]. Since, in Malaysia, only fresh chilies are commercially available, the domestic market and spices industries are solely depending on imported dried chilies for their usage. India and China are the two major countries exporting dried chili to Malaysia, and the varieties supplied from these countries are widely used for domestic consumption and food manufacturing. Among the varieties, Byadgi dried chili originating from Karnataka is well accepted by locals for its mild pungency and intense red color. For domestic use, consumers choose dried chili based on pungency level and appearance, whilst for commercial purposes, more parameters are given importance, such as color, microbial contamination, and aflatoxin level.

Similar to other spices, dried chilies are cultivated and harvested in warm and humid climate, allowing for the growth of a wide range of microorganisms, which can occur at many phases of spice processing, such as cultivation and harvesting. As a result, they are widely known as major food pollutants [3–6]. Fresh chilies are harvested and dried on the ground or tar layered roads, and are almost always contaminated by microorganisms from the soil and windblown dust and by bird droppings. The presence of indigenous microorganisms from plants, an unsanitary food processing area, air, dust, a polluted water source/irrigation with the presence of human/animal excreta, and improper pre- and post-harvest handling during processing, storage, and distribution are all possible microbial contaminations [7].

Another possible health risk caused by dried chili is the toxicity from aflatoxin contamination. Aflatoxins are type of dangerous mycotoxins produced by some strains of *Aspergillus flavus*, *Aspergillus parasiticus*, and *Aspergillus nomius* molds [8], and are extremely carcinogenic to both humans and animals [9]. The most potent aflatoxins found in dried chilies are B1, B2, G1, and G2 [10]. The occurrence and contamination of aflatoxins are widely known in spices, especially in dried chilies and dried paprika. The contamination and its level depend on the type of process, cropland, geographical condition, and climate throughout the year [11–13]. The highest levels are usually found in commodities from warmer regions of the world, where there is a great deal of climatic variation [14,15]. Research has shown that aflatoxin contaminations are higher than the regulated levels allow in dried chili. This can be due to a hot and humid climate in the producing country, poor hygienic conditions during harvesting and processing, mold growth, and poor storage conditions [16]. It was also proven that aflatoxin contamination in dried chili may happen in cold-room storage [17]. Pre-harvest aflatoxin contamination is favored by drought stress, and the post-harvest aflatoxin contamination is favored by raw material handling, especially during rainy season [18]. The hot and humid climate with extended drying periods, improper drying, and inadequate knowledge on handling the dried chilies are the major causes for quality deterioration in dried chilies, which eventually leads to aflatoxin formation [12]. Aflatoxins are able to resist extreme conditions, such as heating at high temperature and freezing [19].

Hygiene or sterilization treatments may help to reduce potential health risks caused by dried chili. Food radiation is a type of physical treatment that uses ionizing radiation to stop the growth of unwanted biological organisms or lower their population [20], and it is one of the more modern food preservation treatments. International institutions such as the World Health Organization (WHO), the United Nations Food and Agriculture Organization (FAO), the International Atomic Energy Agency (IAEA), and Codex Alimentarius have all recognized the technology’s safety. Food radiation has shown promise in extending the shelf life of dried chili, while also preserving its organoleptic and safety features. The initial amount of contamination and the type of bacteria present in dried chili dictates the radiation dose required to disinfect it. Molds, fungus, and coliforms are generally removed at lower dose levels in comparison to total bacteria [21]. Gamma irradiation is recognized as cold sterilization, since it only delivers a small amount of heat or no heat at all to the food item being radiated [22], thus controlled or a reduced amount of gamma irradiation kills microorganisms in dried chili successfully without compromising to its color, flavor, or desired characteristics. The goal of this study was to evaluate the effectiveness of gamma radiation and its storage quality on the basic quality attributes of dried Byadgi chili sold in Malaysian markets.

2. Materials and Methods

2.1. Sample Collection

Dried Byadgi (*Capsicum annuum* L.) chili samples were collected from one of the local leading spices manufacturers. Approximately 2.2 kg of the sample was collected in a polyethylene bag, and sent to the laboratory for analysis. The sample was divided and packed in to 22 packets with 100 g in each, labelled, and sealed tightly before gamma irradiation treatment at Nuclear Laboratory, The National University of Malaysia, UKM, Bangi. A total of four packets (400 g) were radiated at each radiation dose (2.5, 5.0, 7.5, and 10.0 kGy) with cobalt-60 source at ambient temperature using a Gamma Cell 220 Excel irradiator at a dose rate of 2.1 kGy/h. On balance, six packets were kept without gamma irradiation to serve as a control sample (0 kGy). All of the radiated and non-radiated (control) samples were kept at room temperature (25 °C to 35 °C) with a relative humidity of 80% to 85%.

2.2. Analysis of Moisture and Water Activity

The dried Byadgi chili sample was cut into a piece measuring 1 cm in length for moisture and water activity analysis. The moisture content was analyzed referring to AOAC method 925.10, and water activity (a_w) was analyzed using the dew point or chilled mirror method with Aqua Lab 4TE by Decagon, (METER Group, Inc., Pullman, WA, USA) [23]. The results were reported as an average of the triplicate readings.

2.3. Surface Color Measurement

Dried Byadgi chili was cut into pieces of 1–2 cm, and the seeds were removed. The pieces were well arranged to minimize gaps in between them until the cup was full. The samples were measured for surface color properties with CIE L*a*b* color scale using a Hunterlab colorimetric spectrophotometer. The Hunterlab ColorFlex system was used for color characterization, calibrated to a standard green tile (L* = 52.89, a* = −26.98, b* = 12.02). The samples were tested before they were stored at an ambient temperature, and the results were recorded as ‘0 week’ data, considered as the initial data for the study. Overall, at each storage time point, a total of 15 samples were analyzed in triplicate, and the results were reported as the average of the triplicate readings. With L*, a*, and b* color parameters obtained, h* and C* were calculated using the following equations [1]:

$$h_{ab}^* = \tan^{-1}(b^*/a^*) \tag{1}$$

$$C_{ab}^* = (a^{*2} + b^{*2})^{1/2} \tag{2}$$

2.4. Determination of Aflatoxin B1, B2, G1, and G2

Aflatoxins B1, B2, G1, and G2 in the samples were tested according to the method recommended by Vicam [24], using the immunoaffinity column (IAC) cleaning procedure. The determination of aflatoxin presence was made using HPLC with an Agilent 1260 Infinity Quaternary Pump equipped with an Agilent 1260 Infinity Fluorescence Detector (G1312B), with an excitation wavelength of 365 nm and an emission wavelength of 440 nm, reverse phase C18 (4.6 mm × 50 mm, 2.7 µm) Poroshell column from Agilent Technologies and a ‘Phred’ Photochemical Derivatization Reactor from Aura Industries, USA. The column temperature was maintained at 30 °C. The mobile phase used was water-methanol (65:35, *v*/*v*), and 30 µL volume of sample and standards were injected into an HPLC. The separations ran at a flow rate of 1.2 mL/min. The samples were injected in triplicate. Agilent OpenLAB CDS ChemStation software was used to control the system and process signals.

