

# Dissection of QTL for seed anti-nutritional compounds in *Brassica napus* L. seeds



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## 1. Background:

The world-wide demand for additional protein sources for human and animal nutrition keeps rising due to the rapidly growing world population. The by-product of oil production from oilseed rape (*Brassica napus*, OSR) is protein rich rapeseed meal (RSM)



Although the nutritional value of rapeseed protein is high, the use of RSM in poultry feeding is limited due to the presence of anti-nutritional compounds such as seed fibre and glucosinolates

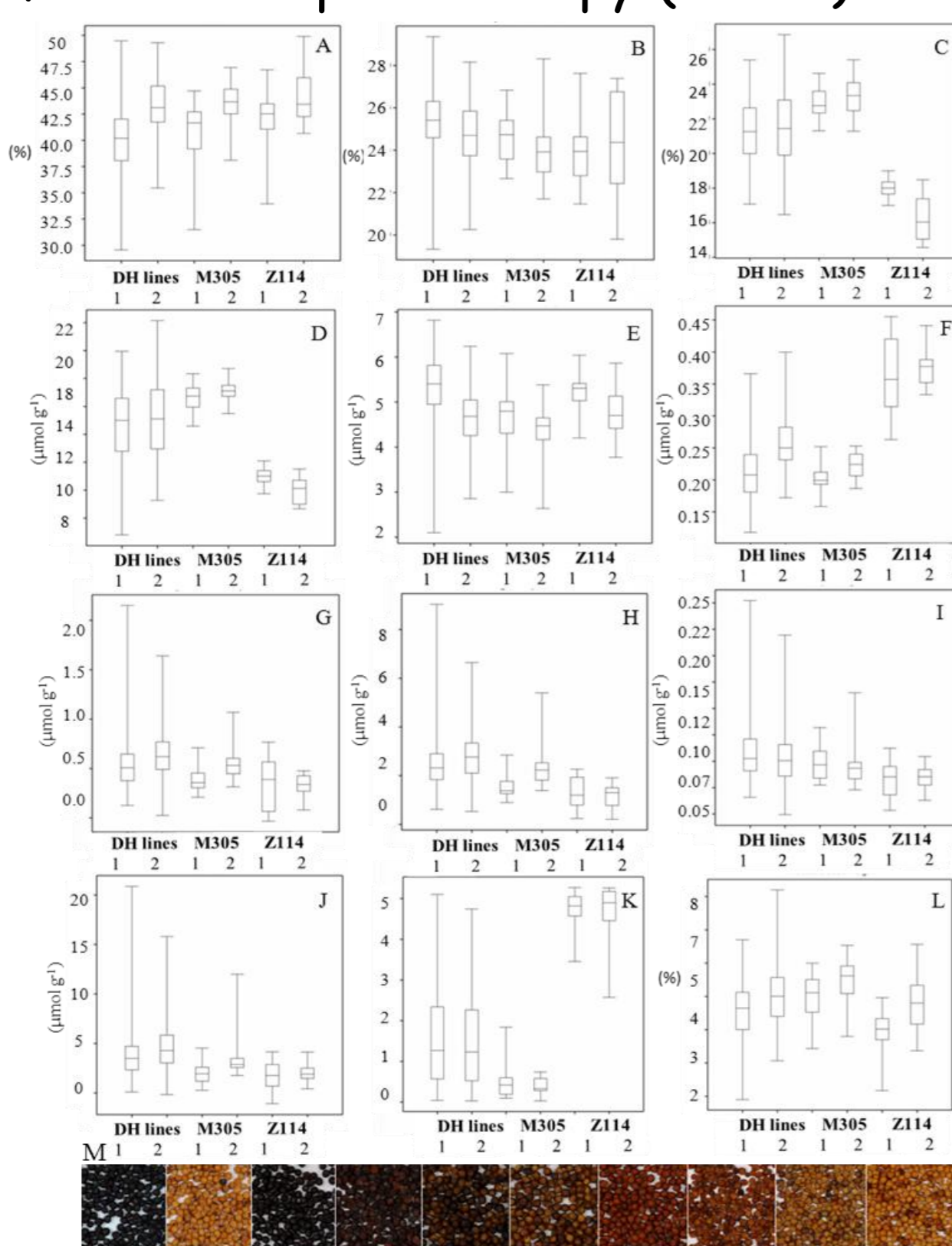


To manipulate the content of traits affecting RSM quality, it is of great importance to understand which genes regulate these traits to be able to incorporate them as molecular markers into OSR breeding improvement programs

