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SEASONAL VARIATION OF THE FOLIAR CONSTITUENTS OF PRIMARY (*MACHILUS BOMBYCINA*) AND SECONDARY (*LITSEA CITRATA*) FOOD PLANTS OF *ANTHERAEA ASSAMENSIS* HELFER (MUGA SILKWORM) IN NAGALAND

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ABSTRACT
Seasonal variation of certain foliar constituents of *Machilus bombycina* King (Som) and *Litsea citrata* Blume (Mejankori), the primary and secondary food plants respectively of *Antheraea assamensis* Helfer (muga silk worm) were estimated in Nagaland. The moisture and total carbohydrate were recorded to be maximum during summer season both in Som (69.81±0.34% and 10.48±0.54%) and Mejankori (68.59±0.60% and 14.16±0.39%) respectively. The mean value of moisture was higher in Som (67.66±0.49%) than Mejankori (65.18±0.91%) leaves while the reverse was observed in case of total carbohydrate (9.82±0.24% and 12.52±0.42% respectively). The crude fibre was recorded maximum during spring in both Som (17.38±1.28%) and Mejankori (20.18±1.76%) with fluctuating trend during summer and winter season. The total nitrogen, crude protein and total ash were higher in Som (3.15±0.06%, 19.66±0.35% and 8.10±0.19% respectively) during spring season, while Mejankori retained higher percentage of these constituents (3.01±0.12%, 18.82±0.74% and 4.11±0.09% respectively) during winter season. The soluble and reducing sugar in Mejankori (4.80±0.29% and 0.39±0.06%) and Som (3.73±0.29% and 1.43±0.12%) was recorded maximum during summer and winter respectively. The mean total soluble sugar content was found to be higher in Mejankori (3.78±0.28%) than Som (3.32±0.13 %) whereas the total reducing sugar content was found to be higher in Som (1.39±0.06%) than Mejankori (0.32±0.03%). Interaction effect between seasons and food plants revealed significant differences for moisture, total carbohydrate, soluble sugar, total nitrogen and crude protein while difference were insignificant for reducing sugar, crude fibre and total ash.

INTRODUCTION

Among the different types of food plants of *Antheraea assamensis* Helfer (muga silkworm), *Machilus bombycina* King (Som) and *Litsea polyantha* Juss. (Soalu) are the principal food plants, while *Litsea salicifolia* Rox ex Wall. (Dighloti) and *Litsea citrata* Blume (Mejankori) are considered as secondary food plants of which the later is grown naturally in Nagaland, a hilly state of North Eastern region of India lying between 25°26´ - 27°40´ N latitude and 93°20´- 95°15´E longitude. Growth pattern and palatability of silkworm species depend on the seasonal variation of foliar nutrients, the quantitative estimate of which have been made in different food plants of Mulberry (Sujathamma and Dandin, 2000; Bose and Bindroo, 2001), Tropical tasar (Sinha and Jolly,1971; Sinha *et al*.,1992; Sinha *et al*.,2005), Oak tasar (Sinha *et al*.,1986; Banerjee *et al*.,1993) and Eri silk worm (Dutta, 2000; Hazarika *et al*., 2005; Baruah, 2013). Most of the food plants may contain all the nutritional requirements but the quantity of each foliar constituent may not be well balanced for proper growth and development of silkworm. While certain information on foliar constituents of food plants of muga silkworm have been reported from the plains of Assam (Yadava and Goswami, 1992; Dutta *et al*., 1997; Choudhury *et al*., 1998; Singh *et al*., 2000; Kakati and Kakati, 2011; Tikadar *et al*., 2013), no comprehensive works are done in higher altitudinal areas. The rearing of *Antheraea assamensis* in Nagaland shows strong seasonality and the larvae being fed upon Mejankori produces a unique creamy-white glossy silk, while on Som it produces golden yellow silk (Kakati *et al*., 2010) which may reflects on the nutritive value of host plants and their seasonal variability. The silk produced by the silkworm grown on Mejankori plant is very attractive and several times costlier than the silk produced by silkworms from other Host plants (Choudhury *et al*., 2012). Hence, it is attempted to evaluate the impact of seasonal variations on certain foliar constituents of the two muga silkworm host plants *Machilus bombycina* (primary) and *Litsea citrata* (secondary) under Nagaland climatic condition.

