Arabidopsis TRM5 Encodes a Nuclear-localised **Bifunctional tRNA Guanine and Inosine-N^1-methyltransferase** that is Important for Growth

Pei Qin (Sabrina) Ng, @PNgsabrina
Qianqian Guo, Shanshan Shi, Diwen Fan, Jun Li, Jing Zhao, Hua Wang, Rakesh David, Parul Mittal, Trung Do, Ralph Bock, Ming Zhao, Wenbin Zhou, Iain R. Searle

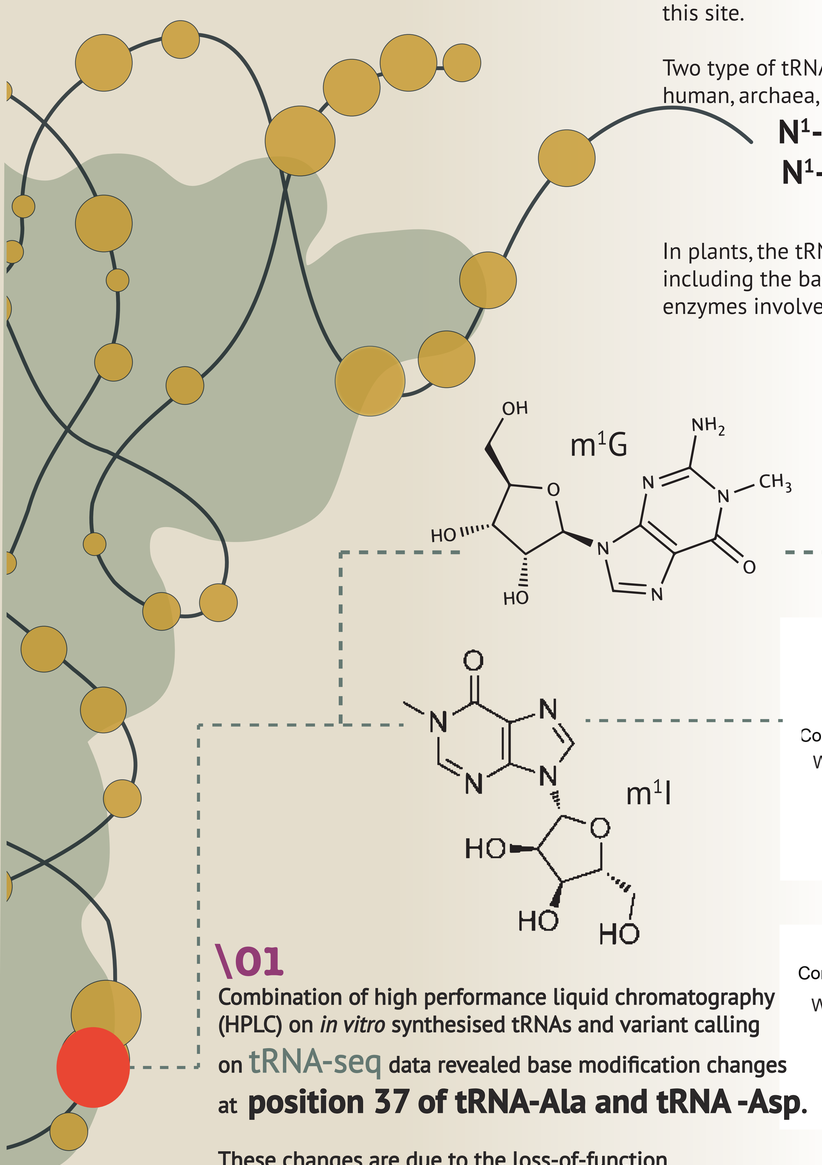
Post transcriptional modifications have been discovered in Transfer RNAs (tRNAs) in both prokaryotes and eukaryotes. However, the precise role of many of these modifications remain unknown.

tRNA anticodon loop position 37 is important to maintain translational fidelity and almost all tRNAs are modified at this site.

Two type of tRNA modications has been reported in yeast, human, archaea, and bacteria at position 37 of tRNAs:

N^1-methylguanosine (M^{1}G) and **N^1-methylinosine (M^{1}I)**.

In plants, the tRNA modification profile is yet to be defined, including the base modifications at tRNA position 37 and the enzymes involved.



