ABSTRACT

Muga silkworm, *Antheraea assamensis* Helfer. is endemic to Northeast India which produces golden silk. They are polyphagous, multivoltine and semi-domesticated in nature which primarily feed on two host plants. They are *Persea bombycina* Kost. commonly known as "Som" and *Litsea monopetala* Roxb. commonly known as "Sualu". In accordance with Assamese calendar the Six different generations of muga silkworm in a year are known as i) *Jaruwa*-Winter (December-January) ii) *Chatuwa*-Early spring (February-March) iii) *Jethuwa*-spring (April-May) iv) *Aheruwa*-Early Summer (June-July) v) *Bhodia*- Summer (August-September) vi) *Kotia* –Late summer or Early winter (October-November). In Assam muga silk culture is practiced in the districts of upper Assam and certain parts of lower Assam. Sericulture in Goalpara district existed almost as a practice among the people since a long time. The following works have been done by taking the following objectives:

1. Survey of different som cultivation sites of the district for collection of diseased samples.
2. Study symptoms, intensity of diseases and quantum of loss caused to the foliage.
3. Epidemiology of the most common diseases of the district.
4. Qualitative and quantitative study of soil, air and phylloplane mycoflora.
5. Management of the dominant disease using systemic, non-systemic fungicides and plant extracts.

Different age leaves of Som, depending upon the size and shape viz. tender, semi mature and mature were randomly collected during rearing season of muga silkworm from February, 2014 to January, 2016. Five major muga silkworm rearing villages of Goalpara district were selected depending upon the direction namely Dorapara Agia on the centre, Budlung pahar on the North, Lengopara on the South, Buraburi on the East and Bhalukdubi Kalyanpur on the West respectively for collecting the leave samples during six muga crop seasons. The infected leaves were categorized into five grades. During the study period four major diseases were encountered in all the 5 study sites which were isolated and identified as Grey blight, Leaf blight, Leaf spot and Leaf rust. The average percentage disease index of two years showed that the disease intensity of Grey blight (12.531%) were highest followed by Leaf blight (9.61%), Leaf spot (9.351%) and Leaf rust (9.268%) during the study period.

It was observed that the Grey blight disease were highest in occurrence during the *Aheruwa* generation and lowest during the *Jaruwa* generation of muga silkworms. Similarly the Leaf spot and Leaf blight disease were highest during the *Bhodia* generation & lowest during the *Jaruwa* generation and Leaf rust were maximum during *Aheruwa* and lowest during the *Jaruwa* generation of the muga silkworms.

The epidemiological study shows that during the *Aheruwa* crop of muga silkworm the Som leaves are highly affected with all the four foliar diseases. During high temperature, humidity and rainfall the disease incidence were the highest. Among all leave types taken Tender, Semimature and Mature, it was observed that the disease incidence were encountered only on the Mature leaves while there were absence of disease on the Tender and Semimature leaves. Among the five study areas , it was seen that the Lengopara site is highly effected with Grey blight disease during the *Aheruwa* crop of muga silkworm while maximum temperature were 34°C and minimum temperature 20.75° C, humidity 92% and minimum 63% and rainfall were 4286.25 ml. During *Chatuwa* crop among the 5 study sites the occurrence of Grey blight disease were maximum in Bhalukdubi, Lengopara and Buraburi, while in Budlung pahar occurrence of Leaf blight were maximum and in Dorapara Agia occurrence of Leaf rust were maximum. During *Jethuwa* crop among the 5 study sites occurrence of Grey blight were maximum in Lengopara and Buraburi. Leaf spot were maximum in Bhalukdubi, Leaf blight were maximum in Budlung pahar and Dorapara Agia. For *Aheruwa* crop , occurrence of Grey blight were maximum for all the study sites. Among them Lengopara is highly effected with Grey blight. During *Bhodia* crop also occurrence of Grey blight disease were maximum in all the sites, among them Buraburi were highly effected with Grey blight. Again for *Kotia* crop Grey blight, Leaf spot and Leaf rust were maximum in Lengopara and leaf blight were maximum in Dorapara Agia . During *Jaruwa* generation of muga silkworm occurrence of Grey blight were maximum in Lengopara, Leaf spot were maximum in Dorapara Agia while Buraburi were highly affected with Leaf blight and Bhalukdubi were affected by Leaf rust. It was seen that the high temperature, humidity, rainfall and topography of the area effects the growth of the pathogens and disease as well. It was also observed that during low temperature, low humidity and low rainfall conditions the disease intensity were less. The Som plant leaves were highly affected by Grey blight, Leaf blight, Leaf spot and Leaf rust during the *Aheruwa* and *Bhodia* generation of muga silkworm.

