Chapter 1 : Application of Somatic Embryogenesis in Woody Plants.

Somatic Embryogenesis in Woody Plants (Forestry Sciences) (Volume 6) by Jain, S M (ed) et al and a great selection of similar Used, New and Collectible Books available now at calendrierdelascience.com

Indirect embryogenesis occurs when explants produced undifferentiated, or partially differentiated, cells often referred to as callus which then is maintained or differentiated into plant tissues such as leaf, stem, or roots. Initiation and proliferation occur on a medium rich in auxin, which induces differentiation of localized meristematic cells. The auxin typically used is 2,4-D. Once transferred to a medium with low or no auxin, these cells can then develop into mature embryos. Germination of the somatic embryo can only occur when it is mature enough to have functional root and shoot apices [2] Factors influencing somatic embryogenesis[edit] Factors and mechanisms controlling cell differentiation in somatic embryos are relatively ambiguous. Certain compounds excreted by plant tissue cultures and found in culture media have been shown necessary to coordinate cell division and morphological changes. Several signaling molecules known to influence or control the formation of somatic embryos have been found and include extracellular proteins, arabinogalactan proteins and lipochitooligosaccharides. Temperature and lighting can also affect the maturation of the somatic embryo. Uses of somatic embryogenesis[edit] Mass propagation [11] Forestry related example[edit] The development of somatic embryogenesis procedures has given rise to research on seed storage proteins SSPs of woody plants for tree species of commercial importance, i. In this area of study, SSPs are used as markers to determine the embryogenic potential and competency of the embryogenic system to produce a somatic embryo biochemically similar to its zygotic counterpart Flinn et al. Seedling shoot height, root collar diameter, and dry weight increased at a greater rate in seedlings than in emblings during the first half of the first growing season, but thereafter shoot growth was similar among all plants. Root dry weight increased more rapidly in seedlings than in emblings during the early growing season During fall acclimation, the pattern of increasing dormancy release index and increasing tolerance to freezing was similar in both seedlings and emblings. Root growth capacity decreased then increased during fall acclimation, with the increase being greater in seedlings. Assessment of stock quality just prior to planting showed that: Grossnickle and Major [15] found that year-old and current-year needles of both seedlings and emblings had a similar decline in needle conductance with increasing vapour pressure deficit. Year-old and current-year needles of seedlings and emblings showed similar patterns of water use efficiency. Rates of shoot growth in seedlings and emblings through the growing season were also similar to one another. Seedlings had larger shoot systems both at the time of planting and at the end of the growing season. Seedlings also had greater root development than emblings through the growing season, but root: Tracking and fate maps[edit] Understanding the formation of a somatic embryo through establishment of morphological and molecular markers is important for construction of a fate map. The fate map is the foundation in which to build further research and experimentation. Two methods exist to construct a fate map: The latter typically works more consistently because of cell-cycle-altering chemicals and centrifuging involved in synchronous cell-division. The zygote is divided asymmetrically forming a small apical cell and large basal cell. The organizational pattern is formed in the globular stage and the embryo then transitions to the cotyledonary stage. Dicots pass through the globular, heart-shaped, and torpedo stages while monocots pass through globular, scuetellar, and coleoptilar stages. Abscisic acid has been reported to induce somatic embryogenesis in seedlings. After callus formation, culturing on a low auxin or hormone free media will promote somatic embryo growth and root formation. In monocots , embryogenic capability is usually restricted to tissues with embryogenic or meristematic origin. Somatic cells of monocots differentiate quickly and then lose mitotic and morphogenic capability. Differences of auxin sensitivity in embryogenic callus growth between different genotypes of the same species show how variable auxin responses can be. Five types of cells were identified from embryonic suspension: Each type of cell multiplied with certain geometric symmetry. They developed into symmetrical, asymmetrical, and aberrantly-shaped cell clusters that eventually formed embryos at different frequencies. Proembryogeny includes all stages prior to suspensor elongation. Early embryogeny includes all stages after suspensor elongation but before root meristem development. Late embryogeny includes development of root and shoot meristems. Proembryogenic masses PEMs , an intermediate between unorganized cells and an embryo composed of cytoplasmic-rich cells next to a vacuolated cell, are stimulated with auxin and cytokinin. Gradual removal of auxin and cytokinin and introduction of abscisic acid ABA will allow an embryo to form. However, the use of this technology for reforestation and tree breeding of conifers is in its infancy.

