GET Journal of Biosecurity and One Health

GET Global Emerging Pathogens Treatment Consortium

GET JOURNAL OF BIOSECURITY & ONE HEALTH

PUBLISHED BY:

Global Emerging Pathogens Treatment Consortium

JOURNAL WEBSITE

www.getjournal.org

The Effect of Storage on the Proximate, Mineral Composition, and Mycoflora of Sundried Pigeon Pea (*Cajanus cajan*) Seeds

Adebayo FO[1*] and Fagbohun ED[2]

[1]Global Emerging Pathogens Treatment Consortium (GET)
[2]Department of Microbiology, Ekiti State University, Ado-Ekiti, Ekiti State, Nigeria

*Corresponding Author: Faith Omolade Adebayo

ORCID ID: 0009-0007-2726-5753

ABSTRACT
This study investigates the impact of a twenty-week storage period on the proximate composition, mineral content, and mycoflora of sun-dried pigeon pea (*Cajanus cajan*) seeds. The storage conditions were observed to promote fungal contamination, leading to qualitative and quantitative changes in the seeds' nutritional and mineral composition. Fungal species isolated included *Aspergillus niger, A. fumigatus, A. flavus, A. oryzae, Fusarium oxysporum, F. solani, F. moniliforme, Rhizopus stolonifer, Alternaria alternata, and Penicillium notatum*. Proximate analysis (g/100g) showed significant increases in moisture content (9.86 to 13.13), ash (2.60 to 3.15), fat (1.25 to 3.17), and carbohydrate (0.22 to 10.56). However, fiber (7.75 to 4.61) and crude protein (76.74 to 68.83) content decreased, suggesting deterioration in the seeds' quality over time. Mineral analysis revealed reductions in sodium (3.49 to 2.47), potassium (21.26 to 15.03), calcium (6.02 to 3.43), magnesium (11.75 to 9.27), zinc (0.09 to 0.05), iron (0.28 to 0.21), copper (0.07 to 0.02), manganese (0.08 to 0.05), and phosphorus (19.02 to 12.03). The findings indicate that prolonged storage at room temperature facilitates mycoflora proliferation, leading to degradation in nutritional and mineral composition. This has significant implications for food security, as pigeon pea is a key protein source in many regions. The study underscores the importance of adopting improved storage techniques to minimize fungal contamination and nutrient loss, thereby enhancing the economic and nutritional value of stored pigeon pea seeds.

Keywords: Pigeon pea; *Cajanus cajan;* Storage effects; Proximate composition; Mineral composition; Mycoflora

Corresponding Author's Email: adebayofaithomolade@gmail.com

GET Journal of Biosecurity and One Health (2025) 4(1) 46-55.
DOI: 10.36108/GJOBOH/5202.40.0150

INTRODUCTION

Cajanus cajan (L.) Millsp., commonly known as pigeon pea, is a leguminous plant belonging to the family Leguminosae (Fabaceae). It is widely known as red gram in English and Arhar or Tur dhal in India [1]. Its seeds vary in size, shape, and color, typically round or oval, ranging from white and greyish to red and brown, with a small white hilum. Production of *Cajanus cajan* in Africa is estimated to account for about 9.3% of global production, which is comparatively small next to the 77.6% contribution from India [2]. The crop is cultivated throughout the tropics and subtropics [3,4] and represents approximately 5% of total world legume production [5], with India remaining the dominant producer [6]. Globally, pigeon pea is cultivated on about 4.67 million hectares, of which 3.30 million hectares are in India alone. In Asia, other significant producers include Myanmar (570,000 ha), China (150,000 ha), and Nepal (20,988 ha) are also important pigeon pea-producing countries. In Africa, significant production occurs in Tanzania, Kenya, Malawi, Uganda, and Mozambique [7]. India remains the world's largest producer and consumer of pigeon pea [8]. In Nigeria, pigeon pea is produced in considerable quantities, particularly in the southeastern, southern, and central regions of the country [8]. Compared with other grain legumes, pigeon pea ranks only sixth in area and production [9,10]. It contains protein (23.77%), Fat (1.1%), and crude fibre (7.49%). Its seed consists of 85% cotyledons, 14% seed coat, and 1% embryo. Cotyledons are rich in carbohydrates (66.7%), while a major proportion (about 50%) of seed protein is located in the embryo. It has a good amount of cysteine and methionine [11]. It provides iron, sulphur, calcium, potassium, manganese, thiamine, niacin, and riboflavin. [7] It is also known to contain genistein and daidzein [3]. Despite its nutritional and economic importance, post-harvest storage poses a major challenge to the quality and safety of pigeon pea and other legumes. During storage, seeds are highly susceptible to microbial contamination, especially by storage fungi such as *Aspergillus, Fusarium, Penicillium, and Rhizopus* species. These spoilage organisms cause biochemical deterioration, leading to loss of nutritional value, seed discoloration, rancidity, and off-flavors [12–14]. More importantly, some of these fungi produce mycotoxins, such as aflatoxins and fumonisins, which have been linked to serious health issues, including hepatotoxicity, nephrotoxicity, immunosuppression, and carcinogenesis in humans and animals [15,16]. Previous studies on other legumes such as cowpea (*Vigna unguiculata*), soybean (*Glycine max*), and groundnut (*Arachis hypogaea*) have documented similar patterns of fungal invasion and nutrient degradation during prolonged storage [17–19]. However, there is limited information on the mycoflora associated with sun-dried pigeon pea during storage, particularly under tropical conditions where temperature and humidity promote rapid fungal proliferation. Therefore, this study was designed to evaluate the changes in proximate composition, mineral content, and mycoflora of sun-dried pigeon pea (*C. cajan*) stored for 20 weeks. The findings aim to provide insight into the nutritional and safety implications of long-term storage and to contribute to strategies for maintaining the quality of pigeon pea during storage.