2.5. Determination of Capsaicin and Dihydrocapsaicin

The sonication method was applied for capsaicin and dihydrocapsaicin extraction, as described by [25]. A total of 100 g dried Byadgi sample was powdered using a blender (Oster 10 speed household blender, Neosho, MO, USA), and sieved using 1 mm sieve to produce a homogeneous particle size. One gram of powdered sample was weighed in to 120 mL glass bottle lined with Teflon lids and mixed with 25 mL methanol. The bottles were sonicated for 20 min at 50 °C with an Elmasonic sonicator bath (model S30H, Singen, Germany). The bottles were removed from the sonicator, and cooled to room temperature. The supernatant was filtered into HPLC vials with 0.45 µm nylon syringe filter, caped, and analyzed within 24 h. The extraction was performed in triplicate.

Capsaicin and dihydrocapsaicin separation was performed with an Agilent HPLC instrument equipped with an Agilent 1260 Diode Array Detector set at 280 nm excitation wavelength and 4 nm band width. Agilent OpenLAB CDS ChemStation software was used to control the system and process signals. Chromatographic separation was performed with Zorbex Eclipse XDB-C18 (4.6 × 50 mm, 3.5 µm) Rapid Resolution column, and the column temperature was maintained at 30 °C. Then, 5 µL of filtrate was injected into the HPLC for capsaicin and dihydrocapsaicin quantification. The mobile phase used was water-acetonitrile (52:48, *v*/*v*), working at 1.0 mL per minute flow rate.

2.6. Microbiological Analysis

Total aerobic count (TAC), coliforms, *Escherichia coli*, yeast, and mold were analyzed using commercially available 3M™ Petrifilm™ plates (3M Microbiology Products, St. Paul, MN, USA), following the manufacturer's instructions. For TAC, the average number of red-colored colonies developed on the 3M™ Petrifilm™ were counted and reported as log cfu/g after multiplying by the dilution factor. Using the same protocol, coliforms, *Escherichia coli*, yeast, and mold counts were measured. The visible, red-colored colonies surrounded by trapped gas indicated coliform bacteria, while *E. coli* appeared as blue-colored colonies with trapped gas on the 3M™ Petrifilm™. Yeast and mold colonies that were developed from 3 to 5 days were calculated. Yeast colonies appeared raised, and were pink-tan to blue-green in color, while mold colonies had multiple colors with diffused age.

2.7. Statistical Analysis

All analysis were performed in triplicate, and the data measured were expressed as means ± standard deviations. Statistical analysis of data was performed using one-way analysis of variance (ANOVA) and a GLM (general linear model) procedure for two-way ANOVA of the SPSS software version 22.0 (IBM Corp., Armonk, NY, USA). The dependent variables TAC, coliform, *E. coli*, yeast, mold, capsaicin, and dihydrocapsaicin were analyzed using one-way ANOVA to identify the significant difference between the two factors individually, 'storage time' and 'gamma irradiation dose'. Meanwhile, with two-way ANOVA, the two factors were evaluated simultaneously to identify significant interaction (storage time x gamma irradiation), and were compared using a Duncan multiple comparison test. A PCA plot was constructed after creating correlation matrix using the data set of 25 observation points (5 samples with different radiation doses × 5 storage intervals) and their corresponding 12 attributes at each storage time. Principal component analysis (PCA) was performed with XLSTAT 2019 software to explore the relationship between the effect of different gamma radiation doses and different storage times corresponding to physical quality attributes (moisture and water activity), surface color attributes (lightness—L*, redness—a*, yellowness—b*, C*—chroma and h*—hue), microbial quality (total aerobic count—TAC, yeast and mold), and pungency components (capsaicin and dihydrocapsaicin) for radiated and control dried Byadgi chilies samples collected.

3. Results and Discussion

3.1. Moisture and Water Activity Content

The moisture content slightly increased for radiated samples compared to the control sample (Table 1). The moisture content in the control sample was 14.87%, and the highest moisture content was observed for 7.5 kGy at 15.66%. At 7.5 kGy and 10.0 kGy, the increase in moisture was 5.3% and 3.09%, respectively. Upon storage, the moisture content increased significantly. After 16 weeks in storage, the increase in moisture for the control, 7.5, and

10.0 kGy radiations were 18.36%, 10.22%, and 9.55%, respectively. The control sample showed a drastic increase in moisture. A similar finding was reported by [26], and could be due to high relative humidity storage condition. The observation from this study shows that radiation did not stop moisture migration in dried chili. However, moisture increases in radiated samples were much lower than the control sample. Overall, 10.0 kGy radiated samples showed much more stable moisture increases compared to other radiated and control samples.

Table 1. Mean (n = 3) values for moisture content (%) and water activity (aw) for irradiated dried Byadgi chili at different irradiation doses (0, 2.5, 5.0, 7.5, and 10.0 kGy) and storage periods (0, 4, 8, 12, and 16 weeks).

Storage (Weeks)		0 kGy	2.5 kGy	5.0 kGy	7.5 kGy	10.0 kGy
0	Moisture	14.87 ± 0.45 ^{cB}	15.31 ± 0.02 ^{dAB}	15.54 ± 0.08 ^{eA}	15.66 ± 0.35 ^{cA}	15.33 ± 0.14 ^{cAB}
	Water activity	0.6047 ± 0.0026 ^{dE}	0.6163 ± 0.0005 ^{eD}	0.6241 ± 0.0009 ^{eC}	0.6276 ± 0.0007 ^{cB}	0.6307 ± 0.0011 ^{bA}
4	Moisture	16.24 ± 0.05 ^{bA}	15.76 ± 0.3 ^{cB}	15.97 ± 0.05 ^{dAB}	16.16 ± 0.02 ^{bA}	15.72 ± 0.2 ^{bB}
	Water activity	0.6571 ± 0.0018 ^{bA}	0.6293 ± 0.0004 ^{dD}	0.6317 ± 0.0015 ^{dD}	0.6495 ± 0.0021 ^{bB}	0.6430 ± 0.0009 ^{cC}
8	Moisture	16.27 ± 0.13 ^{bA}	16.24 ± 0.06 ^{bA}	16.17 ± 0.09 ^{cAB}	16.00 ± 0.16 ^{bcBC}	15.90 ± 0.09 ^{bC}
	Water activity	0.6505 ± 0.0004 ^{cA}	0.6502 ± 0.0010 ^{cAB}	0.6477 ± 0.0016 ^{cC}	0.6480 ± 0.0012 ^{bBC}	0.6309 ± 0.0016 ^{bD}
12	Moisture	16.37 ± 0.17 ^{bB}	16.38 ± 0.1 ^{bB}	16.71 ± 0.05 ^{aA}	16.72 ± 0.05 ^{aA}	16.41 ± 0.12 ^{aB}
	Water activity	0.6562 ± 0.0065 ^{bcA}	0.6589 ± 0.0053 ^{bA}	0.6586 ± 0.0006 ^{aA}	0.6560 ± 0.0015 ^{aA}	0.6595 ± 0.0008 ^{aA}
16	Moisture	17.60 ± 0.05 ^{aA}	16.74 ± 0.08 ^{aB}	16.34 ± 0.14 ^{bC}	16.39 ± 0.25 ^{abC}	16.29 ± 0.17 ^{aC}
	Water activity	0.6684 ± 0.0008 ^{aA}	0.6647 ± 0.0012 ^{aB}	0.6562 ± 0.0007 ^{bC}	0.6561 ± 0.0014 ^{aC}	0.6547 ± 0.0011 ^{bC}

Means within a column followed by different small letters ($^{a–e}$) are significantly different ($p < 0.05$), Means within a row followed by different capital letters ($^{A–E}$) are significantly different ($p < 0.05$).

