

# Using Novel Broccoli Lines to Understand Glucosinolate Production and Sulphur Metabolism

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UKBRC MEETING

MIKHAELA NEEQUAYE

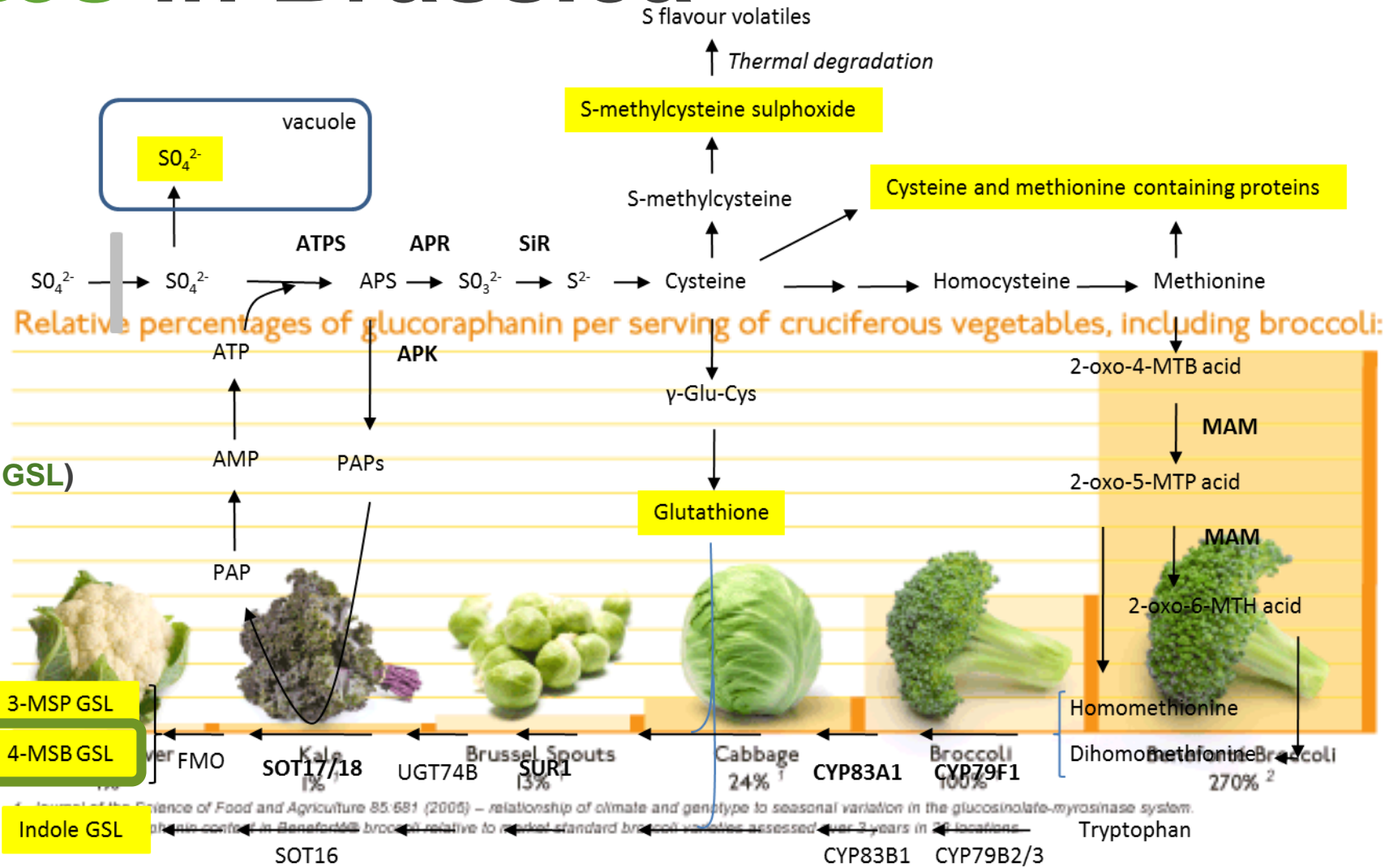
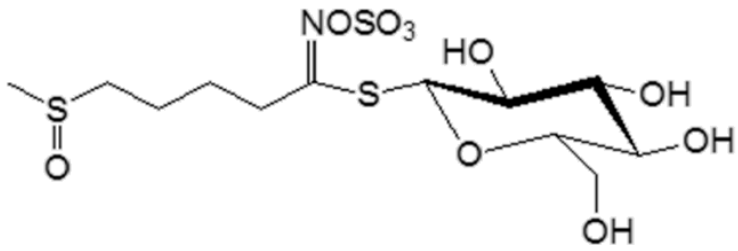
9<sup>TH</sup> MAY 2018



