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ROLE OF TISSUE CULTURE IN FORESTRY

บทบาทของการเพาะเลี้ยงเนื้อเยื่อในทางป่าไม้

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บทคัดย่อ

ในขณะที่ความต้องการไม้เพิ่มขึ้น ปริมาณไม้ที่นำออกจากป่ากลับลดลงทุกขณะ ดังนั้นการนำเอาเทคนิคทางการเพาะเลี้ยงเนื้อเยื่อไปใช้ทางด้านป่าไม้จึงเป็นสิ่งที่ควรจะพิจารณา เทคนิคการเพาะเลี้ยงเนื้อเยื่อมีทั้งส่วนที่เป็นข้อดีและข้อเสีย แต่ในระยะยาวผลดีจะมีมากกว่า ถ้าหากว่านักวิชาการสามารถที่จะหาวิธีการที่เหมาะสมมาใช้สำหรับการเลี้ยงเนื้อเยื่อของต้นไม้แต่ละชนิดได้ โครงการเพาะเลี้ยงเนื้อเยื่อทางด้านป่าไม้ในหลายประเทศได้พัฒนาไปไกลแล้ว แต่ในประเทศไทยกำลังเริ่มต้น อย่างไรก็ดี ผลสำเร็จที่จะมีตามมาก็จะเป็นส่วนหนึ่งที่จะช่วยแก้ปัญหาเกี่ยวกับการขาดแคลนไม้ในอนาคตได้บ้าง

ABSTRACT

While the demand for world wood is increasing, the supply of wood from the forested area is decreasing. Re-afforestation is very necessary to be done in tropical countries, specially in Thailand. Using tissue culture technigues in forestry, then, should be under consideration. Eventhough tissue culture technigues have both advantages and disadvan - tages but in the long run the disadvantages could be overcome if appropriate techniques for each tree species could be found. Forest tree tissue culture technique in many countries have been developing for a long time but they have only recently started in Thailand. However, the accomplishments that should follow will of course help the wood shortage crisis to some extent in the future.

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INTRODUCTION

The important scientific consequences which could follow from the aseptic culture of plant tissues and plant cells was visualised as long ago as 1902 by Gottlieb Haberlant. However, it was, not until the early 1930's that seperate plant cells were induced to grow in liquid media and on agar plates. Experimental work has been proceeding continuously in this field since 1920 but it is only during the last decade that these techniques have begun to play a decisive role in the develop ment of plant science affecting the development of crop yield and possibly the productivity of the forest as well.

Significant gains have occured in growth and disease resistance of many forest trees, but demand for wood products will increase greatly during the last quarter of this century (Winton et. al., 1974). The U.S. Forest Service (1973) has predicted a wood shortage by the year 2000 unless new advances in forest technology are made soon. Greater yield from forest land is thus required and tree-breeding programs for improving the yield of the next generations should be well planned. Accordingly some forest research laboratories are investigating the possibilities of applying the relatively new method of callus and tissue cultures to accelerate current tree-breeding programs.

In the past decade many attempts have been made to produce plantlets from conifer tissues or organs cultured in vitro. Partial success has been reported by Greenwood and Berlyn (1965) by the induction of roots on Pinus lambertiana hypocotyls, by Bethel (1972) with the growth of roots on Pseudotsuga menziesii callus. Now it is possible to grow the cells of many tree species both angiosperms and gymnosperms on completely defined, artificial media from genetically diverse trees (Brown and Sommer, 1975), and numerous plantlets are obtainable from all of the major species of southern pines by using young embryos from mature seeds. (Sommer et. al., 1974, 1975). But neither conifer nor hardwood plantlets from single cells in liquid cultures have been produced, however, consi-

derable progress is being made. Within the near future we may be able to clone or reproduce vegetatively many of our forest trees in mass in small culture flasks. If this were possible, form the economics point of view, it would be save both money and time.

APPLICATION OF TISSUE CULTURE IN FORESTRY

The main objective of using tissue culture in forestry is for increasing yield or productivity of forests in a short time span. How we can reach this objective and how we can manipulate the techniques of tissue culture are things which should be considered.

The application of tissue culture to breeding programs is by using the mass propagation of the desired genotype. This vegetative propagation would allow the transmission of all genetic characteristics to the next generation. Recently, haploid plantlets were obtained in some genera by the anther and pollen culture techniques. Immature pollen of cultured anther has been induced to form embryoids or callus masses and can differentiate to plantlets (Carlson, and Polacco, 1975).

Haploid plantlets are very useful in breeding programs because they permit direct relation of recessive mutant phenotypes, or they provide the screening of the recessive mutant phenotypes in which recessive genes, often masked in diploid individuals, are exposed in haploid plants. Also the chromosome number of haploid plantlets can be doubled to form a true breeding diploid line which would be homozygous. However, most haploid plants formed through anther and pollen culture techniques would be of low vigor or inviable due to the unmasking of the deleterious recessive genes. But small numbers of them would be free of deleterious genes and would be useful in the breeding program. Homozygous diploid lines could then be crossed to obtain heterosis or hybrid vigor.

However, there is also a problem associated with the breeding program: the reduction in genetic variation of a cultivated species through selective breeding would lead to increasing the chances of major epidemics and catastrophic losses. Tissue culture not only can reduce

the genetic variability of crop species, but can increase it as well. Variation can be increased by somatic or controlled parasexual hybridization. The method is by enzymatic digestion removal of cell walls of different plants which are grown together in the same medium. The protoplasts, under the proper conditions, will fuse, followed by nuclear fusion and cell wall reconstitution. This method has been done with tobacco cells (Carlson et. al., 1972) and is being attemped with forest trees (Winton et. al., 1974).

