Introduction

1. History of PTC
2. Explanation of PTC
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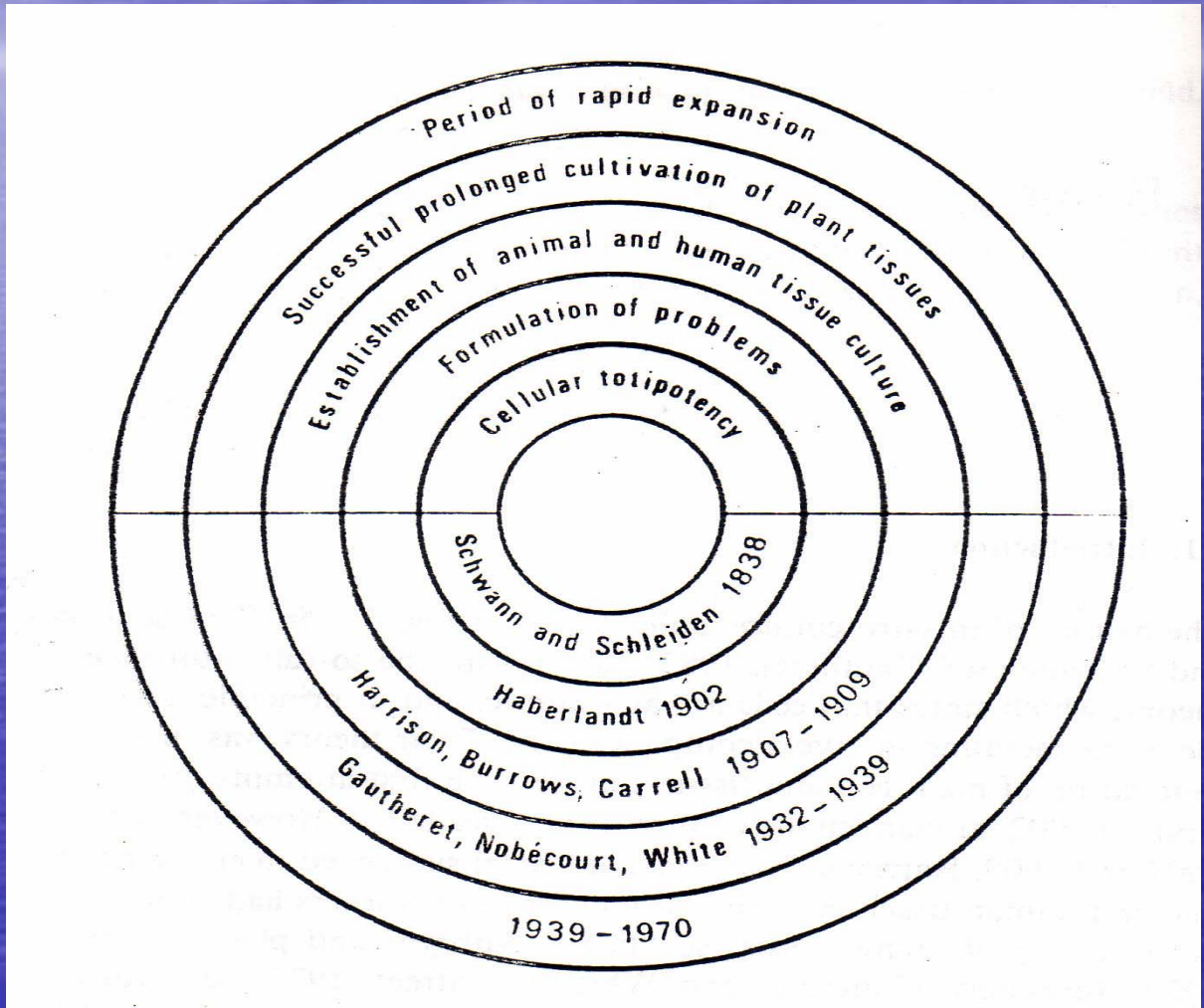
4. Benefit of PTC

