# Regulatory mechanisms of flowering in Sturt's desert pea (*Swainsona formosa*)

by

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# Statement of originality

I certify that the work presented in this thesis has not been previously submitted for a degree or diploma at any other higher education institution.

To the best of my knowledge and belief, this thesis contains no material previously published or written by another author except where due reference is made.

Tanya Tapingkae

# List of abbreviations and glossary of terms

| | |
|---|---|
| ABA | abscisic acid |
| BA | benzyl adenine |
| BAP | benzylamino purine |
| C | carbon |
| $[CH_2O]_n$ | carbohydrate |
| EDTA | ethylenediamine tetra-acetic acid |
| FW | fresh weight |
| GA | gibberellin |
| $GA_3$ | gibberellic acid |
| IAA | indole-3-acetic acid |
| IBA | indole-3-butyric acid |
| LD | long-day |
| LSD | least significant difference |
| mRNA | messenger ribonucleic acid |
| MS | Murashige and Skoog (1962) basal medium |
| N | nitrogen |
| NAA | α-naphthalene acetic acid |
| PAGE | polyacrylamide gel electrophoresis |
| PAR | photosynthetically active radiation (usually μmol $m^{-2}s^{-1}$) |
| PBZ | paclobutrazol |
| PGR | plant growth regulator |
| ppm | parts per million |
| PPFD | photosynthetic photon flux density (μmol $m^{-2}s^{-1}$) |
| rpm | revolutions per minute |
| RNA | ribonucleic acid |
| Rubisco | ribulose-1, 5-bisphosphate carboxylase/oxygenase |
| SD | short-day |
| SDS | sodium dodecylsulphate |

# Abstract

*Swainsona formosa* (G.Don) J. Thompson (Sturt's desert pea) is an Australia native legume which has potential as an ornamental pot plant and is also suitable for hanging baskets and as a cut flower. Accurate identification of the timing and control of growth stages is critically important in making management decisions for floricultural crops. One of the impediments to the commercialisation of *S. formosa* as a pot plant is its inability to produce flowers under low light conditions.

The apical meristems of *S. formosa* were investigated by stereomicroscopy and scanning electron microscopy to identify flowering time and stages of floral development. Conversion from the vegetative to the reproductive stages began within 40 to 46 days after seed germination for axillary branches and 46 to 52 days for central stems. The order of floral organ initiation within each whorl is unidirectional, except for the petal whorl, which is simultaneous; the flower is organised into five whorls and shows a pentamerous arrangement of sepals and petals, ten stamens in two whorls and a central carpel. This is the first description of the complete floral ontogeny of a member of tribe Galegeae in papilionoid legumes.

The effect of high light intensity (800 ± 50 μmol $m^{-2}s^{-1}$) and low light intensity (150 ± 10 μmol $m^{-2}s^{-1}$) on the flowering of *S. formosa* was investigated, with particular emphasis on measuring the changes in sugar concentrations in the transition stages of the shoot apices from the vegetative to the floral, using high-performance liquid chromatography. Plants grown under high light initiated flowers within 45 days from seed germination, while plants grown under low light intensity remained vegetative and produced no flowers during the 60-day experimental period. Trace amounts of glucose (0.52 mg $g^{-1}$) were detected at the beginning of the transition from the vegetative to floral stage (40 days after germination) in the apices of plants grown under high light intensity conditions; this increased to 2.70 mg $g^{-1}$ with progressive floral development. No glucose was detected in the shoot apices of plants grown under low light during the experimental period. The timing of changes in sucrose quantity matched the timing of morphological changes, indicating a key role for sucrose in flower development. The changes in the sugar composition of the shoot apices are associated with quantitative

changes in sugar translocation and changes in enzyme activity related to floral transition. Also, the balance between glucose, fructose and sucrose is important for flowering to occur. Adequate amounts of glucose is required for floral initiation and development in *S. formosa*.

Among many factors, the Carbon:Nitrogen (C:N) ratio is considered to be one of the most important physiological signals that induce flowering of *S. formosa*. Reduced %N and %C at the early transition stage, and an increased C:N ratio in the shoot apices, could be used as indicators for stage changes. To flower, plants need to maintain an appropriate C:N ratio. The mode of N acquisition (fixation versus assimilation) seems to influence %N accumulation in the shoot apices of low light grown plants.

To reach a more precise conclusion on the effects of carbohydrates on flowering of *S. formosa*, the effects of exogenous sugar applications on the flowering of *S. formosa* were tested. Several experiments (spraying, brushing, injecting and absorbent cotton on apex), on the application of exogenous sugars, were conducted to identify the correct methodology of application, and the types and concentrations of sugars needed to induce flowering of *S. formosa* grown under low light intensity conditions. Results confirm that *S. formosa* requires adequate high light intensity for flower induction and development. All the sugar treatments using exogenous application methods failed to induce the transition to flowering in *S. formosa* plants grown under low light conditions. The failure to flower depends on many factors, such as the use of ineffective concentrations of sugars along with inappropriate application methods. *In vitro* flowering was achieved within 4 to 6 weeks by placing shoot apices from high light grown stock plants on Murashige and Skoog medium supplemented with 3.0 to 4.5% sucrose. This could be the result of an interplay of factors, including environmental conditions under which the explant source was grown and the types and concentrations of exogenous sugar used in the culture medium. This is the first report of *S. formosa* producing flowers *in vitro*. It is suggested that future work on exogenous application of sugars be expanded by using *in vitro* techniques.

The key stages in the transition of *S. formosa* plants to flowering is correlated with protein concentrations and patterns. High light intensity caused changes in the concentrations and types of proteins that promoted flower initiation and differentiation.

These protein changes were absent from the vegetative apices of high light plants and all the apices of plants grown under low light. Further studies on protein morphogenetic markers, as well as their genes and conditions for expression, are among the most promising approaches to the regulation of morphogenetic processes in this and other plants.

# Table of contents

# List of tables

# List of figures