UV Treatment on Inactivation of Fungal Contamination of Sri Lankan Black Pepper (*Piper nigrum*) seed and powder

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**Abstract:** A study was carried out to test the efficacy of UV-C treatment to decontaminate fungal species in black pepper. After preliminary tests carried out using a UV-C drum, a canopy type UV-C sterilization unit was designed and fabricated. Black pepper seed and powder samples were given 10, 15, and 20 minutes UV-C treatments. Results showed that UV-C canopy is effective on inactivation of yeast and moulds in black pepper seeds complying the international microbiological standard requirements for spices. Improvement of UV-C Design modification have been carrying out to obtain the potential inactivation level by UV-C light for black pepper powder and improve the inactivation level of bacteria in black pepper seed and powder.

**Keywords:** Black pepper, UV-C canopy.

1. **Introduction**

Black pepper is mainly used to augment the distinctive flavour, aroma, colour and fragrance of food. Sri Lankan black pepper have been popularized throughout the world. Black pepper has a high potential of microbial contamination including fungus during preand post operations till final market packing [1]. Contamination of spices is leading to cause some food spoilage and food borne illnesses [2]. Fungus can degrade the quality of spices since the growth of mycelia on spices, sporulation and production of mycotoxins [3, 4]. Sterilization of black pepper is essential because in many cases black pepper added to food as a raw material without cooking.

Thermal sterilization is achieved with high intensity of heat and still this is the main technique that used in the spice processing industry including black pepper [5]. High heat in thermal sterilization may change quality of spices in cases of sensory, functional and nutritional aspects due to thermal degradation. These thermal losses can be prevented by introducing non-thermal sterilization method [6] such like UV-C (200-280 nm) [7]. UV-light photon at 253 nm has energy of 470 kJ/mol [8]. Absorption of UV light by microorganisms can undergo changes
within their cells due to UV photons, photochemical changes and exposure to heat causing temperature build up within microbes ultimately to inactivate them [9]. Therefore, the objective of this study was to test the UV-C system developed to inactivate fungal species contaminated in black pepper seeds and black pepper powder and identify modifications necessary for to optimize the system to obtain potential decontamination level of UV-C light.

2. Material and Methodology

2.1 Experimental equipment

Preliminary UV-C treatments were carried for black pepper spowder using a UV-C rotating drum fabricated to obtain initial UV-C microbial inactivation data. Based on these data an experimental UV unit (canopy type) was designed and fabricated. It consisted with ten UV lights (Tepro (China) Co. LTD, GPH287T5L/HO; 27W) which were mounted on two side walls and underneath of the canopy. UV unit was fixed on a conveyor for one-way linear movement of black pepper samples. Two adjustable metallic doors for closing the chamber in both sides to prevent the outside exposure of UV radiation during operation.

![Figure 1. UV-C Rotating drum](image)

![Figure 2. UV-C Canopy with 10 UV-C bulbs and coupled to a conveyer belt](image)

2.2. Sample collection and UV-C treatment

Packed black pepper powder and pepper seeds samples were randomly collected representing different brands from different super markets in Sri Lanka. Collected ten pepper seeds and pepper powder samples aseptically were mixed to prepare one composite sample and were divided in to three equal samples (replicates). Each sample was divided into one for control (20g) sample and other three samples (20g) for treatments. 20g of black pepper seed and powder samples were loaded in a sterilized metal tray and were placed on the conveyor. Sample was UV-C treated by 10, 15 and 20 minutes.
2.3 Microbiological study

Treated each sample was transferred to a strainer bag aseptically for microbial analysis. Yeast and mould count tests were carried out following standards of International Standard Organization (ISO 6887-1:1991 and ISO 21527-2:2008). Colony forming units per gram (cfu/g) was taken after counting of the colonies on the plates. Received data was verified with microbiological criteria in recommendation of international microbiological standard to assess the acceptable level of spices. UV dosage was calculated using following formula in each treatment times.

\[
\text{Dose} = \frac{\text{time (s) } \times \text{ output (W)}}{\text{area (cm}^2\text{)}}
\]

