I INTRODUCTION

In recent years the naturally occurring benzenoid compounds such as leuconanthcyanidin, flavonoids, hydroycinnamic acid, and coumarin in plants have been actively investigated for their potential functions in luding phenolic acids containing lignin and other secondary plant products. Apart from the microstructure of plants, phenolic substances have been studied extensively in view of their marked pharmacological applications (Firbank, 1959) and their development in the disease instance in plants (Fridham, 1962). Bate Smith (1959) had demonstrated the naturally occurring phenolic substances in a wide range of higher plants including ferns, gymnosperms, and angiosperms and stands their systematic distribution as well as find out their occurrence in different organs of various plants.

However, in addition to what has been known as pholalic acids, all have been thought to play an important role in the regulation of plant growth and development. For instance, phenolic acids are considered to be the most common growth inhibitors. The hydroybenzoic acids are extremely common plant constituents (Tomazewski, 1960) and cinnamic acid derivatives such as caffeic, coumaric, and ferulic acids are the common growth inhibitory substances among plants.

Many naturally occurring plant growth inhibitors such as phenolics have a synergistic effect on growth of plants. When they are present at low concentrations they promote auxin induced
gr wth while inhibiting at higher on ntrati ns Van Overb ek
t al (1951) stated that cinnami a ids in their cis configura
tion act s weak auxins and as antagonists f auxin in th trans
form Nitsch and Nitsch (1961) hav sh wn synergestic pr moti ns
of growth by phenolics when indol acetic acid was supplied to the
plant as the uxin S licylic acid p-coumaric cid and oth rs
imply inhibit growth without any syn rgestic pr motion, and
it was n t d alw ys that syn rg stic eff ct depends n the r la
tiv con entrati ns of phenoli compound and auxin

Plant g owth regulati n is brought ab ut by the presen
of g owth inhibit rs in the tissu s particl rly in the early
dev lopmental stages Kawase (1961) has n ti ed that the f rma
ti n of increased content of growth inhibit rs is related to
light Konishi (1954) has corr lat d the gr wth rate with
inhibit r content and found that when inhibitor ntent was low
the growth of the plant was high and a lowering of growth
occurred when high phenolic ntent was present This observa-
tion was upported from the investigation of Basyouni and
Towers (1964) who have stated that maximum ntetration of
phenolic acids wa present in the seedlings 9 days after
germination and the amounts fell off with increasing age reach-
ing a minimum four to five weeks later

Downs and Bortheniek (1956) and Phillips and Wareing (1959)
have attributed the suppression of plant growth under various
photo periods to the formation of inhibitors in the leaves and
the presumed translocation of the inhibitor to the growing point
of the stem. The increased inhibitory content in different organs of the plant such as leaves, stems, roots, and fruits with the advancement of age was observed by Varga and Koves (1959) and they suggested that the increased amount of inhibitory substances in stems and fruits was due to translocation from the leaves.

Masuda (1962) has demonstrated the importance of light in the formation of inhibitory substances. In the etiolated seedlings exposed to light, a marked increase in the amount of substances formed was observed. Another evidence has also been presented by Zucker and Ahrens (1958) who have shown that the formation of chlorogenic acid had a definite light requirement. The influence of light on the formation of phenols in the leaves of Prunus domestica was determined by Hillis and Swain (1957) who found that leaves on the sunny side of the tree had notably higher content of phenolic components than leaves on the shady side.

Biosynthesis of simple phenols in higher plants through the shikimic acid pathway (Neish, 1960) apparently does not have a requirement for light directly but it may be related to the formation of carbohydrate which is the precursor, through photosynthesis. Racusen and Aronoff (1954) have demonstrated the effect of light on the formation of aromatic amino acids using $^{14}\text{C}O_2$. They showed that detached leaves of soybean did not synthesize phenylalanine and tyrosine in darkness, but the
exised leaves produced these compounds from the pr ducts formed in one hour photosynthesis in light. Weinstein (1961) had shown by feeding $^{14}$C labelled quinu acid to the leaves of large number of plants in darkness, the formation of labelled tyrosine, phenylalanine and often shikimic acid. Thus in all the above studies light appeared to have only an indirect effect on phenolic biosynthesis.