METHODOLOGY

Collection of Samples

The seeds of *Cajanus cajan* were obtained from Oja Oba market in Ado-Ekiti, Ekiti State, Nigeria. The seeds were sundried for one week. The dried pigeon pea was further stored in an insect-free container, carefully labelled, and kept in the well-ventilated laboratory for 20 weeks in the Department of Microbiology, Ekiti State University, Ado-Ekiti, Nigeria. The seeds were examined monthly for changes in mycoflora, minerals, and nutrient composition during storage.

Proximate Analysis

A sample of the stored pigeon pea was analysed using the AOAC [20] standard procedures for ash, crude fibre, moisture content, Fat, protein, and carbohydrates. All determinations were in duplicates.

Mineral Analysis

The AOAC standard method [20] was used. The minerals were analyzed from a solution obtained by first dry-washing, as follows: about 1.5 g of the flour sample was placed in a petri dish and heated gently on a Bunsen burner in a fume cupboard until the charred mass ceased to emit smoke. It was transferred to a muffle furnace at 550 °C. Heating was continued until all the carbon was burnt away. The dish and ash were transferred to a desiccator to cool, after which 0.1 M HNO^3 solution (10 mL) was added to the crucible to break up the ash. It was then filtered through acid-washed No. 43 Whatmann filter paper into 100 mL with the same dilute acid solution. Mineral concentrations (Ca, Fe, P) were determined by flame Atomic Absorption Spectrophotometry using a Buck Atomic Absorption Spectrometer (Buck Scientific, Model 200A/200, Inc., East Norwalk, Connecticut, U.S.A). Hollow cathode lamps specific for each element were used, and an air–acetylene flame served as the energy source. Calibration curves were prepared using certified standard solutions of each metal, and the sample concentrations (mg/100 g) were calculated from the calibration curves based on their absorbance values [20].

GET Journal of Biosecurity and One Health (2025) 4(1) 46-55.
DOI: 10.36108/GJOBOH/5202.40.0150

Isolation of Fungi from Stored Sundried Pigeon Pea (*Cajanus canjan*)
The mycoflora associated with *Cajanus cajan* during storage were isolated using three methods: direct plating, dilution plate, and washing.

Direct Plating Method
Sun-dried seeds of *Cajanus cajan* were randomly selected and surface-sterilized individually with 70% ethanol for 1 minute, followed by 2 rinses with sterile distilled water to remove residual ethanol. The seeds were then aseptically plated onto potato dextrose agar (PDA) plates using a sterile spatula and incubated at room temperature (25 ± 2°C) for 5–7 days. Emerging fungal colonies were subcultured onto fresh PDA plates to obtain pure cultures through successive hyphal tip transfers. The purified cultures were examined microscopically for morphological features, including hyphae, conidia, and fruiting bodies, to identify the common fungi present [21].

The emerging fungal colonies were subcultured onto fresh PDA plates to obtain pure cultures by successive hyphal tip transfers. The purified cultures were examined microscopically for morphological features, including hyphae, conidia, and fruiting bodies, to identify the common fungi present [22].

Dilution Plate Method
The dilution plate method described by Fagbohun and Ogundahunsi [23] was employed. One gram of the stored *Cajanus cajan* sample was aseptically placed in a test tube containing 9 ml of sterile distilled water and thoroughly shaken to obtain a uniform suspension. One milliliter of this mixture was serially diluted in sterile distilled water to achieve the desired dilution levels. Aliquots of milliliter each from the 10^{-2} and 10^{-3} dilutions were introduced into sterile molten PDA plates supplemented with 100 µg/ml chloramphenicol to inhibit bacterial growth. The plates were gently swirled for uniform mixing, allowed to solidify, and incubated at ambient temperature (25 ± 2°C) for 5 to 7 days. Emerging fungal colonies were observed for morphological features, including fruiting bodies and hyphal characteristics. Hyphal tips of each distinct fungus were transferred successively onto fresh PDA plates until pure cultures were obtained.

