

The role of β -amylase 9 in starch degradation in the CAM plant *Kalanchoe fedtschenkoi*

Acknowledgements: Dr Anne Borland, Erin Casey

Project Aims:

- To analyse the effects of an RNAi line mutation of the BAM-9 gene on plant biochemistry
- To ascertain if BAM-9 plays a role in starch degradation in CAM

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Introduction

- Crassulacean acid metabolism (CAM) is a form of photosynthesis where CO_2 uptake occurs at night, and fixation occurs during the day, making it very water use efficient.
- Starch degradation is a crucial mechanism for CAM functioning, and the enzyme BAM-9 is thought to be involved in starch breakdown.
- In this project the gene for BAM-9 was knocked out so it was no longer expressed, and the resulting mutant plants were analysed.

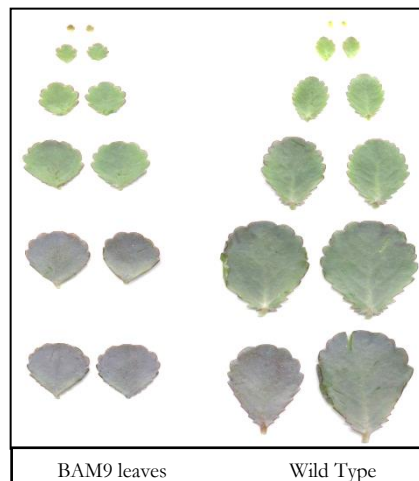


Fig.1. Comparison of leaves 1-6 of the mutant plant (BAM9) and the untreated plant (Wild Type).

Results

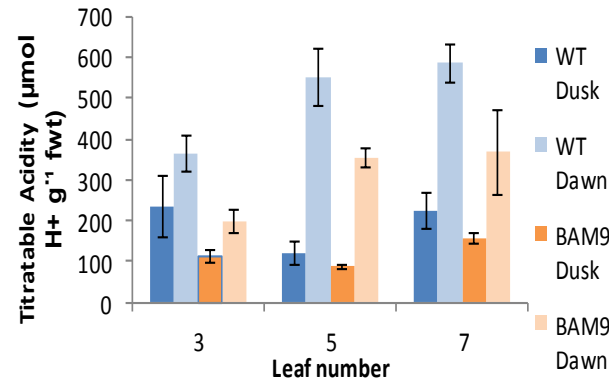


Fig.2. Comparison of the level of acidity ($\mu\text{mol H}^+ \text{g}^{-1} \text{fwt}$) between BAM9 and WT plants at dawn and dusk. Data averaged from 4 replicates, standard error bars shown. Titrations were used to measure the level of acidity.

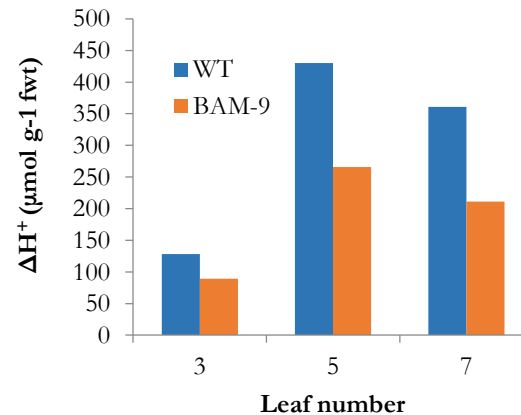


Fig.3. Comparison of the dawn/dusk difference in titrateable acidity, which gives a measurement of CAM activity. Data averaged from 4 replicates.

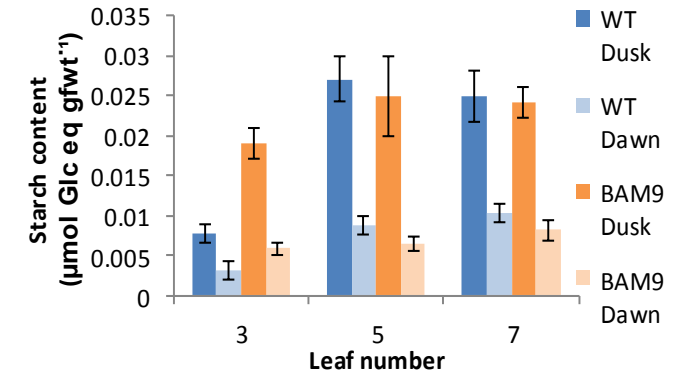


Fig.4. Comparison of the starch content ($\mu\text{mol Glc eq g fwt}^{-1}$) between BAM9 and WT plants at dawn and dusk. Data averaged from 4 replicates, standard error bars shown. Spectrophotometry was used to measure the starch content.

Discussion

- BAM-9 plants had significantly lower acidity levels and less acid accumulation overnight compared to WT plants, indicating that CAM is compromised in the BAM-9 plants.
- Starch content did not differ significantly between BAM-9 and WT plants. This suggests the lower CAM in BAM-9 was not directly related to starch turnover.
- Leaf area was reduced in the BAM-9 plants compared to wild type, indicating the importance of CAM for plant growth.
- Future work: analysis of sugar content and full growth analysis, focusing on younger leaf ages.