**T4149\_TANPA (n=9) , C71A1\_TANPA (n=6), C71B1\_TANPA (n=3). (Tanacetum parthenium) .**

All members of the same biochemical pathway involved in the production of sesquiterpene lactones. All mentioned in this paper: <https://pubmed.ncbi.nlm.nih.gov/24704560/> That seeks to map the biochemical pathway to produce parthenolide in feverfew (Tanacetum parthenium).

* T4149\_TANPA is likely an upstream intermediate pathway member. It is co-expressed with other sesquiterpene lactone pathway genes.
* C71A1\_TANPA is Parthenolide synthase as confirmed in the study.
* C71B1\_TANPA is a downstream Parthenolide pathway member that converts costunolide/parthenolide → 3β‑hydroxy derivatives.

**What does this mean?** – Implies that multiple members of the parthenolide/ sesquiterpene lactone biosynthetic pathway are absent. Which would suggest major inhibition/downregulation of the pathway.

**What is the functional role of parthenolide and related compounds? –** Multiple papers point to parthenolide compounds playing a major role in defence in plants, especially daisy species in defence against herbivores. They have been described as a potent anti-herbivory agent

* <https://arxiv.org/pdf/2402.00548>
* <https://poisonousplants.ansci.cornell.edu/toxicagents/sesqlactone/sesqlactone.html>
* <https://www.tandfonline.com/doi/full/10.1080/07352689.2016.1145956>

A few other roles mentioned but primary role seems to be defence against herbivores and plant-eating insects.

**Link to Darwin’s Daisies.**

The Galápagos Islands have no native mammalian herbivores and insect diversity may be lower and less specialized. These secondary compounds are difficult and expensive to produce so may have been selected against due to the lack of native herbivores. This paper has shown that a member of the Asteraceae family native to the Canary Islands showed a very limited profile of secondary metabolites unlike mainland Asteraceae.

* <https://pubmed.ncbi.nlm.nih.gov/27272544/>

Whilst looking into the terpenoid pathway downregulated genes and defence response I also noted that a few of the genes were involved in **Gibberellin** biosynthesis:

**KS1\_STERE (n=3), KS2\_STERE (n=2)**

Ent-kaurene synthase 1 (KS1) and Ent-kaurene synthase 2 (KS2) are key enzymes in the gibberellin (GA) biosynthetic pathway. Studies have shown that *Arabidopsis* with knock-down/out’s of KS1 result in dwarf phenotypes:

* <https://doi.org/10.1104/pp.116.4.1271> (old but seems relevant).
* <https://doi.org/10.1038/s41438-020-00399-6>
* <https://academic.oup.com/hr/article/doi/10.1038/s41438-020-00399-6/6445672>
* <https://journals.plos.org/plosgenetics/article?id=10.1371/journal.pgen.1008562>

**What does this mean** – inhibition of these enzymes suggests that as well as a global downregulation of terpenoid biosynthesis, the specialised branch leading to the production of gibberellin may also be down-regulated.

Mutants in KS1 show **dwarf phenotypes**, as do downstream pathway components (3 and 4). Gibberellin biosynthesis is a specialized branch of the broader terpenoid biosynthesis pathway. Which is why these genes are associated with the “terpenoid biosynthesis” GO term.

**Functional role of Gibberellin:**

Interestingly this pathway is typically involved in the formation of dwarf phenotypes, so it is surprising to see these genes presenting gene loss, as Darwin’s daisies on the Galapagos show larger forms and gigantism. (<https://www.nature.com/articles/s41467-022-31280-w>).

Gibberellin is a key plant hormone involved in regulation of growth. <https://www.sciencedirect.com/science/article/pii/S1084952119301600>. One main role is promoting stem elongation.

**Link to Darwin’s Daisies.**

As Darwin’s daisy is an island species, there may be selection pressures against gibberellin to reduce plant height to overcome challenges such as reduced nutrient availability, high winds.

Another idea could be that even though gibberellin signalling is reduced, plant size/height may be caused by upregulation in different pathways and down-regulation of this pathway is a trade-off effect involved in the downregulation of sesquiterpene lactones.

**Possible links to phenotypes:**

1. Assess the extent of sesquiterpene lactone pathway loss across species and island. On smaller islands (with presumably less predators) can we observe a more complete loss of the pathway/ is some signalling retained in larger islands?
2. Using the species metadata for each sample, see if absence of gibberellin biosynthesis related genes occurs only in certain species. These species could then be cross-referenced with species height phenotype data that could give further insight into whether gibberellin is causing any change in species size. I am not sure whether all the 18 species in the dataset show island gigantism. Perhaps ones that don’t are the ones that have gibberellin downregulation.
3. We also have “habitat” data. We could also check to see whether production of sesquiterpene and gibberellin have any association with their habitat. How this would link with phenotype is less clear though.