DISCUSSION

Eri silkworm, *Philosamia ricini* is extensively reared in Assam and other parts of N.E. India. It is a household level cottage industry contributing a substantial amount to the rural economy of this region. The primary food plant of the silkworm is castor, *Ricinus communis* locally known as ‘Era’. There are some other plants species which are used as eri silkworm food during scarcity of primary food plant. The present study was designed to evaluate the impact of two different species of food plants (*Ricinus communis* and *Heteropanax fragrans*) on the growth and developmental behaviour of eri silkworm in different seasons (Spring-Summer, Rainy and Winter). The experiments done and the results obtained thereof are being discussed in this chapter.

5.1 MORPHOLOGICAL AND ECONOMICAL CHARACTERS OF *Philosamia ricini* FED IN CASTOR (*Ricinus communis*) AND KESSERU (*Heteropanax fragrans*) DURING THREE SEASONS:

The consumption and utilization of food constitute a sine qua non of growth, development and reproduction of an animal. The amount, rate and quality of food consumed by a larvae influences its performances viz. growth rate, developmental time, final body weight, dispersal ability and probability of survival (Kerkut and Gilbert, 1985). In phytophagous chewing insects, leaf shape may have an affect since some of these insects prefer or require an edge and rolled leaves make feeding difficult (Bernays and Chapman, 1970).

In the present study, it was observed that the size of the eri larvae fed on castor and kesseru did not exhibit much noticeable difference during the early larval
period. However, the kesseru fed larvae shows a slight elevation in length and weight in the 2\textsuperscript{nd} and 3\textsuperscript{rd} instar. This difference may be due to high moisture content in the tender and medium leaves of kesseru compared to castor. In the 4\textsuperscript{th} and 5\textsuperscript{th} instar castor fed eri larvae showed increased larval size as well as higher weight (Table 3, 6, Fig. 1, 2) than the kesseru fed larvae. Earlier Ratte (1985), reported that holo and hemi metabolous insects terminate growth at metamorphosis. Growth curves of the initial development of these insects are remarkably similar, but vary during the final instar. In the present study high larval weight of castor fed eri larvae during the last instars can be explained with reference to percentage of moisture content in food plants. Parpiev (1968) reported that high water content affects both edibility and assimilability of leaves in the silkworm. While low water content affects energy expenditure, nutritional efficiency and growth of herbivorous insects (Martin and Van't Hof, 1988). The present result is similar to the work of Paul \textit{et al.}, (1992) which revealed that absolute consumption and growth rate/day/larva increased with increasing percentage of leaf moisture. Thus high moisture content in matured leaves of castor may cause an elevation of length and weight in castor fed larvae during 4\textsuperscript{th} and 5\textsuperscript{th} instar. High protein content in castor leaves in comparison the kesseru leaves may be another reason which shows this difference. A comparatively higher larval size and weight was recorded during Spring-Summer than Rainy and Winter season (Table 3, 6; Fig. 1, 2). Earlier Ratte (1985) reviewed the effect of temperature on insect size and showed that some insects have direct relationship between weight and temperature. Sweeney and Vannote (1981) produced empirical evidence to explain the interaction of temperature in developmental process and physiology, in determination of size and fecundity of six co-existing mayflies. Thus higher temperature during Spring-Summer than Winter
may be reflected in the higher weight and size of the silkworm. The nutrient contents in the host plants were also recorded high during Spring-Summer than Winter, which may cause this variation. The growth rate during the early instars were found to be quite low for both castor fed and kesseru fed eri silkworm. It is similar to the work of Poonia (1978) who reported that most of the food consumed during early instars is spent in energy for maintenance and very little is use for growth.

The present study revealed that the larval duration was shorter in the castor fed larvae compared to the kesseru fed ones (Table 8, Fig. 4). It can be suggested that due to lower amount of nutrients in the kesseru plant (Gogoi and Yadav, 1995) than the castor plant, the kesseru fed worms cannot invest sufficient amount of metabolites for its growth which leads to prolonged larval duration. It is evident that the deficiency in any essential nutrient in insects usually causes merely cessation of growth and prolonged survival (Gordon, 1959). The shortest and the longest larval duration were found during rainy and winter season,respectively. The larval duration during Spring-Summer was slightly longer than the rainy season. Longer larval duration during Winter may be attributed to low protein and moisture content in the leaves and/or due to the minimal food intake by the larvae during winter. And likewise high protein and moisture content in leaves and increased food intake by the larvae during Spring-Summer and Rainy season gives shortest larval duration.

