

5. DISCUSSION

5.1 . Consumption and Assimilation of Food During Dry Part of Summer Due to Starvation on Different Days

5.1.1. Consumption of food :

Nistari : Without methoprene administration : Analysis of the result indicates that food consumption varies significantly among the larvae subjected to starvation on different days. Lowest consumption value recorded is on day 2. Larval duration of the starved batches of 1-4 days has been at par with that of the control. The duration has been shortened by a day in case of starvation on 5th and 6th day. This indicates that during the dry part of summer the obligatory feeding period for Nistari extends upto the 4th day of 5th larval instar. Since 5th and 6th day starvation were the facultative feeding period, the larvae entered into pupation. The results are in conformity with the obligatory and facultative feeding periods in lepidoteran larvae advocated by Bounhiol (1938), and extended in *B.mori* by Legay (1955). Calvez and Fourche (1980) proposed that this change of signal in larvae from one phase to the other is promoted through a change in food intake and metabolic process. Regression equation for consumption (Y) against the day of starvation (X) indicates a negative trend. The observed values of consumption are higher over the predicted values for the starvation days 5 and 6. Compensation of loss in consumption due to daywise starvation reveals that except in the larvae starved on day 2 only a small amount of loss has been compensated by the larvae of all other treatments. The highest amount of recovery has occurred among the larvae of starvation days 5 and 6. This corroborates the finding of Kogan (1972), Schroeder (1976) and Mathavan *et al.*, (1987) who demonstrated that insect compensate consumption loss due to

restriction in feeding by increasing the consumption rate over the expected limits (predicted values). A similar observation has been reported by Muthukrishnan and Delvi (1974) in *Poeciloceris pictus*. However, the food deprivation on day 2 (within obligatory feeding period) has exerted some stress from which the larvae of Nistari race could not compensate food consumption fully after restoration of feeding.

With methoprene administration: In spite of daywise starvation the JHa markedly increases food consumption in all the batches including the larvae of control batch. A higher amount of leaf consumption over that in control larvae has also been observed due to starvation on days 1, 2 and 4 accompanied with the JHa application. The larval duration of methoprene-treated control larvae is prolonged by one day beyond that of the control larvae without methoprene. However, the larval duration has been prolonged by one day due to JHa over that the larvae without the JHa in cases of starvation on day 1 to day 5. Prolongation of larval duration during fifth instar of *B. mori* due to methoprene application is a well known phenomenon (Akai and Kobayashi, 1971; Chang ^{et al.,} 1972; Zau, 1979; Kamada *et al.*, 1979; Kobari and Akai 1978, 1979; Kurata 1981; Shibukawa and Akai, 1981; Chowdhuri *et al.* 1986; Rao *et al.* 1988; Trivedy *et al.*, 1993). Prolongation of larval duration by one day due to treatment with 0.3125 µg/larva, methoprene to 48 hrs old fifth instar larvae has been reported in case of Indian bivoltine race of *B. mori* (Rao *et al.* 1988). Methoprene is known to exert some morphogenetic effect along with the environmental factors thus inhibiting early metamorphosis in insects (Slama 1971; Gilbert ^{et al.,} 1980; Sehnal, 1983). This also holds good for *B. mori*. Thus, the fifth stage larval period is extended resulting in increased food consumption. Regression equation indicates a significant but negative trend for consumption (Y) against day of starvation (X).

This suggests that daywise starvation followed by treatment with methoprene cannot rescue the nutritional impairment. Predicted values show a good fit with the observed values except for the day 2 starvation where consumption value is higher than the predicted value. This indicates that the loss in consumption due to starvation has been rescued by methoprene. Most of the treatments compensated consumption loss after methoprene treatment. Lowest amount of food consumption was on day 6. The obligatory feeding period has been prolonged due to methoprene. This corroborates a similar observation by Calvez (1981) who concluded that methoprene though acted on both obligatory and facultative feeding periods but did not have a control in the transition.

◦ **KPGB : Without methoprene application :** Bivoltine races consume more food than the multivoltine races. Control larval duration has been 8 days. The rate of food consumption increases at higher temperature (Legay, 1958; Waldbauer, 1968; Upadhyay and Mishra, 1991). The increased consumption of food helps insects to meet increased maintenance cost (Muthukrishnan and Pandian, 1987). Results indicate that the best consumption has been recorded for the starvation day 1 and lowest on the days 3,4 and 5. Larval period has been extended by one day over control in case of starvation day 1 and 5, and by half a day in case of day 6. This implies that the obligatory feeding period of this bivoltine race continues upto day 4 or 5 of fifth stage larval life during the dry part of summer. Starvation during facultative feeding period results in release of signal in larvae to undergo pupation. Hence, there is no prolongation of larval life in cases of starvation on day 6 and 7. Paul and Deb (1993) however, obtained obligatory feeding period of KPGB race upto the first three days of the life span of 7 days of fifth instar during spring season (best season for bivoltine). Regression equation indicates a negative trend suggesting an impairment in food consumption caused by starvation stress. The

predicted values were less than observed values for the days 6 and 7 and higher for 3, 4 and 5. This indicates that the loss in consumption can not be compensated even when full feeding is restored in cases of days 3, 4 and 5. This may be due to starvation imposed on obligatory period (Janarthan *et al.*, 1994) and also due to low energy reserve in larva to overcome the stress (Calvez, 1981).

KPGB : With methoprene administration : Irrespective of the day of starvation the total food consumption has been increased upto 4.688 gm/larva by the action of the JHa and there is prolongation of larval life as has been recorded by Rao *et al.* (1988). The highest consumption was recorded on day 1 and the lowest on the day 4. Regression equation shows negative trend confirming that methoprene too could not rescue impairment of consumption due to starvation. Compensation in consumption loss indicates that except for the days 6 and 7 none of the other starvation days could recover the loss. This supports the view of Calvez (1981) that impairment caused due to restriction of food cannot be repaired by treatment with JHa.

KPGB x P₅ : without methoprene administration : The total food consumption in KPGB x P₅ has been 2.853 gm/larva in control. The least impairment of food consumption has been recorded in the larvae of day 2 starvation and the highest in day 5. Food consumption of KPGB x P₅ during dry part of summer is lower than that of KPGB alone. Larval duration in all the starvation levels has been prolonged for a day over that of control larvae except for the day 8. Regression equation expresses a negative trend like Nistari and KPGB. The observed consumption values are higher in days 1, 2, 6, 7 and 8 but lower in days 3, 4, and 5 as computed against the predicted values. This confirms recovery of the loss in food consumption. for the larvae of days 1, 2, 6, 7 and 8 but not for the days 3,

4 and 5. The highest suffered cases are the days 3, 4 and 5 possibly because of starvation on the days of obligatory feeding period when recovery is not possible. Basu *et al.* (1992) have observed that hybrids of NB₇ X KPGB fed for 5 days and starved afterward spun cocoons with good pupation rate. In the present finding the feeding regulation on the days 1 and 2 though involves obligatory feeding period the recovery of earlier consumption loss has occurred because the single cross hybrid are vigorous (Narasimhanna, 1985) and stronger than the pure lines. Further, the higher hybrid vigour helped the larvae to overcome starvation stress during early larval life, which has not been possible for pure lines during the season.

KPGB x P₅ : with methoprene administration : Methoprene has induced a significant increase in food consumption, and has prolonged larval period by one day. The total consumption has been 3.294 gm/larva in control. The highest consumption has been on day 1 and the lowest on the day 5. No significant recovery of consumption loss has been observed on days 2, 3, 4 and 5 which fall within the obligatory feeding period. This is justified by the negative trend of regression equation.

5.1.2 Assimilation

Nistari : without methoprene administration - The total assimilation of food in control larva has been recorded to be 0.747gm . Analysis of the result reveals that assimilation of food differs significantly due to daywise feeding regulation. The lowest quantity of assimilation has been on day 6 starvation. Some amount of loss in assimilation has been compensated in all the treatments except for the day 6. This compensation has been the highest on the day 4 starvation. Waldbauer (1964) has argued that due to rapid rate of feeding by the fifth instar larvae of *B.mori*, ingested food

has very less time for enzymatic activity, thus affecting proper absorption of digested food in the gut. Apart from this, the higher content of crude fibre in food is also responsible for reduced digestibility (Waldbauer, 1964; Horie *et al.*, 1976; Ito and Kobayashi, 1978; Horie and Watanabe, 1983; Benchamin and Jolly, 1984; Anantharaman *et al.*, 1993). Regression equation for assimilation of food (Y) against day of starvation (X) expresses a negative trend. The observed values have a good fit against the predicted values except for the starvation on days 3 and 4 where a higher quantity of assimilation may have occurred in the starved larvae after resumption of feeding. Impairment in assimilation of food have been recorded in all the treatments as compared to that of control. Mathavan *et al.*, (1987) have reported a decrease in assimilation due to starvation. Yamamoto and Fujimaki (1982) in Japanese breeds and Remadevi *et al.*, (1993) in Indian breeds observed a positive correlation between consumption and assimilation of food. They are of the opinion that inspite of positive correlation higher consumption may not result in higher assimilation as this is a strain specific character of *B.mori*. A similar observation has also been reported by Horie and Watanabe (1983), Per^aswami *et al.* (1984), Sumioka *et al.* (1982), Benchamin and Jolly (1984), Naik and Delvi (1987), Anantharaman *et al.* (1993) in *B.mori* and by Singh and Prasad (1990) in *Dasychria mendosa* (Lepidoptera, Lymantriidae). Assimilation in fifth instar larvae in general is affected by nutritional deficiency caused by high content of crude fibre and deficiency of water in food (Waldbauer 1964). But results indicate a higher assimilation in some daywise feeding regulated batches as compared to predicted values suggesting compensation of assimilation loss. This may be due to a cumulative effect of increased residence time of food in gut (Schroeder, 1976) and accumulation of water in body due to feeding stress (Delvi *et al.*, 1988). But when compared with assimilation of food during

wet part of summer it is found that assimilation is lower during dry part of summer. This is probably due to low water content in mulberry leaves during dry part of summer. Legay (1958) indicated that low water content in mulberry leaves hampered digestion in *B.mori* and high water level accelerated the process.

Nistari : with methoprene administration : The assimilation of food markedly increased in all categories of starved larvae after treatment with the JHa. As obligatory feeding period was prolonged by methoprene (Calvez, 1981), the consumption of food was also increased resulting in higher accumulation of food than in the JHa-untreated control larvae. Treatment with methoprene however, lowered the rate of daily assimilation of food. Regression equation shows a negative trend in assimilation, the observed values show a good fit with the predicted values indicating impairment caused has not been compensated except for the days 4 and 5 where assimilation has been almost similar to the control values.

KPGB : without methoprene administration : The total assimilation of food by a control larvae is 1.654gm. Likewise in Nistari, the impairment in assimilation has also been recorded in the starved larvae of KPGB. In spite of higher consumption by KPGB, as compared with that of Nistari, the assimilation rate has been lower. This corroborates the findings of Yamamoto and Fujimaki (1982), Horie and Watanabe (1983), Periswami *et al.* (1984), Remadevi *et al.* (1993), the assimilation is totally a strain specific character and not dependent on quantity of consumption. Regression equation expresses a negative relation suggesting an impairment of assimilation caused by starvation. During dry part of summer the assimilation rate is lower than that in wet part of summer. This may be due to low water content in the mulberry leaves (Legay, 1957),

high content of crude fibre (Waldbauer, 1964) and dry matters (Anantharaman *et al.* 1993). Due to low rate of assimilation the starved larvae were unable to compensate the assimilation loss. Except for the starvation days 1, 6 and 7 the predicted values in case of other days have been higher indicating impairment of assimilation.

KPGB : with methoprene administration : The total assimilation of food in control larvae was 1.947 gm/larva during dry part of summer. Prolongation of obligatory feeding period (Calvez, 1981) resulted in compensation of assimilation loss in case of days 6 and 7. Regression equation indicates a negative trend suggesting that impairment of assimilation could not be compensated.

KPGB x P₅ : without methoprene administration : The assimilation of food in control larvae has been 1.185 gm/larva. The assimilation rate is higher than that in KPGB. Regression equation shows a negative relation. Comparison of observed and predicted values indicates a compensation of assimilation loss on starvation days 1, 6, 7 and 8. The assimilation of food is lower during dry part of summer than during wet part of summer as in cases of Nistari and KPGB. Higher quantity of assimilation of food in KPGB x P₅ as compared with that of KPGB pure is due to strain-specific difference.

KPGB x P₅ : with methoprene administration : The total consumption in methoprene treated control has been 1.369 gm/larva. Methoprene effectively increased assimilation rate but in a slow pace. Compensation of assimilation loss has been recorded in cases of days 6, 7 and 8. This justifies that methoprene when applied to the starved larvae prolongs facultative feeding period too. Regression equation indicates that impairment of assimilation could not be recorded by methoprene. However,

overall assimilation of food has been higher than that of the pure KPGB.

5.1.3 Absolute consumption rate

Nistari : without methoprene administration : The highest absolute consumption rate (ACR) has been obtained for starvation on day 5 followed by days 6 and lowest on day 2 followed by days 4 and 3 during dry part of summer. It is evident from the result that the highest compensation in consumption loss of food due to daywise starvation has been on day 5 followed by day 6. These batches have increased their feeding rate after starvation, which is in support of the finding of Kogan (1972), Schroeder (1976), Mathavan *et al.* (1987). Again regulation of feeding during day 5 and 6 (considered as facultative feeding period) has no impact on physiological stress for respiration. This may be due to gain in energy from food during obligatory feeding period (upto 3rd or 4th day of fifth instar). Similarly, starvation on day 2 has shown the lowest ACR followed by day 4 and 3. This also has happened due to feeding regulation during obligatory feeding period. Janarthan *et al.* (1994) have observed that starvation during obligatory feeding period lowers consumption rate even after the resumption of feeding. Furthermore, the starvation during early part of fifth instar exerts stress causing imbalance in the rate of consumption though the larval duration has been found to be prolonged (Calvez, 1981).

Nistari: with methoprene administration : The absolute consumption rate has been increased significantly due to methoprene. The highest rate has been obtained in the larvae of starvation day 6 and the lowest rate on day 5. But overall result is very much similar in all the days except for the days 3 and 5. Due to prolongation of obligatory feeding period by methoprene treatment, the absolute consumption rate has been increased

and the loss of consumption due to starvation has been recovered to a great extent.

KPGB : without methoprene administration : The highest ACR has been recorded for the days 1 and 7 followed by the day 6 and the lowest on day 4 followed by days 5 and 3. This low ACR on days 4, 5 and 3 of starvation may be due to non recovery during obligatory feeding period (Janarthan *et al.* 1994) and low level of stored energy in larvae to overcome such stress (Calvez, 1981). These batches have faced a higher amount of loss in food consumption which they could not recover when feeding was resumed. But for the days 1 and 7 the consumption loss has been recovered when feeding was resumed.

KPGB : with methoprene administration : The highest ACR has been recorded in cases of day 7 followed by day 6 and the lowest in days 3 and 4 of methoprene-treated larval batches. Treatment with methoprene enhanced the ACR in days 7 and 6 of the facultative feeding period. But methoprene could not increase the ACR of day 3 and 4 of the obligatory feeding period. This might have resulted due to transition from obligatory to facultative feeding period, controlled by 'All or None' principle, and overcoming from starvation stress is dependent on the energy reserve by larvae before undergoing such stress (Calvez, 1981).

KPGB x P₅ : without methoprene administration : The highest ACR has been recorded for the day 2 followed by 1 and the lowest for the days 4 and 5. Compensation of consumption loss due to starvation during obligatory feeding period has been higher in KPGB x P₅, possibly because of hybrid vigour (Narasimhanna, 1985) and energy reserve in early part of the fifth stage larval life (Calvez, 1981). The ACR of the larvae starved on days 7 and 8 has been recorded to be higher over predicted values

which indicates a higher compensation of consumption loss after restoration of feeding.

KPGB x P₅: with methoprene administration :Methoprene has no influence on the boosting up of ACR when the larvae are starved during obligatory feeding period. This is why the obligatory feeding period has been prolonged. Extension of facultative feeding period resulted in compensation of consumption loss for starvation on days 6, 7 and 8, showing an increase in absolute consumption rate.

Hence, it is evident from the results that starvation during obligatory feeding period impairs the ACR.

5.1.4 Absolute growth rate

Nistari: without methoprene administration: The AGR has been the highest for days 5 and 6 and lowest for the day 2. Assimilation of food by larvae is known to be directly related to the tissue growth (Barah *et al.* 1989). Thus, the starvation on day 2 (obligatory feeding period) resulted in a low assimilation of food due to low growth rate. Starvation on days 5 and 6 (during facultative feeding period) the ACR has been higher resulting in better growth rate. Growth of larvae is directly influenced by percentage of assimilated food in body (Yamamoto and Gamo, 1976 Remadevi *et al.* 1993). Starvation on day 2 though have caused a higher ACR, but the growth rate (0.024 gm/day) is slower, the benefit has turned negative due to increase in respiratory rate and increased maintenance cost. Comparatively, a lower ACR on days 2, 5 and 6 indicates a higher growth rate due to low respiratory loss and maintenance cost. This is in support of the finding of Schroeder (1976) who argued that a slow growth rate enhances respiration rate and thus increases the relative maintenance

cost. Starvation during obligatory feeding period has reduced absolute growth rate due to low ability in compensating the consumption loss by the larvae after resumption of feeding. But starvation during the facultative feeding period does not impose such stress and larvae are able to compensate the consumption loss to a greater extent. Hence, the growth is enhanced. This is in support of the findings of Muthukrishnan and Delvi (1974), Mathavan and Muthukrishnan (1976); Muthukrishnan *et al.* (1978), Grabstein and Scriber (1982, Muthukrishnan and Pandian (1984). The authors are of the opinion that increased consumption rate by starved larvae after restoration of feeding enhances growth rate.