Studies were conducted on air , non-rhizosphere and rhizosphere soil and phylloplane mycoflora of Som during the rearing season of muga silkworm for 2 years. A total of 18

fungal species were isolated and identified from air, 18 fungi from non-rhizosphere soil and 27 fungi from rhizosphere soil depending upon their colony morphology, mycelial characterstics and microscopic observation with the help of standard manuals. While from phylloplane a total of 22 fungi were isolated and identified during the study period. The major aeromycoflora were *Aspergillus niger*, *Cladosporium cladosporioides*, *Rhizopus stolonifer* and *Fusarium oxysporum* during *Chatuwa* generation; *Rhizopus stolonifer* , *Alternaria alternata*, *Fusarium oxysporum* and *Curvularia lunata* during *Jethuwa*; *Aspergillus flavus*, *Rhizopus stolonifer* and *Curvularia lunata* during *Aheruwa*; *Aspergillus flavus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Pestalotiopsis disseminata*, *Colletotrichum gloeosporioides* and *Rhizopus Stolonifer* during *Bhodia*; *Aspergillus niger*, *Cladosporium cladosporioides*, *Aspergillus flavus*, *Aspergillus fumigatus*, *Rhizopus stolonifer*, *Penicillium chrysogenum* and *Pestalotiopsis disseminata* during Kotia while *Penicillium chrysogenum*, *Cladosporium cladosporioides*, *Fusarium oxysporum*, *Rhizopus stolonifer*, *Aspergillus niger*, *Pestalotiopsis disseminata* and *Aspergillus flavus* were the dominat aeromycoflora during the *Jaruwa* generation of muga silkworm. During *Chatuwa* generation a total of 9, during *Jethuwa* a total of 12, during *Aheruwa* a total of 12, during *Bhodia* a total of 8, during *Kotia* a total of 11 and during *Jaruwa* a total of 9 fungal species were isolated and identified from air.

For non-rhizosphere soil, in 3 month age old plantlet a total of 12 fungal species, for 6 months age group of plants a total of 16 fungal species, for 9 month old plantlet a total of 12 no. of fungi and for 12 month old planted a total of 11 no. s of fungal species were isolated and identified. Similarly from rhizosphere soil, a total of 18 fungi from 3 months old plantlet, 20 no. s of fungi from 6 month old plantlet, 14 no. s of fungi from 9 months old plantlet and a total of 17 fungi from 12 month old plantlet were isolated and identified. The dominant non-rhizosphere fungal flora in 3 months age group of plants were *Rhizopus stolonifer, Aspergillus niger, Penicillium chrysogenum, Mucor hiemalis, Trichodema viridae* and *Saccharomyces cereviseae,* in 6 months age group of plantlets the dominant mycoflora were *Rhizopus stolonifer, Aspergillus niger, Aspergillus fumigatus, Trichoderma viridae and Penicillium chrysogenum,* in 9 months age group of plants *Aspergillus fumigatus, Rhizopus stolonifer, Penicillium chrysogenum, Saccharomyces cereviseae, Trichoderma viridae, Aspergillus niger, Mucor hiemalis, Pestalotiopsis disseminata, Mycelia sterila (white), Cladosporium cladosporioides and Aspergillus flavus while in 12 months age group of plants Aspergillus fumigatus, Rhizopus stolonifer, Penicillium chrysogenum, Cladosporium cladosporioides, Aspergillus flavus, Aspergillus niger* were the dominat mycoflora in non-