The abiotic factors and food plants greatly influence the growth and development of muga silkworm _Antheraea assama_, its shell weight, silk content and reelability (Chandrashekar and Thangavelu, 1986). Sarkar _et al._ (1994) observed that high crude protein in mulberry leaves gave high cocoon, shell weight and cocoon yield of _Bombyx mori_. In the present study also cocoon and shell weight was found higher in the castor fed eri silkworm (Table 7, Fig. 3) in comparison to
kesseru fed eri silkworm. This indicates that higher protein content in the castor leaves facilitates better silk spinning performance in eri larvae. The weight of cocoon of castor fed larvae is higher during Spring-Summer and kesseru fed larvae cocoon shows higher weight during Rainy season though it is comparatively lower than castor fed cocoon. Higher cocoon weight of kesseru fed eri larvae during Rainy season may be due to high moisture content in kesseru leaves which is similar to the work of Pandey (1995). During Winter low quantity of proteins and carbohydrates are observed in the leaves of food plants which may be a possible cause of low cocoon and shell weight.

The fecundity rate of castor fed eri larvae was found higher than the kesseru fed ones (Table 9). Seasonally, Spring-Summer was found to be ideal as the fecundity rate of both castor fed and kesseru fed larvae was found high during this season. It was reported that the reproductive dynamics and fecundity of female insects depends upon the climatic conditions as observed in Colorado beetle—"Leptinotarsa decemlineata" (Opyrchalowa et al., 1976).

5.2 PROTEIN, CARBOHYDRATE AND MOISTURE CONTENT IN THE LEAVES OF THE FOOD PLANTS OF ERISILKWORM, CASTOR (Ricinus communis) AND KESSERU (Heteropanax fragrans):

The phytophagous insects utilize plant nutrients for their growth and development. Any change in the nutritional quality of the food plant may result in adverse physiological adaptation. The environmental factors also have a direct impact on the food plants in different seasons (Hering and Taguchi, 1951). It was reported that the growth and development of silkworm B. mori and the economic characters of their cocoons were influenced to a great extent by the nutrient content
of mulberry leaves (Krishnaswami et al., 1971). Moreover, the quantity, rate and quality of food consumed by an insect larvae has a great bearing on its survival, growth rate, developmental duration and final body weight (Ramadevi et al., 1993). In this study, the average protein concentration in the tender leaves was recorded to be higher than the medium and matured leaves in both the food plants *R. communis* and *H. fragrans* (Table 9, 10; Fig. 5). Jawale et al., (1981) had reported that the tender leaves of mulberry were rich in nitrogen in comparison to medium and matured leaves. The protein concentration in *R. communis* was recorded higher than *H. fragrans* in the three seasons. Earlier Mattson (1980) reported that tree leaf contains less amount of protease inhibitors than forbs, which is reflected in low protein content in *H. fragrans*. High amount of soluble protein and total phenol with higher phenylalanine ammonia lyase activity was observed in good quality leaves and on the other hand poor quality leaves contain lower quantities of soluble protein, total phenols as well as phenylalanine ammonia lyase (Hazarika et al., 1995). Protein content was high during Spring-Summer than Rainy and Winter for both the food plants. However, Chaluvachari and Bongale, (1995) reported lower value of leaf nitrogen, protein and sugar content in leaf during Summer, associated with lower value of larval duration and larval weight. It could be attributed to high temperature and low humidity prevailing during Summer crop (Krishnaswami et al., 1971). The temperature and humidity variations prevailing during Spring-Summer and Summer must have caused the variation in protein level in the food plants.