MATERIALS AND METHODS

Fresh leaves of all ages and sizes from the host plants i.e. Som and Mejankori were collected form Ungma sericulture farm (rearing site) separately in spring (April-June), summer (July-August) and autumn (October-December) seasons corresponding to the rearing schedule of muga silkworm. The leaves were properly cleaned and used for different estimations. The moisture content and crude fibre were estimated by using the method of AOAC (1990). Total Nitrogen was determined by micro- Kjeldahl method and the nitrogen percentage was multiplied by 6.25 to calculate crude protein (Kirk and Sawyer, 1991). Total soluble sugar and total carbohydrate was estimated by Anthrone method (Yem and Willis, 1954). Reducing sugar was estimated by the method described by Burner (1964). The total ash was determined using the method as described by Kirk and Sawyer (1991). The estimation was done seasonally in triplicate and mean value was recorded for interpretation of results. The data were statistically analysed following the analysis of variance for seasons as well as host plants in a randomized complete block design and co-efficient of variation (CV %) was calculated following the method described by Gomez and Gomez (1984).

RESULTS AND DISCUSSION

The seasonal variation in nutrients of the leaves of two host plants *Machilus bombycina* and *Litsea citrata* utilized for rearing of *Antheraea assamensis* in different rearing seasons is given in Table 1.

Moisture content

Summer season retained maximum quantity of moisture followed by spring and autumn season in both food plants ($p < 0.05$). The mean moisture content was found to be higher in *Machilus bombycina* (67.66 ± 0.49%) than *Litsea citrata* (65.18 ± 0.91) with no significant difference at 5% level of probability. The interaction effect due to host plant x season was found to be significant. However, Dutta *et al.* (1997) recorded highest moisture content in *Litsea polyantha* (71.84%) followed by *Litsea salicifolia* (69.98%) and *Litsea citrata* (69.55%) and the lowest in *Machilus bombycina* (67.02%) in Jorhat region of Assam. In a study on the status of biochemical constituents in four morphotypes of *Persea bombycina,* Baruah and Baruah (2007) estimated higher moisture content in the tender leaves than that of semi mature and mature leaves, however other mineral contents showed an increasing trend from tender to mature leave. In a comparative analysis among eight food plants of different wild silkmoth species at Mokokchung, Kakati *et al.* (2013) recorded maximum moisture content in *Heteropanax fragrans* (73.52 ± 1.77) and minimum in *Quercus acutissima* (56.09 ± 2.33). The environmental condition may be one of the factors responsible for fluctuation of moisture content of leaves in different seasons.

Carbohydrate

The percentage of total carbohydrate as a whole was higher in the leaves of *Machilus bombycina* (10.48 ± 0.54%) and in *Litsea citrata* (14.16 ± 0.39%) during the summer season. The mean total carbohydrate content was also found to be higher in *Litsea citrata* (12.52 ± 0.42%) than in *Machilus bombycina* (9.82 ± 0.24%) and the former retained more carbohydrate content than the later in all seasons. The carbohydrate content of the leaves of both plants was found to be variable with regard to seasonal fluctuation ($p < 0.05$) and was observed in the order of summer > spring > autumn in the case of *Machilus bombycina* and in the order of summer > autumn > spring in the case of *Litsea citrata*. Probably, the moisture quantity of the leaves maintained a degree of relationship in carbohydrate content in different seasons, because reduction of moisture content of leaves from summer to winter may be due to utilization of moisture to build the carbohydrates and other constituents of leaves, thereby increasing dry matters (Sharma and Devi, 1997). Many researchers also reported favorable effect of total carbohydrate content in different silkworm host plants like Mulberry (Bose and Bindroo, 2001), Tasar (Sinha and Jolly, 1971), Muga (Dutta *et al.*, 1997; Choudhury *et al.*, 1998) and Eri host plants (Pathak, 1988; Dutta, 2000; Hazarika *et al.*, 2005) in different seasons.

Soluble and reducing sugar

The percentage of total soluble sugar as a whole was higher in the leaves of *Litsea citrata* during summer season (4.80 ± 0.29%), while in *Machilus bombycina*, it was higher during the autumn season (4.22 ± 0.16%). The variability of reducing sugar content of the leaves of both plants was none significant with regard to seasonal fluctuation and was observed in order of autumn > spring >summer in the case of *Machilus bombycina* and in the order of autumn > summer > spring in the case of *Litsea citrata*. The mean total soluble sugar content was found to be higher in *Litsea citrata* (3.78 ± 0.28 %) than *Machilus bombycina* (3.32 ± 0.13 %) whereas the

Table 1: Seasonal variation of foliar constituents of host plants of *Antheraea assamensis* Helfer in Nagaland