rhizosphere soil. In rhizosphere the dominant mycoflora for 3 months age group of plants were *Penicillium chrysogenum, Rhizopus stolonifer, Aspergillus niger, Trichoderma viridae, Saccharomyces cerevisae, Mucor hiemalis, Fusarium oxysporum and Mycelia sterila* (white), in 6 months age group of plants the dominant mycoflora were *Rhizopus stolonifer, Penicillium chrysogenum, Aspergillus niger, Trichoderma viridae, Saccharomyces cerevisae and Aspergillus fumigatus*, in 9 months old plantlet the dominant mycoflora were *Rhizopus stolonifer, Aspergillus flavus, Aspergillus niger, Curvularia lunata, Penicillium chrysogenum, Aspergillus fumigatus, Alternaria alternata, Mycelia sterila* (white) and *Trichoderma viridae*. In 12 months age group of plants *Rhizopus stolonifer, Aspergillus flavus, Aspergillus niger, Curvularia lunata, Mucor hiemalis, Penicillium chrysogenum, Aspergillus fumigatus, Cladosporium cladosporioides, Mycelia sterila* (white), *Trichoderma viridae* and *Trichothecium sp.* were the dominant rhizosphere mycoflora.

During *Chatuwa* generation, from the dorsal surface of Tender leaves a total of 11 fungal species and from ventral surface a total of 9 fungal species were isolated and identified. Among them *Rhizopus Stolonifer* were the dominant fungal species which were found in both the dorsal and ventral surface of Tender leaves. In Semimature leaves, on the dorsal surface a total of 11 and on ventral surface a total of 8 fungi were isolated among which *Rhizopus stolonifer* were the dominant fungal species. On Mature leaf dorsal surface a total of 9 fungi and from ventral surface 6 fungi were isolated and identified, among which *Rhizopus stolonifer* were the dominant fungal species encountered. In *Jethuwa* generation of muga silkworm, on dorsal surface of Tender leaves a total of 10 and in ventral surface a total of 8 fungal species were isolated & identified among which, *Alternaria alternata* were the dominant fungal species in the dorsal surface while *Aspergillus flavus* were the dominant fungi on ventral surface of Tender leaves of *Som*. In Semimature leaves on both dorsal and ventral surface a total of 10 no. s of fungi were isolated and identified among which *Rhizopus stolonifer* were the dominant fungal species on both the dorsal and ventral surface of the leaves. In mature leaves on dorsal surface a total of 8 fungi and on ventral surface a total of 10 fungi were isolated and identified, among them *Rhizopus stolonifer* were the dominant fungi in both the dorsal and ventral surface of the Mature Som leaves. In Aheruwa generation of muga silk worm, on the dorsal surface of Tender leaves a total of 8 and on ventral surface a total of 7 fungal species were isolated and identified among which on dorsal surface the dominant fungal species were *Aspergillus niger* and on ventral surface *Rhizopus stolonifer* were the dominant fungi. In Semimature leaves, on the dorsal surface a total of 7

