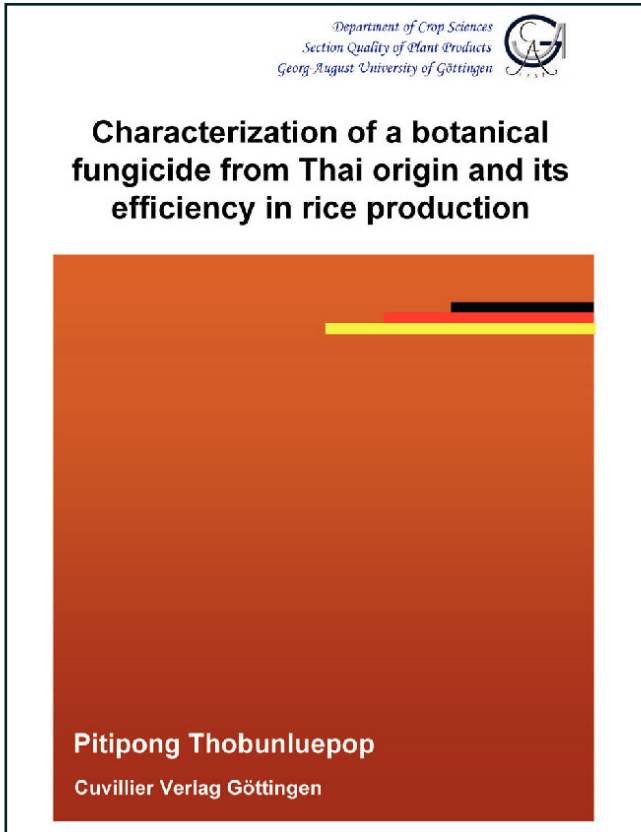




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**Characterization of a botanical fungicide from Thai origin and its efficiency in rice production**



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# **1. Introduction**

## **1.1 Background**

Rice (*Oryza sativa*) is a major staple food and a mainstay for the rural population and their food security in Asia, Latin America, Caribbean and Africa. It is central for the food security of over half the world population (Cheema *et al.*, 1991). In Thailand, rice is mainly cultivated by small farmers in holdings of less than one hectare. Crop yields in the small scale or subsistence farming sector are relatively lower than in the commercial farming sector. This is due to lack of management capacity, unfamiliarity with appropriate technologies, and shortages of farming inputs such as irrigation, fertilizers, pesticides, and certified seeds (Reissig *et al.*, 1986). The International Rice Research Institute (IRRI) has demonstrated that rice yield in Asia can be increased by applying scientific principles and new technologies. For examples, the use of modern varieties and improved cultural practices led to the increase of rice yield throughout tropical Asia (IRRI, 2007).

Plant diseases are most important factors, which challenge problems in commercial agriculture and pose real economics threats to both conventional and organic farming. Since the beginning of agriculture, seeds played a major role in agricultural production, because over 90% of the crop productions are propagated via seeds (Agrios, 1988). At the same time, seeds are the major carriers of plant pathogens, including bacteria, viruses and especially fungi. Thus, disease free seeds provide the front line of defense against the development and spread of plant diseases, which are responsible for large crop losses on worldwide basis (Islaml *et al.*, 2000).

## **1.2 Literature review**

### **1.2.1 Seed borne diseases**

Seed qualities are seriously affected by many factors. The principle causes of seed quality losses are biological factors as fungi, insects, mites and rodents (Cheema *et al.*, 1991). Fungi infection is one of the most important factors affecting storage life and is one of the major causes for seed quality deterioration as well as decreased crop yield. It may also degrade product quality (Farr *et al.*, 1989). Rice crop is known to be attacked by many seed borne fungi, which cause major and minor diseases (Appendix 1). Fungicide seed treatment is a promising way to control the invasion of seed borne fungi. These treatments are used for three main reasons: (1) to control soil-borne fungi that cause seed rots, damping-off, seedling blights and root rot; (2) to control fungi that are surface-borne on the

seed, and (3) to control internally seed-borne fungi. The degree of control with fungicide seed treatment depends on: (1) active ingredients of the fungicide (2) rate of applied fungicide (3) presence of seed- and soil-borne pathogens (4) environmental conditions, and (5) application coverage (Smith *et al.*, 1988).