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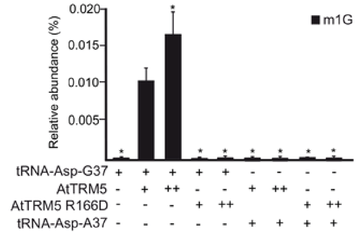
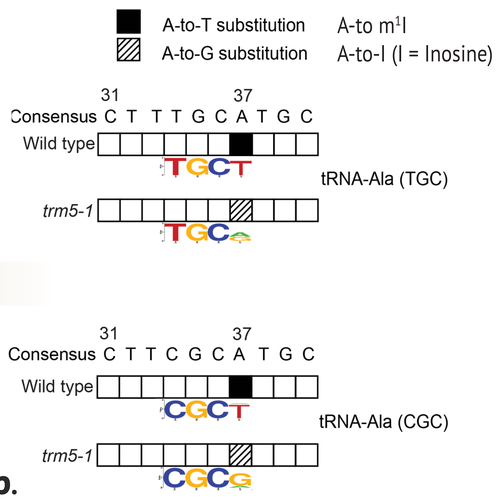
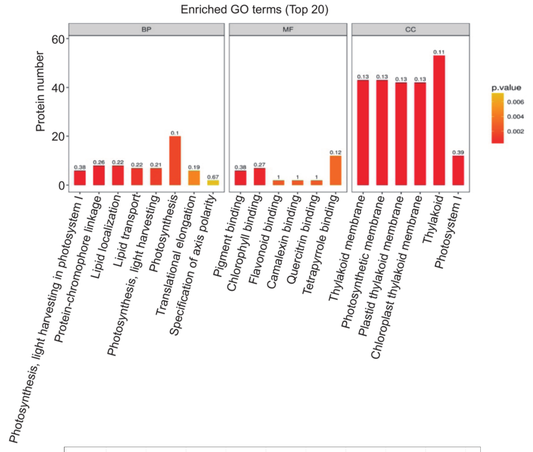
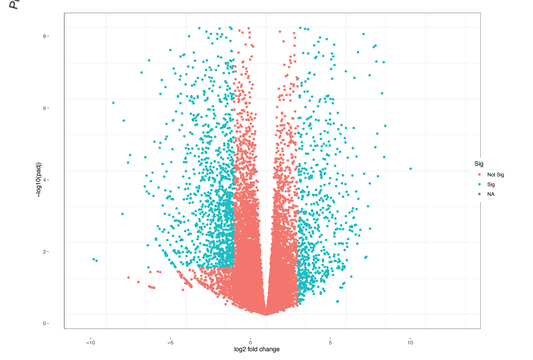
Combination of high performance liquid chromatography (HPLC) on *in vitro* synthesised tRNAs and variant calling on tRNA-seq data revealed base modification changes at **position 37 of tRNA-Ala and tRNA -Asp.**

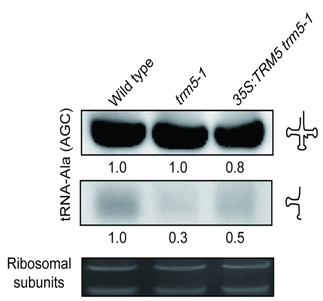
These changes are due to the loss-of-function of TRM5 methyltransferase.

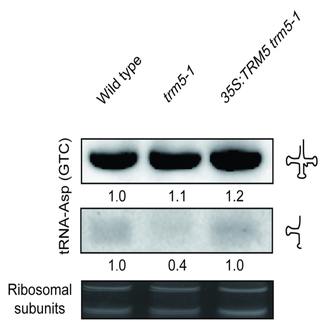


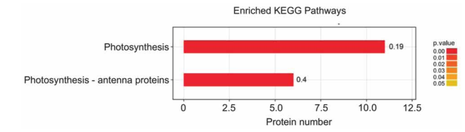












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Analysis of **proteomics** data of *trm5* mutant compared to wild type showed that 102 proteins were upregulated, and 151 proteins were downregulated.
Proteins with $\log_2$FC ⩾1.20 or ⩽0.83 and a significance level of adjusted *p*-value < 0.05 were considered to be differentially expressed.

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Differentially expressed transcripts from **RNA-seq** which have $\log_2$FC > 2 or < -2, BH-adjusted *p*-value < 0.05.
Raw reads were normalised using sample size factor for sequence depth.

580 transcripts upregulated and 1186 transcripts downregulated in *trm5* mutant when compared to wild type.

\04

Comparison of the RNA-seq and proteomics datasets showed that the only protein with DE transcripts,

Pentatricopeptide repeat (PPR-like) protein was shown to be associated with tRNA modification, tRNA fragment processing and protein translation.
(DE, differentially expressed; Non-DE, non-differentially expressed)

\05

Northern blot analysis of RNA from wild type, *trm5-1* and TRM5 overexpression line *35S:TRM5 trm5-1* showed loss-of-function of Trm5 methyltransferase affects 5' tRNA halves steady state abundance. tRNA-Ala (AGC) was inspected for changes in m^{1}G37 and tRNA-Asp (GTC) and m^{1}I37. tRNA-Arg (CCT) and rRNA were used as loading controls. The blots were probed with either tRNA-Arg (CCT), tRNA-Asp (GTC), or tRNA-Ala (AGC).

Our findings suggest that base modification changes in position 37 of tRNAs potentially affect protein translation via 5' tRNA halves.

Affiliations & Funding Bodies

THE UNIVERSITY of ADELAIDE
Australian Government
Australian Research Council