Nistari : with methoprene administration : The highest growth rate has been attained on day 6 and the lowest in day 2. Treatment with methoprene could not enhance the growth rate. Tropical silkworms in general have a slow growth rate (Mathur *et al.* 1989) and this rate varies according to silkworm strain (Yamamoto and Fujimaki, 1982). Methoprene can only prolong the feeding duration but has no influence on the acceleration of assimilation and hence, the growth rate is retarded (Kurata, 1981). Methoprene prolongs the obligatory feeding period so growth rate has been slower in during the obligatory feeding period due to increase in respiration and maintenance cost. Both respiration and maintenance cost are found to be higher in case of day 2 starvation.

KPGB : without methoprene administration : Absolute growth rate has been the highest for day 7 and the lowest for day 4 followed by day 3. KPGB exhibits the trend of growth similar to that of Nistari. Growth rate has been impaired due to starvation during obligatory feeding period as these batches could not recover the consumption loss after restoration of feeding. However, respiration and maintenance costs are not so high as

compared to those of day 7, but low absolute consumption rate on day 2 has retarded the absolute growth rate.

KPGB : with methoprene administration: Treatment with methoprene has no accelerating influence on absolute growth rate in batches starved during obligatory feeding period. This indicates that transition from obligatory feeding period to facultative feeding period has not been altered by the action of methoprene as indicated by Calvez (1981). The lowest growth rate has been recorded for the days 3 and 4. This may be due to a high loss caused by respiration and increased maintenance cost.

KPGB x P₅ : without methoprene administration : The highest absolute growth rate has been found in the larvae starved on day 8 and the lowest in day 3. Results indicate that though this hybrid shows a higher vigour than the pure line, still starvation during obligatory feeding period affects the growth rate due to low absolute consumption rate. Higher absolute consumption rate for starvation on day 8 resulted in higher absolute growth rate.

KPGB x P₅ : with methoprene administration : Results imply that application of methoprene has no impact on growth rate. The highest growth rate has been recorded for days 8 and the lowest for day 3. Starvation on any day during obligatory feeding period has reduced the absolute growth rate in this hybrid.

5.2 Efficiency of Conversion of Digested Food (ECD) During Dry Part of Summer Due to Starvation on Different Days

5.2.1. Larval Body

Nistari: without methoprene administration : The ECD to larval body has been better for the starvation days 5 and 6 followed by 1, 3 and 4, and the lowest for the day 2. However, in all the day the ECD values are lower than that of the control larvae. This is in support of the finding of Mathavan *et al.* (1987). The recorded ECD values for the days 5 and 6 are higher than the predicted values. This signifies that the larvae of *B.mori* can compensate the food deficiency due to starvation to some extent by elevating the ECD as reported earlier by Mathavan *et al.* (1987). A similar increase in ECD under restricted feeding has also been observed in *Poecilocerus pictus* by Muthukrishnan and Delvi (1974). In general, the ECD values are low in the fifth instar larvae of *B.mori* (Anantharaman *et al.* 1993) due to reduced digestibility (Waldbauer, 1964). But in the starved larvae due to longer residence time of food in gut, the ECD values have been increased (Waldbauer, 1964). Such an increase has also been noticed by Sumioka *et al.* (1982). Nath *et al.* (1990) and Tzenov (1993) explained this phenomenon as physiological adaptation for starvation stress. Thus, the ECD of larvae starved on days 5 and 6 has been increased than the predicted values. This also supports the earlier observation of Yamamoto and Gomo (1976) and Remadevi *et al.* (1992, 1993) who indicated that ECD for larval body was not dependent on the consumption of food, rather this was due to the efficiency of assimilation.

But starvation during the obligatory feeding (days 1,2,3) period decreased the ECD values, the highest effect was on the starvation day 2. This justifies that though there is an increase in assimilation rate than the days 5 and 6, this has been of no benefit for the larvae. The lower ECD values have resulted from the increased respiratory and maintenance cost as reported earlier by Schroeder (1976) in *Calocalpa undulata*. Furthermore, during dry part of summer the increased ambient temperature reduces the ECD values. At higher temperature more energy is lost in

metabolism as emphasised by Scriber (1977), Reese and Beck (1978) and Muthukrishnan and Pandian (1987). Waldbauer (1968) ascribed that at low humid condition food is digested using metabolic water hence ECD is lowered. Starvation on day 4 which is the juncture of obligatory and facultative feeding periods, showed almost the same value as predicted. Thus, feeding regulation on any day during obligatory feeding period impairs the ECD in larval body.

Nistari : with methoprene administration: Methoprene has prolonged the obligatory feeding period in *B.mori*. Better ECD for larval body has been observed in the larvae starved on day 6 followed by 3 and the lowest value for day 2. ECD has been effectively lowered in methoprene treated larvae than that in the untreated larvae of both control and starved batches. This may be due to low digestibility of methoprene treated larvae as emphasised by Gaaboub *et al.* (1985). Prolongation of larval feeding period together with increased respiration rate and maintenance cost due to daywise feeding regulation resulted in low ECD values. Starvation on day 6 has caused ECD value higher than prediction. This is perhaps due to the JHa application during facultative feeding period. Wigglesworth (1934), Friends *et al.* (1965) have indicated that a critical minimum ration is essential to trigger the secretion of moulting hormone. Thus, day 6 larvae pupated without much increase of maintenance cost. But in early part of fifth instar (upto day 4th) the ECD has decreased due to relatively high maintenance cost caused by high temperature and low humidity during rearing season. High temperature accelerated metabolism for meeting the energy demand (Muthukrishnan and Pandian, 1987) whereas low water content in food substantially decreased the ECD values as water deficiency for digestion of food is met through metabolic water (Waldbauer, 1968). Thus, methoprene could not repair the starvation impairment caused during obligatory feeding period.

KPGB : without methoprene administration : Higher ECD has been obtained from starvation day 5 followed by 3, and the lowest on day 1. In contrast to lower ECD values of Nistari, in bivoltine KPGB race the values have been higher than that of control. Such higher ECD values due to restricted feeding has also been reported by Muthukrishnan and Delvi (1974) in *Poeciloceris pictus* and by Sumioka *et al.* (1982), Nath *et al.* (1990) and Tzenov (1993) in *B.mori*. Tzenov (1993) explained this phenomenon as physiological adaptation by larva against starvation stress. Increase in ECD values in starved batches is perhaps due to higher assimilation of food as indicated earlier by Yamamoto and Gamo (1976). Except for starvation on day 1 in other days of obligatory feeding period larvae compensated the loss by increasing their assimilation efficiency. Day 1 starved larvae could not do so because of low energy reserve (Calvez, 1981). Thus, the high assimilation of food had no positive role on ECD. The observed ECD values have been lower than the predicted values for day 4, 6 and 7. This may be due to low assimilation and high metabolic loss due to respiration and maintenance cost. This supports the finding of Schroeder (1976). Such metabolic loss has been very low in day 5 starved larvae. Thus, the ECD values have been lowered in case of days 1, 4, 6 and 7.

KPGB: with methoprene administration : The ECD values are better for the larvae starved on day 1 and the lowest on days 6 and 7. The observed values are lower in case of days 2 and 3 than the predicted values though the assimilation efficiency is higher than that of days 6 and 7. This may be due to low reserve of energy to withstand starvation during obligatory feeding period (days 2 and 3). The ECD values are the same in both methoprene treated and untreated control batches. This supports the earlier finding by Gaaboub *et al.* (1985).

KPGB x P₅ : without methoprene administration - The ECD values have been better in all the starvation days except in day 1. The observed values have been higher for days 2, 3, 4 and 5 than the predicted values. The absolute growth rate in KPGB x P₅ is higher than in pure KPGB and assimilation efficiency is also higher. Low metabolic loss due to respiration and maintenance cost has been recorded in batches starved during obligatory feeding period. This is the reason for higher ECD value for the days 2, 3, 4 and 5. The better performance of KPGB x P₅ may be due to more resistance of bivoltine hybrids than the pure races against any stress (Narasimahanna, 1985).

KPGB x P₅ : with methoprene administration : Among the starvation days the ECD to larval body has been higher for the days 3, 4 and 5 and lower for the days 6, 7 and 8. On the whole, the ECD values of methoprene-treated and untreated larvae have been lower than that of the control larvae. This result agrees with the similar observation of Gaaboub (1985). The hybrid (KPGB x P₅) has a superiority to KPGB in respect of the ECD value of JHa-treated larval body.

5.2.2. Male Cocoon shell :

Nistari : without methoprene administration : The ECD for male cocoon shell has been relatively better for the day 1 followed by 2, 4 and 6 and lower for the days 5 and 3. According to Horie and Watanabe (1985) 25% of the absorbed energy is allocated for cocoon shell by the larvae. Excepting for the days 3 and 5 the observed values of ECD have been higher than the predicted values in all other starvation days.

Nistari: with methoprene administration : The ECD to male cocoon shell has been higher in the larvae starved on days 2 and 3 than on other days. The ECD values declined significantly in all the methoprene treated

starved batches except in cases of control and day 3, which have maintained almost a similar value. This corroborates Kurata (1981) that the coefficient of utilization of ingested food into silk protein is nearly the same in JHa treated and untreated controls. The starvation stress accompanied with JHa treatment during the obligatory feeding period has no significant negative effect on ECD. But there has been a reduction in the ECD values to male cocoon shells when starvation stress was applied during the facultative feeding period. Even the JHa could not induce the ECD.

KPGB : without methoprene administration : The ECD to male cocoon shell has been better for the day 5 followed by days 3 and 4 and lower on day 1 followed by 6 and 7. On the whole, a better ECD to cocoon shell has been observed for the days 5, 3 and 4. Where assimilation is also high because of reduced respiratory and maintenance costs. But starvation on days 1, 6 and 7 has increased metabolic loss thus decreasing the ECD. Again, formation of silk protein in larva is dependent on the quantity of food consumed in later half of the fifth stage larvae (Fukuda *et al.* 1963). Hence, starvation during later part, on days 6 and 7 comparatively reduced the synthesis of silk protein by lowering the ECD values.

KPGB : with methoprene administration : The ECD for male cocoon shell has been better on starvation day 1 followed by 4 and 5 and lower in days 6 and 7. Action of JHa prolonged the obligatory feeding period in starved larvae on days 2 and 3 but the ECD values have been decreased due to metabolic loss. Akai *et al.* (1973) and Akai (1988) have implicated that the application of the JHa to early fifth instar helps in the synthesis of RNA in silk gland for a longer period, thus, the total volume of silk is increased. This has helped to show better performance after starvation on days 4 and 5. But starvation during the late facultative period caused adverse effect.

KPGB x P₅ : without methoprene administration : A better ECD for male cocoon shell has been obtained for the day 5 followed by 3 and 4 and lower on day 1 followed by 6, 7 and 8. The ECD values have been reduced due to starvation on days 1 and 4, and on the days 6, 7 and 8 because of higher respiratory and maintenance costs.

KPGB x P₅ :with methoprene administration: A better ECD for male cocoon shells has been resulted in the larvae of day 5 followed by 4, and the lower values for the day 8 followed by 1. Treatment with methoprene significantly lowered the ECD values in all the batches as compared to that of the control. Observed values for the days 2, 3, 4 and 5 are higher than the predicted ones. This justifies that the consumption of food in later part of the fifth instar is utilized for synthesis of silk protein (Akai, 1988). Starvation during later part has impaired the ECD due to metabolic loss.

5.2.3 : Female Cocoon shell

Nistari : without methoprene administration : The ECD to female cocoon shell has been better for the days 1,2,4 and 6 but low for the days 3 and 5. In most of the starved larval batches and the control, the ECD values for female cocoon shell have been higher than the ECD to male cocoon shell. Impairment in ECD to cocoon shell has occurred for the days 3 and 5.

Nistari : with methoprene administration : The ECD to female cocoon shell in methoprene treated batches has been relatively better for the days 2 followed by 3, but the lowest has been for the days 1 and 4. Low ECD values obtained in methoprene treated batches as compared to those of the untreated larvae of all the days except 2 and 3. Methoprene has

prolonged the obligatory feeding period and is able to rescue the impairment due to starvation on day 2 and 3. In both methoprene treated and untreated control larvae the ECD has been almost the same as reported earlier by Kurata (1981).

KPGB : without methoprene administration : Horie and Watanabe (1985) emphasized that ECD values are higher in female larvae as they allocate energy both for cocoon shell and egg production. Thus, the starved female larvae are more sensitive than starved male larvae (Kadono Okuda *et al.* 1986; Tzenov and Petkov, 1993). Hence starvation in early part of fifth instar and also during the days 6 and 7 (facultative period) has reduced the ECD values due to metabolic loss. But starvation on the days 3, 4 and 5 has resulted in relatively better values due to higher synthesis of silk during part when feeding was continued.

KPGB : with methoprene administration: The results are found to be the same as the ECD to male cocoon shell. Methoprene has induced in synthesis of silk protein inspite of lower ECD values compared with the untreated batches.

KPGB x P₅ : without methoprene administration : The ECD to female cocoon shell is better for the larvae starved on day 5 followed by day 3. The lower values obtained has been on day 1 followed by the days 6, 7 and 8. As the female larvae allocate energy for silk production towards the later half of life, the better efficiency for cocoon shell has been obtained for days 2, 3, 4 and to some extent for 5. The result resembles that of the KPGB.

KPGB x P₅ : with methoprene administration : A better ECD for female cocoon shell after treatment with methoprene has been obtained from the

larvae starved on day 5 followed by 4 and lower values for day 8. Methoprene effectively lowered the ECD values in all the treatments as compared to that of untreated.

5.2.4. Female Pupa

Nistari : without methoprene administration : The female larvae of *B.mori* utilize energy for egg production from food assimilated during early part of the fifth instar and whole of the fourth instar (Fukuda *et al.*, 1963). Hence, starvation on days 4 and 6 did not cause much impairment in gaining female pupal weight. But starvation during the obligatory feeding period reduced this efficiency. The conversion of dry matter of food for pupal body has been found to be more in case of female larvae (Horie and Watanabe, 1983; Periswamy and Radhakrishnan, 1985).

Nistari : with methoprene administration : The ECD to female pupa is quite better for the day 3 and low for the day 2. Better ECD has also been obtained for the days 4, 5 and 6 as compared with the predicted values. Prolongation of obligatory period due to methoprene helps the larvae batches starved on early days to store more energy for pupa after restoration of feeding. But starvation on day 2 resulted in more metabolic loss thus reducing the ECD value. This supports the finding of Muthukrishnan and Pandian (1987).

KPGB : without methoprene administration : Higher values of ECD to female pupa has been obtained for day 5 followed by 3 and lower values for day followed by 7. Observed values are higher than the predicted values for the days 2, 3, 4 and 5. The result justifies that irrespective of the day of starvation during obligatory feeding period, the female larvae

first allocate dietary resource for egg production . Hence, starvation during the facultative feeding period significantly reduced the ECD values (days 6 and 7). These results are quite different from that of Nistari. This may be due to high temperature during rearing period which has caused much metabolic loss to late age fifth instar larvae. But Nistari being well acclimatized to such adverse climate could have withstood the temperature. This is in support of the finding of Anantharaman *et al.* (1993) who opined that dry matter production per unit food intake is dependent on acclimatization ability of the race of *B.mori*.

KPGB : with methoprene administration : Higher ECD for female pupa has been recorded for day 1 followed by 2 and 5 and lower values for the days 6 and 7. Methoprene significantly lowered the ECD for female pupa in all the starved batches as compared to the untreated control. Further, in all the treatments from the day 1 to 5 the observed values are higher than the predicted values, justifying that food assimilated during facultative feeding period has also been allocated to enhanced ECD to female pupa. Methoprene treatment could not recover the loss due to starvation on the days 5 and 6.

KPGB x P₅ : without methoprene administration : The result obtained is more or less similar to that of pure KPGB. However, due to increased vigour in hybrid the ECD values are higher.

KPGB x P₅ : with methoprene administration : Higher ECD to female pupa has been obtained for the day 5 followed by 4 and 3, and lower for the day 8 followed by 6. Prolongation of obligatory feeding period by the action of methoprene and higher assimilation of food for pupal body

has been recorded in the hybrid. Thus, ECD values has become higher than that of pure KPGB. Loss of metabolism due to starvation on later part of fifth larval life (days 6, 7 and 8) could not be compensated.

5.3 Economic Characters of *B.mori* caused Due to Daywise Starvation During Dry Part of Summer

5.3.1 Nistari : without methoprene administration : The ERR in case of Nistari, being highly acclimatized to adverse conditions of West Bengal, has exhibited better performance after starvation on day 1 resulting in higher ERR. This supports the observation of Benchamin *et al.* (1992). But starvation on other days of the obligatory feeding period such as on day 2 caused high mortality hence the reduced ERR (Janarthan *et al.* 1994). The ERR has been somewhat consistent when starved on other days, which supports the finding of Anantharaman *et al.* (1993) who observed that the survival of silkworm race under stress feeding depended on its acclimatization power to a particular environment. Compared with the values of control sets a reduction in ERR has been observed for all the starved batches supporting the eralier observatioin of Kurisu *et al.* (1975), Nath *et al.* (1990), Samson *et al.* (1981), Hidesh *et al.* (1982), Sumioka *et al.* (1982), Sukumar and Ramalingam (1986), Haniffa *et al.* (1988), Tzenov and Petkov (1993) and Himantharaj (1994).

The final larval weights have been relatively less impaired on starvation days 4,5,6 in comparison to that of other days of starvation indicating that the larvae were able to compensate a good quantity of food deficiency after the resumption of feeding during facultative feeding period. The result is in support of the finding of Mathavan *et al.* (1987). But larvae starved during obligatory feeding period (days 1,2,3) have not been able to compensate the loss under physiological stress due to increased cost of respiration and maintenance (Schroeder, 1976;

Muthukrishnan and Pandian (1987) though assimilation of food has been higher. Low water content in food during this period is compensated by the utilization of metabolic water, which otherwise would have increased larval body weight (Waldbauer, 1968).

