



Woolf Orchid Culture

Orchid Flasking for the Amateur

by John Woolf

Some basics about in-vitro culture for the Amateur.

POLLINATION OF PLANT

One takes pollen from the POLLEN PARENT and places it onto the stigmatic surface of the POD PARENT.

Pollen is found in a variety of places on the different Orchid Genera and can be dry (as Cymbidium) or wet (as Paphiopedilum).

Pollination may take a few days or a few weeks depending on genera

Fertilisation occurs when the pollen has grown down into the Ovary (seed capsule) this usually takes approximately two thirds of the period to maturity. After this GREEN POD culture can be started. Once the seed capsule has split one has to use the DRY SEED technique. It is far easier to flask Greenpods you simply trim off any damaged areas and old flower parts sterilise in 2% sodium (1/2 strength Snowwhite or similar) take into your clean air cabinet (or Home made box) cut seedpod open and decant seed into sterile flasks. For dry seed one has to sterilise individual seed then wash in sterile water before placing into sterile flasks this can result in over sterilisation and you can kill the seed or under sterilisation and you can lose the seed to Fungi / Bacteria.

MERICLONING OR MERISTEMMING

This is the taking of a piece of tissue from a plant and reproducing many exactly (in theory) the same. The Explant as it is known will be washed in alcohol, sterile water, sodium sterile water trimmed washed again and this treatment continued until you have a sterile minute piece of tissue to place in the sterile flask. Once it begins to grow it is then placed onto a multiplication media and continually cut up until you have produced the required number of plants.

THE FLASK

This can be any glass plastic or similar jar that has a lid with a good seal and can be autoclaved in a pressure cooker. The lid has a hole drilled with a rubber tube packed with non-absorbent cotton wool inserted as a breather (or you can use a Teflon patch). The jar etc. MUST be clean, soak in sodium for 30 minutes and then rinse any dirt and sodium off as the sodium will kill plants if left in flask.



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THE MEDIA

This can be purchased from firms such as Orchid Media Lab who specialise in orchid flasking media (and others see Orchids Australia) or make your own.

Some ingredients that we use are available only on permit from A Q I S.

A simple media.....

- (1) 2 1/2 grams of G F 9 fertiliser - available from Groforce
- (2) 5 beads of Calcium Nitrate - available from Groforce
- (3) 6-7 grams of Agar Agar powder -available from health food shops
- (4) 20 grams of sugar (less for some genera) fructose or glucose
- (5) 1/4 teaspoon activated charcoal (optional but required for some genera)available from Selbys.
- (6) 1 drop of Best Grow - available from Bacto (we have tried Nufarm Growth Formula with equal success)
- (7) Peptone (optional) you will need a permit for this)
- (8) 100 grams of ripe Cavendish bananas (put through a blender). Other fruit used for specific purposes are coconut milk, potato, tomato, pumpkin etc.

Make up to 1 LITRE with Demineralised water (available from supermarkets)

STERILISING THE MEDIA

This is then placed on a stove and brought to near boil stirring continuously decant into flasks and seal with prepared lid. This is then placed into pressure cooker for 15 minutes at 15 p s i to sterilise. Leave to cool remove and tighten loose lids and place in a clean dry area until used. (A new foam esky or icepak box is good)

THE CABINET

This can be a CLEAN AIR UNIT costing from \$2.500.00 to \$10.000.00 or a home made unit costing \$50.00 depending on how serious you get or how many flasks you are going to produce.

Here are two examples.

- (1) a wooden metal or perspex box with a sliding glass top and two armholes.
- (2) a foam icepak box with the top cut out and glass inserted with two armholes.

METHODS OF FLASKING

There are two different methods used

- (1) Using sodium hypochlorite
- (2) using a Bunsen Burner

The inside of the unit should be washed with Alcohol or Sodium before starting.



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Equipment needed includes

- 1a. A plastic food container to hold sodium
- 1b. Or a Bunsen Burner
2. A petri dish or a saucer
3. A scalpel or knife (Stainless Steel)
4. A pair of tweezers (Stainless Steel)
5. A pair of rubber gloves and some Talcum Powder
6. A charcoal impregnated face mask
7. A plastic backed apron or some old clothes
8. Some cloth that will stand sodium (Kimtex Wipes if available)
9. You will find other tools as you become more adapt.

STORAGE OF FINISHED FLASKS :-

Once finished replating (placing seedlings on agar in flasks) place flasks in a dry area where they will get reasonable light from all sides (if possible or turn occasionally) or place under artificial lights.