# Glucosinolates in Brassica

## Glucoraphanin

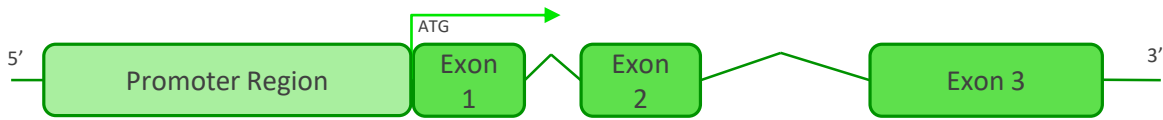
(4-methylsulphanylbutyl glucosinolate, 4-MSB GSL)



Traka et al (2013) Genetic regulation of glucoraphanin accumulation in Beneforte broccoli. *New Phytologist*, 198(4), 1085-1095.



# MYB28 in Brassica



**HAG1, AT5G61420, Bo2g161590**



Augustine, R., (2013). *Plant biotechnology journal*, 11(7), 855-866.



*E.B. 802. Arabis Thaliana. Thale-cress.*

Gigolashvili, T.,(2007). *The Plant Journal*, 51(2), 247-261.

Hirai, M. Y., (2007). *Proceedings of the National Academy of Sciences*, 104(15), 6478-6483.

Sønderby, I. (2007) *PLoS One*, 2(12), e1322.

Harper, A. (2012) *Nature biotechnology*, 30(8), 798-802.

Li, F.,(2014). *DNA research*, 21(4), 355-367.



*Pl. 28. Chou Navel. Brassica Napus L.*

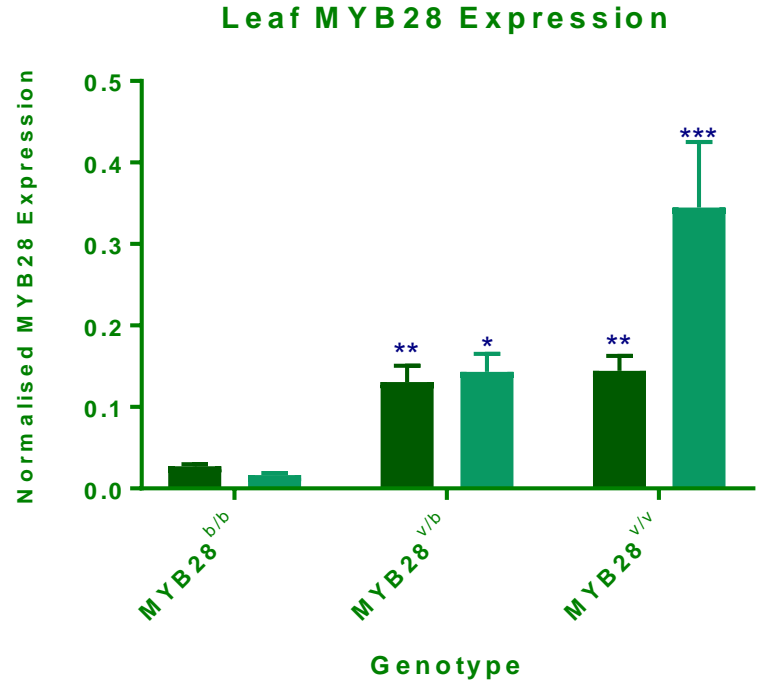
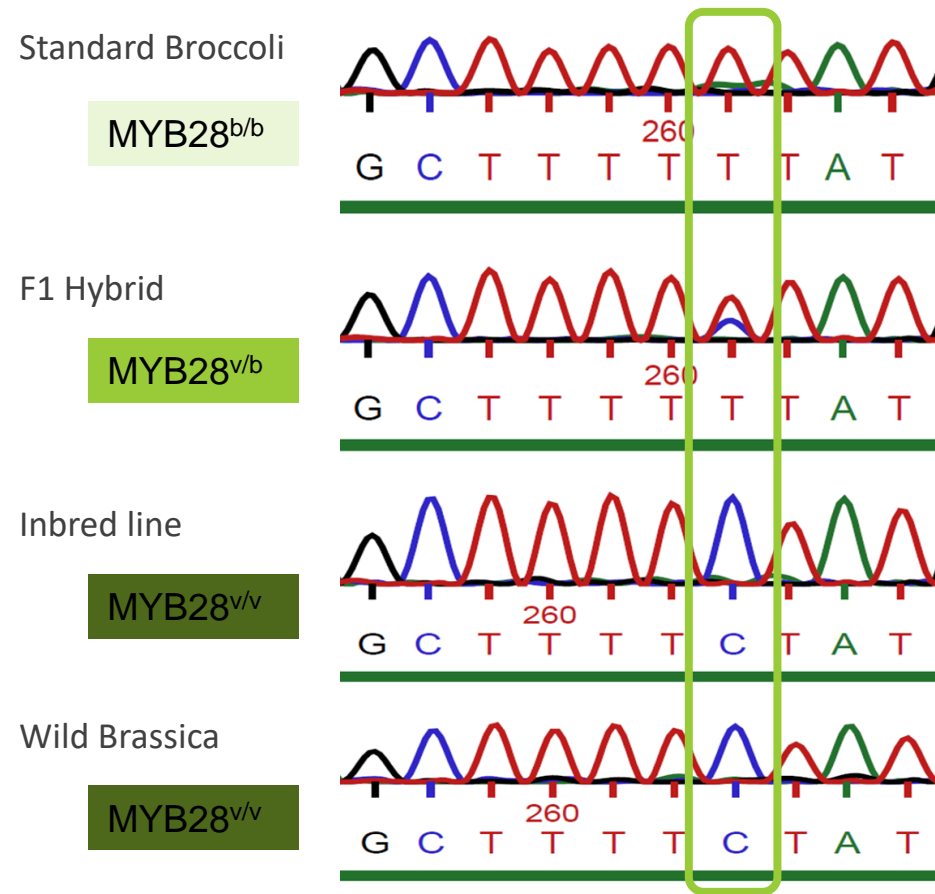