In one of the studies, Ref. [27] compared the effects of steaming and irradiation at 10 kGy on red pepper powder stored at ambient (20 ± 2 °C) and refrigerated (4 ± 2 °C) temperatures. The moisture content reduced to 9.04% in comparison to an initial level of 10% after being radiated at 10 kGy. The moisture content of both control and treated samples reduced further after six months of storage, with the decrease being larger in samples stored at an ambient temperature (7.75% (control), 8.47% (steamed), and 8.74% (radiated), respectively). The author explained that radiation induces significant changes in cellular integrity through the depolymerization of polysaccharides, which leads to softening and easy water release in foods at ambient temperature. However, this observation is in contrast to the findings reported in our study.

In another study, Muhammad et al. (2009) [18] evaluated effect of radiation on aflatoxin-contaminated chilies collected from a local market. The samples were packed in HDEP bags, and irradiated at 2.0, 4.0, and 6.0 kGy with a cobalt 60-gamma irradiator at a dose rate 0.4461 kGy/h. The irradiated and control samples were stored for 90 days at room temperature with a relative humidity of 45–60%. The moisture content before and after 6.0 kGy radiation was 11.41% and 11.39%, respectively. Overall, the moisture content of the samples reacted insignificantly to the administered radiation doses and storage up to 90 days, in a way which contradicted to our findings.

Similar findings were observed by [28], when dried hot peppers were packed and irradiated at 2.0, 4.0, and 6 kGy in a Co^{60} gamma irradiator. The radiated samples were kept at 25 °C for 90 days with a control sample, and examined for mycological, aflatoxins, and moisture. The initial moisture content of the control sample ranged from 11.96–12.52%, and it dropped to 11.87–12.46% after radiation at 6 kGy. In radiated samples, the researcher noticed an average moisture loss of 1%, which contradicts the conclusions of our study.

In correspondence with moisture, water activity rose as the radiation dose and storage period increased. The initial water activity before radiation was 0.6047 ± 0.0026, and at 10.0 kGy the water activity increased to 0.6307 ± 0.0011. Upon storage, the highest value for water activity was observed in the control sample at 16 weeks, 0.6684 ± 0.0008. Water activity for radiated samples were observed to be much more stable than in the control sample. In general, decreased moisture content and lower water activity contribute to storage stability of dried chilis. As a result of diminished free radical generation, this situation has a significantly weaker effect against the fatal effect of ionizing radiation [28]. In contrast to other quality indicators in dried chilies, such as microbial load, color, nutritional value, and spiciness, the effect of radiation on moisture and water activity in dried chilies has not been substantially researched.

3.2. Effect on Surface Color (L, a*, b*, C* and h*)*

The presence of carotenoid pigments gives the dried chili its deep red color. Carotenoids, such as capsanthin and capsorubin, are the main components responsible for this red color. Carotenoids are thermally sensitive in nature, and could easily disappear at room temperature [29]. In this study, it was observed that radiation did not affect the surface color of dried Byadgi chilis. However, surface color was greatly affected as the radiation dose and storage time was increased. Chatterjee et al. (1998) [30] reported insignificant ($p < 0.05$) differences in color coordinates for radiated chilies, but upon storage, significant ($p < 0.05$) color reduction was observed. In our study, the L* value in control sample was 24.16 ± 1.01. Upon storage, a slight decrease in L* was observed. This is an indication that the sample was losing its initial brightness value, which is attributed to a decrease in bright pigment concentration. Following storage for 16 weeks, radiation at 10.0 kGy showed the highest loss in L* value. Overall, lower radiation doses showed better L* values, compared to the higher radiation doses (Table 2).

Table 2. Effect of gamma radiation on L* (lightness), a* (redness), and b* (yellowness) for dried Byadgi chili.

Storage (Weeks)	Parameters	0 kGy	2.5 kGy	5.0 kGy	7.5 kGy	10.0 kGy
0	L*	24.16 ± 1.01^{Aa}	24.11 ± 0.47^{Aa}	24.24 ± 0.3^{Aa}	23.50 ± 0.46^{Aa}	23.91 ± 1.35^{Aa}
	a*	20.85 ± 0.69^{Aa}	18.04 ± 0.13^{Ca}	19.18 ± 0.34^{Ba}	16.54 ± 0.10^{Dab}	16.76 ± 0.06^{Da}
	b*	9.85 ± 0.05^{Ba}	9.03 ± 0.09^{Ca}	10.43 ± 0.07^{Aab}	8.01 ± 0.13^{Eb}	8.40 ± 0.02^{Db}
4	L*	22.87 ± 0.09^{Ab}	21.91 ± 0.67^{BCb}	22.23 ± 0.34^{ABb}	21.21 ± 0.09^{Cb}	21.92 ± 0.45^{BCb}
	a*	16.16 ± 0.12^{Cc}	13.97 ± 0.06^{Ec}	18.00 ± 0.21^{Ab}	15.39 ± 0.10^{Db}	16.69 ± 0.08^{Ba}
	b*	8.63 ± 0.11^{Cb}	9.17 ± 0.06^{Ba}	9.64 ± 0.06^{Ab}	8.24 ± 0.10^{Db}	9.76 ± 0.26^{Aa}
8	L*	22.71 ± 0.04^{Ab}	20.24 ± 0.23^{Cb}	20.39 ± 0.07^{Cc}	19.55 ± 0.22^{Dc}	21.67 ± 0.12^{Bb}
	a*	17.82 ± 0.13^{Ab}	15.84 ± 0.20^{Db}	16.26 ± 0.05^{Cc}	16.97 ± 0.08^{Ba}	17.04 ± 0.19^{Ba}
	b*	8.51 ± 0.11^{Bb}	8.05 ± 0.13^{Cb}	7.10 ± 0.11^{Dc}	9.62 ± 0.12^{Aa}	8.20 ± 0.07^{Cb}
12	L*	21.59 ± 0.25^{Dc}	21.96 ± 0.33^{Cb}	19.74 ± 0.09^{Ed}	23.20 ± 0.08^{Aa}	22.38 ± 0.02^{Bb}
	a*	12.44 ± 0.78^{Bd}	15.70 ± 0.41^{Ab}	12.89 ± 0.98^{Bd}	13.16 ± 0.27^{Bc}	12.89 ± 0.46^{Bb}
	b*	5.22 ± 0.41^{Cd}	7.56 ± 0.80^{Ab}	6.83 ± 0.69^{ABc}	6.34 ± 0.17^{BCc}	6.61 ± 0.77^{ABc}
16	L*	21.34 ± 0.06^{Bc}	22.50 ± 0.57^{Ab}	22.58 ± 0.24^{Ab}	21.43 ± 0.50^{Bb}	21.17 ± 0.59^{Bb}
	a*	11.40 ± 0.84^{ABd}	13.11 ± 0.43^{Ad}	11.24 ± 0.93^{Be}	10.99 ± 1.63^{Bd}	10.88 ± 0.35^{Bc}
	b*	6.76 ± 1.41^{CDc}	7.79 ± 0.25^{BCb}	10.82 ± 0.87^{Aa}	8.37 ± 0.55^{Bb}	5.47 ± 0.34^{Dd}

Means within a column followed by different small letters ($^{a–e}$) are significantly different ($p < 0.05$), Means within a row followed by different capital letters ($^{A–E}$) are significantly different ($p < 0.05$).