Somatic hybridization can be done with both haploid and diploid cells but it would be much more appropriate to use diploid if the future generations were to produce sexually.

Hybrid cells and plantlets that were chosen and tested by exposing them to various kind of stresses and which survived those stresses are potentially interesting individuals. Selective procedures might be designed to recover cells or plantlets tolerant to toxic amounts of specific ions, pollutants, herbicides or to extreme temperature or water stress. This method can also be used for studying the resistance to certain disease or insensitivity to pathogens as done by Harvey and Grasham (1970).

Tissue culture is also useful for conservation of gene resources. The preservation of callus can be done by sequential culturing which will last for a long time, two year for quaking aspen (Winton, 1970), or even longer if the appropriate method can be found. Low temperature storage of cultured carrot cells has also been successful (Nag and Street, 1973). In the same way important individual genotypes for the breeding programs could be preserved.

ADVANTAGE OF TISSUE CULTURE IN FORESTRY

There are some advantages to using the tissue culture techniques in forest. First, the number of plants will be increased in a shorter period of time when compared to those produced by normal propagation procedures. With certain successful preparation of tissue cultures, a

small plantlets can be obtained in about 2-3 months, while the normal propagation procedures or sexual reproduction would take more time than that. Also use of this technique will overcome the problem of difficulties in rooting of some species from vegetative propagation or grafting. Apart from this advantage, tissue culture techniques require a lot less space than the normal propagation procedures.

Second, pertaining to the condition of stock: The stocks from tissue cultures tend to be aseptic because they have been taken care of under certain conditions while field stocks for normal propagation may be subject to various conditions such as drought, cold, diseases etc. which could lead to unsuccessful grafting.

Third, field propagation methods are more susceptible to the infestation of viruses than the tissue culture method. Generally mature tissues are being used in field propagations and they tend to be infested more easily than the younger tissues which are being used in tissue culture.

Fourth, potential for genetic manipulation by tissue culture technique is better than that by traditional propagation methods. By subjecting the cells or plantlets to various stress agents; diseases, salt, temperature, drought and observing their reactions. The resistant cells will be chosen and may be used to fuse with other cells to produce a resistant strain.

DISADVANTAGES OF TISSUE CULTURE

On the debit side of tissue culture techniques, there are also some considerations of which we ought to be aware.

First, each plant species has certain requirements of its own in order to grow. A perfect procedure for each species is required, especially in the condition of the medium, which must be determined seperately for each species.

Second, there is some tendency toward increasing the ploidy and

the chromosome abberations which are mainly due to stimulation from growth factors which will cause unbalanced differentiation in the cells and lead to abnormality. (Mott, 1977)

However, when considered in the long run, these disadvantageous aspects should be overcome as a result of research projects.

CONCLUSION

There are both advantages and disadvantages to the use of tissue cultures which certainly will affect forestry operations. However, as time goes by, researchers tend to work for the advantages and against the disadvantages in order to get the better result.

The potential of tissue culture in increasing forest tree yield is very interesting. Tissue culture is expected to be a great tool for this purpose, however the techniques are just being developed by researchers, especially in Thailand.

In the near future, wood production is not expected to meet the demand of the consumer, therefore increasing the forest tree yield is very desirable. The traditional forest tree breeding program of sexual reproduction is providing significant gains and is already well established (Zobel, 1974). As breeding programs advance into later generations, yield should be increased considerable; however the time span of the operation is an important barrier to providing enough wood materials for the consumer.

For tissue culture techniques, the progress can not now compare to that of traditional breeding method, but when we look at the present roles of tissue culture technique closely, some possibilities are more likely to be speculated. It might be a long time before tissue culture may be used in an operational practice in forestry. When techniques in tissue culture make more progress it can initially be used as a research tool to study genotype and environment interaction. Since the plantlets of the same genotype can be produced in a large number at the same time; ample plantlets will be available for study in different manipulated con-

ditions. But at the present time, the tissue culture of trees are very expensive and would not be economical unless the suspension cultures of tree tissue were possible.

It is now possible to grow plantlets from isolated cells and small clusters of cells detached from callus tissue by means of mechanical agitation, but mostly with herbaceous plants which are induced to form embryos in suspension cultures (Halperin, 1967; Thomas and Street 1970). Winton (1970) was the first to report the success of complete tree formation from root sprout callus tissue of triploid quaking aspen (Populus tremuloides). Plantlets from most major southern pine species are now obtainable by growing young embryo from mature seeds (Sommer and Brown, 1974), but their frequency, control and repeatability have not been fully determined. If there is a possibility that any pine species could be induced in suspension as are carrot embryos (Halperin, 1967), about 73 million plantlets might be produced from just 100 liters of medium (Campbell and Durzan, 1976).

In forestry, the true test of tissue culture is how its application affects plant characters under actual conditions. In this area, tissue culture techniques have not yet had the chance to prove themselves. They have not yet produced any genetic variants or combinations of genetic characters of economic importance, however there is a good possibility and it is only a matter of time and effort before vegetative propagation of forest trees by tissue culture techniques will be achieved. Ofcourse, if this were done it could possibly minimize or make up for the world wood shortage now being predicted.

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