BioLink : Jurnal Biologi Lingkungan, Industri dan Kesehatan, Vol. 10 (1) August (2023)
ISSN: 2356- 458X (print) ISSN: 2597-5269 (online)
DOI: 10.31289/biolink.v10i1.9391

BioLink
Jurnal Biologi Lingkungan, Industri, Kesehatan

Available online http://ojs.uma.ac.id/index.php/biolink

MICROBIOLOGICAL QUALITY OF CAYENNE PEPPER (Capsicum frutescens L.) IN MALANG TRADITIONAL MARKET

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Submitted : 10-04-2023; Reviewed : 31-05-2023; Accepted : 27-07--2023

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Abstract

Cayenne pepper is consumed as a food spice because of its color, spicy taste, aroma and complete nutritional value. Cayenne pepper sold in traditional markets is not separated between whole cayenne pepper and broken cayenne pepper. This study aims to: 1) calculate the ALT of mold colonies on whole and damaged cayenne peppers, 2) identify contaminant mold species found in intact and damaged cayenne peppers. Samples of whole cayenne pepper and damaged cayenne pepper were isolated by grinding 10 grams of cayenne pepper, it was then inoculated on Potato Dextrose Agar (PDA) medium and incubated at 25oC for 7 x 24 hours. Then the ALT of mold colonies was calculated on whole and damaged cayenne pepper. Then, each isolated mold contaminating colony's morphology and microscopic characteristics were described. Finally, each contaminant mold isolate was identified. Furthermore, the study unpacked that: 1) The ALT of contaminant mold colonies on whole cayenne pepper was 1.0x103 colonies/g, while the average ALT of contaminant mold colonies on damaged cayenne pepper was 2.1x105 colonies/g, 2) There were 8 species of mold found in cayenne pepper namely Mucor racemosus Fres, Aspergillus niger van Tieghem, Penicillium griseofulvum Dierckx, Penicillium frequentans Westing, Penicillium citrinum Thom, Penicillium nalgiovense Laxa, Penicillium digitatum Sacc and Penicillium variabile Sopp.

Keywords: *Capsicum frutescens L; contaminant molds; identification; Malang traditional market*

How to Cite: *Noor, A.F., Hastuti, U.S., & Lukiati, B. (2023). Microbiological Quality Of Cayenne Pepper (Capsicum frutescens L). In Malang Traditional Market. BioLink: Jurnal Biologi Lingkungan, Industri dan Kesehatan, Vol. 10 (1): page. 1-7*

INTRODUCTION

Cayenne pepper (*Capsicum frutescens* L.) is preferred and consumed by Indonesian people. It is used as a spice or flavoring in food because of its attractive color, taste, spiciness, aroma and complete nutritional value (Sofiarani & Ambarwati, 2020). Cayenne pepper contains nutrients, including protein, fat, carbohydrates, potassium, phosphorus, iron, vitamin A, vitamin B1, vitamin C and water (Nurhayati & Sari, 2020). Cayenne pepper is widely used by the community since it has good nutrition for humans (Hartono, 2020).

Various kinds of food ingredients not only act as a source of nutrition for humans, but there are also some nutrients for microorganisms (Hastuti, 2010). The nutrients contained in cayenne pepper are also needed by the mold, so that cayenne pepper can be a substrate for the growth of contaminant molds. Cayenne pepper has weaknesses, such as it is easily damaged during pre-harvest and poor storage after harvest. Preharvest damage is caused by insect pests (Meilin, 2014). Damage to chilies is characterized by chilies with holes, brownish black spots and powder (Hastuti, 2010). Glodjinon's research et al., (2020) has identified contaminant molds in chili peppers in West Africa that produce aflatoxins.

The quality of cayenne pepper due to the presence of contaminant molds can be determined based on the standard total plate number (ALT) of mold colonies. Based on Regulations of the Food and Drug Supervisory Agency of the Republic of Indonesia (2019), the maximum ALT limit for mold and yeast colonies in spices is $2x10^4$ colonies/g. Identification of contaminant molds in cayenne pepper needs to be done to find out the species of mold that contaminate cayenne pepper. Several species of aflatoxin-producing mold contaminants include: *Aspergillus flavus, A.parasiticus, A. niger, Penicillium citrinum, P. frequentans, P. fellutanum* (Hastuti, 2010; Ferdiansyah et al., 2020). Aflatoxins are dangerous to health since they are carcinogenic (cancer trigger), hepatotoxic (can poison the liver), and mutagenic (trigger gene mutations) for humans, mammals and poultry (Sumantri, 2022). The type of mycotoxin that is most widely known and gets special attention from experts in the health sector is: aflatoxin (Hastuti, 2006).

The aims of the study were 1) to calculate the ALT of mold colonies in whole and damaged cayenne peppers, and 2) to identify contaminant mold

species found in intact and damaged cayenne peppers.

RESEARCH METHODS

The research was conducted from July 2022 to September 2022 at the Microbiology Laboratory, Department of Biology, State University of Malang. Materials used in this study include: Cayenne pepper (*Capsicum frutescens* L.), PDA medium (Potato Dextrose Agar), 70% alcohol, aluminum foil, 0.1% peptone solution, 95% alcohol, lactophenol solution, lactophenol cotton blue solution, tissue, cotton, and distilled water.