Washing Method
The washing method described by Fagbohun [22] was employed. One gram of *Cajanus cajan* seeds was weighed using a digital weighing balance and dispensed into a test tube containing 9 ml of sterile distilled water. The mixture was shaken thoroughly for about 2 minutes to dislodge surface contaminants and fungal spores. Using a sterile pipette, 1 milliliter of the resulting suspension was aseptically introduced into sterile Petri dishes containing molten PDA. The suspension was evenly spread on the agar surface using a sterile glass spreader to ensure uniform distribution of spores. The plates were allowed to solidify and then incubated at 28°C for 5 to 7 days. After incubation, the plates were examined for visible fungal growth. Distinct colonies were subcultured onto fresh PDA plates until pure cultures were obtained for further identification.

Identification of Mycoflora
Pure cultures of each fungus isolated from *Cajanus cajan* seeds were prepared on nutrient media, and slides were made for microscopic observation. Cultural and morphological characteristics of each isolate, including colony colour, texture, hyphae septation, spore structure (sporangia/conidia), spore arrangement (single or in chains), spore shape, and size, were examined and recorded as identification criteria [24,25]. The isolates were further subcultured to obtain pure strains, and detailed macroscopic and microscopic features (e.g., conidiophore structure, conidial morphology) were used to assign fungal genera and/or species based on contemporary identification schemes [26,27].

Needle Mount Preparation Method
Fragments from the sporulating surface of pure fungal cultures (taken from mid colony to colony edge) were gently teased in a drop of 70% ethanol on a sterile glass slide using a sterile needle. A drop of lactophenol cotton blue stain was added, a coverslip applied, and the preparation examined under 10× and 40× objective lenses [28].

Slide Culture Technique
From a plate approximately 2 mm deep, a 1 cm2 PDA was cut and placed on a sterile glass slide. Each isolated fungus was inoculated into the four vertical sides using a sterile needle. A sterile coverslip was placed on it, overlapping the medium on all sides. The preparation was placed on a suitable support in a petri dish containing blotting paper soaked in 20% glycerol in water. The preparation was kept moist at 28°C until adequate growth was observed. The medium was removed, and the fungus adhering to both coverslip and slide was examined [28]. A drop of alcohol was added, followed by a drop of lactophenol blue, and the preparation was covered and examined under the microscope's low-power objective.

Statistical Analysis
For each sample, the overall Mean, standard deviation (SD), and standard error of the Mean (SEM) were calculated. All analyses were performed in triplicate, and results are expressed as mean ± SD. Data from proximate, mineral, and mycofloral analyses were processed using

Microsoft Excel (version 2019). To compare differences in mean values across storage period treatments, a one-way analysis of variance (ANOVA) was applied, followed by post hoc comparisons of means using a multiple-comparison test. Differences were considered statistically significant at $p < 0.05$, and mean values bearing different superscript letters within the same column or row were considered significantly different at the 5% level [29].

DISCCUSSION

The fungi isolated from stored sun-dried *Cajanus cajan* seeds using the direct plating, washing, and dilution methods are summarized in Tables 1–3. The results revealed a progressive increase in the diversity and frequency of fungal species with increasing storage duration. At the initial stage of storage, only a few fungal species, such as *Aspergillus niger*, *A. fumigatus*, and *A. flavus*, were isolated; however, by the 20th week, ten fungal species belonging to five genera were identified across the three methods. The summary of the results of fungi isolated from stored sundried pigeon pea (*Cajanus cajan*) seeds is shown in Figure 1. Ten (10) fungi comprising five genera were isolated: *Rhizopus* sp., *Fusarium* sp., *Penicillium* sp., *Aspergillus* sp., and *Alternaria* sp. This finding is in agreement with the report of Chaudhari et al. [30], who reported the presence of *Aspergillus niger*, *A. flavus, Fusarium oxysporum, F. moniliforme, F. udum, Drechslera* sp., *Curvularia lunata*, *Rhizoctonia* sp., and *Alternaria alternata* in pigeon pea (*Cajanus cajan*) seeds. A similar observation was reported by Ingle et al. [31], who isolated multiple fungal species, including *Alternaria alternata, Aspergillus flavus, A. niger, Chaetomium globosum, Cladosporium herbarum, Curvularia lunata, Fusarium oxysporum, F. moniliforme, F. roseum, Pythium sp., Rhizoctonia solani, Rhizopus stolonifer, Botrytis cinerea, Macrophomina phaseolina, Penicillium notatum, and Phytophthora cinnamomi* from pigeon pea seeds using agar plate and blotter methods. It was noted that while most fungi were recovered by all methods, *Alternaria alternata* was not isolated using the direct plating method. The predominant fungi belonged to the genus *Aspergillus*, which are commonly classified as storage fungi capable of growth at lower seed moisture content [32]. Among *Aspergillus* species, A. *flavus* was the most frequently isolated. Similar results have been reported in other legume and groundnut studies [33–35]. Several studies have shown that stored legumes and cereal grains are highly susceptible to *Aspergillus flavus* colonization, with fungal loads often increasing as storage duration progresses [36]. This contrasts with the findings of Amsalu et al. [37], who reported a decline in fungal populations during storage of faba beans. Most isolated fungi are known to be surface contaminants of seeds and contribute to decay during storage. Fungi recovered via washing methods are typically surface colonizers, while those recovered by any method may include both field and storage fungi [38]. Stored products remain vulnerable to microbial attack under favourable conditions such as moderate temperatures and high humidity [39]. These fungi also produce mycotoxins as secondary metabolites during growth, harvest, and storage [40].

