



Effects of Gamma Radiation and Electron Beam on Samples of the Food-green Peanuts, Organic Peanuts, and Eco-labelling Green Peanuts Industry Artificially Inoculate with *Aspergillus flavus*

Gisele Ferreira de Souza^{1*} and Jair Ribeiro dos Santos Junior²

¹Faculty of Medicine, University of São Paulo (USP), Brazil

²State University of Maringá (UEM), Brazil

*Corresponding Author: Gisele Ferreira de Souza, Faculty of Medicine, University of São Paulo (USP), Brazil.

Received: March 02, 2020

Published: July 30, 2020

© All rights are reserved by Gisele Ferreira de Souza and Jair Ribeiro dos Santos Junior.

Abstract

The purpose of this research was to assess the effects of Gamma Radiation as well as Electron Beam on samples of Brazil nuts that are contaminated with *Aspergillus flavus* under temperatures of 30°C and a humidity of 93%. The process took place in fifteen days of incubation where aflatoxins and mycobation are analyzed. The samples were further grouped into three groups namely, control, group 1 and group 2 which receive radiation of 0, 5 and 10 kGy dosage of electron beam (EB) and gamma radiation (GR). Some samples of not inoculated were illuminated with a similar dosage to evaluate the sensors. The results indicated that 0.80 of the samples had an average water capacity. Illumination or irradiation of gamma radiation and electron beam at a dosage of 5 and 10 kGy were able to eliminate the *A. flavus* fungi in the samples of Brazil nuts. Analyses of aflatoxin indicated that electron beam doses of 5 and 10 kGy lowers aflatoxins levels by 53.32 and 65.66%, respectively. Moreover, this same dosage of gamma radiation lowered the levels of toxins by 70.61 and 84.15%, respectively, as compared to the control group. Sensory assessment showed that texture and smell of the illuminated samples of Brazil nuts were acceptable.

Keywords: Gamma Radiation; Electron Beam; Brazil Nuts; *Aspergillus flavus*; Aflatoxin

Abbreviations

AFB1: Aflatoxin B1; AFB2: Aflatoxin B2; AW: Water Activities; EB: Electron Beam; GR: Gamma Radiation; PBS: Phosphate Buffered Saline; FAO: United Nations Food and Agriculture Organization; IAEA: International Atomic Energy Agency; WHO: World Health Organization

Introduction and Literature Review

Fungi can contaminate foods in their phases of production under some minimum conditions, such as temperature and humidity [1]. The Brazil nut is a standout amongst the most important prod-

ucts of the Amazon region and are marketed in shelled or stripped structures. The nuts contain 17% protein and 67% lipids, in addition to basic amino acids, such as methionine and cysteine, and minerals, for example, selenium [2]. The fungal disease in Brazil nuts has been studied over the last century, with the aflatoxigenic *Aspergillus flavus* being one of the species that is mostly observed in this commodity. The *A. flavus* is found in *Aspergillus* segment *Fluvi* and develops well on oleaginous substrates, which support the formation of aflatoxins. Aflatoxins are a group of fatal and carcinogenic auxiliary metabolites delivered by specific strains of *A. flavus*. The most generally produced toxins are aflatoxin B1 (AFB1) and

aflatoxin B2 (AFB2) [1]. AFB is especially vital since it is classified as a number 1 human cancer-causing agent by the International Agency for Research on Cancer.

The growth and increase of *A. flavus* is dependent upon water activities (aw) at dimensions of 0.78 to 0.82, a general moistness of 80 to 90%, and temperatures above 25uC [3]. The ideal temperature for the development of this fungus is about 35uC [4]. Aflatoxin formation is perceived at dimensions of 0.83 to 0.87 and temperatures of 12 to 42uC [5]. These variables support fungal contamination in tropical and subtropical districts. The hindrance of fungus development, especially, *A. flavus*, which is aflatoxigenic in nuts, is essential to diminish the potential hazard to human wellbeing. In this circumstance, decontamination techniques utilizing ionizing vitality are being considered as choices.

Ionizing radiation can lessen *A. flavus* contamination in various foods [1]. To mycotoxins, irradiation can diminish aflatoxin content in foods by delivering radicals that follow up on the terminal furan ring of these poisons. Techniques dependent on high energy production provided by machine sources or gamma beams from radionuclides, for example, ⁶⁰Co, convey enough energy to expel electrons from the circles of atoms, in this method creating ions. Gamma radiation (GR) and electron shaft (EB) have been known to demonstrate a comparable impact on foods. However, there are a few contrasts in regards to infiltration and the strategy for their application.

Advantageous impacts of irradiation include decrease of storage loss, an expanded timeframe of nutrition in foods, and disposal of microbial and fungal contaminations from the feed, with further advantages including the absence of surplus impacts after illumination and being useful for the earth [6]. The treatment of food with ionizing radiation, as well as other forms of food processing, can create physical and chemical changes. To guarantee negligible differences, for most prepackaged food, a 10-kGy portion is viewed as the greatest required [6]. Studies researching control techniques for organisms and mycotoxins in Brazil nuts are scanty, and there are no reports on the control of *Aspergillus* and aflatoxins in Brazil nuts by irradiation systems. Thus, the present analysis aims to assess the impact of ionizing radiation (GR and EB) on *A. flavus* disease and AFB1 tainting in Brazil nuts.