Procedure

1. **Isolation of Contaminant Mold in Cayenne Pepper**

All cayenne pepper from the traditional market in Malang were collected. Cayenne pepper that has been collected was separated into 2, namely whole cayenne pepper and broken cayenne pepper. Whole cayenne pepper and broken cayenne pepper were weighed up to 10 grams, then mashed with mortalwith sterile mortar and pestle and dilution in 90 ml of 0.1% peptone aqueous solution to obtain a dilution level of 10^{-1}. Dilution was continued gradually until a solution with dilution levels of 10^{-2}, 10^{-3}, 10^{-4}, and 10^{-5} was obtained. Each of these solutions was incubated on the PDA plate medium as much as 0.1 ml, evenly distributed and incubated at 25^0C for 7 x 24 hours. The molds growing on the PDA plate media were coded and isolated on the PDA slanting medium.

2. **Identification of Contaminant Molds in Cayenne Pepper**

Identification of contaminant molds by macroscopic and microscopic methods. Macroscopic observation was done by observing the morphology of contaminating mold colonies. Each contaminant mold isolate was described with a description of the morphological characteristics of the colony. Microscopic observation was carried out using the slide culture method and observed for its microscopic characteristics. Identification of molds down to the species level used a mold identification key book.

3. **Calculating ALT of Mold Colonies**

How to calculate ALT of mold colonies using the formula:

$$x = yx\frac{1}{z}xm$$

(Modified from Fardiaz, 1993)

Example:
x = Total Plate Count (TPC)
y = Number of mold colonies per species
z = Dilution Rate
m = Suspended volume grown

RESULTS AND DISCUSSION

1. Total Plate Number (ALT) of Mold Colonies in Whole and Damaged Cayenne Pepper

The ALT testing technique for contaminating mold colonies from whole and damaged cayenne pepper samples was carried out by dilution of 10^{-1} to 10^{-5}. Calculation of ALT of contaminant mold colonies of whole and damaged cayenne pepper samples was carried out in 3 replications. The average ALT of contaminant mold colonies in whole and damaged chili pepper samples is shown in Table 1.

Table 1. Contaminant ALT of Mold Colonies in Whole Cayenne Pepper and Broken Cayenne Pepper

Test	ALT Contaminant Mold Colonies (colony/g)	
	Whole Cayenne Pepper	Broken Cayenne Pepper
1	1.0 x 103	3.4 x 105
2	1.0 x 103	4.6 x 103
3	1.0 x 103	2.9 x 105
Average	1.0 x 103	2.1 x 105

The average obtained from the ALT of contaminant mold colonies on whole cayenne pepper was 1.0 x 10^3 colonies/g, while the average The ALT of contaminant mold colonies on damaged cayenne pepper was 2.1 x 10^5 colonies/g. Based on the 2019 Regulations of the Food and Drug Supervisory Agency of the Republic of Indonesia (BPOM RI) 2019, the maximum ALT limit for mold and yeast colonies in spices is 2x10^4 colonies/g, so based on the results of the ALT calculation of mold colonies in Table 1, it shows that the number of contaminating mold colonies in Whole cayenne pepper was still suitable for consumption, while damaged cayenne pepper was not suitable for consumption. The ALT of contaminant mold colonies in damaged cayenne pepper was higher than intact cayenne pepper. It can be caused by damage found in damaged cayenne pepper. This damage makes it easier for contaminant mold spores and mycelium to grow both on the surface and penetrate the skin of cayenne pepper.

2. Identification of Contaminant Mold Species Identification Whole Cayenne Pepper and Broken Cayenne Pepper

Isolation and description of mold through observation of colony morphology and microscopic characteristics on samples of whole and damaged x, found 8 mold isolates, each of which was given a code, namely: A, B, C, D, E, F, G, and H. The results of the description of each contaminant mold isolate found were referred to in the books

"Fungi and Food Spoilage" (Pitt & Hocking., 2009) and "Introduction To Food-Borne Fungi" (Samson, et al., 2004). Contaminant mold species in cayenne pepper that have been identified are shown in Figure 1 below.

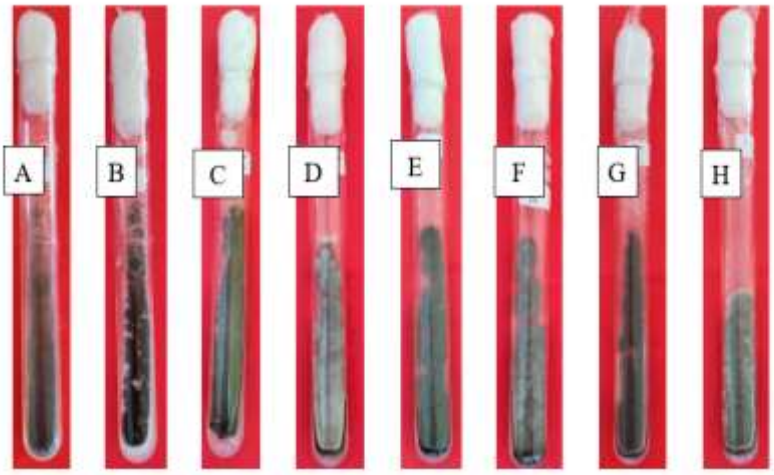

Figure 1. Each Contaminant Mold Isolate in Cayenne Pepper

Pictures of microscopic observation of mold contaminants can be seen in Figure 2.

