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Functional divergence of SnRK1.1 family members in regulating the growth and yield formation in *Brassica napus*

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Background:

As an important oil crop in the world, the yields and low harvest index (HI) are affected by "source-flow-storage" in rapeseed (Brassica napus), which leads to the severe rapeseed industry development. Thus, studying the energy transport and storage are extremely meaningful in B. napus based on high yield seedlings.

Objective:

Based on the extremely high harvest index (HHI) and low harvest index (LHI) materials, the expression and function of BnaSnRK1.1 members were analysed, and the functional differentiation of BnaSnRK1.1 genes affecting yield and energy transport in rapeseed were further clarified, which would contribute to further exploration of high yield and breeding new varieties in rapeseed.

Methods:

Sequences of BnaSnRK1.1 members were analysed by gene cloning. Based on their expression patterns, BnaA01.SnRK1.1 and BnaA05.SnRK1.1 were selected for further study. Using GUS staining, we checked the differences of genes expression. Chromatin accessibility, methylated and histone modification were used to explore the reasons for the expression differences in the harvest index accessions. Phenotype observation and physiological index from transgenic plants suggested the functional differentiation of BnaA01.SnRK1.1 and BnaA05.SnRK1.1. IP-MS, yeast two-hybrid and dual luciferase reporter assay were used to resolve the molecular mechanism of BnaA01.SnRK1.1 in regulating yield and energy transport.

Results:

BnaA01.SnRK1.1 and BnaA05.SnRK1.1 are both DEGs in HHI and LHI materials, while their sequences both remained highly conserved in coding region and 2kb upstream regions (ATG) in the two materials, respectively. These two genes presented different expressions in same materials. Based on the induced GUS staining, the differed expression levels of BnaA01. SnRK1.1 and BnaA05.SnRK1.1 owned to the responding to IAA, ABA, SA and mannitol. Chromatin accessibility suggested the two BnaSnRK1.1 genes may both locate in open and closed chromatin area in LHI and HHI, respectively. And the active H3K27ac and H3K4me3 contributed to the expression differs of BnaSnRK1.1 in the two accessions. The function differentiation of BnaSnRK1.1 members was revealed by transgenic plants, overexpressed BnaA01.SnRK1.1 resulted to more siliques on the main inflorescence but smaller seeds, while overexpressed BnaA05.SnRK1.1 increased the silique number per plant and larger biomass. Moreover, BnaCIPK6 and BnaFBA2 could interact with BnaA01.SnRK1.1, but no interaction with BnaA05.SnRK1.1.

Conclusions:

BnaSnRK1.1 members were conservative in different accessions. Cis-elements in promoters resulted in the different expression levels between BnaA01.SnRK1.1 and BnaA05.SnRK1.1. Chromatin accessibility affected the levels of histone modification for BnaSnRK1.1 in harvest index materials, which made the genes expression pattern differed. BnaA01.SnRK1.1 could function at the flower bud differentiation to affect the silique number in main inflorescence, seed size may be affected by phytohormone, which may be regulated by BnaCIPK6. At the same time, energy metabolism was changed in transgenic seedlings of BnaA01.SnRK1.1 by interacting with BnaFBA2, a key enzyme in sugar and energy metabolism. BnaA05.SnRK1.1 made an effect on biomass and yield through increasing silique number. In this end, functional differentiation occurred during evolution for BnaSnRK1.1 members, which has important influence on the further understanding of rapeseed evolution and development, and also lays insight into improving yield in B. napus.