Table 1: Summary of fungi isolated from stored pigeon pea (C*ajanus cajan)* using the direct plating method

Weeks of Storage	Fungal Species
Freshly prepared samples	A, C
4	A, B, C, F, H
8	A, B, C, D, F, H, I
12	A, B, C, D, F, G, H, I
16	A, B, C, D, F, G, H, I, J
20	A, B, C, D, F, G, H, I, J

Legend: A – *Aspergillus niger*, B – *Aspergillus fumigatus*, C – *Aspergillus flavus*, D – *Rhizopus stolonifer*, E – *Alternaria alternata*, F – *Fusarium oxysporum*, G – *Fusarium solani*, H – *Aspergillus oryzae*, I – *Fusarium moniliforme*, J – *Penicillium notatum*

GET Journal of Biosecurity and One Health (2025) 4(1) 46-55.
DOI: 10.36108/GJOBOH/5202.40.0150

Table 2: Summary of fungi isolated from stored pigeon pea (C*ajanus cajan)* using the washing method

Weeks of Storage	Fungal Species
Freshly prepared samples	A, B
4	A, D, E, G, H
8	A, D, E, G, H, I
12	A, B, C, D, E, G, H, I
16	A, B, C, D, E, G, H, I, J
20	A, B, C, D, E, F, G, H, I, J

Legend: A – *Aspergillus niger*, B – *Aspergillus fumigatus*, C – *Aspergillus flavus*, D – *Rhizopus stolonifer*, E – *Alternaria alternata*, F – *Fusarium oxysporum*, G – *Fusarium solani*, H – *Aspergillus oryzae*, I – *Fusarium moniliforme*, J – *Penicillium notatum*

Table 3: Summary of fungi isolated from stored pigeon pea (*Cajanus cajan)* using the dilution method

Weeks of Storage	Fungal Species
Freshly prepared samples	B, C
4	A, B, C, D, F, H
8	A, B, C, D, E, F, H, I
12	A, B, C, D, E, F, G, H, I
16	A, B, C, D, E, F, G, H, I, J
20	A, B, C, D, E, F, G, H, I, J

Legend: A – *Aspergillus niger*, B – *Aspergillus fumigatus*, C – *Aspergillus flavus*, D – *Rhizopus stolonifer*, E – *Alternaria alternata*, F – *Fusarium oxysporum*, G – *Fusarium solani*, H – *Aspergillus oryzae*, I – *Fusarium moniliforme*, J – *Penicillium notatum*

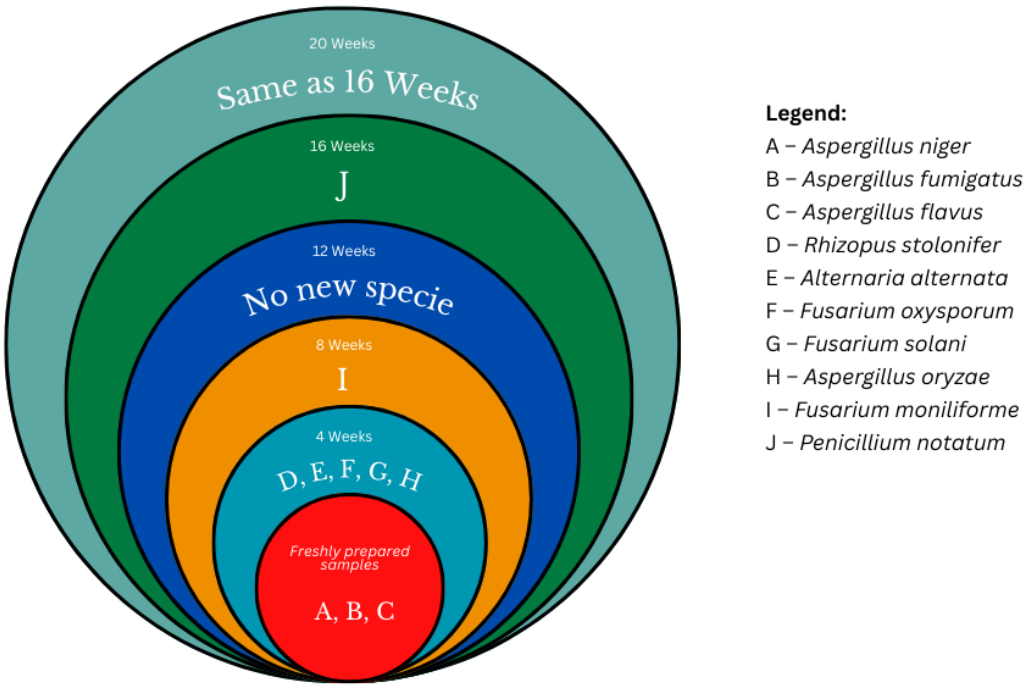

Figure 1: Fungi Isolated from Stored Pigeon Pea (*Cajanus cajan*) Across Different Storage Periods

