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Artikel

Identification *Aspergillus Sp.* on *Vigna Radiata, L.* at Andir Market, Bandung City

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A B S T R A C T

Vigna radiata is a carbohydrate food with a fairly high nutritional content, carbohydrate food has the components needed by a microorganism, one of which is mushrooms. Vigna radiata is also a food ingredient that is often found in traditional markets such as Andir Market. The humid market environment and open storage areas make it easier for Vigna radiata to be contaminated by fungi such as Aspergillus sp. Aspergillus sp is a fungus that is often found in a variety of habitats but is generally saprophytes in soil, feed and food products saved. Aspergillus also often contaminates grains, nuts and produce processed like Vigna radiata. Aspergillus sp produces a toxin called aflatoxin which this toxin is often found contaminating food. Aspergillus sp causes poisoning, respiratory system disorders such as asthma, and lung disease. Infections caused by Aspergillus are called Aspergillosis, this infection is one of the most common opportunistic infections that infect the lungs. This research was conducted in February 2021. The research was carried out in a bacteriology laboratory at Institute Kesehatan Rajawali. The aim of this study determines the types of Aspergillus sp which contaminate Vigna radiata sold at Andir Market, Bandung City. The research method used is descriptive by using the total population. This research was conducted using the sowing method and was observed macroscopically and microscopically. The results in this study are known to Aspergillus sp with a percentage that is Aspergillus fumigatus (40%). Aspergillus niger and Aspergillus terreus (20%) and there is 1 negative sample Aspergillus sp. In this study, it was concluded that there are types of Aspergillus sp in Vigna radiata sold at the Andir Market, Bandung City, the types are Aspergillus fumigatus, Aspergillus niger and Aspergillus terreus.

INTRODUCTION

Mushrooms belong to the lower class of plants that do not have substances green, the fungus acts as a saprophytic parasite. Mold grows more effectively in humid temperatures. The habitat of the fungus on land and in humid places has an optimal temperature ranging from 22ºC-35ºC, with a maximum temperature of 27 ^{0}C-29 ^{0}C and a minimum temperature <5ºC. Fungi also live on organisms or the remains of organisms in sea and freshwater. Fungi can also survive in an acidic environment (Smith, 2015).

Aspergillus sp including eukaryotic microorganisms is currently recognized as one of the few living things with a wide distribution area. Types of molds are common contaminants in various substrates in tropical and subtropical regions. Aspergillus Sp is often found in a variety of habitats, generally in soil, feed products and stored food. *Aspergillus sp* also often contaminates grains and legumes such as Vigna radiata(Utamiet al, 2012).

Aspergillus sp is a fungus that has cosmopolitan properties so this fungus has a very wide distribution area (Srikandi, 2015). Generally, fungi are shaped like threads, each sheet of thread is called a hypha and a group of hyphae is called mycelium. The diameter of the hyphae in fungi is generally 0.5-100µ, but each type of fungus has varying sizes (Syaifuddin, 2017). The vesicles will form brown, green or black coloured chains called conidia (Himedia, 2015). *Aspergillus* can also cause neonatal infections (Cloherty, 2012). Therefore, humans can experience infections in the lower respiratory system, because the conidia are inhaled by humans (Sullivan, 2011).

Aspergillus sp causes poisoning, respiratory system disorders such as asthma, and lung disease. Infections caused by Aspergillus. Also known as Aspergillosis, this infection is one of the most common opportunistic infections that infect the lungs. Symptoms of this infection vary depending on the type of disease. The most common symptoms are symptoms of poisoning caused by toxic metabolism that comes from ingested mushrooms into the body. Ingested poisons can cause fatal disease and some of these poisons have cancer-causing carcinogenic properties. *Aspergillus Sp* produces a toxin called aflatoxin which this toxin is often found contaminating foodstuffs (Smith, 2015).

The fungus Aspergillus sp can produce some mycotoxins. One of which is aflatoxin. Aflatoxin itself is a class of mycotoxin compounds, toxins derived from known fungi lethal and carcinogenic to humans and animals. The high content of aflatoxins in food can cause poisoning (Saputri, 2018). Aflatoxins are harmful to human and animal health because carcinogenic, mutagenic, teratogenic and immunosuppressive (Mobeen et al, 2011).