3. Results and Discussion

Absorbed UV-light energy by microbial cells induces photo-physical, photochemical, and/or photo-thermal effects that necessary for inactivation of pathogenic microorganisms. Base pairs of DNA absorbs UV light efficiently due to their aromatic ring structure. Pyrimidines; thymine (DNA), cytosine (DNA and RNA), and uracil (RNA) are strong absorbers of photons in the ultraviolet range and generate photoproducts which results in microbial inactivation. In photochemical oxidation DNA bonds affected by the UV-C light and split due to shifting of electrons, breaks bonds of nitrogenous bases thereby deplete growth of cells. Pyrimidine dimer formation in DNA strands by DNA cross linking between thiamine and cytosine indicate main effect to inhibit transcription and cell replication thereby delay or prevent reproduction and ending microbial cell depth. The degree of inactivation by UV radiation is directly related to the UV dose, exposure time. The dosages required for inactivation are pathogen specific. Nature of food source, food density, UV absorptivity and surface properties of food material are the variables of germicidal effects of UV-C light [9]-[11].

The International Microbiological Standard recommended limits for contaminants in spices are in the range of \(10^1\) to \(10^3\) cfu/g for yeast and mould. In the preliminary study of black pepper seed using UV-C drum it (Figure 1) was able to reduce Yeast and Mould counts from \(1.5 \times 10^4\) to \(7.72 \times 10^3\) after giving 01 hour UV-C treatment with total power of 24 W using 3 UV bulbs. Again it was able to reduce Yeast and Mould count in black pepper powder from \(5.6 \times 10^4\) to \(6.81 \times 10^3\) after giving 30 minutes UV-C treatment with the total power of 81 W using 03 UV – C bulbs. Results indicated that even under increased wattage the inactivation level is not complying the international microbial standard for spices. In the study carried by using UV-C canopy with increased number of UV –C bulbs into 10 numbers and thereby with increased total wattage of 270 and under 20 minutes’ treatment duration (Table 1) it was able to obtain complying of yeast and mould count in pepper seeds with the international microbial standard for spices.
Table 1. Yeast and Mould counts of pepper seeds and pepper powder

<table>
<thead>
<tr>
<th>UV-C treatment time (Min.)</th>
<th>UV dose (8J/cm²)</th>
<th>Y &amp; M counts (cfu/g) of pepper seeds</th>
<th>Y &amp; M counts (cfu/g) of pepper powder</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>-</td>
<td>5.45x10⁴</td>
<td>1.86x10⁴</td>
</tr>
<tr>
<td>10</td>
<td>10.</td>
<td>3.18x10⁵</td>
<td>1.64x10⁵</td>
</tr>
<tr>
<td>15</td>
<td>16.2</td>
<td>1.36x10⁵</td>
<td>1.14x10⁵</td>
</tr>
<tr>
<td>20</td>
<td>21.6</td>
<td>4.54x10⁵</td>
<td>6.81x10⁵</td>
</tr>
</tbody>
</table>

Results indicated that there is minimal shadow and surface properties effect in case of sterilization intensity of pepper seed. This inactivation enhanced by rolling of pepper seeds during conveyer movement. More inactivation can be obtained if there is a fluidizing mechanisms for higher exposure to direct UV-C light. In case of black pepper powder, 0.05-0.4 log reduction was detected in yeast and mould count compared to pepper seed. This low log reduction indicated that small particles of black pepper powder coincides each other and that prevents direct exposure of UV-C to the targeted microbial cells. It is more suitable if it is coupled with cyclone type air flow to give direct exposure of UV-C radiation to each powder particle. As the UV light penetrates through the food material, its intensity decays along a distance of beneath the food surface. The residual amount of light is dissipated as heat and transferred to the inner layers through conduction. Because of these reasons UV light is more effective for surface sterilization of spices. Shadow effect exerted by microbial lamella and high microbial load can be minimized by applying good Agricultural Practices (GAP) and Good Manufacturing Practices (GMP) during pre and post-harvest operation.

4. Conclusion

Selected brands of spices obtained from different markets of Sri Lanka are contaminated by yeasts and moulds. UV-C treatment given by using UV-C canopy is effective on inactivation of yeast and moulds in black pepper seeds. Modification of UV canopy type sterilizing machine is necessary to increase the inactivation level for black pepper powder.
References


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Conflict of interest: NIL

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