The Chestnut tree

Chestnut trees produce nuts and they are located in different countries in the Amazon region including Colombia, Peru, Guyana,

Suriname, Brazil and Venezuela. These trees are dense and tall arboreal formations, found in forests. The Chestnuts trees are large mature trees that grow on poor soils that are drained, unstructured as well as clayey or sandy [7].

The Amazon regions that produce nuts have humid and hot climates. In these regions, the temperature can vary from minimums between 19.2 and 23.4°C to maximum of 30.6 - 32.6°C [8]. The annual precipitation varies from 1400 and 2800 mm, while the relative humidity is between 79% and 86% [9]. Chestnut trees can grow to a height of fifty meters, and their bases can be up to two meter in diameter. They have a cylindrical, straight and smooth trunk, and branch out into the upper portions of the crowns. During the flowering phase, from November to February, they present creams and white flowers (Garlic, 2010).

The Brazil nut

After the decline of latex exploration in Brazil, the Brazil-Brazil, which belongs to the botanical family of *Lecythidaceae*, became a fundamental resource of extraction and exportation in the northern region of the country. Thus, in order to control the trade in this item, Federal Decree No. 5,975/06 was proposed, which regulates the Forest Code and the National Environmental Policy. This decree establishes principles on the Management Plan, evacuation of vegetation, use of raw wood, reforestation and permission to transport items from the forest [9].

Sustainable forest

The Brazil nut are currently one of the most important plant is the Amazon, given their biological, social, financial and nutritional importance. Its commercialization is done mainly in natura and a smaller part is dry, for conservation. In Brazil, the technology used in food preparation has not undergone critical changes over the years. However, the recent government activities to assist in innovative technologies for food preservations have produced exceptionally positive outcomes, since cooperatives and associations have gained greater bargaining power, since, as it is an extremely delicate product, buyers ended up directing prices [7,10].

The commercialization of Brazil nuts, in connection with foreign to exchange, follows two foreign exchange flows, domestic and foreign exchange; this proportion was changed from 70% to 75% of domestic use and from 25% to 30% for export. The main destinations for marketing Brazil nuts in natura are Bolivia, the United States, Peru, Honk Kong, Europe and Australia [7,11]. Cur-

rently, Brazil has lost its largest nut export station to Bolivia. With the absence of public agreements, there is a decrease in the number of profitable chestnut trees, lack of motivation for advertising approaches, in addition to problems to meet phytosanitary export prerequisites [7,11].

Nutritional estimation of the Brazil nut

According to Gonçalves., *et al.* [12], the Brazil nuts are an oleaginous almond of high vitality and dietary benefit, with different components necessary for optimal nutrition. In addition to its nutritional quality, it has a pleasant taste. Studies have demonstrated that Brazil nut have from 60 to 70% of lipids and around 15 to 20% of highly esteemed organic protein (Table 1), estimated at several times the cow-like milk protein.

Components	Brazil nut (Quantity g. 100g ⁻¹)
Moisture	3.10
Lipids	64.94
Proteins	14.11
Nitrogen	2.62
Dietary fiber	8.02
Carbohydrates	6.27
Embers	3.56
Energetic value	655.98

Table 1: Proximate composition and energy value of the Brazil nut edible.

Source: Freitas [13].

The presence of the albumin fraction, excelsin, makes the nut considered only a plant of whole food with its origin known as “vegetable meat.” Among the essential amino acids are present in nuts, leucine, isoleucine, methionine, cysteine, lysine, tryptophan, valine, and threonine [7,10]. The lipid fraction, divided into mono- and polyunsaturated fatty acids, is presented in the following proportions: 37.75% linoleic acid, and oleic 37.42% oleic acid, which is equivalent to 75.17% of the total fatty acids 24.83% represent saturated fatty acids, containing 13.15% palmitic, 10.36% stearic and 1.32% arachidonic [12].

In addition, the almond also contains minerals that are essential for the human body, such as phosphorus, copper, potassium, zinc, magnesium, manganese, calcium, among other nutrients, however,

the concentration of these compounds changes according to the area where they are located the trees [12]. Another constituent present in the nut is selenium, a vital cell reinforcement, alluded to responsible for the prevention of malignancy and, moreover, associated with the digestion of the thyroid. Its insufficiency is related with cardiomyopathy, solid dystrophy in multiplication in different species of creature, including people, so that admission in satisfactory sums of Brazil nuts has been prescribed for individuals who seek to improve their personal satisfaction [14].

Mycotoxins

In Brazil Resolution - RDC No. 7, of February 18, 2011, of the National Health Surveillance Agency, provides the maximum admissible dimensions for mycotoxins in nourishment.

Aspergillus flavus

The types of the class *Aspergillus* are among the most essential in food spoilage; these are described by the advancement of brilliant and bright provinces and conidia creation in head type “mop-like” (scouring brush) [3]. The primary species having a place with *Aspergillus Flavi* area are *Aspergillus flavus*, *Aspergillus parasiticus*, *Aspergillus nomius*, *Aspergillus oryzae* and *Aspergillus sojae*. In any case, *A. flavus*, *A. parasiticus*, and *A. nomius* get more consideration by the mycotoxin creation capacity. The species *A. oryzae* and *A. sojae* are viewed as sheltered since there was no history of creating poisons and are used in the food business [3].

