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Multiplication of shoots by Seolncr

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At the base of each leaf is a bud that will develop after being contacted with the culture medium. Periodically these new shoots should be subcultured in a new medium through replanting divisions in culture tubes or other suitable containers. These operations are performed in the laminar flow hood or in an isolated place that allows us to maintain aseptic conditions. This increases the number of plants in each ring or division of plants. The number of plants that is obtained depends on the plant species and conditions of the culture medium. The number of plants obtained by means of micropropagation can achieve exponential increases, considering all the factors that affect growth have been optimized.

Choosing a rooting medium of the explants

For root explants individual seedlings are mainly used with an approximate size of 2 cm. The shoots obtained during the multiplication phase was transferred to a medium free of growth regulators or hormones that contains only the type of auxin . Some species of plants do not need to go through this stage and make their roots in the same medium where new buds develop, so the process of multiplication and rooting pass simultaneously.

Acclimatization of rooted explants

The newly rooted explants are very sensitive to environmental changes, so the success or failure of the whole process depends on the acclimation. At this stage the plants suffer different kinds of changes that will allow them to adapt to living in natural conditions. At the time that the explants are removed or rooted cuttings of the bottles, are poorly adapted to grow in a greenhouse, as these explants were rooted and grown in environments with very high humidity and generally have stomata (structures responsible for regulating perspiration and water loss in the plant) that are not fully functional versus the relative humidity decreases, and therefore too slow to avoid desiccation of the explant. On the other hand, grow in humid environments as well typically involves the lack of a well developed wax cuticle, which represents the physical barrier to prevent water leakage along the entire surface of the plant. Rooted seedlings should be acclimatized to the humidity of the greenhouse gradually decreasing relative humidity and progressively increasing the intensity of light. These seedlings will be planted in containers (seedbeds) covered with plastic to maintain high relative humidity.The choice of a substrate with good physical characteristics, is key to the success of this stage. For transplantation, we chose a loose substrate, porous mixture of sand with peat, rice hulls burned to allow root development and growth very fast. The blends are different and varied according to the species with which we work. After carefully removing the agar to avoid damaging the roots, the seedlings were rinsed and placed in seedbeds with the mixture of selected substrates and covered with nylon. Every day we must control the humidity level in the seedbeds. If necessary, is applied with a hand spray irrigation, to maintain a moist environment at the substrate. At 15 days after the transplant, you can start lifting the nylon cover in the cooler hours (early morning or late afternoon). At first the plants are left uncovered half hour per day. The next week is left uncovered for one hour. One month after transplantation, are left covered overnight and if there is growth of new leaves, the plants may remain uncovered. The in vitro culture conditions, driving changes in anatomical and physiological aspects of plants, for this reason, during acclimatization, changes must be very gradual to minimize stress and have higher survival rate.

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