In contrast to our findings, Kispeter et al. (2003) [31], in their research on paprika powder radiation, have highlighted that, generally, there was an increase in L* value upon radiation up to 5.0 kGy, and the significant difference was within a unit that was not detectable by the human eye. The author observed no further changes above 5.0 kGy. In contrast, Jose et al. (2000) [32] reported no changes observed in L* values for the samples radiated at 10.0 and 12.5 kGy. According to their findings, radiation doses less than 5.0 kGy did not cause any obvious effects on the surface color. Similarly, Lee et al. (2004) [33] observed no changes in L* values after radiation up to 7.0 kGy, and the author concluded that irradiation at 7 kGy did not change the essential quality of red color in the samples evaluated.

The redness (a*) value for the radiated dried Byadgi chili showed an inverse trend towards an increase in gamma radiation (Table 2). Upon storage for up to 16 weeks, the a* value dropped from the initial value of 20.85 ± 0.69 to 11.40 ± 0.84, 10.99 ± 1.63, and

10.88 ± 0.35 for the control, 7.5, and 10.0 kGy samples, respectively. A tremendous decrease was observed for the control sample. Radiation below 5.0 kGy showed better a* values in comparison with 7.5 kGy and 10.0 kGy. Similar findings were reported by [32], where, in their study, radiation at 10 and 12.5 kGy decreased the a* value in paprika powder. In contrast to this, Lee et al. (2004) [33] observed no changes in a* value for pepper powders radiated at 10 kGy and 7 kGy.

Beta-carotene, zeaxanthin, violaxanthin and beta-cryptoxanthin are the compounds responsible for the yellow-orange color found in chili carotenoids. In this research, the initial yellowness value, b*, in the control sample was 9.85. Upon radiation, b* did not show a proper trend towards an increase in radiation dose. A slight increase was observed at 5.0 kGy, and an obvious decrease was observed at 7.5 and 10.0 kGy (Table 2). Radiation at 2.5 kGy showed only a slight decrease compared to the control sample. Storage for up to 16 weeks generally showed a tremendous decreasing trend, except at 5.0 kGy. This indicates that radiation at 10.0 kGy did not help in retaining the carotenoids in dried Byadgi chili.

The color saturation index, or C*, and h* give more information about the spatial distribution of colors in radiated and non-radiated samples. As the radiation dose was increased, the C* value decreased (Table 3). This may have been caused by some biochemical process that contributes to color changes. The effect was also evident from the increase in h* measurement (Table 3), which could be due to creation of colored compounds. It appears that as the storage time was increased, a reduction in color quality properties was obvious. This could be well associated with the Maillard reaction or another non-enzymatic browning reaction. It was reported that the non-enzymatic browning in dried red pepper was due to reactions between reducing sugars and amino acids in pericarp. The other possible reason could be enzymatic oxidation of some phenolic compounds present in the red dried chili. This is because all of the study samples were packed under normal conditions, and the availability of oxygen in the packet could have stimulated an ongoing carotenoid oxidation in the radiated and non-radiated samples. The other possible reason for non-enzymatic browning could be due to increase in water activity. The rate and extent of non-enzymatic browning is influenced by water activity values. Minimum browning can be achieved when dried chili is stored below 0.3 water activity level [34].

Table 3. Effect of gamma radiation on C* (chroma) and h* (hue) for dried Byadgi chili.

Storage (Weeks)	Parameters	0 kGy	2.5 kGy	5.0 kGy	7.5 kGy	10.0 kGy
0	C*	22.69 ± 0.17 ^{Aa}	20.17 ± 0.03 ^{Ba}	19.60 ± 0.05 ^{Ca}	17.55 ± 0.03 ^{Ea}	18.91 ± 0.10 ^{Db}
	h*	25.85 ± 0.07 ^{Ca}	26.31 ± 0.05 ^{Bb}	26.74 ± 0.22 ^{Aa}	25.62 ± 0.23 ^{Cd}	26.15 ± 0.06 ^{Bc}
4	C*	18.41 ± 0.40 ^{Bb}	15.42 ± 0.10 ^{Cc}	20.49 ± 1.03 ^{Aa}	17.72 ± 0.20 ^{Ba}	19.78 ± 0.39 ^{Aa}
	h*	26.25 ± 1.00 ^{CDa}	25.93 ± 0.41 ^{Db}	28.20 ± 0.41 ^{Ba}	27.21 ± 0.18 ^{BCc}	30.61 ± 0.43 ^{Aa}
8	C*	15.91 ± 0.10 ^{Bc}	13.83 ± 0.11 ^{De}	14.86 ± 0.14 ^{Cb}	17.27 ± 0.11 ^{Aa}	16.04 ± 0.25 ^{Bc}
	h*	26.63 ± 1.35 ^{Ba}	26.47 ± 0.69 ^{Bb}	26.82 ± 2.39 ^{Ba}	29.35 ± 0.25 ^{Ab}	27.63 ± 0.68 ^{ABb}
12	C*	13.50 ± 0.88 ^{Bd}	17.15 ± 0.19 ^{Ab}	11.98 ± 1.30 ^{Cc}	14.61 ± 0.28 ^{Bb}	14.43 ± 0.11 ^{Bd}
	h*	22.73 ± 0.32 ^{Db}	22.55 ± 0.50 ^{Dc}	27.69 ± 0.47 ^{Ba}	25.94 ± 0.36 ^{Cd}	30.98 ± 0.14 ^{Aa}
16	C*	12.83 ± 0.73 ^{Bd}	14.36 ± 0.59 ^{Ad}	14.77 ± 0.85 ^{Ab}	14.74 ± 0.58 ^{Ab}	12.23 ± 0.35 ^{Be}
	h*	25.92 ± 0.33 ^{Ca}	29.70 ± 0.43 ^{ABa}	28.24 ± 1.45 ^{Ba}	31.09 ± 0.66 ^{Aa}	26.57 ± 1.10 ^{Cbc}

Means within a column followed by different small letters ($^{a–e}$) are significantly different ($p < 0.05$), Means within a row followed by different capital letters ($^{A–E}$) are significantly different ($p < 0.05$).

Very limited studies are available on the effect of gamma radiation on L*, a*, and b* surface color values for dried chilies. Extractable color values and carotenoid content were given more importance in research works, as compared to surface color analysis. In one study, Helga et al. (2018) [35] evaluated the effect of gamma irradiation on carotenoid content and ASTA color value for paprika samples that were irradiated at 1, 5, and 10 kGy. It was observed that the total carotenoids content and ASTA value decreased from its initial value of 2555.6 to 952.5 µg/g and 106.4 to 37.6, respectively, after radiation at 10 kGy. The author concluded that total carotenoid content and ASTA color value in paprika significantly decreased after ionizing radiation. Similarly, Iqbal et al. (2016) [36] reported that the initial carotenoid value in a control hot paprika sample was 76.9 mg/100 g, and declined immediately after radiation at 2, 4, and 6 kGy to 74.9, 72.1 and 70.7 mg/100 g, respectively. For both radiated and non-radiated samples, a loss of an average of 12% was found after 3 months of storage. The researcher highlighted that carotenoid is susceptible to gamma irradiation, and the drop could be attributed to an increase in the oxidation reaction, which is induced by energy absorption during irradiation. In another study, (Ayob et al., 2021) a significant decrease in carotenoid content as the gamma radiation and storage time was increased was observed. The author highlighted that capsanthin decreased the most (29.9%), while capsorubin decreased the least (19.2%) at 9 kGy radiation and 9 months of storage.