*E.B. 2276. Brassica rapa. Common Turnip.*

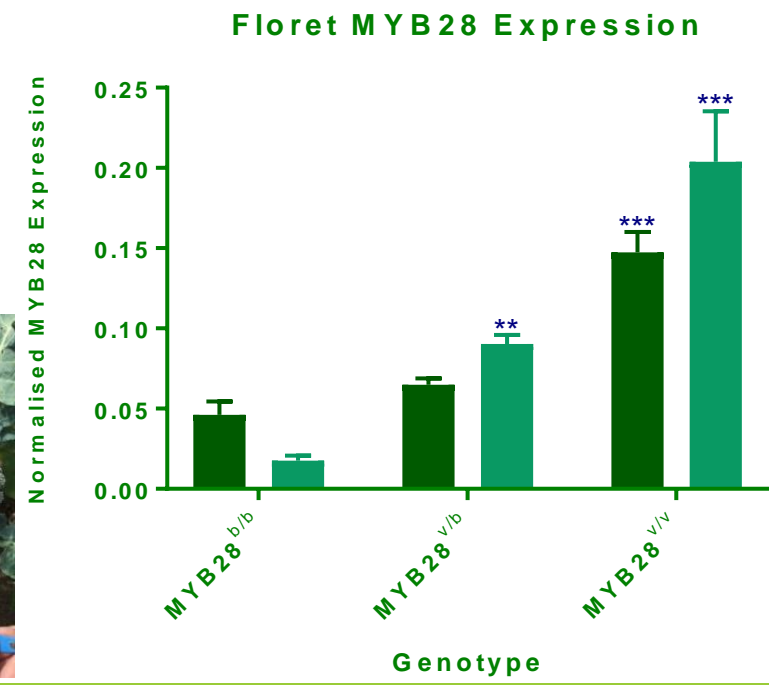
Brown, A. F.,(2015). *Theoretical and Applied Genetics*, 128(7), 1431-1447.