The parasitic contamination in the field begins during harvesting, drying, and storage of the substrate. The genus *Aspergillus* spp. is generally dispersed and frequently happens in tropical and subtropical areas with high humidity. However, it can develop in states of high temperatures and low water action, along these lines contaminating a wide range of substrates and is therefore found during the storage time frame [3]. As indicated by Baquião [9], the parasitic contaminating can occur in the field and continue during storage as well as during the fruits transportation to the processing plants.

The toxigenic growths are those fit for delivering mycotoxins under ideal conditions, inhabit and develop to create poisons in an assortment of substrates, as per the necessities of every species (Stoloff, 1979). *A. flavus* oleaginous extends well on substrates, which have a higher generation limit aflatoxins. They may likewise deliver aflatoxins in lower recurrence rich substrates, for example,

maize starch [15,16]. The water activity (aw) of foods is portrayed as the measure of available free water. It includes water which isn't focused on chemical bonding, and different solutions of solutes [17]. The water activity is estimated to a range of 0 and 1, with 1 being the esteem found in the water. All toxigenic organisms present least, ideal and greatest Aa for development (Jay, 1994), the vast majority of which develops in a water movement around 0.85 [17]. In accordance with Lacey [15], the base Aa esteems for the development of *A. flavus* are in the range of 0.78 to 0.82. The relative air moisture of 80% to 90% and temperatures over 25°C support the development and improvement of *A. flavus*, and the typical development of this growth happens at a temperature around 35°C [18,19]. The species *A. flavus* emerges for being a standout amongst the essential producer of aflatoxins, having the capacity to incorporate aflatoxin B1 and B2 [3].

Aflatoxins

In 1960, more than 100,000 turkeys died in ranches in England, with the disease, initially called "Turkey X Disease." This disease was not constrained to turkeys, yet different winged creatures were additionally affected. After a careful examination to identify the cause of the disease, it was discovered that it was related with clumps of shelled nut supper traded from Brazil that had been consumed by the birds. After an investigation of the husk of the grains, it was found that this product contained something exceptionally poisonous to the creatures, from which point different hypotheses arose in England about the idea of the poison. But it was in 1961 that the poison was recognized as *A. flavus*, the maker of aflatoxin, a name given in light of their beginning (*A. flavus* - Afla) [20,21].

That same year, there was a major investigation on the carcinogenic impacts of aflatoxin, where researchers saw the improvement of dangerous tumors in rodents that consumed poisoned feed in portions that contained low doses, and different doses that caused intense damage [22]. According to the United Nations Food and Agriculture Organization (FAO) evaluation, 25% of the foods consumed worldwide have mycotoxin contamination, the most impressive of which are aflatoxins. The annual misfortune is approximately \$ 1 billion, including spending on human, animals and degraded agrarian items [18]. Aflatoxins, synthetically intertwined to a coumarin core bifurano pentanone ring and a 6-lactone ring.

Ionizing radiation

The ionizing radiation (X-beams, gamma radiation, electron bar) is the structure which has adequate energy to expel electrons

from atoms circles consequently creating ions. High-energy radiation that causes ionization of the medium in which it is absorbed contrasts from non-ionizing radiation (microwaves, radio waves and TV) that does not have enough energy to ionize the material. The energy consumed in excitation and ionization of atoms produce chemical reactions that may forever change the physical and chemical structure of the radiated material. The absorbed dose or treatment is the measure of energy retained per unit mass of radiation material. The unit utilised is called Gray (Gy) or quilogray (kGy), where 1 Gy is identical to 1 Joule of energy consumed per 1 kg of illuminated materia.

The idea of the usage of ionizing radiation in foods emerged after the discovery of radioactivity by Henri Becquerel in 1895. Around the same time, there was a suggestion in the publication of a German medical journal, on the utilization of radiation for the control of pathogens in foods. In mid-1900, studies in the United States and the United Kingdom reported the usage of ionizing radiation in the elimination of microorganisms in diets. Around then, the primary sources of ionizing radiation were the radio that the non-accessibility, made the procedure monetarily. However, there are reports of different investigations using X-beams to kill bugs and larvae in tobacco leaf and furthermore parasites found in pork meat. With the development of atomic reactors to ionizing, radiation has turned out to be increasingly possible, and more studies have been accomplished for the safety of foods applications. The first nation to allow consent for human use on irradiated foods was the former Soviet Union, which in 1958 approved the light of potatoes to repress growing of this food [23].

Pathogenic microorganisms can be eliminated through radiation for the safety of people and animals, this technique is fundamentally used for sanitization of pharmaceuticals and medical products, as well as foods illumination, since ionizing radiation contributes to the control of pests and insects in agricultural products [24]. The committee board of the food radiation formed by the organs of the United Nations Food and Agriculture Organization (FAO), International Atomic vitality Agency (IAEA) and the World Health Organization (WHO) and the Codex general standards for irradiated foods, offers recommendations on the types of ionizing radiation that is appropriate for food irradiation: X-beam with energy up to 5 MeV gamma radiation originating radionuclide cobalt 60 (⁶⁰Co) and Cesium (¹³⁷Cs), electrons with power up to 10 MeV.

As indicated by Tsai [24], the generation of X-beam is done at accelerators where the electron bar focuses around a substantial

material target producing, for braking, X-beams, this impact is otherwise called “Bremsstrahlung impact”. This kind of generator has exceptionally low proficiency, because close to 10% of the energy of the electrons is changed over into energy as electromagnetic waves (X-beams), so the attainability of using radiation electron bar and gamma radiation is higher. Isotopes that always discharge gamma radiation cannot be turned on or off as an X-beam machine, Therefore, the gamma radiation and electron beam has been used for sterilization of food and medical supplies, and new tools have been designed for this reason. Not a wide range of ionizing radiations are used for the irradiation of foods, since they don’t show enough penetration energy into the material (Figure 1), for example, alpha particles [25].

