SUMMARY

The present work was undertaken on the following aspects of germination, growth and development in *Sesamum indicum* L. cv. Limbdi:

1. Ascorbic acid (AA) turnover, synthesis of nucleic acids and proteins in relation to enzymes like general peroxidase, AA-FR-peroxidase, protease, RNase, Catalase and lipase during early stages of germination.

2. Effect of different photoperiods on growth, flowering and metabolic as well as enzymic activities involved in the synthesis of ascorbic acid, nucleic acids and proteins.

3. Influence of varying number of photoinductive cycles (SD) on growth and developmental pattern as well as on ascorbic acid and nucleic acid metabolism.

4. Effect of exogenous application of ascorbic acid, RNA and DNA with and without $\text{H}_2\text{O}_2$ on seedling growth and metabolic changes associated with juvenile differentiation.
Experiment - I

50 seeds of *Sesamum indicum* L. cv. Limbdi were germinated in Petri-dishes (12.5 cm. in diameter) at 30±1°C in an incubator using distilled water as a medium. Whole seedlings were analysed at 24-hourly interval for ascorbic acid turnover, nucleic acids, proteins and enzymic activities such as peroxidase, AA-FR-peroxidase, protease, catalase, lipase and RNase upto 96 hours of germination. Dry weights of the seedlings as well as moisture content were also worked out upto 96 hours.

With the initiation of imbibition, moisture content of the seed increases causing rapid hydration of protoplasmic constituents thus activating enzymic systems and metabolism. Lipase, protease, RNase, peroxidase and AA-FR-peroxidase activities increase with advance in germination causing breakdown of complex reserves into simpler and utilisable substances which are translocated to the growing embryo-axis. Dry weight of the seedling decreases with increase in germination period on account of depletion of food-reserves and losses due to respiration. RNA content decreases as RNase activity increases throughout the 96 hour period of germination resulting in the formation of simpler and
soluble nucleotides necessary for the growth of embryo-axis. High level of AA at 24 hour and 96 hour of germination most probably helps in the mobilization of soluble nutrients towards growing point as shown earlier. Utilisation of AA also increases indicating important role of AA during germination. Fluctuations in the level of ASG and NAB are suggestive of the maintenance of the balance between oxidative and reductive atmosphere within the cell as free radicals of AA are produced due to intensified peroxidative action.

Experiment - II

Seeds of *Sesamum indicum* L. cv. Limbdi were sown in earthenware pots (9" diameter). Watering was done daily and manuring of ammonium sulphate and superphosphate (2:1) was given weekly at a rate of 1 gram per pot. Photoperiodic treatment was started when the seedlings had become well rooted. The treatment consisted of (1) Normal day (ND)-12.5 hours of natural day-light; (2) Short day (SD) having 8 hours of natural daylight and 16 hours of darkness and (3) Long day (LD) - Natural day-light supplemented by artificial fluorescent tube-lights at night (having mean light intensity of 42 ft.C.).

Growth in terms of height of the main stem, number of leaves and branches was recorded under all three
photoperiods for 10 plants at weekly intervals. By the method of random sampling, three plants from each treatment were selected for separately recording fresh and dry weights of root, stem and leaf at weekly intervals. Relative growth rate (R.G.R.), net assimilation rate (N.A.R.) and leaf weight ratio (L.W.R.) were also worked out. Flowering data and harvest data were also recorded.

Study of growth characters clearly reveals the fact that vegetative growth is enhanced under non-inductive photoperiod (LD) which prevents flowering. Under inductive photoperiod (SD) vegetative growth is reduced on account of acceleration in flowering compared to ND. Number as well as dry weights of fruits and seeds are more under SD. 1,000 seed-weight also increases under SD.

Experiment - II B

Levels of ascorbic acid (AA), ascorbigen (ASG), ascorbic acid utilisation (AAU), net ascorbic acid bound (NAB), nucleic acids and proteins as well as activities of enzymes like general peroxidase, AA-FR-peroxidase, protease and RNase were determined at various stages of growth development and seed-maturation. Different reproductive organs and their corresponding leaves were analysed only in the case of SD and ND plants. As LD plants did not
flower, only the terminal apex and the subtending leaf were analysed at the time of different reproductive stages of SD and ND plants. Increase in levels of AA, NAB, RNA, DNA and proteins at the time of transition to flowering and during reproductive differentiation clearly reveals the importance of these metabolites in processes associated with development. Simultaneously AA-FR-peroxidase activity also increases accelerating the formation of free-radical of AA which activates many enzymic systems like protease and RNase causing faster mobilization of important constituents for rapid development. ASG content decreases resulting in release of more AA which is rapidly used for active cell-division and differentiation. All these enzymic and metabolic processes are hastened under SD compared with those in ND creating an actively energised atmosphere resulting in early flowering under SD. Similar enhancement of metabolic processes is maintained in the terminal apex of SD and ND compared with that in LD. In LD, metabolism is maintained at a lower rate keeping the plants in vegetative condition. Higher peroxidative activity and higher AA utilisation at certain stages of growth under LD treatment create an oxidative atmosphere resulting into faster breakdown of reserves which are ultimately mobilized and used for the enhancement of vegetative growth. Lower levels of AA,
RNA, DNA and protein in plants under LD at the time of reproductive differentiation under SD and ND is suggestive of the controlling role of these metabolites on the relative rates of cell-division, cell-elongation and cell-maturation which ultimately control the process of reproductive differentiation.