# MYB28 Allelic Variation



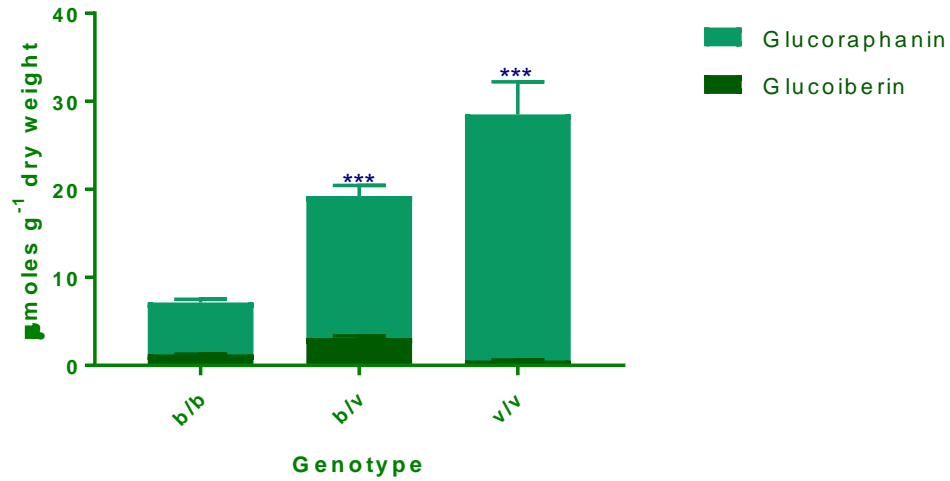
2016  
2017



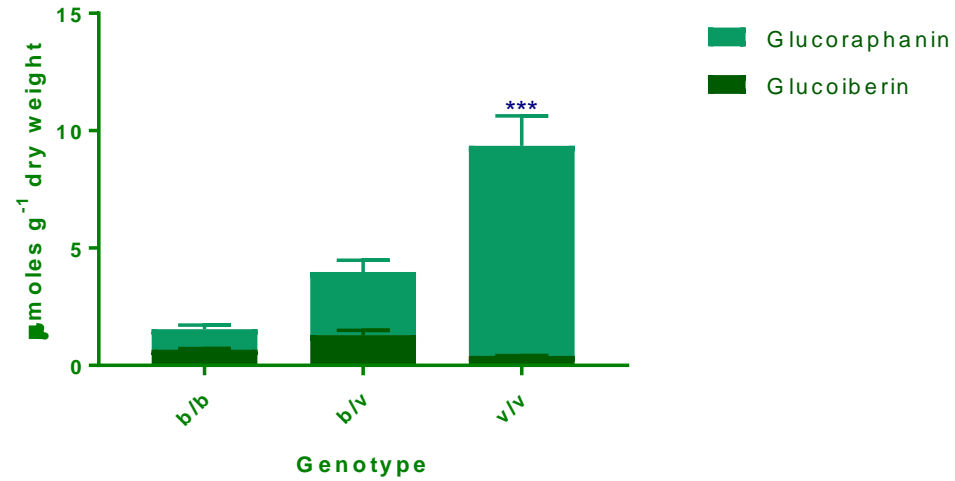
Mikhaela Neequaye, unpublished

# Methionine Derived Glucosinolates

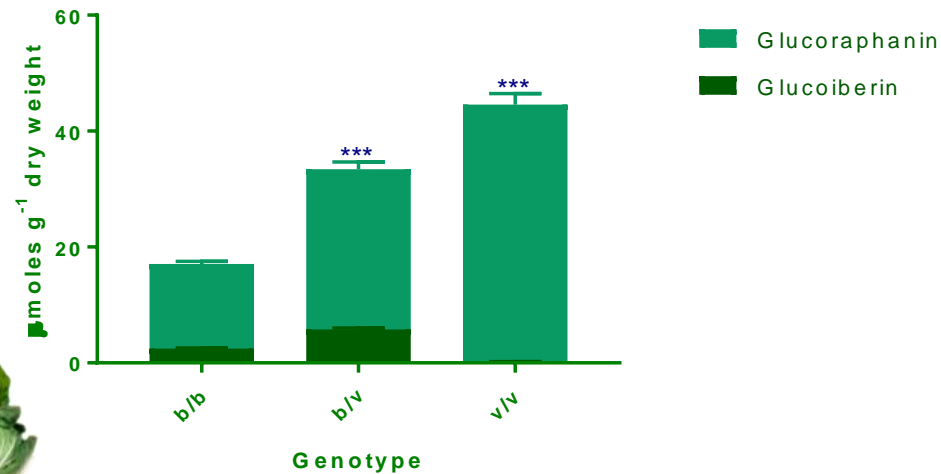
Leaf Methionine-Derived Glucosinolates 2016



