INTRODUCTION

Vegetative propagation of plants by rooting stem cuttings is practised in forestry and horticulture to obtain plants of desired genetic constitution within a short span of time and also for the multiplication of species which do not reproduce well by other means.

The production of adventitious roots on stem cuttings is basically a problem of organogenesis. As each meristematic cell is totipotent in nature, it may be assumed that a detached organ possesses an inherent capacity of restorative regeneration. However, root regenerating capacity of stem cuttings many a time, does not express itself because of the lack of some of the
internal and/or external conditions that are necessary for the expression of this potentiality. The elucidation of the factors that influence regeneration and the role that each of these factors plays has, therefore, been a subject of fundamental and applied interest and has engaged the attention of plant physiologists for a long time. An extensive study of the subject has shown that amongst the various factors that control the process of regeneration plant hormones and nutrition are of utmost significance.

Van der Lek, the Dutch scientist suggested as early as in 1925 that the initiation of roots on stem cuttings was caused by a hormone(s) that was produced in the developing buds and was conducted to the base. After the isolation of auxin, its efficacy as a root initiating substance was tested and it was in the year 1934 that Thimann and Went indicated that root formation in the plant was controlled by endogenous auxin. Indoleacetic acid (IAA) an endogenous auxin and a number of synthetic auxins have since then engaged the attention of researchers in investigation on the physiology of regeneration. A landmark in the history of the study of physiology of regeneration was the discovery of Skoog and Tsue (1948) and Skoog and Miller (1957) who showed that the direction in which cambial cells regenerate is determined by the ratio of auxin to adenine or auxin to kinetin; a high ratio
favouring the formation of roots but a low one the formation of buds and suppression of roots. These studies, therefore, demonstrated the importance of a proper balance between different endogenous factors in problems of regeneration. The mechanism of action of auxins in regeneration, however, is yet to be fully elucidated.

The rooting of stem cuttings is also dependent upon nutritional factors (Jones and Beaumont, 1937; Pearce, 1943; Hyun, 1967). Went and Thimann (1937) showed that sugars exert a positive effect on root initiation. More recently Herman and Hess (1967) showed that etiolated stem cuttings that rooted better than the light grown ones, had substantially higher levels of sucrose, glucose, fructose and proteins. Stoltz and Hess (1966) also reported that there was a marked difference in the total carbohydrate contents of easy- and difficult-to-root varieties of Hibiscus.

Attempts to understand the mechanism of auxin action and its interaction with nutrition in the initiation of root primordia have also been made in more recent years. On the basis of extensive work carried out in this laboratory for the last about 10 years Nanda et al. (1968, 1969, 1970, 1971) considered that auxins play multifarious role. These are concerned in the division of meristematic cells,
in their elongation, in differentiating cambial initials into root primordia and most of all in the mobilization of reserve food materials caused by enhancing in the activity of hydrolyzing enzymes and passing these mobilized sugars to the site of root initiation.

Some attempts have also been made in recent years to elucidate the mechanism of action at cellular and subcellular level. Knypl (1966) reported that the inhibitors of RNA and protein syntheses inhibited the IAA-induced growth of hypocotyl sections of sunflower and reduced the production and subsequent growth of roots on etiolated seedling sections of maize. He concluded from these results that auxin-mediated growth phenomena were dependent on RNA and protein syntheses and suggested that IAA induced the synthesis of mRNA.

In a more recent review of literature relating to hormonal regulation and nucleic acid metabolism, Key (1969) has pointed out that some hormone responses which require the involvement of DNA-dependent RNA synthesis, may be due to direct effect of the hormone at transcriptional level, others at translation level, while certain others are probably not mediated through a mechanism involving RNA or protein synthesis. Although
some excellent reviews on hormone action in general have also appeared from time to time (Trewavas, 1968; Loening, 1967; van Overbeek, 1966; Helgeson, 1968; Osborne, 1965 and Galston and Purves, 1960), surprisingly very little is known of the processes that are involved in the mechanism of auxin action in root initiation at cellular and subcellular level.

It was considered that a study of the interaction effects of auxins with some of the external and internal factors and the changes in the metabolic drifts associated with the production and development of roots on etiolated stem segments treated with inhibitors of nucleic acids and proteins are likely to shed some light on the mechanism of auxin action in the initiation and development of roots. This investigation was, therefore, undertaken using etiolated stem segments of *Populus nigra* and *Salix tetrasperma* and epiphyllous buds of *Bryophyllum tubiflorum* with the following objectives in view:-

i) to study the effects of auxins and nutrition singly and in combination with each other on the production of roots under different environmental conditions;

ii) to investigate the mode of action of IAA and GA₃ on different parameters of growth;
iii) to study the mechanism of auxin action at cellular and subcellular level by employing inhibitors of DNA, RNAs and protein syntheses such as FUDR, FU, actinomycin-D and cycloheximide in the initiation and development of roots; and

iv) to study the metabolic changes that take place in stem segments during the period of initiation and development of roots with a view to correlate them with the contents of DNA, RNA and protein during rooting.

The results obtained in this investigation provide some leads to our understanding of the physiology of rooting in stem cuttings and constitute the subject matter of this thesis.