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Disinfection of *Fusarium*-infected Rice Seeds by Prochloraz and Gaseous Chlorine Dioxide

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Three species of *Fusarium*, *F. fujikuroi*, *F. verticillioides* and *F. proliferatum*, are known to be associated with bakanae disease of rice [1, 2]. *F. fujikuroi* infects rice flowers and survive in endosperm and embryo of the seeds. Infected seed is an important source of primary inoculum of pathogens [3]. Seeds of rice (*Oryza sativa* cv. Boramchan) collected from bakanae-infected field were found to be 96% infected with *Fusarium* sp., 52% with *F. fujikuroi*, 42% with *F. verticillioides*, and 12% with *F. proliferatum* as determined by incubation method and species-specific PCR assays. *F. fujikuroi* was detected at lemma/palea, endosperm and embryo whereas *F. verticillioides* and *F. proliferatum* were recovered only from lemma/palea by means of component plating test.

Seed disinfection methods have been developed to control bakanae disease and prochloraz has been most widely used for rice seeds. Two chemicals formulated with prochloraz (PC 1) and prochloraz + hexaconazole (PC 2) that inhibit biosynthesis of ergosterol strongly reduced the incidence of *Fusarium* spp. on selective media to 4.7% and 2.0%, respectively. Disease symptoms of rice seedlings in nursery soil were alleviated by chemical treatment; seedlings with elongated leaves or wide angle between leaf and stem were strikingly reduced from 15.6 to 3.2% (PC 1) and 0 (PC 2), stem rots were reduced from 56.9 to 26.2% (PC 1) and 32.1% (PC 2), and normal seedling increased from 0.4 to 13.3% (PC 2).

Prochloraz has some disadvantages and risks such as the occurrence of tolerant pathogens [4] and effects on the sterol synthesis in animals and humans [5]. For these reasons, it is necessary to develop new disinfection method that do not induce fungal tolerance and are safe to humans and animals. Chlorine dioxide (ClO_2), that is less toxic, produces no harmful byproducts, and has high oxidizing power, has been reported to be effective at disinfection of several phytopathogenic fungi including *Colletotrichum* spp. and *Alternaria* spp. [6]. Gaseous ClO_2 applied to rice seeds at a concentration of 20 ppm strongly suppressed mycelial growth of *Fusarium fujikuroi, F. verticillioides* and *F. proliferatum*. The incidence of *Fusarium* spp. in dry seed with 8.7% seed moisture content (SMC) tended to decrease as the concentration of ClO_2 increased from 20 to 40 ppm. Applying 40 ppm ClO_2 at 90% relative humidity, incidence was reduced to 5.3% and resulted in significant reduction of disease symptoms on MS media. In nursery soil, stem rot was reduced from 56.9 to 15.4% and the number of normal seedlings increased from 0.4 to 25.5%. With water-soaked seeds (33.1% SMC) holding moisture in the endosperm and embryo, the effectiveness of disinfection using ClO_2 increased, even when treated with only 20 ppm for four hours. This suggests that moisture was a key element for action of ClO_2. Removal of the palea and lemma from seeds significantly decreased the incidence of *Fusarium* spp. to 3.0%. Seed germination appeared to decrease slightly by water-soaking at 30°C because of increased SMC and by physical damage of embryos from hulling. These results indicate that the use of gaseous ClO_2 was effective as a means to disinfect rice seeds infected with *Fusarium* spp. and that moisture around the pathogens in the seed was an important factor for the action of ClO_2. Further investigations should be conducted to ascertain the best conditions for complete disinfection of *Fusarium* spp. that infect deep site of rice seeds.

References

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