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Chapter 2 : Somatic Embryogenesis in Woody Plants : P. K. Gupta :

So far, plant tissue culture technology has largely been exploited in the propagation of ornamental plants, especially foliage house plants, by comÂ- mercial companies. Generally, tissue culture of woody plants has been recalÂ- citrant. However, limited success has been achieved in tissue culture of angiosperm and gymnosperm woody plants.

In the past, most of the information on this subject was scattered in proceedings volumes, journals, biotechnology books, etc. It has been difficult for the researchers and students to obtain comprehensive information on this rapidly growing subject from a single source. These books enable readers to get a clear view of this subject on historical, anatomical, physiological, biochemical and molecular aspects, and applications including protoplasts, cryopreservation, manufactured seed artificial seed , genetic transformation, bioreactors, mutations, and future uses in forest plantations. Each selected woody plant mentioned in Somatic Embryogenesis in Woody Plants is briefly introduced first, covering botany and genetics, importance and geographical distribution, breeding problems, and in vitro propagation and problems of each selected woody plant and then is followed by the description on the initiation and maintenance of embryogenic cultures, embryo development and germination, and field trials if any of these plants. Books like Somatic Embryogenesis in Woody Plants are meant for graduate students and researchers in forestry and horticulture as well as biotechnologists. Somatic embryogenesis in Cycadales; R. Somatic embryogenesis in Norway spruce Picea abies ; S. Somatic embryogenesis in Picea glehnii and P. Somatic embryogenesis in Black spruce Picea mariana ; K. Somatic embryogenesis in Serbian spruce Picea omorika ; R. Somatic embryogenesis in Blue spruce Picea pungens Engelmann ; J. Somatic embryogenesis in Red spruce Picea rubens Sarg. Somatic embryogenesis in Sitka spruce [Picea sitchensis Bong. Somatic embryogenesis in Pinus caribaea; A. Somatic embryogenesis in Slash pine Pinus elliottii Engelm. Somatic embryogenesis in Sugarpine Pinus lambertiana ; P. Somatic embryogenesis in Pinus nigra arn. Somatic embryogenesis in Maritime pine Pinus pinaster ; J. Somatic embryogenesis in Pinus radiata don; S. Somatic embryogenesis in Eastern White pine Pinus strobus ; K. Somatic embryogenesis in Scots pine Pinus sylvestris ; A. Somatic embryogenesis in Loblolly pine Pinus taeda ; M. Somatic embryogenesis in Douglas-fir Pseudotsuga menziesii ; P. Somatic embryogenesis in Larix; J. Somatic embryogenesis in Abies spp. Present state of somatic embryogenesis in Sequioa semervirens; J.

Chapter 3 : Protocol for somatic embryogenesis in woody plants, (Forestry sciences, Vol. 77) Jain Shri Mol

Somatic Embryogenesis in Woody Plants: Volume 2 â€• Angiosperms (Forestry Sciences) th Edition by S. Mohan Jain (Editor), Pramod P.K. Gupta (Editor), R.J. Newton (Editor) & 0 more.