1. History of PTC



1. History of PTC

1929 Embryo culture of *Linum* to avoid cross incompatibility (Laibach).
1934 In vitro culture of the cambium tissue of a few trees and shrubs failed to be sustained since auxin had not yet been discovered (Gautheret).
1934 Successful culture of tomato roots (White).
1936 Embryo culture of various gymnosperms (LaRue).
1939 Successful continuously growing callus culture (Gautheret, Nobécourt and White).
1940 In vitro culture of cambial tissues of *Ulmus* to study adventitious shoot formation (Gautheret).
1941 Coconut milk (containing a cell division factor) was for the first time used for the culture of *Datura* embryos (van Overbeek).
1941 In vitro culture of crown-gall tissues (Braun).
1944 First in vitro cultures of tobacco used to study adventitious shoot formation (Skoog).
1945 Cultivation of excised stem tips of *Asparagus* in vitro (Loo).
1946 First whole *Lupinus* and *Tropaeolum* plants from shoot tips (Ball).
1948 Formation of adventitious shoots and roots of tobacco determined by the ratio of auxin/adenin (Skoog and Tsui).
1950 Organs regenerated from callus tissue of *Sequoia sempervirens* (Ball).
1952 Virus-free dahlias obtained by meristem culture (Morel and Martin).
1952 First application of micro-grafting (Morel and Martin).
1953 Haploid callus of *Ginkgo biloba* produced from pollen (Tulecke).
1954 Monitoring of changes in karyology and in chromosome behaviour of endosperm cultures of maize (Strauss).
1954 First plant from a single cell (Muir et al.).
1955 Discovery of kinetin, a cell division hormone (Miller et al.).
1956 Realization of growth of cultures in multi-litre suspension systems to produce secondary products by Tulecke and Nickell (Staba, 1985).
1957 Discovery of the regulation of organ formation (roots and shoots) by changing the ratio of cytokinin/auxin (Skoog and Miller).
1958 Regeneration of somatic embryos in vitro from the nucellus of *Citrus ovules* (Maheshwari and Rangaswamy).
1958 Regeneration of pro-embryos from callus clumps and cell suspensions of *Daucus carota* (Reinert, Steward).
1959 Publication of the first extensive handbook on plant tissue culture (Gautheret).

1. History of PTC

1960 First successful test tube fertilization in *Papaver rhoeas* (Kanta).
1960 Enzymatic degradation of cell walls to obtain large numbers of protoplasts (Cocking).
1960 Vegetative propagation of orchids by meristem culture (Morel).
1960 Filtration of cell suspensions and isolation of single cells by plating (Bergmann).
1962 The development of the famous Murashige and Skoog medium (Murashige and Skoog).
1964 First haploid *Datura* plants produced from pollen grains (Guha and Maheshwari).
1964 Regeneration of roots and shoots on callus tissue of *Populus tremuloides* (Mathes).
1965 Induction of flowering in tobacco tissue in vitro (Aghion-Prat).
1965 Differentiation of tobacco plants from single isolated cells in micro-culture (Vasil and Hildebrandt).
1967 Flower induction in *Lunaria annua* by vernalization in vitro (Pierik).
1967 Haploid plants obtained from pollen grains of tobacco (Bourgin and Nitsch).
1969 Karyological analysis of plants regenerated from callus cultures of tobacco (Sacristan en Melchers).
1969 First successful isolation of protoplasts from a suspension culture of *Hapopappus gracilis* (Eriksson and Jonassen).
1970 Selection of biochemical mutants in vitro (Carlson).
1970 Embryo culture utilized in the production of monoploids in barley (Kasha and Kao).
1970 First achievement of protoplast fusion (Power et al.).
1971 First plants regenerated from protoplasts (Takebe et al.).
1972 Interspecific hybridization through protoplast fusion in two *Nicotiana* species (Carlson et al.).
1973 Cytokinin found capable of breaking dormancy in excised capitulum explants of *Gerbera* (Pierik et al.).
1974 Induction of axillary branching by cytokinin in excised *Gerbera* shoot tips (Murashige et al.).
1974 Regeneration of haploid *Petunia hybrida* plants from protoplasts (Binding).
1974 Fusion of haploid protoplasts found possible which gave rise to hybrids (Melchers and Labib).
1974 Biotransformation in plant tissue cultures (Reinhard).
1974 Discovery that the Ti-plasmid was the tumour inducing principle of *Agrobacterium* (Zaenen et al.; Larebeke et al.).
1975 Positive selection of maize callus cultures resistant to *Helminthosporium maydis* (Gengenbach en Green).

1. History of PTC

1976 Shoot initiation from cryo-preserved shoot apices of carnation (Seibert).

1976 Interspecific plant hybridization by protoplast fusion for *Petunia hybrida* and *Petunia parodii* (Power et al.).

1976 Octopine and nopaline synthesis and breakdown found to be genetically controlled by the Ti-plasmid of *Agrobacterium tumefaciens* (Bomhoff et al.).

1977 Successful integration of the Ti-plasmid DNA from *Agrobacterium tumefaciens* in plants (Chilton et al.).

1978 Somatic hybridization of tomato and potato (Melchers et al.).

1979 Co-cultivation procedure developed for transformation of plant protoplasts with *Agrobacterium* (Marton et al.).

1980 Use of immobilized whole cells for biotransformation of digitoxin into digoxin (Alfermann et al.).

1981 Introduction of the term somaclonal variation (Larkin and Scowcroft).

1981 Isolation of auxotrophs by large scale screening of cell colonies derived from haploid protoplasts of *Nicotiana plumbaginifolia* treated with mutagens (Siderov et al.).

1982 Protoplasts are able to incorporate naked DNA; transformation with isolated DNA is consequently possible (Krens et al.).

1982 Fusion of protoplasts by electrical stimulus (Zimmermann).

1983 Intergeneric cytoplasmic hybridization in radish and rape (Pelletier et al.).

1984 Transformation of plant cells with plasmid DNA (Paszkowski et al.).

1985 Infection and transformation of leaf discs with *Agrobacterium tumefaciens* and the regeneration of transformed plants (Horsch et al.).

