Bioreactors in Plant Cell Culture

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Bioreactor is a device or system designed to grow cells or tissues in the context of cell culture. It is a closed system used for bioprocessing, that supports the growth of cells or tissues. The process can be aerobic or anaerobic. However for plant cell culture, growth of cells occurs under aerobic condition.

Bioreactor is one of the prerequisites for industrialization of Plant Cell Culture. It is used in the micropropagation of several crops, including ornamental and medicinal plants, vegetables and fruits. By cultivation in bioreactors, different plant parts can be obtained, such as buds, somatic embryos, bulbs, shoots, calluses, protocorm and others. They are also used for making pharmaceutical products such as flavonoids, phenols and other secondary metabolites.

Bioreactors are commonly cylindrical, ranging in size from litres to cubic metres and are often made of stainless steel. They are sterile vessels with temperature controllers and gassing facilities required for starting a biochemical reaction. Control over temperature, moisture, pH level, oxygen levels and stirring rate will yield the most suitable conditions necessary for maximized cell growth and productivity.

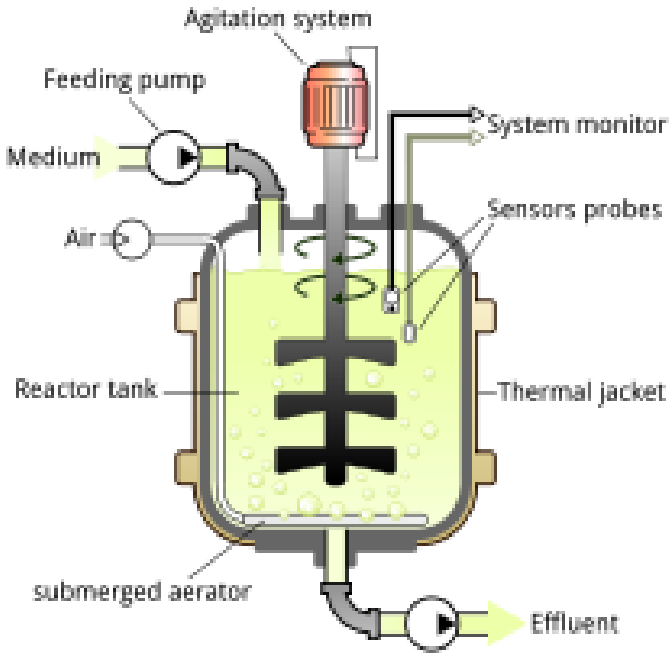

Figure 1. General structure of a stirring bioreactor

Bioreactor design specific for plant cell is important for the production of secondary metabolites using cell culture techniques. Based on the structure, plant cell bioreactor can mainly be divided as 5 types:

- Mechanical stirring,
- Air-lifting,
- Bubbling,
- Nutrient mist and
- Temporary immersion bioreactors.

These bioreactors have some characteristics in common that they can provide well mixed media and sterile air. Different from shaking flask, that the shaking will allow the media contact with air in the flask continuously, the media in bioreactor should be stirred or lifted by air bubbles, or mixed with air and then provide to cultured cells or tissues. Therefore, provision of air becomes very important for bioreactors. In bioreactors, there are several sensors monitoring the pH changes, temperature, dissolved oxygen (DO) and generated bubbles, the automated bioreactor can control these indexes following the order set by the operators.

Usually, the **mechanical stirring bioreactor** can produce highest DO index, however, it is not used widely in plant cell and tissue culture, because they are sensitive to shear force.

In case of **air-lift and bubbling bioreactors**, they have similar structures and used quite often in plant cell and tissue culture. In hairy root culture, the bioreactors are always equipped with layers of meshes made of stainless steel, to provide attaching area for hairy roots.

The **nutrient mist and temporary immersion bioreactors** have similar characteristics and they are both used for tissue culture, such like hairy root, adventitious root, shoots, bud clusters. They are all composed of two parts, media storage part and tissue culture part. After mixing of media and sterile air, for nutrient mist bioreactor, the media is sprayed through atomizer into tiny droplets on the surface of cultured tissues, and for temporary immersion bioreactor, the media is pumped to the culture part and maintain for short time and then the media is recovered to storage tank.

