The backward state of affairs in technology and the wretched and unscientific management of Indian agriculture as a whole have adversely affected our national economy. Therefore, development of a sound fruit industry which could have been an asset to the developing economy of our country in many ways, has remained unfulfilled. This coupled with the unskilled methodology adopted in horticulture and improper facilities of transit and storage have further aggravated the situation and much of the produce is wasted on this account alone. Besides, tropical climatic conditions of our country are best suited to the development of phytopathogenic diseases. This calls for an urgent attention of plant pathologists and policy makers alike. Hence the present problem dealing with the biochemical and pathological investigations on soft rot of pear fruits is of great relevance and applied value.

Pear is one of the most important fruit crops of India and occupies a prominent place next to apple among temperate fruits. Fresh pear fruits are usually relished as dessert. Besides, they are processed into a variety of products such as syrup, jam, jelly, marmalade, juice, wine, murabbas etc. In the form of these preparations, the produce is available to the population throughout the year. Pear is cultivated mainly in the hilly regions from where it has to be transported to various places to
be marketed. It is mainly during this transit that a portion of the produce undergoes damage and biodeterioration. The losses suffered on this account are enormous. The present fruit rot disease of pear was encountered during a survey of post-harvest storage and transit diseases in the local market and godowns and was found to be mainly due to infection caused by *Botryodiplodia theobromae*. On account of its great economic importance due to severe losses caused by this destructive pathogen and lack of information in connection with this disease, this problem has, therefore, been selected for the present work. During these investigations, an attempt has been made to study pathological, physiological and biochemical aspects of this disease and efforts have also been made to find out some suitable control measures for the disease.

The present causal organism, *B. theobromae* was isolated from diseased pear fruits. The appearance of typical water soaked areas on the artificially inoculated fruits leading to the browning of the fruit surface and softening of the internal tissues are the characteristics of soft rot symptoms which could be compared with those of diseased pears normally found in nature. At an advanced stage of rotting, wrinkles, corrugations developed in the fruit rind. Eventually the fruit turned pulpy and dilapidated.
Pathogenicity was successfully established by satisfying the Koch's postulates. *B. theobromae* appeared to be a wound parasite requiring some kind of injury on fruit surface which probably also supplied nourishment to support the pathogenic infection. This is supported by the fact that the agar disc inoculum was more effective in causing the disease than the disc free inoculum. Intact fruit surface frustrated any attack of infection by the pathogen.

Among various environmental factors, temperature and relative humidity caused noteworthy affects on rot development. Maximum rotting was favoured at 26-30°C and 100-67% RH. Temperature below 15°C and above 35°C remained uninfluential so also the RH which proved less effective below 67%.

Age of culture did not show much effect on the virulence of the pathogen and severity of disease. However, comparatively young cultures were found to be more aggressive than the aged cultures.

Several vital metabolic processes are affected by pathogenic invasion bringing about a change in the chemical composition of the host. This has a bearing on the nutritional value of the host rendering it unpalatable and unfit for human consumption. Post-infectional changes in some of the prominent metabolites like sugars, amino acids, organic acids, phenols, ascorbic acid and total carbon and nitrogen contents were observed.
Experimental results showed the presence of sucrose, glucose and fructose in healthy fruits. Out of these glucose and sucrose remained less affected during pathogenesis. However, concentration of fructose was found fluctuating intermittently. Interestingly, raffinose, a trisaccharide along with an unknown sugar appeared in the diseased tissues only which could be detected even after 15 days of infection.

Post infectious alterations in amino acids appeared more remarkable. Nine ninhydrin positive spots were present in the healthy fruits. During pathogenesis, amino acids like lysine, leucine, phenylalanine and cystine first increased up to 8 days and then declined. Most of the amino acids had disappeared by 15th day of disease incubation. These amino acids were either used up by the pathogen or got incorporated into the protein fraction of host-parasite complex. However, appearance of alanine, tyrosine, methionine and the unknown one in the diseased tissues could probably be due to pathogen stimulated proteolytic and transaminative reactions.

Out of five organic acids, tartaric, succinic, maleic, malic and citric observed in healthy tissues, the latter three were found to decline gradually after infection. Consumption of malic acid by the pathogen probably weakened host resistance to the growing infection as this compound is a well known resistant factor in several cases. Fumaric acid appeared temporarily for
a short duration in the diseased tissues and then vanished. None of the organic acids were, however, observed to increase in quantities during disease development.