Alpha toxin poisoning causes death, as many as 500,000 cases every year in the world experience alpha-toxin poisoning. As in the case in Kenya where 125 people were reported to have been

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poisoned (Taufik, 2018). This case is the case with the largest fatalities in the world (Kusumaningrum, 2010). In 2017, 153 cases were reported throughout Indonesia poisoning in 25 provinces in Indonesia. Based on the report from the Food and Drug Supervisory Agency or BPOM, the most food poisoning occurred in West Java province, amounting to 32 cases (21%) (Syaifuddin, 2017).

Food is a source of nutrition for humans, but besides that, food is also a factor in the growth of microorganisms, especially fungi. Foodstuffs that have the composition required by a microorganism will be contaminated by the growth of microorganisms in the food (Noverita, 2009). The growth of microorganisms in foodstuffs can result in adverse physical or chemical changes so that these foodstuffs are unfit for consumption and contaminated foodstuffs will experience spoilage (Amalia, 2013).

Carbohydrate food is an ingredient that has the components needed by a microorganism. These foodstuffs are often found in traditional markets or supermarkets in large quantities, including green beans, peanuts, corn and rice. Foodstuffs that are most often overgrown by microorganisms are fungi. One of the factors that strongly support the growth of fungi in this food is a humid storage area (Nasir, 2017).

Vigna radiata is a carbohydrate food with a fairly high nutritional content and a complete composition. One of the nutritional content found in *Vigna radiata* is Carbohydrate: 62.9 g, Protein: 22.2 g, Fiber: 4.1g and Water: 10 mg. This composition is a growth factor in fungi so that it can contaminate food (Mustakim, 2014). To find out the type of *Aspergillus sp* that contaminates *Vigna radiata* will be observed macroscopically and microscopically, based on this problem, the researchers are interested in conducting research with the title Identification *Aspergillus Sp* on *Vigna radiata* sat Andir Market, Bandung City

METHOD

The research design uses a quantitative descriptive method. The purpose of this research is to find out the existence of Aspergillus sp on Vigna radiata sold in the market. This research was conducted in February 2021. The research was carried out in a bacteriology laboratory at Institute Kesehatan Rajawali.

Tools and materials used in this research Autoclave, Newsprint, Beaker glass, Laminar air flow, Bunsen, mortar pestle, Petri dish, Micropipette, Cover glass, Microscope, Erlenmeyer, Analytical balance, Hot plate, Oven, Incubator, Object glass, Aquades, *Vigna radiata*, Lactophenol Cotton Blue (LPCB), Potato Dextrose Agar (PDA).

The research variables in this study are the independent variable and the dependent variable. The independent variable in this study was *Aspergillus Sp*. The dependent variable of this study was *Vigna radiata*. The population in the study is *Vigna radiata* sellers. The sample used in the study is *Vigna radiata*.

The sampling technique used in This research is total sampling, the technique of taking samples using the entire population (Sugiyono, 2017).

Data processing was carried out during the research in the laboratory, data were obtained from *Vigna radiata* contaminated by *Aspergillus Sp* isolated on PDA (*merck*) media and incubated for 3-5 days, then observed macroscopically and microscopically. Then the data is processed based on the table using Ms Excel.
The data in this study were analyzed using the formula :

$$P = \frac{f}{N} \times 100\ \% \quad (1)$$

P : Percentage

N : Number of samples identified

F : Frequency of contaminated Vigna radiata *Aspergillus sp*

RESULTS AND DISCUSSION

This research begins with taking samples sold at the Andir Market, Bandung City, then inoculation using PDA media and then incubating for 3-5 days after which macroscopic and microscopic observations are carried out, and microscopic observations are stained using LPCB.

Macroscopic observations were carried out to determine the types of fungi that contaminate *Vigna radiata* sold at Andir Market, Bandung City, including the colour, shape, and edges of the colonies growing on the media can be seen in **Table 1**.

