

ISOLATION, IDENTIFICATION AND SELECTION OF CELLULOLYTIC FUNGI FROM BANANA WASTE (*Musa paradisiaca*)

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ABSTRACT

The aims of this reesearch was to determine the type of fungus that can grow and degrade cellulose has the potential banana waste. The research conducted by carrying out isolation and identification and selection of cellulolytic fungi from banana waste. Banana waste has a fairly complete nutritional content is to be a potential raw material for fish feed. However, it is containing high cellulose that are not easily digested by fish, it is necessary to need the microorganisms in the process. The research was conducted using the descriptive method. Isolation of fungi with dilution series method on PDA and SDA medium and identification with the moist chamber method. Cellulolytic fungi selection through iodine test by looking at the diameter ratio of transparent zone and colony diameter.

The result showed that total of 6 fungi were isolated from banana waste : *Penicillium nalgiovense* Laxa, *Aspergillus ochraceus* Wilhelm, *Aspergillus nidulans*, *Aspergillus terreus*, *Aspergillus niger*, and *Aspergillus oryzae*. *Aspergillus terreus* and *Aspergillus oryzae* more potential degrade cellulose banana waste than other fungi that found.

Keywords : *cellulolytic fungi, banana waste*

INTRODUCTION

Banana waste is a potential as source of vegetable protein fish food because it contains a fairly complete nutrition. The result of proximate analysis at the Laboratory of Livestock Ruminant Nutrition and Food Chemistry Faculty of Animal Husbandry, Universitas Padjadjaran (2009) content of nutrients and energy of a banana waste is as follows: protein 10.09%, 18.01% crude fiber, crude fat 5.17 %, BETN 55.59%, Calcium 0.36%, 0.10% phosphorus and gross energy 3727 kcal / kg. Based on the analysis of van Soest banana waste contains 27.36% ADF, NDF 39.27%, lignin 20.21%, cellulose 7,15% and hemicellulose 9.91%. Banana waste also contains tannin 0.0967%. The presence of mineral and protein content is high enough waste illustrates the potential banana waste can be used as an alternative feed ingredient for fish. The main problem

using banana waste as a feed ingredient is content the high crude fiber; so it is difficult to digest. Efforts to increase the value of the benefits of banana waste can be done through a service known microbial fermentation process. Fungus have a greater ability to degrading cellulose, it is necessary for reasons of isolation and selection of fungi from banana waste.

The aims of this research was to determine 1) type of fungus that can grow on a banana waste, 2) isolates fungus that have the potential to degrade cellulose of banana waste, and 3) isolates the most potentialfungus to degrading cellulose of banana waste.

MATERIALS AND METHODS

Materials and Equipment

The materials used in this research is the banana waste , distilled water, iodine solution, physiological NaCl 0.9%, distilled water, PDA (*potatoes dextrose agar*), SDA (*Sabourad Dextrose Agar*) methylated, and tetracycline. The tools used in this research include autoclave, glass beaker, Bunsen, Petri dishes, measuring cups, incubator, needle inoculation, labels, cotton, filter paper, analytical balance, erlenmeyer, ose, test tube rack, test tube, microscope, mikropipet, pipettes, and a spatula.

Procedur

Isolation of Fungi

The research method begins with sampling of the banana waste that already retted, whereas to isolate the fungus is through Dilution Series, it is expected fungus contained in the sample can be grown, separated, isolated and subsequently on PDA and SDA medium.

Identification of Fungi

After the isolates obtained, to identify the fungus through the moist chamber method. Fungi identification is then performed by observing colony characteristics and morphology of fungi with the help of fungi identification book.

Selection of cellulolytic fungi

Each isolate of fungi was inoculated on PDA medium, and incubated at room temperature for 24 hours. At the end of the incubation medium surface drops iodine

solution. If the fungus has the ability cellulolytic it will show the transparent zone around the colony isolates of the fungus.

RESULTS AND DISCUSSION

Types of Fungi

There are 6 types of fungi isolates were isolated from banana waste, screening fungus in PDA and SDA medium, (Figure 1 dan Figure 2).



Picture 1. Screening Fungus in PDA medium



Picture 2. Screening Fungus in SDA medium

Six isolates of the fungus are as follows:

Isolate 1

This Isolate were identified as *Penicillium nalgiovense* Laxa , with the characteristics: colony diameter from 2.5 to 3.5 cm for 5 to 7 days. Colonies are white and sometimes gradually become pale green. Having a yellow-green conidiophores. Conidiophores have two to three or even more branches, have hyalin. Conidia globose to subglobose, smooth, hyalin, 3-4 μm .

Isolate 2

Fungi isolate 2 were identified as *Aspergillus ochraceus* Wilhelm, with the characteristics: colony diameter from 2.5 to 3.5 cm in 5-7 days. Colonies brown-black with white side. Konidiophor solid yellow to brown. Head of young konidiophor globose shaped and spread to two or more groups of columns. Konidiphor height 1.5 mm, yellow to brown and rough-walled. Vesicles, globosa, with hyalin measuring 15-20 x μm . Conidia globosa-subglobosa, hyalin-walled with rough or slippery.