This new embryo can further develop into a whole plant. In woody plants, somatic embryogenesis plays a critical role in clonal propagation and is a powerful tool for synthetic seed production, germplasm conservation, and cryopreservation. A key step in somatic embryogenesis is the transition of cell fate from a somatic cell to embryo cell. Although somatic embryogenesis has already been widely used in a number of woody species, propagating adult woody plants remains difficult. In this review, we focus on molecular mechanisms of somatic embryogenesis and its practical applications in economic woody plants. Furthermore, we propose a strategy to improve the process of somatic embryogenesis using molecular means. Introduction of Somatic Embryogenesis in Plants In flowering plants, the process of double fertilization involves a haploid sperm fertilizing a haploid egg cell to form a diploid zygote. Subsequently, the zygote undergoes a series of morphological, biochemical, and molecular events to develop into an embryo. This stage of development is referred to as embryogenesis Goldberg et al. Somatic embryogenesis is when a somatic cell dedifferentiates to a totipotent embryonic stem cell that can give rise to an embryo in vitro Verdeil et al. Since the original description of somatic embryogenesis in carrot Daucus carot cell cultures Steward et al. The developmental stages of somatic embryogenesis are similar to the process of zygotic embryogenesis in terms of developmental and regulatory mechanisms Dodeman et al. Therefore, somatic embryogenesis can provide an accessible model for studying the earliest developmental events of the zygotic embryo in the lifecycle of higher plants Zimmerman, Somatic and zygotic embryos have similar developmental stages typically passing through globular, torpedo, and cotyledonary stages in dicots, or globular, scutellar, and coleoptilar stages for monocots Mordhorst et al. For conifers, morphogenetic stages include globular, early cotyledonary and late cotyledonary embryos stages Quiroz-Figueroa et al. There are two different ways of inducting somatic embryogenesis including direct somatic embryogenesis and indirect somatic embryogenesis Yang and Zhang, In direct somatic embryogenesis, somatic embryos can be directly induced from the explant under certain conditions without any intermediate callus stage. Conversely, indirect somatic embryo genesis occurs via an intermediate callus stage and has been observed in most species Cuenca et al. Distinguishing between direct and indirect somatic embryogenesis can be difficult as both processes have been observed to occur simultaneously in the same tissue culture conditions Turgut et al. In contrast to primary somatic embryogenesis induced from explant cells, secondary somatic embryogenesis is the phenomenon whereby new somatic embryos are induced through existing somatic embryos Raemakers et al. In woody plants, secondary somatic embryogenesis can maintain the embryogenic competence of cultures for many years and thus provide useful research material Martinelli et al. Indirect somatic embryogenesis is a multi-step regeneration process beginning with a proembryogenic mass PEM , followed by somatic embryo formation, maturation, and conversion von Arnold et al. A key point in indirect somatic embryogenesis is the production of PEM consisting of proliferating embryogenic cells at an intermediate state between callus and somatic embryo and a relatively disorganized structure Halperin, Given that the potential applications of somatic embryo genesis in woody plants span a broad range of topics, this review will focus on briefly introducing practical applications of somatic embryogenesis in economically significant woody plants. Molecular mechanisms to improve the development of somatic embryogenesis in woody plants will be also discussed. Application of Somatic Embryogenesis in Woody Plants The rapid increase in human population size, environmental pollution, and demand for timber products has put enormous pressure on trees. Development of new technologies for tree propagation, improvement, and breeding can help to solve these problems Timmis, This has been achieved in part using biotechnology methodologies like in vitro propagation, genetic transformation, and marker-assisted breeding to gradually genetically improve woody plants Merkle and