Jose et al. (2000) [32] studied the effect of electron beam irradiation on surface color and ASTA extractable color for paprika powder. The ASTA value for irradiation doses less than 5 kGy showed 1 to 2% loss, while at the highest irradiation dose, 12.5 kGy, there was a loss of around 6 to 7%. Lee et al. (2004) [33] evaluated the effect of gamma radiation on extractable color and surface color (Hunterlab L*, a, b) for Korean red pepper powder. In this study, ASTA units did not have significant difference on irradiated red pepper powder up to 7.0 kGy. It was concluded that irradiation at 7.0 kGy did not change the essential quality of red color in samples evaluated. In another study, Ayob et al. (2021) [4] observed a significant decrease in carotenoids as the gamma radiation dose and storage time was increased, in a way which was supportive to the findings in this study. Further, the author highlighted that capsanthin decreased the most (29.9%), while capsorubin decreased the least (19.2%) at 9 kGy radiation and 9 months of storage.

Apart from the above explanation, another possible reason for color loss during storage at an ambient temperature could be due to deterioration in ascorbic acid/vitamin C content that is found naturally in dried chili. Ascorbic acid is very sensitive to environmental conditions, and loss in ascorbic acid is strongly correlated to color loss in dried chili. Fontana (2000) [37] has mentioned that vitamin C degrades as water activity increases, in good agreement with our observation in this study. In another study, ref. [38] highlighted that color loss in dried paprika stored at high temperature could be the consequence of vitamin C breakdown. To support this, Calucci et al. (2003) [39] observed in their study that when diverse aromatic herbs and spices were irradiated at a dose of 10.0 kGy and stored for three months, the concentration of ascorbic acid fell by an average of 21%. Similarly, Iqbal et al. (2016) [36] observed that gamma radiation at 2, 4, and 6 kGy did not significantly affect ascorbic acid concentration as compared to the control sample. However, the author observed that ascorbic acid decreased significantly in radiated and non-radiated sample after storage for 90 days.

3.3. Capsaicin and Dihydrocapsaicin Content

Limited studies are available on the effect of gamma radiation on capsaicin and dihydrocapsaicin levels in dried chili, as most of the time importance has been given to microbial profiling. In this study, a negative correlation was observed between the increase in gamma radiation and its effect on the pungency components analyzed, capsaicin and dihydrocapsaicin (Table 4). The initial value for capsaicin and dihydrocapsaicin were 359.9 ± 3.4 ppm and 170.4 ± 1.6 ppm, respectively. Radiation at 10.0 kGy showed a larger decrease in capsaicin values in comparison to 7.5 kGy radiation, whilst dihydrocapsaicin showed the lowest value at 5.0 kGy. The observed trend for capsaicin content is in good agreement with the findings by [40], where the capsaicin content decreased as the radiation dose was increased in commercial red pepper powder. The author reported that the initial capsaicin value was 56.7 mg, and after radiation at 3, 6, and 9 kGy, the value dropped to 55.3, 50.1, and 48.2 mg, respectively. Further, the author mentioned that high irradiation dose affected capsanthin and capsaicin content, but at the end of the storage time, the differences between irradiated and un-irradiated samples were not impressive.

Table 4. Mean ($n = 3$) values for capsaicin (Cap) and dihydrocapsaicin (DHC) (mg kg^{-1}) for irradiated dried Byadgi chili at different irradiation dose (0, 2.5, 5.0, and 7.5 kGy) and storage period (0, 4, 8, 12, and 16 weeks).

Storage (Weeks)		0 kGy	2.5 kGy	5.0 kGy	7.5 kGy	10.0 kGy
0	Cap	359.9 ± 3.4 ^{Aa}	313.5 ± 1.3 ^{Ca}	340.1 ± 10.2 ^{Ba}	321.2 ± 0.6 ^{Ca}	288.9 ± 8.3 ^{Da}
	DHC	170.4 ± 1.6 ^{Aa}	138.7 ± 2.7 ^{Ca}	124.0 ± 2.1 ^{Eb}	131.1 ± 1.0 ^{Db}	143.4 ± 2.8 ^{Ba}
4	Cap	316.6 ± 2.3 ^{Ab}	312.4 ± 3.0 ^{Aa}	296.4 ± 5.7 ^{Bb}	278.4 ± 2.4 ^{Cb}	271.5 ± 0.9 ^{Db}
	DHC	109.5 ± 2.1 ^{Dc}	123.4 ± 2.2 ^{Cb}	142.3 ± 2.1 ^{Aa}	135.6 ± 1.8 ^{Ba}	121.8 ± 2.1 ^{Cc}
8	Cap	288.7 ± 4.3 ^{Ac}	267.9 ± 1.6 ^{Cb}	287.5 ± 3.9 ^{Ab}	276.3 ± 4.1 ^{Bb}	287.4 ± 3.1 ^{Aa}
	DHC	127.1 ± 0.3 ^{Bb}	112.5 ± 0.4 ^{Dc}	115.5 ± 0.4 ^{Cb}	107.4 ± 1.7 ^{Ec}	136.3 ± 1.9 ^{Ab}
12	Cap	268.5 ± 1.1 ^{BCd}	271.9 ± 4.2 ^{ABb}	263.2 ± 2.4 ^{Cc}	275.1 ± 3.9 ^{Ab}	253.3 ± 3.8 ^{Dc}
	DHC	93.6 ± 1.6 ^{Dd}	104.2 ± 1.1 ^{Bd}	98.4 ± 1.5 ^{Cc}	105.1 ± 1.5 ^{Bc}	110.0 ± 1.1 ^{Ad}
16	Cap	233.6 ± 7.7 ^{De}	269.8 ± 7.0 ^{Cb}	290.7 ± 10.0 ^{Ab}	271.4 ± 11.9 ^{Bb}	254.5 ± 3.3 ^{Cc}
	DHC	107.7 ± 0.9 ^{Ac}	83.6 ± 1.7 ^{Be}	113.7 ± 15.2 ^{Ab}	72.6 ± 0.1 ^{BCd}	66.1 ± 0.8 ^{Ce}

Means within a column followed by different small letters ($^{a–e}$) are significantly different ($p < 0.05$), Means within a row followed by different capital letters ($^{A–E}$) are significantly different ($p < 0.05$).

Contrary to the findings in this study, ref. [36] reported that radiation at 2, 4, and 6 kGy did not affect the capsaicin and dihydrocapsaicin content for sun-dried hot paprika samples that were prepacked with synthetic low-density polyethylene bags. Upon storage for 90 days, the levels of capsaicin and dihydrocapsaicin showed an average reduction of 3.4% and 4.2%, respectively. The author observed that capsaicin and dihydrocapsaicin content were more stable at 6 kGy, in which the lowest loss was observed at 90 days of storage. Similarly, [33], in their study on the effect of gamma radiation on pungency for Korean red pepper powder, concluded that gamma irradiation at 7 kGy did not affect its pungent quality. The initial value of capsaicinoids, the sum of capsaicin and dihydrocapsaicin, was observed as 40.07 mg/100 g. After radiation at 3, 5, and 7 kGy, the value slightly increased to 42.65, 41.89, and 44.99 mg/100 g. However, the observation did not show any significant difference between radiated and non-radiated samples.