The selection behaviour of phytophagous insects is often governed by the distribution of secondary chemicals in plants. The silkworms consumed the secondary chemicals for the fulfillment of their different metabolic activities.
In insects as in higher animals glycogen is the main tissue carbohydrate. It is a necessity for normal growth and development of phytophagous insects. They are probably the most widely distributed and widely occurring compounds in plants in which their quantity and quality vary enormously. Quantitative rather than qualitative content of food plays more decisive role in determining the relationship between insect and plant (House, 1969). However the quantitative requirement of carbohydrate is found to vary in different insects.

The total carbohydrate content was recorded manifold high in secondary host plant *H. fragrans* in comparison to primary host plants *R. communis* (Table 11; Fig. 5). The matured leaves recorded higher carbohydrate content than the tender and the medium leaves of both the food plants. Seasonal variation has shown that carbohydrate content during winter was lower than Spring-Summer for both the plants. It was reported by Pandey (1995) that the carbohydrate concentration of plants varies according to season. Li and Sano (1984) observed that high quantity of carbohydrates and lower levels of water and proteins in the feed resulted in slower larval growth and less body weights. This is similar to the present study were kesseru fed larvae demonstrated prolonged larval duration and lower larval size and weight. It is suggested that the optimum requirement of carbohydrate appears to depend proximally on metabolic rate.

Water content in plants acts as an ideal solvent for many biochemical reactions. Plant cells, which have lost water are at a turgor pressure lower than the maximum value, and are said to be suffering from water stress. The water content of cells, when falls below the optimum value causes some degree of metabolic disturbances. It was indicated that as water stress increases from mild to moderate,
cellular biochemical processes are increasingly affected. The protein and chlorophyll biosynthesis are sensitive to rather mild water stress (Fitter and Hay, 1983).

The body water content of an insect is directly influenced by dietary water (Scriber and Slansky, 1981). The higher growth in the mature larva of *Antheraea assama* and *Philosamia ricini* are influenced by dietary water and which also plays a decisive role in the selection of food. The higher values of leaf moisture in mulberry leaves were associated with higher values of larval weight and moulting ratios (Chaluvachari and Bongale, 1995). Similar results have been obtained in the present study.

The moisture content in this study was found higher in the matured leaves of *R. communis* whereas *H. fragrans* recorded high moisture content in tender and medium leaves. Winter season recorded low moisture content in both the food plants. The difference in the moisture content of *R. communis* during Spring-Summer and Winter was more pronounced in comparison to *H. fragrans*, where Spring-Summer and Rainy season exhibited almost equal moisture content (Table 14, Fig.5). The high moisture content in leaves of food plants increased the palatability and assimibility of nutrients and considering that, leaf moisture content may be a criteria in determining leaf quality (Parpiev, 1968).

It was reported that tree leaves are capable of absorbing significant amounts of liquid water from their surface thereby raising leaf water potential (Conner *et al.*, 1977). Even as the entire water content imbibed by the larvae from the leaves are required by the silkworm for the metabolic activities, they have a mechanism of water conservation bestowed upon them (Sahay and Kapila, 1993). Such as rectal papillae in the alimentary tract and cryptonephric arrangement of malphigian tubules.
5.3 PROTEIN, CARBOHYDRATE, DNA AND RNA IN WHOLE BODY TISSUE OF Philosomia ricini DURING 3<sup>RD</sup>, 4<sup>TH</sup> AND 5<sup>TH</sup> INSTAR FED ON CASTOR AND KESSERU LEAVES DURING TWO DIFFERENT SEASONS:

The diet of phytophagous insects has got a significant influence on the biosynthesis of silk protein. Thus the choice of food, rate of feeding and food utilisation is indispensable for its growth and development. Continuous feeding generally depends on continuous phagostimulation and an inadequate concentration of phagostimulants leads to early cessation of feeding (Bernays and Simpson, 1982; Barton Bowne, 1975). This indicates that the pattern of feeding is not one which is simply switched on and than continue until repletion; it requires continued positive feedback, an important part of which is usually chemical feedback from the food (Kerkut and Gilbert, 1985). During the early period of each instar, increased rate of food consumption causes an influx of protein from the dietary sources, but it was suggested to be largely used to meet the energy demand (Poonia, 1978), which is evident from the moderate increase of the protein content in the early periods.

