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Genome-wide association analysis of seed colour-related metabolites (mGWAS) in *Brassica napus* L

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Background:

Rapeseed is an important source of edible vegetable oil and feed protein; however, seed pigments affect the quality of rapeseed oil and the feed value of the residue from oil pressing. Seed colour is closely related to secondary metabolites such as flavonoids and polyphenols, but the molecular mechanism of these metabolites in rapeseed remains unclear.

Objective:

Seed coat pigment composition differs among varieties with different colours of seeds in Brassica crop species. To identify candidate genes involved in seed colour variation and improve the feeding value of rapeseed we screened candidate genes related to the formation of seed colour in *B. napus* through mGWAS of differential seed colour-related metabolites.

Methods:

187 high-generation inbred lines of *B. napus* with obvious differences in seed colour were used as materials, and UPLC-HESI-MS/MS was used to detect and analyse extensively targeted metabolites at 35 days seeds after flowering. First, comparative analysis was conducted to determine the stable differential metabolic components of seed colour. Second, taking the differential metabolites as the investigated traits, mGWAS analysis was performed based on the genotype information obtained from the 60K chip of *B. napus* to determine the candidate intervals. Thirdly, genomic and transcriptomic data was used to annotate and screen candidate genes.

Results:

We identified 27 differential metabolites that were extremely significantly negatively correlated with yellow-seed trait, and also had a certain significant correlation with some stored components in seeds. Based on mGWAS analysis of 27 differential metabolites in 2016cq, 2017cq, 2018cq and BLUP, we detected 609 significant association SNPs, and the contribution rate of a SNP ranged from 9.5% to 61.23%. A major candidate interval, qTN-A09-21, on chromosome A09 was identified as accounting for over 20% phenotypic variations per loci, which was significantly associated with most differential metabolites. In the genomic regions linked to the mGWAS peaks, we predicted 116 candidate genes including 47 transcription factors (e.g., MYB, bHLH, and MYC), 56 structural genes (e.g., BnaBan, BnaF3H and BnaTT4) and 13 unknown function genes, combined with analysis of differentially expressed genes.

Conclusions:

Through genome-wide association analysis of 27 differential metabolites related to seed colour in *B. napus*, we detected 609 significantly associated SNPs with the contribution rate was from 9.5% to 61.23%. Have covered the whole genome 51.16Mb, of which qTNA09-21 was a major candidate interval. Based on gene annotation and RNA-seq, a total of 116 candidate genes were selected. Our results will help enrich and improve the metabolic regulatory network of flavonoid pathway in *B. napus* and provide genetic resources and ideas for genetic improvement of rapeseed quality and new germplasm creation by genetic engineering.