Higher impairment of male cocoon shell due to starvation during facultative feeding period (days 5 and 6) supports the observation of Fukuda *et al.* (1963). Horie and Watanabe (1985) have argued that food consumed by fifth instar larvae during facultative feeding period is utilized for synthesis of silk. Kurata (1985) has observed that accumulation of RNA in cells of silk gland is reduced when starvation is imposed from day 3 onward of the fifth instar larvae and recovery from this effect has been delayed even if the larvae were refed. Decrease of cocoon shell weight due to starvation has also been reported earlier by Samson *et al.* (1981), Mathavan *et al.* (1987), Haniffa *et al.* (1988), Nath *et al.* (1990), Tzenov (1993) and Janarthan *et al.* (1994). The female cocoon shell weights have also impaired in batches starved during facultative feeding period where food resources are largely allocated for silk gland growth as suggested by Inagaki and Yamashita (1986).

Daywise starvation has reduced the female pupal weight on days 5 and 6. Muthukrishnan and Pandian (1987) are of the opinion that food quality and availability together with ambient rearing conditions influence largely the female pupal weight in *B.mori*. Dry matter utilization is high in female larvae (Horie and Watanabe, 1983) as a high amount of energy is converted towards egg production (Yamamoto and Gomo, 1976; Dingnong *et al.*, 1991, Anantharaman *et al.*, 1993; Remadevi *et al.*, 1993).

With methoprene administration : The ERR% has been found to be somewhat better after application of methoprene only for the days 1 and

6. ERR% due to starvation on days 3, 4 and 5 has been reduced after administration of methoprene. Trivedy *et al.* (1993) have argued that the prolongation of larval period after methoprene administration reduces the ERR during dry part of summer. Methoprene administration simply prolongs the obligatory feeding period (Calvez, 1981). Thus, the larval batches under different days of starvation with sufficient energy reserve can exhibit better ERR as in case of starvation day 1. Hence, methoprene cannot promote better ERR% in starved batches.

Methoprene has no significant role in the improvement of the starvation related impairment of final larval weight suggesting that methoprene is ineffective particularly during the obligatory feeding period (days 1,2,3).

Methoprene administration has induced higher rate of silk synthesis on all the starvation days except for the day 1. This increase in silk synthesis supports the observations of Akai *et al.* (1973) and Akai (1988). The authors argue that the methoprene helps in retaining the activity of RNA in cells of silk gland much longer than untreated larvae, thus in increasing the volume of silk. But under starved condition perhaps the reserve protein synthesis for cocoon shell gets interlocked due to low food consumption and the JHa caused low digestibility that results in low cocoon shell weight as also recorded by Tojo *et al.* (1981). However, increase of cocoon shell weights in methoprene treated batches has been observed in all the treatments as compared with that of untreated control in all the batches in females and excepting for the starvation days 1 and 6 in the males.

In all the cases methoprene administration resulted in higher pupal weights even in case of starvation days 3, 4, 5 and 6. This suggests that Nistari larvae can be starved during dry part of summer on day 4 and 5 and economic loss can well be compensated by application of methoprene.

5.3.2 KPGB : without methoprene administration : The ERR% in KPGB has been quite good when the larvae were starved on days 1, 6 and 7. The values for rest of the days of starvation have been reduced by increase in mortality. Such observation conforms to the earlier record of Janarthan *et al.* (1994). Paul *et al.* (1993) have identified obligatory feeding stage of KPGB upto the day 3 during favourable season. A better ERR in larval batches starved during the facultative feeding period may be due to the initiation of neurohormonal signal for pupal metamorphosis after the attainment of critical minimum weight as has been interpreted by Wigglesworth (1939) and Friends *et al.* (1965). Increase in mortality in starved batches has earlier been observed by Muthukrishnan and Delvi (1974), Mathavan and Muthukrishnan (1976) and Muthukrishnan *et al.* (1978). Upadhyay and Mishra (1991) also held the idea that though food consumption rate in the bivoltine race is higher at a temperature 30°C and above the ERR is reduced due to higher mortality. A positive relationship between mortality and day of starvation had been drawn by Hidesh *et al.* (1982), Sumioka *et al.* (1982), Nath *et al.* (1990) and Tzenov and Petkov (1993).

Daywise starvation during the obligatory feeding period (days 2 and 3) and also during early facultative feeding period (days 4 and 5) have exhibited higher rate of impairment of larval weight. Reduction of larval weight due to starvation in bivoltine silkworm of *B.mori* has also been reported by Yamamoto and Gomo (1976), Muthukrishnan *et al.* (1978), Sumioka *et al.* (1982), Radhakrishnan *et al.* (1985), Mathavan *et al.* (1987) Radhakrishnan and Delvi (1987), Haniffa *et al.* (1988), Nath *et al.* (1990) and in *Antheraea assama* by Bardoloi and Hazarika (1992).

Starvation during obligatory feeding period (days 1,2,3) and early facultative period (day 4) indicate harmful effect on cocoon shell weights as compared with that of the other days. This can be attributed to the

high metabolic loss at higher ambient temperature by the larvae (Muthukrishnan *et al.*, 1976). It is reported that JH secretion is hindered due to starvation during obligatory feeding period resulting in low cocoon shell quantity (Tojo *et al.*, 1981; Tzenov and Petkov, 1993), and lower synthesis of silk in silk gland when starved on days 3, 4 and such impairment is not recovered even when larvae are re-fed (Kurata, 1985). Paul and Deb (1993) also have observed less impairment of cocoon shell weight in batches starved after day 3. Female cocoon shell weights also are decreased due to starvation.

Relatively less impairment of female pupal weight has occurred when the larvae were starved on days 1, 2, 6 and 7. The result indicates that starvation during the days of late obligatory and early facultative feeding periods (days 3, 4 and 5) has suffered the most due to disturbances in energy allocation for female pupal biomass and cocoon shell production.

With methoprene administration : The ERR% in the starved batches after administration of methoprene has not been found to be improved. This result corroborates the finding of Calvez (1981) and Trivedy *et al.* (1993).

Methoprene has no impact on the recovery of loss in final larval weight due to starvation. Janarthan *et al.* (1994) are of the opinion that proper care in feeding during obligatory feeding period only results in successful bivoltine crop. Results obtained, also indicate that starvation during facultative feeding period has lesser impact on larval weight when compared with that of obligatory period except for the day 1.

There has been an increase in male cocoon shell weight in all the batches except in the larvae starved on the days 6 and 7. Such a result has earlier been observed by Magadum *et al.* (1990) in other bivoltine

silkworm. Prolongation of larval life and increase in food intake have helped the larvae for additional growth of silk gland caused by the action of JHa (Prudhomme and Couble, 1979); Prudhomme ^{et al.} (1985), Akai and Kabayashi (1971), Chang ^{et al.} (1972), Kamada *et al.* (1979), Zan (1979), Kobari and Akai (1978, 1979), Kurata (1981), Shibukawa and Akai (1981), Choudhary *et al.* (1986), Kadono - Okuda *et al.* (1986), Rao *et al.* (1988), Trivedi *et al.* (1993) have applied methoprene to the fifth instar larvae of *B. mori* for increasing the production of silk. But the results indicate that application of methoprene followed by starvation during facultative feeding period (days 6, 7) can not improve silk production in both males and females.

Methoprene has no influence on the larvae for increasing female pupal weights. Therefore, starvation accompanied with methoprene application is of no use for compensating the loss in female pupal weight caused due to starvation.

5.3.3 KPGB x P₅ : without methoprene administration : The ERR% has been reduced in all the starved batches except in day 1 which is mainly due to mortality in dry part of summer. The result corroborates the observation of upadhyay and Mishra (1991) indicating higher susceptibility of this hybrid to temperature along with starvation.

Final larval weight has been impaired in all the days of starvation. Slight improvement, though much lower than the control value, has been observed on the starvation days 6, 7 and 8 due to higher consumption of food by way of prolongation of larval period. Prolonged larval period and low final larval weight in starved batches has also been observed by Basu *et al.* (1992) in bivoltine hybrid NB7 x KPGB.

In the present experimental hybrid starvation from the days 2 and 5 has affected most the male cocoon shell weights. Possibly starvation has

affected silk synthesis during obligatory feeding period (Dingnong *et al.*, 1991) by way of reduction in RNA synthesis (Kurata, 1985). But starvation on any day from 3 to 6 has affected the cocoon shell weight in females. Because, starvation affects the female larvae to a greater extent than the male larvae (Watanabe, 1985).

Starvation during the obligatory feeding period (days 3 and 4) has impaired the female pupal weight as the greater part of the food energy obtained during early part of fifth instar might have been utilized for the egg production. Relatively less affected periods are days 1, 2, 7 and 8.

With methoprene administration : Reduction of ERR% in methoprene administered starved batches compared to those of JH-free batches justifies the inability of methoprene during dry part of summer in the bivoltine hybrid of KPGB x P₅.

Maximum impairment in final larval weight has occurred in case of starvation on the obligatory feeding days 2, 3 and 4. On other days, the loss is relatively less as appeared from the predicted values. Exogenous JHa even has no impact in the recovery of larval weight. It has been argued that JHa can prolong the obligatory feeding period but has no control on the transition from obligatory to the facultative feeding period (Calvez, 1981). But in this case possibly there has been no prolongation of obligatory feeding period because of exogenous JHa and hence, recovery of larval weight could not be possible.

Treatment with methoprene has recovered the starvation-loss only on day 1 for male cocoon shell weight. In general, the JHa has the ability to improve the male cocoon shell weights over the JHa-free starved batches except for the day 2. This may be due to inhibition of JHa secretion in larvae when starved during obligatory feeding period as has been argued

earlier by Hwang Hsu (1979). Except for the starvation days 2 and 7 in rest of the days, the female cocoon shell weights are higher than in JHa-free starved batches. Among the methoprene treated starved larvae only the day 1 batch is able to recover the loss as compared with the untreated control.

No impact of the JHa has been observed on the starved batches for improvement of pupal weights. But in starvation days 5, 6 and 8 this character has shown better result as compared with that of JHa-free batches during dry part of summer. This suggests that in KPGB x P, feeding can be withdrawn on these days but the larvae should be treated with methoprene.

5.4 Reproductive Characters of *B.mori* Caused Due to Daywise Starvation During Dry Part of Summer

5.4.1. Nistari : without methoprene administration : The reproductive characters such as live weight of female pupae, total ovariole length, number of eggs in ovariole and fecundity are very much inter-related. Effect on any one of these characters is reflected on the other. Daywise starvation results in a progressive impairment of these characters with the advanced days of starvation of the larvae. Nistari being well acclimatized to the environment of the plains of West Bengal is able to compensate for the loss caused due to starvation on early days 1, 2, 3 and 4. Number of eggs in ovariole and fecundity has appeared to be well related with female pupal weights. This result supports the earlier observations of Mizuta *et al.* (1969), Govindan *et al.* (1991), Shaheen *et al.* (1992). Daywise starvation has reduced the fecundity in all the batches as compared with that of the control, corroborates the opinions of Engelmann (1970), Samson *et al.* (1981), Sumioka *et al.* (1982), Haniffa *et al.* (1988) Nath *et al.* (1990), Kawaguchi *et al.* (1991) Benchamin *et*

al. (1992) and Tzenov and Petkov (1993).

Nistari : with methoprene administration : Application of methoprene highly improved the concerned reproductive character in the daywise starved batches of Nistari when compared to that of JH-free control. Prolongation of obligatory feeding period due to methoprene has resulted in higher female pupal weight which in turn has increased the fecundity in comparison to JH-free batches except for the starvation days 4 and 5. Parlak *et al.* (1992) have recorded that the growth of ovary commences on day 4 of fifth instar. Hence, it can be assumed that the starvation on days 4 and 5 might have affected normal ovarion growth as has also been emphasized earlier by Englemann (1970), Slansky (1980, 1982). Higher fecundity, increased ovariole length and higher number of eggs in ovariole have earlier been observed in methoprene-treated batches by Magadum *et al.* (1990), Yun and Quan (1991).

Thus, it transpires that the farmers cannot be recommended to starve the fifth instar larvae of Nistari during the dry part of summer. In case of exingiency of food shortage starvation can be practised along with methoprene but not for the days 4 and 5.

5.4.2 KPGB : without methoprene administration : Starvation on days 3, 4 and 5 affects badly the female pupal weights to a higher extent though all the days under starvation have affected as compared to the control values. Direct adverse impact of food deprivation on days 3, 4, 5 on development of the ovary (Parlak *et al.* 1992) resulted in low fecundity of this bivoltine breed during dry summer. This may be due to low accumulation of sufficient nutrient and allocation of substantial nutrient energy for egg production as a result of starvation (Engelmann, 1970; Slansky 1980 a, b; 1982).

With methoprene administration : The result indicates that methoprene during this season has no impact on the recovery or improvement of characters impaired due to starvation. However, methoprene has been able to raise the weight of female pupae and other reproductive characters including fecundity on days 3, 4, 5 though the increase is not significant as compared with the JH-free control. This implies that dry part of summer is a constraint for the action of methoprene as a growth promoter in bivoltine.

Thus, starvation in bivoltine during dry part of summer cannot be recommended particularly during the facultative feeding period of prospective seed crop rearing.

5.4.3 KPGB x P₅ : without methoprene administration: In this hybrid significantly less impairment has occurred on the starvation days 5,6,7 (facultative feeding period). This implies that feeding should never be discontinued during the obligatory feeding period and also on day 4, the first day of facultative feeding period. The weights of female pupae are positively correlated with the fecundity. This hybrid has produced greater number of eggs as compared with that of KPGB in all the days of starvation. Such a superiority of the hybrid for egg production compared with that of pure bivoltine races has earlier been recorded by Gupta *et al.*, (1991).

With methoprene administration : Application of methoprene coupled with starvation on different days do not show any impact on the compensation of impairment over JH-free control. However, significant improvement over JH-free treatments on the starvation days 2,3,4 has been recorded. This suggests that the prolongation of obligatory feeding period might be the reason for the improvement. To a small extent there

is an improvement of the fecundity by methoprene action for the starvation day 4 which has suffered the most. However, the overall result suggests that during the dry part of summer methoprene can improve the situation very poorly in case of bivoltine silkworms.

On the whole, if food availability demands, starvation to the larvae may be imposed only during any of the later days of facultative feeding period such as on days, 5, 6 and 7.

5.4.4 Hatching percent : Hatching percent has been found to be independent of the days of starvation. Starvation has no impact on hatching of eggs in all the races and hybrid. The earlier observation by Paul and Deb (1993) indicates that imposed nutritional stress by way of feeding regulation in parental generation can not alter the original genetically determined character in subsequent generation. Even if the larvae are maintained at low ration level for five consecutive generations can not alter the genetically determined trait such as the hatching percent (Pillai, 1989). Irrespective of the quantity of food supplied pupal-imaginal period of metamorphosis has not been altered (Muthukrishnan and Pandian, 1987). Thus, it can be inferred that hatching percent is dependent on other micro and macro environmental factors and is not affected by regulated starvation or food rationing. This is further evident from the histological observation on spermatogenesis and oogenesis which have been normal in the present observation. Egg vigour has also remained unaffected though Band (1973) is of the opinion that starvation has impact on egg weight as it decreases starch content in egg. Methoprene when applied to the starved larvae has no impact on oogenesis and spermatogenesis as studied in the histological preparations. Magadum and Magadum (1993) are of the opinion that methoprene has no adverse effect on the quality of eggs of *B.mori*. Thus, it can be inferred that hatching is an independent genetic trait, controlled by embryological process combined with environment.

5.5 Utilization of Nitrogen by the Fifth Instar Larvae Due to Daywise Starvation During Dry Part of Summer.

5.5.1. Ingestion

Nistari : Ingestion of nitrogen during dry part of summer has been lower than in wet part of summer due to low quantity of food consumption though the quantity of leaf nitrogen has been higher (3.64%). The low level of water in mulberry leaves during dry season affects nutritional efficiency and growth of larvae (Scriber, 1977; Reese and Beck, 1978). The ingestion of nitrogen has been almost the same in all the starvation days except the day 1. This implies that the nitrogen ingestion is not impaired considerably due to starvation. Treatment with methoprene has improved consumption of food significantly in all the starvation days thus, the consumption of nitrogen has also been improved considerably.

KPGB : Nitrogen ingestion has been lowered in all the starvation days very significantly. The impairment has been maximum in the starvation days 3, 4 and 5. Thus, the starvation stress affects nitrogen ingestion worst during both obligatory and facultative feeding days. This result is quite consistent with the earlier observation of Janarthan *et al.* (1994). Treatment with methoprene in starved batches has no good impact on the rescue of the impairments in any of the starved days. However, due to increased food consumption relatively higher amount of nitrogen has been consumed. the total quantity of nitrogen consumed has been higher in dry season when compared with that of wet part of summer.

KPGB x P_s : Except for the starvation days 1 and 2 the consumption of nitrogen has been remarkably impaired. The hybrid has consumed a higher quantity of nitrogen inspite of low water levels in the leaves. Treatment

with methoprene though improved the nitrogen ingestion than in the control in all the starved days the improvement has been very high in days 1, 6, 7 and 8.

5.5.2. Excretion :

Nistari : A higher quantity of nitrogen has been eliminated through excreta during the dry part of summer. Because water imposed a limitation in nitrogen digestion and utilization. Starvation together with low water level of the leaves have reduced the efficiency of conversion of ingested nitrogen to body tissue (Schroeder, 1986). Parallely with the higher quantity of leaf consumption following methoprene application, much higher quantity of nitrogen has been eliminated in all the starved batches.