2. Explanation of PTC

Isolation of part of plant and cultured on medium aseptically

3. Types of culture

1. Culture of intact plants
2. Embryo culture
3. Organ culture
4. Callus culture
5. Single cell culture
6. Protoplast culture

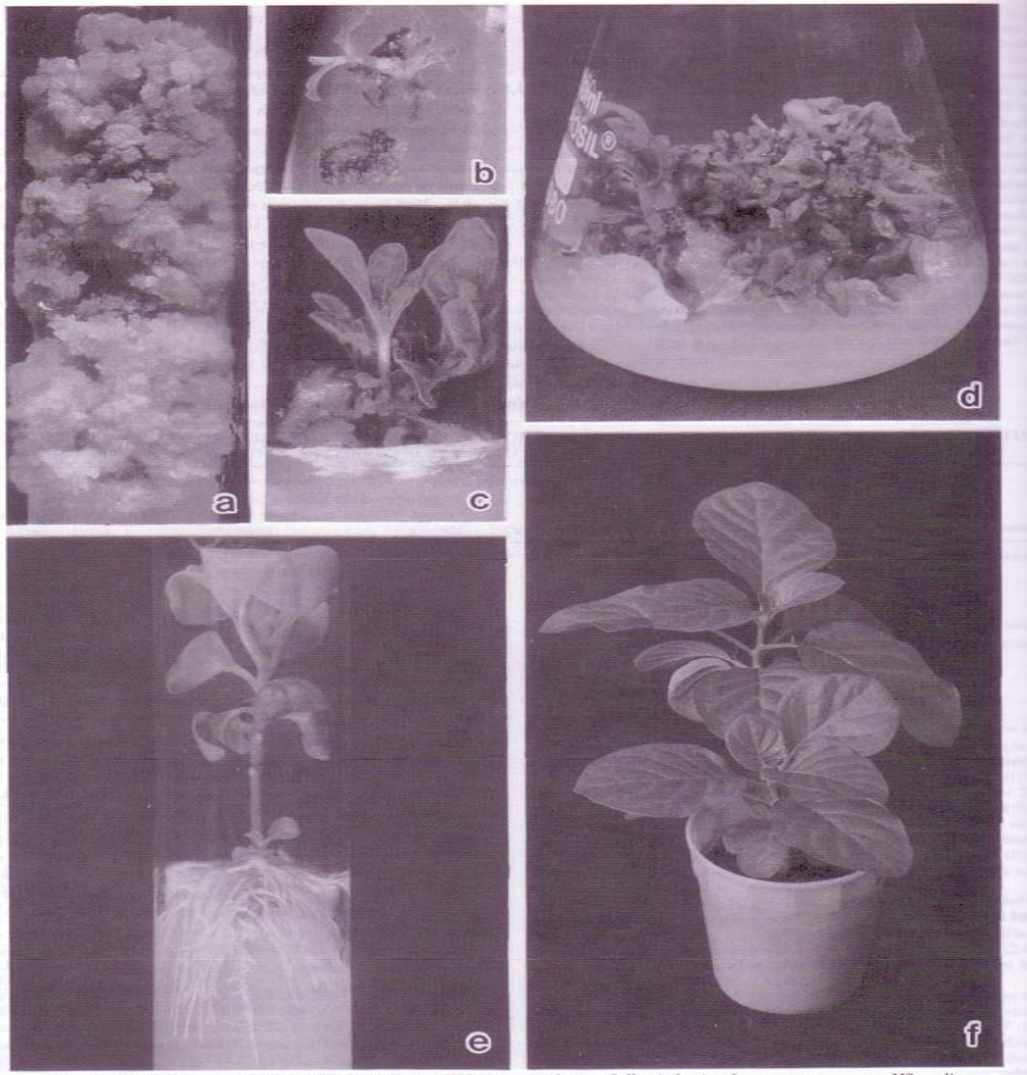


FIG. 1. Callus induction and plantlet regeneration in *Withania somnifera*. *a*, Callus induction from root segments on MS medium containing 2 mg l^{-1} (9.1 μM) 2,4-D and 0.2 mg l^{-1} (0.9 μM) KN. *b*, Shoot induction from hypocotyl callus with 2 mg l^{-1} 2,4-D and 0.2 mg l^{-1} KN. *c*, Shoot multiplication of hypocotyl callus-derived shoots with 2 mg l^{-1} (8.9 μM) BA. *d*, Multiple shoots obtained after 60 d of the second subculture with 2 mg l^{-1} BA. *e*, Root formation with 2 mg l^{-1} (9.9 μM) IBA. *f*, Plantlet transplanted to a plastic pot.