**Experiment - III A**

Sowing, watering and manuring of seeds of *Sesamum indicum* L. cv. Limbdi were done in a similar manner as mentioned in 'Experiment - II' and plants were placed under LD. At the 4th leaf stage (i.e. at 17 days) plants were given inductive treatment of 4-, 8- and 12-SD cycles respectively and at the end of each photoinductive treatment they were again put under continuous light (LD). Growth and developmental studies for different inductive cycles were carried out in the same way as mentioned in 'Experiment II'.

Height of main stem, leaf and branch number as well as dry weights of root, stem and leaf are enhanced under 4-SD compared with those in 8- and 12-SD plants respectively. This suggests that increasing the number of inductive cycles causes acceleration in the developmental process which brings about progressive retardation in vegetative growth.
The vegetative period is progressively shortened by photoinductive treatment of 4-, 8- and 12-SD to 54, 36 and 33 days respectively counting from the day of sowing. If the period is calculated from the day of initiation of the photoinductive treatment, the respective vegetative periods were 37, 19 and 16 days. Plants under LD treatment did not flower.

Experiment - III B

Biochemical estimations were carried out in the same way as mentioned in 'Experiment II'. Terminal apex and the corresponding fully expanded leaf below it were sampled for 4-, 8- and 12-inductive cycles separately on different days for a particular reproductive stage, because the stages were reached after different periods under different treatment. Sampling for LD treatment was done on the same day as in inductive treatment for comparison. Different reproductive organs and corresponding leaves were also analysed biochemically for 4-, 8- and 12-SD.

Metabolic and enzymic studies clearly reveal that synthesis of AA, nucleic acids and protein as well as peroxidase, AA-FR-peroxidase, protease and RNase activities are accelerated in plants receiving photoinductive treatment. Maximum acceleration in metabolism is observed
in plants treated with 12-SD which points towards the influence of increasing inductive effect on metabolism. If metabolic and enzymic activities of terminal apices are compared to that of LD, it is clearly observed that all the constituents concerned with energy level within the plant are lower under LD and also the enzymic activities are at lower levels. It, therefore, becomes clear that after receiving photoinductive cycles plants show greater tendency towards flowering even when placed subsequently under non-inductive photoperiod. The photoinductive stimulus appears to be quantitative in nature up to a certain level.

**Experiment - IV**

50 seeds of *Sesamum indicum* L. cv. Limbdi were germinated in Petri dishes (12.5 cm. diameter) in lower (25 ppm), higher (200 ppm) concentrations of AA, RNA and DNA as well as their combinations and germination process was carried out up to 96 hours in an incubator at (30±1°C). Same set was repeated adding a very low concentration of H$_2$O$_2$ (0.01 v/v) to each medium.

% Moisture content, length of seedlings and fresh and dry weights were recorded in all the media at 24-hourly interval up to 96 hours. There is an increase in moisture
content of seedlings germinated in H$_2$O$_2$ indicating an inductive effect of H$_2$O$_2$ on imbibition process. This increase in moisture content causes greater hydration of protoplasm in the seeds consequently, enzymic activities of general peroxidase, AA-FR-peroxidase, protease and lipase are all accelerated by exogenous application of AA, RNA and DNA and this is further accelerated by the addition of H$_2$O$_2$. This not only suggests the important role of AA and核酸 acids in the cell-division processes associated with juvenile-differentiation but also points towards the inductive effect of H$_2$O$_2$ which operates mainly through the formation of free-radical of AA catalysed by enhanced AA-FR-peroxidase activity coupled with active AA synthesis. Synthesis of proteins, histones and nucleic acids also increase under the effect of AA, RNA and DNA as well as H$_2$O$_2$ thus indicating the presence of an active synthetic system created by the free-radical of AA.

Absence of any significant difference or even a slight reduction in the dry weights of the seedlings under the above mentioned treatments can be attributed to the losses due to the mobilization of reserves and respiratory activity. Enhancement in dry matter of seedlings can only start after the synthesis of chlorophyll and initiation of photosynthesis.
Considering the marked enhancing effect of $\text{H}_2\text{O}_2$ on extension growth as well as on the biosynthesis of metabolites like AA, DNA, RNA, proteins and related enzymic activities as well as taking into account the effect of photoperiodic treatments and inductive photocycles on the metabolic events leading to flowering; it may be postulated that at various stages of growth and development of a plants, free radicals of AA create an integrated mechanism of electron flow and biosynthesis of cell-constituents (including macromolecules) thus enabling the processes of cell-division and cell-differentiation to proceed at a faster or a slower rate depending upon the rate of electron-flow.

Finally, it can be said that this work on *Sesamum indicum* L. cv. *Limbdi* a qualitative short-day plant, confirms the ascorbic acid-nucleic acid-protein metabolism concept of growth and development which was advanced earlier from this laboratory in case of wheat, barley and other thermophobes.