In this study 3<sup>rd</sup> instar castor fed eri larvae was found to contain lower protein content than kesseru fed larvae (Table 15, 18; Fig. 7, 11). This may be assumed to be due to the early development of silk gland of castor fed larva, for which the tissue and haemolymph protein are utilised for its synthesis. In the initial stage the silk gland absorbs proteins from the haemolymph which may be carried from the different parts of the body and at the later stage this proteins are utilized to form the silk protein and the silk gland stops to absorb protein from the haemolymph. During the 2<sup>nd</sup> instar stage the silk gland of both castor and kesseru fed larvae remains at the threshold point of development without much utilization of
protein. The development of the silk gland in kesseru fed 3rd instar larvae was seen to take place at a later stage than the castor fed larvae. However the protein content in the 4th and 5th instar of castor fed larvae was found to be comparatively very high than the kesseru fed larvae (Table 16, 17, 19, 20; Fig. 7, 11). The gradual increase of larval body weight with consumption of food indicate higher level of protein content in the later stages of development. Since the tree leaf contains less amount of protease inhibitors than forbs this may advocate the low protein content in the kesseru fed larvae than the castor fed one.

The protein content during Spring-Summer was higher than the Winter season in both castor and kesseru fed larvae. Low intake of food by the silkworm during winter season may tend to lower the protein content in the larvae. Riley (1980) reported that the feeding activity was likely to be minimal during Winter period forcing the larvae to mobilize energy storage reserve to maintain homeostasis. Low moisture and protein value during Winter of the host plant leaves may be another factor in the availability of low protein in the body of the larvae during this season. In the three instars of both castor and kesseru fed larvae daywise fluctuation of protein concentration was noticed. This fluctuation may be due to metabolic changes taking place in the body of the silkworm. The slightly decreased protein content during the early part of each instar may be attributed due to moulting or shedding of the exuvia. The massive accumulation of protein in the larvae during 5th instar, pre-pupal stage is very important for the overall developmental economy of the insects, since it serves as major protein reserve for adult differentiation.

The fluctuation in the protein content in the whole body tissue may be attributed to a variety of interrelated accessory factors. The primary factors which can affect the protein content on the seasonal basis is diet, reproductive conditions
and moulting cycle status. Other factors, which might also be considered, are temperature, photoperiod, developmental stages and soil condition of host plants. The influence of all these factors will implicit seasonal changes in the protein and FAA pool (Graney and Giesy, 1986). Therefore it may be assumed that the variable nutritional value caused by seasonal variation may be responsible for varied type of protein level as recorded in different seasons.

Many insects are sensitive to changing environmental condition. Of the many significant environmental factors, light, temperature are considered to be of prime importance in the regulation of growth and development of insect (Giespitz and Zarankina, 1963; Danilevskii, 1963).

The principle classes of organic compounds that are found in insects are carbohydrates along with protein and lipids. They are present in free form and combined with other molecules. Carbohydrates contribute to the structure and function of all insect tissues. The insect endoskeleton, is formed of amino polysaccharide – chitin. Glucose, Fructose and Sucrose are nutritionally adequate sugars for most insects but much interspecific variation exists in the utilisation of other dietary carbohydrate. Nutritional studies have shown that quantitative requirements for carbohydrate often vary according to age, sex, and metamorphic stage of the insect.

The amounts of dietary carbohydrate requirement, in insect larvae differ variably. The larvae of some species do not require carbohydrate since they are able to substitute dietary protein or lipid for carbohydrate and meet their energy needs for growth and development from amino acids and fatty acid oxidation. Other species require only moderate amounts of carbohydrate in their larval diet. The adult insects commonly consume large quantities of carbohydrates. These ingested carbohydrates
together with nutrient reserve carried over from the larval stage, are necessary to meet the insects energy demand (Wyatt, 1967).

Carbohydrate in insect is stored in the form of glycogen and trehalose which can be readily converted into glucose (Shimada, 1979). The stored form of carbohydrates is being mobilised during periods of great activities such as prolonged flight, in moulting, maturation of eggs and pupation.