Dean, ; Lelu-Walter et al. In vitro propagation could not only be used for mass clonal propagation of desirable genotypes, but could also provide suitable target material for genetic transformation Rugh et al. This technology is important for woody plants that have a long life cycle and are difficult to propagate by conventional methods Isah, In tissue cultures, plant regeneration via somatic embryogenesis may offer many advantages over organogenesis, such as the feasibility of single cell origin and the possibility of automating the large-scale production of embryos in bioreactors and field planting as synthetic seeds Giri et al. The bipolar nature of embryos allows for direct development into plantlets without the need for the rooting stage required for plant regeneration organogenesis von Arnold et al. Furthermore, single epidermal cell origins for embryos might avoid chimeras, favoring the use of this process for plant transformation Normah et al. In woody plants, pioneering research on somatic embryo genesis was only observed to form the embryo-like structures in sandalwood Santalum album L. However, these embryo-like tissues did not grow into complete plants. Early reports of regeneration via somatic embryogenesis of sandalwood were achieved using hypocotyl and nodal segments Bapat and Rao, ; Sita et al. In conifers, somatic embryogenesis and plantlet regeneration were first reported in Norway spruce Picea acies , for which immature and mature zygotic embryos were used as explants to establish a culture system Chalupa, ; Hakman et al. This was followed by extensive studies into exploiting the potential of somatic embryogenesis of important tree species with examples are described below. Somatic embryo development was then progressed to the precotyledonary stage on media with 2. Embryogenic calli of Japanese larch Larix kaempferi were obtained from mature embryos cultured in dark condition on Quoirin and Lepoivre media with 1. Further improvements of somatic embryo development show that the highest plantlets yield was obtained calli were cultured on half strength Quoirin and Lepoivre media containing 90 mM sucrose and 7. Eucalypts are widely planted hardwood forest trees because of their fast growth and remarkable adaptability Patt et al. Eucalypts can also provide excellent resources in terms of the production of pulp and eucalyptus oil. In Eucalyptus species, most reports were on embryogenic cells derived from immature and mature zygotic embryos or juvenile seedlings Prakash and Gurumurthi, Somatic embryogenesis achieved induction using leaf from mature trees is rarely reported. A recent study has described somatic embryogenesis induction from leaf and shoot apex explants of mature Eucalyptus globulus and hybrid E. The East Indian sandalwood tree S. Somatic embryogenesis provides a system for large-scale plant propagation in bioreactors da Silva et al. Misra and Dey reported an efficient protocol for the mass production of sandalwood biomass by bioreactor based cultivation of somatic embryogenesis. Somatic embryos can be used to produce raw medicinal materials such as santalols, phenolics, and arabinogalactan proteins. A successful rapid protocol for somatic embryogenesis using cultures of nodal segments on media with 2. The strawberry tree Arbutus unedo L. The fruit of the strawberry tree are commonly consumed fresh or processed into jam El-Mahrouk et al. Conventional methods of propagation cannot preserve elite strawberry tree genotypes. Somatic embryogenesis is able to overcome this problem by using of leaves from adult trees to induce somatic embryos Martins et al. Alders species are of minor importance in economic terms, but do have ecological value through land reclamation and reforestation. Alders can fix atmospheric nitrogen through a symbiotic association with the actinomycete Oliveira et al. Induction media were composed of 0. For medicinal woody plants, such as bastard teak Butea monosperma Lam. Kuntze , somatic embryogenesis was able to effectively produce a large number of plantlets and bioactive compounds. Moreover, qualitative analysis using Liquid chromatography electro spray ionization quadrupole time of flight mass spectrometry LC ESI Q-TOF MS showed that the secondary metabolites in vitro developed cultures were the same as those in wild grown leaf samples Sharma et al. The developmental pattern of somatic embryos is very similar among most woody species tested to date. This begins with an immature or mature embryo that is cultured on a nutrient medium containing a high concentration of plant growth regulators like 2,4-D, 6-BA, and Picloram Isah, It is disadvantageous to use immature or mature zygotic embryos as explants because of their unproven genetic value. Although induction of somatic embryo using leaf from mature tree as explants have already been achieved in a small number of species, this difficulty still remains unsolved in initiation of embryogenic cultures from adult woody plants Cuenca et al. Maturation