GET Journal of Biosecurity and One Health (2025) 4(1) 46-55.
DOI: 10.36108/GJOBOH/5202.40.0150

Table 4: Summary of results of proximate analysis of pigeon pea (*Cajanus cajan)* during 20 weeks of storage (g/100 g)

Weeks of Storage	**MC**	**Ash**	**Fat**	**CF**	**CP**	**CHO**
Freshly prepared	9.86 ± 0.02	2.60 ± 0.01	1.25 ± 0.01	7.75 ± 0.04	76.74 ± 0.05	0.22 ± 0.01
4	10.62 ± 0.03	2.89 ± 0.02	1.29 ± 0.02	6.50 ± 0.03	76.27 ± 0.04	0.63 ± 0.02
8	11.34 ± 0.03	2.97 ± 0.02	1.69 ± 0.02	5.96 ± 0.03	73.39 ± 0.03	1.51 ± 0.02
12	12.15 ± 0.03	3.14 ± 0.02	2.27 ± 0.02	5.28 ± 0.03	71.90 ± 0.03	4.86 ± 0.02
16	12.47 ± 0.03	3.14 ± 0.02	3.16 ± 0.02	4.82 ± 0.03	70.30 ± 0.03	8.00 ± 0.02
20	13.13 ± 0.02	3.15 ± 0.02	3.17 ± 0.02	4.61 ± 0.02	68.83 ± 0.02	10.56 ± 0.03

Legend: MC – Moisture content, Ash – Total ash, Fat – Crude Fat, CF – Crude fibre, CP – Crude protein, CHO – Carbohydrate.
Values are expressed as Mean ± Standard Deviation (SD) of triplicate determinations.

GET Journal of Biosecurity and One Health (2025) 4(1) 46-55.
DOI: 10.36108/GJOBOH/5202.40.0150

Table 5: Summary of results of mineral analysis of pigeon pea (*Cajanus cajan)* during 20 weeks of storage (mg/100 g)

Weeks of Storage	**Na (mg/100g)**	**Ca**	**K**	**P**	**Mg**	**Mn**	**Fe**	**Zn**	**Pb**	**Cd**	**Ni**	**Cr**	**Cu**
Freshly prepared	3.49 ± 0.35	6.02 ± 1.09	21.26 ± 2.56	19.02 ± 2.85	11.75 ± 0.95	0.08 ± 0.01	0.28 ± 0.03	0.09 ± 0.01	0.00 ± 0.00	ND	ND	0.00 ± 0.00	0.07 ± 0.02
4 weeks	3.00 ± 0.35	5.52 ± 1.09	19.05 ± 2.56	11.85 ± 2.85	10.83 ± 0.95	0.08 ± 0.01	0.28 ± 0.03	0.07 ± 0.01	0.00 ± 0.00	ND	0.00 ± 0.00	0.00 ± 0.00	0.03 ± 0.02
8 weeks	2.66 ± 0.35	4.86 ± 1.09	16.85 ± 2.56	12.58 ± 2.85	10.77 ± 0.95	0.07 ± 0.01	0.23 ± 0.03	0.06 ± 0.01	ND	ND	ND	0.00 ± 0.00	0.05 ± 0.02
12 weeks	2.56 ± 0.35	4.03 ± 1.09	15.23 ± 2.56	11.85 ± 2.85	9.87 ± 0.95	0.06 ± 0.01	0.22 ± 0.03	0.06 ± 0.01	ND	ND	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02
16 weeks	2.62 ± 0.35	3.45 ± 1.09	15.17 ± 2.56	12.07 ± 2.85	9.46 ± 0.95	0.05 ± 0.01	0.22 ± 0.03	0.06 ± 0.01	ND	ND	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02
20 weeks	2.47 ± 0.35	3.43 ± 1.09	15.03 ± 2.56	12.03 ± 2.85	9.27 ± 0.95	0.05 ± 0.01	0.21 ± 0.03	0.05 ± 0.01	0.00 ± 0.00	ND	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02

Legend: Na – Sodium, Ca – Calcium, K – Potassium, P – Phosphorus, Mg – Magnesium, Mn – Manganese, Fe – Iron, Zn – Zinc, Cu – Copper, Pb – Lead, Cd – Cadmium, Ni – Nickel, Cr – Chromium, *ND – Not detected.

Values are expressed as Mean ± Standard Deviation (SD) of triplicate determinations.

