

Mutant screening of transgenic *Arabidopsis* in genetic pathway of barley metallothionein promoter

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ABSTRACT

In order to search for signaling factors altering the expression of barley metallothionein promoter, the mutant screening technique was used. The promoter region of metallothionein in barley (cv. Hordea) was used to drive the *gus* gene and it was transferred to *Arabidopsis thaliana*. The M₀ seeds was mutagenized using four doses (10, 20, 30, 40 Grays) of fast Neutron radiation. The chemical compound of 3-Amino-1,2,4-Triazole (3-AT) at 20 mM was chosen as the best activator using CRD statistical design. Inheritance of GUS activity was evaluated using histochemical GUS assay. 48 hours after spraying by 20 mM 3-AT the inhibition model of 3:1 GUS expression: non expression was tested. 20 potential mutant lines were selected by carrying out several screening steps. In each stage 30 seedlings of 1500 M₂ lines were checked by GUS staining. The arrangement of promising lines included 3, 5, 9 and 3 which were referred to 10, 20, 30 and 40 Gray of fast Neutron treatments respectively. These lines were analyzed using fluorometric β-glucuronidase technique and were selected as potential mutants in genetic pathways of metallothionein promoter activity for further studies.

Key Words

Barley, Fast Neutron, Mutation, Metallothionein Promoter, *gus* Gene.