and quality of somatic embryos are further limiting factors in conversing embryos into plants. Synthetic Seeds The concept of synthetic seeds was first mentioned by Murashige The synthetic or artificial seed was defined as an encapsulated single somatic embryo inside a matrix covering. Later, synthetic seeds of alfalfa were successfully produced by encapsulating somatic embryos in alginate hydrogel Redenbaugh et al. In early studies, synthetic seeds referred only to the somatic embryos that were used in plant production and were transported to the field. Following this utilization of synthetic seeds, shoot apical tips, axillary buds, and nodal segments have also been employed as appropriative alternatives to somatic embryos Sarkar and Naik, ; Standardi and Piccioni, ; Ara et al. Until now, synthetic seeds have been used in some economically significant woody species including fruit and forest trees Prewein and Wilhelm, ; Malabadi and Staden, ; Singh et al. The encapsulation technology provides the somatic embryo with protection from mechanical damage and a supply of nutrients for the growing embryo. Synthetic seeds could, therefore, be easily handled for storage, transport, and sowing, the same as a zygotic seed Rai et al. Hydrated and desiccated forms of encapsulation technology were employed in synthetic seeds production Sharma et al. Most efforts involve using different coating agents to encapsulate the propagules. These agents include sodium alginate, potassium alginate, sodium pectate, and carrageenan among others, with sodium alginate being the most common Malabadi and Staden, ; Pintos et al. Carbon sources, plant growth regulators, and antimicrobial agents were also added to the hydrogel to facilitate growth and increase the survival rate of the encapsulated propagules Bapat and Mhatre, ; Rai et al. Finally, the encapsulated propagules were cultured on media or in the field for plantlet conversion. Desiccated encapsulation was only suitable for some specious whose somatic embryos are tolerant of desiccation. This indicates that the survival competence of somatic embryos is an important factor for storage and conversion under low moisture conditions. After inducing desiccation tolerance, somatic embryos are coated with a protective and nutritive layer to avoid mechanical damage and provide nutrients during the early stages of conversion Sharma et al. This desiccated encapsulation method of generating synthetic seeds is not, however, widely used in woody plants because the somatic embryos of some woody plants cannot tolerate desiccation. Using somatic embryos of woody plants as explants presents a major problem in that the use of encapsulated embryos results in a lower conversion rate when compare with other explants, such as the shoot tip or nodal segment Tsvetkov et al. Moreover, different woody species also have different conversion rates. The genotype of the somatic embryo, the encapsulating agent used, and the matrix determined the success of synthetic seed technology in woody plants Gantait et al. Evidence also suggests that the conversion rate may decrease with increased storage times and storage temperatures Singh et al. Cryopreservation is an effective technique for long-term conservation of woody plant somatic embryos Lambardi et al. The key requirement of cryopreservation is that the water content of the cells be kept low enough to prevent the formation of ice crystals, ensuring that somatic embryos can easily recover after storage in liquid nitrogen. In contrast with one-step direct immersion in liquid nitrogen, slow cooling is the common method for cryopreservation of somatic embryogenic cultures in conifer and broad-leaf trees Klimaszewska et al. Slow cooling is, therefore, both expensive and tedious, making it essential that a simple and reliable cryopreservation method be developed for widespread use in somatic embryo cultures of woody plants. In order to trigger cell vitrification, two strategies were employed during one-step freezing. Chemical dehydration using highly concentrated vitrification solutions, such as dimethyl sulfoxide or sucrose, is one method, and another is physical dehydration by exposing the somatic embryo to sterile air or silica gel Sisunandar et al. This work was carried out to develop a simple and reliable protocol for cryopreservation of woody plants, and has led to successful cryopreservation of embryogenic cells from a large number woody plant species including both angiosperms and gymnosperms Cyr, ; Touchell et al. To rapidly thaw cryopreserved samples, the cryovials are plunged into warm water. Cryoprotectants are removed from the thawed somatic embryogenic cultures through gradual elution, and the cultures are transferred onto fresh regrowth media. This technique could preserve embryogenic cells for an extended period. For instance, in A. In recent years, cryopreservation has been routinely employed for long-term conservation of plant genetic resources in woody plants Benelli et al.