Surprisingly, only one phenolic compound could be detected in the samples of healthy and diseased fruit extracts. Its concentration remained unaffected by the development of rotting. This indicated that phenolics were not much involved in pathogenesis.

Healthy pear fruits were found to contain meagre amounts of ascorbic acid which however was found to decrease noticeably during pathogenesis. This could be responsible for further lowering the nutritional quality of the fruit.

Analysis of total carbon and nitrogen contents of the healthy and infected pear fruits revealed that C/N ratio underwent a significant change. Organic carbon was found to decrease gradually while total nitrogen contents increased progressively along with the advancing disease.

Cell wall degrading enzymes are intricately involved in the phytopathological process of disease causation and pectolytic and cellulolytic enzymes appear to be the most important ones imperative in fungal pathogenesis. Healthy tissue extracts demonstrated mild activities of glycosidases, PG and PMG but no PME activity could be detected. Pectolytic enzyme activity
increased after infection where PG was found to be more active than PMG indicating more important role of the former during pathogenesis. However, their activities declined after 6th day of incubation. Diseased fruits also exhibited high PME activity which probably supported and contributed to pathogenesis in conjunction with PG. Besides, diseased tissue extracts were also found to be strongly cellulyolytic in nature.

Healthy fruits exhibited very poor cellulyolytic activity. But cellulase activity was observed to increase rapidly in the infected tissues with the advancement of incubation period. Cellulases were considerably active even during the later stages of disease development when PG activity had already sharply declined. This clearly indicates participation of cellulases in the later stages of pathogenesis. Cellulases were also co-dominant with the PG activity in the early phase of fruit rotting. Therefore, a synergism between PG and cellulase could also be conjectured.

Pectolytic enzymes preferred the acid range of pH displaying maximum activity around pH 4.6 while cellulases were found to be less sensitive to the pH conditions of the medium and were quite active between pH 4.0 to 7.0, their peak activity being recorded at pH 5.5.

A poor amylolytic activity was detected in the extracts of 8 day old diseased fruits which could not be traced in extracts
of fruits with more advanced stages of pathogenesis. Healthy fruits were completely devoid of any amylolytic activity. As such it appears that amylases did not play any important role in the diseased host tissues.

With a view to devise suitable control measures to combat the soft rot of pear fruits caused by the *B. theobromae*, certain well known chemotherapeuticants such as fungicides, antibiotics, phenolics and volatile compounds were tried against the test pathogen. A few effective treatments were then selected and employed in the form of 'fruit dips' to assess their efficacy against the disease development 'in vivo'. Besides, their effects on cellulase activity and on mycelial respiration of pathogen were also worked out in order to have some understanding about their mechanism of action.

Out of ten fungicides used, vegoll, thiram, Cu-oxychloride, aretan, miltox, vitavax and difoltan were found to be highly efficaceous against the mycelial growth of the test pathogen. Among these, vegoll, thiram, aretan and Cu-oxychloride could also as successfully control the fruit rot. Vegoll and thiram proved most successful and could very safely be employed for controlling the disease most effectively even at the lowest concentrations. These fungicides were also found to be antirespiratory and anti-cellulolytic to varying extents.

Out of ten antibiotic compounds employed, aureofungin, nystatin and talsutin exhibited maximum inhibitory activity against
the test pathogen. But when these chemicals were put to the 'in vivo' tests, only nystatin was found effective controlling the fruit rot satisfactorily. Nystatin was also found to be anticellulolytic. However, aureofungin and talsutin which also suppressed cellulase activity failed miserably in controlling the fruit rot. It appears that their anticellulolytic activity was in some way modified in the presence of certain host components. Mycelial respiration remained more or less unaffected by these antibiotics although aureofugin caused some stimulation.

In case of phenolic substances, out of the ten compounds tried, only three viz., \( \alpha \)-naphthol, \( \beta \)-naphthol and vanillin proved considerably effective against mycelial growth. Of these, only \( \beta \)-naphthol (1000 ppm) controlled the pear fruit rot significantly.

Amongst eight volatile compounds, six were found to be fungitoxic to varying extents. However, ammonia and acetone were found to be extremely toxic showing very strong inhibition of mycelial growth and therefore these may also be employed to prevent the pear fruit rot. Pyridine on the contrary was found to be stimulatory for fungal growth and hence of no significance.