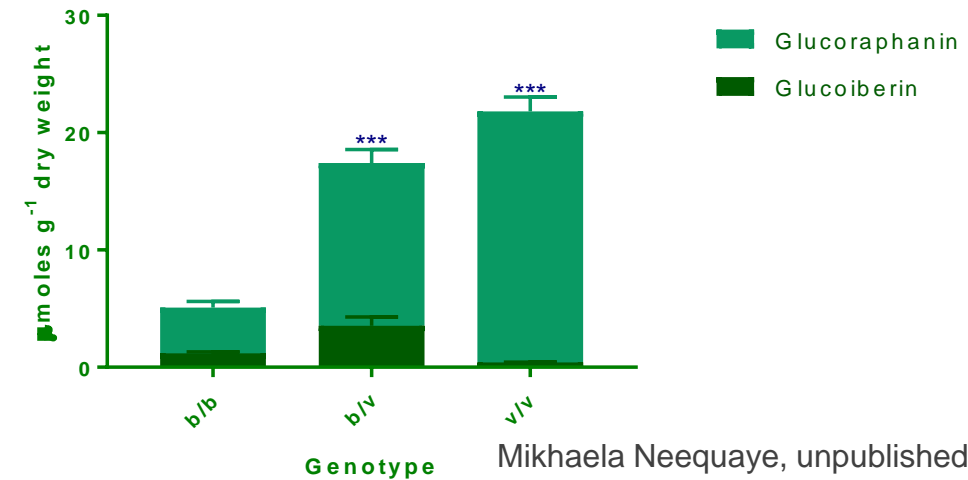
Leaf Methionine-Derived Glucosinolates 2017



Floret Methionine-Derived Glucosinolates 2016



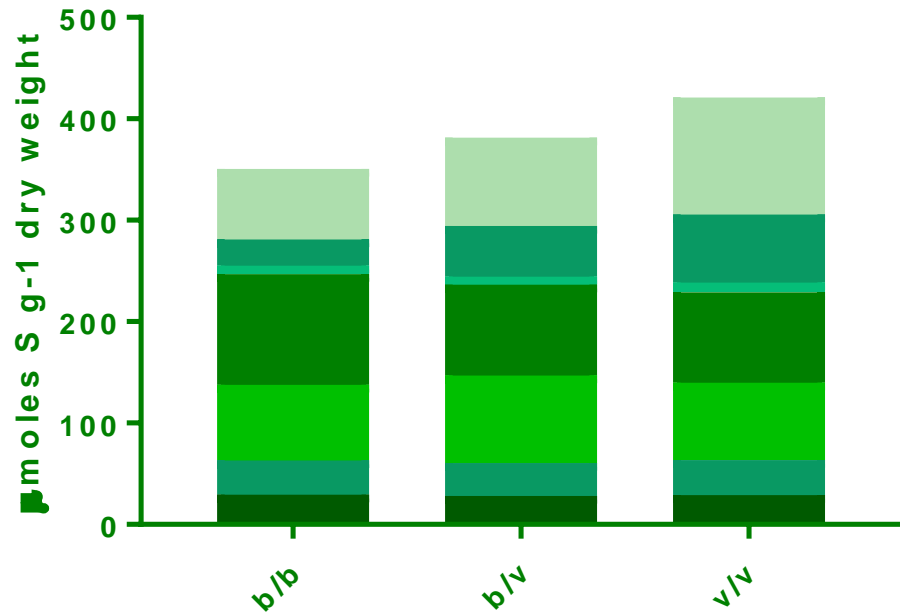
Floret Methionine-Derived Glucosinolates 2017