fungi and on ventral surface a total of 7 fungi were isolated and identified among which on both the dorsal and ventral surfaces the dominant fungal species were *Rhizopus stolonifer*. In Mature leaves, on the dorsal surface a total of 8 and on ventral surface a total of 9 fungal species were isolated and identified among which *Rhizopus stolonifer* were dominant fungal species for both dorsal and ventral surface. In *Bhodia* generation, on Tender leaves dorsal surface a total of 7 and on ventral surface a total of 7 fungi were isolated and identified, among which *Aspergillus flavus* were dominant on both dorsal and ventral surface. In Semimature leaves dorsal surface a total of 6 fungi and on ventral surface a total of 8 fungi were isolated and identified among which on dorsal surface *Aspergillus flavus* were the dominant and on ventral surface *Aspergillus niger* were the dominant fungal species.In Mature leaves dorsal surface a total of 10 fungi and on ventral surface a total of 11 fungi were isolated and identified, among which *Pestalotiopsis disseminata* were dominant on the dorsal surface and *Aspergillus niger* were dominant on the ventral surface of leaves. In *Kotia* generation, on Tender leaves dorsal surface a total of 8 fungi and on ventral surface a total of 7 fungi were isolated and identified. Among which the *Aspergillus niger* were dominant fungal species for both the surfaces. In Semimature leaves dorsal surface a total of 7 fungi and on ventral surface a total of 6 fungi were isolated and identified, among which *Aspergillus niger* were the dominant fungal species on both the dorsal and ventral surfaces of the leaves. In Mature leaves dorsal surface a total of 7 fungi and on ventral surface a total of 8 fungi were isolated and identified, among which *Aspergillus niger* were dominant fungal species for both the dorsal and ventral surfaces. In *Jaruwa* generation in Tender leaves dorsal surfaces a total of 9 fungi and in ventral surface a total of 7 fungi were isolated and identified , among which *Rhizopus stolonifer* were dominant fungi for both the dorsal and ventral surfaces. In Semimature leaves dorsal surfaces a total of 5 fungi and on ventral surface a total of 6 fungi were isolated and identified among which *Rhizopus stolonifer* were the dominant fungi for both the surfaces . In Mature leaves dorsal surface a total of 11 fungi and on ventral surface a total of 9 fungi were isolated and identified. Among which *Rhizopus stolonifer* were the dominant fungal species for both the dorsal and ventral surfaces of the leaves.

The physicochemical properties of soil were analysed using standard procedures in summer as well as in winter seasons. Which shows that all the 5 soil samples of the study area were acidic. During summer season, the soil of Dorapara Agia were highly acidic (P^H 5.65) while during winter season also the soil of Dorapara Agia were highly acidic (P^H 4.52). The organic carbon content during summer season were highest on the soil of Lengopara

(3.11 %) and less at Bhalukdubi Kalyanpur (1.16%). Again during winter, the organic carbon content were highest on the soil of Bhalukdubi Kalyanpur (3.03 %) and less at Dorapara Agia (1.5%). During summer season the available nitrogen content were highest on the soil of Lengopara (1035.53 Kg/ ha) and less on the soil of Bhalukdubi Kalyanpur (385 Kg/ha) and in winter season maximum at Bhalukdubi Kalyanpur (1008.18 Kg/ha) and less at Dorapara Agia (502.43 Kg/ha). Again during summer the available phosphorus was maximum at Lengopara, Dorapara Agia and Bhalukdubi Kalyanpur (44.88 Kg/ha each) and less at Budlung pahar (33.34 Kg/ha) while during winter maximum at Budlung pahar (34.62 Kg/ha) and less at Buraburi and Bhalukdubi Kalyanpur (11.54 Kg/ha for each). Available Potassium were maximum at Lengopara (399.84 Kg/ha) and less at Dorapara Agia (181.44 Kg/ha) during summer season, while during winter maximum at Buraburi (524.16 Kg/ha) and less at Dorapara Agia (423.32 Kg/ha). In summer the water holding capacity were maximum for the soil of Lengopara (42.20 %) and less for the soil of Bhalukdubi Kalyanpur (33.15 %) while during winter season maximum at Bhalukdubi Kalyanpur (50.74%) and less at Budlung pahar (44.30%). The soil texture were also analyzed.

The biochemical estimation of foliar constituents were done with standard methods in three seasons for three leave types namely Tender, Semimature and Mature leaves. Results showed that during summer season i.e the *Jethuwa* and *Aheruwa* generation of muga silkworm, the total nitrogen, crude protein, total carbohydrate, crude fibre and moisture content were more than the winter i.e the *Jaruwa-Chatuwa* and autumn season *Bhodiya-Kotia* generations of muga silkworm. The annual mean of all the three types of leaves shows a total nitrogen content of 4.1332%, crude protein of 25.833%, total carbohydrate 12.429%, crude fibre 15.009 % and moisture 68.27 %.

