

## Differential expression of anthocyanin biosynthetic genes and anthocyanin accumulation in tartary buckwheat cultivars 'Hokkai T8' and 'Hokkai T10'

<sup>a</sup>Department of Crop Science, <sup>c</sup>Department of Bio Environmental Chemistry, Chungnam National University <sup>b</sup>National Agricultural & Food Research Organization, Hokkaido, Japan

Nam Il Park<sup>a</sup>, Xiaohua Li<sup>a</sup>, Tatsuro Suzuki<sup>b</sup>, Sun-Ju Kim<sup>c</sup>, and Sang Un Park<sup>a,\*</sup>

### Objectives

We used anthocyanin biosynthetic genes clones, along with previously isolated clones for phenylalanine ammonia lyase (PAL) and chalcone synthase (CHS), to compare the expression of anthocyanin biosynthetic genes in different organs of tartary buckwheat cultivars 'Hokkai T10' and its parent cultivar 'Hokkai T8.' In addition, we analyzed the anthocyanin and flavonol contents in seedlings of these 2 cultivars.

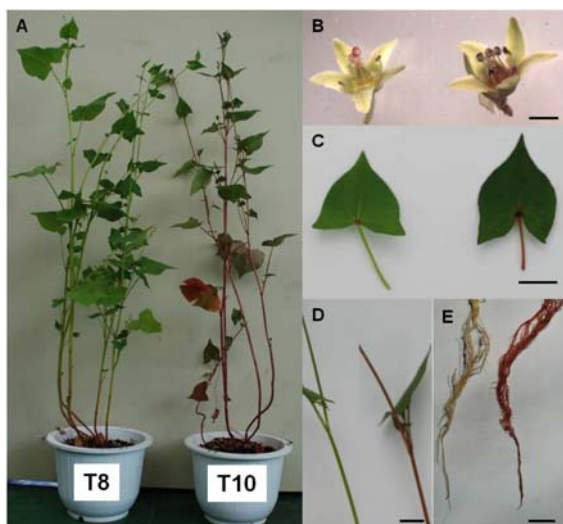
### Materials and Methods

- Two cultivars of tartary buckwheat, 'Hokkai T8' and 'Hokkai T10,' were bred by the National Agricultural Research Center (Hokkaido, Japan).
- RNA extraction and polymerase chain reaction analyses
- Cloning of cDNAs encoding anthocyanin biosynthetic enzymes
- Flavonoid staining and Quantification of anthocyanin content

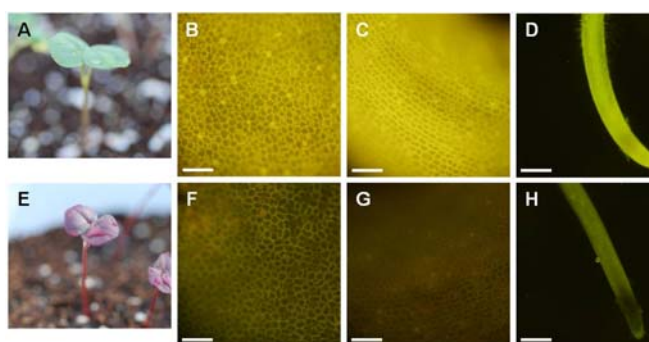
### Results

We cloned and characterized 6 genes involved in anthocyanin biosynthesis in tartary buckwheat, namely *FtCAH*, *FtACL*, *FtCHI*, *FtF3H*, *FtF3'H*, and *FtANS*, which encode cinnamate 4-hydroxylase (C4H), 4-coumarate:CoAligase (4CL), chalcone isomerase (CHI), flavones 3-hydroxylase (F3H), flavonoid 3'-hydroxylase (F3'H), and anthocyanidin synthase (ANS), respectively. Then, we used these cDNAs, along with previously isolated clones for phenylalanine ammonia-lyase (PAL) and chalcone synthase (CHS), to compare gene expression in different organs of tartary buckwheat cultivars 'Hokkai T8' and 'Hokkai T10.' The expression of anthocyanin biosynthetic genes, and consequently, anthocyanin concentrations, was higher in 'Hokkai T10' than in 'Hokkai T8.' However, naringenin chalcone, a flavonoid, was absent from 'Hokkai T10' seedlings.

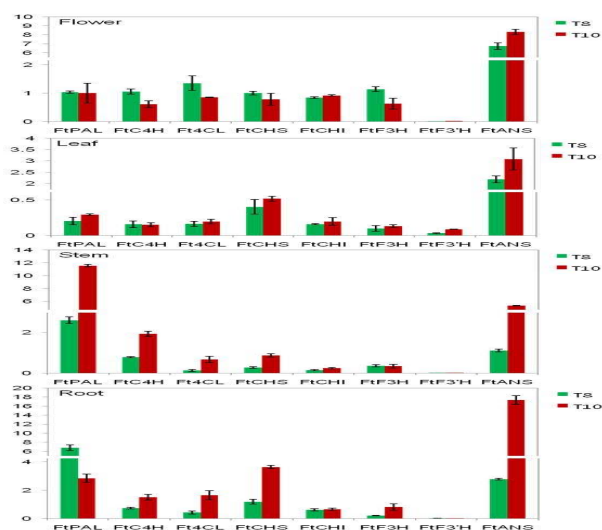
.....  
\* corresponding author: Tel. 042-821-5730, E-mail: [supark@cun.ac.kr](mailto:supark@cun.ac.kr)



**Fig. 1.** Tartary buckwheat cultivars ‘Hokkai T8’ and ‘Hokkai T10.’ (A) Whole plants, (B) flowers, (C) leaves, (D) stems, and (E) roots. The scale bar represents 1 mm in (B) and 1 cm in (C), (D), and (E).



**Fig. 2.** Anthocyanin and flavonoid contents in tartary buckwheat cultivars ‘Hokkai T8’ (A - D) and ‘Hokkai T10’ (E - H) seedlings. Anthocyanin accumulation in ‘Hokkai T8’ (A) and ‘Hokkai T10’ (E) seedlings. Flavonoid staining in cotyledons (B, F), cotyledonary nodes (C, G), and roots of ‘Hokkai T8’ and ‘Hokkai T10’ (D, H). The characteristic bright yellow fluorescence of chalcone–naringenin is strongest in the cotyledons and cotyledonary nodes of ‘Hokkai T8’ (B, C), but is weaker in ‘Hokkai T10’ (F, G). The characteristic bright green fluorescence of kaempferol is strongest in the roots of ‘Hokkai T8’ (D), but is weaker in ‘Hokkai T10’ (H). The scale bars in (B), (C), (F), and (G) represent 100  $\mu\text{m}$  and those in (D) and (H) represent 500  $\mu\text{m}$ .



**Fig. 3.** Differential expression of anthocyanin biosynthetic genes in different organs of tartary buckwheat cultivars ‘Hokkai T8’ and ‘Hokkai T10.’ The height of each bar represents the mean of 3 measurements and error bars indicate the standard deviation.

**Acknowledgements:** This study was supported by Technology Development Program for Agriculture and Forestry, Ministry for Food, Agriculture, Forestry, and Fisheries, the Republic of Korea.