Isolate 3

Fungi isolate 3 identified as *Aspergillus nidulans*, with the characteristics: colonies have a white edge, having grown in 5 days time. Conidiophore brown. Head conidia compact and columnar shaped.

Isolate 4

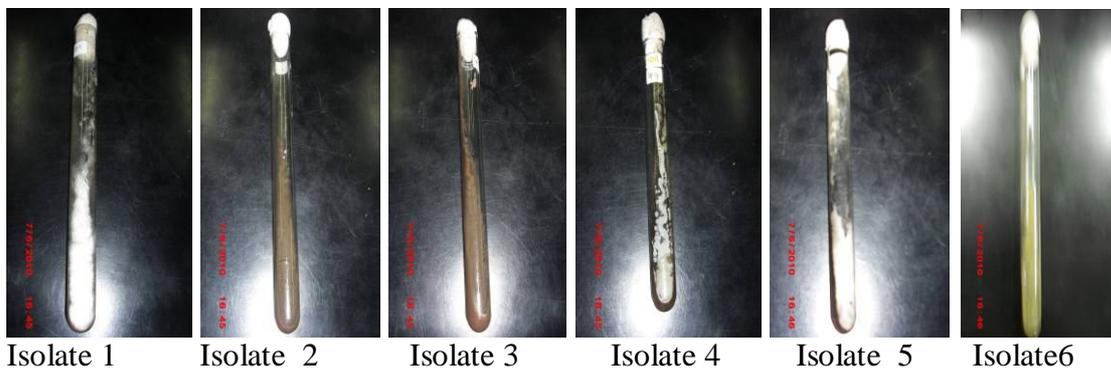
Fungi isolate 4 were identified as *Aspergillus terreus*, with the characteristics: colony diameter from 3.5 to 5.0 cm in 5 to 7 days is green, generally consists of conidiophore solid yellow brown, dark-colored growing increasingly dark. Head conidia compact, high-columna generally 150-500 x 30-50 μm . Conidiophore hyalin, smooth-walled, subglobose vesicles 10-20 μm . Phialide derived from metule 5-7 x 2.0 to 2.5 μm . Metulae 5.5 x 1.5 to 2.0. Conidia globose to ellipsoidal 1.5 to 2.5 μm , yellow slippery hyalin.

Isolate 5

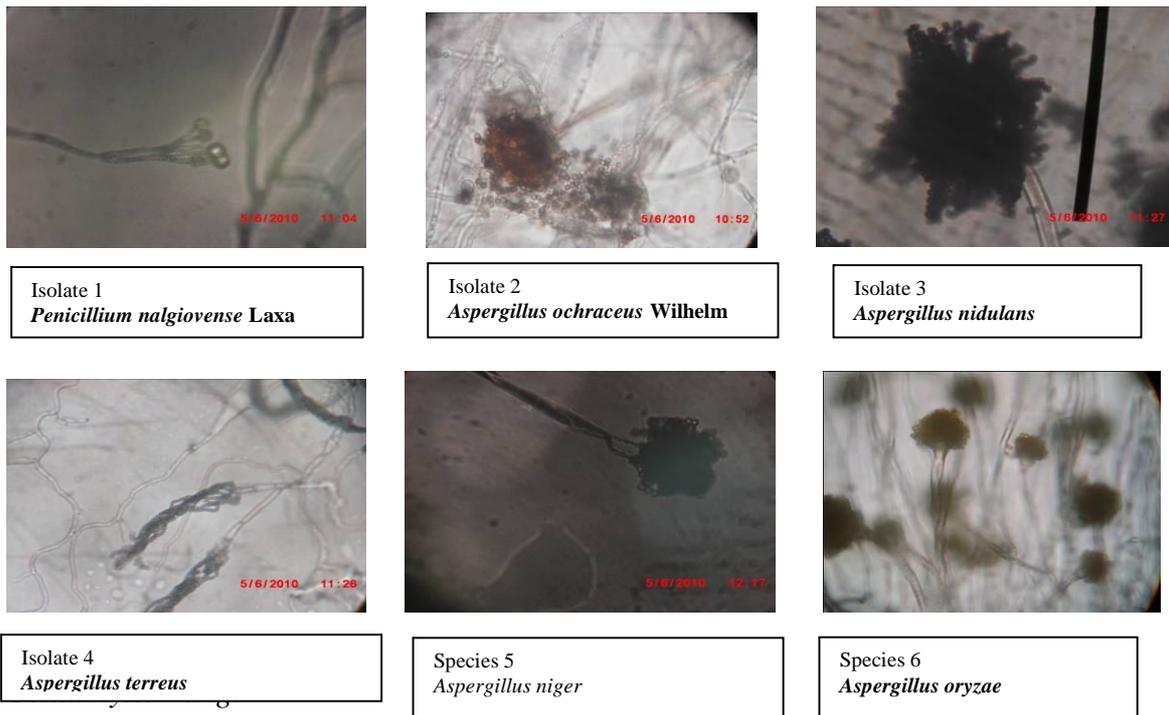
Fungi isolate 5 identified as *Aspergillus niger*, with the characteristics: colony reached a diameter of 4-5cm in 5 to 7 days and consists of a compact basal layer of white to yellow and a wide layer of conidiophores old ciklat colored to black. Head-colored conidia Hiam, round, and tend to split into the fields in the colony of old age. Stipe of conidiophores smooth-walled, colorless hyaline, but can also be brown. Spherical vesicles up to semibulat, and a diameter of 50-100 μm . Phialid formed in metula, and size (7.0 to 9.5) x (3-4) μm . Metula hyaline to brown in color, often bersepta, and size (15-25) x (1.4 to 6.0) μm . Conidia spherical to oval, measuring 3.5 to 5.0 μm , brown, have ornamentation in the form of bumps and spikes that are not uniform.