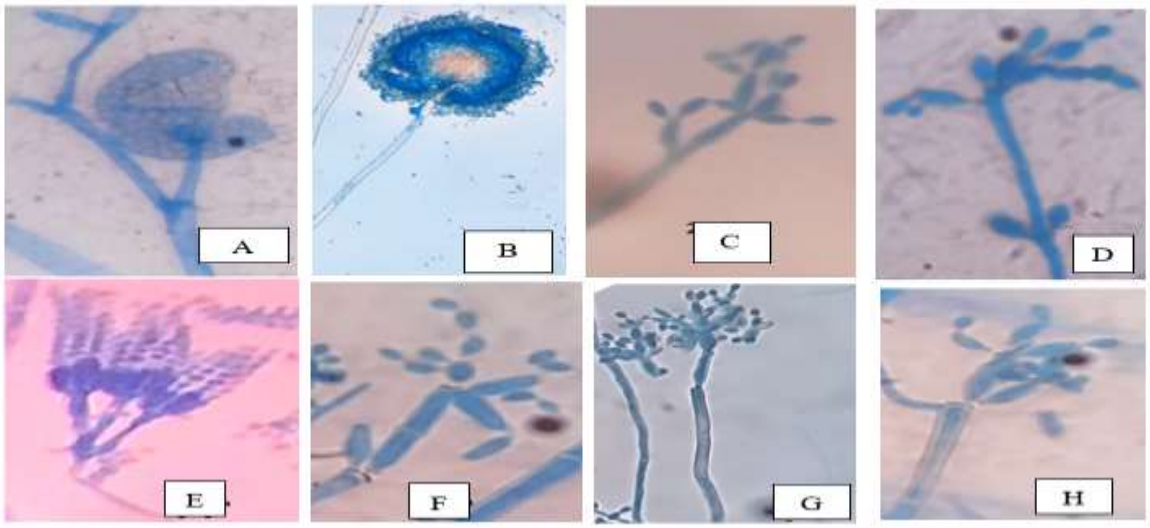

Figure 2. Microscopic Contaminant Mold in Cayenne Pepper

Information: A. *Mucor racemosusfres*.; B. *Aspergillus nigervan* Tieghem.; C. *Penicillium griseofulvum* Dierckx.; D. *Penicillium frequentans* Westing.; E. *Penicillium citrinum* Thom.; F. *Penicillium nalgiovense* Laxa.; G. *Penicillium digitatum* Sac.; H. *Penicillium variabile* Sopp.

Some species of contaminant mold can grow on both whole cayenne pepper and damaged cayenne pepper. Damage to cayenne pepper can occur before harvest which is caused by insect pest attacks, namely fruit flies (Suriana, 2019). It can be due to, among other things: 1) sellers do not separate whole and damaged cayenne peppers, so that the contaminant mold on the damaged cayenne pepper can contaminate whole cayenne pepper; 2) whole and damaged cayenne pepper both contain nutrients that support the growth of the mold. In damaged cayenne pepper, mold hyphae can penetrate the damaged cayenne pepper skin since it is fragile. Damage that occurs such as holes in the skin of cayenne pepper can make it easier for contaminant mold hyphae to penetrate into the cayenne pepper. Indonesia is a conducive condition for the growth of mycotoxin-producing molds (Anastasia & Widiastuti, 2015). In damaged cayenne pepper, *Aspergillus niger van Tieghem* was found. which is a species of mold that produces aflatoxin and ochratoxin which can cause aspergillosis and is hepatotoxic and carcinogenic (Handajani & Purwoko, 2008). Aflatoxin B1 is a mycotoxin that is harmful to the body, is hepatotoxic, teratogenic, mutagenic, and causes hepatitis, bleeding, immunosuppression and liver carcinoma (Min, et al. 2011). In addition, aflatoxin B1 has a melting point at 268–269 °C (Broto, 2018). Based on these properties, the process of cooking food, among others: by boiling, roasting and frying cannot decompose aflatoxin B1 that enters the body.

CONCLUSION

From the results of this study it can be concluded that the ALT of contaminant mold colonies on whole cayenne pepper was 1.0x103 colonies/g, while the average ALT of contaminant mold colonies on damaged cayenne pepper was 2.1x105 colonies/g. There were 8 species of mold found in cayenne pepper, namely: *Mucor racemosus Fres, Aspergillus niger van Tieghem, Penicillium griseofulvum Dierckx, Penicillium frequentans Westing, Penicillium citrinum Thom, Penicillium nalgiovense Laxa, Penicillium digitatum Sacc and Penicillium variabile Sopp*.

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