Recently much attention has been paid to biosynthesis of phenolic constituents from $^{14}$CO$_2$. The formation of phenylpropanoid units has been studied by several workers (Stone, 1953; Brown et al, 1953) while the conversion of $^{14}$CO$_2$ to lignin by wheat plants during the period of rapid lignification was also studied. Investigations on lignin and lignification in wheat plants have shown that the lignin content is very low in young plants reaching a maximum at maturity. This indicates that there was no obvious lateral ship between the amounts of phenolic acids and the system of lignification (Basyouni and Towers, 1964).

The observations of Davis (1955) and his collaborators have shown the pathway by which carbon dioxide is converted to shikimic acid and to related alicyclic compounds in the biosynthesis of aromatic substances in higher plants. Brown and Neish (1955) have synthesized labelled shikimic acid and fed to cuttings of wheat plant, and found the incorporation of radioactivity in the endogenous lignin. This phenomena presented the evidence for the occurrence of shikimic acid pathway of aromatic biosynthesis in
high r plant Brown and Neish (1955b) extended their studies with radioisotopes to a number of phenicolic compounds which they considered potential intermediates in lignification and strongly indicated that phenylpropanoid carboxylic acids are necessary in lignification. Brown and Neish (1956) found p hydroxycinnamic acid and caffeic acid to be present in plants and are undressed as precursors of lignin rather than phenylalanine. Higuchi (1962) labeled phenylalanine, p-hydroxy cinnamic acid and ferulic acids to cambial tissue cultures of Picea strobus and found all these compounds to be good precursors of coniferyl lignin. Stafford (1960) has made a different approach in evaluating potential lignin precursors. He incubated sections of lamina of Phleum pratense with p-hydroxy cinnamic acids. Ferulic and sinapic acids and showed the formation of lignin-like polymers in the presence of hydrogen peroxide at pH 4.5. She characterized the polymers thus formed by several means with natural lignin.

Rate of lignification during plant growth and development is completely dependent upon the interaction of light and auxin and to a certain extent on the enzyme activities. Siegel (1954) suggested that high levels of auxin in rapidly growing organs would suppress lignin deposition, but with declining auxin concentration accompanying maturity lignification would increase.

Plant growth and development would be governed by basic endogenous growth stimulants, such as gibberellins, auxins and
growth inhibitors. Endogenous auxins have been extensively studied in several plant tissues (Bently 1958; Thimann 1963). A good deal of work is also available on the occurrence of endogenous gibberellin-like substances in different plants (Corran and Phinney 1961). Gibberellins A₅ A₆ and A₈ have been identified in flowering plants by MacMillan et al. (1961) and also by West and Reilly (1961).

Gibberellin content in the extracts of developing bean fruit has been studied by Mitchell et al. (1951). The occurrence of endogenous gibberellins in plants has been examined by several workers (West and Phinney 1958; Murakami 1959). Radley (1958) observed the changes in gibberellin content of seeds and fruits and showed that this substance fluctuates in content during development of fruit. Presence of gibberellin-like substances has also been detected in the seedlings grown in darkness by Murakami (1961) who showed that gibberellin content decreased considerably during the first two weeks after germination in Pharbitis. Wheeler (1960) also demonstrated the presence of gibberellin in the seedlings and in mature seeds by Corcoran and Phinney (1962). Rapidly growing tissues such as expanding cotyledons or leaves are expected to have high amounts.

Corcoran (1959) obtained evidence for such substances from acetone extracts of young seed and fruit of 35 species. Murakami (1957, 1959) have analyzed 15 species of Leguminosae and obtained evidence for the occurrence of gibberellin-like substances. Gibberellins of individual parts of plants such as...
Investigations (Phinney and West 1962) have shown the presence of gibberellin-like substances in acetone extracts of the seedlings of onion plants. Ogawa (1964) has studied the occurrence of gibberellin-like substances in different gans of Pha bitis nil using iced seedling bioassay in coryledons of dark grown seedlings where he observed a gradual increase in a e and f esh weight f coryledonal tissue. Relatively small amounts of gibberellin-like substances were noticed in the roots and hypc of the minating seedlings.