In contrast to the findings in this study, ref. [41] reported an increase in pungency value after paprika powder was radiated at 5 kGy. However, the pungency value dropped generously during storage for up to 8 weeks in a way similar to the observation in this study. According to the researcher, a decrease in pungency value during storage could be an effect of milling and a residue enzymatic reaction. Similarly, ref. [42] reported that capsaicin and dihydrocapsaicin increased as the gamma radiation was increased, but decreased upon storage. The researchers highlighted that capsaicin is more stable than dihydrocapsaicin, which is similar to the findings in our study. In another study, ref. [43] mentioned that gamma radiation may help to retain capsaicinoids in chili pepper, in a way which is supportive of the findings reported by [42].

Apart from radiation dose applied, storage condition plays a very important role in the retention of capsaicin and dihydrocapsaicin. Naturally, dried chilies lose their pungency upon storage, critically at an ambient temperature. In this study, after 16 weeks of storage, sample radiated at 10.0 kGy showed 29.29% loss in capsaicin. For 7.5 kGy and the control samples, the decrease were recorded as 24.59 and 35.09%, respectively. Similar findings were reported by [44], where the author observed that capsaicin concentration in polyethylene packed hot peppers gradually and steadily decreased for 150 days of storage by 8.8% at 20 °C, 10.4% at 25 °C, and 12.9% at 30 °C. In correlation to these results, Topuz and Ozdemir (2004) [43] observed a 16% decrease in capsaicin and a 15% decrease in dihydrocapsaicin in powdered paprika after 6 months of storage at room temperature. Similarly, ref. [45] observed a continuous decrease in capsaicin and dihydrocapsaicin, 20.9 to 21.9% and 28.8 to 29.9%, respectively, for 9 months storage at an ambient temperature. Notably, the author also reported a high decrease in capsaicin and dihydrocapsaicin (overall 14–21.2%) at refrigeration temperature (2 °C to 5 °C).

3.4. Microbial Content

The initial total aerobic count (TAC) in dried Byadgi chili was $8.9 \times 10^3 \pm 8.1 \times 10^2$. At 7.5 kGy, TAC growth was observed, but at 10.0 kGy, complete sterilization was successfully achieved (Table 5). At the end of the storage, the control sample had the highest TAC count, $1.0 \times 10^4 \pm 1.7 \times 10^2$. Samples radiated at 10.0 kGy did not show a stable TAC count upon storage. Growth was observed at 4-, 8- and 16-weeks storage, but 12 weeks of storage had zero growth. This shows that there are certain strains of microorganism which are able to resist a radiation dose of 10.0 kGy.

Table 5. Total aerobic count (TAC) and mold of dried Byadgi chili irradiated at different irradiation dose (0, 2.5, 5.0, 7.5, and 10.0 kGy) and storage period (0, 4, 8, 12, and 16 weeks).

Storage (Weeks)		**Irradiation Dose (kGy)**				
		0 kGy	**2.5 kGy**	**5.0 kGy**	**7.5 kGy**	**10.0 kGy**
0	TAC	$8.9 \times 10^3 \pm 8.1 \times 10^{2\ \mathrm{Ab}}$	$6.3 \times 10^2 \pm 4.1 \times 10^{1\ \mathrm{Bc}}$	$2.1 \times 10^2 \pm 1.8 \times 10^{1\ \mathrm{Ba}}$	$6.7 \pm 5.8^{\mathrm{Bab}}$	0^{Bb}
	Mold	$11 \pm 1.7^{\mathrm{aA}}$	$2.0 \pm 1.7^{\mathrm{bA}}$	$3.3 \pm 5.8^{\mathrm{bA}}$	$3.3 \pm 5.8^{\mathrm{bA}}$	0^{bA}
4	TAC	$7.7 \times 10^3 \pm 5.8 \times 10^{2\ \mathrm{Ab}}$	$5.2 \times 10^3 \pm 3.3 \times 10^{1\ \mathrm{Ba}}$	$1.8 \times 10^2 \pm 2.0 \times 10^{1\ \mathrm{Cb}}$	$1.7 \times 10^1 \pm 9.1^{\mathrm{Ca}}$	3.0^{Cb}
	Mold	$7.7 \pm 4^{\mathrm{aAB}}$	$1 \pm 1.7^{\mathrm{bA}}$	0^{bA}	0^{bA}	0^{bA}
8	TAC	$6.8 \times 10^3 \pm 3.5 \times 10^{2\ \mathrm{Ab}}$	$1.1 \times 10^3 \pm 3.0 \times 10^{1\mathrm{Bb}}$	$2.0 \times 10^2 \pm 2.0 \times 10^{1\ \mathrm{Cab}}$	$1.1 \times 10^1 \pm 1.7^{\mathrm{Cab}}$	$4.0 \times 10^1 \pm 8.9^{\mathrm{Ca}}$
	Mold	$4.3 \pm 2.3^{\mathrm{aB}}$	$3.3 \pm 3.5^{\mathrm{abA}}$	0^{bA}	0^{bA}	0^{bA}
12	TAC	$4.7 \times 10^4 \pm 4.9 \times 10^{3\ \mathrm{Aa}}$	$1.1 \times 10^3 \pm 1.2 \times 10^{1\ \mathrm{Bb}}$	$4.3 \times 10^1 \pm 5.8^{\mathrm{Bc}}$	$5.7 \pm 2.3^{\mathrm{Bb}}$	0^{Bb}
	Mold	0^{aC}	0^{aA}	0^{aA}	0^{aA}	0^{aA}
16	TAC	$1.0 \times 10^4 \pm 1.7 \times 10^{2\ \mathrm{Ab}}$	$6.1 \times 10^2 \pm 8.1^{\mathrm{Bc}}$	$3.8 \times 10^1 \pm 6.8^{\mathrm{Cc}}$	$1.0 \times 10^1 \pm 3.0^{\mathrm{Cab}}$	$5.7 \pm 2.3^{\mathrm{Cb}}$
	Mold	0^{aC}	0^{aA}	0^{aA}	0^{aA}	0^{aA}

Means within a column followed by different small letters ($^{\mathrm{a–c}}$) are significantly different ($p < 0.05$), Means within a row followed by different capital letters ($^{\mathrm{A–C}}$) are significantly different ($p < 0.05$).

Mold plays a very important role in dried chili quality because some strains of molds, such as *Aspergillus flavus* and *Aspergillus parasiticus*, produce aflatoxins in dried chili. In this study, the visual appearance of the control sample did not show mold appearance until the end of storage period. This could be due to very low initial mold count, which was 11 ± 1.7 cfug^{-1}. The mold count reduced as the storage time was increased. Mold growth was not observed for the control sample and 2.5 kGy sample from 12 weeks of storage onwards (Table 4). Radiation at 5.0 and 7.5 kGy showed mold growth after radiation, but no growth observed upon storage. Even though mold growth was observed for radiated and non-radiated samples, aflatoxin was not detected in any of the samples until the end of the storage period. Yeast growth was not observed in any samples before and after radiation. The overall microbial count reduction could be due to the production of free radicals, reactive oxygen species, and other products from gamma radiation that might have interacted with cellular composition, leading to microorganism inactivation [41,46].