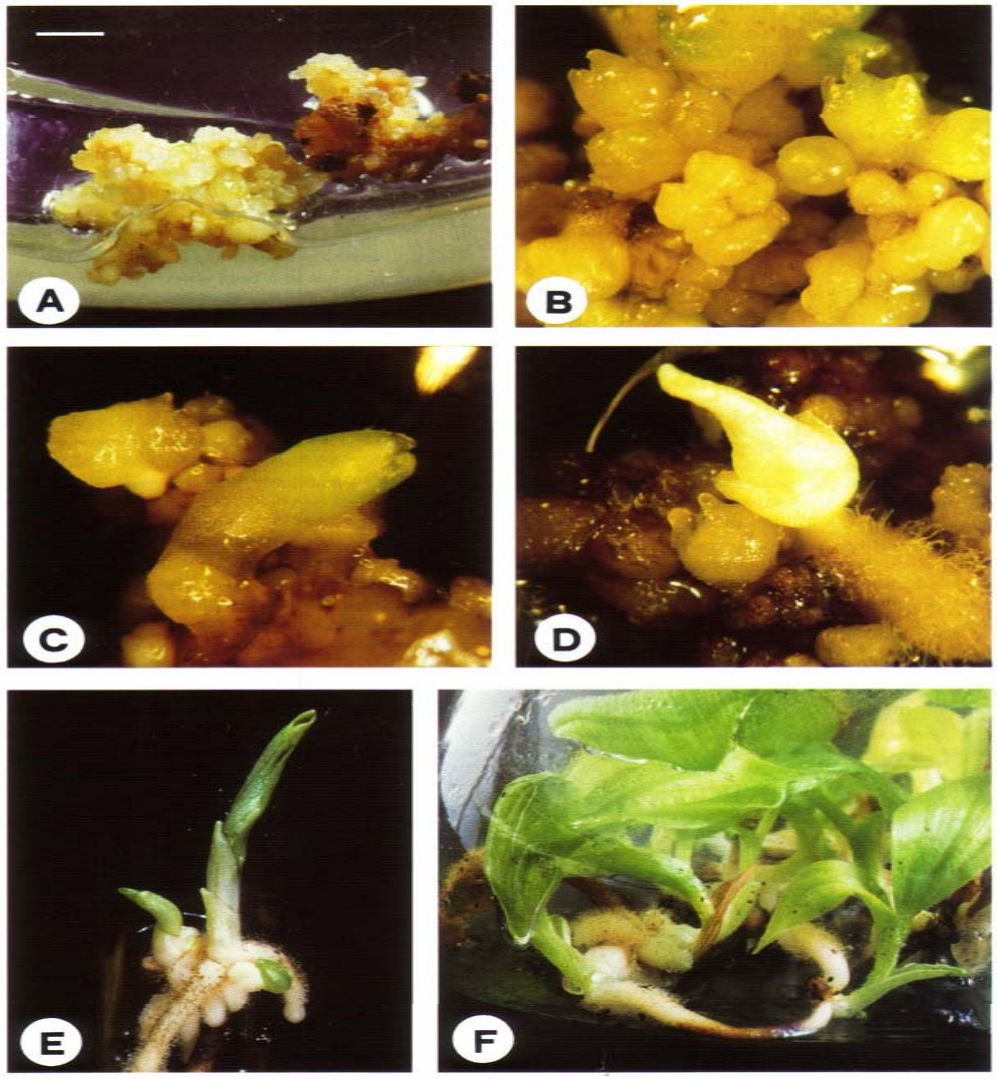
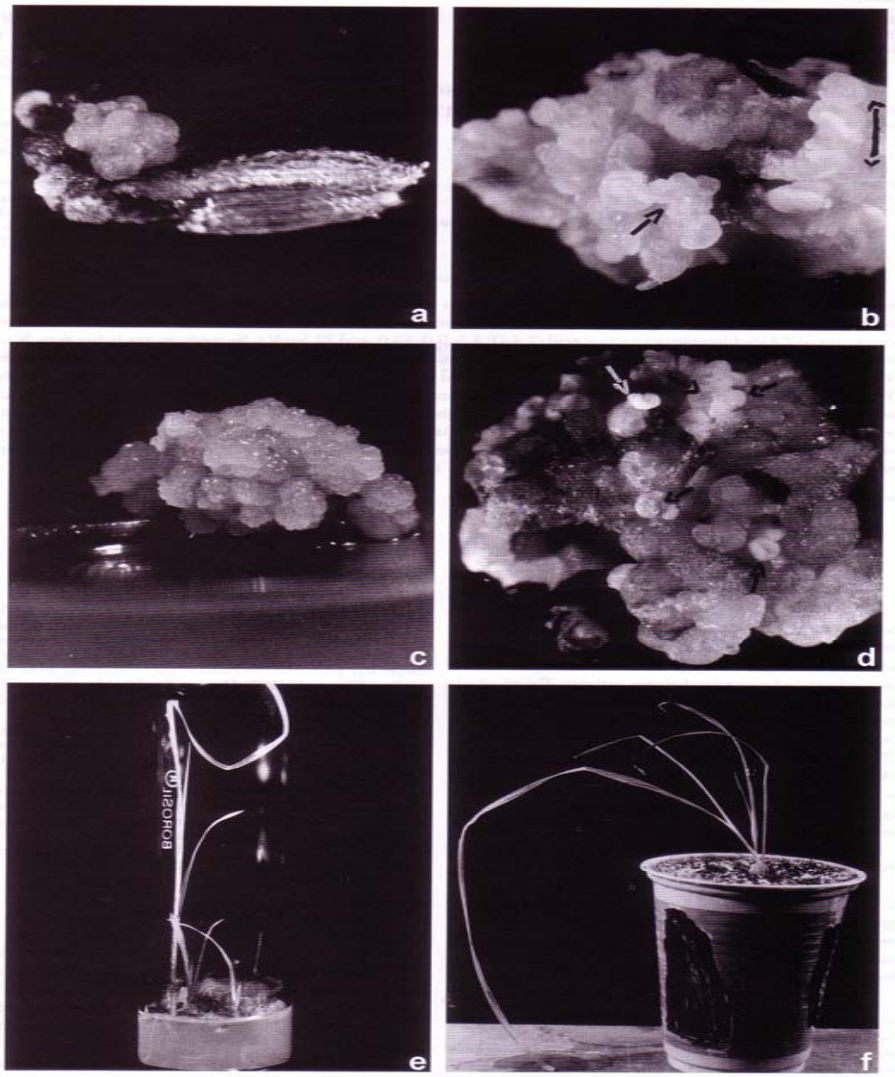


