CHAPTER VI

SUMMARY OF STUDIES ON WHEAT SEEDLINGS
This experimental work was undertaken to investigate the effects of water stresses and low temperature stresses on wheat seedlings and to ascertain the associated metabolic changes in wheat seedlings due to these stresses. Water stresses were induced by germination of the seedlings under osmotica and by desiccation.

Experiment I

Certified and graded seeds of wheat (Triticum aestivum, L.) (Var. Kalyan Sona) were germinated in sterilized, clean petridishes (9.0 cm. diameter) lined with sterilized filter paper (Whatman No.1) under D.W. (which served as the control) and various osmotica like NaCl, Sucrose, Mannitol and PEG (6,000 Mol. Wt.) (A.R. Grade) upto 120 hours at room temperature (28 ± 2°C) in normal day light.

Extension growth of seedlings - root and shoot, fresh weight and dry weight as well as the associated biochemical changes were studied after 48 hours and 120 hours of germination in embryo axis and endosperm separately. The
following metabolites like Starch, RNA, DNA, Protein, Histone, Total Sugar, Reducing Sugar and Non-Reducing Sugar and enzymic activities like Amylase, RNAse, Protease, Invertase, Catalase and Peroxidase were studied.

Germination was arrested by osmotica and was more under 5 osmotica, than 1. Shoot growth and root growth was highly reduced under osmotica except 1 NaCl and 1 Mannitol which enhanced it compared with the seedlings germinated under D.W.

Fresh weight of embryo axis increased with the advancement of germination and fresh weight of embryo axis of seedlings germinated under osmotica for 48 hours showed no much change as compared with those under D.W. Germination under osmotica for 120 hours, however, lowered the fresh weight except under 1 NaCl and 1 Mannitol, wherein they increased it. Fresh weight of endosperm of seedlings germinated under osmotica were lowered.

Dry weight of embryo axis increased with the advancement of germination. Dry weight of embryo axis of seedlings were more when germinated under osmotica than under D.W.
Seedlings under 4 osmotica had more dry weight than 5. Dry weight of endosperm lowered with the advancement of germination. Dry weight of endosperm were more when germinated under osmotica. Seedlings under 5 osmotica had more dry weight than 1.

Per cent (\%) moisture content of embryo axis increased with germination time. Moisture content in embryo axis of seedlings were generally lower when germinated under osmotica than under D.W. However, moisture content in embryo axis of seedlings germinated under 5 PEG for 120 hours increased it. Per cent (\%) moisture content of endosperm increased with germination period. Moisture content in endosperm of seedlings were generally lower when germinated under osmotica. 5 osmotica caused greater lowering effect than 1.

Catalase activity in embryo axis increased with the advancement of germination. This activity was lowered by prolonged germination under osmotica, beyond the activity of seedlings germinated under D.W. However, the activity of embryo axis of seedlings germinated for 120 hours under 1 NaCl was increased. Catalase activity of endosperm increased with germination. This activity was lowered by prolonged
germination under osmotic stress. Peroxidase activity of embryo axis of seedlings under osmotic stress were lower than those germinated under D.W. This activity was lowered more under 1 osmotic than under 5 in the seedlings germinated for 120 hours. Peroxidase activity of endosperm of seedlings under osmotic stress were lowered except in the seedlings germinated for 120 hours under 1 PEG, which increased this enzymic activity.

Amylase activity of embryo axis increased with germination. Amylase activity of seedlings germinated under osmotic stress were lowered than those germinated under D.W. 1 osmotic caused more lowering effect than 5. Amylase activity of endosperm increased with germination. Amylase activity of seedlings germinated under osmotic stress were lowered except in the seedlings germinated for 120 hours under 1 NaCl, 1 PEG and 5. Mannitol wherein it was higher.

Starch content of endosperm declined with the advancement of germination. In seedlings germinated for 48 hours under osmotic stress lowered this content than those germinated under D.W. whereas seedlings germinated for 120 hours under osmotic stress increased this content.
Invertase activity of embryo axis increased with the advancement of germination period. This activity was lowered in the seedlings germinated under osmotica than those germinated under D.W., except in the seedlings germinated for 48 hours under 1 NaCl, in which the activity increased. Invertase activity of endosperm increased with the advancement of germination. This activity was lowered in the seedlings germinated under osmotica. Reducing sugar content of embryo axis increased with germination. This sugar content increased in the seedlings germinated for 48 hours under 1 osmotica and 5 Sucrose as compared with those germinated under D.W. While in the seedlings germinated for 120 hours under osmotica this sugar content was lowered except under 1 Mannitol, 5 Sucrose, and 5 PEG, wherein this sugar content was increased. Reducing sugar content of endosperm increased with germination. This sugar content was increased in the seedlings germinated for 48 hours under 1 osmotica while this sugar content was lowered in the seedlings germinated for 120 hours under osmotica.