Verma et al. (2015) [6] reported that the irradiation of local dried chili at a dose of 4.0 to 6.0 kGy is sufficient to diminish overall microbial load to an acceptable level and eliminate fungal development. Unlike in this study, even though the initial TAC and mold count for control sample was 10^3 and 10^1 CFU g^{-1}, zero growth was not achieved at 7.5 kGy radiation. Only 10.0 kGy radiation showed good resistance against TAC and mold. A similar finding was observed by [40], where the researcher evaluated the effect of gamma irradiation on the sterilization and physiochemical changes of Korean red pepper powder during three months of storage. Samples were radiated at 3.0, 6.0, and 9.0 kGy. Gamma irradiation at 9.0 kGy sterilized the red pepper powder samples from the initial count of 3.83×10^6 and 6.68×10^6 CFU g^{-1}. Further, 9.0 kGy radiated samples did not show any bacterial growth upon storage for up to three months at room temperature.

Rico et al. (2010) [27] reported that irradiation at 10.0 kGy led to a 5 log reduction in microbial load, as compared to steam treatment, which showed 1 to 2 log reduction from the initial microbial load of 10^6 CFU g^{-1}. Further, yeast and mold counts were reduced to 10^{-1} CFU g^{-1} from the initial count 10^{-3} CFU g^{-1}. After storage for 6 months, the researcher observed zero count for yeast and mold, in a way which supports the findings in this study.

In a related study, ref. [28] observed that at 6.0 kGy, the total mold and *Aspergillus* count of whole dried peppers was completely destructed compared to initial count of 3.68 and 2.67 log CFU g^{-1}, and it was also reported no counts were observed for up to 90 days of storage. In another study conducted by [47], it was observed that decontamination by gamma irradiation at 10 kGy effectively eliminated the total bacteria count from the initial value of 4.0×10^7 CFU g^{-1}, and no growth were observed for up to 6 months of storage, while only 5.0 kGy was required for fungal elimination from the initial count of 1.5×10^4 CFU g^{-1}, and proliferation of mold was not observed for up to 6 months of storage, even though the initial count after radiation at 5.0 kGy was 40 CFU g^{-1}. The author observed that microbial quality in irradiated spices were retained up to 6 months of storage, as compared to non-irradiated spices, in a way opposed to the observation in this study, where TAC was observed after 16 weeks of storage.

Helga et al. (2018) [35] from Hungary studied the effect of different decontamination methods on microbial load, bioactive component, aroma, and color of paprika powder. The methods analyzed were gamma irradiation, steam treatment, microwave heating, enhanced microwave treatment, and radio-frequency heat treatment. As for gamma irradiation treatment, the paprika powder samples were exposed to 1.0, 5.0, and 10.0 kGy. Among the treatments performed, the author observed that radiation treatment was highly effective against reducing and eliminating microbial count. However, mold count was not eliminated, even at 10.0 kGy.

Dikkala et al. (2018) [48] observed that 2.5 kGy of gamma irradiation effectively reduced fungal growth in whole grains, with radiation curves showing a decrease in microbial growth with an increase in irradiation dose, indicating that high doses of gamma irradiation were more effective to eradicate total aerobic count, yeast, and mold. The death of microorganisms through radiation might be the result of their high sensitivity to gamma radiation that causes direct and indirect DNA damage [48,49]. Gamma radiation can affect DNA directly through ionizing rays or indirectly by the free radicals (*H, *OH and $e_{aq}{}^-$) formed from water [50].

In this research, *E. coli* was not detected in all the samples analyzed, even though studies have reported its growth in dried chilies. Gamma radiation has been proven to be effective against *E. coli* activation. For example, [36], in his research, observed a reduction in *E. coli* colonies with an increase in gamma radiation dose. The researcher explained that the dose-dependent observation could be due to inability of the pathogen to recover from the injuries caused by gamma radiation and the inability of the injured cells to adapt to their environment. Similar observations were also highlighted by other researchers [51,52].

From this study, it was clearly observed that 7.5 kGy gamma radiation dose was not able to completely sterilize the samples tested. The finding from this study is in line with a study conducted by [47]. The author mentioned that 5 kGy radiation is suitable for decontamination, but emphasized that radiation at 10 kGy is necessary to completely eradicate total bacterial load. To decide on this, the author suggested observing the initial microbial count in the dried chilies designated to be radiated.

To determine the ideal radiation dose for dried chili sterilization, the initial evaluation on total microbial load and the types of microorganism present is very important. For example, in a study conducted by [53] for red pepper powder decontamination, a radiation dose of 18 kGy was required to reduce the TAC to 1.0 $cfug^{-1}$ from the initial count of 1.8×10^6 $cfug^{-1}$. Furthermore, the study sample was also heavily contaminated by coliform (1.3×10^5 $cfug^{-1}$), Bacillus cereus (4.8×10^4 $cfug^{-1}$), and yeast and mold (6.2×10^3 $cfug^{-1}$). However, complete sterilization was not achieved in this study, even though 18 kGy was used. Similarly, Iqbal et al. (2012) [28] was not able to achieve sterilization for dried chili and paprika powder after radiation at 6 kGy, where the initial counts were found to be about 10^7 to 10^4 $cfug^{-1}$, respectively. Helga et al. (2018) [35] highlighted in their findings that, at 10 kGy radiation, the aerobic mesophilic count was reduced to

2.91 log $cfug^{-1}$ from the initial value of 6.84 log $cfug^{-1}$. Lee et al. (2004) [33] reported that a radiation dose of 7 kGy did not eliminate the total mesophilic count from the initial value of 6.71 log $cfug^{-1}$.

Some similar studies reported that radiation at 10 kGy was able to sterilize dried chili or reduce the TAC to an acceptable level. For example, a study conducted by [54] is in good agreement with this research work: complete sterilization was achieved at 10 kGy for Korean red pepper powder from the initial count 5.27 log $cfug^{-1}$. In a related study, ref. [40] observed that a dose of 9 kGy was required to completely sterilize a red pepper powder sample from the initial count 3.83×10^6 $cfug^{-1}$. Thus, the initial level of contamination plays a very important role in determining the irradiation dose required in accordance with the intended purpose, either to sterilize or reduce microbial load to an acceptable level. The decision may be influenced by storage duration and storage condition.