The result of the proximate analysis of sundried pigeon pea (*Cajanus cajan*) seeds during 20 weeks of storage is shown in Table 4. There were changes in the nutritional value compared to the freshly pigeon pea seeds. This is due to fungal activity that caused changes during storage of the product [41]. After twenty weeks of storage, the moisture content, ash, Fat, and carbohydrate increased to 13.13, 3.15, 3.17, and 10.56 (g/100 g), respectively. On the other hand, the fibre and crude protein contents were decreased to 4.61 and 68.83 (g/100 g), respectively, during storage. These findings agreed with the report of Fagbohun and Ogundahunsi [23] who reported an increase in the moisture content (7.21-12.22), carbohydrate content (0.20-2.01), and a decrease in the fiber content (7.75 - 4.61) of stored sundried *Citrullus lanatus* Thunberg (Melon) Seeds. Fagbohun and Lawal [42] also reported an increase in the moisture content of sundried soybean (Glycine max.) from 6.80–8.34. High moisture levels enhance microbial growth, thereby shortening the shelf life of stored products [43]. An increase in moisture content and changes in other nutrient parameters during storage may be attributed to the activity of storage-associated mycoflora, which metabolize seed nutrients over time [41]. Fungal contamination and seed deterioration have been documented in legumes and other crops during prolonged storage, leading to associated losses in nutrient quality and the potential for mycotoxin production [42]. Changes in proximate composition, for instance, fluctuations in carbohydrate, Fat, fibre, and mineral content, have been observed in stored grains and pulses, highlighting that storage fungi can significantly alter seed chemistry [43]. For example, a recent study on seed-borne mycoflora of legumes under different storage conditions reported significant reductions in protein and mineral levels over time, correlated with increased fungal colonization [44]. Mineral analyses of stored pulses have shown that essential minerals such as K, Mg, Ca, Na, and trace elements decline as storage duration increases, likely due to fungal metabolism and associated biochemical transformations [45]. Comparable findings have been reported across different legumes and storage contexts, underscoring the need for rigorous storage hygiene and regular seed health monitoring to preserve nutritional quality [46].

CONCLUSION

Pigeon pea (*Cajanus cajan*) seeds are of great economic and nutritional importance. To preserve their quality and safety, they must be stored under controlled environmental conditions to prevent fungal contamination and nutrient deterioration. This study revealed that sun-dried pigeon pea seeds were contaminated with various fungal species during the 20-week storage period, leading to statistically significant ($p < 0.05$) reductions in key nutritional parameters, including crude protein, fiber, and mineral contents, while moisture, Fat, and carbohydrate levels increased. The isolated fungi, predominantly species of *Aspergillus*, *Fusarium*, *Rhizopus*, *Penicillium*, and *Alternaria*, utilized the stored seeds as substrates, accelerating spoilage and nutrient degradation. These seed-borne fungi are known producers of spores and mycotoxins, which pose serious health risks to humans and animals when consumed through contaminated grains. Therefore, maintaining optimal storage conditions (low humidity, moderate temperature, and adequate ventilation) and implementing effective post-harvest handling practices are critical to minimizing fungal proliferation, preserving nutritional quality, and preventing economic losses associated with pigeon pea storage.

REFERENCES

[1]. Murali R, Anand G, Jolly GE, Jincy M. Impact and Response of Drought Stress in Pigeon Pea (*Cajanus cajan* L.): a Review. Agric Rev. 2025;46(4):555-565. doi:10.18805/ag.R-2713.

[2]. Indian Council of Agricultural Research (ICAR) – Indian Institute of Pulses Research (IIPR). Pigeon Pea Crop. Kanpur: ICAR-IIPR; 2020. https://www.icar-iipr.org.in/pigeonpea-crop/ [Accessed 05 December, 2025]

[3]. Saxena KB, Choudhary AK, Saxena RK, Varshney RK. Breeding Pigeonpea Cultivars for Intercropping: Synthesis and Strategies. Breed Sci. 2018;68(2):159–167.

[4]. Heuzé V, Tran G, Boudon A, Lebas F. Pigeon pea (*Cajanus cajan*) Forage. Feedipedia (INRAE, CIRAD, AFZ, FAO). 2022. Available from: https://www.feedipedia.org/node/22444. [Accessed 05 December, 2025]

[5]. FAO. FAOSTAT Statistical Database: Crops and Livestock Products – Pigeon pea. Rome: Food and Agriculture Organization of the United Nations; 2020. Available from: https://www.fao.org/faostat. [Accessed 05 December, 2025]

[6]. Singh N, Gupta S, Srivastava RK. Trends in Pigeon pea Production in India: A Regional Analysis. Indian J Agric Res. 2019;53(3):375–381.

[7]. Loko YL, Adjatin A, Dansi A, Vodouhè R, Sanni A. On-farm Management and Participatory Evaluation of Pigeon pea Diversity across Agroecological Zones of Benin. J Ethnobiol Ethnomed. 2020;16:24. doi.org/10.1186/s13002-020-00378-0

[8]. Ajeigbe HA, Adigun JA, Singh BB. Productivity and Utilization Trends of Pigeon pea in Nigeria: A Review. J Agric Environ Sci. 2017;6(4):55–63.