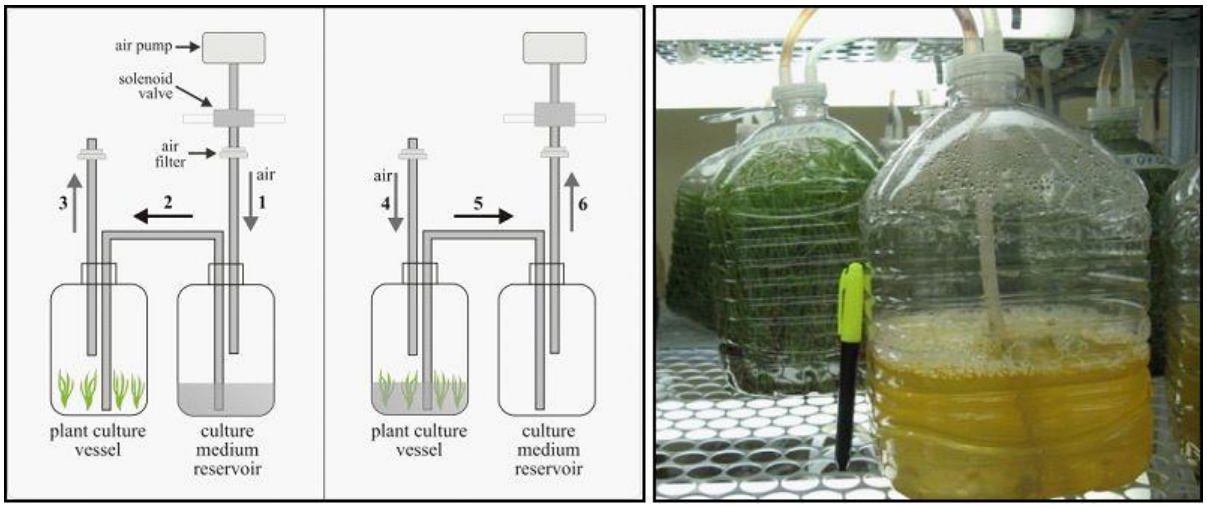

Figure 2. Temporary immersion bioreactor (TIB): The transfer of the medium to the plant culture vessel occurs by a positive pressure of the air pump and the explants are temporarily immersed, after this, the air pump starts and the medium returns to the culture medium reservoir (A) scheme, (B) picture.

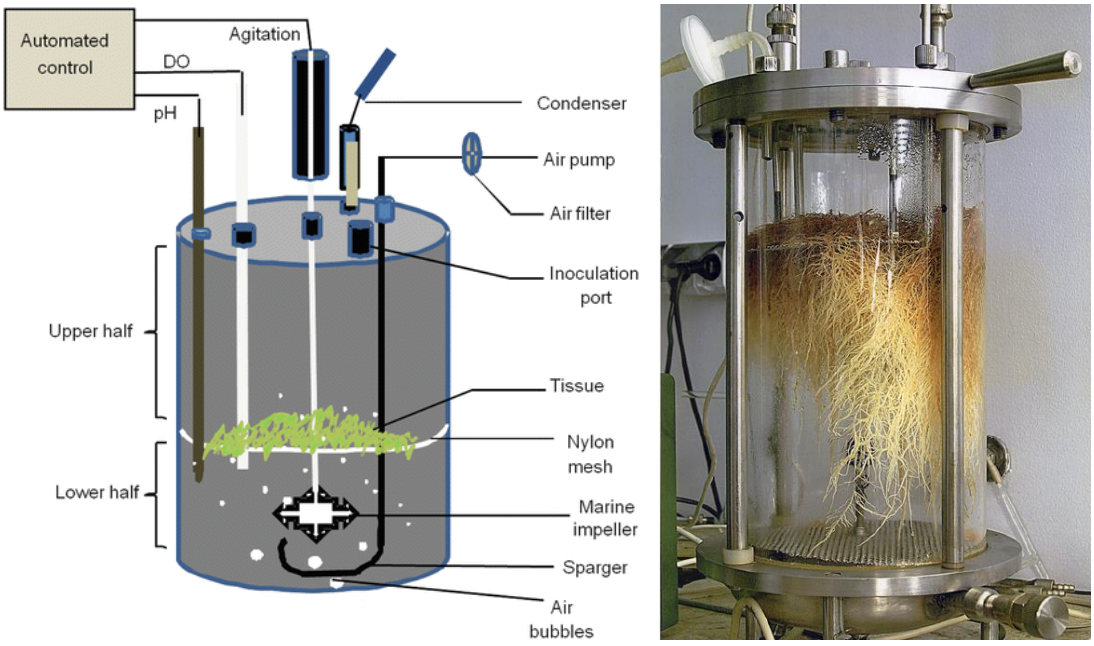

Figure 3: Nutrient Mist Bioreactor: A) Hairy Root culture scheme, B) *Platycodon grandiflorum* hairy roots culture picture by Joanna Giebułtowicz

There are successful examples for cell and tissue culture of medicinal herbs to produce useful secondary metabolites:

1. *Digitalis purpurea* cell line was cultured in airlift bioreactors, and the yield of methyl isopropyl hydroxyl digitoxin reached 430 mg/L;
2. *Phalaenopsis aphrodita* protocorm like body culture in temporary immersion bioreactor for micro-propagation.
3. *Panax ginseng* adventitious root culture in bollon type bubbling bioreactor at scale of 20 tons. All these achievement demonstrated the industrial potential of the application of bioreactor in producing secondary metabolites with high medicinal value.
4. In the propagation of pineapple (*Ananas comosus*), the multiplication rate in bioreactors was four times higher than that obtained in conventional systems. In banana (*Musa acuminata*) this multiplication rate was five times higher and in sugarcane (*Saccharum edule* Hassk.), it was six.

Ref:

1. **Hu Gaosheng and Jia Jingming,** Production of Useful Secondary Metabolites Through Regulation of Biosynthetic Pathway in Cell and Tissue Suspension Culture of Medicinal Plants, 2012.
2. **G. P. P. Lima, R. A. da S. Campos, L.G. Willadino, T. J.R. Câmara and F. Vianello,** Polyamines, Gelling Agents in Tissue Culture, Micropropagation of Medicinal Plants and Bioreactors, 2012
3. **Natalia Urbańska , Joanna Giebułtowicz, Olga Olszowska , Wojciech J. Szypuła,** The growth and saponin production of Platycodon grandiflorum (Jacq.) A. DC. (Chinese bellflower) hairy roots cultures maintained in shake flasks and mist bioreactor, 2014.