Review of: "Locally adaptive temperature response of vegetative growth in Arabidopsis thaliana"

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Potential competing interests: The author(s) declared that no potential competing interests exist.

This study by Clauw et al. analyzes growth responses of Arabidopsis accessions from wide-ranging latitudes to low growth temperature conditions. Accessions from colder climates show distinct patterns of growth, gene expression, metabolite accumulation, and the correlations are suggestive of local adaptation. The slower growth of high latitude plants in the cold is reasoned to reflect a reallocation of energy to cold acclimation. The findings may point to a trade-off between growth and acclimation.

It is a fundamentally important goal to better understand the basis of local adaptation of vegetative growth to low temperature and I think this study could thus be helpful.
I have no knowledge of some procedures, including the tests of broad sense heritability and adaptive differentiation, so I cannot comment on that.
There are some points which for me would need clarification:

1. Growth is quantified by measuring leaf surfaces which may miss growth in leaf thickness. It was previously shown by Adams et al. (doi: 10.3389/fpls.2016.01026) that some accessions from more northern latitudes develop thicker leaves and higher photosynthetic capacity at low temperature, compared to accessions from lower latitudes. Their apparent slower growth of rosette size at low temperature could thus mask an increased growth in leaf thickness. Did the authors consider this possibility? Do the authors, considering this, still see justification for their conclusion that reduced growth in these accessions is the likely consequence of redirection of resources towards acclimation? How?
2. Seedlings of accessions from colder climates showed larger rosettes at early stage. However, in these experiments, seedlings were grown at 21°C, which is well above normal temperatures in the colder climates. Therefore, it seems premature to me to suggest it to be an adaptive trait, as the authors do (line 250). Or is there additional support for this?
3. Transcriptomes were analyzed from plants 35 DAS. Why was this late time point chosen? Aren't these plants bolting? Differences associated with cold acclimation are probably more pronounced shorter after the transfer to low temperature.

Minor:

1. L.154. What are ‘cold acclimation genes’, how were they selected, and why are only genes considered whose expression is affected by CBFs and ZAT12?
2. Fig S6/L.168. I could use more information on what this data represents and how it would support the conclusion that the expression of certain acclimation genes would correlate with winter temperatures of accessions.
3. Please provide the AGI numbers of genes in the respective clusters A-G, depicted in Fig.5.
4. The *COL5* gene was identified as candidate gene in the GWAS, potentially contributing to the growth differences. Is it’s expression at low temperature correlated with rosette size in the accessions?