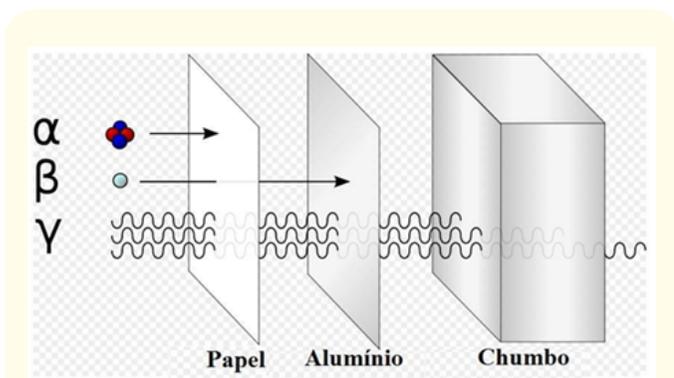


Figure 1: Penetrability of ionizing radiation α particles, β (electrons) and gamma rays.

Source: Wikimedia Modified (2012).

The infiltration of ionizing radiation is inversely proportional to the density of the produced product, and the density and thickness of the material to be used. It is critical for the application of gamma radiation or electron beam where the intensity of the electron bar penetration is not as much as gamma beam, for instance, a material with a thickness of 1 g/cm³ (Similar to water) the penetration intensity of an electron bar with energy of 10 MeV is 5 cm, while utilizing gamma radiation from cobalt-60 and average energy of 1.25 MeV is around 50 cm [24].

There are prescribed strategies for food protection using food technologies, in a sense, food irradiation can be utilized alone or in conjunction with other technologies, for example, cooling, freezing, warming and packaging. The radiation used in food prepara-

tion don’t have enough energy to make any atomic response in the art, that is, don’t respond with cores of atoms of the material hence leave no radioactive waste in the material after irradiation, so the food or some other substances that are exposed to such radiation, don’t end up radioactive (IAEA, FAO, WHO, 2012).

Radiation electron beam

A particle accelerator is a gadget that applies powers charged particles through a combination of electric and magnetic fields, producing particles with fast and high power, while setting up a potential high voltage between a cathode and an anode, in vacuo, the cathode discharges electron beam, as indicated by principle standard like the flat tube however the TV tube utilizing around 25,000 volts of intensity, as accelerators for industrial voltage fluctuates between 200,000 volts to 10,000,000 volts [24,26].

Electron accelerators were not initially used for sterilizing procedures; due to problems that were generated by the limitations associated with the number of factors such as energy, scanning width, beam intensity and belt speed. These variables impacted the ingested dose. However, with the advancement of accelerator technology, it has been possible to adjust different components to machines with complex electronic structures, in order to allow these equipment to meet the standard irradiation of products and their use in sterilization processes [24].

According to Silva, *et al.* (2011), electron accelerators are broadly used in industrial processing, particularly in the safeguarding and disinfestation of food. They have high processing abilities, favorable cost and the likelihood of intrusion of radiation sources. In contrast to gamma radiation, the utilization of electron beams, there is the speed in the processing of products. Electron accelerators are structured and fabricated for explicit uses and are classified by the energy range, and the accelerators with the range between 0.3 to 10 MeV are most regularly used for mechanical application.

In identifying the ionizing radiation procedure to be used, the items presented in table 2 must be evaluated as indicated by the needs of each case.

Proposals and Objectives

Since 1990, Brazil nut production has declined, when the country delivered just over 50 thousand tons per year. During the last

evaluation, Brazilian production has kept up since 2000, with thirty thousand tons per year. As time passes new technologies and innovations are developing for control of molds and mycotoxins in Brazil nuts., Scussel, *et al.* [27] proposed application techniques with a changed environment (ozone, carbon dioxide and adsorbents) in pre-packed nuts, one of the main tested applications, the one that increased the degradation of aflatoxin expansion to restrict the development of microorganisms was the use of ozone. Brazilian law permits the utilization of sources of cobalt-60 radioisotope and electron accelerators to irradiate food.

Radiation type	Benefits	Disadvantages
Gamma	Good penetration	Cannot be focused, bent, with implications efficiency.
	Low energy consumption	It cannot be turned off.
	Does not require specialized operators.	Licensing difficulties
		Recharge and transport source
Electron beam	Driving the material to be further facilitated	Low penetrability
	Can be focused, bent more flexibility in efficiency	High power consumption
	can switched off (lowest waste)	Need training
	Does not use material radiation.	Cost of equipment and installation

Table 2: Comparison of gamma radiation and electron bar. Source: [24,26].