Season	Host Plants	Parameter (%) Moisture	Carbohydrate	Soluble sugar	reducing sugar	Crude fibre	Total nitrogen	Crude protein	Total ash
Spring	A	68.93 ± 0.49	9.66 ± 0.28	2.92 ± 0.11	1.41 ± 0.12	17.38 ± 1.28	3.15 ± 0.06	19.66 ± 0.35	8.10 ± 0.19
(Apr-Jun)	B	66.13 ± 0.67	10.46 ± 0.40	2.32 ± 0.12	0.25 ± 0.01	20.18 ± 1.76	2.42 ± 0.02	15.13 ± 0.12	3.70 ± 0.11
Summer	A	69.81 ± 0.34	10.48 ± 0.54	3.32 ± 0.10	1.33 ± 0.06	15.35 ± 0.97	2.67 ± 0.07	16.70 ± 0.45	7.83 ± 0.25
(Jul-Aug)	B	68.59 ± 0.60	14.16 ± 0.39	4.80 ± 0.29	0.39 ± 0.06	20.06 ± 1.60	2.65 ± 0.03	16.56 ± 0.19	3.78 ± 0.06
Autumn	A	64.24 ± 0.64	9.32 ± 0.24	3.73 ± 0.29	1.43 ± 0.12	17.01 ± 1.98	2.60 ± 0.30	16.25 ± 1.84	7.80 ± 0.21
(Oct-Dec)	B	60.81 ± 1.45	12.93 ± 0.31	4.22 ± 0.16	0.31 ± 0.01	16.19 ± 1.66	3.01 ± 0.12	18.82 ± 0.74	4.11 ± 0.09
Mean + SE	A	67.66 ± 0.49	9.82 ± 0.24	3.32 ± 0.13	1.39 ± 0.06	16.58 ± 0.83	2.81 ± 0.11	17.54 ± 0.71	7.91 ± 0.11
	B	65.18 ± 0.91	12.52 ± 0.42	3.78 ± 0.28	0.32 ± 0.03	18.81 ± 1.01	2.69 ± 0.07	16.84 ± 0.44	3.86 ± 0.07
S. Ed+	Host plant	0.64	0.30	0.16	0.06	1.29	0.11	0.69	0.12
	Season	0.78	0.37	0.19	0.08	1.58	0.14	0.85	0.15
H x S		1.11	0.53	0.27	0.11	2.23	0.19	1.20	0.21
CD-5%	Host plant	NS	0.62***	0.32**	0.13***	NS	NS	NS	0.25***
	Season	1.60***	0.76***	0.39***	NS	NS	NS	NS	NS
	H x S	2.26**	1.08***	0.56***	NS	NS	0.39***	2.45***	NS

* = Significant and **& *** = Highly Significant at the 5% level of probability, NS = Non Significant; A = Machilus *bombycina* (Som), B = *Litsea citrata* (Mejankori)

total reducing sugar content was found to be higher in *Machilus bombycina* (1.39 ± 0.06%) than *Litsea citrata* (0.32 ± 0.03%). Yadava and Goswami (1992) revealed that total sugar percentage was significantly higher in Som when compared to Soalu and observed that medium-aged leaves of Som and Soalu were higher in total sugar percent when compared with tender and mature leaves. However Jolly *et al*. (1974) reported a decreasing trend for total sugar percent with the maturity of the leaves in tasar silkworm food plants. Dutta *et al*. (1997) reported that Som leaves possessed the highest amount of total soluble sugar (4.63%) and was at par with Mejankori leaves (4.62%). Bharali (1984) reported that total soluble and reducing sugar content of different types of som plant leaves increased from spring to autumn. While comparing reducing sugar percentage, Dutta *et al*. (1997) observed that Som leaves contained maximum reducing sugar (2.56%), but did not differ significantly from that of Soalu (2.33%) and Mejankori (2.49%).

Crude fibre

The mean crude fibre content recorded a non significant variation (5%) between the *Machilus bombycina* (16.58 ± 0.0.83%) and *Litsea citrata* (18.81 ± 1.01%). However, *Litsea citrata* (20.18 ± 1.76%) retained more moisture in spring season than *Machilus bombycina* (17.38 ± 1.28%). The seasonal fluctuation of crude fibre content of the leaves of both plants was variable ($p < 0.05$) and observed in the order of spring > autumn > summer in the case of *Machilus bombycina* and in the order of spring > summer > autumn in the case of *Litsea citrata*. Crude fibre is the ash free material and highly composed of cellulose and lignin, but cannot be digested by the silkworm larvae. Agarwal *et al*. (1980) stated that the crude fibre played a very important role in the feeding of larvae. A negative correlation exists between the crude fibre and the feeding of the larvae, showing that the lesser the percentage of crude fibre, the better would be the rearing results. Yadava and Goswami (1992) observed that *Machilus bombycina* and *Litsea polyantha*, the primary food plants of muga silk worms recorded a comparatively low percentage of crude fibre in comparison to tasar silkworm food plants. Dutta *et al*. (1997) recorded the highest percentage of crude fibre in Som leaves (27.18%) followed by Dighloti (23.26%) and Soalu (15.49%) whereas Mejankori leaves possessed the lowest amount (7.91%).