KPGB : In general, the starvation reduces the excretion of nitrogen, particularly in case of days 3 to 5. This is also true in terms of percentage of nitrogen excretion. It appears that under moisture stressed condition lower level of food consumption due to starvation is compensated by restricting nitrogen elimination. Thus, bivoltine race expresses superiority to the polyvoltine in the utilization of dietary nitrogen. Treatment with methoprene though has increased the quantity of nitrogen excretion except in the days 3 to 4 percentage of nitrogen elimination is significantly lower except for the starvation day 2. In spite of low leaf water during dry part of summer and starvation stress the bivoltine race is genetically competent for nitrogen utilization.

KPGB x P₅ : In this hybrid of bivoltine there is a tendency to restrict nitrogen elimination in all the starvation days except in the days 1 and 2 where nitrogen elimination is higher than the control values. But following methoprene application the elimination of nitrogen has been reduced in

all the starvation days. This reveals that this hybrid has the property of higher level of nitrogen utilization even under stressed condition.

5.5.3. Digestion and digestibility percentage :

Nistari : Digestion of nitrogen requires high level of dietary water. The starvation has been found to impair the digestibility percent, in all the batches, due to low water level in the mulberry leaves as emphasized by Scriber (1977) and Reese and Beck (1978). The worst affected day in this respect is the starvation day 6. Methoprene has no ability to improve the digestibility of nitrogen in starved batches. However, during the facultative feeding period the digestibility is relatively better.

KPGB : Better digestion of nitrogen has been observed when the larvae were starved during the late obligatory and the entire facultative feeding periods. The values are relatively poor for the starvation days 1 and 2 indicating that the starvation during the early obligatory feeding period restricts nitrogen digestibility. Digestibility percent has been decreased significantly when the starved batches were treated with methoprene.

KPGB x P₅ : The result indicates that the starvation initially reduces nitrogen digestion on days 1 and 2 followed by significantly higher rates on the days 3, 4 and 5. Again, the rate has been declined on days 6 and 7. Thus, starvation stress has no impact on the nitrogen digestibility on days 1, 2, 6 and 7, i.e. during early obligatory and late facultative feeding periods. In comparison to untreated control, methoprene reduces the nitrogen digestibility. However, the values are higher on starvation days 4 and 5.

5.5.4. Distribution of nitrogen

5.5.4.1 Larval body :

Nistari : Due to low water level in food the expenditure of energy for nutritional efficiency and growth is affected in silkworm (Scriber, 1977; Reese and Beck, 1978). Again, low water also reduces nitrogen assimilation. Thus, during dry part of summer only 30 mg of nitrogen has been accumulated in larval body. Schroeder (1986) has emphasized that the efficiency of conversion of digested nitrogen to larval tissue is reduced by starvation. In case of Nistari the food deprivation on early two days of obligatory feeding period and on days 5 or 6 of facultative feeding period has reduced the flow of nitrogen to larval body. Methoprene though has improved the nitrogen content of larval body, it has no ability to restore the level of nitrogen contents of control larvae.

KPGB : The nitrogen content in the body of control larvae has been 78 mg. Food deprivation during the days 3, 4 and 5 has lowered the nitrogen contents of the larval body. This may be due to high metabolic loss caused by temperature and low water level in food. Treatment with methoprene has been ineffective to recover from impairment but the accumulation of nitrogen has increased as compared with that of the untreated batch.

- **KPGB x P₅ :** The nitrogen content of larval body is about 56 mg. Food deprivation during the late obligatory and early facultative feeding days has reduced the nitrogen content. Treatment with methoprene has improved the body nitrogen in the larvae on days 1 and 8. But the improved value is still lower than the nitrogen content of control larvae. In case of other starvation days there is no significant improvement.

5.5.4.2. Male Cocoon shell :

Nistari : Food deprivation during intermediate days of larval life has resulted in better nitrogen share for male cocoon shell. This indicates better utilization of nitrogen for male cocoon shell from the food consumed during early part of obligatory and later part of facultative feeding periods. Compared with the nitrogen contents of untreated control males methoprene has been able to improve the nitrogen levels in the shells resulted from the starved larvae of later days of facultative feeding period. In rest of the starvation days the values also have improved but could not cross the control value.

KPGB x P₅ : The share of nitrogen for male cocoon shell has been lowered in the batches starved during the days covering late obligatory and early facultative feeding periods. In case of other days the share is also low but of little difference from the value of control. Methoprene could recover the affected batches from impairment. Even in no cases the share has exceeded the value of untreated control individuals.

5.5.4.3. Female Cocoon shell :

Nistari : Starvation on any day has reduced variably the share of nitrogen for the female cocoon shells. The lower range of variability in the share on different days of starvation may be due to differential but rational allocation of larval nitrogen for reproduction and cocoon shell. Treatment with methoprene has rescued the female's share for cocoon shell in all the days of facultative feeding period. This might have happened due to prolongation of feeding period by the action of methoprene.

KPGB : Starvation has lowered the share of nitrogen for female cocoon

shell significantly in all the days with much intense effect on the days 3, 4 and 5 which cover both obligatory and facultative feeding periods. This supports partly the finding of Janarthan *et al.* (1994) who regard that the obligatory feeding period is very crucial in the fifth instar stage of bivoltine silkworm. In case of the worst affected days 3, 4 and 5 even the methoprene has no good contribution to the recovery of female shell share. In all other days of starvation, methoprene has been able to raise the share of nitrogen in the female cocoon shell as compared with the share for untreated control.

KPGB x P₅ : Food deprivation during the days of obligatory and facultative feeding has resulted in lowering of nitrogen content in the female shell, particularly in cases of the days 3-6. Treatment with methoprene has no impact on the recovery of nitrogen share when compared with the result of JHa-free control.

5.5.4.4. Female pupa :

Nistari - Starvation in general has reduced the nitrogen content in female pupal body almost to a half in most of the cases or even of much lower share. This indicates that due to division in the allocation of nitrogen for shell and pupal body the share for the pupa has been reduced considerably. Treatment with methoprene though has increased the share of nitrogen for pupa, the recovery to the level of untreated control has been possible only in case of the starvation days 5 and 6.

KPGB : Starvation has restricted the pupal share of nitrogen only for the days 3 and 4 which constitute later half of obligatory feeding period. Proper feeding is essential up to the day 4 of the fifth stage larvae of *B.mori* for the optimum growth of larvae (Parlak, 1992). Deficiency during this period

certainly affects the biochemical profile of pupal body. This may explain the lower nitrogen share for pupal body from the larvae. Methoprene has no noticeable impact in raising the pupal share of nitrogen over the control value.

KPGB x P₅ : Food deprivation on all the days has impaired the proper flow of nitrogen from larval body to female pupa in this hybrid. Even methoprene has not been able to bring up the nitrogen content to the level of untreated control individuals. This implies that this bivoltine hybrid is too much sensitive to starvation for nitrogen utilization and sharing.

5.6 Consumption and Assimilation of Food During Wet Part of Summer Due to Starvation on Different Days

5.6.1 Consumption

Nistari without methoprene administration : During the wet part of summer both temperature and the humidity remain high. The consumption of food by the control larvae is quite high (1.935 g/larva), much higher than during the dry part (1.271 g/larva). The higher quantity of leaf consumption by the fifth instar larvae of *B.mori* at high temperature and high humidity has also been reported by Mishra and Upadhyay (1992) and Paul *et al.* (1992). The larval duration is also shortened, the life span is only 5.5 days. Paul *et al.* (1992) reported an accelerated larval growth due to high dietary water. In the present result a prolongation of larval duration by about half a day over that of control larvae has been recorded for the starvation days 2, 3 and 6. In the remaining days of starvation the duration has been at par with the control larval duration. Extension of larval period due to starvation on days 2 and 3, being the crucial obligatory feeding days, is a physiological adaptation for food

compensation. But the extension due to starvation on day 6, the later facultative feeding day, is due to a little delay in releasing the signal from the neuro endocrine system for pupation. Benchamin *et al.* (1992) have opined that the multivoltine larvae are more resistant to first day starvation after moult. The wet part of summer being the most adverse season for silkworm rearing (Paul *et al.* 1992) the larvae were affected by daywise starvation as is revealed from the regression equation. Likewise in dry part of summer the predicted values indicate that the worst affected batch is the day 2.

With methoprene administration : Treatment with methoprene has increased significantly the consumption of food in all the batches including the control. However, starvation on days 2-6 has reduced larval duration by about half a day. JH-free larvae starved on day 2 has suffered most in food utilization. When treated with methoprene, the larvae have consumed the maximum quantity of food due to prolongation of obligatory feeding period (Calvez, 1981), thus have compensated the consumption loss to a maximum extent. The lowest consumption has been recorded for the day 6. Regression equation reveals a negative trend of food consumption (Y) against the days of starvation (X) suggesting that the impairment caused by starvation could not be recovered by the action of methoprene. Predicted values show a good fit except for the day 2 where observed values are higher.

KPGB : without methoprene administration: The consumption of food has been less than that in the dry part of summer. In spite of longer larval life of bivoltine, higher quantity of consumption under high temperature and high humidity (Mishra and Upadhyay, 1992) and excess leaf moisture promoting larval growth during wet part of summer (Paul *et al.*, 1992), the rearing performance of the bivoltines is very poor because

of larval diseases. The food consumption in silkworms is a strain-specific character and it totally depends on ambient environmental factor (Horie and Watanabe, 1982; Periswami *et al.* 1984; Vijay, 1985; Remadevi *et al.* 1991). Furthermore, both edibility and assimilability of leaves in silkworm is affected by high water content. Regression equation indicates a negative trend towards consumption against the days of starvation suggesting an impairment caused by starvation. Predicted values are better than the observed values for the days 2-6. This indicates that the consumption due to starvation cannot be compensated after restoration of feeding.

With methoprene administration : Food consumption in methoprene treated control larvae has been increased by more than 1.5 g/larva and the fifth larval duration has been extended by 1 day. Such an extension is in agreement with the observation of Rao *et al.*(1988). Methoprene has no significant influence on the compensation of consumption loss due to starvation.

KPGB x P₅ :without methoprene administration : Though it is a bivoltine hybrid, the total consumption per control larva is only 3.049 gm which is much lower than that of KPGB. However, the larval duration is the same i.e. 9 days in control. Starvation has prolonged this duration by one day only except for the day 8. In all the starvation days the food consumption is less than the control value. Regression equation demands for a negative trend for food consumption. But from the predicted values it can be inferred that larvae of the days 1,2,3, 4 and 6 could not compensate for the consumption loss. But the larvae of other days of starvation could compensate the loss partially.

With methoprene administration : Here too, the consumption of food

has been increased following methoprene administration. Regression equation expresses a slight positive trend proving some improvement over impairment caused in consumption. The highest level of recovery of consumption loss was on the day 8. Larval duration in KPGB x P₅ has been prolonged by 2 days over the control, as against only one day in case of KPGB. Long larval period at an environment of high temperature and high humidity is not desirable for the rearing of this bivoltine hybrid too, as the crop suffers badly from the diseases particularly at the fifth larval stage.

5.6.2 Assimilation

Nistari : without methoprene administration : The total assimilation of food in every control larva has been 1.122 gm. Assimilation of food has been found to vary significantly among the starvation days, the lowest being on day 6. The compensation in the loss of assimilation has occurred for all the starvation days except for the day 6. The maximum level of compensation has been achieved for day 5 followed by 4. Higher water content in food has facilitated a better digestion of crude fibre and dry matters during the season resulting in assimilation of food higher than that of the dry part of summer. Legay (1957) demonstrated that high dietary water together with high ambient temperature and humidity increased assimilation of food. This is also true for Nistari during wet part of summer (Paul *et al.*, 1992). Regression equation indicates a negative trend between assimilation (Y) and the day of starvation (X). A comparison of observed and predicted values justifies an impairment of food assimilation due to starvation except for the days 4 and 5. The result corroborates the earlier observation of Mathavan *et al.* (1987).

With methoprene administration : The total assimilation in methoprene treated control has been higher than that of untreated control. As a consequence, a higher quantity of assimilatory loss has been compensated in all the starved but methoprene-treated batches except for the day 6. This compensation during wet part of summer may be due to an increase in consumption rate after restoration of feeding with leaves having higher moisture content (Paul *et al.*, 1992). High temperature and high humidity have also facilitated better digestibility of food in the larvae of starvation days 1 to 5. But starvation on day 6, the later part of facultative feeding period, perhaps has switched in the larvae a signal to enter into metamorphosis resulting in low level of assimilation.

KPGB: without methoprene administration : Likewise in Nistari, the impairment of assimilation has also occurred in KPGB in all the starved batches. The regression equation indicates a very little negative trend. A non-significant correlation has resulted between the days of starvation and assimilation. Recovery from assimilation loss has taken place for the starvation days 1, 6, 7 and 8. Though during the dry part of summer assimilation loss could not be compensated, during the wet part of summer compensation of loss has occurred in some of the starvation days. The higher water content in the mulberry leaves might have facilitated higher digestibility as the low dietary water is a constraint for digestibility (Waldbauer, 1964). Furthermore, the high ambient temperature and humidity also help in higher consumption and assimilation of food in the bivoltine race (Legay, 1958; Mishra and Upadhyay, 1992).

With methoprene administration : The JHa has been able to compensate the assimilatory loss for the starvation day 1 by way of prolongation of obligatory feeding period (Calvez, 1981). However, recovery from assimilatory loss has also occurred to some extent in cases

of the starvation days 5, 6, 7 and 8 which suggests that this JHa can also influence during facultative feeding period.

KPGB x P₅ : without methoprene administration : Hybrids of bivoltine generally perform better than pure bivoltine (Hamano and Toshihiko, 1989). But compared with the performance of KPGB, the KPGB x P₅ hybrid has performed poor after daywise starvation, the assimilation of food has been lower during wet part of summer. Regression equation shows a negative trend. The observed values provide a good fit against the predicted values, suggesting an impairment in assimilation of food. In none of the cases the loss due to starvation could be recovered.

With methoprene administration : The result reveals that except for the starvation days 7 and 8 none of the other batches has been able to compensate for the loss in assimilation. Regression equation shows a negative trend. The observed values provide a good fit against the predicted values, suggesting that the assimilation loss could not be rescued even after the treatment with methoprene except for the days 7 and 8 which span the extended facultative feeding period.

5.6.3 Absolute consumption rate

Nistari : without methoprene administration : The result reveals that the highest absolute consumption rate (ACR) has been obtained for starvation day 5 and the lowest for the day 2. The ACR is quite high in this season than the dry part of summer. Except for the days 4 and 5, in case of other starvation days the observed values are lower than the predicted values, revealing an impairment of ACR caused due to starvation.

With methoprene administration : The JHa when applied to the larvae of starvation days 6 though induced the prolongation of facultative feeding period, there has been no recovery of consumption loss. Because of attainment of critical weight at this age of larvae pupal programming has been signaled (Calvez, 1981, Sakurai, 1984) This has caused the lowering of ACR. Except for the treatment of the JHa on day 2, the observed values are at par or less than that of the predicted values. This confirms the finding of Paul *et al.* (1992) who held that a high water content in food does not favour higher food consumption as the ambient condition is not conducive to silkworm rearing.

KPGB : without methoprene administration : The ACR has been the highest for the starvation day 1 followed by 8 and is the lowest on the day 4. Except for the day 8 the observed values are lower than the predicted values. This confirms that wet part of summer is not conducive to the rearing of bivoltine silkworms.

- o **With methoprene administration :** The ACR is the highest for the starvation day 1 followed by the day 4 and is the lowest for the day 6. The JHa when applied to the larvae of starvation day 4, has helped in the recovery of the loss of ACR which has suffered the most without the JHa. Such recovery has been possible due to prolongation of obligatory feeding period.

KPGB x P₅ : without methoprene administration : Except for the starvation day 5, the larvae of other starvation days were not able to compensate the loss of ACR. This reveals that, irrespective of obligatory and facultative feeding periods, starvation has caused impairment of ACR in this bivoltine hybrid during the wet part of summer.

With methoprene administration : The ACR has suffered the most even after the administration of the juvenoid on the day 4 larvae followed by day 2. In none of the cases the loss in ACR has been recovered. On the other hand, the loss during the facultative feeding period, days 7 and 8, has been recovered by way of prolongation of the facultative feeding period following the methoprene application. This result is in conformity to the finding of Janarthan *et al.* (1994).

5.6.4 Absolute growth rate

Nistari : without methoprene administration : The absolute growth rate (AGR) has been always lower in all the starved batches than that of the control larvae. However, in case of days 1 the rate has been almost at par with the control because of almost no impairment of food consumption. The highly resistant multivoltine Nistari race suits better to overcome starvation on first day (Benjamin *et al.*, 1992). High dietary water of about 76.6% together with high ambient temperature and relative humidity during this season might also have favoured the growth (Paul *et al.*, 1992) which has led to less impairment of AGR inspite of starvation stress. This is why the starvation on days 4 and 5 (during facultative feeding period) has exhibited better recovery in the AGR. This result supports the arguments of Muthukrishnan and Delvi (1974), Mathavan and Muthukrishnan (1976), Muthukrishnan *et al.* (1978), Grabstein and Scriber (1982) and Muthukrishnan and Pandian (1984) who are of the opinion that the increased feeding rate on restoralism of food by larvae starved during facultative feeding period is followed by higher growth rate.

With methoprene administration : Methoprene has lowered the AGR in all the batches inclusive of the control though the food consumption

and ACR have been increased significantly. However, in comparison to the control values the AGR has been increased significantly in case of the days 3 and 6, the values have been at par with the control AGR and finally in case of days 1 and 4 the AGR has been lowered from the control value. The lowering of the AGR due to methoprene has also been recorded by Kurata (1981). The lowering of AGR than the methoprene-treated control for the starvation days 1 and 4 is associated with high metabolic loss due to respiration and high maintenance cost.