Isolate 6

Fungi isolat 6 identified as *Aspergillus oryzae* , with the characteristics: colony diameter between 4-5 cm in 5 days, generally consisting of the top with a long conodiophore. Conidia heads radiate, yellow-green to brown. Conidiophore Hyalin measuring 4-5 mm, subglobose vesicles with a size of 40-80 μm. Phialide sometimes comes directly from vesicles or metulae, usually measuring 8-12 x 4-5 μm. Conidia initially shaped ellipsoidal but the older the age-globosa fungi to shape subglobosa with size from 4.5 to 8 μm.



Picture 3. Isolate of Fungus

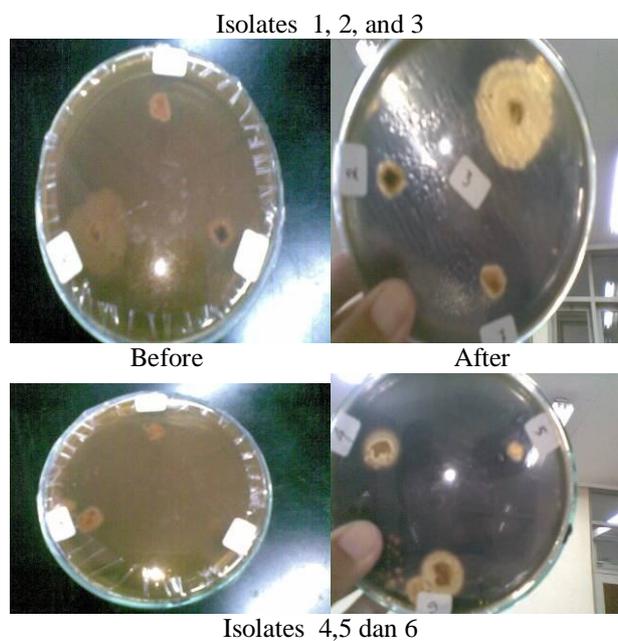


Picture 4. Species of Fungus

Based on a comparison between the transparent zone and the colony diameter formed by each isolate, it obtained two isolates of fungi that have a greater ability to degrade cellulose, namely *Aspergillus terreus* and *Aspergillus oryzae*. As for the comparison between the transparentlear zone and colony diameter can be seen at Table 1.

Table 1. Comparison between Transparent Zone and Colony Diameter

Fungi	Transparent Zone (mm)	Colony Diameter (mm)	Comparison TZ dan CD (mm)	Number of Fungi Candidate
Isolate 1	8	7	1.14	5
Isolate 2	9	6	1.5	3
Isolate 3	27	22	1.22	4
Isolate 4	14	7	2.00	2
Isolate 5	5	5	1,00	6
Isolate 6	17	6	2.83	1



Picture 5. Iodium Test

The potential fungus to degrade cellulose is Isolate 6. The greater the ratio of the diameter of transparent zone and colony diameter, the fungus more potential degrade cellulose (Kader, Omar, 1998). Fungi can degrade cellulose because it can form and secrete the enzyme cellulase. The greater the amount of cellulase enzymes that are

secreted in the faster degradation of cellulose occur (Bagga and Sandhu, 1987 in Zumrotiningrum, et al 2004).

CONCLUSION

Retrieved 6 types of isolates fungus that grows on a banana waste, namely: *Penicillium nalgiovense*, Laxa, *Aspergillus ochraceus* Wilhelm, *Aspergillus nidulans*, *Aspergillus terreus*, *Aspergillus niger*, *Aspergillus oryzae*. *Aspergillus terreus* and *Aspergillus oryzae* were the best candidates with the highest capability in cellulose degradation.

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