Given the high aflatoxin contamination in Brazil nuts and the strict control of importing nations in connection to toxic levels in foods, the European Union nations chose in 2003 for the return of this product from Brazil. Thereafter, they started to request special conditions for the import of the item, for example, consistency with good extractive practices, with separate proof by the Brazilian experts and authorities and determination of aflatoxins by research centers authorized by the Ministry of Agriculture, Livestock and Supply. In Brazil, the necessities for the control of aflatoxins

in Brazil nuts are less prohibitive, so the parcels rejected by European nations can be coordinated to the domestic market, a reality that represent an incredible health risk. Therefore, control of fungal contamination and aflatoxin is critical not just because it keeps away from substantial economic losses and dangerous effect on the environment, but also establishes a matter of general health. The utilization of medicine by the irradiation procedure can be a significant method for containing the danger of fungal contamination and represents the most suitable option for decontamination of this product class. Studies on Brazil nuts are rare, and there are minimal reports on fungal control through irradiation procedures, particularly the electron beam.

Materials and Methods

Samples of Brazil nut

Some samples containing Brazil nuts were analyzed. These samples were obtained in some cities in Brazil, and weighed two hundred and fifty grams and contained approximately fifty nuts. These samples were packed in plastic bags, which were then wrapped in paper bags, sealed and finally sterilized. This sterilization process was carried out with ethylene oxide, used to eliminate fungi that occurs naturally. After that, the samples were then placed in a sterile glass dish and subsequently stored in sterile plastic boxes.

Preparation of inoculation and suspension of spores in samples of Brazil nuts

The toxigenic *A. flavus* that produces AFB1 and AFB2 strain IMI 190 was used in the production of spores. The *A. flavus* sample was acquired from the International Mycological Institute located in London in the United Kingdom. To enable counting of spores, *A. flavus* was planted onto the potatoes dextrose sugar maintaining it in an incubator with temperatures of 25uC for one week to allow its growth. Its cultures were cleaned using sterile phosphate buffered saline (PBS) and Tween 80 solution. The mixture was formed to dissolve 50 ml of Tween 80 in 100ml of PBS. The spores were counted inside a Neubauer chamber. The resulting solution was adjusted 1 out of 150 spores for each milliliter of solution. The Brazil nut samples were covered in with twenty drops of one milliliter of *A. flavus* solution.

Control of temperatures and humidity

For the humidity control, a saturated saline solution with thirty percent of potassium sulfate was used, which maintains a

relative humidity at 93% inside the sterilized plastic box in which the samples were stored. The temperature and humidity control were monitored using the Tualatin thermos-hygrometer, model ETHG880, OR and Oregon Scientific, placed inside the boxes containing the samples. These boxes were incubated for fifteen days at 30°C in a biological oxygen demand incubator.

Determination of water activities (*aw*)

The Aqualab CX-2 instrument was used to measure the *aw* of Brazil nut samples after the relative humidity and temperature had been determined.

Counting the plate

The fungal mycobiota was analyzed after as the samples were triturated. This was done by transferring ten grams of aliquots to the Erlenmeyer flasks with ninety milliliters of sterile water. These flasks were shaken using a Tecnal TE-140 shaker for thirty minutes at 160 rpm. Of these solution, one milliliter was used to make serial dilutions. 0.1 ml aliquot of each solution were added to Petri dishes containing the potato dextrose agar. These plates were incubated at 25°C for ten days to determine the amount of CFU in a one gram.

Determining the aflatoxins in the samples

For extraction, 25 g of every Brazil nut test was standardized. Then, 12.5 ml of an acetonitrile-water (85:15, vol/vol) solution were mixed with 2.5 g of the Brazil nuts sample, for each of the collected samples. This blend was shaken for sixty minutes. When centrifugation was completed, 5 ml of the supernatant holding nuts was blended with 45 ml of fermented water (0.5% chilly acidic corrosive), and 2.5 ml of the supernatant holding the shells was combined with 47.5 ml of J. Sustenance Prot. fermented water. This solutions were washed on a Strata C18-E cartridge (500 mg/3 ml, sorbent mass per volume; Phenomenex, Santa Clara, CA) at a stream rate of one drop for each. The containers contained 12 ml of fermented water. Aflatoxins were eluted with one milliliter of methanol and the eluent was dissipated to buildup. Deposits were dissolved with trifluoroacetic corrosive and hexane vanished once more as well as resuspended in 400 ml of a methanol-water (1:1, vol/vol) arrangement. The blend was infused into the Shimadzu Prominence elite fluid chromatography framework (Shimadzu, Kyoto, Japan) furnished with an RF 10AXL fluorescence identifier (excitation at 365 nm and emanation at 450 nm) and the auto

sampler framework. The investigative section (Shim-Pack VP-ODS, 150 by 4.6 mm; Shimadzu) was attached to a precolumn container (Shim-Pack GVP-ODS, 10 by 4.6 mm; Shimadzu) kept up at 40°C in a broiler. The isocratic portable stage comprised of acetonitrile-methanol-water (1.5:1.5:8, vol/vol/vol) in addition to 0.1% trifluoroacetic corrosive and was eluted at a stream rate of 1 ml/min. The strategy was approved as depicted already. The limits of confinement of measurement and recognition for all aflatoxins were 1.5 and 0.75 ng/g, separately. The recuperation rates were 80.44% for AFB1 and 83.28% for AFB2.