KPGB : without methoprene administration : In this bivoltine silkworms the AGR has been lowered considerably in all the days of starvation. A comparison of the observed and predicted values reveals that except for the day 4, the observed values are higher. This indicates that in spite of starvation stress the AGR is quite good during the wet part of summer. However, in comparison to the control value the low AGR in all the batches, particularly in case of the days 4 and 6, has been due to high maintenance cost (Schroeder, 1976).

With methoprene administration : Likewise in the multivoltine race, the AGR has been lowered by methoprene in all the batches inclusive of the control. The worst affected batches are of the starvation days 3 to 6. In all the days the observed values have been lower than the predicted values, indicating an impairment of AGR due to methoprene. The result suggests that the food utilization efficiencies of different races of silkworm and their genetic property determine the AGR (Yamamoto and Fujimaki, 1982). The ambient conditions are also added factors (Remadevi *et al.*, 1991). On the whole, the loss in AGR due to starvation cannot be recovered by methoprene administration.

KPGB x P₅ : without methoprene administration : The starvation of

larvae of this bivoltine hybrid on all the days has impaired the AGR considerably. Except for the days 2, 3 and 8, the observed values are either higher or at par with the predicted values. Though there is no recovery of the loss in AGR due to starvation, a better AGR has been expressed in the starvation days 1, 3 and 4. This is perhaps due to hybrid vigour of KPGB x P₅. Among all the treatments the larvae of only the day 5 have been able to recover a very low amount of consumption loss and hence, the AGR. These results are quite an exceptional from the results obtained for Nistari and pure KPGB. The better growth rate in KPGB x P₅ may also be due to excess dietary water in food. But starvation during facultative feeding period has retarded the AGR of the hybrid because the larvae could not recover from the heavy consumption loss, as the highest quantity of food was consumed by the control larvae on day 7. Starvation during late facultative feeding period has induced the neurohormonal signal for pupation resulting in low consumption of food and hence, the low AGR. Maintenance costs has been recorded to be lower in treatments on days 1, 3 and 4 than that of the days 7 and 8 which also resulted in better AGR in batches starved during obligatory feeding period.

With methoprene administration : In general, the juvenoid has lowered the AGR of this bivoltine hybrid. The observed values have been lower than the predicted values in case of starvation days 2 to 6 indicating no recovery of the impairment in AGR. This has occurred due to higher maintenance cost during these days of larvae. On the whole, this juvenoid has been able to improve the AGR in case of the days 7 and 8 only.

5.7 Efficiency of Conversion of Digested Food (ECD) During Wet Part of Summer Due to Starvation on Different days.

5.7.1 Larval body

Nistari : without methoprene administration: The ECD to larval body has been higher than the control in case of starvation day 6. In case of day 2 the value has been almost at par with the control. For the remaining days the ECD values have been lowered. The differences in the ECD values are related to the extent of compensation of assimilation loss due to starvation. During wet part of summer the high rearing room temperature and relative humidity favour better growth of larvae. Furthermore, the high water content in leaves increases consumption and assimilation rate due to better digestion (Paul *et al.*, 1992) Energy in larvae is perhaps saved by the reduction of metabolic loss due to non conversion of reserve carbohydrate for the production of metabolic water as has been observed during dry part of summer (Waldbauer, 1968). Nistari, being very much acclimatized to this adverse summer in West Bengal, has a very short larval duration which is one of the characters of acclimatization of race (Anantharaman *et al.*, 1993). So, starvation stress on any day during facultative period releases pupation signal. Because, during this feeding period food is consumed to the ration level for only maintenance. The net result is low ECD value for larval body. A critical ration level of food is essential for maintenance in insects for triggering the secretion of moulting hormone (Wigglesworth, 1939; Friend *et al.*, 1965). However, Kadono-Okuda *et al.* (1986) are of the opinion that the endocrine system directing pupal metamorphosis is determined prior to last larval ecdysis in *B.mori*.

With methoprene administration : Except for the larvae of starvation days 4 and 6, this juvenoid has lowered the ECD to larval biomass in all other days. Except for the day 6 the observed values are always lower than the predicted values showing a greater impairment due to the action of the juvenoid. This conforms to the observation of Gaaboub *et al.* (1985).

- A reduced digestibility of food has resulted in lower ECD values (Remadevi *et al.*, 1992). Thus, the result suggests that this juvenoid has no significant role in the improvement of ECD values impaired as a result of starvation.

KPGB : Without methoprene administration : This bivoltine race is very much sensitive to starvation stress. The ECD value to larval body has been lowered considerably in all the starvation days. The worst effect has occurred on all the days of obligatory and early half of the facultative feeding period. The observed values have been much lower than the predicted values in all the starvation days except the days 2,7 and 8. Due to adverse climatic condition for bivoltine rearing during this period the starved larvae have suffered from high metabolic loss due to respiration and maintenance cost. The assimilation of food after restoration of feeding in these batches could not be improved due to high maintenance cost specially in larvae starved on days 3 to 6. Starvation on day 1 has resulted in higher metabolic loss due to less energy reserve by larvae required to withstand the feeding stress. But on day 2 the larvae could have accumulated energy on day 1, could withstand the starvation stress a little more and have performed a better ECD for larval body. The result also reveals that starvation even during facultative feeding period lowers the ECD values considerably. The ECD values of starved larvae during wet part of summer are always much lower than those of during the dry part. This indicates that the efficiency of conversion of digested food to larval biomass is affected much during this adverse season.

With methoprene : The ECD values to larval body has been lowered further than those during dry part of summer. On the whole, this juvenoid deteriorates the ECD from the values obtained simply after starvation. Thus, methoprene has no use for improvement of loss due to starvation.

KPGB x P₅ : without methoprene administration : With reference to the control the ECD values are much higher in all the batches except in the larvae starved on day 8. The observed values for the days 1, 3, 4, 5 and 6 are higher than the predicted values. Performance of KPGB x P₅ has been found to be comparatively better than pure KPGB during this season. This is perhaps due to hybrid vigour in KPGB x P₅ (Narasimahanna, 1985). Hamano and Toshihiko (1989) have recorded that the larvae of hybrid races grow well than pure lines. The present results reveals significantly high ECD values in feeding regulated batches as compared to the control. Increased ECD in feeding regulated larvae of *B.mori* has also been reported by Sumioka *et al.* (1982), Nath *et al.* (1990) and Tzenov (1993). The authors have explained this phenomenon as a physiological adaptation under stress feeding condition. Furthermore, the high water content in food during this period has promoted larval growth (Paul *et al.*, 1992) than during the dry part of summer. Because high dietary water facilitates digestion (Waldbauer, 1968). Thus, the hybrid of KPGB x P₅ has effectively assimilated food without much metabolic loss due to respiration and maintenance cost except for the starvation days 2, 7 and 8, and this hybrid has shown better ECD than pure KPGB during the season.

With methoprene administration: Methoprene has lowered the values of ECD to larval body considerably even than the untreated control larvae. Thus, methoprene has no importance in the improvement of loss due to starvation.

5.7.2. Male Cocoon shell

Nistari : without methoprene administration: The impairment of ECD to male cocoon shell has been lowered significantly in cases of starvation days 2, 4 and 5, the lowest being on day 2. Significantly the value has been raised over the control in starvation day 6, even higher than the predicted value. As a high percentage of food energy has been utilized by *B.mori* larvae for the cocoon shell, the rate of assimilation of food plays an important role. The ECD values has also been higher on days 1 and 3 than the predicted values. In contrast to the observation of Fukuda (1963), even in the facultative feeding days (except the day 6) starvation has reduced the ECD to male cocoon shell. Due to shorter duration of fifth larval life span during wet part of summer than during the dry part, the larvae starved on day 5 has been deprived of maximum quantity of food ingestion and assimilation which perhaps has resulted in the low ECD of cocoon shell. Food consumed during the latter days of fifth instar is utilised effectively by *B.mori* larvae for the production of silk protein (Fukuda *et al.*, 1963).

With methoprene administration: A higher ECD to male cocoon shell has been recorded in the larvae of starvation day 3, the ECD has been lowered greatly on the days 1 and 4. In all other days, there is no significant difference of the ECD values from the control. Thus, the juvenoid has improved the impairment of ECD to male cocoon shell selectively for some of the days of starvation.

KPGB : without methoprene administration : The ECD values have been higher than control in the larvae starved on days 2 and 5 but lower than the control on days 1 and 7. The observed values have been higher than the predicted values on days 1, 3, 5 and 6. Starvation on day 1 makes the larvae too much exhaustive of reserve resources after a moult (Calvez,

1981) that results in lower ECD to male cocoon shell. Again, starvation on day 6 and 7 impairs silk protein synthesis to the highest extent. Hence, the ECD on these two days are very poor (Fukuda *et al.*, 1963). The bivoltine silkworms are more susceptible to starvation stress than the multivoltines (Benjamin *et al.*, 1992). Starvation on day 4, the beginning of facultative period, has caused a lower synthesis of silk protein due to high metabolic loss and slow rate of accumulation of RNA in silk gland (Kurata, 1985).

With methoprene administration: In comparison to the values of untreated control the ECD to male cocoon shells has been reduced by methoprene in all the starved batches and control larvae. Methoprene when applied to the larva starved on different days has maintained the ECD values to cocoon shell in most the cases at par with the JHa-treated control batch. In case of the days 6 and 8 the values have been much higher than that of the control. The lowering of ECD values due to methoprene supports the record of Gaaboub *et al.* (1985). The notable point is that in spite of low ECD to larval body during the days of facultative feeding period, the ECD values for male cocoon shell are quite good, particularly in the larvae starved on the days 6 and 8.

KPGB x P₅ : without methoprene administration: In this bivoltine hybrid the starvation has promoted the ECD to male cocoon shell to a considerable extent in all the feeding days particularly during the obligatory feeding days. The positive response of this hybrid to the starvation stress appears to be a genetic quality. Even, having a low ECD to larval body in batch starved on day 2 the ECD to male cocoon shell has increased.

With methoprene administration: In contrast to the JHa-free result, methoprene has reduced the ECD to male cocoon shell except in cases of

the larvae starved on the days 4 and 5. This negative effect is more during the obligatory feeding days. The methoprene-induced prolongation of larval duration during obligatory period, impairs the ECD to male cocoon shell by increasing respiratory and maintenance costs. Furthermore, during the adverse rearing season the JHa decreases ECD to cocoon shell (Kurata, 1981). Compared to the pure KPGB race superiority of the hybrid has also been recorded by Horie *et al.* (1976). However, the JHa is to be avoided for the bivoltines.

5.7.3 Female Cocoon shell

Nistari : without methoprene administration : The basic pattern of the impact of starvation on the ECD to female cocoon shell is similar to that of the male cocoon shell. With respect to the individual starvation days the result shows only minor differences.

With methoprene administration : The starvation combined with the JHa has no considerable differences in the results of ECD to cocoon shells between the two sexes. The ECD is only slightly improved in cases of the starvation days during the facultative feeding period.

KPGB : without methoprene : The ECD to cocoon shell shows sexual dimorphism, it is higher in the females. The impact of starvation is somewhat similar in the two sexes. Interestingly, the ECD to female cocoon shell is significantly higher in the starvation batches of days 2 and 8. In cases of other days (except day 5) the values are lower. Except for the days 2, 4 and 7 skipping of feeding for a day may be recommended if exigency demands, but at a little sacrifice of shell value.

With methoprene administration : The pattern of action of the JHa on

the ECD to female cocoon shell is almost similar to that in case of male. Except for the days 2, 6 and 7 the JHa lowers the ECD values from the control. However, in none of the days the application of JHa is suitable.

KPGB x P₅ : without methoprene administration: There is no sex-specific differences in the pattern of ECD values for cocoon shell under day-wise starvation stress. Even the values are not higher in the females than in the males as in the case of pure KPGB. Interestingly, the value has been boosted up to a very high level in the batch starved on days 4 and 5, especially on day 5.

With methoprene administration : The impact of methoprene is the same as in the male batches. The poor result due to the JHa negates its application to the starved larvae during the wet part of summer.

5.7.4 Female Pupa

Nistari : without methoprene administration : Compared with that of the control, higher ECD values for female pupal body has been obtained in cases of the starvation days 1, 3 and 6. The values has been significantly lower in the starvation days 2, 4 and 5 than that of the control. The female larval dietary energy is used for both egg and silk productions. The differential ECD values on different days of starvation have resulted from differential effect of starvation for the allocation of food for different purposes.

- **With methoprene administration :** The ECD to female pupal body has always been boosted up due to the JHa. But in all the days of starvation the values are significantly lower than that of the control. Except for the day 1, in all other days the results are higher than the untreated control.

However, the observed values are lower than the predicted values except in case of day 6, indicating that inspite of prolongation of larval duration due to the JHa, the ECD has been lowered.

KPGB : without methoprene administration: In all the starvation days the ECD to female pupal body has been lower than the control. The observed values have been higher than the predicted values. In general, ECD to female pupal body has been better than the ECD values for female cocoon shell. This is in conformity to the opinioin of Horie and Watanabe (1980) that the dry matter utilization for egg production than silk gland is more in female larvae of *B. mori*. The high metabolic loss due to respiration and maintenance cost on days 1, 3 and 4 may be the reason for lowering the ECD values for female pupa.

With methoprene administration : The ECD to female pupal body in this bivoltine race has a set back due to JHa during the adverse wet part of summer (Paul *et al.*, 1992). However, in all the starvation days the observed values are higher than the predicted values.

KPGB x P_g : without methoprene administration : In all the days of starvation the ECD to female pupal body has always been higher than in the control. This implies that starvation stress has boosted up this allocation of food energy in this bivoltine hybrid, possibly for higher allocation for egg production. Interestingly, the highest ECD value has been recorded on the starvation day 5.

With methoprene administration: In this bivoltine hybrid the JHa has lowered the ECD to female pupal body only slightly than in case of untreated control batch. But in all the starvation days the values have been considerably lower than the control and much lower than the

untreated starved batches. Thus, this juvenoid is of no use in boosting up the starvation loss, rather this has a detrimental effect in this bivoltine hybrid.

5.8 Economic Characters of *B. mori* Caused Due to Daywise Starvation During Wet Part of Summer.

5.8.1 Nistari : without methoprene administration : The ERR% has been relatively lower during wet part of summer than in the dry part of summer in all the daywise starved batches including control. This reveals that mortality percent in fifth instar is higher during this season. Paul *et al.* (1992) have opined that high water level in leaf is one of the reasons for such mortality in all the races of *B. mori*. Kirusu *et al.* (1975) have observed multiplication of enterococcus in midgut lumen during this season acquired along with mulberry leaves into the body of larvae. They are also of the opinion that such multiplication is more in starved larvae than in the healthier one. Sukumar and Ramalingam (1986) have observed high bacterial population on healthy mulberry leaves during wet part of summer. Thus, inspite of well-acclimatization in this environment, Nistari has suffered from impairment of ERR% in all the days of starvation except for the days 3, 4 and 6. This result suggests that optimum feeding during obligatory period may help to develop resistance against the bacterial diseases.

The final larval weight has been impaired in all the starvation days except the days 1 and 6 indicating less assimilation of food on other days of starvation. In spite of high water level in leaves the assimilation rate in the body of silkworm has been reduced due to starvation during the short lifespan of larvae. However, the gain in larval weight does not differ much with that in the dry part of summer in all the batches of starvation.

Cocoon weights are higher during the wet part of summer than during the dry part indicating better utilization of assimilated food under high water level. But starvation has impaired this character on all the days except for starvation days 1 and 3 in males and days 1, 3 and 4 in females. This reveals that food consumed during the facultative period has largely been utilized for silk synthesis (Fukuda, 1963). However, the result obtained has been significantly lower on all the starvation days when compared with that of the control batch.

Starvation during facultative feeding period has highly impaired the female pupal weight in all the starved batches likewise during dry part of summer. This indicates that energy conserved by female is proportionately distributed both for cocoon shell and pupal biomass for egg production.

Thus, starvation during the days 4-6 has higher negative effect for economic parameters in Nistari. Hence, if required, feeding may be avoided on day 3 only during this season.

With methoprene administration : Except for the starvation days 2 and 3 in rest of the days starvation followed by treatment with methoprene, the ERR % has not been significantly impaired when compared to those of JH-free starved days. However, methoprene can not improve the ERR % when compared to those with JH-free control batch. This is due to JHa-induced prolongation of larval period. Furthermore, prolongation of larval obligatory feeding period followed by low digestibility perhaps have increased mortality and reduced ERR% on days 2 and 3.

The final larval weight has been significantly increased over the JH-free control batch on the starvation days 3 and 6. This is perhaps due to better compensation of assimilatory loss during the prolonged feeding

period by the action of methoprene. But on other starvation days due to increased maintenance cost the larval weight has been decreased as has been observed by Gaaboub *et al.* (1985). Calvez (1981) and Trivedy (1993) have argued that starvation during obligatory feeding period results in low larval weight. A similar effect is apparent in the methoprene-treated starved batches during the dry part of summer.

Except for the day 4 in male in all other starvation days larvae of both the sexes with methoprene have been able to improve the cocoon shell weights than JHa-free control. This reveals that the JHa has the ability to increase silk synthesis in starved batches also by inducing better assimilating of food. But the day 4 male larvae might have been affected by starvation and synthesized silk at low level (Kurata, 1985) or have increased maintenance cost due to prolongation of feeding period and thereby decreasing the silk-yield (Muthukrishnan *et al.*, 1978).

Female pupal weights have been improved significantly over the control (untreated) value in all the starved batches. This supports the observation of Yun and Quan (1991) who have argued that JHa significantly increases reproductive bio quality in *B.mori* when applied to the fifth instar larvae.

Thus, the results suggest that if required, starvation combined with the JHa application may be imposed during the facultative feeding period except on day 4.