FIG. 1. *In vitro* plantlet regeneration from protocorm-derived callus of *Cypripedium formosanum*. *A*, Totipotent callus obtained from a seed-derived protocorm segment after 24 wk on basal medium with 4.52 μ*M* 2,4-D and 4.54 μ*M* TDZ following induction (*bar* = 12 mm). *B*, PLBs formed after 8 wk of culture from protocorm-derived callus on basal medium with 4.44 μ*M* BA (*bar* = 1.2 mm). *C*, An elongated PLB (*bar* = 1.2 mm). *D*, A PLB developed into a shoot bud and a root (*bar* = 1.5 mm). *E*, Lateral buds proliferating from a sprouting shoot (*bar* = 4 mm). *F*, Twelve-month-old PLB-derived plantlets ready for potting (*bar* = 12 mm).


a
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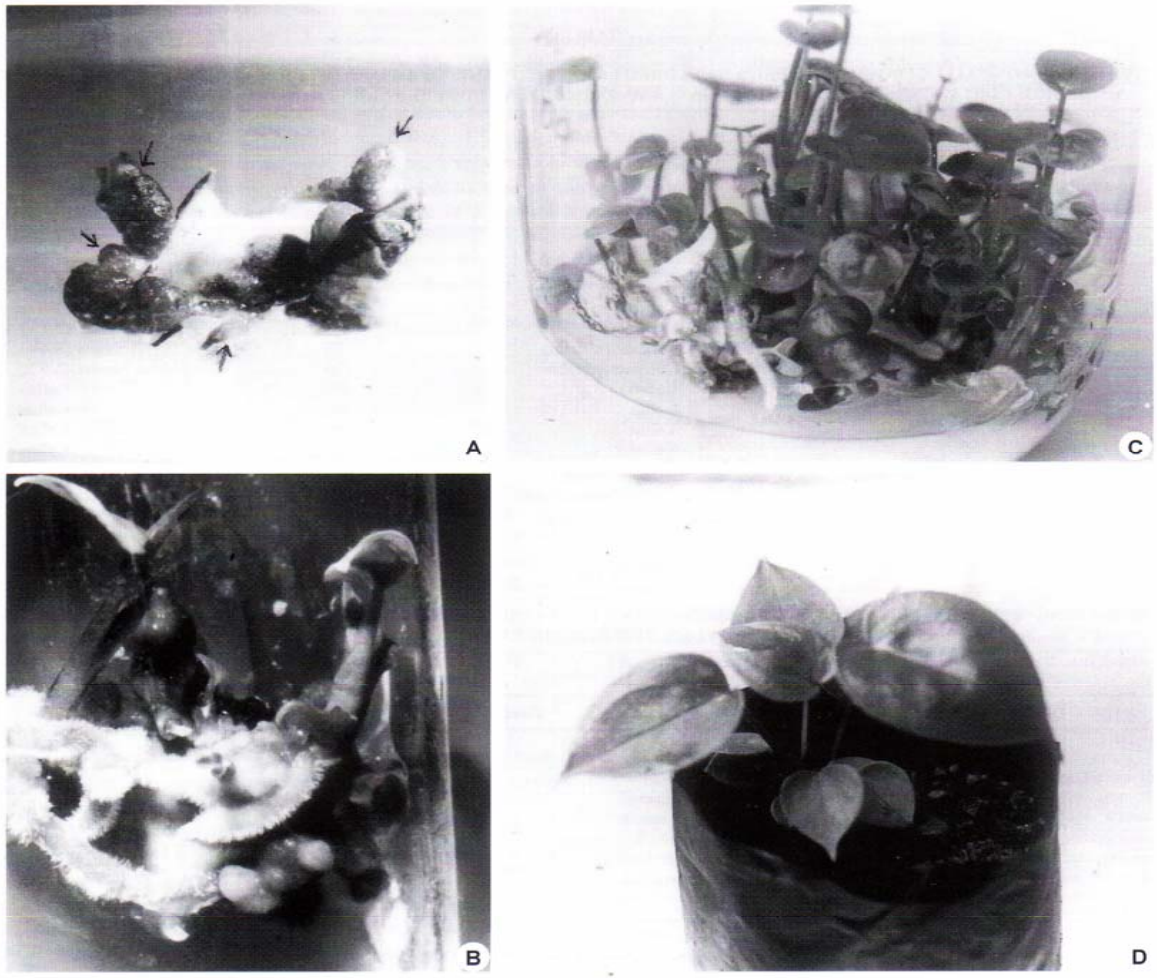


