Date:

Name:

## Topic:Herb MicropropagationInduction of the Culture

<u>Material</u>: Leaves of African violet (*Saintpaulia ionantha* Wendl.) and Cape primrose (*Streptocarpus x hybridus*) (Family *Gesneriaceae*)

<u>Medium</u>: F1 = M-S basal salts, B5 vitamins, BA 1mg.l<sup>-1</sup>, NAA 1mg.l<sup>-1</sup>, sucrose 20 g.l<sup>-1</sup>, 0.65% agar, pH 5.5. Recalculate concentrations of plant growth regulators and sucrose to molar concentration (= SI units).

## Procedure:

- 1. Remove fully expanded leaves from a stock plant and wash them with distilled water.
- 2. Disinfections of the leaves in 70% ethanol for 30 sec to1 min.
- 3. Disinfections in aqueous solution of 15% commercial bleach SAVO (v/v) by gentle agitation for 15 min.
- 4. Rinse 3 times in sterile distilled water for 3 min.
- 5. Transfer disinfected leaves from the bottle on a sterile Petri dish in a flow hood.
- 6. Excise  $1 \text{ cm}^2$  segments of the leaf.
- 7. Place the segments with the adaxial surface of the leaves (**upper layer of epidermis of the leaf**) in contact with the agar medium F1.
- 8. Record the number of segments within a tissue culture jar and the number of the jars.
- Culture under the light (cool white fluorescent light, photoperiod 16/8 hours, PAR 30 μmol.m<sup>-2</sup>.sec<sup>-1</sup>) at 22°C for 4 weeks.

## Evaluation:

Control contamination of the explants in the following period of cultivation. The contaminated cultures should be discarded.

Evaluate frequency of segments with organogenic shoot bud formation (direct organogenesis) and the number of segments with proliferating callus stage (indirect organogenesis). Record the multiplication rate (total number of shoots per explant).

http://www.africanviolet.org.au/streptocarpus.htm

http://www.robsviolet.com/streptocarpus1.htm

http://www.streptocarpussociety.org.uk/

http://en.wikipedia.org/wiki/Streptocarpus

http://www.ext.vt.edu/departments/envirohort/factsheets/pottedplants/strept.html