In this study carbohydrate content of the kesseru fed larvae is found to be higher than the castor fed larvae. During 3rd and 4th instar the difference was comparatively less (Table 15, 16, 18, 19; Fig. 8, 12) but in the 5th instar kesseru fed larvae exhibited very high carbohydrate content (14.26 mg/gm) than the castor fed larvae (5.8 mg/gm) (Table 17, 20; Fig.8, 12). This may be attributed due to the high carbohydrate content in the kesseru leaves in comparison the castor leaves. The carbohydrate in diet of Heliothis zea markedly influenced the haemolymph trehalose level (Friedman et al., 1991).

The carbohydrate concentration was found to fluctuate daywise in different instars. The low level of carbohydrate prior to ecdysis may be due to the affect of refusal to take food by the insect during that period, for which tissue, haemolymph, and fat reserve, carbohydrate, protein and lipids are utilized for its maintenance. Lohr and Gade, (1983) reported high concentration of haemolymph, carbohydrate at the end of last instar in Carausius morosus which decreased towards ecdysis.

In this study it was observed that the carbohydrate content in the castor fed larvae falls prior to spinning. This may be due to the larval – pupal transformation stage. It was reported by Bade and Wyatt, (1962), that a decrease in glycogen concentration was observed during larval-pupal transformation in Cecropia silkworm. The same was reported by Crompton and Birt, (1967) for Lucilia cuprina.
During larval pupal transformation stage histolysis of organs occur, which may cause the fluctuation of carbohydrate concentration. Carbohydrate content was found to be low in winter in comparison to spring-summer (Table 22, 26; Fig.8, 12). It has been generally accepted that many over wintering insects accumulate sugar alcohols such as sorbitol and/or glycerol through the breakdown of glycogen storage (Grubor et al., 1992). In these insects low temperature triggered the synthesis of sugar, alcohol (Baust, 1982; Hayakawa and Chino, 1981). In silkworm *Philosamia cynthia* sugar alcohol are not accumulated appreciably but instead, large amounts of trehalose appear in the haemolymph, the interconversion between glycogen and trehalose is temperature dependent (Wiens and Gilbert, 1967, Fischer et al., 1971).

The total soluble carbohydrate recorded gradual increase from 3rd instar and reached maximum in the 5th instar. This is similar to the observation made by different workers on silkworm, reporting a gradual increasing trend of carbohydrate content in each instar. A similar report was made on the quantitative assay of total body carbohydrate of *Spaeroderma rusticum* (Sharma and Muddir, 1992) which revealed a gradual rise in 3rd instar being significantly higher and with the rising trend in the later instars. The females showed slightly higher body carbohydrate concentration than the male. The ascending trend from the earlier to later stages is attributed to greater metabolic demand and that in adult female for ovarian maturation.

In the present study estimation of DNA of body tissue of 3rd instar larva revealed no significant difference between castor and kesseru fed larvae, though slightly higher content of DNA was observed in kesseru fed larvae. However in the 4th and 5th instar castor fed larvae had higher DNA content than kesseru fed larvae (Table 23, 27; Fig. 9, 13). During Winter DNA content was slightly lower than
Spring-Summer for both castor fed and kesseru fed larvae. DNA showed an increasing variation from 3rd to 5th instar. The increase in DNA content with the growth of the larvae may be due to rapid cell multiplication and continuous increase in cell number (Pant and Kumar, 1979). Therefore, higher DNA content in castor fed larvae meant higher growth rate.

The RNA content of the castor fed larvae was found to be higher than the kesseru fed larvae in the 3rd and 4th instar. But in the 5th instar, kesseru fed larvae showed higher RNA content than the castor fed larvae (Table 24, 28; Fig. 10, 14). High RNA concentration during 5th instar in kesseru fed larvae may be necessary for protein synthesis. As protein synthesis is directly related to RNA which in turn is dependent on DNA, therefore, it would not be unexpected to observe variation of nucleic acid level during the larval and transformational stages. High RNA concentration in the castor fed larvae during 3rd and 4th instar may be due to high protein biosynthesis and its decrease in the 5th instar may be attributed to cessation of growth before metamorphosis. As kesseru fed larvae exhibits longer larval duration than castor fed larvae its metamorphosis takes place later on which may in turn cause high RNA content during that period.