Total nitrogen

The total nitrogen content as a whole was higher in the leaves of *Machilus bombycina* during spring season (3.15 ± 0.06 %) while in *Litsea citrata* it was higher during the autumn season (3.01 ± 0.12%). The nitrogen content of the leaves of both plants was found to be variable with regard to seasonal fluctuation ($p < 0.05$) and observed in the order of spring > summer > autumn in the case of *Machilus bombycina* and in the order of autumn > summer > spring in the case of *Litsea citrata* . The crude protein of both the plants showed seasonal variability at 5% level with maximum of 19.66 ± 0.35% during spring followed by summer and autumn in the case of *Machilus bombycina* while *Litsea citrata* recorded the maximum 18.82 ± 0.74% during autumn followed by summer and spring. The mean total nitrogen (2.81 ± 0.11 % and 2.69 ± 0.07 %) and crude protein content (17.54 ± 0.71 % and 16.84 ± 0.44 %) did not show significant difference between *Machilus bombycina* and *Litsea citrata* respectively. The interaction effect due to host plant x season was significant for both the nutrients as well as host plant ($P < 0.05$). The total nitrogen content of different leaves of the tested plants differs significantly from each other and also in different seasons. Yadava and Goswami (1992) reported that *Litsea polyantha* leaves possessed higher total nitrogen and crude protein than *Machilus bombycina*, but variation between the leaf types for nitrogen was not significant. Dutta *et al*. (1997) recorded maximum amount of total nitrogen and protein in Mejankori (3.08% and 15.53%) followed by Som (2.36% and 12.25%), Soalu (2.02% and 10.36%) and Dighloti (1.39% and 7.50%). The protein contents in different sericigenous plant are highly variable and are greatly influenced by environment and heredity (Agarwal *et al*., 1980; Pathak, 1988).

Ash content

The percentage of total ash as a whole was higher in the leaves of *Machilus bombycina* during spring season (8.10 ± 0.19 %) while in *Litsea citrata* it was higher during the autumn season (4.11 ± 0.09%). The variability of ash content of the leaves of both plants was found to be highly significant with regard to seasonal fluctuation ($p < 0.05$) and was observed in the order of spring > summer > autumn in the case of *Machilus bombycina* and in the order of autumn > summer > spring in the case of *Litsea citrata*. The mean total ash content was found to be higher in *Machilus bombycina* (7.91 ± 0.11 %) than *Litsea citrata* (3.86 ± 0.07%) and the former retained more ash content than the later in all seasons.

Thus the foliar analysis is most essential, which acts as a guideline to muga culture to evaluate the nutritional status of different food plants for their selection based on the nutritional value for the feeding of the silkworm and its development. Further, variation in feeding parameter and palatability among different silkworm species might be due to the variation in quantitative requirement of each of the nutrients and also the required balance of nutrients within the species owing to many factors including the synthetic ability of the organism and metabolic activities involving specific interrelations between certain nutrients (House, 1974). Paul *et al*. (1992) observed that availability of moisture content in the leaves enhanced the feeding efficiency of the larvae which in turn increased the growth rate. The effect of seasons on foliar nutrient concentrations varied depending on the specific nutrient which is usually regarded as an indicator for the nutritional status of plants (Bergmann, 1993). In a fodder tree of Central Himalaya, Singh *et al*. (2010) recorded significant positive correlation between crude fibre (CF) and altitude, indicating increasing trend of CF with an increasing altitude. Singh and Todaria (2012) reported that nitrogen concentration in plant increased with altitudinal gradient and the variations in nitrogen content may partly be attributed to re-translocation of leaf nitrogen into branches before leaf fall and partly due to a dilution factor with expansion and maturity of the leaves (Khosla *et al*., 1992). The comparative studies on seasonal variations on foliar constituents of both Som and Mejankori revealed significant differences for moisture, total carbohydrate, soluble sugar, total nitrogen and crude protein whereas difference were insignificant for reducing sugar, crude fibre and total ash. The results suggest an impact of seasonal variation on the nutritive value of leaves and highlight the importance of Mejankori at par with Som host plant on rearing performance and cocoon production in different seasons (Kakati *et al*., 2010).

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