Non-reducing sugar content of embryo axis increased with germination. In the seedlings germinated under osmotica for 48 hours there was little change in the non-reducing
sugar content whereas in the seedlings germinated for 120 hours under osmotica it decreased except under $\text{1 NaCl}$ where it increased.

Non-reducing sugar content of endosperm increased with germination. In the seedlings germinated under osmotica, this sugar content was lowered. Total sugar content of embryo axis increased with germination. In the seedlings germinated under osmotica this sugar content was lowered but in the seedlings germinated under $\text{1 NaCl}$ for 120 hours this sugar content was increased. Total sugar content of endosperm increased with germination. Endospermic total sugar content was lowered in the seedlings germinated under osmotica.

Protease activity of embryo axis increased with germination. This activity was lowered in the seedlings germinated under osmotica as compared with those germinated under D.W. except under $\text{1 NaCl}$ when germinated for 120 hours wherein this activity was enhanced. Protease activity of endosperm followed the same trend as that of embryo axis.
Protein content of embryo axis of seedlings germinated under osmotica was lowered. Protein content of endosperm of seedlings germinated under osmotica for 48 hours was little affected while in the seedlings germinated for 120 hours this content was lowered except under 1 sucrose wherein there was an enhancement. Histone content of embryo axis and endosperm of seedlings germinated under osmotica was lowered.

RNase activity, RNA and DNA content of embryo axis and endosperm increased with germination. Germination of seedlings under osmotica adversely affected RNase activity, RNA and DNA content than those germinated under D.W.

Experiment II

Certified and graded seeds of Wheat (Eriticum aestivum. L.) (Var. Kalyan Sona) were germinated in sterilized, clean petridishes (90 cms. diameter) lined with sterilized filter paper (Whatman No. 1) for a period of 120 hours at room temperature (28 ± 2°C) in normal day light in D.W. 48 hours and 120 hours germinated seedlings were desiccated for 120 hours. After every 24 hours some of these
desiccated seedlings were taken out and analyzed for the biochemical estimations in embryo axis and endosperm separately. The following metabolites like Starch, RNA, DNA, Protein, Histone, Total Sugar, Reducing Sugar and Non-Reducing Sugar and enzymic activities like Amylase, RNAase, Protease, Invertase, Catalase and Peroxidase also Desiccated Weight and Per cent (%) Moisture Loss were studied. The results were compared with the controlled (undesiccated) seedlings.

Desiccated weight of embryo axis and endosperm of seedlings germinated for 48 hours and 120 hours were more than the dry weight of the undesiccated seedlings. Desiccated weight went on decreasing with increase in treatment. Desiccated weight was maximum at 24 hours of desiccation and was minimum at 120 hours of treatment. Per cent (%) Moisture Loss of embryo axis and endosperm increased with treatment period. Minimum moisture loss was at 24 hours of treatment and the maximum was at 120 hours of treatment.

Catalase activity of embryo axis and endosperm of 48 hours germinated seedlings increased with increase in
treatment period and was more than in the embryo axis of the undesciccate seedlings. In 120 hours germinated seedlings, this activity was suppressed by desiccation except in embryo axis under prolonged desiccation of 120 hours, when it was enhanced. Peroxidase activity of embryo axis and endosperm of 48 hours and 120 hours germinated seedlings was suppressed by desiccation as compared with the activity in the undesciccate seedlings. However, prolonged desiccation enhanced this activity beyond the activity of undesciccate seedlings.

Amylase activity in both embryo axis and endosperm of germinated seedlings of 48 hours and 120 hours was lowered by desiccation treatment as compared with the activity in the undesciccate seedlings. Starch content of endosperm of 48 hours germinated seedling was decreased during desiccation. The lowering effect reduced with increase in the duration of treatment. In 120 hours germinated seedlings, this content was not affected by desiccation.

Invertase activity in embryo axis of 48 hours seedlings was lowered by 24 hours of treatment and other periods did
not affect as compared with the activity in the undesci\ncoated seedlings while in endosperm this activity was suppressed by desiccation. In 120 hours germinated seedlings in embryo axis, prolonged treatment enhanced this activity while in endosperm it was suppressed by desiccation treatment. Reducing sugar content of embryo axis and endosperm of 48 hours germinated seedling was not affected much by desiccation treatments, while in 120 hours germinated seedlings in embryo axis prolonged treatment period enhanced this sugar content while in endosperm it was lowered. Non-reducing sugar and total sugar contents of embryo axis and endosperm of 48 hours and 120 hours germinated seedlings was enhanced by prolonged desiccation treatment except in the endosperm of 48 hours germinated seedlings, wherein desiccation treatment has not affected much this sugar content. Thus on the whole, carbohydrate metabolism was adversely affected by desiccation treatments.

Protease activity in embryo axis of 48 hours germinated seedlings was slightly increased by longer periods of desiccation while in endosperm this activity was suppressed equally when compared with the activity in the
undesiccated seedlings. In embryo axis and endosperm of 120 hours germinated seedlings prolonged treatment enhanced this enzymic activity. Protein content of embryo axis and endosperm of germinated seedlings were lowered by desiccation treatment. However, in endosperm of 48 hours germinated seedlings, 72 hours and 96 hours of treatment enhanced this content. Histone content of embryo axis and endosperm of germinated seedlings of 48 hours and 120 hours was enhanced by prolonged period of desiccation treatment.