Fungicide seed treatments do not control bacterial pathogens and most of them will not control all types of fungal diseases. Therefore, it is important to choose carefully the treatment that provides the best control of the disease organisms present on the seed or potentially present in the soil. The degree of control varies with product, rate of fungicide use, environmental conditions and disease organisms present. The rate of application prescribed by the label should be considered because over-treatment may injure the seed and under-treatment may not provide good disease control (Webster and Gunnell, 1992).

Fungicide seed treatments help to control soil-borne fungi that cause seed decay, seedling blight and root rot. Control of these diseases may result in better stands, more vigorous seedlings, and increased yields. The application of the fungicide captan (N (trichloromethylthio)-tetrahydrophthalimide) to rice seed has been found to be effective in enhancing seed germination and protecting rice seed borne fungi (Jiskani, 1999). However, in Europe, captan has been banned for use in agriculture because it is a known carcinogen. Additionally, chemicals application and their residues can generate tremendous negative side effects on soil, water and atmospheric environment, which may cause either direct or indirect impacts on human being or other living organisms (Baird, 1994).

The chemicals remain from fungicides in the environment occur either from the overdose application or their lengthy persistent effect and are present generally in soil and water with implications on all living beings in both direct and indirect ways. The WHO reported in 2005 that 5,500 Thai people, 50 % Philippines rice farmers, and 42,800 people in China have health problems because of pesticide poisoning (Jitsaguan, 2005). Those who are affected by chemical toxins are prone to high risk of cancer with possibility of genetic inheritance that can weaken the health status of future generations. The heavy and lengthy use of agrochemicals for pest control not only pollutes the environment, but can also affect negatively the ecosystem as well as the food chain and can lead to pesticide resistance problems. The whole process can generate the loss of biodiversity or even the extinction of some organisms. Additionally, agrochemical residues in food products may become

problems for international trade (Kamal and Moghal, 1968). The alternatives to chemical pesticides for pest control that receive attention are the use of mechanical methods, resistant plant varieties, improved cultural practices, and biological pesticides. Biological pesticides are pesticides whose active ingredients are plant-produced chemicals (Jiskani, 1999).

The growing environmental awareness and health concern worldwide at present time have led to consumer demand for safe and clean foods particularly those chemicals-free products. Therefore, nowadays there emerge many academic interests on bio-fungicides as an alternative to toxic synthetic fungicides to minimize undesirable side effects. Biological fungicides offer important advantages compared to chemical pesticides. The use of these agents in agriculture introduces fewer risks to human health and to the environment than chemical pesticides. Additionally, bio-fungicides are generally relatively inexpensive to produce. Therefore, the use of herbal extracts for plant protection could be a promising way in a chemical-free agricultural system (Horrigan *et al.*, 2002).

Many researchers investigated the effect of extracts from *Eugenia caryophyllus* Bullock *et. Harrison* (Myrtaceae), *Acorus caramus* Linn. (Araceae), *Mammea siamensis* (Miq.) T. (Guttiferae), *Eupatorium odoratum* L. (Compositae) and some species of *Stemona* (Stemonaceae) on different fungal diseases. Issakul *et al.* (2003) reported that extracts from *A. caramus*, *M. siamensis*, and *S. curtisii* have insecticidal properties. Chanthayot and Jatisatiern (2003) found in laboratory tests beneficial fungicidal properties with extracts from *E. caryophyllus* and *A. caramus*. On the other hand, *E. odoratum* had unclear biological activity. Wonggirathiti and Jatisatiern (2003) reported that  $\beta$ -asarone, found in *A. caramus* and eugenol from *E. caryophyllus* showed very high fungicidal properties. Moreover, Mungkornasawakul *et al.* (2003 and 2004) described that stemocurtisine, found in *S. curtisii*, and surangin C from *M. siamensis*, showed high biological activity in *Atemia salina* Leach (Anostraca) and in some insects. Furthermore, the analysis of plants treated with extracts of the studied plants did not show any harmful residues. In field studies, farmers were very satisfied with the new botanical pesticides because they could use them instead of harmful synthetic pesticides.