Table 1. Macroscopic Observations

No	Colony Color	Colony Form	Edge of the Colony	Information
1	Black	Semi-round	Uneven	*Aspergillus niger*
2	Beige	Round	Uneven	*Aspergillus terreus*
3	White	Fibre	Uneven	*Negative Aspergillus*
4	Dark green	Velvet	Uneven	*Aspergillus fumigatus*
5	Dark green	Velvet	Uneven	*Aspergillus fumigatus*

Microscopic observations were carried out using staining LPCB to clarify parts of fungi such as conidia and hyphae found in fungi can be seen in **Table 2.**

Table 2. Microscopic Observations

No	Type *Aspergillus*	Conidia	Hyphae
1	*Aspergillus niger*	Globose shaped with brown dark, walled smooth as well contain hyaline	Have a wall thickness and don't disagree
2	*Aspergillus terreus*	Have shape long column and also contains metula for support phialides and smooth walls which contains hyaline	Party with contain hyaline
3	*Aspergillus fumigatus*	Globose shaped and also contains hyaline	Hyphae septate and contain hyaline or called conidiospores

There is 4 contaminated *Vigna radiata Aspergillus sp*, where there are 3 types of *Aspergillus sp*, in the *Vigna radiata* sample 1 there is a type of *Aspergillus niger*, and in the *Vigna radiata* sample 2 there is a type of *Aspergillus terreus*, then in samples of Vigna radiata 4 and 5, there are types of *Aspergillus fumigatus*. In addition, there is 1 *Vigna radiata* that is not contaminated with *Aspergillus sp* but still contaminated with other types of fungi *Rhizopus sp* can be seen in **Table 3.**

The first stage in this study was carried out by sampling which is sold at the Andir Market, Bandung City, the sample used in this study is green beans. In this study, the total sampling technique was used according to Sugiyono (2017), the total sampling technique was carried out by taking the entire population available, at Andir Market, Bandung City, there are 5 green bean sellers, so the sample size used in this study is 5 samples of green beans.

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Table 3. Data on contaminated *Vigna radiata Aspergillus Sp*

No	Sample	Type *Aspergillus*			Information
		A.niger	*A.terreus*	*A.fumigatus*	
1	*Vigna radiata* 1	+	-	-	*Positive Aspergillus Sp*
2	*Vigna radiata* 2	-	+	-	*Positive Aspergillus Sp*
3	*Vigna radiata* 3	-	-	-	*Negative Aspergillus Sp*
4	*Vigna radiata* 4	-	-	+	*Positive Aspergillus Sp*
5	*Vigna radiata* 5	-	-	+	*Positive Aspergillus Sp*

The next stage after sampling is the stage of inoculation of *Vigna radiata* on PDA media with the sowing method or also called seedlings. According to Cappucino (2014), is a medium for fungal growth which has a low pH value ranging from 4.5 to 5.6 with this pH value, PDA media can inhibit the growth of a bacterium where a neutral temperature is needed with a pH value of 7.0 and a temperature of 7.0. Optimum fungal growth is between 25ºC to 30ºC(Murwani, 2015). As explained in Octavia's research (2017), PDA media has a composition such as potatoes as much as 4 grams serves as a carbon source, then 20 grams of dextrose functions as sugar and energy and 15 grams of agar components which are useful for compacting the media.

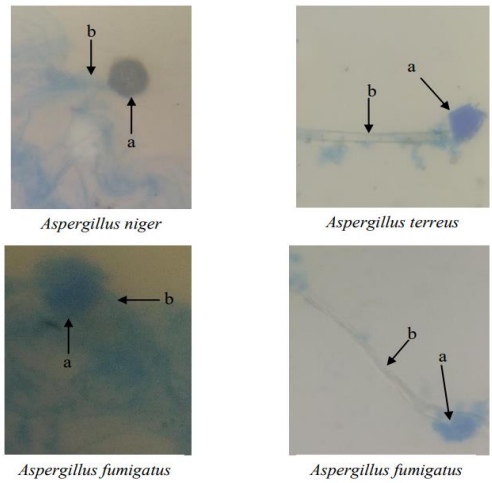

Figure 1. Microscopy With LPCB Staining: a) Conidia; b) Hifa

The stage after making media and inoculation is incubation for 3-5 days, according to Pujiati (2018), there are several factors that can affect the growth of fungi such as humidity where molds such as Aspergilluscan live with a humidity level of 80%, the next factor is the substrate or nutrients in the media then the storage temperature of the mushrooms where for this type of mushroom *Aspergillus sp* can live at room temperature around 25ºC to 30ºC and the last factor is the degree of acidity or commonly referred to as pH. In general, fungi grow at low pH values ranging from 4.5 to 5.5.

