PENGUJIAN KEMAMPUAN ANTAGONISTIK KHAMIR EPIFIT ASAL KEBUN RAYA CIBODAS DAN POTENSI *Candida* sp. Berkhout UICC Y-328 SEBAGAI AGEN BIOKONTROL *Aspergillus ochraceus* Wilhelm PADA TOMAT PASCAPANEN

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TESIS

Diajukan sebagai salah satu syarat untuk
memperoleh gelar magister sains

Oleh:
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Tanggal Lulus : 23 Desember 2009

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Title : STUDY ON THE ANTAGONISTIC ABILITY OF EPIPHYTIC YEASTS OF CIBODAS BOTANICAL GARDEN AND THE POTENTIAL OF *Candida* sp. Berkhout UICC Y-328 AS A BIOCONTROL AGENT OF *Aspergillus ochraceus* Wilhelm ON POSTHARVEST TOMATOES

Thesis Supervisors: I. Ariyanti Oetari, Ph.D. ; II. Dr. Andi Salamah

SUMMARY

Microbial communities usually have mixed populations, only in unique environmental situations do microorganisms live entirely alone. Thus many types of interactions are possible among the members of an ecosystem's community. In general, the constant association of different organisms in an ecosystem is referred to as symbiosis, with the associates being called symbionts. One type of a symbiosis is antagonism. Antagonism is a symbiotic relationship in which one population of microorganisms has a harmful effect on the growth of another microbial population (Batzing 2002: 696).

A number of microorganisms (bacteria, yeasts, fungi) which effectively control postharvest pathogens have been identified as antagonists (Mari and Guizzardi 1998:60). A variety of microbial antagonists were reported to control several different pathogens on various fruits. The organism that suppresses the pest or pathogen is referred to as the biological control agent (BCA) (Pal & Mcspadden Gardener 2006: 1). Biological control may in simple terms be defined as the use of one living organism to control another (Druvefors 2004: 4). Often antagonists are isolated on the surface of plants; this natural presence

makes them more likely to succeed because of their colonization ability and environmental adaptation (Mari and Guizzardi 1998:60).

The use of yeasts as antagonists appears to be quite promising, although the mechanism has not yet been fully elucidated. Some antagonist yeasts have been reported as biocontrol agent of fungal pathogen on fruits. Zhao *et al*. (2008: 115--116) reported that tomato fruit treated with *Pichia guillermondii* had an infection rate of 25% which was caused by *Rhizopus nigricans*, which was significantly lower than the control (41.67%). Kalogiannis *et al.* (2006: 72) reported that *Rhodotorula glutinis* Y-44 significantly reduced disease incidence caused by *Botrytis cinerea* on tomato by 52%, compared to the untreated control. Zhang *et al.* (2004: 84) reported that the application of *Cryptococcus laurentii* resulted in low average decay incidence caused by *B. cinerea* in fruit by 7.1%, compared with 40% in the water-treated control fruit.

University of Indonesia Culture Collection (UICC) collected epiphytic yeasts from plant samples of Cibodas Botanical Garden, and moulds from decayed tomatoes and infected plants. The ability of the epiphytic yeasts as biocontrol agents against tomato spoilage-causing moulds has not been reported.

This study consists of two parts. Part 1 is The Antagonistic Ability of Epiphytic Yeasts of Cibodas Botanical Garden on Tomato Plant Infected-Causing Moulds. Part 2 is The Potential of *Candida* sp. UICC Y-328 as a Biocontrol Agent of *Aspergillus ochraceus* on Postharvest Tomatoes.

The purposes of this study were to investigate the ability of six species of epiphytic yeasts in inhibiting the growth of tomato plant infected-causing moulds, and the potential of *Candida* sp. UICC Y-328 as a biocontrol agent in reducing postharvest tomato spoilage caused by *Asp. ochraceus*.

The media used for growing the yeasts was Yeast Malt Agar (YMA), and maintenance for fungi was Potato Dextose Agar (PDA). The media used for antagonistic test were PDA and Potato Dextrose Broth (PDB).

Antagonistic test by strip method was carried out by using the concentrations of yeast cells at (0.7--4.45) x 10^8 CFU/ml, and *Asp. ochraceus* at (7.0--8.1) x 10^7 CFU/ml, *Asp. terreus* Thom at (7.7--8.6) x 10^7 CFU/ml and *Drechslera* sp. at (0.45--3.5) x 10^5 CFU/ml. The yeast cells were inoculated 4 hours earlier before inoculation of mould spores on PDA in Petri dishes. Results showed that *Candida* sp. UICC Y-328 has highest percentage of colony reduction of *Asp. ochraceus* (56.45%), followed by *Metschnikowia reukaufii* UICC Y-351 on reducing colonies of *Asp. terreus* and *Drechslera* sp. (25.42% and 51.28%, respectively) during 6-day incubation.

Antagonistic test by co-culture method was carried out by using the concentrations of yeast cells at (0.7--4.45) x 10^8 CFU/ml, and *Asp. ochraceus* at (6.0--8.6) x 10^7 CFU/ml, *Asp. terreus* at (4.6--9.5) x 10^7 CFU/ml. The yeast cells were inoculated 8 hours earlier before inoculation of mould spores on PDB. Results showed that *Candida* sp. UICC Y-328 reduced the size of conidial heads (5.52%) and hyphae (8.29%) of *Asp. ochraceus*, at 3-day incubation. *Cryptococcus laurentii* UICC Y-379 reduced the size of conidial heads and

hyphae of *Asp. ochraceus* (15.07% and 11.60% respectively) and *Asp.terreus* (12.35% and 24.47% respectively) at 3-day incubation. Antagonistic test by slide culture method showed that the yeast cells of four strains (*Candida rancensis* UICC Y-326, *Cr. laurentii* UICC Y-319, *Cr. laurentii* UICC Y-379,and *M. reukaufii* UICC Y-351) attached to hyphae of *Drechslera* sp. after 3- and 4-day incubation. Cells of *Candida* sp. UICC Y-328 attached to hyphae of *Drechslera* sp. after 4-day incubation. Cells of *Cr. laurentii* UICC Y-385 was not able to attach to hyphae of *Drechslera* sp.

Candida sp. UICC Y-328 was potential in reducing the growth of *Asp. ochraceus*, and was investigated further for its potential as a biocontrol agent. Wounds on postharvest tomatoes were inoculated with 25 µl of yeast cell suspension and 25 µl of mould spore suspension. The yeast cells were inoculated 24 hours earlier before inoculation of mould spores on wounds of tomatoes. Biocontrol study showed that incidence of spoilage in postharvest tomatoes which were wounded and inoculated with *Candida* sp. UICC Y-328 and *Asp. ochraceus*, were reduced by 20% after 15-day incubation at room temperature. All postharvest tomatoes which were wounded and inoculated with *Asp. ochraceus* as control, were spoiled (100%). Synthetic fungicide Dithane M-45 at a concentration of 0.08% reduced spoilage incidence by 70%. *Candida* sp. UICC Y-328 was not effective as biofungicide in reducing spoilage incidence.

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