Irradiating nut samples

The inoculated samples were isolated into three groups: the control group, group 1 and group 2. Groups 1 and 2 were separated into two subsamples. The control group and each of the subsample were composed of 50 Brazil nuts (with an average of 50 g each), which were fixed independently in polyethylene bags under vacuum. The samples belonging to the control group were not lighted. The samples in groups 1 and 2 were lighted with dosages of 5 and 10 kGy, separately, utilizing GR or EB. The GR treatment was done at the Institute of Energy and Nuclear Research Brazil at a temperature of 25 to 28°C utilizing an adjusted 60Co source (Gammacell 220 N, Atomic Energy of Canada Limited) and a portion rate of 1.87 kGy/h; the vulnerability of the portion estimation was 1.7%. EB, unlike GR, has a constrained penetration depth, which may influence microbial inactivation relying upon the span of the package, and light on just a single side was not adequate to uncover the entire portion to the objective portion; subsequently, for EB, the examples were illuminated on the two sides. A DC 1500/25/4-Job 188 mechanical assembly (Radiations Dynamics, Inc., New York, NY) creating EB vitality of 0.55 to 1.50 MeV and a pillar current of 0.3 to 25 mA was utilized.

Evaluating sensory

Sensory evaluation is the examination of the sensorial qualities of an item that has been exposed to a treatment like the light procedures [28]. The study is used to improve the quality and advancement of foods, as well as to decide the approval of the item by the consumer. The sensory test was performed after treatment with EB and GR of Brazil nut not inoculated under similar conditions to those used for the trial tests; the samples were placed on the white plastic dishes with 3-digit daze codes and exhibited to a board of 30 untrained people (15 men and 15 women, age 25 to 50 years).

Participants received their samples in an irregular presentation and responded to the inquiries. The questions were designed to approach the participants in order to classify each physical characteristic, distributing scores of smell, taste, and texture. Between tests, the mouth was cleaned with water. To test worthiness, the participants evaluated each sample as indicated by a 9-point decedent scale (from 1, detest very, to 9, as incredibly); a score of 5 was viewed as satisfactory [28].

Results and Discussion

The irradiation energy eliminated *A. flavus* in the Brazil nut tests at in all dosages tried (Table 3). The comparative results were explained by different authors who watched a decrease in the development of fungi after treatment with light, as in the case of *A. flavus*; and *Fusarium verticillioides* in maize, and *Alternaria alternata* in sunflower, rice, maize, and wheat. In different investigations, GR has also been proved to be exceptionally successful against organisms that cause waste of cashew nuts, for example, *Aspergillus*, *Cladosporium*, *Penicillium*, *Curvularia*, and *Emericella*, with reduction in these fungal masses depending on the radiation portion connected and the underlying contagious burden.

chain of occasions that lead to organic changes. Then again, the indirect impact comprises radiation associating with water, creating free radicals that can harm DNA. Light additionally decreased the aflatoxin concentrations in the Brazil nut tests examined. Nonirradiated samples (control gathering) limited, on average, 4.75 mg/kg AFB1 (100%), while the concentration of this poison in illuminated samples was specifically corresponding to the radiation portion (Table 3). EB illumination at dosages of 5 and 10 kGy decreased aflatoxin levels by 53.32% (2.21 mg/kg) and 65.66% (1.63 mg/kg), separately, while GR at similar portions diminished toxin levels by 70.61% (1.39 mg/kg) and 84.15% (0.75 mg/kg) as compared to sum in the control samples. The numerical investigation demonstrated that all radiation treatments were successful in lessening Brazil nut aflatoxin ($P < 0.05$). The adequacy of EB and GR light has been exhibited by Shahbazi, *et al.* [29] in maize samples. Temcharoen and Thilly [30] watched decreases in aflatoxin dimensions of 75 and 100% in shelled nut samples through the use of doses of 1 and 10 kGy, correspondingly.

The aw estimations of the 50 samples analyzed ranged from 0.44 to 0.59 (mean of 0.48) before incubation and from 0.78 to 0.88 (mean of 0.80) after incubation, being compatible with the results presented by Reis, *et al.* (2012) in their analysis. The aw is a vital factor for fungal development and impacts the period of usability of items. It was exhibited that an aw esteem higher than 0.70 in Brazil nut tests can increase fungal growth and aflatoxin creation. During the illumination procedure, aw levels are identified explicitly with contagious development and aflatoxin deprivation. The aw additionally assumes a crucial role in the destruction of AFB1 by illumination, since radiolysis of water prompts the development of profoundly receptive free radicals. These radicals can promptly attack AFB1 at the terminal furan ring, making products with lower biological action. The high aw levels experienced in the present investigation may clarify the hindrance of fungal development and decrease in aflatoxin concentration. The biological and chemical impacts of EB are like those of GR, with photons exchanging their vitality to permeable materials by secondary electrons. Nevertheless, the two techniques contrast as far as infiltration. A few authors have indicated contrasts in penetration power among GR and EB.

The effects of sensory assessment for texture demonstrated that all groups were satisfactory (score 5). As to scent, tests treated

Group	Dose (kGy)	Mean <i>A. flavus</i> count (CFU/g 10 ²)	Amt of aflatoxin (mg/kg)			
			Mean*	SD	Min	Max
Control	0	32.9	4.75 A	4.87	0.77	19.82
GR	5	ND	1.39 B	2.55	0.86	8.94
	10	ND	0.75 B	1.71	0.81	6.75
EB	5	ND	2.21 B	3.33	1.01	15.14
	10	ND	1.63 B	2.87	0.95	14.54

Table 3: Mean *A. flavus* counts and aflatoxin levels following treatment of Brazil nuts with GR or EB.

GR: Gamma Radiation; EB: Electron Beam; ND: Not Detected; Min: Minimum Level; Max: Maximum Level.

*Means followed by the same letter are not significantly different by Tukey’s test ($P > 0.05$).