[9]. Nix A, Paull CA, Colgrave M. The Flavonoid Profile of Pigeon pea (*Cajanus cajan*): a Review. SpringerPlus. 2015;4:125. DOI 10.1186/s40064-015-0906-x

[10]. Saxena KB, Kumar RV, Sultana R. Genetic Improvement of Pigeon Pea - a Global Perspective. Plant Breed. 2018;137(4):451–469.

[11]. Haji A, Teka TA, Urugo MM. Nutritional Composition and Bioactive Compounds of Pigeon pea (*Cajanus cajan* L.) Seeds. Legume Sci. 2024;6(2):e233.

[12]. Atanda SA, Pessu PO, Agoda S, Isong IU, Adekalu OA, Echendu MA, Falade TC. Fungi and Mycotoxins in Stored Foods: Implications for Food Safety and Health. Adv J Microbiol Res. 2020;14(5):1–10.

[13]. Kortei NK, Awuah RB, Okyere YA, Kyei-Baffour M. Storage Fungi and Mycotoxins Associated with Cowpea (*Vigna unguiculata* L. Walp) Seeds from Smallholder Farmers in South Africa. Phytoparasitica. 2025;53(2). https://doi.org/10.1007/s12600-025-01267-6

[14]. Kunz A, Winkler J, Warth B. Growth and Toxin Production of *Phomopsis* and *Aspergillus* Species under Different Storage Conditions in a pea (*Pisum sativum*) Model System. Mycotoxin Res. 2021;37(3):259–273.

[15]. Zain ME. Impact of Mycotoxins on Humans and Animals. J Saudi Chem Soc. 2015;19(3):275–283. doi:10.1016/j.jscs.2014.05.002.

[16]. Battilani P, Toscano P, Van der Fels-Klerx HJ, et al. Aflatoxin B1 Contamination in Maize in Europe Increases due to Climate Change. Sci Rep. 2016;6:24328. doi:10.1038/srep24328.

[17]. Onwuka GI, Ogunbanwo ST. Fungal Contamination and Aflatoxin Occurrence in Cowpea (*Vigna unguiculata*) Stored Under Different Conditions in Nigeria. Food Control. 2019;100:129–137. doi:10.1016/j.foodcont.2019.01.019.

[18]. Ogunka-Nnoka CC, Nwosu C, Okeke BC. Post-harvest Fungal Invasion in Stored Soybean (Glycine max) and Associated Mycotoxin Risk. J Stored Prod Res. 2021;92:101826. doi:10.1016/j.jspr.2021.101826.

[19]. Kpodo FM, Wiafe-Kwagyan M, Ntiamoah E. Assessment of Mycotoxin Contamination in Stored Groundnut (*Arachis hypogaea*) from Selected Markets in Ghana. Toxins (Basel). 2022;14(4):262. doi:10.3390/toxins14040262.

[20]. AOAC International. Official Methods of Analysis of AOAC INTERNATIONAL. 22nd ed. Oxford University Press; 2023.

[21]. Chaudhari A, Sharma H, Sharma JK, Mulji J. Seed-borne Fungal Pathogens Associated with Pigeon pea Seeds and their Effect on Seed Quality Parameters. Indian J Plant Protect. 2020;45(3). https://epubs.icar.org.in/index.php/IJPP/article/view/105368

[22]. Fagbohun ED, Durojaiye AO, Popoola OA. Mycoflora, Proximate Composition and Mineral Analysis During the Storage of Smoked Dried Crayfish (*Penaeus natialis*) – Shrimps. Asian J Fish Aquat Res. 2019;4(1):1-8.

[23]. Fagbohun ED, Ogundahunsi AS. Effects of Storage on Nutritional, Mineral Composition, and Mycoflora of Stored Sundried *Citrullus lanatus* Thunberg (melon) Seeds. Int J Biochem Res Rev. 2019;28(4):1-7.

[24]. Rathod SG, Ingle ST, Gurav NV, Chaure NV. Identification and Detection of Seed-Borne Mycoflora of Chickpea using Different Seed Testing Methods. J Plant Dis Sci. 2024;19(1):61–66.

[25]. Hay A, Abebe W, Tadesse G. Mycoflora Associated with Soybean Seeds: Incidence and Detection Methods. Int J Plant Sci. *2023.*

[26]. Singh D, Sharma P, Kumar R. Seed-Borne Fungal Diversity in Legumes and Morphological Identification. Plant Pathol J. 2022;38(2):121–132. doi:10.5423/PPJ.OA.03.2022.0031.

[27]. Chatterjee S, Saha A, Das S. Modern Approaches to Seed-Borne Fungi Identification: Morphology and Molecular Tools. J Mycol Plant Pathol. 2021;51(4):345–356. doi:10.5958/0974-7542.2021.00045.7.

