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Expression of human lactoferrin gene transformed rice(Oryza sativa L.) via Agrobacterium

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Objectives

To produce resistant rice against pathogene expressing human lactoferrin through Agrobacterium and to extend utility of transformed rice expressing in leaves as well as grains of plants by using ubiquitine promoter.

Materials and Methods

- 1. Plant materials: Rice (Oryza sativa L. cv. Dong Jin)
- 2. Methods: Herbicide test, PCR, Southern blot analysis, Western blot analysis, Antibacterial activity

Results and Discussion

Callus derived from mature seeds of rice(Oryza sativa L. cv. Dong Jin) were co-cultivated Agrobacterium tumefaciens EHA105 containing Ubi and genes for HLF and bialaphos resistance(Bar). Transgenic plant survived on medium containing 2.5mg/L bialaphos were resistant to herbicide(Meiji herbiace) at a dosage lethal to wild type plants. PCR and Southern blot analysis confirmed that T-DNA was integrated into the plant genome. The expression of lactoferrin protein was detected by Western blot analysis from leaves and grains of transformants. The expression of a human lactoferrin gene produces a potent antibacterial protein in transgenic rice leaves. T₁ progenies were normally grown on media containing 5mg/L bialaphos.

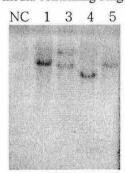


Fig. 1. Southern blot analysis of transgenic (Lines 1,3-5) and control (NC) plants. $40\mu g$ genomic DNA was digested with EcoRI and hybridized to Bar probe

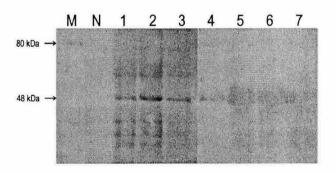


Fig 2. Western blot analysis of human lactoferrin protein in transgenic rice plants.

Lanes 1: Purified human lactoferrin, Lanes 2: non-transgenic rice plant, Lanes 3-6: transgenic rice plants.