Chapter 4 : Somatic Embryogenesis in Woody Plants: Volume 4 (Forestry Sciences) - | SlugBooks

FORESTRY SCIENCES Somatic Embryogenesis in Woody Plants 5 co Maturation of somatic embryos S.M. Jain, P.K. Gupta and R.J. Newton (eds.), Somatic Embryogenesis in Woody Plants.*

Received May 31; Accepted Aug This article has been cited by other articles in PMC. Abstract Oplopanax elatus is a medicinal plant on the verge of extinction because of overexploitation. In the present study, the effects of various factors on enhancing somatic embryogenesis and plantlet conversion were studied. Mature seeds were collected from a total of 13 plants from 4 mountains in South Korea, and the genetic distances were calculated to analyze the effect of genotype on somatic embryogenesis. Results of cluster analysis and the unweighted-pair-group method with arithmetic mean of 13 genotypes indicated the presence of 3 main groups. Both genotype and explant type affected the induction of somatic embryos SEs. Sorak 2 and root were found to be the most suitable genotype and explant type, respectively, for SE induction in O. The formation and development of SEs were significantly influenced by culture density; thus, 10 mg embryonic callus was found to be the most suitable for SE induction. The highest rates of germination and SE conversion were obtained in a germination medium containing 1. Thus, our results showed that the percentage survival of O. Micropropagation, Oplopanax elatus, Conservation, Somatic embryogenesis, Regeneration Background Oplopanax elatus is a valuable medicinal plant and one of the first Far-Eastern plants to be recommended for research as a source of herbal preparations, similar to ginseng. Because of its highly valued medicinal properties against conditions such as asthenia, depressive states, and hypertension, this rare and endangered species has been historically overexploited throughout Asia, and its demand is increasing Lee et al. The distribution of O. Conventional methods for the vegetative propagation of O. Moreover, the distribution of weak, less viable zygotic embryos of this species is restricted to high elevations Lee et al. Therefore, alternative methods for the rapid multiplication of this plant must be developed for its conservation, as well as for medicinal purposes. In vitro culture has always been a preferred, alternative approach for replicating genetically identical plantlets of rare and endangered species Kowalski and Von Staden ; Negash ; Gomes et al. Although callus induction and somatic embryogenesis have been widely explored for Araliaceae, studies on somatic embryogenesis in O. For this species, callus induction and somatic embryogenesis have been reported by only Cho et al. In , plantlets of this species could be successfully regenerated by somatic embryogenesis in our laboratory Moon et al. However, because of the low ratio of induction of somatic embryos SEs and plantlet conversion, this method could not be established for the mass propagation of O. Another limitation was that viable seeds of this species cannot be easily collected from natural habitats. Consequently, somatic embryogenesis is dependent on other somatic tissues, such as leaves, stems, and roots. In this paper, we have reported an improved production system for the SEs of O. Results Cluster analysis and genotype effect The results of the cluster analysis indicated that the genotypes of the studied O. The Bangtae genotype is related to the 2 Taebaek genotypes, whereas the 2 Sorak genotypes and 8 Jiri genotypes form separate clusters.

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Chapter 5 : Frontiers | Application of Somatic Embryogenesis in Woody Plants. | Plant Science

The rapid progress made on somatic embryogenesis and its prospects for potential applications in improving woody plants prompted us to edit this book initially in three volumes, and now to add two more volumes.