FIG. 1. *In vitro* propagation of anthurium cultivars through direct plant regeneration from leaf explants (pH 5.5). *A*, Direct shoots on young brown leaf (proximal) explant of cultivar Senator on half-strength MS medium with 1.11 μ*M* BA, 1.14 μ*M* IAA, and 0.46 μ*M* Kn (60 d; *arrows* indicate shoot buds). *B*, Direct shoots with roots on young brown leaf (proximal) explant of cultivar Tinora Red on half-strength MS medium with 1.11 μ*M* BA, 1.14 μ*M* IAA, and 0.46 μ*M* Kn (60 d). *C*, Shoot multiplication of cultivar Tinora Red on half-strength MS medium with 0.44 μ*M* BA, 1.14 μ*M* IAA, and 0.46 μ*M* Kn. *D*, Plantlet of Tinora Red in polyethylene bag (60 d).

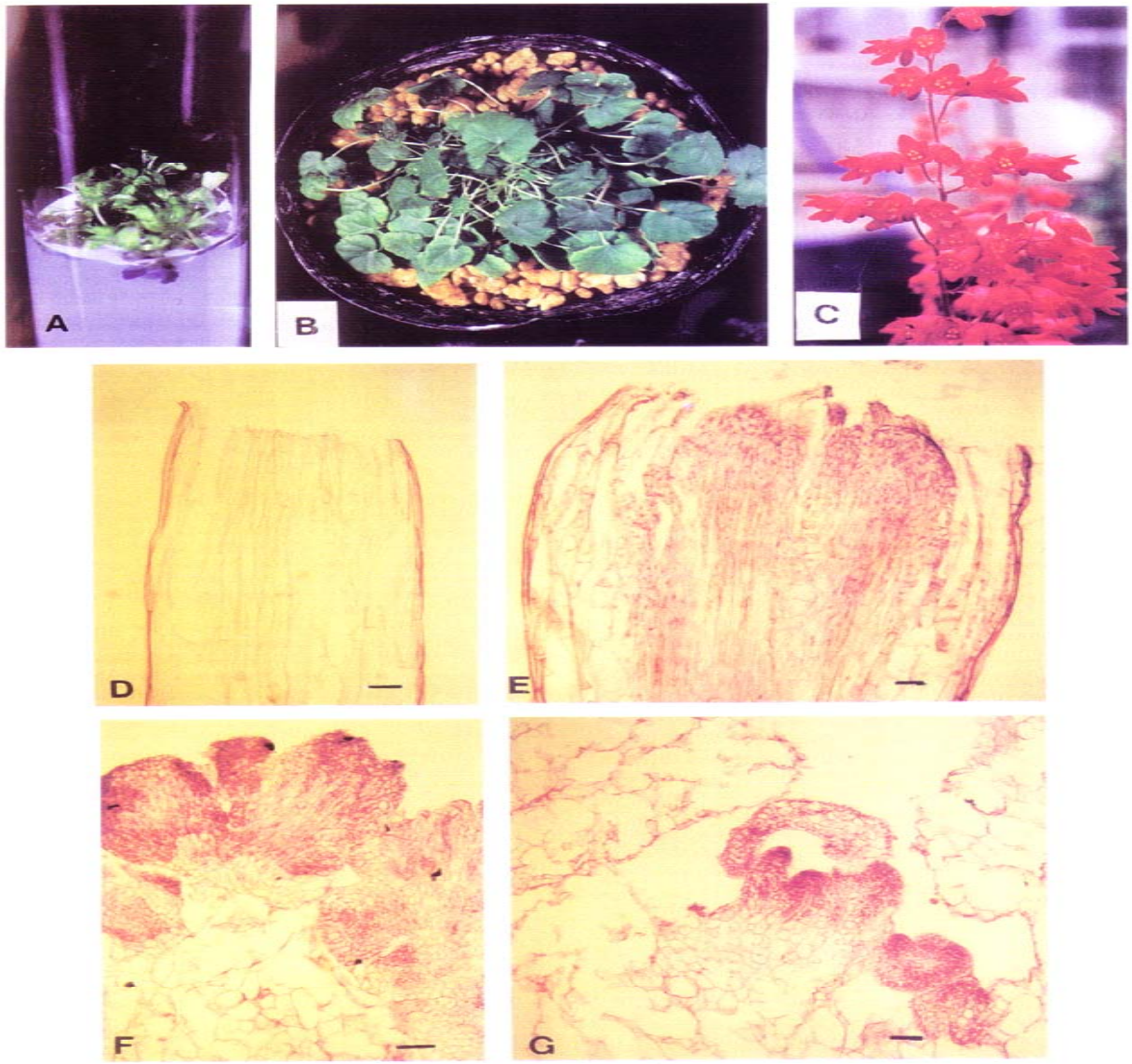
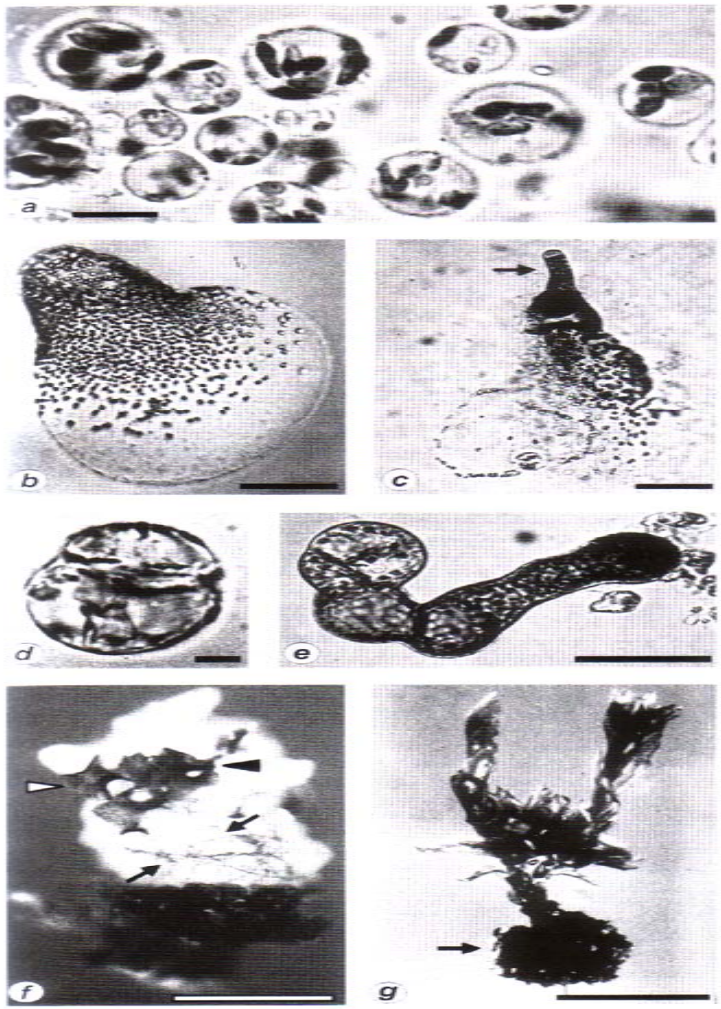


FIG. 1. Shoot regeneration in petiole culture of coral bells and histology, plant growth, and flowering *ex vitro*. *A*, Shoot regeneration from petiole on MS medium containing 0.19 μM NAA and 4.4 μM BA. *B*, An acclimatized plant in a pot. *C*, Flowering of a plant regenerated the following year. *D*, Longitudinal section (15 μM in thick) of petiole showing no cell division at culture initiation. *E*, Dividing cells appearing at the cut end from