5.4 PROTEIN, CARBOHYDRATE, DNA AND RNA CONTENTS IN THE SILK GLAND OF philosamia ricini FED ON CASTOR AND KESSERU DURING 4TH AND 5TH INSTAR:

The silk gland forms an important organ in the silkworm. It constitutes roughly 40% of the body weight of the larva. The growth of the silk gland is the manifestation of accumulation of organic components with particular reference to
proteins (Ito, 1967; Tazima, 1978). The protein biosynthesis in silk gland have been shown to be influenced by photoperiod (Tazima, 1978; Kerkut and Gilbert, 1985).

Among all other growth ingredients protein is one of the most essential and chief growth factor which is correlated with the growth and development of the silkworm. The biochemistry of silk gland of silkworms attracted the attention of Japanese Scientists and systems responsible for the synthesis of silkprotein was studied in detail by them (Hasoda et al., 1963; Kobayashi et al., 1966). The synthesis of silk protein during the growth of the silkworm larva was studied by Pukudawa (1959) and they reported that 70 percent of the silk protein produced by B. mori is directly taken from mulberry leaves, while about 30 percent thereof is derived from tissue and haemolymph protein. In this study protein content of the silk gland was recorded high in the 4th day 4th instar in castor fed larvae, however in the 5th and 6th day of 5th instar kesseru fed larvae demonstrated very high protein content than castor fed larvae (Table 34; Fig. 16). Parenti et al., (1985) reported that the two neutral amino acids, with threonine, proline, aspartate, glutamate and their amides are taken up by the silk gland directly from the haemolymph for silk synthesis. The silk gland of both castor and kesseru fed larvae demonstrated high protein content than the body tissue in this study.

In this study, the enzyme activity was found high in kesseru fed larvae during 5th instar. This high activity of enzyme may be intimately linked with maintenance of size of amino acid pool which determines the synthesis of protein. This may be reflected in the silk gland of kesseru fed larvae where high protein content was recorded during the 5th instar.

The protein content in the silk gland of both castor and kesseru fed larvae during winter is found to be less than that of spring summer. As mentioned earlier
protein biosynthesis in silk gland is influenced by photoperiod. Higher photoperiod 
during Summer induces higher protein biosynthesis and low photoperiod during 

Dhinkar et al., (1991) reported that high percentage of humidity in the 
atmosphere with low photoperiod and cool temperature seems to generate 
favourable activities for silk gland. In contrast, high photoperiod, and hot 
temperature with low humidity seems to generate unfavourable conditions for the 
activities of silk gland. Thus, Spring-Summer seems to be ideal for silk-protein 
synthesis.

The amount of protein in the silk gland was found to increase 
commensurating with the developmental age of different silk worms. An increasing 
trend of protein concentration in the silk gland was also observed in Galleria 
mellonella (Janda, 1975).

In insects as in all other organism when food intake exceeds its energy 
requirement, the excess is stored in the form of carbohydrates, proteins and fats in 
the storage tissue. The energy stores are mobilized under the influence of various 
hormone at different developmental stages of the insects. The haemolymph is the 
extra-cellular fluid in the haemocoel which bathes the fat body and silk gland and 
thus act as a transport medium for the exchange of metabolites.

In this study silk gland of 4\textsuperscript{th} day of 4\textsuperscript{th} instar and 5\textsuperscript{th}, 6\textsuperscript{th} day of 5\textsuperscript{th} instar 
showed higher carbohydrate content in kesseru fed larvae than in castor fed larvae 
(Table 32, 34; Fig. 16). Further it was observed that carbohydrate content in the 4\textsuperscript{th} 
day of 4\textsuperscript{th} instar was higher than the 5\textsuperscript{th} and 6\textsuperscript{th} day of 5\textsuperscript{th} instar in both castor and 
kesseru fed larvae. Crompton and Birt, (1967) observed a decrease in total soluble 
carbohydrate in the entire tissue as well as in different organs (fat body,
haemolymph and silk gland) immediately before pupation, which subsequently accumulated, preparative to its utilisation for the rapid anabolic process involved in the glycogen biosynthesis.