One of the pest control methods that have received scant attention in developing countries and which have been extensively used in developed countries is seed coating technology. The coating of seeds by chemicals or alternative substances can reduce the percentage of plant damage by pests and the level of pesticide application significantly, which could decrease the amount of pesticide application by 85% compared to direct drench application methods (Embaby, 2006). Another advantage of seed coating technique is the preservation of the quality of seeds and the inhibition of seed deterioration (Whipp and Budge, 1993). At thus becomes an interest and a challenge to explore whether the western knowledge of seed coating technique and the available local bio-pesticides can be combined to produce efficient botanical fungicide coated seeds to respond the environmental awareness and meet consumer's demand for clean and safe food.

#### 1.2.2 Effect of seed treatment on seed quality

Planting of high quality seeds is important for an efficient rice production system. In order to obtain optimum stands and yields, early planting, reduction of seeding rates, and the drill planting required high quality and vigorous seeds. Strong seedlings have faster growth rate, greater tolerance to fungi infection and stress than less vigorous ones.

Diseases affect seed quality, yield and may degrade product quality. A fungicide seed treatment protects the seeds and young seedlings from many seed borne and soil borne fungi infections. However, fungicides can produce phytotoxic effects that cause losses of seeds germinability and seedling vigor (Yildirim and Hoy, 2003). These phytotoxic effects may be amplified by the over-application of fungicides. Von Pine *et al.* (1995) reported that fungicide treatments with captan and pirimifos reduced maize seed qualities during storage and the posterior behavior under field conditions, which led to yield reduction. Silva *et al.* (1996) reported that clorpirifos and captan reduced the maize seeds germination and the vigor during the storage.

Therefore, the use of botanical fungicides for plant pathogens control could be a promising way in a chemical-free agricultural system (Bhatti and Soomro, 1996). However, there are also some disadvantages of botanical fungicides:

- (1) They are more difficult to implement than the chemical fungicides,
- (2) They have generally a narrow target range,
- (3) They do not act as quick as chemical fungicides,

- (4) They have a shorter shelf life than the chemical fungicides, and
- (5) They are not compatible with chemical fungicides or herbicides.

Moreover, some essential oils from the botanical fungicides can cause phytotoxic effects on seed viability when used at high concentrations (5 to 10%, v/v) (Tworkoski, 2002).

Thus, the present study was conducted to address the following objectives:

1. To screen and select the best antifungal activity of plant crude extract or essential oil against pathogenic fungi on rice seeds,
2. To compare the effectiveness of conventional chemical seed treatment and botanical fungicide coated seed against rice seed borne fungi during storage,
3. To investigate the effects of various seed coating substances on rice seeds qualities, especially on chemical and biochemical properties,
4. To evaluate the effectiveness of various seed coating substances on seed practice and plant productivity under field conditions,
5. To evaluate the effect of seed coating substances on the quality of the harvested grain,
6. To evaluate the effect of various seed coating substances on environmental pollution

### **1.3 Thesis outline**

In the Chapter 2, screening and selecting of the best antifungal activity of four different plant crude extracts or an essential oil against pathogenic fungi are described. These extracts will be used for seed coating technology in comparison to conventional chemical fungicide seed treatment to evaluate their effectiveness for seed borne fungi control before and during germination. These experiments are described in Chapter 3. Then, in Chapter 4, the effects of various seed coating substances on rice seed properties before and during storage are reported. The effect of various seed coating substances on rice seed properties before and during storage and the relationship between chemical and biochemical properties changes in rice seeds are discussed in Chapter 5. The results of field experiments to compare the effectiveness of the botanical fungicide and conventional chemical seed treatment on seed quality in practice are presented in Chapter 6. Chapter 7 characterizes the influence of various seed coating substances on the quality of harvested rice grain. Chapter 8 contains the conclusion and summary.

**Predicted results:** The tested plant extracts act as botanical fungicides and are an alternative source to replace chemicals for seed coating application in order to control

fungal diseases without physiological injuries in the seeds and to ensure high yields as well as a stable quality of the harvested grains. Moreover, fewer amounts of chemical residues contaminated in both soil and harvested product are expected.

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