Vigna radiata that have been inoculated on PDA media and incubated for 3-5 days will be observed macroscopically and microscopically, aiming to determine the type of *Aspergillus sp* which contaminates the green beans. Macroscopic observations include colour, shape, and the edges of the colonies that grew on the media and then continued with microscopic observations using LPCB staining. This staining uses an LPCB reagent which is specialized for staining fungi, LPCB reagent contains phenol crystals, cotton blue, lactic acid, glycerol and distilled water. cotton blue has a function of giving colour to the fungus, glycerol has a function to keep cells from drying out, then lactic acid serves to maintain the structure of the fungus and clean the tissue and finally phenol has a function as a disinfectant (Himedia, 2015).

In this study, the results were obtained macroscopically and microscopically, microscopically the types of *Aspergillus Sp* The contaminating are *Aspergillus fumigatus*, *Aspergillus niger* and *Aspergillus terreus*. Where almost half of 5 *Vigna radiata* are found Aspergillus fumigatus and a small portion of 5 Vigna radiataare found *Aspergillus niger* and *Aspergillus terreus* then from 5 Vigna radiatathere is one green bean that is not contaminated by *Aspergillus Sp*, but contaminated with other fungi *Rhizopus sp.*

Percentage *Aspergillus fumigatusin* this study 40% including the criteria, almost half of them contaminate green beans. Macroscopically, the colour of the colonies was dark green in the form of velvet with uneven edges and microscopically, the conidia were globose or round which contained hyaline and the hyphae that were septate in the hyphae also contained hyaline. As in Fusvita's research (2019), where it was found that *A.fumigatus* with the characteristics of grey colonies with white edges and has septate hyphae containing hyaline. According to Sullivan (2011), this fungus can cause lower respiratory infections if part of the fungus is inhaled, namely conidia.

On *Aspergillus niger* obtained a percentage of 20% which includes the criteria for a small portion of contaminating green beans. Macroscopically, the results were black colonies with semi-rounded colonies and uneven edges, microscopically, the conidia were globose or round with dark brown colour and smooth walls containing hyaline and then the hyphae have thick walls and do not have septa. As in the research of Oramahi (2006), the colony forms on A. *niger* which are based on white and covered with dark brown to black colour and hyphae that are not septate have thick walls. According to Refai (2014), this fungus can cause respiratory infections if part of this fungus or called spores is inhaled by humans.

*Aspergillus terreu*s have the same percentage as *A. niger* which is 20% which includes the criteria for a small portion of contaminating green beans. Macroscopically, the colony colour was creamy and the colony was round and the edges of the colony were uneven (Indrawati, 2006). Microscopically, the conidia were shaped like long columns containing metula and smooth walls containing hyaline and hyphae. *A. terreusseptate* and also contain hyaline. As explained by Refai (2014), that A. terreushas characteristics of cream-coloured colonies such as cinnamon and velvety-shaped colonies, microscopically have the characteristics of septate and hyaline-containing hyphae with conidian containing metula. A. terreusIt can cause a respiratory infection known as aspergillosis.

In this study, there was one sample of 5 samples of *Vigna radiata* that were not contaminated *Aspergillus sp* but still contaminated with other types of fungi, namely *Rhizopus sp*. The sample was seen macroscopically to form white colonies with black spores and shaped like fibres, microscopic examination was not carried out because this study only focused on the type of fungus. *Aspergillus sp*. The *Vigna radiata* were declared negative *Aspergillus sp* due to storage of *Vigna radiata* in an open place such as stored in a jar while other *Vigna radiata* samples are stored in an

open area such as a basin that is stored outside the shop or on the terrace of the shop and the shop is in a humid environment and is not exposed to direct sunlight.

In a study conducted by Nasir (2017), it was explained that the moist storage of Vigna radiatais one of the factors where fungi can grow on these green beans. And according to Sullivan (2011), the spread of *Aspergillus sp* through the air where the spores of the fungus fall on the surface of the food so that it can contaminate the food. So the storage of *Vigna radiata* covered with spores from *Aspergillus* cannot contaminate the surface of the *Vigna radiata*. However, it can still be contaminated with other types of fungi such as *Rhizopus sp* because these fungi are common fungi that contaminate foodstuffs and these fungi play an important role in food spoilage as described in research conducted by Hidayatullah (2018).

CONCLUSIONS

Based on the research that has been done with the title Identification *Aspergillus sp* in *Vigna radiata* sold at Andir Market, Bandung City, it can be concluded that there are 3 types *Aspergillus sp* in *Vigna radiata*, namely, *Aspergillus fumigatus, Aspergillus niger*, and *Aspergillus terreus*

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