5.8.2 KPGB : without methoprene administration : The ERR% has been significantly reduced during the wet part of summer than during the dry part of summer in both control and starved batches. Starvation on days 1 and 7 has produced somewhat better result, though lower than control, indicating that the season is not conducive to bivoltine rearing.

Mortality is higher when the larvae have been starved during early part of the fifth instar. The result corroborates the observation by Rao (1988), Paul *et al.* (1992). Mishra and upadhyay (1992) have argued that higher relative humidity during the season facilitates higher consumption of food but high temperature induces mortality in bivoltine.

Starvation has reduced the final larval weight in all the batches when compared with the control. However, the effect has been least on starvation day 7. Final larval weight has been higher during wet part of summer than in the dry part. Extension of larval duration due to starvation during obligatory feeding period has resulted in reduced final larval weight on account of increased metabolic cost (Schroeder, 1976).

Starvation on days 7 and 8 in case of male and day 1 in case of female has resulted in relatively better cocoon shell weights though the weights are significantly much lower than that of the control. Thus, the result suggests that feeding during obligatory period plays an important role for synthesis of silk which corroborates the opinions of Fukuda *et al.* (1963), Kurata (1985), Tojo *et al.* (1981), Dingnong *et al.* (1991). The female larvae are required to share the food energy for the formation of both cocoon shell and eggs. This is why the female larvae are more sensitive to starvation than the males (Kadono - Okuda *et al.*, 1986; Tzenov and Petkov, 1993). The lowering of cocoon shell weights due to starvation in females may be related to this reason.

Among the starvation days the female pupal weights have been relatively better on days 7 and 8, though the weights are much lower than the control pupae. This indicates that the allocation of energy for reproduction gets impaired if the larvae are starved during obligatory and early facultative feeding days.

With methoprene administration : Likewise in dry part of summer, during the wet part of summer too, methoprene has reduced the ERR% in all the starved batches as well as in the control. This suggests that methoprene administration is not at all helpful during this adverse season of rearing.

Impairment of final larval weight caused due to daywise starvation during obligatory feeding period has not been recovered after treatment with methoprene. This suggests that the prolongation of larval duration by the action of methoprene does not help the larvae to compensate for the consumption and assimilation loss and promote growth at par with the JH-free control. However, larvae of starvation day 1 and to some extent of day 7 have attained the weight of JH-free control. This result suggests that feeding during obligatory feeding period and early part of facultative feeding period play important role for the larval weight gain.

Only in case of starvation days 1 and 8 in males and in none of the days in female, the application of methoprene has been able to compensate the impairment of cocoon shell weights. This suggests that food consumed during obligatory feeding period and also early part of facultative feeding period in the males and during the entire fifth instar larval life in the females the food energy is distributed for both cocoon shell and reproduction. Therefore, the larvae should not be deprived of food for better cocoon shell.

Likewise in dry part of summer, methoprene has been found to be ineffective during wet part of summer also in recovering the impairment of female pupal weights due to starvation.

Thus, the result suggests that starvation on any day during wet part of summer is not at all advisable. However, if required food deprivation may be done during extreme later part (days 7, 8) of fifth instar.

5.8.3 KPGB x P₅ : without methoprene administration : The ERR% has been significantly reduced in this hybrid during wet part of summer when compared with that of dry part of summer. Such an effect has also been observed in KPGB. This suggests that wet part of summer is not conducive for rearing of bivoltine.

The larval weights have shown non-significant difference from the control larval weight when starved on day 4. In rest of the days the weight gain has been significantly lower than the control. Maximum loss has been recorded from starvation during facultative feeding period. Muthukrishnan and Pandian (1987) have argued that when metabolic loss due to high temperature and high humidity under starvation stress is low the larvae attain good larval weight. Larvae starved on day 4 have suffered least metabolic loss due to respiration and maintenance cost. Starvation on days 7 and 8 resulted in the lowest larval weight. This may perhaps be due to consumption of critical minimum ration by larvae before the starvation stress have been imposed and hence, entered metamorphosis during this adverse season. Such an argument has also been provided by Wigglesworth (1934) and Friends *et al.* (1965).

For male cocoon shell weights, starvation on days 1, 2 and 3 have no significantly different result from the control and within the starvation days, suggesting that feeding during facultative feeding period is largely responsible for male cocoon shell weight. A similar result has been found for the female cocoon shell under starvation on any day from 1 to 5. But starvation on any day from 6 to 8 has resulted in very low cocoon shell weight. Thus, it can be assumed that this hybrid has some vigour to withstand starvation stress during the early part of fifth instar.

Starvation on days 1, 2 and 5 has no gross effect on the female pupal weights. The differences in the weights from the control value

have no significance. In rest of the days of starvation the values have been impaired. the result is almost similar to the one obtained during dry part of summer.

With methoprene administration : The ERR% has not been improved by the application of methoprene. Such a result has also been obtained for Nistari and KPGB during this season.

Significant improvement in larval weights over the JH free control value has been observed in starvation days 1, 7 and 8. This suggests that compensation in food assimilation has been increased by the action of methoprene. Thus, exogenous methoprene has been able to overcome the starvation stress and in recovering from the impairment of larval weight.

Improvement in cocoon shell weights in starvation days 4,5,6,7,8 in males and only in day 4 in female over that of JH-free control individuals indicates that the females are more susceptible to starvation as they are required to allocate energy for both cocoon shell and egg production (Kadono-Okuda *et al.*, 1986; Tzenov and Petkov, 1993).

Impairment in female pupal weights has not been recovered by the action of methoprene suggesting its ineffectiveness for this character on this bivoltine hybrid during wet part of summer. The overall result indicates that in the bivoltine hybrid of KPGB x P₅ starvation is not advisable on any day of the fifth instar. But in case of exigency starvation may be imposed on the days 5, 6 and 8 of facultative feeding period and along with methoprene.

5.9 Reproductive Characters of *B.mori* Caused Due to Daywise Starvation During Wet Part of Summer

5.9.1 Nistari : without methoprene administration : During the wet

part of summer the female pupal weight has been higher in the batches starved on days 1, 3 and 4. In spite of the low ECD to female pupa in batches starved on day 4, the live female pupal weight has been higher than of the control. This is evidently due to accumulation of excess water in pupal body during this season. Such accumulation of excess body water by starved larval batches has been observed by Delvi *et al.* (1988) and Paul *et al.* (1992). Again, the low female pupal weight on starvation days 5 and 6 has resulted from food deprivation after consumption of critical minimum ration required by the larvae undergoing pupal moult. Kawaguchi *et al.* (1991) have also argued that larval maturity to spin cocoons and undergoing metamorphosis in *B.mori* depend on the critical minimum ration. The total ovariole length, total number of eggs in ovariole and fecundity have been directly related to pupal weight as has been evident during the dry part of summer. Likewise in dry part, during the wet part of summer too, the reproductive characters are impaired when starvation is imposed on days 5 and 6, the late of facultative feeding period (Himantharaj, 1994; Kawaguchi *et al.*, 1991).

With methoprene administration : As in the dry part of summer the reproductive characters in the JH treated batches of Nistari have shown a significant improvement than the JH free starved batches. Impairment of pupal weights have been recovered in all the starvation days following methoprene application. Ovariole length has also been increased than in the JH-free control individuals on starvation days 1,2, 3 and 7. Days 4 and 5 have suffered much from the starvation stress because the growth of ovary has been affected (Parlak *et al.*, 1992). Number of eggs in the ovarioles and fecundity have also been reduced in these days of starvation.

Thus, it can be suggested that during wet part of summer starvation can be imposed on Nistari on any day of the fifth larval instar except on

days 4 and 5. However, starvation is to be accompanied with the JHa application.

5.9.2. KPGB : without methoprene administration : Impairment caused due to daywise starvation has been the least on days 7 and 8 though the effect was significant when compared with that of control during the wet part of summer. The larval performance for all the reproductive characters has been relatively improved in this season than in the dry part of summer. High water content in mulberry leaves has facilitated better conversion of digested food to pupal mass. But due to disturbances in the development of ovary when starved on days 4, 5, 6, the fecundity has been reduced than in other days of starvation (Parlak *et al.*, 1992).

With methoprene administration : Recovery of impairment over the values in JH-free control batch has not been observed in any of the JH-treated daywise starved batches. This indicates that methoprene is inefficient to improve the losses caused by starvation during wet part of summer. This is mainly due to lowering of digestibility of food and prolongation of the larval duration during this adverse season. The wet part of summer though facilitates high consumption due to more water in leaves but pupal weight is comparatively reduced due to increase in maintenance cost (Paul *et al.*, 1992).

Thus, it can be suggested that, if required, starvation may be imposed during later part of facultative feeding period of the fifth larval instar but application of methoprene is to be avoided.

5.9.3. KPGB x P₅ : without methoprene administration : In comparison to the dry part of summer the hybrid KPGB x P₅ has suffered more for the reproductive characters during the wet part of summer. The

female pupal weight and other correlated characters have been reduced. Least impairment has been recorded on starvation day 7 indicating that the later part of facultative feeding period can be recommended as an ideal time for starvation of fifth instar larvae if required.

With methoprene administration : Prolongation of larval duration with the application of methoprene in daywise starved batches has no promoting effect during the wet part of summer as in case of pure bivoltine KPGB.

Hence, it can be inferred that the application of methoprene should be avoided and if required starvation may be imposed on day 7 only.

Hatching percent : Hatching percent of the fertilized eggs during wet part of summer in all the three experimental breeds Nistari, KPGB and hybrid of KPGB x P₅ has been found to be an independent trait without having any relation with starvation and starvation accompanied with the JHa application. In this respect there is no differences in the results obtained in these two seasons.

5.10 Utilization of Nitrogen by the Fifth Instar Larvae Due to Daywise Starvation During Wet Part of Summer

5.10.1 Ingestion :

Nistari : There has been no significant differences in the quantity of leaf nitrogen ingested by the larvae of different starvation days except for the day 1. Because of higher adaptability to adverse climate of wet part of summer in West Bengal starvation has no negative effect on nitrogen ingestion. In spite of low level of leaf nitrogen during this season (2.66%)

there is no impairment in the consumption of nitrogen. Even this has been higher than in the dry part of summer. The low nitrogen content of the leaves has been made up by higher quantity of leaf consumption which supports a similar observation of Slansky and Fenny (1977) in *Pieris rapae*. Again, the higher food consumption has been possible due to high water content of mulberry leaves during this season which has facilitated better nitrogen intake (Muthukrishnan and Pandian, 1983; Pandian *et al.* 1978). Treatment with methoprene has further increased the consumption of food in all the treated batches thus, the ingestion of higher quantity of Nitrogen.

KPGB : The wet part of summer is known as the adverse season for bivoltine rearings in West Bengal (Rao *et al.*, 1988; Paul *et al.*, 1992). The amount of nitrogen ingestion has been lowered except for the starvation day 7. Starvation on day 4 which falls in between obligatory and day 4 facultative feeding periods has suffered from maximum impairment. Treatment with methoprene has increased the consumption of leaf due to prolongation of larval span thus, more nitrogen has been ingested.

KPGB x P₅ : This bivoltine hybrid has also suffered from impairment in nitrogen ingestion due to starvation and the impairment could not be recovered by the JHa.

5.10.2 . Excretion :

Nistari : The quantity of nitrogen excretion has been significantly increased only in the starvation day 6. Except for the days 1 and 5 in all other days the excretion of nitrogen has been lowered than in the control larvae. Treatment with methoprene has further increased the nitrogen loss in the excreta than in the control indicating a reduction in the

digestibility of nitrogen during this season.

KPGB : The percentage of nitrogen excretion is quite high in all the days of starvation but the difference is very little in the starvation days 7 and 8. In terms of total quantity, the nitrogen excretion has been very high in all the starvation days except 7 and 8 than in case of control. Treatment with methoprene also has resulted in a similar effect with respect of dietary nitrogen excretion.

KPGB x P_g : Both in terms of percentage and quantity the nitrogen excreted in the faeces has been always significantly lower in the starved larvae of all the batches than in the control larvae. This implies a higher efficiency of nitrogen utilization in this bivoltine hybrid. Treatment with methoprene has increased the percentage of nitrogen excretion only in the starvation days 2, 3, 7 and 8 and that is again of lower magnitude.

5.10.3 Digestion and digestibility percentage

Nistari : There is a little impairment of nitrogen digestion in the starvation days 3, 5 and 6. This is in term of quantity of nitrogen digested. But in term of percentage that is the digestibility has no significant lowering from the control values. Rather the digestibility has been significantly increased in the days 2 and 4. Thus, the starvation has no detrimental effect in Nistari on nitrogen digestibility. Treatment with methoprene however, has reduced the digestion and digestibility of nitrogen during this season.

KPGB : Due to less intake of food the digestion has also been less during this adverse rearing season. Except for the days 7 and 8 the

digestibility of nitrogen has been affected, may be due to high water level in the leaves (Parpiev, 1968) though Legay (1958) suggested that high relative humidity increases digestibility of nitrogen in *B.mori* which may be upto 100%. But in the control batch the value is only 79.06%. Treatment with methoprene has further reduced the digestibility of nitrogen may be due to higher metabolic loss caused by starvation and high temperature.

○ **KPGB x P₅** : This hybrid has shown a better nitrogen digestion as compared with the pure KPGB. Impairment of nitrogen digestion has been observed due to food deprivation on all the days except the day 4. Digestibility of nitrogen has been remarkably better in most of the starvation days except the day 8. Treatment with methoprene significantly decreased the digestibility of nitrogen suggesting that methoprene reduces nitrogen digestibility during this adverse season.

5.10.4. Distribution of nitrogen

5.10.4.1. Larval body

Nistari : There has been a reduction in the efficiency of nitrogen assimilation in larval body on account of starvation on different days. Thus, starvation affects nitrogen utilization by the larval body (Schroeder, 1976). Treatment with methoprene significantly increases the flow of nitrogen in all the starved batches.

KPGB : The quantity of dietary nitrogen utilized in the larval body in the control batch is much lower than the result obtained of Horie and Watanabe (1986). This difference may be due to difference in race of silkworm, season of rearing and nitrogen percent in mulberry leaves. The

flow of nitrogen in general has been affected in all the starved batches with maximum detrimental effect on the late obligatory and early facultative feeding days. The treatment with JHa has no noticeably better effect on the flow of dietary nitrogen to larval body.

KPGB x P₅ : Food deprivation on any day of the larvae has reduced the flow of dietary nitrogen to the larval body. Methoprene even could not improve the situation, though the total quantity of nitrogen has been largely increased than the JHa-free larval batches.

5.10.4.2. Male cocoon shell

Nistari : Among the starved larvae relatively better flow of nitrogen from larval body to the male cocoon shell has occurred in batches fed during early part of obligatory feeding period. Nagata and Kobayashi (1990) have also observed that storage protein in haemolymph is related to the quantity of food and activity of feeding. They have emphasized that starvation reduces the level of storage protein which again increases when feeding is restored and is related to the nitrogen level in food. Treatment with methoprene only has increased level of nitrogen in cocoon shell but cannot recover in any of the batches from impairment.

KPGB : Starvation during obligatory and early facultative feeding periods has reduced the nitrogen flow to male cocoon shell. This result suggests that feeding during obligatory feeding period is essential in bivoltine as has been suggested by Janarthan *et al.*, (1994). Treatment with methoprene has recovered the impairment in the days 2 and 3 but not in other days.

KPGB x P₅ : Starvation during facultative feeding period (days 6, 7,8)

has reduced flow of nitrogen towards male cocoon shell very prominently. This is because food consumed during later part of fifth instar is effectively utilized for cocoon shell production. Methoprene has been able to improve situation in days 6, 7, 8 through the prolongation of larval period.

5.10.4.3. Female cocoon shell

Nistari : Among the starved batches relatively better flow of nitrogen has been observed during obligatory feeding period. This suggests that food nitrogen consumed during facultative feeding period has been utilized for cocoon shell production. Treatment with methoprene has only increased the flow of nitrogen but there has been improvement over the control values due to reduction in digestibility of nitrogen.

KPGB : Starvation has seriously lowered the flow of nitrogen to female cocoon shell on days 2 to 6 covering both obligatory and facultative feeding periods. The day 4 has suffered the most. This is perhaps because of the sharing of nitrogen for cocoon shell and female pupal biomass as ovary development also commences from this day. Treatment with methoprene has only increased flow of nitrogen in all the batches but impairment caused could not be recovered.

KPGB x P₅ : The flow of nitrogen has been severely reduced in the days of facultative feeding period. This suggests that nitrogen consumed during facultative feeding period is effectively utilized for cocoon shell in the bivoltine hybrid. Treatments with methoprene has only increased the flow of nitrogen in cocoon shell but a recovery to the level of control value has not been possible.

5.10.4.4. Female Pupa :

Nistari : Food deprivation has lowered the flow of nitrogen in all the batches with a greater effect during the later part of facultative feeding period. The relatively less affected days are 1, 4 and 5. 34.48% of digested nitrogen has been utilized for female pupa in control batches. Except for the starvation days 5 and 6 more than 30% of the larval nitrogen has been utilized for female pupa. Methoprene has been able to increase the flow of nitrogen from larval body to female pupal body. But the values could not cross that of the JHa-free control batch except for the days, 1 and 3. Therefore, starvation accompanied with methoprene application is quite suggestive for these two days only during wet part of summer and for this character.

KPGB : Except the starvation day 7 the flow has been reduced. But the situation is not alarming in the days 1, 3, 4 and 8. Therefore, in case of exigency starvation can be imposed with a little sacrifice for this character. Except for the days 2 to 4, in all other days the JHa has been able to boost up the nitrogen level upto the JHa-free control value or even higher than that in days 1 and 7. In the days 2 to 4 the flow has been improved much over the JHa-free starved batches of corresponding days.