3.5. Relationship between Dried Byadgi chili Quality Characteristics as Affected by Irradiation Doses and Storage Times

Principal component analysis (PCA) was performed to further elucidate the relationship between the effect of different gamma radiation doses and storage time on quality attributes of dried Byadgi chilies samples. Figures 1 and 2 depict the correlation relationships between variables and samples, as well as observations on specific trends related to the study's combination treatments, respectively. It can be observed that the variation from PC1 and PC2 is greater than 70% (72.4%), showing a good depiction of information which can be extracted from the first 2 PCs (Figure 1). The x—axis revealed a clear separation based on shorter storage times (0, 4, and 8 weeks) and longer storage times (12 and 16 weeks), with shorter storage time clustered in quadrants I and II, and longer storage time clustered in quadrants III and IV (Figure 2). According to Figure 1, samples in quadrants III and IV may have poor microbial and physicochemical properties. Samples in quadrants III and IV, for example, are positively correlated with high moisture content and water activity, which is associated with decreased shelf-life stability. Samples of higher doses of radiation, particularly of 5.0 kGy and 10.0 kGy, were found to be located adjacent with color and microbial properties. As for the non-radiated samples, microbial content, especially TAC, had a drastic count, which is distinguished from the rest of the samples, especially at week 0 (D0W0) and week 12 (D0W12). Furthermore, the effect of storage was discovered to spread along PC1, from quadrant II to quadrant III in the following order: D0-W0, D0-W4, D0-W8, D0-W12, and D0-W16 (Figure 2). This indicates that the quality of the samples deteriorates as storage time increases. The effect of radiation, on the other hand, was observed to spread along PC2, indicating an improvement in quality parameters from D0-W4, D2.5-W4, D5.0-W4, D7.5-W4, and D10.0-W4.

It can also be observed that the longer storage time at an ambient temperature, especially at week 12 and week 16, showed a negative impact for radiated and non-radiated dried Byadgi chili, notably in quadrant III and IV. Hue value at quadrant III could be an indication that the samples change to be darker in color. Similar findings were reported by [55], and the researcher suggested that treatments such as modified atmospheric packaging and cold storage for dried chili may have contributed to delaying color changes. In a related study, ref. [56] reported 42.1% reduction in carotenoids for 10.0 kGy radiated paprika that was stored for 10 months at an ambient temperature. The initial level of microorganisms in the non-radiated sample was very high, as indicated in D0-W0, and the highest count for the control sample was observed at 12 weeks, D0-W12. This indicates that total microorganisms multiplied over the time until reaching the lag phase at 12 weeks storage, and thereafter started to decline. According to PCA results, radiated dried Byadgi chili samples with shorter storage time had better physicochemical attributes in surface color, capsaicin, and dihydrocapsaicin, which was consistent with the findings reported by [43], who reported a 14% and 23.4% decrease in pungency value for radiated paprika after storage for 4 months and 10 months, respectively.

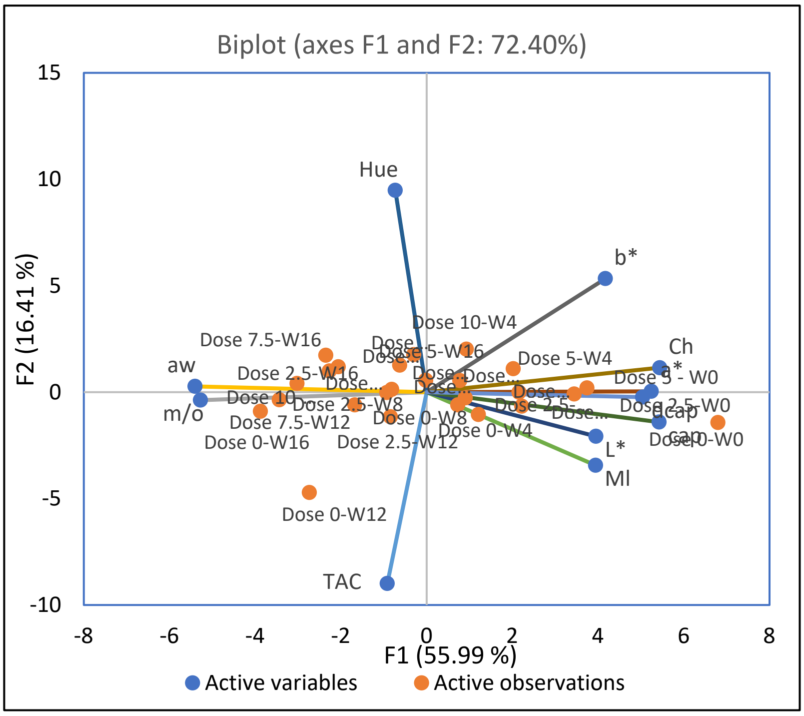

Figure 1. PCA plot indicating the correlation relationship between quality attributes of dried Byadgi chili samples, as affected by irradiation dose and storage times.

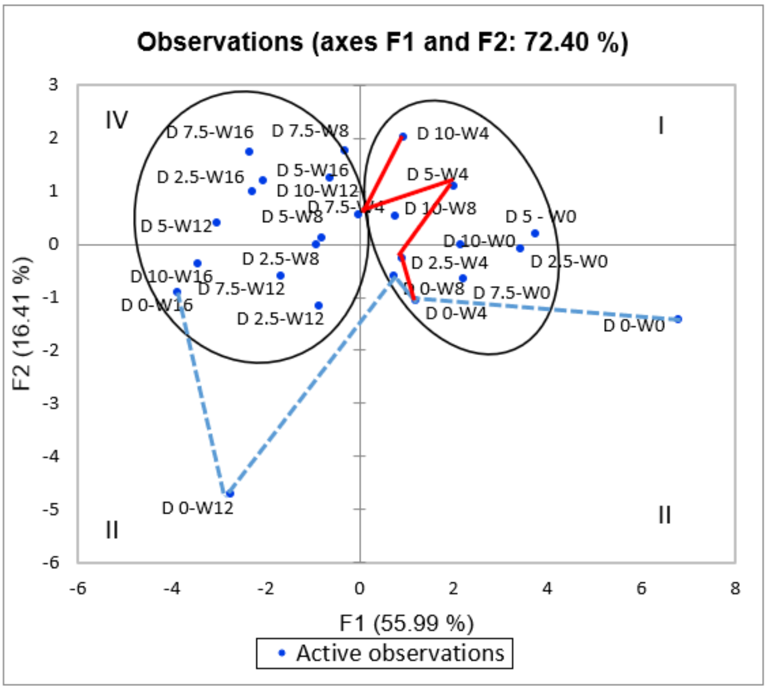

Figure 2. PCA on combination treatments of irradiation doses and storage times. Ellipses indicate sample separation, and lines (blue and red) indicate a potential trend.

4. Conclusions

The efficacy of gamma radiation and its storage quality at an ambient temperature on the microbial and physicochemical quality attributes for dried Byadgi chili was investigated. Moisture and water activity increased with radiation. However, upon storage, radiated samples showed lower moisture and water activity levels in comparison with the control sample. Radiation at 10.0 kGy demonstrated sterilization with good microbial profile until

16 weeks of storage, whereas radiation at 5.0 and 7.5 kGy showed better sanitary effects up to 16 weeks of storage, even though complete sterilization was not achieved. It was observed that radiation did not help in preserving the surface color of dried Byadgi chili. Radiation at 5.0 and 7.5 kGy can be used to reduce microbial load to an acceptable level with minimal changes in physical properties when stored at ambient temperature for 16 weeks.

Author Contributions: Conceptualization, N.B. and S.M.Y.; investigation, N.B. and S.M.Y.; writing—original draft preparation, N.B. and S.M.Y.; writing—review and editing, N.B., E.D., I.A.R. and S.M.Y.; supervision, S.M.Y., I.A.R. and A.A.; facility and support I.A.R. and S.M.Y. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Informed Consent Statement: Not applicable.

Conflicts of Interest: The authors declare no conflict of interest.

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