RNAase activity in embryo axis and endosperm of 48 hours and 120 hours germinated seedlings was suppressed by desiccation except in embryo axis of 120 hours germinated seedlings. 120 hours of desiccation treatment increased this activity as compared with this activity in the undesiccated seedlings. RNA and DNA content of embryo axis and endosperm of germinated seedlings were lowered by desiccation treatment.

Thus, on the whole, desiccation treatment has highly affected the metabolism in the germinated wheat seedlings.
Experiment III

Certified and graded seeds of Wheat (Triticum aestivum, L.) (Var. Kalyan Sona) were germinated in clean, sterilized petridishes (9.0 cms. diameter) lined with sterilized filter paper (Whatman No. 1) for a period of 120 hours at room temperature (28 ± 2°C) in normal day light in D.W. 48 hours and 120 hours germinated seedlings were kept in a refrigerator, the inside temperature of which was maintained constantly at 10°C. The seedlings were kept for 120 hours. After every 24 hours, some of these seedlings were taken out and analyzed for the biochemical estimations in embryo axis and endosperm separately. The following metabolites like Starch, RNA, DNA, Protein, Histone, Total Sugar, Reducing Sugar, and Non-Reducing Sugar and enzymic activities like Amylase, RNAse, Protease, Invertase, Catalase and Peroxidase were studied. The results were compared with the controlled (untreated) seedlings.

Catalase activity in embryo axis and endosperm of 48 hours germinated seedlings was slightly increased by low temperature treatment as compared with this activity in the controlled (untreated) seedlings. While in embryo axis
and endosperm of 120 hours germinated seedlings this activity was highly suppressed by this treatment. Peroxidase activity in embryo axis and endosperm of germinated seedlings of 48 hours and 120 hours was suppressed by low temperature treatment except in the endosperm of 48 hours germinated seedling, longer period of treatment enhanced this activity as compared with this activity in the untreated seedlings.

Amylase activity in embryo axis and endosperm of germinated seedlings of 48 hours and 120 hours was highly suppressed by low temperature treatment as compared with the activity in the untreated seedlings. Starch content of endosperm of both the germinated seedlings depleted with increase in treatment period.

Invertase activity in embryo axis of germinated seedlings of 48 hours and 120 hours was lowered by all periods of treatment and extent of lowering was invariant with the duration of treatment when compared with the activity in the untreated seedlings. While in endosperm of these germinated seedlings, this activity was lowered
by low temperature treatment. Shorter periods of treatment suppressed the activity more than longer periods of treatment. Reducing sugar content of embryo axis and endosperm of germinated seedlings of 48 hours and 120 hours was lowered by low temperature treatment.

In the seedlings of 48 hours of germination, the fall in sugar content increased with duration of treatment. The fall in sugar content was more in endosperm than in embryo axis. Non-reducing sugar content of embryo axis of 48 hours germinated seedling increased with the duration of treatment so that prolonged periods of treatment enhanced this sugar content more than that of the untreated seedlings. While in 120 hours germinated seedlings this sugar content was lowered by low temperature treatments. In endosperm of these germinated seedlings this sugar content was lowered by this treatment but the lowering effect decreased with the duration of treatment. Total sugar content of embryo axis and endosperm in the seedlings of 48 hours and 120 hours of germination was lowered by all periods of treatment, the extent of suppression being more by shorter periods of treatment. Thus carbohydrate
metabolism was adversely affected by low temperature treatment.

Protease activity in embryo axis and endosperm of 48 hours and 120 hours germinated seedlings was lowered by all periods of treatment as compared with the activity in the untreated seedlings. The extent of lowering being invariant with the duration of treatment. Protein content of embryo axis and endosperm of these germinated seedlings was lowered by all periods of treatment. The fall in protein content was more in 120 hours germinated seedlings than in 48 hours germinated seedlings.

Histone content of embryo axis and endosperm of 48 hours and 120 hours germinated seedlings was lowered by all periods of treatment and 24 hours of treatment caused the maximum lowering effect. Thus the protein metabolism was adversely affected by low temperature treatment.

RNAase activity in embryo axis of 48 hours and 120 hours germinated seedlings was lowered by low temperature treatment as compared with the activity in the untreated seedlings. The lowering effect decreased with increase
in treatment. The same effect was observed in endosperm also. RNA content of embryo axis and endosperm of these germinated seedlings was lowered by low temperature treatment and the lowering effect being invariant with the duration of treatment. DNA content of embryo axis as well as of endosperm of these germinated seedlings was lowered by all periods of treatment more or less equally. Thus the Nucleic Acid Metabolism was adversely affected by low temperature treatment.

Thus, on the whole, low temperature treatment has adversely affected the metabolism in the germinated wheat seedlings.