KPGB x P₅ : Starvation during the obligatory feeding days has affected less than in the facultative feeding days when total flow of nitrogen is considered. But in term of percentage of nitrogen flow the situation is relatively better in the starvation days 1, 3, 4 and 5. During wet part of summer starvation if required may be imposed on days 1 and 4. The starvation days in which the flow was lowered to a greater extent has been improved considerably by action of methoprene. Compared with the value of JHa-free control batch, starvation along with JHa-application can be recommended for this character during wet part summer on the days 1 and 4 without any impairment and with little impairment on the days 2,3 and 8.

5.11 Determination of Doses of the Imidazoles KK-42, KK-22 and KK-110 for the Induction of Trimoulters in the bivoltine hybrid of KPGB x P₅ During Wet Part of Summer

5.11.1 Induction of Precocious Metamorphosis and Formation of Trimoulter Cocoons Due to Different Doses of Imidazoles

Induction of precocious metamorphosis by the use of imidazoles are known to be dependent on dose and time of application (Kuwano *et al.*, 1984; Asano *et al.*, 1986). The present result of precocious moulting with the imidazoles when applied to 0-hr-old 4th stage larvae corroborates almost the similar observation of Asano *et al.*, (1986). Result indicates that the highest rate of precocious metamorphosis is induced by topical application of only 3 µg KK-42/larva which resulted in a 92% cocoon formation and out of this 92% pupation has been successfully taken place at 94.56%. A lower dose of 1 µg/larva of KK-42 causes only 70% precocious metamorphosis with 94.28% pupation. Higher dose of 5 µg of KK-42 causes 39% larval mortality, only 82% precocious metamorphosis and 93.90% pupation. But Kawaguchi *et al.* (1993) have reported no larval mortality even at a dose of 30 µg/larva. 5 µg/larva of KK-42 have been considered as a standard dose by many authors (Akai and Mauchamp, 1989; Gu *et al.*, 1992) for the induction of 100% precocious metamorphosis. It has also been suggested that among the imidazoles known so far, KK-42 is found to be the most potent compound which can induce precocious metamorphosis at a relatively low dose of 1 ~ 4 µg/larva (Kuwano *et al.*, 1988). Yamashita *et al.* (1987) have observed that a dose of 30 µg KK-42 causes 100% precocious metamorphosis. It is suggested that KK-42 acts directly on the prothoracic gland cells without killing the cells in *B.mori* larvae (Kadono-Okuda *et al.*, 1987; Yamashita *et al.*, 1987; Darvas *et al.*, 1989; Oshiki *et al.*, 1990). But no information

is available on the spectrum of effect of KK-42 under high temperature and high humidity environment. It seems that due to smaller size of Indian bivoltine silkworms than their Japanese and Chinese counterparts, the freshly ecdysed fourth instar larvae of present investigation cannot withstand higher dose of KK-42 may be due to sudden decrease of ecdysteroid level in haemolymph (Reese, 1985) or may be due to some delayed cytotoxicity promoted under high temperature and high humidity leading to larval death. Delayed toxicity with KK-42 treatments have also been reported in *Oncopeltus fasciatus* nymph by Kuwano *et al.* (1992).

KK-110 has been reported to act at a lower dose of 1 ~ 4 $\mu\text{g}/\text{larva}$ and is known to be less sensitive than KK-42 (Kuwano *et al.*, 1988). But in this finding a dose of 5 $\mu\text{g}/\text{larva}$ cannot induce expected precocious metamorphosis in KPGB x P₅ larvae. A dose of 10 $\mu\text{g}/\text{larva}$ has been found to be the best to induce 93% precocious metamorphosis with 94.62% pupation. A higher dose of 15 $\mu\text{g}/\text{larva}$ induces 91% precocious metamorphosis with 92.30% pupation.

KK-22 appears to be ineffective at a lower dose of 5 $\mu\text{g}/\text{larva}$ which resulted in only 26% precocious metamorphosis and 84.69% pupation. But a dose of 10 $\mu\text{g}/\text{larva}$ induces 75% precocious metamorphosis and 80.68% pupation. Whereas 20 $\mu\text{g}/\text{larva}$ results in 86% precocious metamorphosis and 90.10% pupation. The results agree with the finding of Asano *et al.* (1986, 1987) who have recorded that KK-22 at a dose from 20 to 40 $\mu\text{g}/\text{larva}$ gives the highest percentage of precocious cocoons.

5.11.2 Rearing Performance of KPGB x P₅ Due to Different Doses of Imidazoles.

Average cocoon weight : The size of the cocoons produced are smaller

than that of the control in all the treatments with different doses of the three imidazole compounds applied. A similar observation has been made earlier by Kanda *et al.* (1986) and Himantharaj *et al.* (1996) who also recorded lower cocoon weights after treating the *B.mori* larvae with different kinds of imidazoles. In all the treatments the cocoon size has also varied in both the sexes according to the doses applied, a decrease in size with the increase of the doses of the imidazoles. The present finding corroborates this observation, the cocoon weights have also been reduced with the increase in dose of all the three imidazole compounds (Kawaguchi *et al.*, 1993). In *B.mori* the accumulated food in larval body is converted to the cocoon shell and pupal biomass. Again, larval body weight is related to food intake (Ding Nong *et al.*, 1991) and finally to storage protein concentration in larval haemolymph. This is also influenced by nutrition (Nagata and Kobayashi, 1990). An overall lower food consumption by induced trimoulters due to skipping off of the usual physical fifth stage causes a reduced cocoon weight. The fifth larval instar alone consumes about 87% of the food required by all the larval instars of *B.mori* (Matsumura and Takeuchi, 1950). Though the imidazoles prolong the duration of 4th instar larvae by 3-6 days over the duration of control larvae, the total food consumption is much less than the tetramoulter larvae. Hence, imidazole compounds certainly put a stress on normal development of larvae forcing them to the neurohormonal response leading to precocious pupation, provided the larva has attained the critical weight. Ecdysteroid (MH) in haemolymph responsible for moulting and metamorphosis (Reese, 1985) and the juvenile hormone responsible for the control of cellular polymorphism and growth (Riddiford, 1985) are effectively lowered by the treatment with imidazoles. According to Wu^{et al.} (1993) corpora allata are metabolically inhibited by the imidazole compounds to synthesize JH. MH level is also lowered for several days following KK-42 application. The low level of JH later acts on the brain

neuro-secretory cells to release PTH and thereby increases MH. This late MH peak under low titre of JH leads to precocious pupation. Although the attainment of larval critical weight is a prerequisite for the induction of trimoulter (Asano *et al.*, 1987), allocation of low amount of larval nutrients for cocoon shell certainly lead to reduce cocoon shell weight which has been impaired proportionately much than for the pupal biomass allocation. This justifies the higher impairment of shell weights than those of the pupal weights as compared with these weights of the control tetramoulter.

Cocoon shell weights : Reduced cocoon shell weights have been observed in imidazole treated batches. Increase of shell weights is directly related to the amount of food intake, its allocation and conversion efficiency for shell by the fifth instar larvae (Dingnong *et al.*, 1991). Cocoon shell formation is affected by a change in the profile of storage protein in haemolymph due to the impact of imidazole. A faster rate of development of the posterior silk gland in trimoulters (Zhou Yuan Shen, 1992; Hao Lin *et al.*, 1992) demands for a higher quantity of storage protein within a short time (Nagata and Kobayashi, 1990). Compared with the control values the male and female cocoon shell weights have been on the average 34.6% and 37.7% after KK-42, 31.9% and 35.4% after KK-110 and 34.3% and 37.2% after KK-22 treatment respectively. He and He (1994) have obtained a shell weight of 35% of the control value from the trimoulters. Kiuchi *et al.* (1985) has described these low cocoon shell weights as one third that of the control. The present result corroborates these previous observations. The lowering of the cocoon shell weights at higher dose of KK-42 also supports the earlier observation of Kanda *et al.* (1986). A similar result has been obtained with KK-110 treatments. However, treatment with KK-22 at 20 µg/larva resulted in heavier cocoon shell weights in both males and females though the difference in the

results obtained from 10 and 20 μg doses has been non significant in case of female shell weight.

Silk ratio (SR %) : SR% is calculated from cocoon and cocoon shell weights. It is obvious that when cocoon shell weights are lower as in case of induced trimoulters the values of SR% recorded are also lower when compared with the shell weights of control tetramoulters.

Filament length : Reduced filament length has been obtained for all the treated batches. As cocoon shell weights are very low, the filament lengths have been reduced correspondingly. Though the cocoon shell weights are on an average 30-35% that of control, compared with the control values the average filament length has been 42.3% in case of KK-42, 39.4% in case of KK-110 and 51.1% in case of KK-22 treatments. This indicates that the cocoon filaments are of fine denier but having a high reelability. This qualitative superiority of filaments supports the finding of Kanda *et al.* (1986), He and He (1994), Xue (1992), Tan *et al.* (1992) and Himantharaj *et al.* (1996).

Demier : The size of cocoon filament can easily be regulated within the same silkworm race by the application of different doses of imidazole compounds (Akai *et al.*, 1986). This also holds good for the Indian bivoltine KPGB x P₅. In this race the demier ranges from 1.57-1.71 for KK-42 (1- 5 μg /larva), 1.46-1.51 for KK-110(10-15 μg /larva) and 1.9 - 1.94 for KK-22 (10-20 μg /larva) against the value of 2.01 in case of control tetramoulters.

5.11.3. Food Consumption and Assimilation by the Larvae of KPGB x P₅ Due to Different Doses of the Imidazoles

Food consumption : The results reveal that depending on the dose of

different imidazoles different quantities of food have been consumed by the trimoulter destined larvae. The quantities were less than 50% (dry basis) of the consumption by the control tetramoulter larvae during 4th and 5th stages taken together. But Xue *et al.* (1990) have reported that the food consumption by all the larval stages of trimoulter-destined generation induced by imidazole is 30% less than all the larval states of control tetramoulter. The consumption has been lowest in KK-22 treated trimoulters. There is a saving of about 50% of leaves (dry basis) due to treatment with imidazoles. Prolongation of fourth larval duration by 3-6 days in induced trimoulters beyond the control fourth stage larvae has been reported earlier by Kuwano *et al.* (1984), Kiuchi *et al.* (1985), Asano *et al.* (1986, 1987) and Yamashita *et al.* (1987). In the present experiment this prolongation ranges from 3 to 3.75 days. This low limit of extended duration in this investigation may be due to a combined effect of differences in silkworm race, mulberry cultivars, dose of imidazoles and rearing environment.

Assimilation : Assimilation has been increased with the increase of dosages of KK-42 and KK-22. But in case of KK-110, treatment with lower dosages have shown higher assimilation values. Assimilation in treated larvae is higher than that of control fourth stage larvae alone. This is essential for attaining the critical weight to undergo precocious pupation.

Absolute consumption rate : Absolute consumption rate is well related to the total consumption and larval duration. When compared with the combined rate of control fourth and fifth stage larvae, the absolute consumption rate are 62.0, 61.8 and 55.0% in case of treatments with KK-42, KK-110 and KK-22 respectively. This increased rate justifies the resource storage for silk gland and reproductive energy allocation of the trimoulters within the reduced period of larval life.

Absolute gain in larval weight : Gain in larval weight in comparison to the control larvae, has been 45% in KK-42 treatments, 49.3% in KK-110 treatments and 39.4% in KK-22 treatments. However, gain in larval weight by the treated larvae is many times over that of control 4th stage for attaining the critical weight for pupation (Aanso *et al.*, 1987; Gu *et al.*, 1992). A constant of 100:40 in gain in larval weight between control tetramoulter and induced trimoulter has been established by Kiuchi *et al.* (1986). The present finding corroborates this result though the experiment has been carried out in tropical humid condition.

Absolute growth rate : Absolute growth rate of induced trimoulters has been more than double that of the control fourth stage larvae. But compared with the growth rate of tetramoulter control fourth and fifth stages taken together, the growth rate of trimoulters have been 64.9%, 68.9% and 56% following the treatments with KK-42, KK-110 and KK-22 respectively. Prolongation of larval period accompanied with increased body weight is indispensable for indication of precocious metamorphosis (Asano *et al.*, 1987) through inhibition of ecdysteroid synthesis without any large influence on the general sterol metabolism for growth (Kadono-Okuda *et al.*, 1994).

5.11.4 Efficiency of Conversion of Digested Food (ECD) Due to Different Doses of the Imidazoles.

Assimilation efficiency : On an average the assimilation efficiency has been 56.3% , 59.02% and 50.16% after treatment with KK-42, KK-110 and KK-22 respectively as against the 50.46% in control larvae. This reveals an increase in 6.3% assimilation efficiency due to KK-42 and 9.02% due to KK-110. An increase in 4-9% dietary efficiency has been recorded over the control by Zhuang *et al.* (1992). With the exception of KK-22 the results obtained from other two imidazoles are in agreement with

those of Zhuang *et al.* (1992). On the whole, higher assimilation efficiency in larvae due to imidazole treatments is a physiological compensation within the shortened feeding regime of one generation.

Larval body : With comparison to the control values of 5th stage larvae the averages mature weights attained by induced trimoulters have been 32.7%, 33.2% following the treatments with KK-42, KK-110 and KK-22 respectively. However, the weights have been double that of the fourth instar control larvae. The growth in the trimoulter is influenced by general sterol metabolism in larvae independent of the impact of imidazole compounds (Kadono-Okuda *et al.*, 1994). Asano *et al.*, (1987) have recorded in the induced trimoulters an increase in body weight by 1.6 times that of the 4th stage. The present result shows an increase of 1.9 times due to KK-42 and KK-110 and 1.75 times due to KK-22. These differences might be attributed to the racial difference. Asano *et al.* (1987) also have reported an increase in 40% larval body weight in trimoulters due to KK-22. But in the present study the increase has gone upto 75% of the fourth instar control larval weights. A prolongation of larval life including the phagoperiod beyond the span of control fourth instar larvae causes such increase. This is evident from the observation of Asano *et al.* (1987) who claims that the increase of body weights of trimoulters is at par with the control larvae upto 72 hrs of feeding and thereafter, the induced trimoulter larvae start gaining further weights by increasing consumption of food during the extended larval duration. A notable observation in this experiment is that lower doses of imidazoles have resulted in higher larval weights except in case of 1 μ g KK 42/larva. This may imply that the different doses of imidazoles have an impact on the initiation of the programme of precocious pupation, accordingly the weights vary. ECD to larval body on an average are 41.19% , 41.55% and 43.19% in cases of treatments with K-42, KK-110 and KK-22 respectively as against 44.29%

in case of control larvae. This implies that the imidazoles have no impact on the ECD to larval body. There is no significant difference among the values.

Cocoon shell : In comparison to the tetramoulter control individuals the cocoon shell weights have been lower in all the induced trimoulters due to less intake of food. Correspondingly the ECD to cocoon shells are relatively lower in the induced trimoulters. On an average the values attained are 11.57% and 10.70% due to KK-42, 9.75% and 9.23% due to KK-110 and 13.62% and 12.57% due to KK-22 in cases of males and females respectively. Due to low allocation of food resources for cocoon shell, the ECD values in induced trimoulters have been significantly lowered from the control values. This result corroborates the finding of Kubota *et al.* (1988) and Kiuchi *et al.* (1986) who also have recorded a lower ECD for cocoon shell in trimoulters induced by imidazole compounds.

Pupa : With comparison to the control tetramoulter, the average pupal weights have been 58.3% and 67.2% in case of treatments with KK-42, 64.2% and 66.9% in KK-110 and 60.85% and 75% in KK-22 treatments for males and females respectively. Treated larvae consumed less than 50% of food consumed by the tetramoulter. But the loss of weight is not more than 40% particularly in case of female pupa. Slansky and Scriber (1985) have advocated that under stressed condition larvae are forced to act on the neurohormonal response, attain critical weights soon and pupation results. Maximum energy from food is allocated to the pupae for maintaining reproductive vitality and perpetuation of the race. Kiuchi (1996) has proposed that due to short larval period than control and also due to overall less consumption of food a greater part of resources are allocated to pupa of induced trimoulters. Concurrently, the shell weights have been proportionately impaired. With reference to the control values

the ECD to pupa on an average are 19.40% and 27.09% for KK-42, 19.46% and 24.71% for KK-110 and 24.34% and 35.97% for KK-22 applications in cases of males and females respectively. Reproductive allocation for the treated female pupae is always higher than in the treated male pupae and in control tetramoulter pupae (Kiuchi *et al.*, 1986, 1996) for the production of miniature but viable reproducing adults (Staal, 1986). Present finding corroborates the finding of Kiuchi *et al.*, (1986).

From the overall analysis of the results it transpires that the doses of 3 μg of KK-42/larva, 10 μg of KK-110/larva and 20 μg of KK-22/larva are the most suitable and effective doses for the induction of trimoulter in KPGB x P₅ bivoltine hybrid of *B.mori* during the unfavourable wet part of summer of tropical humid regions such as in West Bengal.

5.12 Comparative Nutritional Efficiencies of Larvae Due to Selected doses of the Imidazoles

5.12.1 Food consumption : Food consumption by induced trimoulters after treatment with different imidazoles has been less than 50% of control (Fourth and fifth larval instars taken together). However, there has been significant differences in the amount of consumption due to the three compounds indicating a differential action of the imidazoles. Xue *et al.* (1994) have reported a 30% less leaf consumption by induced trimoulters but the authors did not refer to the season of the experiment. As the present experiment has been conducted during the wet part of summer when both temperature and humidity are high, the consumption ratio is low, and hence, a 50% decrease of food consumption by the larvae. However, the total leaf consumption by the apparent fourth instar larvae alone after treatment with the imidazoles are considerably higher due to

prolongation of the fourth instar larval period. This is inevitable for precocious metamorphosis by trimoulters (Kuwano *et al.*, 1984 ; Kiuchi *et al.*, 1985; Yamashita *et al.*, 1987; Asano *et al.*, 1986, 1987).