There are two components by which radiation can influence cells. These two components are normally called direct and indirect impacts. As for its direct impact, radiation executes the cells through ionization and excitation of nucleic acids, setting off a

with all doses were viewed as adequate. Tactile assessment of the taste demonstrated that just GR (5 kGy) – treated samples were approved as acceptable. There were critical contrasts between the qualities for the control groups and every single exploratory group ($P < 0.05$), except for GR at a 5-kGy dose, for taste and EB at a 10-kGy dose for texture. Their qualities demonstrated minimal distinction from the qualities for the control samples (Table 4). In the findings, in spite of the factual contrast intangible qualities between the control and the treatment used, Brazil nuts lighted with a GR portion of 5 kGy were viewed as acceptable.

Parameter	Sensory score for				
	Control group	Group dosed with 5 kGy of		Group dosed with 10 kGy of	
		EB	GR	EB	GR
Odor	7.5 A	5.5 B	5.6 B	5.9 B	5.8 B
Taste	7.2 A	3.9 B	5.8 A	3.8 B	4.8 B
Texture	7.5 A	5.6 B	5.6 B	6.3 A	6.1 B

Table 4: Mean results of sensorial evaluation of Brazil nut samples treated with GR and EB.

GR: Gamma Radiation; EB: Electron Beam. To test acceptability, 30 panelists evaluated each sample according to a 9-point hedonic scale (from 1, dislike extremely, to 9, like extremely). A score of 5 was considered acceptable. The control, GR, and EB groups each consisted of 50 g of Brazil nuts.

*Means followed by the same letter are not significantly different by Tukey’s test ($P > 0.05$).

Studies have stated the impacts of light at doses extending from 5 to 10 kGy on fungal development and inactivation of mycotoxins in foods; however, investigations of sensory assessment with these dosages are deficient. In the present examination, both illumination forms at 5-and 10-kGy portions demonstrated proficiency in *A. flavus* and elimination of aflatoxin. The treatments in GR and EB provoked adjustments in the sensory qualities of samples with the doses utilized in the study; nevertheless, Brazil nut samples illuminated with a GR dose of 5 kGy were viewed as suitable.

To test acceptability, 30 panellists evaluated each sample according to a 9-point hedonic scale (from 1, dislike extremely, to 9, like extremely). A score of 5 was considered acceptable. The control, GR, and EB groups each consisted of 50g of Brazil nuts.

Means followed by the same letter are not significantly different by Tukey’s test ($P > 0.05$).

EB and GR efficiently lowered aflatoxins and *A. flavus* in the samples of Brazil nuts. Furthermore, GR was more efficient in minimizing the aflatoxins due to the intensive penetration of GR. The illumination treatments encouraged changes in sensory features of Brazil nuts samples. However, the samples that were illuminated with GR using a dosage of 5 kGy were acceptable by the specialists [31-35].

Conclusion

The methodology proposal here for the elimination of aflatoxins in the food industry is proven to be used in more than 50 countries, being validated by them. Our results that prove the effect of radiation, which acts directly or indirectly on fungi, more specifically on the RNA and DNA molecules, in this way, it is proved that irradiation acts by inhibiting the reproduction and growth of fungi. In addition, the elimination of aflatoxins was total in food.

Based on our results, we observed the need for further research, and the formation of a commission to standardize a safe dose of radioactive material to be used for the treatment of this type of food product. This standardization can be based on the laws in force in different countries and approved by the World Health Organization (WHO), United Nations Food and Agriculture Organization (FAO) and International Atomic Energy Agency (IAEA). It is also suggested the standardization and standardization of seals to be inserted in the packages that indicate the absence of aflatoxins in the products.

Conflict of Interest

There is no conflict of interest.

Bibliography

1. Assunção E., et al. "Effects of gamma and electron beam radiation on Brazil nuts artificially inoculated with *Aspergillus flavus*". *Journal of Food Protection* 78.7 (2015): 1397-1401.
2. Baquião AC., et al. "Mycoflora and mycotoxins in field samples of Brazil nuts". *Food Control* 28 (2012): 224-229.
3. Pitt JI and Hocking AD. "Fungi and food spoilage". Blackie Academic Professional, London (2009).

