Technical improvement of the plantform bioreactor for in vitro culture and large scale micropropagation.

Intellectual rights

Trademark: Plantform bioreactor
(applied)
Patented process

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Description of the invention

Conventional micropropagation is expensive and labour intensive. The production systems utilized the last 20 years are still in use by most laboratories. Until now few laboratories have been truly profitable. However with development of a new biological pathway using liquid culture systems in bioreactors it is now possible that micropropagation become profitable. A small in vitro culture container, named RITA, based on this system was developed by a French company and patented in France 1995. This container was mainly developed for somatic embryogenesis and is less suitable for large scale in vitro plant propagation. We have now developed a commercially attractive bioreactor (plantform) for large scale in vitro plant production. The bioreactor has been designed in a way that is easy to handle, it has low weight, it is transparent, it is autoclavable, the gas exchange including oxygen and carbon dioxide can be controlled using air pumps and a timer. The units can be placed above each other saving place in the climate chamber. Fig 1 shows the construction and the details of the bioreactor.

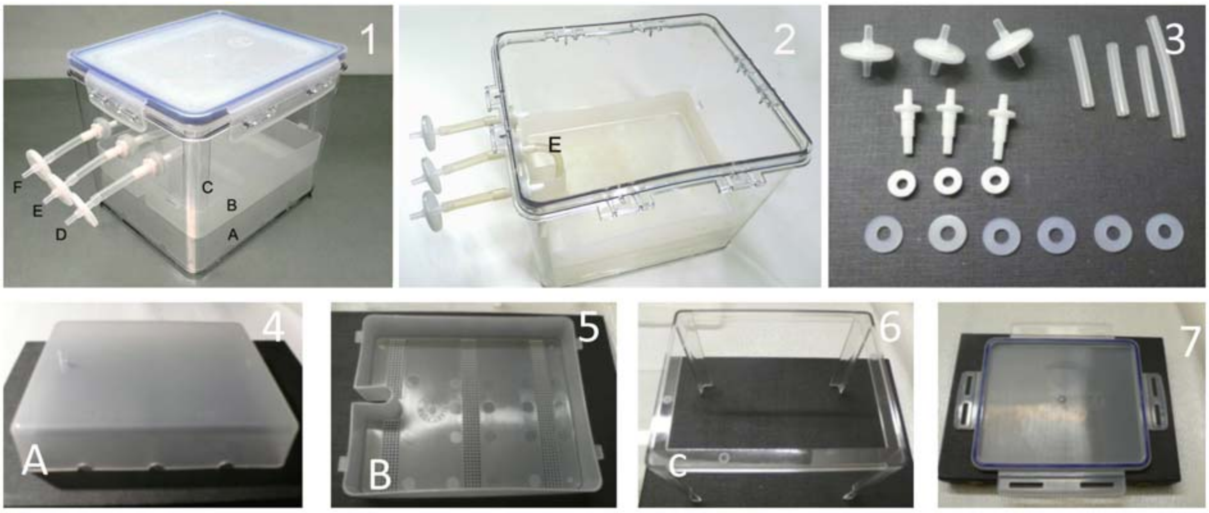

Fig.1.1: Bioreactor and its different parts numbered from A-F. 1:2 Outer container with 3 inlets/outlets for gas exchange, E shows the middle filter connected to a plastic tube on the inner chamber, 1:3 filters, plastic tubes, nuts, clamps and silicon rings to be connected to the 3 inlets/outlets on the outer container, 1:4A inner chamber with 3 grooves on the long side and connection to the middle filter, 1:5B basket with 3 rows of small holes, 1:6C frame with 4 legs, 1:7 lid with 4 flaps and an inner silicon ring.

Description

The bioreactor consists of a body made of polycarbonate which is transparent and can stand heating 120^0 C. The size is 180x150x150 mm. Gas exchange is controlled through three inlets/outlets anchored to the side by nuts and clamps through holes in the body and sealed with heat resistant silicon o-rings. The gases are passed through sterile filters connected by silicon tubes to the inlets. The middle filter is connected to a plastic tube on the inner chamber and goes through the basket (fig 1:2 E)

An inner chamber with 3 grooves on the long side and 2 grooves on short side is placed at the bottom of the bioreactor. It is designed in a way to allow nutrients to rise efficiently when a pressure is applied (fig 1:4 A).

A basket containing the plant material is placed above the inner chamber. The small holes in the basket is placed in three rows which allow the nutrients to flow efficiently through the basket (fig 1:5 B)

A frame with four legs is placed at the bottom of the basket to press down the basket when a pressure is applied. (Fig1:6 C)

The body is closed by a lid which is air tight using silicon seal inside a groove of the lid. The lid is a snap-on lid easy to put on and remove (1:7).

The chamber, basket, legs and the lid are made of polypropylene, more flexible plastic, which can be autoclaved at 120^0 C.

After sterilizing all parts the bioreactor is ready to use. The body is filled with generally 500ml of nutrient solution under the inner chamber. Then sterile plant material is placed on the basket. The frame with the four legs is placed on the basket and the lid is secured at the top of the body. The filters are connected to 2 timers, and 2 air pumps via silicon tubes to regulate gas exchange. When a pressure is applied to the middle filter the nutrient solution is forced upwards and covers the plant material. When pressure is relieved the nutrients will drain back through the holes in the basket. In order to facilitate air to go out from the tube connected to the middle filter an electric valve can be applied. No liquid will be left on the basket which is important to avoid hyperhydricity. The immersion time and frequency like duration and frequency of ventilation are set by the timers. Since the body has feet at the bottom several units can be placed above.

The advantage of the plantform bioreactor.

The bioreactor will reduce production costs and improve plant quality.

1. The use of liquid nutrient solution will eliminate costs for agar, the gelling agent, which is used in conventional in vitro propagation.

2. The size of the bioreactor will allow a larger amount of plants in each unit which will reduce labour costs for transferring plants to new containers.

3. The amount of nutrient supply can be regulated due the different growth phases. Bigger plants require more nutrients than small plants. Better quality of plants.

4. The gas phase in plant tissue culture is important and with this system enrichment of oxygen and other gases can be regulated. Several plants produce ethylene especially during rooting after application of auxin. Deleterious gases can be removed by this system.

5. The rooting will also take place in liquid medium which facilitate planting in soil afterwards. Acclimation of in vitro plants when transferred to soil condition generally cause great losses of plants. This is caused by sensitivity to low humidity after in vitro production. With this system the stomata in the plants are functional which means that acclimation in soil is improved.