the inside of the petiole tissue after 1 wk of culture. *F*, Many dividing cell clusters appearing at the cut end after 3 wk of culture. *G*, A shoot apex appearing at the periphery of the cell clusters after 5 wk of culture. *Bars* $(D-G) = 0.1$ mm.



1. Protoplasts and regeneration of *Sphagnum fallax*. *a*. Protoplasts subprotoplasts (with fewer than four plastids) from an apical bud. protoplasts were formed accidentally by cell fractionation during the ation for cell wall digestion (*bar* = 10 µm). *b*. Giant cell with paired ids derived from a capitulum protoplast; 35 d in V-KM (*bar* = 50 µm). ad giant cell and germinating cell (*arrow*) derived from an apical bud gradual osmotic readjustment with B5S for 32 d (*bar* = 50 µm). *d*. Cell er derived from a single apical bud protoplast; 8 d old (*bar* = 10 µm). *e*. nning of the formation of a protonemal plate; 14 d old; B5S = 50 µm). *f*. Filamentous protonemata (*arrows*) and protonemal plates *heads*) regenerated from *Sphagnum* capitulum protoplasts on *Solanum* s; on B5BS for 6 wk (*bar* = 0.5 mm). *g*. Regenerant from a protonemal protoplast with a protonemal plate cluster (*arrow*) and a plantlet; on for 10 wk (*bar* = 1 mm).