## 2. Aim

To understand the genetic regulation of traits affecting RSM quality in *B. napus*

3. Analysis of oil, protein, fibre, glucosinolates, seed coat colour in seeds of a mapping population of doubled haploid (DH) lines derived from a cross between black-seeded M305 (DH) x yellow-seeded Z114 (DH) parental lines (Fig. 1) measured with near-infrared reflectance spectroscopy (NIRS)



A number of DH lines had higher or lower content of seed fibre, glucosinolates, oil and protein, which indicates a transgressive segregation in this population

Figure 1 Boxplots showing content of (A) oil, (B) protein, (C) NDF fibre, (D) ADF fibre, (E) 4OH glucobrassicin, (F) glucobrassicin (G) glucobrassicinapin (H) gluconapin, (I) napoleiferin, (J) progroitrin, (K) seed coat colour and (L) thousand seed weight in seeds of the M305xZ114 mapping population (DH lines) and the parental lines M305 and Z114 in two growing seasons (1) 2015/2016 (2) 2017/2018 (M). The images panel shows variation in seed coat colour in the parental lines: M305, Z114 and selection of DH lines representing colours black (DH52), dark brown (DH24), dark brown with single light colour seeds (DH17), light brown (DH32), red (DH4, DH73), yellow with single brown seeds (DH66), yellow seeds (DH71)

4. High resolution Skim Genotyping by Sequencing (SkimGBS)<sup>1</sup>: the parental lines were sequenced 50x and 78 DH lines 1-2x leading to the identification of 187 794 SNPs, using SGSautoSNP<sup>2</sup>, across the mapping population M305xZ114

5. Single loci r/QTL mapping allowed association of SNPs with all tested traits across different A and C chromosomes of *B. napus*. Candidate genes underlying QTL regions were identified and are listed in Table 1.

Table 1. List of candidate genes associated with traits affecting RSM quality: seed ADF & NDF fibre, glucosinolates, protein and oil content, seed coat colour.

Trait	QTL	Chr	SNPs pos (Darmour_v4)	LOD	Candidate genes	<i>A. thaliana</i>	Function
ADF	qADF-6	A01	1657792	4,560633	BnaA01g03570D	AT4G33330.2	Plant glycogenin-like starch initiation protein 3 - glucuronyltransferase responsible for the addition of GlcA residues onto xylan and for secondary wall deposition
ADF	qADF-3	C01	7340553	4,249909	BnaC01g11740D	AT4G19410.1	Pectinacetyltransferase family protein, carboxylesterase activity; plant type cell wall
ADF	qADF-3	C01	7340553	4,249909	BnaC01g11750D	AT4G19420.1	Pectinacetyltransferase family protein, carboxylesterase activity; plant type cell wall
NDF	qNDF-2	A01-random	2629509	2629509	BnaA01g37370D	AT3G11410.1	Protein phosphatase 2CA - negative regulator of ABA signalling, expressed in seeds during germination
Seed colour	qSCC-2	C02	25688721	4,117965	BnaC02g27750D	AT4G01070.1	UDP-glycosyltransferase, anthocyanin biosynthesis in grape
Seed colour	qSCC-2	A06	21600770	4,660092	BnaC01g12560D	AT4G21150.1	HAPLESS 6, glycotransferase activity
Sum gluk	qSGSL-2	A05	3888328	4,914159	BnaA02g09500D	AT5G54680.1	BASIC HELIX-LOOP-HELIX 105, BHLH105/ILR3-glucosinolate biosynthesis
Protein	qPROT-1	A06	22723671	4,281476	BnaA06g34380D	AT2G02040.1	PTR2, peptide transporter 2, peptide and NO3 <sup>-</sup> transporters, expressed during seed development
Oil	qOIL-5	C04	45355640	5,006092	BnaC04g45730D	AT2G38180.1	SGNH hydrolase-type esterase, lipid metabolic process

The associated genes include those known to be involved in regulation of fibre, glucosinolates, protein and oil content in seeds

## 6. Conclusions:

- The results of ANOVA revealed that the main effect of years and lines were significant for all the traits affecting RSM quality.
- Applying SkimGBS and QTL analysis in the mapping population M305xZ114 allowed identification of QTL/SNPs on different A and C chromosomes associated with traits affecting RSM quality in *B. napus*
- The identified genes contained those already known to be involved in regulation of traits affecting RSM quality and will be used as candidate genes for future analysis

7. Future work includes validation of candidate genes and development of molecular markers for MAS

## References:

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## Acknowledgments