During the study period the most dominant disease in the study area were the Grey blight disease. Hence, the pathogen, *Pestalotiopsis disseminata* which causes the foliar Grey blight of som was isolated from freshly infected som leaves, collected randomly from the study area of Goalpara district of Assam, India. Pure culture of the fungus were maintained by sub culturing periodically on fresh potato dextrose agar (PDA) medium. The pathogen was treated with both systemic and non systemic fungicides to observe the control of mycelial growth of the fungus *in-vitro*. Four systemic fungicides i.e. chemicals namely Bavistin, Copper oxychloride, Mancozeb and Topsin M and leaf extracts from five locally available plant species i.e. *Azadirachta indica*, *Lantana camara*, *Eupatorium odoratum*, *Lucas aspera* and *Bougainvillea spectabilis* were selected as nonsystemic fungicides. Both the treatments

were made 1:100(0.01), 5:100(0.05), 10:100(0.10), 15:100(0.15) and 20:100(0.20) concentration and tested against *Pestalotiopsis disseminata* by the poisoned food method and the percentage of growth inhibition was calculated. Among the systemic fungicides used it was seen that at 0.1% concentration of Bavistin and Topsin M showed 100% growth inhibition of *Pestalotiopsis disseminata*. While Mancozeb at 0.2% concentration showed 100 % growth inhibition against the test fungus. Again the copper oxychloride showed less inhibitory effect against *Pestalotiopsis disseminata*. Among the non-systemic fungicides i.e the plant extracts it was seen that at concentration 0.20% , *Azadirachta indica* showed 95.56% of inhibition against *Pestalotiopsis disseminata* while *Bougainvillea spectabilis* and *Eupatorium odoratum* showed 83.33% and 80% of growth inhibition against the test fungus. The *Lantana camara* and the *Lucas aspera* showed comparatively less inhibitory effect against *Pestalotiopsis disseminata*. Statistical analysis using SPSS software showed that there was a significant difference found among the chemical fungicides used ($F_{3,60}$= 33232.794, $p<0.05$) and inhibition at different concentrations ($F_{4,60}$=19703.914, $p<0.05$) .Furthermore, multiple comparison Tukey HSD also showed that there is a significant difference ($p<0.05$). Similarly for plant extract the statistical analysis showed that there was a significant difference found among the botanicals used ($F_{4,75}$= 4675.620, $p<0.05$) and inhibition at different concentrations ($F_{4,75}$=2266.798, $p<0.05$) .Furthermore, multiple comparison Tukey HSD also showed that there is a significant difference ($p<0.05$).

Diseased leaves were collected from different Som cultivation sites of Goalpara district of Assam and both the qualitative and quantitative data were observed. The symptoms and the disease intensity of the four major foliar diseases Grey blight, Leaf blight, Leaf spot and Leaf rust of Som and quantum of loss caused to the foliage were recorded. The epidemiological studies of the most common diseases of the district showed that the disease occurrence were maximum during the Aheruwa generation of the muga silkworm. This study also gives the qualitative and quantitative data on air, non-rhizosphere & rhizosphere soil and phylloplane mycoflora of Som from the selected villages of Goalpara district of Assam during all the six generations of muga silkworm in a year i.e. the *Jaruwa*, *Chatuwa*, *Jethuwa*, *Aheruwa*, *Bhodia* and *Kotia*. This study also revealed the Physicochemical properties of soil samples of Som plantation area as well as biochemical constituents of *Som* leaves. An attempt has been made to control the dominant fungal pathogen *Pestalotiopsis disseminata* which causes Grey blight disease of Som with the help of both systemic and non-systemic fungicides. As most of the muga seed crops from Goalpara district is supplied to the other

parts of Assam, hence control of various diseases of the food plant som is very important. Due to easy availability of the plants viz. *Azadirachta indica*, *Bougainvillea spectabilis*, *Eupatorium odoratum*, *Lantana camara* and *Lucas aspera* mentioned in the experiment, farmers can use this to control the disease. The residual effects of plant extracts do not cause any adverse effects on the silkworms and can be used in place of systemic fungicides as they may cause adverse effects and mortality of the silkworm. Use of plant extracts to control the Grey blight disease is cost effective.