Carbohydrate content in the silk gland during spring-summer was found to be higher than the winter season for both castor and kesseru fed larva. This falls in the pattern reported earlier (Yamashita and Hasegawa, 1974; Kerkut and Gilbert, 1985) that the carbohydrate content in the silk glands of the winter worms are low in comparison to the summer worms.

A significant increase in the concentration of carbohydrates was earlier noted by Unni, (1988) in the silk gland and the haemolymph of *P. ricini*, which decreased during the 1st day of spinning. The fluctuations of carbohydrate in different organs, supports the speculation about the conversion of fat into carbohydrate during developmental stages.

At the end of larval life, the fifth larval instar is characterised by hypertrophy of the silk gland and an increasing rate of silk synthesis until cocoon spinning. Following food intake after the 4th ecdysis the exponential growth of the gland is accompanied by a parallel increase in the cellular content of DNA, RNA and proteins (Prudhomme and Couble, 1979).

The DNA content in the silkgland of 4th day of 4th instar kesseru fed larvae was higher than the castor fed larvae during Spring-Summer and Winter. In the 5th instar too it followed the same trend (Table 32, 34; Fig. 16). However DNA content in castor fed larva has shown descending pattern in the 5th and 6th day of 5th instar. Since DNA content is considered as the estimate of cell number, its depletion in the silk gland suggest less growth rate and/or less multiplication of cells (Pant and Kumar, 1979).
Low DNA content during 4th day of 4th instar in castor fed larvae may be suggested due to early moulting of castor fed larvae because of its higher growth rate. But as kesseru leaves contain lower amount of nutrients, the kesseru fed worms cannot invest sufficient amount of metabolites for growth for which its larval duration is prolonged.

Study of the posterior silk gland of developing China oak silkworm larva revealed the rate of RNA accumulation in the gland increased correspondingly with growth (Shubnikova and Ginstberg, 1954). Thereafter a number of workers made detailed studies on RNA and DNA of posterior silk gland (psg) of last instar larvae of B. mori. Evidence suggests that about 200,000 fold increase of DNA content in each of psg of B. mori during larval development of the insect (Neulat, 1967; Chizei and Tojo, 1972; Gage, 1974). It was demonstrated by Tashiro et al., (1968), Morimoto et al., (1968) and Gillot and Daillie (1968) that the nucleic acid content of psg of B. mori increases keeping pace with the developmental days of the last larval instar (Wu et al., 1978; Maekawa et al., 1980).

In this study it was observed that the RNA content in the silk gland of the 4th day 4th instar castor fed larva was higher than the kesseru fed larva. On the other hand in the 5th and 6th day of 5th instar RNA content was found high in the kesseru fed larva than the castor fed larva. The high protein content in the silk gland of kesseru fed larvae during 5th instar may be reflected in the high RNA content. Michalik et al., (1984) reported only RNA content seems to change in the silk gland of the insect during the last larval instar.
5.5 ASPARTATE AMINOTRANSFERASE (AsAT), ALANINE AMINOTRANSFERASE (AAT) AND FREE AMINO ACIDS (FAA) IN HAEMOLYMPH OF CASTOR FED AND KESSERU FED LARVA OF Philosamia ricini.

Enzymes are remarkably effective catalyst responsible for the thousands of co-ordinated chemical reactions involved in biological processes of living organisms. All enzymes are proteinous in nature and there is evidence that many of the haemolymph protein function as enzymes (Laufer, 1960).

Since amino acid pool is responsible for protein synthesis and protein is directly related to growth and silk synthesis, it is therefore essential to study the aminotransferase activity in regard to environmental or seasonal fluctuation.

One of the biochemical characteristics of insects in general is the high concentration of free amino acids in their haemolymph as well as in the tissues. Amino acids serve as units for complex molecules of proteins or they get metabolised. An early step in the breakdown of amino acids is their loss of amino groups which are transferred to a suitable compound, the process being known as the transamination. Transamination, involving glutamate, aspartate, alanine and their corresponding keto acids are most active. Pyridoxal phosphate is utilised as the coenzyme and the optimum pH is about 7.5.

McAllan (1958, cited by Price 1961) showed that the transamination activity in the cockroach and the housefly increases during larval development and the adult differentiation parallel to the increase of protein synthesis.