5.12.2. Digestion and assimilation : Except for KK-22 the other two compounds have raised the assimilation rate upto 8% than the control (combined 4th and 5th stage). An increase of 4-9% assimilation efficiency over the control have been recorded by Zhuang *et al.* (1992) which supports this finding. Digestion and assimilation efficiencies are found to be different and significant among the treatments except in case of KK-22 (20 µg) where the efficiency is almost the same as in the control larvae.

5.12.3 Gain in larval weight : Gain in larval weight is related to food intake and assimilation by the larvae. Growth of induced trimoulter larvae is influenced by general sterol metabolism which is not affected by the action of imidazoles (Kadono Okuda *et al.* 1994). Gains in larval weights and growth as indicated by Asano *et al.* (1987) have been at par with those of control larvae upto 72 hrs of age. Thereafter the rate has been accelerated for attaining larval weight to the critical level required for pupation after a little prolongation of larval period, thus gained a higher weight over the control. However, the larval weights in the trimoulters have always been lower than the values of control tetramoulters. The significant differences in the results obtained from different imidazoles indicate a differential impact of the compounds.

5.12.4 Efficiency of conversion of digested food (ECD%):

ECD% to larval body : ECD to larval body has been almost the same in the larvae treated with different imidazoles but the efficiencies has been slightly lower than that of control tetramoulters. The highest value

has been obtained in case of KK-110 (10 µg) treated larvae. The result indicates that the imidazoles have very little effect in lowering the ECD to larval body.

ECD% to cocoon-shell : ECD% to cocoon shell in trimoulters has been found to be greatly reduced. This may be due to more allocation of food towards pupal biomass, Kiuchi *et al.* (1985), Kobota *et al.* (1988) and Himantharaj *et al.* (1996) also have recorded lower cocoon shell percentage in the imidazole induced trimoulter cocoons. ECD to male cocoon shell are relatively higher than the values of female cocoon shells in the present result. The difference in the values resulted from three imidazoles indicate a differential action of the compounds used.

ECD% to pupal biomass : Compared with the control values and irrespective of the sexes the ECD to pupal biomass of induced trimoulters has been higher than that of tetramoulter control individuals. It appears that the imidazole compounds induce the conversion of greater share of digested food to pupal body than to the cocoon shell, so that the viability of the resultant adults is not impaired to a greater extent. Almost a similar result was obtained by Kiuchi ^{*et al.*} (1986) ^{*and Kiuchi*} (1996). The values due to KK-22 (20 µg/larva) shows a difference with the results obtained from the other imidazoles.

5.13 Comparative Effect of the Imidazoles on Rearing Performance

5.13.1 Effective Rate of Rearing (ERR%) : The ERR% has been increased after the treatment with different selected doses of imidazoles in the hybrid of KPGB x P₅. From 62% in case of control the ERR has been increased to 85% due to KK-42 (3 µg), 78% due to KK-110 (10µg) and 81% due to KK-22 (20 µg). He and He (1994) have argued that the

physique of the induced trimoulters are stronger than the control tetramoulters. Treatment with imidazoles during adverse rainy season reduces the larval mortality. This is one of the reasons for better ERR. This supports the observation of Xue and Li (1987), Xue (1990, 1992, 1994) who recorded a lower incidence of disease and hence a reduced mortality in induced trimoulter. They have argued that induced trimoulters are less susceptible to diseases due to non entrance into the disease prone fifth larval instar. The authors recorded a better rearing performance of induced trimoulters during adverse seasons like summer and autumn. However, in the present result the ERR values obtained from the three imidazoles show no significant difference in Indian bivoltine during wet part of summer.

5.13.2 Total Larval Period : Total larval duration has been shortened by about 5 days in comparison with the tetramoulter control larval life span. Kiuchi *et al.* (1985, 1986) in their experiment have recorded a shortening of larval duration in treated trimoulters by 4 days than that of the tetramoulters. However, the authors used a different imidazole, the SSP-11. A 3.50 to 3.75 days longer larval duration beyond the duration of control larva (5.75 days) in the treated 4th stage larvae, supports the results obtained by other authors (Kiuchi *et al.*, 1985; Akai *et al.*, 1986; Asano *et al.*, 1986, 1987).

5.13.3 Cocoon weights : Cocoon weights have been reduced by about 50% of the cocoon weights of tetramoulter control insects. This is obvious due to less food intake during short larval duration of trimoulters following imidazole treatments. Imidazole induced reduction of cocoon weight corroborates the earlier finding of Staal (1985), Kanda *et al.* (1985), Akai *et al.* (1986), Kiuchi *et al.* (1986), He and He (1994) and Himantharaj

et al. (1996) . However, the cocoon weights obtained due to KK-22 are higher than those obtained due to other two imidazoles.

5.13.4. Cocoon shell weight : Cocoon shell weights have been 35% of control weights. Lower cocoon shell weights due to imidazoles have also been reported by Kanda *et al.* (1985), Kiuchi *et al.* (1986) and Himantharaj *et al.* (1996). Compared with the 50% reduction of cocoon weights the impairment of shell weights alone due to the imidazoles used is less. Furthermore, there is no significant difference in shell weights obtained due to optimum doses of the three imidazoles.

5.13.5. Filament Length : Filament lengths obtained from the trimoulters cocoon shell are about 50% that of control. In spite of the reduced shell weights the fineness and higher reelability of the trimoulters cocoons are important aspects in practical sericulture. The fineness has also contributed to the less impairment of filament length. Production of fine filament accompanied by high reelability in case of trimoulters have also been reported by Akai *et al.* (1986), Xue (1992), Tan *et al.* (1992), He and He (1994) and Himantharaj *et al.* (1996).

5.13.6. Denier : Denier of filaments obtained from trimoulters is finer than that obtained from the control tetramoulters. Variation of denier size due to application of imidazoles has been reported by Akai *et al.* (1986), Tan *et al.* (1992) and Xue (1992).

5.14 Reproductive Performance of Induced Trimoulters of KPGB x P₅ with Selected Dosage of Imidazoles During Wet Part of Summer

Pupal Weight (Live) : Pupal weight, particularly that of female pupa, has a bearing on the fecundity (Mizuta *et al.*, 1969; Kotikal *et al.*, 1989;

Jayaswal *et al.*, 1991). Metamorphic efficiency for adult development is directly related to the efficiency of conversion of digested food into larval body and then to pupal body (Muthukrishnan and Pandian, 1987). Again, during stressed feeding condition larvae after attainment of critical weight, neurohormonal changes lead to pupation (Nijhout, 1975). The resultant small pupae produce adults that make some contribution for the perpetuation of generations (Slansky and Scriber, 1985; Staal, 1986). In the present result the pupal weights recorded after treatments with different imidazoles have been 65-70% that of control. This supports the finding of Staal (1986) who has recorded the emergence of small viable and reproductive adults by treatment with imidazoles. Pupal weights have been significantly different for different imidazoles indicating again the differential action of the imidazoles applied.

Emergence Period : Metamorphic efficiency of pupa to adult depends on the quality of food storage during larval period for viable adult emergence (Muthukrishnan and Pandian 1987). The efficiency is accelerated or modified by the effect of temperature. As the ambient temperature and humidity were on higher side during the time of experiment the tetramoulter (control) moth emergence occurred between 11.12 and 11.45 days. Emergence of moth in imidazole treated batches has been preponed by about 1.5 days. This may be due to less amount of food reserves in the smaller pupae. The moth emergence period is important from hybridization point of view, so as to synchronise the larval duration with emergence of two different races. No significant differences in emergence period have been observed in different imidazole treated batches indicating that the imidazoles have no impact on the moth emergency period.

Fecundity : The efficiency of egg production in induced trimoulters is more than in tetramoulters (Kiuchi, 1996). Xue *et al.* (1994) have observed

a better egg recovery percent with low unfertilized egg rate in trimoulters. Results in this study indicate that fecundity ratios of control and imidazole treated batches have been 100 (control) : 69.37 (KK-42, 3 μg), 67.19 (KK-110, 10 μg), 61.98 (KK-22, 20 μg) Kawaguchi *et al.* (1993) have reported a 40% egg laying by induced trimoulters. In this finding, the increase in egg laying efficiency by trimoulters may be due to lower fecundity in tetramoulter during adverse rearing season for bivoltine. However, this observation of greater agility of trimoulter moth corroborates the finding of Xue *et al.* (1994) who have emphasized that the trimoulter moths are more active than the tetramoulters.

Weight of 100 eggs : Kawaguchi *et al.* (1993) have reported that the size and weight of eggs are reduced by the application of 5 ~ 20 μg /larva of KK-42. As the lower effective dose has been applied in this study, the weight of 100 eggs shows no significant difference from that of tetramoulter control except in case of KK-22 treated batch. Reduction of weight of eggs has also been recorded by Xue *et al.* (1994).

Weight of 100 ants (newly hatched larvae) : Weight of 100 ants has no significant difference from that of control. This indicate fertilization, mating and oviposition are almost the same as that of control tetramoulter.

5.15. Utilization of Nitrogen in Food by Induced Trimoulters of the Hybrid of KPGB x P₅

5.15.1 : Ingestion : Silkworms can utilize nitrogen in food very efficiently for the synthesis of silk protein during its fifth larval stage (Horie and Watanabe, 1986). Leaf nitrogen during the season under study has been only 2.66%. This low level may be due to high rainfall during this season. As more than 90% of the total larval food requirement is consumed by silkworms during the fourth and fifth stage, the utilization of nitrogen in

food during these two stages can be considered as almost the total amount of nitrogen consumed by the larvae in life time. Horie and Watanabe (1986) observed an ingestion of 231 mg of nitrogen during the 4th and 5th stages. But in this present study the nitrogen ingestion is only 93 mg by the control larvae. This may be due to variation in rearing condition and also lower quantity of nitrogen in the leaves. Imidazole treated trimoulter-destined larvae have consumed almost 50% or a little less amount of nitrogen in food as compared to that of control tetramoulter during the 4th and 5th stages taken together.

5.15.2. Excretion : Excretion of nitrogen through faecal matters by trimoulter-destined larvae as compared to that of the control is less. This implies that nitrogen in food has been efficiently utilized by the imidazole treated larvae destined for precocious pupation, for both growth of body tissues and formation of silk protein in silk gland. Horie and Watanabe(1986) have claimed that during early part of the fifth stage the ingested nitrogen from food is utilized by larvae for the formation of body tissue and during later half of fifth instar for the formation of silk protein in silk gland.

5.15.3 Digestion and digestibility percentage : The digestibility is quite high during the wet part of summer. This may be due to high water content in the leaves promoting higher nitrogen utilization (Mishra and Upadhyay, 1992). Horie and Watanabe recorded 65.1% nitrogen digestibility which has gone upto 69.89% in this finding. Induced trimoulter-destined larvae have higher digestibility over the control tetramoulter. The reason may be the precocious programming in trimoulters for the formation of cocoon shell and pupa within a shorter larval life span.

5.15.4 . Distribution of nitrogen from larval body to cocoon shell and pupa

Larval body : Compared with the results obtained by Horie and Watanabe (1986) the nitrogen assimilation by the larvae in the present investigation is much less. This difference may be due to difference in silkworm hybrid reared, season of rearing, total nitrogen in the leaves and larval conversion efficiency. Efficiency of conversion of digested nitrogen in larval body has been recorded to be 92.30% in control tetramoulters 96.98% in KK-42, 94.28% in KK-110 and 93.10% in KK-22. These values indicate that ECD (nitrogen) is quite high in the induced trimoulters leading to higher nitrogen percent in larval body. The values are 7.01% in control, 8.09% in KK-42, 7.71% in KK-110 and 8.40% in KK-22 treated larval body.

Male Cocoon shell : 70% of the larval nitrogen has been utilized by control tetramoulters for male cocoon shell. In the induced trimoulters the values are 41.93% in KK-42, 36.36% in KK-110 and 51.85% in KK-22 treated individuals . Horie and Watanabe (1986) have recorded a 75.5% conversion of larval nitrogen for male cocoon shell. The lower value obtained in the present experiment can be attributed to the completely different hybrid silkworm, mulberry cultivar and rearing environment.

Female cocoon shell : 55% of the larval nitrogen has been utilised for the female cocoon shell in control tetramoulters, whereas the 35.48% in KK-42, 30.30% in KK-110 and 37.03% in KK-22 treated individuals. Utilization of nitrogen for female cocoon shells is always lower than in the males. Horie and Watanabe (1986) have recorded this conversion value as high as 64.5%.

Male Pupa : A 30% of the larval nitrogen has been utilized for male pupal biomass of control tetramoulters. The values are 45.16% in KK-

42, 51.51% in KK-110 and 37.03% in KK-22 treated individuals. This indicates a higher allocation of nitrogen towards pupal growth in trimoulters than in case of tetramoulters.

Female Pupa : A 43.33% of the larval nitrogen has been utilized for female pupal biomass of control tetramoulter. The values are 64.51% in KK-42, 69.69% in KK-110 and 59.25% in KK-22 treated larvae. This implies that the induced trimoulter larvae are more programmed for utilization of nitrogen in food for the production of yolk protein of eggs rather than for cocoon shell. Horie and Watanabe (1986) have recorded utilization of 34.88% of larval nitrogen for female pupa in tetramoulters. In this experiment the value (43.33%) in the tetramoulters has been recorded to be higher than the reported value. This perhaps due to unfavourable wet part of summer when nitrogen percentage in the leaves is low. A physiological compensation has been developed by way of boosting up of the efficiency of utilization.

5.16 Rearing Performance at the Next Generation of Induced Trimoulters of KPGB x P₅

During wet part of summer(unfavourable) : The tetramoulters obtained in the next generation have maintained the general qualitative and morphological traits. Thus, the return to the usual tetramoulter development from the induced trimoulter in this experiment supports the finding of He and He (1994). The authors are of the opinion that the qualitative characters and morphological characters of *B.mori* when reared with imidazole treatments for continuous and alternate generations, show normal performance at par with the control tetramoulters when reared without imidazole treatment. Neither the genotype nor the other efficiencies of the race are found to be affected due to the imidazoles used.

During November-December (favourable) : From the result it is evident that the qualitative characters of *B.mori* have not been influenced at the followings generation in which no imidazole has been applied. Wigglesworth (1972) suggested that in lepidopteran larvae the number of larval instars and their morphological characters are genetically fixed. These can only be modified by alteration of external factors. the imidazoles have altered the larval development programme with the formation of precocious cocoons. The action of imidazole compounds is anti juvenile in nature and this action can be rescued by application of juvenile hormone analogue, such as methoprene as have been argued by Asano *et al.*(1986) and Darvas *et al.* (1989). He and He (1994) have advocated for the use of imidazoles during unfavourable rearing season for the production of eggs from trimoulter moths and use of this eggs for rearing during favourable season. This result justifies the use of imidazole compounds during unfavourable season for the production of bivoltine eggs which can be reared during the following favourable season without further application of imidazoles for achieving desired yield in tropical plains such as in West Bengal.

5.17. Rearing Performance of the Hybrids of Three-way Crosses of Polyvoltine (Nistari) X Induced Trimoulter Bivoltine Male (KPGB X P₂)

Fecundity : Successful mating is one of the important factors for fecundity of female silk moths (Askari *et al.*, 1984). The oviposition stimulating substance and transfer of sperm into female genital tract induce ovoposition (Yamaoka and Hirao 1981). Virgin females and the females mated with sterile males show low level of fecundity (Fugo and Arisawa, 1992; Danthanarayana and Gu, 1991) and sterility in males may be caused by exposure of male silkworm larvae to high temperature during spinning

(Sugai and Hanaoka, 1972; Sugai and Takahashi, 1981; Dutta *et al.*, 1982). But in the present result the fecundity in all the three way crosses of Nistari X induced trimoultar bivoltine males treated with 3 μ g of KK-42, 10 μ g of KK-110, or 20 μ g of KK-22 has been almost the same as in the case of control (Nistari X tetramoultar bivoltine male). The Nistari (a polyvoltine race) though having a low yielding trait, is very resistant to the adverse climate of West Bengal and has a very high combining ability with any improved race/line showing positive heterosis (Jayaswal *et al.*, 1990). Xue^{et al.} (1994) after using induced trimoulters for the preparation of F₁ commercial hybrid eggs has obtained the result at par with that of control. His conclusion is that the health of the induced trimoultar male moths is not impaired at all. The present result also substantiates this conclusion.

Weight of 100 eggs and weight of 100 ants (newly hatched larvae) :

Weight of 100 eggs has no significant difference either within the treatments or with the control. So, hybridization with induced trimoultar male has no adverse effect on the egg vigour. Hence the newly hatched out larval weight has not been affected.

Hatching percent : The nonsignificant difference in the percentage of hatching of eggs obtained from imidazole-treated and control females implies that these compounds have no sterilizing effect on the males of *B. mori*.

Single cocoon and single shell weights : Single cocoon and single shell weights of treated individuals are almost at par with those of control in all the treatments. This indicates that the hybrid vigour and combining ability of Polyvoltine with trimoultar bivoltine for economic traits are not affected by the imidazoles. This finding is in support of He and He (1994) who have observed that neither the genotype nor the combining

ability are affected by the use of imidazoles for the production of trimoulters.

SR% (Silk ratio) and filament length : Both the economic traits have not been affected by the imidazoles applied. These are almost at par with the control values.

From the analysis of all the observations it is imperative that during the wet part of summer in West Bengal imidazoles can be used for the production of trimoulter males of bivoltine races. The F_1 hybrid eggs can also be effectively raised as seed during this season by crossing polyvoltine with the resultant trimoulter bivoltine males. The suitable doses of the compounds are 3 μg of KK-42, 10 μg of KK-110 and 20 μg of KK-22.