Fossard (1977)
3 types of in vitro culture in higher plants :

1.Organized
resembles in vivo vegetative propagation
2. Non-organized
organized part of a plant ---- non organized
3. Non-organized/organized
organ/tissue --- callus or organ or whole individu

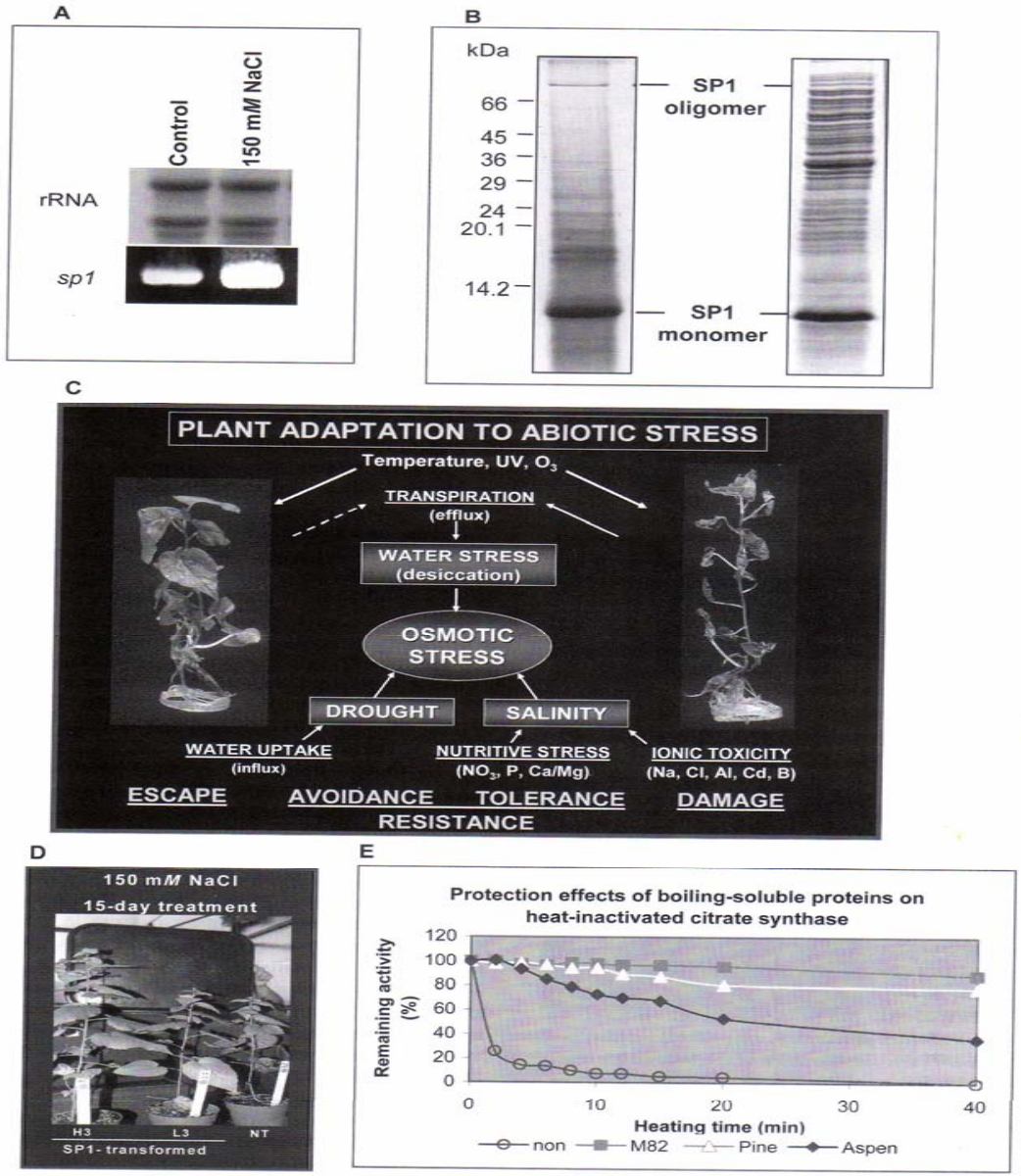


FIG. 1. Aspects of drought and salinity stress. *A*, RT-PCR analysis of control and salt stress (150 m*M* NaCl, 4 h). *B*, SDS-PAGE analysis of aspen total boiling-soluble proteins (*left lane*) and recombinant SP-1 protein in *E. coli* (*right lane*). *C*, Schematic depiction of plant adaptation to abiotic stress. *D*, *sp1*-transgenic aspen line H3, which expresses elevated levels of SP-1, tolerates NaCl stress better than either non-transformed plants (NT) and other *sp1*-transformed plants (e.g., L3) that express normal levels of SP-1. *E*, Boiling-soluble proteins from different plant species (M82-tomato, pine, and aspen) protect citrate synthase from heat-inactivation (Wang et al., 2002b).


THANK YOU