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Investigation of Shoot Development Using Promoter Trap System in Arabidopsis

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Promoter trap system provides a powerful tool for useful promoter identification as well as insertional mutagenesis. The promoter trap system contains a promoterless repoter gene that expression can occur only when the insertion is within a transcriptional unit and in the correct orientation.

We have developed a vector, pFGL561, which can be used for promoter trap in dicot such as Arabidopsis. The binary vector contains a promoterless synthetic green fluorescence protein (sGFP). In addition, one splice donor/acceptor (D/A) element preceding the reporter gene can allow expression even if insertion occurs within an intron.

A total of 604 T_1 lines of Arabidopsis have been generated using the pFGL561 so far. Using GFP Imaging System with argon laser as light source, among 300 T_1 plants out of the 604 T_1 plants, we found 137 lines showing the GFP signals in shoot apex (33 lines), hypocotyl (27), root (12), shoot apex and hypocotyl (24), shoot apex and root (6), hypocotyl and root (2), and whole seedlings (32). For 11 T_1 lines, the GFP activity in T_2 plants has been reconfirmed under fluorescence microscope.

As a result of the analysis of phenotypes in T_2 plants, we have found 7 lines showing abnormal shoot development such as dwarfism, retarded shoot growth. As well as abnormal vegetative development, we also found T_2 plants showing abnormal reproductive development such as semi-sterility and flowers with four stamens.

Further analysis of the phenotypes of T_2 plants and the identification of the tagged genes by inverted PCR are undergoing.