The role of indoleacetic acid oxidase (auxin oxidase) has received good interest in connection with plant growth regulation. This enzyme is of wide spread occurrence in plants and converts physiologically active auxin to physiologically inactivating. Peroxidases seem to be intimately associated in controlling the level of auxin activity. Some components of indoleacetic acid oxidase system act as most important factors (W sting 1960). Severi and workers have studied the activity of auxin oxidase (Stutz 1957; W sting 1960). Galston (1954) stated that auxin oxidase system appears to consist of light activable flavoprotein coupled through hydrogen peroxide to peroxidase. Peroxidase was considered to be the actual component for the destruction of indoleacetic acid. The oxidative decarboxylation of indoleacetic acid by pea enzyme requires a phenolic cofactor as an oxidant for the metallic oxidizer.
Sagi and Gay (1961) Watanabe and Stut (1960) have studied the influence of gibberellin and vitamin B to peri ds on the activity of indoleacetic acid oxidase in *Lupinus albus*. They have also shown an inverse relationship between phenol content and auxin oxidase activity. Sagi and Garay (1964) stated that correlation between IAA oxidase and total phenol content was conspicuous. When IAA oxidase activity decreased there was a sudden rise in phenol content. This observation suggests that IAA oxidase plays an active role in the regulation of plant growth and the phenolics indirectly as growth stimulants by controlling the endogenous levels of IAA. The enzymatic breakdown of auxin was dependent upon the relative amounts of phenolic activators and inhibitors. The activity of auxin oxidase was based on the levels of individual phenolic substances present in the tissues. Caffeic and chlorogenic acids act as IAA oxidase inhibitors (Robin and Klein, 1957) whereby sparing IAA and resulting in growth promotion. On the other hand, coumaric and ferulic acids activate auxin oxidase thus inhibiting growth (Gortner et al., 1958). These native phenols can exert their growth regulation in terms of influencing the enzymatic auxin decomposition. Thus it appears phenols would act both as activators and inhibitors of IAA oxidase by controlling the level of endogenous auxin.

As regards the relation between gibberellin and auxin oxidase there are different views. Gibberellin is supposed to
act as an auxin sparing mechanism by bringing about an inhibition of auxin oxidase activity (Vlotos and Meudt, 1957) Contrary to this observation gibberellin has been shown to have no effect on auxin oxidase activity (Sagi and Garay 1961) Siroi and Parups (1965) have evidence to suggest an altogether different mechanism for gibberellin action independent of auxin oxidase system Thus the relationship between auxin oxidase and gibberellin on the one hand and the relation of both these to phenolic compounds in controlling plant growth is not clearly understood at the moment

The principal object of the present investigation, therefore, is to understand the interaction of endogenous phenolic substances gibberellins and auxin oxidase in controlling plant growth

With this object in view, a study of the qualitative and quantitative changes in phenolic constituents endogenous gibberellins and auxin oxidase (as measured by peroxidase) in the seedlings of Cucumis melo grown under different environmental conditions was made in relation to growth and elongation To understand the mobilization and translocation of phenolics within the plant, individual parts of the seedlings were analyzed for phenolic constituents Experiments were also conducted to gain an insight into the rate of phenolic biosynthesis in relation to age
II MATERIALS AND METHODS

A. Plant Materials

The plant materials selected for this investigation were
(i) **Cucumis melo** L
(ii) **Coleus aromaticus** Benth
(iii) **Allium cepa**, L

Growth conditions

(i) **Cucumis melo** L

(1) Seeds were sown in pans of soil (two parts of red soil mixed with one part of compost manure) and were maintained under natural conditions with 10-12 hour photoperiod at 30° 32°C for seven days (till the emergence of first leaf). They were then divided into three batches and were treated as follows:

(a) This batch continued to be maintained under natural conditions as above

(b) Seedlings in this case were transferred to a static condition of continuous light (2,000 lux) at 26°C

(c) The third batch was transferred to continuous darkness at 26°C

(2) Seeds were germinated on wet filter papers in 6 petridishes at 30°C in the dark and were allowed to grow under etiolated conditions at the same temperature.

In all the above conditions samples were taken at regular two-day intervals for the analysis.