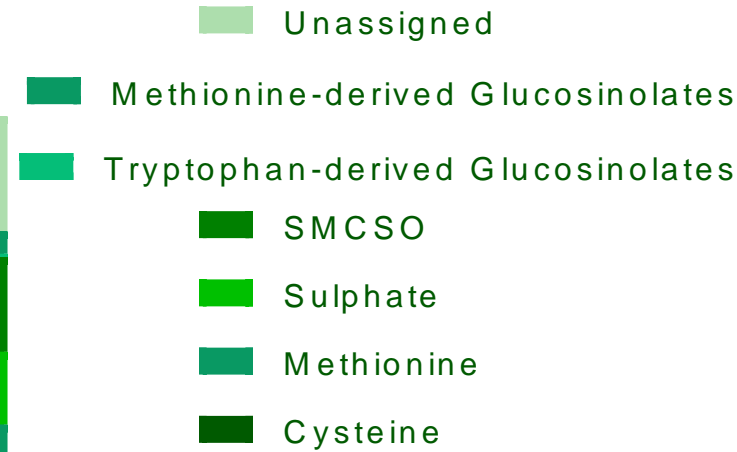
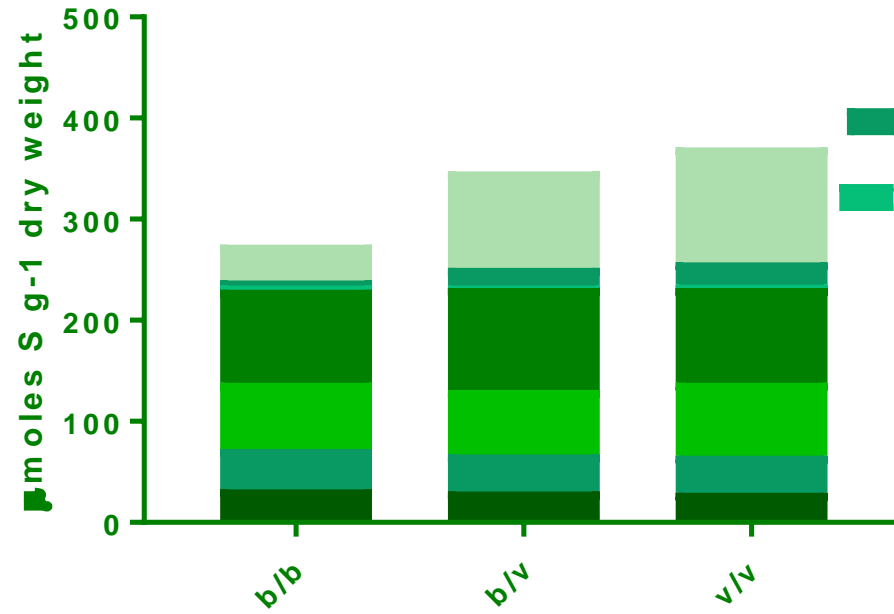
Mikhaela Neequaye, unpublished