In this study, both AsAT and AAT in the haemolymph of 4th and 5th day of 5th instar of kesseru fed larva is found to be higher than the castor fed larva. The low activity of both the enzyme in the present study in castor fed larvae compared to
kesseru fed larva is due to high amounts of carbohydrate and protease inhibitions concentration in the castor leaves (Joshi, 1986). This is further supported by our findings. As it is reported by Adibi, (1968); Davidson and Longslow, (1975) that since protein synthesis require a balanced amino acid pool. The AAT activity forms a general index of amino acid breakdown and AsAT marks towards the mobilization of amino acids into the glucogenesis.

In general, the enzyme system in insects bears a great similarity to that known in both bacteria and mammals (Chen et al., 1964). The activity of both AsAT and AAT were found to be almost par with one another during the developmental stages in Sarcophaga ruficornis (Pant and Kumar, 1980). In the present study the activity level of AsAT is found higher than AAT for both castor fed and kesseru fed larva. It is similar to the observation made by (Pant and Unni, 1978) where barring 2nd day old 5th instar, AsAT was predominant over AAT is haemolymph throughout the development till pupation. According to Pant and Pandey (1980) the high activity of aspartate and alanine aminotransferase were observed throughout the larval development and spinning period in A. mylitta. The elevated activity of these enzymes during larval developmental stages parallel the protein synthetic rate in the gut, fat body and silk gland. The most important physiological function of L-alanine aminotransferase is the maintenance of amino acid pool at a proper level for protein synthesis (Meister, 1965) and the supply of metabolites for energy metabolism (Sacktor, 1974).

In our study aminotransferase activity was found to be low during winter in comparison to Spring-Summer for both castor fed and kesseru fed larva. It was reported by Unni et al., (1983) that photoperiodism had a significant influence on aminotransferase activity of erisilkworm. Therefore low photoperiod during winter...
must have a negative affect for aminotransferase activity. Unni and Pant (1985) reported low activities of aminotransferase when larvae of *P. ricini* was exposed to cold stress.

Insects are known to contain a high concentration of FAA in their haemolymph and in other tissues (Florkin and Jeuniaux, 1974). It is likely that the levels of these amino acids are affected by degradation of proteins to FAA during histolysis.

Probably Narumi (1950) was the first worker to make detailed investigation on haemolymph FAA in the sericogenous insect *B. mori*. High titre and wide variety of FAAs in the haemolymph of the insect was observed by Wyatt (1961). Variations of FAAs pattern during larval growth and moulting have been observed qualitatively and quantitatively in insects (Levenbook, 1962; Chen, 1966). Among the 20 haemolymph FAA, glycine, alanine, serine and tyrosine are reported to being actively utilised for silk production in the sericogenous insect *B. mori* (Shimura et al., 1958; Tashiro et al., 1968).

In this study FAA during 5th and 6th day of 5th instar is found to be higher in castor fed larva than the kesseru fed larva. Variation in their concentration is probably due to different growth related functions, like metamorphosis (Kondo, 1957; Duchateau and Florkin, 1958). Detoxification (Friedler and Smith, 1954; Cauda, 1955); organ formation (Goldberg and Demeillon, 1948); energy production (Sackett, 1961); histogenesis (Jolly et al., 1972); Silk production (Tazima, 1978) and also due to their multiple function during different metabolic, metamorphic and stressful situations like osmoregulation (Bishop et al., 1925), protein synthesis (Buck, 1953), cocoon spinning (Wyatt, 1968).
As noted by Burnet and Sang (1968) the composition of amino acids in *Drosophila* larvae can be altered by different diets. Similar nutritional effects on the level of free amino acids have been reported for the aphid, *Myzus persicae* (Strong, 1964).

In this study FAA is found to be slightly higher during winter than in Spring – Summer lower protein content during winter may cause higher FAA during winter. However the difference is quite negligible. In contrary to this Cook *et al.*, 1972 reported that the fall of FAA during winter season is associated with the carbohydrate metabolism.

In kesseru fed larvae, the low level of FAA may be because they are utilized for generating energy via gluconeogenesis, which is further supported by the increased level of enzyme activity in the present study.