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Although somatic embryogenesis has already been widely used in a number of woody species, propagating adult woody plants remains difficult. In this review, we focus on molecular mechanisms of somatic embryogenesis and its practical applications in economic woody plants. Furthermore, we propose a strategy to improve the process of somatic embryogenesis using molecular means. Subsequently, the zygote undergoes a series of morphological, biochemical, and molecular events to develop into an embryo. This stage of development is referred to as embryogenesis Goldberg et al. Somatic embryogenesis is when a somatic cell dedifferentiates to a totipotent embryonic stem cell that can give rise to an embryo in vitro Verdeil et al. Since the original description of somatic embryogenesis in carrot Daucus carot cell cultures Steward et al. 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Application of Somatic Embryogenesis in Woody Plants The rapid increase in human population size, environmental pollution, and demand for timber products has put enormous pressure on trees. Development of new technologies for tree propagation, improvement, and breeding can help to solve these problems Timmis, This has been achieved in part using biotechnology methodologies like in vitro propagation, genetic transformation, and marker-assisted breeding to gradually genetically improve woody plants Merkle and Dean, ; Lelu-Walter et al. In vitro propagation could not only be used for mass clonal propagation of desirable genotypes, but could also provide suitable target material for genetic transformation Rugh et al. 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These agents include sodium alginate, potassium alginate, sodium pectate, and carrageenan among others, with sodium alginate being the most common Malabadi and Staden, ; Pintos et al. Carbon sources, plant growth regulators, and antimicrobial agents were also added to the hydrogel to facilitate growth and increase the survival rate of the encapsulated propagules Bapat and Mhatre, ; Rai et al. Finally, the encapsulated propagules were cultured on media or in the field for plantlet conversion. Desiccated encapsulation was only suitable for some specious whose somatic embryos are tolerant of desiccation. This indicates that the survival competence of somatic embryos is an important factor for storage and conversion under low moisture conditions. After inducing desiccation tolerance, somatic embryos are coated with a protective and nutritive layer to avoid mechanical damage and provide nutrients during the early stages of conversion Sharma et al. This desiccated encapsulation method of generating synthetic seeds is not, however, widely used in woody plants because the somatic embryos of some woody plants cannot tolerate desiccation. Using somatic embryos of woody plants as explants presents a major problem in that the use of encapsulated embryos results in a lower conversion rate when compare with other explants, such as the shoot tip or nodal segment Tsvetkov et al. Moreover, different woody species also have different conversion rates. The genotype of the somatic embryo, the encapsulating agent used, and the matrix determined the success of synthetic seed technology in woody plants Gantait et al. Evidence also suggests that the conversion rate may decrease with increased storage times and storage temperatures Singh et al. Cryopreservation is an effective technique for long-term conservation of woody plant somatic embryos Lambardi et al. The key requirement of cryopreservation is that the water content of the cells be kept low enough to prevent the formation of ice crystals, ensuring that somatic embryos can easily recover after storage in liquid nitrogen. In contrast with one-step direct immersion in liquid nitrogen, slow cooling is the common method for cryopreservation of somatic embryogenic cultures in conifer and broad-leaf trees Klimaszewska et al. Slow cooling is, therefore, both expensive and tedious, making it essential that a simple and reliable cryopreservation method be developed for widespread use in somatic embryo cultures of woody plants. In order to trigger cell vitrification, two strategies were employed during one-step freezing. Chemical dehydration using highly concentrated vitrification solutions, such as dimethyl sulfoxide or sucrose, is one method, and another is physical dehydration by exposing the somatic embryo to sterile air or silica gel Sisunandar et al. This work was carried out to develop a simple and reliable protocol for cryopreservation of woody plants, and has led to successful cryopreservation of embryogenic cells from a large number woody plant species including both angiosperms and gymnosperms Cyr, ; Touchell et al. To rapidly thaw cryopreserved samples, the cryovials are plunged into warm water. Cryoprotectants are removed from the thawed somatic embryogenic cultures through gradual elution, and the cultures are transferred onto fresh regrowth media.

Chapter 6 : Somatic Embryogenesis in Woody Plants : Pramod P. K. Gupta :

Earlier, we edited a well received series on "Somatic embryogenesis in woody trees", volumes 1 to 6. These volumes provided readers detailed extensive reviews on somatic embryogenesis of important angiosperm and gymnosperm tree species, an excellent source of information for newcomers or those already engaged in research.

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