# Sulphur Partitioning

2016



2017



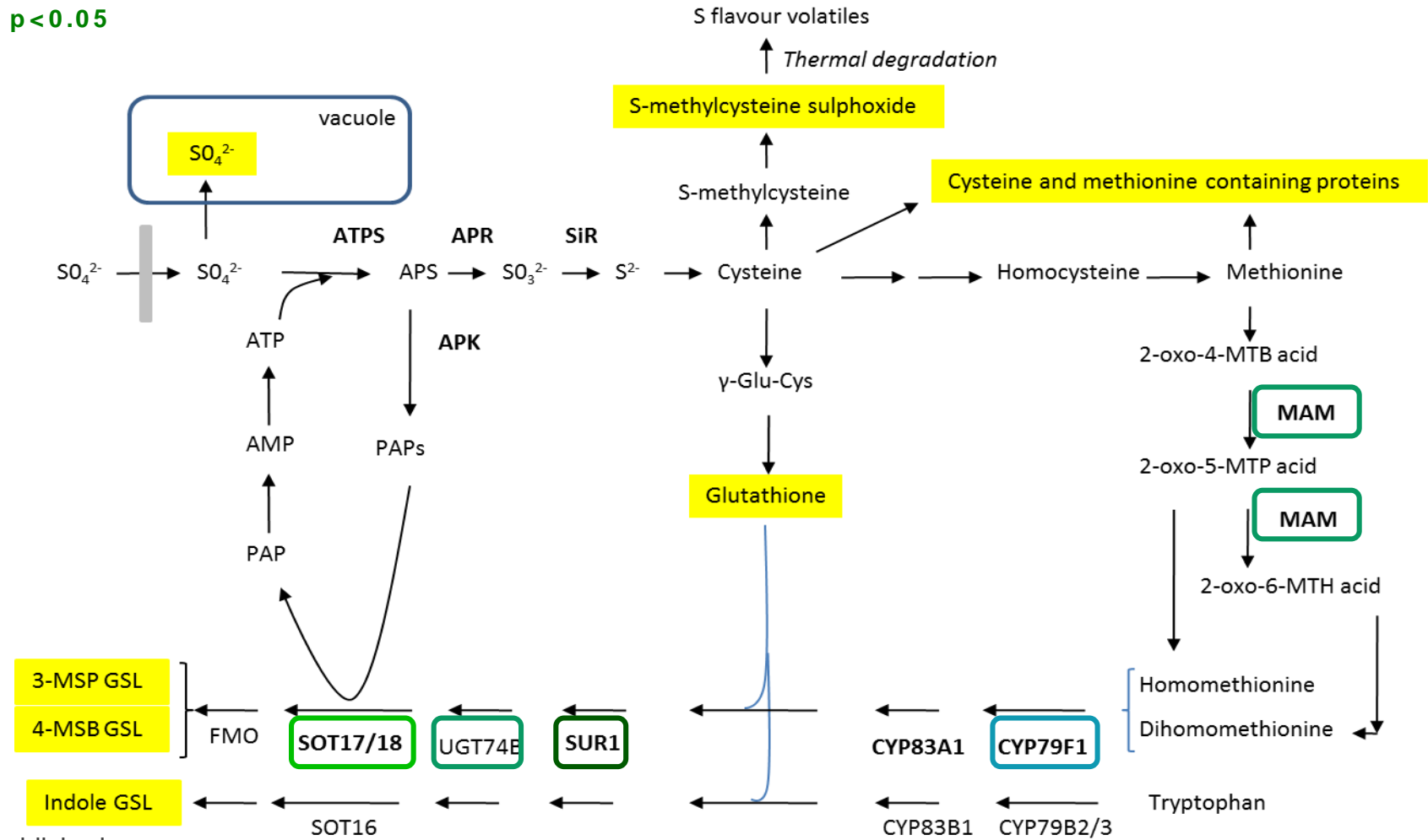
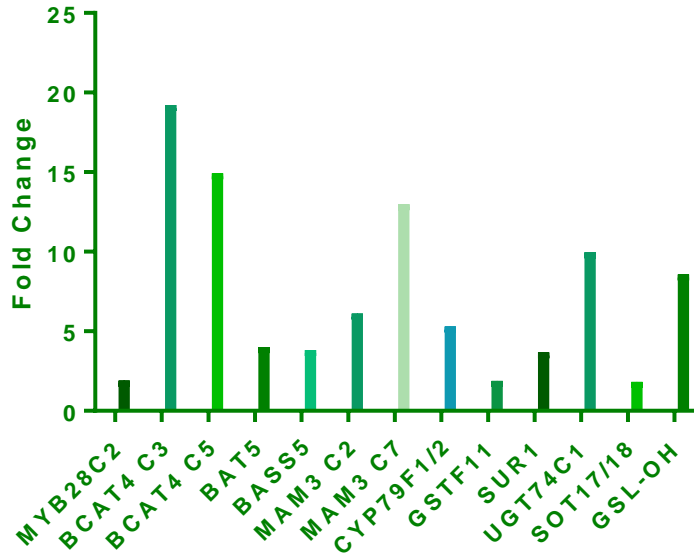
Broccoli Myb28 genotype



Mikhaela Neequaye, unpublished

# Glucosinolate Biosynthesis Genes

RNAseq Glucosinolate Biosynthesis Genes  $p < 0.05$



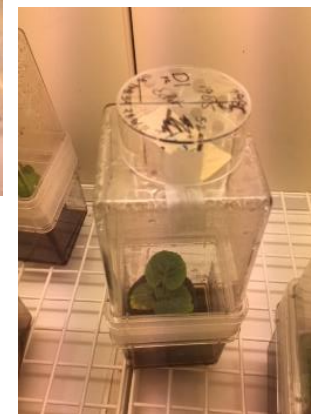
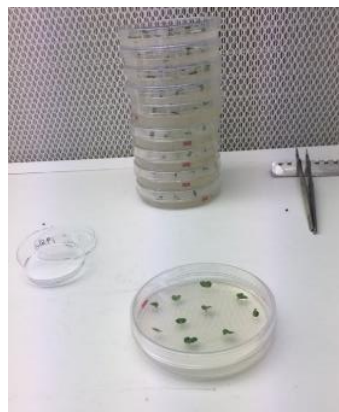
Mikhaela Neequaye, unpublished

# CRISPR Mutants

# (BRACT)

Construct Design

- 2 Constructs
- 2 guides per construct



Transformation

- 182 Transformants

Screening & Selection

- 14 Positive Plants
- Allelic series of mutations



# Acknowledgements



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