[28]. Agu KC, Chidozie CP. An Improved Slide Culture Technique for the Microscopic Identification of Fungal Species. Int J Trend Sci Res Dev. 2021;6(1):243–254.

[29]. Quirk TJ, Quirk MH, Horton HF. Excel 2016 for Environmental Sciences Statistics: A Guide to Solving Practical Problems. Cham (Switzerland): Springer; 2017.

[30]. Chaudhari A, Sharma H, Jehani M, Sharma JK. Seed Mycoflora Associated with Pigeon pea (*Cajanus cajan* L.) Millsp., their Significance and the Management. J Pure Appl Microbiol. 2017;11(1):567-575.

[31]. Ingle HD, Ingle ST, Gurav NV, Chaure NV. Detection and Diagnosis of Seedborne Mycoflora Associated with Pigeon Pea. J Plant Dis Sci. 2024;19(1):22–26.

[32]. Kumar A, Pathak H, Bhadauria S, et al. Aflatoxin Contamination in Food Crops: Causes, Detection, and Management - a Review. Food Prod

Process Nutr. 2021;3:17. https://doi.org/10.1186/s43014-021-00064-y

[33]. Gebisa LA, Wodimagegnehu sadik GT. Identification of Microflora Associated with Groundnut Seeds and its Impact on Seed Germination. Adv Agric Food Sci. 2024.

[34]. Amsalu N, Ararsa Leta, Amin Mohammed Y, et al. Seedborne Mycoflora of Faba bean (*Vicia fabae* L.) and Control of Selected Fungi. Agri Res Rev. 2023;9(6):e17291.

[35]. Yadav S, Sharma R, Singh P. Occurrence of *Aspergillus* and *Fusarium* Species in Stored Legumes and their Mycotoxin Production. Int J Food Microbiol. 2022;367:109544. doi:10.1016/j.ijfoodmicro.2022.109544.

[36]. Pitt JI, Hocking AD. *Fungi and Food Spoilage*. 3rd ed. New York: Springer; 2009.

[37]. Amsalu N, Ararsa Leta, Amin Mohammed Y, et al. Storage Fungi Dynamics and Mycoflora Changes in Faba Bean Seeds Over Time. Agri Res Rev. 2023;9(6):e17292.

[38]. Chandra S, Singh RK. Seed-Borne Fungal Pathogens in Legumes: Field vs Storage Fungi. J Plant Pathol Microbiol. 2021;12:555875. doi:10.4172/2157-7471.1000555.

[39]. Tadesse D, Alemayehu G, Bekele T. Effect of Storage Conditions on Microbial Contamination of Legume Seeds in Ethiopia. Int J Food Sci. 2020;2020:8894123. doi:10.1155/2020/8894123.

[40]. Kumar A, Pathak H, Bhadauria S, et al. Mycotoxins in Legumes: Occurrence, Health Effects, and Management Strategies. Food Prod Process Nutr. 2021;3:17. doi:10.1186/s43014-021-00064-y.

[41]. Ingle HD, Ingle ST, Gurav NV, Chaure NV. Detection and Diagnosis of Seedborne Mycoflora Associated with Pigeon pea. J Plant Dis Sci. 2024; 19(1):22–26. Doi:10.48165/jpds.2024.1901.04.

[42]. Amza I, Nazari M, Faraji M. Seed Borne Fungi; Food Spoilage, Health Risk and Control Strategies - a review. Food Sci Qual Manag. 2021;114:1–12.

[43]. Megersa B, Seid H, Esatu A, Debeli G. Effects of Seed Dressing and Storage Duration on Different Seed Quality Parameters of Faba Bean (*Vicia faba* L.). Reports. 2023;3(3):23–28. doi:10.11648/j.reports.20230303.11.

[44]. Amsalu N, Ararsa L, Amin MY, Tahir M. Seedborne Mycoflora of Faba Bean and Evaluation of Plant Extracts & *Trichoderma* Species against Selected Fungi. Heliyon. 2023;9(6):e17291. doi:10.1016/j.heliyon.2023.e17291.

[45]. Ajadi I, Olahan GS. Mycoflora Associated with Groundnut Seeds Collected from Three Senatorial Districts of Kwara State, Nigeria. J Jewel Sci Res. 2023;8(1-2):62–69. https://journals.fukashere.edu.ng/index.php/jjsr/article/view/190

[46]. Elaigwu M, Oluma HOA, Onekutu A. Storage Mycoflora in Sesame Seed Production in Benue State, Nigeria. J Bot Res. 2021;3(4):22–28. doi:10.30564/jbr.v3i4.3482.

[47]. Samal I, Bhoi TK, Raj MN, Majhi PK, Murmu S, Pradhan AK, et al. Underutilized Legumes: Nutrient Status and Advanced Breeding Approaches for Qualitative and Quantitative Enhancement. Front Nutr. 2023;10:1110750. doi:10.3389/fnut.2023.1110750.