INTRODUCTION

Forests, which are the main source of biomass, fuel, fodder, wood, medicinal plants and raw materials for many industries, are decreasing at an alarming rate due to rapid deforestation. As a result, the gap between supply and demand of the above products is widening. In addition, rapid deforestation deprives employment to many and has serious ecological consequences. Therefore, under the prevailing circumstances there is an urgent need to initiate reforestation programmes on a wide scale to ensure sufficient availability of forest products and long range ecological security.

Hence rapid and massive afforestation is a high priority programme in our country. To meet the requirements of the programme, plant tissue culture of promising trees is recognised as one of the best techniques. Seasons influence the various stages of life activities such as growth, metabolism, reproduction, movement, distribution, behaviour, death etc. Seasonal variation also have an impact on tissue culture, mainly it affects during culture initiation.

The present investigation on in vitro studies and seasonal variation was taken up on two forest and one medicinal plants of considerable economic and ecological

*Terminalia arjuna* (Roxb.) W. & A. commonly known as Arjuna myrobalan belongs to the family Combretaceae. It is a forest tree having both timber and medicinal values. It is of importance in sericulture as its leaves are used as substitute for mulberry leaves to feed the silk worms (Anonymous, 1986). Further this plant is recommended for afforestation programme on saline, alkaline and semi-arid tracts and ravines (Tenjarla et al., 1990).

Seeds of *T. arjuna* have very thick seed coat. So, for germination it takes several days to months. Propagation through cuttings need more plant material that would disturb the growth of parent plants and would lead to the transmission of pathogens.

In order to overcome the above handicaps in natural propagation of the plant, *in vitro* techniques might be beneficial.

*Madhuca longifolia* (Koenig) Macbr. syn. *Bassia longifolia* Koenig is a multipurpose tree species belongs to the family Sapotaceae. It is of immense medicinal, industrial and agricultural significance (Anonymous, 1986). Further, it
has the ability to grow in saline, alkaline and water logged soils which are otherwise unsuitable for cultivating the other crops (Tenjarla et al., 1990).

In Madhuca, seed setting and fruit setting are very low (Kuruvilla, 1989). Conventional methods of propagation, sexual as well as vegetative, have many problems which restricts their multiplication in a large scale. Seed propagation is unreliable because of disease and pest problems, short viability and heavy rains in the natural habitat during the seedling season (Rout and Das, 1993). In nurseries, the seedlings are attacked by Loranthus spp. Moreover, seedling transplantation is risky, since, it has long delicate tap root (Tenjarla et al., 1990). Hence tissue culture techniques offer a viable alternative to overcome the above disadvantages.

Mentha piperita Linn. emend. Huds., (peppermint) perennial herb of the family Lamiaceae, is very commonly used in traditional medicine and is of immense pharmaceutical and industrial significance (Anonymous, 1986).

*M. piperita* is a triploid (2n=72) sterile plant (Sato et al., 1989). It is commonly propagated by runners, which have more chances for transmission of pathogens. Moreover, oil quality and quantity are reduced by pests associated with mint cultivation.
Therefore, in vitro techniques were found to be essential, to produce improved plants on large scale. In the light of the above, the proposed programme of work is initiated on Terminalia arjuna, Madhuca longifolia and Mentha piperita with the following objectives:

1. To obtain multiple shoots from axillary bud explants by micropropagation,

2. to induce organogenesis from leaf calli,

3. to identify somaclonal variants derived from calli by catalase isozyme analysis,

4. to induce rooting in shoots derived from multiple shoots and calli, and transfer of the plants to soil,

5. to regenerate plants through somatic embryogenesis,

6. to isolate protoplasts from in vitro harvested leaves and culture the protoplasts,

7. to analyse the molecular basis of embryogenic calli developed from protoplasts by SDS-PAGE technique and

8. to study the effect of seasons on in vitro culture.