4. Reis TA., et al. "Mycobiota and mycotoxins in Brazil nut samples from different states of the Brazilian Amazon region". *International Journal of Food Microbiology* 159 (2012): 61-68.
5. World Health Organization (WHO). 3.2.1.1. Moisture content and temperature. In Mycotoxins. Environmental health criteria 11. World Health Organization, Geneva (1979).
6. Inamura PY., et al. "Mediate gamma radiation effects on some packaged food items". *Radiation Physics and Chemistry* 81 (2012): 1144-1146.
7. Pennacchio HL. "Castanha-of-Brasil proposta de preço minimum 2006/2007". *Magazine* 1 (2007): 124-127.
8. Tonini H. "Manejo de produtos florestais não madeireiros na amazonia - (castanheira-do-brasil) resultados de pesquisa". *Boletim de pesquisa e desenvolvimento, Embrapa* 2 (2008): 39.
9. Baquião AC. "Fungi and mycotoxins in nuts the Brazil, the harvest storage". 2012. 141 f. Thesis (Doctorate in Microbiology) - Institute of Biomedical Sciences, University of São Paulo, São Paulo (2012).
10. Souza MLD and Menezes HCD. "Processing of Brazil nut and meal and cassava flour: quality parameters". *Food Science and Technology* 24.1 (2004): 120-128.
11. Brazil. Ministry of Agriculture, Livestock and Supply. Scenario Brazil-Brazil 2000-2010 exports. Presentation sectoral chamber of fruit growing, Ministry of Agriculture, Livestock and Supply. Brasilia, (2010): 15.
12. Gonçalves JFDC., et al. "Primary metabolism components of seeds from Brazilian Amazon tree species". *Brazilian Journal of Plant Physiology* 14.2 (2002): 139-142.
13. Freitas JB. "Composição química de nozes e sementes comestíveis e sua relação com a nutrição e saúde". *Revista de Nutrição* 23.2 (2010): 269-279.
14. Coutinho VF., et al. "Supplementation with brazil nuts (CP, *Bertholletia excelsa*, H.B.K.) in capoeira players on selenium (Se) concentration and glutathione peroxidase activity (GSH-PX, E.C.1.11.1.9)". In: Trace elements in man and animal. Springer, Pt. 2, New York, (2002): 405-406.
15. Lacey J and Magan N. "Fungi in cereal grains: their occurrence and water and temperature relationships". In: Chelowski, J. (ed.) *Cereal grain: mycotoxins, fungi and quality in drying and storage*. Amsterdam: Elsevier (1991): 77-118.
16. Xavier JG., et al. "Equine leukoencephalomalacia: report of five cases". *Brazilian Journal of Veterinary Research and Animal Science* 28 (1991): 185-189.
17. Taniwaki MH and Silva N. "Fungos em alimentos: ocorrência e detecção". *Campinas: Núcleo de Microbiologia do ITAL* (2001): 82.
18. CAST - Council for Agricultural Science Technology. "Mycotoxins Risks in plant, animal, human systems". Task Force Report, Ames, Iowa, 139 (2003): 1-199.
19. Christensen CM and Sauer DB. "Microflora". In: *Storage of Cereal Grains and their Products*, 3rd ed., ed. C. M. Christensen, (1982): 219-240.
20. ANSCI - Department of Animal Science. "Aflatoxins: occurrence and health risks". *Plants poisonous to livestock*. Cornell University.
21. Blount WP. "Turkey "X" Disease". *Journal of the British Turkey Federation* 9.52 (1961): 52-61.
22. Lancaster MC. "Toxicity associated with certain samples of groundnuts". *Nature* 192 (1961): 1095.
23. Nordion. "The history of food irradiation". (2012): 5.
24. Tsai D. "Application of electron beam radiation as a sterilizing agent microorganisms in peaty substrate". 2006. 119 f. Dissertation (Masters in Nuclear Technology) - Institute of Energy and Nuclear Research, University of São Paulo, São Paulo (2006).
25. Ferreira-Castro FL. "Interaction between toxigenic fungi (*Aspergillus flavus* and *Fusarium verticillioides*) And weevils (*Sitophilus Sitophilus*) in corn grain samples". 2011 Thesis (Doctorate in Microbiology) - Institute of Biomedical Sciences, University of São Paulo, São Paulo (2011).
26. Borrelly SI. "Tratamento de esgoto sanitário com uso de acelerador de elétrons". 1995. 104 f. Dissertation (Masters in Nuclear Technology) - Institute of Energy and Nuclear Research, University of São Paulo, São Paulo (1995).
27. Scussel VM., et al. "Effect of oxygen-reducing atmospheres on the safety of packaged shelled brazil nuts during storage". *International Journal of Analytical Chemistry* 2011 (2011): 1-9.
28. Rodrigues FT., et al. "A sensory evaluation of irradiated cookies made from flaxseed meal". *Radiation Physics and Chemistry* 81 (2012): 1157-1159.

29. Shahbazi HR., *et al.* "Effects of gamma and electron-beam irradiation on aflatoxin B1 content of corn grain". *Animal Science Journal* 3 (2010): 56-61.
30. Temcharoen P and Thilly WG. "Removal of aflatoxin b1 toxicity but not mutagenicity by 1 megarad gamma radiation of peanut meal". *Journal of Food safety* 4 (1982): 199-205.
31. Bennet JW. "Aspergillus: a primer for the novice". *Medical Mycology* 47.s1 (2009): S5-S12.
32. Chourasia R. "Effect of temperature, relative humidity and light on aflatoxin b1 production in neem and datura seeds". *International Journal of Pharmacognosy* 29 (1991): 197-202.
33. Jelinek CF. "Distribution of mycotoxin - An analysis of world-wide commodities data, including data from FAO/WHO/UNEP food contamination monitoring programme". In: International Conference on mycotoxins, 1987, Bangkok, thailand. Anais... bangkok, thailand (1987).
34. Lindsey DL and Turner RB. "Inhibition of growth of aspergillus flavus and trichoderma viride by peanut embryos". *Mycopathologia* 55 (1975): 149-152.
35. Ramakrishna N., *et al.* "Effect of surface sterization, fumigation and gamma irradiation on the microflora and germination of barley seeds". *International Journal of Food Microbiology* 13 (1991): 47-54.

Assets from publication with us

- Prompt Acknowledgement after receiving the article
- Thorough Double blinded peer review
- Rapid Publication
- Issue of Publication Certificate
- High visibility of your Published work

Website: www.actascientific.com/

Submit Article: www.actascientific.com/submission.php

Email us: editor@actascientific.com

Contact us: +91 9182824667